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Use of Aqueous Extract and Essential Oil of Citrus Aurantifolia Leaves in the Protection of Vegetables

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Summary

Our study is devoted to the valorization of the essential oil and the aqueous extract of Citrus aurantifolia leaves as biopesticides. The extraction of oil from the leaves was produced by hydrodistillation. The aqueous extract of the leaves was obtained by maceration. The insecticidal activity of the essential oil and the aqueous extract of the leaves of the plant was tested by spraying on two plots containing 10 Ndrowa eggplant plants. (*Solanum aethiopicumm gilo*) each. Leaf oil extraction yields 1.02%. On the plants sprayed with essential oil and aqueous extract of lemon tree leaves, it was observed a very significant reduction in insect pests of eggplant crops (less than 13% and 2% respectively for plots B and C). The havoc on the parties respectively with the aqueous extracts and the essential oil of the leaves of the lemon tree. The damage to plants in the treated plots was 13% and 3% for plots B and plots C respectively. Regarding the fruit conditions of Ndrowa eggplants, the frequencies of good fruits were 96%, 87% and 75% respectively for plots A, B and C. Regarding yield, plots treated with essential oil (C) and the aqueous extract (B) of lemon leaves were respectively 44% and 38% compared to 18% for the control plot (A). These oils are therefore better agents for protecting and preserving vegetables and fruits. Further study would be necessary to determine effective doses by working on a large population of insects and eggplant plants.

Keywords: Essential oil, Ndrowa eggplant, Citrus aurantifolia, Hydrodistillation, Insect, Solanum Aethiopicumm gilo

Introduction

Agriculture is one of the main sectors of activity which contributes to the socio-economic development of populations (more than 52%) in Africa [1]. Côte d'Ivoire, a country in West Africa, is an agricultural country. Its economy is mainly based on agriculture. Alongside cash crops, food crops including market garden products occupy an important place in the daily diet of populations [2,3,4]. Market gardening plays a key role in most nutrition and poverty reduction programs, fight against poverty and contribute significantly to family income [5,6]. However, the cultivation of food crops is experiencing increasing difficulties which are affecting its level of production. These include diseases and parasites (insects, fungi, viruses) which affect yield [7,8,9]. The fight against crop pests until now has been carried out using chemicals from plants of different crops [10,11]. Unfortunately, the use of pesticides in agriculture is not without consequences. The environment can be contaminated by these substances [12]. They lead to the destruction of useful species and constitute a major risk of human and animal poisoning [13,14]. Today, chemical pesticides which are undeniably effective reveal their limits through perverse effects on the ecosystem, fauna, flora [15,16]. Furthermore, the handling of chemicals by populations without any prior training are factors that expose not only the health of the consumer but also that of the producer [15]. It is therefore necessary to seek alternatives to the use of synthetic products [17]. The use of pesticidal plants is likely to significantly reduce pests and the need for synthetic pesticides [18]. Indeed, many plants are known and used for their biocidal activities (toxic, repellent, anti-feedant) against a wide range of pests. They can be used in the form of plant extracts for foliar protection [19]. Essential oils or whole plants are also used in stored food attics [20].

It is for this reason that in Ivory Coast, like many countries, studies are being carried out to find alternatives to chemical pesticides. Our study is part of the valorization of natural substances in the treatment of agricultural products against parasites harmful to market gardening crops.

The main objective is to bring food crop producers in rural areas to the use of biopesticides as alternatives to chemicals in crop pest control. It is with this in mind that our research aims to contribute to the management of pest parasites in eggplant crops by promoting the leaves of Citrus aurantifolia, a plant resource available in all rural areas of Côte d'Ivoire. This specifically involves validating the effectiveness of the aqueous extract and essential oil of Citrus aurantifolia leaves on the main pest parasites encountered on eggplant cultivation and evaluating the effect of the extracts on eggplant yield.

Materials and Methods

Study site

The test was conducted in Yamoussoukro (Lakes Region), located between 6°15-7°35 N and 4°40 and 5°40 W and 95 m altitude. The soil of the experimental plot is ferrallitic and has a sandy texture. It is highly desaturated, poor in exchangeable bases, particularly phosphorus, and comes from tertiary sands [7]. The soil is a ferralitic, gravelly and gravelly soil, more or less deep, rejuvenated, clayey-sandy (Ekou and Djidji, 1997). The climate is the Baoulean climate and straddles the pre-forest savannah and the rainforest. The terrain is generally very uneven. The distribution of rainfall is variable and is between 900 and 1100 mm 3. The average temperature is 26°C [21]. The locality has an equatorial climate with two rainy seasons (March to July and September to October) and two dry seasons (November to March and July to September). This hot and humid climate is conducive to the proliferation of fungal diseases and insect pests of vegetable plants.

Cultivation practices

Ndrowa seeds *(Solanum aethiopicumm gilo)* were germinated in a nursery on a small plot of 2 m^2 after plowing the soil. After sowing, the plot was regularly watered. After 40 days in the nursery, the plants were transplanted into three different plots of 25 m^2 each separated by 100 meters. To do this, the plots were cleared, plowed and watered. During this operation, the soil was enriched with a mineral fertilizer based on NPKs, at a rate of 1 g/30 m^2 [22] and disinfected with the different solutions using a 2 L manual pressure sprayer. After transplanting, the plot was regularly watered every morning during the short dry season from July to September. In each plot, 10 eggplant plants (plants are approximately 15 cm high with at least three leaves) were transplanted 0.5 m × 1 m apart.

The transplants were made on July 20, 2021. A week later, the transplanting of the plants, fertilizer was again brought to the different plots. Herbs were removed throughout the experiment. From sowing to the end of observations lasted 4 months including 3 months, one week of flowering and 3 weeks of fruit harvest.

Observations and measurements

In each plot, observations focused on the following parameters: sensitivity to bacterial wilt, the level of attack on the plants, namely attacks on leaves, buds and fruits, the presence of insects, caterpillars…., the total number of fruits harvested, the average weight of the fruit.

The vegetative development of the plants was noted visually on some parameters: Susceptibility to bacterial wilt was assessed by weekly counting of all withered plants from the period of transplanting the plants until the end of the observation. The cumulative rate of wilted plants per week per plot was calculated.

To assess the damage caused by pests, a weekly count of all attacked plants (plants, leaves, buds and fruits) was carried out from the appearance of the attacks. Fruit attacks concerned fruit bores, attacks on leaves and buds concerned shredding and rolling. The cumulative rate of attacked games has been determined. The number of fruits per plant as well as the average weight of the fruit were measured on a scale. Observations relating to the number of insects were carried out during three periods (one week after the second treatment, during the flowering period and during the harvest period). The count is carried out for one week in each period. Data on pests were taken in the mornings between 6:30 a.m. and 9:30 a.m. for three days at different periods (P1, P2, P3) (Fauquet et al., 1987).

Preparation of biopesticide products

Citrus leaves aurantifolia were recovered from citrus trees aurantifolia present in the villages of Yamoussoukro. The aqueous extract was obtained by maceration of 100 g of dried leaf powder of citrus aurantifolia in one liter of water by maceration. Thus, the collected and dried leaves were ground to obtain a fine powder, then this ground material was soaked in distilled water for 24 hours. Finally, this ground material was filtered to obtain aqueous extracts. As for the essential oil of lemon leaves, it was obtained by hydrodistillation in an abininc from the fresh leaves of citrus aurantifolia.

For each of the cultivation plots, eight treatments based on plant extracts and essential oils were compared to the untreated control. Three universal manual pressure sprayers, brand DCRAFT, with a capacity of 2 liters, were used. One was used to apply the essential oil of citrus aurantifolia at a rate of 3 ml/l of water to the plants in plot C, the second to apply aqueous extracts of citrus aurantifolia leaves at a rate of 100 g /l of water on the plants of plot B. The third to be used for the application of liquid soap water on plot A. 1 liter of water was added to each mixture then 10 ml of liquid soap was added respectively to the different mixtures of leaves and essential oil of lemon tree leaves allowing them to attach to the leaves, stems, buds and fruits of the plants. So that the treatment is homogeneous and with the aim of reaching

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a large population of pests. All parts of the plant including leaves, buds, twigs, branches and fruits were sprayed (Bhyan et al., 2007). Furthermore, the jets were oriented so as to cover the lower and upper surfaces of the leaves of each plant, also targeting the underside of the leaves where pests tend to hide. Treatments began on crops just before transplanting, then every week after transplanting, until 14 days before the first harvest.

Data analysis

Data entered into the Microsoft Excel spreadsheet. Graphical representation and data processing were carried out using Excel software. Data were analyzed with Graph Pad Prism 8.0 software. The differences between the means were determined using the Newman- Keuls test at the 5% threshold.

Figure 1: pest insects encountered on the sites.

Results

Plant-destroying parasites identified on the plots

The results of the parasites encountered are presented by the figures (Figure 1/A, B, C, D, E, F, G, H). The pests encountered in the different plots are beetles, jassid beetles, caterpillars, whiteflies, orthoptera and aphids (Figure 1). The whiteflies encountered are whiteflies and blackflies. The aphids encountered are the genera Aphis, Myzus , Mcrosiphum . The orthopterans encountered are grasshoppers, locusts and crickets. Beetles include beetles, weevils, ladybugs and black ground beetles. Caterpillars are the most numerous pests with a frequency of 22%. They are followed by whiteflies and beetles (19%), then aphids (17%) (Figure 1A).

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The last groups of parasites encountered in the plots are orthoptera and jassids with respective frequencies of 12% and 11%. The beetles experienced considerable growth ($p < 0.05$) from the first period p1 (15%) to the third period p3 (23%). On the other hand, aphids, whiteflies and caterpillars experienced a non-significant regression from period p1 to period p3 ($p> 0.05$) (Figure 1/C,D,E,F,G). Regarding the presence of parasites on each of the plots, on plot A, which is the control plot, the different parasites were strongly encountered during the three periods with frequencies very significantly higher ($p < 0.0001$) than those of plots B and C treated respectively with the aqueous extract of the leaves of C. aurantifolia and the essential oil of the leaves of the plant.

On plot B, the frequencies of parasites found on site were less than 13% during all periods. Orthoptera are the parasites most encountered in this plot with frequencies of 12.7%, 6.83% and 3.8% respectively at the different periods p1, p2 and p3.

As for plot C, parasite frequencies were less than 2%. Jassids and aphids were absent during the p1 period on this plot.

A : Frequency of insects encountered on the sites B : Distribution of insects according to the period C : Distribution of pucerons on the plots D : Distribution of caterpilla on the plots E : Distribution of beetles on the plots F : Distribution of whiteflies on the plots G : Distribution of orthoptera on the plots H : Distribution of jassids on the plots

Effects of aqueous extract and essential oil of Citrus aurantifolia against parasite attacks

The yield of essential oil extraction from Citrus leaves aurantifolia by hydrodistillation was 1.02%. The results of insect pest attacks on crops in the different sites are presented in Figure 2 (Figure 2/A, B, C, D). The control plot was the plot which followed a lot of attacks ($p<0.0001$) during this experiment with a frequency of 87% compared to 10% and 3% respectively for plot B and plot C. The plot was significantly less attacked ($p<0.05$) compared to plot C (Figure 2/A). Regarding plot A, the majority of attacks were leaf attacks with a frequency of 64%. After the leaves, come the attacks of the buds and the fruits with respective frequencies of 23% and 10%. Problems linked to eggplant plants represent the lowest attacks (Figure 2/B). In plots B and C, attacks are dominated by damage to leaves and buds. These ravages are followed by attacks on the fruits with frequencies of 17% and 8% respectively for plot B and plot C. Attacks on the feet were very rarely observed in plots B and C (4% and 2% respectively). Wilted plants were not observed in these two plots (Figure 2/C, D). Statistical analyzes revealed a strong significance between attacks on leaves and buds compared to attacks on fruits and then on eggplant plants.

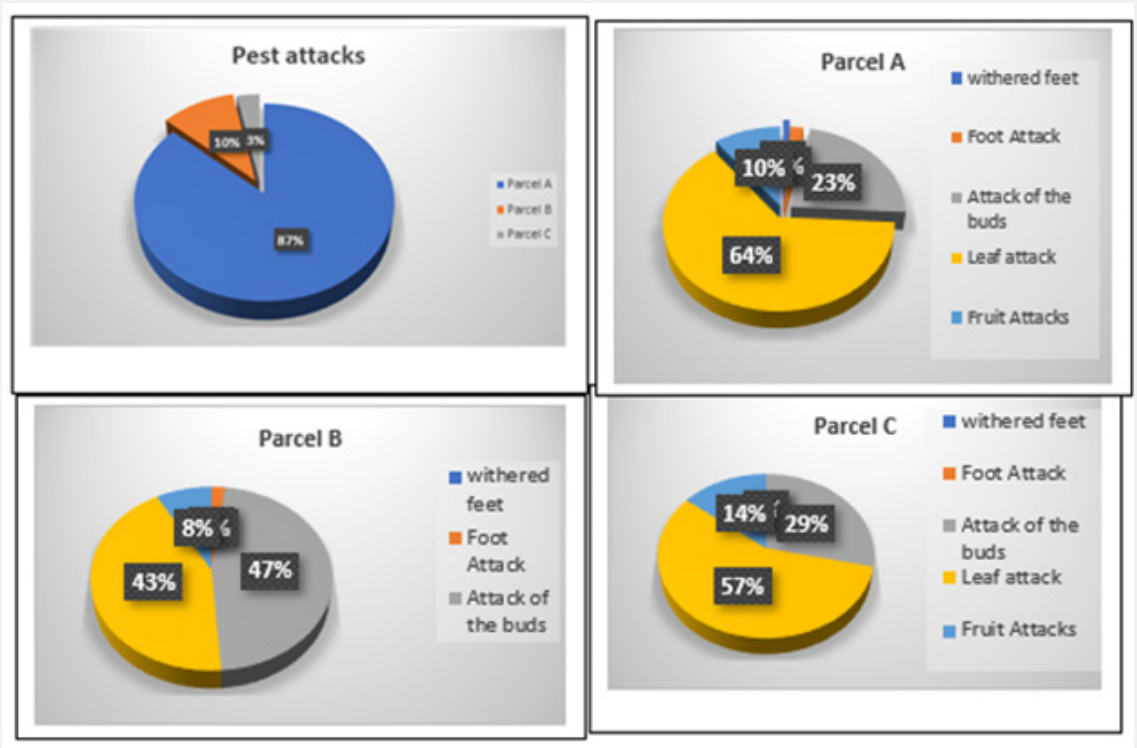

Figure 2: Pests of insects on crops.

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A : Distribution of insect damage on plots B : Insect damage on plot A C : Insect damage on plot B D : Insect damage on plot C

Effects of aqueous extracts of leaves and essential oil of leaves of *citrus aurantifolia* on the yield of different plots

On control plot A, the fruits were 75% in good condition (skin and interior) compared to 87% and 96% respectively for the fruits of plots B and plot C. the analyzes showed a small significant difference ($p< 0.05$) between the good condition of the fruits of plot A and the two other plots treated with *C. aurantifolia* leaves (Figure 3). Statistical analyzes also showed strong significant differences ($p<0.0001$) between good and bad fruits in all plots.

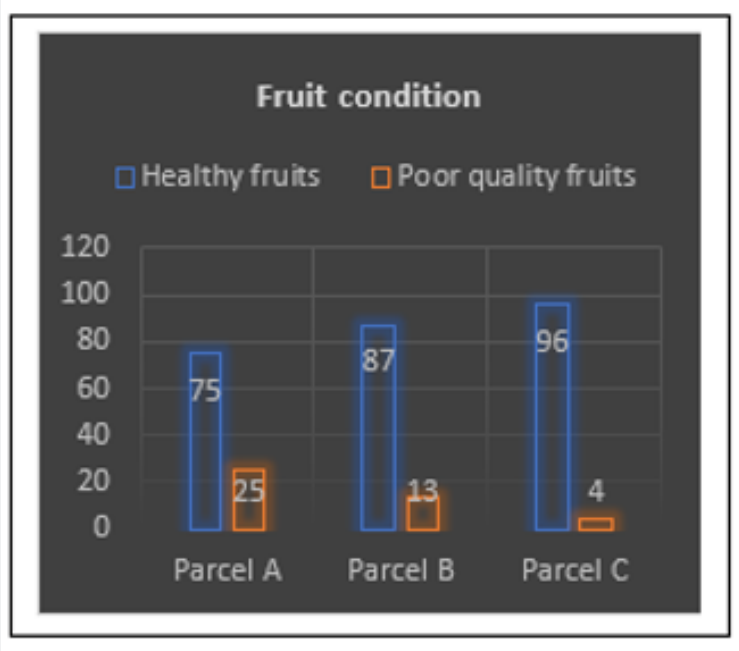

Figure 3: Condition of mature eggplants.

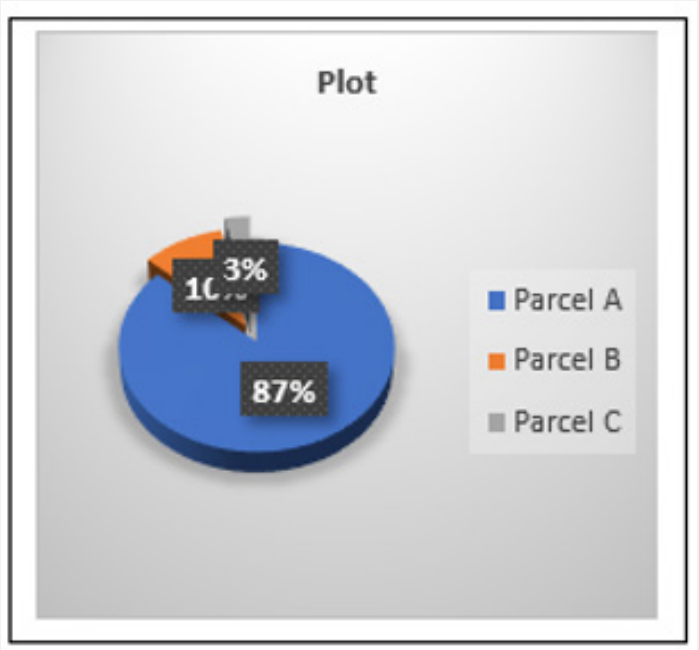

Figure 4: Yield in the plots.

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In terms of yield, plot C had the best yield (44%), followed by plot B with a yield of 38%. The lowest yield was observed in plot A (18%). Statistical analyzes showed a significant difference ($p<0.001$) between the yield of the control plot A and the two other treated plots (plot B and C) (Figure 4).

In terms of average weight of eggplants, the values are between 98 g and 104 g. Statistical analyzes show no significant difference ($p>0.05$) between the average weights of eggplants from the different plots (Figure 5).

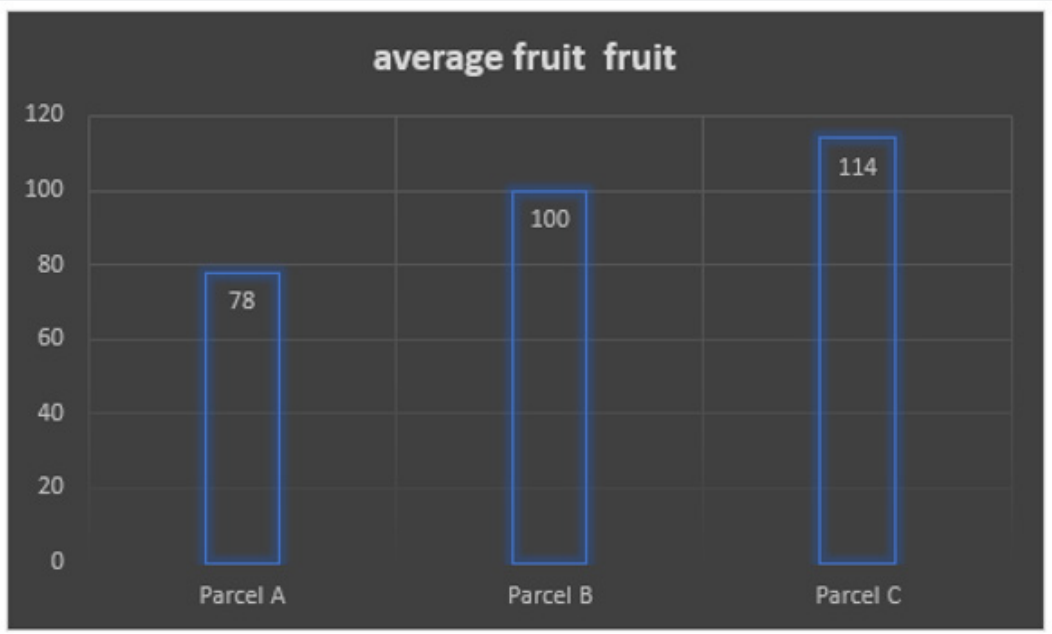

Figure 5: average weight of eggplants harvested at maturity.

Discussion

Plots treated with aqueous extract and essential oil of *C. aurantifolia* were very lightly infested by eggplant insects at all periods. Its broad spectrum of action on parasites on sites shows that it is not selective [23]. However, the essential oil has been shown to be very effective against insects on different sites during all periods. In addition, during the first period, the essential oil was very repellent against aphids and jassids (0%) [24]. These results are similar to those of Gnago et al. [25], who showed that neem seed extract behaves as a repellent. The leaves of C. aurantifolia exhibited high pesticidal activity by effectively reducing the populations of whiteflies, beetles, caterpillars, orthoptera, aphids and jassids as well as reducing the incidence and severity of pest pests. leaves, buds and fruits. This plant would therefore have repellent and anti-appetizing properties (Nevala, 2000). The leaves would therefore contain compounds that deter feeding, repellent, inhibit oviposition and growth regulatory activities against a wide variety of crop pests [26]. The aqueous extract and essential oil of lemon leaves were able to reduce the presence and activities of insects on the plots and therefore allowed a good yield, because there is a positive correlation between the incidence and severity of crop damage and the pest population at the sites [27]. This suggests that the aqueous extract and essential oil of lemon leaves can be used as a good pesticide to protect vegetable crops. Compounds from the leaves of C. Aurantifolia have regulatory effects (antibioses/ anticenoses) on insects [28]. Similarly, Amtul [29] reported that A. indica contains compounds that act as inhibitors of the digestive enzyme alpha-amylase in beetles. Indeed, numerous studies have shown the effects of pesticidal plants on vegetable crop aggressors. Okereke et al. [2] showed that the application of aqueous extracts of Azadirachta indica on tomato plants infected by Sclerotium rolfsii Saccardo helps reduce the severity of the disease and obtain better growth of the plants. Kankam and Sowley [30] demonstrated that amending chili plants infested with A. indica induces a significant drop in nematode populations resulting in better growth of treated plants. Echereobia et al. (2010) showed that aqueous extracts of Piper guineense induced repellent activity on arthropod pests. Kambou and Guissou [31] also noted that spicy substances (Sinapis nigra, Mochiah et al. [19] showed that Allium and *C. papaya* extracts led to a considerable reduction in okra and eggplant pests. Bolou et al. [32] demonstrated that the essential oil of *X. aethiopica* fruits strongly inhibited the mycelial growth of *S. rolfsii*, resulting in a

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considerable reduction in the incidence of the disease on treated tomato plants.

The high N'drowa yields obtained in plots B and C can be explained using different factors. First, the good vegetative development linked to the low sensitivity of these plants to bacterial wilt allowed many plants to return to production. Then, the low presence of pests on the treated sites and the very reduced number of attacks on buds, leaves and fruits. However, the yield of plot A would be linked on the one hand to the ravages of plants, buds and leaves by pests. Also, it would be linked to rotting or perforation of fruits by insect pests (caterpillars, aphids) [33]. Asare-Bediako et al. (2014) showed that extracts of A. indica, C. papaya , *Allium sp* . , *Capsicum sp* . minimized the severity of whitefly damage and increased the yield of treated plots. Adesina et al., [34] showed that the use of aqueous extracts of Loncarpous cyanescens and Dalbergia lactea reduced the ravages of leaf beetles in okra cultivation. Habou et al. [35] showed that the application of *Jatropha sp.* on cowpea pests made it possible to reduce the abundance of thrips and bugs and the severity of their attacks, thus increasing yield.

Furthermore, the use of biopesticides is gaining ground as these plants provide alternative means of controlling crop pests, also preserve the ecosystem [36]. Natural products from plants can also increase yields with a cost/benefit ratio comparable to that of synthetic pesticides [37]. Thus, the improvement in the yield of treated plants could be due to the release of nutrients (nitrogen, phosphorus) by the leaves for significant vegetative growth and better fruit development. Also, these properties would be due to the presence of secondary metabolites such as saponins, polyesters and phenols which would influence the metabolism of the plant and allow an improvement in plant growth and yield [38]. These results are similar to those obtained by Habou et al. [39,40,41,42] who in open fields, curcas oil significantly reduced the number of cowpea pests.

Conclusion

Plant pesticides are far from quickly replacing synthetic pesticides, particularly in large crops. In market gardening, however, they can be an alternative solution and contribute to preserving the health of populations. Pesticidal plant extracts are less dangerous than synthetic pesticides due to their fairly rapid decomposition and low polluting action. They also make it possible to keep the pest population below the harmful threshold and reduce the use of synthetic pesticides used on market gardening. The aqueous extract and the essential oil of the leaves of C. aurantifolia have proven to be powerful pesticides by the considerable reduction in the presence of pests and damage on treated crop plots but also in the good yield of eggplants observed in this study. It would therefore be necessary to explore the capabilities of biopesticides (notably lemon leaves) to control pests of vegetable crops and to optimize their use. We must encourage the use of plant pesticides by producers and raise awareness about the enhanced safety of products treated with plant-based pesticides and their long-term benefits.

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