NOVEMBER 2019 M.Sc. Food Engineering BÜŞRA BELTEKİN

REPUBLIC OF TURKEY

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GRADUATE SCHOOL OF NATURAL & APPLIED SCIENCES

EFFECT OF CALCIUM CHLORIDE APPLICATION ON SOME PHYSICAL AND SENSORIAL PROPERTIES OF PUMPKIN DESSERT

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IN

FOOD ENGINEERING

BY

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in

Food Engineering

Gaziantep University

Supervisor

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GAZİANTEP UNIVERSITY
GRADUATE SCHOOL OF NATURAL & APPLIED SCIENCES
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I hereby declare that all information in this document has been obtained and presented in accordance with academic rules and ethical conduct. I also declare that, as required by these rules and conduct, I have fully cited and referenced all material and results that are not original to this work.

Büşra BELTEKİN

ABSTRACT

EFFECT OF CALCIUM CHLORIDE APPLICATION ON SOME PHYSICAL AND SENSORIAL PROPERTIES OF PUMPKIN DESSERT

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Pumpkin dessert is one of the most popular desserts in our country. In this study, effects of calcium chloride on sensorial and physical properties of pumpkin dessert was investigated. *Cucurbita maxima* was used to obtain traditional pumpkin dessert. Response surface methodology was applied to optimize pumpkin dessert conditions. Temperature (20-40 °C), calcium chloride concentration (0.50-2.50 %), and holding time (10-60 minute) were the independent process variables. The optimum conditions of pumpkin dessert were based on the maximum and minumum levels of responses. It was found that calcium chloride concentration had higher effect on hardness of the pumpkin dessert than temperature and holding time. On the other hand, temperature and holding time increased L* values of pumpkin dessert increased. For total carotenoid content analysis, the increase in temperature and holding time lead to lower total carotenoid content. The optimum temperature, calcium chloride concentration and holding time were estimated as 29.89^{0}C, 2.14 % and 56.22 minute for pumpkin dessert. The maximum and minmum levels of responses under optimum conditions were hardness 28.31 N, L* 45.75, a* 9.2, b* 28.44, 112.77 μg/ml. It was found that temperature, holding time and calcium chloride concentration were important parameters in production of pumpkin dessert.

Key Words: Pumpkin Dessert, Calcium Chloride, Hardness, Color, Optimization

ÖZET

KALSİYUM KLORÜR UYGULAMASININ BAL KABAĞI TATLISININ BAZI FİZİKSEL VE DUYUSAL ÖZELLİKLERİ ÜZERİNE ETKİSİ

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52 sayfa

Kabak tatlısı ülkemizdeki en popüler tatlılardan biridir. Bu çalışmada, kalsiyum klorürün balkabağı tatlısının duyusal ve fiziksel özellikleri üzerine etkisi araştırılmıştır. Geleneksel balkabağı tatlısı elde etmek için *Cucurbita maxima* kullanıldı. Kabak tatlısı koşullarını optimize etmek için tepki yüzey metodolojisi uygulandı. Sıcaklık (20- 40 °C), kalsiyum klorür konsantrasyonu (0.50-2.50 %) ve bekleme süresi (10-60 dakika) bağımsız proses değişkenleri olarak alındı. Klasiyum klorür konsantrasyonun, balkabağı tatlısının sertliği üzerinde sıcaklığa ve bekletme süresine kıyasla daha etkili olduğu tespit edildi. Öte yandan, sıcaklık ve bekletme süresi arttıkça balkabağı tatlısının L * değerinin de arttığı gözlendi. Toplam karotenoid içerik analizi için sıcaklık ve bekletme süresindeki artış, daha düşük toplam karotenoid içeriğine yol açtı. Balkabağı tatlısı için optimum sıcaklık, kalsiyum klorür konsantrasyonu ve bekletme süresi 29.89^{0}C, 2.14 % ve 56.22 dakika olarak hesaplandı. Optimum koşullar altında maksimum tepki seviyesi sertlik 28.31 N, L * 45.75, a * 9.2, b * 28.44, 112.77 µg/ml idi. Balkabağı tatlısı üretiminde, sıcaklık, bekletme süresi ve kalsiyum klorür konsantrasyonun önemli etkileri olduğu saptandı.

Anahtar Kelimeler: Balkabağı Tatlısı, Kalsiyum Klorür, Sertlik, Optimizasyon

To my beloved family...

Sincerely yours...

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LIST OF SYMBOLS

A	Absorbance
^{0}Brix	Total soluble solids
Da	Dalton
E 440	Pectin
KCl	Potassium Chloride
K_3PO_4	Tripotassium Phosphate
mM	Millimolar
nm	Nanometer
w/v	Weight/volume
w/w	Weight/weight
µg	Microgram
µm	Micrometer
µmol	Micromole

LIST OF ABBREVIATIONS

ANOVA	Analysis of variance
BHT	Butylated hydroxy toluene
DPPH	2,2-dipheynl-1-picrylhydrazyl
EDTA	Ethylenediamine Tetraacetic Acid
FAOSTAT	Food and Agriculture Organization of the United Nations
FRAP	The Ferric Reducing Ability of Plasma
GAE	Gallic acid equivalents
HPLC	High Performance Liquid Cromotograpy
SEM	Scanning Electron Microscopy
THF	Tetrahydrofuran
TUIK	Turkish Statistical Institute

CHAPTER 1

1.1 Introduction

Pumpkin is one of the most popular fruits due to its healthy and nutritious ingredients. Pumpkin is a seasonal product that is used as an animal feed together with the indispensable part of human diet. It is a highly productive product grown in tropical and subtropical countries (İncedayi et.al., 2009). Pumpkin which belongs to the Cucurbitaceae family, has 119 type and 825 species in the taxonomic classification (Hyun-Il Jun et.al., 2006). Of these, the most in the world species with economical prescription; Citrullus, Cucumis, Cucurbita and Lagenaria species, which are known as *Citrullus lanatus* (*Citrullus lanatus (Thunb.) Matsum & Nakai*), *Cucumis sativus L.*, *Cucumis melo L.*,*Cucurbita pepo L.*, *Cucurbita maxima Duch.*, *Cucurbita moschata duch.* and *Lagenaria siceraria* (*Molina*) *Standl* (Figure 1.1).

Cucurbita maxima

Cucurbita pepo

Cucumis melo L

Cucurbita moschata

Lagenaria siceraria

Cucumis sativus L

Figure 1.1 Some species of Cucurbitaceae

Studies on the history of *Cucurbita moschata* were reportedly cultivated first in the west of Mexico (3400 BC) and later in the southwest of the USA (900 BC). With the

arrival of Spanish explorers in America, species diversity moved to Japan and then to Asia. On the other hand, there is no evidence before *Cucurbita moschata* reach Europe in the 17th century. The rich climatic diversity that our country has enabled a number of cucurbit species to be cultivated in our country without problems (Balkaya et al., 2010). Production of pumpkin is demostrated according to years in Turkey in Table 1.1 (TUIK).

Table 1.1 Production of pumpkin in Turkey (TUIK)

Pumpkin	Ton
2009	82.552
2011	93.099
2012	93.612
2013	95.076
2014	93.672
2015	95.363
2016	96.268
2017	89.737

Pumpkin is used to obtain juice, pomace, dried product, cake, filling material in pie, bread and soup and dessert, generally consumed as dessert in Turkey. Also, there exist several researches about the improvement of pumpkin based foods as its use in enlarged snack and breakfast cereal products. Pumpkin is also utilized as the thickener, flavoring and coloring agent (Dirim, 2012). Pumpkin has wide production area all around the world. China is first in the world in pumpkin production, followed by India, Russia, Ukraine, USA and Mexico respectively (Table 1.2, FAOSTAT, 2017).

Table 1.2 Worldwide pumpkin production (FAOSTAT, 2017)

Country	**Ton**	**Country**	**Ton**
China	8,051,495	Algeria	307,861
India	5,142,812	Korea	299,804

Russia	1,165,834	Rwanda	290,134
Ukraine	1,164,660	Philippines	287,425
USA	1,091,121	Argentina	266,878
Mexico	710,632	Pakistan	264,150
Spain	702,278	Peru	197,203
Italy	590,541	Cameroon	193,092
Turkey	580,624	Japan	190,238
Indenosia	566,857	Morocco	179,519
Malawi	545,769	Iran	151,392
Cuba	537,545	Niger	141,979
South Africa	433,966	Canada	136,286
Egypt	428,335	Germany	133,561
Bangladesh	340,908	Thailand	125,002

1.2 General Properties of Pumpkin

Pumpkin is a one-year culture fruit. It can be long cylindrical, flat round and pear shaped. The color of the crust is yellow, orange yellow and orange, and the color of the beef can change from light orange to dark orange. The shells are flat, along with grooved and sliced ones. Their average weight ranges from 5 to 60 kg (Kaya, 2006). 60-70% of the root of the pumpkin is found at a depth of 30 cm. The roots of the flowers can be reduced to 1-1.5 m depth. The flowers are best and most fruitful in the pH range between 6,5-7,5 pH. climatic conditions must be mild and require. Temperatures are between 18-27°C. Approximately 3-4 months are required to survive (Özel, 2010).

Pumpkin (*Cucurbita maxima*) is a seasonal crop that has been used traditionally both as dietary food and animal feed (Incedayı et al., 2009). Pumpkin health benefits have been proven as a result of investigations. Pumpkin is very rich in minerals such as fiber, calcium, potassium, phytosterols, polyunsaturated fatty acids, antioxidant

vitamins such as carotenoids and tocopherols, zinc and trace elements such as zinc and the sodium content is quite low. The lifophylic components such as carotenoids in pumpkin give orange color to pumpkin and pumpkin makes it a beneficial nutrient. Carotenoids play a role as free antioxidants and free radical scavengers and play an important role in cancer prevention. The sense of sight increases the absorption of Vitamin A, which is essential for growth and embryo development. (İlgöy Gözükara, 2013). Carotenoids are known to be the main source of vitamin A. Deficiency of Vitamin A is one of the widespread causes of blindness and infant mortality, and is a major health problem in some parts of Southeast Asia, Africa, South and Central America. Carrots, potatoes, parsley, chicory, red pepper, spinach, lettuce, mago, apricot and pumpkin have high carotenoid contents (Drammeh et al.,2002 and Benadi et al.,2003). These fruits and vegetables, as well as pumpkins are very rich in carotenoids and are used in diets such as vision impairment in vitamin A deficiency. It has been proven that diet with rich pumpkin reduces risk of stomach, chest, lung and colon cancer and pumpkin oil reduces hypertension and high cholesterol risk (İlgöy Gözükara, 2013). β-carotene, small amounts of α-carotene, lycopene, lutein and trace amount of cyrptoxanthin and cis-β-carotene are the major carotenoids in pumpkin (Zhou et al., 2007). Chemical composition of fresh pumpkin is showed at Table 1.3 (Kaya, 2006; Gözükara, 2013).

Table 1.3 Chemical composition of fresh pumpkin (Kaya, 2006; Gözükara, 2013)

Nutritional value	**Unit**	**Amount/100 g**
Water	g	91.60
Energy	kcal	26
Protein	g	1.00
Total fat	g	0.10
Ash	g	0.80
Carbohydrate	g	6.50
Fiber	g	0.5
Mineral		
Calcium	mg	21
Iron	mg	0.80

Magnesium	mg	12
Phosphor	mg	44
Potassium	mg	340
Sodium	mg	1
Zinc	mg	0.32
Copper	mg	0.127
Manganese	mg	0.125
Selenium	µg	0.3
Vitamin		
Vitamin C	mg	9.0
Thiamine	mg	0.050
Riboflavin	mg	0.110
Niacin	mg	0.600
Pantothenic acid	mg	0.298
Vitamin B_6	mg	0.061
Total folate	µg	16
Folic acid	µg	0
Vitamin B_{12}	µg	0.00
Vitamin A	IU	1600
β_caroten	mcg	3100
α_caroten	mcg	515
Lutein+zeaxantin	mcg	1500
Retinol	µg	0
Vitamin A, RAE	µg_(RAE)	80
Vitamin E	mg (ATE)	1.060
Amino acid		
Tryptophan	g	0.012
Threonine	g	0.029
Isoleucine	g	0.031
Leucine	g	0.046
Lysine	g	0.054
Methionine	g	0.011
Cysteine	g	0.003

Phenylalanine	g	0.032
Tyrosine	g	0.042
Valine	g	0.035
Arginine	g	0.054
Histidine	g	0.016
Alanine	g	0.028
Aspartic acid	g	0.102
Glutamic acid	g	0.184
Glycine	g	0.027
Proline	g	0.026
Serine	g	0.044

In Table 1.4, pumpkin, apricot, carrot, pumpkin and tomato products were compared in terms of their nutritional content. It contains more calcium, α-carotene, β-carotene, Vitamin A than pumpkin, zucchini, tomato and apricot. Sodium content is very low, especially when compared to carrots. The content of potassium and Vitamin E of pumpkin is more than carrots, apricots, zucchini and tomato products (Gözükara, 2013).

Table 1.4 Nutritional component of some fruits (Gözükara, 2013)

Component (100 g)/Product	Pumpkin	Zucchini	Carrot	Apricot	Tomato
Calcium(mg)	21	15	33	13	10
Potassium(mg)	340	262	320	259	237
Sodium(mg)	1	2	69	1	5
α-caroten(mcg)	515	0	3477	19	101
β-caroten(mcg)	3100	120	8285	1094	449
Vit A (IU)	7384	200	16811	1926	833
Vit E (mg)	1,06	0,12	0,66	0,89	0,54

1.3 Application of Calcium Chloride Salt on Foods

1.3.1 General Information About Calcium

Calcium is one of the most important minerals for human health. 99% of the calcium contained in our body is located in our bones and teeth, while the remaining 1% is in

the blood, which provides blood clotting, structuring the joints, regulating heartbeats and helping to repair the nervous system (Altıner and Şahan, 2016). Dietary calcium is highly effective in prevention of disease and health promotion. For increased absorption of calcium, Vitamin D plays an active role as a compound. Vitamin D is converted to dihydroxycholecalciferol, which is an active hormone form in the and kidney. This active hormone works in harmony with the calcium-binding protein and parathyroid hormone and directly activates the absorption of calcium in the small intestine and regulates the elevation of phosphorus absorption (Tayfur, 1991). Deficiency of this nutrient has been caused chronic diseases, implicating osteoporosis, osteomalacia, hypertension and colon cancer (Anino, S.V et al., 2006). 50-73% of the total Ca intake in humans is obtained from milk and milk products. 25-27% of total Ca intake is supplied by cereal products, fruits and vegetables. Nowadays in industrialized societies, enrichment of foods processings have been applied to eliminate nutritional deficiencies and related health problems. One of the most common applications of this is mineral enrichment. Calcium, magnessium, iodine, iron, zinc, selenium are some major minerals, which used commonly in the world, in enrichment processes in order to protect human health. In England flour has begun to enrich with Ca in 1943. Calcium carbonate, calcium citrate, calcium lactate, calcium chloride, calcium phosphate, calcium propionate, calcium ascorbate, calcium gluconate are calcium salts using to enrichment of foods. Flour, juice, breakfast cereals, milk and milk products, chocolate, cake, muffin are some of the processing foods (Altıner and Şahan, 2016).

1.3.2 General Properties of Calcium Chloride

Calcium chloride is one of the ionic salts in solid form at room temperature. It will be produced directly from limestone as well as by Solvay method. Due to its hygroscopic structure, calcium chloride must be kept in air-tight containers. According to the Communiqué on the Purity Criteria of Additives Other than Colorants and Sweeteners Used in Foodstuffs of the Turkish Food Codex (2002/28), the following definition is made for calcium chloride: E 509; Einecs: 233-140-8; chemical formulation is $CaCl_2.nH_20$ (n=0, 2 or 6); molecular weight is 110.99 (anhydrous), 147.02 (dihydrate), 219.08 (hexahydrate); white, odorless, hygroscopic powder or dust when air contacted crystals and the content of it on anhydrous basis should not be less than 93.0%. Anhydrous calcim chloride is freely soluble in water

and ethanol. Dihydrate form is freely soluble in water, soluble in ethanol and hexahydrate is very soluble in water and ethanol. Calcium chloride, acts as a origin of calcium ions in solution and its inability soluble calcium compounds, causing precipitation.

$$3CaCl_2 + 2K_3PO_4\ (aq) \rightarrow Ca_3(PO_4)_2\ (s) + 6\ KCl(aq) \quad (1.1)$$

Liquefied $CaCl_2$ can be electrolyzed to obtain calcium metal.

$$CaCl_2(l) \rightarrow Ca(s) + Cl_2(g) \quad (1.2)$$

1.3.3 Role of Calcium Chloride Applications on Foods

Calcium chloride is used with the function of stabilizer, thickener and flavoring agent in cheese, canned fruits and vegetables, jam, marmalade, jelly and beer industry. However, it used in construction, agriculture and mining sectors. Previous studies have found that calcium chloride is effective agent in increasing the strength of many fruits and vegetables. The effects of calcium chloride on fruits and vegetables are delaying aging, reducing post-harvest deterioration, controlling the formation of many physiological diseases. Moreover, the added calcium was observed to increase the nutritional value of the product. The main role of the addition of calcium to increase the resistance to softening is to stabilize the membrane systems and increase the hardness and strength of the cell wall by providing calcium pectate formation (Güldaş and Dağlıoğlu,2008).

1.4 Calcium Pectate

1.4.1 Pectin

Pectin is a complex polysaccharide and an important structural constituent of plant cell wall (King, 1993). These substances are highly polysaccharides with high water retention capacity consisting of complex and colloidal carbohydrate derivatives consisting mainly of anhydrogalacturonic acid units. In this case, the unit forming the pectic substances is polygalacturonic acid, and they make α-1,4 bonds with each other to make a straight chain. They are heteropolysaccharides with molecular masses of 30000 - 300000 Da. Pectic substances are a general name given to a group of substances including pectinic acid, pectin, pectic acid and salts of them. (Willants et al. 2001). Figure 1.3 shows that the structure of cell wall (Chan et al., 2017).

Pectin are frequently associated with other cell wall components such as cellulose or hemicellulose in plant tissues (Chan et al., 2017). Pectin is produced in the plant's Golgi apparatus (Kashyap *et al.* 2001). It is also a natural component of the human diet, where it contributes to soluble fibre (Maxwell et al., 2012). Pectin was first discovered in apple juice by Vauquelin (1790) and named by Braconnot (1825a,b), borrowing from the Greekword pektikos which means to congeal or solidify.

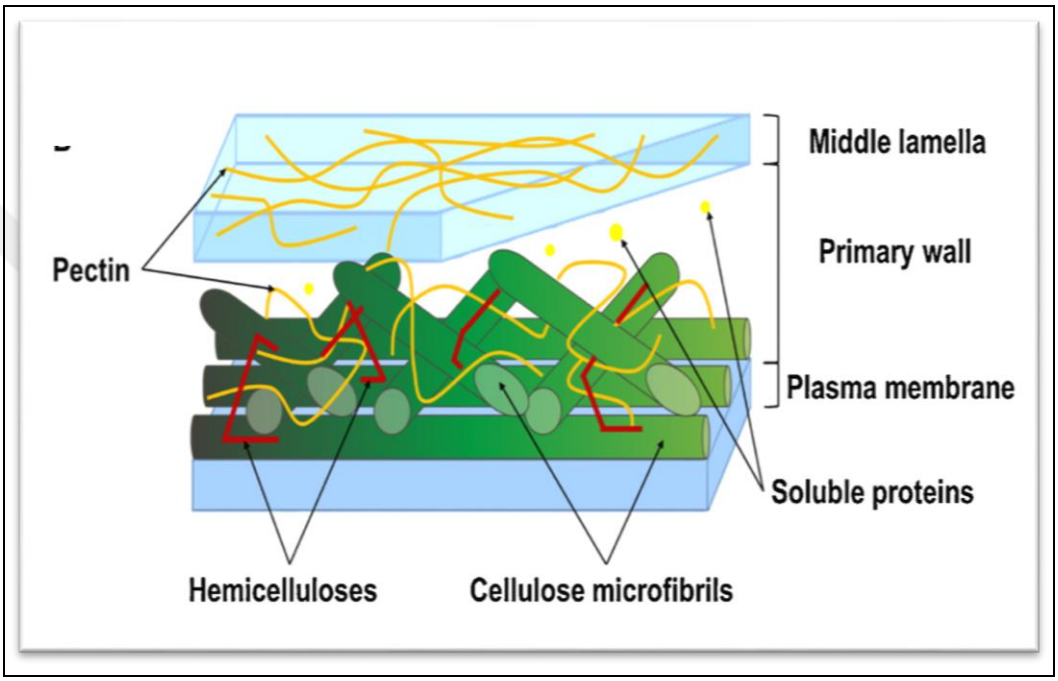

Figure 1.2 Structure of cell wall (Chan et al., 2017)

Although more than 210 years have elapsed since the discovery of pectins, the chemical and structural properties of this polymer family have been investigated. To begin with the basic structure, the pectin is combined by at least 17 different monosaccharides of which D-galacturonic acid (GalA) (Figure 1.3, Chan et al., 2017) is the most plentiful, followed by l-arabinose, d-galactose, l-rhamnose and others (Kaya et al., 2014 and (Chan et al., 2017)). These monosaccharides can be interconnected through 20 different link-ages (Kaya et al., 2014).

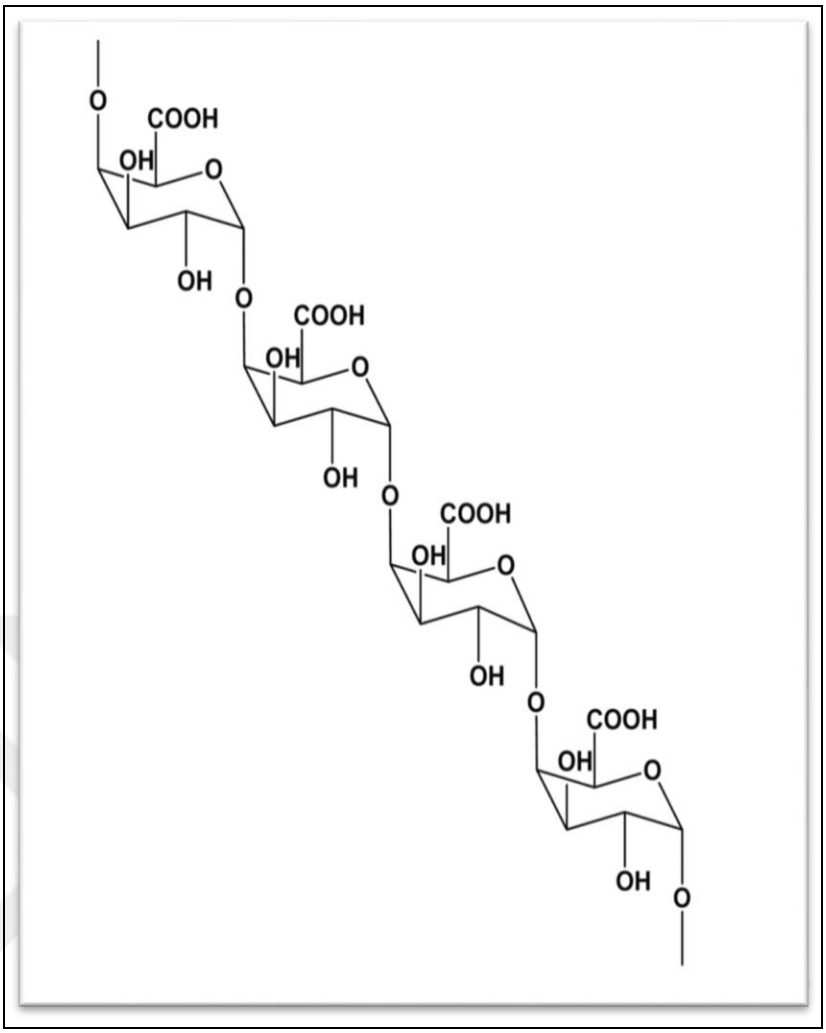

Figure 1.3 α-1,4-linked D-galacturonic acid (GalA) units (Chan et al., 2017)

Within GalA monomers, the carboxylicgroups or hydroxyls may be methyl-esterified (Chan et al., 2017). Figure 1.4 shows that Methylated GalA (Chan et al., 2017).

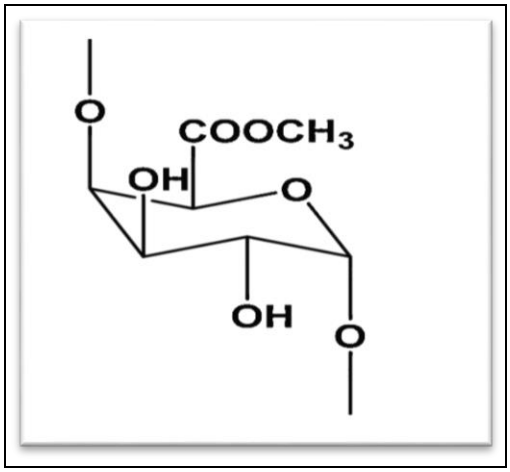

Figure 1.4 Methylated GalA (Chan et al., 2017)

Pectin, which is the substrate of pectinase enzyme, was examined with scanning electron microscopy (SEM) by Maraş et al.(2004). The crystal size of pectin was 3 μm in x14000 magnification view. It is shows that in Figure 1.5 (Maraş et al., 2004).

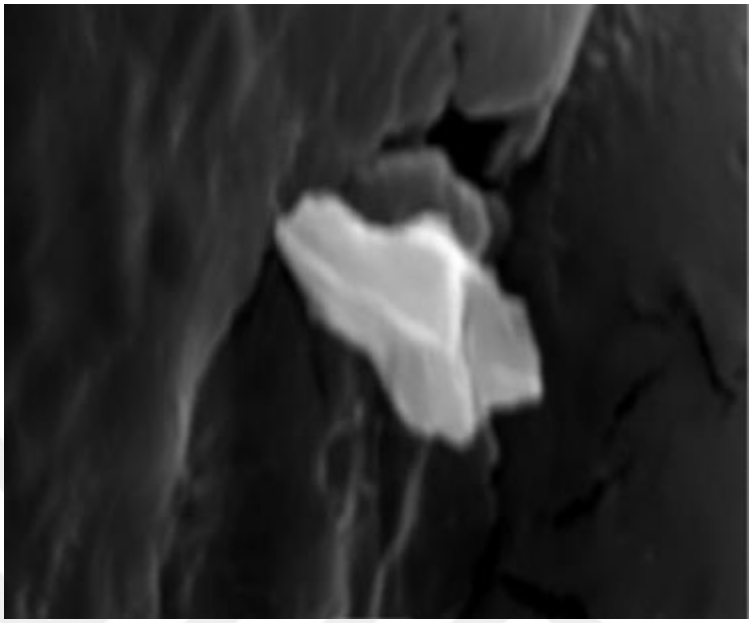

Figure 1.5 SEM images of pectin (Maraş et al., 2004)

Pectin is used in the food industry to provide the production of jam and marmalade, canned fish, ice cream, mayonnaise and sauce (Aslan, 1994). Pectin (E 440) is a food additive, which is particularly important in fruit and vegetable technology, especially in the production of clear juice (Ekşi, 1988).

Pectin is obtained industrially from apple and citrus fruit pulp. Pectin was originated in the 1900s in Germany when an apple juice manufacturer tried to cook dried apple pomace, the by-product from apple juice processing (Ciriminna et al., 2015). The protopectin in the crude material is solubilized by acid extraction and then the soluble pectin is separated from the aqueous solution with isopropyl alcohol. Pectin is used as a liquid or dried powder in trade (Acar and Gökmen, 2004).

Ptitchkina et al. (1994) studied the gelation properties of high methoxyl pectin and used pumpkin pectin. The properties of pectin obtained from pumpkin were compared with citrus pectin and the degree of esterification of both pectins were found to be close to each other, and pumpkin pectin was found to have better gelling properties. Pectin is also used in as an emulsifier, stabilizer and/or thickener (Codex Alimentarius, 2015). All of these functional terms are called ‘rheology modifiers’.

Pectin has beneficial effects on health because it contains dietary fiber. Pectin is slightly digested in the small intestine and fermented by colon bacteria in the large intestine. Pectin has prebiotic effect due to this function (Yılmaz et al., 2016).Table 1.5 shows that pectin content of some fruits (Aehle, 2004; Kaya, 2006).

Table 1.5 Pectin content of some fruits (Aehle, 2004; Kaya, 2006)

Fruit	Pectin, %
Apple	0.5-1.6
Currant	1.0-1.2
Grape	0.1-0.4
Orange Peel	3.5-5.5
Pear	0.7-0.9
Pineapple	0.04-0.1
Strawberry	0.5-0.7

1.4.2 Calcium Pectate

Pectic substances may chemically change during ripening of fruits. As maturation proceeds, pectinic acid is hydrolyzed to pectic acid. Therefore, a weak gel structure is formed in jams and marmalades produced from extremely ripe fruits (McCready, 1966; Yigit, 1975; Whitaker et al., 1984). The active pectin methlyesterase enzyme converts the pectin molecule into the calcium pectate structure (Figure 1.6, Özel, 2009).

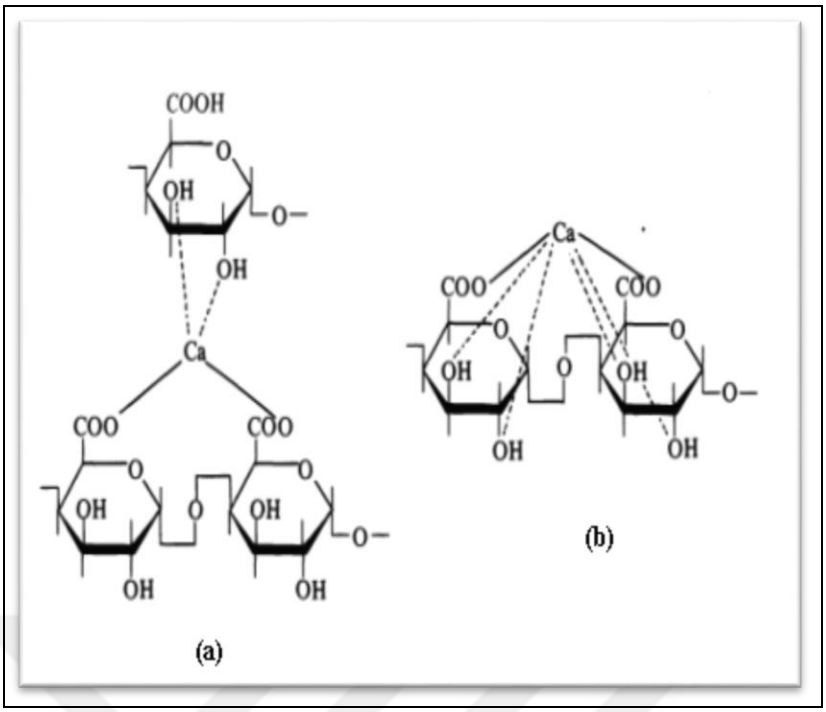

Figure 1.6 Calcium binding to the pectin molecule (Özel, 2009)

a) intermolecular binding

b) intramolecular bonding

1.5 Literature Survey

It has been briefly mentioned some scientific studies about calcium chloride and some different salts how they are applied in different vegetables and fruits.

Turhan et al. (2018) aimed to determine effects of irrigation water salinity and Ca^{2+} applications on carrot (Daucus carota L.) yield and some quality characteristics of in their study. In addition to five different sodium chloride concentrations (0, 20, 40, 60 and 80 mM) in irrigation water, trials were repeated with 0, 4 and 8 mM Ca^{2+} applied to soil for each NaCl level. According to the results of the study, the marketable yield of carrot decreased as irrigation water increased salinity. In the quality parameters (fruit water content, water soluble dry matter, reducing sugar, β-carotene and vitamin C), the negative effects of salt were determined at increasing salt concentrations of 20 mM. Ca^{2+} application of the carrot water soluble dry matter, reducing sugar, β-carotene and vitamin C in the content of the negative effects of salt was provided. Calcium applications had no positive effect on marketable carrot yield, fruit water content and total acid values. Accurate management of Ca^{2+} application can be used to improve some quality parameters of carrot.

Jan et al. (2016) studied effect of different calcium salt concentration with diffent storage time on the some properties of stored apple. 0, 3, 6, and 9 % $CaCl_2$ solution was prepared and apples were immersed for 3, 6, 9, and 12 min to these solutions, then stored for 150 days at 5 ± 1°C with 60–70% relative humidity. Percent weight loss, total sugar, ascorbic acid and firmness of apples were determined after the storage time. Results showed that Ca content of samples increased importantly during the storage time. Percent weight loss increased during the 150 days storage but holding in the $CaCl_2$ solution controlled weight loss. Total sugar content (%) was increased during storage; in this study, total sugar content of apples were decreased by $CaCl_2$ solution. The lowest amount was observed in apples that holding in 9 % of $CaCl_2$ solution. Ascorbic acid content decreased during the storage and maximum amount of ascorbic acid was observed at 9 % concentration of $CaCl_2$ solution. Firmness of apples were decreased significantly during storage but it was retained with calcium chloride treatment, the higher the $CaCl_2$ concentration lead to the more firm apples.

Mahmud et al. (2015) studied CaC_2 and different calcium salt effects on ripening of mango in Bangladesh. They used four mango types harvested in *Bangladesh - lokhna-mangefera indica, hemsagor-mangefera indica, gopalvogh – mangefera indica,* and *langra- mangefera indica.* Fruits were immersed different concentration (0, 2.5, 5.0, 7.5 and 10 %) of calcium chloride ($CaCl_2$), calcium sulphate ($CaSO_4$) and calcium ammonium nitrate $Ca[NH_4NO_3]_2$ and calcium carbide (CaC_2) for 10 min. The fruits were ripened at 27°±4°C in the paper carton–boxes lined and covered with newspapers. Results showed that CaC_2 had highest positive effect on ripening of mango fruits but it is carcinogenic chemical and highly hazardous for human health. Sensory properties were changed with storage time. Color of the skin was turned to the yellowish color, but not uniform color with increasing concentration. Flavor, pulp color and aroma showed different results. 2,5 and 5 % concentration caused to same as control but less attractive results. They investigated deep yellow and less attractive results as a result of 7,5 and 10 % concentration treatments and the taste of fruits of in all concentrations were determined some portion sweet and some sour while control was sweet but not sour. According to results calcium chloride was more effective when compared other salts and it delayed ripening 3.5 days than untreated control group, calcium sulphate delayed 3.00 days and and calcium ammonium

nitrate had no effect on mango. After 6 days taste of fruits was satisfactorry for all concentrations. CaC_2 treatment caused to spoilage of 75% of mango fruits after 1 day and after 10 days 25% of mango fruits were spoiled result of $CaCl_2$ and $CaSO_4$ treatment. After 7 days 25% of mango fruits were spoiled when treated $Ca[NH_4NO_3]_2$. Aroma, flavor, texture properties was found good but not better both $Ca[NH_4NO_3]_2$ and $CaCl_2$. $CaSO_4$ showed better taste conversely CaC_2 not so good. Hardness and softness was observed CaC_2 causing very very soft texture and other salts was found like as control group.

Chong et al. (2015) investigated chitosan, calcium chloride, and their combination on delaying weight loss, improved firmness, delayed colour changes and moderately inhibited microbial growth of postharvest fresh-cut honeydew melon. Treatments with 2% chitosan, 1% $CaCl_2$ and combination of 2% chitosan and 1% $CaCl_2$ showed that importantly decreasing weight loss, evolved firmness, delaying colour changes, and inhibiting microbial growth on fresh-cut honeydew melon during storage time (13 days at $7 \pm 2\ ^0C$) when compared to control group. The best result was obtained combination of 2% chitosan and 1% $CaCl_2$ all properties of honeydew melon.

Khaliq et al. (2015) researched how gum arabic covering combined with calcium chloride affects some properties of mango (*Mangiferaindica L.*) fruit during low temperature storage. In this study gum arabic (GA) 10% , calcium chloride (CA) 3% + gum arabic 10% were used, fruits were stored at $6 \pm 1\ ◦C$ and $90 \pm 3\%$ relative humidity for 28 days and then transferred to $25 \pm 2\ ^oC$ for 5 days shelf life were examined. Control group was distilled water. According to results the combined treatment of calcium chloride 3% + gum arabic 10% significantly relieved the increase in decay relation. Morever, weight loss, color changes, soluble solid concentration (SSC), respiration rate, ethylene production and maintained high firmness, titratable acidity and ascorbic acid were effeciently decreased by only gum arabic 10% or combined with calcium chloride 3%. GA 10% and CA 3% + GA 10% treated groups indicated higher firmness when compare the control fruits. Weight loss of untreated mango fruits was notable ($p \leq 0.05$) higher than treated fruit after 14 days up to the end of storage period. SSC of control fruit was found highest after CA 3% and CA 3% + GA 10% treated fruits at the end of storage period, but these treatments were not notable different. GA 10% significantly ($P \leq 0.05$) hindered ascorbic acid loss when compared to the control fruits after storage time. The loss of

ascorbic acid of control fruits was the highest. There were no notable differences in ascorbic acid between GA 10% and CA 3% + GA10% treatments along the storage period.

Karemera and Habimana (2014) studied on effect of pre-harvest calcium chloride application on some post harvest behavior of mango fruits (Alphonso). Different concentration of $CaCl_2$ and spray day groups were determined and these groups were; control (no spray), 0.50 % spray of calcium chloride at 30 days before harvest, 1.00 % spray of calcium chloride at 30 days before harvest, 1.50 % spray of calcium chloride at 30 days before harvest, 0.50 % spray of calcium chloride at 15 days before harvest, 1.00 % spray of calcium chloride at 15 days before harvest and 1.50 % spray of calcium chloride at 15 days before harvest. According to results they observed the best result from the last group, 1.50 % spray of calcium chloride at 15 days before harvest. 1.50% $CaCl_2$ at 30 days before harvest retarded the ripening 19.22 days, 1.00% $CaCl_2$ sprayed trees at 30 days before harvest and it delayed 18.33 days and 1.50% $CaCl_2$ sprayed trees at 15 days before harvest delayed 18.44 days. The mango fruits ripened much earlier 14.77 days on normal (no sprayed) trees.

Incedayi et al. (2009) researched on the dessert produced from modified atmosphere packaged pumpkins. Firstly pumpkin was minimally handled and stored after modified atmosphere packaging. Processed pumpkins were cut into small pieces and put into 1500 ppm Na-metabisulphide + 1% NaCl solution. The cut pieces were divided into two groups and both of them were diptreated with 150 ppm Na-hypochloride for 5 minutes and washed with tap water. Then when the first group was immersed into the citric acid (1.5%) solution for 5 minutes; the second group was submerged into the L-cysteine (0.5%) solution for 10 minutes. Approximately 200 g samples were packed in polypropylene dishes and BOPP film packages. Packages were also separated into two groups. The first group was stamped with 80% vacuum with 80% N_2 and the second group was stamped with 80% vacuum with 70% N_2+ 10% CO_2. Samples were stored at 4±2°C. After 10 days storage, traditional Turkish dessert called as “kabak tatlısı” were cooked with these pumpkins and chemical composition and organoleptic properties of both of the pumpkins and the desserts were analyzed. They found that during the storage period, ascorbic acid, total phenolics and total carotenoids content of pumpkins decreased. Color values of the samples were very close to each other. The samples treated with citric acid and

packaged under nitrogen atmosphere contained higher amount of carotenoids and had higher a value. Nontheless, there was not important difference between b and L values of the samples during the storage period.

Ergun and Ergun (2007) investigated extension shelf life of sliced cucumber with modified atmosphere packaging and calcium lactate application. Cucumbers (Cucumber sativus L. 'Beith Alpha') were cut into slices after being peeled and stored in modified atmospheric conditions at 4 ºC for 7 to 10 days. When positive results were obtained from modified atmosphere packaging (MAP), cucumber slices were added to calcium lactate (5%) solution, calcium lactate (5%) + ascorbic acid (5%) was immersed in solution or water only and maintained at the same temperature under modified atmosphere conditions. Weight loss or dehydration was found to be one of the most important factors in decreasing quality parameters in sliced cucumber and it was decided that MAP was the best way to prevent this. MAP increased the shelf life of sliced cucumbers at 4 ºC from 2 days to 5 to 7 days. Softening was delayed by calcium applications. Calcium lactate application resulted in 9 days of shelf life. Consequently, MAP was found to increase the shelf life of calcium lactate together with quadrupling the shelf life.

Ergun and Ergun (2006) examined in their study the reactions of grated carrot to calcium applications. Grated carrots were immersed in 5% calcium lactate concentration, 5% calcium chloride concentration or water (control) for 5 minutes and stored at 4 ºC for 8 to 9 days. During storage, sensory changes in taste, friability and odor were determined. In addition, the amount of dry matter dissolved in water was determined. In both calcium treatments, loss of friability and malodour were delayed. But calcium chloride caused little bitterness when compared to other treatments. While the amount of dry matter dissolved in water was not affected by calcium applications, the decrease in pH value was prevented by calcium application. Calcium treatments also delayed the blacking of grated carrots. According to the results, shelf life of grated carrots at 4 ºC can be increased by calcium applications, especially calcium chloride applications. The non-demanding odor development was blocked or suppressed by both calcium lactate and calcium chloride. Calcium lactate delayed the development of odor for 4 days while calcium chloride did not allow an undesirable odor development during storage.

Garcia et al. (2006) stated that dipping strawberries in calcium chloride solution after harvest could extend the shelf life of strawberries. The most effective application is to immerse in 1% calcium chloride solution at 25 °C for 15 minutes. It was observed that the hardness of the fruit did not change significantly during the first days of dipping applications and at the end of 7 days fruit hardness increased.

Anjum and Ali (2004) studied on the effect of different calcium salts on Pakistani mango. They used calcium chloride ($CaCl_2.2H_2O$), calcium sulphate ($CaSO_4.2H_2O$) or calcium ammonium nitrate {$Ca(NH_4NO_3)_2$} solutions. Fruits were immersed for 10 minutes in 2.5, 5.0 or 7.5% these solutions and then fruits were stored at ambient temperature 25 ± 3 ^{0}C. According to results calcium chloride was more effective than other salts, it retarded about 3.33 days later than the control fruits, calcium sulphate was retarded only for 2.67 days, calcium ammonium nitrate was not enough effective. Calcium chloride at the concentrations of 2.5 % and calcium sulphate at 5.0 and 7.5% acts as the same and delayed ripening for 2 days. On the other hand, calcium chloride at the concentrations of 5.0 and 7.5% and calcium sulphate at 7.5% were the most effective and mango in these processing ripened 4 days later than control group. Calcium ammonium nitrate was only effective at lower concentration of 2.5% and retarded ripening for only 2 days. 5% $CaCl_2$ solution delayed in ripening for 4 days and had better skin and pulp colour but it negatively affected the eating quality.

Mishra (2002) determined calcium application 6-year period at the end, pear trees yielded an increase of over 13%. Fruit weight, quality and lowered the product value in the fruit reduced the formation of deterioration. At the same time, the process reduced the abnormal hardening of certain regions by more than 50%.

Desrumaux et al. (2000) studied about to extent of shelf life of mushroom after postharvest and during storage period. They showed the mushroom yield was not influenced by the use of hard, middle-hard or relatively soft irrigation water. Yields tended to be smaller as the calcium chloride dose added to the irrigation water was increased from 0 to 0.4%. The mushroom color at picking was not influenced by the type of water or by the administering of calcium chloride. During conservation, it had been found that $CaCl_2$ had a positive influence on the mushroom color. The color improvement was based on a slowing down of the yellowing and a brighter color.

The effect was only noticeable in the 2nd flush after long-lasting conservation (6 days), in older flushes somewhat earlier (4 days). The mushroom dry matter level was slightly positively influenced by the administering of calcium chloride. The mushroom calcium level increases as a result of the calcium chloride administering.

Qi et al. (2000) researched effect of different salts and cooking temperature on banana and plantain texture. Dessert banana was cooked in distilled water with different temperature (60, 70, 80, 90, and 100 °C) for 10 min. Furthermore, dessert banana and plantain pulp were cooked at 100 °C for 15 min with different concentrations; 0.5% (w/v) $CaCl_2$, 0.5% (w/v) NaCl, or 0.5% (w/v) ethylenediaminetetraacetic acid (EDTA). They found that cooking dessert banana at 60-70 °C for 10 min had little effect on pulp firmness than at 80-100 °C. when the temperature reached at least 80°C, pulp rupture force of dessert banana decreased importantly, by 29%. In addition, they observed cooking at 90 °C caused 36% loss of firmness and cooking at 100 °C in distilled water caused the softest pulp texture, the firmness decreasing by 77% by the end of the cooking. When compared the salts, it was resulted 0.5% $CaCl_2$ significantly more firmness than cooking in distilled water, 0.5% NaCl or 0.5% EDTA For dessert banana, they examined that $CaCl_2$ caused only 44% loss of the original firmness compared to 67.5% in distilled water, 66.4% in NaCl and 71.4% in EDTA solution.

Alonso et al. (1997) researced preventing on the lossing of firmness, color and turgidity of sweet cherry after freezing. For this aim they applied different pretreatments with and without $CaCl_2$ solution. Fruits were immersed at 50 ^{0}C for 10 min. with or without by immersion in 100 mM $CaCl_2$ and thermal pretreatment at 70 ^{0}C for 2 min with or without dipping in 100 mM calcium chloride solution to prevent firmness loosing. According to results there was not any significant differences in color of fruits between the control and pretreatment groups. Freezing method causes loss of firmness was greatest in the control cherries. The firmness of skin had not significant differences than control groups. However, fibrosity and firmness of pulp, were significantly higher in cherries treated at 70 ^{0}C for 2 min and then immersed in 100 mM $CaCl_2$.It was detected that generally cherries with thermal treatments especially with $CaCl_2$ firmer than controls.

Schirra et al. (1994) determined maturity of mandarins with calcium chloride application it can be delayed and the storage time can be extended. In the study, while the fruit was still green in the tangerine trees, 4 - 6% calcium chloride was sprayed four times in the same day and the harvested fruits were stored at 5^0C and 90% relative humidity. Fruits were followed for 75 days of cold storage; changes in weight loss and ascorbic acid content were determined. The control group not sprayed with calcium chloride was deteriorated after 30 days, while the group treated with $CaCl_2$ remained intact until day 75. The amount of ascorbic acid in fruits not being applied to calcium was gradually decreased and the value reached on the 30th day was reached on the 75th day in those who received calcium.

Robson et al. (1989) and Souza (1999) showed that the application of 1% $CaCl_2$ solution at a pressure of 3 psi increased the ripening time of the peaches by approximately 30% 14 days. Application of $CaCl_2$ in peach led to a decrease in enzyme levels and an increase in sugar levels during the harvest period. Besides, browning (decay) decreased by 34% and disease index decreased by 29%. The spraying process during the growth period of peach trees has a positive effect on the product quality. It was determined that the fruits of the calcium treated fruits retained their quality longer than those not applied.

Mishra (2002) and Luna-Guzman et al. (1999) found that the rate of respiration was decreased in the melon slices treated with 2.5 % $CaCl_2$ and the hardness of the slices that were processed was significantly higher than the control group. Striping, rod-shaped cutting and slicing methods in foods increase the absorption of calcium due to the increase of surface area.

Izumi and Watada (1994) showed that, the value of carrots texture immersion with the process; compared to the control group, it was 69% for those stored at 0 ^{0}C and 93% for those stored at 5 ^{0}C. Weight loss rates decreased by 2.5 times in the group stored at 0 ^{0}C and by 33% in those stored in 5 ^{0}C. The 1% calcium solution helped to reduce the microbial load in the carrot strips as it increased the resistance in the tissue.

Carvalho et al.(2014) assessed the total carotenoid, α- and β- carotene and 9 and 13-Z-β- carotene isomers contents in C. moschata after some different cooking processes. For this aim, samples of raw different type of pumpkins were peeled and

divided into four parts by two longitudinal cuts, to obtain four sections. From these sections, they discarded the two sections opposite from each other, and the remaining two sections were used in the cooking.. Two landrace pumpkins of *C. moschata* were washed, peeled off, cut in small pieces, and divided into four different groups; the pumpkin pieces were then cooked in boiling water (2:1 for 4 minutes), steamed cooked (5 minutes), or cooked with a 60% sucrose solution in 200 mL of water. After that they analyzed total amount of carotenoids in the pumpkin samples using a spectrophotometer at 450 nm. Their results showed that the samples cooked in boiling water contained 258.50, 184.80, 43.97, 6.80, and 0.77 μg.g–1 of total carotenoids, β-carotene, α-carotene, 13-*Z*-β-carotene, and 9-*Z*-β-carotene, respectively. The steamed samples contained 280.77, 202.00, 47.09, 8.23, and 1.247 μg.g–1 of total carotenoids, β-carotene, α-carotene,13-*Z*-β-carotene, and 9-*Z*-β-carotene, respectively. The samples cooked with sugar had 259.90, 168.80, 45.68, 8.31, and 2.03 μg.g–1 of total carotenoid, β-carotene, α-carotene, 13-*Z*- β-carotene, and 9-*Z*- β-carotene, respectively. The total carotenoid and carotenoid isomers increased after the cooking methods, most likely as a result of a higher availability induced by the cooking processes.

1.6 The Aim of The Study

Pumpkin is known to include high amount of beneficial component for human health in particular carotenoids. The major carotene in pumpkin is β- carotene, with small amounts of α- carotene, lutein, lycopene, and trace amount of cryptoxanthin and cis-β-carotene. There are several studies about the carotenoids, hardness, color, total phenolics, total sugar determination after $CaCl_2$ treatments of pumpkin fruit however about some properties of pumpkin dessert, studies have been insufficient. Pumpkin dessert is so popular dessert in Turkey. Incedayi et al. (2009) examined production of dessert from modified atmosphere pumpkins and determined some properties of dessert such as total carotenoids, ascorbic acid, total pheolics and color values during storage time. Carvalho et al. (2014) investigated that total carotenoid, α- and β-carotene, 9 and 13-Z-β- carotene isomers contents in C. moschata by cooking with sucrose solution. The objectives of this study are also to investigate effect of calcium chloride treatment on physical and sensorial properties of pumpkin dessert and to optimize temperature, holding time and calcium chloride concentration conditions of pumpkin dessert by use of response surface methodology.

CHAPTER 2

MATERIALS AND METHODS

2.1 Materials

Fruit of this study was provided from Bingöl, East Anatolian, Turkey. Pumpkin seeds were gotten from Bolu, West Blacksea, Turkey and cultivated in Bingöl. Plants were harvested in mid November 2017 (Figure 2.1).

Figure 2.1 Preharvest pumpkin

Calcium chloride dihydrate was purchased from VWR ProlaboChemicals. Tetrahydrofuran (THF) for high performance liquid cromotography (99.9 %) was provided from Chemsolute. Metanol was purchased from VWR Prolabo Chemicals. Butylated hydroxy toluene, BHT (2,[6]-Di-tert-Butyl-p-cresol) (min 99.0 %) was purchased from Sigma.

2.2 Experimental Design and Data Analysis for Pumpkin Dessert

The central composite design (CCD) for three independent variables was performed for pumpkin dessert. The independent variables were calcium chloride concentration (0.50-2.50 %), temperature (20-40 ^{0}C) and holding time (10-60 min). The complete

design was composed of 19 runs, including five replications of the center points for the three independent variables. The outline of experimental design and the independent variables is represented in Table 2.1. Dependent variables are hardness, color and total carotenoid as responses. Response surface methodology extensively use in food industries for product development and process optimization when there are two or more factors affecting the responses of product (Koh et al., 2017).

Table 2.1 Design of experiment

Run	Temperature (^{o} C)	Time (min)	Concentration (%)
1	**24.05**	**49.87**	**0.91**
2	**30.00**	**35.00**	**1.50**
3	**30.00**	**35.00**	**1.50**
4	**30.00**	**35.00**	**2.50**
5	**30.00**	**60.00**	**1.50**
6	**24.05**	**20.13**	**2.09**
7	**35.95**	**20.13**	**2.09**
8	**24.05**	**49.87**	**2.09**
9	**30.00**	**35.00**	**1.50**
10	**35.95**	**20.13**	**0.91**
11	**30.00**	**10.00**	**1.50**
12	**35.95**	**49.87**	**0.91**
13	**24.05**	**20.13**	**0.91**
14	**30.00**	**35.00**	**1.50**
15	**20.00**	**35.00**	**1.50**
16	**35.95**	**49.87**	**2.09**
17	**30.00**	**35.00**	**0.50**
18	**30.00**	**35.00**	**1.50**
19	**40.00**	**35.00**	**1.50**

2.3 Preperation of Pumpkin Dessert

Pumpkin fruit was washed with tap water to remove foreign materials from it. The cleaned pumpkin was cut and seperated from its peels and seeds, then minimized in

cube form approximately 3x3x3 cm^3 by hand. Pumpkin cubes were put in plastic bag and stored in refrigarator at -18 °C until using (Figure 2.2).

Figure 2.2 Cutted and stored pumpkin

Stored pumpkin cubes were prepared in different conditions (Figure 2.3). All groups were hold their temperature, calcium chloride concentration (w/v) and holding time before cooking. Then 20 °Brix sucrose syrup (2:1) (w/w) was prepeared and pumpkins cooked in this sucrose syrup.

After that some of different pumpkin desserts were used for sensory, hardness and color analysis (Figure 2.4). Others were putted in plastic bags, and stored at -18 °C until using other analysis (Figure 2.5).

Figure 2.3 Frozen raw pumpkin cubes

Figure 2.4 Different types of pumpkin dessert

Figure 2.5 Pumpkin dessert in plastic bags

2.4 Methods

2.4.1 Sensory Evaluation of Pumpkin Dessert

Pumpkin desserts were organoleptically evaluated for their hardness, color, taste and general pleasant properties with 7 panelists. All parameters were scored from 1 to 5. Hardness, color, flavor and general appreciation parameters were analyzed by 15 panelists.5 degrees for hardness (5= very hard, 4= hard, 3= medium, 2= soft, 1= very soft), 5 degrees for color ((5= red, 4= orange, 3= light orange, 2= yellow, 1= light yellow), 5 grade for taste (5= very sweet, 4= sweet, 3= normal, 2= tasteless, 1= very unpleasant) and 5 degrees for general appreciation (5= very like, 4= like, 3= normal, 2= not liked, 1= not at all) hedonic scales in the form of were used.

2.4.2 Hardness Analysis

Hardness analysis of pumpkin desserts was determined with Agrosta (Digital Firmness Tester, France) and recorded carefully.

2.4.3 Color Measurement

Color determination of all types of pumpkin desserts was done on transparent surface by Konica Minolta (Color Reader, CR-10 Japan). The instrument was standardized before measurement. The color values were expressed as L^* (lightness) y-axis, a^* (redness/greenness) x-axis, and b^* (yellowness/blueness) z-axis (CIE, 1976)

2.4.4 Determination of Total Carotenoid Content

For extraction of carotenoids; 0,5 g of sample was taken, 3 ml of THF-methanol (50:50) and % 0,1 BHT was added. This mixture was homogenized with ultraturrax (Heidolph Instruments, D-91126 Schwabach, Germany) and supernatant was filtered with hydrophilic PTFE syringe filter (pore 0,45 nm, diameter 25 mm) to glass cuvette. Then absorbance was readed at 450 nm by spectrophotometer (UV-1800 Shimadzu Corporation, Kyoto, Japan). Calibration curve of β- carotene was demostrated in Figure A.1.

2.4.5 Statistical Analysis

All experiments data were analyzed by using Design expert (Stat- Ease, Version 7.0.0, 2004) according to response surface methodology. All results were compared by using the analysis of variance (ANOVA). The differences between individual means were counted to be significant at $\alpha = 0.05$.

CHAPTER 3

RESULTS AND DISCUSSION

Studies with pumpkin dessert aimed to determined sensory, hardness, color and total carotenoid of pumpkin dessert. The effect of independent variables on some properties of pumpkin dessert was investigated. The results were given Table 3.1. For this target, multiple regression analyses were succeeded using response surface analysis to fit mathematical models to the experimental data and define the relationship between three independent variables and the response variables.

Table 3.1 Experimental results for responses of pumpkin desserts

Run	Hardness(N)	L*	a*	b*	Total carotenoid(µg/ml)
1	17±3.21	50.63±0.5	10.4±0.36	26.1±0.15	92.8±2.42
2	27±2.65	47.67±0.58	6.47±0.06	23.8±0.50	112.6±1.45
3	24±1.00	46.5±0.58	6.66±0.15	24.17±0.38	116.3±2.9
4	29.33±3.05	41.57±0.51	6.9±0.07	25.83±0.15	117.7±0.97
5	24.33±1.15	45.2±2.63	9.63±0.2	29.3±0.42	117.7±1.93
6	26.67±2.52	43.5±3.21	7.26±0.12	29.1±0.23	149.5±14.98
7	25.33±2.52	45.23±1.60	8.63±0.21	28.7±0.15	142.3±14.49
8	29.67±2.52	46.06±1.19	8.9±0.15	30.06±0.15	117.7±1.93
9	25±2.00	46.57±0.51	6.96±0.36	23.6±0.60	120.1±2.41
10	16.33±1.53	47.23±3.03	9.03±0.41	28.2±0.1	89±0.97
11	21±2.65	46.23±0.51	8.86±0.15	24.4±0.3	148.5±8.7
12	15±2.00	51.47±5.33	8.53±0.15	26.63±0.21	109.8±5.31
13	17.67±2.52	46.26±0.26	7.83±0.21	25.53±0.46	105.7±0.48
14	22.67±3.05	46.26±0.26	8.03±0.21	23.7±0.53	87.3±2.42
15	20±2.00	47.6±0.58	7.33±0.1	28.67±0.15	152.5±5.78
16	24.67±4.04	45.4±0.53	9.23±0.12	27.53±0.45	98.6±5.79
17	15.33±1.53	46.36±1.57	7.56±0.15	21.17±0.38	93.8±0.96

18	26.33±1.53	47.4±1.64	7±0.1	24.4±0.3	96.5±2.9
19	23±1.00	48.3±0.17	10.6±0.25	32.6±0.15	81.8±4.35

3.1 Effect of Independent Variables on Properties of Pumpkin Dessert

3.1.1 Effect of Temprature and Holding Time on Hardness

Effect of temperature and holding time on the hardness of the pumpkin dessert was shown in Figure 3.1. The hardness of pumpkin dessert varied from 15 to 29.67 N. The hardness of control pumpkin dessert which not treated with calcium chloride concentration or temperature and holding time was measured 12 N. As it can be seen from Figure 3.1, increase in temperature, and holding time caused a decrease and increase in the hardness respectively. As shown in equation 3.1, the model obtained for predicting the hardness of pumpkin dessert explained the main linear effects of factors affecting the hardness ($p<0.05$). The lowest hardness value was obtained at the maximum temperature and minimum holding time. At low holding time, the increase in temperature resulted in a decrease in hardness, while at high holding time, the increase in temperature led to decrease in hardness. Morever, at low and high temperature, the increase in holding time caused to sharp increase in hardness. According to Figure 3.1, holding time had higher effect on hardness of the pumpkin dessert than temperature. According to results, higher calcium chloride concentration affected positively on hardness of desserts.

In addition to the main quality factors such as appearance, flavor and nutritional properties, texture is one of the most important parameters for fresh and processed foods. (Gouriou et al., 2017). One of the most frequently seen problems in minimally processed fruits and vegetables is tissue softening (Türkmen and Sarı, 2004) and it is not contributed by consumer acceptability. Jan et al.(2016), investigated the how calcium salts affect soft rot, bitter pit and physicochemical propeties of stored apple. It was reached dipping of apples in 9% calcium chloride solution for different time intervals had considerably affected firmness. At the same time they deduced at 9% concentration and 12 minutes. Thermal treatment notably reduced the pumpkin firmness and energy. Gonçalves et al.(2007), indicated textural parameters decreased as process time and temperature increased, when pumpkin (Cucurbita maxima L.) were blanched in the temperature range of 75–95 °C. Calcium chloride supplied

being harder structure of the pumpkin desserts (Equation 3.1). Pectic substances change during ripening of fruits. The pectin methlyesterase enzyme converts the pectin molecule into the calcium pectate structure. Consequently, calcium pectate makes the cell membrane durable and that causes the cell wall and the middle parts of the fruit to gain resistance, will harden the texture of the pumpkin dessert. Likewise, Mahmud et al. (2015) found out high calcium chloride concentration improved the firmness of mango. In this study control pumpkin dessert that not treated with any calcium chloride concentration or temperature and holding time was found more softer than other treated pumpkin dessert.

Hardness = +22.65 + 7.90*C (3.1)

where C is the calcium chloride concentration.

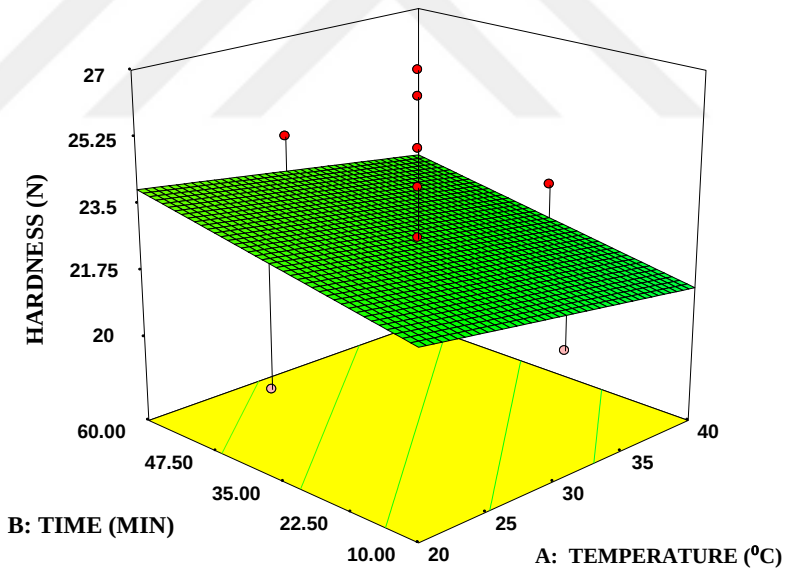

Figure 3.1 Effect of temperature and holding time on hardness of pumpkin dessert

3.1.2 Effect of Temprature and Holding Time on Total Carotenoid Content

Effect of temperature and holding time on total carotenoids of the pumpkin dessert was shown in Figure 3.2. As shown in equation 3.1, the model obtained for

predicting the total carotenoids of pumpkin dessert explained the main linear effects of factors affecting the total carotenoids ($p<0.05$). A negative coefficient means a decrease in response when the level of the variable is increased, whereas a positive coefficient indicates an increase in response. The total carotenoids of pumpkin desserts varied from 81.8 to 152.5 μg/ml. The total carotenoid content of control pumpkin dessert was recorded 124.92 μg/ml. As shown in Figure 3.1 the increase in temperature and holding time lead to lower total carotenoid content. Previous studies showed pumpkin is a rich source of carotenoids, especially in carotene and other carotenoids that are good precursors of Vitamin A (Seo et al., 2005). Carotenoid losses occur during processes such as shredding, grinding, heat treatment, boiling, cooking, drying, freezing, irradiation and storage applied to foods for various purposes (Akdoğan and Özdemir, 2006; Chen et al., 1996; Lin & Chen, 2005). Ribeiro et al. (2015) studied in some style of home cooking on retention and bioaccessibility of pro-vitamin A carotenoids in biofortified pumpkin. They applied boiling, steaming and boiled with sugar processings. The results showed that pro-vitamin A carotenoids of different pumpkin genotypes which cultivated from Brazil had different values. In general boiling process caused decreasing carotenoid of biofortified pumpkin. Boiling with sugar had lower caretonoid than other processed pumpkin and raw samples of all genotypes. Carvalho et al. (2014) found that total carotenoid content of raw pumpkin (Landrace pumpkins (*C. moschata*)), cooked in boiling water, steamed cooked, cooked with additioning sugar groups. They found results as 236.10, 258.50, 280.77 and 251.90 μg.g–1. Dutta *et al.* (2006) showed the total carotenoid content of pumpkin pieces thermally treated for different blanch times. They reported that at temperatures of 85°C and 95°C, 4.13-13.35% decrease in the total carotenoid content was observed when compared with the control. This is because prolonged bleaching at elevated temperatures causes water-soluble carotene-bound protein molecules to infiltrate into the globular form. On the other hand it was observed that calcium fertilizer applications may had positive effects on some quality characteristics such as water soluble dry matter, reducing agent sugar, β-carotene, vitamin C of carrot plants grown in salty conditions (Turhan et al., 2018). Similarly in this study calcium chloride treatment affected positvely the total carotenoid content of pumpkin dessert.

Total carotenoid = + 113.17 – 17.83*A + 18.66*C (3.2)

where A is the temperature, C is the calcium chloride concentration.

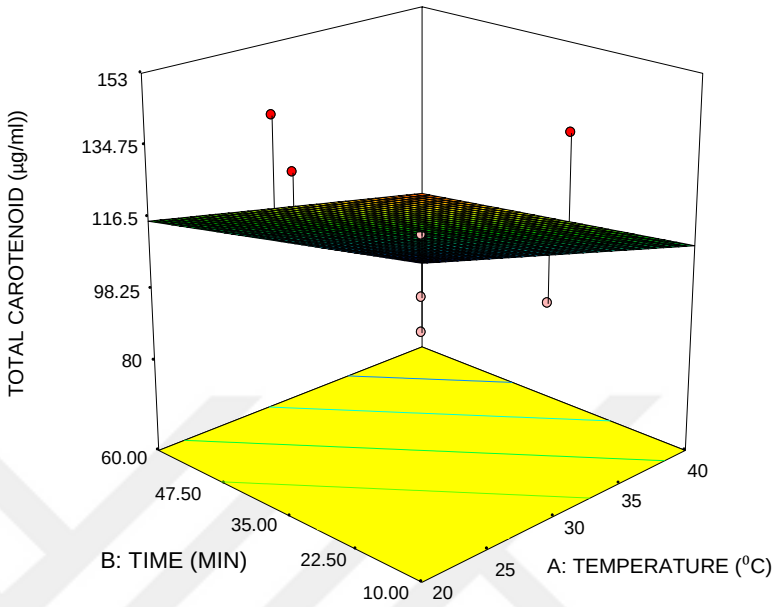

Figure 3.2 Effect of temperature and holding time on total carotenoid of pumpkin dessert

3.1.3 Effect of Temprature and Holding Time on Color

The color of processed products is an important quality factor that reflects sensory attractiveness. Although functional foods can provide consumers with many benefits in terms of health, it cannot be preferred to consumers without visual attraction (Dirim and Çalışkan, 2012). Lightness value, L*, indicates how dark/light the sample is (varying from 0—black to 100—white), a* is a measure of greenness/redness (varying from -60 to +60), and b* is the grade of blueness/yellowness (also varying from -60 to +60) (Gonçalves et al., 2007). L*, a* and b* values were 46.56 ± 0.11, 9.5 ± 0.17, 29.33 ± 0.30 for control pumpkin dessert respectively. Figure 3.3 showed that effect of temperature and holding time on L* values of the pumpkin dessert. L* values of pumpkin desserts changed between 41.57 and 51.47. As shown in equation 3.3, the model obtained for predicting the L* values of pumpkin dessert explained the main linear effects of factors affecting the L* values ($p<0.05$). As indicated that in Figure 3.3 when temperature and holding time increased L* values of pumpkin

dessert increased. Maximum L* values was observed at highest temperature. Similar result recorded by Alonso et al. (1997). They determined that for color changes of frozen sweet cherry which applied thermal pretreatment with calcium chloride and without it. Altough high temperature caused increased color, $CaCl_2$ treatment lead to poor color. As in equation 3.3 calcium chloride negative coefficient indicated a decrease in responses. Also Mahmud et al. (2015) showed that mango which treated with calcium salts (calcium chloride, calcium sulphate and calcium ammonium nitrate) had a poor influence on the pulp color.

$$L^* = +46.58 - 2.66^*C \quad (3.3)$$

where C is the calcium chloride concentration.

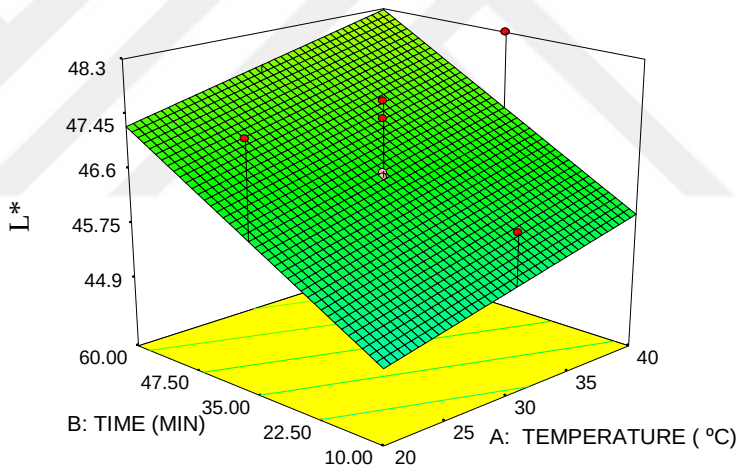

Figure 3.3 Effect of temperature and holding time on L* value pumpkin desserts

The model obtained for predicting the a* values explained the main linear, quadratic and interaction of variables ($p<0.05$, equation 3.4, Figure 3.4). a* values varied between 6.66 and 10.6. Linear effect of temperature (A) had positive effect, quadratic (A^2) and (B^2) had positive effects;

$$a^* = +7.02 + 0.80^*A + 2.04^* A^2 + 2.32^* B^2 \quad (3.4)$$

where A is the temperature, B is the holding time.

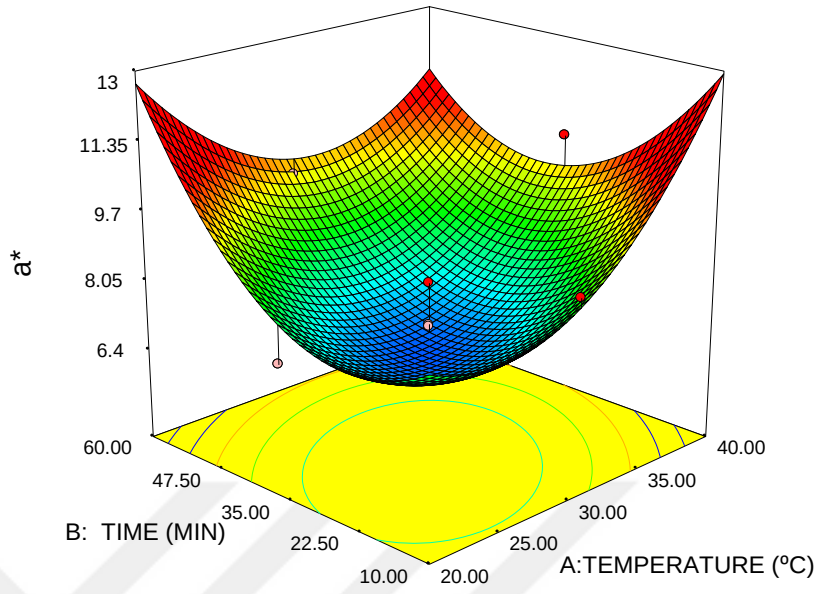

Figure 3.4 Effect of holding time and temperature on a* values of pumpkin dessert

a* value changed negatively with decreasing temperature and time. b* values of pumpkin desserts changes between 23.6 and 32.6. The model obtained for predicting the b* values explained the main linear, quadratic and interaction of variables ($p<0.05$, equation 3.5, Figure 3.5). Linear effect of calcium chloride concentration (C), quadratic (A^2) and (B^2) had positive effects. As a* value, decrease temperature and time caused to low of b* value. Gonçalves et al.(2007) found the effects of blanching treatment on peroxidase inactivation, colour and texture of pumpkin (Cucurbita maxima L.) in the temperature range of 75–95 °C. Coversely our results they determined for blanched pumpkins, CIE L*a*b* and C* colour factors decreased significantly ($P<0.05$) as the time of process and temperature increased. The samples became darker, and lost redness, yellowness and vivid characteristics. These colour alterations may be explained by heat carotenoid degradation.

$$b^* = +23.91 + 2.08*C + 7.03* A^2 + 3.24* B^2 \quad (3.5)$$

where A is the temperature, B is the holding time and C is the calcium chloride concentration.

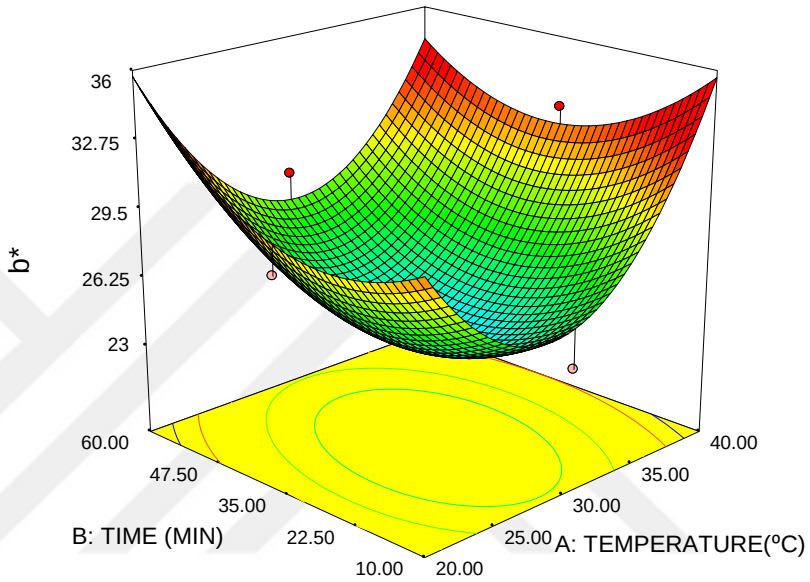

Figure 3.5 Effect of temperature and holding time on the b* values pumpkin desserts

3.1.4 Effect of Temprature and Holding Time on Sensory Properties

Sensory analysis is an important distinguishing factor in minimal and heat- treated foods. Odor was not an important factor affecting the results in this study and therefore was not taken into consideration. Statistically result of sensory analyis of control pumpkin was given in Table 3.2.

Table 3.2 Sensory analysis of control pumpkin dessert

Control Group of Pumpkin Dessert	
Parameters	**Mean±Standart Deviation**
Hardness	1.29 ± 0.49
Color	2.86 ± 0.38
Taste	1.86 ± 0.69
General appreciation	2.57 ± 0.53

Table 3.3 Experimental results of sensory analysis (hardness) of pumpkin desserts

Run	N(Number of Panelist)	Mean± Std. dev.
1	7	3.43±0.98
2	7	3.71±0.76
3	7	3.86±0.69
4	7	4.00±0.58
5	7	3.57±0.53
6	7	4.00±0.00
7	7	3.14±0.38
8	7	3.29±0.49
9	7	3.29±0.49
10	7	2.29±0.49
11	7	2.86±0.69
12	7	2.57±0.53
13	7	2.57±0.53
14	7	3.71±0.76
15	7	4.00±0.00
16	7	3.71±0.49
17	7	1.71±0.49
18	7	3.43±0.54
19	7	3.14±0.38

According to hardness analysis of pumpkin desserts, 8. pumpkin dessert run, was selected more harder than other. This group had temperature 24.05 °C, holding time 49.87 minute and calcium chloride concentration 2.09 %. This result was close to hardness analysis. In many processed foods, a combination of hardening agents such as calcium salts is used to prevent food softening. Ca^{+} combines with the pectic acid in the cell wall to form calcium pectate and protect the cell wall. In addition, stabilization of cell membrane with Ca ions and improving cell turgor pressure cause hardening (Türkmen and Sarı, 2004). All hardness results were given in Table 3.3. In color analyis, 3. and 16. runs had highest color values, the lowests were 2. and 8. runs. These results was not same with color analysis ($p>0.05$). Results were given in Table 3.4.

Table 3.4 Experimental results for sensory analysis (color) of pumpkin desserts

Run	N(Number of Panelist)	Mean±Std.dev.
1	7	2.86±0.69
2	7	2.71±0.49
3	7	3.43±0.79
4	7	2.86±0.38
5	7	3.00±0.00
6	7	2.86±0.89
7	7	3.14±0.38
8	7	2.71±0.49
9	7	3.00±0.58
10	7	3.14±0.38
11	7	3.29±0.49
12	7	2.86±0.38
13	7	3.14±0.69
14	7	3.14±0.69
15	7	3.29±0.49
16	7	3.43±0.53
17	7	3.14±0.69
18	7	3.00±0.58
19	7	3.29±0.49

For taste analysis it was determined that 17. group found more sweeter and this group had low calcium chloride concentration(0.50 %). 4. group pumpkin dessert

was found that most tasteless group and had 2.50% calcium chloride concentration. Anjum and Ali (2004) analyzed immersing mango friuts with different calcium salts solution; calcium chloride ($CaCl_2.2H_2O$), calcium sulphate ($CaSO_4.2H_2O$) or calcium ammonium nitrate {$Ca(NH_4\ NO_3)_2$} solutions. (2.5, 5.0 or 7.5%) for 10 min and determine some quality parameter during storage. The increase in the concentration of calcium salts caused delayed ripening, on the other hand, it was found to have a negative effect on mango quality by increasing the shrinkage of the skin and lowering the flavor and taste of the fruits. Mahmud et al. (2015) showed the distinct effect of some calcium salts and CaC_2 on the ripening of the mango fruits in Bangladesh. According to thier results, concentration of calcium salts had negative effects on taste of the mango fruits. For general appreciation of pumpkin dessert 13. and 17. groups selected better than other. Lower $CaCl_2$ concentration lead to convenient general appreciation and taste. Taste and general appreciation results were given in Table 3.5 and Table 3.6 respectively.

Table 3.5 Experimental results for sensory analysis (taste) of pumpkin desserts

Run	N(Number of Panelist)	Mean±Std. dev.
1	7	2.71±0.49
2	7	2.57±0.54
3	7	2.29±0.76
4	7	1.29±0.49
5	7	2.14±0.38
6	7	1.57±0.54
7	7	1.71±0.49
8	7	2.29±0.76
9	7	2.57±0.54
10	7	3.00±0.58
11	7	2.43±0.53
12	7	2.71±0.49
13	7	2.57±0.54
14	7	1.86±0.69
15	7	1.86±0.69
16	7	1.43±0.54
17	7	3.29±0.49
18	7	2.29±0.49
19	7	2.29±0.49

Table 3.6 Experimental results for sensory analysis (general appreciation) of pumpkin desserts

Run	N(Number of Panelist)	Mean±Std.dev
1	7	2.29±0.49
2	7	2.14±0.38
3	7	2.29±0.49
4	7	1.14±0.38
5	7	2.29±0.49
6	7	1.43±0.54
7	7	2.14±0.38
8	7	1.57±0.54
9	7	2.00±0.00
10	7	2.57±0.54
11	7	1.71±049
12	7	2.57±0.54
13	7	2.86±0.38
14	7	1.86±0.69
15	7	2.43±0.54
16	7	1.43±0.54
17	7	2.86±0.38
18	7	2.43±0.54
19	7	2.57±0.54

3.2 Optimization

Optimization can be defined as the processing conditions that get the optimum (maximum or minimum) value of a function of certain decided variables subject to constraints that are imposed. Optimization may be the process maximizing a desired quantity or minimizing an undesired one. The values of the processing variables that produce the desired optimum value are called optimum conditions (Altan et al., 2008). Product responses such as hardness, color, total carotenoid content were the foremost major parameters determining quality of pumpkin dessert. Therefore, optimum conditions for pumpkin dessert were determined to obtain maximum color, maximum total carotenoid content and hardness. To determine the optimum pumpkin

dessert process conditions, response surface of desirability function was used for numerical optimization. The desirability function of the response surface is shown in Figure 3.6 for obtaining optimal conditions in pumpkin dessert. By applying the desirability function method, covering our criteria, one solution was obtained for the optimum conditions to produce pumpkin dessert with the highest desirability value obtained. The optimum temperature, holding time and calcium chloride concentration were 29.89°C, 56.22 minute, and 2.14 % calcium chloride concentration, respectively. By applying these optimal conditions, pumpkin dessert with a 28.31 N hardness, 45.75 L*, 9.2 a*, 28.44 b* and 112.77 µg/ml total carotenoid content could be put out. The product with these specifications could be acceptable from consumer perspective. The product with these specifications could be acceptable from consumer perspective. The estimated L* value and a* value (45.75 and 9.2 respectively) were close to 46.56 and 9.5 which is L* value and a* value of the control group of pumpkin dessert. Desirability was 0.526 for pumpkin dessert.

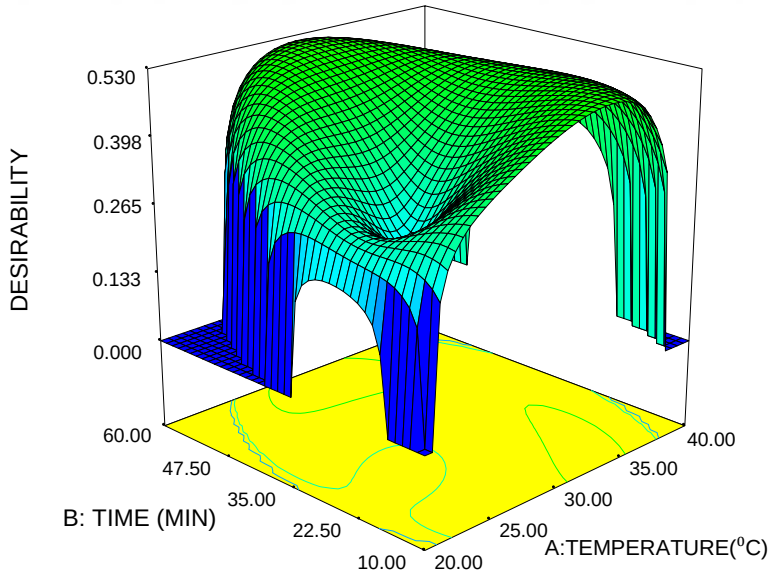

Figure 3.6 Effect of temperature and holding time on desirability function

CHAPTER 4
CONCLUSION

The consequences of this study can be summarized as :

1) The hardness of pumpkin dessert varied between 15 N and 29.67 N. High temperature caused to soft texture and increase calcium chloride concentration and holding time improve hardness of pumpkin dessert.

2) Total carotenoid content of pumpkin dessert decreased with rising temperature and holding time, and also increasing calcium chloride concentration affected negatively.

3) It was examined that high temperature and holding time lead to improving of L*, a* and b* value of pumpkin dessert. Increased calcium chloride concentration was adversely affects color values.

4) In sensory analysis, results represented that increasing calcium chloride pan out affirmative hardness on pumpkin desserts. While color of desserts varied with according to parameters, it was determined that bitterness level increased with increasing calcium chloride concentration for taste of pumpkin desserts.

5) The optimum temperature, holding time and calcium chloride concentration were 29.89°C, 56.22 minute, and 2.14 % calcium chloride concentration, respectively. By applying these optimal conditions, pumpkin dessert with a 28.31 N hardness, 45.75 L*, 9.2 a*, 28.44 b* and 112.77 μg/ml total carotenoid content could be produced.

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APPENDICES

APPENDIX A
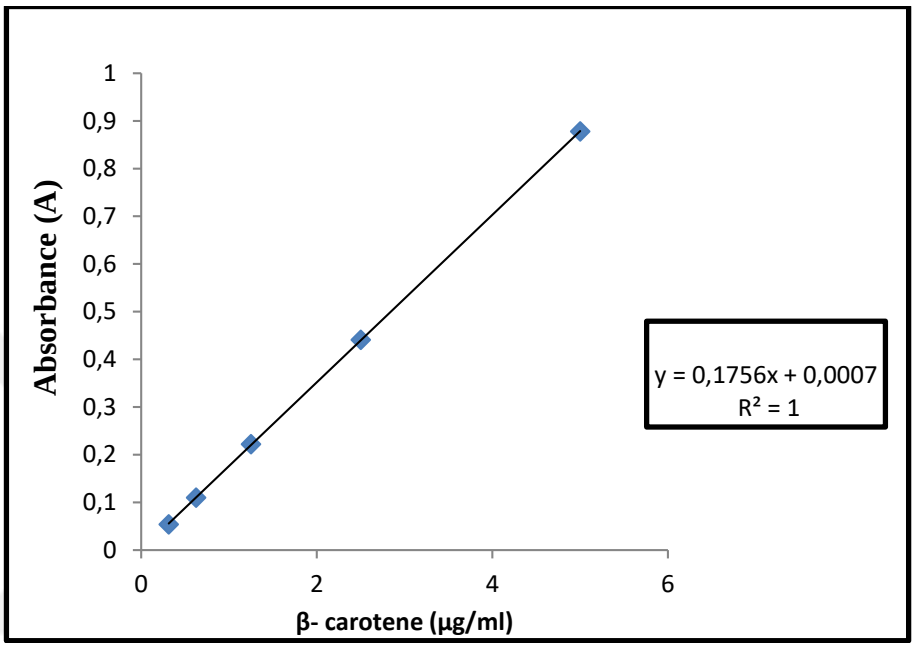
Figure A.1 Calibration curve of β- carotene