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Impact of abamectin, *Bacillus thuriengiensis* and Neem oil extract on *Aphis gossypii* glover and *Bemissia tabaci* pests of the watermelon (*Citrullus lanatus*) in Dschang

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Abstract

A study on the control of watermelon pests was conducted in Western region of Cameroon. The trial was laided on a randomised complete block design with three replications at the University of Dschang farm from December 27, 2019 to April 10, 2020 (dry season). The treatments were T1 (control plot), T2 Biotrin (Abamectin 5% at 375ml/ha), T3 Antario (*Bacillus thuringiensis* 1.4%+abamectin 0.1% at 375g/ha), T4 Neem oil (0.3 to 1% Azadirachtin) at 7l/ha, 400 H2O/ha) and T5 (Emamectin benzoate 50g/kg, 250g/ha). Pest abundance, leaf infestation rate, plant growth parameters and yields were assessed weekly from 26 days after sowing to three weeks before harvest. The results showed that *Aphis gossypii* (Hemiptera: Aphididae) the major pest (39.54±7.15) and *Bemisia tabaci* (Hemiptera: Aleyrodidae) (1.64±0.16) the second host infested watermelon. The peak of the leaf infestation rate is 20.97% at 40 DAS. The number of aphids was higher at 54DAS (71.34±26.30) and for whiteflies at 68 DAS (2.83±0.54). The effect of the treatments was significant (P>0.05) for all parameters. The control plot was more infested with highest mean leaf infestation rate, number of Aphids and whiteflies respectively 29.6± 2.30%; 158.03±29.59; 1.04±0.20 than Biotrin less attacked (4.11 0.89%; 2.21±0.4; 0.89±0.2). Biotrin, neem oil, Antario and Emacot are comparable for all parameters. In summary, the lowest yield was obtained in the control plot (35.71±2.66t/ha) but was not significantly different (P>0.05) from those of the other plots. Biotrin, neem oil and Antario are bioinsecticides alternatives for Emacot and it should be applied before the 40DAS.

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Introduction

Vegetables play a fundamental role in food and nutrition security programmes (James *et al.*, 2010). They are the most important and inexpensive component of a balanced diet that are taken into account because of their richness in nutrients essential for the body (Andreas Ebert, 2014). In Africa, vegetable production is one of the dynamic sectors, favourable to producers and exporters in terms of both income and employment (Toutchap, 2018). Watermelon (*Citrullus lanatus* Thumb (Sankar *et al.,* 2020) represents one of the most important vegetable crops of the cucurbit family, widespread throughout the world. From a nutritional standpoint, it is rich in water or 90.9% water on average with moisturising properties and low fat without cholesterol. It contains citrulline, which is used to synthesize another key amino acid in the body, with argine playing a key role in the vision, healing and elimination of ammonia (Arvanitoyannis *et al.*, 2008). FAO (2014) reported that watermelon production has quadrupled in the last 40 years. In 2016, it stood at 117,022,560t representing 3,507,243 ha with an average yield of 3.34kg (FAOSTAT). In Cameroon, watermelon production increased from 44,527 t in 2010 to 51,893 t in 2013 (National Tropical Institute, 2015).

Watermelon production is a profitable sector like any other vegetable. Watermelon producers face insect pests as a major constraint as other vegetable in the world (Charleston *et al.*, 2005; Djomaha, 2018). Theirs are the group of pests whose damage is significant because of climatic conditions that are favourable to their development (Imam *et al.*, 2010). These watermelon pests are in particular the melon or cotton aphid (*Aphis gossypii* Glover), the whitefly (*Bemisia tabaci*), (National Tropical Institute, 2015).

Pests of the Hemiptera's families are a major economic and agricultural problem in many tropical crops. They cause direct damage by sucking the contents of the leaves for food, thereby slowing down the growth and development of the plant. Indirectly, they secrete honeydew that produces fumagin and provides access to viruses (Blackman and Eastop, 2007). In the case of *Aphis gossypii*, the losses are up to 20% for direct attacks and 85% when the attacks are indirect and those of *Bemisia tabaci* go up to 100%. To address this problem, many farmers resort to the use of synthetic insecticides despite their adverse effects on the environment and human health (Grzywas *et al.,* 2002).

One of the problems associated with insecticides is the mechanism of resistance of pests to different active ingredients (Charleston *et al.*, 2005). Thus, this study focuses on increasing yields while respecting the environment through integrated pest management. Hence our test, that can justify the use of bioinsecticides such as biotrin, antario and neem oil to the detriment of synthetic insecticides.

Materials and methods

The test was set up at the FAAS's Application and Research Farm (FAR) of the University of Dschang from December 20, 2019 to April 27, 2020 in an experimental plot opposite amphitheatre 1000. Annual rainfall is between 1800 and 2000 mm and average annual temperatures vary between 10°C and 35°C.

Plant material

The plant material used is watermelon seed (*Citrullus lanatus*); more precisely the standard variety BELLA CAOLACK, variety very appreciated by the population of West Cameroon where it is sold in all local markets and is widely consumed by households.

Insecticides

The chemical insecticide used was Emacot. The active ingredient is Emamectin benzoate 50g/kg WG-dispersible granules. Emacot is a preventive and curative insecticide that acts by ingestion, very effective against insect larvae. Foliar application was done at a dose of 250g/ha every 7 days.

Biotrin

The active ingredient of biotrin is abamectin 5%. Biotrin works by poisoning insect larvae, quickly preventing the insect from feeding, which leads to their death. The application (foliar) was done every 7 days at a dose of 375ml/ha.

Antario

The active ingredient of antario is *Bacillus thuringiensis* var. Kurstaki (132000 $IU.mg^{-1}$) and abamectine (0,1%) in the form of wet powder. The application (foliar) was made every 7 days at a dose of 375 g/ha. Antario acts through two synergistic modes of action of *B. thuringiensis* which with its natural isolates is able to destroy certain species of insects (Lepidoptera, Diptera and Coleoptera) and abamectin whose mode of action has been described in the description of the biotrin (Russell IPM, 2018).

Neem oil

The equipment was purchased from a distributor in Dschang from northern Cameroon. The active ingredient of neem oil is 0.3 to 1% Azadirachtin and 18% Neem oil. This solution can act as a repellent, anti-feeder or as a growth regulator that can affect egg laying in females as well as the molting and growth of larvae in some arthropods. Its application was done once a week at the dosage $16ml.l^{-1}$ or 240ml per sprayer of 15l of H20; or $7l.ha^{-1}$for 400l of water.

Experimental design

It was in randomised complete blocks having five treatments and three replications. The total area is $400m^2$, with three blocks spaced 1m apart. Each block had 5 experimental units of 3m long and 5m wide giving an area of $15m^2$, separated from each other by 1m. The gaps between the plants were 1m*1.5m or a density of plants of 6,667 plants ha^{-1}. Treatments were T1 (control), T2 (biotrin), T3 (antario), T4 (neem oil) and T5 (emamectin benzoate).

Technical details

Clearing was done manually with cutlass to remove *Tithonia diversifolia*, *Mimosa* spp and others weeds.

Staking and ploughing: the experimental plot after clearing was cleaned and the stakes were placed. The staking of the plot consisted of firstly drawing the right angles of the plot from a base line following the 3-4-5 method of the Pythagorean Theorem. The ploughing of the plot was done manually using hoes, daba, picks, forks because the soil was very hard.

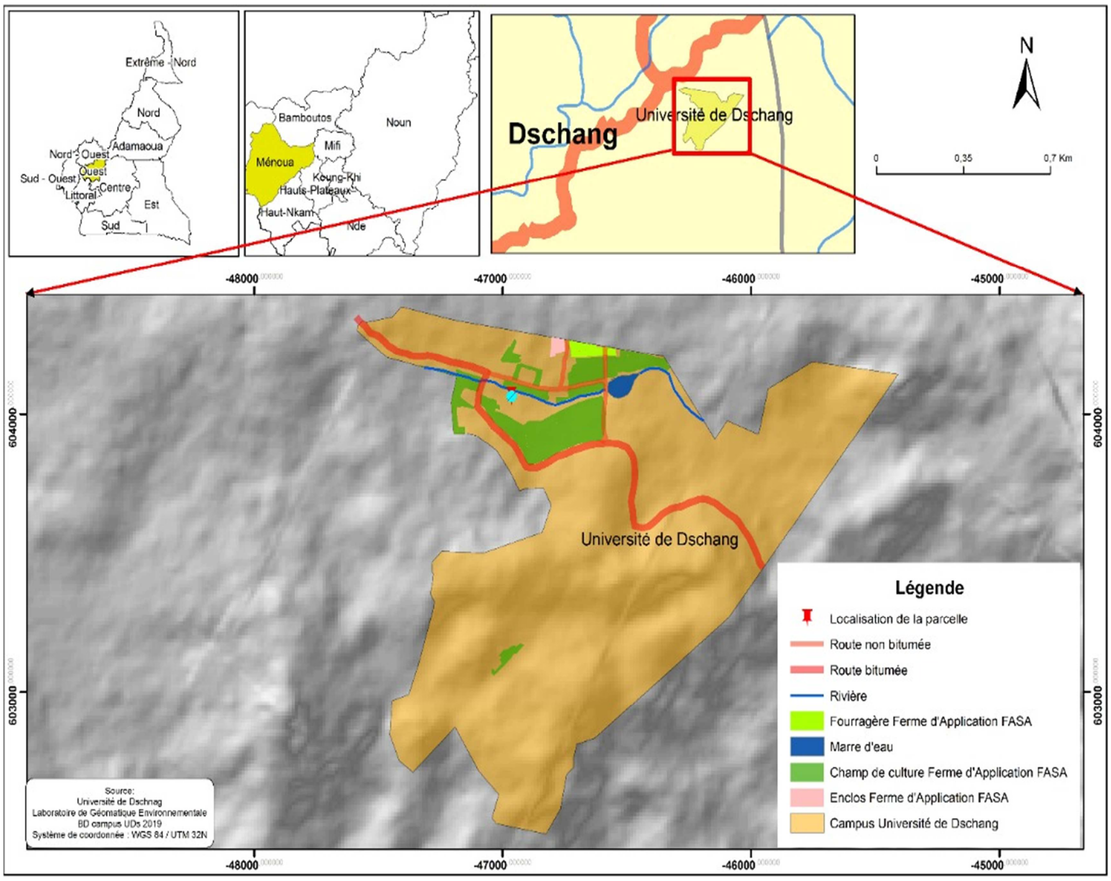

Fig. 1. Location of the research area.

Organic fertilization: 1Kg of poultry manure mixed with Arabic soil was introduced into the hole and watered for two weeks for decomposition.

Chemical fertilization: the fertilizer formula used is 13-13-21 (NPK), applied at the rate of 10g per hole 3 weeks after seed emergence and repeated after every 3 weeks.

Fungicide application: PYRAM (65% Metiram + 5% pyraclostrobin at 33 g for 15l of H20) which is systemic and contact was applied every two weeks. Then AGREB 80WP to avoid the effect of resistance, composed of 800g.kg-1 of Maneb was used in foliar pulverization at the dose of 80g for 15l H20 every two weeks.

Irrigation: Manual watering was done twice a week with watering cans of 12 l of water per m^2 until the rains returned in March.

Weeding: Throughout the production cycle, two weeding operations were carried out on January and February.

The sanitary harvest: All plants showing signs or symptoms of diseases on leaves and fruits were pulled out in the March 6th.

Data collection

Sampling starts on January 29, 2020 and continues at the weekly basis. The plants were randomly sampled in each experimental unit, 8 plants inside each plot to the detriment of those of the borders. The data collection was done by visual counting one a week at morning time before the spread of insecticides.

Evolution aphid and whitefly populations of on plants

The counting of nymphs and adults of aphids, eggs, larvae, nymphs and adults of whiteflies was done to determine the abundance of each pest. The upper and lower sides of the total leaves of each plant were observed.

Leaf infestation rate

The total leaves and the coiled leaves of each plant were counted and the leaf rate infestation was obtained according to the formula of Balajas *et al.* (2008):

$$\text{Infestation rate} = \frac{\text{number of leaves rolled}}{\text{number of leaves}} \times 100$$

For the growth parameters of the plant

The diameter at the stem collar was measured with the calliper and the length of the stem by the tape measure

Yield

The Fresh Mass (MF) of the Fruits taken from the eight plants sampled from each plot was obtained from a scale.

Data analysis methods

MS Excel software was used for processing prior to a descriptive analysis of parameters. The graphs of the parameters of the infestation rate according to treatments and time and that of the level of loss according to treatments and time were made using the MS Excel software. For data analysis, the JMP SAS 8.02 software was used to compare different treatments. The ANOVA tests were done to compare treatments when the data followed the normal distribution

Results and discussions

Evolution of the average leaf infestation rate, umber of Aphis gossypii and Bemisia tabaci and the growth parameters of plant according to time

The following fig. (Fig.2A) shows the evolution of the average leaf infestation rate over time; it appears that the curve evolves in an increasing way until it reaches its peak before decreasing. At 26 DAS, the plot is already infested by pests at a rate of 10% and will be evolving until 33 DAS thus reaching 14.61%. At 40DAS, the peak is reached with a value of 20.97%. At 54 DAS, the curve decreases considerably and the rate of leaf infestation drops to 9.44% and continues to decrease until the end of the evaluation to 7.75%. The high infestation rate at this date would be due to the low number of leaves. The plant still at the young stage would have undergone a heavy pressure of the two pests on the stems and leaves. Thus, the most sensitive period of watermelon growth is between emergence and 40 DAS.

Aphids (Fig.2A) are present at 26 DAS the number of aphids suddenly rises to 47 DAS and peaks at 58 DAS. The number of aphids gradually decreases to 68 DAS without crossing the average of 60 aphids per plant.

As concerns the whiteflies, the number is 1.5 at 26 DAS but decreases to 33 DAS, then increases abruptly to 40 DAS and goes down and up to reach the peak at 68 DAS.

As for the white flies, the number is 1.5 at 26 DAS but decreases at 33 DAS, then increases abruptly at 40 DAS and goes down and up to reach the peak at 68 DAS. Probably the flowering period would be the best time for white flies on watermelon. But the evaluation cannot be continued on flowering plants because of the loss of flower buds. The treatment of the plot against the rise of the whiteflies is thus to be considered. This result shows that whiteflies can be harmful to the plant. They suck the juice of the plants to feed and excrete the excess sugars into a sticky substance. The heavily affected fruits become sticky. The adult insects and their larvae secrete this sticky substance, called honeydew. On the honeydew develops fumagine. In case of a heavy whitefly infestation, the plant will not be able to develop anymore because of the damage caused by their stings and the harvest will remain limited. The leaves become pale and may wilt and fall off. Whiteflies can also transmit viruses, which in turn can cause a lot of damage to plants (Djomaha and Ghogomu, 2016).

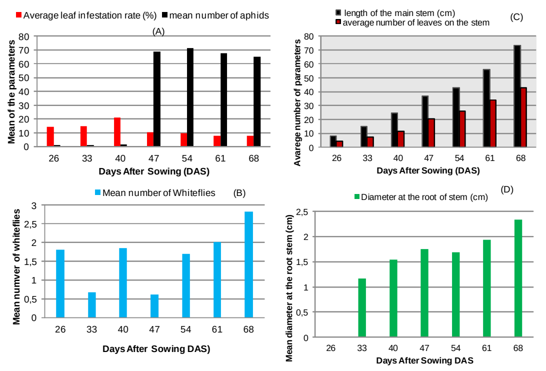

Fig. 2. Evolution of the populations of aphids, leaf infestation rate (A), *Bemisia tabaci* (B), length and leaves on the stem (C) and diameter at the root stem (D) according to time.

In terms of growth parameters, Fig. (Fig.2C) shows that stem length and average leaf number increase gradually over time to 68 DAS without interruption. Watermelon is an indeterminate growth plant and the increase in leaves can influences fruit size and weight.

The diameter at the collar (Fig. 2D) increases gradually from 26DAS to 54DAS where there is a slight drop then goes up gradually until flowering. This drop coincides with the peak of aphid numbers (Fig.2A). The raw sap comes from the roots and transits through the stem to reach the leaves. It is therefore said that the suction of the aphids combined with that of the whiteflies present on the leaves would have disturbed the circulation of the sap and would have reduced the diameter of the stem by a few centimeters.

In general, the increase of aphids population is due to parthenogenic reproduction helping to reduce the duration of generation compare to sexual reproduction for whitefly (Dixon 2007). Their high fertility rate ranging from 7 to 29 days, played on the significant multiplication of the population of aphids on the leaves. This information corroborates the results of (Djomaha 2018) on the asexual reproduction of aphids on cabbage crop.

Evolution Aphis gossypii at its different stages of development

The following fig. (Fig.3) shows the curves of aphid nymphs and adults.

Adults were more numerous than nymphs. From 26 DAS onwards, individuals are present but their number remains constant until 40 DAS when there is a strong expansion. The curve of the adults being always situated above that of the nymphs. This result shows that the nymphs and the adults cause the damage on the watermelon. The expansion of the numbers would be due to the increase of stem length with more the leaves as observed in fig. 2.

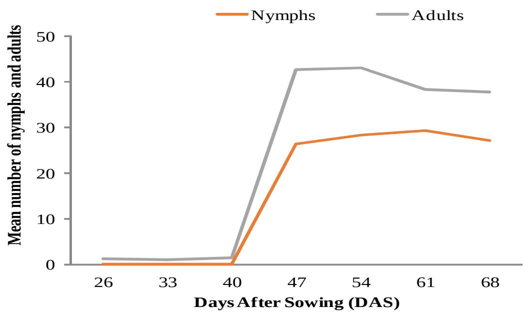

Fig. 3. Evolution of nymph and adult populations of *Aphis gossypii*"according to time.

Evolution of Bemisia tabaci populations at its different stages of development

The following fig. (Fig.4) shows the evolution of the whitefly at its different stages of development.

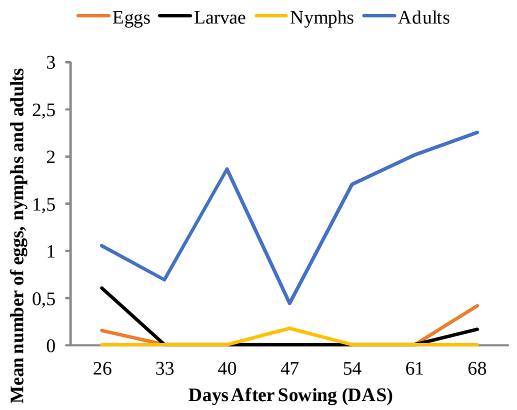

Fig. 4. Evolution of the average eggs, nymphs, larvae and adults of *Bemisia tabaci*.

Generally, all stages of the whitefly are present from the eggs to the adults.

Specifically, adults are numerous throughout the trial period, followed by larvae, eggs and nymphs. The peaks are reached on 68 DAS, except that of the nymphs, which is reached on 47 DAS. In summary during the period of this trial, in the case of the white fly, the adults cause more damage to the plant.

Abundance of aphids, whiteflies and average leaf infestation rate (%) per treatments and time

In general, all treatments have average parameters from the beginning to the end of the trial.

The fig. (Fig. 5A) below shows the control curve above the Antario curve and the other three curves merged on the x-axis. Peak numbers of aphids is at 47 days after sowing and numbers increase in control plots until flowering unlike those treated with Antario. This shows that the treatment against aphids is necessary and Biotrin, neem oil and Emacot significantly reduced aphid numbers in the field.

As for the whitefly (Fig. 5B), the numbers were not important compared to aphids. The curves evolve progressively but take a big jump at flowering (68 days after sowing). At that point, the control curve is higher than the other curves. This indicates that whiteflies are more active during the flowering period. They can affect fruit formation and therefore require control method. Emacot was closer to the control curve showing its ineffective action on whitefly as opposed to aphids. The neem oil curve is lower than the other curves followed by Biotrin and antario.

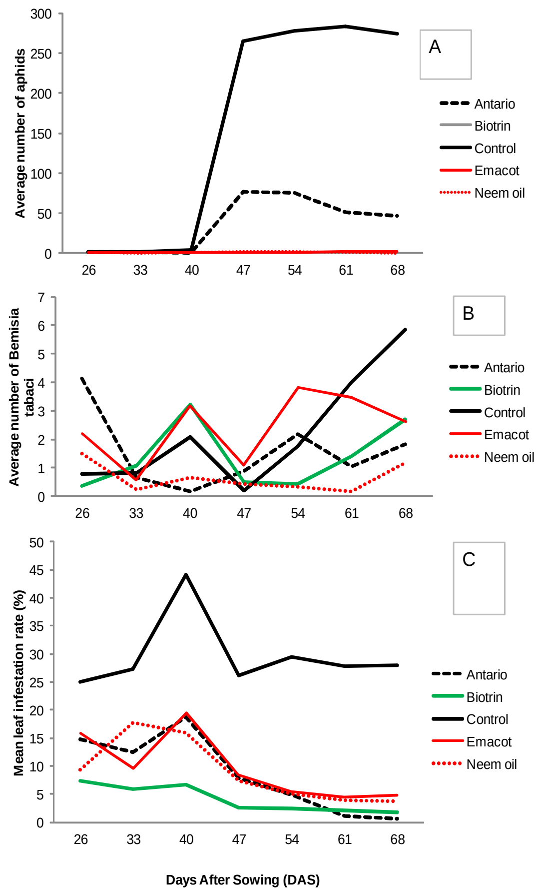

Fig. 5. Evolution of the average number of *Aphis gossypii* (A), *Bemisia tabaci* (B) and mean leaf infestation rate (C) according to treatments and time.

Aphids and whiteflies are piercing and sucking insects and their damage is manifested by leaf curling. The following fig. (Fig.5C) shows that the leaf infestation rate follows a zig zag pattern with a peak at 40 DAS. The control curve is higher than the others and the Biotrin curve is the closest to the x-axis during the whole sampling period. This expresses a proved efficacy of Biotrin on both pests against the three other insecticides. The curves of Antario and neem oil are similar to the one of Emacot.

Consequently, the population is reduced to the level of all other treatments and slightly to the level of T1 negative treatment. This reduction in the infestation rate at the 5th collection would be linked on the one hand to the return of rains which would have had an influence on the pests. These results go hand in hand with those of (Djomaha and Ghoghomu, 2016) which show that water droplets wash cabbage pests. In the same vein, (Leslie *et al.*, 2009) who worked on cabbage aphid, have shown that, rains lead to high mortality of nymphs in the rainy season and on the other hand to the fact that T2, T3, T4 and T5 treatments have been effective to aphids. The best treatment against this aphid would therefore be T5 positive control which records a general average of 0.9 unlike the negative control T1, which records up to 78.90, then compared to these two controls, T2 is the best treatment, then T4 and finally T3.

Watermelon growth parameters per treatment

The table 1 below shows that the Emacot treated plots showed the most significant values for all four watermelon growth parameters. The coiled leaved are reduce, the number of leaves are high, the diameter at the stem root is great and stem length is more important than in any other plot. The plots treated with Biotrin and neem oil, which are comparable to each other, came after Emacot and recorded significant values compared to the untreated plots. In untreated plots, there are more curled leaves, fewer leaves, shorter stem length and reduced diameter at the stem root. Antario is the treatment that presents similar values as the control except for the average number of coiled leaves where it is classified with Emacot, neem oil, Biotrin and Antario. These three bioinsecticides are thus detectable as alternatives to Emacot the chemical insecticide.

Table 1. Average length of main stem, diameter at the root of stem, average number of leaves en curled leaves per treatment.

Treament	length of the main stem (cm)	Diameter at the root of stem (cm)	average number of leaves on the stem	Average number of curled leaves
Control	30.50±2.07b	1.37±0.07a	17.69±1.15b	5.85±0.54a
Biotrin	40.80±2.98ab	1.39±0.06a	21.54±1.49ab	0.64±0.11b
Antario	32.97±2.29b	1.48±0.15a	18.53±1.35b	0.91±0.12b
Neem oil	37.80±2.21a	1.59±0.06a	21.83±1.21ab	1.14±0.17b
Emacot	41.10±2.54a	1.62±0.07a	24.74±1.37a	1.51±0.17b
	F=7.79 P=0.0001	F=3.11 F=0.01	F=4.58 P=0.0001	F=63.29 P=0.0001

a, b: values affected with the same letters in the same column are not significantly (P>0.05) different.

Yields per treatment

Yield per treament

The following table (table 2) shows the average fresh weight yields (t/ha) of watermelon fruits according to treatments. Watermelon produced in all plots. The statistics show that the means are not significantly different (P=0.05%). However the greatest yields were registered in the Emacot and Biotrin parcels followed by Antario and neem oil plots. The lowest yield was in the untreated one.

Table 2. Average total yields (t/ha) of watermelon fresh fruits per treatments.

Treatment	Average total yield (t/ha)
Control (T1)	35.71±2.66a
Biotrin(T2)	39.85±1.84a
Antario(T3)	39.50±1.86a
Neem oil(T4)	37.23±1.58a
Emacot(T5)	40.21±2.28a

a, b: values affected with the same letters in the same column are not significantly (P>0.05) different.

Without insecticides, the infestation of the leaves by aphids and whiteflies has high so plant. This has reduced the leaf area, resulting in a reduction in photosynthesis because the light that reaches the leaves is insufficient. The growth being slowed down, this will play on the filling of the fruits (their weight inkg). This result coincides with the work of El Shafie *et al.* (2012) which shows an increase in yield losses as the leaf infestation rate increases. Taking into consideration the data in Fig. 7, Biotrin is therefore the most appropriate biological insecticide that can be alternated with Antario for aphids or with neem oil for whitefly.

Conclusion

The work carried out in this trial focused on evaluating the effectiveness of integrated pest management, combining three bioinsecticides and a chemical insecticide against *Aphis gossypii* and *Bemisia tabaci*. At the end of this study, the results obtained revealed that:

Aphid and whitefly populations were present from the beginning to the end of the trial. Untreated plots had more infestation, more curled leaves, fewer leaves, shorter stem length, and smaller collar diameter. This resulted in a decrease in fresh weight yield of watermelon. The best yields were obtained with Emacot followed by Biotrin and neem oil. These two bioinsecticides can be applied alternately with Emacot in an IPM program.

The most sensitive period for watermelon is before 40 DAS to both pests attacks and after 68 DAS to whitefly damages. So control measurements to limit the damage on plant should be taken before that period.

Farmers should use neem oil because not only is it organic but also, it is a local product which gives an advantage at the economic level given the price of imports of biotrin.

Africa IPM should continue the extension of biotrin to producers. The Ministry of Agriculture and Rural Development of Cameroon should register biotrin and reduce the cost to encourage producers who are not in the high production area of neem oil.

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