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The UV-C lowered natural moth infestation of date palm throughout commercial life

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SUMMARY

Some methods commonly used to avoid postharvest pest diseases and microbial growth in dates have shown harmful effects on humans and/or on the environment and their use will be restricted or even forbidden in a short delay. However little information is available about sustainable sanitation techniques to maintain overall quality and safety of dates throughout commercial life. This work study the effects of 6 KJ m⁻² UV-C, on natural infestation by moth of pyrale and on overall quality (including safety) of DegletNour dates stored for 30 days at 20°C. As control, 100 mg L⁻¹ NaClO and untreated samples were used. The skin color, pH, titratable acidity, sugars content microbial counts, sensory quality and moth infestations were monitored. Both sanitizing treatments significantly lowered moth infestation and microbial counts without inducing any detrimental effects on overall quality of the dates. After shelf life results reached with the UV-C were considerably better than with NaClO. In addition this sustainable sanitizer treatment could be implemented without excessive costs in existing disinfection facilities for handling and shipping dates.

Keywords: DegletNour cv., pyrale, storage period, quality attributes, weight loss, NaClO, safety.

INTRODUCTION

The date palm (*Phoenix dactylifera* L.) (2n = 36) is a monocotyledon, dioecious and perennial plant. It is a fruit tree and the only cultivated species of the genus Phoenix family Areaceae (Munier, 1973). DegletNour is the most produced date palm cv. in Tunisia, being its cultivation, picking and handling the major activity of people in the Southern areas of the country. In the 2012/2013 season dates production was 192,850 t of which more than 70% were of DegletNour cv. (Gifruits, 2013). The most common biological attack of dates in Tunisia and Algeria is by the moth of pyrale (*Ectomyeloisceratoniae* Zeller). In Tunisia, the *E. ceratoniae* infests 20% of the harvestable crop annually being the major insect pest of dates both in field and in storage. It degrades the stored dates and causes weight loss downgrading the commercial value of the fruit (Haouel et al., 2010).

In order to avoid the postharvest pest diseases and microbial attacks of dates, a number of methods have been developed including fumigants (methyl bromide -CH₃Br-, phosphine -PH₃-, sulfur dioxide -SO₂-, sulfate carbon -CS₂-, phostoxin, carbon dioxide -CO₂-, ethylene oxide mixed with carbon dioxide), vacuum storage, and application of microwave, chlorine -NaClO-, ozone -O₃-, UV-C radiation, heat treatment, freezing or irradiation (Zouba, 2009; Dehghan-Shoar et al., 2010; Haouel et al., 2010; Ben-Lalli et al., 2013). Currently CH₃Br is the most widely used fumigant as quarantine treatment against *E. ceratoniae* and other insects on stored dates. Even though due to be harmful on human health as well as for the environment its use is scheduled for worldwide withdrawal application in 2015 under the Montreal Protocol of the United Nations Environment Program on ozone-depleting substances (UNEP, 1995; Bell, 2000). Due to this, the search of commercial alternatives is urgently required.

Among different antimicrobials NaClO is considered as a potent sanitizing with powerful oxidizing properties, being generally effective, comparatively inexpensive, and the most used by the food industry for sanitizing both products and equipment. The NaClO in water increases pH and generates HOCl, which is the active disinfectant, being more efficient at a pH range of 6.5–7.5. However, NaClO may incompletely oxidize organic food constituents to produce toxic byproducts in process

water, such as chloroform, haloacetic acids or other trihalomethanes. Due to this, in some European countries like Germany, The Netherlands, Denmark, Switzerland and Belgium, the use of NaClO in plant products is currently forbidden, and alternative sanitizers for the food industry must be found (Artés et al., 2009; Betts and Everis, 2005).

An efficient physical technique for surface sanitizing treatment of fruit and vegetables is the non-ionizing, artificial ultraviolet radiation at a wavelength of 190–280 nm (UV-C). In the range of 240–260 nm has been approved in the USA to be used in food (USDA/FDA, 2002). In addition, it delays senescence and extends shelf-life. The UV-C affects several physiological processes in plant tissues and damages DNA at doses from 0.5 to 20 kJ m⁻², of some microorganisms affecting their multiplication (Nakajima et al., 2004) inhibiting their growth (Allende and Artés, 2003; Pan and Zu, 2012). The UV-C treatment offers as main advantages that it is sustainable not leaving any residue, lethal to most types of microorganisms and easy to use, does not have legal restrictions, and does not require extensive safety equipment to be implemented (Artés et al., 2009). Even though, more research is needed to optimize the UV-C application on plant produce (Artés-Hernández et al., 2009; Martínez-Hernández et al., 2011).

Since very little information is available on the effect of UV-C on date palm, the objective of this work was to assess their comparative effects with NaClO on survival of moth of pyrale and on natural microflora growth, as well as on quality attributes of Deglet Nour dates throughout commercial shelf-life.

2. MATERIALS AND METHODS

2.1. Plant material

Deglet Nour dates were hand harvested at the end of October at fully mature ('Tamar' stage) from a farm located in an Oasis of the Governorate of Kébili (South of Tunisia). Immediately after harvest, the bunch of dates was cut into spikelet and about 50 kg fruits were placed in ventilated polystyrene boxes and transported at ambient temperature by car and plane to the Pilot Plant of the Technical University of Cartagena (Spain). Total transport duration was about three days. After arrival, dates were inspected, damaged fruit were discarded and sound dates were sorted to achieve uniformity in the whole lot.

2.2. Sample preparation, treatments and storage conditions

The dates were processed in a disinfected area at room temperature (about 20°C). The following treatments were applied:

UV-C: The UV-C equipment consisted of two batches of 15 reflectors with unfiltered germicidal emitting lamps (TUV 36W/G36 T8, Philips, Holland) fixed to a chamber frame as fully described in Artés-Hernández et al. (2010). According to our preliminary studies on dates the selected UV-C radiation dose was 6 kJ m⁻². In this treatment no water washing was used.

b) NaClO: dates were dipped for 4 min in a water solution at about 15°C containing 100 mg L⁻¹ NaClO at pH 6.0, followed by 1 min rinsing in tap water.

c) Control: dates were dipped in tap water at about 15°C for 2 min.

After sanitizing treatments, dates were manually drained with absorbent paper to eliminate the excess of surface water. Then, 200 g of dates from each treatment were placed in 750 mL PP baskets that were thermally sealed at the top with a 30 µm thickness BPP film (Plásticos del Segura S.L., Murcia, Spain) which was perforated with a 0.7 mm φ needle in order to assure an air atmosphere with high relative humidity within packages.

Three replicates per treatment were prepared and then stored in darkness at 20°C. This adverse temperature, which shortens the shelf-life of dates, was chosen to simulate the normally used at commercial distribution and retail sale scale in Europe.

2.3. Detection of pyrale infestation and microbial analyses

The natural pyrale infestation was calculated in three replicates of 20 dates each by the ratio between the number of dates naturally infested by larvae of *E. ceratoniae* observed by eye and the total number of dates of each replicate expressed in percentage. Three randomized samples from each treatment were taken on the processing day and after 30 days at 20°C to find out natural microflora growth on

dates. By using saboraadoxytetracycline agar base (ScharlauChemie S.A. Barcelona, Spain), mold and yeast colonies were counted three days after incubation at 25°C (NF V 08-059, 1995) and totals mesophilic were counted on plate count agar after 48 h of incubation at 30°C (NF V 08-05, 1996). Coliforms bacteria were counted on violet red bile dextrose agar (VRBD, pH 7.2) (ScharlauChemie S.A. Barcelona, Spain) after 24h at 37°C (NF V 08-015, 1991). All microbial counts were reported as \log_{10} colony forming units per g of sample (\log cfu g^{-1}).

2.4. pH, titratable acidity and sugar concentration

After removing the pits, the dates were cut into small pieces, and ground into a uniform mash. The pH was potentiometrically measured with a digital pH-meter (Crison501, Barcelona, Spain) equipped with temperature control probe (NF V 05-108, 1970). Titratable acidity (TA, g citric acid 100 g^{-1} fw) was monitored by titrating the sample extract with 0.1N NaOH (NF V 05-101, 1974).

In order to quantify the sugar composition, according to Aguayo et al. (2006) it was used an HPLC (Merck Hitachi, Darmstadt, Germany) equipped with a refractive index detector (Hitachi, L-7490 model, Tokyo, Japan), L7100 pump, a Hitachi L7200 automatic sample injector and a LiChroCART 250-4 Purospher STAR NH 2 column (5 μ) (Merck, Darmstadt, Germany). A 20 μ L extract sample was injected using a mobile phase of acetonitrile: water (85:15, v/v) (Merck, Germany) at a flow rate of 1.5 mL min^{-1} flow. The concentrations of sucrose, glucose and fructose were determined and reported as g sugar 100 g^{-1} dry matter (dm).

2.5. Color

The color measurement was made on three randomized sides of 10 dates randomly selected from each treatment. A compact tri-stimulus colorimeter (Minolta CR 300, Ramsey, NJ, USA) with an aperture diameter of 8 mm, previously calibrated with a white calibration plate (C standard C.I.E. illumination, 0° viewing) was used. Results were expressed as CIEL*a*b* color space units. For each fruit, the skin color was determined as L*(lightness) or calculated as Chroma ($C^* = [(a^*^2 + b^*^2)^{0.5}]$) and Hue angle ($H^\circ = \arctg b^*/a^*$) according to CIELAB (1976).

2.6. Sensory evaluation

Sensory analyses were performed according to Martínez-Hernández et al. (2013) on the processing day and after 30 days of shelf-life at 20°C. The panel test consisted of five assessors (three women and two men, aged 25–65 years) screened for sensory ability. Overall quality, color, texture and flavor were evaluated on a five-point hedonic scale (1: extremely poor, 2: poor, 3: acceptable and limit of usability, 4: good and 5: excellent).

2.7. Statistical analysis

For each quality attribute an ANOVA was performed and values reported for treatment and storage period were compared to find significant differences. By the use of Info Stat (version 1), the least significant difference multiple range test at $p < 0.05$ was conducted.

3. RESULTS AND DISCUSSION

3.1. Pyrale infestation and microbial analysis

The infestation level of dates by *E. ceratoniae* was significantly affected by the sanitizing treatments and the storage period (Table 1). As expected, the storage period increased the natural infestation and both sanitizing treatments lowered the natural infestation. In particular, UV-C was the most effective against moth proliferation ($8.33 \pm 2.89\%$ infestation after treatment) compared to NaClO ($15.0 \pm 2\%$) and Control ($26.67 \pm 2.89\%$). As according to Dhoubi (2000) the eggs of pyrale are attached to surface of dates, it could be expected that the UV-C inhibited their growth. In a similar way *Ceratitiscapitata* eggs were eliminated in infested guavas with 16 $kJ m^{-2}$ UV-C and *in vitro* with 1.4 $kJ m^{-2}$ UV-C (Vieira et al., 2009).

Compared to Control samples immediately after both sanitizing treatments the microbial counts of molds, yeasts, total mesophilic and coliforms were significantly lowered (Table 1). The most efficient antimicrobial effect after shelf-life was found in UV-C treatment with a reduction of 1.6 \log cfu g^{-1} for yeast and molds, 2.1 \log cfu g^{-1} for total mesophilic and 1.5 \log cfu g^{-1} for coliforms compared to

Control. The monitored coliforms counts must be considered as relevant due to the risks of these microorganisms for consumers. In this way, results reached with UV-Care particularly interesting.

Table 1. Effect of UV-C and NaClO treatments on moth infestation (%) and microbial counts of molds, yeasts, total mesophilic and coliforms (log cfu g⁻¹) of Deglet Nour date after 30 days at 20°C. Data are means (n = 3) ± SD.

| | UV-C | | NaClO | | Control | |
|------------------|-----------|-----------|-----------|-----------|-----------|------------|
| | Day 0 | Day 30 | Day 0 | Day 30 | Day 0 | Day 30 |
| Moth infestation | 1±0.5 | 8.3±2.9 | 1±0.5 | 15±2.0 | 1±0.5 | 26.66±2.88 |
| Yeasts and molds | 4.72±0.18 | 6.81±0.07 | 4.48±0.19 | 6.85±0.05 | 6.14±0.08 | 8.44±0.005 |
| Total mesophilic | 3.86±0.04 | 6.62±0.05 | 3.91±0.01 | 7.02±0.01 | 5.15±0.16 | 8.69±0.03 |
| Total coliforms | 3.24±0.01 | 3.9±0.006 | 3.18±0.1 | 4.19±0.04 | 3.88±0.1 | 5.42±0.02 |

3.2. Titratable acidity, pH sugar concentration

The effects of selected treatments on pH, TA and sugars concentration are reported in Table 2. No significant differences in TA (about 0.10 ± 0.02 g citric acid 100 g⁻¹ fw on the processing day) among treatments and storage time were found. The storage period slightly lowered the pH (about 5.60 ± 0.10 on the processing day) without differences among treatments.

The current pH values agree with those from Besbes et al. (2009), and confirm that the storage time induced a slight decrease in the pH of dates (Dehghan-Shoar et al., 2010), without significantly changing the TA. Azelmat et al. (2006) reported that the pH decrease throughout storage of dates could be due to the activity of microorganisms and insects.

Table 2. Effect of UV-C and NaClO treatments on pH, acidity (g citric acid/100g FW) and sugars concentration (g 100 g⁻¹ dm) of Deglet Nour date after 30 days at 20°C. Data are means (n = 3) ± SD.

| | UV-C | | NaClO | | Control | |
|--------------|------------|------------|------------|------------|------------|------------|
| | Day 0 | Day 30 | Day 0 | Day 30 | Day 0 | Day 30 |
| pH | 5.56±0.17 | 5.12±0.1 | 5.67±0.11 | 5.18±0.11 | 5.53±0.33 | 5.11±0.04 |
| Acidity | 0.101±0.04 | 0.110±0.01 | 0.083±0.01 | 0.105±0.01 | 0.110±0.01 | 0.120±0.02 |
| Total sugars | 79.06±0.6 | 59.08±0.7 | 79.56±0.5 | 59.10±0.8 | 79.57±1.2 | 58.50±0.7 |
| Fructose | 14.0±1.79 | 10.24±0.5 | 13.5±1.15 | 11.51±1.36 | 14.93±0.64 | 10.29±0.61 |
| Glucose | 19.32±2.81 | 16.8±0.8 | 21.11±1.76 | 21.52±3.99 | 23.57±0.97 | 15.61±1.45 |
| Sucrose | 45.74±3.7 | 32.04±1.23 | 44.95±2.44 | 26.07±1.95 | 41.07±0.35 | 32.60±1.11 |

The sugars concentration (about 79 ± 0.5 g 100 g⁻¹ dmon the processing day) was not affected by treatments (Table 2). Sucrose was the predominant with 32.04 ± 1.23 to 45.75 ± 3.7g 100 g⁻¹ dm. The sugars concentration of DegletNour dates varied with ripening stage, firmness and water content, and different sugar levels have been found. Besbes et al. (2009) reported for total sugars, sucrose and reducing sugars 87.55 ± 0.10 g 100 g⁻¹ dm, 53.59 ± 0.13 g 100 g⁻¹ and 33.96 ± 0.23 g 100 g⁻¹ respectively. Chaira et al. (2007) found total sugars in DegletNour date flesh of 72.82 ± 0.25 %, sucrose 55.08 ± 0.14 % and reducing sugars 17.74 ± 0.33%. Elleuch et al. (2008) reported similar values with total sugars 79.1 ± 0.8 %, sucrose 52.7 ± 0.15 %, glucose 13.7 ± 0.5 % and fructose 12.6 ± 0.2 %. El Arem et al. (2011) reported 63.16 ± 1.59% total sugars, 33.32 ± 1.91% sucrose and 29.79 ± 0.35% reducing sugars.

A decrease of the total sugars concentration after shelf-life from 79.56 ± 0.09 g 100 g⁻¹ to 58.51 ± 0.83 g 100 g⁻¹ was found. A degradation of sucrose and reducing sugar was also found. These results agree with those of Khali et al. (2007) in DegletNour cv.who found decreased values of total sugars from 70.5 to 45.75% fw after 5 months at room temperature. This trend was very probably due to consumption of sugars as respiratory substrates.

3.3. Color

The combined decreases of L*, Chroma and H° values, slightly accentuated in the last two parameters, reflected well the color changes from light brown to dark brown of dates after storage. Compared to Control, at the processing day neither after shelf-life, no changes in L*, Chroma and H° values due to both sanitizing treatments were found (Table 3). It could be cited just as a trend, without significant differences among treatments, that UV-C treated dates showed high L* and Chrome values at the beginning and at the end of the storage period. The L* value found here (33.58 ± 2.05) agrees with that reported by Elleuch et al. (2008) which showed that DegletNour cv. ($L^* = 31.71 \pm 0.57$) was lighter than Allig cv. ($L^* = 22.89 \pm 0.45$).

Results reached in color changes generally agree with those found for others plant products. Kim et al. (2010) showed that NaClO did not affect the strawberries color, although Martin-Diana et al. (2007) found that NaClO reduced L* of fresh-cut lettuce. Fresh processed lettuce tissue became shinier when $8.14 \text{ UV-C kJ m}^{-2}$ was applied possibly due to induction of lignification-like processes started by the lettuce tissue to protect itself against the UV-C stress (Allende and Artés, 2003). However, up to 7.2 kJ m^{-2} UV-C had no significant effect on color of watermelon pieces (Fonseca and Rushing, 2006; Artés-Hernández et al. 2010).

Table 3. Effect of UV-C and NaClO treatments on skin color parameters after 30 days at 20°C. Data are means (n = 3) ± SD.

| | UV-C | | NaClO | | Control | |
|--------|------------|------------|------------|------------|------------|------------|
| | Day 0 | Day 30 | Day 0 | Day 30 | Day 0 | Day 30 |
| L* | 34.55±1.78 | 33.65±2.27 | 32.60±2.77 | 30.98±1.28 | 32.78±2.39 | 32.07±2.92 |
| Chrome | 20.01±2.64 | 15.98±3.05 | 20.37±4.47 | 15.65±2.30 | 20.04±2.72 | 15.60±3.12 |
| Hue° | 61.85±4.24 | 58.31±5.72 | 61.57±5.56 | 59.34±5.98 | 62.29±4.79 | 55.30±4.45 |

3.4. Sensory analysis

Storage conditions (duration, temperature, and RH) are crucial factors for keeping sensory quality of dates (Ismail et al., 2008). In particular, the dark color of date fruits after storage, due to the polyphenoloxidase enzyme activity on polyphenols, depends of the above mentioned conditions. It could be expected that fruit color, appearance, texture, and flavor commonly and progressively deteriorated throughout prolonged storage. However, after 30 days at 20°C only a no significant changes in different sensory scores of the dates for all treatments, without differences among them, were found (Table 4). These results agree with those obtained on Tunisian dates by Khali and Selselet-Attou (2008).

Table 4. Effect of UV-C and NaClO treatments on flavor, texture, color and overall quality of DegletNour date after 30 days at 20°C. Data are means (n = 3) ± SD.

| | UV-C | | NaClO | | Control | |
|-----------------|-----------|-----------|-----------|-----------|-----------|-----------|
| | Day 0 | Day 30 | Day 0 | Day 30 | Day 0 | Day 30 |
| Color | 2.73±0.9 | 3.36±0.67 | 2.60±0.8 | 2.95±0.65 | 2.20±0.94 | 2.86±0.77 |
| Texture | 2.83±0.95 | 3.04±1.15 | 2.56±0.62 | 2.60±0.91 | 3.06±0.97 | 2.59±0.97 |
| Flavor | 3.16±1.09 | 3.23±1.08 | 2.86±0.74 | 3.10±1.04 | 3.30±1.2 | 3.09±1.02 |
| Overall quality | 3.0±0.9 | 3.5±0.8 | 2.86±0.61 | 3.05±0.98 | 2.93±0.96 | 3.11±0.68 |

4. CONCLUSIONS

The treatment of DegletNour dates with UV-C and NaClO showed a positive effect for lowering their natural infestation by *E. ceratoniae* as well as the microbial growth after a shelf life of 30 days of storage at 20°C. Particularly UV-C was the most and highly efficient treatment against moth proliferation without adversely affect all quality attributes. It was remarkable the effect of UV-C for reducing coliforms counts due to their risks for consumers, underlying the possible commercial relevance of this cost effective technique which could be easily put into practice in existing handling and shipping dates facilities.

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