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A COMPREHENSIVE CHARACTERIZATION OF GUELMOISE, A NATIVE CATTLE BREED FROM EASTERN ALGERIA

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Abstract

The present study was a global and a first characterization (Typology, biometric, biochemical and contribution on the genetic characterization) of the Guelmoise local cattle breed. A structured questionnaire surveys were carried out in 42 farms spread over the four wilayas of the north-eastern region of Algeria in order to characterize the local cattle population of Guelmoise and their mode of rearing. These surveys focused on the characteristics of the farms (socio-economic condition of the farmer, mode of breeding, reproduction and also to take samples of milk and blood). A total of 164 adult animals were selected for morphological description and body measurements (148 females and 16 males). The livestock system has an extensive nature and men are the main responsible for these farms. The height at withers (HaW), body length (BL), Heart Girth (HG), Spiral Tower (ST) and estimated weight (W) in Guelmoise cattle were 115 ± 0.07, 118 ± 0.08, 154 ± 0.09, 187 ± 0.11 cm and 258 , 12 ± 43.12 kg respectively in females and 171 ± 0.16, 176 ± 0.18, 187 ± 0.08, 225 ± 0.08 cm and 462 ± 59.67 kg respectively in males. Multivariate statistical analyses have allowed identifying four different classes were defined as follows: morphometric measurements, weight and age in females as in males. Estimated age at first calving was 39.40 months while the calving interval varies between 14 to17 months and the reproductive life of the cows (13,7 years).The average daily production of milk 5,55 ± 1,67 liters in spring with a duration of lactation varies between 6 to 7 month. The physico-chemical characteristics of milk were focused on the Fat content (FC), Protein content (PC), Lactose (Lact), Dry extract (DE), Defatted dry extract (DDE) and freezing point depression (FPD) were respectively 2.85 ± 0.89%, 3.60 ± 0.39%, 4.82 ± 0.13%, 11.84 ± 0.90%, 9.13 ± 0.41% and 0.56 ± 0,01°C. The genotyping of 24 individuals of the Guelmoise population using the Illumina BovineSNP50 BeadChip, we found that the observed heterozygosity was globally equivalent to 0.30. The inbreeding measured by Fis was slightly positive (~ 0.04) indicating the absence of a phenomenon of selection. The data contribute to the feasibility of a conservation and selection programme for this breed and the results are useful for the implementation of a conservation strategy that should aim to conserve animals. The Guelmoise cattle can be used as an alternative genetic resource for production improvement programs.

Keywords: Cattle, Guelmoise, morphological characterization, milk analysis, genetic characterization, SNP, Algeria.

Introduction

Guelmoise is Algerian autochthonous bovine cattle, generally found in mountainous and forested areas in northeastern Algeria. It is a small sized animal, particularly hardy and well adapted to difficult environmental conditions. It possesses valuable breeding attributes, such as resistance to infectious diseases and parasites. (Geoffroy S.H., 1919, Benyoucef M.T., 1986, Aissaoui C and al., 2003). Adults

are mainly light- to dark-gray coat-coloured although some pale- to dark-reddish animals may be observed. Head, neck, extremities and tail tips are usually darker, with a light muzzle ring and black nasal mucosa. Calves are usually born fawn and change to the adult coat color after few months (**Figure 1**).

Figure 1. Phenotypic appearance of the Guelmoise cattle: a typical grey coat-coloured adult animal (on the left) and a fawn calf with its mother (on the right).

Like all native cattle in North Africa, it belongs to the type *brown of the Atlas*, its main ancestor being *Bos primigenius mauritanicus*. Several authors (Geoffroy SH., 1919, Diffloth., 1924, Sadler S., 1931 and Magneville Y., 1949) affirm that the *brown of the Atlas*, and hence Guelmoise, represent a variety of the Iberian cattle. Despite local cattle breeding plays an important role in the family economy of rural households in Algeria, it remains among the least modernized sectors and, in recent years, there has been a gradual decline in livestock numbers. The rearing system of Guelmoise is of an extensive, or semi-extensive, nature, based on the pasture as the primary source of food (Abdelguerfi and al., 2000), with the specificity of being independent of public aid (Ghozlane F et al., 2010).

According to FAO (2012), description of morphological and phenotypic traits, combined with reconstruction of the genetic structure by the use of molecular markers, allow to characterize, identify and differentiate populations, which represent the first step toward a sustainable use of animal genetic resource (FAO, 2011). To this regard, the characterization of the breeding system in its Specific environment also represents a fundamental knowledge for the definition and implementation of a suited conservation program. Hence, the present study aims to characterize the local Algerian Guelmoise breed using a comprehensive approach based on (i) analysis of its breeding system; (ii) morpho-biometric characterization of Guelmoise cattle; (iii) study of some physicochemical parameters of milk and (iv) Analysis of the genetic diversity and structure of the Guelmoise population by genome-wide SNP markers.

Materials and Methods

The study area

The study was performed over four Algerian administrative provinces (wilayas) in north-eastern Algeria (Figure 2): El-Tarf, Annaba, Souk Ahras and Guelma, characterized by different geo-pedological conditions, making up areas of different terrain (mountain, plain and lakes). The climate is sub-humid to mild-wet, with temperature ranging 2°C to 4°C in winter and 25°C to 30 °C in summer. The annual rainfall is between 600 and 1000 mm.

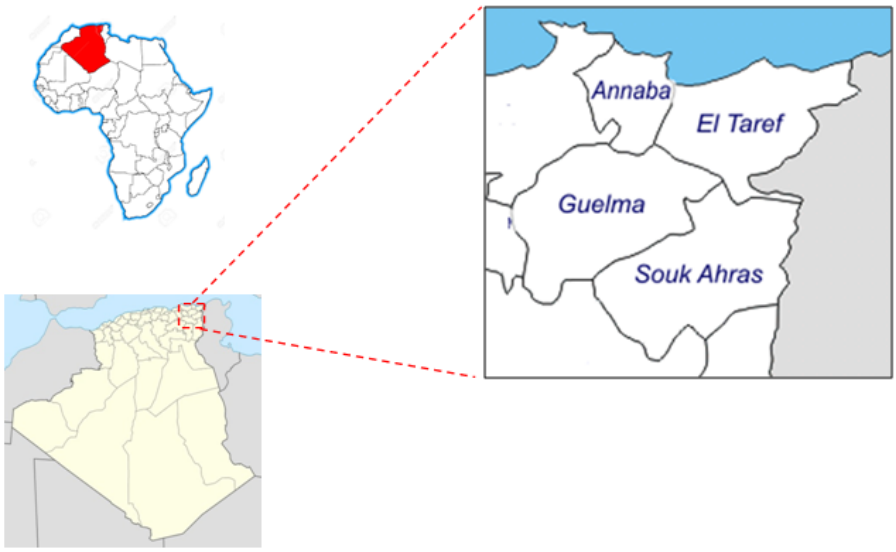

Figure 2. Localization of the study areas.

Field investigations and measurements

Surveys were carried out, on a total of 42 farms spread over the above-mentioned *Wilayas*, adopting a hybrid approach (written questionnaires and oral interviews), and focusing on (i) socio-economic status of the breeder, (ii) breeding system, (iii) reproduction and (iv) milk production . Morphometric measurements were performed on adult animals (aged >4 years), for a total of 148 cows and 16 Bulls. The considered variables were body length (BL), height at withers (HW), Spiral Tower (ST) and Heart Girth (HG) (FAO; 2012). Measurements were taken to the nearest centimeter on animals placed on a flat ground using a tape measure and a measuring stick. All measurements were carried out by the same person in order to avoid between-individual variations. To estimate the weight (W), we used the formula of Crevat**:** **W=** index × **HG**$^{3.}$ According to (Boulahbal JB, 1999) the weight index of our local populations is **69.54**, which allowed the establishment of the Crevat formula: **W= 69, 54× HG**^{3}.

Milk analysis

The study involved 58 cows of different farms, aged between 4 and 14 years, raised in an extensive system, and was performed in the El-Tarf region during the month of May 2016. Samples were collected in sterile vials and immediately transported to the laboratory for subsequent analyses.

The physico-chemical analyses of the milk samples were carried out at the Food Analysis Laboratory of the Biotechnology Research Center in Algeria**,** using a MilkoScan Minor (6) type 78110. The following parameters were measured: fat content (FC), protein content (PC), lactose (Lact), dry extract (DE), defatted dry extract (DDE) and freezing point depression (FPD).

Genetic characterization using genome-wide SNP markers

Twenty-four representative unrelated animals were selected from different farms. Whole blood samples were collected, for each animal, in 5 ml vacutainer tubes containing EDTA as anticoagulant and stored at - 20°C until subsequent analyses were performed. DNA extraction was carried out from whole blood according to the method described by Miller et al; (1988). Nucleic acids were extracted using a saturated NaCl solution. The DNA was picked up and washed in 70% ethanol. The DNA was hence dissolved in 1X TE buffer. DNA concentration was determined, using a NanoDrop1000 Thermo Scientific spectrophotometer, and then diluted to the working concentration of 50ng/µl. The samples were genotyped at 54609 Single Nucleotide Polymorphisms (SNPs) using the Illumina Bovine SNP 50K Bead Chip (Illumina, Inc.), following standard operating procedures recommended by the manufacturer. Genotyping was performed at the facilities of the Department of Agriculture and Foresty Sciences, University of Palermo, Italy. SNPs and animals were pruned using the following parameters: (1) SNP

call rate ⩽ 97%, (2) SNP minor allele frequency ⩽ 1% and (3) animals displaying ⩾ 10% of genotype missingness. Quality controls were performed using PLINK v1.07 software (Purcell et al., 2007).

Statistical analyses

Statistical analyses of data from morphological measurements and milk physico-chemical attributes were performed using XLSTAT software (Copyright 2014). We first carried out a descriptive analysis, and then a Multiple Correspondence Analysis (MCA), followed by a hierarchical clustering (HCA), was carried out to identify the classes.

The level of genetic diversity of Guelmoise cattle was estimated from the observed percentage of heterozygous genotypes per individual (Ho) and the expected heterozygosity (He) using PLINK (Purcell et al., 2007). The software was also used to estimate individual inbreeding coefficients (FIS).

Results and Discussion

Sociological status of the breeder

According to the surveys, the totality (N = 42) of the cattle breeders were males. The age of the breeders varied between 16 and 74 years. In terms of educational attainment, 9 (21.4%) were illiterate and, generally, old men, 27 (64.2%) received primary education, 6 (14.2%) had completed secondary studies.

Cattle management practices

The Guelmoise management system is generally semi-extensive. Housing of the Guelmoise animals consists of simple traditional structures **(Figure 3)**, usually called '*Zriba*', made up of reeds and tree trunks, and roughly built by the cattle owner itself, who, in the majority of cases, does not give much importance to the control of climatic factors. The animals are so rustic that, in the mountains areas, they can spend most of the winter in the forest.

Figure 3. Examples of typical cattle sheds ("*Zriba*") for the Guelmoise cattle.

Cattle feed composition is strongly affected by the altitudinal zone (BNEDER, 1978, Boulahbal, 1999). Feed of animals living in the mountainous areas mainly consists of foliage and shrubs. The farmer drives his herd in the morning in the forest and then returns in the evening to recover them. In some forest areas, the breeders do not even need to perform this daily ritual, as the animals come and go instinctively (Yacheur, 1986; Benchaar, 1987; Kacha, 1988). Animals living around lakes are generally feed on natural or flooded meadows (Figure 4).

Figure 4. Examples of natural (on the left) and flooded (on the right) meadows.

Animals browsing in mountainous forest areas are less affected by seasonal variation in feed availability as they can find poor, though year-round, pasture. On the contrary, in the plains and around the lakes, winter represents the unfavorable season in terms of pasture availability. Generally, spring is synonymous with grazing abandoned. Dairy production is increasing because of the abundance of green grass and the animals recover weight quickly.

Herds' size per farm is around 8-50 cows (on average, X±DS). Cows are mainly kept to provide milk for the household while calves are raised and sold to allow additional incomes.

Among the parameters related to cattle reproductive and productive efficiency, a special attention is given to age at first service, age at first calving, total milk yield, average milk yield per day and calving interval (Dematawawa et al., 1998). The mean age at puberty in this study is 30.1 months for female and 36. 2 months for male Guelmoise cattle. Our results are lower than 55.6 and 57 months reported for Semien and Wegera female cattle (Wuletaw, 2004), respectively, and 42.6 months for Mursi cattle in Southwest Ethiopia (Terefe et al.; 2015). This variation may be due to different feeding management and genetic makeup of the different cattle breeds.

Mean age at first calving (AFC) for Guelmoise breeding females in the present study was 39. 40 months. Our results is lower than some indigenous cattle types reported by (Taye, 2005) who reported 54.1 months for Sheko breed whereas, (Tadesse, 2005) reported 53.1 months for Raya-Sanga cattle and 54.8 months for Mursi cattle (Terefe E and al; 2015). The differences in the reproductive performance of indigenous Guelmoise cows **(Table 1**) might be attributed to the differences in nutritional and reproductive managements among the smallholder cattle keepers in different parts of the country.

The mean reproductive lifetime of Guelmoise cow reported to be 13.7 years. Within this reproductive lifetime, the mean number of calves born per cow was estimated to be nine. The mean reproductive lifetime of Guelmoise cow is the same as the taurine Sheko cattle breed (Taye et al., 2007); however, the number of calves born in a lifetime of the Sheko cow is eight calves and is lower than the Guelmoise cow. The large number of calves per cow in Guelmoise cattle might be due to breed and management differences, with Guelmoise cattle being managed in open grazing compared with the restricted feeding management of Sheko cattle (Taye et al., 2007). This uncontrolled breeding results in a longer reproduction season of Guelmoise cattle as compared with the controlled breeding system (Mekonnen et al., 2012).

Table 1. Mean and standard error (SE) of reproductive performances of the Guelmoise cattle

Reproductive trait	Mean ±SE
Age at puberty in males (months)	36.20 ± 0.20
Age at puberty in females (months)	30.10 ± 3.40
Age at first calving (months)	39.40 ± 1.20
Calving interval (months)	15.20 ± 2.10
Female reproductive life (years)	13.70 ± 2. 40
Male reproductive life (years)	7.20 ± 1.60

Body measurements

Females - The average height at withers (HW), body length (BL), heart girth (HG), spiral tower (ST) and estimated weight (W) in female Guelmoise cattle were 115 ± 0.07,118 ± 0.08, 154 ± 0.09, 187 ± 0.11 cm and 258.12 ± 43.12 kg, respectively (data not shown). The above results confirmed that Guelmoise cattle have a small size. Our measurements were similar to those reported by (Boujenane, 2015) on Oulmes-Zaer and Tidili cattle (115 cm for HW, 126 cm for BL), and (Endashaw et al., 2015) on Mursi cattle (122 cm for BL, 114 cm for HW and 145 for HG). Also, our results were lower than those found by (Ftiwi et al., 2015) on Indigenous Begait Cattle in Western Tigray (131.48 ± 0.25cm for HW, 128.13 ± 0.16 cm for BL and 159 .55 ± 0.24 cm for HG). However, our results were higher than those found by (Kanai et al., 2013) on Bunaji cows in Nigeria (151 cm for HG, 107 cm for BL and 249 for weight).

Significant positive correlations were observed between body weight and the linear body measurements (BL, HG, ST and WH) (**Figure 5)**. The highest correlation coefficient (0, 9705) was between body weight (W) and heart girth (HG) (**Table 2)**. The positive) correlation of weight with linear body measurements indicates that linear body measurements can be used as a marker to estimate weight for different purposes.

Table 2: Correlation matrix of body measurements in female animals.

Variables	Age	MP	HW	ST	HG	BL	WE
Age	1	0,1214	0,2013	0,2099	0,3013	0,2870	0,2719
MP (l/d)	0,1214	1	0,0775	0,0735	0,1229	0,0898	0,1127
HW (m)	0,2013	0,0775	1	0,4456	0,4971	0,4938	0,4880
ST (m)	0,2099	0,0735	0,4456	1	0,7053	0,5561	0,6774
HG(m)	0,3013	0,1229	0,4971	0,7053	1	0,6107	0,9705
BL(m)	0,2870	0,0898	0,4938	0,5561	0,6107	1	0,5589
WE (Kg)	0,2719	0,1127	0,4880	0,6774	0,9705	0,5589	1

The values in bold are different from 0 to a significance level alpha = 0.05

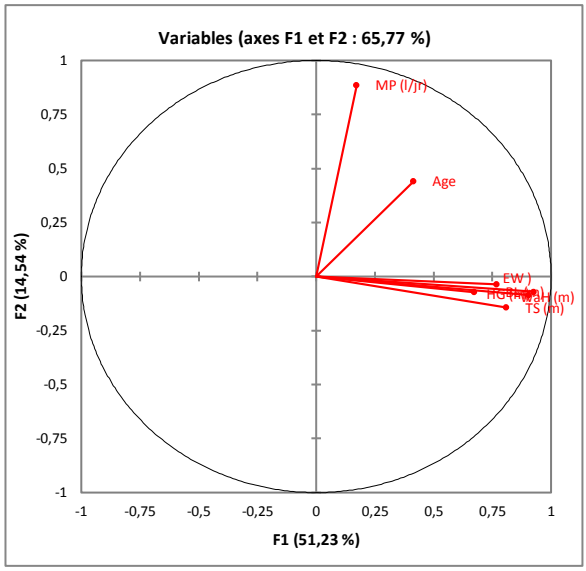

Figure 5. Projection of the morphometric variables characterizing the female animals of the Guelmoise population on factorial plane1-2.

A multidimensional analysis was hence performed. Seven quantitative variables, correlated with each other and illustrating the diversity of the population, were considered for principal component analysis (PCA) (**Figure 6)**. They were designed to accurately identify the reality of class choices (age, measurements, weight and milk production). Four classes were identified (**Table 3)**. Class (1) included 26 cows, aged ≥ 9 years, with a dairy production of 7 l/d, and body measurements values and weight lower than that of the second class. Class (2) included the largest number of individuals (60 cows), aged between 5 and 6 years, with the highest body measurements and weights compared to the other classes. Class (3) included 30 cows, aged between 5 and 6 years, with body measurements and weight close to the average. Class (4) included the youngest animals, i.e. 23 cows aged between 4 and 5 years, displaying the lowest values for body measurements and weight.

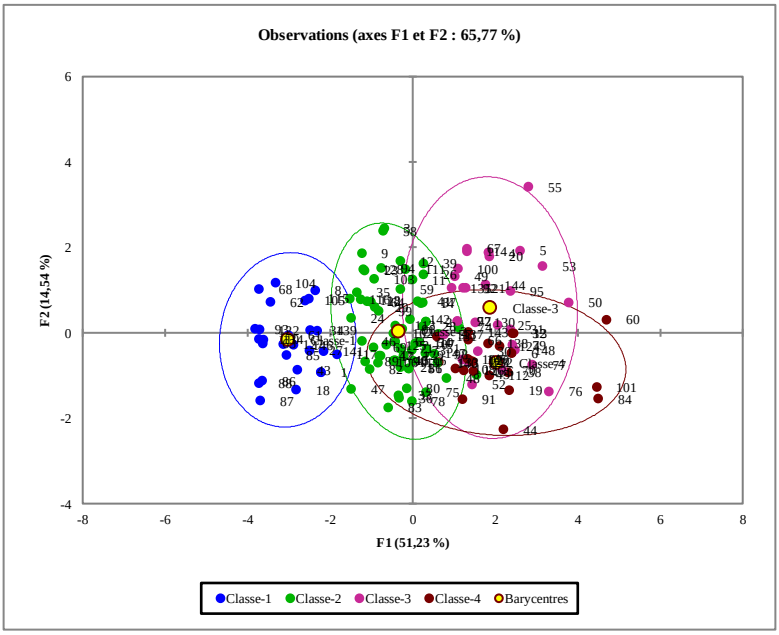

Figure 6: The different classes in female animals.

Table 3. Comparison of the different classes in females using an analysis of variance test.

	Class 1	Class 2	Class 3	Class 4
Age	9,1000[a]	5,1304[c]	6,0652[b]	4,5769[c]
MP	7,3000[a]	6,6522[a]	6,8116[a]	6,0000[a]
HW	1,1990[a]	1,2009[a]	1,1471[b]	1,0838[c]
ST	1,9193[b]	1,9978[a]	1,8696[c]	1,7238[d]
HG	1,6150[b]	1,6504[a]	1,5328[c]	1,4123[d]
BL	1,2600[a]	1,2291[a]	1,1651[b]	1,1015[c]
EW	289,7496[b]	312,7373[a]	249,5470[c]	196,1027[d]

The averages assigned to the same letter on the same line are not significantly different (p> 0.0001)

Males - Average height at withers (HW), body length (BL), heart girth (HG), spiral tower (ST) and estimated weight (W) in male Guelmoise cattle were 171 ± 0.16, 176 ± 0.18, 187 ± 0.08, 225 ± 0.08 cm and 462 ± 59.67 kg, respectively (data not shown). We found higher values compared to those found by **(**Ftiwi et al., **2015)** on Indigenous Begait Cattle in Western Tigray (136. 99 ± 0.10 cm for HW, 135.60 ± 0.09 cm for BL and 168. 91± 0.10 cm for HG) and (Endashaw et al., 2015**)** on Mursi cattle (129.3 ± cm for BL, 121.3 ± 1.9 cm for HW and 154. 6 ± 1.6 for HG). Significant (positive) correlations were observed between body weight and the linear body measurements (HG and ST) (**Figure7**). The highest correlation coefficient (0.9989) was between body weight (W) and heart girth (HG).

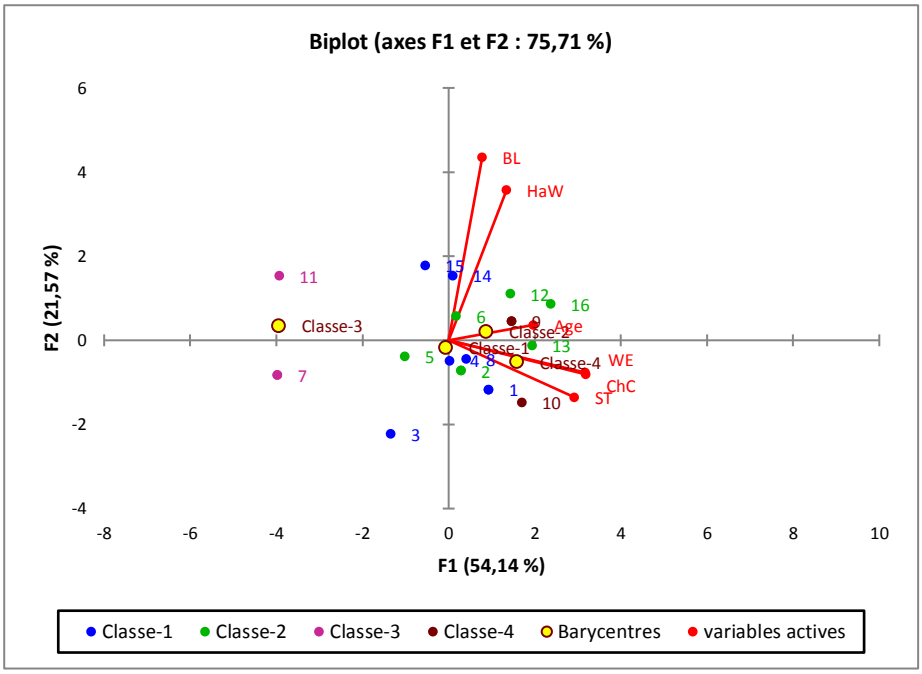

Figure 7. Projection of the morphometric variables characterizing males of the Guelmoise population on factorial plane1-2 and the different classes

Concerning males, three classes were identified. The differences between these classes were due to weight estimates (W), heart girth (HG) and age. Class (1) included 7 bulls, aged between 5 and 6 years, having an average weight of 512.631 kg and high body measurements compared to the other two classes. Class (2) also included 7 bulls, aged 4 to 5 years, with an average weight of 467.432 kg, and high body

measurements. Class (3) included 2 bulls displaying the smallest body measurements and weight (379,265 kg) compared to the other two classes (**Table 4).**

Table 4. Comparison of the different classes in males using an analysis of variance test.

	Class 1	**Class 2**	**Class 3**
Age	5,7143[a]	4,4286[b]	4,5000[b]
HW	1,7686[a]	1,7729[a]	1,7450[a]
ST	2,3029[a]	2,2743[a]	2,1100[b]
HG	1,9457[a]	1,8871[b]	1,7600[c]
BL	1,8357[a]	1,8143[a]	1,8000[a]
EW	512,631[a]	467,4329[b]	379,2650[c]

The averages assigned to the same letter on the same line are not significantly different (p> 0.0001)

Milk analysis

Average fat content (FC), protein content (PC), lactose (Lact), dry extract (DE), defatted dry extract (DDE) of the milk samples were respectively 2.85 ± 0.89%, 3.60 ± 0.39%, 4.82 ± 0.13%, 11.84 ± 0.90%, 9.13 ± 0.41% .

The mean freezing point (FPD) value of the Guelmoise cow's milk samples was 0.56 ± 0.01°C. The above values were close to those reported by **(**Imran et al., 2008**)** on the XX breed (PC and Lact being 3.28 ± 1.02 % and 4.38 ±1.36 %, respectively) but lower than those found by **(**Hamad et al., 2009**)** on "Baladi" cow's milk in Qena governorate (Egypt), where FC, PC and Lact were,, respectively, 4.28 ± 0.1, 3.37 ± 0.5, and 4.47 ± 0.58.

The dry extract (DE) is very important particularly in the cheese industry, since extracting the lipid fraction from the milk provides a better calculation of the protein fraction. 11.84 ± 0.90%,

The cow's milk DDE is, however, as in the milk of sheep, a very stable value. This may explain the similarity of our samples for this parameter.

Table 5. Correlation matrix of dairy traits.

Variables	**Month of lactation**	**Quantity**	**Age**	**Fat content**	**Protein content**	**Lactose**	**Dry Extract**	**DDE**	**FPD**
Month of lactation	1	-0,5015	0,1996	-0,5940	-0,0467	0,4016	-0,4528	0,1912	0,5176
Quantity	-0,5015	1	0,1784	0,1666	0,1233	-0,1369	0,2096	0,0157	-0,2435
Age	0,1996	0,1784	1	-0,2014	-0,0888	0,1220	-0,2118	-0,0482	0,1025
Fat content	-0,5940	0,1666	-0,2014	1	-0,0756	-0,2512	0,8375	-0,1960	-0,2944
Protein content	-0,0467	0,1233	-0,0888	-0,0756	1	-0,1205	0,2090	0,7310	-0,0897
Lactose	0,4016	-0,1369	0,1220	-0,2512	-0,1205	1	-0,2635	0,1875	0,6927
Dry Extract	-0,4528	0,2096	-0,2118	0,8375	0,2090	-0,2635	1	0,2038	-0,1355
DDE	0,1912	0,0157	-0,0482	-0,1960	0,7310	0,1875	0,2038	1	0,3454
FPD	0,5176	-0,2435	0,1025	-0,2944	-0,0897	0,6927	-0,1355	0,3454	1

The values in bold are different from 0 to a significance level alpha = 0.05

Significant (negative) correlations were observed between month of lactation and fat content (FC), quantity and dry extract (DE) (**Figure 8)**. The highest correlation coefficient (0.8375) was between dry extract (DE) and fat content (FC). A significant (positive) correlation found between the defatted dry extract (DDE) and protein content (PC). Also, the freezing point (FPD) was significantly correlated with the month of lactation, lactose (Lact) and defatted dry extract (DDE) (**Table 5)**.

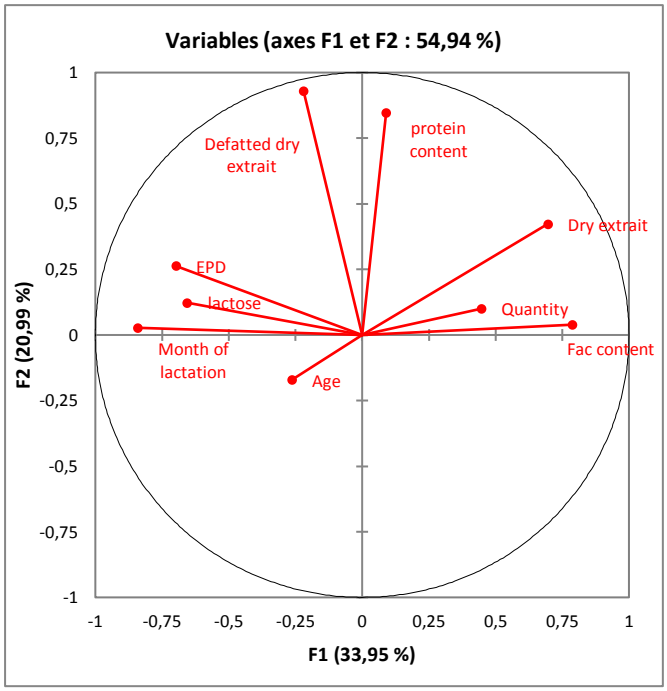

Figure 8. Projection of the variables characterizing the milk compositions of the Guelmoise population on the factorial plane1-2.

Genetic diversity

In this study, we aimed to investigate the genetic diversity of the Guelmoise local breed using genotype data at genome-wide SNP markers. The observed heterozygosity (H_o) was close to 0.30, similar to that of the three Tunisian populations, the Blonde of the Cap Bon (BLCAP), the Brune of the Atlas Grise (BRATG) and the Brown of the Atlas Fauve (BRATF) genotyped using the same SNP chip (Ben Jemaa et al., 2015). Our results are also almost similar to those of European breeds (Ho ~ 0.32). Populations of Indian origin (GIR, BRA, BRM and NEL) had the lowest rates, ranging from 0.16 to 0.2, while African populations had slightly higher rates ranging from 0.19 to 0.24 (Ben Jemaa et al., 2015). Inbreeding, measured as F_{IS}, was only slightly positive (~ 0.04), indicating that mating among relatives is generally avoided (**Table 6)**.

Table 6. Observed Heterozygosity (H_o) and inbreeding, measured as F_{IS}, for the Guelmoise population.

Population	H_o	F_{IS}
GUE	0.303	0.04

Conclusion

Our study highlighted that the Guelmoise cattle population is still reared under rather primitive management systems in marginal areas of northeastern Algeria characterized by a quite static rural society, with milk being mainly produced for subsistence and calves for sale as a source of cash income. Productive and reproductive parameters highlighted generally poor performances compared to other local cattle breeds in North Africa, likely the consequence of both a low-input management system and the lack of effective directional selection (or herd improvement through crossbreeding with European more productive breeds, which, instead, characterized most of the originally autochthonous cattle stocks in the Maghreb area). The above results, when considered together with the recognized rusticity and resistance to infectious diseases and parasites, and the presence of ancestral coat-color traits, point to an original, primitive cattle genetic stock. Description of the typology, physical and biochemical characteristics, of livestock breeds is very important for developing economic and breeding strategy in

a particular production system. The Guelmoise cattle breed shows morphological variability and conducted with an ancestral method. This variability indicates absence of directional selection towards particular objective traits. Genetic studies investigating the population structure of autochthon Algerian cattle are almost inexistent, except some preliminary studies. Our study is the first of its kind aiming to assess the genetic structure of Algerian cattle using the BovineSNP50 BeadChip. Our results are preliminary because they are limited by a single population and should be studied in depth by an exhaustive study with all the local populations existing in Algeria (Cheurfa, Setifienne, Chelifienne, Tlemcenienne and Zebu) and relationships between other breeds.

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