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PYROLIGNEOUS EXTRACT OF *EUCALYPTUS* SP. IN THE TREATMENT CHICKPEA SEEDS

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Abstract: Seed treatment is one of the main measures for controlling fungal diseases. Pyroligneous extract is derived from burning wood, resulting in a liquid condensed from the smoke, which has biostimulant, chelating and phytosanitary properties, with potential in agriculture. The aim of this study was to evaluate different concentrations of pyroligneous extract from *Eucalyptus* sp. in the treatment of chickpea seeds. The research was conducted at the Phytopathology Research Laboratory of the Federal University of Minas Gerais, Montes Claros-MG. A completely randomized design (DIC) with four replications was used. The treatments consisted of different concentrations (0, 5, 25, 50 and 75% v/v) of pyroligneous extract from *Eucalyptus sp*. Aleppo chickpea seeds were immersed for 5 minutes in 100 mL of the different concentrations of pyroligneous extract. The filter paper method or "blotter test" was used for the seed health test. In each gerbox referring to 25 seeds were placed in each box. The seeds were incubated in a BOD chamber at 20 °C with a 12-hour photoperiod for eight days. After this period, the genera of fungi in the seeds were identified and the incidence of fungi in the treatments was determined. The fungi associated with the seeds were *Aspergillus* sp. (12%), *Penicillum* sp. (33%), *Rhizopus* sp. (3.6%) and *Fusarium* sp. (9.5%). The highest concentrations of *Eucalyptus* sp. pyroligneous extract showed greater fungal control, reaching 91% at a concentration of 75%. The pyroligneous extract of *Eucalyptus* sp. has potential for treating chickpea seeds.
Keywords: *Cicer arietinum*, health test, fungi, control.

INTRODUCTION

Chickpeas, a legume of the genus *Cicer*, are one of the most cultivated species within the "pulses" group, which is characterized by legumes whose dried seeds are widely used in human nutrition (FAO, 2016). As well as being an important source of vegetable protein, this food contains essential nutrients such as fiber, vitamins and minerals, contributing to a balanced and healthy diet.

In addition, its cultivation has agronomic advantages, such as the ability to fix nitrogen in the soil, which contributes to agricultural sustainability (Pereira et al., 2020). In this way, chickpeas not only play a key role in nutrition, but are also a promising option for more sustainable agricultural practices.

Currently, chickpeas rank third in terms of production volume in the pulses ranking, with an annual production of 10.1 million tons. It is second only to beans and peas, accounting for 70% of global pulse production and 17% of the total. In Brazil, chickpeas were introduced at the beginning of the 20th century, but their production area has only started to grow recently, representing an opportunity for both domestic consumption and export. Currently, the country still depends on imports to meet the annual demand of around 8,000 tons (Reyes, 2020).

The occurrence of diseases is one of the main causes of the reduction in chickpea production. These diseases, caused by fungi, bacteria, viruses and nematodes, can result in total losses, reduce the quality of the product or make some areas unsuitable for cultivation. Chickpeas are vulnerable to various diseases, such as ascochyta burn, sclerotium wilt, fusarium, root rot, viruses and nematode infestations, which represents a major challenge for production.

In conclusion, the control of pests and diseases that affect chickpeas requires an integrated approach that combines chemical and biological methods to ensure sustainable production. The use of fungicides, although effective, can be complemented with more sustainable alternatives, such as the use of eucalyptus pyroligneous extract. This extract, obtained through the distillation of eucalyptus wood, is known for its natural antifungal and insecticidal properties and is a promising alternative for controlling diseases and pests in an environmentally friendly way. Its application can reduce dependence on synthetic chemicals, promote soil health and increase plant resistance, contributing to more sustainable agricultural practices that are less aggressive to the environment. Adopting strategies such as these, combined with integrated pest management (IPM), represents an effective and responsible way forward for chickpea production.

The aim of this study is to evaluate the potential of eucalyptus pyroligneous extract as a sustainable alternative for disease control, with the aim of reducing the incidence of infected seeds. The research also aims to increase productivity by treating seeds with the extract, promoting the use of effectively treated seeds for cultivation, thus contributing to healthier and more sustainable production in the field.

METHODOLOGY

In the context of assessing seed quality, the health test is used to determine the phytosanitary state of a batch, along with other tests that assess germination capacity, vigor, physical purity and genetic identity.

There are various tests that can be carried out to test seed health. In this work, which was carried out with chickpeas, the Incubation on Paper Substrate test or Filter Paper method, better known as the "blotter test", was used.

The procedure to be followed is based on the Seed Health Analysis Manual, made available by the Ministry of Agriculture, Livestock and Supply (MAPA).

The experiment was carried out at the Phytopathology Research Laboratory at the Federal University of Minas Gerais (UFMG), Montes Claros-MG *campus*.

The treatments consisted of different concentrations (0, 5, 25, 50 and 75% v/v) of pyroligneous extract from *Eucalyptus sp*. Aleppo chickpea seeds were immersed for 5 minutes in 100 mL of the different concentrations of pyroligneous extract.

PREPARATION OF PYROLIGNEOUS EXTRACT

The pyroligneous extract was diluted in distilled water to obtain the desired concentrations. The dilution was carried out using precise volumetry, ensuring the accuracy of the concentrations prepared. The extract was homogenized to ensure uniform distribution of the bioactive compounds throughout the solution.

SEED TREATMENT

The seeds were immersed in the different concentrations of pyroligneous extract for 5 minutes in 100 mL of solution for each concentration. The seeds were immersed in sterile beakers to avoid any contamination. The seeds in the control group were subjected to the same procedure, but immersed in distilled water (0% v/v pyroligneous extract) to compare the effects of the concentrations of pyroligneous extract with the conventional treatment.

Image 1: Seeds before being immersed.

Image 2: Seeds immersed in the solution.

TEST INSTALLATION AND EVALUATION

Incubation on Paper Substrate or Filter Paper method ("blotter test")

PROCEDURES

The seeds were placed separately on a layer of damp filter paper, using two sheets of blotting paper. They were placed in gerbox containers, keeping a distance of 1 to 2 cm between them. These containers have transparent lids that allow all the incident light to pass through.

Image 3 and 4: Incubation of chickpea seeds treated with pyroligneous extract Eucalyptus sp.

INCUBATION CONDITIONS

The containers with the seeds were placed under white fluorescent light bulbs at a distance of 30-40 cm in chambers with a 12-hour photoperiod for 8 days at a temperature of 20°C.

SEED EVALUATION

The seeds were examined individually, considering each treatment, using a stereomicroscope with a magnification of between 30 and 80 times to identify the presence of typical fungal growth structures, such as conidiophores with conidia and fruiting bodies (e.g. pycnidia, acervuli, perithecia), as well as their inhibition at each concentration. These characteristics are essential for identifying fungal species.

After the incubation period, the fungal incidence was evaluated under a stereoscopic microscope and/or slides were mounted for the morphological identification of fungal genera under an optical microscope.

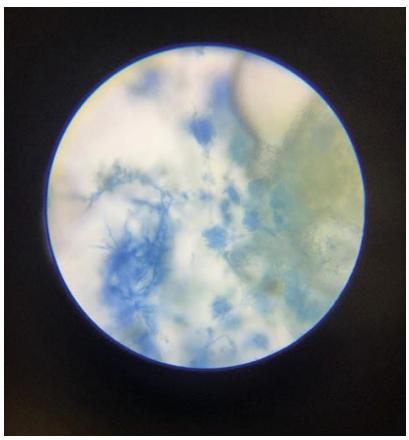

Image 5: slide of *Penicillium* sp**.**

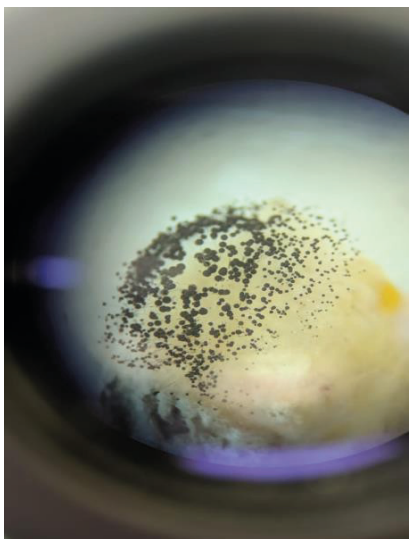

Image 6: *Aspergillus* sp.

RESULTS AND DISCUSSION

Analysis of the fungal incidence associated chickpea seeds revealed the presence of four predominant fungal genera: *Penicillium sp.*, *Aspergillus sp.*, *Rhizopus sp.* and *Fusarium sp.*, with *Penicillium sp*. showing the highest incidence, reaching 33% of the total, followed by *Aspergillus sp.* (12%), *Fusarium sp*. (9.5%) and *Rhizopus sp*. (3.6%). The highest incidence of *Penicillium sp*. (69%) and *Rhizopus sp.* (12%) was observed in the , i.e. in seeds that had not been treated with eucalyptus pyroligneous extract.

INCIDENCE (%)				
Concentrations (%)	*Penicillium* sp.	*Rhizopus* sp.	*Aspergillus* sp.	*Fusarium* sp.
0	69	12	2	0
5	56	8	0	0
25	22	1	0	0
50	6	0	0	10
75	0	0	0	9

Table 1: Fungal incidence in chickpea seeds after treatment with *Eucalyptus* sp. pyroligneous extract at different concentrations.

The application of *Eucalyptus sp.* pyroligneous extract proved to be effective in fungal control, especially as the concentration of the extract increased. Although the increase in concentration contributed to a significant reduction in the incidence of *Penicillium sp.*, *Rhizopus sp.* and *Aspergillus sp*., the *Fusarium sp.* species did not show such a significant reduction, as shown in table 1. With the extract concentrated at 75%, a reduction of up to 91% in fungal incidence was observed, indicating that eucalyptus pyroligneous extract has significant potential for controlling fungal infections associated with chickpeas.

This fungal control was most evident in the genera *Penicillium sp.*, *Rhizopus sp.* and *Aspergillus sp*., which suggests that the pyroligneous extract of *Eucalyptus sp.* may have a more robust effect against these fungi in par-

ticular. The phenolic compounds and organic acids present in the extract probably play an important role in inhibiting fungal growth, interfering with the pathogens' cell membranes and the metabolic processes essential for their survival.

The reduction in fungal incidence, especially with the higher concentration of pyroligneous extract, is an indication that the use of the extract could be a promising strategy for controlling fungal diseases in chickpeas, especially during the storage phase, where fungal contamination is more prevalent. In addition, the use of a natural product such as pyroligneous extract offers ecological and safety advantages, providing a sustainable alternative to synthetic fungicides.

It is important to note that, despite the promising results, more studies are needed to evaluate the efficacy of *Eucalyptus sp.* pyrolene extract in field conditions and in different types of soil, as well as investigating the interaction of the extract with other integrated pest and disease management practices. Additional tests should also be carried out to analyze possible effects of the extract on plant health and the viability of plants seeds, ensuring that fungal control does not harm the productivity and quality of the chickpeas.

CONCLUSION

Analysis of the fungal incidence on chickpea seeds showed that the pyroligneous extract of *Eucalyptus* sp. has significant potential for controlling fungal infections, especially against the genera *Penicillium* sp., *Aspergillus* sp. and *Rhizopus* sp. Application of the extract, especially at the higher concentrations, resulted in a significant reduction in fungal incidence, with a decrease of up to 91% at the 75% concentration. This effectiveness suggests that pyroligneous extract could be a promising alternative to the use of synthetic fungicides, offering ecological and safety benefits.

The phenolic compounds and organic acids present in the eucalyptus extract play an important role in inhibiting fungal growth, demonstrating the feasibility of using natural products in plant health management, particularly in seed storage situations. However, further studies are needed to assess the efficacy of the extract in field conditions, in different types of soil and in conjunction with other integrated pest and disease management practices. Furthermore, additional research is essential to assess the possible impacts of the extract on plant health and seed viability, ensuring that fungal control does not negatively interfere with chickpea productivity and quality.

Therefore, the use of pyroligneous extract from *Eucalyptus* sp. appears to be a promising strategy for the sustainable control of fungal diseases, although more tests and evaluations are needed to consolidate its application on a large scale. In addition, more advanced studies should be carried out to discuss the factors that lead to the increase in Fusarium sp., in order to arrive at a more stable concentration that controls all the fungi mentioned equally.

Furthermore, it is imperative that more in-depth studies are conducted in order to investigate the factors that contribute to the increase in the incidence of *Fusarium sp.*, with the aim of determining conditions that allow a stable concentration of the pathogen to be achieved. These studies should explore the mechanisms that favor the proliferation of this fungus, as well as the environmental, genetic and agronomic factors that influence its dynamics. A detailed understanding of these aspects is crucial for developing more effective management strategies capable of controlling not only *Fusarium sp.* but also other pathogenic fungi that affect crops. In this way, it will be possible to establish balanced and sustained control, minimizing the impact on agricultural production and ensuring plant health stability.

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