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Performance of Sesbania–Sunflower under Different Cropping Systems using ^{15}N and ^{13}C

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A field experiment on sesbania (ses) and sunflower (sun) plants grown in monocropping and intercropping systems (1ses:1sun; 1ses:2sun, and 2ses:1sun, row ratio) was conducted to evaluate growth and N_2-fixation using ^{13}C and ^{15}N natural abundance techniques. The results showed that 1ses:1sun surpassed the other treatments in terms of nitrogen (N) and dry matter yields (DM). However, 1ses:2sun gave the highest seed and oil production and showed the greatest N_2-fixation. Sesbania plants in the 1ses:2sun fixed almost identical amounts of N_2 in mono and intercropping systems. Moreover, soil N-uptake was the lowest among other treatments. These results give an advantage to the 1ses:2sun treatment over other treatments in terms of soil N consumption and N_2 fixation to meet sesbania's N requirements. Nevertheless, 2ses:1sun seemed to be an inappropriate treatment. On the other hand, it could be suggested from ‰$\Delta^{13}C$ data that plants in the intercropping systems appeared not to be subjected to any abiotic stress during the growing period.

Key words: Sesbania, Sunflower, intercropping, N2- fixation, 13C, 15N

Multiple cropping system of legumes and non legumes is a traditional farming practice in many countries and represents a promising agricultural practice enabling better use of lands for sustainable agriculture (Muturi *et al.*, 2016). Benefits of this system may arise from increased nitrogen (N), dry matter yield and greater N_2 fixation efficiency (Fujita *et al.*, 1990; Tobita *et al.* 1994; Kurdali *et al.*, 1996; Kurdali 2009). However, N_2 fixed by the legume component in the intercropping system depends on the species, morphology, density of legume, type of management, and the competitive abilities of the component crops (Ofori and Stern, 1987).

Sunflower was evaluated as a component of strip, row, and relay intercropping systems that would be practical for mechanized farming (Robinson, 1984). Several researchers have reported on sunflower intercropping with legumes. Robinson (1984) reported that yield was greater for some intercropping systems than from sole cropping. Fieldbean (*Phaseolus vulgaris* L.) as a row intercrop with sunflower increased in yield as sunflower population decreased, but its highest intercrop yield was 59% that of sole cropped fieldbean. Strip intercropping sunflower with corn or soybean compared with sole cropping showed that the percentage gain in sunflower yield was offset by about the same percentage reduction in corn or soybean yield. Shivaramu and Shivashankar (1992) reported that legumes reduced sunflower seed yield, achen per head and head diameter when they were sown at the same time. However, Kandhro *et al.*, (2007) showed that mungbean/ sunflower intercropping can be practiced to maximize the crop production from the unit area.

Dhaincha (*Sesbania aculeata* L. Pers.) is a fast growing leguminous plant, and is adapted to a variety of soil conditions, varying from waterlogged to saline, and from sandy to clay soils (Sandhu and Haq, 1981; Kurdali and Al-Ain, 2002; Kurdali *et al.*, 2003). It is a native to Pakistan and India and was introduced to Syria to produce green manure (Kurdali *et al.*, 2007) and fodder (Zarkawi, Al-Masri and Khalifa 2003) under saline and non saline conditions. Moreover, the inclusion of sesbania in multiple cropping systems with non legumes represents promising agricultural practices enabling better use of lands for sustainable agriculture (Kurdali *et al.*, 2003). However, Kurdali, (2009) showed that growth, N_2-fixation and soil N uptake in this legume species was affected by the companion crop (*e.g.,* sorghum as C_4 or sunflower as C_3). Intercropping of two rows of sesbania with one row of sorghum (2sesbania/ 1sorghum) showed a greater efficiency over monocropping; whereas, the 2sesbania/1sunflower intercropping was similar to that of the monocropping (Kurdali 2009). Hence, the assessment of different planting patterns of sesbania and sunflower is required. The objectives of this field experiment were to: 1) evaluate dry matter production, total N yield and land-use efficiency in *Sesbania aculeata* (ses.) and *Helianthus annuus* L. (sun.) grown either solely or as intercrops using three different combinations of sesbania and sunflower row ratio (2ses: 1sun; 1ses: 1sun and 1ses: 2sun), 2) measure N_2-fixation in sesbania using the ^{15}N natural abundance technique, 3) assess the intraspecific competition for soil N uptake and balance under intercropping system, 4) examine the possibility of N transfer from sesbania to the associated non-legumes, and 5) determine if the carbon isotope discrimination ($\Delta^{13}C$) can be used to assess factors responsible for crop performance variability in different cropping systems.

MATERIALS AND METHODS

Site description

The field experiment was conducted at Der-Alhajar Research Station located south east of Damascus, Syria (33°21'N, 36°28' E) at 617 m. above sea level. Soil texture was sandy clay (50.4% sand, 13.1% silt and 36.5% clay) with an average of pH 8.6, Ec_e 0.16 dS/m, organic matter 0.82%, available P 10.8 µg /g, NH_4^+ 8.3 µg /g, NO_3^- 4.6 µg /g and total N 0.7 mg/g of the top 25 cm. The average minimum temperature in winter is 1.3 °C in Jan., while it increases to an average of 36 °C during August.

Field procedures and treatments

Seeds of dhaincha (*Sesbania aculata*) and sunflower (*Helianthus annuus*) were hand sown at 3 cm depth in the first week of July, in rows with row- to- row distance of 40 cm and plant to plant distance of 40 cm for sesbania, and 20 cm for sunflower. Two crops and three intercropping systems were studied: 1) sole crop of sesbania, 2) sole crop of sunflower, 3) sesbania and sunflower grown in alternating rows of each species (1ses:1sun), 4) species were grown in alternating two rows of sesbania with one row of sunflower (2ses:1sun), and 5) sesbania and sunflower were grown in alternating one row for the former with two rows for the latter (1ses:2sun). Plant densities were 60,000 and 120,000 plant/ha for solely grown sunflower and sesbania, respectively. In the intercropping systems, plant density in the 1ses:1sun treatment was 1/2 that of the solely grown species. Whereas, sesbania and sunflower in the 1ses:2sun were 1/3 and 2/3 that of the sole cropping plants, respectively. In contrast, they were 2/3 and 1/3 that of the sole cropping in the 2ses:1sun treatment. Barley was the previous crop. Each plot, representing one replicate, was 5×5m. Since abundant nodules have been previously observed on the roots of *Sesbania aculeata* grown at the same site, the seeds were not inoculated. Surface irrigation was initiated immediately after planting and preceded according to soil moisture content which was determined by neutron scattering (CPN 503 DR) in access tubes installed to 105 cm depth in a central row of each treatment (Kurdali, 2009).

Intercropping system effectiveness assessment

Land equivalent ratio (LER) was used for estimating the efficiency of intercropping relative to sole cropping (Mead and Willy, 1980), and was calculated using the following equation (Offori, and Stern, 1987):

$$LER = (Yij/Yii)+(Yji/Yjj)$$

Where, Y is the yield per unit area (kg/ha), Yii and Yjj are sole crop yield for the component crops i (sunflower) and j (sesbania). Yij and Yji are mixed-crop yield.

Plant sampling and isotopic compositions analyses

Plant samples were harvested 85 days after planting (DAP) coinciding with the physiological maturity stage. Whole aboveground plant samples were collected from the corresponding subplots by cutting the main stem immediately below the cotyledonary node, then separated into its main components (leaves, stem and reproductive parts), dried at 70 °C, weighed and ground to a fine powder. Plants from the mixed stands were separated into the component species. Furthermore, grain yield of the different plant species was determined at maturity (115 DAP). In addition, oil content was determined in sunflower seeds originated from pure and mixed stands as extractable component in Soxhelt apparatus using standard method (A.O.A.C, 1990). Concentration of total N and C, and $\sigma^{15}N$ and $\sigma^{13}C$ were determined on sub-samples (7 and 2 mg dry weight for N and C determination, respectively) of leaves, stem and reproductive parts using the continuous-flow isotope ratio mass spectrometry (Integra-CN, PDZ Europea Scientific Instrument, UK). Isotopic compositions are expressed using delta notation (σ) in parts per thousand (‰):

$$\sigma (‰) = [(R_{sample}/R_{standard}) - 1]\ 1000$$

where R is the ratio of $^{15}N/^{14}N$ or $^{13}C/^{12}C$. The nitrogen isotope ratios are expressed relative to atmospheric air as the standard. The carbon isotope ratio in the plant sample is expressed relative to Pee Dee belemnite (PDB) standard.

Although $\sigma^{13}C$ provides information on the $^{13}C/^{12}C$ in tissues, it is often preferable to express the values as leaf carbon isotope discrimination ($\Delta^{13}C$) (Farquhar, O'Leary and Beny 1982):

$$\Delta^{13}C = (\sigma\ ^{13}C_{air} - \sigma\ ^{13}C_{sample}) / (1 - \sigma\ ^{13}C_{sample}/1000)$$

where $\sigma\ ^{13}C_{air}$ is the $\sigma\ ^{13}C$ value in air (-8‰) and $\sigma\ ^{13}C_{sample}$ is the measured value in the plant.

Estimate of atmospheric N_2 fixation by sesbania

The fractional contribution of fixed N_2 derived from air (%Ndfa) in sesbania grown as a single crop and in intercropped stands was calculated from the ^{15}N abundance of the legume and non N_2-fixing reference plants (‰) as indicated in the following equation (Shearer and Kohl, 1986; Amarger *et al.*, 1979; Domenach and Chalamet 1979):

$$\%Ndfa = (\sigma^{15}N_{ref} - \sigma^{15}N_{fp}) / (\sigma^{15}N_{ref} - B)\ 100$$

where $\sigma^{15}N_{ref}$ represents the level of $\sigma^{15}N$ detected in the reference plants (*e.g.,* solely grown sunflower) growing on the same soil at the same time as the test legume, $\sigma^{15}N_{fp}$ is the $\sigma^{15}N$ of the test fixing plant (*e.g.*, sesbania) and *B* is the value of $\sigma^{15}N$ (-1.37‰) in sesbania plants that was solely dependent on atmospheric N_2 (*e.g.,* nodulated plants grown on N-free medium), (Kurdali, 2009). The sole crops of sunflower served as a reference crop for measuring N_2-fixation by sesbania plants.

Whole plant $\sigma^{15}N$(‰) was calculated as an average of shoots (Sh), stems (St) and reproductive parts (Rp) $\sigma^{15}N$ weighted by the total N content (kg) of shoots, stems and reproductive parts:

$\sigma^{15}N$= [(Sh $\sigma^{15}N$x Sh N) + (St$\sigma^{15}N$ x St N) + (Rp $\sigma^{15}N$ x Rp N)]/ (Sh N + St N + Rp N)

Whole plant $\sigma^{13}C$ (‰) was calculated as follow:

$\sigma^{13}C$ = [(Sh $\sigma^{13}C$ x Sh C) + (St $\sigma^{13}C$ x St C) + (Rp $\sigma^{13}C$ x Rp C)]/ (Sh C + St C + Rp C)

Statistical analysis

The experimental design was a randomized complete block with four replicates. Data was subjected to analysis of variance (ANOVA) test, and means were compared using the Least Significant Difference (Fisher's PLSD) test at the 0.05 confidence level.

RESULTS AND DISCUSSION

Dry matter yield

It has been reported that production efficiency in intercrop systems could be improved by minimizing inter specific competition between the component crops for growth limiting factors (Offori and Stern, 1987). Therefore, a balance between non legumes and legumes in the mixed stand is desirable. Competition between component crops for growth limiting factors may be regulated by agronomic factors, such as the proportion of crops in the mixture (Offori and Stern, 1987; Kurdali *et al.*, 2003; Yilmaz *et al.*, 2008; Kurdali (2009) reported that when component crops in mixed cropping systems belong to different photosynthetic carbon dioxide metabolism pathway (*e.g*., sesbania, C3 and sorghum, C4), productivity and efficiency appeared to be determined by the more aggressive crop, usually the C4. However, the undertaken study showed that when component crops belong to the same photosynthetic pathway (C3), productivity appeared to be determined by their corresponding densities in the intercropping system. The 1ses:1sun treatment exhibited a similar distribution of total DM in the sesbania (4.7Mg/ha) and sunflower (4.9 Mg/ha), (Table 1). This result indicated that a balance in the dry matter yield between the legume and non legumes species (C3) was obtained when they were grown in alternating rows. However, the contribution of sunflower to dry matter production (5.7 Mg/ha) was more than that of sesbania (3.7 Mg/ha) in the 1ses: 2sun treatment. In contrast, the contribution of sesbania to dry matter production (5.3 Mg/ha) in the 2ses: 1sun treatment was higher than that of sunflower (3.3 Mg/ha), (Table. 1). Similarly, the latter values were relatively close to those obtained in a previous study where DM yield of sesbania (5.9 Mg/ha) was almost two-fold that of sunflower (3.1 Mg/ha) using 2ses:1sun row ratio (Kurdali, 2009). Also, Shivaramu and Shivashankar (1992) showed that the soybean yield increased along with its increased density in mixed cropping system with sunflower. Therefore, it can be concluded that dry matter production in a given species is determined by their corresponding densities in the intercropping system and/or by the more competitive crop.

The total above ground dry matter accumulation in the solely grown sunflower was higher than that of solely grown sesbania as well as of the intercropping treatments. Both 1ses:1sun and 1ses:2sun treatments significantly ($P<0.05$) surpassed 2ses:1sun and solely grown sesbania in terms of total dry matter yield. Dry matter yield of each component crop within the intercropping treatment was significantly lower ($P<0.05$) than that of the sole cropping (Table 1). This indicates that the component crops compete with each other for the limited resources under the intercropped conditions. The reduced dry matter of the intercropped plants compared to solely grown plants in our study is a common observation in multiple cropping systems (Danso, Zapata, and Hardarson 1987; Tobita *et al.* 1994; Kurdali *et al.*, 2003; Kurdali, 2009).

Grain and oil yield

Grain yield data followed a similar trend to that of the dry matter yield (Table 2). Grain yield of each plant species grown in the mixed stands was lower than solely grown crops (Table 2). Moreover, oil content and yield in sunflower grown in the mixed stands were also lower than solely grown crops. Similarly, Kandel *et al.*, (1997) showed that intercropping of hairy vetch, sweet clover alfalfa and medic with sunflower, at the same time, reduced sunflower achene's yield and head diameter.

Nevertheless, the 1ses:2sun treatment gave the greatest seed and oil production of sunflower plants comparing to the other two intercropping treatments.

Intercropping system effectiveness assessment

According to Offori and Stern (1987), the land equivalent ratio (LER) directly reflects the land performance under mixed cropping as a function of plant density, competitive ability of the component crops, land management and surrounding environment and water availability (Kurdali *et al.*, 2003; Kurdali 2009). In this study, LERs (total) in the three intercropping systems were close to one indicating that the efficiency of producing dry matter in the sesbania /sunflower intercropping was similar to that in the monocropping system (Table 3). In the 1Ses:2Sun treatment, the LERi value of sunflower (0.51) was almost equal to that of sesbania (LERj 0.47). This indicates that both plant species performed well in this mixed stand from the land equivalent ratio point of view. The 1ses:1sun was next in order where LER values were 0.44 and 0.59 in sunflower and sesbania, respectively. Nevertheless, 2ses:1sun treatment seemed not to be an appropriate treatment due to the divergence of LER values in both species, where sunflower plants had a much smaller value (0.30) comparing to that of sesbania (0.66). Similarly, these values were relatively close to those obtained in a previous study (Kurdali, 2009) confirming that 2ses:1sun row ratio is not an appropriate intercropping treatment from the land equivalent ratio point of view.

Table 3 shows that the greatest LER total value of seed yield was in 1Ses:2Sun treatment (1.07). Moreover, LER values of sunflower seeds (0.69) and oil (0.62) yields in this intercropping treatment were greater than 0.5. These results indicate that 1ses:2sun treatment was more appropriate than the others because of its higher LER value for seed and oil yield of sunflower plants.

Total Nitrogen Yield

Nitrogen yield of different plant parts of sesbania, and sunflower, grown either alone or in intercropping systems is given in Table 4. Whole plant N yield of the solely grown plants did not differ from each other ($P<0.05$). Nitrogen yield of each component crop within the intercropping treatment was significantly lower than that of the sole cropping. With regards to the intercropping treatments, 1ses:1sun gave the greatest N yield and together with 1ses:2sun treatment were satisfactory in terms of N uptake in both species having almost similar values. The observed values of N yield were 110 and 100 kg N/ha in the 1ses:1sun and 87.7 and 88 kg N/ha in the 1ses:2sun treatment for sesbania and sunflower plant species, respectively (Table 4). Nevertheless, 2ses:1sun treatment seemed not to be an appropriate treatment due to the divergence of N yield in sesbania (119 kg N/ha) and sunflower (56 kg N/ha) species.

Atmospheric N_2 fixation

$\sigma^{15}N$ (‰) values in shoots and reproductive plant parts of sesbania, and sunflower, grown either alone or in intercropping systems are given in Table 5. Whole plant $\sigma^{15}N$ (‰) data is summarized in Fig.1. The $\sigma^{15}N$ value of fixed nitrogen determined in sesbania plants grown on a N-free medium was -1.37±0.5. This value was significantly different from those of sunflower plants (+14‰ on the average); whereas, $\sigma^{15}N$ in sesbania plants ranged between -0.32 and +7.19‰. The lower $\sigma^{15}N$ values in sesbania plants compared to sunflowers indicated a significant contribution of N_2 fixing in this plant species. Moreover, the lower $\sigma^{15}N$ values (-0.32‰) in sesbania plants grown in the intercropping system (*e.g.*, 1ses:2sun) compared to other cropping reflected a higher %Ndfa. Similar observations were reported in previous studies, where the natural abundance of the ^{15}N in the intercropped legumes compared with non-legumes was found to be considerably less than that of sole cropping (Berkasm *et al.*, 1988, Kurdali, 2009).

Data of the proportion and amounts of N derived from N_2-fixation (Ndfa) in the different plant parts of sesbania are given in Table 6. The percentage contribution of biological nitrogen fixation (BNF) to the amount of N accumulated by sole sesbania (45%) did not significantly differ from intercropped sesbania plants in 1Ses:1Sun (43%) and 2Ses:1Sun (44%) treatments. However, a significant increase in %Ndfa rates (93%) was obtained when growing one row of sesbania with two rows of sunflower (1ses:2sun). The enhanced % N_2

fixation in the 1ses:2sun stand compared with the other stands might be attributed to the depletion of soil N resulting from the greater apparent competitiveness of sunflower for soil N, and consequently, a greater sesbania dependence on N_2 fixation as previously reported for other mixed cropping systems (Izaurralde *et al.*, 1992; Kurdali *et al.*, 1990; Kurdali *et al.*, 1996; Hardarson *et al.*, 1988).

The amount of N symbiotically fixed by solely grown sesbania (83 kg N/ha) was not significantly different from that of 1ses:2sun (81 kg N/ha) in spite of higher %Ndfa values in the intercrop (Table 6). This could be attributed to the decrease in total dry matter yield when intercropped (Table 1). This observation is consistent with studies on cereal/legume intercropping systems (Danso *et al.*, 1987; Tubita *et al.*, 1994). Although there were no significant differences in the amount of Ndfa by sesbania plants among monocropping and intercropping (1ses:2sun) treatments, the number of rows in the mixed stand was only 1/3 of that in the pure stand. This gives an advantage to this intercropping system over sole cropping with regards to N_2-fixation. Nevertheless, 1ses:1sun and 2ses:1sun treatments seemed to be unappropriate in terms of N_2-fixation due to the lower amounts of fixed N (48 and 53 kg N/ha, respectively). On the other hand, the possibility of N-transferred from sesbania to the adjacent sunflower is excluded, due to the insignificant differences between $\sigma^{15}N$ values in the whole plant of sunflower plants grown in the different cropping systems (Fig. 1).

Soil nitrogen uptake

The percentage contribution of soil N (Ndfs) to the amount of N accumulated by sole sesbania (55%) did not significantly differ from sesbania plants in 1ses:1sun (57%) and 2ses:1sun (56%) treatments. However, a significant decrease in %Ndfs rate (7%) was obtained in sesbania when growing one row of sesbania with two rows of sunflower (1ses:2sun). Amount of N derived from soil in solely grown sesbania was 100 kg N/ha, whereas, it decreased to 62 and 66 kg N/ha in 1ses:1sun and 2ses:1sun treatments, respectively. However, soil N uptake by sesbania in 1ses:2sun treatment was only 7 kg N/ha (Table 6).

Soil N uptake in the 1ses:2sun treatment (95 kg N/ha) was less than that of the other treatments (162, 122, 176 and 100 kg N/ha in 1ses:1sun, 2ses:1sun, sole sunflower, and sole sesbania, respectively) (Tables 6 & 4). Correspondingly, soil N uptake by sunflower (88 kg N/ha) in the latter intercropping system was 12.5 times greater than that of sesbania (7 kg N/ha) indicating a high competitiveness for soil N among the component crops when grown together. Such a decrease in soil N uptake by sesbania plants was associated with a higher amount of N_2-fixation (81 kg/N ha). These results give an advantage to the 1ses:2sun treatment over other treatments in terms of soil N consumption and N_2 fixation to meet sesbania's N requirements. Consequently, total N uptake was almost the same between the two plant species (88 kg N/ha).

Competitiveness for soil N among the component crops in the 1Ses:1Sun treatment seemed to be less than that of 1ses:2sun because soil N uptake by sunflower (100 kg N/ha) was only 1.6 times greater than that of sesbania (62 kg N/ha). However, amounts of Ndfs were almost the same between the two species (56 and 66 kg N/ha for sunflower and sesbania, respectively) in the 2ses:1sun treatment (Tables 4&6). Similarly, Kurdali *et al.*, (2003) reported a relatively similar amount of soil N uptake when growing two rows of sesbania with one row of sorghum plants. Overall, from ecological standpoint, the best intercropping treatment seemed to be 1ses:2sun which showed greatest N_2-fixation and lowest soil N consumption.

Soil nitrogen balance

A positive soil N balance would be normally expressed when the total amount of N_2 fixed in plant residues exceeded the total amount of N removed. Therefore, it is important to adopt suitable intercropping system to obtain a high potential yield of residual N, which gives an advantage to the subsequent crops. The total amount of fixed N_2 in shoots and stems of sesbania plants were: 24, 12, 17 and 14 kg N/ha in sole sesbania, 1ses:1sun, 1ses:2sun and 2ses:1sun, respectively (Tables 6 & 7). While, for the same treatments, amounts of nitrogen absorbed from soil and stored in sesbania's pods were: 77, 50, 3, 52, kg N/ha (Table 7). Consequently, estimated N balance values of sesbania plants following pod harvest were -53, -38, +14.3 and -

38 kg N/ha for the above mentioned treatments, respectively.

Regarding sunflower plants, amounts of nitrogen absorbed from soil and stored in the reproductive parts were 105, 66, 57 and 38 kg N/ha in sole sunflower, 1ses:1sun, 1ses:2sun and 2ses:1sun, respectively (Tables 4 & 6). Consequently, a negative N balance resulted in all treatments. The net soil N balance in different cropping system were -43, -53, -76, -104 and -105 kg N/ha for 1ses:2sun, sole sesbania, 2ses:1sun, 1ses:1sun and sole sunflower, respectively. However, the best treatment was 1ses:2sun because of its highest N balance and N_2-fixation.

Table 1. Dry matter yield (Mg/ha) in leaves, stems, reproductive parts and whole plant of sesbania (Ses) and sunflower (Sun) grown either solely or as intercropping systems

Cropping system	leaves	stem	Rep. parts	Whole plant
(A) Comparisons among cropping systems for each plant species				
Sesbania				
Sole	0.91a	2.81a	4.3a	8.00a
(1ses:1sun)	0.53c	1.66b	2.51b	4.69b
(1ses:2sun)	0.46c	1.33c	1.95c	3.73c
(2ses:1sun)	0.68b	1.82b	2.75b	5.25b
LSD 0.05	*0.13*	*0.29*	*0.46*	*0.76*
Sunflower				
Sole	2.48a	4.29a	4.34a	11.12a
(1ses:1sun)	1.20b	1.82c	1.91b	4.93c
(1ses:2sun)	1.23b	2.45b	2.05b	5.71b
(2ses:1sun)	0.72c	1.29d	1.30c	3.30d
LSD 0.05	*0.23*	*0.29*	*0.36*	*0.50*
(B) Comparisons among cropping systems				
Sole crop				
Sesbania	0.91d	2.81c	4.28a	8.00c
Sunflower	2.48a	4.29a	4.34a	11.12a
intercropped				
(1ses:1sun)	1.73b	3.47b	4.42a	9.62b
(1ses:2sun)	1.67b	3.78b	4.00a	9.44b
(2ses:1sun)	1.39c	3.11c	4.05a	8.56c
LSD 0.05	*0.23*	*0.35*	*N.S*	*0.77*

Note. For each crop species (A), and the cropping systems (B), means within a column followed by the same letter are not significantly different (*P>0.05*).

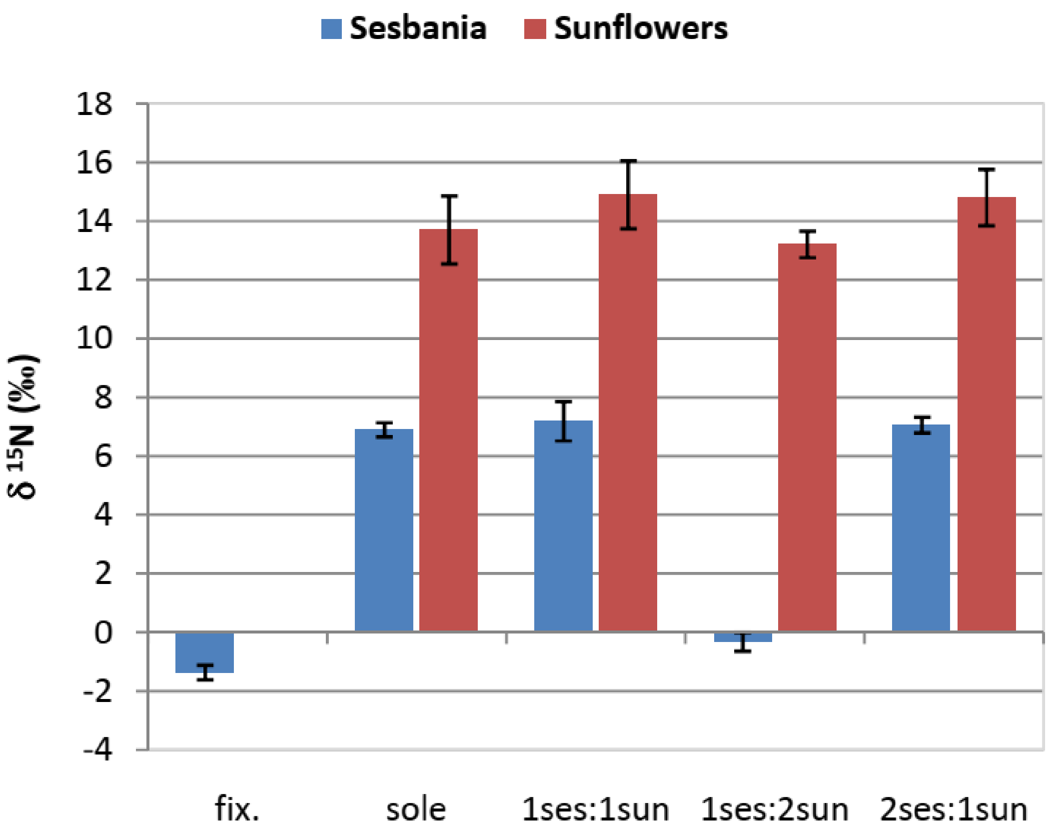

Figure 1. $\delta^{15}N$ (‰) values of the entire plants of sesbania (Ses) and sunflower (Sun) grown either solely or as intercropping systems. $\delta^{15}N$ (‰) of N_2-fixed (fix) was obtained from sesbania plants grown on N-free medium. Bars mean standard deviations.

Table 2. Seed yield (Mg/ha) of sesbania (Ses), and sunflower (Sun) grown either solely or as an intercropping system, in addition to oil content and yield in sunflower seeds under different cropping systems

Cropping system	Sesbania	Sunflower		
	Seed yield (Mg/ha)	Seed yield (Mg/ha)	Oil content (mg/g)	Oil yield (Mg/ha
Sole	1.58a	3.03a	478a	1.45a
(1ses:1sun)	0.93b	1.38c	437b	0.60c
(1ses:2sun)	0.59c	2.09b	427b	0.89b
(2ses:1sun)	1.02b	1.06d	431b	0.46d
LSD 0.05	*0.17*	*0.26*	*22.04*	*0.13*

Note. Means within a column followed by the same letter are not significantly different (*P>0.05*).

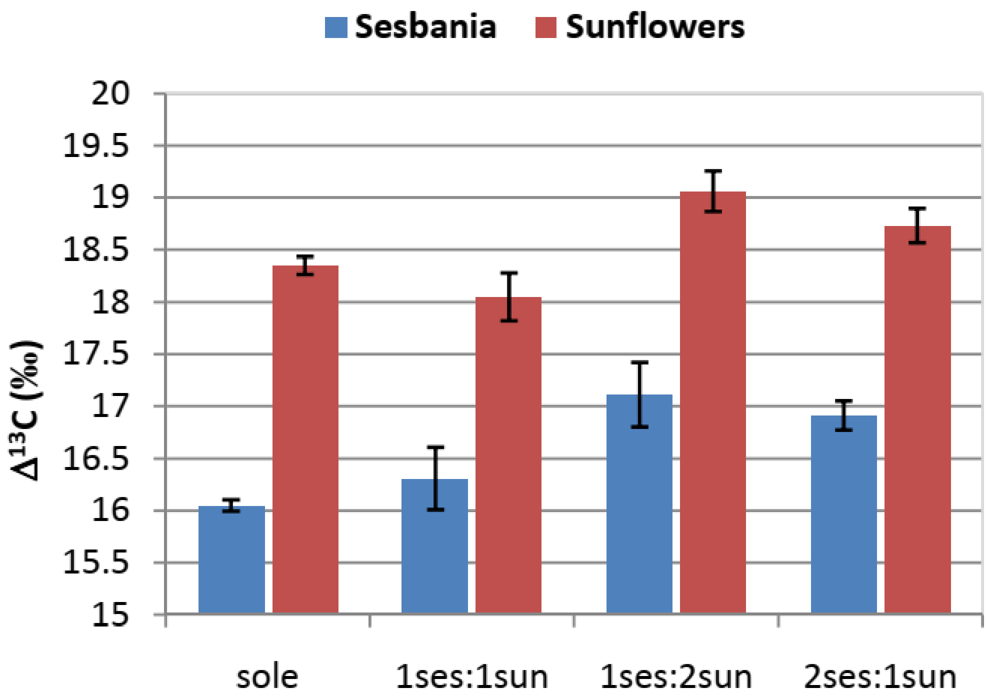

Figure 2. Carbon isotope discrimination ($\Delta^{13}C$‰) in the whole plants of sesbania (Ses) and sunflower (Sun) grown either solely or as intercropping systems. Bars mean standard deviations.

Table 3. Land Equivalent Ratio (LER) of the dry matter, seed and oil yields for sunflower (LERi) and Sesbania (LERj) either solely or as intercropping systems

LER	Intercropping		
	1ses:1sun	1ses:2sun	2ses:1sun
Total dry matter yield			
LERi (sunflower)	0.44	0.51	0.30
LERj (sesbania)	0.59	0.47	0.66
LER (total)	**1.03**	**0.98**	**0.96**
Seed yield			
LERi (sunflower)	0.46	0.69	0.35
LERj (sesbania)	0.59	0.38	0.65
LER (total)	**1.05**	**1.07**	**1.00**
Oil yield			
LERi (sunflower)	**0.42**	**0.62**	**0.31**

Table 4. Nitrogen yield (kg N /ha) in leaves, stem, reproductive parts and whole plant of sesbania (Ses) and sunflower (Sun) grown either solely or as intercropping systems

Cropping system	leaves	stem	Rep. parts	Whole plant
(A) Comparisons among cropping systems for each plant species				
Sesbania				
Sole	28.7a	18.3a	136.3a	183.3a
(1ses:1sun)	13.5c	10.5b	86.0bc	110.1b
(1ses:2sun)	12.4c	8.5b	66.7c	87.7c
(2ses:1sun)	18.4b	10.1b	90.6b	119.1b
LSD 0.05	*4.2*	*4.5*	*19.3*	*21.9*
Sunflower				
Sole	50.4a	20.5a	105.0a	175.9a
(1ses:1sun)	25.3b	8.8b	65.7b	99.8b
(1ses:2sun)	21.8b	9.3b	57.2b	88.2b
(2ses:1sun)	12.3c	5.7b	37.9c	55.8c
LSD 0.05	*8.7*	*3.8*	*14.7*	*20.6*
(B) Comparisons among cropping systems				
Sole crop				
Sesbania	28.7c	18.3a	136.3ab	183.3b
Sunflower	50.4a	20.5a	105.0c	175.9b
intercropped				
(1ses:1sun)	38.8b	19.4a	151.7a	209.9a
(1ses:2sun)	34.2bc	17.8a	123.9bc	175.9b
(2ses:1sun)	30.7bc	15.7a	128.6b	175.0b
LSD 0.05	*8.4*	*4.8*	*20.9*	*25.8*

Note. For each crop species (A), and the cropping systems (B), means within a column followed by the same letter are not significantly different (P>0.05).

Carbon isotope discrimination

Data presented in Table 5 showed that ^{13}C discrimination ($\Delta^{13}C$‰) in the above ground plant materials (shoots, stems and reproductive parts) was affected by the plant species and the cropping system. Moreover, $\Delta^{13}C$‰ values of the whole plants are shown in Fig.2. It has been reported that carbon isotope discrimination ($\Delta^{13}C$‰) was considered to be a powerful tool for studying the effect of different abiotic parameters on C3 plant stomata functioning. Water deficit, temperature, salinity and sunlight (Farquhar *et al.*, 1989; Ehleringer *et al.*, 1986) decrease ‰$\Delta^{13}C$ through their influence on stomatal aperture. In this study, ‰$\Delta^{13}C$ values did not decrease in the intercropping compared to monocropping treatments, indicating that, under prevailing conditions, plants in the intercropping systems appeared not to be subjected to any abiotic stress during the experimental period. On the other hand, in some cases, whole plant ‰$\Delta^{13}C$ values in mixed stands (e.g., 1ses:2sun) were higher than that of solely grown plants. Such increments may be related to the amount of light intercepted by the component crops in the mixed stands. Ehleringer *et al.*, (1986) reported that carbon isotope discrimination decreased with increasing sunlight Correspondingly, Kurdali, (2009) suggested that stomata of the partly shaded leaves in mixed stands were more opened and carbon isotope discrimination values were higher.

Table 5. Natural abundance of nitrogen ($\delta^{15}N$‰) and carbon isotope discrimination ($\Delta^{13}C$‰) in leaves, stems and reproductive parts of sesbania (Ses) grown either alone or as intercropping systems with sunflower (Sun)

Cropping system	leaves	stem	Rep. parts
Natural abundance of nitrogen ($\delta^{15}N$‰)			
Sesbania			
Sole	+6.89a	+4.50ab	+7.17a
(1ses:1sun)	+6.89a	+6.07a	+7.37a
(1ses:2sun)	-0.41b	+3.62b	-0.82b
(2ses:1sun)	+7.51a	+4.03b	+7.3a
LSD 0.05	*1.7*	*1.7*	*1.74*
Sunflower			
Sole	+12.35ab	+13.31ab	+14.40a
(1ses:1sun)	+13.72ab	+13.58ab	+15.70a
(1ses:2sun)	+ 9.28b	+16.79a	+14.06a
(2ses:1sun)	+14.20a	+10.17b	+15.63a
LSD 0.05	*4.6*	*4.6*	*N.S*
Carbone isotope discrimination ($\Delta^{13}C$‰)			
Sesbania			
Sole	16.37b	16.50c	15.71a
(1ses:1sun)	17.13a	16.65bc	15.86a
(1ses:2sun)	16.44b	18.36a	16.38a
(2ses:1sun)	17.46a	17.68b	16.06a
LSD 0.05	*0.71*	*0.47*	*N.S*
Sunflower			
Sole	18.86b	18.26b	18.16ab
(1ses:1sun)	18.71b	18.01b	17.63b
(1ses:2sun)	18.45b	19.96a	18.38a
(2ses:1sun)	19.52a	19.04a	17.97ab
LSD 0.05	*0.53*	*0.71*	*0.72*

Note. For each crop species, means within a column followed by the same letter are not significantly different (*$P>0.05$*). Mean value of $\delta^{15}N$ of fixed N_2 determined in sesbania plants grown on N-free medium was ‰-1.37 (Kurdali 2009)

In sesbania plants, the inclusion of sunflower in the intercropping stands; particularly in 1ses:2sun treatment is accompanied with increments of $\Delta^{13}C$ values (Fig. 2) and with a decrease of $\sigma^{15}N$ (Fig.1). Such an observation was previously reported by Kurdali and Al-Shammaa (2010) in lentil plants subjected to different soil moisture levels. It is worthy to mention that the decline of $\sigma^{15}N$ was an indication of dilution of the ^{15}N isotope through a higher N_2 fixation. Moreover, the increase of $\Delta^{13}C$ values (Fig. 2) was associated with an enhancement in the amount of N_2 fixed (Table 7). This observation is consistent with that of Knight *et al.*, (1993) who reported a positive correlation between $\Delta^{13}C$ and the amount of N_2 fixed in lentil inoculated with different strains of rhizobia.

Table 6. Proportions (%) and amounts (kg N/ha) of nitrogen derived from atmosphere (Ndfa) and from soil (Ndfs) in leaves, stems, reproductive parts and the whole plant of *sesbania aculeata* (Ses) grown either solely or as intercropping systems with sunflower (Sun)

Cropping system	leaves	stem	Rep. parts	Whole plant
		%Ndfa		
Sole	45.3b	61.1ab	43.5b	45.3b
(1ses:1sun)	45.3b	50.7b	42.2b	43.4b
(1ses:2sun)	93.6a	67.0a	96.4a	93.1a
(2ses:1sun)	41.3b	64.3a	42.6b	44.2b
LSD 0.05	*11.2*	*11.5*	*11.5*	*8.9*
		Ndfa (kg/ha)		
Sole	12.7a	11.2a	59.1a	83.0a
(1ses:1sun)	6.1b	5.6b	36.0b	47.6b
(1ses:2sun)	11.6a	5.7b	63.7a	81.1a
(2ses:1sun)	7.5b	6.5b	38.8b	52.8b
LSD 0.05	*1.9*	*3.5*	*14.0*	*15.6*
		%Ndfs		
Sole	54.7a	38.9ab	56.5a	54.7a
(1ses:1sun)	54.7a	49.3a	57.9a	56.6a
(1ses:2sun)	6.4b	33.0b	3.7b	6.9b
(2ses:1sun)	58.8a	35.7b	57.4a	55.8a
LSD 0.05	*11.2*	*11.5*	*11.5*	*8.9*
		Ndfs (kg/ha)		
Sole	16.0a	7.1a	77.2a	100.3a
(1ses:1sun)	7.4b	5.0b	50.1b	62.4b
(1ses:2sun)	0.8c	2.8c	3.0c	6.6c
(2ses:1sun)	10.9b	3.6bc	51.9b	66.3b
LSD 0.05	*4.3*	*1.5*	*15.7*	*15.7*

Note. Means within a column followed by the same letter are not significantly different (*P>0.05*).

Table 7. Nitrogen balance (kg/ha) in sesbania (ses) and sunflower (Sun) grown either alone or as intercropping systems

N-Status	Sole cropping		intercropping systems					
			1ses:1sun		1ses:2sun		2ses:1sun	
	Ses	Sun	Ses	Sun	Ses	Sun	Ses	Sun
N input From N_2 fixation (leaves and stem)	+24	-	+12	-	+17	-	+14	-
N loses from soil in reproductive parts	-77	-105	-50	-66	-3	-57	-52	-38
N balance in each species	-53	-105	-38	-116	+14	-57	-38	-38
Net N balance in the cropping systems	**-53**	**-105**	**-104**		**-43**		**-76**	

CONCLUSUSION

This study provides valuable information on the impact of intercropping systems of dhaincha (*Sesbania aculeata*) and sunflower (*Helianthus annuus*) on their growth and N_2-fixation by the legume, and the interaction between both species for soil N uptake. The following conclusions were obtained from this research:

From a productivity standpoint (dry matter, N, seed and oil yield):

The 1ses:1sun surpassed the other treatments in terms of N and DM yields and exhibited a similar distribution of total DM and N uptake in the sesbania and sunflower plant species. The 1ses:2sun was second in terms of DM and N uptake showing also a similar distribution of total N in both plant species.

The 1ses:2sun gave the highest seed and oil production and together with 1ses:1sun treatment, were satisfactory in terms of LER for DM in both species having almost similar values. However, the former treatment was more appropriate than the latter because of its higher LER value for seed and oil yield of sunflower plants.

The 2ses:1sun treatment seemed to be inappropriate due to the divergence of LER values in both species, where sunflower plants had a low value as compared to sesbania.

From an ecological standpoint (N_2-fixation, soil N-uptake and balance):

The $\sigma^{15}N$ method seemed to be suitable for estimating symbiotic nitrogen fixation in sesbania plants grown in monocropping or intercropping systems with non-legumes. The best treatment was 1ses:2sun which showed the maximum N_2-fixation. Sesbania plants fixed almost identical amounts of atmospheric N_2 in both the monocropping and intercropping systems although the density of these plants in the latter was only 1/3 that of the former system.

Soil N-uptake in the 1ses:2sun was the lowest among other treatments. These results give an advantage to the 1ses:2sun over other treatments in terms of soil N consumption and N_2 fixation to meet sesbania's N requirements.

Based on ‰$\Delta^{13}C$ data, plants in the intercropping systems appeared not to be subjected to any abiotic stress.

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REFERENCES

Amarger, N., Mariotti, F., Durr, J.C., Bourguignon, C. and Lagacherie, B. (1979) Estimate of symbiotically fixed nitrogen in field grown soybeans using variations in ^{15}N natural abundance. *Plant and Soil*, **52**, 269-280.

Association of Official Analytical Chemists A.O.A.C. (1990) Official methods of analysis 966.23 (15thed.,) Association of Official Analytical Chemists., Washington D.C. 951-960.

Danso, S.K.A., Zapata, F. and Hardarson, G. (1987) Nitrogen fixation in fababeans as affected by plant population density in sole or intercropped systems with barley. *Soil Biol. Bioch.*, **19**, 411-415

Domenach, A.M., and Chalamet, A. (1979) Estimates d'azote par le soja a l'aide de deux méthodes d'analyses isotopiques. *C. R. Acad. Sci*., Paris, 289, 291-294.

Ehleringer, J.R., Field, C.B., Lin, Z.F. and Kuo, C.Y. (1986) Leaf carbon isotope and mineral composition in subtropical plants along an irradiance cline. *Oecologia,* **70**, 520-526.

Farquhar, G.D., Ehleringer, J. R. and Hubick, K.T. (1989) Carbon isotope discrimination and photosynthesis. *Annual Rev. Plant Physiol. Plant Molecul. Biol.,* **40**, 503–537.

Farquhar, G.D., O'Leary, M.H. and Berry, J.A. (1982) On the relationship between carbon isotope discrimination and the intercellular carbon dioxide concentration in leaves. *Aust. J. Plant Physiol.*, **9**, 121-137.

Fujita, K., Ogata, S., Matsumoto, K., Masuda, T., Ofosu-Budu, G.K. and Kuwata, K. (1990) Nitrogen transfer and dry matter production in soybean and sorghum mixed cropping system at different population densities. *Soil Science and Plant Nutrition*, **36**, 233-241.

Hardarson, G., Danso, S.K.A. and Zapata, F. (1988) Dinitrogen fixation measurements in alfalfa–ryegrass swards using nitrogen–15 and influence of the reference crop. *Crop Sci.*, 28, 101 –105.

Izaurralde, R.C., Mcgill, W.B. and Juma, N.G. (1992) Nitrogen fixation efficiency, interspecies N transfer, and root growth in barley field pea intercrop on a black chernozemic. *Biol. Fertil. Soil,* **13**, 11-16.

Kandel, H.J., Schneiter, A.A. and Johnson, B.L. (1997) Intercropping legumes into sunflower at different growth stages. *Crop Sci.,* 37, 1532-1537.

Kandhro, M.N., Tunio, S.D., Memon, H.R. and Ansari, M.A. (2007) Growth and yield of sunflower under influence of mungbean intercropping. *Pak. J. Agr.: Agr. Eng. Veterin. Sci.*, **23 (1),** 9-13

Knight, J.D., Verhees, F., Van Kessel, C. and Slinkard, A. E. (1993) Does carbon isotope discrimination correlate with biological nitrogen fixation? *Plant and Soil*, **153**,151-153.

Kurdali, F. (2009) Growth and Nitrogen fixation in dhaincha/sorghum and dhaincha/sunflower intercropping systems using ^{15}Nitrogen and ^{13}Carbon natural abundance techniques. *Comm. in Soil Sci. Plant Anal.*, **40 (19-20),** 2995-3014.

Kurdali, F., Al-Ain, F., and Al-Shammaa, M., and Razzouk, A.K. (2007) Performance of sorghum grown on a salt affected soil manured with dhaincha plant residues using ^{15}N isotopic dilution technique. *J. Plant Nutr.*, **30**, 1605-1621.

Kurdali, F., and Al- Shammaa, M. (2010). Natural abundances of ^{15}Nitrogen and ^{13}Carbon indicative of growth and N_2 fixation in potassium fed lentil grown under water stress. *J. Plant Nutr.*, **33 (2),** 157-174.

Kurdali, F., and Al-Ain, F. (2002) Effect of different water salinity levels on growth, nodulation and N_2-fixation by dhaincha and on growth of sunflower using a ^{15}N tracer technique. *J. Plant Nutr.*, **25**, 2483-2498.

Kurdali, F., Domenach, A.M. and Bardin, R. (1990) Alder-poplar association: determination of plant nitrogen sources by isotope techniques. *Biol. Fertil. Soil*, **9**, 321-329.

Kurdali, F., Janat, M. and Khalifa, K. (2003) Growth and nitrogen fixation and uptake in dhaincha/sorghum intercropping system under saline and non saline conditions. *Comm. in Soil Sci. Plant Anal*., **34**, 2471-2494.

Kurdali, F., Sharabi, N.E. and Arslan, A. (1996) Rainfed vetch-barley mixed cropping in the Syrian semi-arid conditions. I. nitrogen nutrition using ^{15}N isotopic dilution. *Plant and Soil*, **183**, 137-148.

Mead, R., and Willy, R.W. (1980). The Concept of a "Land Equivalent Ratio" and Advantages in Yield from Intercropping. *Exp. Agr.*, **16**, 217-228.

Muturi, E.W., Opiyo, A.M., Aguyoh, J.M. (2016): Economic efficiency of green maize intercropped with beans grown under Tithonia and inorganic fertilizer. *African Journal of Agriculure Research*. **11**, no 18, 1638-1645

Offori, F., and Stern. W.R. (1987) Evaluation of N_2-fixation and nitrogen economy of maize/ cowpea intercrop system using ^{15}N dilution method. *Plant and Soil*, **102**, 149-160.

Robinson, R.G. (1984) Sunflower for Strip, Row, and Relay Intercropping. *Agron. J.*, **76**, 43-47.

Sandhu, G.R., and Haq, M.I. (1981) Economic utilization and amelioration of salt-affected soils, In *Membrane biophysics and salt tolerance in plants*, eds. Qureshi, R.H., Muhammad, S. and Aslam, M., University of Agriculture: Faisalabad, Pakistan. pp 111-114.

Shearer, G., and Kohl, D.H. (1986) N_2 Fixation in field settings: estimates based on natural ^{15}N abundance (review). *Aust. J. Plant Physiol.*, **13**, 699-756.

Shivaramu, H.S and Shivashankar, K. (1992) Performance of sunflower (*Helianthus annuus*) and soybean (*Glycine max*) in intercropping with different plant populations and planting patterns. *Ind. J. Agron.*, **37 (2),** 231-236.

Tobita, S., Ito, O., Matsunaga, R., Rao, T.P., Rego, T.J., Johansen, C. and Yoneyama, T. (1994) Field evaluation of nitrogen fixation and use of nitrogen fertilizer by sorghum/ pigeonpea intercropping on an alifisol in the Indian semi- arid tropics. *Biol. Fertil. Soils*, **17**, 241-248.

Yilmaz, F., Atak, M., and Erayman, M. (2008) Identification of advantages of maize-legume intercropping over solitary cropping through competition indices in the East Mediterranean region. *Turk. J. Agr. Forest.*, **32**, 111-119.

Zarkawi, M., Al-Masri, M.R. and Khalifa, Kh. (2003) Research note: An observation on yield and nutritive value of *Sesbania aculeata* and its feeding to Damascus does. *Trop. Grasslands*, **37**, 187-192.