Increasing the Feed Values of Barley, Vetch, and Safflower Mixtures in Hydroponic Fodder Systems

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Abstract

Cool-climate grains have been germinated under controlled climatic conditions and used in feeding animals for many years. This study was conducted to determine the effect of adding different amounts of vetch and safflower seeds to barley seed in fresh green feed machines and administering liquid fertilizer (LF) through irrigation water on feed values. Mixtures were prepared by adding vetch and safflower to barley at ratios of 10, 20, and 30%.

Dry matter (DM) losses were observed in all mixtures. The use of LF reduced the loss of crude protein (CP) in 100% barley and the 70% barley + 30% vetch mixture. However, the CP yield increased in 90% barley + 10% vetch mixture. A 10% increase was observed in ether extract (EE) yield in 80% barley + 20% vetch mixture with LF application. An increase of 60.3–143.7% in acid detergent fiber values was observed in all mixtures. Neutral detergent fiber yields decreased by 17.8–45.9%, and ash loss was observed to be higher in all mixtures compared to that in seeds. All mixtures exhibited an increase in total carotene, except for the mixtures of 90% barley + 10% vetch and 70% barley + 30% vetch with LF. An increase in vitamin E values was observed in 100% barley group with or without LF, and in 90% barley+10% vetch and 90% barley + 10% safflower groups compared to those in the seeds.

The addition of up to 30% vetch to the barley and the use of LF were more effective in terms of reducing nutrient losses than other mixtures. In addition, we concluded that the use of LF had a positive effect and that the feed values of the seeds could be increased using this production system.

1. Introduction

Seeds have been germinated and used as animal feed since 1939. Since the early 1970s, there has been feed production systems with up to 400 different seed-germination methods, and those using these methods have raised the question of whether this type of production is economical. Germination is a complex metabolic event involving the breakdown of structural proteins, oxidation of lipids, and conversion of carbohydrates into simple sugars to provide the energy and the essential components needed for the growth and development of the seed or grain (Sneath and Melntosh, 2003).

In hydroponic systems, the seeds to be germinated are kept in water until they are completely saturated. Complex biochemical changes occur during hydration and sprouting. Chemical components, such as proteins, starches, and lipids, are used to make new compounds through enzymatic reactions and transferred to other parts of the plant. The seeds sprout within 24 h and green parts form on the second or third day. Although the plants can be harvested on the fifth day, the seventh day is the most suitable for harvest because plant growth decreases thereafter (Kafkafı, 1988; Chavan and Kadam 1989; Cuddeford, 1989).

The temperature of the holding water, holding time, light levels and duration, irrigation techniques and frequency, LF support, and effect of seed-sowing rates on dry matter (DM) were examined, and our results suggested that an increase in DM would not be possible over a short-term (5–8 d) growth cycle. In different studies, DM losses ranging from 25 to 94% have been reported within 5- to 7-d germination processes (Peer and Leeson, 1985; Chung et al., 1989; Chavan and Kadam, 1989; Cuddeford, 1989); however, Sneath and Melntosh (2003) have reported that no changes in DM were observed over a 6-d germination process. The studies showed that more than 90% of DM can be recovered using the appropriate methods and under the appropriate conditions and, in terms of feed value, quality-based rather than cost-based production must be considered.

Hydrolytic enzyme activities, total proteins, fat content, some essential amino acids, total sugars, and group B–vitamin values increase whereas DM, starch, and antinutrient levels decrease during grain-seed germination. The increases in protein, fat, fiber, and total ash is correlated with reduced starch; however, amino acid composition, group-B vitamins, sugars, protein and starch mobilization, phytate, and protease enzyme inhibitors are the result of metabolic reactions during the germination process. The importance of these changes as they relate to animal nutrition is worth examining (Peer and Leeson, 1985; Chavan and Kadam, 1989; Sneath and McIntosh, 2003; Tan-Wilson and Wilson, 2012; Sharif et al., 2013).

Unless nutrients are externally supplied to the seeds, germinating seeds consume only water and oxygen. Under optimum moisture, oxygen, and temperature conditions, sugars are used as a source of energy for cell wall synthesis and growth. Grain loses carbon through respiration during its growth period. In addition, because photosynthesis during the germination process is not very efficient, carbon accumulation is very low. The positive nutritional changes during the germination process are mainly a result of the conversion of complex compounds into simple forms, and the breakdown of undesirable antinutritional substances. There is no actual increase in the majority of nutrients; rather, their higher values are the result of a loss of DM in the form of carbohydrates in combination with respiration during germination and the decrease in total carbohydrates (MacLeod and White, 1960; Chavan and Kadam, 1989; Cuddeford, 1989; Kubicka et al., 2000; Tan-Wilson and Wilson, 2012; Tonguç et al., 2012; Akbağ et al., 2014).

The amount of energy decreases as the germination time increases because of an increase in fiber and a decrease in starch. Although it was reported that more energy was lost after 4 d, the nutritional yields had better performance than dry grass (Peer and Leeson, 1985; Sneath and McIntosh, 2003).

In the production of commercial green feed, cool cereal grains, especially barley, are used. No studies have been found in the literature on yield production using legumes alone or in mixtures; however, in studies on the germination physiology of legumes, the researchers emphasized that this germination produced shoots that were richer in phenolic compounds with antioxidant and antimicrobial properties, and that the total phenolic, flavonoid, and antioxidant activities significantly increased. It has been reported that inhibition of trypsin increased during germination of cowpea grains and reached a maximum on the fifth day, the total content of soluble sugars increased, and the vitamin C content increased. It has also been reported that the taste of the germinated cowpeas was positively affected, and that the nutritional quality and bioavailability of carbohydrates and proteins increased. In addition, it was reported that polyphenols decreased after germination, the oligosaccharide content decreased, and the phytic acid content decreased as a result of cowpea germination (Özkaynak, Kanmaz and Ova, 2014);

One of the most important forage crops that can be grown in field agriculture is vetch, which is a protein-rich legume. Green and dry grass yield has an important place in forage crops due to its value in animal nutrition. The creeping habitus of vetch species, which is a valuable forage plant, is a negative factor in roughage production. However, this negativity is eliminated by growing them in a mixture with upright habitus grassy forage crops such as barley. Safflower is an oil seed plant that can grow in barren and arid lands contains approximately 20-40% oil and 10-20% crude protein. Its unsaturated fatty acids ratio in total fatty acids is around %90-93. Safflower is an important alternative feed resource that can be used as both roughage and concentrated feed because of its seeds and oil. Besides, the Safflower have also the importance due to its rich unsaturated fatty acid content that is used in the synthesising of Conjugated Linoleic Acid (Schmid et al, 2006; Ingale and Shrivastava, 2011)

Therefore, the main purpose of this study is to investigate the effects of mixtures consisting of protein-rich legume vetch and energy-rich wheatgrass barley grains on feed value with hydrophonic fodder system.

2. Materials And Methods

This study was conducted in Van, Turkey, on 38.50 x 43.36 coordinates. Barley, vetch, and safflower were used in this study. Before the study began, it was understood that the structure of vetch and safflower roots was looser than that of the barley, and that distribution and harvesting were very difficult because their roots did not cling to each other; therefore, the changes in the nutrients of the fresh green feed obtained by mixing vetch and safflower seeds in different ratios were examined.

The mixtures used were as follows: 100% barley, 90% barley + 10% vetch, 80% barley + 20% vetch, 70% barley + 30% vetch, 90% barley + 10% safflower, 80% barley + 20% safflower, and 70% barley + 30% safflower.

Preparation of the hydroponic system

The hydroponic system for growing livestock feed (Ekohasıl 98: Ecological Livestock Systems) was used in this study. Before adding seed, the irrigation system, trays where seeds would be sowed and 14 rubber buckets in which the seeds would be kept in water were washed with water and detergent and disinfected with 10% formaldehyde. Before the seeds were added, the machine was set to 21ºC, irrigated for 90 sec every 2 h, received 19 h of light, and allowed to run for 2 d. To prevent mold growth during the study, 50 mL bleach was added to the irrigation water daily.

Sowing and sampling

Three-section trays measuring 1.4 m^2 were prepared using seven plates for each mixture and one rubber 20-L bucket for each tray. Before sowing, the seed mixture was weighed in the rubber buckets using a 1 g–sensitive balance to ensure that 3.3 kg mixture was used, after which 15 L water was added to the bucket, and the mixture was let stand for 24 h. After 24 h, the straw-hay and seeds that had accumulated on the surface of the water were filtered using using 1mm mesh screen. The filtered residues were collected for each mixture and dried to determine washing losses, and a ~1 kg sample was taken for each mixture for analysis. The seeds in the rubber buckets were planted in the trays and allowed to incubate for 7 d.

Harvest and sampling

At the end of the seventh day, the fresh green feed was removed from the trays and spread out for 3 h to allow the water to drain. Samples weighing 750–800 g were collected from three different points in the fresh green feed and kept in a drying room for 3 d to pre-dry, after which they were kept at 80ºC for 48 h to complete the drying process.

Application of LF

The LF comprised 8% nitrogen (N), 4% water-soluble phosphorpentaoxide, and 3% water-soluble potassium oxide. The amount of LF used was twice the amount recommended for field use calculated as follows:

Recommended ratio: 1 L LF for 4000 m^2 field

Calculated for hydroponic system: 20 m^2 area x 7 layers = 140 m^2 area at 35 ml LF for 7 days = 5 ml

For doubling this value = 10 mL was calculated as the daily amount.

Chemical analyses

DM, crude protein (CP), and ash in the feeds were analyzed according to the Association of Official Agricultural Chemists International (1991) standards. Crude oil (CO) was analyzed according to the American Oil Chemists' Society (2004), and acid detergent fiber (ADF) and neutral detergent fiber (NDF) were analyzed according to Van Soest et al. (1991). Total carotene and vitamin E were analyzed using high-performance liquid chromatography with the lutein (DSM Nutrition, Heerlen, The Netherlands) standard at a flow rate of 1.05 mL/min with the Spherisorb ODS type, 3-tip C18 column, and methanol-water (97:3, v/v) mobile phase (Surai and et al., 1996; Surai and Speake, 1998a; b; Karadaş et al., 2005; 2011).

Statistical analyses

The study was conducted with seven replications based on the fully randomized experimental design. Statistical analyses were performed using SAS 9.4.1 (https://www.sas.com/en_us/software/sas9.html). The differences between the seven factors with or without LF were tested using one-way analysis of variance (ANOVA). Duncan's multiple range test was used to determine the differences among the factors found to be significantly different after conducting the ANOVA analyses using the following equation:

$$, y_{ij} = \mu + a_i + e_{ij} ,$$

where y_{ij} = dependent variable; (I = 1,2,...,7), (j = 1,2,...,7); μ = overall mean; a_i = the fixed effect of groups; and e_{ij} = random error.

3. Results

In this study, 90% barley + 10% vetch with LF ($90B10V_{lf}$) produced the lowest green-grass yield; however, it also produced the highest DM yield ($P \le 0.05$). Green grass yield increased in the groups containing 90% barley + 10% vetch (90B10V) and 80% barley + 20% vetch (80B20V) with no LF ($P \le 0.05$) (Table 1) and in the groups containing 80% barley + 20% safflower with fertilizer ($80B20S_{lf}$) ($P \le 0.05$) (Table 2).

Table 1
Changes in nutrient content in fresh green feeds with no liquid fertilizer (g).

	100B		**90B10V**		**80B20V**		**70B30V**		**90B10S**		**80B20S**		**70B30S**	
	Seed	**Sprout**	**Seed**	**Sprout**	**Seed**	**Sprout**	**Seed**	**Sprout**	**Seed**	**Sprout**	**Seed**	**Sprout**	**Seed**	**Spr**
Green grass	3300	17554.9 ±439ac	3300	18265.6 ±273.1 a	3300	18230.1 ±164.9 a	3300	16939.9 ±285.5bc	3300	17769.0 ±256.3 ab	3300	17135.1 ±266.9 bc	3300	167 ±18 c
DM	3061.5	1771.8 ±41.9c	3087.3	1754.3 ±57.2 c	3078.3	2067.3 ±135.5 ab	3106.7	2232.5 ±80.7 a	3064.6	1814.6 ±66.6 bc	3086.3	2183.1 ±124.4 a	3093.8	205 ±85 ab
CP	336.8	290.6 ±11.8 c	401.4	358.6 ±9.4 b	443.1	436.2 ±30.8 a	492.6	483.2 ±28.4 a	337.2	333.6 ±11.0 c	346.2	375.6 ±21.7 b	366.0	377 ±16
EE	214.9	96.2 ±23.0 c	169.6	127.6 ±14.2 c	165.1	136.5 ±69.34 c	187.3	180.3 ±39.1 bc	238.4	156.2 ±28.7 bc	326.4	233.4 ±19.5 ab	397.4	289 ±35
ADF	268.7	618.7 ±42.1 c	275.9	664.5 ±41.9 bc	348.4	756.5 ±29.6 b	411.1	696.1 ±20.05 bc	407.3	747.0 ±23.4 b	478.6	942.9 ±55.6 a	471.5	925 ±42
NDF	1730.3	1164.8 ±45.4	1882.9	1130.6 ±75.3	2062.0	1217.6 ±83.9	1940.1	1239.4 ±54.6	2032.4	1099.4 ±44.1	1477.9	1143.9 ±79.7	1643.0	125 ±41
ASH	68.8	67.6 ±2.8 c	79.0	69.5 ±3.07 c	87.1	80.2 ±4.0 b	99.5	90.4 ±3.0 a	73.2	67.3 ±1.7 c	79.6	75.0 ±2.5 bc	74.8	75. ±3.
Total Car*	6.9	11.8 ±1.0 c	11.5	11.3 ±1.5 c	14.3	16.3 ±5.7 bc	17.3	23.4 ±2.6 a	6.3	13.7 ±1.4 c	6.5	19.9 ±1.4 ab	6.1	21. ±2.
Vit E*	65.8	74.0 ±5.7 d	96.9	75.7 ±6.8 d	130.5	109.5 ±11.2 bc	156.5	140.1 ±10.1 ab	98.9	86.3 ±8.8 cd	138.3	127.3 ±9.7 ab	185.1	144 ±16

B: barley, V: vetch, S: safflower, DM: dry matter, CP: crude protein, EE: ether extract, ADF: acid detergent fiber, NDF: neutral detergent fiber; Car: carotene, Vit: vitamin.

*mg/kg

Numbers before abbreviated mixtures represent percentages.

$^{a, b, c, d, e}$Values with the different letters in the same row differ significantly ($P < 0.05$).

Table 2
Changes in nutrient content in fresh green feeds with liquid fertilizer (g).

	$100B_{lf}$		$90B10V_{lf}$		$80B20V_{lf}$		$70B30V_{lf}$		$90B10S_{lf}$		$80B20S_{lf}$		70
	Seed	Sprout	Seed	Sprout	Seed	Sprout	Seed	Sprout	Seed	Sprout	Seed	Sprout	
Green grass	3300	17473.9 ±255.3 bc	3300	16940.6 ±275.7 c	3300	17462.4 ±294.7 bc	3300	17943.4 ±210.9 ab	3300	18101.1 ±336.3 ab	3300	18505.0 ±278.1 a	33
DM	3061.5	2107.4 ±142.9 b	3087.3	2441.1 ±116.2 a	3078.3	2274.7 ±25.1 ab	3106.7	2217.9 ±47.0 ab	3064.6	2084.4 ±103.7 b	3086.3	2161.2 ±137.4 ab	30
CP	336.8	332.1 ±22.0 c	401.3	405.7 ±13.5 b	443.1	432.9 ±5.8 b	516.9	493.9 ±7.5 a	337.0	322.8 ±9.2 c	346.2	352.6 ±20.6 c	36
EE	214.9	166.3 ±22.4 b	169.6	155.5 ±23.9 b	165.1	181.5 ±25.0 ab	167.1	149.3 ±36.5 b	238.4	190.5 ±21.5 ab	326.4	214.4 ±30.2 ab	39
ADF	268.1	653.2 ±86.6 b	275.9	636.8 ±29.7 b	348.4	636.4 ±39.0 b	399.9	640.9 ±29.1 b	407.3	776.7 ±34.8 ab	478.6	883.8 ±40.0 a	47
NDF	1730.3	1226.7 ±117.0	1882.9	1405.7 ±112.7	2062.0	1300.4 ±39.9	2017.1	1344.7 ±85.7	2032.4	1261.3 ±33.6	1477.9	1215.1 ±75.6	16
ASH	68.8	65.8 ±2.1 d	79.0	78.1 ±3.6 bc	87.1	82.4 ±3.5 ab	102.8	87.5 ±2.2 a	73.1	71.4 ±3.0 cd	79.6	71.9 ±3.3 cd	74
Total Car*	7.0	20.4 ±2.6	11.5	20.1 ±1.8	14.3	16.3 ±3.0	19.1	15.8 ±1.8	6.3	13.6 ±2.0	6.5	18.4 ±2.1	6.
Vit E*	65.8	100.4 ±8.4	96.9	104.0 ±9.1	130.5	100.3 ±12.1	159.5	106.7 ±5.3	98.9	99.0 ±15.6	138.3	109.3 ±7.5	18

Notes: lf, liquid fertilizer; DM, dry matter; CP, crude protein; ADF, acid detergent fiber; NDF, neutral detergent fiber; Car, carbohydrate, Vit, vitamin.

[a,b,c,d,e]Values with the different letters in the same row differ significantly (P < 0.05).

*mg/kg

DM losses were observed in all mixtures and applications. Compared to the seeds, the lowest losses at 20.9% were from the $90B10V_{lf}$ mixture; whereas, the highest losses were from the 90B10V mixture (43.2%) (P ≤ 0.05) (Tables 1). LF increased DM efficiency in all mixtures except 70% barley + 30% safflower ($770B30S_{lf}$) (P ≤ 0.05) (Table 2).

The seeds increased CP by 1.1% in the $90B10V_{lf}$, 8.5% in the 80B20S, 1.8% in the $80B20S_{lf}$, and 3.1% in the 70BA30S mixtures (Table 4). The use of LF reduced CP loss in $100B_{lf}$ and in the $70B30V_{lf}$ mixture, and resulted in CP yields higher than that in seed in the $90B10V_{lf}$ mixture (P ≤ 0.05) (Table 2).

In the sprouts, only the $80B20V_{lf}$ mixture increased ether extract (EE) by 10% (Table 4); a decrease was observed in all other samples. EE losses were between 3.7 and 55.2% in mixtures without LF and between 8.3 and 34.4% in mixtures with LF. The $70B30V_{lf}$ group yielded the highest EE values (P ≤ 0.05).

We observed a 60.3–143.7% increase in ADF in all mixtures compared to that in seeds (Tables 3 and 4). ADF yields especially increased with an increase in the proportion of safflower in the mixtures. In this study, the highest ADF yields were obtained in the 80B20S, 70B30S, $80B20S_{lf}$, and $70B30S_{lf}$ groups (P ≤ 0.05) (Tables 1 and 2). Higher levels of ADF production were also obtained with the use of LF.

Table 3
Differences in nutrients in fresh green feeds with or without liquid fertilizers (g).

	LF	100B	90B10V	80B20V	70B30V	90B10S	80B20S	70B30S
Green grass	N	17555 **AC**	18266**a A**	18230**a A**	16940**bBC**	17769 **AB**	17135**b BC**	16791**b C**
	Y	17474 **BC**	16941**bC**	17462**bBC**	17943**aAB**	18101 **AB**	18505**a A**	18166**a AB**
DM	N	1771.8**b C**	1754.3**bC**	2067.3**AB**	2232.5**A**	1814.6bB**C**	2183.1 **A**	2057.4 **AB**
	Y	2107.4**a B**	2441.1**aA**	2274.7**AB**	2217.9**AB**	2084.4a **B**	2161.2 **AB**	1963.1 **B**
CP	N	290.6 **C**	358.6**bB**	436.0 **A**	483.2 **A**	333.6 **C**	375.6 **B**	377.5 **B**
	Y	332.2 **C**	405.7**aB**	432.9 **B**	493.9 **A**	322.8 **C**	352.6 **C**	351.1 **C**
EE	N	96.2**b C**	127.6 **C**	136.5 **C**	180.3 **BC**	156.0 **BC**	233.4 **AB**	289.6 **A**
	Y	166.3**a B**	155.5 **B**	181.5 **AB**	149.3 **B**	190.5 **AB**	214.4 **AB**	260.5 **A**
ADF	N	618.7 **C**	664.5 **BC**	756.5**aB**	696.1 **BC**	747.0 **B**	942.9 **A**	925.0 **A**
	Y	653.2 **B**	636.8 **B**	636.4**bAB**	640.9 **B**	776.7 **AB**	883.8 **AB**	859.8 **A**
NDF	N	1164.8	1130.6	1217.6	1239.4	1099.4**b**	1143.9	1253.7
	Y	1226.7	1405.7	1300.4	1344.7	1261.3**a**	1215.2	1152.2
Ash	N	67.4 **C**	69.5 **C**	80.2 **BC**	90.4 **A**	67.3 **C**	75.0 **BC**	75.6 **BC**
	Y	65.8 **D**	78.1 **BC**	82.4 **AB**	87.5 **A**	71.4 **CD**	71.9 **CD**	73.9 **BCD**
Total Car*	N	11.8**b C**	11.3**b C**	16.3 **BC**	23.4**a A**	13.7 **C**	19.9 **AB**	21.3 **AB**
	Y	20.4**a**	20.1**a**	16.3	15.8**b**	13.6	18.4	20.4
Vit E *	N	74.0 **D**	75.7**b D**	109.5 **BC**	140.1**aAB**	86.3 **CD**	127.3 **AB**	144.3 **A**
	Y	100.4**A**	104.0**a**	100.3	106.7**b**	99.0	109.3	127.2

Notes: LF, liquid fertilizer; DM, dry matter; CP, crude protein; ADF, acid detergent fiber; NDF, neutral detergent fiber; Car, carbohydrate, Vit, vitamin; N, unused liquid fertilizer; Y, used liquid fertilizer.

*mg/kg.
a, b, c, d: Values with these different letters in the same column differ significantly (P < 0.05).

A, B, C, D: Values with these different letters in the same row differ significantly (P < 0.05).

Table 4
Green-grass and nutrient changes compared to seeds in fresh green feeds with or without liquid fertilizer (%).

	100B		90B10V		80B20V		70B30V		90B10S		80B20S		70B30S	
LF	**N**	**Y**	**N**	**Y**	**N**	**Y**	**N**	**Y**	**N**	**Y**	**N**	**Y**	**N**	**Y**
Green grass	432.0 ↑	429.5 ↑	453.5 ↑	413.3 ↑	452.4 ↑	429.2 ↑	413.3 ↑	429.1 ↑	438.4 ↑	448.5 ↑	419.2 ↑	429.2 ↑	408.8 ↑	429.2 ↑
DM	42.1 ↓	31.2 ↓	43.2 ↓	20.9 ↓	32.8 ↓	26.1 ↓	28.1 ↓	28.6 ↓	40.8 ↓	32.0 ↓	29.3 ↓	30.0 ↓	33.5 ↓	36.5 ↓
CP	13.7 ↓	1.3 ↓	10.6 ↓	1.1 ↑	1.6 ↓	2.3 ↓	1.9 ↓	4.4 ↓	1.0 ↓	4.2 ↓	8.5 ↑	1.8 ↑	3.1 ↑	4.0 ↓
EE	55.2 ↓	22.6 ↓	24.71 ↓	8.3 ↓	17.3 ↓	10.0 ↑	3.7 ↓	10.6 ↓	34.5 ↓	20.1 ↓	28.5 ↓	34.3 ↓	27.1 ↓	34.4 ↓
ADF	130.8 ↑	143.7 ↑	140.8 ↑	130.8 ↑	117.1 ↑	82.7 ↑	69.3 ↑	60.3 ↑	83.4 ↑	90.7 ↑	97.0 ↑	84.6 ↑	96.2 ↑	82.3 ↑
NDF	32.7 ↓	29.1 ↓	39.9 v	25.3 ↓	40.9 ↓	36.9 ↓	36.1 ↓	33.3 ↓	45.9 ↓	37.9 ↓	22.6 ↓	17.8 ↓	23.7 ↓	29.9 ↓
ASH	1.6 ↓	4.4 ↓	12.0 ↓	1.1 ↓	7.8 ↓	5.3 ↓	9.2 ↓	14.9 ↓	7.9 ↓	2.3 ↓	5.7 ↓	9.7 ↓	1.1 ↓	1.1 ↓
Total Car*	70.6 ↑	194.2 ↑	1.6 ↓	75.4 ↑	14.5 ↑	14.1 ↑	35.6 ↑	16.9 ↓	116.4 ↑	115.8 ↑	205.4 ↑	181.6 ↑	249.8 ↑	233.6 ↑
Vit E*	12.5 ↑	52.6 ↑	21.9 ↓	7.3 ↑	16.0 ↓	23.1 ↓	10.4 ↓	33.1 ↓	12.9 ↓	0.1 ↑	7.9 ↓	30.0 ↓	22.0 ↓	31.3 ↓

DM, dry matter; CP, crude protein; ADF, acid detergent fiber; NDF, neutral detergent fiber; Car, carbohydrate, Vit, vitamin. LF, liquid fertilizer;

N, unused liquid fertilizer; Y, used liquid fertilizer.

*mg/kg.

↑: increase

↓: decrease

The results also showed a decrease in NDF of 17.8–45.9% in all mixtures (Table 4). The differences among the mixtures in terms of NDF yields were not statistically significant (Tables 1 and 2).

A decrease in crude ash value was observed in the mixtures compared to that in the seeds in all samples except for 70B30S (Tables 1, 2, and 3). As the amount of vetch and safflower in the seeds increased, the ash value increased by up to 20%. The highest ash loss was observed in 70B30V$_{lf}$ at 14.9%. Crude ash loss increased in the 100B$_{lf}$, 70B30V$_{lf}$, and 80B20S$_{lf}$ groups (Table 4). The highest ash yield was observed in the 70B30V and 70B30V$_{lf}$ groups ($P \leq 0.05$) (Tables 1 and 2).

Total carotene contents increased in all mixtures except for the 90B10V and 70B30V$_{lf}$ groups (Table 4). Fertilization had an effect on the increase in carotene in the 90B10V$_{lf}$ and 80A20F$_{lf}$ groups. Total carotene production reached the highest value in the 70B30V group ($P \leq 0.05$) (Table 1); the difference in the yield among the groups with LF was not significant (Table 2).

Vitamin E values increased in the 100B, 100B$_{lf}$, 90B10Vlf, and 90B10S$_{lf}$ groups compared to that in seeds ($P \leq 0.05$) (Tables 1and 2); whereas, these values decreased in all other mixtures.

4. Discussion

Using 3.3 kg of each of the different mixtures produced between 18.50 and 16.94 kg green grass, which corresponded to 5.6–5.13 kg green grass from 1 kg mixture (Tables 1 and 2). The DM contents ranged from 10.8 to 14.40%. In the studies, between 6 and 10 kg green grass was produced from 1 kg grain containing 6.4 to 20.0% DM. The results of Dung et al. (2010) showed a yield of 4.7 kg green grass with 19.7% DM; those of Peer and Leeson (1985) showed a yield of 5.7 kg green grass from barley. Many factors, such as irrigation water quantity and quality, pH, seed preparation, seed quality and type, sowing density, ambient temperature, hygiene status, plant nutrients, and light, affect DM production. It has been reported that as a result of the use of energy reserves, oxidation, nutrient exchange, and nutrient synthesis, any increase in DM would not be possible during the 7-d germination process and DM losses would be between 7 and 64%. (Lewis and Chadwick, 1983; Chavan and Kadam, 1989; Sneath and Melntosh, 2003; Karaşahin, 2014).

One study has found that adding N-containing LF to the irrigation water of hydroponically produced barley fresh green feed had a positive effect on the accumulation of total N and organic N in all plants and in different plant components (Lewis and Chadwick, 1983). During germination, an increase in proteins is proportional to the loss of DM through respiration. Previous studies have shown that, from the fourth day of germination, the absorption of minerals and nitrates from the roots increased and the synthesis of N compounds from the seed carbohydrate reserves increased, which led to an increase in CP levels. The increase in proteins through DM loss was ~24% from day 4 to day 8 and was proportional to the loss of DM, and that one-half of this increase was a result of protein synthesis from the N absorbed from irrigation water. In addition, it has been reported that there were CP losses of 7–24% after the eight day, and that CP content could be approximated to the seed after the second day of germination (Sneath and Melntosh, 2003; Chung et al., 1989). Previous studies have reported that there was a 3% average increase in DM in 7-d fresh green feed compared to that in barley seeds (Peer and Leeson, 1985; Dung et al., 2010;

Shaarif et al., 2013). The results of the study by Karaşahin (2014) showed 16.5–19.9% CP in grass 7 d after germination with different applications compared to that in barley seeds with 13.5% CP; however, in terms of total yield, these results reported CP losses from 4.7 to 21.58%. During 7 d of germination, proteins were metabolized by enzymes, yielding an increase in water-soluble proteins and amino acids (Nielsen et al., 1977; Pathirana et al., 1983; Chavan and Kadam, 1989). This conversion to albumin and globulins increases protein quality; however, in terms of ruminant feeding, whether within the structure of proteins, the total amount of N is more important than this increase because the microorganisms in the rumen break down the majority of high-quality proteins into ammonia.

With the addition of safflower seeds and their high oil content, substantial fat loss was observed during germination. During plant germination, oils re-metabolize into hydrolytic enzymes and essential fatty acids. Carbon, hydrogen, and oxygen molecules released as a result of the breakdown of fats are used in the synthesis of nutrients that are required for the formation of different parts of the plant. It has been reported that linoleic acid and oleic acid decrease; linolenic, palmitic, and stearic acids increase; and total lipid values decrease compared to those in seeds, although there was a rapid decrease until the fourth day of germination and a rapid increase from the fourth to the seventh days. In a study conducted on two safflower varieties, it was reported that fat content decreased from 58 to 45% in the first 72 h of germination (MecLeod et al., 1962; Peer and Leeson 1985; Kubicka et al., 2000; Öztürk et al., 2012; Tonguç et al., 2012). In hydroponics systems, it is believed that the seeds with a high amount of fat, such as safflower seeds, will experience fat losses because of the amount of irrigation water and the frequency of irrigation during germination.

Different studies have reported that ADF value increased between 214.3 and 402.8% in barley fresh green feeds produced using different systems (Cuddeford, 1989; Shaarif, 2013). In particular, it has been reported that the starch contained in the seed is metabolized by enzymes and used in the synthesis of structural carbohydrates, such as cellulose and hemicellulose, thus increasing the amount of ADF with germination. The use of fats in the structure of the seeds and cell wall material explains the increase in ADF yield when the ratio of safflower is increased in the mixtures (Table 4).

Sharif et al. (2013) reported that the results of several studies indicated that the NDF values in fresh green feeds were 57.4–138.6% lower than those in the seeds. The NDF values of the products increased when the vetch and safflower ratio in the mixtures with no LF increased (Table 1). In the mixture groups with LF, the use of vetch increased NDF; whereas, the use of safflower decreased it (Table 2). It can be argued that the decrease in NDF in all mixtures compared to that in the seeds indicates increased intracellular material, which also indicates an increase in the digestible parts of the intracellular material (Tekce and Gül, 2014).

Different researchers and different production systems have reported ash values in barley fresh green feeds that ranged from 3 to 4.6%. (Bulletin, 1917; Peer and Leeson, 1985; Dung et al., 2010; Sharif et al., 2013; Akbağ et al., 2014). Similarly, in the present study, the ash content in all mixtures changed by 3.12 to 4.05%; however, DM losses were also observed. It is believed that the minerals found in the structure of the seeds are metabolized by enzymes, become water soluble, and are washed away by irrigation water during the germination period.

Several studies have shown that vitamin values in seeds increase with germination, but these increases can be minimal (Chavan and Kadam, 1989). Cuddeford (1989) has reported that the amount of carotene is 4.1 mg/kg DM in barley seeds and 42.7 mg/kg DM in 6-d-old fresh green feeds. In other studies, it has been reported that the carotene content increases with germination (Leontovich and Bobro, 2007) in low-carotene rice, wheat, and corn as a result of 7-d germination, with an approximately 10-fold increase in rice and wheat and a >50% increase in corn (Chattopadhyay and Banerjee, 1951). The amount of β-carotene increased by 4- to 6-fold as a result of seed germination in soybeans (Young et al., 2012; Lee et al., 2013).

Previous studies have reported that the losses of protein and vitamins are minimized with germination compared with that of other conservation methods (Leontovich and Bobro, 2007). As a result of 6-d germination, vitamin E increased from 7.4 mg/kg DM to 62.4 mg/kg DM, and it was observed that within a few days of germination, vitamins can increase by up to 20 times (Cuddeford, 1989). As a result of 2-d germination, α-tocopherol significantly increased in rice and wheat seeds (Öztürk et al., 2012; Kim et al., 2017). The addition of vetch to the barley reduced the amount of oil in the seed mixture compared to that in barley alone, but with germination, this was more advantageous in terms of vitamin E than the fat-rich safflower mixtures. In the present study, adding vetch and safflower to barley reduced vitamin E production.

Conclusion

Evaluating the results of the present study together with those of the literature, we observed that adding up to 30% vetch and LF to barley was more efficient than other mixtures in reducing nutrient losses; however, we determined that the use of LF containing plant nutrients positively affected production and that the feed values of the seeds can be increased using this production system. Therefore, we suggest that more studies are needed to determine the optimal seed mixture and the necessary systems for obtaining higher feed value from that mixture.

Declarations

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All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Cüneyt Temür and Hikmet Özdemir. The first draft of the manuscript was written by Cüneyt Temür and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

This is an observational study. The Yüzüncü Yıl University Animal Experiments Local Ethics Committee (YUHADYEK) has confirmed that no ethical approval is required.

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