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EFFECT OF *Azadirachta indica*- LEAVES, *Allium sativum*- BULBS AND *Ocimum sanctum*- LEAVES ON THE GERMINATION OF MAIZE SEEDS

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ABSTRACT

The present study evaluated the effect of aqueous and ethanolic extracts of leaves of *A.indica,* bulbs of *A. Sativum* and leaves of *O. sanctum* for maize seed germination at 2.0, 4.0, 6.0, 8.0 and 10% concentration for 12, 24, and 48 hours of treatment. Maximum and significant activity was observed in ethanolic extract of *A. Indica* at 6.0 % concentration at 24 hours of duration and the maximum germination percentage was recorded as 90.15 ± 0.2 followed by aqueous extract of *A. Indica (*90.02 ± 0.2) and than by ethanolic extract of *A. Sativum (*78.15 ± 0.2) . The lowest maize seed germination (60.45 ± 0.1) was observed in aqueous extract of *O. Sanctum* at 10 % concentration during 12 hours of treatment. The concentration (10%) at duration of treatment (24 and 48 hours) resulted in total germination failure suggesting that the higher concentrations with longer duration of treatment are highly phytotoxic to maize seeds. This phytotoxicity may be due to organic acids, chemical decomposition or microbial degradation of organic compounds.

KEYWORDS: *A. indica*, *A. sativum, O. Sanctum,* Phytotoxic and Seed Germination.

Maize (*Zea mays* L.) is one of the main sources cereals for food, forage and processed industrial products. World production of maize is around 790 million tones it serves as a staple food providing more than one-third of the calories and proteins in some countries. Maize Seed is living entity and is subjected to various environmental stresses which affect the quality. Despite the high yielding potential and various advantages of maize, the yield per unit area of the crop is low in India. Delay in germination and low seed viability is the serious problems limiting the production of maize. Highly vigorous seeds germinate rapidly, uniformly and are able to withstand environmental adversity after sowing. However, the use of maize seeds of low physiological quality is a common practice under tropical and subtropical production conditions, leading to inadequate plant population in the field. So, there is an urgent need to search for alternative strategies for the management of pre and post sowed maize seed diseases. Nature has bestowed on us a very rich botanical wealth large numbers 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). 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). Plant metabolites and plant based pesticides appear to be one of the better alternatives as are known to have minimal environmental impact and danger to consumers in contrast to the synthetic pesticides. Plant-based biopesticides (botanicals) and their metabolites are now being extensively used as a component of IPM (Padmaja, 2005). Control measures by using plant extracts are cost effective and non-toxic methods. In the present study, aqueous and ethanolic extract of *A. Indica*- leaves, *A. Sativum*- bulbs and *O. Sanctum*- leaves were evaluated for their potency in maize seed germination.

MATERIALS AND METHODS

Experimental Seeds

Maize seeds were used for the present investigation.

Experimental Plant Product Used For Extract

Following plant products were used for extract preparation:

Azadirachta indica- Leaves

Allium sativum - Bulbs

Ocimum sanctum- Leaves

Collection of Plant Materials

Selected plant materials i.e. leaves of *Azadirachta indica* and *Ocimum sanctum* were collected from the botanical garden of Govt. Holkar science college in poly bags and brought to lab and their botanical identity was established and *Allium sativum* was brought market and it was also identified at the department of botany, Govt.

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Holkar science college, Indore (M.P.). The collected material was washed, shade dried under room temperature (27 ± 2^0C) and *Azadirachta indica* and *Ocimum sanctum* was powdered using electric blender and the paste of *Allium sativum* was made which were further processed for phytochemical analysis.

Soxhlet Extraction

The ordinary method of extraction was not efficient to yield good amount of active principle of the plant material. To extract more active principle from all the plant materials, Soxhlet extraction given by Sharma and Gupta (2009) was used. The plant extract were prepared in two solvent one was distilled water and second was ethanol.

Phytochemical Screening

Phytochemical screening was done in order to detect the presence of bioactive constituents such as alkaloids, tannins, saponins, phenols, flavonoids and glycosides using the methods described by Sofowora (1978), Trease and Evans (1989).

Maize seed germination effect (Duangpatra, 1986)

Randomized block design was used for the present investigation with three replications of treatments.The germination test was performed by the use of between-paper method (rolled method).

Germination percentage was calculated as described in the following Formula **(Patel, 2001)**.

$$GP = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds kept}} \times 100$$

All data thus obtained was tabulated and graphically presented as per the required statistical methods. The data was analyzed using statistical software.

RESULTS

10 % concentration of both aqueous and ethanolic extracts of *A. indica* at 24 and 48 hours of duration showed the complete failure (00.00 ± 0.0) of maize seed germination. The highest seed germination effect was shown by ethanolic extract at 6.0 % concentration (90.15 ± 0.2) followed by aqueous extract at 6.0 % concentration (90.02 ± 0.2) both at 24 hours of treatment and than by ethanolic extract at 6.0 % concentration (88.97 ± 0.2) at 48 hours of treatment and the least effect was shown by aqueous extract at 10 % concentration (64.00 ± 0.0) at 12 hours of duration. The result showed that there was increase in maize seed germination percentage from 2.0 % - 6.0 % concentration but 8.0 % concentration showed gradual decrease in maize seed germination percentage. The result showed that higher concentrations with longer duration of treatment are highly phytotoxic to maize seeds.

10 % concentration of both aqueous and ethanolic extracts of *A. sativum* at 24 and 48 hours of duration showed the complete failure (00.00 ± 0.0) of maize seed germination. The highest seed germination effect was shown by ethanolic extract at 6.0 % concentration (78.15 ± 0.2) followed by aqueous extract at 6.0 % concentration (77.65 ± 0.2) both at 24 hours of treatment and than by ethanolic extract at 6.0 % concentration (76.80 ± 0.2) at 48 hours of treatment and the least effect was shown by aqueous extract at 10 % concentration (63.33 ± 0.0) at 12 hours of duration. The result showed that there was increase in maize seed germination percentage from 2.0 % - 6.0 % concentration but 8.0 % concentration showed gradual decrease in maize seed germination percentage. The result showed that higher concentrations with longer duration of treatment are highly phytotoxic to maize seeds.

10 % concentration of both aqueous and ethanolic extracts of *O. sanctum* at 24 and 48 hours of duration showed the complete failure (00.00 ± 0.0) of maize seed germination. The highest seed germination effect was shown by ethanolic extract at 6.0 % concentration (74.93 ± 0.2) followed by aqueous extract at 6.0 % concentration (73.56 ± 0.2) both at 24 hours of treatment and than by ethanolic extract at 6.0 % concentration (72.92 ± 0.0) at 48 hours of treatment and the least effect was shown by aqueous extract at 10 % concentration (60.45 ± 0.1) at 12 hours of duration. The result showed that there was increase in maize seed germination percentage from 2.0 % - 6.0 % concentration but 8.0 % concentration showed gradual decrease in maize seed germination percentage. The result showed that higher concentrations with longer duration of treatment are highly phytotoxic to maize seeds.

The control value for each plant extract at different concentrations with respect to different time duration was 21.33± 0.2.

Evaluation of different concentration of different plant extract on seed germination by between-paper method (rolled method) revealed that 10 % concentration at 24 and 48 hours of treatment resulted in complete failure in seed germination suggesting that higher concentrations with longer duration of treatment is highly phytotoxic to

maize seeds. This is evident from the fact that all plants extract at 24 hours of treatment at 6.0 % concentrations resulted in highest seed germination. In view of the above result, the ethanolic extract of *A. indica* at 6.0 % concentration resulted in highest seed germination effect at 24 hours of treatment followed by *A. sativum* and the least effect was shown by *O. Sanctum.*

Table 1: Effect of aqueous and ethanolic extracts of *A. indica* on maize seed germination.

Duration of seed Treatment (hours)	Concentration (%)	Seed Germination (%) in Aqueous extract	Seed Germination (%) in Ethanolic extract
12 hours	2.0	68.66 ± 0.1	69.23 ± 0.0
	4.0	70.00 ± 0.3	72.10 ± 0.2
	6.0	75.06 ± 0.2	75.11 ± 0.2
	8.0	70.33 ± 0.0	71.86 ± 0.0
	10	64.00 ± 0.0	64.09 ± 0.3
	Control	21.33 ± 0.2	21.33 ± 0.2
24 hours	2.0	72.12 ± 0.5	74.07 ± 0.5
	4.0	75.09 ± 0.0	76.00 ± 0.0
	6.0	90.02 ± 0.2	90.15 ± 0.2
	8.0	78.23 ± 0.3	80.15 ± 0.2
	10	0.0 ± 0.0	0.0 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2
48 hours	2.0	72.68 ± 0.0	75.88 ± 0.2
	4.0	77.36 ± 0.1	79.06 ± 0.0
	6.0	88.12 ±0.2	88.97 ±0.2
	8.0	82.00 ± 0.3	86.00 ± 0.0
	10	0.0 ± 0.0	0.0 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2

Values are the mean of three replicates, ± standard error

Table 2: Effect of aqueous and ethanolic extracts of *A. sativum* on maize seed germination.

Duration of seed Treatment (hours)	Concentration (%)	Seed Germination (%) in Aqueous extract	Seed Germination (%) in Ethanolic extract
12 hours	2.0	65.54 ± 0.5	66.06 ± 0.3
	4.0	66.70 ± 0.0	68.00 ± 0.0
	6.0	74.98 ± 0.1	75.04 ± 0.0
	8.0	71.03 ± 0.2	72.63 ± 0.2
	10	63.33 ± 0.0	64.92 ± 0.1
	Control	21.33 ± 0.2	21.33 ± 0.2
24 hours	2.0	70.08 ± 0.0	71.20 ± 0.0
	4.0	71.37 ± 0.1	75.00 ± 0.0
	6.0	77.65 ±0.2	78.15 ± 0.2
	8.0	74.70 ± 0.3	73.31 ± 0.0
	10	0.0 ± 0.0	0.0 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2
48 hours	2.0	68.22 ± 0.0	69.19 ± 0.2
	4.0	71.00 ± 0.1	72.14 ± 0.0
	6.0	75.63 ± 0.2	76.80 ± 0.2
	8.0	73.28 ± 0.5	73.89 ± 0.5
	10	0.0 ± 0.0	0.0 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2

Values are the mean of three replicates, ± standard error.

Table 3: Effect of aqueous and ethanolic extracts of *O. sanctum* on maize seed germination.

Duration of seed Treatment (hours)	Concentration (%)	Seed Germination (%) in Aqueous extract	Seed Germination (%) in Ethanolic extract
12 hours	2.0	64.40 ± 0.1	66.00 ± 0.2
	4.0	65.53 ± 0.2	66.84 ± 0.0
	6.0	67.24 ± 0.0	68.61 ± 0.1
	8.0	66.19 ± 0.2	67.00 ± 0.2
	10	60.45 ± 0.1	63.88 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2
24 hours	2.0	65.88 ± 0.3	67.09 ± 0.0
	4.0	67.17 ± 0.0	68.00 ± 0.5
	6.0	73.56 ± 0.2	74.93 ± 0.2
	8.0	69.06 ± 0.1	70.68 ± 0.1
	10	0.0 ± 0.0	0.0 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2
48 hours	2.0	67.83 ± 0.0	68.22 ± 0.2
	4.0	69.32 ± 0.2	70.96 ± 0.1
	6.0	70.66 ± 0.1	72.92 ± 0.0
	8.0	69.44 ± 0.2	71.13 ± 0.2
	10	0.0 ± 0.0	0.0 ± 0.0
	Control	21.33 ± 0.2	21.33 ± 0.2

Values are the mean of three replicates, ± standard error.

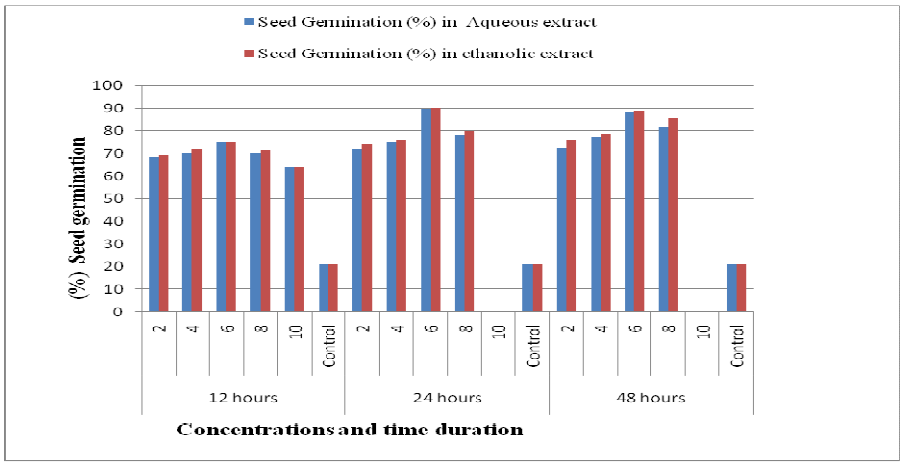

Figure 1: Effect of aqueous and ethanolic extracts of *A. indica* on maize seed germination.

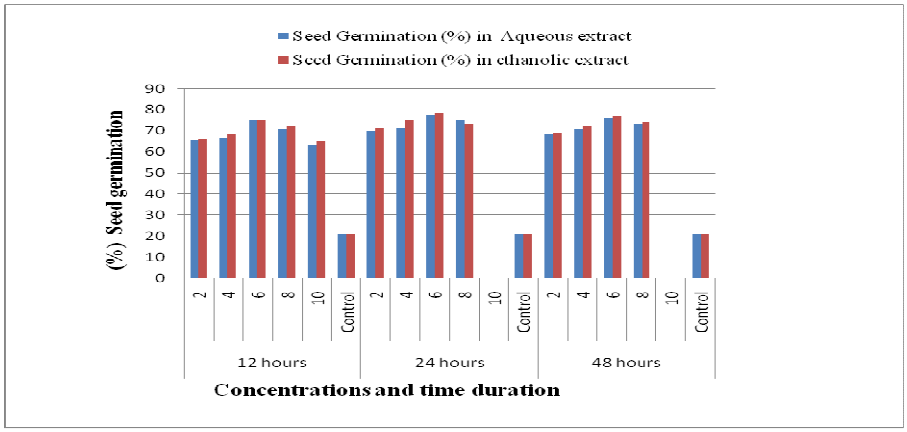

Figure 2: Effect of aqueous and ethanolic extracts of *A. sativum* on maize seed germination.

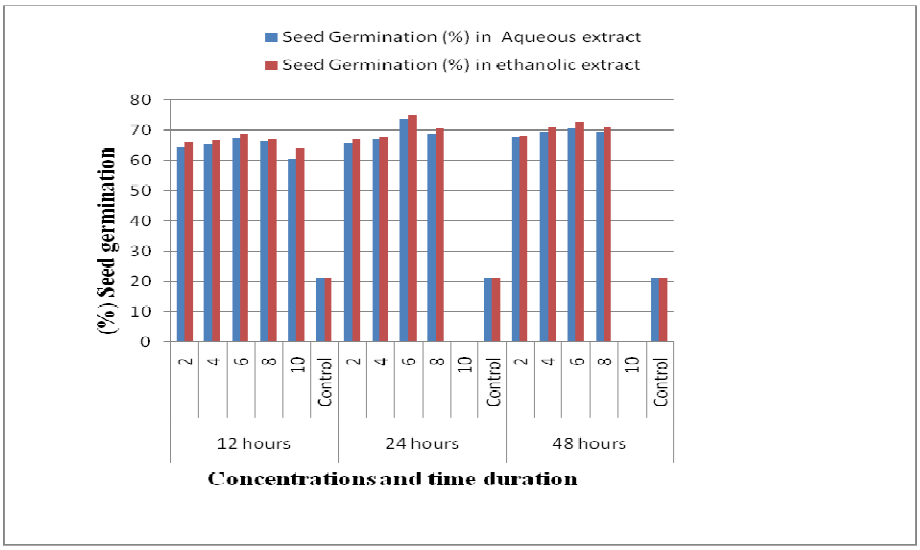

Figure 3: Effect of aqueous and ethanolic extracts of *O. sanctum* on maize seed germination.

DISCUSSION

Evaluation of different concentration of different plant extract on maize seed germination by between-paper method (rolled method) revealed that 10 % concentration at 24 and 48 hours of treatment resulted in zero seed germination suggesting that higher concentrations with longer duration of treatment is highly phytotoxic to maize seeds. This phytotoxicity may be due to organic acids, chemical decomposition or microbial degradation of organic compounds. This is evident from the fact that all plants extract at 24 hours of treatment at 6.0 % concentrations resulted in highest seed germination. In view of the above result, the ethanolic extract of *A. indica* at 6.0 % concentration resulted in highest seed germination effect at 24 hours of treatment followed by *A. sativum* and the least effect was shown by *O. Sanctum.*

Kiran *et al.* (2012) found highly significant increase in vegetative growth parameters in seeds treated with 20% concentration of the aqueous extract of *Psoralea corylifolia* for 6 hrs duration suggesting that treatment of maize seeds with 20% concentration of the aqueous extract for 6 hours is the most ideal treatment for growth promotion, germination increase and significant decrease of seed mycoflora. They also suggested that 50% concentration of the aqueous extract is highly phytotoxic causing 100 % seed germination failure. They suggested that longer duration and high concentration cause the total germination failure. So, this result is in agreement with the present investigation.

Ahmed (2002) was also in agreement with the present investigation that garlic extracts as more effective against *Bipolaris oryzae* at 1:1 dilution. Extracts of different plant species may contain phytotoxic compounds. The interactions of crop species with extracts indicate that phytotoxic effects may be due to more than one chemical component present in the different extracts and the crop species that reacted differently to these compounds. Inhibition might have been due to the presence of allelochemicals as reported by Chaturved (1969). Swaminathan *et al.* (1989) reported that the potential compounds which are able to induce inhibitory effect and seed germination are identified as phenolic acids (Rejila *et al.*, 2012).

Verma *et al.* (2012) and Lawan *et al.* (2011) also reported the same result that Phytotoxic effects of sweet basil (*Ocimum basilicum* L.) extracts on germination and seedling growth of commercial crop plants. They also found that *Ocimum sanctum* showed low grain germination effect. They also reported that different concentrations of leaf extract inhibited the germination of the seeds to a certain extent which in some cases found to cause complete inhibition of germination of the seeds. Overall growth rate of seedlings was also reduced in almost all the treatments compared with control. So, it was observed that the inhibition of seed germination and seedling growth is dependent on the concentration i.e. inhibition was more as the concentration increased. These findings coincided with the reports of Einhelling (1996), Daniel (1999), Lawan *et al.* (2010) and Sazada *et al.* (2009) who reported that allelopathy is concentration dependent phenomenon.

The present findings corroborate the earliest report by (Bora *et al.,* 1999), who found that the inhibitory effect of leaf extracts of *Acacia auriculiformis* on germination of some agricultural crops was proportional to the concentration of the extract. Also noted by (Jadhar and Gayanar, 1992), percentage of germination, plumule and radicle length of rice and cowpea decreases with the increasing concentration of *Acacia auriculiformis* leaf extract.

The present findings are also in agreement with Rajendra *et al.* (2014) who also reported the Efficacy of Plant Extracts on the Germination of Wheat Seeds. The interaction effects of different doses and different plant extracts showed that the average germination was the highest and the lowest dose level. Germination of wheat seeds decreases gradually with the increase of doses. The present findings are almost in agreement with those of Islam (2001), Khaire *et al.* (1992) and Gupta *et al.* (1988), where they reported that seeds treated with plant materials did not adversely affect the seed germination. Therefore, the results of present investigation are supported by above mentioned authors.

In the present investigation, it is now clear that as the concentration and duration of treatment increases the seed fails to germinate. Thus, based on the result of present investigation, it is recommended that *A. Indica* followed by *A. Sativum* and than *O. Sanctum* at 6.0 % concentration with the duration of 12, 24 and 48 hours are effective for the maize seed germination as it showed the highest seed germination effect. The result of the present investigation also suggests that 10% concentration of the aqueous and ethanolic plant extracts are highly phytotoxic at 24 and 48 hours of treatment. Thus Farmers may use these plant extracts in their storage structure for management of stored grain pests without any adverse effect on germination of treated seeds.

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