Effect of different dietary protein sources on digestibility and growth performance parameters in lambs

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Abstract
Background: The aim of this study is a comparison between some different protein sources in sheep rations to identify the best protein source that achieves the highest production performance and the lowest economical feed efficiency. Twenty Farafra male lambs weighed on average 41.39 ± 1.66 kg were divided randomly into four similar groups of 5 animals each. All groups were fed on concentrate feed mixture (CFM) at 3% of their body weight, and wheat straw fed ad lib. to replace 100% of the soybean meal (SBM) as a source of protein, black cumin seed meal (BCSM), cottonseed meal (CSM) and sesame seed meal (SSM) were incorporated into the CFM. The feeding trial extended for 66 days followed by digestibility and nitrogen (N) balance trials.

Results: The results indicated no difference in total feed intake between experimental groups. Digestibility of crude protein, ether extract and crude fiber for animals fed CSM ration was higher (p < 0.05) than those fed the other rations. On the other hand, the CSM ration recorded a higher value of digestible crude protein than SBM and BCSM rations. Lambs gave rations containing CSM was higher (p < 0.05) average daily weight gain compared with those fed the other rations. Lambs that fed CSM ration were better to feed conversion as kg dry matter intake/kg gain than those fed the other rations. Relative economic efficiency was the best for CSM ration as compared to other protein sources. Nitrogen balance value of sheep fed CSM diet had the highest (p < 0.05) value compared with those fed different sources. Rumen fermentation showed that the pH value was the lowest with SSM, NH3-N concentration was higher for BCSM and SSM, and total volatile fatty acids were higher for SBM compared with the other rations.

Conclusion: It could be concluded that cottonseed meal was the best treatment for digestibility coefficient, average daily gain, highest nitrogen balance, better feed conversion ratio and also the best economic efficiency compared to soybean meal, black cumin seed meal and sesame seed meal treatments. It can replace 100% soybean meal by cottonseed meal in sheep ration when economics is to be considered.

Keywords: Protein seed meal, Farafra sheep, Nutrients digestibility, Growth performance

Background
Although the progress has been achieved in livestock nutrition worldwide, most producers and nutritionists still think about crude protein only when evaluating protein feed and animal requirements. All livestock production interests either scientists or producers aimed to reduce feed costs, improved efficiency of production and dietary protein use (Schwab and Broderic 2017). The two major components of feed are protein and energy that effect on young growing and fattening animals. Protein is an essential nutrient for growing and fattening lambs and it is more expensive than other components, but optimal use of protein is necessary (Dabiri and Thonney 2004). Ruzic-Muslic et al. (2014) reported...
that as a result of increased soybean prices, import trends and production fluctuations, consumer interest in alternative sources of protein has increased. Condition of the quality of protein in animal diets improves their performance and ensures profitable productivity.

Chemistry of protein sources is differing, along with amino acid (AA) profile and crude protein (CP) availability in the rumen, also post-ruminal level (Gleghorn et al. 2004; Bateman et al. 2005). Jørgensen et al. (1984) showed that the effect of different protein sources has varied on animals’ performance characteristics biochemical indices because of changes of rumen ecology and amino acid profile; the responsible for in performance may be varied (Hall and Huntington 2008).

Solomon et al. (2008) reported that additives soybean meal (SBM), canola meal (CM), cottonseed meal (CSM) and sunflower meal (SFM) in lamb diets as different protein sources supply the condensed nutrients that are successfully used at a ruminal level. Khalid et al. (2012) showed that a greater by-pass degree for protein source has recorded higher effects on nitrogen balance, growth and muscle mass accretion compared with weaker protein by-pass. The inclusion of protein sources with AA profiles connecting closely to the AA needs of the growing animals results in greater growth performance and nitrogen utilization by the lambs.

The aim of this study is a comparison between some different protein sources (SBM, *Glycine max*.; BCSM, *Nigella sativa*; CSM, *Gossypium spp*. and SSM, *Sesamum indicum*) in Farafra sheep rations to identify the best protein source that achieves the highest production performance and the lowest economical feed efficiency. The following evaluation of alternative protein sources will provide nutritionists with information on the advantages and disadvantages of feed ingredients as well as correctly including them in the sheep’s feed.

**Methods**

This study was carried out at the Nubaria Experimental Station, Nubaria, Behera Governorate and on the Laboratories of Animal Production Department, National Research Centre, Dokki, Giza, Egypt.

**Protein sources**

In this study, four conventional and unconventional protein sources (soybean meal SBM, black cumin seed meal BCSM, cottonseed meal CSM and sesame seed meal SSM) were used on sheep rations.

**Nitrogen fractions**

The true protein nitrogen of different protein sources was determined according to AOAC (2005). Non-protein nitrogen (NPN) was calculated by subtracting the true protein nitrogen value from the total nitrogen value.

The insoluble protein of different protein sources was determined according to Waldo and Goering (1979). Soluble protein was calculated as the difference between CP and insoluble ones orderly.

**Amino acids analysis**

Amino acid content was determined as described by Spackman et al. (1958). The analysis was performed in the Central Service Unit, National Research Center, Egypt, using LC 3000 Amino Acid Analyzer (Eppendorf-Biotronik, Germany).

**Feeding trials**

Twenty fattening mature male Farafra sheep weighed in average 41.39 ± 1.66 kg randomly allocated into four similar groups (5 animals in each). SBM, BCSM, CSM and SSM were incorporated into the experimental concentrate feed mixtures (CFM) as sources of protein. The formulation and the chemical composition of the experimental CFMs are shown in Table 4. The experimental CFMs were fed to all groups at 3% of their body weight and wheat straw was fed *ad lib*. Freshwater was freely available to animals at all times. The feeding trial lasted for 66 days (around 9 weeks); during this period, animal’s body weight was recorded bi-weekly and the feed intake was adjusted.

**Digestibility trials**

At the end of the feeding trial, three animals were randomly selected from each group to determine nutrients digestibility, nutritive values and nitrogen balance. Animals were housed in individual metabolic cages; the cages allowed collection feces separately from the urine which was collected in attached glass containers containing 50 ml sulfuric acid solution 10%. The digestibility trial extended to 14 days as a preliminary period followed by 7 days for feces and urine collection. The animals were fed on 3% of live body weight, the CFM was offered in two portions at 8.30 a.m. and 2:00 p.m., and wheat straw was offered *ad lib*. During the collection period, feces and urine were quantitatively collected from each animal once a day at 8:00 a.m. before feeding. The actual quantity of feed intake and water consumption was recorded. A sample of 10% of the collected feces from each animal was sprayed with 10% sulfuric acid and 10% formaldehyde solutions, and then, it dried at 60 °C for 24 h. Samples were mixed and stored for chemical analysis. Composite samples of feeds and feces were finely ground before analysis. Also, 10% of the daily collected urine from each animal was preserved for nitrogen determination. The nutritive values expressed as total digestible
nutrients (TDN) and digestible crude protein (DCP) of the experimental rations were determined. Rumen fluid samples were collected from all animals at the end of the digestibility trial before feeding, 3 and 6 h post-feeding via stomach tube and strained through four layers of cheesecloth. Samples were separated into two portions; the first portion was used for immediate determination of ruminal pH and ammonia nitrogen (NH₃–N), concentration, while the second portion was stored at −20 °C after adding a few drops of toluene and a thin layer of paraffin oil till analyzed for total volatile fatty acids (TVFA’s). Animal welfare statements (housing, management and rations preparation) were applied in this work.

Analytical procedures
Chemical analysis of ingredients, experimental concentrate feed mixture and feces samples were analyzed according to AOAC (2005) methods. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined in ingredients and experimental CFM according to Goering and Van Soest (1970). Ruminal pH was immediately determined using a digital pH meter. Ruminal NH₃–N concentrations were determined by applying the NH₃ diffusion technique using the Kjeldahl distillation method according to AOAC (2005). Ruminal TVFA’s concentrations were determined by steam distillation according to Warner (1964).

Statistical analysis
Collected data of water consumption, feeding trials, digestibility trials and nitrogen balance were subjected to statistical analysis as one-way analysis of variance using the general linear model (Yijk = µ + Ti + eij) procedure of SPSS (2008). On the other hand, collected data of ruminal fluid parameters (pH, NH₃–N and TVFA’s concentrations) were subjected to statistical analysis as two factors-factorial analysis of variance using the general linear model (Yijk = µ + Ti + Sij + (T × S)ij + eijk) procedure of SPSS (2008), where Yij or Yijk = the observation, µ = overall mean, Ti = protein source (treatment), Sij = sampling time, (T × S)ij = treatment × sampling time interaction, and eij or eijk = experimental error. Duncan’s multiple range test (Duncan 1955) was used to separate means when it was significantly different. Because group feeding was used in the feeding trial, the data of feed intake did not subject to statistical analysis.

Results
Protein sources
Chemical composition and cell wall constituents of different protein sources are illustrated in Table 1. Data presented that SBM has the highest value of CP and nitrogen-free extract (NFE); however, it recorded the smallest value of ether extract (EE) or crude fiber (CF). However, the BCSM noted the greatest value of EE content. CSM reported the highest value of CF, while recorded the smallest value of ash content. CSM contained the highest values of NDF and ADF, but SBM showed the lowest value of NDF and ADF. BCSM contained the largest values of gross energy, while CSM noted the smallest value.

Amino acids profiles of different protein sources
Amino acid profiles of different protein sources are found in Table 2. The SBM had the biggest value of most AA’s compared with various protein sources. On the other hand, BCSM and CSM showed the lowest value of most AA’s content.

Nitrogen fraction of the experimental protein sources
The nitrogen fraction of protein sources is illustrated in Table 3. The data offered that SBM showed the biggest value of total protein and true protein; however, it recorded the smallest value of NPN as % of total nitrogen. However, BSCM contained the lowest value of insoluble protein (52.40%).

Nutritional evaluation of experimental CFMs
Chemical composition and cell wall constituents of the tested CFMs are shown in Table 4. Results concluded that CFM contained CSM showed the highest value of CF, followed by BCSM and SSM mixtures, compared with SBM that showed the smallest values. Furthermore, CP content of SBM, BCSM, CSM and SSM mixtures was closed (14.03, 13.99, 14.06 and 14.10%), respectively. BCSM mixture had the biggest value of EE content.

Feed intake
Dry matter intake of the tested groups is clarified in Table 5. The data reported that the inclusion of SBM, BCSM, CSM and SSM in ration did not significantly affect feed intake of lambs, either concentrate or roughage and subsequently total dry matter intake (DMI) intake. Sheep feeding on SSM recorded the smallest values of gross energy, while CSM noted the highest values of concentrate and total DMI, but sheep fed on SBM showed the lowest content of concentrate intake and total DMI.

Digestibility
The nutrients digestibilities of the experimental diets are also presented in Table 5. Digestibility of DM, OM, CP, CF and EE and feeding value as DCP were significantly different across the various dietary groups. The digestibility of DM, OM and CP was lower (p < 0.05) for the BCSM diet compared to the rest diets. No significant (p > 0.05)
differences were found in NFE and feeding value as TDN among the different experimental diets. In our study, CP digestibility was higher \((p < 0.05)\) for CSM and SSM compared to the SBM diet (77.24) compared to rest diets.

### Table 1 Chemical composition, cell wall constituents and gross energy of the experimental protein sources

| Item                        | Protein sources       | Soybean meal (SBM) | Black cumin seed meal (BCSM) | Cotton seed meal (CSM) | Sesame seed meal (SSM) |
|-----------------------------|-----------------------|--------------------|-----------------------------|------------------------|------------------------|
| Moisture                    |                       | 10.15              | 8.63                        | 9.62                   | 8.92                   |
| Components, % on DM basis   |                       |                    |                             |                        |                        |
| Organic matter (OM)         |                       | 92.55              | 92.47                       | 94.49                  | 91.86                  |
| Crude protein (CP)          |                       | 44.5               | 30.09                       | 26.24                  | 30.66                  |
| Crude fiber (CF)            |                       | 7.33               | 10.21                       | 29.92                  | 9.6                    |
| Ether extract (EE)          |                       | 5.01               | 21.08                       | 8.39                   | 15.9                   |
| Nitrogen free extract (NFE) |                       | 35.71              | 31.09                       | 29.94                  | 35.7                   |
| Ash                         |                       | 7.45               | 7.53                        | 5.51                   | 8.14                   |
| Cell wall constituents, % on DM basis |
| Neutral detergent fiber (NDF) |                       | 33.74              | 35.63                       | 48.58                  | 35.23                  |
| Acid detergent fiber (ADF)  |                       | 16.12              | 18.74                       | 36.72                  | 18.19                  |
| *Gross energy, kcal/kg DM    |                       | 4771               | 5396                        | 4756                   | 5107                   |

*Gross energy (kcal/kg DM) was calculated according to Blaxter (1968), where each g of CP = 5.65 kcal, each g of EE = 9.40 kcal, each g of CF and NFE = 4.15 kcal.

Nitrogen utilization

Table 6 shows the results of the nitrogen utilization of Farafra sheep fed the tested diets. Nitrogen utilization was affected by dietary protein sources. Sheep fed diet contained BCSM significantly \((p < 0.05)\) highest fecal nitrogen then SBM and SSM compared with CSM. The lowest value of nitrogen retention was reported with

### Table 2 Amino acids content of tested protein sources

| Amino acid, g/100 g | Protein sources | SBM | BCSM | CSM | SSM |
|---------------------|-----------------|-----|------|-----|-----|
| *Essential amino acids* |
| Threonine           |                 | 1.81| 1.45 | 1.07| 1.16|
| Valine              |                 | 2.59| 0.04 | 1.63| 1.40|
| Methionine          |                 | 0.68| 0.55 | 0.24| 0.38|
| Isoleucine          |                 | 2.05| 1.42 | 1.02| 4.41|
| Leucine             |                 | 5.88| 2.44 | 3.01| 4.24|
| Phenylalanine       |                 | 5.42| 1.69 | 2.97| 2.71|
| Histidine           |                 | 2.32| 1.42 | 1.25| 0.83|
| Lysine              |                 | 4.45| 1.48 | 2.18| 1.51|
| Arginine            |                 | 2.86| 3.24 | 1.75| 5.48|
| Proline             |                 | 1.80| 1.36 | 1.11| 1.25|
| *Nonessential amino acids* |
| Aspartic            |                 | 4.83| 2.97 | 2.62| 3.34|
| Serine              |                 | 2.70| 1.35 | 1.47| 1.88|
| Glutamic            |                 | 11.79| 6.22 | 6.68| 10.84|
| Glycine             |                 | 0.94| 1.28 | 0.72| 2.58|
| Alanine             |                 | 4.01| 1.74 | 2.88| 3.51|
| Cysteine            |                 | 1.60| 5.28 | 1.50| 2.10|
| Tyrosine            |                 | 2.75| 1.69 | 1.10| 1.16|
SBM. On the other hand, better digestion and nitrogen balance (NB) were observed with improving the protein concentration in the rations of growing lambs.  

**Water balance**  
Table 7 shows results of water balance for groups fed different protein sources. A nonsignificant difference \((p < 0.05)\) was observed between groups in drinking or water balance.

### Table 3 Nitrogen fraction and protein solubility of the experimental protein sources

| Item                                | Protein sources |
|-------------------------------------|-----------------|
|                                     | SBM             | BCSM    | CSM     | SSM     |
| **Nitrogen fraction**               |                 |         |         |         |
| Total protein                       | 44.50           | 30.06   | 26.24   | 30.69   |
| True protein (g)                    | 39.44           | 25.37   | 23.12   | 23.13   |
| True protein, % of total protein    | 88.63           | 84.40   | 88.11   | 75.37   |
| Non-protein nitrogen (NPN) (g)      | 5.06            | 4.69    | 3.12    | 7.56    |
| NPN, % of total protein             | 11.37           | 15.60   | 11.90   | 24.63   |
| **Protein solubility**              |                 |         |         |         |
| Insoluble protein (g)               | 41.88           | 15.75   | 22.81   | 24.25   |
| Insoluble protein, % of total protein| 94.11           | 52.40   | 86.93   | 79.02   |
| Soluble protein (g)                 | 2.62            | 14.31   | 3.43    | 6.44    |
| Soluble protein, % of total protein | 5.89            | 47.60   | 13.07   | 20.98   |

### Table 4 Formulation, chemical composition, cell wall constituents and gross energy of the experimental concentrate feed mixtures

| Item                                | Concentrate feed mixtures |
|-------------------------------------|---------------------------|
|                                     | SBM           | BCSM           | CSM           | SSM           |
| **Formulation (%)**                 |               |                |               |               |
| Yellow corn                         | 50            | 50             | 50            | 50            |
| Soybean seed meal                   | 16            | –              | –             | –             |
| Black cumin seed meal               | –             | 23             | –             | –             |
| Cottonseed meal                     | –             | –              | 28            | –             |
| Sesame seed meal                    | –             | –              | –             | 21.5          |
| Wheat bran                          | 31            | 24             | 19            | 25.5          |
| Limestone                           | 2             | 2              | 2             | 2             |
| Common salt                         | 1             | 1              | 1             | 1             |
| Total                               | 100           | 100            | 100           | 100           |
| **Chemical composition, % on DM basis** |       |                |               |               |
| Moisture (%)                        | 10.00         | 9.77           | 10.29         | 9.26          |
| OM                                  | 94.89         | 95.23          | 93.89         | 93.16         |
| CP                                  | 14.03         | 13.99          | 14.06         | 14.10         |
| CF                                  | 10.40         | 11.30          | 12.84         | 11.20         |
| EE                                  | 3.11          | 6.84           | 4.61          | 5.66          |
| NFE                                 | 67.35         | 63.10          | 62.38         | 62.20         |
| Ash                                 | 5.11          | 4.77           | 6.11          | 6.84          |
| **Cell wall constituents, % on DM basis** |       |                |               |               |
| NDF                                 | 35.76         | 36.35          | 37.36         | 36.28         |
| ADF                                 | 18.92         | 19.74          | 21.14         | 19.65         |
| *Gross energy, kcal/kg DM*          | 4312          | 4521           | 4349          | 4375          |

*Gross energy (kcal/kg DM) was calculated according to Blaxter (1968), where each g of crude protein (CP) = 5.65 kcal, each g of EE = 9.40 kcal, each g of CF and NFE = 4.15 kcal**
Table 8 records the results of body weight (BW) gain, feed intake and feed conversion. A maximum \((p < 0.05)\) daily BW gain was observed for lambs fed CSM diet compared to lambs fed BCSM, SSM and SBM diets, respectively. But, no significant difference was showed in BW gain for lambs fed diets of BCSM and SSM. However, average daily gain (ADG) for lambs fed BCSM and SSM was higher \((p < 0.05)\) than those fed SBM diets.

Table 8 illustrates the feed conversion for different experimental diets. There were no significant effects were observed in feed conversion of lambs fed different diets. However, average daily gain (ADG) for lambs fed BCSM and SSM was higher \((p < 0.05)\) than those fed SBM diets.

Table 8 illustrates the effect of feeding Farafra sheep on different protein sources on rumen activity. Ruminal pH, the concentration of TVFA’s and \(\text{NH}_3\)-N were significantly different \((p < 0.05)\) with experimental treatments. Lambs that received a diet containing SSM reported the smallest pH value. On the contrary, BCSM improved significantly \((p < 0.05)\) pH value. Ruminal pH values were unchanged \((p > 0.05)\) between lambs received CSM, SBM or BCSM diets. Ruminal pH was ranged between 5.92 and 6.47.

A diet containing BCSM increased significantly \((p < 0.05)\) \(\text{NH}_3\)-N value than those fed SBM, or CSM diets. Results of TVFA’s demonstrated that the diet containing SBM was bigger in TVFA’s value in comparison with the other diets. Results also showed that 3 h post-feeding increased significantly \((p < 0.05)\) \(\text{NH}_3\)-N and TVFA’s concentrations followed by 6 h post-feeding. However, the lowest values were obtained before feeding. The highest \((p < 0.05)\) pH values were shown before feeding, while the values obtained at 3 and 6 h post-feeding were nearly similar.

Discussion
Chemical composition of protein sources: in this study, the chemical composition of mixtures varied due to the differences in the chemical composition of ingredients was used in the experimental rations. Variations in the chemical composition of different tested rations related to different portions of ingredients used in ration formulation and also related to differences in their chemical composition.

Table 5 Feed intake, nutrients digestibility and feeding value of Farafra sheep fed different experimental diets

| Item                     | Experimental diets | SE  | Sig  |
|--------------------------|--------------------|-----|------|
|                          | SBM  | BCSM | CSM | SSM |
| Av. body wt. (kg)        | 58.17 | 52.17 | 51.58 | 54.00 | 1.39 | NS |
| DM intake (g/head/day)   |       |      |     |      |
| Concentrate             | 831.00 | 928.46 | 924.01 | 936.43 | 25.71 | NS |
| Roughage                 | 239.08 | 156.39 | 212.99 | 217.74 | 22.69 | NS |
| Total                    | 1070.08 | 1084.85 | 1137.00 | 1154.17 | 30.82 | NS |
| DM intake (g/kg w0.75)   |       |      |     |      |
| Concentrate             | 14.44 | 18.00 | 18.05 | 17.35 | 0.73 | NS |
| Roughage                 | 4.05  | 3.02  | 4.24  | 4.05  | 0.41 | NS |
| Total                    | 18.49 | 21.02 | 22.29 | 21.40 | 0.91 | NS |
| Nutrients digestibility %|       |      |     |      |
| DM                       | 69.18a | 62.14b | 70.58a | 71.49a | 1.44 | * |
| OM                       | 72.20a | 64.85b | 75.05a | 74.12a | 1.47 | * |
| CP                       | 66.20b | 59.28c | 77.24a | 71.09ab | 2.23 | ** |
| CF                       | 54.14a | 49.46b | 56.82a | 48.73b | 1.44 | * |
| EE                       | 51.90c | 61.84b | 70.18a | 56.37bc | 2.65 | * |
| NFE                      | 80.89 | 79.42 | 83.47 | 83.36 | 1.04 | NS |
| Feeding value %          |       |      |     |      |
| TDN                      | 69.27 | 65.82 | 73.51 | 71.91 | 1.24 | NS |
| DCP                      | 8.05b | 7.58b | 9.67a | 8.93ab | 0.31 | * |

a, b and c: Means in the same row with different superscripts are significantly different at \((p < 0.01)\)
SE, standard error; NS, nonsignificant difference
*: Significant difference at \((p < 0.05)\), **: Significant difference at \((p < 0.01)\)
analysis (Omer et al. 2019). The chemical composition of the cottonseed meal is very good and one of the supplemental sources of protein for farm animals, especially for ruminants (Osti and Pandey 2006). Zagorakis et al. (2018) found that SBM in animal diets can be replaced by many feeds and other by-products, such as oilseeds and oilseeds cake. However, these two feed categories contain different content for CP and EE. The data recorded that SBM showed the biggest value of most amino acids compared with various protein sources. These results are rationally similar to Heuze et al. (2019) who observed that CSM protein is lower in lysine than SBM (4% vs. 6%) and generally due to the lower protein content, the total content for CSM is lower in lysine and essential AA’s. Cottonseed meal is a relatively rich source of protein (30–50%) and amino acids (He et al. 2015). Ramachandran et al. (2007) reported that CSM has around 40% protein and the fiber content of 11–13% is seemly in lysine, methionine, threonine and tryptophan. The results of the nitrogen fraction of protein sources cleared that the protein degradability of CSM was found to be 57 percent at a rumen outflow rate of 0.05 per hour (Osti and Pandey 2006).

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The feed intake of sheep fed different protein sources: the data reported that the inclusion of SBM, BCSM, CSM and SSM in ration did not significantly affect on feed intake of lambs. This result agreed with those obtained by Alves et al. (2010) who showed that when CSM substituted up to 35% of SBM there were no differences in intake. Sheep feeding on SSM recorded the highest values of concentrate and total DM intake, probably due to more palatability of SSM compared to the other sources of protein. Sheep fed on SBM showed the lowest content of concentrate intake and total DM intake; this probably due to the lower palatability of SBM compared to the other protein sources. In the same way, Baile and Forbes (1974) stated higher DMI caused by higher digestibility and less gut fill effect on reticulo-rumen (Plassance et al. 1997) or influence DMI by the composition of ingredients (Carneiro et al. 2006).

The digestibility of sheep fed different protein sources: digestibility of DM, OM, CP, CF and EE and feeding value as DCP were significantly different for diets containing different protein sources. These data disagreed with those obtained by Khorasani et al. (1990); Zinn and Depeters (1991); they showed that DM and CP digestibility of SBM were 2–10% more than canola meal in growing lambs. The CP digestibility was higher ($p < 0.05$) for CSM and SSM as compared to SBM and BCSM diets. Brown (1990) reported the same results of crude protein digestibility which may be due to high CF in CSM, compared with SSM or SBM. The results of low CF digestibility of SSM and BCSM compared to CSM and SBM may be due to the high relatively high EE content. For low nutritive value forages and fibrous by-products, CSM is a good protein supplement due to its high protein digestibility.

### Table 6 Nitrogen balance of Farafra sheep fed different experimental diets

| Item                              | Experimental diets | SE | Sig          |
|-----------------------------------|--------------------|----|--------------|
|                                   | SBM    | BCSM | CSM   | SSM   |
| Nitrogen balance (g/head/day)     |        |      |        |       |
| Nitrogen intake                   | 20.80  | 22.19 | 22.70  | 23.08 |
| Fecal nitrogen                    | 7.01b  | 9.02a | 5.19c  | 6.69b |
| Urinary nitrogen                  | 12.32a | 9.31b | 5.46c  | 10.37b|
| Digestible nitrogen               | 13.79c | 13.17c| 17.51a | 16.39b|
| Nitrogen retention                | 1.47c  | 3.86bc| 12.05a | 6.02b |
| N retent./N intake (%)            | 7.07c  | 17.40b| 53.08a | 26.08b|
| N retent./digest. N (%)           | 10.66c | 29.31b| 68.82a | 36.73b|
| Nitrogen balance (mg/kg w0.75)    |        |      |        |       |
| Nitrogen intake                   | 360.52 | 429.99| 444.13 | 427.80|
| Fecal nitrogen                    | 120.34b| 174.58a| 101.52b| 124.11b|
| Urinary nitrogen                  | 212.96a| 180.10a| 106.69b| 192.07a|
| Digestible nitrogen               | 240.18 | 255.41| 342.61 | 303.69|
| Nitrogen retention                | 27.22c | 75.31bc| 235.92a| 111.62b|
| N retent./N intake (%)            | 7.55c  | 17.51b| 53.12a | 26.09b|
| N retent./digest. N (%)           | 11.33c | 29.49b| 68.86a | 36.75b|

a, b and c: Means in the same row with different superscripts are significantly different at ($p < 0.01$)

SE, standard error; NS, nonsignificant difference

*Significant difference at ($p < 0.05$), **Significant difference at ($p < 0.01$)
(Brown and Pate 1997; Bonsi and Osuji 1997). Significant effects of CM, CSM and SBM on DM, CP and CF digestibility were recorded by Khan et al. (1997) in growing Afghani lambs.

Nitrogen utilization of sheep fed different protein sources: sheep fed diet contained BCSM significantly (p < 0.05) highest fecal nitrogen then SBM and SSM compared with CSM. This looks like another result (Fahmy

### Table 7 Water balance of Farafra sheep fed different experimental diets

| Item                          | Experimental diets | SE  | Sig |
|-------------------------------|--------------------|-----|-----|
|                               | SBM    | BCSM | CSM | SSM |
| Water balance (mL/head/day)   |        |      |     |     |
| Drinking water                | 6604.33| 6125.00| 6550.00| 6900.00| 264.62| NS |
| Feed water                    | 118.33 | 117.33| 129.33| 119.33| 3.41  | NS |
| Urinary water                 | 1074.67a| 583.33c| 475.00c| 750.00b| 82.14 | * |
| Fecal water                   | 413.00a| 342.00b| 158.33c| 200.67bc| 37.04 | * |
| Balance                       | 5234.99| 5317.00| 6046.00| 6068.66| 285.96| NS |
| Water balance (mL/day/kg w0.75) |       |      |     |     |
| Drinking water                | 114.17| 117.73| 128.30| 127.87| 5.78  | NS |
| Feed water                    | 2.07  | 2.30  | 2.50  | 2.23  | 0.21  | NS |
| Urinary water                 | 18.87a| 11.30ab| 9.30b| 13.90ab| 1.46  | * |
| Fecal water                   | 7.03a | 6.60ab| 3.03b| 3.70b | 0.60  | ** |
| Balance                       | 90.34 | 102.13| 118.47| 112.50| 5.90  | NS |
| Drinking water (L/kg DM intake/day) | 6.17 | 5.60 | 5.76 | 5.98 | 0.21 | NS |

a, b and c: Means in the same row with different superscripts are significantly different at (p < 0.01)
SE, standard error; NS, nonsignificant difference
* Significant difference at (p < 0.05). **Significant difference at (p < 0.01)

### Table 8 Growth performance, feed intake and feed conversion of Farafra sheep fed different experimental diets

| Item                          | Experimental diets | SE  | Sig |
|-------------------------------|--------------------|-----|-----|
|                               | SBM    | BCSM | CSM | SSM |
| No. of animals                | 5  | 5 | 5 | 5 |
| Feeding period (day)          | 66 | 66 | 66 | 66 |
| Initial body wt. (kg)         | 43.50 | 41.50 | 41.50 | 39.00 | 1.66 | NS |
| Final body wt. (kg)           | 55.40 | 55.40 | 57.00 | 52.60 | 1.34 | NS |
| Body wt. gain (kg/period)     | 11.90b | 13.90b | 15.50a | 13.60b | 0.47 | * |
| Av. weight gain (g/h/day)     | 180.30c | 210.61b | 234.85a | 206.06b | 7.36 | * |
| Feed intake (g DM/head/days)  |      |      |     |     |
| Concentrate                   | 1035.32| 1162.50| 1158.20| 1170.46|
| Roughage                      | 308.25| 242.35| 300.00| 295.70|
| Total                         | 1343.57| 1404.85| 1458.20| 1466.16|
| Feed conversion               | 7.45 | 6.67 | 6.21 | 7.12 | 0.24 | NS |
| Economic evaluation           |      |      |     |     |
| Income                        | 380.80| 444.80| 496.00| 435.20| – | – |
| Feed cost                     | 195.10| 203.98| 211.55| 212.92| – | – |
| Revenue                       | 185.70| 240.82| 284.45| 222.28| – | – |
| Efficiency (L.E./h)           | – | 55.12 | 98.75 | 36.58 | – | – |
| Relative efficiency (%)       | 100 | 129.68 | 153.18 | 119.70 | – | – |

a, b and c: Means in the same row with different superscripts are significantly different at (p < 0.01)
SE, standard error; NS, nonsignificant difference
*Significant difference at (p < 0.05), **Significant difference at (p < 0.01). Because group feeding was used in the feeding trial, the data of feed intake did not subject to statistical analysis.
El-Nomeary et al. Bull Natl Res Cent (2021) 45:40

El-Nomeary et al. 1992; Phillips and Rao 2001) who reported that a great amount of CP diets probably result in higher fecal and urinary N excretion. Williams et al. (1991) discussed that improving N due to the result from accretion post-ruminal AA’s absorption that is more than to the tissue needs or ruminal or post-ruminal absorption of ammonia. Although CSM increased digestible nitrogen (DN), nitrogen retention (NR), NR% of nitrogen intake (NI) and NR% of DN than the other protein sources, the lowest value of N retention was reported with SBM; these probably connect with increased the total nitrogen excretion of SBM diet. Sheep that received diet contained CSM showed a better value of NR and NR % of DN, and the other groups were positive nitrogen balance (NB). Lambs fed diets with protein sources were reported a positive NB (4.1–6.4 g/h/days) compared to those lambs fed an un-supplemented diet (Ward et al. 2008). On the other hand, better digestion and NB were observed with improving the protein concentration in the rations of growing lambs. The NB or NR probably refers to normal CP synthesis (Fahmy et al. 1992). Caton et al. (1988) recorded that rations supplemented with high CSM as a protein source with lambs improving NR compared with low CSM diets.

Water balance of sheep fed different protein sources: A nonsignificant difference (p < 0.05) was observed between groups in drinking or water balance, due to the positive relationship between water intake and DMI (NRC 1996). Allen (1997) noticed that when water was not perfect mixing with ruminal contents the effects of higher water intake would tend to lessen. On the other hand, Galyean and Defoor (2003) concluded that improved water intake probably nearly shift site absorption of acid (i.e., intestines vs. rumen) and thereby not highly alter load of total metabolic acid; however, the temporal pattern of acid absorption probably be altered to reach the metabolic acid load again evenly much time.

Growth performance of sheep fed different protein sources: The final BW of lambs fed CSM diet was higher (p < 0.05) than lambs fed SBM, BCSM and SSM diets. Cottonseed meal can replace sesame or groundnut meal as the protein source in diets for rams with a similar daily weight gain of 76.3 g/days and a better feed conversion ratio of 0.85 and also cottonseed meal used in diets for growing lambs gave the same performance as other oilseed meals such as groundnut, sesame and soybean (Ahmed and Abdalla 2005). Yunus et al. (2004) reported that CSM has higher weight gain than sunflower meal in buffalo calves feeding. They also concluded that CSM resulted in greater weight gain than sunflower meal because of interactions in the diet (i.e. degree of undegradable protein in the rumen or contents of lignin). Variation in gain probably extent of rumen degradation of different protein sources which in turn, leads to variable AA’s supply (Urbaniaik 1995), while the diet containing CSM recorded the highest feed conversion. However, no significant differences were shown in feed intake for lambs fed different sources of protein (Suliman and

| Item                          | Experimental diets | Mean | SE | Sig |
|-------------------------------|--------------------|------|----|-----|
| pH                            |                    |      |    |     |
| 0 h                           | 6.60               | 6.67 | 6.70 | 6.17 | 6.54A | 0.30 **  |
| 3 h                           | 6.10               | 6.27 | 6.20 | 5.77 | 6.09B | 0.27 **  |
| 6 h                           | 6.40               | 6.47 | 6.40 | 5.83 | 6.28B | 0.34 **  |
| Mean                          | 6.37a              | 6.47a| 6.43a| 5.92b|      | 0.35 **  |
| Ammonia-N, mg/100 mL          |                    |      |    |     |
| 0 h                           | 14.00              | 16.67| 11.00|      | 15.77 | 14.36C | 2.92 **  |
| 3 h                           | 25.70              | 36.67| 23.53|      | 35.26 | 30.29A | 6.77 **  |
| 6 h                           | 19.33              | 23.76| 23.00|      | 23.00 | 22.27B | 3.38 **  |
| Mean                          | 19.68b             | 25.70a| 19.18b|      | 24.68a| 5.04 *  |
| Total VFA’s, meq/100 mL       |                    |      |    |     |
| 0 h                           | 13.00              | 8.73 | 10.07|      | 9.73  | 10.38C | 2.05 **  |
| 3 h                           | 45.33              | 29.00| 31.33|      | 30.67 | 34.08A | 7.11 **  |
| 6 h                           | 22.53              | 20.13| 21.10|      | 20.40 | 21.04B | 2.49 **  |
| Mean                          | 26.95a             | 19.29b| 20.83b|      | 20.27b| 6.58 *  |

a and b: Means in the same row with different superscripts between treatments are significantly different. A, B and C: Means in the same columns with different superscripts between times are significantly different.
SE, standard error; NS, *: Significant difference at (p < 0.05). **: Significant difference at (p < 0.01)

Table 9 Rumen fluid parameters of Farafra sheep fed different experimental diets
In the same way, Negesse et al. (2001) cottonseed meal supplementation did not affect rumen fermentation and its higher intake might be attributed to a positive relationship between DM and CP intake.

Ruminal parameters of sheep fed different protein sources: Weimer (1996) reported that the values of 6.0–7.0 were the optimal pH for microbial growth and fiber digestion. If the pH value was below 6.0 the digestion of cellulose was limited (Mould and Ørskov 1984). But the optimum ruminal pH was ranged between 5.8 and 6.3 for cellulolytic bacteria (Staples et al. 1984). The same values were recorded in our study. Kajananpruthipong and Leng (1998) reported that ruminal protozoa, fungi and bacteria were related by the levels of NH$_3$–N concentration in the rumen. Results showed that 3 h post-feeding decreases significantly ($p < 0.05$) pH than that at 0 h, while improved NH$_3$–N and TVFA’s concentrations. These data were in the same way as Khorsheed (2008), Taie et al. (2005) showed that the decrease in pH with sampling time post-feeding was probably because of increased fermentation after feeding, in which they were connected with a decrease in rumen pH. The peak of NH$_3$–N at 3 h post-feeding is probably due to protein degradation and hydrolysis of NPN substances (Reddy et al. 1989), or because of the deamination of ruminal AA’s (Chandra et al. 1991). On the other hand, El-Shafie et al. (2007) concluded that the relationship between NH$_3$–N and TVFA’s concentrations reflects the ruminal pH and fungi effect probably connected to the optimum ruminal utilization of the dietary energy and positive ruminal fermentation because pH value is a major factor affecting the rumen fermentation and its functions.

Conclusion

It can be concluded that using cottonseed meal led to improve digestibility, nitrogen balance, growth performance and improved feed conversion, as well as reduce the costs of feeding. Cottonseed meal is the best protein source to feed sheep as compared to other sources of protein such as soybean meal, black cumin seed meal and sesame seed meal. Palatability and availability make cottonseed meal a very common protein source. It can replace 100% soybean meal by cottonseed meal in sheep ration when economics is to be considered. Future study is needed to determine the proper level used of these protein sources in rations for sheep.

Abbreviations

AA: Amino acid; ADF: Acid detergent fiber; ADG: Average daily gain; AOAC: Official methods of analysis; BCSM: Black cumin seed meal; CF: Crude fiber; CFM: Concentrate feed mixture; CP: Crude protein; CSM: Cottonseed meal; DCP: Digestible crude protein; DE: Digestible energy; DM: Dry matter; DMI: Dry matter intake; DN: Digestible nitrogen; EE: Ether extract; NB: Nitrogen balance; NFE: Nitrogen-free extract; NH3-N: Ammonia nitrogen; NDF: Neutral detergent fiber; NR: Nitrogen retention; NRC: National Research Council; OM: Organic matter; SBM: Soybean meal; SPSS: Statistical package for social sciences; SSM: Sesame seed meal; TDN: Total digestible nutrients; TVFAs: Total volatile fatty acids.

Acknowledgements

Our deep thanks are due to the Sheep Experiment Unit, Research and Production Station, National Research Centre for saving facilities that make this work possible.

Authors’ contributions

YAA co-operated in fieldwork, collected samples, prepared data, statistical analysis, written the manuscript, laboratory analysis and follow-up the publication with the journal. HHA co-operated in the plan of work, revision of the manuscript and written the manuscript. MMS co-operated in the revision of the manuscript. AAA co-operated in fieldwork, prepared data and collected samples and the revision of the manuscript. FMS co-operated in written and revision manuscript. All authors read and approved the final manuscript.

Funding

All authors equally shared in financing the cost of the research paper.

Availability of data and materials

*Not applicable* for this study.

Ethics approval and consent to participate

*Not applicable* for this study.

Consent for publication

*Not applicable* for this study.

Competing interests

The authors declare that they have no competing interests.

Received: 3 February 2020 Accepted: 3 January 2021 Published online: 12 February 2021

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