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Plant metabolites as potential food preservatives

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INTRODUCTION

Inadequate techniques of culture, recollection and storage of food and forages, can lead to fungi development producing toxic chemical compounds. Fungal species producers of mycotoxins mainly belong to the genera *Aspergillus*, *Penicillium* and *Fusarium*, being aflatoxins, ochratoxins, phomopsins, slaframine and patuline the best known mycotoxins. Occurrence of mycotoxins in food causes loss of the nutritious value, and produces harmful effects on human and animal health.

The aims of this study were: (a) To determine the ability to produce aflatoxins in four (4) *Aspergillus* strains isolated from balanced food used for farm animals. (b) To analyse *in vitro* fungitoxic effect of alcoholic extracts of *Tripodanthus acutifolius* and *Larrea divaricata*, plants used in the popular medicine of Tucumán region (Argentine).

METHODOLOGY

Aspergillus strains used in this work were *A. flavus*, *A. parasiticus* NRRL22, *A. nomius* VSC23 and *A. nomius* 13137. They were cultured in a basal medium which contained 0.5% peptone, 2% malt extract and 2% glucose, added with 0.6 or 1.8% agar.

Tested plant species were *Tripodanthus acutifolius* (Ruiz & Pav) Van Thieghem ('corpo') and *Larrea divaricata* Cav ('jarilla'). Extracts were prepared with dry and powdered aerial parts (10% w/v) with 96° ethanol according to the Pharmacopeia Argentina 6th edition.

Aflatoxicity of the fungal species was determined by thin-layer chromatography (TLC) and observation under long UV light at 366 nm (González *et al.,* 2005); by pigment production (Shier *et al*., 2005), and by observation of mycelium colour changes in the presence of alkaline and acid solutions (Saito and Machida, 1999).

In vitro fungitoxic activity was tested by several methods: bioautography, radial growth inhibition in plates and inhibition percentage determination (Reyes Chilpa *et al*., 1997), and zonal inhibition of hyphas development. Minimum Inhibitory Concentration (MIC) of the extracts was determined by microdilution in 96-wells plates. Likewise MIC values of the substances used for food protection against polluting microorganisms were determined.

RESULTS AND DISCUSSION

Of the four *Aspergillus* strains isolated from balanced food, only two of them (*A. nomius* VSC23 and *A. nomius* 13137), showed to be aflatoxigenic strains. From the culture medium where both strains developed, a compound was isolated and identified as Aflatoxin B_1 by TLC and comparison with a mycotoxin standard. These findings were confirmed by isolation of the yellow pigment that changed colour according to pH to which was exposed.

Previous reports had shown the antifungic effect of several alcoholic extracts from plants collected in Tucumán and used in popular medicine on the 4 isolated strains of *Aspergillus* (Table 1). The tinctures of *T. acutifolius* and *L. divaricata* were selected to carry out this work

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because of their highest biocide activity.

The fungitoxic efficiency of the mentioned extracts was proved by determining MIC values of both extracts, which were compared with the MICs of organic acids (ascorbic acid and sorbic acid) usually used as food preservatives. Quite similar values were obtained between both types of antifungic substances as shown in Table 2.

CONCLUSIONS

These results suggest that some metabolites may be isolated from plant species of northwestern Argentine (*T. acutifolius* and *L. divaricata*), which could be used to avoid polluting fungi growth and further synthesis of mycotoxins, thus being useful for food and stored forages conservation.

Table 1. Percentage of radial growth inhibition in plates (0.3 mg phenolic compounds/mL of culture medium)

Plant extracts	Fungi			
	1	2	3	4
Tripodanthus acutifolius	89.2	87.5	88.4	76.8
Larrea divaricata	54.2	36.3	49.2	54.1
Zuccagnia punctata	29.1	16.0	30.0	31.2
Geoffroea decorticans	18.6	27.7	39.6	18.4
Phytolacca dioica	5.0	0.0	39.0	6.4
Schinus molle	0.2	0.3	1.4	5.2

1. Aspergillus flavus, 2. Aspergillus parasiticus NRRL22
3. Aspergillus nomius 13137, *4. Aspergillus nomius* VSC23
n=12

Table 2. MICs of plant extracts and synthetic preservatives agents

Antifungal Substances	Fungi	
	A. nomius 13137	*A. nomius* VSC23
T. acutifolius (tincture)	50 µg PC/mL	50µg PC/mL
L. divaricata (tincture)	100 µg PC/mL	50 µg PC/mL
Ascorbic acid	200 µg/mL	150 µg/mL
Sorbic acid	100 µg/mL	400 µg/mL

MICs of plant extracts are given in microg of Phenolic Compounds/ mL
Synthetic agent' MICs in microg/mL
PC: Phenolic Compounds n= 8

REFERENCES

González O., Hinojo M. J., Mateo R., Medina A. and Jiménez M. (2005) Occurrence of mycotoxin producing fungi in bee pollen. *International Journal of Food Microbiology* **105**: 1-9.

Reyes Chilpa R., Quiroz Vázquez R.I., Jiménez Estrada M., Navarro Ocaña A. and Cassani Hemández J. (1997) Antifungal activity of selected plant secondary metabolites against *Coriola versicolor*. *Journal of Tropical Forest Products* **3**: 110-113.

Saito M. and Machida S. (1999) A rapid identification method for aflatoxin-producing strains of *Aspergillus flavus* and *Aspergillus parasiticus* by ammonia vapor. *Mycoscience* **4**: 205-208.

Shier W. T, Lao Y., Steele T. W. and Abbas H. K. (2005) Yellow pigments used in rapid identification of aflatoxin-producing *Aspergillus* strain are antraquinones associated with the aflatoxin biosynthetic pathway. *Biorganic Chemistry* **33**: 426-438.