Research Article

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Efficacy of hot water treatment for postharvest control of western flower thrips, *Frankliniella occidentalis*, in French beans

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Abstract

Background: The western flower thrips, *Frankliniella occidentalis*, is a quarantine pest of French beans that requires phytosanitary treatment to meet quarantine requirements for strict lucrative markets. In this study, the efficacy of hot water treatment against *F. occidentalis* eggs and its effects on the postharvest physicochemical quality parameters of French beans was evaluated.

Results: The immersion time of 8.01 min (95% critical limits CL 7.77–8.24) was predicted by the probit model as the minimum time required to achieve a 99.9968% control level. Confirmatory tests with a large number of *F. occidentalis* eggs were performed to validate the estimated time to achieve probit-9 control level, and there were no survivors from the 50 103 eggs treated. Likewise, none of the 55 364 eggs exposed to 45 ± 0.2 °C for 7 min (observational time) survived. The effect of the treatment schedule on French beans quality parameters was assessed and there were no differences in weight loss, moisture content, total soluble solids, titratable acidity, pH, and reducing sugars between treated and untreated samples.

Conclusion: Our results indicate that hot water treatment (at 45 ± 0.2 °C for a duration of 8.01 min is an effective phytosanitary treatment for the control of *Frankliniella occidentalis* on French beans, with no significant impact on pods quality.

Keywords: phytosanitary treatment; egg disinfestation; thripidae; *Phaseolus vulgaris*; physicochemical parameters

1 INTRODUCTION

French beans, *Phaseolus vulgaris* L. (Fabales: Fabaceae), also known as snap or green beans, and locally in Kenya as '*mishiri*', is a commercially important horticultural crop. It approximately constitutes 20% by volume and 10% by value of all fresh horticultural exports in Kenya and ranks second only to cut flowers.[1,2] Despite the importance of French beans, their production and trade by smallholder farmers are continually hampered by myriad phytosanitary and technical quality challenges in sub-Saharan African, primarily the damage and presence of invasive pests such as thrips in the fresh produce, especially when the produce is for export.[3]

Western flower thrips (WFT), *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae), is one of the most important agricultural invasive pests of highly valued agronomic and horticultural crops, including tomatoes, capsicums, cucumbers and French beans.[4–7] This pestiferous and ubiquitous thrips species is indigenous to the southwestern states of the USA but has since spread around the world, including East Africa.[8] The rapid global dispersal, colonization and invasiveness of *F. occidentalis* are mainly attributed to the international trade of fresh fruits and vegetables that harbour *F. occidentalis*.[8] In addition, *F. occidentalis* possesses traits such as a short generation cycle, cryptic behaviour and high reproductive capacity that influence its success in invasion.[7] Adult female *F. occidentalis* lay 20–40 eggs into plant parenchyma tissue of leaves, flower parts or fruits using a blade-like ovipositor to insert eggs. After hatching, two active larval-feeding stages and adults feed by piercing plant cells with their mouthparts and sucking out the contents, consequently causing extensive damage that leads to both qualitative and quantitative yield losses.[5,6,9] Indirectly, *F. occidentalis* can vector destructive plant pathogenic viruses, especially tospoviruses, including *Tomato spotted wilt orthotospovirus* and *Impatiens necrotic spot orthotospovirus*.[10]

For sustainable management of *F. occidentalis*, a major pest of many horticultural crops such as French bean in East Africa, sustainable preharvest control strategies have been developed and incorporated into integrated pest management (IPM) packages.[11–16] Several studies have evaluated the effectiveness of the thrips IPM components in Kenya and some of the options have been very effective with significant economic impacts when diligently and consistently applied.[17] However, due to limited adoption of sustainable pest management and lack of knowledge, especially among smallholder farmers, 100% efficacy is not always

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guaranteed and often the life stages of the pest find their way postharvest into the fresh produce for export.[13,17] Globally, *F. occidentalis* is an important crop pest and is included in the quarantine pest lists of many regional plant protection organizations. For instance, *F. occidentalis* is species no. 177 on the list of A2 pests regulated as quarantine pests in the European Plant Protection Organisation (EPPO) region.[18] Therefore, the EU and other export markets have implemented stringent quarantine restrictions for invasive pest species. Fresh agricultural produce being imported into each country is meticulously monitored and quarantined to isolate and eliminate the accidental introduction of invasive insects in pest-free regions. Such introductions cause significant crop loss and economic hardship due to the need for increased pest control.[19,20] National Plant Protection Organisations (NPPOs) ensure that fresh produce destined for export is subjected to a standard postharvest phytosanitary inspection and treatment against *F. occidentalis* in the country of origin before it is cleared for export.[20]

There are numerous postharvest phytosanitary treatments for quarantine control of *F. occidentalis* that have been developed and commercially adopted or are at different stages of adoption. These include irradiation, low oxygen treatments, heat/cold treatments and chemical fumigations using methyl bromide, sulfuryl fluoride, phosphine, ethanedinitrile, methyl benzoate and nitric oxide.[21–25] The efficacy and acceptability of some of these methods are challenged by their ozone depletion potential, unacceptable phytotoxic effects on some fresh products, high levels of residue on fresh commodities, pesticide tolerance/resistance and/or ineffectiveness resulting from complex fumigation procedures.[26–31] Compared to these techniques, the hot water treatment (HWT) technique offers the best non-chemical alternative since it is reliable, efficient, viable and environmentally friendly.[32,33] The postharvest HWT technique is a suitable treatment for the control of other pests of quarantine importance such as codling moths and fruit flies in fresh produce.[34–39] Similarly, HWT was recently considered as a potential phytosanitary treatment for postharvest control of *F. occidentalis* in French beans.[40]

The International Standards for Phytosanitary Measures (ISPM) require that postharvest treatments are effective and the level of efficacy quantified statistically.[41] Often probit-9 is the benchmark for the efficacy of any phytosanitary treatment and this requires that 99.9968% mortality of the target pest in fruits or vegetables be demonstrated.[42–44] However, until now there has been a lack of a non-chemical alternative HWT that guarantees 99.9968% efficacy against *F. occidentalis* in French beans. Conventional commercial postharvest processing of French beans results only in the removal of adult and larval stages of thrips. However, the egg stage of the pest, which is hidden within the bean tissue presents the highest risk for the spread of thrips into export markets.[7,36] Thus, our main objective was to develop optimal hot water disinfestation treatment schedule for *F. occidentalis* eggs in the French bean crop that is widely grown in Kenya for export.[3] Herein, we determined the development rate of eggs in the French beans, described the heat tolerance of the egg stage and estimated the minimum time required to achieve 99.9968% mortality of *F. occidentalis* eggs. Furthermore, we validated the estimated time with a large sample size of thrips eggs and finally evaluated the impact of the HWT schedule on the physicochemical quality parameters of French beans. The development of a non-chemical postharvest quarantine treatment protocol for French beans and the subsequent adoption will guarantee *F. occidentalis*-free French beans for high-end export markets globally.

2 MATERIALS AND METHODS

2.1 Thrips colony rearing

Field-collected *F. occidentalis* were reared on French bean pods and maintained in ventilated plastic jars (17 cm in height, 8 cm diameter) at 25 ± 1 °C, 60 ± 10% relative humidity (RH) and 12 h light:12 h dark (12L:12D) photoperiod, at the Animal Rearing and Containment Unit (ARCU) of the International Centre of Insect Physiology and Ecology (*icipe*), Nairobi, Kenya as described by Nyasani *et al.*[45] The laboratory-reared adult thrips used in our experiments had been maintained for more than 30 generations in the laboratory with a 3-month interval infusion of field-collected thrips during the 1 year of this experiment, to keep the original behavioural characteristics of the species.

2.2 Experimental French beans

French bean samples were obtained from the counties of Kirinyaga [00°36′12.5″ S, 037°22′11.6″ E, 1219 m above sea level (asl)], Machakos (01°14′17.8″ S, 037°27′51.1″ E, 1189 m asl) and Murang'a (00°48′55.8″ S, 037°14′48.3″ E, 1138 m asl). These are among the leading French bean producing counties in Kenya.[2] The pods were selected based on commercial maturity indices such as colour, firmness, shape and size.[46,47] Only pods measuring between 4 and 9 mm in diameter and 10–15 cm long, free of diseases, pests and injuries were used in the experiments. The freshly harvested French bean pods were treated within 24 h postharvest.

2.3 Hot water treatment tank

A 1600-L double-walled stainless-steel hot water treatment tank (Desbro Engineering Ltd, Nairobi, Kenya) with tap water (pH 7.5 ± 1) was used in the experiment. The tank was connected to a Grant Squirrel data logger (SQ2020-2F8) with 16 thermocouple probes (Tempcon Instrumentation Ltd, Arundel, UK) to give temperature recordings throughout the treatment (Fig. 1) as described by Mwando *et al.*[39]

2.4 Determination of the development rate of *F. occidentalis* eggs in French beans

The development rate of *F. occidentalis* is temperature and host-dependent. We therefore sought to determine the development of eggs of *F. occidentalis* in French beans under ambient temperature (25 ± 1 °C). Briefly, a batch of 120 freshly harvested French bean pods, *P. vulgaris*, was surface disinfected to remove any fungal spores using 0.5% sodium hypochlorite (NaClO) and rinsed with tap water. They were then divided into six groups of 20 each for easy handling. Each group of 20 pods was placed in a plastic jar containing approximately 200–300 male and female *F. occidentalis* adults (1♂:5♀) aged between 2 and 5 days. The thrips were allowed 12 h to lay eggs on the bean pods. Finally, the pods were removed from the jar containing adult thrips and then incubated at 25 ± 1 °C, 60 ± 10% RH and a photoperiod of 12L:12D. Observation on the larval emergence was executed each day for six consecutive days from 20 French beans pods picked at random from the 120. The pods were dissected under a stereomicroscope with LED and HD Camera Leica EZ4 HD (Leica Microsystems, Heerbrugg, Switzerland) after staining according to Nyasani *et al.*[45] for easy visualization of unhatched eggs (Fig. 2(B)). All eggs of *F. occidentalis* that were found were recorded. The developmental times of the egg to the first instar were determined

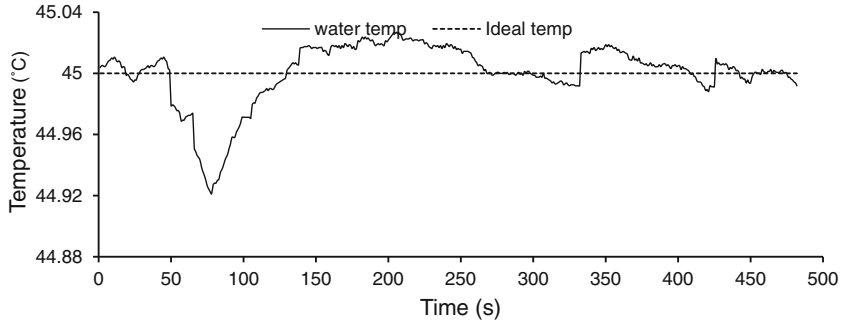

Figure 1. Mean temperature of water in the hot water immersion tank recorded every second during the treatment process by a Grant Squirrel data logger connected to the tank.

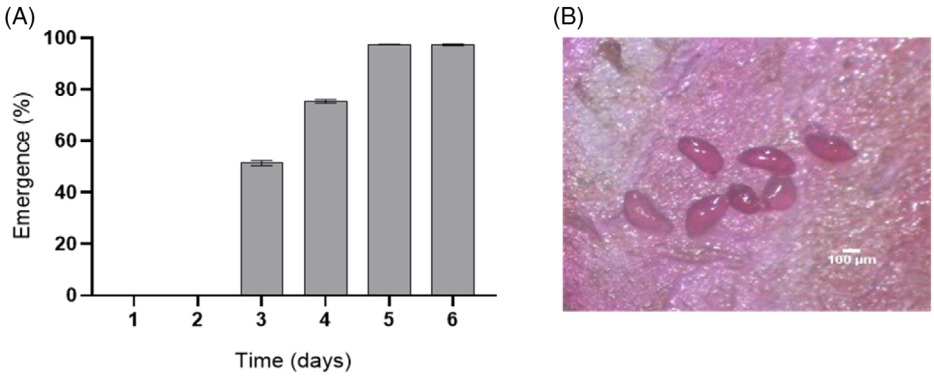

Figure 2. Hatchability of *Frankliniella occidentalis* eggs in French beans over time (days) under ambient conditions (25 ± 1 °C and 60 ± 10% relative humidity) (A) and the image of stained eggs in the pod (B).

using the morphological features described in Reitz[5] and CABI.[8] Our focus was on the egg (opaque, reniform/kidney-shaped and about 200 µm long, inserted into the epidermis and mesophyll layer of host plant) and first-instar larva (spindle-shaped and creamy-white to yellow).

2.5 Determination of the natural mortality of *F. occidentalis* eggs in French beans

Twenty-five pods were prepared, infested and incubated (25 ± 1 °C and 60 ± 10% RH) as above. After 5 days, the number of emerged larvae was recorded. They were then removed from the pod using a soft camel-hair brush and each pod was stained and dissected under the microscope to count the number of unhatched eggs. The experiment was replicated five times. This data was eventually used to correct for control mortality in the subsequent experiment using Abbott's formula.[48]

2.6 Determination of the lethal time of *F. occidentalis* eggs in French beans

French beans infested as described above were held under the same conditions for 12–24 h before the hot water treatment. To confirm the number of eggs in treated infested pods, an equal number of pods (control) infested in the same way were incubated for 4 days then first-instar larvae were counted on the fifth day. Groups of 25 pods were picked at random from the holding jars and placed in each of seven perforated stainless-steel crates measuring 57 × 40 × 23 cm (Desbro Engineering Ltd, Kenya) which allowed the hot water to circulate freely around the pods.

The experiment was replicated 30 times and a total of 5250 French bean pods were treated (175 pods per replicate). An equal number of infested pods were set aside as control for use as an estimation of the number of eggs in the treated pods. The timing of the HWT commenced when all the pods in a treatment batch were completely submerged in the hot water treatment tank[39] containing water already at 45 ± 0.2 °C. The choice of the water temperature was based on pilot experiments conducted to determine the temperature that does not result in the scalding of pods post-treatment. The period between loading all the seven crates was approximately 30 s. The infested pods were immersed in the water at 45 ± 0.2 °C for 1, 2, 3, 4, 5, 6 and 7 min and temperature recordings were taken throughout the treatment (Fig. 1). After treatment, pods were stored under the pretreatment ambient room conditions (25 ± 1 °C and 60 ± 10% RH) to allow them to dry and cool naturally. On day 5 the numbers of first-instar larvae in treated and untreated samples were recorded. Time–mortality relationships were established and the probit-9 quarantine security level was estimated.[43]

2.7 Large-scale confirmatory test

French bean pods were infested as above and validation of the 99.9968% quarantine security level was carried out at 45 ± 0.2 °C for 8.01 min, i.e. the time that is theoretically required to kill 99.9968% of eggs of *F. occidentalis* in French beans. The experiment was replicated 15 times and a total of 1875 French beans pods infested with the eggs were used, and an equal number were set aside as an untreated control. Thereafter, the pods were carefully examined and the number of emerged larvae was determined. In addition, we also investigated the efficacy of HWT at 45 ± 0.2 °C for 7 min, which was the observational time at which complete egg mortality was obtained. For this validation,

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1875 pods were infested and treated while the same number of pods was infested but not subjected to hot water treatment (control). The experiment was also replicated 15 times. The assumption was that in each test, the number of eggs in each treatment and the control was not significantly different and therefore egg mortality was calculated based on the number of larvae counted on the pods in the treatment as compared with the larvae counted on the pods in the control.

2.8 Assessment of the impact of the HWT schedule on quality parameters

French beans pods (approx. 1000) were harvested at the mature-green stage and carefully selected based on uniformity of colour, firmness and size, and randomly divided into two groups. The first group (approx. 500) was subjected to a HWT for 8.01 min at 45 ± 0.2 °C as described above. The other group was held untreated at ambient conditions (25 ± 1°C). Pods were randomly picked from each batch and subjected to several tests to assess the impact of the treatment protocol on weight loss, moisture content, total soluble solids, titratable acidity, pH and reducing sugar, from the first day and every second day for 11 days, following the methodology described in AOAC,[49] with slight modifications. The experiment was repeated three times.

Briefly, the weight loss rate was determined using a digital scale (Ohaus CS2000; Melrose, MA, USA) by weighing a composite of 30 pods before and after treatment, and the results of the cumulative weight loss were expressed as a percentage. The pH of the French beans was measured by a digital pH meter (Orion 5 Star; Thermo Scientific, Waltham, MA, USA) at ambient temperature using extract directly from the pods. Total soluble solids (TSS) was determined from the samples that were homogenized in a blender, thoroughly mixed and filtered through layers of cheesecloth using a manual juicer, then a drop was used to measure direct °Brix content using a digital refractometer (Atago PR-101a; Cole-Parmer/Antylia Scientific, Vernon Hills, USA). Titratable acidity (TA) was measured in the pods through titration of an aliquot of 10 mL of the filtered extract against 0.1 N sodium hydroxide (NaOH) using phenolphthalein at 1% as an indicator. The results were expressed as a percentage. The moisture content of the pods was determined by first recording the initial weight of the pureed sample preweighed aluminium dish. The samples were then dried in an oven (Nabertherm, RT-120; Lilienthal, Germany) overnight at 105 °C and weighed again. The moisture content was expressed as a percentage. Reducing sugars content was quantified using the Lane–Eynon titration method. A test sample (5 g) was dissolved in 100 mL warm distilled water. The filtrate was transferred into a 50-mL burette, then titrated with 10.0 mL

Table 1. Natural mortality of *Frankiniella occidentalis* eggs in French bean pods (25 ± 1 °C and 60 ± 10% relative humidity)

Replicate	No. of individuals	No. hatched	No. dead	% Mortality
1	1009	980	29	2.87
2	899	871	28	3.11
3	905	879	26	2.87
4	873	849	24	2.75
5	889	862	27	3.04
Total	4575	4441	134	2.93 ± 0.065

Table 2. Time–mortality relationship for the egg stage of *Frankiniella occidentalis* in French beans after immersion in hot water at 45 ± 0.2 °C

Stage	Time (min)	No. alive	No. dead	% Mortality
Egg	1	20 202	8433	41.74
	2	19 865	17 484	88.01
	3	20 036	18 839	94.03
	4	20 363	19 478	95.65
	5	20 568	19 909	96.80
	6	19 305	19 066	98.76
	7	19 525	19 525	100.00

Table 3. Mortality of egg stage of *Frankiniella occidentalis* in French beans subjected to hot water at 45 ± 0.2 °C for 8.01 min

Replicate	No. treated	No. dead	No. alive	% Mortality
1	3720	3720	0	100
2	3373	3373	0	100
3	2648	2648	0	100
4	3024	3024	0	100
5	3550	3550	0	100
6	2641	2641	0	100
7	2674	2674	0	100
8	3697	3697	0	100
9	4307	4307	0	100
10	2769	2769	0	100
11	4399	4399	0	100
12	3297	3297	0	100
13	3543	3543	0	100
14	3205	3205	0	100
15	3256	3256	0	100
Total	50 103	50 103	0	100

Table 4. Mortality of egg stage of *Frankiniella occidentalis* in French beans subjected to hot water at 45 ± 0.2 °C for 7 min

Replicate	No. treated	No. dead	No. alive	% Mortality
1	4497	4497	0	100
2	4306	4306	0	100
3	4439	4439	0	100
4	3703	3703	0	100
5	3421	3421	0	100
6	3044	3044	0	100
7	4238	4238	0	100
8	3335	3335	0	100
9	4542	4542	0	100
10	3014	3014	0	100
11	2892	2892	0	100
12	3037	3037	0	100
13	3340	3340	0	100
14	3610	3610	0	100
15	3946	3946	0	100
Total	55 364	55 364	0	100

of boiling Fehling's solution (5.0 mL of Fehring's Solution A and 5 mL of Fehring's Solution B) and 50 g of NaOH in distilled water, diluted to 500 mL, kept for 2 days and filtered through a 0.45 µm filter paper. The endpoint was determined by mixing 1% methylene blue until the blue colour of the indicator disappeared to a brick red endpoint. Titration was completed within 3 min and blank titration was performed without hydrolysing the sucrose sample. Titration was performed in triplicate and the average value was taken.

2.9 Data analysis

The data on the development rate of *F. occidentalis* in French beans were scored as a percentage. To estimate the minimum lethal time required to attain different levels of efficacy, data from the heat tolerance of the egg were first corrected for control mortality[48] and then analysed using the generalized linear model of regression with a probit function (dose.p function from MASS library). The validation phase data were expressed as % mortality. Data on the physicochemical quality parameters of French beans were analysed by *t*-test after being subjected to the Shapiro normality test. All analyses were performed using R software version 4.0.0.[50]

3 RESULTS

3.1 Development rate of *F. occidentalis* in French beans

Based on the morphological characteristics described in the Material and Methods section 2.4, on days 1 and 2 after oviposition only eggs were present. The eggs started hatching by day 3 and by day 5 more than 99% of the larvae were in the first instar (Fig. 2(A)). The average natural egg mortality was 2.93 ± 0.065% (Table 1).

3.2 Heat tolerance of *F. occidentalis* eggs

The mortality of all of *F. occidentalis* eggs increased as treatment time increased, with 100% mortality of all eggs recorded after 7 min of hot water treatment at 45 ± 0.2 °C (Table 2). Based on time–mortality relationships, the estimated minimum time (using probit analysis) required to attain 50%, 90%, 99%,

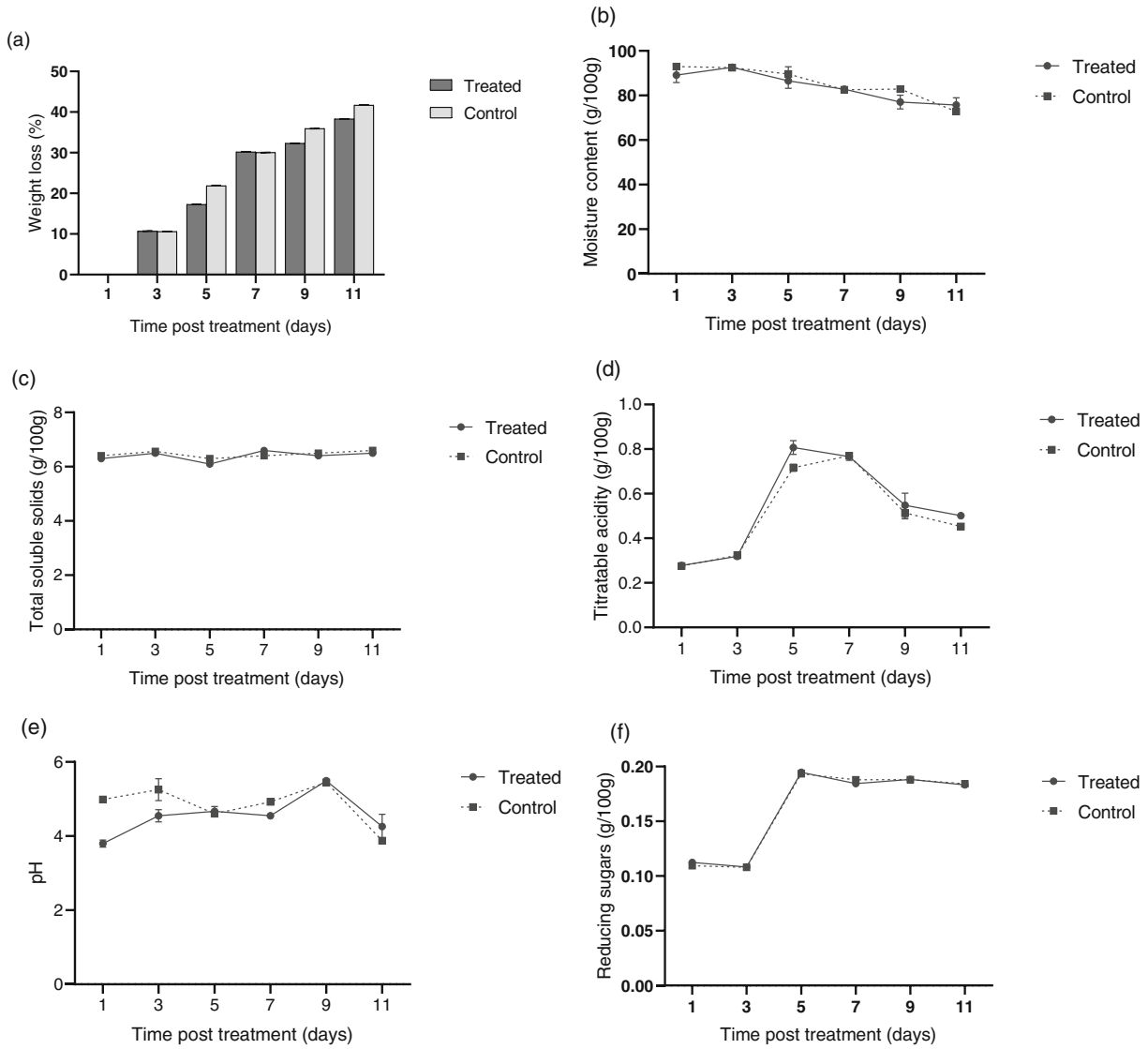

Figure 3. Changes in French bean weight loss (a), moisture content (b), total soluble solids (c), titratable acidity (d), pH (e) and reducing sugars (f) 11 days post hot water treatment.

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99.9%, 99.99% and 99.9968% mortality was found to be 0.81 (range 0.74–0.89 min), 3.12 (range 3.06–3.18 min), 5.00 (range 4.88–5.12 min), 6.37 (range 6.20–6.54 min), 7.50 (range 7.29–7.72 min) and 8.01 (range 7.77–8.24 min), respectively.

3.3 Large-scale confirmatory test

Based on these results, large-scale validation experiments were carried out on French bean pods infested with eggs and treated for 8.01 and 7 min in 45 ± 0.2 °C water. In the large-scale validation trials there were no survivors from 50 103 and 55 364 *F. occidentalis* eggs treated at 45 ± 0.2 °C for 8.01 or 7 min, respectively (Tables 3 and 4).

3.4 Change in French bean physicochemical quality parameters after HWT

After treatment, the quality of the French bean pods was assessed. Basically, there were no visual signs of heat injuries or acceleration in skin colour development detectable in either untreated or heat-treated pods over the 11 days post-treatment. There were no significant differences between the treated and untreated French bean pods recorded in weight loss ($t = -0.216$, df = 9.941, $P = 0.834$), moisture content ($t = 0.38$, df = 6.779, $P = 0.715$), total soluble solids ($t = -0.756$, df = 8.788, $P = 0.47$), titratable acidity ($t = 0.228$, df = 9.927, $P = 0.824$), pH ($t = -1.322$, df = 9.999, $P = 0.216$) and reducing sugars ($t = 0.003$, df = 9.992, $P = 0.998$) 11 days post-HWT (Fig. 3(a)–(f)).

4 DISCUSSION

Postharvest disinfestation treatments are imperative to prevent the introduction and spread of agricultural pests to new regions through movement of fresh fruits and vegetables during international trade.[19,20] The greatest challenge, however, is the development of a safe and environmentally friendly treatment protocol that will serve as a viable alternative to commonly used chemical fumigations. Previous life-history parameter studies have investigated the development and reproduction of *F. occidentalis* on many different plants, including beans,[51,52] *Chrysanthemum*,[53] cotton,[54] cucumber[55] and peanut.[56] The performance, including developmental times of *F. occidentalis*, depends both on temperature and the host plant on which the thrips are feeding.[57–59] The incubation period of the egg stage of *F. occidentalis* in vegetables, including French beans, at ambient conditions (25 ± 1 °C) of 3–4 days reported in our study is comparable to results reported in previous studies. Zhi *et al*.[60] reported the egg incubation time for this pest on bean pods of 3 days at 24 ± 1 °C, 36 ± 8% RH. Similarly, thrips reared individually on bean leaves at 24–25 °C and 16L:8D had an egg incubation period of 2–4 days.[51,52] In the present study, infested French beans pods were subjected to hot water treatment 12–24 h after exposure to *F. occidentalis*.

Temperature is one of the major abiotic factors that affects the performance of insects, including their development and survival.[5,61–64] Equally, adult and immature life stages of thrips in different crop hosts or even varieties are known to respond differently when subjected to heat.[65] For instance, immersion of *Dendrobium* orchid inflorescences in water at 49.5 °C for 15 and 20 s reduced the mean number of adult thrips per blossom by 88.1% and 95.3%, respectively. Studies by Hara *et al*.[66] established that hot water dips of propagative material of *Anthurium* at 49 °C for 10 min effectively disinfect some *Anthurium* cultivars of thrips before planting. Similarly, a HWT of 50 °C was able to kill all thrips life stages on peaches and nectarines in 1 and 2 min, respectively.[67] In this study, 99.9968% mortality corresponding to probit-9 for the egg stage of *F. occidentalis* in French beans was achieved at 8.01 min (95% CL 7.77–8.24 min) with a water temperature of 45 °C. However, 100% mortality was demonstrated already after 7 min at 45 °C, 1 min before the estimated efficacy time. The lower limit of the confidence interval (7.77 min) is higher than the tested range of 1–7 min by 0.77 min, which translates to 46.2 s. Although expectations would have required that the estimate falls within the tested range, this is often not the case in most cases as even going back to the drawing board to test higher durations gives similar results, not only in our case but in various studies.[37,68,69] These differences are expected because several studies have reported that modelling mortality response data sometimes results in overestimating the dose above that obtained experimentally.[70] Thus, we suppose the use of confidence intervals becomes necessary when choosing an effective treatment time period. We report effective treatment times of 7 and 8.01 min, which could be adopted for French beans of 4–9 mm diameter, 10–15 cm long, which is within the basic range of category preferred for export.[20,46,47] Both times were tested in large-scale validation and resulted in no pest survivors as required in phytosanitary treatments. In the interest of saving cost, 7 min would be ideal for adoption in large-scale commercial treatments.

The heat susceptibility of *F. occidentalis* may vary according to the environment that the insects are initially exposed to., for example laboratory-reared insects may respond to heat differently compared to their wild type counterparts.[65,71] Laboratory-reared insects often exhibit inbreeding leading to genetic drift compared to their wild counterparts, thus allelic diversity and heterozygosity are often lost.[72] However, periodic outcrossing with insects collected from the wild usually is adequate to restore genetic and phenotypic deviations, especially in non-modified insect strains. In our case we used laboratory-reared *F. occidentalis* which had been kept in the rearing facility for 30 generations, a period which is relatively inadequate to produce highly felt genetic modification. Furthermore, in the current experiment, we infused the laboratory colony with field-collected thrips at 3-month intervals. This is a standard practice which ensures that the effects of selection and genetic drift are minimized and genetic vigour is enhanced. We therefore do not expect a marked difference in the response of the insects used in our study compared to their wild counterparts.

Previous studies on disinfestation of thrips in fresh produce using hot water treatments largely focused on the larval and adult life stages of the pest.[66,67] Conversely, *F. occidentalis* eggs are laid concealed within plant tissue, including the French bean pod. This presents the highest risk for the spread of thrips into other destinations unnoticed, since casual inspection may not reveal the presence of thrips eggs. In addition, conventional processing and chemical fumigants may be ineffective because chemicals fail to contact the hidden thrips eggs. Thus, if an infestation of fruits and vegetables occurs shortly before harvest, chances are high that the infected fresh produce could be packed for export, posing a risk of phytosanitary concern for certain import markets where the climate may be conducive for the survival of the pest. The hot water dips have been shown to be more effective against cryptic insect pests than chemical pesticides because heat penetrates the plant tissue to reach the pest.[73]

Studies by Speckhahn *et al*.[40] focused on the disinfestation of French beans against *F. occidentalis* eggs and they observed that treatment of French bean pods with hot water at 50 °C for at least

5 min resulted in 100% mortality of thrips eggs. Pilot studies in the present study worked on temperatures below 50 °C as used in Speckhahn *et al.*[40] and concluded that a temperature of 45 °C was ideal for the disinfestation of French beans without causing any damage to the pods. Their experiments were conducted in a smaller tank (25 L) with one thermos-regulator, thus temperature control may have been inadequate as compared to the current work, which had multiple sensors and adequate heating as explained in section 2.3. The ISPM no. 28 requires that fresh fruits and vegetables meant for export be subjected to phytosanitary treatments that are effective and the level of efficacy quantified statistically.[19,74] Speckhahn *et al.*[40] did not statistically demonstrate whether probit-8.72 or probit-9 efficacy levels, which are widely adopted in various treatments, were attained in their study.[19,20,44] Similarly, the study does not indicate the number of individuals that were treated. The current study has demonstrated how the probit-9 efficacy level was achieved after subjecting a total of 5250 *F. occidentalis*-infested French beans pods to a water temperature of 45 °C for seven different times. Additionally, during the efficacy determination, a total of 180 262 eggs were treated.

Postharvest heat treatments may influence phytochemical changes in fresh produce after storage and ultimately their marketing.[75] However, this depends on several factors, including temperature/duration combination, fresh produce variety, maturity stage at harvest and other preharvest treatments.[76] In the current study, HWT of 45 °C for 8.01 min did not influence the loss of weight and moisture in the treated French bean samples 11 days post-treatment. These results are comparable with the findings of other studies. Ismail *et al.*[77] reported a similar observation in green beans subjected to HWT of 45 °C for 15 min. The loss of moisture content and subsequent weight loss is a common phenomenon in fresh produce under storage, albeit influenced by storage conditions.[78] This is due to the high rate of respiration and heat emission in green beans. Similarly, HWT did not have a significant impact on the pH change 11 days post-treatment. Nevertheless, a temperature effect was noted 1-day post-treatment, which diminished with time. The results corroborate the findings of Khalil *et al.*,[79] who reported that HWT did not cause significant changes in the pH levels in peach fruit 6 days post-treatment. The lack of significant changes in total soluble solids and titratable acidity observed in this study 11 days after treatment is comparable to the findings of other studies. There were no differences between green beans subjected to HWT (45 °C for 15 min) and control samples 21 days post-treatment.[77] Majomot *et al.*[80] observed no difference in change in TSS between samples pepper-sprayed with warm water (55 °C) and control samples at ambient conditions. Similarly, there was no difference in the levels of TA between hot water treated green beans and control samples 14 days post-treatment.[77] In the present study, HWT did not influence the change in reducing sugars in French beans. Conversely, the content of reducing sugars was reduced after green beans were subjected to HWT of 45 °C for 15 min in Ismail *et al.*[77] This difference could be due to differences in exposure time.

5 CONCLUSIONS

Postharvest control of insects such as *F. occidentalis* in harvested French beans is critical to preserve product quality and market value, and to meet phytosanitary requirements for export markets. The global phase-out of several chemical fumigants, such as methyl bromide, necessitates alternative treatment for postharvest pest control. Hot water treatment has shown strong efficacy against a wide range of postharvest insect pests of fresh fruits and vegetables. Our results showed that at 45 °C, the minimum time required for 99.9968% quarantine security level, which corresponds to probit-9 at the 95% confidence level, in French beans is 8.01 min. We also observed that a shorter exposure time of 7 min at 45 °C was sufficient to result in no survivors in 55 364 tested individual eggs. We therefore recommend the adoption of both 7 and 8.01 min as effective durations which ensure freedom of French beans from thrips. However, 7 min may be favoured to reduce cost at a commercial level without any detrimental effect on treatment results. The present study has established that HWT is effective against *F. occidentalis* for the tested temperature–time treatment regime and had no adverse effect on French bean quality parameters such as weight loss, moisture content, pH, total soluble solids, reducing sugars and titratable acidity. This demonstrates that HWT is a promising novel alternative non-chemical treatment for postharvest thrips control. More research and development efforts are necessary to accelerate its commercial applications, including the development of safe and effective treatment protocols against a wide range of thrips species on French beans and other different fresh produce infested in the field.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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