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Radio Frequency Treatment for Postharvest Disinfestation of Dates

Francesco Garbati Pegna*[a], Patrizia Sacchetti[b], Valentina Canuti[a], Serena Trapani[a], Carlo Bergesio[a], Antonio Belcari[b], Bruno Zanoni[a], Ferdinando Meggiolaro[c]

[a] GESAAF - Department of Agricultural, Food and Forestry Systems - Università degli Studi di Firenze, Italy
[b] DISPAA - Department of Agri-food Production and Environmental Sciences - Università degli Studi di Firenze, Italy
[c] Stalam S.p.A. - Nove, Italy
francesco.garbati@unifi.it

Disinfestation is a necessary step in the packing chain of dates for fresh consumption and exposure to radio frequency radiation allows to carry out this operation minimizing the effect on the organoleptic characteristics of the fruits. The authors in a previous work evaluated the possibility of using this treatment for the disinfestation of dates finding that the exposure time should be at least 6 minutes. These results were confirmed by the present work where dates of different varieties were irradiated for 6 min obtaining 100 % mortality of insects, without affecting the aspect of the treated dates. The trials were conducted on dates of Siwi variety, infested in the laboratory by adults of *C. hemipterus* and *P. interpunctella*, and of Deglet Nour variety, naturally infested in the place of origin. The results showed the efficacy of this method for controlling arthropod pests of dry fruits with no undesirable effect on fruit quality and can be considered for the future definition of a disinfestation protocol based on this technology

1. Introduction

The use of Radio Frequency (RF) electromagnetic fields appears to be an interesting alternative to traditional disinfestation methods for dry fruits, including dates, though no industrial process is known to be in use until now for this fruit. RF potentialities for disinfestation have been explored by various authors, but mainly on dry seeds and in fruits of high moisture content. Nelson (1996) reviewed experimental findings of the previous 50 years with respect to entomological and physical factors affecting the response of insects exposed to RF and microwave electromagnetic fields, in grain and grain products, while Marra et al. (2009) reviewed more recent advances in RF treatment of foods. Wang and Tang (2001) compared different disinfestation methods for nuts, outlining the potentialities of dielectric heating and investigated, together with other authors, the effects of RF irradiation for pest control in legumes (Wang et al, 2010a, 2010b) and fresh (Wang et al, 2001, 2003) and dried fruits (Wang et al, 2007).
The date (fruit of *Phoenix dactylifera* L,) is an important agricultural product for many countries of Northern Africa and Western and Southern Asia: in 2012 the land under cultivation in the world has been over 1.1 Mha and the production more than 7.5 Mt (FAO, 2012). As other fresh or dry fruits, dates are exposed to insect damage, either on the tree or during storage, and disinfestation is actually carried out mainly by conventional hot air, cold treatment, fumigation or treatment in controlled atmosphere. Concerning exposure of dates to electromagnetic radiation, the dielectric properties of dry dates exposed to 2.45 GHz microwave radiation (Ali et al, 2000) and to 27 MHz RF radiation (Alfaifi et al, 2013) have been determined and found comparable with those of other fruits at similar moisture content.
The authors of this paper, in a previous research (data not published), have found that exposure to RF radiation for 6 min is lethal for adults, larvae and pupae of *Carpophilus hemipterus* (L.) (Coleoptera: Nitidulidae), artificially inserted in date fruits, without significant alteration of aspect and moisture content of the

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fruits, while other authors (Johnson et al, 2003) determined that temperature above 50 °C is lethal for larvae of *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae). The objective of the present study is to confirm these results by evaluating the efficacy of 6 min RF treatment on dates infested by different insects and the possible effects on fruit quality in order to develop a disinfestation protocol based on this technology.

2. Materials and methods

2.1 Dates and infestation

Tests have been carried out on dates of the Siwi and Deglet Nour varieties. Siwi is a semi-dry variety indigenous from Egypt while Deglet Nour is a semi-dry variety common in all date producing Countries, especially in North Africa. The Siwi dates used for the trials were harvested in 2012 in the Oasis of Siwa in Egypt and preserved in sealed commercial packages until infested in the laboratory by the dried fruit beetle, *C. hemipterus* and the Indian meal moth *P. interpunctella*, two common insect pests of dates that feed on ripe dates in the bunches, on the ground and in the packinghouses. The Deglet Nour dates were harvested in 2014 in the Biskra Province in Algeria and preserved without precautions before shipping to Italy as they had been naturally infested in their original environment.
The in-the-laboratory infestation has been obtained by exposing two groups of 100 dates to insect infestation by introducing fruits into adult cages. In order to obtain dates infested by different developmental stages of each pests, for each group 50 dates have been infested four weeks before the treatment and 50 dates have been infested two weeks prior the experiment. Adults were allowed to infest dates for three days. Each sample was composed by dates of each group.

2.2 Process

The trials were conducted in the laboratory of Stalam S.p.A. in Nove (Vicenza, Italy) using 4 samples of Siwi dates and 4 samples of Deglet Nour. Each sample of Siwi was composed by 15 dates infested with *C. hemipterus*, 15 dates infested with *P. interpunctella* and 70 non-infested dates: the weight of the single fruits varied from 9 to 14 g and the length between 30 and 39 mm, the moisture content was 13.5 % and the total weight of the sample was about 1,100 g. Each sample of Deglet Nour was composed by 80 dates: the weight of the single fruits varied from 6 to 10 g and the length between 35 and 43 mm, the moisture content was 17.5 % and the total weight was about 650 g.
The control samples for Siwi were constituted by 40 dates infested with *C. hemipterus* and 40 dates infested with *P. interpunctella* and by 60 dates non-infested for color control. The control sample for Deglet Nour was constituted by 200 dates picked up from the original batch. Table 1 shows the composition of the samples.
Each sample was processed singularly, by exposing the dates adjusted in a single layer on a plastic tray, to radiofrequency radiations for 6 min in a 27.12 MHz, 3.5 kW lab scale RF tunnel, produced by Stalam S.p.A. and equipped with parallel-plate electrodes with adjustable distance and with a 600 mm wide shielded mouths for conveying the dates to and fro inside the irradiation chamber, to simulate the continuous process. The irradiation chamber measured 1,200 x 800 mm and was heated at 60 °C with a thermostatic temperature control, the electrodes were kept at a fixed distance and the potential difference was 5,000 V. Each sample was weighted before and after the trial and photographed with a 16 Mpixel digital camera (Power Shot A2500, Canon Inc., Tokyo, Japan) and with an infrared camera (i7, FLIR Systems, Wilsonville, OR, USA) with f=6.8 mm IR lens, from a distance of about 0.5 m before and after exposure. Temperature was monitored during the exposure and for 3 minutes after the end of the treatment, with a 4-channel signal conditioner (Reflex, Neoptix Inc., Québec, QC, Canada) for fiber optic temperature measurement with 4 fiber optic probes and with a 1-channel signal conditioner (Nomad, Neoptix Inc., Québec, QC, Canada) with 1 fiber optic probe; the two systems were adjusted before the beginning of the trials. The 5 probes were inserted in 5 dates positioned in 5 different areas of the tray as showed in Figure 1.

Table 1: Test layout

Date type	RF treatment (n./sample)	Control (n./sample)	Replicates (n.)	Total RF treated (n.)	Total control (n.)
Siwi infested by *C.h*	15	10	4	60	40
Siwi infested by *P.i.*	15	10	4	60	40
Siwi non-infested	70	15	4	280	60
total	100	35	4	400	140
Deglet Nour	80	50	4	320	200

Figure 1: Sample of Siwi dates with probes before treatment (tags indicate different types of infestation)

2.3 RF treatment evaluation

After treatment the samples of Siwi infested dates and Deglet Nour have been controlled, for determining insect presence and mortality, in the laboratory of DISPAA, entomology section, and compared with the untreated samples. Samples of treated and untreated non-infested Siwi dates have been analyzed in the laboratory of GESAAF, food technology section, for detection of aspect alterations due to the treatment.
Treated and untreated dates have been checked within three days after the RF treatment in order to verify the presence of live or dead insects, identifying the insect order and the developmental stage. The infestation reduction due to the RF treatment has been calculated according to Abbott's formula (Abbott, 1925)
The aspect of treated and untreated Siwi dates was defined through the measurements of color. Three fruits from each treated sample were randomly selected and longitudinally cut in order to both remove stone and obtain two uniform sections of the fruit. Each section was subjected to color measurement at two different surface locations. In order to measure fruit color a sample holder was build consisting of a Teflon plate (76 mm x 76 mm x 2 mm) with a glass window. CIELAB color coordinates were measured by a spectrophotometer (Lambda 35 UV/VIS, Perkin Elmer Inc., Waltham, MA, USA) equipped with Labsphere RSA-PE-20. A dark brown standard was used for each analysis session to evaluate measurement performance. UV WinLab Softwares were used to both record spectra (version 2.85.04, Perkin Elmer Inc.) and L*, a* and b* coordinates (version 3.00, Perkin Elmer Inc.). Data on the quality evaluation of dates were processed by Statgraphics Centurion 285 (Version XV, Statpoint Inc., USA) with multifactor ANOVA considering the factors: date fruit sample, replicate and treatment. Fisher's least significant difference (LSD) procedure has been used to discriminate between means.

3. Results and discussion

Deglet Nour dates were attacked mainly by Lepidoptera with an overall infestation of 21.54 % (larvae or dates with frass). Observation of full grown larvae revealed that the most representative species was presumably the carob moth, *Apomyelois ceratoniae* (Zeller). Besides these few dates infested by Coleoptera (2.50 %) were found, most likely Nitidulids.
The effects of RF treatment on insects infesting Deglet Nour dates (Table 2) have been remarkable since no living insect was found in the four replicates, showing an efficacy of 100 %, according to Abbott's formula.
Table 3 and Table 4 report the effects of RF treatment on Siwi dates infested by *P. interpunctella* and *C. hemipterus*. In both cases, in the four replicates, 100 % of mortality was obtained where the control replicates showed 85 % of dates containing small and/or full grown larvae of the moth and 27.5 % containing adults of the beetle. Actually dates exposed to *C. hemipterus* were infested only by adults who entered the fruits; moreover, due to the aggregation behavior of this species, very few dates were infested with tens of adults each. In any case the results are meaningful since the adult is the most resistant stage to high temperatures (Al-Azawi et al., 1984). Similar remarkable results have been obtained with the Indian meal moth since the 6 minutes treatment killed all the full grown larvae present in the samples (about 10 % of the overall larvae)

being the fifth instar the most heat-tolerant stage of this species (Johnson et al, 2003; Mahroof and Subramanyam, 2006).

Table 2: Infestation assessment of Deglet Nour dates in the control and RF treated replicates. "Infested dates %" includes both dates with insects and dates with frass.

	Replicate	Dates (n.)	Dates with living insects (n.)	Dates with dead insects (n.)	Infested dates (%)	Dates with dead insects (%)	Dates with living insects (%)
control	1	50	6	1	24.00	2.00	12.00
	2	50	6	2	26.00	4.00	12.00
	3	50	9	2	32.00	4.00	18.00
	4	50	8	1	24.00	2.00	16.00
	overall	200	29	6	26.50	3.00	14.50
RF treatment	1	80	0	16	23.75	20.00	0
	2	80	0	18	27.50	22.50	0
	3	80	0	20	33.75	25.00	0
	4	80	0	9	12.50	11.25	0
	overall	320	0	63	24.38	19.68	0

Table 3: Infestation assessment of Siwi dates infested by *P. interpunctella in the control and RF treated replicates*

	Replicate	Dates (n.)	Living larvae (n.)	Dead larvae (n.)	Dates with living larvae (n.)	Dates with dead larvae (%)	Dates with living larvae (%)
control	1	10	19	0	7	0	70
	2	10	23	0	9	0	90
	3	10	46	0	10	0	100
	4	10	28	0	8	0	80
	overall	40	116	0	34	0	85
RF treatment	1	15	0	26	0	60.00	0
	2	15	0	30	0	66.67	0
	3	15	0	27	0	60.00	0
	4	15	0	23	0	66.67	0
	overall	60	0	106	0	63.33	0

Table 4: Infestation assessment of Siwi dates infested by C. hemipterus in the control and RF treated replicates

	Replicate	Dates (n.)	Living adults (n.)	Dead adults (n.)	Dates with living adults (n.)	Dates with dead adults (%)	Dates with living adults (%)
control	1	10	1	0	1	0	10.00
	2	10	57	0	4	0	40.00
	3	10	8	1	4	10.00	40.00
	4	10	5	0	2	0	20.00
	overall	40	71	1	11	2.50	27.50
RF treatment	1	15	0	0	0	0	0
	2	15	0	2	0	13.33	0
	3	15	0	1	0	6.67	0
	4	15	0	68	0	33.33	0
	overall	60	0	71	0	13.33	0

The effectiveness of the process is due to the short time in which the relatively high temperatures inside the fruits are reached. in all the dates in which the temperature was controlled through the fiber-optic probes, the initial temperature was in the range 17.5-19.5 °C, the maximum recorded temperature was in the range 61.0 - 62.2 °C for the Siwi dates and in the range 62.5 – 67.2 °C for Deglet Nour and was reached 410-430 s after the beginning of the irradiation process in the first case and after 370-410 s in the second case, as shown in Table 5 and Table 6. The higher temperatures reached by Deglet Nour and the higher rate of heating can be explained by their higher moisture content.
Table 7 shows the results of the color analysis: this table applies a multiple comparison procedure to determine which means are significantly different from which others. The bottom half of the output shows the estimated difference between each pair of means. An asterisk has been placed next to 1 pair, indicating that this pair shows a statistically significant difference at the 99,0 % confidence level.
The table shows that there aren't significant differences between the sample reference (C) and samples treated (T) if the trichromatic coordinates a* (red - green) and L (brightness) are considered. The samples are rather statistically different for the coordinate b* (yellow - blue). It is possible to consider that the differences between C and T aren't significant: this is confirmed also by the ΔE since all values are less than 5 and then the differences are negligible. Finally it can confirm that the differences are minimal and that aren't perceptible to the naked eye.

4. Conclusions

Six minutes RF treatment allowed to achieve complete disinfestation of dates attacked by moths and beetles without significant alteration of color and moisture content of the fruits. The treatment was effective for dates of the variety Siwi, infested by *C. hemipterus* adults and *P. interpunctella* full grown larvae, which are considered as the most resistant stages of these two common insect pests, as well as on various other insects colonizing dates of the variety Deglet Nour and this makes it a very interesting option for those date packing industries which include the disinfestation phase in their process. As a matter of fact RCF treatment constitutes an effective alternative to the actually widespread methods, based on hot or cold air treatment, fumigation or controlled atmosphere, which show several drawbacks in terms of cost, timeliness and effect on the organoleptic characteristics of the treated product. The effectiveness of the tested conditions opens the way to the definition of a protocol for RF disinfestation of dates on industrial scale.

Table 5: Maximum temperatures reached by Siwi dates in relation to the beginning and end of irradiation

Trial n.	Temperature (°C)	Time after start (s)	Time after stop (s)	Duration (s)
1	61.2	410	50	20
2	62.2	420	60	20
3	61.0	430	70	10
4	61.0	410	50	10

Table 6: Maximum temperatures reached by Deglet Nour dates in relation to the beginning and end of irradiation

Trial n.	Temperature (°C)	Time after start (s)	Time after stop (s)	Duration (s)
1	62.5	410	50	20
2	63.2	380	20	40
3	67.2	370	10	20
4	62.6	370	10	10

Table 7: Average results of the trichromatic coordinates of Siwi dates samples untreated (C), treated (T) and LSD (Fisher's least significant difference).

	C	T1	T2	T3	T4	LSD
a*	4.48 ab	5.19 b	3.63 a	4.82 ab	4.98 ab	± 0.69 ns
b*	2.50 bc	3.87 c	0.39 ab	0.30 a	-0.26 a	± 1.08***
L	25.02 a	24.35 a	25.84 a	26.15 a	24.70 a	± 1.17 ns
ΔE		1.68	2.42	2.50	2.82	

values followed by different letters within the same row differ significantly (*** P ≤0.001, ns = not significant)

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