

FUNGICIDAL EFFECT OF *Equisetum hyemale* EXTRACT OBTAINED BY ALKALINE HYDROLYSIS

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ABSTRACT

Concern about preserving the environment and human health is growing in the agro-industrial sector. Therefore, the search for alternatives that are beneficial to these aspects has received significant attention. In this project, the objective was to produce an extract of organic components that would be beneficial to the environment, human health and effective in plant nutrition. The *Equisetum hyemale* (Horsetail) plant was selected due to the presence of Silicon in its composition, which has fungicidal properties. The study aimed to evaluate the alkaline Horsetail extract in combating the fungus *Exserohilum turcicum*, known as leaf spot, which can negatively affect agricultural production. The extract production process involved steps such as harvesting, pre-drying, grinding, alkaline hydrolysis and filtering. To test its effectiveness, the fungus was inoculated in contact with the extract in Petri dishes containing specific culture medium. The results showed that the alkaline extracts obtained an IVCM of 50% and PIC of 65%, proving their effectiveness in antifungal control.

Keywords Environment preservation, Human health, Agro-industrial sector, *Equisetum hyemale* (Horsetail), Antifungal control.

1 INTRODUCTION

In Brazil, losses in soybean production due to competition with weeds represent a major challenge, affecting up to 30% of the main crops and resulting in significant losses of around 9 billion reais. Furthermore, plantations face threats from various pathogens, which generally require the use of agrochemicals for control, although this can have harmful effects on the environment and human health^{3,5}.

Given this scenario, the evolution of agrochemicals is essential to meet the country's food demand without compromising environmental preservation and public health. In this context, studies indicate the potential of medicinal plants as an alternative to conventional agricultural pesticides, presenting advantages such as lower costs, lower environmental impact and greater accessibility for farmers^{2,3}.

Equisetum hyemale (Horsetail), a medicinal plant known for its therapeutic properties in humans, also shows promise as an agricultural pesticide due to the presence of Silicon in its composition, which assists in the plants' immune response. This study aims to evaluate the effect of soluble horsetail extracts on the growth of the phytopathogen *Exserohilum turcicum*, indicating a significant potential for activating the plant defense response against diseases in agricultural crops, especially soybean^{4,7,8}.

2 MATERIAL & METHODS

For extraction, the solvent used was KOH (Potassium Hydroxide), diluted in concentrations of 1, 2, 4, 8, 12 and 16%. The mackerel mass used was set at 10 g for 90 g of solvent, varying the concentrations of KOH (Potassium Hydroxide).¹

The preparation of the solutions was carried out in a 500 mL Erlenmeyer flask, subsequently closing the entrance hole with cotton. In this way, the glassware with KOH and Horsetail solution inside was prepared for autoclave extraction.

Alkaline extraction occurred using pressure of 1.21 kgf cm⁻² for 2 hours in an autoclave. After treatment, the solution was subjected to vacuum filtration, separating the solid part from the liquid part of the solution, obtaining the horsetail extract through alkaline means.

For the inoculation of the *Exserohilum turcicum* fungus, the extract that showed the best use of silicon absorption was selected. This analysis was carried out once for each type of extract, to define the result with the best extraction of silicon from the plant. Within this, the production of all proposed extracts followed the flowchart shown in figure 1.

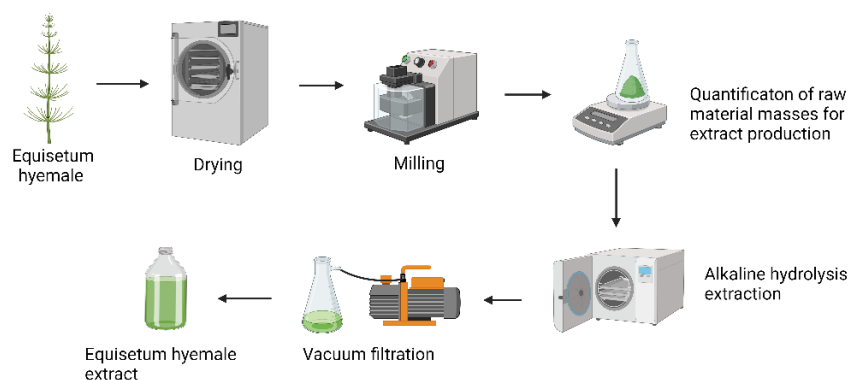


Figure 1 Production flowchart of *Equisetum hyemale* extract by alkaline hydrolysis.

After producing the extracts, they were subjected to the vacuum filtration process, where it was possible to separate the solid part from the liquid part. Therefore, analyzes of PIC (Mycelial Growth Inhibition), IVCM (Mycelial Growth Speed Index) and silicon were carried out to prove the effectiveness of the horsetail extract.

3 RESULTS & DISCUSSION

Os After extract production using KOH, silicon in the extract was quantified. Table 1 shows the percentage of silicon found in the extracts in which the concentration of KOH was varied, so that the mass of horsetail used in the extracts was 10 g for each one

Table 1 Percentage of silicon in KOH extract

% KOH	m Horsetail	% Si	m extract	g extracted silicon
1	10	0,26	42,65	0,11089
2	10	0,41	66,67	0,27335
4	10	0,65	66,06	0,42939
12	10	0,62	73,51	0,45576
16	10	0,66	64,05	0,42273

With the data obtained in the extraction and percentage of silicon in the extract, the relationship between the percentage of KOH concentration and the percentage of silicon and extract mass is found. These data are presented in figure 2.

Analyzing Figure 2, it can be stated that the best combination of solvent and horsetail solid was 4% KOH, obtaining a standard extract mass between the solvents of 66.06 g and 0.65% silicon, demonstrating better performance among the other concentrations analyzed. The extract with 12% KOH obtained similar values, however, more than twice as much KOH was used and a lower percentage of silicon in the extract.

The extract with 16% demonstrated a higher percentage of silicon, however, it provided a lower mass of extract during extraction and required four times more KOH compared to the best result. With 1% KOH, the extract showed lower mass yield and silicon percentage, with 2% an improvement in extract mass yield was obtained, however, satisfactory values for silicon percentage were not obtained compared to 4% KOH.

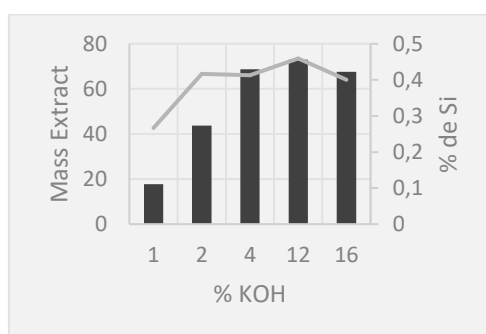


Figure 2 Graph of the concentration relationship of % KOH with % Si

Using the best combination of alkaline extract, microbiological analysis was carried out, using the compound to attack the fungus *Exserohilum turcicum*. The results were analyzed by the Mycelial Growth Velocity Index (IVCM) and the Percentage of Mycelial Growth Inhibition (PIC). The microbiological test was carried out using the same parameters as the alcoholic extract. Figure 3 shows the IVCM of the alkaline extract, which demonstrates a reduction in mycelial growth speed of around 50%.

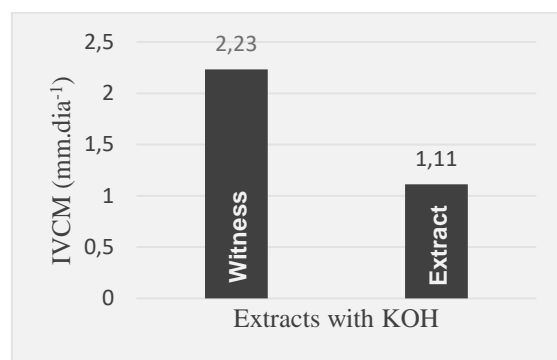


Figure 3 Graph of mycelial growth rate index (ivcm) with KOH

The PIC analysis of the alkaline extract was presented in table 2, where we obtained an average of 63.63% inhibition in the first 4 days. Subsequently, the extract began to decrease in effectiveness.

Table 2 Results of Percentage of Mycelial Growth Inhibition (PIC) with KOH

	2ºdia	4ºdia	6ºdia	8ºdia
PIC	65,57%	61,68%	52,19%	14,02%

4 CONCLUSION

Based on the Mycelial Growth Speed Index (IVCM) and the Mycelial Growth Inhibition Percentage (PIC), *Equisetum hyemale* extract has potential as a substitute for pesticides, acting as an inducer of plant resistance. Future research should focus on agronomic tests to prove its effects in the field.

REFERENCES

- HIJAZIN, Carlos Atalla; SIMÕES, Aline Toniai; SILVEIRA, Diogo Rhoden. (2010). Hidrólise ácida, alcalina e enzimática. Revista Atitude. 7. ed. Rio de Janeiro. Disponível em: <https://www.tratamentodeagua.com.br/wp-content/uploads/2016/10/Hidr%C3%B3lise-%C3%A1cida-alcalina-e-enzim%C3%A1tica.pdf>.
- CARNEIRO, Fernanda Melo; SILVA, Maria José Pereira da; BORGES, Leonardo Luiz; ALBERNAZ, Lorena Carneiro; COSTA, Joana Darc Pereira. (2014). Tendências dos estudos com plantas medicinais no Brasil. p. 1-32.
- FOWLER, João. 2020. Defensivos Agrícolas: saiba o que são, os tipos e a importância dos agroquímicos para a produção rural. Tecnologia do Campo, Agricultura, abr.
- GONÇALVES, Ricardo Marcelo *et al.* (2013). Mancha-foliar-de-Phaeosphaeria (mancha-branca-do-milho): fungo ou bactéria? Embrapa Milho e Sorgo. Boletim de Pesquisa e Desenvolvimento, Sete Lagos-Mg, v. 21, n. 79, p. 1-37.
- GUIMARÃES, SS; MAZARO, SM; FREDDO, ÁR; WAGNER JÚNIOR, A. (2015). Potencial de preparados de cavalinha (*Equisetum* sp.) na síntese de metabólitos de defesa em cotilédones de soja (*Glycine max* L.) e o efeito sobre o crescimento de *Rhizoctonia solani* Kuhn, in vitro. Revista Brasileira de Plantas Mediciniais. p. 1-7.
- OLIVEIRA, João Almir. (2020). Efeito do tratamento fungicida em sementes e no controle de tombamento de plântulas de pepino (*Cucumis sativus* L.) e pimentão (*Capsicum annum* L.). 111 f. Dissertação (Doutorado) - Curso de Agronomia, Escola Superior de Agricultura de Lavras, Lavras, 1991. Disponível em: <http://repositorio.ufla.br/handle/1/33483>. Acesso em: 26 ago. 2020.
- QUEIROZ, G. M.; SOUZA-MOREIRA, T. M.; SALGADO, H. R. N.; MOREIRA, R. R. D.; UTRERA, S. H.; MARTINS, C. H. G.; PIETRO, R. C. R. L. (2014). Antimicrobial activity and toxicity in vitro and in vivo of *Equisetum hyemale* extracts. Revista de Ciências Farmacêuticas Básica e Aplicada, v. 35, n. 4, p. 559-563.
- PEREIRA, Liliana Avelar *et al.* (2020). Fungitoxicidade in vitro de Iprodione sobre o crescimento micelial de fungos que se associam a sementes de arroz. 2002. Disponível em: <https://www.scielo.br/pdf/rbs/v24n1/v24n1a10.pdf>.

ACKNOWLEDGEMENTS

I thank SATIS (SATIS Industry and Commerce Ltda), FAZU (Associated Faculty of Uberaba), UNIUBE (University of Uberaba) and FAPEMIG (Minas Gerais State Research Support Foundation), for all the equipment and materials made available to the project, and to all the help I received from these institutions.