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Table: 1, Figs : 8

Allelopathic effects of *Acacia nilotica* leaf residue on *Pisum sativum* L.

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ABSTRACT

A greenhouse pot experiment was conducted to assess the allelopathic effects of *Acacia nilotica* leaves on the growth and metabolic activities of 45-day-old pea (*Pisum sativum* L.) plants. Qualitative and quantitative HPLC analysis of water extract of *Acacia nilotica* leaves revealed that protocatechuic and caffeic acids were the principal phenolic compounds accompanied by major amounts of ferulic, cinnamic acids and apigenin; whereas, pyrogallic, *p*-coumaric, syringic acids and coumarin were found in trace amounts. The lower doses of *Acacia* leaf residue (0.25 and 0.5 %, w/w) stimulated the growth of pea shoot and root, but the higher doses (0.75, 1.0, 1.5 and 2 %, w/w) were inhibitory to seedling growth and the effect was concentration dependent. The total phenolic content of pea shoots (particularly phenolic glycosides), increased at lower doses of *Acacia* residue and decreased with higher ones While, the phenolic aglycones increased with higher doses than lower ones. Chlorophyll *a*, *b* and carotenoids accumulated in pea shoot at lower doses of *Acacia* leaf residues, accompanied by accumulation of total sugar, mainly the insoluble fraction. On the other hand, the inhibition in the contents of photosynthetic pigments at higher doses of *Acacia* residues was paralleled by significant reduction in all sugar fractions. The contents of total nitrogen and phosphorus (their insoluble forms), increased with lower *Acacia* residues (0.25 and 0.5 %); whereas all nitrogen and phosphorus fractions declined by increasing *Acacia* doses up to 1 %. The total nucleic acids, including DNA and RNA increased with lower *Acacia* residue doses and gradually declined with the increase in *Acacia* level up to 1%.

Key Words: *Acacia nilotica*, agroforestry, allelopathy, growth, natural phenolics, nitrogen, nucleic acids, photosynthetic pigments, phosphorus, *Pisum sativum* L.

INTRODUCTION

Allelopathy is more pronounced in forest plantations, by affecting the growth of understory plant spp. (3,7,25,31,36). *Acacia* (Leguminosae: Minosoideae) spp. are of important economic value (the leaves are animal fodder, timber as fuel wood, charcoal, gum and tannins) in arid and semi-arid environments (15), hence, planted on field borders as windbreaks or grow natively in fields (26). In Egypt, *Acacia* trees are found on the canal banks in the Nile region (4). The leachates of various parts of *Acacia* species have allelopathic activity ranging from stimulation to inhibition on seedling growth of many

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crops, depending on the concentration of allelochemicals and on the sensitivity of target species (20,24,32). *Acacia nilotica* suppresses the seedling establishment of many agricultural plants, the bark extract was more inhibitory than leaf extracts (35). Moreover, Tripathi *et al.* (37) demonstrated that the lower concentrations of foliar extract of *Acacia nilotica* stimulated the seedling growth of soybean, but the higher ones had suppressive effects.

Acacia species contain allelopathic compounds, which have strong allelopathic potential on the plants grown under the tree canopy. Sundaramoorthy *et al.* (34), Fagg and Stewart (15), and Gonzalez *et al.* (19) identified several allelochemicals, mainly phenolic acids and flavonoids, in various parts of *Acacia* species that were phytotoxic to the seedling growth of many crop plants. The physiological and metabolic actions of various natural products on higher plants have received little attention (11,27). Therefore, we assessed the allelopathic effecs of *Acacia nilotica* leaf residue on some physiological and biochemical aspects of pea (*Pisum sativum*) plant and to identify the responsible allelochemicals present in *Acacia* leaves.

MATERIALS AND METHODS

Pea (*Pisum sativum* L. cv. Master B.) seed was obtained from the Department of Vegetable Research, Ministry of Agriculture, Egypt. The mature leaves of *Acacia nilotica* tree were collected from a 6-years old stand at Wadi El-Natron, Egypt. The leaves were air dried in shade and ground into a fine powder. A pot experiment was done in plastic pots (13 cm dia and 15cm depth), each containing 7 kg of clay and sand mixture (w /w, 3:1). The pots were divided into 7 groups, the first was control and the in other six groups *Acacia* powdered leaves were added to give final concentration of 0.25, 0.5, 0.75, 1.0, 1.5 and 2.0% (w/w, residue/soil). Ten healthy uniform pea seeds were sown at 2cm. soil depth per pot. After emergence, the seedlings were thinned to 5 plants per pot. All pots were arranged in randomized complete block design in greenhouse under natural condition of light with 10 h photoperiod and mean 20/15 °C day/night temperature and 50-60% relative humidity. The plants were watered with tap water as needed.

The plants were harvested at 45 days after sowing, then washed thoroughly with distilled water and divided into root and shoot organs. Lengths of the main root and shoot and their fresh weights were measured. The samples were oven dried at 85°C and their dry weights were estimated and water contents were calculated on dry weight basis. Photosynthetic pigments were extracted from fresh pea shoot with 100% acetone using the method of Fadeel (14), then measured and calculated using the equation of Sestak *et al.* (30). Extraction of nucleic acids from fresh pea shoot was done as per the method of Mohamed and El-Sayed (23). DNA and RNA were measured using diphenylamine and orcinol reactions, respectively as described by Clark and Switzer (9). One ml of nucleic acid extract was mixed with 6 ml of freshly prepared diphenylamine reagent and heated in a boiling water-bath for 10 min., then the absorbance of blue colour was measured at wave length 600 nm. Six ml of orcinol acid reagent and 0.4 ml alcoholic orcinol were added to 0.2 ml of nucleic acid extract, then the mixture was heated in boiling water-bath for 20 min. and its optical density was measured at 660 nm wavelength.

Soluble sugars were extracted from dried powdered pea shoot tissue with 80% ethanol by refluxing for 1 h (38). The reducing value of the sugar extract was determined by Nelson's reaction (9). A mixture of 1 ml of sugar extract and 1 ml of freshly prepared Nelson's alkaline copper reagent (Nelson's A and B; 25:1 ratio) was heated in boiling water-bath for 20 min. After cooling, 1 ml arsenomolybdate reagent was added, shaked well and completed up to volume with distilled water, then colour intensity was measured at 540 nm. The non-reducing sugars were hydrolyzed in the ethanol extract with 6N HCl (17), whereas the insoluble carbohydrates in the remaining dry residue were hydrolyzed with 0.2N H_2SO_4 in a boiling water bath for 1 h (33). All data are expressed in terms of glucose equivalents. Phenolic compounds were extracted from dry shoot with 80% hot methanol and hydrolyzed with 2N HCl for 15 min in boiling water bath to cleave the glycosidic linkages (21). Phenolic contents in the methanolic extract before and after hydrolysis were estimated using Folin-Ciocalteau phenolic reaction (2).

Total nitrogen and phosphorus were determined after the acid digestion with 1 ml 50% H_2SO_4 and 1 ml 30% perchloric acid, using Berthelot reaction (8) and modified Fiske-Subborow method (9), respectively. Soluble nitrogen and phosphorus were extracted from the dried pea shoot tissue with 10% trichloroacetic acid (TCA) and the remaining dried residue was acid digested to obtain the insoluble components. Total amount of free amino acids was estimated in the TCA extract as amino-N as per method of Russell (28).

HPLC analysis of phenolic compounds in the aqueous extract of *Acacia nilotica* leaves was carried out on Hewlett-Packard HPLC system fitted with a reverse-phase ultrasphere C18 hypersil column (250 x 4.69 mm) of 25 μm particle size. Phenolic compounds in the sample were identified by comparing their retention times with standards mixture and their concentrations were calculated on the basis of area measurments. All data were statistically analyzed, using one-way ANOVA program.

RESULTS AND DISCUSSION

The HPLC analysis of *Acacia nilotica* leaves (Table 1) revealed the presence of 10 phenolic aglycones. The protocatechuic and caffeic acids were most abundant, while ferulic, cinnamic acids and apigenin were detected in lesser amounts. Pyrogallic, *p*-coumaric, syringic acids, catechol and coumarin were present in trace amounts. Tripathi *et al.* (37) demonstrated that phenolic acids are the most active allelochemicals in *Acacia nilotica* leaves, to which the allelopathic activity was ascribed. Three phenolic compounds, mainly coumaric, vanillic and phloretic acids were detected in *Acacia tortilis* leaf extract (34). Moreover the *Acacia melanoxylon* leaf extract contained vanillic and ferulic acids, vanillin, 4-hydroxy-3-methoxybenzyl alcohol, quercetin-3-glucoside, quercitrin, luteolin and apigenin (19).

The lower concentrations of *Acacia* leaf residue (0.25 and 0.5%) stimulated the length, fresh and dry weights of pea shoot and root over the control, while their water content were insignificantly affected. The shoot elongation was greatly stimulated than root (Figs. 1 and 2). However, the promotion of fresh and dry weights was more pronounced in root than in shoot. Conversely, increasing the rate of *Acacia* residue up to 2.0% caused a gradual decline in all measured growth parameters, the medium inhibition

Table 1. The qualitative and quantitative analysis of phenolic aglycones of *Acacia nilotica* leaves using HPLC-Values are expressed as mean ± Standard error.

Phenolic Compound	Retention Time (min)		Concentration ($\mu g\ g^{-1}$ dry wt.)
	Standard	Sample	
Pyrogallic acid	3.18	3.074	48.36± 0.02
Resorcinol	9.61	-	-
Protocatechuic acid	10.31	10.332	532.78± 0.14
Catechol	13.47	13.346	105.66± 0.06
p-Hydroxybenzoic acid	14.13	-	-
Caffeic acid	14.57	14.725	568.35± 0.11
Chlorogenic acid	15.75	-	-
p-Coumaric acid	20.13	20.050	77.02± 0.01
Syringic acid	20.28	20.937	64.76± 0.02
Ferulic acid	22.46	22.561	392.81± 0.21
Coumarin	29.86	29.972	20.98± 0.03
Salicylic acid	33.61	-	-
Cinnamic acid	34.11	34.132	346.38± 0.07
Apigenin	36.04	36.139	288.50± 0.13
Kaempferol	39.29	-	-

occurred at 2.0% concentration. The inhibition of cell elongation may be related to the direct action of allelochemicals by interfering with cell division either directly or through interaction with hormones. These results are in accordance with Tripathi *et al.* (37), who demonstrated stimulation in the root and shoot length of 30-day-old soybean plants irrigated with lower concentration of *A. nilotica* leaf extract, while the higher concentration were inhibitory. Moreover, Srinivasan *et al.* (32) showed that the growth of soybean was significantly inhibited with *A. holosericea* than the growth of cowpea and pigeon peas. Swaminathan *et al.* (35) ascribed the poor growth of eight arable crops under *A. nilotica* canopy to the tannins leached from the bark. Growth and productivity of crops in association with *A. nilotica* tree belt were adversely affected by foliar leachates of *A. nilotica* (26). In earlier study, Geissman and Phinny (18) attributed the reduction in plant growth to the interference of phenolic compounds with gibberellin function. Moreover, Mersie and Singh (22) and Baziramakenga *et al.* (5) showed that several phenolic acids suppressed the growth of tomato and soybean plants.

The total phenolic content, mainly phenolic glycosides of 45-day-old pea shoot increased when raised with 0.5% *Acacia* leaf residue. Such accumulation can be attributed to the stimulation of phenolic biosynthesis and their binding with sugar units forming phenolic glycosides (non-phytotoxic). At higher concentrations of *Acacia* leaf residue (0.75 and 1.0%), the decline of total phenolic content may be due to inhibition of phenolic biosynthesis and their glycosylation that leading to increase in the level of phenolic aglycones (phytotoxic). In this respect, Sato *et al.* (29) showed that the activity of phenylalanine ammonia- lyase (catalyzed the first step in phenolic biosynthesis) decreased with increasing the cinnamic acid derivatives and related compounds.

Reigosa *et al.* (27) and Einhellig (11) reviewed that allelochemicals affected the growth and many vital processes due to their interference with several metabolic activities,

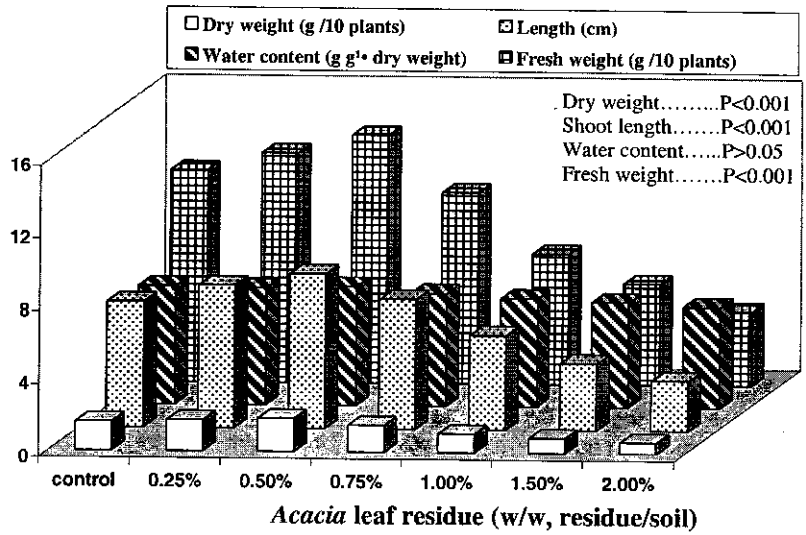

Figure 1. Effects of different concentrations of *Acacia* leaf residue on the shoot growth of 45-day-old pea plant.

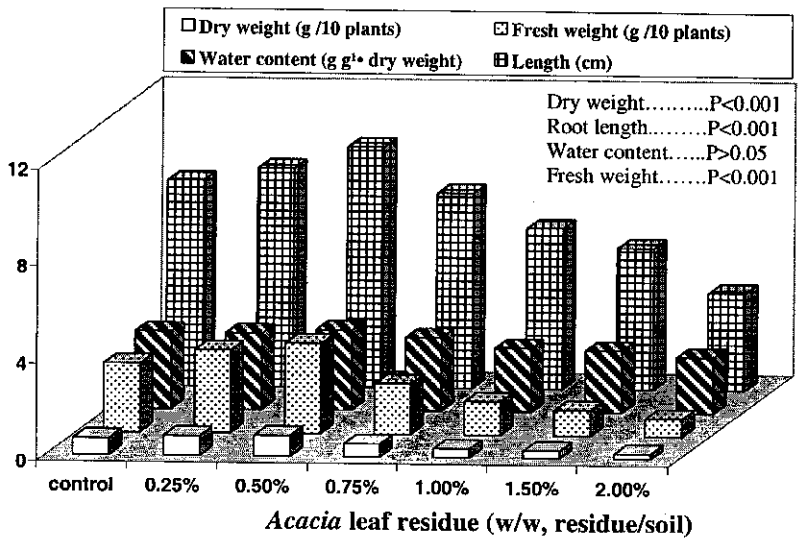

Figure 2. Effects of different concentrations of *Acacia* leaf residue on the root growth of 45-day-old pea plant.

where the effects are concentration dependant. Thus, the reduction in pea growth under the higher concentrations of *A. nilotica* residue was related to the increasing in the level of the internal phenolic aglycones.

Increasing *Acacia* leaf residue up to 0.5% level increased the contents of chlorophyll *a* and *b* as well as carotenoids relative to their controls, whereas the higher treatments reduced their contents particularly chlorophyll *a* (Fig. 4). The allelochemicals released from *Acacia* residue may interfered with the biosynthesis of photosynthetic pigments. Similarly, Tripathi *et al.* (37) showed that the higher concentrations of *A. nilotica* leaf extract reduced the chlorophyll *a* content in the leaves of 30-day-old soybean. Other studies demonstrated a reduction in chlorophyll *a* and *b* contents coupled with a chlorotic appearance in the leaves of plants treated with phenolic acids or plant residues (5,11,12,13,22).

A significant increase in the total carbohydrate content, particularly the insoluble sugar, in pea shoot treated with *Acacia* leaf residue up to 0.5% concentration, despite reduction in the total soluble sugars as compared to their respective controls (Fig. 5). This effect may be attributed to the activation of the enzyme system involved in the conversion of the soluble sugars to insoluble form. On the other hand, the higher levels of *Acacia* residues significantly reduced the contents of all sugar fractions and the reduction was concentration dependent. This is in accordance with Tripathi *et al.* (37), who showed that *A. nilotica* leaf extract at 5% concentration, increased the carbohydrate content in soybean leaves, while minimum content was found at 10 and 20% concentrations. A similar trend was found in soybean treated with different concentrations of lupine seed extract (1).

With respect to the nitrogen content of pea shoots, the increasing levels of *Acacia* leaf residue up to 1.0% gradually decreased the total soluble nitrogen content, including the total amount of free amino acids and other soluble nitrogen fraction (Fig. 6). However, the contents of total nitrogen and the insoluble nitrogen reached maximum value at 0.5% *Acacia* treatment, then declined by higher doses, where the effect was concentration dependent. This could be due to the higher levels of *Acacia* allelochemicals, which have harmful effect on nitrogen metabolism. This effect may be attributed to the stimulation of the conversion of soluble nitrogen into the insoluble form under the lower *Acacia* treatments. In this respect, Bazairamakenga *et al.* (5) found that incorporation of *S*-methionine into proteins was increased in soybean seedlings by low concentrations of *p*-hydroxybenzoic and *p*-coumaric acids. However, Tripathi *et al.* (37) reported that the protein and amino acid contents in soybean increased linearly with increasing leaf extract concentrations of *A. nilotica*. Moreover, Duhan *et al.* (10) and Balasubramanian and Ravichandran (3) showed that allelochemicals of *Acacia nilotica* and other agroforestry trees inhibited legume-*Rhizobium* symbiosis and interfered with subsequent nodulation and nitrogen metabolism.

The maximum increase in total phosphorus content over the control, occurred in pea shoot treated with 0.5% *Acacia* leaf residue (Fig. 7). This was related to the increases in both total soluble and insoluble phosphorus levels despite decline in the content of inorganic soluble phosphorus and increase in the organic soluble form. It may be attributed to the enhancement of conversion of inorganic soluble phosphorus into organic forms. On the other hand, the treatment with higher *Acacia* residue (0.75 and 1%) severely attenuated all phosphorus fractions (Fig. 7). This reduction may be due to the inhibition of the

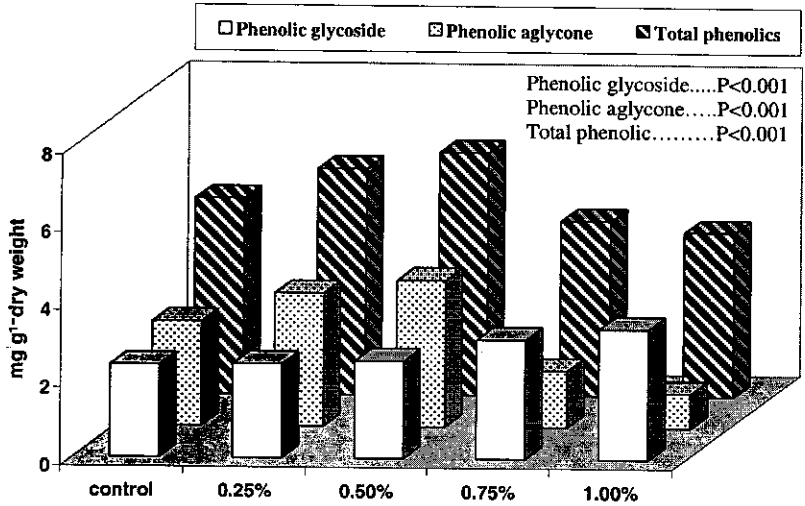

Figure 3. Effects of different concentrations of *Acacia* leaf residue on the phenolic content (mg g^{-1} dry weight) of 45-day-old pea shoot.

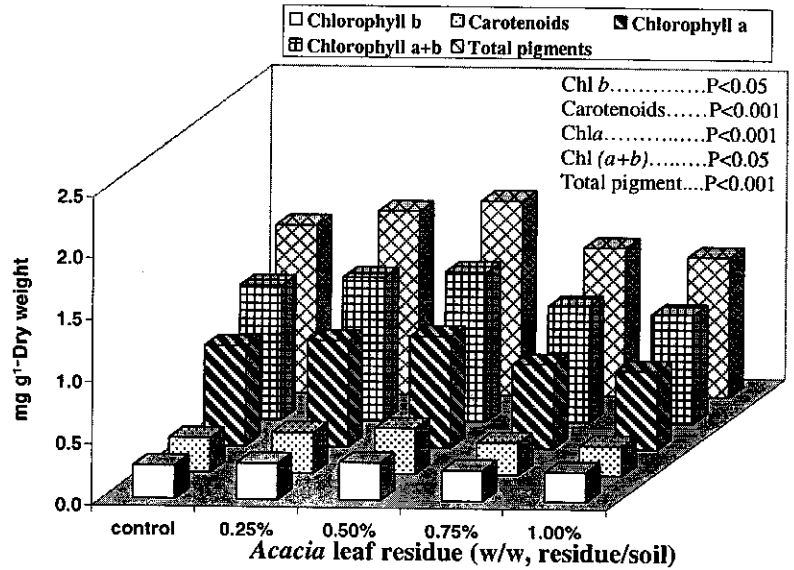

Figure 4. Effects of different concentrations of *Acacia* leaf residue on the various photosynthetic pigments (mg g^{-1} dry weight) of 45-day-old pea shoot.

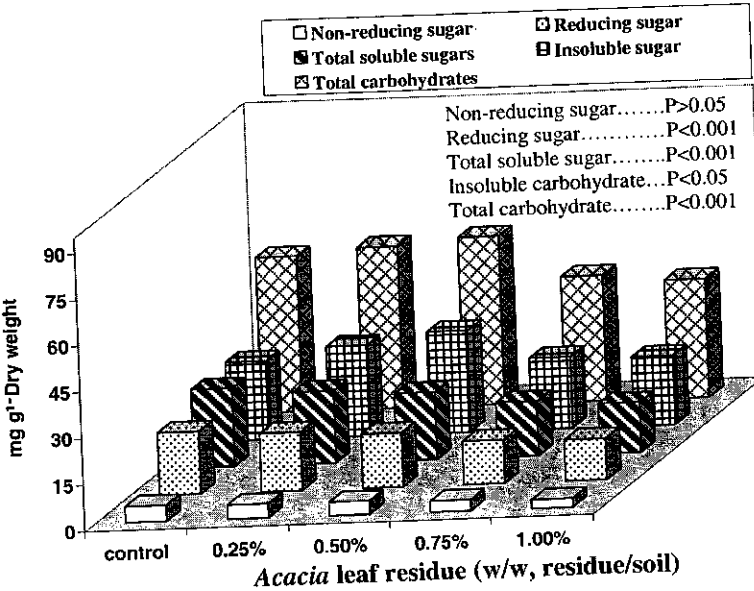

Figure 5. Effects of different concentrations of *Acacia* leaf residue on the various carbohydrates contents (mg g^{-1} dry weight) of 45-day-old pea shoot.

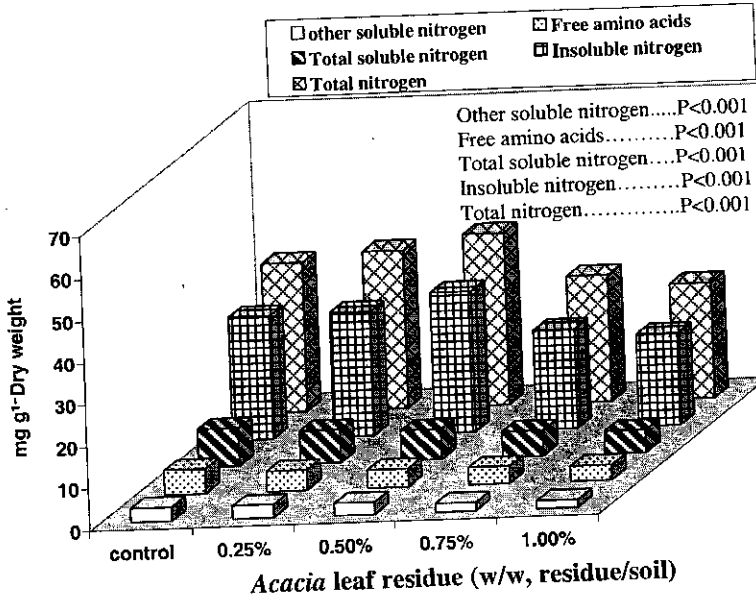

Figure 6. Effects of different concentrations of *Acacia* leaf residue on the various nitrogen contents (mg g^{-1} dry weight) of 45-day-old pea shoot.

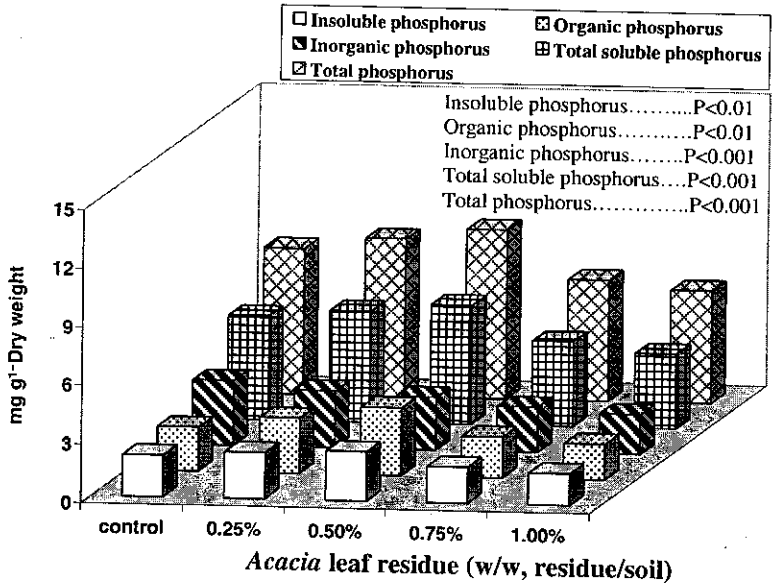

Figure 7. Effects of different concentrations of *Acacia* leaf residue on the various phosphorus contents (mg g^{-1} dry weight) of 45-day-old pea shoot.

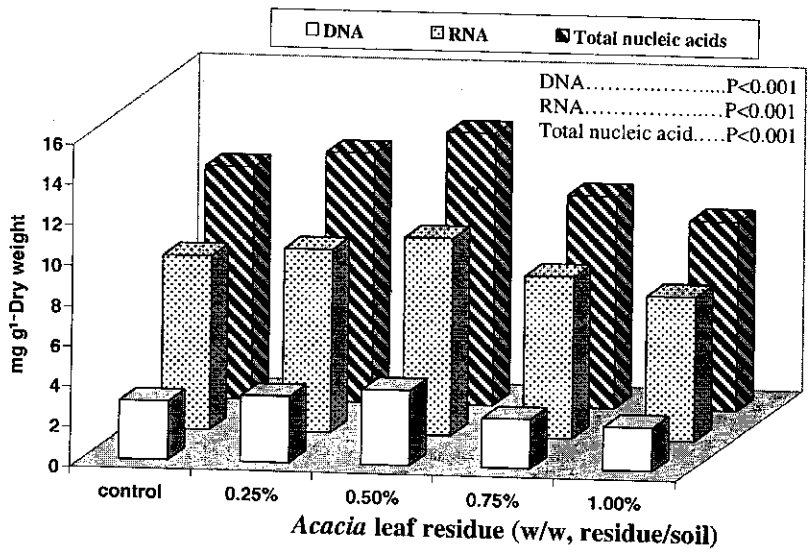

Figure 8. Effects of different concentrations of *Acacia* leaf residue on various nucleic acid contents (mg g^{-1} dry weight) of 45-day-old pea shoot.

phosphorus uptake and assimilation. Our results are supported by the observation of Mersie and Singh (22) and Vaughan and Ord (39), that the phenolic acids may interfere with the phosphorus uptake. In this connection, Friebe *et al.* (16) demonstrated that phenolic compounds inhibited the plasma membrane ATPase proton pump that affects the phosphorus uptake.

The incorporated *Acacia* residues at 0.25 and 0.5% levels considerably stimulated the accumulation of nucleic acids, compared to control (Fig. 8). The increase in phosphorous content under these treatments may be explained that the lower *Acacia* residue levels could stimulate the incorporation of phosphorus into nucleotides and/or stimulate the enzyme system involved in the biosynthesis of new nucleic acids (6). Inversely, the higher *Acacia* residues (0.75 and 1%) reduced the nucleic acid content of pea shoot. As phosphorus is main constituent of nucleic acids, the reduction in phosphorus content of pea shoot under higher *Acacia* treatments, may limit nucleic acids biosynthesis. Similarly, Baziramakenga *et al.* (5) reported many phenolic acids reduced the incorporation of ^{32}P into nucleic acid in soybean seedlings. This response suggests that the higher *Acacia* residues levels inhibited the rate of DNA and RNA production, thereby reduced the levels of protein synthesis and the productivity of pea plant.

In conclusion, the allelopathic activity of *Acacia* tree partly depends on the amount and type of allelochemicals released from the decomposed leaves, as well as the uptake of these compounds by plant root.

REFERENCES

1. Al-Wakeel, S.A.M. and Soliman, M.H. (1994). Allelopathic effects of lupine extracts on some physiological aspects in soybean plants. *Bulletin Egyptian Society of Physiological Science* **14**: 124-138.
2. AOAC (1990). *Official Methods of Analysis.* The Association of Official Analytical Chemists. Tanni, 15th Edn. pp. 746. Washington, D. C.
3. Balaubramanian, A. and Ravichandran, V.K. (1996). Allelopathic significance of six agroforestry trees on *Casuarina equiestifolia* growth and nodulation. *Madras Agriculture Journal* **83**: 83-87.
4. Batanouny, K. H. (1999). *Wild Medicinal Plants in Egypt.* pp. 98-101. Palm Press, Cairo, Egypt.
5. Baziramakenge, R.; Simard, R.R. and Leroux, G.D. (1994). Effects of benzoic and cinnamic acids on growth, mineral composition, and chlorophyll of soybean. *Journal of Chemical Ecology* **20**: 2821-2833.
6. Baziramakenga, R.; Leroux, G.D.; Simard, R.R and Nadeau, P. (1997). Allelopathic effects of phenolic acids on nucleic acid and protein levels in soybean seedlings. *Canadain Journal of Botany* **75**: 445-450.
7. Carballeira, A. and Reigosa, M.J. (1999). Effects of natural leachates of *Acacia dealdata* Link in Galicia (NW Spain). *Botanical Bulletin Academia Sinica* **40**: 87-92.
8. Chaney, A.L. and Marbach, E.P. (1962). Modified reagents for determination of urea and ammonia. *Clinical Chemistry* **8**: **130-132.**
9. Clark, J.M. and Switzer, R.L. (1977). *Experimental Biochemistry.* 2nd Edn. Freedman and Company, San Francisco.
10. Duhan, J.S., Sharma, P.K. and Lakshminarayana, K. (1994). Allelopathic effect of *Acacia nilotica* on nodulation and nitrogen fixation by *Rhizobium* (Cowpea). *Allelopathy Journal* **1**: 47-52.
11. Einhellig F.A. (2002). The physiology of allelochemical action: clues and views. In: *Allelopathy From Molecules to Ecosystems,* (Eds., M.J. Reigosa and N. Pedrol), pp. 1-23. Science Publishers Inc. USA.
12. Ervin, G.N. and Wetzel, R.G. (2000). Allelochemical autotoxicity in the emergent wetland macrophyte *Juncus effuses* (Juncaceae). *American Journal of Botany* **87**: 853-860.
13. Eyini, M., Jayakumar, M. and Pannirselvam, S. (1989). Allelopathic effect of bamboo leaf extract on the seedling of groundnut. *Tropical Ecology* **30**: 138-141.
14. Fadeel, A. A. (1962). Location and properties of chloroplasts and pigment determination in roots. *Physiological Plantarum* **15**: 130-147.

15. Fagg, C.W. and Stewart, J.L. (1994). The value of *Acacia* and *Prosopis* in arid and semi-arid environments. *Journal of Arid Environment* **27:** 3-25.
16. Friebe, A., Roth, U., Kuck, P., Schnabl, H. and Shulz, M. (1997). Effects of 2, 4-dihydroxy-1, 4-benzoxazin-3-ones on the activity of plasma membrane H^+-ATPase. *Phytochemistry* **44**: 979-983.
17. Gaines, T.P. (1973). Automated determination of reducing sugars, and starch in plant tissue from one weighed sample. *Journal Association of Official Analytical Chemists* **56**: 1419-1424.
18. Geissman, T.A. and Phinny, B. O. (1972). Tannins as gibberellin antagonists. *Plant Physiology* **49**: 323-330.
19. Gonzalez, L., Souto, X.C. and Reigosa, M.J. (1995). Allelopathic effects of *Acacia melanoxylon* R. Br. Phyllodes during their decomposition. *Forestry Ecological Management* **77**: 53-63.
20. Jadhav, B.B. and Gaynar, D.G. (1992). Allelopathic effects of *Acacia ouriculiformis* A. Cunn. on germination of rice and cowpea. *Indian Journal of Plant Physiology* **35**: 86-89.
21. Jindal, K.K. and Singh R.N. (1975). Phenolic content in male and female *Carica papaya*: A possible physiological marker for sex identification of vegetative seedlings. *Physiological Plantarum* **33**: 104-107.
22. Mersie, W. and Singh, M. (1988): Effects of phenolic acids and ragweed Parthenium (*Parthenium hysterophorus*) extracts on tomato (*Lycopersicon esculentum*) growth, nutrient and chlorophyll content. *Weed Science* **36:** 278-281.
23. Mohamed, Y.A.H. and El- Sayed, M.A. (1982). The role of cobalt in nucleic acid content of *Azotobacter chroocaccum*. *Iraqi Journal of Science* **23**: 421-424.
24. Nsolomo, V.A.; Mrecha, M.S. and Maghembe, J.A. (1995). Effect of *Acacia xanthophloea* leachates on seed germination of some agricultural and multipurpose tree crops. *Journal of Tropical Forest Science* **7:** 398-404.
25. Pellissier, F., Gallet, C. and Souto, X.C. (2002). Allelopathic interaction in forest ecosystem. In: *Allelopathy: From Molecules to Ecosystems*, (Eds., M. J. Reigosa and N. Pedrol), pp. 254-269. Science publishers, Inc. USA.
26. Puri, S., Singh, S. and Kumar, A. (1994). Growth and productivity of crops in association with an *Acacia nilotica* tree belt. *Journal of Arid Environment* **27**: 37- 48.
27. Reigosa, M.J., Moeiras, A.S. and Gonzalez, L. (1999). Ecophysiological approach in allelopathy. *Critical Review of Plant Sciences* **18**: 577- 608.
28. Russell, G.A. (1944). Note on the colorimetric determination of amino nitrogen *Journal of Biological Chemistry* **156**: 467- 468.
29. Sato, T.,Kiuchi, F. and Sankawa, U. (1982). Inhibition of ~phenylalanine ammonia-lyase by cinnamic acid derivatives and related compounds. *Phytochemistry* **21**: 845-850.
30. Sestak, Z., Catsky, J. and Jarvis, P.G. (1971). Determination of chlorophylls *a* and *b*. In: *Plant Photosynthetic Production: Manual Methods*. (Eds., Z. Sestak, J. Catsky and P.G. Jarvis) pp. 672-697. Dr. W. Junk Publ. The Hague.
31. Souto, X.C., Gonzalez, L. and Reigosa M.J. (1995). Allelopathy in forest environment in Galicia, NW Spain. *Allelopathy Journal* **2:** 67-78.
32. Srinivasan, K., Ramasamy, M. and Shantha, R. (1990). Tolerance of pulse crops to allelochemicals of tree species. *Indian Journal Pulses Research* **3**: 40-44.
33. Streeter, J.G. and Jeffers, D.L. (1979). Distribution of total non-structural carbohydrates in soybean plants having increased reproductive load. *Crop Science* **19** : 729-734.
34. Sundaramoorthy, S., Kalra, N. and Sen, D.N. (1992). Allelopathic effect of *Acacic tortilis* on seedling metabolites of clusterbean. Proceedings, First National Symposium on *Allelopathy in Agroecosystems, Agriculture and Forestry*. (Eds., P. Tauro and S. S. Narwal), pp. 125-126. CCS, Haryana Agricultural University, Hisar, India.
35. Swaminathan, C., Rai, R.S.V. and Suresh, K.K. (1989). Allelopathic proclivities of *Acacia nilotica* (L.) Willd. ex Del. *Journal of Tropical Forestry Science* **2: 56 – 60.**
36. Swaminathan, C., Sivagnanam, K. and Srimathi, P. (1993). Allelopathic proclivities of multipurpose trees. *Myforest* **29:** 147-149.
37. Tripathi, S., Tripathi, A., Kori, D.C. and Tiwari, S. (1998). Effect of tree leaves aqueous extracts on germination and seedling growth of soybean. *Allelopathy Journal* **5:** 75-82.
38. Upmeyer, D.J. and Koller, H. R. (1973). Diurnal trends in net photosynthetic rate and carbohydrate levels of soybean leaves. *Plant Physiology* **51**: 871-874.
39. Vaughan, D. and Ord, B.G. (1991). Extraction of potential allelochemicals and their effects on root morphology and nutrient contents. In : *Plant Root Growth: An Ecological Perspective* (Ed., D. Atkinson), pp. 399-421, Black, London.