



Antifungal Activity of Biopesticides and their Effects on the Growth Parameters and Yield of Maize and Pigeon Pea

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ABSTRACT

In recent years, the search for biological methods to avoid the application of chemical products in agriculture has led to investigating the use of plant extracts and biopolymers-based materials, such as chitosan. The present work was aimed at evaluating the antifungal effect of chitosan and the aqueous extracts of the three aloe species, namely: *Aloe vera*, *A. ferox*, and *A. marlothii* against *Cochliobolus heterostrophus* on maize and *Colletotrichum lindemuthianum* on pigeon pea, as well as their effect on the growth parameters, severity degree and grain yields. The experiment was laid out in a Randomized Complete Block Design with three replications and 9 treatments. The spraying of all tested biopesticides at 7th and 14th days showed a significant increase in growth parameter (increase of shoots) on both species. The highest grain yield was observed by applying samples QTZ at 25% (6.0 t.ha⁻¹) and AF_13 (4.2 t.ha⁻¹) on maize, AF_13 (1.2 t.ha⁻¹) and AF_10 (1.6 t.ha⁻¹) on pigeon pea when compared with the positive control - Manconzeb (4.5 and 0.5 t.ha⁻¹ for maize and pigeon pea, respectively).

Keywords: Chitosan, Aloe species, Biopesticides, Crop protection, Growth parameters, Grain yield.

Introduction

Synthetic pesticides are agents used to control pests in agriculture such as fungi, bacteria, insects, etc. These pesticides have a quick mode of action and a broad spectrum of activity. They are a very important part of agriculture. Their long-term use may have drawbacks in the environment by harming all forms of life, including the targeted pest as well as the non-targeted plant species. They may have different fates in the various components of the environment such as soil, air, water, and biota, and therefore impact public health. In the search for alternative solutions to maintain human and plant health, as well as environmental sustainability, the interest in plants and their chemo-biodiversity as a source of bioactive secondary metabolites has increased.^{1,2}

Botanical insecticides are a viable alternative to synthetic insecticides, and they degrade quickly, retard the development of insecticide resistance, and can be used shortly before harvest and are environment-friendly.³

Many researches have focused on chitosan as a source of bioactive material in recent years. However, the high molecular weight results in its poor solubility at neutral pH, and its high viscosity limits its use in food, cosmetics, agriculture, and the pharmaceutical industry. Chitosan is natural, non-toxic, and a polymer prepared from chitin by deacetylation, which in turn is a major component of the shells of crustaceans. It has been reported to have antitumor, antibacterial, antifungal, and antimicrobial properties.⁴

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Previous studies have shown that chitosan has fungicidal effects against *Penicillium* on peanuts and other plant species,^{5,6} enhance the growth of Black gram plant (*Vigna mungo*).⁷

Plants belonging to the genus *Aloe* have been used in many countries for the folklore treatment of various illnesses including inflamed eyes and wound healing by topical application of the fresh gel from the plant, as purgative due to its anthraquinone glycosides (barbaloin, β -barbaloin, and homonatinoloin), treatment of chronic skin ulcers, and hypoglycemic activity. Resin is the most variable component in the range of 40 to 80%. The resin does not have a medicinal interest; it corresponds to an ester of *p*-coumaric acid and the resin alcohol, aloeresinetanol. Besides, there are up to 20% of aloins. On hydrolysis, aloins yield emodin that is the active compound of the acibar.⁸⁻¹⁰ Although there are 300 *Aloe* species, only a few are important in medicine. The gel of the aloe species is known to have antifungal activity against plant pathogenic fungi.¹¹

Maize (*Zea mays*) and pigeon pea (*Cajanus cajan*) are important crops for the supply of human nutritional requirements. These two types of basic food nutritionally complement each other. A dietary combination of these plants provides higher quality and about 50% of the protein in a diet compared to individual plants. Food Agriculture Organization designated half of the Sub-Saharan African countries as having a short supply of these grains or seeds, due in part to the low yield caused by pests, including the effects of two pathogenic fungi, *Cochliobolus heterostrophus* and *Colletotrichum lindemuthianum* on maize and pigeon pea, respectively. This has caused notable food shortages. In Sub-Saharan Africa, one of the constraints to sustained production of these crops is the lack of synthetic pesticides due to their high cost. This leads to poor plant growth which results in a decline in agricultural food production.¹²⁻¹⁵

This research work aimed to evaluate *in situ* the fungicidal effects of the aqueous extracts of chitosan, *A. vera*, *A. marlothii*, and *A. ferox* against *Cochliobolus heterostrophus* (on maize) and *Colletotrichum lindemuthianum* (on pigeon pea) and their crop yield enhancement. Before now, no antifungal properties have been reported on *Aloe* species against *Cochliobolus spp* on maize and *Colletotrichum spp* on pigeon pea.

Materials and Methods

This research was conducted *in situ* at Rural Development High School (ESUDER) in Vilankulo District, Inhambane Province in Mozambique from May to July 2018. The samples of the leaves of *A. vera* (AV), *A. marlothii* (AM), and *A. ferox* (AF) were collected in May 2018 in Namaacha District in Maputo Province. The samples were prepared as aqueous solutions or without adding water, as follows: *Aloe vera* (AV), *A. ferox* gel (AF_gel), *A. marlothii* 10% (AM_10), *A. marlothii* 13% (AM_13), *A. ferox* 10% (AF_10), and *A. ferox* 13% (AF_13). Chitosan (Qtz) at 25% was obtained previously in our laboratory using crab shells. Three spraying dates were used: S1 (spraying on the 7th day after the emergence of the crops), S2 (spraying on the 14th day after the emergence of the crops), and S3 (spraying on the 21st day after the emergence of the crops). The two parallel trials were laid in a Randomized Complete Block Design with three replications and 9 treatments corresponding to the aqueous or without adding water solutions. Data were collected one week after each spraying to determine the severity degree against *Cochliobolus heterostrophus* in maize and the antracnose causing *Colletotrichum lindemuthianum* in Pigeon pea. The severity of the disease was determined by using diagrammatic scales.¹⁶ Simultaneously, the assessment of the following growth parameters such as plant height, stem diameter, and the number of leaves per plant was measured.

Statistical analysis

The statistical analysis consisted of ANOVA and post-hoc tests implemented in SPSS Statistics (v.20, IBM SPSS Chicago).

Results and Discussion

Maize

The biopesticides effects on the growth parameters of maize were not significantly different according to the Kruskal-Wallis test ($P > 0.05$) after S1. These similarities in the growth parameters could be attributed to the fact that the corn seedlings are still in the establishment phase and at the beginning of their growing process. However, after S2, only the plant diameter was significantly affected ($P = 0.01$). The severity of blight leaf disease on maize increased after S1 on application of all extracts, except those treated with Manconzeb (Figure 1). After S2, a decrease in severity degree of blight disease (caused by *Cochliobolus heterostrophus*) on maize treated with all the extracts including Manconzeb (Figure 2) was observed. According to the Kruskal-Wallis test, there was a significant difference ($P < 0.05$) in the severity distribution of the blight leaf disease after S1 and S2. An increase in the severity of the blight disease on maize (Figures 1 and 2) was observed up to S2, while there was a decrease in disease severity between S2 and S3. These results showed that the tested biopesticides and the Manconzeb had a positive effect on reducing the blight leaf disease on maize.

The yield component of maize were significantly influenced ($P < 0.05$) by the interactive effect of the different samples. The multiple mean comparison test revealed that the highest mean yield was obtained by the treatment with chitosan and AM_13. The yields, in both cases, were found to be 6.0 and 4.2 t.ha⁻¹ (Figure 3) and were significantly different when compared with Manconzeb (4.5 t.h⁻¹). The high yield obtained by treating maize crops with chitosan reinforces the effect of this biopesticide in reducing infections which were observed after S3. This suggested that the observed effect could be responsible for the protection of the plant against blight leaf disease and the corresponding high grain yield. Pena and co-workers¹⁷ reported good agronomical performance and grain yield of maize when the seeds were incubated with chitosan before seeding. The AM_10 and AM_13 formulations also showed similar performance to chitosan with yields above 4.0 t.ha⁻¹.

Due to their wide range of biological activities, including antifungal properties, Aloe extracts have been evaluated for their potential use as biopesticides. The methanol extracts of *A. vera* showed repellent and toxicity effects against *Sitophilus oryzae*, a maize storage fungus pest,

and aloin A, a major secondary metabolite found in Aloe species was the most active compound.¹⁸ Also, the *A. vera* gel was found to enhance the shelf-life of tomato.¹⁹

Pigeon pea

The yield components of pigeon pea were not significantly affected by the different treatments (Table 1), suggesting that they did not interfere with the development of this crop. However, there is a possibility that the treatments had interfered with a number of growth parameters. This possibility could be described by significant differences in growth parameters as highlighted in Table 1. The sample AF_10 influenced positively the highest number of these parameters, which produced the highest grain yield observed (1.6 t.ha⁻¹) because the grain yield depends on the number of growth parameters. A decrease in severity degree of *Colletotrichum lindemuthianum* on pigeon pea was observed in all the biopesticides tested suggesting that the application of the treatments at intervals of seven days decreased the infestation of pigeon pea by the targeted fungus. According to ANOVA, there were no significant differences ($P > 0.05$) in all biopesticides after S1. However, after S2, significant differences between all biopesticides tested were observed. The Dunnett C test revealed significant differences ($P < 0.05$) of both AM_13 and chitosan when compared to Manconzeb. While the other biopesticides extracts did not show statistically significant different.

Anthracnose, a postharvest disease caused by the fungus *Colletotrichum lindemuthianum* is the most devastating disease of bell pepper that causes great economic losses especially in tropical climates.²¹ About 60% control of anthracnose on papaya fruit achieved with chitosan was found to be considered as a natural product to control postharvest diseases of papaya²² and bell pepper.²¹ The application of chitosan to the leaves of cucumber helps to control anthracnose disease caused by *Colletotrichum spp.*²³ The antifungal properties of *A. vera* extracts against various fungi species, including *Colletotrichum* pathogens, showed a broader spectrum of antifungal activity. Aloe species extracts may be an attractive alternative for use as a natural product for the control of fungi that attack industrial crops, thereby limiting the application of chemical fungicides.²⁴

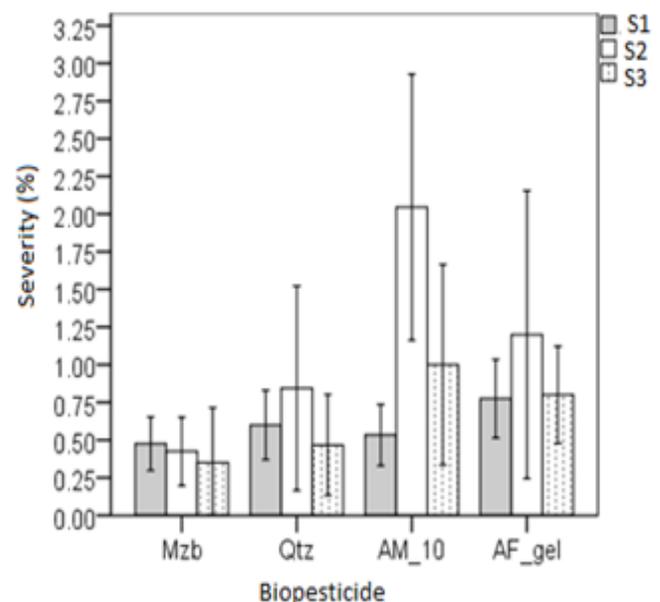


Figure 1: Severity degree of blight disease on maize after the third week

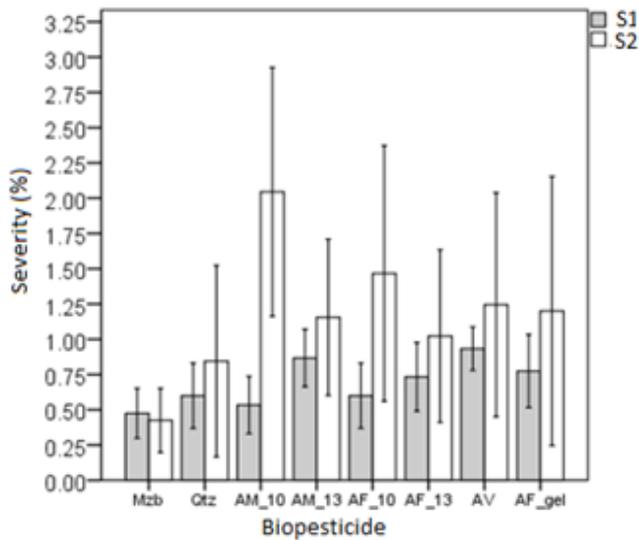


Figure 2: Severity degree of blight disease on maize after two weeks.

Table 1: Yield and number of pods per pigeon pea plant

| Treatment | Yield (t.ha ⁻¹) | Pods per plant |
|-----------|-----------------------------|----------------|
| test | 0.59 | 19 bc† |
| AF_gel | 0.32 | 11c |
| AV | 0.86 | 32abc |
| AF_13 | 1.20 | 27abc |
| AF_10 | 1.59 | 49a |
| AM_13 | 0.74 | 40ab |
| AM_10 | 0.83 | 17bc |
| Qtz | 0.70 | 13cb |
| Mzb | 0.52 | 22abc |

† Means followed by the same letter are not significantly different, Tukey HSD ($\alpha = 0.05$).

Aloe vera (AV), *A. ferox* gel (AF_gel), *A. marlothii* 10% (AM_10), *A. marlothii* 13% (AM_13), *A. ferox* 10% (AF_10), *A. ferox* 13% (AF_13), Chitosan 25% (Qtz), and Mancozeb (Mzb).

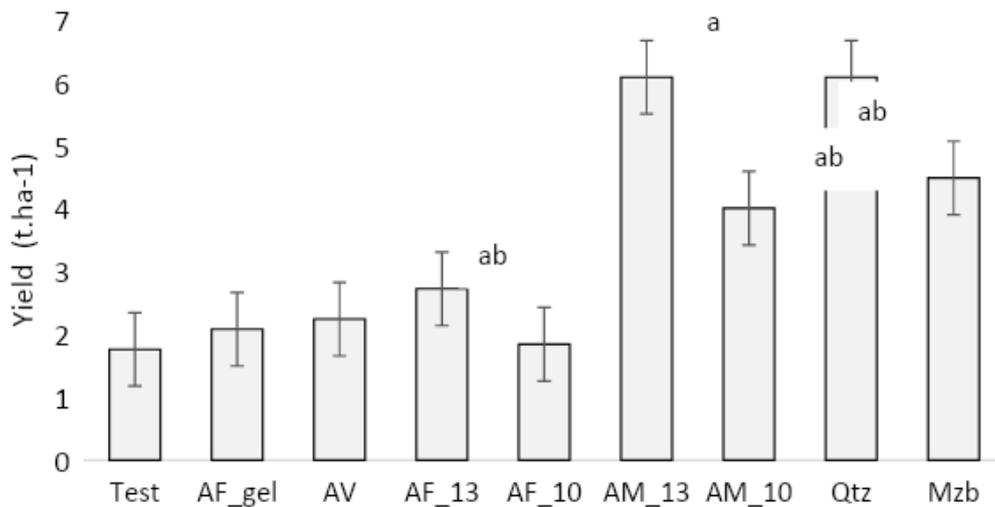


Figure 3: Average yield of the dry corn cob. The vertical lines represent an average confidence interval of 95%. Bars followed by the same letter are not statistically different by Tukey HSD test ($\alpha = 0.05$).

Conclusion

The results obtained in this work indicate the antifungal properties of chitosan and Aloe spp. extracts against the fungi tested in maize and pigeon pea crops. Aloe species and chitosan are available in large quantities and at a low price, and they could be promising in the development of sustainable agricultural practices as well as in food production and preservation (as highlighted by other authors). In particular, given the always-growing demand for food worldwide, the ongoing climate change, and the consumption of farmlands, biopesticides appear to be a promising tool for cultivation under stress conditions and to permit the cultivation of varieties of crops.

Conflict of interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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