INNSPUB

International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print), 2222-5234 (Online)
http://www.innspub.net
Vol. 8, No. 3, p. 29-34, 2016

RESEARCH PAPER OPEN ACCESS

Potential allelopathic inhibitory effect of *Salix nigra* on growth of *Triticum aestivum*

Wasim Ahmed*, Mushtaq Ahmad, Rahmat Ali Khan

Department of Bio-Technology, University of Science and Technology Bannu, KPK, Pakistan

Key words: *Salix nigra*, Allelopathy, *Triticum aestivum*.

http://dx.doi.org/10.12692/ijb/8.3.29-34 Article published on March 23, 2016

Abstract

The importance of the role of allelopathy in agro-eco system cannot be denied. Molisch, in 1937, put forwarded the concept of allelopathy to explain how a plant can impart its harmful/beneficial effects upon the other with the release of certain molecules in their surroundings. The present work was carried out as a preliminary study to investigate any possible herbicidal activity of the plant's extract on growth of *Triticum aestivum*. Modified protocol of McLaughlin and Rogers was used to carry out petri plates study. Similarly, open field study was also carried out to get more precise results. Our results showed that the crude extract of our plant significantly ($P > 0.05$) inhibited the shoot (hypocotyls) growth of wheat compared to control after 3rd & 5th days. The data also indicated that the extract significantly inhibited roots growth as compared to non-treated water (control) group. After complete treatment with sample solutions, fresh and dry weight of all replicates was calculated and this exposed that our plant extract extensively ($P < 0.05$) controlled the fresh as well dry weight. Similarly, in field study, results indicated that it markedly ($P<0.05$) inhibited shoot growth compared to control. After complete treatment, fresh and dry weight of all replicates were measured which revealed that the extract significantly ($P<0.05$) decreased fresh as well dry weight of wheat.

* **Corresponding Author:** Wasim Ahmed ✉ waseem_bnu57@yahoo.com

Introduction

The dispersal and plenty of plant species is determined by their ability to strive for resources (Huston and Smith 1987, Tilman 1988, Casper and Jackson 1997; Grime 2001) and by other types of biotic interactions (Callaway, 2007). Some plant species subdue the progression or establishment of their entrants by releasing toxic composites, a mechanism referred to as allelopathy (Inderjit *et al*., 2011). Allelopathic compounds can directly impair other species by dwindling their germination or growth (Callaway and Aschehoug, 2000; Prati and Bossdorf, 2004). Alternatively, these compounds can act indirectly through effects on the soil microbial community, i.e. by suppressing mutualists or by promoting antagonists of other plant species (Mangla and Inderjit, 2008; Stinson *et al*., 2006). Thus, allelopathic compounds can play a major role in plant–plant interactions (Wardle *et al.* 1998). Moreover, allelopathic compounds can be persistent in soils. The resulting legacy effects of allelopathic species can affect future plant species and potentially entire ecosystems in the long term (Prati and Bossdorf, 2004; Stinson *et al*., 2006).

During the last decade, the concept of allelopathy has received increased attention. A no of plant species are being explored for their allelopathic potential in Pakistan (Ahmed *et al*., 2014). It is therefore, the most appropriate time to contemplate the development and organization of our medicinal plants industry to become independent in order to provide communal native natural drugs that are under use by the healers of rural areas to treat various diseases (Ahmed *et al*., 2014-15). The objective of the study was to assess the potential allelopathic interference/inhibition of *Salix nigra* against Triticum aestivum.

Material and methods

Plant collection and extraction

The leafy plant was mowed from a rural area of district Bannu and was identified and recognized by a botanist at department of Botany, UST Bannu. The voucher specimen was deposited in herbarium of the department. After drying the plant, it was crushed into fine powder. The powder was extracted in 70% methanol for a period of 7 days. Filtration was done after this one week period and filtrate was applied to rotary evaporator to make it concentrated. Freeze drying was applied in order to get powder of the extract.

Sample preparation

To precede allelopathic assay, 10mg of plant's powder was dissolved in 10ml methanol. From this stock solution, further dilution was made to 10µg/ml, 100µg / ml and 1000µg /ml (a 15ml volume was made).

In vitro allelopathic assay: petri plates study

Modified protocol of McLaughlin and Rogers (1998) was used for determining the allelopathic interference/inhibition of methanolic fraction of plant extract with triticum aestivum. The experiment was executed in triplicates. In this experiment, filter papers were set in the autoclaved petri plates. 2ml solution from 10, 100 and 1000µg/ml was sprayed/poured on the filter paper. Petri plates were already labeled. The control was not treated by the sample solution.

In order to evaporate the methanol from the sample treated plates, they were placed in the oven at 40°C. After complete evaporation, 5ml of distilled water was sprayed over all experimental and control group's plates. 5 wheat seeds, washed by distilled water and then by 1%$HgCl_2$ solution, were placed in each plate at equal distance by scientific way. All the petri plates were incubated in growth chamber for five days. After 3 days, hypocotyls/shoot and radical/root inhibition was noted with respect to control. Similarly after 5 days the inhibition was noted by the same way and mean was taken.

Allelopathic assay: open field study

Preparation of samples

For allelopathic open field study, stock solution was prepared by dissolving 5mg of plant's powder in 5ml distilled water instead of methanol due to its toxicity.

From this stock solution, further dilutions were made to 10, 100 and 1000µg/ml each one of 15ml in volume.

Assay procedure

The experiment was also performed in triplicates.102g mud was put in each disposable glass labeled as control and experimental. 7 clean wheat seeds were propagated in each glass and added 25ml dis.H_2O. All the glasses were kept in the open environment. After 7 days, length of hypocotyls/shoot of all the grown wheat seeds were noted & mean was taken.

5ml sample solution of each concentration was sprayed separately on the concerned hypocotyls/shoot of grown wheat. The control (not treated) was sprayed by 5ml of distilled water. After 7 days, the length of hypocotyls/shoot of grown wheat of all the treated and control was measured and average was taken. The difference was noted between the treated and non-treated with respect to the control.

The fresh weight of the entire treated and control fraction was recorded very carefully by digital electronic balance. After this, all the treated and control fractions were packed separately and were placed in the oven at 80°C until complete dryness. The total dry weight was noted & the difference was taken between the weights of treated fractions with respect to control.

Statistical analysis

Statistical analysis was done using SPSS 16.0. All the experiments were performed in triplicates and results mentioned as ±mean SD.

Results

Petri plate study

Three different concentrations (10µg/ml, 100µg/ml and 1000µg/ml) were used for the assessment of allelopathic interference/inhibition of *S. nigra*. The result revealed that crude extract of our plant significantly ($P > 0.05$) inhibited the shoot (hypocotyls) growth of wheat compared to control after 3rd & 5th days (Fig 1). The data also indicated that the extract significantly inhibited roots growth as compared to non-treated water (control) group as shown in Fig 2. After complete treatment, fresh and dry weight of all replicates were calculated which exposed that our plant extract extensively ($P < 0.05$) controlled the fresh as well dry weight (Table 1).

Table 1. Fresh and dry weight of wheat in the absence and presence of different concentrations of *Salix nigra* extract.

S.No	Treatments	Fresh weight	Dry weight
1	control	1.39 ±0.03	0.14 ±0.02
2	10 µg/ml	1.24±0.05	0.10±0.08
3	100 µg/ml	1.09 ±0.01	0.06 ±0.06
4	1000 µg /ml	0.96±0.03	0.03±0.05

Table 2. Fresh and dry weight of wheat in the absence and presence of different concentrations of *Salix nigra*.

S.No	Treatments	Fresh weight	Dry weight
1	control	1.26±0.04	0.18±0.032
2	10 µg/ml	1.22±0.08	0.16±0.07
3	100 µg/ml	1.12±0.09	0.10±0.02
4	1000 µg/ml	1.07±0.02	0.07±0.05

Field study

Various concentrations of *Salix nigra* methanolic extract were used for phytotoxic activity against wheat growth. The presented data indicated that it markedly ($P<0.05$) inhibited shoot growth compared to control (Fig 3). After complete treatment, fresh and dry weight of all replicates were measured which revealed that the extract significantly ($P<0.05$) decreased fresh as well dry weight of wheat (Table 2).

Discussion

Plants bearing medicinal value and their bioactive components are used worldwide for managing a no of human illnesses including inflammation, cancer, infections, cardiac diseases and several new. Natural products use for the control of all kind of diseases can be credited to their minimal adverse events as compared to drugs synthesized in the laboratory. In most areas of the world, locally used herbal treatments are common and have been under investigation to get active principal of these medications. Pakistan, a rich country in natural resources and medicinally important plants which have been in use by native healers to treat many diseases including diseases of digestive system, cardiovascular, skin diseases and infectious illnesses.

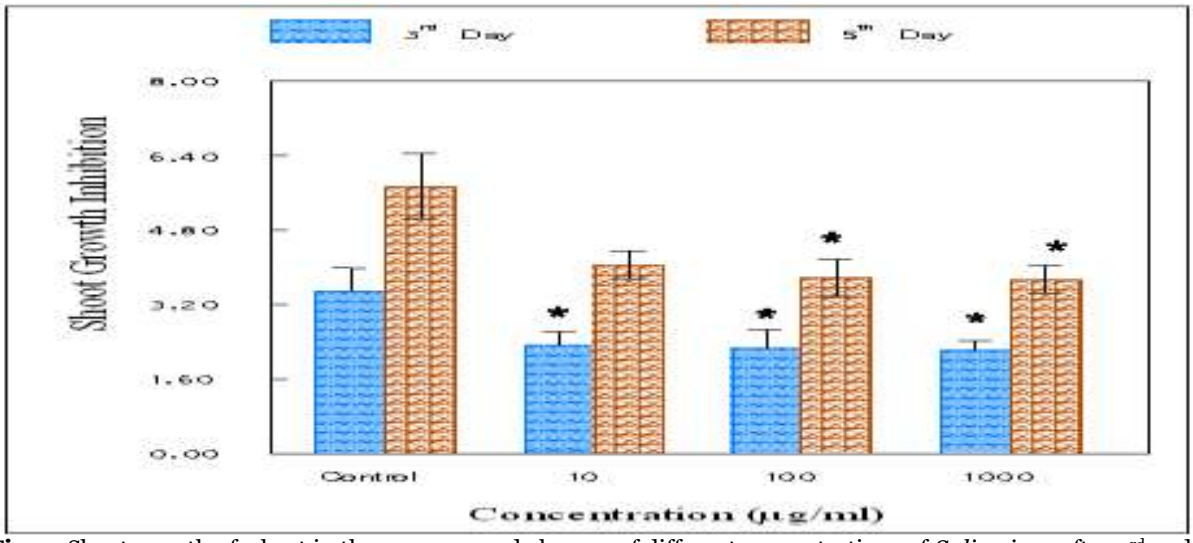

Fig. 1. Shoot growth of wheat in the presence and absence of different concentrations of *Salix nigra* after 3rd and 5th day treatment.

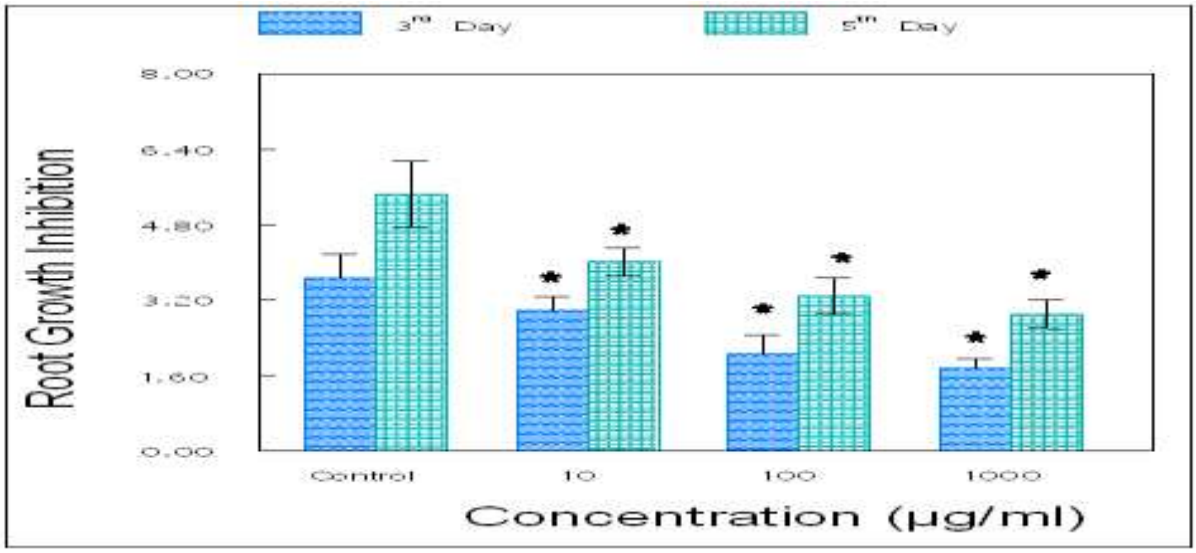

Fig. 2. Root growth of wheat in the presence and absence of different concentrations of *Salix nigra* after 3rd and 5th day treatment.

Scientists performed numerous allelopathic experiments on plants having medicinal importance. The research work of Nazir *et al.*, 2007 revealed the allelopathic potential of three herbal species (*Saussaurea lappa, Rheum emodi* and *Potentilla fulgens*). The researchers studied the allelopathic effects of these three herbs on a few food crops. It was noted that the germination of all the traditional food

crops was reduced to great extant. In recent years, exploration of allelopathic potential of the medicinal plants is a field of exceptional curiosity. Khan *et al.*, 2009 worked to check the allelopathic effects of four medicinal plants by using sandwich method. Similarly, sandwich method was also used by Fujii and Hiradate, (2007) to study the allelopathic activity of 239 plants on lettuce. The main purpose of Aziz and Fujii, (2005) to examine 14 medicinal plant species of plane areas with semi-arid conditions was of their allelopathic potential by using sandwich method. They reported that at field levels, allelochemicals play a very good role in the agricultural and functional ecology.

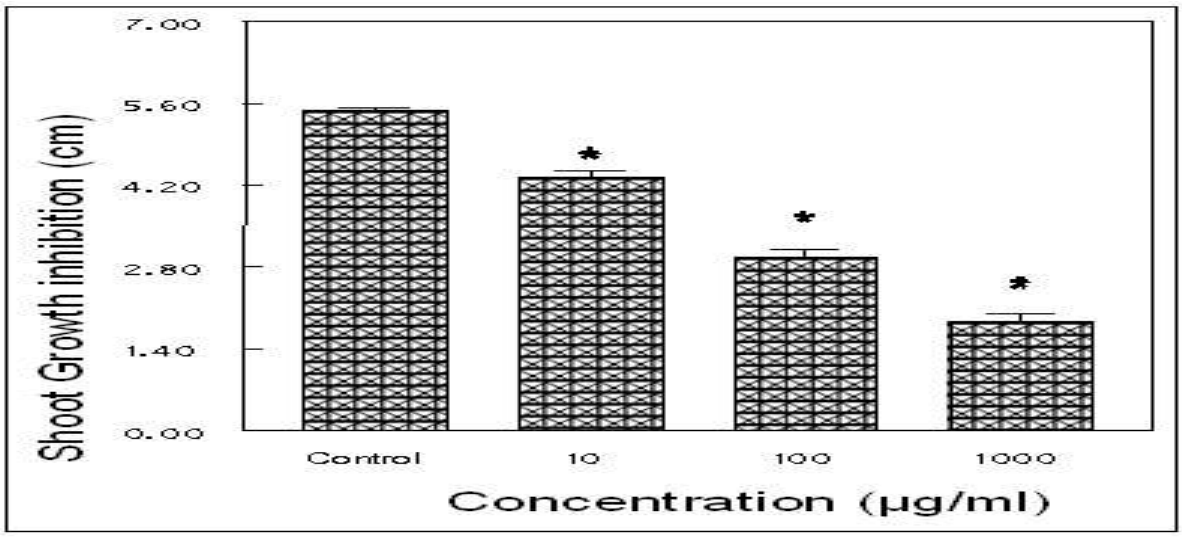

Fig. 3. Shoot growth of various groups after complete treatment.

Including Pakistan, approximately in entire world, there is a marked reduction in crop production due to weeds. The extent of losses caused by weeds was found to be more as compare to the insects and other diseases but their facts are usually ignored. Weeds control through synthetic drugs has caused various human health problems and soil water pollution (Barkatullah *et al.*, 2001). So weeds control through harmless means is required to protect environment and to increase crop yield. Crude methanolic extract of *Salix nigra* showed good phytotoxicity as compared to control which proved the presence of phytochemicals in the plant which can be further isolated and purified for future use.

Conclusion

Salix nigra could be an impending source of new phytotoxic agent. As effective inhibitory activity was observed, more research should be directed towards isolation of phytotoxic bioactive compounds as well as further field trials can be carried out to confirm the present findings.

References

Aziz M, Fujii Y. 2005. Allelopathic effect of some medicinal plant substances on germination of Amaranthus retroflexus and Portulaca. InProceeding, International Symposium on Improving the Performance of Supply Chains in the Transitional Economies. ISHS Acta Horticulturae 699.

Barkatullah M, Ibrar N, Muhammad. 2011. Evaluation of Zanthoxylum armatum DC for invitro and invivo pharmacological screening. African journal of pharmacy and pharmacology **5(14),** 1718-1723. http://dx.doi.org/10.5897/AJPP11.405

Callaway RM, Aschehoug ET. 2000. Invasive plants versus their new and old neighbors: a mechanism for exotic invasion. Science **290,** 521–523.

Callaway RM. 2007. Positive interactions and interdependence in plant communities. Springer, Dordrecht.

Casper BB, Jackson RB. 1997. Plant competition underground. Annual Review of Ecological System. **28,** 545–570.

Daniel P, John NK. 2006. Invasive plant suppresses the growth of native tree seedlings by disrupting belowground mutualisms. PLoS Biology. **4,**727–731. http://dx.doi.org/10.1371/journal.pbio.0040140

Fujii Y, Hiradate S. 2007. Allelopathy : New concepts and methodology, Science publishers. 382 p.

Grime JP. 2001. Plant strategies, vegetation processes, and ecosystem properties. John Wiley & Sons, Chichester.

Huston M, Smith T. 1987. Plant succession: life history and competition. American Naturalist **130,** 168–198.

Inderjit. 2011. Plant phenolics in allelopathy. Bot. Rev. **62,** 186–202.

Jung EH, Lee IJ. 2009. Assessment of allelopathic potential of selected medicinal plants of Pakistan. African Journal of Biotechnology **8(6),** 1024-1029.

Khan Al, Hamayun M, Hussain J, Khan H, Gillani SA, Kikuchi A, Watanab KN, Kristina AS, Stuart AC, Jeff RP, Benjamin EW, Ragan MC, Giles CT, Steven GH, Mangla S, Inderjit, Callaway RM. 2008. Exotic invasive plant accumulates native soil pathogens which inhibit native plants. Journal of Ecology. **96,** 58–67.

McLaughlin JL, Rogers LL. 1998. The use of biological assays to evaluate botanicals. Drug Information Journal **32,** 513-524.

Nazir T, Uniyal AK, Todaria NP. 2007. Allelopathic behaviour of three medicinal plant species on traditional agriculture crops of Garhwal Himalaya, India. Agroforestry systems **69(3),** 183-7.

Prati D, Bossdorf O. 2004. Allelopathic inhibition of germination by Alliaria petiolata (Brassicaceae). American Journal of Botany. **91,** 285–288.

Tilman D. 1988. Plant strategies and the dynamics and structure of plant communities. Princeton University Press, Princeton.

Wardle DA, Nilsson MC, Gallet C, Zackrisson O. 1998. An ecosystem-level perspective of allelopathy. Biological Reviews. **73(3),** 305-319. http://dx.doi.org/10.1111/j.1469185X.1998.tb00033.x

Wasim A, Mushtaq A, Nadia M, Abdul R, Zakir K. 2014. Phytotoxic and leishmanicidal activity of Salix nigra. International Journal of Biosciences. **5(7),** 75-79. http://dx.doi.org/10.12692/ijb/5.7.75-79

Wasim A, Mushtaq A, Nadia M. 2014-2015. Phytochemistry and Anti-Acetyl Cholinesterase Activity of Salix nigra. International Journal of Pharmacognosy and Phytochemical Research **6(4),** 942-944.