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Table: 1, Figs : 5

Allelopathic effects of *Panax notoginseng* plant extracts on germination and seedling growth of soybean (*Glycine max*) and corn (*Zea mays*)

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ABSTRACT

We investigated the allelopathic effects of aqueous extracts of seeds and different parts of *P. notoginseng* on seed germination and seedling growth of soybean (*Glycine max*) and corn (*Zea mays*). In soybean, leaf extracts hastened the germination and significantly stimulated the shoot and root length than other extracts, the mean stimulation rate in shoot length was 90%, 49%, 50% and 20% with extracts of leaf, stem, root and seed, respectively. While in root length, it was 70%, 29%, 43% and 18% at extracts of leaf, stem, root and seed, respectively. In corn, the minimum germination was observed with *P. notoginseng* leaf extracts. Corn seeds treated with leaf extract showed the maximum shoot and root length than stem, root and seed extracts and *P. notoginseng* seed extract strongly increased the dry weight than other extracts, the mean increasing rate of dry weight reached 7.0%, 0.64%, 8.3% and 22% at extracts of leaf, stem, root and seed respectively. The mean sensitivity index (MSI) indicated that *P. notoginseng* extracts inhibited the corn seed germination but slightly stimulated the seedling growth. These extracts stimulated both soybean seed germination and seedling growth. The results suggested that the *P. notoginseng* extracts contain allelochemicals, which affect the germination and seedling growth of corn and soybean, and sowing of soybean instead of corn is a practical approach to avoid inhibition in *P. notoginseng* cropped soil.

Key words: Allelopathic effects, corn, germination, *Glycine max, Panax notoginseng*, seedling growth, soybean, *Zea mays*

INTRODUCTION

Allelopathy refers to the beneficial or harmful effects of one plant on another through release of biochemicals and has an important function in agricultural ecosystems, as evident by its wider use in crop protection as an environment-friendly approach to replace herbicides, fungicides and insecticides (9,20,25). *Panax notoginseng* is mainly grown in Yunnan Province, China, and its root is widely used in traditional Chinese medicine. Saponins and steroid glycosides, the major bioactive compounds in this plant are used in pharmacological uses (anti-inflammation, antisenile and immunological functions) (16,22). Wenshan in Yunnan Province is major growing region of *P. notoginseng* in China,

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however, its continuous cropping causes soil sickness due to allelopathy (30). Lands planted once with *P. notoginseng* can no longer grow *P. notoginseng* over the next 8 to 10 years, so farmers grow other crops in these lands, and soybean and corn are two common crops grown by farmers. Considering the lack of information on the allelopathic effects of *P. notoginseng* on germination and seedling growth of soybean and corn, we studied the allelopathic effects of *P. notoginseng* seed, root, stem and leaf extracts on germination and early growth of soybean and corn, to select is more suitable for planting, and further to help farmers choose suitable crop and obtain higher yields.

MATERIALS AND METHODS

The study was conducted in the Laboratory of Chinese Medicine College, Beijing University of Chinese Medicine, China. The *P. notoginseng* samples were collected in November 2013 from Wenshan, Yunnan Province. Soybean (ZhongHuang-13) and corn (LiYu-16) seeds were purchased from Chinese Academy of Agricultural Sciences, Beijing.

Aqueous extraction: *P. notoginseng* seeds, roots, stems and leaves were air-dried separately, ground to powders (4,13), and sieved through a 60-mesh net screen to remove larger particles. To prepare 20 g/L solution, 20 g of powdered material was immersed in 1 L deionised water and stored at 4 °C for 24 h (3,5,12). The solution was then filtered and the extracts were diluted with sterile distilled water to obtain final concentrations of 0%, 0.1%, 0.5%, 1.0%, and 2.0% (1,2,6). Sterile water was used as the control.

Experimental: Soybean and corn seeds were surface-sterilised with 0.5% potassium permanganate for 20 min and rinsed with distilled water (15,29). The seeds were then soaked in diluted aqueous extracts for 12 h at room temperature. Twenty seeds were then planted in a 90 mm Petri dish containing 30 g sterilised silica sand and incubated at 25 °C. Each treatment was replicated thrice. Silica sand was irrigated with 5 mL distilled water every day. Seed germination defined as the emergence of 1 mm long radicles from the testa, was recorded daily. The soybean experiment was terminated 10 d after sowing, while the corn experiment was terminated 15 d after sowing (8,21). Shoot and root length and dry weight (drying conditions, 45°C) were also determined.

Germination % (G) and mean germination time (MGT) were calculated as under:

$$G = (n / N) \times 100\ \% \qquad (14)$$
$$MGT = [\sum (Ni \times Di)] / (\sum Ni) \qquad (26)$$

Where, G: Germination %, n: number of seeds that germinated, N: number of seeds planted, Ni: germinated seeds at each observation, Di: Days of each observation, MGT: Mean germination time (days).

The allelopathic response index (RI), which measures each treatment response (T) to control (C), was calculated. RI values were then averaged arithmetically to obtain the mean sensitivity index (MSI). The RI and MSI were determined as under:

If $T \geq C$ then $RI = 1 - (C/T)$
If $T < C$ then $RI = (T/C) - 1$ (27)

$$MSI_R = \frac{\sum RI}{n} \quad (17,33)$$

Where RI: Response index, T: Treatment response, C: Control response, R: the level of mean sensitivity index (MSI), n: Total number of data (RI) at a specific level.

RI and MSI_R values ranged from –1 to +1, with positive values indicating stimulation by the treatments and negative values indicating inhibition relative to the controls.

Statistical analysis: The data were statistically analysed by analysis of variance. Significant differences amongst treatments were identified using the LSD test. Statistical analyses were done using SPSS for Windows, version 16.0.

RESULTS AND DISCUSSION

Germination

The effects of *P. notoginseng* extracts on soybean and corn seed germination depended on the tissue type and concentration of *P. notoginseng* applied.

Comparing the effects of extracts from different tissues on various parameters, the maximum germination of soybean (2.5 %) was observed with root extract. Leaf extract also increased and hastened the germination (MGT) in soybean, and the increase in germination was 1.8%. While, in corn, the minimum germination (0 %) was with leaf extracts, which was equal to the germination of control group (Fig. 1).

In this study, the minimum germination (0 %, equal to the germination of control group) was with leaf extracts) and leaf extract also slowed the germination (MGT) in corn. Similar results have been reported in other crops, the MGT of *Sorghum bicolor* L. was delayed by high concentration of some plant extracts (19). This finding also similar with previous reports in which wheat germination was delayed and germination decreased significantly after treatment with leaf extracts of *Calotropis procera* and *Morettia philaeana* (1). The extracts of several weeds and lemon balm extracts are also reported to show strong inhibitory effects on wheat seed germination (6, 23). Some studies have also reported that germination of some weeds and lentil are inhibited when treated with aqueous extracts of *Rottboellia cochinchinensis* and goosefoot (18, 24). Their results demonstrate that aqueous plant extracts contain allelochemicals that can inhibit germination. However, the germination and MGT of soybean seeds was stimulated after treatment with *P. notoginseng* leaf extract. The results of the present study suggested that *P. notoginseng* contains allelochemicals that promote soybean seed germination while inhibit corn seed germination.

Seedling growth

The effects of *P. notoginseng* extracts on soybean and corn seedling growth depended on the tissue type and concentration of *P. notoginseng* applied.

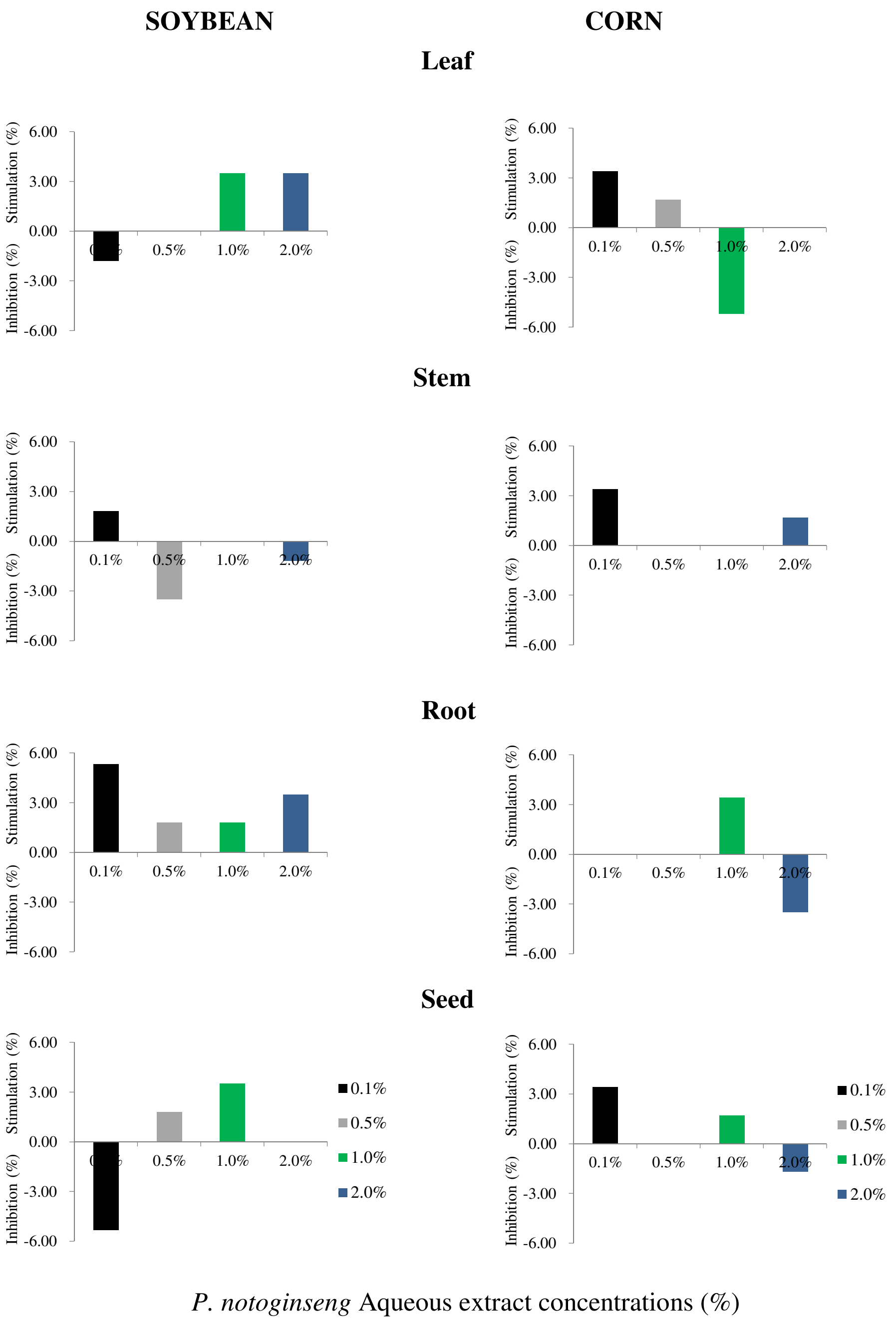

Figure 1. Inhibitory/Stimulatory effects of *P. notoginseng* extracts on germination of soybean and corn.

Comparing the effects of extracts from different tissues on various parameters, leaf extract significantly stimulated ($p < 0.05$) the shoot and root length in soybean than other extracts. With the increasing concentration, the shoot and root length showed significant increase. The increasing concentrations of aqueous leaf extracts resulted in initial increases followed by gradual decreases in shoot and root length in soybean (Fig. 2 and 3). The shoot length was stimulated by 90%, 49%, 50% and 20% with extracts of leaf, stem, root and seed respectively. While the increase in root length was 70%, 29%, 43% and 18% with extracts of leaf, stem, root and seed respectively.

In corn, seed extract showed significant ($p < 0.05$) stimulated the dry weight than other extracts, while leaf extract showed the maximum stimulation in shoot (0.99%) and root length (1.8%) than stem, root and seed extracts. The increasing concentrations of aqueous extracts resulted in initial increases followed by gradual decreases in dry weight in corn (Fig. 4). Most extracts stimulated the dry weight than control, the mean increasing rate of dry weight reached 7.0%, 0.64%, 8.3% and 22% with extracts of leaf, stem, root and seed respectively.

Seedling establishment was stimulated in both soybean and corn after treatment with *P. notoginseng* extracts. Leaf extract significantly promoted shoot and root length of soybean and exhibited the phenomenon of the higher the concentration, the more significant role in promoting; seed extract significantly promoted dry weight of corn. Several reports have demonstrated that extracts from some weeds such as *Ixora* and *Cyperus tuberosus* show stimulatory effects on seedling growth of *Oryza sativa* L. and *Arachis hypogaea* (8,21). The results suggested that the difference in the effect of various extracts is due to the content and concentration of chemicals and *P. notoginseng* leaf and seed may be containing allelochemicals which affect soybean and corn.

The main bioactive components in leaf, stem, root and seed include ginsenosides, notoginsenosides, flavonoids and dencichine. Ginsenosides Rb and Rd, flavonoids and dencichine are the main components reported in leaves and ginsenosides Rg_1, Rg_2, Rb and Rd, notoginsenoside R_1 and R_2, flavonoids and dencichine were the main components in stems (11,28). While, Ginsenosides Rg_1, Rg_2, Rb and Rd, notoginsenosides R_1, R_2, R_3, R_4 and R_6, flavonoids and dencichine are the main components in roots (10,31,32) and ginsenosides Rb and Rd were the main components in seeds (7,28). Ginsenosides Rb and Rd are present in all tissue of *P. notoginseng*. Notoginsenosides R and ginsenosides Rg were present in stems and roots, while ginsenosides Rg were absent in leaves and seeds and notoginsenosides are scarce in leaves and absent in seeds. It suggested that the difference in result of this study maybe due to variability in the components in each tissue of *P. notoginseng*. In this study, the maximum increase in shoot (90%) and root length (70%) of soybean were induced by leaf extract; the largest dry weight of corn was induced by seed extract. All the largest indicators appeared in leaf extract or seed extract. It indicated that notoginsenosides R or ginsenosides Rg may be the allelochemicals that inhibit germination and seedling growth. Further researches should be done to investigate, which components are the allelochemicals.

Response index (RI) of soybean and corn

Leaf extracts proved the most stimulatory to soybean, followed by root, stem and seed extracts (Fig. 5), and the higher the concentration, the more was stimulation. Thus

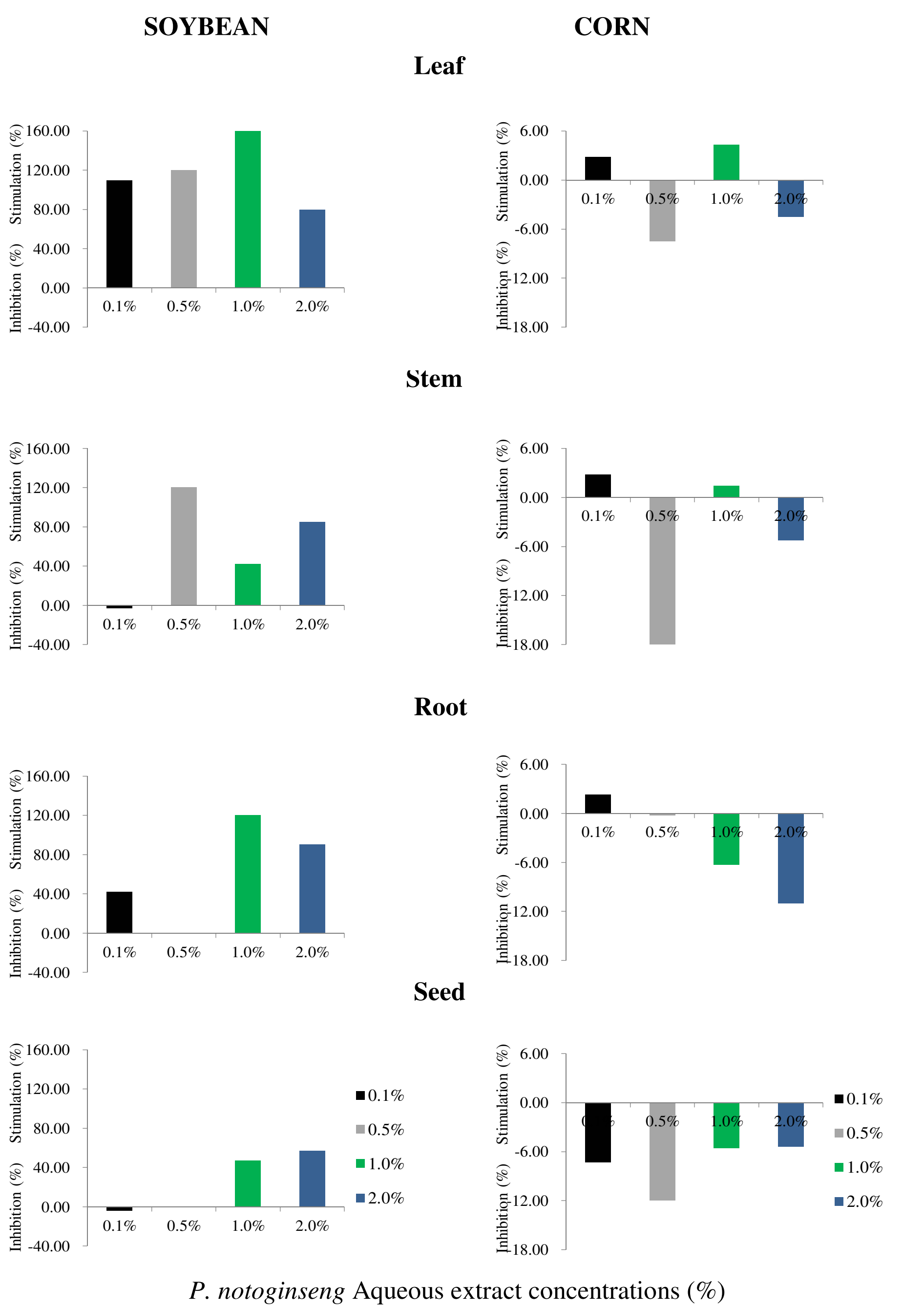

Figure 2. Inhibitory/Stimulatory effects of *P. notoginseng* extracts on shoot length of soybean and corn.

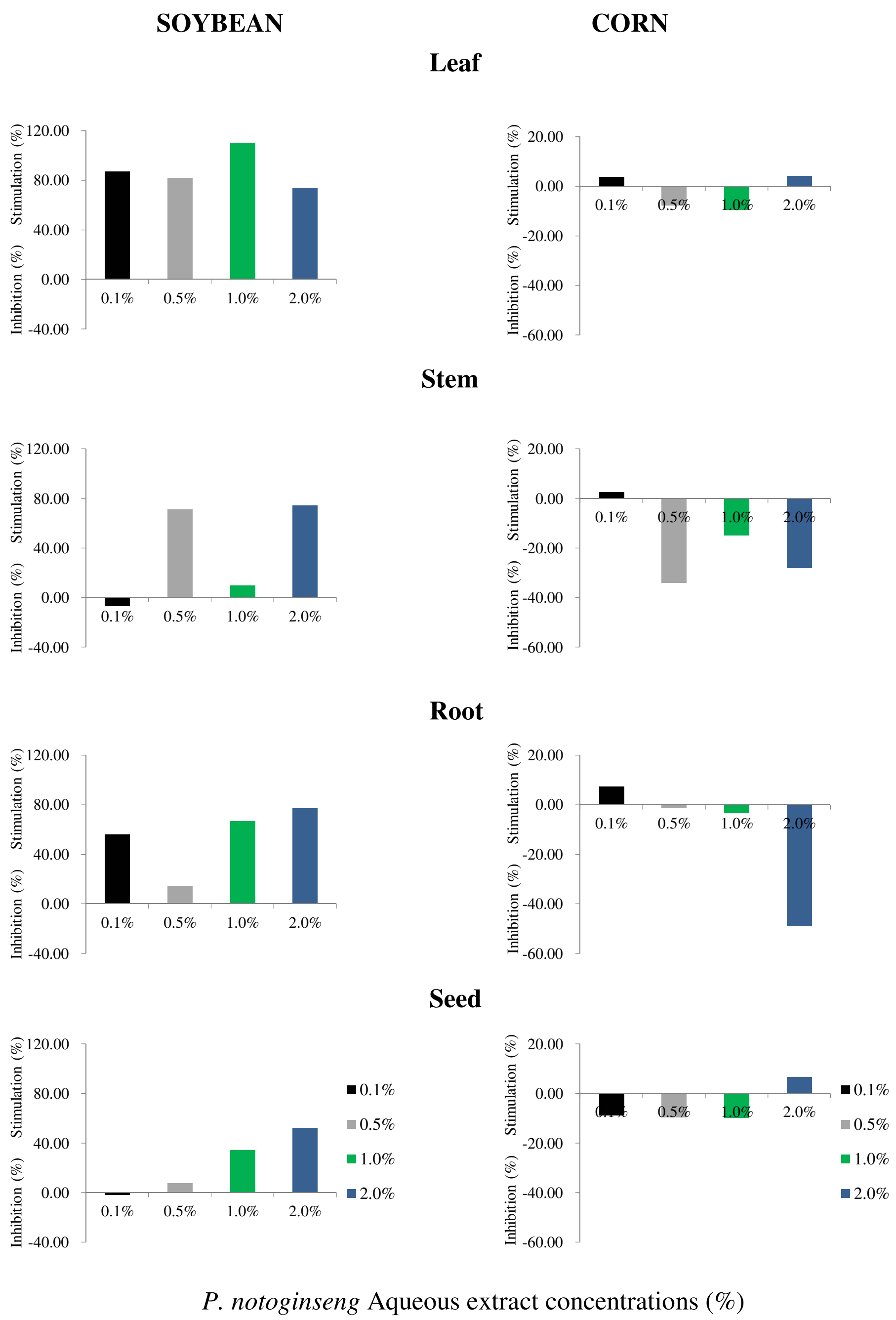

Figure 3. Inhibitory/Stimulatory effects of *P. notoginseng* extracts on root length of soybean and corn.

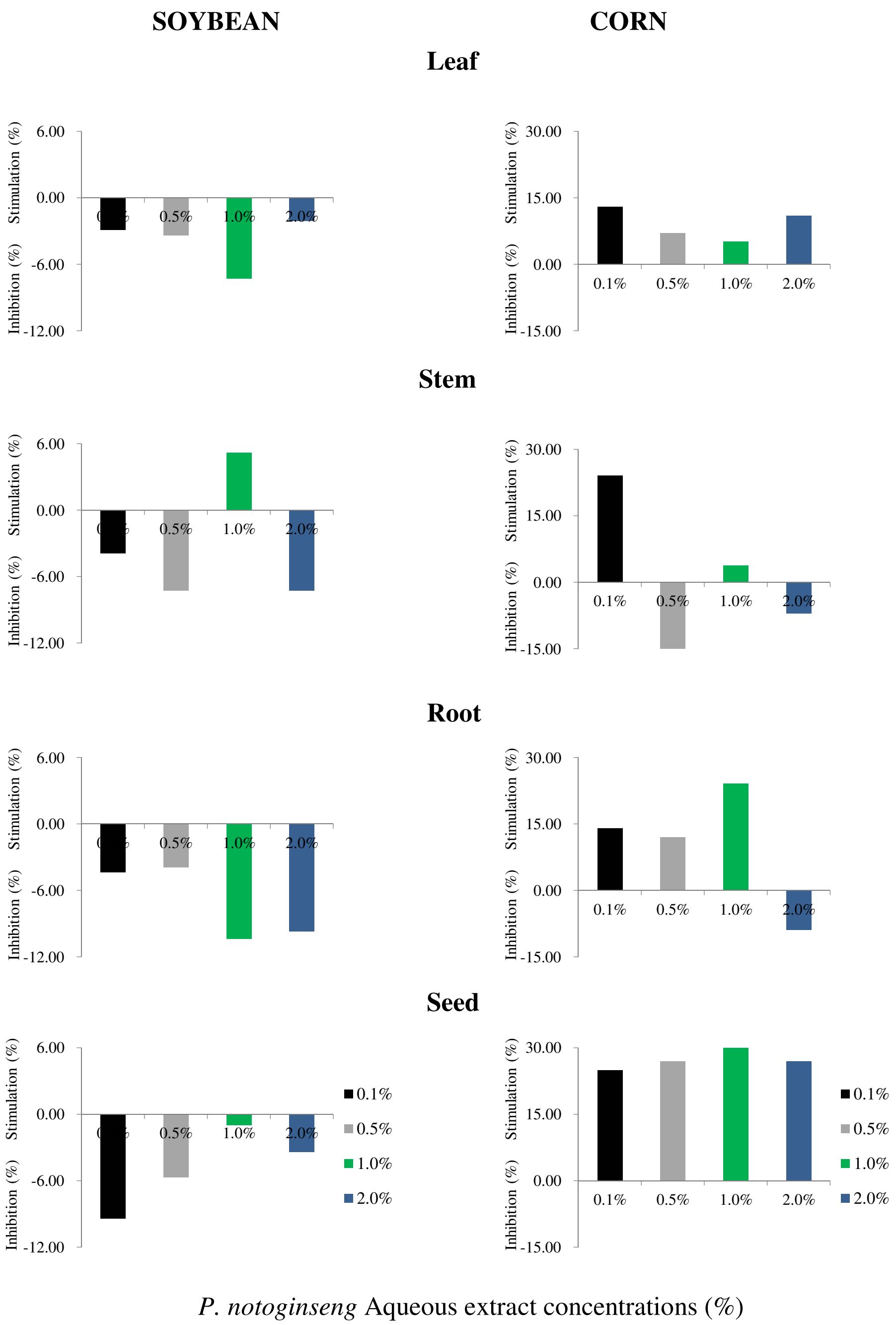

Figure 4. Inhibitory/Stimulatory effects of *P. notoginseng* extracts on dry weight of soybean and corn.

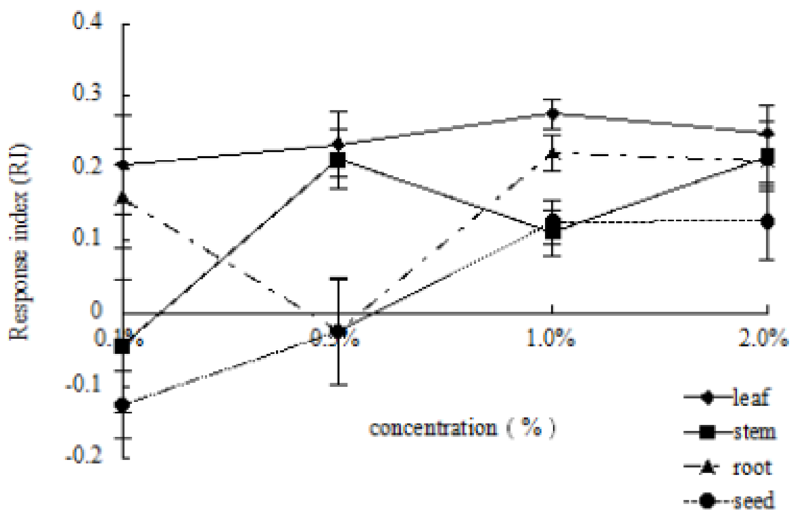

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Figure 5. Allelopathic effects of aqueous extracts of *P. notoginseng* on RI of soybean

Table 1. Effects of *P. notoginseng* extracts on sensitivity of soybean and corn

Crop			MSI_1				MSI_2		MSI_3
	Germi-nation	MGT	Length (cm)		Dry weight (g)		Stage		Crop species
			Shoot	Root	Shoot	Root	Germin-ation	Seedling growth	
Soybean	0.00990	0.0549	0.312	0.274	-0.0548	0.0350	0.0324	0.142	0.0872
Corn	0.00470	-0.0244	-0.044	-0.100	0.0716	0.113	-0.00985	0.0100	0.0000750

MSI: Mean Sensitivity Index

Note: there are 4 parts (leaf, stem, root and seed), and each part has 4 concentrations with three replicates.

n = 48 in MSI_1; n = 48 * 2 = 96 in Seed germination stage in MSI_2 (calculating from germination rate and mean germination time two indicators), n = 48 * 4 = 192 in seedling growth stage (calculating from shoot length, root length, shoot dry weight and root dry weight four indicators); n = 96 + 192 = 288 in MSI_3 (calculating from seed germination and seedling growth of two stages).

allelochemicals released by *P. notoginseng* leaves produce the most stimulatory effects on soybean. The RI and MSI_3 data were both greater than 0 (Table 1); such results confirmed that soybean germination and seedling growth were enhanced by *P. notoginseng*.

However, in corn, the MSI_2 in seed germination stage was less than 0 while the MSI_2 in seedling growth stage was greater than 0 (Table 1). Thus *P. notoginseng* extracts were inhibitory to corn during the seed germination stage, but were stimulatory in seedling growth stage.

The MSI_R in this study indicates that *P. notoginseng* promotes germination and seedling growth and shows overall stimulatory effects on soybean. In conclusion, *P. notoginseng* extracts exert stimulatory effects on soybean. Thus, planting crops such as soybean instead of corn is a practical approach to avoid inhibition in *P. notoginseng* cropped soil. Allelochemicals appear to exert important effects on crop growth and production yield. Further studies must be carried out to investigate the effect of *P. notoginseng* on other crop species in field experiments.

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