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Jubilee of Professor Juraj Pivko

On 20 January 2014 outstanding Slovak scientist and teacher DVM Juraj Pivko, MD., member of the editorial board of the Slovak Journal of Animal Science celebrated his 70th anniversary.

Prof. DVM Juraj Pivko, MD was born 20. 1. 1944 in Pezinok. He graduated in the field of General Veterinary Medicine at the University of Veterinary Medicine in Košice in 1968. In the same year he joined the Research Institute of Animal Production in Nitra, where he graduated in 1978 as a Postgraduate in the field of veterinary morphology and physiology.

His research continued in the reproduction of farm animals and in 1988 he defended his Doctoral dissertation. In 1992 he was qualified as Associate Professor in the reproduction of farm animals and in 1996 was appointed as a Professor of General Animal Breeding.

His research activities from the beginning were devoted to the reproduction of farm animals.

His dissertation in this topic was focused on the reproductive process of infantile gilts. The results of his work formed the basis for the successful solution of the research projects in the field of reproduction of farm animals, which resulted in the preparation of a doctoral dissertation „The processes of oocyte maturation and embryo transfer in cattle, pigs and sheep“.



Professor Pivko significantly contributed to the establishment of biotechnical methods, such as embryo transfer and artificial insemination in farm animals, in the former Czechoslovakia and then in Slovakia. Another important area of his activity was the development of electron microscopy in the field of animal science.

Prof. Pivko underwent research stays and lecturer tours in leading European research centres in Germany, France and Russia. He has been the principal investigator in many international research projects and grants, organizer and coordinator of eminent international conferences. A portfolio of his scientific publications has very good

international response and contains more than 400 bibliographic units (among them 21 scientific monographs and over 200 original scientific papers).

A significant part of the activities of professor Pivko is represented by his teaching activities on the Faculty of Biotechnology and Food Sciences of Slovak University of Agriculture and Faculty of Natural Sciences of Constantine the Philosopher University in Nitra, where he taught mainly subjects in the field of biotechnology and animal reproduction. He was the supervisor of a significant number of undergraduate theses.

Since 1988 he was Director of the Institute of Genetics and Reproduction of Farm Animals at the Research Institute for

Animal Production, where he created his own scientific school being a supervisor of 12 PhD students; 3 scientists in his research team were awarded the degree Doctor of Sciences.

Prof. Pivko is a member of several scientific boards, academic, expert committees and editorial boards; he is the holder of significant domestic and international awards.

At the occasion of his jubilee we wish professor Pivko in the following years a lot of optimism, health, creative scientific invention and enjoy in work activity and family life.

Prof. Ing. Ladislav Hetényi, PhD.
Editor-in-Chief

CHARACTERIZATION OF GENETIC RESOURCE IN CHICKEN OF ORAVKA BREED

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ABSTRACT

Growth and some reproduction traits (fertilization, hatchability and egg weight) of Oravka breed were measured in two chicken lines (OR2 and OR3). The in situ conservation flock kept in Animal Production Research Centre Nitra (APRC Nitra) during the period between 2009 and 2012 was included in the experiment. During each season the breeding males not from the APRC Nitra flock were used. The breeding females originated from the mating between females raised in the APRC Nitra and those new males. The weight at age of 5, 12 and 20 weeks was monitored. The average weights of OR2 were ranged from 371.3 ± 68.9 g to 538.1 ± 79.4 g at 5 weeks, from 1246.1 ± 254.0 g to 1464.5 ± 242.2 g at 12 weeks and from 2076.3 ± 381.8 g to 2286.1 ± 535.4 g at 20 weeks of age. The fertilization rate for each line and season was higher than 84 % except for OR2 line in the season 2011/2012. The hatchability from fertilized eggs was higher than 80 % except for OR2 line in the season 2011/2012. The average weights of eggs in the middle of laying period (from March to May) were ranged from 53.9 ± 3.5 g to 56.6 ± 4.3 g for OR2 and from 52.9 ± 4.5 g to 56.4 ± 3.6 g for OR3 during the whole experiment.

Key words: Oravka; growth; fertilization; hatchability; egg weight; genetic resources

INTRODUCTION

The intensive selection of laying and meat breeds and lines can cause that some genes may disappear. Gardini and Villa (2003) reported that local breeds are an evidence of great achievement of many generations of breeding. For centuries, farmers have been adapting chickens to local conditions, cultural needs and preferences. Unfortunately, over last decades, as a result of the industrialization of agriculture, the old poultry breeds in Europe mostly suffered a graduate decrease in numbers.

The Research Centre of Animal Production in Nitra (APRC Nitra) deals with the conservation of poultry genetic resources not only on a methodology basis but also contributes to the maintaining of the local Oravka breed. This breed was formed by crossbreeding of the local hens in the Orava region with Rhode Island,

Wyandotte and New Hampshire breeds. The breeding programme aimed at forming a dual-purpose breed with good egg production, growth ability and adaptability to harsh environment started in 1950ies (Kadlečík *et al.*, 2004). The breeding programme consisted of three consecutive phases (Chmelničná, 2004), and Oravka breed was recognized in 1990.

Oravka is a dual-purpose breed kept for egg and meat production, respectively. The animals are of yellowish-brown colour and of rectangular body frame. The live weight of adult females is 2.2 to 2.7 kg and that of males is 2.8 to 3.3 kg. About 180 to 200 eggs per female and year are produced. The egg shell is brownish. The minimum hatching egg weight is 55 g (Hrnčár, 2008).

A survey of Oravka living animals (breeding males and females) was done by Hrnčár and Weis (2007) and Oravcová *et al.* (2010), respectively.

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Hrnčár and Janesová (2006) compared growth intensity between Oravka breed and some chosen breeds (New Hampshire, Plymouth, Rhode Island and Sussex). The growth of Oravka breed differed only from New Hampshire breed. The same results were confirmed by Hrnčár *et al.* (2010), who referred that New Hampshire chicken had the higher live weight from 8 weeks of age. The effect of sire on live weight of descendants was investigated by Hanusová *et al.* (2012). The production parameters (egg weight, growth) of Oravka breed were observed by Weis and Hrnčár (2009) and Hanusová *et al.* (2010). A study on Oravka reproduction traits over the recent years has not been done until now.

Therefore, the goal of this study was to analyse growth and some reproduction (fertilization, hatchability and egg weight) traits in Oravka population reared in the APRC Nitra.

MATERIAL AND METHODS

Growth and some reproduction traits (fertilization, hatchability and egg weight) of Oravka breed were measured in two lines (OR/2 and OR/3). The in situ conservation flock kept in the Animal Production Research Centre Nitra (APRC Nitra) during the period between 2009 and 2012 was included in the experiment. Each season the breeding males outside the APRC Nitra flock were used. The breeding females originated from mating between females raised in the APRC Nitra flock and these new males.

Chicken were kept until 12 weeks of age indoor in a heating room. Birds were placed in the weaning pen with wood shavings litter. From 12 weeks of age, they were kept outdoor in a heatless hen-house with covered yard. Feeding and watering was *ad libitum*. They were

fed with standard feed mixture which differed between age categories, but was the same within each category during the analyzed period. The chickens were exposed to the natural light.

The animals were weighed individually at 5, 12 and 20 weeks of age. The reproduction traits: the number of setting eggs, the number of fertilized eggs and the number of hatched chickens were recorded. The fertilization and hatchability were calculated. The eggs were weighed individually each month for a period of 7 days. Each season the parent's reproductive traits and growth of their offspring were measured.

The basic statistic characteristics were calculated using the SAS/STAT 9.2. software (2002-2008).

RESULTS AND DISCUSSION

The Oravka chicken growth during the three seasons is given in Tables 1 and 2. The growth traits of two lines at the age of 5, 12 and 20 weeks, regardless of sex, are given in Table 1. In Table 2, the growth traits by sex are shown. The lowest weight of 5 weeks of age had animals in season 2011/12. Line OR/2 had higher weight during the whole experiment in 20 weeks of age. The males had more intensive growth than females from 5 weeks of age (except for OR/3 line in season 2011/2012).

The body weight in our experiment was similar to that observed by Hrnčár *et al.* (2010) till the age of 12 weeks. As a difference, 20 weeks old females were heavier than those of the same age referred by Hrnčár *et al.* (2010).

Table 1: Live weight of animals of Oravka breed by season and line

Season	Line	n	Live weight (g)				
			5-week old		12-week old		20-week old
			$\bar{x} \pm SD$	n	$\bar{x} \pm SD$	n	$\bar{x} \pm SD$
2009/	OR/2	107	450.6 ± 64.4	88	1421.1 ± 245.4	33	2286.1 ± 535.4
2010	OR/3	78	468.2 ± 72.1	53	1464.5 ± 242.2	16	2076.3 ± 381.8
2010/	OR/2	93	538.2 ± 79.4	93	1318.7 ± 240.1	31	2258.5 ± 447.1
2011	OR/3	62	486.9 ± 85.2	62	1246.1 ± 254.0	22	2161.8 ± 255.0
2011/	OR/2	70	371.3 ± 69.0	60	1281.3 ± 233.8	27	2129.6 ± 468.3
2012	OR/3	103	383.6 ± 81.8	84	1373.9 ± 234.2	35	2106.9 ± 358.6

Table 2: Live weight of animals of Oravka breed by season, line and sex

Season	Line	Live weight (g)											
		5-week old				12-week old				20-week old			
		n	$\bar{x} \pm SD$	♀	♂	n	$\bar{x} \pm SD$	♀	♂	n	$\bar{x} \pm SD$	♀	♂
2009/2010	OR/2	41	419.8 ± 40.8	45	486.0 ± 54.7	43	1260.2 ± 146.9	45	1574.9 ± 221.4	23	1993.0 ± 209.6	10	2960.0 ± 433.4
	OR/3	30	444.00 ± 55.9	22	502.3 ± 73.2	28	1313.2 ± 158.7	25	1364.0 ± 205.2	14	1977.1 ± 285.9	2	2770.0 ± 155.6
2010/2011	OR/2	41	499.5 ± 67.5	52	575.4 ± 72.6	41	1134.8 ± 162.2	52	1463.7 ± 186.4	21	2054.7 ± 274.3	10	2605.0 ± 481.5
	OR/3	32	445.7 ± 74.8	30	523.4 ± 79.0	32	1076.6 ± 154.5	30	1427.0 ± 211.3	14	2042.9 ± 216.8	8	2370.0 ± 173.2
2011/2012	OR/2	30	343.3 ± 64.2	38	390.0 ± 66.1	26	1116.2 ± 129.0	34	1407.6 ± 217.2	15	1766.0 ± 246.3	12	2584.2 ± 188.3
	OR/3	40	368.9 ± 77.2	53	366.7 ± 87.3	38	1243.2 ± 215.1	46	1482.0 ± 191.8	27	1962.6 ± 226.5	8	2593.8 ± 287.7

The reproduction traits of the two Oravka lines are given in Table 3. The fertility of eggs during the experiment was higher than 84 per cent except for season 2011/2012 in line OR/2. In this line, fertility was only 40.18 % since the cock had hormonal disorder. It had a higher level (82.72 pmol/l) of estradiol (predominant sex hormone present in females) and low level (0.861 pmol/l) of testosterone (male sex hormone). The cock in line OR/3 had 1.809 pmol/l of testosterone and no estradiol. Hatchability from pickled eggs was similar in both lines within the two seasons. Only in the third season, the hatchability was higher in line OR/3.

The eggs were weighed individually for a period of 7 days each month (10 laying months in total). The average egg weight within the three most intensive laying months is given in Table 4. The average weights of eggs were from 53.9 ± 3.5 g to 56.6 ± 4.3 g in OR/2 and from 52.7 ± 4.2 g to 56.4 ± 3.6 g in OR/3 during the whole experiment. The average egg weight had an increasing tendency in line OR/2 during the whole experiment. Benková *et al.* (2003) detected the average egg weight 49.48 g in 1996, 54.87 g in 1998 and 55.24 g in 2001 in Oravka breed. Weis and Hrnčár (2009) observed the average weight of eggs 52.40 g in 2004. This value increased to 55.70 g in 2008.

CONCLUSION

The experiment showed good growth and reproduction traits of Oravka breed, which is known for good adaptability to harsh environment. The average live weights of OR/2 animals were from 371.3 ± 68.9 g to 538.1 ± 79.4 g at 5 weeks of age, from 1246.1 ± 254.0 g to 1464.5 ± 242.2 g at 12 weeks of age and from 2076.3 ± 381.8 g to 2286.1 ± 535.4 g at 20 weeks of age. The fertilization in each line and season was higher than 84 % except for OR/2 line in season 2011/2012. In the analyzed period, the improvement in egg weight in line OR/2 from 53.9 ± 3.5 g to 56.6 ± 4.3 g was observed. It is recommended to continue in selection aimed at an increasing egg production and egg weight to be in line with the standard of breed, which is as high as 180 - 200 eggs for egg production and 58 g for egg weight.

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Table 3: The reproduction traits of Oravka breed by season and line

Characteristic	Season 2009/2010		Season 2010/2011		Season 2011/2012	
	Line		Line		Line	
	OR/2	OR/3	OR/2	OR/3	OR/2	OR/3
Number of setting eggs	183	133	158	132	224	164
Number of fertilized eggs number	164	112	142	127	90	143
Fertilization (%)	89.6	84.2	89.9	96.3	40.2	86.7
Hatchability						
(%) Fr. pickled eggs	72.1	67.7	79.8	84.9	33.9	84.4
Fr. fertilized eggs	80.5	80.3	88.7	88.2	73.9	85.3

Table 4: Average egg weights of Oravka breed by line in the most intensive laying period (March-May)

Season	Line	Month	Eggs number	Weight (g) $\bar{x} \pm SD$
2009/2010	OR/2	III/10	69	53.9 \pm 3.5
		IV/10	70	54.8 \pm 3.8
		V/10	59	54.4 \pm 4.3
	OR/3	III/10	40	54.7 \pm 3.2
		IV/10	45	54.4 \pm 4.8
		V/10	36	55.7 \pm 7.1
2010/2011	OR/2	III/11	86	54.5 \pm 3.9
		IV/11	83	55.3 \pm 4.1
		V/11	62	54.0 \pm 4.1
	OR/3	III/11	59	56.4 \pm 3.6
		IV/11	53	55.8 \pm 3.9
		V/11	46	54.8 \pm 3.7
2011/2012	OR/2	III/12	75	55.3 \pm 3.8
		IV/12	61	56.3 \pm 5.3
		V/12	60	56.6 \pm 4.3
	OR 3	III/12	59	53.0 \pm 4.5
		IV/12	42	53.3 \pm 5.4
		V/12	40	52.7 \pm 4.2

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EFFECT OF GENOTYPE ON EGG QUALITY CHARACTERISTICS OF JAPANESE QUAIL (*COTURNIX JAPONICA*)

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ABSTRACT

A research was carried out to determine effect of different genotypes of Japanese quail (*Coturnix japonica*) in genetic resource on external and internal egg quality parameters. The birds were housed as 1 male and 3 females per cage of 0.12 m² area at Animal Production Research Centre Nitra and fed with a mixture of 9.0 MJ ME and 145.0 g of crude protein during the experiment. Feed and water were given *ad libitum*. Analysis of external and internal characteristics of Japanese quail eggs was performed in the laboratory of the Department of Poultry Science and Small Animal Husbandry at the Faculty of Agrobiological and Food Resources of the Slovak University of Agriculture in Nitra. This research was conducted to investigate the effects of genotype on egg weight, egg length, egg width, egg shape index, shell weight, percentage of shell, shell thickness, shell strength, albumen weight, percentage of albumen height, albumen width, albumen length, albumen index, Haugh unit, yolk weight, percentage of yolk, yolk height, yolk width, yolk index and yolk colour. We have found significantly higher values for meat type in terms of all egg parameters ($P \leq 0.05$). In case of shell parameters, we observed significant ($P \leq 0.05$) difference between genotypes only in shell weight in benefit of the meat type and significant ($P \leq 0.05$) higher value in shell strength for laying type. There were significant ($P \leq 0.05$) differences found between the genotypes in points of albumen height and albumen index for laying Japanese quail. The significant ($P < 0.05$) difference in benefit of the meat type was found in yolk weight, yolk percentage, yolk height and yolk index. For all other characteristics no significant differences in egg quality between the laying and the meat type of Japanese quail were observed.

Key words: Japanese quail; egg; external quality; internal quality

INTRODUCTION

The Japanese quail, *Coturnix japonica* is known to have been domesticated since the 12th century AD in Japan, mainly for its ability to sing. Intensive production of the species started in Japan in the 1920s. The first egg lines were then developed by selection. They were successfully introduced from Japan to America, Europe and Middle East between the 1930s and 1950s, where specific lines were bred for egg and meat production (Ashok and Prabakaran, 2012). Extensive research on *Coturnix japonica* has showed that it was a valuable animal for avian research (Baumgartner *et al.*, 2007; Jung *et al.*, 2009).

A Japanese quail, the smallest farmed avian

species (Panda and Singh, 1990), is becoming popular in commercial poultry sector for meat and egg production. Distinct include rapid growth – enabling quail to be marketed for consumption at 5-6 wks of age, early sexual maturity - resulting in short generation interval, high rate of egg laying and much lower feed and space requirements than domestic fowl. The Japanese quail is a bird with a high production potential which can lay up to 350 eggs of 10-12 grams each, which means 20 times her body weight.

Egg quality has been defined as the characteristics of an egg that affect its acceptability by the consumers. Egg quality is the more important price contributing factor in table and hatching eggs. Therefore, the economic success of a laying flock solely depends on

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the total number of quality eggs produced (Monira *et al.*, 2003).

Egg quality is composed of those characteristics of an egg that affects its acceptability to consumers, it is therefore important that attention is paid to the problems of preservation and marketing of eggs to maintain the quality. Among many quality characteristics, external factors including cleanliness, freshness, egg weight and shell weight are important in consumer's acceptability of shell eggs (Song *et al.*, 2000; Adeogun and Amole, 2004, Dudusola, 2010). On the other hand, interior characteristics such as yolk index, Haugh unit, and chemical composition are also important in egg product industry as the demand for liquid egg, frozen egg, egg powder and yolk oil increases (Scott and Silversides, 2001).

The most studies of egg production in quail was in laying type of Japanese quails (Garcia *et al.*, 2000; Ribeiro *et al.*, 2003; Murakami *et al.*, 2006; Araujo *et al.*, 2007; Murakami *et al.*, 2007), whilst there are few studies on the egg production potential in meat type of Japanese quails (Mori *et al.*, 2005; Barreto *et al.*, 2007).

From the point of view of consumers, egg weight is the most essential quality trait. In Japanese quails, this trait is related to genetic structure of flock (Rajkumar *et al.*, 2009), sexual maturity (Kumar *et al.*, 2000), production type (Panda and Singh, 1990), nutrition (Güçlü *et al.*, 2008), the stage of production cycle (Yanakopolous and Tserveni-Gousi, 1986, Silversides and Scott, 2001; Nowaczewski *et al.*, 2010), housing density (Bhanja *et al.*, 2006) and other. Another important exterior

trait is the egg shell integrity.

According to the extent of its damage, eggs could be divided into three groups – with broken external and internal cracks. Egg shell integrity is important not only from economic point of view, but also with regard to human health safety (Genchev, 2012).

This study was designed to determine the effect of genotype on some internal and external quality characteristics of Japanese quail eggs.

MATERIAL AND METHODS

The study was performed in the laboratory of the Department of Poultry Science and Small Animal Husbandry at the Faculty of Agrobiological and Food Resources of Slovak University of Agriculture in Nitra. In the experiment we used the eggs laying type and meat type of Japanese quail obtained from the experimental farm at Animal Production Research Centre Nitra in Lužianky.

Interior and exterior parameters of eggs quality were evaluated at 20 weeks of age, within 24 h of collection. We analysed thirty eggs at each evaluation time. Throughout the study, 35 birds were maintained in normal environmental conditions and housed in the proportion 1 male/3 females per cage of 0.12 m² area. During the egg production period, Japanese quails were fed *ad libitum* commercial feed mixture HYD-10 for laying hens and quails (Tekro, Slovak Republic). Nutritional value of diets is shown in Table 1.

Table 1: Nutritional value of complete feed mixture HYD-10

Nutrient	Unit	HYD-10
Crude protein	g/kg	min. 145.0
ME	MJ/kg	min. 9.0
Lysine	g/kg	min. 5.0
Methionine and cistine	g/kg	min. 5.0
– from that methionine	g/kg	min. 2.0
Calcium	g/kg	min. 27.0
Phosphorus	g/kg	min. 4.0
Sodium	g/kg	min. 1.0
Manganese	mg/kg	min. 50.0
Iron	mg/kg	min. 70.0
Copper	mg/kg	min. 50.0
Zinc	mg/kg	min. 40.0
Vitamin A	i.u./kg	min. 8000
Vitamin D ₃	i.u./kg	min. 1500

Egg weight was individually determined to 0.01g accuracy using a laboratory scale Owa Labor (VEB Wägetechnik Rapido, Germany). Egg length (along the longitudinal axis) and egg width (along the equatorial axis) were measured with a micrometer. Egg shape index was calculated as the ratio of egg width to length (%) by the method of Anderson *et al.* (2004).

After the eggs were broken, egg shells were washed with water and dried in order to clean the remaining albumen. Following this procedure, shell weight (with membrane) was measured using a laboratory scale Owa Labor (VEB Wägetechnik Rapido, Germany) and the percentage proportion of the shell in the egg was determined. Shell thickness (with membrane) was measured at the sharp poles, blunt poles and equatorial parts of each egg. Shell thickness was obtained from the average values of these three parts. The egg shell strength was determined manually using an Egg Crusher device (VEIT Electronics, Czech Republic).

The albumen weight was calculated from the difference between the egg weight, and the yolk and shell weight and the percentage proportion of the albumen in the egg was determined. Albumen index (%) was determined by the method of Alkan *et al.* (2010) on the basis of the ratio of the thick albumen height (mm) measurement taken with a micrometer to the average of width (mm) and length (mm) of this albumen with 0.01mm accuracy. Haugh unit was calculated according to the procedure of Haugh (1937).

Yolk weight with 0.01 g accuracy was determined using the laboratory scale Owa Labor (VEB Wägetechnik Rapido, Germany) and its percentage proportion was calculated.

Yolk index (%) was measured on the basis of the ratio of the yolk height (mm) to the yolk width (mm) by the method of Funk (1948) using micrometer with 0.01mm accuracy.

Yolk colour was determined with the scale of Hoffman La Roche (Hoffman–La Roche, Switzerland).

The evaluated variables were submitted to analysis of variance using Statistical Analysis System software package (SAS, 2003). The significance of differences between the genotypes was tested by the Tukey's test at the levels of significance.

RESULTS AND DISCUSSION

Exterior and interior quality characteristics of eggs have also been investigated in several studies on quails (Yanakopolous and Tserveni-Gousi, 1986; Uluocak *et al.*, 1995; Altan *et al.*, 1998; Minvielle *et al.*, 2002; Mignon-Grasteau and Minvielle, 2003; Bardakçioğlu *et al.*, 2005).

Egg weight is among the most important parameters not only for consumers, but for egg producers as well (Genchev, 2012). In our experiment, average egg weight was significantly ($P \leq 0.05$) affected by the quail type. The eggs meat type of Japanese quail weighed 13.06 g in average, similarly to that reported by Mori *et al.* (2005) and Santos *et al.* (2011). Whereas the eggs laying type of Japanese quail weighed 11.48 g in average, what is consistent with those obtained by Garcia *et al.* (2000), Kadam *et al.* (2006), Murakami *et al.* (2006), Oliveira *et al.* (2007) and Murakami *et al.* (2008). Our results correspond to the findings by Gonzales (1995). The egg shape index was significantly ($P \leq 0.05$) influenced by the genotype in benefit of meat type. In contrast, in the shell strength we found statistically significant value for laying type in comparison with meat type of Japanese quail.

Table 2: The mean values of the exterior egg quality parameters in Japanese quails

Parameter	Laying type	Meat type
Egg weight (g)	11.48 ± 1.72	13.06 ± 2.05 ^b
Egg width (mm)	25.71 ± 0.75	26.94 ± 0.77 ^b
Egg length (mm)	33.52 ± 1.89	34.46 ± 1.92 ^b
Egg shape index (%)	76.70 ± 0.67	78.18 ± 0.69 ^b
Shell weight (g)	1.02 ± 0.05	1.16 ± 0.07 ^b
Shell percentage (%)	8.88 ± 0.26	8.89 ± 0.25
Shell thickness (mm)	0.25 ± 0.11	0.23 ± 0.10
Shell strength (N.cm ⁻²)	6.59 ± 1.35 ^a	6.46 ± 1.37

Values shown are mean ± SD (standard deviation)

a, b values in rows with no common superscripts differ significantly ($P < 0.05$)

Table 3: The mean values of the interior egg quality parameters in Japanese quails

Parameter	Laying type	Meat type
Albumen weight (g)	6.75 ± 0.24	7.52 ± 0.31 ^b
Albumen percentage (%)	58.78 ± 0.55	58.39 ± 0.52
Albumen length (mm)	50.14 ± 0.49	49.82 ± 0.44
Albumen height (mm)	4.82 ± 0.14 ^a	4.16 ± 0.10
Albumen width (mm)	38.27 ± 0.61	38.31 ± 0.58
Albumen index (%)	10.12 ± 0.38 ^a	9.45 ± 0.32
Haugh Unit	87.28 ± 0.49	87.56 ± 0.51
Yolk weight (g)	3.72 ± 0.11	4.28 ± 0.14 ^b
Yolk percentage (%)	32.43 ± 0.48	35.84 ± 0.56 ^b
Yolk height (mm)	11.19 ± 0.12	12.11 ± 0.14 ^b
Yolk width (mm)	25.88 ± 0.15	26.41 ± 0.17
Yolk index (%)	43.22 ± 0.31	45.86 ± 0.28 ^b
Yolk colour (°LR)	4.30 ± 0.84	4.40 ± 0.86

Values shown are mean ± SD (standard deviation)

a, b values in rows with different superscripts differ significantly ($P < 0.05$)

Higher egg weight in the meat type of Japanese quail caused statistically ($P \leq 0.05$) higher egg shell weight. The most important quality traits of the egg shell are its strength and thickness. There were no significant ($P > 0.05$) differences between the genotypes for egg shell thickness. The egg shell thickness values in this research (0.22 and 0.23 mm) were somewhat higher in comparison to Kostova *et al.* (1993), Gonzales (1995), Altan *et al.* (1998) and Orhan *et al.* (2001), who reported values from 0.19 to 0.22 mm. In case of shell strength we observed similar values (6.47, respectively 6.46 N.cm⁻²). Compared to hens' eggs, those from quail had poorer shell quality, as judged by shell thickness and shape (Fletcher *et al.*, 1983; Zita *et al.*, 2013).

Quail eggs have higher proportions of yolk than those from hens (Fletcher *et al.*, 1983; Zita *et al.*, 2013). From yolk characteristics of eggs, the genotype significantly ($P \leq 0.05$) affected yolk weight and yolk index for the meat type of quail. Yolk index values in this study (43.22, respectively 45.86 %) were in agreement with the data reported for yolk index in quail in the literature (Orhan *et al.*, 2001; Erensaym and Camci, 2002). The yolk colour was not affected by the genotype.

The significantly ($P \leq 0.05$) higher albumen index for laying Japanese quail was in accordance with albumen height differences. There was no significant ($P > 0.05$) difference determined between the laying and meat type for the Haugh Unit. The Haugh Unit values in this study (87.28 and 87.56) were in agreement with the data reported in literature for Haugh Unit, such as

85.53-95.21 in quail (Altan *et al.*, 1998; Türkmüt *et al.*, 1999). The higher the Haugh unit and yolk index, the more desirable is the interior quality of the egg (Adeogun and Amole, 2004). Quail eggs have lower proportions of albumen than those from hens (Fletcher *et al.*, 1983; Zita *et al.*, 2013).

CONCLUSION

According to the results obtained in this work we can conclude that for several parameters of the external and internal egg quality statistically significant differences between the laying and meat type of Japanese quail were observed. The most significant differences in benefit of the meat type were recorded for egg, shell, albumen and yolk weights and also for the ratio of each parts of the egg. In contrast, the laying type showed a better value of the egg strength and albumen parameters. The genotype along with nutrition, health, age, maintenance, storage condition of eggs and storage period can affect characteristics of egg quality.

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MODEL COMPARISONS AND GENETIC PARAMETER ESTIMATES OF GROWTH TRAITS IN BALUCHI SHEEP

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ABSTRACT

Genetic and non-genetic parameters were estimated for growth traits and average daily weight gains of Iranian Baluchi lambs using univariate and multivariate models. Data on body weight collected for a period of 25 years (1984-2009) were used to model the growth trajectory and estimate genetic parameters. Studied traits were birth weight (BW), 3-month weight (3MW), 6-month weight (6MW), 9-month weight (9MW), yearling weight (YW), pre-weaning average daily gain (ADG1) and post-weaning average daily gain (ADG2). Genetic parameters were estimated using the restricted maximum likelihood (REML) procedure under univariate and multivariate animal models. Random effects were explored by fitting additive direct genetic effects, maternal additive genetic effects, maternal permanent environmental effects, the covariance between direct and maternal genetic effects and common litter effects in twelve different models for analysis of each trait. The heritability, estimated from the most appropriate model for BW, 3MW, 6MW, 9MW, YW, ADG1 and ADG2 trait, were 0.062 ± 0.02 , 0.12 ± 0.02 , 0.16 ± 0.03 , 0.21 ± 0.03 , 0.17 ± 0.03 , 0.08 ± 0.02 and 0.1 ± 0.02 , respectively. The maternal heritabilities of these traits were 0.09 ± 0.02 , 0.04 ± 0.01 , 0.045 ± 0.017 , 0.015 ± 0.02 , 0.02 ± 0.012 , 0.03 ± 0.01 and 0.05 ± 0.02 , respectively. The present study shows the importance of inclusion of maternal effects in designing appropriate breeding programs for genetic improvement in Baluchi lambs for growth traits.

Key words: growth traits; average daily weight gain; variance components; heritability; Baluchi sheep

INTRODUCTION

The sheep population in Iran in 2011 was about 54 million heads, including 27 breeds and ecotypes (the Iranian ministry of agriculture, 2011). Among them Baluchi sheep is one of the most widely occurred breed, which represents approximately 30 % (near to 15 million head) of total sheep population (Madad and Ghazanfari, 1999). The body colour is generally white with black spots at the end of the muzzle, ears, eyes, and metacarpus and metatarsus area. This breed is widely distributed from north-east to south-east of the country and is reared mainly for meat purposes.

Growth rate of animals is influenced not only by direct additive genetic effects but also affected by

maternal genetic and maternal permanent environment. Results of several studies showed that including of the maternal effects into models caused more accurate estimation of (co)variance and genetic parameter of production and reproductive traits (Miraei-Ashtiani *et al.*, 2007; Zamani and Mohammadi, 2008; Mohammadi *et al.*, 2013ab).

Thus, accurate estimation of (co)variance components is outcome for designing any breeding program and genetic evaluation system. Because of lack of such comprehensive estimates for growth traits of Baluchi sheep in Iran this study has been performed with the objective of accurate estimation of (co)variance components and corresponding genetic parameters for growth traits of Baluchi sheep.

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MATERIAL AND METHODS

Flock management and data sources

Data used in the present study were collected from Breeding Sheep Center, located in North East of Iran in Mashhad, Khorasan Razavi province. During the spring and summer, the flock was kept on pastures and in the autumn it was grazed on wheat and barley stubbles. During the winter, the lambs were kept indoors and hand-fed. Supplementary feed, offered to all animals during winter and to ewes late in pregnancy, consisted of wheat and barley straw, alfalfa hay, sugar beet pulp and concentrate. The investigated traits in this study were: birth weight (BW), 3-month weight (3MW), 6-month weight (6MW), 9-month weight (9MW), yearling weight (YW), pre-weaning average daily gain (ADG1) and post-weaning average daily gain (ADG2) with using records of 45,656 lambs of 1,380 sires and 13,988 dams born between 1984 to 2009. The structure of the data used in the analysis is shown in Table 1.

Statistical analysis

Data were analyzed by a least squares analysis of variance using the general linear model (GLM) procedure of the SAS software package (SAS, 2004). The fixed effects considered were: sex of lambs in two classes (male-female), type of birth in three classes (single, twins, triplets), age of the dam at lambing in seven classes (2 to 8 years old), year of birth in 26 classes (1984 to 2009) and number of flocks in eight classes (1 to 8), respectively. The interactions between fixed factors were not significant and, therefore, these factors were excluded from the final model. Moreover, the age of lambs was placed in the model as a covariate factor. (Co)variance components and corresponding genetic parameters for the studied traits were estimated with the help of twelve univariate animal models. Tested models (in matrix notation) were as follows:

$$\begin{array}{ll}
 y = Xb + Z_a a + e & \text{Model (1)} \\
 y = Xb + Z_a a + Z_c c + e & \text{Model (2)} \\
 y = Xb + Z_a a + Z_m m + e & \text{Cov(a,m) = 0 Model (3)} \\
 y = Xb + Z_a a + Z_m m + e & \text{Cov(a,m) = } A\sigma_{am} \text{ Model (4)} \\
 y = Xb + Z_a a + Z_m m + Z_c c + e & \text{Cov(a,m) = 0 Model (5)} \\
 y = Xb + Z_a a + Z_m m + Z_c c + e & \text{Cov(a,m) = } A\sigma_{am} \text{ Model (6)} \\
 y = Xb + Z_a a + Z_4 l + e & \text{Model (7)} \\
 y = Xb + Z_a a + Z_c c + Z_l l + e & \text{Model (8)} \\
 y = Xb + Z_a a + Z_m m + Z_l l + e & \text{Cov(a,m) = 0 Model (9)} \\
 y = Xb + Z_a a + Z_m m + Z_l l + e & \text{Cov(a,m) = } A\sigma_{am} \text{ Model (10)} \\
 y = Xb + Z_a a + Z_m m + Z_c c + Z_l l + e & \text{Cov(a,m) = 0 Model (11)} \\
 y = Xb + Z_a a + Z_m m + Z_c c + Z_l l + e & \text{Cov(a,m) = } A\sigma_{am} \text{ Model (12)}
 \end{array}$$

Where y is a vector of records for the different traits; a , b , c , m , l and e are vectors of direct additive genetic effects, fixed effects, maternal permanent

environmental effects, maternal additive genetic, common environmental and residual effects, respectively; X , Z_a , Z_m , Z_c and Z_l are design matrices associating the fixed effects, direct additive genetic effects, maternal permanent environmental effects, maternal additive genetic effects and common environmental effects to vector of y , respectively. All the means of random effects are equal to zero. In the matrix notation, the (co)variance structure was as follows:

$$v \begin{bmatrix} a \\ m \\ c \\ l \\ e \end{bmatrix} = \begin{bmatrix} A\sigma_a^2 & A\sigma_{am} & \circ & \circ & \circ \\ A\sigma_{am} & A\sigma_m^2 & \circ & \circ & \circ \\ \circ & \circ & Id\sigma_c^2 & \circ & \circ \\ \circ & \circ & \circ & Il\sigma_l^2 & \circ \\ \circ & \circ & \circ & \circ & In\sigma_e^2 \end{bmatrix},$$

where A is the additive numerator relationship matrix, σ_a^2 , is the direct additive genetic variance, σ_m^2 is the maternal additive genetic variance, σ_{am} is the direct-maternal additive genetic covariance, σ_c^2 is the maternal permanent environmental variance, σ_l^2 is the common environmental variance, σ_e^2 is the residual variance and I_d , I_c and I_n are identity matrixes with orders equal to number of dams, litters and records, respectively. Also, in these models σ_{am} is the (co)variance of direct and maternal additive genetic effects. All traits were analyzed with WOMBAT software package by AI-REML algorithm (Meyer, 2006). The most appropriate model for each trait was selected based on Akaike's information criterion (AIC) (Akaike, 1974):

$$AIC_i = -2 \log L_i + 2 p_i$$

where $\log L_i$ represents the maximized log likelihood, and p_i is the number of parameters obtained for each model. The model that has the lowest AIC, is the appropriate model for that trait. Total heritability was estimated according to the following equation:

$$h_t^2 = \frac{\sigma_a^2 + 0.5 \sigma_m^2 + 1.5 \sigma_{a,m}}{\sigma_p^2}$$

Genetic and phenotypic correlations were estimated using bivariate analyses applying the best model determined in univariate analyses. If the values of $-2 \log$ likelihood variance in the Simplex function were below 10^{-8} , it was assumed convergence had been achieved (Mohammadi *et al.*, 2013b).

Table 1: Descriptive statistics for traits studied

Traits ^a	No. of records	Mean (kg)	SD ^b (kg)	CV (%)	No. of dams	No. of sires
BW	13682	3.9	0.8	20	3648	371
3MW	10015	22.9	5.27	23	2842	267
6MW	8150	29.8	5.56	18	2624	249
9MW	7194	33.1	5.4	16	2480	247
YW	6615	38.8	6.83	17	2394	246
ADG1	10015	0.20	0.061	30	2842	267
ADG2	8119	0.034	0.022	64	2624	249

Traits: BW: birth weight, 3MW: 3 month weight, 6MW: 6 month weight, 9MW: 9 month weight, YW: yearling weight, pre-weaning average daily gain: ADG1 and post-weaning average daily gain: ADG2

Table 2: Least square means \pm SE of pre- and post-weaning growth traits of Baluchi lambs

Fixed effects	Traits ^a						
	BW(kg)	3MW(kg)	6MW(kg)	9MW(kg)	YW(kg)	ADG1(kg)	ADG2
Overall mean	3.9 \pm 0.80	22.9 \pm 5.27	29.8 \pm 5.56	33.1 \pm 5.40	38.8 \pm 6.83	0.2 \pm 0.061	0.03 \pm 0.022
Sex	**	**	**	**	**	**	**
Male	4.06 ^a \pm 0.81	23.9 ^a \pm 4.87	31.29 ^a \pm 4.73	34.61 ^a \pm 5.55	41.23 ^a \pm 6.96	0.214 ^a \pm 0.062	0.037 ^a \pm 0.020
Female	3.81 ^b \pm 0.87	21.87 ^b \pm 5.44	28.41 ^b \pm 5.91	31.55 ^b \pm 4.77	36.4 ^b \pm 5.70	0.19 ^b \pm 0.058	0.030 ^b \pm 0.012
Type of birth	**	**	**	**	**	**	**
Single	4.30 ^a \pm 0.74	25.24 ^a \pm 4.9	31.83 ^a \pm 5.30	34.57 ^a \pm 5.39	40.06 ^a \pm 6.9	0.22 ^a \pm 0.06	0.033 ^a \pm 0.02
Twin	3.66 ^b \pm 0.66	21.04 ^b \pm 4.6	28.25 ^a \pm 5.05	31.80 ^{ab} \pm 4.90	37.7 ^a \pm 6.5	0.18 ^b \pm 0.05	0.035 ^a \pm 0.03
Triplet	3.02 ^c \pm 0.70	19.43 ^b \pm 4.8	26.67 ^a \pm 5.23	30.50 ^a \pm 5.40	36.50 ^a \pm 6.7	0.17 ^b \pm 0.05	0.034 ^a \pm 0.03
Age of dam (Year)	**	**	*	*	**	**	*
2	3.81 ^a \pm 0.78	22.65 ^{ab} \pm 5.10	29.87 ^{ab} \pm 5.52	32.87 ^a \pm 5.42	38.18 ^a \pm 6.87	0.20 ^{ab} \pm 0.050	0.035 ^{ab} \pm 0.02
3	3.98 ^b \pm 0.80	23.13 ^{ab} \pm 5.21	30.07 ^a \pm 5.61	33.22 ^b \pm 5.44	38.94 ^b \pm 6.94	0.20 ^a \pm 0.059	0.033 ^{ab} \pm 0.02
4	3.97 ^b \pm 0.81	22.86 ^{ab} \pm 5.30	29.68 ^b \pm 5.44	33.23 ^b \pm 5.45	39.15 ^b \pm 6.80	0.20 ^b \pm 0.060	0.033 ^a \pm 0.02
5	3.99 ^{bc} \pm 0.79	22.94 ^{ab} \pm 5.20	29.95 ^{ab} \pm 5.63	33.05 ^{ab} \pm 5.21	38.63 ^{ab} \pm 6.65	0.20 ^a \pm 0.061	0.035 ^{ab} \pm 0.02
6	3.98 ^{bc} \pm 0.83	22.84 ^{ab} \pm 5.40	29.86 ^{ab} \pm 5.69	33.00 ^{ab} \pm 5.50	39.2 ^b \pm 6.90	0.20 ^{ab} \pm 0.060	0.034 ^{ab} \pm 0.02
7	4.04 ^{bc} \pm 0.80	22.92 ^{ab} \pm 5.10	29.97 ^{ab} \pm 5.49	33.4 ^{ab} \pm 4.94	39.5 ^b \pm 6.30	0.20 ^{ab} \pm 0.070	0.037 ^b \pm 0.02
8	4.18 ^c \pm 0.85	23.21 ^{ab} \pm 5.60	30.39 ^{ab} \pm 5.69	33.4 ^{ab} \pm 5.20	40.2 ^b \pm 6.50	0.18 ^{ab} \pm 0.070	0.039 ^b \pm 0.02
Year of birth	**	**	**	**	**	**	**

^afor trait abbreviations see footnote of Table 1. *P < 0.05; **P < 0.01; and ns: non-significant (P > 0.05)

RESULTS AND DISCUSSION

Fixed Factors

Least square means for studied traits are shown in Table 2. The result of variance analysis showed that the year of birth had significant effects on all studied

traits (p < 0.01). Sex of lamb had significant effect on all traits (p < 0.01). The significant effect of fixed factors in these characters could be assigned partly to the differences in the endocrine system of female and male lambs. Also, age of dam had significant effect on BW, 3MW, 6MW, 9MW, YM, ADG1, and ADG2 (p < 0.05).

Type of birth had a significant effect on weight changes in all traits ($p < 0.01$). Single born lambs had higher body weights and pre-weaning growth rate than twins and triplets.

Heritability estimates

Estimates of phenotypic variance using different models were generally similar for all considered traits. Residual variance was also similar in models 1 to 6, but was reduced when models 7 to 12 were fitted. The estimations of (co)variance components and corresponding genetic parameters are presented in Table 4. Also, determination of the most appropriate model of each trait is shown in bold in Table 3.

The most appropriate models for BW, ADG1, and 3MW were Model 12, 11 and 5 respectively. The most appropriate models for ADG2, 6MW, 9MW and YW were Model 9, 5, 9 and 10 respectively.

Maternal permanent environmental effects had a considerable impact on variation for BW, 3MW, 6MW, 9MW and ADG1. Maternal permanent environmental estimates of 0.13 were obtained for both ADG and 3MW. Estimated correlations between direct and maternal genetic effects for various traits are presented in Table 4. Estimates of the genetic correlation between direct and maternal genetic effects varied between traits and ranged from 0.47 for BW to 0.96 for YW, and 0.84 for ADG2, respectively.

Correlation estimates

Estimates of correlations between growth traits are presented in Table 5. There was no contrast

relationship between these traits in terms of phenotypic, genetic and environmental correlations accordingly, selection for any of these body weights will bring out positive response to selection for others. Estimates of additive genetic correlations between body weights were positive and high; varied from 0.60 for BW and YW to 0.97 for 9MW and YW. Phenotypic correlation estimates ranged from 0.30 for BW and YW to 0.79 for 9MW and YW and estimates of environmental correlation from 0.18 for BW and YW to 0.71 for 9MW and YW.

In general, the values observed in this study are in agreement with the estimates reported by the other researchers (Zamani and Mohammadi, 2008; Mohammadi *et al.*, 2013a). Maternal additive genetic correlation estimates between body weights were positive and ranged from 0.67 (between BW and 3MW) to 0.98 (between 9MW and YW).

Differences in managing practice, feed availability, climatic conditions and breeding systems through years, are possible reasons for significant effects of year on the considered traits (Mohammadi *et al.*, 2013a). According to the previous reports, the growth rate of female lambs was slower than in male lambs, and thus their weight was less, respectively (Mohammadi *et al.*, 2013b). Also, competition for milk consumption can be effective between twins and triplets particularly in pre-weaning period, which was consistent with other reports (Ozcan *et al.*, 2005). Including of birth age as a correlated variable into the statistical model (covariate) had a significant effect on all traits ($p < 0.01$).

The estimate of direct heritability for BW in

Table 3: AIC values ^a under different models for the body weight traits^b

Model	Traits						
	BW	3MW	6MW	9MW	YW	ADG1	ADG2
Model 1	-747.83	38393.30	32585.600	28200.62	27692.470	85775.160	55535.36
Model 2	-1290.25	38281.20	32524.440	28148.97	27673.068	85670.260	55535.36
Model 3	-1276.83	38304.90	32528.880	28152.35	27673.068	85702.006	55517.68
Model 4	-1275.82	104259	43013.640	28152.35	27670.480	85702.006	55517.68
Model 5	-1336.49	38275.6	32519.040	28150.53	27675.032	85666.060	55533.70
Model 6	-1337.63	38275.60	32519.060	28141.84	27667.900	85667.950	55519.10
Model 7	5182.90	38372.61	32549.740	28183.18	27677.130	85748.030	55509.18
Model 8	-1561.26	38281.14	32519.272	28144.00	27666.192	85667.740	55511.18
Model 9	-1579.16	38299.86	32528.890	28137.43	27668.078	85691.580	55495.32
Model 10	-1581.24	105059.50	69195.322	28138.45	27660.470	85691.580	55495.32
Model 11	-1608.06	38282.23	32526.774	28147.44	27668.000	85663.300	55510.64
Model 12	-1612.29	38283.06	32526.924	28138.45	27660.550	85665.230	55497.32

^a as deviations from the model with the lowest AIC value

^b for trait abbreviations see footnote of Table 1

Table 4: Estimates of (co) variance components and genetic parameters for the body weight traits with the best model

Traits ^a	Model	σ_a^2	σ_m^2	σ_{pe}^2	σ_l^2	$\sigma_{a,m}$	σ_e^2	σ_p^2	$h_a^2 \pm S.E$	$m^2 \pm S.E$	$c^2 \pm S.E$	$l^2 \pm S.E$	$r_{a,m} \pm S.E$	h_t^2
BW	12	0.023	0.034	0.03	0.87	0.013	0.18	0.37	0.062 ± 0.02	0.09 ± 0.02	0.09 ± 0.02	0.23 ± 0.01	0.47 ± 0.02	0.15
3MW	5	2.28	0.70	1.46	-	-	13.13	17.75	0.12 ± 0.02	0.04 ± 0.01	0.08 ± 0.01	-	-	0.15
6MW	5	3.56	0.95	1.29	-	-	15.32	21.11	0.16 ± 0.03	0.045 ± 0.10	0.06 ± 0.016	-	-	0.19
9MW	9	4.11	0.28	1.43	1.17	-	12.62	19.63	0.21 ± 0.03	0.015 ± 0.20	0.07 ± 0.02	0.06 ± 0.03	-	0.21
YW	10	4.41	0.50	-	2.43	1.44	16.90	25.79	0.17 ± 0.03	0.02 ± 0.02	-	0.09 ± 0.03	0.96 ± 0.37	0.25
ADG1	11	174	57.80	152	85.69	-	1542.6	2013	0.08 ± 0.02	0.03 ± 0.01	0.07 ± 0.01	0.13 ± 0.03	-	0.01
ADG2	9	35.26	16.54	-	45.007	-	273.4	349.9	0.10 ± 0.02	0.05 ± 0.02	-	0.13 ± 0.02	0.84 ± 0.08	0.24

σ_a^2 : direct additive genetic variance; σ_m^2 : maternal additive genetic variance; σ_{pe}^2 : maternal permanent environmental variance; σ_l^2 : common litter variance; $\sigma_{a,m}$: covariance between direct and maternal additive genetic effects; σ_e^2 : residual variance; σ_p^2 : phenotypic variance; h_a^2 : direct heritability; m^2 : maternal heritability; c^2 : ratio of maternal permanent environmental variance to phenotypic variance; l^2 : ratio of common litter variance to phenotypic variance; $r_{a,m}$: direct - maternal genetic correlation; *S.E*: standard error; h_t^2 : total heritability

^a for trait abbreviations see footnote of Table 1

Table 5: Correlation estimates among studied traits under bivariate animal models

Trait 1	Trait 2	r_{a12}^b	r_{p12}	r_{m12}	r_{e12}
BW	3MW	0.72 ± 0.08	0.40 ± 0.010	0.67 ± 0.04	0.29 ± 0.01
	6MW	0.65 ± 0.09	0.36 ± 0.010	0.69 ± 0.05	0.25 ± 0.02
	9MW	0.71 ± 0.08	0.35 ± 0.010	0.71 ± 0.06	0.21 ± 0.02
	YW	0.60 ± 0.10	0.30 ± 0.010	0.75 ± 0.08	0.18 ± 0.02
	ADG1	0.52 ± 0.12	0.25 ± 0.010	0.54 ± 0.05	0.17 ± 0.01
	ADG2	-0.11 ± 0.14	0.034 ± 0.010	0.40 ± 0.14	0.025 ± 0.02
3MW	6MW	0.89 ± 0.03	0.72 ± 0.010	0.97 ± 0.02	0.65 ± 0.01
	9MW	0.82 ± 0.05	0.63 ± 0.010	0.94 ± 0.03	0.55 ± 0.01
	YW	0.85 ± 0.04	0.58 ± 0.090	0.88 ± 0.05	0.49 ± 0.01
	ADG1	0.85 ± 0.03	0.88 ± 0.010	0.87 ± 0.02	0.89 ± 0.01
	ADG2	0.13 ± 0.14	-0.22 ± 0.010	0.18 ± 0.21	-0.30 ± 0.02
6MW	9MW	0.96 ± 0.02	0.78 ± 0.050	0.97 ± 0.02	0.70 ± 0.01
	YW	0.95 ± 0.02	0.71 ± 0.010	0.93 ± 0.03	0.63 ± 0.01
	ADG1	0.82 ± 0.05	0.58 ± 0.010	0.73 ± 0.05	0.51 ± 0.01
	ADG2	0.52 ± 0.10	0.55 ± 0.010	0.51 ± 0.17	0.58 ± 0.01
9MW	YW	0.97 ± 0.01	0.79 ± 0.050	0.98 ± 0.03	0.71 ± 0.03
	ADG1	0.70 ± 0.07	0.49 ± 0.010	0.78 ± 0.06	0.12 ± 0.02
	ADG2	0.66 ± 0.09	0.32 ± 0.010	0.52 ± 0.09	0.27 ± 0.02
YW	ADG1	0.82 ± 0.05	0.46 ± 0.010	0.63 ± 0.09	0.37 ± 0.02
	ADG2	0.60 ± 0.10	0.27 ± 0.012	0.26 ± 0.26	0.23 ± 0.02
ADG1	ADG2	0.31 ± 0.05	-0.21 ± 0.040	-0.12 ± 0.21	-0.27 ± 0.05

^a the symbols are the same as Table 1

r_{a12}^b : direct genetic correlation between trait 1 and trait 2; r_{p12} : phenotypic correlations between trait 1 and 2; r_{m12} : maternal additive genetic correlation between trait 1 and 2; r_{e12} : residual correlations between trait 1 and 2

the current study (0.062) is lower than in the report of Mohammadi *et al.* (2013b) (0.15). Lower heritability of birth weight compared to the other weights is related to the following reasons. Fetal growth is influenced by genetic and environmental factors such as the placenta and the fetal nutrition by a dam. Therefore, environmental factors affecting dam growth, especially the quality and quantity of food and the storage of food for dam can influence the growth of the embryo. The obtained direct heritability estimate of 0.08 for ADG1 agrees with those reported by Ozcan *et al.* (2005) and Ghafouri-Kesbi *et al.* (2008). There is higher estimate reported for direct heritability of ADG1 (Mohammadi *et al.*, 2013b; Abegaz *et al.*, 2007). In the present research the estimate of direct heritability for 3MW (0.12) corresponds to the data of Jafaroghli *et al.* (2010). Higher estimate (Mohammadi *et al.* 2013a; 0.16; Mohammadi *et al.* 2013b; 0.19) have also been reported. The reason for low heritability is that the lambs are more affected by breast milk during infancy. Estimated m^2 for birth weight, which is the ratio of maternal additive variance to phenotypic variance, is 0.09. Estimated maternal heritability of 0.03 for ADG1 agrees with that reported by Ghafouri-Kesbi *et al.* (2008). Thus, maternal effects and maternal power led to the increase in error variance and thus decrease in the heritability. The estimated m^2 for 3MW was 0.04, whilst Maria *et al.* (1993) stated it to be 0.34. Also, total heritability estimate for BW and 3MW (0.15) corresponds to those reported by Mohammadi *et al.* (2013a).

Low estimate of direct heritability obtained for ADG2 in the present study (0.1) is similar to the estimate reported by Ghafouri-Kesbi *et al.* (2008) -0.09. In contrast to present estimate, Abegaz *et al.* (2007) obtained lower values. The estimate of direct heritability for 6MW in this study (0.16) is higher than the estimate by Mohammadi *et al.* (2013b; 0.21) and is lower than by Ghafouri-Kesbi *et al.* (2008).

Also, the estimate of direct heritability for 9MW in this study (0.21) is approximately compatible with previous results in the Shal breed by Mohammadi *et al.* (2013a; 0.18). Moreover, the obtained direct heritability value for YW (0.17) was in accordance with the estimate of Mohammadi *et al.* (2013a; 0.19). As it is explicit, direct heritability has had upward trend, which has been proved by different researchers. The estimated value for maternal heritability of ADG2 (0.05) was in concordance with estimates of Mohammadi *et al.* (2013a) in Shal sheep. Also, maternal heritability for 6MW was estimated to be 0.07 (Abegaz *et al.*, 2007), whilst in our study this parameter was estimated to be 0.045. The estimate of maternal heritability for 9MW in the present study (0.015) is higher than the estimate published by (Ghafouri-Kesbi *et al.*, 2008) -0.05. The obtained maternal heritability value for YW (0.02) was

in accordance with the estimate of Notter *et al.* (1997; 0.05).

In addition, c^2 for 6MW was estimated to be 0.06, that was lower than the results reported by others researches (Mohammadi *et al.* (2013b; 0.06). The rate of c^2 for 9MW was estimated to be 0.07, which is in accordance with results of others researches (Ghafouri-Kesbi *et al.*, 2008; 0.02).

The results indicate that maternal additive genetic effects, which regard to the growth of fetus, could have some beneficial effect on the post-natal growth traits too. In the other words, body weight from birth to 6MW of age is partly influenced by similar genes of the dam in terms of maternal genetic effects.

Maternal genetic correlation for BW-3MW was 0.67, which is in agreement with the estimates of Abegaz *et al.* (2007) and Mohammadi *et al.* (2013b).

The estimates of correlations between growth rate and body weights are presented in Table 5. Phenotypic and direct genetic correlation estimates between post-weaning and pre-weaning growth rate was negative implying that different mechanisms are responsible for the expression of respective pre-weaning and post-weaning traits. Negative phenotypic and genetic correlation estimates were obtained for ADG1-ADG2.

It appears that lambs with higher gain in the pre-weaning period have less gain and are also less efficient during the post-weaning period at the phenotypic and genetic level. Similar to our estimate, a negative correlation between ADG1 and ADG2 has been reported by several authors (Abegaz *et al.*, 2007, Mohammadi *et al.*, 2010).

Direct genetic correlation estimates of post-weaning growth rate with BW was negative, whilst pre-weaning growth rate with BW was positive. Several authors have been reported results similar to our estimates (Abegaz *et al.*, 2007; Mohammadi *et al.*, 2013b). They stated that 3MW and ADG1 are genetically the same traits, and the selection can be performed based on one of them. Because Iranian farmers generally sell their lambs at 3MW, if selection is performed on 3MW, an improvement in 3MW and all correlated traits would be expected. Phenotypic correlations were varied from -0.22 between 3MW and ADG2 to 0.88 between ADG1 and 3MW. In general, these values were consistent with the published estimates of other researches (Abegaz *et al.*, 2007; Mohammadi *et al.*, 2013b).

CONCLUSION

The present research contributes to the model comparison and estimation of genetic parameters in fat-tailed sheep. It was observed that models containing both maternal genetic effects and direct genetic effects could better explain the genetic variation observed in

early growth traits. The genetic correlation between ADG1 and 3MW was positive, indicating that 3MW and ADG1 are genetically the same traits, and thus selection can be performed based on one of them. Because Iranian farmers generally sell their lambs at 3MW, if the selection is performed on 3MW, an improvement in 3MW and all correlated traits would be expected.

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EFFECT OF GENOTYPE ON PRODUCTION TRAITS IN BROILER CHICKENS

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ABSTRACT

The study was carried out on two broiler chicken genotypes Ross 308 and Cobb 500 in the nucleus poultry farm of the Institute of Agriculture – Stara Zagora between April and June 2012. For this purpose, 300 eggs of each genotype were set for incubation to compare the meat traits of two of the most popular broiler chicken hybrids: Cobb 500 and Ross 308 reared at the nucleus poultry farm of the Institute of Agriculture – Stara Zagora. The fertility rate, embryonic death rate, weight loss between incubation days 0 and 18, and the hatchability of set and fertilized eggs were determined. The number of chickens included in the study was 100 from each genotype (50 male and 50 female). Experimental birds were reared on wooden shavings bedded floor, with constant access to compound feed according to the age until 49 days of age. The live body weight was determined individually by weighing birds at 1, 14, 28, 42 and 49 days of age. By the end of the experiment, a slaughter analysis of three female and three male broilers with a live weight close to the group average was performed. For integral assessment of broiler combinations, the European Poultry efficiency factor (EPEF) was calculated. The results of the present experiments showed a number of differences in meat and slaughter traits between studied broiler chicken hybrids. The weight of hatchlings differed significantly according to the genotype ($p < 0.05$). One-day-old Cobb 500 broilers were heavier than Ross 308 broilers. At the end of the experiment, Cobb 500 broilers attained a higher live weight, and were heavier than Ross 308 birds by 6.29 %. The feed intake per kg weight gain over the entire experimental period was 2.178 kg and 2.181 kg for Ross 308 and Cobb 500, respectively. Higher values of the European Poultry Efficiency Factor (EPEF) were established in Cobb 500 broilers, which were more economically efficient than Ross 308 by 14.87 points (6.18 %). The performed slaughter analysis showed higher values of slaughter traits in Cobb 500, which had higher growth potential: roasting weight 1810.67 g and grilling weight 1710.50 g; whereas the respective values in Ross 308 chickens were 1547.67 g for grilling and 1645 g for roasting. In male Cobb 500 broiler chickens, the roasting percentage was 74.02 %, which was 1.41 % more than that of Ross 308 males. The same trend was observed in female birds as well, i.e.

Key words: broiler; European Poultry Efficiency Factor (EPEF); body weight; Cobb 500; Ross 308; slaughter analysis

INTRODUCTION

The modern broiler chicken production is an extensive and rapidly developing sector, supplying the market with relatively cheap and high-quality dietetic food. Due to contemporary selection programmes, a considerable improvement of weight gain, feed conversion, slaughter yield and breast meat yields were achieved during the past decades (Chambers *et al.*, 1981, Havenstein *et al.*, 1994a, 1994b). The progress in the selection of meat type chickens resulted in significantly

shorter fattening period up to 42 days of age at slaughter weight of 2 kg (Havenstein *et al.*, 2003).

Regardless of genetic improvements performed by breeders, broiler hybrids still differ with regard to their efficiency due to the specific selection practices (Emmerson, 1997). Hence the evaluation of promising crosses selected for high live weight, high weight gain, feed conversion, carcass traits and adaptation potential would highly contribute to the high efficiency of broiler chicken produce.

The aforementioned traits depend on numerous

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factors, including the genotype and the gender. Many researchers have reported a substantial effect of the genotype on live weight (Ojedapo *et al.*, 2008; Razuki *et al.*, 2011), feed conversion, carcass composition (Havenstein *et al.*, 2003; Santos *et al.*, 2004; Marcato *et al.*, 2006; Nikolova and Pavlovski, 2009), carcass weight (Rondelli *et al.*, 2003), and abdominal fat (Barbato, 1992; Fontana *et al.*, 1993).

A number of experiments have showed that the live body weight was also influenced by the gender (Sheuerman *et al.*, 2003; Musa *et al.*, 2006), feed intake and utilization (Smith *et al.*, 1998), abdominal fat content and the carcass composition. In a study of slaughter traits of five different turkey genotypes, Hristakieva *et al.* (2005) established a higher percentage of the grill from the live weight in females compared to male broilers. A number of studies demonstrated that female broilers have higher breast proportions, while in males the proportion of thighs was higher (Young *et al.*, 2001; Nikolova and Pavlovski, 2009; Abdullah *et al.*, 2010). In addition, Mendes *et al.* (2004) established lower abdominal fat percentage in males than in females.

The purpose of the present experiment was to compare the meat traits of two of the most popular broiler chicken hybrids Cobb 500 and Ross 308.

MATERIAL AND METHODS

The experiment was carried out in the nucleus poultry farm of the Institute of Agriculture - Stara Zagora between April and June 2012. Two broiler chicken genotypes were studied: Ross 308 and Cobb 500. For this purpose, 300 eggs of each genotype were set for incubation. The fertility rate, embryonic death rate, weight loss between incubation days 0 and 18, and the hatchability of set and fertilized eggs were determined. The number of chickens included in the study was 100 from each genotype (50 male and 50 female). The sexing

was done at one day age. Experimental birds were reared on wooden shavings bedded floor, with constant access to compound feed according to the age until 49 days of age.

The live body weight was determined individually by weighing birds at 1, 14, 28, 42 and 49 days of age. Feed conversion was calculated for each genotype and for the periods between 1-14, 14-28, 28-42 and 43-49 days of age on the basis of feed intake and weight gain.

By the end of the experiment, a slaughter analysis of three female and three male broilers with a live weight close to the group average was performed. After a 12-hour fasting the live body weight as well as the weight after grilling, weight of different carcass parts (breast, thighs, wings), weight of edible offal (heart, liver, gizzard) and weight of abdominal fats were determined. The slaughter yield and carcass ratios were calculated.

For integral assessment of broiler combinations, the European Poultry Efficiency Factor (EPEF) was calculated according to the formula:

$$\text{EPEF} = \frac{\text{live body weight (kg)} \times \text{livability (\%)} \times 100}{\text{fattening period (days)} \times \text{feed efficiency}}$$

Data were statistically processed according to the gender and genotype by ANOVA/MANOVA and LSD post hoc test using Statistica 8 software (StatSoft, 2009). Results were considered significant when $P < 0.05$. The percentages were arc sine transformed prior to the analysis.

RESULTS AND DISCUSSION

Table 1 presents the weight of incubated eggs, the loss in their weight for the first 18 days of incubation and the outcome of incubation. The weight of Cobb 500 incubation eggs was significantly higher than the average weight of Ross 308 eggs by 2.12 g.

Table 1: Incubation traits of eggs from Ross 308 and Cobb 500 genotypes

Genotype	Weight of incubation eggs (g)	Weight loss of incubation eggs % (days 0-18)	Fertility rate %	Embryonic death rate %	Hatchability %	
					from eggs set	from fertilized eggs
Ross 308	66.54 ± 0.34 ^b	15.91 ± 0.15 ^a	84.17 ± 0.84 ^a	8.42 ± 0.58 ^a	77.08 ± 1.25 ^a	91.58 ± 0.58 ^a
Cobb 500	68.66 ± 0.46 ^a	14.74 ± 0.36 ^b	86.36 ± 3.76 ^a	9.34 ± 1.91 ^a	78.18 ± 2.32 ^a	90.66 ± 1.91 ^a

^{a-b} – different letters within a column indicate statistically significant differences at $P < 0.05$

Table 2: Live body weight of broilers (g) depending on the genotype and the gender at different ages

Factors	Age, days				
	1 day	14 days	28 days	42 days	49 days
Genotype					
Ross 308	42.86 ± 0.39 ^b	381.66 ± 3.83 ^a	1115.95 ± 11.49 ^a	2019.60 ± 19.67 ^b	2435.29 ± 19.50 ^b
Cobb 500	44.96 ± 0.38 ^a	334.52 ± 3.89 ^b	1069.29 ± 9.42 ^b	2188.54 ± 24.63 ^a	2598.91 ± 24.76 ^a
Gender					
Male					
Ross 308	43.04 ± 0.48 ^b	384.21 ± 5.15 ^a	1119.96 ± 16.53 ^a	2052.89 ± 30.69 ^b	2462.31 ± 31.58 ^b
Cobb 500	45.87 ± 0.46 ^a	330.98 ± 4.93 ^b	1075.74 ± 13.25 ^b	2257.93 ± 33.78 ^a	2672.14 ± 34.43 ^a
Female					
Ross 308	42.68 ± 0.61 ^{ab}	379.10 ± 5.69 ^a	1111.94 ± 16.10 ^a	1986.30 ± 24.12 ^b	2412.39 ± 23.90 ^a
Cobb 500	44.03 ± 0.62 ^a	338.05 ± 6.36 ^b	1062.84 ± 12.68 ^b	2119.14 ± 28.95 ^a	2485.00 ± 23.70 ^a

^{a-b} – different letters within a column indicate statistically significant differences at $P < 0.05$

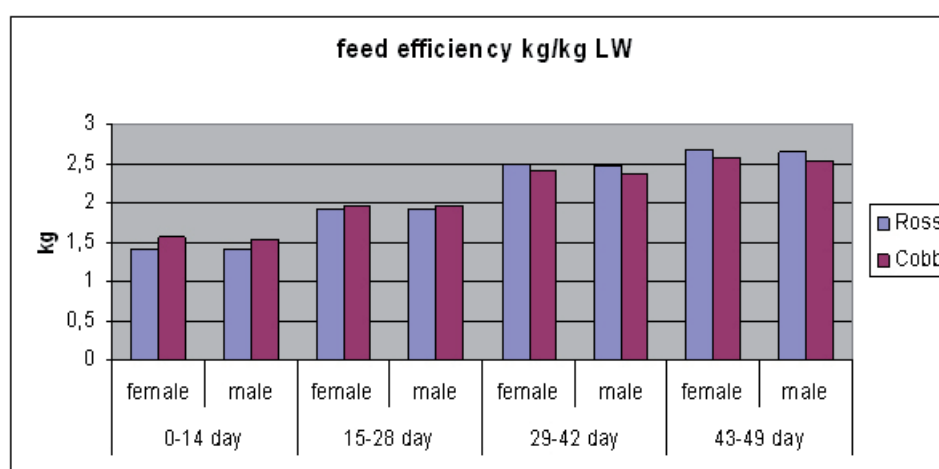
The relative loss of egg weight until the 18th incubation day was lower in Cobb 500 (14.74 %) than in Ross 308 (15.91 %). These data confirmed the earlier results of Tona *et al.* (2010) that the weight loss of Ross eggs was higher than that of Cobb eggs. The results did not demonstrate any statistically significant differences with regard to embryonic death rate, fertility and hatchability of set and fertilized eggs between studied genotypes.

Table 2 presents the data about live body weight of chickens at different ages depending on the genotype and the gender. The weight of hatchlings differed significantly between genotypes. Higher values were obtained for Cobb 500 broilers (44.96 g) than for Ross 308 (42.86 g). These differences could be associated

at the highest extent to differences in the weight of incubation eggs (Table 1).

Comparison of genotypes showed that Ross 308 hybrids were superior to Cobb 500 at the beginning of the fattening period despite the lower live weight at hatching of both genders which was preserved until the 28th day of age. Therefore, their weight gain rate until that age was more rapid. By the end of the experiment, Cobb 500 broilers attained higher average live weight, which was 6.29 % more than that of Ross 308.

This trend was also valid for male Cobb 500 broilers when the combined effects of genotype and gender were accounted for. Cobb 500 males were by 7.85 % heavier than Ross 308 males, whereas the differences between Cobb and Ross female broilers were

**Fig. 1: Feed efficiency**

statistically insignificant.

The dynamics in live body weight depending on the gender showed substantial differences between male and female broilers by the 42nd day of age by 6.15 % for Cobb 500 at 49 days of age. The differences between male and female Ross 308 broilers were 3.2 % at 42 days of age and even lower (2.02 %) by the end of the experiment.

Feed conversion between 1 and 14 days of age was more efficient (9-19 % higher) in male and female Ross 308 broilers than Cobb 500, whereas during the second period the values were comparable and the difference was very small (2 %) (Fig. 1).

Within the period from the 29th to the 42nd day

of age, there were no differences in feed conversion between genders. Ross 308 chickens exhibited a slightly lower feed conversion compared to Cobb 500. This trend was also present between the 43rd and the 49th day of age. Over the entire experimental period, the feed intake for 1 kg weight gain for Ross 308 and Cobb 500 birds was 2.178 kg and 2.181 kg, respectively.

For better evaluation of studied broiler combinations the European Poultry Efficiency Factor was calculated, which indicates the level of genetic potential utilization of a hybrid. The EPEF data (Table 3) demonstrated that Cobb 500 birds had higher values. i.e. a higher economic efficiency than Ross 308 by 14.87 points (6.18 %).

Table 3: European Poultry Efficiency Factor

Genotype	Body weight at 49 days of age, kg	Livability, %	Feed conversion (kg/kg)	EPEF	
				Absolute	Relative
Ross 308	2.435	99	2.178	225.89	93.82
Cobb 500	2.599	99	2.181	240.76	100

Table 4 presents the slaughter analysis results depending on the genotype and the gender. The differences in pre-slaughter weight reflected upon roasting and grilling weights. The slaughter analysis showed higher values of these traits in both genders of Cobb 500 broilers that are outlined with a higher growth potential (1810.67 g for roasting and 1710.50 g for grilling weight) in comparison to Ross 308 broiler chickens (1645 g for roasting and 1547.67 g for grilling weight).

In the present experiment, the difference between pre-slaughter live weight of male and female birds did not entail statistically significant changes in either grilling or roasting weights. Evidently, the body weight increase was due to body parts that do not participate in slaughter yield formation. A similar opinion was reported by Abdullah *et al.* (2010). In general, female broilers had higher breast weight and breast proportion from the grill than males, while the males had higher absolute and relative thigh weight. Our data were in agreement with those reported by Mendes *et al.* (2004), Santos *et al.* (2004) and Abdullah *et al.* (2010).

The comparison of breast and thigh weight with regard to the genotype once again showed the superiority of Cobb 500, which exhibited higher breast weight by 81 g and higher thigh weight by 25.33 g than Ross 308 broilers. The wings' weight was higher in Cobb 500

(188.33 g) than in Ross 308 (178.33 g).

Table 5 presents relative proportions of studied carcass traits. Roasting and grilling, presented as percentage of the live weight, attained statistically higher values in Cobb 500 broilers both with regard to the genotype and the gender. The roasting of male Cobb 500 was 1.41 % higher than that of male Ross 308. Similar trends were seen in female Cobb 500 which was superior to female Ross 308. The differences with regard to other traits between both genders of Cobb 500 and Ross 308 were insignificant. In general, female broilers had higher grilling percentage from the live, higher breast and weight and percentage weight, but lower thigh weight and percentage than males. Our results support those of Mendes *et al.* (2004), Santos *et al.* (2004), Hristakieva *et al.* (2005) and Abdullah *et al.* (2010).

CONCLUSION

The results of present experiment showed a number of differences in meat and slaughter traits between studied broiler chicken hybrids. Cobb 500 broilers had higher productive performance compared to Ross 308 under the same growing conditions.

The weight of hatchlings differed significantly according to the genotype. One-day-old Cobb 500

Table 4: Slaughter traits of broiler chickens depending on the genotype and the gender

Factors	Live weight, g	Roasting, g	Grilling, g	Breast, g	Thighs, g	Wings, g
Genotype						
Ross 308	2241.67 ± 20.72 ^b	1645.00 ± 16.45 ^b	1547.67 ± 17.73 ^b	406.67 ± 14.69 ^b	527.67 ± 12.46 ^b	178.33 ± 3.22 ^{ab}
Cobb 500	2411.67 ± 29.71 ^a	1810.67 ± 23.41 ^a	1710.50 ± 24.21 ^a	487.67 ± 23.45 ^a	553.00 ± 20.95 ^a	188.33 ± 4.38 ^a
Gender						
Male						
Ross 308	2276.67 ± 29.63 ^b	1654.00 ± 29.46 ^b	1556.00 ± 31.56 ^b	396.33 ± 23.05 ^{ab}	546.33 ± 20.54 ^{ab}	183.67 ± 1.76 ^{ab}
Cobb 500	2456.67 ± 42.56 ^a	1819.67 ± 47.81 ^a	1716.00 ± 52.14 ^a	438.67 ± 8.21 ^a	586.67 ± 32.19 ^a	192.33 ± 3.18 ^a
Female						
Ross 308	2206.67 ± 6.67 ^{abc}	1636.00 ± 20.11 ^b	1539.33 ± 22.48 ^b	417.00 ± 21.00 ^{bc}	509.00 ± 2.52 ^a	173.00 ± 4.51 ^a
Cobb 500	2366.67 ± 24.04 ^a	1801.67 ± 19.34 ^a	1705.00 ± 13.43 ^a	536.67 ± 16.76 ^a	519.33 ± 4.98 ^a	184.33 ± 8.35 ^a

^{a-b-c} – different letters within a column indicate statistically significant differences at $P < 0.05$

broilers were heavier than Ross 308 broilers. At the end of the experiment, Cobb 500 broilers attained a higher live weight, and were heavier than Ross 308 birds by 6.29 %. The feed intake per kg weight gain over the entire experimental period was 2.178 kg and 2.181 kg for Ross 308 and Cobb 500, respectively. Higher values of the European Poultry Efficiency Factor were established in Cobb 500 broilers, which were more economically efficient than Ross 308 by 14.87 points (6.18 %). The

performed slaughter analysis showed higher values of slaughter traits in Cobb 500, which had higher growth potential (roasting weight 1810.67 g and grilling weight 1710.50 g in Cobb 500 while 1645 g for roasting and 1547.67 g for grilling in Ross 308 broilers, respectively). In male Cobb 500 broiler chickens, the roasting percentage was 74.02 %, which was 1.41 % higher than that of Ross 308 males. The same trend were observed in female birds as well, i.e. superiority of Cobb 500 over Ross 308.

Table 5: Slaughter yield and slaughter traits (%)

Factors	Roasting, % of live weight	Grilling, % of live weight	Breast, % of grill	Thighs, % of grill	Wings, % of grill
Genotype					
Ross 308	73.39 ^b	69.04 ^a	26.27 ^b	34.09 ^b	11.40 ^a
Cobb 500	75.10 ^a	70.94 ^b	28.53 ^a	32.31 ^a	11.02 ^a
Male					
Ross 308	72.66 ^a	68.33 ^a	25.47 ^a	35.11 ^a	11.81 ^a
Cobb 500	74.05 ^b	69.83 ^b	25.58 ^a	34.15 ^a	11.23 ^a
Female					
Ross 308	74.14 ^a	69.75 ^a	27.07 ^b	33.08 ^b	10.98 ^a
Cobb 500	76.14 ^a	72.06 ^a	31.48 ^a	30.46 ^a	10.81 ^a

^{a-b} – different letters within a column indicate statistically significant differences at $P < 0.05$

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SIMULTANEOUS STUDY OF SOME OF MALE BREEDING SOUNDNESS INDICES AND SEXUAL URGE ON THE CROSSBREED RAMS

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ABSTRACT

The aim of this trial was investigation of the relationship between seminal parameters, sexual urge (SU) and some of body measures (BMs) in five ArkharMerino×Ghezel (AM×GH) and five Ghezel×Baluchi (GH×BL) rams during 5 month. The semen samples were evaluated for semen volume (SV), total sperm/ejaculate (TSE), spermatozoa concentration (SC), color, wave motion, spermatozoa progressive motility, percentage of live and abnormal spermatozoa, pH and metabolic activity of spermatozoa (MBRT). SU of the rams was measured by two indices including reaction time (RT) and refractory period (RP). BMs of the rams consisted of body weight (BWT), body length (BL), hip width (HW) and height at withers (HTW), which were recorded in monthly intervals. No significant differences were found between the two hybrid groups in any traits except for SU indices. RT only showed a significant correlation with SV and pH ($r = -0.14$ and $r = -0.17$, $P < 0.05$ respectively). RP showed a significant correlation with semen traits except for SV, TSE, pH, semen color. A significant correlation was revealed between the all BMs except for BWT with HTW. Semen quantity characteristics had a significant correlation with HTW, HW and BWT. RP showed a negative correlation with BMs. These results suggest that BMs can be used to predict the SU of the rams and also they will confirm the necessity of synchronized selection for the breeding soundness indices in the herd.

Key words: crossbreed ram body measurements; sexual urge; semen characteristics

INTRODUCTION

Reproduction is one of the most important factors for the economics of livestock production (Chenoweth, 1994; Makarechian *et al.*, 1985). Evaluation of reproductive ability of rams is an integral part of management programs of sheep flocks. The objective of a breeding soundness examination (BSE) in rams is to evaluate and classify their breeding ability. Hence, evaluation of male fertility prior to breeding is one of paramount factors to achieve breeding success (Ford *et al.*, 2009). The potential fertility of breeding males can be evaluated in the field by assessment of mating ability, testicular and physical examination and semen quality evaluation (Hoflack *et al.*, 2006). Semen evaluation has been used as an index of ram fertility especially in

those used in AI programs. Strongly sexual urge or libido of rams influences overall flock fertility (Matos and Thomas, 1991).

Differences in sexual behavior among rams have been recognized since long ago (Hafez, 1951) and positive associations between rams with high scores for sexual performance and ewe fertility have been reported (Mattner *et al.*, 1971; Perkins *et al.*, 1992). Study of relevance between fertility and quality of sexual desire can be useful for selection purposes and also for obtain an optimize fertility in the herd. Many studies indicated that sexual urge is an important factor affecting male fertility and there are some evidences that it is strongly influenced by genetic factors e.g. breed or genetic group (Ologun *et al.*, 1981; Chenoweth, 1983). Quirino *et al.* (2004) reported that direct selection for libido would

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be effective and it would lead to desirable correlated response in body weight, physical and morphological characteristics of spermatozoa and undesirable correlated response for scrotal circumference. In contrast, Galina *et al.*, (2007) showed that libido is neither related to semen quality nor to scrotal circumference, so that it is possible to obtain an excellent semen sample in bulls with low libido. This incoherence in results between the probers may be caused by various methods of libido testing (Landaeta-Hernandez *et al.*, 2002; Landaeta-Hernandez *et al.*, 2001; Bertram *et al.*, 2002). Therefore, there is a need for a standard libido testing in all breeding and commercial flocks to remove the rams with poor performance before than serving with female. Seminal physicochemical characteristics of these genetic groups have been well studied previously (Asadpour *et al.*, 2012a; Asadpour *et al.*, 2012b; Moghaddam *et al.*, 2012a; Moghaddam *et al.*, 2012b). However, there is low data of some aspects of the reproductive characteristics of ArkharMerino×Ghezel and Ghezel×Baluchi genetic group. The present study designed to determine the relationship between semen characteristics, sexual urge and body conformation traits. Therefore, the data from the crosses were studied according phenotypic correlation between these traits.

MATERIAL AND METHODS

Animals and management

This project was performed using 5 AM×GH and 5 GH×BL rams (3-5 years old) and via a female teaser (from Oct 2011 to Feb 2012). The males were trained to mounting and serving with anoestrus ewe with quiet temperament. The location for performing this study was in suburb of Tabriz, Iran (38° 02' N, 46° 27' E). During 15 days the rams were trained (in peak of breeding season) to semen collection by artificial vagina (AV) by the presence of the operator and in the mating pen (210 cm length, 60 cm width, 120 cm height). All the examinations were done by the same technician. The rams separated from the herd were housed in a large cover shelter with an open precinct in order to walk freely. All of the rams were kept under natural photoperiod. Any of rams were not able to seeing mounting and serving of other rams. Levels of nutrition remained equal and without changes as each ram's diet daily consisted of 20 % concentrate (75 % barley, 25 % corn, soya, bran, supplement and lime) and 80 % alfalfa hay. Also, all the rams had free access to salty stones and fresh water twice or three times a day. Hoof trimming, shearing, crutching, dipping, disease prevention and other general management were checked during the study.

Assessment of body measurements (BMs)

Height at wither (HTW) was measured vertically from thoracic vertebrae to the ground using a metal ruler. Body length (BL) was measured from the sternum to the aitch bone. Hip width (HW) was measured using a plastic measuring tape. BWT, BL, HW and HTW were recorded in monthly intervals.

Estimation of sexual urge (SU)

Two traits reaction time (RT) and refractory period (RP) were used for assessment of sexual urge (SU) of the rams. Simultaneous with semen collection the SU indices were evaluated at five-day intervals. The rams were reared under similar conditions from birth until the examination period. The testing of SU is based on the time taken by a particular ram to react to a sexual stimulus ewe. A camera was used for recording time to the SU indices. Each ram that did not mount the stimulus ewe within 5 minutes was considered inactive. The reactions are included by two criteria: a) Reaction time; measured as the amount of time between first contact with the teaser ewe and first false mount with the penis erected (Hoflack *et al.*, 2006). b) Refractory period; measured as the time taken between first ejaculate till the second false mount (Prado *et al.*, 2002). Each ram was allowed to mount with the stimulus ewe and following the time was recorded for the RT and then the RP.

Semen evaluation

Concurrent with video recording for the ram's sexual activity, the ram semen samples were collected. Ejaculates of rams were collected in the intervals of five days and it was constant throughout the study. Artificial vagina (AV) with internal temperature maintained at about 40 - 42°C was used for semen collection. Collecting glass was warmed at 37°C before the operation and it was maintained at this temperature until processed. A ewe with quiet temperament was used for mounting by the rams. Immediately after ejaculation the fresh semen samples were transferred to the laboratory (keeping out of direct sun light) and evaluated. SV (semen volume) was recorded using a graduated collecting glass (0.1cc accuracy). Semen pH was measured by the Pen form pH-meter (with 0.1 grades, model 8685, AZ Instrument, Taiwan). SC (spermatozoa concentration) was determined by use of a Thoma chamber following haemocytometer counter method. The fresh semen was diluted using 0.1 M sodium citrate dehydrate 2.9 % (pH = 6.7 - 6.9) plus one drop of formalin (1:400) at 400×magnification. TSE (total sperm/ejaculate) was then calculated (volume×density). Wave motion of fresh semen was evaluated (100 × magnification) according to Evans and Maxwell (1987). The assessment of the spermatozoa progressive motility was done using a visual scale from 0 to 100 % on the basis of suspended droplet slide and on a heated (37°C)

stage using phase-contrast optics ($\times 400$). Suspended droplet slide showed individual spermatozoa with more lucidity. For spermatozoa morphology and spermatozoa live/dead ratio, semen was stained with eosin-nigrosin stain and examined microscopically ($\times 400$). About 300 spermatozoa were counted from several parts of the slide. Metabolic activity of spermatozoa was measured using the Methylene Blue Reduction Time (MBRT). It was estimated by use of the method adopted by Herman and Madden (1953). Semen index was calculated according to Talebi *et al.*, (2009).

Statistical analysis

All statistical analyses were performed using the Statistical Analysis System (SAS, 1996). There were a few outliers on some of the traits (SV, SC, sperm abnormality, MBRT and SU). Therefore, to reduce the effect of sampling error, we have removed the outlier data. The Proc Mixed procedure of SAS was used for analysis of the repeated measurement data. The mean values were compared using a Tukey' test. Pearson correlation coefficient was calculated to evaluate the relationship between the traits. The mean values were considered to be statistically significant at $P \leq 0.05$.

RESULTS AND DISCUSSION

The minimum, maximum and mean \pm SE of seminal characteristics of AM \times GH and GH \times BL rams are presented in Table 1. AM \times GH rams showed best semen quality than the other genotype but it was not significant ($P > 0.05$). According to the descriptive statistics, AM \times GH genetic group had a higher time scores (equivalent with the lower libido) than GH \times BL rams in case of the SU traits (Table 2). The large range for all traits indicated the wide variation between individual rams. An inconspicuous and non-significant dominance of BMs ($P > 0.05$) was observed in the mean values of AM \times GH rams than GH \times BL genetic group (Table 2). The rams with high SU presented the highest live spermatozoa, motility, spermatozoa metabolic activity, SC and the fewest spermatozoa abnormalities. However, these relationships were ranged from - 0.13 to 0.24 and were not significant (Table 3). Small and negative correlation was observed between RT with SV and pH (varying from 0.14 - 0.17, $P < 0.05$). RP did not show a significant correlation with TSE, semen pH, color and volume ($P > 0.05$). Correlation coefficient between the SU indices (RT and RP) demonstrated that, the rams with fewer RT had a shorter RP ($r = 0.13$, $P = 0.04$). Thus, reaction time could be a factor for estimating refractory period of the rams. A highly significant correlation was revealed between BMs e.g. BWT and BL ($r = 0.54$), HW and HTW ($r = 0.835$) and HW with BL ($r = 0.49$), indicating high

level of association between these variables (Table 4). As it is shown in table 4 a high and significant correlation coefficients between HTW and HP vs. RP were observed ($r = - 0.47$). Data of semen evaluation as a determining factor for breeding soundness examination did not indicate any high and clear correlation with body sizes except for some of semen quantity traits e.g. TSE with HTW and HP ($r = 0.39$ and 0.31 respectively), SV with HTW and HW ($r = 0.36$ and 0.30 respectively) and also SC with BWT ($r = 0.29$, $P < 0.05$).

Many researchers emphasized that genetics plays an important role in determining sexual urge and it has a clear effect on sexual urge (libido) and inherent fertility differences between individual males (Ologun *et al.*, 1981; Chenoweth, 1997; Petherick, 2005). These studies show that in *Bos indicus* and *Bos taurus*, crossbred bulls generally exhibited higher libido scores in pen-tests than did their parental purebreds, providing further evidence of genetic influence on libido (Chenoweth and Osborne, 1965). Contrary to the results of Ford *et al.*, (2009) who did not observe significant difference between Boer and Kiko bucks in terms of SU indices ($P > 0.05$), in our work it was found that GH \times BL rams were better compared to the other genotype. The non-significant difference between the two genetic groups (in body weight and body length) was in agreement with results of Lavvaf *et al.*, (2012). In our study SU was found to be useful in semen quality estimating. These findings also coincide with the results of Quirino *et al.*, (2004) who used scoring system from 0 (no sexual interest) to 10 (two services followed by sexual interest, including mounts, mounting attempts or further services) for the assessment of sexual urge. Deen (2008) revealed that there is a high correlation between, copulation time and semen volume in camels ($r = 0.957$). The results of Wiggins *et al.*, (1953) showed exists a significant correlation between some of libido criteria (including number of ejaculates per trial, ejaculate time for first, second and third mating) and percentage of ewes lambing.

Wiggins *et al.*, (1953) reported that significant correlation was revealed between semen volume ($r = 0.062$, $P < 0.05$), estimated motility count ($r = 0.077$, $P < 0.01$), percentage of normal sperm ($r = 0.432$, $P < 0.01$), percentage of abnormal heads ($r = - 0.35$, $P < 0.01$) and percentage of ewes lambing. These findings indicated that the sexual urge indices are correlated with fertility and also the fertility parameters have a relatively correlation with some semen characteristics. This simultaneous trend between SU and physical semen characteristics in our study is in agreement with findings of Barkawi *et al.*, (2006). Anzar *et al.*, (1993) after study on 44 buffalo bulls reported that semen production was correlated with sexual behavior urge only in the fair and poor categories of buffalo bulls ($r = 0.84$, $P < 0.005$). Galal

Table 1: Range of seminal measurements of Ghezel×Baluchi and ArkharMerino×Ghezel rams

Genetic groups		AM × GH			GH × BL		
Semen parameters	N	Mean ± S.E.	Min	Max	Mean ± S.E.	Min	Max
Semen volume (ml)	145	1.12 ± 0.18	0.45	2.00	1.17 ± 0.25	0.48	2.20
Wave motion (0-5)	143	4.05 ± 0.18	2	5	3.82 ± 0.24	2	5
Progressive motility (%)	145	69.60 ± 4.21	50	90	67.75 ± 3.83	45	85
Semen color (0-5)	145	3.61 ± 0.41	2	5	3.55 ± 0.45	2	5
Total sperm output (×10 ⁹)	144	4.275 ± 0.73	1.654	19.55	4.616 ± 1.21	1.506	21.6
Sperm density (×10 ⁹)	145	3.623 ± 0.39	1.950	5.56	3.45 ± 0.44	1.85	5.42
Live sperm (%)	145	73.52 ± 3.42	50	90	72.91 ± 3.46	54	90
Abnormal sperm (%)	143	10.50 ± 1.53	4	28	11.30 ± 1.72	4	29
Semen index (×10 ⁹)	145	21133 ± 3923	4088	48876	20460 ± 3659	1040	64255
Semen pH	143	6.45 ± 0.27	5.8	7.1	6.69 ± 0.35	5.9	7.7
MBRT (s)	143	107.47 ± 7.07	55	190	119.16 ± 8.4	64	281

Means within each row within each factor without letters did not differ significantly from each other.

Table 2: Range of sexual behavior urge and body feature traits of Ghezel×Baluchi and ArkharMerino×Ghezel rams

Genetic groups		AM × GH			GH × BL		
SU parameters	N	Mean ± S.E.	Min	Max	Mean ± S.E.	Min	Max
Reaction Time (s)	144	24.45 ± 7.51 ^a	3	110	11.76 ± 7.02 ^b	2	48
Refractory Period (s)	144	234.47 ± 109.1 ^a	42	983	79.01 ± 108.5 ^b	20	305
Body Measurements	N	Mean ± S.E.	Min	Max	Mean ± S.E.	Min	Max
Body weight (kg)	50	69.79 ± 5.74	49.8	90	74.37 ± 5.74	53.2	92
Body length (cm)	50	77.78 ± 3.45	70	83	74.07 ± 3.73	68	80
Height at withers (cm)	50	71.30 ± 4.74	63.5	87	69.64 ± 4.72	64.5	77
Hip width (cm)	50	18.78 ± 1.93	15	25	18.00 ± 1.79	16	21

^{a,b} – significant difference at P<0.05

Table 3: Correlation coefficient (r) between sexual behaviour urge and seminal traits in ArkharMerino×Ghezel and Ghezel×Baluchi rams

r	SV	WM	PM	SL	SAB	MBRT	pH	TSE	Conc	Color	RT	RP
RP	-0.06	-0.24	-0.22	-0.20	0.22	0.19	-0.12	-0.02	-0.13	-0.11	0.13	1
P value	0.76	0.002	0.009	0.001	0.008	0.002	0.07	0.686	0.04	0.10	0.04	1
RT	-0.14	-0.02	-0.01	-0.004	0.01	0.005	-0.17	0.01	-0.05	-0.05	1	0.13
P value	0.03	0.72	0.85	0.94	0.83	0.96	0.01	0.84	0.44	0.47	1	0.04

SV = semen volume, WM = wave motion, PM = progressive motility, TSE = total sperm per ejaculate, Conc = sperm concentration, SL = Percentage of live spermatozoa, SAB = Percentage of abnormal spermatozoa, MBRT = methylene blue reduction time, RP = refractory period, RT = reaction time

et al., (1978) in their seasonal study on Merino, Ossimi and their crosses stated that relationship between semen quality and libido is not clear across breeding groups. It is not surprising that the findings on the relationship between measures of libido and fertility are inconspicuous, with some workers reporting positive correlations (Lunstra, 1984, 1986; Crichton and Lishman, 1988) and others, contradictory or negative (Christensen *et al.*, 1982; Boyd *et al.*, 1989; Bertram *et al.*, 2002; Holroyd *et al.*, 2002). The high and significant correlation among BMs and BWT will provide a valuable data for early selection of the crossbred rams in genetic improvement schemes. Due to the strong correlation between hip width and body length, these criteria (HP and BL) could be used for prediction of the ram body weight. These results are in agreement with results of Keith *et al.*, (2009). Maksimovic *et al.* (2012) in their study reported that body mass of three crossbred rams (Wurtemberg, Il-de-France and Pirot Pramenka) has a significant correlation with their body length ($r = 0.58$, $P < 0.01$). Also they stated that HTW did not have a significant correlation with the ram body mass. In the other study expressed that many Belgian Blue bulls with poor semen quality were failed in breeding soundness evaluations (Hoflack *et al.*, 2006). Hassan *et al.*, (2009) reported there are

a significant correlation between body weight and SV, SC and sperm motility ($r = 0.568$, 0.664 , 0.494 respectively). Fields *et al.*, (1979) reported a non-significant correlation between BWT with SV, sperm motility and SC and these results are in agreement with our work except for SC. Previously was also reported a positive correlation between sperm production and body condition score (Ikhatua and Olayiwole, 1982). Okere *et al.*, (2011) indicated that semen production is fairly independent of most body conformation traits. A positive correlation between hip width and height at withers with semen quantity characteristics ($r = 0.27$ to 0.39), indirectly indicate that the rams with bigger HW and HTW may have more semen output. Overall in the present study the correlations between seminal traits and body measurements were quite low. Unlike the results of Ford *et al.*, (2009), in our research SU scores and especially refractory period were correlated to the body size traits ($P < 0.05$). Refractory period could be defined as a period of time during which testis are incapable of repeating another ejaculation. Among two libido traits, RP showed more correlations with the other traits than RT and probably this trait of sexual urge (RP) could be an appropriate clue for male libido estimating. This discrepancy in the libido results of

Table 4: Correlation coefficient (r) between body conformation traits with seminal and sexual urge traits in the both genetic groups

Traits	r	Body weight	Height at withers	Hip width	Body length
Body conformation traits	Body weight	1	0.21	0.44**	0.54**
	Height at withers	0.21	1	0.835**	0.36**
	Hip width	0.44**	0.835**	1	0.49**
	Body length	0.54**	0.36**	0.49**	1
Sexual urge traits	Reaction time	- 0.40**	- 0.19	- 0.27*	- 0.22
	Refractory period	- 0.22*	- 0.47**	- 0.47**	- 0.37**
Seminal Traits	Semen volume	0.03	0.36**	0.30*	0.02
	Wave motion	0.22	0.20	0.18	0.002
	Progressive motility	0.17	0.21	0.21	0.01
	Live sperm	0.15	0.21	0.21	0.05
	Abnormal sperm	0.17	- 0.22	- 0.22	- 0.05
	MBRT	0.29*	- 0.22	- 0.22	0.05
	pH	0.16	- 0.08	- 0.13	- 0.03
	Total sperm/ejaculate	0.11	0.39**	0.31*	0.03
Sperm concentration	0.22*	0.27*	0.23	- 0.08	
Semen color	0.17	0.22	0.19	0.07	

** $P < 0.01$, * $P < 0.05$, ns Non-significant. MBRT: methylene blue reduction time.

different probers may be caused by various methods used for testing libido such as the latency (refractory period) for males to copulate, or reaction time (Chenoweth, 1981; Landaeta-Hernandez *et al.*, 2001), counts and durations of interest, such as sniffing at the vulva and time spent with females (Bertram *et al.*, 2002), the number of mounts and/or serves during a set period of time (Landaeta-Hernandez *et al.*, 2001; Bertram *et al.*, 2002) and scores assigned according to various combinations of these measures (Blockey, 1981; Chenoweth, 1981; Landaeta-Hernandez *et al.*, 2001). Therefore, there is a need for the development of a predictive standardized test for estimating sexual urge of males. Overall the interpretation and comparison of the results of these researches will be very difficult.

CONCLUSION

There is a paucity of data on breeding soundness evaluations in ArkharMerino×Ghezel and Ghezel×Baluchi rams. Therefore, this trial compared some of breeding soundness indices (BMs, semen evaluations), SU and their relationship with each other. Striking correlation between semen characteristics and RP in the crosses confirms the fact that probably this parameter of SU is an adequate index for libido testing. Nevertheless, ambiguities and inconsistency in results of the researchers made a commitment for numerous investigations in these fields. Generally, our results indicated that measurements of external body dimensions, body weight, sperm output characteristics and sexual urge can accurately guide the assessment of the reproductive performance of the crossbred rams.

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MASTITIS PATHOGENS AND THEIR RESISTANCE AGAINST ANTIMICROBIAL AGENTS IN DAIRY COWS IN NITRA, SLOVAKIA

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ABSTRACT

The objective of this study was to evaluate the effectiveness of different antibiotics against mastitis causing microorganisms in lactating dairy cows in and around Nitra region, Slovakia. Milk samples from quarters were cultured and bacteriologically evaluated. All the bacteria isolated through microbiological procedures were subjected to antimicrobial susceptibility test by disc diffusion method to a large number of antibiotics. The results revealed higher sensitivity against tetracycline (100 % of *Streptococcus agalactiae* and *uberis*, *Escherichia coli* (*E. coli*), Coagulase Negative Staphylococci (CNS)), (97.37 % of *Staphylococcus aureus*) with highest number of bacterial isolates, followed by enrofloxacin (100 % of *Strep. agalactiae* and *uberis*), (97.37 % *Staph. aureus*), (97.14 % of CNS), cefalexin + kanamycin (100 % of *Strep. agalactiae* and *uberis*), (97.14 % of CNS), (96.0 % of *E. coli*) and amoxicillin + clavulanat (100 % of *Strep. agalactiae* and *uberis*), (98.57 % of CNS), (94.74 % of *Staph. aureus*), (94.0 % of *E. coli*). Maximum resistance was observed against penicillin (96.0 % of *E. coli*) and streptomycin (66.67 % of *Strep. uberis*). In conclusion, *in vitro* antibiogram studies of bacterial isolates revealed higher sensitivity for tetracycline, enrofloxacin, a combination of cefalexin plus kanamycin and amoxicillin plus clavulanat acid.

Key words: dairy cows; mastitis; antimicrobial agents; bacterial strains; disc diffusion

INTRODUCTION

Mastitis is an inflammation of the mammary gland characterized by physical, chemical, bacteriological and cytological changes in milk. Pathological changes in glandular tissues of the udder and effects on the quality and quantity of milk have been observed (Amir, 2013). This disease is mainly caused by microorganisms usually bacteria, including gram-negative and gram-positive bacteria, mycoplasmas, yeasts and algae (Zadoks *et al.*, 2011).

The majority of mastitis incidences are caused by only a few common bacterial pathogens involved: *Staph. spp.* (*Staph. aureus* & *Staph. epidermidis*), *Strep. spp.* (*Strep. agalactiae*, *Strep. dysgalactiae*, *Strep. uberis* & *Strep. bovis*), coliforms (mainly *E. coli* & *Klebsiella pneumoniae*) and *Actinomyces pyogenes* (Sharma,

2010). Coagulase Negative Staphylococci (CNS) and *Corynebacterium bovis*, two other highly prevalent pathogens, are historically considered to be of limited importance and are therefore often described as minor pathogens. The impact of CNS is increasing (Pyörälä and Taponen, 2009), probably because prevalence of major pathogens are decreasing (Sampimon *et al.*, 2009).

The most effective procedures to control contagious mastitis pathogens can be obtained by using dry cow therapy, post milking teat disinfectants and effective pre-milking hygiene (Fox and Gay, 1993). The incidence of streptococcal mastitis has been greatly reduced by using antibiotics and improving herd hygiene, but the incidence of staphylococcal mastitis has increased greatly. Treatment of all quarters with antibiotics during drying off is very important (Sharif *et al.*, 2009). The majority of antibiotics used are broad-

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spectrum antibiotics acting against Gram-positive and Gram-negative bacteria (NCCLS, 2002). Control of environmental mastitis can be achieved by reducing the number of bacteria to which teat is exposed, increasing immune resistance of the cow, pre milking teat dipping with a germicidal. Animal environment should be as clean and dry as possible.

Antimicrobials are routinely used for treatment of dairy cattle affected with clinical and subclinical infections (Aarestrup, 2005). The use of antimicrobials have, over time, increased the number of antimicrobial-resistant microbes globally, and any use of these agents will to some extent benefit the development of resistant strains and also inappropriate usage of antimicrobials such as wrong dose, drug or duration may contribute the most to the increase in antimicrobial resistance without improving the outcome of treatment (Williams, 2000).

In recent years, antimicrobial susceptibility testing has become under scrutiny because of concerns about antimicrobial resistance, changes in methodology and the relationship between *in vitro* results and on-farm clinical outcomes. Susceptibility tests of milk samples submitted to state diagnostic laboratories that use the disk-diffusion method have demonstrated remarkable agreement but vary from results of a small survey processed using broth dilution (Constable and Morin, 2003).

Our recent study also dealt with the frequency

of distribution of pathogens in positive milk samples (Idriss *et al.*, 2013). The present work aimed to study the effectiveness of different antibiotics against isolated microorganisms.

MATERIAL AND METHODS

The study was conducted during the period from 2010-2012 in and surroundings of Nitra region in Slovakia. A total of 390 milk samples were collected from udder quarters of dairy cows at some different small holder dairy farms, and pathogenic bacteria were examined and sensitivity of microorganisms against antibiotics had been tested.

Milk sample collection and laboratory analysis

After a quarter had been cleaned up by removing any possible dirt and washed with tap water, the teat end was dried and swabbed with cotton soaked in 70 % ethylalcohol. Approximately 100 ml of milk was collected aseptically into sterile bottles, after discarding the first 3 milking streams. Milk samples from each quarter were transported to the Animal Production Research Center Laboratory in an ice cooled box at 4°C and analysed immediately (max. 4 h after collection) either for identification of the clinical mastitis

Table 1: Results of microbiological culture of milk samples collected from mastitis cows in Nitra region

Isolated microorganisms	Total. No.	%
<i>Staphylococcus aureus</i>	38	9.74
<i>Streptococcus agalactiae</i>	6	1.54
<i>Streptococcus uberis</i>	16	4.10
<i>E. coli</i>	50	12.82
<i>Enterococcus</i> spp.	12	3.08
<i>Bacillus</i> spp.	25	6.41
<i>Corynebacterium pyogenes</i>	5	1.28
CNS	70	17.95
<i>Pseudomonas aeruginosa</i>	13	3.33
<i>Staphylococcus epidermidis</i>	14	3.59
<i>Staphylococcus chromogenes</i>	4	1.03
Yeasts	22	5.64
Others (bacteria and mould)	13	3.33
infected quarters	288	73.85
non-infected quarters	102	26.15
Total dairy cows in herd	390	100

T. no- Total number of isolate, %- percentage of bacteria, T.no. , CNS- Coagulase Negative Staphylococci.

pathogen or to determine the reason for an increased somatic cell count (SCC). The milk samples were investigated for pathogenic mastitis in accordance with a standard procedure (IDF, 1981).

Antimicrobial susceptibility test

All the bacteria isolated through microbiological procedures were subjected to antimicrobial susceptibility test by disc diffusion method to identify the most effective drugs for mastitis treatment in the study area (Hameed, 2008). The sensitivity against amoxicillin, amoxicillin + clavulanat acid, cefalexin + kanamycin, ceftiofur, cloxacillin, enrofloxacin, lincomycin, nafpenzal, neomycin, penicillin, rifaximin, streptomycin and tetradelta were determined on Mueller Hinton agar as described by National Committee for Clinical Laboratory Standards (NCCLS, 2002). The results were obtained by measuring the diameter of the growth inhibition zone around the antibiotic disc for each isolated bacterial strain and recorded as sensitive, intermediate and resistant.

Statistics: Statistical evaluation of data was done by Excel program.

RESULTS AND DISCUSSION

From our previous study a total of 390 milk samples were investigated, 288 (73.85 %) samples were positive. No pathogens were isolated from 102 (26.15 %) milk samples as given in Table 1 (Idriss *et al.*, 2013).

The study of the frequency of susceptibility of *Staph. aureus* (n = 38) to antibiotics has revealed a higher sensitivity to the enrofloxacin, tetradelta (97.37 % to each), combinations of amoxicillin plus clavant acid and cefalexin plus kanamycin (94.74 % to each) and rifaximin (94.74 %). A certain resistance has been noted to amoxicillin and streptomycin (18.42 % to each), lincomycin (13.16 %) and penicillin (10.53 %). More number of isolates showed moderate sensitivity or resistance to streptomycin (10.53 %), amoxicillin and penicillin (2.63 % to each) (Table 2).

Staphylococci were mostly susceptible to antimicrobials tested but, Muhamed *et al.* (2012) found that *Staph. aureus* was resistant to penicillin and streptomycin (41.44 % and 25.65 % respectively). Similar results were obtained by Sumathi *et al.* (2008) where *Staphylococcus* and *Streptococcus* spp. were resistant to streptomycin and penicillin. Those results are in accordance with our findings.

In contrast, CNS (n = 70) have been found to show a complete sensitivity to the rifaximin and tetradelta (100 % to each), and higher sensitivity to amoxicillin combination plus clavulanat acid (98.57 %), cefalexin plus kanamycin, ceftiofur, cloxacillin, enrofloxacin, lincomycin, nafpenzal (97.14 % to each). Apart from these unexpected results of CNS strain sensitivity for all antibiotic except to streptomycin (14.29 %), penicillin and amoxicillin (5.71 % to each), some strains showed intermediate sensitivity or resistance to amoxicillin and penicillin (7.14 % to each). Whereas the antibiogram

Table 2: Frequency of susceptibility of *Staphylococcus aureus* (n = 38) and Coagulase negative staphylococci (CNS) (n = 70) to antibiotics

Bacterial strains Antibiotic agent	<i>Staphylococcus aureus</i> (n = 38)			CNS (n = 70)		
	S %	IM %	R %	S %	IM %	R %
Amoxicillin	78.95	2.63	18.42	87.14	7.14	5.71
Amoxicillin + clavulanat	94.74	0.00	5.26	98.57	0.00	1.43
Cephalexin + kanamycin	94.74	0.00	5.26	97.14	1.43	1.43
Ceftiofur	94.74	0.00	5.26	97.14	0.00	2.86
Cloxacillin	92.11	0.00	7.89	97.14	0.00	2.86
Enrofloxacin	97.37	0.00	2.63	97.14	0.00	2.86
Lincomycin	86.84	0.00	13.16	97.14	0.00	2.86
Nafpenzal	94.74	0.00	5.26	97.14	0.00	2.86
Penicillin	86.84	2.63	10.53	87.14	7.14	5.71
Rifaximin	94.74	0.00	5.26	100.00	0.00	0.00
Streptomycin	71.05	10.53	18.42	85.71	0.00	14.29
Tetradelta	97.37	0.00	2.63	100.00	0.00	0.00

CNS- Coagulase negative staphylococci, n- number of bacteria strains, S- Sensitivity, IM- Intermediate, R- Resistant.

test to various antibiotics revealed that the isolates of CNS was resistant to streptomycin (14.29 %), followed by amoxicillin and penicillin were (5.71 % to each) (Table 2).

In the present study *Staph. aureus* was resistant to amoxicillin, streptomycin, lincomycin and penicillin and CNS was resistant to streptomycin, penicillin and amoxicillin, which is consistent with previous findings (Bengtsson *et al.*, 2009).

It is interesting to note that the present study has revealed a complete susceptibility (100 %) of *Strep. agalactiae* and *Strep. uberis* to all antibiotics, except *Strep. agalactiae* was resistant to lincomycin (16.67 %) and streptomycin (33.33 %), and *Strep. uberis* to cloxacillin (20 %) and streptomycin (66.67 %) (Table 3).

In our study we have found that all *Strep. agalactiae* and *Strep. uberis* were susceptible to a lot of antibiotics. In contrast, Erskine *et al.* (2002) and Makovec and Ruegg (2003) have found congruent results that *Staph.* other than *Staph. aureus* were sensitive to penicillin, ceftiofur and cephalothin and *Staph. aureus* was sensitive to ceftiofur and cephalothin and resistant to penicillin.

Vasil' (2009) tested 14, 52 and 30 strains of *Strep. agalactiae*, *Strep. uberis* and CNS and has found that *Strep. agalactiae* strains were sensitive to all antibiotics except to neomycin, streptomycin, while *Strep. uberis* was a complete sensitive to a combination of amoxicillin + clavulanat and ampicillin, followed by cefalotin, lincomycin, whilst it is resistant to streptomycin,

novobiocin and neomycin and CNS was sensitive to a combination of amoxicillin + clavulanat and resistant to streptomycin and penicillin. These results are in accordance with our findings that CNS, *Strep. agalactiae*, *Strep. uberis* and *E. coli* were completely sensitive (100 %) to tetracycline, while *Staph. aureus* showed sensitivity of 97.37 %. *Strep. agalactiae*, *Strep. uberis* and *E. coli* were complete sensitive (100 %) to enrofloxacin, followed by *Staph. aureus* and CNS (97.37 %) and (97.14 %), respectively. *Strep. agalactiae* was (100 %) sensitive to cefalexin + kanamycin, followed by CNS, *E. coli* and *Staph. aureus* (97.14 %), (96.0 %) and (94.74 %), respectively. *Strep. agalactiae* was (100 %) sensitive to amoxicillin + clavulanat, followed by CNS, *Staph. aureus* and *E. coli* (98.57 %), (94.74 %) and (94.0 %), respectively.

The percentage of susceptibility of *E. coli* (n = 50) isolates, revealed complete sensitivity to ceftiofur, enrofloxacin and tetracycline (100 %) isolates, followed by a combination of amoxicillin plus clavulanic acid and neomycin (96 % to each). A highly resistance has been noted to cloxacillin (98 %), lincomycin and penicillin with (96 % to each) and amoxicillin (82 %). Among the *E. coli* isolates, intermediate susceptibility was observed with streptomycin (6 %) and combinations of amoxicillin plus clavulanic acid (4 %) (Table 4).

Results of the current study demonstrated that *E. coli* was resistant to amoxicillin and penicillin. Similar result was obtained by Onerba (2006) who reported that *E. coli* was resistant to amoxicillin (85 %).

Table 3: Frequency of susceptibility of *Streptococcus agalactiae* (n = 6) and *Streptococcus uberis* (n = 15) to antibiotics

Bacterial strains	<i>Streptococcus agalactiae</i> (n = 6)			<i>Streptococcus uberis</i> (n = 15)		
	S %	IM %	R %	S %	IM %	R %
Amoxicillin	100	0	0	100	0	0
Amoxicillin + clavulanat	100	0	0	100	0	0
Cephalexin + kanamycin	100	0	0	100	0	0
Ceftiofur	100	0	0	100	0	0
Cloxacillin	100	0	0	80	0	20.00
Enrofloxacin	100	0	0	100	0	0
Lincomycin	83.33	0	16.67	100	0	0
Nafpenzal	100	0	0	100	0	0
Penicillin	100	0	0	100	0	0
Rifaximin	50	0	50	100	0	0
Streptomycin	66.67	0	33.33	33.33	0	66.67
Tetracycline	100	0.0	0	100	0	0

S- Sensitivity, IM- Intermediate, R- Resistant, n- number of bacteria strains

Table 4: Frequency of susceptibility of *Escherichia coli* (n = 50) to antibiotics

Bacterial strains Name of antibiotic	<i>Escherichia coli</i> (n = 50)		
	S %	IM %	R %
Amoxicillin	18.00	0.00	82.00
Amoxicillin + clavulanat	94.00	4.00	2.00
Cephalexin + kanamycin	96.00	2.00	2.00
Ceftiofur	100.00	0.00	0.00
Cloxacillin	2.00	0.00	98.00
Enrofloxacin	100.00	0.00	0.00
Lincomycin	4.00	0.00	96.00
Nafpenzal	90.00	0.00	10.00
Neomycin	96.00	0.00	4.00
Penicillin	4.00	0.00	96.00
Rifaximin	62.00	0.00	38.00
Streptomycin	84.00	6.00	10.00
Tetradelta	100.00	0.00	0.00

S- Sensitivity, IM- Intermediate, R- Resistant, n- number of bacteria strains

Foltys and Kirchnerová (2005) tested 60, 62 and 77 strains of *Staph. aureus*, *Strep. agalactiae* and *E. coli*, respectively to various antibiotics and they reported that *Staph. aureus* was sensitive to all antibiotics except lincomycin and streptomycin, whilst *Strep. agalactiae* was 100 % sensitive to amoxicillin and ampicillin and resistant to streptomycin, neomycin and tetracycline and *E. coli* was resistant to all antibiotics. These findings are in complete accordance with the results of the present study except *E. coli* which was sensitive to ceftiofur and enrofloxacin (100 % to each of them) and to neomycin (96.0 %).

CONCLUSION

Antibiotic susceptibility tests should be done to determine the effectiveness of drug that can be used for successful treatment of diseases. Proper isolation and identification of the causative organism play significant role in prevention and control of the diseases. In our study a combinations of amoxicillin plus clavulanat acid, cefalexin plus kanamycin, enrofloxacin and tetradelta were the most effective antibiotics for control of bovine mastitis in Nitra area. Thus, there is a need to routinely investigate and record the epidemiology of bovine mastitis and antibiogram sensitivity of bacterial isolates in various parts of Slovakia.

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DETERMINANTS OF ECONOMIC EFFICIENCY IN DAIRY CATTLE AND SHEEP

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ABSTRACT

The objective of this study was to analyze the determinants of economic efficiency in milk production and milk and lambs production on dairy cattle and sheep farms, respectively. Economic efficiency was evaluated by the synthetic indicator of the total profit to cost ratio and by the individual indicator of the profit to individual costs-items for the database of farmers of the Animal Production Research Centre Nitra for the period 2006 to 2012. Economic efficiency with and without direct subsidies was expressed per kg of milk in dairy cattle and per ewe and year in dairy sheep. The average value of profit to cost ratio was - 9 % and - 48 % for cattle and sheep farms, respectively. Costs of feeds, depreciations and other direct costs were of higher proportion on the total costs in cattle and sheep. The profit to cost ratio on these costs items was the lowest. On the contrary, proportion of profit per unit of costs for repairs and services, management of overhead costs and for other direct material costs was higher in dairy and sheep analysed farms. Economic efficiency of milk production calculated in 2007 and 2008 for cattle farms was positively determined by lower value of costs per milk unit along with increase in milk price. The sharp fall in milk price, reduction in the number of cows per herd and savings in the feeds consumption resulted in the lower economic efficiency of milk production in period 2009 - 2012. In sheep farms, positive impact of demand for dairy products on the sheep milk price over the whole time period was found. Contrary, price of lambs remained on its low value. Size of flock and milk yield increased in the consequence. In spite of these facts and of reduction in some inputs, it was not sufficient for profitability in sheep. Level of animal performance, market price of dairy cattle and sheep commodities, input prices (feed, labour, other direct costs and depreciations) along with the value and scheme of subsidies were found as the most important determinants of economic efficiency in dairy cattle and sheep farms.

Key words: economic efficiency; profit to cost ratio; ruminants; milk; lambs

INTRODUCTION

Economy of animal production is closely associated with the biological efficiency of breeding. It is generally understood as the company's ability to change the material inputs (expressed as costs) into the marketable product under the common production conditions (Samuelson and Nordhaus, 1992; Tess and Davis, 2002; Gunlu *et al.*, 2003). Some of the biological aspects of the animal production efficiency were summarized previously (Tess and Davis, 2002; Krupová *et al.*, 2012). Profit to cost ratio is usually used as the

indicator of the economic efficiency (Foltýn *et al.*, 2010). Many papers dealing with the analyses of profitability using these parameters in dairy cattle (e.g. Ubrežiová and Mihina, 1995, 1998; Chrastinová *et al.*, 2011;) and in sheep (Jávor *et al.*, 2005; Vláčil, 2005; Benoit and Laignel, 2011) have been published till now. To the best of our knowledge, neither the value of profit to cost ratio for individual cost items defined in the calculation formula nor the detailed analysis of the development of base macro and microeconomic factors (determinants) have been evaluated until now for dairy cattle and sheep. The objective of this study was to analyze the economic

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efficiency and to identify key factors (determinants) of dairy cattle and sheep in Slovakia for the period 2006 to 2012.

MATERIAL AND METHODS

Data description

The economic efficiency of milk in cattle and of milk and lamb in sheep was evaluated in period 2006 to 2012. In total, data from 141 dairy cattle and 51 dairy sheep farms recorded in the database of Animal Production Research Centre (APRC) Nitra were analysed. These farms were chosen randomly to represent the individual production regions and breeds of dairy cattle (Holstein, Slovak dual purpose cattle - Simmental and Slovak Pinzgau and crosses) and sheep (Improved Valachian and Tsigai) in Slovakia. The basic production characteristics of dairy cattle and sheep farms for the period 2006 to 2012 are summarised in Table 1. For dairy cattle farms a classical indoor production system was typical with the cows in a free housing system. Integrated intensive indoor fattening of surplus male progeny and selling of the surplus pregnant breeding heifers was practised. Age at first calving reached 940 days and the average number of lactations finished per cow was 3.0 during the evaluated period. For analyzed dairy cattle herds as well

as for all dairy cattle herds in Slovakia the continuous milk production during the year was typical. Dairy sheep flocks were kept mainly in semi-extensive (so-called Carpathian) production system. Farming of domestic multi-purpose breeds (Improved Valachian and Tsigai) was characterised by a seasonal lambing in winter and by pasture grazing during the summer. Lambs were weaned and sold before Easter at the average age of 50 days. After weaning of lambs, ewes were milked until the end of the breeding season (autumn). Natural mating was used only. Ewes gave birth to lambs for the first time at 2 years of age and average length of productive life of ewes was 3.85 years in the widespread production system.

Basic economic indicator

The base indicator of economic efficiency (profit or loss) in dairy cattle and sheep for period 2006 to 2012 was calculated as the difference between total revenues and total cost per animal products with and without including of direct subsidies¹. Profit or loss was defined in € per kg of milk and in € per ewe and year in cattle and sheep farms, respectively. Total costs in cattle and sheep were quantified by a countdown calculation method

¹Payment per livestock unit (2007-2012), additional national direct payment per dairy cow (2010-2012) and support per dairy cow - help in milk crisis (2010). For more details see Krupová *et al.* (2013).

Table 1: Basic production indicators in analysed dairy cattle and sheep farms from 2006 to 2012

Indicator	2006	2007	2008	2009	2010	2011	2012	Mean value	v (%) ^a
Dairy cattle									
Number of cows in herd	308	334	328	312	350	314	296	320	6
Losses of cows (%)	7	6	5	5	6	5	6	6	13
Milk yield in kg per FD	14.55	16.36	17.27	16.47	15.31	16.16	17.19	16.19	6
Fertility (%)	89	90	90	89	87	97	98	91	5
Calving interval (days)	491	410	431	433	431	418	421	434	6
Age at first calving (days)	1035	1018	919	921	906	885	899	940	6
Dairy sheep									
Number of ewes in flock	349	366	512	482	436	452	437	433	13
Losses of ewes (%)	12	6	10	13	7	9	11	10	24
Milk yield (kg per ewe and year)	61.78	73.79	65.17	45.58	70.82	66.04	58.77	63.14	14
Lambs born per ewe ^b	1.15	1.24	1.04	1.11	1.08	1.39	1.18	1.17	9
Lambs sold per ewe and year	0.67	0.71	0.59	0.52	0.68	0.86	0.73	0.68	15
Weaning weight of lambs (kg per lamb)	12.54	12.60	11.78	11.72	12.14	12.01	10.41	11.89	6
Wool production (kg per ewe)	2.50	2.70	3.20	2.90	3.51	3.04	3.39	3.03	11

Source: economic database of APRC Nitra, own calculations

^aCoefficient of variation

^bParameter is influenced by the average litter size and proportion of ewes which give birth to lambs in the flock per year

when by-product value (manure and live born calf in cattle and manure, wool and live born lambs in sheep, respectively) were eliminated from the direct and indirect costs (Krupová *et al.*, 2012).

In cattle, total revenues based on the market price per kg of milk and total costs per kg of milk were defined (Table 2). In sheep, milk and lambs were the two main products of the farming. Therefore, milk yield per milking period and number of lambs sold per ewe and year and market price per milk unit and per lamb (Table 1 and 3) were considered when calculating the total revenues. Regarding the direct subsidies, value of the subsidies per kg of milk in dairy cattle was based on the sum of all direct subsidies (payments per livestock unit

and per dairy cow) and the amount of milk produced by the individual farmers during the evaluated years. Contrary in dairy sheep farms, direct payments per livestock unit (ewe = 0.15 livestock unit) were only provided for farmers. In 2006, subsidies were not taken into account due to the absence of direct payments to dairy farmers (MA SR, 2013). Other subsidies (e.g. LFA, SAPS) were not considered to analyse the direct impact of costs, market prices and animal performance on the economic efficiency in the evaluated period. The average exchange rate of 30.126 Slovak Crowns (SKK) per Euro was used in the calculations for the period from 2006 to 2008. For more details see Table 2 and 3 where basic economic indicators of dairy cattle and sheep farms

Table 2: Basic economic indicators of milk production in analysed cattle farms from 2006 to 2012 (in € per feeding day (FD), in € per kg of milk, respectively) and average proportion of individual costs items on the costs (%)

Indicator	2006	2007	2008	2009	2010	2011	2012	Mean value	v (%) ^a	Cost proportion (%)
Labour costs	0.370	0.459	0.404	0.399	0.548	0.531	0.550	0.466	17	8
Own feed	1.358	1.687	2.179	1.819	0.984	2.316	1.954	1.757	26	29
Purchased feed	0.430	0.635	0.717	0.513	1.025	0.600	0.814	0.676	29	11
Other material costs ^b	0.215	0.209	0.236	0.193	0.212	0.352	0.310	0.247	24	4
Repairs and services	0.079	0.110	0.079	0.081	0.095	0.058	0.086	0.084	19	1
Depreciation of tangible property	0.309	0.276	0.363	0.405	0.423	0.354	0.663	0.399	32	7
Depreciation of basic stock	0.640	0.647	0.599	0.609	0.746	0.900	0.737	0.697	15	11
Other direct primary costs ^c	0.457	0.614	0.541	0.526	0.573	0.690	0.729	0.590	16	10
Other direct secondary costs ^d	0.458	0.538	0.570	0.534	0.639	0.802	0.726	0.609	20	10
Production overhead	0.238	0.265	0.307	0.243	0.386	0.461	0.469	0.338	30	6
Management overhead	0.172	0.227	0.201	0.191	0.145	0.404	0.431	0.253	46	4
Costs together	4.727	5.669	6.196	5.511	5.775	7.467	7.470	6.116	17	100
By-product ^e	0.273	0.268	0.273	0.273	0.274	0.281	0.289	0.276	2	-
Total costs per FD	4.454	5.401	5.923	5.239	5.502	7.186	7.181	5.841	17	-
Total costs per kg of milk	0.306	0.330	0.343	0.315	0.359	0.445	0.418	0.359	15	-
Subsidies in € per kg of milk ^f	0	0.009	0.006	0.030	0.034	0.042	0.015	0.019	75	-
Market price per milk without subsidies	0.321	0.348	0.348	0.252	0.284	0.331	0.307	0.313	11	-
with subsidies	-	0.356	0.355	0.282	0.318	0.373	0.323	0.334	10	-
Profit or loss per milk without subsidies	0.015	0.017	0.006	-0.063	-0.065	-0.114	-0.110	-0.045	-128	-
with subsidies	-	0.026	0.012	-0.033	-0.042	-0.072	-0.095	-0.034	-138	-

Source: economic database of APRC Nitra, own calculations

^aCoefficient of variation

^bPurchased medicines, disinfectants, other material used in the office

^cInclude breeding and veterinary treatments, energy, social costs and other services

^dInclude own trucking and other own services

^eValue of manure (0.036 t of manure per FD * 3.65 € per t) and calf born alive (35 kg * 1.66 € per kg of live weight * average number of calves) per FD of cow

^fSum of all direct subsidies (payments per livestock unit and per dairy cow) per milk unit. For more details see section "Material and Methods"

for the analyzed period are given.

Profit to cost ratio

Detailed analysis of economic efficiency in cattle and sheep was based on the synthetic indicator of profit to cost ratio and on the individual indicators of profit to cost ratio. The synthetic parameter of profit to cost ratio including the direct subsidies (PCR) of milk production in cattle was measured as follows:

$$PCR = \frac{\text{profit}}{\text{total costs}}$$

and the synthetic parameter of profit to cost ratio without direct subsidies (PCR_2) was calculated as:

$$PCR_2 = \frac{\text{profit} - S}{\text{total costs}}$$

where: *profit* is profit or loss in milk production (€ per kg) with including direct subsidies (*S*) and costs are total costs per kg of milk (Chrastinová *et al.*, 2009, 2011; Foltýn *et al.*, 2010). In dairy sheep farms, the synthetic parameter of profit to cost ratio with and without direct subsidies (PCR and PCR_2) was calculated as defined before, where *profit* was profit or loss in € per ewe and year with including direct subsidies (*S*) and costs were total costs per ewe and year.

The same algorithm was used for calculation the individual indicators of profit to cost ratio. The only difference being that the values of individual cost items of the calculation formula were used. Absolute values of profit to cost ratio were applied to compare the significance of individual costs items given in the calculation formula over the analyzed period and to objectify proportion of the profit or loss on the individual cost items.

RESULTS AND DISCUSSION

Basic economic indicator

Basic economic indicators in cattle farms over the analyzed period (Table 2) showed that the profit in milk production was only achieved in the year 2007 and 2008. Market price of milk and milk yield in dairy cattle were higher (by 0.041 € per 1 kg milk on average and by 0.88 kg milk per feeding day, respectively) compared the rest of the studied period. In addition, the lower level of costs per feeding day (FD) in dairy cattle (by 0.250 € per FD) were achieved in the mentioned years. Due to combination of these factors the profit in milk production was achieved. It is very important to note that the higher value of loss was achieved in the years 2011 and 2012. It was related to the higher costs per FD (+ 35 %) compared to the rest of the mentioned years. The value of unit costs

in milk production increased mainly due to the higher feed prices and the cancellation of tax benefits for fuel (2011) which were implicated in the agriculture sector in previous period.

Compared to dairy cattle herds, economic efficiency in sheep farms was influenced by two products. Therefore combination of production and economic parameters of the individual sheep commodities on the economic efficiency should be considered. In dairy sheep farms negative efficiency (loss) was found over the whole time period (Table 3). However, the loss value was not constant. At the beginning of the evaluated period, the loss per ewe deepened and reached the bottom in 2009 (- 99 € without subsidies and - 77 € with subsidies). In the next three years, positive impact of milk yield (+ 20 kg per ewe and year), number of lambs sold per ewe (+ 0.24) and market price of lambs (+ 5 €) was found. Compared to 2009, total revenue per ewe and year finally increased by 18 € on average in these years but it was still not sufficient for profit. Considering the whole time period, increase of costs value (+ 41 %) compared to revenues (+ 4 %) probably plaid a role in sheep farms. Moreover, mentioned disproportion was not absorbed by subsidies, especially if its value declined in the last three years (Table 3).

Profit to cost ratio - synthetic indicator

Synthetic indicator of profit to cost ratio (profitability) of milk production in cattle (Figure 1) ranged within the interval from - 26 % (without subsidies in 2010 and 2011) to + 8 % (with subsidies in 2007) during the analyzed period (Figure 1). This range is in accordance with the results of Chrastinová *et al.* (2009) and Foltýn *et al.* (2010). In our study, the negative value of profit to cost ratio in milk production (with and without direct subsidies) was found in the years from 2009 to 2012. The average market price of milk dropped down (by 0.033 € per 1 kg milk on average) during this period. The lower value of revenues was not compensated even the higher value of subsidies (+ 0.025 €) per 1 kg of milk (Table 2). Profit to costs ratio in analysed dairy cattle herds reduced in the individual year by 5 p.p. (percentual point) after adding of subsidies (Figure 1).

Wider range of interval for profit to cost ratio (from - 42 % without direct subsidies to 22 % with direct subsidies) was noted by Ubrežiová and Mihina (1995, 1998) and Chrastinová *et al.* (2011). It was mainly due to the higher variability of production and economic indicators of the herds they evaluated. For example, the milk yield varied from 7.56 kg to 16.68 kg per FD and unit costs from 0.270 € to 0.380 € per 1 kg of milk. The system of regulation within the economic reform practised in the nineties of the past century was an important factor for these results. Appropriate values of these indicators valid for dairy cattle farms of APRC are

Table 3: Basic economic indicators of milk and lamb production in analysed sheep farms from 2006 to 2012 (in € per ewe and year) and average proportion of individual costs items on the costs (%)

Indicator	2006	2007	2008	2009	2010	2011	2012	Mean value	v (%) ^a	Cost proportion (%)
Labour costs	16.59	21.86	31.32	33.64	40.40	37.81	34.49	30.87	26	18
Own feed	36.48	45.91	45.95	48.11	44.11	34.91	32.76	41.18	14	24
Purchased feed	4.30	14.52	9.08	3.61	2.05	6.19	10.82	7.22	57	4
Other material costs ^b	5.60	1.91	7.02	2.05	4.66	5.67	6.27	4.74	39	3
Repairs and services	1.96	2.62	3.92	1.39	3.00	4.05	1.48	2.63	38	2
Depreciation of long-term tangible property	17.19	17.85	14.85	16.29	9.42	12.15	15.40	14.74	19	9
Depreciation of basic stock	11.96	13.91	18.74	15.33	14.97	10.65	12.24	13.97	18	8
Other direct primary costs ^b	13.82	15.90	19.33	27.58	27.60	28.06	25.90	22.60	25	13
Other direct secondary costs ^b	17.48	25.84	10.93	12.39	14.51	19.04	21.11	17.33	28	10
Production overhead	6.79	2.99	5.90	8.66	9.72	10.52	12.66	8.18	36	5
Management overhead	1.71	0.38	2.49	7.87	6.80	8.99	6.45	4.96	63	3
Costs together	133.88	163.70	169.54	176.92	177.23	178.04	179.59	168.42	9	100
By-product ^c	20.72	18.99	21.51	21.63	19.15	21.59	19.47	20.44	5	-
Total costs per ewe and year	113.16	144.72	148.03	155.29	158.08	156.45	160.12	147.98	10	-
Market price per kg of milk	0.707	0.701	0.766	0.835	0.745	0.836	0.883	0.782	8	-
Market price per lamb	38.17	29.59	28.70	20.61	22.96	28.31	27.15	27.93	19	-
Total revenues per ewe and year ^d	69.25	72.74	66.85	56.37	68.37	79.56	71.71	69.26	9	-
Subsidies per ewe and year ^e	0	21.41	20.89	22.20	21.45	16.43	5.37	15.39	54	-
Profit or loss per ewe and year without subsidies	-43.91	-71.98	-81.18	-98.92	-89.71	-76.90	-88.41	-78.71	-21	-
Profit or loss per ewe and year with subsidies	-43.91	-50.57	-60.29	-76.72	-68.26	-60.47	-83.04	-63.32	-20	-

Source: economic database of APRC Nitra, own calculations

^aCoefficient of variation

^bFor more details see notes to Table 2

^cValue of manure (0.0055 t * 3.65 € per t), wool (production in kg * 0.664 € per kg) and lambs born alive (3.8 kg of live weight per lamb * 3.319 € per kg * number of lambs) per ewe and per year

^dBased on the milk yield, milk price, number of lambs sold per ewe and year and lamb price

^eAppropriate value of subsidies paid per livestock unit (LU; one ewe = 0.15 LU). For more details see section "Material and Methods"

summarized in Table 1 and 2. The higher values of profit to cost ratio of milk production (from 63 % to 72 %) was published by Arbel *et al.* (2001) in spite of the comparable value of market prices of milk and of costs per cow and feeding day. High level of milk yield (26.71 kg to 31.70 kg per feeding day) which finally reduced the unit cost per kg of milk (0.190 € per 1 kg) was the main determinant of difference in this case. Contrary to our study, almost two times higher value of cost per milk unit was found (0.359 € per kg, Table 2). On the other hand, Roest (2000) noted comparable value for the profit to cost ratio (- 6 %) in milk production in spite of extremely low milk yield (6.73 kg per feeding day) per cows reared in mountain and foothill regions. Positive impact of higher market price of milk (0.510 € per kg) on the profit to cost ratio was confirmed in this study.

Total profit to cost ratio in dairy sheep varied from - 64 % (without subsidies in 2009) to - 35 % (with subsidies in 2007) over the analysed period (Figure 1). Negative value of profit to cost ratio - 40 % and - 38 % was found also for dairy sheep farms in 2002 and 2003 (Vláčil, 2005) based on comparable value of production (58 kg of milk and 0.69 of lambs per ewe and year) along with market prices of dairy sheep commodities (0.594 € per kg of milk and 33 € per lamb). Economic situation in these farms changed to profitable (10 % and 16 %) when support per sheep breeding and cheese production (95 € and 102 € per ewe and year) was considered (Vláčil, 2005). Negative ratio of economic efficiency in sheep farms analysed in our study reduced in the individual year by 10 p.p. after adding of subsidies (Figure 1). Positive influence of subsidies on profitability was confirmed also

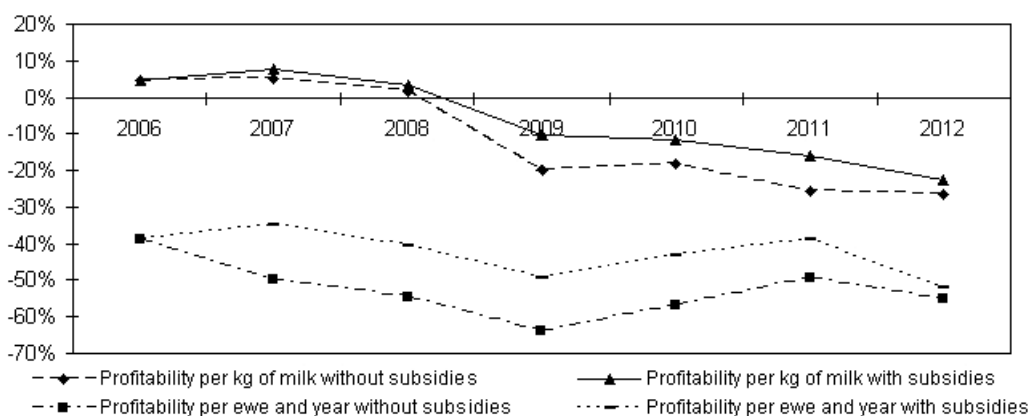
in other dairy (Benoit and Laignel, 2011) as well in meat sheep farms (Milerski *et al.*, 2006; Cehla *et al.*, 2012).

Positive value of profit to cost ratio (+ 119 % and + 54 %) was published for extensive and intensive dairy sheep farms in Hungary, respectively (Jávor *et al.*, 2005). Apart from the higher value of milk yield in these flocks (90 L and 280 L per ewe and year, resp.) compared to our study (63 kg per ewe and year, Table 1), the main reason is that gross margin (9.800 and 15.100 HUF per ewe and year) was used as the basic economic indicator in this paper. Based on this methodology, all of the costs items were not considered and therefore value of annual costs per ewe was lower (8.300 HUF \approx 33 €) compared to our study (148 € per ewe and year on average over the analysed period, Table 3). Positive economic results in family farms were also found in meat sheep for period 2005 to 2008 (Benoit and Laignel, 2011). Likewise in previous paper, gross margin was used in the economic evaluation. Moreover, revenues were not drawn exclusively from sheep farming, since there were also other activities (e.g. crops production). These authors also stated a lower value of revenues and incomes per meat sheep farms compared to dairy cattle herds. Likewise, value of incomes in meat sheep farms located in upland zones (which is typical for dairy sheep farms in our study) was lower than those in plainland farms in their paper. This would be also the case of disproportion in the economic efficiency between cattle and sheep farms analysed in our study. Trend of profitability ratio declined in period 2006 – 2009 in analysed dairy cattle (by 15 % and 25 % with and without of subsidies) and sheep farms (by 11 % and 25 % with and without of subsidies) which is comparable with findings of Benoit and Laignel (2011) for cattle and meat sheep farmers.

Profit to cost ratio - individual indicator

For the profit to cost ratio of the individual cost items similar characteristic were found in dairy cattle and sheep farms. The lowest proportion of the economic result (profit or loss) on the own feeds, depreciations in basic stock and on the other direct costs for the analyzed farms was recorded (Table 4 and 5). On the contrary, the higher share of economic efficiency was calculated per repair and services costs, overheads and other material costs. Results published by Ubrežiová and Mihina (1995; 1998) for cattle and by Vláčil (2005) for dairy sheep correspond to our founding. Differences between the cattle and sheep were in profit to cost ratio for purchased feeds and labour costs only. Higher intensity of production in dairy cattle compared to sheep (semi-extensive farms) lead to upper consumption of purchased feeds. Therefore, purchased feeds belonged to the costs items with lower value (0.07 and 0.08 with and without of subsidies) of profit to cost ratio in cattle (Table 4). Contrary, for dairy sheep farms, a higher need of human labour is typical compared to cattle. According to this, labour costs took the place among the cost items with lower value (2.12 and 2.61 with and without of subsidies) of profit to cost ratio in sheep (Table 5). In respect of labour costs it should be also mentioned that investment into the technological equipment for milking will be accompanied with higher material consumption (disinfecting, spare parts), energy consumption (electricity and water), and the cost of repairs and depreciation of fixed assets. However, savings in labour costs and charges will be higher than operating costs for parlours (Vláčil and Mihina, 2007).

Generally it can be said that value of profit to cost ratio of the individual cost item (given in Table 4



Source: economic database of APRC Nitra, own calculations

Fig. 1: Profit to cost ratio (profitability) of milk production in cattle and in sheep flocks from 2006 to 2012

and 5) was preliminary determined by the value of the individual costs items per FD of cow as well per ewe and year (see last column of Table 2 and 3). Profit to cost ratio was lower for the cost items with higher value in farming and vice versa. Moreover, higher value of loss reached in sheep farms compared to cattle (Table 3 and 2) resulted to higher absolute values of profit to cost ratio in sheep (Table 5 and 4). Nevertheless, values intended inside the production system were only relevant for evaluation of the individual indicators of profit to cost ratio. When negative profit (loss) was calculated (from 2009 to 2012 in cattle farms and over the whole time period in sheep) a slightly lower ratios of profit to the individual cost items was found after including of subsidies.

Determinants of economic efficiency

Level of animal performance (e.g. milk yield,

number of sold lambs), price of the main inputs (feeds, other direct costs, labour and depreciations), market price of products along with the value and type of subsidies are the most important determinants of economic efficiency in dairy cattle and sheep farms. Individual influence of these factors on the economic efficiency of cattle and sheep production was outlined above. Therefore a comprehensive analysis along with development of further micro and macro economic factors will be taken into account in the following text.

During the period 2006 - 2008, milk yield per cow and number of dairy cows in the analyzed dairy cattle herds increased (Table 1). Average level of milk yield in Slovak cattle herds slightly increased as well, but the number of dairy cows decreased nearly by 9 % during this period (Figure 2). Similarly in dairy sheep farms, an increase in milk yield and in size of analysed dairy

Table 4: Profit to cost ratio of the individual cost items and its basic statistical characteristics in the analysed dairy cattle farms from 2006 to 2012 (€)

Individual items of cost's formula	2006	2007	2008	2009	2010	2011	2012	Mean value	v (%) ^a
Labour costs without subsidies	0.04	0.04	0.01	0.16	0.12	0.21	0.20	0.11	73
with subsidies	-	0.06	0.03	0.08	0.08	0.14	0.17	0.09	57
Own feed costs without subsidies	0.01	0.01	0.00	0.03	0.07	0.05	0.06	0.03	80
with subsidies	-	0.02	0.01	0.02	0.04	0.03	0.05	0.03	61
Purchased feed costs without subsidies	0.03	0.03	0.01	0.12	0.06	0.19	0.14	0.08	82
with subsidies	-	0.04	0.02	0.06	0.04	0.12	0.12	0.07	66
Other direct material costs without subsidies	0.07	0.08	0.03	0.33	0.31	0.32	0.36	0.21	69
with subsidies	-	0.12	0.05	0.17	0.20	0.20	0.31	0.18	49
Repair and services costs without subsidies	0.19	0.15	0.08	0.78	0.68	1.96	1.28	0.73	95
with subsidies	-	0.24	0.15	0.41	0.44	1.24	1.10	0.60	77
Depreciation of long-term									
tangible property without subsidies	0.05	0.06	0.02	0.16	0.15	0.32	0.17	0.13	77
with subsidies	-	0.09	0.03	0.08	0.10	0.20	0.14	0.11	53
Depreciation of basic stock without subsidies	0.02	0.03	0.01	0.10	0.09	0.13	0.15	0.08	74
with subsidies	-	0.04	0.02	0.05	0.06	0.08	0.13	0.06	60
Other direct primary costs without subsidies	0.03	0.03	0.01	0.12	0.11	0.16	0.15	0.09	71
with subsidies	-	0.04	0.02	0.06	0.07	0.10	0.13	0.07	55
Other direct secondary costs without subsidies	0.03	0.03	0.01	0.12	0.10	0.14	0.15	0.08	69
with subsidies	-	0.05	0.02	0.06	0.07	0.09	0.13	0.07	53
Production overhead costs without subsidies	0.06	0.06	0.02	0.26	0.17	0.25	0.24	0.15	67
with subsidies	-	0.10	0.04	0.14	0.11	0.16	0.20	0.12	45
Management overhead costs without subsidies	0.09	0.07	0.03	0.33	0.45	0.28	0.26	0.22	72
with subsidies	-	0.11	0.06	0.17	0.29	0.18	0.22	0.17	47

Source: own calculations

^a Coefficient of variation

Table 5: Profit to cost ratio of the individual cost items and its basic statistical characteristics in the analysed dairy sheep farms from 2006 to 2012 (€)

Individual items of cost's formula	2006	2007	2008	2009	2010	2011	2012	Mean value	v (%) ^a
Labour costs without subsidies	2.65	3.29	2.59	2.94	2.22	2.03	2.56	2.61	15
with subsidies	-	2.31	1.92	2.28	1.69	1.60	2.41	2.12	17
Own feed costs without subsidies	1.20	1.57	1.77	2.06	2.03	2.20	2.70	1.93	23
with subsidies	-	1.10	1.31	1.59	1.55	1.73	2.53	1.58	28
Purchased feed costs without subsidies	10.21	4.96	8.94	27.42	43.78	12.42	8.17	16.56	78
with subsidies	-	3.48	6.64	21.27	33.31	9.77	7.67	13.19	74
Other direct material costs without subsidies	7.84	37.72	11.56	48.25	19.24	13.56	14.10	21.75	65
with subsidies	-	26.50	8.59	37.42	14.64	10.66	13.24	16.99	60
Repair and services costs without subsidies	22.41	27.49	20.73	71.14	29.87	19.00	59.60	35.75	54
with subsidies	-	19.31	15.40	55.18	22.73	14.94	55.98	29.42	57
Depreciation of long-term tangible property without subsidies	2.55	4.03	5.47	6.07	9.53	6.33	5.74	5.67	35
with subsidies	-	2.83	4.06	4.71	7.25	4.98	5.39	4.54	33
Depreciation of basic stock without subsidies	3.67	5.17	4.33	6.45	5.99	7.22	7.22	5.72	22
with subsidies	-	3.63	3.22	5.00	4.56	5.68	6.78	4.65	25
Other direct primary costs without subsidies	3.18	4.53	4.20	3.59	3.25	2.74	3.41	3.56	16
with subsidies	-	3.18	3.12	2.78	2.47	2.15	3.21	2.87	13
Other direct secondary costs without subsidies	2.51	2.79	7.42	7.99	6.18	4.04	4.19	5.02	40
with subsidies	-	1.96	5.51	6.19	4.71	3.18	3.93	4.00	36
Production overhead costs without subsidies	6.46	24.10	13.76	11.42	9.22	7.31	6.98	11.32	51
with subsidies	-	16.93	10.22	8.86	7.02	5.75	6.56	8.83	41
Management overhead costs without subsidies	25.74	188.22	32.63	12.57	13.19	8.55	13.71	42.09	143
with subsidies	-	132.24	24.23	9.75	10.04	6.73	12.87	31.66	131

Source: own calculations

^aCoefficient of variation

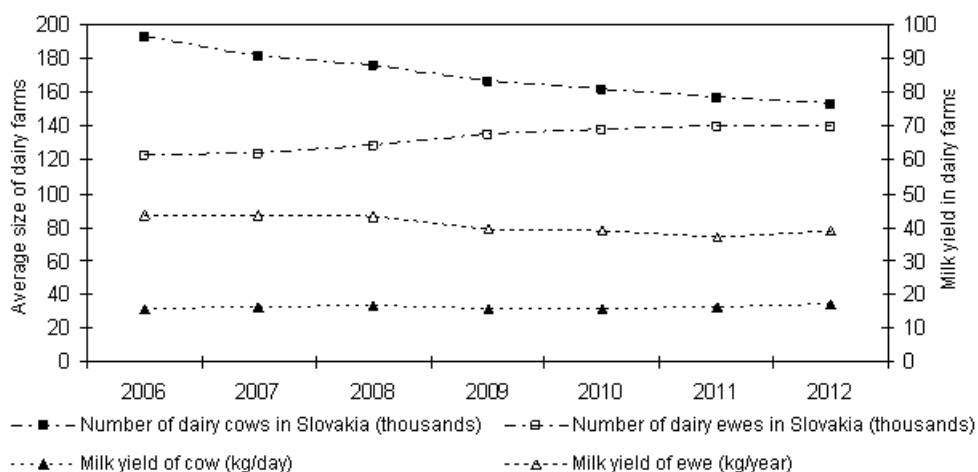
sheep farms (Table 1) along with stabilisation in these parameters (- 1 % in milk and + 5 % in number of ewes in farm) in Slovakia was found in this period (Figure 2). According to these trends, higher stability in the agricultural sector can be indicated for the sheep farms together with cattle farms analyzed in our study.

When analyzing the determinants of economic efficiency in cattle and sheep it is also necessary to take into consideration the price of diesel, which creates predominant part of costs for roughage and concentrates (Gunlu *et al.*, 2003; Blaskó *et al.*, 2012). Costs for grain and forage feeds represent from 30 to 35 % of total costs in dairy cattle and sheep farms (Krupová *et al.*, 2012). In Slovakia, price of diesel slightly increased (from 1.320 to 1.380 € per litre) during the period 2006 - 2008 mainly due to the reduction of its supply at world market. At the same time, increase in diesel price was slightly taken

care of by strengthening of USD exchange rate against the EUR (Figure 3). Increase in the level of diesel price influenced the costs for feed production (Figure 4 and 5) and also the level of costs for own (mostly forage) and purchased feeds used in analyzed dairy cattle and sheep farms (Table 1). For comparison, decrease in production costs for forage feeds was officially published in Slovakia for this period (Figure 5). The costs of grain feeds at first jumped to 176 € and then decreased to 162 € per tonne (Figure 4). It was not possible to quantify the real costs for feed production in database of evaluated farmers. Nevertheless, it is supposed that the mentioned disproportion could be caused by the difference between the real costs for feeds production and the value (price of intermediate goods) they were accounted in cattle and sheep economic evidence. This assumption is partly confirmed by the fact that average price of grain feeds

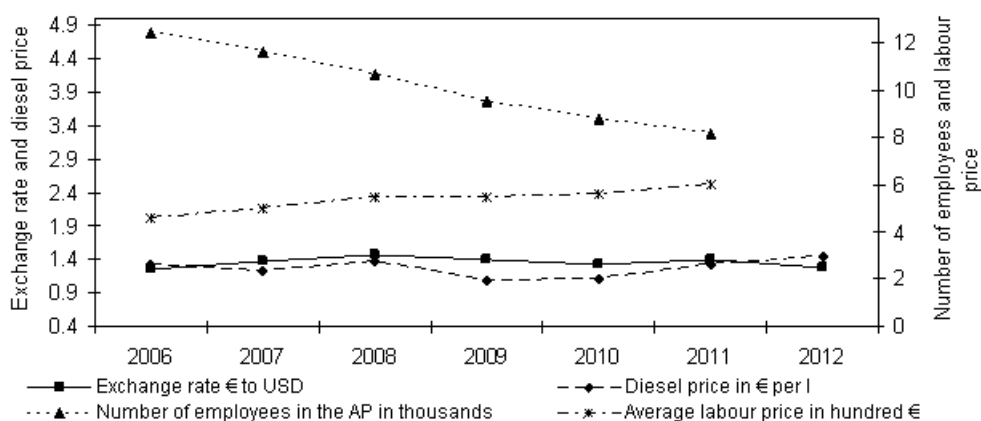
on the market slightly raised over the whole time period (Figure 4). This trend could presumably result in rise of intradepartmental price of all plant commodities, inclusive of forage feeds. Additionally, increase of milk yield in dairy cattle and sheep connected with rise of nutrients requirements was another factor that influenced the increase of feed costs in analyzed farms (Table 2 and 3) which was also confirmed in paper Kuipers *et al.*

(1999). In analysed farms, the unit costs per kg of milk finally raised by 12 % in cattle (Table 2) and costs per ewe and year by 31 % during the years 2006-2008 (Table 3). Regarding the value of own feed costs, they should be calculated only in the own cost value for given plant commodities. Finally, it seems to be a very useful solution to optimize the value of own feed costs in animal production.



Source: RIAFE (2013); economic database of APRC Nitra, own calculations

Fig. 2: Milk yield and average number of dairy cattle and sheep in Slovakia in 2006 to 2012



Source: EUROSTAT (2013); SO SR (2013)

^aNumber of employees in the AP and the average labour price have not been available for 2012 until now

Fig. 3: Development of the exchange rate of € to USD, diesel price, number of employees and average labour price^a in animal production (AP) in Slovakia from 2006 to 2012

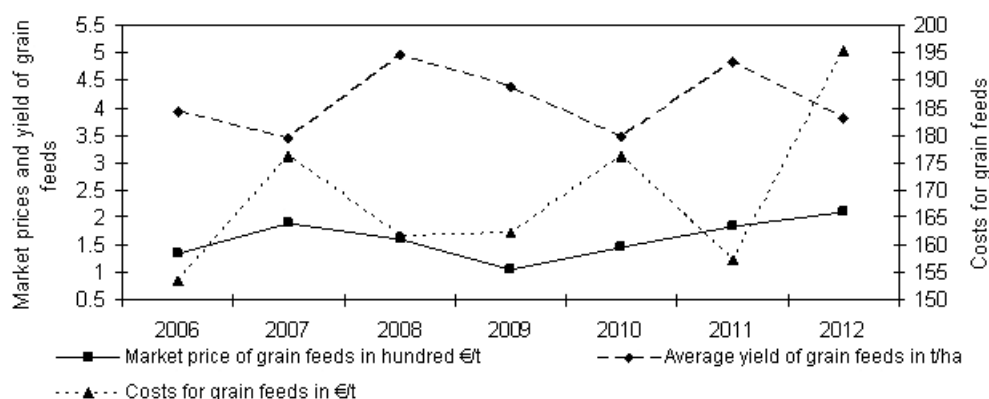
Labour costs, similarly to feed costs, are an important item in calculation formula of dairy cattle and sheep farms (Table 2 and 3). Average monthly wages in the branch of animal production slightly rose (nearly by 90 €) during the period 2006 to 2008 in Slovakia. On the other hand, a number of employees in animal production during this period decreased (Figure 3). This disproportion could cause the irregular development of labour costs (increase in 2006-2007 and decrease in 2007-2008) in dairy cattle herds (Table 1). Contrary, value of labour costs in sheep was exempted from this disproportion probably due to existence of seasonal employees and external personnel typical for this production system.

Market supply (production) of milk increased until 2008 (Table 1) with simultaneous increase of market price of milk (Table 2) in cattle producers. This situation resulted in surplus commodity on the market in 2009 and 2010 and in decrease of demand for milk (Table 2) which was also confirmed in paper Blaskó (2012). The consequence of these events caused to a drop in milk price in 2009 and 2010 (Table 2). This negative situation was partly compensated by the addition of national direct payment per dairy cow and support per dairy cow - help in milk crisis - paid in dairy sector in 2010 (Table 2). In dairy sheep, raised demand for dairy sheep products over the whole time period positive expressed in the milk price. These economic conditions focused farmers more on milk production compared to producing lambs especially if the price of lambs was close to its minimum. Number of ewes in the flock and milk yield per ewe slightly increased in the consequence (Table 1).

However, uncertainty in overall economic situation in 2009 lead to reduction in inputs mainly these

for feeds. Yield of forage and grain feeds per hectare slightly decreased in 2009 - 2012 and unit costs for feed production increased by 5 % (Figure 4 and 5). This situation was related with the higher feed prices (+ 34 %) in 2010 and 2011 compared to the rest of the mentioned years and with the cancellation of tax benefits for fuel in 2011 which were implicated in the agriculture sector in previous period. At first, dairy cattle farmers tried to solve this unfavourable situation mainly by reduced amount of purchased feeds and their substitution by own feeds. In addition, the producers who supply the most of the required amount of purchase the own feeds, probably have an important advantage in decreasing the production costs comparing the ones who buy from outside (Gunlu *et al.*, 2003). At the end of evaluated period, the situation in cattle nutrition, especially in purchased feeds, returned to the state before 2009. Total increase of feeding costs in cattle (+ 55 %) was based on change in cost for own (44 %) and for purchased feeds (89 %) over the whole period (Table 2). Regard to the situation in 2009, further reaction of dairy cattle farmers was a short-term decreasing of the size in the analyzed dairy cattle herds by 5 % in 2009 (Table 1). However, according to average Slovakian data reduction in numbers of dairy cows took place almost over the whole period (Figure 2). However, these changes were not effective from the complex point of view mainly due to the milk yield per cow slowly decreased (Table 1, Figure 2).

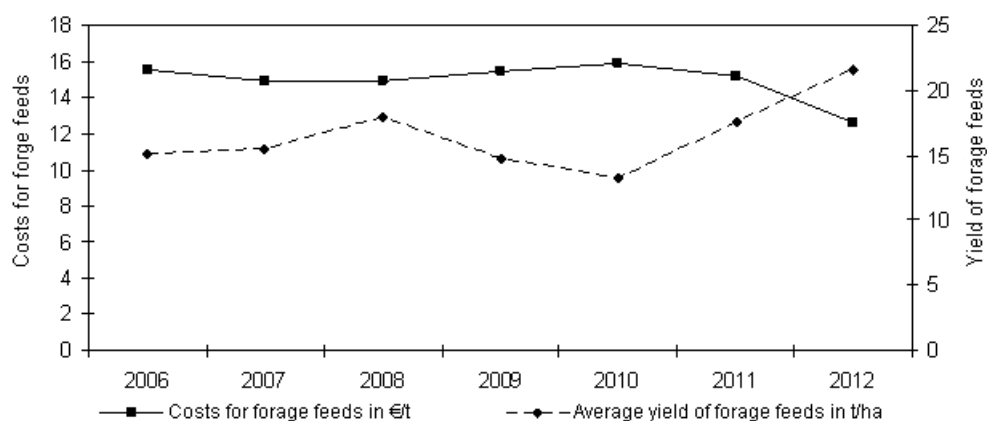
Similarly in sheep farms, value of costs for purchased feeds was strongly influenced by overall economic conditions, mainly by market prices of grain feeds after 2009 (Figure 4). Reduction was observed even in costs for own feeds (Table 3). In the context of these facts, average milk yield per ewe and year in



Source: RIAFE (2013)

^aAverage values for wheat, rye, barley, grain maize and oats

Fig. 4: Base production and economic parameters of grain feeds^a from 2006 to 2012



Source: RIAFE (2013)

^aAverage values for green maize, multiannual forage crops, meadows and pasture

Fig. 5: Base production and economic parameters of forage feeds^a from 2006 to 2012

Slovakia reached only the 64 % of the yield in analysed sheep farms and its value slightly declined over the time (Figure 2). This situation was not compensated even by the fact that increase in size of flock was found in dairy sheep farms analysed in our study as well according to average Slovakian data (+ 10 %, Table 1 and Figure 2) in the last four years compared to pervious period. In addition of reduced value of subsidies, farm profitability remained in red numbers (Table 3, Figure 1).

Value of labour costs changed by + 23 % and + 57 % in last four years compared to the previous period and by + 49 % and + 108 % over the analyzed period in cattle and sheep farms, respectively. In comparison, number of employees decreased by 24 % and the average value of monthly wages increased by 14 % in Slovakia (Figure 3). Existence of over-employment along with alternatively less effective utilization of labour power in the production process in analyzed farms can be indicated.

Concerning the value of revenues in 2009 and 2012, the unit milk price changed + 22 % and + 15 % (with and without subsidies, respectively) in dairy cattle (Table 2). Total revenues increased by 27 % in sheep farms in this period. However, total incomes remained almost the same (77 € in 2012 vs. 79 € in 2009) when considering of subsidies (Table 3). Finally, combination of the above mentioned micro and macro economic factors and animal performance resulted in the increase of loss by more then two times (to 0.10 € per each kg of milk or to 1.62 € per FD of cow on average, Table 2) in analysed cattle farms and loss in sheep remained almost at the same negative level (85 € per ewe and year on average, Table 3).

CONCLUSION

Dairy cattle and sheep farmers should concentrate on accounting the costs only for categories to which they belong (especially overhead costs) to define objective value of cost for given value of production. Moreover, dairy farmers should connect to marketing associations to promote higher market prices of milk commodities. Experience suggests that milk price is higher by 20 % on average for farmers cooperating in marketing associations compared to the individual sellers. Nevertheless, possibilities to increase milk price individually per additional milk fats and proteins paid to farmers by dairies are small. Regarding the revenues, it seems to be useful to focus on diversifying their structure by farmers. Diversification (on cow-calves/meat sheep, plant, biogas production and services) can spread business risk to the widest base of outputs. Moreover, universal orientation of production can reduce the response time to market changes and lead to higher flexibility of organizational and cost systems.

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Review

METHODS OF METHANE MEASUREMENT IN RUMINANTS

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ABSTRACT

This review is devoted to methodology, which can help direct and indirect measurement of methane emissions. This paper will be useful for expanding the knowledge base of researchers, farm planners, and policymakers as they work to develop and maintain sustainable environment conditions for farming systems in Slovakia. The following methods like respiration chamber, SF₆ technique, alternative methods, micrometeorological methods, proxy methods, in vitro gas production technique, and models for predicting methane production are described. Above mentioned methods are compared and their advantages and disadvantages are enlisted.

Key words: methane; emission; method

INTRODUCTION

Animals contribute to global warming by releasing of greenhouse gas emissions. The major greenhouse gas produced from enteric fermentation of ruminants during the normal digestive process is methane (CH₄). Fermentation CH₄ is the sum of enteric CH₄ and manure CH₄ (Veysset *et al.*, 2010; Mihina *et al.*, 2012). Enteric fermentation from livestock is a large source of methane, which has a global warming potential 23 times that of carbon dioxide (Bhatta *et al.*, 2007; Loh *et al.*, 2008). Methane from agriculture arises primarily from enteric fermentation; therefore, ruminants (especially beef and dairy cattle) are mainly responsible for enteric emissions of CH₄ (Kebreab *et al.*, 2006). Enteric CH₄ from ruminant livestock accounts for 17-37 % of anthropogenic CH₄ (Beauchemin *et al.*, 2010; Sejian *et al.*, 2011).

Methodologies for measuring CH₄ emissions range from animal respiration chambers to estimation

of model techniques. While chambers provide a simple measurement technique that is ideal for testing treatment differences there are disadvantages, too as only a small area or number of animals may be studied (McGinn *et al.*, 2008; van Haarlem van *et al.*, 2008; Flesch *et al.* 2007). The latest technology developed to estimate CH₄ more accurately is the micrometeorological mass difference technique (Harper *et al.*, 1999; Sejian *et al.*, 2011).

Emission of CH₄ in ruminants differs depending on factors like animal species, breed, pH of rumen fluid, ratio of acetate: propionate, methanogen population, composition of diet and amount of concentrate fed. Among the ruminant animals, cattle contribute the most towards the greenhouse effect through methane emission followed by sheep, and goats, respectively (Charmley *et al.*, 2008; Bhatta *et al.*, 2008).

The purpose of the current study was to describe new methods for direct and indirect measurement of methane emissions.

Respiration chamber

The principle of the chamber is to collect exhaled CH_4 emissions from all sources of enteric fermentation (mouth, nostrils, and rectum) from the animal and to measure the concentration. Chambers are divided into two types, the closed-circuit and the open-circuit. The closed-circuit system is almost not used and preferred are open-circuit chambers. An air pump removes all air from the space through a flow meter and gas sensors in the open-circuit system. Each chamber is fitted with internal ventilation fans for efficient mixing of expired gases and incoming air. Air inlet is located at the front and an air outlet at the back. Fresh air to chamber is directly drawn from outside or through an air conditioning system to control humidity and temperature. The chamber is equipped with sensors for measuring relative humidity, temperature and barometric pressure. These allow air flow data to be adjusted for dry, standard temperature and pressure conditions. Outlet gas from each chamber is continuously sampled for analysis. Air flow is ducted via flexible polyurethane hoses. Air circulation is provided throughout the chambers at continuous but adjustable flow rates (usually 100-250 $\text{L}\cdot\text{min}^{-1}$) (Chagunda *et al.*, 2011; Storm *et al.*, 2012).

Methane emission is calculated from flow and gas concentration in inlet and outlet air from the chamber. The difference between the outgoing and incoming amount of methane expresses the methane emission (Muñoz *et al.*, 2012). Outlet gas from each chamber is continuously sampled for analysis. A multigas analyser with capability for measurement of methane and other gasses as carbon dioxide, and oxygen is used for the gas analyses (Pinares-Patino *et al.*, 2008a; Chagunda *et al.*, 2011).

SF_6 tracer

The principle is that methane emission can be measured if the emission rate of a tracer (non-toxic, physiologically inert, stable) gas from the rumen is known (Hegarty, 2013). SF_6 was selected from many comparisons, because it has an extremely low detection limit (Muñoz *et al.*, 2012). The gas should mix with rumen air in the same way as methane. The SF_6 technique involves the use of a SF_6 permeation tube dosed into the reticulo-rumen (Lassey *et al.*, 2001). The calculation of daily CH_4 emission is based on the $\text{CH}_4:\text{SF}_6$ ratio of concentrations (adjusted for background concentrations) and the specific pre-calibrated permeation rate of SF_6 from the particular permeation tube deployed in the animal.

SF_6 is filled into small permeation tubes. The rate of diffusion of SF_6 out of the permeation tubes is measured by placing them in a 39°C water bath and measuring the daily weight loss until it is stable. The permeation tube containing ultra-pure SF_6 is placed in

the rumen of an animal before the experimental period (Martin *et al.*, 2008). The sampling apparatus consists of a collection canister, a halter and capillary tubing. A representative of breath gas sample, containing respired and eructated gas is collected through a capillary tube placed at the nose of the animal, fitted to a halter, or behind the head and connected with the evacuated canister (approximately 2.5 L); the tubing regulates the sampling rate for 24 hours (Lassey *et al.*, 2001). This strategy requires two suites of canisters (the one removed became free once the collected samples were transferred to the analysis laboratory) (Bárbaro *et al.*, 2008). The concentration of SF_6 and CH_4 in the canister is determined then by gas chromatography. The methane emission is calculated from the release rate of SF_6 and concentration of SF_6 and CH_4 in the containers in excess of background level (Storm *et al.*, 2012).

Pinares-Patiño, Clark (2008) and Laubach *et al.* (2008) recommended the use of SF_6 method in grazing cattle involving large herds. The tracer technique is now widely used in New Zealand and many other countries for CH_4 emission measurements on grazing and pen-fed cattle, sheep, deer and alpacas (Pinares-Patiño *et al.*, 2008b). CH_4 emission estimates SF_6 method revealed slightly lower (by 5-10 %) than the respiration chamber measured values. However, other studies with cattle using hoods or respiration chambers (Grainger *et al.*, 2007) reported SF_6 tracer estimates slightly higher (by 1-2 %) than calorimetric estimates.

Alternative methods

More applications of alternative methods are combined with milking and feeding. The animals entering in automatic milking or feeding system are recognized and concentrations of CH_4 and CO_2 are measured. Air is continuously pumped through the equipment to quantify flow and thereby CH_4 and CO_2 emitted during milking and feeding.

Garnsworthy *et al.* (2012a) developed a novel technique based on sampling air released by eructation during milking. Methane analyzers are installed in automatic milking stations. Belching frequency and methane released per eructation are used to estimate methane emission rate. Air is sampled continuously from the feed mangers in the milking stations at 1 $\text{L}\cdot\text{min}^{-1}$ via an 8-mm diameter polyethylene tube, approximately 3 m in length, connected to the gas inlet port of the infrared methane analyzer with a range of 0 to 10.000 $\text{mg}\cdot\text{kg}^{-1}$.

The same authors (Garnsworthy *et al.*, 2012b) recorded methane emissions of cows during milking using methane analyzers installed in automatic milking stations, modified as respiration chamber. Methane concentrations in air released by eructation are measured continuously at each milking and eructation data are used to calculate individual daily means for methane

emission rate during milking. Air blows through the instrument by the pump between the gas inlet port and analyzer. Air is sampled continuously during the stay in the milking stations via a polyethylene tube, connected to the gas inlet port of analyzer. The port for the exhaust air from the analyzer is vented into the space at least 3 m from any sampling point.

Hegarty (2013) describes the device patented in USA called Emission monitoring unit, which measures emissions from individual cattle repeatedly over short timed periods whenever they visit the unit to consume a delivered mixture. Air is continuously drawn into the space where cattle received feed, and CH₄ and CO₂ flux are calculated continuously by multiplying the CH₄ or CO₂ concentration by the flow rate of air.

Other methods under development include the micrometeorological technique, combined feeder and CH₄ analyzer. An additional method for estimating methane emissions from livestock is based on the use of CO₂ as a tracer gas. Instead of using externally some gas, the naturally emitted CO₂ is used to quantify CH₄ emission (Madsen *et al.*, 2010). The exhaled air contains both the gases CO₂ and CH₄ (Laubach *et al.*, 2004).

The calculations are the similar as for the SF₆ tracer technique (just replacing SF₆ with CO₂). Corrections can be made for growing and lactating animals. The CO₂ method can be used to quantify methane production under different circumstances, for example from a dairy cow's barn and individual estimates for cows visiting an automated milking system (Storm *et al.*, 2012). Lassen *et al.* (2012) recorded individual methane (CH₄) and CO₂ production repeatedly on high number of dairy cows during milking also in an automatic milking system. They used a portable air sampler and analyzer unit based on transform infrared detection. The ratio between CH₄ and CO₂ was used as a derived measure with the idea of using CO₂ in breath as a tracer gas to quantify the production of methane. The repeatability was sufficient. The results of their study suggested that the CH₄ to CO₂ ratio measured using the non-invasive method is suitable and may be useful in both management and genetic evaluations. The instruments combined with automatic milking system may be useful to generate large data for genetic evaluation of CH₄ production in dairy cattle.

Micrometeorological methods

Micrometeorological methods are defined as measuring fluxes of gas in the free atmosphere and relating these fluxes to animal emissions. The methods are based on measurements of wind velocity and methane concentration, but the number of measuring points and the theories used to calculate emission rates differ between methods. The external tracer ratio technique can be used, where a tracer gas is released in the paddock or barn, and the concentrations of tracer and methane are

measured in the surroundings (Harper *et al.*, 2011). This category of methods also includes the technique of mass balance in enclosed barns, where ventilation rate and concentrations in inlet and outlet are used to estimate the emission. While it is relatively easy to estimate emission rates from mechanically ventilated closed barns, naturally-ventilated buildings are problematic because of difficulties with measuring air exchange rates (Derno *et al.*, 2009). These types of buildings are commonly used for cattle since they are not especially susceptible to draughts and temperature changes and no extra heating is required. Air exchange rates in these buildings depend on the temperature gradient, temperature humidity index, and the air velocity. In this case, the release rates of harmful gases may also depend on external and uncontrollable parameters such as wind speed and the other parameters of outside environment. This method is particularly important in the current period; the present trend in milk production in Europe is to change to systems with loose housing in naturally-ventilated buildings (Ngwabie *et al.*, 2009).

Bjorneberg *et al.* (2009) used an open-path spectrometer operating in the monostatic mode for measuring methane. In this instrument, radiation from an incandescent silicon carbide source is collimated and passed into an interferometer. The exit ray from the interferometer leads onto an external beam splitter, so half the radiation is conducted into a 250 mm telescope that expands the beam due to magnification of its collimation. The diameter of the expanded beam at a distance of 50 m from the telescope is less than 400 mm. A cube-corner retro reflector is mounted at an appropriate distance from the telescope (usually between 150 and 250 m) and is aligned so that the reflected beam is returned to the telescope. The telescope reduces the beam back to a diameter of about 40 mm. The beam is driven from the telescope to the external beam splitter, which passes the beam to a cooled mercury cadmium telluride detector. Interferograms are measured at 70 s intervals. Quantitative determinations of CH₄ concentrations (also NH₃ and N₂O) are performed by partial least squares regression of the open-path spectra (Bjorneberg *et al.*, 2009).

A significant improvement in methane measurement accuracy is contributed by micrometeorological techniques which allow accurate emission estimates from agricultural sources via a dispersion technique (also called inverse dispersion technique) (Flesch *et al.*, 2005). This method has the advantages, which include non-interference, and the ability to incorporate the measurement footprint over larger areas. Inverse-dispersion methods have been used with success in several studies of feedlot gas emissions (Flesch *et al.*, 2007; Loh *et al.*, 2008; McGinn *et al.*, 2011). However, there are several limitations to using

inverse dispersion methods including wind conditions and the need for source homogeneity (van Haarlem van *et al.*, 2008).

Lagrangian Stochastic (bLS) method, belonging to category of dispersion techniques (but also in the category of micrometeorological techniques), is usually used in conjunction with global positioning system information from individual animals, to evaluate CH₄ emissions from pens of cattle (Laubach *et al.*, 2005). CH₄ concentration is measured using an open-path laser. Each laser path is located at a height of 1.5 m about 1 to 1.5 m outside the perimeter of the pens (McGinn *et al.*, 2009). The gas dispersion model contains vertical concentration profiles (Laubach *et al.*, 2008).

Methane emissions from grazing cattle are determined in a field experiment using paddock-scale (also belonging to micrometeorological) methods. The paddock-scale methods exploit how the gas, once emitted from the cattle, is transported and dispersed by the wind. Therefore, the emission rate may be calculated from measurements of wind speed, wind direction and turbulence, as well as CH₄ concentration upwind and downwind. The paddock-scale methods include a mass-budget approach, flux-gradient method and gas dispersion model. Accuracy is dependent on certain conditions, particularly whether the place is usually windy and free of obstructions that alter the turbulent airflow (Laubach *et al.*, 2008).

Loh *et al.* (2008) applied open path spectroscopic concentration measurements and a bLS dispersion model for evaluation of methane and total greenhouse gasses in situ from feedlot beef production for the first time. Their results are consistent with other studies using a similar approach to measure emissions on a farm scale.

Proxy methods

Proxy methods were developed with the purpose of examining many animals at a same time without complex and expensive equipment. Close relationship of methane emissions with parameters that can be measured in easily obtainable from samples of milk or feces is used (Dehareng *et al.*, 2012). Usually, the fatty acid profiles of milk are examined for correlations with methane production of the cows. The principle is that some fatty acids or fats in the milk or feces are correlated with either the feed composition or the amount of methanogens in the rumen (Vlaeminck *et al.*, 2006; Chilliard *et al.*, 2009).

The two challenges in using short-term breath measures as a proxy for measures of emissions are collecting data for an adequate period to provide a repeatable estimate of emission rate and scaling up from a short-term emission rate to methane production for whole day. These efforts resulting from the fact that the measurement is not entirely reliable and that a short term

enteric methane emission measurement is not identical to a measure of daily methane production made in a respiration chamber.

Use of spectrometry to predict the CH₄ emission of dairy cows has got high potential, too. (Dehareng *et al.*, 2012) investigated the feasibility to prognosticate CH₄ emissions using milk mid infrared spectra. The experiments aimed to induce a large variation in CH₄ emission by feeding different diets (fresh grass and sugar beet pulp; maize silage and hay; grass and corn silage with cracked corn, soybean meal and dried pulp). Milk sample of 50 ml was collected from each cow and analyzed by spectrometry. Results suggest the feasibility of direct CH₄ prediction from milk mid infrared spectra. This alternative method could be useful to predict the CH₄ emissions at farm level or at the regional scale and it also could be used to identify cows with low CH₄ emission.

In Vitro gas method

The gas measuring technique has been widely used for evaluation of nutritive value of feeds. More recently, the increased interest in the efficient utilization of roughage diets has led to an increase in the use of this technique due to the advantage in studying fermentation kinetics. Gas measurement provides a useful data on digestion kinetics of both soluble and insoluble fractions of feedstuffs (France *et al.*, 2000). This method has been modified for methane creation (Navarro-Villa *et al.*, 2011; Storm *et al.*, 2012).

The principle is to ferment feed under controlled laboratory conditions by natural rumen microbes. Feedstuffs are incubated at 39°C with a mixture of rumen fluid, buffer and minerals for a certain time period. The amount of total gas produced during incubation is measured and its composition analyzed, to obtain data on the *in vitro* production of methane. The method requires access to fresh rumen fluid, which is typically obtained from fistulated cows or other ruminants. The calculations are the same as for the CO₂ tracer technique.

Pellikaan *et al.* (2011) showed the gas production equipment which offers the possibility to determine total gas production, as a measure of organic matter fermentation, and methane synthesis simultaneously. With this system the maximum level of total gas production and methane synthesis can be determined, as well as the kinetics of synthesis. A fast screening of feedstuffs and additives for methane synthesis and total gas production is possible.

Models for predicting methane production

In many cases of scientific trials using the total national emissions calculation is not possible. Therefore there is an interest in being able to predict methane production using models based on existing data, such

as animal characteristics (weight, age, breed), feed characteristics (nutrient and energy content), intake data (dry matter or nutrients) or digested nutrients. Such models often use data derived from experiments conducted with cattle in respiration chambers, but not techniques for measuring methane which were applied in recent years. Tremendous progress has been made in the field of designing simulation models for predicting CH₄ emissions, and the latest integrated farm system models offer greater scope to accurately predict greenhouse gas emissions with the incorporation of climatic and management information (Ellis *et al.*, 2009; Sejian *et al.*, 2011). Dry matter intake (DMI), metabolizable energy intake, neutral detergent fibre, acid detergent fibre, ether extract, lignin, and forage proportions were considered in the development of models to predict CH₄ emissions (Ellis *et al.*, 2007).

Majority of methane models were developed from measurements obtained in respiration chambers. Some models require the proportion of roughage in the ration, while the other models require digested amounts of different nutrients. Total CH₄ production (L/d) in the cattle data set has been closely related to dry matter intake. Ramin and Huhtanen (2013) concluded that feed intake is the main determinant of total CH₄ production and that gross energy intake is negatively related to feeding level and dietary fat concentration and positively to diet digestibility, whereas dietary carbohydrate composition has only minor effects. CH₄ production was positively related to diet digestibility and negatively related to dietary fat concentration, whereas dietary carbohydrate composition had only minor effects. When authors expressed as a proportion of gross energy intake, CH₄ production was negatively related to feeding level and dietary fat concentration and positively related to diet digestibility and dietary concentrations of non-fibre carbohydrate and neutral detergent fibre.

A comparison of the above mentioned models leads to large differences in the estimates of methane emission. The model estimates are also associated with errors. The best equations developed by Ellis *et al.* (2007) for beef cattle, dairy cattle, and cattle in general had prediction errors of 14.4, 20.6 and 28.2 %, respectively. When models were evaluated with independent datasets, the prediction errors were increased.

The results of Ramin and Huhtanen (2013) indicate that CH₄ production can be predicted accurately from a set of variables that are available at the time of prediction. Equations predicting CH₄ production per unit of feed intake (gross energy or dry matter) are biologically more valid, and therefore it is recommended that CH₄ production is predicted as intake of gross energy (GE) or dry matter (DM) × production per unit (MJ of GE or kg of DM) of intake.

Methods of choice for estimating enteric methane

emission depend on aim, equipment, knowledge, time and money available, but interpretation of results obtained with a given method can be improved if knowledge about the disadvantages and advantages are used in the planning of experiments (Ramin and Huhtanen, 2013). The prediction models should use to predict emissions for each strategy (Legesse *et al.*, 2011; Aljaloud *et al.*, 2011; Kebreab *et al.*, 2006, 2008).

An inverse dispersion model was utilized to calculate CH₄ emissions from a commercial cattle feedlot and an adjacent runoff retention pond. The feedlot measurements were collected within the interior of the feedlot enabling a near continuous emissions record over the 12 d of the study period (van Haarlem *et al.*, 2008).

There have been several attempts to formulate mathematical models to predict CH₄ emissions from cattle. The models can be classified into 2 principal groups: empirical (statistical) models that relate nutrient intake to CH₄ output directly and dynamic mechanistic models that attempt to simulate CH₄ emissions based on a mathematical description of ruminal fermentation biochemistry (Kebreab *et al.*, 2008; Alemu *et al.*, 2011). A synthesis of the available literature suggests that the mechanistic models are superior to empirical models in accurately predicting the CH₄ emission from dairy farms. The latest development in prediction model is the integrated farm system model which is a process-based whole-farm simulation technique (Sejian *et al.*, 2011).

The model proposed by Moe and Tyrrell (cit. Kebreab *et al.*, 2006) is an empirical one developed using data from cattle, and the model relates intake of carbohydrate fractions to CH₄ production as follows: Methane (MJ/d) = 3.41 + 0.51 NFC + 1.74 HC + 2.65 C, where NFC = non-fibre carbohydrate (kg/d); HC = hemicellulose (kg/d); and C = cellulose (kg/d). In cases in which NFC values were not available, it was calculated as NFC = 100 – (CP + ether extract + ash + NDF), where CP = crude protein and NDF = neutral detergent fibre.

MOLLY model is a dynamic mechanistic model of nutrient utilization in cattle. Ruminal CH₄ production was predicted based on hydrogen balance. Excess hydrogen produced during fermentation of carbohydrates and protein to lipogenic volatile fatty acids (acetate and butyrate) is partitioned between use for microbial growth, biohydrogenation of unsaturated fatty acids, and production of glucogenic volatile fatty acids (propionate and valerate). The assumption is made that the remaining hydrogen is used solely and completely for methanogenesis (Kebreab *et al.*, 2004).

The rumen model of Dijkstra *et al.* (cit. Kebreab *et al.*, 2006) is the basis for the mechanistic model used in the present evaluation. The model is based on a series of dynamic, deterministic, and nonlinear differential equations. Kebreab *et al.* (2004) incorporated the rumen

model to a whole animal model that included nitrogen and phosphorus utilization. Bannink *et al.* (2011) developed a new stoichiometry for fermentation within the rumen based entirely on experimental observations with lactating dairy cows; therefore, model COWPOLL was modified to accommodate these stoichiometric coefficients. One of the fundamental differences in estimating CH₄ emissions between MOLLY and COWPOLL is the representation of microbes in the rumen and the coefficients of fermentation for transformation of substrate to volatile fatty acids. The MOLLY model uses 1 group of microbes, whereas COWPOLL separates the microbial community into 3 groups: amylolytic, cellulolytic bacteria, and protozoa (Kebreab *et al.*, 2008).

Charmley *et al.* (2008) described a modelling approach that estimates cattle methane emissions for various bioregions. The approach incorporates a metabolizable energy based model of animal production linked to a property herd economic model. This provides a flexible tool to evaluate animal and property herd dynamics on regional methane yields and live weight productivity, as well as to assess financial impacts. The model predicts that an important determinant of methane output per unit of product is reduced days to market. Reduced days to market may be achieved through a range of energy supplementation and marketing strategies. The modelling framework can be applied to a wide range of production, management and marketing scenarios to generate information on possible changes in methane emissions and financial gross margins. While these changes can be quantified, the output should be considered in light of the data deficiencies (Charmley *et al.*, 2008).

Many governments have implemented policies to reduce greenhouse gas emissions from agriculture and significant efforts are now being directed towards developing animal husbandry methods that lower enteric CH₄ emissions (Beauchemin *et al.*, 2010). To adequately assess greenhouse gas mitigation strategies, it is necessary to use a whole system modelling approach (Beauchemin *et al.*, 2010).

Three primary areas require refinement and relate to a better understanding of the forage base that makes up the major component of the diet. They include estimation of diet quality under selective grazing conditions; estimation of dry matter intake under heterogeneous grazing conditions; and precision of predicting methane yield from cattle grazing forages (Charmley *et al.*, 2008).

Mathematical models allow us to predict CH₄ production from cattle without undertaking extensive and costly experiments. The models used can be classified as either statistical models, which relate nutrient intake to CH₄ production directly, or dynamic

mechanistic models, which estimate CH₄ production using mathematical descriptions of rumen fermentation biochemistry (Kebreab *et al.*, 2004, 2006). Although many statistical models have been fairly successful in predicting CH₄ production, many have inputs that are not commonly measured and some may have difficulty predicting CH₄ production outside the range of values on which they were developed. These problems may be addressed by using commonly measured equation input variables and by developing models on expansive data sets compiled from multiple sources (Ellis *et al.*, 2007).

Advantages and inefficiencies of methods

Respiration chambers are regarded as the standard method for estimation of CH₄ methane emission from ruminants, because the environment can be controlled and the reliability and stability of instruments can be measured. However, results obtained in chambers cannot be extrapolated to loose housing animals, nor on pasture. This method is extremely slow and expensive (Hegarty, 2012), requires trained animals, restricted animal movement, causes stress, and have a high labour input (Pinares-Patiño, Clark, 2008). Respiration chambers are not used for determining methane production on farm.

The SF₆ method can be used to investigate nearly all aspects of feeding and nutrition, effect of chemical and physical composition, restricted or *ad libitum* feeding, different additives and grazing. However, using the method for investigation of dynamics of methane emission may be problematic. The following cons are maintaining a constant release rate from permeation tubes, effect of release rate upon emission rate of methane, background level determination, inconsistency between CH₄ measurements determined in chambers and with SF₆ (Storm *et al.*, 2012; Hegarty, 2013). The SF₆ method gives more variable results of methane emission than chamber measurements. The method is the only available method for measuring individual free ranging animals on pasture (Muñoz *et al.*, 2012). The number of animals is limited to 30 (Laubach *et al.*, 2008). The CO₂ technique is a newly developed approach for estimation of methane emissions from ruminants. It can be used under different conditions on large numbers of animals or for the overall estimation of herd emissions. However, this method is less precise than the respiration chamber methods.

The micrometeorological methods are still new and further development and documentation on reliability is needed, but the methods are valuable in evaluating whole dairy systems and interactions between animals and landscape. Unfortunately, all these methods are influenced by instabilities like non-steady state wind or movement of point-emission sources (McGinn *et al.*, 2008). It is also difficult to relate the CH₄ production to feed intake for grazing animals.

A disadvantage of *In Vitro* gas production

technique is that it only simulates the ruminal fermentation of feed, not emissions and digestibility by the entire animal. Furthermore, under normal conditions it does not include long-term adaptation of the ruminal microorganisms to the tested feedstuffs. During live animal experiments it is usually a practice to have adaptation periods to new feeds of at least 14 days and animals' output is not considered stable in this method (Pellikaan *et al.*, 2011). Results should therefore always be interpreted with care (Storm *et al.*, 2012). Fortunately, the method can easily be applied to many animals making it possible to reduce the standard error of means from experiments. It is possible to determine *in vitro* degradation of the feedstuffs and find if the reduction in methane production is at the cost of total feed degradation. Screening large amounts of feeds and additives is the best application of the *in vitro* method. This method has a large capacity, making it possible to test many different combinations of feedstuffs.

The mathematical models are essential for estimating national or global emissions. They are easy to apply and will give estimates of the average emission of the unit in question. The models are based on experimental data and as such are limited in their application. However, a model based on respiration chamber experiments can therefore not be directly applied to free ranging cattle. Also, our understanding of ruminal digestion is not yet complete. Therefore a continuous need exists for more data to increase our knowledge of this complex system.

CONCLUSION

Many suitable methods for CH₄ measuring are already in use and new ones are being developed. Some, however, are only useful for a particular environment. It is extremely important to compare several methods for accurate assessment. Further research is needed to better understand the CH₄ measurement and evaluation in progressed managements.

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