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Evaluation of phytoextracts against seed mycoflora of Indian bean (*Lablab purpureus* L.) cultivars under *in vitro* condition

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ABSTRACT

In vitro evaluation of seven phytoextracts as seed dresser against seed mycoflora of Indian bean revealed custard apple leaf extract proved to be most effective with least number of fungal infected seeds followed by turmeric rhizome extract. *Neem* leaves extract also proved their potential against seed mycoflora.

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INTRODUCTION

Indian bean (*Lablab purpureus* L.) (Sweet) usually known as *Dolichos* bean, Hyacinth bean or Field bean is one of the most ancient crop among cultivated plants. It is a bushy, semi-erect, perennial herb, mainly cultivated either as a pure crop or mixed with finger millet, groundnut, castor, corn, Bajra or sorghum in Asia and Africa. It is a multipurpose crop grown for pulse, vegetable and forage purpose (Gowda, 2013).

In India, *Lablab* is a field crop mostly confined to the peninsular region and cultivated to a large extent in Karnataka and adjoining districts of Tamil Nadu, Andhra Pradesh and Maharashtra. Karnataka contributes a major share, accounting for nearly 90 per cent in terms of both area and production in the country. Outside India, the crop is cultivated in East Africa, with similar uses and in Australia as a fodder crop (Gowda, 2013).

Indian bean has been reported to suffer from various types of disease and majority of them are known to be caused by fungi which are seed borne in nature. Among them, anthracnose caused by *Colletotrichum* sp., blight caused by *Alternaria* sp., vascular wilt caused by *Fusarium* sp. and other molds like *Rhizopus* sp. and *Mucor* sp. are major seed borne mycoflora observed in Indian bean (Saxena and Kumari, 2017). Seed borne fungi are the most important plant pathogens that cause direct and indirect losses of the bean crop throughout the world (Schwartz and Galvez, 1980).

MATERIAL AND METHODS

Collection of seed samples:

Seeds of Indian bean cultivars collected from major Indian bean growing areas of Navsari as well as Surat district and also from the Pulse Research Station, Navsari Agricultural University, Navsari. There were five major varieties were collected named GV 1, GV 2,

GNIB 21, GNIB 22, VAL 125-26. The trial was laid out in Completely Randomized Design under laboratory condition.

Isolation of pathogens:

For isolation of seed mycoflora from Indian bean seeds two different seed health testing methods *viz.*, standard blotter method and agar plate method (ISTA, 1985) were used. Further isolated fungi were pure cultured and stored in PDA slants and stored under 5^{0}C for further use.

Preparation of phytoextracts:

Fresh and healthy plant parts *i.e.* leaves, bulbs, cloves and rhizomes of respective plants were collected and washed thoroughly with tap water and finally rinsed with sterile distilled water. Fifty grams of leaves, bulbs, cloves and rhizomes cut into small pieces and then macerated separately in sterile distilled water (1:1 w/v basis) by blender. Thus, prepared extracts of each were filtered through double layer sterilized muslin cloth to remove extraneous material and were considered 100% extract. Standard extracts were further diluted to the required concentration using sterile distilled water.

Management of seed mycoflora:

Seeds of Indian bean (400 as per standard method) were inoculated by soaking the seeds into mixed spore suspension of fungi followed by shade drying and then treated by soaking them into solution of various phytoextracts as mentioned in Table A. for 15-20 minutes. These treated seeds were evaluated by standard agar plate method and incubated at 25±2^{0}C for seven days. After ending of incubation period observations were recorded as percentage of infected seeds.

Table A : List of phytoextracts used for treatment

Sr. No.	Treatments	Dose ml/100g seeds
T_1	*Neem* leaf extracts (*Azadirachta indica*)	1.5ml
T_2	Garlic clove extracts (*Allium sativum*)	1.5ml
T_3	Dhatura leaf extracts (*Datura stramonium*)	1.5ml
T_4	Castor leaf extracts (*Ricinus communis*)	1.5ml
T_5	*Neem* oil (*Azadirachta indica*)	1.5ml
T_6	Turmeric rhizome extracts (*Curcuma longa*)	1.5ml
T_7	Custard apple leaf extracts (*Anona reticulate*)	1.5ml
T_8	Control (Without treatment)	-

RESULTS AND DISCUSSION

In vitro evaluation of seven phytoextracts (four leaf extracts *viz.*, castor, custard apple, datura, neem also garlic clove extract, turmeric rhizome extract and Neem oil) as seed treatment at their respective concentrations against seed mycoflora showed significant differences in per cent seeds showing mycoflora growth (Table 1). None of the treatments gave complete control of all

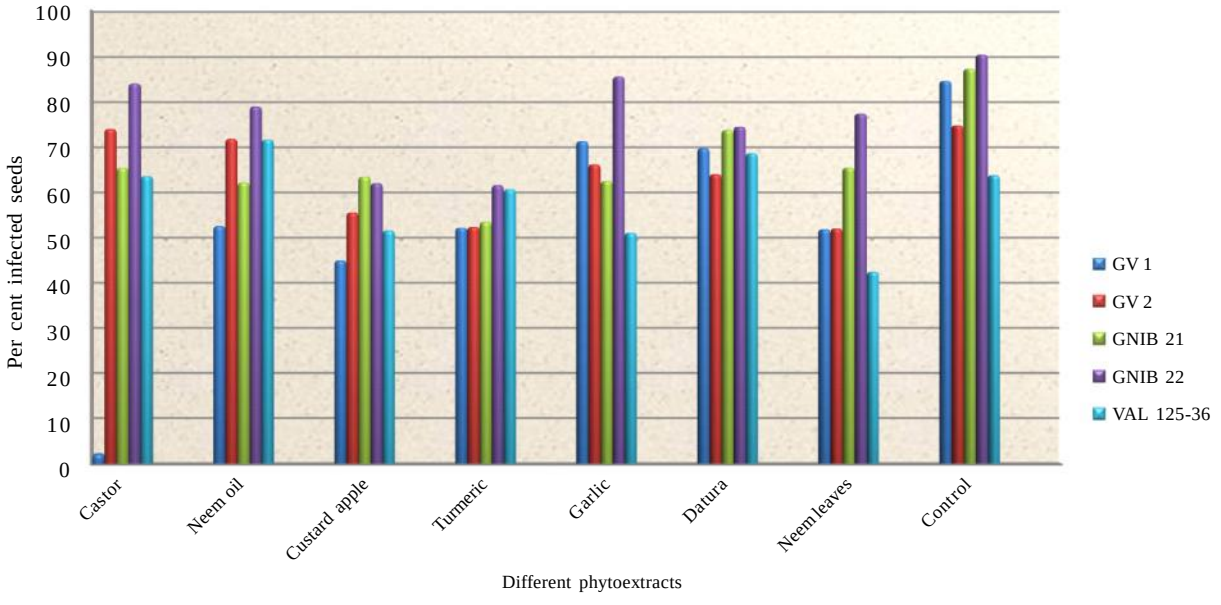

Fig. 1: Effect of phytoextracts on seed mycoflora

Table 1 : *In vitro* management of seed mycoflora of Indian bean using phytoextracts

Sr. No.	Treatments	Dose	Per cent infected seeds					Mean
			Cultivars					
			GV 1	GV 2	GNIB 21	GNIB 22	VAL 125-36	
1.	Castor	15ml	58.21*	59.26	53.95	66.38	52.83	58.13
			(2.27)	(73.92)	(65.39)	(83.93)	(63.53)	(57.81)
2.	Neem oil	15ml	46.44	57.90	52.00	62.65	57.78	55.35
			(52.55)	(71.81)	(62.14)	(78.92)	(71.62)	(67.41)
3.	Custard apple	15ml	42.04	48.16	52.77	51.93	45.87	48.15
			(44.89)	(55.53)	(63.43)	(62.01)	(51.56)	(55.48)
4.	Turmeric	15ml	46.23	46.33	46.96	51.65	51.11	48.46
			(52.18)	(52.35)	(53.46)	(61.53)	(60.63)	(56.03)
5.	Garlic	15ml	57.55	54.40	52.22	67.64	45.56	55.47
			(71.22)	(66.14)	(62.50)	(85.52)	(51.02)	(67.28)
6.	Datura	15ml	56.57	53.09	59.13	59.61	52.96	56.86
			(69.69)	(63.97)	(73.70)	(74.41)	(63.75)	(70.08)
7.	Neem leaves	15ml	46.00	46.11	53.97	61.52	40.62	49.64
			(51.78)	(51.97)	(65.43)	(77.29)	(42.43)	(57.78)
8.	Control	-	66.85	59.84	69.09	71.85	55.91	64.12
			(84.56)	(74.75)	(87.22)	(90.32)	(68.62)	(80.12)
	S.E.±		0.72	0.78	0.80	0.82	0.62	-
	C.D. (P=0.05)		2.18	2.37	2.44	2.49	1.87	-
	C.V%		2.38	2.56	2.54	2.31	2.13	-

*Figures outside parentheses are arcsine transformed values. Figures in parentheses are original values

fungi. However, custard apple leaf extract @ 15 per cent showed significantly minimum per cent seeds mycoflroa (48.15%) followed by turmeric rhizome extract @ 15 per cent (48.46%) and *Neem* leaf extracts (49.64%). Control recorded significantly highest (64.12%) per cent seeds showing mycoflora among all treatments. Significantly lowest per cent infected seeds found in VAL 125-36(40.62%) in the treatment of custard apple leaf extract and GV 1(42.04%) and GV 2(46.11%) in *Neem* leaf extract, respectively (Fig. 1).

Custard apple and turmeric extracts served as best treatments as the leaf extracts of custard apple have antifungal and antioxidant activities due to acetogenins present in leaves, which inhibit growth of the fungi. Whereas, the antifungal effect of turmeric includes fungal cell membrane disruption and inhibition of growth hormone (ergosterol) synthesis and respiration, which leads to death of fungi.

The present findings were more or less similar with the earlier findings of Kalidindi *et al.* (2015) that aqueous extract of custard apple were found to express inhibition against five different strains of fungi (*Alternaria alternata, Candida albicans, Fusarium solani, Microsporum canis* and *Aspergillus niger.*

Furthermore, Chen *et al.* (2018) found a strong inhibitory effect of turmeric extract on various pathogenic fungi *Fusarium graminearum, Fusarium chlamydosporum, Alternaria alternata, Fusarium tricinctum, Sclerotinia sclerotiorum, Botrytis cinerea, Fusarium culmorum, Rhizopus oryzae, Cladosporium cladosporioides, Fusarium oxysporum* and *Colletotrichum higginsianum,* which indicates a broad-spectrum antifungal effect of turmeric.

Chaudhari (2017) found the seed treatment with *Neem* seed phytoextracts gave more seed germination (78.67%) compared to control in pigeonpea.

Conclusion:

Seven phytoextracts were evaluated under *in vitro* condition as seed dresser against seed mycoflora where mean data indicated custard apple leaf extract @ 15 per cent revealed minimum (48.15%) seeds mycoflora followed by, Turmeric rhizome extract @ 15 per cent (48.46%). Control recorded significantly highest (64.12%) infected seeds.

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