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EVALUATION OF AQUEOUS EXTRACT OF SEEDS OF *PSORALEA CORYLIFOLIA* L. ON SEED MYCOFLORA, SEED GERMINATION AND SEEDLING VIGOUR OF MAIZE SEEDS.

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ABSTRACT

In vitro evaluation of seed germination and seedling vigour of maize seeds when tested at 3,6,12 and 24 hours of duration at 10,20,30,40 and 50% concentration of aqueous extract. At 12 hours of treatment, 20% concentration of aqueous extract showed high increase in seed germination (88.0%) and vigour index (1398.5). Significant activity was also observed in 6 hours of treatment compared to 3 and 24 hours of treatment. In pot experiment at 12 hours of treatment and at 20% concentration of aqueous extract, maximum and significant activity was observed in germination (90.0%), height(17.77cms), fresh weight(1.95 grams), dry weight(0.506 grams) and leaf area (16.27cms) compared to control. Moderate activity was observed in 3 and 6 hours of duration of treatment and least activity was observed in 24 hours of treatment.

Key words: *Psoralea corylifolia*, Aqueous extract, germination, vigour index.

INTRODUCTION

Nature has been a source of medicinal agents for thousands of years and an impressive number of modern drugs have isolated from natural sources (Vasu K and Singaracharya MA , 2008). Herbal drugs are still the mainstay of about 75-80% of the world population mainly in the developing countries for primary health care (Vasanthi S et al. 2008). Traditional medicine is an important source of potentially useful are a source of great economic value all over the world. Nature has bestowed on us a very rich botanical wealth and large number of diverse types of plants grow in different parts of the country. Plant products still remain the principal source of pharmaceutical agents used in traditional medicine (Prince L and Prabakaran P, 2011). Medicinal plants were used as excellent antimicrobial agents because it contain a variety of chemical constituent is nature. Recently much attention has directed towards extracts and biologically active compounds isolated from popular plant species. In recent years, secondary plant metabolites (Phytochemicals) previously with unknown pharmacological activities have been extensively investigated as source of medical agents (Prince L and Prabakaran P, 2011). Plants are used medicinally in different countries and are a source of many potent

and powerful drugs. Medicinal plants are used by 80% of the world population as the only available medicines especially in developing countries. Current research on natural molecules and products primarily focuses on plants since they can be sourced more easily and be selected based on their ethno-medicinal uses .A wide range of medicinal plants parts is used to extract as raw drugs and they possess varied medicinal properties (Dash BK et al. 2011). Many synthetic chemicals are widely used for the management of seed borne fungi which are both efficient and effective. Many of synthetic fungicides are known for their non-biodegradable nature and residual toxicity (Pak L, 2003). The ill effects associated with the use of chemical fungicides like carcinogenicity and teratogenicity which cause a serious health problem. There is an urgent need to search for alternative strategies for the management of pre and post harvest crop diseases. Medicinal plants are the local heritage with global importance and world is endowed with a rich wealth of medicinal plants (Suriyavathana M ,2010). In the present study, aqueous extract of seeds of *Psoralea corylifolia* L belongs to family Fabaceae were evaluated for their potency in seed germination and seedling vigour of maize.

MATERIALS AND METHODS

1.Test Plant

Shade dried, healthy seeds of *P. corylifolia* were collected from seed market, Mysore. The seeds were washed thoroughly 2-3 times with running tap water and once with sterile distilled water, air dried at room temperature on a sterile blotter, and used for preparation of extracts (Verma S and Dohroo NP, 2003).

1.1. Extraction

1.1.1. Aqueous extraction

One hundred grams of the thoroughly washed and air dried healthy seeds of *P. corylifolia* were macerated with 100ml sterile distilled water in a waring blender (Waring international, new hart-ford, CT, USA) for 5 min. The macerate was filtered through double-layered muslin cloth, and then centrifuged at 4000g for 30 minutes. The supernatant was filtered through Whatman No.1 filter paper and sterilized at 120^0 C for 10 minutes, which served as 100% aqueous mother extract. The extract was preserved aseptically in a sterile brown bottle at 5^0 C until further use (Verma S and Dohroo NP, 2003).

1.1.2. Seed treatment

Maize seeds (Local) were soaked in 10, 20, 30, 40 and 50% concentration of the aqueous seed extract for 3, 6, 12 and 24 hours duration. Seeds treated with sterile distilled water and soaked for 3, 6, 12 and 24 hours served as control

1.1.3. Seed germination and seedling vigour

Effect of aqueous seed extract of *P.corylifolia* on seed germination and seedling vigour of maize under laboratory condition was studied by treating the seeds and subjecting them to germination test and vigour index analysis. Seeds treated with extract and untreated seeds were subjected to germination test following the procedure of paper towel method (ISTA, 1999). Seedling vigour was determined at the end of 14 days of incubation following the method of Abdul Baki AA and Anderson JD (1973). The experiment was carried out with four replicates of 100 seeds each and repeated three times**.**

1.1.4. Pot experiments

The treated and untreated seeds of maize were sown in plastic pots of 10 cms diameter of 250 ml capacity, filled with autoclaved soil and sand in the ratio 2:1, each pot were sown with 5 seeds. They were maintained under greenhouse conditions at 25-30^0C and >95% relative humidity and watered regularly. No artificial fertilizer was provided to the plants. The setup was observed everyday for the growth parameters and recorded. Emergence data was recorded from the two days of sowing. Numbers of seeds germinated were counted on the 7^{th} day of sowing and the percentage germination was calculated. Later from each pot all the seedlings except one were removed and the setup was maintained upto 14 days. After 14^{th} days, the vegetative growth parameters viz., height, fresh weight, dry weight and leaf area were recorded. Height of the plant was measured from the base to the tip of the plant. Fresh weight was determined by uprotting the plant carefully, washing them

thoroughly to remove remnants of soil particles and weighing. Dry weight was determined by drying the plants in an oven at 65^{o} C until the weight remained constant. The leaf area was measured using the instrument ∇T area meter, MK2, Burwell, Cambridge, England (Kulshreshtha M and Khan MW, 2002).

Statistical analysis

The data were subjected to Tukey's HSD analysis. Data on percentages were transformed to arcsine and analysis of variance (Anova) was carried out with transformed values. The means were compared for significance using Tukey's HSD (P=0.05).

RESULT

Seed germination and seedling vigour

Total germination failure was observed at 6,12 and 24 hours of treatment with 50% concentration of the extract and at 24hours treatment with 40% concentration of the extract. Slight increase in seed germination and seedling vigour was observed in seeds treated with 10% concentration of the extract in all the duration of treatments tested. Highly significant increase in seed germination and seedling vigour was observed in the seeds treated with 20% concentration of the extract for the different durations tested. The seeding vigour increased with increased period of soaking in this concentration upto 12hours. No significant increase in seed germination or seedling vigour at 24hours treatment in this concentration over 12hours treatment. At 30% concentration treatment, marginal increase in seed germination and seedling vigour was observed at 3,6 and 12hours of treatment over control. However, total germination failure of seeds was observed in this concentration at 24hours treatment. At 40% concentration, significant decrease in seed germination and vigour index were observed in all the duration of treatment tested compared with control. At 24 hours period of soaking, total germination failure was observed in this concentration. At 50% concentration, highly significant reduction in seed germination and vigour index was observed at 3hours treatment, total germination failure was observed in this concentration at 6, 12 and 24hours treatment compared with control (Table 1).

Table 1 Effect of aqueous seed extract of P. corylifolia L. on seed germination and seedling vigour of aize

Duration of seed treatment (hours)	Concentration (%)	Germination (%)	Vigour index (MRL + MSL) x germination %
3 hours	10	74.00^{e} ±0.0	1017.5^{g}±0.0
	20	76.00^{g} ±.5	1029.8^{i}±0.3
	30	73.00^{d} ±0.1	1010.7^{d}±0.0
	40	71.00^{b} ±0.3	1004.6^{b}±0.1
	50	71.00^{b} ±0.3	1004.6^{b}±0.1
	Control	71.00^{b} ±0.2	1015.2^{f}±0.1
6 hours	10	74.00^{e} ±0.0	1036.0^{k}±0.0
	20	77.00^{h} ±0.5	1031.9^{j}±0.5
	30	72.00^{c} ±0.1	1029.6^{i}±0.1
	40	72.00^{c} ±0.5	1017.5^{g}±0.0
	50	0.0^{a} ±0.0	0.0^{a}±0.0
	Control	72.00^{c} ±0.2	1036.0^{k}±0.1
12 hours	10	75.00^{f} ±0.1	1008.5^{c}±0.2
	20	88.00^{i} ±0.2	1398.5^{l}±0.3
	30	74.00^{e}±1.0	1021.2^{h}±0.1
	40	73.00^{d}±0.5	1011.6^{e}±0.0
	50	0.0^{a}±0.0	0.0^{a}±0.0
	Control	72.00^{c}±0.2	1010.1^{d}±0.1
24 hours	10	75.00^{f}±0.1	1008.5^{c} ±0.2
	20	88.00^{i}±0.2	1398.5^{l}±0.3

	30	0.0^{a}± 1.0	0.0^{a}±1.0
	40	0.0^{a}±0.0	0.0^{a} ±0.0
	50	0.0^{a}±0.0	0.0^{a}±0.0
	Control	72.00^{c}±0.2	1010.1^{d}±0.1

- ***Values are the mean of three replicates, ± standard error.***
- ***The means followed by the same letter (S) are not significantly different at P<0.05 when subjected to Tukey's HSD.***

Pot experiments

Maize seeds treated with different concentrations of the aqueous extract for 3hours duration did not show any significant improvement in germination compared with control. Similarly no significant improvement in the other vegetative growth parameters like height, fresh weight, dry weight and leaf area were observed (Table 2).

The seeds treated with different concentrations of the aqueous extract of *P. corylifolia* for 6 hours duration recorded significant increase in germination at 20% concentration only. While the other concentration tested viz., 10, 30 and 40% did not show any significant improvement in germination. On the other hand at 50% concentration, total failure of germination was observed. The vegetative growth parameters like height, fresh weight, dry weight and leaf area also did not show any significant increase over the control except at 20% concentration. At 20% concentration, highly significant increase in germination and all the other growth parameters tested were significantly increased (Table 3).

Table 2 : Effect of aqueous extract of seeds of P. corylifolia on vegetative growth of maize seeds- 3 hours

Parameters	Aqueous Extract					
	Control (untreated)	**10%**	**20%**	**30%**	**40%**	**50%**
Germination (%)	55.00^{d}±0.0	54.50^{bc}±0.1	56.00^{e}±0.2	54.00^{b}±0.0	53.50^{a}±0.0	53.50^{a}±0.0
Height (cms)	11.23^{d}±0.5	11.24^{b}±0.5	11.28^{c}±0.5	11.24^{b}±0.0	11.24^{b}±0.0	11.24^{b}±0.0
Fresh weight (gms)	0.880^{b}±0.0	0.889^{c}±0.0	0.910^{e}±0.0	0.892^{d}±0.0	0.879^{a}±0.0	0.879^{a}±0.0
Dry weight (gms)	0.210^{a}±0.2	0.216^{b}±0.2	0.217^{b}±0.0	0.215^{b}±1.0	0.210^{a}±0.0	0.210^{a}±0.0
Leaf area (cm^2)	7.03^{a}±0.0	7.10^{b} ±0.2	7.25^{e}±0.1	7.15^{d}±0.0	7.11^{c}±0.0	7.11^{c}±0.0

- ***Values are the mean of three replicates, ± standard error.***
- ***The means followed by the same letter (S) are not significantly different at P<0.05 when subjected to Tukey's HSD.***

Table 3: Effect of aqueous extract of seeds of P. corylifolia on vegetative growth of maize seeds- 6 hours.

Parameters	Aqueous Extract					
	Control (untreated)	**10%**	**20%**	**30%**	**40%**	**50%**
Germination (%)	55.00^{c}±0.0	57.50^{d}±0.2	68.50^{e}±0.0	58.00^{b} ±1.0	56.00^{b}±0.0	0.0^{a}±0.0
Height (cms)	11.35^{b} ±0.1	11.38^{d}±0.1	12.39^{e}±0.2	11.37^{c}±0.5	11.37^{c}±0.5	0.0^{a}±0.0
Fresh weight (gms)	0.892^{b} ±1.0	0.900^{e} ±1.5	1.210^{f}±1.0	0.896^{d}±0.0	0.891^{b} ±0.0	0.0^{a}±0.0
Dry weight (gms)	0.223^{b}±0.0	0.228^{c}±0.0	0.315^{f} ±0.2	0.236^{e}±0.5	0.233^{d}±0.2	0.0^{a}±0.0
Leaf area (cm^2)	7.20^{b}±0.0	7.27^{c}±0.0	8.96^{e}±0.2	7.28^{d}±0.0	7.21^{c}±0.0	0.0^{a}±0.0

- ***Values are the mean of three replicates, ± standard error.***
- ***The means followed by the same letter (S) are not significantly different at P<0.05 when subjected to Tukey's HSD.***

Highly significant increase in germination and other vegetative growth parameters were observed at 10% and 20% concentration treatments. The germination percentage was highest (90%) at 20% concentration treatment with increasing concentration. Significant decrease in germination percentage was observed at

30% and 40% concentration treatment and total failure of germination was observed at 50% conc. Significant increase in vegetative growth parameters was not observed at 10, 30 an 40% compared with control. However highly significant increase in the vegetative growth parameters like Height (17.77cms) , fresh weight (1.95grams) , Dry weight (0.506grams) and leaf area (16.27cm^2) over control was observed at 20% concentration treatment (Table 4).

Table 4: Effect of aqueous extract of seeds of P. corylifolia on vegetative growth of maize seeds- 12 hours

Parameters	Aqueous Extract					
	Control (untreated)	**10%**	**20%**	**30%**	**40%**	**50%**
Germination (%)	58.00^{c}±0.0	66.00^{d}±0.1	90.00^{e}±0.1	44.00^{b}±0.0	44.00^{b}±0.0	0.0^{a}±0.0
Height (cms)	11.53^{c}±0.2	11.58^{d}±0.5	17.77^{e}±0.5	11.58^{d}±0.5	11.30^{b}±0.1	0.0^{a}±0.0
Fresh weight (gms)	0.92^{e}±1.0	1.44^{b}±1.0	1.95^{f}±1.0	0.91^{d}±0.0	0.89^{c}±0.0	0.0^{a}±0.0
Dry weight (gms)	0.226^{b} ±0.0	0.381^{e} ±0.0	0.506^{f}±0.0	0.275^{d} ±0.0	0.243^{c}±0.1	0.0^{a}±0.0
Leaf area (cm^2)	7.58^{b}±0.0	9.96^{d}±0.0	16.27^{e} ±0.0	8.02^{c}±0.0	8.02^{c}±0.0	0.0^{a}±0.0

- ***Values are the mean of three replicates, ± standard error.***
- ***The means followed by the same letter (S) are not significantly different at P<0.05 when subjected to Tukey's HSD.***

Marginal increase in germination and different vegetative growth parameters were observed at 10% concentration treatment. Highly significant increase in germination and other vegetative parameters were observed at 20% concentration. At 30% concentration, significant decrease in germination without much variation in vegetative growth parameters was observed. At 40% and 50% concentration, total germination failure was observed (Table 5).

Table 5: Effect of aqueous extract of seeds of P. corylifolia on vegetative growth of maize seeds - 24 hours

Parameters	Aqueous Extract					
	Control (untreated)	**10%**	**20%**	**30%**	**40%**	**50%**
Germination (%)	58.00^{c}±0.0	66.00^{d}±0.1	90.00^{e}±0.1	44.00^{b}±0.0	0.0^{a}±0.0	0.0^{a}±0.0
Height (cms)	11.53^{b} ±0.2	11.58^{c}±0.5	17.77^{d}±0.5	11.58^{c}±0.5	0.0^{a}±0.0	0.0^{a}±0.0
Fresh weight (gms)	0.92^{c}±1.0	1.44^{d}±1.0	1.95^{e} ±1.0	0.91^{b}±0.0	0.0^{a}±0.0	0.0^{a}±0.0
Dry weight (gms)	0.226^{b}±0.0	0.381^{d}±0.0	0.506^{e}±0.0	0.275^{c}±0.0	0.0^{a}±0.0	0.0^{a}±0.0
Leaf area (cm^2)	7.58^{b}±0.0	9.96^{d}±0.0	16.27^{e} ±0.0	8.02^{c}±0.0	0.0^{a}±0.0	0.0^{a}±0.0

- ***Values are the mean of three replicates, ± standard error.***
- ***The means followed by the same letter (S) are not significantly different at P<0.05 when subjected to Tukey's HSD.***

DISCUSSION

Further investigations conducted on the germination and vegetative growth parameters of seeds treated with different concentrations of aqueous extract of *P. corylifolia* for 3,6,12 and 24 hours durations also reveals that 20% concentration of the aqueous extract promoted germination and vegetative growth of maize seeds. Highly significant increase in vegetative growth parameters have been observed in seeds treated with 20% concentration of the aqueous extract for 6hrs duration suggesting that treatment of maize seeds with 20% concentration of the aqueous extract for 6hours is the most ideal treatment for growth promotion, germination increase and significant decrease of seed mycoflora. The results of the present investigations also suggests that 50% concentration of the aqueous extract is highly phytotoxic. Thus based on the results of the present investigations, it is recommended that 20% concentration of the aqueous

extract treatment of maize seeds for 6hours duration could be exploited as an important ecofriendly approach for the management of seed borne fungi of the maize seeds in particular and other seeds in general.

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