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KAHRAMANMARAŞ SÜTÇÜ İMAM UNIVERSITY
GRADUATE SCHOOL OF NATURAL AND APPLIED SCIENCE

**DETERMINATION OF OXIDATIVE
STABILIZATION AND BIOACTIVITIES OF
SOME NATURAL PLANTS AND THEIR EFFECTS
ON HEALTH**

HOLEM HASHM RASUL

Ph.D. THESIS

DEPARTMENT OF BIOENGINEERING AND SCIENCES

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Medicinal Plants and Food Analysis

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DECLARATION

I declare and guarantee that all information in this document has been obtained and presented in accordance with academic rules and ethical conduct. Based on these rules and conduct, I have fully cited and referenced all material and results that are not original to this work.

HOLEM HASHM RASUL



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**BAZI DOĞAL BITKİLERİN OKSİDATİF STABİLİZASYON VE
BİYOAKTİVİTELERİNİN BELİRLENMESİ VE SAĞLIK ÜZERİNE
ETKİLERİ
(DOKTORA TEZİ)
HOLEM HASHM RASUL
ÖZET**

Bu çalışmada, *Eremurus spectabilis* Bieb, *Prosopis farcta*, *Capparis spinosa*, *paliurus spina-christi*, *Purslane portulaca*, *Artemisia haussknechtii* boiss, *Prunus microcarpa*, *Silybumista yacarpa*, *Lecarpa lecarnaury* bitkilerinin oksidatif stabilizasyon ve biyoaktivite etkileri araştırıldı. Bu doğal bitkiler Irak'ın Soran kentindeki Balakyan, Hawdian ve Kawna Gund köylerinde toplanmıştır. Bu bitkilerin ekstraksiyonu için üç yöntem; geleneksel Clevencer, mikrodalga ve maserasyon yöntemleri ile birlikte saf su, etanol:saf su (%50/50), etanol, hekzan:etanol (%50/50) ve hekzan çözücülerini kullanılarak ekstraksiyon işlemleri tamamlandı. Bitki ekstraktlarının fitokimyasal analizleri sıvı kromatografi-kütle spektrometresi/kütle spektrometresi (LC-MS/MS) ile gerçekleştirilmiş ve iyon Kromatografi (IC) kullanılarak bazı mineral bileşikler tanımlanmıştır. Ayrıca toplam ekstraksiyon verimi, toplam tanen, toplam fenol, toplam flavonoid, toplam antisiyanidin ve DPPH serbest radikal süpürme, antibakteriyel aktivite özellikleri araştırıldı. Ayrıca antibakteriyel testler için altı patojenik bakteri türleri uygun şartlarda uygulanmıştır. Elde edilen sonuçlara göre, *Prosopis farcta*'nın ekstraksiyon verim değeri (CE: 0,81 gr/gr), *Capparis Spinosa* bitkisinin toplam tanen(CE: 0,462 kg/mg), *Artemisia haussknechtii* boiss bitkisinin toplam fenol (OSE: 3,980 mg GAE/g), *Paliurus Spina Christi* bitkisinin toplam flavonoid, (OSE:0,328 mg QE/g), *Artemisia haussknechtii* boiss bitkisinin toplam antosiyanin (OSE:176,59 mg/kg) içerikleri en yüksek değerler olarak elde edildi. Bitki ekstraktlarının antioksidan kapasitesinin belirlenmesinde serbest radikal (DPPH) olarak 1-difenil-2-pikrilhidrazil kullanılmıştır. Ekstraktların antioksidan aktivitesi, *Artemisia haussknechtii* boiss ve *Pistacia eurycarpa* yale'nin antioksidan aktivite (DPPH) yüzdesi %85'in üzerinde olan en yüksek ve en etkili olduğunu göstermiştir. LC-MS/MS sonuçlarına göre, en yüksek fitokimyasal miktarları *Capparis Spinosa* meyvesinden kersitin maddesi (OSE: 2224.114 µg/g), *Prunus microcarpa* ve *Silybum Marianum* bitkilerindeki Silymarin maddesi(CE: 29082.0 µg/g), *Paliurus Spina Christi* meyvesinden elde edilen kateşin hidrat (CE:10368.99 µg/g), *Artemisia haussknechtii* ve *Pistacia eurycarpa* yale bitkilerinden elde edilen luteolin (CE:5172.051), (MWE: 97336.1 µg/g) bileşikleri en yüksek değerler olarak tesbit edildi. İyon kromatografi sonuçlarına göre K⁺, Mg²⁺, Cl⁻, PO₄³⁻, SO₄²⁻ ve Ca²⁺ iyonları en yüksek miktarları da tüm yöntemlerle *Artemisia haussknechtii* ve *Portulaca oleracea* ekstraktları için harika bir kaynak olduğu tespit edilmiştir. *Artemisia haussknechtii* bitkisinin tüm çözücülerinde, tüm bakterilere karşı ve *Eremurus Spectabilis* bitkisini *Pseudomonas aeruginosa*'ya (ATCC 27853) karşı en yüksek inhibisyon zonu(22.85 mm) oluşturduğu belirlendi. Standartlara kıyasla en iyi MİK değerleri 1,95-125 µg/ml ≥500 arasındadır. Sonuç olarak: doğal biyoaktif kaynağı olan bitki ekstraktları, oksidatif stabilitenin neden olduğu patolojilerin risklerini azaltmak için tıptan ve yenilebilir bitkilerden doğal terapötikler olarak antioksidanlar ve antibakteriyel ajanlar olarak kullanılabileceği anlaşıldı.

Anahtar Kelimeler: Biyoaktif, Fitokimyasal, Antibiyotik, Yenilebilir bitki, Şifalı bitki, 1, 1-difenil-2-pikrilhidrazil

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**DETERMINATION OF OXIDATIVE STABILIZATION AND
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(PhD. THESIS)

HOLEM HASHM RASUL

ABSTRACT

This study was investigated the effects of oxidative stabilization and bioactivities of *Eremurus Spectabilis* Bieb, *Prosopis farcta*, *Capparis Spinosa* fruit *paliurus spina-christi*, *Purslane* *Portulaca oleracea*, *Artemisia haussknechtii* boiss, *Prunus microcarpa*, *Silybum marianum* seed, *Pistacia eurycarpa yale* collected in villages of (Balakian, Hawdian, and Kawna Gund) at Soran city, Iraq. Three methods were employed for the extractions of these natural plants; conventional extraction (CE), microwave-assisted (MW), and orbital shaker (OS) methods along with five solvents including distilled water, ethanol: distilled water (50/50%), ethanol, hexane: ethanol (50/50%), and hexane. The analyses of phytochemicals of plant extracts were performed by liquid chromatography with tandem mass spectrometry (LC-MS/MS) and identified some mineral compounds by using ion chromatography (IC). As well, total extracts yield, total condensed tannin, total phenol, total flavonoid, total anthocyanin, and assessment DPPH free radical, antibacterial activity were evaluated. Furthermore, six pathogenic bacterial species were applied for the antibacterial tests. According to the results, the extraction yield value from *Prosopis farcta*, total condensed tannin from *Capparis Spinosa*, total phenol from *Artemisia haussknechtii* boiss, a total flavonoid from *Paliurus Spina Christi*, total anthocyanin contents from *Artemisia haussknechtii* boiss were the highest values as (CE: 81.56 mg\kg), (CE: 0.462 kg/mg), (OSE: 3.980 mg GAE/g) (OSE:0.328 mg QE/g) (OSE:176.59 mg/kg) in solvents (distilled Water: ethanol v/v), and (distilled water) respectively. The determination of the antioxidant capacity of the plant extracts was used 1-diphenyl-2-pikrilhidrazil as free radical (DPPH). The antioxidant activity of extracts showed that the *Artemisia haussknechtii* boiss, and *Pistacia eurycarpa yale* were the highest and the most effective with antioxidant activity (DPPH) percentage greater than 85%. Likewise, the highest quantities of phytochemicals were quercitin from *Capparis Spinosa* fruit (OSE: 2224.114 µg/g), Silymarin from *Prunus microcarpa* and *Silybum Marianum* (CE: 29082.0 µg/g), and (OSE: 12006.66 µg/g) in (ethanol: distilled water) extracts, also Catechin hydrate from *Paliurus Spina Christi* fruit (CE:10368.99 µg/g), luteolin from *Artemisia haussknechtii* and *pistacia eurycarpa yale* (CE:5172.051), (MWE: 97336.1 µg/g) in ethanol extract according to LC-MS/MS results. Too, the highest quantities of ions of K^+ , Mg^{2+} , Cl , PO_4^{3-} , SO_4^{2-} and Ca^{2+} were detected as a great source for *Artemisia haussknechtii* and *Portulaca oleracea* extracts by all methods. The antibacterial activity was shown the highest inhibition zone at diameter (22.85 mm, 18.97) during conventional and microwave methods in all solvents from *Artemisia haussknechtii* boiss against all bacteria and the *Eremurus Spectabilis bieb* against the *Pseudomonas aeruginosa* (ATCC 27853). With best the MIC values between 1.95-125 µg/ml $\geq$ 500, compared to the standards. In conclusion: plant extracts natural source of bioactive could be used as antioxidants and antibacterial agents as natural therapeutics from medicine and edible plants to reduce the risks of pathologies induced by oxidative stability.

Keywords: Bioactive, Phytochemical, Antibiotic, Edible plant, natural plant, 1, 1-diphenyl-2-picrylhydrazyl

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SYMBOLS AND ABBREVIATIONS

WHO:	World Health Organization
S.E	Standard Error
S.D	Standard division
MDC	Media Diagnostic Center
CAP*	Collage of American Pathology
B:	Bacteria
CE	Conventional extraction
MWE	Microwave extraction
OSE	Orbital shaker extraction
D.W	Distilled water
E: DW	Ethanol: Distilled water
E	Ethanol
H: E	Hexane: Ethanol
H	Hexane
PDA	Potato dextrose agar
CLSI	Clinical Laboratory Standard
MDC	Media Diagnostic Center
CAP*	Collage of American Pathology
B	Bacteria
G–ve:	Gram-negative bacteria
G+ve:	Gram-positive bacteria
MHB	Mueller Hinton broth
MHA	Mueller Hinton agar
PDA	potato dextrose agar
ATCC:	American Type Culture Collection
MIC:	Minimum Inhibitory Concentration
BHT:	Butylated hydroxytoluene
DPPH:	1, 1-diphenyl-2 picrylhydrazyl
HPLC:	High Performance Liquid Chromatography
ROS:	Reactive Oxygen Species
LC:	Liquid Chromatography

MS:	Mass Spectrometry
IC	Ion Chromatography
OD	Optical density
UV	Ultraviolet
L:	liter
ml:	Milliliter
μl:	Microliter
Mm:	millimeter
mg:	Milligram
Kg:	Kilogram
g:	Gram
GAE:	gallic acid equivalent
QE:	quercetin acid equivalent
ppm:	parts per million
ppb:	part per billion
μg:	Microgram
Cm:	Centimeter
nm	Nano meter
rpm	Round per minute
oC	Celsius

1. INTRODUCTION

For thousands of years, medicinal plants have been used in various cultures of the world as a safe therapeutic modality. The operation of medicinal plants is based on the rich experiences of innumerable healers over centuries, inherited from ancestors, healer-to-healer transfer, or developed through personal experiences over time. Modernity or cultural revolutions have not altered the in-depth wisdom of this natural medical paradigm. Consequently, no modern system of medicine can ordinarily lay claim to it. The traditional way of therapy exemplifies well-developed systems such as allopathic, homeopathic, ayurveda, and Chinese systems of therapy, which vary in definition and procedure (Gurib-Fakim 2006; Schippmann *et al.* 2002; Schippmann *et al.* 2005).

Many civilized countries have developed their materia medica, which provides knowledge on various plants used for medicinal purposes. The synthesis of this natural-source human pharmacopeia with remarkable advances in different fields of clinical medicine lays the foundations for a much-needed transformation in the global healthcare system (Kesarwani and Gupta 2013; Olurishe and Mati, 2014). Plants have been one of the first and most widely used resources for curing illnesses since ancient times. Humans and plants have always had a close friendship, and everyone is aware of the healing benefits of plants and their uses (UNESCO 1997).

Therefore, plant products and folk medicine based on the use of seeds, stems, fruit, bark, leaves, plant extracts, and flowers for medicinal purposes is known as herbal medicine. that used to manufacture human and veterinary medicines in the form of extracts and chemical compounds from medicinal plants, and the Clay tablets from the Sumerian and Babylonian cultures, which used several treatment methods for medicinal plants, indicate that medicinal plants have been used in Iraq for thousands of years (Noaema and Sawicka 2018; Nikbakht and Kafi 2008).

The presence of secondary metabolites, which are complex chemical compounds of differing compositions, gives medicinal plants their properties. Alkaloids, glycosides, flavonoids, saponins, tannins, carbohydrates, and essential oils are the various forms of alkaloids. Medicinal and aromatic plants make up a wide group of economically valuable plants that supply the raw materials for the herbal, perfumery, flavor, and cosmetic industries in their native countries (Najafi *et al.* 2010).

Essential oils and extracts from a variety of culinary and medicinal plants, herbs, and spices are powerful biologically active natural agents. The usage of essential oils as antimicrobial agents in food processes may be seen as a new intrinsic determinant for improving food safety and shelf life. (Ebrahimi *et al.* 2008). Chemical drugs are gradually being substituted by active substances from plants, due to their toxic and permanent impact on humans (Banerjee 2002). Throughout the tradition, herbal medicines have been used to cure a variety of infectious diseases. In several developed nations, plant materials continue to play an important role in primary health care as medicinal remedies. (Nezhadali and Zarrabi-Shirvan 2010)

Recently, Medicinal and aromatic plants have received much attention in many areas and make up a wide group of economically valuable plants that supply the raw materials for the herbal, perfumery, flavor, pharmaceutical industries, and cosmetic industries in their native countries (Khorasaninejad *et al.* 2011).

More than 10% of the world's estimated 258,650 varieties of higher plants are used to treat ailing populations. Many well-known drugs (such as tubocurarine, reserpine, aspirin, and morphine) were discovered using common knowledge (Shinwari 2010). On the other side, Over 422000 flowering plant species have been recorded from around the world, with 5000 species used for medicinal purposes (Mozaffarian 2005). According to international statistics, the volume of medicinal herbs exchanges increases at a rate of 12-15 % yearly (De Silva 1997). Traditional drugs are used by more than 80% of the world's population to treat different skin diseases.

The research world has increasingly paid heed to the conventional use of plants for wound healing (Ghasemi Pirbalouti *et al.* 2010). In Germany, a big center of chemical drugs production, herbal medicines are increasingly used by patients and recommended by doctors (Pande *et al.* 2004). The use of non-phytotoxic, more systemic, and easily biodegradable active compounds from higher plants instead of chemical fungicides is revealed (Gottlieb *et al.* 2002). Several plant spices and herbs have a preservative effect, proposing the presence of anti-oxidant and antimicrobial constituents in their tissues. Many medicinal plants contain large amounts of antioxidants other than vitamin C, vitamin E, and carotenoids (Javanmardi *et al.* 2003).

Since plants contain a number of antimicrobial agents, including antifungal activity, which could lead to new antimicrobial drug candidates (Pirbalouti *et al.* 2009). Also, medicinal plants generally contain a variety of compounds that can form a natural antibacterial mixture

and serve as an effective, inexpensive, and safe antibacterial substitute for treating common bacterial infections (Ngemenya *et al.* 2006). Secondary metabolites of plants contained antibacterial activity against important pathogens (Raghavendra *et al.* 2006; Kiran and Raveesha 2005). Considering, pathogenic microorganisms have developed resistance to antibiotics, researchers are interested in biologically active compounds isolated from plant species for the eradication of pathogenic microorganisms (Yousefzadi *et al.* 2011). Oxidation reactions in the human body, especially during times of stress, create free radicals. These toxic radicals can trigger chain reactions that can kill cells and lead to severe diseases like cancer, cardiovascular disease, and neurodegenerative disease (Astley 2003; Ahmad *et al.* 2012; Adebayo *et al.* 2012).

1.1. The objective of this study

1. Extraction of the 10 plants by using different methods and different polarity and non-polarity of solvents to evaluate the feasibility of each other.
2. Chemical tests of the extract to acknowledge the chemical groups existing in the plant extract.
3. Total yield extracts were also evaluated.
4. Isolation, purification, and identification of phytochemicals, especially condensed tannin.
5. Determination and identification of total phenol, total flavonoid, and total anthocyanin contents.
6. Find out the (DPPH) free radical scavenging of root extracted and antioxidant activity.
7. Characterization of isolated phytochemicals compounds by using physical and chemical analyses with different spectroscopic techniques by using LC-MS/MS .
8. Analysis of mineral components Ion Chromatography IC
9. The current investigation was assumed to Isolation and identification Bacteria from Oral of mouth patients in Soran city. Too, the biological activity of compounds isolated from the extract. Identify and discover some of the important antibacterial and their influences on health then compare with some antibiotics by the study of antibacterial activity with a MIC.

1.2. The important points of this investigate

The importance of doing this investigation to study as below:-

1. To explore the effectiveness of dissimilar extraction methods and comparisons were made to determine the best extraction method .
2. To do an opening study on the parameters that can impact extraction efficiency to obtain a better yield .
3. To obtain the optimum financial benefits of the plant by the chance in a research study to produce a marketable product .
4. To open up chances for the investigation study that is related to the therapeutic value of these new natural plants in order to the activity it commercially in the pharmacological interest .
5. To inform and provide sufficient information to the growers of these plants on the actual potential and economic benefits of the plant, especially on its diverse uses.
6. To open up chances for the investigation study that is related to the therapeutic value of these new natural plants in order to the activity it commercially in the pharmacological interest.
7. To inform and provide adequate information to the growers of these plants on the actual potential and economic benefits of the plant, especially on its various uses.

2. LITERATURE REVIEW

2.1. Drug Medicinal plant

In terms of serving humankind, drug medicinal plants have a long and illustrious history on almost every continent. The extraordinary wisdom has been passed down over the generations by traditional healers. The depth wisdom of this natural medical model has not been altered by modernity or cultural revolutions. Pharmacological reasoning based on existing applications, along with phytochemical research, could usher in a new era in disease care (Khan 2014). Even for many scientists, the word "pharmacognosy" remains unfamiliar. The topic is commonly known as the study of primitive drugs of plant and animal origin, according to dictionary definitions. The pharmacognosy specialized for several years in the authentication and consistency assessment of crude products, often of plant origin, using macroscopically and macroscopically methods. Pharmacognosy is the study of biogenic or naturally produced pharmaceuticals and toxins, according to a modern description. It deals with medicinal plants in the form of raw herbs or extracts, as well as pure natural compounds and health-promoting food (Heinrich 2004)

This modern definition, and explanatory rider, reflects the changes that have taken place in the past 50 years, particularly with the focus on phytochemistry and biological activities of natural products. The scope of pharmacognosy is also defined as the study of physical, chemical, biochemical, and biological properties of drugs, drug substances, or potential drugs or drug substances of natural origin as well as the search for new drugs from natural sources (Phillipson 2007). However, at present nearly 30% or more of the modern pharmacological drugs are derived directly or indirectly from plants and in homeopathic or ayurvedic medicines, medicinal plants, their parts, and extracts dominate the scenes. The commercial value of various innumerable drugs and pharmaceuticals derived from tropical forest systems on a worldwide basis is projected at 20 billion dollars a year (Sharif and Banik 2006)

Plants have been used in medicine since the origin of human civilization (Phillipson 2001). The teas, tinctures, powders, poultices, and infusions, as well as other preparations, can also be used to make medicinal plants (Balunas and Kinghorn 2005). Herbal therapy is an ancient tradition in Iraqi civilization, and it is a source of healing for a variety of diseases. Many people believe in its use in prevention and healing (Noaema and Sawicka 2018). Also, herbal therapies are now the most common form of conventional medicine, and they are immensely successful on the global market. Western Europe's annual sales peaked in 2003-2004. (WHO

2014). Therefore, herbal products have been more common in more rich countries in recent decades. In 1997, Germany was the European country with the largest share of the herbal medicine industry (Phillipson 2007).

The public access to these herbal medicinal products (HMP) led to the need for up-to-date monographs and to the use of standardized materials to prevent adverse effects including drug interactions in patients taking other over-the-counter or prescription medicines. The United States of America (USA) and Europe (EU) have been working together to control and authorize the selling of medicinal herbs to patients who order them (Shabir 2003)(Gurib-fakim 2006)

According to a (WHO) Fact Sheet published in 2008, traditional medicine is used by 80 % of the population of many Asian and African countries for primary health care. In several developed nations, 70 % to 80% of the population has used traditional or alternative treatments such as phytotherapy or acupuncture (Phillipson 2001). The European Scientific Cooperative on Phytotherapy was established in 1989 in the European Union. The principal goal of the ESCOP is to advance phytomedicine's scientific standing and to help in the harmonization of their regulatory status at the European level. Based on leading experience from across Europe, the ESCOP publishes state-of-the-art studies of the clinical application of herbal medicinal products.(Gurib-fakim 2006) Also, both food and drug products are governed by the Food and Drug Administration (FDA) in the United States (Frueh *et al.* 2008).

The FDA Scientific Committee released a recommendation paper for the safety evaluation of herbs and botanical formulations planned for use as ingredients in food supplements in June 2008, after a public consultation. The EFSA Scientific Cooperation (ESCO) Working Group was formed in the EU to guide the adequacy of the proposed method for botanical preparations safety evaluation. Standardization is a method that ensures a minimum level of active ingredients in an ingredient, and it's becoming more necessary as a way to ensure a stable supply of high-quality phytopharmaceutical items. It is characterized as the process of establishing repeatable pharmaceutical consistency by comparing a substance to known standard compounds and determining minimum quantities of one or more compounds. Standardization only refers to extracts in the field of phytomedicines. Monographs and/or pharmacopeias can include standards for active ingredients to be used in medicinal products. (Gurib-fakim 2006; Gouveia *et al.* 2013; Konczak 2014).

Therefore, Traditional medicine is a collection of skills, expertise, and procedures based on ideas, values, and interactions from various traditions that are used to promote health and to avoid, diagnose, enhance, or manage physical and mental disorders (Phillipson 2007).

Any medicinal product with one or more herbal substances or one or more herbal preparations as active ingredients, or one or more such herbal substances in combination with one or more such herbal preparations, is known as a herbal medicinal product. Pharmacognosy has provided research on natural substances and diets that are beneficial to one's health (Phillipson 2007), which combines various areas such as phytochemistry, microbial chemistry, biosynthesis, biotransformation, herbal, and analytical chemistry to find new medicines from natural sources. Wherever we go, health and food agencies are implementing policies that promote the rational use of medicinal plants. Traditional medicine relies on the separation, characterization, certainty of synergy, and confirmation of mode of action of active substances, and the discovery of new biologically active compounds remains a challenge because only a limited number of plants have been thoroughly investigated in a systematic manner around the world (David 2014).

2.1.1. *Eremurus Spectabilis Bieb leaf*

The scientific name of *Eremurus* is *Eremurus Spectabilis* the common names of *Eremurus spectabilis bieb* (Rıdvan Polat¹, Bülent Güner, 2017; Ahmed 2016)

Kingdom: Plantae

Clade: Angiosperms

Class: liliopsida

Order: Asparagales

Family: Asphodelaceae Juss

Subfamily: Asphodeloideae

Genus: *Eremurus* S bieb

The *Eremurus spectabilis Bieb* leaf belongs to the family of Liliaceae and geographically distributed in the region of South Asia and Central Asia, including Iran, West Pakistan, Afghanistan, Iraq, Turkey, Palestine, Lebanon, Syria and Caucasus. Their roots accumulate high levels of fructans during their growth and are traditionally used to cure jaundice, liver disorders, stomach irritation, pimples and bone fractures and even as a glue for industrial application (Crockett 1972; Brickell 1996; Dashti *et al.*, 2005).

Individuals use this herb as a vegetable and use it to cure a variety of ailments. The leaves of this plant (which are referred to as "iriş, Gullik" by different people) are used to treat eczema, fungal infections, and diabetes. (Ozturk 2014; Zekiye Ceren Arıtuluk 2011). A local name of plant *Eremurus Spectabilis* in Kurdish language such as Sıtırk (سییتیرک) and (خوي)Xwzhe (Çakilcioğlu *et al.* ,2015). Also, the *Eremurus Spectabilis bieb* plant was Kurdish folkloric herbal medicine, sometimes, plant remedies could be directly collected in the wild by the final users, and only a small number of people retain the knowledge about the correct identification of the plants and their proper use. For these reasons, people usually rely on local markets to get rapid insight into the use of plants in traditional medicine and to purchase them (Cunningham AB, 2001).

However, there is insufficient research is available on the mineral and nutrient content of *Eremurus spectabilis* specimens. The anti-oxidative or radical-scavenging property is particularly intriguing among physiological functions because it can provide health protection against reactive oxygen species and free radicals, which have been linked to over 100 diseases. (Halliwell 1993). As far as we know, only one study has been done on the chemical content of *Eremurus spectabilis* (Gungor 2002). When considering both anti-proliferative and anti-tumor necrosis factor-alpha results and in accordance with the analytical results, thus validating the ethno medical use of *Eremurus Spectabilis* roots and leaf in traditional Kurdish medicine.

Moreover, its anti-inflammatory effect appeared to be intermediate compared to the other two extracts examined (Raffaella *et al.* 2015). The *Eremurus spectabilis* could be a valuable source with high antioxidant capacity, phenolic, and minerals (Tosun, Ercisli, Ozer, Turan, *et al.* 2012a). The *Eremurus spectabilis* extract by hexane ethanolic was stronger affected to the cancer cell lines than an aqueous extract because had high antioxidant capacity with The average amount of total phenolic in samples. Protein, K, Ca, Mg, Fe and Cu contents (Hazha J. Hidayat 2015). also the water extracts of roots and leaf that had higher antioxidant capacity and showed higher radical scavenging activity than ascorbic acid and BHA (Dervişoğlu *et al.* 2014).

The *Eremurus spectabilis* bieb have different proportional antimicrobial activities against bacteria and microorganisms (Kirba 2015). However, the consumption of *Eremurus spectabilis* plant might prove to be beneficial for human health (Murathan *et al.* 2018).

2.1.2. *Prosopis farcta* fruit

The scientific name of *Prosopis* , and the Syrian mesquite (Bovey 2016), the species of the common name is *Prosopis farcta* , growing in and around the Middle East, (Banks & Sol.) J.F. Macbr. – Syrian mesquite (Legume *et al.* 2017).

Kingdom: Plantae – Plants
 Super division: Spermatophyta – Seed plants
 Division : Magnoliophyta – Flowering plants
 Class : Magnoliopsida – Dicotyledons (Sprent 2009)
 Order :Fabales
 Family: Fabaceae/Leguminosae – Pea family
 Genus : *Prosopis* L. – mesquite
 Species: *Prosopis farcta*.

The genus *Prosopis* L., Synonyms of it are *Lagonychium factum* (Ikramov *et al.* 1990), *Prosopis stephaniana* (Gouda *et al.* 1984). *Mimosa farcta*; and *Mimosa stephaniana* (Sprent 2009). It has belonged to the Mimosaceae family and *Prosopis* genus (Mozaffarian 2010). That is widely spread in different dry and semi-dry regions worldwide like; Africa, Australia, America, and Asia (Pérez-garcía, *et al.*,2012; Pinos-rodriguez 2014; Abdelmoteleb *et al.* 2017). Approximately, this genus comprises from 44 to 50 species (William and Jafri 2015).

Prosopis farcta (Banks ET Sol.) Eig. (Family Fabaceae) is a little prickly spiny shrub; it is native to United States, Kuwait, Turkey, Iraq, Iran, Northern Africa, and South Western Asia (Sharifi-rad *et al.* 2014). Another side, the different extracts of *Prosopis farcta* aerial part

showed promising antimicrobial, anticancer, and antioxidant activities, which may be returned to their identified bioactive secondary metabolites (Amal *et al.* 2017).

A literature survey revealed that the *Prosopis farcta* has been used in the remedy of neurological disorders because of antioxidant rule and cholesterol level (Mollashahi *et al.* 2013; Omid *et al.* 2013). Moreover, different parts of the plant (seeds, pods, and leaves) were reported to show a wide range of biological activities including; antioxidant (Poudineh *et al.* 2015), antibacterial (Sharifi-rad *et al.* 2014).

The plant of *Prosopis farcta* that Liver protection capability may be due to their high contents of various flavonoids that act as powerful antioxidants and free radical scavengers (hepatoprotective) (Alharbi *et al.* 2016), to reduce cardiac pain and anti-parasitic (Asadollahi *et al.* 2014; B. Gulalp O. Karcioğlu 2008).

That the selective extraction of plant of *Prosopis farcta* by appropriate solvents could be very important to obtain bioactive fractions (Poudineh *et al.* 2015). The phytochemicals in the hexane and dichloromethane extracts of plant *Prosopis farcta* have shown a potent activity against three isolates of Gram negative and Gram positive bacteria (Layla A. and Ibrahim a., 2018).

2.1.3. *Capparis Spinosa* fruit

The scientific name of capparis is *Capparis spinosa* the common names of caper bush (Bachmann 2009; Inocencio *et al.* 2006; Tlili *et al.* 2011)

Kingdom: Plantae
Clade: Angiosperms
Family: Capparaceae
Family-siceinctific: Capparidaceae
Family common: caper
Genus: *Capparis*
Species: *C. spinosa*
Common name: caper bush
Binomial name *Capparis spinosa*

Capparis spinosa L. is well known with its common name ‘Capers’ or *Capparis* genus in different countries (Azaizeh 2003; Inocencio *et al.* 2006), and includes 350 species of tropical or subtropical origin, many of them distributed in the Mediterranean regions (Inocencio *et al.* 2006). This annual winter deciduous species, also known as caper bush, has rounded, fleshy leaves and broad white to pinkish flowers. (Ramezani and Aghel 2008). Caper had many

Common names: Kabar, Shafallah (كبار شفالله), Mar Gir, Mara Gira And Kabaruk (Blakelock and Townsend ,1980). A caper is also the pickled bud of this plant. Caper bush found practically everywhere in Iraq (Chakravarty 1976) Capparis species, as a large spontaneous plant, that information concerning the morphology, ethnopharmacology, phytochemistry, and biological activities of caper plants (Tlili *et al.* 2011).

Capparis spinosa has been investigated for its biochemical contents, which are affected by multiple factors such as geographical and environmental conditions, harvest date and size, preservation procedures, genotype, and processing methods of extraction (Tlili, Khaldi, and Triki 2010). In addition, it has a considerable economic importance through the uses of its roots, buds and fruits in many food and pharmaceutical industries (Sher and Alyemeni 2010). Extracts of different parts of *Capparis spinosa* have been shown to possess biological activity against a large number of pathogens (Chopra *et al.* 1956), and is considered as a very important source of medicine for antifungal (Ghdeib 1999). Antifungal, antibacterial, anti-amoebic, and antiworm activities have been demonstrated (Asolkar *et al.* 1992; Guha Bakshi *et al.* 1999) antidiabetic, antihyperlipidemic (Eddouks *et al.* 2005).

Ant hepatotoxic (Gadgoli and Mishra 1999), potential inhibitor of NF-kappa B (Zhou *et al.* 2011a), anti-inflammatory (Zhou *et al.* 2010) and anticarcinogenic (Kulisic-Bilusic *et al.* 2012). Anti leishmania, antihepatotoxic, and anti-allergic activities (Jacobson and Schlein 1999; Gadgoli and Mishra 1999; Trombetta *et al.* 2005). In addition, a methanolic extract of the plant *Capparis spinosa* tends to be a healthy source of antioxidants and minerals, which can help to protect health and fight disease (Aliyazicioglu *et al.* 2013).

Moreover, *capparis spinosa* contained an appreciable level of vitamin C and The significant level of all these antioxidants. that exist the important nutritional and medicinal value of this caper to be used in the Mediterranean basin (Tlili, Khaldi, and Triki 2010). The Capers of *Capparis spinosa* are rich in phenolic compounds and flavonoids as reported in several studies. Such secondary metabolites generally play a role in abiotic stress responses widely associated with tolerance to heat (Wahid 2007).

also that the plant of *Capparis spinosa* extract by Hexane and Chloroform was rich in minerals like K, Ca, Mg and P. (Brindha.P and Manikandaselvi .s, 2014). the *Capparis spinosa* is suggested to be a good candidate for use in natural medicine with historical background (Bakr and El Bishbishy 2016).

2.1.4. *Paliurus spina-christi* mill fruit

The scientific name of *Paliurus* is *paliurus spina-christi* Mill , the classification of plant .(Tuzlacı and Tolon 2000; Tutin *et al.* 1976)

Kingdome: Plantae

Division: magnoliopsida

Sub classs: Rosidae

Order: Roales

Family: Rhamnaceae

Genus and species: *Paliurus spina-christi*

Paliurus spina-christi belonging to Rhamnaceae family commonly known as Jersalem thorn, Garland thorn or crown of thorns (Rushforth 1999) is a traditional Mediterranean and Asiatic medicinal plant (Polunin *et al.* 1965), and is a perennial thorny shrub of wide spread distribution in dry and rocky places in the Mediterranean region and Asia (Brantner A, Males Z 1996),this plant grows along the Adriatic coast and on the islands (Polunin *et al.* 1965), and is one of the few woody species from the Dobrogea dry plane (Ciocârlan 2011)

This species is known by local names such as “Karaçalı”, “draga dikenî”, “öküz gözü”, “çaltı”, “ilme”, “çaltı dikenî”, “sarı çalı”, “sarı diken”, “çalı bakıldağı”, “çantı dikenî” (Tuzlacı and Tolon, 2000; Bulut and Ad 2011).It's used as diuretics, against diarrhea and rheumatism in traditional Croatian herbal medicine (Grlic 1986) and is used as diuretic, antirheumatic, hypo cholesterol emic, tonic, expectorant for children, anti-inflammatory, antitussive, tonic as well as in the treatment of cough, diarrhea, chronic obstructive pulmonary disease, abdominal pain, stomach pain, nausea, diabetes, nephralgia, kidney stones, sore throat, bronchitis, atherosclerosis, haemorrhoid, urinary tract infection in traditional Turkish herbal medicine (Bulut and Ad 2011; Zor *et al.*, 2017).

also that different extracts of this species have antimicrobial, hypolipidemic, antioxidant, and antigenotoxic activities(Zor *et al.*, 2017; Brantner A, Males Z 1996). It is also used against rheumatism in ethno medicine (Gene *et al.* 2013).

that ethanol and ethyl acetate extracts from branches and leaves of *Paliurus spina-christi* have significant antioxidant and anti-inflammatory activity(Ali ŞEN 2018a).However, the methanolic extract of plant *paliurus spina-christi* at low concentration it act as antioxidant but while at higher concentration it act as pro-oxidant (Taha Ahmed *et al.* 2013). In addition, the fruits contain alkaloids (Velcheva *et al.* 1993), generally plant parts contain flavonoids and tannins (Brantner and Males 1999)

The chemical investigation of *paliurus spinachristi* fruits and leaves were rich source of flavonoids (isoquercetin hexoside, quercetin, naringenin-c-diglycoside, quercetin 3-orhamnoglucoside 7-o-rhamnoside, quercetin-3-o-triglycoside, quercetin 3-o- α -rhamnosyl (1 $\rightarrow$ 6)- β -galactoside, quercetin-3 o- α -rhamnosyl (1 $\rightarrow$ 6)- β -galactoside-7-o- rhamnoside, hyperoside, quercetin-3-o-[- β -xylosyl(1 $\rightarrow$ 2)- α -rhamnosyl (1 $\rightarrow$ 6)]- β -glucoside, quercetin-3-rutinoside-7-rhamnoside, isoquercitrin, rutin, kaempferol-3- glycoside), tannins (epigallocatechol, gallicatechol, catechol, gallicatechin, gallicatechin, (+) and (-) catechin, epigallocatechin, catechin hydrate, proanthocyanidin b dimer), phenolic acids (5-caffeoyl quinic acid 5-pcoumaroylquinic acid), coumarins, amino acids, fatty acids, and alkaloids (Kayalar E *et al* 2016),(Zor *et al.* 2017),(Kemertelidze EP , et al 1999),(Brantner A, Males Z, 1996; Brantner and Males 1999b).

Antibacterial activity, mainly on Gram-positive bacteria, of the ethanolic extracts from different plant parts was found ,*Paliurus spina-christi* demonstrate a clear inhibition zone against gram positive and negative bacteria (Brantner A, Males Z, 1996).

2.1.5. Purslane *Portulaca oleracea* herb

The plant of species *oleracea* and Preferred Scientific Name *Portulaca oleracea* herb Linnaeus , the Preferred Common Name Purslane names of wild purslane, moss rose (Iwu 1993; Byrne, R. & McAndrews 1975; Ahmad and Askari 2015; Azuka *et al.* 2014; Cherukuri Vidyullatha *et al* , 2013).

Kingdom: Plantae

Superdivision: Spermatophyta

Division - magnoliophyta

Clade: Angiosperms

Class: magnoliopsida

Subclass - caryophyllidae

Order: Caryophyllales

Family: Portulacaceae

Species: *P. oleracea*

Genus: *Portulaca*.

The definition *Portulaca* comes from the Latin words "portare" which means "to bear" and "lac" which means "milk," and refers to the plant's milky sap (Rashed *et al.* 2003; Iwu 1993) *Portulaca oleracea* L is listed in the World Health Organization as one of the most used medicinal plants and it has been given the term 'Global Panacea'(Dweck 2001). The use of this plant as a vegetable, spice and medicine has been known since the times of the ancient

Egyptians and was popular in England during the middle Ages (Lanska 1992). It is an annual green herbaceous plant widespread in Mediterranean, central European and Asian countries (Rashed *et al.* 2003). Although orthodox medicine has been accepted by some populations of the world yet greater percentage still relies on natural remedies because of their cheap and easy availability *Portulaca oleracea* has been reported as a global panacea due to its several medicinal uses (Lim and Quah 2006). The aerial parts of the plant are used in many countries as a diuretic, febrifuge, antiseptic, antispasmodic and vermifuge (Xiang *et al.* 2005). Garden purslane, pigweed, moss rose, pusley, little hogweed, common purslane, and purple flowered purslane are some of the common names for the herb. The above-ground sections of common purslane, known as *Portulaca oleracea* L. (Portulacaceae; K: Pallpena) in Kudistan Iraq, are used as a fresh vegetable in salads or combined with lentils to make a special soup called “Pallpena Batrsh.” (Ahmad and Askari 2015). Also, this plant has different names in various ethnic groups in Nigeria. It is known as “Ntioke”, “Ntilimoke”, “Ntiike” or “Idiridi” in Igbo; “Esan omode” or “Papasan” in Yoruba; “Babbajibji” and in another country (Burkill 1985).

It is widely used not only as an edible plant, but also as a traditional Chinese herbal medicine. The main active ingredients of *P. oleracea* are flavonoids (Yan *et al.* 2012). Previous pharmacological investigations have indicated that flavonoids from *P. oleracea* have antioxidant (Erkan 2012), anti-inflammatory (Tadic VM *et al.* 2012), wound healing (Anitha *et al.*, 2011), anti-tumour effects (Xu *et al.* 2011) and anti-bacterial (Cushnie and Lamb 2005). Some of these activities have also been shown in different *portulaca oleracea* extracts (Chan *et al.* 2000; Rashed *et al.* 2003; Lim and Quah 2006; Shen *et al.* 2013; Ullah *et al.* 2013).

Portulaca oleracea cultivars could be used as a source of minerals and antioxidants, especially for functional food and nutraceutical applications (Uddin *et al.* 2012), that purslane *Portulaca oleracea* grown in different region has different antioxidant capacities according extract solvent (Güngören *et al.* 2017). In addition the *Portulaca oleracea* L. plant of to the highest values of the minerals as calcium, magnesium, phosphorus and potassium. thus, the inclusion of these parts as food and drug because replete with therapeutic agents (Ezeabara *et al.* 2014), also the common purslane *Portulaca oleracea* has more mineral composition than ornamental ones. that, both types of purslane are safe for human consumption and very good vegetable food crops for natural minerals, antioxidants, and medicinal purposes (Alam *et al.* 2014), therefore, that the aqueous extract of *Portulaca oleracea* contains several pharmacologically active constituents, that act in part on postsynaptic α -adrenoceptors and by interference with trans membrane calcium influx (O. PARRY, 1988).

That ethanol extract of *Portulaca oleracea* leaves play a beneficial role in the treatment of alcohol induced tissue damage, which could be one of its therapeutic values.(Hefnawy and Ali 2015), and also this Ethanol extract of this plant is preferable for extraction of saponin, tannin and terpenoid whereas both aqueous and ethanol extracts would produce good level of alkaloid, flavonoid, phenol and sterol (Ezeabara *et al.* 2014)

Therefore, the use of this Water-Methanol Extract of part plant *P. oleracea*, it can be applied in other food industries as a natural antioxidant to prevent the oxidation of vegetable oils instead of synthetic antioxidants (Jalali Mousavi *et al.* 2015). Also Methanol extract of the leaf and aerial parts of *P. oleracea* showed antimicrobial, antioxidant and anti-inflammatory activities and these pharmacological properties may justify the folkloric uses of the plant.(Agyare *et al.* 2015a). Another hand, the extract methanol of plant fraction is very rich in total polyphenols and exhibits a good antioxidant activity especially for scanning free radicals (IC₅₀ = 78.44 mg/ml). in addition, that encouraging for the recovery of this species as an excellent medicinal and alimentary plant (Karoune 2017).

In addition, a health properties of this plant content a high of nutrients and the nutraceuticals, especially antioxidants, purslane is also a very likely candidate as a useful cosmetic ingredient.(Sicari *et al.* 2018; Liu *et al.* 2000; Gallo *et al.* 2017). The *purslane P. oleracea* as a raw material at affordable cost and a good source of several compounds that have a positive impact in human health (Erkan 2012). Many varieties of purslane grow in a wide range of climates and regions. It is known to have higher nutritional value than major cultivated vegetables, with its considerable b-carotene, ascorbic acid and a-linolenic acid content (Liu *et al.* 2000). Thus, the purslane as the highest phenolic compounds include kaempferol, quercetin, apigenin and lipid peroxidation and the richest source of a-linolenic acid as a vegetable and a rich source of antioxidants. (Simopoulos *et al.* 2013; Erkan 2012). Water was reported as the major constituent of purslane stems and leaves. Among the 27 fatty acids detected in the plant, the most abundant was linolenic acid, followed by palmitic and oleic acids. Oxalic, citric, acotinic, fumaric, citric, malic and oxalic acids were the organic acids that were found to exist in the plant (Oliveira *et al.* 2009)

The MWE/UPLC–MS/MS method, an effective method for quality evaluation of *Portulaca oleracea* will be of great importance for quality assessment and should, therefore, be useful in assisting clinical applications(Liang *et al.* 2014). The anti-microbial ability of *portulaca oleracea* is also uniquely noted given the various pathogens this plant has been demonstrated to inhibit. The prevention of bacterial growth such as *Neisseria gonorrhea*,

Staphylococcus aureus, *Escherichia coli* and *Bacillus subtilis* with a 70% ethanolic extract of *portulaca oleracea* (Elkhayat *et al.* 2008).

2.1.6. *Artemisia haussknechtii* herb

The plant scientific Name *Artemisia haussknechtii* Boiss or *Artemisia hololeuca* Boiss herb. & Hausskn The plant was described in Boiss. 1875 (Dolatyari and Vallès 2014; Nafisi and Ghahremaninejad 2014).

Kingdom: Plantae

Tribus: Anthemideae

Subtribus: Artemisiinae

Synonyms: *Artemisia ucranica* Besser ex Nyman Class

Order: Asterales

Family: Asteraceae

Genus: *Artemisia*

Species: *Artemisia haussknechtii*

The genus *Artemisia* (compositae), with the common Persian name of ‘DERMANE’ (Schaffer and Mozaffari 1996; Ahmadi *et al.* 2002). All *Artemisia* Spp. are used for treatment of infectious diseases such as malaria, hepatitis and other diseases causing by helminthes, fungi, bacteria and viruses.(Yu *et al.* 2003; Juteau *et al.* 2002; Aniya *et al.* 2000), includes 34 species that can be found as wild plant all over Iran of which two are endemic (Zargari 1993). Local people in the western part of Iran, thats used some *Artemisia* species s tonic, and as anti-helminthic and *Artemisia haussknechtii* (Boiss) in the treatment of dyspepsia and other gastrointestinal disorders as traditionally , and also Antimicrobial effects of some endemic *Artemisia* in Iran such as *A. diffusa*, *A. oliveriana*, and *A. turanica* (Khanahmadi M *et al*, 2009; Ramezani, et al, 2004).

That *Artemisia haussknechtii* leaf extract with various groups of phytochemicals such as phenols and flavonoids had suitable ability in green synthesis of Ag, Cu and TiO₂ NPs. Also, Ag and Cu NPs had more antibacterial activities compared to TiO₂ NPs (Alavi and Karimi 2017). on the other hand, the *Artemisia haussknechtii* ethanolic extract showed good anti-oxidant activity and also extract inhibited the growth of both grams positive and gram negative bacteria and fungi (Khanahmadi M *et al*, 2009). The extract and essential oil of *A. haussknechtii* have been shown to possess antioxidant and anti-microbial activities(Khanahmadi M *et al*, 2009). *Artemisia haussknechtii* Boiss. is used in food as a flavoring, in perfumes and in pharmaceutical products (for its functional properties) in Iran (Abad *et al.* 2012). However, according to the outcomes obtained from previous researches on ten *Artemisia species*

(*Artemisia santonicum* L., *Artemisia taurica* Willd., *Artemisia spicigera* K. Koch, *Artemisia herba-alba* Asso, *Artemisia haussknechtii* Boiss., *Artemisia campestris* L., *Artemisia araratica* Krasch., *Artemisia armeniaca* Lam., *Artemisia austriaca* Jacq, and *Artemisia abrotanum* L.), collected from different localities throughout Turkey. However, that artemisinin or any of its related analogues were not detected in all investigated *Artemisia* species of Turkish origin (Nurgün *et al.* , 2007). In addition, the ethno-medical uses and health benefits of various *Artemisia* spp. and general information about bioactive compounds and free radicals (Nigam *et al.* 2019).

2.1.7. *Prunus microcarpa* seed

The scientific name of plant *Prunus microcarpa* C. A. Meyer (var. *pubescens* Bornm.) and Synonyms is *Prunus antilibanotica* (Uzun 2016)(Mati 2009)

Kingdom: Plantae

Glade: Angiosperms

Order: Rosales

Family: Rosaceae

Subfamily: Amygdaloideae

Genus: *Prunus*

Species: *P. microcarpa*

Binomial name: *Prunus microcarpa*

Prunus microcarpa one of the wild stone fruits worth exploring is boiss (syn. *Cerasus microcarpa*). It belongs to the Rosaceae family and has a shrubby habit with a height range of 0.5 to 3.5 meters. It can be found in abundance in the Kurdistan Mountains, as well as in the forest zone of Kurdistan, and on rare occasions in the lower margin of the thorn - cushion zone. (Townsend 1964). It has white or red flowers with erect sepals. The fruits are ovate in shape, black and red in color, and have a scrobiculate stone with a shallow depth. It is well suited to harsh winter and dry-hot summer conditions, and it may have the ability to be a dwarfing rootstock with early bearing, or a valuable new source of germplasm (Nas *et al.* 2010; Ahmad and Askari 2015)

A local name of *prunus microcarpa* in Kurdish language is Belaluk (بلاروك), Arabic name is (كرز صغير الثمرة), and english name is little cheny. Nothing is known about its chemical composition, and literature survey indicates that no work has been reported with regard to the flavonoids on the fruit of *prunus microcarpa* (Harborne 1965). The orthodox diabetes drugs have been discovered to have important antioxidant activity. These *Prunus microcarpa* extracts

may have the ability to reduce the impact of free radicals, which may be one of the causes of tissue distraction and insulin tolerance in diabetics (Lachin and Reza 2012).

That the plant *Prunus microcarpa* was content a high compound of rutin (glycoside flavonoid) as concentration 57.081% (Hussain 2004). The *prunus microcarpa* can be considered as characterizing the large gene pool that contributed to the domestication process of cherries. Furthermore, the conservation of the highly diverse native populations of Iranian wild the plant of *prunus microcarpa* species should be mainly considered (Mohammadi *et al.* 2019).

2.1.8. *Silybum Marianum seed*

The Scientific name of plant *Silybum Marianum* (L.). Fruct. Sem. Pl. 2:378 1791(*Carduus marianum*), the Species: *S. marianum* (Hammouda *et al.* 1968; Pieroni *et al.* 2016)

Kingdom: Plantae

Clade: Asterids

Order: Asterales

Family: Asteraceae

Genus: *Silybum*

Species: *S. marianum*

Binomial name: *Silybum marianum* (L.)

Silybum marianum L. (milk thistle) is an important herb native to the region extending from Southern Europe to North Africa. The plant is cultivated throughout the world for the medicinal potential of its seeds. The main active compound in the seeds is a flavonoid known as silymarin, Silibinin is the major pharmacologically active compound extracted from the seeds of the milk thistle *silybum marianum* (Mourelle *et al.* 1989; Valenzuela and Garrido 1994; Al-Anati *et al.* 2009; Pieroni *et al.* 2016; Holm *et al.* 1997; Greenlee *et al.* 2015). A local name of plant *silybum marianum* were given in the Latin alphabet, following the rules of Sorani Kurdish as chaw baz (چاو باز) and milk thistle *Silybum marianum* , Arabic: Qortom (قرطوم), Harshaf barri (حرفش بري), Shouk el-diman (شوك الدمن), Shouk el-nasara (شوك النصاري) Berber: Tatoura Doujnilourman. English: *Carduus marianus* L. (botanical synonym), Milk thistle, Holy thistle. French: Chardon marie, Chardon argente, Artichaut sauvage (Hammouda *et al.* 1968; Pieroni *et al.* 2016; Ahmad and Askari 2015).

Milk thistle (*Silybum marianum*) is a widely used traditional herbal/dietary supplement around the world for its strong hepatoprotective activity against several toxins and for regenerating damaged hepatic tissues (Mourelle *et al.* 1989; Valenzuela and Garrido 1994), including cirrhosis, acute or chronic viral hepatitis and necrosis (Jalali and Najafzadeh

2016). Also that is increasingly used in oncology research and treatment settings,(Al-Anati *et al.* 2009; Pieroni *et al.* 2016; Holm *et al.* ., 1997; Greenlee *et al.* 2015), and to be a good source of healthy edible sprouts (Vaknin *et al.* 2008).

That indicated that having multiple pharmacological activities including antioxidant, hepatoprotectant, anti-inflammatory agent, antibacterial, antiallergic, antiviral, antimutagenic, antineoplastic, and antithrombotic agent (El-demerdash *et al.*, 2015). this plant have the potential to be transposed successfully in the context of modern medical interventions such as liver transplantation surgery(Ligeret *et al.* 2008),also be responsible for possible herbal based dietary supplements-induced liver injury (Fenclova *et al.* 2019), and It protect against drugs and chemicals-induced hepatotoxicity (Upadhyay *et al.* 2007; Res 2011),

Additionally, the treatment with methanolic extract of *S. marianum* could significantly attenuate the nickel induced oxidative hepatotoxicity through its antioxidant properties. suggesting their anti-oxidant and immunity powerful (Bouhalit and Kechrid 2018). Also useful as natural antioxidants are ethanol extracts of various parts of the pappi and roots of *Silybum marianum* L. (Sun *et al.* 2016), the silybin B is abundant in the seeds of *Silybum marianum*, which may be a good source of the compound. The silibinin may have a chemopreventive effect against mediated colon carcinogenesis (Rodriguez *et al.* 2018; Sangeetha *et al.* 2012).

The analysis of *silybum marianum* stems showed lower concentration of all ingredients except analysis of metal ion of Si which is found in higher quantity compared to other parts of the herb (Ghafor *et al.*, 2014). That *S. marianum* of blue and white flowering data showed nearly the same values of phytochemicals and antimicrobial active against all gram positive bacteria and demonstrated moderate to significant antibacterial activity against tested pathogens, while inactive against gram negative bacteria and fungi (Muhammad *et al.* 2011). these weeds could be a potential source of natural antioxidants in future for the pharmaceutical industry, because all the species of plant having high phenol and flavonoid content, and strong antioxidant potential in terms of DPPH radical scavenging activity and total antioxidant capacity. (Singla and Pradhan 2019).

2.1.9. *Pistacia eurycarpa* Yale

The Taxon Name is *Pistacia eurycarpa* Yaltirik and Synonym *Pistacia atlantica* Desf ssp. kurdica Rech.f, *Pistacia atlantica* Desf var. kurdica Zohary. it is closely related to *Pistacia atlantica* (Rhodes and Maxted 2016; AL-Saghir 2010), and the common name is Pistache, Pistachio

Kingdom: Plantae
Phylum: Tracheophyta
Class: Magnoliopsida
Order: Sapindales
Family: Anacardiaceae
Genus: *Pistacia* L.

The *Pistacia eurycarpa* Yale, is a member of Anacardiaceae family (Kafkas 2002). *Pistacia eurycarpa* belongs to the secondary Gene Pool of *Pistachio P. vera* (El Zerey-Belaskri 2019), and also *Pistacia eurycarpa* has also been reported to be present in the Key Biodiversity Area (KBA) and Important Plant Area (IPA) of Sargalu in Iraq, which consists of forest steppe in the Zagros Mountains; although this area was deemed 'unprotected' in 2010 (Nature Iraq 2010).

Various researches have shown that this plant, an endemic plant in Turkey, is rich in fat content and contains high levels of fatty acids (Demirci *et al.* , 2001; Kafkas *et al.* 2007). A Common names in Iraq of Kurdish is Dareben or Dar qezwan (قزوان), Arabic botom, Iran Baneh, Pakistan *khinjuk* (خنجک), Turkey Bittım, Cyprus Gum Tree, English Mastic tree or Terebinth (Ahmed, 2017).

There are few studies on *Pistacia khinjuk* regarding to the chemical composition of essential oils (Tavakoli *et al.* 2013) and flavonoids (Kawashty *et al.* , 2000; Pietta 2000). The extract of hull plant represents a high antioxidant activity and total phenolic content, suggesting their possible application in pharmaceutical and food technology (Hatamnia, *et al.* , 2015). Therefore, many studies were carried out on antioxidant and free radical scavenging activities of phenolic compounds in genus *Pistacia* L. (Hosseini *et al.* ,2005; Gourine *et al.* 2010; Tomaino *et al.* 2010; Farhoosh *et al.* , 2011; Hatamnia and Abbaspour, 2014). It was determined that the extracts obtained from this plant have antimicrobial and antifungal effects. There are soaps which used in the area of cosmetics are made from *Pistacia eurycarpa* oils (Kordali *et al.* 2003; Diğrak *et al.* 2001).

The antimicrobial activity of the hydro distilled oil was determined for ethno medical uses of the oleo-gum resin on skin diseases (Demirci *et al.* , 2001), also the extract (Essential Oil) from *Pistacia eurycarpa* Yalt. especially in vapor phase as a natural inhibitor to control *A. flavus* growth in food materials such as maize.(Bahramian 2019). Furthermore, that the extracts of *P. eurycarpa* fruit and its peel significantly accelerated the growth of *M. tuberculosis* by

shortening the growth duration of *M. tuberculosis* at least by one third. and so it has the potential for use as a gene donor for crop improvement (ŞİMŞEK *et al.* 2013).

2.2. Phytochemical of plants

Plant metabolites are divided into two categories: primary and secondary metabolites. Compounds known as primary metabolites play essential roles in plant growth as photosynthesis, respiration and growth (Crozier *et al.* 1997).

Primary metabolites include phytosterols, acyl lipids, nucleotides, proteins, amino acids, and organic acids. Secondary plant metabolites have a wide variety of structures and are found in a small number of plant species. They are an expression of a species' individuality, though they can be detected in high concentrations in some situations. Some of these compounds were discovered to play an important role in plant resistance in a variety of ways. (Crozier *et al.* 1997).

They can aid reproduction by attracting pollinators, serve as deterrents against herbivores, and/or provide protection against harmful sun radiation during the plant's life cycle. Some play a part in human well-being as well. Moderate long-term intakes of some groups of these compounds may help to suppress and/or eliminate cancer and other chronic diseases, as neurodegenerative disease, cardiovascular cancer, type 2 diabetes, hypertension, and the aging phenomenon (Katalinic, *et al.*, 2010).

2.2.1. Antioxidant activity of Medicinal Plants

An antioxidant is a molecule that prevents or substantially slows the oxidation of substrates, even though it is present at lower concentrations than the oxidized substrate (Matkowski 2008; Gouveia *et al.* 2013). Plant antioxidant compounds play an important role as health protection factors. The main characteristic of an antioxidant is its ability to trap free radicals. Antioxidants such as phenolic compounds including flavonoid, phenolic acids, tannins, and phenolic diterpenes scavenge free radicals such as peroxide, hydroperoxide, or lipid peroxyl, and thus inhibit the oxidative mechanisms that led to degenerative diseases and are of great benefits to our health (Pietta 2000; Chung *et al.* 1998; Amarowicz *et al.* 2004).

Antioxidant activity against reactive species implicated in aging and chronic, infectious, inflammatory, neurological, and degenerative disorders is one of the most important biological properties of phenolic compounds. (Soto *et al.* 2011; Vermerris. and Nicholson. 2007). The

relation between oxidative stress and human health, namely in the pathogenesis of various diseases and disorders has become a serious issue and as such has attracted much attention in the scientific community. Below pressure, the human body will create more harmful species, such as reactive oxygen species (ROS), (hydroxyl radicals, hydrogen peroxide, and superoxide anion radicals), than enzymatic antioxidants (superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase), and non-enzymatic antioxidants (α -tocopherol (vitamin E), ascorbic acid (vitamin C), glutathione, carotenoids, and flavonoids), producing cell damage (Katalinic, *et al.*, 2010).

When antioxidants are in short supply, this effect is amplified. As a result, the human diet should be supplemented with antioxidant compounds, which may be synthetic or natural. Foods are vulnerable to oxidation in and of themselves (Matkowski 2008). Therefore, by inhibiting and delaying oxidation, antioxidants have become common in food products as a way to extend shelf life and reduce waste and nutrient losses. (Tsuda *et al.* 1994). Through a long time, synthetic antioxidants such as butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), propyl gallate (Pg) and tert-butyl hydroquinone (TBHQ) were utilized as additives in foods and beverages. However, since they are linked to elevated levels of cytotoxicity and carcinogenic effects, their use is now limited. As a result, there is a vital need to explore natural compounds with antioxidant properties and low toxicity (Branen 1975; Matkowski 2008; Gouveia *et al.* 2013), and there has been a widespread desire to switch to natural food additives in favor of synthetic ones (Howell 1986).

Antioxidants are classified into three categories: blocking antioxidants, scavenging antioxidants, and restore and de novo antioxidants, and they all perform essential roles throughout the body's protection mechanism (Niki 2010). The preventing antioxidants, which inhibit the production of reactive oxygen and nitrogen species (ROS/RNS), is the first line of protection. Active organisms are easily removed by scavenging antioxidants so they can harm biologically important molecules. Carotenoids, for example, can mechanically or chemically scavenge singlet oxygen (Howell 1986; Niki 2010). There is a developing interest in materials showing antioxidant properties, which can be gotten as food ingredients or as specific preventive pharmaceuticals (Konczak and Sydney 2014). The antioxidant activity of a biological substance can be measured using a many methods. The ABTS⁺ and DPPH radicals are the two most often used radicals. DPPH is a free radical that can be obtained without any preparation, whereas ABTS⁺ requires enzymatic or chemical reactions. In the terms of ease, speed and sensitivity the most used. Because presence of the antioxidant leads to the

disappearance thr nature of radical chromogens to stimulate RONS (Brand-Williams *et al.* 1995; Miller and Rice-Evans 1997).

2.2.2. Phenolic and Tannin compounds

Plants that have one or more phenol units in their molecules are called phenolics. based on their structure (Naczk and Shahidi 2006). Also, the phenolics are primary sources of natural antioxidants are, which can be found in vegetables, nuts, beans, leaves, stems, barks, and in bananas that may occur in all parts of the plants (Pratt and Hudson 1990).

Plant phenolics are multifunctional and can serve as reducing agents (free radical terminators), metal chelators, and singlet oxygen quenchers, among other things. Consumption of foods and drinks high in phenolic content has been linked to a lower risk of heart disease (Verhagen 1989). Also, the flavonoid compounds, cinnamic acid derivatives, coumarins, tocopherols, and polyfunctional organic acids are the most common plant phenolic antioxidants (Hertog *et al.* 1993).

Phenolic compounds are a class of low and medium molecular weight plants develop secondary metabolism both during normal growth and in reaction to stress conditions such as inflammation, wounding, and UV radiation. (Naczk and Shahidi 2006). Free radicals are scavenged from phenolic compounds and aromatic amines. The second line of protection in vivo is this kind of system. The third line of protection consists of a variety of enzymes that restore damage, eliminate waste by-products, and re-establish lost function (Niki 2010).

Since phenolic compounds are bioavailable in foods (fruits, teas, herbs, grains, coffees, spices, and so on), humans can eat them on a regular basis to the recommended daily intake ranges from 25 mg to 1 g (Stalikas 2007). More than 5000 phenolic compounds have been identified (Pyrzynska and Biesaga 2009). In recent decades, the impact of phenolic compounds on human health has been well known. Phenolic compounds have been linked to important biological and pharmacological properties such as anti-inflammatory, antimutagenic, and anticarcinogenic activities (Fang *et al.* ., 2007; Stalikas 2007). However, the phenolic compounds have been used in various medical uses since ancient civilizations. Willow bark, for example, was used in the centuries B.C. to relieve aches and pains, as well as alleviate fevers and inflammation; the active ingredient is a phenolic acid (Mahdi 2010). Therefore, when consumed in significant amounts, certain phenolic compounds can be toxic. The ability of phenolic compounds to precipitate proteins, shape complexes with polysaccharides, influence lipid metabolism, and interfere with metal ion bioavailability are all recorded negative

properties (Fang *et al.*., 2007) (Stalikas 2007). Besides, the existence of at least one aromatic ring with one or more hydroxyl groups characterizes the chemical composition of phenolic compounds. They can be present in the free form or conjugated to sugar which organic acid residues, and are categorized depending on the number and organization of carbon atoms in the basic structure. They can also be divided into classes based on the number of phenol compounds in each, as well as the structural elements that hold these rings together. As a result, flavonoids and phenolic acids are distinguished (e.g. hydroxybenzoic and hydroxycinnamic acids), the simple phenolics, phenolic acids, coumarins, flavonoids, stilbenes, lignans, lignins, and tannins are many of the phenolics that can be divided into different classes (Naczki and Shahidi 2006).

Tannins that use tannic acid as a building block, on the other hand, have a relatively high molecular weight and are divided into two types: hydrolysable and condensed tannins. Proanthocyanidins is another name for condensed tannins. Fruits and fruit oils have a lot of condensed tannins in them. Fruit astringency and color are linked to this special group of phenolics (Landete 2012; Serrano *et al.* 2009; Koleckar *et al.* 2008). Moreover, proanthocyanidins are considered to have a number of biological properties that are critical to human health (Chung *et al.* 1998). Tannins, which are naturally found in the human diet as a component of plant-based foods, were once thought to be anti-nutrients. These compounds tend to bind to carbohydrates and proteins, thus decreasing the absorption and ingestion of these nutrients (Chung *et al.* 1998). On the other hand, Tannins have been shown to have anticarcinogenic, antimutagenic, antimicrobial (antibacterial, antiviral), and antioxidant properties (Chung *et al.* 1998; Cos *et al.* 2004; Koleckar *et al.* 2008; Serrano *et al.* 2009).

Proanthocyanidins were also shown to have cardioprotective properties, as their use reduced the chance of heart attacks. Tannins have also been shown to help with diabetes. Tannin use was observed to reduce blood glucose levels by slowing glucose absorption in the intestine. As a result, those with diabetes should consume a diet high in tannins. Tannins, on the other hand, have been discovered to have anti-inflammatory properties. (Koleckar *et al.* 2008; Serrano *et al.* 2009).

Tannins have also been related to biochemical effects such as lowering blood pressure, lowering serum lipids, and regulating immune responses (Chung *et al.* 1998). Tannins have also been linked to anti-oxidant properties. Free radical scavenging, transition metal chelation, inhibition of prooxidative enzymes, and lipid peroxidation are among the antioxidant properties of these substances (Cos *et al.* 2004; Koleckar *et al.* 2008).

Based on knowledge of the physiological and health effects of concentrated tannins present in plant food products, it is important to estimate the relative content of these tannins in plant food products. Fruits and vegetables, which are high in phenolic compounds, are considered the richest sources (Kapur and Kapoor 2001). The degree of ripening of the plants, as well as the environmental conditions of the production fields, have the greatest effect on the concentrated tannin content of fruits. Furthermore, how fruits are stored and handled will influence their tannin content (Serrano *et al.* 2009).

2.2.3. Flavonoid Plant Content

Flavonoids are secondary metabolites found in a variety of plant tissues, and best known for their distinctive red, blue, and purple pigments, it including the seeds, berries, nuts, bark, roots, stems, and flowers. Plants have been shown to contain over 4000 structurally distinct flavonoids (Brahmachari 2011). Because of their high capacity to donate electrons or hydrogen atoms, many of them have been shown to outperform many well-known antioxidants, such as ascorbate (vitamin C) and -tocopherol (vitamin E), based on *in vitro* antioxidant assays. (Hernández *et al.* 2009).

The molecular composition of flavonoids determines their ability to function as antioxidants. Flavonoids' free radical scavenging behaviors and antioxidant properties are influenced by the arrangement of hydroxyl groups and other molecular features in their chemical structure. (Saxena *et al.* 2012). Plant-derived flavonoids are consumed in the amount of one gram a day by humans (Kuhnau 1976), with drinks such as tea, chocolate, red wine, and alcohol containing large quantities of flavonoids, and herbal remedies containing flavonoids being used all over the planet (Gurib-Fakim 2006; Harborne 1986). Also, the Tea, fruits (apples, blueberries), dark chocolate, and red wine all have moderate to high levels of flavonoids, while broccoli and fruit juices (cranberry and orange) have comparatively low levels (Beecher 2003)

The flavonoids are a complex category of polyphenolic compounds found in the plant kingdom that have a broad variety of bioactive properties, like antioxidant and antibacterial properties as well as cardiovascular system protection. Flavonoids are antioxidants that can scavenge free radicals, boost antioxidant enzyme levels, and protect body processes from free radical harm. (Sandhar *et al.* 2011). Furthermore, flavonoids have a typical biosynthetic basis, which results in a structural feature: the 2-phenyl chromone moiety (or 3-phenyl chromone for isoflavonoids). The degree of oxidation of the central pyranic moiety, which can be opened or

recyclized in a furanic moiety, can be used to classify flavonoids into different classes. Several thousand flavonoids have already been identified, the majority of which are water soluble. And if a few of them, such as rutinoid and hesperidoid, are poorly soluble in these solvents, they are easily extracted from plant material using water or alcohol as hetero sides. (Min-Hsiung *et al.* 2010; Wu *et al.* 2016).

2.2.4. Anthocyanin Plant Content

Anthocyanins are polyphenylated pigments with many phenyl groups in their composition. Flavonoids with an open C-ring are closely related. They're called "anthocyanidins," and the family of compounds that give plants their purple, blue, and red colors, pigments. Anthocyanins (glycosides), and anthocyanidins (aglycones) are two types of anthocyanins, which are both water soluble and provide many flowers and fruits (Di Carlo *et al.* 1999; Gurib-fakim 2006; Ignat *et al.* 2011; Wallace *et al.* 2013).

Anthocyanins can be found in flower petals, roots, leaves, and fruits. They're polyphenolic pigments that come in a variety of shades, ranging from red to purple. A review of the literature shows the existence of over 700 anthocyanin compounds (Castañeda-Ovando *et al.* 2009). A high consumption of foods high in anthocyanins can have health benefits for cancer, ageing diseases, obesity, neurological diseases, arthritis, diabetes, and bacterial infections. Anthocyanins are found mainly in fruits with red or blue color such as strawberries and other berries, grapes, wine, and tea and their examples include cyanidin and pelargonidin (Hudson *et al.* 2000; Olsson *et al.* 2004; Yi *et al.* 2006; De Pascual-Teresa and Sanchez-Ballesta 2008; Hassan and Abdel-Aziz 2010; Sancho and Pastore 2012; Nijveldt *et al.* 2018).

A balanced diet high in anthocyanins has a number of notable health benefits. Certain chronic diseases, such as hyperglycemia, are protected by anthocyanins (Tsuda *et al.* 2003), and inhibit the increase of tumor cells in humans (Wang *et al.* 2012; Gao *et al.* 2013). Because of these advantages, anthocyanins are becoming more common and are being used in drinks (Zhang *et al.* 2003), and in the treatment of a many of human pathologies (Wang *et al.* 2012) (Tsuda *et al.* 2003), the measurement of anthocyanins may be useful in deciding when vegetables should be harvested (Chen *et al.* 2015) as well as evaluating the consistency of fresh and manufactured goods for industries and rural producers (Poiana *et al.* 2012), since the composition of this pigment and other phenolic compounds is directly linked to their efficiency UV-visible spectrophotometry is one method for determining plant anthocyanin material (Lees

and Francis 1972; Vera de Rosso 2008), and nuclear magnetic resonance (NMR) (Ella Missang *et al.* 2003).

Some of these procedures, however, necessitate lengthy processing times, high costs, and specialized reagents. The biological properties of anthocyanins, as well as their health benefits, are driving efforts to develop and improve extraction, quantification, and identification approaches in natural sources using systematic and speedy methods. (Ignat *et al.* 2011; Pappas *et al.* 2011). Depending on the pH of the organelle, the same anthocyanin may appear in various colors in different plants. They differ from their plastic equivalents, who follow the Beer–Lambert law and produce consistent colors. Anthocyanin color stability is determined by a number of variables, including the anthocyanin's composition and concentration, pH, temperature, and the existence of complex agents such as phenols and metals. Antioxidant activity of anthocyanins has been shown to be higher (Castañeda-Ovando *et al.* 2009), than vitamins C and E (Bagchi *et al.* 1998). Free radicals can be captured by the phenolic hydrogen atoms in these compounds (Rice-Evans *et al.* 1996); this is the reason for its anticarcinogenic activity (Kamei and Kopeček 1995).

2.3. Pathogenic Bacteria

Bacterial infections are widespread in the tropics, and they account for a large part of the disease burden. (Kumar, P. and Clark 1990). In the treatment of bacterial infections, chemotherapy is the most popular solution. Resistance to antibiotics is a big issue for antibiotics in clinical practice, as it can lead to drug failure (McKeegan *et al.* 2002). In vitro, a large number of phytochemicals from various chemical groups have been found to have inhibitory effects on various microorganisms. (Cowan 1999), and some plant extracts have been found to be effective against both gram negative and gram positive bacteria (Nascimento *et al.* 2000). Presently there is no single antibacterial chemical entity extracted from plants that is used in clinical practice (Gibbons 2004).

The toxicity, high cost, and poor effectiveness are among the other issues of antibiotics. As a result, new antibacterials must be sought on a regular basis to address these issues. The human body includes approximately 10^{13} cells and hosts approximately 10^{14} bacteria, which are predominantly commensals and play an important role in human biological activities. These bacteria are mostly settled in certain areas of the body (known as niches) such as the skin, conjunctiva of the eye, oral cavity, upper respiratory, gastrointestinal, and urogenital tracts (CP Davis 1996). Antibiotics, which are strong against pathogenic bacteria, can disrupt the usual

microflora's ecology in both quantitative and qualitative ways (Edlund and Nord 2000). Therefore, any microflora can be selectively inhibited by medicinal plants (Ahn *et al.* 1990). As a result, medicinal plants can have benefits over antibiotics. Medicinal plants contain a wide range of compounds with well-known beneficial properties (RojAS and IYENGAR 1985; Chopra *et al.* 1992). Candidates for developing new antimicrobial drugs are agents that can either prevent or destroy pathogens while causing least or minimal toxicity to host cells. Antimicrobial effects of medicinal plants have been increasingly identified from various parts of the world in recent years (Grosvenor *et al.* 1995; Ratnakar and Murthy 1995; Diniz *et al.* 1996; Nimri *et al.* 1999; Sharma *et al.* 2001).

Plant extracts with target sites other than those used for antibiotics are expected to be effective against drug-resistant bacterial pathogens. However, there is a scarcity of knowledge on medicinal plants' operation in this regard (Nair *et al.* 1995; Lee *et al.* 2003; Saxena and Kumar 2007). Other research mentioned, that chosen 45 Indian medicinal plants to be tested against bacteria that are multi-drug resistant, including *Staphylococcus aureus*, *Salmonella paratyphi*, *Escherichia coli*, *Shigella dysenteriae*, and *Candida albicans*. The medicinal plants were chosen based on their common uses in India (32 plants) as well as 13 plants' antimicrobial activity (Chopra *et al.* 1992; Ahmad *et al.* 1998; Mehmood *et al.* 1999).

So many natural antioxidants have been derived from different varieties of plants; phenolic compounds, which are commonly distributed in the plant kingdom, are at the forefront. In fact, anticancer, antibacterial, antifungal, and antiviral properties have been related to these compounds (Ksouri *et al.* 2009).

3. MATERIALS AND METHODS

3.1. Materials

3.1.1. Apparatus and equipment

Table 3.1. The list of laboratory apparatus and equipment

Apparatus and Equipment	Company	Country
Water bath	Memmert	Germany
Water distillatory	DLabTech	Korea
Centrifuge	Hettich	England
Magnetic stirrer hot	Labtech	India
Refrigerator	Revco (Thermo)	USA
Sensitive Balance	Mettler	Switzerland
pH meter	Gallen-Kamp	England
Moisture analyzer DBS	KERN & Sohn GmbH	Germany
Digital Camera	SONY	Japan
Candle Jar	Merck	Germany
Micropipette different volumes	Gallen-Kamp	Germany
Fume Hood	Hyland	Germany
Micro tubes 1.5ml clear	Axygen, INC.	U.S. A
Sterile transport swabs	Capon S.P.A	Italia
Disposable micropipette tip		Germany
Disposable plastic Petri dishes (15 x100)	Sun	Jordan
Glass screw- cupped tube (16 x 13 mm)		
Disposable plastic test tube	Barco pharma company	China
Glass tube	Samco	
Glass Slide	Sail Brand	China
UV-vis spectrophotometer	SHIMADZU	Germany
Bunsen burner	Shenma	China
Nexera model Shimadzu UHPLC	HiMedia	India
Forceps		China
Filter papers	Cito test	China
L shaped spreader	Citotest	China
Syringe	Aysecure	UK
Para film	Bemis	U.S.A.
Autoclave	Lab Tech	Korea
Incubator	Memmert 2000	Germany
Electric Oven	Memmert 2000	Germany
Densi Check (Turbidity meter)	Bio Merieux	French
Microscope (CH30	Olympus	Japan

3.1.2. Reagents, stain and solution

Table 3.2. Culture media used in this study

Medium	Company	Country
Hilton agar	Merack KGaA	Germany
Hilton broth	Merack KGaA	Germany
Antibiotic Disk	Bioanalyse	MDC (Erbil)
Petri dish		Turkey

Table 3.3. Chemicals and solutions

Reagents	Company	Country
Distilled water	KSU	KSU
Ethanol	Alpha- chemica	Germany
Hexan	Fisher scientific	Germany
N-butanol	Alpha- chemica	Germany
FeSo4	Merack KGaA	Germany
HCl	Prolabo	England
DPPH	Himedia	India
Folin	Merack KGaA	Germany
Na2CO3	Merack	Germany
NaNO2	Merack	Germany
AlCl3	Merack	Germany
NaOH	Merack	Germany
CH3COONa	Merack	Germany
KCl	TEKIM	Turkey
Methanol	Merack	Germany
Formic acid	Sigma Aldrich	Germany
BHT	Samarra	Iraq
Normal saline		India

3.1.3. Microorganism

All bacteria were obtained from (Media Diagnostic Center) MDC Erbil Accredited America, CAP*(Collage of American Pathology), these strain are external quality control from CAP except all bacteria which is referred to ATCC strain of ATCC 29213 to (*Klebsiella pneumoniae* FMC).

Table 3.4.All the organisms of bacteria

Organism Code	Organism	Source
B ₁	<i>Staphylococcus aureus</i> (ATCC 29213)	CAP*
B ₂	<i>Pseudomonas aeruginosa</i> (ATCC 27853)	CAP
B ₃	<i>Bacillus subtilis</i> (ATCC 6633)	CAP
B ₄	<i>Enterococcus faecalis</i> (ATCC 29212)	CAP
B ₅	<i>Enterobacter aerogenes</i>	CAP
B ₆	<i>Klebsiella pneumonia</i>	CAP

CAP*: College of American Pathology

3.2. Method and preparation

3.2.1. Collected Plants and Identification

This plant survey occurred in villages (Balakian, Hawdian, and Kawna Gund) at Soran city from the North of Iraq. This work is included survey, identification and collection of plants during their season. Plants parts such as seed, flower, leaf, and fruit were collected for the identification process as shown in (Figure 3.1.), as well as the taxonomist identification was by (Dr. (Abdullah Sh. Sardar), Erbil, Iraq: College of Education, Salahaddin University. The plants were collected carefully for one year's in difference months as illustrated in (Table 3.5.), after that storage and dried of plants parts under shade to be prepared for experimental analysis.

3.2.2. Types of plants

The plants were used for our study *Eremurus Spectabilis* leaf (Estrelek), *Prosopis farcta* fruit (Xrnuk), *Capparis Spinosa* fruit (Talamar-kabar), *Paliurus, spinachristi* fruit (Drka Ze-Estre), *Portulaca oleracea* herb (Palpena), *Artemisia haussknechtii* Boiss herb (Giyaband), *Prunus microcarpa* seed (Blaruk), *Silybum Marianum* seed (kaluxan-pshelok) and *Pistacia eurycarpa* Yale (Qasuwaan).

Table 3.5. Identified and collecting plants

No.	Plant Binominal Names	Common Names	During Collect	Plants Herbarium Code
1.	<i>Eremurus Spectabilis</i>	Foxtail lilly	20-April- to 10-july/ 2017	7706
2.	<i>Prosopis farcta</i>	Syrian mesquite	2-Agust to 10-Oct/ 2017	7707
3.	<i>Capparis Spinosa fruit</i>	caper bush	5-july to 10-september/ 2017	7708
4.	<i>Paliurus spinachristi fruit</i>	Jerusalem- thorn, garland thorn, Christ's thorn	05-june to 20-july/ 2017	7709
5.	<i>Portulaca oleracea</i>	Common purslane, moss rose	09-julyto 25-Agust/ 2017	7710
6.	<i>Artemisia haussknechtii</i>	Dermane	10-mayto 20-june/ 2017	7711
7.	<i>Prunus microcarpa seed</i>	mall-fruited cherry	15-juneto 5-Agust/ 2017	7712
8.	<i>Silybum Marianum seed</i>	Cardus marianus, milk thistle, Marian thistle, blessed milk thistle, Marian thistle, Mary thistle, Saint Mary's thistle	27-May to 10-july/ 2017	7713
9.	<i>Pistacia eurycarpa Yale</i>	Buotim , Pistache, Pistachio	20-sep. to 20 Dec/ 2017	7714



1. *Eremurus Spectabilis* Bioss leaf



2. *Prosopis farcta* fruit



3. *Capparis Spinosa* fruit



4. *Paliurus spinachristi* fruit



5. *Portulaca oleracea* herb



6. *Artemisia haussknechtii* herb



7. *Prunus microcarpa* seed



8. *Silybum Marianum* seed



9. *Pistacia eurycarpa* Yale fruit.

Figure 3.1. Medicinal natural plants displayed in aerial part

3.3. Chemical Analysis and Extraction Methods

Six analysis crude extraction was used, which is involved percentage yield extraction, total condensed tannin, total phenolic, total flavonoid, total anthocyanin determination, and DPPH radical scavenging activity. All steps of this analysis we're doing at Kahramanmaraş Süçü Imam University. Also, the Phytochemical analyses were done at Harran university (HÜBTAM LABSİS).an electronic weighing balance and soaked with five different solvents Distilled water, (Ethanol: Distilled Water v/v) aqueous, Ethanol, (Hexane: Ethanol v/v) aqueous, and Hexane, as shown in (Table 3.6.), and (Figur 3.2.). In this study, four methods utilized for extraction as following in below:

3.3.1. Conventional extraction

For this method, 6.25 grams of plant powder sample and 125 ml of Distilled Water, (Ethanol: Distilled Water v/v) aqueous, Ethanol, (Hexane: Ethanol v/v) aqueous, and Hexane were mixed in around the bottom flask separately. The mass of the plant (g) to the amount of the solvent (ml) was 1:10. In addition, stirring is used. The extraction is performed at boiling point temperature for 1 h. At the end of the extraction cycle, this mixture of solvent and sample was separated using the Buchner funnel and filter paper. Finally, filtrates were collected and stored in the cabinet (Fume Hood) to evaporate (Dhanani *et al.* 2017; Ollanketo *et al.* 2002; Dhobi, *et al.*, 2009).

3.3.2. Microwave Assisted Extraction

Microwave milestone NEOS system was utilized. 6.25grams of the plant was added and mixed with 125 ml of Distilled Water, (Ethanol: Distilled Water v/v) aqueous, Ethanol, (Hexane: Ethanol v/v) aqueous, and Hexane separately in the flask. Additionally, a magnetic stirrer bar was applied. Extraction was carried out for 30 min and temperatures were standard for solvents and different powers (250,300 watts) were used. Finally, this mixture of solvent and sample was separated by using Buchner funnel and filter paper. Finally, filtrates were collected and stored in the cabinet (Fume Hood) to evaporate (Mandal, et al., 2007)(Dhobi, et al., 2009; Latha 2007).

3.3.3. Orbital Shaker Extraction

In this method, 6.25 grams of powder plants extracted by using 125 mL of each solvent Distilled Water, (Ethanol: Distilled Water v/v) aqueous, Ethanol, (Hexane: Ethanol v/v)

aqueous, and Hexane are mix in around the bottom flask separately for 24 hours at 35 0C in a temperature-controlled orbital shaker (Gallenkamp, UK). Then this mixture of solvent and sample was separated by using Buchner funnel and filter paper. Finally, filtrates were collected and stored in a cabinet (Fume Hood) to evaporate (Rohman *et al.* 2010; Ashraf *et al.* 2016).

3.3.4. Fume Hood

One of the primary safety devices in laboratories that handle chemicals in the laboratory fume hood. A fume hood removes gases, vapors, and fumes from the laboratory. A well-designed fume hood, properly installed and maintained, will provide substantial protection for an experimenter (provided the experimenter is aware of the capabilities and limitations of the particular fume hood). This datasheet explains the functions and the various safety features of fume hoods, and it provides sufficient information to enable the researcher to evaluate the protection afforded by a particular fume hood (Ii 1980).

Table 3.6. During heating and time for all method and solvents extraction

Common Solvents used	Conventional Extraction	Microwave Extraction	orbital shaker Extraction
Distilled water	After Boiling point 1 hour, 250 C ⁰	After Boiling point 30 Minute, 250 watt, 150 rpm, 110 C ⁰	24 hour, 150 rpm
Ethaol: Distilled (v/v) water (50/50%) ml	After Boiling point 1 hour, 250 C ⁰	After Boiling point 30 Minute, 250 watt, 150 rpm, 110 C ⁰	24 hour, 150 rpm
Ethaol	After Boiling point 1 hour ,100 C ⁰	After Boiling point 30 Minute, 250 watt, 150 rpm, 78 C ⁰	24 hour, 150 rpm
Hexane: Ethaol (v/v) (50/50%) ml	After Boiling point 1 hour, 100 C ⁰	After Boiling point 30 Minute, 250 watt, 150 rpm, 78 C ⁰	24 hour, 150 rpm
Hexane	After Boiling point 1 hour, 50 C ⁰	After Boiling point 30 Minute, 300 watt, 150 rpm, 69 C ⁰	24 hour, 150 rpm

Notes: P=Plant; CE= Conventional Extraction; MWE= Microwave Extraction; OSE= Orbital Shaker Extraction; S= Solvent; W=Distilled water; W: E= distilled water: ethanol aqueous (v/v) ; E=Ethanol; H: E= hexane: ethanol aqueous (v/v); H=Hexane; Plant = 10 (g); Solvents =200(ml); Solvent: Solvent = 100+100 (ml)



Conventional Technique



Microwave Technique



Orbital shaker Technique

Figure 3.2. All techniques of (Conventional, Microwave, orbital shaker) extraction for all plants.

3.3.5. Determenation of Extraction Yield %

The extraction yield was a measure of the solvent's efficiency to extract specific components from the original material and it was defined as the amount of extract recovered in mass compared with the initial amount of dry plant. It is presented in percentage (%) and was determined for each technique tested .

Plant powder (6.25 g) was extracted with 125 ml Distilled Water, (Ethanol: Distilled Water v/v) aqueous, Ethanol, (Hexane: Ethanol v/v) aqueous, and Hexane separately, by using various extraction methods described in Conventional extraction, Extraction by Microwave-Assisted Extraction (MAE) technique and Orbital shaker (OSE). The yield percentage of the extract was determined by using the following formula (Murugan and Parimelazhagan 2014; Zhangetal.,2009;Anwar,etal.,2009).

$$\text{Extraction Yield \%} = \frac{X}{Y} * 100$$

Where,

X is the oven dry weight of extract (g),

Y is the oven dry weight of the sample (g).

3.3.6. Determination of total condensed tannin

This parameter was carried out by Shimadzu UV-vis spectrophotometer. The extraction solution was prepared by mixing 0.05 g of Fe₂SO₄, 95 ml N-butanol, and 5 ml HCl (35%). For determining the condensed tannin, 0.01 g of both plant powder and mimosa tannin were put separately in a test tube and 10 ml of extraction solution was added and placed in a water bath for heating for 1 h. The absorbance was measured at 580 nm wavelength (Karaogul *et al.* 2018; Makkar and Singh 1995).

3.3.7. Determination of total phenolic compounds

The total phenolic compounds were quantified following the Folin Ciocalteu method as described by Dewanto et al (2002). An aliquot of the diluted extract was added to 180 mL of distilled water and 20 mL of Folin–Ciocalteu reagent. The mixture was then shaken and allowed to stand for 6 min before adding 1.60 mL of 7% Na₂CO₃. The solution was then adjusted with distilled water to reach a volume of 3 mL and thoroughly mixed, after incubation in the dark, absorbance at 760 nm was read against the blank. The total phenol content of plant extracts was expressed as milligrams of Gallic acid equivalents per gram of dry weight (mg GAE/g DW)

from a calibration curve with Gallic acid. All samples were analyzed in three triplicate (Dewanto *et al.* 2002).

3.3.8. Determination of Flavonoid Contents

The flavonoid contents of individual extracts were measured as per the Dowd method (Arvouet *et al.* 1994) An aliquot of 1 mL of extract solution (25–200 µg/mL) or quercetin (25–200 µg/mL) were mixed with 6.4 mL distilled water, and then 0.3 mL of 10% (w/v) AlCl₃ was added with 0.3 mL (1 M) CH₃CO₂K. After that, 2 ml of NaOH was added; then the solution was then incubated for 30 min at ambient temperature. The absorbance was recorded at 510 nm against the control. The data are expressed milligrams quercetin per gram (mg QE/g) of dry extract.

3.3.9. Total Anthocyanin measurement using pH differential method

Total anthocyanin was determined following the method described previously (Rapisarda *et al.* 1999) with some amendment . The extract was mixed with a buffer solution of pH 1.0 (0.02 M KCl and 980 ml of distilled water with 1.86g KCl, with added 27M HCl), and a buffer solution of pH 4.5 (960 ml of distilled water with 54,43g sodium acetate, with added 20 M HCl) added respectively. The prepared mixture was homogenized and centrifuged twice for 15 min at 4°C at 5000 rpm; the absorbance was read at 512 nm and 700 nm. The anthocyanin was quantified following the spectrophotometric technique suggested; the concentration of anthocyanin was specified to stratify the Lambert-Beer law. The recorded spectra in a Heλios spectrophotometer were amount at 25°C, Total anthocyanin amount was determined by the following formula (Rapisarda *et al.* 1999). The concentration (mg/L) of every anthocyanin was measured according to the following:

$$A = (A \cdot MV \cdot DF \cdot 1000) / (\epsilon \cdot 1)$$

Where

A: is the absorbance = (A_{λvis-max}) pH 1.0 – (A_{λvis-max}) pH 4.5

MW is the molecular weight (g/mol) = 449.2 g/mol for Cy-3-glc,

DF: is the dilution factor (0.2 ml sample is diluted to 2 ml, DF = 100), and ε is the extinction coefficient (L × cm⁻¹ × mol⁻¹) = 26,900 for Cy-3-glc, where L (path length in cm) = 1. For comparison, the same extinction coefficient was used for other standards to calculate the concentration of each anthocyanin and thus results reported is expressed as Cy-3 glc equivalents.

3.3.10. DPPH radical scavenging activity

The free radical scavenging activity of Distilled Water, (Ethanol: Distilled Water) aqueous, Ethanol, (Hexane: Ethanol) aqueous, and Hexane separately, of the crude plant 0.004 g was determined by the DPPH assay described by Blois (1958) with slight modification. In its radical form, (DPPH) 1, 1-diphenyl-2-picrylhydrazyl absorption level decreases at 517 nm with reduction of an antioxidant or a radical species. Briefly, 0.1 mM DPPH was prepared for ethanol or methanol extractions. Then 0.1, 0.2, and 0.3 ml of sample solutions mixed with methanol and ethanol up to 3 ml in a test tube, separately. Then, 1 ml of DPPH was added. The mixture was then shaken vigorously and placed at a dark room temperature for 30 min. Later, the absorbance was measured by Shimadzu UV-vis 1240 spectrophotometer at 517 nm. Butylated hydroxyl toluene (BHT) was used as a reference. Radical scavenging activity was expressed as the inhibition percentage of free radical by the sample and calculated with the following equation (Gülçin *et al.* 2010; Hamad *et al.* 2017; Ak and Gülçin 2008).

$$\text{Inhibition of DPPH radical scavenging activity (\%)} = \frac{A-B}{A} \times 100$$

Where,

A is the absorbance of DPPH,

B is the absorbance in the presence of sample and BHT.

3.3.11. Phytochemicals analysis in plant extraction by Liquid chromatography mass spectrometry LC-MS/MS

The plants were dried under the shade and ground into fine powder by using an electric blender, then, 6.25 g of powder was extracted with 125 ml solvent distilled water. Which by the method: Conventional extraction during 1 hour after starting boiling. After that the extraction solvents and then evaporated by Fume Hood at 30 °C for 24 h. until preparing extractive yield. hence, the crude extract plant was weight amount (0.005 g) add into tube were add (5 ml) of Methanol into the tube and then mixed by vortex instrument that takes 1 ml of the solution to add by micro peptide to vial after that put in LC-MS/MS Technique to get the result of the phytochemical analysis. LC-MS/MS analyses of the phenolic compounds were performed using a Nexera model Shimadzu UHPLC coupled to a tandem MS/MS instrument. The liquid chromatography was equipped with an LC-20AD two pieces' pump, DGU-20A3R degasser, CTO-10ASVP column oven, and SIL-20AC autosampler. The chromatographic separation was performed on a C18 reversed-phase Inertsil ODS-4 (30 mm × 100 mm, 2 µm) analytical column. The column temperature was fixed at 40oC. The elution gradient consisted of mobile phase A water (5 mM ammonium formate and 0.1 % formic acid), and mobile phase B methanol (5 mM ammonium formate and 0.1% formic acid). The solvent flow rate was maintained at 0.3 mL/min and the injection volume was settled as 2 µL. (Figure 3.3.). The gradient programmer of the LC device used to be constant at time (0,4,7 and 7.01) respectively, with the solvent A % (95,5,5,95) and solvent B% (5,95,95,5). MS detection was performed using Shimadzu LCMS 8040 model triple quadrupole mass spectrometer equipped with an ESI source operating in both positive and negative ionization modes. LC-MS/MS data were collected and processed by Lab Solutions software (Shimadzu, Kyoto, Japan). The multiple reaction monitoring (MRM) mode was used to quantify the analysts: the assay of investigated compounds was performed following two or three transitions per compound. The optimum ESI conditions were determined temperature at 250oC, heat block temperature,400 oC, nebulizing gas flow (nitrogen); 3 L/min and drying gas flow (nitrogen); 15 L/min (Ertas *et al.* 2015) (Koyuncu *et al.* 2021)



Figure 3.3. Technique of LC-ESI-MS/MS chromatogram

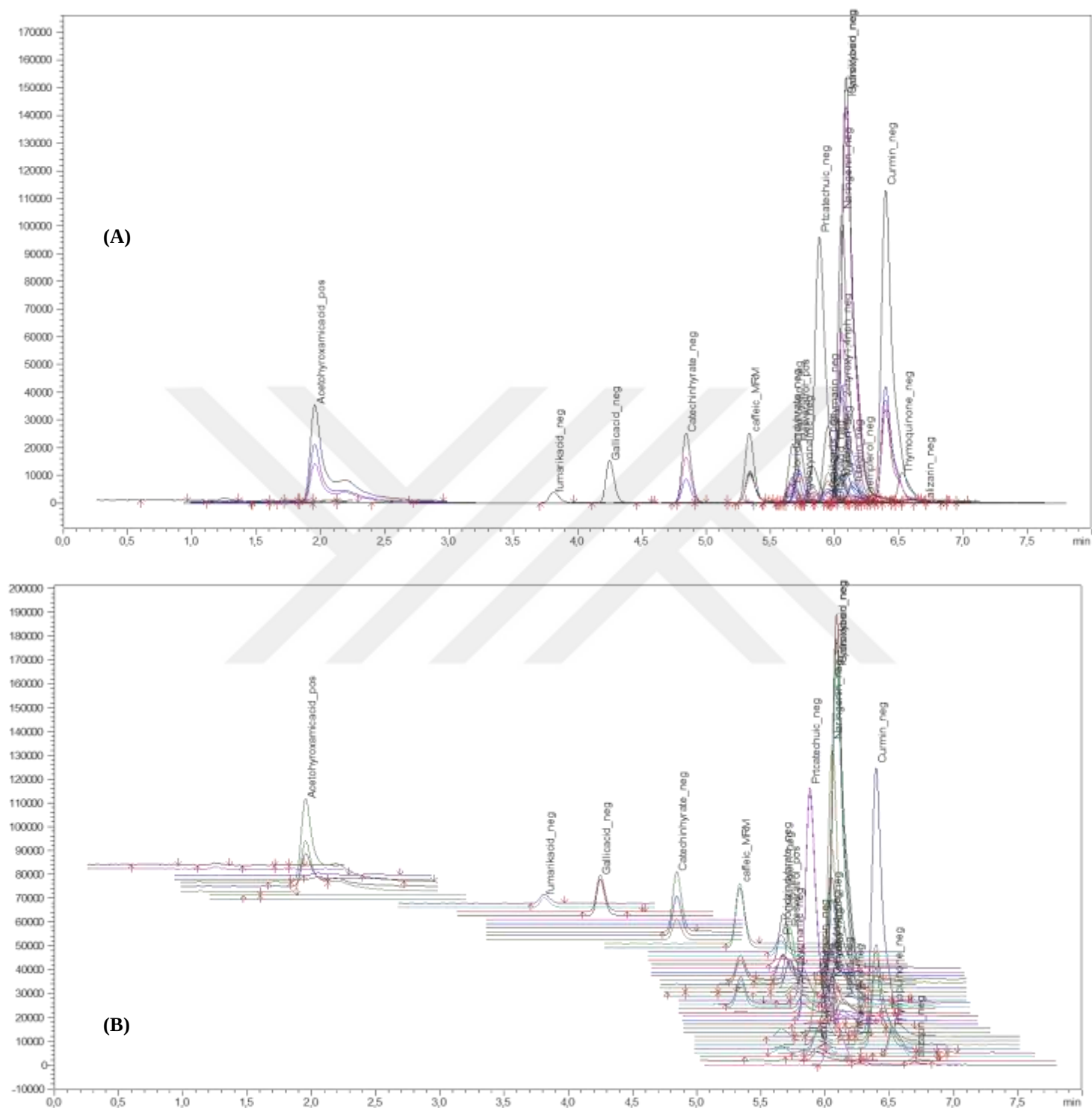


Figure 3.4. LC-MS/MS chromatograms of A: 250 ppb standard mix, B: *plant* extract. 1: Catechin hydrate, 2: Quercetin, 3: Acetohydroxamic acid, 4: Vanillic acid, 5: Resveratrol, 6: Fumaric acid, 7: Gallic acid, 8: caffeic acid, 9: Phloridzin dehydrate, 10: Oleuropein, 11: hydroxy cinnamic, 12: Ellagic acid, 13: Myricetin, 14: Silymarin, 15: Curmin, 16: Naringenin, 17: Kaempferol, 18: salicylic acid, 19: Hydroxy benzoik acid, 20: Butein, 21: luteolin, 22: alizarin, 23: Thymoquinone, 24: Protocatechuic acid ethyl ester, 25: 2-hydroxy 1,4 aftokiniton.

3.3.12. Analysis in solvents in extraction of Plants by Ion Chromatography (IC)

Ion chromatography (IC) is a rapid and sensitive technique for separating and analyzing solutions containing complex mixtures of ions (Basta and Tabatabai 1991; Kubáň *et al.* 2005; Small *et al.* 1975). The study was carried out with Shimadzu Prominence (HIC-20A Dual IC) Super Ion Chromatograph device brand Dual Flow-Line IC/ HPLC.

Approximately 0.001 g of the crude plant was weighed and then 10 mL distilled water was added and shaken by vortex instrument for 10 minutes to precipitate crude plant after that take 4 ml of solution added by micro peptide into vial injected into Ion chromatography technique. A representative separation using IC-CD of a standard solution containing the inorganic compounds identified in the crude plant. The optimized columns shim pack IC-SA3 and shim pack IC-C4 were performed (250 mm x 4.6 mm x 0.25 μ m), the elution of mobile phase of 3.6 mM sodium carbonate and 2.5 Mm oxalic acid at the 450C Column Oven Temperature. The solvent flow rate was maintained at 0.8 mL/min with a sample size of 50 μ L and injection volume was settled as 1600 μ L. the solvent flow rate at a flow rate of this technique was used for the separation of mineral compounds as anions and cation ((F⁻, ClO₂⁻, BrO₃⁻, Cl⁻, NO₂⁻, Br⁻, NO₃⁻, PO₄³⁻, SO₄²⁻, Li⁺, Na⁺, NH₄⁺, K⁺, Mg²⁺, and Ca²⁺) (Masson *et al.* 2005).



Figure 3.5. Technique of Ion Chromatography (IC)

3.4. Biological Study

3.4.1. Test on antibiotic activity of Oral rinse technique

3.4.1.1. Bacteria Oral rinse

In healthy people, the oral cavity had a well-balanced microbiome of roughly 1.0×10^{11} microbes/g of dental plaque and over 700 organisms on the tongue dorsum, buccal epithelium, rough and soft palates, and other surfaces (Aas *et al.* 2005). Several experiments shown that a disruption of this equilibrium, resulting in a rise in harmful microbes and a decline in beneficial microbes, induces oral diseases like periodontal disease and dental caries (Do *et al.* 2013)

3.4.1.2. Sample Collection and study area

The procedures were conducted aseptically according to the directorate of health - Erbil approved for healthcare, the samples were collected from thirteen healthy peoples, by collecting oral liquid wash, and diluted by normal saline in an anaerobic transport medium. After that, the collected samples were transferred to microbiological laboratory and preserved in refrigerator until running the experiments.

The isolated samples of bacteria were cultivated on nutrition agars, blood agars, and MacConkey agars. The isolated bacteria were morphologically characterized after 24 hours' incubation under certain conditions (37°C). moreover, the isolated bacteria were characterized on selective and differential media such as mannitol salt agar (P.Tille 2014).

3.4.2. Sterilization of media and materials

3.4.2.1. Autoclaving

Media was autoclaved for 15 minutes at 15 psi pressure (Bailey and Linko 1990). Except for CaC, which was sterilized by water bath if the autoclave temperature was less than 100°C and no pressure was specified by the manufacturer.

3.4.2.2. Dry sterilization

Materials such as glassware, conical flask and L shaped spreader were washed with detergent and rinsing with water and allowed to dry, the glassware was sterilized in a hot air oven at 200°C for two hours.

3.5. Culturing on Mueller Hinton broth and Mueller Hinton agar

100 ml of distilled water was added to the 21g media of Mueller Hinton broth and 38g Mueller-Hinton agar individually, after that put their on steering for the 15 minutes as well as sterilized by autoclaving at 15 psi pressure (121°C) for 15 min (MN Baily 1990).



Figure 3.6. Petri dish culture on mueller hinton broth and mueller hinton agar

3.5.1. Prepared of bacteria

2 ml of Mueller Hinton broth was homogenized into the tube after that take stock bacteria by swab to into tube, and incubated at 37°C for 24 h.



Figure 3.7. Preparation of the bacterial strains B1; *Staphylococcus aureus* (ATCC 29213), B2; *Pseudomonas aeruginosa* (ATCC 27853), B3; *Bacillus subtilis* (ATCC 6633), B4; *Enterococcus faecalis* (ATCC 29212), B5; *Enterobacter aerogenes*, and B6; *Klebsiella pneumoniae* (CCM 2318)

3.5.2. Disc diffusion method to assess the antimicrobial property of crude extract antibacterial test susceptibility testing and inoculated onto petri dishes

3.5.2.1. Determination of the antibacterial activity of plant extraction by disk diffusion method

The disk diffusion method also known as Kirby Bauer, was carried out according to the Clinical and Laboratory Standard Institute Guidelines National Committee for Clinical Laboratory Standard (CLSI). Firstly, the selective medium (Mueller-Hinton agar) was aseptically prepared and poured into sterilized petri plates, and the broth culture containing the isolated bacteria were compared to McFarland 0.5 to ensure the turbidity of the young cultures of bacteria (Cruz *et al.* 2006; Deliorman Orhan *et al.* 2012). After that, the bacteria were streaked continuously on MHA by using sterilized disposable swabs. The cultures were leaved to dryness, then the suspended discs with various concentrations of plant extract (50, 70, and 100 μ L/disk) were subjected on to culture media and directly incubated under certain conditions for 24 hours. The inhibition zones were detected in millimeters.

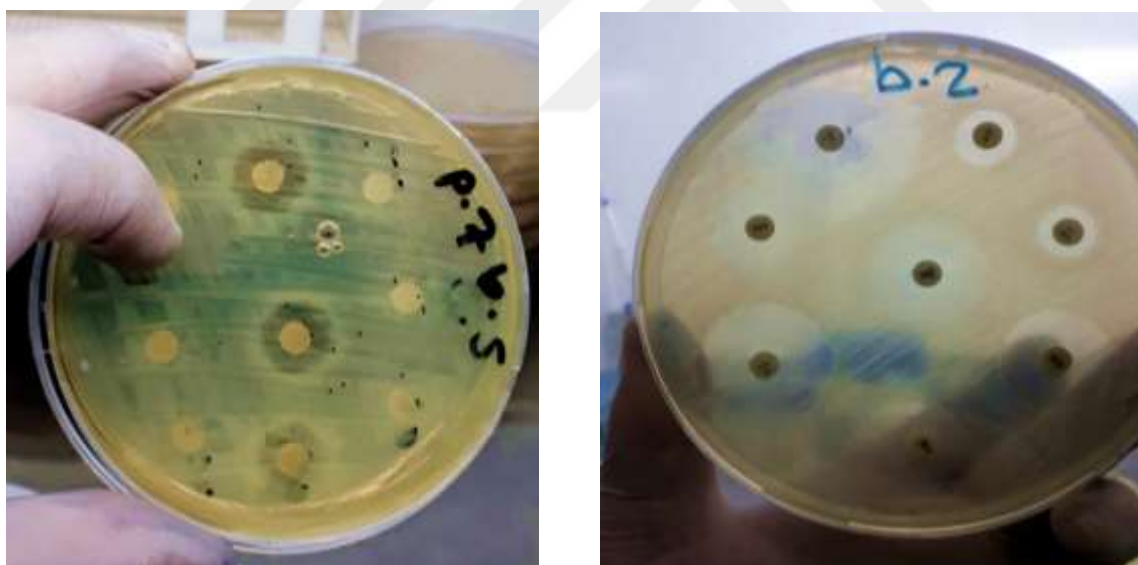


Figure 3.8. Zones of inhibition at dilution of plant extract against bacteria



Figure 3.9.Preparation of Microorganism.

3.5.2.2. Minimum inhibitory concentration (MIC)

The MIC is the lowest concentration of an antimicrobial that inhibits the visible growth of a microorganism after overnight incubation (Kaya *et al.* 2012). MICs were defined as the lowest concentration of boric acid and borax inhibiting visible growth of the bacteria.

One mililitre of extract concentration 100 mg/ml for five extract solvents were mixed with agar in serial decimal dilutions diluting 1 ml in 9 ml of ml in ml agar to get concentration range of 0.5 to 512mcg/ml. This is usually hours. All the bacteria suspensions were prepared by suspending 24h bacteria culture in sterile no of the bacteria suspension was adjusted to 1.0 McFarland standard (equivalent to 1.5×10^8 CFU/ml). may also be standardized based on Mueller Hinton broth, the Bacterial suspensions were adjusted to the logarithmic-phase growth to match the turbidity of a 0.5 McFarland standard (Cruz *et al.* 2006; Deliorman Orhan *et al.* 2012), yielding approximately 10^8 CFU/mL .

The same amounts of bacteria were added to all tubes and the tubes were incubated at 37 °C for 24 h. Each tube was examined for growth and compared to the control. Bacterial suspension added to a tube filled with nutrient broth without boric acid and borax was used as a positive growth control. A tube not containing nutrient broth was used as a negative growth control. The absence of growth was defined as antibacterial activity.



Figure 3.10. Minimum inhibition concentration (MIC) of plant extract against bacteria

3.6. Statistical Analysis

All of the statistical parameters calculated using (IBM SPSS) for (version 20), the obtained all values were exhibited as means $\pm$ S.E were calculated. The results of the two groups were compared using the analysis of the independent-samples T-test. An Expectation value (P-value) less than ($p < 0.01$) was considered as statistically significant (*). Whereas, the P-value more than ($p > 0.01$) was adopted as the criteria for a significant difference as statistically not significant.

4. RESULTS AND DISCUSSION

4.1. Determination of extraction yield %

4.1.1. Determination extraction yield % of *Eremurus Spectabilis Bieb*

In this research, the Extraction Yield of plant *Eremurus Spectabilis B.* leaf prepared by Conventional Extraction (C/E), microwave extraction (MW/E), and orbital shaker extraction (OS/E) techniques by different solvents such as distilled water, ethanol: distilled water (v/v), ethanol, hexane: ethanol (v/v), and hexane. The higher amount of extraction yield observed by conventional extraction technique with solvent distilled water was (CE: 65.37 %), while the lowest value found by the same method with hexane was (21.30 %). On the other hand, the higher amount of yield extraction by microwave extraction method with solvent of ethanol was (MWE 47.33 %), otherwise, the highest value extraction yield was obtained in orbital shaker hexane (OSE: 75.00 %), compared to the same method with different solvents and compared to other methods with different solvents in plant *Eremurus Spectabilis B.* as investigated in Table and figure (4.1).

Table 4.1.Extraction yield % of plant *Eremurus Spectabilis Bieb* areal parts

	C/ E (%)	MW/E (%)	OS/E (%)
Solvent	Mean ±S.E	Mean ±S.E	Mean ±S.E
DW	37.66±0.5	24.13±0.1	45.30±0.2
DW:E (v/v)	65.37±0.1	40.33±0.4	33.33±0.2
E	23.00±0.5	47.33±0.2	44.33±0.2
H:E (v/v)	21.41±0.3	43.50±0.5	70.33±0.3
H	21.30±0.2	22.66±0.2	75.00±0.5

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H).

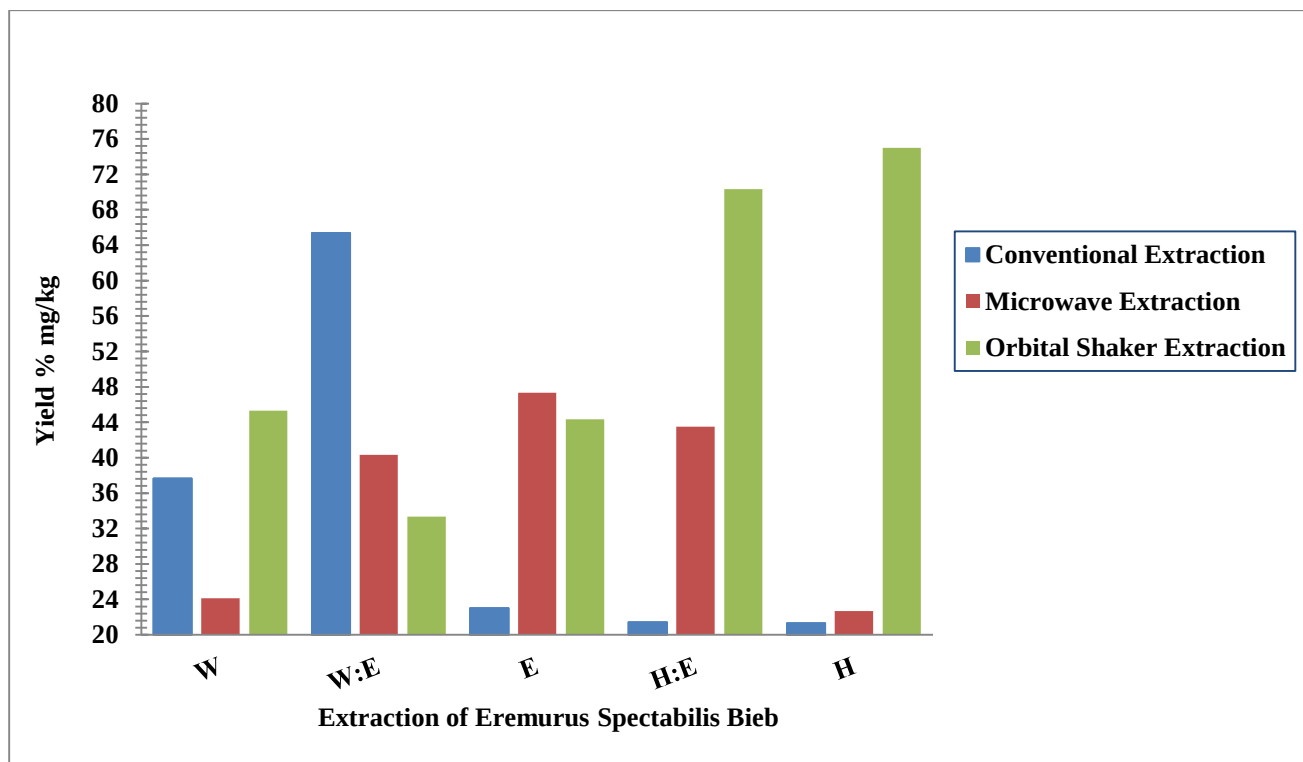


Figure 4.1. Extraction yield % of plant *Eremurus Spectabilis Bieb*

4.1.2. Determination extraction yield % of *Prosopis farcta*

Three different methods such as conventional, microwave, and orbital shaker extraction, were used for topmost extraction in seed of plant *Prosopis farcta* with five solvents. Our study that the topmost extraction values ranged between 20.63- 81.56 %, 28.85-42.05 %, and 28.0- 43.55 % by utilized techniques, with dissimilar solvents CE/H and DW/E, MW/E with solvents E and H, and OS/E with solvents DW and H, respectively. The results shown that, C/E with solvent E was more efficiency for the topmost extraction in seed of the plant *prospis farcta* due to the highest value (81.56 g/g) as shown in Table and Figure (4.2)

Table 4.2. Extraction yield % of plant *Prosopis farcta*

	C/ E (%)	MW/E (%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	31.95 $\pm$ 0.64	41.36 $\pm$ 0.28	28.00 $\pm$ 0.64
DW:E (v/v)	81.56 $\pm$ 0.28	35.50 $\pm$ 0.64	31.60 $\pm$ 0.40
E	41.50 $\pm$ 0.64	28.85 $\pm$ 0.47	38.20 $\pm$ 0.40
H:E (v/v)	20.63 $\pm$ 0.00	32.95 $\pm$ 0.62	41.15 $\pm$ 0.47
H	21.00 $\pm$ 0.64	42.05 $\pm$ 0.62	43.55 $\pm$ 0.85

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E), and Hexane (H).

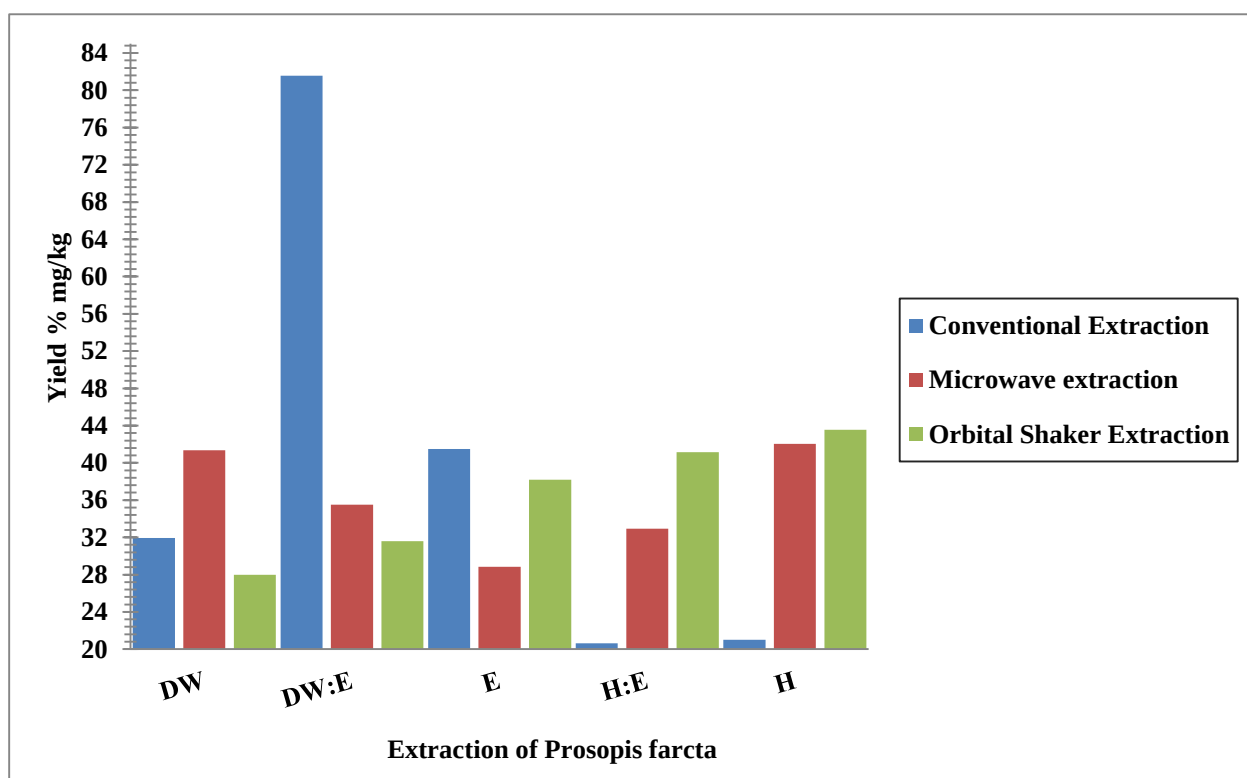


Figure 4.2. Extraction yield of *Prosopis farcta*

4.1.3. Determination extraction yield % of *Capparis Spinosa* fruit

The extract yield of plant *Capparis Spinosa* fruit was prepared by three different techniques with dissimilar solvents such as: distilled water, distilled water: Ethanol (v/v), Ethanol, and Hexane: Ethanol (v/v) and Hexane are summarized in Table and Figure (4.3). Among the extraction methods, a distilled water: ethanol conventional extraction showed the highest yield percentages (CE DWE: 65.99 %), (OSE: 58.90 %), and (MWE: 51.20 %) followed by orbital shaker and microwave extraction methods. Whereas, the orbit shaker showed the topmost extract yield with all the extract solvents, respectively.

Table 4.3.Extraction yield % of plant *Capparis Spinosa* fruit

	C/ E (%)	MW/E (%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	37.20 $\pm$ 0.40	51.20 $\pm$ 0.40	47.25 $\pm$ 0.47
DW:E (v/v)	65.99 $\pm$ 0.25	28.48 $\pm$ 0.40	29.10 $\pm$ 0.28
E	32.32 $\pm$ 0.40	37.60 $\pm$ 0.40	41.60 $\pm$ 0.70
H:E (v/v)	30.74 $\pm$ 0.28	45.90 $\pm$ 0.28	58.58 $\pm$ 0.28
H	31.15 $\pm$ 0.47	24.05 $\pm$ 0.47	58.90 $\pm$ 0.28

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H).

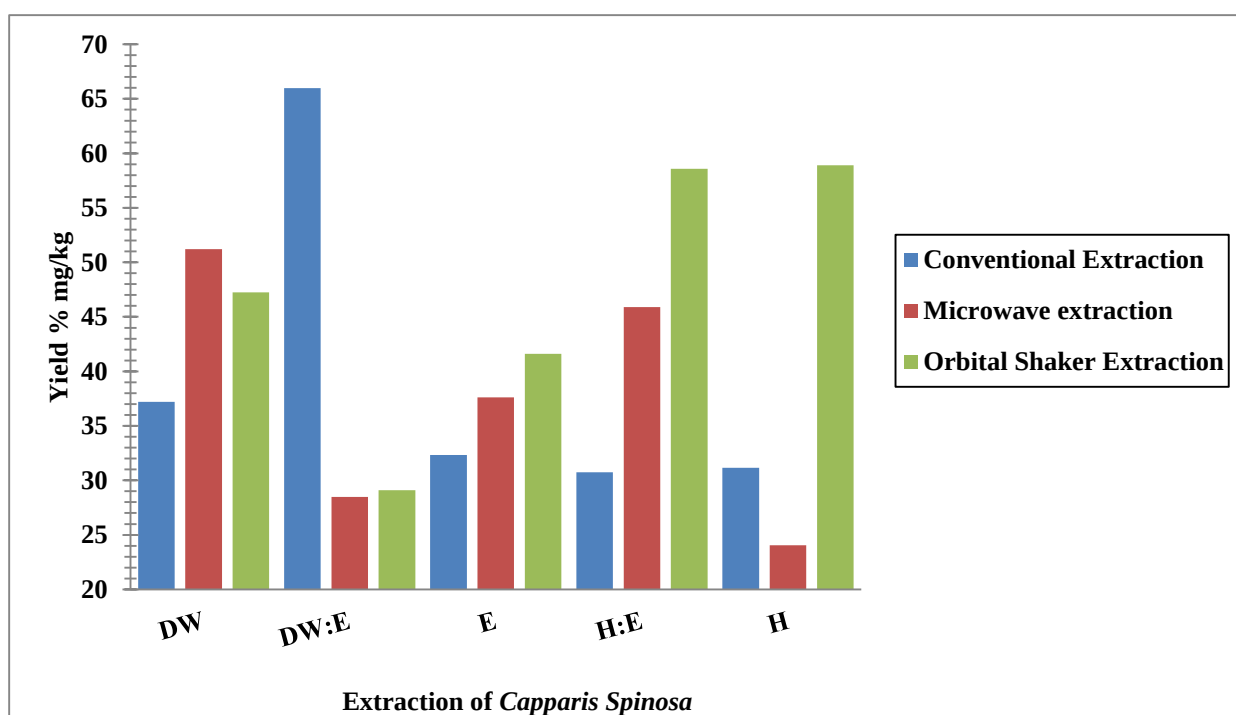


Figure 4.3. Extraction yield % of *Capparis Spinosa*

4.1.4. Determination extraction yield % of *Paliurus spinachristi* fruit

The yield proportion of the plant *Paliurus spinachristi* seed extracts prepared by three different techniques with five solvents. The results were provided in Table and Figure (4.4). The highest extraction yield was found for the usage of Distilled water: Ethanol in the conventional extraction method (OS/HE: 66.88 %). On the contrary, the smallest yields percentages were (CE/DWE: 61.60 %), and (MW/E: 40.00 %) respectively. However, the yield extraction from the method of conventional extraction was observed significant amount with all solvents, in comparison with other methods respectively. Furthermore, some of the other results in some solvent extracts with the method of extraction were proximity value to each other or same.

Table 4.4.Extraction yield % of plant *Capparis Spinosa* fruit

	C/ E (%)	MW/E (%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	37.92 $\pm$ 0.40	17.12 $\pm$ 0.40	36.64 $\pm$ 0.40
DW:E (v/v)	61.60 $\pm$ 0.40	28.96 $\pm$ 0.40	24.16 $\pm$ 0.40
E	27.90 $\pm$ 0.28	40.00 $\pm$ 0.40	29.60 $\pm$ 0.40
H:E (v/v)	49.05 $\pm$ 0.47	32.32 $\pm$ 0.40	66.88 $\pm$ 0.40
H	50.05 $\pm$ 0.25	25.12 $\pm$ 0.40	52.48 $\pm$ 0.40

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E), and Hexane (H).

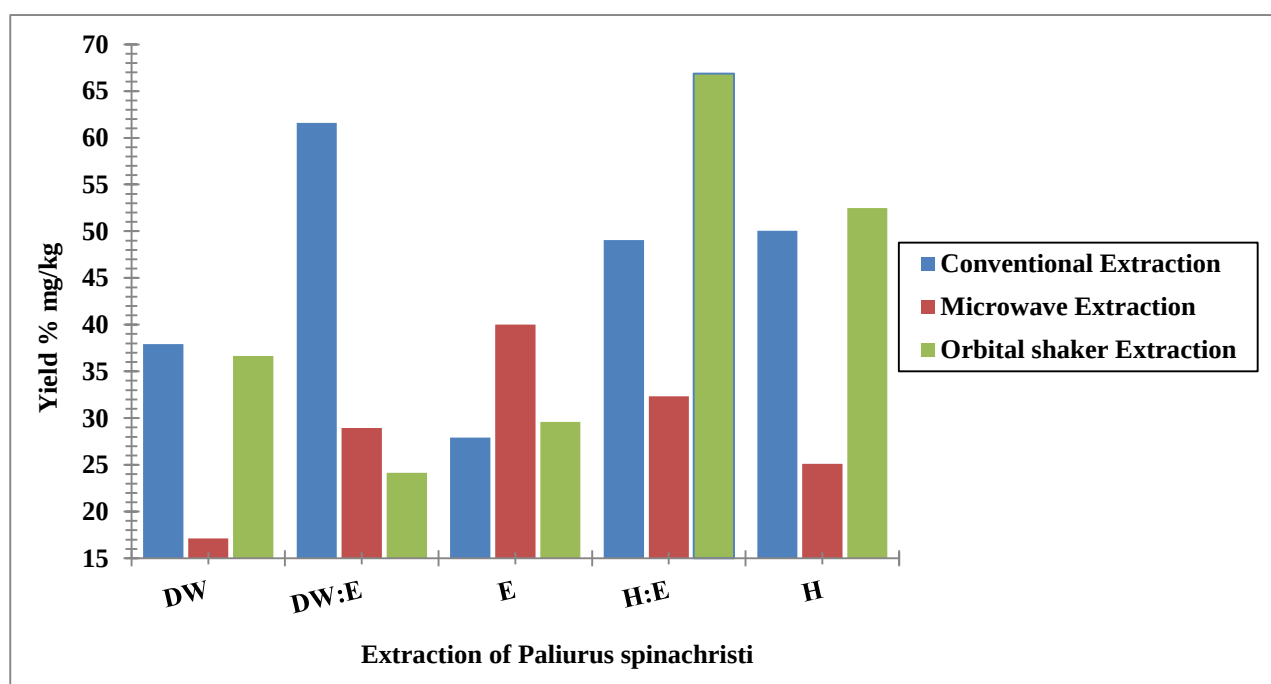


Figure 4.4. Extraction yield of *Paliurus spinachristi*

4.1.5. Determination extraction yield of *Purslane Portulaca oleracea* herb

The yield percentage of the *Purslane Portulaca oleracea* herb extracts prepared by conventional, microwave, and orbital shaker extraction techniques by using different solvents Distilled water, distilled water: Ethanol (v/v), Ethanol, and Hexane: Ethanol (v/v) and Hexane are potted in Table and Figure (4.5). The uppermost extraction yield was found in orbital shaker distilled water: ethanol (55.15 %), conventional distilled water (50.57 %), and microwave distilled water (38.01 %), with all values respectively. Whereas, the smallest extraction yield was observed in Hexane: Ethanol with microwave and with other methods of extraction solvents.

Table 4.5.Extraction yield % of plant *Purslane Portulaca oleracea*

	C/ E (%)	MW/E(%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	42.99 $\pm$ 0.40	38.01 $\pm$ 0.64	55.15 $\pm$ 0.64
DW:E (v/v)	50.57 $\pm$ 0.64	34.47 $\pm$ 0.62	33.31 $\pm$ 0.47
E	29.05 $\pm$ 0.47	30.73 $\pm$ 0.65	28.18 $\pm$ 0.64
H:E (v/v)	39.28 $\pm$ 0.25	12.61 $\pm$ 0.67	35.85 $\pm$ 0.64
H	39.55 $\pm$ 0.62	35.53 $\pm$ 0.28	23.54 $\pm$ 0.47

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E), and Hexane (H).

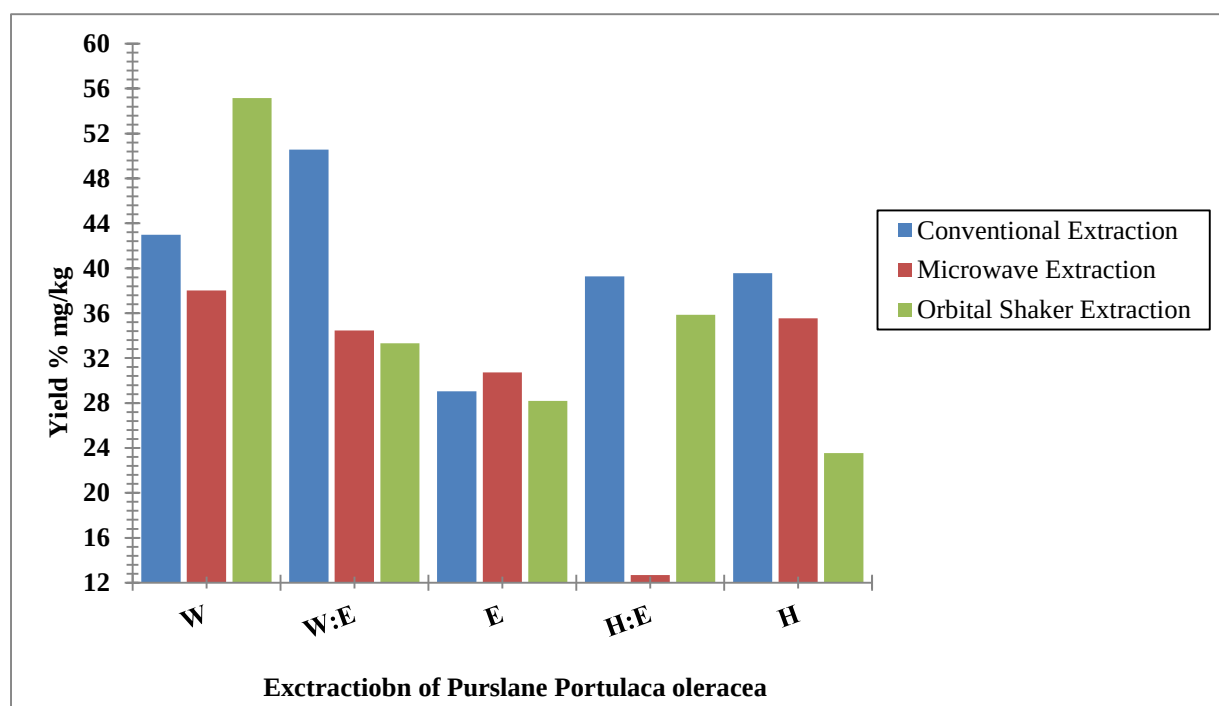


Figure 4.5.Extraction yield % of *Purslane Portulaca oleracea*

4.1.6. Determination extraction yield % of *Artemisia haussknechtii* Boiss

The yield determinations of plant *Artemisia haussknechtii* Boiss herb in different solvents were presented in Table and Figure (4.6). The result extraction yield in distilled water (OSE: 63.50 %), appeared to be highly significant comparing to distilled water: Ethanol solvents from (MWE: 57.25 %) and (CE: 42.35 %), respectively. On the other hand, the minimum extraction yield was observed in solvent of hexane with the (MWE: 13.53 %), and (CE: 21.53 %) respectively, that was an extensive range of yields among extracts was uncovered depending on the extraction methods and solvents.

Table 4.6.Extraction yield % of plant *Artemisia haussknechtii* Boiss

	C/ E (%)	MW/E(%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	31.88 $\pm$ 0.83	28.35 $\pm$ 0.88	63.50 $\pm$ 0.33
DW:E (v/v)	42.35 $\pm$ 0.83	57.25 $\pm$ 0.88	28.10 $\pm$ 0.33
E	30.17 $\pm$ 0.83	37.73 $\pm$ 0.88	43.46 $\pm$ 0.57
H:E (v/v)	23.58 $\pm$ 0.83	16.71 $\pm$ 0.88	34.77 $\pm$ 0.57
H	21.53 $\pm$ 0.83	13.02 $\pm$ 0.88	34.38 $\pm$ 0.57

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H).

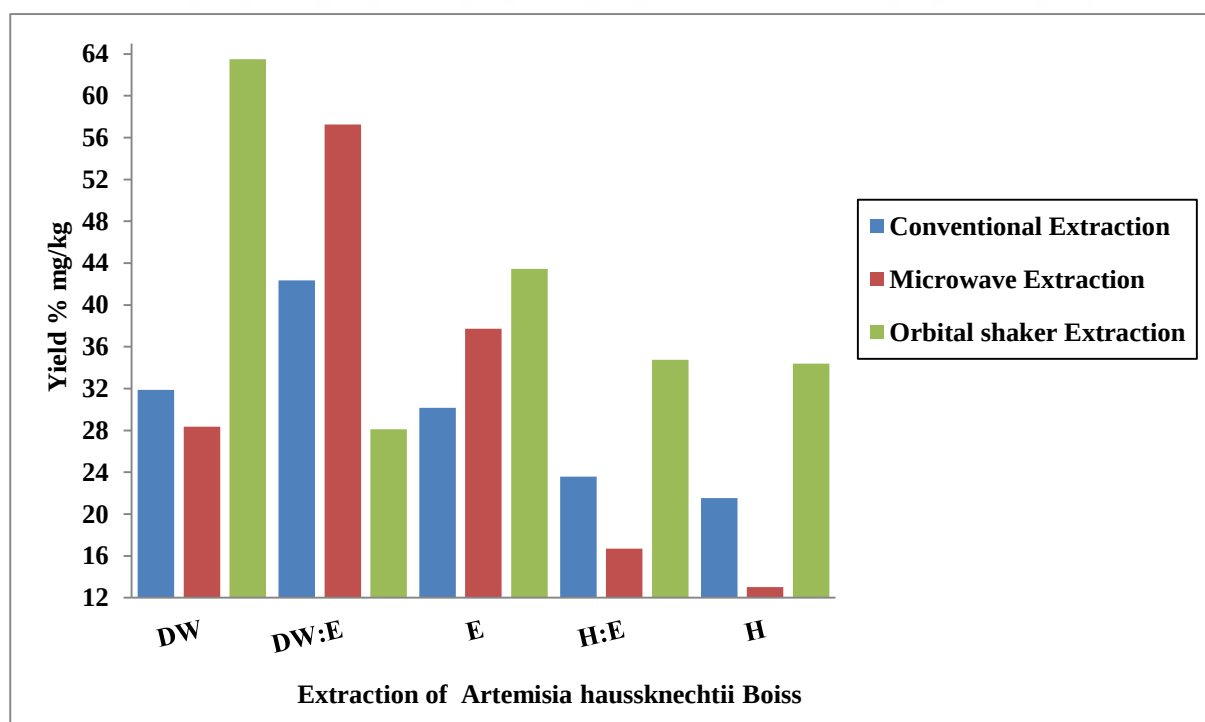


Figure 4.6.Extraction yield % of *Artemisia haussknechtii* Boiss

4.1.7. Determination extraction yield % of *Prunus microcarpa*

The extract yield of *Prunus microcarpa* seed prepared by different methods and dissimilar solvents. Extraction yield (mass of extract/mass of dry matter) was used as a pointer of the impacts of the extraction conditions. The orbital shaker hexane extract yield (OS/E: 43.46 %) was maximum yield. On the other hand, the upper most extracts yield were (MW: 38.10 %) and (CE: 38.37 %) from distilled water: ethanol solvent. While two conventional extracts of distilled water and ethanol solvents same amounts (26.38 %). Furthermore, the lowest extraction yield was observed for extracts by orbital shaker ethanol solvent as (12.98 %), are observed in Table and Figure (4.7).

Table 4.7.Extraction yield % of plant *Prunus microcarpa*

	C/ E (%)	MW/E(%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	26.38 $\pm$ 0.33	31.68 $\pm$ 0.32	34.40 $\pm$ 0.57
DW:E (v/v)	38.37 $\pm$ 0.33	38.10 $\pm$ 0.66	12.98 $\pm$ 0.66
E	26.38 $\pm$ 0.66	34.05 $\pm$ 0.66	40.17 $\pm$ 0.33
H:E (v/v)	35.63 $\pm$ 0.57	28.65 $\pm$ 0.32	34.06 $\pm$ 0.88
H	36.07 $\pm$ 0.33	29.29 $\pm$ 0.33	43.46 $\pm$ 0.66

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H).

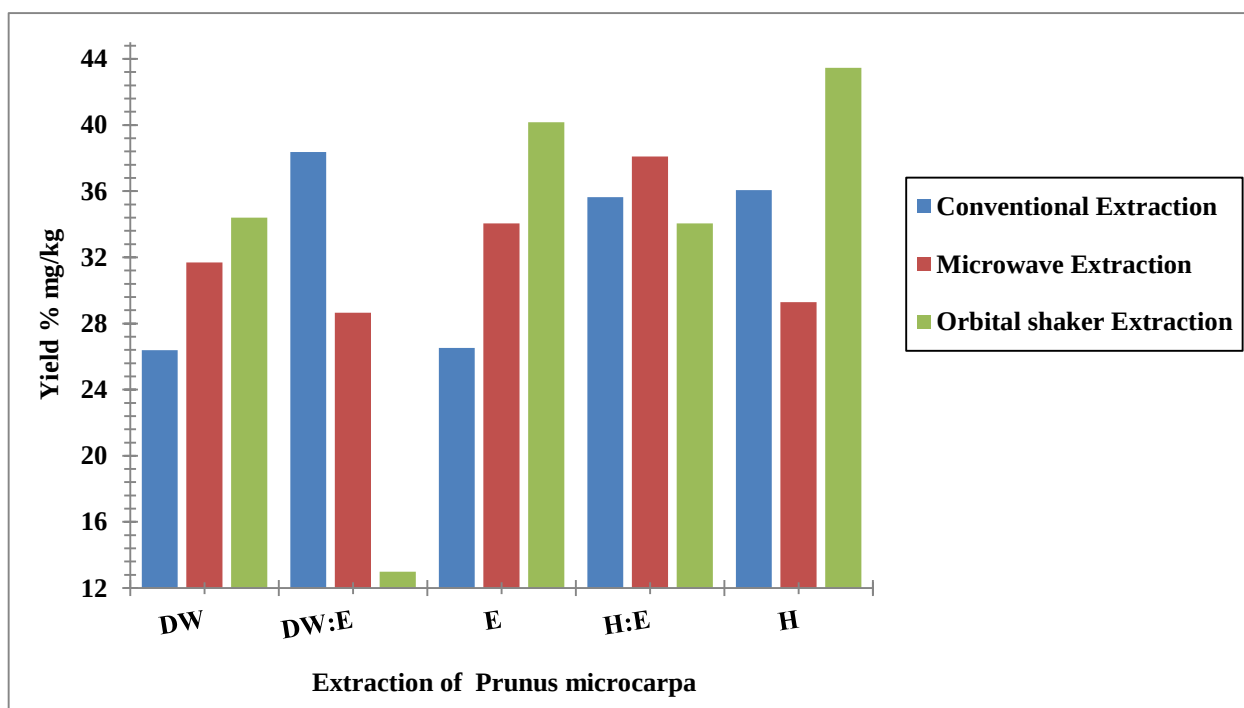


Figure 4.7. Extracts yield % of *Prunus microcarpa*

4.1.8. Determination extraction yield % of *Silybum Marianum*

The yield proportions of *Silybum Marianum* seed extracts were shown in Table and Figure (4.8). The topmost extraction yield in this plant seed extracts was found for the usage of (distilled water: ethanol) solution in microwave extraction method (MW: 55.46 %), with the orbital shaker (hexane: ethanol) yield percentage (OS:53.43 %), and (distilled water), with the conventional extraction (CE: 32.89 %), respectively. On the other hand, the lowest yield percentage was (CE:17.72 %) by conventional (water: ethanol) extraction, compare with other methods. However, all two solvents extract as (H: E and H) of the conventional method were as same percentages (31.54 %).

Table 4.8.Extraction yield of plant *Silybum Marianum*

	C/ E (%)	MW/E(%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	32.89 $\pm$ 0.33	28.68 $\pm$ 0.33	36.84 $\pm$ 0.33
DW:E (v/v)	17.72 $\pm$ 0.33	55.46 $\pm$ 0.33	28.20 $\pm$ 0.32
E	29.18 $\pm$ 0.34	46.72 $\pm$ 0.33	43.71 $\pm$ 0.32
H:E (v/v)	31.54 $\pm$ 0.66	34.34 $\pm$ 0.33	53.43 $\pm$ 0.32
H	31.54 $\pm$ 0.67	43.74 $\pm$ 0.33	47.82 $\pm$ 0.33

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H).

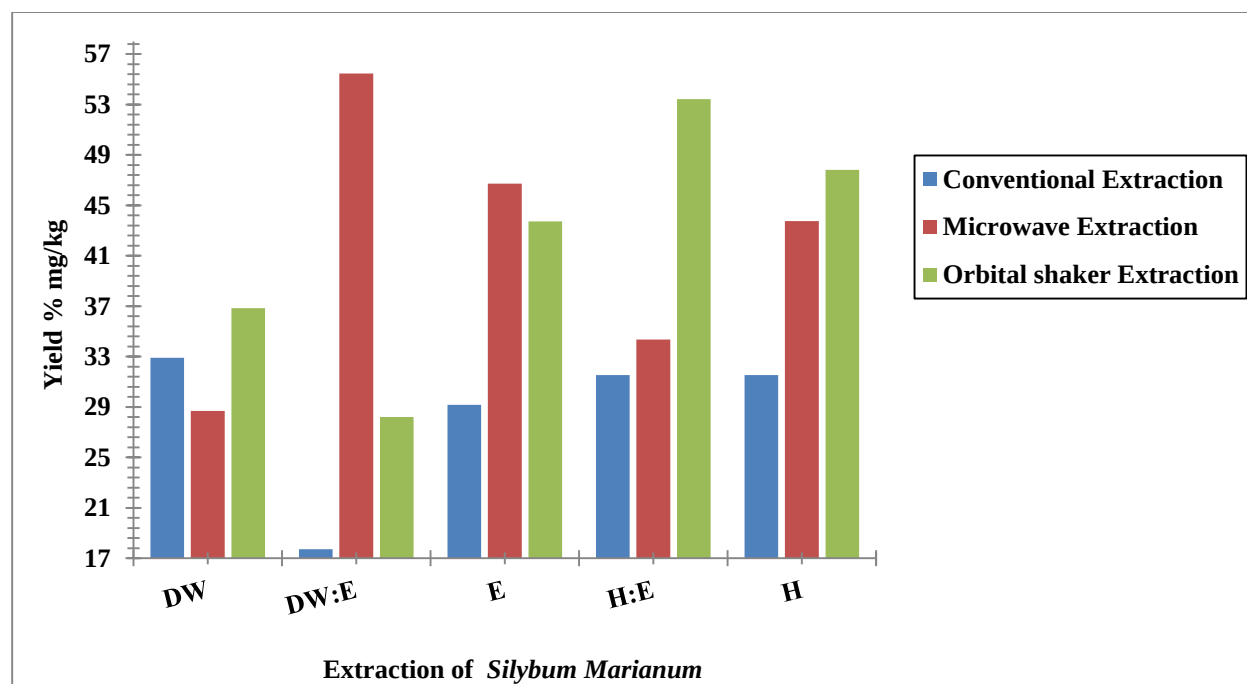


Figure 4.8.Extracts yield % of *Silybum Marianum*

4.1.9. Determination extraction yield % of *Pistacia eurycarpa* Yale

Extract yield of *Pistacia eurycarpa* seed prepared by dissimilar methods with utilized different solvents such as (distilled water), (ethanol: distilled water) (v/v), (ethanol), (hexane: ethanol) (v/v), and (hexane) are summarized in Table and Figure (4.9). The orbital shaker (OSW: 71.98 %), conventional (CEW: 64.77 %), and microwave (MWW: 60.94 %) as the same solvent as water extracts yield, were maximum yield followed by with other solvents respectively. However, although, the orbital shaker method was significant values with five solvents, compare with other methods. On the other hand, the lowest extract yield was (OSE: 25.26 %) with ethanol solvent. While all three solvents extract as (water: ethanol, ethanol, and hexane) of the conventional method was same percentages (34.86 %). Also, the minimum extract yield was (MWE: 29.60 %), and ethanol solvent.

Table 4.9.Extraction yield % of plant of *Pistacia eurycarpa* yale

	C/ E (%)	MW/E(%)	OS/E (%)
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	64.77 $\pm$ 0.66	60.94 $\pm$ 0.33	71.98 $\pm$ 0.33
DW:E (v/v)	34.86 $\pm$ 0.33	38.35 $\pm$ 0.66	48.81 $\pm$ 0.33
E	34.86 $\pm$ 0.88	29.60 $\pm$ 0.57	25.26 $\pm$ 0.88
H:E (v/v)	49.10 $\pm$ 0.33	48.14 $\pm$ 0.33	44.98 $\pm$ 0.66
H	34.86 $\pm$ 0.88	60.49 $\pm$ 0.33	43.21 $\pm$ 0.33

Note: Mean standard Error; Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H).

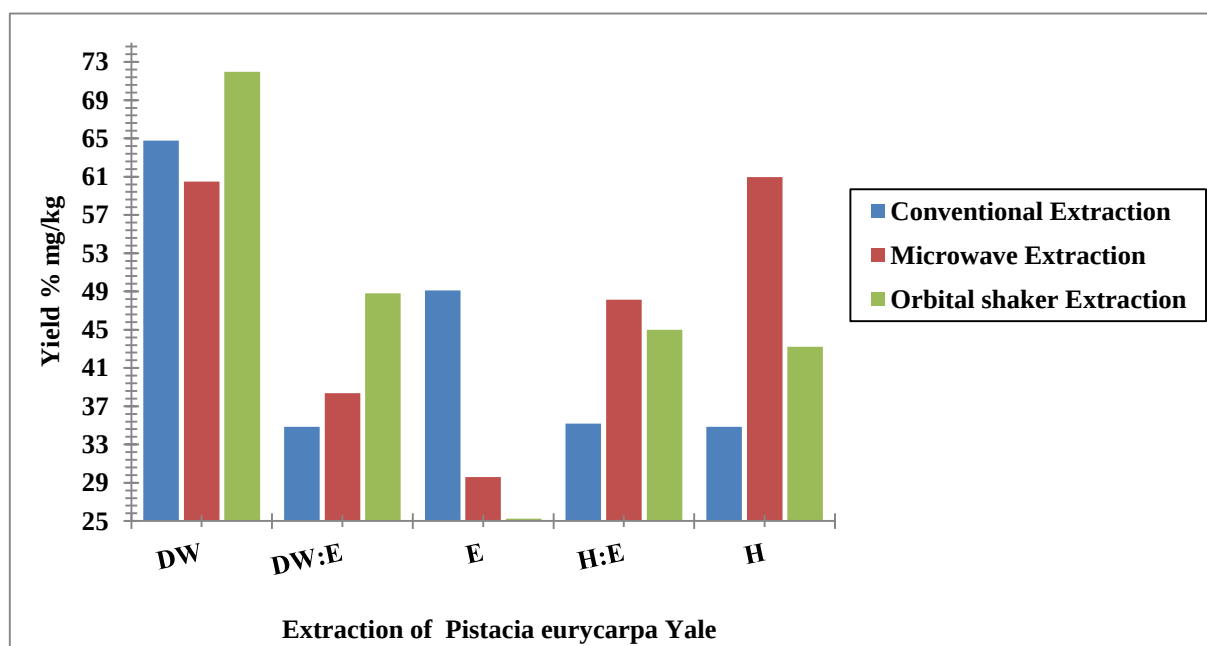


Figure 4.9.Extracts yield % of *Pistacia eurycarpa* yale

Yield extraction is the separation of medicinally active portions of the plant using selective solvents through standard procedures (Handa *et al.* 2008). In our research showed some solvents suitable for the extraction of particular classes of plants compounds. The aqueous solvents mixture (mixture of water, ethanol, and hexane in varying proportions) is generally considered to give high extraction yields according to different plants in all tables, which is owing to their expanded polarity range. Sample preparation is the crucial first step in the analysis of plant herbs because it is necessary to extract the desired chemical components from the herbal material for further extraction and characterization (Huie 2002). Furthermore, the polar solvents will extract out polar substances and non-polar material will be extracted out by non-polar solvents. Solvent extraction is the most popular method of extraction (Gupta *et al.* 2012).

Our results are in agreement with (Hazha J. Hidayat 2015) study in which the plant of *Eremurus spectabilis* extract by hexane ethanolic was stronger affected and similar with the parameter investigated was 86.62–91.35 mg/kg, by water extract. (Tosun, Ercisli, Ozer, Turan, *et al.* 2012).

Although, that the selective extraction of the plant of *Prosopis farcta* approval with (Poudineh *et al.* 2015; Layla A 2018) by appropriate solvents hexane and dichloromethane extracts could be very important to obtain bioactive fractions. Also from plants of *Capparis spinosa* extract by hexane and Chloroform and plant *Paliurus spinachristi* extracts ethanol and ethyl acetate was approval with (Brindha.P 2014; Şen 2018), because have significant yield extracts from the plant of *Silybum marianum* not approval with the studies (Bouhalit and Kechrid 2018; Sun *et al.* 2016).due the different technique. On the other hand, in all three plants as *Prunus microcarpa*, *Silybum Marianum* and *Pistacia eurycarpa yale* were a significant value with extract hexane solvent. Because non-polar solvents perform a good extraction, also in combination among them. According to the solubility of the bioactive compounds, there are a large number of inorganic, organic, polar and non-polar extract (De Monte *et al.* 2014). Also, the effects of solvents polarity on extraction yield both qualitatively and quantitatively was confirmed by (Franco *et al.* 2008),

The process of yield extraction are equally important with all solvents in the study of medicinal plants,, that agreement with (Balaky *et al.* 2020; Holem *et al.* 2021). Therefore, an extraction method is that which can obtain minimal changes to the functional properties and obtaining extracts with a high yield of the extract required (Quispe-Condori *et al.* 2008).

Generally, our results were observed that the yield extraction from all plants was found a great significant value that obtained through different techniques with five solvents.



4.2. Determination Total Condensed Tannin of all plant studies

Tannin was quantified by using the n-butanol-HCl-iron Procedure. Using this measurement to determine tannin, the rating in the figure was quantitatively released from the analysis was displayed in Figure (4.10). The highest widely used standard for n-butanol/HCl the mimosa tannin assay is determined using the regression equation ($y = 300.83x - 33.559$), under typical reactions/conditions. $R^2 = 0.9973$ was previously calculated based on a linear calibration curve comparison of the tannins present in all plant species.

Calculation: percent = $A / 3 m$, A = Value of extinction, m = Weight of mass (Makkar and Singh 1995; Elena 2011; Wang and Franz 2015; Karaogul *et al.* 2017).

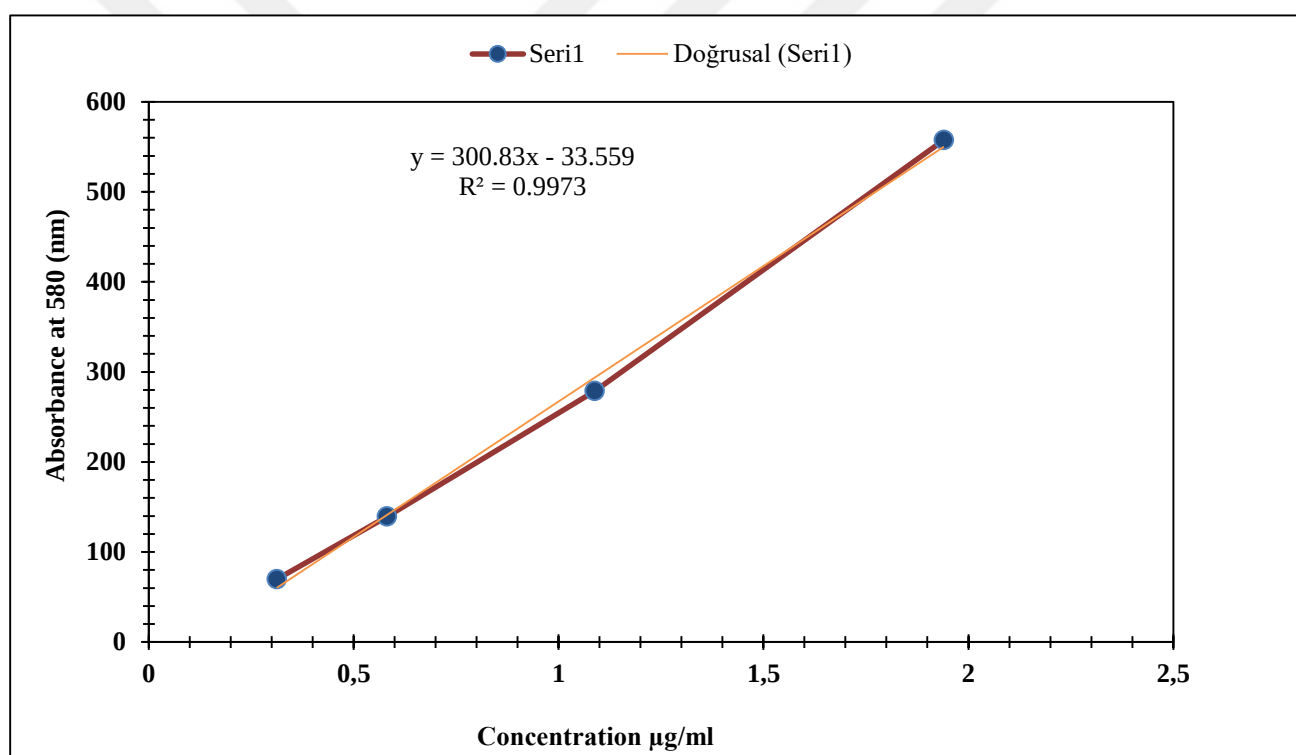


Figure 4.10. Mimosa tannin standard calibration curve

4.2.1. Determination Total Condensed Tannin of *Eremurus Spectabilis* Bieb

From the Table (4.10), and figure (4.11), the total condensed tannins were the highest value in Hexane: ethanol extract of the conventional method (C/E: 0.307 mg/kg), also the hexane extract of microwave technique to give (MW/E:0.187 mg/kg) and (OS/E:0.186 mg/kg) from the orbital shake of Hexane extract respectively. Contrarily, the method of conventional extraction was higher in five solvents from distilled water to hexane respectively, compared with orbital shaker extraction and microwave extraction. Then, the distilled water: ethanol from methods of conventional extract, microwave extract, and orbital shaker obtained lower values of total condensed tannin 0.042 mg/kg, 0.059 mg/kg, and 0.040 mg/kg from plant *Eremurus Spectabilis* Bieb, respectively. The methods and solvents were observed a high significant for this plant.

Table 4.10. Analysis Total condensed tannin concentration of *Eremurus Spectabilis* Bieb

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.068 $\pm$ 0.006	0.099 $\pm$ 0.002	0.041 $\pm$ 0.00
DW:E (v/v)	0.042 $\pm$ 0.000	0.059 $\pm$ 0.004	0.040 $\pm$ 0.00
E	0.107 $\pm$ 0.004	0.080 $\pm$ 0.000	0.159 $\pm$ 0.00
H:E (v/v)	0.307 $\pm$ 0.008	0.147 $\pm$ 0.006	0.173 $\pm$ 0.00
H	0.272 $\pm$ 0.004	0.187 $\pm$ 0.004	0.186 $\pm$ 0.00

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

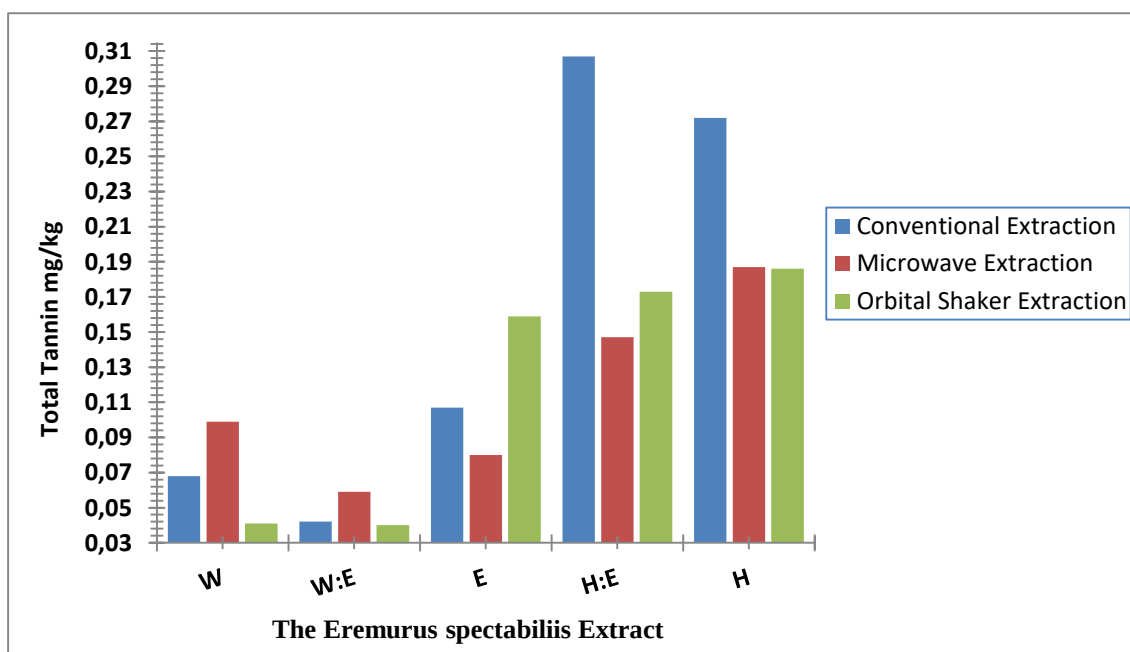


Figure 4.11. Total condensed tannin concentration of *Eremurus Spectabilis* Bieb

4.2.2. Determination Total Condensed Tannin of *Prosopis fracta*

The results indicated that the total condensed tannin prepared by different methods with five solvents are illustrating in Table (4.11), and Figure (4.12). Our results were observed the total condensed tannin was a higher amount from hexane extract with techniques to give the values of (C/E: 0.186 mg/kg), (OS/E: 0.158 mg/kg), and (MW/E: 0.103 mg/kg) respectively. Then the conventional extraction was given a high result with solvents, respectively. On the other hand, the distilled water extract was lowest amount followed by (C/E: 0.049 mg/kg), (OS/E: 0.045 mg/kg), and (MW/E: 0.053 mg/kg), respectively. Furthermore, most of our results of total condensed tannin in other solvents of *Prosopis fracta* extract were discovered a significant value.

Table 4.11. Analysis Total condensed tannin concentration of *Prosopis fracta*

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.049 $\pm$ 0.000	0.053 $\pm$ 0.002	0.045 $\pm$ 0.0004
DW:E (v/v)	0.117 $\pm$ 0.004	0.095 $\pm$ 0.002	0.050 $\pm$ 0.0006
E	0.120 $\pm$ 0.000	0.068 $\pm$ 0.000	0.067 $\pm$ 0.0002
H:E (v/v)	0.071 $\pm$ 0.002	0.099 $\pm$ 0.004	0.084 $\pm$ 0.0006
H	0.186 $\pm$ 0.004	0.103 $\pm$ 0.004	0.158 $\pm$ 0.0004

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.

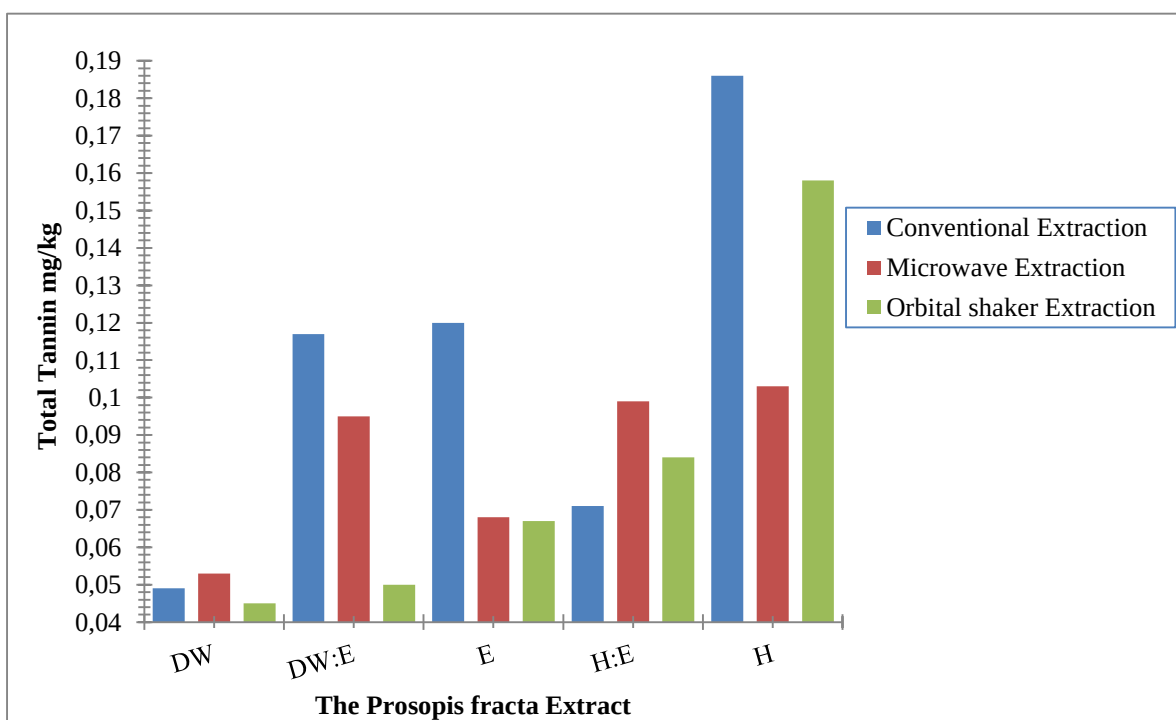


Figure 4.12. Total condensed tannin concentration

4.2.3. Determination Total Condensed Tannin of *Capparis spinosa*

According to Table (4.12), and Figure (4.13), the results showed that the amount of total condensed tannin was a minimum extract from (C/E: 0.462 mg/kg) ethanol: distilled water extract. Also observed high-value Ethanol solvents (OS/E: 0.116), and hexane: ethanol extract (MW/E: 0.113 mg/kg), respectively. On the other hand, the lowest value of total tannin was found from ethanol: distilled water (OS/E: 0.043 mg/kg), (MW/E: 0.057 mg/kg), and ethanol extract (C/E: 0.061 mg/kg), although, the conventional method was significant values with five solvents, compared with other methods microwave extraction and orbital shaker extraction, respectively.

Table 4.12. Analysis Total condensed tannin concentration of *Capparis spinosa*

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.237 $\pm$ 0.002	0.067 $\pm$ 0.004	0.051 $\pm$ 0.0004
DW:E (v/v)	0.462 $\pm$ 0.004	0.057 $\pm$ 0.000	0.043 $\pm$ 0.0002
E	0.061 $\pm$ 0.004	0.107 $\pm$ 0.006	0.116 $\pm$ 0.0002
H:E (v/v)	0.075 $\pm$ 0.000	0.113 $\pm$ 0.002	0.050 $\pm$ 0.004
H	0.165 $\pm$ 0.002	0.110 $\pm$ 0.000	0.068 $\pm$ 0.0000

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

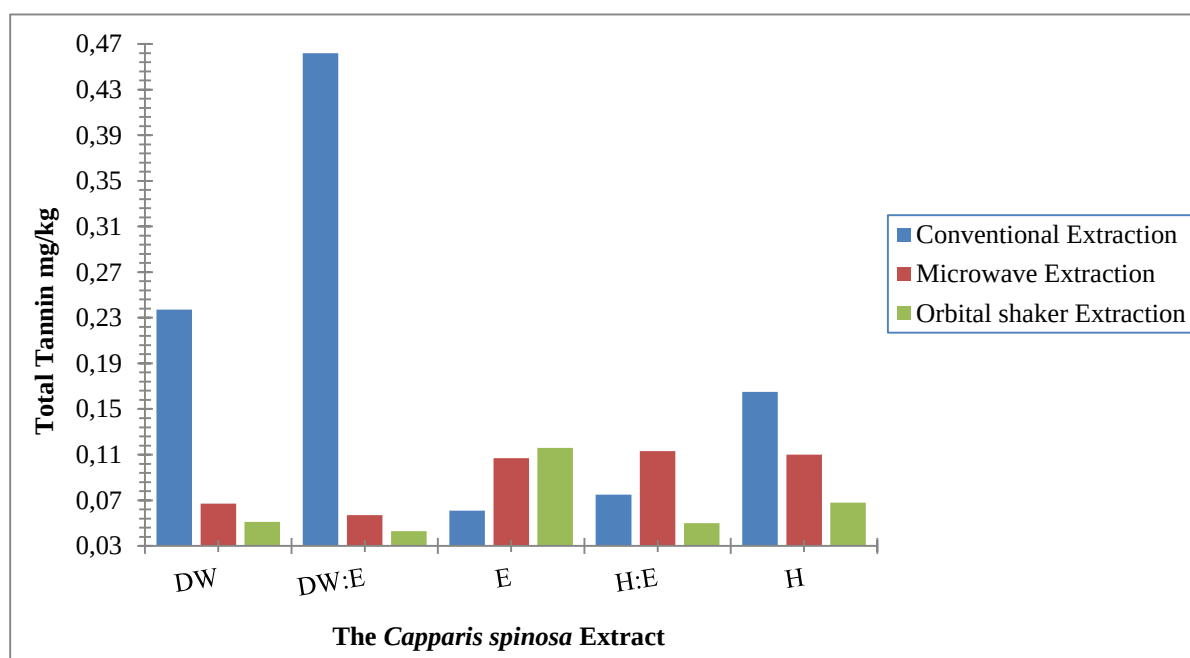


Figure 4.13. Total condensed tannin concentration of *Capparis spinosa*

4.2.4. Determination Total Condensed Tannin of *Paliurus spina christi*

Our results for dissimilar solvents of condensed tannin from *Paliurus spina christi* according to an experimental different methods extraction are shown in Table (4.13), and Figure (4.14). the topmost value was found with hexane extracts (0.145 mg/kg), from conventional extraction, also other techniques showed the highest condensed tannin from microwave, orbital shaker methods with ethanolic extraction to gives (0.140 mg/kg), and (0.132 mg/kg). Despite that, the method of conventional solvents from *Paliurus spina christi* was a significant value compare with both methods of microwave and orbital shake extraction respectively. Whereas, the lowest values were observed from ethanol: distilled water (OS/E: 0.053 mg/kg), with other methods of extraction, respectively. Furthermore, the values of distilled water from conventional and orbital shaker extractions were obtained the same results (0.065 mg/kg).

Table 4.13. Analysis Total condensed tannin concentration of *Paliurus spina christi*

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.065 $\pm$ 0.002	0.078 $\pm$ 0.004	0.065 $\pm$ 0.0002
DW:E (v/v)	0.078 $\pm$ 0.000	0.083 $\pm$ 0.000	0.053 $\pm$ 0.0004
E	0.102 $\pm$ 0.000	0.140 $\pm$ 0.004	0.132 $\pm$ 0.0006
H:E (v/v)	0.092 $\pm$ 0.000	0.112 $\pm$ 0.000	0.100 $\pm$ 0.0006
H	0.145 $\pm$ 0.004	0.098 $\pm$ 0.000	0.052 $\pm$ 0.0004

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

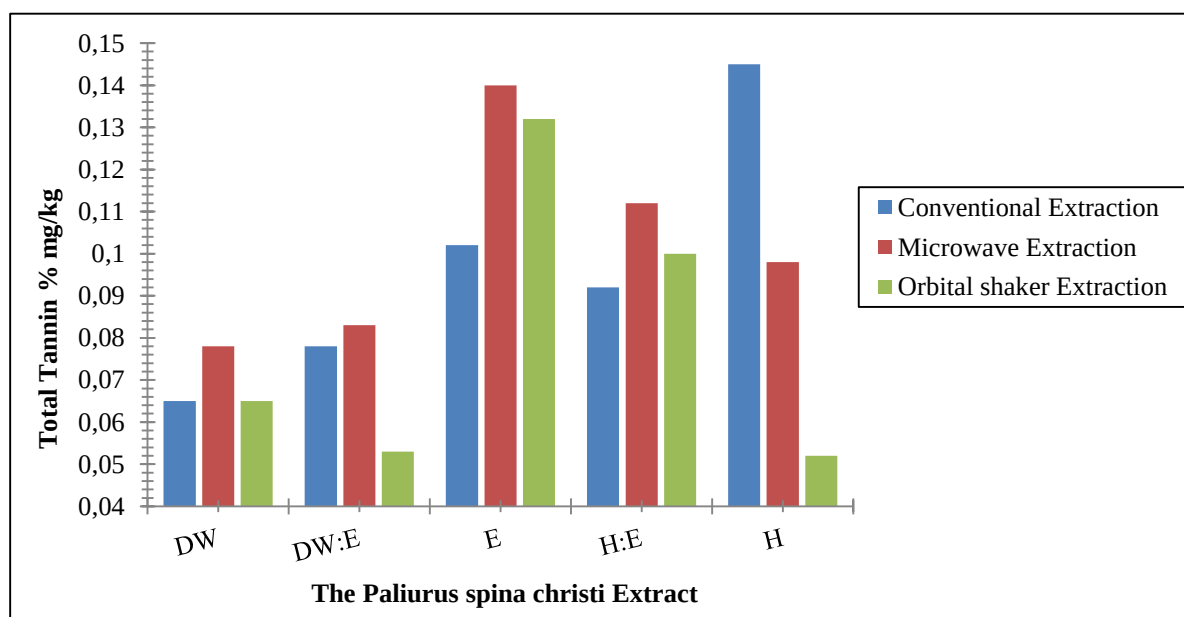


Figure 4.14.Total condensed tannin concentration of *Paliurus spina christi*

4.2.5. Determination Total Condensed Tannin of *portulaca oleracea* herb

Provides information regarding the contents of total condensed tannin in the *portulaca oleracea* herb, are given in Table (4.14), and Figure (4.15). The results showed that the total condensed tannin was found maximum amount from (C/E: 0.341 kg/mg), (MW/E: 0.328 kg/mg), with the solvent of (hexane: ethanol) and the amount of (OS/E: 0.193 kg/mg) with ethanol solvent. Whereas, the total condensed tannin recorded lowest amount from all methods (C/E: 0.046 kg/mg), (MW/E: 0.062 kg/mg), and (OS/E: 0.086 kg/mg) with solvent of distilled water. Generally, the total condensed tannin from *portulaca oleracea* was got significant results with all method techniques and different solvents, especially. Ethanol, hexane: ethanol, and hexane extraction.

Table 4.14. Analysis Total condensed tannin concentration of *portulaca oleracea*

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.046 $\pm$ 0.002	0.062 $\pm$ 0.000	0.086 $\pm$ 0.0000
DW:E (v/v)	0.065 $\pm$ 0.000	0.066 $\pm$ 0.002	0.092 $\pm$ 0.0004
E	0.190 $\pm$ 0.004	0.130 $\pm$ 0.004	0.193 $\pm$ 0.0006
H:E (v/v)	0.341 $\pm$ 0.004	0.328 $\pm$ 0.006	0.181 $\pm$ 0.0004
H	0.285 $\pm$ 0.000	0.117 $\pm$ 0.004	0.104 $\pm$ 0.0004

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

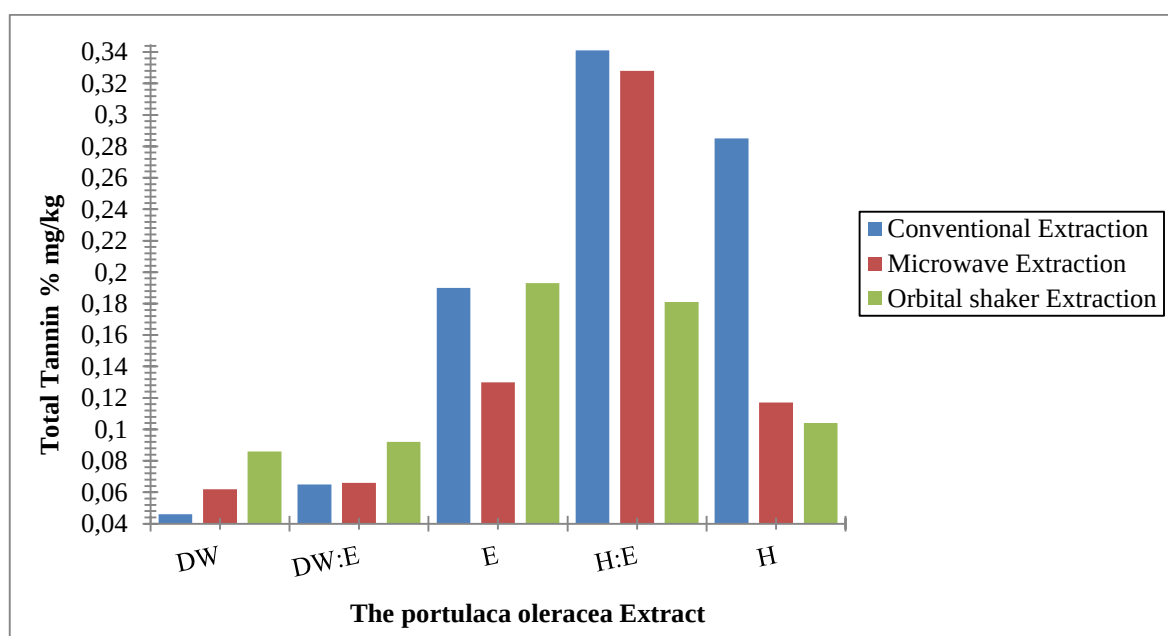


Figure 4.15.Total condensed tannin concentration of *portulaca oleracea*

4.2.6. Determination Total Condensed Tannin of *Artemisia haussknechtii* boiss

The results from Table (4.15), and Figure (4.16). Indicate that even though the *Artemisia haussknechtii* boiss herb was identified as a condensed tannin significant variety, by using the dissimilar organic solvents with the different methods. The values of extractable and total condensed tannin obtained in these experimental techniques were highest with all solvents, respectively. The condensed tannin from *Artemisia haussknechtii* boiss was observed a high value with Hexane solvent from techniques (C/E: 0.188 mg/kg), (OS/E: 0.195 mg/kg), and hexane: ethanol from microwave extraction to give (0.172 mg/kg). However, the solvent of (DW: E) with all methods was observed the lowest value (MW/E: 0.040 mg/kg), (OS/E: 0.060 mg/kg), and (C/E: 0.068 mg/kg), on the other hand, our results showed from all methods and solvents extraction that increases the values of ethanol, hexane: ethanol and hexane, respectively.

Table 4.15. Analysis total condensed tannin concentration of *Artemisia haussknechtii* boiss

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.072 $\pm$ 0.004	0.043 $\pm$ 0.004	0.076 $\pm$ 0.0002
DW:E (v/v)	0.068 $\pm$ 0.004	0.040 $\pm$ 0.004	0.060 $\pm$ 0.0004
E	0.090 $\pm$ 0.000	0.111 $\pm$ 0.004	0.134 $\pm$ 0.0004
H:E (v/v)	0.111 $\pm$ 0.002	0.172 $\pm$ 0.006	0.140 $\pm$ 0.0006
H	0.188 $\pm$ 0.004	0.090 $\pm$ 0.004	0.195 $\pm$ 0.0004

Notes/ Plants

=P;

C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction W=Distilled water;

DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane; Mean standard

Error=M $\pm$ S.E

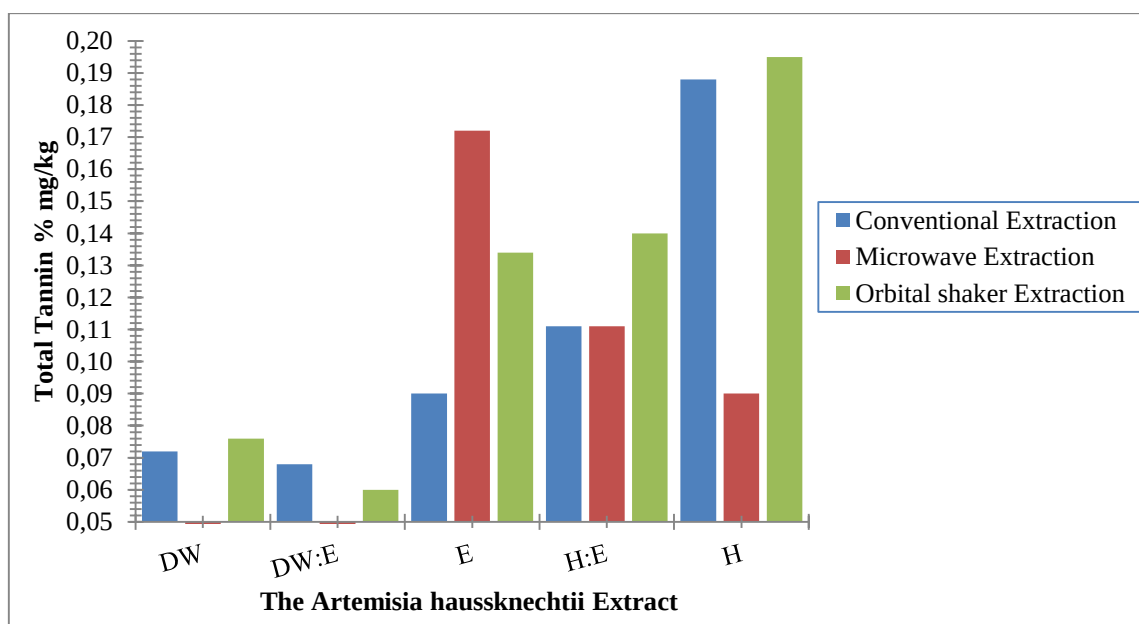


Figure 4.16. Total condensed tannin concentration of *Artemisia haussknechtii boiss*

4.2.7. Determination Total Condensed Tannin of *Prunus microcarpa*

Total condensed tannin contents of the *Prunus microcarpa* according technique methods and extraction solvents are shown in Table (4.16), and figure (4.17). Among the extraction solvents tested, the C/E: 0.160 mg/kg, MWE: 0.166 mg/kg, and OSE: 0.195 mg/kg were highest with hexane efficient solvent systems, when compared with all other solvent systems used, furthermore, the solvent of (distilled water: ethanol), that was showed the lowest values of total condensed tannin from all methods as arrange (CE: 0.057 mg/kg, MWE: 0.045 mg/kg, and OSE: 0.045 mg/kg), on the other hand, The total condensed tannin from microwave extraction was observed a similar amount with solvents as DW: E, E, and H:E: 0.045 mg/kg. Additionally, the total tannin of *Prunus microcarpa* from conventional extraction was a significant value with all solvents.

Table 4.16. Analysis Total condensed tannin concentration of *Prunus microcarpa*

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.068 $\pm$ 0.002	0.053 $\pm$ 0.002	0.049 $\pm$ 0.0002
DW:E (v/v)	0.057 $\pm$ 0.002	0.045 $\pm$ 0.005	0.045 $\pm$ 0.0000
E	0.090 $\pm$ 0.004	0.045 $\pm$ 0.000	0.055 $\pm$ 0.0006
H:E (v/v)	0.098 $\pm$ 0.004	0.045 $\pm$ 0.000	0.048 $\pm$ 0.0004
H	0.160 $\pm$ 0.004	0.166 $\pm$ 0.004	0.195 $\pm$ 0.0004

Notes/ Plants=P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

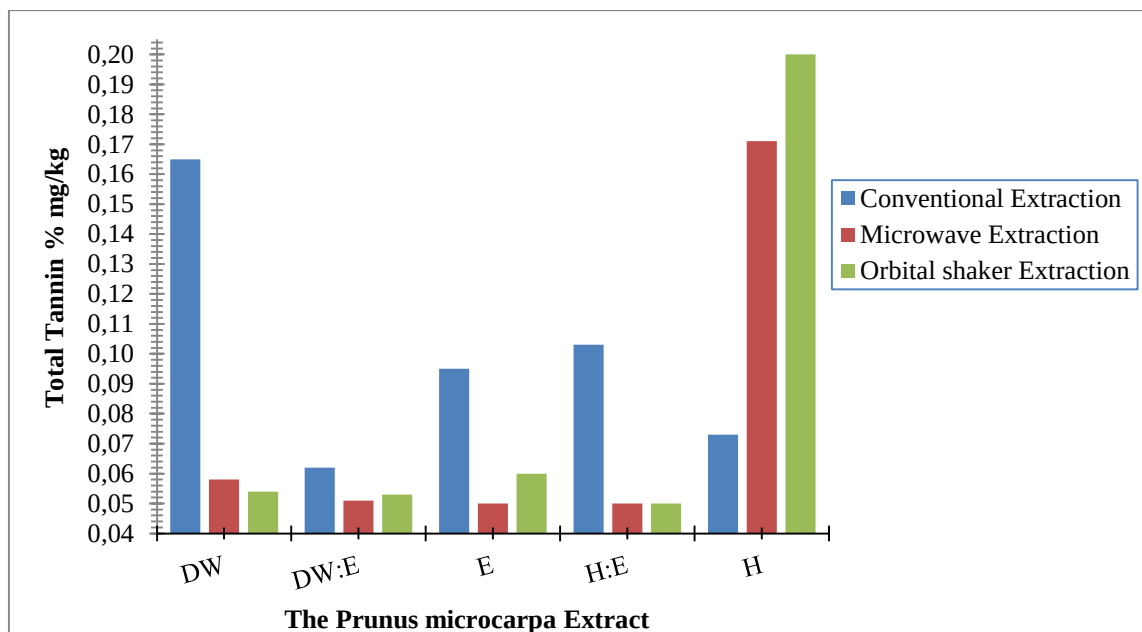


Figure 4.17. Total condensed tannin concentration of *Prunus microcarpa*

4.2.8. Determination Total Condensed Tannin of *Silybum Marianum*

Great variation was observed in the three methods of *Silybum Marianum* from all different solvents are summarized in Table (4.17) and Figure (4.18). The total condensed tannin ranged uppermost 0.178 mg/kg in an orbital shaker and 0.063 mg/kg in conventional methods with the distilled water solvent, also, in microwave extraction was observed 0.082 mg/kg with the hexane: ethanol solvent. Whereas, the lowest total condensed tannin was observed in hexane solvent with all methods of extraction as arranging (OS/E: 0.041 mg/kg, MW/E: 0.042 mg/kg, and C/E: 0.054 mg/kg), respectively. Generally, that the orbital shaker, microwave, and conventional extraction were significant values with all solvents respectively.

Table 4.17. Analysis Total condensed tannin concentration of *Silybum Marianum*

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.063 $\pm$ 0.000	0.052 $\pm$ 0.004	0.178 $\pm$ 0.0006
DW:E (v/v)	0.057 $\pm$ 0.000	0.057 $\pm$ 0.004	0.068 $\pm$ 0.0006
E	0.058 $\pm$ 0.004	0.051 $\pm$ 0.004	0.095 $\pm$ 0.0004
H:E (v/v)	0.053 $\pm$ 0.004	0.082 $\pm$ 0.004	0.049 $\pm$ 0.0004
H	0.053 $\pm$ 0.004	0.042 $\pm$ 0.004	0.041 $\pm$ 0.0004

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

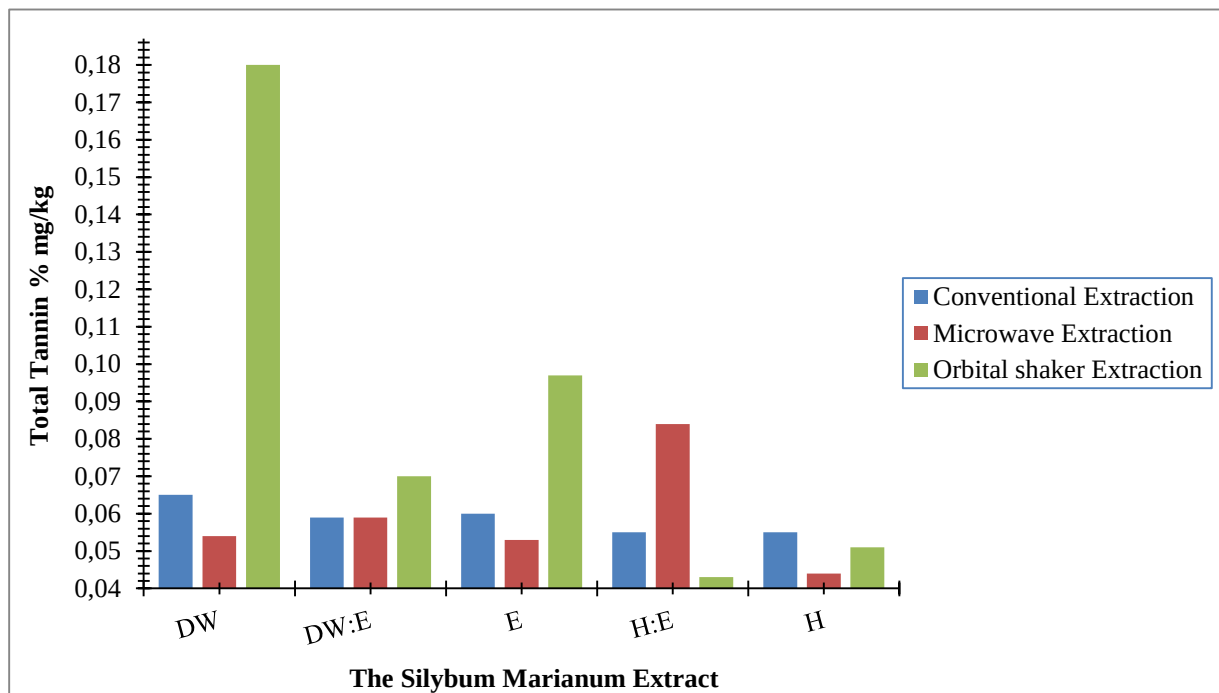


Figure 4.18. Total condensed tannin concentration of *Silybum Marianum*

4.2.9. Determination Total Condensed Tannin of *Pistacia eurycarpa Yale*

Total condensed tannin of *Pistacia eurycarpa Yale*, in different methods, is influenced by the dissimilar solvents was illustrated in Table (4.18) and Figure (4.19). Total tannin content was maximum in the orbital shaker extract prepared with Distilled water-ethanol (0.144 mg/kg), followed by hexane: ethanol (0.124 mg/kg) of conventional extraction and water (0.097mg/kg) from microwave extraction. Whereas the total condensed tannin was observed the lowest amount from microwave extraction (H/E: 0.055 mg/kg) followed by orbital shaker extraction (DW: 0.063 mg/kg), and conventional extraction (H: 0.066 mg/kg). However, that the conventional method was got a significant value with all solvents compare with other methods of extraction and solvents. In general, the concentration values of total condensed tannin of *Pistacia eurycarpa Yale* were higher and lower according to the prepared extraction methods and solvents extract.

Table 4.18. Analysis Total condensed tannin concentration of *Pistacia eurycarpa* Yale

	C/ E mg/kg	MW/E mg/kg	OS/E mg/kg
Solvent	Mean $\pm$ S.E	Mean $\pm$ S.E	Mean $\pm$ S.E
DW	0.106 $\pm$ 0.0006	0.097 $\pm$ 0.0006	0.063 $\pm$ 0.0002
DW:E (v/v)	0.099 $\pm$ 0.0004	0.073 $\pm$ 0.0000	0.144 $\pm$ 0.0004
E	0.079 $\pm$ 0.0002	0.063 $\pm$ 0.0004	0.076 $\pm$ 0.0004
H:E (v/v)	0.124 $\pm$ 0.0004	0.055 $\pm$ 0.0006	0.074 $\pm$ 0.0002
H	0.066 $\pm$ 0.0004	0.068 $\pm$ 0.0002	0.115 $\pm$ 0.0006

Notes/ Plants =P; C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction
W=Distilled water; DW: E=Ethanol: Distilled water; E=Ethanol; H: E=Hexane: Ethanol; H =Hexane, Mean standard Error=M $\pm$ S.E

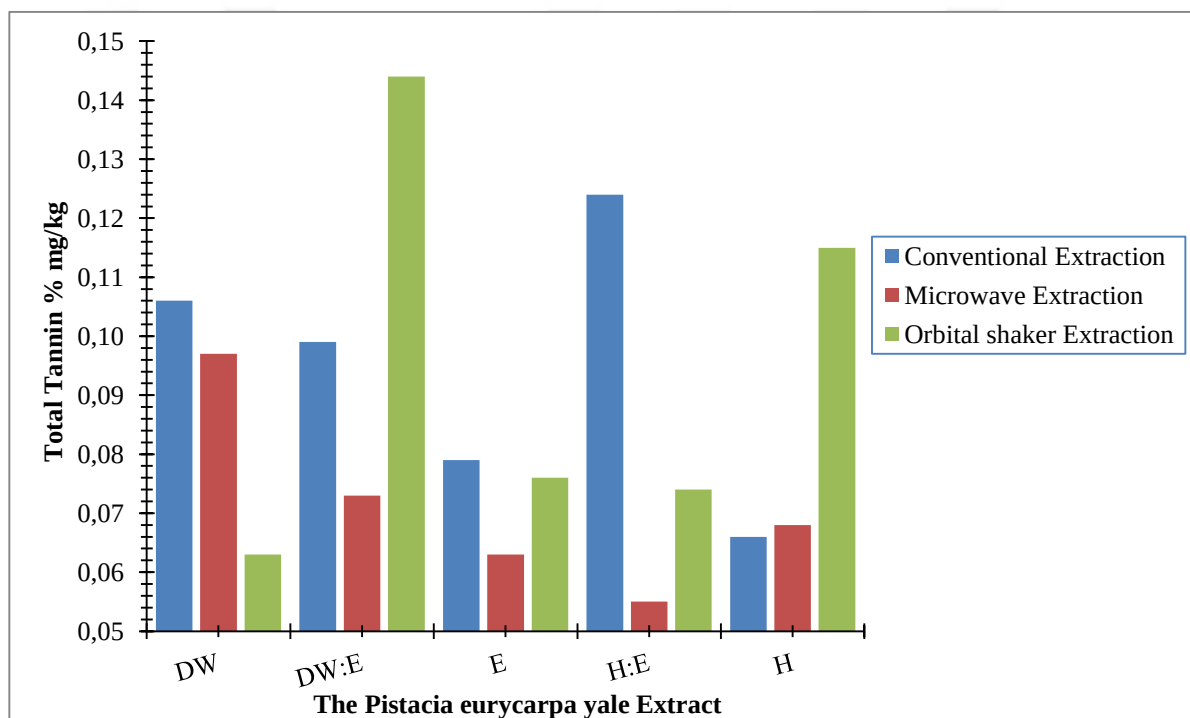


Figure 4.19. Total condensed tannin concentration of *Pistacia eurycarpa* Yale

The condensed tannin is secondary metabolism that occurs naturally in a variety of plants, found in cell walls or stored in vacuoles, stems, bark, leaves, flowers, or seeds, mainly in dicotyledonous plants. From the chemical point of view, they are complex organic compounds derived from flavonol, soluble in water (McMahon *et al.* 2000).

In our study, that the plant of *Eremurus Spectabilis* Bieb, that the methods of conventional extraction were higher in five solvents from distilled water to hexane respectively. There were no findings on complete condensed tannin of *Eremurus Spectabilis* Bieb by this process methods, as far as our literature investigation could discover. Therefore, this research may have provided new findings on this plant. Furthermore, the *Prosopis farcta* is in disagreement with other reports (Sharifi-rad *et al.* 2014), due to the high concentrations of tannins, alkaloids, glycosides, and moderate concentrations of flavonoids, saponins, and phenols in the *Prosopis farcta* of polar solvent methanolic extract, but agreement with (Fariba Jafari *et al.* 2013).

Also, the result *Capparis spinosa* is therefore consistent with of (Ghdeib 1999), Although, that the condensed tannin with solvent extraction of the plant of *Capparis spinosa* approval with (Mahboubi and Mahboubi. 2014), and (Tlili *et al.* 2017), due to the *Capparis spinosa* is a source of phytochemicals with important biological advantages. also the plant of *Paliurus Spina Christi* in our research not significant (Ali ŞEN 2018). On the other hand, the *portulaca oleracea* phytochemical screening of solvents extract was showed the highest values and agreement with (Agyare *et al.* 2015; Tadić *et al.* 2012; Holem *et al.* 2021). Also, another report mentioned, both aqueous and ethanol extracts of *portulaca oleracea* contain a strong amount of alkaloid, flavonoid, phenol, and sterol, but ethanol extract is preferred for the extraction of saponin, tannin, and terpenoid, this study inline with our study (Anthonia Ezeabara 2014a). But not compatible with (Karoune *et al.* 2017; Okafor and Ezejindu 2014), Due to the lowest amount of condensed tannin. Also, approval with ((Ezeabara *et al.* 2014; Balaky *et al.* 2020), because the ethanol aqueous and ethanol extracts of this plant are preferable for extraction of tannin, saponin, and terpenoid whereas would produce a good level of alkaloid, flavonoid, phenol, and sterol.

The *Artemisia haussknechtii* have the highest values were found from all methods and solvents. That for reason, don't accept with (Alavi and Karimi 2017b), but accepted with (Singh *et al.* 2012), because the ethanolic extract has significantly high ($P<0.05$) concentration phenolic, flavonoids and tannin comparison to other solvents used. The plant of *Prunus microcarpa* is disagree with a report (Belhadj *et al.* 2016). The *Silybum Marianum* agree with

(Shah *et al.* 2011). Furthermore, the *Pistacia eurycarpa* Yale was got a significantly high value from the conventional method with all methods extraction and solvents.

As a general rule, there has been a subtle distinction (solvent-wise) between the procedures of methods used for the extraction of condensed tannins when extracts are intended for quantification or when isolating condensed tannins for other, more specific measurements.



4.3. Determination Total Phenol of all plant studies

Phenolic or polyphenols are secondary plant metabolites that are all over the place present in plants and plant products. Many of the phenolics have been shown to contain high levels of antioxidant activities (Razali *et al.* 2008). The Plant phenolic compounds are presently among the most studied phytochemicals because of their biological functions, including antioxidant and anti-inflammatory characteristics (Koleckar *et al.* 2008).

4.3.1. Determination Total Phenol of *Eremurus Spectabilis Bieb*

The results showed the total phenol content of distilled water extract of *Eremurus Spectabilis Bieb* was observed significant high values (OS/E: 3.197 mg GAE/g), the (MW/E: 2.577 mg GAE/g), and (OS/E: 1.386 mg GAE/g) at ($p < 0.01$), respectively. Whereas the total phenol was lower than and near values from ethanol: distilled water extract was showed a significant at ($p < 0.01$) to get (OS/E: 2.918 mg GAE/g), the (MW/E: 2.652 mg GAE/g), and (OS/E: 2.226 mg GAE/g), dry extract respectively. are shown in Table (4.19), and Figure (4.20). Furthermore, the Orbital shake technique with both polar solvents water and water: ethanol extract of *Eremurus Spectabilis Bieb* was showed maximum significant amount at ($p < 0.01$), that the distilled water extract of *Eremurus Spectabilis Bieb* was a major effect of the anti-proliferate agent as compared to non-polar solvents, thus, the polar extracts were found to be strong inhibitors of at different concentrations, in comparison with ethanol extract, this finding are in agreement with this reports by (Abubaker and Hidayat 2015; Tosun, Ercisli, Ozer, Polat, *et al.* 2012), also disagreement with (Tuzcu and Agca 2017), and approval with (Murathan *et al.* 2018), due to the presence of high total phenol content and the consumption of *Eremurus Spectabilis Bieb* plant might prove to be beneficial for human health.

Table 4.19.Total phenolic contents of *Eremurus Spectabilis Bieb* areal parts

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	1.386 $\pm$ 0.005315	2.236 $\pm$ 0.000629	-110.207-	0.000
MW/E (mg GAE/g)	2.577 $\pm$ 0.005583	2.652 $\pm$ 0.000645	-82.928-	0.000
OS/E (mg GAE/g)	3.197 $\pm$ 0.000479	2.918 $\pm$ 0.000645	347.483	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$)

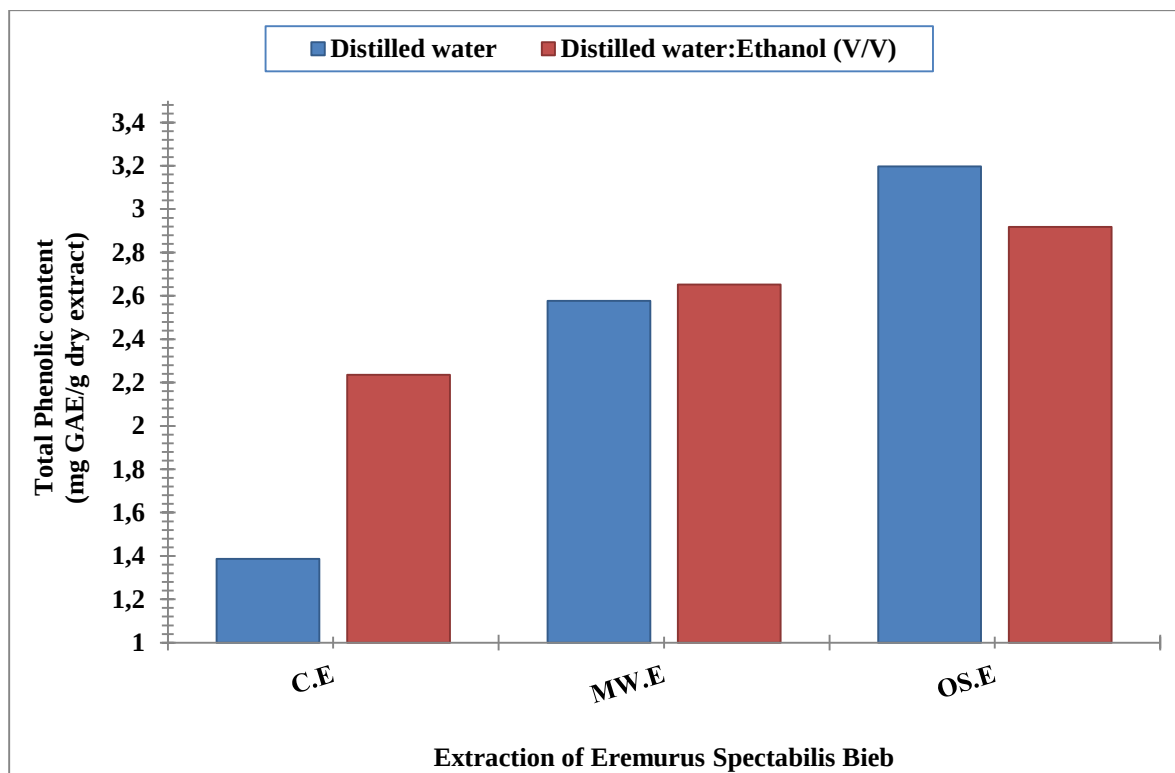


Figure 4.20. Total phenolic contents of *Eremurus Spectabilis Bieb* areal parts

4.3.2. Determination Total Phenol of *Prosopis fracta*

Table (4.20), and Figure (4.21). are investigated that the amount of the total phenol of *Prosopis fracta* which was the highest value in (MW/E: 3.276 mg GAE/g) with distilled water that significant levels of ($P < 0.01$) were used for determining statistical difference. The second highest value due to ethanol: distilled water extract of *Prosopis fracta* was (C/E: 3.011 mg GAE/g) significantly ($P < 0.01$). Whereas, the comparison with orbital shaker and conventional extraction were showed the lowest amount of total phenol content from *Prosopis fracta*. On the other hand, the solvent of ethanol: distilled water extract was observed maximum values of total phenol content with all three techniques significantly ($P < 0.01$), respectively. Our study agreement with those reports (Sharifi-rad *et al.* 2014; Mohammed *et al.* 2018). Thus, the results of this work referred that the selective extraction of *Prosopis farcta* by convenient solvents could be very important to obtain bioactive fractions. our study agrees with (Poudineh *et al.* 2015).

Table 4.20.Total phenolic contents of *Prosopis frakta*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	1.637 $\pm$ 0.000629	3.011 $\pm$ 0.000645	-1.373750-	0.000
MW/E (mg GAE/g)	3.276 $\pm$ 0.000645	2.933 $\pm$ 0.000854	0.343250	0.000
OS/E (mg GAE/g)	0.721 $\pm$ 0.000854	2.183 $\pm$ 0.000854	-1.461500-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at (p<0.01).

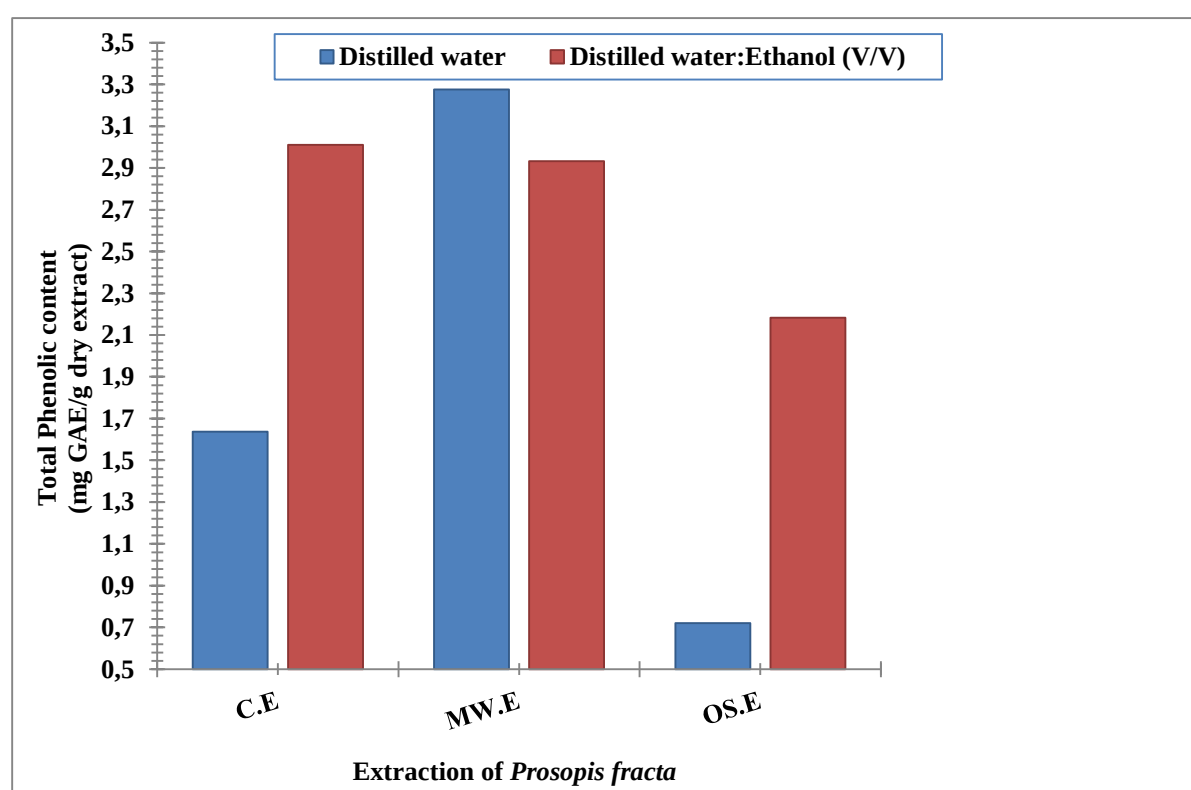


Figure 4.21. Total phenolic contents of *Prosopis frakta*

4.3.3. Determination Total Phenol of *Capparis spinosa*

The ethanol: distilled water extract, with high total phenol content by (OS/E:2653 mg GAE/g), with other (MW/E:2.260 mg GAE/g) and (C/E:1.233 mg GAE/g) extraction respectively, while the total phenol content was lowest in *capparis spinosa* distilled water extract followed by (C/E: 1.040 mg GAE/g), MW/E:1.251 mg GAE/g), and (OS/E: 1.796 mg GAE/g) with significance levels of (P< 0.01), that results illustrated in Table (4.21), and Figure (4.22). Therefore, the total phenol content in *Capparis spinosa* extract was observed at a

significantly high value ($P < 0.01$) with polar solvents with the orbital shaker technique. Our study agrees with (Mahboubi and Mahboubi, 2014). Our experiment showed that phenol content was located in *capparis spinosa*, approval with those reports (Allaith 2016; Tlili *et al.* 2017), and (Tlili, Khaldi, Triki, *et al.* 2010), also dissimilar with (Rezzan *et al.* 2013), thus in disagrees with (Oudah *et al.* 2014).

Table 4.21.Total phenolic contents of *Capparis spinosa*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	1.040 $\pm$ 0.000645	1.233 $\pm$ 0.000645	-211.421	0.000
MW/E (mg GAE/g)	1.251 $\pm$ 0.000645	2.260 $\pm$ 0.000645	-1105.304-	0.000
OS/E (mg GAE/g)	1.796 $\pm$ 0.000645	2.653 $\pm$ 0.000479	-1066.090-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

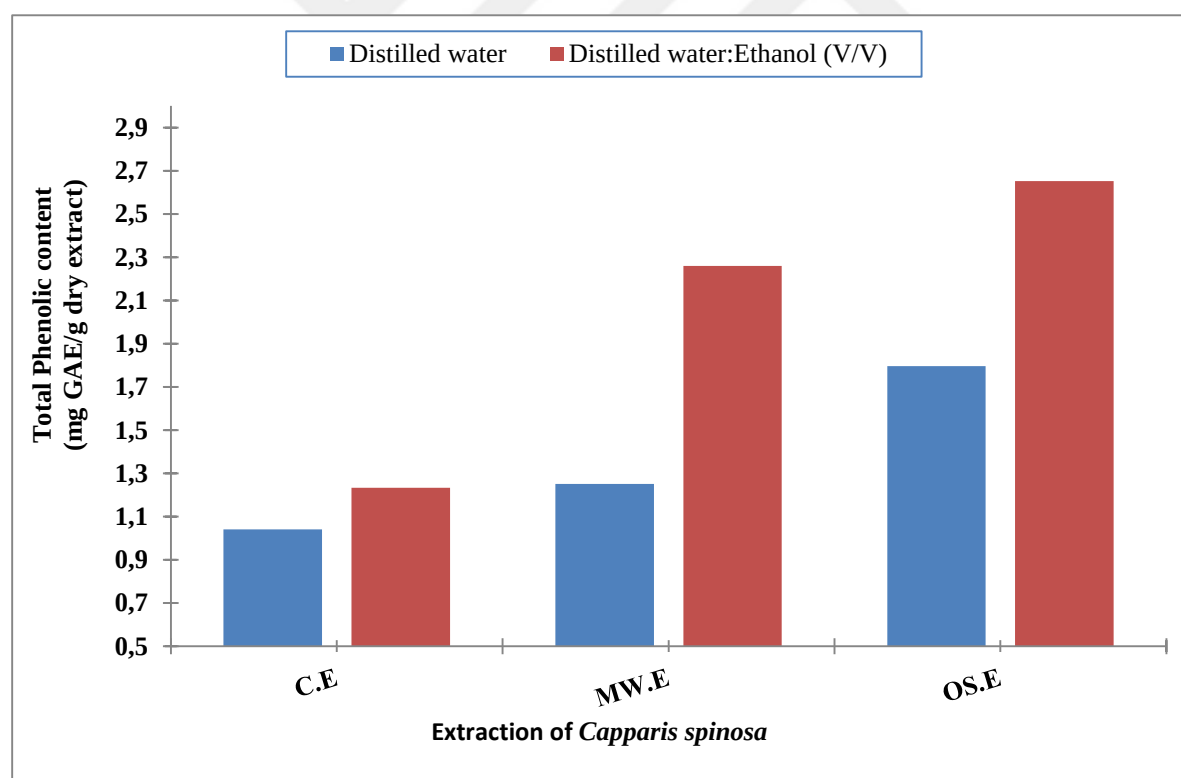


Figure 4.22.Total phenolic contents of *Capparis spinosa*

4.3.4. Determination Total Phenol of *Paliurus spina Christi*

From Table (4.22), and figure (4.23). Shows the total phenol content of ethanol: distilled water extract prepared from *Paliurus spina Christi* was variable but high value was observed in the three techniques (OS/E: 3.232), (MW/E:2.977), and (C/E:2.422)mg GAE/g respectively, with statistically significant of ($P < 0.01$), thus, the total phenol of water extract was observed equally lowest amount from *Paliurus spina Christi* with all methods extraction, our study agrees with reports (Ali ŞEN 2018), and (Taha Ahmed *et al.* 2013). The interesting biological activity of an extract of the *Paliurus spina Christi* is compelled us to study phytochemical analysis (Whiting and Alvin 1987). In addition, the presence of phenolic content of *Paliurus spina Christi* with polar solvents was good as a natural antioxidant activity.

Table 4.22.Total phenolic contents of *Paliurus spina Christi*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	2.861 $\pm$ 0.000289	2.422 $\pm$ 0.000707	575.441	0.000
MW/E (mg GAE/g)	2.353 $\pm$ 0.000629	2.977 $\pm$ 0.000408	-831.667-	0.000
OS/E (mg GAE/g)	2.269 $\pm$ 0.000408	3.232 $\pm$ 0.000479	-1531.027-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

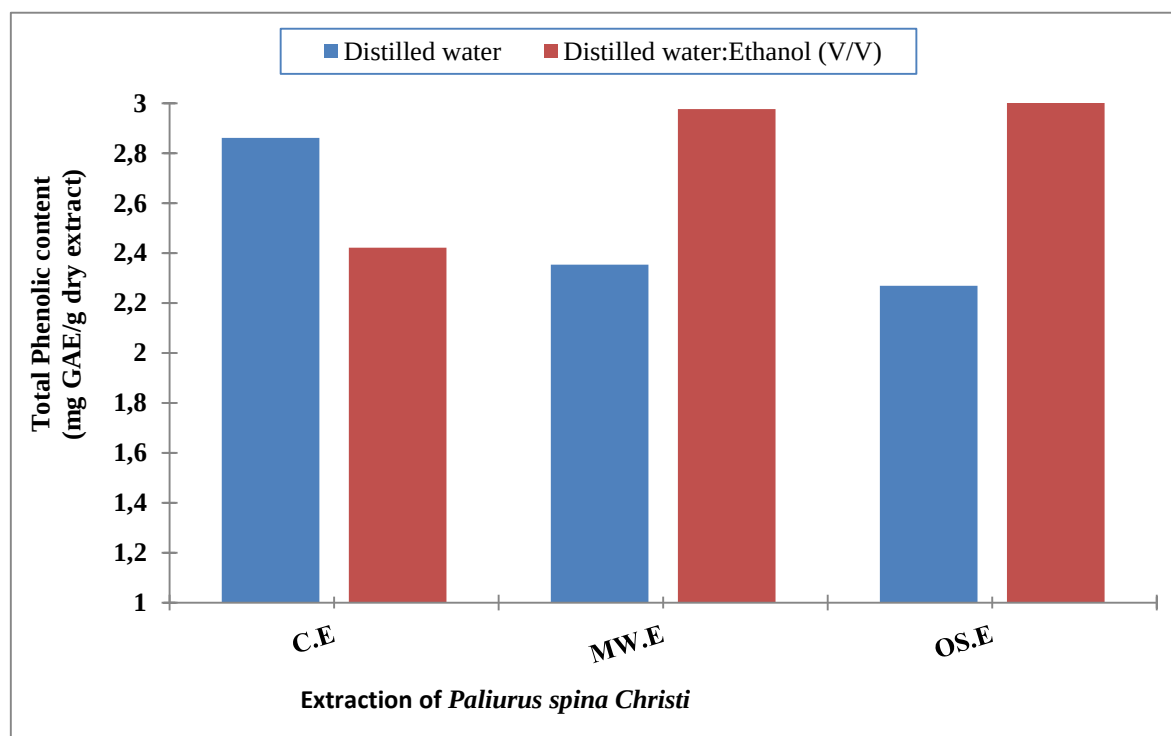


Figure 4.23.Total phenolic contents of *Paliurus spina Christi*

4.3.5. Determination Total Phenol of *portulaca oleracea* herb

Among all the tested of techniques extracts with dissimilar solvents the highest amounts of total phenol content were found in the ethanol: distilled water (MW/E: 2.543 mg GAE/g). The total phenol contents of other extracts distilled water were minimum values between MW/E: 1.079, OS/E: 0.981, and C/E: 0.953, mg GAE per g extract Table (4.23), and Figure (4.24). The extract of *portulaca oleracea* herb. by microwave method was great significant values of total phenol content at statistically significant of ($P < 0.01$). our study agrees with reports (Lim and Quah 2006; Hutadilok-Towatana *et al.* 2006). That approval with (Hefnawy and Ali 2015),(Karoune *et al.* 2017), and (Balaky *et al.* 2020) that the polar solvent ethanol extract of *portulaca oleracea* is rich in phenolic content and plays a beneficial role in the treatment of alcohol-induced tissue damage as therapeutic value. Furthermore, our study similar to (Anthonia Ezeabara 2014), because the aqueous and ethanol extracts from *Portulaca oleracea* L. would produce a good level of total phenol, alkaloid, flavonoid, and sterol.

Table 4.23.Total phenolic contents of *portulaca oleracea* L.

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	0.953 $\pm$ 0.000408	1.874 $\pm$ 0.000479	-1464.271-	0.000
MW/E (mg GAE/g)	1.079 $\pm$ 0.000408	2.543 $\pm$ 0.000750	-1715.340-	0.000
OS/E (mg GAE/g)	0.981 $\pm$ 0.000289	1.741 $\pm$ 0.000479	-1359.977-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

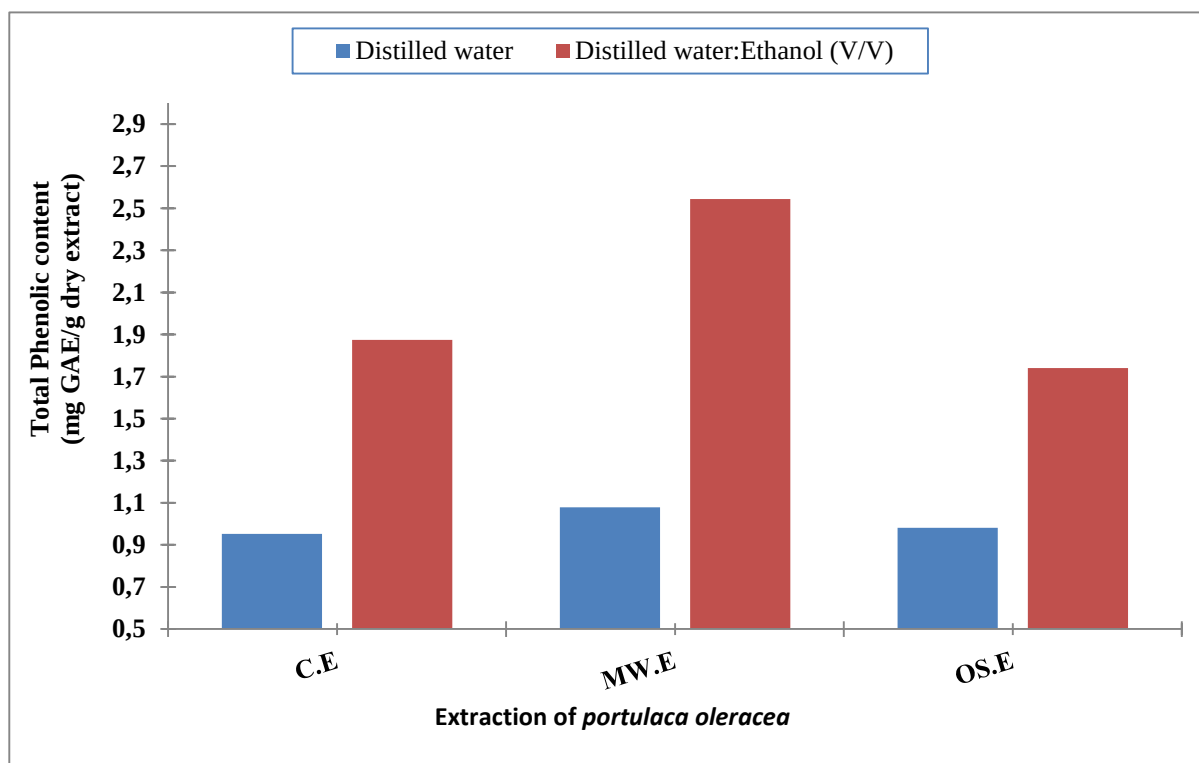


Figure 4.24.Total phenolic contents of *portulaca oleracea* L.

4.3.6. Determination Total Phenol of *Artemisia haussknechtii* boiss

The orbital shaker method with solvents (ethanol: distilled water) extract showed that the best total phenol content in *Artemisia haussknechtii* boiss herb (3.980 mg GAE/g) value significantly ($P < 0.01$). Thus, the conventional methods with solvent water extract was showed a high value (2.976 mg GAE/g) are presented in Table (4.24) and Figure (4.25). On the other hand, the extraction with microwave method with solvents of (distilled water) and (ethanol: distilled water) of total phenol contents were lowest (2.342 mg GAE/g), and (1.964 mg GAE/g). Whereas, the highest and lowest amount of phenol content extracted was obtained through the methods, which provided for the three best extractions. The present study in agreement with (Singh *et al.* 2012; Alavi and Karimi 2017b). Also, approval with (Khanahmadi *et al.* 2009).

Table 4.24.Total phenolic contents of *Artemisia haussknechtii* boiss

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	2.976 $\pm$ 0.000629	2.322 $\pm$ 0.000479	827.884	0.000
MW/E (mg GAE/g)	2.342 $\pm$ 0.000289	1.964 $\pm$ 0.000479	676.634	0.000
OS/E (mg GAE/g)	2.862 $\pm$ 0.000629	3.980 $\pm$ 0.000289	-1615.458-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at (p<0.01)

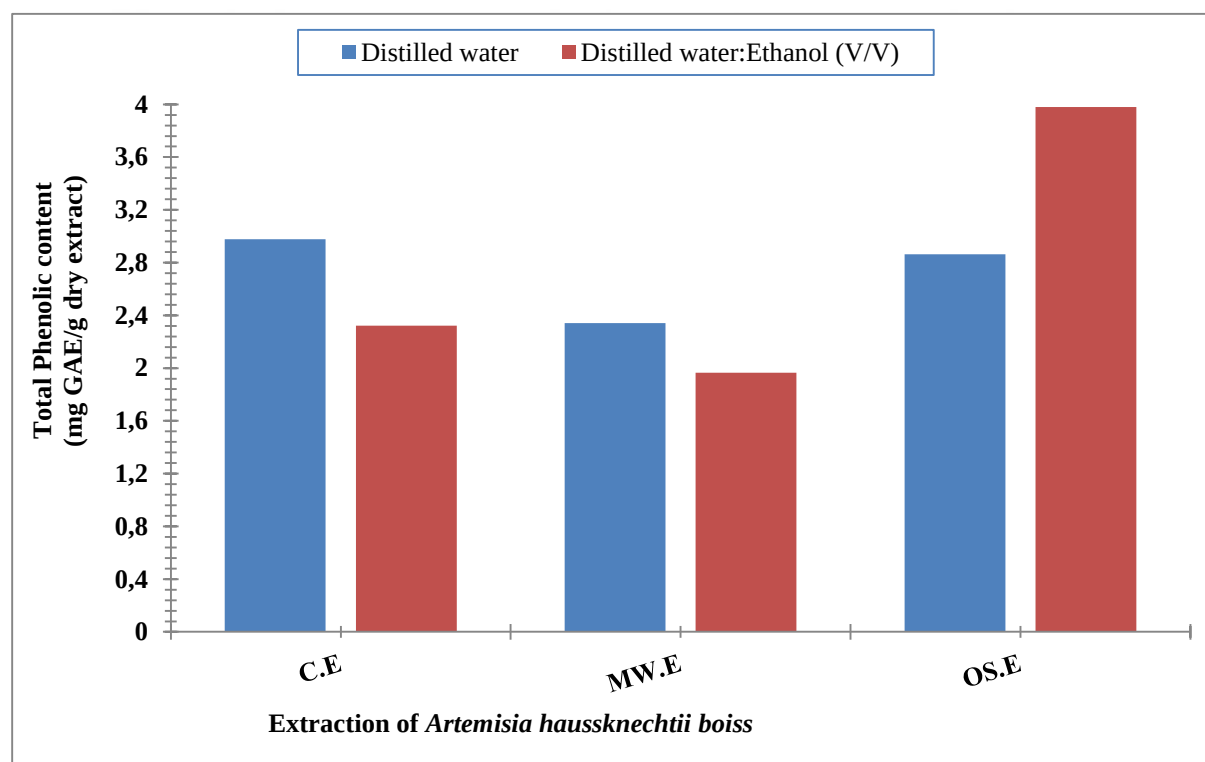


Figure 4.25.Total phenolic contents of *Artemisia haussknechtii* boiss

4.3.7. Determination Total Phenol of *Prunus microcarpa*

The current work, shows the total phenolic content of *Prunus microcarpa* (ethanol: distilled water) extracts was the significant high amount at ($P < 0.01$), (MW/E: 3.374 mg GAE/g) as a function of their concentrations, illustrate Table (4.25), and Figure (4.26), compare with the other methods extraction and solvents. Also, the conventional technique with solvent of distilled water extract was observed high amount as (C/E: 2.614 mg GAE/g) and significantly at level ($p < 0.01$). Furthermore, the total phenol content of the orbital shaker with solvents of (D.W 1.939 mg GAE/g) and (E: DW 0.729 mg GAE/g) indicated the lowest amounts in *Prunus microcarpa*. In general, our research may have revealed new total phenol findings on this herb. Because no have results on total phenolic content in *Prunus microcarpa* by these techniques.

Table 4.25. Total phenolic contents of *Prunus microcarpa*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	2.614 $\pm$ 0.000408	1.963 $\pm$ 0.000629	867.667	0.000
MW/E (mg GAE/g)	0.915 $\pm$ 0.000408	3.374 $\pm$ 0.000479	-3908.827-	0.000
OS/E (mg GAE/g)	1.939 $\pm$ 0.000408	0.729 $\pm$ 0.000408	2095.781	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

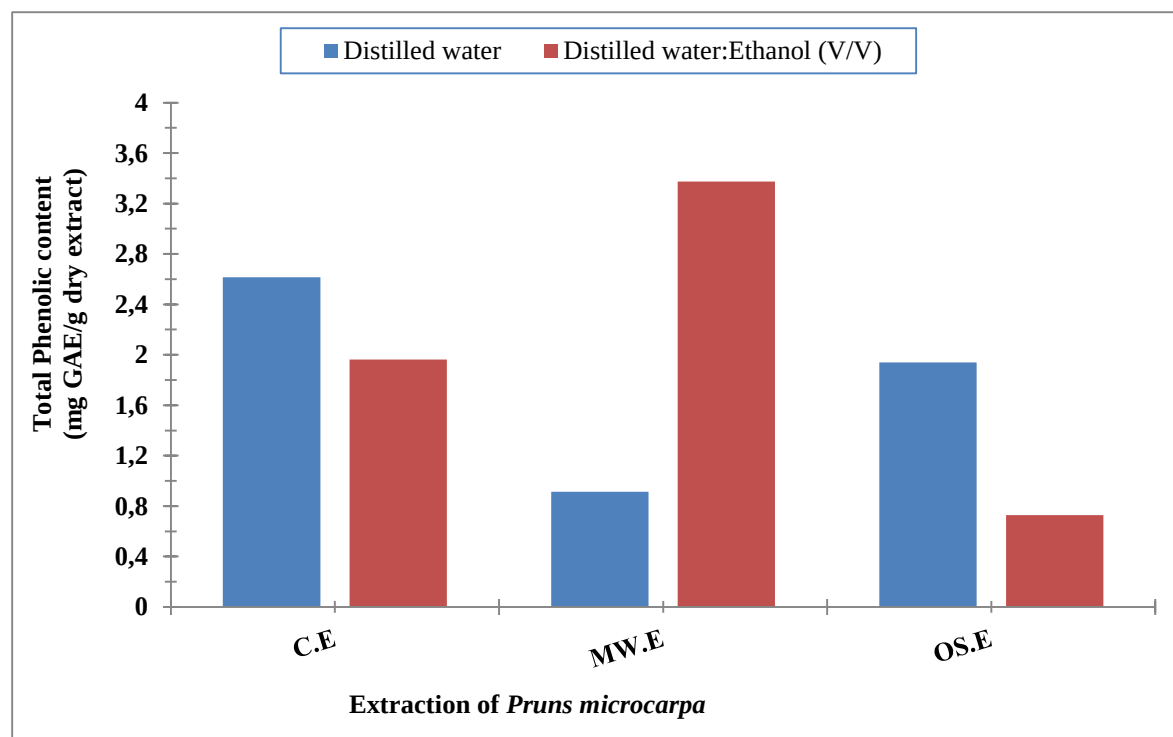


Figure 4.26. Total phenolic contents of *Prunus microcarpa*

4.3.8. Determination Total Phenol of *Silybum Marianum*

Table (4.26) and Figure (4.27), illustrated that the highest total phenol content of *Silybum marianum*, was found from ethanol: distilled water solvents as (C/E: 3.341 mg GAE/g), (MW/E: 3.227 mg GAE/g), and (OS/E: 3.131 mg GAE/g) at (p0.01) respectively. while that the distilled water extract of total phenol content revealed that high or low values were observed (3.120 mg GAE/g) in conventional extraction, (2.562 mg GAE/g) in microwave method, and (1.684 mg GAE/g) in orbital shaker respectively. Consequently, the ethanol: distilled water solvent of total phenol content was significantly (p<0.01), the topmost values which compare with of distilled water solvent with three techniques of extraction. Our study in agrees with (Sun *et al.* 2016), and also approval with (Vaknin *et al.* 2008), that the method was different but the same plant.

Table 4.26. Total phenolic contents of *Silybum Marianum*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	3.120 $\pm$ 0.000479	3.341 $\pm$ 0.000479	-231.166-	0.000
MW/E (mg GAE/g)	2.562 $\pm$ 0.000645	3.277 $\pm$ 0.000479	-890.016-	0.000
OS/E (mg GAE/g)	1.684 $\pm$ 0.000408	3.131 $\pm$ 0.000629	-1929.667-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker
Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water

Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at (p<0.01).

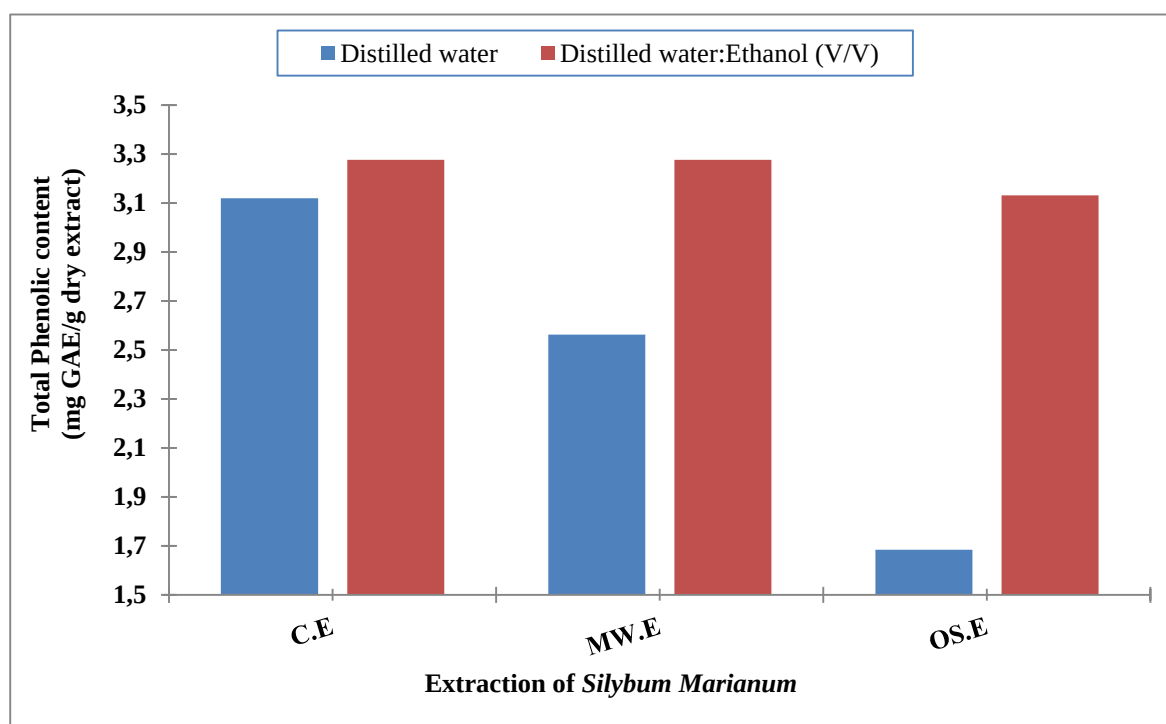


Figure 4.27.Total phenolic contents of *Silybum Marianum*

4.3.9. Determination Total Phenol of *Pistacia eurycarpa* Yale

The total phenol content of the different methods ranged from (CE:3.264 mg GAE/g), (MWE:2.733 mg GAE/g), (OSE: 2.746 mg GAE/g) was significantly high values with solvent ethanol: distilled water ($p<0.01$), also the total phenol content with distilled water was indicate a significant value with both techniques, (CE:3.566 mg GAE/g), (MWE: 2.976 mg GAE/g), while the orbital shaker had the lowest total phenol content (OSE: 1.942 mg GAE/g) with distilled water solvent. The total phenol content in *Pistacia eurycarpa* Yale, maybe influenced by different solvents and extraction techniques, which are shown in Table (4.27), and Figure (4.28). Our study an agreement (Balaky *et al.* 2020), because there are the same extraction methods but the type of plant was different. Thus, our study has indicated a new conclusion of total phenol on *Pistacia eurycarpa* yale

Table 4.27.Total phenolic contents of *Pistacia eurycarpa* Yale

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg GAE/g)	3.566 $\pm$ 0.000408	3.264 $\pm$ 0.000707	369.873	0.000
MW/E (mg GAE/g)	2.976 $\pm$ 0.000854	2.733 $\pm$ 0.000645	226.777	0.000
OS/E (mg GAE/g)	1.942 $\pm$ 0.000645	2.746 $\pm$ 0.000479	-1000.762-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p<0.01$)

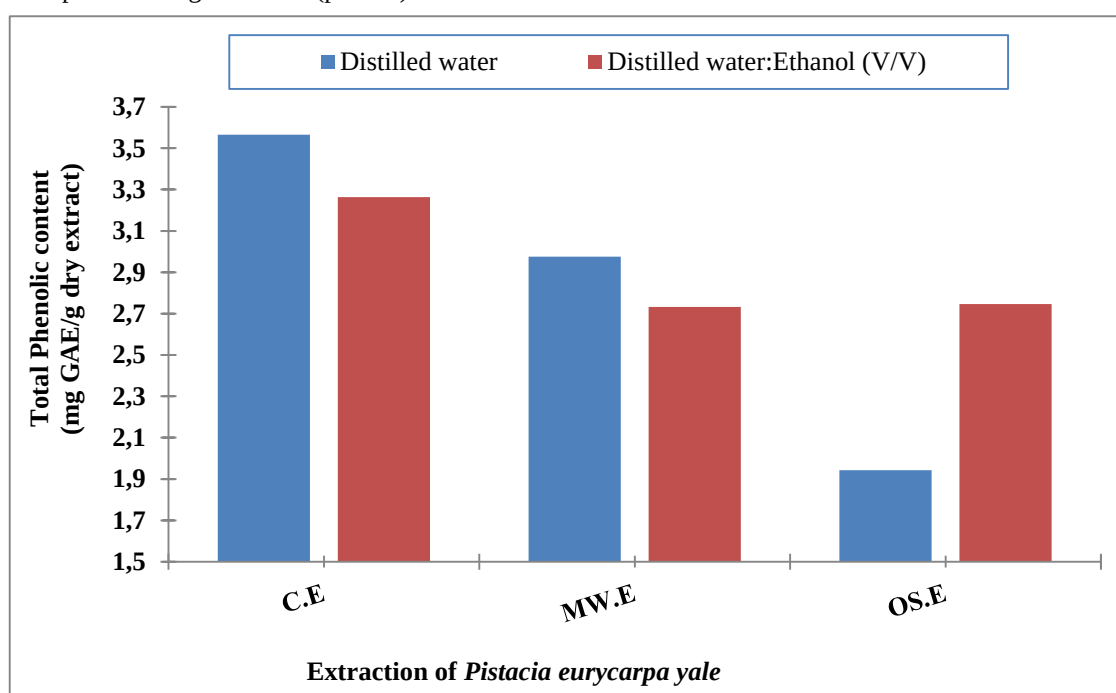


Figure 4.28.Total phenolic contents of *Pistacia eurycarpa* Yale

In our study, all plants a good source of total phenol content and also a significant value ($p < 0.01$) through various techniques and solvents. In addition, the orbital shaker extraction of total phenol content was better than to compare with other methods as the microwave method and conventional extraction. That agrees with this report (Balaky *et al.* 2020; Holem *et al.* 2021).

Therefore, the phenolic compounds the most important biologically active compound found in many plant extract (Aly and Bafeel 2009), thus the Phenolic compounds play an important role in stabilizing lipid oxidation and are linked to antioxidant activity. The mechanism of phenolic compounds for the production of antioxidants belongs to their radical scavenging ability, which is mediated by hydroxyl groups (Hatann *et al.* 1989), furthermore, Many plants contain phenolic compounds that have been shown to have important activities, including potent antioxidant activity (Chandini *et al.* 2008; Duh *et al.* 1999).

The retrieval of phenolic compounds was purely dependent on the solvent used and its polarity for different plant materials and also affected by their solubility in the specific solvent. Moreover, the solubility of the solvent plays a fundamental role in increasing the solubility of phenolic compounds in it (Alothman *et al.* 2009). Briefly, our study is indicated a new conclusion of total phenol content from most plants, also showed that types of extraction and solvents are extremely influenced.

4.4. Determination Total flavonoid of all plant studies

Flavonoids, a big family of compounds with a C6-C3-C6 framework, are the most abundant polyphenols in the human diet. Polyphenols are considered fundamental to human health and have far-reaching pharmacological effects on human health, especially in the prevention of age-related diseases (Govindarajan *et al.* 2005), thus the flavonoids are currently subgroups of phenolic compounds. They are the largest and best-studied classes of secondary metabolites (Grotewold 2006)

4.4.1. Determination Total flavonoid of *Eremurus Spectabilis Bieb*

As for as different techniques and solvents are concerned, illustrate Table (4.28), and Figure (4.29), the extracts obtained by the application of conventional extraction with polar ethanol: distilled water significantly ($p < 0.01$), (0.238 mg QE/g) higher flavonoid content as compared to the extracts obtained as a result of using microwave extraction (0.183 mg QE/g), and the orbital shaker (0.137 mg QE/g) of *Eremurus Spectabilis Bieb*, while as compared with distilled water solvent was arranged from high to low values followed (CE; 0.185 mg QE/g), (MW/E: 0.166 mg QE/g) and (OS/E: 0.101 mg QE/g) respectively. Consequently, our study clearly showed that the total flavonoid content of the conventional technique with (ethanol: distilled water) solvent was significantly highest values from *Eremurus spectabilis Bieb*. This research is consistent with those found in the literature (Tuzcu and Agca 2017; Dervişoğlu *et al.* 2014), thus approval with (Bircan and Kırbağ 2015).

Table 4.28. Total Flavonoid content in *Eremurus Spectabilis Bieb*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.185 $\pm$ 0.000289	0.238 $\pm$ 0.000854	-59.076-	0.000
MW/E (mg QE/g)	0.166 $\pm$ 0.000479	0.183 $\pm$ 0.000645	-20.843-	0.000
OS/E (mg QE/g)	0.101 $\pm$ 0.000479	0.137 $\pm$ 0.000289	-63.952-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

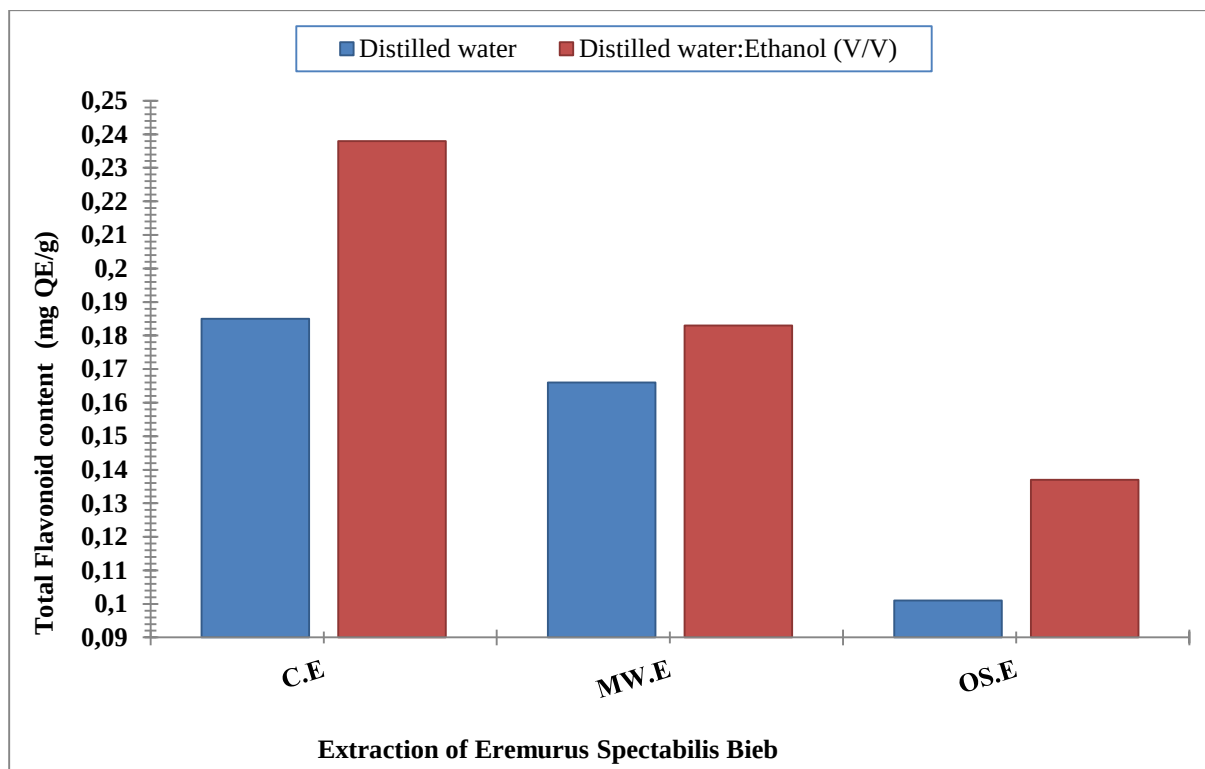


Figure 4.29. Total Flavonoid content in *Eremurus Spectabilis Bieb*

4.4.2. Determination Total flavonoid of *Prosopis frakta*

Total flavonoid content of *Prosopis frakta* from Table (4.29), and Figure (4.30), was determined by utilizing the different methods and solvents in the *Prosopis frakta*, the conventional method with polar water solvent was observed highest values from both (C/E: 0.182 mg QE/g), Microwave method (MW/E:0.086 mg QE/g), also the total flavonoid content showed significantly high value (0.098 mg QE/g) at ($p < 0.01$), with (ethanol: distilled water) solvents of orbital shaker, while a compare with water solvent was less amount (0.056 mg QE/g) in *Prosopis frakta*. In general, the polar water of total flavonoid extracted through method conventional was had the significant highest value at ($p < 0.01$).

Our study approval with (Mohammed *et al.* 2018), also agreement with research (Fariba Jafari *et al.* 2013), and the (Alharbi *et al.* 2017), studied total flavonoids content from *Prosopis frakta* act as a powerful antioxidant and reduce total cholesterol, triglycerides, malondialdehyde, and rise HDL-C in rats with tetrachloride-induced hepatotoxicity.

Table 4.29.Total Flavonoid content in *Prosopis frakta*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.182 $\pm$ 0.000408	0.071 $\pm$ 0.000645	144.678	0.000
MW/E (mg QE/g)	0.086 $\pm$ 0.000645	0.078 $\pm$ 0.000645	8.764	0.000
OS/E (mg QE/g)	0.056 $\pm$ 0.000479	0.098 $\pm$ 0.000408	-66.359-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at (p<0.01).

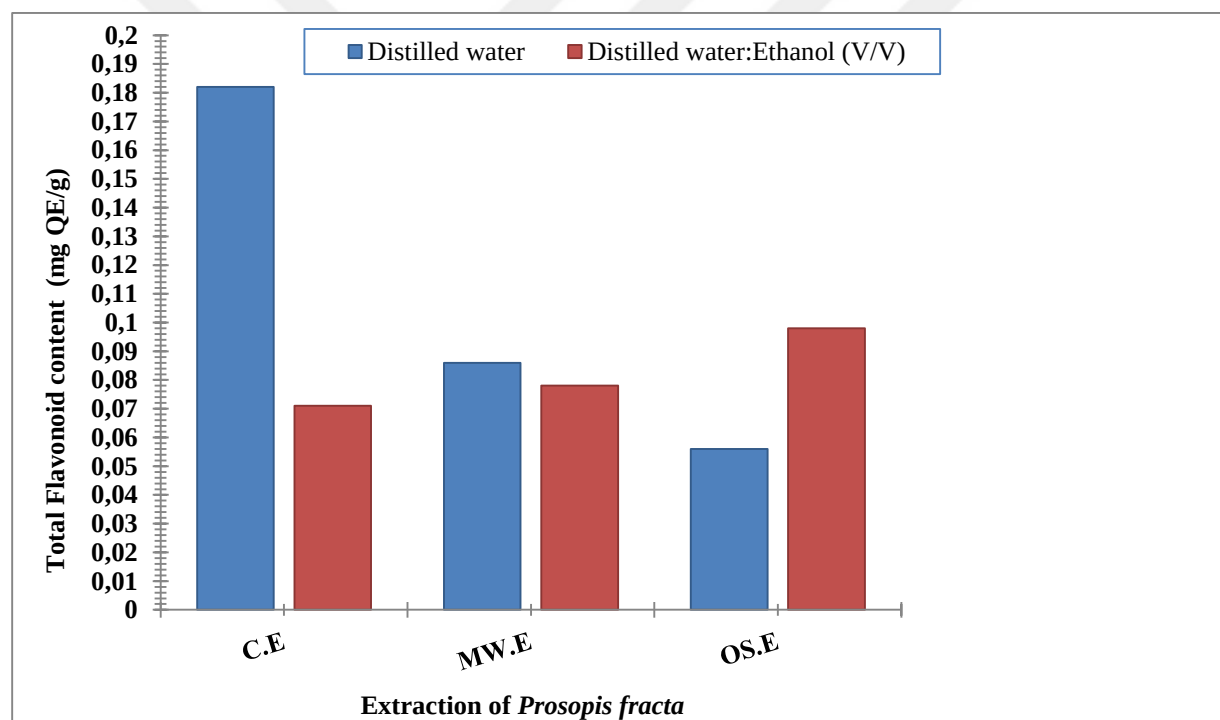


Figure 4.30.Total Flavonoid content in *Prosopis frakta*

4.4.3. Determination Total flavonoid of *Capparis spinosa*

The total flavonoid content values of *Capparis spinosa* extract with (ethanol water) through of techniques (conventional, orbital shaker, and microwave) extractions were showed significantly ($p<0.01$) highly amount (C/E: 0.142 mg QE/g), (OS/E: 0.098 mg QE/g), and (MW/E: 0.086 mg QE/g) respectively, that compared with the distilled water solvent and other techniques extraction. However, that the lowest total flavonoid content belongs to *Capparis spinosa* of distilled water extract was (MW/E: 0.065 mg QE/g), (OS/E: 0.078 mg QE/g), and (C/E: 0.077 mg QE/g), respectively. Completely, in our study, the microwave extraction with solvent (ethanol: water) was significantly ($p<0.01$) the best technique to extract total flavonoid content from *capparis spinose*, as shown in Table (4.30), and Figure (4.31).

The present study agrees with what has been reported (Mahboubi and Mahboubi. 2014), that is approved with (Allaith 2016; Tlili *et al.* 2017) had reported, that the Total free phenolics, total flavonoids, and total carotenoids have a high correlation with antioxidant capacities of *Capparis spinosa*. In addition, these findings are in line with the study of (Arrar *et al.* 2013), and (Zhou *et al.* 2011; Mohebbali *et al.* 2018), who reported that flavonoids in *Capparis spinosa* have anti-inflammatory activity (Sultan and Çelik 2009), which reported, that *Capparis spinosa* was shown to contain quantities of antioxidant phytochemicals like flavonoids and other polyphenols. Further, naturally occurring products from the order *Capparales*, also known as flavor compounds, anticarcinogenic agents, and biopesticides (Mikkelsen *et al.* 2000).

Table 4.30.Total Flavonoid content in *Capparis spinose*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.077 $\pm$ 0.000645	0.142 $\pm$ 0.000645	-71.204-	0.000
MW/E (mg QE/g)	0.065 $\pm$ 0.000479	0.086 $\pm$ 0.000629	-26.563-	0.000
OS/E (mg QE/g)	0.078 $\pm$ 0.000408	0.098 $\pm$ 0.000479	-32.186-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p<0.01$).

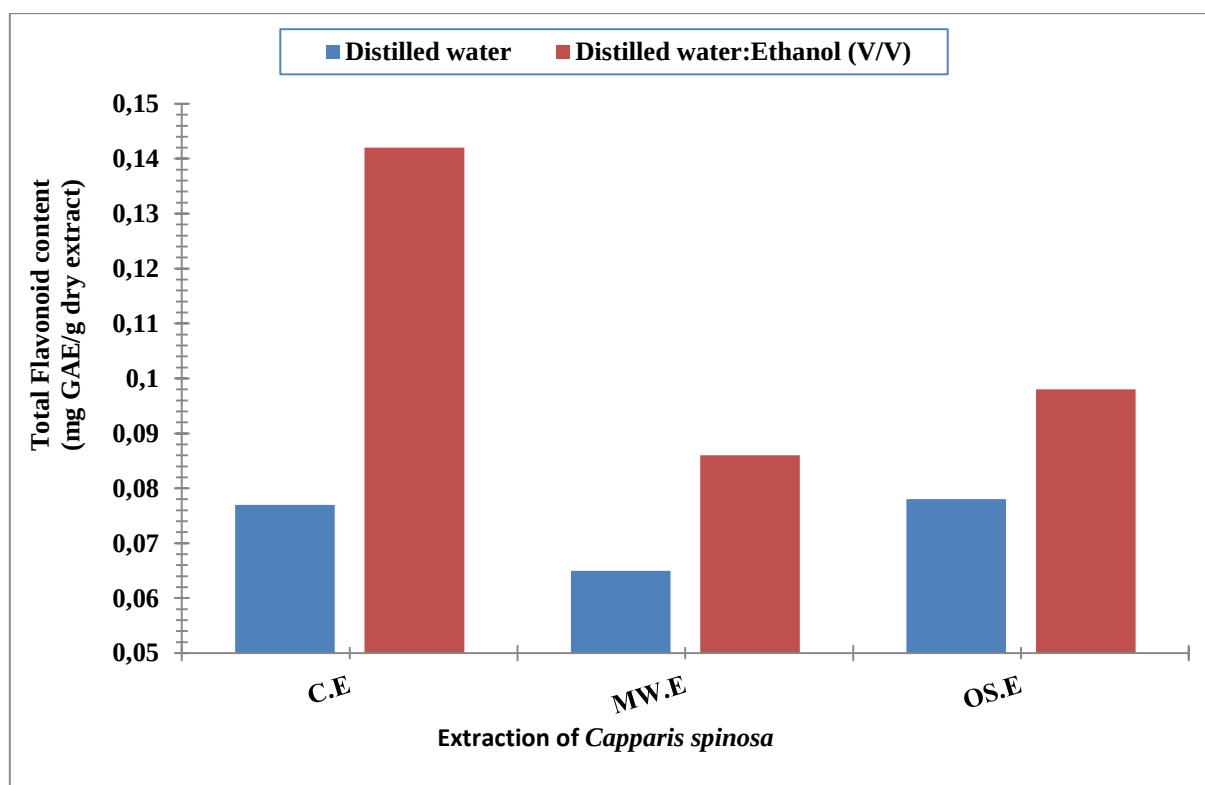


Figure 4.31. Total Flavonoid content in *Capparis spinosa*

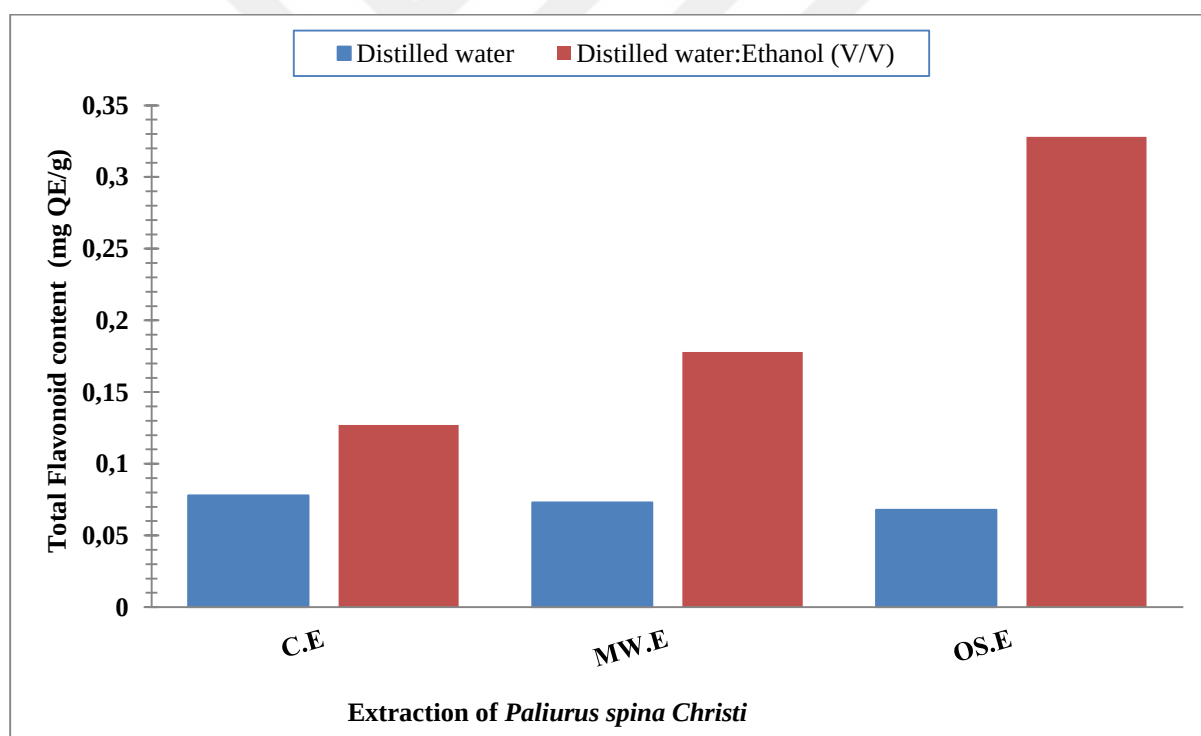
4.4.4. Determination Total flavonoid of *Paliurus spina Christi*

Our findings are shown in Table (4.31), and Figure (4.32). The extraction of the total flavonoid content of *Paliurus spina Christi* by different techniques with dissimilar solvents revealed that the orbital shaker technique with solvent of (ethanol: distilled water) had the significantly highest value (0.328 mg QE/g) at ($p < 0.01$), and also had high values with (MW/E: 0.178 mg QE/g), and (C/E: 0.127 mg QE/g), in comparison with the (distilled water) solvent and three methods extraction, that revealed the lowest values (C/E: 0.078 mg QE/g), (MW/E: 0.073 mg QE/g), (OS/E: 0.068 mg QE/g), respectively. our results are in line with the report of (AHMED *et al.* 2013), who reported that *Paliurus spina-christi* is a rich source of the flavonoids quercetin, rutin, hyperoside, and quercetin-3-O- diglucoside (Dalakishvili *et al.* 1986; Brantner and Maleš 1990). in conclusion, the *Paliurus spina Christi* extract with solvent of (ethanol: distilled water), especially with orbital shaker technique, was presented a significantly total flavonoid content.

Table 4.31.Total Flavonoid content in *Paliurus spina Christi*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.078 $\pm$ 0.000408	0.127 $\pm$ 0.000408	-84.870-	0.000
MW/E (mg QE/g)	0.073 $\pm$ 0.000479	0.178 $\pm$ 0.000629	-132.816-	0.000
OS/E (mg QE/g)	0.068 $\pm$ 0.000479	0.328 $\pm$ 0.000289	-465.549-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

Figure 4.32Total Flavonoid content in *Paliurus spina Christi*

4.4.5. Determination Total flavonoid of *portulaca oleracea* herb

The highest value of total flavonoid content of the *portulaca oleracea* herb was observed for the solvent (ethanol: distilled water) through techniques of (C/E: 0.214 mg QE/g), (MW/E: 0.147 mg QE/g), and (OS/E: 0.107 mg QE/g), while compare with (distilled water) extract was showed a lowest total flavonoid with other methods of extraction. Thus, the total flavonoid content from methods (C/E and OS/E) with solvent (distilled water) extract was observed same amounts 0.087 mg QE/g, shown in Table (4.32), and Figure (4.33). Our study does not agree with the report (Liang *et al.* 2014). But agree with (Anthonia Ezeabara 2014), because a reported both aqueous and ethanol extracts would produce a suitable level of alkaloid, flavonoid, phenol, and sterol. On the other hand, the main active ingredients of *portulaca oleracea* L are flavonoids (Yan *et al.* 2012). Also approval with (Nagarani *et al.* 2014).

Earlier pharmacological studies have also shown that flavonoids from *Portulaca oleracea* L. have anti-oxidant (Erkan 2012), anti-bacterial (Cushnie and Lamb 2005), and anti-inflammatory (Tadić *et al.* 2012). Basically, the methods of conventional extraction and other techniques were showed significantly ($p < 0.01$) best results through solvent of (ethanol: distilled water) extract in *portulaca oleracea* L.

Table 4.32.Total Flavonoid content in *portulaca oleracea* L

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.087 $\pm$ 0.000289	0.214 $\pm$ 0.000645	-179.605-	0.000
MW/E (mg QE/g)	0.065 $\pm$ 0.000479	0.147 $\pm$ 0.000408	-129.937-	0.000
OS/E	0.087 $\pm$ 0.000479	0.107 $\pm$ 0.000479	-28.803-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

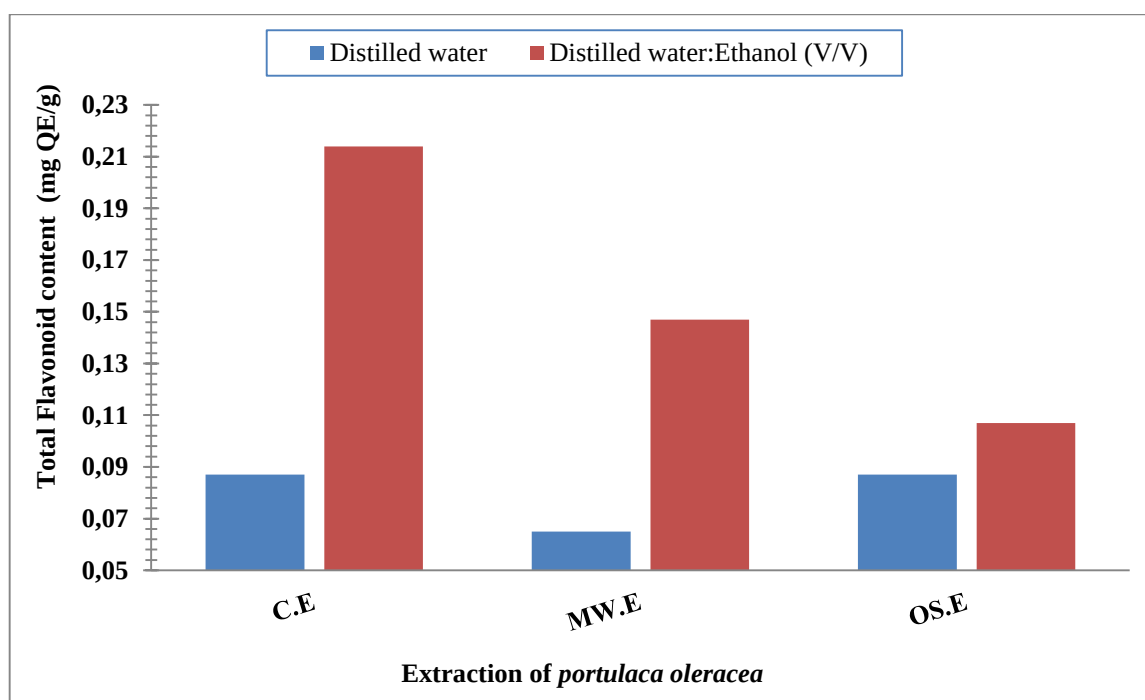


Figure 4.33.Total Flavonoid content in *portulaca oleracea L*

4.4.6. Determination Total flavonoid of *Artemisia haussknechtii boiss*

The total flavonoid content in *Artemisia haussknechtii boiss* herb, which the extracted by solvent (distilled water) using the conventional method and solvent (ethanol: distilled water) using the microwave method, was observed a same significantly highly values (0.236 mg QE/g), at ($p < 0.01$), compared with other extraction methods. On the other hand, obtaining the total flavonoid content of the solvent (distilled water) with two methods (Microwave and orbital shaker) was given the same quantities (0.132 mg QE/g), shown in Table (4.33), and Figure (4.34).

Our study agrees with (Amiri and Goodarzi 2017), also Another report showed that *Artemisia haussknechtii-Boiss* extract which contains flavonoids and phenols had a suitable ability to synthesize greens, among other phytochemicals. (Alavi and Karimi 2017b). Consequently, the microwave and conventional techniques with both solvents to extract the total content of flavonoids were the significant values of *Artemisia haussknechtii boiss*

Table 4.33.Total Flavonoid content in *Artemisia haussknechtii* boiss

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.236 $\pm$ 0.000629	0.208 $\pm$ 0.000479	35.418	0.000
MW/E (mg QE/g)	0.131 $\pm$ 0.000479	0.236 $\pm$ 0.000645	-129.723-	0.000
OS/E (mg QE/g)	0.132 $\pm$ 0.000408	0.168 $\pm$ 0.000479	-57.617-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

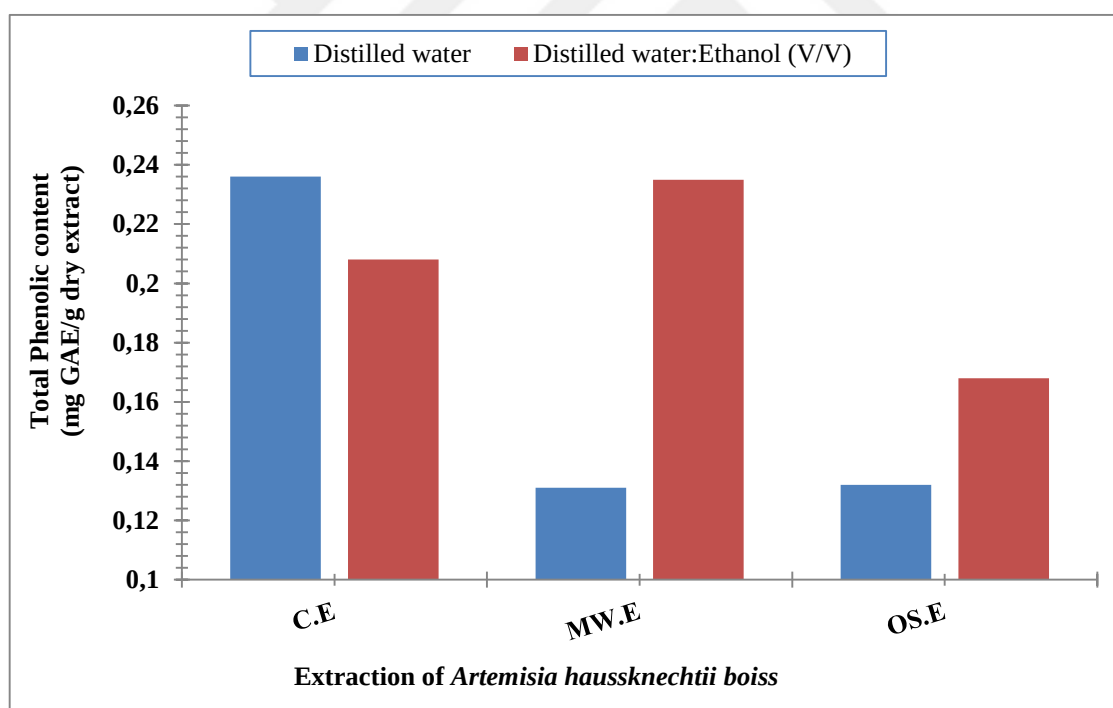


Figure 4.34.Total Flavonoid content in *Artemisia haussknechtii* boiss

4.4.7. Determination Total flavonoid of *Prunus microcarpa*

It was observed that the solvent (ethanol: distilled water) to extract the total content of flavonoid in *Prunus microcarpa* that has a higher level by applying the techniques (OS/E: 0.217 mg QE/g), (CE: 103 mg QE/g), and (MWE: 0.107 mg QE/g), with all significant ($p < 0.01$) value. That is illustrated in Table (4.34), and figure (4.35). Thus, also the total content of flavonoid was observed low values with the solvent (distilled water) by all methods respectively. In addition, the microwave technique gives a lower value through the solvent (distilled water). Compared to another method and (ethanol: distilled water) solvent.

Essentially, that the techniques extraction by solvent (ethanol: distilled water) of *Prunus microcarpa* were got the significant values ($p < 0.01$) of total flavonoid content. According to the study of Jissain and Majid (2003), that the *Prunus microcarpa* was the rich source to the presence of rhamnose and glucose. This work agreement with some other studies on different species such as (Zahid *et al.* 2019) and (Belhadj *et al.* 2016).

Table 4.34.Total Flavonoid content in *Prunus microcarpa*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.091 $\pm$ 0.000479	0.117 $\pm$ 0.000645	-32.042-	0.000
MW/E (mg QE/g)	0.068 $\pm$ 0.000408	0.103 $\pm$ 0.000250	-74.679-	0.000
OS/E (mg QE/g)	0.081 $\pm$ 0.000645	0.217 $\pm$ 0.000289	-192.333-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

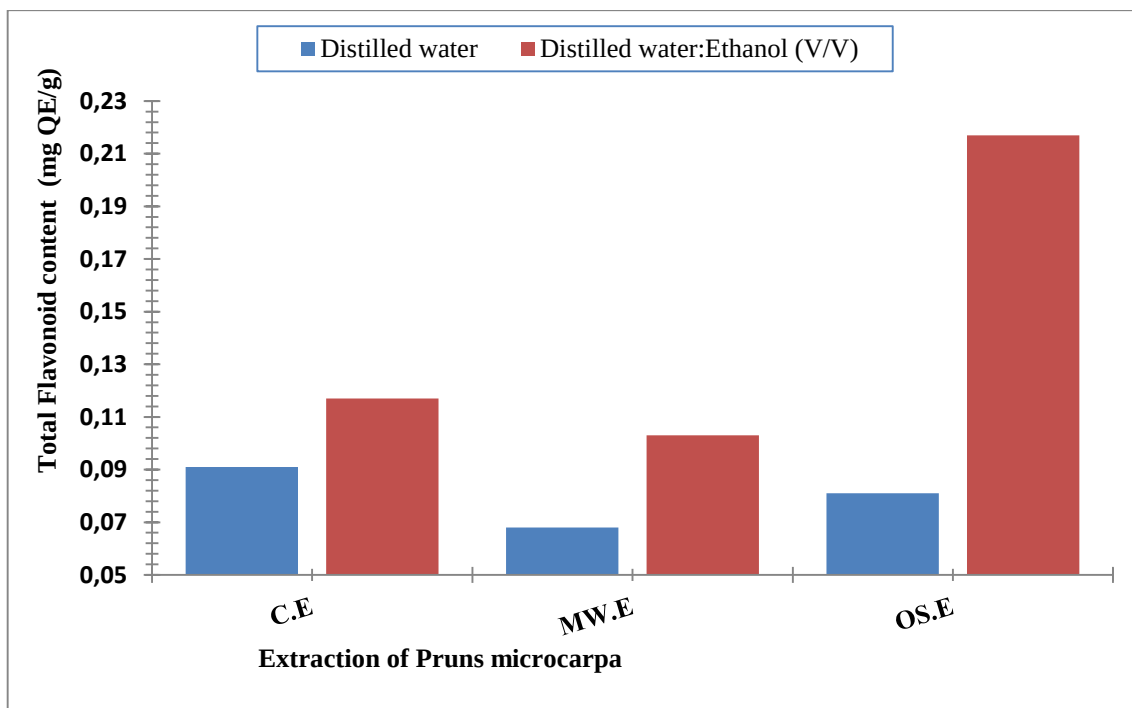


Figure 4.35. Total Flavonoid content in *Prunus microcarpa*

4.4.8. Determination Total flavonoid of *Silybum Marianum*

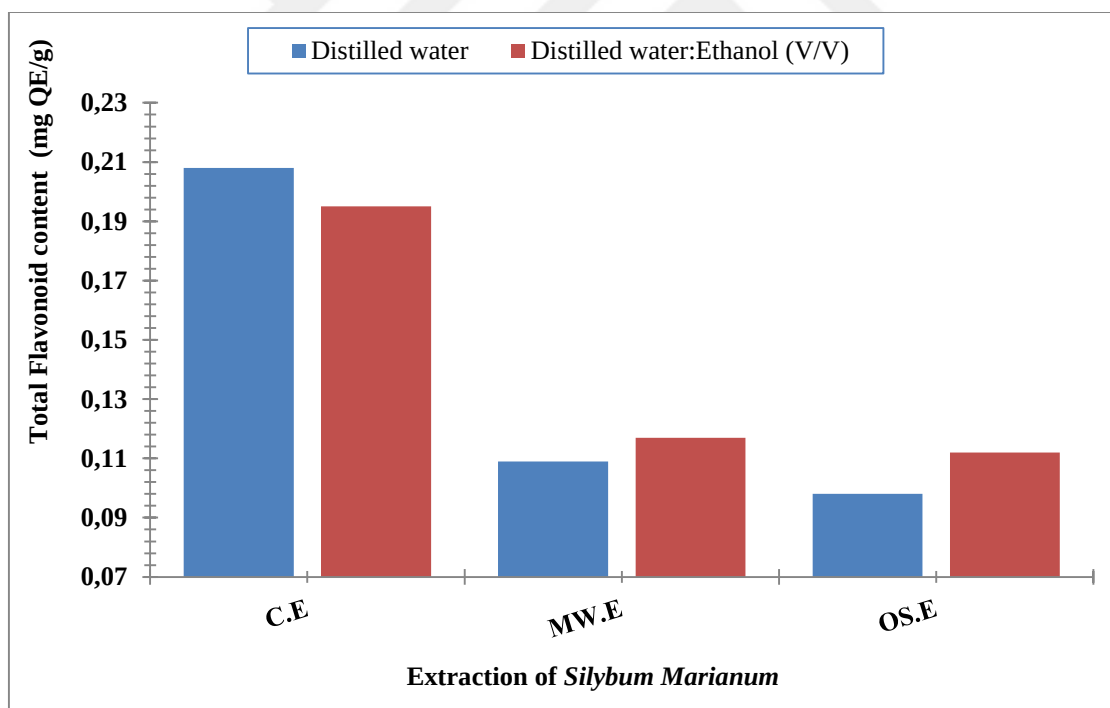
Depend on the Table (4.35), and Figure (4.36). statistically, the highest significant value ($p < 0.01$) of total flavonoid content using the conventional method with solvent were observed (DW: 0.208 mg QE/g) and the solvent (E: DW 0.195 mg QE/g) respectively, on the other hand, also the method of microwave extraction with solvents (distilled water 0.109 mg QE/g), and (ethanol: distilled water 0.117 mg QE/g) was observed high values ($p < 0.01$), respectively. Whereas the lowest total flavonoid content through using the orbital shaker with solvents (distilled water: 0.098 mg QE/g), and (ethanol: distilled water: 0.012 mg QE/g) was observed from *Silybum Marianum*.

Lastly, our results indicated that the total flavonoid content of *Silybum Marianum*, was shown significantly values with all methods extraction through solvents distilled water and ethanol: distilled water. Our results agree with (Sun *et al.* 2016; Ghafor *et al.* 2014). Another report by Vaknin *et al.* (2008), with that the milk thistle *Silybum marianum* is a good source of healthy edible sprouts. Although, the pharmacological activity of *Silybum marianum* is based on a mixture of flavonoids known as *silymarin* as described by all researchers (Smith *et al.* 2005; Morelli 1978; Lee and Liu 2003).

Table 4.35.Total Flavonoid content in *Silybum Marianum*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.208 $\pm$ 0.000408	0.195 $\pm$ 0.000289	25.000	0.000
MW/E (mg QE/g)	0.109 $\pm$ 0.000408	0.117 $\pm$ 0.000479	-13.908-	0.000
OS/E (mg QE/g)	0.098 $\pm$ 0.000289	0.112 $\pm$ 0.000645	-19.799-	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

Figure 4.36.Total Flavonoid content in *Silybum Marianum*

4.4.9. Determination Total flavonoid of *Pistacia eurycarpa Yale*

Table (4.36), and Figure (4.37). The total flavonoid content of *Pistacia eurycarpa Yale*, by using the conventional method with solvents (ethanol: distilled water), and (distilled water) were observed significantly ($p<0.01$) maximum values as (0.129 mg QE/g) and (0.125 mg QE/g), respectively. Thus, also the total flavonoid content was observed high value through the microwave method with solvents (ethanol: distilled water 0.097 mg QE/g), and (distilled water 0.087 mg QE/g), when compared with the lowest total flavonoid content was obtained from *Pistacia eurycarpa Yale* extracted by solvents (ethanol: distilled water 0.065 mg QE/g) and (distilled water 0.073 mg QE/g) with using the orbital shaker, respectively.

Obviously, that the conventional method was better than the other methods (microwave and orbital shaker) to given the highest significant ($p<0.01$) values of total flavonoid content in *Pistacia eurycarpa yale*. Our study agrees with reports but different species of plant (Amel *et al.* 2016; Ayşenur *et al.* 2016). Therefore, this finding may have provided new studies on this plant. In another study (Maskan and Karataş 1998), report that the great source of essential unsaturated fatty acids was *Pistacia eurycarpa yale*.

Table 4.36.Total Flavonoid content in *Pistacia eurycarpa Yale*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E (mg QE/g)	0.125 $\pm$ 0.000289	0.129 $\pm$ 0.000408	-7.000-	0.000
MW/E (mg QE/g)	0.087 $\pm$ 0.000289	0.097 $\pm$ 0.000645	-14.142-	0.000
OS/E (mg QE/g)	0.073 $\pm$ 0.000645	0.065 $\pm$ 0.000289	11.314	0.000

Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p<0.01$).

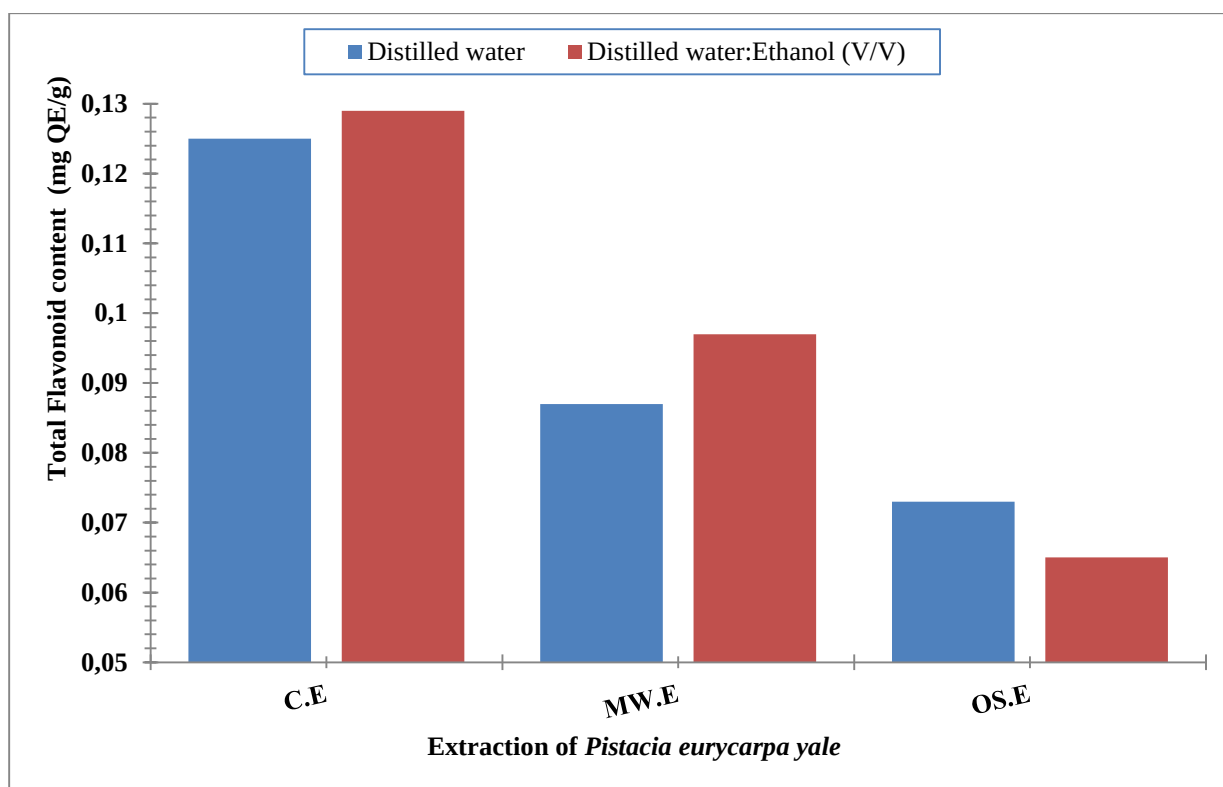


Figure 4.37. Total Flavonoid content in *Pistacia eurycarpa Yale*

According to our study, the method of conventional extraction from all plants was observed as the best significantly ($p < 0.01$) through extracted total flavonoid content with solvents, compared with other techniques (Balaky *et al.* 2020).

Flavonoids are divided into two categories: flavones and flavonols. Flavonols are antioxidants that serve as both radical scavengers and metal chelators (Bors and Saran 1987). Plant flavonoids, phenolic acids, terpenoids, and other polyphenolic compounds also have a variety of biological properties, such as anti-inflammatory, antimicrobial, antitumor, and platelet aggregation inhibition (Akdemir *et al.* 2011; Prior 2003; Formica and Regelson 1995).

On the other hand, total flavonoids and total phenols in plants are the most important bioactive compounds that are an important source of antioxidants (Zhang *et al.*, 2016). Recently, in some of our plants, no studies have been identified in the literature on the essential raw extract obtained with solvents (ethanol: pure water) and (distilled water) with various techniques (conventional, microwave, and orbital shaker).

4.5. Determination Total Anthocyanin of all plant studies

Anthocyanins are an important category of flavonoids that represent a large group of secondary metabolites of plants (Tanaka and Ohmiya 2008), anthocyanins are a group of water-soluble blue, purple, and red pigments whose hue, a property of color, is influenced by 600 anthocyanins have been identified in nature (Smeriglio *et al.* 2016; Ignat *et al.* 2011), anthocyanin's are believed to offer a wide variety of health benefits, such as prevention of heart disease, inhibition of carcinogenesis and inflammatory activity in the brain (Bagchi *et al.* 2004; Galli *et al.* 2006; Tsang *et al.* 2005), cardiovascular diseases and cancer (Caridi *et al.* 2007).

4.5.1. Determination Total Anthocyanin of *Eremurus Spectabilis Bieb*

Depend on the Table (4.37) and, Figure (4.38). That indicates the total anthocyanin content of *Eremurus Spectabilis Bieb*, by using the orbital shaker technique was detected in solvent (ethanol: distilled water) a highest significant value (OSE: 165.32 mg/kg) ($p < 0.01$), and more effective in comparison with solvent distilled water, thus by the using microwave method was detected significant value of total anthocyanin content (MWE: 87.67 mg/kg), in the same solvent while using in orbital shaker extraction, while the total anthocyanin content wasn't detected in conventional method due to the presence of high heating. Consequently, the orbital shaker technique was more effective than other techniques to given total anthocyanin content in solvent (Ethanol: distilled water) extract.

Table 4.37. Total Anthocyanin content in *Eremurus Spectabilis Bieb*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E mg/kg	Mean $\pm$ S.E mg/kg	T-test	P-value
C/ E	-	-	-	-
MW/E	58.86 $\pm$ 0.00064	87.67 $\pm$ 0.00047	-35849.805-	0.000
OS/E	27.97 $\pm$ 0.00085	165.32 $\pm$ 0.00085	-113736.605-	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$)

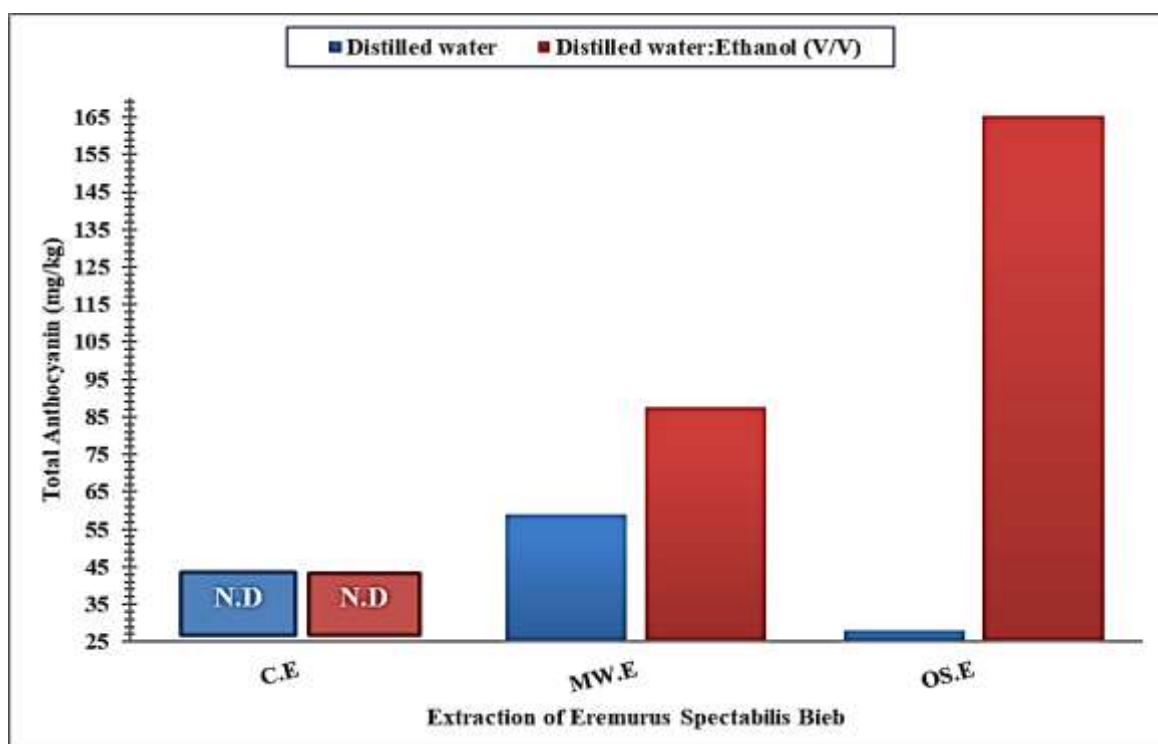


Figure 4.38. Total Anthocyanin content in *Eremurus Spectabilis Bieb*

4.5.2. Determination Total Anthocyanin of *Prosopis fracta*

In our study, the total anthocyanin of *Prosopis fracta* was detected significant quantities in solvent (distilled water) by using the orbital shaker (OSE: 10.85 mg/kg) ($p < 0.01$), compared with the same solvent (distilled water) was not detected value by using the methods conventional and microwave extraction. On the other hand, the total anthocyanin content was detected in (ethanol: distilled water) by using the microwave method (MWE: 2.50 mg/kg), while compared with other methods. Completely, the orbital shaker effective method with (ethanol: distilled water) gives strong quantities of total anthocyanin content in *Prosopis fracta*, illustrate in Table (4.38), and Figure (4.39).

Table 4.38. Total Anthocyanin content in *Prosopis fracta*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	-	-	-	-
MW/E	-	2.50 $\pm$ 0.00	-3873.758-	0.000
OS/E	10.85 $\pm$ 0.00	-	16811.07	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$)

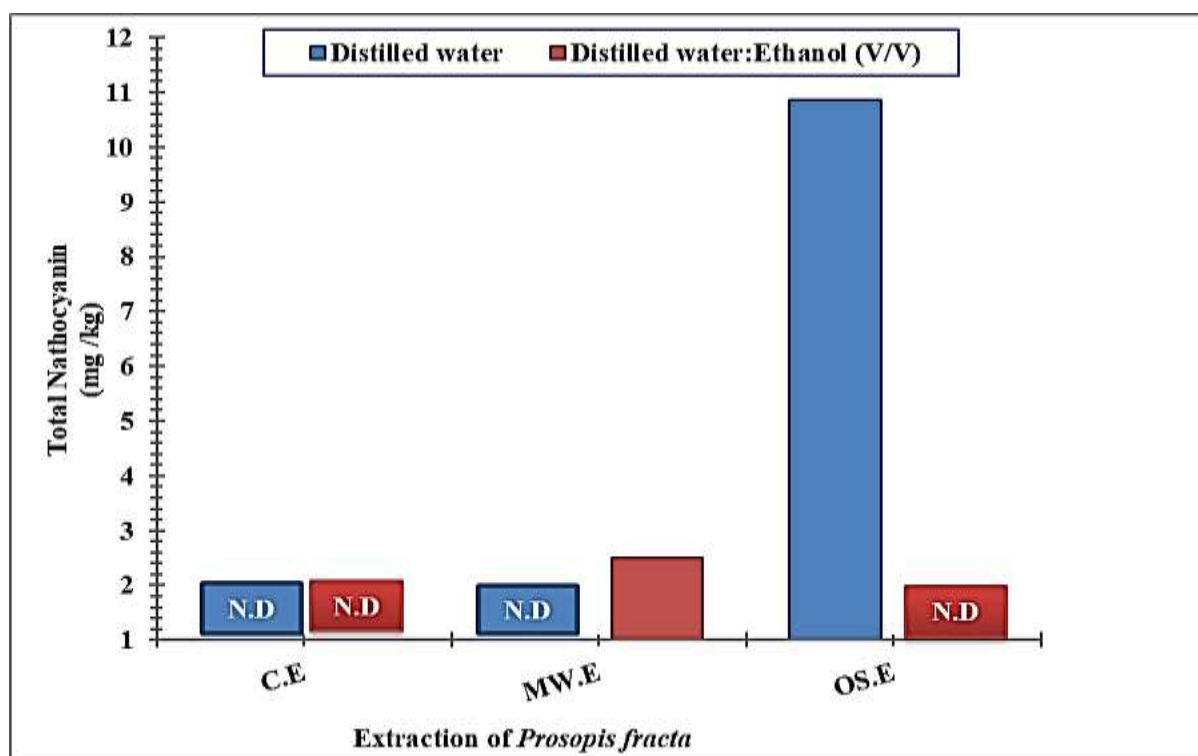


Figure 4.39. Total Anthocyanin content in *Prosopis fratta*

4.5.3. Determination Total Anthocyanin of *Capparis spinosa*

The total anthocyanin content of *Capparis spinosa* in solvent (distilled water), was detected a significant ($p < 0.01$), highly values by using the methods (OS/E: 10.02 mg/kg), (C/E: 23.38 mg/kg) respectively. In comparison with solvent (ethanol: distilled water) was not detected by methods of (conventional and microwave) extraction. Whereas, that the orbital shaker in solvents (distilled water) and (ethanol: distilled water) was gives detected quantities of total anthocyanin content OS/E (10.02 mg/kg) and (18.37 mg/kg) respectively. Table (4.39) and, Figure (4.40). Eventually, that the conventional method more effective than orbital shaker extraction.

Table 4.39. Total Anthocyanin content in *Capparis spinosa*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E mg/kg	Mean $\pm$ S.E mg/kg	T-test	P-value
C/ E	23.38 $\pm$ 0.00	-	48839.749	0.000
MW/E	2.92 $\pm$ 0.00	-	6102.292	0.000
OS/E	10.02 $\pm$ 0.00	18.37 $\pm$ 0.00	-9262.107-	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at (p<0.01).

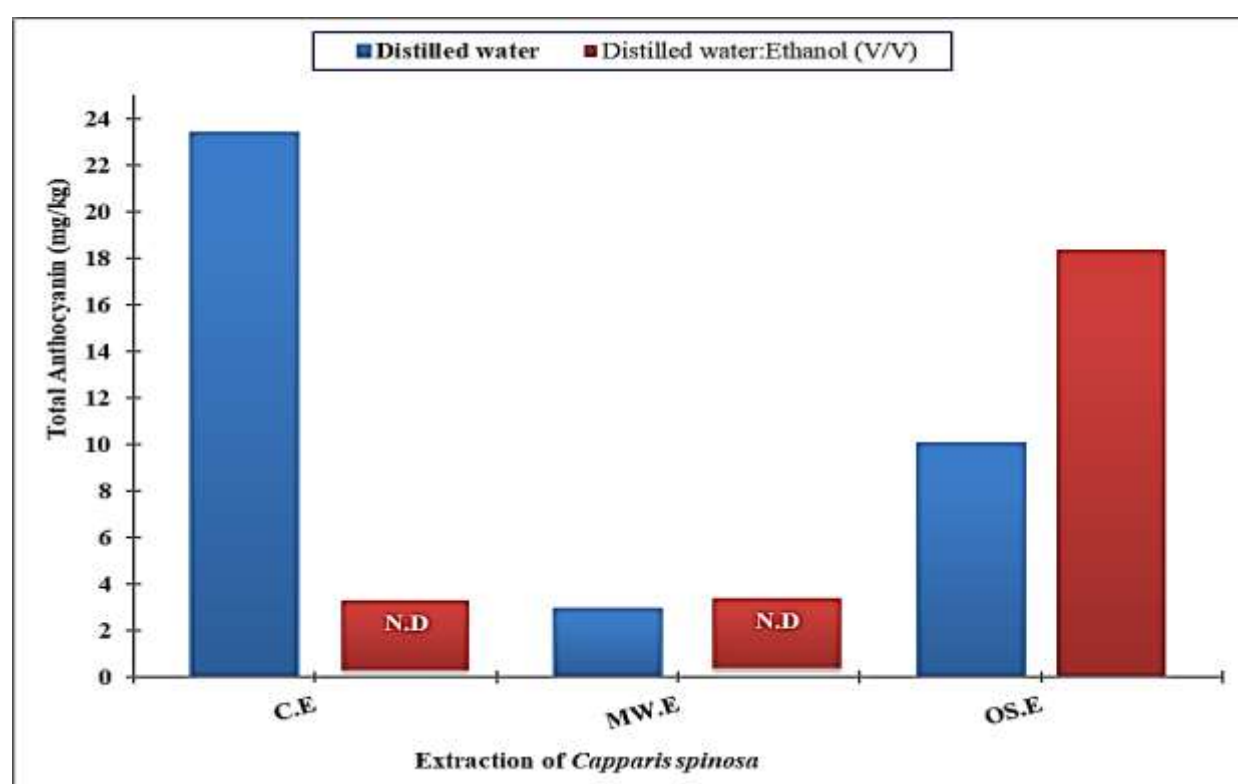


Figure 4.40. Total Anthocyanin content in *Prosopis fratta*

4.5.4. Determination Total Anthocyanin of *Paliurus spina Christi*

Our finding was observed that the orbital shaker in solvents (distilled water) and (ethanol: distilled) was given effective quantities of total anthocyanin content (OSE: 46.760 mg/kg) and (29.227 mg/kg) (p<0.01) respectively, compare and contrast with the methods (conventional and microwave) extraction in solvent (ethanol: distilled water) was no detected a quantity of total anthocyanin content of *Paliurus spina Christi*. On the other hand, the methods of microwave and conventional were observed significant values in solvents (MWE: 33.822 mg/kg) and (CE: 15.025 mg/kg) respectively.

Totally, the orbital shaker in solvents (distilled water) and (ethanol: distilled) was detected in strong quantities compare with another method extraction, in Table (4.40) and Figure (4.41).

Table 4.40. Total Anthocyanin content in *Paliurus spina Christi*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	15.025 $\pm$ 0.00	-	2327.663	0.000
MW/E	33.822 $\pm$ 0.00	-	7065.290	0.000
OS/E	46.760 $\pm$ 0.00	29.227 $\pm$ 0.00	1852.381	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

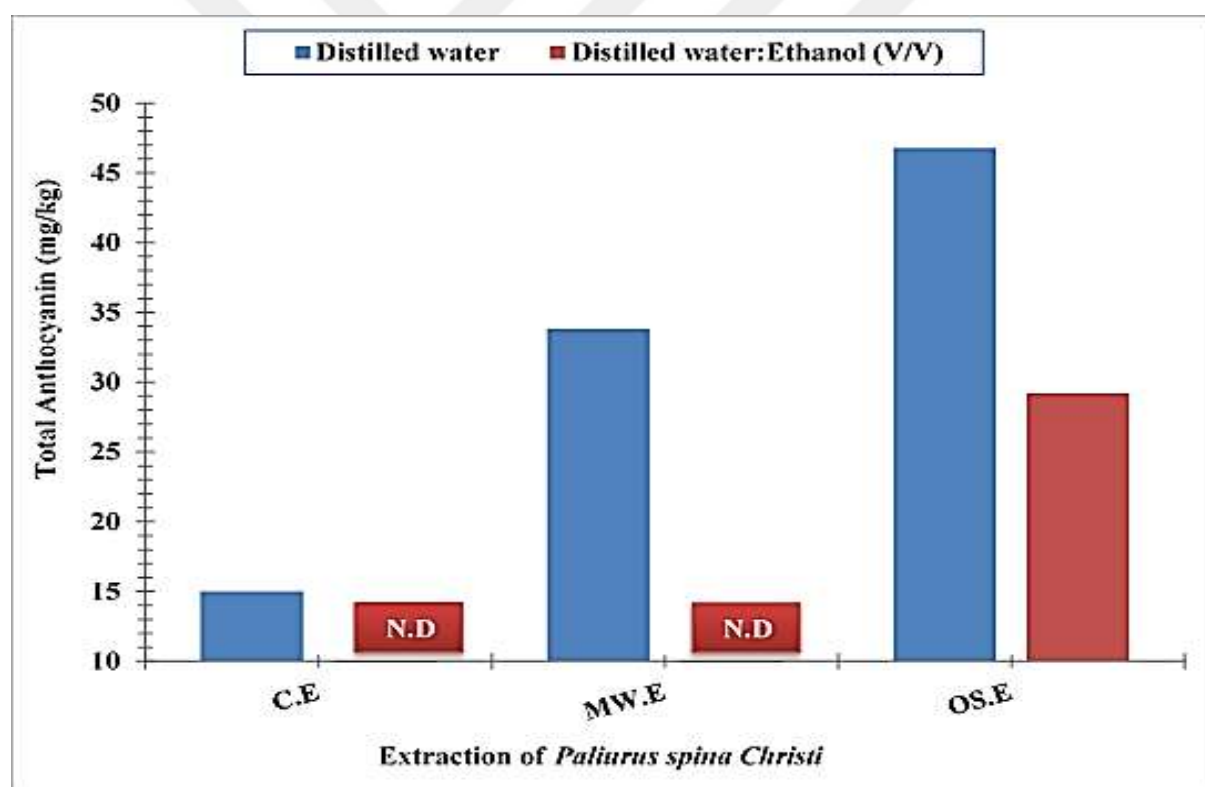


Figure 4.41. Total Anthocyanin content in *Paliurus spina Christi*

4.5.5. Determination Total Anthocyanin of *portulaca oleracea* herb

From the Table (4.41), and Figure (4.42). That the solvents of (distilled water) and (ethanol: distilled) were detected highest significant values (75.98 mg/kg) and (74.31 mg/kg) at ($p < 0.01$), by using the orbital shaker respectively, in comparison with the methods of (conventional extraction) in solvents of (distilled water) was detected quantity (CE: 62.62 mg/kg), whereas low quantity (1.67 mg/kg) was detected, by using (microwave extraction) of total anthocyanin content in *portulaca oleracea*. On the other hand, the (microwave and conventional extraction) was no detected values of total anthocyanin in solvent (ethanol: distilled water). Therefore, that the orbital shaker technique with the solvent of distilled water, was given great effective value of total anthocyanin in *portulaca oleracea* extract.

Table 4.41.Total Anthocyanin content in *portulaca oleracea*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	62.62 $\pm$ 0.00	-	130810.480	0.000
MW/E	1.671 $\pm$ 0.00	-	3491.127	0.000
OS/E	75.98 $\pm$ 0.00	74.31150 $\pm$ 0.00	1560.345	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

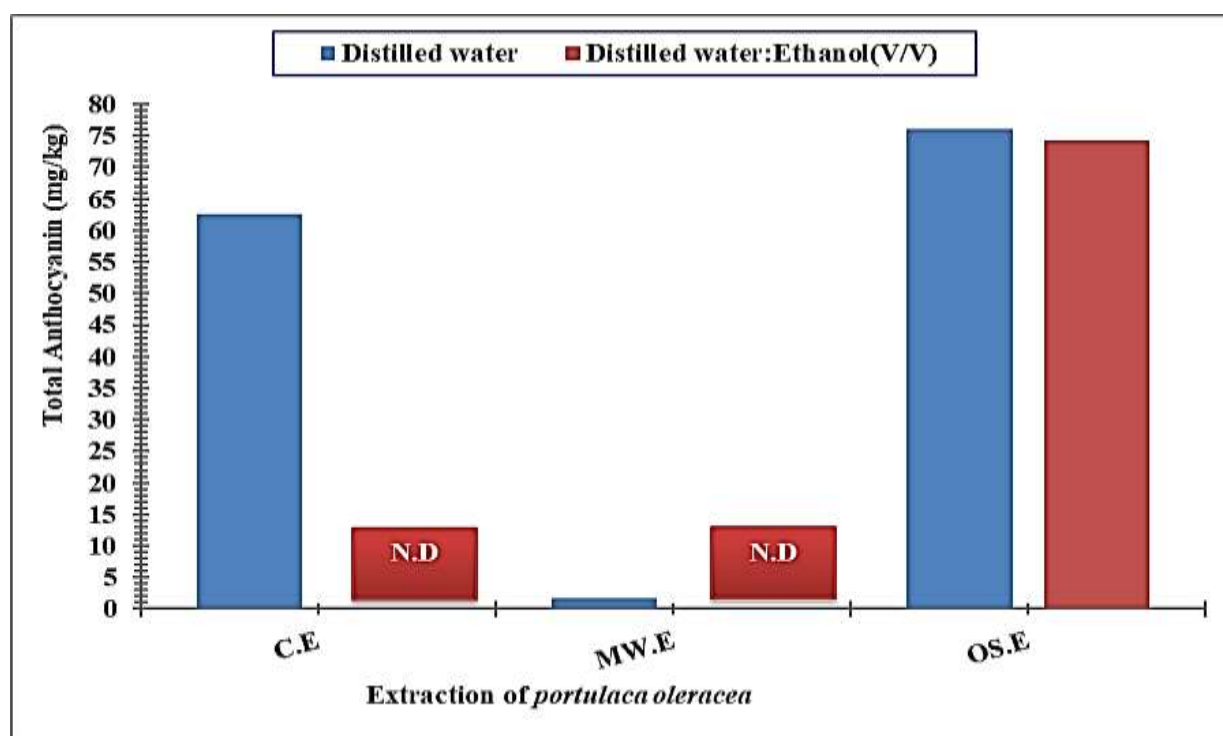


Figure 4.42.Total Anthocyanin content in *portulaca oleracea*

4.5.6. Determination Total Anthocyanin of *Artemisia haussknechtii* boiss

The total anthocyanin content was illustrated in Table (4.42), and Figure (4.43). Our results show that solvent of (distilled water) was detected strong quantity by using method orbital shaker (176.59 mg/kg). In comparison with the conventional method in solvents (distilled water), and (ethanol: distilled water) was observed low values of total anthocyanin content (53.84 mg/kg), and (32.56 mg/kg) respectively. On the other hand, the solvent of (ethanol: distilled water), using microwave technique (90.17 mg/kg) was shown highly significant value ($p < 0.01$), whereas, with solvent (distilled water) was not detected quantity. Consequently, the total anthocyanin content of *Artemisia haussknechtii* boiss herb, by orbital shaker was observed as the preferred effective amount with distilled water solvent.

Table 4.42.Total Anthocyanin content in *Artemisia haussknechtii* boiss

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	53.84 $\pm$ 0.00	32.56 $\pm$ 0.00	22234.069	0.000
MW/E	-	90.17 $\pm$ 0.00	-105598.341-	0.000
OS/E	176.59 $\pm$ 0.00	43.42 $\pm$ 0.00	211662.787	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

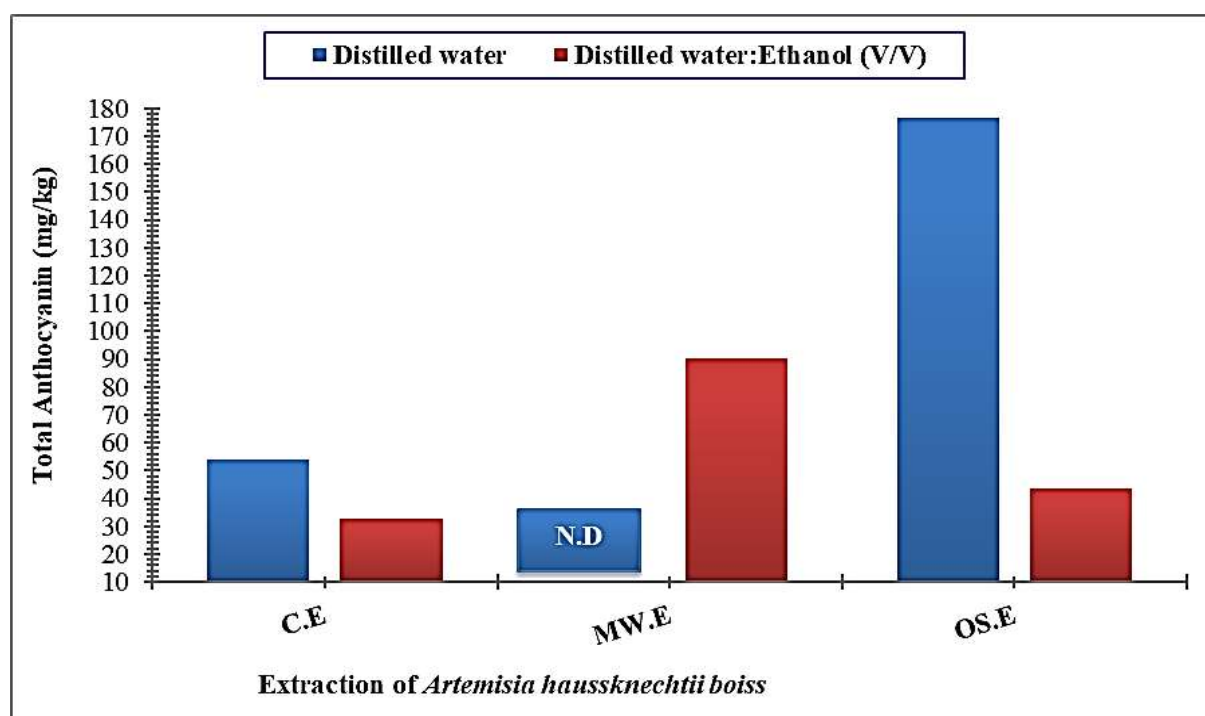


Figure 4.43.Total Anthocyanin content in *Artemisia haussknechtii* boiss

4.5.7. Determination Total Anthocyanin of *Prunus microcarpa*

In the current study, that the orbital shaker with solvent (distilled water) of *Prunus microcarpa* was detected a strong quantity of total anthocyanin content (32.15 mg/kg) ($p < 0.01$), thus, the method of microwave technique was observed as a significant value (13.36 mg/kg) in the same solvent, comparison with the solvent of (ethanol: distilled water) was not detected by using three methods extraction, illustrated in Table (4.43), and Figure (4.44).

Table 4.43.Total Anthocyanin content in *Artemisia Prunus microcarpa*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	-	-	-	-
MW/E	13.361 $\pm$ 0.00	-	20699.547	0.000
OS/E	32.152 $\pm$ 0.00	-	35220.751	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

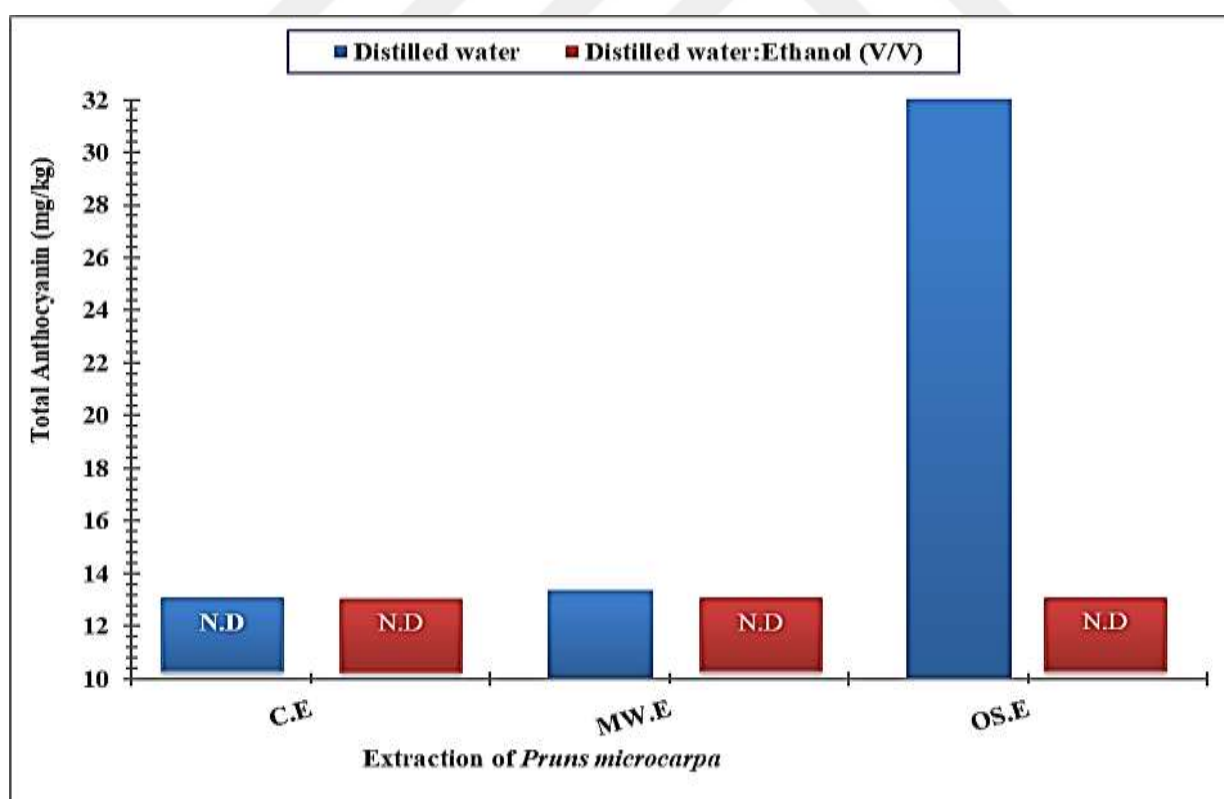


Figure 4.44.Total Anthocyanin content in *Prunus microcarpa*

4.5.8. Determination Total Anthocyanin of *Silybum Marianum*

The results of anthocyanin content of *Silybum Marianum* varied by solvents and techniques extractions, shown in (Table 4.44 and figure 4.45). The conventional techniques with solvents (E: DW=32.151 mg/kg), and (DW=20.87 mg/kg) was discovered higher significant values ($p < 0.01$). On other hand, the total anthocyanin content was detected in significant quantities with solvent (distilled water) by using an orbital shaker (30.89 mg/kg), and also was found low values in solvent (ethanol: distilled water) while using microwave extraction (5.01 mg/kg). Moreover, the solvent (distilled water) by MW/E and (ethanol: distilled water) by orbital shaker was not appeared the amount of anthocyanin content.

Table 4.44.Total Anthocyanin content in *Silybum Marianum*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	20.87200 $\pm$ 0.00091	32.15150 $\pm$ 0.00064	-10088.692-	0.000
MW/E	-	5.01175 $\pm$ 0.00085	-5869.161-	0.000
OS/E	30.89200 $\pm$ 0.00040	-	75669.637	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at ($p < 0.01$).

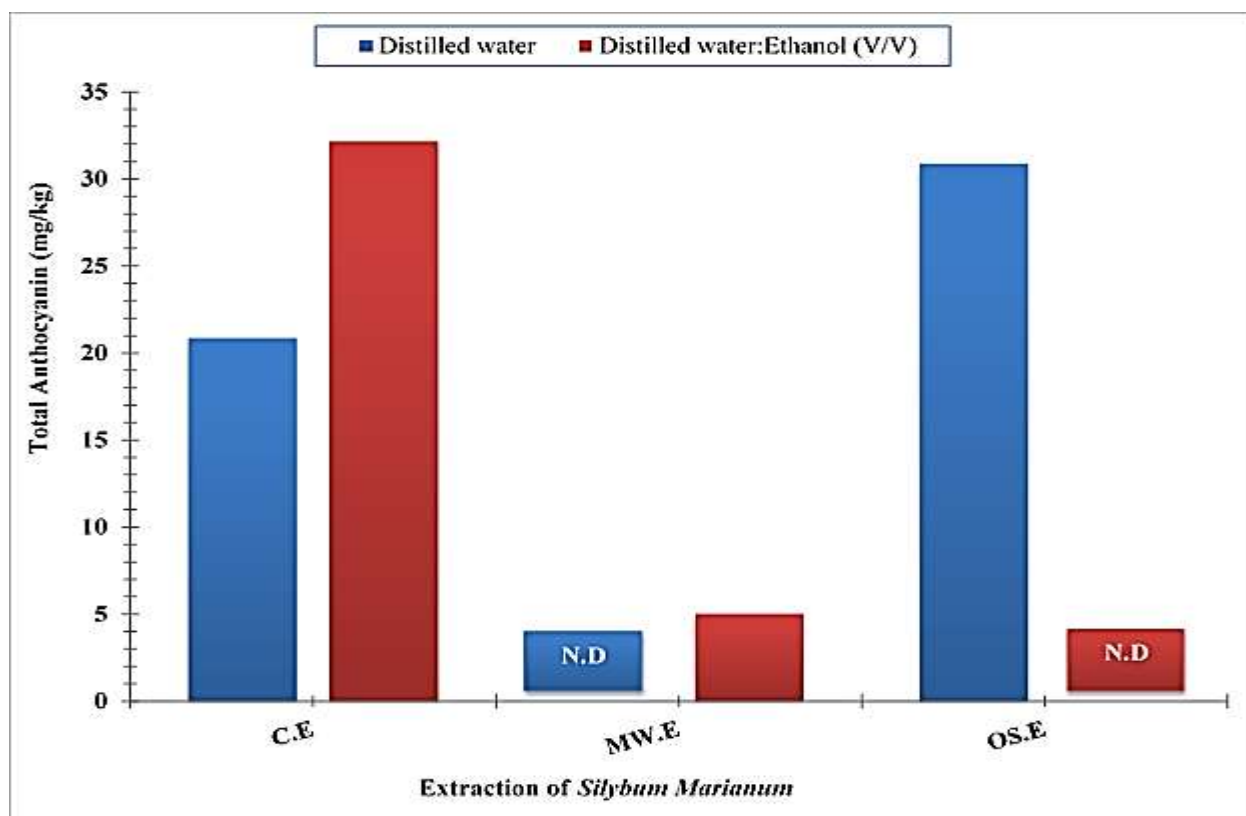


Figure 4.45. Total Anthocyanin content in *Silybum Marianum*

4.5.9. Determination Total Anthocyanin of *Pistacia eurycarpa Yale*

Our study from Table (4.45), and figure (4.46). The significant results in terms of total anthocyanin content were obtained from *pistacia eurycarpa yale* (MWE: 34.23 mg/kg) by using the microwave method with solvent (ethanol: distilled water), this study detected the highest significant values ($p < 0.01$). Thus, the content of anthocyanin in solvent (distilled water) by using the conventional technique was observed a significant quantity, but due to solvent ethanol: distilled water was not discovered. On the other hand, that the method of orbital shaker in solvent (DW) not detected the value, while solvent (E: DW 0.83 mg/kg) was given the lowest values of total anthocyanin. Additionally, the highest total anthocyanin content was found in *Pistacia eurycarpa Yale* extracts, a strong quantity by orbital shaker with solvent (ethanol: distilled water).

Table 4.45.Total Anthocyanin content in *Pistacia eurycarpa Yale*

Solvent	Distilled water	Distilled water: Ethanol	Statistic	
Method	Mean $\pm$ S.E	Mean $\pm$ S.E	T-test	P-value
C/ E	20.87 $\pm$ 0.00047	-	43597.575	0.000
MW/E	27.55 $\pm$ 0.00085	34.23 $\pm$ 0.00064	-6240.214-	0.000
OS/E	-	0.83050 $\pm$ 0.00028	-2876.936-	0.000

Not Detected: N.D; Notes/ Plants =P; Methods: C/E=Conventional Extraction; ME= Microwave Extraction; OS/E= Orbital shaker Extraction Solvent: W=Distilled water; DW: E=Ethanol: Distilled water; Values are Mean $\pm$ SE: standard Error, One-sample T-test significant at (p<0.01).

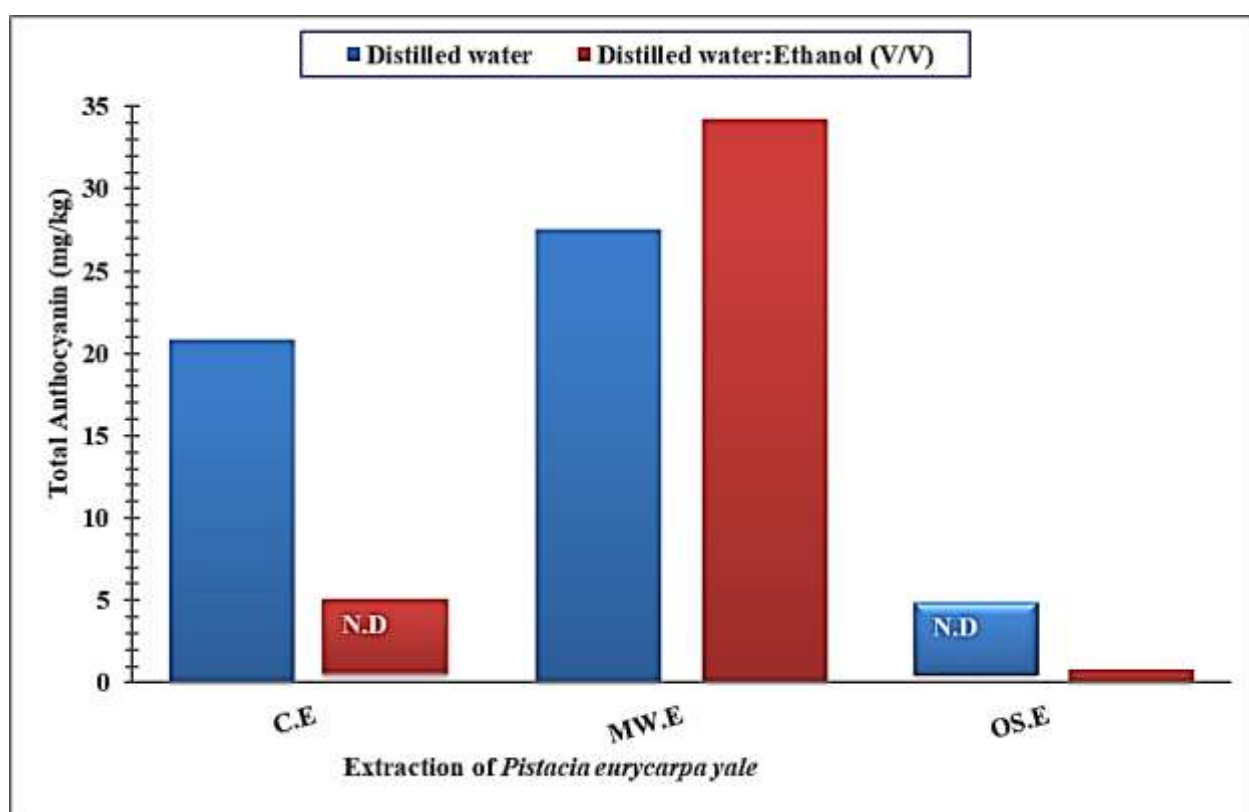


Figure 4.46.Total Anthocyanin content in *Pistacia eurycarpa Yale*

The anthocyanins are usually obtained by plant extraction and the techniques of extraction currently practiced are the use of solvents such as methanol, ethanol, acetone, water, or mixtures. However, structural changes with hydroxyl, methoxyl, glycosyl, and, in particular, acyl groups and environmental conditions such as temperature and light can quickly influence the stability of this anthocyanin (Vankar and Srivastava 2010). In another study, 96 % of ethanol solvent is used since the content of the water in the solvent is not so high to make the extraction more viscous. Methanol can extract anthocyanins 20 percent more efficiently than ethanol and 73 % more effective than water, but the use of methanol in large amounts is discouraged because it is not ideal for the health and the plant environment (Elaeocarpus *et al.* 2011)

In conclusion, the orbital shaker with solvent (distilled water) of total anthocyanin content in all plants was detected as a preferred powerful effective method for comparison with other techniques. However, the total anthocyanin content of the conventional method was not detected, due to the influence of the temperature of a conventional technique on the crude extracts. This study agrees with researchers (Niu *et al.* 2017; Liu *et al.* 2018), who reported the effect of high temperature and increased deterioration on the decrease of anthocyanin content in fruit and plants. Also, our study has shown, that relationship between the anthocyanin, flavonoid with total phenolic content is an Inverse relationship, that is approved with (Wang and Lin 2000) studied that anthocyanin and flavonoid increase, while the total phenolic content decrease. In general, our study is the first new experiment on the total anthocyanin content of our plants (Balaky *et al.* 2020)

4.6. Free Radical scavenging activity of DPPH Antioxidant

In this research, the DPPH assay has been widely used as a quick, reliable, efficient, reactive, accurate, simple, and reproducible parameter to search for the in vitro antioxidant activity of pure compounds as well as plant extracts (Ara and Nur 2009; Hassan 2011). Also, the DPPH is a monitor of chemical reactions involving radicals, and the method of free radical has been widely used for different natural and synthetic compounds antioxidant and free radicals scavenging potential (Sharma and Bhat 2009; Zhu *et al.* 2018). Furthermore, the role of an antioxidant is removing free radicals (Wang *et al.* 2008). On the other hand, The stable free radical 2, 2-Diphenyl-1-picrylhydrazyl (DPPH) has been widely used to assess the reducing substances or antioxidant activity of plant extracts and accepts hydrogen radical to transform a constant diamagnetic molecule, yellow-colored biphenyl picrylhydrazine (Öztürk *et al.* 2007; Dinis and Almeida 1997; Cotellet *et al.* 1996; SHIRWAIKAR *et al.* 2006). Researchers discovered, Plants' phenol content and antioxidant activity also increased at the same time (Andarwulan *et al.* 1999; Fathabad *et al.* 2015).

4.6.1. Assessment of antioxidant activities of *Eremurus Spectabilis Bieb*

It is known that determined *Eremurus Spectabilis Bieb*, extracts by three methods with the five solvents to have a significant antioxidant and free radical scavenging ability using DPPH as shown in Table (4.46) and Figure (6.10).

In this study, the ethanol and (hexane: ethanol) extracts had the highest antioxidant activity in concentration 0.1ml, 0.2ml, and 0.3ml values according to all methods (C/E, MW/E, and OS/E). Therefore, the antioxidant activity from ethanol (MWE: 82.65µg/mL) in 0.2ml concentration, (CE: 91.40µg/mL), (OSE: 92.14µg/mL), and the (hexane: ethanol) extracts (CE: 89.26µg/mL), (MWE: 94.12µg/mL) (OSE: 87.08 µg/mL), was significant highest values in concentration 0.3ml respectively, while it was observed that the lowest solvents of DW (CE: 34.41µg/mL),(MWE: 48.83 µg/mL), and (OSE: 30.23µg/mL), E: DW (CE: 24.32µg/mL),(MWE: 42.88µg/mL), and (OSE: 42.68µg/mL), were recorded. On the other hand, the hexane extracts with all (CE: 73.15 µg/mL) (MWE: 83.03µg/mL), and (OSE: 84.74µg/mL) respectively, showed a significant antioxidant activity. Actually, the (ethanol), and (hexane: ethanol) extracts was better than that of BHT in all concentration 0.1ml, 0.2ml, and 0.3ml that gives a great free radical scavenging activity of the *Eremurus Spectabilis Bieb*.

These findings agree with (Tosun, Ercisli, Ozer, Turan, *et al.* 2012), these results report from DPPH assay, the highest free-radical scavenging capacity was observed in a sample of *Eremurus Spectabilis Bieb* 86.66µg extract .ml⁻¹, that an important antioxidant and free radical scavenging activity. Also, Our study approval with (Murathan *et al.* 2018), that the DPPH of methanolic extracts (polarity index: 5.1) by Soxhlet apparatus where antioxidant activity was highest and lowest values indicated, that the *Eremurus Spectabilis Bieb* has beneficial impacts on DPPH radical scavenging capacity and may, thus protect against oxidative damage. Further, Our study doesn't agree with (Dervişoğlu *et al.* 2014) because the antioxidant capacity in water extracts obtained the high value of *Eremurus spectabilis*.

Generally, our finding was observed a great significant effect of DPPH scavenging activity compared to BHT standard, In all solvents especially of (ethanol), and (hexane: ethanol) with all techniques of *Eremurus Spectabilis Bieb*.

Table 4.46.DPPH free radical scavenging activity of *Eremurus Spectabilis Bieb* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	13.95	18.76	71.6	80.28	70.53
	0.2ml	16.04	24.12	83.06	86.49	73.15
	0.3ml	34.41	24.32	91.4	89.26	71.2
Trolox	0.1ml	271.39	294.64	255.44	179.7	351.78
	0.2ml	265.41	277.37	162.42	130.53	344.48
	0.3ml	212.92	276.7	94.651	108.6	323.21
R2		0.82	0.77	0.99	0.95	0.92
IC50		4.887586	11.93165	-1.2343	-5.8861	-14.503
MWE $\mu\text{g/mL}$	0.1ml	37.67	39.38	75.53	88.67	80.29
	0.2ml	41.62	35.87	83.14	92.03	81.87
	0.3ml	48.83	42.88	82.65	94.12	83.03
Trolox	0.1ml	203.62	228.2	223.55	113.25	245.48
	0.2ml	192.32	239.5	161.76	86.67	228.2
	0.3ml	171.72	216.90	165.74	70.06	215.57
R2		0.97	0.25	0.69	0.98	0.99
IC50		3.3056	8.0623	-6.5516	13.2617	-21.11
OSE $\mu\text{g/mL}$	0.1ml	24.18	28.04	81.91	76.17	81.5
	0.2ml	27.44	35.67	83.96	85.4	80.47
	0.3ml	30.23	42.68	92.14	87.08	84.74
Trolox	0.1ml	242.15	264.74	171.72	212.25	232.19
	0.2ml	232.85	240.16	155.11	139.17	243.48
	0.3ml	224.88	217.57	88.671	125.88	196.97
R2		0.99	0.99	0.89	0.86	0.52
IC50		9.512784	3.98861	-5.04305	-4.03303	-18.0186
BHT $\mu\text{g/mL}$	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.2. Assessment of antioxidant activities of *Prosopis fracta*

Depending on Table (4.47), and Figure (6.11), the conventional technique in (ethanol), and (hexane: ethanol) extracts of *Prosopis fracta* were highest and greatest significantly impact antioxidant activity in all concentrations of 0.1, 0.2, and 0.3ml. Especially in 0.3 ml concentration, observed maximum antioxidant activity at values (95.58 μ g/mL, and 93.20 μ g/mL) in compared with a minimum of BHT at values (92.30 μ g/mL, 91.94 μ g/mL) respectively. Whereas, the hexane extract showed an effective antioxidant activity value of 85.11 μ g/mL in 0.3ml of concentration. Reversely, the solvent of (distilled water) and (ethanol: distilled water) was obtained minimum values at various concentration 0.1, 0.2, and 0.3ml, free radical scavenging activity, showed less antioxidant activity than compared with the highest of BHT.

Thus, The DPPH free radical scavenging activity by microwave technique in different solvent extracts was significant values at 0.1, 0.2, 0.3 ml concentrations. therefore, the (hexane: ethanol) extracts, observed the highest and strongest scavenging activity at (95.63 μ g/mL) were performed at 0.3ml concentration, in comparison with the lowest of BHT at measure (91.94 μ g/mL), while the ethanol and hexane were obtained less effective an antioxidant activity in the concentration of 0.1, 0.2, 0.3 ml to compare with highest of BHT, oppositely, the ethanol: water and water showed the smallest antioxidant activity in all concentrations.

on the other hand, the orbital shaker technique in all solvents except for (distilled water) had obtained the highest and strong antioxidant activity in three concentrations especially in the concentration of 0.3ml at values (65.81 μ g/mL, 60.61 μ g/mL, 95.41 μ g/mL, 95.63 μ g/mL, and 95.85 μ g/mL) compared with the least of BHT control, at (52.98 μ g/mL, 92.30 μ g/mL, 91.94 μ g/mL, and 96.40 μ g/mL) successively.

Our finding in line with (Poudineh *et al.* 2015) reported, to obtain bioactive fractions, it may be necessary to selectively extract *Prosopis farcta* using appropriate solvents. Furthermore, the antioxidant effect of *Prosopis fracta* can be further utilized for the isolation of active components used in the treatment of many diseases induced by free radicals, the antioxidant effect of *Prosopis fracta* can be further exploited for the isolation of active components used in the treatment of many diseases induced by free radicals (Mohammed *et al.* 2018).

Commonly, the *Prosopis fracta* extracts by orbital shaker method in all solvents and conventional extraction with solvent (ethanol) and (hexane: ethanol), were given the highest and better antioxidant activities in comparison to BHT against free radical.

Table 4.47. DPPH free radical scavenging activity of *Prosopis fracta* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	mg/ml	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	76.92	7.44	7.83	84.2	93.7	77.3
	0.2ml	157.92	15.58	31.54	93.2	92.36	75.65
	0.3ml	157.89	31.16	35.25	95.58	93.2	85.11
Trolox	0.1ml	76.92	289.993	329.859	153.12	73.3888	278.033
	0.2ml	157.92	266.738	253.449	93.2079	84.0197	192.986
	0.3ml	157.89	222.221	241.49	60.7646	77.3754	295.973
R2			0.96	0.84	0.89	0.13	0.02
IC50			4.692243	3.832239	—	—	37.80488
MWE $\mu\text{g/mL}$	0.1ml	76.92	38.6	49.48	77.16	95.21	85.96
	0.2ml	157.92	45.11	62.06	77.74	96.3	84.56
	0.3ml	157.89	46.74	70.1	83.14	95.63	87.3
Trolox	0.1ml	76.92	200.959	195.644	210.261	61.4291	183.684
	0.2ml	157.92	182.355	155.114	205.61	52.7915	198.966
	0.3ml	157.89	177.7043	70.1031	161.75798	58.1069	169.06671
R2			0.89	0.98	0.82	0.14	0.23
IC50			3.5999312	0.976816	—	—	—
OSE $\mu\text{g/mL}$	0.1ml	76.92	9.76	60.2	94.51	88.17	84.74
	0.2ml	157.92	37.44	40.82	94.76	89.09	94.75
	0.3ml	157.89	65.81	60.61	95.41	95.63	95.85
Trolox	0.1ml	76.92	283.349	161.094	69.4022	117.24	196.97
	0.2ml	157.92	204.282	223.55	67.4089	109.93	88.996
	0.3ml	157.89	123.221	159.765	62.0935	58.107	76.047
R2			0.99	0.0003	0.93	0.84	0.26
IC50			2.439853	—	—	—	—
BHT $\mu\text{g/mL}$	0.1ml	76.92	78.37	38.96	88.05	79.94	91.03
	0.2ml	157.92	79.53	50.72	91.73	88.84	95.72
	0.3ml	157.89	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.3. Assessment of antioxidant activities of *Capparis spinosa*

According to results in Table (4.48), and Figure (6.12), the conventional technique in ethanol extract for *Capparis spinosa* showed the maximum value of antioxidant activity at (78.15µg/mL, 90.26µg/mL, and 93.04µg/mL), in 0.1, 0.2, and 0.3ml of concentrations, in compared with the least of BHT.

Furthermore, the highest value was hexane extracts in all three concentrations at measure (84.56µg/mL, 82.73µg/mL, and 82.48µg/mL) respectively, while the smallest values were obtained solvents of (distilled water, ethanol: distilled water, and hexane: ethanol) showed less antioxidant activity compared to standard of BHT. Hence, the (hexane: ethanol), (hexane), and (ethanol) extracts of the microwave method displayed the highest antioxidant activity at values (79.19µg/mL, 87.91µg/mL, and 86.91µg/mL), (83.22µg/mL, 88.77µg/mL, and 87.91), and (78.47µg/mL, 80.44µg/mL, and 84.53µg/mL) respectively, in concentrations of 0.1, 0.2, 0.3ml. Whereas, the (distilled water), and (ethanol: distilled water) extracts the antioxidant activity was fewer values compared with highest of efficient BHT.

Another side, observed that the antioxidant activities by orbital shaker extraction was effective values while compared with BHT standard from our solvent. The hexane extract was significant DPPH scavenging activity in concentrations of 0.1, 0.2, and 0.3ml at (82.73µg/mL, 85.35µg/mL, and 84.07µg/mL) whereas, the (hexane: ethanol), and (ethanol), observed the same near values of antioxidant activity but not correlate proportionally with (distilled water) and (ethanol: distilled water) extract, due to the lowest effective values, Compared with the highest efficiency of BHT.

Our study agrees with (Bonina *et al.* 2002; Rezzan *et al.* 2013) had reported in several in vitro tests, the *Capparis spinosa* extract exhibited strong and effective antioxidant /free radical scavenging in methanolic extract. Also, approval with (Allaith 2016; Arrar *et al.* 2013), reported that the *Capparis spinosa* in the methanolic extract was greatly antioxidant capacities, compared with total flavonoids, the total free phenolic, and total carotenoids.

In addition, the conventional technique of *Capparis spinosa*, extracts, showed strong antioxidant activity, in all solvents except (distilled water), and (ethanol: distilled water), while the (microwave and orbital shaker) techniques had obtained significant values to compare with BHT against free radical.

Table 4.48. DPPH free radical scavenging activity of *Prosopis fratta* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	3.95	7.62	78.15	66.86	84.56
	0.2ml	16.97	7.83	90.26	72.48	82.73
	0.3ml	36.27	16.28	93.04	76.25	82.48
Trolox	0.1ml	299.959	330.523	202.288	286.006	198.966
	0.2ml	262.751	329.859	103.953	241.49	218.899
	0.3ml	262.751	302.617	81.362	211.59	221.557
R2		0.75	0.76	0.88	0.98	0.83
IC50		7.737675	36.42424	–	–	34.27184
MWE $\mu\text{g/mL}$	0.1ml	48.37	35.05	78.47	79.19	83.22
	0.2ml	41.86	38.35	80.44	87.91	88.77
	0.3ml	53.02	52.16	84.53	86.91	87.91
Trolox	0.1ml	173.053	242.154	199.63	188.335	213.584
	0.2ml	191.657	187.006	183.684	119.234	153.12
	0.3ml	159.76469	231.52313	150.46267	127.20762	162.4224
R2		0.17	0.033	0.96	0.65	0.61
IC50		2.9665462	6.941497	–	–	–
OSE $\mu\text{g/mL}$	0.1ml	17.67	31.54	73.64	75.75	82.73
	0.2ml	23.72	37.52	78.15	74.24	85.35
	0.3ml	30	46.39	80.85	79.78	84.07
Trolox	0.1ml	3.95	7.62	78.15	66.86	84.56
	0.2ml	16.97	7.83	90.26	72.48	82.73
	0.3ml	36.27	16.28	93.04	76.25	82.48
R2		299.959	330.523	202.288	286.006	198.966
IC50		262.751	329.859	103.953	241.49	218.899
BHT $\mu\text{g/mL}$	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.4. Assessment of antioxidant activities of *Paliurus spina Christi*

Free radical scavenging ability results of *Paliurus spina Christi* extracts with five solvents by different methods in comparison to BHT are shown in Table (4.49), and Figure (6.13).

The results by the conventional method in all solvents were significant free radical scavenging ability in 0.1, 0.2, 0.3ml. The highest values of antioxidant activity were obtained in 0.3ml concentration at measure (80.36 μ g/mL) from the ethanol extract, hexane: ethanol (75.16 μ g/mL), and hexane (72.54 μ g/mL), while the (distilled water) and (ethanol: distilled water) was the nearest minimum of antioxidant, all solvents showed near and low antioxidant activity compared with BHT to resistance-free radical.

Consequently, all solvent extracts by microwave technique were got the strong efficient antioxidant activity in concentrations of 0.1, 0.2, and 0.3ml, except distilled water extract, which is showed weak antioxidant activity. in ethanol and hexane extracts showed the greatest antioxidant activity in DPPH assays with values of (95.97 μ g/mL), and (93.53 μ g/mL) respectively, in compared to smallest butylated hydroxytoluene (BHT), while the (E: DW), and (H: E) observed a significant antioxidant activity with values of (87.01 μ g/mL) and (86.91 μ g/mL), respectively. Compared with BHT against DPPH radicals.

The orbital shaker was given significant values of antioxidant activity with all solvents at concentrations 0.1, 0.2, and 0.3. Furthermore, the (ethanol) and (ethanol: distilled water) extracts in concentration 0.3ml, showed the biggest and more antioxidant activity in compared to BHT at values (92.63 μ g/mL), and (56.08 μ g/mL) respectively. Moreover, the (hexane: ethanol), and (hexane) extracts were significantly values of antioxidant activity at (89.93 μ g/mL), and (83.64 μ g/mL), while the distilled water extract observed the lowest antioxidant activity to compared butylated hydroxytoluene. Our investigator agrees with (Ali ŞEN 2018b), had studied, that the ethanol and ethyl acetate extracts from *Paliurus spina-christi* branches and leaves have strong antioxidant and anti-inflammatory capacity,

Commonly, the microwave way with all solvent extracts of this plant was more effective antioxidant activity than the orbital shaker extract even at three concentrations, in comparison with BHT to remove free radical.

Table 4.49. DPPH free radical scavenging activity of *Paliurus spina Christi* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	22.79	8.65	59	67.95	69.73
	0.2ml	40	10.92	75.2	75.08	71.75
	0.3ml	48.6	25.97	80.36	75.16	72.54
Trolox	0.1ml	246.141	327.201	357.765	277.369	360.423
	0.2ml	196.973	319.892	226.208	220.892	338.496
	0.3ml	172.389	271.389	184.349	220.228	329.859
R2		0.96	0.84	0.91	0.75	0.94
IC50		2.999225	6.02659	–	–	–
MWE $\mu\text{g/mL}$	0.1ml	14.41	62.68	82.81	76.93	88.77
	0.2ml	53.02	73.19	93.69	76.09	95.36
	0.3ml	54.18	87.01	93.53	86.91	95.97
Trolox	0.1ml	270.06	153.12	164.416	206.275	153.12
	0.2ml	159.765	119.234	76.0465	212.919	81.362
	0.3ml	156.44254	74.71765	77.37537	163.08684	74.71765
R2		0.77	0.99	0.73	0.63	0.81
IC50		2.47559847	0.003288	–	–	–
OSE $\mu\text{g/mL}$	0.1ml	10.69	38.76	84.61	76.17	82.18
	0.2ml	22.55	51.13	92.06	84.89	80.59
	0.3ml	27.44	56.08	92.63	89.93	83.64
Trolox	0.1ml	280.691	230.194	149.798	212.25	224.88
	0.2ml	246.805	190.328	89.3351	143.15	242.15
	0.3ml	232.852	174.382	84.6841	103.29	208.93
R2		0.94	0.94	0.8	0.97	0.0004
IC50		5.555548	2.15478	–	–	–
BHT $\mu\text{g/mL}$	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.5. Assessment of antioxidant activities of *portulaca oleracea* herb

The results were investigated in Table (4.50), and Figure (6.14). Among the different solvents by the conventional technique of *portulaca oleracea* herb, showed a significant percentage of scavenging activity in concentration 0.3ml, the (ethanol) and (hexane) extracts were obtained high efficiencies antioxidant activity ($84.28\mu\text{g/mL}$), and ($83.95\mu\text{g/mL}$), whereas the lowest of antioxidant activity with (DW $15.34\mu\text{g/mL}$, $40.20\mu\text{g/mL}$, and $68.79\mu\text{g/mL}$) extracts successively, in comparison with highest of BHT. Furthermore, the antioxidant potential of solvent extracts by microwave technique was evaluated at three concentrations of 0.1, 0.2, 0.3ml. Therefore, the (ethanol: distilled water) was extracts showed the strongest antioxidant activity of free radical scavenging characteristics against 1,1-Diphenyl-2-picrylhydrazyl (DPPH) than BHT. Also, the solvents of (E; $87.47\mu\text{g/mL}$, H; $83.64\mu\text{g/mL}$, and H/E; $82.38\mu\text{g/mL}$) extracts were more potent as scavengers' free radical than distilled water $49.30\mu\text{g/mL}$ extract in 0.3ml of concentration, showed the antioxidant activity smallest than butylated hydroxytoluene (BHT). However, the DPPH assay of the various extracts by orbital shaker at concentrations 0.1, 0.2, 0.3ml showed had a proficiency high antioxidant activity of $83.64\mu\text{g/mL}$, with (hexane: ethanol) extract, and $81.87\mu\text{g/mL}$ with (hexane) extract respectively, while extraction with (ethanol), and (ethanol: distilled water) observed a significant antioxidant activity at ($72.50\mu\text{g/mL}$), and ($70.72\mu\text{g/mL}$) respectively. Hence, the water extract was the smallest antioxidant activity in 0.3ml concentration, therefore, all extract solvents showed less effective antioxidant activity than the capacity of BHT to eliminate the free radical.

Our reports agree with those researchers (Sicari *et al.* 2018b; Uddin *et al.* 2014) studied, that the antioxidant activity of dried leaves of *portulaca oleracea*, with (ethanol) extract, was found to be higher than dried leaves with (methanol: water) extracts, and fresh leaves with (methanol: water) extract were also higher. Therefore, the highest antioxidant potential was found in dried leaves, which were used for solvent extraction. This result is partially similar to the findings (Hefnawy and Ali 2015) had reported, that the ethanolic of *portulaca oleracea* leaves extract antioxidant activities against DPPH and ABTS radicals at various concentrations (50, 100, 150, and 200 g ml⁻¹). Also, our study approval with (Alam *et al.* 2014; Youssef and Mokhtar 2014) indicated that the findings showed that ornamental *Portulaca oleracea* L was more source in antioxidant properties. Moreover, the methanol extract of the leaves and aerial part of *portulaca oleracea* displayed wide spectrum antioxidant, and antimicrobial adose-dependent anti-inflammatory properties (Agyare *et al.* 2015). Furthermore, the solvent of

ethanol extract of *portulaca oleracea* by methods of (microwave and orbital shaker) showed more effective antioxidant activity than (conventional technique) and with other solvents to compared with BHT to remove the free radical.

Table 4.50. DPPH free radical scavenging activity of *Portulaca oleracea* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration (µg/mL).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE µg/mL	0.1ml	11.86	8.04	67.51	61.66	81.94
	0.2ml	13.02	28.65	72.91	68.62	79.43
	0.3ml	15.34	40.2	84.28	68.79	83.95
Trolox	0.1ml	277.369	329.194	288.664	327.201	227.537
	0.2ml	274.047	262.751	244.812	272.053	254.778
	0.3ml	267.402	225.543	152.456	270.725	205.61
R2		0.96	0.97	0.95	0.76	0.19
IC50		23.03448	3.514925	–	–	–
MWE µg/mL	0.1ml	25.11	50.3	82.73	72.98	83.52
	0.2ml	33.95	69.48	85.43	78.52	82.42
	0.3ml	49.3	73.4	87.47	82.38	83.64
Trolox	0.1ml	239.496	192.986	167.08	237.503	210.261
	0.2ml	214.248	131.194	143.154	193.651	222.221
	0.3ml	170.39557	118.57003	126.54319	228.20098	208.93251
R2		0.97	0.87	0.99	0.04	0.0082
IC50		3.14744067	0.752988	–	–	–
OSE µg/mL	0.1ml	10.69	49.27	67.83	74.07	79.43
	0.2ml	14.65	70.72	70.04	85.31	79.19
	0.3ml	18.83	70.72	72.5	83.64	81.87
Trolox	0.1ml	280.691	196.308	286.006	228.87	228.2
	0.2ml	269.396	136.51	268.067	139.89	257.44
	0.3ml	257.436	127.208	248.134	153.12	227.44
R2		0.99	0.94	0.99	0.62	0.75
IC50		10.66659	–	–	–	–
BHT µg/mL	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.6. Assessment of antioxidant activities of *Artemisia haussknechtii* boiss

The *Artemisia haussknechtii* boiss herb extracts during three methods with different solvents are shown in Table (4.51), and Figure (6.15). It is observed that a maximum antioxidant activity at the concentrations 0.1, 0.2, and 0.3ml. (C/E: 93.94 μ g/mL, 91.27 μ g/mL), (MW/E: 95.90 μ g/mL, 94.21 μ g/mL), (OS/E: 95.74 μ g/mL, 92.70 μ g/mL) successively, showed more effective antioxidant activity compared BHT to remove free radical. While the hexane extract was observed antioxidant activity in all methods at (CE: 69.12), (MWE: 86.02), and (OSE: 80.29) successively, but this antioxidant activity less than BHT, therefore, the (ethanol: distilled water) extracts during conventional method more effective antioxidant activity at value (79.79 μ g/mL) compared than BHT at (52.98 μ g/mL) in 0.3ml of concentration. Whereas, the (ethanol: distilled water) extracts by (microwave and orbital shaker) techniques were obtained less antioxidant activity at (MWE: 51.75 μ g/mL), and (40.41 μ g/mL) compared with the highest of BHT standard. Thus, the distilled water extract with all methods, got a minimum antioxidant activity to compared BHT, against free radical.

In our study, indicate that the conventional technique more effective than the microwave technique, and the microwave technique more effective than the orbital shaker technique with all solvent except distilled water extract, respectively.

Consequently, the (ethanol), and (hexane: ethanol) showed great antioxidant activity compared with BHT. This finding agrees with (Khanahmadi *et al.* 2009) reported, that the antioxidant activity of the ethanolic extract of *Artemisia haussknechtii* was found to be very high, and it increased as the concentration of the plant extract increased. Moreover, In a DPPH free scavenging radical assay, the antioxidant activity of the polar sub-fraction of *Artemisia haussknechtii* was discovered to be higher than that of the others (Amiri and Goodarzi 2017), that approval with our study. Also, our study in line with (Carvalho *et al.* 2011) studied, that the antioxidant activities of *Artemisia* were found to be higher (reflecting a 2-fold difference) than that of other plants, according to DPPH assays. When compared to main commercially available leafy vegetables.

Table 4.51. DPPH free radical scavenging activity of *Artemisia haussknechtii boiss* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	17.67	31.54	79.95	88.42	66.68
	0.2ml	19.06	67.83	93.78	92.11	67.72
	0.3ml	20.46	79.79	93.94	91.27	69.12
Trolox	0.1ml	260.758	253.449	187.671	115.248	393.644
	0.2ml	256.771	136.51	75.3821	86.013	382.349
	0.3ml	252.785	97.9727	74.0532	92.6573	367.067
R2		1	0.92	0.75	0.54	0.99
IC50		24.26619	1.59743	–	–	–
MWE $\mu\text{g/mL}$	0.1ml	39.53	41.85	95.09	94.79	84.62
	0.2ml	41.39	42.06	95.9	94.96	85.41
	0.3ml	49.3	51.75	95.9	94.21	86.02
Trolox	0.1ml	198.302	220.228	64.7512	64.7512	183.02
	0.2ml	192.986	219.563	58.1069	63.4223	189.664
	0.3ml	170.39557	188.33518	58.1069	69.40221	183.01974
R2		0.88	0.76	0.75	0.54	0.99
IC50		3.34930483	2.965343	–	–	–
OSE $\mu\text{g/mL}$	0.1ml	22.79	29.89	89.11	80.2	79.43
	0.2ml	29.3	32.37	95.09	90.85	79.43
	0.3ml	34.65	40.41	95.74	92.7	80.29
Trolox	0.1ml	246.141	258.765	113.255	180.36	254.78
	0.2ml	227.537	250.792	64.7512	95.979	254.78
	0.3ml	212.255	224.879	59.4358	81.362	245.48
R2		0.99	0.91	0.82	0.85	0.66
IC50		5.555593	5.0001	–	–	–
BHT $\mu\text{g/mL}$	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.7. Assessment of antioxidant activities of *Prunus microcarpa*

The DPPH free radical scavenging activities were expressed as a percentage of inhibition and result from *Prunus microcarpa* extracts by three methods with five solvents In three different concentrations 0.1, 0.2, and 0.3ml are shown in Table (4.52), and Figure (6.16). Further, the conventional extraction with all solvents extract as the range of (DW 13.02 μ g/mL), (E: DW 19.38 μ g/mL), (E 66.53 μ g/mL), (H: E 57.55 μ g/mL), and (H 69.85 μ g/mL) respectively, in 0.3 of concentration, showed less effective antioxidant activity in compared BHT standard.

Hence, the microwave technique with (ethanol: distilled water) extracts was maximum antioxidant activity at value (64.53 μ g/mL) in 0.3 ml of concentration to compared lowest of BHT at values (52.98 μ g/mL) to remove free radical, while the solvents of (ethanol), (hexane: ethanol), and (hexane) were given high antioxidant activity at values (85.76 μ g/mL), (79.44 μ g/mL), and (84.62 μ g/mL) successively, but showed less antioxidant activity compared with BHT. On the other hand, the water extract showed minimum antioxidant activity.

On the other hand, all solvents by the orbital shaker technique exhibited high antioxidant activity at values (W: E 48.04 μ g/mL), (E 86.25 μ g/mL), (H: E 75.08 μ g/mL), and (H 80.53 μ g/mL) respectively, except the water extract, showed less antioxidant activity at value (13.48 μ g/mL), all the solvents by orbital shaker extracts observed less antioxidant activity compared with the standard of BHT against DPPH radical. Our results are don't comparable to this study (Liu *et al.* 2014), Due to the different technologies work and different species of plant with the same family. Also agrees with (Belhadj *et al.* 2016), who studied, that the variety *Prunus* species extract displayed the highest antioxidant activity compared to the other samples. The GC and GC–MS were used to analyze the n-hexane extract of *Prunus domestica* shoots. It has been effective in scavenging DPPH radicals as an antioxidant (Mahmood *et al.* 2009).

Our study indicated that the *Prunus microcarpa* extracts of microwave technique with solvent (ethanol: water), gave better antioxidant activity than BHT to prevent create of DPPH free radicals.

Table 4.52. DPPH free radical scavenging activity of *Prunus microcarpa* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration (µg/mL).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE µg/mL	0.1ml	7.9	11.95	58.75	55.03	64.73
	0.2ml	13.95	16.08	61.37	56.79	65.46
	0.3ml	13.02	19.38	66.53	57.55	69.85
Trolox	0.1ml	288.664	316.57	359.758	379.691	414.906
	0.2ml	271.389	303.282	338.496	365.738	406.933
	0.3ml	274.047	292.651	296.637	359.758	359.094
R2		0.61	0.005	0.96	0.94	0.85
IC50		17.0549	11.75741	—	—	—
MWE µg/mL	0.1ml	39.06	55.67	81.09	76.84	82.48
	0.2ml	58.37	54.63	84.94	78.52	83.03
	0.3ml	60.69	64.53	85.76	79.44	84.62
Trolox	0.1ml	199.63	175.711	178.369	193.651	221.557
	0.2ml	144.483	179.033	147.141	193.651	215.577
	0.3ml	137.8385	147.14052	140.49622	186.34189	198.30163
R2		0.82	0.66	0.87	0.97	0.92
IC50		10.4272326	0.131739	—	—	—
OSE µg/mL	0.1ml	24.88	32.37	77.74	69.547	77.97
	0.2ml	34.41	46.18	77.82	74.83	80.84
	0.3ml	13.48	48.04	86.25	75.08	80.53
Trolox	0.1ml	272.718	250.792	205.61	264.74	270.72
	0.2ml	212.919	206.275	204.946	222.89	239.5
	0.3ml	240.161	200.295	136.51	220.89	242.82
R2		0.29	0.83	0.75	0.78	0.69
IC50		6.517016	2.99562	-5.19882	-6.38043	-21.2656
BHT µg/mL	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.8. Assessment of antioxidant activities of *Silybum Marianum*

The DPPH scavenging activity of *Silybum Marianum* extracts by different techniques with various solvents in the concentrations 0.1, 0.2, and 0.3ml, are shown in Table (4.53), and Figure (6.17). The conventional method in (ethanol: distilled water), and (ethanol) extracts have been observed maximum effectiveness of the antioxidant activity at values (E: DW 87.62 $\mu\text{g/mL}$), and (E 95.17 $\mu\text{g/mL}$) in 0.3ml of concentration, compared with the lowest of BHT at values (52.98 $\mu\text{g/mL}$), and (92.30 $\mu\text{g/mL}$) successively, while the (hexane: ethanol), and (hexane) extracts, showed significant values of antioxidant activity at (H: E 84.31 $\mu\text{g/mL}$), and (H 71.20 $\mu\text{g/mL}$), successively, the distilled water extract showed low antioxidant activity at (25.81 $\mu\text{g/mL}$), to comparison with highest BHT against free radical.

Furthermore, the microwave method with solvent (ethanol: distilled water), (ethanol), and (hexane: ethanol) extracts obtained a strong efficiency antioxidant activity at measures (E: DW 91.95 $\mu\text{g/mL}$), (E 96.07 $\mu\text{g/mL}$), and (H: E 95.21 $\mu\text{g/mL}$) in concentration 0.3ml, in compared with of BHT standard against free radical, while the significant values of antioxidant activity were observed in hexane and distilled water extracts at (84.99 $\mu\text{g/mL}$), (66.74 $\mu\text{g/mL}$) in concentration 0.3ml, compared with the maximum of BHT.

Furthermore, the orbital shaker technique with solvent (ethanol: distilled water), (ethanol), extracts, gave a great ability antioxidant activity at values (E: DW 89.89 $\mu\text{g/mL}$), and (E 95.58 $\mu\text{g/mL}$), in concentration 0.3ml, showed more antioxidant activity compared Butyl hydroxytoluene (BHT) to remove free radical. Whereas, the (hexane: ethanol), and (hexane) extracts, obtained a significant antioxidant activity at values (76.25 $\mu\text{g/mL}$), and (82.67 $\mu\text{g/mL}$) respectively, in comparison with the highest values of BHT. Therefore, the distilled water extract displayed the antioxidant activity lower than BHT at a value (27.90 $\mu\text{g/mL}$), in 0.3ml concentration.

Our study higher than this report (Sun et al. 2016) That the ethanol extract of *Silybum Marianum* had the maximum 1, 1-Diphenyl-2-picryl-hydrazyl (DPPH) radical scavenging activity (69.68%), accompanied by the roots (66.02%), indicating strong scavenging activity as important natural antioxidants. Also agrees with (Anthony and Saleh 2012) had reported, that the antioxidant activity was calculated by estimating their antioxidant potential as Trolox equivalent and measuring radical scavenging activity with DPPH. Our results were suggested, that the microwave technique of *Silybum Marianum* in solvents (W: E, E, H: E, and H) was the

high antioxidant activity and better than the BHT standard against free radical, showed more antioxidant activity compared with the (conventional and orbital shaker) techniques.

Table 4.53. DPPH free radical scavenging activity of *Silybum Marianum* extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	6.27	69.48	88.87	67.11	67.9
	0.2ml	22.79	85.56	90.09	78.43	68.09
	0.3ml	25.81	87.62	95.17	84.31	71.2
Trolox	0.1ml	293.315	131.194	115.248	284.013	380.355
	0.2ml	246.141	79.3687	105.281	194.315	378.362
	0.3ml	237.503	72.7244	64.0868	147.805	344.476
R2		0.86	0.83	0.88	0.96	0.79
IC50		5.25	—	—	—	—
MWE $\mu\text{g/mL}$	0.1ml	44.65	90.3	94.76	85.06	82.24
	0.2ml	62.09	91.75	95.49	88.84	83.83
	0.3ml	66.74	91.95	96.07	95.21	84.99
Trolox	0.1ml	183.684	64.0868	56.778	141.825	224.214
	0.2ml	133.852	59.4358	61.4291	111.926	206.939
	0.3ml	120.56332	58.77133	67.40892	61.42905	206.93922
R2		0.89	0.84	0.99	0.97	0.99
IC50		1.29121028	—	—	—	—
OSE $\mu\text{g/mL}$	0.1ml	19.06	65.15	88.54	71.14	80.23
	0.2ml	26.27	86.39	94.43	74.32	80.04
	0.3ml	27.9	89.89	95.58	76.25	82.67
Trolox	0.1ml	256.771	145.147	117.906	252.12	219.56
	0.2ml	236.174	65.4156	70.0666	226.87	248.13
	0.3ml	231.523	65.4156	60.7646	211.59	219.56
R2		0.88	0.85	0.86	0.98	0.75
IC50		7.789571	—	—	—	—
BHT $\mu\text{g/mL}$	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

4.6.9. Assessment of antioxidant activities of *Pistacia eurycarpa* Yale

As shown in Table (4.54), and Figure (6.18). The antioxidant activity of *Pistacia eurycarpa* Yale extracts was measured using different assays: (C/E, MW/E, and OS/E) with five solvents at 0.1, 0.2, and 0.3ml of concentrations. The results indicated that the (ethanol), (ethanol: distilled water), and (hexane) extracts, showed maximum antioxidant capacity at values (96.07 μ g/mL), (74.43 μ g/mL), and (97.55 μ g/mL), respectively, in 0.3ml of concentration, better than BHT to prevent create of free radical. While the (distilled water), and (hexane: ethanol) extracts obtained a significant antioxidant activity at values (73.48 μ g/mL), and (74.43 μ g/mL) but less than BHT standard.

Therefore, the results of the microwave technique exhibited an antioxidant activity better and biggest values in (ethanol: distilled water), (ethanol), and (hexane: ethanol) extract at (93.19 μ g/mL), (97.70 μ g/mL), and (90.35 μ g/mL) with 0.3ml of concentration, the results showed more antioxidant activity of free radical scavenging against (DPPH) compared with BHT, to prevent the free radical. Whereas, the water and hexane extracts have remarkably significant antioxidant activity compared with BHT in concentration 0.3ml at values (80.93 μ g/mL), and (85.90 μ g/mL) respectively.

Also, the highest free radical scavenging activity of DPPH was found in all solvents during the orbital shaker method, in concentration 0.1, 0.2, and 0.3ml, the highest antioxidant activity (88.86 μ g/mL), (95.82 μ g/mL), and (96.22 μ g/mL) was found in (ethanol: distilled water), (ethanol), and (hexane: ethanol) extracts in 0.3ml concentration, this results showed antioxidant activity better than with BHT at values (52.98 μ g/mL), (92.30 μ g/mL), and (91.94 μ g/mL) to against free radical. Hence, the antioxidant activity of water and hexane extracts are generally significant values at (72.32 μ g/mL), and (87.55 μ g/mL) while, lower than BHT at (81.86 μ g/mL), and (96.40 μ g/mL) respectively, in 0.3 of concentration.

Our results are dissimilar with (Amel *et al.* 2016), who had studied, that the *Pistacia lentiscus* L, methanol extract exhibited the lowest values of antioxidant activity and lowest activities to scavenge DPPH radical. Also, current study in line with (Hatamnia *et al.* 2014), showed, that the highest antioxidant activity of hull species *pistacia* extract can be related to its higher total phenolic content. According to our findings, the methods of (CE, MWE, and OSE) with all solvents of *Pistacia eurycarpa* Yale, extracts gave the highest antioxidant activity compared with BHT to remove the DPPH free radical

Table 4.54. DPPH free radical scavenging activity of *Pistacia eurycarpa* Yale extracts, compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).

Methods	Con.	SCV (W)	SCV (W/E)	SCV (E)	SCV (H/E)	SCV (H)
CE $\mu\text{g/mL}$	0.1ml	26.04	24.32	88.87	57.71	88.95
	0.2ml	50	49.89	92.22	57.21	96.58
	0.3ml	73.48	74.43	96.07	65.01	97.55
Trolox	0.1ml	236.839	276.704	115.248	358.429	151.127
	0.2ml	168.4	194.315	88.0063	362.416	57.4425
	0.3ml	101.29	115.248	56.778	300.624	68.0734
R2		1	0.99	0.99	0.69	0.83
IC50		2.209106	–	–	–	–
MWE $\mu\text{g/mL}$	0.1ml	73.72	92.57	96.8	81.29	83.83
	0.2ml	76.04	93.81	97.05	88.5	85.11
	0.3ml	80.93	93.19	97.7	90.35	85.9
Trolox	0.1ml	100.63	56.778	50.7982	171.724	192.986
	0.2ml	93.9861	52.7915	48.8049	114.583	192.986
	0.3ml	80.03309	54.78475	43.48944	99.96599	184.3486
R2		0.95	0.25	0.93	0.89	0.75
IC50		–	–	–	–	–
OSE $\mu\text{g/mL}$	0.1ml	52.79	83.91	95.99	95.3	82.79
	0.2ml	64.88	87.21	95.33	95.72	83.64
	0.3ml	72.32	88.86	95.82	96.22	87.55
Trolox	0.1ml	160.429	84.6841	57.4425	60.765	208.93
	0.2ml	125.879	74.0532	62.7579	57.442	208.93
	0.3ml	104.617	68.7378	58.7713	53.456	166.41
R2		0.98	0.96	0.057	0.99	0.75
IC50		0.634969	–	565.5556	–	–
BHT $\mu\text{g/mL}$	0.1ml	78.37	38.96	88.05	79.94	91.03
	0.2ml	79.53	50.72	91.73	88.84	95.72
	0.3ml	81.86	52.98	92.3	91.94	96.4

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW), Distilled water: Ethanol (DW: E), Ethanol (E), Hexane: Ethanol (H: E) and Hexane (H); BHT: Butylated Hydroxy Toluene; IC50: Inhibitory concentration 50

Our study indicates that all plants obtained a high antioxidant activity in all different solvents during the (microwave, conventional, and orbital shaker). Especially, the (ethanol), and (hexane: ethanol) extracts. this study in agreement (Balaky *et al.* 2020) had reported, that the ethanol extraction ability higher than distilled water during Microwave-Assisted Extraction of DPPH free radical activity of plants. On the other hand, ethanol is known as a good solvent to extract phenol and be safe for human consumption (Dai and Mumper 2010).

Therefore, the extraction techniques, solvent type, and polarity, plant parts selected for extraction mediate antioxidant and free radical scavenging capacity (Chekroun-Bechlaghem *et al.* 2019). During times of stress, free radicals and reactive metabolites are formed. Many diseases, especially cancer, are linked to oxidative stress. Furthermore, there is a connection between the occurrence of chronic diseases and the level of oxidative stress. (Sosa *et al.* 2013). Antioxidants are created as a part of the protection mechanism of higher organisms and inhibit oxidative damage associated with diseases (Adebayo *et al.* 2012; Astley 2003; Ahmad *et al.* 2012).

As a general result of the current study, it has been concluded that our plants have an extremely higher level of antioxidant activity depending upon the methods and solvents. This finding approval to (Katalinić *et al.* 2004) had reported, that fruits, herbs, and grains, are contain a broad range of antioxidant components, including phenolic compounds. Also, in our study, the antioxidant ability of these compounds is strongly associated. Though, some of our plants seem to have limited amounts than the artificial antioxidants as BHT standard on solvents

Furthermore, artificial antioxidants including butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), propyl gallate (Pg), and tertbutyl hydroquinone (TBHQ) were commonly used as food and beverage additives for a long time. Since they are related to elevated levels of cytotoxicity and carcinogenic effects, their use is now limited. As a result, there is a major need to discover natural compounds with antioxidant properties and low toxicity (Gouveia *et al.* 2013). Although, our natural antioxidant activity had great ability to remove and stable free radical and better than BHT, which can be used in several food and health applications. Therefore, antioxidants play a critical role in human health protection (Lambert and Yang 2003).

However, the concentration of antioxidants needed to decrease the initial DPPH radical's concentration by 50%. The inhibition of concentration 50 (IC₅₀) is a parameter widely used to measure antioxidant activity. A decrease or comparatively IC₅₀ value corresponds with

a higher antioxidant power or shows more antioxidant potential. Therefore, the relationship between antioxidants activity and IC₅₀ is negative and reverse associations. This result is in agreement with reports (Concepción *et al.* 1998; Kumawat *et al.* 2012; Djenidi *et al.* 2020).

The antioxidant activities varied largely among all the plants used in this study. Indeed, the highest antioxidant activity, with the smallest IC₅₀ (-4.30 mg/mL) value for 1,1-diphenyl-2-picrylhydrazyl studies. Additionally, the superiority of all plants was more pronounced as compared to positive controls. Therefore, antioxidant activity and Trolox were shown to have strong and positive associations, with correlation coefficients ranging from ($R \geq 0.99$), and ($R \geq 1$). Too, this study in line with (Chaouche *et al.* 2014).

Finally, our results indicate that the *Pistacia eurycarpa yale*, *Prosopis frakta*, and *Paliurus Spina Christi* were obtained the highest amounts of antioxidant activity. Especially, the *Pistacia eurycarpa yale* giving maximum (80.93 µg/mL, 93.19 µg/mL, 97.70 µg/mL, 90.35 µg/mL, and 85.90 µg/mL) in all solvents, that better and more than an antioxidant activity to compare with BHT standard, IC₅₀, and Trolox standard.

4.7. Identification and Quantification of specific Phenolic Compounds of our plants extracts by LC-MS/MS

In the current study, the Analytical parameter by LC-MS/MS was dependent on Twenty-five standards types of specific phenolic compounds that have been using with our plants by three techniques with five solvent extracts. Further, that used to analyze by a mass spectrometer (LC-MS/MS) equipped with the triple-quadruple analyses, because of its sensitivity and selectivity for compound precise phenolic and flavonoids, negative ionization mode used to be desired in the present-day research. The specific phenolic compounds have been recognized following their retention and quantified by their respective trendy calibration curves. Many researchers in the literature on LC-MS/MS-based quantitative analysis (Ertas *et al.* 2014; Ertas *et al.* 2015; Cavaliere *et al.* 2005)

4.7.1. Identification and quantification of analysis specific phenolic compounds of *Eremurus Spectabilis Bieb* extract by LC-MS/MS

Among all identified specific phenolic compounds from 25 standard kinds of phenolic, was only detected twelve phenolics by LC-MS/MS as (acetohydroxamic acid, vanillic acid, resveratrol, fumaric acid, gallic acid, caffeic acid, hydroxycinnamic acid, silymarin, curmin, Salicylic acid, hydroxybenzoic acid, and luteolin) from *Eremurus Spectabilis Bieb*, in solvent of distilled water with the techniques of (conventional extraction, microwave extraction, and orbital shaker extraction) individually, as shown in Table (4.55).

Therefore, the nine kinds of phenolic were obtained in **distilled water** extract at conventional techniques, it contained the highest amount of vanillic acid (CE: 2219.132 µg/g), while the distilled water was showed significant of hydroxycinnamic acid (346.966 µg/g), and resveratrol (180.932 µg/g) respectively, and with a minimum amount of phenolic as acetohydroxamic acid, fumaric acid, gallic acid, caffeic acid, salicylic acid, and hydroxybenzoic acid) respectively. Also, the distilled water at microwave technique obtained ten kinds of phenolic compound, it showed a maximum of hydroxycinnamic acid (MWE: 1465.097 µg/g) while, the vanillic acid (MWE: 820.150 µg/g), resveratrol (MWE: 191.826 µg/g), and luteolin (MWE: 113.938 µg/g) respectively, with smallest of phenolic as (fumaric acid, caffeic acid, silymarin, curmin, and hydroxybenzoic acid). On the other hand, nine kinds of phenolic standards were displayed at the orbital shaker method in distilled water. The highest amount of vanillic acid was (OSE: 1001.372 µg/g), the significant amounts were fumaric acid (OSE: 136.696 µg/g), luteolin (OSE: 200.078 µg/g), and resveratrol (OSE: 167.375 µg/g)

respectively. Whereas, has been obtained a minimum amount of (acetohydroxamic acid, gallic acid, caffeic acid, hydroxycinnamic acid, and hydroxybenzoic acid).

Hence, the **(ethanol: distilled water extract v/v)** was given ten kinds of phenolic compounds at all three methods. According to study results, vanillic acid (CE: 789.360 $\mu\text{g/g}$), (MWE 776.319 $\mu\text{g/g}$), and (OSE: 355.54 $\mu\text{g/g}$), and the hydroxycinnamic acid (CE: 485.548 $\mu\text{g/g}$), (MWE 725.205 $\mu\text{g/g}$), and (OSE: 131.550 $\mu\text{g/g}$), show very high quantities among all methods. While the other kinds of phenolic compounds at different methods were showed significant amounts of resveratrol (CE: 39.744 $\mu\text{g/g}$), (MWE: 47.838 $\mu\text{g/g}$), (OSE: 74.794 $\mu\text{g/g}$), fumaric acid (CE: 60.478 $\mu\text{g/g}$), (MWE: 75.247 $\mu\text{g/g}$), and (OSE: 76.684 $\mu\text{g/g}$), gallic acid (CE: 47.745 $\mu\text{g/g}$), caffeic acid (CE: 17.765 $\mu\text{g/g}$), (MWE: 22.780 $\mu\text{g/g}$), and (OSE: 8.338 $\mu\text{g/g}$), silymarin (CE: 77.944 $\mu\text{g/g}$), (OSE: 80.390 $\mu\text{g/g}$) respectively. Otherwise, the (luteolin, acetohydroxamic acid, and hydroxybenzoic acid) showed the smallest quantities of phenolic compounds, especially the hydroxybenzoic acid at (CE: 3.965 $\mu\text{g/g}$), and (3.353 $\mu\text{g/g}$) respectively.

Also, the solvent of **ethanol** extract was detected twelve types of phenolic at conventional technique compounds, eight types of phenolic compounds at microwave technique, and thirteen compounds of phenolic, respectively. The hydroxycinnamic acid, vanillic acid, resveratrol, fumaric acid, and caffeic acid was detected as the main highest phenolic with conventional technique at amounts (3118.368 $\mu\text{g/g}$), (825.264 $\mu\text{g/g}$), (350.313 $\mu\text{g/g}$), (114.619 $\mu\text{g/g}$), and (114.922 $\mu\text{g/g}$) respectively, while the silymarin, quercetin, Phloridzin dihydrate, gallic acid, and catechin hydrate showed sensitive amounts (CE: 69.134 $\mu\text{g/g}$), (CE: 55.148 $\mu\text{g/g}$), (CE: 48.174 $\mu\text{g/g}$), (CE: 44.835 $\mu\text{g/g}$), and (CE: 23.893 $\mu\text{g/g}$) respectively. But, the acetohydroxamic acid, and hydroxybenzoic acid there were given the littlest quantities (CE: 7.166 $\mu\text{g/g}$), (CE: 7.113 $\mu\text{g/g}$) respectively. Furthermore, the microwave technique was obtaining maximum amounts of hydroxycinnamic acid, vanillic acid at (MWE: 754.478 $\mu\text{g/g}$), and (MWE: 494.623 $\mu\text{g/g}$). whereas, the resveratrol (MWE: 87.892 $\mu\text{g/g}$), gallic acid (MWE: 46.391 $\mu\text{g/g}$), fumaric acid (MWE: 42.226 $\mu\text{g/g}$), and caffeic acid (MWE: 21.035) were found to be the abundant amount, and, the acetohydroxamic acid (MWE: 3.416 $\mu\text{g/g}$), hydroxybenzoic acid (MWE: 5.222 $\mu\text{g/g}$) few abundant amounts the extract method, but each of the catechin hydrate, quercetin, Phloridzin dihydrate, silymarin) weren't detected at microwave method. The orbital shake method gave the maximum phenolic compounds of hydroxycinnamic acid (OSE: 2055.213 $\mu\text{g/g}$), and with the high amounts of resveratrol (OSE: 303.684), vanillic acid (OSE: 287.561 $\mu\text{g/g}$), ellagic acid (OSE: 267.526 $\mu\text{g/g}$), silymarin (OSE:

142.829 $\mu\text{g/g}$) respectively. While the fumaric acid (OSE: 74.801 $\mu\text{g/g}$), caffeic acid (OSE: 66.010 $\mu\text{g/g}$), Phloridzin dihydrate (OSE: 51.492), quercetin (OSE: 50.470), and gallic acid (OSE: 45.727) were obtained the most abundant amounts of phenolic with least abundant amounts of salicylic acid (OSE: 3.849 $\mu\text{g/g}$), acetohydroxamic acid, (OSE: 10.596 $\mu\text{g/g}$), and hydroxybenzoic acid (OSE: 13.633 $\mu\text{g/g}$) respectively.

According to the results were showed the conventional technique fifteen kinds of phenolic compounds, the microwave technique seven kinds of phenolic, and the orbital shaker eight kinds of compounds phenolic, in (**hexane: ethanol**) extracts. The biggest compound of phenolic was observed in conventional technique as hydroxycinnamic acid (CE: 4860.090 $\mu\text{g/g}$), and with the highest amounts of vanillic acid (CE: 756.776), resveratrol (CE: 677.986 $\mu\text{g/g}$), ellagic acid (CE: 264.876 $\mu\text{g/g}$), luteolin (CE: 222.968 $\mu\text{g/g}$), silymarin (CE: 218.978 $\mu\text{g/g}$), and quercetin (CE: 136.745 $\mu\text{g/g}$) respectively. Whereas it has been containing a least of compounds hydroxybenzoic acid (CE: 3.981 $\mu\text{g/g}$), butein 4.041 $\mu\text{g/g}$), salicylic acid (CE: 4.829 $\mu\text{g/g}$), and acetohydroxamic acid (CE: 16.910 $\mu\text{g/g}$). Also, it gives a significant value of fumaric acid (CE: 59.693 $\mu\text{g/g}$), Phloridzin dihydrate (CE: 45.944 $\mu\text{g/g}$), and gallic acid (CE: 44.653 $\mu\text{g/g}$) respectively.

Additional, the microwave method was giving the highest amount of vanillic acid (MWE: 796.163 $\mu\text{g/g}$), resveratrol (MWE: 107.552 $\mu\text{g/g}$), while the compounds of luteolin (MWE: 68.904 $\mu\text{g/g}$, fumaric acid (MWE: 58.487 $\mu\text{g/g}$), hydroxycinnamic acid (MWE: 25.382 $\mu\text{g/g}$), and caffeic acid (MWE: 15.664 $\mu\text{g/g}$), have been a plentiful amounts and hydroxybenzoic acid (MWE: 8.236 $\mu\text{g/g}$) least plentiful amount of plant extracts. Instead, that the microwave in (hexane: ethanol) extract was no detected the catechin hydrate, quercetin, acetohydroxamic acid, gallic acid, Phloridzin dihydrate, salicylic acid, and butein.

However, the orbital shaker was obtained a largest phenolic as hydroxycinnamic acid (OSE: 2023.298 $\mu\text{g/g}$), among all compounds, and with high amounts of vanillic acid (OSE: 770.078 $\mu\text{g/g}$), and resveratrol (OSE: 348.320 $\mu\text{g/g}$), respectively. While the fumaric acid (OSE: 79.131 $\mu\text{g/g}$), silymarin (OSE: 74.801 $\mu\text{g/g}$), caffeic acid (OSE: 54.901 $\mu\text{g/g}$), and catechin hydrate (OSE: 30.522 $\mu\text{g/g}$) showed the effective quantity of phenolic compounds, also the acetohydroxamic acid and hydroxybenzoic acid have been the least amount (OSE: 8.181 $\mu\text{g/g}$), and (OSE: 5.092 $\mu\text{g/g}$), conversely, it has been no detected any compounds as quercetin, gallic acid, Phloridzin dihydrate, ellagic acid, butein, and luteolin.

Lastly, the **hexane** extract showed twelve types of compounds of phenolic with all three methods. It contained high quantities of vanillic acid (CE: 893.619 $\mu\text{g/g}$), resveratrol (CE: 311.142 $\mu\text{g/g}$), and hydroxycinnamic acid (CE: 267.353 $\mu\text{g/g}$). Whereas, it recorded the low amounts of fumaric acid (CE: 61.582 $\mu\text{g/g}$), gallic acid (CE: 46.385 $\mu\text{g/g}$), curmin (CE: 45.395 $\mu\text{g/g}$), caffeic acid (CE: 13.848 $\mu\text{g/g}$), hydroxybenzoic acid (CE: 3.981 $\mu\text{g/g}$) at conventional technique.

The microwave method with hexane extract was showed a maximum amount of hydroxycinnamic acid at (MWE: 3766.054 $\mu\text{g/g}$). Also, showed high amounts of vanillic acid (MWE: 773.442 $\mu\text{g/g}$), resveratrol (MWE: 427.428 $\mu\text{g/g}$), caffeic acid (MWE: 103.874) respectively. While it has been obtained effective amounts of fumaric acid (MWE: 72.560 $\mu\text{g/g}$), silymarin (MWE: 68.520 $\mu\text{g/g}$), quercetin (MWE: 48.101 $\mu\text{g/g}$), gallic acid (MWE: 47.454 $\mu\text{g/g}$), and Phloridzin dihydrate (MWE: 46.052 $\mu\text{g/g}$) respectively. Though, it was exhibited the smallest amounts of acetohydroxamic acid (MWE: 14.284 $\mu\text{g/g}$), hydroxybenzoic acid (MWE: 11.106 $\mu\text{g/g}$), and butein (MWE: 2.310 $\mu\text{g/g}$) respectively.

On the Other hand, the high amounts of phenolic compounds detected in hexane extract at orbital shaker method it was vanillic acid (OSE: 839.529 $\mu\text{g/g}$), resveratrol (OSE: 136.602 $\mu\text{g/g}$), hydroxycinnamic acid (OSE: 100.992 $\mu\text{g/g}$), and fumaric acid (OSE: 54.956 $\mu\text{g/g}$) respectively. While the acetohydroxamic acid and hydroxybenzoic acid were low amounts (OSE: 3.004 $\mu\text{g/g}$), and (OSE: 2.037 $\mu\text{g/g}$) respectively, Therefore, the phenolic compounds of quercetin, gallic acid, caffeic acid, Phloridzin dihydrate, and silymarin were not detected in hexane extract at orbital shaker technique.

There is no research on *Eremurus Spectabilis Bieb* of phytochemical analysis. Our study indicated that the *Eremurus Spectabilis Bieb* was containing high specific phenolic compounds. Especially, vanillic acid in water extracts higher than other solvents at all three methods.

Table 4.55. Identification and quantification of analysis specific phenolic compounds of **Five solvents** with three methods of *Eremurus Spectabilis* Bieb extract by LC -MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	-	-	-	-	-	-	23.893	-	-	-	-	30.522	-	-	-
2	-	-	-	-	-	-	55.148	-	50.470	136.745	-	-	-	48.101	-
3	2.627	9.768	2.791	3.965	-	3.353	7.166	3.416	10.596	16.910	-	8.181	-	14.284	3.004
4	2219.132	820.150	1001.372	789.260	776.319	355.54	825.264	754.478	287.561	756.776	796.163	770.078	893.619	773.442	839.529
5	180.932	191.826	136.696	39.744	47.838	74.794	350.313	87.892	303.684	677.986	107.552	348.320	311.142	427.428	136.602
6	59.693	88.211	200.078	60.478	75.247	76.684	114.619	42.226	74.801	59.693	58.487	79.131	61.582	72.560	54.956
7	42.481	-	42.632	47.745	-	-	44.835	46.391	45.727	44.653	-	-	46.385	47.454	-
8	14.197	39.887	15.494	17.765	22.780	8.338	114.922	21.035	66.010	176.245	15.664	54.901	13.848	103.874	-
9	-	-	-	-	-	-	48.174	-	51.492	45.944	-	-	-	46.052	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	346.966	1465.097	0.669	485.548	725.205	131.550	3118.368	494.623	2055.213	4860.090	25.382	2023.298	267.353	3766.054	100.992
12	-	-	-	-	-	-	-	-	267.526	264.876	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	-	68.376	-	77.944	-	80.390	69.134	-	142.829	218.978	-	74.801	-	68.520	-
15	-	44.870	-	-	-	-	-	-	-	-	-	-	45.395	-	-
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
18	0.449	-	-	-	-	-	-	-	3.849	4.829	-	-	-	-	-
19	5.151	6.372	6.695	4.992	4.952	6.213	7.113	5.222	13.633	3.981	8.236	5.092	3.981	11.106	2.037
20	-	-	-	-	-	-	-	-	-	4.041	-	-	-	2.310	-
21	-	113.938	167.375	-	-	13.308	-	-	-	222.968	68.904	-	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1.4 aftokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.2. Identification and quantification of analysis specific phenolic compounds of *Prosopis farcta* extract by LC-MS/MS

The determination of the phenolic compounds in the *Prosopis farcta* were detected thirteen phenolics from 25 kinds by the LC-MS/MS analysis, in five dissimilar solvents with three different methods, exhibited in Table (4.56). It was determined that the **distilled water** extract had a high content of vanillic acid amounts (CE: 756.254 $\mu\text{g/g}$, MWE: 706.480 $\mu\text{g/g}$, OSE: 756.877 $\mu\text{g/g}$) at all three methods. Further, the fumaric acid, caffeic acid, hydroxycinnamic acid, and Phloridzin dihydrate detected an effective amount at (conventional and microwave) techniques, and no data detected in an orbital shaker at (CE: 104.018 $\mu\text{g/g}$, MWE: 111.165 $\mu\text{g/g}$, OSE: N.D),(CE: 11.463 $\mu\text{g/g}$, MWE: 5.593 $\mu\text{g/g}$, OSE: N.D), (CE: 31.253 $\mu\text{g/g}$, MWE: 128.239 $\mu\text{g/g}$, OSE: N.D), and (CE: 76.816 $\mu\text{g/g}$, MWE: 106.778 $\mu\text{g/g}$, OSE: N.D) respectively, while the distilled water extract was given the smallest amount of acetohydroxamic acid at microwave technique (MWE: 1,242 $\mu\text{g/g}$), and the hydroxybenzoic acid at all three methods. Hence, it was detected high of quercetin at the orbital shaker technique (OSE: 132.104 $\mu\text{g/g}$).

On the other side, the quantified result in (**ethanol: distilled water**) extracts obtained thirteen types of phenolic compounds. The vanillic acid, gallic acid, catechin hydrate, Phloridzin dihydrate, and hydroxycinnamic acid was containing high amounts of phenolic at (CE:975.413 $\mu\text{g/g}$, MWE: 755.090 $\mu\text{g/g}$, OSE: 346.816 $\mu\text{g/g}$), (CE: 431.489 $\mu\text{g/g}$, MWE: 228.533 $\mu\text{g/g}$, OSE: 500.301 $\mu\text{g/g}$), (CE: 292.830 $\mu\text{g/g}$, MWE: 208.032 $\mu\text{g/g}$, OSE: 428.832 $\mu\text{g/g}$), (CE: 172.778 $\mu\text{g/g}$, MWE: 132.857, OSE: 354.271 $\mu\text{g/g}$), and (CE: 149.999 $\mu\text{g/g}$, MWE: 46.700 $\mu\text{g/g}$, OSE: 105.771 $\mu\text{g/g}$). Whereas the resveratrol, fumaric acid was obtained effective amounts of phenolic compounds in all three methods at (CE: 47.361 $\mu\text{g/g}$, MWE: 44.031 $\mu\text{g/g}$, and OSE: 82.570 $\mu\text{g/g}$), and (CE: 72.587 $\mu\text{g/g}$, MWE: 58.237 $\mu\text{g/g}$, and OSE: 68.494 $\mu\text{g/g}$) respectively, also the minimum amounts of their were salicylic acid and hydroxybenzoic acid in all three methods. However, no data detected of quercetin in conventional extract, except at the orbital shaker technique, while the acetohydroxamic acid, caffeic acid, silymarin were found in (microwave and orbital shaker) techniques.

Also, considered the results in **ethanol** extract identified fifteen kinds of phenolic compounds at all three techniques. In general, it was a very rich phenolic content because of its high level of luteolin amount at conventional technique (CE: 2686.674 $\mu\text{g/g}$). Furthermore, the catechin hydrate (CE: 265.090 $\mu\text{g/g}$, MWE: 129.616 $\mu\text{g/g}$, OSE: 211.691 $\mu\text{g/g}$), vanillic acid (CE: 933.044 $\mu\text{g/g}$, MWE: 854.015 $\mu\text{g/g}$, OSE: 797.275 $\mu\text{g/g}$), gallic acid (CE: 879.453 $\mu\text{g/g}$,

MWE: 240.065 $\mu\text{g/g}$, OSE: 531.495 $\mu\text{g/g}$), and Phloridzin dihydrate (CE: 519.326 $\mu\text{g/g}$, MWE: 109.203 $\mu\text{g/g}$, OSE: 586.812 $\mu\text{g/g}$) respectively, were found to be the most abundant compounds in the ethanol extract. Also, all three methods detected an effective amount of phenolic compounds, resveratrol, fumaric acid, hydroxycinnamic acid, and hydroxybenzoic acid, while, some other phenolic of ellagic acid, kaempferol, silymarin, quercetin, and caffeic acid, was shown at specific values with conventional technique at (419.997 $\mu\text{g/g}$, 303.563 $\mu\text{g/g}$, 110.012 $\mu\text{g/g}$, 103.767 $\mu\text{g/g}$, and 7.701 $\mu\text{g/g}$), respectively. Though, the orbital shaker observed the quercetin, acetohydroxamic acid, silymarin, salicylic acid, and curmin in microwave technique.

The (**hexane: ethanol**) extract showed fifteen phenolic compounds in all techniques. It gave the highest amounts of phenolic compounds with (conventional and microwave) methods of vanillic acid, gallic acid, and Phloridzin dihydrate, and hydroxycinnamic acid (CE: 844.662, MWE: 861.847 $\mu\text{g/g}$), (CE: 811.513 $\mu\text{g/g}$, MWE: 1009.884 $\mu\text{g/g}$), (CE: 536.357 $\mu\text{g/g}$, MWE: 579.168 $\mu\text{g/g}$), and (CE: 687.706 $\mu\text{g/g}$, MWE: 733.445 $\mu\text{g/g}$), respectively. Also, both conventional and microwave extraction were displayed as effective quantities of catechin hydrate, quercetin, resveratrol, fumaric acid, and silymarin (CE: 25.439 $\mu\text{g/g}$, MWE: 26.978 $\mu\text{g/g}$), (CE: 61.433 $\mu\text{g/g}$, MWE: 129.840 $\mu\text{g/g}$), (CE: 52.089, MWE: 82.658), (CE: 50.990 $\mu\text{g/g}$, MWE: 73.418 $\mu\text{g/g}$), and (CE: 168.571 $\mu\text{g/g}$, MWE: 110.285 $\mu\text{g/g}$) respectively.

Furthermore, the hydroxycinnamic acid and hydroxybenzoic acid detected a strong abundant amount at the orbital shaker method (OSE: 309238 $\mu\text{g/g}$), and (OSE: 29782 $\mu\text{g/g}$). On the other hand, the high and low amounts at orbital shaker technique was containing vanillic acid, gallic acid, catechin hydrate, quercetin, resveratrol, fumaric acid, and myricetin, at (OSE: 909.761 $\mu\text{g/g}$, OSE: 762.788 $\mu\text{g/g}$, OSE: 208.075 $\mu\text{g/g}$, OSE: 126.298 $\mu\text{g/g}$, OSE: 66.963 $\mu\text{g/g}$, OSE: 64.628 $\mu\text{g/g}$, OSE: 60.133 $\mu\text{g/g}$) respectively. The acetohydroxamic acid, caffeic acid, and salicylic acid were showed the smallest amount in all three methods.

The results indicated that all treatments of **hexane** extract of *Prosopis farcta* by using three ways were found ten phenolic compounds, the results of vanillic acid (CE: 844.811 $\mu\text{g/g}$), (MWE: 922.373 $\mu\text{g/g}$), and (OSE: 758.403 $\mu\text{g/g}$) by all three methods was the maximum quantities and catechin hydrate, resveratrol, and gallic acid (CE: 38.378 $\mu\text{g/g}$, MWE: 27.613 $\mu\text{g/g}$, and OSE: 85.233 $\mu\text{g/g}$), (CE: 47.971 $\mu\text{g/g}$, MWE: 48.370 $\mu\text{g/g}$, and OSE: 55.339 $\mu\text{g/g}$), and (CE: 124.667 $\mu\text{g/g}$, MWE: 65.493 $\mu\text{g/g}$, and OSE: 161.404 $\mu\text{g/g}$) a limited amounts of abundant compounds in all extract methods.

On the other hand, the ellagic acid detected a high quantity of phenolic compounds by conventional extraction as (CE: 252.207 $\mu\text{g/g}$). Whereas, the Phloridzin dihydrate and fumaric acid were detected from the methods of (conventional and orbital shaker) extraction as (CE: 49.070 $\mu\text{g/g}$, OSE: 130.607 $\mu\text{g/g}$), and (CE: 64.623 $\mu\text{g/g}$, OSE: 47.997 $\mu\text{g/g}$) respectively. However, the hydroxybenzoic acid was found the smallest amount from all three techniques and found an effective and little amounts of curmin and caffeic acid by microwave extraction.

According to our results, the vanillic acid was displayed of *Prosopis farcta* maximum amount in ethanol extract at all methods. Moreover, there is no study on *Prosopis farcta* phytochemical analysis. But there are different studies regarding the phenolic and flavonoid components in the *Prosopis farcta* beans, which were determined flavonoid investigation identified by HPLC. (Aydin *et al.* 2014).

Table 4.56. Identification and quantification of analysis specific phenolic compounds of five solvents with three methods of *Prosopis farcta* extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	57.009	75,853	103,349	292.830	208.032	428.832	265.090	129.616	211.691	38.378	27.613	85.233	25.439	26.978	208.075
2	-	-	132,104	-	-	54.916	103.767	-	70.953	-	-	-	61.433	129.840	126.298
3	-	1,242	-	-	0.478	1.845	-	-	2.846	-	3.532	2.725	3.119	2.533	1.590
4	756,254	706,480	756,877	975.413	755.090	346.816	933.044	854.015	797.275	844.811	922.373	758.403	844.662	861.847	909.761
5	36.621	55.383	90.430	47.361	44.031	82.570	79.368	47.731	71.062	47.971	48.370	55.339	52.089	82.658	66.963
6	104.018	111.165	-	72.587	58.237	68.494	61.622	58.305	44.904	64.623	-	47.997	50.990	73.418	64.628
7	262.025	429.187	77.875	431.489	228.533	500.301	879.453	240.065	531.495	124.667	65.493	161.404	811.513	1009.884	762.788
8	11.463	5.593	-	-	5.036	5.515	7.701	-	-	-	8.671	-	12.227	18.855	5.651
9	76.816	106.778	-	172.778	132.857	354.271	519.326	109.203	586.812	49.070	-	130.607	536.357	579.168	370.338
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	31.253	128.239	-	149.999	46.700	105.771	496.559	65.286	66.536	-	-	-	687.706	733.445	309238
12	-	-	-	-	-	-	419.997	-	-	252.207	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	26.525	60.133
14	-	-	-	-	77.917	-	110.012	-	72.646	-	-	-	168.571	110.285	-
15	44.870	-	-	-	-	-	-	44.870	-	-	45.402	-	-	-	-
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	303.563	-	-	-	-	-	-	-	-
18	-	0.307	-	7.362	2.608	17.531	-	-	5.317	-	-	-	4.315	26.039	13.517
19	11.981	7.335	7.175	11.061	9.586	23.056	9.573	9.815	17.426	4.141	6.371	8.620	14.758	31.633	29782
20	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
21	-	-	-	-	-	-	2686.674	-	-	-	-	-	464.611	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1.4 aftokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.3. Identification and quantification of analysis specific phenolic compounds of *Capparis Spinosa* fruit extract by LC-MS/MS

In this study, the quantification and determination of phenolic acids in five solvents with all three different methods of *Capparis Spinosa* were displayed by LC-MS/MS. As shown in Table (4.57). The twelve phenolic compounds in **distilled water** extract were catechin hydrate, quercetin, acetohydroxamic acid, vanillic acid, resveratrol, fumaric acid, gallic acid, Phloridzin dihydrate, silymarin, curmin, salicylic acid, and hydroxybenzoic acid at all three methods. The results showed that vanillic acid highest value among all methods with concentration (CE: 807.148 µg/g, MWE: 825,918 µg/g, and OSE: 849,534 µg/g), while the resveratrol, gallic acid, and Phloridzin dihydrate were showed the significant concentrations as (CE:47.517 µg/g, MWE: 42.792 µg/g, OSE: 31.814 µg/g) in all methods, (MWE: 49.640 µg/g, OSE: 44.072 µg/g), and (MWE: 47.544 µg/g, OSE: 45.944 µg/g) at microwave and orbital shaker techniques. Thus, the hydroxybenzoic acid was a smallest concentration (CE: 15.550 µg/g, MWE: 8.465 µg/g, OSE: 10.836 µg/g) at all methods. Also, the significant amounts of catechin hydrate, and curmin (CE: 65.339 µg/g, MWE: 27.346 µg/g), and (CE: 46.451 µg/g, MWE: 45.395 µg/g) at methods of conventional and microwave extraction, respectively. On the other hand, all three methods identified fumaric acid and fumaric acid in the distilled water extract.

The results showed ten phenolic acids in (**ethanol: distilled water**) extract with all techniques. That quercetin displayed a maximum amount in the orbital shaker method (OSE: 2224.114 µg/g). Also, the vanillic acid and fumaric acid highest amount among all specific phenolic in all methods (CE: 775.700 µg/g, MWE: 838.925 µg/g, OSE: 826.338 µg/g), and (CE: 191.398 µg/g, MWE: 183.839 µg/g, OSE: 329.042 µg/g), while the hydroxybenzoic acid gave the lowest quantity (CE: 5.517 µg/g, MWE: 10.217 µg/g, OSE: 9.245 µg/g). On the other hand, the high amounts of silymarin catechin hydrate, and resveratrol found in microwave technique as (MWE: 180.475 µg/g, MWE: 156.590 µg/g, MWE: 117.983 µg/g,) was extra than the amounts of other compounds as catechin hydrate (CE: 62.554 µg/g), resveratrol (CE: 66.830 µg/g), gallic acid (CE: 45.438 µg/g), and curmin (OSE: 45.402 µg/g) whereas the hydroxybenzoic acid was the smallest amount in all methods.

From table (4.57), the fourteen phenolic compounds were obtained with all three methods in **ethanol** extract. The highest quantity and the significant amount of phenolic compounds was vanillic acid (CE: 827.950 µg/g, MWE: 824.154 µg/g, OSE: 778.510 µg/g), fumaric acid CE: 339.115 µg/g, MWE: 491.319 µg/g, OSE: 125.327 µg/g), and catechin hydrate

(CE: 42.756 $\mu\text{g/g}$, MWE: 69.559 $\mu\text{g/g}$, OSE: 24.859 $\mu\text{g/g}$, also with obtained smallest amounts of salicylic acid (CE: 4.494 $\mu\text{g/g}$, MWE: 8.395 $\mu\text{g/g}$, OSE: 6.653 $\mu\text{g/g}$) respectively, at all three-technique. On the other hand, a significant amount was observed of resveratrol, gallic acid, and silymarin with (conventional and microwave) methods as (CE: 43.632 $\mu\text{g/g}$, MWE: 47.828 $\mu\text{g/g}$), (CE: 45.432 $\mu\text{g/g}$, MWE: 46.267 $\mu\text{g/g}$), and (CE: 69.010 $\mu\text{g/g}$, MWE: 62.769 $\mu\text{g/g}$) respectively. Further, the resveratrol and silymarin were containing high amounts of phenolic compounds by the orbital shaker method, whereas the acetohydroxamic acid and butein detected a least of phenolic acid in the microwave and orbital shaker methods.

The (**hexane: ethanol**) extract by using three methods were found 14 specific phenolic compounds. It included maximum quantities of vanillic acid and fumaric acid (CE: 778.964 $\mu\text{g/g}$, MWE: 853.513 $\mu\text{g/g}$, OSE: 731.643 $\mu\text{g/g}$) and, (CE: 245.192 $\mu\text{g/g}$, MWE: 370.343 $\mu\text{g/g}$, OSE: 251.206 $\mu\text{g/g}$). Besides, that the significant amounts of resveratrol and gallic acid (CE: 71.681 $\mu\text{g/g}$, MWE: 55.655 $\mu\text{g/g}$, OSE: 78.989 $\mu\text{g/g}$), and (CE: 44.310 $\mu\text{g/g}$, MWE: 46.076 $\mu\text{g/g}$, OSE: 43.620 $\mu\text{g/g}$), while the minimum amount of it was salicylic acid and hydroxybenzoic acid at all three methods. Also, some other phenolic was showed of catechin hydrate, Phloridzin dihydrate, hydroxycinnamic acid, and butein in methods of (microwave and orbital shaker). Whereas, quercetin, acetohydroxamic acid, and caffeic acid displayed the lowest values in the microwave method.

However, the results were indicated nine kinds of phenolic compounds are abundant in hexane extract by three methods. Under this condition, it contained a great amount of it was vanillic acid and fumaric acid at all three ways (CE: 711.712 $\mu\text{g/g}$, MWE: 719.757 $\mu\text{g/g}$, OSE: 745.311 $\mu\text{g/g}$), and (CE: 182.132 $\mu\text{g/g}$, MWE: 102.95 $\mu\text{g/g}$, MWE: 498.889 $\mu\text{g/g}$). Also, the effective amount of catechin hydrate and resveratrol were showed in all three ways (CE: 70.288 $\mu\text{g/g}$, MWE: 42.839 $\mu\text{g/g}$, OSE: 56.681 $\mu\text{g/g}$) (CE: 63.086 $\mu\text{g/g}$, MWE: 82.641, OSE: 82.286) successively. While the least quantity was acetohydroxamic acid in the microwave method (MWE: 0.856 $\mu\text{g/g}$), and hydroxybenzoic acid at all three methods. Therefore, the silymarin, curmin, and butein were detected by the orbital shaker methods.

Generally, our plant was rich in specific phenolic compounds. Also, our findings in all solvents and with all methods. contradict those of a previous study (Germano *et al.* 2002), which found 0.39 percent w/w rutin in a methanolic extract of *Capparis spinosa* using HPLC analysis. While our study agrees with (Rezzan *et al.* 2013), because identified many phenolic compounds in *Capparis spinosa* of gallic acid, vanillic acid, chlorogenic acid, p-OH benzoic acid, caffeic acid, syringic acid, vanillin, syringaldehyde, p-coumaric acid, ferulic acid, proto-catechuic

acid, and proto-catechuic aldehyde. Also our finding in agreement with (Oudah *et al.* 2019), mentioned, that the presence of (Gallic acid, Caffeic acid, Coumaric acid, Vanillic acid, Syringic acid, Ferulic acid, Chlorogenic acid, Rutin, and Quercetin) in the extract was confirmed by UV-Vis Spectra and high-performance liquid chromatography of Capparis Spinosa derived polyphenols.



Table 4.57. Identification and quantification of analysis specific phenolic compounds of five solvents with three methods of *Capparis Spinosa* fruit extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	65.339	27.346	-	62.554	156.590	-	42.756	69.559	24.859	32.056	52.331	113.831	70.288	42.839	56.681
2	-	-	123.219	-	-	2224.114	-	34.754	-	-	34.157	-	-	-	-
3	2.919	-	9.274	3.989	-	-	-	7.481	1.625	-	9.031	2.681	-	0.856	22.547
4	807.148	825.918	849.534	775.700	838.925	826.338	827.950	824.154	778.510	778.964	853.513	731.643	711.712	719.757	745.311
5	47.517	42.792	31.814	66.830	117.983	274.448	43.632	47.828	121.729	71.681	55.655	78.989	63.086	82.641	82.286
6	457.369	312.183	63.090	191.398	183.839	329.042	339.115	491.319	125.327	245.192	370.343	251.206	182.132	102.954	98.889
7	-	49.640	44.072	45.438	46.544	-	45.432	46.267	-	44.310	46.076	43.620	-	-	-
8	-	-	-	5.773	-	-	5.855	-	-	-	7.091	-	6.571	-	-
9	-	47.544	45.944	-	-	-	56.399	-	-	57.395	47.472	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	-	-	-	-	-	-	55.097	25.693	2.077	128.225	129.612	-	-	-	-
12	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	68.649	-	-	-	180.475	-	69.010	62.769	237.817	-	-	-	-	-	81.200
15	46.451	45.395	-	-	-	45.402	-	-	-	-	-	-	-	-	45.391
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
18	8.833	-	1.230	-	-	1.723	4.494	8.395	6.653	12.407	19.268	3.199	-	-	-
19	15.550	8.465	10.836	5.517	10.217	9.245	2.708	15.933	16.413	16.017	29.869	12.433	4.885	7.303	6.565
20	-	-	-	-	-	4.351	-	11.115	8.601	12.945	12.307	-	-	-	3.979
21	-	-	-	-	-	-	120.953	-	-	-	273.169	-	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1,4 aftokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.4. Identification and quantification of analysis specific phenolic compounds of *Paliurus spinachristi* fruit extract by LC-MS/MS

In this study, the LC-MS/MS was carried to the quantification and determination of phenolic compounds in *Paliurus spinachristi* extract with five solvents by all three methods, as shown in Table (4.58).

Many phenolic compounds in **distilled water** extract were detected by using all three methods. The results observed a rich and high amount of catechin hydrate in the conventional and microwave methods with concentration (CE: 3392,999 $\mu\text{g/g}$), and (MWE: 347,402 $\mu\text{g/g}$). Also, the vanillic acid and resveratrol demonstrated high amounts (CE: 749.724 $\mu\text{g/g}$, MWE: 761.421 $\mu\text{g/g}$, OSE: 901.170 $\mu\text{g/g}$), and (CE: 78.705 $\mu\text{g/g}$, MWE: 35.655 $\mu\text{g/g}$, OSE: 44.460 $\mu\text{g/g}$), while the hydroxybenzoic acid showed the minimum amount in all three methods. On the other hand, the fumaric acid, gallic acid, Phloridzin dihydrate hydroxycinnamic acid, and caffeic acid detected the highest and lowest of amounts phenolic (CE: 140.774 $\mu\text{g/g}$, MWE: 101.351 V), (CE: 54.479 $\mu\text{g/g}$, MWE: 56.169 $\mu\text{g/g}$), (CE: 62.274 $\mu\text{g/g}$, MWE: 89.472 $\mu\text{g/g}$), and (CE: 77.875 $\mu\text{g/g}$, MWE: 16.137 $\mu\text{g/g}$) respectively, in (conventional and microwave) methods. Besides, acetohydroxamic acid, curmin, and salicylic acid have been detected in conventional methods but not detected in the microwave and orbital shaker methods.

Therefore, the twelve compounds of phenolic in (**ethanol: distilled water**) extract were detected by all three methods. It contained maximum quantities of catechin hydrate and vanillic acid at all three methods (CE: 1873.085 $\mu\text{g/g}$ $\mu\text{g/g}$, MWE: 3845.669 $\mu\text{g/g}$, OSE: 160.095 $\mu\text{g/g}$), and (CE: 788.542 $\mu\text{g/g}$, MWE: 729.109 $\mu\text{g/g}$, OSE: 803.947 $\mu\text{g/g}$), with hydroxybenzoic acid was a minimum amount at all three methods. On the other side, quercetin, caffeic acid, and silymarin detected a significant amount in microwave methods. Whereas, the fumaric acid and Phloridzin dihydrate displayed in conventional and microwave methods (CE: 59.520 $\mu\text{g/g}$, MWE: 76.089 $\mu\text{g/g}$), and (CE: 81.274 $\mu\text{g/g}$, MWE: 116.988 $\mu\text{g/g}$). Alternatively, there was no data detected acetohydroxamic acid and curmin at the microwave and orbital shaker methods that were detected in the conventional method, while the gallic acid resveratrol was detected in the conventional and orbital shaker methods.

From the **ethanol** extract by using three methods exhibited fifteen kinds of the phenolic compounds, the all three methods obtained the biggest quantity of catechin hydrate (CE: 10368.997 $\mu\text{g/g}$, MWE: 6084.363 $\mu\text{g/g}$, OSE: 3343.005 $\mu\text{g/g}$), further the Phloridzin dihydrate, vanillic acid, quercetin, hydroxycinnamic acid, and fumaric acid displayed high amounts(CE:

441.906 µg/g, MWE: 225.071 µg/g), (CE: 210.316 µg/g, MWE: 263.925 µg/g), (CE: 197.261 µg/g, MWE: 103.431 µg/g), (CE: 150.993 µg/g, MWE: 12.961 µg/g), and (CE: 112.628 µg/g, MWE: 89.905 µg/g) respectively, and with low amounts of resveratrol, hydroxybenzoic acid, salicylic acid, caffeic acid, and acetohydroxamic acid all specific compounds were detected at the conventional and microwave methods. Also, gallic acid and silymarin were identified in significant amounts at the microwave technique. Whereas, the catechin hydrate was observed a maximum amount (OSE: 3343.005 µg/g) with the high amounts of (vanillic acid 812.174 µg/g), (Phloridzin dihydrate 332.599 µg/g), (resveratrol 142.207 µg/g), and (luteolin 131.736 µg/g) respectively at orbital shaker method. Instead, there was (fumaric acid 74.311 µg/g), (quercetin 57.016), (hydroxybenzoic acid 52.032 µg/g), (salicylic acid 34.837 µg/g), and (oleuropein 28.239 µg/g) in the ethanol extract at orbital shaker methods.

Also, the phenolic compounds were discovered fifteen kinds in (**hexane: ethanol**) extract in all three methods. The (hexane: ethanol) extract containing the biggest amounts of catechin hydrate in all three methods (CE: 10290.24 µg/g, MWE: 9623.260 µg/g, OSE: 7034.122 µg/g), Also, the phenolic compounds of vanillic acid, Phloridzin dihydrate, quercetin, hydroxycinnamic acid, and resveratrol displayed high quantities (CE: 806.614 µg/g, MWE: 770.207 µg/g, OSE: 773.113 µg/g), (CE: 586.861 µg/g, MWE: 442.244 µg/g, OSE: 525.373 µg/g), (CE: 174.648 µg/g, MWE: 117.121 µg/g, OSE: 180.566 µg/g), (CE: 158.629 µg/g, MWE: 127.472 µg/g, OSE: 24.259 µg/g), (CE: 95.925 µg/g, MWE: 125.846 µg/g, OSE: 161.580 µg/g), while the significant and low amounts were observed of each fumaric acid, gallic acid, salicylic acid, hydroxybenzoic acid, and caffeic acid (CE: 80.876 µg/g, MWE: 73.877 µg/g, OSE: 87.249 µg/g), (CE: 46.219 µg/g, MWE: 43.468 µg/g, OSE: 60.305 µg/g), (CE: 52.680 µg/g, MWE: 1.862 µg/g, OSE: 54.661 µg/g), (CE: 43.928 µg/g, MWE: 30.667 µg/g, OSE: 41.233 µg/g), and (CE: 12.990 µg/g, MWE: 6.632 µg/g, OSE: 6.444 µg/g) respectively. On the other side, the maximum amount of phenolic compounds was luteolin (OSE: 2647.491 µg/g), and high amounts of it was silymarin (OSE: 80.769 µg/g, MWE: 149.950 µg/g) respectively, and the kaempferol in an orbital shaker (OSE: 173.239 µg/g). Contrarily, a little amount of acetohydroxamic acid was detected in conventional and microwave techniques.

The results of the **hexane** extract obtained twelve specific compounds of phenolic in all methods. However, the value of vanillic acid found the maximum amount (CE: 1147.294 µg/g), with the high amounts of catechin hydrate and resveratrol in the conventional method (CE: 611.901 µg/g), (CE: 115.679 µg/g). Furthermore, the silymarin, Phloridzin dihydrate, fumaric

acid, gallic acid, and curmin were exhibited (CE: 62.506 $\mu\text{g/g}$, 58.095 $\mu\text{g/g}$, 48.485 $\mu\text{g/g}$, 46.310 $\mu\text{g/g}$, and 44.870 $\mu\text{g/g}$) respectively, in the conventional method. While the microwave method was observed a high amount of vanillic acid (MWE: 919.850 $\mu\text{g/g}$). Also, the obtained amounts were (silymarin 68.412 $\mu\text{g/g}$), (fumaric acid 60.252 $\mu\text{g/g}$), (gallic acid 47.627 $\mu\text{g/g}$), (resveratrol 36.237 $\mu\text{g/g}$), (catechin hydrate 25.824 $\mu\text{g/g}$), and the lowest amount of (caffeic acid 6.578 $\mu\text{g/g}$) respectively. On the other hand, the highest amount obtained by the orbital shaker was vanillic acid catechin hydrate, and quercetin (OSE: 798.584 $\mu\text{g/g}$, 290.090 $\mu\text{g/g}$, and 122.987 $\mu\text{g/g}$) respectively. Also, the specific amount was (resveratrol 73.342 $\mu\text{g/g}$), and (fumaric acid 60.177 $\mu\text{g/g}$) respectively. Nonetheless, salicylic acid and acetohydroxamic acid were given in the smallest amounts (OSE: 3.916 $\mu\text{g/g}$, OSE: 0.360 $\mu\text{g/g}$), and the lowest amount detected at all three methods was hydroxybenzoic acid.

In general, the *Paliurus Spina Christi* fruit extracts was containing a high of phenolic compounds quantitatively analyzed by LC-MS/MS. Our study is not agreement with (Ads *et al.* 2018) had reported, that the *zizith's spina-Christi*, have been detected and recognized of compounds botulin, quercetin, stigmasterol, sitosterol, ethyl oleate, and phytol through using GC-MS tests.

Table 4.58. Identification and quantification of analysis specific phenolic compounds of five solvents with three methods of *Paliurus spinachristi* fruit extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	3392.999	347.402	-	1873.08	3845.66	160.095	10368.99	6084.36	3343.005	10290.24	9623.26	7034.122	611.901	25.824	290.090
2	-	-	-	-	53.796	-	197.261	103.431	57.016	174.648	117.121	180.566	-	-	122.987
3	3.973	-	-	4.647	-	-	4.063	2.388	-	9.736	2.987	-	4.665	-	3.916
4	749.724	761.421	901.170	788.542	729.109	803.947	210.316	263.925	812.174	806.614	770.207	773.113	1147.29	919.850	798.584
5	78.705	35.655	44.460	47.264	-	70.993	55.292	90.560	142.207	95.925	125.846	161.580	115.679	36.237	73.342
6	140.774	101.351	-	59.520	76.089	-	112.628	89.905	74.311	80.876	73.877	87.249	48.485	60.252	60.177
7	54.479	56.169	-	47.640	-	42.647	-	48.375	-	46.219	43.468	60.305	46.310	47.627	-
8	8.451	12.468	-	-	7.476	-	8.581	5.675	-	12.990	6.632	6.444	-	6.578	-
9	62.274	89.472	-	81.274	116.988	-	441.906	225.071	332.599	586.861	442.244	525.373	58.095	-	-
10	-	-	-	-	-	-	-	-	28.239	-	-	-	-	-	-
11	77.875	16.137	-	-	-	-	150.993	12.961	-	158.629	127.472	24.259	-	-	-
12	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	-	-	-	-	67.852	-	-	208.144	-	-	149.950	80.769	62.506	68.412	-
15	45.402	-	-	45.402	-	-	-	-	-	-	-	-	44.870	-	-
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-	-	-	-	173.239	-	-	-
18	38.208	-	-	-	4.184	-	44.102	12.031	34.837	43.928	30.667	41.233	-	-	0.360
19	42.441	9.126	2.762	11.100	18.515	6.594	55.093	27.673	52.032	52.680	1.862	54.661	7.021	1.749	11.341
20	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
21	-	-	-	-	-	-	3.124	-	131.736	15.976	-	2647.491	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1,4 aftokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.5. Identification and quantification of analysis specific phenolic compounds of *Portulaca oleracea* herb extract by LC-MS/MS

In this study, the five solvents through all three methods of *portulaca oleracea* have detected many types of phenolic compounds analyzed by LC-MS/MS, as displayed in Table (4.59). The **distilled water** extract has been detected twelve types of phenolic compounds at all three methods. The maximum amount of vanillic acid was obtained (CE: 864.844 $\mu\text{g/g}$, MWE: 856.071 $\mu\text{g/g}$, OSE: 915.551 $\mu\text{g/g}$) at all three techniques. Also, the resveratrol, gallic acid, and catechin hydrate were displayed definite amounts in all three methods at (CE: 63.759 $\mu\text{g/g}$, MWE: 60.052 $\mu\text{g/g}$, OSE: 98.317 $\mu\text{g/g}$), (CE: 46.653 $\mu\text{g/g}$, MWE: 45.229 $\mu\text{g/g}$, OSE: 44.534 $\mu\text{g/g}$), and (CE: 26.519 $\mu\text{g/g}$, MWE: 54.984 $\mu\text{g/g}$, OSE: 23.073 $\mu\text{g/g}$) while the lowest amount was acetohydroxamic acid and hydroxybenzoic acid (CE: 1.426 $\mu\text{g/g}$, MWE: 1.650 $\mu\text{g/g}$, OSE: 3.179 $\mu\text{g/g}$), and (CE: 14.658 $\mu\text{g/g}$, MWE: 10.396 $\mu\text{g/g}$, OSE: 10.548 $\mu\text{g/g}$) in all methods. On the other hand, it was detected high amounts of fumaric acid (CE: 125.46 $\mu\text{g/g}$, MWE: 396.161 $\mu\text{g/g}$), and hydroxycinnamic acid (CE: 130.815 $\mu\text{g/g}$, MWE: 32.661 $\mu\text{g/g}$) in conventional and microwave methods. Whereas, the ellagic acid detected high quantity (CE: 289.861) at conventional methods, as well as the curmin (OSE: 45.402 $\mu\text{g/g}$), and quercetin (OSE: 156.668 $\mu\text{g/g}$) in an orbital shaker.

The (**ethanol: distilled water**) extracts shown fourteen kinds of phenolic compounds at different three methods. The topmost quantity compound was vanillic acid (CE: 815.498 $\mu\text{g/g}$, MWE: 760.757 $\mu\text{g/g}$, OSE: 754.215 $\mu\text{g/g}$), and the specific amounts were resveratrol (CE: 92.346 $\mu\text{g/g}$, MWE: 36.248 $\mu\text{g/g}$, OSE: 96.493 $\mu\text{g/g}$), gallic acid (CE: 44.915 $\mu\text{g/g}$, MWE: 42.730 $\mu\text{g/g}$, OSE: 43.130 $\mu\text{g/g}$), catechin hydrate (CE: 24.961 $\mu\text{g/g}$, MWE: 23.809 $\mu\text{g/g}$, OSE: 24.058 $\mu\text{g/g}$) at all three methods. Instead, the hydroxybenzoic acid and acetohydroxamic acid detected lowest amounts (CE: 13.083 $\mu\text{g/g}$, MWE: 3.129 $\mu\text{g/g}$, OSE: 16.455 $\mu\text{g/g}$), and (CE: 1.701 $\mu\text{g/g}$, MWE: 3.465 $\mu\text{g/g}$, OSE: 4.781 $\mu\text{g/g}$) respectively. Hence, the luteolin, silymarin, and quercetin were detected high amount at orbital shaker technique (OSE: 740.347 $\mu\text{g/g}$, 112.628 $\mu\text{g/g}$, 38.095 $\mu\text{g/g}$) respectively, and the smallest quantity of butein (OSE: 10.683 $\mu\text{g/g}$). Also, the conventional method was identified hydroxycinnamic acid, caffeic acid, and curmin, from maximum value to the minimum value, respectively.

Furthermore, the phenolic compounds were detected in twelve types in **ethanol** extract at all three methods. It contained the greatest amounts were vanillic acid at all three-technique (CE: 961.285 $\mu\text{g/g}$, MWE: 723.759 $\mu\text{g/g}$, OSE: 961.329 $\mu\text{g/g}$). As well as, the high amounts of

resveratrol found by all methods (CE: 187.461 $\mu\text{g/g}$, MWE: 135.779 $\mu\text{g/g}$, OSE: 170.749 $\mu\text{g/g}$) while the each of gallic acid, butein, hydroxybenzoic acid, salicylic acid, and acetohydroxamic acid showed specific and low amounts (CE: 44.990 $\mu\text{g/g}$, MWE: 43.111 $\mu\text{g/g}$, OSE: 42.596 $\mu\text{g/g}$, (CE: 42.130 $\mu\text{g/g}$, MWE: 19.131 $\mu\text{g/g}$, OSE: 8.211 $\mu\text{g/g}$), (CE: 22.132 $\mu\text{g/g}$, MWE: 13.546 $\mu\text{g/g}$, OSE: 10.343 $\mu\text{g/g}$), (CE: 12.497 $\mu\text{g/g}$, MWE: 1.863 $\mu\text{g/g}$, OSE: 1.771 $\mu\text{g/g}$), and (CE: 6.128 $\mu\text{g/g}$, MWE: 2.963 $\mu\text{g/g}$, OSE: 1.481 $\mu\text{g/g}$) respectively. On the other side, the conventional method was observed definite amounts of hydroxycinnamic acid, oleuropein, fumaric acid, catechin hydrate, caffeic acid, and curmin, but not data found in other techniques.

Hence, the (**hexane: ethanol**) extracts gave fifteen kinds of phenolic compounds in all three ways. The uppermost and high quantities were vanillic acid and resveratrol (CE: 995.175 $\mu\text{g/g}$, MWE: 865.787 $\mu\text{g/g}$, OSE: 885.471 $\mu\text{g/g}$), and (CE: 306.745 $\mu\text{g/g}$, MWE: 88.599 $\mu\text{g/g}$, OSE: 280.096 $\mu\text{g/g}$) in all three ways. Also, the definite amount was detected of hydroxycinnamic acid, gallic acid, and butein (CE: 99.154 $\mu\text{g/g}$, MWE: 44.550 $\mu\text{g/g}$, OSE: 42.968 $\mu\text{g/g}$), (CE: 42.457 $\mu\text{g/g}$, MWE: 43.111 $\mu\text{g/g}$, OSE: 42.596 $\mu\text{g/g}$), and (CE: 59.118 $\mu\text{g/g}$, MWE: 9.658 $\mu\text{g/g}$, OSE: 49.402 $\mu\text{g/g}$). While it has been obtained the minimum amounts of salicylic acid and hydroxybenzoic acid, and caffeid acid (CE: 13.472 $\mu\text{g/g}$, MWE: 8.380 $\mu\text{g/g}$, OSE: 18.862 $\mu\text{g/g}$), and (CE: 16.721 $\mu\text{g/g}$ CE: 15.887 $\mu\text{g/g}$ CE: 28.829 $\mu\text{g/g}$), and (CE: 7.464 $\mu\text{g/g}$, MWE: 6.466 $\mu\text{g/g}$, OSE: 11.185 $\mu\text{g/g}$) respectively. On the other hand, silymarin (OSE: 87.789 $\mu\text{g/g}$), quercetin (OSE: 53.310 $\mu\text{g/g}$), and acetohydroxamic acid (OSE: 1.852 $\mu\text{g/g}$) were displayed in orbital shaker methods. Further, the fumaric acid (CE: 52.925 $\mu\text{g/g}$), catechin hydrate (CE: 25.714 $\mu\text{g/g}$), and oleuropein (CE: 36.287 $\mu\text{g/g}$) detected by the conventional method. Instead, the luteolin has been detected by microwave and orbital shaker method, but not identified at conventional method.

The solvent of **hexane** extract was found eight compounds of phenolic by using three methods, the phenolic compound as vanillic acid was detected high amount at all three techniques (CE: 969.854 $\mu\text{g/g}$, MWE: 883.583 $\mu\text{g/g}$, OSE: 794.341 $\mu\text{g/g}$), and the resveratrol displayed the specific quantity (CE: 46.380, MWE: 90.543, OSE: 70.872) while, it has been least amount of hydroxybenzoic acid (CE: 10.898 $\mu\text{g/g}$, MWE: 2.303 $\mu\text{g/g}$, OSE: 6.128 $\mu\text{g/g}$) thru all techniques. On the other side, the catechin hydrate was discovered the biggest amount (CE: 3211.209 $\mu\text{g/g}$) at conventional method, but the fumaric acid was shown specific amount at conventional and orbital shaker methods (CE: 84.092 $\mu\text{g/g}$, and OSE: 53.043 $\mu\text{g/g}$) in compare with smallest amounts of the salicylic acid and caffeic acid (CE: 0.343 $\mu\text{g/g}$, OSE:

0.193 $\mu\text{g/g}$), and (CE: 6.282 $\mu\text{g/g}$, and OSE: 5.424 $\mu\text{g/g}$). However, it obtained a high quantity of phloridzindihydrate (CE: 101.073 $\mu\text{g/g}$) by the conventional method.

Our finding agrees with (Gallo *et al.* 2017) reported, that the phenolic compounds present in the *Portulaca Oleracea* leaves studied were qualitatively analyzed utilizing Liquid Chromatography-Tandem Mass Spectrometry (LC/MS/MS). However, for quality valuation of phenolic compounds from *Portulaca Oleracea* extract. Which utilizing the permanent LC-MS/MS method, because the speedy and effective method., our study agrees with (Liang *et al.* 2014a) But, our finding more effective than the results (Liang *et al.* 2014a) analyzed by LC-MS/MS.



Table 4.59. Identification and quantification of analysis specific phenolic compounds in five solvents with three methods of *Portulaca oleracea L* extract by LC ESI MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	26.519	54.984	23.073	24.961	23.809	24.058	25.023	-	23.013	25.714	-	-	3211.20	-	-
2	-	-	156.668	-	-	38.095	-	-	-	-	-	53.310	-	-	-
3	1.426	1.650	3.179	1.701	3.465	4.781	6.128	2.963	1.481	-	-	1.852	-	1.889	3.426
4	864.844	856.071	915.551	815.498	760.757	754.215	961.285	723.759	961.329	995.175	865.787	885.471	969.854	883.583	794.341
5	63.759	60.052	98.317	92.346	36.248	96.493	187.461	135.779	170.749	306.745	88.599	280.096	46.380	90.543	70.872
6	125.463	96.161	-	124.624	-	149.513	50.398	60.545	-	52.925	-	55.268	84.092	-	53.043
7	46.653	45.229	44.534	44.915	42.730	43.130	44.990	43.111	42.596	42.457	44.550	42.968	-	-	-
8	-	6.456	-	8.567	5.881	-	14.070	5.792	-	7.464	6.466	11.185	6.282	-	5.424
9	-	-	-	-	-	-	-	-	-	-	-	-	101.073	-	-
10	-	-	-	-	-	-	60.744	-	-	36.287	-	-	-	-	-
11	130.815	32.661	-	15.188	-	7.750	81.074	3.369	-	99.154	24.632	84.511	-	-	-
12	289.861	-	-	-	-	-	-	-	-	-	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	-	-	-	-	-	112.628	-	-	-	-	-	87.789	-	-	-
15	45.931	-	45.402	46.444	-	-	45.395	45.931	-	-	-	-	-	-	-
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
18	-	3.317	-	-	-	8.072	12.497	1.863	1.771	13.472	8.380	18.862	0.343	-	0.193
19	14.658	10.396	10.548	13.083	3.129	16.455	22.132	13.546	10.343	16.721	15.887	28.829	10.898	2.303	6.128
20	-	-	-	-	-	10.683	42.130	19.131	8.211	59.118	9.658	49.402	-	-	-
21	-	-	-	-	-	740.347	-	-	-	-	363.839	449.786	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1.4 aftokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.6. Identification and quantification of analysis specific phenolic compounds of *Artemisia haussknechtii* Boiss herb extract by LC-MS/MS

In the current study, several types of phenolic compounds were carried out by LC-MS/MS for five solvents of *Artemisia haussknechtii* Boiss extract, at all three methods, as presented in Table (4.60).

The twelve kinds of compounds were discovered in **distilled water** extract in all three techniques. The value of vanillic acid was showed highest in all three methods (CE: 744.776 µg/g, MWE: 728.202 µg/g, OSE: 793.161 µg/g), and the resveratrol and caffeic acid gave specific amounts as (CE: 74.759 µg/g, MWE: 46.606 µg/g, OSE: 75.744 µg/g), and (CE: 24.195 µg/g, MWE: 24.490 µg/g, OSE: 7.772 µg/g), while the minimum of phenolic compounds was hydroxybenzoic acid (CE: 5.955 µg/g, MWE: 7.017 µg/g, OSE: 21.340 µg/g) respectively. On the other hand, the orbital shaker methods observed particular amounts for each gallic acid (OSE: 56.950 µg/g, hydroxycinnamic acid 56.269 µg/g, curmin 44.870 µg/g, hydroxycinnamic acid 24.069 µg/g, salicylic acid 16.362 µg/g, and acetohydroxamic acid 4.811 µg/g), respectively. Also, the distilled water in (conventional and microwave) methods revealed many phenolic as (gallic acid CE: 46.006 µg/g), (catechin hydrate CE: 24.564 µg/g), (fumaric acid CE: 220.021 µg/g), (fumaric acid MWE: 251.956 µg/g), and (silymarin MWE: 61.705 µg/g) respectively.

Furthermore, the (**ethanol: distilled water**) extract identified nine types of phenolic compounds by using three methods. In general, the maximum was vanillic acid (CE: 867.843 µg/g, MWE: 852.239 µg/g, OSE: 312.411 µg/g), and resveratrol (CE: 43.567 µg/g, MWE: 67.066 µg/g, OSE: 74.606 µg/g) was specific abundant compounds at all three methods, while hydroxybenzoic acid was the smallest values (CE: 4.167 µg/g, MWE: 2.670 µg/g, OSE: 6.561 µg/g) through all methods. On the other side, the highest and lowest amounts of orbital shaker method of (ethanol: distilled water) extract were luteolin (OSE: 281.480 µg/g), fumaric acid (OSE: 223.420 µg/g), hydroxycinnamic acid (OSE: 7.003 µg/g, caffeic acid OSE: 6.664 µg/g), and acetohydroxamic acid (OSE: 2.356 µg/g,) respectively. Also, the fumaric acid, curmin, caffeic acid, acetohydroxamic acid were detected amounts in microwave methods (MWE: 158.771 µg/g, 45.402 µg/g, 9.806 µg/g, and 1.926 µg/g) respectively. Rather, the conventional method no detected any acetohydroxamic acid, fumaric acid, caffeic acid, hydroxycinnamic acid, curmin, and luteolin.

Also, the seventeen kinds of phenolic have been identified in ethanol extract with all three methods. The conventional method gave the greatest quantities of luteolin (5172.051 $\mu\text{g/g}$), quercetin (1513.400 $\mu\text{g/g}$), hydroxycinnamic acid (1071.642 $\mu\text{g/g}$), and fumaric acid (1004.286 $\mu\text{g/g}$) respectively. Also, vanillic acid, kaempferol, caffeic acid, oleuropein, and resveratrol have been obtained high amounts (CE: 737.589 $\mu\text{g/g}$, 543.738 $\mu\text{g/g}$, 586.883 $\mu\text{g/g}$, 227.093 $\mu\text{g/g}$, and 168.005 $\mu\text{g/g}$). Whereas, the low and specified amounts were salicylic acid 10.963 $\mu\text{g/g}$, acetohydroxamic acid (5.326 $\mu\text{g/g}$), and butein (2.094 $\mu\text{g/g}$), catechin hydrate (84.195 $\mu\text{g/g}$), silymarin (68.382 $\mu\text{g/g}$), Phloridzin dihydrate (46.412 $\mu\text{g/g}$), curmin (45.371 $\mu\text{g/g}$), hydroxybenzoic acid (30.729 $\mu\text{g/g}$) respectively, through conventional methods. On the other side, the microwave and orbital shaker were given a maximum amount of luteolin (MWE: 2342.891 $\mu\text{g/g}$, OSE: 1668.65 $\mu\text{g/g}$). Hence, the high amounts of vanillic acid, fumaric acid, kaempferol, quercetin, hydroxycinnamic acid, caffeic acid, resveratrol, and oleuropein were recorded (MWE: 848.204 $\mu\text{g/g}$, OSE: 863.393 $\mu\text{g/g}$), (MWE: 622.794 $\mu\text{g/g}$, OSE: 403.146 $\mu\text{g/g}$), (MWE: 438.835 $\mu\text{g/g}$, OSE: 341.713 $\mu\text{g/g}$), (MWE: 442.261 $\mu\text{g/g}$, OSE: 439.100 $\mu\text{g/g}$), (MWE: 395.753 $\mu\text{g/g}$, OSE: 403.030 $\mu\text{g/g}$), (MWE: 172.672 $\mu\text{g/g}$, OSE: 120.414 $\mu\text{g/g}$), (MWE: 121.674 $\mu\text{g/g}$, OSE: 177.669 $\mu\text{g/g}$), and (MWE: 103.490 $\mu\text{g/g}$, OSE: 115.943 $\mu\text{g/g}$) respectively, at microwave and orbital shaker methods, while the salicylic acid and hydroxybenzoic acid detected the lowest amounts at microwave and conventional methods. Otherwise, the acetohydroxamic is not sensed in the orbital shaker method, but detected in the microwave method.

The **(hexane: ethanol)** extract was showed fourteen kinds of phenolic acid by all various methods. It contained the biggest amounts of luteolin and hydroxycinnamic acid at conventional method were (CE: 4979.472 $\mu\text{g/g}$, 1647.117 $\mu\text{g/g}$) respectively. Additionally, the highest quantities and the specified quantities of phenolic acid were (quercetin 921.528 $\mu\text{g/g}$, vanillic acid 765.268 $\mu\text{g/g}$, kaempferol 670.911 $\mu\text{g/g}$, caffeic acid 620.094 $\mu\text{g/g}$, fumaric acid 300.018 $\mu\text{g/g}$, and resveratrol 206.923 $\mu\text{g/g}$), and (Phloridzin dihydrate 46.834 $\mu\text{g/g}$, gallic acid 43.114 $\mu\text{g/g}$) at conventional method. Also, the minimum quantities of butein, hydroxybenzoic acid, salicylic acid, and acetohydroxamic acid were recorded. However, the microwave way had obtained the maximum amount of luteolin (MWE: 1636.162 $\mu\text{g/g}$), and high amounts of several compounds of phenolic acids such as vanillic acid, hydroxycinnamic acid, quercetin, kaempferol, fumaric acid, caffeic acid, and resveratrol by microwave mode were (MWE: 820.000 $\mu\text{g/g}$, 331.676 $\mu\text{g/g}$, 287.736 $\mu\text{g/g}$, 247.926 $\mu\text{g/g}$, 128.519 $\mu\text{g/g}$, 122.723 $\mu\text{g/g}$, and 81.868 $\mu\text{g/g}$), while the salicylic acid and hydroxybenzoic acid displayed least amounts at

microwave way. On the other hand, the results of the orbital shaker mode presented that vanillic acid and fumaric acid in the highest amounts (OSE: 786.472 $\mu\text{g/g}$, 179.282 $\mu\text{g/g}$). Further, the resveratrol, luteolin, and quercetin were given specified amounts of (OSE: 78.652 $\mu\text{g/g}$, 70.527 $\mu\text{g/g}$, and 54.924 $\mu\text{g/g}$), while the caffeic acid, hydroxycinnamic acid, hydroxybenzoic acid, and acetohydroxamic acid showed the lowest amounts at the orbital shaker.

Thus, through our study thirteen types of phenolic compounds in **hexane** extract have been detected by all methods. The maximum kinds of phenolic acids of hydroxycinnamic acid and vanillic acid were found (CE: 1634.574 $\mu\text{g/g}$, and CE: 1139.121 $\mu\text{g/g}$). Though, the high amounts of each resveratrol, hydroxycinnamic acid, fumaric, and gallic acid found in (CE: 259.605 $\mu\text{g/g}$, 1634.574 $\mu\text{g/g}$, 105.485 $\mu\text{g/g}$, and 44.953 $\mu\text{g/g}$) was extra than the amounts of other compounds, while caffeic acid and hydroxybenzoic acid were minimum amounts (CE: 5.332 $\mu\text{g/g}$, and 5.649 $\mu\text{g/g}$) through a conventional method. On the other hand, the specified amounts of phenolic were resveratrol (66.878 $\mu\text{g/g}$), quercetin (54.580 $\mu\text{g/g}$), curmin (45.292 $\mu\text{g/g}$), caffeic acid (27.453 $\mu\text{g/g}$), catechin hydrate (24.626 $\mu\text{g/g}$), luteolin (24.676 $\mu\text{g/g}$) and hydroxybenzoic acid (1.310 $\mu\text{g/g}$) at microwave mode. So, the orbital shaker was containing a high amount of vanillic acid (OSE: 768.872 $\mu\text{g/g}$), and fumaric acid (OSE: 170.090 $\mu\text{g/g}$), also, the resveratrol and hydroxycinnamic acid were shown specified quantities (OSE: 80.628 $\mu\text{g/g}$), and (OSE: 64.162 $\mu\text{g/g}$), while the minimum amount of caffeic, hydroxybenzoic acid, and acetohydroxamic acid in an orbital shaker mode were investigated.

Our study mentioned, that *Artemisia haussknechtii* Boiss are different studies about the species of plant and phenolic compound with another study when carried out analyzed by LC-MS/MS. (Erdemoglu *et al.* 2007) that the number of *Artemisia* species, *artemisinin* was not identified in all investigated *Artemisia* species of *Artemisia haussknechtii* Boiss, it was observed lower than LOD value measured by HPLC. Furthermore, various chromatographic analytical techniques were used to study the crude ethanolic extract of *Artemisia brasiliensis* to determine its major chemical constituents. Many compounds were described using GC-MS analysis and LC-MS/MS, repeating the study's importance and contribution to the information of chemistry (Alencar Filho *et al.* 2019) this study in line with our study. The plant of *Artemisia aucheri* Boiss (Mohammadpoor *et al.* 2002). Another study on *Artemisia haussknechtii* Boiss plants produces 56 volatile compounds. Camphor (41.01 %), 1,8-cineole (32.35 %), cis-davanone (3.68 %), 4-terpineol (2.99 %), linalool (2.84 %), beta-fenchyl alcohol (2.72 %), and borneol are the key constituents of the essential oil extracted using the HD-SHLPME technique (2.58 %) (Jalali Heravi and Sereshti 2007), (Sereshti and Samadi 2007).

Table 4.60. Identification and quantification of analysis specific phenolic compounds in five solvents with three methods of *Artemisia haussknechtii* Boiss extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	24.564	-	-	-	-	-	84.195	-	-	-	-	-	-	24.626	-
2	-	-	-	-	-	-	1513.400	442.261	439.100	921.528	287.736	54.924	-	54.580	-
3	-	-	4.811	-	1.926	2.356	5.326	-	5.342	5.123	-	4.663	-	-	1.239
4	744.776	728.202	793.161	867.843	852.239	312.411	737.589	848.204	863.393	765.268	820.000	786.472	1139.12	851.311	768.872
5	74.759	46.606	75.744	43.567	67.066	74.606	168.005	121.674	177.669	206.923	81.868	78.652	259.605	66.878	80.628
6	220.021	251.956	-	-	158.771	223.420	1004.286	622.794	403.146	300.018	128.519	179.282	105.485	106.377	170.090
7	46.006	-	56.950	-	-	-	-	-	-	43.114	-	-	44.953	-	-
8	24.195	24.490	7.772	-	9.806	6.664	586.883	172.672	120.414	620.094	122.723	13.418	5.332	27.453	5.238
9	-	-	-	-	-	-	46.412	-	-	46.834	-	-	-	-	-
10	-	-	-	-	-	-	227.093	103.490	115.943	-	-	-	-	-	-
11	-	24.069	56.269	-	N.D	7.003	1071.642	395.753	403.030	1647.117	331.676	7.696	1634.57 4	507.583	64.162
12	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	-	61.705	-	-	-	-	68.382	-	-	-	-	-	-	-	-
15	-	-	44.870	-	45.402	-	45.371	-	-	-	-	-	-	45.292	-
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	543.738	438.835	341.713	670.911	247.926	-	-	135.624	-
18	-	-	16.362	-	-	-	10.963	6.904	4.377	11.908	1.339	-	-	-	-
19	5.955	7.017	21.340	4.167	2.670	6.561	30.729	11.812	13.378	21.383	9.506	5.317	5.649	1.310	4.765
20	-	-	-	-	-	-	2.094	-	-	18.964	-	-	-	-	-
21	-	-	-	-	-	281.480	5172.051	2342.89	1668.651	4979.472	1636.16	70.527	-	24.676	-
22	-	-	-	-	-	-	84.195	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	1513.400	442.261	439.100	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1,4 aetokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.7. Identification and quantification of analysis specific phenolic compounds of *Prunus microcarpa* extract by LC-MS/MS

The consequences were showed that various phenolic compounds are abundant in the five solvents of *Prunus microcarpa* extract at all three techniques by LC-MS/MS analysis, are summarized in Table (4.61).

Generally, all three methods in **distilled water** extract were observed ten phenolic compounds. The vanillic acid and resveratrol (CE: 748.113 $\mu\text{g/g}$, MWE: 758.992 $\mu\text{g/g}$, OSE: 774.115 $\mu\text{g/g}$), and (CE: 109.712 $\mu\text{g/g}$, MSE: 125.612 $\mu\text{g/g}$, OSE: 39.566 $\mu\text{g/g}$) at all three methods were found to be the most abundant and the caffeic acid, hydroxybenzoic acid, and acetohydroxamic acid (CE: 12.698 $\mu\text{g/g}$, MWE: 19.986 $\mu\text{g/g}$, OSE: 5.579 $\mu\text{g/g}$), (CE: 7.709 $\mu\text{g/g}$, MWE: 10.251 $\mu\text{g/g}$, OSE: 9.901 $\mu\text{g/g}$), and (CE: 2.963 $\mu\text{g/g}$, MWE: 1.209 $\mu\text{g/g}$, MSE: 4.197 $\mu\text{g/g}$) respectively. The other compounds of phenolic have been showed in different methods as the quercetin 55.626, Phloridzin dihydrate 48.164 $\mu\text{g/g}$, curmin 46.451 $\mu\text{g/g}$, gallic acid 43.485 $\mu\text{g/g}$, and salicylic acid 4.912 $\mu\text{g/g}$, at microwave method, while the quercetin, gallic acid, and catechin hydrate in the conventional method (51.240 $\mu\text{g/g}$, 43.924 $\mu\text{g/g}$, 37.273 $\mu\text{g/g}$), and the curmin 45.931 $\mu\text{g/g}$, in orbital shaker was detected.

The results were intimated twelve compounds of phenolic were plentiful in (ethanol: distilled) water extract by three methods. It carried the highest amount of vanillic acid (CE: 906.382 $\mu\text{g/g}$, MWE: 875.010 $\mu\text{g/g}$, OSE: 709.558 $\mu\text{g/g}$), were obtained as well as, the containing quantities of resveratrol (CE: 62.797 $\mu\text{g/g}$, MWE: 70.727 $\mu\text{g/g}$, OSE: 44.602 $\mu\text{g/g}$), and fumaric acid were recorded (CE: 53.468 $\mu\text{g/g}$, MWE: 46.130 $\mu\text{g/g}$, OSE: 76.023 $\mu\text{g/g}$), at all three techniques. Whereas, hydroxybenzoic acid and acetohydroxamic acid had shown the lowest quantities in all three techniques. On the other hand, the catechin hydrate, quercetin, and salicylic acid detected in the (microwave and orbital shaker) methods (MWE: 24.549 $\mu\text{g/g}$, OSE: 23.768 $\mu\text{g/g}$), (MWE: 127.818, OSE: 38.319 $\mu\text{g/g}$), and (MWE: 2.003 $\mu\text{g/g}$, OSE: 0.972 $\mu\text{g/g}$), while the caffeic acid and hydroxycinnamic acid were discovered at conventional and orbital shaker methods. Also, the butein was detected a low amount in the microwave method, and the gallic acid was noticed in conventional and microwave methods.

The **ethanol** extract was identified thirteen kinds of phenolic in all three methods. The high amounts were detected of vanillic acid silymarin, and quercetin (CE: 828.939 $\mu\text{g/g}$, 168.530 $\mu\text{g/g}$, and 118.025 $\mu\text{g/g}$) by the conventional method. Moreover, the specified amounts of the conventional method were resveratrol, gallic acid, curmin, fumaric acid, and catechin hydrate (CE: 86.217 $\mu\text{g/g}$, 45.075 $\mu\text{g/g}$, 45.395 $\mu\text{g/g}$, 44.962 $\mu\text{g/g}$, and 28.196 $\mu\text{g/g}$)

respectively. On the other hand, the maximum amounts were obtained luteolin (MWE: 923.404 $\mu\text{g/g}$), vanillic acid (MWE: 797.435 $\mu\text{g/g}$), kaempferol (MWE: 703.604 $\mu\text{g/g}$), and quercetin (MWE: 604.013 $\mu\text{g/g}$) respectively, while the resveratrol and fumaric acid detected the specified amounts (MWE: 86.597 $\mu\text{g/g}$, and 62.260 $\mu\text{g/g}$) by microwave method. Therefore, the orbital shaker method was given the highest amount of vanillic acid (893.050 $\mu\text{g/g}$), and the defined amounts of resveratrol (70.271 $\mu\text{g/g}$), curmin (45.395 $\mu\text{g/g}$), and catechin hydrate (23.556 $\mu\text{g/g}$). Whereas, for each acetohydroxamic acid, caffeic acid, and hydroxybenzoic acid obtained low quantities at all three methods.

In addition, ten types of phenolic compounds have been detected in (**hexane: ethanol**) extract by three techniques. It limited a great amount of vanillic acid at all three methods (CE: 827.400 $\mu\text{g/g}$, MWE: 791.143 $\mu\text{g/g}$, OSE: 810.907 $\mu\text{g/g}$). Also, the resveratrol was given a specified amount (CE: 58.282 $\mu\text{g/g}$, MWE: 70.996 $\mu\text{g/g}$, OSE: 59.128 $\mu\text{g/g}$) in all three methods, while the lowest amount of it was hydroxybenzoic acid, and acetohydroxamic acid in all methods. On the other side, the kaempferol was detected a high amount (CE: 219.760 $\mu\text{g/g}$), and quercetin (CE: 67.292 $\mu\text{g/g}$) respectively, with the lowest of caffeic acid at conventional method. So, the orbital shaker detected catechin hydrate, salicylic acid, curmin, and gallic acid by microwave method. In comparison, no fumaric acid, phloridzin dihydrate, hydroxycinnamic acid, ellagic acid, myricetin, silymarin, and butein were detected using any of the three methods.

The results were showed eight varieties of phenolic compounds in **hexane** extract analyzed by LC-MS/MS, at all three methods. The highest amount and the lowest amounts of phenolic compounds were vanillic acid (CE: 808.539 $\mu\text{g/g}$, MWE: 821.900 $\mu\text{g/g}$, OSE: 854.649 $\mu\text{g/g}$), and the hydroxybenzoic acid, acetohydroxamic acid (CE: 5.397 $\mu\text{g/g}$, MWE: 2.598 $\mu\text{g/g}$, OSE: 4.238 $\mu\text{g/g}$), and (CE: 1.849 $\mu\text{g/g}$, MWE: 4.415 $\mu\text{g/g}$, OSE: 3.142 $\mu\text{g/g}$) at all three methods, while the resveratrol, gallic acid, curmin have been detected a specific amount in all methods (CE: 58.421 $\mu\text{g/g}$, MWE: 53.993 $\mu\text{g/g}$, OSE: 85.846 $\mu\text{g/g}$), (CE: 45.363 $\mu\text{g/g}$, MWE: 45.582 $\mu\text{g/g}$, OSE: 42.726 $\mu\text{g/g}$), and (CE: 44.395 $\mu\text{g/g}$, MWE: 45.402 $\mu\text{g/g}$, OSE: 45.291 $\mu\text{g/g}$) successively. Also, the method of orbital shaker extraction was given a high amount of catechin hydrate (80.501 $\mu\text{g/g}$), and the lowest amount of caffeic (5.528 $\mu\text{g/g}$).

Contrarily, all three methods have no data detected any quercetin, fumaric acid, Phloridzin dihydrate, oleuropein, hydroxycinnamic acid, ellagic acid, myricetin, silymarin, kaempferol, salicylic acid, and butein.

Table 4.61. Identification and quantification of analysis specific phenolic compounds in five solvents with three methods of *Prunus microcarpa* extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	37.273	-	-	-	24.549	23.768	28.196	-	23.556	-	-	23.242	-	-	80.501
2	51.240	55.626	-	-	127.818	38.319	118.025	604.013	-	67.292	-	-	-	-	-
3	2.963	1.209	4.197	4.507	2.427	1.077	1.426	3.796	-	3.648	2.050	2.166	1.849	4.415	3.142
4	748.113	758.992	774.115	906.382	875.010	709.558	828.939	797.435	893.050	827.400	791.143	810.907	808.539	821.900	854.649
5	109.712	125.612	39.566	62.797	70.727	44.602	86.217	86.597	70.271	58.282	70.996	59.128	58.421	53.993	85.846
6	-	-	-	53.468	46.130	76.023	44.962	62.260	-	-	-	-	-	-	-
7	43.924	43.485	-	44.512	44.017	-	45.075	-	-	-	47.216	-	45.363	45.582	42.726
8	12.698	19.986	5.579	11.527	-	8.895	10.877	16.904	5.110	6.454	-	5.058	-	-	5.528
9	-	48.164	-	-	-	-	-	-	-	-	-	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
12	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	-	-	-	-	-	-	168.530	-	-	-	-	-	-	-	-
15	-	46.451	45.931	-	-	-	45.395	-	45.395	-	-	45.402	44.395	45.402	45.291
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	703.604	-	219.760	-	-	-	-	-
18	-	4.912	-	-	2.003	0.972	-	-	-	-	-	18.099	-	-	-
19	7.709	10.251	9.901	7.301	4.461	7.975	5.397	8.820	3.172	4.853	5.396	38.351	5.397	2.598	4.238
20	-	-	-	-	3.539	-	-	-	-	-	-	-	-	-	-
21	-	-	-	-	-	-	-	923.404	-	-	-	-	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1,4 aftokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H)

4.7.8. Identification and quantification of analysis specific phenolic compounds of *Silybum Marianum* seed extract by LC-MS/MS

The several phenolic compounds identified from twenty-five standards were identified and quantified for all three methods with five solvents for *Silybum Marianum* seed extract by liquid chromatography with mass spectrometry LC-MS/MS, as presented in Table (4.62).

The **distilled water** by all three methods was given seventeen kinds of phenolic acids by LC-MS/MS. The conventional method was obtained the highest amounts of hydroxycinnamic acid (852.037 µg/g), vanillic acid (760.224 µg/g), luteolin (733.665 µg/g), silymarin (5506.374), quercetin (476.308 µg/g), ellagic acid (306.441 µg/g), and caffeic acid (103.960 µg/g), also the method of conventional containing the specified amounts of acetohydroxamic acid (1.588 µg/g), resveratrol (70.722 µg/g), fumaric acid (65.909 µg/g), myricetin (56.431 µg/g), gallic acid (43.851 µg/g), and catechin hydrate (33.320 µg/g). While the lowest amounts of salicylic acid have been showed (CE: 3.030 µg/g), and hydroxybenzoic acid (CE: 8.281 µg/g) respectively.

On the other hand, the vanillic acid (MWE: 824.061 µg/g), silymarin (MWE: 620.350 µg/g) was detected in high quantities at microwave methods, whereas, the low amounts and limited amounts was detected caffeic acid (MWE: 5.556 µg/g), hydroxybenzoic acid (MWE: 5.333 µg/g), quercetin (MWE: 56.329 µg/g), and resveratrol (MWE: 52.919 µg/g) respectively. Hence, the high values of silymarin and vanillic acid were showed by way of orbital shaker (OSE: 561.757 µg/g), (OSE: 757.009 µg/g), while limited quantities and lowest values gallic acid 43.433, curmin (OSE: 44.870 µg/g), hydroxybenzoic acid (OSE: 5.494 µg/g), caffeic acid (OSE: 5.574 µg/g), and butein (OSE: 6.567 µg/g) respectively. Instead, the methods of microwave and orbital shaker no detected any data of catechin hydrate, acetohydroxamic acid, fumaric acid, Phloridzin dihydrate, oleuropein, hydroxycinnamic acid, ellagic acid, myricetin, curmin, kaempferol, salicylic acid, and butein.

Moreover, the demonstration of seventeen types of phenolic in (**ethanol: distilled water**) extract in all methods by LC-MS/MS. Results showed that the luteolin, silymarin, and quercetin contained the greatest amounts at the conventional method (CE: 3123.491 µg/g, CE: 29082.01 µg/g, and CE: 1311.375 µg/g). So, the hydroxycinnamic acid, vanillic acid, naringenin, kaempferol, myricetin showed high content in ethanol: distilled water as (978.535 µg/g, 637.389 µg/g, 198.104 µg/g, 188.960 µg/g, and 179.331 µg/g) respectively, at the conventional method.

Other compounds were identified and showed the limited amounts and lowest values of catechin hydrate (44.277 $\mu\text{g/g}$), resveratrol (51.364 $\mu\text{g/g}$), gallic acid (47.181 $\mu\text{g/g}$), fumaric acid 61.094 $\mu\text{g/g}$, Phloridzin dihydrate 53.039 $\mu\text{g/g}$, salicylic acid (5.775 $\mu\text{g/g}$), acetohydroxamic acid (3.03 $\mu\text{g/g}$), hydroxybenzoic acid (9.095 $\mu\text{g/g}$), and caffeic acid (15.342 $\mu\text{g/g}$). Further, the microwave method gave a big amount of silymarin 5787.578, and high amounts of vanillic acid, quercetin, luteolin, resveratrol, and hydroxycinnamic acid (MWE: 892.557, 132.964, 174.752, 127.507 $\mu\text{g/g}$, and 80.543 $\mu\text{g/g}$) respectively. While the myricetin, caffeic acid, and hydroxybenzoic acid were detected in minimum amounts at the microwave method. Another way of the orbital shaker was showed the highest quantities of silymarin (12006.661 $\mu\text{g/g}$), luteolin (1161.103 $\mu\text{g/g}$), vanillic acid (738.135 $\mu\text{g/g}$), and hydroxycinnamic acid (186.919 $\mu\text{g/g}$). Also, the myricetin, resveratrol, quercetin, phloridzin dihydrate, and fumaric acid have been the defined amounts (OSE: 84.556 $\mu\text{g/g}$, 55.240 $\mu\text{g/g}$, 51.494 $\mu\text{g/g}$, 47.218 $\mu\text{g/g}$, and 47.052), with the lowest amounts of it were acetohydroxamic acid, caffeic acid, kaempferol, hydroxybenzoic acid.

through all three methods in **ethanol** extract discovered the sixteen kinds of phenolic acids by LC-MS/MS. The phenolic compounds were silymarin (CE: 24529.834 $\mu\text{g/g}$), and luteolin (CE: 1676.075 $\mu\text{g/g}$), which had shown the greatest quantities in the conventional method. Also, the vanillic acid, hydroxycinnamic acid, quercetin, ellagic acid, naringenin, and myricetin were most plentiful at the conventional method (725.923 $\mu\text{g/g}$, 694.448 $\mu\text{g/g}$, 496.123 $\mu\text{g/g}$, 306.451 $\mu\text{g/g}$, 272.481 $\mu\text{g/g}$, and 199.390 $\mu\text{g/g}$) respectively, while the other compounds of resveratrol, curmin, gallic acid, catechin hydrate, and Phloridzin dihydrate were little abundant (58.442 $\mu\text{g/g}$, 46.451 $\mu\text{g/g}$, 44.305 $\mu\text{g/g}$, 29.863 $\mu\text{g/g}$, and 64.436 $\mu\text{g/g}$), with smallest amounts of caffeic acid and hydroxybenzoic acid. Thus, the microwave technique was containing high amounts of silymarin (969.642 $\mu\text{g/g}$), vanillic acid (930.684 $\mu\text{g/g}$), resveratrol (117.972 $\mu\text{g/g}$), quercetin (75.968 $\mu\text{g/g}$), and gallic acid (44.382 $\mu\text{g/g}$) respectively, whereas the minimum quantities of luteolin were appeared (25.042 $\mu\text{g/g}$), catechin hydrate (22.975 $\mu\text{g/g}$), hydroxycinnamic acid (17.324 $\mu\text{g/g}$), caffeic acid, (9.373 $\mu\text{g/g}$), hydroxybenzoic acid (3.699 $\mu\text{g/g}$), and acetohydroxamic acid (1.178 $\mu\text{g/g}$) respectively at microwave method. On the other side, the highest amount at way orbital shaker was silymarin (OWE: 6264.859 $\mu\text{g/g}$), and the most abundant of it has been showed vanillic acid (OSE: 827.374 $\mu\text{g/g}$), and kaempferol (OSE: 137.308 $\mu\text{g/g}$), while the limited amounts and lowest amount of it has been invetigated (resveratrol 78.683), (hydroxycinnamic acid 21.825 $\mu\text{g/g}$), (luteolin 44.849 $\mu\text{g/g}$), and (hydroxybenzoic acid 4.638 $\mu\text{g/g}$) respectively. Rather, in the way of an orbital shaker was

not detected any data of some phenolic as catechin hydrate, quercetin, fumaric acid, gallic acid, caffeic acid, Phloridzin dihydrate, ellagic acid, myricetin, and curmin, naringenin.

Also, the **(hexane: ethanol)** extract displayed fourteen kinds of phenolic compounds at all various methods. The methods conventional, and microwave, and orbital shaker have been obtained a maximum amount of silymarin (CE: 14953.13 $\mu\text{g/g}$, MWE: 8328.583 $\mu\text{g/g}$, OSE: 1909.230 $\mu\text{g/g}$) respectively. Hence, the vanillic acid, hydroxycinnamic acid, and resveratrol showed high quantities at all three methods (CE: 779.671 $\mu\text{g/g}$, MWE: 954.155 $\mu\text{g/g}$, OSE: 909.915 $\mu\text{g/g}$), (CE: 504.606 $\mu\text{g/g}$, MWE: 204.480 $\mu\text{g/g}$, OSE: 21.537 $\mu\text{g/g}$), and (CE: 62.991 $\mu\text{g/g}$, MWE: 63.101 $\mu\text{g/g}$, OSE: 78.759 $\mu\text{g/g}$) respectively. Whereas, the lowest amounts were detected of acetohydroxamic acid and hydroxybenzoic acid in all three ways. On the other hand, the conventional methods observed a maximum amount of luteolin (CE: 1246.343 $\mu\text{g/g}$), and high amounts of myricetin (CE: 111.399 $\mu\text{g/g}$), and quercetin (CE: 156.249 $\mu\text{g/g}$), also the limited amounts have been detected of naringenin (CE: 78.105), and gallic acid (CE: 45.017 $\mu\text{g/g}$), with the smallest amount of caffeic acid (CE: 11.603 $\mu\text{g/g}$), phloridzin dihydrate (CE: 8.965 $\mu\text{g/g}$). While the microwave way gave high and low amounts of luteolin (MWE: 241.217 $\mu\text{g/g}$), myricetin (MWE: 57.911 $\mu\text{g/g}$), quercetin (MWE: 61.537 $\mu\text{g/g}$), catechin hydrate (MWE: 25.092 $\mu\text{g/g}$), naringenin (MWE: 5.498 $\mu\text{g/g}$). Further, the Phloridzin dihydrate (OSE: 48.630 $\mu\text{g/g}$), and gallic acid (OSE: 44.075 $\mu\text{g/g}$), were displayed in way of an orbital shaker.

Analysis of six kinds of phenolic compounds in **hexane** extract with many methods was supported out by liquid chromatography with mass spectrometry (LC-MS/MS). The top quantity of it was vanillic acid (CE 831.196 $\mu\text{g/g}$, MWE: 368.027 $\mu\text{g/g}$, OSE: 838.983 $\mu\text{g/g}$) by all three methods. Hence, the resveratrol acid was showed a limited amount in all three methods (CE: 78.894 $\mu\text{g/g}$, MWE: 61.186 $\mu\text{g/g}$, OSE: 55.354 $\mu\text{g/g}$). Though it has been getting small quantities of hydroxybenzoic acid, acetohydroxamic acid, and caffeic acid were detected. As an alternative, there was no data detected catechin hydrate, quercetin, fumaric acid, gallic acid, Phloridzin dihydrate, oleuropein, hydroxycinnamic acid, ellagic acid, myricetin, curmin, naringenin, kaempferol, salicylic acid, hydroxybenzoic acid, butein, and luteolin at all three methods. On the other hand, the low amount of caffeic acid (CE: 9.961 $\mu\text{g/g}$) in the conventional method, and with the high amount of silymarin (MWE: 93.543 $\mu\text{g/g}$) in the microwave, method were detected.

No research has been conducted on *Silybum Marianum* seed extracts. However, there have been a variety of studies on the phenolic compounds in other techniques (Rodriguez *et al.* 2018) has also suggested that the extract of *silybum marianum* seeds contains the maximum

amount of (silybin B 7.434 mg/g DW) analyzed by HPLC-UV. Our finding inline with (Ghafor *et al.* 2014) reported, that The phenolic and flavonoids compounds of kaempferol, luteolin, thymoquinone, gentisic acid, coumaric acid, ferulic acid, sinapic acid, chlorogenic acid were showed in the Stems of *Silybum Marianum* through using the HPLC method.



Table 4.62. Identification and quantification of analysis specific phenolic compounds in five solvents with three methods of *Silybum Marianum* seed extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C /E: W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	33.320	-	-	44.277	-	-	29.863	22.975	-	-	25.092	-	-	-	-
2	476.308	56.329	-	1311.37	132.964	51.494	496.123	75.968	-	156.249	61.537	-	-	-	-
3	1.588	-	-	3.038	-	2.101	-	1.178	-	3.133	1.586	4.952	3.923	1.640	3.021
4	760.224	824.061	757.009	637.389	892.557	738.135	725.923	930.684	827.374	779.671	954.155	909.915	831.196	368.027	838.983
5	70.722	52.919	-	51.364	127.507	55.240	58.442	117.972	78.683	62.991	63.101	78.759	78.894	61.186	55.354
6	65.909	-	-	61.094	-	47.052	-	-	-	-	-	-	-	-	-
7	43.851	-	43.433	47.181	-	-	44.305	44.382	-	45.017	-	44.075	-	-	-
8	103.960	5.556	5.574	15.342	5.444	7.170	6.286	9.373	-	11.603	-	-	9.961	-	-
9	-	-	-	53.039	-	47.218	64.436	-	-	58.965	48.630	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	852.037	-	-	978.535	80.543	186.919	694.448	17.324	21.825	504.606	204.480	21.537	-	-	-
12	306.441	-	-	-	-	-	306.451	-	-	-	-	-	-	-	-
13	56.431	-	-	188.960	25.807	84.556	199.390	-	-	111.399	57.911	-	-	-	-
14	5506.374	620.350	561.757	29082.0	5787.57	12006.66	24529.8	969.642	6264.859	14953.13	8328.58	1909.230	-	93.543	-
15	-	-	44.870	-	-	-	46.451	-	-	-	-	-	-	-	-
16	-	-	-	198.104	-	-	272.481	-	-	78.105	5.498	-	-	-	-
17	-	-	-	179.331	-	13.210	-	-	137.308	-	-	-	-	-	-
18	3.030	-	-	5.775	-	-	-	-	-	-	-	-	-	-	-
19	8.281	5.333	5.494	9.095	4.681	3.993	6.489	3.699	4.638	6.124	4.074	4.423	6.548	5.204	4.364
20	-	-	6.567	-	-	-	-	-	-	-	-	-	-	-	-
21	733.665	-	-	3123.49	174.752	1161.103	1676.07	25.042	44.849	1246.343	241.217	-	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1,4 aetokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

4.7.9. Identification and quantification of analysis specific phenolic compounds of *Pistacia eurycarpa* Yale extract by LC-MS/MS

It was revealed that the solvent extracts in all three methods of *Pistacia eurycarpa* Yale give many compounds through using 25 standard compounds by LC-MS/MS, as displayed in Table (4.63).

The results in **distilled water** to be identified twelve compounds at all three ways, the luteolin was observed maximum amount in all methods (CE: 37868.688 µg/g, MWE: 3533.255 µg/g, OSE: 13225.072 µg/g). Whereas, a high amount has been detected of vanillic acid CE: (797.780 µg/g, MWE: 306.911 µg/g, OSE: 764.272 µg/g), kaempferol (CE: 668.318 µg/g, MWE: 221.570 µg/g, OSE: 699.787 µg/g) in all methods. However, the limited amount and low amount of resveratrol, hydroxybenzoic acid were identified (CE: 74.964 µg/g, MWE: 59.180 µg/g, OSE: 63.045 µg/g), and (CE: 4.633 µg/g, MWE: 2.738 µg/g, OSE: 5.454 µg/g) respectively. On the other hand, the other compounds of quercetin, gallic acid, catechin hydrate, Phloridzin dihydrate, and hydroxycinnamic acid were detected in the conventional method (CE: 493.615 µg/g, 360.458 µg/g, 149.917 µg/g, 45.944 µg/g, and 18.168 µg/g) respectively, while the microwave methods detected gallic acid, catechin hydrate, and acetohydroxamic acid. So, fumaric acid and quercetin were obtained by the orbital shaker method (OSE: 49.385 µg/g, and 417.556 µg/g).

The fourteen compounds of phenolic were gained in (**ethanol: distilled water**) extract using three methods. The greatest amounts found were the luteolin and kaempferol at all methods (45196.51 µg/g, 44302.86 µg/g, 45085.90 µg/g), and (1493.649 µg/g, 3001.281 µg/g, 2357.353 µg/g). Also the vanillic acid, quercetin, gallic acid, Catechin hydrate, and butein obtained high values in all three methods.(CE: 790.452 µg/g, MWE: 973.745 µg/g, OSE: 813.926 µg/g), (CE: 1109.553 µg/g, MWE: 560.707 µg/g, OSE: 533.187 µg/g), (CE: 194.943 µg/g, MWE: 122.094 µg/g, MSE: 154.630 µg/g), (CE: 116.134 µg/g, MWE: 84.826 µg/g, OSE: 105.154 µg/g), and (CE: 187.489 µg/g, MWE: 51.772 µg/g, OSE: 30.308 µg/g) respectively, while the resveratrol and hydroxybenzoic acid were detected amounts at all methods. Other compounds found high values in the way conventional method of ellagic acid and silymarin have illustrated (264.856 µg/g, and 62.761 µg/g). Also, the high amounts were showed ellagic acid and fumaric acid in the way of the orbital shaker. But, the lowest amounts were acetohydroxamic acid (OSE: 1.568 µg/g) in the orbital method, hydroxycinnamic acid (CE: 3.400 µg/g) in the conventional method, and acetohydroxamic acid (MWE: 3.480 µg/g), in microwave method respectively.

Otherwise, any caffeic acid, Phloridzin dihydrate, oleuropein, hydroxycinnamic acid, myricetin, curmin, naringenin, and salicylic acid were not detected in all three ways.

The phenolic compounds obtained fourteen kinds by three different methods in **ethanol** extract. The luteolin, kaempferol, and quercetin were getting the greatest amounts in different methods (CE: 83461.207 $\mu\text{g/g}$, MWE: 97336.19 $\mu\text{g/g}$, OSE: 70523.74 $\mu\text{g/g}$), (CE: 5080.096 $\mu\text{g/g}$, MWE: 6691.929 $\mu\text{g/g}$, OSE: 3763.944 $\mu\text{g/g}$), (CE: 1521.190 $\mu\text{g/g}$, MWE: 2761.181 $\mu\text{g/g}$, OSE: 1139.338 $\mu\text{g/g}$) respectively. So, it contains high quantities and limited amounts of vanillic acid, gallic acid, butein, resveratrol, and catechin hydrate were (CE: 760.630 $\mu\text{g/g}$, MWE: 797.694 $\mu\text{g/g}$, OSE: 825.422 $\mu\text{g/g}$), (CE: 180.211 $\mu\text{g/g}$, MWE: 245.721 $\mu\text{g/g}$, OSE: 98.198 $\mu\text{g/g}$), (CE: 29.660 $\mu\text{g/g}$, MWE: 152.142 $\mu\text{g/g}$, OSE: 60.747 $\mu\text{g/g}$), (CE: 45.717 $\mu\text{g/g}$, MWE: 105.444 $\mu\text{g/g}$, OSE: 8.371 $\mu\text{g/g}$), and (44.357 70.926 86.483), with the minimum amount of hydroxybenzoic acid at all three methods. However, the ellagic acid and silymarin (CE: 315.291 $\mu\text{g/g}$, MWE: 413.014 $\mu\text{g/g}$), and (CE: 81.324 $\mu\text{g/g}$, MWE: 411.400 $\mu\text{g/g}$) in both methods as conventional and microwave were showed high amounts. While the hydroxycinnamic acid and acetohydroxamic acid in conventional methods, acetohydroxamic acid in the orbital shaker, and the fumaric acid in the microwave method were detected, respectively. Instead, each of caffeic acid, Phloridzin dihydrate, oleuropein, myricetin, curmin, naringenin, and salicylic acid was not discovered at all three methods.

Also, the solvent of (**hexane: ethanol**) extract was given fifteen compounds of phenol by three methods. The detected topmost amounts of luteolin (CE: 72384.36 $\mu\text{g/g}$, MWE: 16853.66 $\mu\text{g/g}$, OSE: 41869.82 $\mu\text{g/g}$), and kaempferol (CE: 5241.874 $\mu\text{g/g}$, MWE: 1256.109 $\mu\text{g/g}$, OSE: 2454.117 $\mu\text{g/g}$) were recorded in three methods. Though the quercetin, vanillic acid, and gallic acid found high values in all methods (CE: 1950.079 $\mu\text{g/g}$, MWE: 276.044 $\mu\text{g/g}$, OSE: 515.186 $\mu\text{g/g}$), (CE: 725.224 $\mu\text{g/g}$, MWE: 815.573 $\mu\text{g/g}$, OSE: 815.076 $\mu\text{g/g}$), and (CE: 269.223 $\mu\text{g/g}$, $\mu\text{g/g}$ 52.193 $\mu\text{g/g}$, OSE: 68.316 $\mu\text{g/g}$) respectively, also all three methods noticed limited amounts of resveratrol and low amount hydroxybenzoic acid. Additionally, the conventional way displayed high amounts of ellagic acid (CE: 310.558 $\mu\text{g/g}$), hydroxycinnamic acid (CE: 32.850 $\mu\text{g/g}$), and butein (CE: 34.099 $\mu\text{g/g}$), and low amounts of caffeic acid (CE: 6.311 $\mu\text{g/g}$, salicylic acid (CE: 4.025 $\mu\text{g/g}$) respectively. Further, the microwave technique has given the high amount of silymarin (MWE: 102.256 $\mu\text{g/g}$), and small amounts of acetohydroxamic acid (MWE: 2.101 $\mu\text{g/g}$), with the amount of butein in the orbital shaker method. Any fumaric acid, phloridzin dehydrate, oleuropein, myricetin, curmin, and naringenin were no identified in all three methods.

Ultimately, the eight phenolic compounds were discovered in **hexane** extract by LC-MS/MS in all three ways. The highest amounts of phenolic were identified in hexane extract which was luteolin (CE: 858.402 $\mu\text{g/g}$, MWE: 384.642 $\mu\text{g/g}$, OSE: 1200.966 $\mu\text{g/g}$), and vanillic acid (CE: 767.026 $\mu\text{g/g}$, MWE: 852.850 $\mu\text{g/g}$, OSE: 810.452 $\mu\text{g/g}$). Too, the specified amount and least amount of it has been resveratrol (CE: 59.320, MWE: 35.984, OSE: 36.238 $\mu\text{g/g}$) and hydroxybenzoic acid (CE: 4.355 $\mu\text{g/g}$, MWE: 2.133 $\mu\text{g/g}$, OSE: 3.278 $\mu\text{g/g}$) respectively, in all three ways. Besides, the gallic acid 42.678 $\mu\text{g/g}$, quercetin 41.697 $\mu\text{g/g}$, acetohydroxamic acid 1.207 $\mu\text{g/g}$, and salicylic acid 0.929 $\mu\text{g/g}$, were detected in the conventional method. But the acetohydroxamic acid 2.075 $\mu\text{g/g}$, in the microwave method, and the gallic acid in the orbital shaker way were detected. While, the methods of conventional, microwave, and orbital shaker were not recorded any data of catechin hydrate, caffeic acid, Phloridzin dihydrate, oleuropein, hydroxycinnamic acid, ellagic acid, myricetin, silymarin, curmin, naringenin, kaempferol, and butein.

There are no any investigations on *Pistacia eurycarpa* Yale extracts. Although there were various studies about the phenolic compounds that agree with our study (Amel *et al.* 2016) reported that gallic acid, ascorbic acid, coumaric acid, catechin, and quercetin were discovered for the first time in the leaves of *Pistacia lentiscus*, as were ferulic acid, vanillic acid, and naringenin.. In another study,(Kafkas *et al.* 2007) the report suggests that the GC-MS found several compounds of fatty acid in the *Pistacia eurycarpa* Yale, including 18:2 (9, 12; 52.2 percent) methyl oleate (17.7%) and methyl palmitate (16.7 %), methyl stearate (4.3 percent), methyl palmitoleate (2.0%), 11-eicosenic acid methyl ester (1.4 %), and heptadecene-(8)-carbonic acid (1.3 %).

Table 4.63. Identification and quantification of analysis specific phenolic compounds in five solvents with three methods of *Pistacia eurycarpa* Yale extract by LC-MS/MS

NO	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E:W (µg/L)	M/E:W (µg/L)	OS/E: W (µg/L)	C/E (µg/L)	M/E (µg/L)	OS/E (µg/L)	C/H: E (µg/L)	M/H: E (µg/L)	OS/H: E (µg/L)	C/H (µg/L)	M/H (µg/L)	OS/H (µg/L)
1	149.917	24.119	-	116.134	84.826	105.154	29.660	152.142	60.747	31.419	-	23.809	-	-	-
2	493.615	-	417.556	1109.55	560.707	533.187	1521.190	2761.18	1139.338	1950.079	276.044	515.186	41.697	-	-
3	-	8.918	-	-	3.480	1.568	3.978	-	2.572	-	2.101	-	-	1.207	2.075
4	797.780	306.911	764.272	790.452	973.745	813.926	760.630	797.694	825.422	725.224	815.573	815.076	767.026	852.850	810.452
5	74.964	59.180	63.045	78.662	58.348	59.004	44.357	70.926	86.483	58.771	94.525	43.535	59.320	35.984	36.238
6	-	-	49.385	-	-	55.246	N.D	51.158	-	-	-	-	-	-	-
7	360.458	58.401	-	194.943	122.094	154.630	180.211	245.721	98.198	269.223	52.193	68.316	42.678	-	61.245
8	-	-	-	-	-	-	-	-	-	6.311	-	-	-	-	-
9	45.944	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	18.168	-	-	3.400	-	-	0.615	-	-	32.850	-	-	-	-	-
12	-	-	-	264.856	-	317.604	315.291	413.014	-	310.558	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	-	-	-	62.761	-	-	81.324	411.400	-	-	102.256	-	-	-	-
15	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
17	668.318	221.570	699.787	1493.64	3001.28	2357.353	5080.096	6691.92	3763.944	5241.874	1256.10	2454.117	-	-	-
18	-	-	-	-	-	-	-	-	-	4.025	-	-	0.929	-	-
19	4.633	2.738	5.454	6.221	4.180	5.796	6.755	6.808	8.435	8.140	4.917	7.60	4.355	2.133	3.278
20	34.003	-	-	187.489	51.772	30.308	45.717	105.444	8.371	34.099	-	2.956	-	-	-
21	37868.68	3533.255	13225.07	45196.5	44302.8	45085.90	83461.20	97336.1	70523.74	72384.36	16853.6	41869.82	858.402	384.642	1200.96
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

¹RT: Retention time. ²Parent ion (*m/z*): Molecular ions of the standard compounds (mass to charge ratio). ³MRM (Multi reaction module) MS₂ (CE): fragments for the related molecular ions (CE refers to related collision energies of the fragment ions). ⁴R²: coefficient of determination. SD: relative standard deviation. ⁵Quantification (mg analyte/g extract): values in g/g (w/w) of plant extract. ⁶N.D. not detected. ⁷Specific phenolic plant extract. 1: Catechin hydrate. 2: Quercetin. 3: Acetohydroxamic acid. 4: vanillic acid. 5: Resveratrol. 6: Fumaric acid. 7: Gallic acid. 8: caffeic acid. 9: Phloridzin dehydrate. 10: Oleuropein. 11: hydroxycinnamic. 12: Ellagic acid. 13: Myricetin. 14: Silymarin. 15: Curmin. 16: Naringenin. 17: Kaempferol. 18: salicylic acid. 19: Hydroxybenzoic acid. 20: Butein. 21: luteolin. 22: alizarin. 23: Thymoquinone. 24: 2-hydroxy 1,4 aftarokiniton. 25: Protocatechuic acid ethyl ester. ⁸Method technique: Conventional Extraction (C/E). Microwave Extraction (MW/E). Orbital Shaker Extraction (OS/E). Solvent: Distilled water (DW). Distilled water: Ethanol (DW: E). Ethanol (E). Hexane: Ethanol (H: E) and Hexane (H).

Generally, in current study, the first analysis dealing with the identification and characterization of specific phenolic components and no study has been conducted on our plant extracts, in five solvents extracts (distilled water, ethanol: distilled water, ethanol, hexane: ethanol, and hexane) by all three methods by LC-MS/MS. Because of the great sensitivity and selectivity, liquid chromatography coupled to ESI-MS (LC ESI-MS/MS) has received considerable attention for the determination of phenolic acids and flavonoids, the negative ionization method was preferred in the modern study (Ertas *et al.* 2015; Sandín-España *et al.* 2016). Hence, the HPLC-MS-MS and MS-MS analysis can be used to further characterize the structure. It can also be used to assess the presence of previously known substances, minimizing the amount of time spent isolating them (Constant and Beecher 1995).

Also, our plants indicated the high and significant amounts of specific phenolic compounds in all five solvents by three methods. However, from the twenty-five standards of phenolic compounds, our plants detected only twenty kinds of compounds. Accordingly, our plants were containing the highest amounts of phenolic especially (vanillic acid, resveratrol, gallic acid, acetohydroxamic acid, luteolin, naringenin, quercetin, catechin hydrate, silymarin, salicylic acid, Phloridzin dihydrate, kaempferol, and caffeic acid. Rather, other phenolic compounds were not detected in all plants as alizarin, thymoquinone, hydroxy-1,4-naphthoquinone, and protocatechuic acid ethyl ester. according to the study (Nugroho *et al.* 2009), had mentions that the presence of phenolic compounds of plants such as quinic acid in methanol extract solvents can play an important role in increasing its primary antioxidant activity. Upon the findings, Tannin content tends to be associated with average annual temperature. Because of variations in root structure, there is extremely variance in phenolic composition within and within plants. Furthermore, the phenolic composition and therefore the antioxidant potential of plants belonging to the same species may vary, because of variations in environmental conditions, plant physiopathology, water intensity, and soil quality, among other factors (Zielinski *et al.* 2014). To our conclusion, none of these previous studies have used LC-MS/MS to determine the phytochemical compositions of our plants.

4.8. Analysis of mineral components of all Plants by Ion Chromatography (IC)

Ion chromatography (IC) is a preferred and commonly used technique for the determination of inorganic and small organic anions occurring or created under the conditions used, and that used in a wide range of samples for the determination of ionic compounds (Kubáň *et al.* 2005; Small *et al.* 1975). The focus on the ion chromatographic application of inorganic by the following fields are mainly determined: chemistry of power plants, Environmental analysis, pharmaceuticals, biotechnology, semiconductors Industry, food and beverages, metal manufacturing, mining, agriculture, electroplating. Industry of pulp and paper (Michalski 2009), Our study, the ion chromatography applications have been used for representative isolation of the regular solution of the identified fifteen mineral ion compounds (F^- , ClO_2^- , BrO_3^- , Cl^- , NO_2^- , Br^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}) by using different methods, conventional extraction and microwave-assisted extraction with five different solvents in all plants.

4.8.1. Analysis of mineral components of *Eremurus Spectabilis Bieb* extract

From Table (4.64), and Figure (6.1), the conventional extraction methods with five solvents of *Eremurus Spectabilis Bieb* have detected the six mineral compounds as F^- , Cl^- , Na^+ , NH_4^+ , K^+ , and Ca^{+2} , respectively, however, other ions as (NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ and Mg^{2+}) was detected from two to four solvents. therefore, the Ions of K^+ , NH_4^+ , and Cl^- was detected great significant values as solvents, distilled water (10.287) (4.670) (1.308) ppm, ethanol: distilled water (10.320), (4.137) (1.531) ppm, ethanol (2.798) (1.272) (1.904) ppm, hexane: ethanol (1.018), (0.684) (1.338) ppm, and hexane (1.608) (0.412) (0.643) ppm, respectively. in comparison with the mineral ions of F and Na^+ was detected lowest significant quantities in all solvents.

Secondly, the mineral components with the method of microwave extraction from 15 mineral compounds were detected ten ions in five solvents as (F^- , Cl^- , NH_4^+ , K^+ , Mg^{2+} , Ca^{+2} , NO_3^- , PO_4^{3-} , SO_4^{2-} , and Li^+), respectively. The ion of mineral compounds with all solvents was identified highly significant values of K^+ , NH_4^+ , SO_4^{2-} , and Cl^- , as followed DW (8.985), (3.807) (2.270) (1.361) ppm, E: DW (8.165, 0.935, 2.139, and 1.102) ppm, E (9.393, 1.136, 2.477, and 1.766) ppm, H: E (3.824, 1.017, 3.065, and 2.512) ppm, and (0.302, 0.050, 0.928, and 0.543) ppm. Respectively, in compared with the lowest ion components as Ca^{2+} , Mg^{2+} , and F^- , that shown with all solvents. Furthermore, other ions were detected by some solvent NO_3^- , PO_4^{3-} ,

Li^+ , Mg^{2+} , and Ca^+ . Whereas, the ion of Na^{2+} was not detected in solvent (distilled water and ethanol: distilled water).

In addition, the mineral compounds by using the method of orbital shaker extraction were identified eleven ions in all solvents. The ions of K^+ , NH_4^+ , PO_4^{3-} , SO_4^{2-} , Cl^- were detected in great significant quantities with all solvents. Furthermore, the solvents of (distilled water) and (ethanol: distilled water) were identified the highest values as K^+ (14.872, 7.673 ppm), NH_4^+ (5.263, 3.552 ppm), PO_4^{3-} (5.869, 2.680 ppm), SO_4^{2-} (3.835, 3.342 ppm), and Cl^- (2.217, 1.652 ppm), in comparison with lowest values of ions (NO_3^- , Mg^{2+} , F^- , and Li^+ ,) respectively. However, the ion of Na^+ was not detected in solvent (ethanol: distilled water) and hexane. This current report is the first experience to found mineral compounds by techniques of ion chromatography in *Eremurus Spectabilis Bieb*

Generally, our study indicates that the ion chromatography of by method of orbital shaker extraction, in solvents distilled water, and ethanol: distilled water of *Eremurus Spectabilis Bieb*, was significant analysis to given the ions, in comparison with techniques of (conventional and microwave), and other solvents. Hence, the *Eremurus Spectabilis Bieb* was a rich source of K^+ , Ca^{2+} ions, our study approval with the report (Tosun, Ercisli, Ozer, Polat, *et al.* 2012; Arslan and Erbil 2018), and (Bircan and Kırbağ 2015), reported that the *Eremurus Spectabilis Bieb*, could be a valuable source of antioxidants, phenolic and minerals of K^+ , Ca^{2+} , Mg^{2+} , Fe^{2+} Cu^+ contents, flavonoids, vitamin K, vitamin D, and polysaccharides, were found higher than in some other commonly used vegetables.

Table 4.64. Analysis of mineral components of *Eremurus Spectabilis Bieb* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.521	0.579	0.581	0.493	0.535	0.529	0.536	0.570	0.499	-	0.838	0.517	1.818	0.943	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	1.308	1.531	1.904	1.338	0.643	1.361	1.102	1.766	2.512	0.543	2.217	1.652	2.325	1.336	1.061	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	-	-	0.432	-	-	0.417	0.000	0.434	0.460	-	0.456	0.000	0.467	-	-	mg/L
8.	PO ₄ ³⁻	2.931	2.604	-	-	-	3.074	4.179	4.036	1.585	-	5.869	2.680	1.020	2.114	1.066	mg/L
9.	SO ₄ ²⁻	2.114	2.726	0.604	-	-	2.270	2.139	2.477	3.065	0.928	3.835	3.342	1.418	2.957	2.226	mg/L
10.	Li ⁺	0.000	0.218	-	-	-	0.203	-	-	0.454	0.234	0.248	0.219	0.308	0.409	0.235	mg/L
11.	Na ⁺	0.056	0.212	0.403	0.263	0.008	-	-	0.123	0.559	0.168	0.086	-	0.388	0.084	-	mg/L
12.	NH ₄ ⁺	4.670	4.137	1.272	0.684	0.412	3.807	0.935	1.136	1.017	0.050	5.263	3.552	1.017	0.546	1.006	mg/L
13.	K ⁺	10.287	10.320	2.798	1.018	1.608	8.985	8.165	9.393	3.824	0.302	14.872	7.673	3.463	3.231	3.374	mg/L
14.	Mg ²⁺	0.541	0.404	0.000	0.000	0.147	0.516	0.463	0.398	0.195	-	0.737	0.913	0.168	0.240	0.455	mg/L
15.	Ca ²⁺	0.748	0.512	0.168	0.136	0.201	0.425	0.708	0.413	0.569	-	0.982	11.176	0.423	0.472	0.949	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.2. Analysis of mineral components of *Prosopis farcta* extract

According to the *Prosopis farcta*, the method of conventional extraction has detected eight significant mineral compounds as ions (K^+ , PO_4^{3-} , SO_4^{2-} , Cl^- , Ca^{2+} , Mg^{2+} , NH_4^+ , F^- , and Li^+) from fifteen in all solvents, respectively. Furthermore, the solvent of distilled water was observed highest quantity of ions (K^+ 6.272 ppm), (PO_4^{3-} 2.289 ppm), (SO_4^{2-} 2.048 ppm), (Cl^- 1.842 ppm), (Ca^{2+} 0.798 ppm), (Mg^{2+} 0.736 ppm), (NH_4^+ 0.627 ppm), and (F^- 0.490 ppm), in compared with solvent of hexane was lowest values (NH_4^+ 0.073 ppm), (Mg^{2+} 0.308 ppm), (Ca^{2+} 0.455 ppm), (F^- 0.503 ppm), (Cl^- 0.747 ppm), (SO_4^{2-} 0.827 ppm), (PO_4^{3-} 0.829 ppm), and (K^+ 1.377 ppm). On the other hand, the ion of Na^+ was not detected in solvent (ethanol: distilled water) and hexane, while has been detected with other solvents.

Also, the microwave technique was observed ten mineral compounds of ions of (K^+ , SO_4^{2-} , PO_4^{3-} , Cl^- , Ca^{2+} , Mg^{2+} , Li^+ , NH_4^+ , Na^+ and, F^-) in all various solvents. Moreover, the ions of (K^+ 7.513 ppm), (Cl^- 1.894 ppm), (F^- 0.595 ppm), (Mg^{2+} 0.544 ppm), (NH_4^+ 0.471 ppm), (Li^+ 0.313 ppm), was observed highest quantities in solvent ethanol respectively, while the ions of SO_4^{2-} 2.383 ppm, PO_4^{3-} 1.803 ppm, and Ca^{2+} 0.736 ppm, was high values in solvent of distilled water, respectively. Therefore, all the mineral ions have been showed lower value with hexane and excepting of ion F^- . Otherwise, the Ion of Na^+ was not detected with solvents of (distilled water and hexane), whereas, that is shown in other solvents.

The orbital shaker method was showed nine mineral ions (K^+ , SO_4^{2-} , Cl^- , Ca^{2+} , Mg^{2+} , PO_4^{3-} , F^- , NH_4^+ , and, Li^+) by the Ion chromatography. However, the ions of (K^+ 6.009 ppm), (SO_4^{2-} 3.124 ppm), (Cl^- 2.404 ppm), (Ca^{2+} 3.610 ppm), (PO_4^{3-} 2.213 ppm), (Mg^{2+} 1.053 ppm), (NH_4^+ 0.387 ppm), and (Li^+ 0.237 ppm) in all solvents respectively, in (ethanol: distilled water v/v) was a higher value than comparing with other solvents. Thus, the ions of (Cl^- , Ca^+ , SO_4^{2-} , Mg^{2+} , NH_4^+ , and, Li^+) were the lowest values viewed with the solvent of hexane, except for ions (PO_4^{3-} and F^-), because the ions of (PO_4^{3-} and F^-) had high quantities in solvent of hexane. On the other hand, the Na^+ ion was not detected in all solvents by using the orbital shaker method, illustrated in Table (4.65), and Figure (6.2).

This research was showed that the microwave technique in all solvents was a significant method to identify mineral compounds from *Prosopis farcta*, In comparison with other methods.

Table 4.65. Analysis of mineral components of *Prosopis farcta* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H:E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc.(pp m)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.490	0.545	0.580	0.561	0.503	0.518	0.516	0.595	0.637	0.614	0.509	0.614	0.550	0.618	0.520	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	1.842	1.251	2.191	1.584	0.747	1.343	1.660	1.894	1.390	0.804	1.186	2.404	1.328	1.425	0.847	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
8.	PO ₄ ³⁻	2.289	1.152	0.656	-	0.829	1.803	1.669	1.736	0.758	1.313	0.775	2.213	0.145	0.749	0.949	mg/L
9.	SO ₄ ²⁻	2.048	1.289	1.387	0.594	0.827	2.383	2.142	2.218	1.602	1.511	1.672	3.124	0.703	1.505	0.881	mg/L
10.	Li ⁺	0.271	0.230	0.309	-	0.213	0.300	0.292	0.313	0.299	0.253	0.206	0.237	0.223	0.226	0.217	mg/L
11.	Na ⁺	0.067	-	0.331	0.303	-	-	0.011	0.188	0.314	-	-	-	-	-	-	mg/L
12.	NH ₄ ⁺	0.627	0.380	0.772	0.651	0.073	0.455	0.441	0.471	0.451	0.126	0.202	0.387	0.392	0.369	0.124	mg/L
13.	K ⁺	6.272	3.395	1.694	1.029	1.377	3.857	4.524	7.513	1.029	1.688	2.111	6.009	1.078	1.683	1.907	mg/L
14.	Mg ²⁺	0.736	0.394	0.196	0.156	0.308	0.458	0.447	0.544	0.177	0.273	0.560	1.053	0.179	0.458	0.291	mg/L
15.	Ca ²⁺	0.798	0.449	0.304	0.173	0.455	0.736	0.417	0.512	0.238	0.396	1.049	3.610	0.247	0.699	0.385	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.3. Analysis of mineral components of *Capparis spinosa* extract

The ion chromatography was detected twelve mineral compounds from *Capparis spinosa*, in Table (4.66)M and Figure (6.3), as ions (F^- , BrO_3^- , Cl^- , Br^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), by using the different techniques and with five solvents. The technique of conventional extraction in all solvents was given three ions as (NO_3^- , NH_4^+ and K^+), whereas had detected the eight mineral compounds (BrO_3^- , Cl^- , Br^- , PO_4^{3-} , SO_4^{2-} , Na^+ , Mg^{2+} , and Ca^{2+}), not in all solvent. Thus, the ions of (NH_4^+ 25.091 ppm, NO_3^- 5.475 ppm, Br^- 1.784 ppm, and BrO_3^- 0.585 ppm), was detected in significant quantities in solvent of ethanol: distilled water, and the ions of (SO_4^{2-} 16.219 ppm, K^+ 14.459 ppm, Ca^{2+} 1.328 ppm, NH_4^+ 1.261 ppm, and F^- 0.694 ppm) in solvent of distilled water. in compared with the other solvents, respectively. on the other hand, the ion of Na^+ was not detected in solvents (distilled water, hexane: ethanol, and hexane), and the ion of Li^+ , in solvent of (ethanol: distilled water).

Additionally, ion chromatography was discovered eleven mineral compounds (SO_4^{2-} , K^+ , PO_4^{3-} , NH_4^+ , NO_3^- , Li^+ , Na^+ , F^- , Mg^{2+} , Cl^- , and Ca^{2+}), in way technique of microwave extraction in all solvents. Therefore, the microwave methods identified 7 ions of SO_4^{2-} , K^+ , PO_4^{3-} , NH_4^+ , Ca^{2+} , NO_3^- , and Mg^{2+} in all solvents, generally the solvents of (distilled water), and (ethanol: distilled water) was given the high significant values in ions (SO_4^{2-} 21.399, 14.867 ppm), (K^+ , 18.303, 16.639 ppm), (PO_4^{3-} 5.085, 4.485 ppm), (NH_4^+ 2.000, 1.535 ppm), (Ca^{2+} 1.819, 0.693 ppm), (NO_3^- 1.367, 1.393 ppm), and (Mg^{2+} 1.098, 0.925 ppm), although, (distilled water) is a better solvent than contrasting it with other solvents. However, the hexane solvent was lower values for all ions, while not data detected Na^+ ion comparing with other solvents.

In this study, the method of orbital shaker extraction was identified 10 mineral compounds of ion (K^+ , SO_4^{2-} , PO_4^{3-} , NO_3^- , F^- , NH_4^+ , Mg^{2+} , Cl^- , and Li^+), in all solvents. the ions (K^+ , SO_4^{2-} , and PO_4^{3-}), had significant quantities in all solvents. Furthermore, the solvent of (ethanol: distilled water) was given the highest values of ions K^+ (18.369 ppm), SO_4^{2-} (14.186 ppm), PO_4^{3-} (3.804 ppm), NO_3^- (1.706 ppm), Mg^{2+} (1.015 ppm), Cl^- (0.579 ppm), and Li^+ (0.281 ppm), respectively. Whereas, the solvent of distilled water was given significant values of ions (F^- 1.387 ppm, NH_4^+ 0.722 ppm, and Ca^{2+} 0.754 ppm), with other solvents. Contrarily, the solvents (hexane) and (hexane: ethanol), were given low ions of (F^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , NH_4^+ , Mg^{2+}) and (Cl^- , Li^+ , and Ca^{2+}), respectively. on the other hand, the ion Na^+ was not detected in all solvents.

In the current study, all techniques are best and significant of *Capparis spinosa*, to identify the mineral compounds in solvents of (ethanol: distilled water),(distilled water), and other solvents by ion chromatography.



Table 4.66. Analysis of mineral components of *Capparis spinose* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.694	-	0.598	0.522	0.523	0.898	0.778	0.538	0.533	0.503	1.387	0.670	0.513	0.539	0.504	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	0.585	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	0.538	-	0.556	0.533	0.542	0.632	0.556	0.534	0.560	0.674	0.547	0.579	0.931	0.529	0.681	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	1.784	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	1.116	5.475	1.197	0.938	0.505	1.367	1.393	0.784	0.945	0.465	1.101	1.706	0.530	0.797	0.433	mg/L
8.	PO ₄ ³⁻	3.392	22.275	-	-	0.982	5.085	4.485	0.000	0.000	0.736	2.938	3.804	-	1.646	0.769	mg/L
9.	SO ₄ ²⁻	16.219	-	0.830	0.793	2.661	21.399	14.867	0.709	0.693	1.479	10.950	14.186	1.840	4.884	1.742	mg/L
10.	Li ⁺	-	-	-	-	-	-	0.200	0.194	0.000	0.197	0.000	0.281	0.201	0.193	0.207	mg/L
11.	Na ⁺	-	2.150	0.009	-	-	0.052	0.103	0.098	0.136	-	-	-	-	-	-	mg/L
12.	NH ₄ ⁺	1.261	25.091	0.538	0.257	0.087	2.000	1.535	0.273	0.283	0.098	0.722	0.513	0.152	0.233	0.109	mg/L
13.	K ⁺	14.459	1.086	1.093	0.753	2.486	18.303	16.639	1.125	0.702	2.154	12.230	18.369	0.443	5.957	1.170	mg/L
14.	Mg ²⁺	0.789	0.894	0.157	-	0.329	1.098	0.925	-	-	0.246	0.697	1.015	0.438	0.443	0.374	mg/L
15.	Ca ²⁺	1.328	-	-	-	0.343	1.819	0.693	-	0.199	0.235	0.754	0.743	0.739	0.432	0.680	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.4. Analysis of mineral components of *Paliurus spina Christi* extract

Depend on Table (4.67), and Figure (6.4), the method of conventional extraction was identified 11 ions (F^- , Cl^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}) in all different solvents. Further, the ion of K^+ : 8.355, 7.707, 6.720, 2.792, and 2.578 ppm, was highest values from the solvent of distilled water to hexane, and the Cl^- : 1.420, 1.394, 2.892, 0.826, and 0.789 ppm, in comparison with the lowest values of ions (F^- , NO_3^- , Mg^{2+} , and Ca^{2+}), respectively. Thus, the ions of SO_4^{2-} and PO_4^{3-} , had significant amounts in solvents (distilled water 2.153, 1.323 ppm), and (hexane: 4.048, 1.241 ppm) respectively. On the other hand, the solvent of hexane was detected all the mineral compounds in except the ion F^- , whereas the ion Na^+ was not detected in all solvents excluding hexane.

The second technique was microwave extraction which has been identified eleven mineral compounds in all various solvents. The solvent of Ethanol: Distilled water was given significant-high quantities in all ions K^+ (13.977ppm), SO_4^{2-} (2.906 ppm), Cl^- (2.359 ppm), PO_4^{3-} (1.463 ppm), Mg^{2+} (0.817 ppm), NO_3^- (0.731 ppm), F^- (0.669 ppm), Ca^{2+} (0.532 ppm), Li^+ (0.200 ppm), NH_4^+ (0.280 ppm), Na^+ (0.089 ppm), to compare with solvents of (Ethanol), and (Hexane: ethanol), respectively. On the other hand, the solvent of hexane had detected lower values of all ions. Furthermore, the ion of Na^+ detected limit values with all solvents except for distilled water.

Also, the orbital shaker technique was detected eight ions (F^- , Cl^- , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , Mg^{2+} , and Ca^{2+}), in all solvents. The ions of Ca^{2+} (10.993, 14.242, 2.388, 2.828 ,and 1.483), Na^+ (1.848, 2.590, 1.615, 1.984, 0.732), and Cl^- (1.945, 1.711, 1.924, 1.654, 0.605), were identified highest significant values in all solvents (W , W: E , E, H:E , and H), while, the ion of Ca^{2+} better than in comparison with ions of Na^+ , Cl^- , and other ions, respectively. In this technique, the ion of K^+ was not detected in all solvents. Otherwise, the solvent of hexane was showed the lowest value in all solvents, while comparing with other solvents.

In our study, that the techniques of (microwave and conventional) extraction were significant ways to recognize the mineral compounds of *Paliurus spina christi*, in solvent ethanol: distilled water, and with other various solvents.

Table 4.67. Analysis of mineral components of *Paliurus spina Christi* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.590	0.591	0.717	0.525	0.000	0.541	0.669	0.580	0.484	0.546	0.000	0.693	0.505	0.486	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	1.420	1.394	2.892	0.826	0.789	0.934	2.359	1.831	1.228	0.694	1.945	1.711	1.924	1.654	0.605	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	0.555	0.558	0.869	0.439	0.455	0.463	0.731	0.656	0.539	-	-	0.688	0.562	0.521	-	mg/L
8.	PO ₄ ³⁻	1.323	0.936	-	-	1.241	0.686	1.463	-	-	0.578	-	1.110	0.000	0.000	0.700	mg/L
9.	SO ₄ ²⁻	2.153	1.829	-	0.806	4.048	1.443	2.906	0.584	0.906	0.957	1.848	2.590	1.615	1.984	0.732	mg/L
10.	Li ⁺	-	-	-	-	0.437	0.216	0.200	0.202	0.212	0.238	0.198	0.205	0.198	0.204	0.198	mg/L
11.	Na ⁺	-	-	-	-	0.107	-	0.089	0.187	0.025	0.058	-	-	-	-	-	mg/L
12.	NH ₄ ⁺	0.214	0.178	0.310	0.065	0.066	0.604	0.280	0.112	0.084	0.046	-	2.040	0.231	0.152	0.088	mg/L
13.	K ⁺	8.355	7.707	6.720	2.792	2.578	4.318	13.977	4.342	3.242	0.371	10.993	14.242	2.388	2.828	1.483	mg/L
14.	Mg ²⁺	0.633	0.571	0.209	0.290	0.279	0.416	0.817	0.183	0.277	-	0.471	1.165	0.476	0.627	0.233	mg/L
15.	Ca ²⁺	0.719	0.493	0.197	0.282	0.369	0.528	0.532	0.165	0.296	0.144	0.452	0.895	0.736	0.852	0.180	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.5. Analysis of mineral components of *portulaca oleracea* extract

The technique of conventional extraction of *portulaca oleracea* herb, was detected eleven mineral compounds in various solvents, as shown in Table (4.68), and Figure (6.5). The ions of K^+ (32.305, 33.099, 16.999, 10.193, 2.632 ppm), Cl^- (3.205, 2.933, 8.128, 4.918, 0.772 ppm), NO_3^- (1.497, 1.380, 1.743, 0.802, 0.449 ppm), Mg^{2+} (1.228, 0.707, 0.319, 0.206, 0.308 ppm), have been identified the highest significantly values, to compare with other ions (SO_4^{2-} , Ca^+ , and NH_4^+) in all solvents (distilled water, ethanol: distilled water: ethanol, hexane: ethanol, and hexane), respectively. On the other hand, the ion of PO_4^{3-} was found to have significant values in solvents (distilled water: 3.342 ppm) and (Ethanol: distilled water: 3.029 ppm). Also, the ions of F^- and Li^+ , was obtained a lower value in all solvents, from the solvent of ethanol and hexane have not detected ion Na^+ .

the *portulaca oleracea* with microwave technique was observed 11 ions (F^- , Cl^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), during all solvents. The ion of K^+ in all solvents (DW 41.508 ppm, E: DW 30.542 ppm, E 13.988 ppm, H: E 7.693 ppm, and H 1.805 **ppm**) were detected largest and better than among all ions, as the ions of Cl^- (3.833 ppm), (2.591 ppm), (6.648 ppm), (3.821 ppm), and (0.602 ppm), NO_3^- (1.804 ppm), (1.314 ppm), (1.552 ppm), (0.726 ppm), and (0.420 ppm), Mg^{2+} (2.394 ppm), (0.704 ppm), (0.195 ppm), (0.184 ppm), and (0.180 ppm), SO_4^{2-} (1.093 ppm), (1.796 ppm), (0.577), (0.882 ppm), and (0.684 ppm), respectively, in comparison with other ions (F^- , Li^+ , Na^+ , and Ca^{2+}). Also, the ion of PO_4^{3-} in solvents ethanol: distilled water and **distilled water have been** found the highest values while comparing with other solvents. On the other hand, all the ions were showed a lower value with a solvent of hexane.

Additionally, the orbital shaker technique was showed ten ions of (Cl^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), throughout all solvents. All the solvents of (W, W: E, E, H: E, and H), gave significant quantities of K^+ ion (62.030, 50.123, 14.510, 13.757, and 8.428) ppm, respectively. Thus, the ions of Cl^- (5.610 ppm), (5.478 ppm), (5.791 ppm), (3.176 ppm), and (0.906 ppm), were observed high values in all solvents in comparison with other ions. Therefore, the solvents of distilled water, and **ethanol: distilled water**, were detected significant values of ions (PO_4^{3-} 1.604 ppm, 5.503 ppm), (SO_4^{2-} 2.355, 1.841 ppm), and (Mg^{2+} : 1.940, 0.729 ppm), in compare with ions of (NO_3^- , NH_4^+ , and Ca^{2+}), identified a lower value in all solvents, respectively. On the other hand, the ion of Na^+ was not detected in all solvents, with the exception of solvent (distilled water). In our study showed, that the *portulaca oleracea*

was obtain the good and highest values of mineral compounds with the all techniques, therefore, the ion of Potassium (K^+), was shows significant and beneficial value with all techniques and solvents. Because of its low carbohydrate content and high potassium content, it can be considered a healthy food for diabetic patients. It has also been linked to stronger blood pressure regulation (Anthonia Ezeabara 2014). Our study, approval with reports (Alam *et al.* 2014; Siriamornpun and Sutt 2010), had reported, that the common *purslane*, *Portulaca oleracea L*, contains more minerals content than ornamental purslanes.. Also, agreement with (Uddin *et al.* 2012), reported, the *Portulaca olerecea*, cultivars could be utilized as a source of minerals and antioxidants, particularly for nutraceuticals applications and useful foods.



Table 4.68. Analysis of mineral components of *portulaca oleracea* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.512	-	0.586	-	-	0.480	0.510	0.484	0.579	-	-	-	-	-	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	3.205	2.933	8.128	4.918	0.772	3.833	2.591	6.648	3.821	0.602	5.610	5.478	5.791	3.176	0.906	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	1.497	1.380	1.743	0.802	0.449	1.804	1.314	1.552	0.726	0.420	0.000	1.941	1.612	1.052	0.544	mg/L
8.	PO ₄ ³⁻	3.342	3.029	-	-	-	2.780	3.266	-	-	-	1.604	5.503	0.616	1.077	1.226	mg/L
9.	SO ₄ ²⁻	0.991	1.481	0.699	0.000	0.945	1.093	1.796	0.577	0.882	0.684	2.355	1.841	0.622	0.774	0.777	mg/L
10.	Li ⁺	0.199	0.215	0.207	0.197	-	0.199	0.200	0.199	0.219	0.199	0.197	0.205	0.197	0.215	0.200	mg/L
11.	Na ⁺	0.216	0.221	-	0.151	-	0.471	0.208	0.165	0.151	0.085	0.320	-	-	-	-	mg/L
12.	NH ₄ ⁺	0.091	0.103	0.118	0.099	0.058	0.114	0.073	0.045	0.082	-	0.045	0.050	-	0.063	0.126	mg/L
13.	K ⁺	32.305	33.099	16.99	10.19	2.632	41.508	30.542	13.98	7.693	1.805	62.030	50.123	14.510	13.757	8.428	mg/L
14.	Mg ²⁺	1.228	0.707	0.319	0.206	0.308	2.394	0.704	0.195	0.184	0.180	1.940	0.729	0.410	0.542	0.585	mg/L
15.	Ca ²⁺	0.295	0.247	0.237	0.223	0.356	0.263	0.156	0.228	0.219	0.180	0.240	0.241	0.209	0.255	0.314	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.6. Analysis of mineral components of *Artemisia haussknechtii* boiss extract

From Table (4.69), and Figure (6.6). The method of conventional extraction had detected eleven mineral compounds of ions (F^- , Cl^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), through all solvents. According to the analysis of our results, it was found that (K^+ E: DW 46.592, DW 12.667, E 5.069, H: E 3.580, H 0.844 ppm), the content was the biggest significant quantity among all ions and in all solvents. Therefore, the ions of SO_4^{2-} : (5.924, 2.640, 0.717, 0.606, 1.397 ppm), Ca^{2+} : (2.666, 0.301, 0.269, 0.319, 0.269 ppm), and Cl^- : (1.454, 3.980, 1.764, 1.510, 0.574 ppm), was given some significant quantities in all variety of solvents, in comparison with the lower values of ions (F^- , NO_3^- , Li^+ , and Mg^{2+}), respectively, on the other hand, the ion of PO_4^{3-} , gives a high value in solvents distilled water 2.755 ppm, and ethanol: distilled water 4.556 ppm. The Na^+ content was not detected with solvent (distilled water), while the detected in other solvents.

Depend on the technique of microwave extraction, was identified eleven mineral compounds in all different solvents. The Potassium ion was given a big significant amount in all solvents as (DW 11.770), (E: DW 22.240), (E 4.994), (H: E 1.158), and (H: 1.035 ppm) respectively. Thus, the ions of Ca^{2+} (2.142 ppm), (2.753 ppm), (0.301 pm), (0.332 ppm), and (0.396 ppm), Cl^- (1.385 ppm), (2.426 ppm), (1.945 ppm), (0.797 ppm), (1.385 ppm), SO_4^{2-} (1.873 ppm), (2.930 ppm), (0.630 ppm), (0.683 ppm), (0.786 ppm), were discovered a high significant values. In comparing with lower values of ions (NO_3^- , Li^+ , Na^+ , and NH_4^+), furthermore, the phosphate ion has detected a significant amount in solvent (distilled water) and (ethanol: distilled water), On the other side, the smallest values were obtained of all mineral compounds in hexane solvent.

The technique of the orbital shaker had detected eleven mineral components by ion Chromatography in all varying solvents. however, the ions of K^+ (13.915, 8.838, 4.664, 3.220, 2.188 ppm), SO_4^{2-} (2.242, 1.654, 0.637, 1.400, 1.009 ppm), and Cl^- (2.242, 1.654, 0.637, 1.400, 1.009 ppm), was obtained highest values compared with the lowers values as following ions (F^- , NO_3^- , Li^+ , NH_4^+ and Mg^{2+}), with all solvent. Therefore, the solvents of (distilled water 2.633 ppm), and (ethanol: distilled water 1.198 ppm) was showed significant values of ion potassium in all mineral components, thus the sodium was not detected in all solvents in except solvent of ethanol.

In our investigation, the solvents distilled water and ethanol: distilled water with different techniques, were observed a new study of mineral compounds by ion chromatography in *Artemisia haussknechtii* boiss.



Table 4.69. Analysis of mineral components of *Artemisia haussknechtii* boiss extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.784	-	0.667	0.563	-	0.747	1.038	0.639	0.505	0.496	0.612	0.825	0.604	0.548	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	1.454	3.980	1.764	1.510	0.574	1.385	2.426	1.945	0.797	0.605	1.478	1.284	1.988	0.978	0.678	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	0.669	1.853	0.794	0.769	-	0.687	1.049	0.846	0.451	0.425	-	0.683	0.895	0.530	0.427	mg/L
8.	PO ₄ ³⁻	2.755	4.556	-	-	-	1.280	1.200	-	-	0.578	2.633	1.198	0.000	0.746	-	mg/L
9.	SO ₄ ²⁻	5.924	2.640	0.717	0.606	1.397	1.873	2.930	0.630	0.683	0.786	2.242	1.654	0.637	1.400	1.009	mg/L
10.	Li ⁺	0.500	0.282	0.232	-	0.256	0.209	0.239	-	0.220	0.201	0.228	0.240	0.234	0.262	0.228	mg/L
11.	Na ⁺	-	0.344	0.132	0.116	0.050	0.279	0.239	0.164	0.069	0.047	-	-	0.109	-	-	mg/L
12.	NH ₄ ⁺	0.343	0.133	0.384	0.426	0.045	0.071	0.512	0.240	0.139	0.044	0.262	0.178	0.439	0.197	-	mg/L
13.	K ⁺	12.667	46.592	5.069	3.580	0.844	11.770	22.240	4.994	1.158	1.035	13.915	8.838	4.664	3.220	2.188	mg/L
14.	Mg ²⁺	0.885	0.953	0.208	0.250	0.197	0.700	1.325	0.193	0.180	0.233	0.840	0.687	0.226	0.283	0.217	mg/L
15.	Ca ²⁺	2.666	0.301	0.269	0.319	0.269	2.142	2.753	0.301	0.332	0.396	2.375	1.178	0.217	0.719	0.548	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.7. Analysis of mineral components of *Prunus microcarpa* extarct

In our study, the *Prunus microcarpa*, illustrate in Table (4.70), and Figure (6.7), the conventional extraction in all solvents was identified ten mineral compounds of ions (F^- , Cl^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{+2} , and Ca^{2+}). The solvents of distilled water, gave the highest ions of K^+ PO_4^{3-} , and NH_4^+ (3.656 ppm), (1.563 ppm), and (0.190 pmm), in comparison with the lower values (2.536 1.270 and 0.062 ppm) in solvent (ethanol: distilled water), respectively. Thus, the ions of (SO_4^{2-} , Cl^- , Ca^{2+} , F^- , Mg^{2+} , Na^+ , and Li^+), were detected significant quantities (3.951, 1.003, 0.549, 0.518, 0.503, 0.432, and 0.362 ppm) in solvent (ethanol: distilled water), with various solvents. Furthermore, the Na^+ content was not detected in solvents of (Distilled water, Hexane: ethanol, and Hexane), whereas identified in another solvent, on the other hand, the solvent of Hexane was given a lowest amount in all mineral compounds.

The methods of Microwave extraction in some solvents were detected ten mineral components of ions (F^- , Cl^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}). Furthermore, the ions of SO_4^{2-} , Li^+ , and Na^+ , were detected amounts in all solvents (0.918, 0.993, 0.677, 0.573, 0.722 ppm), (0.213, 0.228, 0.203, 0.202, 0.215 ppm), and (0.113, 0.279, 0.101, 0.002, 0.061) respectively. On the other hand, the ions of (K^+ and PO_4^{3-}), was detected highest significant values in solvents of (Distilled water 4.347, 2.246 ppm), (Ethanol: distilled water 2.928, 1.240 ppm), and (Ethanol 1.803, 1.012 ppm), While compared with the lowest values of ions (F^- , Cl^- , NH_4^+ , Mg^{+2} , and Ca^{2+}) with some solvents, respectively.

Furthermore, the orbital shaker extraction had detected eight ions (F^- , Cl^- , SO_4^{2-} , Li^+ , Na^+ , K^+ , Mg^{+2} , and Ca^{2+}), in different solvents. The solvents of (distilled water), and (ethanol: distilled water), were identified high significant values of ions SO_4^- (5.204, 2.783 ppm), and Mg^{2+} (2.681, 4.199 ppm), in comparison with the smallest quantities of ions Cl^- , Li^+ , and Ca^{2+} in all various solvents, respectively. On the other hand, the ions of K^+ and Na^+ were detected with a solvent of distilled water, while the ion sodium was not detected in all solvents.

In this study, there was obtained significant quantities of mineral compounds, by using conventional extraction in solvents of (distilled water), and (ethanol: distilled water), Hence, this study is the initial experience to discovered mineral compounds by processes of ion chromatography in *Prunus microcarpa*.

Table 4.70. Analysis of mineral components of *Prunus microcarpa* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.550	0.518	0.525	-	0.520	0.516	0.505	-	-	-	-	2.028	-	-	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	0.562	1.003	0.636	0.539	0.612	0.587	0.577	0.582	-	-	0.595	0.617	0.513	0.511	0.507	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
8.	PO ₄ ³⁻	1.563	1.270	-	-	-	2.246	1.240	1.012	-	-	5.204	2.783	-	-	0.476	mg/L
9.	SO ₄ ²⁻	0.812	3.951	0.870	0.589	1.279	0.918	0.993	0.677	0.573	0.722	0.924	0.715	0.512	0.670	0.513	mg/L
10.	Li ⁺	0.202	0.362	0.204	0.196	0.235	0.213	0.228	0.203	0.202	0.215	0.224	-	-	0.215	-	mg/L
11.	Na ⁺	-	0.432	0.182	-	-	0.113	0.279	0.101	0.002	0.061	-	-	-	-	-	mg/L
12.	NH ₄ ⁺	0.190	0.062	0.052	0.000	0.047	0.239	-	0.081	-	0.046	0.294	-	-	-	-	mg/L
13.	K ⁺	3.656	2.536	0.536	0.149	0.704	4.347	2.928	1.803	-	0.327	2.681	4.199	-	-	-	mg/L
14.	Mg ²⁺	0.643	0.503	0.237	0.163	0.270	0.867	0.258	0.204	-	0.156	0.760	0.553	-	-	-	mg/L
15.	Ca ²⁺	0.411	0.549	0.333	0.552	0.434	0.509	0.273	0.166	-	0.147	0.539	0.461	-	-	-	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.8. Analysis of mineral components of *Silybum Marianum* extract

Data from Table (4.71), and Figure (6.8), showed that the techniques of conventional extraction had identified ten ions in different solvents (F^- , Cl^- , SO_4^{2-} , Li^+ , Na^+ , K^+ , Mg^{2+} , and Ca^{2+}). The ions of (K^+), and (SO_4^{2-}), were identified high significant values (4.182, 2.190, 0.303, 0.256, 0.147ppm), and (1.659, 0.894, 0.894, 0.772, 0.614 ppm), compare with the decrease values of ions (F^- , Cl^- , Li^+ , NH_4^+ , Mg^{2+} , and Ca^{2+}) in all solvents, thus, the ion of Na^+ was detected in solvent distilled water whereas, not data recorded in solvents of (E: W, E, H: E, and H). However, the solvent of distilled water and ethanol: distilled water, were got significant values of ion PO_4 (1.444, 0.694 ppm) respectively, compared with the lower all ions with solvent of hexane.

According to the method of the microwave extraction had observed eight mineral components (Cl^- , SO_4^{3-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), by ion Chromatography in dissimilar solvents. Therefore, the ions of Potassium (K^+) recieved the highest quantity (2.632, 0.731, 1.147, 0.282, 0.649 ppm) with five solvents. Hence, the ions of (Cl^- , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , Mg^{2+} , and Ca^{2+}), obtained significant values, while in comparison with the ions of K^+ were the smallest values. On the other hand, the ion sodium was detected in all solvents except (ethanol: distilled water), whereas the solvent of hexane was giving the lowest values in all ions.

The orbital shaker extraction was detected seven mineral compounds (Cl^- , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{+2} , and Ca^{2+}), in five solvents. The solvents of (DW, E: DW, E, H: E), were identified as high significant values of ion K^+ (2.440, 1.514, 0.335, and, 0.146 ppm), while in compared with the lowest amounts of ions (Cl^- , SO_4^{2-} , Na^+ , NH_4^+ , and Mg^{2+}), respectively in solvents of (DW, E: DW, and E). Thus, the ion of sodium was the smallest amount in solvents of (DW 0.028, E: DW 0.127, and E: 0.029 ppm), as well as there was not detected in solvents of (H: E, and H), respectively. On the other hand, the solvent of hexane was showed the lowest values in some solvents.

Our study Correspond to (Ghafor *et al.* 2014), the report indicates, that the *Silybum marianum* stems have recorded the highest concentrations of Si 398.4 ppm, and Al 166.1 ppm, respectively, the lowest concentration of As (Ca. 0.028 ppm). Milk thistle seeds were well-known for providing a good source of minerals when consumed, the present study showed that the conventional extraction was the best and significant technique with solvent (distilled water), and (ethanol: distilled water), in comparison with the microwave and orbital shaker techniques of *Silybum Marianum*

Table 4.71. Analysis of mineral components of *Silybum Marianum* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.540	0.505	0.504	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	0.695	0.643	0.684	0.637	0.552	0.608	0.600	0.548	0.553	0.622	0.610	0.591	0.550	-	0.525	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
8.	PO ₄ ³⁻	1.444	0.694	-	-	-	-	-	-	-	-	0.682	-	-	0.000	0.517	mg/L
9.	SO ₄ ²⁻	1.659	0.894	0.894	0.772	0.614	0.638	0.607	0.559	0.855	0.636	0.577	0.540	-	0.505	0.547	mg/L
10.	Li ⁺	0.250	0.207	0.199	0.199	-	0.202	0.201	0.203	0.229	0.207	0.000	0.202	-	-	-	mg/L
11.	Na ⁺	0.066	-	-	-	-	0.248	-	0.050	0.130	0.079	0.028	0.127	0.029	-	-	mg/L
12.	NH ₄ ⁺	0.487	0.107	0.062	0.059	-	0.270	0.059	0.084	-	-	0.767	-	0.053	-	-	mg/L
13.	K ⁺	4.182	2.190	0.303	0.256	0.147	2.632	0.731	1.147	0.282	0.649	2.440	1.514	0.335	0.146	-	mg/L
14.	Mg ²⁺	0.688	0.410	0.229	0.219	0.169	0.503	0.248	0.386	0.153	0.321	0.616	0.477	0.194	-	0.216	mg/L
15.	Ca ²⁺	0.994	0.648	0.603	0.490	0.305	0.516	0.170	0.377	0.151	0.155	0.518	0.323	-	-	0.283	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

4.8.9. Analysis of mineral components of *Pistacia eurycarpa yale* extract

Interestingly of *Pistacia eurycarpa Yale*, from Table (4.72), and Figure (6.9), the ten of mineral compounds (F^- , Cl^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), were detected by using the conventional method with all solvents. The ion of K^+ in solvent (DW), and (E: DW) were observed largest amount (4.053, 4.270 ppm), in comparison with solvents of (E, H: E, and H). On the other hand, the ions of Cl^- , Mg^{2+} , Ca^{2+} , NH_4^+ , and SO_4^{2-} obtained significant values in all solvents. Therefore, the ions of (F^- , PO_4^{3-} , and Li^+) were detected in both solvents (distilled water), and (ethanol: distilled water). Additionally, the solvent of hexane was given the highest significant values in ion (SO_4^{2-} 4.279, and PO_4^{3-} 1.228 ppm) whereas, the sodium was detected with all solvent while in excepting not detected in solvent of (distilled water).

Furthermore, the seven mineral compounds (F^- , Cl^- , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}) was found in using microwave extraction in four solvents ((DW, E: DW, E, and H: E). the solvent of (ethanol: distilled) was gives a high significant quantities of ions (K^+ 5.698), (Cl^- 1.415 ppm), (SO_4^{2-} 1.218 ppm), (Ca^{2+} 0.933 ppm), (Na^+ 0.616 ppm), (Mg^{2+} 0.593 ppm), and F^- 0.518 ppm), in comparison with lowering values with solvent of (distilled water). Also the solvent of ethanol was obtained a significant values of ions (K^+ 1.085 ppm), (Li^+ 0.196 ppm), (Ca^{2+} 0.149 ppm), (Na^+ 0.106 ppm), and (NH_4^+ 0.047 ppm), respectively.

The techniques of orbital shaker extraction had discovered nine mineral compounds as followed (F^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}), through various solvents. The distilled water was detected highly values of ions (K^+ 4.656, PO_4^{3-} 1.028, F^- 0.607, SO_4^{2-} 0.563, Mg^{2+} 0.350, and, Ca^{2+} 0.249), while comparison with lower ions in the other solvents, respectively. Moreover, the solvent of (ethanol: distilled water) was given the significant of ions (K^+ 2.420 ppm, F^- 0.560 ppm, SO_4^{2-} 0.531 ppm, Mg^{2+} 0.301 ppm, and Ca^{2+} 0.267 ppm), respectively. On the other hand, the ion of sodium was not detected in all solvents, in exclusion of solvent ethanol.

In consequence of our study, the conventional extraction and solvent of (ethanol: distilled water) have been concluded a good and important technique to be obtained significant mineral compounds of *Pistacia eurycarpa Yale*, in comparison with the other methods and solvents.

Table 4.72. Analysis of mineral components of *Pistacia eurycarpa yale* extract

Detector	Name	C/E					MW/E					OS/E					Units
		W	W: E	E	H: E	H	W	W: E	E	H: E	H	W	W: E	E	H: E	H	
		Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	Conc. (ppm)	
1.	F ⁻	0.497	0.499	0.505	-	-	-	0.518	-	-	-	-	-	-	-	-	mg/L
2.	ClO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
3.	BrO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
4.	Cl ⁻	0.714	0.593	0.580	0.533	0.589	0.569	1.415	0.567	0.000	0.000	0.607	0.560	0.560	0.532	0.000	mg/L
5.	NO ₂ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
6.	Br ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
7.	NO ₃ ⁻	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	mg/L
8.	PO ₄ ⁻	0.699	0.661	-	-	1.228	-	-	-	-	-	1.028	-	-	-	-	mg/L
9.	SO ₄ ⁻	0.805	0.745	0.542	-	4.279	0.564	1.218	-	0.520	0.000	0.563	0.531	0.629	-	0.596	mg/L
10.	Li ⁺	0.205	0.198	-	-	0.494	-	-	0.196	-	-	-	-	0.206	-	0.206	mg/L
11.	Na ⁺	-	0.173	0.015	0.103	0.248	0.073	0.616	0.106	0.063	0.063	-	-	0.018	-	-	mg/L
12.	NH ₄ ⁺	0.150	0.144	0.058	0.047	-	-	-	0.047	-	-	-	-	0.093	0.043	-	mg/L
13.	K ⁺	4.053	4.270	0.617	0.401	0.720	3.065	5.698	1.085	-	-	4.656	2.420	0.756	0.381	-	mg/L
14.	Mg ⁺²	0.357	0.337	0.151	-	0.164	0.301	0.593	-	-	-	0.350	0.301	-	-	-	mg/L
15.	Ca ⁺²	0.442	0.291	-	-	0.357	0.143	0.933	0.149	-	-	0.249	0.267	-	-	-	mg/L

Method technique: Conventional Extraction (C/E), Microwave Extraction (MW/E), Orbital Shaker Extraction (OS/E); **Solvents:** DW: Distilled water, DW: E, Distilled Water-Ethanol, E: Ethanol, H-E: Hexane-Ethanol, and H: Hexane; **Mineral Ions:** 1. F⁻ (Fluoride), 2. ClO₂⁻ (Chlorite), 3. BrO₃⁻ (Bromate ion), 4. Cl⁻ (Chloride), 5. NO₂⁻ (nitrate), 6. Br⁻ (Bromide), 7. NO₃⁻ (Nitrate), 8. PO₄³⁻ (Phosphate), 9. SO₄²⁻ (Sulfate), 10. Li⁺ (Lithium), 11. Na⁺ (sodium), 12. NH₄⁺ (Ammonium), 13. K⁺ (potassium), 14. Mg²⁺ (Magnesium), and 15. Ca²⁺ (Calcium); **not detected:** N.D

This work that is summarized the ions of (F^- , Cl^- , NO_3^- , PO_4^- , SO_4^- , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{+2} , and Ca^{+2}), were detected in great significant quantities in our plants. While, the ions of (ClO_2^- , BrO_3^- , NO_2^- , and Br^-), were given zero ranges with all analysis methods in some plants, our finding agrees with the reported by researchers (Holem, Eyyüp, *et al.* 2020). Furthermore, sodium (K^+) contributes to the protection of tissue excitability and the ion regulation of the human body. It is an essential food and necessary roles in the synthesis of amino acids in proteins, as well as the ion K^+ is necessary for its diuretic nature (Chen *et al.* 2005).

Moreover, in our study, the ion of sodium (Na^+) content had detected in some solvents, whereas was not detected in some other solvents. Therefore, the ion Na^+ plays roles an important function in the transport of metabolites (Chen *et al.* 2005). Also, sodium (Na^+) is a basic components in fibrin construction which types fibrinogen and in consequence fibrin and collagen (Schalm *et al.* 1975). Fibrin is a dotting issue responsible for the balance. K^+ and Na^+ ions are recognized activators of energy potentials during the nerve membrane collectively with ion Ca^+ , ions may also serve as intestine peristalsis, replenishment in diarrheic conditions, and maintenance of ordinary apprehensive function.

Hence, The ratio K^+ and Na^+ in any food is an essential factor in the prevention of hypertension and arteriosclerosis with K depressed and no enhanced blood stress (Saupi *et al.* 2009; Chen *et al.* 2005), also the mutual effects of K^+ and Na^+ have shown that a low diet in Na^+ and a large diet in K^+ (low urinary Na^+ and K^+ ratio) favor lower blood pressure. and would reduce the development of cardiovascular disease (Luft 1990). On the other hand, an increase in dietary potassium (K^+) as the Cl^- (salt) has shown to decrease blood pressure in some hypertensive people (Bangash *et al.* 2011).

Despite, both the ions Magnesium (Mg^{+2}), and calcium (Ca^{+2}), were indicated the high limited values in our plant .that is a play a large function in biomolecules metabolism, photosynthesis, and binding agents of cell walls (El Khatib *et al.* 2003) Therefore, the ion of Mg^{+2} plays an important role in the structure and the function of the human body (Cerklewski 1997). Further, the Mg^{+2} ions are recognized hormone activators in kind of second diabetes (Schalm *et al.* 1975). Also, calcium (Ca^{+2}) was obtained a significant value in our plant, It is very greatly required for the regular functioning of the blood coagulation of the heart muscle, milk clothing, and the permeability mechanism. In addition, the Ca^{+2} is a big ingredient of bone and assists in tooth improvement, human blood and extracellular fluid (Indrayan *et al.* 2005; El Khatib *et al.* 2003), due to the Lack of Ca^{+2} , such as vitamin D, leads to the classic bone

symptoms associated with rickets, such as bowlegs, knock knees, the curvature of the spine, pelvic and thoracic deformities (NORMAN 2001). In the present work, a highly sensitive, effective, precision, and fast ion chromatography technique way was established and proven as suitable for separating, quantifying, and identifying mineral compounds. the mineral compounds has been displayed of ions (F^- , ClO_2^- , BrO_3^- , Cl^- , NO_2^- , Br^- , NO_3^- , PO_4^- , SO_4^- , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{+2} , and Ca^{2+}), that was providing a very helpful method for chemical analysis and biological study purposes.,(Masson *et al.* 1996).

Generally, this current report was the first experimental to find mineral compounds by techniques of ion chromatography in our plants.



4.9. Antibacterial activities of plant extract

4.9.1. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Eremurus Spectabilis Bieb* extracts against the bacteria with antibiotic (Antimicrobial Agent)

The several bacteria activity were inhibited by three different methods and with solvents, the significant inhibition zones were seen of *Eremurus Spectabilis Bieb*, extracts with the concentrations of 50, 70, and 100µL/disk, shown in Tables (4.73).

In our study, the mean of inhibition zones against effects bacteria in three concentrations by the conventional technique with distilled water solvent was ranged a significant highest inhibitory zone in the concentrations of 50, 70, and 100µL/disk inhibited growths of *Staphylococcus* (9.36, 13.18 and 17.33 mm) in diameter of inhibition zone respectively. Furthermore, the concentration of 100µL/disk of the extract was given zones *Staphylococcus aureus* 17.33 mm, *Pseudomonas aeruginosa* 14.62 mm, *Bacillus subtilis* 15.8 mm, *Enterococcus faecalis* 15.83 mm, *Enterobacter aerogenes* 12.85 mm, and *Klebsiella pneumonia* 15.16 mm, in diameter of inhibition zone against all bacteria respectively, which is effectively more than the tested standard antibiotics as P 10, AMP10, Eyr 10, and CN 10. Whereas, with the positively significant inhibitory zone in concentration 70µL/disk extract. In comparison with the concentration of 50µL/disk extract was the lowest and doesn't give the inhibition zones in some bacteria as follows *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *Klebsiella*, respectively.

On the other hand, the solvent of ethanol: distilled water extract of *Eremurus Spectabilis Bieb* showed the maximum inhibition zone in the concentration of 100µL/disk, with diameter 14.03 mm, 16.15 mm, 16.04 mm, 11.98 mm, 15.27 mm, and 16.16 mm), against six bacteria respectively. Whereas, with the concentration of 50, and 70 µL/disk were showed the comparatively limit and lowest inhibition zones of resistance with bacteria. Otherwise, the MIC values regarding the antibacterial activity of crude extraction were against bacteria species. In addition, the water extract was showed the least MIC values 3.90 µg/ml, 500 µg/ml, and 250 µg/ml against *Staphylococcus aureus*, *Enterococcus faecalis*, and *Enterobacter aerogenes*, while the solvent (ethanol: distilled water) extract was showed the great efficiency MIC values 31.25µg/ml, 15.62 µg/ml, 3.90µg/ml, 7.81µg/ml, 31.25µg/ml, 1.95µg/ml, against all bacteria respectively. Secondly, the microwave technique with a solvent of (distilled water) extracts in concentrations 50, 70, and 100µL/disk showed maximum significant inhibitory zones in

diameter 18.97mm, 18.53 mm, 17.44 mm, 16.47 mm, 16.04 mm, and 16.60 mm by the concentration of 100µL/disk. Whereas, the similarity and significant inhibitory zone with a diameter of 15.3, 15.89, 14.9, 11.77, 15.77, and 16.53 mm of inhibition zones by concentration 70µL/disk were against the varieties of bacteria.

However, the inhibitory zones showed low activity in the concentration of 50µL/disk was able to kill bacteria better than antibiotics. In comparison with (ethanol: distilled water v/v) extract was observed lowest inhibition zone with all diameters in concentrations of 70 and 100µL/disk. In except concentration 50µL/disk not detected inhibition zone to resist all bacteria. Furthermore, the concentration of 100µL/disk extract was observed as a suitable inhibition zones in diameter 14.03mm, 16.15mm, 16.04mm, 11.98mm, 15.27mm, 16.16mm against the organisms tested, respectively. Whereas, in the concentration of 70µL/disk of extracts, was detected a significant zone inhibition against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *Enterobacter aerogenes* respectively. Therefore, our extract of *Eremurus Spectabilis Bieb* was effectively more antibacterial activity than the tested standard antibiotics as P 10, AMP10, Eyr10, and CN 10. The data of MIC showed a proficient value against all bacteria tested were between 31.25-500µg/ml, in solvent (distilled water) extract, while the solvent (ethanol: distilled water) was ranged between 62-500µg/ml, to resistance all bacteria in except B.4, and B.6 respectively.

In the other methods of orbital shaker in distilled water was showed not detected inhibitory zones from all concentrations of 50, 70, and 100µL/disk extract, whereas the ethanol: distilled water showed maximum antibacterial activity of inhibition zones in the concentration 100µL/disk extract with a diameter (15.97mm, 18.29mm, 16.71mm, 12.74mm 15.43mm, and 13.61mm against all pathogenic bacteria strains, respectively. Furthermore, the MIC values were provided to have the highest activity at ranged 250µg/ml, 31.25µg/ml, 125µg/ml, 500µg/ml, and 250µg/ml. which sometimes resist antibiotics to prevent the growth of the organism's activity, this finding is supported and in agreement with (Murathan *et al.* 2018) that study revealed the extracts of *Eremurus Spectabilis Bieb*, possess antimicrobial and antimutagenic properties. Therefore, the consumption of *Eremurus Spectabilis Bieb* may prove to be beneficial for human health. Also dissimilar with (Bircan and Kırbağ 2015) Reported, that the *S. aureus*, *E. coli*, *Epidermophyton spp.*, and *Candida albicans* all have been different proportional antimicrobial activities of *Eremurus Spectabilis Bieb*.

Generally, that the data of inhibition zones and the MIC values showed that (MWE) and (CE) with (distilled water) have been higher antimicrobial activity compared to (OSE) in

(ethanol: distilled water). Therefore, showed better antibacterial activity extracts than antibiotic (Antimicrobial Agent) to resist the growth of bacteria activity.



Table 4.73. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Eremurus Spectabilis* Bieb solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis /Extract Solvents of Plant with bacteria dilution assay %	C/E I.Z. (mm)	W	50%	9.36	-	-	12.84	10.2	-
			70%	13.18	12.58	14.64	13.69	11.5	-
			100%	17.33	14.62	15.85	15.83	12.85	15.16
		W:E	50%	-	10.47	11.51	12.12	11.67	-
			70%	11.12	11.7	12.56	14.13	13.98	12.62
			100%	11.55	13.95	14.21	14.88	15.09	14.52
	C/E MIC (µg/ml)	W	≥500	3.90	-	-	500	250	-
		W:E	≥500	31.25	15.62	3.90	7.81	31.25	1.95
	MW/E I.Z. (mm)	W	50%	12.4	12.17	12.34	10.72	13.95	15.01
			70%	15.3	15.89	14.9	11.77	15.77	16.53
			100%	18.97	18.53	17.44	16.47	16.04	16.6
		W:E	50%	-	-	-	-	-	-
			70%	11.6	13.99	9.42	-	12.03	-
			100%	14.03	16.15	16.04	11.98	15.27	16.16
	MW/E MIC (µg/ml)	W	≥500	31.25	62.5	500	125	250	125
		W:E	≥500	62.5	500	500	-	250	-
	OS/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	8.51	14.94	-
			70%	10.20	-	-	-	-	-
			100%	15.97	18.29	16.71	12.74	15.43	13.61
	OS/E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	250	31.25	125	500	250	-
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	-	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=*Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa* (ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4=*Enterococcus faecalis* (ATCC 29212),B.5=*Enterobacter aerogenes*, B.6=*Klebsiella pneumonia*; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : Not Detected.

4.9.2. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Prosopis farcta* extracts against the bacteria with antibiotic (Antimicrobial Agent)

According to the Table (4.74). The conventional technique with solvents in concentrations of 50, 70, and 100µL/disk extract were not recorded any data growth of inhibition zone of *Prosopis farcta*, against five bacteria activity, except the water extract in the concentration of 100µL/disk, which showed the moderate antibacterial activity of inhibition zone (relative to the used standard antibiotic) against *Pseudomonas aeruginosa*, with inhibition zones of 10.60 mm, and with the comparatively efficient MIC value of 500µg/ml, compared to antibiotic as Ampicillin, and Gentamycin, with inhibition zones of (10.59mm, and 10.19) mm respectively.

On the other side, the method of microwave extraction with the solvent water was not detected any inhibitory zone to resist all bacteria. Whereas, the (ethanol: distilled water) extract in a concentration of 100µL/disk extract, showed the greatest inhibition zone in diameter 11.34mm, 17.27mm, 10.50mm, and 11.40mm, with the strong effect of MIC values 62µg/ml, 31µg/ml, 62µg/ml, and 500µg/ml, against four bacteria namely; *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *Enterobacter aerogenes*, in comparison with inhibition zones of Penicillin, Ampicillin, Erythromycin, and Gentamycin, respectively.

Another technique of orbital shaker with water extraction appeared a maximum effect inhibition zones in concentrations of 50, 70, and 100µL/disk extract. Therefore, the concentration of 100µL/disk showed the highest significant inhibitory zones with a diameter 13.12mm, 18.81mm, 17.22mm, 16.84mm, 18.13mm, and 21.15 mm, with the great of efficiency of MIC values of 125µg/ml, 31.25µg/ml, 62.5µg/ml, 125µg/ml, 31.25µg/ml, 15.62µg/ml, to resist all the bacteria activity, in compared with the artificial antibiotics as P 10, AMP10, Eyr 10, and CN 10 respectively.

Our study dissimilarity with (Poudineh *et al.* 2015), this studies showed that there was no antibacterial activity in the seed of *Prosopis farcta*, extracts while certain concentrations 10µL, 20µL, 40µL, and 80µL. Thus, approval with Sharifi-rad *et al.* (2014), reported the *Prosopis farcta*, extracts possess potent and high antibacterial activity against Methicillin-resistant *Staphylococcus aureus*, and may be used as an antibacterial to treat diseases. Also, that is disagree with these reports (Amal *et al.* 2017; Layla *et al.* 2018). Additionally, our study has indicated that the extract of *Prosopis farcta*, by the orbital shaker technique in distilled water,

showed a significant way to give the highest inhibitory zone against all the bacteria, in comparison with other techniques

Table 4.74. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Prosopis farcta* solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis /Extract Solvents of Plant with bacteria dilution assay %	C/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	10.60	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
	C/E MIC (µg/ml)	W	≥500	-	500	-	-	-	-
		W:E	≥500	-	-	-	-	-	-
	MW/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	15.49	-	-	-	-
			70%	-	16.82	-	-	-	-
			100%	11.34	17.27	10.50	-	11.40	-
	MW/E MIC (µg/ml)	W	≥500	-	-	-	-	-	-
		W:E	≥500	62.5	31.25	62.5	-	500	-
	OS/E I.Z. (mm)	W	50%	-	12.96	12.86	11.74	14.50	15.58
			70%	-	14.26	12.95	13.89	16.01	16.59
			100%	13.12	18.81	17.22	16.84	18.13	21.15
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	16.34	-	19.21
	OS/E MIC (µg/m)	W	≥500	250	250	62.5	125	31.25	15.62
		W:E	≥500	-	-	-	62.5	-	15.62
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	-	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=*Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa* (ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4=*Enterococcus faecalis* (ATCC 29212),B.5=*Enterobacter aerogenes*, B.6=*Klebsiella pneumonia*;Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : Not Detected.

4.9.3. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Capparis Spinosa* fruit extracts against the bacteria with antibiotic (Antimicrobial Agent)

The antibacterial activities from *Capparis Spinosa*, after determination was, illustrated in Table (4.75). In this study, the distilled water extract of the *Capparis Spinosa*, by the technique of (CE), in the concentration of 50, 70, and 100 μ L/disk extract, observed the greatest and better antibacterial was obtained in the inhibition zone to the resistance of *Staphylococcus aureus*, in the diameter 13.06mm, 14.96mm, and 18.74mm, that more effective to compare with an antibiotic (P10, AMP10, Eyr10, and CN10) in diameter 12.94 μ g/ml, 12.83 μ g/ml, 11.6 μ g/ml, and 16.9 μ g/ml respectively. The MIC was the greatest value 3.90 μ g/ml, to able kill microorganisms. Whereas, the solvent of (ethanol: distilled water) was given significant highly inhibitory zones in the concentration of 100 μ L/disk extract with diameter 12.72mm, 13.01mm, and 13.80, that effective on microorganisms namely; *Enterobacter aerogenes*, *Bacillus subtilis*, and *Klebsiella pneumonia*, compared with artificial antibiotics (AMP10, Eyr10, and CN10) respectively. However, The MIC has high values (15.62 μ g/ml, 31.25 μ g/ml, 31.25 μ g/ml inhibitors), which prevents the growth of bacteria activity.

Also, the microwave technique with distilled water extract showed a moderate inhibitory zone in concentration 100 μ L/disk with a diameter of 13.90 mm, against the *Enterococcus faecalis*, more effective on the bacteria than the antibiotics (P10, AMP10, C30, Eyr10, and CN10), in diameter 8.27 μ g/ml, 7.8 μ g/ml, 13.7 μ g/ml, 13.14 μ g/ml, and 10.71 μ g/ml, respectively. Instead, they observed an antibacterial activity by (ethanol: distilled water) highest inhibition zone against *Bacillus subtilis*, in concentration of 100 μ L/disk with diameter 15.62mm, *Enterococcus faecalis*, in concentration 70 μ L/disk, and 100 μ L/disk with diameter 13.03mm, and 14.69mm, and *Klebsiella*, in concentrations of 50,70,100 μ L/disk in diameter; 13.70mm, 14.14mm, and 17.29mm, respectively.

Furthermore, the (ethanol: distilled water) was the more effective extract compared with antibiotics like Penicillin, Ampicillin, Chloramphenicol, Erythromycin, and Gentamycin, respectively. The MIC values of water (62.5 μ g/ml), and ethanol: water (500 μ g/ml, 15.62 μ g/ml, and 7.81 μ g/ml), were strong extracts against *Enterococcus faecalis*, *Bacillus subtilis*, *Klebsiella*, respectively. On the other hand, no data detected growth of inhibitory zone by the orbital shaker technique with solvents to extract *Capparis Spinosa*, against all the bacteria activity. Our study agrees with (Oudah *et al.* 2019) had reported, that the highest activity of isolated polyphenolic for *Capparis spinosa* was against pathogens *E.coli* and *Pseudomonas*

(12mm), with the lowest activity in *Staphylococcus* (10mm) and an inhibition region against *Bacillus* (11mm). Also, our study intimated that the good finding of *Capparis Spinosa* extract, by microwave technique with solvent (E: DW), may give the strongest zone of inhibition as an antibacterial against bacteria activity.



Table 4.75. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Capparis Spinosa fruit* solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis /Extract Solvents of Plant with bacteria dilution assay %	C/E I.Z. (mm)	W	50%	13.06	-	-	-	-	-
			70%	14.96	-	-	-	-	-
			100%	18.74	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	8.70	-	12.21	-
			100%	-	-	12.72	-	13.01	13.80
	C/E MIC (µg/ml)	W	≥500	3.90	-	-	-	-	-
		W:E	≥500	-	-	15.62	-	31.25	31.25
	MW/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	13.90	-	-
		W:E	50%	-	-	-	-	-	13.70
			70%	-	-	-	13.03	-	14.14
			100%	-	-	15.33	14.69	-	17.29
	MW/E MIC (µg/ml)	W	≥500	-	-	-	62.5	-	-
		W:E	≥500	-	-	500	15.62	-	7.81
	OS/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
	OS/E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	-	-	-	-	-	-
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	-	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml); ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1= *Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa* (ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4= *Enterococcus faecalis* (ATCC 29212), B.5= *Enterobacter aerogenes*, B.6= *Klebsiella pneumoniae*; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : Not Detected.

4.9.4. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Paliurus spinachristi* fruit extracts against the bacteria with antibiotic (Antimicrobial Agent)

The six bacteria species were inhibited by the *Paliurus spinachristi* fruit extracts during three methods and different solvents of distilled water and, ethanol: distilled water. Furthermore, the zones of inhibition significantly were observed by the concentrations 50, 70, 100 μ L/disk of extracts, presented in Table (4.76).

In this study, the antibacterial activity of the distilled water extracted by conventional technique showed maximum inhibitory zones in concentrations 70, and 100 μ L/disk against *Staphylococcus aureus*, with a diameter (9.63mm, 11.81mm), *Pseudomonas aeruginosa*, with a diameter (11.78mm, 12.26mm), and *Bacillus subtilis* with a diameter (10.78mm, 13.96mm), in except *Enterobacter aerogenes* in diameter (10.03mm) with concentration 100 μ L/disk, and *Klebsiella pneumonia*, with a diameter (16.48mm, 17.80mm, and 19.76mm) in concentrations of 50, 70, and 100 μ L/disk, respectively. In comparison with the ethanol: distilled water was observed highest significantly inhibition zone in concentrations of 50, 70, and 100 μ L/disk to resistance bacteria from *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Enterococcus faecalis*, and *Enterobacter aerogenes*, with highest the diameter 14.33mm, 18.06mm, 16.78mm, 15.50mm, and 14.47mm respectively.

Also, the results of MIC distilled water as (7.81 μ g/ml, 15.62 μ g/ml, 125 μ g/ml, 62.5 μ g/ml, and 7.81 μ g/ml), and (ethanol: distilled water) as (500 μ g/ml, 31.25 μ g/ml, 7.81 μ g/ml, 125 μ g/ml, and 125 μ g/ml), were showed the strength and proficient values into preventing the bacteria activity, However, the detected activities of (DW, and E: DW), were more effective antibacterial activity compared to the antibiotic (Eyr10, P10, AMP10, CN10, and C30) which, showed obvious inhibitory impacts against all the tested bacteria.

Next, the microwave technique in solvent of (distilled water) was not recorded any inhibition zone. Whereas, the (ethanol: distilled water), showed a maximum inhibitory zone in concentrations 50, 70, and 100 μ L/disk of extracts. The highest activity (inhibition zones between 11.75 and 18.1mm) was found in *Paliurus spinachristi* extracts. Antibacterial activity of the extracts against all bacteria was measured at arranging between *Staphylococcus aureus* (12.75 and 16.97mm) in diameter, *Pseudomonas aeruginosa* (13.72 and 17.37mm) in diameter, *Bacillus subtilis* (11.75 and 13.09) in diameter, *Enterococcus faecalis* (13.50 and 16.81) in diameter, *Enterobacter aerogenes* (14.46 and 18.01) in diameter, and *Klebsiella pneumonia*

(12.79 and 14.75) in diameter, the antibacterial activities of (ethanol: distilled water) more efficient to compare with antibiotics (Eyr10, P10, AMP10, CN10, and C30) against the efficacy of bacteria. However, the MIC was great and more effective values (15.62µg/ml, 62.5µg/ml, 62.5µg/ml, 125µg/ml, 31.25µg/ml, 250µg/ml), to kill all the bacteria.

On the other hand, the orbital shaker method with (ethanol: distilled water) had a higher inhibitory effect on (*Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Enterococcus faecalis*, *Enterobacter aerogenes*, and *Klebsiella*, in concentration of 100µL/disk of extracts with diameters (12.50mm, 11.10mm, 10.72mm, 14.73mm, 14.45mm, 10.32mm) respectively, which have been more effective to compared with antibiotics to prohibit the growth of bacteria. Also, the MIC values have significant concentration 500µg/ml, 500µg/ml, 62.5µg/ml, 250µg/ml, 2505µg/ml, 62.55µg/ml) to kill microorganisms.

Our study of the current research is in agreement-in partial-with the results of (Brantner *et al.* 1996) studies that the flavonoids have been some influence on the antibacterial activity *Paliurus spina-christi*. Also, polyphenols can hence be considered as antibacterial agents, and there was an inhibitory effect resulting mainly on Gram-positive bacteria. Also, These findings are in line with (Motamedi *et al.* 2014) reported that results can be concluded that hydroalcoholic extracts yielded from *Ziziphus spina-christi* are suitable for fighting bacterial pathogens especially *Staphylococcus aureus* , which is against proffer antibiotics. Thus, our study accepted (Adzu and Haruna 2007) has explained that leaf extracts of this *Paliurus spina-christi* have antimicrobial, antidiabetic, antinociceptive, and antihyperglycaemic effects.

Generally, the (CE) and (ME) with (ethanol: distilled water), were obtained a strong zone of inhibition as antibacterial activity and showed better than in compared some antibiotic to destroy all growth of bacteria activity.

Table 4.76. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Paliurus spinachristi* fruit solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis / Extract Solvents of Plant with bacteria dilution assay %	C E I.Z. (mm)	W	50%	-	-	-	-	-	16.48
			70%	9.63	11.78	10.78	-	-	17.80
			100%	11.81	12.26	13.96	-	10.03	19.76
		W:E	50%	11.90	10.97	12.01	9.15	11.62	-
			70%	12.77	15.34	15.25	14.60	13.61	-
			100%	14.33	18.06	16.78	15.50	14.47	-
	C E MIC (µg/ml)	W	≥500	7.81	15.62	125	-	62.5	7.81
		W:E	≥500	500	31.25	7.81	125	125	-
	M W E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	12.75	13.72	11.75	13.50	14.46	12.79
			70%	14.29	14.91	12.34	16.04	14.95	13.71
			100%	16.97	17.37	13.09	16.81	18.01	14.75
	M W E MIC (µg/ml)	W	≥500	-	-	-	-	-	-
		W:E	≥500	15.62	62.5	62.5	125	31.25	250
	O S E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	12.50	11.10	10.72	14.73	14.45	10.32
	O S E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	500	500	62.5	250	250	62.5
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	N.E	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1= *Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa* (ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4= *Enterococcus faecalis* (ATCC 29212), B.5= *Enterobacter aerogenes*, B.6= *Klebsiella pneumonia*; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : No zone of inhibition.

4.9.5. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the Purslane *Portulaca oleracea* herb extracts against the bacteria with antibiotic (Antimicrobial Agent)

Our results of *Purslane Portulaca oleracea* herb, during three methods of (C/E, MW/E, OS/E), with five solvents in the concentration of 50, 70, and 100µL/disk, were given in Table (4.77), the distilled water extract in concentrations 70µL/disk and 100µL/disk, showed highly significant inhibitory zone activities in diameter (13.23mm, and 17.03mm), comparison with antibiotic of Penicillin, Ampicillin, Erythromycin, and Gentamycin, in diameter (10.02 µg/ml, 11.42µg/ml, 8.8µg/ml, and 17.66µg/ml) respectively, against of Gram-negative bacteria *Enterobacter aerogenes*, with obtain a greatest efficacy of MIC values 62.5 µg/ml. Whereas, the (ethanol: distilled water) extracts had maximum zone activity in concentration 50, 70, and 100µL/disk against six tested of bacteria, The highest inhibition zone concentration 100µL/disk in diameters (18.99mm, 19.57mm, 17.13mm, 16.14mm, 15.09mm, and 18.96mm) were more effective compared with concentration 50, 70µL/disk, and the antibiotic (Eyr10, P10, AMP10, CN10, and C30), to fight all bacteria respectively, which the effective MIC values (125µg/ml, 15µg/ml, 250µg/ml, 62.5µg/ml, 500 µg/ml, and 31.25) respectively.

Thus, the results of antibacterial activities were measured as a zone of inhibition in mm by microwave technique in solvents (distilled water) and (ethanol: distilled water) with concentrations 70µL/disk and 100µL/disk of extracts. The antibacterial activities of distilled water extract were estimated in vitro against *Enterococcus faecalis*, which highly inhibition zone in the concentrations of 70, and 100µL/disk with a diameter (10.07mm, 12.81mm), compared with antibiotics of Penicillin Ampicillin Gentamycin in diameter zone (8.27µg/ml, 7.8µg/ml, and 10.71µg/ml) respectively, Despite that, the MIC was effective value as 500µg/ml to kill the bacteria. Also, the (ethanol: distilled water) extracts, showed maximum inhibition zones in concentrations 50, 70, and 100µL/disk, to inhibit the growth of bacteria and the inhibition zones ranged between 9.03-17.46 mm. In addition, the extracts of concentration 100µL/disk, determine any antibacterial effects against used 6 bacteria in diameter (17.45mm, 12.14mm, 17.46mm, 13.23mm, 12.81mm, and 14.25) respectively in compared with lowest antibiotics (P 10, AMP10, C30, Eyr10, and CN 10) respectively, with the great sensitive of MIC values as (125µg/ml, 500µg/ml, 250µg/ml, 125µg/ml, 500µg/ml, and 125µg/ml) in (ethanol: distilled water) extracts respectively.

On the other side, the orbital shaker extraction in solvent of distilled water never any inhibition zone, Whereas, the (E: DW), showed an effective highly inhibitor activity in the

concentration of 100µL/disk of extract in diameter (18.42mm, 9.44mm, 7.44mm, 14.26mm, and 9.93mm), compared with lowest antibiotics (P10, AMP10, C30, Eyr10, and CN 10) respectively, to prevent the growth of activity five bacteria as namely; (*Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Enterococcus faecalis*, *Enterobacter aerogenes*, and *Klebsiella*), with the got greatest of MIC values in diameter (31.25µg/ml, 125µg/ml, 500µg/ml, 250µg/ml, and 62.5µg/ml) respectively.

Our finding in line with another lecturer (Nayaka *et al.* 2014) indicated, that the (MIC) of the flavonoid and apigenin isolated from *Portulaca oleracea* L. Therefore, can be used to produce antibacterial drugs. Also, our study approval with (Agyare *et al.* 2015b), the MIC values of 12.5, 12.5, 50.0, 50.0, and 50.0 mg/mL, the methanol extract showed antimicrobial activity against *S. aureus*, *S. pyogenes*, *E. coli*, *P. aeruginosa*, and *C. albicans*, respectively. Given the numerous pathogens this plant has been shown to inhibit, the anti-microbial potential of *Purtolaca oleracea* 70 percent ethanolic extract is also noteworthy. *Bacillus subtilis*, *Neisseria gonorrhoeae*, *Staphylococcus aureus*, and *Escherichia coli* (Elkhayat *et al.* 2008), this research approval with our study. On the other hand, the *Portulaca oleracea* has been used in traditional medicine to treat ailments like abscesses, bilious disorders, and coughs, and it also indicates the possibility of isolating antibacterial agents (Islam *et al.* 2018).

Additionally, the conventional and microwave techniques with solvent (ethanol: distilled water) were more efficient antibacterial activity than compared orbital shaker extract and antibiotics against the growth of all bacteria.

Table 4.77. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Purslane Portulaca oleracea L* extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis /Extract Solvents of Plant with bacteria dilution assay %	C/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	13.23	-
			100%	-	-	-	-	17.03	-
		W:E	50%	14.48	12.08	10.01	12.81	12.79	10.50
			70%	15.17	14.81	11.97	14.26	13.42	16.76
			100%	18.99	19.57	17.13	16.14	15.09	18.96
	C/E MIC (µg/ml)	W	≥500	-	-	-	-	62.5	-
		W:E	≥500	125	15	250	62.5	500	31.25
	MW/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	10.07	-	-
			100%	-	-	-	12.81	-	-
		W:E	50%	13.61	9.03	14.86	9.24	10.32	10.96
			70%	15.08	10.75	16.77	11.51	10.92	12.44
			100%	17.45	12.14	17.46	13.23	12.81	14.25
	MW/E MIC (µg/ml)	W	≥500	-	500	-	500	-	-
		W:E	≥500	125	500	250	125	500	125
	OS/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	18.42	9.44	-	7.44	14.26	9.93
	OS/E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	31.25	125	-	500	250	62.5
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	N.E	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=*Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa* (ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4=*Enterococcus faecalis* (ATCC 29212),B.5=*Enterobacter aerogenes*, B.6=*Klebsiella pneumonia*;Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : Not Detected.

4.9.6. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Artemisia hasskchitii* boiss extracts against the bacteria with antibiotic (Antimicrobial Agent)

The results of the antibacterial activity in concentrations 50, 70, and 100µL/disk with standard antibiotics are presented in Table (4.78). Interestingly, the results showed that the conventional technique in distilled water in a concentration of 100µL/disk of the extract was a maximum antibacterial activity to resistant to six of the tested bacteria. Furthermore, the highest concentration of 100µL/disk of distilled water extract, showed a great and high inhibition zone with the diameter (18.69mm, 17.70mm, 20.52mm, 20.05mm, 21.08mm, 18.82), compared with the lowest seven antibiotics (Penicillin, Ampicillin, Tetracycline, Chloramphenicol, Erythromycin, Ciprofloxacin, and Gentamycin) respectively, against all growth of isolate bacteria. Whereas, that the ethanol: distilled water gave a high inhibition zone in concentration 50, 70, and 100µL/disk of extract measured at (11.82mm to 21.04mm) in diameter, therefore, the concentration 100µL/disk was observed maximum zones of inhibition with the diameter (18.34mm, 21.04mm, 16.75mm, 17.59mm, 18.48mm, 19.05mm) showed more antibacterial activity compared to the minimum of antibiotics (P10, AMP10, TET10, C30, Eyr10, CF5, and CN10) respectively, to the killing of all bacteria, with great of significant effect values of MIC from (distilled water) in diameter (3.90µg/ml, 31.25µg/ml, 15.62µg/ml, 31.25µg/ml, 62.5µg/ml, and 3.90µg/ml), and (ethanol: distilled water) in diameter (7.81µg/ml, 3.90µg/ml, 125µg/ml, 500µg/ml, 125µg/ml, and 7.81µg/ml) respectively, to prevent of growth six microorganisms.

Besides, the microwave technique with a solvent of (distilled water) was a high significance of inhibition zone in the concentration of 100µL/disk of extracts with diameter 14.50mm, 16.28mm, 15.43mm, 18.41mm, 15.41mm, in comparison with the concentration of 50,70µL/disk and with lowest antibiotic against five bacteria as follows *Staphylococcus aureus* (ATCC 29213), *Pseudomonas aeruginosa* (ATCC 27853), *Enterococcus faecalis* (ATCC 29212), *Enterobacter aerogenes*, and *Klebsiella pneumonia*. As well, the (ethanol: distilled water) extracts given a great and efficacious inhibition zone in concentrations 100µL/disk in diameter (18.86mm, 19.76mm, 21.54mm, 20.34mm, 22.85mm, and 20.02mm) to resistance all bacteria activities, that stronger antibacterial activity compared with the lowest of sensitive antibiotics P10, AMP10, TET10, Eyr10, CF5, and CN10, respectively. Actually, the MIC was given good values ((125µg/ml, 62.5µg/ml, 125µg/ml, and

125µg/ml) in solvent (distilled water) and (31.25µg/ml, 62.5µg/ml, 3.90µg/ml, 7.81µg/ml, 1.95µg/ml, and 62.5µg/ml) with (solvent ethanol distilled water) by (MWE) respectively.

However, the orbital shaker technique with distilled water extract never given zones of inhibition. While the (ethanol: distilled water) extract had given a maximum antibacterial activity against the growth of bacteria in the concentration of 100µL/disk in diameter zones (19.28mm, 18.93mm, 13.87mm, 14.88mm, 15.45mm, and 15.14), with the highest sensitivity of MIC values (500µg/ml, 31.25µg/ml, 250µg/ml, 62.5µg/ml, 500µg/ml, and 125µg/ml) respectively, compared with least of antibiotics. Our study agrees with (Ramezani, Fazli-Bazzaz, *et al.* 2004) Indicated all the extracts were given antibacterial activity against two gram-positive bacteria, and this finding may partly justify the traditional use of *Artemisia* species.

In fact, in our finding the *Artemisia hasskchitii boiss* extract by conventional technique more significant compared to microwave technique and orbital shaker extraction, to give highest antibacterial activity, and better than antibiotic to prevent growth and bacteria activity.

Table 4.78. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Artemisia hasskchitii* boiss solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis / Extract Solvents of Plant with bacteria dilution assay %	C E I.Z. (mm)	W	50%	13.30	13.69	13.13	15.55	13.93	11.89
			70%	14.60	14.56	19.18	18.97	16.96	14.37
			100%	18.69	17.70	20.52	20.05	21.08	18.82
		W:E	50%	12.38	17.95	11.82	15.81	15.51	15.52
			70%	16.40	19.86	13.83	16.34	18.82	18.63
			100%	18.34	21.04	16.75	17.59	18.48	19.05
	C E MIC (µg/ml)	W	≥500	3.90	31.25	15.62	31.25	62.5	3.90
		W:E	≥500	7.81	3.90	125	500	125	7.81
	MW E I.Z. (mm)	W	50%	11.35	11.35	-	-	-	-
			70%	13.67	15.09	-	-	14.69	14.61
			100%	14.50	16.28	-	15.43	18.41	15.41
		W:E	50%	16.85	15.98	15.52	18.76	15.33	17.24
			70%	18.58	17.48	18.57	20.32	21.64	18.69
			100%	18.86	19.76	21.54	20.34	22.85	20.02
	MW E MIC (µg/ml)	W	≥500	125	62.5	-	125	-	125
		W:E	≥500	31.25	62.5	3.90	7.81	1.95	62.5
	OS E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	12.50	-
			70%	12.09	-	11.35	15.32	14.62	-
			100%	19.28	18.93	13.87	14.88	15.45	15.14
	OS E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	500	31.25	250	62.5	500	125
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	N.E	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

Note: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=*Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa* (ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4=*Enterococcus faecalis* (ATCC 29212),B.5=*Enterobacter aerogenes*, B.6=*Klebsiella pneumonia*; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10µg , (CF) 5µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : Not Detected

4.9.7. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Prunus microcarpa* extracts against the bacteria with antibiotic (Antimicrobial Agent)

The *Prunus microcarpa* extracts were investigated to evaluate their antibacterial activity against six bacteria by three techniques with various solvents in concentrations 50, 70, and 100µL/disk. Evaluation of the antibacterial activity of these plant extracts was recorded in Table (4.79).

The results of conventional technique with distilled water revealed extract in the concentration of 50, 70, and 100µL/disk was observed maximum zone of inhibition with a diameter (23.07mm, 13.11mm, 18.29mm, 19.43mm, and 23.18mm), to resistance against of *Staphylococcus aureus* (ATCC 29213), *Pseudomonas aeruginosa* (ATCC 27853), *Enterococcus faecalis* (ATCC 29212), *Enterobacter aerogenes*, and *Klebsiella pneumonia* respectively, in comparison with the smallest of antibiotics. Thus, the results of MIC were higher effective values at (1.95µg/ml, 125µg/ml, 62.5µg/ml, 31.25µg/ml, and 1.95µg/ml) to against the activity of bacteria respectively. Whereas, the (ethanol: distilled water), showed a great variable of inhibitory zone against the effectiveness of bacteria (B.1, B.4, B.5, and, B6).in concentration 100µL/disk with diameter zones (12.93mm, 12.56mm, 12.93mm, and 16.74mm), with high proficiency of MIC values (500µg/ml, 500, 250µg/ml, and 500µg/ml), showed higher antibacterial activity compared to the lowest values of antibiotics (P10, AMP10, Eyr10, and CN10) respectively.

Secondly, that the microwave method in solvent of water was not discovered any zone of inhibition, while the (ethanol: distilled water) had given a maximum antibacterial activity with concentrations of 50, 70, and 100µL/disk, in diameter zones as the range between (8.61mm and 14.26mm), also the concentration of 100µL/disk, had an inhibitory effect in diameter zones (14.26mm, 10.75mm, 12.62mm, 13.79mm, 13.54mm, and 11.35), on the *Staphylococcus aureus* (ATCC 29213), *Pseudomonas aeruginosa* (ATCC 27853), *Enterococcus faecalis* (ATCC 29212), *Enterobacter aerogenes*, and *Klebsiella pneumonia*, respectively. The MIC of the most effective plant extracts at values of (125µg/ml, 500µg/ml, 250µg/ml, 500µg/ml, 125µg/ml, and 125µg/ml) showed more antibacterial activity compared to the least of antibiotics against all the organisms respectively.

However, the orbital shaker with solvent (DW) did not detect any inhibition zones, While that the (E: DW), showed a high antibacterial in the concentration of 100µL/disk with

diameter zone(10.09mm), to resistance against *Enterococcus faecalis* (ATCC 29212), also the antibacterial extraction of *Prunus microcarpa* was more effective than antibiotics Penicillin, Ampicillin, Gentamycin at measure (8.27 µg/ml, 7.8µg/ml, and 10.71µg/ml), thus, the MIC value was highly sensitive and its minimum inhibition concentration reached to 250µg/ml to prevent the growth microorganism.

In general, our study suggests that *Prunus microcarpa*, obtained the highest inhibitory zone during microwave technique with (ethanol solvent) and conventional extraction with (distilled water), which was more effective antibacterial activity than antibiotics to avoid the bacteria.



Table 4.79. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Prunus microcarpa* solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis /Extract Solvents of Plant with bacteria dilution assay %	C/E I.Z. (mm)	W	50%	17.31	10.18	-	11.50	14.50	18.05
			70%	22.09	11.47	-	16.92	16.10	21.68
			100%	23.07	13.11	-	18.29	19.43	23.18
		W:E	50%	11.96	-	-	-	-	-
			70%	11.10	-	-	-	9.69	-
			100%	12.93	-	-	12.56	12.93	16.74
	C/E MIC (µg/ml)	W	≥500	1.95	125	-	62.5	31.25	1.95
		W:E	≥500	500	-	-	500	250	500
	MW/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	10.85	8.61	9.65	12.35	11.14	11.07
			70%	11.92	10.38	9.97	13.38	12.52	10.36
			100%	14.26	10.75	12.62	13.79	13.54	11.35
	MW/E MIC (µg/ml)	W	≥500	-	-	-	-	-	-
		W:E	≥500	125	500	250	500	125	125
	OS/E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	10.09	-	-
	OS/E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	-	-	-	250	-	-
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	No	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=Staphylococcus aureus (ATCC 29213), B.2= Pseudomonas aeruginosa (ATCC 27853), B.3= Bacillus subtilis (ATCC 6633), B.4=Enterococcus faecalis (ATCC 29212),B.5=Enterobacter aerogenes, B.6=Klebsiella pneumonia; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : Not Detected.

4.9.8. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Silybum Marianum* extracts against the bacteria with antibiotic (Antimicrobial Agent)

According to the results in Table (4.80), The conventional method with(distilled water) of *Silybum Marianum* extract, gave high effectiveness of antibacterial zones in the concentration of 100µL/disk, were examined against *Pseudomonas aeruginosa* (ATCC 27853), and *Klebsiella pneumonia* at measures (15.44mm), and (15.51mm), in comparison better than antibiotics (Penicillin, Ampicillin, Erythromycin, and Gentamycin), and the best results of MIC were obtained for B.2 and B.6 at values 62.5µg/ml, and 1.8µg/ml respectively. While the (ethanol: distilled water) extract, had got maximum inhibitory zones in the concentration of 100µL/disk, at diameter (15.4mm, 9.44mm, and 12.34mm) respectively, with stronger effective of MIC at values (250µg/ml, 500µg/ml, and 125µg/ml), obtained a more antibacterial activity in comparison with lowest values of antibiotic; Penicillin, Ampicillin, and Erythromycin, to resistance against three bacteria *Pseudomonas aeruginosa* (ATCC 27853), *Bacillus subtilis* (ATCC 6633), and *Enterobacter aerogenes*, respectively.

On the other hand, the antibacterial activity in distilled water did not obtain any zone of inhibition by microwave technique, whereas in with (ethanol: distilled water) was obtained an effective antibacterial activity in the concentration of 100µL/disk with a diameter (9.58mm, 9.49mm, and 8.09mm), with sufficient of MIC at values (500µg/ml, 62.5µg/ml, Although, and 250µg/ml) respectively. Although, the antibacterial of *Silybum Marianum* extract of less than in compared with antibiotics, to prevent growth three of bacteria.

Also, the technique of orbital shaker with (crude distilled water) was observed not detect of inhibition zone, excepting of solvent (ethanol: distilled water) showed maximum inhibition zones in the concentration of 100µL/disk at (13.22mm, 14.32mm, 11.02mm, and 10.86mm) of plant extract are observed against *Staphylococcus aureus* (ATCC 29213), *Enterococcus faecalis* (ATCC 29212), *Enterobacter aerogenes*, and *Klebsiella pneumonia*; compared with the smallest zone of antibiotic P10, AMP10, C30, Eyr10, and CN 10 respectively. Besides the MIC values were high proficiency at 125µg/ml, 62.5µg/ml, 250µg/ml, and 500µg/ml respectively, to against the bacteria. Our finding agreement with (Khan *et al.* 2014) reported that the *silybum marianum*, was particularly effective for liver disorders among these herbal formulations. Furthermore, the *Silybum Marianum* (*silymarin*) was found to be highly effective against all gram-positive bacteria and to have moderate to strong antibacterial activity against pathogens

studied(Shah *et al.* 2011). Our study mention that the orbital shaker technique with solvent (ethanol: distilled water) better and more effective than the (microwave and conventional extraction) to give a great inhibition zone on the bacteria activity.



Table 4.80. Inhibition zones (I.Z. mm) and minimum inhibitory concentration (MIC) of the *Silybum Marianum* solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis / Extract Solvents of Plant with bacteria dilution assay %	C E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	12.21	-	-	-	-
			100%	-	15.44	-	-	-	15.51
		W:E	50%	-	-	-	-	-	-
			70%	-	-	7.77	-	-	-
			100%	-	15.4	9.44	-	12.34	-
	C E MIC (µg/ml)	W	≥500	-	62	-	-	-	1.8
		W:E	≥500	-	250	-	-	-	-
	MWE I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	9.58	-	9.49	8.09
	MW E MIC (µg/ml)	W	≥500	-	-	-	-	-	-
		W:E	≥500	-	-	500	-	62.5	250
	OSE I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	13.22	-	-	14.32	11.02	10.86
	OS E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	125	-	-	62.5	250	500
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	N.E	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=*Staphylococcus aureus* (ATCC 29213), B.2= *Pseudomonas aeruginosa*(ATCC 27853), B.3= *Bacillus subtilis* (ATCC 6633), B.4=*Enterococcus faecalis* (ATCC 29212),B.5=*Enterobacter aerogenes*, B.6=*Klebsiella pneumonia*; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : No zone of inhibition.

4.9.9. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Pistacia eurycarpa* Yale extracts against the bacteria with antibiotic (Antimicrobial Agent)

From the Table (4.81). The crude extracts obtained by a conventional method with (distilled water) from the *Pistacia eurycarpa* Yale, tested for antibacterial activities against five bacteria, *Pseudomonas aeruginosa* (ATCC 27853), *Bacillus subtilis* (ATCC 6633), *Enterococcus faecalis* (ATCC 29212), *Enterobacter aerogenes*, and *Klebsiella pneumonia* in the concentration of 100 μ L/disk at measure (12.87mm, 18.03mm, 12.21mm, 9.65mm, and 10.96mm), showed higher antibacterial activity compared with antibiotics Penicillin, Ampicillin, Erythromycin, and Gentamycin. While (ethanol: distilled water) extract had antibacterial activity against as follows; *Staphylococcus aureus* (ATCC 29213), *Pseudomonas aeruginosa* (ATCC 27853), *Bacillus subtilis* (ATCC 6633), *Enterobacter aerogenes*, and *Klebsiella pneumonia*, in the concentration of 100 μ L/disk, with effective inhibition zones of (9.85mm, 7.98mm, 9.42mm, 9.38mm, and 10.11mm) showed less antibacterial activity. Thus, the antibacterial activity of the crude distilled water, and ethanol: distilled water by microwave extraction in the concentration of 100 μ L/disk were showed high effectiveness of the antibacterial activity against (B.1) at measure zone (10.26mm) and (B.1, B.2, B.3, B.4, B.5, and B.6) at diameters (16.53mm, 17.39mm, 17.2mm, 15.01mm, 19.32mm, and 21.27mm) respectively, obtained in distilled water, and ethanol: distilled water. However, the antibacterial extract from *Pistacia eurycarpa yale*, was better than in comparison to the antibiotic, therefore the MIC was shown the highest concentrations at (250 μ g/ml) in (DW), and (125 μ g/ml, 500 μ g/ml, 15 μ g/ml, 500 μ g/ml, 125 μ g/ml, and 390 μ g/ml) with (E: DW) respectively, to prevents the growth of a microorganism.

Furthermore, the orbital shaker technique no registered any data of inhibition zone with a solvent of water. Whereas, with a solvent of (ethanol: water) detected a large growth of inhibition zones in concentration 100 μ L/disk to resistance against four bacteria as namely (*Staphylococcus aureus*, *Enterococcus faecalis*, *Enterobacter aerogenes*, and *Klebsiella pneumonia*) at diameter (15.41mm, 14.70mm, 11.69mm, and 16.08mm), had better than compared with antibiotics (P10, AMP10, C30, Eyr10, and CN10) respectively. However, the MIC was determined a big effective at values (31 μ g/ml, 125 μ g/ml, 500 μ g/ml, and 62.5 μ g/ml) against the growth of bacteria. Our finding in line with (Kordali *et al.* 2003), reported that the extracts of *Pistacia vera*, *Pistacia terebinthus*, and *Pistacia lentiscus* significantly inhibited growth against pathogenic agricultural fungi.

Our results showed the inhibition zone of antibacterial activities was obtained highest zone with microwave technique with solvent (distilled water) better than to compare with an orbital shaker, conventional extraction, and antibiotics against the bacterial activity.



Table 4.81. Inhibition zones (I.Z.mm) and minimum inhibitory concentration (MIC) of the *Pistacia eurycarpa* Yale solvents extract against the bacteria with some Antibiotic Sensitivity Agent.

	Techniques Analysis	S.	Con.	B.1	B.2	B.3	B.4	B.5	B.6
Technique Analysis / Extract Solvents of Plant with bacteria dilution assay %	C E I.Z. (mm)	W	50%	-	9.04	13.03	9.93	8.58	8.75
			70%	-	9.34	15.43	10.06	8.72	8.55
			100%	-	12.87	18.03	12.21	9.65	10.96
		W:E	50%	-	-	-	-	-	7.2
			70%	7.71	-	-	-	-	7.89
			100%	9.85	7.98	9.42	-	9.38	10.11
	C E MIC (µg/ml)	W	≥500	-	31	15	250	500	500
		W:E	≥500	500	125	250	-	250	125
	MW E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	10.26	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	13.62	-	-	-	18.63
			100%	16.53	17.39	17.2	15.01	19.32	21.27
	MW E MIC (µg/ml)	W	≥500	250	-	-	-	-	-
		W:E	≥500	125	500	15	500	125	3.90
	OS E I.Z. (mm)	W	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	-	-	-	-	-	-
		W:E	50%	-	-	-	-	-	-
			70%	-	-	-	-	-	-
			100%	15.41	-	-	14.70	11.69	16.08
	OS E MIC (µg/m)	W	≥500	-	-	-	-	-	-
		W:E	≥500	31	-	-	125	500	62.5
Antibiotic (Antimicrobial Agent) / Susceptible	Penicillin	P 10	10 µg	12.94	11.09	14.3	8.27	10.02	10.61
	Ampicillin	AMP10	10 µg	12.83	10.59	N.E	7.8	11.42	12.64
	Tetracycline	TET10	10 µg	21.73	21.66	20.08	22.23	22.37	21.54
	Chloramphenicol	C30	30 µg	24.7	20.72	24.91	13.7	26.13	28.49
	Cefotaxime	CTX 30	30 µg	29.53	26.8	28.25	26.36	27.48	28.41
	Erythromycin	Eyr 10	10µg	11.6	16.45	11.37	13.14	8.8	12.98
	Ciprofloxacin	CF 5	5 µg	22.76	18.58	21.85	21.5	21.29	19.42
	Gentamycin	CN 10	10 µg	16.9	10.19	11.1	10.71	17.66	14.21

NOTE: Solvents extract and synthetic activities the bacteria dilution assay. Values are mean± SE of triplication sample, Technique analysis -: CE= Conventional Extraction; ME= Microwave Extraction; and OS/E = Orbital shaker Extraction, S =Solvent: W= Distilled water, and W: E= aqueous of Distilled water: Plant = 4 (g); Solvents =80(ml); Solvent: Solvent = 40+40 (ml): ZI: zone of inhibition Concentration, (50,70, and 100) %; MIC: minimum inhibition concentration = (500, 250,125,62.5, 31.25, 15.62, 7.81, 3.90, and 1.95)%; Bacteria: B.1=Staphylococcus aureus (ATCC 29213), B.2= Pseudomonas aeruginosa(ATCC 27853), B.3= Bacillus subtilis (ATCC 6633), B.4=Enterococcus faecalis (ATCC 29212),B.5=Enterobacter aerogenes, B.6=Klebsiella pneumonia; Antibiotic (Antimicrobial Agent) / Susceptible: Penicillin G: P10 µg, Ampicillin (other) : AMP10 µg, Tetracycline : TET 10 µg, Chloramphenicol : C 30 µg, Cefotaxime : CTX 30 µg, Erthromycin : Eyr 10 µg , (CF) 5 µg : Ciprofloxacin, and, Gentamycin : CN10 µg; - : No zone of inhibition.

Our study showed, that the inhibition zone (I.Z) and Minimum Inhibitory Concentration (MIC) of our plant extracts were obtained the highest antibacterial activity during the three methods as (C/E, MW/E, and OS/E) with solvents (distilled water), and (ethanol: distilled water). Our study showed, that the inhibition zone (I.Z) and Minimum Inhibitory Concentration (MIC) of our plant extracts were obtained the highest antibacterial activity during the three methods as (C/E, MW/E, and OS/E) with solvents (distilled water), and (ethanol: distilled water). While in our study no utilized hexane extract to antibacterial activity compared with other analysis, because the hexane extract was shown to have a very weak activity to gainst all growth bacteria in our research. That is inline with (Sprague *et al.* 2014), also agreement with (Islam *et al.* 2018) had reported that the methanol extract demonstrated considerable to modest antibacterial activity than n-hexane extract and dichloromethane in comparison with the standard antibiotic. Therefore, that the microwave and conventional techniques were given high antibacterial activity than the orbital shaker technique, moreover, all the three techniques by different solvents were showed antibacterial activity great compared with the least inhibition zone of antibiotics.

However, some of our plant extracts seem to have less antibacterial activity comparing to antibiotics such as (TET10, C30, CTX30, and CF5). Although, the positive point is it is natural. Because it helps in confirming the resistance of microorganisms to an antimicrobial factor and it observes the activity of new antimicrobial agents, by described as the lowest dilution or highest concentration of the extracts that inhibit the growth of organisms. (Omar *et al.* 2010). Moreover, the antimicrobial analysis using the agar well diffusion method and Minimum Inhibitory Concentration (MIC) values is been used by several researchers (Pavithra *et al.* 2010; Arora and Kaur 2007; Gurudeeban *et al.* 2010), this reports agreement with our study. Also our findings, approval with reports (Balaky *et al.* 2020; Galali *et al.* 2017), that indicate the antimicrobial activity in plants can be linked to the existence of biologically active compounds that inhibit bacterial growth. These findings may be especially important because as bacteria develop, they produce toxins that cause septicemia and enteritis As part of their survival strategy, plants may generate a large number of secondary molecules. (Houghton 2001).

Therefore, the existence of secondary molecules known as phytochemicals, such as alkaloids, flavonoids, terpenoids, tannins, saponins, anthraquinones, and glycosides, is thought to be responsible for certain plants' antimicrobial properties (Omojate *et al.* 2014). Since plants produce a variety of compounds with antimicrobial properties such as antibacterial activity,

they are good candidates for exploring new antibacterial drugs (Dahiru *et al.* 2006). Furthermore, the medicinal use of these plants resides in the phytochemical components that give rise to a definite pharmacological action on the human body (Akinmoladun *et al.* 2007), this report support the results we obtained from our study.

To the best of our knowledge, few studies have been published on the antimicrobial activity of our plants but not the same process. However, our study indicated, that the extract of our plants is a considering natural product and antibacterial source in the development of phytomedicines against drug-resistant infections.



5. CONCLUSION

To summarize, the studied plant extracts registered a significance in the chemical and biological characteristics such as extraction yield %, total condensed tannin, total phenol content, total flavonoid, total anthocyanin, phytochemical identification by LC/MS/MS, and analysis of mineral compounds by IC. The important pharmacological antibacterial activity of plant extracts on six bacteria was contrasted with nine synthetic antibiotics using the agar disk diffusion process and MIC value in this study.

1. The extraction yield % from all plant extracts significant value was seen through different techniques with five solvents. Especially, from plant *Prosopis farcta* highest value was observed using (ethanol: distilled water v/v) CE: 81.56 mg/kg. Also, the (DW: E v/v) with other solvents was given high amounts of yields in all plants.
2. Although, the total condensed tannin from all plants was significantly high using different methods with all the solvents. But the *Capparis Spinosa* in (distilled water: ethanol extract) shows the greatest amount (0.462 kg/mg), with the other plants during conventional extraction as P.1, P.2, and P.4 respectively. Also, the orbital shaker technique was containing high amounts of total condensed tannin in plants as P.6, P.7, P.8, and P.9. respectively, during utilized different solvents.
3. The total phenol content was a suitable source and a significant value ($p < 0.01$) through the various techniques and solvents. Especially, the orbital shaker obtained the highest value of total phenol from *Artemisia haussknechtii boiss* in (DW: E 3.980 mg GAE/g) extract, and better than comparing with other plants.
4. Consequently, the *Paliurus Spina Christi* extract in (distilled Water: ethanol v/v) has given the maximum value (0.328 mg QE/g) of total flavonoid through an orbital shaker method. While the conventional extraction displayed a significant amount ($p < 0.01$) from all plants.
5. Furthermore, the *Artemisia haussknechtii boiss* extract in solvent (distilled water) was detected to be the highest amount (176.59 mg/kg) ($p < 0.01$) of total anthocyanin contents during utilized orbital shaker method, as the preferred powerful effective method in comparison with other techniques. However, the total anthocyanin content of the conventional method was not recorded, possibly due to the intervention of the temperature of a conventional extraction on the crude extracts.
6. Our study observed that all plants possess high antioxidant activity in all solvents during (microwave, conventional, and orbital shaker) especially, the (ethanol), and (hexane:

ethanol) extracts. Also, our results indicated that the *Pistacia eurycarpa yale*, *Prosopis fracta*, and *Paliurus Spina Christi* were obtained the highest amounts of antioxidant activity. Especially, the *Pistacia eurycarpa yale* giving maximum (80.93 $\mu\text{g/mL}$, 93.19 $\mu\text{g/mL}$, 97.70 $\mu\text{g/mL}$, 90.35 $\mu\text{g/mL}$, and 85.90 $\mu\text{g/mL}$) in all solvents, that better and more than an antioxidant activity to compare with BHT standard, IC₅₀, and Trolox standard. Certainly, the highest antioxidant activity against,1-diphenyl-2-picrylhydrazyl radical with the lowest IC₅₀ $\leq$ - 4.30 mg/ml). Additionally, the superiority of all plants was more pronounced as compared to standard controls. Therefore, antioxidant activity and Trolox were shown to have strong and positive associations with correlation coefficients ranging from ($R \geq 0.99$), and ($R \geq 1$).

7. The results of LC-MS/MS analysis of all plant extract indicates that it can be used as a new source of vanillic acid and fumaric acid. Furthermore, the highest phenolic compounds including (resveratrol, gallic acid, quercetin, phloridzin dehydrate, ellagic acid, and silymarin, caffeic acid) were seen in *Eremurus Spectabilis*, *Prosopis farcta*, *Capparis Spinosa* fruit, *Paliurus spinachristi* fruit, *Portulaca oleracea*, and *Silybum, Marianum* respectively. On the other hand, for the phenolic compounds, the *Artemisia haussknechtii*, and *Pistacia eurycarpa yale* was the highest source.
8. Chemical ions such as (F^- , ClO_2^- , BrO_3^- , Cl^- , NO_2^- , Br^- , NO_3^- , PO_4^{3-} , SO_4^{2-} , Li^+ , Na^+ , NH_4^+ , K^+ , Mg^{2+} , and Ca^{2+}) were detected in all the plants. Moreover, the ions of (K^+ , NO_3^- , PO_4^{3-}) were the highest quantities in all the plants, particularly, the plants of *Artemisia haussknechtii* and *Portulaca oleracea* were the richest sources of ions (K^+ , Mg^{2+} , Cl^- , PO_4^{3-} , SO_4^{2-} and Ca^{2+})
9. Finally, the inhibition zone and minimum inhibitory concentration of our plant extracts possessed the highest antibacterial activity during three methods in (distilled water), and (ethanol: distilled water v/v). Additionally, the plant *Artemisia haussknechtii* and *Eremurus Spectabilis* Bieb had the strongest antibacterial activity with zones inhibition between (15.14 mm - 22.85 mm), and (11.55 mm-18.97 mm) respectively, with best the Minimum Inhibitory

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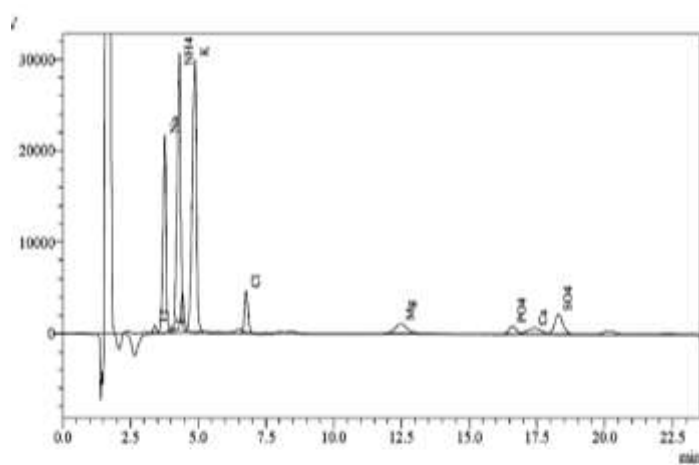
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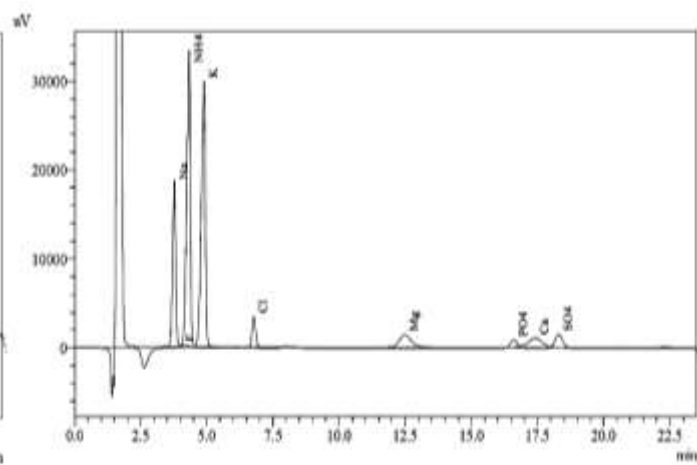
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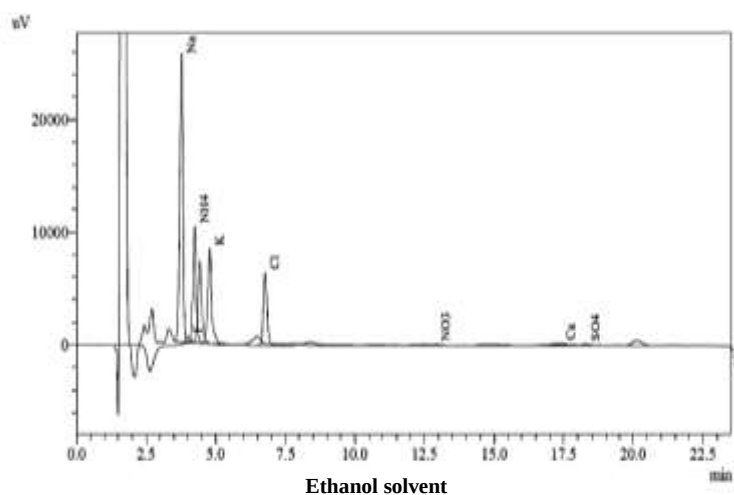
APPENDIX



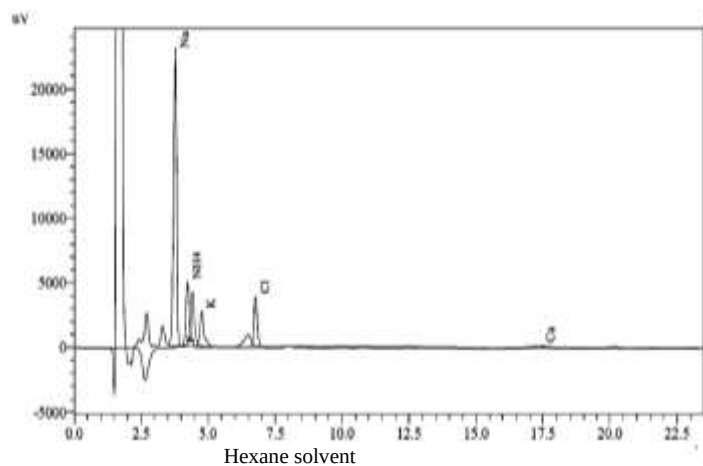
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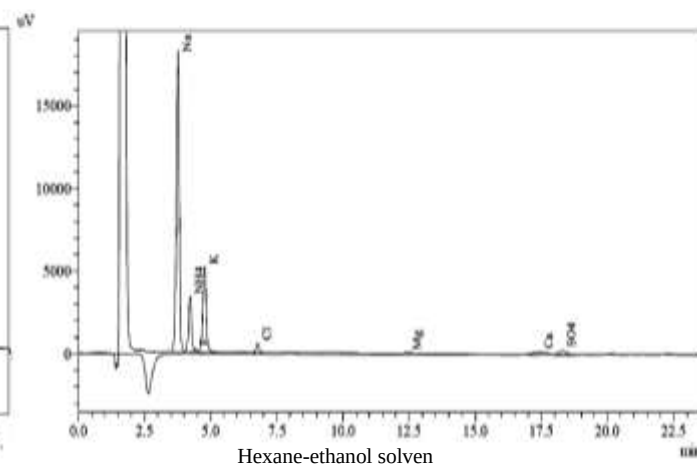
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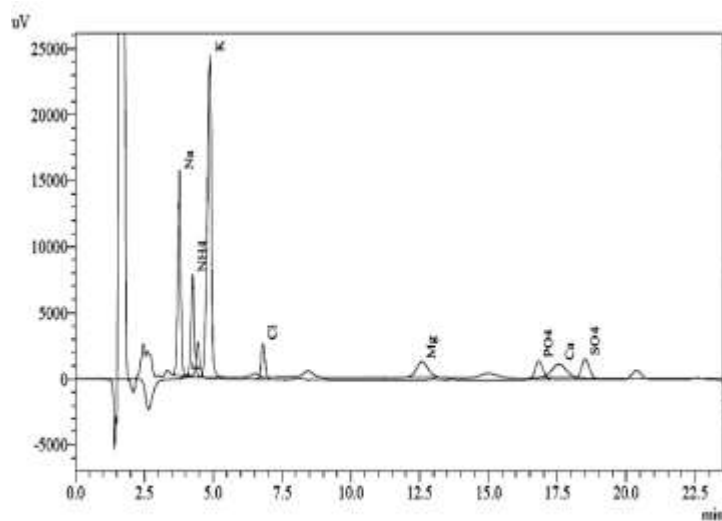
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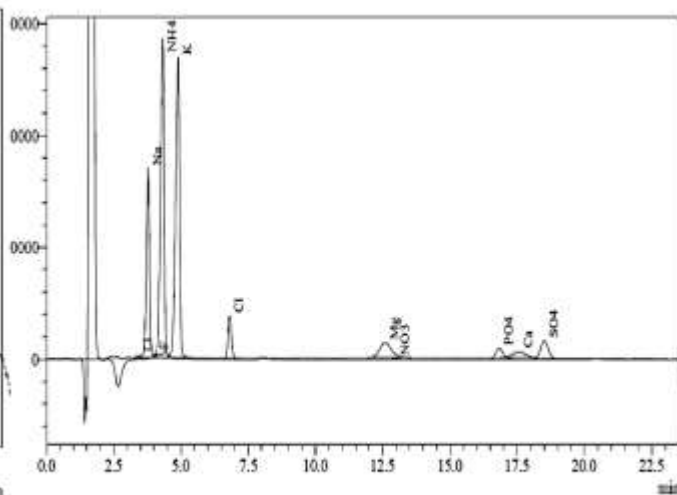
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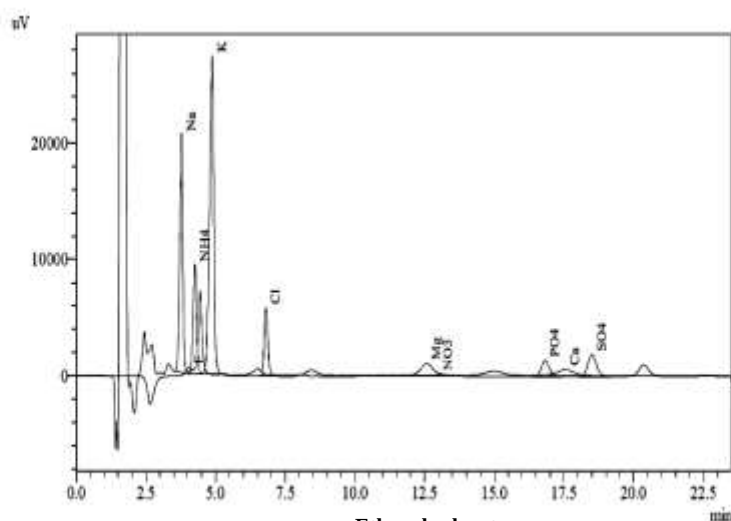
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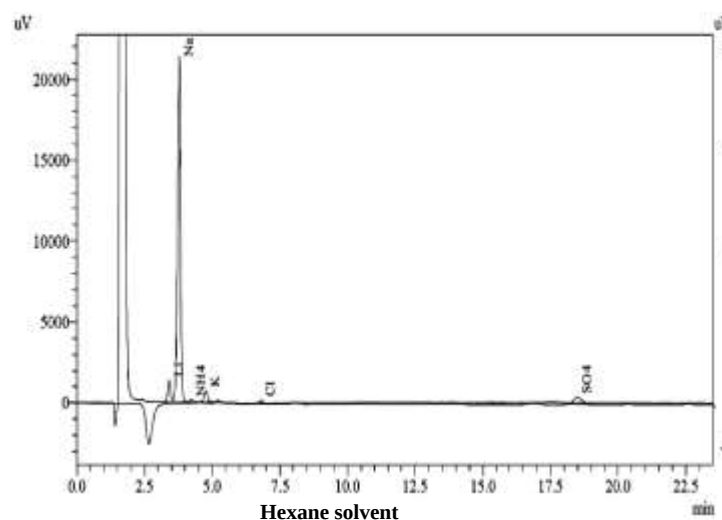
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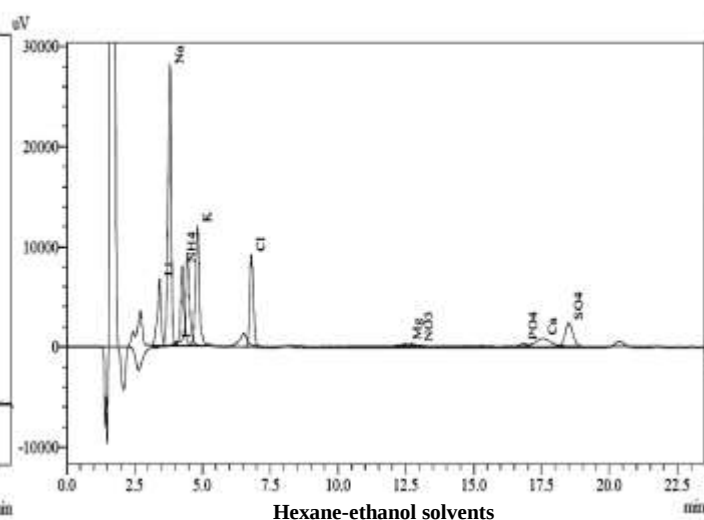
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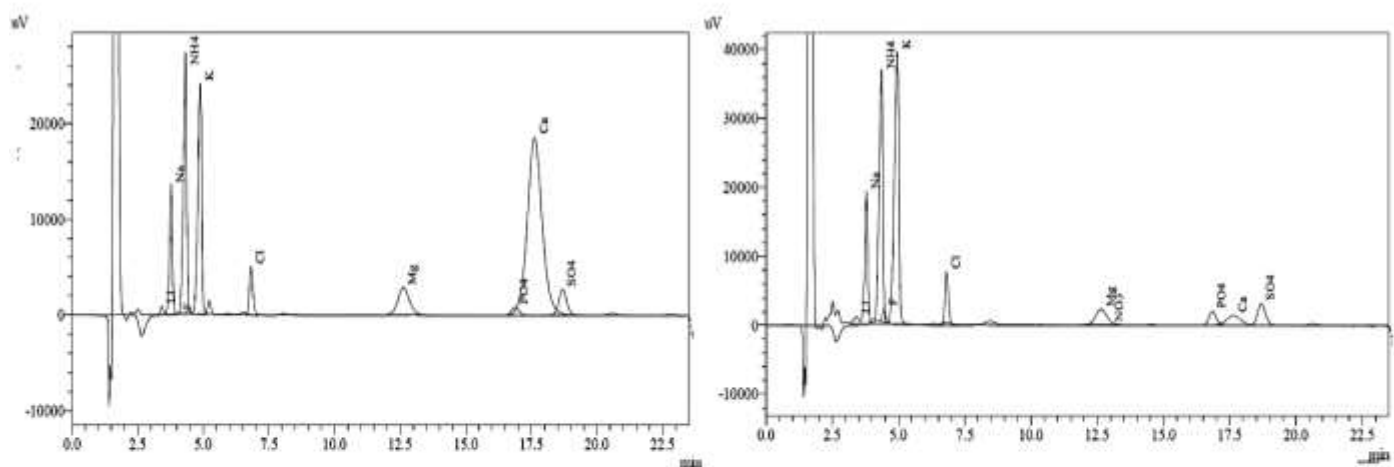
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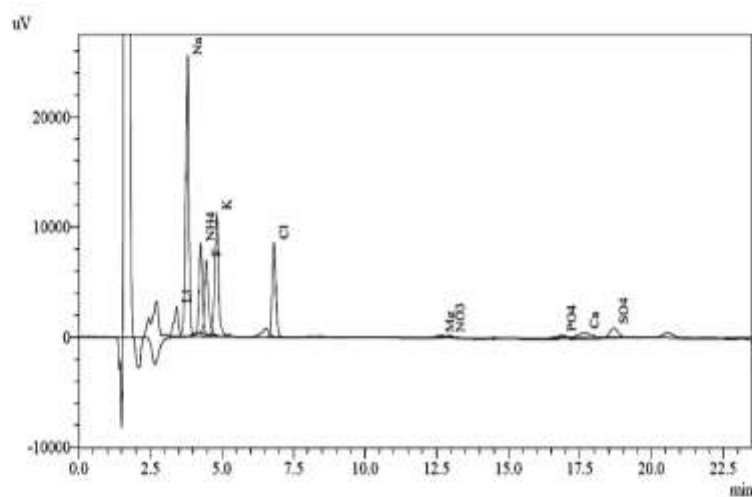


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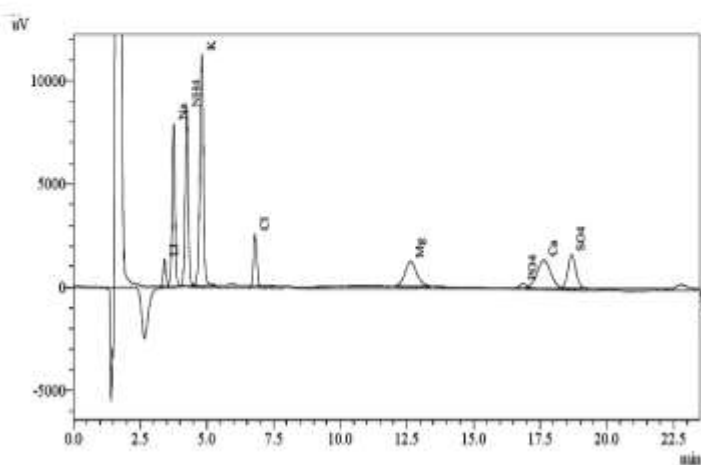


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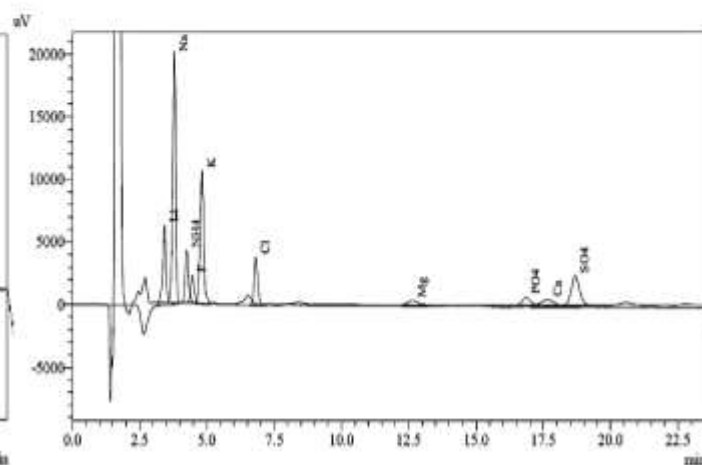
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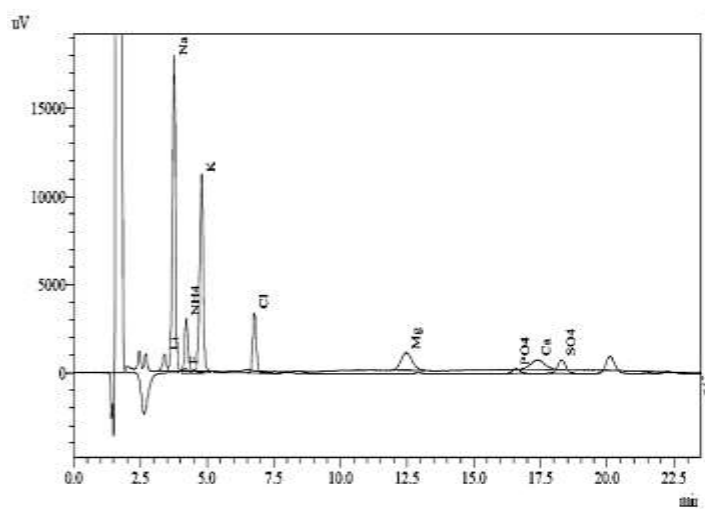


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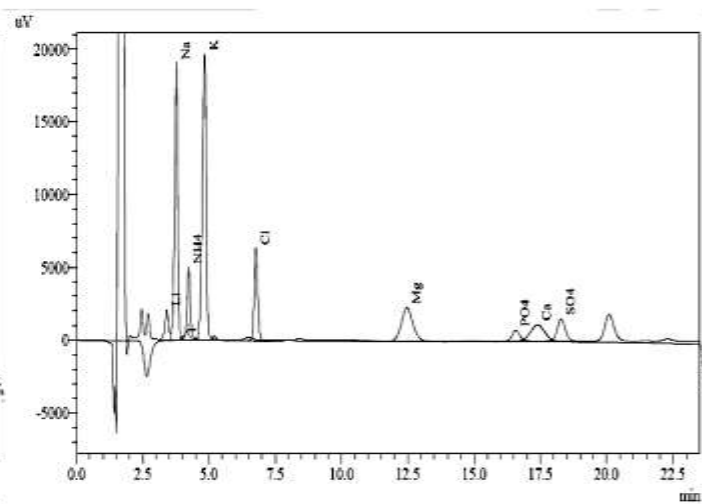


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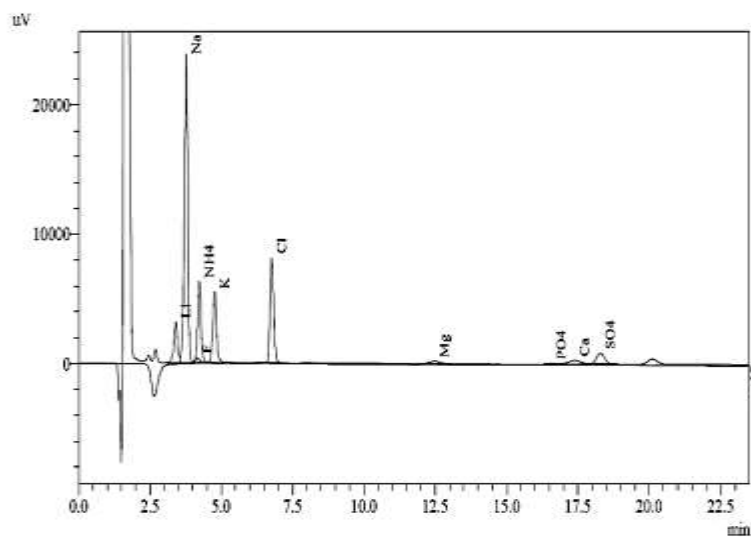
Figure 0.1. Quantitative result of *Eremurus Spectabilis Bieb* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by Ion Chromatography



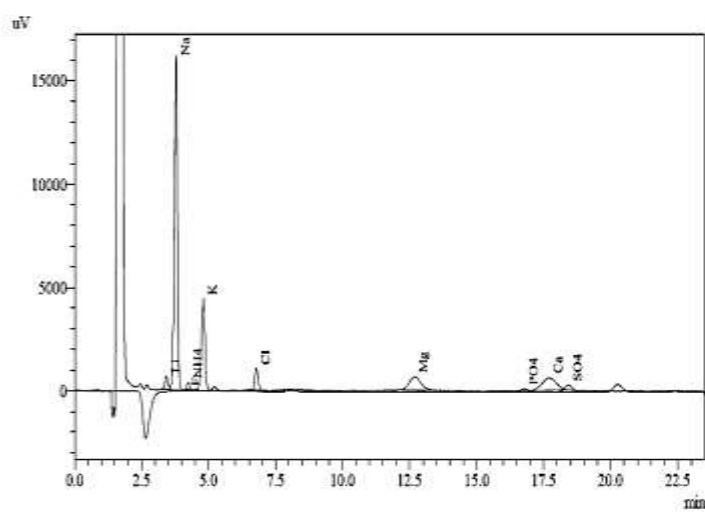
Water-ethanol solvents



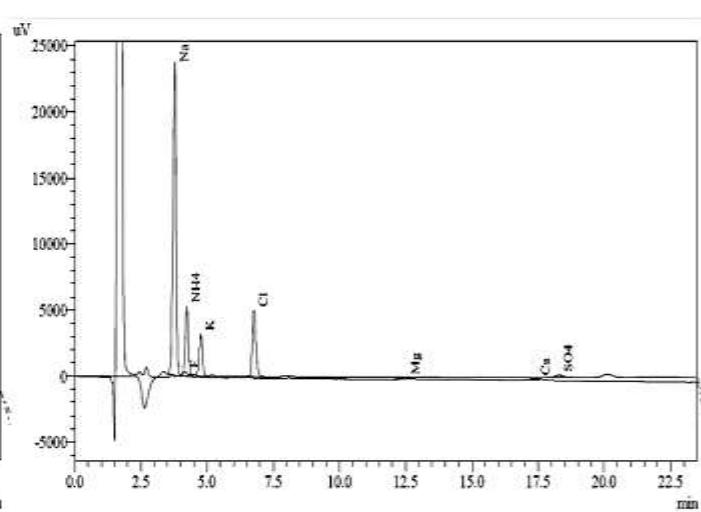
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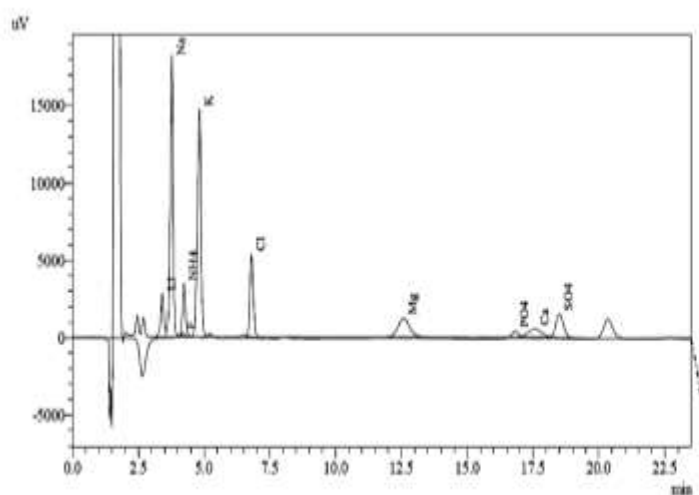
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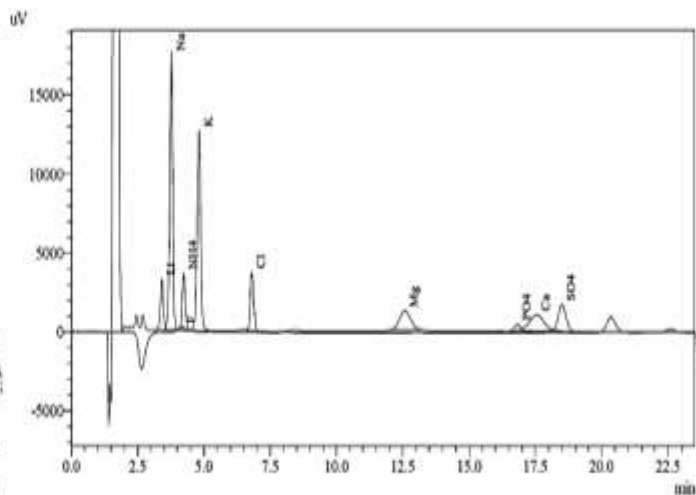
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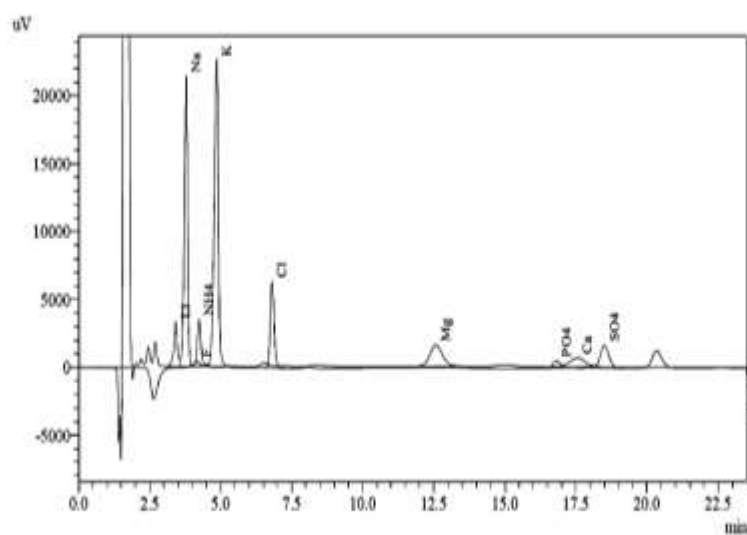
Hexane-ethanol solvents



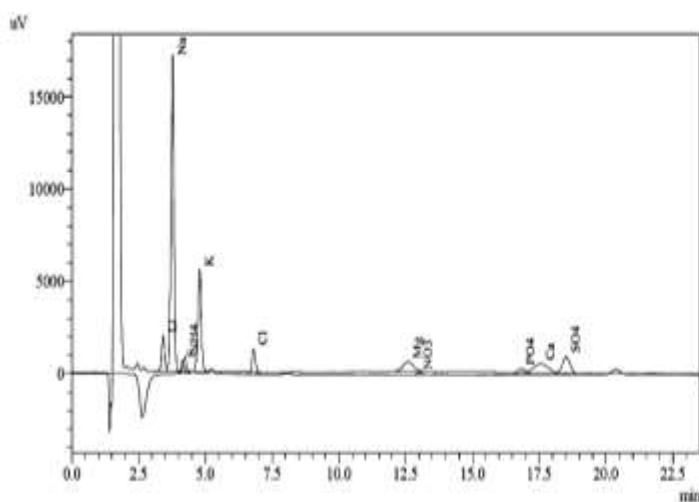
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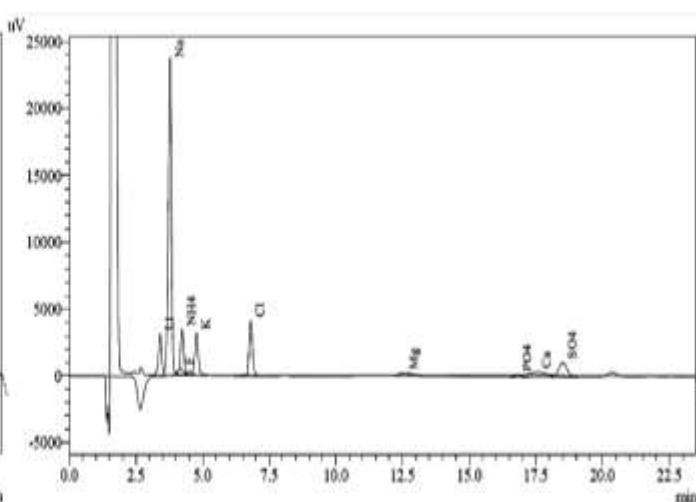
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Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

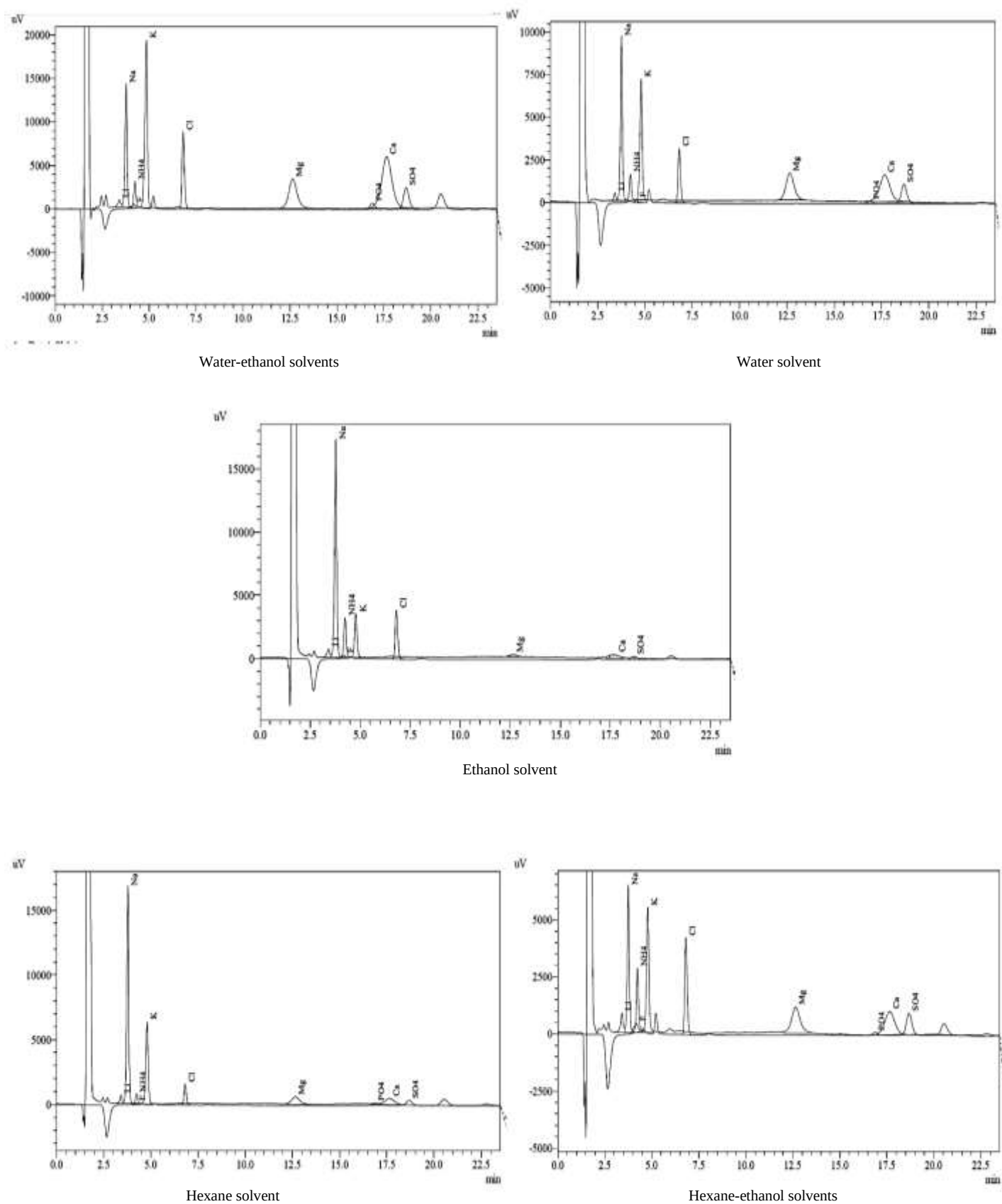
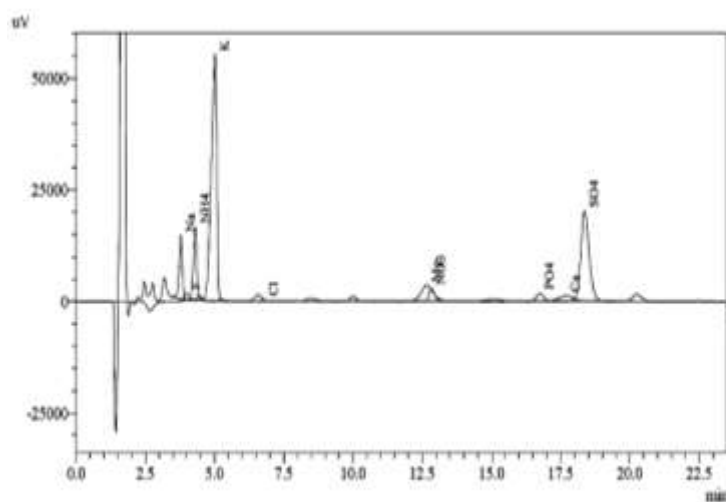
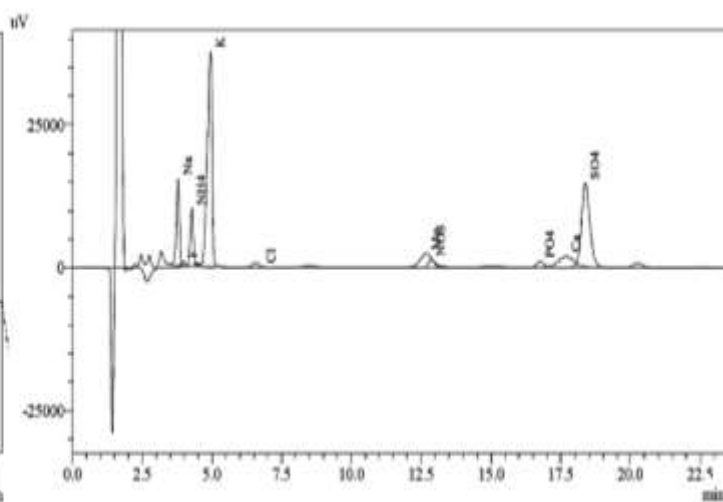


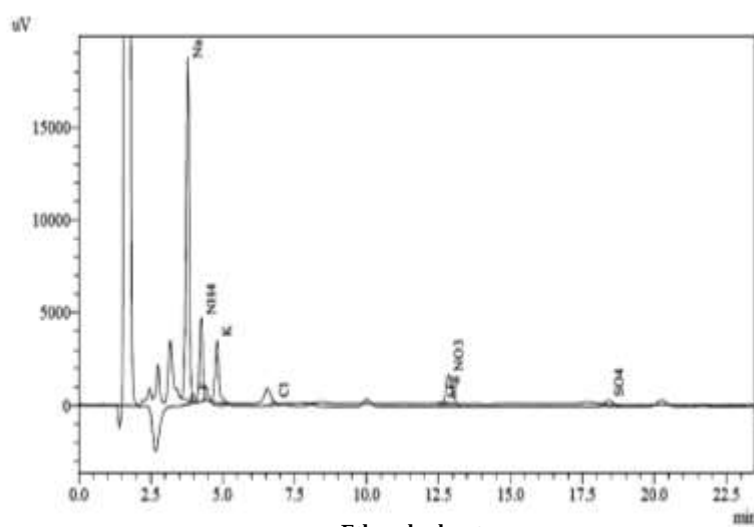
Figure 0.2. Quantitative result of *Prosopis farcta* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



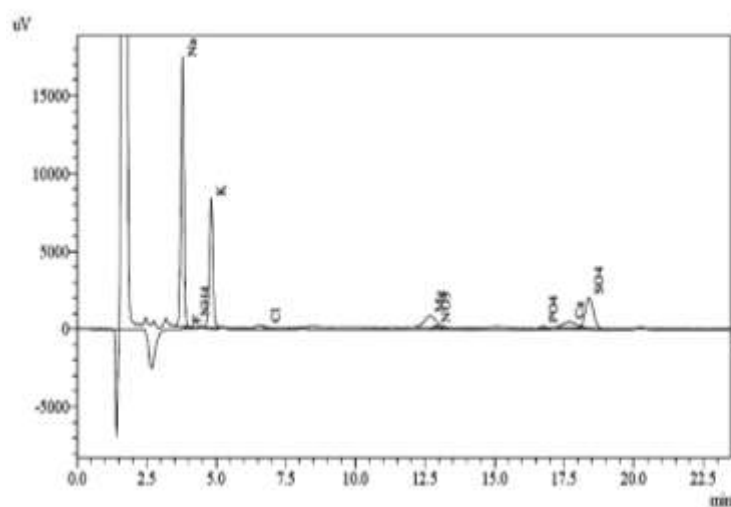
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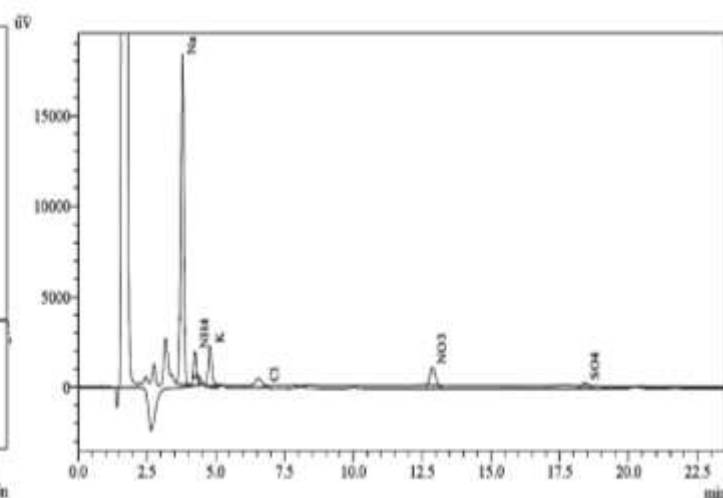
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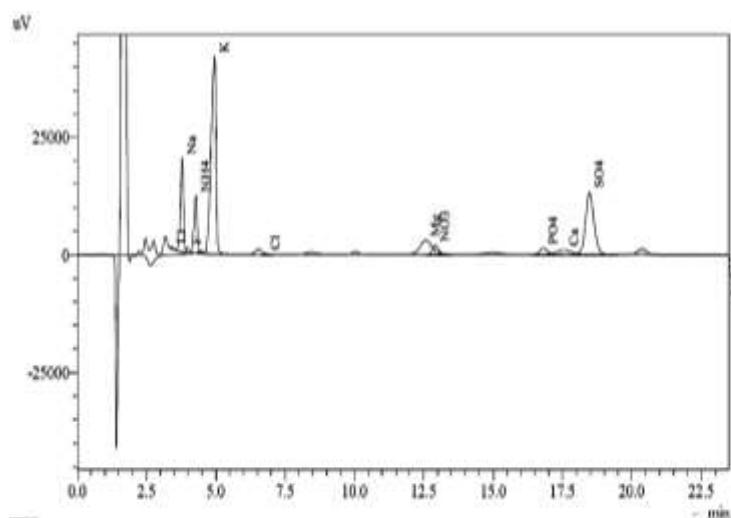
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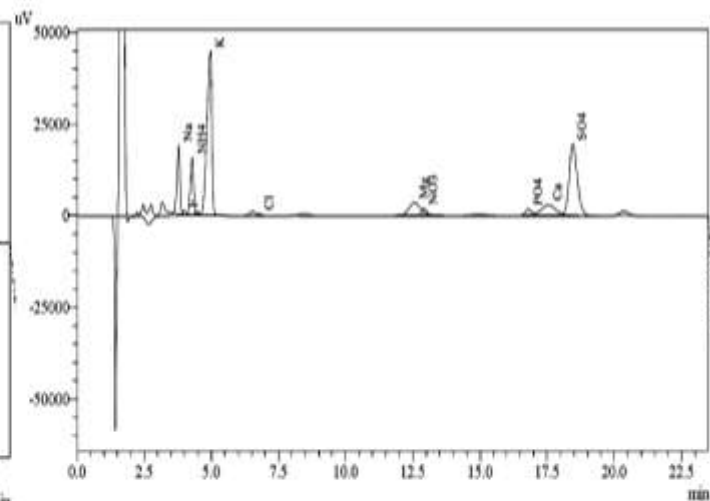
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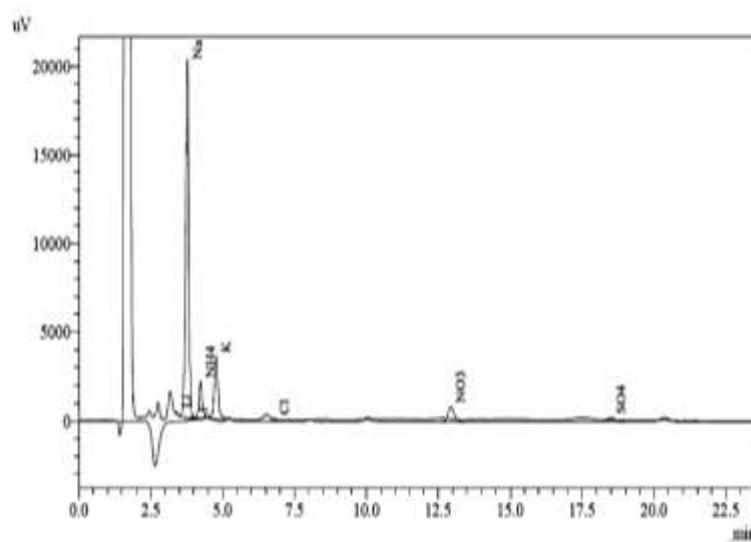
Hexane-ethanol solvents



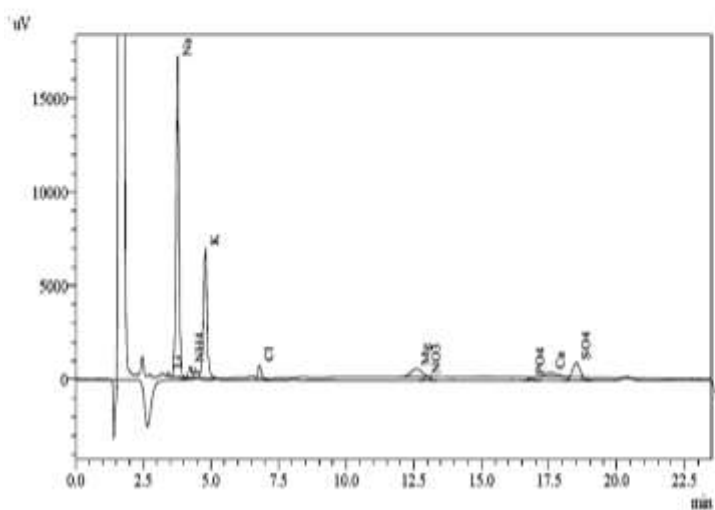
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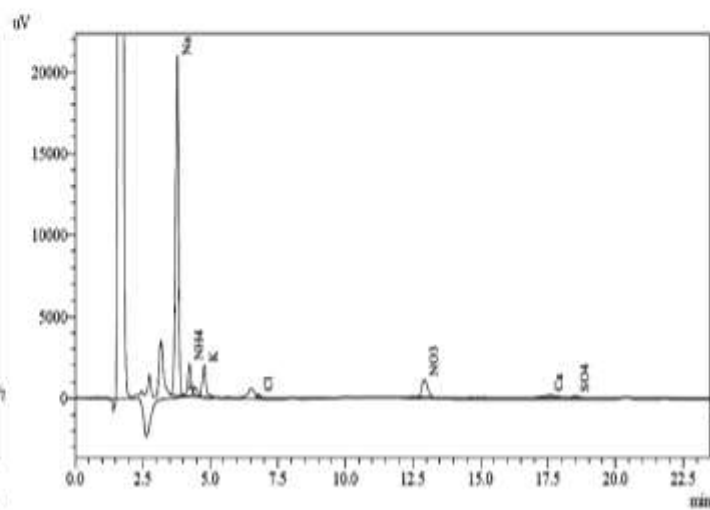
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Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

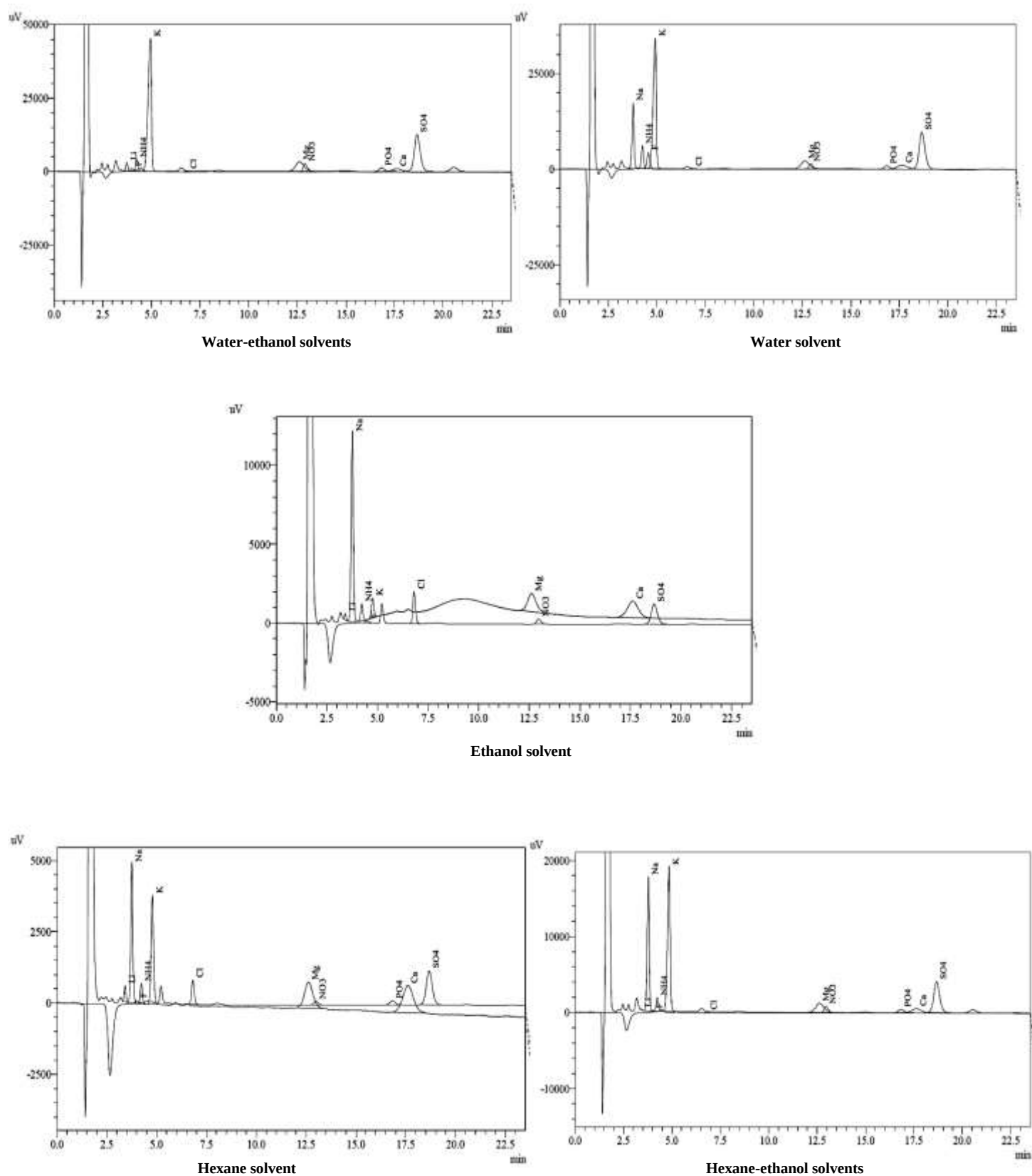
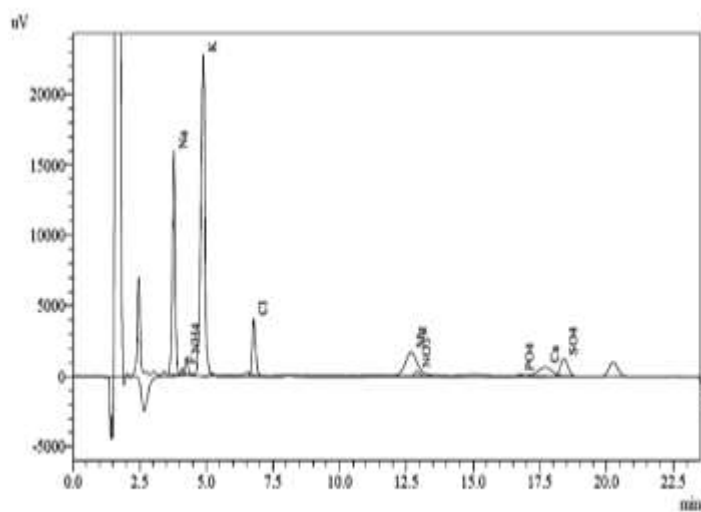
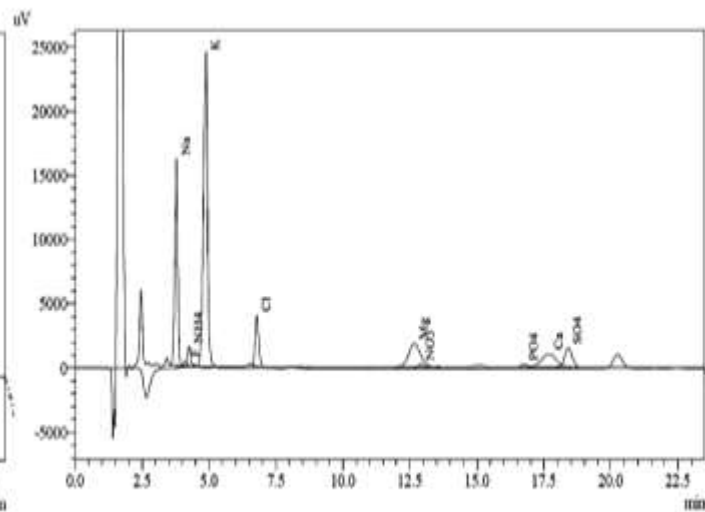


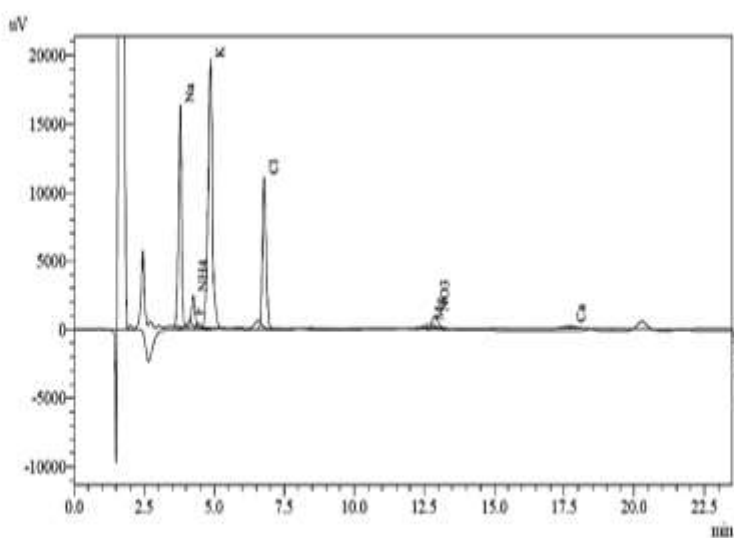
Figure 0.3. Quantitative result of *Capparis spinosa* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



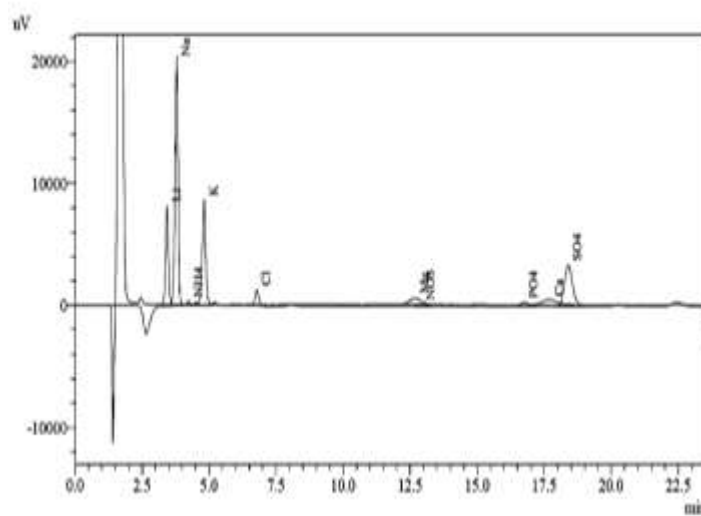
Water-ethanol solvents



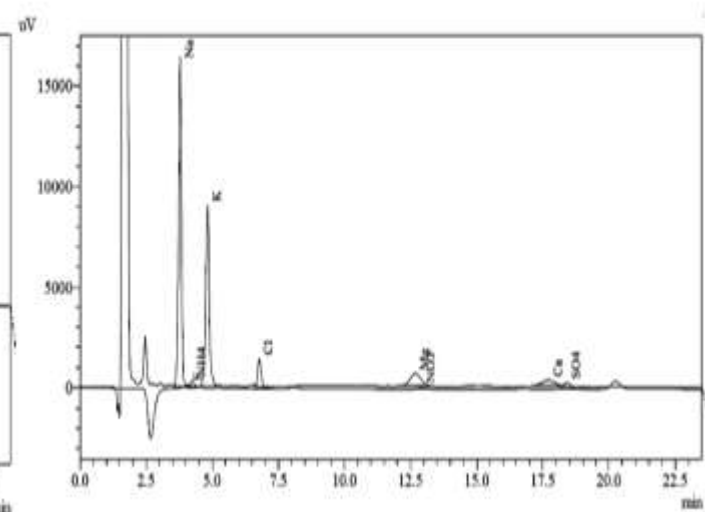
Water solvent



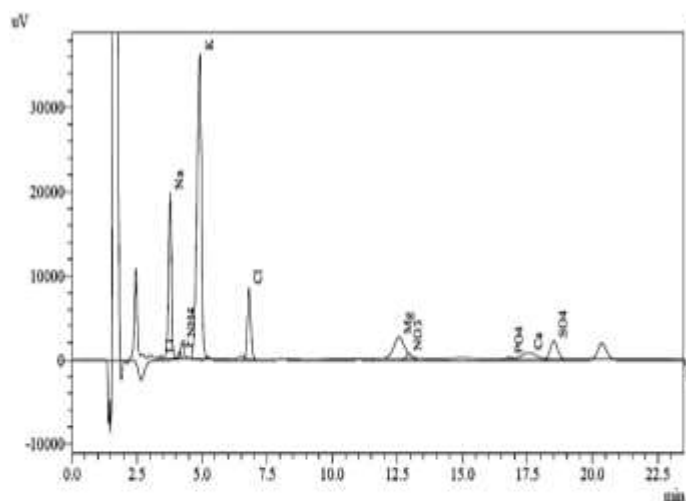
Ethanol solvent



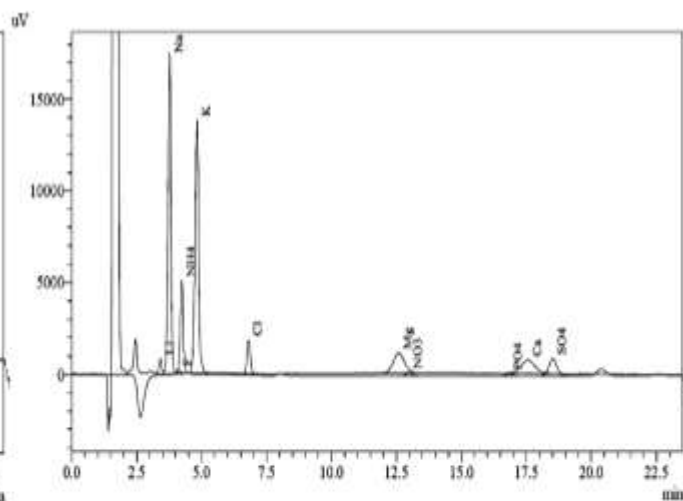
Hexane solvent



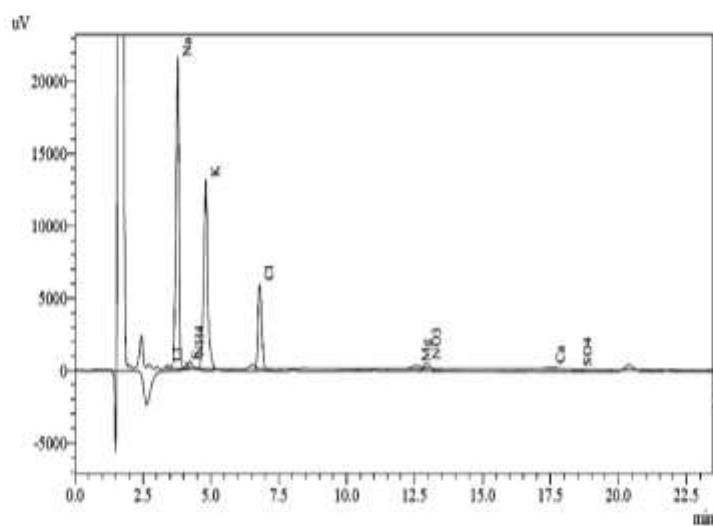
Hexane-ethanol solvents



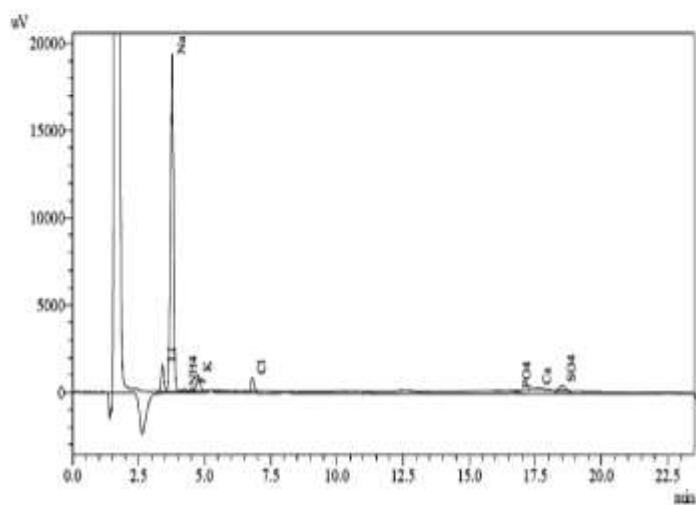
Water-ethanol solvents



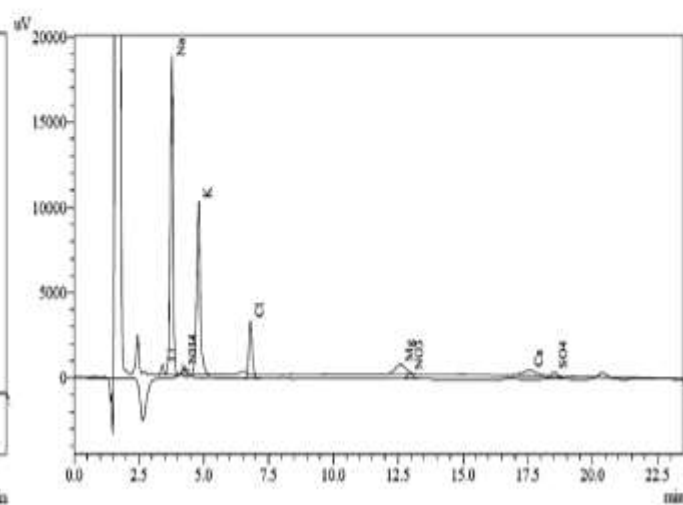
Water solvent



Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

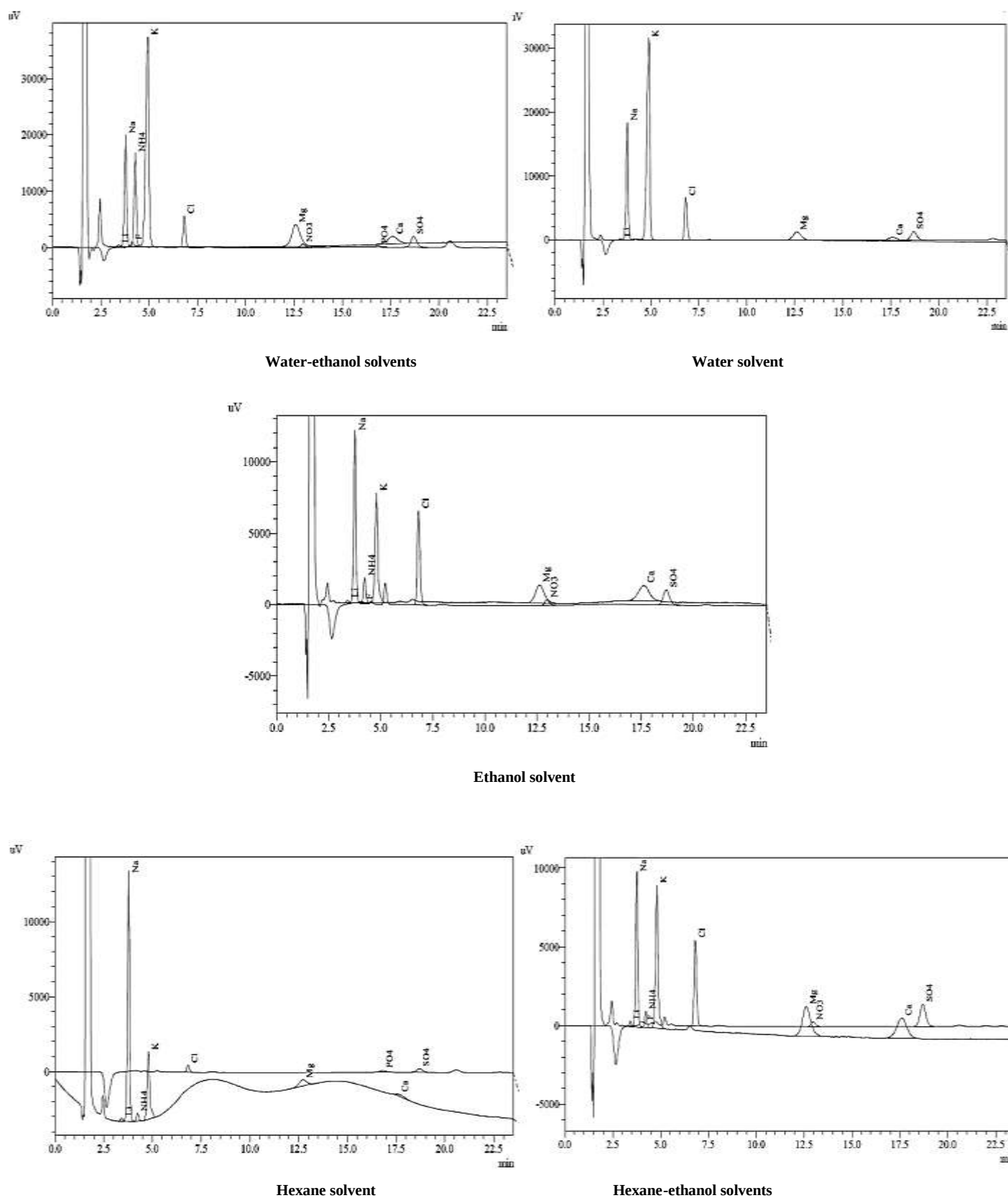
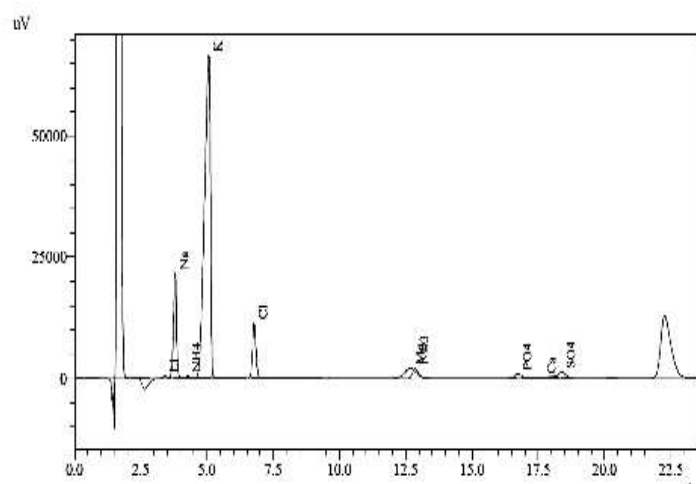
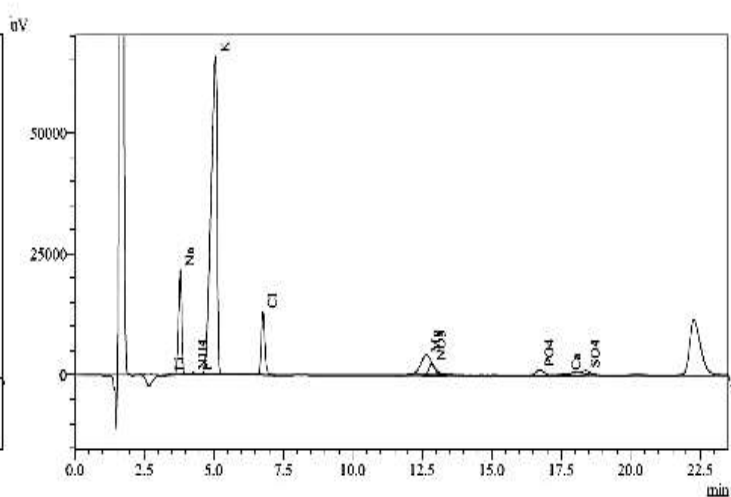


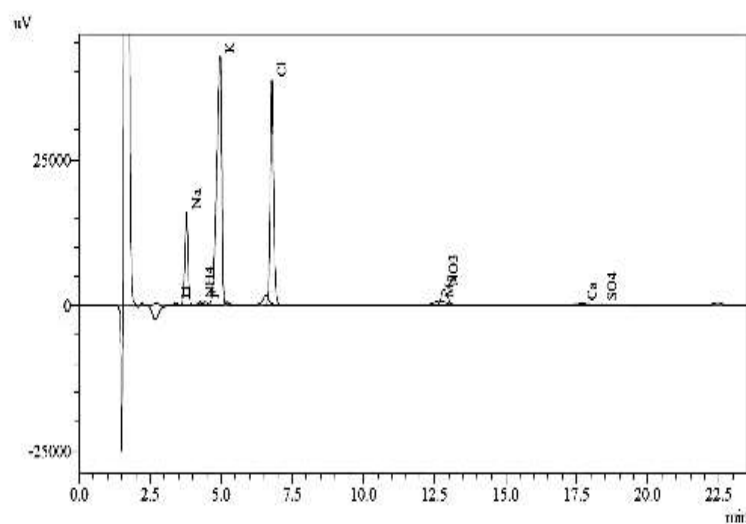
Figure 0.4. Quantitative result of *Paliurus spina Christi* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



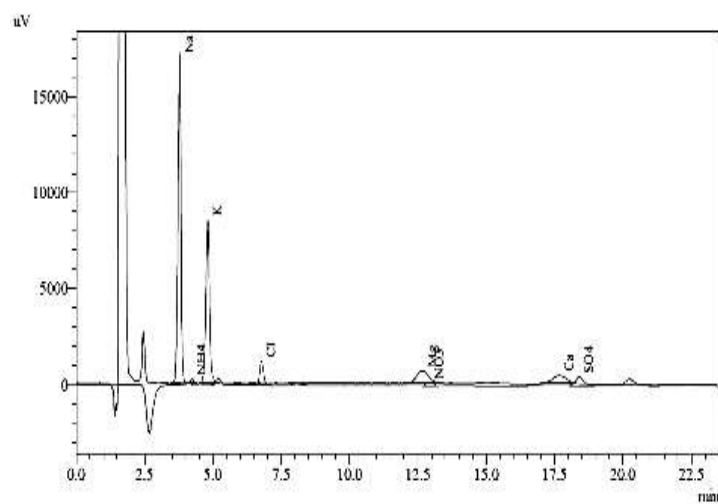
Water-ethanol solvents



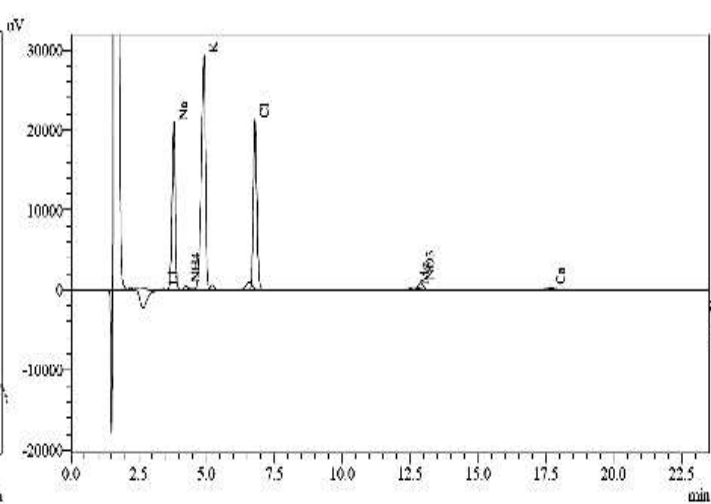
Water solvent



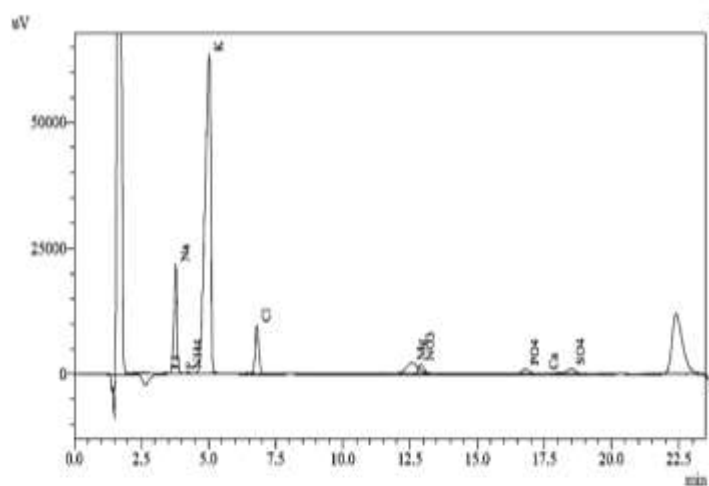
Ethanol solvent



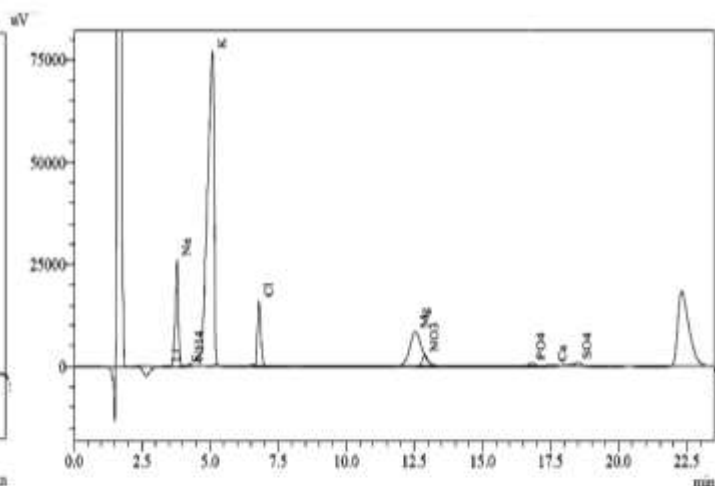
Hexane solvent



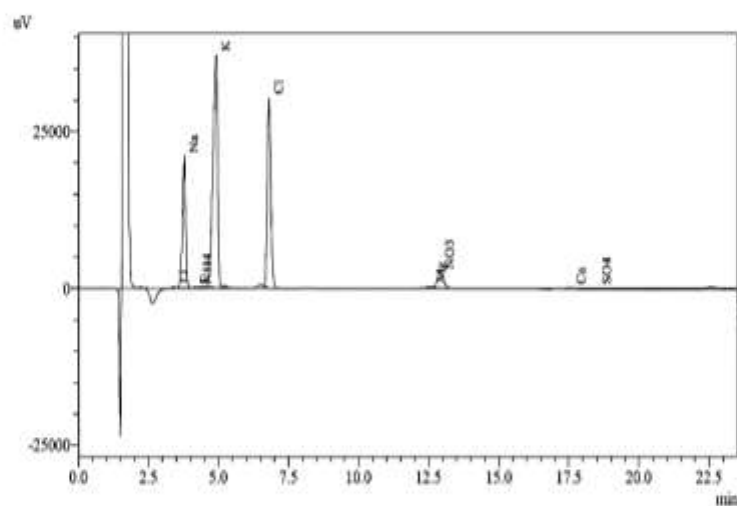
Hexane-ethanol solvents



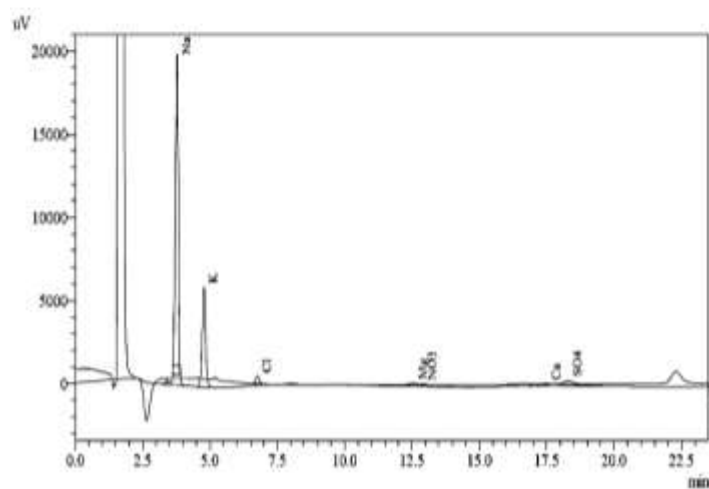
Water-ethanol solvents



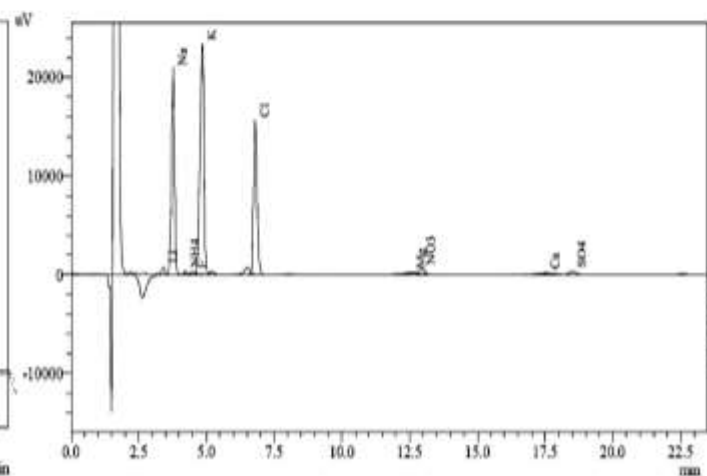
Water solvent



Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

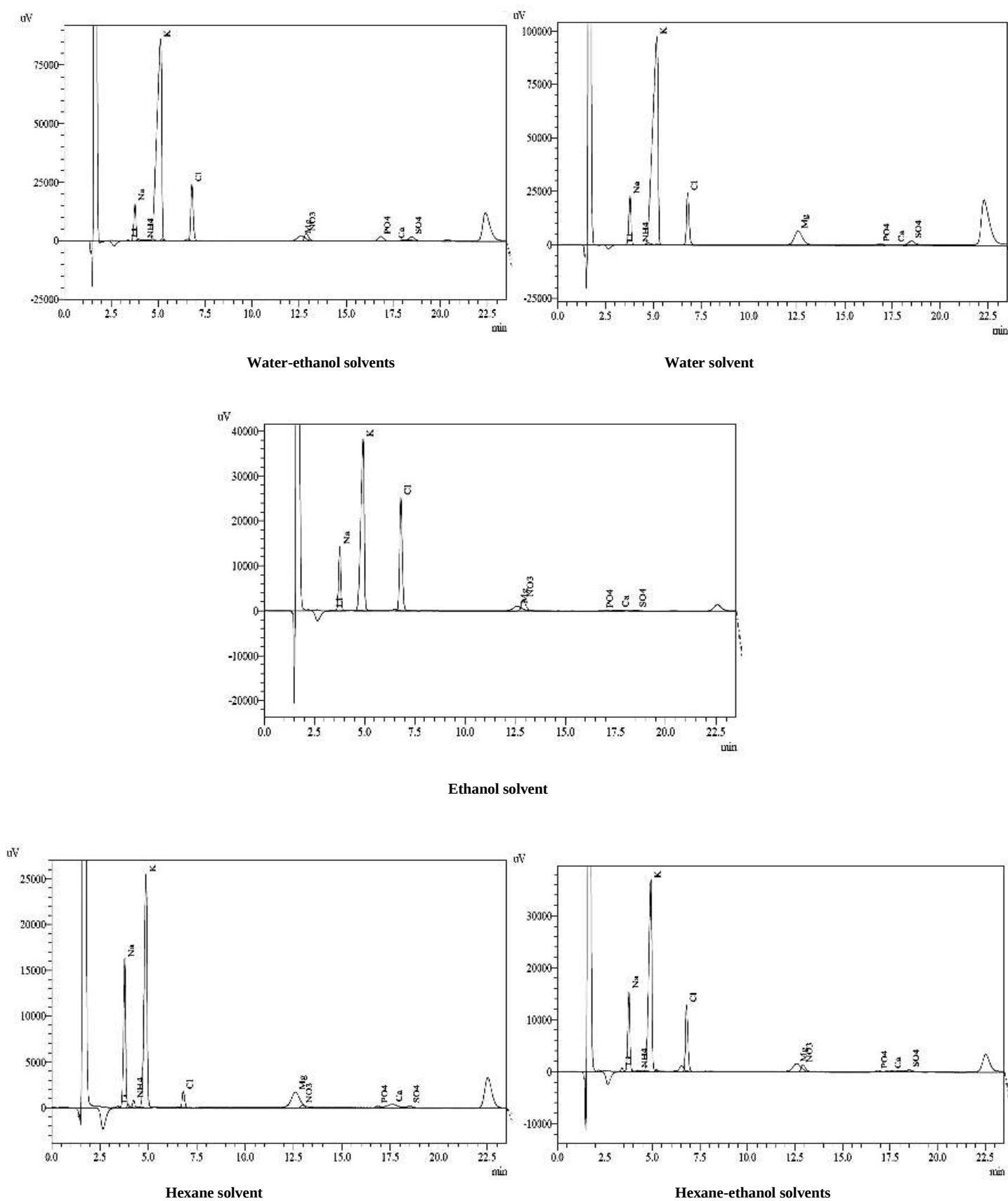
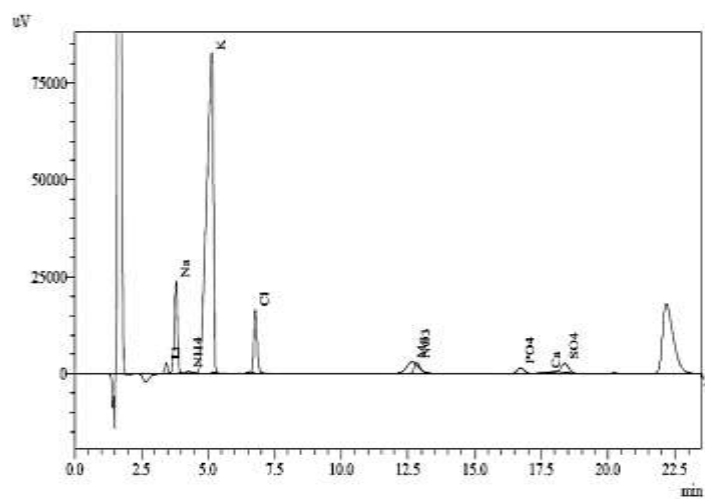
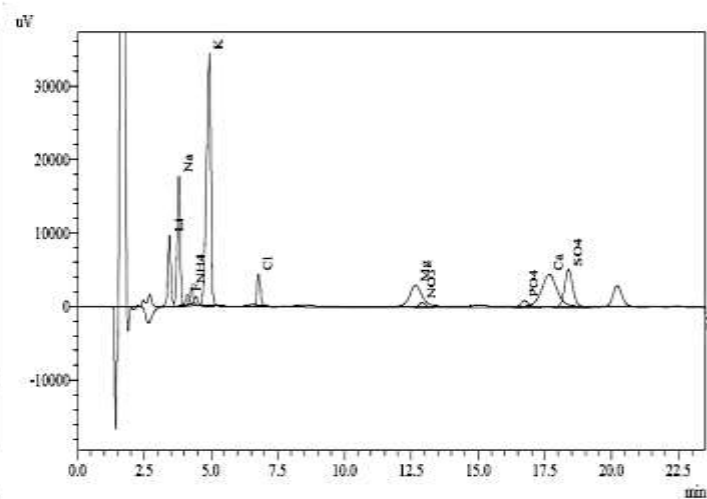


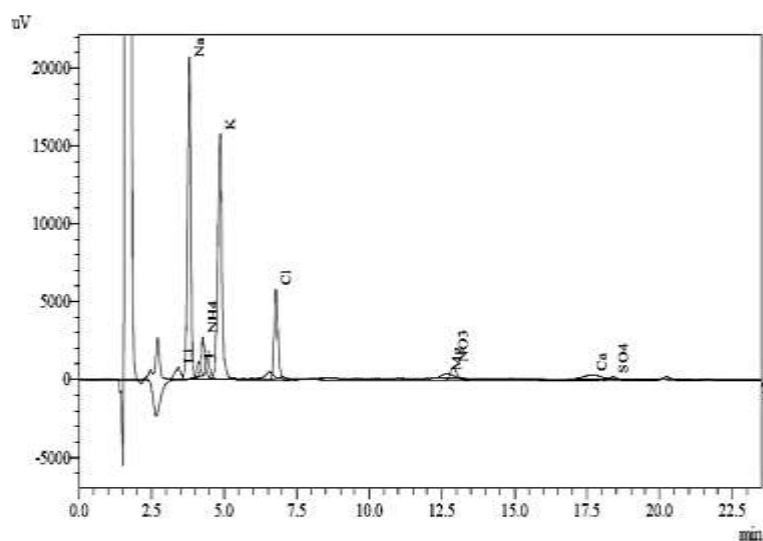
Figure 0.5. Quantitative result of *Portulaca oleracea* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



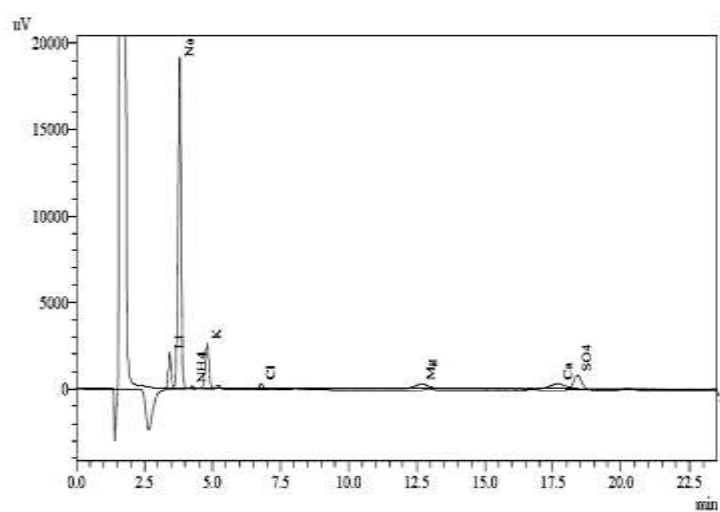
Water-ethanol solvents



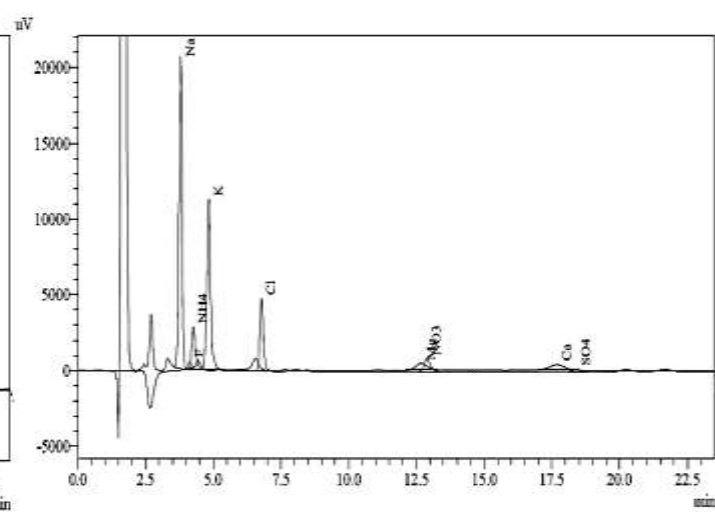
Water solvent



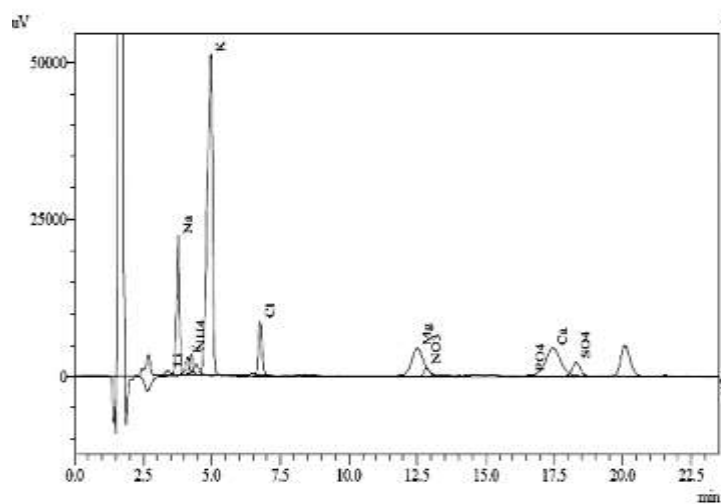
Ethanol solvent



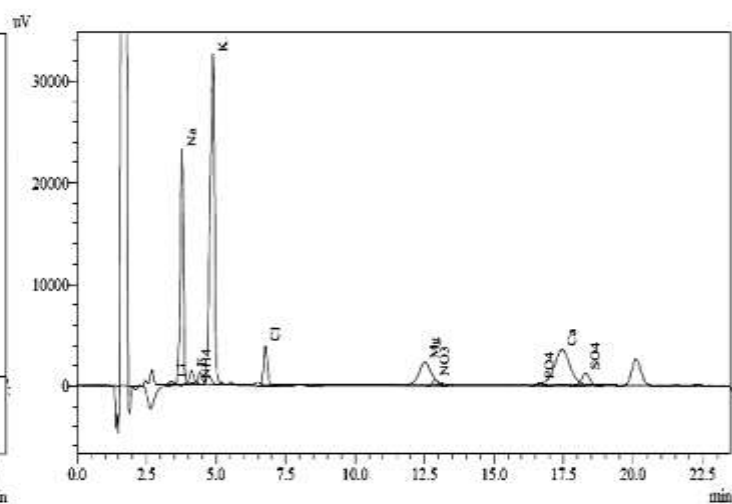
Hexane solvent



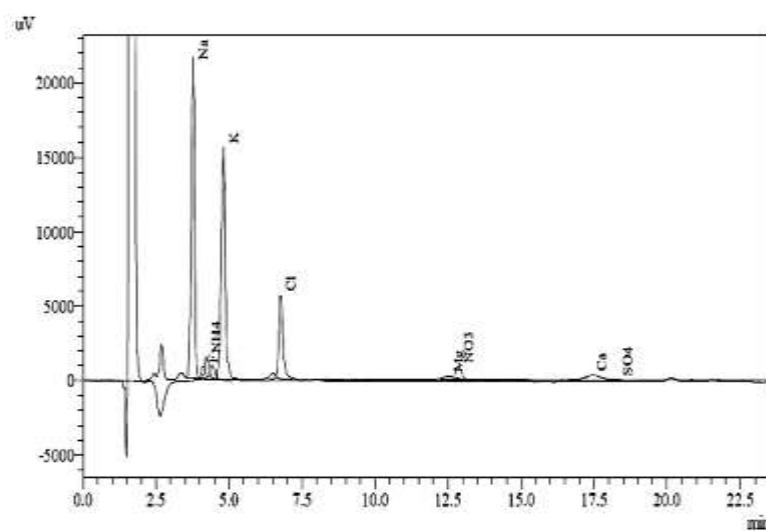
Hexane-ethanol solvents



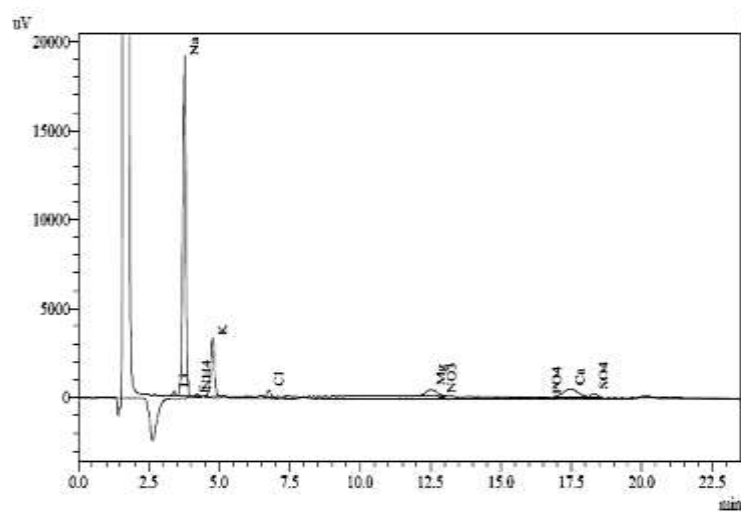
Water-ethanol solvents



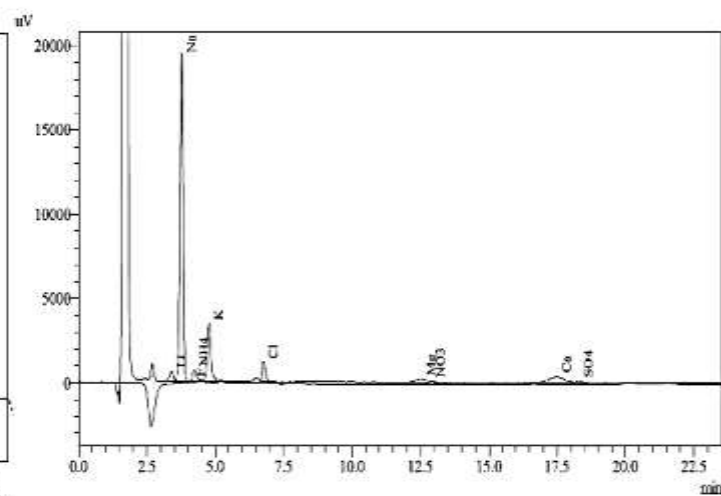
Water solvent



Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

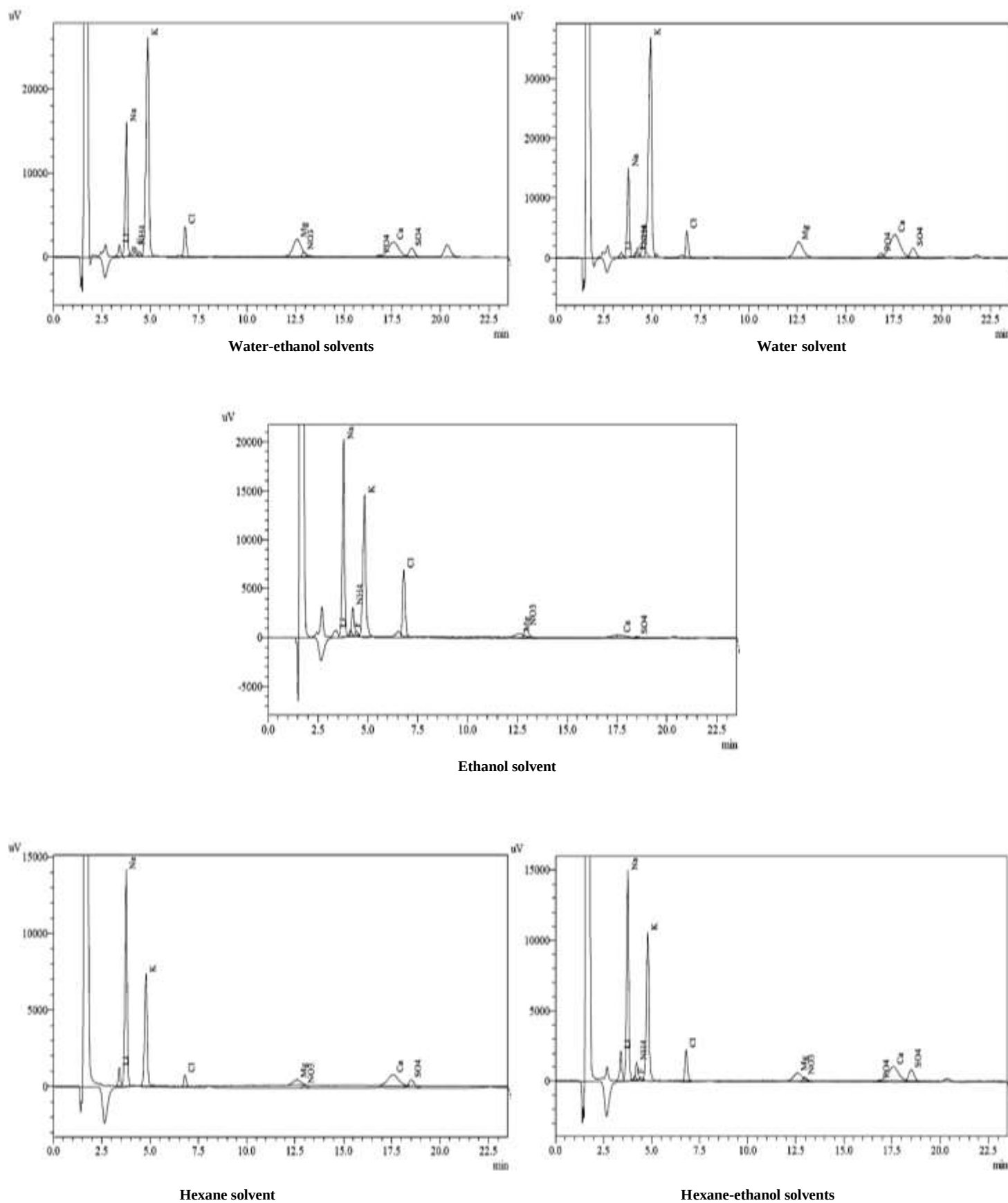
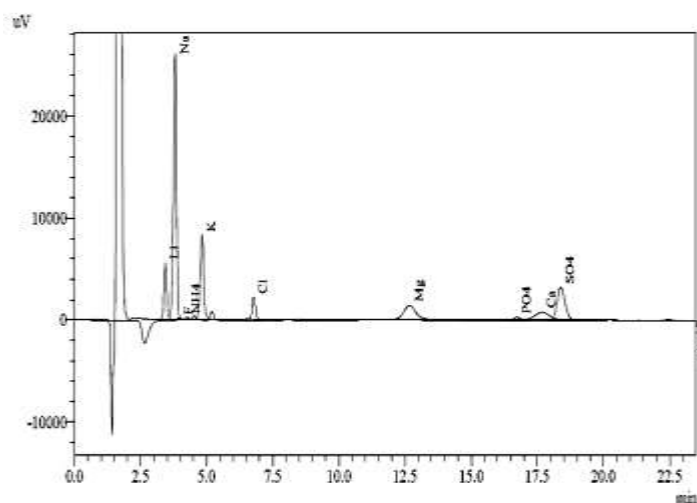
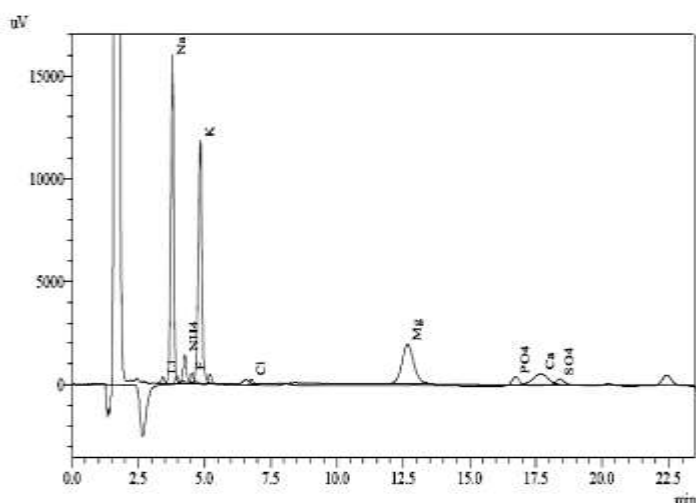


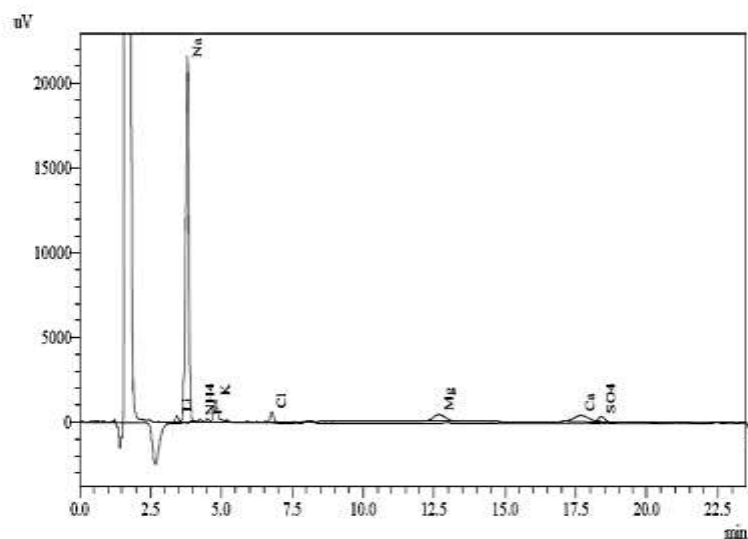
Figure 0.6. Quantitative result of *Artemisia haussknechtii boiss* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



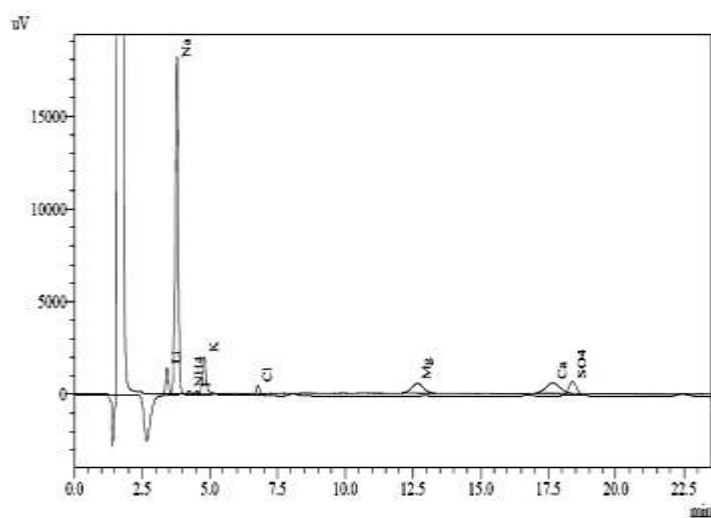
Water-ethanol solvents



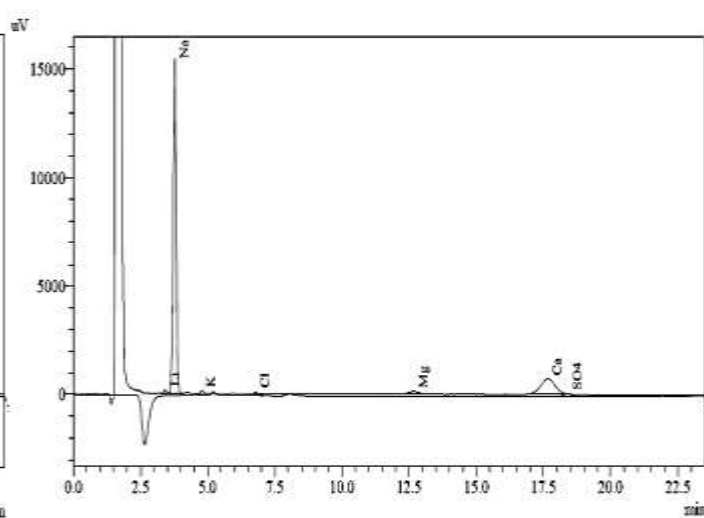
Water solvent



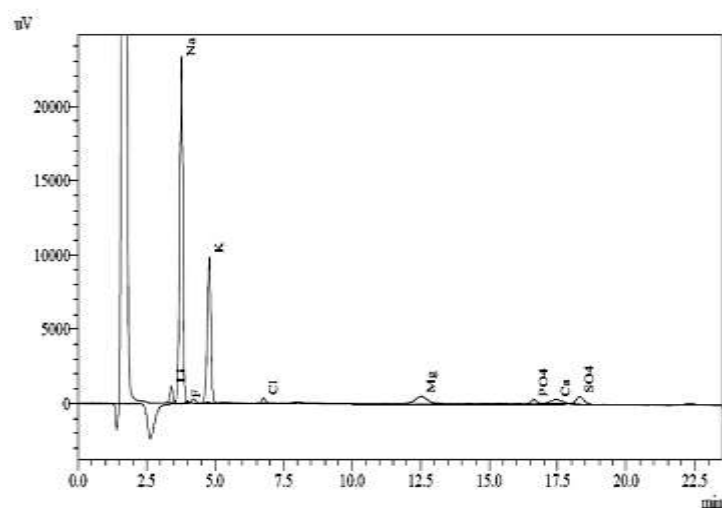
Ethanol solvent



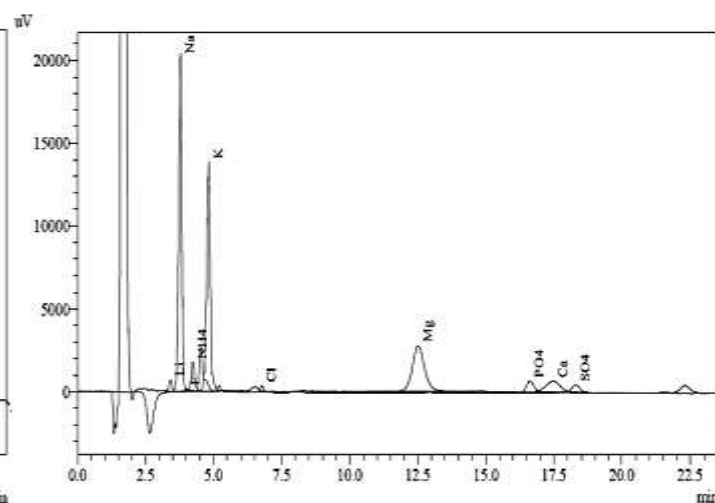
Hexane solvent



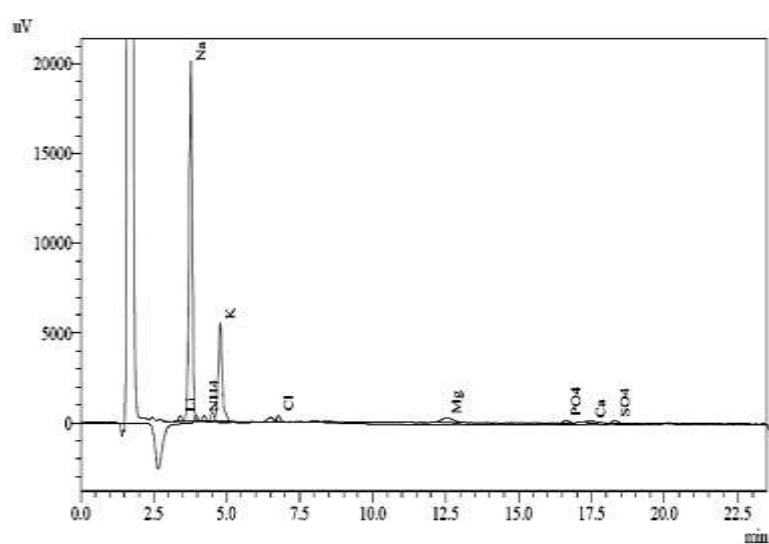
Hexane-ethanol solvents



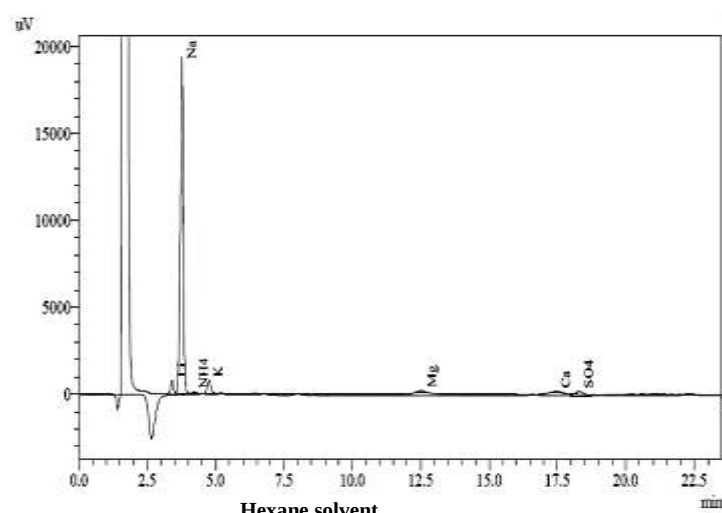
Water-ethanol solvents



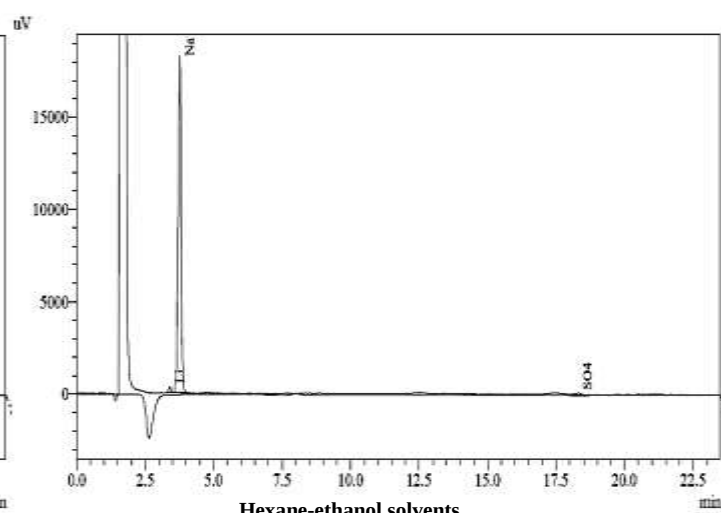
Water solvent



Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

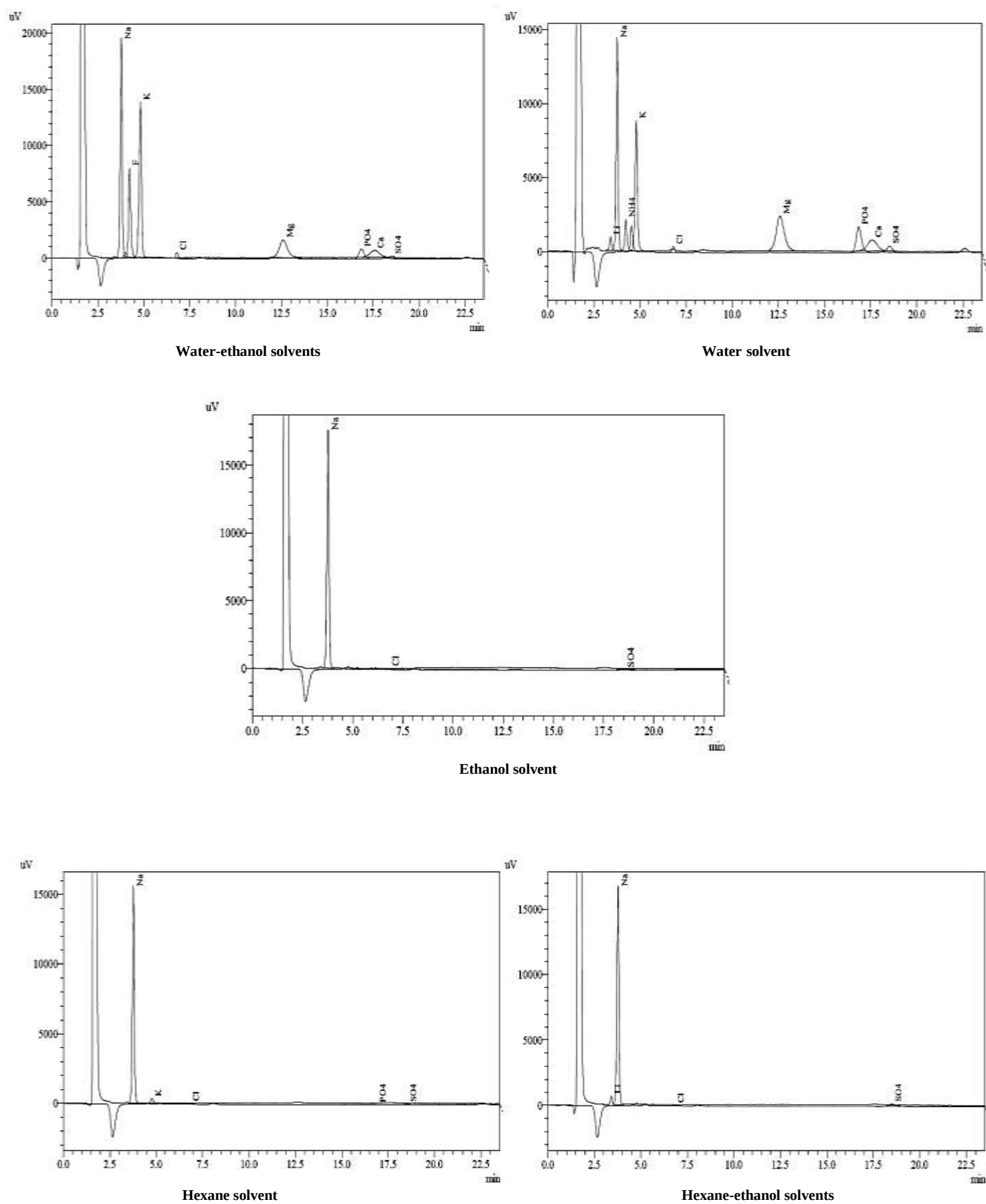
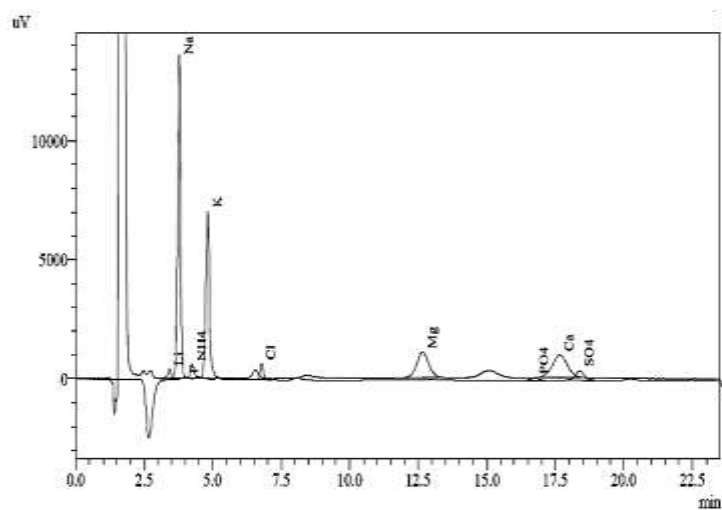
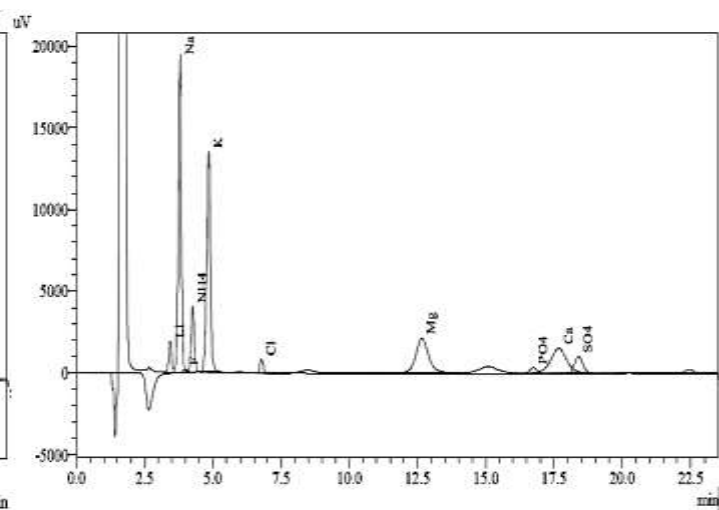


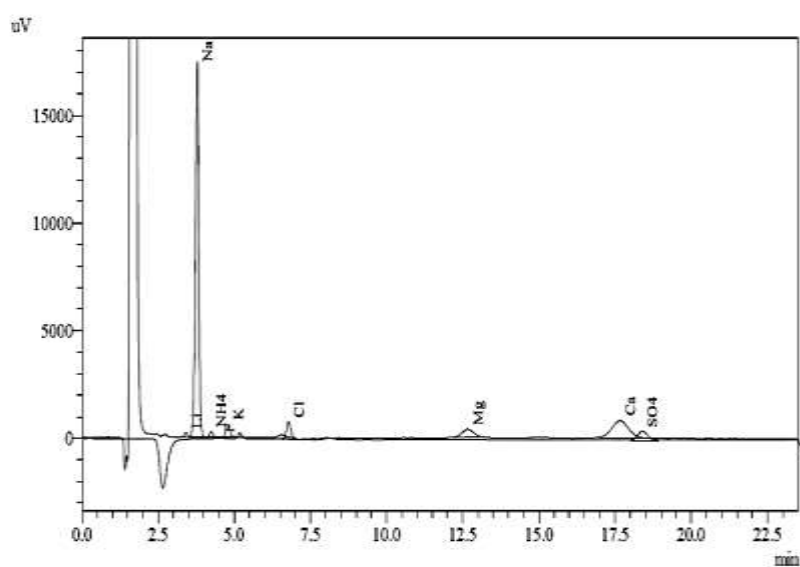
Figure 0.7. Quantitative result of *Prunus microcarpa* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



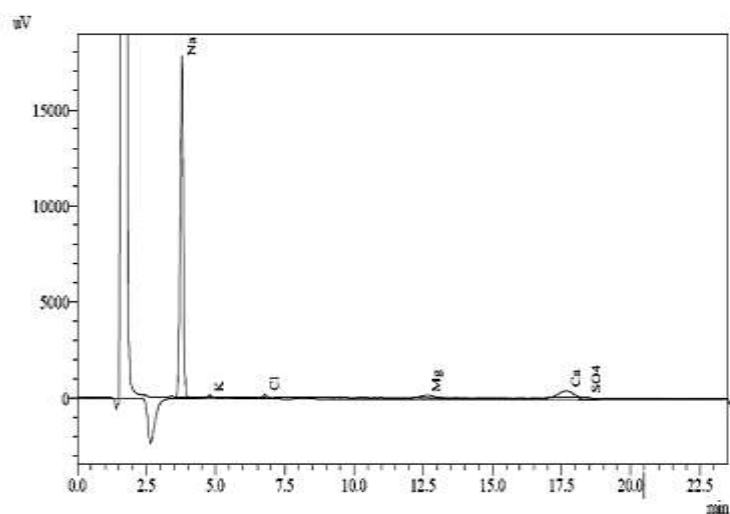
Water-ethanol solvents



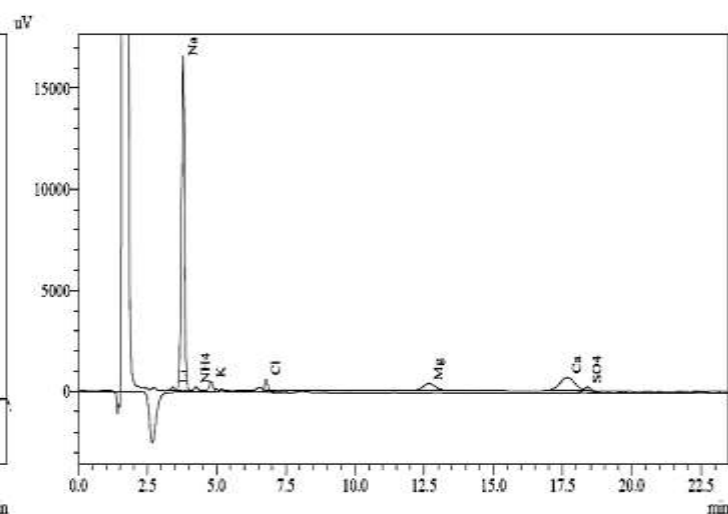
Water solvent



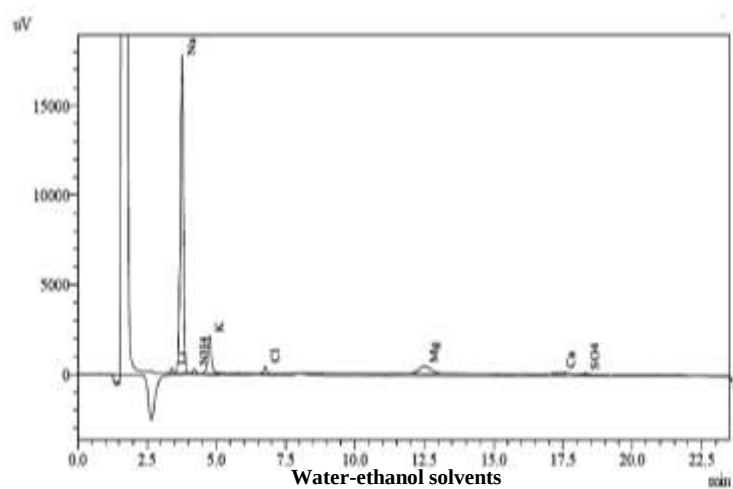
Ethanol solvent



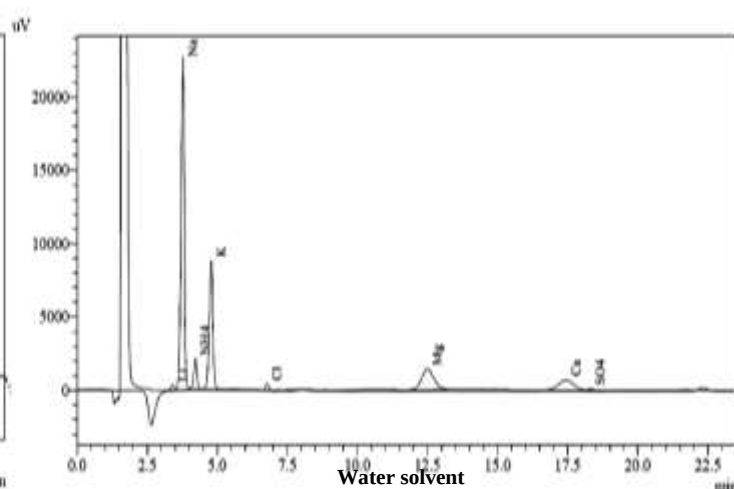
Hexane solvent



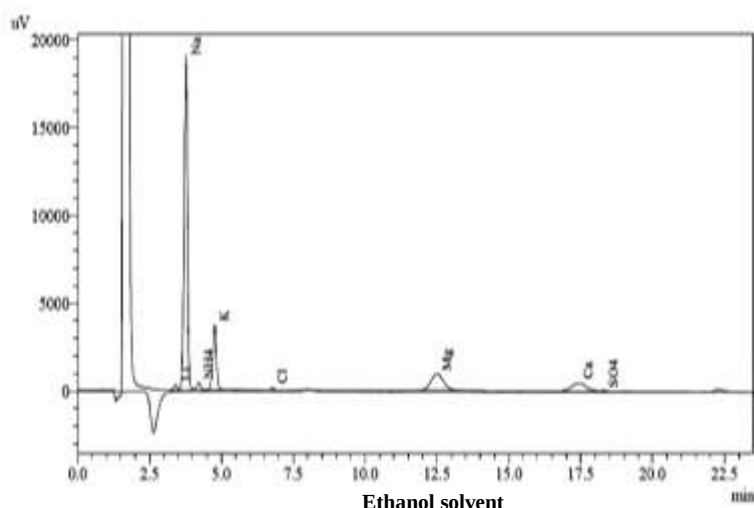
Hexane-ethanol solvents



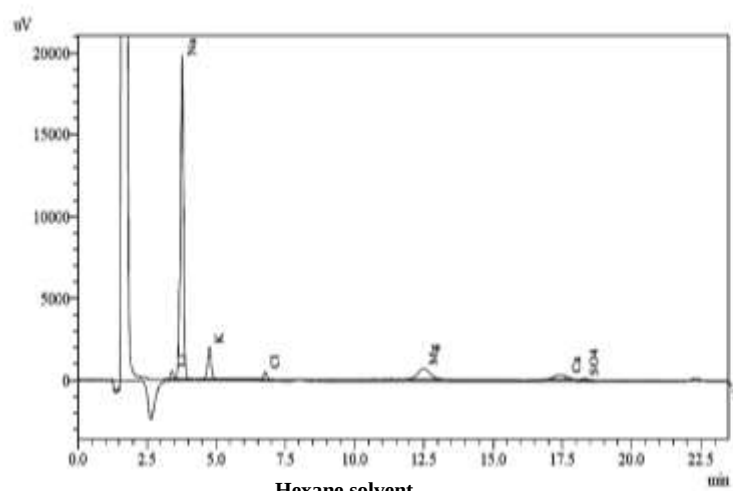
Water-ethanol solvents



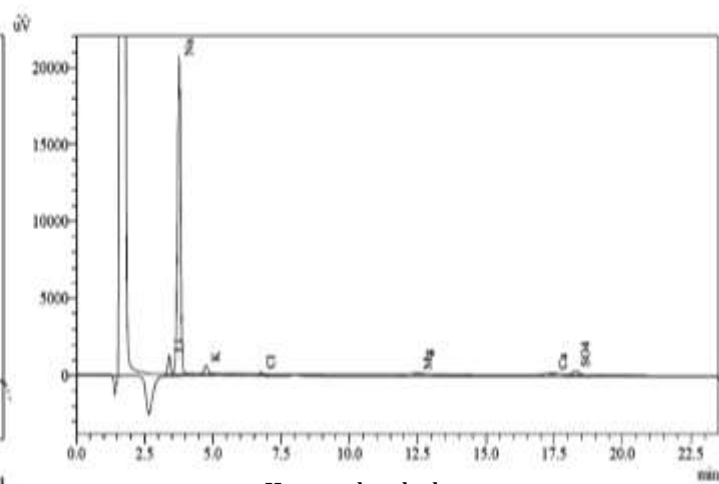
Water solvent



Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

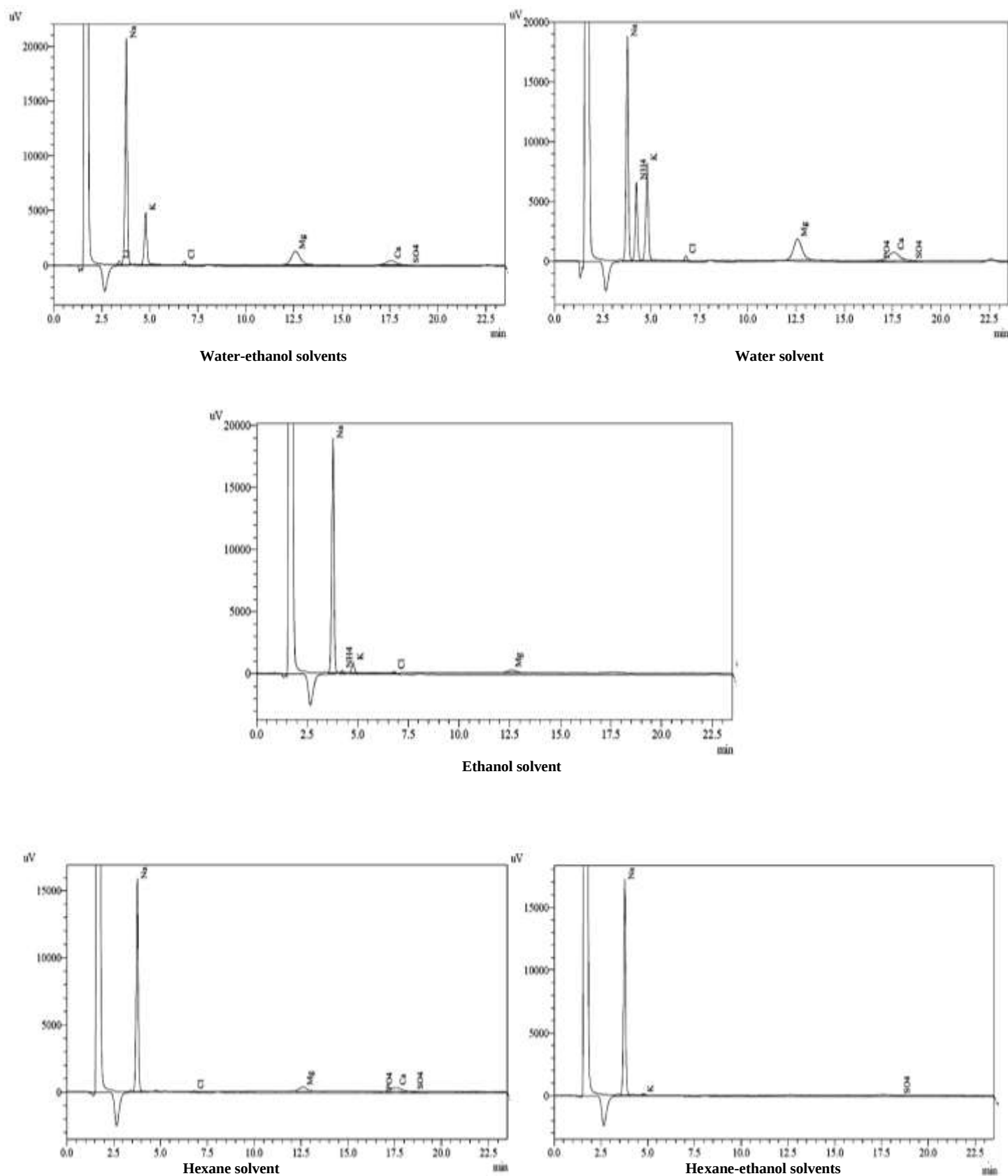
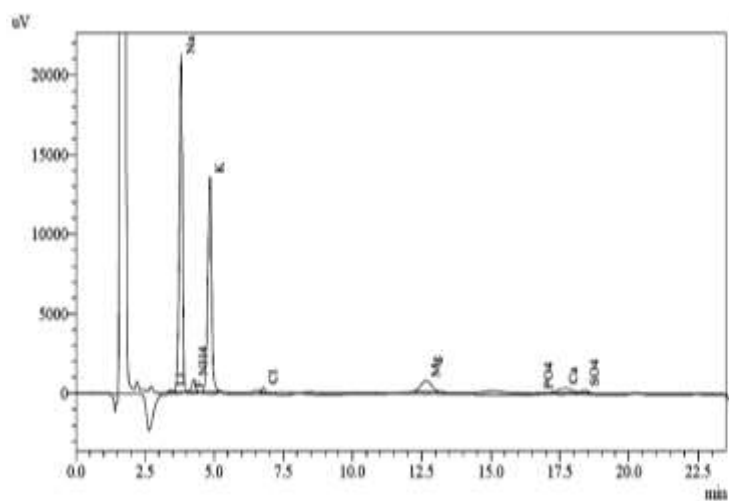
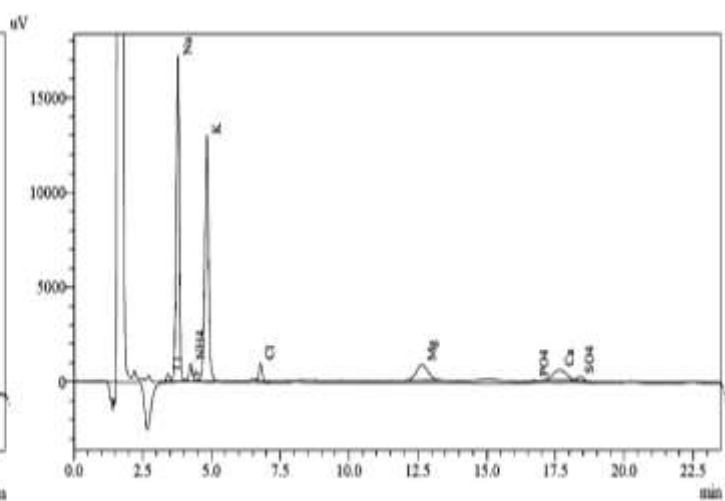


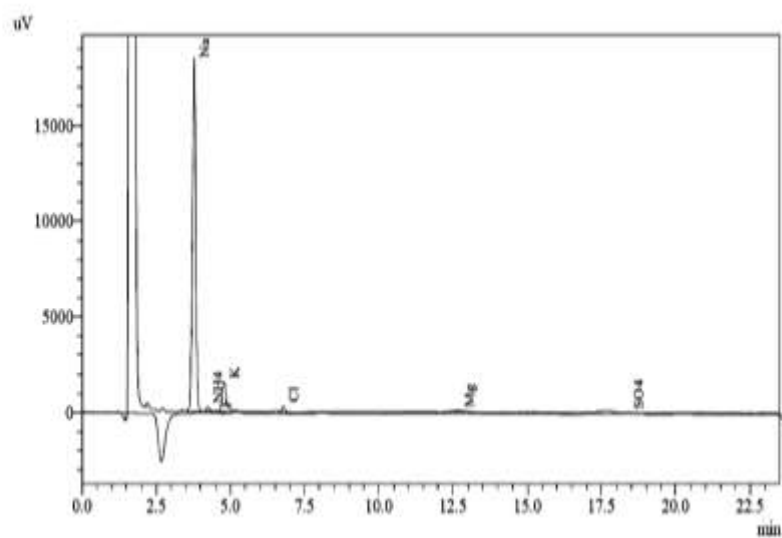
Figure 0.8. Quantitative result of *Silybum Marianuma* Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



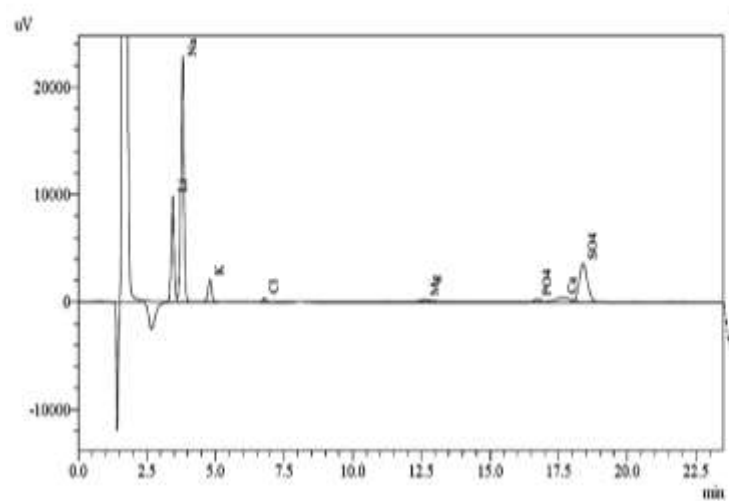
Water-ethanol solvents



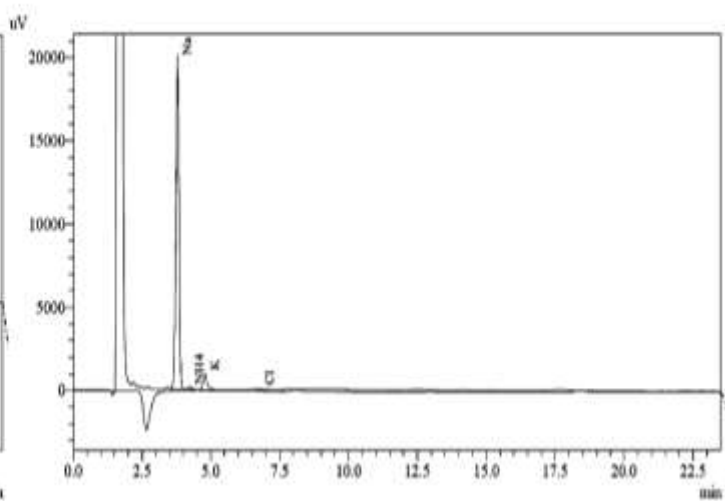
Water solvent



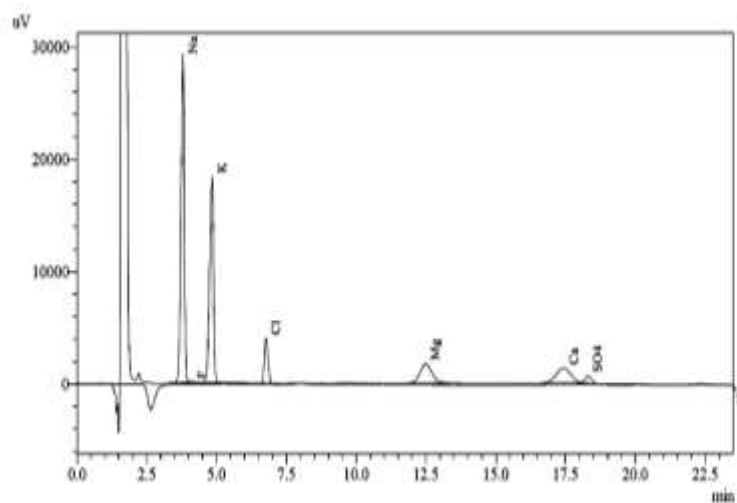
Ethanol solvent



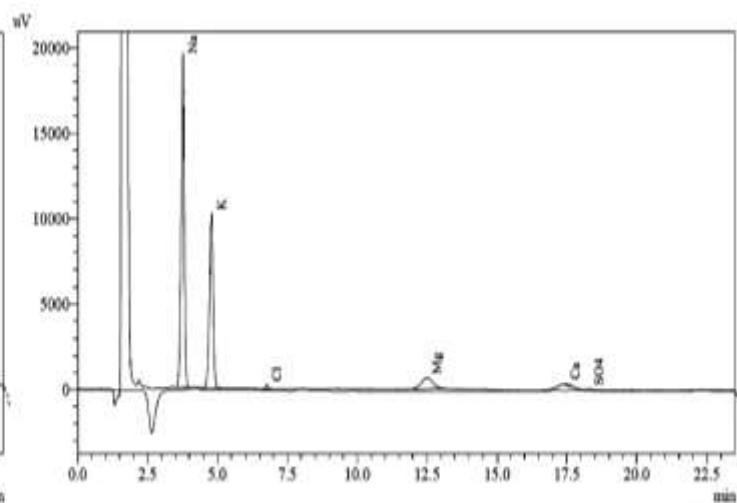
Hexane solvent



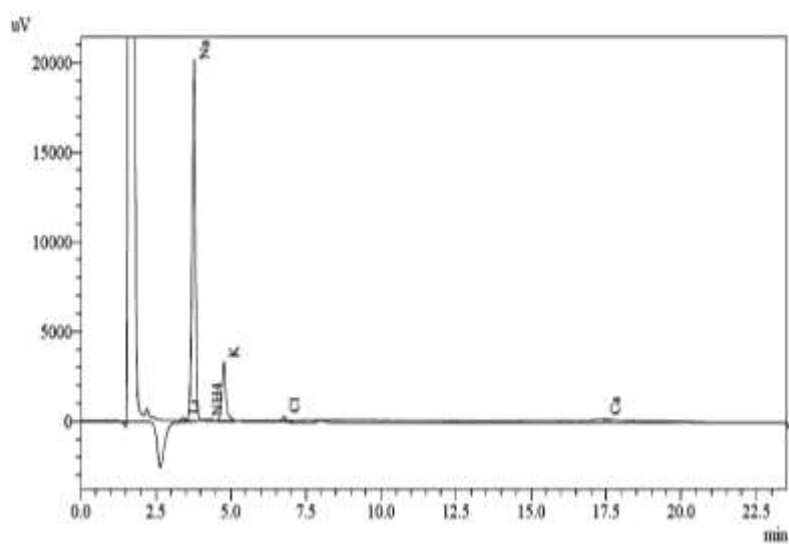
Hexane-ethanol solvents



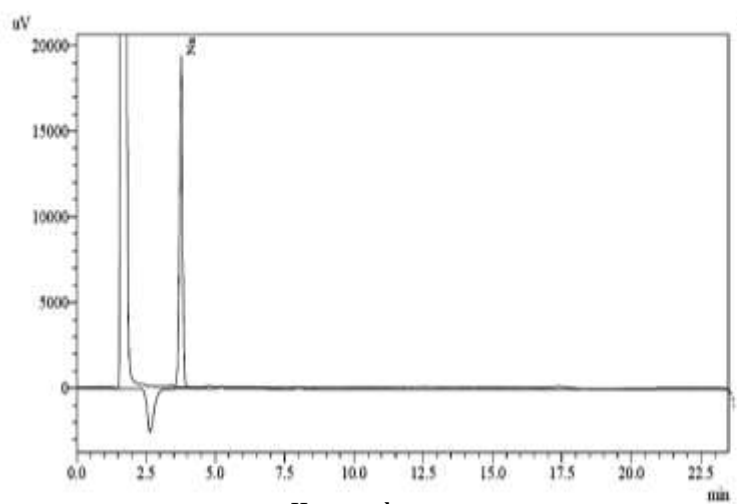
Water-ethanol solvents



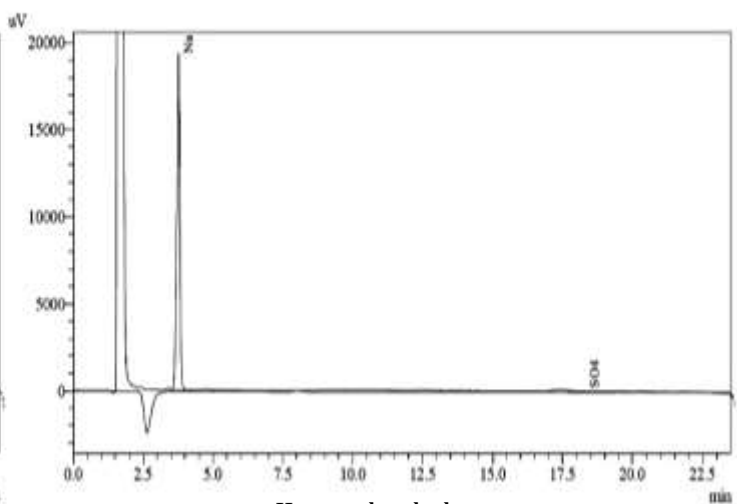
Water solvent



Ethanol solvent



Hexane solvent



Hexane-ethanol solvents

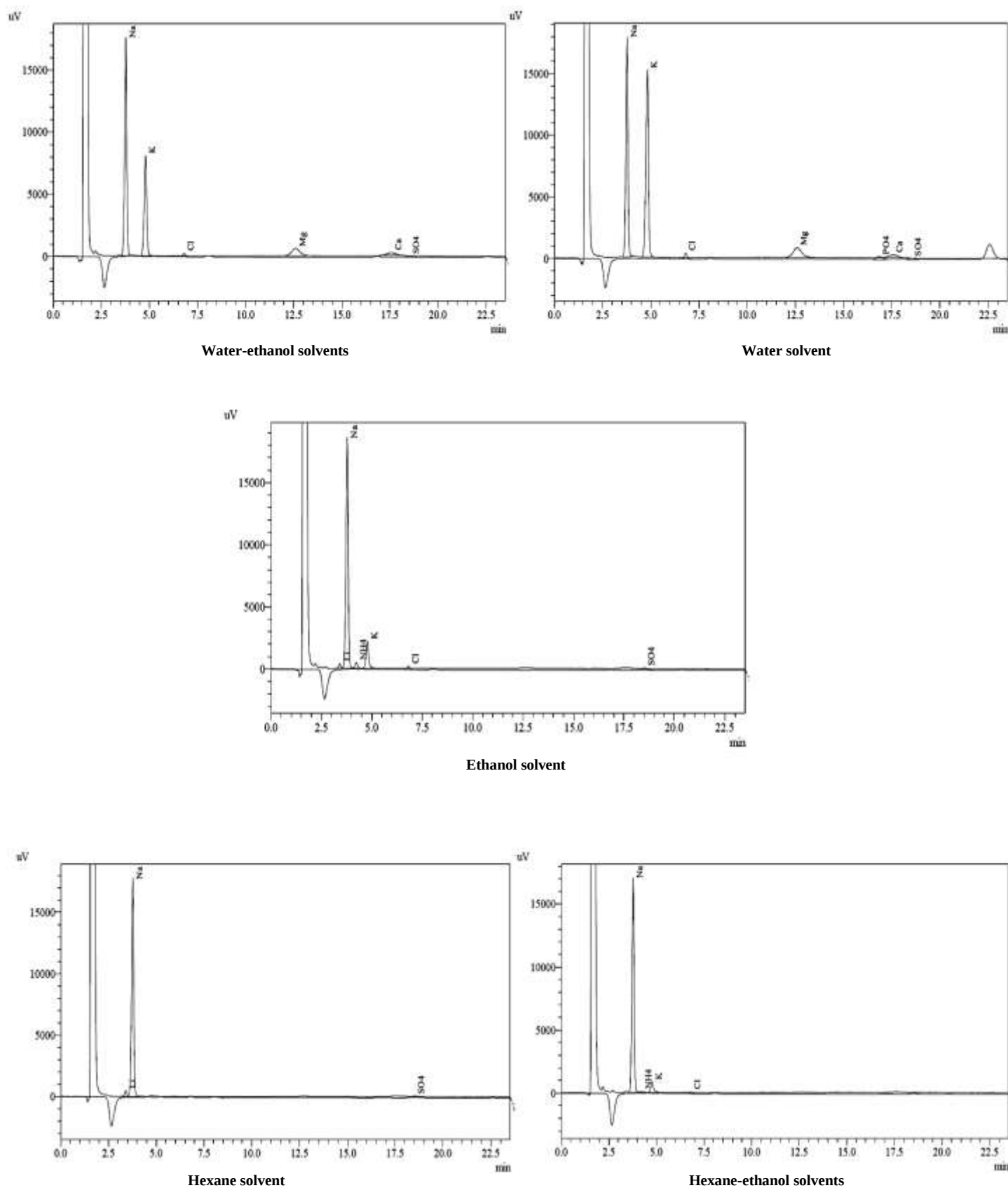
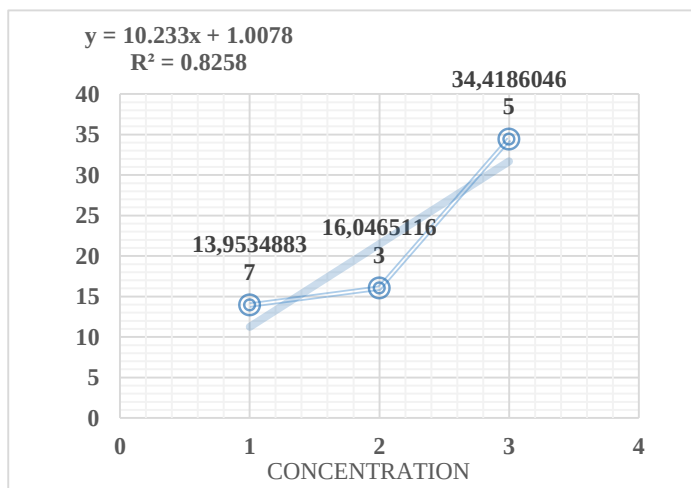
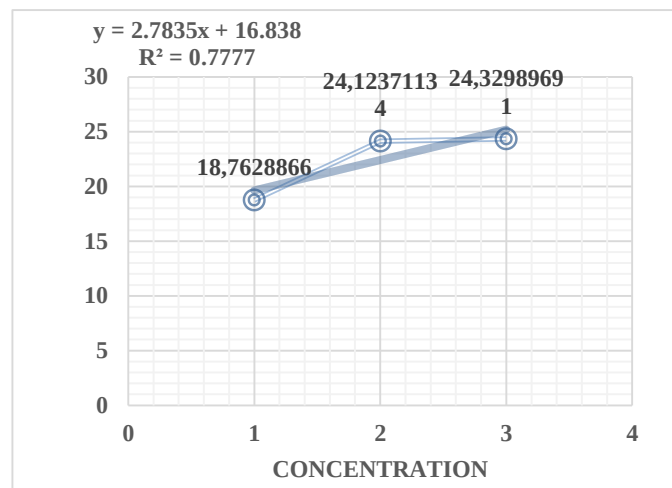


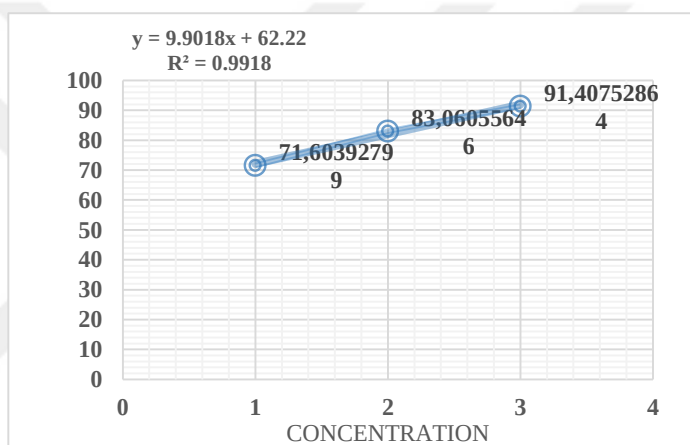
Figure 0.9. Quantitative result of *Pistacia eurycarpa* Yale Extract in five solvents during three methods of (Conventional, Microwave and Orbital shaker) extraction by ion chromatography



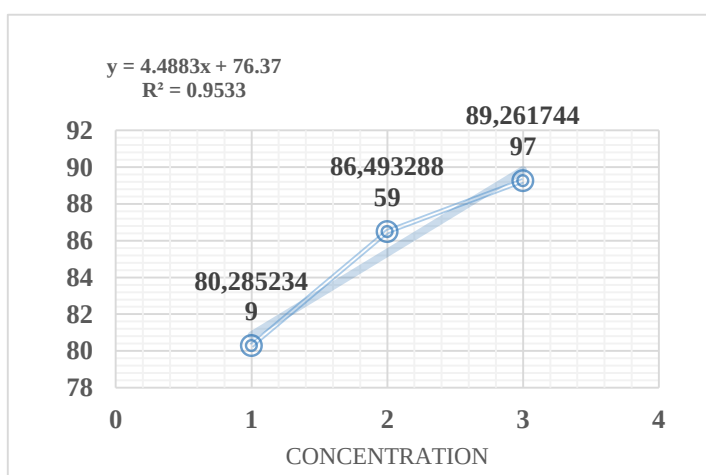
Water extract



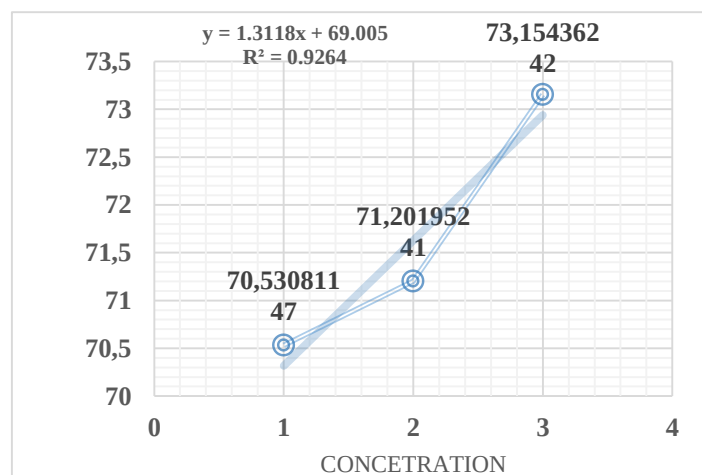
Water: ethanol extract



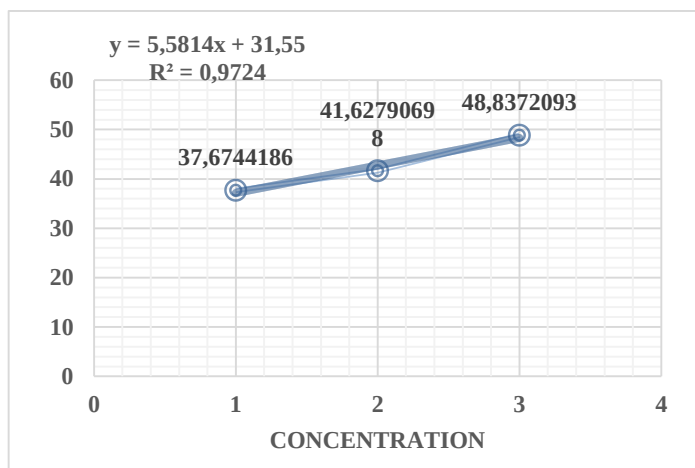
Ethano extract



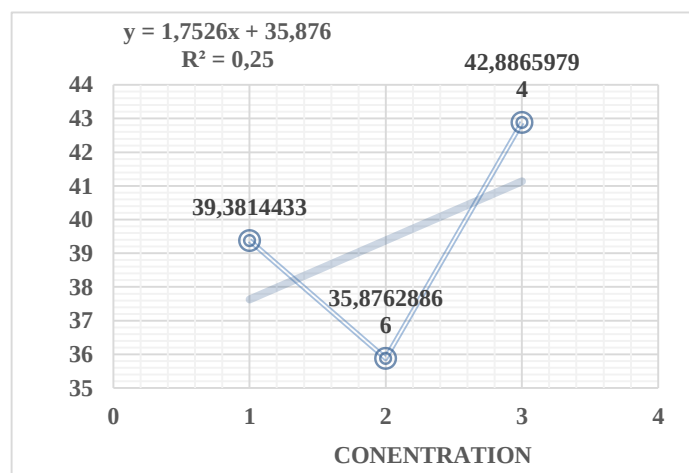
Hexane: ethanol extract



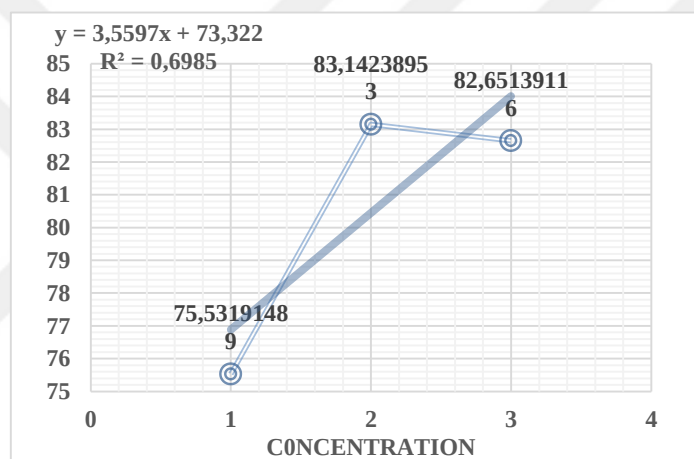
Hexane extract



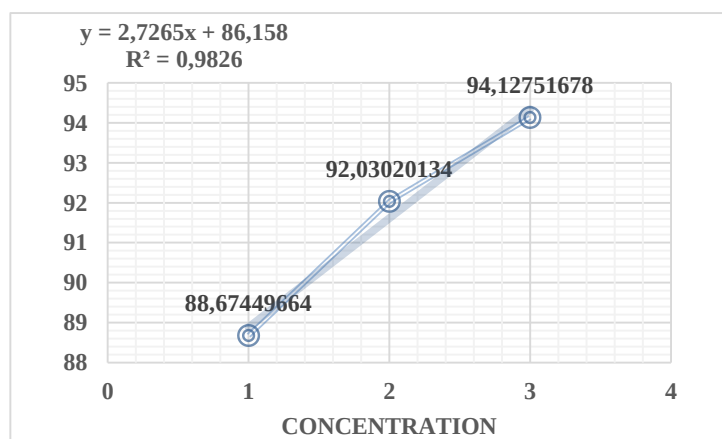
Water extract



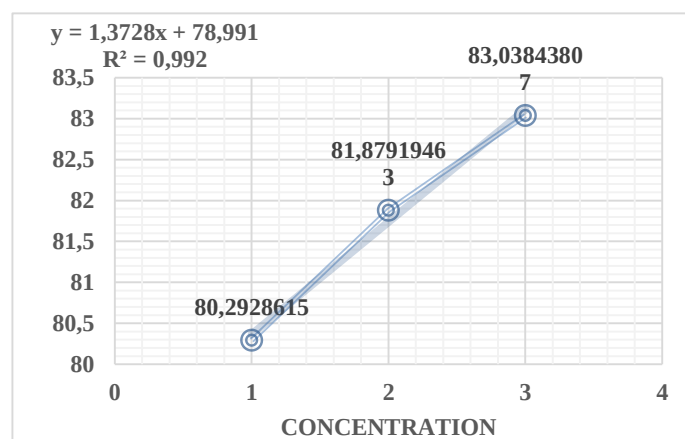
water: ethanol extract



Ethanol extract



Hexan: ethanol extract



Hexane extract

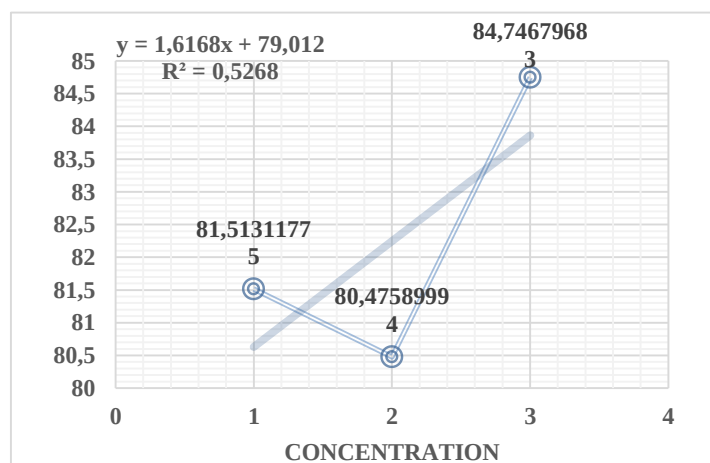
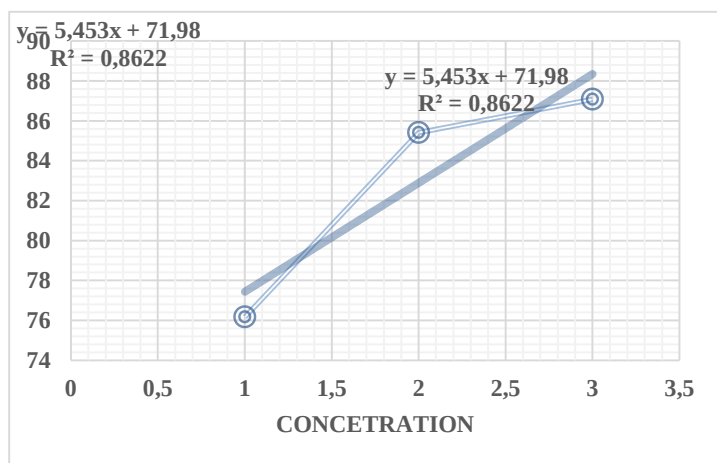
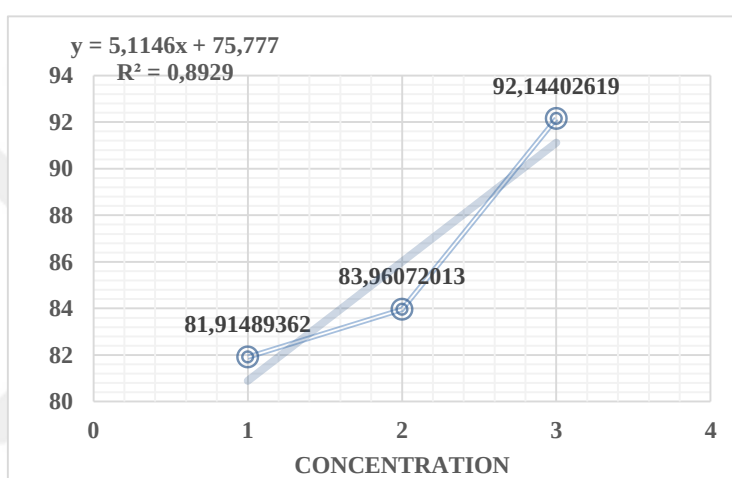
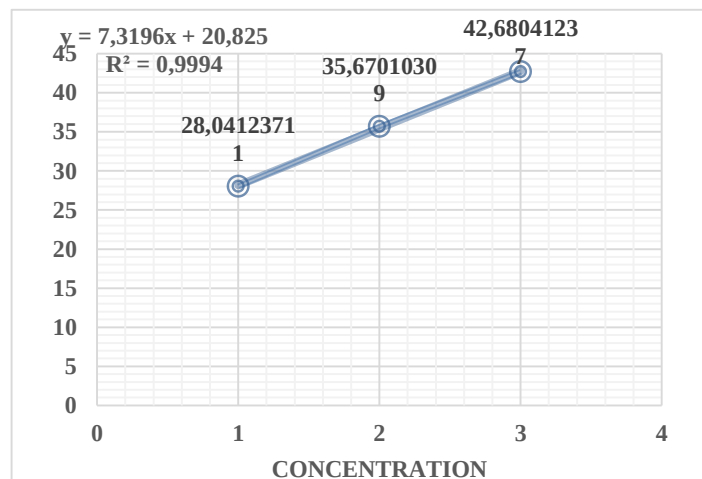
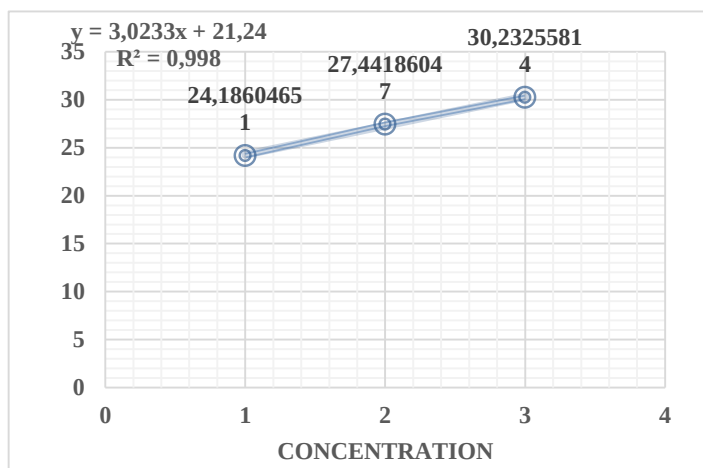
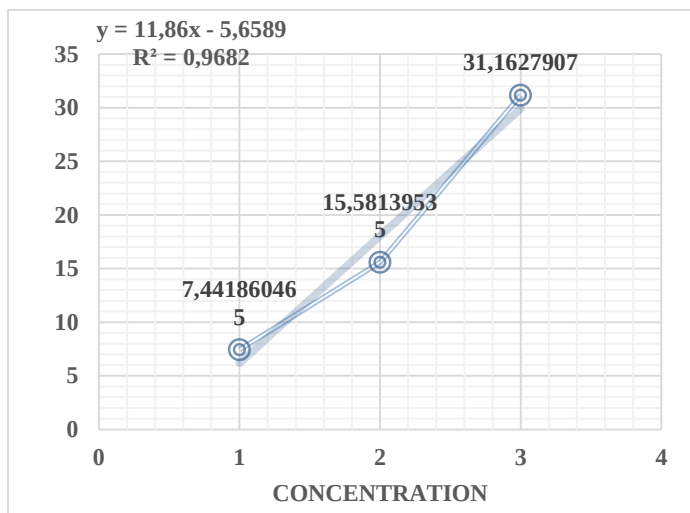
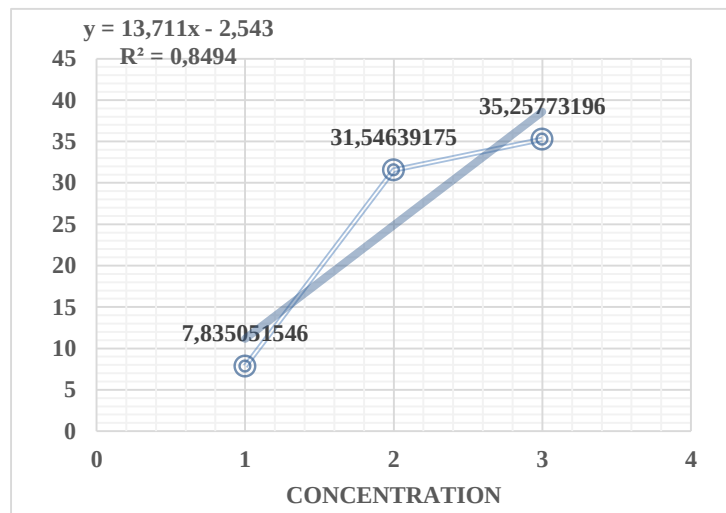


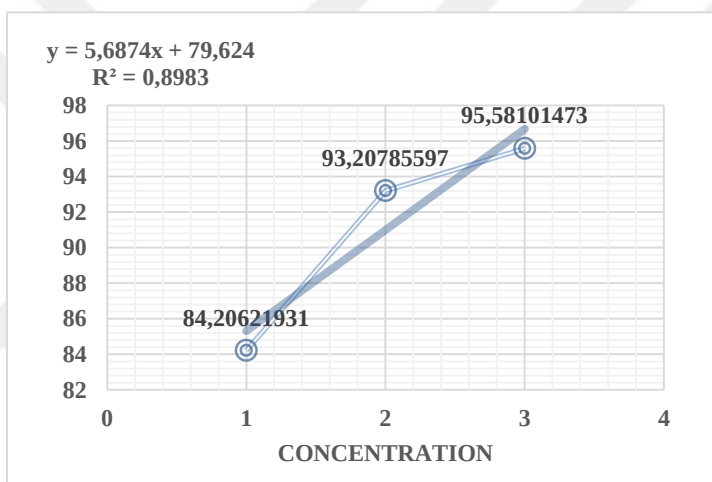
Figure 0.10. DPPH free radical scavenging activity of *Eremurus Spectabilis Bieb* extracts by methods of (Conventional, Microwave, and orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).



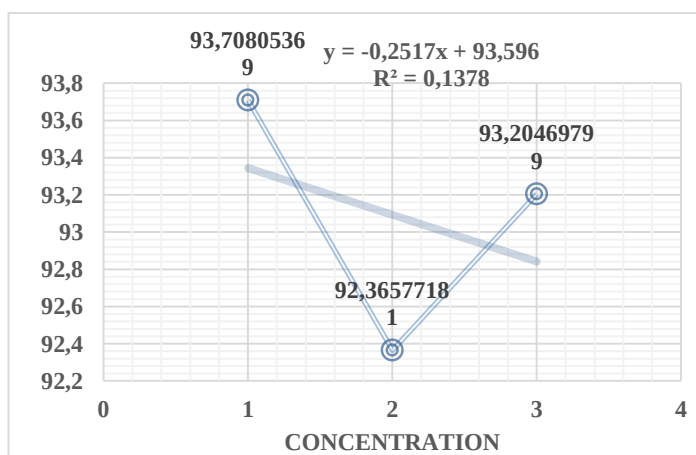
Water extract



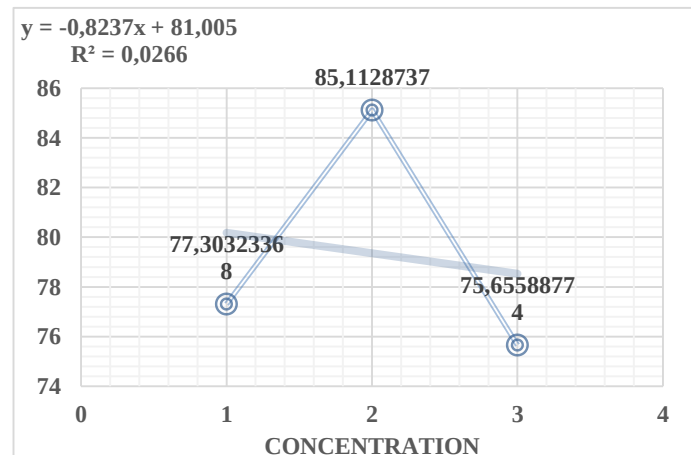
Water: Ethanol extract



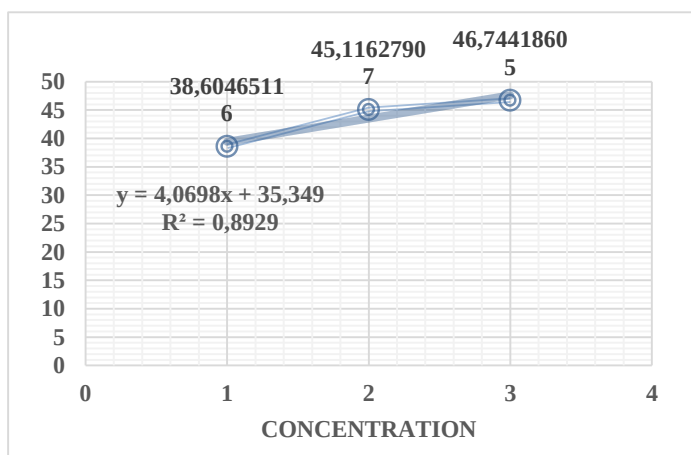
Ethanol extract



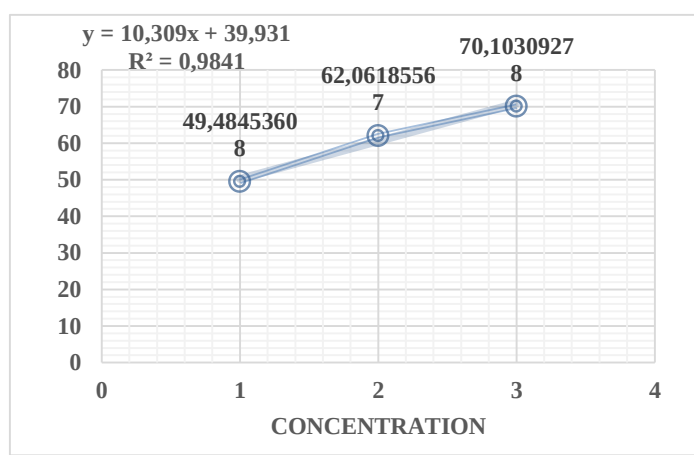
Hexane: ethanol extract



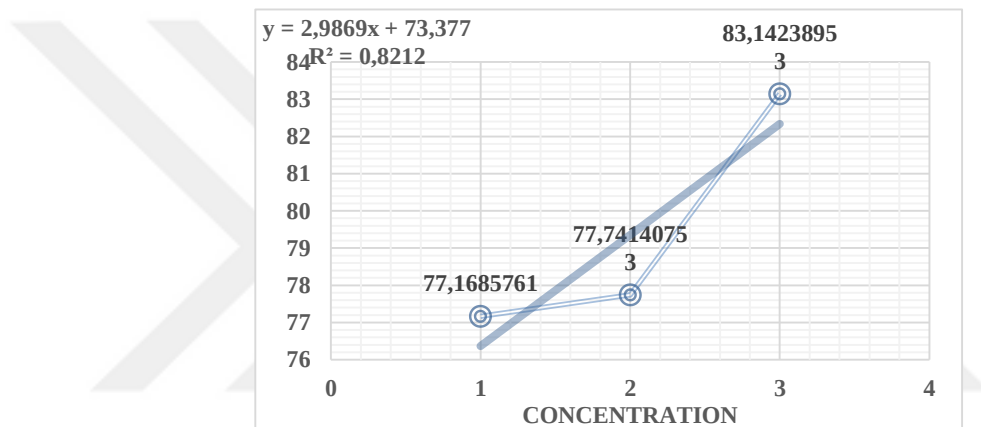
Hexane extract



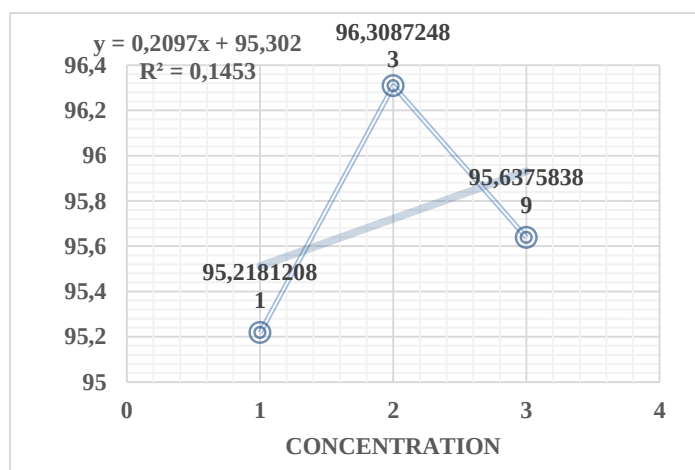
Water extract



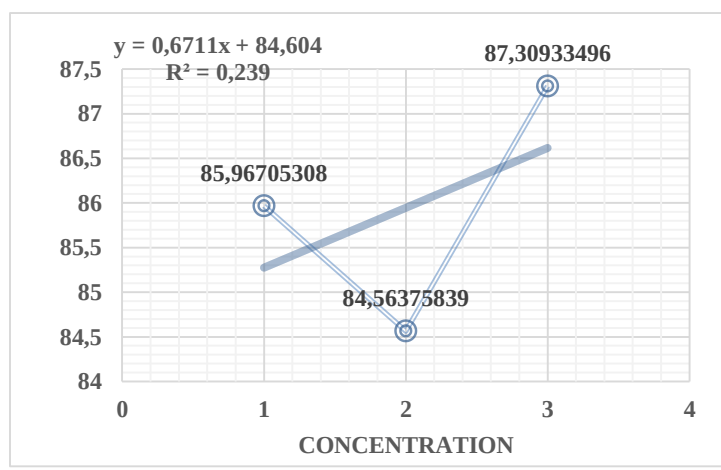
Water ethanol extract



Ethanol extract



Hexane: ethanol extract



Hexane extract

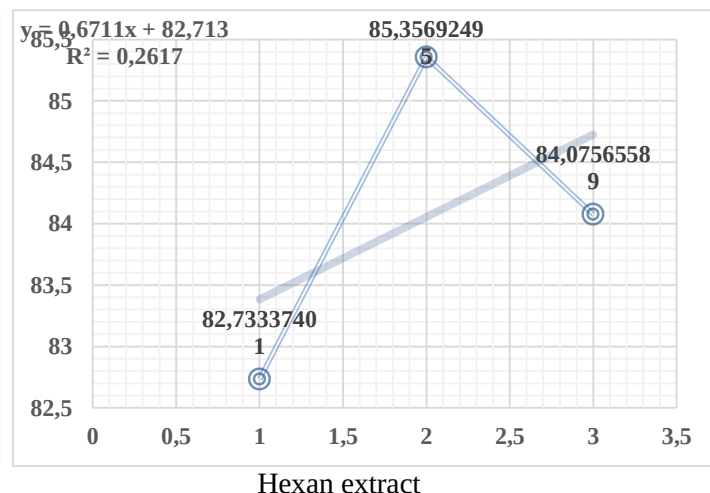
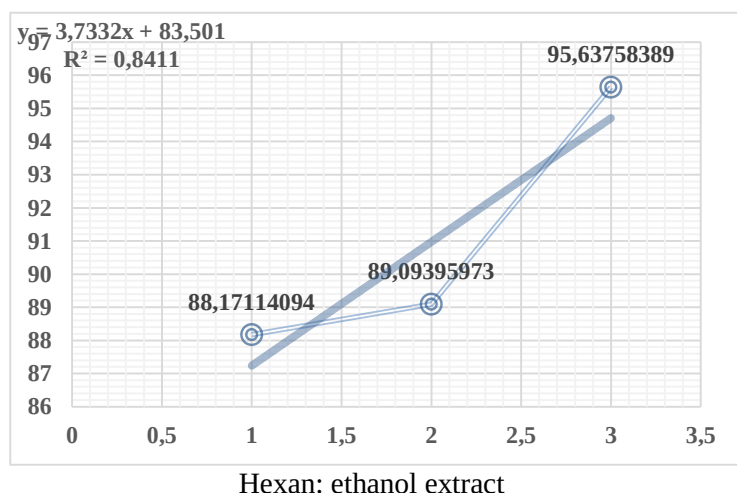
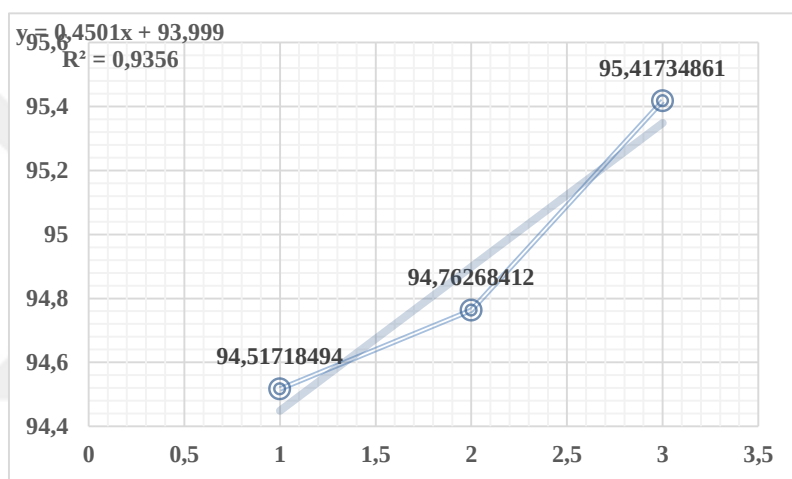
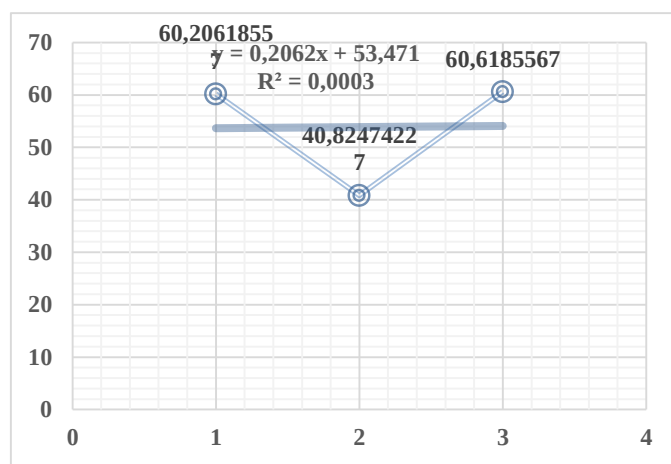
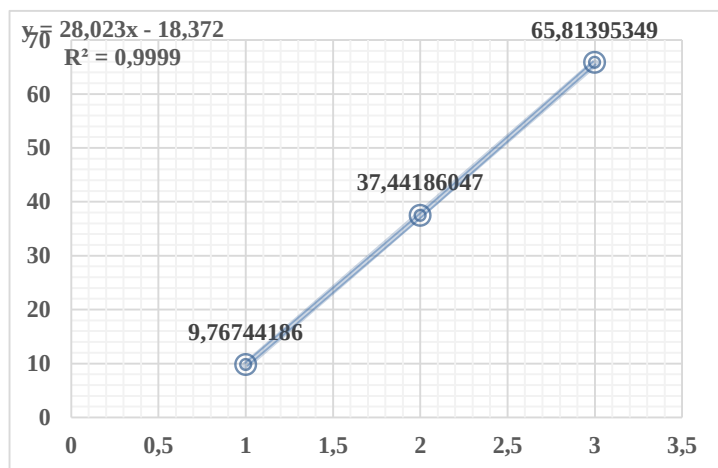
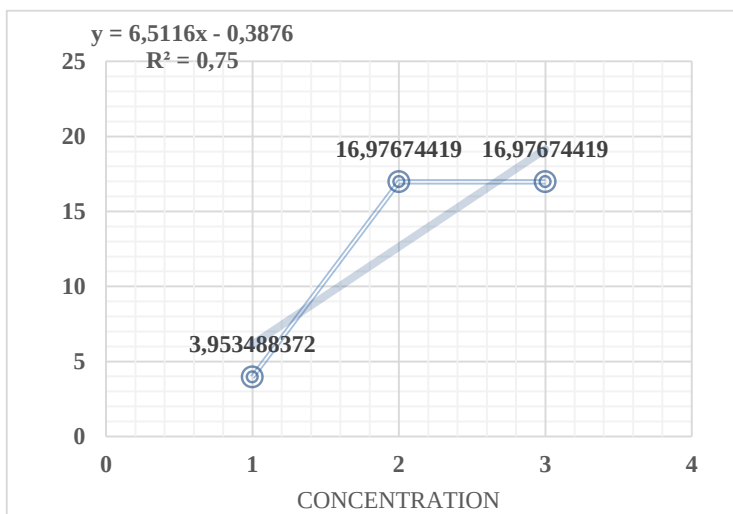
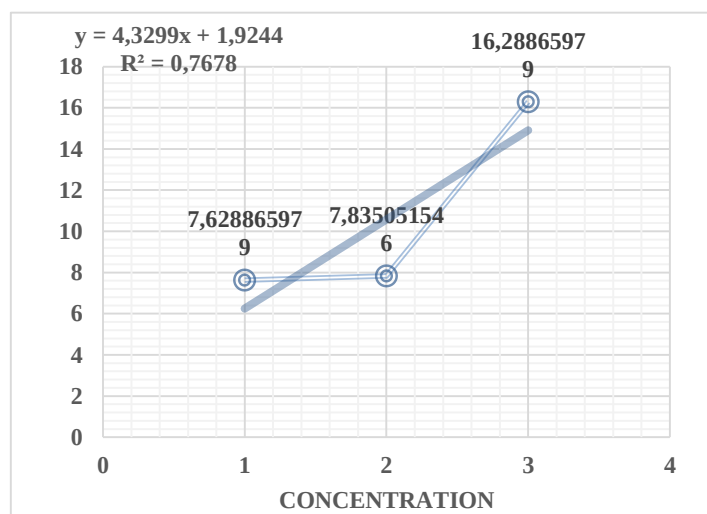


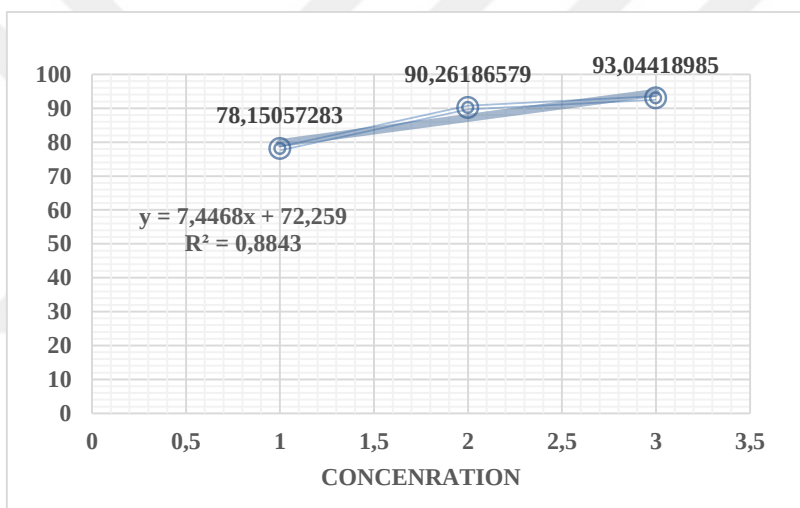
Figure 0.11. DPPH free radical scavenging activity of *Prosopis frakta* extracts, by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).



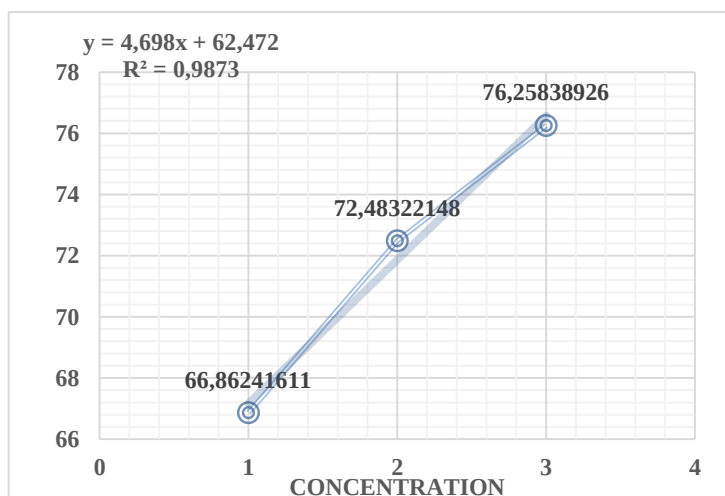
Water extract



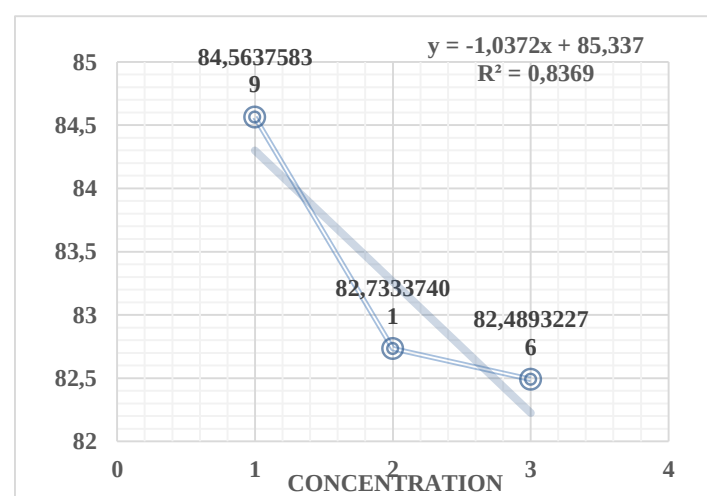
Water: ethanol extract



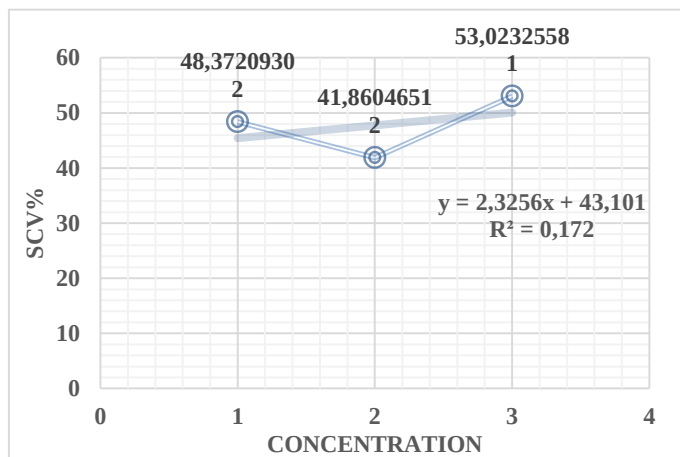
Ethanol extract



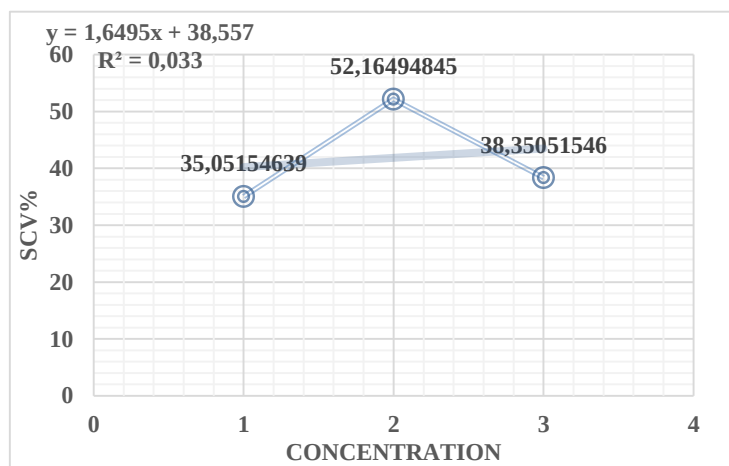
Hexan: ethanol extract



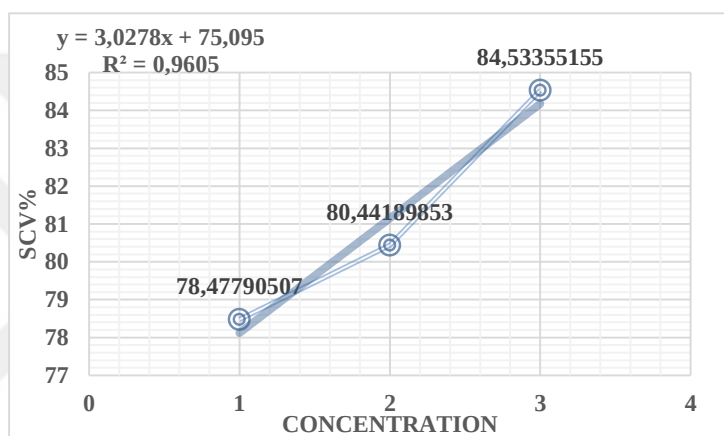
Hexane extract



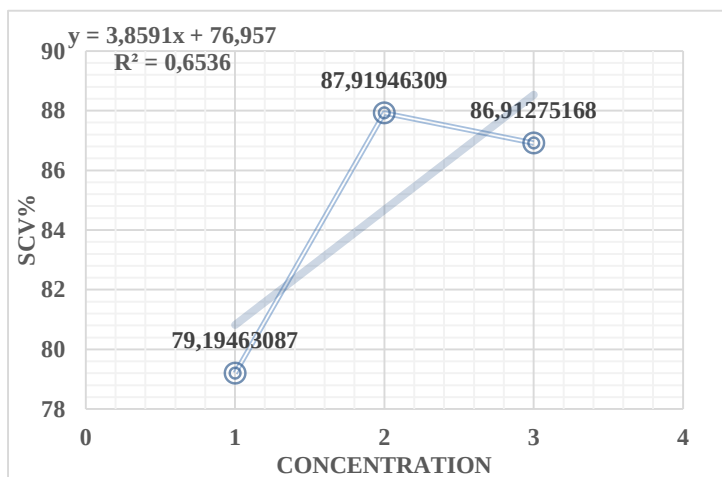
Water extract



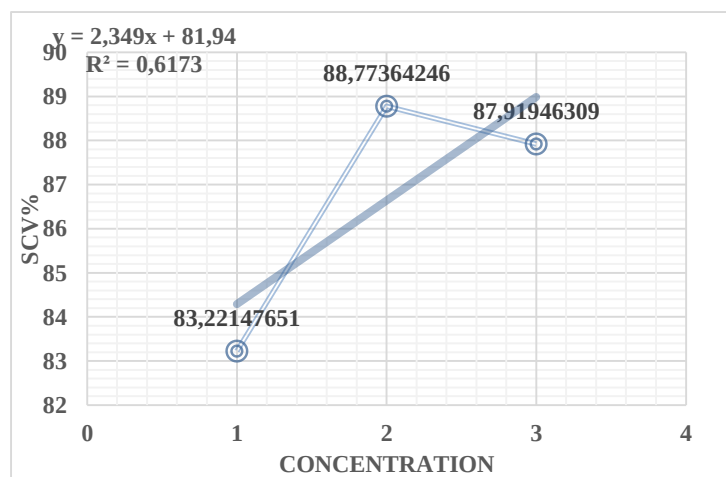
water: emanol extract



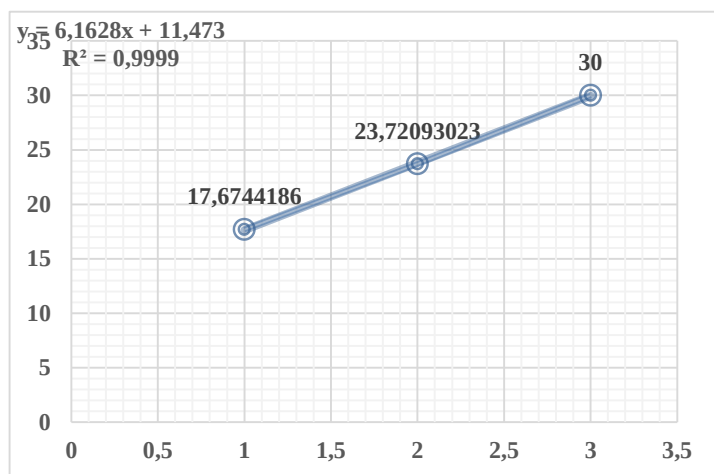
Ethanol extract



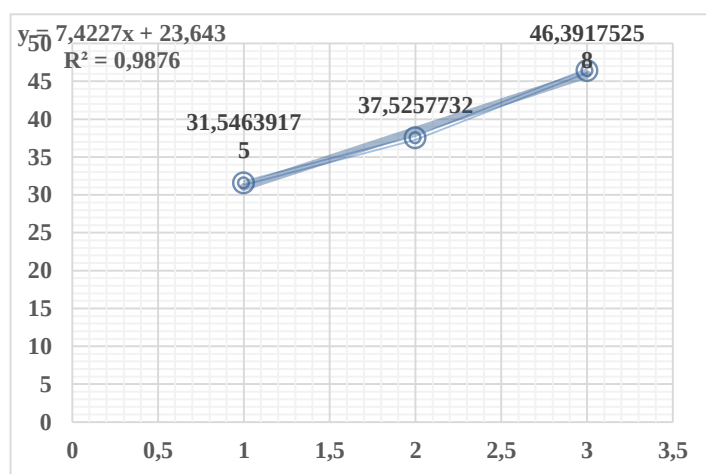
Hexane: ethanol extract



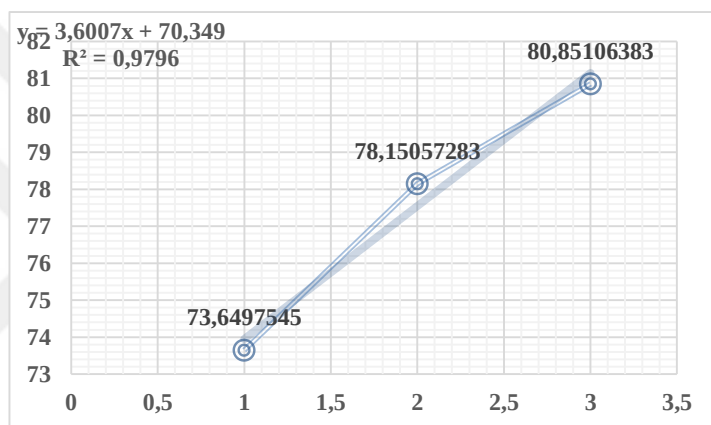
Hexane extract



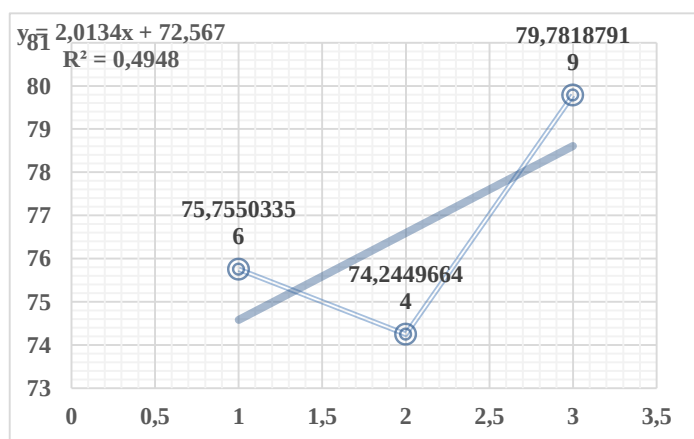
Water extract



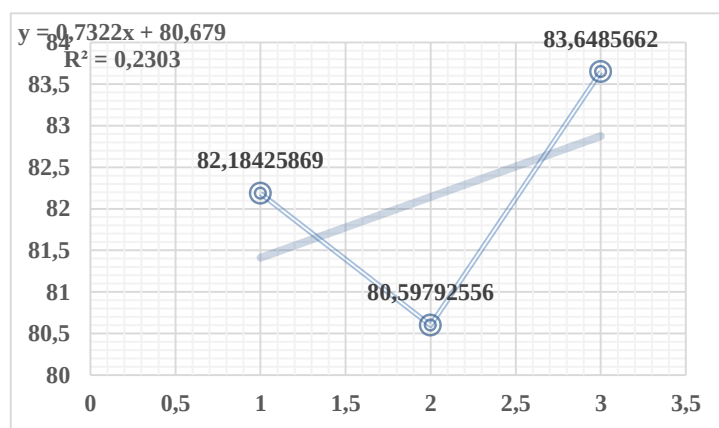
Water: ethanol extract



Ethanol extract

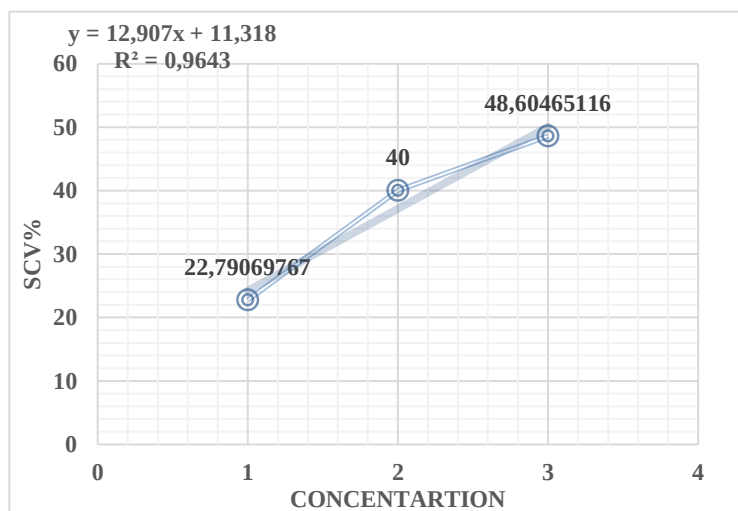


Hexane: ethanol extract

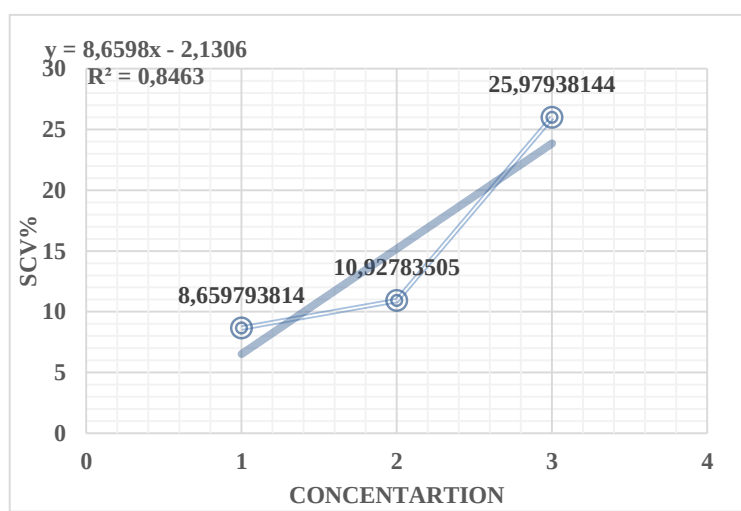


Hexane extract

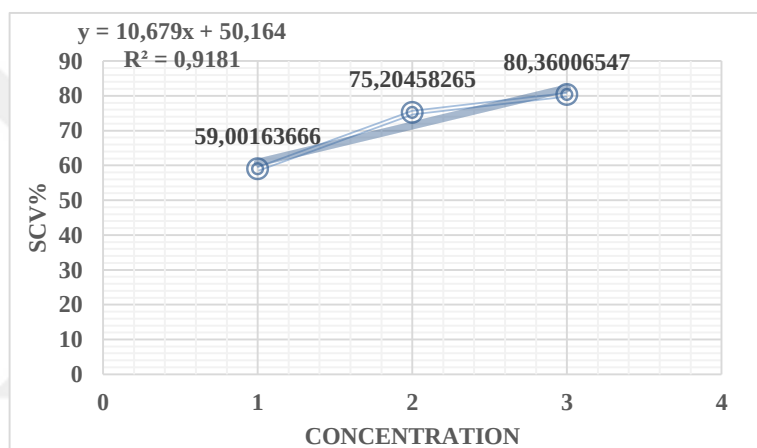
Figure 0.12. DPPH free radical scavenging activity of *Capparis spinosa* extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC₅₀ expressed as median inhibitory concentration (µg/mL).



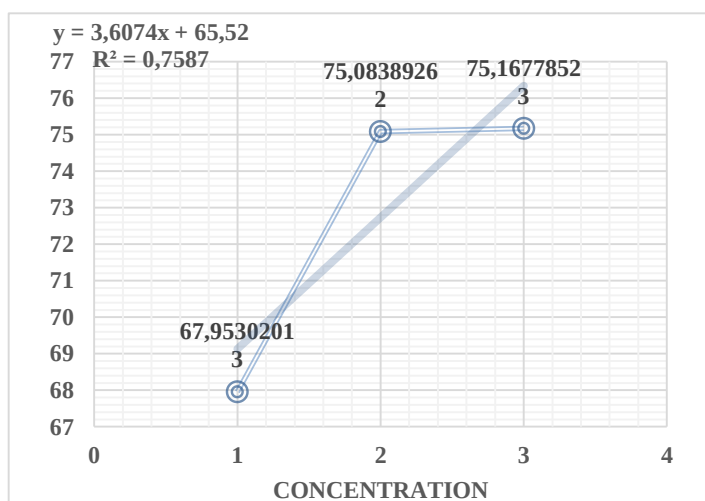
Water extract



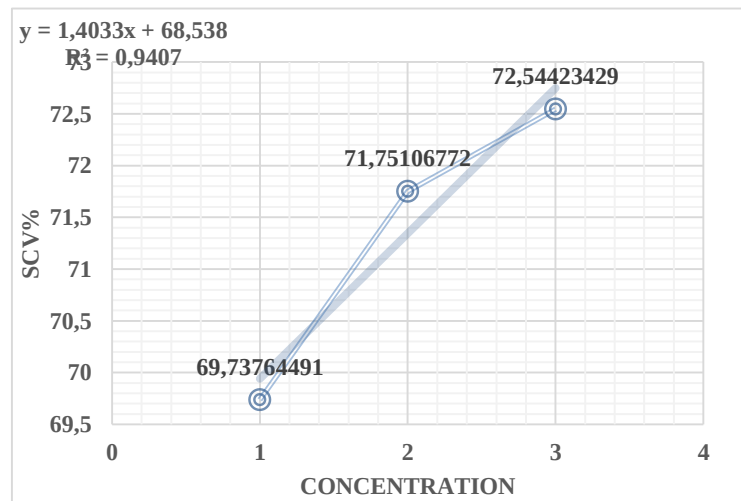
Water: ethanol extract



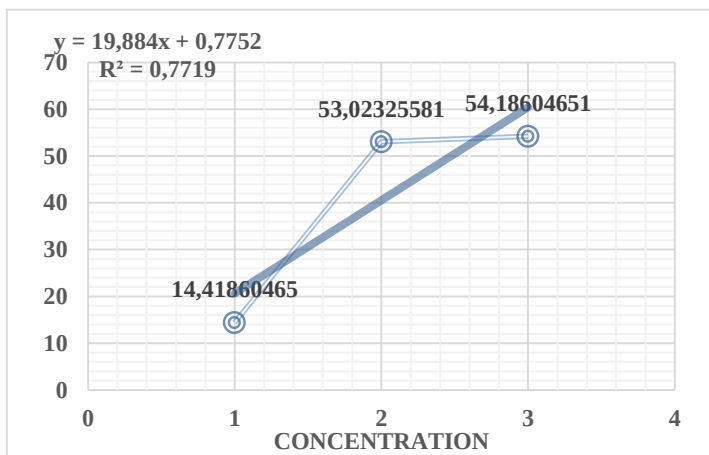
Ethanol extract



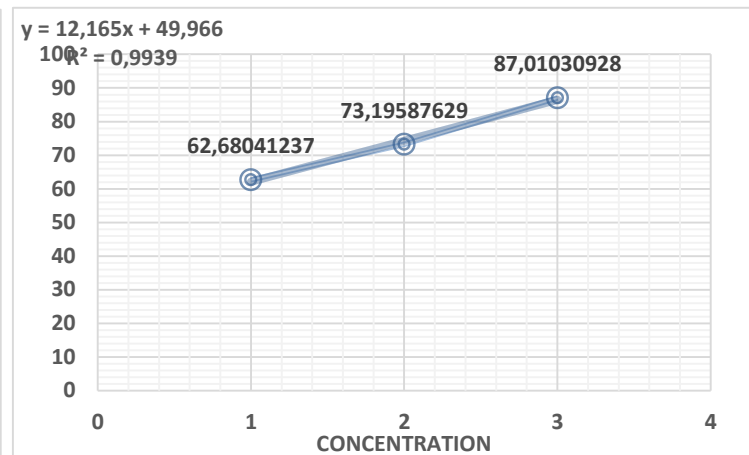
Ethanol: hexane extract



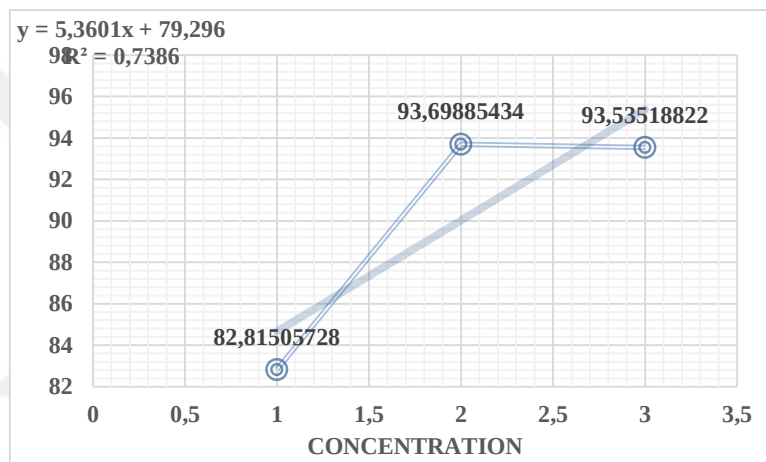
Hexane extract



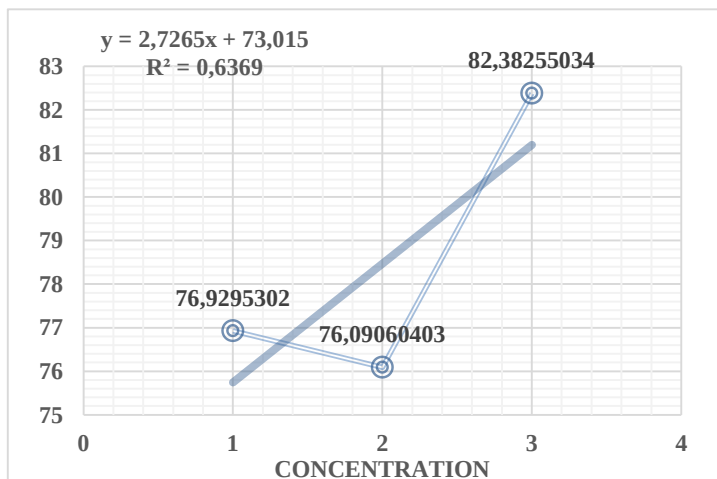
Water extract



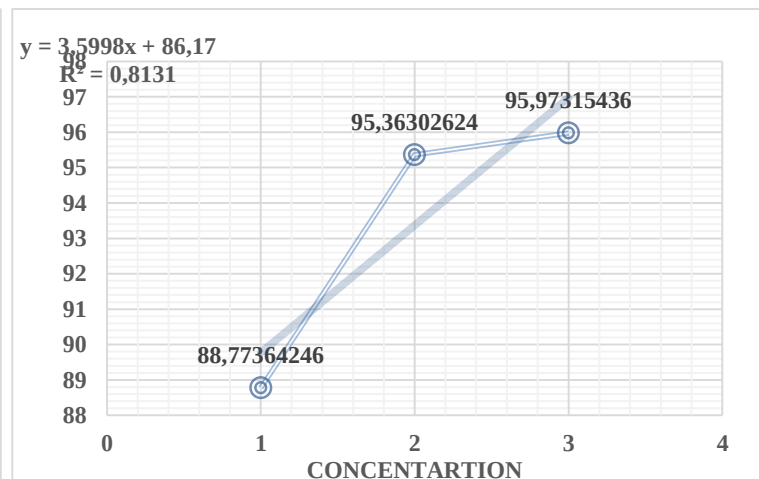
Ethanol: water extract



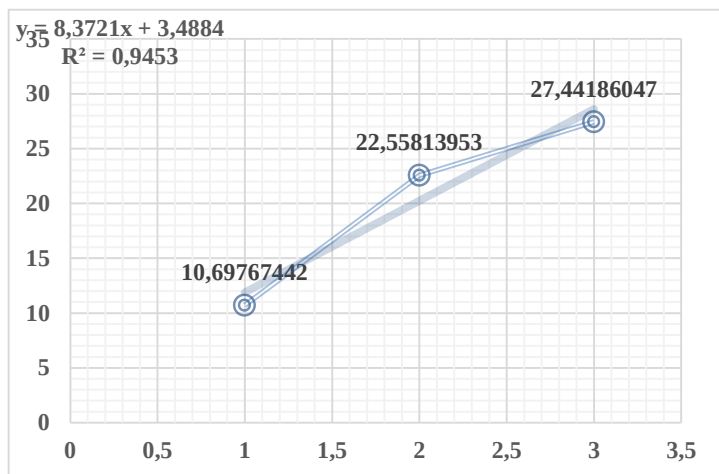
Ethanol extract



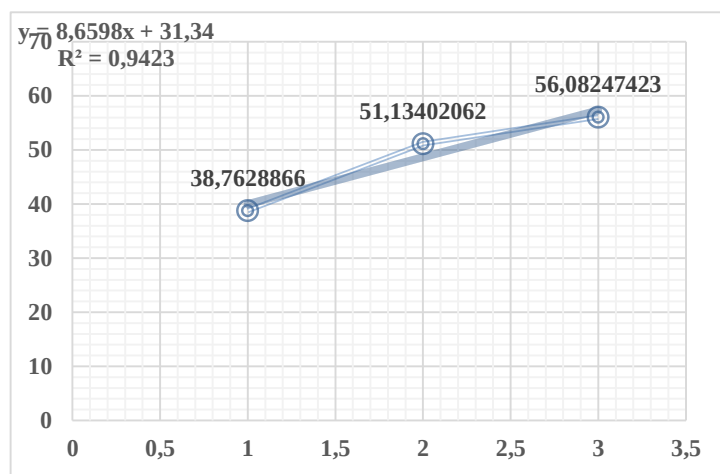
Hexane: ethanol extract



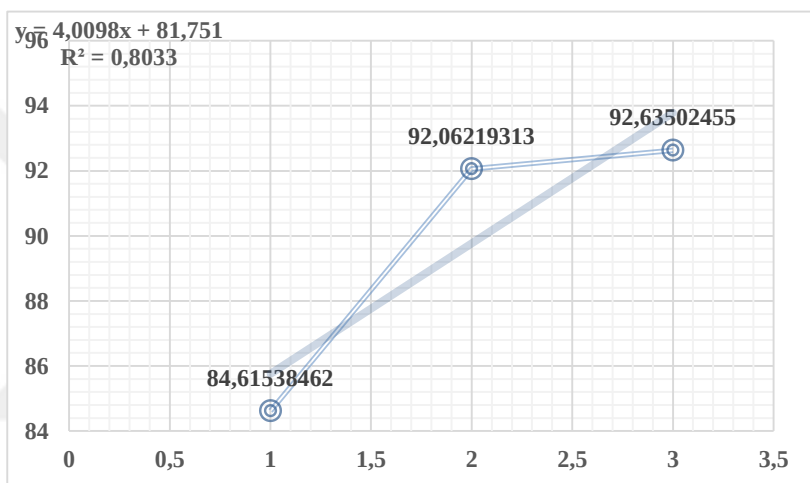
Hexane extract



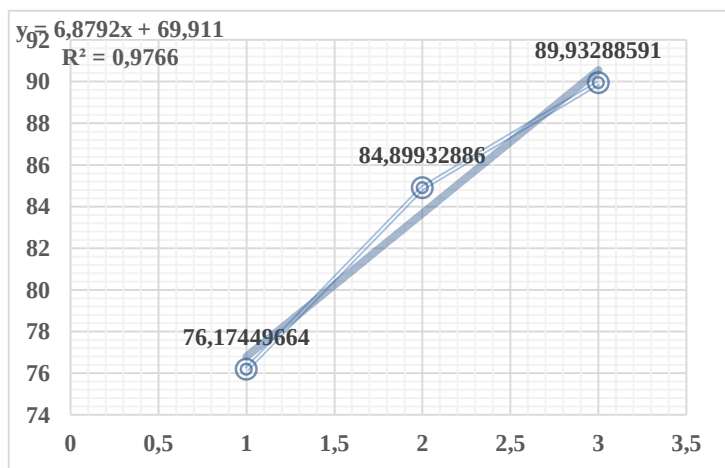
Water extract



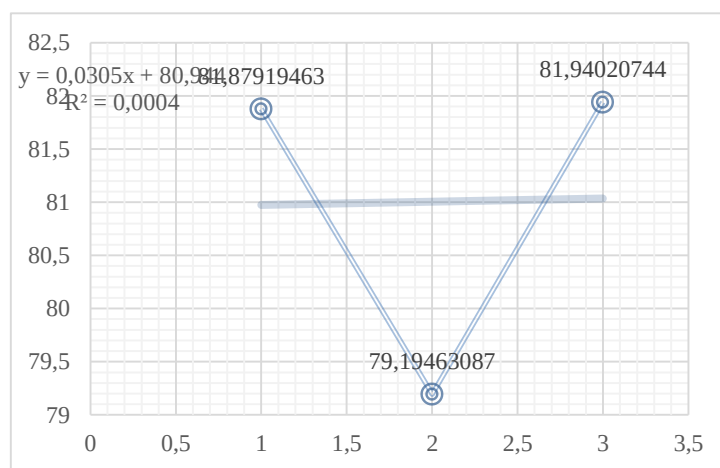
Water: ethanol extract



Ethanol extract

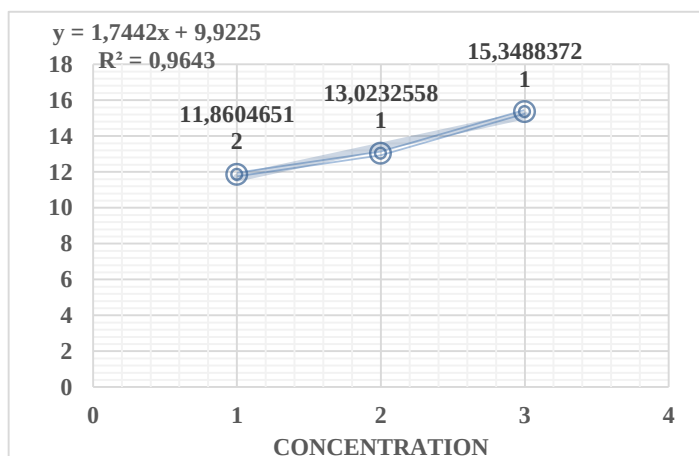


Ethanol: hexane extract

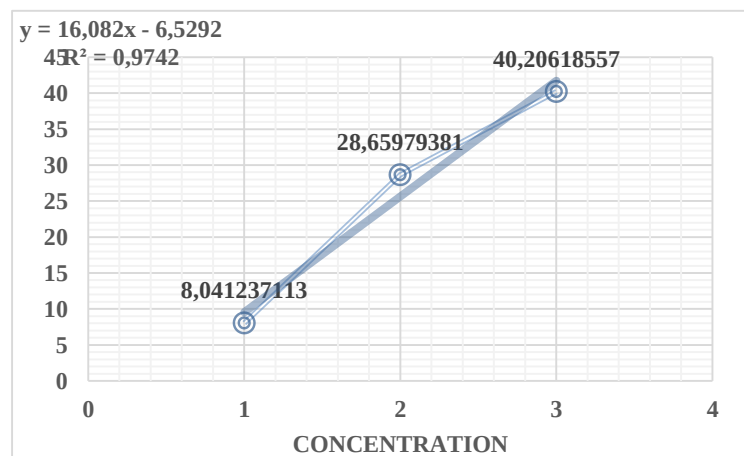


Hexane extract

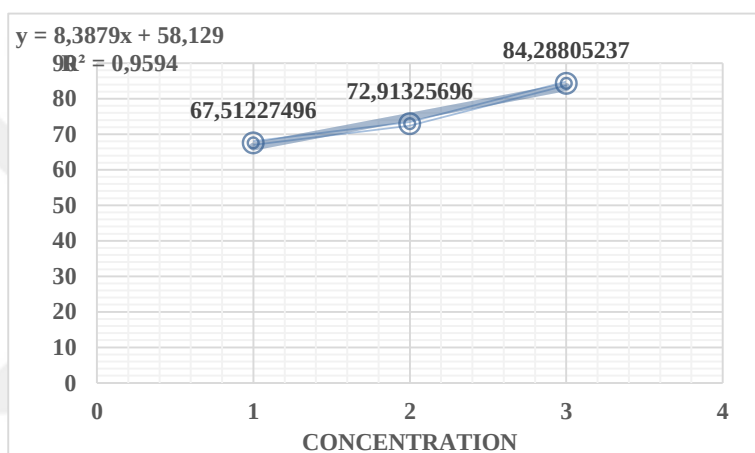
Figure 0.13. DPPH free radical scavenging activity of *Paliurus spina Christi* extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).



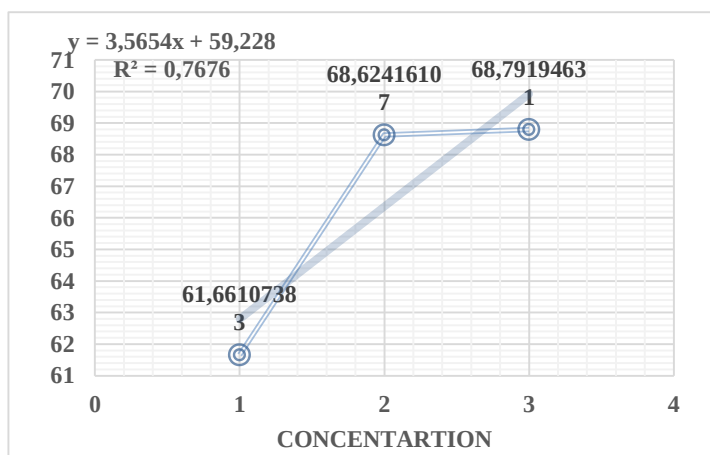
Water extract



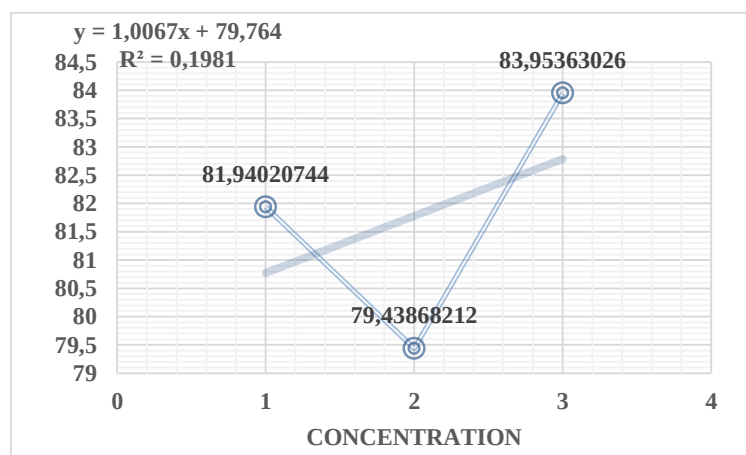
Water: ethanol extract



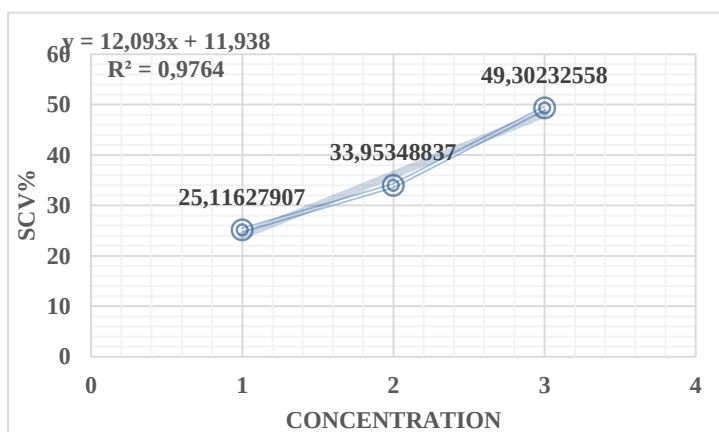
Ethanol extract



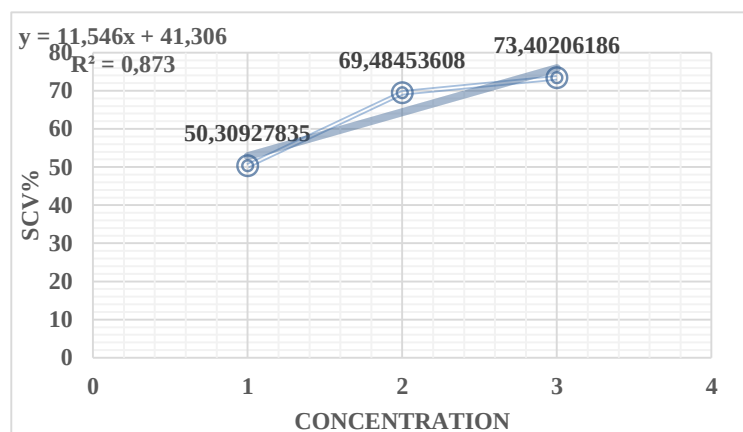
Hexane: ethanol extract



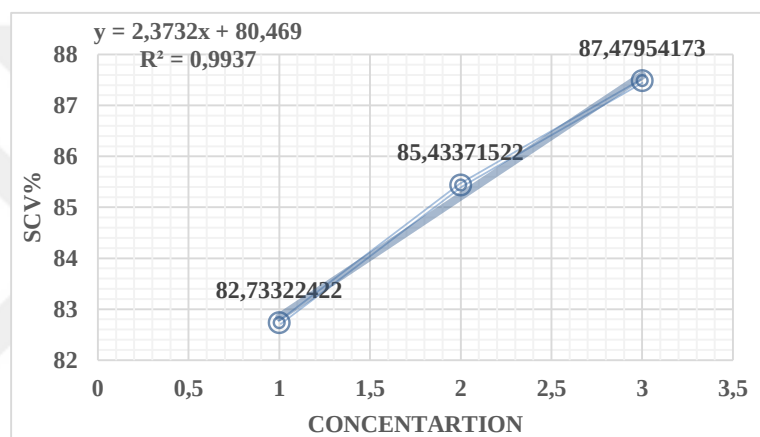
Hexane extract



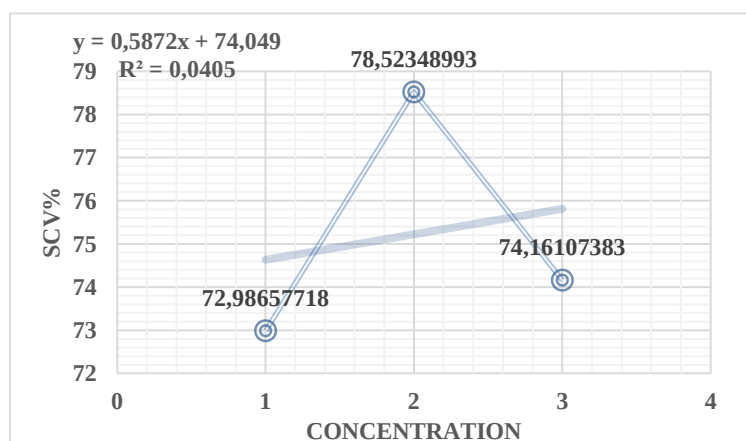
Water extract



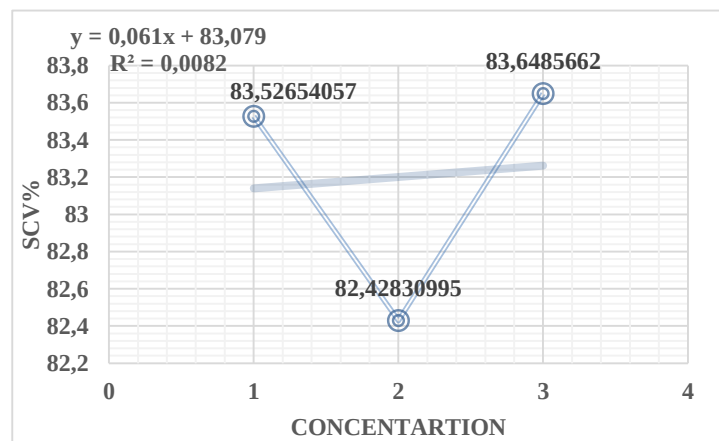
Ethanol: water extract



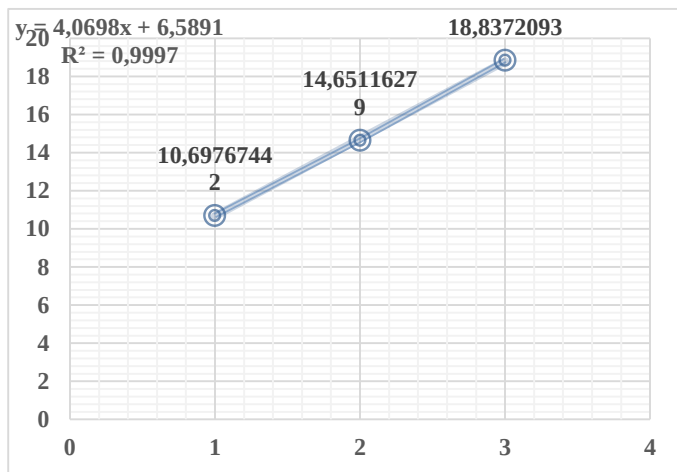
Ethanol extract



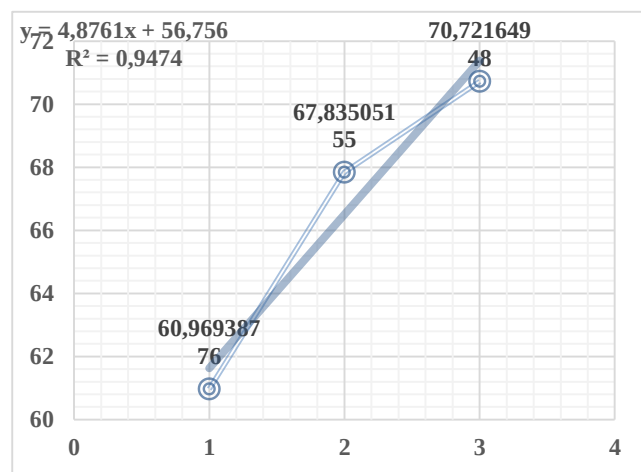
Hexane: ethanol extract



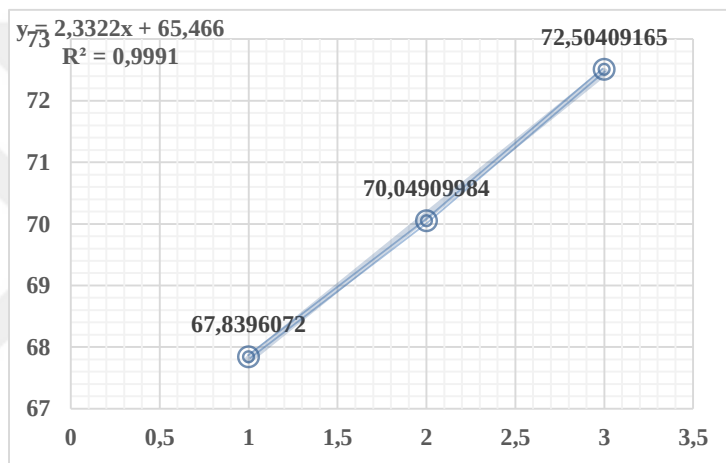
Hexane extract



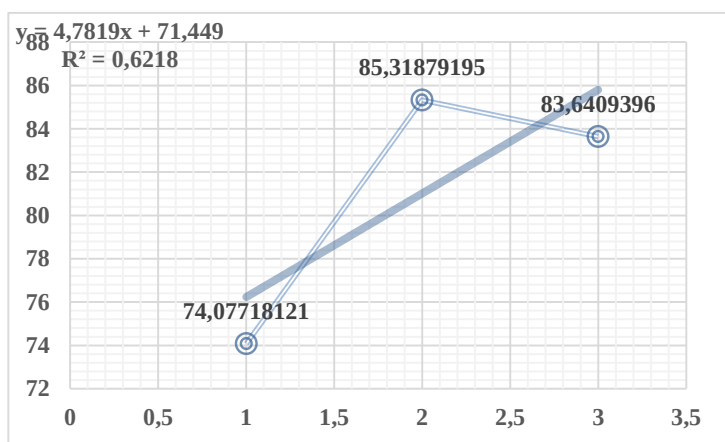
Water extract



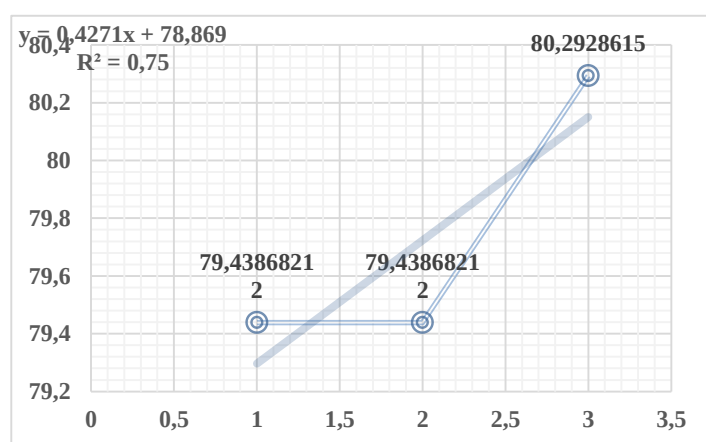
Water: ethanol extract



Ethanol extract

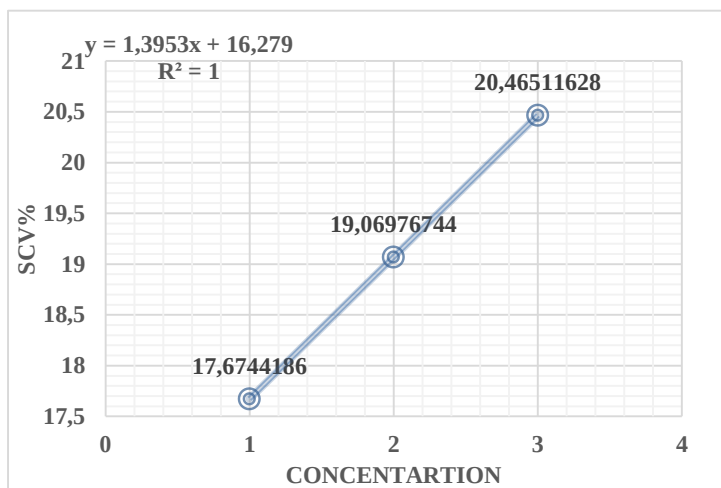


Hexane: ethanol extract

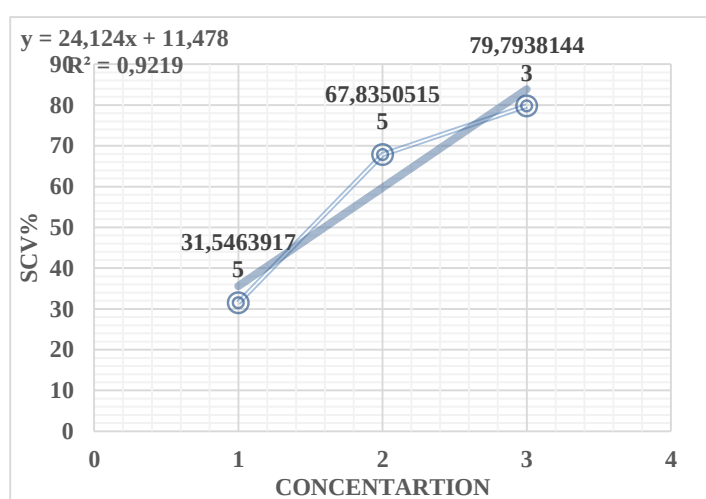


Hexane extract

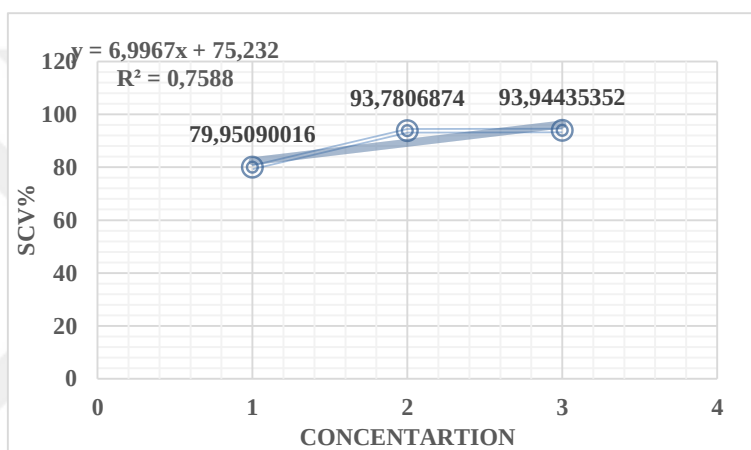
Figure 0.14. DPPH free radical scavenging activity of *Portulaca oleracea* extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration ($\mu\text{g/mL}$).



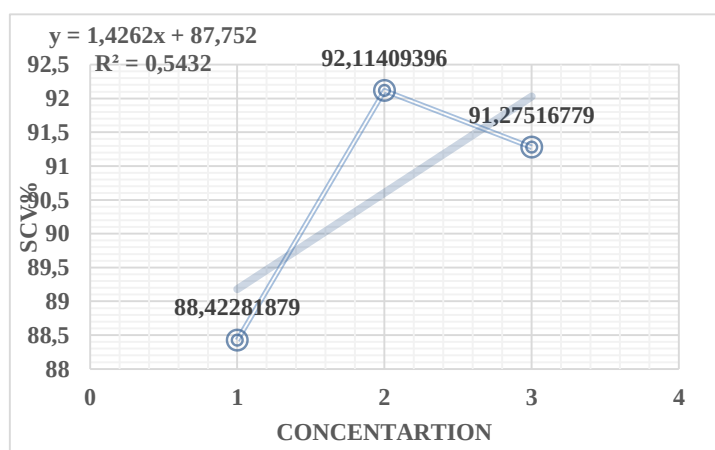
Water extract



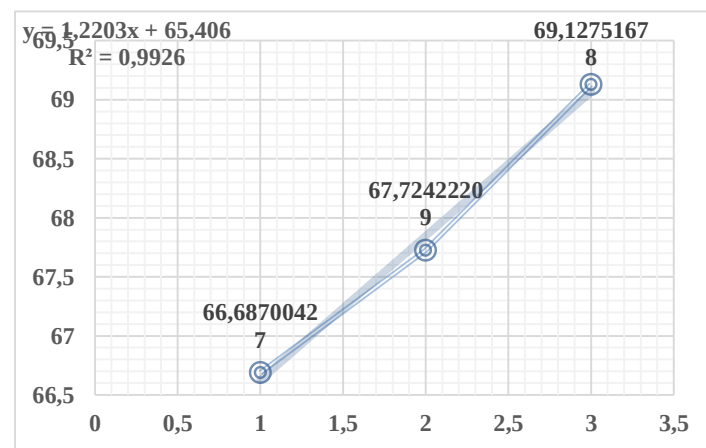
Water: ethanol extract



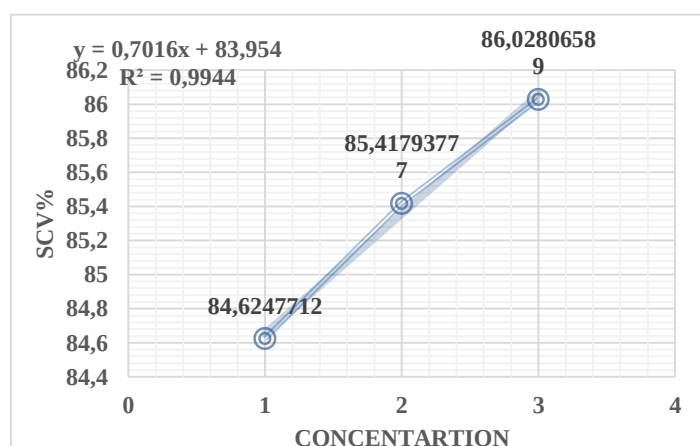
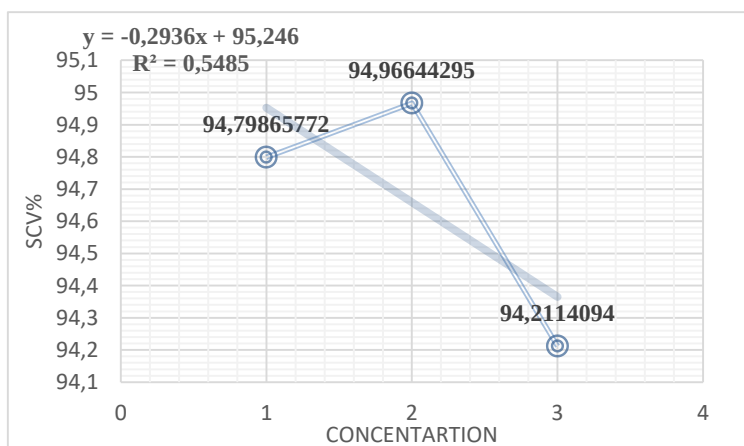
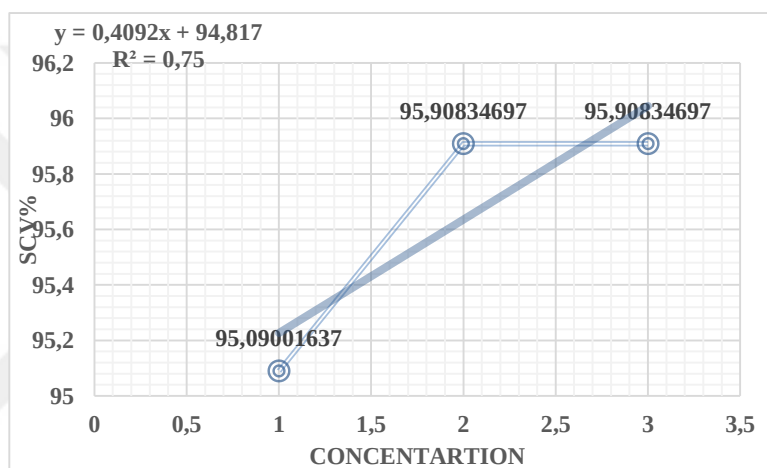
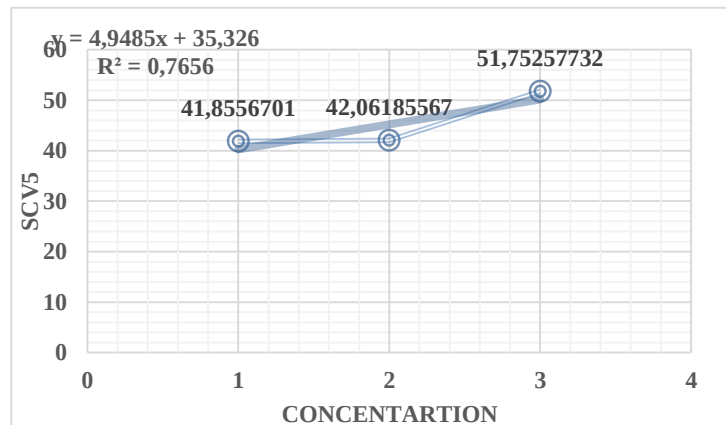
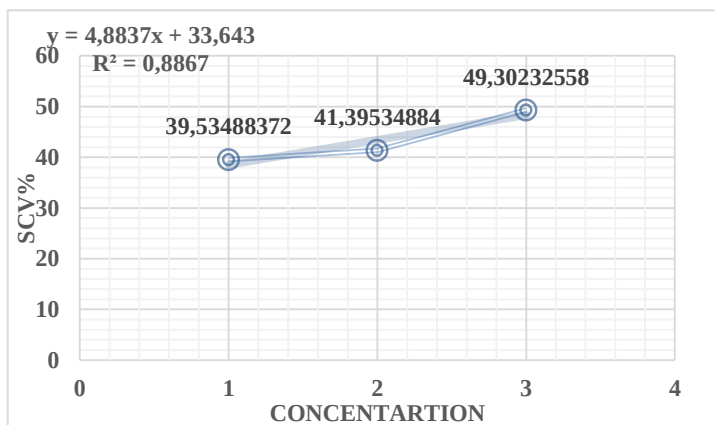
Ethanol

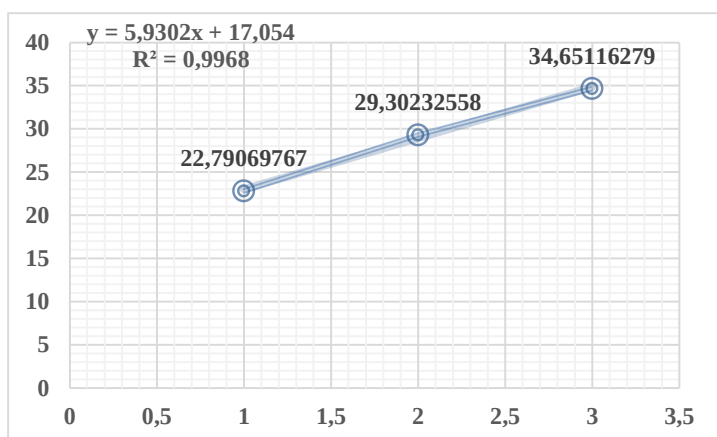


Hexane: ethanol extract

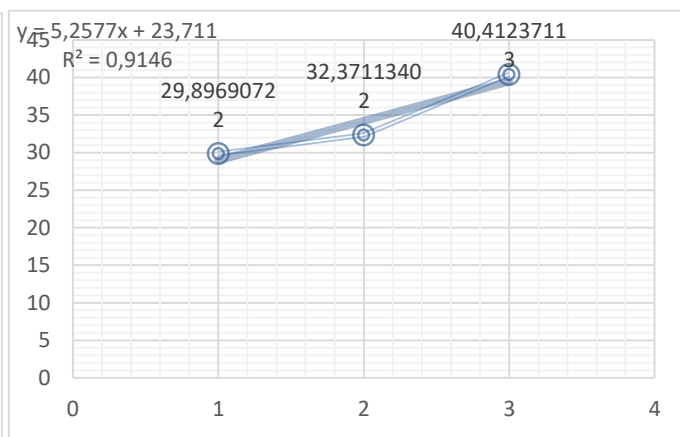


Hexane extract

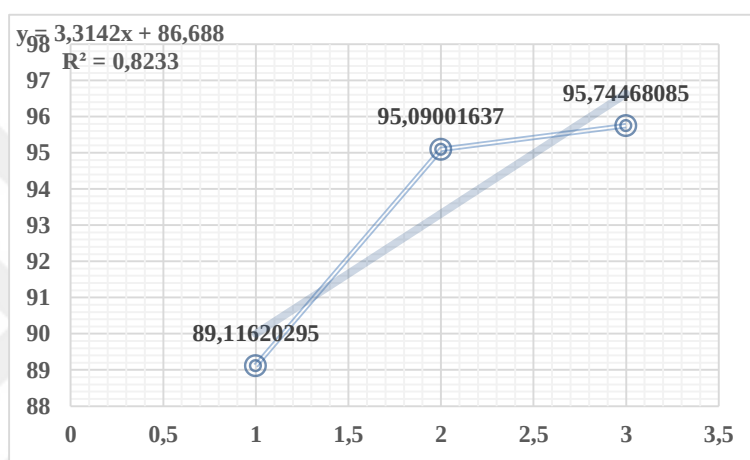




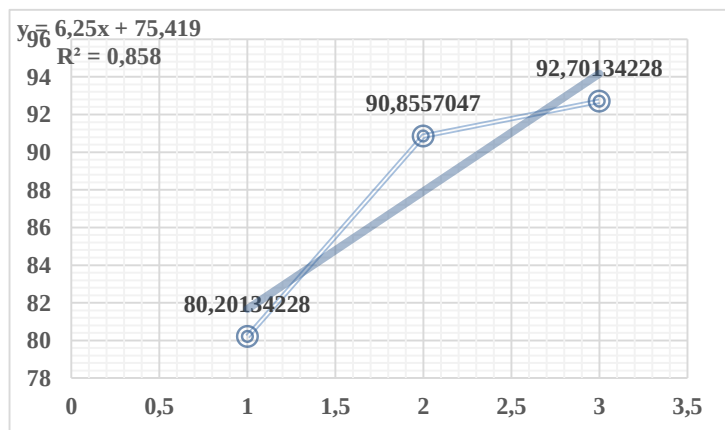
Water extract



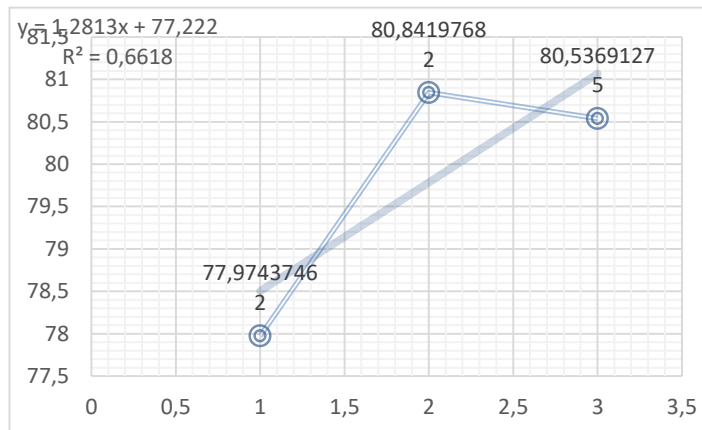
Water: ethanol extract



Ethanol extract

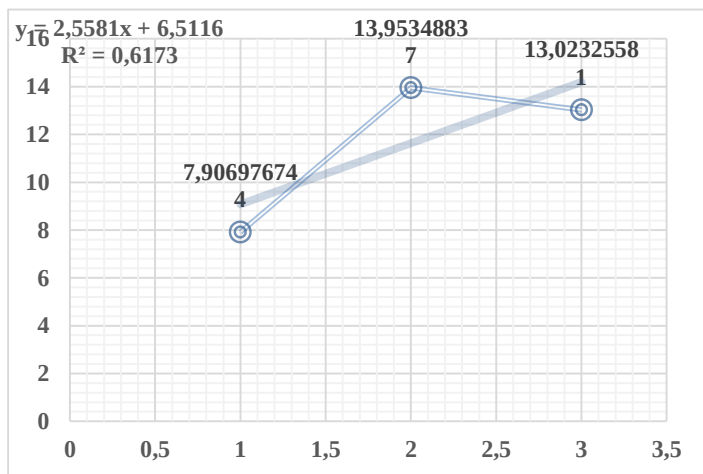


Hexan: ethanol extract

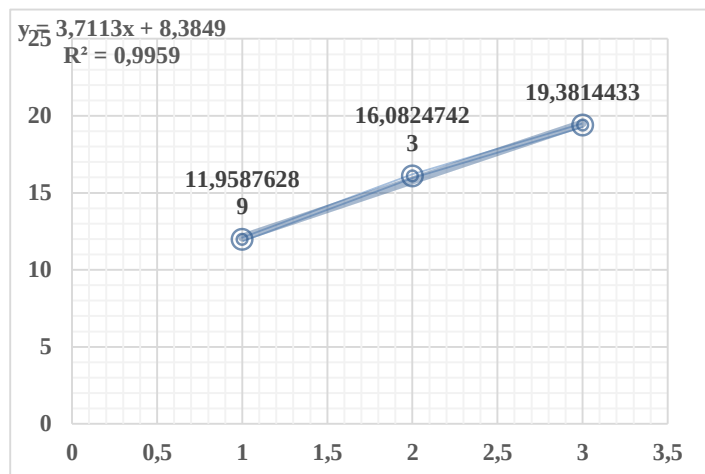


Hexane extract

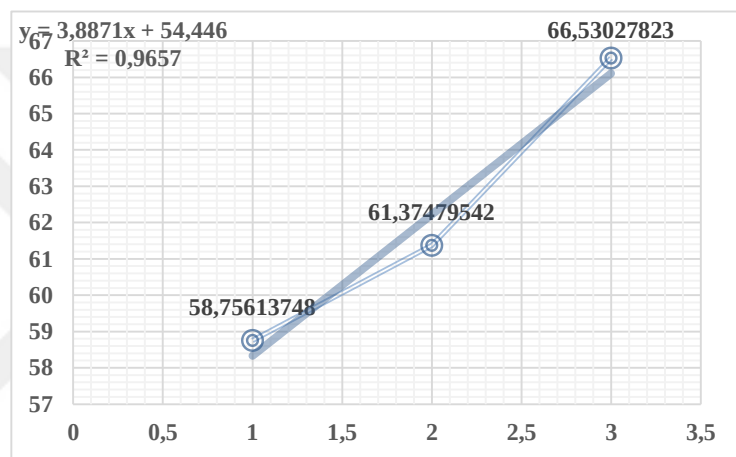
Figure 0.15. DPPH free radical scavenging activity of *Artemisia haussknechtii* boiss extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration (µg/mL).



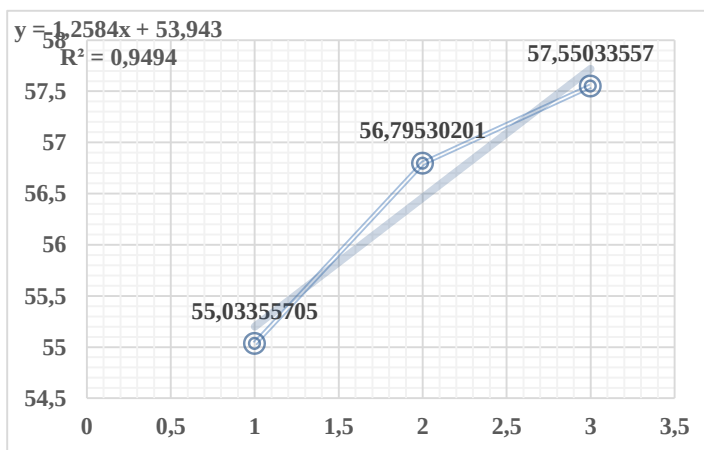
Water extract



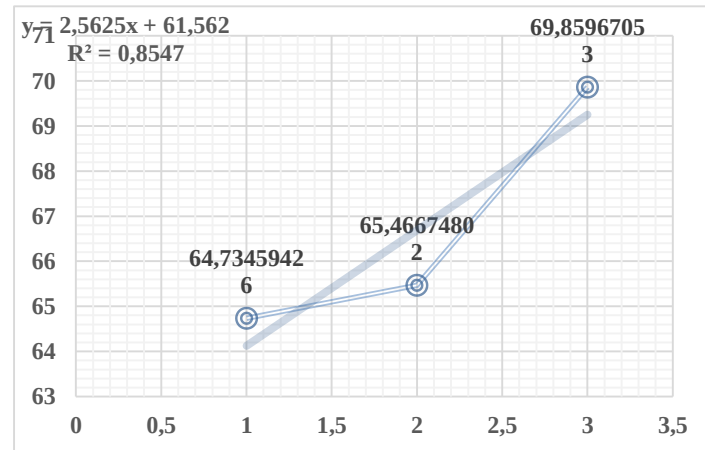
Water: ethanol extract



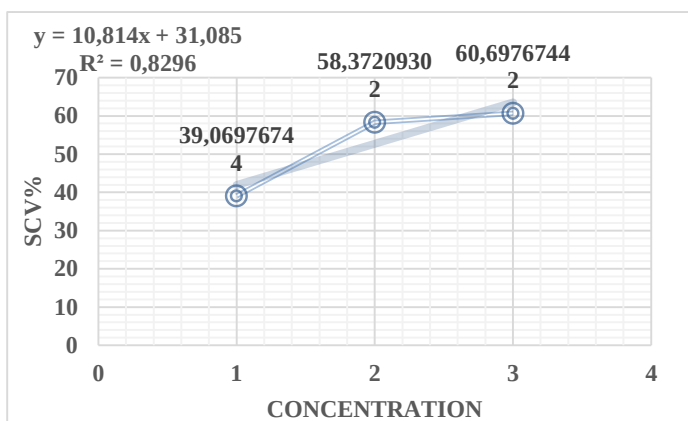
Ethanol extract



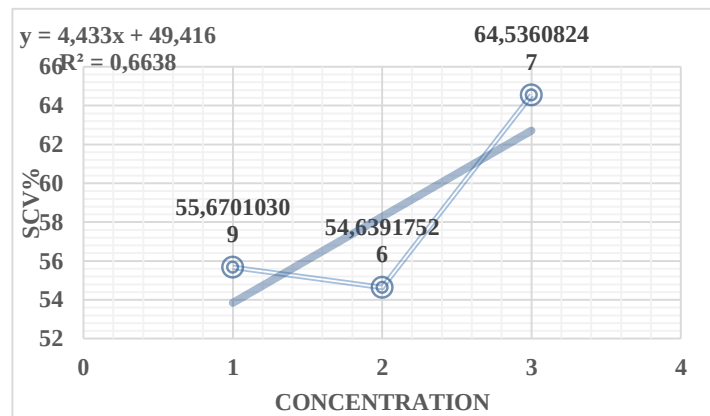
Hexane: ethanol extract



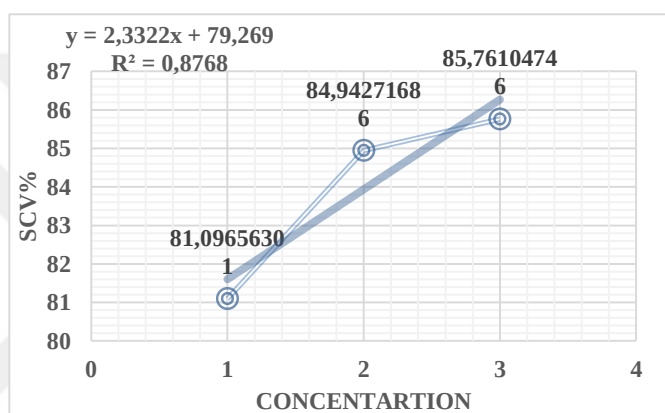
Hexane extract



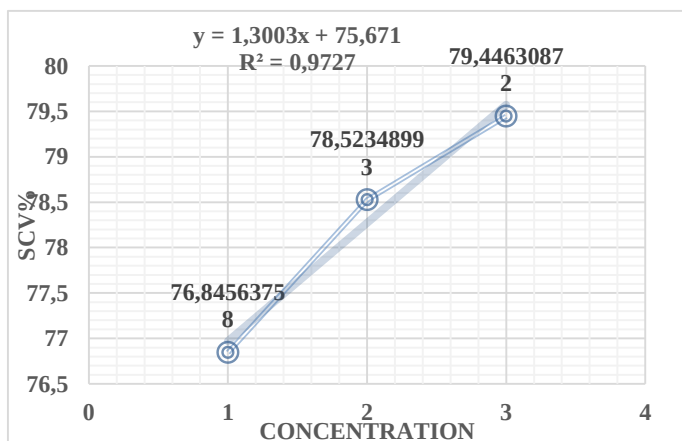
Water extract



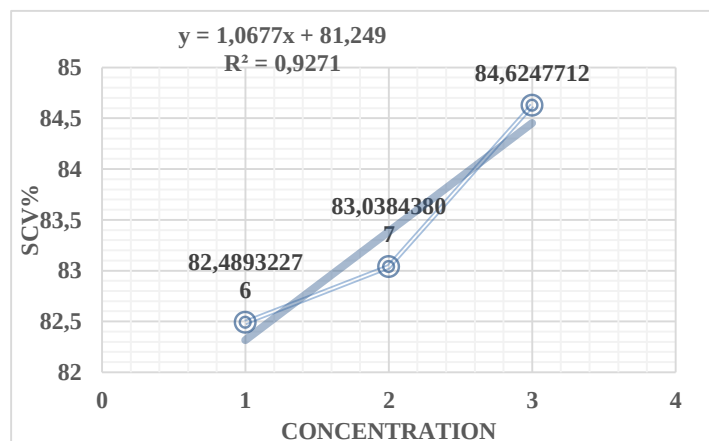
Water: ethanol extract



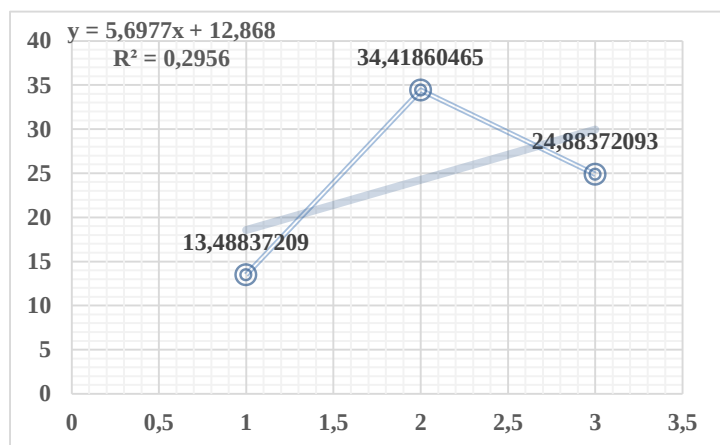
Ethanol extract



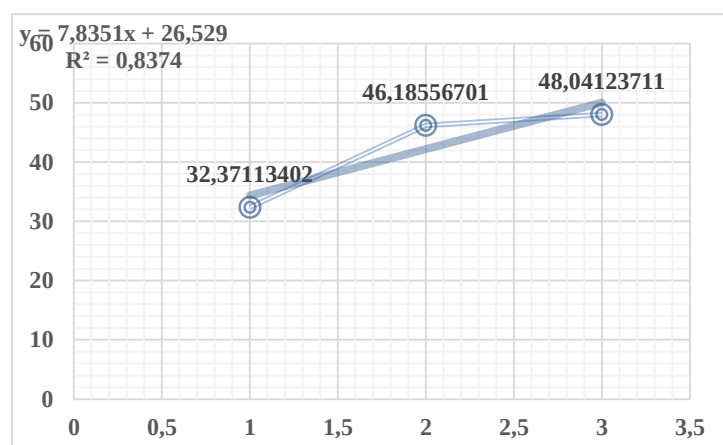
Hexane: ethanol extract



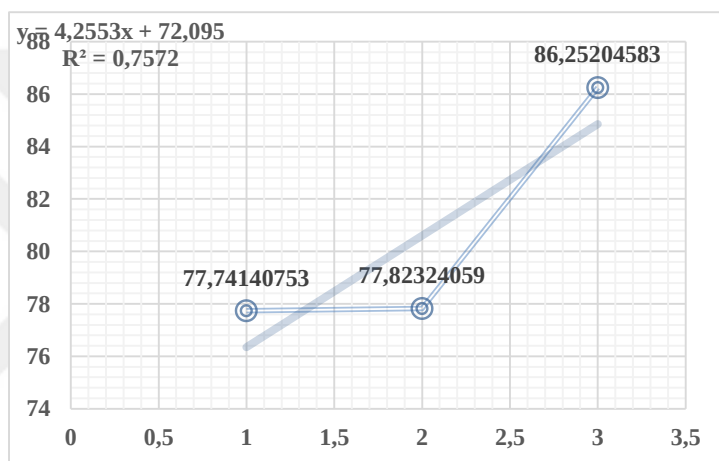
Hexane extract



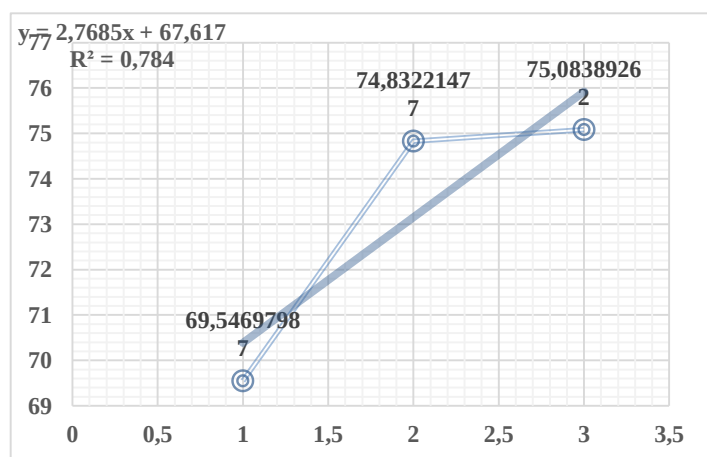
Water extract



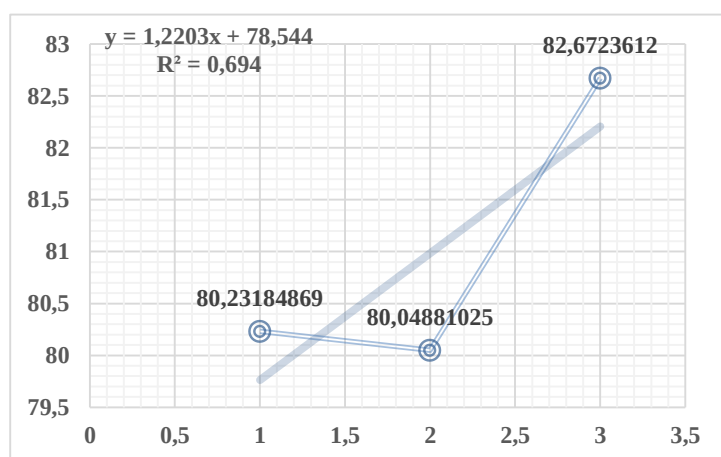
Water: ethanol extract



Ethanol extract

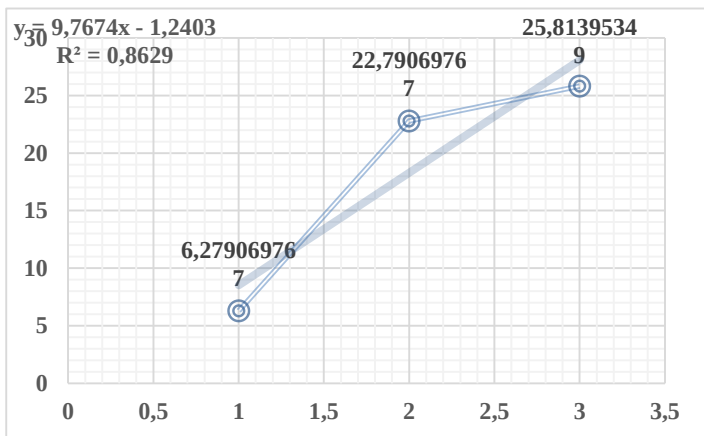


Hexane: ethanol extract

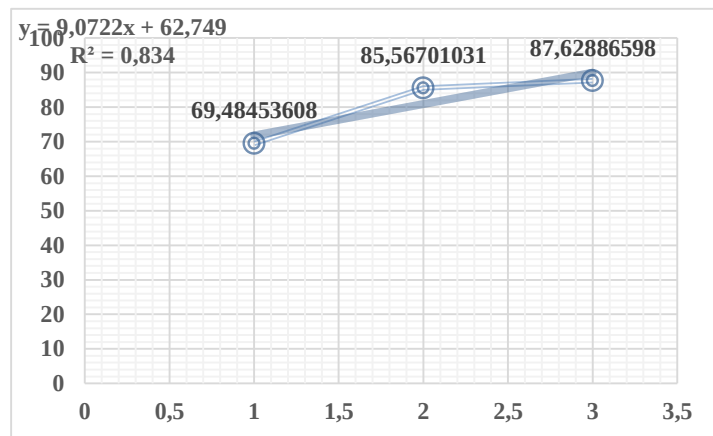


Hexane extract

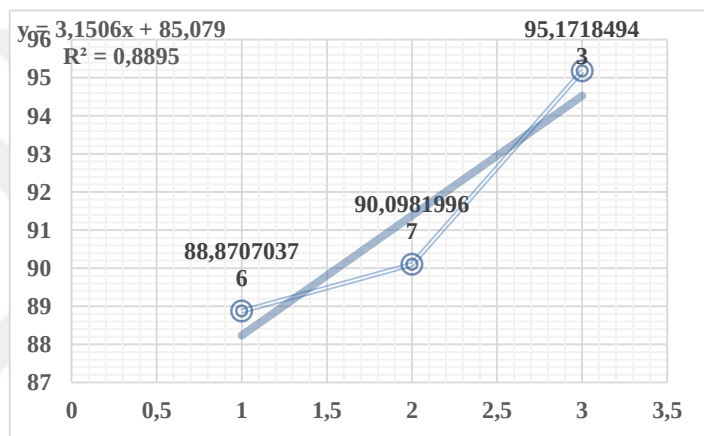
Figure 0.16. DPPH free radical scavenging activity of *Prunus microcarpa* extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration (µg/mL).



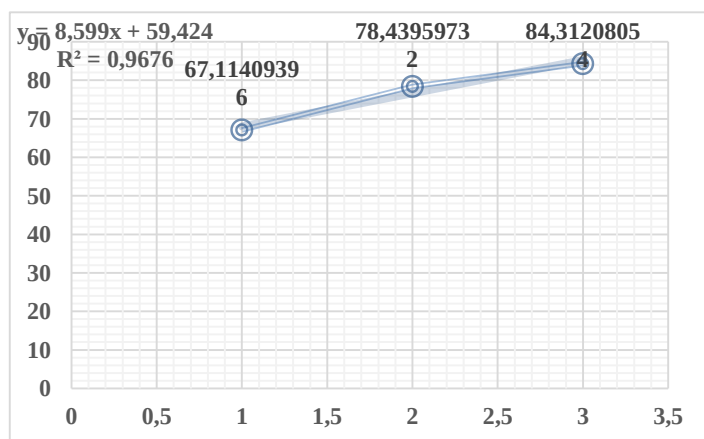
Water extract



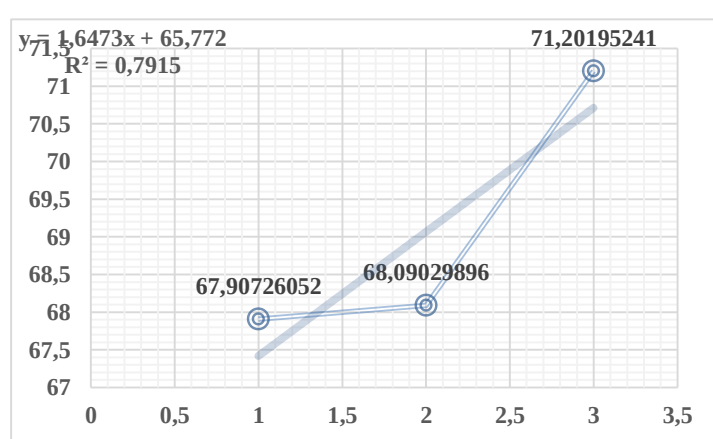
Water: ethanol extract



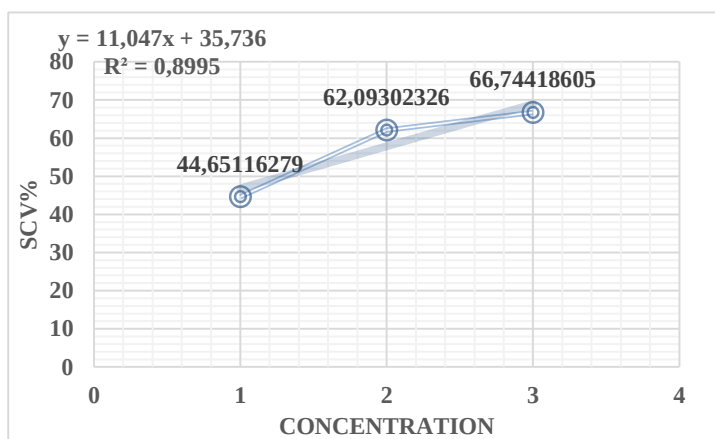
Ethanol extract



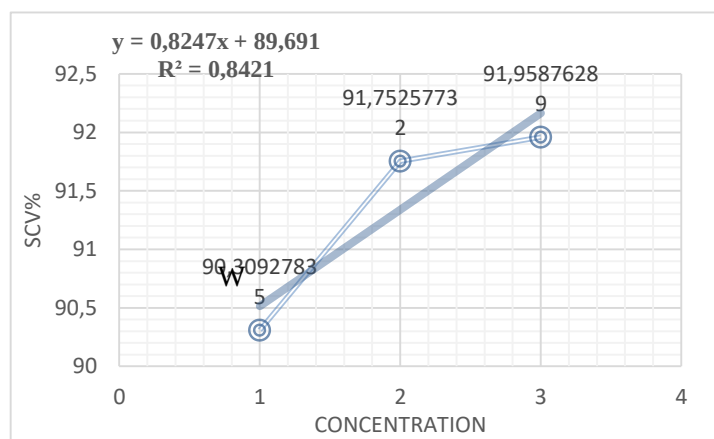
Hexane: ethanol extract



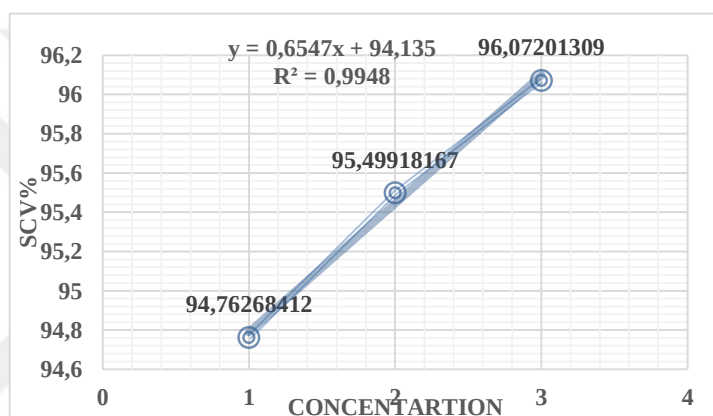
Hexane extract



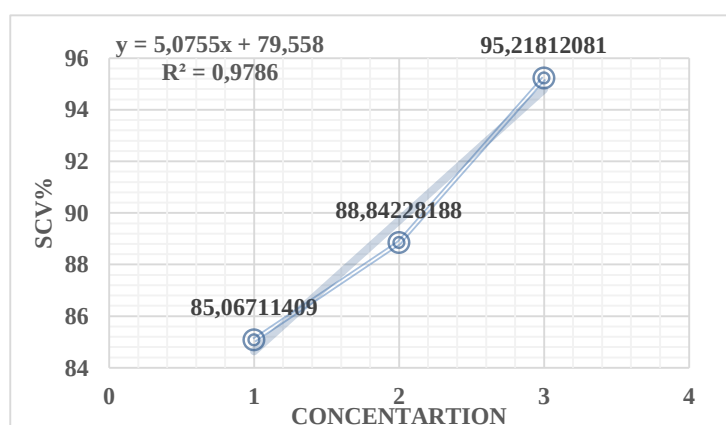
Water extract



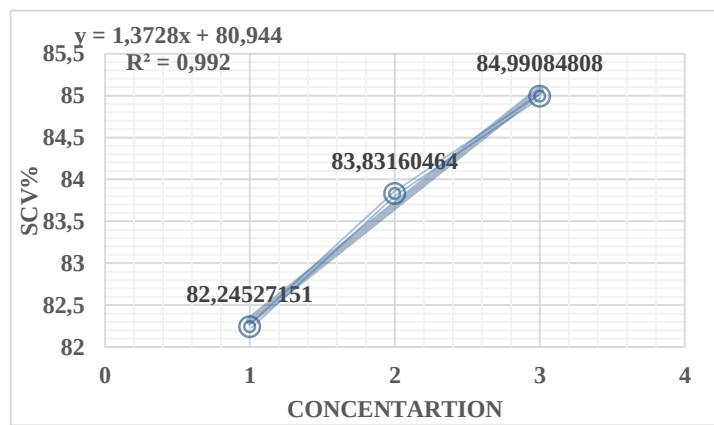
Water: ethanol extract



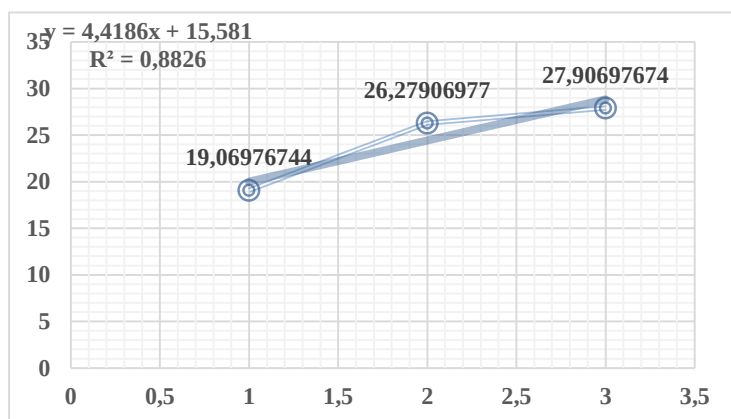
Ethanol extract



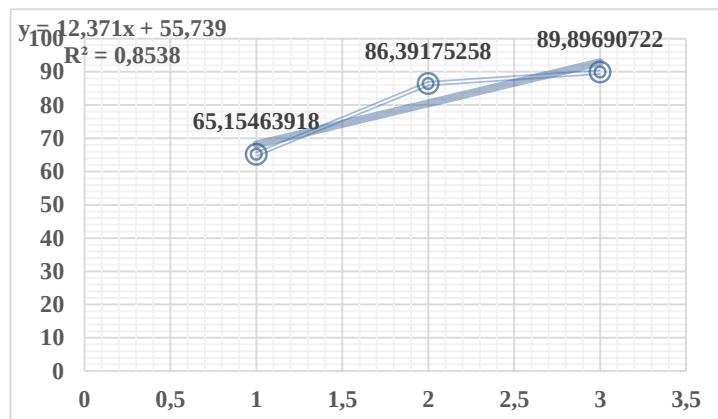
Hexane: ethanol extract



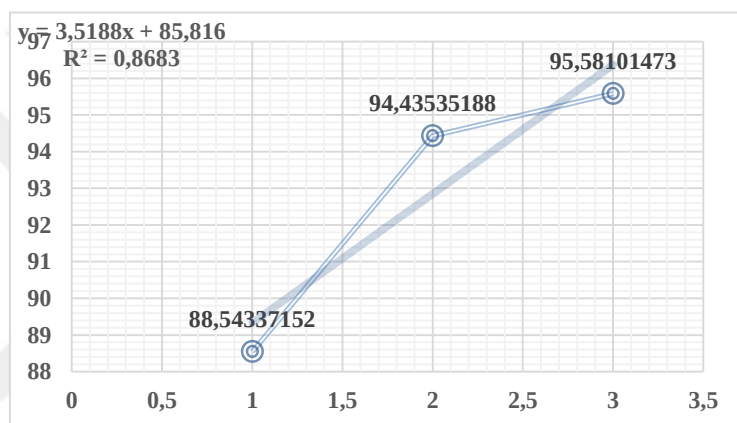
Hexane extract



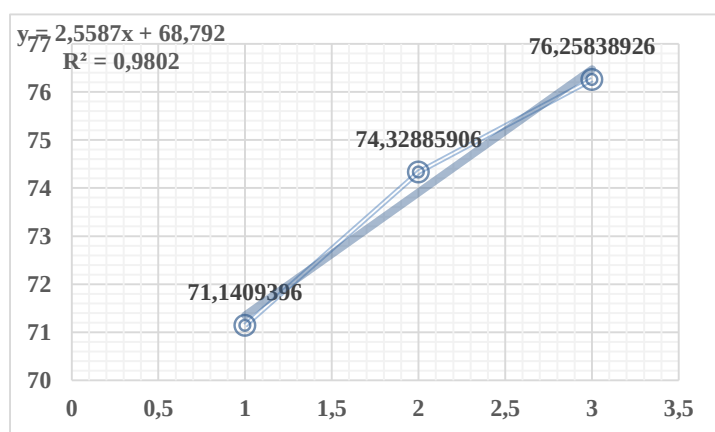
Water extract



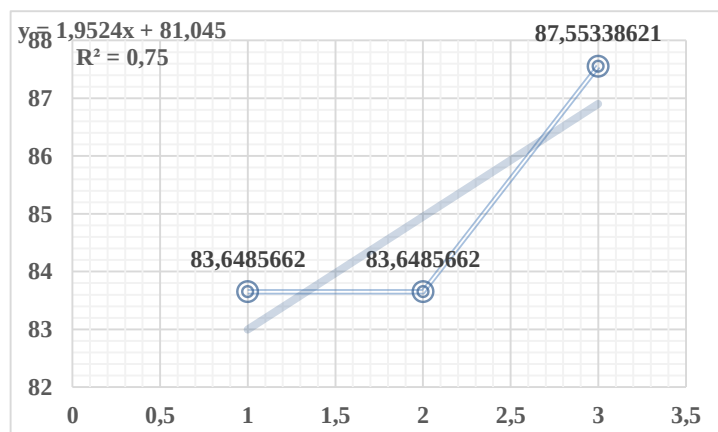
Water: ethanol extract



Ethanol extract

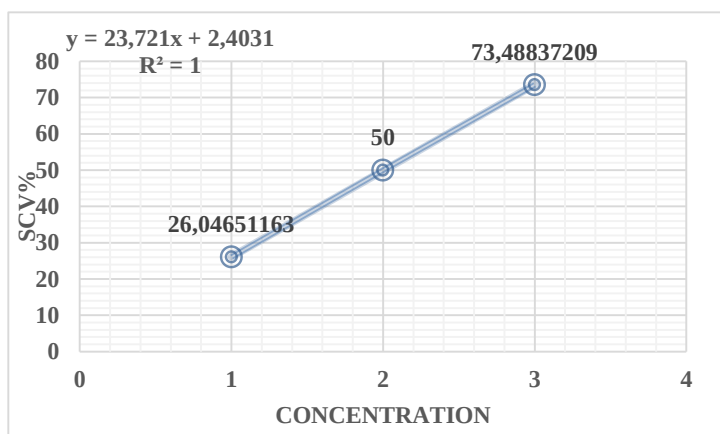


Hexane: ethanol extract

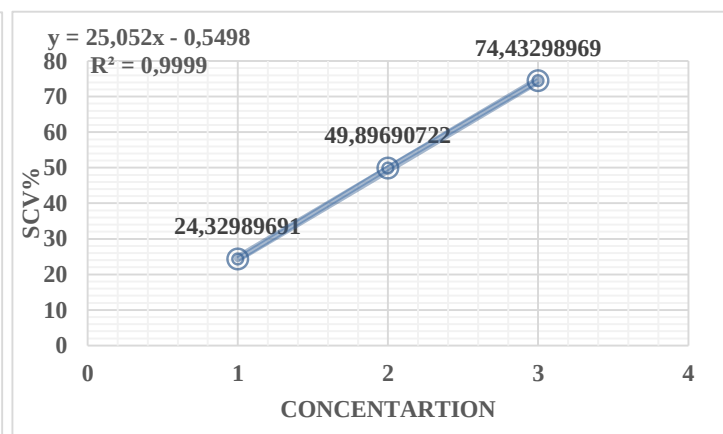


Hexane extract

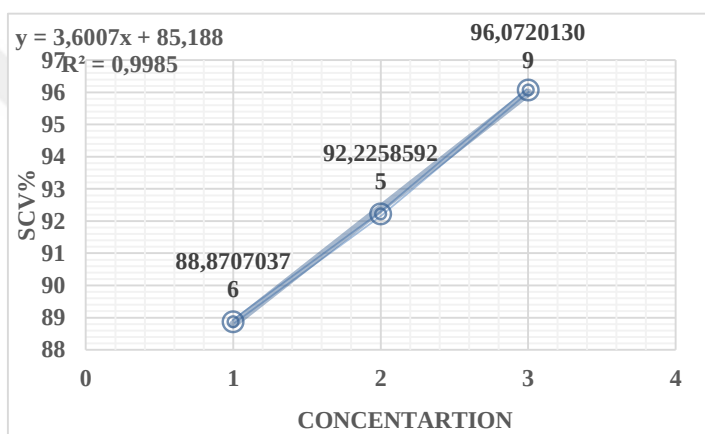
Figure 0.17. DPPH free radical scavenging activity of *Silybum Marianum* extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration (μg/mL).



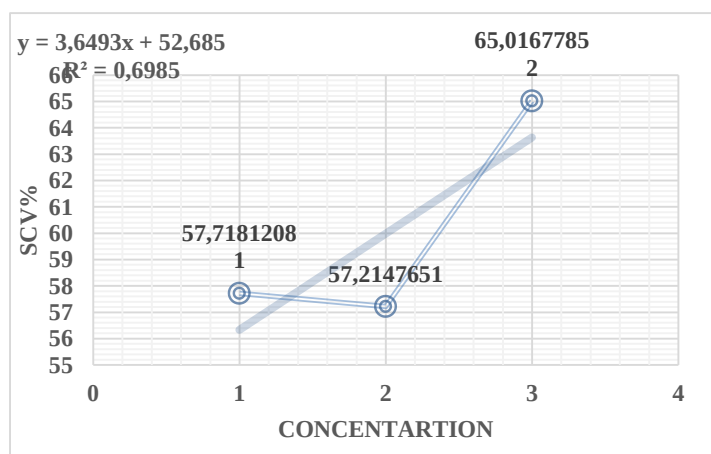
Water extract



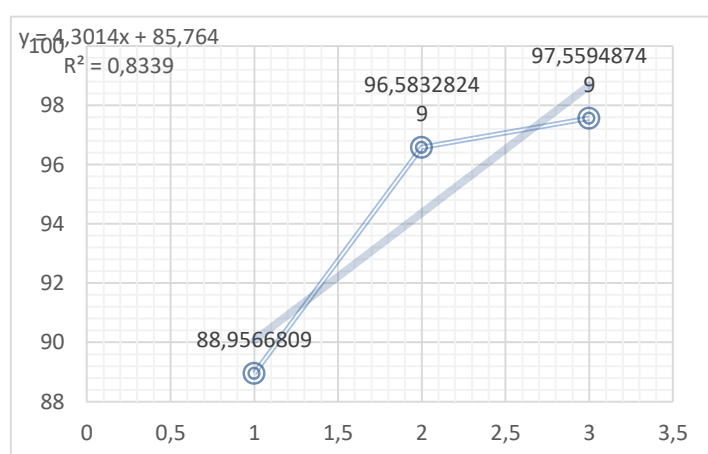
Water: ethanol extract



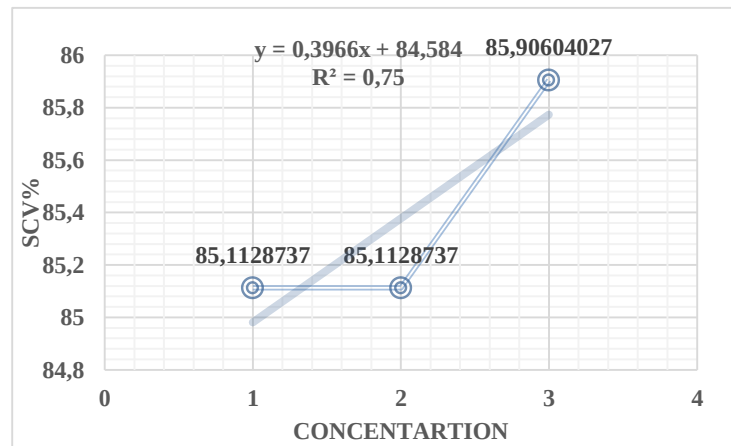
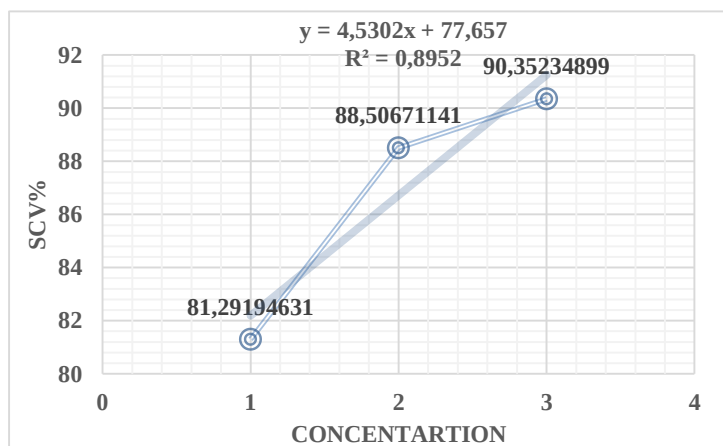
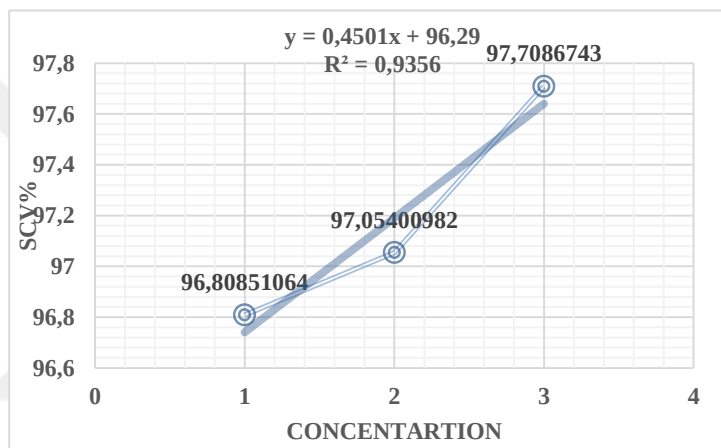
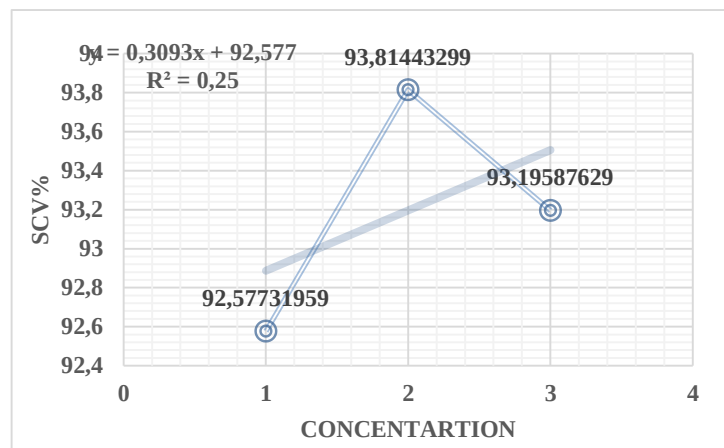
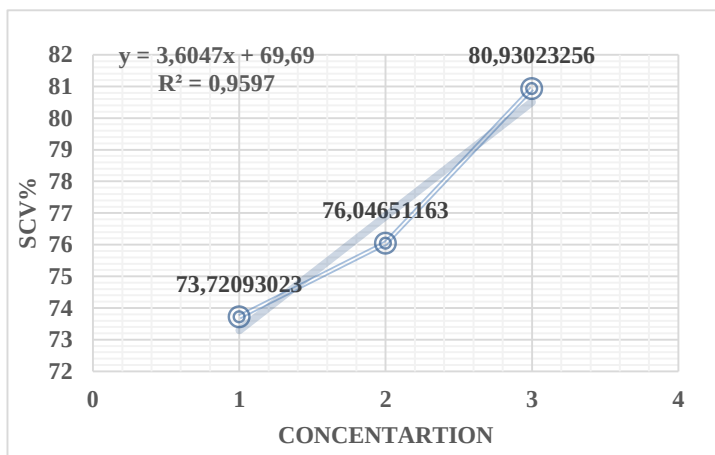
Ethanol extract

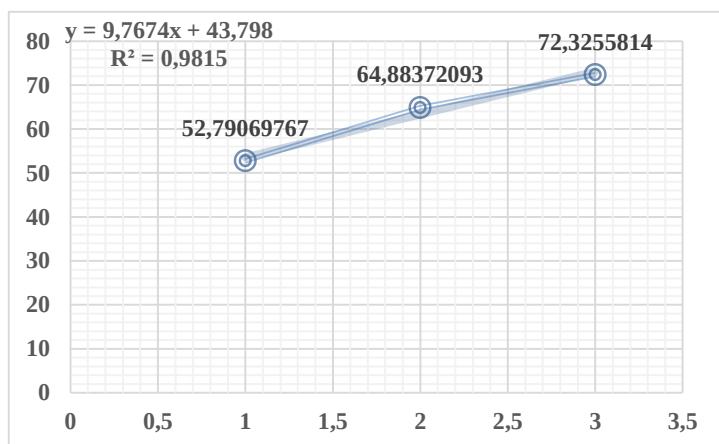


Hexane: ethanol extract

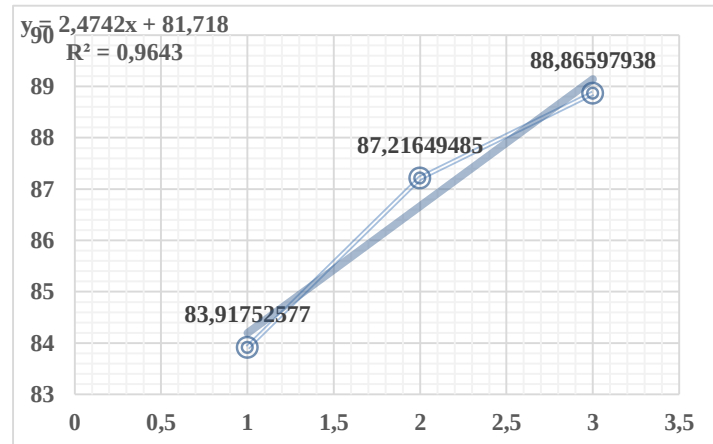


Hexane extract

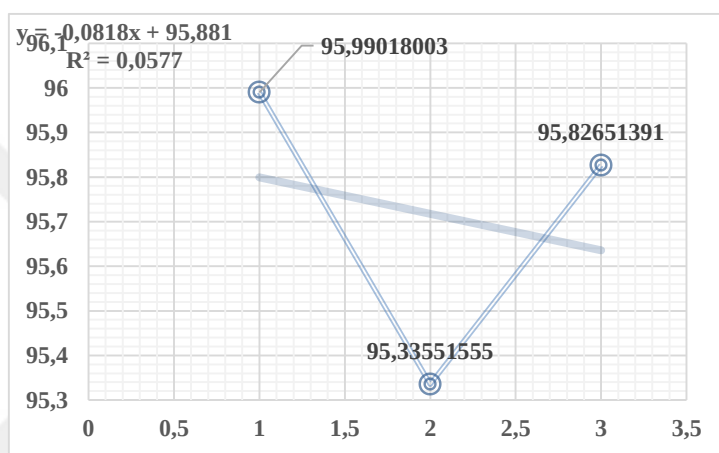




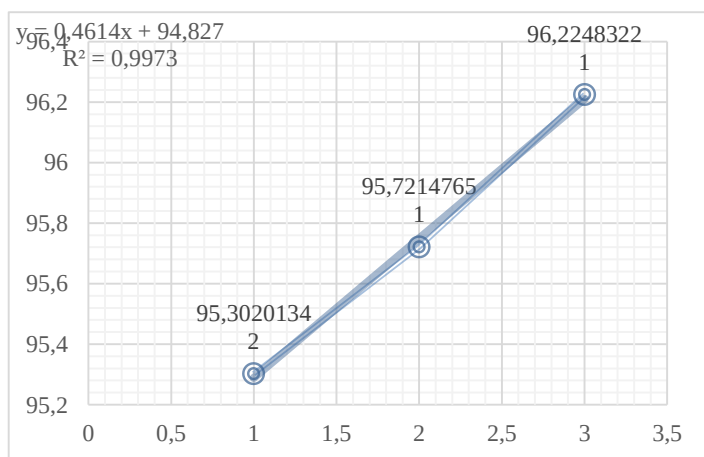
Water extract



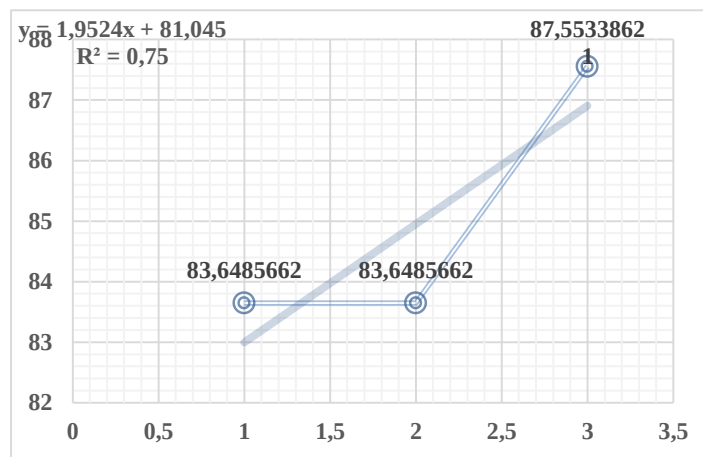
Water: ethanol extract



Ethanol extract



Hexane: ethanol extract



Hexane extract

Figure 0.18. DPPH free radical scavenging activity of *Pistacia eurycarpa* Yale extracts by methods of (Conventional, Microwave, and Orbital shaker), compared with the standard BHT, Trolox, and IC50 expressed as median inhibitory concentration (μg/mL).