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ALLELOPATHIC EFFICACY *OF BUTEA MONOSPERMA* LEAF EXTRACT ON GROWTH & BIOCHEMICAL CONSTITUENTS OF *PHASEOLUS AUREUS*

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ABSTRACT

Butea monosperma is known to influence the growth of plants in its vicinity by the release of the allelochemicals during the decomposition of its litter. The present study intended to investigate the effect of aqueous extract from *B.monosperma* on the seedling growth and biochemical constituents of *Phaseolus aureus*. For the study, different concentrations viz. 0. 1%, 0.2%, 0.3%, 0.4% and 0.5% of *B.monosperma* aqueous leaf extract was prepared, while tap water served as control. *P.aureus* seeds were incubated in different concentrations of leaf extracts and biochemical analysis performed. After 15th & 30th days of exposure to the extract, seedlings were harvested for growth parameters and biochemical characteristics determination. Germination and phenotypic results showed no negative effect by the extract in T1, T2 treatments. The amount of total carbohydrate, chlorophyll and protein content were found to be stimulated in T2. Lower concentration (T2) that appears to be suitable for *P.aureus* growth, showed a decrease in stress preventing compounds such as total phenol and free proline. Optimum and healthy growth were observed in T2 treatment. The results indicate that the application of *B.monosperma* aqueous leaf extract at optimal concentration enhances the growth and may act as natural fertilizer.

Key words: *B.monosperma,* aqueous extract, allelochemicals, *Phaseolus aureus,* Biochemical characteristics

INTRODUCTION

Allelopathy is alluring and paradoxical subject that concern with the plant interaction influenced by the chemical substances that they release into the decomposition of plant material (Zeng et al., 2008, Rice, 1984). In natural ecosystems and agro-ecosystems, allelochemicals may either influence or inhibit the growth and development of recipient plants (Inderjit and Duck, 2003).

In common allelopathy is accepted as notable ecological factor in determining the chemistry of plant communities (Seigler, 1996). Some plants may exhibit inhibitory effects on seed germination and seedling growth of other neighboring plants by either releasing allelopathic substances as exudates from living plant tissues or through decomposition of plant residues (Rice, 1984; Butnariu, 2012).There are scientific work which report that the secondary metabolites such as alkaloids, tannins, flavonoids, phenolics acid, lignins and coumarins of some plants exert allelopathic action (Mathela, 1994).

Allelochemicals are highly attractive as new classes of herbicides due to a variety of advantages. However, in the perspective of bioherbicide based on allelopathic effects caused by these compounds on target plants are also classified as “phytotoxic”. Most of allelopathins are totally or partially environment (Bais et al., 2003) Allelochemicals are secondary metabolites of plants that are released into the environment by exudation from roots, leaching from stems and leaves or water-soluble which makes them easier to apply without additional surfactants. Their chemical structure is more environmentally friendly than synthetic ones (Urszula krasuska,2007). High enough the negative impact of the use of synthetic pesticides, encouraging efforts to pursue empowerment / utilization of natural pesticides as an alternative to synthetic pesticides is of great importance (Gloria Jemmi Christobel et al.2017)

Butea monosperma (Lam,) is a medium-sized deciduous tree belongs to family Luminosae- papilineae. This tree is also called ‘Flame of the Forest’ and Bastard Teak. Medicinal properties of this plants are due to presence of several chemical compounds like carbohydrate, phenol, tannin, saponin, alkaloids, steroids, and flavonoids (Rajput et al., 2011). Numerous studies have revealed a wide range of beneficial effects of *B.monosperma* seed extract on animals, microbes, and humans. Currently, no published information exists relating to the optimized use of aqueous leaf extract of *B.monosperma* on plants to promote the growth by evaluating the beneficiary plant biochemical constituents. The present study represents the first concerted attempt to assess the relative efficiency of different concentration of leaf aqueous extract of *B.monosperma* on the germination, growth and biochemical constituents of *Phaseolus aureus.*

Materials and methods

Preparation of aqueous extract

Insect-free, disease-free leaves of *B.monosperma* were collected from local area. They were washed thoroughly with distilled water and shade-dried (Fig1). The dried sample was then ground to fine powder and stored in an airtight container for further use. 0.1g (T1), 0.2g (T2), 0.3g (T3), 0.4g (T4), 0.5g (T5) of B.monosperma leaf powder was dissolved in 100 ml of tap water respectively and kept for 48 hrs (Wardle et al., 1992). The extracts were then filtered with muslin cloth .Tap water was used as control treatment(C).

Figure 1: Fresh and dry powdered leaves of *B.monosperma*

Growth Experiment

Twenty *Phaseolus aureus* seeds were placed in each of six sterile petridish lined with whatman No.1 filter paper. 5 ml of the different concentration levels of *B.monosperma* aqueous leaf extract were added per petri – dish. 5 ml of distilled water added per petri dish served as acontrol. The pot experiments were conducted under laboratory conditions with different treatments of aqueous extract of *B.monosperma* leaves.

Morphological Parameters

Morphological parameters that include shoot and root length, shoot and root weight were measured in the samples.

Figure 2: Allelopathic effect of leaf extract on of *B.monosperma* on green gram (15thday)

Figure3: Allelopathic effect of leaf extract on of *B.monosperma* on green gram (30th day)

Biochemical Analysis

Biochemical analysis to quantify Carbohydrates (Fales, 1951), Chlorophyll (Arnon, 1949),Protein(Lowry *et al.*, 1951), Phenol (Bray and Thorpe, 1954) and Proline (Bates *et al.*, 1973) were carried out in fresh samples of germinated seeds and in the leaves collected during pot culture for the period of 15th and 30th day of growth.

Statistical Analysis

Statistical analysis was performed using one-way analysis of variance (ANOVA) and the values are presented as mean ± SD for 3 samples in each group.

RESULTS AND DISCUSSION

In the present study, aqueous leaf extracts at different concentration of *B.monosperma* were tested for their allelopathy activity against *Phaseolus aureus*. The allelopathic effect was tested both at seed germination and seedling growth stage. Polyphenols and flavonoids in the *B.monosperma* leaf extracts were found to have a close correlation with scavenging and total antioxidant activities through which the plant exhibit its efficient protective mechanism such as pest control activity (Simmonds , 2003). This lead to the luminous direction to adapt allelochemicals in *B.monosperma* aqueous leaf extract *as* herbicides, pesticides and as growth stimulants.

Table 1: The effect of aqueous *B.monosperma* on morphological parameter Phaseolus aureus. All values are mean ± standard error of three independent replicates.

ROOT & SHOOT GROWTH							
Age(day)	**Treatment (w/w)**	**Root Length(cm)**	**Shoot Length (cm)**	**Root weight (g)**		**Shoot weight (g)**	
				Fresh	**Dry**	**Fresh**	**Dry**
15	Control	7.5	11	0.12	0.05	0.15	0.06
	T1	10	10.5	0.11	0.04	0.16	0.08
	T2	12	13	0.24	0.10	0.20	0.14
	T3	7	11	0.18	0.07	0.15	0.05
	T4	8.5	9.8	0.20	0.075	0.14	0.12
	T5	6	6.5	0.10	0.03	0.13	0.04
30	Control	3.5	22.5	0.04	0.02	0.62	0.11
	T1	4.2	20.2	0.05	0.03	0.58	0.12
	T2	6.5	24.3	0.08	0.05	0.74	0.17
	T3	4	21.5	0.06	0.04	0.60	0.12
	T4	4.4	20.8	0.066	0.03	0.64	0.13
	T5	3	16.2	0.03	0.01	0.55	0.10

Effect of aqueous leaf extracts of *B.monosperma* on Morphological Parameters of *Phaseolus aureus*

The treatment of aqueous *B. monosperma leaf* extract at lower rate of (0.2%) induced a stimulatory effect in growth parameters of both root and shoot of *Phaseolus aureus*. The promotion of growth was more pronounced in shoot than root as compared with their respective control at each growth stage (Figure 2&3). But increasing the level of up to 0.5% (T5) induced a pronounced reduction in shoots and root length. The reductions in plant dry and fresh weight were found increasing with the increased treatment of *B.monosperma* aqueous leaf extract. T5 treatment exhibited lowest plant fresh weight which was significant. (Table 1).

Our finding coincides with the results of Jayakumar et al., 1990 who reported that the irrigation of groundnut and maize with 5, 10, 15 and 20% water extract of abscised Eucalyptus globules leaf greatly reduced plant height. Also evidences (Abu El-Soudet al., 2001) demonstrate that stimulation in pea growth parameters by increasing the rate of the incorporated *Acacia nilotica* leaf residue from 0.25 – 0.5% (w/w), but gradual suppression at 1.5 to 2% (w/w). Similar findings have been communicated by Reigosa et al., 1999 who illustrated that allelopathic compounds are affecting many different physiological processes simultaneously and these effects are concentration dependent. Thus our finding suggest that T2 concentration of *Butea monosperma* aqueous leaf extract is considered to be the optimum concentration for the growth promotion of beneficiary plant.

Effect of aqueous leaf extract of *B.monosperma* on Carbohydrate and Protein and Chlorophyll contents of *Phaseolus aureus*

The results exhibited that the levels of total carbohydrate in leaves were found to be elevated in T2 treatment compared to the control sample. These observations were made on 15 and 30 days old green gram plants raised. Not much drastic reduction was noticed in T5 treatment than control (Fig : 4). Similar findings were obtained by E1-Darier et al., 2009 in *Triticum aestivum* L. treated with aqueous extract of *Achillea santolina* L. shoot which further supports our study.

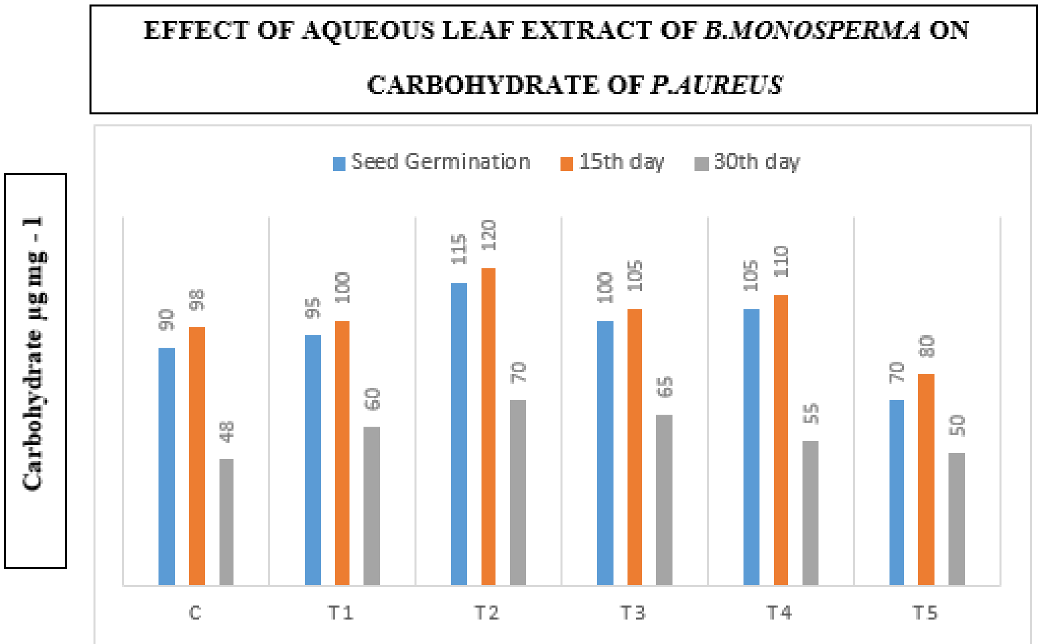

Figure 4: Effect of aqueous leaf extract of *B.monosperma* on Carbohydrate of *Phaseolus aureus*

The increased protein levels continued its trend in 30 days in treated plants T1 to T2 than the control. Protein content of *P.aureus* increased to reach 1.2 fold high at T2 treatment (Fig.5). These results are supported by the findings of Salama et al (2015) who stated that the allelopathic effects is a natural phenomenon in which the plant produces substances and metabolites that may benefit other plants when released.

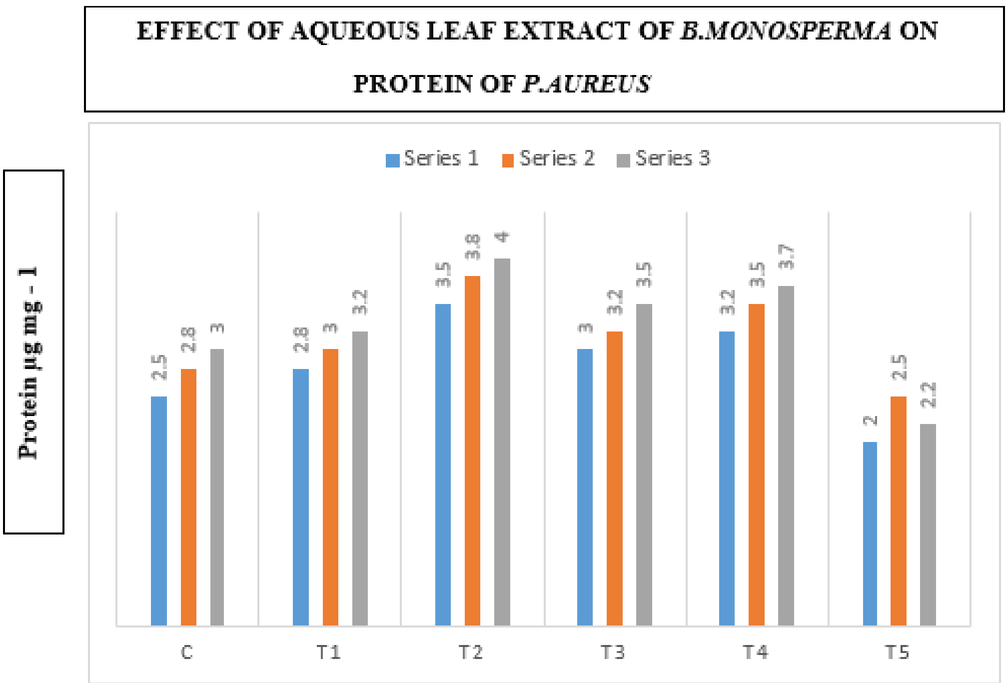

Figure 5: Effect of aqueous leaf extract of *B.monosperma* on Protein of *Phaseolus aureus*

The chlorophyll contents in the leaf imparted similar trend as that of carbohydrate which is consistent with the findings of Johnson et al., 2009 that depict a significant increment in photosynthetic pigment especially chlorophyll a and carotenoids with the application of minerals in barley thus substantiating the presence of growth promoting minerals in the aqueous leaf extract of *B.monosperma.* The chlorophyll content was reduced by 41% when *Phaseolus aureus* seedlings were treated with T5 concentration of B.M leaf aqueous extract (Fig.6). Based on the results procured in this study, we infer that morphological parameters of *Phaseolus aureus* were all stimulated at T2 specifically due to the plant's capacity to accumulate chlorophyll which is an essential component of food manufacturing process; the photosynthesis.

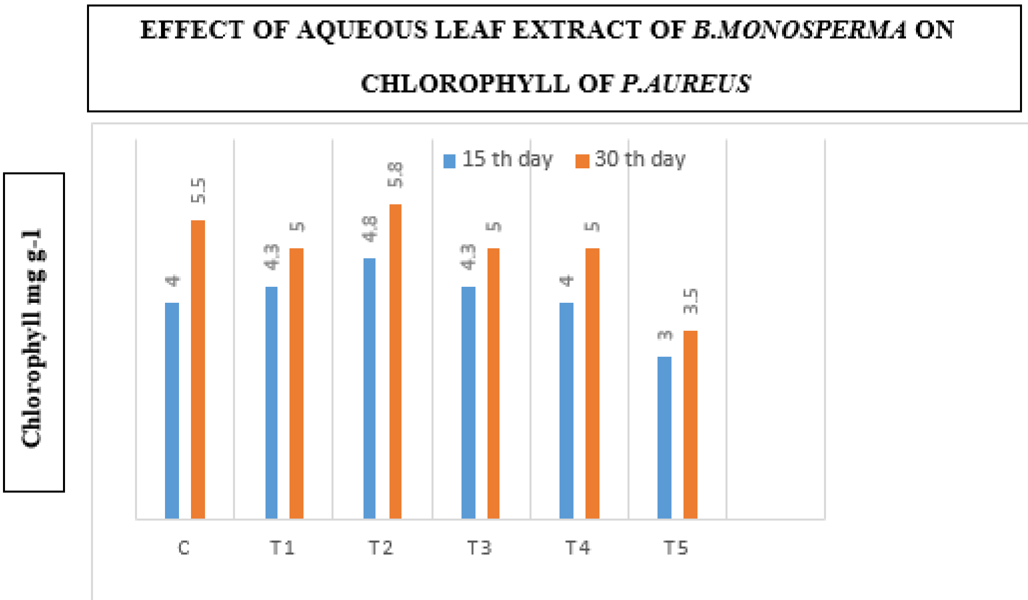

Figure 6: Effect of aqueous leaf extract of *B.monosperma* on Chlorophyll of *Phaseolus aureus*

Effect of aqueous leaf extract of *B.monosperma* on Proline and Phenol of *Phaseolus aureus*

Concomitantly, the decrease in proline levels in treated samples was also found in 30 days accelerated treatments. Free proline contents of P.aureus were the lowest at T2 and showed a decrease of 71.92% over the control indicating that the beneficiary plant is not under stress. Higher application of extract (0.5%) resulted in increased free proline contents (Figure 7). Tolulope et al., 2016 observed a significant increment in proline contents in *V.unguiculata* with the treatment of *Tithonia diversifolia* shoot extract at higher concentration. Phenol induction was also ascertained in rice plant in response to *T.diversifolia* leaf extract (Iloriet al, 2007). Increase in proline content at T5 treatment might be due to the protein breakdown. Report by Guerrier 1998 documents that proline is synthesized from glutamate that resulted due to protein breakdown.

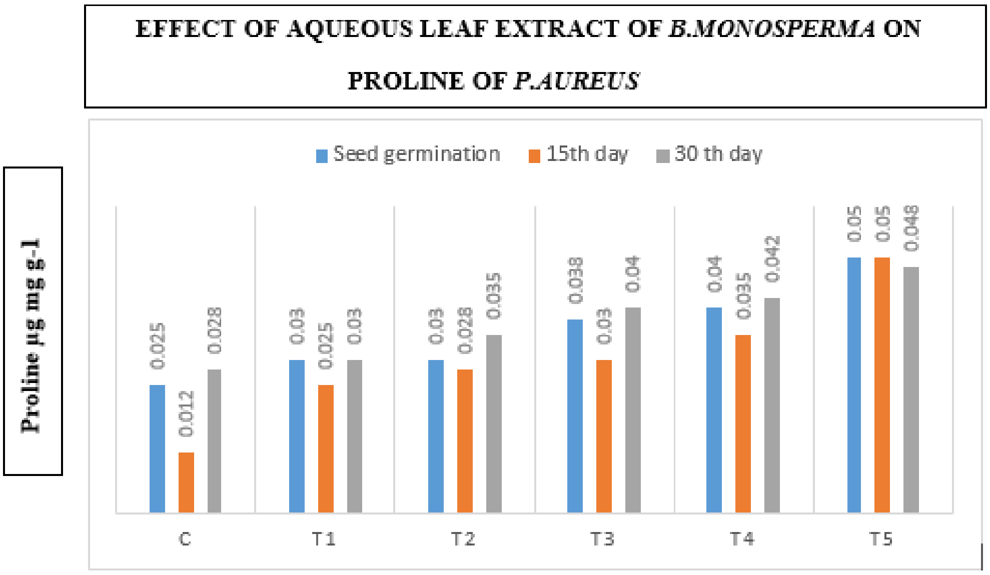

Figure 7: Effect of aqueous leaf extract of *B.monosperma* on Proline of *Phaseolus aureus*

Phenol content from T1 to T2 treatment was found to be relatively similar and quite higher than control but lower than T5 (Figure 8). Results of the present study indicate that effects of *B.monosperma leaf extract* on phenolic compound accumulation depends on the species and the applied extract (Magdalena Szwed et al., 2014). Phenolic compounds, in addition to antioxidant properties, may also display prooxidant effect (Sakihama et al., 2002.). *B.monosperma leaf extract* are a rich source of those substances, which is acknowledged as a basis of allelopathic properties of that species (Rajendra Prasad et al 2016). Inhibition of the growth of acceptor plant (*Phaseolus aureus*) under the effect of *B.monosperma* leaf extract may therefore be related to disturbance in oxidoreduction transformation in their tissues (Magdalena Szwed et al., 2014). This may lead to the oxidation damage of proteins, lipids, and nucleic acids (Sharma et al., 2012). But this perception was slightly different in our results obtained which clearly portray that application of extract at lower concentration might neutralize the inhibitory effect of allelochemicals thus highlighting its beneficial effect. Similar finding was observed by Mewis et al., 2012 whose work stated that some biotic factors may also cause a decrease in the content of antioxidants. The increased Phenol content of the treated plant may attribute to its pest and weed resistance (Khawar Jabran et al 2015).

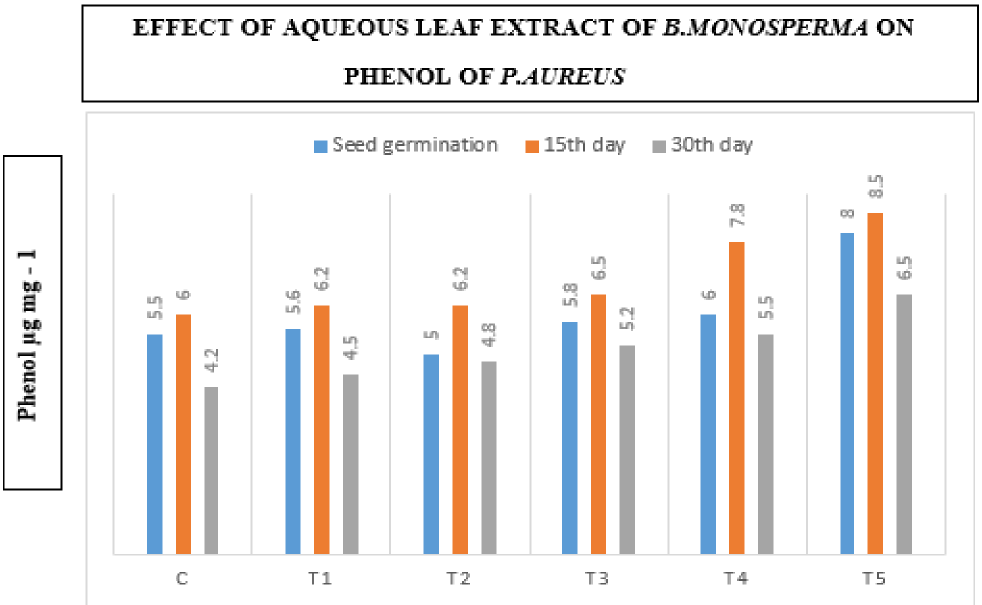

Figure 8: Effect of aqueous leaf extract of *B.monosperma* on Phenol of *Phaseolus aureus*

CONCLUSION

The allelochemicals in *B.monosperma* leaf extract had significant effects on seedling growth and on the biochemical parameters during germination and growth of *Phaseolus* Authors would hereby like to declare that there is no conflict of interests that could *aureus*. Moreover the plants were found to be healthy which may be a basis for further research on the use of *B.monosperma* leaf extract as natural fertilizer and this is an economic procedure. Conflict of interest possibly arise.

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