

## Exploiting Egyptian dates waste extract as a preservative to improve the quality and safety of chilled chickens

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### Abstract

Egypt is one of the richest countries in the world producing *Phoenix dactylifera* fruit. *Phoenix dactylifera* fruit wastes are not yet extensively utilized in the industry. Chicken safety and quality are prerequisites for current meat production and a basic requirement for consumers. Therefore, this study was carried out to evaluate the effect of 2% date seed extract on the safety and quality of the chicken meat marketed in Port-Saied markets, Egypt using chemical and bacteriological parameters. A total of forty chicken carcasses were equally divided into two groups. The first group was treated by dipping the carcasses in 2% date seeds extract for 15 mins, while the second group was kept as a control. Chicken samples were evaluated after one hour of treatment, as on the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> days of storage at 3±1°C. The obtained results revealed that pH, total volatile nitrogen (mg/100 g), and Thiobarbituric Acid reactive substances (mg-MDA/kg) values of treated samples were 6.4, 13.6 and 0.303 respectively on the 9<sup>th</sup> day of chilled storage. The treatment of chicken samples with 2% date seeds extract showed significantly ( $p < 0.05$ ) lower total volatile nitrogen and Thiobarbituric Acid reactive substances values, as compared with control ones due to their content of antioxidants and phenolic compounds. Additionally, treatment with 2% date seed extracts significantly reduced the aerobic bacterial counts and extended the shelf-life of stored chicken meat to the 9<sup>th</sup> day of storage. Furthermore, using 2% date seed extract effectively improved the safety of the chicken meat as *Salmonella* and *Campylobacter* spp. were not detected in treated samples. It could be concluded from this study that additional studies are recommended to investigate the nature of the preservative materials in dates seeds extract to expand its use on an industrial scale to ensure the safety and quality of chilled chicken meat.

### 1. Introduction

Egypt is one of the richest countries in *Phoenix dactylifera* fruit production in the world (Bekheet and El-Sharabasy, 2015). The date production industry of Egypt represents 20% of the entire world production (FAO, 2009). This information contributed to the motivation of food hygiene researchers to make optimal use of the huge dates by-products produced in various fields of industry, including food safety, as they may contain natural preservatives.

Chicken meat is considered a good source of high biological value protein, considerable price, delicious taste, and a low level of fat content when compared with other types of meats (Ahmad *et al.*, 2018). In addition, the consumption of chicken meat as part of a vegetable-rich diet is linked to reducing the risk of developing obesity, cardiovascular diseases, and type 2 diabetes

mellitus (Marangoni *et al.*, 2015). On the other hand, chicken meat is considered a highly perishable food with a limited shelf-life even when stored at chilling temperatures (Mercier, 2019).

Processed raw chicken meats naturally carry bacteria, most of that are square measure chargeable for spoilage and deterioration. Chemical evaluation of chicken meat is useful for determining their quality such as pH values (Hathout and Aly, 2010), total volatile nitrogen (Urmila *et al.*, 2015) and thiobarbituric acid values (Nawar, 1996).

The importance of chicken meat as vectors of food-borne pathogens is essential to public health and a challenge to food safety approaches (Alvarez-Astorga *et al.*, 2003). Thus, chicken meat can harbour a large number of pathogens such as *Salmonella* spp., *Campylobacter* spp. and *Escherichia coli* (Del-Río *et al.*,

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2007). Moreover, an increase in bacterial contamination may be added from poor personal hygiene in retail shops and cross-contamination (Nossair *et al.*, 2015). Food-borne pathogens are the main cause of illnesses and deaths in most countries, affecting approximately 1.8 million people annually (Faruque, 2012).

Herbs have been utilized in meat for a long time as flavouring agents and food preservatives due to their antimicrobial activity against pathogens (Mohanka and Priyanka, 2014). Date seeds were utilized in several old-fashioned medicines for their potential health benefits (Orabia and Shawky, 2014). Date seeds extract contains various antioxidants and antimicrobial agents such as ascorbic acid, phenolics, flavonoids, chlorophyll and carotenoids found in different values (Frag *et al.*, 2014). Recently, Sayas-Barberá *et al.* (2020) used various concentrations of date pit for enhancing the safety and quality of beef burgers. They confirmed that the date pit was able to improve the shelf-life of the burgers during the storage time, due to their antioxidant activity and phytochemical content.

The existence of spoilage or pathogenic microorganisms in chicken meat is harmful but inescapable. Therefore, the study was aimed to investigate the quality and safety of broiler carcasses treated with 2% date seed extract under chilling storage  $3\pm 1^{\circ}\text{C}$  using chemical indicators, i.e. pH, total volatile nitrogen value and thiobarbituric acid reactive substances value. Also, bacteriological determination of aerobic colony counts and *Escherichia coli* counts, as well as detection of *Salmonella* and *Campylobacter* spp. were conducted.

## 2. Materials and methods

### 2.1 Preparation of date seed extract

The fresh date stage of the date palm (*Phoenix dactylifera* L. var *Barhee*, Figure 1) was procured from the local fruit markets in Port-Said province, Egypt, from July to September 2020. Seeds (Figure 2) were manually removed from the fruit's flesh, washed with distilled water and dried in the shade for one week in the open air (Jaganathan *et al.*, 2018). The dried pits were crushed, and ground using a grinder (Knifetec 1095 laboratory mill) at 10,000 rpm to make fine powder and stored in air sealed, labelled polythene bags at  $4^{\circ}\text{C}$  until extraction was carried out.



Figure 1. date palm, *Phoenix dactylifera* L. var *Barhee*



Figure 2. Seeds of date palm

The palm date seeds ethanolic extract was prepared as described by Alugaili *et al.* (2019). Whereby, 450 g of palm date seeds powder was macerated in 2000 mL of 99.9% ethanol in a flask and was placed on a magnetic stirrer for 72 hrs at ambient temperature, then was filtered through a filter paper (Whatman No.1). The solution was then evaporated to dryness utilizing a rotary evaporator (R-100 Buchi – Switzerland) at  $40^{\circ}\text{C}$ . The yield after complete evaporation of ethanol was 33 g of date seeds extract stored at  $4^{\circ}\text{C}$  until further use. The dipping solutions with a concentration of 20 g/L were intended by dissolving date seeds extract in sterile distilled water.

### 2.2 Determination of the antimicrobial activity of the prepared extract

The antimicrobial activity determined by the agar well diffusion method according to NCCLS (1993), Muller-Hinton agar medium (Hi Media, M173) were plated by streaking with 24-hour old cultures of the bacterial strains such as *Staphylococcus aureus* ATCC25923 and *E. coli* ATCC25922 ( $10^5$  CFU/mL). Wells of 6 mm in diameter was made in the Mueller Hinton agar by using the opposite side of micropipette tips, then 20  $\mu\text{L}$  of different concentrations of extracts (1%, 2% and 3%) were pipetted into each well (Figure 3). The plates were incubated at  $4^{\circ}\text{C}$  for 2 hrs to permit diffusion of the active components in the medium (Tagg and Mcgiven, 1971). The standard Levofloxacin 5 mcg/disk (Oxoid, UK) was used as a positive control, the distilled water was used as a negative control. The inoculated plates were incubated at  $37^{\circ}\text{C}$  for 24 hrs. The diameters of the inhibition zones in mm were measured for each plate and the average reading of the triplicates for each antibacterial species was calculated.



Figure 3. Inhibition zones by different concentrations of DSE

### 2.3 Experimental trials

A total of 40 broilers were randomly collected from

private small-scale manual poultry processing shops with an average weight of 1000-1200 g, then divided into two groups. The first group was treated by dipping the chicken carcasses for 15 mins in 2% date seeds extract. While the second group was kept without treatment as a control. After dipping, the chicken carcasses were aseptically drained and separately packed in sterile polyethylene packs for chemical and bacteriological evaluation after one hour of treatment and on the 3<sup>rd</sup>, 6<sup>th</sup> and 9<sup>th</sup> days of storage at 3±1°C.

## 2.4 Chemical evaluation

### 2.4.1 Determination of pH

A total of 10 g of chicken meat was homogenized with 90 mL of distilled water at 25°C. The homogenate was left at room temperature for 10 mins with continuous shaking. The pH value was determined using a digital pH meter (Mettler Toledo, Switzerland), calibrated at pH buffers (4.00 - 7.00 - 10.00) (ISO, 1999).

### 2.4.2 Determination of total volatile basic nitrogen

Ten grams of chicken meat were added to the heating flask containing 300 mL distilled water plus 2 g magnesium oxide (Fisher Chemical, M300-500) and an antifoaming agent. In the receiving flask 25 mL of boric acid (2%) (Fisher Chemical, B/P560/60), and a few drops of methyl red indicator were added. The two flasks (heating and receiving) were linked to the evaporator and the water bath was managed. After 25 mins, distillation was stopped. The content of the receiving flask was moved to another flask and titrated to the end-point by 0.1 N H<sub>2</sub>SO<sub>4</sub> (Fisher Chemical, J/8460/15). The % total volatile nitrogen was determined as followed:

TVN (mg/100 g of chicken meat) = (volume (mL) of H<sub>2</sub>SO<sub>4</sub> used for titration of the distilled - volume (mL) of H<sub>2</sub>SO<sub>4</sub> used in control sample) × 14 (AOAC, 1990).

### 2.4.3 Determination of thiobarbituric Acid reactive substances values

For extraction, 10 g of minced chicken meat mixed with 50 mL of distilled water was added to 47.5 mL of distilled water and 2.5 mL of hydrochloric acid 4 N (Fisher Chemical, H/1150/PB17) to bring the pH to 1.5. Few glass beads were added as an antifoaming agent into a distillation flask. The flask containing the mixture was heated for 10 mins from the time boiling commences collecting 50 mL of distillate. Five mL of distillate was collected into tubes with the stopper, and 5 mL of TBA reagent (0.2883 g of 2-Thiobarbaturic acid (Merck, MC1081800025)/100 mL of 90% glacial acetic acid (Fisher Chemical, A/0360/PB15) was added to them. Then the tubes were closed with stoppers, shaken and

heated in boiling water for 35 mins. Afterwards, the tubes were cooled in water for 10 mins. Eventually, the absorbance against the blank (5 mL distilled water and 5 mL TBA reagent) was measured at a wavelength of 538 nm using one cm cells in a spectrophotometer (Specord 250, Germany) (Pearson, 1984).

TBRS No. = Power of light absorption × 7.8 (results expressed as mg malonaldehyde per kg of chicken meat sample).

## 2.5 Bacteriological evaluation

### 2.5.1 Samples preparation

The technique recommended by USDA/FSIS (1996), after determining the weight, each carcass was rinsed with 400 mL of 0.1% sterile buffered peptone water, and allowed to drain for 15 s. An aliquot (1 mL) of rinse fluid was aseptically transferred to a test tube containing 9 mL sterile buffered peptone water 0.1% to prepare tenth-fold serial dilutions up to 10<sup>-8</sup> for determining aerobic colony count and *E. coli* count.

From the aforementioned rinsed fluid, 25 mL was pre-enriched in 225 mL of sterile buffered peptone water then incubated for 18±2 hrs at 37°C for the detection of *Salmonella* spp., the same technique was used for the detection of *Campylobacter* spp. using Preston selective enrichment broth under microaerobic conditions.

### 2.5.2 Determination of aerobic colony count

One mL of each prepared serial dilution was separately inoculated into sterile duplicate Petri dishes. About 12 to 15 mL of tempered melted standard plate count agar (Oxoid-CM0463) was cooled to 44-47°C and was added to each plate. Afterwards, the plates were thoroughly and uniformly mixed with the inoculum and left to solidify. After solidification, the inoculated plates were incubated at 30±1°C for 72±3 hrs (ISO, 2013). Results were expressed as colony-forming units (CFU/mL), then was converted to CFU/cm<sup>2</sup> by using the formula in FSIS/USDA microbial baseline survey (USDA, 1996).

Total colony-forming units (CFU) of bacteria/Total Surface Area (cm<sup>2</sup>) = CFU/mL recovered × mL used to rinse the carcass / (0.87 × w) + 635, Where “w” is the weight of the carcass in grams.

### 2.5.3 Determination of *Escherichia coli* count

An aliquot (1 mL) of each prepared serial dilution was separately inoculated into sterile duplicate Petri dishes. Approximately 15 mL of tempered melted Tryptone Bile X glucuronide Agar (TBX Medium) (Oxoid, CM0945) previously cooled at 44°C to 47°C in the water bath were added to each inoculated plate.

Immediately after pouring, they were thoroughly and uniformly mixed with the inoculum and allowed to solidify by leaving the Petri dishes on a cool horizontal surface. After solidification, plates were incubated at 37°C for 4 hrs, then the incubation was continued at 44°C for 18-24 hrs (ISO, 2001).

#### 2.5.4 Detection of *Salmonella* spp.

According to ISO (2017a), the pre-enrichment culture was transferred to 10 mL of Rappaport Vassiliadis broth (Oxoid-CM0669). The inoculated tubes were incubated at 41.5±1°C for 24 ±3 hrs, another 1 mL of pre-enrichment culture was moved to a tube containing 10 mL of Muller-Kauffmann tetrathionate/novobiocin broth (MKTTn) (Oxoid-CM1048) and incubated at 37±1°C for 24±3 hrs. Then, a loopful (10 µL) from each of the RV and MKTTn broths was streaked on Hektoen enteric agar (Oxoid-CM0419) and Xylose lysine desoxycholate agar (Oxoid-CM0469). Suspected colonies were exposed to biochemical and serological confirmation.

#### 2.5.5 Detection of *Campylobacter* spp.

According to ISO (2017b), a loopful of Preston broth was streaked on modified Charcoal Cefoperozone Deoxycholate agar (Hi Media, M887I), then incubated in a microaerophilic condition at 41.5°C for 44±4 hrs. A typical colony of *Campylobacter* spp. on mCCDA agar is greyish with a metallic sheen, flat and moist with a tendency to spread. The well-isolated freshly grown colonies were used for examination of morphology and motility, absence of aerobic growth at 25°C, and presence of oxidase activity.

#### 2.6 Statistical analysis

Statistical analysis of the data was performed using Statistical Package for the Social Sciences (SPSS) version 16.0 (SPSS, Inc., Chicago, IL, USA). Data were analysed for the significant differences between control and treated samples. Data were subjected to independent samples T-test and Mann-Whitney U test. The differences in means between control and treatment were carried out using Duncan's multiple range test at 0.05 level.

### 3. Results and discussion

#### 3.1 Determination of the antibacterial activity of the date seeds extract

The result obtained from Table 1 revealed that there was a statistically significant difference ( $p < 0.05$ ) between the inhibitory zones of the various concentrations of date seed extract (DSE). The inhibition

zones (mm) by palm date seeds extract against *Staphylococcus aureus* were 11.6, 16 and 17 mm at concentration levels 1%, 2% and 3 %, respectively. Relatively similar results were recorded by Radfar *et al.* (2019), while our results of 2 and 3% concentrations were higher than those found by Ammar *et al.* (2009), Al-daihan and Bhat (2012), Bentradi *et al.* (2017) and Jaganathan *et al.* (2018). The obtained results were lower than the results reported by Shakiba *et al.* (2011), Perveen *et al.* (2012) and Alrajhi *et al.* (2019).

Table 1. Sensitivity test of palm date seed extract against *Staphylococcus aureus*

Treatment	Conc. Level (%)	Inhibition zone (mm)
Palm date seed extract	1%	11.6 <sup>d</sup>
	2%	16 <sup>c</sup>
	3%	17 <sup>b</sup>
Levofloxacin	5 <sub>mcg</sub> /disc	26 <sup>a</sup>

Levofloxacin was considered a control positive. Values are presented as means. Values with different superscripts are statistically significant ( $p < 0.05$ ) by one-way ANOVA, tracked by Duncan's Multiple Range Tests (DMRTs).

The results showed in Table 2 revealed that there was a significant difference ( $p < 0.05$ ) between the inhibitory zones at concentrations of 1%, 2% and 3% of DSE. There were non-significant differences ( $p > 0.05$ ) between 2% and 3% of date seeds extract. Whereas the inhibitory zones (mm) of DSE against *Escherichia coli* at 1%, 2% and 3% concentration levels were 8, 15 and 15 mm, respectively. The results of 2% and 3% concentration of DSE were nearly similar to those reported by Yassein (2012), Ado *et al.* (2017), Idris *et al.* (2017) and Sundar *et al.* (2017), while the obtained results were higher than those reported by Ammar *et al.* (2009), Saddiq and Bawazir (2010), Al-daihan and Bhat (2012), Saleh (2016) and Alrajhi *et al.* (2019). Also, these results were lower than those recorded by Shakiba *et al.* (2011), Perveen *et al.* (2012) and Metoui *et al.* (2019). On the other hand, these results disagreed with results found by Radfar *et al.* (2019).

Table 2. Sensitivity test of palm date seed extract against *Escherichia coli*

Treatment	Conc. Level (%)	Inhibition zone (mm)
Palm date seed extract	1%	8 <sup>c</sup>
	2%	15 <sup>b</sup>
	3%	15 <sup>b</sup>
Levofloxacin	5 <sub>mcg</sub> /disc	28 <sup>a</sup>

Levofloxacin was considered a control positive. Values are presented as means. Values with different superscripts are statistically significant ( $p < 0.05$ ) by one-way ANOVA, tracked by Duncan's Multiple Range Tests (DMRTs).

#### 3.2 Effect of 2% DSE on chemical evaluation

##### 3.2.1 pH

The results displayed in Table 3 showed the pH

Table 3. Mean values of pH, TVN and TBRS for chicken carcasses (n = 40) treated with 2% DSE during storage at 3±1°C.

Storage	Parameters					
	pH		TVN (mg/100 g)		TBRS (mg-MDA/kg)	
	Control	2% DSE	Control	2% DSE	Control	2% DSE
After one hour	6.0±0.10	5.9±0.10	10.5±0.40	6.4±0.70*	0.168±0.008	0.031±0.01*
3 <sup>rd</sup> day	6.5±0.15	6.2±0.15	12.7±0.78	8.0±1.00*	0.514±0.050	0.122±0.02*
6 <sup>th</sup> day	6.7±0.10	6.3±0.10	15.1±0.83	9.2±0.44*	0.762±0.070	0.204±0.01*
9 <sup>th</sup> day	7.1±0.10	6.4±0.06*	22.0±2.00	13.6±0.70*	1.0±0.1000	0.303±0.06*

\*Significant difference at  $p < 0.05$  between control and treated chicken samples at end of the storage period by independent samples T-test.

values of the treated chicken samples with 2% DSE compared with control samples after one hour, as well as on the 3<sup>rd</sup>, 6<sup>th</sup> and 9<sup>th</sup> day of the chilling period. Whereas the pH values of control chicken carcasses samples were 6, 6.5, 6.7 and 7.1, respectively. While the pH values of treated samples were 5.9, 6.2, 6.3 and 6.4, respectively. The pH values of control and treated chicken carcasses samples on the first day were nearly similar, and this was agreed with Sayas-Barberá *et al.* (2020). Moreover, the pH values of control and treated samples increased during the storage period, because of the accumulation of ammonia as the end product of amino acid decomposition, and this was compatible with Jay (1972). Also, the high pH value of control samples was characterized by the dark colour, this agreed with Pearson and Gillette (1996). It was noticeable from the current results that the pH values of the control samples recorded 7.1 on the 9<sup>th</sup> day of chilling at 3±1°C, while the pH value of treated samples did not increase over 6.4 until the 9<sup>th</sup> day of storage. This might be attributed to the inhibitory effect of antimicrobial ingredients presented in DSE, which limited the growth and proliferation of spoilage microorganisms that utilize the basic nitrogen compounds.

### 3.2.2 Total Volatile Basic Nitrogenous

Total Volatile Nitrogen (TVN) values (mg/100 g) in control chicken carcasses samples after one hour, and on the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> days of storage were 10.5, 12.7, 15.1 and 22.0, respectively. While TVN values in treated samples were 6.4, 8, 9.2 and 13.6, respectively. Total volatile nitrogen values of control and treated samples gradually increased through the storage period. Moreover, TVN values of control exceeded the permissible limit (20 mg/100 g) recommended by EOS (2005) on the 9<sup>th</sup> day of storage (Table 3). This could be due to the presence of microbial contamination or enzymatic degradation, which is in agreement with Urmila *et al.* (2015). While the treated chicken samples with 2% date seed extract showed significantly ( $p < 0.05$ ) lower TVN values than control samples throughout the storage period samples. These results were compatible with Mohammadi *et al.* (2018), which could be explained by the antioxidant activity of date seed extract,

which prohibited lipid peroxidation and putrefaction of amino acids.

### 3.2.3 Thiobarbituric acid reactive substances

The treatment with 2% DSE significantly ( $p < 0.05$ ) decreased the TBRS value in treated chicken samples as compared with control ones throughout the chilling storage period (Table 3). Whereas Thiobarbituric Acid reactive substances (mg-MDA/kg) in control chicken carcasses samples after one hour, on the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> days of storage were 0.168, 0.514, 0.762 and 1, respectively. While the values of TBRS in treated samples were 0.031, 0.122, 0.204 and 0.303, respectively. Thiobarbituric Acid reactive substances (TBRS) values of control and treated samples increased during the chilling storage period. While TBRS values of control exceeded the acceptable limit (0.9 mg malondialdehyde/kg) recommended by EOS (2005), on the 9<sup>th</sup> day of storage at 3±1°C, because of the oxidation of unsaturated fatty acids and formation of lipid hydroperoxide which reflected the spoilage and degradation in products, and these findings were compatible with Nawar (1996).

In contrast, TBRS values of treated chicken samples with 2% DSE remained within the permissible limit because date seed extract may contain phenolic groups which acted as antioxidants and delayed lipid oxidation during the storage. Nearly similar results were attained by Essa and Elsebaie (2018), and Sayas-Barberá *et al.* (2020).

## 3.3 Effect of 2% DSE on bacteriological parameters

### 3.3.1 Aerobic colony counts

Results illustrated in Table 4 exhibited that the treated chicken samples with 2% DSE significantly ( $p < 0.05$ ) decreased aerobic colony count (CFU/cm<sup>2</sup>) throughout the storage period at 3±1°C, where the mean value of aerobic colony counts of control chicken carcasses samples after one hour and on the 3<sup>rd</sup> day at 3±1°C was  $6.7 \times 10^4$ , and  $8.3 \times 10^5$  CFU/cm<sup>2</sup>, respectively. While, treated samples results after one hour, as well as on the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> day at 3±1°C were  $2.2 \times 10^4$ ,  $5.3 \times 10^4$ ,  $3.7 \times 10^5$ , and  $7.8 \times 10^5$  CFU/cm<sup>2</sup>, respectively.

Table 4. Statistical analytical results of aerobic colony counts and *Escherichia coli* counts (CFU/cm<sup>2</sup>) in chicken carcasses (n = 40) treated with 2% date seed extract during storage at 3 ± 1°C.

Storage period	Aerobic colony counts (CFU/cm <sup>2</sup> )		<i>Escherichia coli</i> counts (CFU/cm <sup>2</sup> )	
	Control	2% DSE	Control	2% DSE
After one hour	6.7×10 <sup>4</sup> ±1.4×10 <sup>4</sup>	2.2×10 <sup>4</sup> ±4.2×10 <sup>3</sup> *	3.9×10 <sup>2</sup> ±9.5×10 <sup>1</sup>	1.1×10 <sup>2</sup> ±0.25×10 <sup>2</sup>
3 <sup>rd</sup> day	8.3×10 <sup>5</sup> ±2.1×10 <sup>5</sup>	5.3×10 <sup>4</sup> ±7×10 <sup>3</sup> *	8.3×10 <sup>2</sup> ±1.7×10 <sup>2</sup>	3.2×10 <sup>2</sup> ±0.72×10 <sup>2</sup>
6 <sup>th</sup> day	Rejected	3.7×10 <sup>5</sup> ±7×10 <sup>4</sup> *	Rejected	5.8×10 <sup>2</sup> ±0.8×10 <sup>2</sup>
9 <sup>th</sup> day	Rejected	7.8×10 <sup>5</sup> ±1.2×10 <sup>5</sup> *	Rejected	8.6×10 <sup>2</sup> ±0.96×10 <sup>2</sup>

\*Significant difference at p < 0.05 between control and treated chicken samples at end of the storage period by independent samples T-test.

It was clear from the results that aerobic colony counts of control samples were higher than corresponding counts of treated samples. Also, aerobic colony counts progressively increased during refrigeration in all treated samples but were still lower than control samples. In addition, control samples exceeded the acceptable limit (10<sup>5</sup> CFU/g) stated by EOS (2005) on the 6<sup>th</sup> and 9<sup>th</sup> day of chilling storage. The obtained results agree with those reported by Sundar *et al.* (2017), Jaganathan *et al.* (2018), Alrajhi *et al.* (2019), Metoui *et al.* (2019) and Sayas-Barberá *et al.* (2020).

### 3.3.2 *Escherichia coli* counts

The obtained results in Table 4 exhibited that the mean values of *E. coli* count in control chicken carcasses samples after one hour and on the 3<sup>rd</sup> at 3±1°C were 3.9×10<sup>2</sup>, and 8.3×10<sup>2</sup> CFU/cm<sup>2</sup>, respectively. While, the counts in treated samples subsequently one hour, as well as on the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> day at 3±1°C were 1.1×10<sup>2</sup>, 3.2×10<sup>2</sup>, 5.8×10<sup>2</sup>, and 8.6×10<sup>2</sup> CFU/cm<sup>2</sup>, respectively. Furthermore, it appeared from the previous results that *Escherichia coli* counts of control samples were higher than treated samples with 2% DSE. This result could be explained by what was stated by Shrestha *et al.* (2012), as the presence of hydroxyl group of the phenolic compound has a suppressed impact on the growth of *E. coli*. Although *Escherichia coli* counts progressively increased during refrigeration in all treated samples, they were lower than control samples. Moreover, control samples surpassed the allowable limit (10<sup>2</sup> CFU /gm) stated by EOS (2005) on the 6<sup>th</sup> day of storage. The obtained results were compatible with those reported by Sundar *et al.* (2017), Alrajhi *et al.* (2019) and Metoui *et al.* (2019). On the other hand, these results disagreed with Radfar *et al.* (2019), which could be attributed to using different species of date seed palm with 1% concentration.

### 3.3.3 Detection of *Salmonella*

Results of the incidence rates of *Salmonella* spp. in control and treated samples subsequently one hour was 60 and 0%, respectively. Results in Table 5 revealed the absence of *Salmonella* spp. in the treated samples. These

results agree with those reported by Shakiba *et al.* (2011) and Metoui *et al.* (2019).

### 3.3.4 Detection of *Campylobacter*

The incidence rates of *Campylobacter* spp. in control and treated samples after one hour were 100 and 0%, respectively. The obtained results could be explained by biologically active components of the date seeds including alkaloids, tannins, phenols, phenolic acids, anthocyanins, carotenoids, sterols, and flavonoids which acted as antibacterial agents. These findings are compatible with Roller (2003), Shahidi and Naczka (2004), Alfarsi *et al.* (2007), Farag *et al.* (2014) and Jaganathan *et al.* (2018).

Table 5. Effect of 2% DSE on *Salmonella* and *Campylobacter* spp. in chicken carcasses (n = 5 of each) after one hour.

Incidence / storage period	<i>Salmonella</i> spp.		<i>Campylobacter</i> spp.	
	Control	2% DSE	Control	2% DSE
After one hour	3 (60%)	0 (0.00%)	5 (100%)	0 (0.00%)

## 4. Conclusion

The broiler chicken meat is highly perishable food that is spoiled during prolonged refrigeration storage. Dipping of chicken carcasses in 2% DSE for 15 mins improved the chemical and bacteriological quality and extended the shelf-life of chicken carcasses up to 9 days during chilling storage. Using 2% date seed extract effectively improved the safety of the chicken meat. Additional studies are recommended to investigate the nature of the preservative materials in dates seeds extract in order to expand its application on an industrial scale to ensure the safety and quality of chilled meat chicken.

## Conflict of interest

The authors declare no conflict of interest.

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