SHELF LIFE EXTENSION OF PERSIMMON FRUIT (*DIOSPYROS KAKI*) BY AN EDIBLE COATING, SEMPERFRESH™, COLD STORAGE AND MICROWAVE TREATMENT

by
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ABSTRACT

SHELF LIFE EXTENSION OF PERSIMMON FRUIT (*DIOSPYROS KAKI*) BY AN EDIBLE COATING, SEMPERFRESH™, COLD STORAGE AND MICROWAVE TREATMENT

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The persimmon fruit (*Diospyros kaki*), mainly common in China and Japan has an economic importance due to medicinal bioactive compounds and delicious nutritive properties. In this study, 20 kg of fruits were harvested from Aydın, Turkey and treated with Microwave (M), Semperfresh™ (S) and Cold Storage (C), as solely or in combination, in triplicates.

When all treatments were considered with respect to the control, it was found that the triple combination, Semperfresh™ - Cold Storage - Microwave (S+C+M) exhibited low microbial counts, in addition, the pH, respiration rate and

sensory panel showed that none of these quality criteria were changed, therefore FT-IR, sugar, firmness and the weight analysis of the fruits varied.

Keywords: Persimmon, *Diospyros kaki*, Semperfresh^{TM}, Cold Storage, Microwave.

ÖZET

TRABZON HURMASININ (*DIOSPYROS KAKI*) YENİLEBİLİR BİR KAPLAMA MADDESİ OLAN SEMPERFRESH™, SOĞUKTA SAKLAMA VE MİKRODALGA İLE MUAMELE EDİLEREK RAF ÖMRÜNÜN UZATILMASI

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Çoğunlukla Çin ve Japonya'da yaygın olan Trabzon hurması (*Diospyros kaki*), içerdiği iyileştirici biyoaktif bileşenleri ve besleyici özellikleri sebebiyle ekonomik bir öneme sahiptir. Bu çalışmada, Aydın'da yetiştirilmiş 20 kilogram Trabzon hurması; Mikrodalga (M), Semperfresh™ (S) ve Soğukta Saklama (C) ile birli, ikili ve üçlü birleşimler halinde işlem görmüştür.

Tüm işlemler kontrol esas alınarak dikkate alındığında, üçlü birleşim, Semperfresh™ - Soğukta Saklama - Mikrodalga (S+C+M) düşük mikrobiyal sayım sergilemiş olup, ek olarak pH, solunum hızı ve tat paneli sonuçları bu

kalite ölçütlerinin hiçbirinin değişmediğini göstermiş, bununla birlikte meyvelerin FT-IR, şeker, sertlik ve ağırlık analizleri ise değişmiştir.

Anahtar Kelimeler: Trabzon hurması, *Diospyros kaki*, Semperfresh^{TM}, Soğukta Saklama, Mikrodalga.

Dedicated to My Family

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LIST OF ABBREVIATIONS

°C: Degree Celsius
A: Astringent
CO_2: Carbon Dioxide
cm: Centimeter
DNS: Dinitrosalicylic Acid
FAO: Food and Agricultural Organization
G: Gram
Ha: Hectare
IU: International Units
Kg: Kilogram
Kgf: Kilogram-force
kGy: KiloGray
Kj: Kilojoule
Kcal: Kilo Calorie
Log: Logarithm
L: Liter
Mcg: Microgram
Mcg_DFE: Micrograms of Dietary Folate Equivalent
Mcg_RAE: Micrograms of Retinol Activity Equivalents
Mg: Milligram
Mg/L: Milligram per Liter
mL: Milliliter
mm: Millimeter
mm/sec: Millimeter per second
NA: Non-Astringent
nm: Nanometer
O.D.: Optical Density
PC: Pollination Constant
PCA: Pollination-Constant, Astringent
PCNA: Pollination-Constant, Non-Astringent
PV: Pollination Variant
PVA: Pollination-Variant, Astringent
PVNA: Pollination-Variant, Non-Astringent
RH: Relative Humidity

TMAB: Total Mesophilic Aerobic Bacteria
VDG: Volatile Dependent Group
VIG: Volatile Independent Group
W/v: Weight per Volume

1. INTRODUCTION

Persimmon is a subtropical fruit, widespread grown in the world. It belongs to *Ebenaceae* family and *Diospyros* genus. The origin of persimmon is China and Japan. Its color and apple-like shape is very popular in Far East and consumed high amounts. Due to its ascending popularity parallel with the production rate, the fruit was cultivated far from homeland. Different types of cultivars appear as different brand names in the market (Mallavadhani, 1998).

In general persimmon fruit has been classified by its astringency. Non-astringent type is more desirable for consumers by the reason of its sugary taste (Yonemori *et al*., 2000).

In Turkey the production rate of persimmon has been significantly increasing in the last years in the domestic market (Llacer and Badenes, 2001). Unfortunately, limited non-astringent types of persimmon, in addition the production and storage problems negatively affect capacity of external trade (II. Agriculture Congress, 2004)

Persimmon is an important fruit against cancer due to its antioxidant activity. It is strongly effective on the regulation of cholesterol (Gorinstein *et al.*, 1994). Moreover, persimmon arranges the function of intestines. Persimmons

are also reported to be good sources of sodium, potassium, magnesium, manganese and iron (Gorinstein *et al.*, 2001).

Because of its medical and biochemical importance, the shelf life of the persimmon was tried to be extended by different applications like the use of packaging plastic materials (Cia *et al*., 2006) exposing to the gaseous ozone (Salvador *et al*., 2006), carbon dioxide, nitrogen (Bibi *et al.*, 2006), and air treatment (Luo, 2005).

1.1. Status of Persimmon in the World and Turkey

Through with beautiful look and delicious taste, the persimmon is named the “food of the God” (from Greek, Dios meaning God and Spyros meaning food). In Turkey, the fruit has been known by different names in Turkish; as Trabzon hurması, cennet meyvesi, cennet elması (Sütyemez and Ergenoğlu, 2000). Persimmon is somewhat common in warm regions of the world. Persimmon have originated in China and cultivated in some centuries before Christ. The fruit was introduced to Japan in the 7th century and to Korea in the 14th century. European countries have met in the 17th century and later, in the 18th century, it was already known world-wide (Sugiura, 1997, in Llacer and Badenes, 2001). At present it has been mainly grown in China, Japan and Korea where more than 2000 local cultivars have developed (Yamada *et al*., 2002). It has also been cultivated in the Mediterranean coast of France, Italy, and other European countries, as well as Turkey, and in southern Russia (Bellini and Giordani, 2005).

Tables 1 and 2 show the area and production values for the countries that have more than 100 ha and/or produce more than 1000 t per year according to FAO (2006). In 1961, only five countries had production: China, Japan, Korea, Italy and Brazil. In 2000, two countries were added to producer countries list: Israel, beginning in 1980, and New Zealand, starting from 1990. Other countries, still according to FAO (2006), are close to those numbers: Australia, Iran and Mexico. Some countries like Spain, Portugal, Turkey and others do not appear in the persimmon fruits' statistics of FAO, although at present they largely surpass the production of 1000 tons, probably because persimmon is included in their statistics together with other minor fruit crops.

Table 1. Harvested area (ha) of persimmon fruit from the main producer countries in the last 45 years.

Time	World	China	Korea	Japan	Italy	Brazil
1961	122,381	76,230	2,346	36,700	4,600	2,500
1970	180,611	140,270	5,192	35,900	3,800	3,444
1980	154,432	110,684	6,590	29,400	3,300	4,051
1985	239,055	191,119	9,838	29,800	3,313	3,692
1990	215,107	162,993	13,581	29,500	3,055	3,960
1995	331,752	269,568	25,009	28,000	2,878	4,819
2000	545,738	471,463	31,193	25,000	2,227	6,230
2006	729,095	653,200	28,436	23,500	2,668	8,309

Table 2. Harvested amount (tons) area of persimmon fruit from the main production countries in the last 45 years.

Time	World	China	Korea	Japan	Brazil	Italy
1961	990,079	497,250	13,271	393,500	15,298	70,740
1970	911,635	457,341	30,310	342,700	21,659	59,600
1980	968,198	566,638	31,837	265,200	39,958	61,100
1985	1,186,165	690,945	97,031	289,700	43,658	56,200
1990	1,157,241	640,230	95,758	285,700	46,712	68,770
1995	1,562,487	985,803	194,585	254,100	51,685	61,300
2000	2,417,854	1,615,797	287,847	278,800	63,300	42,450
2006	2,972,349	1,987,000	352,822	232,700	164,849	52,863

Persimmon is widely grown in many areas of Turkey. Though the exact date of introduction to Turkey is not known, it is clear that dates back to rather old times (Onur, 1990, in Yıldız *et al*., 2007). As to the available records, persimmon was introduced to Turkey from Russia by the route of Black Sea. Agricultural Ministry of Turkey was the spearhead to the introduction of different varieties after 1967 and with the help of selection studies, the number of varieties reached to 14. After 1989 with the introduction of new varieties from Italy, Israel, Japan, France and Pakistan the known number of varieties attained 74 (Tuzcu and Şeker, 1997, in Yıldız *et al*., 2007).

In Turkey, persimmon has been cultivated in mostly Mediterranean region. Even though the trees are largely spread around the Mediterranean area, its culture was based on seedlings located in home gardens of small towns

near the larger cities. The main producer districts are located on the eastern part of the region (Aksoy, 1995). Especially Hatay, Adana, and Mersin are the main production centers in Mediterranean region, where harvested fruits are exported to the Arabian countries. In the coastal areas of the Mediterranean region, persimmon can be found at the market for about 7 weeks starting from the third week of September with the early varieties and enduring until mid November. The fruits of the late ripening varieties await on the tree even after the leaf fall (Onur, 2007).

Black Sea region is the second biggest production area of Persimmon fruit. The mountains lying parallel to the Black Sea line ensure a rather mild and humid climate in a narrow strip of land enabling the production of subtropical varieties (Aksoy, 1995). In this area, Ordu, Artvin, Samsun, and Kastamonu are main cultivation regions. Marmara region has the third biggest production capacity in Turkey. Notably Kocaeli is very important for the market (Onur, 2007). Inadequacy of variety of persimmon and amiss application of agricultural techniques increase the production cost. Over and above cold storage and transportation techniques must be improved (II. Agriculture Congress, 2004).

In Turkey, almost all of the commercially grown varieties are astringent type. The first study on Japanese persimmon was started in 1984 at the Citrus Research Institute in Antalya. Alata Horticultural Research Institute helped to carry out the research. Aim of the research select the cultigens present in the western Mediterranean Region resulted in 6 types that have high quality (Onur,

1985: Onur and Taşdemir, 1987, in Aksoy, 1995). 68 different species persimmon fruit are brought from Italy, Japan and Pakistan by Çukurova University. Thus South and Southeast region adaptation works and Blacksea region selective studies were accomplished (Gülcan *et al.,* 2000).

1.2. Plant and Fruit Morphology

Persimmon is a deciduous, monoecious plant, and set fruit parthenocarpically or with pollination. The fruit consists of a berry, as large as an apple, orange in color, with soft, juicy pulp, sweet if ripe (Prandini and Marchesi, 1999). Persimmons generally ripen from late August until early December, depending on climate and region. The fruit is eaten both fresh and dried (McEachern and Hancock, 1997). Also fruits are consumed as jam, marmalade, and molasses.

The appearance of persimmon can be seen in Figure 1, and also cross-section of persimmon can be seen in Figure 2.

Figure 1. The appearance of persimmon fruit.

Figure 2. Cross-section of persimmon fruit.

1.2.1. The classification of persimmon fruit

Persimmons are classified into different groups based on fruit characteristics. The first classification, consists of two groups of cultivars of oriental persimmons with different degrees of astringency at harvest; one is an astringent type (A) and the other is non-astringent (NA) (Yamada, 1994, in Harima *et al*., 2003).

The second classification relates to fruit flesh color when seeds are present. The flesh of pollination variant (PV) types becoming dark around the seed whereas the flesh color of pollination constant (PC) types in uninfluenced seed (Yonemori *et al*., 2000).

Astringency classification has popular trademarks; Fuyu, Gosho, and Jiro are trademarks of NA, Hachiya, Fuji and Saijo are the main trademarks of A cultivators. Astringency and the influence of seed on pulp color are combined to classify the persimmon fruits. There are four groups of cultivators: PCA, PCNA, PVA and PVNA. Fuyu is classified as pollination-constant, non-astringent (PCNA); Zenji Maru as pollination-variant, non-astringent (PVNA); Hachiya and Fuji as pollination-constant, astringent (PCA); and Hiratanensh and Tonewase are pollination-variant astringent (PVA) (Yamada and Kurihara, 1984, in Ray, 2002).

Another classification of persimmon is proposed according to the quality of tannin in the flesh and the capability of seeds of releasing volatile compounds during fruit development. Two main groups can be defined in this method

(Sugiura, 1983, in Giordani, 2001). Volatile independent group (VIG), of cultivars in which the process of natural loss of astringency is not the formation and accumulation of volatile compounds in flesh. PCNA cultivars are included this group.

Volatile dependent group (VDG), of cultivars in which the natural process of astringency loss is contingent on the formation and accumulation of volatile compounds in the fruit. PVNA, PVA and PCA cultivators are included this type. PVNA cultivars bring seeds with high capability of releasing volatile compounds, whereas PVA cultivars bring seeds with low capability of releasing volatile compounds, in addition PCA cultivars show very low or no release of volatile compounds (Sugiura, 1983; Sugiura and Tomana, 1983, in Giordani, 2001). Table 3 shows all classification types of the persimmon and criteria of its evaluation.

Table 3. Classification of persimmon fruit by astringency and flesh color (Yonemori *et al*., 2000).

	NA	**A**
PC	**PCNA** Non-astringent at maturity whether seeded or not. Flesh color unaffected by seed at maturity. Tannins are not coagulated by ethanol treatment at immature stages when fruit is still astringent.**(VIG)**	**PCA** Astringent at maturity unless treated. Flesh color unaffected by seed at maturity. Tannins are coagulated by ethanol treatment even at immature stages.**(VDG)**
PV	**PVNA** Non-astringent at maturity only if seeded. Flesh turns brown at maturity if seeded. Tannins are coagulated by ethanol treatment even at immature stages. **(VDG)**	**PVA** Astringent at maturity unless treated. Brown flesh color only around seed at maturity. Tannins are coagulated by ethanol treatment even at immature stages. **(VDG)**

1.2.2. Tannin structure

The astringency sensation of persimmon comes from the presence of soluble tannins that are grouped together in large cells named "tannin cells". When these cells are broken by a bite during eating, they release soluble

tannins that interact with protein on the surface of the tongue, producing the astringency sensation (Matsuo and Itoo, 1978, in Testoni, 2002).

Tannins are amorphous, rarely crystalline substances which are widely distributed in the plant kingdom. It is a group of phenolic substrates of polyphenol oxidase, containing gallic acid derivatives and glucose units are joined by glycosidic bonds (Özen *et al*., 2004).

Even if more than 0.5% soluble tannins exist in persimmon fruit, they cannot be eaten. The differences between sweet and astringent persimmon fruits are in the quality and size of tannin cells. Tannin cells of perfect sweet persimmon are smaller than those of imperfect sweet persimmons, perfect or imperfect astringent persimmon (Itoo, 1986, in Ray 2002).

The strong binding capacity of persimmon tannin with proteins and carbohydrates has led to several practical applications in Japan. Umbrellas used to be made from paper soaked in crude persimmon tannin. Persimmon tannin has also been widely used to remove protein in brewing of ‘sakè’ which is Japanese rice wine (Nunokawa and Saitoh, 1974, in Nakajima and Sakaguchi, 2000).

1.3. The Ripening and Storage of Persimmon

Harvest season of astringent persimmon fruit is related with the soft stage of fruit development. If picked during this time, the fruit is either soft or will become soft and the astringent tannins are coagulated, so that the fruit will be suitable for eating. Mostly, fruit can be picked and softened at room temperature about 7 to 10 days before it would be softening on the tree. The

time varies slightly with cultivars, and is about the same for both astringent and non-astringent types (Miller and Crocker, 1994).

Non-astringent persimmon does not require softening stage as it can be eaten firm. Non-astringent fruit are distinguished by color and can be rated for ripeness by a color chart. The elimination of green by the increasing development of yellow and/or orange to orange-red is a general indication of marketable fruit. Generally, since astringent persimmons must be soft, or near that state at harvest, they cannot be stored well; but, non-astringent types can be stored up to 30 days at room temperature (Miller and Crocker, 1994).

1.3.1 The removal of astringency from the fruit

Astringent fruit must be soft or artificially treated before astringency is removed so they are considered as 'suitable for eating' but non-astringent types lose astringency while still hard and can be eaten hard or soft (Miller and Crocker, 1994). Astringency can be removed by various treatments, including ethanol vapour treatment of fruit on the tree or after harvest (Taira, 1996, in Asgar *et al*., 2003).

In this manner, treatment with a high CO_2 concentration for one day has been used commercially for some leading astringent cultivars. The rate of the deastringency process is directly proportional to the amount of acetaldehyde accumulation (Pesis and Ben-Arie, 1984; Pesis *et al*., 1988, in Yamada *et al*., 2002).

Hot water treatment (40°C for 15 - 24 h) is another method of removing astringency; however, the products were not of good quality. Freezing treatment (25°C for 10 - 90 days) resulted in the decrease of soluble tannins but with incomplete removal of astringency. Treatment with carbon dioxide gas in a polyethylene bag at normal pressure was also been used before or after harvesting with good results. Irradiation with 1.5–2.5 kGy also resulted in partial removal of astringency. On tree removal of astringency by enclosing the fruit in polyethylene bags containing a small amount of ethanol and post harvest ethanol treatment rendered the fruit very pleasant to eat (Chaudry, 2002, in Bibi *et al*. 2006).

1.4. The Biochemical Structure and the Nutritional Facts of Persimmon Fruit

Mallavadhani (1998) reported that persimmon contains many medicinally bioactive compounds, such as carotenoids, tannins, flavonoids, terpenoids, steroids, naphthoquinones, sugars, amino acids, minerals, and lipids (in Kawase *et al*., 2003).

Table 4 shows the nutritional value of raw persimmon fruit according to USDA nutrient database (USDA, 2007).

Table 4. Nutritional value of 100 g persimmon fruit (USDA, 2007).

Nutrient	Units	Value per 100 grams
Proximates		
Water	G	80.32
Energy	Kcal	70
Energy	Kj	293
Protein	G	0.58
Total lipid (fat)	G	0.19
Ash	G	0.33
Carbohydrate, by difference	G	18.59
Fiber, total dietary	G	3.6
Sugars, total	G	12.53
Sucrose	G	1.54
Glucose (dextrose)	G	5.44
Fructose	G	5.56
Minerals		
Calcium, Ca	Mg	8
Iron, Fe	Mg	0.15
Magnesium, Mg	Mg	9
Phosphorus, P	Mg	17
Potassium, K	Mg	161
Sodium, Na	Mg	1
Zinc, Zn	Mg	0.11
Copper, Cu	Mg	0.113
Manganese, Mn	Mg	0.355
Selenium, Se	Mcg	0.6
Vitamins		
Vitamin C, total ascorbic acid	Mg	7.5
Thiamin	Mg	0.030
Riboflavin	Mg	0.020
Niacin	Mg	0.100
Vitamin B-6	Mg	0.100
Folate, total	Mcg	8
Folate, food	Mcg	8
Folate, DFE	mcg_DFE	8
Vitamin A, IU	IU	1627
Vitamin A, RAE	mcg_RAE	81
Vitamin E (alpha-tocopherol)	Mg	0.73
Vitamin K (phylloquinone)	Mcg	2.6

1.5. The Biomedical Importance of Persimmon Fruit

Tannin-rich fruits have been traditionally used for the treatment of hypertensive diseases (Kameda *et al*., 1987, in Sakanaka *et al*., 2005) and evidence showed that persimmon tannins prolong life, and reduce the incidence of stroke in hypertensive rats (Uchida *et al*. 1989, in Gorinstein *et al*., 1998).

Gorinstein (1994) claimed that persimmon fruit is also rich in antioxidant phenolic compounds other than tannins, and it has been demonstrated that these compounds may reduce the risk of chronic diseases by protecting tissues against free radical-mediated damage. This effect was attributed to the fact that persimmon tannins are 20 times more potent than vitamin E- a classical antioxidant (Gorinstein *et al*., 1998).

Persimmon fruit affects the function of homeostasis and also affects the acceleration of secretion of intestinal juice and intestinal contraction (Yu, 1976, in Kim *et al*., 2006).

1.6. The Studies on Extending Shelf Life of Persimmon

IFST (1993) reported that shelf life is defined as the period of time under defined conditions of storage for which a food product remains safe and fit for use. In other words, during this period, it should retain its desired sensory, chemical, physical, functional or microbiological characteristics (Guizani *et al*., 2005). Various methods have been in use for extending the shelf life of foods.

Up to date, several studies had been conducted on persimmon fruit on extending its shelf-life, including use of chemicals like ozone (Salvador *et al*., 2006), 1-methylcyclopropene (1-MCP) (Ortiz *et al*., 2005), 1-methylcyclopropene and ethylene (Pang *et al*., 2007) and physical treatments like dry air heat (Woolf *et al*., 1997a), dry air (Woolf *et al*, 1997b), and hot air application (Luo, 2005), and physical and chemical combinations like hot water and controlled atmosphere (Burmeister *et al*., 1997). Using of packaging plastic materials (Cia *et al*., 2006), exposition to the gaseous ozone (Salvador *et al*., 2006), carbon dioxide and nitrogen (Bibi *et al*., 2006) are other possible methods for the extension studies of the shelf life of the persimmon fruit.

1.6.1. Microwave

Microwave energy has been widely used in food processing, not only for its heating properties without damaging the nutritional characteristics but also its reduced processing time and costs, enhancing product uniformity and yields, improving unique microstructure and protecting food from surface browning and crusting (Anastasis, Katerina, and Michael, 1999, in Huang *et al*., 2005).

Ideas for the application of microwave heating to the processing of food are reviewed. Pasteurization, sterilization, defrosting, cooking, dehydration and other applications are reported (Sale, 1976).

Unlike other energy delivery systems, microwave heating involves a rapid and direct heating process that reduces the time required to achieve a desired temperature. Hence, the total cumulative thermal treatment is much reduced, better preserving the thermolabile constituents of foods, such as aromas,

vitamins, and pigments (Mudgfett, 1986, in Clare *et al*., 2005). In-package microwave heating treatment of fresh soybean curds effectively extended shelf-life (Wu and Salunkhe, 1977).

1.6.2. Cold storage

Cold storage is based on the idea that refrigerated temperatures slow down most microbial growth and are effective to reduce enzyme activity (Wiley, 1994). Cold storage positively effects the antioxidant properties (Pérez-Ilzarbe *et al*.,1997) and provide long storage life (Lara and Wendell, 1998) to apples, increase shelf life and quality of cherries (Yaman and Bayındırlı, 2002).

1.6.3. Semperfresh ™

Semperfresh™ is used mainly for composite coatings comprised of the sodium salt of carboxymethyl cellulose as the film former, with sucrose fatty acid ester as the emulsifiers and mixed mono and diglycerides (Baldwin *et al*., 1995, in Farber *et al*. 2003) (Yaman and Bayındırlı, 2002). Semperfresh™ delays ripening by altering the permeability of the coated fruit so that internal oxygen levels decrease and carbon dioxide levels increase (Smith and Stow, 1984, in Bauchot *et al*., 1995). It has been commercially available for coating fruits and vegetables since the 1980s.

Research on Semperfresh™ treatment showed that the coating reduced apple ripening rate, as observed by several parameters including texture and color (Santerre *et al*., 1989). Semperfresh™ treatment was also applied on

cherries and conclusions showed that treatment reduce the weight loss and increase firmness, ascorbic acid content, titratable acidity and protect the skin color of cherries during storage time, and increase the shelf-life of cherries (Yaman and Bayındırlı, 2002).

Semperfresh™ treatment is significantly effective on mandarin to reduce the change of ascorbic acid, soluble solids, titratable acidity, weight loss and respiration rate (Bayındırlı *et al*., 1995). Semperfresh™ treatment also affected the shelf life and quality of limes (Motlagh and Quantick, 1998), oranges (Baldwin *et al*., 1995), mango (Ketsa and Prabhasavat, 1992), banana (Truter and Combrink, 1990), apple and pear (Köksal *et al*., 1994).

2. THE AIM AND SCOPE OF THE STUDY

In the present study, our aim is to compare a sucrose-based coating material (Semperfresh^{TM}), together with microwave and cold storage applications on persimmon fruit (*Diospyros kaki*) for extending its shelf life. In addition, when Semperfresh^{TM} application is considered, a recommended dose for persimmon fruit has also been specified.

3. MATERIALS AND METHOD

3.1. Fruit

Twenty kg of mature persimmon fruits were harvested from Aydın, Turkey, and were transported to the laboratory in 6 hours. All samples were washed and dried; the defective ones were eliminated before the treatments.

Fruits were divided in eight lots of treatment groups: Semperfresh™ (S), microwave (M), cold storage (C), microwave - Semperfresh™ (M+S), Semperfresh™ - cold storage (S+C), microwave - cold storage (M+C), Semperfresh™ - cold storage – microwave (S+C+M), and control (CO).

3.2. The Applications of Cold Storage, Microwave and Semperfresh™

3.2.1 The Cold storage treatment

The cold storage process was carried out using a modified storage chamber (Nuve, type ES 500) at -18 ^{0}C for 60 minutes. The required relative humidity (RH) was provided by pans filled with water around different parts of the chamber (RH ≅ 90-95%), which had a ventilation system.

3.2.2 The microwave processing

The microwave (M) applications were performed by exposing all samples at 150 W power for two minutes in a microwave oven (Arçelik, MD 50).

3.2.3 Semperfresh™ fruit coating

Semperfresh™ edible fruit coating, composed of sucrose esters of fatty acids, sodium carboxymethyl cellulose and mono-diglycerides of fatty acids was obtained from ACC (Ankara) in liquid concentrate form (50.0 % w/v). The diluted solutions were mixed for 30–45 minutes with occasional stirring and then applied on the persimmon fruit (Özden and Bayındırlı, 2002). As no recommended concentration of Semperfresh™ was defined for persimmon by the vendor, 0.05, 0.1, 0.5, 1 and 2 % (w/v) levels were subjected to trials with relevance to microbiological and chemical quality criteria. A previous taste panel showed that 0.1% (w/v) can be preferred as a recommended concentration, after dilution with water. For the samples treated with Semperfresh™ along one minute, one mL Semperfresh™ (S) containing solution was used by completing to 1000 mL with distilled water.

3.3. The Microbiological Assays

Fruit (100g) was blended with 100 ml of sterile 0.1% peptone solution. One gram sample from this mixture was thoroughly mixed with 9 ml of sterile distilled water. The total mesophilic aerobic bacteria (TMAB) counts were carried out on plate count agar (PCA-OXOID) at 37 °C for 24 hours and mold and yeast counts were carried out on potato dextrose agar (PDA-DIFCO) plates

at 30 °C for 24 hours, as in triplicates. The assays were repeated in the first and 11th days.

3.4. The pH

The pH of the persimmon fruit samples were measured by a portable pH meter (Isolab), in squeezed persimmon juice.

3.5. The Sugar Content

The sugar content of the persimmon fruit was determined by dinitrosalicylic acid (DNS) method. One mL aliquot from each treatment group was mixed with three mL DNS and one mL of distilled water in triplicates. These groups were kept in water bath at 50° C in 30 minutes incubation (Nüve NB20, Turkey). The absorbance was determined by a spectrophotometer (CADAS 50S Dr. Bruno Lange GmbH-Berlin) at O.D. 550 nm.

A linear standard calibration curve (Figure 9) was constructed according to the following glucose concentrations: 50, 100, 150, 200 mg/L and these were plotted against the optical density. Finally, the amount of glucose in each sample was determined.

3.6. The Weight Determination

Three fruits, from each group were weighed during storage with a digital balance (Precisa) to determine weight loss.

3.7. The Firmness

Three fruits from each group were randomly selected for the firmness test, for which a fruit hardness tester (model FT011, Everwell, Japan) was used. The values were obtained at the same six points on the circumference of each fruit and results were stated in kgf.

3.8. The Respiration Rate

The respiration rates of persimmon fruits were measured with an oxygen analyzer (YSI). Two persimmon fruits, from each group were randomly selected per replication and enclosed in the sealed jars for three days. Oxygen concentration of each jar was determined with oxygen analyzer after each three days up to 11 days and respiration rate was expressed in mL of O_2 per kg hour.

3.9. Fourier Transform Infrared Spectroscopy (FT-IR)

All the treatment groups were analyzed by FT-IR (Jasco, Model, FT-IR 430) to determine their possible functional groups and their shifts. A pellet with potassium bromide (Merck) was prepared under constant pressure application of 10 tons with a 13 mm die before analytical preparation in FT-IR. The FT-IR analysis was performed according to the following criteria: accumulation: 16, resolution: 4 cm^{-1}, apodization: Cosine, gain: 128, scanning speed: 2mm/sec.

3.10. Taste Panel

A group of twenty panelists rated the persimmon fruits according to the criteria of taste, color, texture, appearance, flavor, bitterness, saltiness,

sourness, sweetness, pungency and overall evaluation with a 10 point scale per replication. A score of 10 represented most liked, and one corresponded to least liked (Table 7).

3.11. Statistical Analysis

The statistical analysis was performed by a multivariate one-way Anova software program.

4. RESULTS AND DISCUSSION

4.1. The Microbial Assay

When the three different treatments (Semperfresh^{TM}, microwave and cold storage) were considered, the microbial quality of the persimmon fruit was highly preserved, as an approximate of one log cycle reduction was observed for the triple combination of Semperfresh^{TM} - microwave - cold storage, which provided the most effective treatment for the total mesophilic aerobic bacteria as shown in Figure 3.

The sensory evaluation and statistical findings supported them in consistency ($p<0.05$).

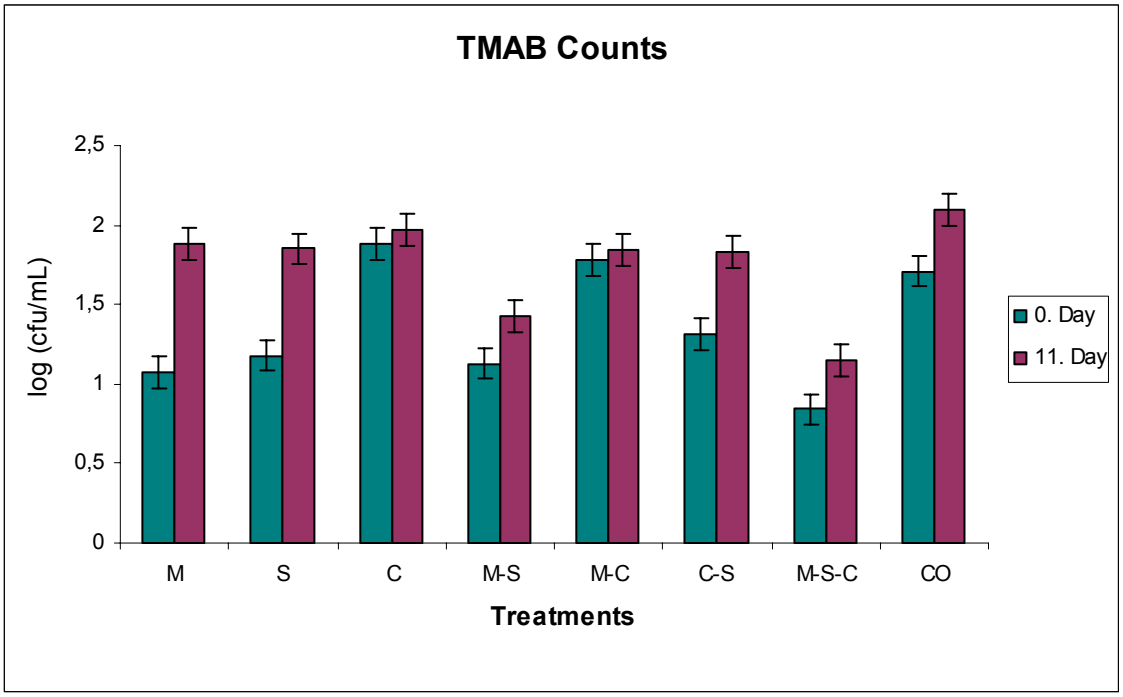

Figure 3. The effect of sole and combined treatment of Semperfresh^{TM}, cold storage and microwave on the total mesophilic aerobic bacteria (TMAB) levels of persimmon, with respect to control. The bars above the treatments indicated the standard deviations.

When the yeast counts were evaluated, it was observed that approximately 0.9 log cycle reduction was achieved in the triple application of Semperfresh^{TM} - microwave - cold storage (Figure 4). The sensory evaluation and statistical findings supported them in consistency ($p<0.05$).

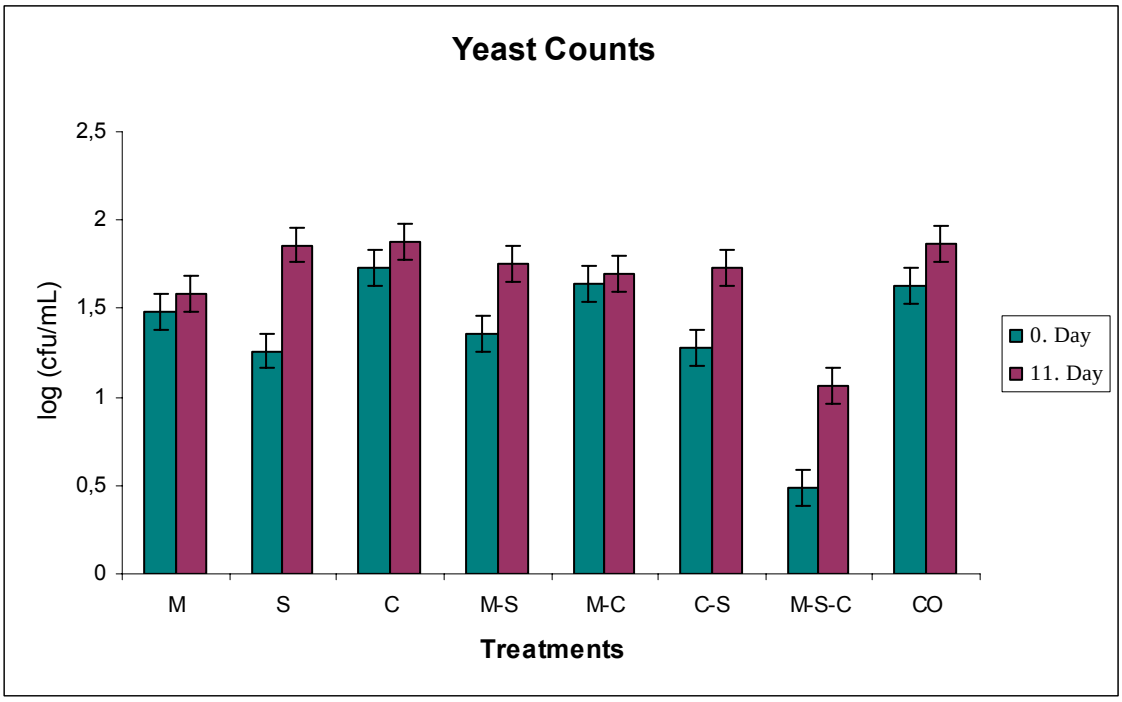

Figure 4. The effect of sole and combined treatment of Semperfresh^{TM}, cold storage and microwave on the yeast levels of persimmon fruit, with respect to control. The bars above the treatments indicated the standard deviations.

Moreover, the mold counts indicated a 1-1.1 log cycle reduction, following the triple treatment of Semperfresh^{TM} - microwave - cold storage (Figure 5). The total mesophilic aerobic bacteria and mold-yeast counts were taken into account while evaluating the microbial quality, with reference to the control group and the other treatments at the end of 11 days. The sensory evaluation and statistical findings supported them in consistency ($p<0.05$).

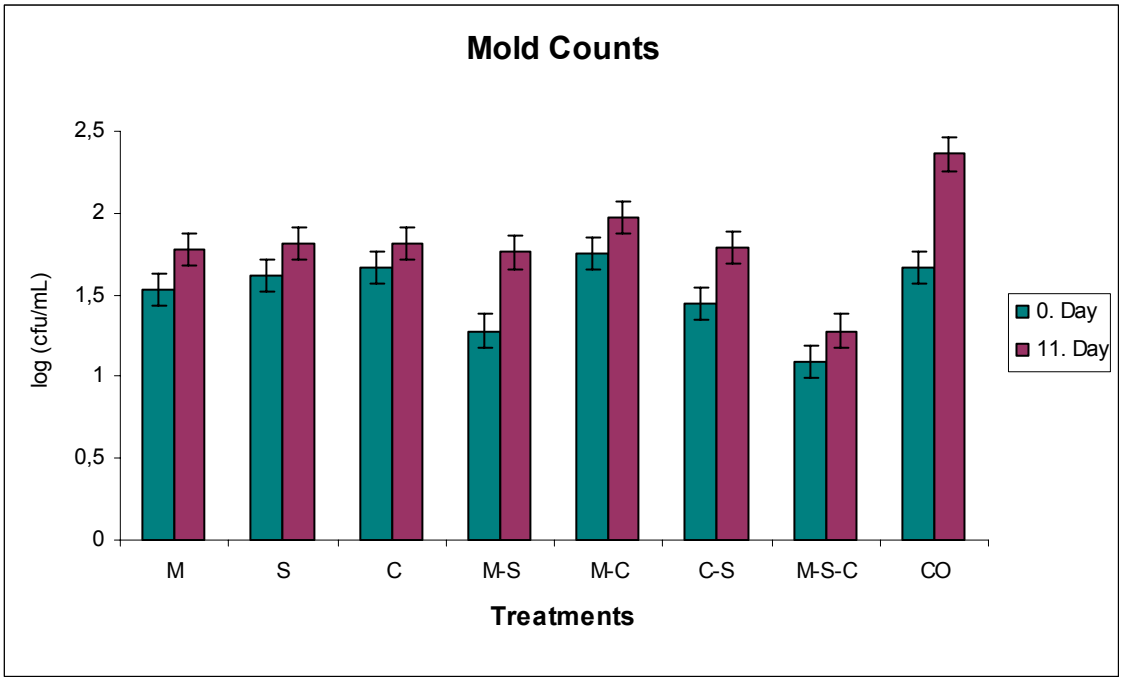

Figure 5. The effect of sole and combined treatment of Semperfresh^{TM}, cold storage and microwave on the mold levels of persimmon fruit, with respect to control. The bars above the treatments indicated the standard deviations.

4.2. The pH

The pH values of the persimmon fruit indicated that during the whole experimental procedure, a slight decline in pH was observed at the end of 11th days, while in control, sole applications of Semperfresh^{TM}, Semperfresh^{TM} - microwave, Semperfresh^{TM} - cold storage and for the combination, a slight increase in pH was observed when compared with the initial up to 11th days measurement, supported by the studies of Worrell *et al* (2002) on breadfruit, and Yurdugul (2005) on quinces. This is mainly due to creation of an anaerobic environment provided by the edible coating material which increases the

proliferation of the anaerobic yeast and mold (Figure 6). The sensory evaluation and statistical findings supported them in consistency ($p<0,05$).

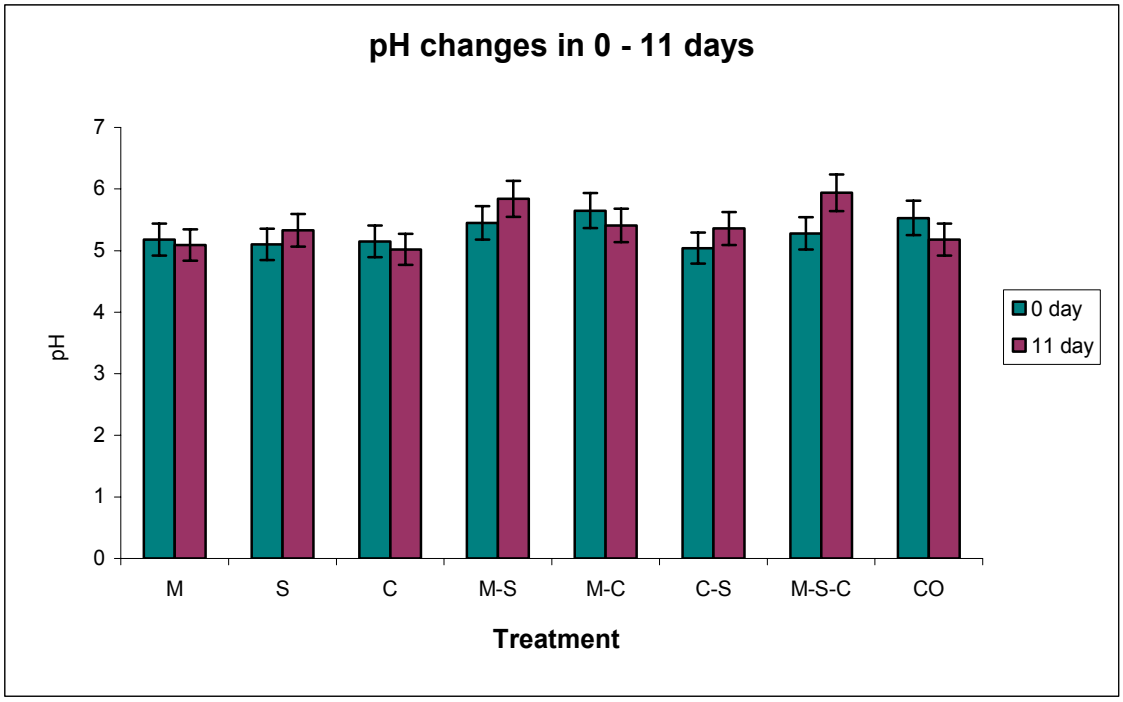

Figure 6. The effect of sole and combined treatment of Semperfresh^{TM}, cold storage and microwave on the pH levels of the persimmon fruit, with respect to control. The bars above the treatments indicated the standard deviations.

4.3. The Determination of Sugar Content

The sugar content amounts of persimmons, as given in milligrams per 100 g, implied that an increase was observed in sugar concentration of the all persimmons treatment groups at end of the 11th days, including the control. The percentages of changing of sugar content by samples were calculated (Table 5).

Table 5. 0^{th} and 11^{th} days' amount of glucose changing in all groups.

Group	0^{th} Day O. D. $_{550}$	Glucose Content	11^{th} Day O.D $_{550}$	Glucose Content	Changing %
C	0,064	71,41	0. 079	76,58	7,23
S	0,024	57,62	0,035	61,41	6,57
M	0,061	70,37	0,085	78,65	11,76
C – S	0,035	61,41	0,045	64,86	5,61
C – M	0,052	67,27	0,066	72,10	7,18
M – S	0,042	63,82	0,055	68,31	7,03
C – S – M	0,022	56,93	0,030	59,68	4,83
Control	0,035	61,41	0,052	67,27	9,54

This increase is mainly due to the anaerobic barrier created in the fruit by the effect of Semperfresh^{TM}, which do not allow the gas exchange between the fruit and the external environment. Triple combination showed the least glucose content changing conclusion otherwise microwave and control groups were the most affected ones.

4.4. The Weight Determination of Treatments

When the weight loss of the persimmon fruits were in consideration, a deviation was observed in the control group and cold storage when compared with the other groups (Figure 7). The highest losses were obtained in those two whereas the best weight retention was observed during the triple combination effect of Semperfresh^{TM} - microwave - cold storage, and the Semperfresh^{TM} - microwave application, mostly due to the protection of water level as the barrier action of the coating material, supported with the research of Bai *et al*. (2003) on

apples and Yurdugul (2005) on quinces where any type of coating material had eliminated the weight loss. The statistical findings supported this in consistency ($p<0.05$).

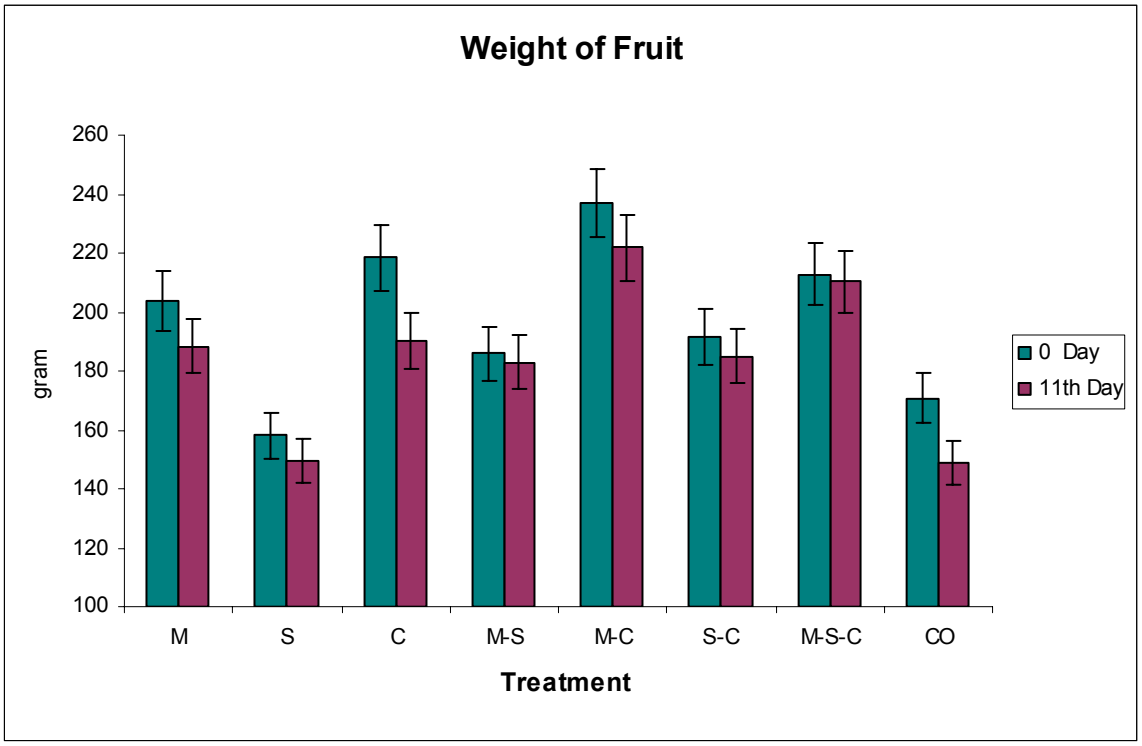

Figure 7. The effect of sole and combined treatment of Semperfresh^{TM}, cold storage and microwave on the weight of persimmon fruit, with respect to control. The bars above the treatments indicated the standard deviations.

4.5. The Respiration Rate

The triple combination and Semperfresh^{TM} - microwave showed sufficient oxygen consumption rate when compared with the other groups, whereas the cold storage and the control application indicated a high rate, respectively (Figure 8). The protective action mechanism of Semperfresh^{TM} as a barrier eliminates the oxygen consumption and controls respiration rate, in line with the

findings of Xu *et al*. (2001). The microwave application showed a slight decrease in oxygen consumption when compared with the Semperfresh^{TM}, whereas the cold storage application decreased the oxygen rate when used together, indicating a slight penetration effect on the coating material when compared with other Semperfresh^{TM} applications. The statistical findings supported this in consistency ($p<0.05$).

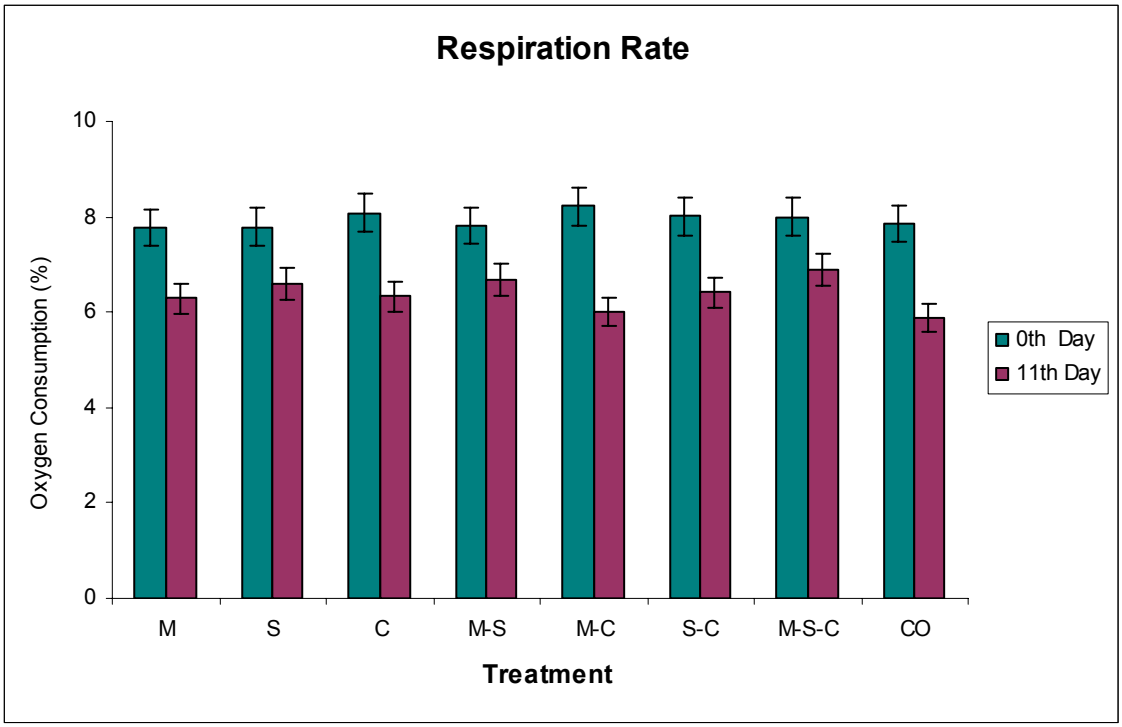

Figure 8. The effect of sole and combined treatment of Semperfresh^{TM}, cold storage and microwave on the respiration rate of persimmon fruit, with respect to control. The bars above the treatments indicated the standard deviations.

4.6. The Firmness

During the entire experimental procedure, a decline from 2.0 kgf to 1.2 kgf in the firmness values of the external coat of the persimmons was observed at the end of day 11 in triple combination treatment, in the control where the

highest decrease was observed up to 0.4 kgf at the end of the 11th days. This finding was supported by Luo (2007) where a similar effect was seen on the persimmon fruits which were treated by 1-methylcyclopropene.

The least affected group was Semperfresh™, where cold storage - Semperfresh™, and triple combination follows respectively, as mainly related to a relatively dry environment which provides a mechanical strength on the external tissue of persimmons when compared with other trials. The findings have demonstrated that sole Semperfresh™ application exhibited a qualified firmness rate, as similar in the research of Schreiner *et al*. (2003) on pepino-dulce fruit, Bai *et al*. (2003) on apples and Yurdugul (2005) where firmness is maintained through shelf life due to sole and combined treatments of cold storage and Semperfresh™, and high firmness rate is achieved with 'zein' coating, respectively.

Table 6. The change in firmness in all groups between 0th and 11th days.

Treatment	0th day	11th day	Degradation %
C	1.8 kgf	0.8 kgf	65,6
M	2.4 kgf	0.6 kgf	75,0
S	1.7 kgf	1.0 kgf	41,1
S – C	2.0 kgf	1.0 kgf	50,0
C – M	2.3 kgf	0.6 kgf	73,9
M – S	2.5 kgf	1.0 kgf	60,0
C – S – M	2.0 kgf	1.2 kgf	40,0
Control	2.2 kgf	0.4 kgf	81,8

The sensory evaluation (Table 7) and statistical findings supported them in consistency ($p<0.05$).

4.7. Fourier Transform Infrared Spectroscopy

Although some shifts were recorded due to FT-IR analysis with respect to control (Figure 10, 11 and 12) no such changes were observed as since the most important groups were preliminarily characterized as alkenyl, alcohol and the aromatics in the treatments (URL 1).

4.8. Statistical Analysis

When all treatments were evaluated, no significant differences were observed between the control and the others. The highest scores were mainly given by the control group (Table 7).

Table 7. Mean scores of sensory evaluation of persimmons ($p<0.05$ for all application).

	M	**S**	**C**	**M-S**	**M-C**	**S-C**	**M-S-C**	**Control**
Color	6,15	7,95	6,7	6,1	5,8	6,85	6	7,45
Sweetness	7,85	7,35	6,45	7,05	6,2	7,5	6,4	8,05
Sourness	1,45	1,55	2,35	2,05	2,05	1,75	2,45	1,4
Bitterness	1,2	1,4	1,55	1,8	1,7	1,5	2,25	1,3
Saltiness	1,2	1,45	1,55	1,35	1,7	1,55	2,01	1,6
Pungency	4,25	5,25	4,1	4,75	4,85	4,4	5,05	3,9
Appearance	6,7	8,05	6,6	6,35	6,25	6,8	5,7	7
Flavor	7,45	7,15	6,4	6	6,3	7	6,3	6,75
Texture	7,1	7,3	6,5	6,45	6,35	7,1	5,95	6,15
Overall	7,45	7,4	6,85	6,55	6,4	7,4	6,2	7,14

All groups were compared with the one-way Anova and interpreted by Dunnet t-test. According to the Dunnet t-test results, triple combination of Semperfresh^{TM} - microwave - cold storage indicated better values of the bitterness and the sourness with respect to control. The others represented no significant differences. All one-way Anova and Dunnet t-tests can be seen in Table 8.

5. CONCLUSIONS

By treatments of Semperfresh^{TM}, microwave, cold storage, the shelf life period of persimmon fruit was extended and fresh look was attained, mainly in Semperfresh^{TM} applications.

Besides the fruit's firmness and deposition of sugar content was improved and protected, as well as an improvement was observed in the taste panel with respect to control. The combinational treatments were found to be more effective than single treatments.

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URL 1: http://orgchem.colorado.edu/hndbksupport/specttutor/irchart.html

APPENDIX

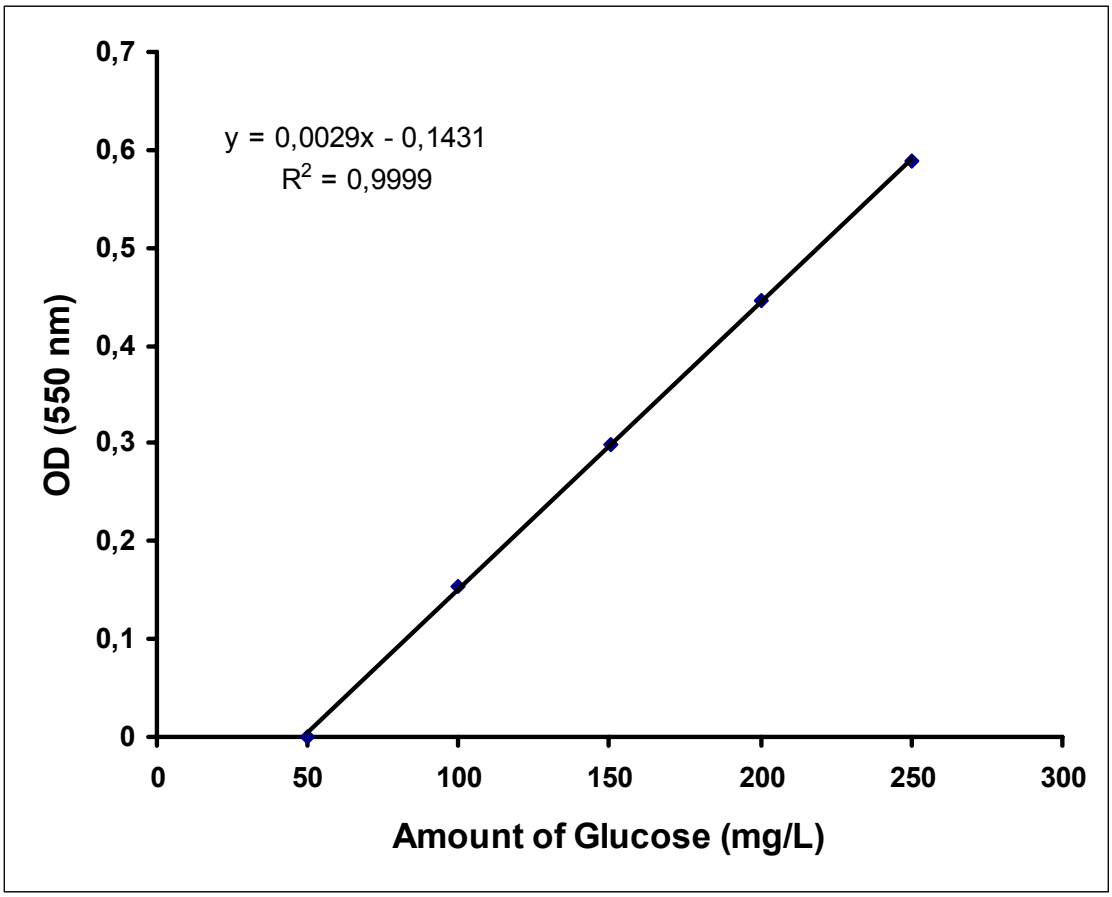

Figure 9. Calibration curve for glucose.

Table 8. Comparisons of all treatment groups in Dunnet t-test.

ANOVA

Color

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	81,300	7	11,614	4,142	,000
Within Groups	426,200	152	2,804		
Total	507,500	159			

Multiple Comparisons

Dependent Variable: Color

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	-1,300	,530	1,000	-2,55
S	CONTROL	,500	,530	,503	-,75
C	CONTROL	-,750	,530	,998	-2,00
M-S	CONTROL	-1,350	,530	1,000	-2,60
M-C	CONTROL	-1,650	,530	1,000	-2,90
S-C	CONTROL	-,600	,530	,994	-1,85
M-S-C	CONTROL	-1,450	,530	1,000	-2,70

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Sweetness

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	68,244	7	9,749	3,771	,001
Within Groups	392,950	152	2,585		
Total	461,194	159			

Multiple Comparisons

Dependent Variable: Sweetness

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	-,200	,508	,949	-1,40
S	CONTROL	-,700	,508	,998	-1,90
C	CONTROL	-1,600	,508	1,000	-2,80
M-S	CONTROL	-1,000	,508	1,000	-2,20
M-C	CONTROL	-1,850	,508	1,000	-3,05
S-C	CONTROL	-,550	,508	,993	-1,75
M-S-C	CONTROL	-1,650	,508	1,000	-2,85

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Sourness

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	22,882	7	3,269	2,287	,030
Within Groups	215,847	151	1,429		
Total	238,730	158			

Multiple Comparisons

Dependent Variable: Sourness

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	,050	,378	,839	-,84
S	CONTROL	,150	,378	,749	-,74
C	CONTROL	,950*	,378	,035	,06
M-S	CONTROL	,653	,383	,187	-,25
M-C	CONTROL	,650	,378	,182	-,24
S-C	CONTROL	,350	,378	,513	-,54
M-S-C	CONTROL	1,050*	,378	,018	,16

*. The mean difference is significant at the .05 level.

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Bitterness

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	15,475	7	2,211	2,009	,057
Within Groups	167,300	152	1,101		
Total	182,775	159			

Multiple Comparisons

Dependent Variable: Bitterness

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	-,100	,332	,936	-,88
S	CONTROL	,100	,332	,783	-,68
C	CONTROL	,250	,332	,593	-,53
M-S	CONTROL	,500	,332	,255	-,28
M-C	CONTROL	,400	,332	,380	-,38
S-C	CONTROL	,200	,332	,662	-,58
M-S-C	CONTROL	,950*	,332	,014	,17

*. The mean difference is significant at the .05 level.

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Saltiness

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	9,975	7	1,425	2,728	,011
Within Groups	79,400	152	,522		
Total	89,375	159			

Multiple Comparisons

Dependent Variable: Saltiness

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	-,400	,229	,999	-,94
S	CONTROL	-,150	,229	,975	-,69
C	CONTROL	-,050	,229	,922	-,59
M-S	CONTROL	-,250	,229	,994	-,79
M-C	CONTROL	,100	,229	,732	-,44
S-C	CONTROL	-,050	,229	,922	-,59
M-S-C	CONTROL	,500	,229	,074	-,04

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Pungency

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	32,094	7	4,585	,853	,545
Within Groups	817,150	152	5,376		
Total	849,244	159			

Multiple Comparisons

Dependent Variable: Pungency

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	,350	,733	,716	-1,38
S	CONTROL	1,350	,733	,147	-,38
C	CONTROL	,200	,733	,793	-1,53
M-S	CONTROL	,850	,733	,402	-,88
M-C	CONTROL	,950	,733	,341	-,78
S-C	CONTROL	,500	,733	,626	-1,23
M-S-C	CONTROL	1,150	,733	,232	-,58

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Appearance

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	65,094	7	9,299	3,186	,004
Within Groups	443,650	152	2,919		
Total	508,744	159			

Multiple Comparisons

Dependent Variable: Appearance

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	-,300	,540	,966	-1,58
S	CONTROL	1,050	,540	,121	-,23
C	CONTROL	-,400	,540	,980	-1,68
M-S	CONTROL	-,650	,540	,996	-1,93
M-C	CONTROL	-,750	,540	,998	-2,03
S-C	CONTROL	-,200	,540	,946	-1,48
M-S-C	CONTROL	-1,300	,540	1,000	-2,58

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Flavor

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	34,994	7	4,999	1,936	,068
Within Groups	392,450	152	2,582		
Total	427,444	159			

Multiple Comparisons

Dependent Variable: Flavor

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	,700	,508	,306	-,50
S	CONTROL	,400	,508	,578	-,80
C	CONTROL	-,350	,508	,977	-1,55
M-S	CONTROL	-,750	,508	,998	-1,95
M-C	CONTROL	-,450	,508	,987	-1,65
S-C	CONTROL	,250	,508	,710	-,95
M-S-C	CONTROL	-,450	,508	,987	-1,65

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Texture

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	34,175	7	4,882	1,996	,059
Within Groups	371,800	152	2,446		
Total	405,975	159			

Multiple Comparisons

Dependent Variable: Texture

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval Lower Bound
M	CONTROL	,950	,495	,127	-,22
S	CONTROL	1,150	,495	,054	-,02
C	CONTROL	,350	,495	,615	-,82
M-S	CONTROL	,300	,495	,660	-,87
M-C	CONTROL	,200	,495	,745	-,97
S-C	CONTROL	,950	,495	,127	-,22
M-S-C	CONTROL	-,200	,495	,950	-1,37

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

ANOVA

Overall

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	37,844	7	5,406	2,425	,022
Within Groups	338,850	152	2,229		
Total	376,694	159			

Multiple Comparisons

Dependent Variable: Overall

Dunnett t (>control)[a]

(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval
					Lower Bound
M	CONTROL	,050	,472	,846	-1,07
S	CONTROL	,000	,472	,875	-1,12
C	CONTROL	-,550	,472	,995	-1,67
M-S	CONTROL	-,850	,472	1,000	-1,97
M-C	CONTROL	-1,000	,472	1,000	-2,12
S-C	CONTROL	,000	,472	,875	-1,12
M-S-C	CONTROL	-1,200	,472	1,000	-2,32

a. Dunnett t-tests treat one group as a control, and compare all other groups against it.

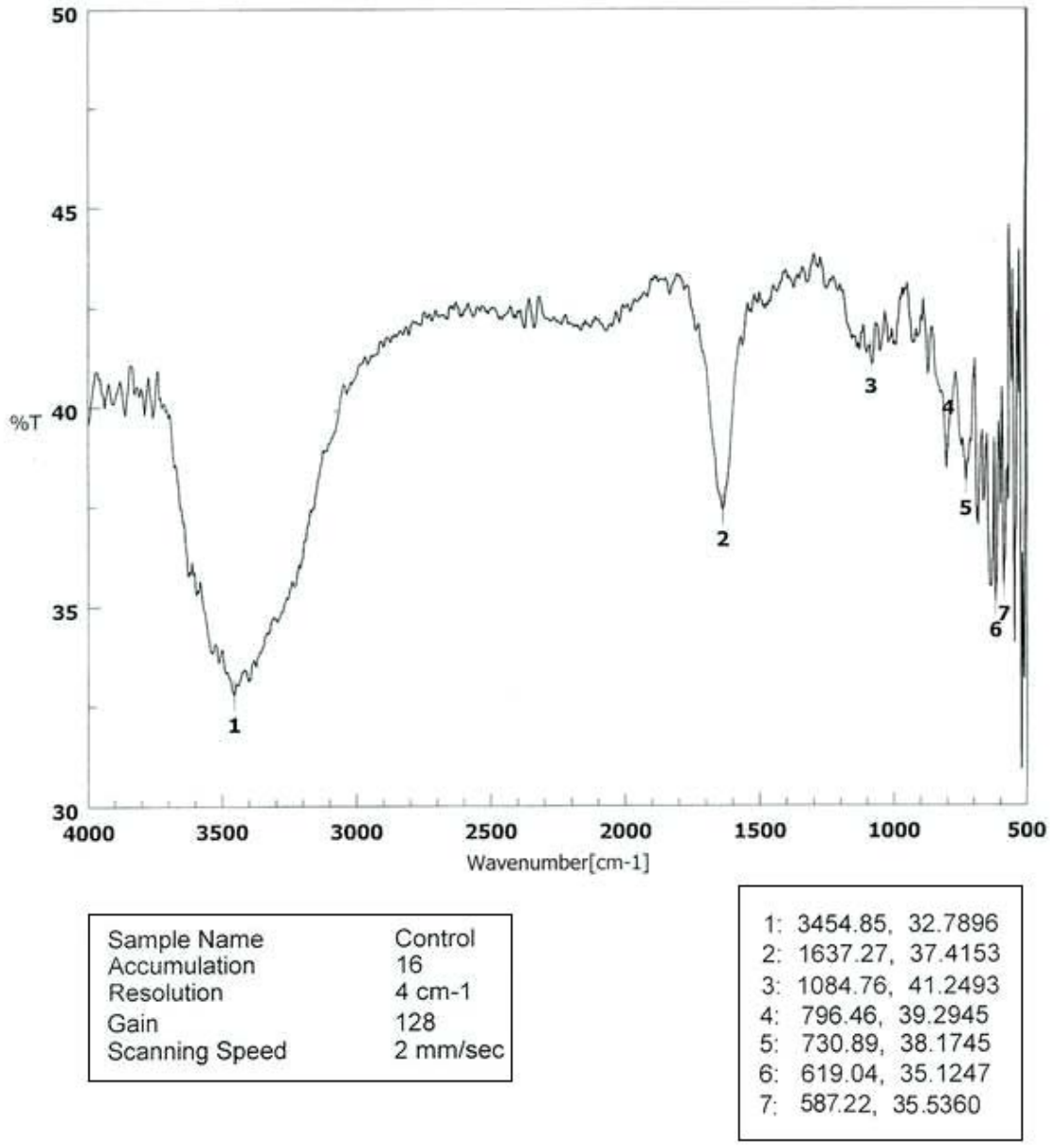

Figure 10. FT-IR analyzes result of control group sample.

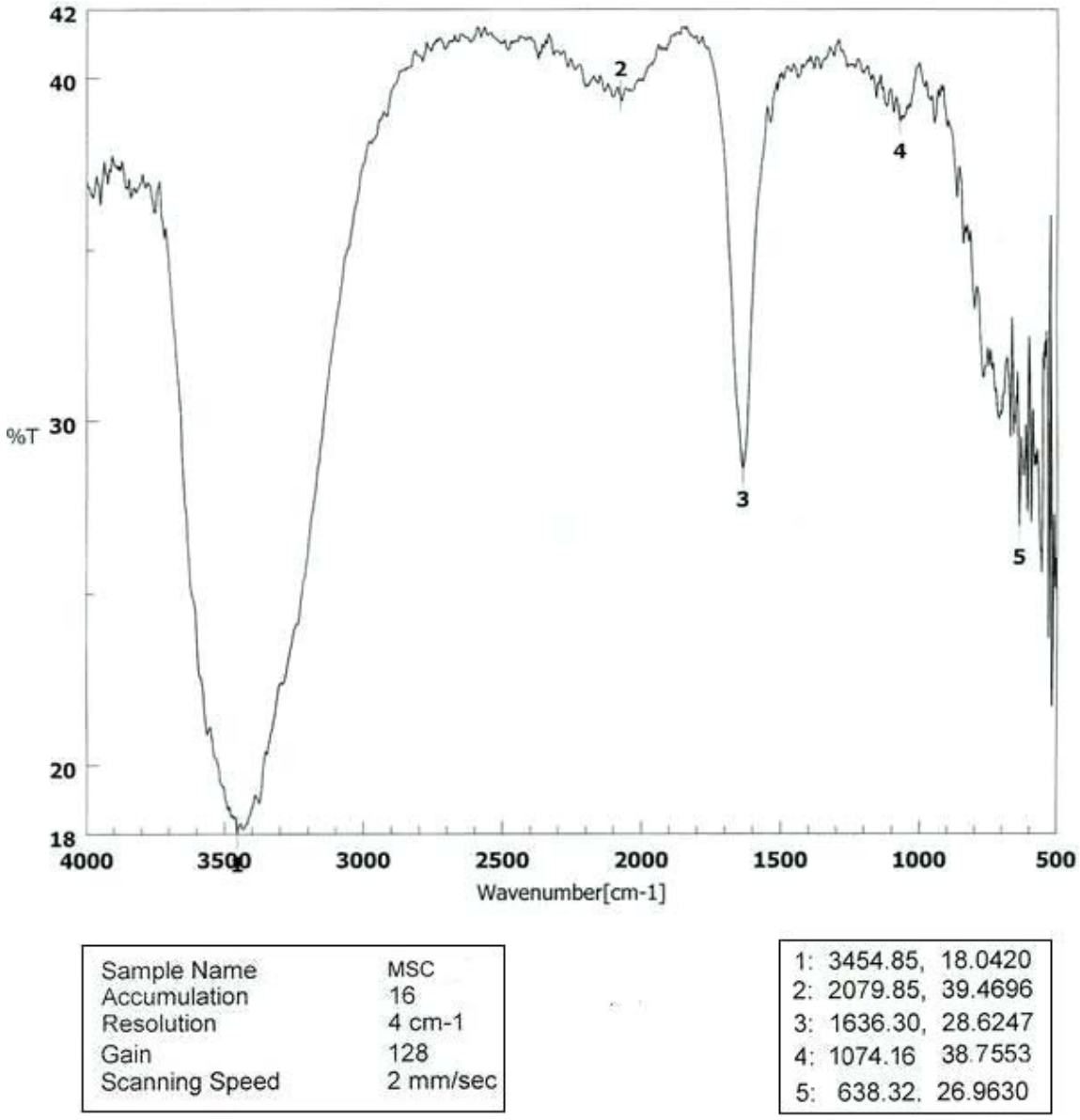

Figure 11. FT-IR analyzes result of triple combination of Semperfresh^{TM} - microwave - cold storage treatment sample.

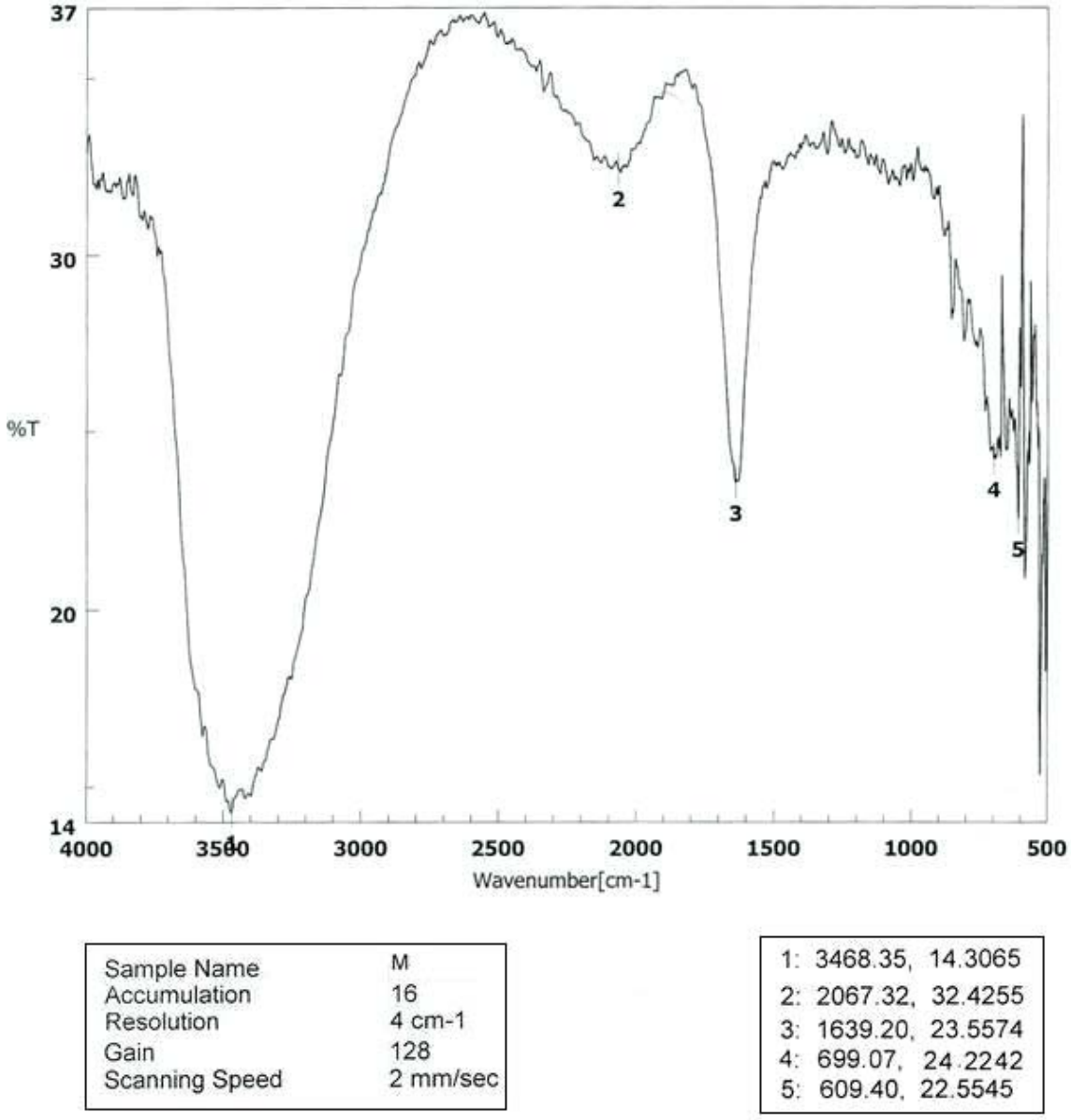

Figure 12. FT-IR analyzes result of microwave treatment sample.