

Use of Heat for Disinfestation and Control of Insects in Dates: Laboratory and Field Trials

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Heat treatment of dates at the time of harvest, was examined as an alternative to fumigation with methyl bromide, which has been phased out in Israel under the Montreal Protocol. In laboratory studies, the influence of 40°, 45°, 50° and 55°C on the levels of disinfestation and mortality of *Carpophilus hemipterus* larvae was examined over a 2-h exposure period. The ratio of the number of insects found outside the feeding sites to the total number of insects, was greatest at 50°C (92.3%), significantly greater than at 40° and 55°C. At 50° and 55°C, 100% mortality was obtained. Since drying temperatures for most date varieties is between 45° and 50°C, and because percent disinfestation and control was most effective at 50°C, these findings were examined under field conditions. Field trials were carried out at a commercial drying station where the crated dates are placed on the ground inside a plastic-clad hothouse. The stacks were covered with plastic liners to form 'drying ducts' through which heated air was sucked using fans positioned at the opposite end of the ducts. It was shown that from 1 to 2 h were required for the dates to reach the set temperature of 50°C. During the following 2-h aeration, the dates were exposed to 50°C heated air, after which an examination of infested dates inserted into the drying ducts, and natural infestations showed that successful control and emigration were obtained. This method produced results comparable to those obtained with methyl bromide fumigation, and was suitable as a replacement technology for infestation control.

KEY WORDS: Dried fruits; nitidulid beetles; heat; insect control; methyl bromide alternatives.

INTRODUCTION

In Israel, the date variety Madjoul is very popular because of its large size, texture, and particular taste. Consequently, its cultivation is on the increase, particularly in the Jordan Valley, where technologies on maturation and drying, employing solar energy, have been developed and practiced (J. Kanner *et al.*, 1998, Annu. Rep. to Israel Min. of Agriculture, Res. Project no. 416-0435-97). However, field infestations of nitidulid beetles in all date varieties pose a serious contamination problem which may compromise exports. Until now, this problem has been addressed successfully using the fumigant methyl bromide (MB). Its use has been mandatory in Israel, particularly because it causes a high proportion of larvae and adults to emigrate from the fruit before they succumb (5,15; S. Navarro *et al.*, 1989, Final Rep. BARD Project no. I-1095-86). This emigration phenomenon is no less important than the toxic effect of the treatment, because minimum acceptance tolerances have been set for the presence of both dead and live insects in dried fruits.

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For several decades MB has been a mainstay treatment to kill a wide array of quarantine pests as well as those encountered in orchards, packinghouses, and food plants. However, this potent fumigant is now associated with depletion of the Earth's ozone layer and, under the terms of the Montreal Protocol (17), it was phased out in January 2005 for Non-Article 5 (developed) countries, and will be phased out in the year 2015 for Article 5 (developing) countries, except for quarantine and pre-shipment fumigations. Nevertheless, the Methyl Bromide Technical Options Committee (MBTOC) of the UNEP (17) recognizes the problem of MB phase-out in Non-Article 5 countries and, in consequence, has recently published a handbook on Critical Use Nominations (CUNs) for MB (16). This is to help parties submit requests for exemptions on a yearly basis under circumstances where no technically or economically feasible alternative treatment to MB yet exists. However, the parties that submit these nominations must demonstrate to the evaluation panel that they are making intense efforts to search for suitable alternatives. The present study was conducted for the express purpose of finding such a feasible alternative for the disinfestation and control of insect pests in dried dates.

The only other universally available fumigant for control of durable commodities is phosphine (PH_3). Regrettably, although PH_3 is a very useful fumigant, it is slow acting, it has not been shown to cause insects to emigrate from the dates (disinfestation), and insects in various countries have developed resistance to this gas (18,19). Other treatments have been examined for their disinfestation effect (1,5–7,14; S. Navarro *et al.*, 1989) including the use of high carbon dioxide concentrations, low oxygen concentrations and low pressures. Although all three treatments do cause high levels of disinfestation, they are not readily suitable for practical application at the point of entry into the date packing houses. In contrast, although high temperatures have been studied for insect control, their capacity to produce disinfestation has not been investigated previously, even though the use of heat is already part of the postharvest handling procedure. If shown to be effective, disinfestation by heat treatment might be applicable at the date drying stage.

The use of heat has been revived as a potential alternative to MB in a variety of situations. Insect pests of stored products survive and multiply over narrow ranges of temperatures. For each species there is a minimum and maximum temperature at which it is able to develop. At certain low temperatures, oviposition and larval growth cease; and at specific high temperatures egg sterility occurs and mortality increases. The lower and upper limits and optimal temperatures of most of the important stored-product species have been studied and are well known. Howe (12) lists minimum and optimum critical temperature and humidity ranges for the major grain storage insects.

Heat treatments mainly against insect pests of stored grain and flour have been studied in the laboratory (7–10) and implemented in the field (3,11). With regard to the dried fruit industry, heat treatments are not presently recognized as a recommended process for disinfestation. Laboratory studies by Lindgren and Vincent (13) showed that to obtain 90% mortality of adult nitidulids, exposure to 49°C for 4 to 20 min was needed, depending upon the ambient relative humidity. Al-Azawi *et al.* (2) showed that under laboratory conditions, adults of the dried fruit beetle, *Carpophilus hemipterus* (L.), are tolerant to heat, and that exposures from 25 to 60 min at 50°C are required to achieve complete mortality. They found that for complete mortality of all stages of *Cadra cautella* on stored dates, exposure to 60°C for 33 min was required (1). However, studies that consider not only control but also emigration of nitidulid beetles from dates using heat during the drying process are

lacking in the literature. In the drying process of the date industry temperatures are usually kept moderate (35° to 55°C) to avoid damage. For example, temperatures used for drying the Madjouli variety in Israel should be kept within the range of 45° to 55°C to avoid discoloration and a blistering effect that separates the skin from the flesh of the fruit.

We consider here the possibility that heat treatment may be effective in producing both emigration and control of nitidulid beetles in dates. The study evaluates the results of laboratory experiments using *C. hemipterus*, followed by field trials to integrate this technology into the existing date drying procedures in Israel.

MATERIALS AND METHODS

LABORATORY EXPERIMENTS

The insects Individuals of *Carpophilus hemipterus* L. were collected from infested dates. The beetles were reared in a rearing room at 26°C and 75% r.h. on an artificial diet composed of 1 l water, 125 g cornstarch, 90 g glucose, 44 g sugar, 50 g brewer's yeast, 18 g agar extract, 3.1 ml propionic acid and 1 g methyl 4-hydroxybenzoate in 10 ml of ethanol. Cultures were set up by placing adult beetles in 200 ml jars containing about 150 g of food medium mixed with sawdust. Following 2 days of oviposition, adults were removed and the insects were reared in these jars until required for the experiment. Larvae 6–8 days old were removed from the cultures for the exposure tests, as needed.

Although both adults and larvae may be found within infested dates, it was shown in preliminary observations that adult mortality is more sensitive to high temperatures and that emigration of adults was greater than that of larvae. Consequently, only larvae were used in the laboratory experiments.

In previous studies (5,6; S. Navarro *et al.*, 1989) the high variance in infestation levels of dates in the laboratory posed significant limitations in using dates for experiments on disinfestation. Therefore, the effectiveness of an artificial feeding site was compared with that of naturally infested dates and found suitable for disinfestation studies (6). The artificial feeding sites destined to simulate the dates consisted of cardboard rectangles placed on food medium contained in petri dishes (6). Larvae to be used in experiments were removed from the culture medium and transferred to the artificial feeding sites 24 h before exposure to the different treatments. All larvae that had not penetrated into the feeding sites were then removed immediately prior to exposure. The feeding sites to be used in the experiments were cut out from the petri dishes. Based on examination of 420 feeding sites used in the tests, the average number of larvae was 3.7 per feeding site.

Experimentation Ten feeding sites, each containing up to four larvae, were introduced into the exposure chambers consisting of 2.54 l desiccators. Each exposure chamber (desiccator) used in the treatments served as a replicate. These chambers were placed in thermostatically controlled incubators. Temperatures of 40°, 45°, 50° and 55°C were tested for determining the effect of percent disinfestation and mortality of larvae. Prior to the exposure experiments, feeding sites were placed in the chambers into which had been inserted thermocouples connected to temperature loggers in order to determine the rates at which the feeding sites reach the target temperatures. For each treatment, an exposure time of 2 h after the feeding sites had reached the test temperature, was employed. This exposure time was chosen based on the findings of Fields (8) and after preliminary tests showed that at this duration, mortality took place within the chosen range of temperatures.

The ratio of the number of insects found outside the feeding sites to the total number of insects, was used to describe the term 'percent disinfestation'. Immediately after each treatment, the number of survivors both inside and outside the sites was recorded and percent mortality was calculated. From 4 to 13 replicates of these experiments were made at each temperature. The data obtained were analyzed using a one-way analysis of variance, and significance of differences between the means was analyzed by the least significant difference test.

FIELD TRIALS

The field trials were carried out in the drying facility of the Timura Company located at the agricultural cooperative Moshav Mehola in the northern Jordan Valley during October 2003. Here we report on two field experiments using artificially infested dates. The experiments were conducted in order to evaluate the efficiency of the technology. A third trial was conducted on naturally and heavily infested dates received from date plantations.

The drying facility The facility consisted of a polyethylene clad hothouse 40 m long \times 10 m wide \times 3 m high, specially prepared for large-scale commercial drying of dates (Fig. 1). The hothouse can accommodate up to 12 rows of stacked dates positioned in-parallel across the hothouse and covered over their top and sides with polyethylene liners to form drying ducts (a). Each row consists of ten pallets, arranged five pallets lengthwise and two pallets across. Each pallet holds crates stacked 20 layers high with five crates (40 \times 60 \times 10 cm) per layer. Each crate holds 3 kg of dates one layer deep of the variety Madjoul. Thus, a standard row consisting of ten pallets holds 3 tons of dates. One open face of each row (b) is set opposite the hot air outlet duct running along one wall, while a large axial fan (c) is positioned against the opposite open face of the row (d) leaving a walk-around distance of approximately 50 cm from the facing wall of the hothouse. For the drying procedure a heater (e) consisting of a chamber equipped with electrical heating elements is used to raise the ambient air temperature when needed, and is controlled by a thermostat. The air is directed through a polyethylene sleeve distribution duct (f), 60 cm in diameter, along the outer wall of the hothouse. Outlets in the distribution duct (g) 30 cm in diameter, positioned opposite each row, enable the hot air to be diverted into the drying ducts. The fans (c) set against the opposing face of the drying ducts create an air-flow through the crated dates inside the ducts. The fan casing measures 1.38 \times 1.38 m, the fan diameter is 1.28 m, and fan specification is 1.5 HP, 1.1 KW. The whole system takes advantage of the ambient solar heat during the harvest season, supplemented by a heater to compensate for drops in temperature during the cooler times of the day and night.

This centralized setup is a recent development of the original method whereby maturation and drying of the Madjoul dates were achieved by stacking crated dates on pallets to a height of 2 m. The stack is then sealed with 'shrink' sheeting except for entry and exit apertures at the bottom and top, respectively. The stack is exposed to vertical convection currents caused by solar-heated air within the column (the chimney effect) (J. Kanner *et al.*, 1998). When MB fumigations were carried out, the dates were first fumigated in a chamber and then transferred to the drying facility.

Dates for bioassay Ripe and intact Madjoul dates were selected and artificially infested with *C. hemipterus* larvae in the laboratory. For each sampling site, 30 to 50 dates were placed in a standard crate, which was then placed over another empty crate lined with a

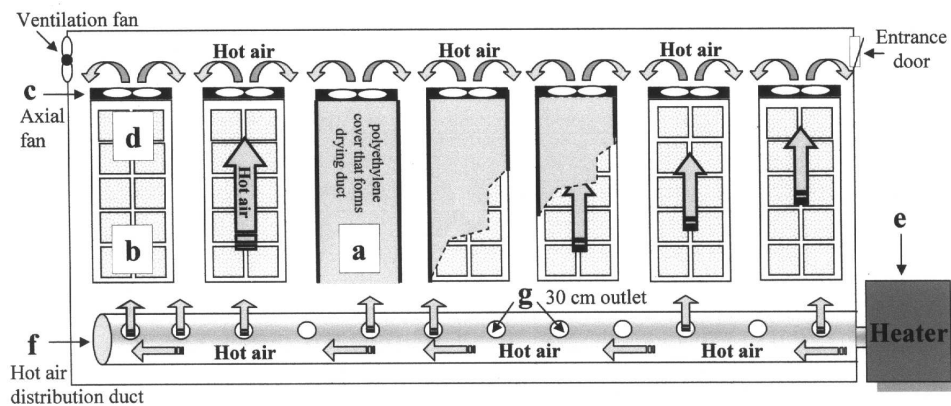


Fig. 1. Schematic diagram of the date drying hothouse. a, drying ducts; b, open face of a row; c, axial fan; d, opposite open face of a row; e, heater; f, distribution duct; g, outlets in distribution duct.

white plastic liner. The infestation level of these dates was approximately ten insects per date. These double-crates were then inserted into the stacked dates in the drying duct at the following four points estimated to be of critical importance: front-top, front-bottom, rear-top and rear-bottom, where 'front' refers to the fan end and 'rear' refers to the opposite end. All these peripheral points were critical because they were suspected to be outside the region of direct air-flow. Mapping of air-flow rates at the fan end showed that for points within the fan diameter, rates from 13.1 to 14.6 m sec^{-1} were recorded, whereas for points outside the fan dimensions, flow rates ranged from 3.2 to 8.6 m sec^{-1} . At the end of the 2-h 50°C exposure period, these double-crates were removed from the drying duct. The number of dead and live larvae collected in the lower crates was counted, and each test date was opened and examined for dead and live larvae. The data obtained were then analyzed for disinfestation levels and mortality.

Natural infestations The third trial was conducted to evaluate the disinfestation levels and mortality of natural infestations during the drying procedure. For this purpose a white plastic sheet 7.2 m^2 in area was placed under six pallets within a drying duct. At the end of the drying period of 72 h which included the 2-h disinfestation treatment at 50°C , the removed larvae were collected from the plastic sheeting, and also date samples from eight crates were taken for analysis of disinfestation and mortality. Due to the large numbers of insects collected on the sheeting, the insects were weighed and weight factors were applied to obtain approximate numerical counts.

Temperature monitoring For trials 1 and 2 the dryer thermostat was set to turn the heater off at 50°C . Temperature loggers were placed in each infested crate (situated at peripheral points suspected to be most variable in temperature), and these were recovered after each trial and downloaded in the laboratory. Data-loggers (HOBO Pro Series) also recorded ambient temperatures inside and outside the hothouse throughout the drying period.

RESULTS

LABORATORY EXPERIMENTS

Temperature time-delay The time-delay required for the artificial feeding sites to attain the target temperature from the initial ambient temperature of 26°C, was ~60 min for all the temperatures tested. This is illustrated in Figures 2 and 3, similar curves being obtained at other temperatures. However, it can be seen that temperatures within the sites were always 1° to 2°C lower than the nominal test temperature

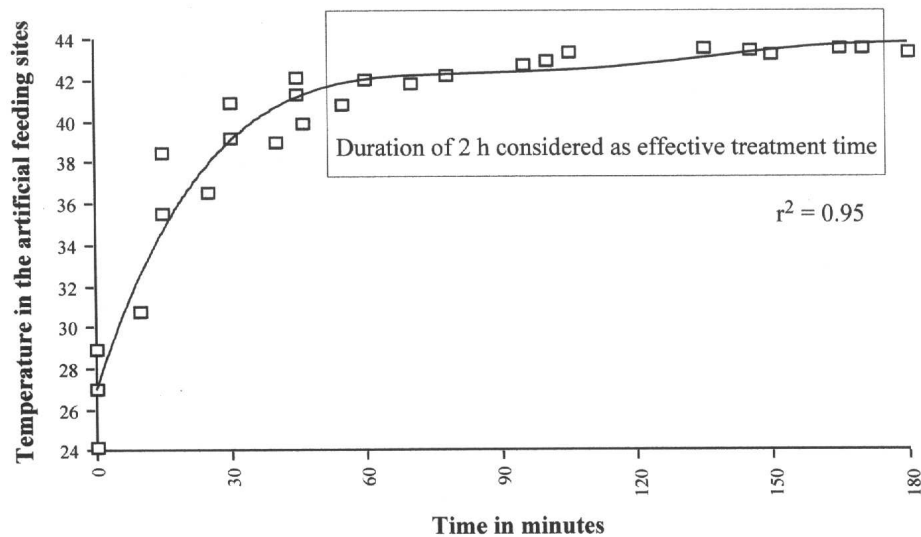


Fig. 2. The time-delay required for the artificial feeding site to attain the target temperature of 45°C from the initial ambient temperature of 26°C.

Disinfestation Table 1 shows that the highest disinfestation value was obtained at exposure to 50°C and averaged 92.3%, whereas at 55°C significantly lower (72.8%) disinfestation was obtained in comparison with the result at 50°C. Exposure to 26°C served as control that resulted in 37.4% of the larvae abandoning the feeding site during the test. One-way analysis of variance for disinfestation data in Table 1 resulted in an F ratio value of 25.02.

Mortality The highest mortality values reached 100% and were obtained at 50° and 55°C (Table 1). At 45°C, mortality ranged from 24.5% to 65.2%, whereas at 40°C no mortality was recorded. One-way analysis of variance for disinfestation data in Table 1, resulted in an F ratio value of 371.08.

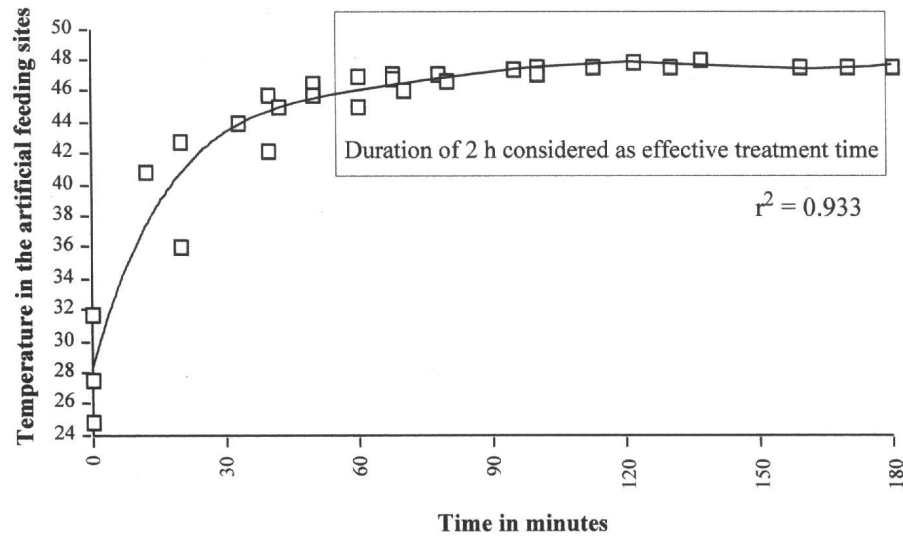


Fig. 3. The time-delay required for the artificial feeding site to attain the target temperature of 50°C from the initial ambient temperature of 26°C.

TABLE 1. Percent mortality and percent disinfestation of *Carpophilus hemipterus* larvae from artificial feeding sites after exposure for 2 h at different temperatures

Temp. (°C)	No. of Replicates	Disinfestation ^z		Mortality ^y	
		Mean (%)	S.E. ^x	Mean (%)	S.E. ^x
26	13	37.4	3.16	0.9	1.7
40	7	48.4	4.3	0.0	2.3
45	6	72.9	4.6	43.7	2.5
50	4	92.3	5.7	100	3.1
55	4	72.8	5.6	100	3.1

^zAnalysis of variance for disinfestation data was: df and mean square for model were 4 and 3240.9, respectively. df and mean square for error were 29 and 129.5, respectively; *F* ratio was 25.02.

^yAnalysis of variance for mortality data was: df and mean square for model were 4 and 14374.4, respectively. df and mean square for error were 29 and 38.7, respectively; *F* ratio was 371.08.

^xStandard errors are pooled estimates of error variance.

FIELD TRIALS

Date temperatures Temperature data derived from data logger compilations in trial 2 are given in Table 2, and are also representative of results obtained in the other trials. These data indicate that the target temperature of 50°C was not achieved in these peripheral regions within 1 h after the introduction of the dates into the dryer. In these peripheral regions, temperatures were from 2° to 3°C lower than the target temperature. Although there was a delay in temperature rise, the target temperatures were approached with a delay of approximately half an hour. The most problematic region was close to the fan at the bottom, probably due to a low air-flow rate at this point.

TABLE 2. Temperatures recorded at bioassay sampling sites during trial 2, after 1 h pre-heating period, with thermostat set at 50°C

Time (min)	Temperature (°C)				
	Top front inside the date	Bottom front inside the date	Top rear inside the date	Bottom rear inside the date	Ambient air inside the hothouse
0	48.4	45.9	44.7	44.6	49.5
30	48.4	45.9	50.9	47.7	49.5
60	48.4	47.1	49.5	48.5	^z
85	49.0	47.1	49.5	48.0	44.5 ^y
95	50.6	48.1	50.2	51.7	46.7
140	51.7	48.8	51.2	50.7	53.0
185	51.2	49.0	51.0	49.8	52.0
230	50.4	49.1	50.7	51.9	50.0

^zNot recorded due to power failure for 20 min.

^yThermostat was raised to 53°C for 20 min.

Disinfestation levels and mortalities in trials 1 and 2 Results of the bioassays carried out in trials 1 and 2 at critical test points that were suspected to be outside the region of direct air-flow, are given in Tables 3 and 4. Bioassays of the infested dates resulted in total emigration and mortality of larvae at the top layers of the drying row. For the dates that were placed in the lower peripheral region, where it was found that air-flow rates were lower, these slight differences allowed survival of larvae at the end of both the 3 h and 4 h exposures.

Disinfestation levels and mortalities in the trial using naturally infested dates The counts of insects recovered from beneath the crated stacks at the end of the date drying procedure are given in Table 5. From these counts, calculation of the initial infestation was made. At the end of the drying period, of the eight samples each containing approximately 50 dates, only one sample contained three infested dates totaling 22 dead larvae. This sample was located at the bottom of the stack.

DISCUSSION

In an earlier study on the effect of MB on disinfestation and mortality (5), the highest level of disinfestation obtained with *C. hemipterus* and *Carpophilus mutilatus* larvae did not exceed 90%. Therefore, the disinfestation level (92.3%) reached in the laboratory experiments of our study (Table 1) are in line with the best known method for disinfestation using MB. This effect was further supported in field trials using laboratory naturally infested dates (Tables 3 and 4). The fumigation conditions for this disinfestation level were 16 mg l^{-1} for a 4-h duration at 26°C. With regard to mortality using MB, Donahaye and Navarro (4) showed that for *C. hemipterus* larvae, exposed for 2 h to MB at 26°C, 22.4 mg l^{-1} was required to obtain 95% mortality (LD_{95}). Mortality data provided in Table 1 also show that complete control of the beetle larvae can be achieved at temperatures of 50° and 55°C. The low disinfestation and mortality levels recorded at the extremities of the drying tunnel (Tables 3 and 4, bottom front and rear) were derived from the poor air distribution. Airflow rates recorded in these areas of the drying tunnel were negligible compared to 1 to 2 m sec^{-1} measured at the center inside the tunnel. To overcome this problem, the company has altered the airflow configuration at these critical points to enable better air

TABLE 3. Disinfestation levels and mortality of *Carpophilus hemipterus* larvae after 3 h at 50°C at four sites in the drying duct in trial 1 using laboratory infested dates that served as bioassay

Location of sample in the drying ducts	No. of dates experimentally infested	No. of insects removed from the dates	No. of insects that remained in the dates	Dates containing insects after treatment	Disinfestation % of the total insect population	Disinfestation % of the dates	Mortality % of insects that remained in the dates
1 (top front)	33	0 (317) ^z	0 (12)	7	100	96.4	100
2 (bottom front)	35	4 (124)	154 (0)	9	44.0	45.4	0
3 (top rear)	35	0 (248)	0 (8)	5	100	96.9	100
4 (bottom rear)	30	2 (117)	61(0)	10	59.1	66.1	0

^zLive insects (in parentheses, dead insects)TABLE 4. Disinfestation levels and mortality of *Carpophilus hemipterus* larvae after 4 h at 50°C at four sites in the drying duct in trial 2 using laboratory infested dates that served as bioassay

Location of sample in the drying ducts	No. of dates experimentally infested	No. of insects removed from the dates	No. of insects that remained in the dates	Dates containing insects after treatment	Disinfestation % of the total insect population	Disinfestation % of the dates	Mortality % of insects that remained in the dates
1 (top front)	35	0 (571) ^z	0 (20)	5	100	96.6	100
2 (bottom front)	35	3 (230)	143 (9)	28	44.0	60.5	5.9
3 (top rear)	35	0 (475)	21 (17)	6	100	96.9	100
4 (bottom rear)	30	0 (370)	61(0)	16	59.1	92.6	0

^zLive insects (in parentheses, dead insects)

TABLE 5. Insects recorded after 72 h drying on 7.2 m² liner placed beneath six pallets of crated dates

Insect	Insects on floor	Insects kg ⁻¹ ^z
<i>Carpophilus mutilatus</i> adults	6,451	3.6
<i>Carpophilus hemipterus</i> adults	1,873	1.0
<i>Epuraea (Haptoncus) luteola</i> adults	208	0.1
<i>Carpophilus</i> spp. larvae	12,276	6.8
<i>Lasioderma serricorne</i> adults	209	0.1
<i>Tribolium castaneum</i> adults	4	0.002
<i>T. castaneum</i> larvae	8	0.004
<i>Cryptolestes</i> sp. adults	3	0.002
Total	20,791	11.6

^zOriginal infestation based on 1,800 kg stacked on six pallets above the liner, each holding 100 crates containing 3 kg dates per crate.

distribution. No subsequent trials with infested dates were carried out by the authors of this study. However, verbal reports by the company on the following date harvest season indicate that correction of airflow configuration solved this problem.

Carpophilus hemipterus was selected as the test insect in the present laboratory experiments to explore the response of this species to heat. As can be seen in Table 5, the dates were naturally infested by three nitidulid species, and *C. mutilatus* was the most numerous followed by *C. hemipterus*, although this was not always the case. The intensity of infestation by a specific species may vary during the season and is influenced by the location of the date grove. *Lasioderma serricorne*, the tobacco beetle, is known to infest dried fruit and also to initiate infestations in the field. The records of low infestations of *Tribolium castaneum* and *Cryptolestes* sp. are particularly interesting in that these species are generally regarded as postharvest pests.

Although postharvest quarantine treatments using high temperatures have been studied on various commodities, this is the first study of dates to evaluate the emigration of nitidulid beetles from the fruit. Dates were used in this study but a range of commodities could be considered as targets where emigration of other insect pests would be beneficial. However, the effectiveness of heat treatment against other stored-product insects may vary considerably and a better knowledge of heat sensitivity of both the insects and the commodity is desirable. In this laboratory study a heating time of ~60 min was necessary before the temperature reached its target level. This heating time may also affect the emigration rate of the larvae. Respiration, as to be expected, is also affected by heat. As the body temperature of the insect increases, there is a concomitant increase in respiration up to an upper critical thermal limit. It has been suggested (5) that as the temperature rises above its optimum level and larval metabolism accelerates, the insect enters a condition of stress characterized by involuntary muscular contractions that cause the legless larva to leave its feeding site and eventually fall from the date. We found that 50°C produced the highest disinfestation level (92.3%) whereas at 55°C a disinfestation level of only 75% was reached, possibly because at that higher temperature mortality is more rapid and the insects die before they are able to escape from the dates. These effects of heat on the nervous and endocrine systems are areas where elevated temperature damage needs further investigation. Among the most studied responses of insects to heat is the elicitation of heat-shock proteins. The impact of these proteins on thermo-tolerance also requires study.

Considering that, until now, most dates, including the favored Madjoul variety, are first disinfested by MB fumigation and then dried when necessary, it is expected that this treatment will be most suitable for dates that require drying before storage. The commercial scale trials showed that the use of heat for insect disinfestation and control was even more effective than the use of MB. Two critical points were identified at the bottom front and rear of the drying duct where disinfestation was not fully successful but those locations represent only a negligible proportion of the quantities of dates treated.

With regard to dates that are harvested dry from the date palm and are already at a moisture content suitable for storage (such as the Deglet-Nour variety), exposure to heat would last no more than 2 h after the dates have reached the desired temperature.

Since conventional drying temperatures for most date varieties are in the range of 45° to 55°C, application of heat appears to be an encouraging solution for the treatment of dates as a replacement for MB. This approach was shown to be feasible using an existing commercial drying installation, with no modification required.

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