# Quantitative Genetics Researches on Milk Protein Systems of Romanian Grey Steppe Breed

V. Maciuc, Șt. Creangă, I. Gîlcă, V. Ujică

**Abstract**—The paper makes part from a complex research project on Romanian Grey Steppe, a unique breed in terms of biological and cultural-historical importance, on the verge of extinction and which has been included in a preservation programme of genetic resources from Romania. The study of genetic polymorphism of protean fractions, especially kappa-casein, and the genotype relations of these lactoproteins with some quantitative and qualitative features of milk yield represents a current theme and a novelty for this breed. In the estimation of the genetic parameters we used R.E.M.L. (Restricted Maximum Likelihood) method.

The main lactoprotein from milk, kappa - casein (K-cz), characterized in the specialized literature as a feature having a high degree of hereditary transmission, behaves as such in the nucleus under study, a value also confirmed by the heritability coefficient ( $h^2 = 0.57$ %). We must mention the medium values for milk and fat quantity ( $h^2=0.26$ , 0.29%) and the fat and protein percentage from milk having a high hereditary influence  $h^2 = 0.71 - 0.63$ %.

Correlations between kappa-casein and the milk quantity are negative and strong. Between kappa-casein and other qualitative features of milk (fat content 0.58-0.67 % and protein content 0.77-0.87%), there are positive and very strong correlations. At the same time, between kappa-casein and  $\beta$  casein ( $\beta$ -cz),  $\beta$  lactoglobulin ( $\beta$ -lg) respectively, correlations are positive having high values (0.37 – 0.45 %), indicating the same causes and determining factors for the two groups of features.

*Keywords*—breed, genetic preservation, lactoproteins, Romanian Grey Steppe

### I. INTRODUCTION

THE conclusions of different bibliographical sources in this field highlight some contradictions in terms of the signification of relations between the genetic variants of lactoproteins and the features of milk yield, due to diverse factors influencing the milk quantity and its composition and underline that it is difficult to establish the singular influence of protein polymorphism [1, 2, 3].

In the first part of our researches, we established the

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V. Ujică is with the University of Agricultural Sciences and Veterinary Medicine, Iasi-700489, Romania, Faculty of Animal Sciences (E-mail: vasileujica@yahoo.com). average values and variability of milk yield indices and lactoproteins under study. On this occasion, we made a full analysis of locus of kappa-casein (K-cz), establishing the gene frequency, genotype frequency, the standard error of the gene frequency and the state of genetic balance according to Hardy-Weinberg law. In the second part, we present the results of quantitative genetics researches (heritability, phenotypic, genetic and environment correlations) and we analyze the significance of relations between the studied lactoproteins with the milk yield by normal lactation (305 days), the milk content in terms of fat protein and casein,.

#### II. MATERIAL AND METHOD

Researches were effectuated on 30 Romanian Grey Steppe cows, genetically preserved, raised semi-intensively, tied-up stalling, at the Research-Development Station for Bovine Growing Dancu, Iași (S.C.D.C.B. Dancu, Iași). In the second part of our researches, we focused on the quantitative genetics elements for some features and lactoprotein systems of milk. The data were taken from observations and direct determinations on the farm and from the primary data bank of the farm and O.A.R.Z. Iaşi (Office for Improvement and Reproduction in Animal Science, Iasi). The study of polymorphism of milk proteins was made by PCR-RFLP technique, and for the study of polymorphism of all bovine lactoproteins we also used the isoelectric focalization technique (I.E.F.) [3]. To estimate the genetic parameters, we used R.E.M.L. method (Restricted Maximum Likelihood). This relies on an iterative process of maximization of a function. The calculation techniques vary depending on the optimization algorithm chosen, but all need BLUP solutions for each iteration cycle for different effects of model. It is necessary to have a large number of iterations to arrive to convergence, but this can be avoided if we want to have an effective evaluation. It is usually accepted as a convergence criterion a difference between the solutions resulted from the last and the before last iteration less than a percentage from the average of the values registered for the respective indicator [4, 5]. In our case, the final convergence was 99.99 % and the number of iterations was 1687.

The biometric model used has the following form [5, 6, 7]:  $J_{iikl}=\mu+F_i+L_J+A_k+e_{iikl}$  where:

J<sub>ijkl</sub>=performance "l" of individual "k" achieved in lactation "j", on farm "i",

 $\mu$  =general average,

 $F_I$  = effect of farm "i" (fixed factor); i=1.

L<sub>j</sub>= effect of lactation rank "j" (fixed factor); j=1.

A<sub>ik</sub>=additional genetic effect of individual "k"; (random factor).

e<sub>ijkl</sub>=error associated to every measured performance.

It is a mixed model since it includes a random factor (the animal) and two fixed factors (farm and lactation rank).

#### III. RESULTS AND DISCUSSIONS

The milk quantity per normal lactation for Romanian Grey Steppe population under study has an ascending evolution from 1589.64 Kg in the 1<sup>st</sup> lactation to 2535.43 Kg in the 5<sup>th</sup> lactation also representing the maximum lactation. The first lactation represents 62.69 % from the maximum lactation, a value highlighting the tardiness of Romanian Grey Steppe breed in terms of milk yield. Staring from the 6<sup>th</sup> lactation, the milk quantity decreases and in the 8<sup>th</sup> lactation the quantity is 1078.5 kg. We must mention that in the nucleus under study there were individuals with a maximum yield of 4019 kg of milk by normal lactation or 4080 kg of milk. The fat percentage reaches its maximum value in the 5<sup>th</sup> lactation, namely 4.73% and the protein percentage has the same evolution reaching 3.71% in the 5<sup>th</sup> lactation [8, 9, 10].

In the genetic structure of the livestock under stock, we identified three groups of paternal semi-sisters with a genetic value between 1548.22 kg (code 79009) and 1752.33 kg (code 79005), proving the weak genetic value of the breeding bulls in terms of milk yield. However, we may notice a good body development of the genetic groups with values of body weight between 549.38 kg (87027) and 626.67 kg (code 79005). These data are favorable for the selection of the nucleus under study in order to improve the meat yield of Romanian Grey Steppe breed.

Studies of quantitative genetics on the characteristics of milk yield of members of the Bovidae family show that both the protein content and the type of casein or seric proteins have a strong genetic determinism (0.63 - 0.57 %) and their variants may positively or negatively influence the total protein content of milk.

After having initially established the phenotypic values and total variance of the main indicators studied for Romanian Grey Steppe breed, we started to determine the genetic variance and heritability coefficients [11] and results are presented in table I.

7	CABLE I

HERITABILITY COEFFICIENT FOR THE MAIN FEATURES AND MILK PROTEAN SYSTEMS

		Variance due to	-	
~	. 2	additive	"Intralot"	Total
Character	h <sup>2</sup>	genes	variance	variance
Normal lactation duration day	0.22	1799.0387	4628.2904	6427.3292
Milk quantity Kg	0.26	46247.036	340772.72	387019.76
Fat percentage	0.71	0.9212	0.6745	1.5957
Fat quantity Kg.	0.29	166.5806	614.5592	781.1397
Protein percentage	0.63	0.9084	0.7005	1.7089
Protein quantity kg	0.27	203.1983	479.9655	683.1637
K_cz %	0.57	0.204	0.3	0.504
β_cz %	0.21	0.048	0.8	0.848
β_lg %	0.19	0.132	0.3	0.168
αS1-cz %	0.29	0.696	1.8	1.104

Analyzing the value of the heritability coefficient for kappa-casein (K\_cz), this has a stronger level of hereditary

determination ( $h^2 = 0.57$  %) and a guarantee that the phenotypic values established largely correspond the potentiality of component genotypes. The environment, in case of this feature, participates with a lower share in determining the total variance, a fact triggering different orientations, methods and managerial systems in the genetic improvement process as against the lactoproteins with a poor hereditary transmission.

The high "intralot" variance of  $\beta$ -casein ( $\beta$ \_cz) and  $\beta$  lactoglobulin ( $\beta$ \_lg) and the lower one due to the additive genes determined smaller values of genetic variance and, implicitly, of heritability coefficients for these lactoproteins. The low genetic determination rate of these lactoproteins reflects the high variability of the female material from where we took off the milk samples and the low genetic variance of the male breeding stock. An intermediate situation is that of  $\alpha S_1$  casein ( $\alpha S1$ - cz) having a medium hereditary transmission coefficient ( $h^2 = 0.29$  %).

By examining the hereditary transmission indices of the main milk protean systems for Romanian Grey Steppe breed, we confirm the observation made on the basis of phenotypic data according to which this population is made of more homogenous or heterogeneous genotypes with a higher or less genetic variability, with features more or less hereditarily fixed.

The main milk lactoprotein, kappa-casein (K\_cz), characterized in the specialized literature as a feature having a high level of hereditary transmission, for Romanian Grey Steppe nucleus from Dancu farm, Iaşi, is as such, a value also confirmed by the heritability coefficient ( $h^2$ =0.57 %).

On the basis of the heritability coefficient values of the main features and milk lactoproteins of Romanian Grey Steppe cows, we may affirm the favorable effect of selection in accordance with the improvement and genetic preservation objectives of this nucleus having a patrimony genetic value for Romania.

Among the objectives for the improvement of Romanian Grey Steppe breed, there is the increase of milk yield and the improvement of the relations between the main quantitative features of milk.

The relatively low heritability of milk yield makes rather ineffective the selection of Sura de steps cows based on their own phenotype or the yield of their descendants; taking into consideration the test according to progeny extends too much the time to obtain the selection effect especially in the fist stage of genetic improvement by genotype. That is why, in the genetic improvement process of Romanian Grey Steppe cattle, an immediate interest, beside the one of establishing the heritability coefficient, consists in establishing the values of correlation coefficients between characters and the features subject to improvement. This triggers the possibility to apply some methods by which the effect of selection may be obtained faster and the simultaneous improvement of more characters that are correlated among them.

For this purpose, we determined the phenotypic, genetic and environment correlations existing between the main milk protean systems and the milk yield indicators in the 1<sup>st</sup>

lactation and among the main milk protean systems as well and results are presented in tables II and III.

In the first stage, we studied the phenotypic, genetic and environment correlations existing between the main milk protean systems and the milk yield indicators (table II) and then the same correlations among the main milk protean systems (table III).

TABLE II PHENOTYPIC, GENETIC AND ENVIRONMENT CORRELATIONS BETWEEN THE MAIN LACTOPROTEIN SYSTEMS AND THE MILK

Charact	er couples	Phenotypic correlation	Genetic correlation	Environmen t correlation
K-cz	Milk Kg	-0.55	-0.46	-0.56
K-cz	Fat %	0.58	0.67	0.64
K-cz	Fat Kg	-0.59	-0.58	-0.61
K-cz	Protein %	0.77	0.83	0.87
K-cz	Protein Kg	-0.68	-0.69	-0.71
β-cz	Milk Kg	0.17	0.25	0.16
β-cz	Fat %	0.64	0.56	0.65
β-cz	Fat Kg	0.24	0.23	0.25
β-cz	Protein %	0.29	0.24	0.28
β-cz	Protein Kg	0.18	0.19	0.17
β-lg	Milk Kg	-0.23	0.17	-0.25
β-lg	Fat %	-0.24	-0.21	-0.13
β-lg	Fat Kg	-0.22	0.18	-0.23
β-lg	Protein %	-0.24	-0.19	-0.26
β-lg	Protein Kg	-0.25	0.20	-0.26
αS1- cz	Milk Kg	0.49	0.27	0,53
αS1- cz	Fat %	0.43	0.30	0.44
αS1- cz	Fat Kg	0.51	0.28	0.54
αS1- cz	Protein %	-0.19	-0.27	-0.18
αS1- cz	Protein Kg	0.21	0.25	0.27

To establish the relations between the main protean systems, we researched by means of the phenotypic, genetic and environment correlation coefficients the manner in which we should calculate the main milk protean systems and results are given in table 3.

TABLE III PHENOTYPIC, GENETIC AND ENVIRONMENT CORRELATIONS

BETWEEN THE MAIN MILK PROTEIN SYSTEMS				
Indicator	couples	Phenotypic correlation	Genetic correlation	Environment correlation
αS1- cz	K-cz	-0.21	0.27	-0.25
αS1- cz	β-cz	0.56	0.48	0.65
αS1- cz	β-lg	-0.26	0.25	-0.28
K-cz	β-cz	0.37	0.43	0.39
K-cz	β-lg	0.39	0.43	0.45

Analyzing the values of the phenotypic and environment correlation coefficients in system  $\alpha S_1$  casein ( $\alpha S_1$ -cz) and other three milk protean systems( K-cz,  $\beta$ -cz,  $\beta$ -lg), it results that in case of kappa-casein (K-cz) and  $\beta$  lactoglobulin ( $\beta$ -lg) correlations are negative and of medium intensity ther existing a significant antagonism between the two features; with  $\beta$ casein ( $\beta$ -cz) correlations are positive and with high values (0.48 - 0.65%), indicating the same causes and determining factors of the two groups of features. As for the genetic correlations, all variants analyzed comply with the limits of positive and significant correlations.

TABLE IV
CORRELATION BETWEEN $\alpha S_1$ -cz și $\beta$ -cz

		$\alpha S_1$ -cz	β-cz
С	orrelations		
αS1-cz	Pearson Correlation	1.000	.585
	Sig. (2-tailed)		.076
	N	30	30
β-cz	Pearson Correlation	.585	1.000
	Sig. (2-tailed)	.076	•
	Ν	30	30

COI	TABLE V RRELATION BETWEEN	αS1-cz ŞI K-	cz
С	correlations	$\alpha S_1$ -cz	K-cz
$\alpha S_1$ -cz	Pearson Correlation	1.000	237
	Sig. (2-tailed)	30	.511 30
	N		1.000
K-cz	Pearson Correlation Sig. (2-tailed)	237	
	N	30	30

TABLE VI CORRELATION BETWEEN K-cz ŞI β-cz				
	-	K-cz	β-cz	
Ĺ	Correlations			
K-cz	Pearson Correlation	1.000	.278	
	Sig. (2-tailed)	•	.565	
	Ν	30	30	
β-cz	Pearson Correlation	.278	1.000	
	Sig. (2-tailed)	.565	•	
	Ν	30	30	

CO	TABLE VII DRRELATION BETWEEN	NK-cz ŞIβ-l	g
(	Correlations	K-cz	β-lg
L L	orrelations	1.000	
K-cz	Pearson Correlation	1.000	.347
	Sig. (2-tailed)	•	.397
	Ν	30	30
β-lg	Pearson Correlation	.347	1.000
	Sig. (2-tailed)	.397	
	Ν	30	30

To confirm the values obtained for the relations between the main milk protean systems by R.E.M.L. method, given in tables IV, V, VI and VII, we present several Pearson correlations obtained by SPSS 16.

We may say that in case of the protean systems under study, selection of the milk protean systems may be made concomitantly since there are stronger and positive genetic relations between these features also favorable for the improvement of milk quality for Romanian Grey Steppe breed.

## IV. CONCLUSION

- In the fist lactation 62.69 % from the maximum lactation was achieved, a value highlighting the tardiness of Romanian Grey Steppe breed in terms of milk yield. The milk quantity ranged between 1589.64 kg (1<sup>st</sup> lactation) and 2535.43 kg in the 5<sup>th</sup> lactation which also was the maximum lactation.
- 2. The main lactoprotein from milk, kappa casein (Kcz), characterized in the specialized literature as a feature having a high degree of hereditary transmission, behaves as such in the nucleus under study ( $h^2 = 0.57$ %) and the protein percentage has a higher hereditary influence ( $h^2 = 0.63$  %)
- 3. In case of the milk protean systems under study, selection of the milk protean systems may be made concomitantly since there are stronger and positive genetic relations between these features also favorable for the improvement of milk quality for Romanian Grey Steppe breed.

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