

## Effect of bio pesticides in the management of populations of the mango mealybug *Rastrococcus invadens* williams 1986 in northern Ivory coast

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### Abstract

The mango mealybug, *Rastrococcus invadens* Williams 1986, is one of the main constraints on mango production in northern Ivory Coast. The aim of this study was to control the mealybug population using biopesticides. Three mango orchards were selected in the localities of Lenvogo, Lataha, and Katia. A Fisher block design with three replicates and four objects was used. Each orchard was divided into four elementary plots consisting of one dose of each biopesticide (limocide,  $2.98 \times 10^{-4}$  g/ml; carapa,  $4.98 \times 10^{-6}$  g/ml; and neem,  $1.49 \times 10^{-6}$  g/ml) and a reference product, cypercal, at a dose of  $2.49 \times 10^{-4}$  g/ml. The mortality rate varied from one treatment to another. Limocide was the most effective against mealybugs, with a mortality rate of  $71.76\% \pm 2.78$ . Limocide also significantly reduced the attack rate in orchards, from 51% to 6.6%, as well as the level of infestation, from more than 15 mealybugs per leaf to around 3. Neem and carapa resulted in moderate reductions of 22.33% and 28.66%, respectively. These biological control methods could be an alternative to chemical insecticides.

**Keywords:** Mango tree; *Rastrococcus invadens*; Biological control; Ivory Coast

### 1. Introduction

In Ivory Coast, all mango (*Mangifera indica*) production areas are home to the mango mealybug (*Rastrococcus invadens* Williams, 1986), a pest that threatens mango productivity. It can cause production losses of up to 100% in smallholder farming if no treatment is applied [5]. In order to protect orchards from this insect pest and reduce production losses, producers first implemented mechanical control methods, which proved to be ineffective, before turning to conventional chemical control based on the use of pesticides. However, this chemical control method has limited effectiveness and has negative impacts on the environment and its components [17]. The intensive use of synthetic insecticides can lead to changes in the organoleptic properties of plant products, as well as potential toxic effects on human health. Insecticides can reduce the mobility, or even directly kill, antagonists (competitors, predators, and parasites) that naturally regulate target pest populations. By eliminating prey or hosts, insecticides deprive antagonists of food, causing them to eventually die. Beneficial insects, including pollinating bees, are highly vulnerable to this, which disrupts the structure and productivity of ecosystems, thereby reducing biodiversity [7]. Faced with this situation, an alternative control method using biopesticides that respects the environment and biodiversity is therefore necessary for the sustainable management of this insect pest.

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## 2. Materials and methods

### 2.1. Study site

The department of Korhogo is located in northern Côte d'Ivoire, between 8°26' and 10°27' north latitude and 5°17' and 6°19' west longitude. It covers an area of 12,500 km<sup>2</sup> (Figure 1), or 3.9% of the national territory [8]. The climate is tropical Sudano-Guinean, marked by two main seasons, one rainy and the other dry, with average temperatures between 24.42°C and 30.53°C [15]. Three orchards were selected in the same agro-ecological zone in the northeast, in the Poro region (Lenvogo, Lataha, and Katia). The distance between the orchards was more than 20 km.

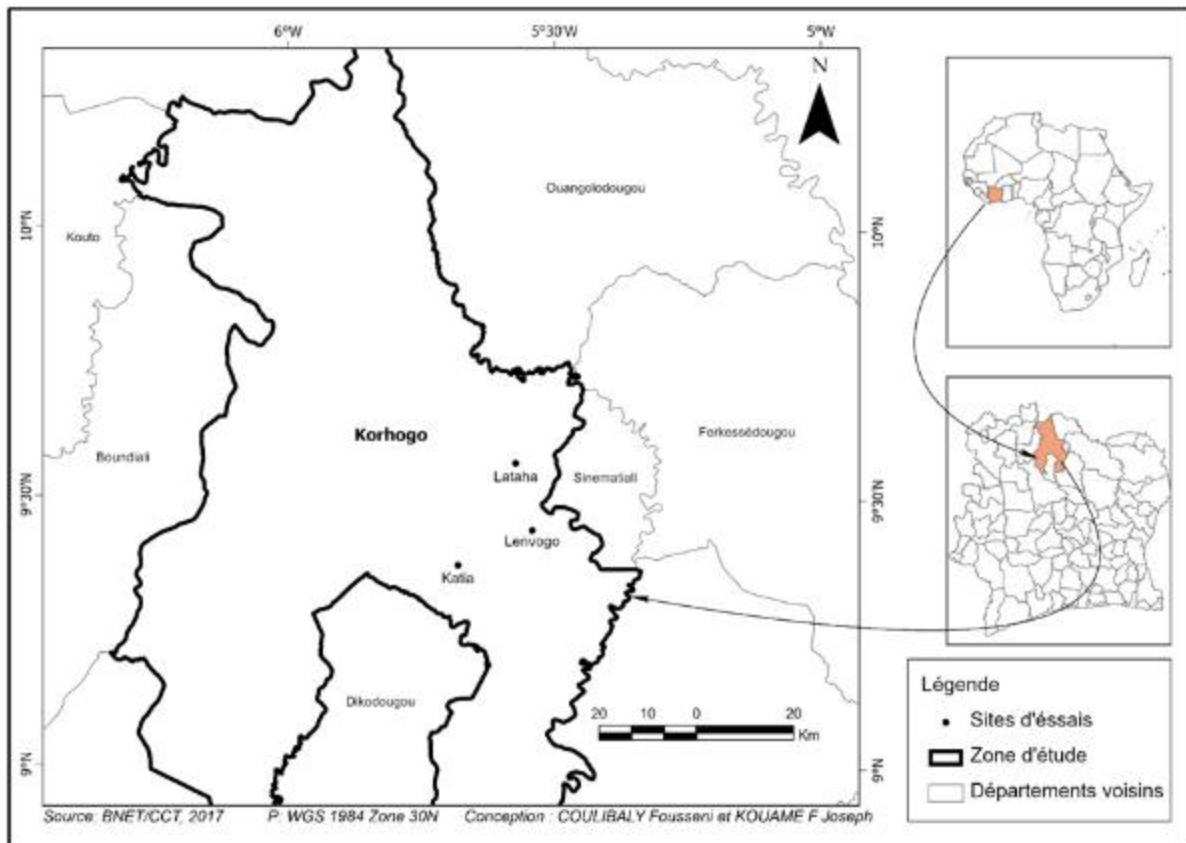


Figure 1 Map of study sites

### 2.2. Materials

#### 2.2.1. Biological materials

Mango orchards of the Kent, Keitt, Palmer, Brooks, Lipens, and Amelie varieties were used as plant material. The animal material used consisted of mealybugs at all stages of development, as well as their natural enemies.

#### 2.2.2. Technical materials

The handling of treatment products required the use of personal protective equipment consisting of coveralls, disposable gloves, face masks, and boots.

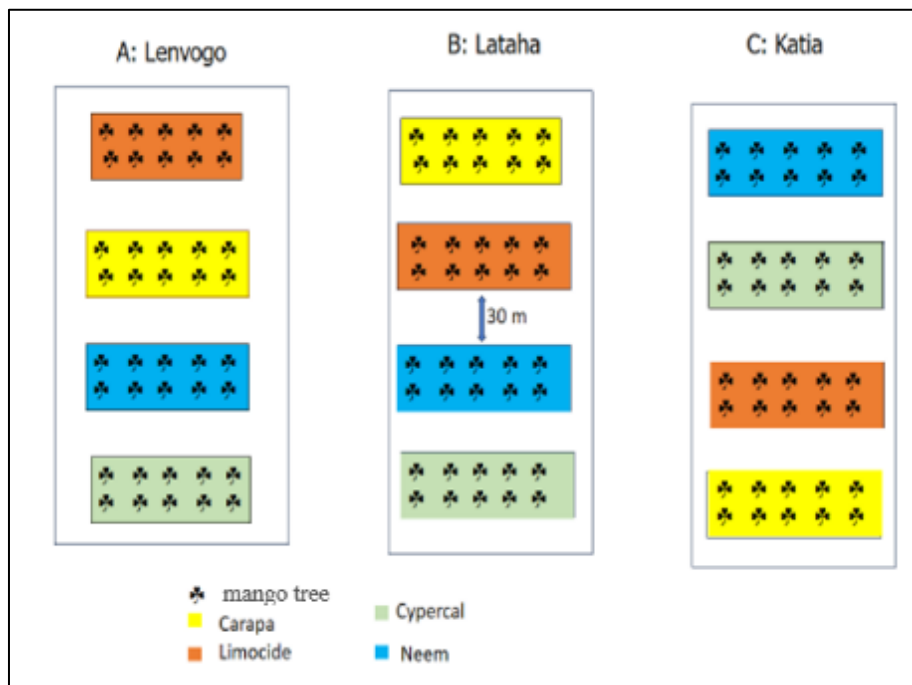
A dissection kit and data collection sheets were used, and a GPS was used to record the geographical coordinates of the study sites. A thermo-hygrometer was used to measure temperature and relative humidity, and a rain gauge was used to record rainfall at the study site. A Bresser handheld magnifying glass (10 x 30 magnification) was used to observe and count scale insects, larvae, and eggs on the underside of the leaflets. Binocular magnifying glasses were used to identify insects, and a digital camera was used to take pictures.

### 2.2.3. Products tested

The biopesticides used are citrus essential oil (limocide) with D-limonene as the active ingredient; neem essential oil (azadirachtin) and carapa essential oil (sesquiterpenes, germacrene D (13.2–21%) and  $\alpha$ -humulene (13.4–31.7%)).

### 2.3. Methods

The device was a Fisher block with four objects and three repetitions. Each orchard was subdivided into four elementary plots consisting of 25 mango trees (1/4 hectare). The distance between trees was 10 meters and 30 meters between elementary plots (Figure 2). A zero rating was given to the elementary plots before product application. On each mango tree, a square of 20 infested leaves was selected and the scale insects on these leaves at all stages were counted. Three applications of the products were made at ten-day intervals. After the third application, another count of live scale insects was made to assess the number of scale insects killed by the treatments.



**Figure 2** Experimental setup for testing products in a real-world environment

#### 2.3.1. Application of products

Each dose of product (150 ml for limocide, neem, and carapa, then 75 ml for cypercal) was diluted in 15 liters of water and poured into a sprayer for application to the trees (Table 1). One milliliter of acetone was added to each sprayer to facilitate emulsification of the essential oils. A separate sprayer was used for each product to avoid interference. The sprayers were calibrated before each application. Three applications were carried out at 10-day intervals.

72 hours after application of the products, a count of live scale insects was carried out to assess the mortality rate for each stage of mealybug development.

**Table 1** Concentrations of the different products to be tested

Products	Product quantity (ml)	Dilution in water (l)	Concentrations (g/ml)
Limocide 60 ME	150	15	$5,94.10^{-4}$ g/ml
Neem	150	15	$2,97.10^{-6}$ g/ml
Crarapa	150	15	$9,90.10^{-6}$ g/ml
Cypercal	75	15	$1,86. 10^{-4}$ g/ml

### 2.3.2. Parameters studied

#### Attack rate in orchards

The attack rate corresponds to the proportion of infested trees in relation to the total number of trees on the plot (formula a). The severity of damage was assessed using the scale proposed by Minhobo *et al.* in 2015. The different gradations of the scale are as follows:

- High rate when more than 30 trees per hectare are infested;
- Medium rate when the number of infested trees per hectare is between 10 and 30;
- Low rate when fewer than 10 trees per hectare are infested.

$$\text{a) Attack rate} = \frac{\text{Number of infested trees}}{\text{Total number of trees}} \times 100$$

#### Level of infestation of plants

The level of infestation was assessed by counting the number of scale insects at all stages relative to the number of leaves examined (formula b). Infestation thresholds were established according to the scale developed by Minhobo *et al.* [9].

- Low infestation level: when there are fewer than 5 scale insects per leaf;
- Medium infestation level: when the number of scale insects per leaf is between 5 and 10;
- High infestation level: when there are more than 10 scale insects per leaf.

$$\text{b) Level of infestation} = \frac{\text{Total number of scale insects}}{\text{Total number of leaves observed}} \times 100$$

#### Mortality rate

The mortality rate was calculated for each treatment and for each stage of mealybug development. It is used to show whether the treatments applied reduced the number of mealybugs on the plants [11]. The formula c below was used for this calculation.

$$\text{c) Mortality rate} = \frac{\text{Number of dead scale insects}}{\text{Total number of scale insects}} \times 100$$

### 3.3.3. Statistical analyses

The comparison of the means for the number of trees attacked and the number of scale insects on the leaves was performed using the one-way ANOVA parametric test at a 5% significance level when the distribution followed a normal distribution and the homogeneity of variances was established.

Pairwise comparisons between the means were performed using Fisher's test, applied at a significance level of 5%, when significant differences were detected.

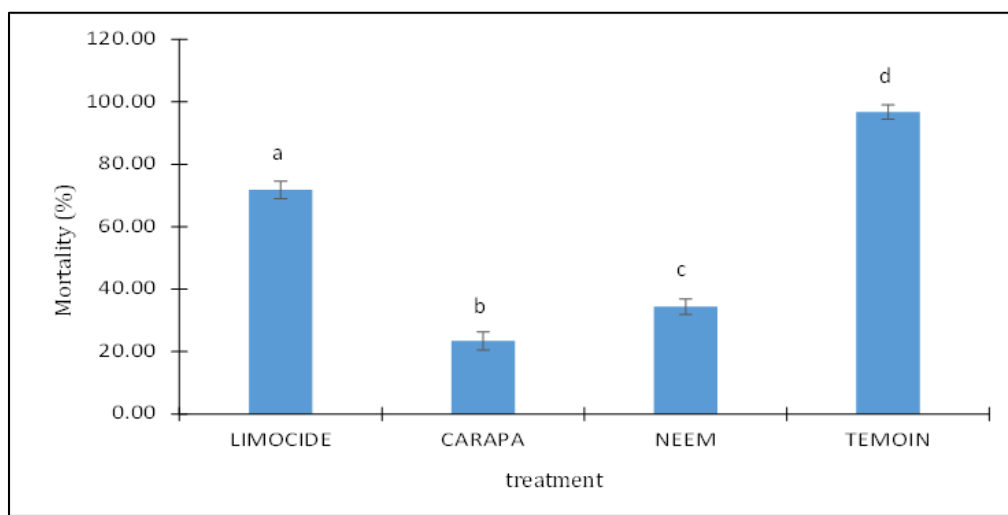
Data processing was performed using Statistica 7.1 and XLSTAT software.

### 3. Results

#### 3.1. Effect of biopesticides on the mangrove mealybug

##### Mortality rate

The mortality rate varied from one treatment to another. The mortality rate of mangrove mealybugs after 72 hours was  $96.72 \pm 2.26\%$  for the reference control (cypical),  $71.76 \pm 2.78\%$  for limocide,  $34.33 \pm 2.5\%$  for neem, and  $23.39 \pm 2.90\%$  for carapa. Limocide had a higher mortality rate than the other biopesticides (Figure 7). The ANOVA variance analysis shows that there is a highly significant difference between the three pesticides ( $P = 0.001$ ).



**Figure 3** Histogram showing the effectiveness of three biopesticides against mealybugs on mango trees

#### 3.2. Attack rate in orchards

The orchards in Lataha, Lenvogo, and Katia used for testing had attacked rates of 70%, 45%, and 38%, respectively. Twenty-one days after treatment, limocide reduced the attack rate in orchards to 6.6%. However, after treatment for the same period, carapa and neem achieved attack rates of 28.66% and 22.33% respectively (Table 2).

**Table 2** Attack rates in orchards after treatment with different products

Treatments	Attack rate after treatment
Limocide	Low = 6,6%
Carapa	Medium = 28,66%
Neem	Medium = 22,33%
Cypical	Low = 3%

#### 3.3. Level of infestation in orchards

The level of infestation in the orchards in Lataha, Lenvogo, and Katia, which were used for testing, varied, with an average infestation of 15.33 scale insects per leaf. Twenty-one days after treatment, the limocide significantly reduced the level of infestation in orchards from 15.33 scale insects per leaf to 3 scale insects per leaf. In contrast, Carapa and Neem resulted in a moderate reduction in the level of infestation in the orchards, with 7.66 scale insects per leaf and 5.33 scale insects per leaf, respectively (Table 3).

**Table 3** Level of infestation by location

Level of infestation in orchards before treatment	Treatment	Level of infestation in orchards after treatment
High (15,33)	Limocide	Low = 3
	Carapa	Medium = 7,66
	Neem	Medium = 5,33
	Cypercal	Low = 1,33

#### 4. Discussion

In the treated plots, limocide, neem, and carapa all caused the death of mango mealybugs at varying rates. This observed mortality is thought to be linked to the insecticidal properties of the active substances contained in the insecticidal plant extracts. According to Philogène *et al.* [12], neem (*Azadirachta indica*) has been used in the tropics for over 4,000 years to protect stored cereals. Diabaté *et al.* [3] reported that the insecticidal effect of *Jatropha curcas* and *Azadirachta indica* extracts on *Plutella xylostella* larvae was due to the active substances present in the extracts. According to Tounou *et al.* [16] and Harouna *et al.* [6], plant extracts with insecticidal effects (such as neem) caused toxicity effects similar to those of synthetic chemicals. Zongo *et al.* [18] and Addea [1] also indicated that there are more than one hundred (100) compounds in neem extracts that were responsible for insect mortality.

On plants that had been treated with neem, scale insects left the various organs and were found on parts that had not been treated. This highlights the repellent effect of neem oil. According to Regnault-Roger *et al.* [14], insect repellency is defined as the ability of an insect repellent substance to reduce the normal contact time of the insect with the treated surface. This repellency of scale insects could be due to a substance contained in neem, azadirachtin [4], which stimulates specific deterrent cells in chemoreceptors. This substance also blocks the triggering of sugar receptor cells that normally promote feeding. This causes the food to be rejected [2]. This assertion has been confirmed by the work of Mondédji *et al.* [10], who reported the anti-appetitive properties of azadirachtin against *Plutella xylostella* (Lepidoptera: Plutellidae) and *Salhbergella singularis* (Heteroptera: Miridae). Cypercal, the reference control, induced a mortality rate of over 95% at all stages of development of the mango mealybug. This very high rate could be explained by the fact that this chemical insecticide, whose active ingredient is cypermethrin, has a long residual effect and acts by contact and ingestion. These results are similar to those of RECA [13], who demonstrated its lethal effect on adult males and females of *Omnivora mutabilis*. It is non-systemic and blocks the insect's nervous system when it comes into contact with or ingests it. In this case, the insect stops feeding, remains immobile and then dies quickly [13]. Mortality was observed from the first day after treatment because these piercing-sucking insects absorb the substance when trying to feed.

#### 5. Conclusion

The results of this study showed that biopesticides (limocide, neem, and carapa) caused mortality in scale insects at varying rates 72 hours after treatment. Limocide proved to be the most effective biopesticide, with an average mortality rate of  $71.76 \pm 2.78\%$  in the field. However, its effectiveness decreases when the scale insect is covered with wax in the adult stage. Limocide reduced the attack rate in orchards from 51% to 6.6%. In contrast, carapa and neem achieved post-treatment infestation rates of 28.66% and 22.33%, respectively.

These biopesticides also reduced the level of infestation in orchards from 15.33 scale insects per leaf to 3 scale insects per leaf for limocide, carapa, and neem, respectively. This work has highlighted a biological control method involving the use of biopesticides in the management of mealybug populations on mango trees in northern Ivory Coast.

#### Compliance with ethical standards

##### Disclosure of conflict of interest

No conflict of interest to be disclosed.

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