



Impact of Intercropping of Different Crops with Two Faba Bean Cultivars on Infestation with Broomrape

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ABSTRACT

Background: Broomrape (*Orobanche crenata* Forsk.) is a major biotic stress of faba bean and affects the yield adversely. The impact of intercropping to curb the spread and progress of parasite was tested by using two faba bean cultivars under heavily soil infested conditions of broomrape. The present study was aimed to evaluate intercropping of fenugreek, flax, garlic or radish with two faba bean cultivars on broomrape incidence to increase faba bean yield, land usage and economic return in heavy soil infestation of broomrape.

Methods: This study was carried out at Giza Agricultural Research Station (Lat. 30°00' 30" N, Long. 31°12' 43" E, 26 m a.s.l), Agricultural Research Center, Giza, Egypt. During winter seasons of 2017/2018 and 2018/2019, fourteen the treatments were the combinations of two faba bean cultivars (Nubaria 1 and Nubaria 2) and four intercrops (fenugreek, flax, garlic and radish), as well as sole plantings of all the tested crops under heavily infestation of broomrape were tested in field conditions. A split-plot design with three replicates was used. Faba bean cultivars were assigned in the main plots and intercropping and sole plantings of faba bean were arranged in sub plots.

Result: Results showed that Nubaria 2 had two mechanisms to broomrape offensive, mainly including cell wall build-up and stamping of vascular tissues. It also had higher soil oxalic and ascorbic acid concentrations than Nubaria 1. Sole Nubaria 1 rhizosphere had higher soil formic acid whereas, fenugreek + Nubaria 2 had higher soil oxalic, ascorbic and salicylic acids concentrations in faba bean rhizosphere. Lower number of spikes per m² and their dry weight per m² were obtained by radish + Nubaria 2 followed by flax + Nubaria 2. Radish + Nubaria 2 recorded higher land equivalent ratio and economic return followed by flax + Nubaria 2. Growing one row of radish or flax between two rows of Nubaria 2 in ridges 60 cm width could be an integrated control strategy to increase faba bean productivity, land usage and economic return under heavily soil infested with broomrape.

Key words: Broomrape infestation, Economic return, Faba bean cultivars, Intercropping, Land usage, Soil organic acids.

INTRODUCTION

Most of the life cycle of broomrape (*Orobanche crenata* Forsk.) depends on connection with the root of faba bean (*Vicia faba* L.) plant and is, therefore very difficult to be controlled either by agronomic practices or herbicides (Goldwasser *et al.*, 2003). If this parasite reaches the root of faba bean plant, the apex penetrates the root tissue and subsequently makes a connection with the vascular tissues in the root and a haustorium is developed on faba bean plant. Due to unique nature of parasite, the available methods of control against root parasitic plants have not proven as effective, economical and applicable as predicted (Goldwasser and Kleifeld, 2004). According to Attia *et al.* (2009), it should be sowing of faba bean around November to produce higher seed yield and its attributes by escaping from broomrape infestation period.

Previous studies (Abdalla and Darwish, 2008; Ismail, 2013; Kandil *et al.*, 2015; Trabelsi *et al.*, 2015; Mabrouk *et al.*, 2016 and Safina, 2017) reported that there were significant differences among faba bean genotypes for number and dry weight of *Orobanche* spikes, the parasitism index, total chlorophyll, number of pods per plant and seed yield per unit area. Thus, broomrape control mainly depends on the use of tolerant cultivars of the host plants as reported by Elsakhawy *et al.* (2020). Accordingly, growing fenugreek (*Trigonella foenumgraecum* L.), flax (*Linum usitatissimum* L.), garlic (*Allium sativum* L.), or radish (*Raphanus sativus*

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L.) with faba bean in the same ridge could enhance systemic defense responses of faba bean plant to control broomrape infestation in their fields. This biological interaction can reduce the appearance of the broomrape in the field or inhibited the broomrape growth during its parasitism on faba bean roots.

Trap or companion crops can be defined as false-host plants turn out compound exudates that support broomrape germination, but they don't permit the broomrape to attach the roots or develop tubercles and do not hamper the growth and yield of crops (Dhanapal *et al.*, 1996). In this concern, Abbes *et al.* (2008) recognized flax and fenugreek as trap crops after observing broomrape seed germination on their roots in vitro, but no tubercle formation was observed on

roots of any plant after 67 days from inoculation. Meanwhile, Abu-Shall and Ragheb (2014) revealed that intercropping radish with broad bean recorded higher productivity of broad bean with lower broomrape infestation followed by garlic and fenugreek than sole broad bean due to some chemicals that secreted from radish, garlic and fenugreek roots. The trap or companion crop can be from the same or another family group, than that of the main crop. According to Aksoy *et al.* (2016), flax was the most effective treatment by decreasing 52% and 71% in shoot number and 55% and 26% in dry weight of *O. crenata* in the first and second seasons, respectively, in legume crop fields. However, Zeid and Komeil (2019) showed that growing fenugreek with faba bean cultivars Giza 843 and Misr 3 reduced seed germination of broomrape by almost 30% than their sole cultures, meanwhile this percentage reached 73% by growing radish with Misr 3 only.

Accordingly, some organic acids in the rhizosphere of intercropped faba bean could play an important role to tolerate broomrape infestation through choice the suitable intercropping system. In this concern, El Bassiouny *et al.* (2005) reported that spray the bean plants after 45 and 60 days of transplanting with ascorbic acid concentrations at 45 and 60 days after sowing, length of internodes and number of nodes at last picking, weight of pods per plant whereas ascorbic acid 200 ppm has given best results for yield per plot and yield per hectare. Thus, ascorbic acid has been shown to play multiple roles in plant growth such as in cell division, cell wall expansion and other developmental processes (El-khayate *et al.*, 2015). It also acts as an antioxidant (Kasim Wedad *et al.*, 2017). Particularly, Madany *et al.* (2020) found that the use of natural substances like (salicylic and ascorbic acid) improved and increased plant growth. The accumulation of citric, lactic, maleic and succinic acids was markedly improved by treatment with silicon dioxide nanoparticles to reduce broomrape infection. Consequently, organic acid plays an important role in plant metabolism as an early source of photosynthesis and precursors of the synthesis of many other compounds (Muktadir *et al.*, 2020).

Maleic hydrazide (systemic herbicides) reduced broomrape spikes at 0.25 - 0.75 kg a.i./ha applied at 30 or 40 days after transplanting tobacco (Dhanapal *et al.*, 1996). Oxalates can be found widely in many plant families in relatively small amounts, but some plant species can accumulate oxalate in a range of 315% (w/w) of their dry weight (Nakata, 2003; Franceschi and Nakata, 2005). Moreover, endogenous salicylic is a plant hormone that has important physiological roles in promoting plant growth and increasing the efficiency of photosynthesis and flowering (Hayat *et al.*, 2007). It may be involved in the expression of specific proteins or defense-related enzymes (Zawoznik *et al.*, 2007). It plays an important role in the regulation of plant growth, development and immune responses (An and Mou, 2011). It participates in many physiological processes in plants as it improves plant growth, enzyme activity, ion absorption and transport (Jalal *et al.*, 2012). It is gaining attention as a regulator of plant metabolism and

physiological processes and is involved in both local and systemic plant defense responses in plants (Kawano and Bouteau, 2013). On the other hand, Laursen and Poudyal (2015) showed that an active formic acid dehydrogenase results in a rapid conversion of formic acid to photosynthetic CO₂ fixation. In other words, the photocatalytic reduction of CO₂ into methane, formaldehyde, formic acid and methanol as main products has been reported over several heterogeneous and homogeneous catalysts. Therefore, the present study was conducted to evaluate intercropping of fenugreek, flax, garlic or radish with two faba bean cultivars on broomrape incidence to increase faba bean yield, land usage and economic return in heavy soil infestation of broomrape.

MATERIALS AND METHODS

The study was carried out at Giza Agricultural Research Station (Lat. 30°00' 30" N, Long. 31°12' 43" E, 26 m a.s.l), Agricultural Research Center, Giza, Egypt during winter seasons 2017/2018 and 2018/2019 seasons to evaluate intercropping of fenugreek, flax, garlic or radish with two faba bean cultivars on broomrape incidence to increase faba bean yield, land usage and economic return in heavy soil infestation of broomrape. Fourteen the treatments were the combinations between two faba bean cultivars (Nubaria 1 "Individual plant selection from Giza Blanca" and Nubaria 2 "ILB 1550 x Radiation 2095 / 76") and four intercrops (fenugreek, flax, garlic and radish), as well as sole plantings of all the tested crops. Seeds of faba bean cultivars Nubaria 1 and Nubaria 2, fenugreek cultivar Giza 2, flax cultivar Sakha 3, garlic cultivar Sids 40 and white radish cultivar Balady were planted on 5th November and 24th October in 2017 and 2018 seasons, respectively. Faba bean plants were harvested on 28th April and 12th April in 2018 and 2019 seasons, respectively. Meanwhile, fenugreek seed, flax seed, garlic bulbs and radish seed yields were harvested on 15th March and 9th March, 5th April and 30th March, 15th March and 7th March and 5th April and 28th March in 2018 and 2019 seasons, respectively. The plant densities of intercropped crops were 50, 50, 67 and 50% of the recommended sole cultures for fenugreek, flax, garlic and radish, respectively. The treatments were as follows:

Intercrops

Intercropping system

Two rows of faba bean were grown (two plants/hill spaced at 25 cm) in both sides of ridges 60 cm width (four seeds/hill) and were later thinned to 16 plants per one meter, meanwhile one row of fenugreek seeds (8.5 g), flax seeds (15 g), radish seeds (11 g) or two rows of garlic cloves (one clove/hill spaced at 10 cm) or were drilled in the middle of ridge.

Sole plantings

➤ Sole faba bean

Two rows of faba bean were grown (two plants/hill spaced at 25 cm) in both sides of ridges 60 cm width (four seeds/hill) and were later thinned to 16 plants per one meter.

➤ Sole intercrops

Two rows of fenugreek seeds (17 g), flax seeds (30 g) or radish seeds (22 g) were drilled in both sides of ridges 60 cm width, meanwhile three rows of garlic cloves were grown (one clove/hill spaced at 10 cm) in ridges 60 cm width.

The preceding summer crop was maize in both seasons. Furrow irrigation was the irrigation system in the region. All cultural practices such as fertilization and irrigation were performed whenever necessary. Calcium super phosphate (15.5% P_2O_5) at rate of 476 kg/ha and potassium sulfate (K_2SO_4 "48.0 % K_2O ") at rate of 238 kg/ha were applied during soil preparation. Mineral nitrogen (N) fertilizer was applied for faba bean in form of ammonium nitrate (NH_4NO_3 "33.5% N") at rate of 107.1 and 107.1 kg/ha under intercropping and sole plantings, respectively, in three equal doses before the first, second and third irrigation. Also, Mineral N fertilizer was applied for fenugreek in form of ammonium nitrate (NH_4NO_3 "33.5% N") at rate of 53.5 and 107.1 kg/ha under intercropping and sole plantings, respectively, in three equal doses before the first, second and third irrigation. With regard to radish, mineral N fertilizer was applied in form of ammonium nitrate (NH_4NO_3 "33.5% N") at rate of 285.6 and 571.2 kg/ha under intercropping and sole plantings, respectively. Meanwhile, mineral potassium (K) fertilizer was applied for radish in form of K_2SO_4 at rate of 238 and 476 kg/ha under intercropping and sole plantings, respectively. Mineral N and K fertilizers for radish were applied in three equal doses before the second, third and fourth irrigation. However, mineral N fertilizer was applied for garlic in form of NH_4NO_3 at rate of 214.2 and 285.6 kg/ha under intercropping and sole plantings, respectively, in two equally doses before the second and third irrigation. With respect to flax, mineral N fertilizer was applied for flax in form of NH_4NO_3 at rate of 89.2 and 178.5 kg/ha under intercropping and sole plantings, respectively, in two equally doses before the second and third irrigation. A split-plot design with three replicates was used. Faba bean cultivars were assigned in the main plots and intercropping and sole plantings of faba bean were arranged in sub plots. The area of sub-plot was 10.8 m², each plot consisted of 6 ridges and each ridge was 3.0 m in length and 0.6 m in width.

Parameters

➤ Anatomical characteristic of faba bean root cells

At 120 days from planting, the cross-section of faba bean root was photographed by light microscopy in Plant Dept., Faculty of Sciences, Cairo University, Giza, Egypt.

➤ Soil organic acids

The determination of oxalic, citric, ascorbic, maleic, formic and salicylic acids (mg/l) was determined in the faba bean rhizosphere for each plot. These analyses were recorded by Environment, Water and Soil Research Institute, Agricultural Research Center, Giza, Egypt at 120 days from planting. These analyses were recorded by Environment, Water and Soil Research Institute, Agricultural Research Center, Giza, Egypt.

➤ Chlorophyll pigments

The leaf chlorophylls a (mg/g FW) and b (mg/g FW) contents were analyzed by the General Organization for Agricultural Equalization Fund, ARC, Giza, Egypt according to Holden (1965) at 120 days from planting.

➤ Broomrape infestation

At 120 days from planting, number of emerged broomrape spikes per m²: it was counted per m² and referred as number of emerged broomrape spikes per host plant in each treatment (Rubiales *et al.*, 2006) and dry weight of broomrape spikes per m² (g). Penetration of broomrape into vascular system of faba bean roots was done by using SEM Model Quanta 250 FEG (Field Emission Gun) in the Egyptian Mineral Resources Authority Central Laboratories Sector. Meanwhile, surface preparations and transverse sections of the root of faba bean cultivars were used for observation of various microscopic features.

➤ Faba bean seed yield and its attributes

At 173 and 168 days from planting in the first and second seasons, respectively, harvested plants (%) were estimated by number of plants at harvest compared to their numbers at planting in each plot. Samples of five plants were collected randomly from each plot to estimate the following criteria: plant height (cm), number of branches/plant, number of pods/plant, seed yield/plant (g) and 100-seed weight (g). Percentage of the harvested plants was estimated by counting number of faba bean plants at harvest on the basis of the experimental plot and expressed as percentage of the harvested plants per ha (%). Seed yield/ha (t) was recorded on the basis of the experimental plot and expressed as ton per hectare (t/ha).

➤ Intercrop yield

At harvest, fenugreek seed, flax seed, garlic bulbs and radish seed yields per ha (t) at 130 and 135, 150 and 156, 130 and 133 and 150 and 154 days from planting in 2017 and 2018 seasons, respectively, were recorded on the basis of the experimental plot and expressed as tons per hectare (t/ha).

➤ Competitive relationships and intercropping economic advantage

Land equivalent ratio (LER): It defines as the ratio of area needed under sole cropping to one of intercropping at the same management level to produce an equivalent yield (Willey, 1979). It is calculated as follows:

$$LER = (Y_{ab}/Y_{aa}) + (Y_{ba}/Y_{bb}),$$

where

Y_{aa} = Pure stand yield of crop a (faba bean), Y_{bb} = Pure stand yield of crop b (fenugreek, flax, garlic or radish), Y_{ab} = Intercrop yield of crop a (faba bean) and Y_{ba} = Intercrop yield of crop b (fenugreek, flax, garlic or radish). To evaluate the economic viability, total return and monetary advantage index (MAI) were calculated as follows: Total return per ha (USD) = Faba bean seed yield x price of faba bean seed + intercrop yield x price of intercrop yield. The prices of faba bean seeds and fenugreek seeds and flax seeds, garlic

bulbs were presented by Bulletin of Statistical Cost, Production and Net Returns (2019). Meanwhile, the price of radish seeds was presented by market prices (2019). One ton of faba bean seeds, fenugreek seeds, flax seeds, garlic bulbs and radish seeds were 720, 840, 614, 186 and 1250 USD, respectively. MAI, it suggests that the economic assessment should be in terms of the value of land saved; this could probably be most assessed on the basis of the rentable value of this land. MAI was calculated according to the formula, suggested by Willey (1979).

$$\text{MAI} = [\text{Value of combined intercrops} \times (\text{LER} - 1)] / \text{LER}$$

MAI value indicates the profit of the cropping system.

Statistical analysis

The statistical analysis of variance according to Snedecor and Cochran (1980) and the least significant differences (LSD) at 5% level of significance tests were done according to Freed (1991).

RESULTS AND DISCUSSION

Anatomical characteristic of faba bean root cells

The vascular system in faba bean root cells is shown in Fig (1). It can play an important role in tolerance of broomrape infestation. Before broomrape infestation, the root cells of Nubaria 1 or Nubaria 2 contain interstitial spaces

that allow the passage of water, nutrients and metabolites to the different parts of the plant with no negative effect on the vascular cylinder (transport vessels; xylem and phloem). Meanwhile, the infestation with broomrape increased the thickness of root cell wall of Nubaria 2 and almost complete closure of the interstitial spaces of its root (stamping) to prevent the translocation of water and photosynthates from the plant to the parasite. These results show that the vascular system of Nubaria 2 root cells played a major role in tolerance of broomrape infestation than the other one. Several pleading mechanisms have been reported to tolerate broomrape infestation, mainly including cell wall build-up and stamping of vascular tissues (Pérez-De-Luque *et al.*, 2005 and 2006).

Soil organic acid

> Faba bean cultivars

Faba bean cultivars significantly affected soil oxalic and ascorbic acid concentrations in faba bean rhizosphere, meanwhile soil citric, maleic, formic and salicylic acid concentrations were not affected (Table 1). These results could be due to the genetic makeup of faba bean cultivars.

> Intercropping systems

Soil organic acid concentrations were significantly affected by intercropping systems (Table 1). Sole faba bean had a higher soil formic acid concentration in the rhizosphere

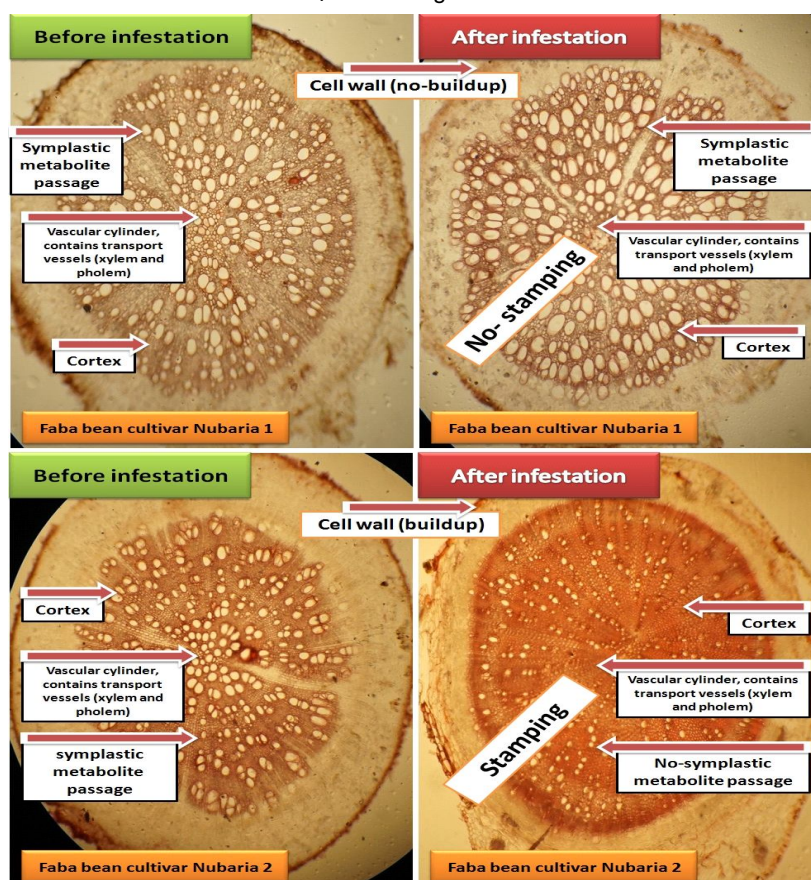


Fig 1: Vascular system of root cells of faba bean cultivars before and after infestation with broomrape.

Table 1: Effect of faba bean cultivars, intercropping systems and their interaction on soil organic acids concentrations in 2018/2019 season.

Faba bean cultivar	Intercropping system	Concentration (mg/l)					
		Soil oxalic acid	Soil citric acid	Soil ascorbic acid	Soil maleic acid	Soil formic acid	Soil salicylic acid
Nubaria 2	Fenugreek	259.50	101.60	2.80	1.30	205.70	3.90
	Flax	163.50	109.00	13.50	3.40	198.60	4.60
	Garlic	101.90	102.20	4.40	1.80	544.20	4.40
	Radish	182.40	101.60	9.00	2.80	184.40	5.10
	Sole faba bean	97.20	100.30	2.40	0.90	1209.20	3.70
	Mean	160.90	102.14	6.42	2.04	468.42	4.34
Nubaria 1	Fenugreek	225.90	82.10	0.70	0.40	279.70	3.20
	Flax	120.90	105.60	5.80	1.30	248.80	4.30
	Garlic	83.70	86.30	1.20	0.60	503.50	3.90
	Radish	105.60	99.70	3.60	1.10	195.00	4.30
	Sole faba bean	76.10	82.10	0.60	0.10	1288.60	3.10
	Mean	122.44	91.16	2.38	0.70	503.12	3.76
Average of intercropping system	Fenugreek	242.70	91.85	1.75	0.85	242.70	3.55
	Flax	142.20	107.80	9.65	2.35	223.70	4.45
	Garlic	92.80	94.25	2.80	1.20	523.85	4.15
	Radish	144.00	100.65	6.30	1.95	189.70	4.70
	Sole faba bean	86.65	88.70	1.50	0.50	1248.90	3.40
F test 0.05 Faba bean cultivar		*	N.S.	*	N.S.	N.S.	N.S.
L.S.D. 0.05 Intercropping system		16.79	10.21	1.37	0.81	11.85	0.23
L.S.D. 0.05 Interaction		23.75	N.S.	1.94	N.S.	16.76	0.74

followed by garlic + faba bean than the others. It is evident that sole faba bean do not excrete the other organic acids in the soil except in a small percentage due to their higher consumption in the metabolic reactions of the plant. With respect to sole faba bean or faba bean + garlic, broomrape probably played a negative role in the fixation efficiency of carbon dioxide which led to higher amounts of formic acid in the rhizosphere. These results are in parallel with Ritota *et al.* (2012) who reported that formic acid was not detected in the garlic. Conversely, flax increased citric, ascorbic and maleic acids concentrations in faba bean rhizosphere that consumed these acids in their metabolic reactions. Ascorbic acid of flaxseed was increased in the early stages of germination and then remained constant (Herchia *et al.* 2015), meanwhile flaxseed oil has citric and ascorbic acids (Roschel *et al.*, 2019) which explain the presence of citric and ascorbic acids in faba bean rhizosphere. Finally, Thiele *et al.* (2019) investigated structural properties of resins based on epoxidized linseed oil and they found varying amounts of the hardener components methyl-tetrahydro phthalic anhydride, pyromellitic dianhydride and maleic acid which explains the presence of maleic acid in faba bean rhizosphere.

Fenugreek leaves contained oxalate value of 527.5 ± 12.5 mg/100 g dry matter (Radek and Savage, 2008) which explains higher soil oxalic acid concentration in faba bean + fenugreek. Finally, radish + faba bean recorded higher citric and salicylic acids concentration followed by

oxalic acid than the others where radish root had salicylic acid (Stoehr and Herrmann, 1975) and oxalic and maleic acids (Gutiérrez and Perez, 2004). Also, Lyda *et al.* (2019) showed that oxalic acid is the predominant compound in wild radish, followed by quinic, citric and maleic acids. Garlic + faba bean came in the second rank for citric acid concentration and the third rank for maleic acid concentration in faba bean rhizosphere. These results are in the same context with Ritota *et al.* (2012) who indicated that citric and maleic acids were detected in some garlic varieties.

➤ The interaction between faba bean cultivars and intercropping systems

Soil oxalic, ascorbic, formic and salicylic acids concentrations in faba bean rhizosphere were significantly affected by faba bean cultivars x intercropping systems, meanwhile, soil citric and maleic acids concentrations were not affected (Table 1). Fenugreek + Nubaria 2 gave higher soil oxalic, ascorbic and salicylic acids concentrations than the others. Meanwhile, sole Nubaria 1 recorded a higher soil formic acid concentration than the others.

Broomrape infestation and leaf chlorophyll pigments

➤ Faba bean cultivars

Expressing tolerance by the number of spikes and dry weight of spikes per m² showed considerable variation between the two cultivars. Nubaria 2 had a lower number of spikes per m² and spikes dry weight per m² and a higher leaf chlorophylls a and b than the other one (Table 2). Meanwhile,

the level of broomrape infestation was high and uniform in the field with Nubaria 1. Nubaria 2 had an osmotic regulation (cell wall build-up and stamping of the vascular tissues) which negatively affected broomrape growth (Fig 1). Consequently, this will maintain the chlorophyll pigment from deterioration. These results are in accordance with Safina (2017) who found that faba bean cultivar Giza 843 was infested by the lower number of *Orobancha crenata* m² than Giza 2 cultivar.

➤ Intercropping systems

Sole faba bean had a higher number of spikes per m² and spikes dry weight per m² and a lower leaf chlorophylls a and b than the other cropping systems (Table 2). These results could be due sole faba bean had lower soil organic acid concentrations than the others (Table 1), which negatively affected emerged broomrapes and their dry weight per unit area. This biological situation will maintain chlorophyll pigment from deterioration. These results are in parallel to Elsakhawy *et al.* (2020) who showed that broomrape infestation significantly decreased leaf chlorophylls a and b of faba bean plants.

Radish + faba bean decreased number of spikes per m² and spikes dry weight per m² and increased leaf chlorophylls a and b compared with sole faba bean. These results were due to radish increased soil citric, maleic and salicylic acids concentrations in faba bean rhizosphere than the others (Table 1) which reduced broomrape infestation and maintained chlorophyll pigment from deterioration. These results are similar to Karpilov *et al.* (1977) who reported that the activity of decarboxylating NADP-malate dehydrogenase in green etiolated pea and barley leaves and in green leaves of a pea mutant lacking photosystem II is found to be 3-fold increased after the injection of maleic acid into cut plants. Although broomrape absorbed water from the faba bean root, the presence of a high concentration of salicylic acid in faba bean rhizosphere can reduce the effect of water shortage on the different tissues of faba bean (Hussein *et al.*, 2009). The application of citric acid may enhance photosynthesis in plant (Rodriguez *et al.*, 2012). Improvement in biomass and plant growth might be accredited to the ability of citric acid to enhance the uptake of essential nutrients by the formation of complexes with nutrients (Kim *et al.*, 2016). Increasing salicylic acid increased chlorophyll content in faba bean (Al-Hilfy *et al.*, 2017). Salicylic acid induced the catabolism of polyamines and fatty acids in the host root (Madany *et al.*, 2019) which prevented it from being moved to the parasite. With respect to soil citric and maleic acids, broomrape was reduced by maleic acid as reported by Dhanapal *et al.* (1996), meanwhile, the citric acid application significantly improved all the gas exchange characteristics and it has been linked to the suppression of broomrape (Mallhi *et al.*, 2019 and Masteling *et al.*, 2019).

Flax + faba bean decreased number of spikes per m² and spikes dry weight per m² and increased leaf chlorophylls a and b compared with the others. These results were due

to flax increased soil ascorbic acid concentration in faba bean rhizosphere than the others (Table 1) which reduced broomrape infestation and maintained chlorophyll pigment from deterioration. The root exudates of flax involved allelopathic substances that had a negative effect on broomrape (Chittapur *et al.*, 2001). The stress imposed in faba bean by *Orobancha* could generate oxidative stress, a state that could be indicated by the decline of ascorbic acid (Nemat Alla *et al.*, 2007). They added that ascorbic acid had positive influences not only because of overcoming the infection effect but also of improving plant metabolites and antioxidative defense mechanism. Ascorbic acid is found in the cytosol, chloroplasts, vacuoles and mitochondria of plant cells (Farjam *et al.*, 2014). These results are in the same context by Safina (2017) who revealed that flax + faba bean could be formed unfavorable under-grown conditions for *Orobancha crenata* growth. Similar results were observed by Mohamed *et al.* (2020) who showed that foliar application of ascorbic acid caused significant increases in chlorophyll a compared with control faba bean plants.

On the other hand, fenugreek + faba bean had a lower number of spikes per m² and spikes dry weight per m² and a higher leaf chlorophylls a and b than the others under heavily soil infested with broomrape. Fenugreek + faba bean decreased number of spikes per m² and spikes dry weight per m² and increased leaf chlorophylls a and b compared with sole faba bean. These results were due to fenugreek increased soil oxalic acid concentration in faba bean rhizosphere than the others (Table 1) which reduced broomrape infestation and maintained chlorophyll pigment from deterioration. Accumulation of oxalate in plants leads to a change of structure that renders the plant less appetizing and toxic (Franceschi and Loewus, 1995). Oxalates have different roles in plants such as ion balance, plant protection and heavy metal detoxification (Franceschi and Nakata, 2005). Certainly, oxalates prevent nutrients from being lost by binding with iron, zinc and manganese (Radek and Savage, 2008).

➤ The interaction between faba bean cultivars and intercropping systems

Spike dry weight per m² and leaf chlorophyll a were significantly affected by faba bean cultivars x intercropping systems in the both seasons, meanwhile number of spikes per m² and leaf chlorophyll b were significantly affected in the second one (Table 2). Fenugreek, flax, garlic or radish + Nubaria 2 recorded lower number of spikes per m² and spike dry weight per m² and higher leaf chlorophyll b than the others. Nubaria 2 had an osmotic regulation in faba bean root cells (Fig 1) that reduced broomrape growth. This negative effect was enhanced by increasing salicylic acid (Radish or garlic), ascorbic acid (flax) or oxalic acid (fenugreek) concentration in faba bean rhizosphere (Table 1). Ascorbic has a great effect on the biosynthesis of the cell wall (Farjam *et al.*, 2014) which enhanced the tolerance to broomrape infestation. These results are in the same context with Abbes *et al.* (2019) who showed a significant reduction

Table 2: Effect of faba bean cultivars, intercropping systems and their interaction on leaf chlorophylls a and b, as well as number of spikes per m² and spikes dry weight per m² in both seasons.

Faba bean cultivar	Intercropping system	Leaf chlorophyll a (mg/g FW)		Leaf chlorophyll b (mg/g FW)		Spikes/m ² (no.)		Spikes dry weight/m ² (g)	
		1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
		season	season	season	season	season	season	season	season
Nubaria 2	Fenugreek	3.57	3.49	0.61	0.57	12.23	16.63	26.48	73.61
	Flax	3.89	3.82	0.69	0.66	9.42	11.49	19.53	54.29
	Garlic	3.46	3.45	0.58	0.56	16.19	27.03	34.74	113.25
	Radish	3.62	3.55	0.86	0.81	2.85	2.53	7.18	16.51
	Sole faba bean	3.22	3.16	0.49	0.47	33.72	71.82	89.57	306.32
	Mean	3.55	3.49	0.64	0.61	14.88	25.90	35.50	112.79
Nubaria 1	Fenugreek	3.46	3.40	0.55	0.51	19.28	29.30	41.56	128.42
	Flax	3.64	3.57	0.60	0.58	11.45	16.14	26.15	76.61
	Garlic	3.33	3.26	0.52	0.49	22.76	41.65	51.87	177.39
	Radish	3.52	3.49	0.67	0.64	7.61	8.90	17.49	44.07
	Sole faba bean	3.09	3.03	0.39	0.37	41.37	93.08	97.04	353.22
	Mean	3.40	3.35	0.54	0.51	20.49	37.81	46.82	155.94
Average of intercropping system	Fenugreek	3.51	3.44	0.58	0.54	15.75	22.96	34.02	101.01
	Flax	3.76	3.69	0.64	0.62	10.43	13.81	22.84	65.45
	Garlic	3.39	3.35	0.55	0.52	19.47	34.34	43.30	145.32
	Radish	3.57	3.52	0.76	0.72	5.23	5.71	12.33	30.29
	Sole faba bean	3.15	3.09	0.44	0.42	37.54	82.45	93.30	329.77
F test 0.05 Faba bean cultivar		**	N.S.	*	*	*	**	**	**
L.S.D. 0.05 Intercropping system		0.10	0.14	0.07	0.04	2.87	3.11	2.56	3.71
L.S.D. 0.05 Interaction		N.S.	N.S.	N.S.	0.06	N.S.	4.40	3.62	5.24

in *Orobancha foetida* infestation in the susceptible (Badi) and resistant (Najeh) faba bean cultivars when intercropped with fenugreek in field, pot and petri dish experiments.

Faba bean seed yield and its attributes

➤ Faba bean cultivars

The differences in responses of faba bean cultivars to broomrape infestation were observed for percentage of the harvested plants, plant height, seed yield per plant, 100-seed weight and seed yield per ha (Table 3). Nubaria 2 recorded higher percentage of the harvested plants, seed yield per plant, 100-seed weight and seed yield per ha than the other one. Nubaria 2 gave higher percentage of the harvested plants by 21.13 and 17.15% in the first and second seasons, respectively, than the other one. Nubaria 2 had an osmotic regulation in the root cells (Fig 1) which reduced broomrape infestation (Table 2). Nubaria 1 produced taller plants by 10.38 and 10.23% in the first and second seasons, respectively, than other one. This result was due to the genetic makeup of faba bean cultivars. Nubaria 2 had higher seed yield per plant than the other one by 44.04 and 23.69% in the first and second seasons, respectively. Also, Nubaria 2 had heavier 100-seed weight than the other one by 9.60 and 7.85% in the first and second seasons, respectively. Nubaria 2 had an osmotic regulation in the root cells (Fig 1) which decreased infestation with broomrape and maintained leaf chlorophyll pigment from deterioration (Table 2) and

thereby more dry matter accumulation during growth and development. Similar results were observed by Abou-El-Seba *et al.* (2016) who found that faba bean cultivars showed highly significant variation of 100-seed weight and seed yield/plant. Also, Safina (2017) showed that faba bean cultivar Giza 843 had higher seed yield per plant by 8.46% than the other one.

Also, Nubaria 2 had higher seed yield per ha by 12.87 and 14.30 % in the first and second seasons, respectively, than the other one. These results were due to higher percentage of the harvested plants that integrated with seed yield of Nubaria 2 to produce high seed yield per unit area. Similar results were observed by Abou-El-Seba *et al.* (2016) who found that faba bean cultivars showed highly significant variation of 100-seed weight and seed yield/plant. Also, Safina (2017) showed that faba bean cultivar Giza 843 had higher seed yield per plant by 8.46% than the other one.

➤ Intercropping systems

Intercropping systems significantly affected percentage of the harvested plants, plant height, number of pods per plant, seed yield per plant, 100-seed weight and seed yield per ha in both seasons, meanwhile number of branches per plant was not affected (Table 3). Sole faba bean had lower percentage of the harvested plants, number of pods per plant, seed yield per plant, 100-seed weight and seed yield per ha than the other treatments. These results were due to

severe infestation with broomrape that negatively affected leaf chlorophylls a and b (Table 2). It is known that *Orobanche* induced yield reductions are not primarily due to higher competition for water and carbohydrate than faba bean (Bayoumi *et al.*, 2014). These results are in harmony with Elsakhawy *et al.* (2020) who showed that plant height, pod number per plant, seed yield per plant and 100-seed

weight were significantly decreased under broomrape infestation.

Conversely, radish + faba bean recorded higher percentage of the harvested plants, plant height and seed yield per ha compared with the others. Radish + faba bean increased percentage of the harvested plants by 104.38 and 77.05%, plant height by 6.35 and 5.86% and seed yield per

Table 3: Effect of faba bean cultivars, intercropping systems and their interaction on faba bean seed yield and its attributes in both seasons.

Faba bean cultivar	Intercropping system	Harvested plants (%)	Plant height (cm)	Branches /plant (no.)	Pods /plant (no.)	Seed yield/plant (g)	100-seed weight (g)	Seed yield/ha (kg)
1st season								
Nubaria 2	Fenugreek	46.42	104.13	3.53	10.57	29.88	90.26	2246
	Flax	51.61	103.68	3.70	11.71	31.36	92.05	2561
	Garlic	33.26	103.23	3.38	10.50	28.67	88.59	1542
	Radish	62.70	110.19	2.31	10.57	28.99	89.40	2740
	Sole faba bean	31.85	103.46	3.61	7.85	23.72	86.89	1081
	Mean	45.16	104.93	3.30	10.24	28.52	89.43	2034
Nubaria 1	Fenugreek	40.73	114.67	3.51	12.83	20.89	83.57	1963
	Flax	42.16	114.39	3.65	14.66	23.62	84.35	2252
	Garlic	27.42	114.13	3.29	12.19	20.15	80.18	1310
	Radish	51.88	121.56	2.18	12.37	20.32	81.22	2495
	Sole faba bean	24.21	114.44	3.50	9.63	14.06	78.63	990
	Mean	37.28	115.83	3.22	12.33	19.80	81.59	1802
Average of intercropping system	Fenugreek	43.57	109.40	3.52	11.70	25.38	86.91	2104
	Flax	46.88	109.03	3.67	13.18	27.49	88.20	2406
	Garlic	30.34	108.68	3.33	11.34	24.41	84.38	1426
	Radish	57.29	115.87	2.24	11.47	24.65	85.31	2617
	Sole faba bean	28.03	108.95	3.55	8.74	18.89	82.76	1035
F test 0.05 Faba bean cultivar		*	*	N.S.	N.S.	**	*	*
L.S.D. 0.05 Intercropping system		4.11	4.70	N.S.	2.91	3.67	2.89	222.3
L.S.D. 0.05 Interaction		N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
2nd season								
Nubaria 2	Fenugreek	41.32	102.92	3.82	9.25	27.12	82.82	1993
	Flax	45.95	102.55	3.93	10.22	29.66	84.11	2268
	Garlic	32.77	102.37	3.66	9.02	26.12	78.43	1363
	Radish	54.10	107.27	2.46	9.45	26.97	80.81	2626
	Sole faba bean	29.39	101.72	3.88	7.03	18.78	77.02	902
	Mean	40.70	103.36	3.55	8.99	25.73	80.63	1830
Nubaria 1	Fenugreek	34.50	112.81	3.73	11.39	17.83	76.73	1708
	Flax	37.13	112.47	3.81	13.00	20.16	78.28	2022
	Garlic	29.80	112.02	3.60	11.33	17.07	72.98	1169
	Radish	45.44	119.73	2.42	11.37	18.28	74.32	2301
	Sole faba bean	26.83	112.70	3.79	8.51	11.98	71.50	806
	Mean	34.74	113.94	3.47	11.12	17.06	74.76	1601
Average of intercropping system	Fenugreek	37.91	107.86	3.77	10.32	22.47	79.77	1850
	Flax	41.54	107.51	3.87	11.61	24.91	81.19	2145
	Garlic	31.28	107.19	3.63	10.17	21.59	75.70	1266
	Radish	49.77	113.50	2.44	10.41	22.62	77.56	2463
	Sole faba bean	28.11	107.21	3.83	7.77	15.38	74.26	854
F test 0.05 Faba bean cultivar		*	**	N.S.	N.S.	**	*	**
L.S.D. 0.05 Intercropping system		4.37	3.28	N.S.	1.43	3.44	3.58	240.6
L.S.D. 0.05 Interaction		N.S.	N.S.	N.S.	N.S.	N.S.	5.06	N.S.

ha by 152.85 and 188.40% in the first and second seasons, respectively, compared with sole faba bean. Also, radish + faba bean increased percentage of the harvested plants by 31.48 and 31.28%, plant height by 5.91 and 5.22% and seed yield per ha by 24.38 and 33.13% in the first and second seasons, respectively, compared with faba bean + fenugreek. Moreover, radish + faba bean increased percentage of the harvested plants by 22.20 and 19.81%, plant height by 6.27 and 5.57% and seed yield per ha by 8.76 and 14.82% in the first and second seasons, respectively, compared with faba bean + flax. Finally, radish + faba bean increased percentage of the harvested plants by 88.82 and 59.11%, plant height by 6.61 and 5.88% and seed yield per ha by 83.52 and 94.54% in the first and second seasons, respectively, compared with faba bean + garlic. These results were attributed to radish increased salicylic, oxalic and citric acids concentrations in faba bean rhizosphere (Table 1) which reduced number of spikes per m² and spikes dry weight per m² than the others (Table 2) and this biological situation increased percentage of the harvested plants and seed yield per ha. Meanwhile, radish + faba bean increased plant height probably due to improve endogenous plant hormones which positively reflected on internodes length and number. Moreover, soil organic acids analyzes showed that the highest concentration of salicylic acid was found by intercropping radish with faba bean compared with the other treatments (Table 1). It seems that salicylic acid activated the consumption of soluble carbohydrates to form a mechanism against broomrape infestation; particularly Farjam *et al.* (2014) indicated that soluble carbohydrates content in salicylic acid application was significantly lower than ascorbic acid application.

Fenugreek + faba bean had higher percentage of the harvested plants, number of pods per plant, seed yield per plant, 100-seed weight and seed yield per ha than faba bean + garlic and sole faba bean. These results were due to higher soil oxalic acid concentrations in the faba bean rhizosphere that negatively affected number of spikes per m² and spikes dry weight per m² (Table 2). The nutrient concentration directly affects host resistance potential towards broomrape (Labrousse *et al.*, 2010). These results reveal that nutrients cannot be absorbed by broomrape as they are bound with oxalic acid that secreted by fenugreek or even from radish which positively reflected on chlorophyll pigments. Meanwhile, flax + faba bean had the highest number of pods per plant, seed yield per plant and 100-seed weight compared with the others. Flax + faba bean increased number of pods per plant by 50.80 and 49.42%, seed yield per plant by 45.52 and 61.96% and 100-seed weight by 6.57 and 9.33% in the first and second seasons, respectively, compared with sole faba bean. Also, flax + faba bean increased number of pods per plant by 12.64 and 12.50%, seed yield per plant by 8.31 and 10.85% and 100-seed weight by 1.48 and 1.78% in the first and second seasons, respectively, compared with faba bean + fenugreek. Moreover, flax + faba bean increased number of pods per

plant by 16.22 and 14.15%, seed yield per plant by 12.61 and 15.37% and 100-seed weight by 4.52 and 7.25% in the first and second seasons, respectively, compared with faba bean + garlic. Finally, flax + faba bean increased number of pods per plant by 14.90 and 11.52%, seed yield per plant by 11.52 and 10.12% and 100-seed weight by 3.38 and 4.68% in the first and second seasons, respectively, compared with faba bean + radish. These results were attributed to high soil ascorbic and citric acids concentrations in faba bean rhizosphere (Table 1) that reduced number of spikes per m² and spikes dry weight per m² (Table 2) which positively reflected on leaf chlorophyll a in leaves of faba bean plants. Ascorbic acid application decreased rate of damage in cell membrane than salicylic acid application (Farjam *et al.* 2014). Furthermore, garlic + faba bean had higher number of pods per plant, seed yield per plant, 100-seed weight and seed yield per ha than sole faba bean. These results were due to garlic increased soil organic acids concentrations in faba bean rhizosphere than sole faba bean (Table 1) which negatively affected broomrape infestation (Table 3).

➤ The interaction between faba bean cultivars and intercropping systems

The interaction between faba bean cultivars and intercropping systems significantly affected 100-seed weight in the second season (Table 3). Flax + Nubaria 2 had heavier 100-seed weight than the others. These results were due to Nubaria 2 had an osmotic regulation in faba bean roots cells (Fig 1) which reduced infestation with broomrape (Table 2). This negative effect was enhanced by increasing ascorbic acid concentration in the faba bean rhizosphere (Table 1). This biological situation allowed soil water and nutrients to move to different parts of the plant which maintained photosynthetic integrity of faba bean plant (Table 3) and thereby increased seed filling rate during growth and development.

Competitive relationships and intercropping economic advantage

➤ Land equivalent ratio (LER)

The total LER values were better than one in all the studied treatments. Intercropping fenugreek, flax, garlic or radish with two faba bean cultivars increased LER compared with sole plantings of faba bean in both seasons (Table 4). LER ranged from 1.58 by garlic + Nubaria 1 to 3.09 by radish + Nubaria 2 in the first season. Also, LER ranged from 1.73 by garlic + Nubaria 1 to 3.48 by radish + Nubaria 2 in the second season. The advantage of higher LER by intercropping fenugreek, flax, garlic or radish with two faba bean cultivars over sole plantings of faba bean could be due to fenugreek, flax, garlic or radish had positive effect on faba bean productivity under heavily soil infested with broomrape that reduced intra – specific competition between faba bean plants for basic growth resources. Radish + Nubaria 2 recorded higher values of LER (3.09 and 3.48 in

Table 4: Relative yields of all species, LER and economic return of intercropping four crops with two faba bean cultivars in both seasons.

Faba bean cultivar	Intercropping system	Faba bean seedyield/ha	Intercrop		Relative yield		LER	Total return/ha	MAI
			yield/ha (kg)		RY _{faba bean}	RY _{intercrop}			
			(kg)	Inter.					
1 st season									
Nubaria 2	Fenugreek	2246	304	1011	2.07	0.30	2.37	1872.4	1082.4
	Flax	2561	332	1419	2.37	0.23	2.60	2047.7	1260.1
	Garlic	1542	7364	23014	1.42	0.32	1.74	2479.9	1054.6
	Radish	2740	1255	2212	2.53	0.56	3.09	3541.5	2395.4
	Sole	1081	-	-	1.00	1.00	1.00	778.3	-
Nubaria 1	Fenugreek	1963	276	1011	1.98	0.27	2.25	1645.2	914.0
	Flax	2252	328	1419	2.27	0.23	2.50	1822.8	1093.6
	Garlic	1310	6123	23014	1.32	0.26	1.58	2082.0	764.3
	Radish	2495	1247	2212	2.52	0.56	3.08	3355.1	2265.8
	Sole	990	-	-	1.00	1.00	1.00	712.8	-
2 nd season									
Nubaria 2	Fenugreek	1993	282	959	2.21	0.29	2.50	1671.8	1003.1
	Flax	2268	309	1336	2.51	0.23	2.74	1822.6	1157.4
	Garlic	1363	6974	21134	1.51	0.33	1.84	2278.5	1040.1
	Radish	2626	1180	2067	2.91	0.57	3.48	3365.7	2398.5
	Sole	902	-	-	1.00	1.00	1.00	649.4	-
Nubaria 1	Fenugreek	1708	265	959	2.11	0.27	2.39	1452.3	844.6
	Flax	2022	302	1336	2.50	0.22	2.73	1641.2	1040.0
	Garlic	1169	5917	21134	1.45	0.28	1.73	1942.2	819.5
	Radish	2301	1172	2067	2.85	0.56	3.42	3121.7	2208.9
	Sole	806	-	-	1.00	1.00	1.00	580.3	-

the first and second seasons, respectively) followed by flax + Nubaria 2 (2.60 and 2.74 in the first and second seasons, respectively). Fenugreek + Nubaria 2 came in the third rank. The advantage of the highest LER by radish or flax + Nubaria 2 over the others was due to Nubaria 2 had an osmotic regulation in the root cells (Fig 1) which negatively affected broomrape infestation. This negative effect was enhanced by increasing salicylic acid (radish) or ascorbic acid (flax) concentration in faba bean rhizosphere. This biological situation maintained leaf chlorophyll pigments of Nubaria 2 from deterioration (Table 2) which increased faba bean productivity under infestation with broomrape. These results are in accordance with Safina (2017) who found that flax + Giza 2 cultivar gave higher LER than flax + Giza 843 cultivar.

➤ Intercropping economic advantage

Data presented in (Table 4) indicate that intercropping fenugreek, flax, garlic or radish with two faba bean cultivars was more profitable to farmers than sole plantings of faba bean in both seasons. Total return varied from 1645.20 USD/ha by fenugreek + Nubaria 2 to 3541.55 USD/ha by radish + Nubaria 2 in the first season. Also, total return varied from 1452.36 USD/ha by fenugreek + Nubaria 1 to 3365.72 USD/ha by radish + Nubaria 2 in the second one. With respect to MAI, MAI varied from 764.30 by garlic + Nubaria 1 to 2395.41 by radish + Nubaria 2 in the first season. Also, MAI varied from 819.55 by garlic + Nubaria

1 to 2398.55 by radish + Nubaria 2 in the second one. Radish + Nubaria 2 recorded the highest economic return followed by flax + Nubaria 2.

CONCLUSION

Growing of one row of radish or flax between two rows of faba bean cultivar Nubaria 2 in ridges 60 cm width increased faba bean productivity, land usage and economic return, hence it should be recommended under heavily soil infested with broomrape.

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