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# Evaluation of intercropping indices and fatty acid composition of safflower (*Carthamus tinctorius* L.)

**Faride Salari[1], Shiva Khalesro[1]✉, Gholamreza Heidari[1], Hamed Ghobari[2]**

1- Department of Agronomy and Plant Breeding, Faculty of Agriculture, University of Kurdistan, Sanandaj, Iran. Email: sh.khalesro@uok.ac.ir

2- Department of Plant Protection, Faculty of Agriculture, University of Kurdistan, Sanandaj, Iran

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## ABSTRACT

Intercropping plays an essential role in enhancing the biodiversity and stability of agroecosystems. The aim of this study was to investigate the optimal pattern and arrangement in safflower/chickpea intercropping in a semi-arid region of Iran. Treatments evaluated in this study were sole cropping of safflower and chickpea, their replacement series (4:4, 2:2, 1:1, 3:1, 1:3), and additive series (20% and 40% of chickpea in both situations in the middle (I) and around (II) of safflower rows). The results showed that the greatest intercropping indices such as land equivalent ratio (LER), area time equivalent ratio (ATER), and system productivity index (SPI) belonged to 40%I additive intercropping pattern. These mentioned values were 1.9, 1.8, and 307.8, respectively. The intercropping patterns had a significant effect on the fatty acid composition of safflower oil. Unsaturated fatty acids including linoleic and oleic acids were higher in the intercropping patterns, whereas, saturated fatty acids consisting of palmitic, myristic, and stearic acids were higher in the safflower sole cropping. Linoleic and oleic acid increased by 9.6% and 16.1% in 40%I compared to sole cropping. Overall, 40%I additive intercropping pattern is more promising in grain and oil yield, intercropping indices, and oil quality than the other intercropping patterns.

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## Introduction

Sustainable agriculture is one of the effective ways to promote sustainability, improve crop production, and provide food security around the world. Its aim is to create equilibrium and sustainability of ecological, economic, and social aspects in the long term (Allahyari *et al.*, 2016; Garibaldi *et al.*, 2016; Rose *et al.*, 2019). Intercropping is one of the most stable agricultural systems that promote the efficient use of land, solar radiation, and nutrients (Yin *et al.*, 2017). Intercropping enhances soil fertility (Chen *et al.*, 2019) and has a positive effect on the environment (Sharma *et al.*, 2017). Further, intercropping increases biodiversity (Afrin *et al.*, 2017; Ellahi *et al.*, 2017). In sustainable farming systems, natural self-regulatory processes (such as pest control and the nutritional cycle), are facilitated through biodiversity. On the other hand, the sole cropping has interrupted these self-regulation features, and as such, it changes into an input-dependent system (Altieri, 2004).

Safflower (*Carthamus tinctorius* L.) is a multi-purpose herb belonging to the Asteraceae family, which is vital in agriculture, industry, and medicine. This plant has high drought tolerance and can undergo dry farming (Gupta, 2016). Chemical insecticides are usually used to control pest damage in safflower farms. Meanwhile, regarding the presence of chemical pesticide residues and their impact on human health and the environment, particular attention should be paid to the methods and inputs used in the production and it justifies the need for changing the conventional agricultural systems (Kremen and Miles, 2012).

Leguminosae plants play an essential role in stabilizing nitrogen in intercropping. Chickpea (*Cicer arietinum* L.) is one of the most valuable legumes that is an essential source of nutrition for humans and animals (Muimba-Kankolongo, 2018). Thus, the use of legumes in intercropping promotes soil fertility and has led to a reduction in the use of chemical fertilizers as well as environmental contamination (Ordóñez-fernández *et al*., 2018).

There are some studies on the effect of improvement safflower yield and advantageous indices in intercrop with chickpea. Moreover, there are no reports on the effect of both additive and replacement series on safflower and chickpea without chemical inputs in intercropping system. Thus, the aim of this study was to investigate suitable intercropping patterns to reach optimal safflower production and the maximum quality of safflower oil. Further, the advantageous indices of intercropping were compared in these crop patterns.

## Material and methods

### *Experimental site*

This research was carried out at the research field of Kurdistan University in Sanandaj, Iran (longitude 47 °18' E, the latitude of 35 ° 19'N and 1865 m altitude) during the 2017 growing season. According to the Koppen climate classification, the climate in the area where the experiment was conducted is temperate with a warm and dry summer. The meteorological information on this site is shown in Fig. 1. The physicochemical properties of soil at the experimental site are given in Table 1.

### *Experimental design description*

The experimental design was a randomized complete blocks design with 11 treatments and 3 replications. Treatments included different cropping patterns: safflower sole cropping (1:0), chickpea sole cropping (0:1), intercropping replacement series: 4:4 4 rows of safflower and 4 rows of chickpea, 2:2 2 rows of safflower and 2 rows of chickpea, 1:1 1 row of safflower and 1 row of chickpea, 3:1 3 rows of safflower and 1 row of chickpea, 1:3 1 row of safflower and 3 rows of chickpea, and intercropping additive series 20%(I) 2 rows of chickpea in the middle of 8 rows of safflower, 20%( II) 2 rows of chickpea around of 8 rows of safflower, 40%(I) 4 rows of chickpea in the middle of 8 rows of safflower, 40%( II) 4 rows of chickpea in the around of 8 rows of safflower. The total rows number were 8, 10 and 12 in sole cropping and replacement intercropping patterns, 20%(I) and 20%(II), 40%(I) and 40%(II), respectively. The distance between rows was 35cm in sole cropping patterns and intercropping replacement patterns. The distance between safflower rows was 35cm and the distance between safflower and chickpea rows was 17.5 cm in the additive intercropping patterns (table 2). The length of the rows was 3.15 m. To enhance the accuracy of the biodiversity study, the distance between plots and blocks was 4 and 5 m, respectively. Both plants were sown on 13 March 2016. The experiment was conducted in dryland conditions. Weeds were controlled by hand weeding during the growing season. Both plants were planted without any fertilizer and pesticide to achieve sustainable agriculture goals.

*Determining yield, safflower oil composition, and intercropping indices*

Total numbers of chickpea plants in each plot were harvested at the maturity stage (3 July 2016) to investigate biological and seed yield. Also, safflower plants were collected on 31 August 2016 to determine biological, grain, and oil yield. To extract the safflower oil, a Soxhlet extraction with n-hexane solvent was used. The extracted oil was isolated through the rotary liquid solvent evaporation (Leal et al., 2009). Then the oil was collected in a specific glass container to analyze the composition.

Fatty acids were transformed to their methyl ester (FAME), they were analyzed using GC/MASS (Agilent 7890A). Nitrogen was the carrier gas. The fused silica capillary column DB WAX (60 m × 0.25 i.d) with 0.25 µm film thickness (Wilmington, DE, USA). The injector temperature was 260 °c and the detector temperature was 220°c.

The advantages of safflower and chickpea intercropping were assessed by calculating intercropping indices as the following formulas. Land equivalent ratio (LER) is described as the relative land area of growing sole cropping required to produce the yields achieved when growing mixes. Equation 1 (Xu et al., 2008) was used to calculate the LER.

$$\mathrm{LER} = \mathrm{Y}_{SC}/\mathrm{Y}_{SS} + \mathrm{Y}_{CS}/\mathrm{Y}_{CC} \quad (1)$$

Where $\mathrm{Y}_{SC}$ and $\mathrm{Y}_{CS}$ are the yields of safflower and chickpea in intercropping, and $\mathrm{Y}_{SS}$ and $\mathrm{Y}_{CC}$ represent their yield in sole crops, respectively.

Generally, crop production is a function of time and area, but only the area is considered for calculating the LER, while the time is not included in the calculations. When the harvesting time of two plants is different in intercropping, the index calculation of the area time equivalent ratio (ATER) is more logical alongside the land equivalent ratio. Thus, as the harvest time of safflower was different compared with chickpea about 57 days, the ATER index was calculated. To calculate this index, Equation 2 (Metwally, 2016) was used.

$$\mathrm{ATER} = {[(\mathrm{Y}_{SC} \times \mathrm{t}_S/\mathrm{Y}_{SS}) + (\mathrm{Y}_{CS} \times \mathrm{t}_C/\mathrm{Y}_{CC})]}/{t} \quad (2)$$

In this equation, $\mathrm{Y}_{SC}$ and $\mathrm{Y}_{CS}$ correspond to the yield of safflower and chickpea in intercropping, and $\mathrm{Y}_{SS}$ and $\mathrm{Y}_{CC}$ denote their yield in sole crops, respectively. The $\mathrm{t}_S$ and $\mathrm{t}_C$ are also the duration of growing safflower and chickpea in the field, respectively, with t being the total time of intercropping.

Another indicator commonly used in the economic evaluation of intercropping systems, which ultimately represents the efficiency of the intercropping system, is the system productivity index (SPI). SPI data are calculated by standardizing the secondary crop based on the main crop, whose higher values indicate the enhanced efficiency of the intercropping system. To calculate this index, Equation 3 (Agegnehu et al., 2006) was used.

$$\mathrm{SPI} = (\mathrm{Y}_{SS}/\mathrm{Y}_{CC})\mathrm{Y}_{CS} + \mathrm{Y}_{SC} \quad (3)$$

In this equation, $\mathrm{Y}_{SC}$ and $\mathrm{Y}_{CS}$ correspond to the yield of safflower and chickpea in intercropping, and $\mathrm{Y}_{SS}$ and $\mathrm{Y}_{CC}$ denote their yield in sole crops, respectively.

Statistical analysis

The obtained data underwent analysis of variance (ANOVA) using SAS statistical software (SAS Version 9.1), where means were compared using the least significant difference test (L.S.D).

## Results

*Safflower and chickpea yield*

seed yield, biological yield, and oil yield of safflower were significantly affected by different cropping patterns (P≤0.01) (Table 3). The greatest seed, biological, and oil yields (266.32, 1146.7, and 78.33 g/m$^2$) were obtained from 40%I and the lowest values (74.04, 321, and 22.13 g/m$^2$) from the 4:4 cropping pattern, respectively (Table 4). The results showed that the effect of cropping patterns on the seed yield, and biological yield of chickpea was significant at 1% probability (Table 3). The highest and lowest seed as well as biological yield of chickpea belonged to sole cropping of chickpea and 20%I ratio, respectively (Table 4). The higher yield of chickpea is expectable, considering its higher proportion in sole cropping.

*Safflower oil composition*

Results showed that the cropping patterns had a significant effect on the safflower oil composition (P≤0.01) (Table 5). Polyunsaturated linoleic acid was the major component followed by monounsaturated oleic acid. The intercropping patterns significantly increased unsaturated fatty acids. The highest percentage of linoleic (75.97%) and oleic acid (14.61%) belonged to the 40%I ratio. This mentioned pattern increased the content of linoleic and oleic acid by 9.56% and 16.11% compared to sole cropping, respectively. Whereas, saturated fatty acids including palmitic, myristic, and stearic acids decreased significantly affected by the intercropping patterns. The highest percentage of palmitic acid (6.02%) and stearic acid (2.67%) belonged to safflower sole cropping (Table 6). Thus, the intercropping patterns improved safflower oil quality compared to sole cropping.

*Intercropping indices*

*Land Equivalent Ratio (LER)*

Based on the results, the intercropping patterns had a significant effect on the LER at 1% probability (Table 7). The greatest amount of LER (1.91) was obtained from the 40%I ratio, and its lowest value (0.75) belonged to the 4:4 cropping pattern (Figure 2.A). Assigning the greatest LER to the additive cropping pattern 40%I can be attributed to better use of resources.

*Area Time Equivalent Ratio (ATER)*

The ATER values calculated for intercropping patterns showed a similar trend to the LER index in these treatments, and it indicated that intercropping could be more efficient than monoculture. Note that the success of intercropping varies with respect to the plant species used in the field. The greatest (1.82) and lowest (0.65) values of ATER were obtained from the 40%I and 4:4 cropping patterns, respectively (Figure 2.B). Thus, the 40%I cropping pattern had the highest productivity and efficiency.

*System productivity index (SPI)*

The cropping patterns had a significant effect on the system productivity index (SPI) at a probability level of 1% (Table 7). The greatest (308.72) and the lowest (12.29) SPI were obtained from the 40%I and 4:4 cropping patterns, respectively (Figure 2.C).

## Discussion

The results of this study revealed that intercropping improved the grain, biological, and oil yield of safflower. In the previous studies, it was determined that peppermint yield increased in the intercropping system with fenugreek (Ebrahimghochim *et al*., 2018). It seems that intercropping pattern plays a vital role in increasing grain and oil yield per unit area. Increasing the number of safflower rows in additive intercropping patterns and 3:1 replacement series enhanced the safflower yield at these patterns. This increment can be due to biological nitrogen fixation by chickpea, lower competition of safflower with chickpea, and optimal uptake of nutrients by safflower in the proper intercropping patterns. Other researchers have reported a positive effect of nitrogen-fixing plants in intercropping (Duchene *et al*., 2017). The interspecies competition was less than the intra-species competition in additive intercropping pattern and thus, the yield increased. Thus, the ecological superiority of intercropping is the result of more efficient use of resources (Moghbeli *et al*., 2019).

Linoleic and oleic acids have a positive effect on human health. The results showed that additive and replacement intercropping patterns increased these unsaturated fatty acids significantly. The highest amount of them belonged to the 40%I ratio. This increase was attributed to the greater light, water, and nutrient received by plants in the intercropping system with those grown in sole cropping. Further, safflower oil quality may be influenced by the cropping system (Saeidi *et al*., 2018). The saturated fatty acids decreased significantly in all of the intercropping patterns. Consequently, the intercropping system improved safflower oil quality. The findings of this study were similar to the results previously in fennel intercropped with common bean[41] and safflower intercropped with faba bean (Rezaei-Chiyaneh *et al*., 2020). In terms of chickpea, the greatest grain and biological yield were obtained from a sole crop of it. Chickpea was the non-dominant plant in these intercropping patterns. Our results are in agreement with Other researchers who have reported that yield reduction is related to competition for absorbing resources, including light for a non-dominant plant (Zhang *et al*., 2008).

In general, total LER values for most of the intercropping patterns in this study have been more than one, suggesting the benefits of intercropping in comparison with sole cropping. This result is in line with the other studies (Ramkat *et al*., 2008; Jamshidi, 2011; Mao *et al*., 2012). The higher value of LER could be related to the positive interaction between two plants, biological nitrogen fixation, and availability of this macroelement (Bhatti *et al*., 2006). In other words, along with the presence of species beside each other, the competition for using environmental resources is increased, but if one of the species has the ability of nitrogen fixation, competitive pressure is reduced, with legume species showing less competition with the main crops (Vandermeer, 1992). The reason for LER reduction in the 4:4 cropping pattern was a decline in the facilitating effects of two species with increasing the strip width. LER's superiority in the additive series over replacement series indicates that the yield was elevated in these treatments. According to other researchers, the most crucial reason for LER elevation in additive series is the enhanced yield in these treatments (John and Mini, 2006). The results of ATER were similar to LER. It can prove the usefulness of the intercropping system of safflower and chickpea compared to their sole cropping.

Generally, we had the highest SPI in the treatments with the higher safflower density, including all additive series and 3: 1 replacement intercropping pattern. Similar results have been reported in barley and faba bean intercropping (Agegnehu *et al*, 2006). Similarly, the other researchers found a superior intercropping system of fennel and dill with a higher density of fennel compared to their sole cropping (Carruba *et al*., 2002).

## Conclusion

The results indicated that safflower/chickpea intercropping increased seed, biological, and oil yield of safflower significantly compared with sole cropping. The maximum values of mentioned yields belonged to a 40%I additive intercropping pattern. Furthermore, intercropping indices such as ATER, LER, and SPI, proved the superiority of intercropping compared to sole cropping of safflower. The greatest values of intercropping indices were obtained from the 40%I additive intercropping pattern. The additive and replacement intercropping patterns had a positive effect on the safflower oil quality. The highest percentage of unsaturated fatty acids included linoleic and oleic acids and the lowest saturated fatty acids such as palmitic, myristic, and stearic acids belonged to a 40%I pattern. Overall, 40%I additive intercropping pattern was the efficient pattern that enhanced both yields and oil quality compared with sole cropping of safflower.

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