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Research Article

Efficacy of Essential Oil Formulations in Rice Seed Treatment Against *Magnaporthe oryzae* B.C Couch, a Rice Blast Pathogen

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Abstract

Rice is one of the most widely produced cereals in Burkina Faso. However, the fungi transmitted by rice seeds, *Oryza sativa* L, cause enormous production losses in rice fields. The aim of this study was to assess the efficacy of powdery formulations of essential oils against *Magnaporthe oryzae* in rice. Thus, rice seeds of the FKR64 variety were disinfected with 15% sodium hypochlorite for 10 minutes and soaked in the suspension of 10^4 conidia/ml for 24 hours. These contaminated rice seeds were coated into powdery formulations of essential oils of *L. multiflora*, *C. schoenanthus* and their combination at a dose of 10g/kg of seed. A contaminated control was used. The experimental set-up was a completely randomised block with four treatments in three replicates, one of which was an absolute control. These treatments were carried out on *L. multiflora* essential oil, *C. schoenanthus essential oil* and their combination at a proportion of 50%. The results showed that the essential oil of *L. multiflora* recorded the highest percentage emergence, average height and average number of tillers of the plants, with values of 97.33%, 47.67 cm and 7.89 tillers respectively. This same formulation recorded the lowest percentage of seedling melt and foliar incidence, at 1.33% and 11.67% respectively. These essential oil formulations can be recommended for seed treatment against *Magnaporthe oryzae*.

Keywords

Oryza sativa L., Rice Blast, *Magnaporthe oryzae*, Essential Oils, Burkina Faso

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1. Introduction

Rice production in Burkina Faso faces a number of constraints, including fungal diseases, which account for 70% of crop diseases. Rice blast is the most dangerous of these diseases [1]. It can cause seedling melting and even leaf spot [2]. Infection can reach the seeds before harvest via the panicle, or after harvest, during drying and storage. These affections are among the main causes of production quality deterioration and yield reduction [3]. The use of chemical seed treatment products remains the main means of combating this disease, but their use causes problems to human and animal health and to the environment [4]. An alternative control method that is respectful of health and the environment therefore needs to be considered. Research into the fungicidal potential of aromatic plants has enabled the development of essential oil formulations for field or post-harvest protection of rice seeds [5]. The substances in these plants also have fungicidal properties [6, 7]. The aim of this study is to assess the efficacy of essential oils of *Lippia multiflora* and *Cymbopogon schoenanthus* against *Magnaporthe oryzae*, in rice seed treatments.

2. Material and Methods

2.1. Biological Material

Rice seeds of the FKR64 rice variety were used. The strain of *M. oryzae*, pathogen of rice blast, was used.

2.2. Essential Oil Formulations

Three (03) Powder formulations were manufactured from the oils of *L. multiflora*, *C. schoenanthus* and their combination at the respective minimum inhibition doses of 0.6 µl/ml i.e. 30 µl/100g, 1.5 µl/ml i.e. 75 µl/100g and 0.3 µl/ml i.e. 15 µl/100g. Rice flour was used as a solid adjuvant in the manufacture of powder formulations of essential oils.

2.3. Preparation of the Inoculum

M.oryzae, isolated from infested leaves showing symptoms, was grown on PDA medium in the dark at 25 ℃. After 14 days of incubation, the conidium-laden surface was scraped with a sterile metal spatula. The mycelium was then suspended in distilled water and vortexed for one minute. The resulting suspension was filtered through muslin to separate the conidia from the mycelial fragments. After counting with a Malassez cell, the conidial suspensions were adjusted with sterile water to obtain final concentrations of^{104} conidia/ml.

2.4. Seed Treatment Procedure

Rice seeds were disinfected with 15% sodium hypochlorite for 10 minutes [8]. followed by a sanitary assessment to ensure the absence of pathogens. These seeds were soaked in the suspension of 10^4 conidia/ml for 24 hours. Control seeds were transferred to Erlenmeyer flasks containing distilled water for 24 hours. The inoculated seeds and the control seeds, dried in a laboratory hood for 24 hours, were coated in powdered formulations of essential oils and then sown in pots.

2.5. Preparing the Potting Soil

The potting mix consisted of a mixture of organic fertiliser and soil in proportions of one third (1/3) and two thirds (2/3) respectively. The potting soil is collected at the Farakoba station to a depth of 20 cm then sterilised in an autoclave at 121 ℃ for 45 minutes.

2.6. Experimental Set-up

The experimental set-up was a completely randomised block with four treatments in three replicates, one of which was an absolute control (T0). These were treatments with the essential oil of *L. multiflora* (T1), with *C. schoenanthus* (T2) and with their combination at 50% proportion (T3).

2.7. Parameters Measured

The percentage of seedling emergence was assessed on the 14th day after sowing, the percentage of seedling melt on the 28th day after sowing, the percentage of plants with leaf spots, the number of tillers and the height of the plants on the 42nd day after sowing.

2.8. Statistical Analysis

Analysis of variance (ANOVA) was performed using XLSTAT software. Means were compared using the Fisher test at the 5% probability level.

3. Results

Table 1 shows the percentage of seedling emergence, seedling melt, foliar incidence, plant height and average number of tillers per plant according to the different essential oil formulations. For each parameter assessed, the analysis of variance showed a very highly significant difference between treatments at the 5% probability threshold using Fisher's test. The results showed that treatment T1 recorded the highest percentage of seedling emergence, plant height and number of tillers, with 97.33%, 47.67 cm and 7.89 tillers/plant respectively. The results showed that treatment T0 recorded the highest values for seedling melt and leaf incidence with 7.57% and 36.45% respectively. Treatment T0 recorded the lowest values for percentage emergence, plant height and number of tillers with 86.96%, 27.33 cm and 3.67 tillers/plant respectively. Treatment T1

recorded the lowest percentages of seedling melt and foliar incidence with 1.33% and 11.67% respectively.

Table 1. *Parameters assessed.*

Treatments	Emergence percentage	Percentage of seedling melt	Foliar incidence
T0	86,96^{c}	7,57^{a}	36,45^{a}
T1	97,33^{a}	1,33^{d}	11,67^{c}
T2	92,21^{b}	3,33^{b}	24,33^{b}
T3	92,54^{b}	2,67^{c}	23,67^{b}
Prob	< 0,001	< 0,001	< 0,001
Signification	THS	THS	THS

Legend: Values in the same column with the same letter are not statistically different at the 5% probability level according to the Fisher test.

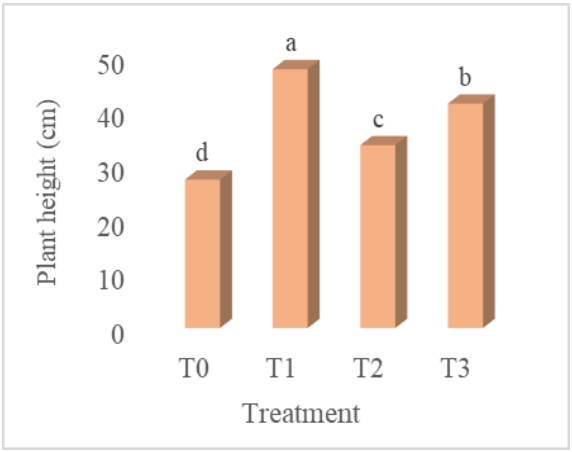

Figure 1. *Plant height.*

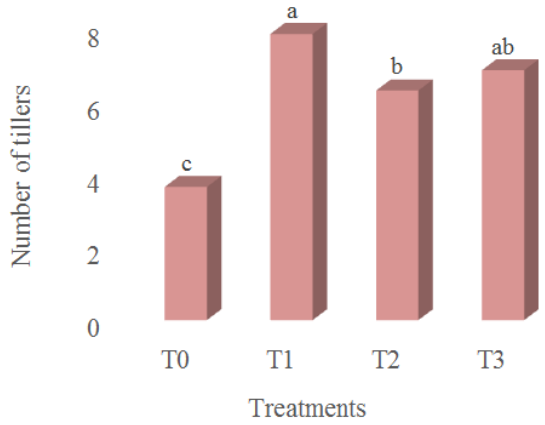

Figure 2. *Nomber of tillers.*

Graphs marked with the same letter are not statistically different at the 5% probability level according to the Fisher test.

4. Discussion

The application of powdery formulations of *L. multiflora* and *C. schoenanthus* essential oils and their combination to seeds artificially contaminated by *M. oryzae* demonstrated their antifungal effect. They reduced seed infection by improving the emergence rate and reducing the rate of seedling melt and foliar incidence compared to the control. The effectiveness of essential oils can be explained by their content of major elements, their antifungal properties and their chemical compositions, which enable them to stop or slow down the development of the fungus, or by the synergistic or additive effect of the chemical compounds. This effectiveness of essential oils has been reported by [9].

Similar results have been obtained by [2] who demonstrated that the fungicidal effect of three formulations based on essential oils of *L. multiflora*, *C. schoenanthus* and *O. americanum* in the protection of cowpea seeds. In addition, the work of [10] on fungi isolated from groundnuts in Benin showed the antifungal activity of essential oils and aqueous extracts of *O. americanum* on several toxigenic strains of the genus *Aspergillus* and *Fusarium*. The work of [11] demonstrated the efficacy of *C. schoenanthus* essential oils against sorghum and millet seed-borne fungi. The work of [12] carried out on the seeds of some cereals such as maize in the presence of phytopathogenic and toxinogenic fungi in Togo showed that the essential oils of *O. basilicum* and *C. schoenanthus* have antifungal activity by improving the germination rate of the seeds of these two crops.

The results showed that the essential oils and their combination had a remarkable effect on the morphological parameters of the rice plants by improving the plant height and number of tillers. However, the essential oil of *L. multiflora* had a more marked effect in improving the number of tillers and plant height. This can be explained by the fact that essential oil formulations, depending on their chemical composition, do not regulate *M. oryzae* in the same way. Similar work has been reported by [13]. The work carried out by [14, 15] respectively showed that the application of essential oils of *O. gratissimum* and *C. citratus* to cotton resulted in good fibre yields, while the application of essential oil of *X. Aethiopica* against *F. oxysporum* produced high leaf and root biomass compared with the control.

5. Conclusion

This study demonstrated the efficacy of essential oils as seed treatments against *M. oryzae*. All the essential oil formulations of *L. multiflora*, *C. schoenanthus* and their combination showed an antifungal effect and improved the mor-

phological parameters of the rice plants. However, the essential oil of *L. multiflora* had a more marked effect in antifungal activity and in improving morphological parameters. These essential oil formulations can be recommended for the treatment and conservation of seeds against fungi.

Abbreviations

FKR : Farakoba Rice
PDA: Potato Dextrose agar

Author Contributions

Ouattara Souleymane: Conceptualization, Resources, Data curation, Software, Formal Analysis, Funding acquisition, Methodology, Writing – original draft, Project administration, Writing – review & editing

Kassankogno Abalo Itolou: Conceptualization, Resources, Validation, Visualization, Writing - review & editing

Zougrana Sylvain: Conceptualization, Writing - original draft, Methodology

Séréme Abdoulaye: Conceptualization, Resources, Supervision, Validation, Investigation

Koïta Kadidia: Conceptualization, Resources, Supervision, Validation, Investigation

Conflicts of Interest

The authors declare no conflicts of interest.

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