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The Effect of Green Manures on Some Agronomic and Chemical Aspects of Sunflower (*Helianthus annuus* L.) under Different Nutrition Systems in Ahvaz Climatic Conditions

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ABSTRACT

In sustainable agriculture, the application of green manure, serving as an alternative to chemical fertilizers, holds great importance. To investigate the effect of green manure plants and different nutrition systems on some agronomic traits and seed chemical composition of sunflower, a field experiment was conducted during the 2023–2024 growing season at the Research Farm of the Faculty of Agriculture, Shahid Chamran University of Ahvaz, Iran. The experiment was arranged in a split-plot design in a randomized complete block with three replications. The main plots consisted of green manure plants at four levels: 1- control, 2- barley green manure, 3- berseem clover green manure and 4- mixed green manure of barley and berseem clover. The sub-plots consisted of different nutrition management systems: 1- control, 2- 100% chemical system 3- integrated system and 4- organic system. Results indicated that the highest seed yield and number of seeds per head, the lowest empty seeds percentage and the highest seed oil yield per unit area were obtained from the mixed barley and berseem clover green manure treatment under the chemical system, with values of 6.29 tons ha^{-1}, 876.17 seeds, 1.87%, and 1914.76 kg per hectare, respectively. The highest protein yield per unit area was obtained from the berseem clover green manure treatment under chemical fertilization, with a value of 718.84 kg per hectare. The lowest seed yield of 3.5 tons ha^{-1} was obtained from the barley treatment under the control without fertilization. Choosing the mixture of barley and clover along with chemical fertilizer resulted in the highest seed and oil yields and based on LSD comparison, placed it in the superior statistical group for protein yield as well. However, for sustainable agriculture and high seed yield, while also achieving high oil and protein yields, cultivation of berseem clover green manure with the integrated system is recommended.

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1. Introduction

Due to the continuous expansion of the human population, sunflower (*Helianthus annuus* L.) and other oilseeds currently dominate the international oilseed market (Islam *et al.*, 2024). Sunflower is recognized as a significant source of superior edible oil, primarily used for culinary purposes. Sunflower oil is in high demand due to its favorable fatty acid profile, particularly its richness in polyunsaturated fatty acids. (Pal *et al.*, 2015; Ramamoorthy *et al.*, 2023). The cultivation of sunflower for the commercial production of oil from its seeds began in Iran with the establishment of a joint-stock company for large-scale oilseed development in 1996. The provinces of Ardabil, East Azerbaijan, West Azerbaijan, and Fars are the most important sunflower producers (Fatahi *et al.*, 2022). Sunflower meal is one of the most abundant agricultural-industrial residues produced worldwide, rich in protein, fiber, and minerals, and is currently used mainly as a component in livestock feed formulations. However, its potential for human nutrition remains largely untapped (Becze *et al.*, 2025). Sunflower seeds contain approximately 15 g of protein, 58 g of total fat, 3 g of ash and 24 g of carbohydrates,

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with a total energy of 675 kcal per 100 g (do Prado *et al*., 2021). Ash obtained from sunflower plant waste, particularly from material used as fuel, has the potential to become an important natural fertilizer containing plant nutrients. Leaf and stem ash are also considered chemical traits for evaluating the nutritional status of sunflower. Sunflower waste ash has a very high content of phosphorus and potassium, and these ashes can be used as fertilizer, especially in soils deficient in these elements (Gul, 2019).

Achieving sustainable agricultural intensification is crucial to meet the food demand of the growing population, which is projected to exceed 10 billion by 2100 (FAO, 2018; UN, 2019). To meet future food needs, a significant 48.6% increase in global food production is required. However, in modern agricultural practices, the overuse of mineral fertilizers not only leads to extensive pollution of soil, atmosphere, and water resources but also results in the degradation of soil properties and food production (Zhao *et al*., 2024). In recent years, the expansion of legume and cereal cultivation has become a major focus in modern sustainable agriculture to improve land fertility (Patil *et al*., 2020). Intercropping legumes and cereals in sustainable farming systems offers numerous advantages, including soil conservation, weed control, livestock feed, efficient land use, and higher system yield and quality (Ton, 2021). Cultivating leguminous plants as green manure, due to their ability for biological nitrogen fixation (Jalilian *et al*., 2021), and harvesting protein-rich seeds are critical strategies farmers use to increase plant protein production (Mukhametov *et al*., 2021). In addition to biological nitrogen fixation, these plants, with their deep and dense root systems, have a greater capacity to extract and recycle nutrients (Jalilian *et al*., 2021). Cereals are not only an important source of plant protein but also an essential component of alternative agricultural systems (Mukhametov *et al*., 2021).

Although green manures are effective in supplying nutrients, the release of nutrients may not coincide with the peak demand of the crop. Furthermore, the mineralization process of nitrogen in green manures depends on the carbon-to-nitrogen ratio. Therefore, the success of green manure application depends on selecting the appropriate species (Jalilian *et al*., 2021). Organic fertilizers also play a vital role in soil fertility, microbial communities, and sustainable agricultural strategies. In organic farming, fertilizers are often applied without a precise calculation of the nutrients added to the soil. The amount of nutrients supplied by organic fertilizers to the crop must equal that supplied by chemical fertilizers for them to be viable replacements. Since organic fertilizers release nutrients slowly over time (Joseph and Babu, 2025) and fertilizers are scarce and expensive inputs, their judicious application is essential to achieve greater benefits (Mallick and Majumder, 2023).

As no single source can supply the required amount of plant nutrients, the integrated use of all nutrient sources is essential to provide balanced crop nutrition. In this context, integrated soil nutrient management, involving the intelligent combination of organic, biofertilizers, and chemical fertilizers, appears to be a practical option for sustainable and profitable commercial-scale agriculture. Therefore, emphasis is placed on using organic fertilizers such as vermicompost and biofertilizers like *Azotobacter*, *Azospirillum*, and phosphate-solubilizing bacteria (PSB) (Mallik and Majumder, 2023). Organic fertilizer compositions using animal bone meal, neem cake, farmyard manure, and vermicompost recorded better growth and yield in sunflower plants compared to mineral chemical fertilizers (Joseph and Babu, 2025). In a study of various treatments including: 100% chemical fertilizers, farmyard manure (10 and 5 t ha^{-1}), vermicompost (5 and 2.5 t ha^{-1}), biofertilizers, and foliar spray, it was found that the treatment of 2.5 t ha^{-1} vermicompost + 50% recommended chemical fertilizers + biofertilizers + foliar spray produced the maximum seed yield (2.48 t ha^{-1}), number of seeds per head (1051), and seed weight (29.31 g) (Mallik and Majumdar, 2023).

In a study on the effect of using two organic fertilizers from different sources (co-composted with biofertilizer, CCB; filter mud cake, FMC) compared to common mineral fertilizers on the quality of sunflower seeds and oil, data indicated that the use of organic fertilizer, compared to the mineral treatment, contributed to a significant increase in the production yield of sunflower seeds and oil. Results of the chemical composition of sunflower seeds showed no significant difference in protein and ash content among all treatments, while a significant difference was observed in oil content, where FMC recorded the highest oil content, followed by CCB, and finally the mineral treatment (Alzamel *et al.*, 2022). An examination of two levels (with and without vetch green manure) and four different nutritional systems revealed that the presence of green manure in the cropping system increased the number of seeds per sunflower head. Evaluation of green manures, including pea, soybean, and triticale, revealed that sunflower seed yield increased by more than 1000 kg ha^{-1} compared to the control. Chemical fertilizer application resulted in a 398 kg ha^{-1} increase relative to the unfertilized treatment. The triticale treatment exhibited the highest seed oil content and the greatest 1000-seed weight. These findings confirm the role of green manures in enhancing soil health, yield, and seed quality, while also promoting sustainable agricultural practices (Peter-Balázs *et al.*, 2024). Over 90% of the country's demand for oil is met through imports (Piravi-Vanak *et al*., 2024). Additionally, the use of plant ash as a sustainable cementitious material in concrete composites is a topic widely researched in the construction field (Șerbănoiu *et al.*, 2021).

Iran also has high potential to increase the cultivated area of sunflower. Therefore, with the objectives of reducing the effects of chemical fertilizers in sunflower production, meeting the country's oil needs, and investigating sunflower plant ash, an experiment titled "The effect of different green manures and various nutrition systems, including mineral, organic, and biofertilizers and their combinations on some agronomic and chemical aspects of sunflower" was conducted in Ahvaz.

2. Materials and methods

2.1. Experimental site and conditions

This research was conducted to investigate the effect of green manure and different nutrition management systems on some agronomic and chemical aspects of sunflower under the climatic conditions of Khuzestan province. The experiment was carried out during the 2023-2024 growing season at the educational-research farm of the Faculty of Agriculture, Shahid Chamran University of Ahvaz. The site is located in the southwestern part of Ahvaz, on the western bank of the Karun River, at a latitude of 31° 19' N, longitude of 48° 41' E, and an altitude of 22 meters above sea level. The city of Ahvaz, in southern Khuzestan province, is based on the classification of the hierarchical cluster analysis method, it is considered part of the hot and dry climate (Movahedi *et al.*, 2012). The meteorological data for the experimental year are presented in Table 1.

Table 1. Mean monthly meteorological conditions during the growing season in the experimental year (2023-2024)

Parameters	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun
Maximum temperature (°C)	30.8	23.3	22.4	22.4	22.6	30.5	26.2	45
Minimum temperature (°C)	17.4	10.6	10.1	10.4	11.5	16.7	21.8	27.2
Mean daily temperature (°C)	24.1	17	16.3	16.4	18	23.7	29	36.1
Rainfall (mm)	48.6	9.9	7.9	21.6	86.9	38.8	36.1	0

Source: Iran Meteorological Organization

2.2. Soil characteristics

Prior to initiating the experiment, soil samples were collected from a depth of 0-30 cm to determine the physical and chemical properties of the soil. The samples were transferred to the laboratory for analysis. The results are presented in Table 2.

Table 2. Physical and chemical characteristics of the experimental site soil.

Soil texture	EC (ds m^{-1})	PH	Organic matter	Nitrogen	Phosphorus	Potassium
			(%)		(mg kg^{-1})	
Sandy loam	0.67	7.62	0.25	0.01	8.36	263.91

2.3. Experimental design and treatments

The experiment was arranged as a split-plot design based on a randomized complete block design with three replications. The main plot factor consisted of four green manure treatments: Control (no green manure), Barley green manure (*Hordeum vulgare* L.), Berseem clover green manure (*Trifolium alexandrinum* L.), and a mixture of barley and berseem clover green manure. The sub-plot factor comprised four different nutrition management systems: Control (no chemical or organic fertilizers), Full chemical fertilizer system (nitrogen, phosphorus, and potassium based on soil test recommendations), Integrated system: 50% of the recommended nitrogen, phosphorus, and potassium + two nitrogen-fixing bacteria (*Bacillus* sp. strain SS4 and *Staphylococcus xylosus* strain SR2) + phosphate biofertilizer (Phosphate Barvar-2) + 50% vermicompost (equivalent to 4 tons ha^{-1}).

The organic system consisted of two nitrogen-fixing bacteria (*Bacillus* sp. strain SS4 and *Staphylococcus xylosus* strain SR2) + phosphate biofertilizer (Phosphate Barvar-2) + 100% vermicompost (equivalent to 8 tons ha^{-1}) + soil application of humic acid (30 kg ha^{-1}) at both vegetative and reproductive stages + foliar application of seaweed extract (*Ascophyllum nodosum*, commercial name: Super Gro) at both vegetative and reproductive stages.

2.4. Agronomic practices

Land preparation for the green manure crops began after initial irrigation and upon reaching optimal soil moisture (field capacity) in late November. Primary tillage was performed using a moldboard plow, followed by disking and land leveling. Irrigation channels were created, and main plots were delineated. Each main plot measured 12 m in length and 4 m in width, separated by bunds. Green manure crops (barley, berseem clover, and their mixture) were sown manually on November 29, 2023. The seeding rates were 190 kg ha^{-1} for local barley (Karoun cultivar), 25 kg ha^{-1} for berseem clover, and half the monoculture density for each in the mixed treatment. Irrigation was performed every 7 to 10 days. The green manure plants were later incorporated into the soil using a hand cultivator. Subsequent land preparation for sunflower cultivation included creating planting rows using a furrower. Sunflower seeds cultivar "Lakomka" (obtained from the Seed and Plant Improvement Institute, Karaj) were sown on February 13, 2024. Planting holes, 4-5 cm deep, were made manually. The seeding rate was 6 kg ha^{-1}, with a target plant density of 8 plants m^{-2}, achieved by spacing rows 60 cm apart and plants 20 cm apart within rows. Each sub-plot consisted of 5 rows.

2.5. Fertilizer and amendment application

In the full chemical system, based on soil test results and critical nutrient levels, fertilizers were applied at rates of 200 kg N ha^{-1} (from urea, split into three equal applications: at emergence, two weeks after planting, and early bud formation), 100 kg P_2O_5 ha^{-1} (from triple superphosphate), and 100 kg K_2O ha^{-1} (from potassium sulfate). Phosphorus and potassium were applied basally during land preparation for sunflower.

In the integrated system, 50% of the recommended phosphorus and potassium chemical fertilizers, along with 4 tons ha^{-1} of vermicompost, were applied basally at planting. Sunflower seeds were inoculated with Phosphate Barvar-2 and the nitrogen-fixing bacterial mixture (*Bacillus* sp. SS4 and *S. xylosus* SR2). Nitrogen was applied as a chemical system.

In the organic system, seed inoculation was performed as in an integrated system. The full dose of vermicompost (8 tons ha^{-1}) was applied basally. Humic acid (30 kg ha^{-1}) was applied to the soil at the 4-leaf stage and at early flowering via irrigation water. Seaweed extract (15-20 drops per liter) was sprayed at the vegetative (2-4 leaf) and flowering stages.

2.6. Crop management

The first irrigation was applied immediately after sowing using the basin method. Subsequent irrigations were scheduled based on crop demand and critical phenological stages, approximately every 7 days during the first three months and every 10 days towards the end of the season (excluding rainfall events). Thinning was performed at the 3-4 leaf stage to achieve the target plant density. Weed control was carried out manually (hand weeding) uniformly across all plots before flowering. No significant plant diseases were observed during the growing season. To prevent bird damage (sparrows and parrots) during head formation and seed filling, all experimental plots were covered with protective nets.

2.7. Harvest and measured parameters

Final harvest took place in late June 2024 at physiological maturity, indicated by the yellowing and browning of bracts and the browning of the back of the sunflower heads. To eliminate border effects, 15 plants were harvested from the three middle rows of each sub-plot (sampling area: 1.2 m²). The following parameters were measured:

Seed number per head was determined by counting all seeds and calculating the mean per head. Thousand-seed weight (TSW) was measured after drying samples in an oven at 40°C to constant weight; six random subsamples of 100 seeds were weighed using a digital balance (0.001 g precision), and TSW was calculated proportionally. Seed yield was determined based on the harvested area. Empty seed percentage was calculated as the proportion of empty seeds relative to total seeds per head. Oil content was determined using a Soxhlet extraction apparatus according to the method described by Latif and Anwar (2008). Oil content was calculated using Equation 1.

1) $$Oil\ (\%) = \frac{W1 - W2}{Weight\ of\ ground\ sample} \times 100$$

In this equation, W1 is the initial weight of the sample and filter paper, and W2 is the weight of the final sample and filter paper. Protein percentage was determined using the Kjeldahl method (Jones, 2001). Oil yield and protein yield were calculated by multiplying seed yield by oil and protein percentages, respectively. The crude ash percentage of vegetative parts (a mixture of approximately 70% stems and 30% leaves) was determined using the method described by Khoshgoftarmanesh et al. (2010).

2.8. Statistical analysis

Analysis of variance (ANOVA) was performed on the collected data using SAS statistical software (version 9.4). Mean comparisons were conducted using the Least Significant Difference (LSD) test at the 5% probability level.

3. Results and discussion

3.1. Number of seeds per head

The effects of green manure, nutrition system and their interactions on the number of seeds per head were significant (Table 3). Among the treatment combinations, the mixture of barley and berseem clover green manure combined with chemical management produced the highest number of seeds per head, with an average of 876.17. In contrast, the lowest number of seeds per head was recorded in the mixture of barley and berseem clover combined with the organic system, with an average of 566.54 seeds (Fig. 1). The reason for the increase in the number of seeds in the superior treatment was the initial nitrogen supply by clover in mixed cultivation and the correction of nitrogen and other nutrient deficiencies at sensitive growth stages, especially in the vegetative and head emergence stages, using the chemical nutrition system.

The findings of this study are consistent with the results of Shoghi Kalkhoran et al. (2012), who showed that the number of seeds per head in sunflower in systems containing green manure with 50% manure and 50% chemical fertilizer was significantly higher than in systems with only chemical or organic fertilizers. Also, Keshta et al. (2008) reported that the application of chemical nitrogen fertilizer (45 kg of nitrogen per 4200 m²) in saline soil increased the number of sunflower seeds compared to other treatments. In another superior treatment, including clover green manure and control without fertilizer, clover, as a legume with nitrogen fixation ability, by storing and gradually releasing nitrogen in the soil, provided conditions for the formation of more reproductive florets. Legume plants are able to fix nitrogen through symbiosis with Rhizobium bacteria (Nazarian *et al.*, 2019). This issue shows the importance of legume green manure in nitrogen supply and its residual effect. As a result, the treatments of clover green manure and the control system (without fertilizer) were statistically placed at the same level as the mixed treatment of barley and clover with chemical management, in terms of the number of seeds per head. Therefore, to reduce the effects of chemical fertilizers, the clover green manure treatment and the control system (without fertilizer) can be used to achieve a higher number of seeds per head. Also, Daryayi et al. (2012) in their study reported that the highest number of seeds per head in sunflower was observed in green manure treatments with 100% zeoponic and a combination of zeoponic and chemical fertilizer.

Table 3. Analysis of variance for some agronomic traits of sunflower under experimental treatments

S.O.V.	d.f.	Number of seeds per head	1000 seed weight	Empty seed percent	Seed yield
Block	2	675.32^{ns}	11.09^{ns}	0.34^{ns}	0.001^{ns}
Green manure (G)	3	35418.25**	228.56**	23.64**	3.257**
Error a	6	2718.26	28.14	0.41	0.249
Nutrition system (N)	3	23376.62**	255.88**	2.30**	3.559**
G × N	9	30714.41**	87.81**	1.06*	1.711**
Error b	24	2378.47	24.66	0.39	0.226
CV (%)		7.13	5.97	18.48	10.11

*, ** and ns indicate significance at the 5%, 1% and no probability levels, respectively

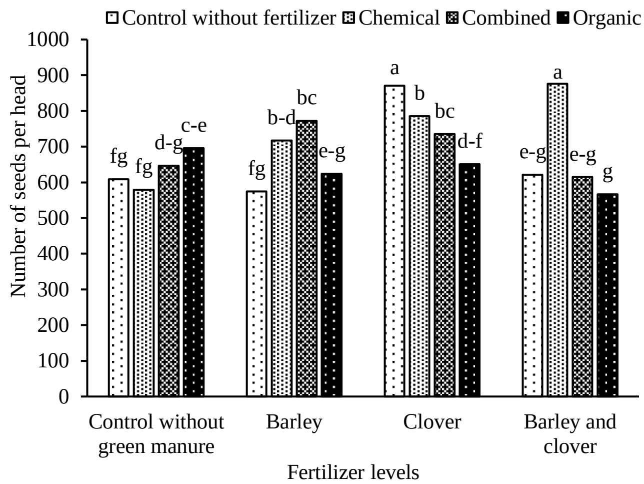

Figure 1. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the number of seeds per head of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

3.2. 1000-seed weight

Analysis of variance in Table 3 revealed significant effects of green manure, nutrition system, and their interaction on 1000-seed weight. The highest 1000-seed weight (96.48 g) was recorded for the mixed barley and clover green manure treatment under the integrated nutrition system, placing it in the superior statistical group. Treatments with desirable values, such as the mixed barley and clover with the chemical nutrition system weighing 91.75 g and clover green manure with the integrated nutrition system weighing 91.85 g, were also placed in the same statistical group (Fig. 2).

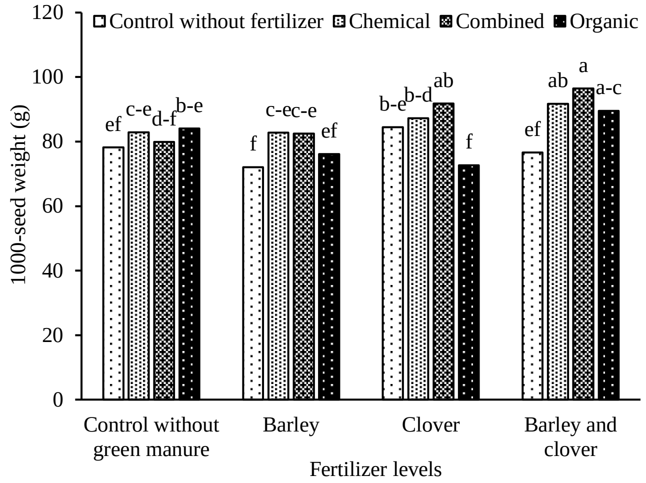

Figure 2. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the 1000-seed weight of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

The lowest 1000-grain weight (72.01 g) was observed in the green manure treatment of barley without any chemical or organic fertilizer application, which was not significantly different from the clover treatment under the organic nutrition system (72.7 g). In the barley treatment without supplemental fertilization, there was a severe deficiency of nitrogen and phosphorus. This appears to be due to the consumption of soil nitrogen by microorganisms during the decomposition of barley residues with a high carbon-to-nitrogen (C/N) ratio, which is consistent with the report by Loomis et al. (2020). As a result, sufficient photosynthetic assimilates were not available for grain filling. In the clover treatment under the organic system, phosphorus was likely unavailable for plant uptake, possibly due to salinity or incomplete decomposition of organic manure. The mixed barley and clover green manure improves root penetration and soil structure, creating a strong root system that can effectively absorb water and nutrients during the seed-filling period, which is often associated with heat stress. Moreover, during the seed-filling stage, optimal nutrient supply through the integrated nutrition system directs assimilates toward the seeds, where they are converted into oil and protein.

Izan et al. (2019) reported that in an integrated system combining 50% chemical fertilizer with phosphorus and potassium supply, biofertilizers, and vermicompost (providing gradual nutrient release and improving soil health), favorable conditions for seed filling are established. The slow release of nutrients from vermicompost reduces the risk of nutrient leaching and supports plants until the end of the growing season. In the study by Souques et al. (2025), it was found that cover crop mixtures can control nitrogen release timing and prolong the seed-filling period, directly influencing 1000-seed weight and crop yield. The present finding of increased 1000-seed weight aligns with the results of Shoghi Kalkhoran et al. (2012), who reported that green manure and the integrated system (50% chemical and 50% organic nutrition) significantly increased sunflower 1000-seed weight. Similarly, in the study by Daryaei et al. (2012), the highest number of seeds per head was observed in treatments with green manure combined with 100% zeoponic fertilizer or a mixture of zeoponic and chemical fertilizers.

An inverse and compensatory relationship often exists between the number of seeds per head and 1000-seed weight. In the treatment with clover green manure and no fertilizer, the seed count is high, but the 1000-seed weight (84.5 g) is lower compared to the mixed barley and clover green manure with the integrated system, which recorded 96.48 g. When the number of seeds increases significantly, plant resources may be distributed among more seeds, resulting in reduced individual seed weight. In treatment clover green manure and no fertilizer, the plant focuses its energy on producing a larger number of seeds (though smaller in size), whereas in treatment mixed barley and clover green manure with the integrated system, optimal and balanced nutrient supply throughout the growing season leads to heavier seeds. The study by Arif et al. (2017) demonstrated that the increase in 1000-seed weight, attributable to an extended seed-filling period, may reflect the influence of plant growth-promoting bacteria and the integrated nutrition system on seed yield through enhanced accumulation of photosynthetic reserves during the prolonged seed-filling phase under varying nutrition and fertilizer regimes.

3.3. Empty seeds percentage

The results indicated that the main effects of green manure and nutrition system, as well as their interaction, had a significant impact on empty seed percentage (Table 3). Among the treatment combinations, barley green manure with the integrated nutrition system showed the highest empty seed percentage, with a mean of 5.85%. In contrast, the lowest empty seed percentage (1.87%) was observed in the treatment combining mixed barley and berseem clover with the chemical nutrition system (Fig. 3).

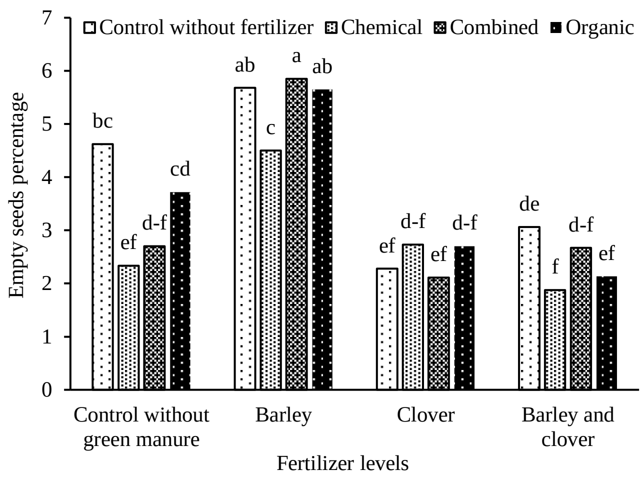

Figure 3. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the empty seeds percentage of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

Empty seeds indicate unsuccessful pollination or incomplete seed filling, and a lower value of this trait reflects more balanced plant nutrition and field health during pollination and seed-filling periods. The cultivation of clover green manure and mixed barley and clover at all nutrition levels had a greater effect on reducing empty seed percentage compared to barley green manure. In contrast, using barley green manure alone led to increased empty seeds due to limited nutrient supply at certain growth stages. The superior treatment barley-clover mixture and the chemical nutrition system demonstrate that the combination of mixed green manure and chemical nutrition created favorable conditions for seed formation and filling, thereby reducing empty seeds. This finding aligns with the results of Mohammadi et al. (2011), who reported that the interaction between green manure and nutrition system significantly influenced seed yield and the concentration of nitrogen, iron, and magnesium in seeds, which can contribute to balanced plant nutrition and reduced empty seeds. Furthermore, Yousefpoor and Yadavi (2014) showed that the full application of required potassium sulfate along with urea and seed inoculation with two types of nitrogen-fixing biofertilizers (*Azotobacter* and *Nitroxin)* had the most positive effect on increasing the kernel percentage of sunflower seeds.

3.4. Seed yield

The effect of green manure, nutrition system and their interaction on seed yield was significant (Table 3). Among the treatment combinations, the mixture of barley and berseem clover with chemical management showed the highest seed yield with an average of 6.29 tons per hectare, which was not significantly different from clover green manure under the no fertilizer, chemical and integrated nutrition systems. The lowest seed yield was observed in the combination of barley green manure without the use of chemical and organic inputs, with an average of 3.5 tons per hectare (Fig. 4).

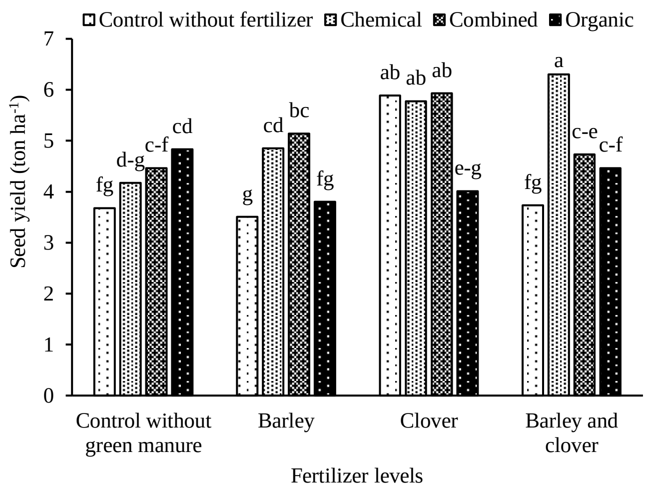

Figure 4. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the number of seed yield of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

It can be inferred that in clover green manure, due to the efficiency of clover in supplying the nitrogen required by the plant through biological fixation and rapid decomposition of residues with a low carbon-to-nitrogen (C/N) ratio, there is no significant difference among the control (no fertilizer), chemical, and integrated nutrition treatments. Therefore, if clover is used as green manure, from an economic perspective, there is no need for chemical or integrated fertilization. However, if the goal is to achieve the maximum grain yield in clover green manure, the integrated system resulted in a higher grain yield due to better nutrient balance. This finding is consistent with the results of Brtnicky et al. (2021) regarding the importance of green manure, as well as the study by Zamanian and Yazdandoost (2021) on increasing sunflower grain yield through the use of chemical fertilizers and seed inoculation with Nitroxin and Biosfer fertilizers. However, with the introduction of the chemical nutrition system alongside barley green manure and mixed barley and clover green manure, seed yield increased.

In the barley-clover mixture and the chemical nutrition treatment, the highest seed yield was achieved, where the mixed green manure improved soil structure and provided gradual nitrogen release (from clover), creating an enriched initial substrate. Subsequently, the chemical system precisely and directly addressed any remaining nutrient deficiencies, driving the plant to its maximum yield potential. This indicates that even with barley or mixed barley-clover green manure, sunflower as a high-demand crop still requires significant amounts of readily available nutrients (especially at critical stages). Eskandari (2017) reported that intercropping cereals and legumes is superior to their monoculture.

3.5. Oil percentage and yield

The effects of green manure, nutrition system, and their interaction on both oil percentage and yield were significant (Table 4).

Table 4. Analysis of variance for some chemical traits of sunflower under experimental treatments.

S.O.V.	d.f.	Seed oil percent	Seed oil yield	Seed protein percent	Seed protein yield	Ash percent
Block	2	6.91^{ns}	11878^{ns}	10.08**	22644*	9.50^{ns}
Green manure (G)	3	79.31**	92486**	2.32^{ns}	25615*	4.24^{ns}
Error a	6	6.41	5393	0.77	3201	12.28
Nutrition system (N)	3	155.58**	987291**	0.08^{ns}	29175**	21.42*
G × N	9	43.16*	235728*	0.09*	18716**	16.05*
Error b	24	15.61	73502	0.041	5201	5.06
CV (%)		14.68	21.36	8.84	12.49	14.2

* and ** indicate significance at the 5% and 1% probability levels, respectively.

Among the treatment combinations, the highest oil percentage (35.99%) was recorded in the control without green manure under the integrated nutrition system, while the lowest (16.63%) was observed in the clover treatment under the organic system. (Fig. 5). In the absence of green manure, less energy is spent on residue decomposition, vegetative growth is reduced, and the plant focuses more on seed and oil production. The addition of the integrated system provides a better balance of nutrients suitable for oil synthesis. Shoghi Kalkhoran et al. (2012) reported that seed inoculation with growth-promoting bacteria improved sunflower oil percentage compared to the control.

Similarly, Khodaei Joghan et al. (2018) found that the highest seed oil percentage under optimal irrigation was obtained from integrated organic treatments containing vermicompost. The control treatment without green manure and fertilization also ranked in the superior statistical group in terms of oil percentage, indicating a defensive plant response under nutrient stress, as oils and fatty acids play protective roles and the plant accelerates oil synthesis through early maturation (Tong *et al*., 2025). However, despite the high oil percentage in this treatment, lower seed yield ultimately resulted in reduced oil yield per hectare (Fig. 5).

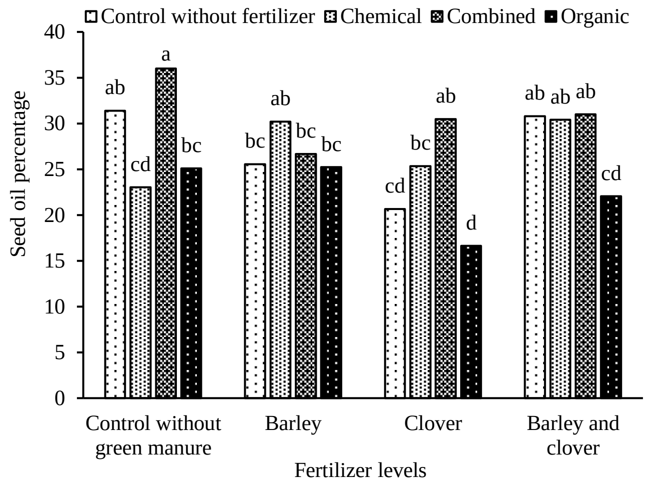

Figure 5. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the seed oil percentage of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

The highest oil yield (1914.76 kg ha^{-1}) was achieved with the mixed barley and clover green manure under the chemical system, while the lowest (658.82 kg ha^{-1}) was recorded for clover under the organic system (Fig. 6). The synergistic effect of green manures, through improved soil conditions and nitrogen supply, combined with the rapid availability of mineral elements at critical times via chemical nutrition, maintains a balance between vegetative and reproductive growth essential for oil synthesis. Treatments including clover with the integrated system, barley with the chemical system, and mixed barley-clover with the integrated system also ranked high statistically, highlighting the role of green manures alongside chemical and integrated nutrition in enhancing oil synthesis.

Javanmard et al. (2022) reported that the highest oil percentage and yield were obtained from a green manure treatment consisting of 50% pea + 50% barley. The control treatment with the integrated system also showed an oil yield of 1603.99 kg ha^{-1}, indicating that even without green manure, an appropriate oil yield can be achieved solely through an integrated nutrition system. Kazemalilou et al. (2023) also reported that integrated application of phosphorus and sewage sludge improved sunflower oil yield and quality.

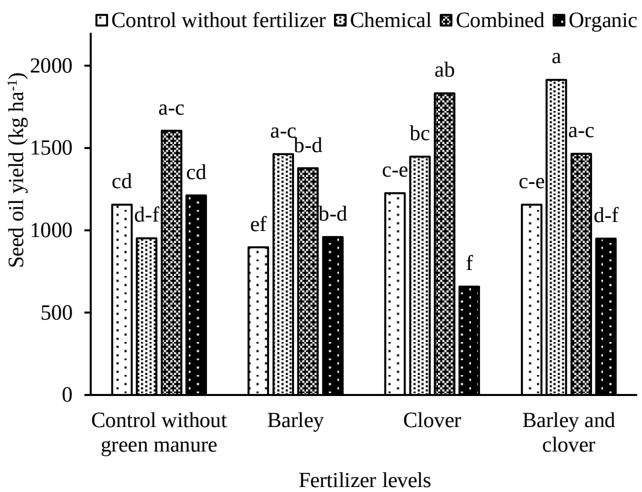

Figure 6. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the number of oil yield of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

3.6. Seed protein percentage and yield

The main effects of nutrition and green manure on protein percentage were not significant, but their interaction was significant (Table 4). The highest protein percentage (14.45%) was observed in the control without green manure and nutrition, while the lowest (10.97%) was recorded for barley green manure under the integrated system (Fig. 7). In the control treatment, limited vegetative growth results in nitrogen being distributed within a smaller dry matter volume, increasing protein percentage; however, due to lower seed yield, protein yield per unit area decreased (Fig. 8).

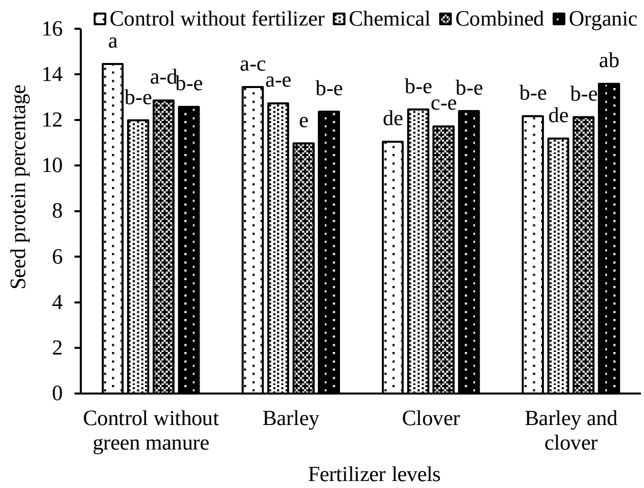

Figure 7. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the number of Protein percentage of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

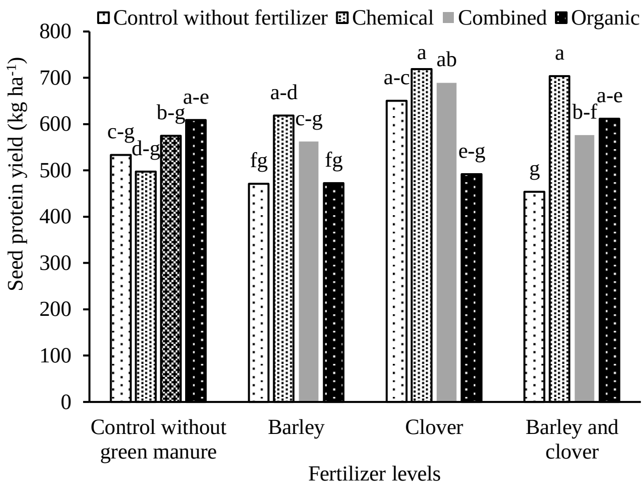

Figure 8. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the seed protein yield of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

Previous studies indicate that nitrogen concentration in plant tissues decreases with increasing dry matter, even under luxury nitrogen consumption (Siadat and Derakhshan, 2022). The lower protein percentage in barley green manure with the integrated system, compared to barley without green manure, is due to increased seed yield and the dilution of absorbed nitrogen across a larger number of seeds, reducing protein percentage per seed. The effects of green manure, nutrition system, and their interaction on protein yield per unit area were significant (Table 3).

The highest protein yield (718.84 kg ha^{-1}) was achieved with clover green manure under the chemical system, while the lowest (453.59 kg ha^{-1}) was obtained with mixed barley and clover green manure under the no-fertilizer control (Fig. 8). The mixed barley-clover green manure with chemical nutrition also ranked high statistically. These results indicate that the presence of clover as a nitrogen-fixing legume enhances seed protein, and supplementing mineral elements to clover or mixed barley-clover green manure increases protein yield. This finding aligns with the report by Gerami et al. (2013), who found that legume green manure (mung bean and cowpea) combined with 50 kg nitrogen resulted in the best protein yield compared to solely chemical or unfertilized systems.

3.7. Leaf and stem ash percentage

The effects of the nutrition system and the interaction between green manure and nutrition system on leaf and stem ash percentage were significant, but the simple effect of green manure was not significant (Table 4). The highest ash percentage (21.02%) was observed in the clover green manure under the organic nutrition system (Fig. 9).

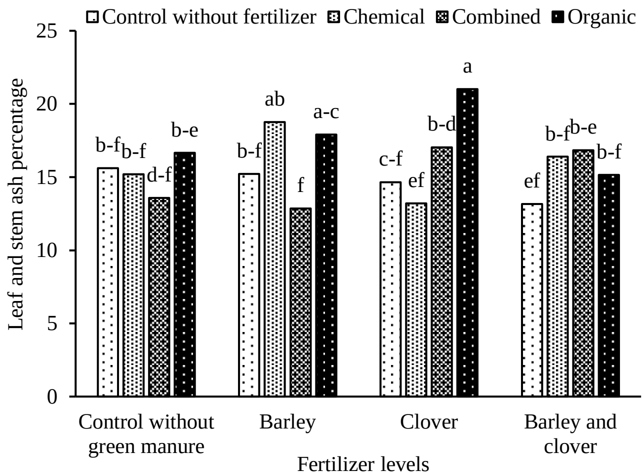

Figure 9. Comparison of the mean interaction effect of green manure crops and different nutrition levels on the leaf and stem ash percentage of sunflower plants (Similar letters indicate no significant difference based on the LSD test).

Improved nutritional status and soil fertility in the organic system, likely due to gradual nutrient release, improved soil structure, enhanced biological activity, and better mineral uptake, contributed to higher ash content. Barley treatments under chemical and organic nutrition systems also ranked in the superior statistical groups. Higher ash percentage indicates greater mineral richness in aerial plant parts. Adekiya et al. (2020) reported that sunflower green manure increased okra fruit ash percentage from 5.6% to 6.9% in 2017 and from 5.1% to 6.5% in 2018. In the study Aboyeji (2019) on radish root, the application of sole and mixed green manure derived from the leaves of (*Vernonia amygdalina* L.) and (*Chromolaena odorata* L.) produced statistically similar ash content values compared to the control treatment without green manure. Alzamel et al. (2022) reported no significant difference in ash content of sunflower seeds among different organic and mineral fertilizer treatments. These results indicate that the effects of green manure and nutrition systems are not absolute or consistent but depend on management practices, plant species, fertilizer type, soil conditions, and climate.

3.8. Correlation results of seed yield with other traits

According to Table 5, seed yield showed a very strong, significant positive correlation with the number of seeds per head (r=0.83**). The number of seeds per head is one of the main components of seed yield, and increasing it directly enhances yield. This represents the strongest positive correlation with seed yield. Seed yield also exhibited a strong, significant positive correlation with 1000-seed weight (r=0.67**). Larger seeds have greater weight and directly increase seed yield. Seed yield showed a moderate, significant negative correlation with the percentage of empty seeds (r=-0.37**). An increase in the percentage of empty seeds reduces the number of filled seeds, consequently decreasing seed yield. Seed yield demonstrated a negligible and non-significant correlation with oil percentage (r=0.05^{ns}). Oil percentage is a trait independent of seed yield, and an increase in seed yield does not affect it. Seed yield revealed a strong, significant positive correlation with oil yield (r=0.7**). Oil yield is derived from the product of seed yield multiplied by oil percentage. Given this strong correlation, an increase in seed yield directly enhances oil yield. Seed yield showed a moderate to strong, significant negative correlation with protein percentage(r=-0.4**). Due to the dilution effect, an increase in seed yield (primarily through carbohydrate storage) leads to a relative decrease in protein percentage. Seed yield exhibited a very strong, significant positive correlation with protein yield (r=0.82**). Despite the reduction in protein percentage, the substantial increase in seed yield results in a remarkable rise in the total protein produced per hectare. Seed yield had an almost zero, non-significant correlation with leaf and stem ash content (r=-0.01^{ns}), indicating that ash in vegetative tissues has no direct relationship with seed yield.

Table 5. Correlation matrix between the studied traits

Traits	NSH	TSW	ES	SY	SO	SOY	SP	SPY
TSW	0.31*							
ES	-0.15^{ns}	-0.41**						
SY	0.83**	0.67**	-0.37**					
SO	0.01^{ns}	0.21**	0.03^{ns}	0.05^{ns}				
SOY	0.56**	0.58**	-0.23^{ns}	0.70**	0.73**			
SP	-0.41**	-0.20^{ns}	0.17^{ns}	-0.40**	-0.06^{ns}	-0.32*		
SPY	0.63**	0.61**	-0.29*	0.82**	0.01^{ns}	0.55**	0.17^{ns}	
LSA	0.02^{ns}	0.01^{ns}	0.07^{ns}	-0.01^{ns}	-0.11^{ns}	-0.04^{ns}	0.06^{ns}	0.32^{ns}

NSH: Number of seeds per head; TSW: 1000 seed weight; ES: Empty seed percentage; SY: Seed yield; SO: Seed oil percentage; SOY: Seed oil yield; SP: Seed protein percentage; SPY: Seed Protein yield; LSA: Leaf and stem ash percentage. ns, * and ** indicate non-significant and significant at the 5% and 1% probability levels, respectively.

4. Conclusion

This experiment demonstrated that the choice of cropping system and fertilizer type significantly affects yield and product quality, with outcomes depending on the cultivation objective. In general, for quantitative increase, selecting a barley-clover mixture combined with chemical fertilizer resulted in the highest seed, oil, and protein yields, along with an acceptable ash percentage. However, for sustainable agriculture aimed at improving soil fertility while achieving high oil and protein yields, cultivating clover with an integrated nutrition system is recommended.

Conflict of interests

All authors declare no conflict of interest.

Ethics approval and consent to participate

No humans or animals were used in the present research. The authors have adhered to ethical standards, including avoiding plagiarism, data fabrication, and double publication.

Consent for publications

All authors read and approved the final manuscript for publication.

Availability of data and material

All the data are embedded in the manuscript.

Authors' contributions

Idea and design of the topic: Dabaghzadeh M., Fateh E. and Aynehband A.; Proposal writing and editing: Dabaghzadeh M. and Fateh E.; Executive section: Dabaghzadeh M. and Fateh E.; Statistical analysis: Dabaghzadeh M. and Zare A.; Article writing and review: Dabaghzadeh M., Fateh E., and Zare A.; Material section: Dabaghzadeh M., Fateh E. and Monsefi A.

Informed consent

The authors declare not to use any patients in this research.

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