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Improving milk antioxidant activity and nutritional composition through the incorporation of *Argania spinosa* (L.) by-products in goat's diet

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Abstract. Many studies reported that beneficial bio molecules compounds have been identified from various parts of the *Argania spinosa* (L.) Skeels and can play an important role in fighting diseases. Certainly, Morocco includes the most important rate of production, but the different by-products of this tree are not valorised. In this present study, we investigate the effect of the incorporation of the Argane tree's by-product in goat's diet, on the antioxidant activity, the microbiological quality, the physicochemical and mineral composition of Alpine goat milk collected from Essaouira, Morocco. The preliminary results of this study revealed a remarkable improvement in several of the analysed parameters, especially the fat content with an increase of 51.21%, which remains the most noteworthy. Microbiological analysis showed that the mean mesophilic aerobic flora count for goats consuming the Argane by-product diet (AD) was $1.38.10^7 \pm 2.67.10^7$ Cfu.mL^{-1} comparing to $2.96.10^7 \pm 5.42.10^7$ Cfu.mL^{-1} for those consuming the Control Diet (CD). The total phenolic content of milk collected from goats consuming AD varied from 17.29 ± 0.61 to 22.65 ± 1.15 mg GAE/g of DM, while that the total phenolic content of milk collected from goats consuming CD varied from 10.74 ± 0.39 to 12.46 ± 0.33 mg GAE/g of DM. Furthermore, mineral analysis demonstrated that AD milk samples are rich in potassium, magnesium and other minerals with many health benefits.

Keywords. Goat milk – *Argania Spinosa* – By-product – Antioxidant activity.

Amélioration de l'activité antioxydante et la composition nutritionnelle du lait de chèvre par l'incorporation des co-produits d'*Argania Spinosa *dans la ration des chèvres

Résumé. *De nombreuses études ont montré que des composés de biomolécules bénéfiques avaient été identifiées dans différentes parties de l'*Argania spinosa *(L.) Skeels et qu'elles pouvaient jouer un rôle important dans la lutte contre les maladies. Certes, le Maroc comprend le taux de production le plus important, mais les expériences de culture dans de nombreux pays, sont prometteuses, et les différents co-produits de cet arbre ne sont pas valorisés. Dans la présente étude, nous étudions l'effet de l'incorporation des co-produits de l'arganier dans l'alimentation des chèvres, sur l'activité antioxydante, la qualité microbiologique, la composition physicochimique et minérale du lait de chèvre alpines collecté à Essaouira - Maroc. Les résultats préliminaires de cette étude ont révélé une amélioration remarquable de plusieurs paramètres analysés, notamment le taux de matière grasse avec une augmentation de 51,21%, qui reste le plus remarquable. Les analyses microbiologiques ont montré que la valeur moyenne de la flore aérobie mésophile totale chez les chèvres consommant le régime contenant les co-produits d'Argane (AD) était de 1,38.107 ± 2,67.107 Cfu.mL^{-1}, comparé à 2,96.107 ± 5.42.107 Cfu.mL^{-1} pour le lait du Régime contrôle (CD). La teneur en composés phénoliques totaux du lait des chèvres consommant la ration alimentaire à base des co-produits de l'arganier variait de 17,29 ± 0,61 à 22,65 ± 1,15 mg GAE / g de matière sèche, tandis que la teneur en composés phénoliques totales du lait des chèvres consommant la ration contrôle variait de 10.74 ± 0.39 à 12.46 ± 0.33 mg GAE/g de matière sèche. En outre, l'analyse des minéraux a montré que les échantillons de lait issu des chèvres consommant le régime AD étaient riches en potassium, magnésium et autres minéraux offrant de nombreux avantages pour la santé.*

Mots-clés. *Lait de chèvre – Co-produits –* Argania spinosa *– Activité antioxydante.*

I – Introduction

In Morocco, goat population is estimated at 5.3 million head, occupying the 2nd place in local farming (Zaaraoui *et al*., 2016), and the 13th place on a world scale (El Moutchou *et al*., 2013).These goats are adaptable to the climatic conditions and ensure the production of several products such as meat, milk and skins (Badis *et al*., 2004). Goat milk represents a food of high nutritional value, due to its composition and recognized benefits for human well-being (Lakram *et al.,* 2019), Therefore, goat milk is considered to possess a high antioxidant potential that resists to oxidative stress which is an important characteristic of numerous cardiovascular related diseases (Alyaqoubi *et al*., 2014). However, livestock production in Morocco suffers from serious feeding problems, during the limited forage availability period, due to shortage, low protein content resources and lack of development of alternative feed resources (Mercha *et al.,* 2019). Among these unconventional alternative resources is the Argan press oil cake that represent an excellent nutrient source with 43.1 % of protein and 21.3% of crude fiber (Mercha *et al.,* 2019). It is therefore an efficient alterative feed resource to improve production and nutritional value of goat milk and its integration in human diets, with health promoting components. This work aims to study the effect of the incorporation of argan by-products in dairy goat on antioxidant activity and nutritional composition of goat milk produced in Essaouira region.

II – Material and methods

An experiment was carried out in the region of Ounagha (Essaouira, in the region of Marrakech, Safi), during. a period of 90 days, on two groups of twelve (12) alpine goats , divided randomly, each with an average age of 2 years. The first group of goats consumed a diet prepared basically from by-products of the Argane tree (press cake (25%) and pulp (20%) as dry matter basis (DM)), while the second fed a diet without Argane by-products (control group). Both groups were receiving 1.5 kg of prepared diets twice a day and a fifteen (15) days adaptation period was respected.

The tested diet is composed of Argane press cake , Argane pulp , wheat bran , barely , chopped straw and a complex of minerals and vitamins (Table 1), Dry matter (DM), ash, ether extract (EE), crude fiber (CF), and crude protein (CP) were determined according to the Association of Official Analytical Chemists methods (AOAC , 2011).The animals were fed normally, and the water was available all time. The nutritional balance was established according to the energy and protein requirements of ewes in the lactation phase following the INRA system.

Table 1. Composition of the experimental diets

	Argane Diet (AD)	Control Diet (CD)
Ingredients (g/100 g of fresh matter)		
Argane press cake	250	0
Argane pulp	200	0
Ground straw	100	170
Wheat bran	250	180
Barley grain	180	200
Minerals and vitamins	20	0
Commercial feed	0	250
Lucerne	0	200
Chemical composition, % (DM basis)		
Crude protein	16.4	11.5
Dry matter	87.2	78.7
Ash	6.56	7.37
Crudefiber	12.9	10.30
Ether extract	3.50	2.12

Raw goat milk samples were collected twice at morning and evening periods in sterile bottles and maintained at 4°C condition during their transport to the food technology laboratory of regional center of agronomic research- INRA, rabat. The pH of the milk samples was determined electrometrically with a pH-meter (Micro pH 2002, Crison, Barcelona, Spain). The physicochemical parameters (fat, solids-non-fat, density, protein, lactose, salts and freezing point were measured using a Lactoscan Milk Analyzer calibrated for goat milk.Thus, a microbiological quality follow-up was provided on the day of the collection of themilk samples, for a count of contaminating microorganisms following the International Dairy Federation protocol (IDF.1987), to be able to evaluate the effect of the incorporation of the prepared food diet on the development rate of bacterial strains.

The antioxidant capacity of each sample through total phenolic content TPC was determined according to (Shori *et al.,* 2011) with minor modification. 200 µl of fresh goat milk extract was added to 1 mL diluted Folin – Ciocalteu reagent and 800 µl of Sodium carbonate solution. After 2 hours incubation, the absorbance was measured at 765 nm wavelength.

Trace elements of milk, including iron, copper, manganese, and zinc, were analyzed by atomic absorption spectrometryin a flame air-acetylene. The measured absorption was done at a specific wavelength of 248.3 nm.

All analysis were carried out in triplicate, data was statistically treated using variance analysis (ANOVA), results are shown as mean, and differences among means were ranked using Duncan's New Multiple Range Test. The significance level was 5%.

III – Results and discussion

1. Milk physicochemical and microbiological quality

Milk quality and quantity are mainly affected by several factors including breed, stage of lactation, milking system and sheep feeding (Mouhaddach *et al*., 2016). Physicochemical results of analyzed milk samples collected during the trial are presented in Table 2. The incorporation of the Argane by-product's diet offers a milk with more fat (4.20 % compared to 2.92% for the control group), more protein content (3.27 % compared to 2.56 % for the control group). For all criteria, the composition of milk from goats fed by the Argane diet exceeded those from the control animals. Our research shows that the number of microbes of technological interest is very high, and this presents an economic and qualitative advantage for industrialists (Lakram *et al*., 2019).

Table 2. Physicochemical content of goat milk from Argane and control diets

	Argane Diet	Control Diet	SEM	*p*
Density, g/cm^3	1.028^a	1.019^b	0.09	<0.001
Fat, %	4.20^a	2.92^b	0.05	<0.001
pH	6.67^a	6.61^b	0.299	<0.001
Protein, %	3.27^a	2.56^b	0.20	<0.001
Lactose, %	4.16^a	4.04^b	0.72	<0.001

Microbial quality is important for milk preservation and its transformation. The results obtained by an enumeration of the different microbial flora of raw goat milk samples are summarized in Table 3.

Overall, the microbiota of raw milk can be influenced by several factors, including animal feeding, seasonal variation, geographical farm location and milking procedures. Since all the factors were stabilized, our results showed that the feeding factor had an effect on goat milk flora. Microbial counts in the AD were lower than those of CD, likely due to the presence of antimicrobial factors in the Argane diet.

Table 3. Microbiological analysis (mean)in function of diet type

	Argane Diet	Control Diet	SEM	*p*
Total aerobic flora	$1.1{\cdot}10^{4\ b}$	$1.1{\cdot}10^{5\ a}$	0.10	<0.001
Total coliform	$3.1{\cdot}10^{4\ b}$	$1.1{\cdot}10^{5\ a}$	0.74	<0.001
Fecal coliform	$2.5{\cdot}10^{5\ b}$	$4.2{\cdot}10^{5\ a}$	0.26	<0.001
Staphylococcus	$3.8{\cdot}10^{1\ b}$	$5.7{\cdot}10^{1\ a}$	0.95	<0.001
Yeasts and molds	$2.8{\cdot}10^{2\ b}$	$1.6.10^{4\ a}$	0.47	<0.001

2. Total phenolic and mineral content

Many studies reported that beneficial bio molecules compounds have been identified from various parts of *Argania spinosa*'s (L.) Skeels and can play an important role in fighting diseases and could be used in pharmaceutical and personal care products (Elbabili *el al.*, 2010). Figure 1 illustrates phenolic contents of both diets.

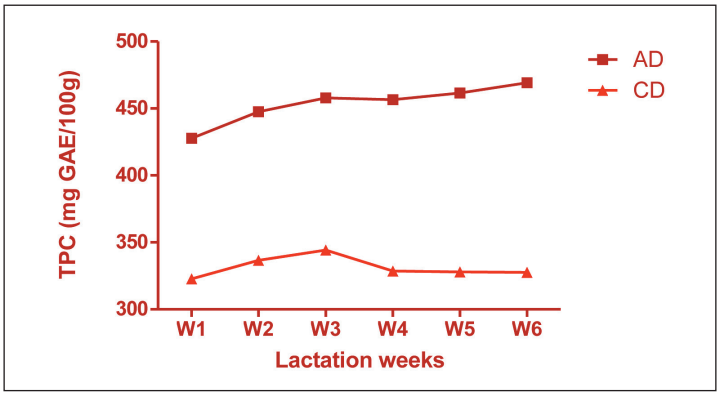

Fig. 1. Phenolics contents of goat milk (mg GAE/100 g DM) as related to diet type and lactation stage. Data are presented as mean ± SEM, n = 3 experiments, p values; *p < 0.05, **p < 0.01, ***p < 0.001. AD: milk collected from goat fed by diet including Argane by- products; CD: milk received from goat fed by local diet.

Among the diets offered, the experimental diet based on Argane byproducts supplied the largest amount of total phenolic compounds with averages of 447.28 ± 1.43 mg GAE/g for AD group, and 344.78 ± 1.26 mg GAE/g for CD group. These results are in accordance with those reported by (Alyaqoubi *et al*., 2014).

Following previous studies, milk and dairy products supply all essential mineral elements needed for newborn development and growth. In this work animal feed and lactation week had a significant effect (P < 0.001) on goat milk trace elements (Cu, Zn, Mg and Fe). Milk iron is naturally low. In our case, the average values of milk iron were 0.09, and 0.05 mg/100 g for the AD, and CD diets, respectively. The iron concentration reported in the literature for goat milk was 0.07 mg/100 g (Park *et al.,* 2007).

IV – Conclusion

The present study revealed that the inclusion of the Argane by-products in goat's diet improved the global quality as well as the antioxidant potential of collected milk, in addition to its mineral composition. Therefore Argane by-products must be considerated as a promising alternative for dry season feeding systems and, thereby undoubtedly reduce the pressure on silvopastoral resources and production costs and consequently improve the income of livestock farmers in dry areas. However, further testing is required on secondary metabolites production in collected milk.

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