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EFFECT OF BIOCIDAL SUBSTANCES ON POPULATION DYNAMICS OF *PLUTELLA XYLOSTELLA* LINNAEUS, 1758, AUXILIARY INSECTS, AND CABBAGE PRODUCTION IN THE SUBURBAN AREA OF DAKAR

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ABSTRACT

This study was conducted to determine the effects of biopesticides, used in the control of *Plutella xylostella*, major bioaggressor of cabbage, on its auxiliaries as well as on cabbage quality. It took place in a field of 65 m2, located in Malika, peri-urban area of Dakar. Treatments, based on *Crateva religiosa* (200g/l), *Calotropis procera* (400g/l), a mixture of the two products, and the control, were performed every two weeks on a random block device. Analysis of the results was done with the Rstudio software. Results showed that untreated plots were more infested with *Plutella xylostella* larvae (p-value = 0.01271). Efficacy of *Calotropis procera* on *P. xylostella* larvae has been demonstrated. The effect was all the more interesting since auxiliaries were not affected by treatments, as the regular sampling revealed. A satisfactory agronomic result was obtained at the end of this test, with as indicator, a much larger harvest from the treated plots.

Contribution/ Originality: This study contributes to strengthen the number of plants that could be used in the biological control of the main cabbage pest *Plutella xylostella.* Calotropis *procera* and *Crataeva* religiosa leaf-based biocidal products are added to those used against *Plutella xylostella.*

1. INTRODUCTION

Agriculture is a very important sector in Senegal's economy. It includes horticulture, which is the main sector strongly concerned with market gardening. Among crops, Brassicaceae represent an important part, with an annual production of 152 million tons of cabbage, cauliflower, and rapeseed over 37 million hectares in 2011 [1]. In West Africa, especially in Senegal, cabbage is grown over 3500 hectares with an estimated annual production of 60,000 tonnes [2]. Cabbage is currently one of the most cultivated vegetables and, with its short life cycle (about 60-90 days after transplanting), is an important source of income for rural populations. Behind Niger, Senegal is the second-largest producer of cabbage in West Africa [3]. Despite the importance of this crop, its production is heavily affected by a number of pests, the main of which is commonly known as the "Cabbage Moth", *Plutella xylostella* [4]. Larvae of this micro-Lepidoptera are defoliating and can cause up to 90% loss of production [5, 6]. In Senegal, this rate is estimated to be between 51% and 94% [7]. Consequently, market gardeners, in order to protect their crops, use chemicals in an utterly disastrous way. Unfortunately, unwarranted use of chemicals is becoming a threat to human health, the environment and auxiliary wildlife [8]. Given this pressure, it turned out to be necessary to find alternatives to synthetic chemical insecticides [9]. On that account, other methods of control are developing, favouring a sustainable management of agro ecosystems. They prevent from economic damage and preserve the environment and human health [7]. Currently, recourse to insecticides of biological origin such as plant extracts is attempted. The most effective against *P. xylostella* and the least harmful to auxiliary wildlife appears to be the extract of *Azadirachta indica* [10]. However, there are other formulations such as those based on *Calotropis procera*, which has appeared highly poisonous to *Caryedon serratus* [11]. In order to encourage producers to adopt biological control practices, it is necessary to provide them with effective and economically viable strategies. In accordance with this context, the purpose of our study is to contribute to the improvement of the *P. xylostella* cabbage moth biological control using bio-insecticides based on extracts from *Calotropis procera* and *Crateva religiosa* leaves.

2. MATERIALS AND METHODS

2.1. Presentation of the Study Area

The commune of Malika covers an area of 805ha 24 Ares 17 CA, that is to say 9.4% of the territory of the Department of Pikine. It is located in Niayes belt, a subdivision of the city of Pikine, and 22 km from Dakar. It is limited to the east and south by the arrondissement of Keur Massar, to the west by Yeumbeul North and to the north by the Atlantic Ocean Figure 1.

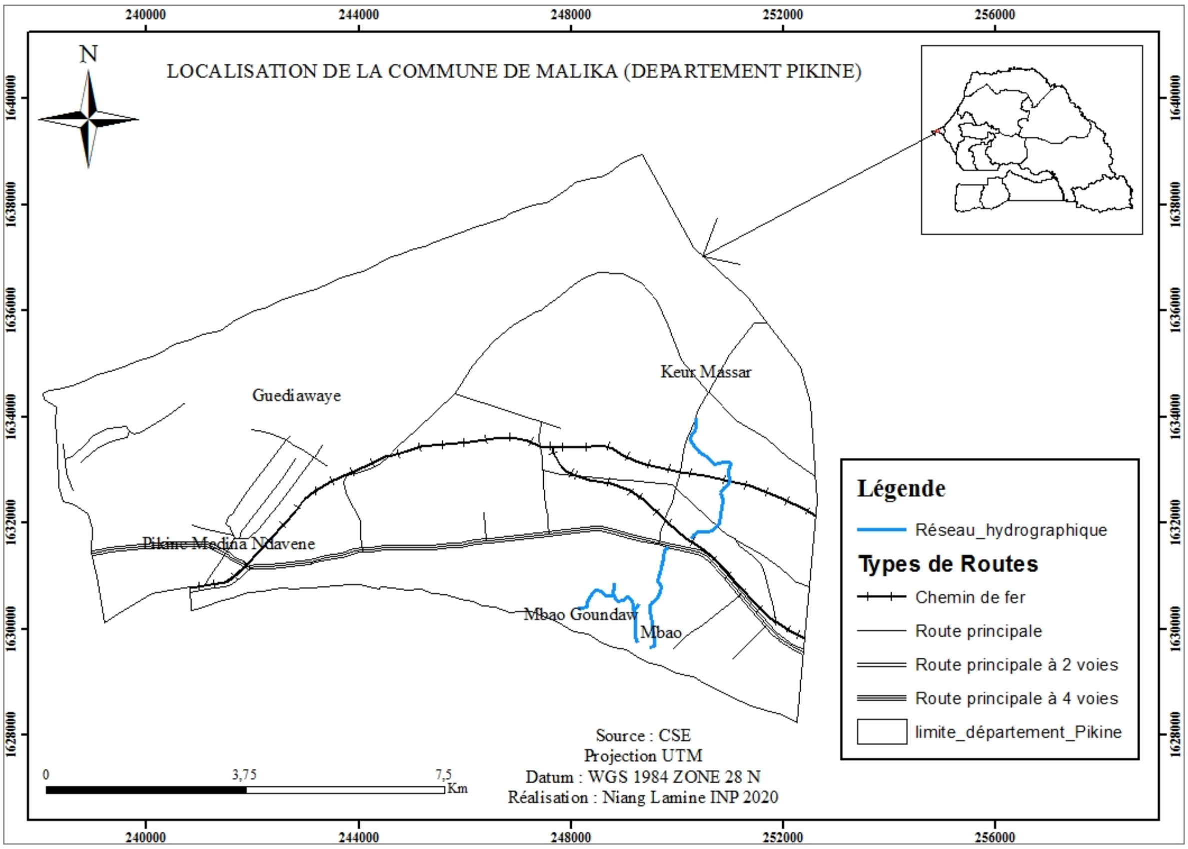


Figure 1. Location of Malika commune.

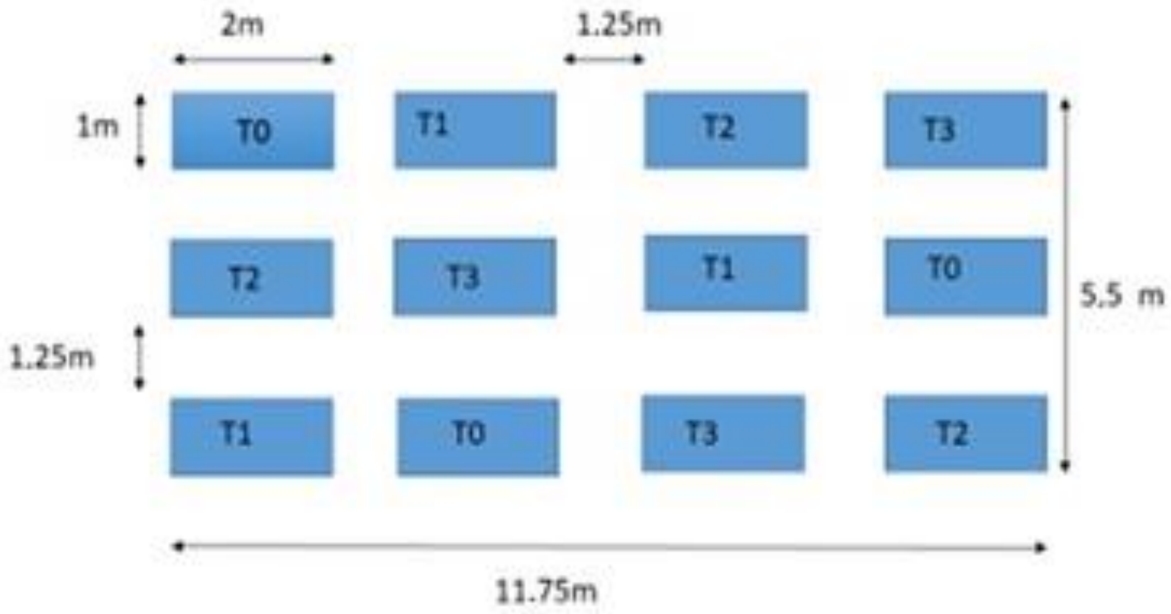
2.2. Biological Materials

In this study, we used *Brassica oleracea* (Copenhagen market variety: grown in dry season, it is one of the varieties commonly used in the country), *Plutella xylostella*, and *Plutella xylostella* auxiliaries as biological material.

3. METHODS

3.1. Experimental Device

Our experimental device Figure 2 consists of complete random blocks with four treatments and three replicates, for a total of 12 experimental plots. The latters were numbered from T0 to T3 depending on the applied treatment. T0 is the control that received only water, T1 was treated with the *C. religiosa* solution at a concentration of 200g/l [12] T2 was treated with the *C. procera* solution at a concentration of 400 g/l [13], and T3 was treated with the mixed solution, which consists of a combination of the two biocides. In each elementary plot, there were 3 lines of 7 plants (numbered from 1 to 7), for a total of 21 plants. The spacing was 10 cm between lines and between plants. The surface of each elementary plot was 2 m2 (1m by 2m) with 1.25 m of spacing between them to manage biocidal losses. All the elementary plots had the same cabbage variety but with different treatments. The block was 11.75 m long and 5.5 m wide, for a total surface of 65 m2.



5.5 X 11.75 = 65 m2

Figure 2. Fisher experimental device.

3.2. Farming Practices

Activities began on November/16 /2019 with cabbage seeding. The nursery was set up in alveoli containing substrate for fertilization. It has been protected by mosquito net panels to limit insect attacks. During the three weeks corresponding to the duration of the nursery, a diagnosis was made two days a week to monitor germination and attacks. Healthy and vigorous plants, aged 25 days, were transplanted in elementary plots previously ploughed.

3.3. Field Transplanting

The field was made-up in December 2019 by measuring an area of 60 m2 divided into 12 elementary plots of 2 m2 each. Using a watering can, a volume of 10 L of water was poured into each elementary plot. After a few minutes, for the soil to moisten, the spacing between cabbage stalks was measured by taking 40 cm spacing of either 21 feet per elementary plot, corresponding to a total of 252 cabbage plants.

3.4. Preparation of Bio Pesticides

The fresh leaves of *Calotropis procera* and *Crateva religiosa* were harvested between 6am and 7am. In fact, active substances can be destroyed beyond these hours by the sun's rays. The leaves of *C. religiosa* were harvested in the

UCAD botanical garden and those of *C. procera* around the VDN road. The preparation took place as follows: weighing, rinsing and then mixing the leaves with a blender before mixing them in a solvent. After harvest, for every 200 g of fresh *C. religiosa* leaves, a volume of 1L of water was added. Then the mixture was strained after 24 hours of rest. Similarly, the *C. procera* leaves, after weighing, were ground in a blender and then macerated in a plastic bucket for 24 hours at a concentration of 400 g per litre. The solvent used for the extraction of biocidal substances was tap water (Tw). The mixture was filtered using a fine mesh sieve (0.01 mm 0.01 mm). The finished product was kept in plastic bottles.

3.5. Phytosanitary Treatments

These treatments, aqueous extracts from fresh leaves of *C. procera* (400 g/l) and fresh leaves from *C. religiosa* (200 g/l) based, were applied with a sprayer 15 days after transplanting, and then repeated every 15 days until the 15th day before harvest. The treatments took place late at night. The biocide application doses were 1l/2m2. Watering of the plants was regularly done.

3.6. Observations and Inventories

Observations on insect populations were made two days before and after each treatment to see how the caterpillars evolved in terms of the treatment. Counts of caterpillars, cabbage moth pupae, and their auxiliaries were conducted twice, before and after each treatment. This inventory was done randomly selecting ten feet on each elementary plot and for each formulation, including the control. For each selected foot, the number of *P. xylostella* larvae and predators present was recorded and pupa samples collected. These were taken to the UCAD Entomology Laboratory where they were incubated, in order to identify emerging parasitoids.

This enabled us to determine the abundance and frequency of occurrence of *P. xylostella* and its auxiliaries.

3.7. Estimation of Damage and Determination of Agronomic Parameters

To quantify the damage, the Nagawa [14] scale was used:

1 = no damage.

2 = < 25% of leaf surface destroyed.

3 = 25 – 50% of leaf surface destroyed.

4 = 51 – 75% of leaf surface destroyed.

5 = > 75% of leaf surface destroyed.

Observations permitted identifying the following parameters:

- Biological efficiency was calculated from the formula:

Biological Efficiency = [(Number of Caterpillars or Damage (Untreated) – Number of Caterpillars or Damage (Treated)) *100/ Number of Caterpillars or Damage (Untreated)] [15].

- Incidence of attacks: It was determined using the following formula:

I% = PA/PT*100.

I% = percentage incidence; AP = number of plants attacked; PT = total number of plants.

Relative Abundance: it is defined as the ratio of the species' abundance (ni) to the total number of individuals of different stand species (N):

$$Pi = ni/N$$

The percentage of loss at harvest (%) was calculated by subtracting the average weight at harvest from the average marketable weight (kg) of cabbage heads without leaves damaged by caterpillars.

3.8. Statistical Analyses

Microsoft Excel 2013 spreadsheet software was used to classify the data obtained in the field and, through its dynamic cross-tabulation, to generate the histograms. Analysis of the results was done with the Rstudio software. At the basis of any analysis, a normality test of data was carried out with the Shapiro-Wilks test. Following this test, data do not follow the normal law; that is why we will use nonparametric tests for the significance between the different treatments. Effect of the treatment on the larvae of *Plutella xylostella* and on the weight of the harvest was determined with the Kruskal Wallis test. The latter, having been significant, was completed by the Man-Withney-Wilcoxon two-to-two comparison test.

4. RESULTS AND DISCUSSION

4.1. Results

4.1.1. Inventory of Species Encountered in the Environment

Throughout the duration of the test, 300 individuals were surveyed and distributed among six (6) species. The majority species was *Plutella xylostella*: 174 individuals with an abundance of 58%, followed respectively by Syrpha with 82 individuals (27%), Spiders with 17 individuals (6%), Ladybugs with 16 individuals (5%), Ants with 8 individuals (3%), and *Cotesia plutellae* with 3 individuals (1%) Table 1.

Table 1. Abundance of bio aggressors and auxiliaries.

Species	*P. xylostella*	Syrpha	Spider	Ladybugs	Ants	*C. plutellae*	Total
Number (ni)	174	82	17	16	8	3	300
Relative abundance (Pi) %	58	27	6	5	3	1	100

4.1.2. Biocidal Effects of Botanical Extracts on Plutella Xylostella

Analysis of Figure 3 revealed that *P. xylostella* populations were more abundant in the control plot than in the treated ones. Thus, *Crateva religiosa*, *Calotropis procera* and the mixture of these two products made it possible to maintain the infestation by *Plutella xylostella* at a lower level. However, the Kruskal Wallis test revealed a significance between the different treatments used (p-value = 0.01271). Similarly, the results obtained with the Wilcoxon test showed a significant difference between T0 and T2 (p-value=0.0081) but no significant difference between T0 and T1 (p-value=0.3970), T0 and T3 (p-value=0.2284), T1 and T2 (p-value=0.3970), T1 and T3 (p-value=0.6683), T2 and T3 (p-value=0.4344).

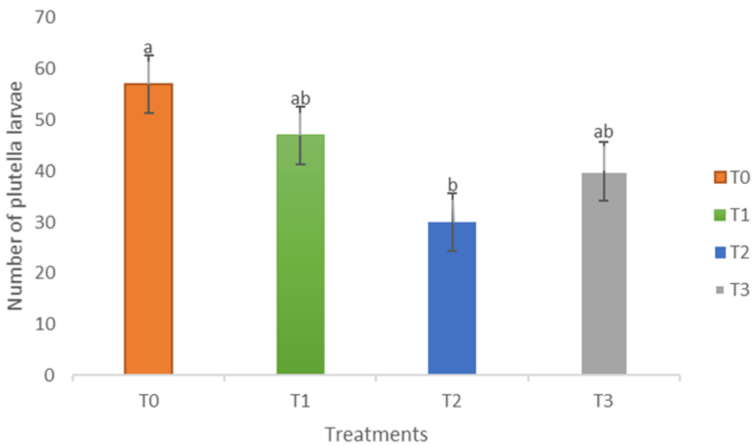


Figure 3. Effect of botanical extracts on larval numbers of *Plutella xylostella*.

4.1.3. Biocidal Effects of Botanical Extracts on the Auxiliary Population

The results presented are based on observations of predators and parasitoids.

4.1.4. Effect of Botanical Extracts on Predators

Table 2 shows the effect of treatments on predators. Analysis of the data obtained with the four species of predators encountered (syrpha, ladybugs, spiders and ants) does not detect any difference between the treated plots (*Crateva religiosa*, *Calotropis procera*) and the control.

Table 2. Effect of bio pesticides on predators.

Treatments	Mean±Sd of Syrpha	Mean±Sd of Ladybugs	Mean±Sd of Spiders	Mean±Sd of Ants
T0	0.083 ± 0.59	0.025 ± 0.20	0.020 ± 0.14	0.004 ± 0.06
T1	0.116 ± 0.93	0.012 ± 0.11	0.020 ± 0.14	0.012 ± 0.14
T2	0.133 ± 1.03	0.008 ± 0.09	0	0.016 ± 0.20
T3	0.008 ± 0.091	0.020 ± 0.19	0.029 ± 0.16	0
P- value	0.5043	0.8765	0.09365	0 .5295
Significance	NS	NS	NS	NS

Note: Sd: Standard deviation.

4.1.5. Effect of Botanical Extracts on Parasitoids

Nymphs of *Plutella xylostella* were weakly parasitized. Parasitoids that emerged from the nymphs evolved from the control and mixture of Calotropis procera and Crateva religiosa (Table 3). *Cotesia plutellae* is the only parasitoid identified in this study.

Table 3. Parasitoid tracking survey.

Total of nymphs	Total emergence of *Plutella xylostella*	Total of parasitoids (*Cotesia plutellae*)	Number of aborted nymphs
14	7	3	4

4.1.6. Biological Efficacy of Natural Compounds for Control of Plutella Xylostella

Application of the different biocides resulted in a reduction in the level of infestation of Plutella xylostella. Thus, *Calotropis procera* treatments have reduced the infestation levels of the *Plutella xylostella* caterpillar by 47% Table 4. While *Crateva religiosa* showed an ability to reduce infestation by 18%. The mixture of the two biocides had an efficiency of reducing infestation levels of Plutella xylostella by 30%.

Table 4. Biological efficacy of botanical extracts on the Plutella xylostella caterpillar.

Traitements	Efficacite Biologique (%)
Crateva religiosa	18
Calotropis procera	47
Crateva religiosa + Calotropis procera	30

4.1.7. Effect of Treatment on the Number of Leaves Attacked

Incidence is the rate of plants with Plutella xylostella damage. Thus, untreated plots had a high incidence of 17.4%, unlike treated plots where the rate of attacked plants was 7.2% for the Calotropis procera, 7.5% for the Crateva religiosa, and 10.61% for the mixed Table 5.

Table 5. level of damage to the plant over the different bio insecticide treatments.

Treatments	Incidence (%)
Non treated	17.4
Calotropis procera	7.2
Crateva religiosa	7.5
Calotropis procera+ Crateva religiosa	10.6

4.1.8. Effect of Treatment on Harvest Weight

Out of a total of 252 transplanted feet, 110 cabbage heads were harvested, the rest being attacked by rodents, aphids, *Hellula undalis*, *Hecoverpa armigera*, *Plutella xylostella* and *Spodoptera littoralis.* Head-free feet were identified in this study. The total harvest weight was estimated at 26.2 kg. The histogram below Figure 4 shows the effect of different biocides on cabbage weight at harvest time. It can then be seen that the largest crops estimated at 12.3 Kg and 8 Kg came from plots treated with Calotropis procera and the mixture of *Crateva religiosa* + *Calotropis procera.*

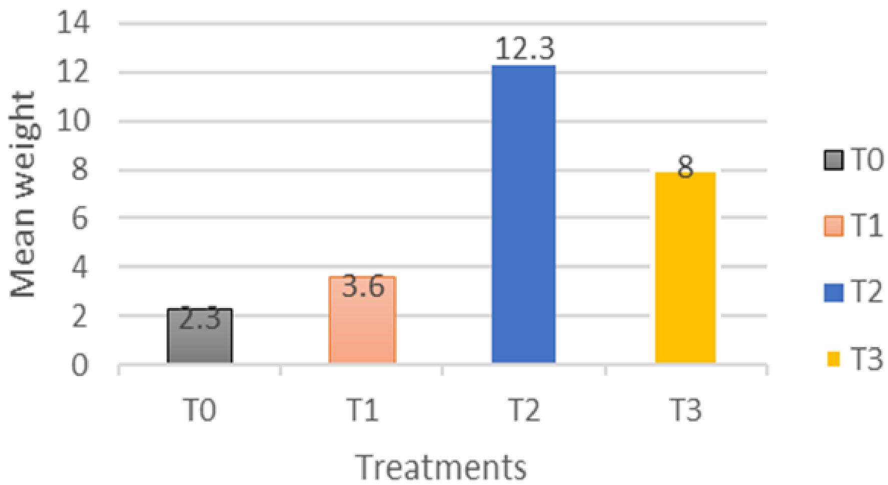


Figure 4. Effect of products on crop yield.

5. DISCUSSION

5.1. Effects of Treatments on Plutella xylostella

During this study, several insects were inventoried. *Plutella xylostella* kept its primary pest status due to the significant increase in its population during the last week of monitoring.

According to Silva-Torres, et al. [8] it represents the most dangerous destroyer among cruciferous parasites worldwide. Prior to treatment, when cabbage plants were relatively young, *P. xylostella* was the most abundant species in the plot. According to Sow, et al. [7] females of *P. xylostella* have a preference for young plants during spawning. This can be related to cabbage content in glucosinolate, which increases when the plant develops, and this would function as a limiting factor in female spawning [7]. These results corroborate those of Ngom, et al. [13] which illustrate that aqueous extracts of *C. procera* leaves have shown efficacy on *P. xylostella* larvae. This efficacy of different extracts of *C. procera* has also been demonstrated on *C. serratus* eggs by Thiaw [11]. *Crateva religiosa* is effective, but its effect is slower than that of *Calotropis procera.* It is the same for the mixed product. According to the study of Faye [16] an efficacy of *Crateva religiosa* has been shown against the bean beetle, *Callosobruchus maculatus.*

5.2. Effect of Treatments on Auxiliary Wildlife

5.2.1. Effect of Treatments on Plutella xylostella Predators

Syrphidae, general predators, are more represented on untreated plots than on treated plots. This trend is explained by the fact that the availability of this predator depends on the availability of its prey, of which *Plutella xylostella* is more common in untreated plots. Their rising number over time could also be explained by the increase in glucosinolate levels in cabbages that could attract *P. xylostella* (females for laying and males for mating). However, their number dropped in the last day, due to the scarcity of *Plutella xylostella* because of the quite entire disappearance of the glucosinolate.

5.2.2. Effect of Treatments on Parasitoids

Cotesia plutellae was the only parasitoid that emerged during this study. Thus, *Cotesia plutellae* were recorded on the control plots and one individual was encountered on T3. Being a parasitoid of *Plutella xylostella*, it is likely to be found where its prey is more present and this, in order to multiply and perpetuate the species. In our test, we found that a small number of Cotesia plutellae. However, although this number is very limited, they were all observed in the final days of sampling. This could be explained by the fact that they would have been the target of other insects they managed to exterminate in the last days of sampling.

5.3. Effect of Treatment on Cabbage-Heads Quality and Maturity

Analysis of agronomic data reveals that untreated plots had a higher infestation level of P. xylostella and lower yield. Our results are similar to those of Sow, et al. [7] which showed that agronomic parameters are heavily affected by the infestation level of *Plutella xylostella.* The inversely proportional evolution of leaf and crop weight impacts has shown that larval attack of *P. xylostella* could indirectly impact the weight of the crop and therefore yield. The larger number of leaves attacked in the control plots, as well as the number of *P. xylostella* larvae, would indicate the need for treatment for better yield. The application of *Crataeva religiosa*-based phytosanitary treatment appears to have an effect on harvest yield, which is consistent with studies by Amoabeng, et al. [17] which have shown that natural products from plants can also increase yields with a cost/benefit ratio comparable to that of synthetic pesticides. An accelerated effect of ripening of cabbage is noted in the treated plots compared to the control ones. We can deduce from this that the treatment with *Calotropis procera* and *Crateva religiosa* seems to have a positive effect on yield.

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Authors' Contributions: All authors contributed equally to the conception and design of the study.

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