

ISSN : 1812-5379 (Print)
ISSN : 1812-5417 (Online)
<http://ansijournals.com/ja>

JOURNAL OF AGRONOMY



ANSI*net*

Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

**Allelopathic Effect of Different Concentration of Water Extracts of
Acacia auriculiformis Leaf on Some Initial Growth Parameters of
Five Common Agricultural Crops**

A.T.M. Rafiqul Hoque, R. Ahmed, M.B. Uddin and M.K. Hossain
Institute of Forestry and Environmental Sciences,
University of Chittagong, Chittagong-4331, Bangladesh

Abstract: Allelopathic effect of leaf extracts (different concentration) of *Acacia auriculiformis* and its possible phytotoxicity was tested in a laboratory experiment by using some agricultural crops e.g. *Brassica juncea* (L.) Czern & Coss; *Phaseolus mungo* L.; *Raphanus sativus* L. and *Vigna unguiculata* (L.) Walp. and *Cicer arietinum* L. as a bioassay material. The experiment was conducted in sterilized petri dishes with a photoperiod of 24 hours on an average of 29°C. The effects of the different concentrations of aqueous extracts were compared to distill water (control.). The aqueous extracts caused significant inhibitory effect on germination, root and shoot elongation and development of lateral roots of receptor plants. Bioassays indicated that the inhibitory effect was proportional to the concentrations of the extracts and higher concentration (50-100%) had the stronger inhibitory effect whereas in some cases the lower concentration (10-25%) showed stimulatory effect. The study also revealed that inhibitory effect was much pronounced in root and lateral root development rather than germination and shoot growth.

Key words: Allelopathic effect, *Acacia auriculiformis*, germination, root length and shoot length

Introduction

Acacia auriculiformis, a leguminous species (Mimosoideae), naturally distributed in Australia, Papua New Guinea and Indonesia (Pinoyopusarerk, 1990), introduced in Bangladesh for afforestation and reforestation of degraded and wasteland areas (Rahman, 1984 and Das, 1985). Although the species is fast growing and can grow in wide ranges of soils (Davidson, 1985) the ground vegetation under its canopy indicated that it has some allelopathic potentials which might have been caused either by fallen leaves (through decomposition of litter) or plant leachates or root exudates. Consequently, the release of allelochemicals (organic substances) into the soil inhibits seed germination and establishment of agricultural crops and vegetation (Rice, 1979). Such inhibitory effects on the under storey vegetation have been exhibited in Pines (Rice, 1974). Joyakumar *et al.* (1978a, 1978b), Swami Rao and Reddy (1984), Melkania (1987), Eyini *et al.* (1989), Koul *et al.* (1991) and Bansal *et al.* (1992) studied the inhibitory effect of *Eucalyptus*, Bamboo, Teak, *Acacia nilotica*, *Dalbergia sissoo*, *Bauhinia variegata*, *Ficus bengalensis*, *Morus alba*,

Populus deltoides, *Salix babylonica* and *Leucaena leucocephala* on the germination and seedling growth of certain food crops. Petmark and Williams (1991) recommended *Acacia auriculiformis* as agroforestry species. King (1979) pointed out the need for investigations of allelopathy in various tree species used in agroforestry where there is a good chance of allelochemicals release by the intercrop trees affecting food and fodder crops. Therefore, it seems essential that the allelopathic compatibility of crops with trees should be checked before introducing in agroforestry system (Gaba, 1987; Khan and Alam, 1996). Very few researches were done on this aspect in case of *Acacia auriculiformis* (Jadhav and Gaynar, 1992; Rao *et al.*, 1994; Bora *et al.*, 1999) in this sub-continent. So the purpose of the present study was to elucidate the allelopathic potential of different concentration of *Acacia auriculiformis* leaf extracts on some common agricultural crops used in Bangladesh.

Materials and Methods

Acacia auriculiformis was considered as the donor plant and the receptor agricultural crops selected were Indian mustard (*Brassica juncea* (L.) Czern & Coss), Chickpea (*Cicer arietinum* L.), Black gram (*Phaseolus mungo* L.), Radish (*Raphanus sativus* L.) and Falen (*Vigna unguiculata* (L.) Walp).

The aqueous extracts of were prepared from fresh leaf of the donor plant. 100 gram of fresh senescent leaves of each species were soaked in 500 ml of distill water and kept at room temperature. After 24 hours the aqueous extract was filtered through the sieve. Some extracts were diluted to make the concentration of 10%, 25%, 50% and 75% and stored for seed treatment experiments. The following treatments were used in the experiment:

- T₀ = Seeds of receptor plants grown in distill water only (Control),
- T₁ = Seeds of receptor plants grown in leaf extracts of 10% concentration
- T₂ = Seeds of receptor plants grown in leaf extracts of 25% concentration
- T₃ = Seeds of receptor plants grown in leaf extracts of 50% concentration
- T₄ = Seeds of receptor plants grown in leaf extracts of 75% concentration
- T₅ = Seeds of receptor plants grown in leaf extracts of 100% concentration

Germination and growth records

The germination test was carried out in sterile petri dishes of 12 cm in size placing a Whatman no.3 filter paper on petri dishes. The extract of each concentration was added to each Petridish of respective treatment daily in such an amount just to wet the seed. The control was treated with distill water only. 20 seeds of an agricultural crop were placed in each Petridish replicating five times. The petri dishes were set in the analytical laboratory of the Institute of Forestry and Environmental Sciences, Chittagong University, Bangladesh at an average room temperature of 29°C. The experiment extended over a period of ten days to allow the last seed germination and the measurement of the shoot and root length. The seed was considered as germinated when the radicle emerged and the germination was recorded daily. The results were determined by counting the number of germinated seeds, number of lateral roots and measuring the length of

primary root and main shoot on 10th day of the experiment. The data were subjected to Analysis of Variance and Duncan's Multiple Range Test (DMRT).

Ratio of germination and elongation were calculated as suggested by Rho and Kil (1986):

$$\text{Relative Germination Ratio (RGR)} = \frac{\text{Germination ratio of tested plant}}{\text{Germination ratio of control}} \times 100$$

$$\text{Relative Elongation Ratio (RER) of shoot} = \frac{\text{Mean shoot length of tested plant}}{\text{Mean shoot length of control}} \times 100$$

$$\text{Relative Elongation Ratio (RER) of root} = \frac{\text{Mean root length of tested plant}}{\text{Mean root length of control}} \times 100$$

For the calculation of percentage of inhibitory effect on the germination and growth parameter to the control was calculated as per formula evolved by Surendra and Pota, (1978):

$$I = 100 - \frac{E_2 \times 100}{E_1}$$

Where: I = % inhibition, E₁ = Response of control plant, E₂ = Response of treatment plant

Results

Germination

Table 1 shows the germination percentage of the five receptor plants. The study revealed that the inhibitory effect increases with the increase of extract concentration. In all cases the most inhibitory effect was found at T₅ treatment (100% conc.). The highest inhibitory effect (-90.39%) was found in *C. arietinum* at T₅ treatment while the lowest was (-1.76%) found in *B. juncea* and *P. mungo* at T₃ treatment. The stimulatory effects were mostly observed at T₁ (10% conc.) treatment. This result supports the findings of Rice (1984) who inferred that chemicals that inhibit the growth of some species at certain concentrations could stimulate the growth of the same or different species at lower concentrations. Significant inhibition of seed germination was not observed in *P. mungo* and *B. juncea* upto 50% concentration. Maximum relative germination (RGR) ratio (105.26%) was observed in *P. mungo* at T₄ treatment while the minimum (9.61%) was found in *C. arietinum* at T₅ treatment (Fig.1).

Growth of seedlings

Shoot elongation (cm)

The shoot lengths of five bioassay species are shown in Table 2. It was observed that in most cases the stimulatory effect was found at T₁ and T₂ treatment. The inhibitory effect was much more pronounced at T₅ treatment followed by T₄, T₃ and T₂ treatment respectively. The highest inhibitory effect (-41.56%) was found on *B. juncea* at T₅ treatment while the lowest (-1.20%) was

Table 1: Germination percent of receptor agricultural crops to distill water (T₀) and different concentrations of *Acacia auriculiformis* extracts (T₁-T₅)

Treatment	Agricultural crops				
	<i>C. arietinum</i>	<i>R. sativus</i>	<i>V. unguiculata</i>	<i>B. juncea</i>	<i>P. mungo</i>
T ₀	86.67a*	83.33 a	95.00a	95.00a	95.00a
T ₁	90.00a (+3.84)	91.67 a (+10.00)	91.67a (-3.51)	95.00a (0.00)	98.00a (+3.16)
T ₂	81.67a (-5.77)	80.00 a (-3.99)	95.00a (0.00)	98.33a (+3.50)	98.33a (+3.51)
T ₃	35.00c (-59.62)	71.67ab (-13.99)	85.00ab (-10.52)	93.33a (-1.76)	93.33a (-1.76)
T ₄	50.00b (-42.31)	73.33ab (-12.00)	83.33ab (-12.28)	85.00a (-10.52)	100.00a (+5.26)
T ₅	8.33d (-90.39)	50.00b (-39.99)	76.67b (-19.29)	56.67b (-40.35)	85.00b (-10.52)

Table 2: Shoot elongation (cm) of receptor agricultural crops to distill water (T₀) and different concentrations of *Acacia auriculiformis* extracts (T₁-T₅)

Treatment	Agricultural crops				
	<i>C. arietinum</i>	<i>R. sativus</i>	<i>V. unguiculata</i>	<i>B. juncea</i>	<i>P. mungo</i>
T ₀	4.98ab*	7.51b	13.94a	3.97ab	18.28a
T ₁	6.13a (+23.09)	9.29a (+23.70)	13.22ab (-5.16)	5.1 a (+28.46)	17.63ab (-3.56)
T ₂	5.25ab (+5.42)	9.82b (+30.76)	12.65ab (-9.25)	4.24ab (+6.80)	15.86bc (-13.24)
T ₃	4.92ab (-1.20)	9.41a (+25.30)	12.67ab (-9.11)	3.97ab (0.00)	16.45ab (-10.01)
T ₄	4.57bc (-8.23)	9.09ab (+21.04)	11.35b (-18.58)	3.56bc (-10.33)	14.91c (-18.43)
T ₅	3.33c (-33.13)	4.43c (-41.01)	8.19c (-41.25)	2.32c (-41.56)	12.20d (-33.26)

Values in the parenthesis indicates the inhibitory (-) or stimulatory (+) effects in comparison to control (T₀)

* Values in the columns followed by the same letter(s) are not significantly different (P_≤0.05) according to Duncan's Multiple Range Test (DMRT)

in *C. arietinum* at T₃ treatment. Maximum elongation of shoot (18.28 cm) was observed in *P. mungo* followed by (13.94 cm) in *V. unguiculata* both at control. Maximum Relative Elongation Ratio (RER) of shoot (130.76%) was observed in *R. sativus* at T₂ treatment while among the survivors the minimum (58.43%) was in *B. juncea* at T₅ treatment (Fig. 2).

Root elongation (cm)

The mean root lengths (cm) as well as the percentage of inhibition of root elongation of all the receptor agricultural crops are shown in Table 3. The study revealed that in all cases the root length of the crops were greatly inhibited with the increasing extract concentration except *P. mungo* T₁ and T₂ treatment. Statistically pronounced significant inhibitory effect

Table 3: Root elongation (cm) of receptor agricultural crops to distill water (T₀) and different concentrations of *Acacia auriculiformis* extracts (T₁-T₅)

Treatment	Agricultural crops				
	<i>C. arietinum</i>	<i>R. sativus</i>	<i>V. unguiculata</i>	<i>B. juncea</i>	<i>P. mungo</i>
T ₀	18.29a*	17.41a	14.09a	9.93a	8.23ab
T ₁	13.49 b	16.86ab	11.05a	8.39b	9.43a
	(-26.24)	(-3.46)	(-21.58)	(-15.51)	(+14.58)
T ₂	13.63b	16.63ab	8.28ab	7.69 b	8.62 ab
	(-25.48)	(-4.48)	(-41.23)	(-22.56)	(+4.74)
T ₃	9.86bc	11.57bc	6.61bc	3.41c	6.49ab
	(-46.09)	(-33.54)	(-53.08)	(-65.66)	(-21.14)
T ₄	8.21c	9.79c	4.11c	2.76c	6.03bc
	(-55.11)	(-43.77)	(-70.83)	(-72.21)	(-26.73)
T ₅	6.42c	3.96b	2.47c	0.8d	4.70c
	(-64.90)	(-77.25)	(-82.47)	(-91.94)	(-42.89)

Table 4: Number of lateral roots developed in receptor agricultural crops to distill water (T₀) and different concentrations of *Acacia auriculiformis* extracts (T₁-T₅)

Treatment	Agricultural crops				
	<i>C. arietinum</i>	<i>R. sativus</i>	<i>V. unguiculata</i>	<i>B. juncea</i>	<i>P. mungo</i>
T ₀	24.00a*	42.73a	29.13a	5.27a	11.13a
T ₁	21.53ab	41.93a	17.93b	4.41ab	10.80ab
	(-10.29)	(-1.87)	(-38.45)	(-16.32)	(-2.96)
T ₂	16.73bc	30.87b	16.60b	4.33ab	10.47ab
	(-30.29)	(-27.76)	(-43.01)	(-17.83)	(-5.93)
T ₃	7.33d	18.60c	11.47bc	4.27ab	7.13c
	(-69.46)	(-56.47)	(-60.62)	(-18.98)	(-35.94)
T ₄	8.87d	16.73c	10.23bc	3.47b	8.20bc
	(-63.04)	(-60.85)	(-64.88)	(-34.15)	(-26.33)
T ₅	10.00cd	6.13d	5.27c	2.00c	6.87c
	(-58.33)	(-85.65)	(-81.91)	(-62.05)	(-38.27)

Values in the parenthesis indicates the inhibitory (-) or stimulatory (+) effects in comparison to control (T₀)

*- values in the columns followed by the same letter(s) are not significantly different (P≤0.05) according to Duncan's Multiple Range Test (DMRT)

was found at T₅ treatment followed by T₃ and T₄ treatment. The highest inhibitory effect (-91.94 %) was observed in *B. Juncea* at T₅ treatment while the lowest (-3.45%) was found on *R. sativus* at T₁ treatment. Maximum Relative Elongation Ratio (RER) of shoot (114.58%) was observed in *P. mungo* at T₁ treatment while the minimum (8.05%) was found in *B. juncea* at T₅ treatment (Fig.3).

Number of lateral roots developed

Table 4 presents the number of lateral root developed in different treatments. It was found that in all cases, higher concentration of water extracts had pronounced inhibitory effect on lateral root development. The highest number of lateral root developed at control

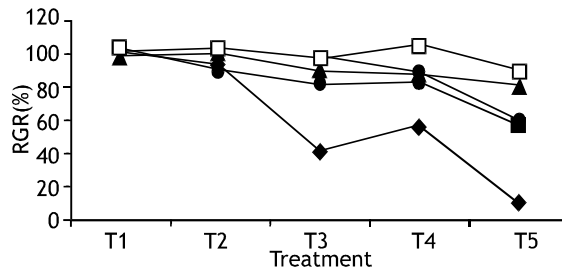


Fig. 1: Relative Germination Ratio (RGR) of bioassay species grown in petri dishes at different concentrations of *Acacia auriculiformis* extracts

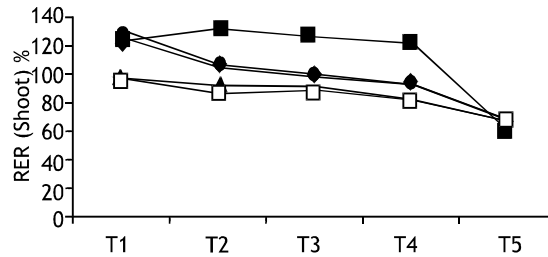


Fig. 2: Relative Elongation Ratio (RER) of shoot of bioassay species grown in petri dishes at different concentrations of *Acacia auriculiformis* extracts

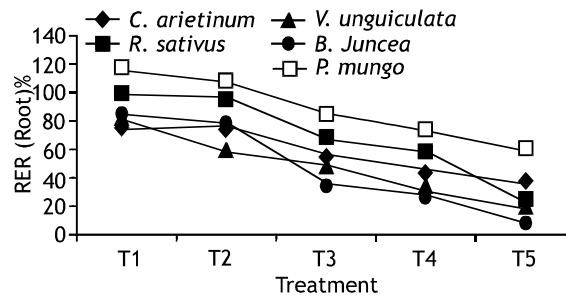


Fig. 3: Relative Elongation Ratio (RER) of root of bioassay species grown in petri dishes at different concentrations of *Acacia auriculiformis* extracts

(T₀). Maximum (42.73 nos.) root developed in *R. sativus* at control while the minimum (2.00 nos.) was recorded in *B. juncea* at T₅ treatment. The most inhibitory effect (-85.38%) was found in *R. sativus* at T₅ treatment followed by (-81.91%) in *V. unguiculata* at the same treatment while the lowest inhibitory effect (-1.87%) was in *R. sativus* at T₁ treatment followed by (-2.96%) *P. mungo* at the same treatment.

Discussion

Considering the foregoing results, it seemed that there is phytotoxic effect of *Acacia auriculiformis* leaf extract on the bioassay species germination, shoot elongation, root elongation and development of lateral root. These results correlated with the findings of Bora *et al.* (1999) who found the allelopathic effect of leaf extracts of *Acacia auriculiformis* on seed germination of some agricultural crops. This observation also confirmed the findings of Mousawi and Al Naib (1975) who reported inhibition of seed germination and seedling growth of some herbaceous plants by leaf extracts of *Eucalypts*. It was also observed that the leaf extracts of *Acacia auriculiformis* delayed as well as hindered the germination significantly in all the receptor plants compared to the control. The result also revealed that root elongation was much more inhibited than shoot elongation and germination. This confirmed the findings of Meissner *et al.* (1982) who also found that the undiluted tuber extract of *Cyperus rotundus* impeded the radicle elongation of cucumber, radish, onion and tomato. Antonio *et al.* (1999) reported the effects of aqueous extracts of *Leucaena* on germination and radicle elongation of three forage grasses in which radicle elongation was a more sensitive indicator for extracts effects rather than seed germination. The results are also in conformity with the earlier findings of Chou and Waller (1980a, 1980b); Swami Rao and Reddy (1984), Chou and Kuo (1986), Alam (1990), Chaturvedi and Jha (1992), Zackrisson and Nilsson (1992) and Uddin *et al.* (2000). All those studies supported that root growth and lateral root development was more sensitive and responds more strongly to the increasing concentration of the aqueous extract in comparison to shoot. Many bioassay species loss their ability to develop normally as a result of reduced radicle elongation and root necrosis. Based on the overall findings it can be concluded that allelopathy is a concentration-dependent phenomenon. This confirmed the report of Ballester *et al.* (1982), Rai and Tripathi (1984), Rizvi and Rizvi (1987) and Daniel (1999).

The studies provide the evidence of *Acacia auriculiformis* has allelopathic potential. Again its inhibitory effect on agricultural crops in absence of fungi and bacteria is an added evidence for allelopathy. The allelochemicals present in the *Acacia auriculiformis* can have an allelopathic inhibitory effect on different agricrops including trees and weeds species associated with *Acacia* plantations and also different agroforestry systems in field conditions. However, long-term field based studies must be carried out.

References

- Mousawi, A.H. and F.A.G. Al-Naib, 1975. Allelopathic effects of *Eucalyptus microtheca* F. Muell. J. Univ. Kuwait Sci., 2: 59-66.
- Alam, S.M., 1990. Effect of wild plant extract on germination and seedling growth of wheat. Rachis, 9: 12-13.
- Antonio, P.S.S.F., J.D.R.R. Teresinha, A.R. Luis and A.R. Ricardo, 1999. Effects of aqueous extracts of *Leucaena* on germination and radicle elongation of three forage grasses. Recent Advances in Allelopathy: A Science for the Future, 1: 391-396.
- Ballester, A., A.M. Vieitez and E. Vieitez, 1982. Allelopathic potential of *Erica vagans*, *Calluna vulgaris* and *Daboecia cantabrica*. J. Chem. Ecol., 8: 851-857.

- Bansal, G.L., H. Nayyer and Y.S. Bedi, 1992. Allelopathic effect of *Eucalyptus macrorrhyncha* and *E. yoymanii* on seedling growth of wheat (*Triticum aestivum*) and Radish (*Raphanus sativus*). Ind. J. Agric. Sci., 62: 771-772.
- Bora, I.P., J. Singh, R. Borthakur and E. Bora, 1999. Allelopathic effect of leaf extracts of *Acacia auriculiformis* on seed germination of some agricultural crops. Ann. For., 7: 143-146.
- Chaturvedi, O.P. and A.N. Jha, 1992. Studies on allelopathic potential of an important agroforestry species. Forest Ecology and Management. Elsevier Science Publishers B.V., Amsterdam, 53: 91-98.
- Chou, C.H. and Y.L. Kuo, 1986. Allelopathic exclusion of under storey by *Leucaena leucocephala* (Lam.) de wit. J. Chem. Ecol., 12: 1431-48.
- Chou, C.H. and G.R. Waller, 1980a. Possible allelopathic constituents of *Coffea arabica*. J. Chem., Ecol., 6: 643-53.
- Chou, C.H. and G.R. Waller, 1980b. Isolation and Identification by mass spectrometry of phytotoxins in *Coffea arabica*. Bot. Bull. Acad. Sinica, 21: 25-34.
- Daniel, W.G., 1999. Historical review and current models of forest succession and interference. CRC Press LLC, pp: 237-251.
- Das, S. 1985. Prothomik Bono Biggan, Kachari Road, Mymensingh. (a manual in Bengali).
- Davidson, J., 1985. Species and Sites : What to plant and where to plant. UNDP/FAO Project BGD/79/017. Field Document-5.
- Eyini, M., M. Joyakumar and S. Pannirselvam, 1989. Allelopathic effect of bamboo leaf extracts on the seedling of groundnut. Trop. Ecol., 30: 138-141.
- Gaba, R.K., 1987. Role of allelopathy in social forestry. In : P.K. Khosla and R.K. Kohli, (Eds.), Social forestry for rural development. ISTS, Solan, pp: 228-234.
- Jadhav, B.B. and D.G. Gaynar, 1992. Allelopathic effects of *Acacia auriculiformis* on germination of rice and cowpea. Indian J. Plant. Physiol., 35: 86-89.
- Joyakumar, M., M. Eyini and S. Pannirselvan, 1978a. Allelopathic effect of teak leaf extract on the seedling of groundnut and corn. Geobios., 14: 66-69.
- Joyakumar, M., M. Eyini and S. Pannirselvan, 1978b. Allelopathic effect of bamboo root extract on the seedling of groundnut and corn. Geobios., 14: 221-224.
- Khan, M.S. and M.K. Alam, 1996. Homestead flora of Bangladesh. BARC, IDRC, SDC., Dhaka, Bangladesh.
- King, K.F.S., 1979. Agroforestry and the utilization of fragile ecosystems. For. Ecol. Management., 2:161-168.
- Koul, V.K., A. Raina, Y.P. Khanna, M.L. Tickoo and H. Singh, 1991. Evaluation of allelopathic influence of certain farm grown tree species on rice. (*Oryza sativa* L. cv. PC-19). Ind. J. For., 14 : 54-57.
- Meissner, R., P.C. Nel and N.S.H. Smith, 1982. The residual effect of *Cyperus rotundus* on certain crop plants. Agroplanta, 14: 47-53.
- Melkania, N.P., 1987. Allelopathy and its significance on production of agroforestry plant associations. In : P.K. Khosla and D.K. Khurana, (Eds.), Agroforestry for rural needs. ISTS, Solan, pp: 211-224.

- Petmark, P. and E.R. Williams, 1991. Use of Acacia species in Agroforestry. In Turnbull, J.W. (Ed.), Advances in Tropical Acacia Research. Proceeding of an International Workshop held in Bangkok, Thailand.
- Pinoyopusarerk, K., 1990. *Acacia auriculiformis*: an annotated bibliography. Winrock International Institute of Agricultural Development, Australian Centre For International Agricultural Research, pp:154.
- Rahman, M., 1984. Forest Tree Improvement in Bangladesh. A Baseline Study. UNDP/FAO, Las Banos. Philippines, RAS/91/004. Working Paper No.6.
- Rai, J.P.N. and R.S. Tripathi, 1984. Allelopathic effects of *Eupatorium riparium* on population regulation of two species of Galinsoga and soil microbes. Plant and Soil, 80:105-117.
- Rao, O.P., A.K. Sexana and B.P. Singh, 1994. Allelopathic effects of certain agroforestry tree species on the germination of wheat, paddy and gram. Ann. For., 2: 60-64.
- Rho, B.J. and B.S. Kil, 1986. Influence of phytotoxin from *Pinus rigida* on the selected Plants. J. Nat. Sci. Wankwang Univ., 5: 19-27.
- Rice, E.L., 1974. Allelopathy. Academic Press, New York.
- Rice, E.L., 1979. Allelopathy- an update. Bot. Rev., 45: 15-109.
- Rice, E.L., 1984. Allelopathy. 2nd ed. Academic Press, San Diego, pp: 422.
- Rizvi, S.J.H. and V. Rizvi, 1987. Improving crop productivity in India. Role of Allelochemicals. In: Waller, G.R. (Ed.), Allelochemicals: Role in Agriculture and Forestry. ACS Symposium Series 330, pp: 69-75.
- Surendra, M.P. and K.B. Pota, 1978. The allelopathic potentials of root exudates from different ages of *Celosia argenta* Linn. Nat. Acad. Sci. Lett., 1: 56-58.
- Swami Rao, N. and P.C. Reddy, 1984. Studies on the inhibitory effect of *Eucalyptus* (hybrid) leaf extracts on the germination of certain food crops. Ind. Forester, 110: 218-222.
- Uddin, M.B., R. Ahmed and M.K. Hossain, 2000. Allelopathic potential of water extracts of *Leucaena leucocephala* leaf on some agricultural crops in Bangladesh. The Chittagong Univ. J. Sci., 24: 121-127.
- Zackrisson, O. and M.C. Nilsson, 1992. Allelopathic effects by *Empetrum hermaphroditum* on seed germination of two boreal tree species. Can. J. For. Res., 22: 44-56.