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Full Length Research Paper

Allelopathic potential of rhizosphere soil of *Croton bonplandianum* on growth and establishment of some crop and weed plants

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This experiment was carried out to study various biological and ecological features of *Croton bonplandianum* Baill and physico-chemical characteristics of its rhizosphere soil, and determine the effect on the growth of some crop and weed plants that is *Triticum aestivum* L., *Brassica rapa* L., *Brassica oleracea* var. *botrytis* L. and *Spinacea oleracea* L., *Melilotus alba* Medik., *Vicia sativa* L., and *Medicago hispida* Gaertn. While comparing the features of the weed between pre- and post flowering stages, it was observed that the growth pattern favours the fast spread and high density of the weed. Root length, shoot length and dry weight of seedlings of test plants decreased significantly when plants were grown in rhizosphere soil of *C. bonplandianum* Baill. The rhizosphere and control soil were analyzed for pH, electrical conductivity, organic carbon, organic matter, available nitrogen, phosphorus, potassium, and micro-nutrients such as sodium, iron, manganese and zinc at pre- and post flowering stage. The pH of rhizosphere soil decreased compared to control whereas the conductivity, organic carbon and organic matter increased. The presence of significantly high amount of phenolics in rhizosphere soil indicated their possible interaction with soil chemical properties. This was also indicated by the correlation analysis between phenolics and various properties.

Key words: Allelopathy, nutrients, phenolics, soil chemistry.

INTRODUCTION

The *Croton bonplandianum* is an obnoxious weed of family Euphorbiaceae, native of South America and was reported from India during late 1890 by Kaul (1967). It now occurs widely along roadsides, railway abandoned field in wide open ravines, and paddy or sugarcane fields and on sandy or sandy clay soils. This species is seldom found in areas enclosed by shrubs and trees where free movement of air is hindered. Flowering: April to August and Fruiting: September to December. In areas invaded by this weed, sustainability and soil health are deteriorated because of its fast spread and utilization of nutrients; it thus damages the whole ecosystem. In uncultivated lands, upon invasion, it spreads vigorously covering the large area forming pure stands and thereby reducing grazing area. It checks the growth of other grasses and weeds and causes the thinning of floral diversity. It was visually observed during the field survey that the weed and crop plants which are selected as test plants were severely affected by *C. bonplandianum.*

For obvious reasons, *C. bonplandianum* is a problem weed for farmers, ecologists/biologists, horticulturists, environmentalist and common man especially in north India. It over competes other plant species upon invasion resulting in adverse effect on natural vegetation and standing crops. Its invasion in crop field hinders preparation of fields while ploughing. Due to its competitive nature, it results in reduction in availability of nutrients to crops thereby reducing productivity both quantitatively as well as qualitatively (Datta and Ray, 1973, 1975).

It was visually observed that any field left to fallow is likely to be invaded by this weed and thus affecting the growth of other plants. The reason for this impact of the weed in the area it invades can not be ascertained, in the absence of any study, It is hypothesized that the success of the invasive tendencies of the weed are due to its allelopathic properties. This study therefore is an attempt to investigate the cause of the invasive nature of *C. bonplan-*

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dianum and the possible allelopathic effect of the soil from its rhizosphere on growth and establishment of selected crop and weed plants.

MATERIALS AND METHODS

Measurement of plant

Plants of *C. bonplandianum* growing locally were uprooted at pre- and post flowering stages and measured for features like average height of the above ground part, primary root, secondary roots, average number of leaves per plant etc. These parameters were measured with the help of measuring tape. Dry weights of the samples were determined following oven drying method. The numbers of secondary roots, inflorescence, seeds etc. were counted directly. Basal area of stem (small), lengths and widths of the seeds were determined by using Vernier caliper. For measurement of any of the feature, 100 plant samples were used.

Collection of soil and evaluation of its physico-chemical characteristics

At the time of termination of the experiment, soil samples from five randomly selected pots (control as well as test pots) were analyzed for pH, conductivity, phenolics, organic carbon, organic matter and nutrients. The pH and conductivity were measured in a 1: 5 soil water (w/v) paste with the help of digital pH and conductivity meter, respectively. Organic carbon and organic matter were measured by using the rapid titration method (Walkley and Black, 1934), total phenolics by (Swain and Hills, 1959). Available nitrogen (Kjeldahl's method), available phosphorus (molybdenum blue method), potassium and sodium (ammonium acetate extract, pH 7) and also available iron, manganese and zinc (using an atomic absorption spectrophotometer) were measured (Allen, 1989). The measurements were taken after 1 week of incorporation. At least three replicates were maintained for each analysis.

Growth studies

For growth studies, seeds of crop plants that is *Triticum aestivum*, *Brassica rapa*, *Brassica oleracea* var. *botrytis, Spinacea oleracea* and weed plants that is *Melilotus alba, Vicia sativa* and *Medicago hispida* were procured from Indian Agriculture Research Institute (New Delhi) and National Research Centre for Weed Science Adhartal, Jabalpur (M.P.) respectively. They were subjected to growth studies in pots filled with soil samples (from *C. bonplandianum* inhabited area as well as control). For each test plant and treatment, five replicates were maintained. The whole set-up was maintained under green house conditions. After one month, seedlings were uprooted carefully, keeping the root system intact. Their root and shoot lengths were measured and biomass quantified after oven drying.

Statistical analysis

The data were subjected to ANOVA followed by Duncan's Multiple Range Test (DMRT) (Duncan, 1955) and 2 sample t-test, wherever applicable.

RESULTS

Growth characteristics

Plants of *C. bonplandianum* exhibited variation between pre- and post flowering stages with regards to many of its characteristics. Aerial cover area in the pre-flowering stage was 0.02 ± 0.003 m^2 while at post flowering stage, it was 0.41± 0.03 m^2 (Table 1). The indicating spread of roots was 24.13 ± 3.17 cm^2 and it increased to 84.26 ± 2.20 cm^2 at post flowering stage (Table 1). Rhizosphere area change from pre to post flowering stage was about 249.19%. However, the pre-flowering stage after bolting under favourable conditions, the leaf number was 680.9 ± 18.13. The number of leaves was found to be more (nearly three times) at the post flowering stage nearly 200% (Table 1). The number of branches, plant height and length of primary roots was increased with 89.63, 271.36 and 539.49% respectively. Likewise, the average length of secondary and tertiary root was increased with 58.79 and 109.43% compared to pre-flowering stage.

The average fresh weight of the one month old plant at pre-flowering stage was 34.16 ± 5.48 g. In a span of two months when plants changed from pre-flowering to post flowering stage, the fresh weight increased to 175.7 ± 26.9 g showing an increase of about 414.34% (Table 1) The stem at pre-flowering stage was less in biomass with an average value of 4.71± 0.67 g /plant. When plant matured, the biomass was measured 92.15 ± 10.9 g /plant. There was a drastic change in the fresh weight of stem. The fresh weight of the leaves in pre-flowering phase/plant was 21.19 ± 5.44 g /plant compared to 40.69 ± 7.58 g /plant during post-flowering stage approximately 92% increase (Table 1). The increase in root fresh biomass from pre- to post flowering stage was drastic. During pre-flowering stage, it was 0.96 ± 1.82 g, which increased to 28.46 ± 2.13 during post flowering stage (Table 1). Increase in biomass from pre- to post-flowering stage was drastic. During pre-flowering stage, it was 0.96 ± 1.82 g /plant which increased to 28.46 ± 2.13 g /plant during post flowering stage (Table 1).

The dry biomass of the above ground plant parts during pre-flowering stage was 6.52 ± 2.14 g /plant while at post flowering stage 35.47± 4.29g /plant. The values of different parts of the plant that is stem, leaves and root during pre-flowering stage were indicating approximately 961. 79, 151.31 and 660.29% respectively (Table 1).

The flowers of C*. bonplandianum* were glabrous and bearing on an average of 159.9±12.6 inflorescences/plant while average number of flowers/inflorescence were 11.19 ± 0.15 with fresh and dry biomass of 27.23 ± 3.13 and 11.46 ± 2.62 g respectively (Table 1).

On an average number of seeds/plant 345.8± 9.76 was counted. Each seed measured about 4.7 ± 0.22 mm in length and 2.8 ± 0.17 mm wide, with a weight 1.02 ± 5.36 g /100 seeds (Table 1).

Elemental analysis

A significant difference in the amount of various elements was observed in *C. bonplandianum* at both pre- and post flowering stages. Exception, however, this was in the case of a, e and Mn (Table 2) where the variation was statistically insignificant at 5% level. Except N and Cu, the amount of

Table 1. Characteristic of weed *C. bonplandianum* Baill collected from study site.

Features	Pre Flowering stage	Post flowering stage	% Variation
Growth Features			
Rhizosphere area (cm^2)	24.13 ± 3.17	84.26 ± 2.20	249.19
Basal area (cm^2)	6.87 ±0.53	15.39 ± 0.77	124.02
Aerial spread (m^2)	0.02 ± 0.003	0.41 ± 0.03	1950.00
Average number			
Leaves/plant	680.9 ±18.13	2040 ±22.26	199.60
Branches /plant	8.49 ± 2.46	1 6.10 ± 5.60	89.63
Average length (cm)			
Above ground part	18.12 ± 2.68	67.29± 9.23	271.36
Primary root	6.28 ± 2.13	16.16 ± 5.16	157.32
Secondary root	7.11 ± 1.19	11.29 ± 3.26	58.79
Tertiary root	2.12 ±1.10	4.44 ± 2.13	109.43
Fresh biomass (g)/ plant			
Above ground part	34.16 ± 5.48	175.7 ± 26.9	414.34
Stem	4.71 ± 0.67	92.15 ± 10.9	1856.48
Leaves	21.19 ± 5.44	40.69 ± 7.58	92.02
Roots	0.96 ± 1.82	28.46 ± 2.13	2864.58
Dry biomass (g)/plant			
Above ground part	6.52 ± 2.14	35.47 ± 4.29	444.02
Stem	2.46 ± 0.33	26.12 ± 4.92	961.79
Leaves	8.79 ± 0.16	22.09 ± 3.17	151.31
Root	0.68 ± 0.43	5.14 ±0.99	655.88
Inflorescence			
Number of inflorescences / Plant	-	159.9± 12.6	
Number of flowers/ inflorescence	-	11.19 ±0.15	
Fresh biomass/plant (g)	-	27.23 ±3.13	
(d) Dry biomass/plan (g)	-	11.46± 2.62	
seeds			
Number/plant	-	345.8± 9.76	
Length (mm)	-	4.7± 0.22	
Width (mm)	-	2.8± 0.17	
Weight of 100 seeds (mg)	-	1.02 ± 5.36	

*The data between the Pre-and post-flowering stage were significantly different applying 2 sample t-tests ± represent standard deviation.

of elements per unit dry weights at post flowering stage were relatively less than that of pre-flowering stage. In case of N and Cu, however, the trend was opposite that is compared to 0.384% of N /unit dry weight during vegetative phase, an increase in the amount was observed (0.572%) during post flowering stage. The percent increase was 48.96% (Table 2). Likewise, in case of Cu, the amount was more during post flowering stage, indicating 100% increase compared to pre-flowering stage (Table 2). There was significant percent decrease in the amount of P, K, Ca, Mg and Zn (that is about, 28.84, 26.37, 15.01, 58.59 and 76.47%, respectively) at post flowering stage compared to pre-flowering stage. In case of Fe and Mn, there was very less change in amount (1.88 and 3.15%) during pre- and post flowering stage, respectively.

Soil characteristics

Electrical conductivity was found maximum in *C. bonplandianum* invaded site at post flowering stage followed in sequence by pre-flowering stage and control (Table 3). The differences among these three were also statistically significant. The soils were also analyzed for phenolic content. The maximum amount of phenolics was found in *C. bonplandianum* invaded site at pre-flowering stage followed by post flowering stage and least in control. The differrences were also statistically significant and similar differrences in regards to organic carbon and organic matter were observed (Table 3). Thus, the amount of organic carbon were in a maximum in soil of *C. bonplandianum* invaded site at pre-flowering stage followed by post

Table 2. Elemental analysis of weed *C. bonplandianum* at pre-flowering and post-flowering stages.

Element (units)	Pre-flowering stage	Post flowering stage	% Variation
N (%)	0.384± 0.02	0.572± 0.05	48.96
P (%)	0.579 ±0.04	0.412± 0.030	-28.84
K (%)	4.02± 0.19	2.96± 0.58	-26.37
Na (%)	0.022 ±0.002	0.020 ±0.003^{ns}	-9.09
Ca (%)	34.32 ±3.48	29.17 ±3.63	-15.01
Mg (%)	22.12± 3.44	9.16 ±2.34	-58.59
Zn (ppm)	1.87± 0.15	0.44± 0.11	-76.47
Fe (ppm)	16.45 ±2.13	16.140±1.37 ^{ns}	-1.88
Mn (ppm)	1.27 ±0.55	1.23± 0.07 ^{ns}	-3.15
Cu (ppm)	0.18 ±0.003	0.36± 0.004	100.00

ns represent insignificant different between pre-flowering and post-flowering stage applying 2 sample t-test.
± represent standard deviation.

Table 3. General characteristics of soil collected from *C. bonplandium* infested areas.

Soil characters	Control	Pre-flowering stage	Post-flowering stage
pH	7.63^{a}	7.47^{a}	7.62^{a}
Conductivity (µS)	132.1^{c}	173.6^{b}	187.5^{a}
Phenolic content (mg/100g soil)	0.21^{c}	1.08^{a}	0.74^{b}
OC (%)	1.08^{c}	1.84^{a}	1.63^{b}
OM (%)	1.37^{c}	3.13^{a}	2.83^{b}
N (kg/ha)	175^{c}	203^{b}	224^{a}
P (kg/ha)	162.4^{b}	188^{a}	189^{a}
K (ppm)	104^{c}	143^{b}	152^{a}
Na (ppm)	37.4^{b}	59.4^{a}	61.1^{a}
Ca (g/100g)	2.31^{c}	6.9^{a}	4.29^{b}
Mg (g/100g)	1.51^{b}	2.59^{a}	2.78^{a}
Cl (g/100g)	3.74^{b}	5.26^{a}	2.68^{c}
HCO_3 (g/100g)	13.74^{c}	30.41^{a}	20.42^{b}
Zn (ppm)	2.4^{c}	5.73^{a}	6.1^{b}
Fe (ppm)	5.4 ^{a}	7.4 ^{c}	10.13 ^{b}
Mn (ppm)	10.2^{c}	10.73^{a}	13.4^{b}
Cu (ppm)	0.24^{c}	0.7^{b}	0.76^{a}

Different superscript symbols in a row represent significant difference at P<0.05 applying DMRT.

flowering stage and control.

The soils under observation (that is pre, post flowering and control) were also assessed to determine the macro and micro nutrients amount. In general, maximum amount was calculated in the soil of *C. bonplandianum* site at pre-flowering stage, followed by post flowering stage and control. Exception, however, were observed in the content of K, Na, Mg, Zn, Fe and Cu where the maximum amount of respective element or nutrient was found in soil at post flowering stage, followed by pre-flowering stage and control. The differences in the amount of almost all macro- and micronutrients among the control, pre and post flowering stage were found to be statistically significant (Table 3).

Growth studies in rhizosphere soil of *C. Bonplandianum*

Germination

Seeds of each test plants (crops and weeds) were germinated in the soil, collected from *Croton* invaded field as well as the control. Since, found there was no change in germination. Data have not been presented.

Root length

In general, root length of test plants emerging from the seeds sown in rhizosphere soil of *C. bonplandianum* was

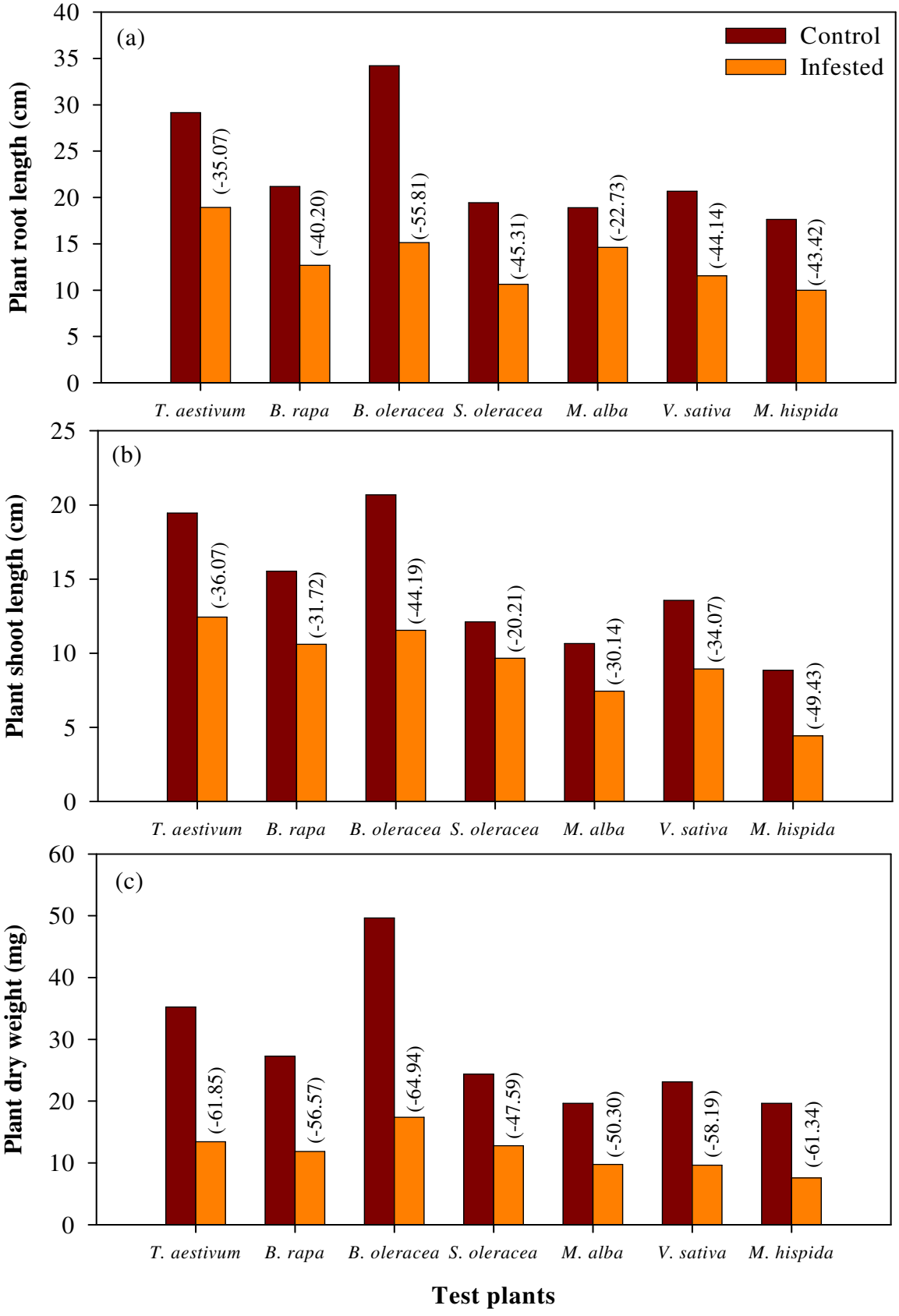

Figure 1. Root length (a),shoot length (b) and dry weight (c) of test plants one month after sowing in soil collected Croton invade area or control.

shorter than those of control. The root length of *B. oleracea* var. *botrytis* was found to be 34.22 ± 0.19 cm in control. Compared to this, when grown in soil collected from *C. bonplandianum* invaded area these root length was 15.12 ± 0.57 cm reduced by 55.81% as compared to control (Figure 1a).This reduction was statistically significant. In case of *T. aestivum* root length in *C. bonplandianum* invaded soil was 18.92 ± 0.25 cm compared to 29.14 ± 0.68 cm in control, reduced by 35.07% (Figure 1a). Similarly, the reduction in root length was also noticed in the paplionaceous weed like *M. alba, V. sativa and M. hispida* and it was 22.73% in *M. alba*, 44.14% in *V. sativa* and 43.42% in *M. hispida*. Likewise, in *B. rapa* and *S. oleracea* reduced by 40.2 and 45.3% in root length respectively (Figure 1a).

Shoot length

The shoot length of each test plant grown in the soil collected from *C. bonplandianum* rich was less than the respective values of those grown in soil collected from *C. bonplandianum* free area or control. In case of *B. oleracea* var. *botrytis,* the shoot length of seedlings was 20.68 ± 0.24 cm, when grown in control (Figure 1b). Compared to it, when grown in the soil collected from *C. bonplandianum* infested area, the shoot length was 11.54 ± 0.76 cm (Figure 1b) reduced by 44.1%. In *T. aestivum*, shoot length was reduced by 36.07% as it was 12.44 ± 0.75 in *C. bonplandianum* invaded field soil in comparison to 19.46 ± 0.28 cm in control soil (Figure 1b). While in case of *B. rapa* and *S. oleracea* when grown invaded field soil was reduced by 31.7% (10.61 ± 0.34 cm) and 20.2% (12.12 ± 0.11 cm) respectively in control (Figure 1b). In weed plants maximum reduction 49.8% was observed in *M. hispida* that is 4.44 ± 0.52 cm compared to 8.85 ± 0.32 cm in control followed by 34.07% reduction in *V. sativa* and 30.1% in *M. alba* compared with the values of 13.56 ± 0.11 cm and 10.65 ± 0.39 cm, respectively in control soil. This reduction was statistically significant.

Dry biomass

The dry biomass, of seedling grown in *C. bonplandianum* invaded soil was less as compared to control. Among all test plants maximum reduction was observed in crop plants that is about 64.9% in *B. oleracea* var. *botrytis* but in *T. aestivum* the reduction was 61.8%, in *B. rapa* 56.5% maximum reduction was observed in *M. hispida* about 61.3%, followed by in *V. sativa* about 58.1% and about while in *S. oleracea* 47.5%. In the weed test plants 50.3% in *M. alba* (Figure 1c). In all cases, a significant reduction in plant dry weight was noticed.

DISCUSSION

The studies indicate that some inhibitors are present in the rhizosphere soil of *C. bonplandianum* that adversely affects the early growth of test plants compared to control. The presence of phenolics detected in the soil inhabited by *C. bonplandianum* indicates that it might be adversely affecting the growth of other plants grown in rhizosphere soil (Leela, 1992; Singh and Thaper, 2002). Likewise the amount of all the nutrients (whether macro- or micro- or ions) was higher in *C. bonplandianum* field soil compared to control soil and hence they are not responsible for growth retardatory effects of crops. On the other hand, the pheolics, a well known group of secondary metabolites (Harborne, 1989; Mizutani, 1999) were found in appreciable amount in rhizosphere soil from *Croton* invade area compared to control. Several studies have indicated that these phenolics are responsible for growth retardatory effect on other plants includeing crops thus causing appreciable injury in the growing plants (Rice, 1984, 1995; Qasem and Foy 2001; Weston and Duke, 2003).

Rhizosphere soil is an active root zone of soil, which is densely populated and where most of the biotic interacttions among microorganisms occur (Walker et al., 2003). It is also an abundant source of organic material on which fauna and flora is dependant for food (Ryan and Delhaize, 2001). Most of the chemicals especially allelochemicals released from plants also accumulate in this zone. These may be released by roots as exudates or from above ground parts through leachation or microbial degradation. Roots, however, are known to serve as one of the major source of organic chemicals released through root exudation. These exudates may contain a diversity of chemicals that regulate the biotic communities of soil besides its physical and chemical properties. These also inhibit growth of competing species (Rovira, 1969). Verma and Rao (2006) have reported that plants release a number of low (phenolics) and high (polysaccharides, proteins) in this respect. The presence of phenolics in rhizosphere soil of *Croton* invaded fields indicates that these might have been release from the plants through any of the mode. Based on these observations, the growth retardatory effects of crops may be attributed to phenolics in the rhizosphere soil of *Croton* invaded fields.

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