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INVESTIGATION OF THE EFFECT OF PHYTIC ACID AND ITS RELATIONSHIP WITH INHIBIN B, TRACE ELEMENT AND SOME BIOCHEMICAL MARKERS IN INFERTILE MEN

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INVESTIGATION OF THE EFFECT OF PHYTIC ACID AND ITS RELATIONSHIP WITH INHIBIN B, TRACE ELEMENT AND SOME BIOCHEMICAL MARKERS IN INFERTILE MEN

By Hussein Ali Nayyef NAYYEF

August 2022

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ABSTRACT

INVESTIGATION OF THE EFFECT OF PHYTIC ACID AND ITS RELATIONSHIP WITH INHIBIN B, TRACE ELEMENT AND SOME BIOCHEMICAL MARKERS IN INFERTILE MEN

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Doctor of Philosophy in Chemistry

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August 2022

Fertility is closely associated with the goodl desire of humans to live, reproduce, and prolong their existence. Objective: To measure the concentration of phytic acid and to study the effect of changes in its concentration on inhibin B and trace elements and some biochemical markers in infertile men, to assess spermatogenesis and semen parameters. There were high levels of phytic acid in men who had problems in the process of spermatogenesis, and thus its direct relationship to male infertility. That the presence of phytates in human food sources can form complexes in the metabolic stages of humans mainly insoluble with cations such as Ca, Mg and Zn, thus limiting or decreasing the bioavailability, these minerals are important in the formation of sperm. Men with high levels of phytic acid can experience changes in concenteration of inhibin B, LH, and FSH and thus directly affect the function of the male reproductive organs, which is evident in the third level of infertile men in our study. As for the low levels of inhibin B and LH, we found that it affects sperm motility and semen volume and thus limits sperm motility in males, which leads to incomplete fertilization. The study concluded that phytic acid can affect the metabolism of trace minerals, inhibin B hormone, LH and FSH, and thus lead to infertility or the development it or increase inability in treatment it. Because food is mainly based on plant products that contain phytate. There is a need for more studies on the effect of phytic acid on humans.

2022, 113 pages

Keywords: Inhibin B, Phytic acid, Zinc, Spermatogenesis, Sperm

ÖZET

KISIR ERKEKLERDE FİTİK ASİTİN ETKİSİ VE İNHİBİN B, ESER ELEMENT VE BAZI BİYOKİMYASAL İŞARETLERLE İLİŞKİSİNİN İNCELENMESİ

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Doğurganlık, insanın yaşama, üreme ve varlığını sürdürme arzusuyla yakından ilişkilidir. Amaç: İnfertil erkeklerde fitik asit konsantrasyonunu ölçmek ve konsantrasyonundaki değişikliklerin inhibin B ve eser elementler ve bazı biyokimyasal belirteçler üzerindeki etkisini incelemek, spermatogenez ve semen parametrelerini değerlendirmektedir. Spermatogenez sürecinde sorun yaşayan erkeklerde yüksek düzeyde fitik asit ve dolayısıyla erkek kısırlığı ile doğrudan ilişkisi varmıştır. İnsan besin kaynaklarında fitatların varlığının, insanların metabolik evrelerinde, esas olarak Ca, Mg ve Zn gibi katyonlarla çözünmeyen kompleksler oluşturabilmesi, bu nedenle biyoyararlanımı sınırlandırması veya azaltması nedeniyle, bu mineraller sperm oluşumunda önemLidir. Fitik asit düzeyi yüksek olan erkekler, inhibin B, LH ve FSH düzeylerinde değişiklikler yaşayabilir ve bu nedenle çalışmamızda infertil erkeklerin üçüncü düzeyinde belirgin olan erkek üreme organlarının işlevini doğrudan etkiler. Düşük inhibin B ve LH seviyelerine gelince, bunun sperm hareketliliğini ve semen hacmini etkilediğini ve dolayısıyla erkeklerde sperm hareketliliğini sınırladığını ve bunun da eksik döllenmeye yol açtığını bulduk. Çalışma, fitik asidin eser minerallerin, inhibin B hormonunun, LH ve FSH'nin metabolizmasını etkileyebileceği ve dolayısıyla kısırlığa veya gelişmesine yol açabileceği veya tedavide komplikasyonları artırabileceği sonucuna varmıştır. Çünkü gıda esas olarak fitat içeren bitkisel ürünlere dayanmaktadır. Fitik asidin insanlar üzerindeki etkisi konusunda daha fazla çalışmaya ihtiyaç vardır.

2022, 113 sayfa

Anahtar Kelimeler: İnhibin B, Fitik asit, Çinko, Spermatogenez, Sperm

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Hussein Ali Nayyef NAYYEF

Çankırı-2022

CONTENTS

LIST OF SYMBOLS

µL	Microliter
ng	Nonogram
L	Liter
-	Minus
%	Percent
*	Star
/	Divide
℃	Silesian
+	Plus
<	Less-than
=	Equal
Rpm	Round per minute
>	More-than
dL	Deciliter
G	Gram
mg	Milligram
-	minus
Min	Minute
mL	Milliliter
ºC	Degrees celsius

LIST OF ABBREVIATIONS

ANOVA	Analysis of Variance
ATP	Adenosine Triphosphate
BMI	Body mass index
Ca	Calcium
Ca^{2+}	Calcium ion
CE	Commission number
DHEAS	Dehydroepiandrosterone sulfate
ELISA	Enzyme linked immunosorbent assay
FSH	Folicle stimulating hormone
GnRH	Gonadotrophin-releasing hormone
HDL	High density lipoprotein
HRP	Horseradish peroxidase
INHB	Inhibin B
K	Potassium
LDL	Low density lipoprotein
LH	luteinizing hormone
MedCalc	Medical calculate
Mg	Magnesium
Mg^{2+}	Magnesium ion
MI	Myo-inositol
MIP6	Myoinositol hexaphosphoric acid
OD	Optical density
P	Phosphorus
PA	Phytic acid
P-B	Phosphorus-binding
PCOS	Polycystic ovarian syndrome
PH	potential of hydrogen
ROC	Receiver operating characteristic
SD	Standerd deviation
Sig.	Significant
SPSS	Statistical Package for Social Sciences
Tc	Total cholesterol
Testeo	Testosterone
Tg	Triglyceride
TMP	substrate solution
VLDL	Very low density lipoprotein
WHO	World human organization
Zn	Zinc

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1. INTRODUCTION

Fertility is closely associated with the goodl desire of humans to live, reproduce, and prolong their existence. The desire for human beings to reproduce has been noticed since the first tales or myths about the world and mankind were written down in history. Women have been associated with fertility from ancient times, and this has been the case for the majority othe time (Hasanpoor-Azghdy *et al.* 2014). Having trouble getting pregnant is a sign of infertility (conceive) (Loftus and Andriot 2012). Present, As a result, it is difficult to compare infertility rates between countries or over time. In most cases, The inability to conceive is what is meant by the term "infertility" (or to anticipate a child) after one year (or more) of unprotected intercourse. So that women's fertility is known to be decreasing significantly as they get older (Gurunath *et al.* 2011). Direct ofoof on age patterns of infecund and infertility cannot get from contemporary populations because such large of couples use contraception (Menken 1986). An assessment of a woman for infertility is appropriate for women who have not become pregnant after having twelve months of regular, unprotected intercourse (Luciano *et al.* 2013).

Male factors account for 22% of infertility in developed countries but have a role in reproductive issues in both sexes in underdeveloped nations. Laboratory tests that might result in a diagnosis of infertility in more than half of all couples who check out are being investigated (Williams *et al*. 2003). In the laboratory diagnosis some tests asuch s an anti-sperm antibody test, body mass index; DHEAS (dehydroepiandrosterone sulfate), follicle-stimulating hormoluteinizingsing hormone, steroid hormone-binding globulin, thyroid function test; used as a marker for diagnosis of infertility according to World Health Organisation (Williams *et al.* 2003).

Zinc is an essential mineral for men's sexual health, and medical reports show that a deficiency of this mineral is associated with decreased sperm production. Women's fertility declines as they get older, which is well known testosterone and prolactin, for example, and it is also a component of prostate fluids (Fallah *et al.* 2018).

In foods like cereal and pulses, you'll find the anti-nutrient phytic acid (PA). The mineral bioavailability is decreased, and the starch-digesting amylase enzyme, which requires calcium, is inhibited in humans (Thavarajah 2010).

In this study, we will measure study the effect of alterations in the concentration of phytic acid on inhibin B and trace elements and some biochemical markers in infertile men, to assess spermatogenesis and semen parameters. After performing the statistical analysis of the results, it looks at the phytic acid levels. We will compare this with the rest of the chemical tests and examine the effect of these test results on the rest of the test, and thus its effect on male infertility. We used the following steps to achieve the objectives of this study: After taking the original approvals from the hospital administration, Following the completion of the study's target population, blood samples were drawn from those who had the disease under study conditions, and a questionnaire was created. Clinical diagnosis by the specialist doctor. Chemical tests were carried out: Inhibin B, Zinc, phytic acid, FSH, LH, Testosterone, Ca +, Mg and Phosphorous to investigate the study objective.

2. LITERATURE REVIEW

2.1 Infertility

Infertility in men or woman (inability to conceive children) is a usual problem, affecting perhaps one couple in six (Irvine 1998). Infertility is one of the most important medical problems. To have infertility, one must be unable to conceive and carry a pregnancy to term. The inability of the man and the woman to conceive children, and this condition is also said of lack of fertility or the presence of a married couple who are not able to have a child even though they are not sterile, or it is the absence of pregnancy despite the regularity of the marital relationship, there is also Absolute infertility, which is the inability to get pregnant at all, is one type of infertility. The causes are not treatable, one of which is the failure to form the uterus or the ovaries, and the second is relative infertility, and it means that there are obstacles to pregnancy that can be treated, and the relative infertility is in turn divided into two types, which are the primary and the second Delayed childbearing despite the occurrence of a previous pregnancy, regardless of the mechanism of pregnancy (Dunson *et al.* 2004).

Male infertility problems often result from problems in the process of sperm production. Sperm production is the primary cause of male infertility. Infertility in the men, can divided it into three types depends on the number of sperms. It is necessary to analyse the underlying causes of male infertility. Problems with sperm reaching and fertilizing the egg, or the inability of the sperm to move properly due to defects in its shape or structure. A man may be born with problems that affect his sperm, and at other times these problems begin during his life due to illness or injury. Male infertility may be caused by lifestyle, environment, or other health-related factors including age and stress, exposure to high temperatures in the testicles that affect sperm movement and fertilization (de Kretser 1997, Poongothai *et al.* 2009).

2.1.1 Type of infertility

World Health Organization data on the prevalence and types of infertility, infertility 3ertili into two types of infertility. Couples who have not had sex for at least a year

without using birth control methods are considered to have primary infertility and secondary infertility respectively. This type of infertility affects only couples who have tried to get pregnant at least once before but have been unsuccessful. Secondary infertility can be brought on by pregnancy waste and vaginal delivery (Schrijvers *et al.* 1991).

2.1.2 Etiology and caused of infertility

Infertility in humans can be caused by a variety of factors, both physical and emotional. It could be a result of a woman's or man's inability to conceive. Infertility in humans can be caused by a variety of factors, both physical and emotional. It could be a result of a woman's or man's inability to conceive. (Dohle *et al.* 2005).

2.1.3 The male reproductive system

The male reproductive organs are mostly situated in the external genitalia. The penis, scrotum, and testicles are the exterior organs in question (Creasy Chapin 2013). The vas deferens, prostate, and urethra are all examples of internal organs. In addition to urine, the male reproductive system is in charge of sexual activity. A collection of organs responsible for urination and reproduction in men is known as the male reproductive system (Emanuele and Emanuele 2001, Rosen *et al.* 2005). It performs the following functions in the body:

- Sperm (male reproductive cells) and seminal fluid production, storage, and transfer (a protective fluid around the sperm),
- The elimination of sperm by means of the female reproductive system,
- Male sexual hormones are produced and secreted (Emanuele and Emanuele 2001).

Hormones are molecules that stimulate or control the operation of cells or organs, and they are essential to the whole male reproductive system. Follicle-stimulating hormone (FSH), luteinizing hormone (LH), and testosterone are the primary hormones involved in the maintenance of a healthy male reproductive system (Meriggiola *et al.* 1996).

The pituitary gland, situated near the base of the brain, secretes FSH and LH and regulates a wide range of physiological processes. An inability to produce sperm (azoospermia) is caused by a lack of follicle-stimulating hormone (Oduwole *et al.* 2018). Luteinizing hormone is essential for maintaining sperm production because it boosts testosterone synthesis. Muscle and strength development, lipid distribution, bone growth, and sexual drive are all aspects of male maturation that benefit from adequate testosterone levels. The male reproductive organs are mostly situated outside the pelvic and abdominal cavities. Penis, scrotum, and testicles are the exterior male reproductive organs (Santi *et al.* 2020, Oduwole *et al.* 2018).

The sperm is created, stored, and transported by the male reproductive system. The testes are the organs responsible for sperm production (Bernardino *et al.* 2019). These sperm then bind with fluid produced by the prostate gland and seminal vesicles; it's important to note that while a man is born with a fully developed male reproductive system, it doesn't start to operate until adolescence (Silber *et al.* 1995). What follows is a breakdown of the inside components of a man's reproductive system:

- Urine is expelled from the body through the urethra, which also serves as the exit point for sperm.
- The epididymis is the long, coiled tube at the rear of each testicle that is responsible for transporting, storing, and maturing the sperm that are created in the testicles.
- Urine and sperm are expelled from the body through the vas deferens. It is a lengthy tube that begins at the epididymis and extends into the pelvis and the bladder's urethra.
- The ejaculatory ducts are the remnants of the vas deferens and seminal vesicles joining together.
- The glands of the bulbo-urethra: The thick fluid produced by these glands is excreted through the urethra. They sit directly below the prostate glands, on each side of the urethra.
- The prostate gland, which aids in ejaculation and nourishes sperm, is situated underneath the urinary bladder and in front of the rectum.
- The vesicles are where the sugar fructose is made, which gives the sperm their energy to swim (Castilho *et al.* 2008, Ruiz *et al.* 2020).

2.1.4 Men's infertility

When a guy is infertile, it means that he is unable to bring about a pregnancy in a fertile female partner. In humans, it is responsible for 40–50 percent of all cases of infertility. It affects around 7% of all males in the population. Man's infertility is often caused by deficits in the characteristics of the sperm (Figure 0.0), as well as semen quality being substituted for male 6ertility in many situations (de Kretser 1997).

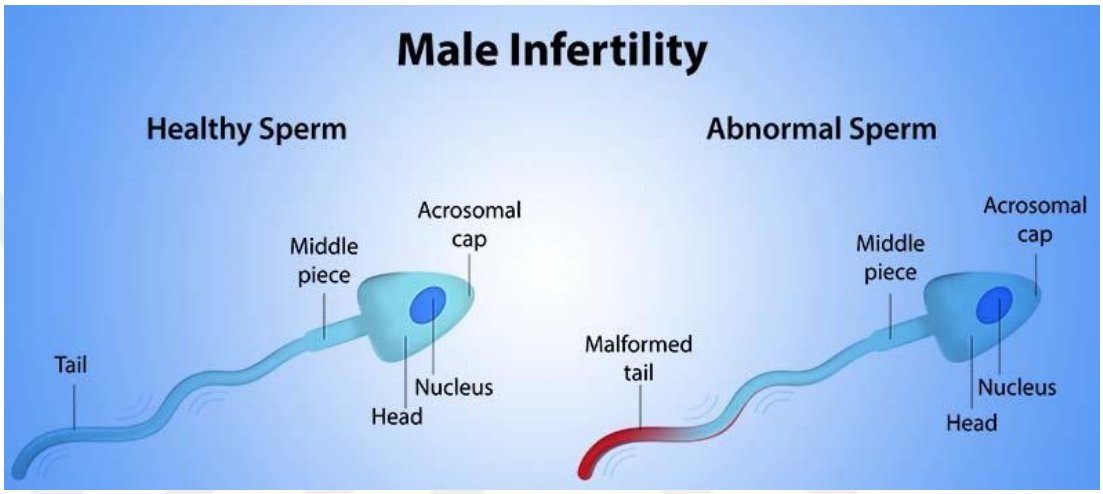

Figure 2.1 Showen the form of sperim in in human male (Life 2021)

2.1.5 Male fertilization

When female infertility occurs, it is possible that an egg that has been fertilized will not survive once it has been attached to the uterine lining (uterus). An egg that has been fertilized will not adhere to the uterine lining. Embryos are unable to move from ovaries to wombs because of this. For example, the ovaries are having trouble ovulating because of this. When the egg and spermatozoon of a mother and a father join together to form a fertilized egg, the process of fetal development begin. The oviduct is the place where mammalian oocytes and spermatozoa meet, a sequence of events takes place that eventually result in the fertilization of the oocytes and the birth of a new person. Fertilization triggers a series of crucial processes that culminate in the formation of the zygote (a fertilized egg) (Neri *et al.* 2014). The typical sperm concentration ranges from 40 million, a milliliter of semen contains up to 300 million sperm A drop sperm count is defined as anything between 10 and 20 million sperm per milliliter of

sperm-containing fluid. If the sperm are in good health, twenty million (10) sperm per milliliter may be sufficient for a successful pregnancy (Diniz *et al.* 2005).

Following cumulus perforation, to fertilize an ovulated egg, free-swimming spermatozoa must first locate and adhere to the zona pellucida. This extracellular coat must be penetrated and fused with by the sperm cell in order for it to activate the ootid. In order for the sperm to penetrate the extracellular coat, the acrosome reaction must be completed. According to current theories, ion channel modulation, receptor-ligand interactions, and many more, membrane fusions, and proteolysis gs. In addition, autoimmune illnesses such as antiphospholipid syndrome, which affect the lining of the uterus, may induce female infertility (APS), Birth abnormalities that affect the reproductive tract are classified as follows: Whether it's cancer or a tumor, Disorders of clotting, Diabetes, drinking an excessive number of alcoholic beverages When you overexert yourself, you risk injury. Eating problems or malnutrition are among the most common. Fibroids and polyps in the uterus and cervix are examples of growths in the reproductive system. Medications such as chemotherapy medications, for example. Inconsistencies in hormone levels, being overweight or underweight are both problematic. Ovarian cysts and polycystic ovary syndrome are two conditions that affect the ovaries (PCOS). A decrease in sperm count may be to blame for male infertility, a blockage that stops the sperm from being released, or flaws in the sperm (Yan 2009). Detailed explanation of male infertility will be provided in the following parts.

2.2 Spermatogenesis

Spermatogenesis is a multifaceted process that occurs in the testicles that results in the production of large quantities of normal spermatozoa (Sperm) via a process known as spermatogenesis (Figure 2.1). The process of spermatogenesis may be broken down into three primary phases, which are as follows: The first is the process of mitosis, in which spermatogonia replicate; the second is meiosis, in which spermatogonial type B cells enter the prophase of the first meiotic division and the number of chromosomes is decreased from diploid to haploid; and the third is ovulation. Now called primordial spermatocytes, these cells eventually become the progenitors of sperm via a process of

cell division. In the third stage, which is known as spermiogenesis, the round spermatid successfully transitions into the complex structure of the spermatozoon after being divided once more to produce round spermatids (the process of creating spermatozoa) (de Kretser *et al.* 1998).

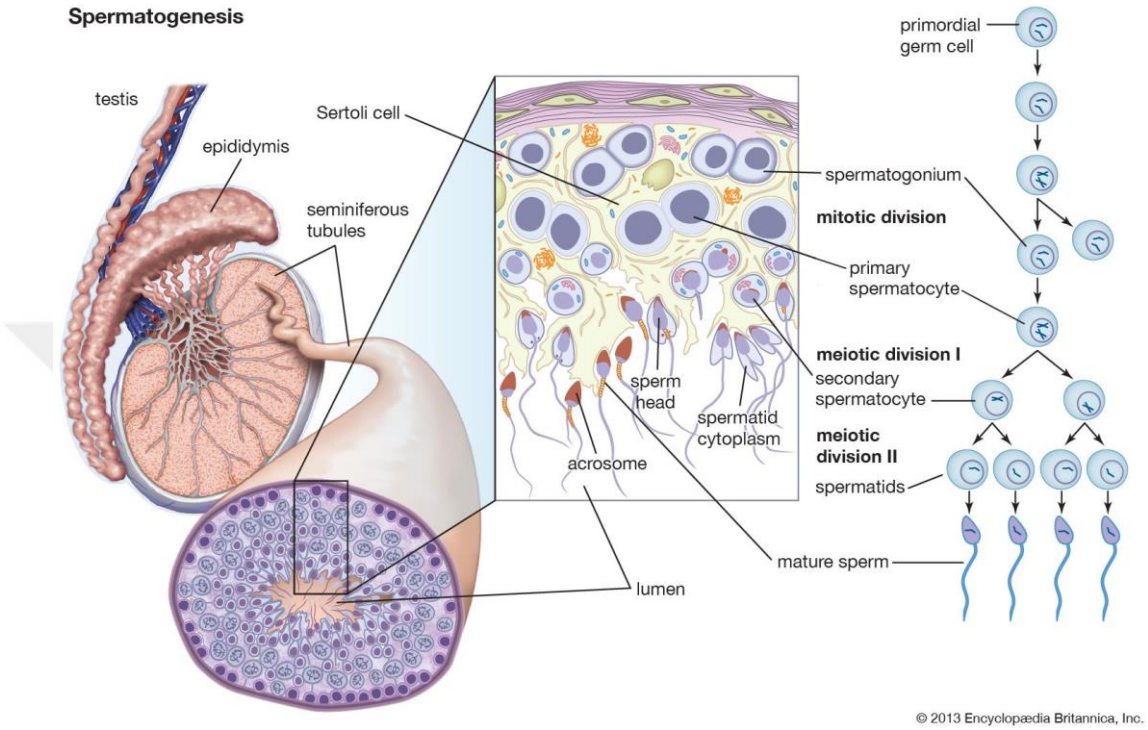

Figure 2.2 spermiogenesis (de Kretser *et al.* 1998)

The development of sperm may be defined as the origin and development of sperm cells inside the male reproductive organs, namely the testicles, according to one definition. It is also possible to define spermatogenesis as the process by the testicles produce what kind of sperm cells (the seminiferous tubules of the body). Seminiferous tubules of the testicles transport mature sperm to the epididymis, where it remains until it can be expelled from the male reproductive system (Healy 1989).

2.2.1 Human testis and hormonal control of spermatogenesis

Follicle stimulating hormone (FSH) and testosterone hormone (TTH) release are necessary for completion of spermatogenic processes. There are in-between stages in the activities of follicle stimulating hormone (FSH) and testosterone hormone on germ

cells due to the lack of receptors on these cells. It is now known what these intermediary processes are like. It is believed that FSH and TTH govern the activity of the Sertoli cell, which in turn affects the activity of the follicles, and hence exercise their impact on germ cells. Synergistic effects on germ cells are shown when follicle stimulating hormone (FSH) and testosterone hormone (TTH) are present, with TTH having a pronounced influence on spermatid maturation towards the end of the process. It is possible that FSH, by its potential to induce Sertoli cell mitosis during testicular development, will have an effect on the spermatogenic capability of the adult testis (McLachlan *et al.* 1995). Both rats and men (humans) have been shown to have a much higher concentration of testosterone in their testicles than in their bloodstreams. Rat spermatogenic regulation has been studied extensively in recent years in an effort to better understand how hormones influence this process. This research has more precisely described the phases at which the hormones testosterone and follicle stimulating hormone (FSH) function, and it has become clear from these findings that developing models to assess FSH insufficiency is challenging. In patients with insufficient gonadotrophin-releasing hormone (GnRH), several complications may arise as a result of the ability of high testosterone dosages to raise FSH levels (Jarow and Zirkin 2005).

2.2.2 Semen

About 0.2 mL of sperm cell and seminal plasma is produced over a period (Katigbak *et al.* 2019). In humans and animals alike, the fluid that is discharged from the male reproductive system is called sperm fluid or seminal fluid because it contains sperm cells that may fertilize the eggs of the female. Besides liquids, sperm (sperm cell) also includes liquids that mix to create seminal plasma, which serves to maintain the viability of the sperm cells (Figure 2.3) (Ax 2000).

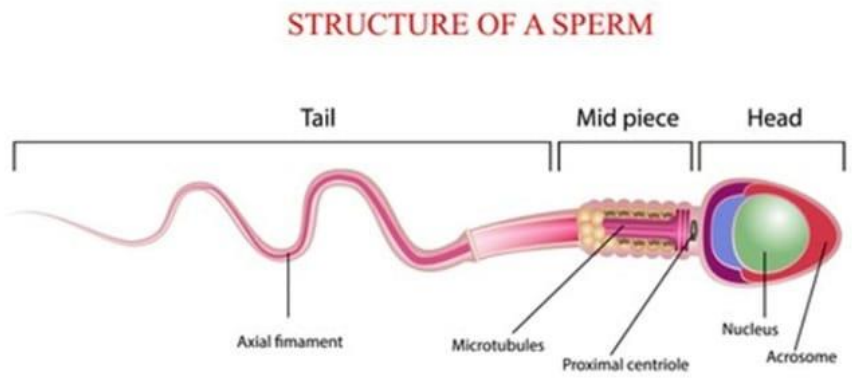

Figure 2.3 Human sperm cells (Mann 2012)

Semen is produced by combining a concentrated suspension of spermatozoa, which is kept in two epididymis, with fluid secretions from the accessory genitals and diluting the mixture. Several boluses of sperm are given at a time. A comparison of semen quantities before and after vasectomy reveals that accessory gland, namely the prostate and seminal sacs, account for around 90 percent of the total semen volume, with minor amounts of secretions from the bulbourethral glands (Cowper's gland) and epididymis (Cooper *et al.* 2010).

2.2.2.1 The biochemistry of semen

Semen (sperm cell) has been the topic of intensive nowadays, not only because of the quick growth of artificial insemination throughout the world, but also because spermatozoa are excellent material for research by the (chemistry) biochemist and the cell physiologist. Studies dealing with semen, spermatozoa and the medium in which they live, biochemical tests seminal plasma, are widely scattered in the scientific literature, in veterinary journals and those concerned with “pure" biochemistry and physiology for treatments infertility in both women and males (Kehres and Maguire 2003, Attia *et al.* 2020).

After masturbation, it is possible to analyze the semen that has been collected. Both the seminal vesicules and the prostatis secretion (which contains acids phosphates, magnesium, zinc, and citric acid) are sources of components used in sperm production

(fructose). Researchers are trying to figure out what makes up seminal plasma by looking at proteins, prostglandins, amoniacids, and sex hormones. The latter are especially interesting since oligospermia is characterized by lower amounts of estradiol and progesterone. There are a variety of seminal plasma biochemical tests used to identify male infertility, and the results might vary widely. When the semenal plasma is collected by masturbation, it may be tested. Prostatial secretion contributes to sperm production by providing acid phosphates, magnesium, zinc, and citric acid, in addition to seminal vesicles (fructose). Seminal plasma is now being studied to determine the amounts of proteins, prostglandines, amoniacide, and sperm hormones. Due to low levels of estradiol and progesterone, the latter is especially relevant when discussing oligospermia. Male infertility in seminal plasma can be diagnosed using a variety of biochemical assays, each with a unique outcome (Katigbak *et al.* 2019).

2.1.5.1 Factors affecting male infertility

After two years of regular, frequent intercourse and the absence of any known reproductive disorder, a couple's sterility may be established. However, infertility refers to a more lasting problem with reproduction. There has been a worldwide increase in the difficulties of dealing with societal problems such as the trend toward later marriage during the last thirty years (Mahat *et al.* 2016). There were many things that affect the process of childbearing, especially the problems that men suffer from, we will talk more clearly within our specialization in clinical chemistry, in which we are looking for an accurate and early diagnosis of infertility and solving its problems through specialized doctors.

2.2.3 The effect of the mineral on infertility

Trace elements have a variety of roles in the body, including serving as cofactors, enzyme activators, and stabilizers of secondary molecular structure. The studies have evolved for a recognition of the trace elements and their essential function in cell metabolism. The effects of dietary deficiencies (of trace elements) on physiological functions in general and reproduction in particular have attracted a lot of attention (Hidiroglou 1979). One of the factors that contribute to male infertility is a lack of or

low sperm quality. In order to establish the type of sperm, three basic end-points are often used: the number of sperm, the sperm's shape, and the activity of the sperm. Male factor infertility and micronutrient therapy options are only addressed in a few studies, researchers say, despite the wide availability of micronutrient supplementation products claiming to improve the spirogram (Buhling 2019).

2.2.3.1 The effect of zinc on infertility in men

An abundant microelement in the human body, zinc may be found in a variety of foods such as nuts, legumes, shellfish and fortified cereals. Mushrooms, low-fat yoghurt, and animal proteins like meat, fish, and dairy products all contain it. The intake of these natural foods has been shown to increase the proliferation of germinal cells and a deficiency in Zn nutrition may be a major risk factor for poor sperm quality (activity, count and morphology), as well as male infertility due to unknown causes (Colagar *et al.* 2009).

This may be caused by a variety of risk factors, included in this list are varicocele and obstructive lesions, cystic fibrosis and cryptorchidism as well as environmental factors like trauma and genitourinary infection and dietary deficiencies in trace elements like zinc and selenium. Incidences Approximately 30 to 55 percent of all infertility occurrences in couples are due to male infertility. Unexplained male infertility affects a large proportion of these sub-fertile individuals, who are categorized as such. The significance of trace elements in the preservation of the quality of human sperm is a topic that is attracting a lot of attention these days. Although it has been shown that trace elements have a function in male human fertility, the biological significance of these components has not yet been extensively explored. For example, Zinc (Zn) is a trace element that can be found in high concentration in mammalian sperm and plays an important role in male human spermatogenesis among other things (Kumar and Singh 2016).

2.2.3.2 The effect of calcium on infertility in men

Among the principal processes through which calcium shortage causes male infertility are impaired sperm motility, impaired chemotaxis, capacitation, acrosome response, and steroidogenesis, to name a few. To improve sperm activity and all of the processes leading to successful fertilization, an appropriate seminal Calcium content is necessary (Harchegani *et al.* 2019). There has been some research done on the influence of seminal calcium on male infertility, and the results have been reported. Seminal TCa concentrations in fertile and infertile males were nearly identical, according to the results of this study (quality of human semenRegardless of spermatozoa motility, the total calcium concentration in the seminal fluid was not statistically different; however, the TCa concentration in the semen of men with hypomotility was significantly lower when compared to the TCa concentration in the semen of men with normal motility (Logoglu *et al.* 1997).

2.2.3.3 The effect of phosphorus and magnesium on infertility in men.

As phosphate transfer enzymes use magnesium as an activator ion, it has been defined as "essential for the survival of all living things." In reality, magnesium plays a role in a number of biological processes, with serum containing the highest concentrations. In the myoneural system, magnesium dioxide (Mg^{2+}) has an immediate effect. It is estimated that about 50 percent of the body's magnesium is found in bone. Only 5 percent is found outside the cells, and the remaining 45 percent is found inside the cells. There are two-thirds of the cation free and one-third protein bound in normal plasma concentrations of 1.5–2.5 meq/L. Magnesium ions are excreted through the kidneys (Omu *et al.* 2001).

2.3 General Information of Phytate Acid

Phytic acid or phytates is «inositol hexakisphosphate», and it is found in all legumes in the form of a mono or divalent salt, it was discovered in 1903 and stored mainly in the layer of Aleuron and is considered Phytic acid is the source of this information. The phosphorus in seeds accounts for around 80% of the total phosphorus in seeds, making them a rich source of both phosphorus and inositol. The plant (vegetable) component in

human diet is substantial, making up between 1% and 5% of the weight of the legumes, grains, oil seeds, and nuts that are the principal sources of human and animal sustenance. There is a considerable variation in the amounts of the phytonutrient phytate found in wheat, nuts, legumes, and seeds, and this variation is likely to be influenced by variables such as the food's growth circumstances and harvesting procedures, processing processes, testing methods, and even the food's age (Saburov et al. 1989, Nissar et al. 2017). Phytic acid (PA) is the principal form of phosphorus storage in many seeds; its chemical name is myoinositol hexaphosphoric acid (IP6). C6H18O24P6 is its chemical formula, and its molecular weight is 660.03 grams. 660.03 grams is its molar mass. The graphic depicts the many ways in which phytic acid may interact with different molecules, including metal cations and protein residues. There are snow-flake-like molecules in it that contain mineral phosphorus securely bonded (Saburov *et al.* 1989, Urbano *et al.* 2000). It also contributes to the preservation of low inorganic P levels in the endosperm, which aids in the promotion of starch production. During germination, these phytates are digested, releasing phosphorus and minerals that may be used by the plant. Unwanted traits of several low-phytate mutants have been observed, stress sensitivity, lower germination rates, and lower yields, among others (Bregitzer *et al.* 2006, Dong *et al.* 2020).

2.3.1 The chemical structure of phytic acid

The terms phytic acid (PA) and phytate are frequently used interchangeably in scientific literature. Phosphate (IP6), the anion "phytate," and the proton donor (H+) "phytic acid" (IP6) are all terms used to describe the same chemical compound in the context of water. This IP6, which has six phosphate groups encircling one inositol ring, is shown in detail in its chemical structure (Figure 2.4) (Angel *et al.* 2000).

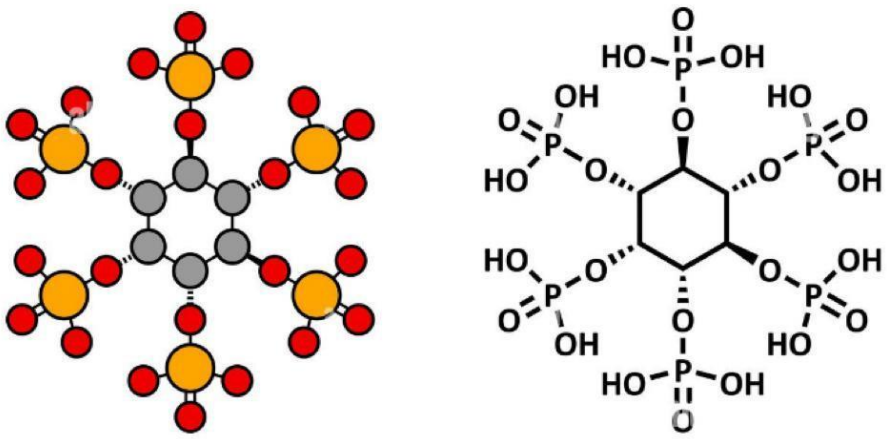

Figure 2.4 Phytic acid structure (Alamy Stock Photo 2022)

With two dissociable OH groups present in each phosphate, IP6 has a total of twelve protons or reactive sites that can be replaced. A pKa of 3 is considered strongly acidic, and two are moderately acidic (pKa about 5 to 6), and the other four of which are very weakly acidic (pKa > 9) As a consequence, totally protonated IP6 can only be found at very low pH values (pH 1.3 or below). As the pH of the solution increases to roughly 10.5, the phosphate groups may be deprotonated sequentially. With this knowledge, it is possible to conclude that the acid-base characteristics of IP6 are influenced by factors other than pH. Furthermore, Factors such as ionic strength have a significant impact on the pKa values of IP6, supporting electrolytes, kind of cations, and other variables. Deprotonation (decreased pKa) is often facilitated by increasing ionic strength, however certain organic salts are suppressive of it (increased pKa) (Rodrigues-Filho *et al.* 2005; Crea *et al.* 2008).

2.3.2 The effect of phytate on humans

Mineral elements are present in food in several chemical forms, and they include their salts with organic acids to form complex compounds with organic origins. Proteins, peptides, amino acids, flavones, hydroxylated carboxylic acids and derivatives of polysaccharides are important components ions that have the ability to interact with them. In most cases, the physiological consequences of the effects of these associations are harmful or unknown. There are two main types of organic substances that reduce the vital utilization of the basic nutrients and trace elements in food, phytic acid and oxalic

acid. We intend to investigate the role of phytic acid in our study (Sprague *et al.* 2012, Kim *et al.* 2009).

Most plant foods contain phytic acid, especially food grains where there is an appropriate concentration of phytate salts in the rice bran, which may reach between 9.5 - 14%. Peas and soybeans of all kinds also contain it, and it is also found in oilseeds such as sesame seeds and radish. Phytic acid may be considered an unwanted component of food that can be completely or partially eliminated during food processing processes, where thermal treatments lead to partial destruction of phytic acid in treated foods (Sprague *et al.* 2012, Kim *et al.* 2009), for example:

Roasting coffee seeds when preparing coffee leads to phytate content has dropped significantly and the heat treatment of soybean flour when removing fat leads to a loss of about 23% of phytate, as if heating in the microwave for 15 minutes leads to a loss of 46% of the phytate in the soybeans removed. The anointing. Likewise, cooking the rice in water greatly reduces phytate content (Liu *et al.* 2018; Schlemmer *et al.* 2009)

Phytic acid catabolism may also occur during food processing and storage because of the enzymatic decomposition The enzyme phytase breaks down phytates, which is common in plant materials, especially in seeds and grains. Phytic acid is also present in legumes in a water-soluble form, so soaking the seeds before cooking them reduces their phytic acid content. By breaking down and analyzing this compound during germination. The enzymes in the yeast eliminate most of the phytic acid during the baking process, and the heat destroys most of the phytic acid found in bread and breakfast cereals rich in the bran. Thus, these processed foods are safer than fresh bran (Sprague *et al.* 2012, Ma *et al.* 2007).

Phytic acid. It is a very important acid for the body to function properly, as the elements of calcium and magnesium mix to form phytin salts in addition to organic phosphates, as they form the basis of the energy required for the body to carry out the vital processes that it carries out at every moment (Sprague *et al.* 2012, Gillespie and Van den Bold 2017**)**.

Phytic acid is an important compound that works as an antioxidant in the body. From a dietary perspective, a diet rich in phytic acid helps to keep kidney and gallbladder stones at bay, as well as a variety of cancers and alcohol-related poisonings, all of which can be devastating. It also improves the taste of meat and fish when eaten (Liu *et al.* 2018, Schlemmer *et al.* 2009).

Phytic acid also works to reduce the metals accumulated in the body, or what is known as chelation, by which work is done to get rid of these harmful minerals, for instance: when iron and phytic acid are mixed together and calcium in excess of the body's need to form phytate salts and thus help the body get rid of them Which protects the body from harm (Sprague *et al.* 2012, Ma *et al.* 2007).

2.3.3 Damage phytic acid

It is considered as one of the anti-nutrients with its ability to chew or hold or reduce the ability to absorb some important metals and elements such as ferrous and nonferrous metals (such as zinc and copper) and phosphorous, in addition to forming complexes with protein. The binding of phytic to a protein depends on the pH and dichotomous cations. In the acid medium, the protein molecule bears a positive charge and the phytic charge is negative, which allows the formation of a complex of them. The increase in the double cations, especially calcium, leads to its entry as a competitor to the protein in the complex, Phytic acid binds to proteins, reducing the likelihood of proteolytic enzymes interacting with the protein. The work of the proteolytic enzymes may thus be inhibited because the enzyme is proteins and therefore it is also exposed to binding to phytic acid, which affects its activity. And his motivational action. It should be noted that calcium phytate and magnesium phytate have the ability to chelate zinc, iron and copper, which reduces the availability of these elements and the body's use of them (Sprague *et al.* 2012, Ma *et al.* 2007).

It has been confirmed to nutritionists that adding one gram of wheat bran powder to each person's food at every meal and to foods that are described as difficult to digest or that do not give a good nutritional benefit as this simple amount gives very useful

results for treating indigestion and colic and can be eaten for adults and children on Both (Xu *et al.* 1992, Harinarayan *et al*. 2021**).**

The bran also strengthens the nerves, the reproductive system and bones, modifies the function of the thyroid gland, stimulates digestive juices, saves the body from many diseases, and makes it vital and active. It also prevents diverticulitis, an intestinal disease characterized by the formation of inflamed sacs protruding from the walls of the colon. Just as the bran prevents constipation, it is very useful for people who suffer from hemorrhoids (Sprague *et al.* 2012, Kim *et al.* 2009).

Food containing fewer wheat grains also helped reduce cholesterol levels in the blood, and it also seemed to improve glucose metabolism in people with diabetes, and this would reduce insulin and other glucose-lowering drugs. Recent studies also indicated that the use of rice bran in meals also helps to reduce cholesterol levels in the blood, but researchers were unable to ascertain whether the reason for this benefit was due to the insoluble fibers present in the bran or the large amounts of unsaturated oils present in the rice endosperm. Which is not separated from the rice husk during the milling process as it is firmLy attached to the bran (Liu *et al.* 2018, Schlemmer *et al.* 2009)

In general, we've found that foods high in fiber are helpful in weight management because they make you feel full without consuming a lot of calories. which explains the reason for the decrease in cancer and heart attacks resulting from obesity in people whose diet contains large amounts of fiber (Oberleas *et al.* 1981, Harinarayan *et al.* 2021).

When people learned about the benefits of wheat bran, many of them deliberately increased its quantity beyond the limit that we mentioned, as a result, it became clear that the rise in IBD could be linked to this, a condition in which the intestine becomes inflamed and has small ulcers. In addition, Phytic acid, found in fresh bran, acts as a preventative measure. The absorption of calcium, iron, zinc, magnesium and other minerals important to the body (Sprague *et al.* 2012, Harinarayan *et al.* 2021**)**.

2.3.4 The role of phytic acid in infertility men

There are a number of infertilety problem and need to reguleted their food. Humans need nutrients for the proper functioning of their cells and for the maintenance of their general health. Nutritional value and quality of food are determined by whether or not the food contributes to preserving health and avoiding illness while also supplying a suitable number of nutrients in its composition. The food matrix also contains additional metabolites (including secondary metabolites that display anti-nutritional qualities), but to a lesser extent, depending on the type of food and the method of preparation used. In humans, it has been demonstrated that the absorption and bioavailability of nutrients are reduced when foods contain anti-nutrients such as phytic acid (Schlemmer *et al.* 2009; Frontela *et al.* 2011).

Since its discovery, the effects of phytic acid on mineral absorption have attracted considerable attention due to its influence on the absorption of specific minerals. PA, on the other hand, has a strong affinity for nearly all of the minerals in the human diet, making it an excellent supplement. As a result, iron, zinc, and calcium absorption and differentiation are hampered, which could lead to a shortage of these essential minerals in humans. This has led to the classification of it as being a human anti-nutrient (Schlemmer *et al.* 2009).

2.4 General Information of Phytase Enzyme

In the presence of phytate, Phosphates are released from the compound by phytases (such as myo-inositol hexakisphosphate phosphohydrolase, EC 3.1.3.8). Many cereal grains, legumes, and oilseeds contain phytate, a phosphorus-storing compound. Botanical, animal, and microbiological sources of phytases include plants, animals, and bacteria. Those derived from microorganisms, on the other hand, most promising for commercial and biotechnological purposes. In the field of animal nutrition, this enzyme is commonly utilized to both boost phosphorus feeding while simultaneously decreasing phosphorus pollution generated by animal wastes (Vats *et al.* 2005).

Phytolases are used in the food industry, the production of myo-inositol phosphates, the paper industry, and the improvement of soil. New phytases with better properties are being made using biotechnology more often, and this is becoming more common. Phytase enzyme uses a wide spectrum in human and animal nutrition. In 1991, the first commercial product from phytase enzyme came to the market. At the same time (around 150 Mio Euro) ten amount is in the market. 30% phosphorus is stored in the root of the plants and seeds and grains are stored from 80% (Table 1). Phytic acid (Figure 2.5) is the gelatina factor in complexes accompanied by many binary ketones, it is important from essential food ingredients, for example (Ca^{2+}, Mg^{2+}, Zn2 +, Cu2 +, Fe2 +, Mu2 +) (Haefner *et al.* 2005, Vats *et al.* 2005).

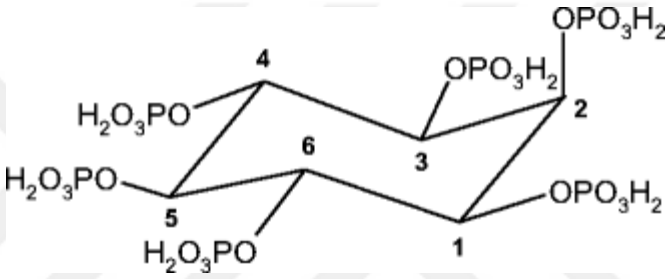

Figure 2.5 Structural formula of phytic acid

In addition, proteins and complexes with amino acids, their pH, are required in the case of acid and base states (Selle *et al.* 2006).

Table 2.1 The phytate content of grains and roots (Ravindran *et al.* 1995)

	Phytate P [g/100 g dry matter]	Phytate P [% of total P]
Cereals		
Corn	0.24	72
Wheat	0.27	69
Barley	0.27	64
Oats	0.29	67
Sorghum	0.24	66
Rice, unpolished	0.27	77
Roots and tubers		
Cassava	0.04	28
Sweet potato	0.05	24

In the last two decades, phytases have proven to be a remarkable enzyme in the fields of human health, nutrition, and environmental protection. Myo-inositol phosphates, myo-inositol and inorganic phosphates are produced by phytases, enzymes that catalyze the

hydrolysis of phytate, the primary phosphate storage form in plant seeds (Figure 2.6). Phytases are a subclass of phosphotases that use phosphohistidine intermediates in the phosphoryl transfer reaction and are typically found in the histidine acid phosphatase family (Mitchell *et al.* 1997).

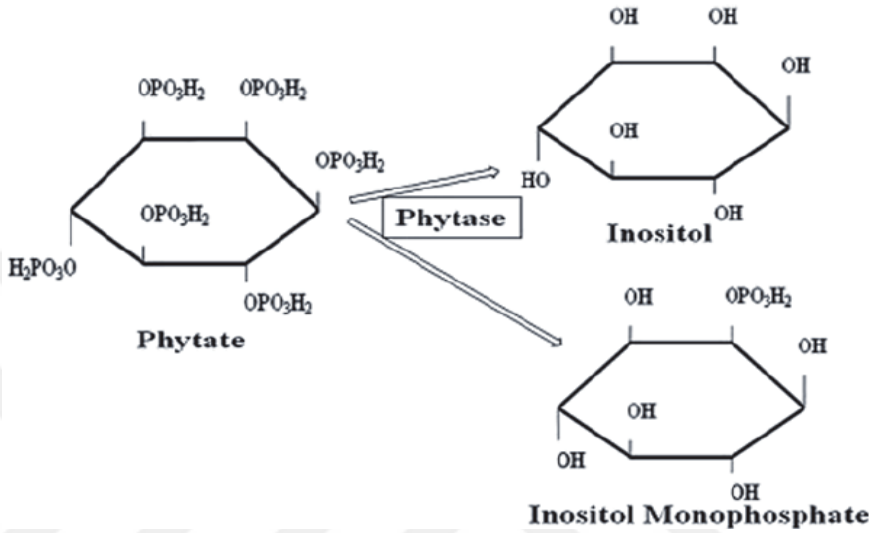

Figure 2.6 Phytic acid hydrolysis by phytase enzyme (Kumar *et al.* 2015)

2.4.1 Temperature and PH of phytases

Phytases are generally active at temperatures in the 45-60 oC range and are thermostable in a wide pH range. Phytases have optimum pH ranging from 2.2-8.0. Most of the microbial phytases have optimum pH between 4.5-5.6. There are some Bacillus strain-specific phytases that have an ideal pH range of 6.5 to 7.5. While the optimum pH of phytase enzymes obtained from plant seeds is in the range of 4.0-7.5, their optimum pH is generally between 4.0 -5.6 (Woyengo *et al.* 2010, Kumar *et al.* 2021).

2.4.2 Enzymatic degradation of phytate

Myo-inositol (1, 2, 3, 4, 5, and 6) hexacyphosphate phosphohydrolases, or phytases, are enzymes that catalyze the gradual release of phosphate from phytate in the presence of

other nutrients, such as amino acids and fatty acids. Debnath and colleagues (Debnath *et al.* 2005) discovered that phytoplankton phytase sequencers have the intositol ring inositol ring, the intermediary chain of myo-inositol phosphates (from pentaphosphate to monophosphate of inositol), and the ability to create free inorganic phosphorus. As well as aiding the release of phosphorus from plant-based foods, phytase also aids in the release of other nutrients like calcium and magnesium as well as protein and lipids. As a result, by secreting phosphorus bound in plant-derived feed materials, phytase increases the amount of phosphorus available for bone formation while simultaneously protecting the environment from phosphorus pollution (Baruah *et al.* 2007). In the early 1900s, rice bran and calf blood were used to detect the presence of phytase activity. Later, it was discovered in plants, bacteria, yeast, and fungus, among other things. Both animals and humans produce their own phytase in the small intestine and the associated microorganisms, making them an excellent source of this enzyme. The endogenous phytase activity of humans and animals is far less than that of plant and microbial phytase. It was in 1991 when Natuphos, the first commercial phytase, was released to the market. Natuphos was derived from the fungus Aspergillus Niger. Following the commercialization of phytase, its acceptability as an animal feed addition in the market has garnered widespread interest across the globe. In addition, the potential to improve human nutrition and other areas, such as aquaculture, is extensively explored (Kumar *et al.* 2010).

2.4.3 Effects of phytic acid on the mineral in humans

Phytate, a dietary compound, impairs the body's ability to absorb and use minerals. Zinc, iron, calcium, magnesium, manganese, and copper are among the metals and elements that fall under this umbrella (Konietzny *et al.* 2003, Persson *et al.* 1998). The bioavailability of Zn^{2+} was shown to be the most harmful of these effects in humans, according to the research. Plant-based meals containing phytate are a significant contributor in lowering Zn^{2+} absorption and Zn^{2+} - homeostasis, which in humans and other animals can lead to dwarfism and hypogonadism. Because of the insoluble phytate mineral complex formed by dietary phytate, the bioavailability of minerals is reduced, according to this theory. Few individuals mean that in the human stomach, phyta-mineral complex is only partially dissolved (Iqbal *et al.* 1994, Lopez *et al.* 2002).

Human studies have also shown that phytate has a significant impact on the absorption of non-haem iron from the intestine (Brune *et al.* 1992). Phytate is also shown to be an absorption inhibitor of Ca^{2+}, however the effect is less dramatic than the reduction in Zn^{2+} and $Fe^{+2/+3}$ absorption seen in people (Lopez *et al.* 2002).

According to Sandström and colleagues (Sandström *et al.* 1990), the colonic bacteria have the capacity to phosphorylate phytate and produce Ca + 2 as a result, which is then taken up by the body. There have only been a few studies on the effects of phytate on the diets of Cu^{+2}, Mn^{+2}, and Mg^{+2} (Lopez *et al.* 2002, Lönnerdal 2002). As well as phytate-cation molar ratio and concentration, the stability and solubility of complexes are determined by the solution pH and various other compounds (Oberleas 1983). Phosphate's solubility can be influenced by the pH of a solution (Cheryan 1980; Cheryan *et al.* 1983); Phytate is less soluble at lower pH values than it is at higher pH values (Torre *et al.* 1991). When pH is less than 4-5, the Ca^{2+}, Cd^{2+}, Zn2 +, and Cu2 + salts tend to dissolve more readily, but Mg-phytate dissolves at acid pH levels up to pH 7.5. (Brown *et al.* 1961, Nolan *et al.* 1987). However, ferric phytate does not dissolve in the pH range 1.0-3.5 when the pH values are the same, Solubility and solubility rise above pH 4 in the range of 1.0-3.5, while phytate ratios increase. Zinc is most likely the mineral that is most sensitive to the phytate complex. Early investigations in chicks, pigs, rats, and other species revealed that dietary phytate had an effect on the amount of zinc required by the animals. In order to avoid chronic permanent deficits, according to Oberleas and Harland 1996, the majority of internally released minerals must be reabsorbed. In their 1996 paper, According to Oberleas and Harland, phytate is a significant factor in the occurrence of zinc deficiency in humans. Because Ca increased the impact of phytate on zinc consumptionBecause of the formation of an insoluble Ca-phytate-Zn complex, it is hypothesized that zinc absorption was limited (Askar *et al.* 1983, Kumar *et al.* 2010).

2.4.4 Effect of phytate on carbohydrates in humans

It has been shown that phytate consumption lowers the rise in blood sugar (glycemic index) (Lee *et al.* 2006). According to some theories, this is due to the fact that phytate forms complexes with the carbohydrates in the feedglucose digestion and absorption are severely affected by this reduction in their solubility. It is possible to bind phytate to

starch either directly, via hydrogen bonds, or indirectly, through proteins that are associated with starch. As a consequence of eating grains and legumes, diabetics may reduce their glucose reaction, which is measured by their low glycemic index. This can help diabetics maintain better control of their blood glucose levels (Yoon *et al.* 1983, Thompson *et al.* 1987).

Minerals, proteins, and carbohydrates can be bound to it because it has the ability to do so at physiological Ph, phytic acid is regarded as an antinutrient by nutritionists. Recent data, while on the contrary, shows that phytic acid may have beneficial benefits. With the addition of endogenous phytate or pure sodium phytate eaten in meals, researchers have seen a decrease in Lower levels of plasma cholesterol and triglyceride in the bloodstream. Aside from that, phytic acid has been shown to have anticancer properties in mouse colon and mammary gland models, in vitro, as well as in a variety of tumor cell lines. A reconsideration of the word "antinutrient," which is used to characterize dietary components such as PA, is necessary in light of these positive benefits (Rickard *et al.* 1997).

After taking out the phytic acid and adding calcium, the starch digestion in vitro improved and the glycemic response in vivo increased, but after adding phytic acid back in, the opposite effect occurred. Respiratory H2 measurement was used to test carbohydrate malabsorption; the results were shown to be inversely linked with the glycemic response; however, the observed alterations were considerably less than those detected in the glycemic response (Thompson *et al.* 1987).

2.4.5 Effect of phytase on lipid in humans

When combined with other nutrients, phytate creates 'lipophytes' (complexes including lipids and derivatives) (Vohra and Satanarayana 2003). Metallic soaps, which are formed in the poultry house lumen by the interaction of lipid and calcium phytate, are a substantial limitation on the amount of energy that can be taken from lipid sources (Leeson 1993).

On the other hand, there is a great deal of evidence that lipid phytate complexes exist in humans. The complexes formed by phytic acid with positively charged food components, such as proteins, carbohydrates, minerals, and trace elements, are responsible for the acid's antinutritional effects. This is evident from what has been said previously (Leeson *et al.* 1993, Vohra 2003).

2.5 General Information of Inhibin B Hormone

The first time the term "inhibin" was used in literature was in 1932, by D. Roy McCullagh, who published a book of the same name. A hormone found in testicular extracts, urine, and blood, he hypothesized, inhibited pituitary hyperplasia and hypertrophy in rats. However, removing the seminiferous tubule walls had the opposite effect, proving his hypothesis wrong (Meachem *et al*. 2001).

An inhibitor of FSH production by the pituitary gland, the pituitary gland's release of FSH is inhibited by inhibin, a dimeric glycoprotein complex. This hormone is generated by the gonads that have been suppressed; it is mostly produced by Sertoli cells in males and granulosa cells in women. Inhibin is essential in the regulation of gametes and embryonic formation, as well as in the development of the fetal embryo. When it comes to Inhibin in the blood. Inhibins come in two forms: A and B (De Jong 1988).

However, Inhibin is a gonadal hormone that has been shown to strongly inhibit FSH synthesis by gonadotropic cells in the pituitary gland (pituitary hormone). Inhibin plays a crucial role in insect differentiation as a part of a wider family of (glyco) protein hormones and growth factors that also includes Mullerian inhibitory substance and growth factor-beta, as well as dimin and activin as two inhibitory beta subunits. Inhibin's effects on pituitary cells are neutralized by the second chemical, according to research. The presence of inhibin molecules in the placenta is similar to that found in the testes. Gonadal inhib has been identified as a distinct compound in seminal plasma, prostate, and gastric juice, according to the findings (Kolb *et al.* 2000).

In humans, a drop in peripheral FSH levels has been reported in many cases, however it is widely believed that these amino acid sequences have a biological significance apart

from this. Inhibitin is an inhibitory neurotransmitter produced by Sertoli cells in the testicles and granulosa cells in the ovary. Although it is generally accepted that FSH stimulates inhibin production, the question of whether or not other variables also play a role in this regulation is now up for dispute. Unfortunately, there are currently no accurate techniques for quantifying peripheral inhibin levels in people and experimental animals, therefore almost all evidence supporting inhibin's physiological role in the control of reproductive processes comes from observation rather than lab studies or research. At the period when the target cells for FSH divide, namely ovarian follicle development in women and puberty in sperm, peripheral FSH levels seem to be controlled through suppressing Sertoli cells and granulosa cells (De Jong 1988).

The term "stromal sex cord tumor" refers to tumors that are mostly composed of granulosa cells. Granulosa cell tumor patients may have elevated levels of serum Inhibin A or B, which could be dangerous. Inhibin B levels are found to be up to 60 times higher in 90-100 percent of granulosa cell tumors, which is a significant increase. Inhibin B is also elevated in 55-60% of mucinous epithelial tumors and can be considered a marker. Inhibins can be used as a complement to the CA 125 test, which is an ovarian cancer marker. Additionally, Inhibin B is a marker of ovulation. Every woman is born with a certain number of follicles containing Oocytes, these follicles constantly and naturally decline with age. The number of follicles remaining in the ovary at any given time is the ovarian reserve. As part of the infertility assessment, there are several ways to estimate a woman's ovarian reserve (Au *et al.* 1985, Meachem *et al.* 2001).

2.5.1 The chemical structure of inhibin B

These hormones are glycoproteins that belong to the TGF-beta family. As previously stated, inhibitins are made up of two subunits, one of which is the 20-kilodal mass and the other of which is the 13-kilodal mass, a disulfide bridge connects the two molecules. Two major isoforms of -subunit, inhibin A (also known as inhibin A) and inhibin B (also known as inhibin B), exist in the mature 32-kDa inhibin protein. A dimer of activins, which are closely linked to each other, has two subunits. Activins were first discovered in swine follicular fluid while purifying the homologous peptide, inhibin.

They were later discovered in human saliva (Ling *et al.* 1985). As a means of gaining knowledge about Inhibin. Glycoprotein hormone inhibin inhibits the production of pituitary gonadotrophins, principally follicle stimulating hormone (FSH). It consists of two subunits, each with a molecular weight of 20 kilodaltons (kDa), separated by a disulfide bond (FSH). There has been a lot of research on the inhibin molecule, which has a molecular weight of 31-32kD and has been cloned from bovine, pig, ovine, and human sources. Ovarian follicular fluids and granulosa and Sertoli cell culture mediums include forms with a higher molecular weight. These versions have subunits with greater molecular weights than the 31kD form, and are therefore denoted by their molecular weights, such as "44" for the higher molecular weight inhibin form (58kD). Both the -subunits and the inhibin dimers have been given the names A and B. Activins are dimers of the -subunit that activate FSH; they are classified as activin A, B, or AB depending on whether they are homodimers of subunits A or b, or heterodimers of the b subunit with another dimer. Female granulosa cells and male Sertoli cells are responsible for the majority of inhibin production in the gonadal system. We plan to investigate more possible cellular origins in the future (Meachem *et al.* 2001).

2.6 General Information of Zinc

The trace elements (sometimes known as significant trace metals) are minerals that are found in minute levels in living tissues. Some of them (Trace elements) are recognized to be nutritionally required for the human body, others may be essential but the evidence for this is just suggestive or inadequate at this time, and the others are deemed to be nonessential by the scientific community. At least one essential role in enzyme catalysis is played by trace elements (Cordain *et al.* 2002). Zinc, copper, and selenium are three of the nine biological trace elements that are vital in the functioning of spermatogenisis in males and the functioning of the ovaries in females. The adult testis has a high quantity of zinc, and the prostate contains a greater concentration of zinc than any other organ in the human body saves the brain (Bedwal *et al.* 1994, Khoobbakht *et al.* 2018). The oxidation-reduction processes of some metallic ions, such or like as zinc, iron, and copper, are involved in the energy metabolism process. Additionally, zinc, as an essential component of human diet, is critical to sperm production in humans and other animals (Cordain *et al.* 2002).

A vital mineral and heavy metal, zinc (Zn) is found in most multivitamins and mineral supplements sold over the counter today. It is also used therapeutically in greater dosages in men because of its ability to penetrate sperm cells and aid in the generation of sperm in males. When it comes to elements in the earth's crust, zinc ranks as one of the most abundant. It can be found in the atmosphere, soil, and water, and it can be found in all foods (Weirum *et al.* 2010, Chae *et al.* 2016). Zinc (2+) is a divalent metal cation, a zinc cation, and a monoatomic dication. It has three different functions. It serves as a metabolite in humans as well as a cofactor (Kim *et al.* 2001).

It is a chemical element that is one of nine trace elements that are vital to life. It is a Group 12 metal with a low melting point (IIb, or zinc group) of the periodic table and is one of the most extensively utilized metals in medicine, accounting for one crucial trace element out of nine. A significant amount of zinc is used in industry (Kim *et al.* 2001, Zia *et al*. 2014).

2.7 General Information of Calcium

When looking back over the history of the calcium element, Lime (calcium oxide, CaO) was a valuable substance that could be collected and formed by heating limestone. It has been used for hundreds of years to manufacture plaster and mortar, as well as other construction materials. In order to avoid further reduction, Antoine Lavoisier classed it as an "earth" since it seemed impossible to reduce it any more. However, he thought it was the oxide of an undiscovered element (Fleckenstein 1983, Ji *et al.* 2021).

1808 saw Humphry Davy experiment with electrolysis in an attempt to reduce damp lime. much as he had done with the two elements sodium and potassium the year before, but he was unsuccessful. Consequently, he attempted a combination of lime and mercury oxide, and although this generated an amalgam of calcium (Ca) and mercury in his investigations in the same topic, it was not enough to demonstrate that he had gotten a new element and to announce his findings to the scientific community. Also, Davy experimented with increasing the amount of lime in the combination, which resulted in more amalgam, which he then distilled to remove the mercury, leaving just calcium (Dorozhkin 2013, Ji *et al.* 2021).

2.8 The Phosphorus and Chemical Structure

In the human body, the element phosphorus (P) is a critical and primary element that is required for a variety of physiological and functional activities to take place. Phosphorus is an element that aids in the creation of bones, the storage of energy, and the production of nerves and muscles in the human body. The mineral phosphorus is available in a variety of foods, particularly meat and dairy products, making it very simple to get sufficient / adequate amounts of this element in the diet. Dietary digestible phosphorus is inversely proportional to total protein content in a non-enhanced unsettled diet, Phosphorus is primarily found in protein-rich foods in the human diet. Phosphorus-rich meals should be cooked by boiling, which decreases the phosphorus as well as the sodium (Na) and potassium (K) content, or by other methods of cooking that result in demineralization of the phosphorus and other minerals. Every meal should have its dosage of phosphorus-binding (P-B) treatment modified to account for the quantity and absorbability of phosphorus (P) present. A decrease in the phosphorus load in humans can only be achieved by dietician counseling that addresses the developing features of dietary phosphorus (P) control. Block and colleagues 1998, Kalantar-Zadeh 2010, and Cupisti *et al.* 2013. The majority of the phosphorus in the human body is found in the bones and teeth. There is, however, some phosphorus present in the bloodstream. A serum phosphorus test may be performed in labrotary to determine the amount of phosphorus in the blood (P test). When there is an excessive amount of phosphorus in the human blood, it is referred to as hyperphosphatemia (phosphorus divided). Hypophosphatemia is the reverse of hyperphosphatemia, which is having too little phosphorus in the serum of blood. Many conditions, comprehensive chronic alcohol abuse and vitamin D deficiency, can cause the blood phosphorus concentration or level to fall too low in the body (Block *et al.* 1998, Dhingra *et al.* 2007).

2.8.1 The effect of phosphor on infertility in men

The mineral phosphorous actually increases fertility in men, as studies indicate that a severe deficiency in phosphorus leads to hypophosphatemia disease, which leads to the emergence of many symptoms, including stress, explaining that phosphorus is the main component of ATP. Or “adenosine triphosphate”, which is an important source of

energy for the majority of the body's cells, and when phosphorous levels in the body decrease, the muscles cannot get the energy to contract, and this leads to weakness of the body's muscles and a feeling of fatigue and stress (Sun *et al.* 2015, Chotchutima *et al.* 2016).

The lack of phosphorous can also lead to anemia or anemia, where a severe deficiency of phosphorus leads to a negative effect on cells. When cells cannot obtain an adequate amount of adenosine triphosphate, which is the most important source of energy, this will lead to a lack of a fuel source. Necessary for many biochemical reactions important for life. The first effect that results from a deficiency of this compound is an increase in the red blood cells' eruption, which leads to anemia. Where it leads to a group of symptoms, including the symptoms that accompany anemia "fainting, pallor of the body, weak bones, and a feeling of pain in the bones and joints, and may lead to the destruction of the nervous system, loss of appetite and weight loss." (Chotchutima *et al.* 2016, Tsao *et al.* 2020).

2.9 General Information of Follicle-Stimulating hoHrmone

Women produce eggs and men produce sperm through the production of the follicle-stimulating hormone (FSH). It is connected with the process of reproduction and the development of eggs in women. The pituitary gland in humans produces a hormone known as folicle-stimulating hormone (FSH). With regards to the pituitary gland, it is a small gland that sits right in front of the sinus cavity, and it produces the hormones that control the body's metabolism and growth. In females, FSH is produced in the ovaries, whereas in men it is produced in the testes (FSH). It takes a complex system including the hypothalamus in the brain, the pituitary gland, and other hormones to regulate production of follicle-stimulating hormone (FSH). The hypothalamus secretes a number of reproductive-related endocrine hormones, including as gonadotropin-releasing hormone (GnRH) and luteinizing hormone (LH), which in turn stimulate the pituitary gland to generate follicle-stimulating hormone (FSH) (Zhang *et al.* 2013, Wang *et al.* 2018).

3. MATERIAL AND METHOD

The purpose of this investigation was to learn more about the issues of male infertility and their identification and detection methods. The main objective was to research the relationship between phytate acid and endocrine B hormone and the extent of the relationship between them and the rest of the chemical parameters. We will mention in this chapter the materials and devices used in this study. We will also briefly mention the methods of collecting samples and methods of preserving them until the final results are obtained.

3.1 Instrement

The equipment and materials that were used in the research and as mentioned in the Table 3.1, Table 3.2 and Table 3.3.

Table 3.1 Equipment used in study

Name	Use	Company – Contray
MiniVADIS	For hormones measurement	France
Microscop	For semen examination	China
Centrfuge Tube	For blood separation	India
ELAIS Reader	For hormones measurement	USA
ELAIS Washer	For hormones measurement	USA
Cobas C111	For hormones measurement	Germany
Incubator	For sample incubation	India
Water bath	For sample incubation	China
Refrigetor	For keep samples	China

Table 3.2 Materials used in study

Name	Use	Company-Contary
Standard Pipette 1mL	Blood (Serum) drawing	Germany
Standard Pipette 100 µL	Blood (Serum) drawing	Germany
Yellow Tip	Blood (Serum) drawing	China
Blue Tip	Blood (Serum) drawing	China
Distal Water	For sample dilution & clean	Germany
Slide	For semen measurement	China
Cover Slide	For semen measurement	China
WBC chamber	For semen measurement	China
White Tube	For blood keep	China
Eppendrof	For blood keep	China

Table 3.3 Kits used in study

Name		Use	Company-Contary
Phytic acid, PA	Kit	PC measurement	Catalogue Number: SL0055PI
INHB Hormone	Kit	INHIB measurement	Catalogue Number: SL0928Hu
Total Cholestrol	Kit	Tc measurement	FUJIFILM M. Systems
Triglyceride	Kit	Tg measurement	FUJIFILM M. Systems
High density lipoprotein	Kit	HDL measurement	FUJIFILM M. Systems
Luteinizing Hormone	Kit	LH measurement	Catalog Number: MBS2025524
Follicle-stimulating hormone	Kit	FSH measurement	Catalog: MBS494606
Testosterone	Kit	Testeo measurement	Catalog Number: MBS580035
Glucose	Kit	RBS measurement	Leaner glucose kit
Phosphor	Kit	P measurement	MyBioSource phosphours kit
Magnesium	Kit	Mg measurement	Cobas Phosphorus kit
Zinc	Kit	Zn measurement	Leaner zinc kit
Calcium	Kit	Ca measurement	Leaner calcium kit

3.1.1 Phytase enzyim kit

The phytic acid, (PA) ELISA kit is to assay phytic acid levels in serum or any biological fluid or cell culture.

Principle: This kit is an ELISA set that use the Sandwich-ELISA technique. Included in this kit is a Microelisa plate that has already been coated with an antibody specific for phytic acid. Add the necessary standards or patient samples to the bare wells of a Microelisa instrument and then incubate with the appropriate antibody to complete the analysis. Each well is then treated with a phytic acid-specific antibody coupled to horseradish peroxidase (HRP) at 37 °C. The unbound antibodies may be removed by washing. TMB substrate solution is utilized as the first step in the reaction in each well. Only the wells that contain the conjugated PC and HRP antibodies will first show blue, and then yellow with the addition of the stop solution. Spectrophotometric analysis of a sample reveals the optical density (OD) (ELISA) at 450 nm. The phytic acid content correlates directly to the OD reading. The phytic acid concentration was determined by comparing the OD of the samples to the standard curve.

Procedure: According to the provided kit from the production company for Human phytic acid ELISA Kit, PA ELISA Kit which it's Catalogue Number: L0055PI.

Dilution of Standards: Dilute the standard (Table 3.4) with micro-tubes first in order to obtain serial concentrations of the standard solution, then pipette a volume of 50ul from

each tube to a microplate well, each tube using at least two wells, where the total number of standard solutions will be a total of ten wells.

Table 3.4 Standard of phytic acid

120 ng/L	Standard-1	300µl OS + 150µl S-diluents
80 ng/L	Standard-2	300µl SN-1 + 150µl S-diluents
40 ng/L	Standard-3	150µl SN-2 + 150µl S-diluents
20 ng/L	Standard-4	150µl SN-3 + 150µl S-diluents
10 ng/L	Standard-5	150µl SN-4 + 150µl S-diluents

3.1.2 INHB hormone

The human Inhibin B (INHB) ELISA kit that we will use in this study aims to examine the levels of INHB in human blood serum, plasma, culture media, or any biological fluid and to determine the levels of this hormone.

Principle: We employed a Sandwich-ELISA ELISA kit, which is based on the same premise as the one used in this investigation. This kit's Microelisa assay plate was pre-coated with an antibody specific to the assay, so all that was needed to detect hormone levels was to add INHB standards or samples to the corresponding Microelisa wells and mix in the relevant antibody. Each well of the Microelisa stripping plate is then treated with an INHB-specific antibody coupled to horseradish peroxidase (HRP) for a total of three hours at 37 degrees Celsius. The components undergo a washing process to remove any dirt or debris. Each well receives a colorant—a TMB substrate solution—that is then allowed to react. Only the wells that contain the antibodies to INHB conjugated to INHB and HRP will show blue, and with the addition of the stop solution, the wells' colors will change to yellow, indicating that the greater the concentrations, the more intense the blue. Spectrophotometric analysis at 450 nm is used to determine optical (absorbance) density (OD). As the concentration of INHB increases, so does the value of the optical density. By comparing the optical density (OD) of samples to the OD of the standard curve, the concentration of INHB hormone in samples (research samples or patients) may be determined.

Procedure: According to the provided kit from the production company for Human Inhibin B (INHB) ELISA Kit which it's Catalogue Number: SL0928Hu.

Dilution of Standards: Dilute the standard (Table 3.5) with micro-tubes first in order to obtain serial concentrations of the standard solution, then pipette a volume of 50ul from each tube to a microplate well, each tube using at least two wells, where the total number of standard solutions will be a total of ten wells.

Table 3.5 Standard of phytic acid

180 pg/mL	Sta.N-1	300µl OS + 150µl S-diluents
120 pg/mL	Sta.N-2	300µl SN-1 + 150µl S-diluents
60 pg/mL	Sta.N-3	150µl SN-2 + 150µl S-diluent
30 pg/mL	Sta.N-4	150µl SN-3 + 150µl S-diluent
15 pg/mL	Sta.N-5	150µl SN-4 + 150µl S-diluent

3.2 Patient and Control Groups

Phytic acid and inhibin B levels and some chemistry tests were determined in the serum of healthy and low sperm counts. The study was divided into five levels, and each level contained (n = 60) samples. A pre and post study was also conducted for sterile people who depended on their feeding on type of food; statistical analysis was done for them at the conclusion of their study. which we will talk about in the previous chapter in detail (the results chapter). The study design was as shown in Figure 3.1.

3.3 Sample Preparation

After collecting whole blood from patients or healthy people (controls) of 5-7 mL, the whole blood was left to coagulate by leaving it undisturbed at room temperature. The blood clotting time usually takes 10 to 20 minutes. We removed the clot and extracted the serum by centrifugation at 2000-3000 rpm for 20 minutes. Serum was extracted into Abendrov tubes. In some samples, sediments appeared after centrifugation, and

according to the scientific methods recommended by the companies that supplied the number of tests, we re- centrifuged the sample again. Samples were stored at -20 °C. During the study period, we avoided repeated freeze-thaw cycles.

3.4 Semen Sample

Semen parameters, following the World Health Organization's guidelines, there are many factors that must be considered when collecting and analyzing semen, which can be divided into the first section that can be evaluated by the naked eye (microscopy) and those that cannot be evaluated by the naked eye (microscopy) (WHO 2010). Below are the requirements that a semen sample must meet to be classified as normozoospermia:

3.4.1 Macroscopics factors

Liquefaction: After the semen is collected, it is in a coagulated state and needs to be liquefied at a temperature of 37 in order to study it. The complete liquefaction time of a semen sample is approximately 15-20 minutes at 37°C.

Viscosity: This is a reference to the overall fluidity of the semen sample; the result can be either normal or high. The methods of reducing the viscosity of semen are the same as the methods of liquefaction. If the semen sample shows high levels of viscosity, that result may be due to an abnormality in the prostate.

The semen volume: The normal volume of semen, after 3 to 5 days of abstinence (intimacy), ranges from 1.5 to 6 mL. Low semen volume when it is larger is called Hypospermia, while semen volume when it is larger is called Hyperspermia. The absence of semen during ejaculation is also known as aspermia.

Colour: It can be said that the usual color of the semen is bright white, slightly yellowish. In some cases where the color is changed, in such a case it is advisable to study the possible reasons behind the change of color in the semen. Red blood cells (Hematospermia) or purulent cells are two examples of changes in color in the semen.

3.4.2 Microscopic assessment

Aggregation of spermatozoa: The adhesion of non-motile spermatozoa to each other or those motile spermatozoa with mucous sutures as well as non-sperm cells or debris is considered to be a non-specific accumulation.

Head-to-head, tail-to-tail, or mixed agglutination are all examples of specific agglutination, as are sperm adhesions. The sperm can be extremely sticky, limiting their ability to move beyond the sperm in some cases, but this is not always the case. Any signs of sperm-to-sperm contact can be found on the heads, tails, or midsections of motile sperm The results of the main type of agglutination (grade inversion (grades 1-4) and location of association (grades A to E) should be recorded (see Figure 2.3):
Grade 1: isolated <10 sperm per agglutination, many free sperm,
Grade 2: Average 10-50 sperm per agglutination, free sperm,
Grade 3: large clumps of >50 sperm, some sperm still free,
Fourth row: Total of all adherent and interconnected spermatozoa.

3.5 Digsin of Study

The levels of study of study

A. L1C (60) = According to the number of sperm (from more than 100)

B. L2C (60) = According to number of sperm (from 40 - 100)

C. L1P (60) = According to number of sperm (from 15 - 40)

D. L2P (60) = According to the number of sperm (from 1 to 14)

E. L3P (60) = According to the number of sperm (Zero)

F. Total (300)

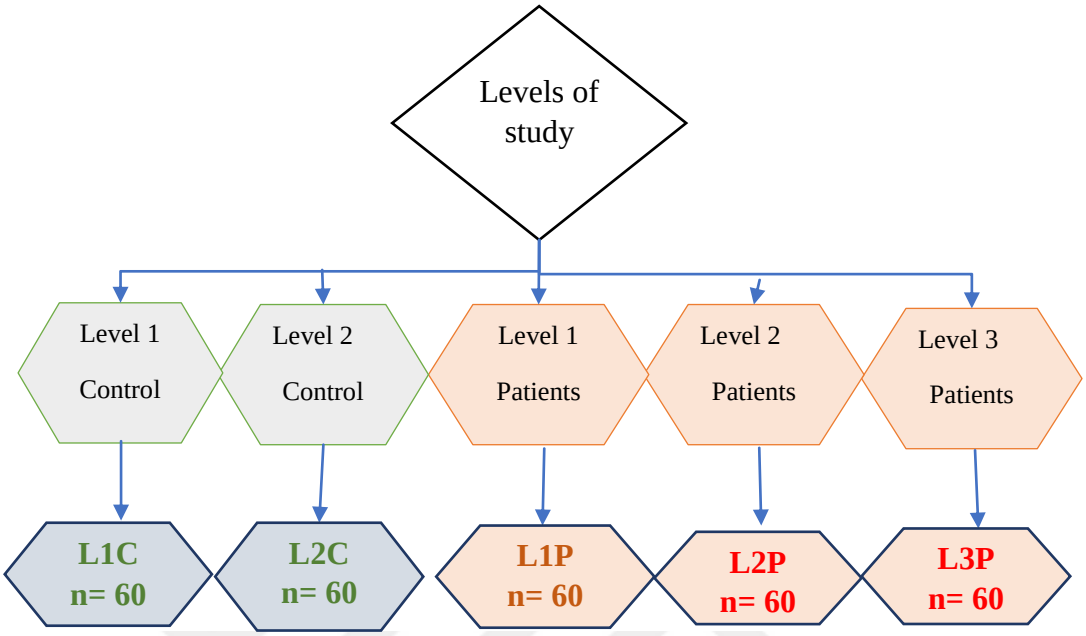

Figure 3.1 Digsin of study and samples number

3.6 The statistical Analysis

The statistical analysis program (MedCalc version 20.111) was used to find out the percentages that could be or be considered a positive result. While the (SPSS version 25) program was used to find the averages between the five levels of study by ANOVA one test. The same program was also used to find the results of the Pearson test for found the correlation between levels study with parameters study.

4. RESULTS AND DISCUSSION

Infertility and its problems are among the things that many studies are looking for in finding a solution to it, whether by early detection and identifying the disease and the type of problem or in following up on patients during treatment. For this reason, we have studied phytase acid and its effect it on some chemical variables whose increase, decrease, or presence in the first place can causing an obstacle in the formation of sperm or affect the effectiveness, number, shape, or movement of sperms. The parameters of the study will be explained starting from age and weight up to the last parameter according to the study tables listed in the following:

4.1 Age, Weight and BMI

Table 4.1 The mean+SD for age, weight and BMI in study effect of phytic acid with infertility men

Parameters		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Sig.
						Lower Bound	Upper Bound	
Age	L1C	60	25.6667	5.00951	1.29345	22.892	28.4408	0.033
	L2C	60	25.4000	4.80773	1.24135	22.737	28.0624	0.033
	L1P	60	28.0667	3.47371	0.89691	26.143	29.9903	0.033
	L2P	60	30.1333	5.46243	1.41039	27.108	33.1583	0.033
	L3P	60	35.0667	6.32982	1.63435	31.561	38.5720	0.033
	Total	300	28.8667	6.11452	0.70604	27.459	30.2735	0.033
Weight	L1C	60	80.1333	7.99881	2.06528	75.703	84.5629	0.049
	L2C	60	77.4667	8.00773	2.06759	73.032	81.9012	0.049
	L1P	60	78.4000	2.52982	0.65320	76.999	79.8010	0.049
	L2P	60	83.2000	5.96657	1.54056	79.895	86.5042	0.049
	L3P	60	90.5333	7.97197	2.05836	86.118	94.9481	0.049
	Total	300	81.9467	8.17030	0.94343	80.066	83.8265	0.049
BMI	L1C	60	26.0667	1.85372	0.47863	25.040	27.0932	0.050
	L2C	60	25.3440	1.60618	0.41471	24.454	26.2335	0.050
	L1P	60	26.1687	0.58155	0.15016	25.846	26.4907	0.050
	L2P	60	26.2867	1.48691	0.38392	25.463	27.1101	0.050
	L3P	60	31.5340	3.50473	0.90492	29.593	33.4749	0.050
	Total	300	27.0800	3.01338	0.34796	26.386	27.7733	0.050

4.1.1 The study of age

- **The mean age of the target samples in the study**

The research that we conducted on a group of men found that male fertility begins at an early age of 11-13 years, according to the questionnaire that we conducted, while it appears to decline or, so to speak, begins to decrease from the age of 76 and above. the average age of male infertility in our study, was approximately 28 to 35 years, and the poor quality of the sperm was a major factor, and the reason, according to the interpretation of the results of the questionnaire, the exposure of most men with infertility in this age to the gases of the battles that took place in Iraq, and the men were severely beaten in his back area during childhood or younghood period, and also, the late of men in performing a sperm examination and quality test for semen. The study concluded that the exposure of most men at this age to toxic gases, and the lack of semen examination and sperm quality measurements can be one of the reasons why infertility is difficult to treat, and thus leads us to early infertility in men. the greater the infertility problems in men in the formation of sperm and become difficult to treat increased the infertility problem as shown in Figure 4.1 and Table 4.1 below.

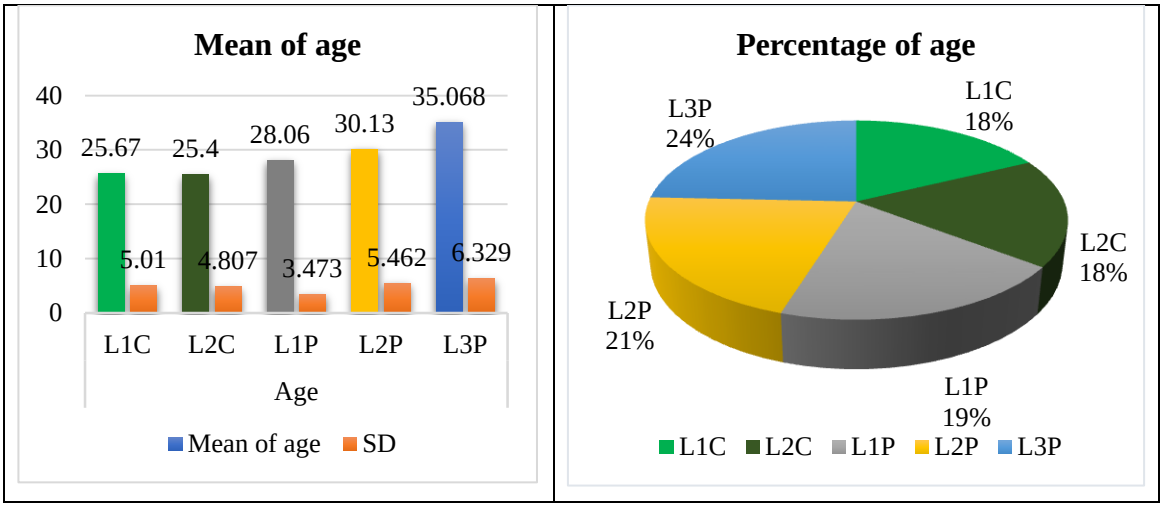

Figure 4.1 The mean for age between study levels in men

- **Post HOC tests for age**

When conducting post-tests, the results indicated that the significant differences were between L1C vs. L3P and L2C vs. L3P. L3P and L1P vs. L3P. L3P, which confirms that the third level was more susceptible to the disease. This can be interpreted as that fertilization or treatment of infertility can occur with less effective treatments in people at the third level, which is among the levels of patients (Table 4.2).

Table 4.2 The post-tests for age between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Age	L1C	L2C	0.26667	1.86319	1.000
		L1P	2.40000	1.86319	0.699
		L2P	4.46667	1.86319	0.128
		L3P	9.40000*	1.86319	0.000
	L2C	L1P	2.66667	1.86319	0.610
		L2P	4.73333	1.86319	0.093
		L3P	**9.66667***	**1.86319**	**0.000**
	L1P	L2P	2.06667	1.86319	0.801
		L3P	7.00000*	1.86319	0.003
	L2P	L3P	4.93333	1.86319	0.073

- **The receiver operating characteristic curve of age**

In the ROC curve for age, a higher value for a test result variable means that there is more evidence for a positive actual state, and a lower value means that there is less evidence for a positive actual state. The negative area and the positive area for age are shown in Table 4.3 and Appendix 1. The positive actual state is >28.

Table 4.3 The information the ROC curve for age

Variable	Age
Classification variable	Levels of study men infertility
Sample size	300
Positive level	120 (40.00 %)
Negative level	180 (60.00%)
Disease prevalence (%)	58.7
Area under the curve-ROC	
AUC-ROC	0.756
SE[a]	0.0675
0.95 Confidence-interval [b]	0.721 to 0.903
z statistic	4.825
Sig. Level P (Area=0.5)	<0.0001
The Youden (detecting disease) index	
Y-index J	0.5833
Associated (Impact point) criterion	>28
Sensitivity- postive area	63.64
Specificity- negative area	77.42

4.1.2 The study of weight in an infertility

- **The mean weight with levels of semen according to cuont nt of sperm**

According to the male fertility divisions, were divided into levels to know the relationship of obesity in men. Where the results of the five levels = L1C =20 %, L2C = 19 %, LP1= 19 %, LP2 = 20 %, and LP3 = 22 %, indicated that there was no statistical significance between the first and second levels. Differences in the third and fourth levels were statistically significant; the largest differences were found at the fifth level. Body mass index (BMI) and excess weight with sperm parameters may be affected by the metabolism of some important elements involved in the formation or activity of sperm in men such as zinc, phosphorous and magnesium. There was a negative association between weight gain and body mass index on the one hand and sperm activity (effectiveness) or total sperm count (total number) through the statistical results we obtained from our study. There is a clear correlation between weight gain and sperm count and body mass index. From here we conclude that the association between sperm parameters with weight gain and body mass was clear, from this we conclude that overweight and obese men may be exposed to greater than their peers Those with normal weight are subject to a pathological condition related to infertility, including low sperm production, or even no sperm. Which ultimately leads to an increase in the

likelihood that they will face difficulty in conceiving, and thus infertility (Figure 4.2 and Table 4.1).

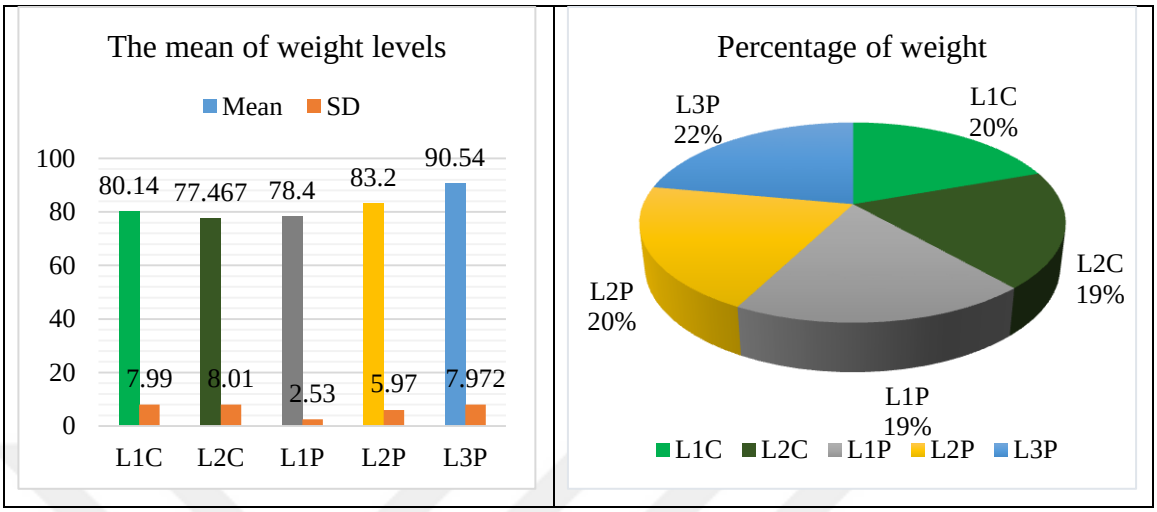

Figure 4.2 The mean for weight between study levels in men

- **Post HOC tests for weight**

When conducting post-tests for weight in Table 4.4, the results indicated that the significant differences were between L1C *vs.* L3P, L2C *vs.* L3P, and L1P *vs.* L3P, which confirms that the third level (L1P *ve.* L3P) was more susceptible to the disease.

Table 4.4 The post-tests for weight between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	$= \frac{\sqrt{MS}}{Nh}$	Tukey HSD.	Sig.
Weight	**L1C**	**L2C**	1.8	1.765	1.019830	0.954
		L1P	1.73333	1.765	0.982056	0.96
		L2P	3.06667	1.765	1.737490	0.746
		L3P	10.4*	1.765	5.892351	0.001
	L2C	**L1P**	0.06667	1.765	0.037773	1
		L2P	4.86667	1.765	2.757320	0.318
		L3P	12.2*	1.765	6.912181	0.000
	L1P	**L2P**	4.8	1.765	2.719546	0.331
		L3P	**12.13333***	**1.765**	**6.874407**	**0.00**
	L2P	**L3P**	7.33333	1.765	4.154861	0.04

- **ROC curve of weight**

The ROC curve for weight, higher test result variables indicate stronger evidence for a positive actual state and lower values indicate a negative case. The negative area and the positive area for age are shown in Table 4.5 and Appendix 2. The positive actual state is >77.

Table 4.5 The information the ROC curve for weight

Variable	Weight
Classification variable	Levels of study
Sample size	300
Positive level	120 (40.00 %)
Negative level	180 (60.00%)
Disease prevalence (%)	58.7
Area under the curve-ROC	
AUC-ROC	0.674
SE[a]	0.0650
0.95 Confidence-interval [b]	0.556 to 0.778
z statistic	2.680
Sig. Level P (Area=0.5)	0.0074
Youden index	
Y-index J	0.2925
Associated (Impact point) criterion	>77
Sensitivity- Postive area	84.09
Specificity- Negative area	45.16

4.1.3 The study of BMI in an infertility

- **BMI with levels of semen according to count of sperm**

Men with a BMI of less than 19 are infertile, people who have a digestive or eating disorder, such as: anorexia nervosa, bulimia nervosa, and those who follow a strict diet and used to rely on plant products in their diet are more exposed for their infertility. Weight gain also had a strong and direct effect, with a BMI between 25-30. Men follow an unhealthy eating pattern that increases their weight, as weight gain in men affects the shape, effectiveness and number of sperm when traveling, and thus affects the level of testosterone and inhibin B to be a catalyst for infertility in men at an early age. Male infertility is more common in obese men with a BMI over 30, who don't exercise, eat a

lot of chicken, and aren't in good health, according to the findings of this study (Figure 4.3 and Table 4.1).

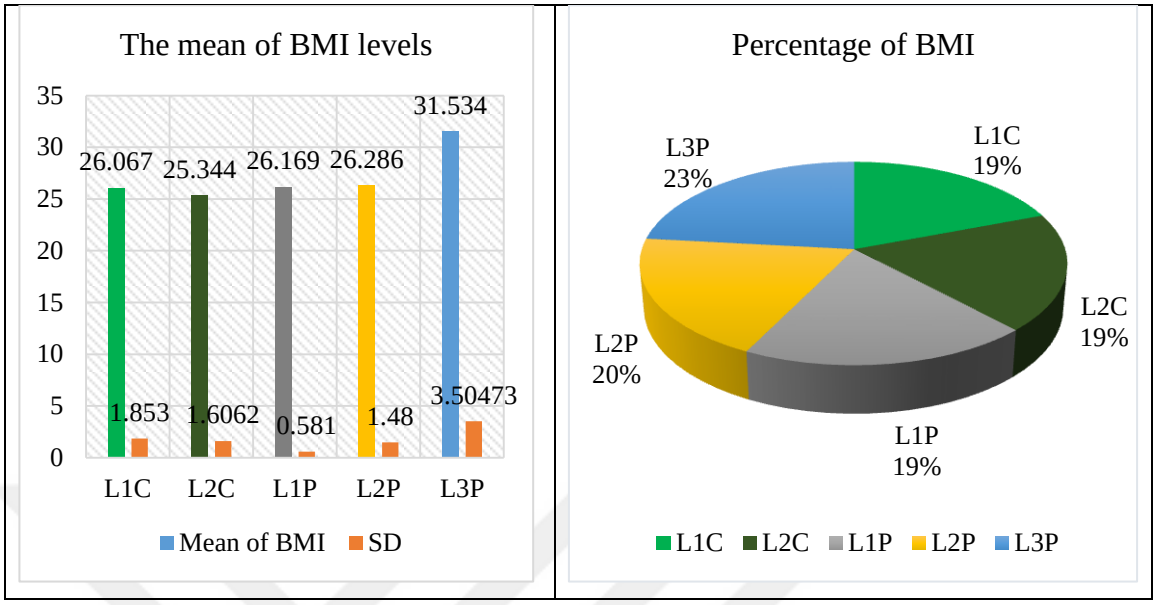

Figure 4.3 The mean for BMI between study levels in men

- **Post HOC tests for BMI**

When conducting post-tests for BMI in Table 4.6, the results indicated that the significant differences were between L1C *vs.* L3P, L2C *vs.* L3P, L1P *vs.* L3P and L2P *vs.* L3P, which confirms that the third level (L2C *vs.* L3P) was more susceptible to the disease.

Table 4.6 The post-tests for BMI between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Body mass index	**L1C**	**L2C**	0.72267	0.7456	0.868
		L1P	0.10200	0.7456	1.000
		L2P	0.22000	0.7456	0.998
		L3P	5.46733*	0.7456	0.000
	L2C	**L1P**	0.82467	0.7456	0.803
		L2P	0.94267	0.7456	0.714
		L3P	**6.19000***	**0.7456**	**0.000**
	L1P	**L2P**	0.11800	0.7456	1.000
		L3P	5.36533*	0.7456	0.000
	L2P	**L3P**	5.24733*	0.7456	0.000

- **ROC curve of BMI**

Higher test result variables' ROC curves for BMI show more evidence of a positive actual state and less evidence of a negative one. The negative area and the positive area for age are shown in Table 4.7 and Appendix 3. The positive actual state is >26.47.

Table 4.7 The information the ROC curve for BMI

Variable	BMI
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
AUC-ROC	0.713
SE[a]	0.0591
0.95 Confidence-interval [b]	0.597 to 0.811
z statistic	3.595
Sig. Level P (Area=0.5)	0.0003
Youden index	
Y-index J	0.4142
Associated (Impact point) criterion	>26.47
Sensitivity- postive area	70.45
Specificity- negative area	70.97

4.2 The study of main parameters - PA and INHB

Table 4.8 The mean+SD for phytic acid and inhibin B in study effect of phytic acid with infertility men

Parameters		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Sig.
						Lower Bound	Upper Bound	
PA	L1C	60	23.7333	9.66190	2.49469	18.3827	29.0839	0.001
	L2C	60	29.8667	6.28907	1.62383	26.3839	33.3494	0.001
	L1P	60	70.6667	15.89549	4.10420	61.8640	79.4693	0.001
	L2P	60	53.3333	7.64074	1.97283	49.1020	57.5646	0.001
	L3P	60	46.0667	5.87326	1.51647	42.8142	49.3192	0.001
	Total	300	44.7333	19.40071	2.24020	40.2696	49.1970	0.001
INHB	L1C	60	143.1333	15.18865	3.92169	134.722	151.5445	0.028
	L2C	60	110.4667	27.75883	7.16730	95.0943	125.8390	0.028
	L1P	60	44.3333	11.66599	3.01214	37.8729	50.7937	0.028
	L2P	60	36.4000	11.03760	2.84990	30.2876	42.5124	0.028
	L3P	60	29.3333	12.30370	3.17680	22.5198	36.1469	0.028
	Total	300	72.7333	48.71326	5.62492	61.5254	83.9412	0.028

4.2.1 The study of phytic acid in an infertility man

- **Phytic acid with levels of semen according to count of sperm**

Lately, emphasis has been placed on PA (Myo-inositol) as an anti-nutritional agent that causes problems in the human diet due to the inability to use phytates. Phytic acid is the main parameter in our research, which is coming in the first level after the hormone Inhibin B in our study. The results of sampling and statistical analysis (L1C = 11 %, L2C = 13 %, L1P = 31 %, L2P = 24 % and L3P = 21 revealed significant differences in means between groups, especially in the third level (LP1) where phytic acid levels rose to their highest levels, followed by the fourth level (LP2) and Then the fifth level (LP3), this leads us to the interpretation that the levels of phytic acid when they were at their highest levels had a strong influence on the process of spermatogenesis and the level of its activity, and thus its direct relationship to infertility in men. These results lead us to the following:

These results indicate that the presence of phytates in human food sources can form complexes in the metabolic stages in humans that are mainly insoluble with cations such as Ca, Mg, and Zn, and thus limit or reduce the Bioavailability of these minerals important in spermatogenesis. Because of this strong chelation of Ca, Mg, and Zn, phytates can limit and be a strong inhibitor in the absorption and digestion of important nutritional minerals in the process of sperm formation in humans. As a result of these averages in phytate levels and as a result of causing a decrease in the bioavailability of calcium (Ca), magnesium (Mg) and zinc (Zn), it can lead to an increase in male infertility. Especially that the basic foodstuffs depend mainly on chicken, whose feed contains large amounts of phytates, as well as relying mainly on phytate-containing vegetable products such as rice, wheat and corn, which is the main source of nutrition in our days (Figure 4.4 and Table 4.8).

- **Comparison with previous studies**

A new study published in Andrology journal has found that the phytate acid (which is a part of many plant foods) may play a crucial role in fertility. Phytic acid binds zinc,

calcium, magnesium, and iron. This leads to poor absorption by the body. As a result, men who have low levels of these essential minerals are more likely to experience infertility (Vazquez-Levin and Verón 2020).

The development of zinc's importance since 1963, when many scientists pointed out its clinical importance. And in order to search for inhibitors or things that prevent the absorption of zinc, many studies have been conducted on humans and animals, and some experiments have concluded that phytate acid has zinc and other element absorption, thus preventing its absorption and utilization by the body (Prasad 2014).

The research and focus were on information about the determination of phytate acid and its derivatives Myo-inositol (MI) (PI, PIP, GPI, IPG) in human tissues in men and body fluids, including semen in men. Phytate acid research on the functions of the male reproductive system and thus understanding its effect on sperm. The study concluded that the subjects who took plant food supplements containing large amounts of phytate acid had sperm parameters decline, and that the subjects who appeared to have low levels of phytate acid had an increase in the quality of the sperms (Vazquez-Levin and Verón 2020).

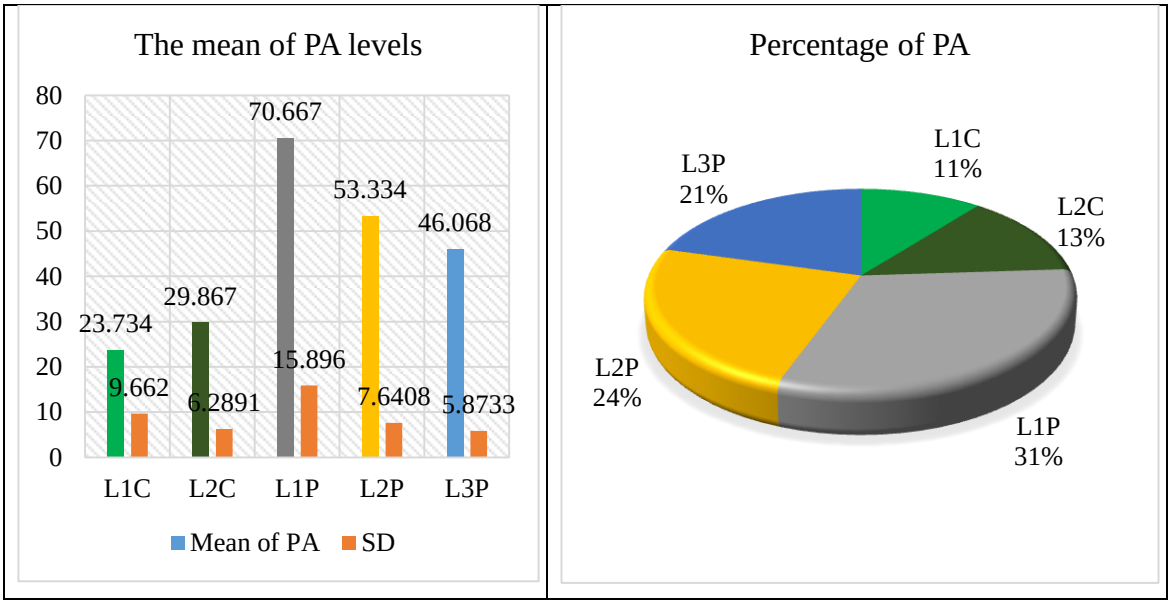

Figure 4.4 The mean for phytic acid between study levels in men

- **Post HOC tests for phytic acid**

When conducting post-tests (Table 4.9) for PA, the results indicated that the significant differences were between L1C *vs.* L1P, L1C *vs.* L2P, L1C *vs.* L3P, L2C *vs.* L1P, L2C *vs.* L2P and L1P *vs.* L3P which confirms that the third, fourth and fifth levels are affected by changes in the levels of phytic acid concentration. Thus, the increase in averages of phytic acid in the levels representing the first patients was more prone to infertility.

Table 4.9 The post-tests for phytic acid between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Phytic acid	**L1C**	**L2C**	6.13333	3.5719	0.430
		L1P	**46.93333***	**3.5719**	**0.000**
		L2P	29.60000*	3.5719	0.000
		L3P	22.33333*	3.5719	0.000
	L2C	**L1P**	40.80000*	3.5719	0.000
		L2P	23.46667*	3.5719	0.000
		L3P	9.20000	3.5719	0.072
	L1P	**L2P**	7.33333	3.5719	0.060
		L3P	24.60000*	3.5719	0.000
	L2P	L3P	7.26667	3.5719	0.261

- **ROC curve of phytic acid**

The ROC curve for phytic acid, this means that higher test results indicate stronger evidence for the positive actual state, and lower results indicate a negative one. The negative area and the positive area for age are shown in Table 4.10 and Appendix 4. The positive actual state is >37.

Table 4.10 The information the ROC curve for phytic acid

Variable	Phytic acid
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
AUC-ROC	0.975
SE[a]	0.0210
0.95 Confidence-interval [b]	0.909 to 0.997
z statistic	22.622
Sig. Level P (Area=0.5)	<0.0001
Youden index	
Y-index J	0.9032
Associated (Impact point) criterion	>37
Sensitivity- postive area	100.00
Specificity- negative area	90.32

4.2.2 The study of inhibin B in an infertility man

- **INHB with levels of semen according to count of sperm**

The study also found, for INHB, that there was a statistically high degree of statistical significance between patient's levels (L1P =12%, L2P = 10% and L3P = 8%) with levels of controls (LC1= 40% and LC2= 30%). The results of the study averages indicate that the enzyme B (protein B) hormone can be used as a diagnostic indicator to assess the status or presence of sperm in testicular tissues, as well as assess their function in men who were suffering from undescended testicles, after a period of surgery and lower them back to their normal position. We also found that subjects with anorexia had an undetectable or very low concentration of coenzyme B in their blood serum. Final conclusion:

Inhibin B hormone with testosterone, FSH and LH can be useful in early detection of the presence sperm inside of testicular tissue or the presence of sperm inside the testicle by measuring their concentration. By measuring testosterone and inhibin B together, we can inquire about the functional state of the Leydig cell. Thus, it leads us to estimate and evaluate the function of the Sertoli cell. Inhibin and AMH reflect Sertoli cell function and may provide a clear assessment of seminiferous tubule quality and the presence of immature sperm (Figure 4.5 and Table 4.8).

- **Comparison with previous studies**

Inhibin B, which can be defined as a dipeptide polypeptide hormone (A and B), regulates and aids in the production and secretion of follicle stimulating hormone (FSH) in women additionally men in a negative feedback loop in order to contribute to the reproductive process. The study found that Inhibin B is high at the beginning of life, then decreases before puberty, and then repeats again during puberty, whose levels were associated with follicle-stimulating hormones and LH. The study concluded that inhibin B plays a major and important role in regulating the hormonal axis of the hypothalamus, pituitary, and gonadal glands in men from infancy to adulthood. Inhibin B is a direct marker of Sertoli cell function in men and thus reflects testicular function in prepubertal men (Chada *et al.* 2003).

It seems that Inhibin B, about which there has been a lot of research recently, is a physiological feedback signal for FSH and HELL. It has been reported and concluded that here, Inhibin B, FSH, and LH levels in the blood, and testosterone in infertile men are correlated with each other, and as a result, they have a correlation with semen variables. It was found that men with Sertoli cell syndrome had lower levels of inhibin B (Bohring and Krause 1999). From the above results, we find the agreement with the interpretations of the results of our study that we reached.

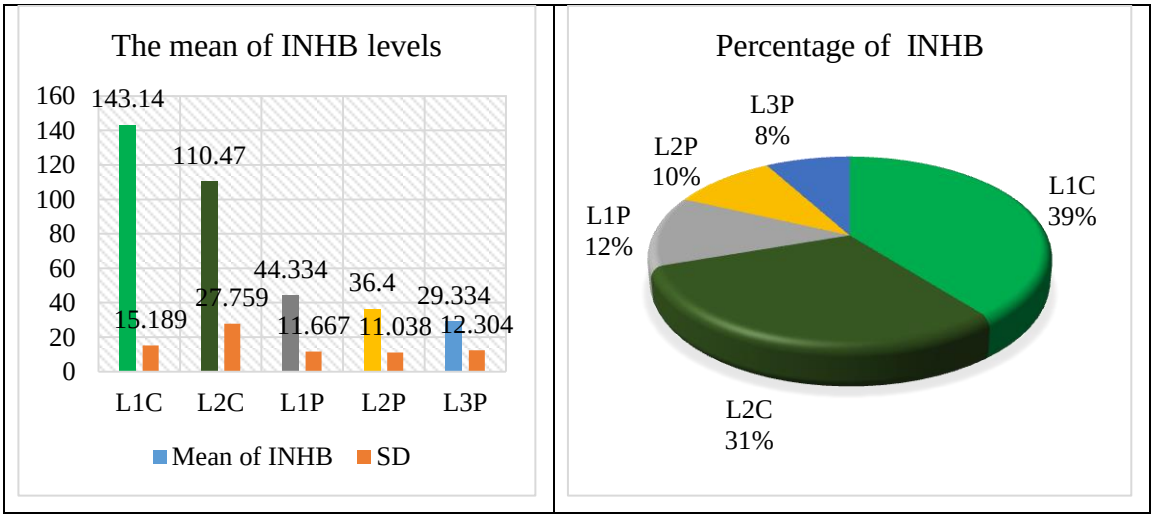

Figure 4.5 The mean for inhibin B between study levels in men

- **Post HOC tests for INHB**

When conducting post-tests for INHB in Table 4.11, the results indicated that the significant differences were between L1C *vs*. L1P, L1C *vs.* L2P, L1C *vs.* L3P and L2C *vs.* L2P, L2C *vs.* L3P. The results indicated that the most effective correlation in infertility patients was between the first and the fifth level (L1C *vs.* L3P).

Table 4.11 The post-tests for INHB between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Inhibin B hormone	**L1C**	**L2C**	13.66667	6.1338	0.038
		L1P	98.80000*	6.1338	0.000
		L2P	106.7333*	6.1338	0.000
		L3P	**113.8000***	**6.1338**	**0.000**
	L2C	**L1P**	66.13333*	6.1338	0.000
		L2P	74.06667*	6.1338	0.000
		L3P	81.13333*	6.1338	0.000
	L1P	**L2P**	7.93333	6.1338	0.696
		L3P	15.00000	6.1338	0.115
	L2P	**L3P**	7.06667	6.1338	0.778

- **ROC curve of INHB**

The ROC curve for inhibin B, A more positive actual state is more strongly supported by higher values of the test result variables and less indicate the negative case. The negative area and the positive area for age are shown in Table 4.12 and Appendix 5. The positive actual state is ≤63.

Table 4.12 The information the ROC curve for INHB

Variable	INHB
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The Error [a] of Intention	0.000
Confidence interval [b] of 95 percent	0.952 to 1.000
Perceived importance P (Area=0.5)	<0.0001
The ROC curve's slope (AUC)	1.000
Youden index	
Y-index J	1.0000
Associated (Impact point) criterion	≤63
Sensitivity- postive area	100.00
Specificity- negative area	100.00

4.3 The lipid profile with phytic acid in infertilety men

Table 4.13 The mean+SD for lipid profile in study effect of phytic acid with infertility men

Parameters		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Sig.
						Lower Bound	Upper Bound	
Tg	L1C	60	150.800	58.8511	15.1953	118.209	183.36	0.615
	L2C	60	131.333	65.9249	17.0217	94.8253	167.841	0.615
	L1P	60	124.533	41.3173	10.6681	101.652	147.414	0.615
	L2P	60	143.400	71.4200	18.4405	103.848	182.951	0.615
	L3P	60	121.266	54.7559	14.1379	90.9439	151.589	0.615
	Total	300	134.266	58.8236	6.79237	120.732	147.800	0.615
Tc	L1C	60	181.557	23.7214	6.12485	168.421	194.694	0.001
	L2C	60	180.514	35.6058	9.19339	160.796	200.232	0.001
	L1P	60	154.638	20.5133	5.29652	143.279	165.998	0.001
	L2P	60	157.026	28.7175	7.41484	141.123	172.929	0.001
	L3P	60	147.795	20.7876	5.36734	136.283	159.306	0.001
	Total	300	164.306	29.3636	3.39062	157.550	171.062	0.001
HDL	L1C	60	46.6000	10.8614	2.80442	40.5851	52.6149	0.208
	L2C	60	47.6000	6.96727	1.79894	43.7417	51.4583	0.208
	L1P	60	45.2000	4.55443	1.17595	42.6778	47.7222	0.208
	L2P	60	41.2000	6.75278	1.74356	37.4604	44.9396	0.208
	L3P	60	46.0667	8.32781	2.15023	41.4549	50.6785	0.208
	Total	300	45.3333	7.88144	.91007	43.5200	47.1467	0.208
LDL	L1C	60	102.786	27.0432	6.98254	87.8106	117.762	0.447
	L2C	60	98.0200	36.2923	9.37064	77.9220	118.118	0.447
	L1P	60	97.6933	31.1516	8.04333	80.4421	114.944	0.447
	L2P	60	92.5200	25.7833	6.65724	78.2417	106.798	0.447
	L3P	60	112.720	30.8808	7.97340	95.6188	129.821	0.447
	Total	300	100.748	30.4030	3.51065	93.7529	107.743	0.447
VLDL	L1C	60	34.0733	11.4689	2.96127	27.7221	40.4246	0.157
	L2C	60	26.0800	13.2462	3.42017	18.7445	33.4155	0.157
	L1P	60	24.9067	8.26347	2.13362	20.3305	29.4828	0.157
	L2P	60	28.6800	14.2840	3.68812	20.7698	36.5902	0.157
	L3P	60	24.2533	10.9511	2.82758	18.1888	30.3179	0.157
	Total	300	27.5987	12.0511	1.39154	24.8260	30.3714	0.157

4.3.1 The study of triglyceride in an infertility

- **Triglyceride with levels of semen according to count of sperm**

The results of triglyceride where we find that is a non-statistical significance although the percentage is clearly lower at the third level of study between patients' levels, percentage (LP1=19 %, LP2 = 21 % and LP3 = 18 %) with controls levels (LC1 = 22 % and LC2 = 20 %) at P = less 0.05 (Figure 4.6 and Table 4.13).

- **Comparison with previous studies**

It was observed upon conducting the statistical analysis that the inverse associations The prevalence of normal or abnormal sperm morphology was found in a J-shaped (low - high - high - low) relationship between high-density lipoprotein, triglyceride, and very low-density lipoprotein in the study (Liu *et al.* 2017).

The results indicated that the levels of TG, TC, and LDL were positively associated with the age of the subjects who had problems with semen parameters. Also, serum TC, TG, and LDL were positively associated with obesity in subjects with semen problems. For the levels of lipids in general in the serum and seminal plasma of men, the level of triglycerides had a slight positive relationship between them and the correlation was very weak (Lu *et al.* 2016).

In the above study, which was conducted by a number of researchers, it was found that triglycerides did not have any statistical significance and had a slightly ineffective inverse correlation, which is consistent with the outcomes of our study in terms of the result. Conclusion: Irregular fat metabolism in the male reproductive system in men may contribute to and aggravate infertility problems.

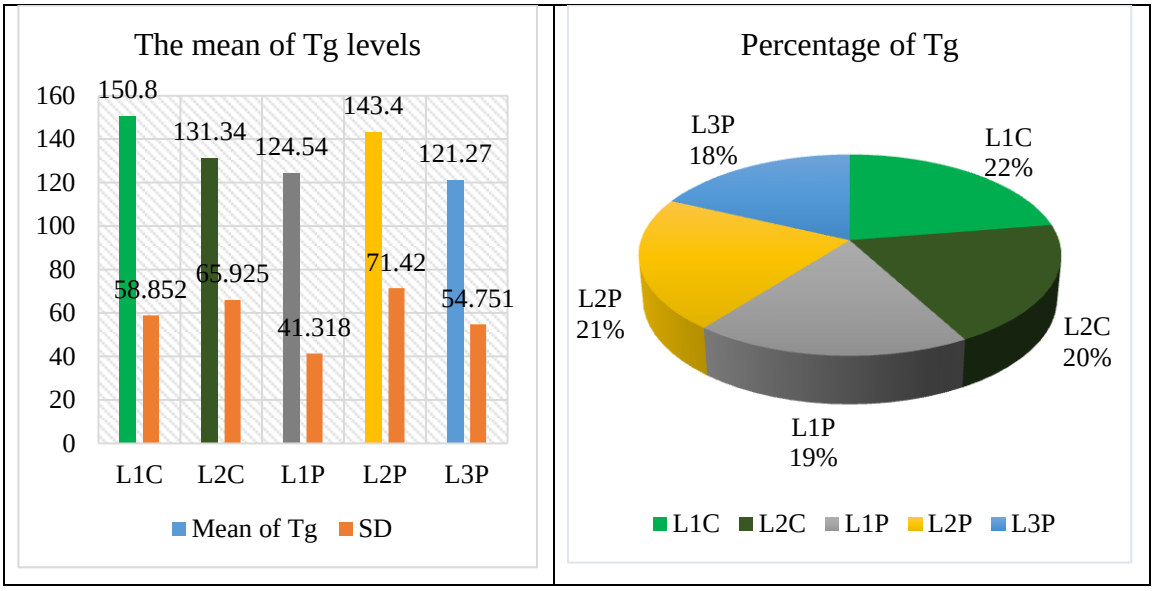

Figure 4.6 The mean for triglyceride between study levels in men

- **Post HOC tests for triglyceride**

When conducting post-tests for triglyceride in Table 4.14), the results indicated that the significant differences were between L1C *vs.* L1P, L1C *vs.* L3P, L1P *vs.* L2P and L2P *vs.* L3P. The results indicated that the most effective correlation in infertility patients was between the first and the fifth level (L1C *vs.* L3P).

Table 4.14 The post-tests for triglyceride between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Triglyceride	**L1C**	**L2C**	9.46667	21.67377	0.897
		L1P	26.26667*	21.67377	0.745
		L2P	7.40000	21.67377	0.997
		L3P	**29.53333**	**21.67377**	**0.013**
	L2C	**L1P**	6.80000	21.67377	0.998
		L2P	12.06667	21.67377	0.981
		L3P	10.06667	21.67377	0.990
	L1P	**L2P**	18.86667*	21.67377	0.907
		L3P	3.26667	21.67377	1.000
	L2P	**L3P**	22.13333*	21.67377	0.845

- **ROC curve of triglyceride**

In the ROC curve for triglyceride, larger values of the test result variables indicate stronger evidence for a positive actual state, and less indicates the negative case. The negative area and the positive area for age are shown in Table 4.15 and Appendix 6. The positive actual state is ≤106.

Table 4.15 The information the ROC curve for triglceride

Variable	Triglyceride
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.567
The Error [a] of Intention	0.0694
Confidence interval [b] of 95 percent	0.448 to 0.681
The z-score	0.966
Significance level P (Area=0.5)	0.3341
The Youden (detecting disease) index	
Y-index J	0.1870
Associated (Impact point) criterion	≤106
Sensitivity- Postive area	47.73
Specificity- Negative area	70.97

4.3.2 The study of total cholesterol in an infertility

- **Total cholesterol with levels of semen according to count of sperm**

The study also found, for total cholesterol, that there was a high statistical significance at a P-value < 0.047 between patient levels (LP1 = 19%, LP2 = 19%, and LP3 18%) with levels of controls (LC1 = 22% and LC2 = 22 %). These results explain that the increased consumption of phytic acid contributes to lowering cholesterol levels, which is the main substance in the formation of steroid hormones, including testosterone. This effect in turn leads to the fact that the percentage of total fat directly affects the concentration, quality, and number of sperm in men (Figure 4.7 and Table 4.13).

- **Comparison with previous studies**

It was found that in food supplements approved (especially containing a percentage of soy protein) About 25 g / day is the average daily intake in different countries. In the feed reduces the levels of total cholesterol and LDL. Soybeans contain different active substances such as trypsin inhibitors, saponins and phytic acid (Ahsan *et al.* 2018).

One of the important studies is to search for how to control the possible intervention strategies to control the levels of cholesterol and other types of fats in the blood. Since phytic acid is a friendly vegetarian ingredient in many diets for the elderly, those who are more susceptible to changes in lipids. Phytic acid has had a role in reporting its antioxidant effects on lipid metabolism and nutrients. Total cholesterol and low-density lipoprotein cholesterol in the P15 group (the supplementation group) were lower or lower than in the P0 group, according to a study (the group of controls - without supplementation) (Lee *et al.* 2007). These results indicate in this study Kidd and agree with our study, which confirmed that phytate affects the levels of lipids in the blood.

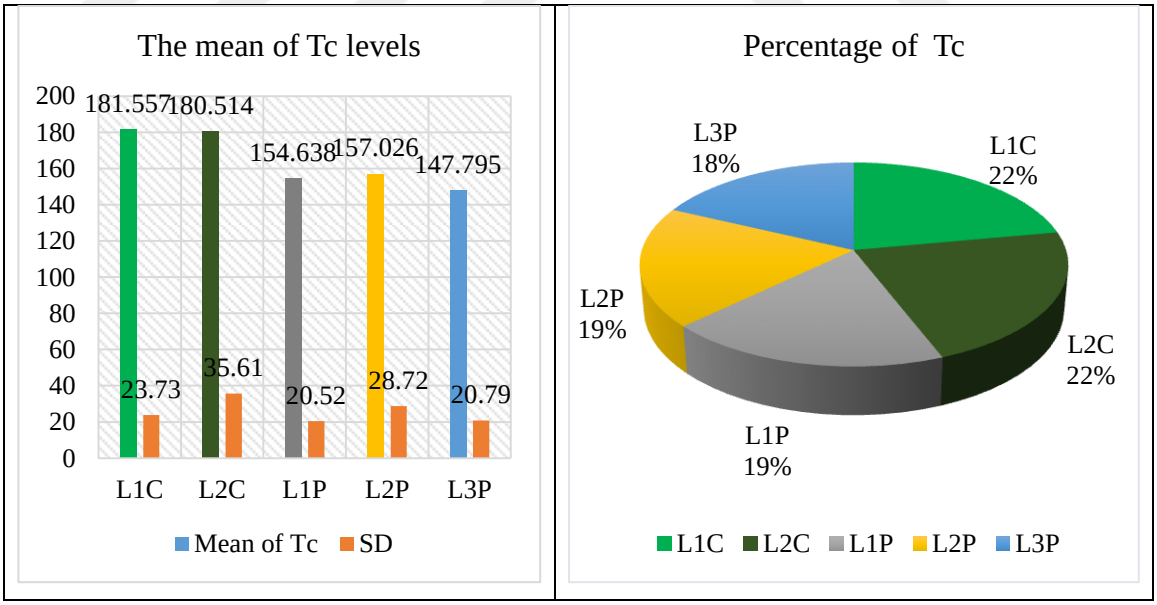

Figure 4.7 The mean for total cholesterol between study levels in men

- **Post HOC tests for total cholesterol**

When conducting post- for total cholesterol in Table 4.16, the results indicated that the significant differences were between L1C *vs.* L1P, L1C *vs.* L2P, L1C *vs.* L3P, L2C *vs.* L1P, L2C *vs.* L2P and L2C *vs.* L3P according to the HSD level, the more affected were L1C vs L3P.

Table 4.16 The post-tests for total cholesterol between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Total cholesterol	L1C	L2C	1.04303	9.67209	1.000
		L1P	26.91883*	9.67209	0.042
		L2P	24.53109*	9.67209	0.094
		L3P	**33.76276***	**9.67209**	**0.007**
	L2C	L1P	25.87580*	9.67209	0.018
		L2P	23.48807*	9.67209	0.120
		L3P	32.71973*	9.67209	0.010
	L1P	L2P	2.38773	9.67209	0.999
		L3P	6.84393	9.67209	0.954
	L2P	L3P	9.23167	9.67209	0.874

- **ROC curve of total cholesterol**

In the ROC curve for total cholesterol, this means that higher test results indicate stronger evidence for the positive actual state, and lower results indicate a negative one. The negative area and the positive area for age are shown in Table 4.17 and Appendix 7. The positive actual state is ≤168.

Table 4.17 The information the ROC curve for total cholesterol

Variable	Total cholesterol
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.769
The Error [a] of Intention	0.0542
Confidence interval [b] of 95 percent	0.657 to 0.859
The z-score	4.968
Perceived importance P (Area=0.5)	<0.0001
The Youden (detecting disease) index	
Y-index J	0.4274
Associated (Impact point) criterion	≤168
Sensitivity- Postive area	75.00
Specificity- Negative area	67.74

4.3.3 The study of HDL in infertility

- **HDL with levels of semen according to count of sperm**

The study also, for the High Disney Lipoprotein found that there is a non-statistical significance between patient's levels (L1P, L2P, and L3P) with controls levels (L1C and L2C), the means of the study was (L1C= 21%, L2C = 21%, L1P= 20%, L2P = 18% and LP3 = 20% at P = 0.208 (Figure 4.8 and Table 4.13).

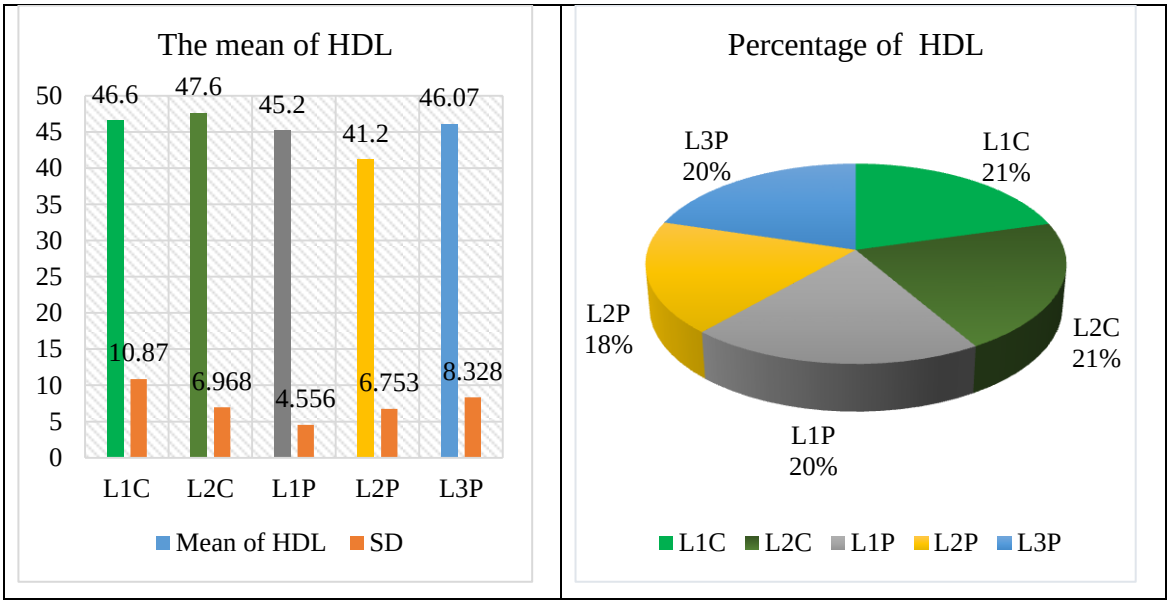

Figure 4.8 The mean for HDL between study levels in men

- **Comparison with previous studies**

The level of HDL in patients with oligospermia, who have azoospermia after the semen tests, was higher than those who had a normal concentration of sperm after the tests, especially those who had normal sperm motility or morphology (Lu *et al.* 2016).

- **Post HOC tests for HDL**

When conducting post-tests (Table 4.18) of HDL, the results indicated that the significant differences were between L2C vs L2P, according to the HSD level, this value was more affected by HDL.

Table 4.18 The post-tests for HDL between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
High density lipoprotein	**L1C**	**L2C**	1.00000	2.83882	0.997
		L1P	1.40000	2.83882	0.988
		L2P	5.40000	2.83882	0.326
		L3P	0.53333	2.83882	1.000
	L2C	**L1P**	2.40000	2.83882	0.915
		L2P	**6.40000**	**2.83882**	**0.172**
		L3P	1.53333	2.83882	0.983
	L1P	**L2P**	4.00000	2.83882	0.624
		L3P	0.86667	2.83882	0.998
	L2P	**L3P**	4.86667	2.83882	0.432

- **ROC curve of HDL**

The ROC curve for age, larger values of the test result variables indicate stronger evidence for a positive actual state and less indicate the negative case. The negative area and the positive area for age are shown in Table 4.19 and Appendix 8. The positive actual state is ≤52.

Table 4.19 The information the ROC curve for HDL

Variable	HDL
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.595
The Error [a] of Intention	0.0704
Confidence interval[b] of 95 percent	0.476 to 0.707
The z-score	1.353
Perceived importance P (Area=0.5)	0.1760
The Youden (detecting disease) index	
Y-index J	0.2639
Associated (Impact point) criterion	≤52
Sensitivity- Postive area	90.91
Specificity- Negative area	35.48

4.3.4 The study of LDL in an infertility

- **LDL with levels of semen according to count of sperm**

Also, for the Low-Density Lipoprotein found that there is a non-statistical significance between patient's levels (LP1, LP,2, and LP3) with controls levels (LC1 and LC2), the means othe f study was (LC1 = 21%, LC2 = 20, L1P= 19%, L2P= 18%, L3P= 22% at P = 0.447 (Figure 4.9 and Table 4.13).

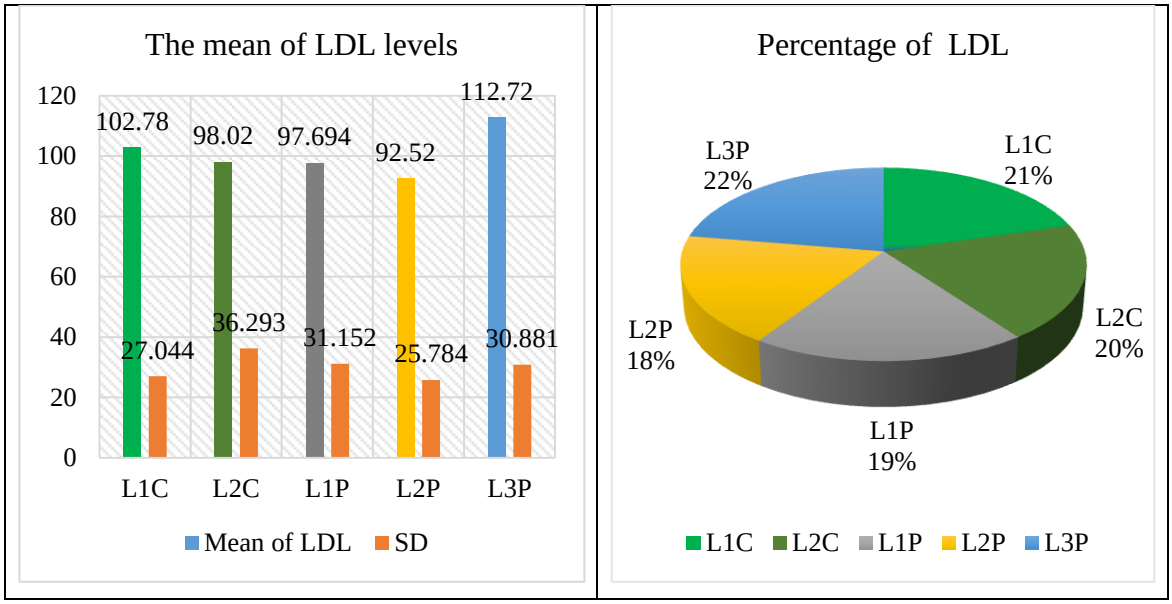

Figure 4.9 The mean for LDL between study levels in men

- **Post HOC tests for LDL**

When conducting post-tests (Table 4.20) of LDL, the results indicated that the significant differences between L2P *vs.* L3P according to the HSD level.

Table 4.20 The post-tests for LDL between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Low density lipoprotein	**L1C**	**L2C**	4.76667	11.12032	0.993
		L1P	5.09333	11.12032	0.991
		L2P	8.26667	11.12032	0.126
		L3P	9.93333	11.12032	0.898
	L2C	**L1P**	0.32667	11.12032	1.000
		L2P	5.50000	11.12032	0.988
		L3P	14.70000	11.12032	0.009
	L1P	**L1C**	7.26667	11.12032	0.067
		L2C	5.50000	11.12032	0.988
		L1P	5.17333	11.12032	0.990
	L2P	**L3P**	**20.20000**	**11.12032**	**0.037**

- **ROC curve of LDL**

In the ROC curve for LDL, this means that higher test results indicate stronger evidence for the positive actual state, and lower results indicate a negative one. The negative area and the positive area for age are shown in (Table 4.21 and Appendix 9). The positive actual state is >99.6.

Table 4.21 The information the ROC curve for LDL

Variable	LDL
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.531
The Error [a] of Intention	0.0693
Confidence interval[b] of 95 percent	0.412 to 0.647
The z-score	0.444
Perceived importance P (Area=0.5)	0.6570
The Youden (detecting disease) index	
Y-index J	0.2170
Associated (Impact point) criterion	>99.6
Sensitivity- Postive area	63.64
Specificity- Negative area	58.06

4.3.5 The study of VLDL in an infertility

- **VLDL with levels of semen according to count of sperm**

The Very Low-Density Lipoprotein found that there is a non-statistical significance between patients' levels (L1P, L2P, and L3P) with controls levels (L1C and L2C), the mean of study was (L1C= 25%, L2C= 19%, L1P= 18%, L2P= 21% and L3P= 17%) at P 0.157 (Figure 4.10 and Table 4.13). Our study concluded and found that an increase in VLDL leads to weak sperm parameters, especially in the shape and activity of sperm in men with abnormal sperm parameters.

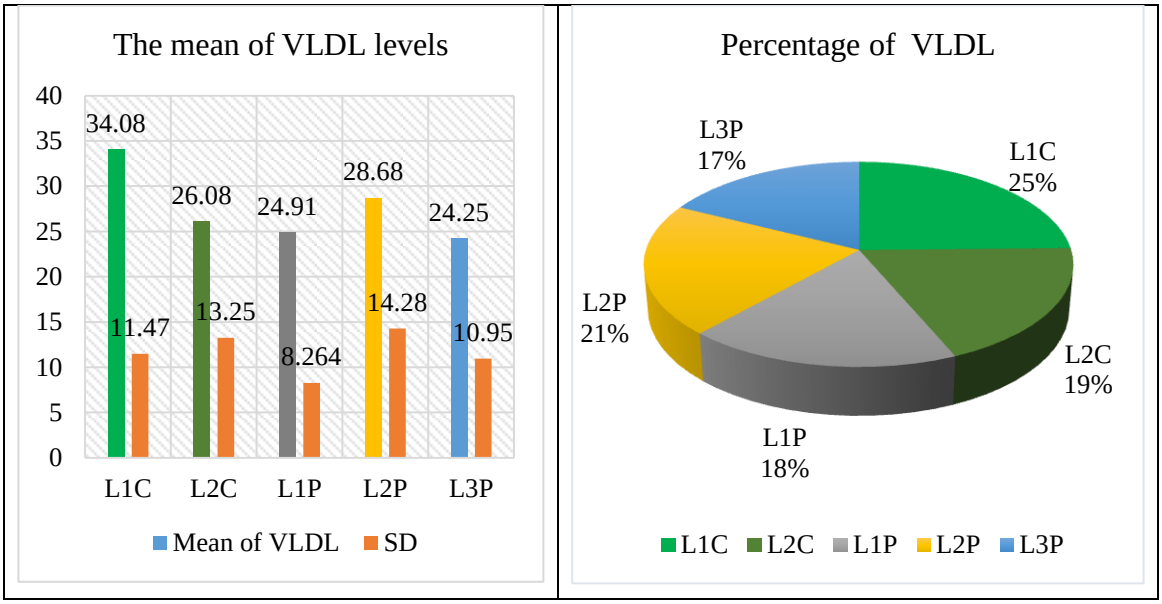

Figure 4.10 The mean for VLDL between study levels in men

- **Comparison with previous studies**

Under the influence of fat, the composition of the phospholipids and fatty acids of the sperm changes, and thus affects the parameters of the seminal fluid and the result is weak and infertility in men. When measuring a range of clinical chemical parameters such as blood glucose, total cholesterol, HDL, LDL, VLDL, triglyceride, FSH, LH, and testosterone levels. The study indicated that the increase in blood VLDL, total triglycerides, and total testosterone was significantly associated with decreased sperm count and motility in men. Also in the same study, FSH values in the serum of infertile men were inversely correlated with normal sperm morphology in men (Ergün *et al.* 2007).

- **Post HOC tests for VLDL**

When conducting post-tests (Table 4.22) of VLDL, the results indicated that the significant differences between L1C *vs*. L3P according to the HSD level.

Table 4.22 The post-tests for VLDL between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Very low-density lipoprotein	**L1C**	**L2C**	7.99333	4.31817	0.353
		L1P	9.16667	4.31817	0.022
		L2P	5.39333	4.31817	0.723
		L3P	**9.82000**	**4.31817**	**0.016**
	L2C	**L1P**	1.17333	4.31817	0.999
		L2P	2.60000	4.31817	0.974
		L3P	1.82667	4.31817	0.993
	L1P	**L2P**	3.77333	4.31817	0.906
		L3P	0.65333	4.31817	1.000
	L2P	**L3P**	4.42667	4.31817	0.843

- **ROC curve of VLDL**

The ROC curve for age, this means that higher test results indicate stronger evidence for the positive actual state, and lower results indicate a negative one. The negative area and the positive area for age are shown in Table 4.23 and Appendix 10. The positive actual state is ≤25.4.

Table 4.23 The information the ROC curve for VLDL

Variable	VLDL
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.587
The Error [a] of Intention	0.0708
Confidence interval[b] of 95 percent	0.467 to 0.699
The z-score	1.222
Perceived importance P (Area=0.5)	0.2217
The Youden (detecting disease) index	
Y-index J	0.2720
Associated (Impact point) criterion	≤25.4
Sensitivity- Postive area	65.91
Specificity- Negative area	61.29

4.4 The Hormones with Phytic Acid

Table 4.24 The mean+SD for LH, FSH, testeosterone and glucose in study effect of phytic acid with infertility men

Parameters		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Sig.
						Lower Bound	Upper Bound	
LH	**L1C**	60	4.2593	1.37090	.35397	3.5002	5.0185	0.003
	L2C	60	7.7947	5.34596	1.3803	4.8342	10.7552	0.003
	L1P	60	5.4193	1.17565	0.3035	4.7682	6.0703	0.003
	L2P	60	5.0940	1.59881	0.4128	4.2086	5.9794	0.003
	L3P	60	12.6053	5.60285	1.4466	9.5026	15.7081	0.003
	Total	300	7.0345	4.65915	0.5379	5.9625	8.1065	0.003
FSH	**L1C**	60	4.2527	2.27669	0.5878	2.9919	5.5135	0.002
	L2C	60	6.6812	3.79138	0.9789	4.5816	8.7808	0.002
	L1P	60	9.9060	5.25140	1.3559	6.9979	12.8141	0.002
	L2P	60	10.1305	4.94590	1.2770	7.3916	12.8695	0.002
	L3P	60	16.1544	5.9714	4.1238	7.3097	24.9991	0.002
	Total	300	9.4250	8.83636	1.0203	7.3919	11.4580	0.002
Testeo	**L1C**	60	5.3573	2.06198	0.5320	4.2154	6.4992	0.004
	L2C	60	5.3227	2.37207	0.6124	4.0091	6.6363	0.004
	L1P	60	3.5593	.59875	0.1546	3.2278	3.8909	0.004
	L2P	60	4.0353	1.69006	0.4363	3.0994	4.9713	0.004
	L3P	60	3.3533	1.76266	0.4551	2.3772	4.3295	0.004
	Total	300	4.3256	1.95190	0.2253	3.8765	4.7747	0.004
Glucose	**L1C**	60	100.7333	19.14407	4.9429	90.131	111.335	0.411
	L2C	60	101.2000	18.78145	4.8493	90.799	111.600	0.411
	L1P	60	93.9333	13.90512	3.5902	86.232	101.633	0.411
	L2P	60	99.0667	15.71835	4.0584	90.362	107.771	0.411
	L3P	60	91.6667	14.32115	3.6977	83.735	99.5975	0.411
	Total	300	97.3200	16.52366	1.9079	93.518	101.121	0.411

4.4.1 The study of LH in an infertility

- **LH with levels of semen according to count of sperm**

The LH found that there is high a statistical significance and showedimportancet between patient's levels (LP1, LP2, and LP3) with controls levels (LC1 and LC2), the means of the study was (L1C= 12%, L2C= 22%, L1P= 15%, L2P= 15%, L3P= 36% at P= 0.016.

The results indicate the importance of the hormone LH, because it increases testosterone production in the testicles (stimulates the Leydig cells in the testicles to produce testosterone, which has a significant impact on sperm production) in males. Men with high levels of luteinizing hormone can suffer from infertility, because the hormone LH

directly affects the functions of the male reproductive organs, which is evident in the third level of infertile men in our study. As for the low levels of the hormone LH, we found that it affects the volume of semen and thus limits the movement of sperm in males, which leads to the incomplete fertilization process (Figure 4.11 and Table 4.24).

- **Comparison with previous studies**

Only LH remained inversely associated with sperm motility after adjusting for FSH and TT levels (P for trend = 0.04), according to mutual adjustment analysis. Lower sperm progressive motility was also linked to higher LH concentrations (P for trend = 0.04). sperm morphology was negatively correlated with LH and FSH levels (Zhao *et al.* 2020).

Fetal follicle-stimulating hormone (FSH) and luteinizing hormone (LH) levels are both lower in semen than in blood, but there is no correlation between these two hormones and sperm counts. The FSH levels in semen appear to correlate well with blood levels; this correlation is only seen in azoospermic patients, however. When spermatogenesis is impaired, the mechanisms that allow LH to enter the semen may be ineffective. If this is the case, patients undergoing infertility treatment who have FSH and LH levels in their semen undetectable would have obstructive forms of azoospermia, whereas patients with elevated LH levels would have returned to a hormonal spermatogenic process (Abbaticchio and Giorgino 1983).

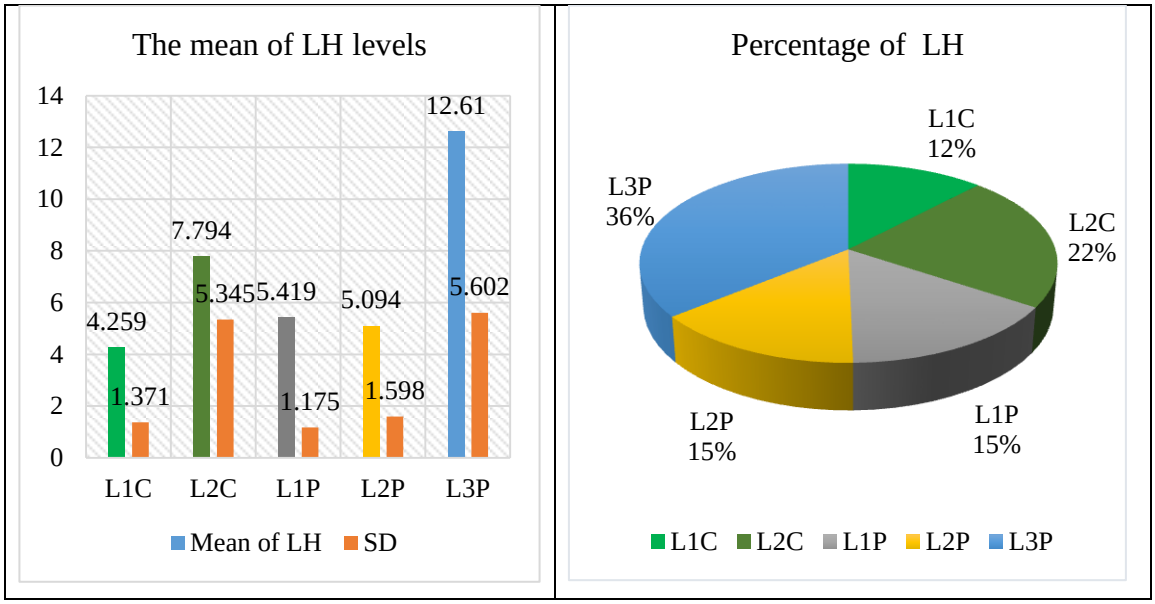

Figure 4.11 The mean for LH between study levels in men

- **Post HOC tests for LH**

When conducting post-tests (Table 4.25) of LH, the results indicated that the significant differences were between L1C *vs.* L3P, L2C *vs.* L3P, L1P *vs.* L3P, and L2P *vs.* L3P according to the HSD level, the L1C vs. L3P value was more affected by LH.

Table 4.25 The post-tests for LH between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Luteinizing hormone	**L1C**	**L2C**	3.53533	1.32453	0.069
		L1P	1.15993	1.32453	0.905
		L2P	0.83467	1.32453	0.970
		L3P	**8.34600***	**1.32453**	**0.000**
	L2C	**L1P**	2.37540	1.32453	0.386
		L2P	2.70067	1.32453	0.259
		L3P	4.81067*	1.32453	0.005
	L1P	**L2P**	0.32527	1.32453	0.999
		L3P	7.18607*	1.32453	0.000
	L2P	**L3P**	7.51133*	1.32453	0.000

- **ROC curve of LH**

The ROC curve for LH, larger values of the test result variables indicate stronger evidence for a positive actual state and less indicate the negative case. The negative area and the positive area for age are shown in Table 4.26 and Appendix 11. The positive actual state is >4.31.

Table 4.26 The information the ROC curve for LH

Variable	LH
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's corresponding area (AUC)	0.885
The Error [a] of Intention	0.0451
Confidence interval[b] of 95 percent	0.790 to 0.947
The z-score	8.539
Perceived importance P (Area=0.5)	<0.0001
The Youden (detecting disease) index	
Y-index J	0.2735
Associated (Impact point) criterion	>4.31
Sensitivity- Postive area	88.64
Specificity- Negative area	38.71

4.4.2 The study of FSH in an infertility

- **FSH with levels of semen according to count of sperm**

The FSH found that there is a statistical significance between the patient's levels (L1P= 21%, L2P = 22%, and L3P = 34%) with controls levels (L1C= 9% and L2C = 14% at P 0.039. In both men and women, the pituitary gland secretes the hormone FSH. Stimulates male testicular growth and, as a result, protein production, which is essential for male fertility because proteins aid in the formation of normal sperm cells, maintain their properties, and are, therefore, prepared for secretion when sperm are released. A change in FSH levels from the normal values (normal FSH levels in adult men is generally between 1.5 to 12.4 mIU/mL) was observed in infertile people who have an increase in phytic acid levels, and since the increase in FSH levels increases in Men indicate that the testicles do not function properly (Ramasamy *et al.* 2009), which leads

us to explain that elevated levels of phytic acid directly affect the secretion of FSH, which if its levels change may be difficult or impossible to produce or create sperm capable of fertilization (Figure 4.12 and Table 4.24).

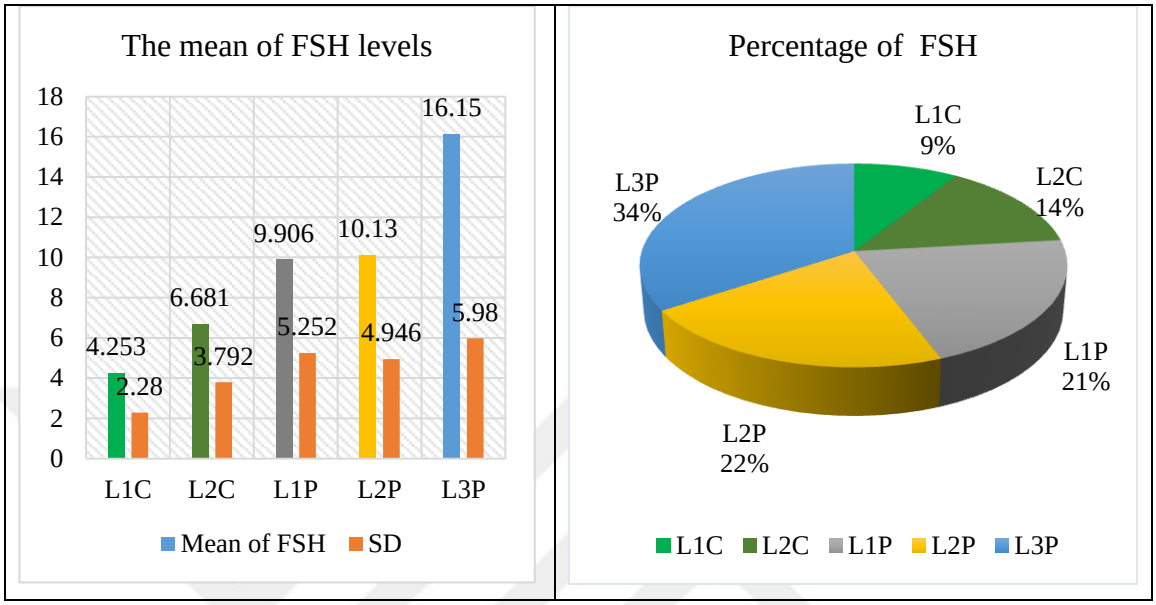

Figure 4.12 The mean for FSH between study levels in men

- **Post HOC tests for follicle-stimulating hormone**

Post-tests for FSH (Table 4.27) revealed significant differences between L1C *vs.* L3P and L2C *vs.* L3P with the latter being more affected by FSH at the HSD level.

Table 4.27 The post-tests for FSH between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Follicle-stimulating hormone	L1C	L2C	2.42853	2.95154	0.923
		L1P	5.65333	2.95154	0.319
		L2P	5.87787	2.95154	0.281
		L3P	9.90173*	2.95154	0.001
	L2C	L1P	3.22480	2.95154	0.810
		L2P	3.44933	2.95154	0.769
		L3P	11.47320*	2.95154	0.017
	L1P	L2P	0.22453	2.95154	1.000
		L3P	6.24840	2.95154	0.225
	L2P	L3P	6.02387	2.95154	0.258

- **ROC curve of FSH**

The ROC curve for FSH, larger values of the test result variables indicate stronger evidence for a positive actual state and less indicate the negative case. The negative area and the positive area for age are shown in Table 4.28 and Appendix 12. The positive actual state is 0.5132.

Table 4.28 The information the ROC curve for FSH

Variable	FSH
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.785
The Error of Intention [a]	0.0528
Confidence interval of 95 percent [b]	0.675 to 0.872
The z-score	5.400
Perceived importance P (Area=0.5)	<0.0001
The Youden (detecting disease) index	
Y-index J	0.5132
Associated (Impact point) criterion	>9.75
Sensitivity- Postive area	54.55
Specificity- Negative area	96.77

4.4.3 The study of testosterone in infertility

- **The testosterone with levels of semen according to the count of sperm**

The testosterone (ng/ml) found that there is high a statistical significance between patient's levels (LP1= 16%, LP2 = 19% and LP3 = 15%) with controls levels (LC1= 25% and LC2 = 25%) at P = 0.004. Depending on the results, phytic acid can be harmful to testosterone levels in men and through a direct effect on semen parameters. Incomplete cooking of vegetable products, for example, can cause an increase in blood levels of phytate acid concentrations. There were 29 samples consuming alcohol in large quantities for two weeks out of 150 samples representing infertile men, where the study averages recorded a sharp decrease in the testosterone hormone in the plasma of

infertile men and a rise in plasma luteinizing hormone concentration (Figure 4.13 and Table 4.24).

- **Comparison with previous studies**

The study group was treated daily with IP6 for 15 days and the same group without treatment for another 15 days. Sera were taken from the experimental group before and after the experiment and measured for testosterone, follicle-stimulating hormone (FSH), and luteinizing hormone (LH) to determine the effect on their toxic concentration. The study revealed changes in the pathological tissues through degeneration and severe mitosis that occurred within the spermatogonial nuclei of males, in addition to the enlargement of the spermatogenetic nucleus. We conclude from this that the toxicity of free radicals can be reduced by the use of phytic acid, which may have a significant protective role (Abu El-Saad Mahmoud 2009).

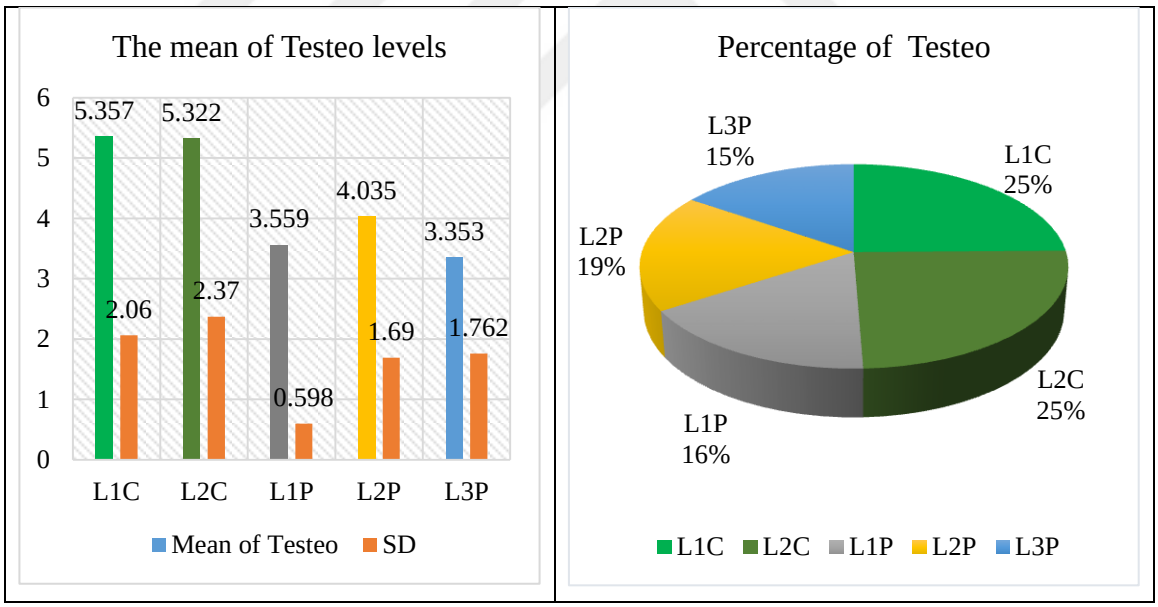

Figure 4.13 The mean for total testeosterone between study levels in men

- **Post HOC tests for testeosterone**

When conducting post-tests (Table 4.29) of total testosterone, the results indicated that the significant differences were between L1C *vs.* L3P, and L2C *vs.* L3P according to the HSD level, the L2C vs L3P value was more affected by testosterone

Table 4.29 The post-tests for testeosterone between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Testeosterone hormone	**L1C**	**L2C**	0.03467	0.65727	1.000
		L1P	1.79800	0.65727	0.059
		L2P	1.32200	0.65727	0.271
		L3P	**2.00400***	**0.65727**	**0.026**
	L2C	**L1P**	1.76333	0.65727	0.067
		L2P	1.28733	0.65727	0.297
		L3P	1.96933*	0.65727	0.030
	L1P	**L2P**	0.47600	0.65727	0.950
		L3P	0.20600	0.65727	0.998
	L2P	**L3P**	0.68200	0.65727	0.837

- **ROC curve of testosterone**

The ROC curve for total testeostrone. This means that higher test results indicate stronger evidence for the positive actual state, and lower results indicate a negative one. The negative area and the positive area for age are shown in Table 4.30 and Appendix 13. The positive actual state is ≤4.63.

Table 4.30 The information the ROC curve for total testeosterone

Variable	Testosterone
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.720
The Error of Intention [a]	0.0630
Confidence interval of 95 percent [b]	0.604 to 0.818
The z-score	3.490
Perceived importance P (Area=0.5)	0.0005
The Youden (detecting disease) index	
Associated (Impact point) criterion	≤4.63
Sensitivity- Postive area	88.64
Specificity- Negative area	54.84

4.4.4 The study of glucose in an infertility

- **The glucose with levels of semen according to the count of sperm**

The glucose found that there is a non-statistical significance between patient's levels (LP1 = 19 %, LP2 = 20 and LP3 = 19) with controls levels (LC1 = 21% and LC2 = 21%) at P = 0.411. Effective and important control of blood glucose through phytate supplements may be an alternative to controlling changes in diabetes levels in diabetic patients, as well as managing disorders in carbohydrate metabolism in humans in general and in men in particular (Figure 4.14 and Table 4.24), as some studies, including the study is at the below, where it found a difference in our results, and the reason is due to the difference in experience and the targeted patients in our study.

- **Comparison with previous studies**

To give more information on phytic acid in diabetic patients. This study examined the impact of phytate acid supplementation on blood sugar levels and was carried out by a team of researchers. Plevelsts participating in the research were tested for diabetes and glucose tolerance. Hemoglobin A1c levels were affected, but insulin levels remained unaffected, in the groups fed a phytate-acid diet. The results of the study indicate that phytate in the target samples reduced fasting blood glucose levels in the study samples with diabetes (Lee *et al.* 2006).

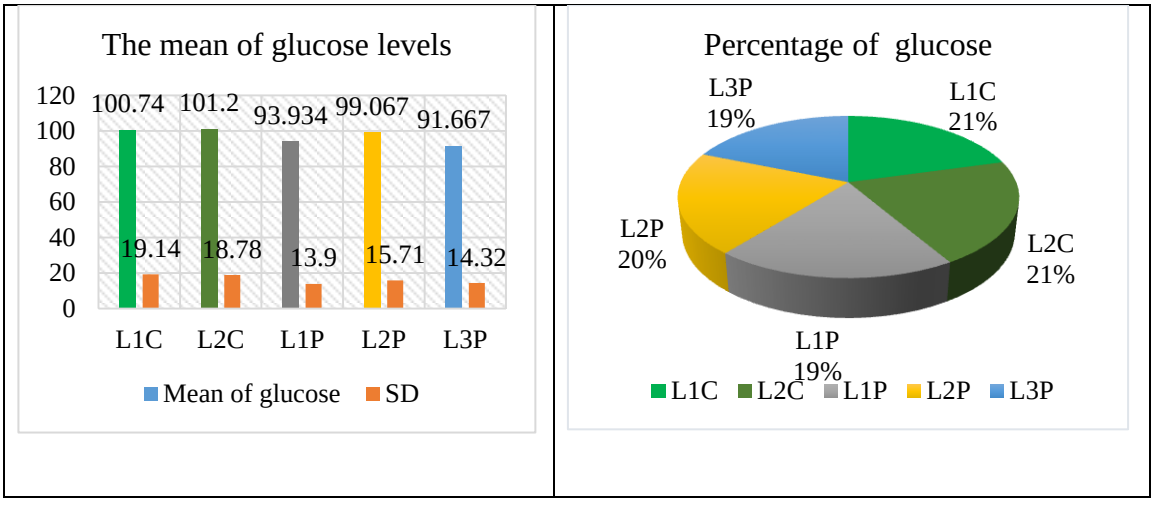

Figure 4.14 The mean for glucose between study levels in men

- **Post HOC tests for glucose**

When conducting post-tests (Table 4.31) of glucose, the results indicated that the significant differences between L1C *vs.* L3P, L2C *vs.* L3P and L1P *vs.* L2P, L2P *vs.* L3P according to the HSD level, L2C *vs.* L3P this value was more affected by HDL.

Table 4.31 The post-tests for glucose between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Glucose	L1C	L2C	0.46667	6.03269	1.000
		L1P	6.80000	6.03269	0.792
		L2P	1.66667	6.03269	0.999
		L3P	9.06667	6.03269	0.564
	L2C	L1P	7.26667	6.03269	0.749
		L2P	2.13333	6.03269	0.997
		L3P	9.53333	6.03269	0.515
	L1P	L2P	5.13333	6.03269	0.914
		L3P	2.26667	6.03269	0.996
	L2P	L3P	7.40000	6.03269	0.736

- **ROC curve of glucose**

The higher the values of the test result variables on the ROC curve for glucose, the more evidence there is that the actual state is positive, while lower values indicate the opposite. The negative area and the positive area for age are shown in Table 4.32 and Appendix 14. The positive actual state is ≤86.

Table 4.32 The information the ROC curve for glucose

Variable	Glucose
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.590
The Error of Intention [a]	0.0682
Confidence interval of 95 percent [b]	0.470 to 0.702
Perceived importance P (Area=0.5)	0.1881
The Youden (detecting disease) index	
Indicator J of Youden index	0.1928
Adjoining criteria	≤86
Sensitivity	38.64
Specificity	80.65

4.5 The Trace Elements with Phytic Acid

Table 4.33 The mean+SD for trace element in study effect of phytic acid with infertility men

Parameters		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Sig.
						Lower Bound	Upper Bound	
P	**L1C**	60	4.0120	0.63894	0.16497	3.6582	4.3658	0.013
	L2C	60	4.3407	0.76009	0.19625	3.9197	4.7616	0.013
	L1P	60	3.0703	0.61002	0.15751	2.7324	3.4081	0.013
	L2P	60	3.0171	0.89908	0.23214	2.5192	3.5150	0.013
	L3P	60	2.6532	0.73610	0.19006	2.2456	3.0609	0.013
	Total	300	3.4186	0.96546	0.11148	3.1965	3.6408	0.013
Mg	**L1C**	60	1.6713	0.59408	0.15339	1.3423	2.0003	0.718
	L2C	60	1.6933	0.62730	0.16197	1.3459	2.0407	0.718
	L1P	60	1.9049	0.28420	0.07338	1.7476	2.0623	0.718
	L2P	60	1.6420	0.42341	0.10932	1.4075	1.8765	0.718
	L3P	60	1.7127	0.73638	0.19013	1.3049	2.1205	0.718
	Total	300	1.7249	0.54934	0.06343	1.5985	1.8512	0.718
Zn	**L1C**	60	78.5333	31.96844	8.25421	60.8298	96.2369	0.011
	L2C	60	62.9333	26.57191	6.86084	48.2183	77.6484	0.011
	L1P	60	51.6667	7.44184	1.92147	47.5455	55.7878	0.011
	L2P	60	69.4667	29.32787	7.57242	62.2254	94.7079	0.011
	L3P	60	60.3333	17.56485	4.53522	50.6062	70.0604	0.011
	Total	300	66.3867	25.91822	2.99278	60.4234	72.3499	0.011
Ca	**L1C**	60	8.5987	1.00079	0.25840	8.0444	9.1529	0.037
	L2C	60	8.9633	0.89943	0.23223	8.4652	9.4614	0.037
	L1P	60	8.0640	0.47464	0.12255	7.8012	8.3268	0.037
	L2P	60	7.8020	0.51977	0.13420	7.5142	8.0898	0.037
	L3P	60	7.6333	0.83386	0.21530	7.1716	8.0951	0.037
	Total	300	8.2123	0.90495	0.10449	8.0041	8.4205	0.037
Fe	**L1C**	60	103.0444	26.67383	6.88715	88.2730	117.8159	0.033
	L2C	60	101.0285	21.03731	5.43181	89.3784	112.6785	0.033
	L1P	60	126.8613	48.15988	12.4348	100.191	153.5314	0.033
	L2P	60	137.9750	51.22697	13.2267	109.606	166.3435	0.033
	L3P	60	105.6451	35.84308	9.25464	85.7959	125.4944	0.033
	Total	300	114.9109	40.22998	4.64536	105.654	124.1670	0.033

4.5.1 The study of iron in an infertility

- **The iron with levels of semen according to the count of sperm**

Iron absorption from most plant foods can be reduced by up to 60% when phytates are present, according to the research. People with normal levels of vitamin C were found to be able to counteract the effects of phytates.

Phytic acid had a direct effect on iron absorption in men at levels II, III and IV who had problems in semen parameters. Iron's importance in the human body can now be better understood, and we found that the increase in iron levels directly affected Sperm movement within the semen (Figure 4.15 and Table 4.33).

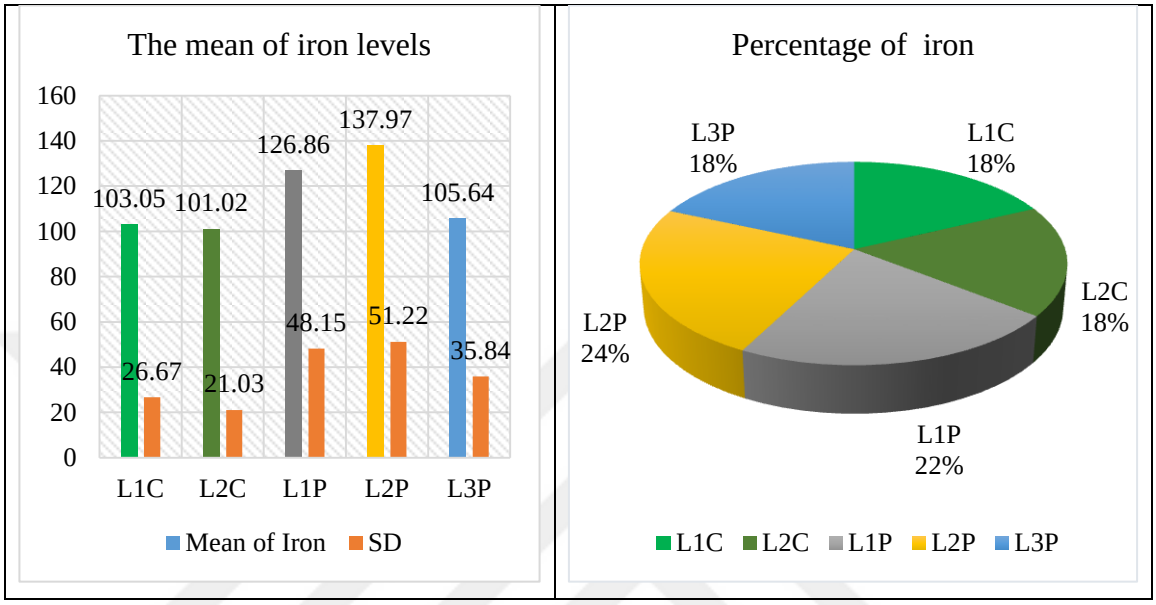

Figure 4.15 The mean for iron between study levels in men

- **Comparison with previous studies**

One of the main causes of anemia in infants is phytic acid, which inhibits iron absorption from plant sources such as cereals and legume-based complementary foods and plant products, both naturally occurring and fortified. It is possible to remove phytates from soy-based foods and supplements in humans, but because phytic acid is strongly inhibited at low concentrations, complete enzymatic hydrolysis by appropriate scientific methods is recommended (Hurrell 2004).

The reproductive system in men is particularly sensitive to excess iron than normal levels. Hypogonadism usually occurs, thus affecting the gonads, infertility and impotence if the excess iron is not removed from the body, and consequently, infertility disorders in men increase as a result of iron overload (Gabrielsen *et al.* 2018).

To test the effects of heavy iron on oxidative stress in the lab, use IP6 or phytic acid, and then study the effects of IP6 on heavy iron in the human body. The results showed that IP6 had chelating and inhibitory iron absorption activity in vitro, while inhibiting iron-induced or lipid peroxidation, and deoxyribose sugar degraded hydroxyl radicals. To conclude, we have discovered that IP6 or phytate acid, a naturally occurring component in cereals, can be used as an iron chelator to combat diseases like thalasis as well a liver-protective supplement (Bhowmik *et al.* 2017).

- **Post HOC tests for iron**

When conducting post-tests (Table 4.34) of iron, the results indicated that the significant differences between L1C *vs.* L1P, L2C *vs.* L2P, L1P *vs.* L3P and L2P *vs.* L3P according to the HSD level, L2C *vs.* L2P this value was more affected by iron, The averages indicated that the effect starts from the second level, in contrast to the above chemical parameters.

Table 4.34 The post-tests for iron between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Iron	L1C	L2C	2.01599	14.03079	1.000
		L1P	23.81689	14.03079	0.442
		L2P	14.93055	14.03079	0.105
		L3P	2.60069	14.03079	1.000
	L2C	L1P	15.83288	14.03079	0.359
		L2P	36.94655	14.03079	0.015
		L3P	4.61668	14.03079	0.997
	L1P	L2P	11.11367	14.03079	0.932
		L3P	21.21620	14.03079	0.558
	L2P	L3P	32.32987	14.03079	0.156

- **ROC curve of iron**

When looking at the ROC curve for iron, higher values of the test result variables indicate more proof of a positive actual state, while lower values indicate a negative

case. The negative area and the positive area for age are shown in Table 4.35 and Appendix 15. The positive actual state is L3P = >142.

Table 4.35 The information the ROC curve for iron

Variable	Iron
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.614
The Error of Intention [a]	0.0649
Confidence interval of 95 percent [b]	0.494 to 0.724
The z-score	1.757
Perceived importance P (Area=0.5)	0.0790
The Youden (detecting disease) index	
Indicator J of Youden index	0.3409
Adjoining criteria	>142
Sensitivity- Psitive area	34.09
Specificity- Negative	100.00

4.5.2 The study of phosphor in an infertility

- **Phosphorus with levels of semen according to count of sperm**

The phosphorus (mg/dl) found that there is a non-statistical significance between patient's levels (LP1, LP2, and LP3) with controls levels (LC1 and LC2) at P = 0.002. There was a percentage (63%) of patients who appeared to have high levels of phytate acid and suffer from a lack of phosphorus, and the decrease in the amount of phosphorus absorbed was associated with the effectiveness and motility of sperms (Figure 4.16 and Table 4.33).

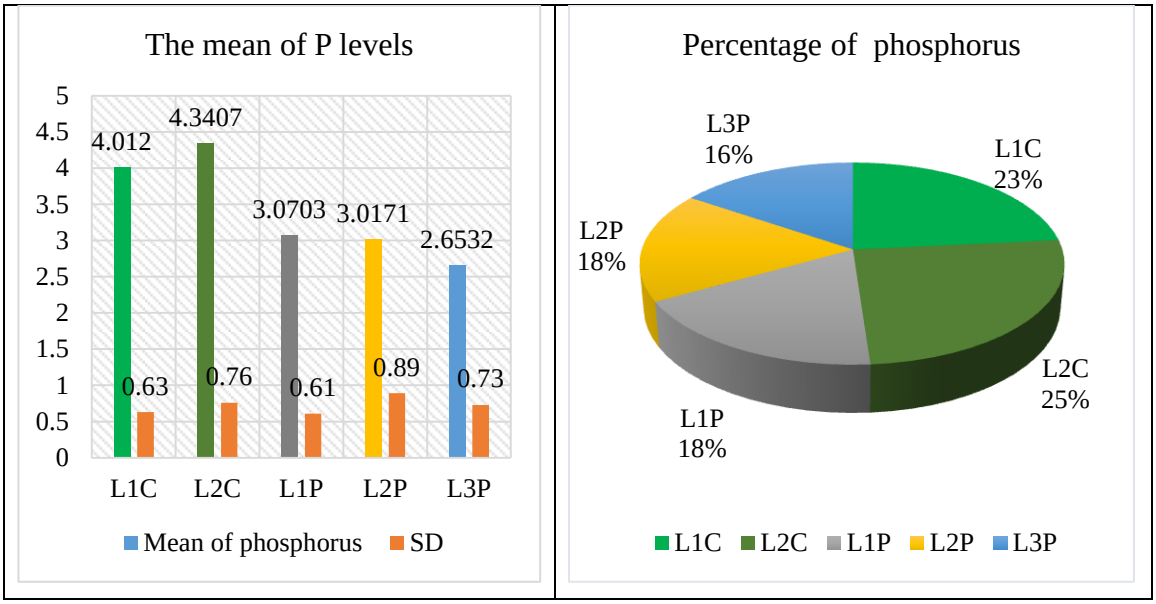

Figure 4.16 The mean for phosphorus between study levels in men

- **Comparison with previous studies**

And it was confirmed by the study of the researcher Levine, et al., who heid: The organic part (components of seminal plasma) It is widely accepted that human seminal plasma contains phosphate esters, making it an essential component, especially glycerylphosphorylcholine (GPC), phosphorylcholine (PCh), and inorganic phosphate (Pi). The epididymis is where he came to the conclusion that male sperm do not reach full maturity (including all of their characteristics) and thus do not have the ability to fertilize. Only if they have enough phosphorus do they become motile, which means that they are capable of sperm's ability to fertilize (Levine, *et al.* 2002).

MI was found to be an effective supplement for men's sperm quality, according to researchers. Larger prospective randomized controlled studies will be required to confirm whether MI supplementation has positive or negative effects on the treatment of male infertility (Vazquez-Levin and Verón 2020).

- **Post HOC tests for phosphorus**

When conducting post-tests (Table 4.36) of phosphorus, the results indicated that the significant differences were between L1C *vs.* L2P, and L2C *vs.* 3P and other levels

according to the HSD level, the L2C *vs.* L3P value was more affected by the phosphorus.

Table 4.36 The post-tests for phosphorus between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Phosphorus	**L1C**	**L2C**	0.32867	0.26874	0.738
		L1P	0.94174*	0.26874	0.007
		L2P	0.99494*	0.26874	0.004
		L3P	1.35877*	0.26874	0.000
	L2C	**L1P**	1.27041*	0.26874	0.000
		L2P	1.32361*	0.26874	0.000
		L3P	**1.68744***	**0.26874**	**0.000**
	L1P	**L2P**	0.05320	0.26874	1.000
		L3P	0.41702	0.26874	0.533
	L2P	**L3P**	0.36383	0.26874	0.659

- **ROC curve of phosphorus**

Phosphor ROC analysis shows that higher values of the test result variables indicate stronger evidence of an actual positive state, while lower values indicate the opposite. The negative area and the positive area for age are shown in Table 4.37 and Appendix 16. The positive actual state is ≤3.08.

Table 4.37 The information the ROC curve for phosphorus

Variable	Phosphorus
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.852
The Error of Intention [a]	0.0436
Confidence interval of 95 percent [b]	0.751 to 0.923
The z-score	8.078
Perceived importance P (Area=0.5)	<0.0001
The Youden (detecting disease) index	
Indicator J of Youden index	0.5946
Adjoining criteria	≤3.08
Sensitivity- Positive area	65.91
Specificity- Negative area	93.55

4.5.3 The study of magnesium in infertility

- **The Magnesium with levels of semen according to the count of sperm**

The Mg found that there is a statistical significance between patient's levels (LP1= 17 %, LP2 = 8 % and LP3 = 9 %) with controls levels (LC1 = 35 % and LC2 = 31 %) at P 0.018. Magnesium in men controls the follicle-stimulating hormone, or FSH, because FSH is dependent on magnesium status, and because FSH is the hormone that stimulates sperm formation in men. Thus, low magnesium in men can indicate low levels of testosterone and thus affect the process of spermatogenesis in men (Figure 4.17 and Table 4.33).

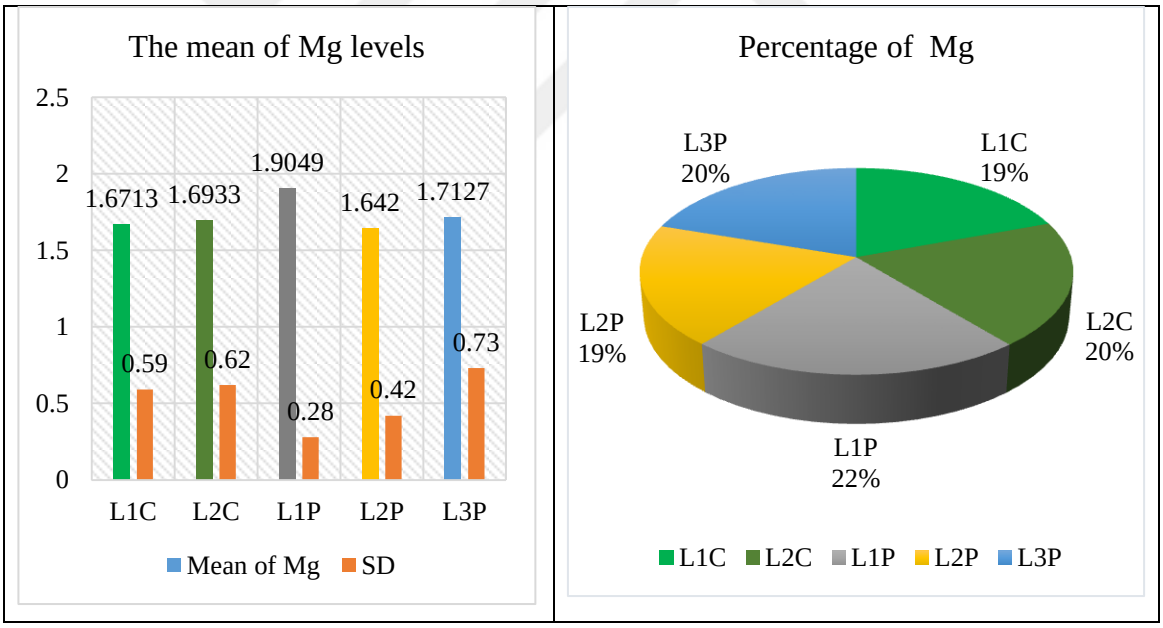

Figure 4.17 The mean for magnesium between study levels in men

- **Comparison with previous studies**

The concentration of some trace elements (calcium and magnesium) in the plasma of normal men (fertile men) was significantly higher when compared with infertile men (Bassey *et al.* 2013).

The levels of magnesium in semen plasma were significantly decreased in men in all infertility groups that were studied. The study concluded that magnesium works in different ways in the body to maintain the natural environment and the continuation of sperm activity until fertilization occurs in men (Abdul-Rasheed 2010).

- **Post HOC tests for magnesium**

When conducting post-tests (Table 4.38) of magnesium, the results indicated that the significant differences were between L1C vs L1P according to the HSD level, the L1C vs L1P value was more affected by magnesium.

Table 4.38 The post-tests for magnesium between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Magnesium	L1C	L2C	0.02200	0.2032	1.000
		L1P	**1.23360***	**0.2032**	**0.047**
		L2P	0.02933	0.2032	1.000
		L3P	0.04133	0.2032	1.000
	L2C	L1P	0.21160	0.2032	0.835
		L2P	0.05133	0.2032	0.999
		L3P	0.01933	0.2032	1.000
	L1P	L2P	0.26293	0.2032	0.696
		L3P	0.19227	0.2032	0.878
	L2P	L3P	0.07067	0.2032	0.997

- **ROC curve of magnesium**

ROC curve for magnesium, the values of the test result variables indicate the no-effect condition towards the positive or to the negative condition. The negative region and the positive region for age are shown in Table 4.39 and Appendix 17. Actual positive state > 1.99.

Table 4.39 The information the ROC curve for magnesium

Variable	Magnesium
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.560
The Error of Intention [a]	0.0682
Confidence interval of 95 percent [b]	0.441 to 0.675
The z-score	0.886
Perceived importance P (Area=0.5)	0.3754
The Youden (detecting disease) index	
Indicator J of Youden index	0.1833
Adjoining criteria	>1.99
Sensitivity- Positive area	40.91
Specificity- Negative area	77.42

4.5.4 The study of Zn in an infertility

- **Zn with levels of semen according to count of sperm**

We found that zinc (μg/dL) has a high statistical significance between the levels of the patient (LP1, LP2, and LP3) with the levels of the controls (LC1 and LC2) at P = 0.011. We found in our study that men who had normal levels of zinc were not suffering from problems in sperm parameters except 9 (5%) cases were slightly low out of a total of 120 (95%) cases in the first and second levels (which are considered as controls). On the contrary, the men who had problems with the number or concentration of sperms had low levels of zinc, it was found that most of these cases were in the third, fourth, and fifth levels. While it was found that the people who had higher zinc levels than the controls had weakness in the effectiveness of sperm (sperm motility). Infertile men can benefit greatly from taking zinc supplements because it improves the quality of their sperm (Figure 4.18 and Table 4.33). The following studies support these explanations:

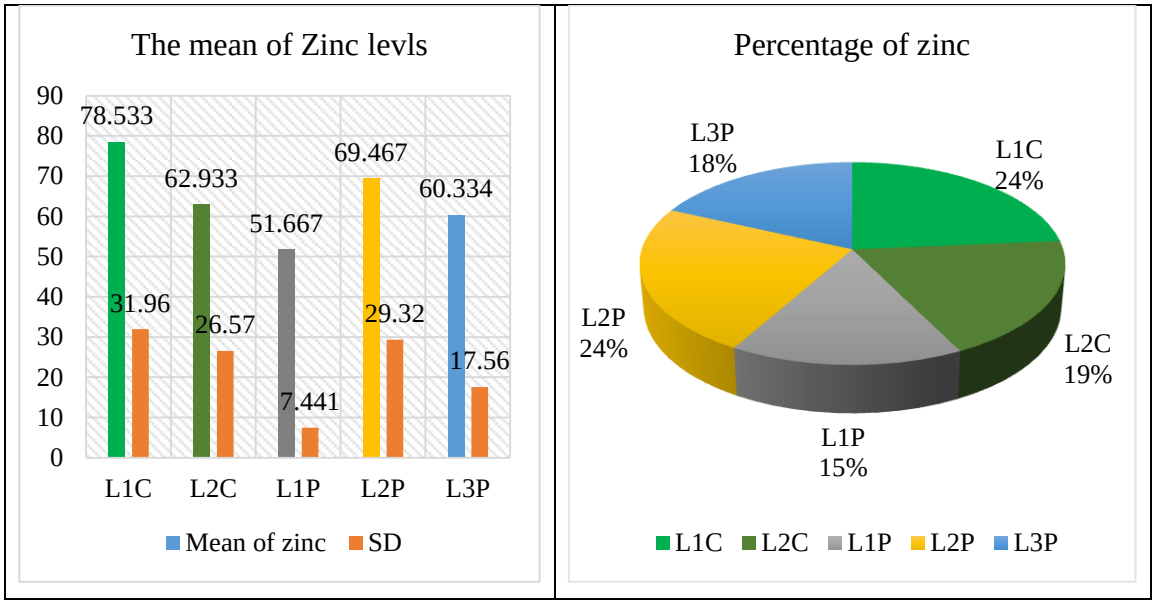

Figure 4.18 The mean for zinc between study levels in men

- **Comparison with previous studies**

The zinc deficiency was a common factor among patients seeking assisted reproductive treatment. Overall, most of the symptoms of zinc deficiency in men are similar to those experienced by women who have undergone infertility treatments. Male factor infertility can be caused by zinc deficiency in men, who are more likely to have low sperm counts and quality. Zinc is not only important for men's fertility, but for their health generally too (Wong *et al.* 2000).

Infertile males had lower zinc concentrations in their seminal plasma compared to healthy males, according to the results of the meta-analysis in men (this interpretation is consistent with what we mentioned regarding the effect of zinc on the concentration and number of Sperm). These researchers also found that zinc supplementation that is within the normal level significantly and understandably increases the volume of semen in men with low sperm count and sperm motility, and thus effect on the percentage of healthy sperm production in men. Zinc levels in the seminal plasma of male infertiles were found to be significantly lower than those in normal males, according to the results of this study (Zhao *et al.* 2016).

The study concluded that high levels of Zn^{2+} in the seminal plasma of infertile men reduce or weaken the movement of sperm in men (This study is matching with our study, and that removing Zn^{2+} and returning to the normal level enhances the movement of sperm (Allouche-Fitoussi and Breitbart).

For men who are infertile, zinc has both physiological and pathophysiological roles, and it has the potential to influence sperm parameters and fertilization. Zinc contains many unique and beneficial properties for men's health. Antioxidants and their types are one of them. Also, elevated levels of ROS in the seminal plasma of infertile men affect the zinc content in the seminal plasma and thus increase the chance of male infertility and can be affected by changes in zinc levels in sperm physiology in men. Zinc is a balancer for male hormones, which helps sex hormones such as testosterone and others in men. Zinc deficiency can impede spermatogenesis in men (and this explanation is consistent with what we found in the interpretation of the results of our study) (Fallah *et al.* 2018).

- **Post HOC tests for zinc**

When conducting post-tests (Table 4.40) of zinc, the results indicated that the significant differences were between L1C vs L1P, L1C vs. L3P and L1P vs L2P according to the HSD level, the L1C vs L1P value was more affected by zinc.

Table 4.40 The post-tests for zinc between in study effect of PA with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Zinc	L1C	L2C	1.60000	8.8725	0.406
		L1P	**26.86667***	**8.8725**	**0.028**
		L2P	0.06667	8.8725	1.000
		L3P	18.20000*	8.8725	0.023
	L2C	L1P	11.26667	8.8725	0.710
		L2P	15.53333	8.8725	0.410
		L3P	2.60000	8.8725	0.998
	L1P	L2P	26.80000*	8.8725	0.028
		L3P	8.66667	8.8725	0.865
	L2P	L3P	1.13333	8.8725	0.256

- **ROC curve of zinc**

Higher test result variables in the ROC curve indicate stronger evidence for positive actual state and less evidence for negative case. The negative area and the positive area for age are shown in Table 4.41 and Appendix 18. The positive actual state is ≤61.

Table 4.41 The information the ROC curve for zinc

Variable	Zinc
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.745
The Error of Intention [a]	0.0589
Confidence interval of 95 percent [b]	0.631 to 0.839
The z-score	4.159
Perceived importance P (Area=0.5)	<0.0001
The Youden (detecting disease) index	
Indicator J of Youden index	0.1562
Adjoining criteria	≤61
Sensitivity- Positive area	70.45
Specificity- Negative area	45.16

4.5.5 The study of calcium in an infertility

- **Calcium with levels of semen according to count of sperm**

When studying calcium, we found that there is a statistical significance between the levels of patients (LP1 = 17 %, LP2 = 8 %, and LP3 = 9 %) when compared with the levels of controls (LC1 = 35 % and LC2 = 31 %) at P = 0.031. The study concluded that calcium increases the chance of trace elements absorption or helps with their absorption in infertile men, where the levels of some trace elements in the first and second levels (which are considered as controls for the study) were within the normal level and calcium was close to (9.962) and the level of phytate acid is low. Conversely, calcium levels decreased at levels fourth and fifth, trace element deficiencies increased, and phytate acid levels were high. This leads us to the interpretation that calcium can act as

an inhibitor of the effect of phytate acid in the human body (Figure 4.19 and Table 4.33).

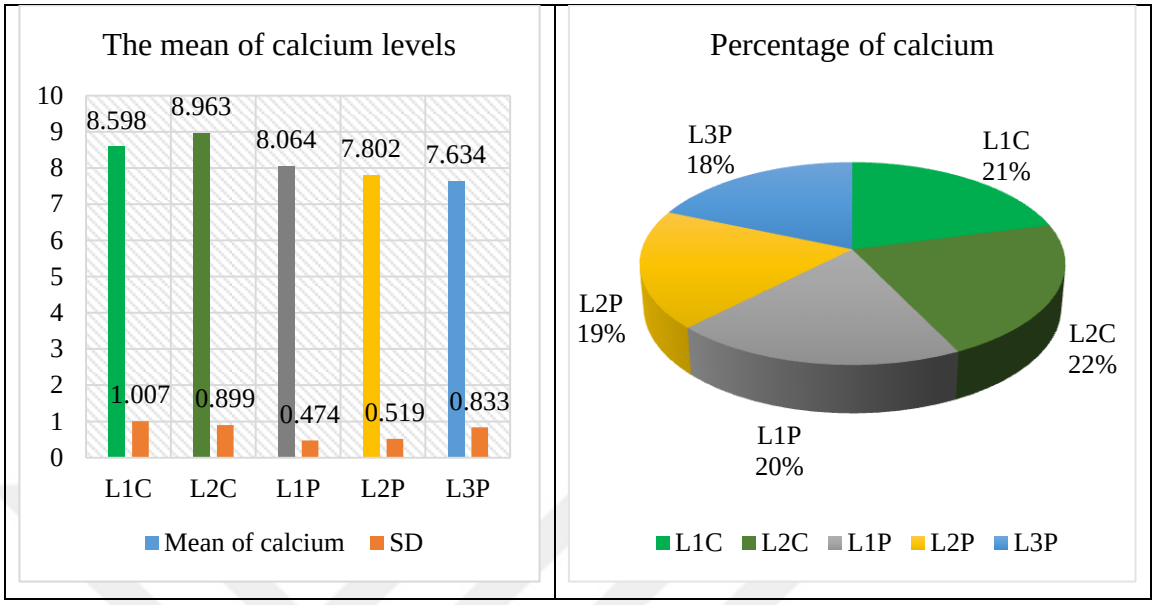

Figure 4.19 The mean for calcium between study levels in men

- **Comparison with previous studies**

Stevenson et al., recommended taking calcium supplements or foods containing calcium after a period of time or several hours after eating wheat bran cereal or fiber-rich plant foods that contain phytate acid (Stevenson *et al.* 2012).

- **Post Hoc tests for calcium**

When conducting post-tests (Table 4.42) of calcium, the results indicated that the significant differences were between L1C vs L2P, L1C vs L3P, L2C vs L1P, and L2C vs L2P according to the HSD level, the L2C vs L2P was more affected by calcium, The averages indicated that the effect begins after a period of time and the development of the disease and is likely due to taking the treatments significantly and in some cases not consulting the treating physician.

Table 4.42 The post-tests for calcium between in study effect of phytic acid with infertility men

Dependent Variable	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.
Calcium	L1C	L2C	0.36467	0.2829	0.699
		L1P	0.53467	0.2829	0.332
		L2P	0.79667*	0.2829	0.048
		L3P	0.96533*	0.2829	0.009
	L2C	L1P	0.89933*	0.2829	0.018
		L2P	1.16133*	0.2829	0.001
		L3P	**1.33000***	**0.2829**	**0.000**
	L1P	L2P	0.26200	0.2829	0.886
		L3P	0.43067	0.2829	0.552
	L2P	L3P	0.16867	0.2829	0.975

- **ROC Curve of calcium**

The higher the values of the test result variables on the ROC curve for serum calcium, the more evidence points to a positive actual state, while the lower the values point to a negative case. The negative area and the positive area for age are shown in Table 4.43 and Appendix 19. The positive actual state is ≤7.74.

Table 4.43 The information the ROC curve for calcium

Variable	Calcium
Classification variable	Levels of study
Sample size	300
Positive level [a]	120 (40.00 %)
Negative level [b]	180 (60.00%)
Disease prevalence (%)	58.7%
Area under the curve-ROC	
The ROC curve's slope (AUC)	0.735
The Error of Intention [a]	0.0581
Confidence interval of 95 percent [b]	0.621 to 0.831
Perceived importance P (Area=0.5)	0.0439
The Youden (detecting disease) index	
Indicator J of Youden index	0.4523
Adjoining criteria	≤7.74
Sensitivity-Positive	61.36
Specificity- Negative	83.87

4.6 The Correlation etween phPytic acid and Inhibin B with Levels tuduy

Table 4.44 Correlation between INHB and phytic acid with author biochemistry

Correlations			
Parameters	Correlation information	PA	INHB
Age	Correlation Coefficient	0.304**	-0.444**
	Sig. (P-value)	0.008	0.0000
Weight	Correlation Coefficient	0.118	-0.162**
	Sig. (P-value)	0.314	0.001
BMI	Correlation Coefficient	0.153	-0.434**
	Sig. (P-value)	0.190	0.0000
PA	Correlation Coefficient	-	-0.629**
	Sig. (P-value)	.	0.0000
INHB	Correlation Coefficient	-0.629**	-
	Sig. (P-value)	0.000	.
Tg	Correlation Coefficient	-0.028	0.155
	Sig. (P-value)	0.812	0.185
Tc	Correlation Coefficient	-0.318**	0.341**
	Sig. (P-value)	0.005	0.003
HDL	Correlation Coefficient	-0.211	0.203
	Sig. (P-value)	0.069	0.080
LDL	Correlation Coefficient	-0.021	-0.142
	Sig. (P-value)	0.861	0.224
VLDL	Correlation Coefficient	-0.086	0.191
	Sig. (P-value)	0.464	0.100
LH	Correlation Coefficient	0.142	-0.385**
	Sig. (P-value)	0.224	0.001
FSH	Correlation Coefficient	0.439**	-0.514**
	Sig. (P-value)	0.000	0.000
Testo	Correlation Coefficient	-0.447**	0.296**
	Sig. (P-value)	0.000	0.010
RBS	Correlation Coefficient	0.003	0.183
	Sig. (P-value)	0.982	0.117
P	Correlation Coefficient	-0.481**	0.584**
	Sig. (P-value)	0.000	0.000
Mg	Correlation Coefficient	0.139	-0.039
	Sig. (P-value)	0.236	0.743
Zn	Correlation Coefficient	-0.649**	0.081
	Sig. (P-value)	0.003	0.492
Ca	Correlation Coefficient	-0.355**	0.441**
	Sig. (P-value)	0.002	0.000
Iron	Correlation Coefficient	0.215	-0.009
	Sig. (P-value)	0.064	0.941

4.6.1 Correlation between phytic acid with author biochemistry

To understand and take more information about phytic acid, we did a Pearson test to see if there are correlations between it and some chemical parameters, and thus interpret the

results clinically. When doing the Pearson test, we found that there is a positivly correlation between phytic acid with age $r = 0.304^{**}$ Sig. 0.001 , which is explained by the accumulation of phytates in the human body with age due to its dependence on plant products, where the percentage of phytic acid increased more and there were significant differences in the averages in 15 samples (2 out of The first level of patients, 4 of the second level of patients, and 9 of the third level of patients) out of 150 samples, and because the percentage is considered low compared to the number of samples, it was referred to the work of a special and detailed study for vegetarians in the recommendations section of this research. No correlation between phytic acid levels and weight was found in Bison's test, which indicates that plant products do not increase weight, and the value of $r = 0.314$ at Sig. = 0.118. Also, was no correlation between phytic acid with LH and BMI, the result was $r = 0.142$ at Sig. = 0.224 and $r = 0.153$ at Sig. = 0.190 (Table 4.44).

A negative correlation was reached between phytic acid with inhibin B $r = -0.629^{**}$ at Sig. = 0.000, which can be explained in the following way the higher the phytic acid, the lower the hormone inhibin B. Since Inhibins are members of the super-amyloid (GFβ) proteins or proteins in the human body, it is because of this binding that FSH and LH secretion is inhibited or stimulated. As the follicle-stimulating hormone (also known as follicle-stimulating hormone or simply FSH), the pituitary gland secretes FSH along with other hormones like luteinizing hormone (LH) to keep men's gonads healthy, with the testicles being the primary gonads. (on the testicles and stimulate the production of sperm), so this explains that the problems in the semen files, where the increase in phytic acid negatively affects the hormone inhibin B, and according to the role which plays it, as mentioned, it greatly affects the levels of FSH and LH Thus, it is a direct cause of the development or occurrence of infertility in men.

The Pearson test also indicated that there is a Correlation between phytic acid and testosterone hormone $r = -0.447^{**}$ at Sig. = 0.001. Since phytic acid affects the hormone inhibin B, as a result, testosterone will be affected, and this conclusion can be summarized by the following: LH is also synthesized in the adenohypophysis and acts with FSH to promote ovulation and secretion of androgens and progesterone. It initiates and maintains the second (secretory) phase of the mammalian estrus and menstrual cycle. In males, it stimulates the development and functional activity of testicular

Leydig cells and testosterone production. That explains to us the correlation of phytic acid with testosterone hormone.

The link between zinc and calcium (r = -0.649** at Sig. = 0.003 and r = -0.355** at Sig. = 0.002). We can say that phytates (IP6) and their low esters (IP5, 4, 3), although they are anti-nutritional in themselves, lead to a direct impact on calcium and zinc levels, and this is explained by the use of phytase enzyme in some animal food products. Up to two to four times that of normal commercial use releases Ca and, P. Zinc has a role in the synthesis and actions of many hormones via zinc transcription factors. Low levels of testosterone are linked to zinc deficiency. Thus, the quality of the semen and the changes in the semen profiles are affected.

4.6.2 Correlation between INHB andauthorr biochemistry

The Pearson test was used in the same experiment to see if there were any correlations between Inhibin B and the other tests and, if so, to assess its usefulness in the diagnosis of male infertility. There was a negativly correlation between age and weight (r = -0.444** at Sig. = 0.0001 and r = -0.162** at Sig. = 0.010 respectively) with inhibin B, as, with increasing age, infertility or weak sperms began to appear, and according to the questionnaire that was conducted, nutrition and the misuse of medicines led to an increase in the rate of infertility, and of course, this problem parallels the increase in weight. Therefore, we believe it is necessary to conduct a study that looks at the relationship between people who take medications or treatments for weight loss and to understand the mechanism of the effect of those drugs on the enzymes involved in the process of spermatogenesis during the period of the taking it.

There was a link between INHB (r = -0.629** at Sig. = 0.000) with phytic acid, which was explained in section (4.6.1). While there was no association between inhibin B and zinc in contrast to its association with phytic acid. There was a link between phytic acid and calcium (r = 0.441** at Sig. = 0.0021).

4.6.3 The correlation between phytic acid with semen parameters

The results of the study of the correlations between semen parameters and phytic acid showed a direct effect on the number and activity of sperm, as shown in Table 4.45. Also, for inhibin B hormone, the results indicated a direct effect on the number, shape and activity of sperms, as shown in Table 4.46.

Table 4.45 Correlation between phytic acid with semen parameters

Parameters	*r*-value	*P-Value*
Count of sperm	-0.572**	0.000
Morphology of sperm	0.0310	0.729
Active of Sperm	-0.405**	0.001
Volume	0.193	0.371

Table 4.46 Correlation between INHB with semen parameters

Parameters	*r*-value	*P-Value*
Count of sperm	0.858**	0.030
Morphology of sperm	0.541**	0.042
Active of Sperm	0.631**	0.001
Volume of semen	0.05800	0.804

4. Conclusion and Recommendation

- **Conclusion**

Many plant-based foods contain phytic acid, which is a substance found in the body in small amounts. Known as IP6, it is also known as inositol hexaphosphate or IP6. This acid is the primary way that many plants, such as beans, seeds, and nuts, store phosphorus.

The research that we conducted on a group of men found that male fertility begins at an early age of 11-13 years, while it appears to decline or, so to speak, begins to decrease from the age of 68 and above. the average age of male infertility was approximately 28 to 35 years, and the poor quality of the sperm was a major factor.

The results indicate the importance of the hormone LH, because it increases testosterone production in the testicles (stimulates the Leydig cells in the testicles to produce testosterone in males.

Men with high levels of luteinizing hormone can suffer from infertility, because the hormone LH directly affects the functions of the male reproductive organs, which is evident in the third level of infertile men in our study. As for the low levels of the hormone LH, we found that it affects the volume of semen and thus limits the movement of sperm in males, which leads to the incomplete fertilization process.

Depending on the results, phytic acid can be harmful to testosterone levels in men and through a direct effect on semen parameters. Incomplete cooking of vegetable products.

There was a percentage (63%) of patients who appeared to have changeslevels of phytate acid and problems of phosphorus levels, and the problem in the amount of phosphorus absorbed was associated with the effectiveness and motility of sperms.

As an anti-nutrient, phytic acid blocks the body's ability to absorb certain minerals like zinc, iron, and others. Phytic acid, which is commonly found in vegetarian diets, has been shown to increase the risk of developing problem of spermatogenesis in men. It forms phytates when combined with other minerals, and since a person cannot secrete or produce enzymes that can break down phytates (the phytase enzyme), and therefore a person cannot absorb its nutrients in the body or a problem in the availability of votive items.

In order to continue the reproductive process in humans, especially men, some of the trace elements play an important role, as zinc and other trace minerals and steroid hormones play a role in the formation of sperm, as well as some protein hormones replace many important functions in the process of sperm formation This, and thus the continuation of the reproductive process in humans. Zinc is necessary for sperm quality in men. Zinc, which exists with high concentrations in the semen of men, plays roles in the functional properties of sperm such as movement, effectiveness, natural shape and even the number of total sperm. Zinc deficiency can affect not only semen parameters but also semen liquidity.

The study concluded that calcium increases the chance of trace elements absorption or helps with their absorption at infertile men, where the levels of some trace elements in the first and second levels (which are considered as controls for the study) were within the normal level and calcium was close to (9.962) and the level of phytate acid is low. Conversely, calcium levels decreased at levels fourth and fifth, trace element deficiencies increased, and phytate acid levels were high. This leads us to the interpretation that calcium can act as an inhibitor of the effect of phytate acid in the human body. In the study, the relationship of zinc, phosphorous, phytic acid, inhibin B and other chemical variables in a striking way to semen quality (the characteristics of semen) was controversial.

Because food is mainly based on plant products that contain phytate. There is a need for more studies on the effect of phytic acid on humans.

- **Recommendation**

1. Study the role of phytate in activating or inhibiting the enzyme trypsin, which helps digestion. To identify the chemical role that the enzyme plays in excluding or reducing cholesterol from the body.
2. Various methods must be followed to reduce the level of phytic acid in foods, such as soaking seeds, legumes and grains well because soaking reduces phytic acid in food.
3. More studies should be done with the nutritionist in order to reach better results
4. The relationship between sperm counts or concentration and low levels of zinc with increase phytic acid levels must be studied in order to know and understand the mechanism and why low zinc levels affect sperm count.
5. Study of the activity and inhibition of phytic acid with glucagon and insulin through biochemistry laboratories.
6. Study of the levels of copper with phytate acid in seminal plasma of men in the azospermia group and in the oligozoospermic group and compared to the control group.
7. A study of the relationship between phytate acid and magnesium, in order to evaluate the possible role of the relationship between them in male infertility when changes in phytic levels.
8. Study of the relationship between selenium and phytate acid in infertile men.
9. Conducting post-studies (supplements of magnesium) for magnesium Changes in magnesium levels may lead to a short period of inactivity in the levels of steroid hormones in men.
10. The relationship between sperm motility and activity with zinc should be studied in order to understand the mechanism by which high levels of zinc can affect their motility.
11. Study of zinc supplementation in with infertility levels divided according to our study where zinc supplementation can significantly increase or enhance sperm quality in infertile male(s)
12. Study of the effect of high levels of reactive oxygen species in the seminal plasma of infertile men on the zinc content in order to find out how much affected by the changes in zinc levels in sperm physiology in men.

13. Conducting a test of the effect of IP6 on iron overload with oxidative stress in the laboratory and studying the levels of iron in the body of men with infertility problems.
14. Conducting a wide and comprehensive survey on vegetarians in order to know the percentage of phytate acid accumulation in them

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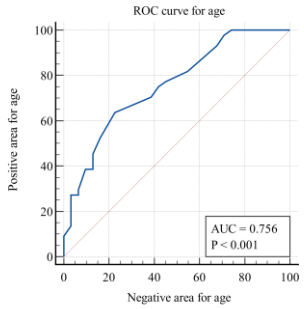
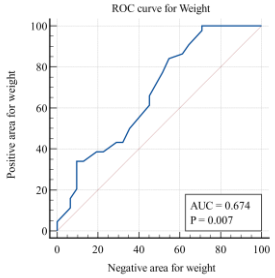
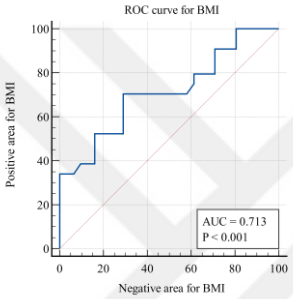
APPENDICES



APPENDIX 1 The graph ROC curve for age



APPENDIX 2 The graph ROC curve for weight



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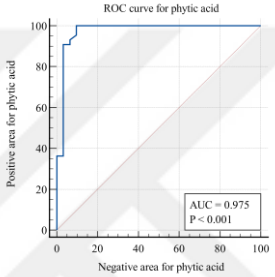
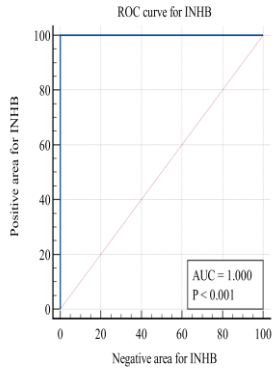
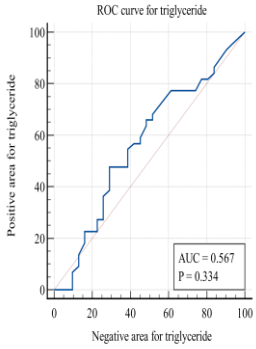


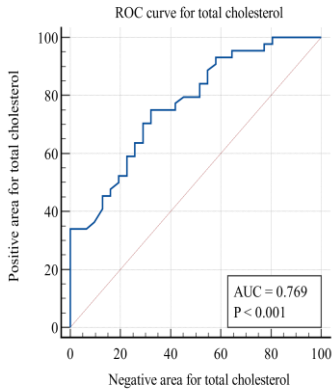
APPENDIX 4 The graph ROC curve for phytic acid



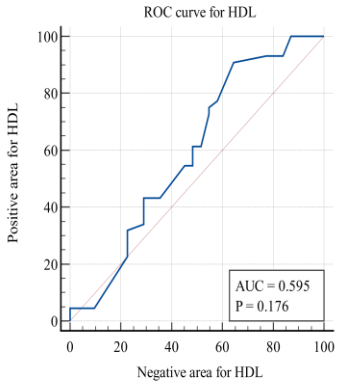
APPENDIX 5 The graph ROC curve for inhibin B



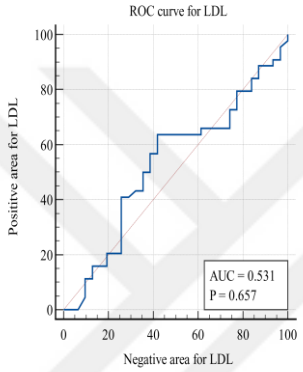
APPENDIX 6 The graph ROC curve for triglyceride



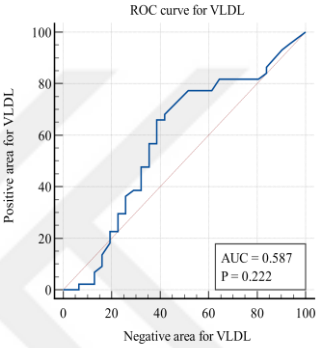
APPENDIX 7 The graph ROC curve for Tc



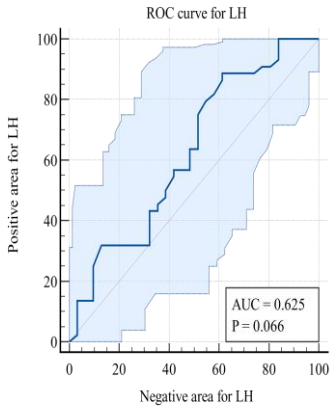
APPENDIX 8 The graph ROC curve for HDL



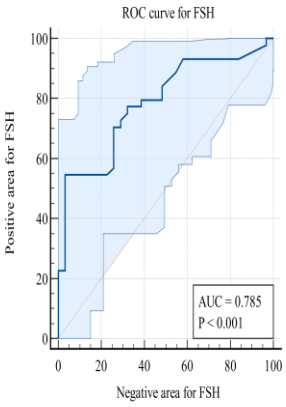
APPENDIX 9 The graph ROC curve for LDL



APPENDIX 10 The graph ROC curve for VLDL



APPENDIX 11 The graph ROC curve for LH



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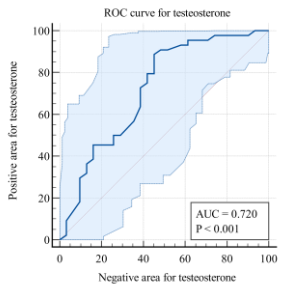
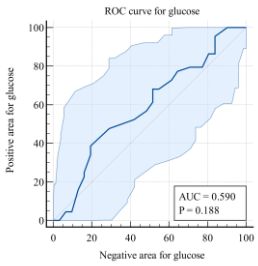
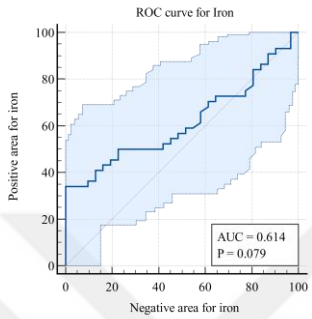


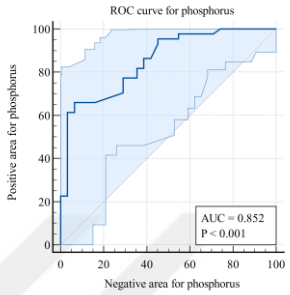
APPENDIX 13 The graph ROC curve for teseosterone



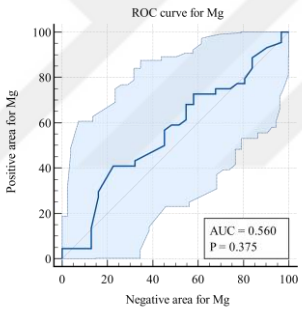
APPENDIX 14 The graph ROC curve for glucose



APPENDIX 15 The graph ROC curve for iron



APPENDIX 16 The graph ROC curve for phosphorus



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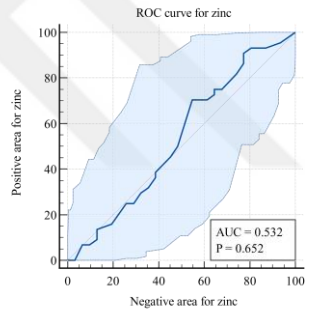
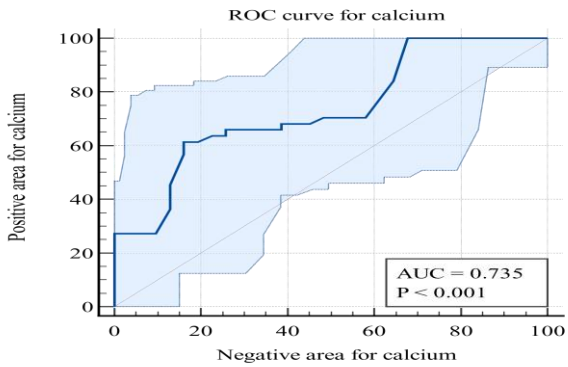


APPENDIX 18 The graph ROC curve for zinc



APPENDIX 19 The graph ROC curve for calcium

CURRICULUM VITAE

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Undergraduate	Al-Mamoun University Faculty of Medical Technology Department of medical techniques	2013-2014

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Nayyef, H. A. and Shubbar, H. H. K. 2020. The study of relation and the effect of IL-1, IL-6 and some biochemical markers on type-1 and type2 diabetes mellitus development risk. 7thinternational Conference on Computational and Experimental Science and Engineering (ICCESEN-7), pp, 298-300. Antalya.