***Pak. J. Bot.*, 45(1): 225-233, 2013.**

ALLELOPATHIC EFFECT OF DECOMPOSED GARLIC (*ALLIUM SATIVUM* L.) STALK ON LETTUCE (*L. SATIVA* VAR. *CRISPA* L.)

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Abstract

Garlic (*Allium sativum* L.) stalk is a byproduct and normally the waste of garlic production but now is thought a good biological resource. It is necessary to utilize this resource efficiently and reasonably to reduce environment pollution and achieve agricultural sustainable development. The allelopathy of decomposed garlic stalk at different ratios (0:100, 1:100, 3:100 and 5:100) was investigated in this study using lettuce (*L. sativa* var. *crispa* L.) as the allelopathic test plant. The results are as follows: (1) Lower concentration of decomposed garlic stalk promoted lettuce plant growth, but higher concentration inhibited growth. (2) The highest ratio of stalk to soil (5:100) significantly increased the root length and fresh weight of lettuce but decreased the shoot length and fresh weight. (3) Lower ratio of stalk to soil significantly increased the content of protein and chlorophyll and the activity of plant enzymes. (4) Increasing the concentration of decomposed garlic stalk increased the activity of soil urease and sucrase. It was concluded that the decomposed garlic stalk with lower concentration could promote lettuce plant growth. This result provides a scientific basis for allelopathic study and application of decomposed garlic stalk.

Introduction

Allelopathy is a natural mechanism in which plants (including microorganisms) influence the growth and development of each other through the release of allelochemicals into the environment (Rice 1984). Allelopathic influence of a plant may result in positive outcomes, but generally it is considered to have negative effects on the other plant (Elijarrat & Barcelo, 2001). Allelopathy plays an important role in interaction among different plant species and in developing specific plant communities. Plant species are in a very complex and multi-directional relation with their neighboring plant species; including competition for nutrient and water, inhibition, stimulation and interdependence (Zeng, 2008). Many higher plant species contain chemicals with an allelopathic activity in different structural organs (Inderjit, 1996; Duck *et al.*, 2000). Under certain circumstance, these allelochemicals are released into environment, either as exudates from living plants or through decomposition of plant residues in sufficient amount in order to affect the neighboring or successional plants (Rice, 1984; Dayan *et al.*, 2000; Einhelling, 2004). Some researches studied that aqeuous of all parts of Lantana camara had strong allelopathy on the growth of various test species (Hussain *et al*., 2011) and the rain leachates of some plants had differential allelopathic effect on various bioasssys (Hussain *et al*., 2011).

Along with the social development and progress of human civilization, the overall cropping system and agricultural approach has drastically changed over the period of time. The usage of crop waste material is becoming more attractive and adoptable because of its economical and crop improvement abilities. Crop waste material also relates to the allelophatic effect of a plant either through its beneficial or inhibitory impact on other plants. Cover crops are considered to highly suitable in such an integrated approach, as they can supplement the agro-ecosystem with many additional benefits, such as improved soil properties, promoted nutrient cycling and managed the pest (Sarrantonio & Gallandt, 2003). Effects of litter are complicated and output of them lies on a delicate balance between promotive and inhibitory effects (Holmgren *et al.*, 1997). The decomposed products of crop residues could promote or inhibit the growth of plants, but the promotion of growth may appear in the later phase of decomposition (Bonanomi *et al.*, 2006). In addition to determining the recognized role of crop residues as a nutrient source (Vitousek & Sanford, 1986), many research studying in natural ecosystems (Meier *et al.*, 2009), laboratory (Hodge *et al.*, 1998) and different farming systems (Putnam, 1994) have indicated that the additives of plant residues and other types of organic matter to the soil may decrease plant growth. About the organic matter applied in the agriculture, Bonanomi *et al*., (2007) indicated that plant litter as the soil amendment decreased the plant growth in their 119 of 1728 experiments (6.9%). Moreover, some allelochemicals released during plant residues decomposition (Bonanomi *et al.*, 2006). Nevertheless, just single compound hardly explained all the inhibitory role of plant residues during the decomposed process (An *et al.*, 2001).

The ecological strategy of plant species was determined by functional traits which response to ground conditions and influence on various ecosystems (Lavorel & Garnier, 2002; Garnier *et al.*, 2007; Suding *et al.*, 2008). The study of functional traits therefore plays an important role in exploring the underlying queries regarding physiology and ecology of plant residue decomposition. Moreover, use of various vegetative residues to amend soil could improve soil biological environment and plant growth situation (McSorley & Gallaher, 1994; Ilieva Makulec *et al.*, 2006). Cover crops during the fallow period in degraded and unfertile fields could reduce nutrient losses, protect the soil, improve biological environments and enhance crop productivity (Guo *et al.*, 2008; Tian *et al.*, 2010; DuPont *et al.*, 2009). The previous studies researched that the crop residues influenced soil nitrogen dynamics and insured the availability of inorganic nitrogen in intensive production systems (Williams & Weil, 2004; Guo *et al.*, 2008).

Garlic (*Allium sativum* L.), a species of *Allium* genus, is one of the major vegetable and medicine plant used around the world. It's not only rich in nutrient substance, but also possesses a natural strength of combating against different bacterial and other microbial disease infections, thus known as a generally good previous crop. Furthermore, stalk of garlic is a byproduct of garlic production but holds a great strength to be used as a good biological resource. However, it is abandoned or burned as waste product in the current garlic cropping production due to ignorance on its importance. As a result, it not only greatly reduces the utilization rate of garlic stalk, but also causes environmental pollution. It is necessary to utilize this resource efficiently and reasonably to reduce environmental pollution and achieve agricultural sustainable development.

Up to now, only few studies on allelopathy of garlic stalk have been found yet but focus on ultrasonic extract of garlic plant (Wang *et al.*, 2009) and garlic straw aqueous extracts (Wei *et al.*, 2008). Thus the overall objective of this research was to investigate the allelopathic effect of different concentrations of garlic stalk decomposition. In this context, the functional traits and plant enzyme activities of lettuce plants as test plant, and enzyme activities of the soil with decomposed garlic stalk were investigated.

Materials and Methods

Site description and experimental design: The experimental field was located in a plastic tunnel at the experimental station (34° 16' N, 108° 4' W) of College of Horticulture, Northwest A & F University, Yangling, Shannxi Province, China. The original soil of the experiment had a pH value (1:1 water) of 7.93, an electrical conductivity (EC) value with the extracting ratio of 1:5 (soil/water) of 199.7 $\mu S{\cdot}cm^{-1}$, available nitrogen (N) content of 60.67 $mg{\cdot}kg^{-1}$, available phosphorus (P) content of 14.77 $mg{\cdot}kg^{-1}$ and available potassium (K) content of 200.90 $mg{\cdot}kg^{-1}$.

Garlic (*Allium sativum* L. cv. G064) stalk was collected from garlic production field in June 2010 after garlic bulb harvest, then dried under field natural conditions and shattered into powder using grinder, subsequently stored in darkness at room temperature until use. It was mixed with soil at three concentrations (1, 3 or 5 g dry weight per 100 g soil) and filled into pots (size 15 cm × 15 cm × 15 cm). Blank control pots were filled with the same soil without garlic stalk. There were three replications and totally 240 pots arranged on a randomized block design and placed in a plastic tunnel. The decomposing process took naturally in 30 days starting from February 2011. Enough water was added to each pot to keep the soil water content not lower than 40% throughout the decomposition periods. After decomposing of garlic stalk, ten seeds of lettuce (*L.sativa* var.*crispa* L., Hongkong Glass Lettuce) were sown separately in each pot and watered regularly during lettuce growth. Meanwhile, each pot was applied 2.5g compound fertilizer (Sakefu, Qinhuangdao, Hebei Province, China) with 7 days interval during whole growth period of lettuce plant. Lettuce plants and soil of each treatment were sampled on 30, 35, 40, 45, 50 and 55 days after sowing, respectively. The plants samples were rinsed with distill water and dried with absorbent paper. Five lettuce plants were sampled randomly from each treatment to measure the morphologic traits. Part of the fresh leaves was used to measure chlorophyll content, and the remaining leaf was freezed in liquid nitrogen for several minutes and then stored at -70°C refrigerator for measuring physiological indices and various enzyme activities. Moreover, the soil samples from different pots of each treatment were homogenized, dried at dark room and sieved through a 1 mm sieve to determine the soil enzyme activity.

Measurement of lettuce plant morphologic traits: The actual length of lettuce shoot and root was measured in laboratory using a straightedge with the readings by estimating the nearest 0.01 cm. And the fresh weight of lettuce shoot and root was measured using an electronic balance (precision 0.001 g).

Determination of lettuce leaf physiological indices and enzyme activities: Chlorophyll content was analyzed as per Ranganna (1986) methods but with little modification. The fresh leaf sample (0.1g) from each treatment was homogenized and extracted with 10ml of 80% acetone over 24 hours until the leaves turned white completely. Afterwards, the concentration of chlorophyll a and b were determined using the extracting solution at 663nm and 645nm wavelength through the spectrophotometer (UV-3802, UNIC, Shanghai, China), respectively.

Ca (mg/L) = 12.7 x D663 – 2.59 x D645
Ca (mg/L) = 22.9 x D645 – 4.67 x D663
Ca (mg/L) = Ca + Cb = 20.3 x D645 + 8.03 x D663
Chlorophyl 1 content (mg/g) = C(mg/L) x total content of extract solution (ml) x dilution factor x 1000/fresh weight of leaf (g)

Enzyme solution was extracted according to Guo *et al.,* (2004) with slight modification. The frozen lettuce leaves from each treatment were homogenized in a pestle and mortar with 0.05M sodium phosphate buffer (pH 7.8). The homogenate was centrifuged at 10000 r/min for 20 min and the supernatant was used for analyzing of soluble proteins content, malonaldehyde (MDA) content, superoxide dismutase (SOD, E.C. 1.15.1.1) activity, peroxidases (POD, E.C. 1.15.1.7) activity, catalase (CAT, E.C. 1.15.1.6) activity, polyphenol oxidase (PPO, EC 1.10.3.1) activity, and L-phenylalanin ammo-nialyase (PAL, E.C.4.3.1.5). The above steps were carried out at the condition of 4°C.

All the physiological indices and enzyme activities were determined as per the methods of Gao (2000). Lipid peroxidation was expressed as the MDA content in μmol per g FW. One unit of SOD activity was defined as the quantity of SOD required producing a 100% reduction of nitroblue tetrazolium (NBT) under experimental conditions and the specific enzyme activity was expressed as units per g FW of leaves. POD, CAT and PPO activity were expressed as absorbance changes at 470 nm, 240 nm and 410 nm per minute per g FW, respectively. PAL activity was defined as absorbance changes at 290 nm per hour per g FW.

Determination of soil enzyme activities: The sucrase and urease activity were assayed on the basis of amount of release and quantitative determination of glucose and NH_3-N products. Soil samples were incubated with 8% sucrose solution and 10% urea solution in suitable buffer solution for 24 hours at 37°C and measured at 508nm and 578nm using spectrophotometry (Tabatabai 1994; Guan 1986). Catalase was measured by ultraviolet spectrophotometry (Yang *et al.*, 2011).

Statistical analysis: The data was statistically assessed by one-way analysis of variance (ANOVA) using SPSS 17.0 software package. Mean separations were performed by Duncan's multiple range tests. Differences at $p = 0.05$ were considered significant.

Results

Effect of decomposed garlic stalk on morphologic traits of lettuce plant: Influence of decomposed garlic stalk on the shoot length (Table 1a) and root length (Table 1b) of lettuce plants was presented in Table 1. The lower concentrations (1:100 and 3:100) of decomposed garlic stalk significantly increased the length of lettuce shoot as compared to the control treatment (0:100), whereas, higher concentration (5:100) of decomposed garlic stalk significantly decreased the length of shoot. Furthermore, root length of lettuce plants was highly promoted by almost all treatments of different concentrations; especially concentration (3:100) held the strongest impact.

The allelopathic effect of decomposed garlic stalk was tested with shoot and root fresh weight of lettuce plant (Table 2a & 2b). Lower concentrations (1:100 and 3:100) of decomposed garlic stalk had significantly promoted the fresh weight of lettuce shoot, whereas an inhibiting effect on fresh weight was observed on application of higher concentration (5:100) of decomposed garlic, as compared with the control (0:100). Additionally, ratio of garlic stalk to soil (3:100) showed the most positive impact on fresh weight of lettuce shoot. Nevertheless, all the treatments of different ratios of garlic stalk to soil significantly promoted fresh weight of lettuce root, except 5:100 ration treatment held only slight promotion on 30 days after sowing.

Table 1a. Effect of different concentrations of decomposed garlic stalk on length of lettuce shoot.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	5.04 ± 0.06 b	5.26 ± 0.07 b	6.02 ± 0.10 c	6.32 ± 0.08 c	8.32 ± 0.09 c	10.50 ± 0.18 c
1 : 100	5.98 ± 0.20 a	5.46 ± 0.09 b	7.24 ± 0.10 b	7.26 ± 0.09 b	9.52 ± 0.07 b	11.46 ± 0.06 b
3 : 100	5.36 ± 0.07 b	6.54 ± 0.09 a	8.14 ± 0.17 a	8.80 ± 0.12 a	10.00 ± 0.20 a	13.36 ± 0.11 a
5 : 100	3.86 ± 0.10 c	4.03 ± 0.12 c	5.94 ± 0.09 c	6.12 ± 0.14 c	7.42 ± 0.13 d	7.46 ± 0.06 d

The data present as the shoot length (cm) of lettuce plants and mean ± standard error. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Table 1b. Effect of different concentrations of decomposed garlic stalk on length of lettuce root.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	4.38 ± 0.26 c	5.26 ± 0.10 d	8.62 ± 0.10 c	8.74 ± 0.11 c	8.84 ± 0.09 d	9.08 ± 0.08 d
1 : 100	5.88 ± 0.15 b	9.18 ± 0.10 b	9.56 ± 0.15 a	10.17 ± 0.08 b	11.62 ± 0.11 b	13.74 ± 0.07 b
3 : 100	7.84 ± 0.09 a	9.94 ± 0.10 a	9.60 ± 0.09 a	10.78 ± 0.12 a	12.48 ± 0.08 a	16.72 ± 0.08 a
5 : 100	4.48 ± 0.11 c	7.12 ± 0.10 c	9.10 ± 0.07 b	10.02 ± 0.08 b	10.36 ± 0.14 c	11.82 ± 0.13 c

The data present as the root length (cm) of lettuce and mean ± standard error. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Table 2a. Effect of different concentrations of decomposed garlic stalk on fresh weight of lettuce shoot.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	0.283 ± 0.002 b	0.359 ± 0.005 c	0.582 ± 0.006 b	0.899 ± 0.004 c	1.563 ± 0.005 c	2.901 ± 0.002 c
1 : 100	0.384 ± 0.006 a	0.448 ± 0.002 b	0.590 ± 0.006 b	1.047 ± 0.001 b	2.543 ± 0.004 b	3.364 ± 0.002 b
3 : 100	0.289 ± 0.003 b	0.528 ± 0.007 a	1.136 ± 0.005 a	2.143 ± 0.002 a	3.903 ± 0.005 a	5.024 ± 0.003 a
5 : 100	0.133 ± 0.003 c	0.194 ± 0.004 d	0.529 ± 0.003 c	0.629 ± 0.002 d	1.246 ± 0.001 d	1.649 ± 0.001 d

The data present as the seedling fresh weight (g) of lettuce and mean ± standard deviation. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Table 2b. Effect of different concentrations of decomposed garlic stalk on fresh weight of lettuce root.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	0.025 ± 0.002 c	0.016 ± 0.001 d	0.049 ± 0.002 d	0.071 ± 0.002 d	0.207 ± 0.001 d	0.223 ± 0.003 d
1 : 100	0.033 ± 0.002 b	0.052 ± 0.001 b	0.104 ± 0.002 b	0.140 ± 0.005 b	0.330 ± 0.003 b	0.416 ± 0.002 b
3 : 100	0.071 ± 0.004 a	0.075 ± 0.001 a	0.119 ± 0.001 a	0.234 ± 0.002 a	0.631 ± 0.003 a	0.718 ± 0.002 a
5 : 100	0.028 ± 0.001 c	0.028 ± 0.002 c	0.057 ± 0.002 c	0.118 ± 0.003 c	0.241 ± 0.002 c	0.343 ± 0.002 c

The data present as the root fresh weight (g) of lettuce and mean ± standard deviation. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Effect of decomposed garlic stalk on lettuce leaf physiological indices and enzyme activities: The test results were summarized in Table 3 for the content of chlorophyll a (Table 3a), chlorophyll b (Table 3b) and total chlorophyll (Table 3c). Garlic stalk to soil ratio of 3:100 significantly increased chlorophyll a content in lettuce leaves during the growth period, in comparison to control plants. However, the concentration 5:100 of garlic stalk to soil significantly decreased chlorophyll a content in lettuce leaves on 30th, 35th and 55th day of sowing, respectively. It was worth to mention that garlic stalk to soil concentration of 5:100 showed no major impact on chlorophyll a content during other growing stages after sowing.

Chlorophyll b content in lettuce leaves was drastically increased by treatment having garlic stalk to soil concentration of 3:100 during 30 to 45 days after sowing, but this enhancing effect was not noticed between 50 to 55 days of sowing time. Furthermore, garlic stalk to soil concentration of 5:100 significantly decreased the chlorophyll b content on 30th, 35th and 50th day of sowing, respectively, while showed no radical effect on 40th and 45th day of sowing. Similarly, treatment with 5:100 ratio of garlic stalk to soil significantly increased chlorophyll b content in lettuce leaves on 55th day of sowing.

Garlic stalk to soil ratio of 3:100 significantly increased the content of total chlorophyll during the whole growth period of lettuce plants. And ratio of 1:100 garlic stalk to soil significantly increased the total chlorophyll content on 30, 35 and 50 days after sowing of lettuce, respectively. Moreover, a significantly decreasing effect on total chlorophyll content was observed from 5:100 ratio of garlic stalk to soil treatment during the whole growth period of lettuce plants, except for 55 days after sowing.

Table 3a. Effect of different concentrations of decomposed garlic stalk on content of chlorophyll a in lettuce leaf.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	0.408±0.012 c	0.512±0.011 b	0.479±0.008b c	0.578±0.006 bc	0.516±0.007 b	0.486±0.018 b
1 : 100	0.155±0.007 b	0.442±0.018 a	0.515±0.004 b	0.593±0.012 b	0.532±0.020 b	0.515±0.009 ab
3 : 100	0.510±0.017 a	0.532±0.010 a	0.606±0.030 a	0.645±0.007 a	0.637±0.014 a	0.551±0.012 a
5 : 100	0.224±0.004 d	0.360±0.005 c	0.433±0.008 c	0.555±0.011 c	0.511±0.006 b	0.111±0.005 c

The data present as content of chlorophyll a ($mg \cdot g^{-1}$) in lettuce leaf and mean ± standard error. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Table 3b. Effect of different concentrations of decomposed garlic stalk on content of chlorophyll b in lettuce leaf.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	0.122±0.003 c	0.131±0.007 b	0.150±0.002 b	0.188±0.008 b	0.214±0.005 a	0.155±0.014 b
1 : 100	0.232±0.005 b	0.151±0.001 b	0.231±0.003 a	0.194±0.008 ab	0.219±0.016 a	0.150±0.007 b
3 : 100	0.271±0.008 a	0.264±0.011 a	0.236±0.020 a	0.217±0.007 a	0.200±0.005 a	0.173±0.011 b
5 : 100	0.054±0.016 d	0.070±0.016 c	0.137±0.003 b	0.177±0.006 b	0.156±0.003 b	0.525±0.002 a

The data present as content of chlorophyll b ($mg \cdot g^{-1}$) in lettuce leaf and mean ± standard error. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Table 3c. Effect of different concentrations of decomposed garlic stalk on content of total chlorophyll in lettuce leaf.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	0.346±0.010 c	0.573±0.012 c	0.629±0.009 c	0.762±0.013 b	0.730±0.010 b	0.641±0.005 b
1 : 100	0.640±0.012 b	0.663±0.018 b	0.746±0.007 b	0.766±0.007 b	0.751±0.006 b	0.665±0.010 b
3 : 100	0.781±0.010 a	0.796±0.017 a	0.842±0.011 a	0.862±0.010 a	0.837±0.010 a	0.724±0.016 a
5 : 100	0.209±0.020 d	0.430±0.012 d	0.570±0.006 d	0.767±0.006 c	0.667±0.005 c	0.636±0.006 b

The data present as content of total chlorophyll (mg·g^{-1}) in lettuce leaf and mean ± standard error. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Effect of the decomposed garlic stalk on protein content (Fig. 1a) and PAL activity (Fig. 1b) of lettuce leaf varied with the variant concentrations of garlic stalk to soil used. At the lower concentration ratios (1:100 and 3:100), the decomposed garlic stalk had significant promotion effect on protein content and PAL activity, whereas at higher concentration ratio (5:100), significant inhibition in protein content and PAL activity was recorded, as compared to control (0:100). At the ratio of 1:100, the decomposed garlic stalk had the strongest promoting effect on protein content and PAL activity.

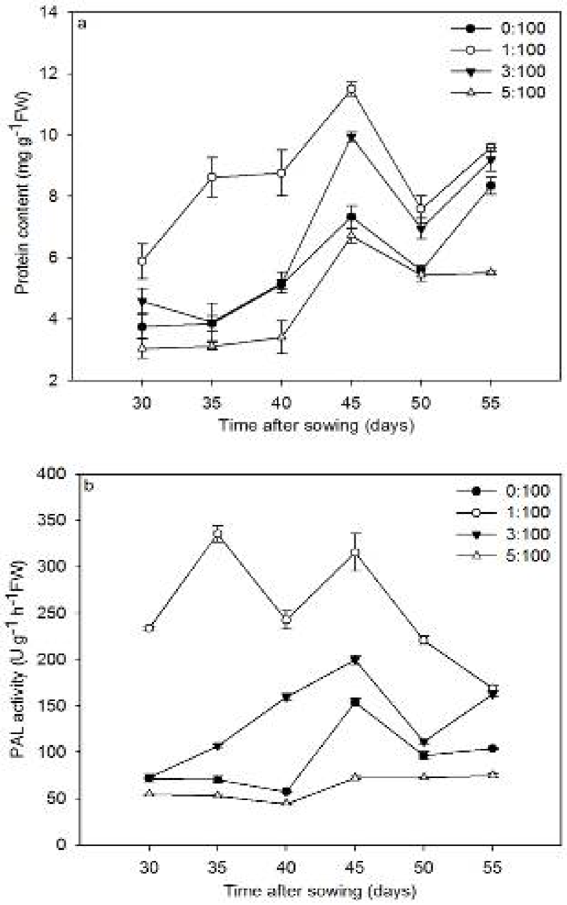

Fig. 1. Effect of different concentrations of decomposed garlic stalk on protein content (a) and PAL activity (b) in lettuce leaf. Error bars present as the standard error of the mean.

Activities of CAT, POD and SOD in lettuce leaves were significantly affected by decomposed garlic stalk at lower concentrations (Fig. 2a, b and c). At the ratio of 3:100, the decomposed garlic stalk had the greatest effect on activity of CAT, POD and SOD, compared with the control. Whereas at higher concentration ratio (5:100), it significantly inhibited the activity of CAT, POD and SOD.

Both PPO activity (Fig. 3a) and MDA content in lettuce leaf (Fig. 3b) decreased with the increasing concentration of decomposed garlic stalk, and the highest values were found in control (0:100) treatment during the whole growth period of lettuce, except for MDA content at garlic stalk to soil ratio of 1:100 on 50 days after lettuce sowing. Additionally, the decomposed garlic stalk at 5:100 ratio prominently decreased the PPO activity and MDA content of lettuce leaves, compared with the control (0:100).

Effect of decomposed garlic stalk on soil enzyme activities: Decomposed garlic stalk showed no significant effect on soil catalase activity on 40 and 50 days after sowing lettuce (Table 4). However, soil catalase activity was significantly inhibited by 1:100 concentration of garlic stalk to soil on 30th and 50th day of sowing. Similarly garlic stalk to soil concentration of 5:100 significantly promoted the soil catalase activity on 50 and 55 days after sowing.

The soil urease activity (Fig. 4a) and sucrase activity (Fig. 4b) were increased with increasing concentration of decomposed garlic stalk. The activity of both urease and sucrase in soil during the whole growth period of lettuce was significantly promoted for all the three treatments (1:100, 3:100 and 5:100) as compared with control and the treatment 5:100 presented the best impact.

Discussion

The results of this research showed that the decomposed garlic stalk at lower concentrations significantly increased the shoot length of lettuce, but the higher concentration significantly decreased the shoot length. Obviously, the degree of inhibition increased as the concentration of garlic stalk increasing. A number of previous studies suggested that the stronger inhibition was observed with the increased concentrations of extracts (Chung & Miller, 1995; Laosinwattana *et al.*, 2007, 2009). However, all concentrations of decomposed garlic stalk in this study markedly promoted the root length of lettuce plants. These results do not agree with earlier research which suggested that root length was more sensitive to allelopathic inhibition of the 0.15 g·ml^{-1} extracts than shoot length (Li *et al.*, 2011), and Noumi & Chaieb (2011) studied that different extracts significantly reduced the seedling growth of the tested species. The reason may be that allelopathy of plant extracts depends on the kind of tissues from which these allelochemicals originate, the concentrations of allelochemicals and the target species (Wu *et al.*, 2009; Mutlu & Atici, 2009). But the findings are in agreement with some of the earlier studies which indicated that water extracts of allelopathic plants causes more pronounced effects on root growth than on shoot growth (Batish *et al.*, 2006; Siddiqui, 2007; Han *et al.*, 2008). Such a result presented might be because that roots are first to absorb the allelochemicals from environment (Turk & Tawaha, 2002). All data on shoot and root length of lettuce indicated that the garlic stalk could release allelochemicals into environment during decomposed period; consequently, it may play an important allelopathy on test plants. Nevertheless, whether or not the higher concentrations of decomposed garlic stalk have promotion effect will be further studied.

A radically enhancing influence of decomposed garlic stalk on physiological indices and plant enzyme activities in lettuce leaves were observed during the study. Chlorophyll content, CAT, POD, and SOD activity were the highest sensitive to ratio of garlic stalk to soil of 3:100. In addition, protein content and PAL activity had the greatest response to the 1:100 ratio of garlic stalk to soil. However, all concentration of garlic stalk decomposition markedly dropped PPO activity and MDA content in lettuce leaf. These findings imply the diversified and variant reaction of physiological indices and plant enzyme activities in lettuce leaves to the decomposed garlic stalk.

Soil enzyme activities are considered as indicators for potential nutrient cycling processes and fertility management, particularly in long-term organic and conventional farming system. Our results indicated that all the ratios of garlic stalk to soil (1:100, 3:100 and 5:100) significantly enhanced both the soil urease and sucrase activity during the whole growth period of lettuce plants. Gu (2009) illustrated that the urease activity of paddy soil was positively influenced by allelopathic rice variety through the release of allelochemicals, but the action only appeared at early growth phases. It suggests that allelochemicals of different plants may have different effective time of action on the test plant.

In general, the decomposed garlic stalk showed marketable allelopathic effect on growth of lettuce plant. These findings provide evidence that the decomposed garlic stalk may contain some allelochemicals and causes allelopathy through releasing the allelochemicals to environment. Furthermore, lettuce plants normally are regarded as model plant to exam the allelopathy of other plant species. So, it is certainly inferred that the decomposed garlic stalk may have allelopathic effect on the great major of vegetable plants from the present study. Nevertheless, further investigation is suggested for exploring the allelopathic effect of decomposed garlic stalk on other crops and weeds.

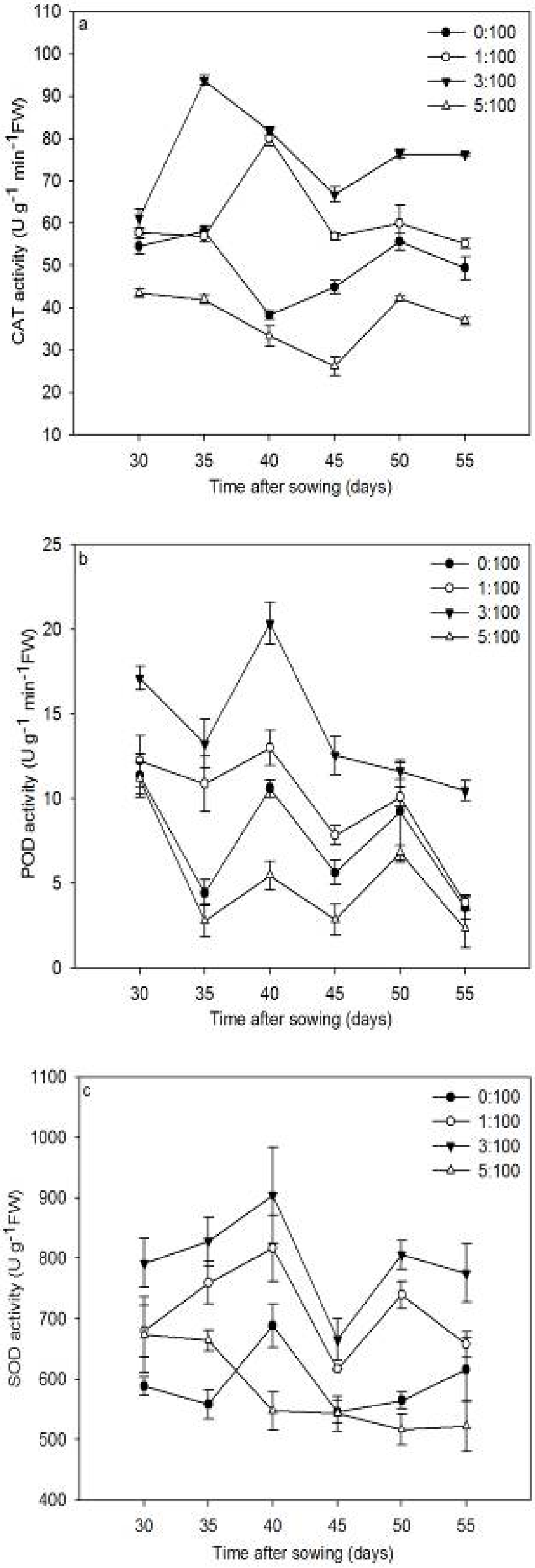

Fig. 2. Effect of different concentrations of decomposed garlic stalk on CAT activity (a), POD activity (b) and SOD activity (c) in lettuce leaf. Error bars present as the standard error of the mean.

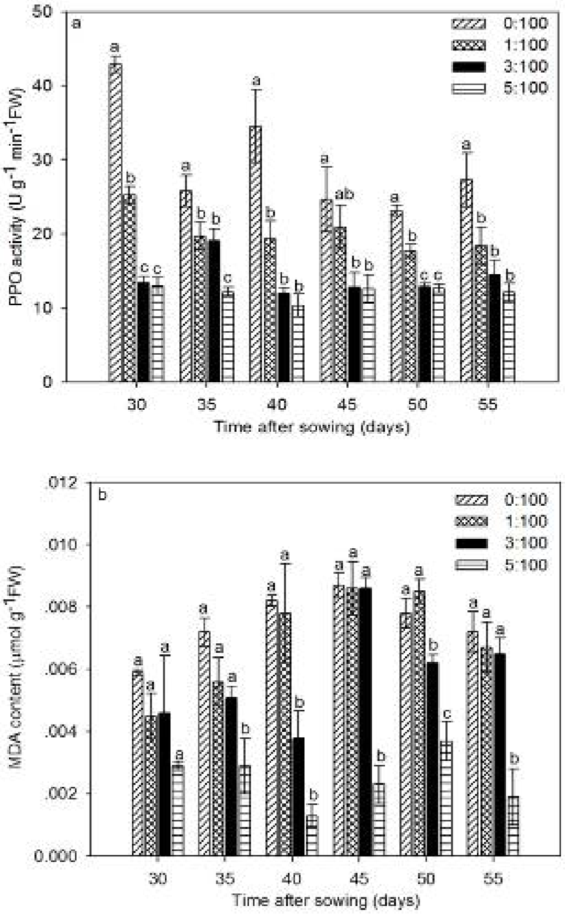

Fig. 3. Effect of different concentrations of decomposed garlic stalk on PPO activity (a) and MDA content (b) in lettuce leaf. Error bars present as the standard error of the mean. Different letters above the histograms indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3.

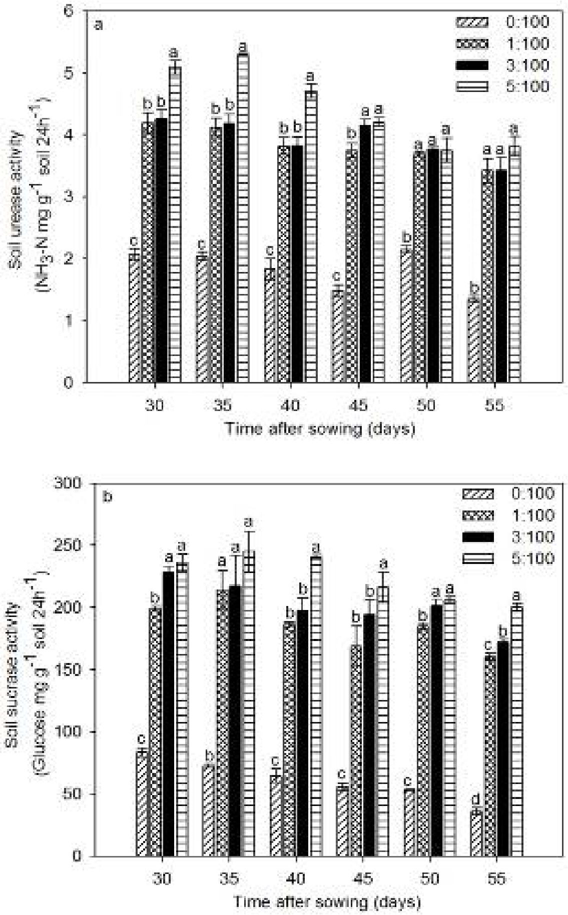

Fig. 4. Effect of different concentrations of decomposed garlic stalk on soil urease activity (a) and sucrase activity (b). Error bars present as the standard error of the mean. Different letters above the histograms indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3.

Table 4. Effect of different concentrations of decomposed garlic stalk on soil catalase activity.

Ratio of garlic stalk to soil	Time after sowing (days)					
	30	35	40	45	50	55
0 : 100	0.0957±0.0015 a	0.0975±0.0004 b	0.0954±0.0020 a	0.0918±0.0023 a	0.0906±0.0008 a	0.0957±0.0020 a
1 : 100	0.0868±0.0029 b	0.0842±0.0030 c	0.0955±0.0021 a	0.0831±0.0009 a	0.0891±0.0026 ab	0.0839±0.0017 c
3 : 100	0.0929±0.0010 ab	0.1372±0.0018 a	0.0868±0.0054 a	0.0874±0.0044 a	0.0906±0.0015 a	0.0883±0.0012 bc
5 : 100	0.0937±0.0028 ab	0.0925±.0025 b	0.0916±0.0024 a	0.0926±0.0021 a	0.0836±0.0018 b	0.0928±0.0029 ab

The data present as soil catalase activity (H_2O_2 mg·g^{-1} soil 20min^{-1}) and mean ± standard error. Different letters in the same column indicate significant differences at 0.05 level (ANOVA and Duncan's multiple range test) n=3

Conclusions

The results of this research showed that the decomposed garlic stalk at lower concentrations significantly increased the growth of lettuce and promoted physiological indices and activities of plant enzymes in lettuce leaf. In addition, the results indicated that all the ratios of garlic stalk to soil (1:100, 3:100 and 5:100) significantly promoted both the soil urease and sucrase activity during the whole growth period of lettuce plants.

Acknowledgements

This research was supported by the project of State Commonwealth (Agriculture) Scientific Research (No.200903018) and the project of State Natural Science Foundation (No.31171949). I thank Dr. Cuinan Wu and Muhammad Alikhan for reading the manuscript prior to submission.

References

An, M., J.E. Pratley and T. Haig. 2001. Phytotoxicity of Vulpia residues: IV. Dynamics of allelochemicals during decomposition of *Vulpia* residues and their corresponding phytotoxicity. *J. Chem. Ecol.*, 27(2): 395-409.

Batish D.R., M. Kaur, H.P. Singh and R.K. Kohli. 2006. Phytotoxicity of a medicinal plant, *Anisomeles indica*, against *Phalaris minor* and its potential use as natural herbicide in wheat fields. *Crop Prot.*, 26: 948-952.

Bonanomi G., M.G. Sicurezza, S. Caporaso, A. Esposito and S. Mazzoleni. 2006. Phytotoxicity dynamics of decaying plant materials. *New Phytol.*, 169: 571-578.

Bonanomi G., V. Antignani, C. Pane and F. Scala. 2007. Suppression of soilborne fungal diseases with organic amendments. *J. Plant Pathol.*, 89: 311-340.

Chung I.M. and D.A. Miller. 1995. Natural herbicide potential of alfalfa residue on selected weed species. *Agron. J.*, 87: 920-925.

Dayan F.E., J.G. Romagni and S.O. Duke. 2000. Investigating the mode of action of natural phytotoxins. *J. Chem. Ecol.*, 26: 2079-94.

Duke, S.O., F.E. Dayan, J.G. Romagni and A.M. Rimando. 2000. Natural products as sources of herbicides: current status and future trends. *Weed Res.*, 10: 99-111.

DuPont, S.T., H. Ferris and M.V. Horn. 2009. Effects of cover crop quality and quantity on nematode-based soil food webs and nutrient cycling. *Appl. Soil Ecol.*, 41: 157-167.

Einhellig, F.A. 2004. Mode of allelochemical action of phenolic compounds. In: *Allelopathy, chemistry and mode of action of allelochemicals*. (Eds.): F.A. Macıas, J.C.G. Galindo, J.M.G. Molinillo and H.G. Cutler, CRC Press, BocaRaton, pp. 217-239.

Elijarrat, E. and D. Barcelo. 2001. Sample handling and analysis of allelochemical compounds in plants. *Trends Anal. Chem.*, 20: 584-590.

Gao, J.F. 2000. *Plant Physiology Experiment Technology*. World Publishing Corporation, China.

Garnier, E., S. Lavorel, P. Ansquer, H. Castro, P. Cruz, J. Dolezal, O. Eriksson, C. Fortunel, H. Freitas, C. Golodets, K. Grigulis, C. Jouany, E. Kazakou, J. Kigel, M. Kleyer, V. Lehsten, J. Leps, T. Meier, R. Pakeman, M. Papadimitriou, V.P. Papanastasis, H. Quested, F. Quetier, M. Robson, C. Roumet, G. Rusch, C. Skarpe, M. Sternberg, J.P. Theau, A. Thebault, D. Vile and M.P. Zarovali. 2007. Assessing the effects of land-use change on plant traits, communities and ecosystem functioning in grasslands: a standardized methodology and lessons from an application to 11 European sites. *Ann. Bot.*, 99: 967-985.

Gu, Y., P. Wang and C.H. Kong. 2009. Urease, invertase, dehydrogenase and polyphenoloxidase activities in paddy soil influenced by allelopathic rice variety. *Eur. J. Soil Biol.*, 45: 436-441.

Guan, S.Y. and G.Q. Shen. 1986. Enzyme activities in main soil in China. *Acta Pedol. Sinica.*, 21(4): 368-381.

Guo, R.Y., X.L. Li, P. Christie, Q. Chen, R.F. Jiang and F.S. Zhang. 2008. Influence of root zone nitrogen management and a summer catch crop on cucumber yield and soil mineral nitrogen dynamics in intensive production systems. *Plant Soil*, 313: 55-70.

Guo, T., G. Zhang, M. Zhou, F. Wu and J. Chen. 2004. Effect of aluminumand cadmium toxicity on growth and antioxidant enzyme activities of two barley genotypes with different Al resistance. *Plant Soil*, 258(1): 241-248.

Han, C.M., K.W. Pan, N. Wu, J.C. Wang and W. Li. 2008. Allelopathic effect of ginger on seed germination and seedling growth of soybean and chive. *Sci. Hortic.*, 116: 330-336.

Hodge, H., J. Stewart, D. Robinson, B.S. Griffiths and A.H. Fitter. 1998. Root proliferation, soil fauna and plant nitrogen capture from nutrient-rich patches in soil. *New Phytol.*, 139: 479-494.

Holmgren, M., M. Scheffer and M.A. Huston. 1997. The interplay of facilitation and competition in plant communities. *Ecol.*, 78: 1966-1975.

Hussain, F., S. Ghulam, Z. Sher and B. Ahmad. 2011. Allelopathy by *Lantana camara* L. *Pak. J. Bot.*, 43(5): 2373-2378.

Hussain, F., I. Iiahi, S.A. Malik, A.A. Dasti and B. Ahmad. 2011. Allelopathic effects of rain leachates and root exudates of *Cenchrus ciliaris* L. and *Bothriochloa pertusa* (L.) A. Camus. *Pak. J. Bot.*, 43(1): 341-350.

Ilieva-Makulec, K., I. Olejniczak and M. Szanser. 2006. Response of soil micro- and mesofauna to diversity and quality of plant litter. *Eur. J. Soil Biol.*, 42: 244-249.

Inderjit. 1996. Plant phenolics in allelopathy. *Bot. Rev.*, 62: 186-202.

Laosinwattana, C., T. Poonpaiboonpipat, M. Teererak, W. Phuwiwat, T. Mongkolaussavaratana and P. Charoenying. 2009. Allelopathic potential of Chinese rice flower *(Aglaia odorata* Lour.) as organic herbicide. *Allelopathy J.*, 24: 45-54.

Laosinwattana, C., W. Phuwiwat and P. Charoenying. 2007. Assessment of allelopathic potential of Vetivergrass (*Vetiveria spp*.) ecotypes. *Allelopathy J.*, 19: 469-478.

Lavorel, S. and E. Garnier. 2002. Predicting changes in community composition and ecosystem functioning from plant traits: revisiting the Holy Grail. *Func. Ecol.*, 16: 545-556.

Li, X.F., J. Wang, D. Huang, L.X. Wang and K. Wang. 2011. Allelopathic potential of *Artemisia frigida* and successional changes of plant communities in the northern China steppe. *Plant Soil*, 341: 383-398.

McSorley, R. and R.N. Gallaher. 1994. Effect of tillage and crop residue management on nematode densities on corn. *J. Nematol.*, 26: 669-674.

Meier, C.L., K. Keyserling and W.D. Bowman. 2009. Fine root inputs to soil reduce growth of a neighbouring plant via distinct mechanisms dependent on root carbon chemistry. *J. Ecol.*, 97: 941-949.

Mutlu S. and O. Atici. 2009. Allelopathic effect of *Nepeta meyeri* Benth. extracts on seed germination and seedling of some crop plants. *Acta Physiol. Plant*, 31: 89-93.

Noumi, Z. and M. Chaieb. 2011. Alleolpathic effects of acacia tortilis (Forssk.) hayne subsp raddiana (Savi) brenan in north africa.*Pak. J. Bot.*, 43 (6): 2801-2805.

Putnam, A.R. 1994. Phytotoxicity of plant residues. In: *Managing agricultural residues*. (Ed.): P.W. Unger, Lewis Publishers, Boca Raton, FL, USA, pp. 285-314.

Ranganna, S. 1986. *Handbook of analysis and quality control for fruits and vegetables*. (2nd Ed) McGraw Hill, New Delhi.

Rice, E.L. 1984. *Allelopathy* (2nd Ed) *Academic Press*, Orlando, FL.

Sarrantonio, M. and E. Gallandt. 2003. The role of cover crops in North American cropping systems. *J. Crop Prod*., 8: 53-74.

Siddiqui, Z.S. 2007. Allelopathic effects of black pepper leachings on *Vigna mungo* (L.) Hepper. *Acta Physiol. Plant*, 29: 303-308.

Suding, K.N., S. Lavorel, F.S. Chapin, J.H.C. Cornelissen, S. Diaz, E. Garnier, D. Goldberg, D.U. Hooper, S.T. Jackson and M.L Navas. 2008. Scaling environmental change through the community-level: a trait-based response-and-effect framework for plants. *Global Chang. Biol.*, 14: 1125-1140.

Tabatabai, A. 1994. Soil enzymes. In: *Methods of soil analysis*, (Eds.): R.W. Weaver, J.S. Angle and P.S. Bottomley. Part 2. Microbiological and biochemical properties, *Soil Sci. Soc. Am.*, Madison, W.I., USA, pp. 775-833.

Tian, Y., J. Liu, X. Zhang and L. Gao. 2010. Effects of summer catch crop, residue management, soil temperature and water on the succeeding cucumber rhizosphere nitrogen mineralization in intensive production systems. *Nutr. Cycl. Agroecosyst*, 88(3): 429-446.

Turk, M.A. and A.M. Tawaha. 2002. Inhibitory effects of aqueous extracts of black mustard on germination and growth of lentil. *Pak. J. Agronom*., 1(1): 28-30.

Vitousek, P.M. and R.L.Jr. Sanford. 1986. Nutrient cycling in moist tropical forest. *Annu. Rev. Ecol. Syst.,* 17: 137-167.

Wang, C.H., Z.H. Cheng, Q. Niu, J.N. Liang and S.H. Xue. 2009. *J. Northwest A &F Univ. Natur. Sci. Edit*., 37(7): 103-109.

Wei, L., Z.H. Cheng and L. Zhang. 2008. *J. Northwest A &F Univ. Natur. Sci. Edit*., 36(10): 139-145.

Williams, S.M. and R.R. Weil. 2004. Crop cover root channels may alleviate soil compaction effects on soybean crop. *Soil Sci. Soc. Am. J.*, 68:1403-1409.

Wu, A.P., H. Yu, S.Q. Gao, Z.Y. Huang, W.M. He, S.L. Miao and M. Dong. 2009. Differential belowground allelopathic effects of leaf and root of Mikania micrantha. *Trees Struct. Funct.*, 23: 11-17.

Yang, L.F., Q. Zeng, H.B. Li and J.J. Yan. 2011. Measurement of catalase activity in soil by ultraviolet spectrophotometry. *J. Soil Sci. Chin.*, 42(1): 207-210.

Zeng, R.S. 2008. Allelopathy in Chinese ancient and modern agriculture. In: *Allelopathy in Sustainable Agriculture and Forestry*, (Eds.): R.S. Zeng, U. Azim, Mallik and S.M. Luo, pp. 37-59.

(Received for publication 22 December 2011)