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EFFICIENCY OF SOME ESSENTIAL PLANT OILS ON SEWEE DATE PALM FRUIT SPOILAGE CAUSED BY *Botrytis cinerea* UNDER COLD STORAGE CONDITIONS

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ABSTRACT: Date palm (*Phoenix dactylifera* L.) fruits is considered among the most storage products playing an important role in economic, nutrition and special life in the Arab countries. Fungi are the main source of fruit rots and spoilage, thus reduces their economic value. Seven fungal species were isolated and identified from date fruits cv. Sewee collected at harvesting of Siwa oasis, Egypt, using dilution plate method. The most frequency identified fungi were *Botrytis cinerea*, *Penicillium* sp., *Aspegillus niger*, *Alternaria alternata* and *Fusarium oxysporium*, while, *Trichoderma* sp. and *Rhizopus* sp. were the lowest detected ones. Washing of date fruits with sterile water decreased the numbers of associated fungi compared with unwashed the control treatment after harvesting (zero time). While pasteurization treatments at 80°C of Sewee date fruits completely reduced the number of fruits associated fungi followed by pasteurization at 70°C compared with zero time. *Botrytis cinerea* Fr. Res was the most frequently isolated fungus causing the highest spoilage incidence on date palm cv. Sewee. *Botrytis cinerea*, inoculated date fruits stored under cooled temperature for six months, increased infection and severity percentage. The lowest date fruits infection was recorded after one month, while the highest infection was calculated after the 6th month of cold storage. *In vitro* experiment revealed that ginger, cinnamon, clove, olive and thyme oils at 5% were the most effective essential plant oils completely inhibiting mycelial growth of *B. cinerea*, while, anise, sesame and fennel oils were the lowest effective ones. *In vivo* evaluation of antifungal activities of essential plant oils were recorded on healthy and *B. cinerea* infected date fruits as a coated treatment. Treatment date fruits with oil highly reduced spoilage caused by *B. cinerea* for 6 months cold storage period. Ginger, lemon, castor, clove, cinnamon, and olive oils were the most effective ones at 6 months of cold storage period. Generally, date fruits of infection were markedly increased with increasing storage period in oil treated and untreated fruits, as well as in inoculated or un-inoculated fruits. *Botrytis cinerea* fungal count on inoculated date palm fruits increased significantly with the prolonged storage period for all treatments. Oils coated fruits significantly recorded the lowest fungal count compared to un-treated inoculated ones. Coated date fruits with essential plant oils recorded the highest fruit firmness (FF), reduced the incensement in juice TSS, increased total sugars, markedly decreased tannin contents, and total phenols percentage as increasing cold storage period.

Key words: Date palm, fruit spoilage, plant oils, *Botrytis cinerea*, fruit firmness, TSS.

INTRODUCTION

Date palm (*Phoenix dactylifera* L.) covers 3% of the world cultivated area (Dowson, 1982). It always play an important role in the economic, nutrition and social life in the Arab countries. Egypt is one of the main date producers in the world followed by Saudi Arabia, Iran, United Arab Emirates and Pakistan (Chao and

Robert, 2007). The cultivated area in Egypt is about 50 thousand hectares, which produce about 1.590.414 tons, representing about 18% of the total worlds production. It is cultivated all over the country especially in New Valley, Aswan, Sinai, Sharkia, and Giza, Governorates. According to FAO STAT data base, Arab countries produced about 95% of the world

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production of dates (FAO, 2016). Date fruits possess high nutritional and therapeutic values with significant antioxidant, anti-proliferative, and antimicrobial properties (Rahmani *et al.*, 2014).

Date palm orchards suffer from serious fruit rot diseases varies from year to year depends on humidity and rainfall. It was estimated that, fruit rot cause 10 to 50% of post-harvest yield losses (Djerbi *et al.*, 1986). Date fruits are one of the largest storage products. Also, Olukayode *et al.* (2017) showed that, date palm fruits contain high population of fungi cause's fruit rots, produce mycotoxins and reduce the economic value of date fruits (Atia, 2011). The most common fungi causing fruit spoilage are *Aspergillus niger* and *Penicillium* sp. during handling, transport, packing and storage (El-Juhany *et al.*, 2009; Atia, 2011).

Fungi are known as the major causal organisms of date palm diseases (Khawla *et al.*, 2019). Different fungi belong to various fungal species were isolated from date fruit samples collected in Libya at zero time and after storage. *A. niger*, *Penicillium* sp. and *Fusarium oxysporium*, were isolated from date fruits (Atia *et al.*, 2009; Atia, 2011). Al-Ahmadi and Ibrahim (2009) reported that *A. niger* was the most frequently fungal species infecting date fruits. Also, *Alternaria alternata*, *A. tenuissima*, *Cladosporium* sp., and *F. oxysporium* were associated with dried date fruits in Iran (Amirmijani, 2018). Irshad *et al.* (2016) isolated *A. niger*, *A. flavus*, *Penicillium* sp. *Cladosporium* sp. and *Rhizopus* sp. from date palm fruits. *A. niger* was dominant and isolated from all the samples, causing fruit rot of different date palm varieties resulting in heavy losses annually. In addition, Al-Ahmadi and Ibrahim (2009) recommended that soft dates should be stored in a cold environment, while dry dates should be stored under dry conditions. It was observed that, the date palm fruits are mostly loaded with mixture of bacteria, molds and yeast. Therefore it is needed to appropriately treat these fruit before consumption, to minimize possible mycotic infection (Colman *et al.*, 2012).

Several plants spices have antifungal activity due to their essential oil fractions. Thyme and

clove oils presented inhibitory effects on food spoilage caused by *A. niger* and *A. flavus* (Viuda *et al.*, 2007). Carnation oil was the most effective one on growth inhibition of tested fungal followed by ginger oil, while olive oil was the lowest one. *Penicillium* spp. was the most sensitive one, followed by *Chlaropsis state* as mentioned by Atia (2011), He also added that, carnation oil treatment reduced fungal load on date whereas the lowest number was detected followed by ginger oil, compared to sodium benzoate and control. Thyme oil had strong fungicidal effect on *Penicillium* sp. and *Alternaria* sp. (Mironescua and Georgescub, 2008) and on *A. alternata* (Hadizadeh *et al.*, 2009). The cinnamon barks and leaves contain a certain fungicidal constituent exhibiting potential anti-mold activity against *A. solani* and *Curvularia lunata* (Ajay *et al.*, 2009). Also, the plant extracts of ginger, had a strong antifungal activity on the growth of *A. alternata* and *F. oxysporum* and their hydrolytic enzymes, *i.e* glucosidase, pectin lyase and protease (Fawzi *et al.*, 2009). In addition, the essential oil of *Mentha spicata* showed cidal effect on mycelial growth of *A. ochraceus*, *P. digitatum*, *Pyricularia oryzae* and *A. alternata*, while clove extracts showed inhibitory effects on different fungi. *In vitro* and *in vivo* control trials with garlic oil showed effective mycelia growth inhibition (Singh *et al.*, 2006) of *A. niger*, *F. solani* and *R. stolonifer* isolated from date fruits and its inhibition increased with increasing concentration (Anjili *et al.*, 2016).

El-Sharony *et al.* (2015) reported that, Arabic gum + black cumin oil and black cumin oil succeeded in reducing weight loss and fruit decay of Zaghoul date fruits, However, the highest value of fruit firmness was scored by Arabic gum + black cumin oil and black cumin oil. Coating date fruits with combination of Arabic gum + black cumin oil or Arabic gum only enhance of fruit chemical properties (total soluble solids and total sugars). In addition, treated orange fruits with black cumin led to reduce weight loss and decay percentages, as well as respiration rate (El-Sharony and Amin, 2014). Barhi' date fruits, at full mature stage of development, initially have a moisture content which is comparatively much higher than the critical value (23%) for yeast fermentation and

fungal attack (Al-Yahia, 1986). Rygg *et al.* (1953) studied factors effecting dates spoilage stored at room temperature. Weight losses and/or fruit decay percentage was a very important factor during post-harvest storage of avocado fruits (Adato and Gazit, 1974).

Al-Redhaiman (2004) and El-Rayes (2009) found that, neither modified atmosphere (MA) conditions nor cold storage temperature had any significant effect on total sugar contents of date fruits. Whereas, clear relationship was observed between fruit stage development and total sugar content. The more advanced stage of fruit development and ripening, the higher sugar content. In addition, tannin contents of date fruits were at maximum concentration in the Khalal stage and gradually decreased to reach a minimum concentration in the ripe stage (Sawaya and Mashadi, 1983). Tannins are water-soluble polyphenol compounds that are reported to serve as a natural defense mechanism against microbial infection. The more advanced stage of ripening the lower fruit tannin contents. Tannins content in Khalas date decreased significantly during 12 months at cold storage (Al-Ogaidi and Mutlak, 1986; Shattir *et al.*, 2002). According to Nagy and Shaw (1980) results, they showed that the major phenolic compounds found in dates are tannins, flavans, coffeoy shikimic acid, flavonols and flavones. Among which tannins are the most complex polyphenols occurring in dates. Tannins content of dates at "Tamr" stage ranges from 0.6 to 3.2% (Sawaya and Mashadi, 1983).

The study aimed to improve date palm fruits storability by testing and evaluate the effect of some essential oils on post-harvest dates palm spoilage caused by *Botrytis cinerea* and post-harvest quality parameters.

MATERIALS AND METHODS

Samples Collection

Samples of date fruits variety Sewee healthy and exhibited rot symptoms were randomly collected from date orchards at harvesting time on El-Wahat El-bahareya, Sewee, Egypt.

Samples were labelled, directly transferred under cooling to the laboratory and then divided into two groups. The first group including rotted fruits was used immediately to isolate the contaminant fungi and considered as zero time. The 2nd group including healthy fruits was stored under cooling (refrigerator) for further studies.

Isolation of Fungi Contaminated Date Fruits

Isolation of the associated fungi was done as follow; date fruits were cut into small pieces and 10 g were taken randomly from each sample separately. Fungi isolation was done using dilution method described by Johnson and Curl (1972) on potato dextrose agar (PDA) medium containing 20 µg/ml chloromphenicol and rose Bengal (30 ppm) at 28± 2°C, with 12 hr., light/dark rhythm. For isolation the (inner) fungi, complete date fruits were taken at random, washed with distilled water, surface sterilized with 0.5 % sodium hypochlorite and/or ethanol for two minutes, washed several times with sterilized distilled water and then dried with a sterilized towels and/or filter papers. Sterilized fruits were cut into small pieces with a sterilized disposable blade and cut surface was placed on PDA medium. Three replicates were used for each sample. Inoculated dishes were incubated at 28°C under 12 hr., light alternating with 12 hr., darkness. The developing fungal colonies were counted, picked up, then subculture and purified on PDA and/or Czapeck-Dox agar medium (Raper and Fennell, 1977).

Purification and Identification Fungi Associated with Contaminated and Rotted Date Fruits

The obtained fungi were purified using the single spore (Hansen, 1926) and/or hyphal tip techniques (Brown, 1924), then identified according to their morphological characters using the description of Booth (1971), Ellis (1976), Raper and Fennell (1977), Pitt (1985), Domsch *et al.* (1980) and Moubasher (1993). Isolated fungi were carried at Plant Pathology Laboratory, Plant Pathology Dept., Fac. Agric, Zagazig University. Stock cultures were maintained on PDA slants in a refrigerator for further studies.

Effect of Pasteurization at 70 and 80°C (Wet Heat) on Fungal Load Contaminated Date Fruits

Effect of pasteurization (wet heat) on fungi count of date fruits were carried out. Date samples (250 g) from the previously healthy mentioned date fruits were transferred into conical flasks 1000 ml. The flasks were tightly closed and incubated separately in a water bath, at 70°C and 80°C for 20 minutes (Lahyani, 1991; Atia, 2011). After pasteurization time flasks were, removed from water bath and immediately cooled. Fungal count was done, using the dilution method as mentioned above.

Effect of Water Washing on Fungal Load Contaminated Date Fruits

To evaluate the effect of water washing treatments on load fungal, washing was carried out using sterile water. Healthy Sewee cv. date fruit samples (100 g) were washed thoroughly with 500 ml sterile water in flasks (500 ml) for 5 minutes, then dried between two filter paper, cut into small pieces, and fungal count was done as previously mentioned above. The efficiency percentage of treatment was calculated according to Atia (2011) as follows:

$$\text{Efficiency (\%)} \text{ of treatment} = 100 - \left(\frac{T}{C} \times 100 \right)$$

Whereas:

C= total count at zero time

T= total count at treatment

Effect of *Botrytis cinerea* on Date Palm Fruits Spoilage during Storage for 6 Months

Botrytis cinerea as the most frequency isolated fungi causing fruits spoilage on date cv. sewee was grown on PDA medium and incubated at 25°C in the dark for 7 days. Resulted conidia were scrapped out from agar surface and suspended in sterile distilled water. The inoculum concentration was adjusted to be 10^6 spore/ml using haemocytometer. Sterilized date fruits were superficially wounded by removing calyx from fruits (0.5 cm in diameter). Sterilized wounded fruits were dipped for five minutes in *B. cinerea* prepared spore suspension and incubated at 20°C for 24 hr., to resemble common fungal infection. Nevertheless, wounded

un-inoculated fruits were dipped in sterilized water and preserved as a control treatment. Fruits were then forced-air cooled to 5°C and placed in white plastic containers (14 × 11 × 5 cm) each contains 10 fruits. *B. cinerea* fungal count, infection percentage and severity were recorder weekly for 6 months at cool storage. The disease severity index was calculated using the scale (0-5), where 0: no apparent rotted area around the point of infection, 1: 1-10% rotted area, 2: 11-25%, 3: 26-50%, 4: 51-75% and 5: 76-100% according to Baudion (1988). Re-isolated was carried out from infected tissues and the isolated fungi were compared with the original cultures used.

In vitro Effect of Some Selected Essential Plant Oils on Growth Reduction of *Botrytis cinerea*

This experiment was conducted to study the effect of some essential plant oils on inhibiting growth of *B. cinerea* isolated from date fruits. Pure essential oils (cinnamon, clove, thyme, ginger, castor, fennel, anise, sesame and olive) were purchased from International Flavor and Plant Oils Res Inst. Giza, Egypt. All plant oils were stored in dark bottles at 4°C, for further studies.

Different concentrations of tested oils were prepared (0 [control], 5, 10 and 15% using the agar poison technique (Raja *et al.*, 2009; Atia, 2011). Different oils concentrations were added aseptically to sterilize PDA medium in obtain required volumes in conical flasks before solidification, agitate will, and then poured in Petri dishes. Oil free medium in Petri dishes was served as control treatment. Three plates for each concentration were used and inoculated at the center with equal discs (5 mm in diameter) taken from 7 days old culture of *B. cinerea*. Then plates were incubated at $28 \pm 2^\circ\text{C}$ until mycelial growth of tested fungus covered the surface medium in control treatment (Qasem and Abu-Blan, 1996). Radial growth of the mycelium in each plate was recorded as the average of two diameters measured at right angles to one another, Percentage of linear growth reduction due to essential plant oils was done using the following formula: Growth inhibition = $R_c - R_t \div R_c \times 100$

Where, R_c is the average radial growth diameter measured in control plates and R_t is the average radial growth diameter measured in treated plates.

***In vivo* Assessment of Antifungal Activity of Selected Essential Plant Oils on Date Palm Fruits cv. Sewee and its Quality Parameters**

The antifungal activity of the selected essential plant oils against the spoilage of date palm fruits cv. Sewee caused by *B. cinerea* were conducted on infection percentage and severity of infection according to **Tian *et al.* (2011) and Tabaestani *et al.* (2013)**. As well as *B. cinerea* fungal count during 6 months of cool storage period was carried out.

The sterilized date fruits were inoculated with *B. cinerea* suspension containing 10^{-6} spore/ml incubated at 20, for 24 hr., to resemble common fungal infection. Nevertheless, wounded un-inoculated fruits were treated and preserved as control treatment. The essential plant oils treatment was done by individually immersion date fruits (inoculated wounded fruits) for five minutes in plant oils using 5% concentration and-or in sterilized distilled water in case of control treatment. Fruits were then forced –air cooled to 5°C for 15 min. and placed in plastic container (14 x 5 x 5 cm) each contains 10 fruits. All storage experiments were conducted in a cold room of Horticulture Dept., Fact. Agric., Zagazig Univ., which maintained at 7°C and 85% relative humidity. *Botrytis cinerea* fungal count (as mentioned above), percentage and severity of infection were recorder and calculated monthly average for 6 months under cold storage condition.

Quality parameters of oil treated fruits were evaluated weekly for 6 months storage. Fruits firmness (FF), total soluble solids (TSS), tannins, sugars fraction and phenolic fraction were detected by photometrical methods described by **Al-Radhaiman (2004)**.

Fruit firmness

Fruit firmness were measured as the maximum force required to penetrate the fruit to a puncturing depth of 10 mm as $Ib/inch^2$, using a 6-mm cylindrical tip of pressure tester. (A

Push Pull Dynamometer Model FD101, Penetrometer). The values were expressed as $Ib/inch^2$.

Juice total soluble solids percentage (TSS%)

TSS % was measured in fruit juice using a hand-held refractometer.

Total sugars were determined in the methanol extract

It was determined using the phenol sulphuric acid the percentage of total sugars was calculated on dry weight basis according to **Dubois *et al.* (1956)**.

Soluble tannins content

The soluble tannins were measured by the Folin-Denis method (**Taira, 1996**). A quantity of five grams of treated sample were placed directly into a solution of 25 ml of methanol 80% and homogenized in a blender. Thereafter, samples were filtered and centrifuged at 14000 rpm for 20 min at 4°C, and the supernatant was reserved. More supernatant was extracted from the precipitant with methanol 80% and added to the first. The total supernatant was brought to 100 ml with distilled water. One ml of this sample solution and 6 ml of distilled water were mixed. Thereafter, 0.5 ml of 1 N phenol reagent (Folin- Ciocalteau reagent) was added and shaken thoroughly. After 3 min, one ml of saturated Na_2CO_3 was added, vortex, and 1.5 ml of distilled water was added. The absorbance was measured after one hr., at 750 nm, soluble tannins were calculated as mg gallic acid equivalents per gram of fresh weight basis.

Total phenol contents

A quantity of 5 g of treated fruit tissue was homogenized in 15 ml ethanol 95% and boiled for 15 minutes. The homogenate was filtrated through Whatman No.1 filter paper. A quantity of 0.5 ml Folin-Denis reagent was added to one ml of the alcoholic extract and after 5 minutes, 7 ml saturated sodium carbonate solution was added, shaken and left for 0.5 hr. Optical density was measured at 750 nm and total phenols were calculated from a standard curve of gallic acid. Obtained data were expressed as the mg gallic

acid equivalents per gram of fresh weight basis according to **Slinkard and Singleton (1977)**.

Statistical Analysis

Data were statistically analyzed by analysis of variance according to **Snedecor and Cochran, (1982)** using SPSS system version 8 (1997).

RESULTS AND DISCUSSION

Isolation and Identification

The mycological experiments of date palm cv. Sewee revealed that the total count of fungi in all samples ranged from 72.50 to 252.80 x 10⁴ colony/g dates at zero time (after harvesting). Seven species fungus belong to seven genera were isolated and identified from the collected date palm fruit samples. Members of *Botrytis*, *Penicillium*, *Aspergillus*, *Fusarium* and *Alternaria* genera were isolated and identified fungi recorded 252.80, 183.90, 160.90, 137.90 and 114.90 colony/g date, respectively. While, *Trichoderma* sp., and *Rhizopus* sp., were the lowest detected ones recorded 72.50 and 91.90 colony/g date, respectively (Table 1).

Results presented in this investigation demonstrate that stored date fruits contained high number of fungi. These fungi include mycotoxin producing genera. Increasing cool storage period increased total fungal contamination. These results are in accordance with those obtained by **Abdul-Rahman et al. (2004) and Atia (2011)**. They reported that dry dates were highly polluted with various fungal genera and species. The most abundant genus found in date fruits was *A. niger* (**Hasnaoui et al., 2010; Atia et al. 2009**). Additionally date fruits was subjected to spoilage fungi genera *e.g.* *Alternaria*, *Botrytis*, *Fusarium*, *Aspergillus* and *Penicillium*, which reported to cause fruits damage (**Bokhary, 2010**).

Effect of pasteurization on fungal count associated with date palm fruits

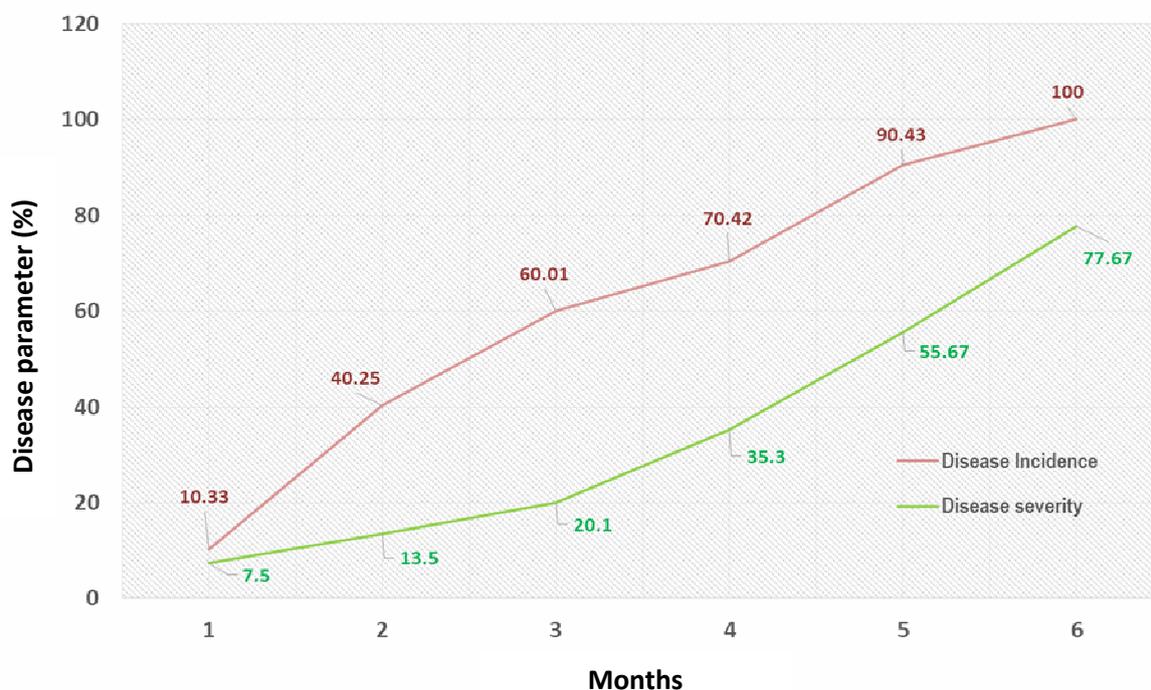
Pasteurization (wet heat) treatments at 80°C of date fruits completely reduced the number of fruits associated fungi (0.00 colony/g date and 100% efficiency) followed by pasteurization treatments at 70°C (40.17 colony/g date and 72.29% efficiency) compared with zero time,

which recorded 144.97 colony/g date (Table 1). *Penicillium* sp, was the sensitive fungus to pasteurization treatments at 70°C which recorded (30.01 colony/g date and 83.68% efficiency) followed by *Rhizoipus* (20.34 colony/g date and 77.87% efficiency) and *Botrytis* (70.02 colony/g date and 72.30% efficiency). While *Fusarium*, was the resistance fungus to pasteurization treatments at 70°C (50.32 colony/g date and 56.21% efficiency) followed by *Aspergillus* (50.01 colony/g date and 68.92% efficiency) (Fig. 1). Similar results were obtained by **Harrak (2003) and Atia (2011)**. They found, that hot air at 60°C for 1-1.5 hr lead to complete control of organisms on stored date and don't change its physical or chemical characters. Also, heating date at 65°C for 20 minutes using water bath completely controlled microorganisms on stored date (**Lahyani, 1991**).

Water washing treatment of cv. Sewee date fruits using sterile distilled water decreased numbers of associated fungi which recorded 83.17 colony/g date and 42.63% efficiency compared with zero time, which recorded 144.97 colony/g date (Table 1). *Fusarium*, was the sensitive fungus to water washing treatments which recorded (80.77 colony/g date and 29.70% efficiency) followed by *Botrytis* (150.02 colony/g date and 40.66% efficiency). While *Alternaria*, was the resistance fungus (70.30 colony/g date and 49.02% efficiency) followed by *Penicillium* (100.05 colony/g date and 45.59% efficiency) (Table 1). Similar results were found by **Atia (2011)**, he found that, washing date fruits with water has kept down or eliminated the initial fungal contaminated fruits. Furthermore, high efficacies against *Neofabraea* spp. and *Penicillium expansum* have been obtained after incubation in hot-water dipping (HWD) for up to 3min. (**Maxin and Klopp, 2004**). Hot-water rinsing (HWR) for 30°C at temperatures above 50°C has been developed to control post-harvest pests and diseases of a range of horticultural products (**Fallik, 2004**). HWD has been introduced into organic apple production (**Maxin et al., 2006**). Furthermore, there is only limited information on the range of fungi that can be controlled by HWD and especially HWR. In preliminary studies, **Maxin and Weber (2011) and Maxin et al. (2012)** have shown that HWD could successfully control various storage rots on artificially inoculated apples.

Table 1. Frequency of fungi isolated from date palm fruits cv. Sewee post-harvesting (zero time), water washing efficiency and pasteurization at 70 and 80°C

Isolated fungus	Zero time – post harvesting	Water washing	Water treatment Efficiency (%)	Pasteurization at 70°C	Pasteurization at 70° C treatment Efficiency (%)	Pasteurization at 80°C
<i>Botrytis sp.</i>	252.80	150.02	40.66	70.02	72.30	0.00
<i>Alternaria sp.</i>	137.90	70.3	49.02	40.22	70.83	0.00
<i>Fusarium sp.</i>	114.90	80.77	29.70	50.32	56.21	0.00
<i>Penicillium sp.</i>	183.90	100.05	45.59	30.01	83.68	0.00
<i>Aspergillus sp.</i>	160.90	90.54	43.73	50.01	68.92	0.00
<i>Rhizopus sp.</i>	91.90	50.26	45.31	20.34	77.87	0.00
<i>Trichoderma sp.</i>	72.50	40.26	44.47	20.25	72.07	0.00
Total	1017.8	582.20		281.17		0.00
Mean	144.97	83.17	42.63	40.17	72.29	0.00

Fig. 1. Infection and severity percentages on *Botrytis cinerea* inoculated date palm fruits cv. Sewee, after 6 months of cold storage period (7°C ±2°C)

Botrytis cinerea Fr. Res., was the most frequency isolated fungus causing spoilage on date palm cv. Sewee. *B. cinerea* fungal identification was done at the Laboratory of Plant Pathology, Faculty Agriculture, Zagazig University. The mycological experiments of dates during cold storage conditions for 6 months revealed that, the percentage and severity of infection of *B. cinerea* were increased monthly with increasing cold storage period. The lowest percentage and severity of infection of *B. cinerea* were resulted after one month of cold storage [10.33 infection (%) and 7.50 severity (%)] compared to healthy fruits. While, the highest infection percentage and severity were obtained after the 6th month of cold storage [100.0 respectively (%) and 77.67 respectively (%)] (Fig. 1). These results are in accordance with those obtained by **Atia (2011)**, who reported that the dry dates were highly polluted with various fungal genera and species. The most abundant genus found in date palm fruits was *A. niger*. Also, date palm fruits in Egypt was subjected to spoilage fungi genera *e.g. Alternaria, Botrytis, Fusarium, Aspergillus* and *Penicillium* (**Bokhary, 2010**).

Antifungal Activity of some Essential Plant Oils *In vitro* and *In vivo*

Essential plant oils at 10 and 15% completely inhibited *B. cinerea* fungal growth (date not shown). While, at 5% concentration, ginger oil was the most effective one followed by cinnamon, clove, olive thyme and castor oils [98.03, 90.22, 88.01, 85.33, 77.25 and 75.66 reduction (%)]. While anise oil was the lowest effective one followed by sesame and fennel oils [10.33%, 24.44 and 45.58 reduction (%)]. Generally, increased oil concentration resulted in complete reduction of mycelia growth (Fig. 2).

The antifungal activity of some essential oils were tested on date cv. Sewee fruits as a coated treatment on inoculated date fruits with *B. cinerea* and un-inoculated fruits (Table 2). Treatments of date palm fruits with selected oils only were highly significant reduced fungal infection and prevented spoilage caused by *B. cinerea* for 6 months cold storage period. Ginger was the most effective in reducing artificial inoculation with *B. cinerea*, followed by lemon, castor, clove, cinnamon, olive [1.67, 1.67, 5.0, 5.0, 6.67 and 6.67 infection (%)]. While fennel, sesame and anise were the lowest effective in reducing artificial inoculation with *B. cinerea* [13.33, 20.00 and 26.67 infection (%)]

compared to non-inoculated and inoculated controls [16.66 and 55% infection (%)] (Table 2).

Damaged date palm fruits increased significantly with the prolongation of the storage period for all treatments. Generally, damaged date palm fruits were markedly increased with increasing cold storage period. The lowest damaged fruits resulted after one month of cold storage. While, the highest damaged fruits were recorded after the 6th month of cold storage. Generally, the *B. cinerea* inoculated fruits recorded the highest damaged fruits than un-inoculated fruits. Healthy date palm fruits treated oils significantly recorded the lowest damaged fruits compared to un-treated fruits. In addition, inoculated date palm fruits treated with oils significantly recorded the lowest damaged fruits compared to un-treated inoculated fruits. The most of tested oil completely prevented damage of date fruits for the 6th month. As well as, the most of tested oil completely inhibited *B. cinerea* infection for three months.

Similar results were obtained by **Viuda (2007)**, **Fawzi *et al.* (2009)**, **Suwitchayanon and Kunasakdakul (2009)** and **Atia (2011)**. Application of essential oils for post-harvest disease control of fresh product, will allow a safer and eco-friendly acceptable strategy of post-harvest diseases (**Hadizadeh *et al.*, 2009**). The antifungal activity of the essential oils is different, depending on the fungal type (**Mironescua and Georgescub, 2008**). The inhibitory effects of plant oils might be regarded to which act as cidal agent against fungal growth and showed abnormal conidia and malformations as swollen, often septated and pale color of hypha (**Suwitchayanon and Kunasakdakul, 2009**). **Raafat *et al.* (2016)**, found that, decay of healthy and inoculated cherry tomato with spoilage pathogens *Alternaria alternata* and *Botrytis cinerea* were decreased in coated fruits treated with essential oils (black seed, fenugreek, cinnamon, spearmint, basil, and thyme).

Effect of Essential Plant Oils on *Botrytis cinerea* Fungal Count on Inoculated Date Palm Fruits

Botrytis cinerea fungal count on inoculated date palm fruits increased significantly with the prolongation of the storage period for all treatments. Generally, *B. cinerea* fungal count was markedly increased as cold storage period increased. The lowest *B. cinerea* count resulted



Fig. 2. Effect of different essential plant oils at 5% concentration on growth reduction of *Botrytis cinerea* isolated from cv. Sewee date fruits

Table 2. *In vivo* antifungal activity of some essential plant oils at 5% concentration on percentage spoilage of cv. Sewee date fruits under cool storage for 6 months

Treatment	Cold storage period (month)						Average	LSD (at 0.05)
	1	2	3	4	5	6		
Control	0.0	0.0	0.0	20	30.0	50.0	16.67	9.77
Inoculated	10	30	40	60	90	100	55.00	18.66
Ginger	0.0	0.0	0.0	0.0	0.0	0.0	0.00	0.0
Ginger + inoculated	0.0	0.0	0.0	0.0	0	10	1.67	3.33
Clove	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Clove + inoculated	0.0	0.0	0.0	0.0	10.0	20.0	5.0	3.44
Cinnamon	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cinnamon + inoculated	0.0	0.0	0.0	0.0	10.0	30.0	6.67	7.35
Olive	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Olive + inoculated	0.0	0.0	0.0	0.0	10.0	30.0	6.67	7.35
Fennel	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Fennel+ inoculated	0.0	0.0	0.0	10.0	30.0	40.0	13.33	14.87
Thyme	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Thyme + inoculated.	0.0	0.0	0.0	0.0	10.0	30.0	6.67	7.35
Lemon	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Lemon + inoculated	0.0	0.0	0.0	0.0	0.0	10.0	1.67	3.33
Anise	0.0	0.0	0.0	0.0	20.0	30.0	8.33	6.45
Anise+ inoculated	0.0	0.0	0.0	40.0	50.0	70.0	26.67	12.80
Sesame	0.0	0.0	0.0	0.0	20.0	50.0	11.67	4.33
Sesame + inoculated	0.0	0.0	0.0	0.0	40.0	80.0	20.0	6.07
Castor	0.00	0.00	0.00	0.0	0.0	0.0	0.0	0.0
Castor + inoculated	0.00	0.00	0.00	0.0	10.0	20.0	5.0	3.33

after one month of cold storage. While, the highest fungal count was recorded after the 6th month (Table 3). Generally, the *B. cinerea* inoculated fruits with *B. cinerea* recorded the highest count than un-inoculated fruits.

In un-inoculated date palm fruits treated with oils significantly prevented fungal count compared to un-treated inoculated fruits. With the excepting of the anise and sesame which were effective till the 4th month most of tested oils treatment completely prevented growth of *B. cinerea* for 6 months cold storage, whereas, in case of inoculated date palm fruits treated with oils significantly prevented fungal count compared to un-treated inoculated fruits, excepting in case of fennel, anise and sesame. Limon was effective till the 5th month.

Lemon was the most effective in reducing count of *B. cinerea* on date fruits followed by olive, clove, castor, thyme, ginger and cinnamon (3.33, 6.67, 15.00, 20.00, 21.67, 23.33 and 25.00 colony/g date). While anise, sesame and fennel were the lowest effective in reducing artificial inoculation with *B. cinerea* (31.67, 50.00 and 51.67 colony/g date) compared to non-inoculated and inoculated controls (75.00 and 386.6 colony/g date) (Table 3).

A similar effect was observed by **Ben-Yehoshua (1969)** in orange fruits coated with wax Also, **Banks (1984)**, reported that, sucrose ester-based coating on banana fruits extended their storage life. In addition, **Tabaestani *et al.* (2013)** suggest that, the cherry tomatoes weight loss can be delayed and storage period life can be extended when tomatoes stored at 20°C. As well as **Raafat *et al.* (2016)**, found that, essential plant oils may improve control decay of tomato fruits during cold storage.

Date Palm Fruit Firmness (FF)

Coated date palm fruits with essential plant oils recorded the highest date palm firmness (FF) in un-inoculated and *B. cinerea* inoculated fruits than those un-treated inoculated control fruits. Generally, date fruits FF was markedly decreased under storage period and increased monthly till the 6th month of cold storage. The highest FF values resulted at the first month while, the lowest FF values were recorded after the 6 months of inoculated and un inoculated

treated oil as well as in un-treated inoculated and -inoculated fruits (Table 4). Clove oil treated date fruits recorded the height FF in un-inoculated and inoculated fruits with *B. cinerea* at storage period for 6 months (459.4 and 386.5), followed by, cinnamon oil and ginger (382.3 and 375.8) in case of untreated fruits. While, fennel and sesame oils recorded the lowest FF, in case of treated inoculated date fruits (280.5 and 294.8).

These results are in agreement with those reported by **Al-Redhaiman (2004)**. He reported that lowest fruit firmness values were observed in control fruits, which totally collapsed after 60 days of storage. Moreover, FF was closely associated with fruit ripening process during the storage period, the more advanced stage of ripening, the lower FF. Also, in other studies FF showed directly proportional values to CO₂ concentration used in the modified atmosphere treatments. Moreover, cold storage delayed fruit ripening and extend the shelf life of dates compared with store at ambient conditions (**Al-Obeed, 2010**). In addition, **Aleid *et al.* (2014)** observed that the main reason for the change of firmness of date fruits during storage period was probably due to the increase in cell rigidity and subsequent strengthening of cell wall bonding. Low temperature storage could therefore be used to effectively strength the firmness of date fruits. Moreover, infiltration of calcium can delay the overall softening process during ripening (**Tirmazi and Wills, 1981; Davenport, 1984**).

Total Soluble Solids (TSS Brix°)

Coated date palm fruits with essential plant oil reduced the increment in juice TSS in un-inoculated and inoculated fruits with *B. cinerea* than of inoculated control and un-treated fruits. TSS was increased in untreated inoculated fruits than in un-inoculated fruits (control). Generally, the highest TSS values was recorded at the 6th month and the lowest TSS values was recorded at the 1st month at cold storage. Castor, clove, cinnamon, fennel, thyme and ginger oils were the most effective on reducing juice TSS in un-inoculated fruits treated with previously oils which recorded the lowest value of TSS (21.31, 21.93, 22.06, 22.28, 22.31 and 22.96). While castor, anise, cinnamon and clove oils were the

Table 3. *In vivo* antifungal activity of some essential oil at 5% concentration on fungal count of *Botrytis cinerea* the causal of fruit spoilage under cool storage for 6 months

Treatment	Cold storage period (month)						Average	LSD
	1	2	3	4	5	6		
Control	0.0	0.0	0.0	80	150	220	75	13.22
Inoculated	70.0	280.0	350.0	470.0	530.0	620.0	386.67	32.44
Ginger	0.0	0.0	0.0	0.0	0.0	0.0	0	0.0
Ginger+ inoculated	0.0	0.0	0.0	0.0	50.0	90.0	23.33	6.33
Clove	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Clove+ inoculated	0.0	0.0	0.0	0.0	30.0	60.0	15.0	4.23
Cinnamon	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cinnamon+ inoculated	0.0	0.0	0.0	0.0	40.0	110	25.0	3.55
Olive	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Olive+ inoculated	0.0	0.0	0.0	0.0	10.0	30.0	6.67	2.75
Fennel	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Fennel+ inoculated	0.0	0.0	0.0	50.0	110.0	150.0	51.67	12.33
Thyme	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Thyme+ inoculated.	0.0	0.0	0.0	0	40.0	90.0	21.67	8.66
Lemon	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Lemon+ inoculated	0.0	0.0	0.0	0.0	0.0	20.0	3.33	3.14
Anise	0.0	0.0	0.0	0.0	20.0	50.0	11.67	4.77
Anise+ inoculated	0.0	0.0	0.0	20.0	60.0	110	31.67	5,87
Sesame	0.0	0.0	0.0	0.0	40.0	130	28.33	2.67
Sesame + inoculated	0.0	0.0	0.0	50.0	90.0	160	50.0	8.5
Castor	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Castor + inoculated	0.0	0.0	0.0	0.0	30.0	90.0	20.0	12.02

Table 4. *In vivo* activity of some essential plant oils at 5% on percentage firmness of un-inoculated and inoculated cv. Sewee date fruits with *Botrytis cinerea* under cool storage for 6 months

Treatment	Cold storage period (month)						Average	LSD (at 0.05)
	1	2	3	4	5	6		
Control	450	322	207.5	112	85	70	207.8	15.1
Inoculated	300	220	194	90	60	30	149	10
Ginger	621	478	391.5	315.3	270.5	178.5	375.8	34.2
Ginger+ inoculated	599	455	370.5	275	213.8	141	342.3	34.1
Clove	667.1	575	491	416.9	362.4	243.38	459.4	53.3
Clove+ inoculated	606.3	491	416	336	276.8	193.2	386.5	44
Cinnamon	598.8	503	374.8	315.3	294.5	207.75	382.3	47.2
Cinnamon+ inoculated	571.8	492	357.5	273.8	256.3	170.25	353.5	33.4
Olive	549.5	442	348.8	276.8	194.3	127.25	323.1	40.5
Olive+ inoculated	502.5	426	331.5	252.3	173	89.5	295.7	39.8
Fennel	549.5	468	308.8	213.5	206.3	86.25	305.4	55.7
Fennel+ inoculated	502.5	442	262.8	160	188.3	128	280.5	42.6
Thyme	572	486	413	313	231.8	160.75	364.4	38.6
Thyme+ inoculated	555.3	475	386.8	296.5	207	88.75	334.9	39.9
Lemon	572	493	398	296.3	194.8	107.25	343.5	43.8
Lemon+ inoculated	554	480	376.3	283.8	182.8	75.75	325.4	43.9
Anise	549.5	468	391	293.8	195.5	142.75	340.1	37.7
Anise+ inoculated	502.5	417	328	262.8	168.8	103	297	37
Sesame	558.5	438	358.8	297.3	193.8	109.25	326	43.2
Sesame + inoculated	517.5	393	319	277.8	167	94.25	294.8	42
Castor	572	496	313	231.8	177.5	126.25	319.4	34.6
Castor + inoculated	555.3	458	296.8	207	146.3	99.75	293.8	38.3

most effective on reducing juice TSS in inoculated oil treated fruits recorded the lowest value (23.08, 23.43, 23.65 and 23.78) (Table 5).

These results are in agreement with those reported by **Al-Kahtani et al. (1998)**, and **Azelmat et al. (2005)**. They reported that, TSS content in date increased gradually with the increasing storage period. Significant increase in TSS content could be due to the degradation in insoluble compounds present in date fruit into soluble compounds such as the conversion of proto pectin into pectin. In addition, **El-Rayes (2009)** found that, a slight increase in TSS occurred in most treatments under investigation. This increase could be due to the conversion of some insoluble compounds into soluble compounds shown by **Thompson and Abboodi (2003)**. Moreover, **Aleid et al. (2014)** studied fruit quality of two date cultivars, under cold storage at 5°C for 12 months, they reported that TSS is a parameter significantly correlated with the perception of sweetness, date flavor and aroma intensity. TSS of date was significantly increased under cold storage at 5°C for 12 months.

Total Sugars

Oils treated date palm fruits significantly increased total sugars in healthy fruits. Total sugars in date palm fruits markedly increased as cold storage period increased. The lowest in fruits total sugars content resulted after one month of cold storage. While, the highest total sugars content was recorded after six months of cold storage.

Total sugars in date palm fruits markedly increased in inoculated fruits with *B. cinerea* as well as un-inoculated fruits. The highest total sugars were recorded in date palm fruits treated with anise oil in *B. cinerea* inoculated and un-inoculated date fruits followed by ginger and clove oils, respectively (Table 6).

These results are in agreement with those reported by **Ozkaya et al. (2009)** who reported that strawberries had higher glucose amount after 10 days of modified atmosphere storage compared with control. **Raafat et al. (2016)** also, found that, cherry tomato fruits inoculated with *A. alternata* and coated with thyme oil caused increase of all sugar fraction concentrations. The same trend heaving was also observed in case of date fruits inoculated with *B. cinerea* and coated

with cinnamon and fenugreek oil, respectively. Increased total sugar content might be due to conversion of starch into sugars (**Tsuda et al., 1999**). Also, **Vesaltalab and Gholami (2012)** and **Nabifarkhani et al. (2015)**, reported that, total sugar content increased during storage period that, may be due to the dehydration and decomposition of organic acids in the fruits

Total Tannin Content

Tannins content in date palm fruits markedly decreased as cold storage period increased. The highest fruit tannins content resulted after one month of cold storage. While, the lowest tannins content was recorded in healthy date palm fruits after six months of cold storage. Treated fruits with oils significantly increased tannins content in fruits compared to un-treated. Total tannins in date palm fruits markedly increased in *B. cinerea* inoculated fruits as well as in inoculated treated with oils. The highest total tannins were recorded in date palm fruits treated with anise oil followed by fennel, ginger, cinnamon and clove in *B. cinerea* inoculated fruits and un-inoculated one (Table 7).

Similar, results were found by **Al-Ogaidi and Mutlak (1986)**, **Abu-Goukh et al. (2003)** and **Al-Redhaiman (2004)**. They cleared that tannin contents of different date fruit cultivars were at maximum concentration in the Khalal (Bisr) stage (full mature stage of development) and gradually decreased to reach a minimum concentration in the ripe stage (Rutab) during cold storage.

Total Phenol

Total phenol percentage (g/100g f.wt.) in date palm fruits markedly decreased as cold storage period increased. The highest fruit total phenol percentage resulted after one months of cold storage. While, the lowest total phenol percentage was recorded after six months of cold storage. Healthy date palm fruits treated oils significantly recorded the highest total phenol percentage in fruits compared to un-treated fruits. Total phenol percentage in date palm fruits markedly decreased in *B. cinerea* inoculated fruits as well as oil treated un-inoculated fruits. The highest total phenol percentage was recorded in date palm fruits treated with anise oil followed by ginger oil inoculated with *B. cinerea* as well as in un-inoculated date fruits.

Table 5. *In vivo* activity of some essential plant oils at 5% concentration on TSS percentage of un-inoculated and inoculated *B. cinerea* Seewe date fruits under coal storage for 6 months

Treatment	Cold storage period (month)						Average	LSD (at 0.05)
	1	2	3	4	5	6		
Control	12	18	25	31	36	39	26.83	7.78
Inoculated	16	21	28	33	38	40	29.33	10
Ginger	9.7	14.3	19.2	29.7	30	34.9	22.97	2.00
Ginger+ inoculated	11.3	17.2	21.4	26.5	31.5	36.5	24.07	3.01
Clove	9.6	14.3	18.8	23.8	29.6	35.5	21.93	2.4
Clove+ inoculated	10.9	16.6	20.5	26.1	30.3	38.3	23.78	3.5
Cinnamon	9.9	16	20.5	25	29	32	22.07	2.1
Cinnamon+ inoculated	11.4	17.1	22	27.2	30.5	33.7	23.65	3.3
Olive	9.7	16.2	23.3	29.8	38.07	39.5	26.10	4.9
Olive+ inoculated	11.3	17.5	24.5	31.3	39.5	42.2	27.72	7.5
Fennel	8.5	13	15.5	25	33.3	38.4	22.28	3.4
Fennel+ inoculated	9.8	15.2	21.2	27.1	39.7	46.9	26.65	3.3
Thyme	9.7	13.2	21.4	26	29.5	34.1	22.32	3.5
Thyme+ inoculated	11.5	17.7	23.7	28.6	33.7	37.4	25.43	4.8
Lemon	8.3	14.7	21.8	28.7	34.7	39.5	24.62	3.3
Lemon+ inoculated	10.2	15.7	23.3	30.2	37.3	41	26.28	5.07
Anise	11.2	16.5	21.8	27.4	30.2	35.3	23.73	2.7
Anise+ inoculated	12.2	18.8	22.7	28.9	31.9	38.3	23.43	3.1
Sesame	9.7	15.2	19.7	24.7	30.6	37.2	22.85	2.3
Sesame + inoculated	11.1	16.8	21	26.3	32.6	39.6	24.57	2.5
Castor	8.8	12.2	17.7	23.7	29.8	35.7	21.32	2.8
Castor + inoculated	9.7	15.5	19.4	26.2	31.3	36.4	23.08	3.2

Table 6. Effect of some essential plant oils on treated infected date fruits with *B. cinerea* at different cold storage periods and their interactions on fruit total sugars percentage of Sewee dates

Treatment	Months	Control	Fennel	Castor	Thyme	Anise	Ginger	Clove	Cinnamon
Uninoculated fruits	1	19.43	21.79	21.19	22.08	24.49	24.08	23.94	23.65
	2	20.93	21.82	21.66	23.52	24.85	24.54	24.01	24.23
	3	20.84	22.70	21.73	23.69	25.51	24.98	24.58	24.69
	4	22.46	23.30	22.74	26.81	26.18	26.14	26.05	26.05
	5	23.35	24.31	21.31	26.24	27.62	27.31	26.51	26.92
	6	25.35	26.31	23.31	28.24	30.62	30.31	28.51	27.92
	Average		22.06	23.37	21.99	25.10	26.55	26.23	25.60
LSD 0.05 %		2.27							
<i>Botrytis cinerea</i>	1	23.96	26.15	25.46	27.23	29.38	28.93	28.4	28.49
	2	25.09	25.92	25.55	28.19	29.79	29.04	28.64	28.92
	3	25.20	27.05	26.26	28.64	30.40	30.18	29.33	29.64
	4	25.91	27.27	26.34	29.79	30.64	30.27	29.95	30.16
	5	27.51	28.72	28.49	30.91	32.64	32.01	31.33	31.72
	6	29.51	29.30	31.49	32.91	34.64	34.01	32.33	32.72
	Average		26.20	27.40	27.27	29.61	31.25	30.74	30.00
LSD 0.05 %		2.20							

Table 7. Effect of some essential plant oils on treated infected date fruits with *B. cinerea* and their interactions on fruit tannins (%) (mg/ 100g fresh weight) of Sewee dates at different storage period.

Treatment	Months	Control	Fennel	Castor	Thyme	Anise	Ginger	Clove	Cinnamon
Uninoculated fruits	1	5.15	5.95	5.29	5.85	6.69	6.01	5.91	6.11
	2	4.64	5.53	4.72	5.27	6.21	5.36	5.3	5.53
	3	4.44	5.05	4.53	5.04	5.74	5.21	5.12	5.27
	4	4.07	4.94	4.35	4.68	5.55	4.89	4.79	4.97
	5	3.77	4.53	3.92	4.24	5.15	4.51	4.38	4.56
	6	2.40	3.04	2.66	3.02	4.15	3.22	2.38	2.56
	Average		4.08	4.84	4.25	4.68	5.58	4.87	4.65
LSD 0.05 %		1.19							
<i>Botrytis cinerea</i>	1	0.59	1.35	0.78	0.67	1.51	0.88	0.80	0.96
	2	0.50	1.17	0.62	0.57	1.31	0.70	0.65	0.80
	3	0.35	1.06	0.47	0.40	1.20	0.53	0.49	0.65
	4	0.32	0.99	0.44	0.37	1.11	0.50	0.44	0.60
	5	0.31	0.79	0.41	0.35	0.90	0.47	0.42	0.53
	6	0.28	0.74	0.50	0.41	0.30	0.42	0.39	0.45
	Average		0.39	1.02	0.54	0.46	1.06	0.58	0.53
LSD 0.05 %		0.25							

Table 8. Effect of some post-harvest treatments; cold storage periods and their interactions on fruit phenols (%) (mg/ 100g fresh weight) of Seewe dates

Treatment	Months	Control	Fennel	Castor	Thyme	Anise	Ginger	Clove	Cinnamon
Uninoculated fruits	1	0.513	0.597	0.563	0.590	0.670	0.640	0.620	0.630
	2	0.463	0.547	0.503	0.523	0.613	0.563	0.540	0.560
	3	0.450	0.503	0.467	0.517	0.563	0.530	0.537	0.536
	4	0.360	0.450	0.440	0.410	0.520	0.490	0.450	0.467
	5	0.343	0.447	0.413	0.390	0.510	0.467	0.433	0.450
	6	0.243	0.227	0.202	0.300	0.420	0.333	0.232	0.334
Average		0.395	0.462	0.431	0.455	0.549	0.504	0.469	0.496
LSD 0.05 %		0.13							
Botrytis cinerea	1	0.090	0.167	0.14	0.107	0.187	0.157	0.137	0.146
	2	0.053	0.107	0.1	0.063	0.120	0.110	0.090	0.095
	3	0.040	0.097	0.073	0.050	0.107	0.083	0.063	0.075
	4	0.040	0.080	0.093	0.047	0.090	0.103	0.057	0.074
	5	0.027	0.063	0.057	0.030	0.073	0.060	0.033	0.049
	6	0.017	0.013	0.017	0.003	0.013	0.040	0.022	0.029
Average		0.045	0.088	0.080	0.050	0.098	0.092	0.067	0.078
LSD 0.05 %		0.049							

These results were similar in trend with **Osman (1984) and Shattir *et al.* (2002)**. They revealed that, Barakawi dates had significant higher total phenolic compounds that make them unpalatable at "Khalaal" and "Rutab" stages, whereas Gondeila dates had significant lower total phenolic compounds that make them palatable at these stages. Moreover, **El-Rayes (2009)** found that carbon dioxide treatment-maintained fruit contents of total phenolic values significantly higher than all other CO₂ treatments. Moreover, low storage temperature-maintained fruit contents of total phenolic values significantly higher than all other used cold storage treatments. Also, during the fruit development, the conversion of Barhy date fruits from full mature stage to ripening stage caused a significant loss in total phenolics, which could be explained as due to the decomposition of natural phenolics in dates during ripening processes. As well as **Raafat *et al.* (2016)** found

that, storage period progressed, a general declining trend in all fruit quality parameters were observed for all treatments except for tomatoes treated with the essential oils, which, showing a lowest weight loss, firmness, TSS, pH, sugar fractions and antioxidant components (Vitam. C, lycopene and phenolic fractions). The obtained results suggested that essential plant oils may improve control decay of fruits, enhancement fruit shelf life and quality-related attributes on top of the well-documented antimicrobial protection during cold storage.

REFERENCES

- Abdul Rahman, A., G. Ibrahim, S. Sulaiman, M. Ahmed and K. Osma (2004). Susceptibility of some varieties of date fruits to support the production of aflatoxins: analysis by high performance liquid chromatography. *Pak. J. Biol. Sci.*, 7 (11): 1937-1941.

- Abu-Goukh, A.A., A.E. Shattir and M.M.E. Balla (2003). Effect of harvesting method on quality and storability of dry dates in Sudan. *Trop. Sci.*, 43: 53-56.
- Adato, I. and S. Gazit (1974). Water-deficit stress, ethylene production ripening in avocado fruits. *Plant Physiol.*, 53: 45-46.
- Ajay, K.M., A. Mishra, H. Kehri, B. Sharma and A.K. Pandey (2009). Inhibitory activity of Indian spice plant *Cinnamomum zeylanicum* extracts against *Alternaria solani* and *Curvularia lunata*, the pathogenic dematiaceous molds. *Ann. Clin. Microbiol. and Anti.*, 8: 1186-1199.
- Al-Ahmadi, S.S. and R. Ibrahim (2009). Application of ozone to control insect pests and molds of date fruits. *Biosci. Biotechnol. Res. Asia.*, 6: 435-446.
- Aleid, S.M., A.M. Elansari, Z.X. Tang and A.A. Sallam (2014). Effect of cold storage and packin type on khalas and sukkary dates quality. *Adv. J. Food Sci. Technol.*, 6 (5): 603-608.
- Al-Kahtani, H.A., H.M. Abu-Tarboush, Y.N. Al-Dryhim, M.A. Ahmed, A.S. Bajaber, E.A. El-Shami and M.A. El-Mojaddidi (1998). Irradiation of dates: Insect disinfestations, microbial and chemical assessments and use of thermos luminescence. *Radiat. Phys. Chem.*, 53: 181-187.
- Al-Obeed, R.S. (2010). Improving fruit quality, marketability and storability of Barhee Date palm. *World Applied Sci. J.*, 9 (6): 630-637.
- Al-Ogaidi, H.K. and H.H. Mutlak (1986). The phenolic compounds of four date cultivars during maturity stages. *Date Palm J.*, 4: 191-196.
- Al-Redhaiman, K.N. (2004). Modified atmosphere improves storage ability, controls decay, and maintains quality and antioxidant contents of Barhi date fruits. *Food Agric. and Environ.*, 2 (2): 25-32.
- Al-Yahia, S.A. (1986). Quality change of 'Barhy' dates during storage at bisr stage. *Proceedings of the Second Symposium on the Date Palm in Saudi Arabia*, March 3-6.
- Amirmijani, A.S.K. (2018). Fungal contamination associated with some dried fruits in Iran. *Novel Res. Microbiol. J.*, 2 (6): 105-113.
- Anjili, S.M., F.K. Channya and I.B. Chimbekujwo (2016). Control of Fungi Isolated from Date Palm Fruit in Yola, Adamawa State. *J. Biol., Agric. and Healthcare*, www.iiste.org, ISSN 2225-093X (Online)
- Atia, M.M.M. (2011). Efficiency of physical treatment and essential oil in controlling fungi associated with some stored date palm fruits. *Aust. J. Basic Appl. Sci.*, 5 (6) 1572: 1587.
- Atia, M.M.M., A.A.M. El-Mahmodi and M. El-Shili (2009). Fungi associated with some stored date palm fruits in Libya. *The Third National Conference on Basic Science, Under the Title: Basic Sciences are the Main Source of Creativity*, Gharian, Libya, from pp 25 - 27.
- Azelmat, K., F. Sayah, M. Mouhib, N. Ghailani and D. El-Garrouj (2005). Effect of gamma irradiation on fourth-instar *Plodia interpunctella* (Hubner) (Lepidoptera: Pyralidae). *J. Stored Prod. Res.*, 41: 423-431.
- Banks, N.H. (1984). Some effects of TAL-Prolong coating on ripening bananas. *J. Exp. Bot.*, 35: 127 - 134.
- Baudion, A.B. (1988). *Laboratory exercises in plant pathology: An Instruction Kit*. APS Press, St. Paul, MN., Pages: 213.
- Ben-Yehoshua, S. (1969). Gas exchange, transportation and the commercial deterioration in storage of orange fruit, *J. Am. Soc. for Hort. Sci.*, 94: 524 - 531.
- Bokhary, H.A. (2010). Seed-borne fungi of date palm, *Phoenix dactylifera* L. from Saudi Arabia. *Saudi J. Biol. Sci.*, 17: 327-329.
- Booth, C. (1971). *The genus Fusarium*. Commonwealth Mycological Institute, Kew Surrey, UK.
- Brown, W. (1924). A method of isolation single strains of fungi by cutting out a hyphal tip. *Ann. Bot.*, 38 : 402-404.
- Chao, C.T. and R.K. Robert (2007). The date palm (*Phoenix dactylifera* L.) overview of

- Biology, uses and cultivation. Hort. Sci., 42: 1077-1082.
- Colman, S., T.H. Spencer, P.E. Ghamba and E. Colman (2012). Isolation and identification of fungal species from dried date palm (*Phoenix dactylifera*) fruits sold in Maiduguri metropolis. Afr. J. Biotechnol., 11 (57): 12063-12066.
- Davenport, T.L. (1984). Studies on avocado fruit ripening using calcium. Proc. Fla. State Hort. Soc., 97: 329-330.
- Djerbi, M., L. Aouad, H. Filali, M. Saaidi, A. Chtioui, M.H. Sedra, M. Allaoui, T. Ham-daoui and M. Oubrich (1986). Preliminary results of selection of high quality Bayoud resistant clones among natural date palm population in Morocco. Proc. Sec. Symp. on the Date Palm. Saudi Arabia, March 3-6: 383-399.
- Domsch, K.W., W. Gams and T.H. Anderson, (1980). Compendium of Soil Fungi. Acad. Press, London.
- Dowson, V.H.W. (1982). Date production and protection with special reference to North Africa and the near East. FAO.
- Dubois, M., K.A. Gilles, J.K. Hamilton, P.A. Rebers and F. Smith (1956). Colorimetric method for determination of sugars and related substances. Anal. Chem., 28:350-356.
- El-Juhany, L. (2009). Degradation of date palm trees and date production in Arab countries causes and potential rehabilitation. Aust. J. Basic Appl. Sci., 4: 3998– 4010.
- Ellis, M.B. (1976). More Dematiaceous Hyphomycetes. Commonwealth Mycol. Inst., Kew, Surrey, UK.
- El-Rayes, D.A. (2009). Effect of carbon dioxide-enriched atmosphere during cold storage on limiting antioxidant losses and maintaining quality of 'Barhy' date fruits. JKAU: Met., Env. and Arid Land Agric. Sci., 20 (1): 3-22.
- El-Sharony, T.F. and O.A. Amin (2014). Effect of some natural substances on fruit quality of Washington navel orange under cold storage, J. Agric. and Ecolo. Res. Int., 2 (1): 58- 68.
- El-Sharony, T.F., O.A. Amin and A.S.E. Abd-Allah (2015). Effect of some post-harvest treatments on quality and storability of date palm fruits zaghoul and Samany cultivars. Egypt. Soc. Environ. Sci., 10 (1): 49-58.
- Fallik, E. (2004). Prestorage hot water treatments (immersion, rinsing and brushing). Post. Biol. and Technol., 32: 125–134.
- FAO (2016). Food and Agriculture Organization of the United Nations (FAO). *Date Palm Production*; FAOSTAT Database; FAO: Rome, Italy, 2016
- Fawzi, E.M., A.A. Khalil and A.F. Afifi, (2009). Antifungal effect of some plant extracts on *Alternaria alternata* and *Fusarium oxysporum*. Afr. J. Biotechnol., 8 (11): 2590-2597.
- Hadizadeh, I., B. Peivastegan and H. Hamzehzarghani (2009). Antifungal activity of essential oils from some medicinal plants of Iran against *Alternaria alternata*. Ame. J. Appl. Sci., 6 (5): 857-861.
- Hansen, H.N. (1926). A simple method of obtaining single spore cultures. Sci., 64: 384-389.
- Harrak, H. (2003). Activités de recherche en matière de la valorisation technologique des dattes, in Rapport d'activités Compagne 2002-2003, INRA, Centre Régional de la Recherche Agronomique de Marrakech, Maroc.
- Hasnaoui, A., M.A. Elhoumaizi, A. Asehraou and A. Hakkou (2010). Chemical composition and microbial quality of main varieties of dates grown in figuig oasis of Morocco. Int. J. Agric. Biol., 12: 311-314.
- Irshad, A., A. Muhammad, H. Faisal, Q.A. Syed and A.R. Toqeer (2016). Pathogenic fungi associated with date palm trees in Turbat, Balochistan. Int. J. Biol. Biotech., 13 (1): 33-38.
- Johnson, L.F. and E.A. Curl (1972). Methods for research on ecology of soil-borne pathogens. Burgess Publishing Co., Minneapolis, MN, USA.
- Khawla, J.A., E.E. Saeed, A. Sham, A.A. Alblooshi, M.M. Alblooshi, K.A. El-Tarabily and S.F. AbuQamar (2019). Secular

- Identification and Disease Management of Date Palm Sudden Decline Syndrome in the United Arab Emirates. *Int. J. Mol. Sci.*, 20 (4): 923; <https://doi.org/10.3390/ijms20040923>
- Lahyani, S. (1991). Conservation des dattes par traitement thermique. Mémoire pour le concours d'accès au grade d'Ingénieur en Chef, Option scientifique. INRA, Rabat, Maroc, 38.
- Maxin, P. and K. Klopp (2004). Economics of hot water dipping. In: Boos, MH. (Ed.), Proc. 11th Int. Conf. Cult. Tech. and Phytopathol. Prob. in Organic Fruit-Growing. FÖKO, Weinsberg, Germ., 75–78.
- Maxin, P. and R.W.S. Weber (2011). Control of Phacidiopyxis washing tonensis storage rot of apples by hot-water treatments without the ethylene inhibitor 1-MCP. *J. Plant Dis. and Prot.*, 118: 222–224.
- Maxin, P., N. Fieger-Metag, B. Benduhn, P. Kruse and P. Heyne (2006). Hot water dipping in Northern Germany-on farm results after four years of scientific work. In: Boos, MH (Ed.), Proc. 12th Int. Conf. Cult. Tech. and Phytopathol. Prob. Organic Fruit-Growing. FÖKO, Weinsberg, Germ., 118–120.
- Maxin, P., S. Huyskens-Keil, K. Klopp and G Ebert (2005). Control of post-harvest decay in organic grown apples by hot water treatment. *Acta Hort.*, 682: 2153–2158.
- Maxin, P., R.W.S. Weber, H.L. Pedersen and M. Williams (2012). Hot-water dipping of apples to control *Penicillium expansum*, *Neonectria galligena* and *Botrytis cinerea*: Effects of Temperature on Spore Germination and Fruit Rots. *Europ. J. Hort. Sci.*, 77 (1): 1–9
- Mironescua, M. and C. Georgescu (2008). Preliminary researches on the effect of essential oils on molds isolated from surfaces. *J. Agro. Aliment. Proc. and Technol.*, 14: 30-33.
- Moubasher, A.H. (1993): Soil fungi in Qatar and other Arab countries. The Scientific and Applied Research Center. University of Qatar, Doha, Qatar.
- Nabifarkhani, S.M., A.D. Garmakhany, E.G. Moghadam and M.A. Shakevi (2015). Effect of nano-composite and thyme oil (*Thymus vulgaris* L.) coating on fruit quality of sweet cherry (Takdaneh CV) during storage period. *Food Sci. and Nut.*, 3 (4): 349-354.
- Nagy, S. and Shaw, P.E. (1980). Tropical and Sub-Tropical Fruits. p. 507-510. AVI, Inc. West Ports, Connecticut, U.S.A.
- Olukayode, O.O, O.O. Ignesta, F. D. Victor, and A. Ohiobo, (2017). Fungal Species Associated with Date Palm (*Phoenix dactylifera* L.) Fruit and Tiger Nut (*Cyperus esculentus* L.) Fruit in Lafia Metropolis, Nasarawa State, Nigeria. *Archives of Current Research International*, 9 (4): 1-7
- Osman, A. M. A (1984). The Performance of Date Palms in the Sudan. *Acta Horticulture* 143:231-234.
- Ozkaya, O., D. Omur, C.S. Giulia and V. Giorio (2009). Evaluation of quality parameters of strawberry fruits in modified atmosphere packaging during storage. *Afr. J. Biotech.*, 8 (5): 789-793.
- Pitt, J.I. (1985): A Laboratory Guide to Common *Penicillium* species. Kew: IMI. pp 184.
- Qasem, J.R. and H.A. Abu-Blan, (1996). Fungicidal activity of some common weed extracts against different plant pathogenic fungi. *J. Phytopathol.*, 144: 157-161.
- Raafat, Shaymaa M., M.I. Abou-Zaid, M.R. Tohamy and H.E. Arisha (2016). Impact of some plant essential oil treatments on controlling cherry tomatoes spoilage, improvement shelf life and quality attributes during storage. *Zagazig J. Agric. Res.*, 43 (3): 785-813
- Rahmani, A. H., M. A. Salah, A. Habeeb, Y B. Ali, S. A. Sauda, (2014). Therapeutic effects of date fruits (*Phoenix dactylifera*) in the prevention of diseases via modulation of anti-inflammatory, anti-oxidant and anti-tumour activity. *International Journal of Clinical and Experimental Medicine*, 7(3): 483-491
- Raja, G.R., C.R. Nirmala and C.H. Ramanamma (2009). Efficacy of phytoextracts and oils of certain medicinal plants against *Cercospora*

- moricola* Cooke, instant of mulberry (*Morus alba* L.) leaf spot. J. Biopesticides, 2 (1): 77-83.
- Raper, K.B. and Fennell, D.I. (1977): The genus *Aspergillus* R.E. Krieger Publishing Company, Huntington, NY, USA.
- Rygg, G.L., J.R. Furr, R.W. Nixon, and W.W. Armstrong, (1953). Factors affecting the spoilage of dates at room temperature. Date Grower's Inst., 30: 10-13.
- Sawaya, W.M. and Mashadi, A.S. (1983). Sugars, tannins, and vitamin contents of twenty-five date cultivars grown in Saudi Arabia, Proceedings of Date Palm Symp. King Faisal Univ., pp: 468-479.
- Shattir, A. E., A. A. Abu-Goukh, and K. M. Karam (2002). Physical and chemical characteristics and yield components of "Barakawi" and "Gondeila" dry dates. Sudan J. Sci. Res., 8 - (1): 119-131.
- Singh, R.Y., S. Kumar and A. Dikshit, (2006). Antifungal properties of essential oil of *Mentha spicata* L. var. MSS-5 Ramesh Singh Yadav. Indian J. Crop Sci., 1(1-2): 197-200.
- Slinkard, K. and V. L. Singleton, (1977). Total phenol analyses: automation and comparison with manual methods. Amer. J. Enol. Vitic., 28: 49-55.
- Snedecor, G.W. and W.G. Cochran, (1982). Statistical Methods. 7th Ed. Iowa State University, Pres, USA.
- Suwitchayanon, P. and K. Kunasakdakul, (2009). *In vitro* effects of clove and turmeric extracts controlling crucifer pathogens. J. Agric. Technol., 5(1): 193-199.
- Tabaestani, H.S., N. Sedaghat, E.S. Pooya and A. Alipour (2013). Shelf life improvement and post-harvest quality of cherry tomato (*Solanum lycopersium* L.) fruit using basil mucilage edible coating and cumin essential oil. Int. J. Agron. and Pl. Prod., 4 (9): 2346-2353.
- Taira, S. (1996) Astringency in persimmon. In: Fruit Analysis, In: Modern Methods of Plant Analysis, Linskens, H.P., Jack-son, J.F. (Ed.), 18. Springer-Verlag, Berlin Heidelberg, 97-110. Technical Bulletin No. 35. 294.
- Thompson, K.A. and A.H. Abboodi (2003). Modified Atmosphere Packaging, Proceedings of the International Conference on Date Palm. King Saud Univ., Qassim, Kingdom of Saudi Arabia, 363-394.
- Tian, J., X. Ban, H. Zeng, B. Huang, J. He and Y. Wang (2011). *In vitro* and *in vivo* activity of essential oil from dill (*Anthem grave lens* L.) against fungal spoilage of cherry tomatoes. Food Control, 22: 1992- 1999.
- Tirmazi, S. I. H. and R. B. H. Wills. (1981). Retardation of ripening of mangoes by post-harvest application of calcium. Trop. Agric., 58: 137-141.
- Tsuda, T., K. Chachin and Y. Ueda (1999). Studies on keeping capacity of imported Carabo mango fruit from the Philippines. J. Jap. Soc. Hort. Sci., 69 (3): 669-674.
- Vesaltalab, Z. and M. Gholami (2012). Effect of essence and extract of *Eugenia caryophyllata* on some qualitative characters of grape during storage period. Irani. J. Hort., 43: 255-265.
- Viuda, M.M., Y.N. Ruiz, J.L. Fernandez and J.A. Perez, (2007). Antifungal activities of thyme, clove and oregano essential oils. Journal of Food Safety, 27(1): 91-101.

تأثير بعض الزيوت النباتية على فساد التمر السيوي المتسبب عن فطر بوتريتس سنريا تحت ظروف التخزين البارد

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تعتبر ثمار البلح من أهم المنتجات التخزينية، ذات الأهمية الاقتصادية والغذائية والاجتماعية الكبيرة بالنسبة للدول العربية، وتعد الفطريات من أهم مصادر أعفان الثمار والتي تقلل من أهميتها الاقتصادية، وقد تم عزل وتعريف عدد سبع أنواع تنتمي لسبعة أجناس فطرية من ثمار التمر السيوي والتي تم الحصول عليها من واحة سيوة، وقت حصاد المحصول مباشرة، وذلك باستخدام طريقة التخفيف، وقد كان أكثر الفطريات المعزولة تكراراً هو من جنس البوتريتس، البنسليوم، الاسبرجلس، الالترناريا، الفيوزاريوم وأقلها تكراراً هو تريكودرما يليه ريزوبس، وقد أدى غسيل التمر السيوي بالماء المعقم إلى تقليل الحمل الميكروبي للفطريات المصاحبة للثمار، مقارنة بالعزل مباشرة بعد الجمع، أدت البسترة على درجه حرارة ٨٠م إلى منع تواجد الفطريات المصاحبة للثمار، تلاها البسترة على درجه حرارة ٧٠م، وكان فطر البوتريتس سنريا هو أكثر الفطريات المسببة لأعفان ثمار التمر تكراراً خلال التخزين البارد للتمر السيوي خلال ٦ أشهر، حيث أظهر نسبة إصابة عالية، وكانت اقل نسبة إصابة خلال الشهر الأول، في حين كانت أعلى نسبة أصابه بعد الشهر السادس، في الدراسة العملية، أدى زيت الجوزبيل والقرفة، يليه زيت القرنفل بتركيز ٥% الي خفض معنوي في نمو فطر البوتريتس سنريا في حين أن تركيز ١٠% و ١٥% أدى إلى منع نمو الفطر تماماً، وقد تلاهم في التأثير زيت السمسم والشمر، أدت معاملة غمر ثمار التمر في زيت القرفة يليه الجوزبيل ثم القرنفل إلى خفض الإصابة وتقليل تعداد فطر البوتريتس المصاحب للثمار السليمة والمعداه، أدت المعاملة بالزيوت إلى تحسين الخواص الشكلية مثل الصلابة وزيادة نسبة المواد الصلبة الذائبة الكلية وزيادة السكريات وتقليل المحتوي من التانينات والفينولات في ثمار التمر السيوي السليمة والمعداه بالفطر بوتريتس خلال مده ٦ أشهر من التخزين البارد.

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