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Effects of replacing corn and soya beans with white sorghum and faba beans on milk quality of Sicilo Sarde dairy ewes in Tunisia

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Abstract. The effects of using white sorghum seeds and faba beans as feed resources were studied on milk performances in dairy Sicilo-Sarde ewes during the milking phase. Twenty ewes were divided into two homogeneous batches according to age, litter size, rank of lactation and live body weight. Ewes received 1.5 kg DM of oat hay /ewe/day supplemented with 500 g/ewe/day of a standard concentrate diet (group C) or a concentrate composed of local white sorghum, faba beans and a sheep vitamin and mineral supplement (group S). Milk quality was determined once a week on samples of bulk milk batch to measure the fat content by Lactoscan. The concentration of urea in milk was determined by the DMAB method. The extraction of lipids was determined following Folch *et al.* (1957) method; the fatty acid composition was determined by gas chromatography. The use of the experimental concentrate (white sorghum and faba beans) affected the protein, non fat solids and lactose contents ($p < 0.05$). The average level of urea was 53.5 ± 8.76 and 35.5 ± 3.4 mg/dl for the C and S ($p < 0.05$), respectively. The main fatty acids were palmitic acid ($27.75 \pm 1.29\%$ vs. $27.77 \pm 1.21\%$ of total fatty acids for the S and C diets, respectively; $p > 0.05$) and miristic acid ($12.06 \pm 0.82\%$ and $12.47 \pm 1.21\%$ for CC and CS, respectively; $p > 0.05$). The average of capric acid was comparable in both concentrates ($7.17 \pm 1.17\%$) in the control group (C) and $7.38 \pm 1.39\%$ in the experimental group (S). The concentration of conjugated linoleic acid (CLA) was 0.48 ± 0.03 and $0.36 \pm 0.09\%$ for the for the C and S ewe groups, respectively ($p > 0.05$).

Keywords. Dairy sheep – Conjugated linoleic acid – Fatty acids – Milk quality – Urea.

Effet du remplacement du maïs et du soja par le sorgho blanc et la féverole sur la qualité du lait, la concentration d'urée, la composition des acides gras et l'acide linoléique conjugué (CLA) dans le lait des brebis Sicilo-Sardes en Tunisie

Résumé. L'effet de l'utilisation du sorgho blanc et de la féverole comme ressources alimentaires locales a été étudié sur les performances laitières de la brebis Sicilo-Sarde durant la phase de traite. Vingt brebis ont été divisées en deux groupes homogènes selon l'âge, la taille de la portée, le numéro de lactation et le poids vif. Les brebis ont reçu 1,5 kg MS/brebis/jour de foin d'avoine complémenté par 500 g/brebis/jour d'aliment concentré commercial (lot C), et 500 g/brebis/jour d'aliment concentré local à base de sorgho blanc, féverole et CMV ovins (lot S). La qualité du lait durant les semaines de contrôle a été déterminée par un Lactoscan Milk analysis. La concentration de l'urée du lait a été déterminée par la méthode DMAB. L'extraction des lipides du lait a été déterminée selon la méthode de Folch *et al.* (1957). Le remplacement du maïs et tourteau de soja par le sorgho blanc et la féverole a affecté l'extrait sec dégraissé et le lactose du lait ($p < 0,05$). La moyenne de l'urée du lait était de $53,5 \pm 8,76$ pour le lot C et $35,5 \pm 3,4$ mg/dl pour le lot S ($p < 0,05$). La moyenne des acides gras était pour l'acide palmitique C16 ($27,75 \pm 1,29\%$ vs. $27,77 \pm 1,21\%$ des acides gras totaux respectivement pour les lots S et C) et pour l'acide myristique C14 ($12,06 \pm 0,82\%$ et $12,47 \pm 1,21\%$ respectivement pour C et S). La moyenne de l'acide caprique C10 était comparable ($p > 0,05$) pour les deux aliments concentrés: $7,17 \pm 1,17$ pour le lot témoin (C) et $7,38 \pm 1,39\%$ pour le lot expérimental (S). La concentration de l'acide linoléique conjugué (CLA) était $0,48 \pm 0,03$ et $0,36 \pm 0,09\%$ respectivement pour les lots C et S. Les acides gras totaux étaient comparables ($p > 0,05$) pour le lait des brebis des deux groupes.

Mots-clés. Acide gras – Acide linoléique conjugué – Brebis laitière – Urée – Qualité du lait.

I – Introduction

Currently, the Tunisian ovine population reached 4 million females (DGPDI, 2005), which is represented mainly by the Barbarine, Noire of Thibar and fine Queue races. These three breeds have been used for meat production. The Sicilo-Sarde, localised in the Northern areas of Tunisia (e.g. Mateur and Beja), is the only breed used for milk production (DGPDI, 2005). Although intensification of livestock in Tunisia is increasing, its integration with crops is limited. Integration of forage into rain-fed farming has not really succeeded.

The performances of dairy production of the Sicilo-Sarde ewes depend largely on food complementation especially during the gestation and suckling (Bocquier and Caja, 2001) periods. In the last few years, the economic world conjuncture involved rising prices of corn and soybean meals which constitute the basic raw materials in concentrated food formulations for Tunisian livestock.

However, with the new policy guidelines in agricultural, the research for alternatives feeds is imperative (Rouissi *et al.*, 2008). The aim of our study was to evaluate the effect of the supplementation with local feed resources (white sorghum and faba bean) on production and quality of Sicilo- Sarde ewe's milk.

II – Materials and methods

1. Animals and diets

Twenty (20) Sicilo-Sarde ewes were divided into two homogeneous batches according to age (5.3 ± 1.25 years as against 5.7 ± 1.15 years), the litter size (1.1 ± 0.31), the rank of lactation (4.3 ± 1.25 vs. 4.6 ± 0.96) and live body weight (33.83 ± 5.63 vs. 33.95 ± 5.58 kg) received a common base ration (oat hay) at 1.5 kg DM/ewe/day supplemented by a 500 g/ewe/day of a standard concentrate [C: 10% barley, 43.3% corn, 25% wheat bran, 17.7% soybean meal, 4% sheep Vitamin and Mineral Mixture (VMM)], or an experimental concentrate (S: 66% white sorghum, 30% faba, 4% sheep VMM). Two weeks of adaptation take place before starting the measurements for 10 weeks control. The chemical composition (organic matter (OM), total nitrogenous matter (CP), crude fiber (CF) and lipids (FAT) (AOAC, 1990) and the nutritive values (Sauvant, 1981) of concentrates and hay are given in the Table 1.

Table 1. Chemical composition and nutritive value of feeds (% DM)

	Concentrate feed		Oat hay
	C	S	
DM %	94.7	94.7	92
Organic matter	91.0	88.3	92.1
Crude protein	16.3	14.65	4.9
Crude fiber	12.7	3.7	35.6
FAT	2.8	7.1	1.8
UFL/Kg DM	0.98	0.99	0.47
PDIN(g/kg DM)	99	96	33.6
PDIE (g/kg DM)	104.9	95	63.9

2. Milk sampling and analysis

Throughout the period of the experiment, milk production was recorded daily with 2 manual milking per day. The study of milk quality was performed once a week on a sample of bulk milk of each batch. During the whole trial milk samples have been collected from each group. The following milk quality parameters (pH, CP, FAT, urea) were determined by Lactoscan (Milkotronic LTD, serial No. 4696, Hungary) and urea by the DMAB method (JOCE, 2009).

Fatty acids in milk were determined by gas chromatography. A sample of 30 g, 200 ml of chloroform and 100 ml of methanol are added and homogenized vigorously for 2 minutes. The mixture was filtered with a filter bucher paper. The residue was dispersed in chloroform-methanol (2/l, v/v, 300 ml), mixed for 3 minutes and filtered again . The remaining solids were washed with chloroform-methanol (2/l, v/v, 60 ml). All filtrates were collected in a funnel. The filtrates were added to 0.2 volume of a solution of NaCl 0.7%. After the formation of two phases. A supernatant containing the non- lipid compounds and an organic phase contain substantially all of the lipids. The organic phase was collected in a calibrated flask and the solvent was removed at 50°C in a rotary vacuum evaporator. The fat content was determined gravimetrically.

In a 20 ml tube, 10 mg of lipid extract, 1 ml hexane and 2 ml 0.5 M sodium methoxide (1.35 g in 50 ml methanol). The mixture was vortexed and placed 15 minutes at 50°C in a heating block and allow to cool and then added 1 ml of methanol/HCl 5% v/v (to be prepared extemporaneously) and was vortexed vigorously. The solution was centrifuged at a rate of 3000 rpm for 5 minutes and recovered hexanoic stage transfer in chromatography vials .The spectrophotometer was adjusted to zero through the reagent blank and read the absorbance at 420 nm. Before the analysis of milk samples by chromatography for fatty acids determination, extraction of lipids took place following the method of Folch *et al.* (1957). The device used was a gas chromatograph FID type of fatty acid methyl esters, with a column OMEGAWAX 250. The temperature at the detector and the injector was 220°C while the level of the column was programmed from 45°C to 190°C.

3. Statistical analysis

The results of the effect of diets on the milk quality parameters were subjected to a one way analysis of variance using the GLM procedure in SAS (1989).

III – Results and discussion

The fat content of milk was $7.58 \pm 0.6\%$ and $7.21 \pm 0.41\%$ for the experimental (S) and the control groups (C), respectively. Statistical analysis revealed that there was no difference between the two groups ($p > 0.05$). This result is consistent with that reported by Rouissi *et al.* (2008) and Selmi *et al.* (2010). This result can be explained by the high energy content in white sorghum seeds and their fat content compared to corn and by the nature of faba bean seeds which are more energetic because of the large amount of starch easily degradable in the rumen (Sauvant, 2004). The average fat content obtained in this trial is higher compared to that reported by Hammami *et al.* (2009) who found an average fat value ranging from 5.34 to 5.83% by supplying a concentrate based on barley during the lactation. This explains the results of Barrillet *et al.* (2002) who reported that the fat content is influenced by the amount of milk produced (decrease causes an increase in these rates) and length of use (the end of the milking phase is accompanied by an increase in the fat content).

The protein content of milk was statistically comparable between diets ($p > 0.05$), ($6.04 \pm 0.57\%$ and $5.86 \pm 0.54\%$ for S and C, respectively). This result converges with those of

Santos *et al.* (1998). This is attributed to the fact that the milk protein content is positively correlated with the energy balance of the diet since energy intake stimulates the synthesis of microbial protein in the rumen (Bocquier and Caja, 2001). The comparable effect of soybean meal and faba bean as protein sources in feed concentrates can be explained by the wealth of faba bean seeds in essential amino acids such as lysine and methionine (Bocquier and Caja, 2001).

Statistical analysis showed that the lactose content is not significantly different ($p>0.05$) according to the dietary regime. However, the average content of lactose in milk of ewes of experimental group is slightly higher compared to that of the control sheep ($4.27\pm 0.43\%$ for S and $4.15\pm 0.5\%$ for C, respectively). This agrees with the result found by Rouissi *et al.* (2008). It is about $4.27\pm 0.43\%$ for S and $4.15 \pm 0.5\%$ for C, this The slight difference in milk lactose content between S and C groups could be due to the richness of faba bean in easily degradable carbohydrate (starch) present especially at the seed coat of faba bean by comparing it with that of soybean meal (Pottier, 2002).

The content of urea in milk is considered as an indicator of protein utilization (Cannas *et al.*, 1998). The average values were 53.5 ± 8.76 and 35.5 ± 3.4 mg/dl respectively for C and S groups ($P<0.01$). This result is consistent with that found by Lagriffoul *et al.* (1999) and different from that reported by Maâmourî *et al.* (2009) who supplied diets rich in tannins. The ratio of protein/energy in the diet that appears to be the factor having the greatest nutritional impact on the rate of urea (Cannas, 2002). This trend could be attributed to soya bean meal rich in protein degradable in the rumen compared to faba beans that contain a large amount of starch but also influences negatively urea concentration. Indeed, the energy level affects the amount of protein and non protein nitrogen to be used by micro-organisms (Benazzouz *et al.*, 2007). Thus, an increase in energy intake in the diet will cause a decrease of urea on the one hand and secondly the tannin content in the integument of faba beans may reduce protein degradation and therefore reduce the amount of urea.

The influence of the inclusion of white sorghum in the ewe's concentrate as an energy source to replace corn was assessed through the analysis of fatty acids in The statistical analysis revealed that there were no differences among fatty acids in milk of both ewe groups regardless of the regime (Table 2). The main saturated fatty acids were palmitic acid C16 ($27.75\pm 1.29\%$ vs. $27.77\pm 1.21\%$ of total FA for diets S and C, respectively) and miristic acid C14 ($12.06\pm 0.82\%$ for C and $12.47\pm 1.21\%$ for S). White sorghum as an energy source rich in fat (Pottier, 2002) maintained the concentrations of key fatty acids compared with corn energy source commonly used as a main ingredient in concentrates made for sheep The stearic acid (C18: 0) represented respectively 6.9 ± 0.74 and $6.82\pm 0.82\%$ of TFA in milk of the C and S ewe groups. The concentration of the oleic acid C18: 1 n-9 was not affected by the diets (20.7 ± 3.2 and $22.5\pm 4.6\%$ for C and S groups respectively; $p>0.05$). The capric acid content was 7.17 ± 1.17 and $7.38\pm 1.39\%$ ($p>0.05$) in the group C and group S respectively. These contents are higher compared to those reported by Luna *et al.* (2005) and Collomb *et al.* (2006) who worked on at different levels of altitude.

The CLA content of sheep milk is dependent on feeding conditions and the nature of the complementation. The concentration of CLA from this study was similar to that reported by Atti *et al.* (2006) who fed ewes with a diet based on concentrate feed but was lower compared with milk from sheep grazing on ryegrass or barley reported by the same authors in the same paper. CLA concentration was not different between the groups (0.48 ± 0.03 and $0.36\pm 0.09\%$, $p>0.05$); this shows that the white sorghum is a good alternative to corn because of the important amount of fat in its seeds.

The percentage of saturated fatty acids was higher compared to the polyunsaturated fatty acids, regardless of the diets. This can be explained by the biohydrogenation of PUFA in the diet. The percentage of monounsaturated fatty acids was 23.9 ± 6.1 and 25.33 ± 4.76 , for C and S, respectively, with no statistical difference ($p>0.05$). All the observed results on milk

fatty acids composition could be explained by the comparable fat composition of maize and white sorghum that are rich in linoleic acid (1305 mg), oleic acid (964 mg) and palmitic acid (407 mg). The ratio PUFA/SFA is similar for both groups ($p>0.05$) and compared to that observed by Atti *et al.* (2006) who worked on sheep fed on pasture. This can be explained by the significant contribution of polyunsaturated fatty acids in feed concentrates compared to forage that are rich in short chain fatty acids (Table 2).

Table 2. Concentration of Fatty acids profile and CLA in Milk (% of total FA)

	C	S	Pr >F	SEM
Capric Acid (C10)	7.17±1.17	7.38±1.39	Ns	0.4
Miristic Acid (C14)	12.06±0.82	12.47±1.21	Ns	1.03
Palmitic Acid (C16)	27.75±1.29	27.77±1.21	Ns	1.06
Stearic Acid (C18)	6.9±0.74	6.82±0.98	Ns	0.87
Conjugated Linoleic Acid (CLA)	0.48±0.11	0.36±0.1	Ns	0.11
SFA	65.76±4.04	66.84±4.92	Ns	0.96
MUFA	23.9±6.1	25.33±4.76	Ns	0.84
PUFA	3.88±1.1	3.7±0.8	Ns	0.56
SCFA	14.62±4.21	14.54±5.35	Ns	1.1
LCFA	53.17±2.42	53.67±3.63	Ns	0.57
PUFA/SFA	0.59	0.55	Ns	0.73
FA omega-3	0.369	0.364	Ns	0.44
FA omega-6	0.09	0.15	Ns	0.82
Ratio n-6/n-3	0.32	0.46	Ns	0.67

SFA: Saturated fatty acids; MUFA: mono-unsaturated FA, PUFA: poly-unsaturated FA; SCFA: Short chain FA; LCFA: Long chain FA. SEM: standard error of the mean; Ns: not significant ($p>0.05$).

IV – Conclusions

The results from this experiment suggest that the effect of the incorporation of local raw materials instead of imported raw materials in the formulation of concentrate feed can maintain or improve some physico-chemical characteristics of Sicilo-Sarde ewes' milk, including its fatty acid concentrations. The higher content of urea in the control group put into evidence the waste of protein used in that diet and the coverage rate of nitrogen needs of this sheep. The elevated urea level in the milk receiving the concentrate feed with a soy protein source reflects the quality level of soybeans protein in one hand, and in the other hand the presence of anti nutritional substances in faba beans.

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