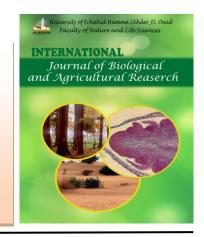
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# Biofungicide Activity of *Datura stramonium* Leaf Extract Against Phytopathogenic Fungi

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### Abstract

The aim of this study is determination of polyphenol contents and biofungicide activity of crude extract of *Datura stramonium*. The results of extraction showed an important yield percentage of crud extract equal 25.22 % wherase, the polyphenol content of the extract was 0,756 (mg AGE/ml Ext). The *In vitro* biofungicide activity carried out by disk diffusion method, the biofungicide effect results performed with crude extract and Milraz WP 76 fungicide (Propineb 70%, Cymoxanil 6%) against fungal strains. The results appear that natural substances possess an antifungal power on the multiresistant germs responsible for infectious diseases for plants. Growth inhibition varies depending on the fungal strains and the concentration of the test product.

Keywords: Datura stramonium, Milraz WP 76, Propineb, Cymoxanil, Biofungicide activity.

### **1. Introduction**

Crop protection plays an essential role in ensuring food safety, and disease is probably the biggest constraint to increase crop production and yield, and one of the major factors limiting their quality. Moulds and their mycotoxins cause losses of 5 to 10% of cereals and their derivatives [1].

Mould reduces technological and health quality (Gluten content, allergy, toxic agents responsible for serious human and animal poisonings: Mycotoxins), reducing the nutritional value, modifying the organoleptic aspect and finally causing economic problems due to the costs of detoxification of grains or discharges of contaminated products [2]

Research and development of new bioactive molecules such as: Phenolic compounds that can more effectively act on new generations of resistant microorganisms and that preserve the environment and humans [3,4]. Phenolic compounds are secondary metabolites of plants. Allowing to resist various attacks. They participate very effectively in the tolerance of plants with various stresses. From a therapeutic point of view, these molecules constitute the basis of the active ingredients found in medicinal plants [5].

This study evaluated the *In vitro* biofungicide activity of phenolic compounds in crude extract of *Datura stramonium* plant on the growth of some phytopathogenic fungi.

### 2. Materials and methods

### 2.1. Microbial material

4 fungal strains pathogenic to plants, they come from the National Plant Protection Institute, Annaba (Algeria). Fungal strains: *Alternaria solani, Aspergillus niger, Septoria nodurum, Fusarium sp.* And for culture media we used Potato Dextrose Agar (PDA).

### 2.2. Fungicide:

In order to compare the biofungicide activity of the isolated extract with those of fungicide, Milraz WP 76 fungicide (Propineb 70%, Cymoxanil 6%) this fungicide is manufactured by Bayer Crop Science. The concentration used (0.50 g/L) was determined based on the manufacturer's recommended doses on the fungicide label.

### 2.3. Preparation of plant extract

50 g of powdered leaves plant were soaked in 1000 ml of Methanol/Water (700/300 ml) for 24 hours in dark. The mixture was stirred by using a glass rod. Resulting extract was filtrated through Whatman  $N^{\circ}$ : 1. and final filtrate obtained was evaporated under reduce pressure then dried. The percentage yield of crud extract was calculated and stored in refrigerator at 4 ± 1 °C for further use. The concentration used in the experimental was based on the dry weight of the extract.

### 2.4. Determination of total phenolic content

100  $\mu$ l of the sample was added to 200  $\mu$ l of Folin–Ciocalteu reagent. After 3 min, 600  $\mu$ l of saturated sodium carbonate solution (about 20%) was added. After 2 h of incubation at room

temperature, the absorbance of the reaction mixture was measured at 765 nm. The same procedure was repeated to all standard gallic acid solutions (0 - 100  $\mu$ g/ml) and standard curve was obtained [6].

## 2.5. Biofungicide activity

We used the solid medium diffusion technique for biofungicide activity [7]. This method consists in determining the sensibility of fungal strains against one or more products. A sterile paper disc with 6 mm of diameter is immersed in the products to be tested (Pure Extract, 1/2 Extract, 1/4 Extract). The extract is diluted in dimethylsulphur DMSO, The three wet discs of three concentrations are placed on the PDA medium that is previously inoculated with strains (Inoculum concentration equal to  $10^6$  à  $10^8$  CFU/ml). The plates are incubated at  $37^{\circ}$ C for 18 to 24 hours. Comparison discs (Milraz WP 76 fungicide) are included in the tests. Each test of the biofungicide activity is effected in triplicata.

### 3. Results

### 3.1. Extraction yield of plant

The Extraction yield for *Datura stramonium* crud extract is in the order of 12.61 g which corresponds to a percentage equal to 25.22 % (Figure 1).

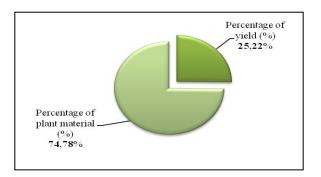


Figure 1: Extraction yield of Datura stramonium crud extract

### 3.2. Total phenol content

The content of phenolic compounds in *Datura stramonium* crud extract determined using regression equation of calibration curve and verified when the correlation coefficient is not less than  $R^2 = 0.992$ . The results are expressed as mg Gallic Acid Equivalent per ml of Extract (mg AGE/ml Ext) (Table 1).

**Table 1:** Total phenolic content of *Datura stramonium* crud extract

	Total phenolic content (mg AGE/ml Ext)		
Crud extract	$0,756 \pm 0,06$		

### 3.3. Biofungicide activity

The results of the various biofungicide activity tests for *Datura stramonium* crud extract are grouped in (Tables 3). All data were compared with Milraz WP 76 fungicide.

	Products tested			
Strains	Pure	1/2	1/4	Milraz
	Extract	Extract	Extract	
Alternaria solani	7,6 ±0,3	$0\pm00$	$0\pm 00$	10,1 ±0,5
Fusarium sp	$7,5\pm 0,2$	$0\pm00$	7,4 ±0,3	8,4 ±0,3
Aspergillus niger	$7,1\pm 0,5$	$0\pm00$	$7,4\pm 0,1$	$8,2\pm 0,4$
Septoria nodurum	$7,3\pm 0,2$	$0\pm00$	$0\pm 00$	8,1 ±0,2

**Table 2:** Diameters of the inhibition zones (in mm) of fungal strains growth against products tested.

*Values represent the mean standard*  $\square$   $\square$  *deviation (n=3)* 

#### 4. Discussion

The pathway to new natural substances as chemical agent alternatives remains a promising solution. The results obtained of crud extract yield from *Datura stramonium* leaves plant is higher (25.22 %). Levels of phytochemical compounds may be affected by the genotype, the conditions for development and growth, the maturity, storage conditions and extraction methods [8].

*Datura stramonium* is widely growing plant and well known to have great pharmacological potential with a great utility and usage in folklore medicine folklore medicinal herbs [9] and use in medicine because of its natural substances such as glycosides, phenol, lignins, saponins, sterols and tannins [10]. The primary biologically active substances in *Datura stramonium* are the alkaloids atropine and scopolamine [11].

According to Usha et *al* [12] research the leaf extract of *Datura stramonium* was found to exhibit excellent antifungal activity against *Fusarium mangiferae*, especially when using methanol-water (70/30 v/v) for obtained of crud extract. Another study by Hadjimi [13] obtained similar results with our results; the crud extract of *Datura stramonium* with  $125*10^{-4}$ ,  $125*10^{-5}$  et  $125*10^{-6}$  mg/ml concentrations, the plant prevents the growth and development of tow strain *Fusarium oxysporum f.sp albedinis (Killian et Maire)*.

### **5.** Conclusion

In biofungicide activity study we have found that *Datura stramonium* leaf extract (Pure Extract) shows antifungicide activity against microorganisms: *Fusarium sp*, *Alternaria solani*, *Septoria nodurum* and *Aspergillus niger* by forming zone of inhibition and lower of the others forming by Milraz. From this study we can conclude that *Datura stramonium* has biofungicide activity against some phytopathogenic fungi.

### 6. Acknowledgement

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