

ALLELOPATHIC EFFECTS OF BLACK NIGHTSHADE (*SOLANUM NIGRUM* L.) ON GERMINATION, GROWTH AND YIELD OF BROAD BEAN (*VICIA FABA* L.) AND COMMON BEAN (*PHASEOLUS VULGARIS* L.)

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Abstract. Germination tests and pot experiments were conducted to explore the allelopathic effect of aqueous extracts of *S. nigrum* on germination, growth and yield of *Vicia faba* (L.) and *Phaseolus vulgaris* (L.). Ten treatments including control were prepared by using extracts obtained from different parts of *S. nigrum*, including roots, leaves, and seeds at 3 concentrations (10, 40, and 60%) to conduct the germination test. The pot experiment was studied by incorporating powder of different plant parts into the soil at 3 concentrations (0.5, 1, and 1.5%). The results of the germination test revealed that the aqueous extracts of roots and leaves significantly reduced germination percentage, radicle, and plumule length of both beans. There was a direct negative relationship between germination test and the plant parts used, including their concentrations. However, seed extract showed a non-significant difference ($p > 0.05$). In the pot experiment, different powders of black nightshade incorporated into the soil decrease plant height, root length, fresh and dry weights. However, the number of leaves was not affected. Also, yield components were decreased with the highest concentrations used. The reduction of germination, growth, and yield of the broad and common beans increased with higher concentrations of root and leaf aqueous extracts, while that of seeds had limited effect. The strongest allelopathic effect of broad and common beans was caused by root extracts of the highest concentration.

Keywords: aqueous extract, germination test, pot experiment, plant height, yield components

Introduction

Allelopathy is a form of interaction between plants, exerted through a chemical inhibitor produced by a plant helping to intervene in the growth and development of another one (Zeng et al., 2010). These chemicals with allelopathic potential can be released into the environment, under suitable conditions, in sufficient amounts to affect neighboring plants (Tahir, 2011). Allelopathy has beneficial or harmful effects on plants due to the release of allelochemicals which are secondary metabolites, which are present in all plant tissues including leaves, stems, flowers, roots, and seeds (Mohsin et al., 2016).

Solanaceae plants have the ability of allelopathy, and so that the species of plants control the behavior, form, and amount of allelopathic compounds (Mushtaq and Siddiqui, 2018). The genera of this family, *Withania*, *Solanum*, *Iochroma*, *Nicotiana*, and *Datura* are rich sources of sesquiterpenoid phytoalexins (Elakovich, 1987). The toxic alkaloids present in certain species of the family have given it its vernacular name of nightshade. The genus *Solanum* is massive, contains species distributed in many parts of the world. The non-growth or growth depletion of neighboring plants is caused by allelopathic constituents of this genus (Mushtaq and Siddiqui, 2018). The phenomena have been found in the two biologically active glycoalkaloids, solasonine, and

solamargine (Fukuhara and Kubo, 2004). *Solanum nigrum* (L.) is a common and troublesome annual weed in many parts of the world, it is one of the most widespread species in Solanaceae family. *S. nigrum* is a common plant due to its toxic content of Solanine, a glycoalkaloid found in most parts of the plant, with the highest concentrations in the unripened berries (Imad et al., 2017). Although it is considered a rich source of one of the most popular plant poisons, it has also proven to be a pharmacologically prospective reservoir of phytochemicals. Leaves and berries of *S. nigrum* are commonly used in South India for the treatment of gastric ulcers, gastritis, and other gastric problems (Hadi et al., 2017; Chen et al., 2009).

Considering the economic importance, and nutritional facts of leguminous crops, broad and common beans in the world especially in developing countries. The article aimed to determine the effect of aqueous extracts obtained from different parts of the black nightshade, at various concentrations, on the germination, growth and yield of common and broad beans cultivated in Egypt, to assess the plant's allelopathic potential.

Materials and methods

Site of experiments

Two experiments were conducted to study the allelopathic effect of different parts of *S. nigrum*, including germination test and pot experiment. The germination test was carried out in the laboratory of Plant Ecology, Faculty of Science, Zagazig University, Egypt. The pot experiment was carried out in the greenhouse of the faculty under field conditions.

Plant collection

Matured black nightshade was collected at the fruiting stage from an agricultural field west of Zagazig University. The plant was separated into roots, leaves, and seeds then oven-dried and ground into a fine powder using mortar. Broad bean (*V. faba*) and common bean (*Ph. vulgaris*) seeds were obtained from Agricultural Research Centre, Egypt.

Chemical analysis

Quantitative estimation of the four major active principles in root, leaf, and seed powders of black nightshade was carried out. Total alkaloid and saponin were determined according to Obadoni and Ochuko (2001). Total flavonoid content was estimated by the method of Srisawat et al. (2010) and tannin according to Edeoga et al. (2005).

Germination test

This experiment was conducted to explore seed germination, length of the radicle, and plumule of beans under the effect of aqueous extract of roots, leaves, and seeds of black nightshade. Aqueous extract of different plant parts was prepared by adding 20 g of the dried part to 800 ml of distilled water, then put in a magnetic stirrer for 24 h. The extracts were filtered (stock solutions) and stored at 4 °C until use later. From each stock solution, three concentrations were prepared by dilution method (10, 40, and 60%), these concentrations were selected depending on a previous study (Stef et al., 2013).

Ten seeds of both beans were put sparsely in a filter paper covered glass petri dish (10 seeds/petri dish). Each dish was applied by 5 ml aqueous extract of each plant part, whereas 5 ml distilled water was applied to the control. The treatments were arranged with three replicates in a fully randomized system. The petri dishes were kept in the laboratory at room temperature for 10 days, the germinated seeds were counted to calculate the germination percentage, and the length of radicle and plumule were measured.

Pot experiment

The pots used were 30 cm in diameter, and 30 cm high filled with equal amounts of sieved soil (2:1 v/v clay and sand). The soil was incorporated with 10, 20, and 30 g of plant parts powder representing 0.5, 1, and 1.5% (residue/soil, w/w) respectively. Soil without *S. nigrum* residue represented control. Each pot was planted with 5 seeds of broad and common beans. After emergence, the seedlings were thinned to 3 seedlings per pot. The pots were kept free from weeds and irrigated uniformly. Each treatment was replicated 3 times and pots were distributed in a complete randomized design. At 30 days after sowing, growth parameters like plant height, root length, number of leaves, fresh, and dry weights of plants were recorded. At harvest broad and common beans were taken to determine the yield components like the number of pods/plant, number of seeds/pod, and weight of 100 seeds (g).

Statistical analysis

Data were statistically analyzed using SPSS program version 23. The averages of different groups and standard error were calculated. The two-way ANOVA test was performed for comparison between the different studied factors (i.e., Aqueous extract and treatments) and followed by post hoc test using Duncan multiple range (DMR) test for comparisons between means of groups. The means followed by the same letter in each column are not significantly different from each other at the 5% probability level (p value at 0.05).

Results

Chemical analysis of black nightshade revealed the presence of many bioactive secondary metabolites in all parts of this plant, particularly alkaloids which are responsible for suppressing the growth of other plants (*Table 1*).

Table 1. Chemical constituents in different parts of black nightshade

Plant parts	Content (%)			
	Alkaloid	Flavonoid	Tannin	Saponin
Roots	1.68	0.73	0.14	0.30
Leaves	1.39	0.88	0.16	0.28
Seeds	0.962	0.49	0.13	0.27

There was no significant difference between the two leguminous plants in their response to residue and extract of the allelopathic plant for all parameters recorded.

Germination test

The results presented in *Figures 1* and *2* showed clearly the effects of different aqueous extracts of black nightshade, different treatments, and interaction effects on germination percentage, and length of radicle and plumule of beans. These parameters significantly ($p \leq 0.05$) decreased after being treated with roots and leaves aqueous extracts of black nightshade. However, the reduction in these parameters increased with the concentration of aqueous extracts, and depend on the part used in the extract. According to the displayed data, aqueous extract of seeds was found to be non-significant ($p > 0.05$) for all parameters. The highest inhibitory effects were exhibited by root extract particularly at 60% concentration, compared to control. The lowest seed germination and length of radicle and plumule of broad bean were 68.2%, 2.80 cm, and 3.65 cm respectively for aqueous root extract (*Fig. 1a, b, c*). In common beans, seed germination and length of radicle and plumule significantly lowered to 84.31%, 3.08 cm, and 4.36 cm respectively for those treated with root extract (*Fig. 2a, b, c*).

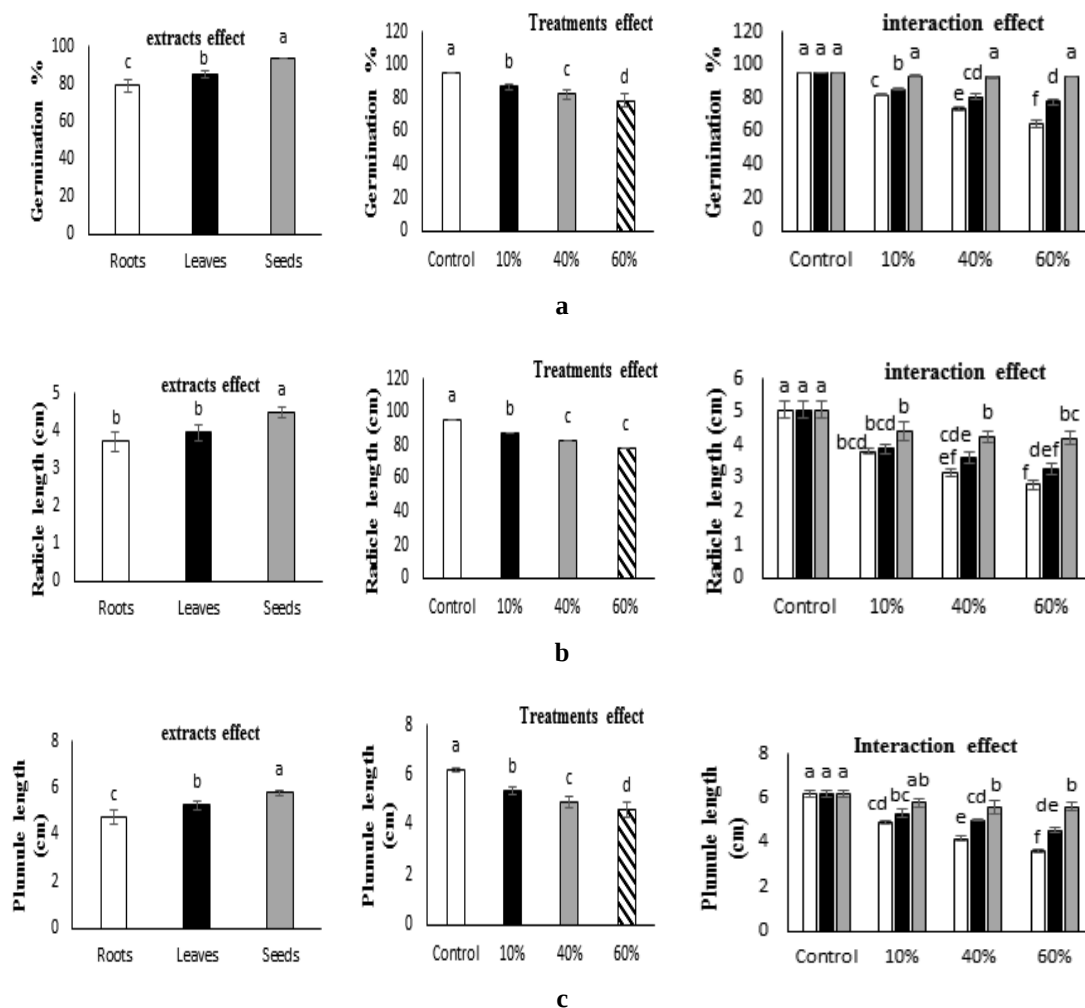


Figure 1. Effects of black nightshade aqueous extracts, different treatments and interaction effect on broad bean, a: germination percent (%); b: radical length (cm); c: plumule length (cm). Error bars indicating the standard error (SE) of three replicates, are significantly* $P < 0.05$

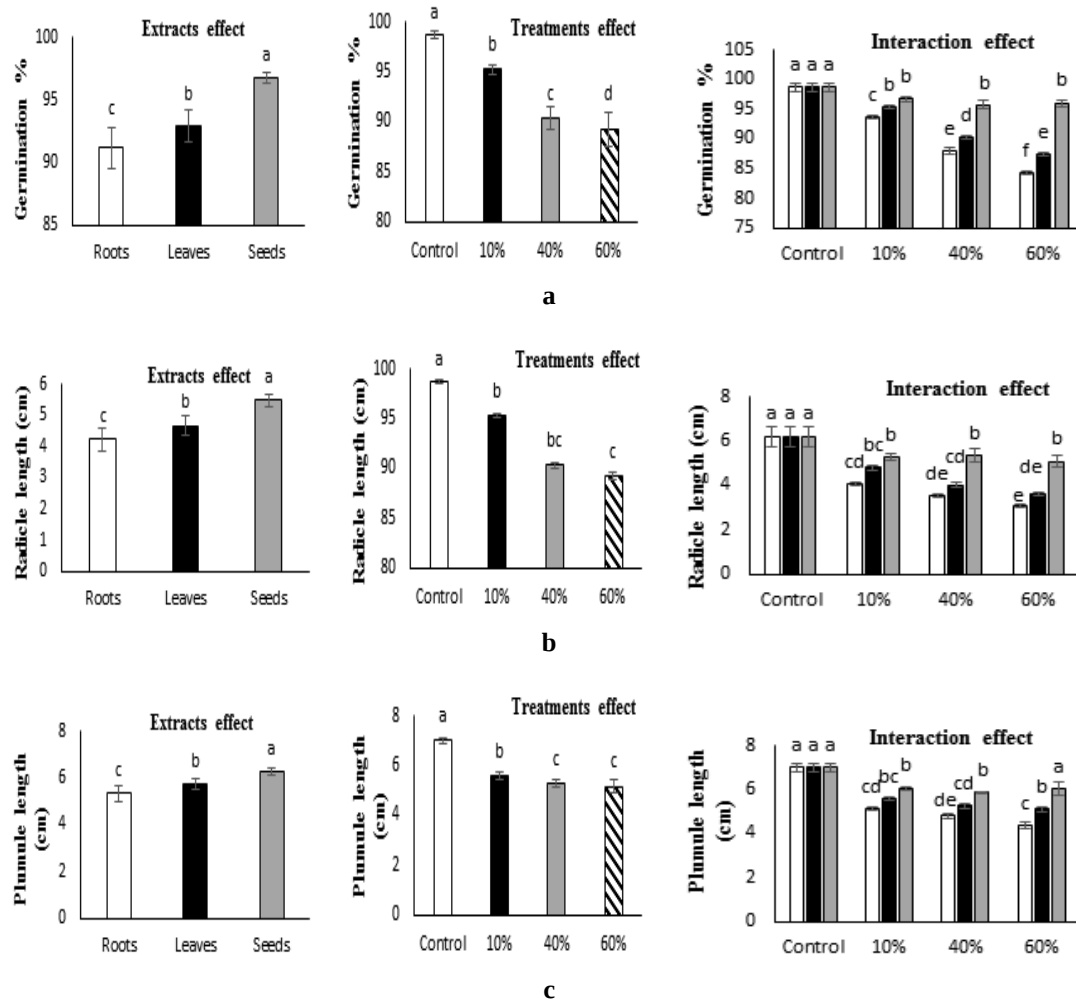


Figure 2. Effects of black nightshade aqueous extracts, different treatments and interaction effect on common bean, a: germination percent (%); b: radical length (cm); c: plumule length (cm). Error bars indicating the standard error (SE) of three replicates, are significantly* $P < 0.05$

Pot experiment

The results presented in Table 2 and 3 showed the effect of root, leaf, and seed powders incorporated into the soil on growth parameters of beans. All parameters investigated in this study except the number of leaves significantly ($p \leq 0.05$) decreased in both plants treated with different powders of black nightshade.

Relative to control, plant height of both beans significantly decreased, and this reduction was increased as different powders increased in the soil. The greatest reduction in plant height was observed when root powder was added to the soil at 1.5%. The plant height of the broad bean lowered to 27.05 cm (Table 2) and the common bean decreased to 26.1 cm (Table 3).

The data of root length showed the same trend of plant height, the greatest reduction in root length was recorded when root powder was incorporated into the soil. The root length of broad and common beans in control was 16.23 and 16.53 cm, respectively. The root length decreased to 10.06 cm in the broad bean, and 13.07 cm in the common bean ($p \leq 0.05$).

Table 2. Growth parameters of broad bean under the effect of different concentrations of black nightshade residues

Treatments (%)	Plant height (cm)	Root length (cm)	Fresh wt. (g)	Dry wt. (g)	Leaves no.
R 0.5	29.58 ± 0.32 ef	12.03 ± 0.03 fg	7.44 ± 0.22 de	2.19 ± 0.13 fg	10 ± 0.33 a
R 1	28.55 ± 0.22 g	10.75 ± 0.25 h	6.64 ± 0.17 f	1.81 ± 0.12 g	9.66 ± 0.57 a
R 1.5	27.05 ± 0.12 h	10.06 ± 0.08 h	6.01 ± 0.01 g	1.13 ± 0.08 h	9.66 ± 0.33 a
Mean	30.09 ± 3.09 c	12.27 ± 2.51 c	7.41 ± 1.45 c	2.226 ± 1.03 c	9.91 ± 0.67 a
L 0.5	31.33 ± 0.33 d	13.58 ± 0.25 d	8.12 ± 0.11 c	3.02 ± 0.03 cd	10 ± 0.57 a
L 1	30.18 ± 0.12 e	12.50 ± 0.28 ef	7.98 ± 0.02 cd	2.66 ± 0.07 de	10 ± 0.33 a
L 1.5	29.35 ± 0.19 f	11.54 ± 0.23 g	7.11 ± 0.11 ef	2.28 ± 0.08 ef	9.66 ± 0.33 a
Mean	31.42 ± 2.22 b	13.46 ± 1.87 b	8.19 ± 0.97 b	2.933 ± 0.6 b	10.00 ± 0.74 a
S 0.5	34.50 ± 0.28 ab	15.51 ± 0.28 ab	9.01 ± 0.09 ab	3.32 ± 0.16 ab	10.33 ± 0.66 a
S 1	33.94 ± 0.05 ab	14.50 ± 0.28 ab	8.80 ± 0.1 ab	3.05 ± 0.05 bc	10.33 ± 0.33 a
S 1.5	32.31 ± 0.33 c	12.77 ± 0.39 bc	8.06 ± 0.03 cd	2.80 ± 0.11 d	10.30 ± 0.66 a
Mean	33.90 ± 1.08 a	14.75 ± 1.43 a	8.86 ± 0.63 a	3.231 ± 0.44 a	10.33 ± 0.78 a
Control	34.84 ± 0.31 a	16.23 ± 0.17 ab	9.58 ± 0.34 a	3.75 ± 0.23 ab	10.33 ± 0.33 a

Mean ± standard error based on ANOVA analysis. Means in the same raw followed by the same letter in each column are not significantly different from each other at the 5% probability level (p value at 0.05) according to Duncan Multiple Range Test (DMRT). R: root; L: leaf; S: seed

Table 3. Growth parameters of common bean under the effect of different concentrations of black nightshade residues

Treatments (%)	Plant height (cm)	Root length (cm)	Fresh wt. (g)	Dry wt. (g)	Leaves no.
R 0.5	27.55 ± 0.22 ef	14.46 ± 0.15 d	6.76 ± 0.15 def	1.92 ± 0.07 d	7.66 ± 0.33 ^a
R 1	26.87 ± 0.13 g	13.78 ± 0.14 e	6.16 ± 0.09 fg	1.34 ± 0.09 f	8.00 ± 1 a
R 1.5	26.10 ± 0.1 h	13.07 ± 0.07 f	5.67 ± 0.08 g	0.94 ± 0.06 g	7.00 ± 1 a
Mean	27.64 ± 1.57 c	14.46 ± 1.37 c	6.78 ± 1.17 c	1.85 ± 0.89 c	7.58 ± 1.16 a
L 0.5	28.81 ± 0.1 c	15.26 ± 0.13 bc	7.35 ± 0.18 cd	2.30 ± 0.14 c	8.00 ± 0.57 a
L 1	27.87 ± 0.23 de	14.80 ± 0.1 bc	6.91 ± 0.08 de	2.01 ± 0.01 d	7.66 ± 0.33 a
L 1.5	27.18 ± 0.17 fg	14.28 ± 0.15 de	6.51 ± 0.08 ef	1.64 ± 0.11 e	7.66 ± 0.88 a
Mean	28.48 ± 1.16 b	15.22 ± 0.91 b	7.32 ± 0.84 b	2.28 ± 0.62 b	7.75 ± 0.87 a
S 0.5	29.91 ± 0.2 ab	16.15 ± 0.14 a	8.25 ± 0.21 ab	3.03 ± 0.03 ab	8.00 ± 0 a
S 1	29.39 ± 0.23 ab	15.39 ± 0.19 ab	7.77 ± 0.13 bc	2.83 ± 0.1 b	7.66 ± 0.66 a
S 1.5	28.26 ± 0.13 d	15.07 ± 0.07 bc	7.21 ± 0.12 cd	2.46 ± 0.02 c	8.00 ± 0.57 a
Mean	29.40 ± 0.79 a	15.78 ± 0.67 a	7.94 ± 0.61 a	2.88 ± 0.3 a	7.83 ± 0.72 a
Control	30.06 ± 0.19 a	16.53 ± 0.25 a	8.53 ± 0.34 a	3.19 ± 0.07 a	7.66 ± 0.33 a

Mean ± standard error based on ANOVA analysis. Means in the same raw followed by the same letter in each column are not significantly different from each other at the 5% probability level (p value at 0.05) according to Duncan Multiple Range Test (DMRT). R: root; L: leaf; S: seed

Fresh and dry weights of both plants follow the same approach as for the plant height and root length. Relative to control, root powder at concentration 1.5% showed the greatest reduction of fresh weight of both plants. The lowest fresh weight of broad bean

was 6.01 g while the highest value was 9.58 g observed in control (Table 2). In common beans, the lowest fresh weight was 5.67 g while the highest value was 8.53 g which was found in control (Table 3). The highest dry weight of broad and common beans were 3.75 and 3.19 g observed in control while the lowest values 1.13, and 0.94 g respectively were found in pots with 1.5% root powder.

The data recorded in beans revealed that the greatest reduction in the previous parameters was observed when roots of black nightshade were applied then the leaves while seeds had limited influence. It is suggested to indicate that the measured properties were not significantly different from the control after treated with seed residues this may be attributed to that this organ has the smallest amount of alkaloid.

For the number of leaves, there was no significant difference between different powders treated with different rates in the soil of both plants ($p > 0.05$). The results showed that the incorporated powders of *S. nigrum* into the soil at rates 0.5, 1, and 1.5% did not affect the number of leaves of broad and common beans.

The results of yield components of beans were recorded in Table 4. All applied powders of black nightshade incorporated into the soil (0.5, 1, and 1.5%) significantly decreased all yield components except the seed powder. Root and leaf powders of black nightshade produced lower yield components than control. Therefore, it could be concluded that *S. nigrum* roots and leaves residues (at 0.5, 1, and 1.5%) incorporated into soil caused a reduction in the yield of broad and common beans.

Table 4. Effect of different residues concentrations of black nightshade on the yield of broad and common beans

Treatments (%)	Broad bean			Common bean		
	No. of pods	No. of seeds	Wt. of 100 seeds (g)	No. of pods	No. of seeds	Wt. of 100 seeds (g)
R 0.5	5.71±0.19 d	3.42±0.16 cd	79.63±0.77 c	7.91±0.13 b	4.95±0.06 c	35.23±0.12 c
R 1	4.27±0.21 e	2.66±0.11 e	75.86±0.26 d	6.86±0.22 d	4.02±0.18 d	32.56±0.31 d
R 1.5	3.27±0.14 f	1.85±0.10 f	72.38±0.14 e	6.10±0.05 e	3.28±0.17 e	30.66±0.17 e
Mean	5.15±1.64 c	3.06±0.98 c	77.79±4.31 c	7.45±1.14 c	4.43±0.93 b	37.62±0.4 a
L 0.5	6.30±0.25 c	3.80±0.10 bc	81.46±0.4 b	8.17±0.09 b	5.05±0.04 bc	36.13±0.20 b
L 1	5.69±0.21 d	3.08±0.04 de	79.67±0.66 c	7.38±0.19 c	4.28±0.22 d	34.73±0.37 c
L 1.5	4.30±0.15 e	2.18±0.10 f	74.98±0.19 d	6.65±0.18 d	3.79±0.14 de	32.63±0.18 d
Mean	5.91±1.2 b	3.34±0.86 b	79.85±3.29 b	7.78±0.92 b	4.65±0.74 b	37.62±0.4 a
S 0.5	7.05±0.07 ab	4.27±0.12 a	82.33±0.25 a	8.01±0.06 a	5.04±0.17 a	37.90±0.09 a
S 1	6.68±0.24 bc	3.98±0.01 ab	83.08±0.11 a	8.10±0.13 a	5.12±0.27 ab	37.66±0.1 a
S 1.5	6.98±0.21 ab	4.14±0.22 ab	82.44±0.31 a	8.03±0.13 a	4.39±0.27 abc	36.3±0.17 a
Mean	7.02±0.38 a	4.18±0.27 a	83.33±0.39 a	8.96±0.18 a	5.58±0.4 a	37.62±0.4 a
Control	7.36±0.2 a	4.32±0.19 a	83.29±0.21 a	8.93±0.11 a	5.49±0.25 abc	37.62±0.39 a

Mean ± standard error based on ANOVA analysis. Means in the same raw followed by the same letter in each column are not significantly different from each other at the 5% probability level (p value at 0.05) according to Duncan Multiple Range Test (DMRT). R: root; L: leaf; S: seed

Discussion

The high nutritional value and protein content make broad and common beans, belonging to the family Leguminosae, the most important crops in the world, especially in developing countries. Weeds cause a substantial decline in agriculture production

through direct and indirect effects. Therefore, it is important to determine the effect of these weeds on the growth of crops. Also, *S. nigrum* is distributed in some field crops in Egypt.

According to the displaying data, black nightshade affects germination, growth, and yield of beans negatively. The probable reason for *S. nigrum* potent allelopathic activity may be due to the presence of many bioactive secondary metabolites in this plant, particularly alkaloids which are responsible for suppressing the growth of other plants (Sabh and Ali, 2010). The inhibitory effect on yield and yield parameters is directly proportional to the increase in the concentration used and this might be due to the presence of toxic allelochemicals like alkaloids. According to Girija and Gowri (2008) the leaf, and fruit extracts of *S. nigrum* contain alkaloids, that may suppress the germination, radicle length, and total protein content of *P. sativum*, *E. coracona*, and *T. foenum*.

Eltayeb et al. (1997) reported that solasodine occurs in roots, stems, leaves, and fruits of *S. nigrum*. The lowest levels are registered in stems and the highest in leaves. In vegetative organs, and particularly in roots, absolute amounts of solasodine rise steadily with age but decrease in fruit. From the results obtained, we found that the root of *S. nigrum* had the strongest allelopathic effect on germination, yield, and growth parameters compared to leaves, and seeds. Likewise, Imad et al. (2017) found that the alkaloidal content of plant parts changes during the development of *S. nigrum*, the absolute amount of alkaloid per leaf increased during leaf development, whereas the concentration declined. Both the concentration and the absolute amount of solasodine per fruit decrease with fruit maturation. Generally, Debnath et al. (2016) found that leaves aqueous extract exhibit more allelopathy for seedlings than those from the flowers of *S. sisymbirifolium* (Lam.) on *V. radiata*, *A. mangostanus*, and *B. campestris*, whereas it is less effective on *O. sativa*. However, the leachates extracted at the flowering stage from *N. plumbaginifolia* provided the greatest growth inhibition and biochemical parameters for seedlings of sunflower depending on concentration (Singh et al., 2015). Also, Morais et al. (2013) displayed that ethanol extract and fractions obtained from ripe fruits of *S. lycocarpum* showed allelopathic potential on the growth of onion radicles; inhibitory effect on the growth of hypocotyls, and radicles of lettuce. The results of germination and pot experiments showed a great reduction increased with the concentration of allelopathic plant, this agrees with Bosch et al. (2004) who showed that leaf leachates of *S. mauritanum* (Scop.) reduced germination in *H. stricta* in a concentration-dependent manner. The growth of both beans was suppressed and radicle and plumule length were decreased, these may be attributed to the allelochemicals which disrupt the growth hormones. El-Shora et al. (2015) found that radicle and plumule of fenugreek, a medicinal plant showed a sensitive response to allelochemicals from aqueous extracts which might have interfered with hormones that encourage growth, development, cell elongation, and cell division especially on younger active root tips.

All growth parameters, plant height, root length, fresh and dry weights except number of leaves reduced after applying root and leaf residues in soil. Allelochemicals from leachates of *S. nigrum* might have interfered with photosynthesis leading to drastic changes in the physiology of plants. El-Shora et al. (2018) found that the decline in photosynthesis may have contributed to decreased growth of plants and decreased accumulation of fresh biomass due to decreased water content in the species studied. In this regard, Fikreyesus et al. (2011) reported that tomato root elongation was inhibited

by extracts of *Eucalyptus camandulensis*, and Gulzar et al. (2014) observed that the root length of *C. album*, *M. alba*, and *N. plumbaginifolia* decreased as the concentration of *C. sophera* increased. The results of the study are consistent with Hamidi et al. (2008) who observed the allelopathic ability of *H. spontaneum* residues incorporated into the soil on the length of seedling, and the dry weight of *T. aestivum*. Also, Ismail and Siddiqui (2011) found that seedling length and weight of *O. sativa* were suppressed by residues of *C. iria*. The greatest reduction in growth parameters was observed at 1.5% concentration residue of *S. nigrum*, and this was consistent with the results of Zohaib et al. (2014) who found that the inhibitory effects of five leguminous weeds against rice caused by the highest concentration of water extracts and residues of these weeds.

Conclusion

Allelopathy can cause substantial decline in the growth and yield of crops. This study was devoted to determining the allelopathic effects of different parts of black nightshade on some leguminous crops, broad and common beans. According to the results of germination and pot experiments, it was found that roots of black nightshade had the highest inhibitory effect on beans. Germination, growth parameters except for the number of leaves and yield exhibited a high level of inhibition by the highest concentration of roots. While seeds showed a limited influence on these parameters. The decline of growth and yield requires further studies on the germinated seeds related to the biosynthesis of secondary metabolites at molecular level. The results confirmed that *S. nigrum* has an allelopathic effect on broad and common beans which depends on plant parts and concentration used in the extract preparation.

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