

## Article

# Analysis of Environmental Factors' Impact on Donkeys' Colostrum Quality

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**Abstract:** Colostrum is a natural product, issued by both mammals and humans in first week of lactation. Among different species, donkey colostrum is considered as having, besides a valuable composition in nutrients and immune factors, an outstanding similitude with human colostrum. In this context, and taking into account the scarcity of available data concerning the interaction between climate factors and colostrum quality, a trial was conducted aiming to identify the possible influence of environmental factors on donkey colostrum nutritional traits. A stock of 16 jennies from 2 farms located in the County of Cluj, during a 7 days postpartum period was analyzed. During experimental period the daily temperature, humidity, and wind velocity data were collected. Strong positive correlations may be reported between fat and lactose, and fat and protein respectively, while moderate to strong correlation is emphasized between lactose and protein content of donkey colostrum. Testing the influence of environmental temperature, relative humidity, and wind velocity influence upon nutritional content of donkey colostrum, results the neglectable influence of the wind velocity, the negative influence of the heat stress upon all studied colostrum components, and complex influence of relative humidity, which has positive influence on fat and lactose increase when it increases, while its increase has negative influence on protein content of donkey colostrum.

**Keywords:** coefficient of variation, correlation, factor, jenny, principal components analysis

## 1. Introduction

In the last decades it can be observed a strong interest for milk coming from other animals than cows [1,2]. Particularly, donkey's milk has recently gained more interest from researchers because of it highly tolerated by infants with protein allergies [3]. Other studies [4] emphasize the particularity of donkey milk because of its rich content in lysozyme, being suitable for probiotic beverages. Cosentino et. al [5] highlighted that in the context of the increasing importance of diversity preservation even in the most vulnerable area from Natura 2000, because of the multiple uses of donkey milk (even for newborns), the donkey rearing should be encouraged.

According to Oxford Dictionary of Food and Nutrition [6], colostrum is "the milk produced by mammals during the first few days after parturition; compared with mature milk, human colostrum contains more protein (2 compared with 1.3 g/100mL), slightly less lactose (6.6 compared with 7.2 g/100mL), considerably less fat (2.6 compared with 4.1 g/100mL), and overall slightly less energy (56 kcal (235 kJ)/100 mL compared with 69 kcal (290 kJ)). Colostrum is valuable source of antibodies for

the newborn infant. Animal colostrum is sometimes known as beestings and human colostrum as foremilk”.

The benefits of both human and mammals’ colostrum result as a consequence of its composition. Beside valuable nutritional components, in colostrum composition are included growth (growth factors as epidermal, insulin like, etc., and growth hormone), immune (lactoferrin and immunoglobulins), and anti-inflammatory factors, with role in reparation of damaged cells, regulation of the immune response, or in reducing inflammatory disorders [7-9]. During the first days from birth, both human and mammal newborns must face a potential harmful environment, due to microbial threat [10]. In this context, we may mention the outstanding colostrum capacity of immunoglobulins postnatal transport, from mother to newborn [11].

The colostrum qualities is of first importance in human nutrition. For this reason, a preoccupation in the field of replacing human colostrum with colostrum from other species is reflected by literature [7-9, 12-13]. This is the appropriate context to mention the opportunity of using colostrum from donkeys as human colostrum replacer. The fundamentals of this assertion is based on the fact already known that donkey milk, as result of its composition, may be successfully used as replacer of human milk [14-21].

According to FAOSTAT [22] statistical data the evolution of donkey rearing worldwide, during the last 25 years, recorded different trends (Fig. 1-2). Africa, Asia and Americas are the continents where donkey rearing is best represented, while Europe and Oceania have fewer stocks (Fig. 1). If in the beginning of the studied period, in the year 1990, respectively, Asia possessed the most important stocks, as number of reared donkeys, in the end of the analyzed period, in 2014, respectively, Africa is placed on the first place. One may also note that except Africa, where a positive trend is emphasized, on the other continents, the donkey rearing records a decreasing tendency from 1990 to 2014 (Fig. 1).

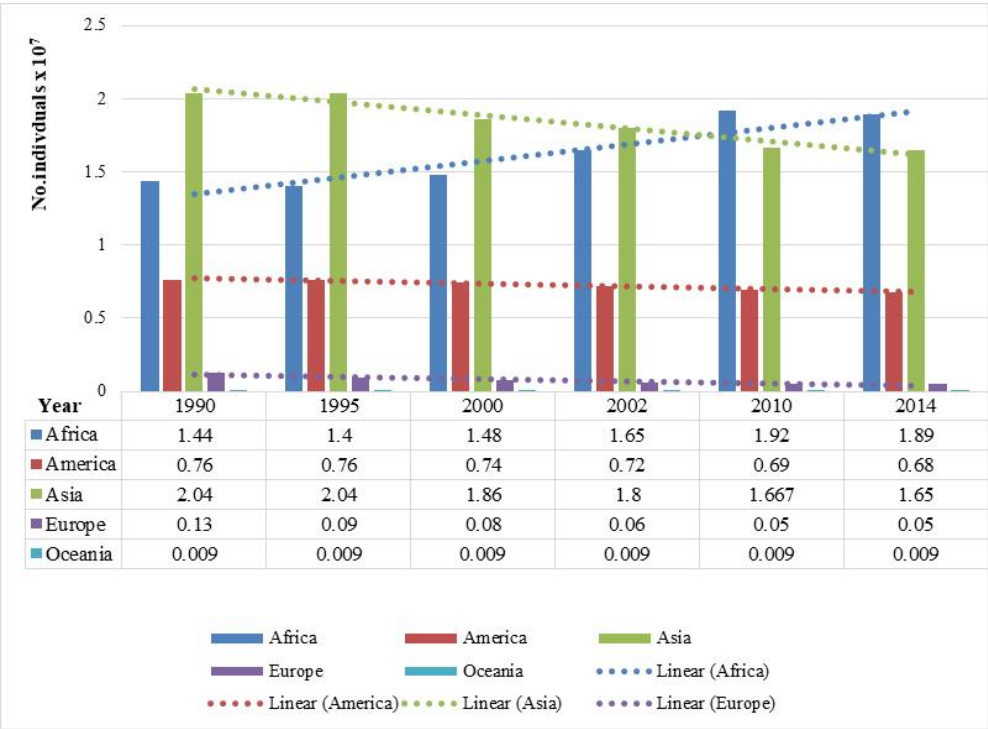
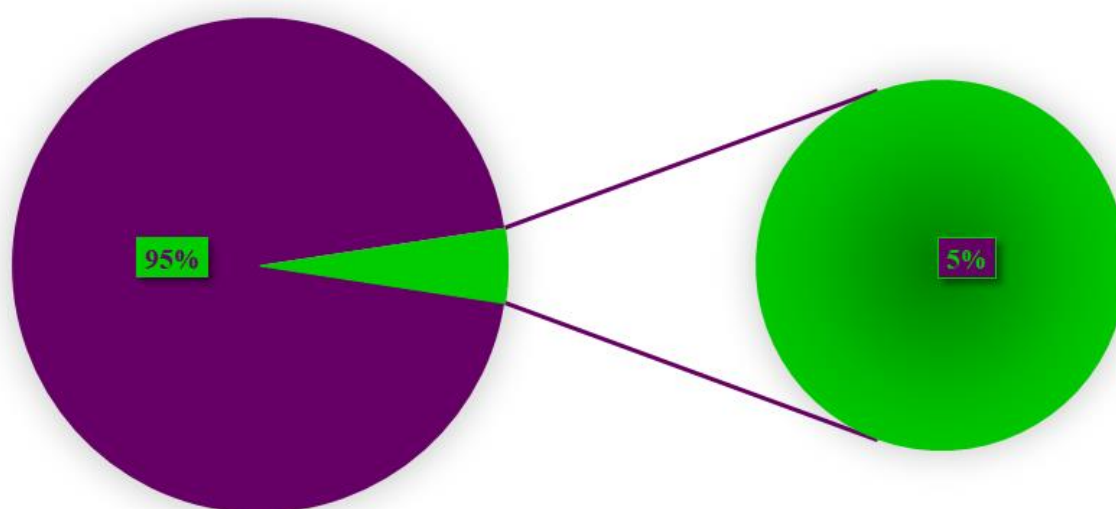


Figure 1. The evolution of donkey stocks by continents, 1990-2014

In European Union (EU), the mean number of donkey stocks by the analyzed 25 years period is of 611,785 individuals, with the minimum number of 411,135 individuals is reported in 2014, and maximum of 928,274 individuals, in 1990 (Fig. 2). This evolution has a large variability (CV = 33.77%), which emphasize the inconsistency of the representativeness of the mean for the analyzed period. Not the same thing may be noted for the evolutions of the donkey rearing in Romania, where 5% of the total EU donkey stocks are reared (Fig. 2). 31,000 donkey stocks is the mean for a 25 years period in Romania, with a minimum of 29,000 reared in 2005, and maximum of 35,000 reared in

1990. The coefficient of variability of 7.07% shows that the mean is representative, and also that the dispersion of data is small. This emphasizes the uniformity of the evolution of the donkey stocks Romania.



**Figure 2.** The basic statistics of donkey stocks evolution in EU and Romania, 1990-2014

The environmental factors, mainly heat stress, affect the behavior and wellbeing of mammals generally speaking, and donkeys, in particular [23-24], the quality of donkey milk [25-30], and also the production and quality of colostrum, generally speaking [31-32]. Studies upon pigs show that valuable components of colostrum, as immunoglobulins, are reported in bigger amounts when low temperatures are recorded [33] corresponding to farrowing that place in winter, compared to values reported in colostrum, when farrowing takes place in spring, summer, or even fall [34]. Even though, in last years, research was conducted in Romania, upon emphasizing donkey milk quality and composition [35-37], no data concerning environmental factors (as noise, temperature, humidity, wind, etc.) influence on colostrum quality and composition are available. The aim of this trial is to quantify, and assess the significance of the influence of temperature, relative humidity, and wind velocity, on fat, protein, and lactose content from donkey colostrum, within regional specific environmental conditions.

## 2. Materials and Methods

### 2.1. The experimental areal

The experiment was developed in two donkey private farms, located nearby an urban area. This urban area is represented by Huedin town. It is located in County of Cluj, being characterized by the coordinates: 46°52'00"North, and 23°02'00"East. The trial was carried out during the jennies 7 days postpartum period, when they fed the offspring with colostrum, from 2<sup>nd</sup> to 8<sup>th</sup> May, 2016.

### 2.2. Sample collection

The colostrum samples were collected once a day, in the morning, from 16 multiparous jennies, during experimental period. The females received the same feed, and were maintained in stabulation, during first 7 days of milking, when colostrum is released. The colostrum samples in

amount of 10 mL, daily collected, were delivered to the laboratory, and maintained at temperature of 4°C. The environmental temperature, relative humidity, and wind velocity were continuously monitored during all 7 days experimental period, using WE900 Weather Station (4-20mA) monitoring station, produced by Global Water, USA. The most important component of the climate traits in experimental area is temperature, because an increase of 0.9°C is reported in last 10 years (Romanian National Administration of Meteorology. Climatic monitoring).

### 2.3. Data analysis

Colostrum qualitative and quantitative traits were analyzed with Lactoscan MCC device, produced by Milkotronic Ltd. Fat, protein, and lactose determinations were performed by the direct measurement, and the basic principle of the methodology is the real time measurement of the speed of the ultrasound in colostrum.

IBM SPSS Statistics v. 20 as used for colostrum and climatic raw data processing. In order to emphasize both colostrum and climatic traits, Descriptive statistics was used. The linearity of the simple correlations between the colostrum traits (fat, protein, and lactose) was tested. The Pearson simple correlations are calculated. Furthermore the factorial analyze, through the Principal Components Analysis (PCA) technique, was used in order to emphasize the main environmental factors (temperature, °C; humidity, %; wind velocity, m/s) affecting the colostrum composition in fat, protein, and lactose.

### 3. Results and discussions

During 7 days experimental period, the mean temperature reaches 12°C, while for mean relative humidity and mean wind velocity, values of 65.43%, and 8.57%, respectively, were reported (Table 1). According to the values of the coefficients of variations, the means of temperature (CV = 9.62%), and relative humidity (CV = 11.09%) were representative for the study, while the mean wind velocity (CV = 21.15%) has satisfactory representativeness. The mean fat content identified in donkey colostrum, during the research period, was 3.77%, while, the protein was 2.36%, and lactose, 2.35%.

The fat content recorded a high variation during the colostrum secretion period, from 1.89% up to 4.88% (CV = 20.21%), smaller variations are identified in protein content (1.47% – 2.75%), and in lactose content (1.43 – 2.71%), both characterized by coefficients of variability of CV = 11.95%, and CV = 11.86%, respectively (Table 2).

**Table 1.** Descriptive statistics concerning the evolution of the climatic parameters within the experimental areal, 2-8 May, 2016

Parameter	N	Temperature (°C)	Humidity (%)	Wind velocity (m/s)
Mean	7	12.00	65.43	8.57
Standard deviation	7	1.15	7.25	1.81
Standard error of mean	7	0.44	2.74	0.69
Minimum	7	10.00	56.00	6.00
Maximum	7	13.00	76.00	11.00
Coefficient of variation	7	9.62	11.09	21.15

**Table 2.** Descriptive statistics for the evolution of donkey colostrum traits, 2-8 May, 2016

Parameter	N	Fat (g/100mL)	Protein (g/100mL)	Lactose (g/100mL)
Mean	16	3.77	2.36	2.35
Standard deviation	16	0.76	0.28	0.26
Standard error of mean	16	0.19	0.07	0.06
Minimum	16	1.89	1.47	1.43

Maximum	16	4.88	2.75	2.71
Coefficient of variation	16	20.21	11.95	11.86

If compared the results of our research (Table 2), with values identified in other mammals and humans (Table 3), it results that donkey colostrum content in nutrients has most quantitative similitudes with human and cow colostrum. Thus, the mean fat content (3.77 g/mL) identified in donkey colostrum is quantitatively close to fat content in cow colostrum reported by Meyer and Kamphues [38], and human colostrum, reported by other researches [39, 40]. Compared to sow colostrum fat content of 5.8% reported by Park [41], ewe and she-goat, reported by Meyer and Kamphues [38]. The donkey colostrum mean protein content of 2.36 g/100 mL resulted from our research (Table 2), frames within the interval of values reported in human colostrum protein [39, 40], while compared to protein content from cow, sow, ewe and she-goat colostrum (Table 3), it has smaller value [38; 41].

**Table 3.** Composition of colostrum in different livestock species and humans

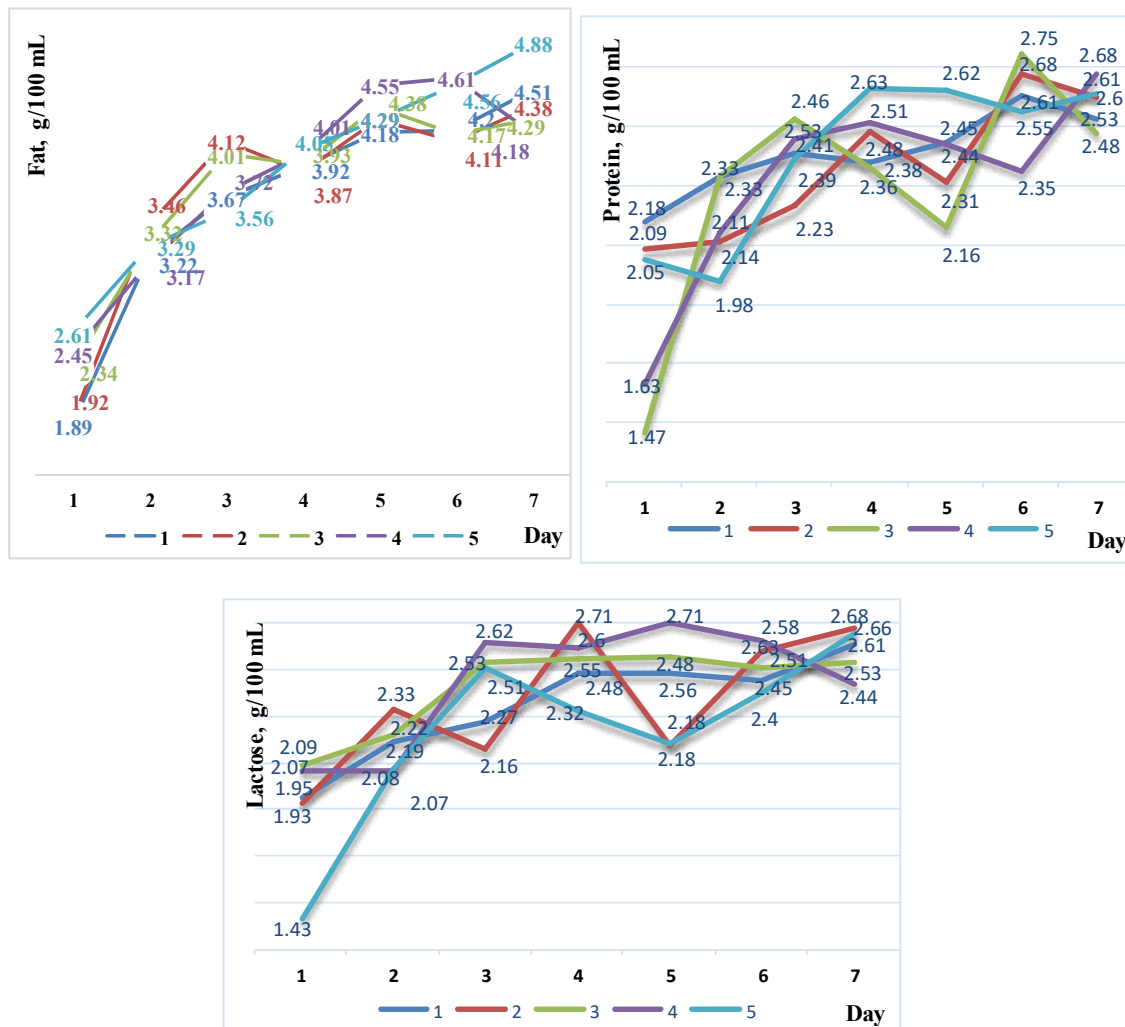
Issue	Fat	Protein	Lactose	Unit	References
Human	2.9-2.95	2.29-3.7	5.3-5.7	g/100mL	[39,40]
Cow	3.6	13	3.1	%	[38]
Pig	5.8	10.6	3.4	%	[41]
Sheep	12.4	13	3.4	%	[38]
Goat	9	8	2.5	%	[38]

According to our research, the mean lactose content in donkey colostrum (2.36 g/mL) is quantitatively close to lactose content from sheep-goat colostrum (Table 2, Table 3) reported by Meyer and Kamphues [38], but lower compared to lactose content from sow, cow, and ewe [38; 41], and much lower (Table 3) compared to lactose content in human colostrum [39-40].

Within our research, the evolution of colostrum nutrients, fat, protein, and lactose, respectively, from the first up to the 7<sup>th</sup> day of colostrum delivery, records an increasing trend, even individual fluctuations may be reported (Fig. 3). Our results are consistent with those reported by Kehoe et al. [40], and Tsioulpas [42], concerning the evolution of the nutrients quantity in cow colostrum, function of delivery day.

Between donkey colostrum mean content in protein and lactose, a positive moderate to strong correlation is identified (Fig. 4). It is statistically very significant ( $p < 0.001$ ), and emphasizes a linear dependence between analyzed traits, being representative for less than half of the sample. It means that when protein from jenny colostrum increases, the lactose also increase in 36.70% of the colostrum collected from the studied jennies population (Fig. 3). Concerning donkey colostrum mean content in fat and lactose, our research reveals a positive strong correlation (Fig. 4). This linear correlation is positive and statistically very significant ( $p < 0.001$ ).

It is representative for more than a half of the studied population, meaning that when fat from colostrum increases, the lactose also increase in 56.70% of the colostrum delivered by the studied jennies population (Fig. 4). The study also emphasizes a strong positive correlation between fat and protein content of donkey colostrum (Fig. 5). This correlation is statistically very significant ( $p < 0.001$ ), and representative for more than half of the sample. In this case, it means that when fat from jennie colostrum increases, an increase of the protein content will be found in colostrum from 52.80% of the studied jennies population (Fig. 5).

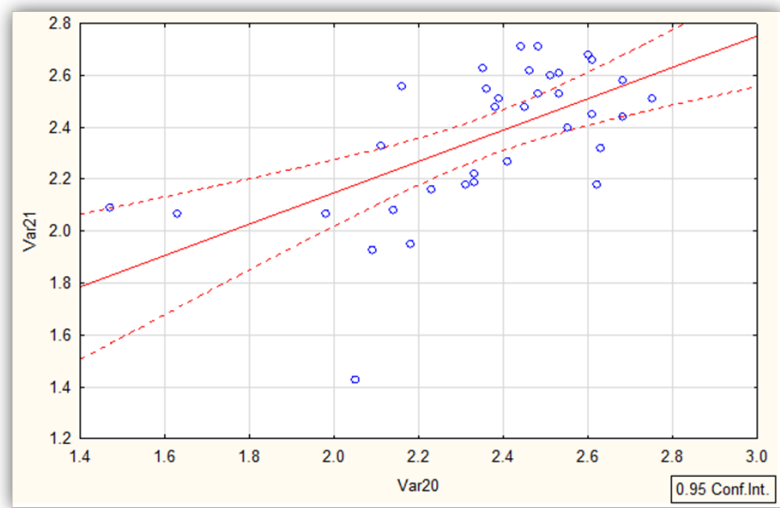


**Figure 3.** The daily evolution of the donkey colostrum nutrients (fat, protein, lactose), 2 – 8 May, 2016

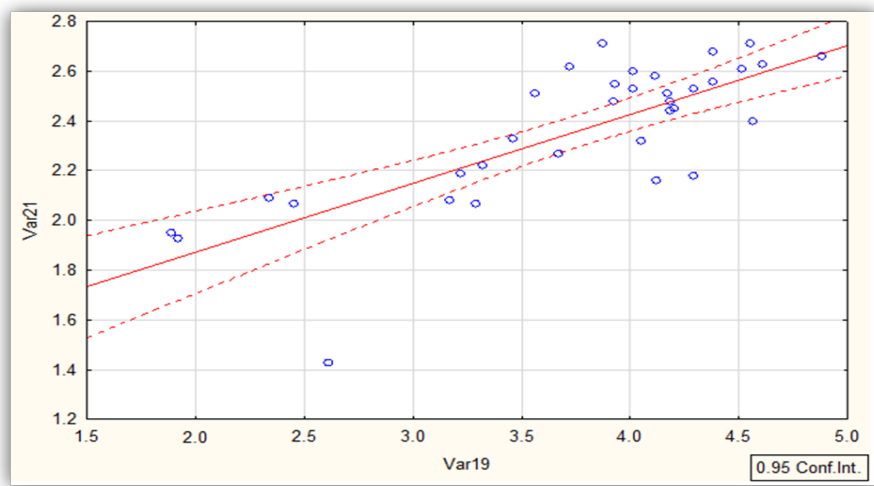
Studies upon the interaction between mammals milk nutrients (fat, protein, lactose, etc.), and climatic factors are few, but concerning the presence of nutrients in mammalian colostrum are fewer.

Thus, if literature mentions the effects of heat stress on nutritional composition of milk from dairy species [27,28], not the same thing may be mentioned when we refer to colostrum, while concerning donkey colostrum, we may note the scarcity of data.

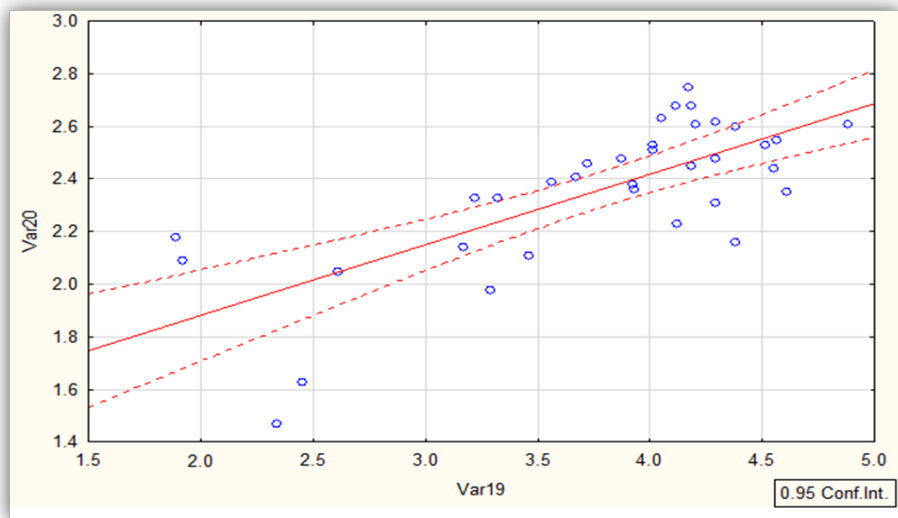
The nutritional characteristics of the donkey colostrum analyzed in this study are moderate to weak correlated with climatic factors (Table 4). The temperature is negatively correlated ( $p < 0.05$ ) with donkey colostrum nutrients, moderate with fat, and lactose, and moderate to strong with protein, while humidity is positively moderate correlated ( $p < 0.05$ ) with fat, and lactose. Between wind velocity and donkey colostrum nutrients, negative weak and very weak correlations ( $p > 0.05$ ) are reported (Table 4).



**Figure 4.** The simple correlation between protein and lactose content from donkey colostrum



**Figure 5.** The simple correlation between fat and lactose content from donkey colostrum, 2 - 8 May, 2016



**Figure 6.** The simple correlation between fat and protein content from donkey colostrum, 2 - 8 May, 2016

Because between colostrum nutritional traits and climatic factors are identified (Table 4), we consider opportune to perform the PCA, in order to identify the principal climatic factors affecting the donkey colostrum nutritional traits studied in this trial. Our study emphasizes two principal components (factors), temperature and relative humidity, respectively, and three variables (Fig. 4).

The first factor, temperature, explains 79.75% of variance, correspondent to 2.392 variables, while the second factor, relative humidity, explains 13.16% of variance, correspondent to 0.394 of variables. Cumulated, both factors explain 92.91% of the commune variance, emphasizing one again the importance of temperature and relative humidity (Table 5).

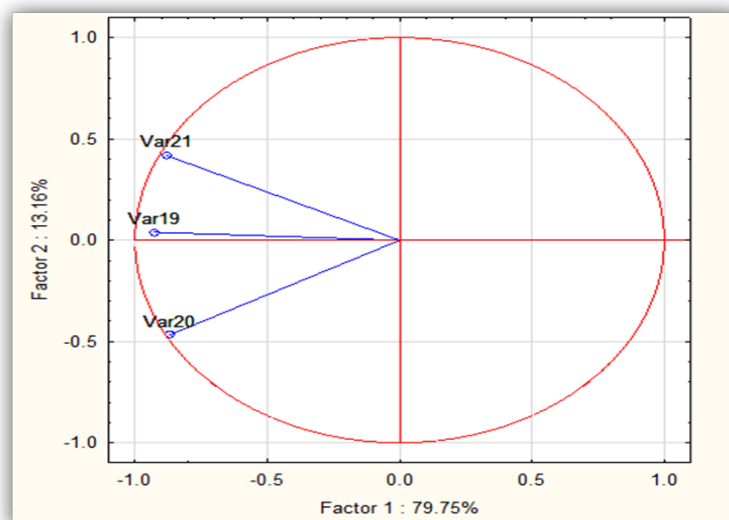
**Table 4.** The correlation matrix of the donkey colostrum traits and climatic traits considered within this experiment, 2 – 8 May, 2016

Issue	Temperature (°C)	Relative humidity (%)	Wind velocity (m/s)
Fat (g/100mL)	- 0.527*	+0.411*	-0.090 <sup>ns</sup>
Protein (g/100mL)	- 0.684*	-0.544*	-0.225 <sup>ns</sup>
Lactose (g/100mL)	- 0.487*	+0.441*	-0.146 <sup>ns</sup>

\* -  $p < 0.05$ ; ns -  $p > 0.05$ .

**Table 5.** The total variance explained

Variable	Eigenvalue	% Total variance	Cumulative - Eigenvalue	Cumulative, %
Fat (mg/100 mL)	2.392	79.751	2.392	79.751
Protein (mg/100 mL)	0.394	13.162	2.787	92.913
Lactose (mg/100 mL)	0.212	7.086	3.000	100.000



Var 19 – fat; Var 20 – protein; Var – 21 – lactose

**Figure 7.** The projection of the variables on the PC1xPC2 plane

Within our experiment, according to variable projections, and correlations between them (Fig. 7, Table 6), protein content of jennies' colostrum may be decreased by increase of both temperature and relative humidity, while the content of other two considered colostrum nutrients, fat, and lactose, may be positively influenced by the decrease of the temperature, and increase of the relative humidity.

**Table 6.** The correlations of PC1 x PC2 variables (factor loadings)

Variables	Principal component (Factor) 1 – temperature (°C)	Principal component (Factor) 2 – relative humidity (%)
Fat (mg/100 mL)	-0.887	-0.437
Protein (mg/100 mL)	-0.937	-0.219
Lactose (mg/100 mL)	-0.854	-0.418
Explained variance	4.185	1.291

According to PCA, all analyzed nutrients, meaning fat, protein, and lactose, are strongly influenced by temperature, while relative humidity affects in a moderate manner fat and lactose, and at low extent, protein (Table 6, Fig. 7).

#### 4. Conclusions

The results of our trial show that donkey colostrum has, at large extent, similar content with human colostrum, in terms of nutrients (fat, protein, and lactose). In the meantime, our study emphasize a relative constant content of fat and lactose in donkey colostrum, while protein exhibits high variations from the first to the last day of production, with linear increasing tendency. Positive strong, and strong to moderate correlations were identified between donkey colostrum nutrients, emphasizing their interrelationship. Our study brings an original contribution to the field of research concerning the possible influence of the climatic factors upon the donkey colostrum content in nutrients. Thus, the PCA emphasizes temperature and relative humidity, at lower extent, compared to temperature, as main factors affecting donkey colostrum content in fat, lactose, and protein. We identified temperature as the main climatic factor influencing the fat, protein, and lactose content in donkey colostrum, meaning that heat stress constitutes a negative environmental input on colostrum nutritional quality. According to our findings, the relative humidity have moderate influence upon nutritional quality of colostrum. The increase of relative humidity has positive influence upon the evolutions of fat and lactose donkey colostrum content, while it has negative influence upon protein content. Taking into account the benefits of donkey milk colostrum, in nutrients, growth, immune, or anti-inflammatory factors, within the national and EU context, characterized by the decrease of donkey stocks, further studies are needed in order to obtain detailed information concerning this natural source of health, and exogenous factors affecting its quality.

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