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Effect of seeding rate days to harvest, and inclusion of hydroponically grown barley fodder in the diet of growing lambs

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THE EFFECTS of seeding rate (2, 3, 4, 5, 6 and 7 kg m⁻²) and days to harvest (7, 10, 15, 20 and 25 days) on the nutrients and mineral contents of hydroponically grown barley fodder (HBF) were assessed. The effects of days to harvest on the nutrient and mineral contents of HBF increased significantly with increasing days to harvest. Seeding rates had no effect on nutrients except for crude fiber (CF). The effect of including HBF (treatment diet) fed to growing Riverine lambs and Rhodes grass hay (RGH) was also evaluated. Results of a 12-week feeding trial on 7-10-month-old lambs indicated that including HBF in the diet of the treatment group significantly improved total voluntary dry matter intake (3850 g wk⁻¹ vs 4713 g wk⁻¹) for the control and treatment diet groups, respectively. However, lambs fed on the diet that included HBF ended with lighter body weight (27396.5 vs. 29528 g). This is despite the observation that they were started with a heavier initial body weight (28392.8 vs. 25317.5 g). This discrepancy may relate to the rates of body weight changes among the lambs in the experimental group. Lambs in the control group gained weight faster (467.8 g wk⁻¹) than the lambs in the treatment group, which lost weight at 110.7 g wk⁻¹. The inclusion of HBF in the diet of growing lambs had no effects on apparent dry matter (DM) or CF digestibility coefficients. However, supplementing the RGH diet with HBF may improve the growth performance.

Keywords: *Hordeum vulgare*, Hydroponic, Nutrition, Proximate analysis, Rhodes grass.

Introduction

Fresh water availability for household and agricultural purposes in arid and semi-arid areas presents a significant constraint to community development. The ever-increasing human population and intense human activities, coupled with an elevated demand for food for both humans and animals in these regions, exert heavy pressure on the meager water resources (Chowdhury & Al-Zahrani, 2015; Al Ahmadi et al., 2019).

Green fodder constitutes the primary component of the livestock diet; however, limited production and low-quality fodder often compromise livestock productivity (Ahamed et al., 2022). Efficient livestock production requires a continuous supply of green fodder throughout the year (Dung et al., 2010). Nevertheless, climatic challenges, prolonged growing periods, limited land resources and competition from cereal crops negatively impact green fodder production (Kumari et al., 2019).

Meanwhile, the surging population demands high meat production, further increasing the need for green fodder availability. Considering the aforementioned challenges, the hydroponic system offers a promising solution for green fodder production under a protected environment within a limited timeframe (Kumar et al., 2018; Ahamed et al., 2022). The hydroponic system is a viable technique for producing cost-effective green fodder year-round in low rangelands or areas with limited water resources, such as deserts, rocky soils and peri-urban and urban areas with high land costs for agricultural infrastructure (Beithou et al., 2022; Abdelraouf & Hamza, 2024). This aligns with findings in Egypt where hydroponic and aquaponic systems were shown to significantly improve water productivity under arid environments and climate challenges (Abdelraouf & Hamza, 2024). Hydroponic forage or fodder production involves sprouting grains in a water or nutrient-rich solution

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within a soilless system under controlled conditions, allowing for the fastest possible growth (Dung et al., 2010; Bulcha et al., 2024). Typically, producing approximately 1 kg of hydroponically grown barley fodder (HBF) requires only 1.5 to 2 liters of water. In contrast, growing the same amount of barley through open field cultivation necessitates around 73 liters of water (Al-Karaki & Al-Momani, 2011). Notably, 1 kg of barley grains can generally produce around 8kg of green fodder, regardless of the season or other environmental factors (Abouelezz et al., 2019). Green hydroponic fodder production is gaining attention for livestock production worldwide. Due to limited resources for green fodder in the Middle East, Africa and Asia, it can be a viable alternative to the meager pastures in these areas (Bakshi et al., 2017).

Barley is the third most important crop after wheat and rice in terms of production (FAO, 2014). Barley's adaptability to diverse environments makes it highly suitable for alternative fodder systems (Sayed et al., 2017). The feed quality of hydroponically grown fodder is higher than dry grains in terms of protein, fibers, vitamins and minerals, which ultimately can improve livestock performance (Al-Baadani et al., 2022; Wu et al., 2024). The process of grain soaking, germination and sprouting produces quality fodder by activating proteases, which break complex proteins into essential carbohydrates and amino acids (Abdel-Wareth et al., 2023). Besides the positive benefits of HBF on the overall performance of livestock in some studies (Abouelezz et al., 2019; Ma et al., 2024; Zang et al., 2024), few other studies either found no significant difference (Abbas & Musharaf, 2008; Sharif et al., 2013; Raeisi et al., 2018; Hashemi et al., 2024) or even lower performance (Fafiolu et al., 2006; Fazaali et al., 2012; Abouelezz et al., 2019; Al-Baadani et al., 2022). The adverse effects of barley sprouts could be due to low germination or excessive sprouting, which promotes poor nutritional factors and produces an undesirable bitterness in the fodder (Fafiolu et al., 2006; Smith et al., 2022). Moreover, if the relative humidity is not controlled, it may encourage mold or fungus growth, reducing fodder yield (Smith et al., 2022) or negatively impacting livestock production (Amani et al., 2020). Hydroponic barley fodder production is generating significant interest among livestock stakeholders as a sustainable option to reduce competition between food and feed production (Tranel, 2013; Bulcha et al., 2022; Smith et al., 2022). Furthermore, it aligns with the European Union (EU) regulations addressing public concerns regarding animal welfare and organic farming, especially emphasizing the accessibility to fresh green fodder (EU, 1999). Arable farming in Saudi Arabia heavily relies on conventional irrigation methods drawing from dwindling underground water reservoirs (Al Ahmadi et al.,

2019). Chowdhury and Al-Zahrani (2015) reported that 83-90% of the total water demands in Saudi Arabia were attributed to cultivating cereals, vegetables, fruits and forage crops during 1990-2009. Camels and small ruminants (sheep and goat) husbandry are the mainstay for a large portion of the rural population in Saudi Arabia. It secures a significant portion of the local market's needs for meat animals, including lamb and mutton (Al-Ghaswyneh, 2022). Communal grazing of camel, sheep and goat herds supplemented with locally grown green fodders or imported hays represents the prominent feature of the husbandry system adopted in rural Saudi Arabia. The arid climate prevailing in Saudi Arabia presents a major constraint to both agricultural production and livestock husbandry. The country receives light seasonal rain showers on sand-rocky rugged soils and lacks freshwater bodies, rivers or streams. The limited and dwindling underground water resources of Saudi Arabia fall short of the current requirements for domestic, industrial and agricultural production (Baig et al., 2022). Therefore, planning and managing water resources sustainably is a prerequisite for agricultural crop production in Saudi Arabia (Mahmoud & Gan, 2019; Rahman et al., 2022). The strategy encourages, among other things, research efforts and proposals to develop and implement non-conventional agriculture, water conserving/water-harvesting technologies for producing fodder and vegetable crops. Hydroponic fodder production has the potential to improve and sustain irrigation water use efficiency, as demonstrated in some African and Asian countries, including Saudi Arabia. However, hydroponic fodder production for livestock production in Saudi Arabia has yet to be fully established (Rahman et al., 2022).

This research investigates some agronomic aspects of HBF and compares its feeding value to that of conventionally cultivated fodder like Rhodes grass. Both *in vitro* and *in vivo* research approaches were employed to facilitate these comparisons. Therefore, the current study aimed to investigate the effects of seeding rate and days to harvest on nutrient content, cell wall composition, specific mineral content of HBF and the performance of growing riverine lambs fed Rhodes grass hay (RGH) supplemented with HBF.

Materials and Methods

Production of hydroponically grown barley fodder (HBF)

Barely seeds (*Hordeum vulgare* L.) of the local cultivar Gesto were cleaned, washed in clean water and sterilized with a 10% sodium hypochlorite solution for approximately one hour. The sterilized seeds were then thoroughly drained twice using clean water and soaked for approximately 12 hours

in germination bags before being transferred to the germination chamber. The soaked seeds were maintained in the germination chambers at a controlled temperature (25°C) and humidity (40-80%) for two days. Afterward, the sprouted seeds were grown in perforated plastic trays within a closed, re-circulating automated hydroponic system. The conveyor culture system was equipped with an automated, controlled microenvironment with solid set sprinklers (BkGreenhouses, Asan, Korea.). The environmental conditions were set to 16-18 hours light, 18-23°C air temperature, 16-23°C water temperature, and 80% relative humidity. To determine the optimal seeding rate for maximum forage production in barley, the seeds were sown at densities of 2, 3, 4, 5, 6 and 7 kg m⁻². The experiment was terminated at 7, 10, 15, 20 and 25 days from sowing to determine the optimal harvesting time for barley in the hydroponic system. Data were collected on the effects of seeding rates and days to harvest on a) nutrient composition, b) cell wall constituents and c) mineral content of HBF.

Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) fractions of the cell wall of hay and forages were determined as described by the Association of Official Analytical Chemists (AOAC) (AOAC, 2005). The mineral content of the tested HBF, including potassium (K); phosphorus (P); calcium (Ca); magnesium (Mg); sodium (Na); zinc (Zn); copper (Cu); manganese (Mn) and iron (Fe) was analyzed using an atomic absorption spectrophotometer (AOAC, 2003).

Proximate analysis methods of the AOAC (AOAC, 2005) were used to determine the nutrient content of pre-prepared and stored fodder samples and refusals. The nutrients analyzed were crude protein (CP), ether extract (EE), crude fiber (CF) and ash. Moisture contents of hays and fecal samples were determined by overnight oven drying at 105°C. The moisture and dry matter (DM) contents of HBF were determined by freeze-drying the lush green samples.

In-Vivo feeding trials

For feeding tests, 4 kg m⁻² of sprouted barley seeds were used to harvest HBF on the 7th day after sowing. Comparative feedlot performance and the apparent digestibility experiments were conducted using eighteen, 7-10-month-old Sudan riverine ecotype lambs (Shugur). The feeding test was extended to 12 weeks, consisting of 3 weeks of preliminary and 9 weeks of experimental periods. The 27 experimental lambs were randomly divided into three groups. The first group was offered only RGH, the second group was given only HBF and the third group was allowed a free choice between RGH and HBF in separate adjacent containers. All the experimental animals received enough of their respective feeds to allow ad-libitum consumption

during the experimental period. Moreover, each animal was vaccinated against endemic diseases and treated with a broad-spectrum anthelmintic drug (Ivomec) after being marked and ear tagged. The lambs were individually housed in 1.5x 2.0 m pens furnished with fine sand under an open-sided barn in the sheep unit of King Faisal University (KFU) Agriculture and Veterinary Research Station, Al-Ahsa, Eastern Province, Saudi Arabia. The pens had an automatic water supply, one or two feed utensils, and salt licks.

Fresh seven-day-old HBF sprouts were harvested, and the lambs were immediately fed at 9 a.m. every day, followed by collecting and weighing previous feed refusals. A sufficient and nutritionally homogenous quantity of RGH was procured to feed the lambs throughout the feeding test periods and was stored appropriately. Daily feed intakes and refusals were recorded, and weekly feed intakes were calculated in grams. Body weight changes were determined weekly. Initial and final body weights of the lambs were recorded at the beginning and end of the experimental period of the feeding test. Voluntary RGH and HBF consumptions, live body weight changes, and feed conversion ratios (FCR) were calculated weekly.

The feed digestibility test began upon the termination of the feedlot performance test and extended to 26 days, consisting of 5 days of adaptation and 21 days of collection periods. Eight lambs were randomly divided into two equal groups of four lambs each and housed in wooden digestibility crates designed for the quantitative total fecal collection. The first group was offered only the RGH control diet, while the second group was offered RGH and HBF in the adjacent containers. Feeds delivered, refusals, and fecal output were collected and recorded every 24 hours. Air-dried feces for lambs were pooled weekly, and the composited samples were picked, ground and stored in stoppered glass jars for proximate analysis after determining the DM content.

Statistical analysis

Two-way analyses of variance (Steel & Torrie, 1980) were employed in a randomized complete block design (RCBD) with individual independent variables (nutrients and minerals) as blocks and two factors (seeding rate and days to harvest). Duncan's Multiple Range Test (DMRT) was used to differentiate between significant treatment means ($P < 0.05$).

In vivo, lamb feedlot performance and digestibility coefficients data were subjected to an online two-tailed unpaired t-test at $P > 0.05$, according to Bruin (2006). Variables tested include fresh (as-is) and DM intakes, body weight changes, FCRs and DM and CP apparent digestibility coefficients.

Results

Compared to RGH alone, HBF in the control diet contained low levels of DM, CF, ash content, and

cell wall components, but higher levels of CP and total digestible content within the DM (Table 1).

Table 1. Nutrients composition of control diet or treatment diet.

Nutrients (%DM)	Control diet	Treatment Diet (RGH & HBF) [†]	
	(RGH alone)	HBF	
Dry matter (DM)	91.21	13.4	75.14
Crud Protein (CP)	7.52	14.4	9.24
ther Extract (EE)	2.26	3.16	2.37
Crude Fiber (CF)	16.69	10.18	15.02
Ash	9.11	2.82	7.53
Neutral detergent fiber (NDF)	71.73	26.34	60.63
Acid detergent fiber (ADF)	38.2	11.96	31.68
Acid detergent lignin (ADL)	6.34	2.83	5.54

Calculated composition of diet consumed by lambs in the treatment group

RGH = Rhodes grass hay

HBF = Hydroponic barley fodde

Chemical composition and quality assessment of HBF

The effects of days to harvest (7, 10, 15, 20 and 25 days) and seeding rates (2, 3, 4, 5, 6 and 7 kg m⁻²) of HBF on nutrient composition and cell wall constituents were investigated (Table 2 and 3). The

results indicate that days to harvest had highly significant ($P < 0.001$) effects on all the tested nutrients and cell wall constituents. However, days to harvest had no impact on saponin-tannin contents in HBF harvested on the specified days (data not shown).

Table 2. Effect of days to harvest on nutrients composition, cell wall constituents of HBF.

Nutrients %	Days to Harvest					MSE
	7	10	15	20	25	
Crude Protein (CP)	13.6 c	14.9 c	16.8 b	18.8 a	18.7a	1.53
Ether Extracted (EE)	2.8 c	3.1 c	4.02 b	4.9 a	5.0 a	0.46
Crude Fiber (CF)	11.4 c	12.8 c	16.5 b	18.1 b	22.2 a	2.3
Ash	2.9 c	3.3 c	4.4 b	5 b	6.1 a	0.63
Neutral Detergent Fiber (NDF)	29.7 c	31.7 c	41.9 b	43.9 b	53.2 a	5.27
Acid Detergent Fiber (ADF)	14.3 c	17.2 c	23.7 b	24.02 b	30.8 a	3.69
ADL - Lignia	3.78 c	4.02 c	6.65 b	7.7 ab	9.85a	1.94

a,b,c = Means in the same row with similar letters are not significantly different at $P \leq 0.05$.

MSE = Means standard error.

It was observed that CP, EE, CF and ash nutrients, in addition to NDF, ADF and ADL components, increased from 13.6, 2.8, 11.4, 2.9, 29.7, 14.3, 3.78% in DM at 7-days to harvest to 18.7, 5.0, 22.2, 6.1, 53.2, 30.8, 9.85% in DM at 25-days to harvest (Table 2), respectively. The highest increments were observed with the advance of harvest days in cell wall constituents, ash and CF (Table 2).

Table 3. Effects of seeding rate on nutrients composition and cell wall constituents of HBF.

Nutrients Composition (%DM)	Seeding Rate (kg m ⁻²)						MSE
	2	3	4	5	6	7	
Crude Protein (CP)	16.6	16.4	16.5	16.7	16.0	17.1	1.526
Ether Extracted (EE)	3.9	3.9	4.1	4.0	4.0	3.90	0.46
Crude Fiber (CF)	15.6 c	14.8 c	17.6 ab	15.0 ab	15.9 ab	18.3 a	2.3
Ash	4.3 ab	4.1 ab	4.5 ab	3.8 ab	4.4 ab	4.8 a	0.627
Neutral detergent Fiber (NDF)	38.2	38.5	42.2	39.3	38.0	44.40	5.274
Acid Detergent Fiber (ADF)	24.6	19.8	21.2	21.9	20.80	23.70	3.69
Acid Detergent Lignin (ADL)	7.8	5.9	5.2	7.4	6.00	6.00	1.94

a,b,c = Means in the same row with different superscripts are significantly different at ($P \leq 0.05$)

MSE = Means standard error.

The graded levels of seeding rate (2, 3, 4, 5, 6 and 7 kg m⁻²) did not affect the nutrient contents of HBF, except for CF and ash (Table 3). The effects of days to harvest and seeding rate on the macro (K, P, Na, Ca and Mg) and micro mineral contents (Fe, Zn, Mn and Cu) in HBF DM were also measured, respectively (Tables 4&5). The reported results provide sufficient evidence that days to harvest had highly significant incremental effects on the macro and micro mineral contents of HBF (Table 4). It was observed that K, P, Na, Ca and Mg increased from 4469.3, 4252.5, 1259.0, 1374.8 and 1635.3 ppm at 7 days to harvest to 6926.0, 6076.0, 5167.2,

4347.2 and 2801.7 ppm, respectively, at 25 days to harvest. The increase in micro minerals was proportionally less as days of harvest progressed. Iron, Zn, Mn and Cu contents of HBF increased from 127.7, 48.2, 22.2 and 1.7 ppm at 7 days to harvest to 145.0, 119.1, 23.4, and 6.4 ppm, respectively, at 25 days.

The seeding rate had a significant ($P > 0.05$) effect on the contents of Fe, Mg, Ca, P, and Cu but no impact on the contents of Mn, Zn, Na and K in HBF (Table 5). Minerals such as K, P, Mn and Cu showed increased content, while the contents of the tested minerals Na, Zn, Ca, Mg and Fe decreased with increasing seeding rate.

Table 4. Effects of days to harvest on the composition of some nutrient in HBF.

Nutrient	Days to Harvest				
	7	10	15	20	25
Macro minerals (ppm)					
Na	1259.0 c	1710.2 c	3624.0 b	3731.20 b	5167.2 a
Mg	1635.3 c	1635.3 cd	1974.8 bc	2018.8 b	2801.70 a
Ca	1374.8 c	1356.8 c	2013.0 b	2287.7 b	4347.2 a
P	4252.5 c	3922.7 c	4751.2 b	4355 bc	6076.0 a
K	4469.3 c	4521.2 c	6055.5 ab	5813.0 b	6926.0 a
Micro minerals (ppm)					
Fe	127.71 a	66.96 b	88.37 ab	88.44 ab	145.02 a
Mn	22.18	14.24	35.87	15.48	23.420
Zn	48.18 ab	36.07 c	58.35 b	68.65 b	119.08 a
Cu	1.68 c	0.43	1.80 c	3.33 b	6.38 a

a,b,c = Means in the same row with different superscripts are significantly different at ($P \leq 0.05$)

MSE = Means standard error.

Feed intake and nutrient digestibility

A comparison of the voluntary feed intakes and body weight changes of the two groups of growing lambs offered RGH alone (first group) and a free choice between RGH or HBF (third group) is presented in Tables 6 and 7, respectively. The second group of lambs, offered only HBF, either refused it entirely or consumed very little. By the end of the first two weeks of the preliminary period, their average daily consumption was only 412.0 g of fresh HBF, which translated to a mere 40.0 g of

DM. Their body condition significantly deteriorated, with an average weight loss of 17% from their initial body weight (Data not shown). Due to these concerning welfare implications, the experimental trial for the HBF control group was discontinued.

In vivo trials proceeded with only the control group (receiving RGH alone) and the treatment group (receiving both RGH and HBF simultaneously). The data showed that the lambs in the treatment group significantly consumed more total DM (4713 g wk⁻¹, equivalent to 2.5% body weight) (Table 6)

compared to the average total DM consumed by the lambs in the control group (3850 g wk⁻¹, equal to 1.9% body weight). However, it must be noted that the lambs in the two groups consumed similar, but insignificantly different amounts of RGH (3850 vs. 3500 g wk⁻¹). Lambs in the treatment group consumed, in addition to RGH, approximately 1277 g wk⁻¹ DM of HBF. This observation suggests that lambs in the treatment group were selected against

HBF and preferred RGH (3500 g wk⁻¹ vs. 1277 g wk⁻¹). The ratio of RGH to HBF consumed by the treatment group was calculated as 3:1. The results also suggest that providing fresh green HBF on animal diets does not improve voluntary consumption of other forages (RGH). Total average voluntary intake by treatment lambs amounted to approximately 12,701 g wk⁻¹ compared to 4420 g wk⁻¹ for lambs in the control group.

Table 5. Effects of seeding rate on the composition of some mineral elements in HBF.

Nutrient	Seeding Rate (kg m ⁻²)						MSE
	2	3	4	5	6	7	
Macro minerals (ppm)							
Na	3676.5	3534.6	3629.4	2251.7	2557.30	2940.40	1029.61
Mg	1879.2 bc	1988.2 bc	2330.4 a	1720 c	1875.2 c	2234.8 ab	288.14
Ca	2356.4 abc	2230.8 bc	2758.6 a	1846.9c	1987.0 c	2479.2 ab	423.19
P	4331.6 b	4632.8 ab	5345.0 a	4300.6 b	4419.0 b	5000.0 ab	Tab
K	5347	5470.4	6096	5078.6 b	5473.2 ab	5896.80	857.48
Micro minerals (ppm)							
Fe	151.04 a	99.6 ab	107.62 ab	87.85 ab	81.96ab	91.72 ab	44.01
Mn	15.83	16.32	20.39	15.6	19.83	41.83	22.97
Zn	74.48	68.86	76.48	59.48	58.04	58.84	17.14
Fe	151.04 a	99.6 ab	107.62 ab	87.85 ab	81.96ab	91.72 ab	44.01
Cu	2.67 ab	2.86 ab	3.64 a	2.10 b	2.40	2.56 ab	0.91

a,b,c = Means in the same row with different superscripts are significantly different at ($P \leq 0.05$)

MSE = Means standard error.

Table 6. Voluntary intake of growing lambs offered control diet or treatment diet.

Variable	Control diet	Treatment diet	Significance
Total voluntary dry matter intake (g wk⁻¹)	3850.0 ± 345.23	4713.4 ± 180.56	*
RGH voluntary dry matter intake (g wk ⁻¹)	3850.0 ± 345.23	3500.3 ± 349.14	N.S
HBF voluntary dry matter intake (g wk ⁻¹)	0000.0 ± 0000.0	1276.9 ± 148.72	***
Total voluntary as - is matter intake (g wk ⁻¹)	4419.6 ± 244.31	12701.0 ± 641.32	*****

N.S = Non-significant at $P \leq 0.05$ * = Significant at $P \leq 0.05$ *** = Significant at $P \leq 0.001$

***** = Significant at $P \leq 0.0001$

The data showed that lambs in the treatment group gained significantly more body weight (28,392 g lamb⁻¹) compared to the control group (25,377 g lamb⁻¹) at the beginning of the experiment (Table 7). However, the situation was reversed by the end of the 12-week feedlot test. The control group of lambs gained a higher average body weight (29528 g) than the treatment group (27397 g). Lambs in the control group gained live body weight at a rate of

467 g wk⁻¹ lamb⁻¹, while lambs in the treatment group lost body weight at an average rate of 111 g wk⁻¹ lamb⁻¹. The difference in FCR was highly significant between the two groups of lambs (Table 8). The control group of lambs had a positive FCR (8.2±2), within the normal range reported for ruminants. Lambs in the treatment group had a negative, significantly high, and highly variable FCR (-42.6±15.4).

Table 7. Initial, final body weights, body weight changes and feed conversion ratios (FCRs) for growing lambs fed a control diet or treatment diet

Variables	Control diet	Treatment diet	Significance
Initial body weight (g)	25317.5 ± 2333.41	28392.8 ± 2816.12	*
Final body weight (g)	29528.0 ± 2283.16	27396.5 ± 2883.76	N.S
Body weight changes (g week ⁻¹ lamb ⁻¹)	467.8 ± 120.22	- 110.7 ± 143.41	*
Feed Conversion ratios	8.2 ± 2.0	- 42.6 ± 15.44	**

N.S = Non-significant at $P \leq 0.05$ * = Significant at $P \leq 0.05$ ** = Significant at $P \leq 0.001$

Table 8. Dry matter, crude protein and crude fiber apparent digestibility coefficients in lambs fed on a trol diet or treatment diet.

Variable	Control diet	Treatment diet	Significance
1. Dry matter			
A. ingested (g d ⁻¹)	702.4 ± 14.84	637.5 ± 15.43	*
B. Excreted (g d ⁻¹)	346.4 ± 75.6	357.6 ± 37.09	N.S
C. Apparent digestibility Coefficient (%)	51.13	43.86	N.S
2. Crude Protein			
A. ingested (g d ⁻¹)	52.8 ± 1.1	58.9 ± 1.43	N.S
B. Excreted (g d ⁻¹)	49.81 ± 10.89	56.5 ± 5.8	N.S
C. Apparent digestibility Coefficient (%)	13.9 ± 12.26	9.61 ± 6.70	N.S
3. CF			
A. ingested (g d ⁻¹)	117.68±2.21	99.03±5.42	*
B. Excreted (g d ⁻¹)	55.91±1.73	38.87±9.18	N.S
C. Apparent digestibility Coefficient (%)	52.46±10.27	59.10±10.40	N.S

N.S = Non-significant at $P \leq 0.05$ * = Significant at $P \leq 0.05$

Comparative data for ingested, excreted (g d⁻¹) and apparent digestibility coefficients of DM (% DM), CP and CF are presented in Table 8. For all variables investigated except DM and CF intake, no significant differences were observed between treatments. The digestibility coefficients of DM and CP were not affected by the diet but were higher for lambs in the control group (53.8 and 13.5%) compared to lambs in the treatment group (43.9 and 9.6%). The relatively low levels of CP apparent digestibility coefficients observed in this study (13.9 and 9.6%) could be associated with the low CP levels of the consumed fodders (Table 1), considering the indigenous CP secreted in the gastrointestinal tract (GIT) during digestion and absorption processes.

Discussion

The HBF is non-conventional roughage with potential benefits for livestock (Ma et al., 2024; Zang et al., 2024). In our study, we found that HBF had lower DM, CF, and ash content than RGH, conventional roughage. However, HBF had higher CP and total digestible content (TDC). These results are consistent with previous studies (Al-Saadi & Al-Zubiadi, 2016; Al-Baadani et al., 2022). Many studies have reported a 7-47% decrease in DM content of sprouted barley compared to seed grain. The decrease in DM content of HBF is likely due to the sprouting process (Bulcha et al., 2024). During sprouting, barley seeds utilize carbohydrates for energy, resulting in the breakdown of cell walls and the release of nutrients. This process also increases CP content (Al-Baadani et al., 2022; Smith et al., 2022; Bulcha et al., 2024). Rhodes grass hay (RGH) is the preferred roughage in Saudi Arabia because it has optimal DM, CF, and cell wall components. However, it has marginal CP

content. HBF can be used to supplement RGH to improve CP content and overall nutritional value.

Previous studies have shown that the nutrient content of HBF increases with the number of days to harvest (Mohamed et al., 2021; Al-Baadani et al., 2022; Bulcha et al., 2022; Bulcha et al., 2024). This trend was also observed in the current study. Compared to the original grains, HBF has a higher nutrient content, particularly in terms of CP, NDF and ADF content (Farghaly et al., 2019; Arif et al., 2023; Bulcha et al., 2024; Wu et al., 2024). The presence of chlorophyll in HBF, known as the "grass juice factor," also contributes to improved livestock performance (Naik et al., 2017; Soufan, 2023). However, the available metabolizable energy (ME) of HBF is lower than that of barley grains (Fazaeli et al., 2012; Bulcha et al., 2024). The current study also observed the least increase in CP content, consistent with previous findings (Al Ajmi et al., 2009; Fazaeli et al., 2012). These changes in nutrient content were accompanied by a decrease in moisture content as the number of days to harvest increased. The protein content of sprouting grains may vary depending on the nitrogen supplementation and the availability of other nutrients (Ortiz et al., 2021; Gümüő et al., 2024). This suggests that the roughage characteristics of HBF improve with longer harvest times (Bulcha et al., 2024). HBF harvested after 15 days could be processed into roughage in the form of hay or silage, containing high CP levels (16.8-18.7% in DM) and being free of saponin-tannin. Conversely, HBF harvested before 15 days has a high moisture content (acting as a diluent), low DM content, diluted nutrients, and high levels of CF and cell wall components (Ortiz et al., 2021; Ahamed et al., 2022; Arif et al., 2023; Bulcha et al., 2024). Variability in HBF composition among studies may also be attributed to inherent genetic differences

among barley cultivars, as genotype has been shown to influence biomass productivity and nutrient assimilation (Abdelwahab et al., 2024).

In our study, we found that different seeding rates had little or no significant effect on the nutrient quality of HBF (Table 3). This aligns with previous research demonstrating that high seed rates do not significantly improve the overall performance of hydroponic fodder. For example, Naik et al. (2017) found that varying seed rates of hydroponically grown maize did not significantly affect the nutritional content of the roots, leaves, or whole plant. Similarly, Assefa et al. (2020) observed that while high seed rates increased fodder yield in terms of DM, medium or low seed rates produced higher DM fodder conversion efficiency and reduced the cost per kg of fodder production. Other studies have reported similar findings concerning fodder yield efficiency and economic outcomes (Iqbal et al., 2021; Elmulthum et al., 2023; Al-Khateeb et al., 2024). Similarly, biomass efficiency and land use optimization in barley-based systems have been demonstrated under Egyptian conditions (Salama et al., 2016), reinforcing the association between seeding density and biological yield efficiency. Our results suggest that high seed rates are unnecessary when growing HBF hydroponically, leading to cost savings and improved fodder conversion efficiency (Mustafa et al., 2023). Furthermore, agronomic variation in response to management factors has also been documented for barley grown in sandy soils in Egypt (Abd El-Hameed, 2011), supporting the relevance of assessing seeding rate effects under differing production environments.

Our study found an increase in the macro and micro mineral content as the days to harvest increased. This suggests that the mineral content also increases with increasing days to harvest. The findings of studies by Bulcha et al. (2022) and Bulcha et al. (2024) support this observation, indicating that the mineral content of HBF depends on the number of days it is grown and the concentration of salt mixtures in the water. This implies that the composition of HBF could be tailored to meet the specific needs of animals. If the minerals in HBF are digestible, they appear to be sufficient to fulfill the maintenance requirements of growing small ruminants.

Increasing the seeding rate of HBF significantly increased the levels of K, P, Mn and Cu. However, it also decreased the levels of Na, Zn, Ca, Mg and Fe in HBF. This likely occurs because the increased number of seedlings compete for these minerals in the salt solution of the irrigation water (McKenzie et al., 2004; Naik et al., 2015; Mustafa et al., 2023). Additionally, a higher seeding rate leads to a higher seed density, which can promote microbial growth in the root mat and negatively affect the quality of the fodder (Mustafa et al., 2023). This is because

the microbes can utilize nutrients and release toxins that can make the fodder less palatable and nutritious (Naik et al., 2015). In extreme cases, high seed density can lead to mold growth, such as *Aspergillus clavatus* (LL Ntsoane et al., 2023). This mold can produce toxins that can cause a variety of health problems in livestock, including posterior ataxia, knuckling of the fetlock, hind legs, stiff gait, hypersensitivity, colonic convulsions, reduced milk production, and even death (McKenzie et al., 2004). The study found that lambs in the treatment group consumed significantly more total DM than lambs in the control group. However, providing fresh green HBF did not improve the voluntary consumption of RGH. The succulent and bulk nature of HBF in the treatment diet may have accelerated the rate of digestion in the GIT, potentially reducing nutrient digestion (Raeisi et al., 2018; Al-Baadani et al., 2022). Furthermore, the negative growth rate observed in the treatment group suggests that the succulent, freshly cut HBF may contain factors that depress growth (McKenzie et al., 2004).

The study also found no significant differences in the lambs' ingested, excreted, and apparent digestibility coefficients for traits other than DM and CF intake. This could be attributed to the increased rate of passage through the GIT caused by the bulkiness of the roughages, particularly those containing hydroponic fodder (Raeisi et al., 2018). Although livestock animals generally prefer green hydroponic fodders due to better digestibility and palatability, some studies have found that it may not have significant benefits. For example, a study by Saidi and Omar (2015) found that HBF had no significant effect on feed intake, body weight gain, milk production, or composition in HBF-fed ewes. Similarly a study by Fazaeli et al. (2011) found that HBF had no significant effect on the average daily gain (ADG) of calves, and it actually increased the cost of feeding by 24%. Additionally, the DM of HBF was lower than the original barley grains. In another trial, Zang et al. (2024) reported that replacing conventional concentrates with HBF in cows had no significant effect on milk yield (fat, protein and lactose), rumination time or other physical activities. Interestingly, however, DM intake was significantly reduced. In terms of sensory characteristics (meat structure, color, tenderness, and smell), Hashemi et al. (2024) found no significant effect on goats when HBF replaced alfalfa. Other studies have found that HBF may have negative effects on livestock. For example, a study by Hosainy-Abrandabadi et al. (2015) found that HBF caused a decrease in DM intake and daily gain in fattening lambs. A more recent study by Fazaeli et al. (2021) found that replacing maize silage with 60% HBF had no significant effect on the overall performance of lactating cows, but it did increase the cost of feeding by 24%. Similar

findings were also reported by Sulser (2015), where HBF could not produce any significant effect on the average daily gain of sheep as compared to other cereal grains and alfalfa.

It is worth noting that the lambs offered the RGH alone readily consumed it throughout the preliminary and experimental periods of the test (12 weeks) with no observable ill effects, morbidities, mortalities. However, the lambs offered the HBF alone were reluctant to eat during the beginning of the preliminary period, and their body condition and health deteriorated significantly as the test progressed. The lambs in the group showed symptoms of profuse watery diarrhea, dehydration, feed refusal-seizure and loss of body condition and weight. Two of the nine lambs in the group died. This suggests that HBF may not be a suitable feed for all livestock animals. Accordingly, the treatment with HBF alone was terminated, and the feedlot test continued with only two lamb groups (RGH group alone and RGH-HBF simultaneous group).

The nutrient profile of HBF is usually altered during sprouting, which can be beneficial for animal production (Bulcha et al., 2024). However, some evidence suggests that HBF may not be as beneficial for ruminants as it is for other animals (Al-Baadani et al., 2022). For instance, HBF is a succulent type with high water content, making it suitable only as a supplement to the total mixed ration for animals (Al-Saadi & Al-Zubiadi, 2016; Al-Baadani et al., 2022). Additionally, changes in the rumen microbiota can disrupt the daily feed intake and digestibility of ruminants fed on HBF (Ren et al., 2022; Ma et al., 2023). Additional joint supplements, Ca, vitamin A&D and specific amino acids like lysine are still needed, as these are unavailable in sprouts (Ortiz et al., 2021; Abdel-Wareth et al., 2023; Wu et al., 2024). Therefore, hydroponic fodder can significantly benefit small animals (non-ruminants) and birds due to their less complex digestive systems (Al-Kanaan, 2022; Abdel-Wareth et al., 2023). Further investigations regarding the biosafety of HBF production and the optimal assessment of its quality are highly recommended. It is important to note that the results of studies on HBF can vary depending on the type of animal being studied, its growth stage, the HBF production method, and other factors. Consulting with a veterinarian or animal nutritionist before feeding HBF to animals is crucial. Moreover, a preliminary economic analysis is necessary to determine the feasibility of starting a hydroponic cultivation project (Elmulthum et al., 2023; Al-Khateeb et al., 2024).

Conclusion

The current study concluded that supplementing a traditional RGH diet with HBF improved the nutritional status and nutrient digestibility of Reverine lambs. However, adding only sprouted

barley in the daily ration negatively affected the daily feed intake, overall growth and performance of the small ruminants. These findings suggest that replacing RGH with HBF in a ratio-dependent manner can maintain the feed intake and growth performance of lambs. Overall, the evidence on the benefits of HBF for ruminants is mixed. More research is needed to determine the optimal way to use HBF in ruminant diets. This is a promising strategy, but further studies are needed to determine the optimal ratio of RGH to HBF.

Consent for publication

All authors declare their consent for publication.

Author contribution

Conceptualization, S.A.A., T.A.M., K.M.A., and F.I.Z.; Methodology, T.A.M., M.N.S., F.I.Z., and N.A.E.; Formal analysis, M.N.S., and T.A.M.; investigation, M.N.S., S.A.A., and T.A.M.; Data Curation, M.N.S., T.A.M., and N.A.E.; Writing-Original Draft Preparation M.N.S., and T.A.M., Writing Review and Editing S.A.A., N.A.E., and F.I.Z. The manuscript was edited and revised by all authors.

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Conflicts of Interest

The authors declare that they have no conflicts of interest to report regarding the present study.

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