

Wax Removal and Diamondback Moth Performance in Collards Cultivars

GA SILVA^{1,2}, RM PEREIRA¹, N RODRIGUES-SILVA¹, TC SOUZA¹, DO FERREIRA¹, EA QUEIROZ¹, GAR SILVA¹, MC PICANÇO¹

¹Lab Entomologia Agrícola, Depto de Entomologia, Univ Federal de Viçosa, Viçosa, Minas Gerais, Brazil

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Correspondence

G.A. Silva, Lab Entomologia e Fitopatologia, Centro de Ciências e Tecnologias Agropecuárias, UENF, Campos dos Goytacazes, RJ 28013-602, Brazil; gasilva@pq.uenf.br

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Abstract

The diamondback moth *Plutella xylostella* (Linnaeus, 1758) (Lepidoptera: Plutellidae) is an herbivorous specialist on Brassicaceae species. *Brassicaceae* spp. plants developed a range of defenses (chemical, physical, and morphological) to prevent herbivores attack. In this study, we reported the antixenotic and antibiotic effects of outermost layer of two species of epicuticular wax of Brassicaceae, *Brassica oleracea* L. var. "Santo Antônio," and Hybrid Kope F1 100MX, on larvae and adult of *P. xylostella*. In the choice experiment, *P. xylostella* adults showed an oviposition preference for collard cultivars Santo Antônio (control) and Hybrid Kope F1 100MX with wax removal. In the no-choice experiment, oviposition was 6.4 times higher in the Hybrid Kope F1 100MX with wax removal than without wax removal. There were significant differences among larvae feeding on leaf disks of Hybrid Kope F1 100MX in the treatments with (65.3 mg) and without wax removal (23.5 mg). The net reproduction rate (R_0), and intrinsic (rm) and finite rates of increase (λ) of *P. xylostella* in the cv. Santo Antônio were bigger in the treatment without wax removal ($R_0 = 50.4$, $rm = 0.23$ and $\lambda = 1.26$) than treatment with wax removal ($R_0 = 28.5$, $rm = 0.20$ and $\lambda = 1.22$). However, only the R_0 value was affected by mechanical wax removal in the Hybrid Kope F1 100MX (with wax removal $R_0 = 43.3$ and without wax removal $R_0 = 30.8$). In conclusion, the results indicate that collard's wax is important to accessibility and development of *P. xylostella*, and its removal changes the resistance of collard's varieties to *P. xylostella*.

Introduction

The diamondback moth *Plutella xylostella* (Linnaeus, 1758) (Lepidoptera: Plutellidae) is an oligophagous pest that specializes in consumption of cultivated and uncultivated plants of the family Brassicaceae (Baek *et al* 2005, Asghari *et al* 2009, Rondelli *et al* 2011). *P. xylostella* has developed resistance to chemical and microbial insecticides (Schuler *et al* 1998, Baek *et al* 2005, Ferré *et al* 1991, Wang *et al* 2015), and alternative strategies to control this insect are required. The knowledge of plant chemicals that plays a role in acceptance or rejection of

potential host plants by insects represents, one, the first step to develop new control strategies of this insect.

Plant surface attributes are expected to be important factors influencing host selection by phytophagous insects (Chapman 1977). Several studies show that *P. xylostella* locate the sites of feeding and oviposition through olfactory, gustatory, and tactile senses triggered by compounds produced in host plants (Bukovinszky *et al* 2005, Hopkins *et al* 2009, Fathi *et al* 2011, Badenes-Perez *et al* 2014). On the other hand, some components of plants' defense system have a direct action on development, fecundity, survival,

and behavior of phytophagous insects (Awmack & Leather 2002, Hasan & Ansari 2011). This defense compounds may reduce the viability and the resource location for eggs, prolong the development time of larvae, reduce weight and viability of pupa, and reduce fecundity and fertility of adults (Müller et al 2010).

Glucosinolates are secondary metabolites found in Brassicaceae and related families with defensive functions against the attack of phytophagous insects (Halkier & Gershenzon 2006, Hopkins et al 2009). Glucosinolates are biosynthesized from several amino acids and different metabolic processes (Kliebenstein et al 2005, Grubb & Steffen, 2006, Ishida et al 2014). Glucosinolates are activated by myrosinases; however, these compounds are stored in different compartments of plant tissue (Koroleva et al 2000, Husebye et al 2002). When the plants are damaged by phytophagous insects, these compounds are converted by plants into toxic forms to insects (Städler 2002, Sun et al 2009). However, *P. xylostella* uses the glucosinolates as cues for the location and recognition of *Brassica* species. Furthermore, previous studies have shown that application of glucosinolates in substrates and on non-host plants stimulates oviposition of *P. xylostella* (Spencer et al 1999, Renwick et al 2006).

The glucosinolates, alkenes, paraffin, and saponins are the main constituents of waxes from the leaves of *Brassica* species (Spencer et al 1999, Renwick et al 2006, Badenes-Perez et al 2014). The composition and quantity of wax vary between cultivars, and they can influence the resistance of *Brassica* species and cultivars against phytophagous insects. Ulmer et al (2002) and Fathi et al (2011) showed that cultivars of *Brassica oleraceae* with high wax content affected negatively the developmental time and survival of larvae and fecundity and choice of oviposition site of females of *P. xylostella*. In addition, mechanical removal of leaf waxes of their host plant affected the oviposition preference of insects (Müller & Hilker 2001). These evidences suggest that epicuticular waxes contain crucial information for host plant choice. However, details about the nature of such compounds and their importance to oviposition and fitness of *P. xylostella* should be better elucidated.

In this study, we aim to examine the influence of epicuticular wax on oviposition choice and fitness of *P. xylostella* on collard cultivars.

Material and Methods

Insect colony

Adults and larvae of *P. xylostella* used in the bioassays were obtained from an insect colony maintained in the Laboratory of Pest Management at the Federal University of Viçosa (UFV). The original population was collected from cabbage

crops in the field experimental station of the University of Viçosa. The colony was maintained at $27 \pm 0.5^\circ\text{C}$, relative air humidity of $75 \pm 5\%$, and a photoperiod of 12 h in four wooden cages ($40 \times 40 \times 40$ cm) covered with organza: one cage for oviposition, one with first instar larvae, one with larger larvae (second, third, and fourth instar), and one for pupation and adult emergence (Bacci et al 2009, Silva et al 2011). The larvae were fed with collard leaves of cv. Manteiga Híbrida grown in greenhouse without insecticide residues.

Plants of collards

The two cultivars of collards (*Brassica oleracea* var. *acephala*) used in the experiment were the cvs. Santo Antônio (SA) and Hybrid Kobe F1 1000MX (Hy). The cv. Santo Antônio has glossy leaves that are indicative of low wax production. The variety Hybrid Kobe F1 1000MX has non-glossy leaves. The variety cv. Santo Antônio was obtained from stem cuttings of plants from a germplasm bank of the Horticulture Department of UFV. The cv. Hybrid Kobe F1 1000MX was obtained through the sowing of seeds in styrofoam trays of 200 cell. The plants used in the bioassays were transplanted to pots of 5 L. Pots were filled with substrate in the proportion of two-thirds of clay and one third of cattle dung. The fertilization and the cultural practices were as recommended to collards (Filgueira 2008). The plants were maintained without application of insecticides.

Methods

The oviposition preference of *P. xylostella* was verified through two experiments (choice and no-choice). In the experiments with choice, four treatments were set as follows: (1) leaves of cv. Santo Antônio used as control (SA), (2) leaves of cv. Hybrid Kobe F1 1000MX (Hy), (3) leaves of cv. Hybrid Kobe F1 1000MX with wax removal (HyWR), and (4) leaves of cv. Hybrid Kobe F1 1000MX with pin holes (HyD) to simulate the injury caused by wax removal of treatment 3. Hydrophilic cotton was moistened with chloroform to remove the wax on both sides of leaves in the treatment number 3.

In the experiment without choice, just leaves of cv. Hybrid Kobe F1 1000MX were used. The collards leaves were divided into two slices (left and right). On one half of the leaf, the wax was removed (HySWR) in the adaxial and abaxial faces, and the other half the wax was not removed (HySW).

The experimental design was in randomized blocks with five replication of each treatment; each replication consisted of a wooden cage with $100 \times 100 \times 80$ cm dimensions covered with organza. In each cage, 25 couples of adult *P. xylostella* (of 2 days old) were released and the number of eggs was counted after 24 h.

The effect of wax removal on foliar consumption by larvae was assessed by running four treatments. The first treatment

consisted of a disk (8 cm diameter) of collards leaves cv. Santo Antônio without the wax removal (SA), the second treatment consisted of disks of collards leaves cv. Santo Antônio with wax removal (SAWR), the third treatment consisted of the leaf disks of collards leaves cv. Hybrid Kobe F1 1000MX without wax removal (HyWR), and the fourth treatment was a disk of collards leaves of cv. Hybrid Kobe F1 1000MX with wax (Hy). The wax was removed using cotton moistened in chloroform. The disks were kept in plastic pots of 500-ml capacity, and then 10 larvae of *P. xylostella* of the third instar were placed in each pot which was kept in a rearing chamber at $27 \pm 0.5^\circ\text{C}$, $75 \pm 5\%$ relative humidity, and 12-h photoperiod. The larval instar was determined by observation of cephalic capsule width (Agerbirk *et al* 2003).

After 24 h, the larvae were removed and the leaf disks masses were measured in an analytical scale (precision of 0.0001 g, Sartorius BP 210D, Gottingen, Alemanha). The leaf disks were then packed in paper bags and dried in an oven with forced air circulation (Marconi, model MA037) at 60°C for 48 h. After drying, the disk masses of leaves were again measured for consumption estimation.

The moisture content calculated for infested disks (Eq. 1) was used to calculate the mass of disk leaf dry matter before the infestation. The calculation was done using Eq. 2:

$$\text{Moisture content (\%)}; \text{MC} = \left[\frac{Wi - Wid}{Wi} \right] \times 100 \quad (1)$$

$$\text{Dry mass consumed (mg)}; \text{DM} \left\{ \left[W - \left(\frac{W \times \text{Mc}}{100} \right) \right] - Wid \right\} \quad (2)$$

where Wi = weight of infested disks before drying, Wid = weight of disks after drying, W = weight of the disks before the infestation.

Biological life table

Life tables were constructed to verify the effect of wax removal in collards on the survival of *P. xylostella* larvae. The life table was built according to Southwood (1978) and Golizadeh *et al* (2009). Neonate larvae (age < 4 h old) were transferred to plastic pots of 500 ml containing a leaf disk of collards of one of four treatments: (1) leaves of collards cv. Santo Antônio (SA), (2) leaves of collards cv. Santo Antônio with wax removal (SAWR), (3) leaves of collards cv. Hybrid Kobe F1 1000MX (Hy), and (4) leaves of collards cv. Hybrid Kobe F1 1000MX with wax removal (HyWR). Each pot received 10 larvae.

The experimental design was completely randomized with 12 repetitions of each treatment. The pots were placed into a rearing chamber at $27 \pm 0.5^\circ\text{C}$, $75 \pm 5\%$ relative humidity, and

12-h photoperiod. When the disks began to change color, they were replaced by others of the same treatment. Larvae mortality was evaluated daily until they become pupae.

The pupae were weighed and sexed and those not giving rise to adults were considered dead by physiological disorders. Adults that emerged were transferred to cages ($40 \times 40 \times 40$ cm) to evaluate fecundity and were offered collard leaves of the same treatment as during the larval development.

Statistical analysis

The results of experiments where the larvae had a choice of leaves were subjected to analysis of variance (one-way ANOVA; PROC ANOVA; SAS System 2002), and averages were compared by Tukey's HSD test ($p < 0.05$). The consumption of leaf area, the weight of pupae, and the oviposition of adults were compared by paired *t* test ($p < 0.05$). The data fit the homogeneity of variance and normality of the errors; therefore, there was no need for the transformation of data. The emergence of adults and the parameters of life table were compared by paired Student's *t* test by two means (SAS System 2002). The program SigmaPlot 12.5 (Systat SoftWare®) was used to build up out the figures.

The life table parameters were calculated by the following formulas:

Net reproductive rate (R_0); $R_0 = \sum l_x m_x$.

Time of generation (T); $T = (\sum l_x m_x x) / R_0$

Intrinsic rate of growth (r_m); $r_m = \ln(R_0) / T$

Finite rate of growth (λ) in day^{-1} ; $\lambda = e^{r_m}$

where x is the age of adults in days, l_x is the survival to age x , and m_x is the number of eggs produced in the age x .

Results

In the choice test, there was a difference in oviposition preference of adults of *P. xylostella* by collards leaves of the different treatments ($F = 7.34$, $df = 12$, $p < 0.001$). SA treatment leaves were most preferred for oviposition followed by HyWR treatment and the Hy and HyD treatments were equally preferred (Fig 1a). In the no choice test, adults of *P. xylostella* had a higher preference for oviposition on the side of the collard leaf in which the wax was removed ($t = 10.72$, $df = 18$, $p < 0.001$) than on the side in which the wax was not removed (Fig 1b).

There was no difference between the foliar mass consumption by *P. xylostella* larvae on the collard cv. Santo Antônio of both treatments, i.e., with and without wax removal ($t = 1.37$, $df = 38$, $p = 0.178$). However, in the comparison between the disks of cv. Hybrid Kope F1 100MX, there were significantly higher consumption on the disks

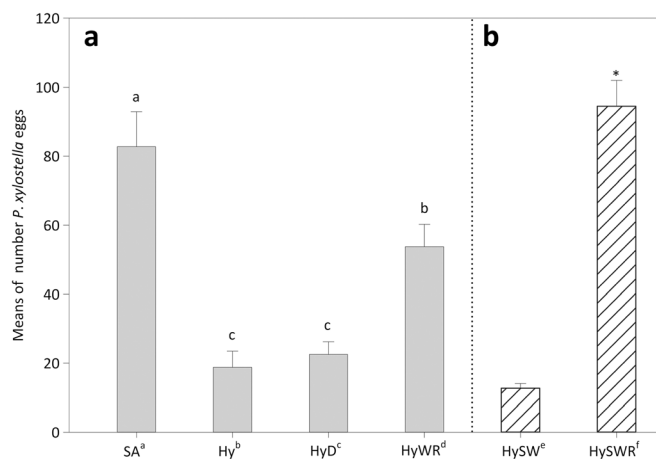


Fig 1 Oviposition of *Plutella xylostella* in collard leaves (27 ± 0.5 ; $75 \pm 5\%$). **a** Choice Test. **b** No-choice Test no. *Means followed by the same letter are not significantly different (Tukey's test, $p < 0.05$). *Significant t test ($p < 0.05$). SA cv. Santo Antônio, Hy cv. Hybrid Kobe F1 1000MX with wax, HyD cv. Hybrid Kobe F1 1000MX with epicuticular wax and damaged by pin, HyWR cv. Hybrid Kobe F1 1000MX with epicuticular wax removed, HySW leaf slice cv. Hybrid Kobe F1 1000MX with epicuticular wax, HySWR leaf slide cv. Hybrid Kobe F1 1000MX with epicuticular wax removed.

with wax removal (HyWR) when compared with disk without wax removal (Hy) ($t = 13.23$, $df = 38$, $p < 0.001$) (Fig 2).

The weight of the resulting pupae in the treatments with and without wax removal in collards leaves cv. Santo Antônio did not differ among themselves ($t = 0.55$, $df = 172$, $P = 0.58$); the same occurred in cv. Hybrid Kope F1 100MX in treatments with and without wax removal ($t = 1.34$, $df = 157$, $p = 0.18$) (Fig 3).

The average number of adult *P. xylostella* emerged from larvae feeding with collards leaves cv. Santo Antônio without removing wax (65.4 ± 3.0) was the same to those with wax

removal (66.4 ± 2.9) ($t = 0.22$, $df = 22$, $p = 0.86$). The same occurred in cv. Hybrid Kope F1 100MX; there was no difference between the emergence of adults from larvae developed in treatment without wax removal (65.4 ± 5.8) and with wax removal (59.3 ± 7.5) ($t = 0.66$, $df = 22$, $p = 0.51$) (Fig 4 and Table 1).

The fecundity of adults of *P. xylostella* emerged in both varieties was affected, with statistical differences among leaves with and without wax removal cv. Santo Antônio ($t = 10.05$, $df = 10$, $p < 0.001$) and cv. Hybrid Kope F1 100MX ($t = 6.94$, $df = 10$, $p < 0.001$). Greatest fertility was observed in the cv.

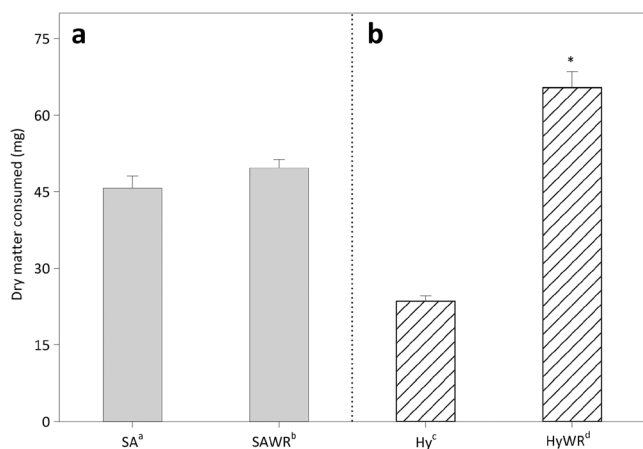


Fig 2 Matter consumed by larvae of *Plutella xylostella* on collard leaf disks (27 ± 0.5 ; $75 \pm 5\%$). **a** Larvae developed on leaf disks of collards cv. Santo Antônio. **b** Larvae developed on leaf disks of collard cv. Hybrid Kobe F1 1000MX. *Significant t test ($p < 0.05$). SA cv. Santo Antônio with epicuticular wax, SAWR cv. Santo Antônio with epicuticular wax removed, Hy cv. Hybrid Kobe F1 1000MX with epicuticular wax, HyWR cv. Hybrid Kobe F1 1000MX with epicuticular wax removed.

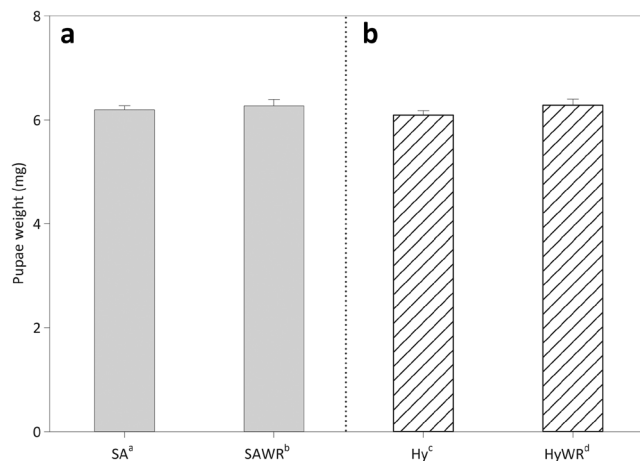


Fig 3 Weight of *Plutella xylostella* pupae (27 ± 0.5 ; $75 \pm 5\%$). **a** Larvae developed on leaf disks of collards cv. Santo Antônio. **b** Larvae developed on leaf disk cv. Hybrid Kobe F1 1000MX. *Significant t test ($p < 0.05$). SA cv. Santo Antônio with epicuticular wax, SAWR cv. Santo Antônio with epicuticular wax removed, Hy cv. Hybrid Kobe F1 1000MX with epicuticular wax, HyWR cv. Hybrid Kobe F1 1000MX with epicuticular wax removed.

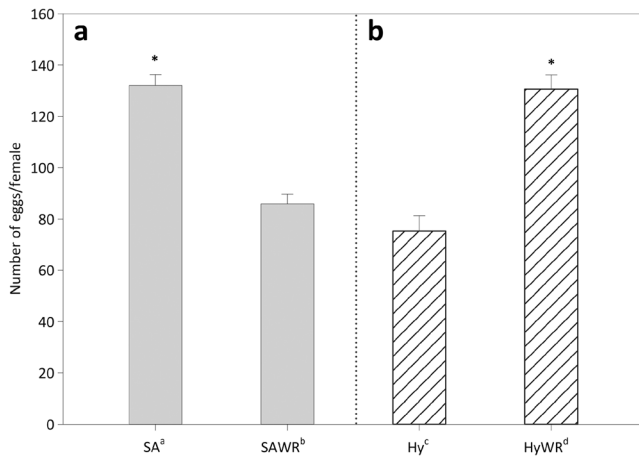


Fig 4 Total number of adult eggs of *Plutella xylostella* originated from larvae developed on collards. **a** Larvae developed on leaf disks of collards cv. Santo Antônio. **b** Larvae developed on leaf disks cv. Hybrid Kobe F1 1000MX. *Significant *t* test ($p < 0.05$). SA cv. Santo Antônio with epicuticular wax, SAWR cv. Santo Antônio with epicuticular wax removed, Hy cv. Hybrid Kobe F1 1000MX with epicuticular wax, HyWR cv. Hybrid Kobe F1 1000MX with epicuticular wax removed.

Santo Antônio without the wax removal and cv. Hybrid Kope F1 100MX when the wax was removed (Fig 4).

The life table parameters in the collards leaves cv. Santo Antônio showed significant differences between treatments in the values of the net reproductive rate ($t = 8.92$, $df = 22$, $p < 0.001$), intrinsic rate of increase ($t = 7.37$, $df = 22$, $p < 0.001$), and finite rate of increase ($t = 7.39$, $df = 22$, $p < 0.001$); however, there was no difference between the generation time ($t = 1.17$, $df = 22$, $p = 0.254$) (Table 2). For the cv. Hybrid Kope F1 100MX, there was significant difference between average net reproductive rate ($t = 3.0$, $df = 22$, $p = 0.006$) of treatment with and without wax removal, but there was no difference between intrinsic growth rate ($t = 1.03$, $df = 22$, $p = 0.314$), finite growth rate ($t = 0.01$, $df = 22$, $p = 0.99$), and the generation time ($t = 1.17$, $df = 22$, $p = 0.254$) (Table 2).

Discussion

In this study, we observed that the wax removal affected the oviposition preference of adults and foliar consumption rate of *P. xylostella* larvae. The wax of leaves of *Brassica* spp. is rich in compounds such as alkenes, paraffin, saponins, and glucosinolates (Spencer *et al* 1999, Renwick *et al* 2006, Badenes-Perez *et al* 2014). The glucosinolates and the saponins are metabolites used in defense against phytophagous insects. These compounds are considered the first and second line of defense used by *Brassica* species to prevent oviposition and feeding of insects (Sarosh *et al* 2010, Fathi *et al* 2011). As *P. xylostella* is an insect specialized in consumption of brassicas, it uses the glucosinolates as host recognition cues (Sun *et al* 2009, Chagas Filho *et al* 2010). In addition, these compounds possess action stimulants of feeding in larvae and oviposition in females of *P. xylostella* (Spencer *et al* 1999, Gigolashvili *et al* 2007, Thuler *et al* 2007, Badenes-Perez *et al* 2014). On the other hand, the saponins have contrary action; they act as repellents to adults and feeding inhibitors to larvae of *P. xylostella* (Badenes-Perez *et al* 2014).

In this study, we did not assess the chemical constitution and the proportion of these compounds in the wax of collards cultivars evaluated. Results showed deterrent effect in female oviposition and an increased leaf feeding rate in the *P. xylostella* larvae of the cv. Kobe F1 1000MX when the wax was removed suggesting a possible higher saponins level. On the other hand, the wax removal on the cv. Santo Antônio did not show the same result, suggesting that this plant has low saponins level. Future studies should be performed with both cultivars, cv. Kobe F1 1000MX and cv. Santo Antônio, to evaluate the amount and composition of their surface waxes.

Epicuticular waxes that cover leaf surfaces have an important function in plant defense against phytophagous insects, impairing mobility, preference, and adhesion of insects and eggs

Table 1 Survival of *Plutella xylostella* developed on leaf disks of collards cvs. Santo Antônio and Hybrid Kobe F1 1000MX variety (27 ± 0.5 ; $75 \pm 5\%$).

Stadium	SA		SAWR		Hy		HyWR	
	Lx	Deads	Lx	Deads	Lx	Deads	Lx	Deads
Larvae 1°	100.0 ± 0.00	7.5 ± 3.50 ^{ns}	100.0 ± 0.00	5.0 ± 1.89	100.0 ± 0.00	4.2 ± 1.90	100.0 ± 0.00	12.5 ± 5.70 ^{ns}
Larvae 2°	92.5 ± 3.50	3.3 ± 1.90	95.0 ± 1.90	4.2 ± 1.87 ^{ns}	95.8 ± 1.85	3.3 ± 2.60	87.5 ± 5.72	5.8 ± 2.89 ^{ns}
Larvae 3°	89.2 ± 4.20	3.8 ± 1.40	90.8 ± 3.14	6.5 ± 1.79 ^{ns}	92.5 ± 2.82	1.6 ± 1.10	81.7 ± 7.31	3.3 ± 1.38 ^{ns}
Larvae 4°	85.4 ± 4.50	3.0 ± 1.72	84.3 ± 2.60	9.8 ± 2.67 ^{ns}	90.9 ± 2.75	6.5 ± 2.20 ^{ns}	78.3 ± 8.33	5.8 ± 2.30
Pupae	82.4 ± 4.70	17.0 ± 4.04 ^{ns}	74.5 ± 2.90	8.1 ± 2.30	84.4 ± 3.04	18.8 ± 4.54 ^{ns}	72. ± 8.94	13.2 ± 2.82
Adults	65.4 ± 3.00		66.4 ± 2.9 ^{ns}		65.6 ± 5.8 ^{ns}		59.4 ± 7.5	

^{ns} no significant *t* test ($p \geq 0.05$), SA cv. Santo Antônio with epicuticular wax, SAWR cv. Santo Antônio with epicuticular wax removed, Hy cv. Hybrid Kobe F1 1000MX with epicuticular wax, HyWR cv. Hybrid Kobe F1 1000MX with epicuticular wax removed, Lx Survival to age *x*.

Table 2 Life table parameters of *Plutella xylostella* developed on leaf disks of collards cvs. Santo Antônio and Hybrid Kobe F1 1000MX variety (27 ± 0.5 ; $75 \pm 5\%$).

Parameters	SA	SAWR	Hy	HyWR
Net reproductive rate (R_o)	$50.39 \pm 2.29^*$	28.54 ± 1.23	30.83 ± 2.74	$43.30 \pm 5.45^*$
Generation time (T)	16.67 ± 0.09	16.85 ± 0.12^{ns}	17.29 ± 0.27	17.30 ± 0.31^{ns}
Intrinsic rate of growth (r_m)	$0.23 \pm 0.003^*$	0.20 ± 0.003	0.20 ± 0.007	0.21 ± 0.013^{ns}
Finite rate of growth (λ)	$1.26 \pm 0.004^*$	1.22 ± 0.004	1.22 ± 0.009	1.24 ± 0.016^{ns}

ns No significant *t* test ($p < 0.05$), *SA* cv. Santo Antônio with epicuticular wax, *SAWR* cv. Santo Antônio with epicuticular wax removed, *Hy* cv. Hybrid Kobe F1 1000MX with epicuticular wax, *HyWR* cv. Hybrid Kobe F1 1000MX with epicuticular wax removed.

*Significant *t* test ($p < 0.05$).

on the leaf surface (Eigenbrode *et al* 1999, Charleston & Kfir 2000). Thus, the wax removal can change physical properties of cv. Kobe F1 1000MX leaf surface leaving it more attractive and vulnerable to *P. xylostella* (Fathi *et al* 2011). Ulmer *et al* (2002) and Fathi *et al* (2011) demonstrated that oviposition of *P. xylostella* was higher in brassica leaves with brightening; a similar effect may have occurred in this work, we observed that the leaves became brighter after removing the wax.

The fertility rate of *P. xylostella* was higher in adults originated from larvae that fed on collards leaf disks of cv. Santo Antônio without wax removal and cv. Hybrid Kope F1 100MX with wax removal (Fig 4). However, there was no difference between the weight of pupae on the comparison of treatments with and without wax removal (Fig 3). Syed & Abro (2003) found a positive relationship between the weight of pupa and fertility of *P. xylostella*. Previously, Hasan & Ansari (2011) did not find any relationship between these variables, corroborating with our result. We expected that the weight of the pupae was influenced by foliar consumption rate (Figs 2 and 3) so that the weight of the pupae was higher in treatments where there was higher leaf consumption.

Some insect specialists have developed mechanisms that enable them to sequester plant's defense compounds and use them for their defense against natural enemies or use these compounds in its biology (Schoonhoven *et al* 2005). Wax removal of cv. Santo Antônio leaves may have eliminated fertility essences compounds to *P. xylostella*, reducing its reproductive potential. The opposite effect occurred with cv. Hybrid Kope F1 100MX, and wax removal may have eliminated toxic compounds on leaf surfaces, with positive effects in the fertility of *P. xylostella*.

The net reproductive rate (R_o), intrinsic rate of increase (r_m), and finite rate of increase (λ) in the collards leaves of cv. Santo Antônio were higher in the treatment without wax removal. Already in the cv. Hybrid Kope F1 100MX, there was a difference only in the comparison between the R_o of treatment with and without wax removal. In both varieties, the generation time (T) was not affected by the treatments (Table 2). The R_o , r_m , and λ are parameters used to describe the population growth, as the larger their values, the faster the population increases (Gotelli 2009).

The r_m is considered the most robust life table parameter to measure antibiosis effects of host plants in insects (Smith 2005); it indicates that a small delay in the organism reproduction can reduce net reproduction more than proportionally (Lewontin 1965). When r_m is low, fecundity becomes a critical factor in altering the rate of population growth (Gotelli 2009). In this context, the *P. xylostella* adults fecundity was the main responsible for difference in the r_m value of treatments with and without wax removal. In general, research shows that adults of moths and butterflies originated from larvae feeding on different *Brassica* species showed variation in the fecundity rate (Fathi *et al* 2011, Hasan & Ansari 2011), corroborating with our result.

In conclusion, the resistance of cv. Hybrid F1 Kope 100MX to *P. xylostella* is overcome by wax removal. Thus, cultural practices that promote direct contact of farmers or tools with the cabbage plants should be minimized in the growing area. Varieties with high content of wax can be grown in strips or mosaic with susceptible brassicas varieties, interfering in the host plant location by *P. xylostella*, or dispersing the infestation. The variety more attractive to oviposition may be used as trap crop; these crops would be planted close to the main crop in order to attract adults and oviposition of *P. xylostella*.

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References

- Agerbirk N, Olsen CE, Bibby BM, Frandsen HO, Brown LD, Nielsen JK, Renwick JAA (2003) A saponin correlated with variable resistance of *Barbarea vulgaris* to the diamondback moth *Plutella xylostella*. *J Chem Ecol* 29:1417–1433
- Asghari A, Fathi SAA, Mohammadi SA, Mohammaddust H (2009) QTL analysis for diamondback moth resistance in canola (*Brassica napus* L.). *Int J Plant Production* 3:29–34

- Awmack CS, Leather SR (2002) Host plant quality and fecundity in herbivorous insects. *Annu Rev Entomol* 47. doi:10.1146/annurev.ento.47.091201.145300
- Bacci L, Picanço MC, Barros EC, Rosado JF, Silva GA, Silva VF, Silva NR (2009) Physiological selectivity of insecticides to wasps (hymenoptera: Vespidae) preying on the diamondback moth. *Sociobiology* 53:151–167
- Badenes-Perez FR, Gershenzon J, Heckel DG (2014) Insect attraction versus plant defense: young leaves high in glucosinolates stimulate oviposition by a specialist herbivore despite poor larval survival due to high saponin content. *PLoS One* 9(4):e95766. doi:10.1371/journal.pone.0095766
- Baek JH, Kim JI, Lee DW, Chung BK, Miyata T, Lee SH (2005) Identification and characterization of ace1-type acetylcholinesterase likely associated with organophosphate resistance in *Plutella xylostella*. *Pestic Biochem Phys* 81:164–175
- Bukovinszky T, Potting RPJ, Clough Y, Van Lenteren JC, Louise EMV (2005) The role of pre- and post-alighting detection mechanisms in the responses to patch size by specialist herbivores. *Oikos* 109:435–446
- Chagas Filho NR, Boiça Junior AL, Alonso TF (2010) *Biologia de Plutella xylostella* L. (Lepidoptera: Plutellidae) em cultivares de couve-flor. *Neotrop Entomol* 39:253–259
- Chapman RF (1977) The role of the leaf surface in food selection by acridids and other insects. *Colloq Int CNRS* 265:133–149
- Charleston DS, Kfir R (2000) The possibility of using Indian mustard, *Brassica juncea* as a trap crop for the diamondback moth, *Plutella xylostella*, in South Africa. *Crop Prot* 19:455–460
- Eigenbrode SD, Nelson NK, Stoner KA (1999) Predation, behavior, and attachment by *Chrysoperla plorabunda* larvae on *Brassica oleracea* with different surface waxblooms. *Entomol Exp Appl* 90:225–235
- Fathi SAA, Bozorg-Amirkalae M, Sarfaraz RM (2011) Preference and performance of *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) on canola cultivars. *J Pestic Sci* 84:41–47
- Ferré J, Real MD, Van Rie J, Jansens S, Peferoen M (1991) Resistance to the *Bacillus thuringiensis* bioinsecticide in a field population of *Plutella xylostella* is due to a change in a midgut membrane receptor. *P Natl Acad Sci* 88:5119–5123
- Filgueira FAR (2008) *Novo manual de olericultura: Agrotecnologia moderna na produção e comercialização de hortaliças*. Editora UFV, Viçosa, p 421
- Gigolashvili T, Yatushevich R, Berger B, Müller C, Flügge UI (2007) The R2R3-MYB transcription factor HAG1/MYB28 is a regulator of methionine-derived glucosinolate biosynthesis in *Arabidopsis thaliana*. *Plant J* 51:247–261
- Golizadeh A, Kamali K, Fathipour Y, Abbasipour H (2009) Life table of the diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) on five cultivated brassicaceous host plants. *J Agr Sci Technol* 11:115–124
- Gotelli JN (2009) *Ecologia*. Editora Planta, Londrina, Brasil, p 302
- Grubb CD, Steffen A (2006) Glucosinolate metabolism and its control. *Trends Plant Sci* 11:89–100
- Halkier BA, Gershenzon J (2006) Biology and biochemistry of glucosinolates. *Annu Rev Plant Biol* 57:303–333
- Hasan F, Ansari MS (2011) Effects of different brassicaceous host plants on the fitness of *Pieris brassicae* (L.). *Crop Prot* 30:854–862
- Hopkins RJ, van Dam NM, van Loon JJ (2009) Role of glucosinolates in insect-plant relationships and multitrophic interactions. *Annu Rev Entomol* 54:57–83
- Husebye H, Chadchawan S, Winge P, Thangstad OP, Bones AM (2002) Guard cell- and phloem idioblast-specific expression of thioglucoside glucohydrolase 1 (myrosinase) in *Arabidopsis*. *Plant Physiol* 128:1180–1188
- Ishida M, Hara M, Fukino N, Kakizaki T, Morimitsu Y (2014) Glucosinolate metabolism, functionality and breeding for the improvement of Brassicaceae vegetables. *Breeding Sci* 64:48–59
- Kliebenstein DJ, Kroymann J, Mitchell-Olds T (2005) The glucosinolate-myrosinase system in an ecological and evolutionary context. *Curr Opin Plant Biol* 8:264–271
- Koroleva OA, Davies A, Deeken R, Thorpe MR, Tomos D, Hedrich R (2000) Identification of a new glucosinolate-rich cell type in *Arabidopsis* flower stalk. *Plant Physiol* 124:599–608
- Lewontin AC (1965) Selection for colonizing ability. In: Baker HG, Stebbins GL (eds) *The genetics of colonizing species*. Academic Press, New York, pp 79–94
- Müller C, Hilker M (2001) Host finding and oviposition behavior in a chrysomelid specialist – the importance of host plant surface waxes. *J Chem Ecol* 27:985–994
- Müller R, Vos M, Sun JY, Sonderby IE, Halkier BA, Wittstock U, Jande G (2010) Differential effects of indole and aliphatic glucosinolates on lepidopteran herbivores. *J Chem Ecol* 36:905–913
- Renwick JAA, Haribal M, Gouinguéné S, Städler E (2006) Isothiocyanates stimulating oviposition by the diamondback moth, *Plutella xylostella*. *J Chem Ecol* 32:755–766
- Rondelli VM, Pratisoli D, Polanczyk RA, Marques EJ, Sturm GM, Tiburcio MO (2011) Associação do óleo de mamona com *Beauveria bassiana* no controle da traça-das-crucíferas. *Pesqui Agropecu Bras* 6:212–214
- Sarosh BR, Wittstock U, Halkier BA, Ekbohm B (2010) The influence of metabolically engineered glucosinolate profiles in *Arabidopsis thaliana* on *Plutella xylostella* preference and performance. *Chemoecology* 20:1–9
- Schoonhoven LM, Van Loon JJA, Dicke M (2005) *Insect-plant biology*. Oxford University Press, New York, p 440
- Schuler TH, Martinez-Torres D, Thompson AJ, Denholm I, Devonshire AL, Duce IR, Williamson MS (1998) Toxicological, electrophysiological, and molecular characterization of knockdown resistance to pyrethroid insecticides in the diamondback moth, *Plutella xylostella* (L.). *Pestic Biochem Phys* 59:169–182
- Silva GA, Picanço MC, Bacci L, Crespo ALB, Rosado JF, Guedes RNC (2011) Control failure likelihood and spatial dependence of insecticide resistance in the tomato pinworm, *Tuta absoluta*. *Pest Manag Sci* 67:913–920
- Smith CM (2005) Plant resistance to arthropods: molecular and conventional approaches. *Springer Science & Business Media, Netherlands*, p 423
- Southwood TRE (1978) *Ecological methods with particular reference to the study of insect populations*. Chapman & Hall, London, p 524
- Spencer JL, Pillai S, Bernays EA (1999) Synergism in the oviposition behavior of *Plutella xylostella*: sinigrin and wax compounds. *J Insect Behav* 2:483–500
- Städler E (2002) Plant chemical cues important for egg deposition by herbivorous insects. In: Hilker M, Meiners T (eds) *Chemoecology of insect eggs and egg deposition*. Blackwell Publishing Ltda, Berlin, pp 171–204
- Sun JY, Sonderby IE, Halkier BA, Jander G, Vos M (2009) Non-volatile intact indole glucosinolates are host recognition cues for ovipositing *Plutella xylostella*. *J Chem Ecol* 35:1427–1436
- Syed TS, Abro GH (2003) Effect of Brassica vegetable hosts on biology and life table parameters of *Plutella xylostella* under laboratory conditions. *Pak J Biol Sci* 6:1891–1896
- Thuler RT, Bortoli SA, Hoffmann-Campo CB (2007) Classificação de cultivares de brássicas com relação à resistência à traça-das-crucíferas e à presença de glucosinolatos. *Pesqui Agropecu Bras* 42:467–474
- Ulmer B, Gillott C, Woods D, Erlandson M (2002) Diamondback moth, *Plutella xylostella* (L.), feeding and oviposition preferences on glossy and waxy *Brassica rapa* (L.) lines. *Crop Prot* 21:327–331
- Wang X, Wu S, Gao W, Wu Y (2015) Dominant inheritance of field-evolved resistance to fipronil in *Plutella xylostella* (Lepidoptera: Plutellidae). *J Econ Entomol* 109:334–338