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Original Research Article

Physiological Allelopathic Effect of Aqueous Extracts of Cucumber, Carrot, Onion, and Garlic Seeds on Germination and Growth of Pea

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ABSTRACT

Allelopathic potential of aqueous extracts of cucumber, carrot, onion and garlic seed on germination and growth of pea was studied. Aqueous extracts at 0.0, 10, 30 and 60% concentrations were used. The results indicate that both cucumber and carrot seeds extracts exerted positive allelopathic effects and improved seed germination, growth and metabolic activities of pea seedlings. Also, the data showed that both cucumber and carrot seeds extracts led to increased auxin content and decreased abscisic acid content of pea seedlings. On the other hand, onion and garlic seed extracts had significant negative allelopathic effects on seed germination, growth parameters and metabolic activities of pea seedlings. Extracts of onion and garlic seeds led to pea seedlings of higher abscisic acid content and lower auxin content. All extracts exerted concentration dependent allelopathic effects on pea growth.

Keyword: Allelochemicals; germination; pea; companion plants; incompatible plants

INTRODUCTION

Pea (*Pisum sativum*) is an annual plant, with a life cycle of one year. It is a cool season crop grown in many parts of the world; it is used as a vegetable, fresh, frozen or canned. The nutritional value of pea is amazing. Pea contains protein, potassium, phosphorus, Magnesium, calcium, sodium, selenium, iron, zinc, manganese, and iron , niacin, folate, vitamins A, C, B1, B6 and other vitamins in small amounts. Allelopathy refers to the beneficial or harmful effects of one plant on another plant through releasing of biochemicals from plant parts.

Allelochemicals are a subset of secondary metabolites not required for metabolism (growth and development) of the allelopathic organism. The term allelopathy is from the Greek-derived compounds *allelo and pathy* (meaning "mutual harm" or "suffering"). Allelopathy can affect many aspects of plant ecology, including occurrence, growth, plant succession, the structure of plant communities, dominance, diversity, and plant productivity [1]. Initially, many of the forestry species evaluated had negative allelopathic effects on food and fodder crops, but in the 1980s research was begun to identify species that had beneficial, neutral, or selective effects on companion crop plants [2]. Commonly cited effects of allelopathy include reduced seed germination and seedling growth. However, known sites of action for some allelochemicals include cell division, nutrient uptake, photosynthesis, and specific enzyme function. The inhibition by these compounds is due to adverse effects on amino acids metabolism and iron concentration equilibrium. Allelopathic inhibition is complex and can involve the interaction of different classes of chemicals, such as phenolic compounds, flavonoids, terpenoids, alkaloids, steroids, carbohydrates, and amino acids. Companion planting is based on the idea that certain plants can benefit others and become healthy when planted in near proximity. Companion plants benefit each other when planted in close proximity. They work well together, attracting good insects and keeping away the unwanted ones. Companion plants also provide nutrients and in some cases natural shade and support to their garden neighbors. Different plant parts, including flowers, leaves, leaf litter and leaf mulch, stems, bark, roots, soil, and soil leachates and their derived compounds, can have allelopathic activity that varies over a growing season. Allelopathic chemicals or allelochemicals can also persist in soil, affecting both neighboring plants as well as those planted in succession. Both cucumber and carrot are very good sources of vitamin C. Vitamin C rescues the cells from an onslaught of destructive free radicals. Cucumber and carrot extracts contain folate, fibers, riboflavin, and potassium, as well as good amounts of copper, vitamin B6, magnesium, manganese, and calcium. Also, cucumber and carrot extracts include detoxifying agents called indoles, and the powerful flavonoids zeaxanthin, lutein, and beta carotene. Garlic and onion are believed to interfere with the growth of beans and peas. Plants that don't like each other may be responding to different environmental needs, could be in direct competition with one another for major resources or one may attract insects that severely harm the other [3]. The objective of the present study was to assess the physiological allechemical effects of aqueous extracts of cucumber, carrot, onion, and garlic seeds on germination and growth of pea.

MATERIALS AND METHODS

Seed extracts of four plants, cucumber, carrot, onion and garlic, are used to study their compatibility and incompatibility effects on germination and growth of pea plant. Seeds of the used plants were obtained from Agriculture Research Centre, Dokki, Giza. Three different concentrations (10, 30 and 60% w/v) were prepared. To prepare 10% seed extract; 50 g fine grind of cleaned air-dried seeds was soaked in 500 ml distilled water for 72 hours at room temperature to allow the auto extraction, then kept in a dark room. The mixture was filtered using muslin cloth. Seeds of pea were surface sterilized with 0.03% formalin for one hour and thoroughly washed with tap water following with distilled water. Pea seeds were divided into groups; one is treated with distilled water alone as a control. The other groups were

treated with different seeds extracts. The sterilized seeds were germinated in Petri dishes (150 mm in diameter x 15 mm deep) containing two sheets of sterile Whatman No. 1 filter paper that had been premoistened with 10 mL of seed extract solution. The Petri dishes, containing the seeds were moved to a controlled environment chamber at 20 ± 2 °C for germination under 18 h light- 6 h dark cycle. Seed germination was observed daily with solutions added to the Petri dishes as necessary to maintain moisture levels. Measurements of seedling growth and metabolic activity were made at 14 days after transfer of the seeds to the Petri dishes. Chemical analyses: Total chlorophylls were estimated in 85% acetone extracted leaves according to Metzner et al. [4]. Total carbohydrates content was determined using the anthrone method described by Umbriet et al. [5]. Soluble proteins content of leaf samples was determined using the method of Lowry et al. [6]. DNA was measured according to Dische and Schwartz [7] by using diphenylamine reagent. RNA was determined using the method adopted by Ashwell [8] using orcinol reagent. Auxin and abscisic acid contents were estimated using the method of Wasfy and Orrin [9]. HPLC was used for identification and determination of hormones. Standards of IAA and ABA were used. Results are the mean values of three replicates of the same sample. Statistical analysis was performed at 5% level using Snedecor and Cochran [10] method.

RESULTS AND DISCUSSIONS

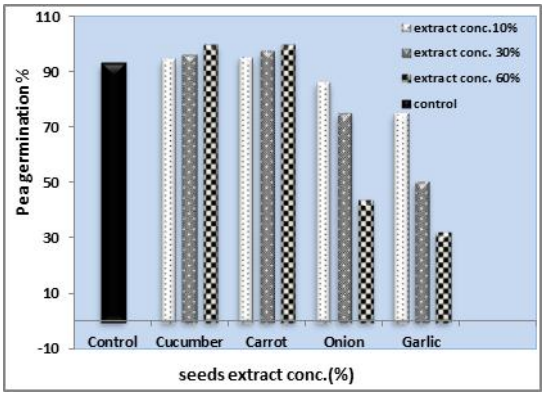
Aqueous extracts of all concentrations of cucumber and carrot seeds significantly stimulated the germination percentage of pea and the stimulation increased with increasing concentration. Whereas, onion and garlic seed extracts significantly inhibited pea germination percentage (Fig. 1a). Also, seeding growth of pea was affected. Cucumber and carrot seeds extracts application led to longer pea seedlings than controls while onion and garlic seed extracts resulted into seedling of shorter lengths (Fig. 1b). The inhibitory effect of onion and garlic seed extracts could be alluded to an increased phytotoxicity by concentration increase. Similarly, the fresh and dry weights of pea seedlings were significantly stimulated by cucumber and carrot seeds extracts and inhibited by garlic and onion seeds extracts (Fig. 1c&d). Higher concentrations of all seeds extracts exerted allelopathic effects on pea seed germination and seedling growth which increased with increasing concentration i.e. it was concentration dependent. Allelopathic nature of seed extracts affects morphological parameters of pea seedlings may be due to stimulated or impaired metabolic activities and growth regulators content of the pea plant which leads to increase or decrease in fresh and dry weights and shoot length depending on the extract and its concentration [11]. These results are in agreement with Thaper and Singh [12], Singh et al. [13] and Lu and Jia [14]. Previous findings support our results in which growth of tomato, radish, cucumber and barnyardgrass inhibited via water extract of *Lantana camara*.

Total chlorophylls, total carbohydrates, soluble proteins and nucleic acids contents of pea seedlings were significantly increased with different concentrations of cucumber and carrot seeds extracts; 10, 30 and 60 % respectively (Table1a & c). The present study demonstrates the phytotoxic influence of allelochemicals present in onion and garlic seed extracts on metabolism of pea seedlings where, total chlorophylls, total carbohydrates, soluble proteins and nucleic acids contents of pea seedlings showed gradual decreases in dose dependent manner. Seedlings of control group contain maximum protein and carbohydrate contents (Table. 1b). The gradual decrease in

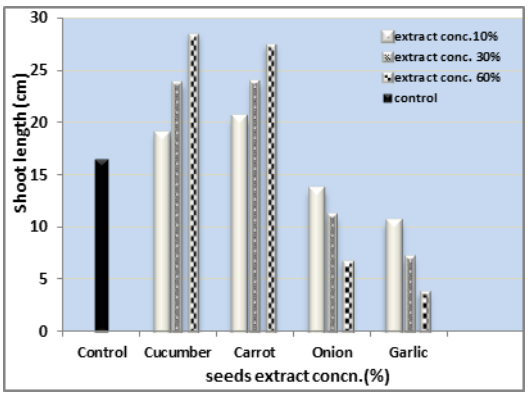
total chlorophylls content which in turn affects carbohydrates content may be due to inhibition in enzyme synthesis, cofactors required for chlorophyll and protein synthesis [15]. Under allelochemical stress, extreme degradation of chlorophyll also arises due to influence of allelochemicals [16]. Allelochemicals reduce the accumulation of chlorophyll content [17]. Similar results were also reported in case of sorghum and radish [18] [19]. Also, nucleic acids (DNA and RNA) contents of pea seedlings were adversely affected by the allelochemical stress caused by onion and garlic seed extracts (Table 1c). Allelochemicals affect plant physiological and cytological processes such as cell wall expansion, protein synthesis, antioxidant enzymatic activities, cell division and nucleic acids level [20]. Decreased rate of protein synthesis is a result of integration of certain amino acids into protein inhibited by phenolic acids [21].

The present study indicates that both cucumber and carrot seed extracts positively affected IAA content of pea seedlings compared with corresponding control. On the contrary, IAA content gradually decreased by onion and garlic seed extracts in dose dependent manner (Fig. 2a). While ABA content of pea seedlings decreased significantly by cucumber and carrot seed extracts, it increased by onion and garlic seed extracts. It is clear that maximum stimulation of auxin content correlates with maximum inhibition of abscisic acid content by 60% seed extracts of carrot and cucumber.

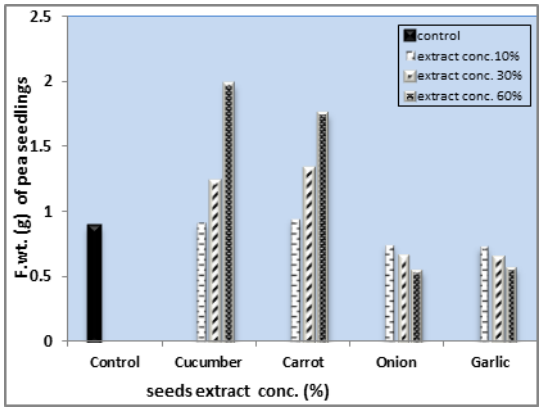
Singh and Sunaina [22] reported that the allelochemicals are known to increase the amount of free amino acids resulted from protein degradation. Also, they added that allelochemicals cause inhibition of cell division in root meristem, inhibition of oxidation of plant hormones, alteration of membrane permeability, reduction of stomatal aperture leading to the impaired photosynthesis, interference with mitochondrial function, reduction of protein synthesis, and clogging of the xylem strand leading to the impaired water balance. Sunflower allelochamicals influenced growth of neighboring mustard plants by induction of oxidative stress and disturbances in hormonal balance between ABA and ethylene in germinating seeds [23]. The decreasing metabolic activity of the mustard seed embryo and blocking its germination and growth may be attributed to the disturbances in phytohormone levels.



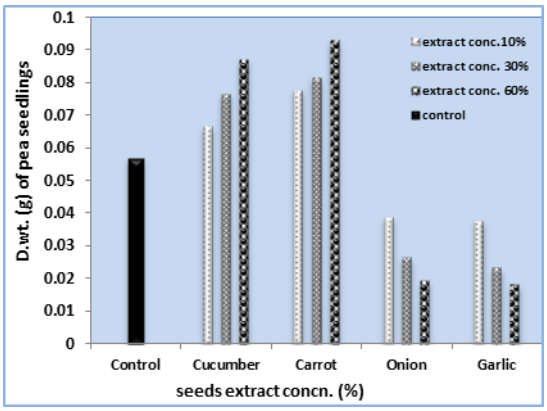
(a) L.S.D. at 5%: 1.32



(b) L.S.D. at 5%: 2.03



(c) L.S.D. at 5%: 0.025



(d) L.S.D. at 5%: 0.007

Fig.1: Allelopathic effect of cucumber, carrot, onion and garlic seed extracts on (a): germination percentage and (b, c, d): seedlings growth of pea.

Table.1.a: Allelopathic effect of cucumber and carrot seed extracts on total chlorophylls, total carbohydrates and soluble proteins contents of pea seedlings (14-d- old).

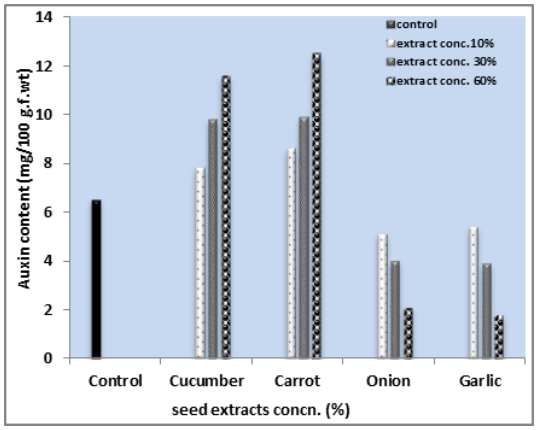
Extract concentration (%)	Total chlorophyll (mg/g.f.wt)		Total carbohydrates(mg/g.d.wt)		Soluble protein (mg/g.d.wt)	
	Cucumber	Carrot	Cucumber	Carrot	Cucumber	Carrot
Control (0.0)	7.5		76.2		67.3	
10	8.13	8.73	81.1	79.4	74.1	72.7
30	9.74	9.82	86.5	87.0	77.3	76.1
60	11.82	12.11	96.8	97.2	83.5	81.6
L.S.D at 5%	0.41	0.45	1.3	1.7	1.4	1.9

Table.1.b: Allelopathic effect of onion and garlic seed extracts on total chlorophylls, total carbohydrates and soluble proteins contents of pea seedlings (14-d- old).

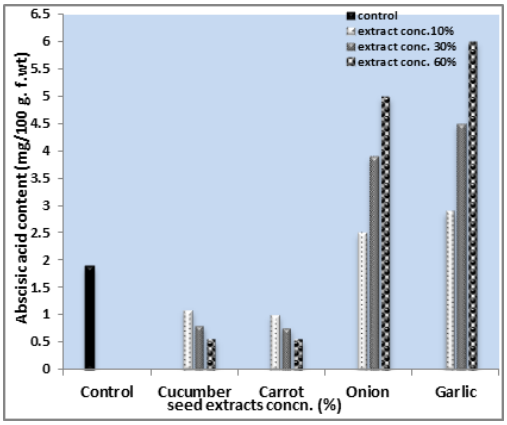
Extract concentration (%)	Total chlorophylls (mg/g.f.wt)		Total carbohydrates (mg/g.d.wt)		Soluble protein (mg/g.d.wt)	
	Onion	Garlic	Onion	Garlic	Onion	Garlic
Control (0.0)	7.5		76.2		67.3	
10	6.3	6.1	68.1	66.4	62.2	60.6
30	5.4	5.0	62.5	61.5	56.7	54.1
60	3.6	3.2	49.4	50.0	50.8	47.7
L.S.D at 5%	0.38	0.33	2.1	2.6	1.9	1.1

Table 1.c: Allelopathic effect of cucumber, carrot, onion and garlic seed extracts on DNA and RNA contents of pea seedlings (14-d- old).

Extract concentration (%)	DNA (mg/g.d.wt)				RNA (mg/g.d.wt)			
	Cucumber	Carrot	Onion	Garlic	Cucumber	Carrot	Onion	Garlic
Control (0.0)	15.3				3.02			
10	16.44	16.91	13.23	13.13	3.96	3.89	2.43	2.33
30	19.12	20.20	11.40	10.33	4.77	4.88	1.21	1.10
60	22.54	23.14	9.12	8.71	6.24	6.35	0.80	0.75
L.S.D at 5%	0.95	0.91	0.88	0.92	0.084	0.082	0.090	0.077



(a) L.S.D. at 5%: 0.47



(b) L.S.D. at 5%: 0.09

Fig. 2: Allelopathic effect of seed extracts of cucumber, carrot, onion and garlic on (a): endogenous auxin (IAA) and (b): abscisic acid contents of pea seedlings

CONCLUSION

The germination and growth of pea was positively affected by cucumber and carrot seed extracts, hence they are believed to be companion plants to pea plant and it is better to be planted in close proximity. The germination and growth of the pea plant was adversely affected by onion and garlic seed extracts and are believed to be incompatible plants to pea, hence it is better to be planted ahead.

CONFLICT OF INTEREST STATEMENT

The authors declare that they have no competing interests.

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