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Alternative Non-Conventional and Environmentally Safe Horticultural, Mechanical and Local Treatments to Control *Cryptoblabes gnidiella* Millière (Lepidoptera: Pyralidae) in Pomegranate Orchards in the Northwestern Region of Egypt

Hashim, S. M.; R. M. Abd El-Moaty and A. W. Tadros
Plant Protection Research Institute, Agric. Res. Center, MOA, Giza, Egypt.

ABSTRACT

Alternative non-conventional and environmentally safe means of control of *Cryptoblabes gnidiella* (Lepidoptera: Pyralidae) in pomegranate orchards by horticultural, mechanical, microbial, and local chemical treatments were evaluated at El-Alamain district, Matrouh Governorate in the northwestern of Egypt during one and two successive years (2015 and 2016). The respective reductions rates of infestation with the following 12 treatments applied for one and two successive years were as follows: dormant pruning (4.04% increased to 4.17%), summer pruning (1.01 increased to 1.39%), dormant and summer pruning (6.06 increased to 8.89%), worming (36.36 increased to 44.44%), bacterial or fungal (14.14 or 10.10 increased to 18.06 or 11.11% ,respectively), local painting or local spraying (72.73 or 78.79 increased to 83.33 or 88.89%, respectively), pruning, worming, together with bacterial or fungal (52.53 or 48.49 increased to 58.33 or 56.94%, respectively), while pruning, worming, and local painting or local spraying treatments (82.83 or 90.91 increased to 91.67 or 95.83% ,respectively). Accordingly, it could be recommended that control of *C. gnidiella* could be effectively achieved by the safe means such as worming and local painting or spraying on the infestation sites in the crown of the stem.

INTRODUCTION

In Egypt, Pomegranate *Punica granatum* L. (Punicaceae: Myrtiflorae) cultivation is progressively increasing all-over the new reclaimed lands year after another, owing to its high nutritional benefits, profitable income, especially as exporting crop as well as high local trading.

However, pomegranate trees are subjected to infestation with several insect pests (Cocuzza, *et al.*, 2016) such as aphids, whiteflis, jassids, mealy bugs, fruit moths (*Virachola livia, Ectomyelois ceratoniae* and *Cryptoblabes gnidiella*), and fruit flies. The most economic insect pests, However, is the stem borer (the Leopard moth *Zeuzera pyrina*) Mesbah, *et al.* (1994), and Abdel Azim, *et al.* (2009).

Recently, *Cryptoblabes gnidiella* is recorded all over the world, and attacks several hosts mainly pomegranate, grapes, citrus, avocado, figs, mango, mulberry, as well as several field and vegetable crops (Hashem, *et al.*, 1996), causing losses reached 30% (Wysoki, *et al.* 1993).

In Egypt, The respective egg, larval, and pupal stages averaged 3, 13-14 and 5-7 days (Swailem and Ismail, 1972).

Abdel-Moaty *et al.* (in press, 2017) in Egypt recorded *C. gnidiella* larvae bore shallow tunnels under pomegranate stem at the crown region, causing weakness, rotten, withered, and finally death of trees. They studied the population level in the Northwestern of Egypt and found that moths started to emerge during March and continued until late November or early December, with 3 - 4 peaks yearly. Summer months recorded the maximum flight activity, medium during spring and autumn but ceased during winter

Although chemical control is costly, adversely affect the natural enemies (parasites, predators, and pathogens), and pollute the environment, yet chemical control treatments are recommended in fruit orchards.

Several attempts were conducted to use environmentally safe treatments to control tree borers in Egypt on several hosts (Tadros, *et al.*, 1993 a; Tadros, *et al.*, 1993 b; Tadros, *et al.*, 2006; Tadros, *et al.*, 2007 a; Tadros, *et al.*, 2007 b, and Abdel Azim, *et al.*, 2009).

The available literature in Egypt included studies on the biology of *C. gnidiella* on pomegranate trees (Swailem and Ismail, 1972) and monitored the population fluctuation in pomegranate orchards (Abdel-Moaty *et al.*, in press, 2017) are essential in determination of the proper timing of the pest control treatments. However, studies concerning the control of *C. gnidiella* in pomegranate orchards in Egypt and abroad are lacking and needs further exclusive work.

The aim of the present investigation is to prevent the pomegranate tree deterioration and yield losses through using non-traditional approaches for controlling *C. gnidiella* to minimize the pesticide residues, reduce the outbreaks of secondary species, decrease the environmental pollution, and magnify the role of the biological control agents.

MATERIALS AND METHODS

At El-Alamain district in the northwestern of Egypt, Matrouh Governorate, experiments were carried out in a pomegranate orchard (25 feddans and 10 years old) highly infested with *C. gnidiella.* Trials were extended during 2 successive years from January 2015 to November 2016. The following 13 treatments were evaluated using completely randomized design (10 trees each treatment and each tree was considered a replicate).

A. Horticultural Treatments:

1. **Dormant pruning treatment:** During January of each year, the regular horticultural winter pruning was carried out including the removal of the severely infested, deteriorated and dead stems infested with the borer and substituted by new branches growing under the crown of the stem (cancer branches).
2. **Summer pruning treatment:** During June, the newly severely infested stems were pruned as described in item (1).
3. **Dormant and summer pruning treatments:** Treatments numbers 1 and 2 were applied together.

B. Mechanical Treatment:

4. **Worming treatment:** During January of each year, a knife (not sharp) and a wire were used to scratch in the infested areas on the crown of the stem for the larvae and pupae inside the tunnels and kill them.

C. Microbiological treatments:

5. **Bacterial treatment:** Bactospeine F.C. (a.i. *Bacillus thuringiensis* (Berliner), 8500 International Units Ak / mg) at the rate of 200 cc/100 liters of water was locally

sprayed on the crown of the stem, 4 times each season (on April, May, June and July) using knapsack sprayer.

6. **Fungal treatment:** Biofly F.C. (a.i., *Beauveria bassiana*, 3 x 10^7 spores / mg) at the rate of 400 cc/100 l. w. were locally sprayed on the crown of the stem, 4 times each season (on April, May, June and July) using knapsack sprayer.

D. Local chemical treatments:

7. **Local painting treatment:** The MOA recommended Basudin (Diazinon) 60% EC and Cidial L (Phenthoate) 50% EC each at the rate of 300 cc/100 l. w. was used to paint the infested sites on stems, 4 times alternatively each season (on April, May, June and July). Painting was practical using a brush.
8. **Local spraying treatment:** The MOA recommended Basudin (Diazinon) 60% EC and Cidial L (Phenthoate) 50% EC each at the rate of 300 cc/100 l. w. was sprayed 4 times alternatively each season (on April, May, June and July). Spraying was practiced by a knapsack sprayer (20 liters capacity) and mainly directed towards the infested sites on stem.

E. Combined treatments:

9. **Pruning, worming, and bacterial treatment:** Treatment numbers 3, 4, and 5 were conducted together.
10. **Pruning, worming, and fungal treatments:** Treatments numbers 3, 4, and 6 were conducted together.
11. **Pruning, worming, and local painting treatments:** Treatments numbers 3, 4, and 7 were conducted together.
12. **Pruning, worming, and local spraying treatments:** Treatments numbers 3, 4, and 8 were carried out together.

F. Untreated:

13. **Check treatment:** Check trees were left untreated as control treatment.

G. Procedures of treatments: The previous 13 treatments were conducted through January to November 2015 seasons. During the 2nd season (January to November 2016), the same previous treatments were repeated on other trees in another nearby area of the same orchard with the same technique for confirmation.

In the meantime, the same previous 13 treatments were carried out on the same last year trees to evaluate the effect of the treatments when applied for two successive years (from January 2015 to November 2016).

Treatments were evaluated at the end of the year (during November) by counting the alive larvae in the treated and untreated pomegranate trees.

H. Evaluation of treatments: The experimental design was completely randomized at significance level 5% split design with 10 trees, each tree was considered as a replicate. The efficiency of treatments was estimated according to the reduction percentage of the borer infestation described by Henderson and Tilton, (1955), as follow:

$$\% \text{ reduction of infestation} = [(C - T) / C] 100$$

Where, **C: the mean number of pupal skins in untreated trees.**
T: the mean number of pupal skins in treated trees.

Grouping of treatments was based on ANOVA test and "Least Significant Difference" (Snedecor and Cochran, 1990).

RESULTS AND DISCUSSION

Trials were conducted at El-Alamain district in the northwestern of Egypt, Matrouh Governorate to evaluate the effect of different horticultural, mechanical, microbial, and local chemical treatments alone or in combination with each other's on the reduction of *C. gnidiella* infestation in pomegranate orchard. The direct effects of treatments were evaluated when applied for only one single year (January to November 2015 or January to November 2016). The cumulative effects were also evaluated for two successive years (January 2015 to November 2016).

1. **Effect of one single year treatments (Direct effect) is presented in (Table, 1):**

1. Effect of horticultural treatments:

1. **Effect of dormant pruning treatment:** Pruning treatment was of no value since *C. gnidiella* larvae feed and habitat inside the crown area of the main stem which always not included in the dormant pruning. Thus, the reduction of infestation was very little (3.92 – 4.17%; mean, 4.04%).

2.Effect of summer pruning treatments: Due to the undetectable symptoms of new infestation inside the crown area of the main stem, and *C. gnidiella* larvae did not occur in the branches, summer pruning was of poor value in reducing the borer infestation (only 0.00 – 1.96%; mean, 0.98%).

3. Effect of dormant and summer pruning treatments: The reduction in *C. gnidiella* infestation very slightly increased when dormant and summer treatments were applied together compared with each treatment alone, their values ranged 5.88 – 6.25% (mean, 6.06%).

2. Effect of mechanical treatment:

1. **Effect of worming treatment:** Worming treatment was of some considered value owing to the shallow larval habitat under the bark of the tree stem, in addition to the advantage as environmentally safe treatment. This treatment additionally exposed the larval tunnels to parasites and predators as well as the weather factors which play an active role in the reduction of borer infestation. The reduction of the borer infestation reached 35.29–37.5% (mean, 36.39%).

2. Effect of microbial treatments:

1. **Effect of bacterial treatment:** Bacterial treatment was relatively of low active in the field as the bacteria highly affected with the weather factors (especially higher temperature and hot wind) and the difficulty of these bacteria to reach the larvae inside their tunnels. Therefore, this treatment was less effective as the reduction reduction percentage of infestation recorded only 12.5 - 15.69% (mean, 14.09%).
2. **Effect of fungal treatment:** As in bacteria, the percentage reduction in *C. varius* infestation due to fungal treatment was as low as 8.33 - 11.76% (mean, 10.04%).

3. Effect of local chemical treatments:

1. **Effect of local painting treatment:** The effect of local painting on the crown areas of stems four times / year with recommended insecticides significantly increased the reduction percentage of *C. gnidiella* infestation showing 70.83 – 74.51% (mean, 72.67%). This high percent reduction was due to the direct treatment of

insecticides to the borer inside these infested sites.

2. **Effect of local spraying treatment:** The effect of local spraying on the crown areas of stems four times / year with recommended insecticides significantly and adequately reduced *C. gnidiella* infestation with 77.08 - 80.39% (mean, 78.73%). This treatment hindered the moth settings, oviposition, hatching and larval entry inside the pomegranate tree stem.

Table 1. Effect of single years treatment on the reduction percentage of *C. gnidiella* infestation in pomegranate orchards in the northwestern of Egypt, during each single years 2015 and 2016 seasons.

Treatments	Mean no. of alive larvae per tree (L/T) and percent reduction of infestation (%RI)						Grouping
	1st year 2015		2nd year 2016		Mean		
	(L/T)	%RI	(L/T)	%RI	(L/T)	%RI	
A- Horticultural Treatments:							
1- Dormant pruning	4.9	3.92	4.6	4.17	4.75	4.04	f
2- Summer pruning	5.0	1.96	4.8	0.00	4.90	0.98	g
3- Dormant & summer pruning	4.8	5.88	4.5	6.25	4.65	6.06	f
B- Mechanical Treatments:							
4- Worming	3.3	35.29	3.0	37.5	3.15	36.36	d
C- Microbial Treatments:							
5- Bacterial	4.3	15.69	4.2	12.5	4.25	14.09	e
6- Fungal	4.5	11.76	4.4	8.33	4.45	10.04	e
D- Local Chemical Treatments:							
7- Local painting	1.3	74.51	1.4	70.83	1.35	72.67	b
8- Local spraying	1.0	80.39	1.1	77.08	1.05	78.79	ab
E-Combined Treatments:							
9- Treatments, 3 + 4 + 5	2.3	54.90	2.4	50.00	2.35	52.45	c
10- Treatments, 3 + 4 + 6	2.5	50.98	2.6	45.83	2.55	48.49	c
11- Treatments, 3 + 4 + 7	0.9	82.35	0.8	83.33	0.85	82.83	ab
12- Treatments, 3 + 4 + 8	0.5	90.20	0.4	91.67	0.45	90.91	a
F- Untreated Treatments:							
13- Check	5.1	--	4.8	--	4.95	--	h

Values within a column followed by different letter are significantly different (P> 0.05), L.S.D. = 0.51, Duncan [1951 as described by Computer Mstat Program, 1987] multiple ranges test.

1. Effect of combined treatments:

1. **Effect of pruning, worming, and bacterial treatments:** Table (1) indicated that pruning and bacterial treatments did not increase the effectiveness of the combined treatments as the reduction percentage in *C. gnidiella* reached 50.00 - 54.90% (mean, 52.45%). The obtained results are mainly due to worming treatment.
2. **Effect of pruning, worming, and fungal treatments:** As mentioned above, the effectiveness of these treatments was mainly due worming but the pruning and fungal treatment did not add noticeable effect. This combined treatment resulted in 45.83 - 50.98% (mean, 48.40%).
3. **Effect of pruning, worming, and local painting treatments:** Excellent results were obtained when these combined treatments were applied together showing 82.35 - 83.33% (mean, 82.83%) reductions of infestation. The main effect was due to worming, and local painting treatments.
4. **Effect of pruning, worming, and local spraying treatments:** As shown in Table (1), almost equal excellent and satisfactory results were achieved when these combined treatments were applied together showing 90.20 - 91.67% (mean, 90.91%) reductions in infestation. Although pruning treatment did not show noticeable effect.

2. **Effect of two successive year treatments (Cumulative effect):** Table (2)

1. **Effect of horticultural treatments alone:** Data in Table (2) indicated that, dormant pruning treatment alone in winter even applied two successive years did not reduced *C. gnidiella* infestation in pomegranate orchards even when applied for two successive years. This relatively low reduction percentage of infestation (4.17%) was due to the larval infestation which was mainly concentrated only in the crown of the stem. Summer pruning had almost no effect in this respect (1.39%), although it was repeated for two successive years. Summer pruning did not share in the reduction of infestation and should be excluded in the integrated control program. Even dormant and summer pruning treatments together for two successive years slightly reduced infestation with 8.89%.
2. **Effect of mechanical treatment alone:** Worming treatment (killing larvae, pre-pupae, and pupal stages) was generally difficult to apply but it had reliable effect in the reduction of infestation (44.44%).
3. **Effect of microbial treatments:** The pathogenic bacteria or fungus was relatively useless in this respect even when applied cumulatively for two successive years (18.06 and 11.11%, respectively) for the previously mentioned reasons.
4. **Effect of local chemical treatments**: Local painting and local spraying 4 times / year was quite effective in the reduction of *C. gnidiella* infestation especially when was applied for two successive years (83.33 and 88.89%, respectively).
5. **Effect of combined treatments:**

Applying dormant pruning, summer pruning, worming, microbial, and/or local chemical treatments in different combinations resulted in adequate

reduction in *C. gnidiella* infestation in pomegranate orchards especially when carried out yearly.

Winter and summer pruning, worming and bacterial treatments showed 58.33% reduction of infestation when conducted for two successive years. Applying winter and summer pruning, worming and fungal treatments for two successive years resulted in almost similar results (56.94%). Winter and summer pruning, worming with local painting for two successive years almost doubled reduction percentage in the *C. gnidiella* borer infestation (91.67%). Winter and summer pruning, worming with local spraying for two successive years resulted in almost similar high reduction percentage in the *C. gnidiella* borer infestation (95.83%).

Table 2. Effect of two successive years treatment on the reduction percentage of *C. gnidiella* infestation in pomegranate orchards in the northwestern of Egypt, during the two successive seasons (2015 and 2016) and differences between one and two year's treatments.

Treatments	Two successive years		Mean one single year	Differences between 1 & 2 years
	No. of larvae	% reduction of infestation	% reduction of infestation	
A- Horticultural Treatments:				
1- Dormant pruning	6.9	4.17	4.04	0.13
2- Summer pruning	7.1	1.39	1.01	0.38
3- Dormant & summer pruning	6.6	8.89	6.06	2.83
B- Mechanical Treatments:				
4- Worming	4.0	44.44	36.36	8.08
C- Microbial Treatments:				
5- Bacterial	5.9	18.06	14.14	3.92
6- Fungal	6.4	11.11	10.10	1.01
D- Local Chemical Treatments:				
7- Local painting	1.2	83.33	72.73	10.60
8- Local spraying	0.8	88.89	78.79	10.10
E- Combined Treatments:				
9- Treatments, 3 + 4 + 5	3.0	58.33	52.53	5.80
10- Treatments, 3 + 4 + 6	3.1	56.94	48.49	8.45
11- Treatments, 3 + 4 + 7	0.6	91.67	82.83	8.84
12- Treatments, 3 + 4 + 8	0.3	95.83	90.91	4.92
F- Untreated Treatments:				
13- Check	7.2	---	---	---

Statistical analysis: Statistical analysis and grouping of the 12 applied treatments for one and two years concluded that there were significant differences between treatments and classified as: {Insignificant differences between the same letters of grouping}

1. Superior group (80 – 100%): MOA Committee approval recommendation

1. Pruning, worming, and local spraying for two years (95.83%) A
2. Pruning, worming, and local painting for two years (91.67%) A
3. Pruning, worming, and local spraying for one year (90.91%)A
4. Local spraying for two years (88.89%) A
5. Local painting for two years (83.33%) A
6. Pruning, worming, and local painting for one year (82.83%) A

2. Sufficient group (50 – less than 80%):

1. Local spraying for one year (78.79 %) B
2. Local painting for one year (72.73 %) B
3. Pruning + Worming + Bacterial for two years(58.33%)C
4. Pruning + Worming + Fungal for two years (56.94%) C
5. Pruning + Worming + Bacterial for one year (52.53%) C

3. Moderate group (30 - less than 50%):

1. Pruning + Worming + Fungal for one year (48.49%) C
2. Worming for two years (44.44%) C
3. Worming for one year (36.36%) C

4. Least group (less than 30%):

1. Bacterial for two years (18.06%) D
2. Bacterial for one year (14.14 %) D
3. Fungal for two years (11.11%) D
4. Fungal for one year (10.10%) D
5. Dormant and summer pruning for two years (8.89%) E
6. Dormant and summer pruning for one year (6.06%) E
7. Dormant pruning for two years (4.17%) E
8. Dormant pruning for one year (4.04%) E
9. Summer pruning for two years (1.39%) F
10. Summer pruning for one year (1.01%) F

From the foregoing results, it could be concluded that the direct effect of one single year treatments on *C. gnidiella* infestation varied from one treatment to another. The cumulative effect of two successive year treatments proved that the infestation could be magnified if these treatments repeated yearly.

The effect of horticultural treatments alone (winter and summer pruning alone or together) was of no value when applied for one season or repeated for two successive years. This is due to *C. gnidiella* larval feeding and habitat inside the crown area of the main stem which not included in the summer or dormant or pruning.

The direct effect of mechanical treatment alone (worming) was of moderate value owing to the shallow larval habitat under the bark of the tree stem, in addition to its environmental safety treatment, and magnified when the treatment was repeated succcessivly.

Microbial treatments with bacteria or fungus showed very low effects for one year and successively for two years. This was owing to the phenomenon that the pest hide inside the tree wood under the bark in addition that the bacteria and fungus were highly affected with the weather

factors in the field and failed to reach the larvae inside.
Local painting and local spraying were quite effective in the reduction of the borers' infestation. The cumulative effect for two years highly increased the reduction of infestation.

Applying the environmentally safe treatments such as dormant pruning in winter with the summer pruning, worming together with pathogenic microbial or local chemical treatments in different combinations highly magnified the reduction of infestation. Applying these combined treatments successively year after another greatly and satisfactory reduced *C. gnidiella* borer infestation in pomegranate orchards.

The obtained results are somewhat in agreement with some authors in Egypt who applied alternative safe means of controlling some insect borers in fruit orchards (horticultural, mechanical, pheromone traps and local chemical treatments). These treatments were applied on *Synanthedon myopiformis* in apple orchards (Tadros *et al.*, 2007), *Zeuzera pyrina* in apple and pear orchards (Tadros *et al.*, 1993) and Tadros *et al.*, 2006), *Paropta paradoxa* in vineyards (Tadros *et al.*, 1993), *Scolytus amygdali* in apricot orchards (Tadros *et al.*, 2007), and *Chlorophorus varius* in citrus orchards .

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الطرق البديلة لمكافحة حفار ساق الرمان "كريبتوبلابس جنديللا" *Cryptoblabes gnidiella* Millière (Lepidoptera: Pyralidae) فى حدائق الرمان فى الساحل الشمالى الغربى بمصر.

صلاح محروس هاشم ، راضي محمدي عبد المعطي و أنطون ولسن تادرس

معهد بحوث وقاية النباتات – مركز البحوث الزراعية – وزارة الزراعة – الجيزة - مصر

تم تقييم فعالية بعض الطرق الغير تقليدية والأمنة بيئيا لمكافحة حفار ساق الرمان "كريبتوبلابس جنديللا" *Cryptoblabes gnidiella* فى حدائق الرمان باستخدام المعاملات البستانية والميكانيكية والميكانيكية والكيماوية الموضعية في منطقة العلمين (محافظة مطروح) في الشمال الغربى بمصر، لمدة عام واحد وعامين متتاليين (2014/2015و 2015/2016). بلغت معدلات خفض الإصابة عند تطبيق المعاملات الإثني عشر الآتية لمدة عام واحد وعامين متتاليين ما يلي، علي الترتيب: معاملات التقليم الشتوي (31.04% إزدادت إلي 45.45%)، ومعاملات التقليم الصيفي (5.7% إزدادت إلي 7.14%)، ومعاملات التقليم الشتوي والصيفي معا (34.9% إزدادت إلي 48.21%)، ومعاملات قتل اليرقات داخل أنفاقها (4.19% إزدادت إلي 6.85%)، والمعاملات البكتيرية أوالفطرية (8.39-6.88% إزدادت إلي 14.58-11.31%)، ومعاملات الدهان الموضعي أو الرش الموضعي (73.32 أو 72.15% إزدادت إلي 84.52 أو 79.46%)، ومعاملات التقليم مع قتل اليرقات مع البكتيرية أوالفطرية (38.76 أو 40.27% إزدادت إلي 49.40 أو 48.81%)، ومعاملات التقليم، مع قتل اليرقات مع الدهان الموضعي أوالرش الموضعي (84.23 أو 82.55% إزدادت إلي 95.24 أو 93.75%). ونتيجة لهذا البحث يمكن التوصية بالمعاملات البيئية الآمنة مثل التقليم الشتوي، مع الدهان الموضعي أو الرش الموضعي لأماكن الإصابة فقط.