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Fungitoxic activity of *Zuccagnia punctata* extracts

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ABSTRACT

The antifungal activities of aqueous (infusion and decoction) and ethanolic (tincture) extracts from a medicinal plant, *Z. punctata*, were evaluated on important fungal pathogens isolated from major crops of Argentina. Fungal species were *Fusarium oxysporum*, *F. thapsinum*, *F. verticillioides*, *Macrophomina phaseolina and Rhizoctonia solani*. Percentage of hyphal growth inhibition was evaluated by agar dilution method. Tincture was the most fungitoxic extractive form assayed. Inhibition of mycelial growth increases with concentration. Our results suggest that *Z. punctata* has a broader fungicidal activity than that previously showed and most fungicidal principles were in the tincture. Further research is needed to identify and characterize these bioactive compounds.

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Keywords: *Z. punctata*, fungal pathogens, extract and crops in Argentina

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Introduction

Phytopathogenic fungi reduce 20% harvest yields in Argentina and are often controlled using synthetic fungicides. Nevertheless, these compounds suffer a loss of biocide effectiveness and have a slow biodegradation (Brady, 1984). Plants can provide more environmentally friendly fungitoxic compounds (Quiroga *et al*., 2001). One example is *Zuccagnia punctata* (Cav.) which showed antifungal activity (Quiroga *et al*., 2001). This medicinal plant is used as antiseptic pedic, and to treat arthritis, rheumatism, fever, edema, infections and tumors (Ratera *et al*., 1980). The aim of this study was to evaluate the antifungal activity of aqueous and ethanolic extracts of *Z. punctata* on pathogens affecting production and marketing of major crops in Argentina.

Experimental

Zuccagnia punctata Cav. was collected in Tucuman province (Argentina) and identified at Instituto de Estudios Vegetales "*Dr. A. R. Sampietro*" (FBQF, UNT, Tucumán, Argentina). Phytopathogenic fungi were isolated by INTA (Pergamino-Leales) and Sección Fitopatología of EEAOC (Tucumán): *Rhizoctonia solani*, *Fusarium oxysporum*, *F. verticillioides*, *F. thapsinum* and *Macrophomina phaseolina*. They were cultured and maintained in modified rose bengal agar (RBA) and modified peptone malt agar (PMA).

Aerial parts of *Zuccagnia punctata* were dried at 45 °C for 5 days. The dry material was powdered and extracted by: 1) Decoction: 5 g of powder in 50 ml of boiling distilled water, boiled for 20 min. 2) Infusion: 5 g of powder in 50 ml of boiling water. Both decoction and infusion were filtered through Whatman No. 1 filter paper and lyophilized. 3) Tincture: 10 g of powder in 100 ml ethanol 96%. After incubation at 37°C (40 cycles/min) for 7 days, suspension was filtered through Whatman No. 1 filter paper and dried at 30 °C.

Dry residues were weighed, dissolved in methanol (concentrated extract, CE) and kept at -20ºC. An aliquot of each CE was evaporated at 40ºC. Dry weight was extracted material (EM) of each extracting form. Extraction efficiency was calculated.

Each CE was diluted with ethanol 96% to 4, 8, and 16 mg of EM/ml. 0.5 ml of each dilution was incorporated into 4.5 ml of warm PMA medium obtaining final concentrations of 0.4, 0.8 and 1.6 mg EM/ml. Controls were 0.5 ml of ethanol 96% in 4.5 ml of PMA. 3 mm diameter-mycelial plugs were placed in the center of PMA plates. Then, they were incubated at 30°C for 3 to 4 days in a moist environment. Average diameter of mycelial growth was determined. Percent of growth inhibition was calculated: % inhibition = [(MGC-MGE)x100]/MGC. Where MGC is average diameter of mycelial growth in the control and MGE is average diameter of mycelial growth in the presence of a tested extract dilution.

Results and discussion

Zuccagnia punctata inhibited growth of the phytopathogenic fungi tested. Antifungal activity was higher at increasing concentrations of assayed extracts (Fig 1,2 and 3).

Tincture had a higher extraction yield (35.7%) than infusion (21.2%) and decoction (24.5%). Tincture exerted a stronger growth inhibition than the aqueous extractive forms. This situation was also observed for other folk argentinian plants (Davicino *et al*., 2007).

In our work, decoction inhibited the growth of *F. thapsinum*, *M. phaseolina* and *R. solani* more than infusion. Conversely, *F. oxysporum* was more sensitive to infusion. In the case of *F. verticillioides*, the inhibitory effect of decoction and infusion was similar.

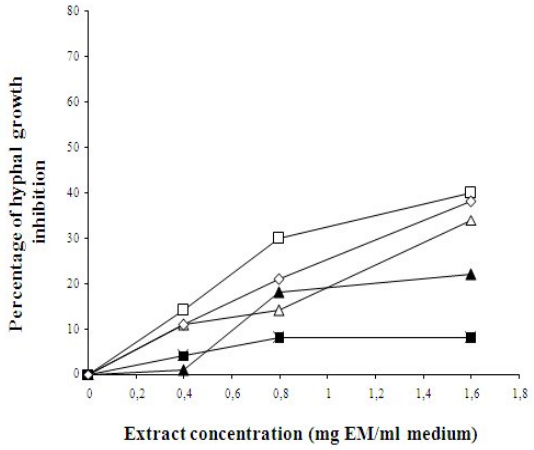

Figure 1. Antifungal activity of infusion obtained from *Z. punctata*.

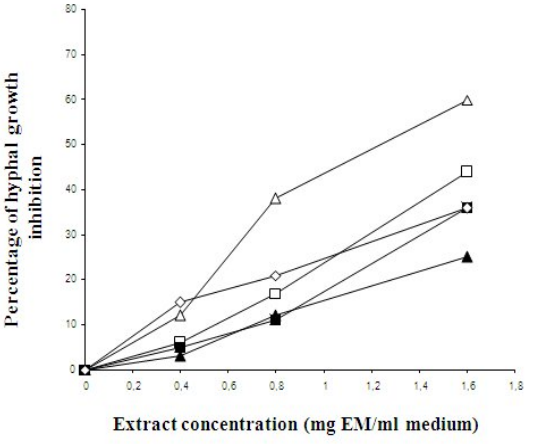

Figure 2. Antifungal activity of decoction obtained from *Z. punctata*.

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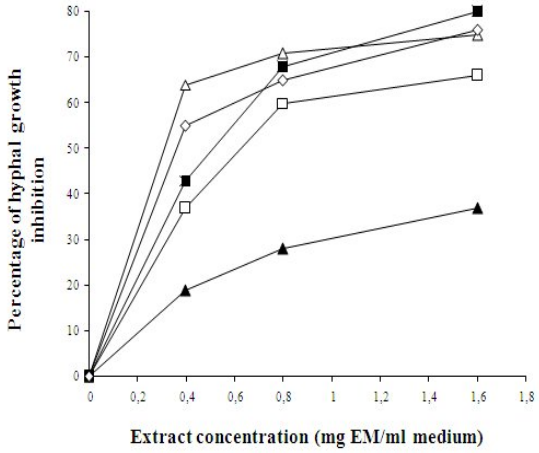

Figure 3. Antifungal activity of tincture obtained from *Z. punctata.*

Conclusions

Our results suggest that *Z. punctata* has a broader fungicidal activity than that previously showed (Quiroga *et al.*, 2001). Most fungicidal principles were in the tincture. Further research is needed to identify and characterize these bioactive compounds.

Acknowledgements

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