# <u>EJEAFChe</u>

# Electronic Journal of Environmental, Agricultural and Food Chemistry

ISSN: 1579-4377

## COMPARISON OF MILK FROM TWO DIFFERENT COW BREEDS BARROSÃ AND FRÍSIA

Preciosa Pires<sup>a</sup>, Élia Fernandes<sup>a</sup>, Manuela Vilarinho<sup>a</sup>, Mário Barros<sup>a</sup>, Rita Ferreira<sup>a</sup>, Luísa Carneiro<sup>b</sup>, Gonçalo Almeida<sup>b</sup>, Manuela Vaz Velho<sup>a</sup>

<sup>a</sup>Escola Superior de Tecnologia e Gestão, Instituto Politécnico de Viana do Castelo, Av. do Atlântico 4901-908 Viana do Castelo, Portugal <sup>b</sup>Escola Superior de Biotecnologia, Universidade Católica Portuguesa, R. Dr. Bernardino de Almeida, 4200-072 Porto, Portugal

### **KEYWORDS**

Milk; Chemical composition; Chemometrics; Pathogens

### **ABSTRACT**

Milk from two different cow breeds, Barrosã and Frísia, was studied. Within the same breed five and four milk samples were taken respectively for chemical and microbiological analysis.

The following chemical parameters were analysed in triplicate:

Total Fat, Fatty Acids Identification and Quantification, Protein, Casein, Albumin, Non-protein Nitrogen, Vitamin A, Vitamin D, Phosphorus, Potassium, Sodium, Iron, Magnesium, Calcium, Ash, Total solids, Lactic acid and Acidity.

In general terms milk from Barrosã breed showed higher values for all analysed parameters except for Total Fat, Total Solids and Vitamin D.

Statistical analysis, by testing the chemical parameters all together using the Hotelling T2 test showed that milk samples from the two cow breeds were significantly different ( $\alpha$ =0,05).

Microbiological analysis (Total Viable Counts (30°C), Escherichia coli, Staphylococcus coagulase + and Listeria monocytogenes) was also performed. Staphylococcus coagulase + was found in one milk sample from Barrosã breed. Listeria monocytogenes was found in two milk samples from Frísia breed.

### INTRODUCTION

The new concepts of natural and organic products and, in 1994, the creation of the Protected Origin Denomination (POD) and Protected Geographical Indication (PGI) for several meat products made possible the conservation of various autochthonous Portuguese breeds.

Barrosã is a cow breed originally from Trás-os Montes but also spread in Minho region. In the second half of XIX century Barrosã breed reached the 200 000 animals having the second place in terms of numbers of national cow breeds. On that time Barrosã meat was very appreciated in the United Kingdom. It was reported a number of 16708 animals exported form January to July 1882 (Carlos Torres, 2001). Barrosã numbers declined almost to extinction until the 1990s when changes on agriculture concepts, with the emphasis on quality rather than quantity and the EU protection for extensive production methods and traditional agro-animal products, together with the creation in 1994 of a POD certification for its meat, contributed for the recovery of this breed (Jordão, 2001). In 2001 there was an effective of 4955 registered reproducers (Matoso-Pereira, 2003). The Barrosã producers are about 2000 (2 to 5 cows each) and are settled in Barroso e Minho areas (Adelino Gouveia 2001). Barrosã breed production relies on extensive rearing. In certain places the animal is only confined during night and

being on free range the all day eating wild pasture and every thing they can get without a dietary supplement unless during pregnancy.

Not very long ago *Barrosã* cow milk was the only milk available in those remote areas. Nowadays, milk is available in every local shop. *Frísia* breed, and no other breed can compete with this on milk quantity, is the common milk producer in Portugal and milk industry relies on it for producing all types of dairy products.

In those rural areas where employment is difficult and desertification becomes a reality, it is important to create alternatives for youth settlement. Touristic exploitation of landscape can be linked to natural food products such as homemade cheese therefore being an added value for the already POD certificated *Barrosã* meat.

The aim of this study was to characterize chemically and microbiologically the  $Barros\tilde{a}$  milk and further comparison with Frisia milk.

### **CHEMICAL ANALYSIS**

### **Materials and Methods**

Sampling

A total a 10 milk samples were collected and analysed. The 5 milk samples from *Barrosã* were collected in local producers from Lindoso(1) and Aveleiras(4) (Arcos de Valdevez). The 5 milk samples from *Frísia* were collected in local producers from Carreço (2), Meadela (2) and Portuzelo (1) (Viana do Castelo).

All the cows were milked manually excepting the 2 samples from Carreço (semi-automatic system). Samples from  $Barros\tilde{a}$  were collected 12-24 hours before than Frisia samples and were analysed simultaneously.

### Analytical Methodologies

The analysed parameters and respective analytical protocols are shown in Table II. All samples were analysed in triplicate.

Table 1. The analysed chemical parameters and the correspondent analytical techniques

Parameter	Analytical techniques		
acidity	NP 470/1983 and AOAC 947.05 - 33.2.06		
lactic acid	NP 470/1983 and AOAC 947.05-33.2.06, conversion factor 0.009		
fat	Method AOAC 905.02, 33.2.25 or NP 468/1990		
	Extraction followed by gravimetry or Roese-Gottlieb method		
non fat solids	% total solids without fat = % total solids - % fat		
fatty acids	Hydrolysis (KOH/ethanol), extraction, acidification, gas chromatography and AOAC		
	960.30, 33.6.14		
ash	AOAC 945.46- 33.2.10 and NP 477/1983		
total solids	AOAC 925.23, NP 580/1970 and NP 475/1983, Gravimetry		
N-total	Kjeldhal method, NP 1986/1991		
N-protein	Kjeldhal method, NP 1986/1991, conversion factor = 6.38		
non-protein-N	AOAC 991.21 - 33.2.12, Kjeldhal method		
albumin	AOAC 925.24 – 33.2.19 and AOAC 927.03-33.2.18 and 991.20 -33.2.11, Kjeldhal		
aibumin	method, conversion factor = 6.38		
total casein	AOAC 927.03 – 33.2.18 and AOAC 991.20 -33.2.11, Kjeldhal method, conversion		
total casein	factor = 6.38		
phosphorus	AOAC 991.25- 33.7.08, UV/vis spectrophotometry method		
vitamins A	Handbook of Food Analysis, Chapter 17, vol.1 Ed. Leo M. L. Nollet High, RP-HPLC		
	method		
Vitamina D	AOAC 981.17- 45.1.21, RP-HPLC method		
Ca	Titrimetry (EDTA)		
K, Na, Fe, Mg	Spectrophotometry by Atomic Absorption (AA)		

### **Results and discussion**

In general terms, milk from " $Barros\tilde{a}$ " breed showed higher values in all analysed parameters, except for Total Fat and Total Solids.

Variations of milk composition, especially in the fat content are dependent on the cow breed (Kirk and Sawyer, 1991). Also the milking intervals significantly influenced the fat content e.g. 2 hours of interval fat =6% compared with 12 hours interval of milking fat=3.6%. Barrosã breed was milked in the evening after being separated from its baby for more than 12 hours (Kirk and Sawyer, 1991).

**Table II**. Chemical analysis results per 100g (values are the average of 5 samples per each type of milk and analysed in triplicate)

Chemical parameter	Barrosã	Frísia
Protein (g)	3.96 (0.50)	3.27 (0.47)
Albumin (g)	0.217 (0.068)	0.185 (0.041)
Casein (g)	3.01 (0.56)	2.48 (0.44)
Non-protein nitrogen (g)	0.029 (0.0029)	0.023 (0.0019)
Total Fat (g)	2.76 (0.43)	4.94 (1.44)
Total solids (g)	11.98 (0.76)	13.37 (1.54)
Non fat solids (NFS) (g)	9.22 (0.48)	8.43 (0.56)
Ash (g)	0.831 (0.061)	0.728 (0.012)
Potassium (K) (mg)	171 (16)	166 (23)
Sodium (Na) (mg)	88 (14)	64 (12)
Iron (Fe) (mg)	0.077 (0.0069)	0.076 (0.0080)
Magnesium (Mg) (mg)	11.6 (1.3)	9.4 (1.0)
Phosphorus (P) (mg)	110 (6.7)	87 (7.6)
Calcium (Ca) (mg)	130 (15)	99 (12)
Acidity (ml NaOH 0.1N)	22 (1.7)	16 (1.7)
Lactic acid (g)	0.20 (0.016)	0.15 (0.015)

<sup>\*</sup>Values between brackets are the Standard Deviations

The higher values for Acidity and Lactic acid of  $Barros\tilde{a}$  milk might be related with the different time collection (12-24 hours before than Frisia samples but they were analysed simultaneously). Those parameters are indicators of milk age. A sour taste is perceptible when acidity (lactic acid) reaches 0,3% and the milk is decidedly sour when it reaches the 0.4% (Kirk and Sawyer, 1991).

The two groups of milks were significantly different, considering the chemical parameters all together (Hotteling  $T^2$ ,  $\alpha$ =0.05). Considering the parameters individually and by applying a F-test, there were no significant differences between analytical parameters from both milks except for total fat, total solids and ash. With a *t*-test, differences between parameters were significant except for albumin, casein, K and Fe contents.

Table III. Total of Fatty acids (FA) and Unsaturated Fatty Acids (UFA)

Milk	FA (mg/100g of Fat)	UFA(+stearic) (mg/100g of fat)	%UFA(+stearic/) /Total FA
Barrosã	1721.59 (114.91)	344.47 (108.07)	19.59 (3.98)
Frísia	1640.98 (207.82)	193.45 (37.54)	11.83 (2.51)

The degree of unsaturation of FA was higher in *Barrosã* milk than in *Frísia* milk.

 $Barros\tilde{a}$  milk showed higher levels of Vitamin A and lower levels of Vitamin D. The average contents of Vitamins A and D in  $Barros\tilde{a}$  and Frisia milks (µg/100g of milk) were 49.07 (sd=2.49) and 1.21 (sd=0.057), and 44.42 (sd=3.59) and 2.60 (sd=0.11), respectively.

### MICROBIOLOGICAL ANALYSIS

### **Analytical procedure**

For microbiological analysis only 4 samples per each type of milk were analysed. Two samples were excluded due to improper sampling. The differences of time collection of  $Barros\tilde{a}$  and Frisia samples, was maintained – analysis for  $Barros\tilde{a}$  milk started one day before than Frisia milk.

The analysed microorganisms and the correspondent analytical protocols are shown in Table V.

Table V. Analytical protocols

Type of microorganisms	Analytical protocol		
Total Viable Counts at 30°C	NP 1995		
Total Coliforms	NFV08-050		
Staphylococcus coagulase	NFV08-057-2		
Listeria monocytogenes	ISO 11290-2 and Vidas-BioMérieux		
Salmonella spp.	Vidas-BioMérieux		
Escherichia coli	NFV 08-053		

### **Results and discussion**

Results are shown in the following table.

Table VI. Number of Microorganisms in milk samples (Barrosã: samples B1-B5; Frísia:samples T1-T5)

Samples	TVC 30°C /ml	Total Coliforms /ml	Staphylococcus coagulase /ml	L. monocytogenes	Salmonella spp. /25 g	E. coli /ml
B1	$<30x10^{2}$	<1.0	<1	<1/ml	nd	nd
B2	$1.4x10^4$	<15	$4.0x\ 10^1$	<1/ml	nd	nd
B4	$<30x10^{2}$	<1	<1	<1/ml	nd	nd
B5	$>30x10^2$	<15	<15	<1/ml	negative	nd
T2	<30	nd	$<1.0x10^{1}$	Negative/25g	negative	<1
Т3	< 30	nd	$<1.0x10^{1}$	Negative/25g	negative	<1
T4	$2.9x10^4$	nd	$<1.0x10^{1}$	Positive/25g	negative	<15
T5	$2.5x10^4$	nd	$<1.0x10^{1}$	Positive/25g	negative	~13

As not every microbiological parameters were performed for the 10 samples it only can be concluded that  $Barros\tilde{a}$  samples showed in general low TVCs; Staphylococcus coagulase + was found in one milk sample from  $Barros\tilde{a}$  milk; Listeria monocytogenes was found in 2 of the 4 Frisia milk samples. These 2 samples were the only ones collected mechanically. This might indicate cross contamination by equipment. L. monocytogenes is a food pathogen. The ingestion of L. monocytogenes in foods can pose a significant health risk, with a relatively high mortality for specific sections of the population, such as foetuses and immuno-compromised people. Several outbreaks linked to dairy products have been reported mainly related with non-pasteurised soft cheeses (Vaz-Velho, 1999).

### **ACKNOWLEDGMENTS**

To Márcio Sousa, an ESTG student, for his help on collection of Barrosã milk samples.

To Isabel Magalhães, Lurdes Videira and Arminda Moreira (ESTG staff), for their help on collection of *Frísia* milk samples.

### REFERENCES

- 1. Gouveia, A. 2001. Os criadores da Raça *Barrosã*. pp. 21-23. In: Raça *Barrosã*. Ed. AMIBA Braga Portugal.
- 2. Jordão, N. 2001. A raça *Barrosã* nos Programas comunitários. pp 17-19. In: Raça *Barrosã*. Ed. AMIBA Braga Portugal.
- 3. Kirk and Sawyer, 1991. Pearson's Composition and Analysis of Foods. Ninth Ed. Longman Scientific & Technical, UK.

- 4. Matoso-Pereira, A. 2003. Produtos tradicionais de Qualidade do Entre Douro e Minho. Ruralidades em Desenvolvimento, p. 8-9.
- 5. Torres., C. 2001. Conservação da Raça. pp. 13-15. In: Raça Barrosã. Ed. AMIBA Braga Portugal.
- 6. Vaz-Velho, M. 1999. Growth and Survival of *Listeria monocytogenes* in Foods. Published on-line-www.expofood.com and in the "Food Borne Illness" Proceedings Book, pp. 25-42. 28 & 29 May, Porto, Portugal.