Evaluation of Antifungal Properties of Leaf Extracts from Botanical Sources against Seed-Borne Fungal Pathogens in Selected Gourd Varieties

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Abstract:

Known for their nutritional abundance in folate, iron, vitamins, calcium, and proteins, gourd vegetables in the Marathwada region of Maharashtra include bottle gourd, bitter gourd, and ridge gourd. Studies show that a large amount of fungal flora is present in these vegetables and their seeds both in the fields and during storage. We investigated several gourd seeds using glucose nutrition agar and regular blotter paper techniques, and we found twelve different kinds of fungus. Primary among these were *Aspergillus niger, Aspergillus flavus, Alternaria alternata,* and *Fusarium oxysporum*. Diseases can be caused by pathogens that are spread by seeds during different phases of plant development, from seed to seedling. Given that biological approaches have less of an environmental effect than synthetic pesticides, we support them as better options. The purpose of this work is to investigate the possibility of plant extracts in protecting seeds and improving germination rates in an ecologically friendly way by fighting seed-borne infections

Keywords: Leaf extract, Gourd vegetables, Seed-borne fungi, Marathwada region, Biological methods

INTRODUCTION

The seed, which functions as a little plant, is crucial to agricultural output. Roughly 90% of the food crops grown worldwide are propagated through the use of seeds. When it comes to calculating the total biological yield per unit of time and per unit of plant surface area, seeds are essential. A wide range of diseases can affect crops, most of which spread through seeds. According to Anwar *et al.* (2012), seed-borne infections can transmit a number of serious diseases that are caused by living organisms and result in considerable losses in crop productivity. These infections fall into two groups: those that live inside the seed and those that stick to the outside seed covering. It has been shown that there are over 1,500 seed-borne bacteria that impact about 600 genera of agricultural plants (Agarwal & Sinclair, 1993). In agricultural fields, seed-borne infections frequently turn into harmful disease agents. Every seed contains a distinct microbiota that can greatly affect the likelihood of disease development over the course of several growing seasons or from one season to the next.

It has been reported all around the world that different fungi can grow on vegetable seeds (Ismail et al., 2012; Idrees et al., 2000; Al Kassim and Monawar, 2000; Esuruoso et al., 1975; Karwasra and

Singh, 1982). Scientific surveys have regularly revealed common seed-borne organisms, such as *Alternaria*, *Aspergillus*, *Rhizopus*, *Penicillium*, *Fusarium*, *Curvularia*, *Humicola*, and *Cladosporium*, on spices, condiments, and vegetable seeds.

Managing crop diseases critically depends on testing the health of seeds to identify seed-borne pathogens and putting control measures in place. In order to discover seed-borne fungi and investigate other strategies for reducing seed bio-deterioration and increasing germination rates in an environmentally responsible manner, the current study concentrated on gourd seeds, namely bottle, bitter, and ridge gourd varieties.

MATERIALS AND METHODS

The current work is divided into two stages:

A) Evaluation of seed-borne pathogens.

B) Evaluation of the efficacy of medicinal plants against seed-borne pathogens.

A) Evaluation of seed-borne pathogens.

Selection of seeds: Gourd seed samples were procured from the town of Udgir's local market.

Sr. No.	Vernacular Name	Scientific Name
1.	Bitter gourd	*Momordica charantia* L.
2.	Bottle gourd	*Lagenaria siceraria* MoL
3.	Ridge gourd	*Luffa acutangula*

Table 1: Gourd seeds

Mycoflora Study:

Two significant, widely used techniques—the glucose nutrient agar method and the conventional blotter paper method—were employed to analyze the mycoflora of seeds.

Blotter paper method:

In the Blotter test method, fungal growth was monitored for eight days after seeds were positioned equidistantly in Petri dishes and incubated.

Glucose Nutrient Agar method (GNA):

Using the GNA method, sterile Petri plates are filled with extract from GNA media, seeds are added, and the mixture is then cultured for eight days. Each Petri dish was checked for the presence of seed borne fungal infections following the eight-day incubation period.

Percentage of incidence of mycoflora was calculated as:

$$\% \text{ incidence} = \frac{\text{No.of Seeds on which a Species Appeared}}{\text{Total Number of Seeds Observed}} \times 100$$

Percentage of seed germination was calculated as:

$$\% \text{ germination} = \frac{\text{No.of Germinating Seeds}}{\text{Total Number of Seeds Observed}} \times 100$$

B) Assessment of effectiveness of medicinal plants:

Collection of plants:

The plants used in this study were gathered from various parts of the town of Udgir. These plants' herbarium sheets were pressed and dried, and then their identification was verified by consulting the Shivaji Mahavidyalaya, Udgir Department of Botany and referring to the "Flora of Marathwada."

Sr. No.	Scientific Name	Family
1.	*Adhatoda vesica L.*	Acanthaceae
2.	*Azadiracta indica A. Juss*	Meliaceae
3.	*Ocimum sanctum*	Lomiaceae
4.	*Calotropis gigantea R.Br*	Apocunaceae
5.	*Datara metal L.*	Solanaceae

Table 2: Selected medicinal plants

Leaf extract Preparation:

Following cleansing under running tap water and then distilled water, the leaves of the chosen plants were cut. 100% leaf extracts of every plant were made by crushing these leaves in a crusher and pestle with distilled water in a 1:1 ratio. The leaf extracts were run through Whatman filter paper No. 1 after passing first through muslin cloth.

Selected samples of vegetable seeds from gourds were treated by a dipping technique. The seeds were submerged for one hour in 1:1 and 1:2 (100% and 50%) dilutions of the previously made leaf extracts. Following treatment, the seeds were given time to dry on filter paper before being examined for mycoflora and seed germination on regular blotter paper.

RESULTS AND DISCUSSION

The results are shown in Table 3 to 5.

Table 3: Mycoflora on Blotter paper linked to untreated gourd seed

Sr. No.	Seed sample	% incidence of Mycoflora	% of seed Germination	Fungi associated
1.	Bitter gourd	86.7	43.3	*Aspergillus niger, A. flavous, Alternaria altarnata, Fusarium oxysporum.*
2.	Bottle gourd	73.3	40.0	*Aspergillus niger, A. flavous, Fusarium oxysporum, Curvularia, Alternaria altarnata, Mucor sp.*
3.	Ridge gourd	83.3	53.3	*Aspergillus niger, A. flavous, Fusarium oxysporum, Alternaria altarnata, Chaetonium sp. Rhizoctonia*
	SE (+/-)	2.01	3.12	

Table 4: Mycoflora associated with untreated Gourd seed on GNA:

Sr. No.	Seed sample	% Incidence of Mycoflora	% of Seed Germination	Fungi associated
1.	Bitter gourd	86.7	40.0	*Aspergillus niger, A. flavous, Fusarium oxyporum, Alternaria sp., Penicillium, Rhizopus stolonifer*
2.	Bottle gourd	80.0	46.7	*Aspergillus flavous, Fusarium oxyporum., Helimanthosporium, Curvularia, Rhizopus stolonifer.*
3.	Ridge gourd	86.7	56.7	*Aspergillus niger, Fusarium oxyporum, Alternaria alternate, Chaetonium sp. Rhizopus stolonifer.*
	SE (+/-)	1.01	2.34	

Table 5: Different leaf extracts' effects on the mycoflora of seeds and their germination rate:

A. Bitter gourd (on Blotter paper)

Sr. No.	Leaf Extract	Concentration used	% of Seed Mycoflora	% of Seed Germination
1	*Azadiracta indica A. Juss*	1:1	13.3	86.7
		1:2	16.7	80.0
2	*Adhatoda vesica L.*	1:1	16.7	83.3
		1:2	20.0	83.3
3	*Calotropis gigantea R.Br*	1:1	20.0	76.7
		1:2	23.3	70.0
4	*Datara metal L.*	1:1	16.7	76.7
		1:2	20.0	73.3
5	*Ocimum sanctum*	1:1	13.3	90.0
		1:2	16.7	80.0
	SE (+/-)		0.12	0.17

B. Bitter gourd (on Blotter paper)

Sr. No.	Leaf Extract	Concentration used	% of seed mycoflora	% of seed germination
1	*Azadiracta indica A. Juss*	1:1	10.0	86.7
		1:2	10.0	83.3
2	*Adhatoda vesica L.*	1:1	13.3	73.3
		1:2	20.0	70.0
3	*Calotropis gigantea R.Br*	1:1	16.7	70.0
		1:2	16.7	66.7
4	*Datara metal L.*	1:1	16.7	83.3
		1:2	23.3	73.3
5	*Ocimum sanctum*	1:1	13.3	86.7
		1:2	20.0	70.0
	SE (+/-)		0.21	0.37

C. Ridge gourd (on Blotter paper)

Sr. No.	Leaf Extract	Concentration used	% of seed mycoflora	% of seed germination
1	*Azadiracta indica A. Juss*	1:1	16.7	86.7
		1:2	16.7	80.0
2	*Adhatoda vesica L.*	1:1	20.0	73.3
		1:2	20.0	66.7
3	*Calotropis gigantea R.Br*	1:1	23.3	80.0
		1:2	26.7	76.7
4	*Datara metal L.*	1:1	16.7	83.3
		1:2	20.0	80.0
5	*Ocimum sanctum*	1:1	13.7	86.7
		1:2	16.7	73.3
	SE (+/-)		0.14	0.21

Using Blotter paper and Glucose Nutrient Agar plates, the fungal incidence in seeds of three distinct gourd vegetables was measured over a course of eight days. Tables 3 and 4 show that twelve distinct fungus were found in all three kinds of gourd seeds: Ridge gourd, Bottle gourd, and Bitter gourd. Similar species of these fungi were detected in a variety of vegetable seeds; the most common was the bitter gourd, followed by the bottle and ridge gourds. Mycoflora was found on the glucose agar plates more so than on the blotter paper approach. Common fungus found included *Aspergillus niger, Aspergillus flavus, Fusarium oxysporum, Alternaria sp., Rhizopus, Curvularia, Chaetomium sp., Helminthosporium, Mucor, Penicillium,* and *Rhizopus stolonifer*. Fungi that predominated *included Fusarium oxysporum, Alternaria alternata, Aspergillus flavus,* and *Aspergillus niger*. Other scientists have also done comparable work; for instance, Survase (2012) assessed methi seeds and Telang (2010) separated mycoflora from Brinjal seeds.

These fungus carried by seeds were linked to seed degradation and reduced the percentage of seeds that germinate. Seeds of bottle gourds germinated less than those of bitter gourds and ridge gours.

As can be seen from Table 5, the leaf extracts of every medicinal plant that was examined both promoted seed germination and showed different degrees of inhibition against the incidence of seed mycoflora. At a concentration of 1:1 (13.3%, 16.7%, and 10% respectively showed the strongest inhibitory impact on seed mycoflora in Bitter gourd, Ridge gourd, and Bottle gourd seeds; *Ocimum sanctum* followed with 13.3% each. With *Calotropis gigantea*, the lowest inhibitory effect against seed mycoflora was seen at a concentration of 1:2 (23.3%, 16.7%, and 26.7%, respectively). Mansur and colleagues (2008) and Nwangburuta (2013) have also carried out comparable research.

There has been notable inhibitory action of *Azadirachta indica* on mycoflora spread by seeds. In 2008, Kadam *et al.* also looked at how well *Azadirachta indica* fought fungus on groundnut seeds. At a 1:1 concentration level, *Ocimum sanctum* exhibited the greatest seed germination rates in gourd seeds (90.0%, 86.7%, and 86.7%, respectively), followed by *Azadirachta indica* (86.7% each).

Additionally showing different degrees of inhibitory activity and boosting seed germination rates were *Adhatoda vasica, Calotropis gigantea,* and *Datura metal.*

Losses from microbial infections can reach 100% for cereals, vegetables, fruits, and agricultural produce both before and after harvest. Quality of seeds is greatly impacted by fungus associated with them (Satish *et al.,* 2007).

Thus, the necessity to find a substitute technique that will not be harmful to the consumer and will stop seed from bio-deteriorating during storage is critical. A more realistic and environmentally friendly approach to plant protection would be to exploit naturally occurring chemicals from plants that slow down the growth of microorganisms. This approach will play a major part in the creation of future crop protection strategies. The results of the current research will be considered as a crucial stage in selecting crop protection methods against the antifungal properties of diseases spread by seeds.

CONCLUSION

Azadiracta indica, Adhatoda vesica, Calotropis gigantea, Datara metal and *Ocimum sanctum* were demonstrated to have higher seed germination and to be highly effective in controlling seed-borne diseases.

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