Nutrient Cycling in Agroecosystems

Unconventional feeds for small ruminants in dry areas have a minor effect on manure nitrogen flow in the soil-plant system

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Abstract:	In dry areas, unconventional feeds are increasingly used for mitigating feed shortages and rangeland degradation. We evaluated how feeding sheep diets containing olive leaves, saltbush leaves and olive cake affects manure quality compared to a barley straw based diet. Soil incubation and plant growth experiments were carried out to measure soil nitrogen (N) mineralization and N uptake by barley plants and to calculate N flow through the feed-animal-soil-plant system. Fresh feces, composts consisting of feces, urine and straw, and ammonium sulfate fertilizer were mixed with soil at rate of 90 mg N kg-1 soil dry matter. Comparisons were made with non-amended soils (control) and soils amended with fresh olive cake applied at 90 and 22.5 mg N kg–1 soil dry matter, respectively. The latter treatment enabled investigation of the effect of passage of olive cake through the digestive tract of sheep on N availability and phenol transformation. Applying fresh olive cake and feces, except the saltbush leaf derived feces, resulted in a net N immobilization. All composts resulted in net N mineralization, although not significantly different from the 0N control soil. Barley growing in soils with amendment that caused N immobilization took up less N than barley growing on the 0N treatment. Reduction in N uptake was most pronounced after amendment with fresh-olive cake. Treatments with net mineralization increased barley N uptake over the 0N treatment with 2 to 16% of N applied being taken up. Dietary composition had a minor effect on N fertilizer value of either feces or compost, but feces N alone was not an efficient N source.
Response to Reviewers:	Unconventional feeds for small ruminants in the dry areas have a minor effect on manure nitrogen flow in the soil-plant system General reply: The authors thank the reviewers for their very pertinent comments and corrections. Reviewer 1: The authors have responded to previous reviewer comments and

suggestions and made very commendable improvements to their manuscript. There are some minor typographical and editorial changes that need to be made in the Abstract, Introduction Materials and methods sections. In the latter section, the calculations are a little confusing to follow and I have taken the liberty of inserting paragraph breaks to clarify the sections to which the equations belong. If these paragraph breaks are not in the correct places them rearrange as required.
Authors: all the comments and suggestions given were considered and included in the text.
Reviewer 1: The reporting of the results has improved. However there are still a number of instances when the authors refer to differences (eg lines 225, 255, 287, 319) or similarities (line 258) that do not exist based on the statistical results shown. Please fix these errors (noted in track changes in the text) as the interpretation of the data is in question. Also inconsistencies occur between the data reported in the text and that in the tables and figures on a number of occasions.
Authors: all the inconsistencies in the results were corrected and checked thoroughly with the data reported in the table. Please find the answers to the different raised questions in the comments (track changes).
Reviewer 1: In the Discussion section, the authors often repeat results, usually as an introductory sentence for a paragraph. The text needs to be re-written so that it does not read as the Results section. Again the authors report results as different or similar that aren't based on the statistical analyses.
Authors: All the small introductions in the discussion part that they are results have been deleted. The different paragraphs in the discussion part have been reformulated accordingly.
Reviewer 1: This is particularly so with the first sentence of the Conclusions, which calls into question the conclusions that are being drawn. I have also rearranged the Conclusions to improve the flow and connectedness of the sentences.
Authors: thank you for the correcting the conclusion. The authors explained in the comment the first sentence of the conclusion. The NUE in there was related to the plant experiment (NUE of barley plant).
Reviewer 1: A number of references in the bibliography haven't been abbreviated. Please check that these meet the Journal's guidelines. Also I couldn't find one reference in the text. However I may have missed it, so please confirm.
Authors: Some of the references were indeed not abbreviated but some others did not have an abbreviation. E.g. Agronomie, Rangelands, they do not have an official abbreviation.
Reviewer 1: Figures and Tables:
Figure 1 looks good. The legends are clear and the graphs are easy to read.
Authors: Thank you.
Reviewer 1: Figure 2 is much improved and easier to read and understand. I would suggest numbering each graph so that they can be referred to in the text individually rather than the reader having to figure out which graph is being referred to. The Legend is only required once, particularly as it did not print fully above all graphs. Above Figure 2 a is fine. Delete the legend above the other graphs. Rearrange the columns in graphs 2a and 2b so that fecal N is between N retention and Urine N and between Unaccounted N and Urine N respectively. This is to make the arrows to the feces amended N box easier to read. I have made some additional changes to the figure. Note that the x-axis labels for Figures 2b and 2c did not print out on my copy, which made understanding the 'Unaccounted' and the 'Lost' and 'Retained' data difficult. Make sure that the whole figure fits on a page.
Authors: All the suggested modifications were taken into consideration in order to improve the figure.
Reviewer 1: The captions to the tables are much clearer now. Some typographical changes have been made to the tables to improve readability.
Authors: Thank you.
Reviewer 1: The manuscript has improved vastly. However the authors have to ensure that their reporting of results takes into consideration the statistical output rather than whether or not the data looks different. The research and results are worthwhile, the interpretation needs to be improved.
My comments and suggestions are included in the Word document, using Track Changes or by inserting comments throughout the text.
Authors: Thank you for all your efforts to help us improving our manuscript. All your suggestions were considered and most of them included in the text, tables and figures. Please find them in the attached file, the manuscript with track changes.

Nutrient Cycling in Agroecosystems

Unconventional feeds for small ruminants in dry areas have a minor effect on manure nitrogen flow in the soil-plant system

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Abstract In dry areas, unconventional feeds are increasingly used for mitigating feed shortages and rangeland degradation. We evaluated how feeding sheep diets containing olive leaves, saltbush leaves and olive cake affects manure quality compared to a barley straw based diet. Soil incubation and plant growth experiments were carried out to measure soil nitrogen (N) mineralization and N uptake by barley plants and to calculate N flow through the feed-animal-soil-plant system. Fresh feces, composts consisting of feces, urine and straw, and ammonium sulfate fertilizer were mixed with soil at rate of 90 mg N kg^{-1} soil dry matter. Comparisons were made with non-amended soils (control) and soils amended with fresh olive cake applied at 90 and 22.5 mg N kg^{-1} soil dry matter, respectively. The latter treatment enabled investigation of the effect of passage of olive cake through the digestive tract of sheep on N availability and phenol transformation.

Applying fresh olive cake and feces, except the saltbush leaf derived feces, resulted in a net N immobilization. All composts resulted in net N mineralization, although not significantly different from the 0N control soil. Barley growing in soils with amendment that caused N immobilization took up less N than barley growing on the 0N treatment. Reduction in N uptake was most pronounced after amendment with fresh-olive cake. Treatments with net mineralization increased barley N uptake over the 0N treatment with 2 to 16% of N applied being taken up. Dietary composition had a minor effect on N fertilizer value of either feces or compost, but feces N alone was not an efficient N source.

Keywords Nitrogen efficiency · Manure · Compost · Saltbush · Olive by-product · Awassi sheep

Introduction

Under-utilized feeds like fodder shrubs and agro-industrial by-products are gaining attention in Mediterranean countries. Their importance lies in their potential role in overcoming animal feed shortages, mitigating rangeland degradation due to overgrazing, and in coping with increasing prices for traditional concentrate feeds and opportunity costs for growing forage on arable land. In dry areas mixed crop-livestock systems with ruminants are common. The main crops are cereals, olive trees (the Mediterranean countries represent 98% of the global area planted with olive trees, Molina-Alcaide and Nefzaoui 1996), vegetables and cotton. In addition, in attempting to re-vegetate degraded and dry rangelands of the dry steppe areas, different saltbush species have been planted because of their tolerance of drought and salinity (Louhaichi and Tastad 2010). Previous studies showed that olive leaves and saltbush leaves constitute suitable forage resources during periods of shortage (e.g., Molina-Alcaide and Yáñez-Ruiz 2008; El Shaer 2010; Abbeddou et al. 2011a).

As a byproduct of the agro-food industry, 2.9 million tons per year of a ligno-cellulosic organic material results from olive oil production (Sansoucy 1985; Sellami et al. 2008). This residue is called olive cake. Studies have addressed its use as an energy source (Oktay 2006), as an animal feed (e.g. Molina-Alcaide and Yáñez-Ruiz 2008; Abbeddou et al. 2011b) or investigated composting before applying it as a soil amendment (e.g. Cayuela et al. 2004; Sellami et al. 2008). Still, it is mostly discarded as a waste and disposed in a non-sustainable manner in the olive oil production areas, where it constitutes an environmental issue because of its composition, large quantities and seasonality (Benitez et al. 2004; Roig et al. 2006).

Soil degradation is increasing in many Mediterranean countries (Benitez et al. 2004). Restoring soil fertility is closely linked to increasing soil organic matter and managing nutrient cycling (Frossard et al. 2006; Sommer et al. 2011). Animal manure is a valuable nitrogen (N) source for crops, although with lower immediate N use efficiency (NUE) by crops than water soluble mineral N (Langmeier et al. 2002; Bosshard et al. 2009). However, organic matter (OM) content and microbial activity are higher in soils regularly receiving animal manure than in soils receiving exclusively mineral fertilizers (Fliessbach et al. 2007; Sommer et al. 2011). It is, however, unclear whether amendments resulting from different feeding options affect soil microbial activity and the NUE by the crops. Previous research indicated that the type of feed affects the availability of N contained in cow feces (e.g., dairy cow diets fed in the Midwest USA; Powell et al. 2006), and the phosphorus (P) content of manure (e.g., sheep fed with bush straw and millet stover; Sangaré et al. 2002) and subsequently, on nutrient uptake by plants from soils amended with these manures (Sangaré et al. 2002; Powell et al. 2006). The availability of manure N to crops could even be more dependent on feed type when feeds contain secondary plant metabolites like phenols. Phenols present in certain feeds may inhibit N availability and nutrient utilization in the animal (Abbeddou et al. 2011b) as well as N mineralization when the feed is applied directly to soils (Benitez et al. 2004; Cayuela et al. 2004). Feeding animals with plants rich in phenols also affected nutrient cycling by reducing N content in urine

(Powell et al. 1994). Phenols bind to proteins to form indigestible complexes, which result in less excretion of N in urine and, additionally, a shift from soluble to insoluble N in the feces (Powell et al. 1994). In contrast, Rufino et al. (2006) did not find a correlation between the phenolic content of the manure and N mineralization.

The present study tested the following hypotheses: (i) Feeding sheep with certain unconventional feeds (olive leaves, saltbush leaves and olive cake) or the conventionally used barley straw affects manure quality in terms of content and availability of nutrients. (ii) Fresh feces, composts (prepared from these feces, urine and barley straw), and fresh olive cake differ in their effect on soil N mineralization and N use by barley plants. These hypotheses were tested in soil incubation and plant experiments in the greenhouse by measuring N mineralization, N use by barley plants, and N flow through the feed-animal-soil-plant system.

Materials and methods

Feeding treatments and manure collection

Four groups of six growing Awassi lambs each were fed diets where large proportions of a traditional barley straw based diet (control) were replaced either by sun-dried leaves (with small twigs) from olive trees (*Olea europaea* L.), or from saltbush shrubs (*Atriplex halimus*), or air-dried olive cake from the first pressing containing also hulls and kernels. The animals received 1.1 kg dry matter (DM) feed per day that covered their maintenance requirements. The composition of the diets is shown in Table 1. The N contents of the four complete diets were in the range of 24.9 ± 0.9 g kg^{-1} DM, with the barley straw, olive leaves, saltbush leaves and olive cake contributing 5.0, 10.7, 18.5 and 6.2 g N kg^{-1} DM, respectively. After a 15-day adaptation to the diets, the animals were held in metabolic crates over 10 days during which all feces (separated from urine) were collected, pooled per group and stored at − 20°C until use. Urine was collected for 2 days, also pooled per group and stored for two weeks at +4°C until use. Further details about the sheep study are given in Abbeddou et al. (2011a, b).

Manure composting procedure

A method of composting manure at a laboratory scale was adapted from Thomsen (2000) with slight modifications. For the barley straw, olive leaves and olive cake diets, mixtures of feces, urine and barley straw were prepared in a fresh weight ratio of 10:2:1. As the feces from the saltbush diet had higher moisture content, more straw was added (fresh weight ratio of 10:2:2). These ratios resulted in moist mixtures approaching the maximum urine holding capacity of straw bedding which has accumulated feces in barns housing sheep. The excreta from the barley straw, olive leaves, saltbush leaves and olive cake diets mixed with barley straw gave initial ratios of feces, urine and straw N of 10.6:2.5:1, 17.7:2.4:1, 3.2:0.4:1; 11.3:2.6:1, respectively. Each feces-urine-straw mixture was composted separately under aerobic conditions by placing 52 kg of each mixture (56 kg for the saltbush diet) in a jute bag with open mesh. These bags were placed on wooden pallets, in a room where the temperature was maintained between 20 to 25°C for 18 weeks. During the composting process, the temperature was recorded weekly. Likewise, the moisture content was adjusted to 450–650 g kg^{-1} DM after having determined the water content, and the mixture was turned manually with a spade, before being placed back into the bags. A subsample of the each compost mixture was collected weekly and stored at – 20°C until analysis. The bags were weighed at the end of the composting.

Soil and preliminary plant experiment

Soil with a low mineral N content was sampled from the upper 20 cm of a field under a chickpea-cereal rotation after chickpea harvesting at the International Center for Agricultural Research in the Dry Areas (ICARDA), Syria. The soil (Table 2), classified as very fine, montmorillonitic, thermic, Chromic Calcixerert (Ryan et al. 1997), was collected using a mechanical soil auger. It was sieved through a mesh (< 2 mm) to obtain a homogenous soil, kept moist and stored at room temperature until the soil incubation and plant growth experiments were established.

In a preliminary experiment, the amount of N amendment to be used for the incubation and the plant experiments was determined. A plant NUE response curve to mineral N fertilization was measured from applications of 0, 45, 90, 135, 270 and 405 mg N kg^{-1} soil after 7 weeks of growth. Ammonium sulfate was used as the mineral amendment, barley as the test crop and each dose was repeated in quadruplicate. The

NUE (see equation 1, below) was plotted against the amount of ammonium sulfate applied. The maximal NUE was found at a dose of 90 mg N kg^{-1} soil, which was then used in the following experiments.

Soil response to the different amendments

The experimental soil was mixed manually with N-free nutrient solutions in order to avoid macro- and micronutrient deficiencies. Mineral compounds supplied to the soil were (mg kg^{-1} soil DM): KCl (85.8), KH_2PO_4 (395.4), $CaCl_2$ (55.1), $MgSO_4$ (162.2), $ZnSO_4$ (4.4), Na_2MoO_4 (0.20), Fe chelate (6.2), H_3BO_3 (5.7), $MnSO_4$ (6.2), $CuSO_4$ (7.9) and $CoSO_4$ (0.48). The soil was mixed with amendment (feces from the four diets, four composts and olive cake; 9 treatments), or ammonium sulfate fertilizer in quantities equivalent to 90 mg N kg^{-1} dry soil. Fresh olive cake was also tested at 22.5 mg N kg^{-1} soil. Control treatment (0N) soil received only the N-free nutrient solutions but no amendment. The amendment, fertilizer and control soil mixtures were transferred to a total of 288 plastic pots (12 treatments × 4 replicates × 6 sampling times) each with a volume of 100 ml. The water content was brought to 350 g kg^{-1} DM of the soil. The pots were stored at 25ºC in a growth chamber in the dark. Lost water was replaced with distilled water every second day by restoring the initial weight of the pots. Four pots from each treatment were withdrawn at every sampling time, i.e., after 1 day, and 1, 2, 4, 7 and 12 weeks. Soils were analyzed immediately afterwards.

Plant response to the experimental amendments

The effect of the amendments on N uptake was assessed by using barley (*Hordeum vulgare L.*, var. Harmal) as a test plant. For this, 48 pots (12 treatments × 4 replicates) were filled with 900 g soil DM prepared and amended as previously described for the soil incubation experiment, except that ammonium sulfate was split in two doses, namely 30 mg N kg^{-1} soil DM at sowing and 60 mg N kg^{-1} soil DM at tillering. Barley seeds treated with a fungizide (Vitavax 200 FF, Chemtura, Italy) were sown at a rate of 10 seeds pot^{-1} with a distance of 2 cm between seeds. The seeded pots were watered to 350 g kg^{-1} DM soil and transferred to a greenhouse set to 14 h daylight at 25°C and 10 h darkness at 15°C. The pots were watered every other day as described for the soil incubation experiment. After 7 weeks of growth, which was at the end of the vegetative stage, the shoots were cut at 1 cm above the soil surface and dried immediately at 65°C for 48 h. The pots were emptied and the roots were washed from the soil under a water-jet, and then also dried at 65°C for 48 h.

Chemical analyses

Feces, compost samples collected during the 18 week composting period, and olive cake were analyzed by standard methods (AOAC 1997) for DM and OM (AOAC index no. 942.05), total N (AOAC 977.02) and, total C with a C/N analyzer (AOAC index no. 977.02). Total phenols were measured in the fresh feces, mature compost and fresh olive cake using the Folin Ciocalteu method (Makkar 2003). Feces and composts were also analyzed for their mineral N content (NO_3^- extracted by deionized water and NH_4^+ extracted with 2 *M* KCl, Bremner and Keeney 1965, and analyzed by titration with diluted H_2SO_4, Keeny and Nelson 1982) as well as total P and K after wet digestion using nitric and perchloric acids (AOAC 935− 13) and analyzed using a UV-Vis spectrophotometer (model U-2000, Hitachi, Tokyo, Japan; AOAC 965− 17) for P and a digital flame analyzer (A. Gallenkamp and Co., London, UK; AOAC 969.23) for K. Compost samples were mixed with water at a ratio of 1:5, and pH and electrical conductivity (EC) of the extract were measured using a pH meter (pH M82, Radiometer, Copenhagen, Denmark) and a conductivity cell (CDM83, Radiometer, Copenhagen, Denmark), respectively. At every sampling date, samples of the incubated soils were analyzed for pH, EC and mineral N (NO_3^- and NH_4^+ extracted as described by Bremner and Keeney 1965, and analyzed by sulfuric acid titration, Keeny and Nelson 1982). Only values recorded at the beginning and the end of the experiment are shown in tables, except for the time course of mineral N. In addition, soil samples collected at the end of the incubation experiment were analyzed for their OM (chromic acid titration method, Walkley 1947), extractable P (sodium bicarbonate extraction, Olsen et al. 1954) and K (ammonium acetate extraction, Richard 1954) contents. Barley shoots and roots were analyzed for DM and total N content by NCS elemental analyzer (Flash EA 1112 Series NCS analyzer, Thermo Electron Corporation, Waltham, USA).

Calculations and statistical analysis

From the incubation experiment, net N mineralization was calculated as the difference between the total mineral N at the end of the incubation time (12 weeks) and that at the beginning of the experiment (day 1). The net mineralization over the 0N control is the net mineralization of the amendments tested minus the net mineralization of the control (0N).

The NUE by barley for each of the amendments was computed by the difference method (Harmsen 2003) as:

$$\text{NUE (\%)} = 100 \times (NP_a - NP_{0N})/N_{ai} \quad (1)$$

where, NP_a = total N uptake by amended plants (mg pot^{-1}), NP_{0N} = total N uptake by unamended plants (mg pot^{-1}) and N_{ai} = total amount of the amendment N applied (mg pot^{-1}).

Selected data from the associated animal experiment (Abbeddou et al. 2011a, b), including N intake, N excretion in feces and urine and the resulting N retention in the body were used in this study to estimate the N flows through the feed-animal-soil-plant system. Nitrogen cycling efficiency was calculated for each diet and was standardized to 1000 g N intake by the animals as follows.

The N excreted in feces and urine was expressed as a proportion of N intake:

$$\text{Fecal N (g kg}^{-1}\text{ N intake)} = 1000 \times (\text{N excreted in feces / N intake}) \quad (2)$$

$$\text{Urine N (g kg}^{-1}\text{ N intake)} = 1000 \times (\text{N excreted in urine / N intake}) \quad (3)$$

$$\text{Excreted N (g kg}^{-1}\text{ N intake)} = \text{Fecal N} + \text{Urine N} \quad (4)$$

$$\text{Body N retention (g kg}^{-1}\text{ N intake)} = 1000 - \text{Excreted N} \quad (5)$$

N retained (fecal N and urine N in uncomposted mixture) from excretion collection, and unaccounted for in composting (i.e. not included in our experimental design), respectively, was calculated according to the formulas given by Rufino et al. (2006), except that they were all expressed based on 1000 g N intake:

$$\text{Fecal N in uncomposted mixture (g kg}^{-1}\text{ N intake)} = 1000 \times (\text{N feces used / N intake}) \quad (6)$$

$$\text{Urine N in uncomposted mixture (g kg}^{-1}\text{ N intake)} = 1000 \times (\text{N urine used / N intake}) \quad (7)$$

$$\text{Unaccounted N (g kg}^{-1}\text{ N intake)} = \text{Excreted N} - (\text{Fecal N in uncomposted mixture} + \text{Urine N in uncomposted mixture}) \quad (8)$$

At the final stage of composting, N efficiency was calculated as the amount of N in compost as a proportion of N contained in the fresh feces and urine used in the mixture for composting, and in relation to N intake:

$$\text{N retained during composting (g kg}^{-1}\text{ N intake)} = 1000 \times ((\text{N in compost / Fecal N and Urine N in uncomposted mixture})) / \text{N intake} \quad (9)$$

N lost during composting (g kg^{-1} N intake) = N in uncomposted mixture – N in compost (10)

For plant N uptake, N flow was calculated as the amount of N in the plant as a proportion of the amount of N applied to the soil with the amendment (feces or compost) again reported as g kg^{-1} N intake of the animal.

Plant N uptake (g kg^{-1} N intake) = NUE (%)/100 × amendment N (g kg^{-1} N intake) (11)

where amendment N is either the fecal N (when feces are used as treatment) or N retained during composting (result from equation 9 when compost is used as treatment).

The GLM procedure (SAS version 9.2, SAS Institute Inc., Cary, NC) was used for analysis of variance of the data from the incubation and the plant experiments, with amendment as a fixed factor. Means were compared with the Tukey test at $P < 0.05$. The tables give arithmetic means, standard errors of the means and *P*-values. Changes in total mineral N in the incubation experiment were analyzed by repeated measurement analysis using the GLM procedure of SAS. Treatment, time and the interaction between the two were considered as fixed factors. Figure 1 gives least square means, standard errors and *P*-values.

Results

Composition of the amendments and changes during the composting process

The mean OM content of the amendments (feces, compost and fresh olive cake) ranged from 683 to 900 g kg^{-1} DM (Table 3), and was lowest for the saltbush leaf compost, and highest for olive cake derived feces, uncomposted mixture and compost derived from this diet. The C/N ratio was on average 25 for fresh feces and 13 for compost and as high as 39 for the fresh olive cake. Accordingly, feces from the olive cake treatment also had a particularly high C/N ratio. Mineral N content (NH_4-N and NO_3-N) tended to be lower in the composts than the corresponding fresh feces, except for compost from barley straw diet. The K content was higher in the composts than in the fresh feces. Likewise P content was higher in the composts, except for saltbush leave compost.. On average, total phenol content in the composts, ranging between 1 to 2 g kg^{-1} DM, was lower than in the feces and the fresh olive cake. Total phenol content in the composts represented on average only 33% of the content in feces from barley straw, olive leaf, saltbush leaf and olive cake diets.

The temperature in the compost bags increased from an average of 7 to 55°C during the first three days (data not shown). The differences in chemical composition between the uncomposted mixtures at the beginning of the composting procedure were largely present at the end of the composting, except for pH and EC. The initial weight of the uncomposted mixture of 52 kg (56 kg for the saltbush treatment) corresponded to 16, 20, 13 and 23 kg on DM basis for composts prepared from the excreta of the sheep fed the barley straw, olive leaf, saltbush leaf and olive cake diets, respectively. The corresponding DM losses during composting were 687, 450, 461 and 565 g kg^{-1} initial DM (data not shown), with OM losses of 729, 507, 539 and 602 g kg^{-1} initial OM, respectively. The decline in OM content was most pronounced with the saltbush leaf and the barley straw treatments (Table 3). The decline in OM was lowest with compost from the olive cake diet and intermediate with the compost from the olive leaf diet. Total N content per unit of DM increased during composting, and the C/N ratio of the four composts decreased from a range of 20 to 32 to between 12 and 17. Contents of both NH_4-N and NO_3-N decreased during the composting process except for the barley straw treatment. While the pH did not change during composting of the excreta obtained from olive leaf and olive cake fed sheep, it decreased for the barley straw diet and increased with the saltbush leaf diet. In all composts, except that produced from feeding the saltbush leaf diet, EC decreased with time.

Soil response to the different amendments

In general, all feces and compost amendments increased the OM contents of the soils, which amounted to greater than 1 g kg^{-1} soil DM on average compared to 0N soil at the end of the incubation period (Table 4). This increase was about 5 times higher with the amendment with fresh olive cake when provided at the full N dose level. The quantities of OM added per kg soil with the amendments were in the range of 3 to 5 g with fresh feces, 2 to 3 g with composts and 8 or 2 g with fresh olive cake provided at the full or the 1/4 N dose.

Feces, except that from the saltbush leaf treatment, and fresh olive cake (at any dose) resulted in lower mineral N levels than in the 0N soil during the 12 week incubation time (amendment × week, $P < 0.001$), i.e., caused immobilization of N (Figure 1). In contrast, composts and feces from the saltbush leaf treatment increased mineral N content over the 0N control, although for NH_4-N not significantly (Figure 1, Table 4). Mineral N (NH_4-N and NO_3-N) in the 0N soil increased from 16 to 27 mg kg^{-1} soil DM from 1 day to 12 weeks incubation (Figure 1). Compared to this change, differences in mineralization caused by the application of the organic amendments were small (Table 4). Net mineralization in compost treatments was not significantly different from the 0N treatment while fresh feces amendments caused significant immobilization, except for the saltbush leave treatment. Amendments of the saltbush leaf treatments (feces and compost) and compost of barley straw treatment resulted in significant increases in extractable K compared to the 0N soil. Finally, only amendment with saltbush leaf feces significantly increased soil P compared to the 0N soil.

The non N amended soil had a pH of 8.4 at the start of the soil incubation experiment. Only few amendments affected pH significantly, which were barley straw feces, olive cake compost and fresh olive cake at any dose (Table 4). Changes in pH during the incubation period were small with only an increase or decrease of up to 0.2 units. The EC was 0.16 mS cm^{-1} in the 0N soil at the beginning, but increased to 0.27 mS cm^{-1} at the end of the incubation period. Changes compared to the 0N soil ranged between 0.02 and 0.15 mS cm^{-1} directly upon amendment application, and between -0.06 and 0.27 mS cm^{-1} at the end of the incubation period. However, EC was significantly higher after incubation of soil amended with mineral fertilizer N, feces and compost of the saltbush leaf and the barley straw treatments, and olive cake compost.

Plant response to the experimental amendments

Total (shoot plus root) barley DM yield was 2.5 g for plants growing on the 0N soil (Table 5). The addition of mineral N fertilizer caused a 1.7-fold increase in yield. Likewise, N yields per pot in shoot and root biomass were increased by mineral N addition. In contrast, the addition of the fresh feces of any origin did not affect or even tended to decrease DM yield compared to 0N, except for the saltbush leaf treatment, which increased shoot and total DM yields by 1.3 and 1.2-fold levels of 0N, respectively. Composts prepared from the barley straw and saltbush leaf treatment increased total DM yields by 1.4- and 1.2-fold respectively. The low and the high dose of fresh olive cake reduced total DM yield by 0.5- and 0.3-fold compared to 0N. The N uptake of barley followed the same trend as DM yield, resulting in a negative NUE for three of the fresh feces (i.e., those derived from the barley straw, olive leaf and olive cake diets) and fresh olive cake applied at both doses. The NUE was positive with the composts but low, ranging from 2% (olive cake diet derived compost) to 16% (barley straw diet derived compost).

N flow in the feed-animal-soil-plant system

The tested feeds resulted in retention of between 158 and 239 g N kg^{-1} N intake in the growing Awassi lambs (Figure 2a). When compared to barley straw, fecal N excretion increased after feeding an olive leaf diet, while intake of saltbush leaf diet tended to increase urinary N. Due to the small amount of urine required for the compost mixtures, between 317 and 581 g urinary N kg^{-1} N intake (32 to 58% of the total excreted N) were unaccounted for in the analysis of N flows (Figure 2b). Composting resulted in very variable N losses from less than 20% with the saltbush compost to more than half of the initial N available with the barley straw compost (Figure 2c). Overall, compost amendment resulted in a positive but small uptake of N by the barley plants (6 to 28 g kg^{-1} N intake), with the majority remaining unused in the soil (Figure 2d). In contrast, the direct application of fresh feces as an amendment resulted in negative N uptake by the barley plants, leading to proportions of unused N ranging between 204 and 425 g kg^{-1} N intake (Figure 2e).

Discussion

In the farming systems typically practiced in dry areas, most of the feces and urine is excreted in the rangelands with the rest deposited in barns where animals are kept during the night and when the weather conditions are extreme (extreme heat at noon, rain or storms). In the study region, excreta deposited in the barns are collected as a mixture of feces and straw bedding which has absorbed some of the urine. This manure is collected usually four times a year, and composted on-farm in heaps. The compost is then used in the cropping system either alone or in combination with a mineral fertilizer (urea) (personal information from the farmers). The traditional forage, consisting of cereal straw, has been replaced occasionally by locally available, low-cost forage alternatives especially during periods of shortage (Abbeddou et al., 2011a). The different chemical composition of the alternative forages and concentrates affect the nutritive value and N use efficiency at the animal level (Abbeddou et al. 2011a, b). No information however is available about the implications of their use on overall N cycling in a mixed crop/livestock system.

Changes in amendment characteristics during composting as affected by the diet fed to sheep

The rate of OM degradation during composting depends on the content of readily biodegradable compounds present in the compost (Sellami et al. 2008). This suggests that the OM in the feces of the sheep fed the saltbush leaf diet should be very degradable, as had been observed for most of the dietary OM in the digestive tract (Abbeddou et al. 2011a). The compost derived from the olive cake diet was higher in OM than other composts. Probably in particular lignin limited its degradation in the digestive tract as feces from olive cake diet contained 630 g neutral detergent fiber kg^{-1} DM, of which 170 g kg^{-1} DM were lignin (Abbeddou et al. 2011b). Additionally, an adverse action on digestion due to the phenols present in this feed had been assumed. The low degradability of OM in the compost derived from the olive cake was consistent with the low OM digestibility of dietary olive cake in the animal. It therefore appears that the properties controlling digestibility of feeds in the animal also control degradation of feces subsequent to excretion, such as during composting (Rufino et al. 2006). The C/N ratios of most composts were in the recommended range of < 12 (Cayuela et al. 2004) for a stable OM of the compost, while it was higher for the compost from the olive cake treatment. The NH_4^+ concentration of all composts was less than 0.40 g kg^{-1} as expected (Cayuela et al. 2004).

Nitrogen retention after composting was greater than the average of 54% reported by Thomsen (2000) for all composts except the barley straw diet derived compost (46% of N in the uncomposted mixture retained after composting compared to 75, 85 and 74% of N retained in olive leaf, saltbush leaf and olive cake derived composts; data not shown), but was in the range of 30 to 87% reported by Rufino et al. (2006).

Soil response to the different amendments

Increase in soil EC results from an increase in the electrolytes Na and K (Mekki et al. 2006), and saltbush leaves are reported to have high contents of these cations (Abbeddou et al. 2011a). An ingested excess of electrolytes is mostly excreted in urine, which explains the elevated EC of both the compost and the soil of the saltbush leaf treatment. Fresh feces from the saltbush leaf diet also had a high K content (Table 3).

Net N mineralization in compost amended soils was similar to net mineralization in the 0N soil. The N immobilization observed with all feces (except that from the saltbush leaves diet) agrees with Wichern et al. (2004), where fresh sheep and goat feces from local farms in Oman resulted in microbial N immobilization. Net N mineralization in soil treated with feces from the saltbush diet may have resulted from relatively low C/N ratio and phenol content, and high mineral N content of this feces. Mineralization of N was related to the C/N ratio of manures as found by Powell et al. (2006). Manures with C/N ratios greater than 19 caused immobilization of soil N, as was observed here with most of the feces having C/N ratios between 22 and 32, while manures with C/N ratio less than 16 caused N mineralization. Mineralization of N might have been further reduced by the presence of polyphenols, especially after amendment by fresh olive cake and feces from the olive cake and leaf diets. Although olive cake has a moderate total phenol content, the special nature of the constitutive phenols (e.g., hydroxytyrosol, tyrosol, and their glucosides) makes it highly antimicrobial (Benitez et al. 2004; Cayuela et al. 2004; Sampedro et al. 2004). Additionally, phenols may bind proteins which are then protected from microbial degradation in the soil as for the rumen (van Bruchem et al. 1999; Tiemann et al. 2009). A regression analysis of all amendments in this study showed that N mineralization was negatively related to total amendment phenol input ($r = -0.75$, $P = 0.012$).

Plant response to the experimental amendments

In the present study 77% of mineral fertilizer N was recovered in the barley roots and shoots, which is in the range of a pot study with ryegrass (Langmeier et al. 2002) but higher than N recovery of a barley crop growing in lysimeters (36% on coarse sand to 49% on sandy loam soil; Thomsen et al. 1997) and recoveries in crops reported from field studies which usually are in the range of 20 to 50% (Crews and Peoples 2005). The NUE for animal manure is usually lower than that of mineral N, because it is mainly composed by organic N forms which are not readily available to plants. In a pot study six harvests of ryegrass recovered 25 to 30% of N applied with fresh cow feces (Langmeier et al. 2002), while wheat growing in microplots in the field recovered 10% of N applied with sheep feces (Bosshard et al. 2009). Also in a microplot field study, barley at maturity (grain and straw) recovered 6% of N-15 added with sheep feces (Jensen et al. 1999). In this experiment, the application of fresh feces reduced plant productivity compared to the 0N soil, resulting in negative NUE except for the amendments from the saltbush leaf treatments. This could be explained by the higher amounts of mineral N (NH_4-N, NO_3-N) found in the feces from the saltbush leaf treatment than the other fecal amendments, and organic fecal N compounds that can be easily mineralized (Bosshard et al. 2011)

In contrast to the feces, a positive NUE was observed for all composts resulting in N recoveries in barley of 2 to 16% of the N applied. This is in the range found for fresh feces in other studies but much less than that reported for slurry composed of all of the feces and urine excreted (Langmeier et al. 2002; Bosshard et al. 2009). The NUE of both olive feed containing diets was lower than those from the barley straw and the saltbush based diet. In their review, Rufino et al. (2006) reported that NUE from composted manure for maize crops in Africa ranged between 3 and 49%, with NUE being higher when urine was included in the composted manure. Nitrogen recovery from compost was higher than from fresh feces probably because soil amended with compost had higher total mineral N than the soil amended with feces. Also, compost had lower phenol contents than feces; the latter also because of dilution by urine and straw. Urine N mostly consists of urea which is a readily available N source for plants (Bosshard et al. 2009). However, part of the urea N gets hydrolyzed during composting (Thomsen 2000; Wichern et al. 2004) and lost in the form of ammonia (Rufino et al. 2006). The high pH in composts and soils may further have intensified ammonia volatilization. As no ^{15}N-labeling was done in the present study, N derived from feces and urine, and their respective contribution to barley N uptake (Bosshard et al. 2009), cannot be separated.

The NUE of fresh olive cake was negative, indicating that the N in olive cake was not directly available for the plants and that olive cake amendment reduced the availability of soil N. The inhibitory effect on plant productivity was less pronounced when less olive cake was added (1/4 of the N dose used in the other treatments). This suggests that feeding olive cake may be relatively more advantageous than use of fresh olive cake as an amendment. When olive cake passes through the rumen and the digestive track, phenols are either metabolized or diluted when mixed with other feed ingredients resulting in excreta that might be less inhibitory to plant growth.

Amendment N not taken up by the crops can remain in the soil (Powell et al. 2006) or be lost from the soil-plant system. Studies using ^{15}N labeling show that with organic amendments often more N is recovered in the soil than with mineral fertilizer N (Sørensen and Thomsen 2005; Gardner and Drinkwater 2009). This is also supported by the studies by Langmeier et al. (2002) and Bosshard et al. (2009), where about 60% of N applied with fresh cattle or sheep feces remained in the soil. This N can become available to subsequent crops (Schröder et al. 2005) although at low release rates. Accordingly, only around 3.3 and 1.5% of feces N were recovered during the two following years in the study by Bosshard et al. (2009). The difference method used in this study does not allow measurement of amendment N recovery in the soil compared to the background soil N.

Nitrogen flow in the feed-animal-soil-plant system as affected by unconventional feeds

The NUE of a mixed crop/livestock system is the result of N conversion at the animal level and the apparent N recovery from soil amended by animal excreta (van Bruchem et al. 1999). The calculated N conversion efficiency at the animal level for growth (body N retention, Figure 2a) was in the range of 150 to 250 g N kg^{-1} N intake reported by van Bruchem et al. (1999). All diets had similar N conversion efficiencies despite the lowest N intake associated with the saltbush leaves and olive cake diets (data taken from Abbeddou et al. 2011a, b). Nitrogen not converted into animal products (milk or body weight gain) is excreted with urine and feces, but also the allocation of excretory N to feces and urine clearly varied between treatments due to phenol content in the diets. As stated above, proteins bound to phenols are indigestible complexes excreted in feces where they constitute the insoluble N fraction (Powell et al. 1994). Based on the allocation of the excretory N, the lowest feces N as a proportion of N intake was found with the saltbush diet ($P < 0.001$) and the highest with diets containing olive-derived feeds. Not all N excreted by the animals was included in manure N flow. Thus a large proportion of N excreted, i.e., urinary N with high plant availability, remained unaccounted for (32 to 58% of the total excreted N; Figure 2b). Our experiment mimicked a situation where all feces would be collected, but where the urine not absorbed by the straw would have been lost. Farming systems with sheep in Syria are quite variable (Rischkwosky et al. 2004), and in turn the collection and use of animals excreta differs between farms and production areas. Likewise, straw is a limited valuable resource, and compost preparation and application may economically not always be viable as straw is in demand as livestock fodder (Sommer et al. 2011). Thus its use as animal bedding in barn is not always applied, especially in seasons with low barley straw yields.

In this study, the NUE of composts and of feces was tested when used as amendments for crop production. As discussed above, NUE by barley of both forms was low or even negative and a large fraction of applied N remained unused (Figures 2d and 2e). The question arises whether the feces and urine would be better used on the rangelands than for crop production. We have no exact data on the proportion of N deposited in the rangelands compared with in the barns. Rufino et al. (2006) estimated that less than 50% of total N excreted can be collected in typical mixed grazing-barn system by smallholders in the sub-Saharan Africa. Based on experimental data of excreta collection and N loss during composting in central Kenya (Lekasi et al. 2001), Rufino et al. (2006) assumed that less than 10% of N in the excreta would be efficiently used in the mixed crop-livestock system in Kenya. Nitrogen deposited in the rangeland can be taken up by plants, be retained in the soil or be lost. Plant N uptake is limited to the short (up to three months in the dry areas of Syria) growing season when water availability sustains plant growth and nutrient uptake. During the six month-long dry period, particularly urine N deposited on the alkaline soils of the study area may largely be lost by ammonia volatilization. For instance, Vallis et al. (1985) reported a loss of 46% of urine N when cattle urine was applied on pasture during the dry season in Australia. Under these conditions, N cycling could be increased when a higher proportion of N is excreted with feces because feces N takes longer time to be mineralized and therefore the risk of ammonia volatilization is lower (Powell et al. 1994; Rufino et al. 2006). Based on this, diets containing olive-derived feeds seemed more efficient than the other diets, especially the saltbush leaf based diet which resulted in the highest proportion of N excreted in urine. Still it has to be shown whether N from olive cake and olive cake derived feces will be available in subsequent years or whether the compositional limitations prevent its release even longer.

Conclusions

Providing unconventional feeds to sheep only slightly affected the NUE by barley of N applied with feces or composts produced from feces, urine and straw, with NUE being higher after treatments with composts than with feces. The NUE obtained in the pot experiment was low or even negative, leaving most of the N applied unused. Feeding olive cake was more efficient as some nutrients were supplied when fed to the sheep increasing overall N cycling efficiency compared with its direct application as soil amendment. Labeling studies with ^{15}N to separate unused N into N retained in soil and N losses are needed, as well as long-term plant experiments under field conditions to test residual amendment N effects. Although reflecting the limited collection practiced on farm, an important proportion of urine N was not considered in the present study. When the intention is to use amendments consisting of animal excreta strategically for crop production and in the growing season, it seems worthwhile to capture as much urine as possible and, therefore, to change excreta storage and application practices.

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Legend of the figures

Figure 1 Changes in total mineral N (mg kg^{-1} DM) of unamended soil (0N), soil amended with mineral N (ammonium sulfate), soil amended with feces from sheep fed barley straw, olive leaves, saltbush leaves or fresh olive cake (A); soil amended with compost made from the feces and urine of sheep fed these four diets (B); and soil amended with fresh olive cake at 90 mg N kg^{-1} and 22.5 mg N kg^{-1} (C). Amendment, $P < 0.001$; week, $P < 0.001$; amendment × week, $P < 0.001$, SEM at treatment level = 1.9

Figure 2 Nitrogen flows in a feed-animal-soil-plant system, where barley straw, olive leaf, saltbush leaf and fresh olive cake diets are fed to Awassi sheep, and the feces and some urine are used to make compost to grow barley plants. Nitrogen flows from the feces to soil is also compared, but most of the urine N is unaccounted for in this experimental system.

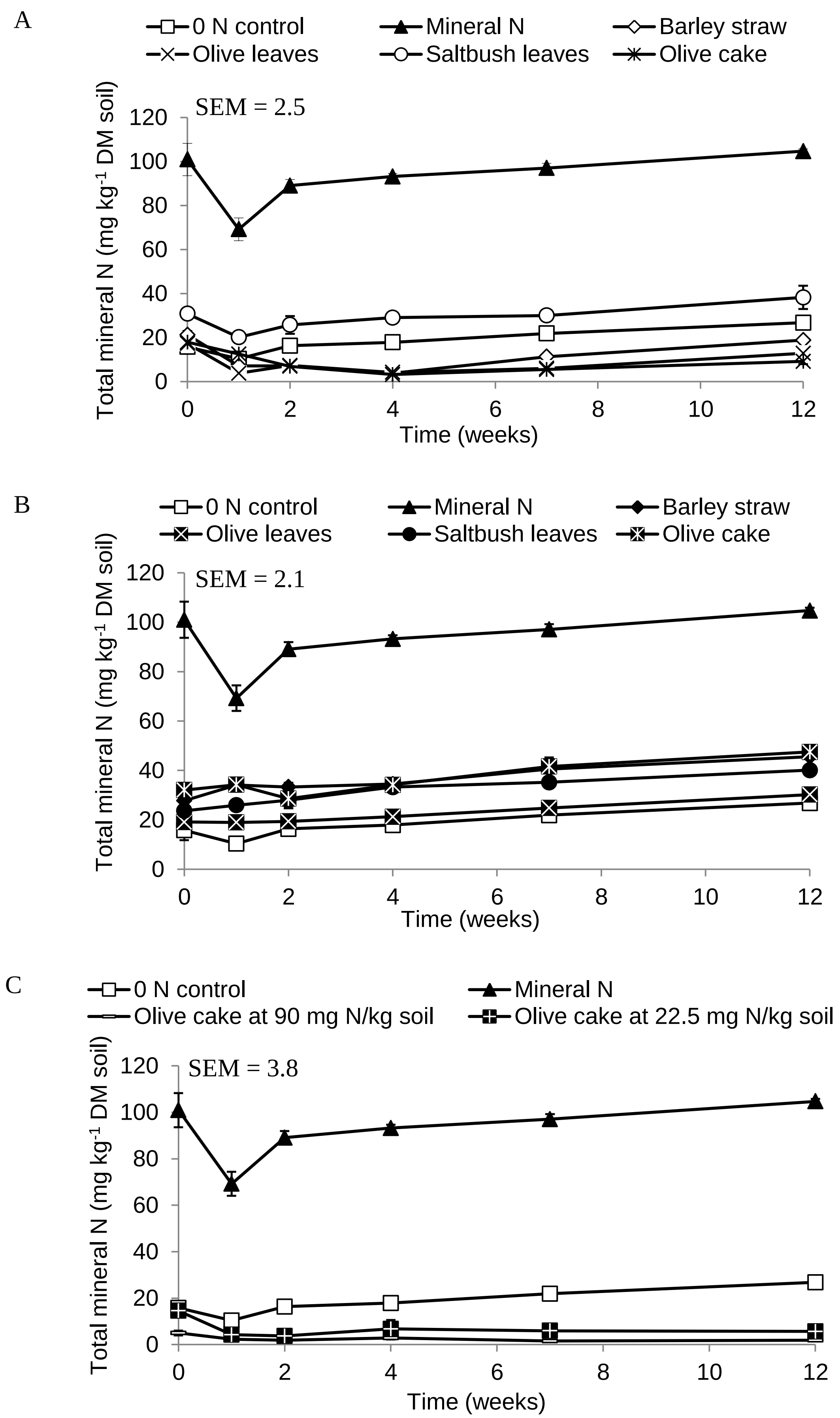
A
0 N control
Mineral N
Barley straw
Olive leaves
Saltbush leaves
Olive cake
SEM = 2.5
Total mineral N (mg kg-1 DM soil)
Time (weeks)
B
0 N control
Mineral N
Barley straw
Olive leaves
Saltbush leaves
Olive cake
SEM = 2.1
Total mineral N (mg kg-1 DM soil)
Time (weeks)
C
0 N control
Mineral N
Olive cake at 90 mg N/kg soil
Olive cake at 22.5 mg N/kg soil
SEM = 3.8
Total mineral N (mg kg-1 DM soil)
Time (weeks)

Figure 1

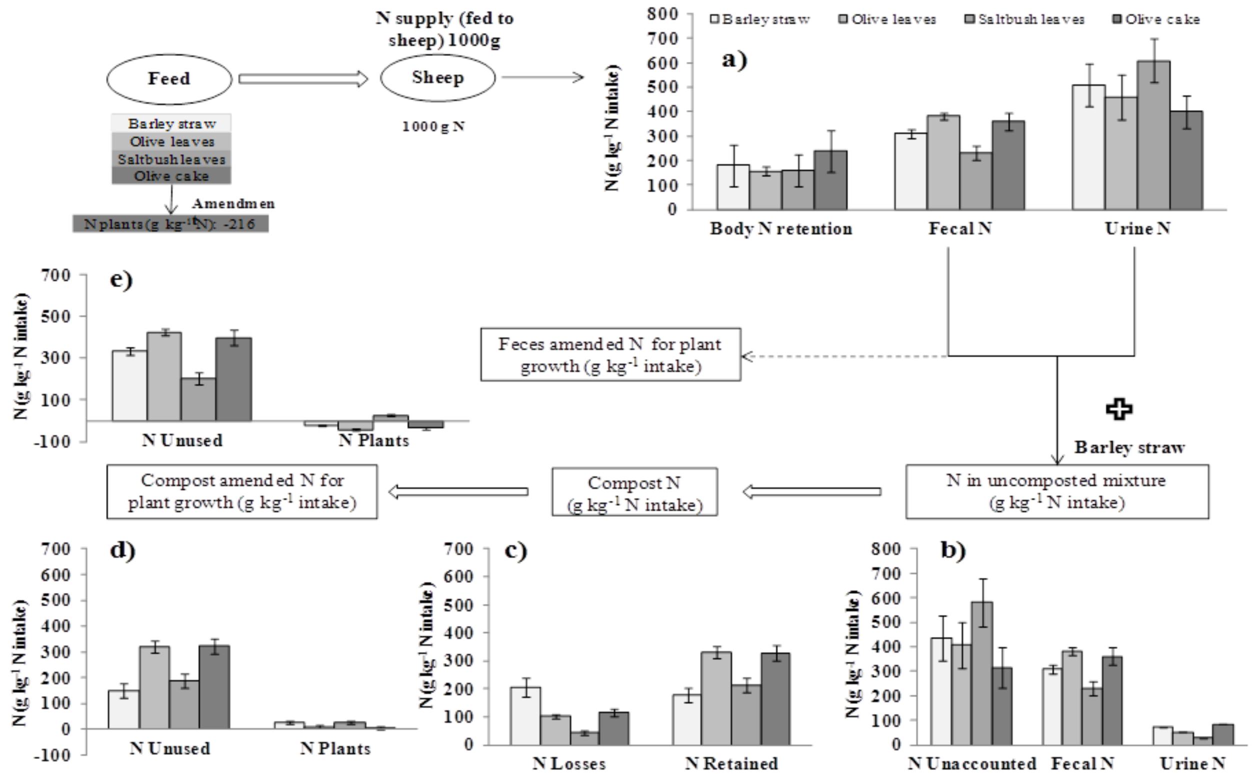
N supply (fed to sheep) 1000g
Feed
Sheep
1000 g N
Barley straw
Olive leaves
Saltbush leaves
Olive cake
Amendmen
N plants (g kg-1 N): -216
Barley straw
Olive leaves
Saltbush leaves
Olive cake
a)
N (g kg-1 N intake)
Body N retention
Fecal N
Urine N
e)
N Unused
N Plants
Feces amended N for plant growth (g kg-1 intake)
Barley straw
N in uncomposted mixture (g kg-1 N intake)
Compost N (g kg-1 N intake)
Compost amended N for plant growth (g kg-1 intake)
d)
c)
b)
N Losses
N Retained
N Unaccounted
Fecal N
Urine N

Figure 2

Table 1 Description of feeding trials and composition of the diets fed to fat-tailed Awassi sheep

Feeding trial	Diet	Feed component description (g kg^{-1} DM)	Nutrients content in feed
Barley straw replaced with olive leaves or saltbush leaves.$^{\Phi}$)	Barley straw	716 g barley straw, 176 g cotton seed meal, 88 g molasses, and 20 g mineral-vitamin mix. 16 g urea kg^{-1} DM.	DM: 1100 g CP: 159 g kg^{-1} DM Metabolisable energy: 7.59 MJ kg^{-1} DM
	Olive leaf	716 g olive leaves, 44 g barley grain, 44 g wheat bran, 88 g cotton seed meal, 88 g molasses, and 20 g mineral-vitamin mix. 11 g urea kg-1 DM.	DM: 1100 g CP: 157 g kg^{-1} DM Metabolisable energy: 7.85MJ kg^{-1} DM
	Saltbush leaf	716 g saltbush leaves, 44 g barley grain, 132 g wheat bran, 88g molasses, and 20 g mineral-vitamin mix. 2 g urea kg^{-1} DM.	DM: 1100 g CP: 159 g kg^{-1} DM Metabolisable energy: 7.59 MJ kg^{-1} DM
Concentrate replaced with olive cake. ψ	Olive cake	490 g barley straw, 340 g olive cake, 100 g cotton seed meal, 50 g molasses, 20 g mineral-vitamin mix.19g urea kg^{-1} DM.	DM: 1100 g CP: 147 g kg^{-1} DM Metabolisable energy: 5.50 MJ kg^{-1} DM

$^{\Phi}$For details see Abbeddou et al. (2011a)
Ψ For details see Abbeddou et al. (2011b)

Table 2 Properties of the soil used in soil and plant growth experiments.

Soil classification	Texture (g kg^{-1})			Composition (g kg^{-1} DM)					pH	Electrical conductivity (mS cm^{-1})
	Clay	Silt	Sand	Organic matter	Total N	Mineral N	Olsen-P[Ψ]	Extractable K[ω]		
Montmorillonitic, thermic, Chromic Calcixerert	539	283	178	10.4	0.78	0.01	0.012	0.27	8.2	0.271

[Ψ]Extraction with sodium bicarbonate (Olsen et al. 1954)
[ω]Extraction with ammonium acetate (Richards 1954)

Table 3 Physico-chemical characteristics of the amendments used for soil and pot experiments. The amendments consist of fresh feces excreted by sheep fed barley straw, olive leaf, saltbush leaf or olive cake diets; uncomposted mixtures of feces (excreted by sheep fed the four diets), urine and barley straw; compost made from the mixtures; and fresh olive cake. The mean and standard error of three analytical replicates are given.

	Organic matter	Kjeldahl N	NH_4-N	NO_3-N	Total K	Total P	Total phenols	C/N	pH	Electrical conductivity
	(g kg^{-1} dry matter)									(mS cm^{-1})
Fresh feces										
Barley straw	870±0.0	18±5.3	1.0±0.10	0.9±0.03	2.4±0.7	7.2±1.2	5.0±0.7	24±7.4	nd	nd
Olive leaves	841±0.5	24±0.3	0.7±0.03	0.6±0.03	1.8±0.6	8.7±1.5	6.1±0.3	21±0.2	nd	nd
Saltbush leaves	762±6.6	21±0.2	1.7±0.09	2.0±0.05	10.1±3.8	12.2±1.7	3.2±0.2	22±0.2	nd	nd
Olive cake	897±0.6	15±0.2	0.4±0.03	0.3±0.02	2.6±0.6	6.0±0.8	3.1±0.9	32±0.4	nd	nd
Uncomposted mixture before composting										
Barley straw	843±0.8	23±3.1	5.8±0.40	5.1±0.40	nd	nd	nd	20±2.2	8.7±0.10	4.6±0.07
Olive leaves	845±0.8	25±1.3	3.8±0.38	3.5±0.27	nd	nd	nd	20±5.9	8.4±0.00	3.7±0.00
Saltbush leaves	798±1.4	19±0.8	0.7±0.20	1.0±0.20	nd	nd	nd	25±6.5	8.6±0.10	4.1±0.00
Olive cake	900±1.4	15±0.2	2.5±0.31	2.3±0.45	nd	nd	nd	32±0.4	8.5±0.07	3.3±0.07
Compost after 18 weeks of composting										
Barley straw	731±0.9	34±0.2	0.03±0.001	4.3±0.40	23.7±2.1	11.4±0.5	1.9±0.2	12±0.1	8.1±0.07	4.3±0.10
Olive leaves	757±2.5	35±0.4	0.00±0.001	0.4±0.02	15.3±1.1	13.0±0.8	1.8±0.2	12±0.1	8.4±0.10	3.1±0.10
Saltbush leaves	683±1.1	30±0.8	0.06±0.002	0.7±0.01	20.2±2.6	9.5±1.0	0.9±0.4	12±0.3	9.0±0.07	5.4±0.07
Olive cake	824±3.4	25±0.3	0.00±0.002	0.4±0.03	12.5±0.5	7.8±0.2	1.2±0.2	17±0.2	8.5±0.00	1.5±0.00
Fresh olive cake	864±0.2	13±1.8	Nd	Nd	8.3±0.7	0.8±0.2	4.1±0.5	39±0.6	nd	nd

nd = not determined

Table 4 Mean organic matter, mineral N (NH_4-N, NO_3-N), calculated net N mineralization and net N mineralization over 0N control, nutrient content (ammonium acetate extractable K, sodium bicarbonate extractable "Olsen"-P), pH, and electrical conductivity of unamended soil (0N), soil amended with mineral N (ammonium sulfate), soil amended with fresh feces excreted by sheep fed barley straw, olive leaf, saltbush leaf or fresh olive cake diets, soil amended with compost (90 mg N kg-1) made with feces from sheep fed these diets or soil amended with fresh olive cake (90 or 22.5 mg N kg-1) at the beginning (Initial) and after 12 weeks (Final) of incubation (n=4).

Treatment	Organic matter (g kg^{-1} soil DM)	NH_4-N (mg kg^{-1} soil dry matter (DM))		NO_3-N (mg kg^{-1} soil DM)		Net N mineral-ization	Net N mineral-ization over control	Extractable-K (g kg^{-1} soil DM)	Olsen-P (g kg^{-1} soil DM)	pH		Electrical conductivity (mS cm^{-1})	
	Final	Initial	Final	Initial	Final			Final	Final	Initial	Final	Initial	Final
0N control	11^{cd}	1.5^{c}	0.1^{b}	14^{e}	27^{e}	11.0^{ab}		0.48^{b}	0.04^{b}	8.4^{ab}	8.2^{c}	0.16^{d}	0.27^{fg}
Mineral N	11^{d}	78.9^{a}	0.4^{ab}	22^{bcd}	104^{a}	3.9^{bcd}	– 7.2^{bcd}	0.48^{b}	0.04^{b}	8.3^{abc}	8.2^{c}	0.25^{ab}	0.54^{a}
Fresh feces - 90 mg N kg^{-1}													
Barley straw	12^{bcd}	2.0^{c}	0.7^{ab}	19^{cde}	18^{f}	– 2.4^{cde}	– 13.4^{cde}	0.48^{b}	0.04^{ab}	8.2^{cd}	8.3^{ab}	0.26^{ab}	0.33^{de}
Olive leaves	12^{bcd}	2.1^{c}	0.6^{ab}	15de	12^{g}	– 4.4^{de}	– 15.4^{de}	0.50^{ab}	0.05^{ab}	8.4^{a}	8.3^{ab}	0.18^{cd}	0.24^{fgh}
Saltbush leaves	12^{bcd}	13.5^{b}	0.3^{ab}	17^{cde}	38^{d}	7.4^{abc}	– 3.6^{abc}	0.54^{a}	0.06^{a}	8.4^{ab}	8.3^{bc}	0.25^{ab}	0.40^{b}
Olive cake	12^{b}	0.9^{c}	0.8^{a}	17^{cde}	8^{gh}	– 8.8^{e}	– 19.8^{e}	0.49^{ab}	0.05^{ab}	8.3^{ab}	8.3^{ab}	0.24^{bc}	0.21^{h}
Composts - 90 mg N kg^{-1}													
Barley straw	11^{bcd}	0.7^{c}	0.4^{ab}	27^{ab}	45^{bc}	17.8^{a}	6.8^{a}	0.54^{a}	0.05^{ab}	8.3^{abc}	8.2^{c}	0.27^{ab}	0.34^{cd}
Olive leaves	12^{bcd}	0.7^{c}	0.5^{ab}	19^{cde}	30^{e}	11.1^{ab}	0.13^{ab}	0.50^{ab}	0.05^{ab}	8.3^{ab}	8.3^{ab}	0.24^{bc}	0.29^{ef}
Saltbush leaves	12^{bc}	0.8^{c}	0.5^{ab}	23^{bc}	40^{cd}	16.4^{a}	5.4^{a}	0.54^{a}	0.05^{ab}	8.3^{bcd}	8.3^{ab}	0.31^{a}	0.39^{bc}
Olive cake	12^{b}	0.8^{c}	0.4^{ab}	31^{a}	47^{b}	15.4^{a}	4.4^{a}	0.50^{ab}	0.04^{b}	8.1^{d}	8.2^{c}	0.29^{ab}	0.36^{bcd}
Fresh olive cake													
90 mg N kg^{-1}	15^{a}	1.0^{c}	0.4^{ab}	4^{f}	1^{i}	– 3.1^{cde}	– 14.1^{cde}	0.52^{ab}	0.04^{b}	8.1^{d}	8.3^{bc}	0.24^{bc}	0.23^{gh}
22.5 mg N kg^{-1}	12^{bc}	1.5^{c}	0.4^{ab}	13^{e}	5^{hi}	– 8.9^{e}	– 11.6^{cde}	0.51^{ab}	0.04^{b}	8.2^{cd}	8.4^{a}	0.25^{ab}	0.23^{gh}
SEM	0.2	3.16	0.04	1.1	3.9	1.5	1.5	0.004	0.001	0.11	0.01	0.01	0.01
P-value treatment	<0.001	<0.001	0.100	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Means carrying no common superscript within each column are different at $P < 0.05$ (Tukey test).

Table 5 Mean dry matter (DM), N yield and N use efficiency by barley grown on unamended soils (0N), soil amended with mineral N (ammonium N), soils amended with fresh feces excreted by sheep consuming barley leaves, olive leaves, saltbush leaves and fresh olive cake, soils amended with compost made using feces from sheep fed these diets, or soils amended with fresh olive cake (n=4)

	DM (g)			N yield (mg pot^{-1})			N use efficiency (%)$^{\psi}$		
Treatment	Shoot	Root	Total	Shoot	Root	Total	Shoot	Root	Total
0N control	1.2^{d}	1.3^{cd}	2.5^{d}	14^{c}	9^{ef}	23^{c}			
Mineral N	2.5^{a}	1.6^{ab}	4.1^{a}	62^{a}	23^{a}	85^{a}	60^{a}	17^{a}	77^{a}
Fresh feces									
Barley straw	0.7^{e}	1.0^{ef}	1.6^{e}	8^{d}	9^{efg}	17^{d}	− 7^{d}	− 1^{ef}	− 8^{d}
Olive leaves	0.5^{f}	0.8^{f}	1.4^{ef}	7de	7^{fg}	14^{d}	− 9de	− 3^{f}	− 11^{d}
Saltbush leaves	1.6^{bc}	1.4^{bc}	3.0^{c}	20^{b}	13^{bcd}	32^{b}	7^{b}	4^{bcd}	11^{b}
Olive cake	0.7^{e}	0.8^{f}	1.5^{ef}	8^{d}	7^{fg}	15^{d}	− 7^{d}	− 3^{f}	− 10^{d}
Compost									
Barley straw	1.7^{b}	1.8^{a}	3.4^{b}	21^{b}	16^{b}	36^{b}	8^{b}	8^{b}	16^{b}
Olive leaves	1.3^{d}	1.3^{cd}	2.6^{d}	15^{c}	11^{cde}	26^{c}	1^{c}	2^{cde}	3^{c}
Saltbush leaves	1.5^{c}	1.4^{bc}	3.0^{c}	19^{b}	13^{bc}	33^{b}	7^{b}	5^{bc}	12^{b}
Olive cake	1.2^{d}	1.1^{de}	2.3^{d}	15^{c}	10^{def}	25^{c}	1^{c}	1^{def}	2^{c}
Fresh olive cake									
90 mg N kg^{-1}	0.3^{g}	0.4^{g}	0.7^{g}	3^{e}	3^{h}	6^{e}	− 13^{e}	− 9^{g}	− 22^{e}
22.5 mg N kg^{-1}	0.5^{f}	0.7^{f}	1.2^{f}	6de	6^{g}	11^{d}	− 40^{f}	− 18^{h}	− 58^{f}
SEM	0.09	0.06	0.14	2.2	0.8	2.9	3.5	1.3	4.7
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Means carrying no common superscript are different at $P < 0.05$ (Tukey test).

$^{\psi}$N use efficiency relative to barley grown in the 0N control soils.

Nutrient Cycling in Agroecosystems

Unconventional feeds for small ruminants in dry areas have a minor effect on manure nitrogen flow in the soil-plant system

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Abstract In dry areas, unconventional feeds are increasingly used for mitigating feed shortages and rangeland degradation. We evaluated how feeding sheep diets containing olive leaves, saltbush leaves and olive cake ~~to sheep~~ affects manure quality compared to a barley straw based diet. Soil incubation and plant growth experiments were carried out to measure soil nitrogen (N) mineralization and N uptake by barley plants and to calculate N flow through the feed-animal-soil-plant system. Fresh feces, composts consisting of feces, urine and straw, and ammonium sulfate fertilizer were mixed with soil at rate of 90 mg N kg^{-1} soil dry matter. Comparisons were made with non-amended soils (control) and soils amended with fresh olive cake applied at 90 and 22.5 mg N kg^{-1} soil dry matter, respectively. The latter treatment ~~allowed~~enabled ~~investigating~~ investigation of the effect of passage of olive cake through the digestive tract of sheep on N availability and phenol~~ic~~ transformation.

Applying fresh olive cake and feces, except the saltbush leaf derived feces, resulted in a net N immobilization. All composts resulted in net N mineralization, although not significantly different from the 0N control soil. Barley growing i~~o~~n soils with amendment~~-~~ that caused N immobilization took up less N than barley growing on the 0N treatment. Reduction in N uptake was most pronounced after amendment with fresh-olive cake~~ amendment~~. Treatments with net mineralization increased barley N uptake over the 0N treatment with 2 to 16% of N applied being taken up. ~~Overall, d~~Dietary composition ~~showed~~ had a minor effect on N fertilizer value of either feces or compost, but feces N alone was not an efficient N source.

Keywords Nitrogen efficiency · Manure · Compost · Saltbush · Olive by-product · Awassi sheep

Introduction

Under-utilized feeds like fodder shrubs and agro-industrial by-products are gaining attention in Mediterranean countries. Their importance lies in their potential role in overcoming animal feed shortages, mitigating rangeland degradation due to overgrazing, and in coping with increasing prices for traditional concentrate feeds and opportunity costs for growing forage on arable land. In dry areas mixed crop-livestock systems with ruminants are common. The main crops are cereals, olive trees (the Mediterranean countries represent 98% of the global area planted with olive trees, Molina-Alcaide and Nefzaoui 1996), vegetables and cotton. In addition, in attempting to re-vegetate degraded and dry rangelands of the dry steppe areas, different saltbush species have been planted because of their tolerance of drought and salinity (Louhaichi and Tastad 2010). Previous studies showed that olive leaves and saltbush leaves constitute suitable forage resources during periods of shortage (e.g., Molina-Alcaide and Yáñez-Ruiz 2008; El Shaer 2010; Abbeddou et al. 2011a).

As a byproduct of the agro-food industry, 2.9 ~~Million~~ million tons per year of a ligno-cellulosic organic material results from olive oil production (Sansoucy 1985; Sellami et al. 2008). This residue is called olive cake. Studies have addressed its use ~~it~~ as an energy source (Oktay 2006), as an animal feed (e.g. Molina-Alcaide and Yáñez-Ruiz 2008; Abbeddou et al. 2011b) or investigated composting before applying it as a soil amendment (e.g. Cayuela et al. 2004; Sellami et al. 2008). Still, it is mostly discarded as a waste and disposed in a non-sustainable manner in the olive oil production areas, where it constitutes an environmental issue because of its composition, large quantities and seasonality (Benitez et al. 2004; Roig et al. 2006).

Soil degradation is increasing in many Mediterranean countries (Benitez et al. 2004). Restoring soil fertility is closely linked to increasing soil organic matter and managing nutrient cycling (Frossard et al. 2006; Sommer et al. 2011). Animal manure is a valuable nitrogen (N) source for crops, although with lower immediate N use efficiency (NUE) by crops than water soluble mineral N (Langmeier et al. 2002; Bosshard et al. 2009). However, organic matter (OM) content and microbial activity are higher in soils regularly receiving animal manure than in soils receiving exclusively mineral fertilizers (Fliessbach et al. 2007; Sommer et al. 2011). It is, however, unclear whether amendments resulting from different feeding options affect soil microbial activity and the NUE by the crops. Previous research indicated that the type of feed affects the availability of N contained in cow feces (e.g., dairy cow diets fed in the Midwest USA; Powell et al. 2006), and the phosphorus (P) content of manure (e.g., sheep fed with bush straw and millet stover; Sangaré et al. 2002) and subsequently, on nutrient uptake by plants from soils amended with these manures (Sangaré et al. 2002; Powell et al. 2006). The availability of manure N to crops could even be more dependent on feed type when feeds contain secondary plant metabolites like phenols. Phenols present in certain feeds may inhibit N availability and nutrient utilization in the animal- (Abbeddou et al. 2011b) as well as N mineralization when the feed is applied directly to soils (Benitez et al. 2004; Cayuela et al. 2004). Feeding animals with plants rich in phenols also affected nutrient cycling by reducing N content in urine

(Powell et al. 1994). Phenols bind to proteins to form indigestible complexes, which result in less excretion of N in urine and, additionally, a shift from soluble to insoluble N in the feces (Powell et al. 1994). In contrast, Rufino et al. (2006) did not find a correlation between the phenolic content of the manure and N mineralization.

The present study tested the following hypotheses: (i) Feeding sheep with certain unconventional feeds (olive leaves, saltbush leaves and olive cake) or the conventionally used barley straw affects manure quality in terms of content and availability of nutrients. (ii) Fresh feces, composts (prepared from these feces, urine and barley straw), and fresh olive cake differ in their effect on soil N mineralization and N use by barley plants. These hypotheses were tested in soil incubation and plant experiments in the greenhouse by measuring N mineralization, N use by barley plants, and N flow through the feed-animal-soil-plant system.

Materials and methods

Feeding treatments and manure collection

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Four groups of six growing Awassi lambs each were fed diets where large proportions of a traditional barley straw based diet (control) were replaced either by sun-dried leaves (with small twigs) from olive trees (*Olea europaea* L.), or from saltbush shrubs (*Atriplex halimus*), or air-dried olive cake from the first pressing containing also hulls and kernels. The animals received 1.1 kg dry matter (DM) feed per day that covered their maintenance requirements. The composition of the diets is shown in Table 1. The N contents of the four complete diets were in the range of 24.9 ± 0.9 g kg^{-1} DM, with the barley straw, olive leaves, saltbush leaves and olive cake contributing 5.0, 10.7, 18.5 and 6.2 g N kg^{-1} DM, respectively. After a 15-day adaptation to the diets, the animals were held in metabolic crates over 10 days during which all feces (separated from urine) were collected, pooled per group and stored at − 20°C until use. Urine was collected for 2 days, also pooled per group and stored for two weeks at +4°C until use. Further details about the sheep study are given in Abbeddou et al. (2011a, b).

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Manure composting procedure

A method of composting manure at a laboratory scale was ~~adopted~~ adapted from Thomsen (2000) with slight modifications. For the barley straw, olive leaves and olive cake diets, mixtures of feces, urine and barley straw were prepared in a fresh weight ratio of 10:2:1. As the feces from the saltbush diet had higher moisture content, more straw was added (fresh weight ratio of 10:2:2). These ratios resulted in moist mixtures approaching the maximum urine holding capacity of straw bedding ~~that~~ which has ~~been~~ accumulated ~~with~~ feces in barns housing sheep. The excreta from the barley straw, olive leaves, saltbush leaves and olive cake diets mixed with barley straw gave initial ratios of feces, urine and straw N of 10.6:2.5:1, 17.7:2.4:1, 3.2:0.4:1; 11.3:2.6:1, respectively. Each feces-urine-straw mixture was composted separately under aerobic conditions by placing 52 kg of each mixture (56 kg for the saltbush diet) in a jute bag with open mesh. These bags were placed on wooden pallets, in a room where the temperature was maintained between 20 to 25°C for 18 weeks. During the composting process, the temperature was recorded weekly. Likewise, the moisture content was adjusted to 450–650 g kg^{-1} DM after having determined the water content, and the mixture was turned manually with a spade, before being placed back into the bags. A subsample of the each compost mixture was collected weekly and stored at − 20°C until analysis. The bags were weighed at the end of the composting.

Soil and preliminary plant experiment

Soil with a low mineral N content was sampled from the upper 20 cm of a field under a chickpea-cereal rotation after chickpea harvesting at the International Center for Agricultural Research in the Dry Areas (ICARDA), Syria. The soil (Table 2), classified as very fine, montmorillonitic, thermic, Chromic Calcixerert (Ryan et al. 1997), was collected ~~by~~ using a mechanical soil auger. It was sieved through a mesh (< 2 mm) to obtain a homogenous soil, kept moist and stored at room temperature until the soil incubation and plant growth experiments were established.

In a preliminary experiment, the amount of N amendment to be used for the incubation and the plant experiments was determined. A plant NUE response curve to mineral N fertilization was measured from applications of 0, 45, 90, 135, 270 and 405 mg N kg^{-1} soil after 7 weeks of growth. Ammonium sulfate was used as the mineral amendment, barley as the test crop and each dose was repeated in quadruplicate. The

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NUE (see equation 1, below) was plotted against the amount of ammonium sulfate applied. The maximal NUE was found at a dose of 90 mg N kg^{-1} soil, which was then used in the following experiments.

Soil response to the different amendments

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The experimental soil was mixed manually with N-free nutrient solutions in order to avoid macro- and micronutrient deficiencies. Mineral compounds supplied to the soil were (mg kg^{-1} soil DM): KCl (85.8), KH_2PO_4 (395.4), $CaCl_2$ (55.1), $MgSO_4$ (162.2), $ZnSO_4$ (4.4), Na_2MoO_4 (0.20), Fe chelate (6.2), H_3BO_3 (5.7), $MnSO_4$ (6.2), $CuSO_4$ (7.9) and $CoSO_4$ (0.48). The soil was mixed with amendment (feces from the four diets, four composts and olive cake; 9 treatments), or ammonium sulfate fertilizer in quantities equivalent to 90 mg N kg^{-1} dry soil. Fresh olive cake was also tested at 22.5 mg N kg^{-1} soil. Control treatment (0N) soil received only the N-free nutrient solutions but no amendment. The amendment, fertilizer and control soil mixtures were transferred to a total of 288 plastic pots (12 treatments × 4 replicates × 6 sampling times) each with a volume of 100 ml. The water content was brought to 350 g kg^{-1} DM of the soil. The pots were stored at 25ºC in a growth chamber in the dark. Lost water was replaced with distilled water every second day by restoring the initial weight of the pots. Four pots from each treatment were withdrawn at every sampling time, i.e., after 1 day, and 1, 2, 4, 7 and 12 weeks. Soils were analyzed immediately afterwards.

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Plant response to the experimental amendments

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The effect of the amendments on N uptake was assessed by using barley (*Hordeum vulgare L*., var. Harmal) as a test plant. For this, 48 pots (12 treatments × 4 replicates) were filled with 900 g soil DM prepared and amended as previously described for the soil incubation experiment, except that ammonium sulfate was split in two doses, namely 30 mg N kg^{-1} soil DM at sowing and 60 mg N kg^{-1} soil DM at tillering. Barley seeds treated with a fungizide (Vitavax 200 FF, Chemtura, Italy) were sown at a rate of 10 seeds pot^{-1} with a distance of 2 cm between seeds. The seeded pots were watered to 350 g kg^{-1} DM soil and transferred to a greenhouse set to 14 h daylight at 25°C and 10 h darkness at 15°C. The pots were watered every other day as described for the soil incubation experiment. After 7 weeks of growth, which was at the end of the vegetative stage, the shoots were cut at 1 cm above the soil surface and dried immediately at 65°C for 48 h. The pots were emptied and the roots were washed from the soil under a water-jet, and then also dried at 65°C for 48 h.

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Chemical analyses

Feces, compost samples collected during the 18 week composting period, and olive cake were analyzed by standard methods (AOAC 1997) for DM and OM (AOAC index no. 942.05), total N (AOAC 977.02) and, total C with a C/N analyzer (AOAC index no. 977.02). Total phenols were measured in the fresh feces, mature compost and fresh olive cake using the Folin Ciocalteu method (Makkar 2003). Feces and composts were also analyzed for their mineral N content (NO_3^- extracted by deionized water and NH_4^+ extracted with 2 *M* KCl, Bremner and Keeney 1965, and analyzed by titration with diluted H_2SO_4, Keeny and Nelson 1982) as well as total P and K after wet digestion using nitric and perchloric acids (AOAC 935− 13) and analyzed using a UV-Vis spectrophotometer (model U-2000, Hitachi, Tokyo, Japan; AOAC 965− 17) for P and a digital flame analyzer (A. Gallenkamp and Co., London, UK; AOAC 969.23) for K. Compost samples were mixed with water at a ratio of 1:5, and pH and electrical conductivity (EC) of the extract were measured using a pH meter (pH M82, Radiometer, Copenhagen, Denmark) and a conductivity cell (CDM83, Radiometer, Copenhagen, Denmark), respectively. At every sampling date, samples of the incubated soils were analyzed for pH, EC and mineral N (NO_3^- and NH_4^+ extracted as described by Bremner and Keeney 1965, and analyzed by sulfuric acid titration, Keeny and Nelson 1982). Only values recorded at the beginning and the end of the experiment are shown in tables, except for the time course of mineral N. In addition, soil samples collected at the end of the incubation experiment were analyzed for their OM (chromic acid titration method, Walkley 1947), extractable P (sodium bicarbonate extraction, Olsen et al. 1954) and K (ammonium acetate extraction, Richard 1954) contents. Barley shoots and roots were analyzed for DM and total N content by NCS elemental analyzer (Flash EA 1112 Series NCS analyzer, Thermo Electron Corporation, Waltham, USA).

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Calculations and statistical analysis

From the incubation experiment, net N mineralization was calculated as the difference between the total mineral N at the end of the incubation time (12 weeks) and that at the beginning of the experiment (day 1). The net mineralization over the 0N control is the net mineralization of the amendments tested minus the net mineralization of the control (0N).

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The NUE by barley for each of the amendments~~,~~ ~~NUE by barley~~ was computed by the difference method (Harmsen 2003) as:

$$\text{NUE (\%)} = 100 \times (NP_a - NP_{0N})/N_{ai} \quad (1)$$

where, NP_a = total N uptake by amended plants (mg pot^{-1}), NP_{0N} = total N uptake by unamended plants (mg pot^{-1}) and N_{ai} = total amount of the amendment N applied (mg pot^{-1}).

~~The mineral fertilizer equivalent of the amendments was calculated following Muñoz et al. (2004) as:~~

~~MFE = NUE amendment / NUE mineral fertilizer × 100~~

~~(2)~~

Selected data from the associated animal experiment (Abbeddou et al. 2011a, b), including N intake, N excretion in feces and urine and the resulting N retention in the body ~~(Abbeddou et al. 2011a, b)~~ were used in this study to estimate the N flows through the feed-animal-soil-plant system. ~~Overall~~ Nitrogen cycling efficiency was calculated for each diet and was standardized to 1000 g N intake by the animals as follows.

The N excreted in feces and urine was expressed as a proportion of N intake:

$$\text{Fecal N (g kg}^{-1}\text{ N intake)} = 1000 \times \text{(N excreted in feces / N intake)} \quad (~~2~~3)$$

$$\text{Urine N (g kg}^{-1}\text{ N intake)} = 1000 \times \text{(N excreted in urine / N intake)} \quad (~~4~~3)$$

$$\text{Excreted N (g kg}^{-1}\text{ N intake)} = \text{Fecal N} + \text{Urine N} \quad (~~5~~4)$$

$$\text{Body N retention (g kg}^{-1}\text{ N intake)} = 1000 - \text{Excreted N}$$

(~~6~~5)

~~N retained or lost during excreta collection or composting was calculated according to the formulas given by Rufino et al. (2006), except that they were all expressed based on 1000 g N intake:~~N retained (fecal N and urine N in uncomposted mixture) from excretion collection, and unaccounted for in composting (i.e. not included in our experimental design), respectively, was calculated according to the formulas given by Rufino et al. (2006), except that they were all expressed based on 1000 g N intake:

$$\text{Fecal N in uncomposted mixture (g kg}^{-1}\text{ N intake)} = 1000 \times \text{(N feces used / N intake)} \quad (~~7~~6)$$

$$\text{Urine N in uncomposted mixture (g kg}^{-1}\text{ N intake)} = 1000 \times \text{(N urine used / N intake)}$$

(~~8~~7)

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Comment [A3]: Authors: Following your suggestion in Table 5 we remove the MFE as it adds no important information to the paper. A reader interested in it can easily compute it fro… the results.

Comment [A4]: This is still very difficult to understand. I presume you mean the excreted N t… was used for making compost versus the excreted… that was discarded. Lost implies non-intentional… whereas (as I understand it and based on your response to the reviewer's comment (A56 & A57)… the excreted N that wasn't used was discarded because it couldn't be used to make compost. Doe… this relate to Figure 2 graph b or c? You refer to… retained or lost during excretion collection (Fig 2… or composting (Fig 2c).

Comment [A5]: Authors: The reviewer is rig… Using lost here was incorrect as it refers to the… unaccounted N used for composting (fig. 2 b). T… authors used the suggestion in the following reviewer comment.

Comment [A6]: Is this 44g based on the examp… you gave in your response to the reviewer's comments ie A57?

Comment [A7]: Authors: The amount of N feces used as explained in our previous answer "A57" would differ with diet. In our example, we said that 40 kg of feces was used in the mixture. This means that N feces used= N content in fec… (g/kg DM) * feces DM (g/100) * 40kg of feces. Which resulted in N feces used ranging from 1… g N for saltbush diet to 460g N for olive leaves diet. However, importantly here we calculate p…

Unaccounted N (g kg^{-1} N intake)~~, i.e., not included in our experimental design~~ = Excreted N – (Fecal N in uncomposted mixture + Urine N in uncomposted mixture)

(~~9~~8)

At the final stage of composting, N efficiency was calculated as the amount of N in compost as a proportion of N contained in the fresh feces and urine used in the mixture for composting, and in relation to N intake:

N retained during composting (g kg^{-1} N intake) = 1000 × ((N in compost / Fecal N and Urine N in uncomposted mixture)) / N intake (~~10~~9)

N lost during composting (g kg^{-1} N intake) = N in uncomposted mixture – N in compost

(~~11~~10)

For plant N uptake, N flow was calculated as the amount of N in the plant as a proportion of the amount of N applied to the soil with the amendment (feces or compost) again reported as g kg^{-1} N intake of the animal.

Plant N uptake (g kg^{-1} N intake) = NUE (%)/100 × amendment N (g kg^{-1} N intake)

(~~12~~11)

~~Where~~ where amendment N is either the fecal N (when feces are used as treatment) or N retained during composting (result from equation ~~10~~ 9 when compost is used as treatment).

The GLM procedure (SAS version 9.2, SAS Institute Inc., Cary, NC) was used for analysis of variance of the data from the incubation and the plant experiments, with amendment as a fixed factor. Means were compared with the Tukey test at $P < 0.05$. The tables give arithmetic means, standard errors of the means and *P*-values. Changes in total mineral N in the incubation experiment were analyzed by repeated measurement analysis using the GLM procedure of SAS. Treatment, time and the interaction between the two were considered as fixed factors. Figure 1 gives least square means, standard errors and *P*-values.

Results

Composition of the amendments and changes during the composting process

The mean OM content of the amendments (feces, compost and fresh olive cake) ranged from 683 to 900 g kg^{-1} DM (Table 3), and was lowest for the saltbush leaf compost, and highest for olive cake derived feces, uncomposted mixture and compost derived from this diet. The C/N ratio was on average 25 for fresh feces

Comment [A8]: Is this the 'lost' N from excretion collection, or discarded as I describe above? If so, then line 193 should read: N retained (fecal N and urine N in uncomposted mixture) from excretion collection and unaccount for in composting (ie not included in our experimental design) was calculated according to formulas given by Rufino et al. (2006), except tha they were all expressed based on 1000 g N intake

Comment [A9]: **Authors: Yes, the reviewer i right and the proposed sentence has been adop by the authors accordingly (paragraph above).**

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Comment [A10]: Presumably the N retained a N lost mentioned here refer to Fig 2c?

Comment [A11]: **Authors: Yes, that is right. The N retained and lost here refer to figure 2c.**

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Comment [A12]: How is time considered in th statistical analysis. Note that time x amendment interactions are reported suggesting that time is al a factor in the statistical analysis. Based on this it would be difficult for a reader to repeat your statistical analysis.

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Comment [A13]: **Authors: This part has escaped our attention although the analysis wa indeed done with the repeated measurement procedure. The sentence is added accordingly.**

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and 13 for compost and as high as 39 for the fresh olive cake. Accordingly, feces from the olive cake treatment also had a particularly high C/N ratio. Mineral N content (NH_4-N and NO_3-N) ~~was generally~~tended to be lower in the composts than the corresponding fresh feces, except for compost from barley straw diet. The K content was higher in the composts than in the fresh feces. Likewise P content was higher in the composts, except for saltbush leave compost.~~, whereas no consistent trend was found in P content~~. On average, total phenol content in the composts, ranging between 1 to 2 g kg^{-1} DM, was lower than in the feces and the fresh olive cake. Total phenol content in the composts represented o~~i~~n average only 33% of the content in feces from barley straw, olive leaf, saltbush leaf and olive cake diets.

The temperature in the compost bags increased from an average of 7 to 55°C during the first three days (data not shown). The differences in chemical composition between the uncomposted mixtures at the beginning of the composting procedure were largely present at the end of the composting, except for pH and EC. The initial weight of the uncomposted mixture of 52 kg (56 kg for the saltbush treatment) corresponded to 16, 20, 13 and 23 kg on DM basis for composts prepared from the excreta of the sheep fed the barley straw, olive leaf, saltbush leaf and olive cake diets, respectively. The corresponding DM losses during composting were 687, 450, 461 and 565 g kg^{-1} initial DM (data not shown), with OM losses of 729, 507, 539 and 602 g kg^{-1} initial OM, respectively. The decline in OM content was most pronounced with the saltbush leaf and the barley straw treatments (Table 3). The decline in OM was lowest with compost from the olive cake diet and intermediate with the compost from the olive leaf diet. Total N content per unit of DM increased during composting, and the C/N ratio of the four composts decreased from a range of 20 to 32 to between 12 and 17. Contents of both NH_4-N and NO_3-N decreased during the composting process except for the barley straw treatment. While the pH did not change during composting of the excreta obtained from olive leaf and olive cake fed sheep, it ~~tended to~~ decreased for the barley straw diet and ~~to~~ increased with the saltbush leaf diet. In all composts, except that produced from feeding the saltbush leaf diet, EC decreased with time.

Soil response to the different amendments

In general, all feces and compost amendments increased the OM contents of the soils, which amounted to greater than 1 g kg^{-1} soil DM on average compared to 0N soil at the end of the incubation period (Table 4).

Comment [A14]: Only saltbush leaf compost was lower than saltbush leaf feces, while barley straw compost was higher than BS feces. The oth were the same based on the SE data given.

Comment [A15]: **Authors: Except for barley straw, the sum of NO3- and NH4+ was lower in compost than fresh feces. But we agree that we have to phrase that more carefully. It is done.**

Comment [A16]: Saltbush leaf compost was lower, the others were higher than the respective feces.

Comment [A17]: **Authors: We agree, correc accordingly.**

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Comment [A18]: This appears to be true only OM, Kjeldahl N, NH4-N, NO3-N and C/N, but no for pH or EC.

Comment [A19]: **Authors: We agree, the sentence has been slightly modified.**

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Comment [A20]: Tended suggests that there w no difference, but the SE indicates that pH of the composts was different to that of the uncomposted mixtures.

Comment [A21]: **Authors: we agree, thank you.**

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This increase was about 5 times higher with the amendment with fresh olive cake when provided at the full N dose level. The quantities of OM added per kg soil with the amendments were in the range of 3 to 5 g with fresh feces, 2 to 3 g with composts and 8 or 2 g with fresh olive cake provided at the full or the 1/4 N dose.

Feces, except that from the saltbush leaf treatment, and fresh olive cake (at any dose) resulted in lower mineral N levels ~~(amendment × week, $P < 0.001$)~~ than in the 0N soil during the 12 week incubation time (amendment × week, $P < 0.001$), i.e., caused immobilization of N (Figure 1) ~~(amendment × week, $P < 0.001$)~~. In contrast, composts and feces from the saltbush ~~leave~~ leaf treatment increased mineral N content over the 0N control, although for NH_4-N ~~often~~ not significantly (Figure 1, Table 4). Mineral N (NH_4-N and NO_3-N) in the 0N soil increased from 16 to 27 mg kg^{-1} soil DM from 1 day to 12 weeks incubation (Figure 1). Compared to this change, differences in mineralization caused by the application of the organic amendments were small (Table 4). Net mineralization in compost treatments was not significantly different from the 0N treatment while fresh feces amendments caused significant immobilization, except for the saltbush leave treatment. Amendments of the saltbush leaf treatments (feces and compost) and compost of barley straw treatment resulted in significant increases in extractable K compared to the 0N soil. Finally, only amendment with saltbush leaf feces significantly increased soil P compared to the 0N soil.

The non N amended soil had a pH of 8.4 at the start of the soil incubation experiment. Only few amendments affected pH significantly, which were barley straw feces, olive cake compost and fresh olive cake at any dose (Table 4). Changes in pH during the incubation period were small with only an increase or decrease of up to 0.2 units. The EC was 0.16 ~~(~ 0.2)~~ mS cm^{-1} in the 0N soil at the beginning, but increased to 0.27 ~~(~0.3)~~ mS cm^{-1} at the end of the incubation period. Changes compared to the 0N soil ranged between 0.02 and 0.15 mS cm^{-1} directly upon amendment application, and between -0.06 and 0.~~13~~ 27 mS cm^{-1} at the end of the incubation period. However, EC was significantly higher after incubation of soil amended with mineral fertilizer N, feces and compost of the saltbush leaf and the barley straw treatments, and olive cake compost.

Plant response to the experimental amendments

Total (shoot plus root) barley DM yield was 2.5 g for plants growing on the 0N soil (Table 5). The addition of mineral N fertilizer caused a 1.7-fold increase in yield. Likewise, N yields per pot in shoot and root

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Comment [A22]: NO3-N was always significantly greater.

Comment [A23]: **Authors: yes, agreed. Sentence modified accordingly**

Comment [A24]: According to Table 4 net mineralization in soils with compost was the same the 0N soil. The feces and olive cake net mineralization were different to the 0N control.

Comment [A25]: **Authors: yes, we agree. Th additional sentence was added accordingly.**

Comment [A26]: What is the significance of rounding up 0.16 and 0.27? Use the original data

Comment [A27]: **Authors: agreed.**

biomass were increased by mineral N addition. In contrast, the addition of the fresh feces of any origin did not affect or even tended to decrease DM yield compared to 0N, except for the saltbush leaf treatment, which increased shoot and total DM yields by 1.3 and 1.2-fold levels of 0N, respectively. Composts prepared from the barley straw and saltbush leaf treatment increased total DM yields by 1.4- and 1.2-fold respectively. The low and the high dose of fresh olive cake reduced total DM yield by 0.5- and 0.3-fold compared to 0N. The N uptake of barley followed the same trend as DM yield, resulting in a negative NUE for three of the fresh feces (i.e., those derived from the barley straw, olive leaf and olive cake diets) and fresh olive cake applied at both doses. The NUE was positive with the composts but low, ranging from 2% (olive cake diet derived compost) to 16% (barley straw diet derived compost). ~~In turn, the mineral fertilizer equivalency of fresh olive cake and most fresh feces, except those derived from the saltbush treatment was negative, and positive with all composts, with a range from 2 to 21% (Table 5).~~

N flow in the feed-animal-soil-plant system

The tested feeds resulted in retention of between 158 and 239 g N kg^{-1} N intake in the growing Awassi lambs (Figure~~.~~ 2a). When compared to barley straw, fecal N excretion increased after feeding an olive leaf diet, while intake of saltbush leaf diet ~~resulted in relatively greater~~tended to increase urinary N. Due to the small amount of urine required for the compost mixtures, between 317 and 581 g urinary N kg^{-1} N intake (32 to 58% of the total excreted N) were unaccounted for in the analysis of N flows (Figure~~.~~ 2b) ~~(32 to 58% of the total excreted N)~~. ~~-~~Composting resulted in very variable N losses from less than 20% with the saltbush compost to more than half of the initial N available with the barley straw compost (Figure 2c). Overall, compost amendment resulted in a positive but small uptake of N by the barley plants (6 to 28 g kg^{-1} N intake), with the majority remaining unused in the soil (Figure 2d). In contrast~~?~~, ~~T~~the direct application of fresh feces as an amendment resulted in negative N uptake by the barley plants, leading to proportions of unused N ranging between 204 and 425 g kg^{-1} N intake (Figure 2e).

Discussion

In the farming systems typically practiced in dry areas, most of the feces and urine is excreted in the rangelands with the rest deposited in barns where animals are kept during the night and when the weather

Comment [A28]: Is this correct? Table 5 indicates 3 to 21% for total MFE, but 2 to 45% fo shoot and root MFE. By the way, how is total MI calculated. Based on the data presented it is not t sum of the shoot and root MFE.

Comment [A29]: Authors: the MFE was obtained by NUE of amendment divided by NU of mineral fertilizer within the columns for sho root and total, respectively (former equation 2) Because shoot and root do not contribute same yields and same NUE, the MFE for the total pl is not equal to the sum. However, referring to your question in Table 5, we agree to remove t MFE results as they add no important information to this paper and can easily be computed it if a reader wants this expression.

Comment [A30]: My recommendation is to indicate which graph in Figure 2 is being describe

Comment [A31]: Authors: we agree and hav adapted Fig. 2 accordingly

Comment [A32]: Is this statistically different? The error bars suggest not, ie the saltbush leaf die did not affect urinary N output.

Comment [A33]: Authors: The reviewer is right, because the difference was indeed not significant (P=0.97). The sentence was correcte accordingly.

conditions are extreme (extreme heat at noon, rain or storms). In the study region, excreta deposited in the barns are collected as a mixture of feces and straw bedding which has absorbed some of the urine. This manure is collected~~ at some intervals,~~ usually four times a year, and composted on-farm in heaps. The compost is then used in the cropping system either alone or in combination with a mineral fertilizer (urea) (personal information from the farmers).~~.~~ The traditional forage, ~~constituted~~ consisting of~~by~~ cereal straw, has been replaced occasionally by locally available, low-cost forage alternatives especially during periods of shortage (Abbeddou et al., 2011a). The different chemical composition of the alternative forages and concentrates affect the nutritive value and ~~the~~ N use~~?~~ efficiency at the animal level (Abbeddou et al. 2011a, b). No information however ~~was~~ is available about the~~ir~~ implications of their use on ~~the~~ overall N cycling in ~~the~~ a mixed crop/livestock system.

Changes ~~of~~ in amendment characteristics during composting as ~~depending~~ affected by the~~on~~ diet fed to sheep

The rate of OM degradation during composting depends on the content of readily biodegradable compounds present in the compost (Sellami et al. 2008). This suggests that the ~~less digested~~ OM in the feces of the sheep fed the saltbush leaf diet ~~was~~ should be very degradable, as had been ~~the case~~observed for most of the dietary OM in the digestive tract (Abbeddou et al. 2011a). The compost derived from the olive cake diet was higher in OM than other composts. ~~It was assumed that~~Probably in particular lignin ~~in particular~~ limited its degradation in the digestive tract as feces from olive cake diet contained 630 g neutral detergent fiber kg^{-1} DM, of which 170 g kg^{-1} DM were lignin (Abbeddou et al. 2011b). Additionally, an adverse action on digestion ~~of~~ due to the phenols present in this feed had been assumed. The low degradability of OM in the compost derived from the olive cake was consistent with the low OM digestibility of dietary olive cake in the animal. It therefore appears that the properties controlling digestibility of feeds in the animal also control degradation of feces subsequent to excretion, such as during composting (Rufino et al. 2006). The C/N ratios of most composts were in the recommended range of < 12 (Cayuela et al. 2004) for a stable OM of the compost, while it was ~~slightly~~ higher for the compost from the olive cake treatment. The NH_4^+ concentration of all composts was less than~~-~~ 0.40 g kg^{-1} as expected (Cayuela et al. 2004).

Nitrogen retention after composting was ~~higher~~ greater than the average of 54% reported by Thomsen (2000) for all composts except the barley straw diet derived compost (46% of N in the uncomposted mixture

Comment [A34]: Authors: Agreed with the modification proposed by the reviewer.

Comment [A35]: Authors: wording slightly modified

Comment [A36]: How did the phenol content the feeds affect composting?

Comment [A37]: Authors: Phenols in additi to lignin are known to reduce microbial degradation. We refer to that effect below, on lines 341-344, and lines 359-365.

Comment [A38]: Table 3 shows that olive cak compost C/N is significantly not slightly greater t that of the other composts.

Comment [A39]: Authors: Agreed

retained after composting compared to 75, 85 and 74% of N retained in olive leaf, ~~atriplex~~ saltbush leaf and olive cake derived composts; data not shown), but was in the range of 30 to 87% reported by Rufino et al. (2006).

Soil response to the different amendments

~~Compared to the 0N soil, only feces derived from the olive leaf diet caused no change in the EC of the soil. All the treatments except feces and compost from olive leaf diet and fresh olive cake caused a significant increase in EC, while feces from olive cake diet caused a significant decrease in EC after 12 weeks of incubation. Mekki et al. (2006) reported observed that an increase~~Increase in soil EC results from an increase in the electrolytes Na and K (Mekki et al. 2006), ~~with~~ and saltbush leaves ~~having~~ are reported to have high contents of these cations (Abbeddou et al. 2011a). An ingested excess of electrolytes is mostly excreted in urine, which explains the elevated EC of both the compost and the soil of the saltbush leaf treatment. ~~But also f~~Fresh feces from the saltbush leaf diet also had a high K content (Table 3).

Net N mineralization in compost amended soils was similar to net mineralization in the 0N soil. The N immobilization observed with all feces (except that from the saltbush leaves diet) agrees ~~Addition of all feces except that from the saltbush leaves diet resulted in N immobilization. These results are in accordance~~ with Wichern et al. (2004), where fresh sheep and goat feces from local farms in Oman resulted in microbial N immobilization. Net N mineralization in soil treated with feces from the saltbush diet may have resulted from relatively low C/N ratio and phenol content, and high mineral N content of this feces. Mineralization of N was related to the C/N ratio of manures as found by Powell et al. (2006). Manures with C/N ratios greater than 19 caused immobilization of soil N, as was observed here with most of the feces having C/N ratios between 22 and 32, while manures with C/N ratio less than 16 caused N mineralization. Mineralization of N might have been further reduced by the presence of polyphenols, especially after amendment by fresh olive cake and feces from the olive cake and leaf diets. Although olive cake has a moderate total phenol content, the special nature of the constitutive phenols (e.g., hydroxytyrosol, tyrosol, and their glucosides) makes it highly antimicrobial (Benitez et al. 2004; Cayuela et al. 2004; Sampedro et al. 2004). Additionally, phenols may bind proteins which are then protected from microbial degradation in the soil as for the rumen (van Bruchem et al. 1999; Tiemann et al. 2009). A regression analysis of all

Comment [A40]: Would this be saltbush, a ne name has been introduced.

Comment [A41]: **Authors: Corrected to saltbush.**

Comment [A42]: Is this data shown?

Comment [A43]: **Authors: This data can be deduced from figures 2 b and c, but directly it i not shown (we were therefore hesitant to insert "data not shown" but agree with the reviewers suggestion). It is the ratio of N retained (compost) / (N Urine + N feces in uncomposted mixture)**

Comment [A44]: These two sentences are resu and shouldn't be in this section. The paragraph needs to be reworded. It seems to me that you are trying to say that the changes in soil EC observed this experiment has been reported in the literature

Comment [A45]: **Authors: Thank you for th correction. The reviewer is right that the first paragraph was a repetition of the results. We limited the discussion to the case reported in th literature for the increase EC in soil.**

Comment [A46]: Again reword this paragraph the first two sentences describe results. Words su as:
The N immobilization observed with all feces (except that from the saltbush leaves diet) agrees with the results of Wichem….
By the way, do you have an explanation for the difference with the saltbush leaf feces? If not then say that there is no explanation. The reader will wonder if you have considered this apparent anomaly.

Comment [A47]: **Authors: Thanks for your proposition which we have included. However, think that the first sentence of this paragraph should remain as it summarizes own results to lead over to discussion. Only then the reader ca clearly differentiate results of this paper from already published results. We do not have a cle explanation for the reason why saltbush feces caused N mineralisation instead of immobilization. Probably it was a combination relatively low C/N and phenol content, and hig initial mineral N. The sentence was added now "Net N mineralization in soil treated with feces fr the saltbush diet may have resulted from relatively low C/N ratio and phenol content, and high miner N content of this feces."**

Comment [A48]: The net mineralization in compost amended soils could have been due to th C/N rations which ranged between 12 and 19. Ha you considered this?

Comment [A49]: **Authors: By manure here, considered both feces and compost. So accordi to Powell et al. (2006), manure with C/N ratios >19 would cause a net immobilization, and manure with C/N <17 would cause net mineralization. Which are in our case feces (between 22 and 32) and compost (12 to 17). T exception indeed was with saltbush feces, and w did not have an explanation (C/N =22).**

amendments in this study showed that N mineralization was negatively related to total amendment phenol input (r = – 0.75, *P* = 0.012).

Plant response to the experimental amendments

In the present study 77% of mineral fertilizer N was recovered in the barley roots and shoots, which is in the range of a pot study with ryegrass (Langmeier et al. 2002) but higher than N recovery of a barley crop growing in lysimeters (36% on coarse sand to 49% on sandy loam soil; Thomsen et al. 1997) and recoveries in crops reported from field studies which usually are in the range of 20 to 50% (Crews and Peoples 2005). The NUE for animal manure is usually lower than that of mineral N, because it is mainly composed by organic N forms which are not readily available to plants. In a pot study six harvests of ryegrass recovered 25 to 30% of N applied with fresh cow feces (Langmeier et al. 2002), while wheat growing in microplots in the field recovered 10% of N applied with sheep feces (Bosshard et al. 2009). Also in a microplot field study, barley at maturity (grain and straw) recovered 6% of N-15 added with sheep feces (Jensen et al. 1999). ~~In contrast, in~~ In this experiment, the application of fresh feces reduced plant productivity compared to the 0N soil, resulting in negative NUE except for the amendments from the saltbush leaf treatments. This could be explained by the higher amounts of mineral N (NH_4-N, NO_3-N) found in the feces from the saltbush leaf treatment than the other fecal amendments, and organic fecal N compounds that can be easily mineralized (Bosshard et al. 2011)

In contrast to the feces, a positive NUE was observed for all composts resulting in N recoveries in barley of 2 to 16% of the N applied. This is in the range found for fresh feces in other studies but much less than that reported for- slurry composed of all of the feces and urine excreted (Langmeier et al. 2002; Bosshard et al. 2009). The NUE of both olive feed containing diets was lower than those from the barley straw and the saltbush based diet. In their review, Rufino et al. (2006) reported that NUE from composted manure for maize crops in Africa ranged between 3 and 49%, with NUE being higher when urine was included in the composted manure. Nitrogen recovery from compost was higher than from fresh feces probably because soil amended with compost had higher total mineral N than the soil amended with feces. Also, compost had lower phenol contents than feces; the latter also because of dilution by urine and straw. Urine N mostly consists of urea which is a readily available N source for plants (Bosshard et al. 2009). However, part of the

Comment [A50]: In this and the subsequent sentence you are discussing plant production relat to 0N soil and state that this is in contrast [to the literature], although in the literature cited previous you describe production from feces not relative to 0N soils. In other words, if these sentences are no 'in contrast' to the literature presented previously, don't start the sentence with 'In contrast'. It is confusing for the reader who is trying to relate thi information with what was presented previously.

Comment [A51]: Authors: The "in contrast" may be somewhat confusing, and we have removed it. Still, to improve clarity, we inserte the "resulting in negative NUE. This also bette links with the next paragraph.

urea N gets hydrolyzed during composting (Thomsen 2000; Wichern et al. 2004) and lost in the form of ammonia (Rufino et al. 2006). The high pH in composts and soils may further have intensified ammonia volatilization. As no ^{15}N-labeling was done in the present study, N derived from feces and urine, and their respective contribution to barley N uptake (Bosshard et al. 2009), cannot be separated.

The NUE of fresh olive cake was negative, showing indicating that the N in olive cake was not directly available for the plants and that olive cake amendment reduced the availability of soil N. The inhibitory effect on plant productivity was less pronounced when less olive cake was added (1/4 of the N dose used in the other treatments). This suggests that feeding olive cake may be relatively more advantageous in terms of laterthan use of fresh olive cake as an amendment. When olive cake passes through the rumen and the digestive track, phenols are either metabolized or diluted when mixed to with other feed ingredients resulting in excreta that might be less toxicinhibitory to plant growth.

Amendment N not taken up by the crops can remain in the soil (Powell et al. 2006) or be lost from the soil-plant system. Studies using ^{15}N labeling show that with organic amendments often more N is recovered in the soil than with mineral fertilizer N (Sørensen and Thomsen 2005; Gardner and Drinkwater 2009). This is also supported by the studies by Langmeier et al. (2002) and Bosshard et al. (2009), where about 60% of N applied with fresh cattle or sheep feces remained in the soil. This N can become available to subsequent crops (Schröder et al. 2005) although at low release rates. Accordingly, only around 3.3 and 1.5% of feces N were recovered during the two following years in the study by Bosshard et al. (2009). The difference method used in this study does not allow measurement of amendment N recovery in the soil compared to the background soil N.

Nitrogen flow in the feed-animal-soil-plant system as affected by unconventional feeds

The NUE of a mixed crop/livestock system is the resultant of N conversion at the animal level and the apparent N recovery from the soil amended by animal excreta (van Bruchem et al. 1999). The calculated N conversion efficiency at the animal level for growth (body N retention, Figure 2a) was in the range of 150 to 250 g N kg^{-1} N intake reported by van Bruchem et al. (1999). All diets had similar N conversion efficiencies despite the lowest N intake associated with the saltbush leaves and olive cake diets The olive cake based diet gave the highest N conversion efficiency of all the tested diets, but this was mainly due to the lowest N

Comment [A52]: Fig 2a shows that there is no difference between the diets based on the error ba ie the N conversion efficiencies is the same for all diets. How about: "All diets had similar N conversion efficiencies despite the lowest N intake associated with the oli cake diet" Also is the N intake info from Abbedd et al. 2011a, b truly different?

~~intake on this diet~~ (data taken from Abbeddou et al. 2011a, b). Nitrogen not converted into animal products (milk or body weight gain) is excreted with urine and feces, but also the allocation of excretory N to feces and urine clearly varied ~~among~~ between treatments due to phenol content in the diets. As~~….. asfor the reasons~~ stated above, proteins bound to phenols are indigestible complexes excreted in feces where they constitute the insoluble N fraction (Powell et al. 1994). ~~The different other diets (barley straw, olive leaves, and saltbush leaves and olive cake) gave similar N conversion efficiency. However, b~~Based on the allocation of the excretory N, the lowest feces N as a proportion of N intake was found with the saltbush diet (*P* < 0.001) and the highest with diets containing olive-derived feeds. ~~In this study, the NUE of composts and of feces was tested when used as amendments for crop production. As discussed above, NUE by barley of both forms was low or even negative and a large fraction of applied N remained unused (Figure 2).~~ Not all N excreted by the animals was included in manure N flow. Thus a large proportion of N excreted, i.e., urinary N with high plant availability, remained unaccounted for (32 to 58% of the total excreted N; Figure 2b). Our experiment mimicked a situation where all feces would be collected, but where the urine not absorbed by the straw would have been lost. Farming systems with sheep in Syria are quite variable (Rischkwosky et al. 2004), and in turn the collection and use of animals excreta differs between farms and production areas. Likewise, straw is a limited valuable resource, and compost preparation and application may economically not always be viable as straw is in demand as livestock fodder (Sommer et al. 2011). Thus its use as animal bedding in barn is not always applied, especially in seasons with low barley straw yields.

In this study, the NUE of composts and of feces was tested when used as amendments for crop production. As discussed above, NUE by barley of both forms was low or even negative and a large fraction of applied N remained unused (Figures 2d and 2e). The question arises whether the feces and urine would be better used on the rangelands than for crop production. We have no exact data on the proportion of N deposited in the rangelands ~~and~~ compared with in the barns. Rufino et al. (2006) estimated that less than 50% of total N excreted can be collected in typical mixed grazing-barn system by smallholders in the sub-Saharan Africa. Based on experimental data of excreta collection and N loss during composting in central Kenya (Lekasi et al. 2001), Rufino et al. (2006) assumed that less than 10% of N in the excreta would be efficiently used in the mixed crop-livestock system in Kenya. Nitrogen deposited in the rangeland can be taken up by plants, be retained in the soil or be lost. Plant N uptake is limited to the short (up to three months in the dry

Comment [A53]: Authors: we apologize for this mistake and thank the reviewer for catchin it. Yes indeed, the N conversion was not significantly different among all the diets. However, the N intake was significantly lower with the olive cake and saltbush diets.

Comment [A54]: Please briefly summarise th reasons so the reader doesn't have to go and find them in the text.

Comment [A55]: Authors, this is done as suggested by the reviewer.

Comment [A56]: Authors: the sentence is a repetition of the modified statement. Deleted.

areas of Syria) growing season when water availability sustains plant growth and nutrient uptake. During the six month-long dry period, particularly urine N deposited on the alkaline soils of the study area may largely be lost by ammonia volatilization. For instance, Vallis et al. (1985) reported a loss of 46% of urine N when cattle urine was applied on pasture during the dry season in Australia. Under these conditions, N cycling could be increased when a higher proportion of N is excreted with feces because feces N takes longer time to be mineralized and therefore the risk of ammonia volatilization is lower (Powell et al. 1994; Rufino et al. 2006). Based on this, diets containing olive-derived feeds seemed more efficient than the other diets, especially the saltbush leaf based diet which resulted in the highest proportion of N excreted in urine. Still it has to be shown whether N from olive cake and olive cake derived feces will be available in subsequent years or whether the compositional limitations prevent its release even longer.

Conclusions

Providing unconventional feeds to sheep only slightly affected the NUE by barley of N applied with feces or composts produced from feces, urine and straw, with NUE being~~-~~ higher after treatments with composts than with feces. ~~Reflecting the limited collection practiced on farm, an important proportion of urine N was not considered in the present study.~~ The NUE obtained in the pot experiment was low or even negative, leaving most of the N applied unused. Feeding olive cake was more efficient as some nutrients were supplied when fed to the sheep increasing overall N cycling efficiency compared with its direct application as soil amendment. Labeling studies with ^{15}N ~~labeling studies are needed~~ to separate unused N into N retained in soil and N losses are needed, ~~and~~ as well as long-term plant experiments under field conditions to test residual amendment N effects. ~~Feeding olive cake turned out to be more efficient by supplying some nutrients when fed to the sheep and increasing overall N cycling efficiency than its direct application as soil amendment.~~ Although reflecting the limited collection practiced on farm, an important proportion of urine N was not considered in the present study. When the intention is to use amendments consisting of animal excreta strategically for crop production and in the growing season, it seems worthwhile to capture as much urine as possible and, therefore, to change excreta storage and application practices.

Comment [A57]: For flow I would rearrange t sentences as follows.

Comment [A58]: **Authors: Thanks, this is fi**

Comment [A59]: **Authors: We here mean th NUE by barley, as shown in Table 5 and Figure d and e. There the main difference clearly is fe against compost , as stated in the second part o the sentence. The sentence has been modified f clarification.**

Acknowledgments This work was funded by the Swiss Development Cooperation, Berne, through the North-South Center, ETH Zurich, Switzerland. We are grateful to Thomas Flura from the plant nutrition group (ETH Zurich) for his help to set up the experiment and his technical advice~~s~~ during soil and plant analysis. Our thanks go to George Estefan, Ahmed Sawass and their teams from ICARDA for their technical help during the whole experiment and to Rolf Sommer (ICARDA) and the anonymous reviewers for ~~his~~ the helpful comments on our manuscript.

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Comment [A60]: Abbreviate

Comment [A61]: Authors: the title does not have an abbreviation as checked by the author

Comment [A62]: Abbreviate

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Comment [A65]: Authors: abbreviated.

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Comment [A67]: **Authors: The reference wa used for the classification of the soil used in thi study. Materials and Methods, Line 123.**

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Comment [A68]: Authors: without t in the official abbreviation.

Comment [A69]: Abbreviate

Comment [A70]: Authors: this is the abbreviation.

Comment [A71]: Abbreviate

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Legend of the figures

Figure 1 Changes in total mineral N (mg kg^{-1} DM) of unamended soil (0N), soil amended with mineral N (ammonium sulfate), soil amended with feces from sheep fed barley straw, olive leaves, saltbush leaves or fresh olive cake~~;~~ (A);~~,~~ soil amended with compost made from the feces and urine of sheep fed ~~by~~ these four diets~~;~~ (B~~),~~); and soil amended with fresh olive cake at 90 mg N kg^{-1} and 22.5 mg N kg^{-1}~~;~~ (C). Amendment, $P < 0.001$; week, $P < 0.001$; amendment × week, $P < 0.001$, SEM at treatment level = 1.9

Figure 2 Nitrogen flows in a feed-animal-soil-plant system, where barley straw, olive leaf, saltbush leaf and fresh olive cake diets are fed to Awassi sheep, and the feces and some urine are used to make compost to grow barley plants. Nitrogen flows from the feces to soil is also compared, but most of the urine N is unaccounted for in this experimental system.

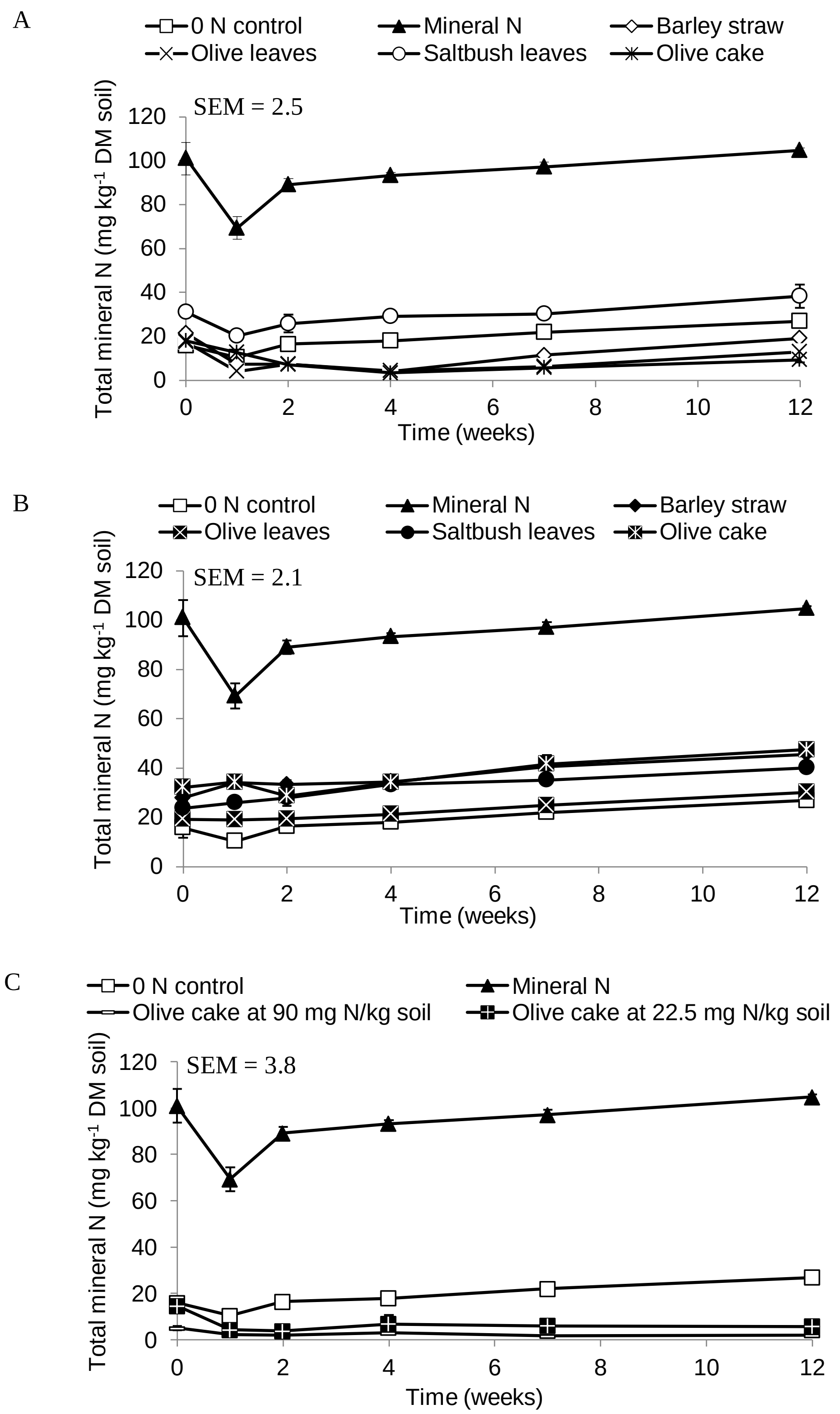
A
0 N control
Mineral N
Barley straw
Olive leaves
Saltbush leaves
Olive cake
SEM = 2.5
Total mineral N (mg kg-1 DM soil)
120
100
80
60
40
20
0
0
2
4
6
8
10
12
Time (weeks)
B
0 N control
Mineral N
Barley straw
Olive leaves
Saltbush leaves
Olive cake
SEM = 2.1
Total mineral N (mg kg-1 DM soil)
Time (weeks)
C
0 N control
Mineral N
Olive cake at 90 mg N/kg soil
Olive cake at 22.5 mg N/kg soil
SEM = 3.8
Total mineral N (mg kg-1 DM soil)
Time (weeks)

Figure 1

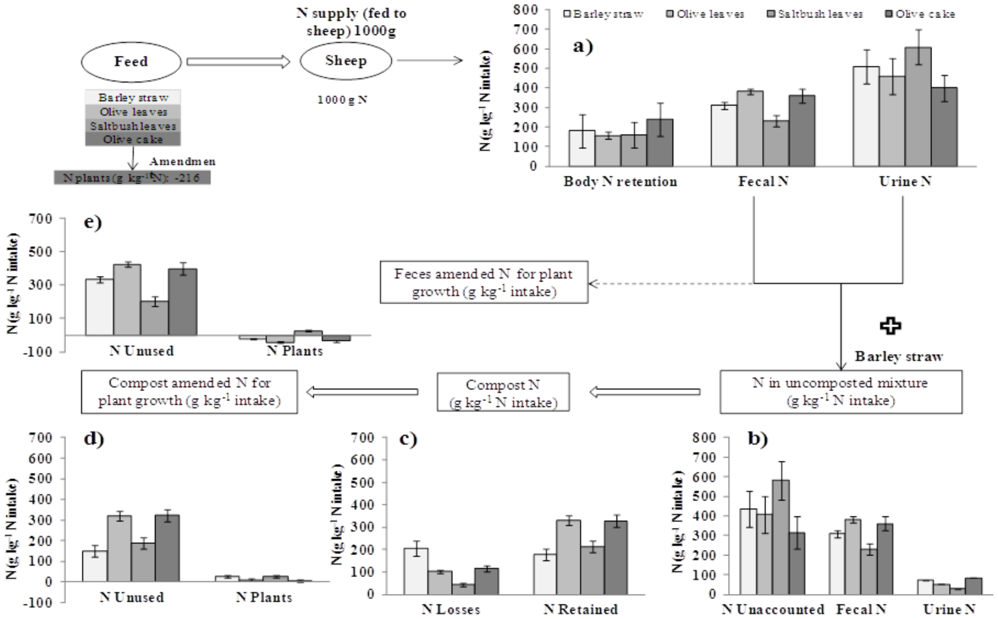
N supply (fed to sheep) 1000g
Feed
Sheep
1000 g N
Barley straw
Olive leaves
Saltbush leaves
Olive cake
Amendmen
a)
Body N retention
Fecal N
Urine N
Feces amended N for plant growth (g kg-1 intake)
Barley straw
N in uncomposted mixture (g kg-1 N intake)
Compost N (g kg-1 N intake)
Compost amended N for plant growth (g kg-1 intake)
e)
N Unused
N Plants
d)
c)
N Losses
N Retained
b)
N Unaccounted
N(g kg-1 N intake)

Figure 2

Table 1 Feeding trials description Description of feeding trials and composition of the diets fed to fat-tailed Awassi sheep

Feeding trial	Diet	Feed component description (g kg^{-1} DM)	Nutrients content in feed
Replacing bBarley straw replaced withby olive leaves and or saltbush leaves in fat-tailed Awassi sheep (. [Φ]Abbeddou et al. 2011a)	Barley straw	716 g barley straw, 176 g cotton seed meal, 88 g molasses, and 20 g mineral-vitamin mix. 16 g urea kg^{-1} DM.	DM: 1100 g CP: 159 g kg^{-1} DM Metabolisable energy: 7.59 MJ kg^{-1} DM
	Olive leaf	716 g olive leaves, 44 g barley grain, 44 g wheat bran, 88 g cotton seed meal, 88 g molasses, and 20 g mineral-vitamin mix. 11 g urea kg-1 DM.	DM: 1100 g CP: 157 g kg^{-1} DM Metabolisable energy: 7.85MJ kg^{-1} DM
	Saltbush leaf	716 g saltbush leaves, 44 g barley grain, 132 g wheat bran, 88g molasses, and 20 g mineral-vitamin mix. 2 g urea kg^{-1} DM.	DM: 1100 g CP: 159 g kg^{-1} DM Metabolisable energy: 7.59 MJ kg^{-1} DM
Concentrate replaced withReplacing the concentrate by Mediterranean by-products, ie olive cake. Ψ in fat-tailed Awassi sheep (Abbeddou et al. 2011b)	Olive cake	490 g barley straw, 340 g olive cake, 100 g cotton seed meal, 50 g molasses, 20 g mineral-vitamin mix.19g urea kg^{-1} DM.	DM: 1100 g CP: 147 g kg^{-1} DM Metabolisable energy: 5.50 MJ kg^{-1} DM

[Φ]For details see Abbeddou et al. (2011a)
Ψ For details see Abbeddou et al. (2011b)

Comment [A75]: Authors agree with review changes; some further minor modifications we added .

Table 2 Properties of the soil used in soil and plant growth experiments.

Soil classification	Texture (g kg^{-1})			Composition (g kg^{-1} DM)					pH	Electrical conductivity (mS cm^{-1})
	Clay	Silt	Sand	Organic matter	Total N	Mineral N	Olsen-P$^{\Psi}$	Extractable K$^{\omega}$		
Montmorillonitic, thermic, Chromic Calcixerert	539	283	178	10.4	0.78	0.01	0.012	0.27	8.2	0.271

$^{\Psi}$Extraction with sodium bicarbonate (Olsen et al., 1954)
$^{\omega}$Extraction with ammonium acetate (Richards 1954)

Comment [A76]: Authors: Additional reference for completeness (see material and method section)

Table 3 Physico-chemical characteristics of the amendments used for soil and pot experiments. The amendments consist of fresh feces excreted by sheep fed barley straw, olive leaf, saltbush leaf or olive cake diets;, uncomposted mixtures of feces (excreted by sheep fed the four diets), urine and barley straw;, the compost made from the mixtures; and fresh olive cake. Mean The mean and standard error of 3 three analytical replicates are given.

	Organic matter	Kjeldahl N	NH_4-N	NO_3-N	Total K	Total P	Total phenols	C/N	pH	Electrical conductivity
	(g kg^{-1} dry matter)									(mS cm^{-1})
Fresh feces										
Barley straw	870±0.0	18±5.3	1.0±0.10	0.9±0.03	2.4±0.7	7.2±1.2	5.0±0.7	24±7.4	nd	nd
Olive leaves	841±0.5	24±0.3	0.7±0.03	0.6±0.03	1.8±0.6	8.7±1.5	6.1±0.3	21±0.2	nd	nd
Saltbush leaves	762±6.6	21±0.2	1.7±0.09	2.0±0.05	10.1±3.8	12.2±1.7	3.2±0.2	22±0.2	nd	nd
Olive cake	897±0.6	15±0.2	0.4±0.03	0.3±0.02	2.6±0.6	6.0±0.8	3.1±0.9	32±0.4	nd	nd
Uncomposted mixture before composting										
Barley straw	843±0.8	23±3.1	5.8±0.40	5.1±0.40	nd	nd	nd	20±2.2	8.7±0.10	4.6±0.07
Olive leaves	845±0.8	25±1.3	3.8±0.38	3.5±0.27	nd	nd	nd	20±5.9	8.4±0.00	3.7±0.00
Saltbush leaves	798±1.4	19±0.8	0.7±0.20	1.0±0.20	nd	nd	nd	25±6.5	8.6±0.10	4.1±0.00
Olive cake	900±1.4	15±0.2	2.5±0.31	2.3±0.45	nd	nd	nd	32±0.4	8.5±0.07	3.3±0.07
Compost after 18 weeks of composting										
Barley straw	731±0.9	34±0.2	0.03±0.001	4.3±0.40	23.7±2.1	11.4±0.5	1.9±0.2	12±0.1	8.1±0.07	4.3±0.10
Olive leaves	757±2.5	35±0.4	0.00±0.001	0.4±0.02	15.3±1.1	13.0±0.8	1.8±0.2	12±0.1	8.4±0.10	3.1±0.10
Saltbush leaves	683±1.1	30±0.8	0.06±0.002	0.7±0.01	20.2±2.6	9.5±1.0	0.9±0.4	12±0.3	9.0±0.07	5.4±0.07
Olive cake	824±3.4	25±0.3	0.00±0.002	0.4±0.03	12.5±0.5	7.8±0.2	1.2±0.2	17±0.2	8.5±0.00	1.5±0.00
Fresh olive cake	864±0.2	13±1.8	nd	Nd	8.3±0.7	0.8±0.2	4.1±0.5	39±0.6	nd	nd

nd = not determined

Table 4 Mean organic matter, mineral N (NH_4-N, NO_3-N), calculated net N mineralization and net N mineralization over 0N control, nutrient content (ammonium acetate extractable K, sodium bicarbonate extractable "Olsen"-P), pH, and electrical conductivity ~~and calculated net N mineralization and net N mineralization over 0N control~~ of unamended soil (0N), soil amended with mineral N (ammonium sulfate), soil amended with fresh feces excreted by sheep fed barley straw, olive leaf, saltbush leaf or fresh olive cake diets, soil amended with compost (90 mg N kg-1) made with feces from sheep fed these diets or soil amended with fresh olive cake (90 or 22.5 mg N kg-1) at the beginning (Initial) and after 12 weeks (Final) of incubation ~~(Final)~~ (n=4).

Treatment	Organic matter (g kg^{-1} soil DM)	NH_4-N (mg kg^{-1} soil dry matter (DM))		NO_3-N (mg kg^{-1} soil DM)		Net N mineral-ization	Net N mineral-ization over control	Extractable-K (g kg^{-1} soil DM)	Olsen-P (g kg^{-1} soil DM)	pH		Electrical conductivity (mS cm^{-1})	
	Final	Initial	Final	Initial	Final			Final	Final	Initial	Final	Initial	Final
0N control	11^{cd}	1.5^{c}	0.1^{b}	14^{e}	27^{e}	11.0^{ab}		0.48^{b}	0.04^{b}	8.4^{ab}	8.2^{c}	0.16^{d}	0.27^{fg}
Mineral N	11^{d}	78.9^{a}	0.4^{ab}	22^{bcd}	104^{a}	3.9^{bcd}	– 7.2^{bcd}	0.48^{b}	0.04^{b}	8.3^{abc}	8.2^{c}	0.25^{ab}	0.54^{a}
Fresh feces - 90 mg N kg^{-1}													
Barley straw	12^{bcd}	2.0^{c}	0.7^{ab}	19^{cde}	18^{f}	– 2.4^{cde}	– 13.4^{cde}	0.48^{b}	0.04^{ab}	8.2^{cd}	8.3^{ab}	0.26^{ab}	0.33^{de}
Olive leaves	12^{bcd}	2.1^{c}	0.6^{ab}	15de	12^{g}	– 4.4^{de}	– 15.4^{de}	0.50^{ab}	0.05^{ab}	8.4^{a}	8.3^{ab}	0.18^{cd}	0.24^{fgh}
Saltbush leaves	12^{bcd}	13.5^{b}	0.3^{ab}	17^{cde}	38^{d}	7.4^{abc}	– 3.6^{abc}	0.54^{a}	0.06^{a}	8.4^{ab}	8.3^{bc}	0.25^{ab}	0[illegible]
Olive cake	12^{b}	0.9^{c}	0.8^{a}	17^{cde}	8^{gh}	– 8.8^{e}	– 19.8^{e}	0.49^{ab}	0.05^{ab}	8.3^{ab}	8.3^{ab}	0.24^{bc}	0[illegible]
Composts - 90 mg N kg^{-1}													
Barley straw	11^{bcd}	0.7^{c}	0.4^{ab}	27^{ab}	45^{bc}	17.8^{a}	6.8^{a}	0.54^{a}	0.05^{ab}	8.3^{abc}	8.2^{c}	0.27^{ab}	0.34[illegible]
Olive leaves	12^{bcd}	0.7^{c}	0.5^{ab}	19^{cde}	30^{e}	11.1^{ab}	0.13^{ab}	0.50^{ab}	0.05^{ab}	8.3^{ab}	8.3^{ab}	0.24^{bc}	0.29^{ef}
Saltbush leaves	12^{bc}	0.8^{c}	0.5^{ab}	23^{bc}	40^{cd}	16.4^{a}	5.4^{a}	0.54^{a}	0.05^{ab}	8.3^{bcd}	8.3^{ab}	0.31^{a}	0.39^{bc}
Olive cake	12^{b}	0.8^{c}	0.4^{ab}	31^{a}	47^{b}	15.4^{a}	4.4^{a}	0.50^{ab}	0.04^{b}	8.1^{d}	8.2^{c}	0.29^{ab}	0.36^{bcd}
Fresh olive cake													
90 mg N kg^{-1}	15^{a}	1.0^{c}	0.4^{ab}	4^{f}	1^{i}	– 3.1^{cde}	– 14.1^{cde}	0.52^{ab}	0.04^{b}	8.1^{d}	8.3^{bc}	0.24^{bc}	0.23^{gh}
22.5 mg N kg^{-1}	12^{bc}	1.5^{c}	0.4^{ab}	13^{e}	5^{hi}	– 8.9^{e}	– 11.6^{cde}	0.51^{ab}	0.04^{b}	8.2^{cd}	8.4^{a}	0.25^{ab}	0.23^{gh}
SEM	0.2	3.16	0.04	1.1	3.9	1.5	1.5	0.004	0.001	0.11	0.01	0.01	0.01
P-value treatment	<0.001	<0.001	0.100	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Means carrying no common superscript within each column are different at $P < 0.05$ (Tukey test).

Comment [A77]: Authors: corrected

Comment [A78]: Why is this negative sign different?

Table 5 Mean dry matter (DM), N yield and N use efficiency by barley grown on unamended soils (0N), soil amended with mineral N (ammonium N), soils amended with fresh feces excreted by sheep consuming barley leaves, olive leaves, saltbush leaves and fresh olive cake, soils amended with compost made using feces from sheep fed these diets, or soils amended with fresh olive cake (n=4)

Treatment	DM (g)			N yield (mg pot^{-1})			N use efficiency (%)$^{\Psi}$			~~Mineral fertilizer equivalent$^{\omega}$ (%)~~		
	Shoot	Root	Total	Shoot	Root	Total	Shoot	Root	Total	~~Shoot~~	~~Root~~	~~Total~~
0N control	1.2^{d}	1.3^{cd}	2.5^{d}	14^{c}	9^{ef}	23^{c}						
Mineral N	2.5^{a}	1.6^{ab}	4.1^{a}	62^{a}	23^{a}	85^{a}	60^{a}	17^{a}	77^{a}			
Fresh feces												
Barley straw	0.7^{e}	1.0^{ef}	1.6^{e}	8^{d}	9^{efg}	17^{d}	– 7^{d}	– 1^{ef}	– 8^{d}	~~– 12^{c}~~	~~– 4^{cd}~~	~~– 10^{d}~~
Olive leaves	0.5^{f}	0.8^{f}	1.4^{ef}	7de	7^{fg}	14^{d}	– 9de	– 3^{f}	– 11^{d}	~~– 14^{c}~~	~~– 16^{d}~~	~~– 15^{d}~~
Saltbush leaves	1.6^{bc}	1.4^{bc}	3.0^{c}	20^{b}	13^{bcd}	32^{b}	7^{b}	4^{bcd}	11^{b}	~~12^{a}~~	~~24^{b}~~	~~14^{b}~~
Olive cake	0.7^{e}	0.8^{f}	1.5^{ef}	8^{d}	7^{fg}	15^{d}	– 7^{d}	– 3^{f}	– 10^{d}	~~– 12^{c}~~	~~– 17^{d}~~	~~– 13^{d}~~
Compost												
Barley straw	1.7^{b}	1.8^{a}	3.4^{b}	21^{b}	16^{b}	36^{b}	8^{b}	8^{b}	16^{b}	~~14^{a}~~	~~45^{a}~~	~~21^{a}~~
Olive leaves	1.3^{d}	1.3^{cd}	2.6^{d}	15^{c}	11^{cde}	26^{c}	1^{c}	2^{cde}	3^{c}	~~2^{b}~~	~~11^{bc}~~	~~4^{c}~~
Saltbush leaves	1.5^{c}	1.4^{bc}	3.0^{c}	19^{b}	13^{bc}	33^{b}	7^{b}	5^{bc}	12^{b}	~~11^{a}~~	~~29^{ab}~~	~~15^{b}~~
Olive cake	1.2^{d}	1.1^{de}	2.3^{d}	15^{c}	10^{def}	25^{c}	1^{c}	1^{def}	2^{c}	~~2^{b}~~	~~4^{c}~~	~~3^{c}~~
Fresh olive cake												
90 mg N kg^{-1}	0.3^{g}	0.4^{g}	0.7^{g}	3^{e}	3^{h}	6^{e}	– 13^{e}	– 9^{g}	– 22^{e}	~~– 22^{d}~~	~~– 50^{e}~~	~~– 28^{e}~~
22.5 mg N kg^{-1}	0.5^{f}	0.7^{f}	1.2^{f}	6de	6^{g}	11^{d}	– 40^{f}	– 18^{h}	– 58^{f}	~~– 67^{e}~~	~~– 106^{f}~~	~~– 76^{f}~~
SEM	0.09	0.06	0.14	2.2	0.8	2.9	3.5	1.3	4.7	~~3.5~~	~~6.4~~	~~4.1~~
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	~~<0.001~~	~~<0.001~~	~~<0.001~~

Means carrying no common superscript are different at $P < 0.05$ (Tukey test).

$^{\Psi}$N use efficiency relative to barley grown in the 0N control soils.

~~$^{\omega}$N use efficiency relative to N use efficiency of the mineral fertilizer.~~

Comment [A79]: The MFE is not discussed. It is also not mentioned in the caption. Should it be included?

Comment [A80]: Authors: we agree with your comment and have removed it as all data to compute it is shown.

Unconventional feeds for small ruminants in the dry areas have a minor effect on manure nitrogen flow in the soil-plant system

General reply:

The authors thank the reviewers for their very pertinent comments and corrections.

Reviewer 1: The authors have responded to previous reviewer comments and suggestions and made very commendable improvements to their manuscript. There are some minor typographical and editorial changes that need to be made in the Abstract, Introduction Materials and methods sections. In the latter section, the calculations are a little confusing to follow and I have taken the liberty of inserting paragraph breaks to clarify the sections to which the equations belong. If these paragraph breaks are not in the correct places them rearrange as required.

Authors: all the comments and suggestions given were considered and included in the text.

Reviewer 1: The reporting of the results has improved. However there are still a number of instances when the authors refer to differences (eg lines 225, 255, 287, 319) or similarities (line 258) that do not exist based on the statistical results shown. Please fix these errors (noted in track changes in the text) as the interpretation of the data is in question. Also inconsistencies occur between the data reported in the text and that in the tables and figures on a number of occasions.

Authors: all the inconsistencies in the results were corrected and checked thoroughly with the data reported in the table. Please find the answers to the different raised questions in the comments (track changes).

Reviewer 1: In the Discussion section, the authors often repeat results, usually as an introductory sentence for a paragraph. The text needs to be re-written so that it does not read as the Results section. Again the authors report results as different or similar that aren't based on the statistical analyses.

Authors: All the small introductions in the discussion part that they are results have been deleted. The different paragraphs in the discussion part have been reformulated accordingly.

Reviewer 1: This is particularly so with the first sentence of the Conclusions, which calls into question the conclusions that are being drawn. I have also rearranged the Conclusions to improve the flow and connectedness of the sentences.

Authors: thank you for the correcting the conclusion. The authors explained in the comment the first sentence of the conclusion. The NUE in there was related to the plant experiment (NUE of barley plant).

Reviewer 1: A number of references in the bibliography haven't been abbreviated. Please check that these meet the Journal's guidelines. Also I couldn't find one reference in the text. However I may have missed it, so please confirm.

Authors: Some of the references were indeed not abbreviated but some others did not have an abbreviation. E.g. Agronomie, Rangelands, they do not have an official abbreviation.

Reviewer 1: Figures and Tables:

Figure 1 looks good. The legends are clear and the graphs are easy to read.

Authors: Thank you.

Reviewer 1: Figure 2 is much improved and easier to read and understand. I would suggest numbering each graph so that they can be referred to in the text individually rather than the reader having to figure out which graph is being referred to. The Legend is only required once, particularly as it did not print fully above all graphs. Above Figure 2 a is fine. Delete the legend above the other graphs. Rearrange the columns in graphs 2a and 2b so that fecal N is between N retention and Urine N and between Unaccounted N and Urine N respectively. This is to make the arrows to the feces amended N box easier to read. I have made some additional changes to the figure. Note that the x-axis labels for Figures 2b and 2c did not print out on my copy, which made understanding the 'Unaccounted' and the 'Lost' and 'Retained' data difficult. Make sure that the whole figure fits on a page.

Authors: All the suggested modifications were taken into consideration in order to improve the figure.

Reviewer 1: The captions to the tables are much clearer now. Some typographical changes have been made to the tables to improve readability.

Authors: Thank you.

Reviewer 1: The manuscript has improved vastly. However the authors have to ensure that their reporting of results takes into consideration the statistical output rather than whether or not the data looks different. The research and results are worthwhile, the interpretation needs to be improved.

My comments and suggestions are included in the Word document, using Track Changes or by inserting comments throughout the text.

Authors: Thank you for all your efforts to help us improving our manuscript. All your suggestions were considered and most of them included in the text, tables and figures. Please find them in the attached file, the manuscript with track changes.