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PRODUCTION OF CHOCOLATE SPREAD WITH PUMPKIN SEED

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PRODUCTION OF CHOCOLATE SPREAD WITH PUMPKIN SEED

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February 2024

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Omar Abbas Fadhil AL-SALIHI

ABSTRACT

PRODUCTION OF CHOCOLATE SPREAD WITH PUMPKIN SEED

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In this thesis study, it was aimed to develop a production method for breakfast chocolate spread containing pumpkin seeds which have beneficial content in terms of health. For this purpose, roasting process of pumpkin seeds was carried out at different temperatures and times before production. For this purpose, 3 different temperature conditions (low: 120 °C, medium: 150 °C and high: 180 °C) and 2 different time conditions (short: 20 minutes and long: 40 minutes) were selected. Pumpkin seeds roasted at different temperatures and times were used 13% in chocolate spread production. Physical, chemical, textural, rheological and sensory analyses were carried out on the chocolate spreads. While roasting temperature and roasting time were found to be effective on AOS, water activity, Casson plastic viscosity, Windhab actual yield stress, infinite viscosity, colour parameters (L^*, a^*, b^*), sensory pumpkin seed aroma and appperance ($p<0.05$). However, pumpkin seed roasting temperature and time were found to be ineffective on textural parameters (firmness, spreadability, stickiness and cohesiveness), particle size distribution parameters (d(0.1), d(0.5), d(0.9), D[3.2], D[4. 3] and span), shear stress, apparent viscosity, Casson yield stress, Windhab linear yield stress, characteristic shear rate, sensory general odour, pumpkin seed odour, general flavour, spreadability, consistency and colour. In this study, sensory parameters were the main determinants. HS and ML spreads scored higher for general odour, pumpkin seed odour, general flavour and pumpkin seed aroma. The HL spread was found to be unsuccessful due to burnt flavour defect. It can be suggested that pumpkin seeds can be applied in the production of spreadable chocolate by roasting at 150 °C for 40 minutes or 180 °C for 20 minutes or by using roasting temperatures and times close to these temperatures and times.

2024, 63 pages

Keywords: Pumkin seed roasting, Chocolate spread with pumpkin seed, Texture, Rheology

ÖZET

KABAK ÇEKİRDEKLİ SÜRÜLEBİLİR ÇİKOLATA ÜRETİMİ

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Bu tez çalışmasında sağlık açısından faydalı içeriğe sahip kabak çekirdeği içeren kahvaltılık sürülebilir çikolata üretim yöntemi geliştirmek amaçlanmıştır. Bu amaçla kabak çekirdeklerinin üretim öncesi farklı sıcaklıklar ve sürelerde kavurma işlemi gerçekleştirilmiştir. Bunun için 3 farklı sıcaklık derecesi (düşük: 120 °C, orta: 150 °C ve yüksek:180 °C) ve 2 farklı süre (kısa:20 dakika ve uzun:40 dakika) koşulları seçilmiştir. Farklı sıcaklık ve sürelerde kavrulan kabak çekirdekleri sürülebilir çikolata yapımında %13 oranında kullanılmıştır. Üretilen sürülebilir çikolatalarda fiziksel ve kimyasal, tekstürel, reolojik ve duyusal analizler gerçekleştirilmiştir. Kavurma sıcaklık ve süresi HYA, su aktivitesi, Casson plastik viskozite, Windhab gerçek akma gerilimi, sonsuz viskozite, renk parametreleri (*L, a, b*), duyusal kabak çekirdeği aroması ve görünüş üzerine etkili bulunurken ($p<0.05$), tekstürel parametreler (sıkılık, sürülebilirlik, yapışkanlık ve tutunabilirlik), parçacık boyut dağılım parametreleri (d(0.1), d(0.5), d(0.9), D[3.2], D[4.3] ve span), kayma gerilimi, görünür viskozite, Casson akma gerilimi, Windhab lineer akma gerilimi, karakteristik kayma hızı, duyusal genel koku, kabak çekirdeği kokusu, genel lezzet, sürülebilirlik, kıvam ve renk üzerine ise etkisiz bulunmuştur. Bu çalışmada, duyusal parametreler ana belirleyici unsur olmuştur. HS ve ML sürülebilir çikolataları genel koku, kabak çekirdeği kokusu, genel lezzet ve kabak çekirdeği aroması bakımından daha yüksek puan almıştır. HL sürülebilir çikolatası ise yanık lezzet kusuru nedeniyle başarısız bulunmuştur. Kabak çekirdeği, 150 °C'de 40 dakika veya 180 °C'de 20 dakika kavurma veya bu sıcaklık ve sürelere yakın kavurma sıcaklık ve süreleri kullanılarak sürülebilir çikolata üretiminde uygulanabileceği önerilebilir.

2024, 63 sayfa

Anahtar Kelimeler: Kabak çekirdeği kavurma, Kakaolu kabak çekirdeği kreması, Tekstür, Reoloji

PREFACE AND ACKNOWLEDGEMENTS

This study titled "Production of Chocolate Spread with Pumpkin Seed" was prepared between 2021-2023 and submitted to Çankırı Karatekin University The Graduate School Of Natural And Applied Sciences as a "Master's Thesis". The aim of the thesis is to determine the roasting temperature and time of pumpkin seeds with the most suitable technological properties for the production of sensory acceptable and chocolate spread with pumpkin seed. Physical, textural, rheological and sensory analyses of the produced chocolate spreads were carried out.

I would like to thank my supervisor Assoc. Prof. Dr. Oğuz AYDEMİR, who always helped me with his knowledge and understanding in the determination of the thesis subject and laboratory studies.

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Omar Abbas Fadhil AL-SALIHI

Çankırı-2024

CONTENTS

ABSTRACT i
ÖZET iii
PREFACE AND ACKNOWLEDGEMENTS v
CONTENTS vi
LIST OF SYMBOLS viii
LIST OF ABBREVIATIONS ix
LIST OF FIGURES x
LIST OF TABLES i
1. INTRODUCTION 1
2. LITERATURE REVIEW 3
3. MATERIAL AND METHOD 20
3.1 Material 20
3.2 Method 20
3.2.1 Pumpkin seed roasting conditions 20
3.2.2 Processing of chocolate spread with pumpkin seed 20
3.2.3 Accelerated oil separation 22
3.2.4 Texture analysis 22
3.2.5 Rheological analyses 23
3.2.6 Particle size distribution 24
3.2.7 Water activity 24
3.2.8 Colour analysis 25
3.2.9 Sensory analysis 26
3.2.9 Statistic analysis 27
4. RESULTS AND DISCUSION 29
4.1 AOS Results 29
4.2 a_w Results 30
4.3 Textural Characteristics 32
4.4 PSD Analysis Results 34
4.5 Rheological Analysis Results 36
4.6 Colour Analysis Results 41

4.7 Sensory Analysis Results 43
5. CONCLUSION **47**
REFERENCES **50**
CURRICULUM VITAE **63**

LIST OF SYMBOLS

$\tau 0$	Actual yield stress
AMP	Ammonium phosphatide
$\dot{\gamma}^*$	Characteristic shear rate
R2	Coefficient of determination
K	Consistency coefficient
°C	Degree centigrade
g	Gram
Hz	Hertz
$\eta\infty$	Infinite viscosity
kg	Kilogram
L*	Lightness
$\tau 1$	Linear yield stress
G″	Loss module
mL	Millilitres
mm	Millimetres
N	Newton
Pa	Paskal
ηpl	Plastic viscosity
a*	Redness-greenness
rpm	Revolutions per minute
s	Saniye
$\dot{\gamma}$	Shear rate
τ	Shear stress
p	Statistical significance
G′	Storage module
a_w	Water activity
b*	Yellowness-blueness

LIST OF ABBREVIATIONS

GLP-1	Glucagon-like peptide-1
LUTS	Lower urinary tract symptoms
PPe-H	Polysaccharide hydrolysate
PUFAs	Polyunsaturated fatty acids
PBPP	Protein-bound polysaccharide

LIST OF FIGURES

Figure 1.1 Data on pumpkin planting and production in Türkiye from 1994 to 2018. 3
Figure 2.1 The mechanism of pumpkin polysaccharide hydrolysate in the treatment of DM-2. 12
Figure 2.2 Antidiabetic synergistic effect of both pumpkin polysaccharide and puerarin 12
Figure 3.1 The oven used in roasting. 21
Figure 3.2 Centrifuge device used in the thesis 22
Figure 3.3 Texture analyzer used in thesis 23
Figure 3.4 Water activity device used in the study 25
Figure 3.5 Colour device used in the study 25
Figure 3.6 Photographs of the produced chocolate spreads with pumpkin seed 27
Figure 3.7 Photographs of the roasted pumpkin seeds 28
Figure 4.1 AOS values of chocolate spread with pumpkin seed (%) 30
Figure 4.2 a_w values of chocolate spread with pumpkin seed 32
Figure 4.3 Shear stress variation versus shear rate of chocolate spreads 37
Figure 4.4 Shear rate versus viscosity variation of chocolate spreads 38
Figure 4.5 Storage (G′) and loss (G″) modulus values against frequency of short-time roasted chocolate spreads. 40
Figure 4.6 Storage (G′) and loss (G″) modulus values against frequency of long-time roasted chocolate spreads 41
Figure 4.7 Sensory analysis results of chocolate spreads with pumpkin seed 46

LIST OF TABLES

Table 2.1 The percentages of bioactive components in pumpkin seeds (food value per 100 g) (Patel *et al.* 2017) 5
Table 2.2 The percentages of bioactive components in pumpkin seeds (food value per 100 g) (Patel, *et al.* 2017) 6
Table 2.3 The content of phenolic acids (mg / 100 g) and fatty acids (%) in pumpkin seed oil (Rezig *et al.* 2012). 7
Table 2.4 The amount of tocopherol in milligrams per 100 grams of pumpkin (Cucurbita maxima var. "Béjaoui") seed oil (Rezig *et al.* 2012). 8
Table 3.1 Experiment plan 20
Table 3.2 Sensory Score Card 26
Table 4.1 AOS Values of Chocolate Spread with Pumpkin Seed (%) 29
Table 4.2 a_w Values of Chocolate Spread with Pumpkin Seed 31
Table 4.3 Textural Parameters of Chocolate Spread with Pumpkin Seed 33
Table 4.4 PSD Parameter values of Chocolate Spread with Pumpkin Seed 34
Table 4.5 Shear stress and apparent viscosity values of spread samples at 30 s-1 shear rate 37
Table 4.6 Casson model parameters of chocolate spreads 38
Table 4.7 Windhab model parameters of chocolate spreads 39
Table 4.8 CIE Colour parameters of Chocolate Spread with Pumpkin Seed 42
Table 4.9 Sensory Parameter Values of Spread Samples 45

1. INTRODUCTION

In recent years, seeds and nuts have received increasing attention due to the high nutritional and health-promoting value of their bioactive components. One example of this is pumpkin seeds. Pumpkin with oil seeds belongs to the family Cucurbitaceae. Although there are many different species cultivated worldwide, the most commercially important are *Cucurbita pepo* (the most common), *C. maxima, C. moschata, C. mixta* and *C. stilbo*. Worldwide, pumpkins are cultivated for many different purposes, such as commercial, decorative, and agricultural uses. The involvement of pumpkin in many traditional treatment systems has attracted the interest of researchers. Although pumpkin seeds are largely regarded as agro-industrial waste, they serve as food stores with important nutritional properties. The consumption of pumpkin seeds is increasing in many countries due to their ethnomedical benefits. Often the seeds are consumed directly as a snack after roasting and salting. They are also used as food additives in the baking industry (Dotto and Chacha 2020). In recent years, pumpkin seed oil has also started to take place in the market due to its health benefits. Pumpkin seeds are generally consumed as snacks or additives to bakery products in Türkiye.

The market for spreads is of great value in Türkiye and in the world. Recently, studies on spreadable products based on different seeds or nuts have increased considerably. The use of pumpkin seeds in this product group, which is especially consumed by children and young people, will not only contribute to the nutrition of the society but also increase the product diversity by bringing a new product to the market. It will also encourage the economic development of pumpkin and pumpkin seed growers.

With this in mind, the use of pumpkin seeds in the production of chocolate spread was tested in this thesis. For this purpose, roasting process was applied to pumpkin seeds at different temperatures and times before production. Pumpkin seeds roasted at different temperatures and times were used in the production of 13% chocolate spread. The effects of roasting

temperature and time on the physical, chemical, textural, rheological, and sensory properties of chocolate spreads were investigated.

2. LITERATURE REVIEW

Many edible plants, including watermelon and squash, belong to the Cucurbitaceae family, and their seeds are frequently discarded as trash (Patel *et al.* 2017). Over 1,174,254 hectares are used to produce 15,057,448 tons of squash worldwide.

Pumpkin seed oil is used in meals (primarily as salad oil) and confectionary and bakery goods as well as in the pharmaceutical sector. In addition to this, pumpkin seeds are utilized as a source of vegetable oil and eaten as edible pumpkin. Türkiye produces 430,402 tons of pumpkin at a 2% percentage, compared to the world's 22,905,868 tons. Figure 1 depicts the production plan of pumpkin seeds in Türkiye from 1994 to 2018.

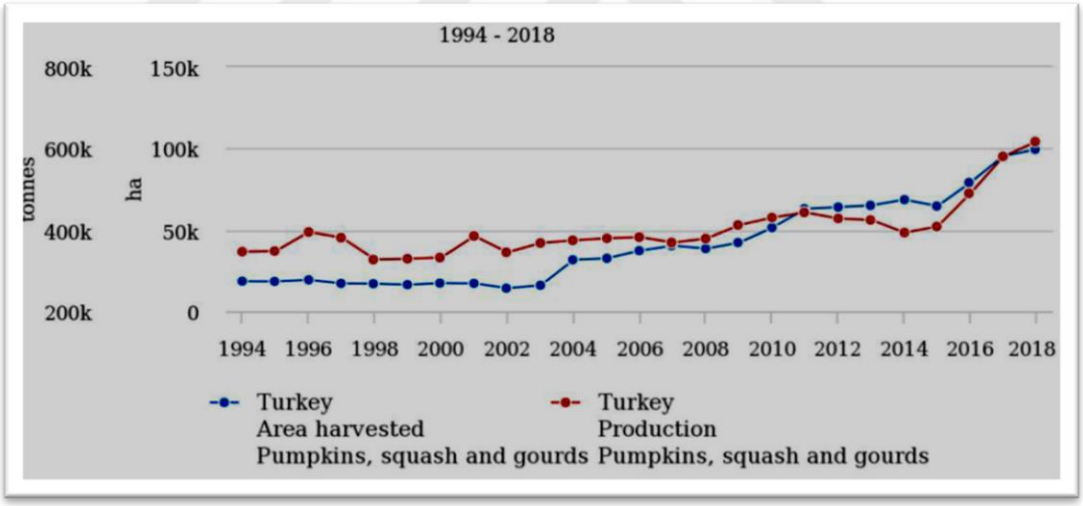


Figure 1.1 Data on pumpkin planting and production in Türkiye from 1994 to 2018

As a member of the Cucurbitaceae family, which also includes squash, pumpkin is a plant that is widely grown throughout most of the world. Its climate need is low, it grows readily in any place with moderate weather, and its abundance in functional components and other nutrients has been observed to be advantageous (Patel *et al.* 2017, Jeffrey *et al.*1990, Karrar *et al.* 2018). Because of its high nutrient content, Cucurbitaceae have been crucial to human

nutrition, particularly in equatorial regions where growing cultivation areas have led to considerable increases in consumption (Karrar *et al.* 2018). Pumpkin seeds are abundant in protein, carotenoids, β-carotene, and γ-tocopherol, as well as minerals such as iron, zinc, manganese, phosphorus, copper, and potassium. The concentration of γ-tocopherol, or vitamin E, is especially high (Murkovic and Hillebrand, 1996). It has been established that pumpkin seeds contain 15.9 mg of tocopherol per 100 grams (Ryan *et al.* 2007). Pumpkin seeds are informed to have about 16.84% crude fibre, 39.25% crude protein, and 27.83% crude fat (Alfawaz *et al.* 2004).

According to Soha *et al.* (2010) pumpkin seeds are a good source of trace minerals like zinc and natural tocopherols. Pumpkin seeds have been found to have higher concentrations of powerful phytochemicals such tocopherols, sterols, and lipochrome (carotenoids), which have been shown to have tremendous potential for immunomodulation, reproductive health, and therapeutic effects in a variety of disease situations. Other research indicates that consuming the entire pumpkin seed extract is strongly linked to a decrease in prostatic hyperplasia symptoms (Glew *et al.* 2006, Stevenson *et al.* 2007).

Numerous studies have also demonstrated pumpkin seeds' ability to arouse sexually (Gundidza *et al.* 2009). Several sources indicate that pumpkin seed oil has a high concentration of vitamin E (tocopherol). In addition to these, it is recognized to have additional health advantages, like preventing prostate enlargement. It has been observed that pumpkin seed oil possesses anti-microbial activity, despite its established health advantages. As a result, it works effectively to enhance the nutritional value of food compositions (Stevenson *et al.* 2007).

The mineral content of the seeds from ten different genotypes of pumpkins was found to be a substantial source of minerals in a prior study. Potassium (K), phosphorus (P), magnesium (Mg), sulphur (S), and calcium (Ca) were the most prevalent minerals in each genotype. While certain genotypes did not exhibit the presence of Al, Co, Cd, Cr, Pb, or Se minerals,

other minerals such as Mo, B, and Ni were present at lesser amounts. Genotype-1 was found to have the highest amount of K (10,332,631 ppm), which is significant for the homeostatic balance of bodily fluids. The most prevalent minerals were magnesium (Mg) and phosphorus (P), with values ranging from 1275.145–3938.163 ppm and 3569.690–9108.835 ppm, respectively (Seymen *et al.* 2016).

Table 2.1 The percentages of bioactive components in pumpkin seeds (food value per 100 g) (Patel *et al.* 2017)

Princples	**Nutritive value**	**Percentage of RDA(%)**
Energy	559 Kcal	28
Carbohydrates	10.71 g	8
Protein	30.23 g	54
Total fat	49.05 g	164
Cholesterol	0 mg	0
Dietary fiber	6 g	16
Vitamins		
Folate	58 µg	15
Niacin	4.987 mg	31
Pantothenic acid	0.750 mg	15
Pyridoxine	0.143 mg	11
Riboflavin	0.153 mg	12
Thiamine	0.27 mg	23
Vitamin A	16 IU	0.5
Vitamin C	1.9 µg	3
Vitamin E	35.10 mg	237

Many nations have used pumpkin seeds to produce oil or protein. Research from Yugoslavia, Austria, Hungary, and Nigeria has led to the understanding that the extraction procedure used to extract pumpkin seeds is the source of cooking oil (Tsaknis *et al.* 1997). Pumpkin seed oil production ensures the use of a renewable resource while adding value to agricultural by-products. The oil content of various pumpkin seed kinds has been found to vary depending on the variety, growth conditions, climatic factors, and agricultural factors (Stevenson *et al.* 2007). Pumpkin seed oil ranges in hue from dark green to brown. Because pumpkin seed oil has a high oil content, hydraulic cold pressing is typically used in industrial production of pumpkin seed oil (Fruhwirth *et al.* 2008). However, the extraction of oil from plant seeds by

A significant drawback of hydraulic cold pressing is the high residual oil content in the seed, which can range from 5 to 15% by weight (Temelli *et al.* 2009).

Table 2.2 The percentages of bioactive components in pumpkin seeds (food value per 100 g) (Patel *et al.* 2017)

Principles	Nutritive value	Percentage of RDA (%)
Minerals		
Sodium	7 mg	0.5
Potassium	809 mg	17
Calcium	46 mg	4.5
Copper	1.343 mg	159
Iron	8.82 mg	110
Magnesium	592 mg	148
Maganese	4.543 mg	198
Phosphorus	1233 mg	176
Selenium	9.4 µg	17
Zinc	7.81 mg	71
Phytonutrients		
Carotenet-β	9 µg	-
Crypto-xanthine –β	1 µg	-
Lutein-zeaxanthin	74 µg	-

The phytochemicals to maintain the ideal body temperature, concentrated energy reserves are provided by fats and oils. About 40 million tons of fats and oils are consumed annually by humans, indicating the significance of fats in a diet and their widespread use in daily life (Dhiman *et al.* 2009).

Table 2.3 The content of phenolic acids (mg / 100 g) and fatty acids (%) in pumpkin seed oil (Rezig *et al.* 2012)

Fatty acid	Composition
Palmitic(C16:0)	15.97±0.39
Palmitoleic(C16:1)	tr.
Stearic(C18:0)	4.68±0.56
Oleic(C18:1)	44.11±0.63
Linoleic(C18:2)	34.77±0.95
Linolenic(C18:3)	Tr
Arachidic(C20:0)	0.41±0.40
SAFA	21.07±1.19
MUFA	44.12±0.57
PUFA	34.78±0.85
Phenolic acid	
Protocatechuic acid	1.81±0.26
Caffeic acid	3.88±0.03
Syringic acid	7.96±0.13
Vanilic acid	2.46±0.37
p-coumaric acid	2.50±0.95
Ferulic acid	4.99±0.29
Unidentified compounds	*

Trace quantities (less than 0.2%), SAFA stands for saturated fatty acids, MUFA for monounsaturated fatty acids, and PUFA for polyunsaturated fatty acids.*: Compound identified, but sample concentration unknown, The values are the mean ± SD of three determinations.

About 30–50% of pumpkin seeds are made up of oil (El-Adaway *et al.* 2001), which is rich in a variety of bioactive substances such carotenoids, sterols, tocopherols, squalene, and unsaturated fatty acids (Younis *et al.* 2000, Rabrenović *et al.* 2014).

Pumpkin seed oil is mostly composed of the fatty acids palmitic, stearic, oleic, and linoleic. According to earlier research, different types of pumpkin seed oil used to produce oil have contents ranging from 10.3-11.7% palmitic, 4.1-5.4% stearic, 30.5-40.8% oleic, and 42.1-51.5% linoleic acid (Wenzel *et al.* 1987). According to Türkmen *et al.* (2015) and Seymen *et al.* (2016) the bioactive characteristics of pumpkin seeds the most well-known and abundant natural sources of tocopherols and phenolics, which are antioxidants, are plants. By

transforming lipid radicals into more stable products, they are regarded as the main antioxidants that break chains in free radical chain reactions or they boost the oxidation stability potential of plant oils (Fruhwirth *et al.* 2008, Rezig *et al.* 2012).

Table 2.4 The amount of tocopherol in milligrams per 100 grams of pumpkin (Cucurbita maxima var. "Béjaoui") seed oil (Rezig *et al.* 2012)

Tocopherol	Composition
α- Tocopherol	128±14.42
γ- Tocopherol	113.66±1.52
δ- Tocopherol	177±14.17
Total	416.66±33.36

Since it has greater biological activity than other tocopherols, α-tocopherol is advised for ingestion by humans and animals, nonetheless, γ-tocopherol exhibits a greater antioxidant capacity than α-tocopherol (Fatnassi *et al.* 2009). In their investigation, (Amin *et al.* 2020) used the DPPH (2,2-diphenyl-1-picrylhydrazyl) test to assess the ability of native and hybrid pumpkin seed oils to scavenge free radicals. Both native and hybrid kinds of pumpkin seed oil demonstrated the percentage of antioxidant activity in the DPPH test at concentrations of 15, 20, 25, 30, 35, 40, and 45 μg/mL. Whereas hybrid pumpkin seed oil ranged from 96.05 ± 1.04% to 74.48 ± 1.10%, domestic pumpkin seed oil revealed a range of 63.21 ± 2.10% to 79.03 ± 6.14%. The total phenol content and antioxidant activity of the genotypes were observed to range from 11.753% to 56.944 mg GAE/100 g and 87.153 mg GAE/100 g, respectively (Seymen *et al.* 2016).

In order to highlight the significance of pumpkin seed oil's antioxidant and gene-protective properties (Yasir *et al.* 2016) looked into three different varieties of pumpkin seed oil. The researchers found that pumpkin seeds had an oil output that ranged from 20–41% and that the seeds' total phenol content was 16-40% comparable to gallic acid. They found that oil with a high phenolic content (149.5–396.4 GAE/g) has a strong ability to inhibit DPPH radicals.

Impact on Human Health pumpkin seed oil's high unsaturated fatty acid content makes it ideal for enhancing the nutritional value of food. Numerous health advantages are facilitated by pumpkin seed oil (Fu *et al.* 2006). Preventing prostate enlargement and reducing prostate size is the most important health benefit associated with pumpkin seed oil (Gossell-Williams *et al.* 2006, Tsai *et al.* 2006). Substantial dichromatic viscous oil from pumpkin seeds has been shown to have substantial antioxidant activity (Stevenson *et al.* 2007), and it has also been found to be an excellent defence against high blood pressure and cancer-causing illnesses (Zuhair *et al.* 2000, Jian *et al.* 2005). In addition to having qualities akin to those of almond oil, pumpkin seed oil also contains minerals, zinc and selenium derivatives, which have strong anti-inflammatory and antioxidant effects. It also has other health advantages, including the assistance of unique sterols. Pumpkin seed oil has characteristics similar to those of almond oil, particularly those carried by minerals, zinc, and selenium derivatives. It also has strong anti-inflammatory and antioxidant qualities, as well as numerous health advantages like lowering blood pressure and blood cholesterol levels thanks to unique sterols in the composition (Stevenson *et al.* 2007). Additionally, there is proof that pumpkin seed oil helps relieve arthritis (Fahim *et al.* 1995), hypercholesterolemia (Zuhair *et al.* 2000), and slow the advancement of hypertension (Zuhair *et al.* 2000).

Pumpkin seed lipid components have been linked to lower human bladder and urethral pressure as well as better bladder compliance (Zhang *et al.* 1994, Schilcher *et al.* 1996). It has been discovered that by encouraging hypoglycaemic activity, pumpkin seed oil helps to manage diabetes (Fu *et al.* 2006). It has been discovered that a significant source of vitamin E (tocopherol) in Japanese diets comes from pumpkin seed oil (Imaeda *et al.* 1999). Natural products like pumpkin seed oil are frequently used in folk medicine to treat various cardiovascular diseases like atherosclerosis and hypertension (Harvath *et al.* 1988, Zuhair *et al.* 1997).

Pumpkin (Cucurbita's) health-promoting properties one of the new issues facing the world is hyperglycaemia, which can be caused by poor insulin response (DM-type 2) or by no insulin at all (DM-type 1). Severe consequences from chronic hyperglycaemia include damage to

the kidneys (nephropathy), brain (neuropathy), and eyes (retinopathy) (American Diabetes Association, 2014). Globally, diabetes is reportedly becoming more prevalent, with estimates of 82 million cases by 2030 (Wild *et al.* 2004). Since over 80% of diabetes-related deaths happen in third-world and middle-class nations, many of them suffer from DM (Xu *et al.* 2014). To treat DM-2, a number of medications are utilized, including metformin, α-glucosidase, and analogues of glucagon-like peptide-1 (GLP-1). However, these medications have an impact on longevity, leading to a number of negative side effects. Because of the adverse impacts healthcare professionals throughout the world concentrate on using dietary components and herbs in place of anti-hyperglycaemic medications to treat DM-2 (Zhang *et al.* 2018).

Herbal preparations, which typically contain pumpkin, are used to treat hyperglycaemia in Mexico and China (Andrade-Cetto *et al.* 2005, Jia *et al.* 2003). Extensive research on the antidiabetic properties of pumpkin flesh, seeds, and peel has been carried out recently (Rolnik and Olas, 2020). It was reported that pumpkin fruit powder had anti-diabetic effects (Ahmad *et al.* 2019). According to this study, pumpkin powder is said to increase the body's insulin production, which lowers blood glucose levels. As a result, it also reduces the chance of renal injury (Chen *et al.* 2005). When complex carbohydrates are broken down in the small intestine, a-glycosidase, an enzyme found in the mucosa of the epithelium, usually takes over. The enzyme A-glycosidase raises blood glucose levels by breaking down glycosidic linkages found in complex carbohydrates (Jaiswal *et al.* 2012).

The fact that using pumpkin can reduce α-glycosidase activity is frequently mentioned. Additionally, it has been noted in the same study that pumpkin exhibits ACE inhibitory properties and can scavenge ROS (Kwon *et al.* 2007). Nevertheless, more investigation is needed to fully comprehend how pumpkin affects α-amylase. One of the most popular methods for examining the antidiabetic effects of any medicinal ingredient is the use of diabetes rats. Alloxan is a toxin that damages the pancreatic B-cells, which causes diabetes in rats (Bonner-Weir *et al.* 2005). Pumpkin's anti-hyperglycaemic qualities are demonstrated in case-control research conducted on rats. It supports the studies that show pumpkin has the

ability to increase the production of insulin and decrease blood glucose levels. In the early stages of diabetes, consuming pumpkin may lower postprandial glucose levels.

Pumpkin's protein-bound polysaccharide (PBPP), which is often dose-dependent, seems to reduce rats' hyperglycaemia (Quanhong *et al.* 2005). An in vivo investigation was carried out in 2019. Pumpkin polysaccharide hydrolysate (PPe-H) and pumpkin polysaccharide (PPe) both demonstrated their hypoglycaemic impact to reduce fasting blood sugar levels in DM-2-affected animals. Because PPe-H is beneficial to diabetics, it has reduced oxidative stress and increased endogenous GLP-1 secretion (Chen *et al.* 2019). Pumpkin has been shown in numerous studies to be beneficial for diabetics (Wang *et al.* 2017). According to one study, because pumpkin tends to raise superoxide dismutase (SOD) levels and shield islet cells against streptozotocin, it exhibits pancreas protective benefits. The combined effect of pumpkin polysaccharide and puerarin was examined in a recent study. It was shown that both of these substances have a tendency to reduce insulin resistance, which is followed by the overexpression of Nrf2/HO-1 and the PI3K/AKT pathway. They also possess hypoglycaemic qualities (Chen *et al.* 2019).

Mechanism of pumpkin polysaccharide hydrolysate in the treatment of DM-2 illustrates the synergistic antidiabetics effect of both pumpkin polysaccharide and puerarin.

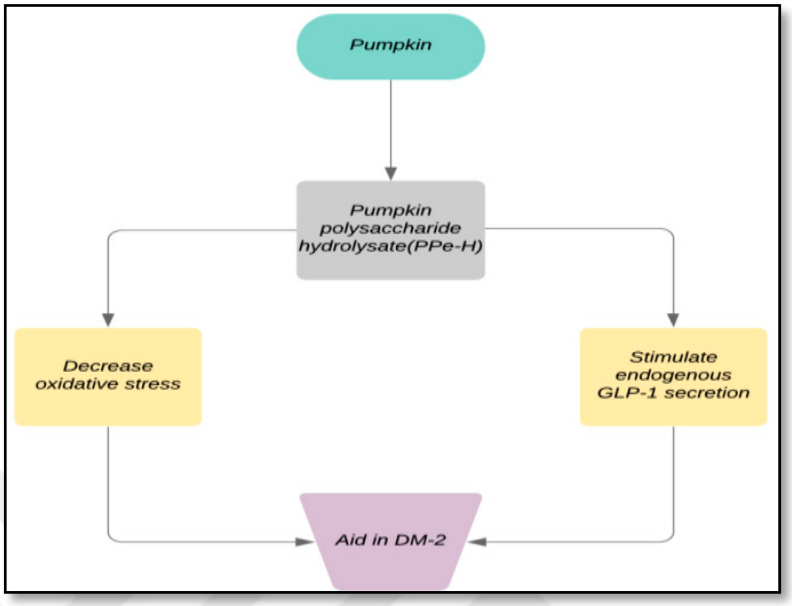


Figure 2.1 The mechanism of pumpkin polysaccharide hydrolysate in the treatment of DM-2

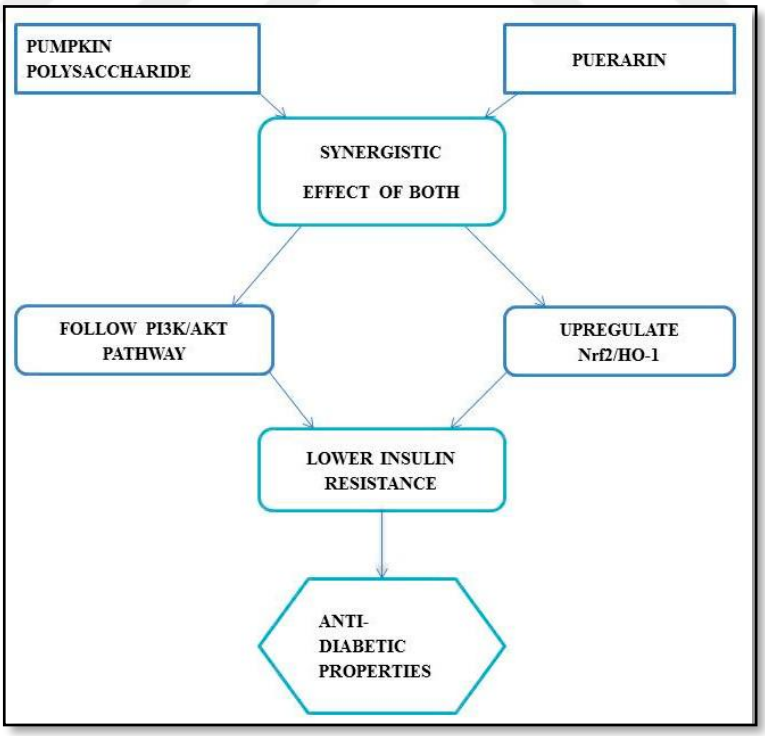


Figure 2.2 Antidiabetic synergistic effect of both pumpkin polysaccharide and puerarin

There have also been reports of the hypoglycaemic qualities of germinated seed oil and seed protein. Ungerminated seeds, however, do not exhibit hypoglycaemic characteristics (Li *et al.* 1956). Several clinical research have been carried out in addition to the in vivo investigations to investigate the effectiveness of pumpkin in treating and managing DM-type 2. It has been found that the consumption of pumpkin fruit juices contributes to fasting blood glucose levels in diabetics. In a similar vein, clinical investigations have demonstrated that pumpkin polysaccharide granules regulate postprandial glucose levels (Zuofeng *et al.* 2008, Yang *et al.* 2003). By encouraging the release of insulin and averting many of the problems linked to diabetes, pumpkin controls blood glucose levels (Askari *et al.* 2016). In light of this, it is claimed that pumpkin possesses antidiabetic qualities and can serve a protective role in preventing hyperglycaemia in people with diabetes. However, these findings did not allow researchers to report the precise mechanism of action of pumpkin against diabetes. There is a strong correlation between cancer and a diet high in oxidants and antioxidants.

Given that oxidative stress is linked to cancer, diet can either make the illness worse or better (Askari *et al.* 2016). There were 9.6 million cancer-related fatalities recorded in 2017. According to a GLOBOCAN report, 18.1 million people have cancer in 2018 and 9.6 million people will die from the disease in 2019 (Khazaei *et al.* 2019). While breast, lung, and prostate cancer are more prevalent in industrialized nations, stomach, cervical, and liver cancer rates are higher in less developed nations (Torre *et al.* 2015, WHO 2022).

It is possible to prevent cancer by following a healthy diet and consuming certain foods. Furthermore, only 5 to 10% of malignancies are inherited, the remainder are brought on by lifestyle choices (Anand *et al.* 2008). The anti-cancer benefits of pumpkin have been studied in a number of studies. According to reports, eating pumpkin seeds inversely correlates with the chance of developing many cancers, including lung, breast, and rectal cancer (Huang *et al.* 2004). According to reports, prostate cancer is the most common cause of death in America among all cancers (Jemal *et al.* 2010). A randomized, double-blind study was carried out in 2009 to look into the connection between prostate cancer and the pumpkin and saw palmetto seed oils. 47 patients, all of whom were 53.3 years old, were split up into groups

for this investigation. Saw palmetto oil and pumpkin seed oil together were found to lower blood prostate-specific antigen after three months (Hong *et al.* 2009).

By preventing testosterone-induced prostate hyperplasia, an oral dose of 20–40 mg/kg of pumpkin seed oil is thought to be beneficial for treating hyperplastic prostate glands for a period of 20 days (Tsaknis *et al.* 1997). It has been found that terpenoids and triterpenoids exhibit anti-tumour cytotoxic effects (Chudzik *et al.*2015). Cucurbitacin B (CuB) and cucurbitacin 163 E (CuE) are two subclasses of cucurbitacin, a class of tetracyclic triterpenes (Gutierrez *et al.* 2016). CuE is said to possess both antitumor and anticancer effects. CuE suppresses prostate cancer by tampering with actin and vimentin's structural integrity (Gutierrez *et al.* 2016). Cucurbitacin induces apoptosis through the PARP, MAPK, and JAK/STAT pathways, exhibiting anticancer effects (Rios *et al.* 2012).

Studies both in vivo and in vitro have shown that pumpkin inhibits the growth of prostate cancer. Additionally, a study conducted on Sprague Dawley rats revealed that pumpkin seed oil might suppress testosterone-induced hyperplasia, indicating that pumpkin seed oil may be useful in the treatment of benign prostatic hyperplasia (Gossell-Williams *et al.* 2008). Moreover, a clinical investigation showed the advantages of using pumpkin seed oil in the early stages of prostate cancer (Hong *et al.* 2009, Friederich *et al.* 2000). A significant source of phytoestrogen, including lignans and isoflavones, is pumpkin seeds. In female bodies, phytoestrogen has a tendency to attach to oestrogen receptors (ER) (Velentzis *et al.* 2008). An investigation into the possible link between phytoestrogen-containing pumpkin seed extract and breast cancer was carried out. It was revealed that MCF7, BeWo, and Jeg3 cells produced more oestradiol, and that MCF7 cells showed a decrease in ER-α (Richter *et al.* 2013). Basically, a study found that the proteins found in pumpkin seeds, called 2S albumins, have anti-cancerous qualities, especially when it comes to breast cancer. Protein was added to MCF7 cells at two distinct doses (10 and 30 μM). The DNA results revealed acridine orange staining and fragmentation tests, where 2S In MCF7-cell lines, albumins from pumpkin seeds have the propensity to induce apoptosis (Tomar *et al.* 2014).

Both directly and indirectly, pumpkin has an impact on serum lipid levels. Lohmann Brown Lite hens were used in the study, and they were fed a flaxseed and pumpkin oil-based diet. According to a report (Herkel *et al.* 2016), hens fed a pumpkin diet produced eggs with the highest MUFA, lowest PUFA, myristic acid, and other saturated fatty acid concentrations. But since the earlier discovery of a rise in myristic acid concentration in eggs following pumpkin supplementation (Hude *et al.* 2012), more research is necessary. In a related study from 2015, pumpkin cake was made with 72% wheat flour and pumpkin extract (5, 10, and 15%). When compared to the standard, pumpkin cake had higher levels of ash (6.45%), carotenoid (41 mg/100 g), and fibre (8%), control cake, which was made of wheat. On a biological basis of analysis, it was reported that pumpkin meal lowers the cholesterol and LDL level and increases the HDL level in a dose-dependent manner (Ali *et al.* 2015). Other researches also support these findings, in which rats with hypercholesterolemia were supplemented with pumpkin seeds. It was reported that PS resulted in a decrease in LDL, whereas an increase in HDL level lowered the level of cholesterol (Abuelgassim *et al.* 2012, Makni *et al.* 2008). In addition to affecting the atherogenicity and blood lipid profile, pumpkin also has a beneficial role in kidney functioning (Barakat *et al.* 2011). A hypercholesterolemic state causes the endothelium layer to malfunction, which raises the amount of vascular cell adhesion molecule (VCAM). Reactive oxygen species (ROS) activation lowers nitric oxide generation and prevents LDL oxidation, which is the cause of this. All of this VCAM amplification process is caused by the oxidation of LDL, which happens when nitric oxide (NO) levels are lower (Al-Showayman *et al.* 2010). 2.6% of pumpkin seed powder contains arginine, the precursor of NO linked to cardiac function, apoptosis, blood pressure maintenance, and inflammation (Glew *et al.* 2006).

An in vivo study that looked at the effects of pumpkin seed extract, namely arginine, found that supplementing rats with hyperlipidaemia with pumpkin seed extract could because arginine is present, increase the expression of NO generation. Furthermore, the generation of NO attenuates the oxidation of LDL, which results in a decrease in VCAM expression (Proboningsih *et al.* 2018). Thus, it can be concluded that altering one's way of life and incorporating pumpkins into one's diet on a regular basis can be a helpful dietary approach

for treating hypercholesterolemia. In addition to all of the above listed characteristics, pumpkin's anti-inflammatory qualities have been shown to have a significant impact on inflammatory illnesses such arthritis (Al-Okbi *et al.* 2017, Vijayalakshmi *et al.* 2018). The majority of males over 50 globally experience stromal and epithelial cell expansion, typically in the periurethral region and transition zone (McVary *et al.* 2011, Caro-Zapata *et al.* 2018).

Pumpkin seed extract, together with saw palmetto and prazosin, has been shown in numerous studies to alleviate lower urinary tract symptoms (LUTS) linked to benign prostatic hyperplasia (BPH) (Edwards *et al.* 2018, Vahlensieck *et al.* 2014). Pumpkin, when given for a full month at a dose of 14 mg/kg, is not only effective in treating LUTS but also in managing testosterone-induced hyperplasia (Tsaknis *et al.* 1997). However, the data are insufficient, and more investigation is needed necessary as these studies, in this instance, ignore a number of the effects of eating pumpkin. Although there are antimicrobial medications on the market today, researchers are concentrating on creating nutritional approaches to treat these infections because using these antimicrobe can result in drug resistance. Pumpkin seed oil has the potential to cure *Salmonella enterica*, *Escherichia coli*, *Klebsiella pneumonia*, *Acinetobacter baumannii*, *Candida albicans*, *Staphylococcus aureus*, and *Serratia marcescens* at a concentration of 2% (v/v) (Ahmad *et al.* 2019). *Mycosphaerella arachidis*, *Fusarium oxysporum*, and *Botrytis cinerea* can all be treated with 375 milligrams of pumpkin seed oil (Xiong *et al.* 2000). Similarly, isolated pumpkin proteins (>2 mM) have the ability to regulate fungal growth (Ng *et al.* 2009).

Similarly, it has been reported that three different pumpkin protein types—MAP11, MAP4, and MAP2—inhibit yeast growth. But MAP2 and MAP4 are not particularly valuable in the management of Gram-negative bacteria (Park *et al.* 2009). As a result, persons who live in places where they are more vulnerable to microbial infections in general should increase their usage of pumpkin, a food with excellent antibacterial qualities in a way that is dose-dependent (Gill *et al.* 2011). Researchers are constantly focusing on finding more efficient and cost-effective ways to cure wounds because they don't seem to be enough helped by medications (Abdel-Azim *et al.* 2011, Narayan *et al.* 2011). Pumpkin seed oil's high

concentrations of PUFA, tocopherols, and plant sterols promote wound healing (Stevenson *et al.* 2007). There have also been reports of analogous discoveries pertaining to pumpkin peel (Bahramsoltani *et al.* 2017).

Male reproduction has been impacted in recent years by lifestyle changes and a few disasters (Inhorn *et al.* 2004). Pumpkins are a useful tool to improve testicular characteristics and boost reproduction because they tend to reduce oxidative stress and decrease lipid peroxidation (Ejete-iroh *et al.* 2019, Fawzy *et al.* 2018). In a recent study, fluted pumpkin seed (FPS) extract was supplemented for 40 days at a rate of 40 mg/kg bodyweight. Extract may protect against oxidative cell damage and increase the quantity of spermatids, spermatocytes, and spermatogonia (Abarikwu *et al.* 2019). FPS extract can therefore be applied as a spermatocyte activator subsequent to chemotherapy. To fully grasp the impact of pumpkin on serum parameters and testes, more research is necessary as the effect is yet unknown (Akang *et al.* 2010).

Despite their small size, pumpkin seeds are incredibly rich in beneficial nutrients and nutraceuticals, including essential minerals, amino acids, phytosterols, unsaturated fatty acids, phenolic compounds, tocopherols, and cucurbitacins. Each and every one of these bioactive substances is essential to overall health and wellbeing. One exists the quickly rising need for novel natural goods to overcome the awareness of customers' health and safety aside from the aspirations to possess a wonderful and pleasurable appearance of ants. Customers are more worried about the components used in the creation of cosmetics. Within this plant seed oils have been utilized extensively as substitutes for due of their renewable nature, tives for raw materials. In order to offer cream emulsions multifunctional benefits on the skin, plant seed oil is typically included. In the current work, an oil-in-water emulsion that was stable was created using pumpkin seed oil (PSO).

Fruit spreads are made by mixing sugar with whole fruit, concentrated fruit juice, or processed fruit juice. The pumpkin, or *Cucurbita maxima*, is a member of the *Cucurbita*

genus and Cucurbitaceae family. Pumpkins come in three varieties: *Cucurbita pepo*, *Cucurbita maxima*, and *Cucurbita moschata*. Pumpkin had between 3.52% and 4.27% seeds. Pepitas are the name for pumpkin seeds, which have a white husk covering them. All grocery stores sell it as a snack in the form of hulled or semi hulled retailers and is employed in the food sector. It has a lot of nutritional and therapeutic properties. It is utilized in medicinal meals for these reasons. In addition to having a high protein content, the seed has pharmacological qualities that include anti-inflammatory, anti-bacterial, anti-fungal, anti-diabetic, and antioxidant capabilities (Nkosi *et al.* 2006).

Pumpkin seeds are frequently seen as industrial waste and thrown away. Whether they are boiled, roasted, or uncooked, seeds are utilized for domestic purposes in many places. Because they are rich in protein, fibre, and minerals like iron, they could be helpful in the food sector.

Polyunsaturated fatty acids (PUFAs), vitamins, phytosterol, potassium, magnesium, manganese, copper, zinc, and salt. The mango has a great taste, scent, and high nutritional content, making it one of the most significant tropical fruits. It is abundant in minerals, vitamins, antioxidants, fibre, carbohydrates, and water. Of the micronutrients present in plants with different health effects, polyphenols, it has the greatest concentration. 1991, Shahidi *et al.* With regard to a range of chronic pathological conditions associated with inflammatory reactions, it offers more phytochemicals, which have anti-inflammatory qualities. With antidiabetic and anticancer capabilities, the pumpkin has higher levels of bioactive compounds. Papaya belongs to the Caricaceae family. It has elevated levels of calcium, iron, and the vitamins A, B, and C.

One popular application of pumpkin seeds is in the production of seed creams, which can be used as spreads, fillings, or toppings in various food products. In recent years, chocolate spread with pumpkin seed has gained popularity among consumers for its rich and distinctive

taste. However, achieving the desired sensory, textural, and rheological properties in these spreads can be challenging.

Roasting is a critical step in the production of pumpkin seed cream as it enhances the flavour and aroma of the seeds. It also affects the texture and overall quality of the final product. Different roasting temperatures and times can lead to variations in the sensory and rheological properties of the pumpkin seeds. Therefore, optimizing the roasting conditions is crucial to producing an acceptable chocolate spread with pumpkin seed.

In some previous studies, some pumpkin seed-based paste type products were produced and some of their properties were analysed. However, these studies are not in the nature of spreadable chocolate, which is completely different from our thesis subject (Nikolic *et al.* 2014, Nikolic *et al.* 2017, Rohini *et al.* 2022).

In this project, we aimed to investigate the effect of different pumpkin seed roasting temperatures and times on the sensory, textural, and rheological properties of chocolate spread with pumpkin seed. Three temperature levels - low (120 °C), medium (150 °C), and high (180 °C) - will be tested, along with two roasting times - short 20 minutes and long 40 minutes. The roasted pumpkin seeds will then be incorporated into the production of chocolate spread with pumpkin seed. This spread will undergo thorough physical, textural, rheological, and sensory analyses to assess its overall quality and acceptability. The findings of this study will provide valuable insights into the optimal roasting conditions for pumpkin seeds in the production of chocolate spread with pumpkin seed. This knowledge can be utilized by food manufacturers and researchers to develop high-quality and desirable seed spreads that cater to consumer preferences. Furthermore, it will contribute to expanding the range of healthy and functional food products available in the market.

3. MATERIAL AND METHOD

3.1 Material

The materials used in the production of chocolate spread with pumpkin seed are powdered sugar (Dr. Oetker, Izmir), cocoa powder (Altınmarka, İstanbul), pumpkin seed (Hasatköy, Denizli), skimmed milk powder (Katkı Dünyası, Gemici Gıda, Istanbul) and cream type oil oil (BES 41, Besler, İstanbul), sunflower lecithin (Katkı Dünyası, Gemici Gıda, İstanbul), ammonium phosphatite (AMP) (4458, Palsgard, İstanbul) salt (Billur Tuz, İzmir) and vanillin (Katkı Dünyası, Gemici Gıda, İstanbul).

3.2 Method

3.2.1 Pumpkin seed roasting conditions

In the study, 3 different temperature (low 120 ºC, medium 150 ºC and high 180 ºC) and 2 different time (short 20 minutes and long 40 minutes) combinations were used for pumpkin seed roasting. Roasting conditions and sample codes for roasting temperatures and times are presented in Table 3.1. An oven (Memmert, UN160 plus, Germany) was used for roasting. After reaching the desired temperature, the pumpkin seeds placed on the tray were kept on the middle shelf of the oven at the specified temperature and time conditions.

3.2.2 Processing of chocolate spread with pumpkin seed

The composition of chocolate spread with pumpkin seed included 42.5% sugar, 28.1% fat, 13% pumpkin seed, 8.5% milk powder, 7% cocoa, 0.4% lecithin, 0.3% AMP, 0.1% vanillin and 0.1% salt. For the production of chocolate spreads, roasted pumpkin seeds were first crushed in a blender. The crushed pumpkin seeds and 20% of the oil to be used were placed in a melanger (a granite stone in a granite bowl mill) (ECGC–12SLTA, Cocoatown, USA).

All the remaining dry ingredients were mixed in a bowl and the dry ingredients, and the remaining oil were gradually added to the melanger. The process took a total of 3 hours for refining and conching. The test samples (1 kg per batch) were packed in 300 g glass jars and stored at 24±0.5 °C for 7 days before analysis. The production of chocolate spread with pumpkin seed was carried out in two replicates.

Table 3.1 Experiment plan

Samples	Temperature (ºC)	Time (minutes)
Low–Short (LS)	120	20
Low–Long (LL)	120	40
Medium–Short (MS)	150	20
Medium–Long (ML)	150	40
High–Short (HS)	180	20
High–Long (HL)	180	40

Figure 3.1 The oven used in roasting

3.2.3 Accelerated oil separation

The accelerated oil separation (AOS) analysis was performed according to method given by Aydemir (2019). 7.5 g of chocolate spread was weighed into a centrifuge tube and subjected to centrifugal force of 20000 g (3–30 KS, SIGMA, Germany) at 20 °C for 10 min. After centrifugation, the separated oil accumulated on the tube was collected with an automatic pipette and weighed. The amount of oil separated from 7.5 g sample was calculated as AOS (g oil/100 g sample) as oil separated from 100 g sample.

Figure 3.2 Centrifuge device used in the thesis

3.2.4 Texture analysis

Spreadability analysis was performed using a TA.XT Plus Texture analyser (Stable Micro Systems, Godalming, UK). Spread samples were placed in the TTC spreadability rig (HDP/SR, 14580) at room temperature (24±1°C) and the analysis conditions were applied: Probe 30 mm distance preset, test speed 3 mm/s, post–test speed 10 mm/s, compression

distance 15 mm. The data obtained were calculated with Texture Exponent Version 4.013.0 (Stable Micro Systems).

Figure 3.3 Texture analyser used in thesis

3.2.5 Rheological analyses

The rheological properties of chocolate spreads were determined in a shear stress/shear rate–controlled rheometer (Anton Paar, MCR 302 model, Austria) equipped with cone and plate geometry (radius: 25 mm, gap: 0.106 mm) and Peltier heater system. The samples were temperature equilibrated by holding at 25 °C for 2 minutes before measurement and analysed at the same temperature. The analyses were performed in two repetitions.

Steady shear properties

Shear stress values versus shear rate between 0.1-100 s^{-1} were recorded to determine the flow properties of the samples. The data obtained were fitted to the Casson (3.1) and Windhab (3.2) model with a software package (RheoCompass v1.21, Anton Paar, Austria).

$$\tau^{0.5}=(\tau_y)^{0.5} + \eta_{pl}(\dot{\gamma})^{0.5} \quad (3.1)$$

where τ is shear stress, τ_y is Casson yield stress, η_{pl} is plastic viscosity, $\dot{\gamma}$ is shear rate.

$$\tau = \tau_0 + \eta_\infty \dot{\gamma} + (\tau_1-\tau_0)\,(1-e^{-\dot{\gamma}/\dot{\gamma}^*}) \quad (3.2)$$

where τ is the shear stress (Pa), τ_0 is the shear stress at zero point i.e. the actual yield stress (Pa), η_∞ is the infinite viscosity (Pa·s), is the shear rate (s^{-1}), τ_1 is the hypothetical shear stress or linear shear stress (Pa) and $\dot{\gamma}^*$ is the shear rate or characteristic shear rate (s^{-1}) corresponding to the infinite viscosity.

Dynamic shear properties

Stress sweep test was performed at a constant frequency of 0.1 Hz to determine linear viscoelastic regions (LVR) of chocolate spreads between 0.01 and 10 Pa at 25 °C, and then frequency sweep tests were performed at this region which was selected as 1 Pa for all samples (Loncarevic *et al.* 2016). Storage (G') and loss (G'') moduli were recorded against angular frequency (ω) between 1 and 10 Hz. All the measurements were carried out in two repetitions.

3.2.6 Particle size distribution

This analysis was performed according to the method given by Mohd Rozalli *et al.* (2015) as a service procurement (Bursa Technical University Central Research Laboratory).

3.2.7 Water activity

Water activity measurements of chocolate spread were carried out at 25°C in a thermostat water activity meter (AquaLab 4 TE, Pullman, WA, USA).

Figure 3.4 Water activity device used in the study

3.2.8 Colour analysis

The determination of CIELab L*, a*, b* colour parameters of chocolate spreads was carried out in Minolta 400 (Konika, Japan) colour measuring device.

Figure 3.5 Colour device used in the study

3.2.9 Sensory analysis

Sensory analyses were evaluated by a partially trained panellist group (30 people) consisting of Çankırı Karatekin University Faculty of Engineering lecturers according to the score card prepared in terms of taste-aroma (5 points), odour (5 points), flavour-aroma, texture-structure and colour-appearance. Sample numbers were coded as 3-digit numbers. During the texture evaluation, the chocolate spreads were also tested with the help of a knife for spreadability of the spreads on bread. Water was given to the panellists to balance the taste between the samples. The scorecard used in the sensory analysis is presented in.

Table 3.2 Sensory score card

Samples	**364**	**847**	**145**	**647**	**492**	**719**
ODOUR (Very bad:0 – Very good:5)						
General Odour →						
Pumpkin seed Odour →						
COLOUR and APPEARANCE (Very bad:0 – Very good:5)						
Colour →						
Appearance →						
FLAVOUR and AROMA (Very bad:0 – Very good:5)						
Flavour →						
Pumpkin Seed Aroma →						
Raw pumpkin seed flavour (none 0-very 5) →						
Burnt pumpkin seed taste (none 0-very 5) →						
STRUCTURE and TEXTURE (Very bad:0 – Very good:5)						
Spreadability →						
Consistency →						

3.2.9 Statistic analysis

Data will be evaluated using SPSS 22 package programme. Analysis of variance (One-way Anova) and Tukey multiple comparison test (95% confidence interval) were used for comparison of means.

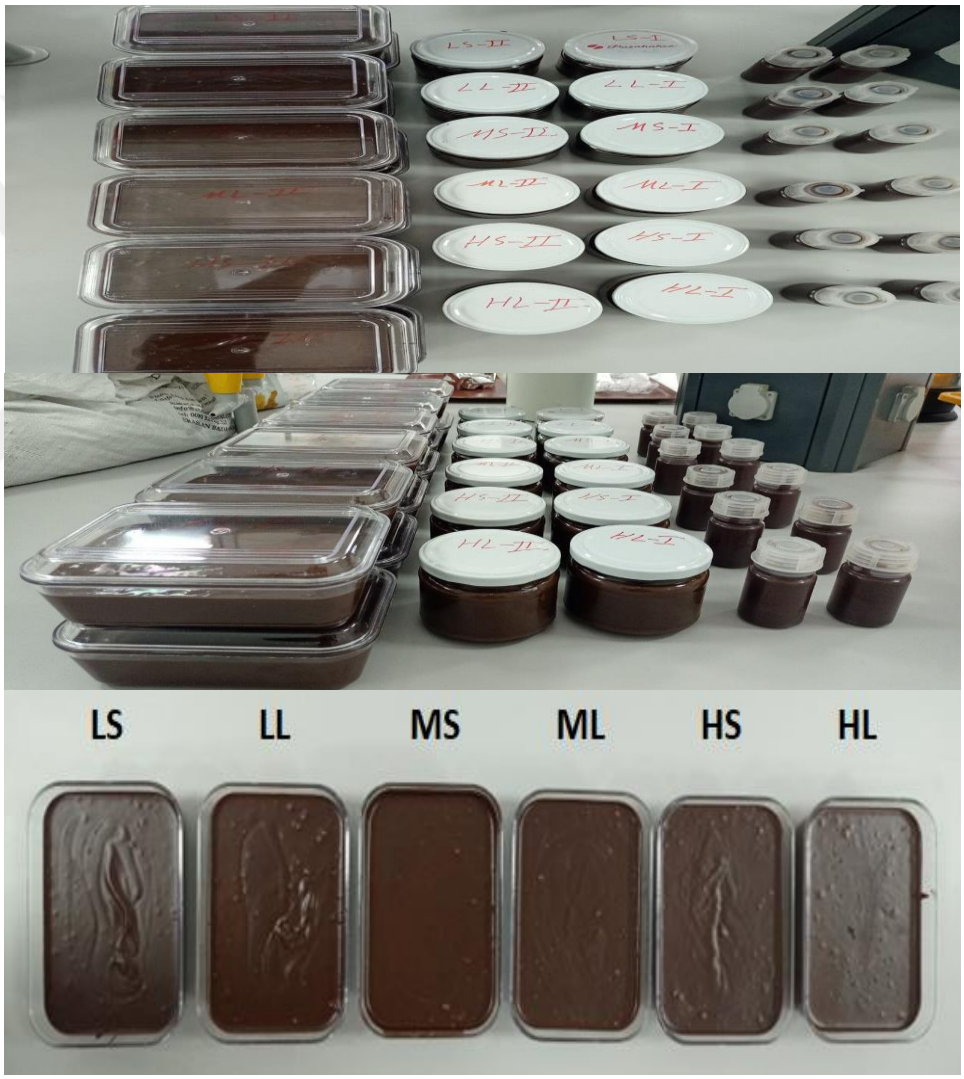
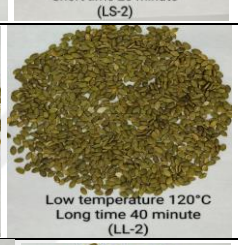


Figure 3.6 Photographs of the produced chocolate spreads with pumpkin seed

Figure 3.7 Photographs of the roasted pumpkin seeds

Raw Pumpkin Seeds	Roasted Pumpkin Seeds	Sample Code
		LS
		LL
		MS
		ML
		HS
		HL

4. RESULTS AND DISCUSION

4.1 AOS Results

The cause of oil separation in chocolate spreads is due to the oil used and the specific gravity difference of the solid particles (Gills and Resurreccion 2000). Free oil separation observed on the surface of such paste products may be perceived as a problem for the consumer. A relationship can be established between AOS value and free oil separation on the surface. The AOS values of the chocolate spreads with pumpkin seed are given in Table 4.1 and Figure 4.1.

Table 4.1 AOS values of chocolate spread with pumpkin seed (%)

Spread samples	AOS (%)
LS	9.80±0.13b
LL	9.70±0.17b
MS	9.30±0.03ab
ML	9.57±0.10ab
HS	9.30±0.10ab
HL	9.07±0.07a

a-b Mean values within each column followed by different letters are significantly different ($p<0.05$)

The AOS values of the samples varied between 9.07% and 9.80%. Aydemir (2019a), Ertuğrul (2023), and Sattari (2023) found the AOS value in cocoa hazelnut creams in the ranges of 4.87–11.94%, 8.27–11.73% and 10.18–11.43%, respectively. Aydemir *et al.* (2021a) also reported very similar results to our findings in cocoa hazelnut creams. Aydemir (2021b) determined 6.27–12.80% AOS values in cocoa carob creams. Although the values between the samples were quite close, there was a statistically significant difference between the spread samples ($p<0.05$). In general, it was observed that fat separation decreased as the temperature and time increased. The spread HL showed significantly lower AOS value than spread LS and LL ($p<0.05$). Aydemir (2019b) found a positive correlation between roasting temperature and time and AOS values in cocoa chestnut cream and Ziaolhagh *et al.* (2017)

in walnut cream. They attributed this to the denaturation of hydrophobic proteins by roasting and the consequent decrease in fat binding properties.

In contrast, Sattari (2023) reported that the lowest fat separation in hazelnut cream with cocoa was found in creams produced from hazelnuts roasted at high temperature. In our study, similar to hazelnut, it is understood that as the roasting temperature of pumpkin seeds increases, the oil separation decreases, that is, the oil binding capacity increases. The fat binding capacity of a protein depends on many different factors such as surface hydrophobicity, lipophilic groups, conformational properties and non-protein compounds in the protein preparation (Pham *et al.* 2017).

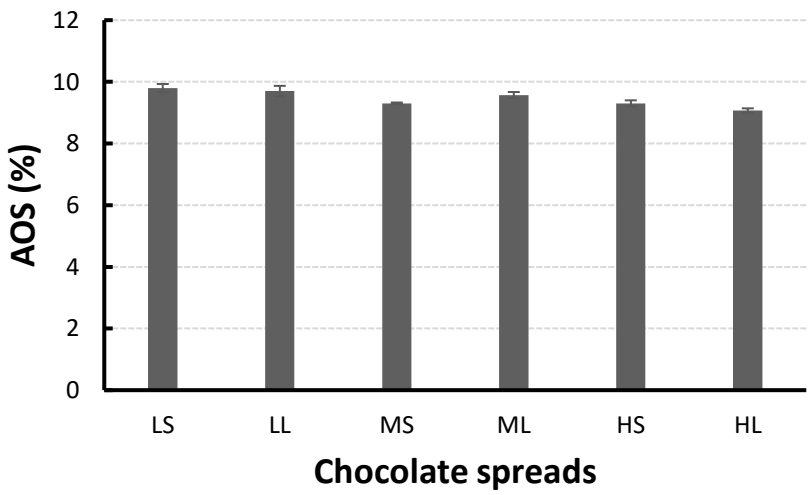


Figure 4.1 AOS values of chocolate spread with pumpkin seed (%)

4.2 a_w Results

The relationship between deterioration and quality defects that occur during the processing and preservation of foodstuffs is successfully expressed by the water activity parameter. Water activity shows how the water in foods is bound to its structure, its potential to be used for microbiological activities through chemical and enzymatic reactions and its level (Us

2014). The water activity values of the chocolate spreads produced were determined in the range of 0.188–0.214 (Table 4.2 and Figure 4.2). Aydemir *et al.* (2021a), Ertuğrul (2023) and (Sattari, 2023) found the water activity value in cocoa hazelnut creams in the ranges of 0.23–0.255, 0.267–0.301, and 0.299–0.337 respectively. Again, Aydemir (2019b) reported water activity values in cocoa chestnut creams and Aydemir *et al.* (2021b) reported water activity values in cocoa carob creams in the range of 0.207–0.264 and 0.274–0.309, respectively. The very low amount of water that may be present in the structure of the product in the production of chocolate spread with pumpkin seed is due to the raw materials, mainly pumpkin seed, powdered sugar, milk powder and cocoa. Since chocolate spread are a suspension product consisting of a distribution of solid particles on a fat base and do not normally contain water, water activity is expected to be very low. In general, there is a tendency to decrease in water activity with increasing roasting temperature and time, but the only difference between the samples was observed between spreads HS and LS ($p<0.05$). There was no statistical difference between other spreads in terms of water activity values ($p>0.05$). Loss of moisture in pumpkin seed is expected with roasting. However, the closeness of the roasting temperature and time combinations and the relatively low pumpkin seed content of 13% did not cause a significant change in moisture levels and therefore in the water activity of the product.

Table 4.2 a_w values of chocolate spread with pumpkin seed

Spread samples	a_w
LS	0.214±0.007b
LL	0.196±0.001ab
MS	0.208±0.001ab
ML	0.188±0.001a
HS	0.188±0.007a
HL	0.192±0.004ab

a-b Mean values within each column followed by different letters are significantly different ($P<0.05$)

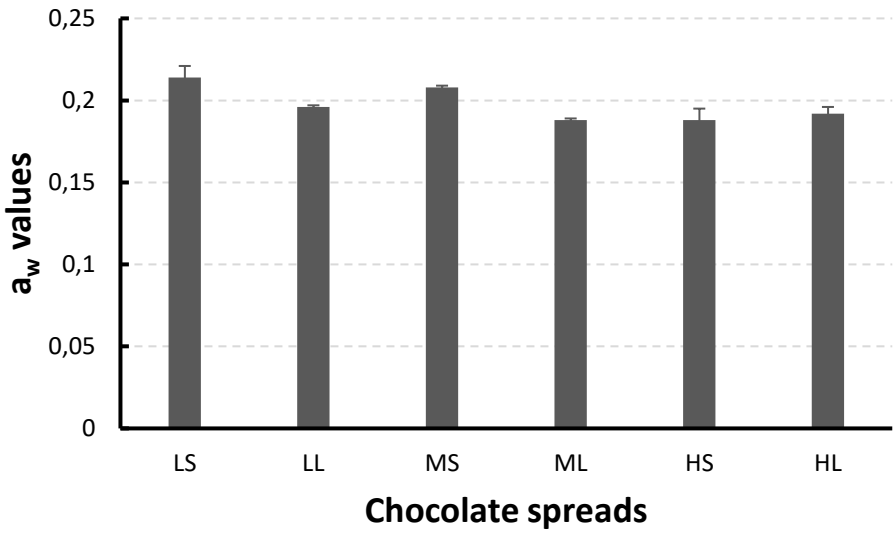


Figure 4.2 a_w Values of chocolate spread with pumpkin seed

4.3 Textural Characteristics

Spreadability, the ease with which a product can be spread, is a desirable characteristic of margarine, butter, jam, jam, chocolate spreads, etc. Firmness (g force) is the maximum force on a product that displays substantial resistance to deformation. Stickiness (g force) is the maximum force necessary to overcome the attractive forces between the surface of the food and the surface of the probe with which the food comes in contact. Work of shear (gs) is the total amount of force required to carry out the shearing process. It is a measure of ease of spreadability. Work of adhesion (gs) is the total amount of the force involved in the withdrawal of probe from the sample (Basu and Shivhare, 2010).

The values of textural parameters of chocolate spreads are presented in Table 4.3. The firmness values of the chocolate spreads were determined in the range of 1420.23–1687.71 g. No significant difference was observed between the spread samples ($p>0.05$). The highest firmness value was found in the spread LL while the lowest value was obtained in the spread HL. It can be said that as the roasting temperature and roasting time increased, a slight tendency to decrease in firmness value was detected. Sattari (2023), Ertuğrul (2023) and

Çoban (2021) reported a firmness value of 979.13–1242.35 g, 267.20–340.28 g and 1619.112–1839.326 g in cocoa hazelnut creams, respectively. This difference is due to the difference in the test conditions selected during the analysis.

Table 4.3 Textural parameters of chocolate spread with pumpkin seed

Chocolate spreads	Firmness (g)	Work of shear (gs)	Stickiness (g)	Work of adhesion (gs)
LS	1681.82±16.66a	1748.96±38.18a	-1495.03±58.56a	-578.22±5.09a
LL	1687.71±109.56a	1705.47±139.09a	-1604.21±62.11a	-583.25±27.57a
MS	1563.10±13.53a	1636.63±68.18a	-1544.59±0.03a	-562.34±0.26a
ML	1684.66±37.51a	1676.22±11.48a	-1660.82±85.27a	-517.77±31.74a
HS	1654.63±36.96a	1631.02±15.41a	-1638.60±127.35a	-523.63±14.21a
HL	1420.23±8.44a	1384.04±27.55a	-1430.89±23.06a	-512.64±17.95a

a-a Mean values within each column followed by same letters are not significantly different ($p<0.05$)

Spreadability (work of shear) values of the chocolate spreads were determined in the range of 1384.04–1748.96 gs. There was no significant difference between the spread samples ($p<0.05$). The highest spreadability value was found in the spread LS while the lowest value was obtained in the spread HL. Similar to firmness, it is observed that spreadability values decrease slightly as roasting temperature and roasting time increase. Aydemir (2019b) obtained similar results in commercial cocoa chestnut creams. Sattari (2023), Ertuğrul (2023) and Çoban (2021) reported spreadability values of 907.58–1171.48 gs, 219.96–302.03 gs and 2174.901–2482.845 gs in cocoa hazelnut creams, respectively. This difference is due to the difference in the test conditions selected during the analysis.

The stickiness values of the spread samples were determined between -1430.89 and -1660.82 g. There was no significant difference between the chocolate spreads ($p>0.05$). The highest stickiness value was found in the spread ML while the lowest value was obtained in the spread HL. No trend was detected in hardness value depending on roasting temperature and roasting time. Sattari (2023), Ertuğrul (2023) and Çoban (2021) reported stickiness values of -1077.25 and -1425.02 g. -204.32 to -236.76 g and -2195.972 to -2529.029 g for cocoa

hazelnut creams, respectively. This difference is due to the difference in the test conditions selected during the analysis.

The work of adhesion (adhesivity) values of the chocolate spreads ranged from -512.64 to -583.25 gs. No significant difference between the spread samples was observed. The highest adhesion value was found in the sample LL while the lowest value was obtained in the sample HL. As the roasting temperature and roasting time increased, a slight decreasing trend was observed in the adhesivity value. Aydemir (2019b) and Coban (2021) obtained very similar results in cocoa chestnut creams and cocoa hazelnut creams, respectively. Sattari (2023), Ertugrul (2023) and reported a stability value of -386.72 to -469.55 gs and -135.99 to -170.61 gs in cocoa hazelnut creams, respectively. This difference is due to the difference in the test conditions chosen during the analysis.

4.4 PSD Analysis Results

PSD parameters values of chocolate spread are given in Table 4.4. The d(0,1) is such a limit value that the volume of particles with a diameter below this value is 10% of the total volume. The d(0,1) values of the chocolate spreads were found to be between 1.13 and 7.66 µm. No significant difference was observed between the spreads samples in terms of d(0.1) values ($p>0.05$). Sattari (2023), Ertuğrul (2023), Coşar (2021) and Aydemir (2019a) determined the d(0.1) value in the range of 1.16–1.26, 0.78–0.99, 0.76–1.39 and 1.00–1.20 µm in cocoa hazelnut creams, respectively. The same parameter was reported to be 1.11–1.17 in cocoa chestnut creams (Aydemir and Atalar 2019) and 2.94–3.59 µm in cocoa creams (Loncarevic *et al.* 2016). Although it is not statistically significant, it is observed that d(0,1) value decreases as the roasting temperature increases.

The d(0.5) value indicates that 50% of the sample volume has a particle size smaller than this value. It was found that d(0.5) values varied between 7.55–54.65 µm in spread samples. No significant difference was found between the samples in terms of d(0,5) values ($p>0.05$).

Sattari (2023), Ertuğrul (2023), Coşar (2021) and Aydemir (2019a) determined d(0,5) values in the range of 5.34–6.89, 3.63–5.77, 3.83–4.38 and 2.24–3.04 µm, respectively. The same parameter was reported to be 3.26–3.90 in cocoa chestnut creams and 12.43–16.45 µm in cocoa creams (Loncarevic *et al.* 2016). Although it is not statistically significant, it is observed that d(0,5) value decreases as the roasting temperature increases.

Table 4.4 PSD Parameter values of chocolate spread with pumpkin seed

Spreads	d(0.1)	d(0.5)	d(0.9)	D[3.2]	D[4.3]	Span
LS	7.66±6.34a	35.95±18.25a	78.65±5.35a	8.77±3.83a	46.35±7.85a	2.67±1.38a
LL	5.55±0.30a	54.65±2.45a	80.90±0.70a	10.40±0.40a	51.45±2.05a	1.38±0.05a
MS	2.35±1.11a	36.55±25.85a	101.00±2.00a	6.46±2.16a	47.55±8.55a	5.55±4.02a
ML	6.51±5.39a	28.41±22.19a	50.20±27.50a	7.81±4.59a	29.57±20.02a	2.28±1.18a
HS	1.17±0.01a	16.31±8.28a	63.25±14.65a	4.26±0.54a	27.65±5.15a	4.52±1.40a
HL	1.13±0.03a	7.55±1.80a	29.55±14.05a	3.46±0.43a	12.06±4.93a	3.52±1.01a

a-a Mean values within each column followed by same letters are not significantly different ($p<0.05$)

The proportion of particles with diameters below d(0.9) is 90% of the sample volume. The d(0.9) levels of the chocolate spread samples were found to be between the limit values of 29.55–101.00 µm. No significant difference was found between the samples in terms of d(0.9) values ($p>0.05$). Sattari (2023), Ertuğrul (2023), Coşar (2021) and Aydemir (2019a) determined d(0.9) values in the range of 20.40–27.30, 17.81–23.85, 12.88–16.99 and 7.32–21.50 µm, respectively, in cocoa hazelnut cream. The same parameter was reported to be 184–247 in cocoa chestnut creams and 55.81–69.66 µm in cocoa creams (Loncarevic *et al.* 2016). Although it is not statistically significant, it is observed that d(0,9) value decreases as the roasting temperature increases.

The surface weighted average particle size D[3,2] values of the spread samples were found to be in the range of 3.46–10.40 µm. The D[3,2] values of the chocolate spread samples were similar to each other ($p>0.05$). Sattari (2023), Ertuğrul (2023), Coşar (2021) and Aydemir (2019a) found that the D[3,2] values of cocoa hazelnut creams were in the range of 3.04–3.46, 2.00–2.53, 2.22–2.83 and 1.93–2.52 µm, respectively. Afoakwa *et al.* (2008) reported

that the D[4,3] volumetric weighted average particle diameter value showed the greatest PBD effect on chocolate rheology.

It was determined that the volumetric weighted average particle size D[4,3] levels of the spread samples were in the range of 12.06–51.45 µm. The D[4,3] values of the chocolate spread samples were similar to each other ($p>0.05$). Sattari (2023), Ertuğrul (2023), Coşar (2021) and Aydemir (2019a) determined that the D[4,3] values in cocoa hazelnut creams were in the range of 8.49–11.07, 6.86–9.29, 6.41–7.29 and 3.76–12.20 µm, respectively.

It was determined that the size distribution width (span) of the chocolate spread samples was in the range of 1.38–5.55. The distribution width values of spread samples were found to be similar to each other ($p>0.05$). Sattari (2023), Ertuğrul (2023), Coşar (2021) and Aydemir (2019a) found that the distribution width values of hazelnut creams with cocoa were in the range of 3,47–4,19, 3.45–6.36, 2.94–3.99 and 2.83–7.69 µm, respectively.

The applied pumpkin seed roasting temperature and time combinations were found to have no effect on PSD parameter values ($p>0.05$).

4.5 Rheological Analysis Results

The shear stress and viscosity variation of chocolate spreads against shear rate are presented in Figure 4.3 and 4.4. A non–linear shear stress value versus shear rate was obtained (Figure 4.3). In other words, a non-Newtonian flow type was obtained in all samples. It was observed that the apparent viscosity values decreased with shear rate (Figure 4.4).

Chocolate spread samples were determined to have a shear thinning (pseudoplastic) flow type. This flow behaviour has been found in many studies investigating spreadable products (Abbasi and Farzanmehr 2009, Li and Liu 2019, Aydemir 2019a, Mieles-Gómez *et al.* 2021, Ertuğrul 2023, Sattari 2023). The shear stress and viscosity values of the samples depending

on the shear rate exhibited very similar profiles. This situation was also consistent with the textural parameters. Shear stress and apparent viscosity values of the samples at 30 s^{-1} shear rate are also given in Table 4.5. As can be seen from Table 4.5, no difference was found between the shear stress and apparent viscosity values of chocolate spreads corresponding to 30 s^{-1} shear rate ($p>0.05$).

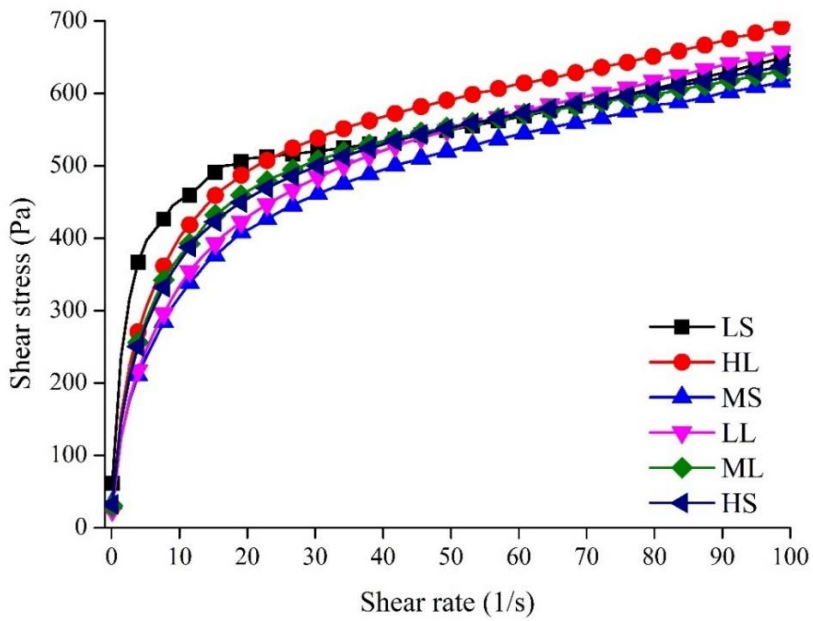


Figure 4.3 Shear stress variation versus shear rate of chocolate spreads

Table 4.5 Shear stress and apparent viscosity values of spread samples at 30 s^{-1} shear rate

Chocolate spreads	Shear stress (Pa)	Apparent viscosity (Pa·s)
LS	524.85±4.21a	17.24±0.14a
LL	486.79±2.46a	15.99±0.08a
MS	497.19±35.95a	16.33±1.18a
ML	514.00±6.08a	16.88±0.20a
HS	543.56±43.74a	17.85±1.44a
HL	503.27±34.89a	16.53±1.15a

a-aThe difference between the same letters in the same column is not significant ($p>0.05$)

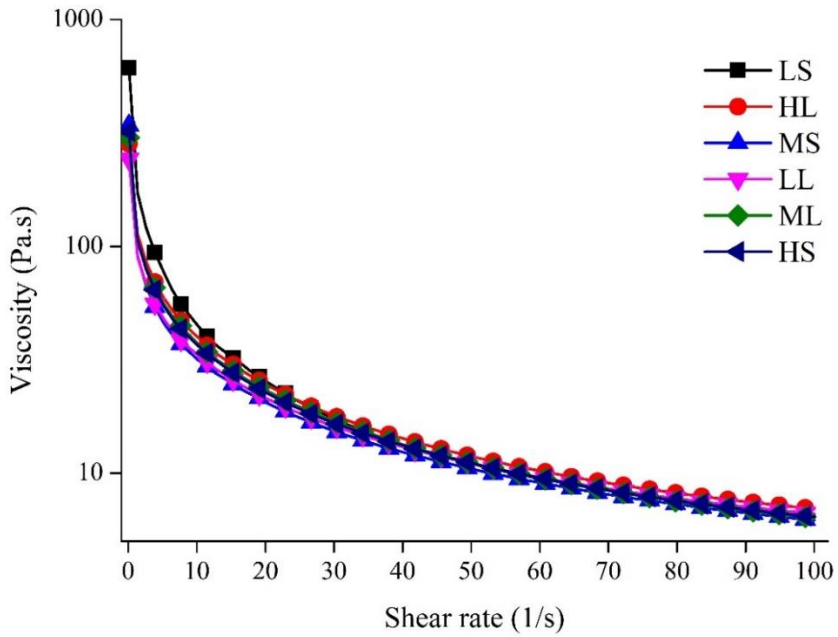


Figure 4.4 Shear rate versus viscosity variation of chocolate spreads

The parameters found by applying Casson and Windhab models to the flow behaviour curves given by the chocolate spread samples are presented in Table 4.6 and Table 4.7 respectively. The R^2 values of the Windhab model showed a better fit than those of the Casson model. When the yield stress (τ_0) values obtained from the Casson model were analysed, no significant difference was observed between the spread samples ($p>0.05$). In terms of plastic viscosity values, the only significant difference was that spread LL showed a higher plastic viscosity value than the other samples ($p<0.05$). There was no difference between the other samples ($p>0.05$).

In terms of the actual (τ_0) yield stress values of the spread samples obtained from the Windhab model, spread LS showed a significantly higher value than the other spreads ($p<0.05$) and the other spread showed similar yield stress values ($p>0.05$). It was determined that the linear yield stress (τ_1) values of the spread samples were similar to each other ($p>0.05$). Many materials used in daily life show solid or liquid properties depending on the

applied stress. At low stresses these materials deform essentially elastically, but when a critical stress is exceeded, they yield, this critical value is called yield stress. The determination of yield stress is critical in industrial processes, for example, yield stress is required to know the minimum pressure required to start a cream flowing in a pipeline or to know the hardness of creams (Dinkgreve *et al.* 2016). A significant difference was found between the infinite viscosity values of the chocolate spread samples ($p<0.05$). This difference was found only between spreads ML and HS. Spread HS gave lower infinite viscosity value. The infinite viscosity values of other spread samples were similar ($p>0,05$). No significant difference was observed between the characteristic shear rate values of the spread samples ($p>0.05$).

Table 4.6 Casson model parameters of chocolate spreads

Chocolate spreads	**τ_0 (Pa)**	**η_{pl} (Pa·s)**	**R^2**
LS	386.05±10.35a	343.13±66.04a	0.978
LL	267.48±1.83a	941.21±7.35b	0.973
MS	283.07±5.60a	656.08±11.03ab	0.985
ML	330.28±2.95a	515.60±4.81a	0.963
HS	324.94±8.27a	593.83±7.15a	0.967
HL	335.62±6.05a	515.28±128.96a	0.980

a-b Mean values within each column followed by different letters are significantly different ($P<0.05$)

When Casson and Windhab model parameters are analysed, it is seen that roasting temperature and roasting time generally have no significant effect on these parameters. No trend was detected between temperature and these parameters.

The frequency dependent storage (G′, elastic behaviour) and loss modulus (G″, viscous behaviour) values of chocolate spread samples are given in Figure 4.5 and 4.6.

Table 4.7 Windhab model parameters of chocolate spreads

Spreads	τ_0 (Pa)	τ_1 (Pa)	η_∞ (Pa·s)	γ^* (Pa)	R^2
LS	46.68±0.51c	455.87±0.16a	2051.95±137.55ab	3.16±0.59a	0.994
LL	19.07±0.73a	451.46±0.73a	2511.75±62.85ab	5.20±0.39a	0.996
MS	29.58±0.13ab	423.89±42.44a	2302.35±239.15ab	5.41±0.79a	0.995
ML	20.56±2.25ab	392.78±2.13a	2875.50±14.30b	5.79±0.00a	0.991
HS	24.29±1.58ab	452.80±17.35a	1837.60±274.30a	4.48±0.63a	0.993
HL	27.92±2.80ab	448.96±27.03a	2294.30±42.90ab	5.82±0.72a	0.994

a-a Mean values within each column followed by same letters are not significantly different ($p<0.05$)

In all samples, G′ values showed a higher change than G″ values along the entire frequency line. This shows that elastic behaviour is more effective than viscous behaviour in chocolate spread samples. It was observed that G′ and G″ values decreased slightly as the roasting temperature increased. This may be related to the slight decrease in particle size as the roasting temperature increases.

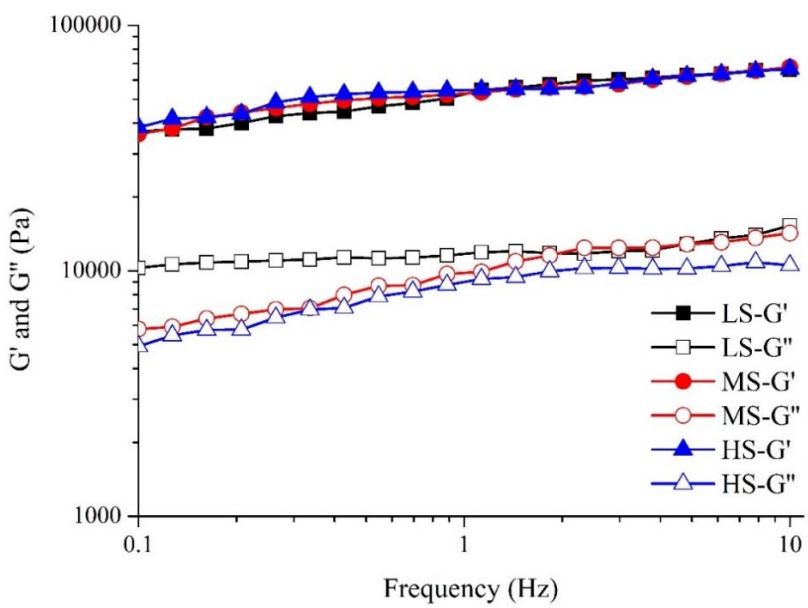


Figure 4.5 Storage (G′) and loss (G″) modulus values against frequency of short-time roasted chocolate spreads

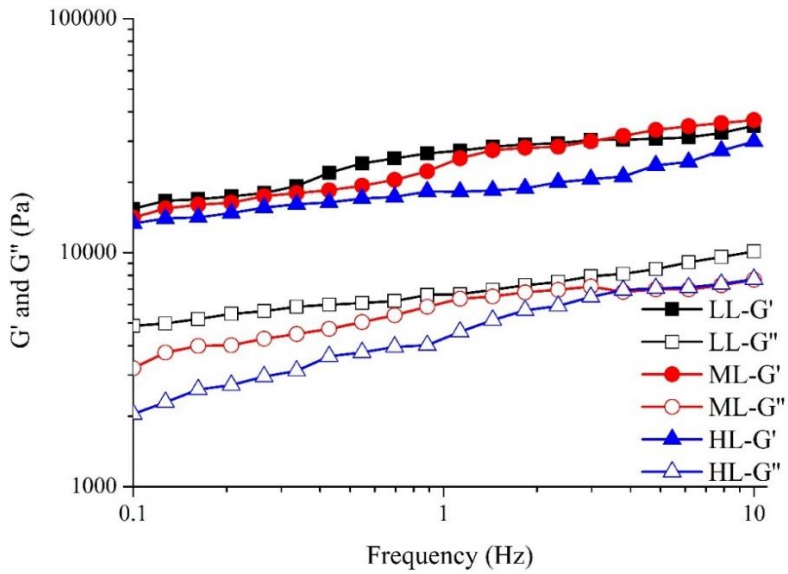


Figure 4.6 Storage (G′) and loss (G″) modulus values against frequency of long-time roasted chocolate spreads

4.6 Colour Analysis Results

The colour analysis results of the spread samples are given in Table 4.8. L^* values of the test samples varied between 27.60–29.13. The L^* values between the spread samples were similar in Low and Medium temperatures batches and the temperature and time conditions of pumpkin seed roasting had no effect on the L^* value ($p>0.05$) among these four samples. However, roasting pumpkin seeds at high temperature significantly decreased the L^* value of chocolate spreads ($p<0.05$). Pumpkin seeds roasted at high temperature caused the colour of the chocolate spreads to darken, that is, to approach black colour.

Sattari (2023), Ertuğrul (2023), Coşar (2021), Acan *et al.* (2021), Şahin and Özata (2022), Tarakçi and Yildirim (2021) and Aydemir (2019a) reported that the L^* values of hazelnut creams with cocoa were in the range of 27.57–28.39, 27.13–28.70, 29.20–34.36, 24.1–27.9, 18.50–32.73, 41.13–42.37 and 19.34–30.48, respectively.

Table 4.8 CIE Colour parameters of chocolate spread with pumpkin seed

Chocolate spreads	*L**	*a**	*b**
LS	29.13±0.01b	5.43±0.07a	5.73±0.01a
LL	28.80±0.01b	5.53±0.16ab	5.59±0.13a
MS	28.55±0.00b	5.66±0.02abc	5.90±0.01a
ML	28.87±0.01b	5.63±0.17abc	5.93±0.05a
HS	27.78±0.01a	6.02±0.05c	7.03±0.16b
HL	27.60±0.01a	5.97±0.12bc	6.97±0.02b

a-b Mean values within each column followed by different letters are significantly different ($p<0.05$)

The *a** values of the test samples ranged between 5.43–6.02. A significant difference was found between the *a** values of chocolate spreads ($p<0.05$). This difference was significant between the samples roasted at low and high temperatures. It was observed that *a** value increased as the roasting temperature of pumpkin seeds increased. This situation occurred due to the caramelization and Maillard reactions with temperature effect and the colour of the pumpkin seeds approaching towards red colour. Sattari (2023), Ertuğrul (2023), Coşar (2021), Acan *et al.* (2021), Şahin and Özata (2022), Tarakçi and Yildirim (2021) and Aydemir (2019a) reported that *a** values of hazelnut creams with cocoa were in the range of 5.10–5.22, 4.88–5.74, 5.84–7.76, 9.61–11.60, 4.01–6.87, 14.09–14.23 and 6.02–11.60, respectively. The *b** values of the test samples varied between 5.59–7.03. A significant difference was found between the *b** values of chocolate spreads ($p<0.05$). The *b** values between the spread samples were similar in Low and Medium temperatures batches and the temperature and time conditions of pumpkin seed roasting had no effect on the *b** value ($p>0.05$) among these four samples. However, roasting pumpkin seeds at high temperature significantly increased the *b** value of chocolate spreads ($p<0.05$). It was observed that *b** values increased as the roasting temperature of pumpkin seeds increased. This situation occurred due to the caramelization and Maillard reactions with temperature effect and the colour of the pumpkin seeds approaching towards yellow colour. Sattari (2023), Ertuğrul (2023), Coşar (2021), Acan *et al.* (2021), Şahin and Özata (2022), Tarakçi and Yildirim (2021) and Aydemir (2019a) reported that *b** values of hazelnut creams with cocoa were in the range of 4.92–5.35, 4.47–5.69, 3.21–6.76, 1.04–3.93, 6.14–12.75, 14.29–14.49 and 5.46–14.70, respectively.

Sattari (2023) stated that roasting temperature and time conditions affected the hazelnut colour to a visually perceptible extent. However, it did not change the colour of chocolate spread. The reason for this is the relatively low proportion of hazelnut in the cream composition (13%), but more importantly, the effect of cocoa, which is the main determining factor on colour, has been the main determining factor. Depending on the cocoa type and production/fermentation conditions, cocoa is the main component that most significantly determines the colour of hazelnut creams with cocoa.

4.7 Sensory Analysis Results

Sensory analysis result values of the chocolate spreads based on panellist scores are presented in Table 4.9 and Figure 4.7. In the odour analysis, the panellists were asked to focus on the general chocolate spread odour and pumpkin seed odour. The general odour scores varied between 2.60 and 3.54. While the spread ML had the highest general odour score, spread LS was rated with the lowest score by the panellists. The general odour scores of the chocolate spreads were similar ($p>0.05$). Pumpkin seed odour scores were found to be between 1.69 and 2.51. The highest pumpkin seed odour score was obtained in the spread HS. No difference was observed between the pumpkin seed odours of the chocolate spreads ($p>0.05$). It was observed that the prominent samples in terms of general odour and pumpkin seed odour were spreads ML and HS.

In the evaluation of flavour and aroma, the panellists were asked to focus on the general chocolate spread flavour and pumpkin seed aroma and to score these two parameters. It was found that the overall flavour scores varied between 3.01 and 4.00. Sample LL had the highest overall flavour score and sample HL had the lowest score. There was no statistical difference between the general flavour scores of chocolate spreads ($p>0.05$). Pumpkin seed flavour scores were found to be in the range of 2.00–3.25. The highest pumpkin seed flavour score was obtained by sample HL. The pumpkin seed flavour score of sample HL was significantly higher than the score of sample LS ($p<0.05$). No difference was observed

between the other samples (p>0,05). It was determined that the pumpkin seed flavour increased as the roasting temperature and time increased.

In the examination in terms of structure and texture, the panellists were asked to focus on spreadability and consistency properties of chocolate spreads and to score these two parameters. As it is known, spreadability is another parameter that consumers attach importance to in chocolate spread. It was determined that the spreadability scores varied between 3.84 and 4.25. The spreadability scores of the spread samples were similar (p>0.05). The selected pumpkin seed roasting temperature and time conditions were not found to be effective on spreadability (p>0,05). The consistency scores of the spreads were found to be in the range of 3.77–4.19. No difference was observed between the consistency scores of the spread samples (p>0,05). The selected pumpkin roasting temperature and time conditions were not found to be effective on consistency (p>0.05). The main determinant of spreadability in chocolate spreads, which is a fat–based suspension product, is the type of oil and the degree of saturation of the fat. In this context, since the oil used was the same, the spreadability and consistency scores were similar. These findings support the texture analysis results.

In the examination of colour and appearance, the panellists were asked to focus on the colour and appearance characteristics of chocolate spreads and to score these two parameters. A bright, homogeneous colour suggestive of cocoa colour and a smooth and uniform appearance are desirable in such products. It was found that the colour scores varied between 3.36 and 4.31. The colour scores of the spread samples were similar (p>0.05). The selected pumpkin seed roasting temperature and time conditions were not found to be effective on colour (p>0.05). This is also consistent with our colour analysis results. The main determinants of colour in chocolate spreads are cocoa and oil. In this context, since the cocoa and fat used were the same, the colour scores were similar. The appearance scores of the spreads were found to be in the range of 3.42–4.42. A difference was found between the appearance scores of the spread samples (p<0.05). The appearance score of spread HS was

significantly higher than that of spread LL ($p<0.05$). No difference was found between the other samples ($p>0.05$).

40% of the panellists detected a burnt pumpkin seed flavour only in the spread HL. Panellists did not detect raw pumpkin seed flavour in any of the samples.

Table 4.9 Sensory parameter values of spread samples

Chocolate spreads	ODOUR	
	General Odour	**Pumpkin Seed Odour**
LS	2.60±0.15a	1.69±0.31a
LL	2.90±0.23a	2.35±0.10a
MS	2.79±0.46a	1.83±0.05a
ML	3.54±0.21a	2.45±0.33a
HS	3.43±0.32a	2.51±0.26a
HL	2.92±0.30a	2.10±0.35a
	FLAVOUR AND AROMA	
	General Flavour	**Pumpkin Seed Aroma**
LS	3.34±0.23a	2.00±0.12a
LL	4.00±0.00a	2.38±0.07ab
MS	3.03±0.59a	2.52±0.15ab
ML	3.61±0.39a	2.70±0.08ab
HS	3.65±0.10a	2.75±0.25ab
HL	3.01±0.12a	3.25±0.26b
	STRUCTURE AND TEXTURE	
	Spreadability	**Consistency**
LS	4.08±0.30a	3.95±0.17a
LL	4.02±0.35a	3.84±0.28a
MS	4.25±0.25a	4.19±0.19a
ML	4.12±0.01a	3.87±0.24a
HS	4.30±0.08a	3.99±0.24a
HL	3.84±0.28a	3.77±0.10a
	COLOUR AND APPEARANCE	
	Colour	**Appearance**
LS	3.60±0.15a	3.49±0.26ab
LL	3.36±0.14a	3.42±0.08a
MS	3.85±0.40a	3.72±0.28ab
ML	4.00±0.00a	3.82±0.07ab
HS	4.31±0.19a	4.42±0.08b
HL	3.94±0.06a	4.01±0.12ab

a-b Mean values within each column followed by different letters are significantly different ($p<0.05$)

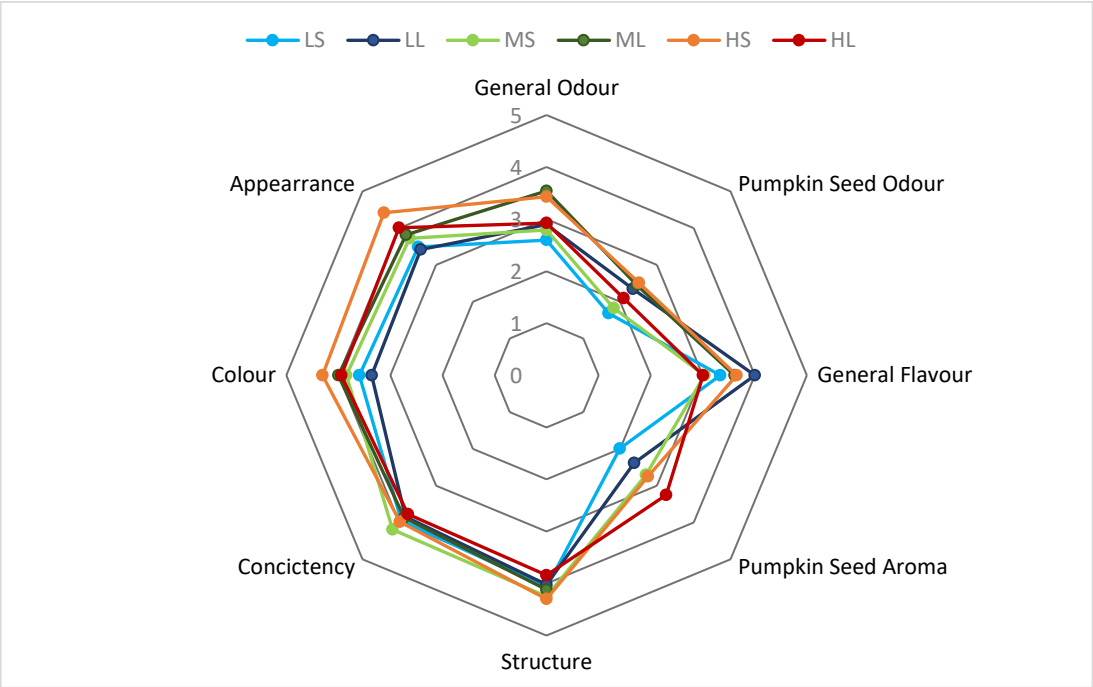


Figure 4.7 Sensory analysis results of chocolate spreads with pumpkin seed

5. CONCLUSION

In this thesis, using different roasting temperature (low: 120 ºC, medium: 150 ºC and high: 180 ºC) and time (short: 20 minutes and long: 40 minutes) norms, the effect of pumpkin seed roasting temperature and time on the physical, textural, rheological and sensory properties of chocolate spread was investigated.

Although the AOS values were quite close between the chocolate spread samples, it was observed that there was a statistically significant difference between the spread samples ($p<0.05$). It was observed that as the roasting temperature of pumpkin seeds increases, the oil separation decreases, that is, the oil binding capacity increases. In the study, the highest oil separation was found in spreads roasted at low temperature ($p<0.05$).

In general, there is a tendency to decrease in water activity with increasing roasting temperature and time, but the only difference between the samples was observed between spreads HS and LS ($p<0.05$). There was no statistical difference between other spreads in terms of water activity values ($p>0.05$). The closeness of the roasting temperature and time combinations in the products and the low percentage (13%) of pumpkin seeds used did not affect the moisture levels and therefore there was no difference in terms of water activity in chocolate spreads ($p>0.05$).

No significant difference was observed between the textural parameters of firmness, spredability, stickiness and adhesivity values of chocolate spread samples ($p>0.05$). Pumpkin seed roasting temperature and roasting time had no effect on textural parameters.

No statistical difference was found between chocolate spread samples in terms of PSD parameters d(0.1), d(0.5), d(0.9), D[3.2], D[4.3] and span values ($p>0.05$). Pumpkin seed roasting temperature and time conditions had no effect on particle size parameter values ($p>0.05$).

Pseudoplastic flow type, which is non-Newtonian flow, was observed in all chocolate spread samples. It was observed that the apparent viscosity values decreased with shear rate. The shear stress and viscosity values of the samples were found to be similar ($p>0.05$). The yield stress ($\tau 0$) values obtained from the Casson model were analysed, no significant difference was observed between the spread samples ($p>0.05$). In terms of plastic viscosity values, the only significant difference was that spread LL showed a higher plastic viscosity value than the other samples ($p<0.05$). A significant difference was found between the infinite viscosity values of the chocolate spread samples ($p<0.05$). This difference was found only between spreads ML and HS. Spread HS gave lower infinite viscosity value. The infinite viscosity values of other spread samples were similar ($p>0.05$). No significant difference was observed between the characteristic shear rate values of the spread samples ($p>0.05$). In all spread samples, G' values showed a higher variation than G'' values along the entire frequency line. It was observed that elastic characteristic was more effective than viscous characteristic chocolate spread samples.

Roasting pumpkin seeds at high temperature significantly decreased the L^* value of chocolate spreads ($p<0.05$). Pumpkin seeds roasted at high temperature caused the colour of the chocolate spreads to darken, that is, to approach black colour. It was observed that a^* value increased as the roasting temperature of pumpkin seeds increased. This situation occurred due to the caramelization and Maillard reactions with temperature effect and the colour of the pumpkin seeds approaching towards red colour. It was observed that b^* values increased as the roasting temperature of pumpkin seeds increased. This situation occurred due to the caramelization and Maillard reactions with temperature effect and the colour of the pumpkin seeds approaching towards yellow colour.

While the spread ML had the highest general odour score, spread LS was rated with the lowest score by the panellists. The general odour scores of the chocolate spreads were similar ($p>0.05$). The highest pumpkin seed odour score was obtained in the spread HS. No difference was observed between the pumpkin seed odours of the chocolate spreads ($p>0.05$). It was observed that the prominent samples in terms of general odour and pumpkin seed

odour were spreads ML and HS. Sample LL had the highest overall flavour score and sample HL had the lowest score. There was no statistical difference between the general flavour scores of chocolate spreads ($p>0.05$). Pumpkin seed flavour scores were found to be in the range of 2.00–3.25. The highest pumpkin seed flavour score was obtained by sample HL. It was determined that the pumpkin seed flavour increased as the roasting temperature and time increased. The selected pumpkin seed roasting temperature and time conditions were not found to be effective on spreadability and consistency ($p>0.05$). The colour scores of the spread samples were similar ($p>0.05$). The selected pumpkin seed roasting temperature and time conditions were not found to be effective on colour ($p>0.05$). The appearance score of spread HS was significantly higher than that of spread LL ($p<0.05$). No difference was found between the other samples ($p>0.05$).

As a result, in this study, no statistically significant difference was observed in textural and rheological tests of different degrees and durations of heat treatment applied to pumpkin seeds on chocolate spread. However, it was determined that spreads HS and ML were superior in terms of general odour, pumpkin seed odour, general flavour and pumpkin seed aroma in sensory tests. Spread HL was found to be unsuccessful due to burnt flavour defect. It is suggested that roasting at 150 °C for 40 minutes or 180 °C for 20 minutes or roasting temperatures and times close to these temperatures and times can be applied in the production of chocolate spread with pumpkin seeds.

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