

**PHYSIOLOGICAL OPPORTUNITY IN SELECTION OF
BREEDING BULLS FOR REPRODUCTION TRAITS**

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Significance of metabolic tests

Due to unilateral selection in breeding of dairy cattle asymmetric genetic progress was achieved in economic traits. Rapid development was made in milk yield per cow. At the same time, however, "secondary" traits did not keep abreast with them even though the former ones might have considerable "primal" economic value in the improvement of factors determining the strength of the animal organism. That is the explanation of the shorter productive life span of cows, decrease of herd life production and reduced reproductive performance in consequence of enhanced overload. The background of the vast majority of cullings might be associated with metabolic disorders, problems of energy mobilization, even though infertility was given as apparent cause for culling. Adverse effect of increased postpartal metabolic mobilization on reproductive performance in dairy cows is well known from literature. Concurrent effect of energy deficit at the early phase of lactation (incongruity of feed consuming capacity of high producing cows and energy requirement of milk production) and aggravating environmental conditions (failures in housing, feeding and management) cannot be compensated by of unstable metabolism. This might be the cause of numerous subclinical and clinical metabolic disorders post partum resulting in decrease of milk yield, reproductive performance and increase of clinical cases, mortality and emergency slaughtering. Having been analyzed the results of relevant investigations Eulitz-Meder and co-workers (1989) stated that genetically the potential milk producing ability might largely be associated with the differences in the endocrine system. Since the selection directed towards high milk yields promotes metabolic processes enhancing glucose production, a precursor in milk synthesis, the capacity and control of gluconeogenesis may play central role. In addition, the whole organism is overloaded by the mammary gland in consequence of deprivation of resources needed for milk synthesis, the fat metabolism due to insufficient glucose supply and energy deficiency, rate of metabolic activity for forming of keton-bodies from fatty acids and ketogenic amino acids increases. Elevated level of keton-bodies leads to ketosis, liver degeneration, decrease of feed consumption and reproductive disorders (Giesecke, 1984). Thus, in complexity undesired association will develop among traits (antagonism of properties) for milk yield, resistance against diseases and fertility.

Physiological principles of reproductive performance and related genetic factors have not been fully cleared yet. For this reason the aim of this study was to promote better understanding of the interactions involved.

Pilot studies were carried out by German researchers (Graf and co-workers, 1981, Roever, 1982, Adam and co-workers, 1985, Feddersen and Kalm, 1985, Feddersen, 1986, Lehmer, 1986, Süphke, 1988, Kalm and Feddersen, 1989, Vollstedt, 1991, etc.) where negative energy balance was stimulated in breeding heifers, bulls and high producing cows by feeding of energy deficient diets. Relation of physiological response to dietetic metabolic load and subsequent reproductive performance of heifers as well as progeny of sires was established in the investigations cited. According to Kalm and Feddersen (1987) reaction ability of metabolism is inherited independent of sex. Consequently it is assumed that the reproduction of progeny can accurately be estimated by test of physiological loading in breeding animals. In spite of small cow herds in FRG you have to take into account marked environmental effects when estimating breeding values for reproduction. In any case reliable correlations were published by the authors cited. Metabolic loading test was elaborated which was applied in progeny testing of potential breeding bulls (Roever, 1982, Feddersen, 1986, Wolter, 1989, Kalm, 1991). Principle of the test involves energy deficient feeding of bulls resulting in similar physiological status as it is the case in dairy cows at the peak of lactation (hunger-test). Apart from the problem outlined above the most harmful factor for the dairy cattle population in this country is the lack of stability of metabolic status of animals (Bozó and co-workers, 1983).

In comparison with any of the West European countries the conditions for the determination of the stability of metabolic status by physio-genetical ways are very favourable in Hungary due to the relatively large herds when taking into account methodological aspects. This prompted our initiative for a comprehensive series of experiments to elucidate the relationship of stability of metabolic status and reproductive performance.

Significance of andrological investigations

1. GnRH test

Extent of secretion of testosterone may differ in several parts of the day. For this reason the so called GnRH test has been developed for different animal species. The quintessence of the test is that testosterone reaction in males to Gonadotrop Releasing Hormone administration shows close relationship to the 24 hours testosterone profile (Post, 1987). This finding allows reliable comparison of testosterone levels in blood samples taken from male animals. In this country Wekerle and co-workers (1989, 1990) described the results of their experiment on GnRH in three species. It has been stated that fertility of sires can be predicted in bulls. Similar results were obtained for boars. It was concluded that the result of the test show characteristic value for a given animal in any part of the day. In addition, seasonal differences were observed in rams, therefore second blood sample should be taken in spring after 2 hours of GnRH administration. This sampling has to be made

1 hour later in autumn. GnRH test was used in rabbits by Gábor and co-workers (1992). The results reveal that the extent of testosterone response is independent of the dose of GnRH and/or body weight of experimental animals. At the same time relationship of β -carotene supply was established to testosterone response. Investigations were directed towards the correlation of weight of testicles and testosterone response to GnRH administration paying attention to the influence on semen quality.

2. Ultrasonic measurement of testicles

Females are given priority at ultrasonic measurements in domestic animals. Nowadays echography has been described as a tool at pathological investigations in males (Traub and co-workers, 1991, Pugh and co-workers, 1990). The workshop led by Prof. Leidl at the International Congress for Reproduction-Biology held in The Hague in 1992 on ultrasonic examinations of sexual organs of males pointed out gaps of this field only and the need for further research in order to make it apt to routine work in praxis. One of our objective was to compare results of GnRH test with ultrasonic pictures of testicles in this study.

MATERIAL AND METHODS

Metabolic loading test have been carried out by Feddersen (1986) and Kalm (1991) can be characterized by two different ways:

- energy deficient feeding for 3 weeks after an adjustment period lasting for 3-4 weeks, —
- sufficient level of feeding for 2 weeks, starvation for 8 days, refeeding for 3 days, resting period for 4 weeks.

In this study we wanted combine advantages of both procedures eliminating the disadvantages. Thus, we applied the procedure as follows:

Adjustment period for 1 week:

1st day - 10% experimental diet+90 % maize meal
2nd-4th day 30-80% experimental diet
from 5th day on - experimental diet

Experimental period for 4 weeks
energy deficient diets

Supplemental feeding without adjustment period.

Apart from the diets bulls were fed barley straw of high quality free choice. The experimental diet contained 3.9 MJ NE_g and 216 g crude protein. This resulted in very low undernutrition. Only partial compensation of this severe starvation could be achieved by the nutrients of barley straw fed ad libitum. Individual live weights of animals were taken prior to the experimental period and after the feeding of energy deficient diets.

During the experiment blood samples were taken at 8 o'clock AM at weekly intervals (7 times). Switching over summer time sampling was made at 9 PM. Glucose, creatinine, FFA, triglyceride, LDH, AST,

ALT, γ -GT, T3 and T4 values were determined in blood serum. At GnRH test 100 μ g GnRH analogue (Ovurelin, Reanal) was administered i.m. 2nd blood samples was taken 90 minutes after GnRH analogue administration. Experimental animals were slaughtered thereafter and weight and volume of testicles were also taken. Findings were compared with testosterone response to GnRH administration. Ultrasonic measurement of testicles was conducted by real time echograph ALOKA attached with 5 Mhz linear transducer.

RESULTS AND DISCUSSION

Only small change in body weight of breeding bulls was recorded during the energy deficient feeding period. Conformation, however, showed some signs of alteration, i.e. musculature of animals became more marked as compared to previous status. This phenomenon seemed to be supported by changes in creatinine, FFA and triglyceride levels in blood serum samples (Table 1.). At the same time Doornenbal and co-workers (1987) stated that creatinine level of serum depends also on testicles, better to say, on production of testosterone, because significantly higher creatinine levels were recorded in serum of bulls than in oxes. Statistical analysis of this experiment seems to reconfirm previous findings. Even though glucose values remained among physiological range (2.3-4.1 nmol/l, Fraser, 1986), levels closely reflect feeding conditions due to energy deficient diets (Table 1.). Intermediate correlation was established for triglyceride of serum. It is assumed that interrelationship reflected by statistical analysis was caused by individual sensivity. This would mean that it could be considered as a valuable parameter in detection of metabolic sensivity of animals. Similar situation was shown for statistical evaluation of FFA values in serum as it was the case for triglyceride. This reveal extended individual variability was recorded in enzymes of the liver (Table 2.) and the hormones of thyroid gland (Table 3.) as it was the case with the metabolites of fat turnover, our hypothesis is considered being reconfirmed. The close correlation between the extent of response in testosterone to GnRH treatment and volume of testicles (Table 4.) suggests determinative significance of structure of testicles in testosterone levels, consequently in semen production. This enables the andrological examinations of young sires and prediction of semen quality. In order to be objective, digitalization of ultrasonic pictures of testicles is planned with computer aided comparison of testosterone response to GnRH test and objective control of semen quality. In case of success of this procedure opportunity for selection of secondary traits increases in combination with metabolic parameters which may contribute to the improvement of the reproductive performance of cattle. This works made by supporting of National Scientific Research Foundation (OTKA 2429 topic).

List of references is available at the authors.

Table 1. Glucose, FFA, triglyceride (TG) and creatinine (CR) levels in blood plasma during the experiment (n=52)

Blood sample	Glucose nmol/l	FFA mmol/l	TG mmol/l	CR μ mol/l
1.	4.10 \pm 0.44	0.06 \pm 0.04	0.17 \pm 0.06	120.2 \pm 24.3
2.	3.97 \pm 0.41	0.21 \pm 0.14	0.13 \pm 0.04	152.9 \pm 24.3
3.	3.90 \pm 0.39	0.21 \pm 0.13	0.10 \pm 0.02	168.8 \pm 29.8
4.	3.47 \pm 0.39	0.18 \pm 0.13	0.13 \pm 0.04	184.7 \pm 28.9
5.	3.23 \pm 0.41	0.23 \pm 0.11	0.15 \pm 0.04	181.3 \pm 29.9
6.	3.32 \pm 0.33	0.24 \pm 0.15	0.12 \pm 0.04	175.2 \pm 23.9
7.	3.60 \pm 0.32	0.05 \pm 0.03	0.10 \pm 0.04	151.3 \pm 20.1

Table 2. Level of enzymes of liver in plasma during the experiment (n=52)

Blood sample	AST NE/l	ALT NE/l	τ -GT NE/l	LDE NE/l
1.	121.1 \pm 55.4	27.3 \pm 8.8	22.6 \pm 5.1	2241 \pm 401.5
2.	114.6 \pm 50.4	28.9 \pm 7.4	24.5 \pm 5.8	2451 \pm 503.5
3.	109.3 \pm 43.5	29.9 \pm 5.9	27.8 \pm 6.6	2316 \pm 261.7
4.	90.5 \pm 24.1	29.6 \pm 5.4	27.3 \pm 5.9	2260 \pm 269.8
5.	96.8 \pm 25.4	29.8 \pm 5.3	30.4 \pm 7.1	2031 \pm 336.7
6.	90.6 \pm 22.5	28.9 \pm 5.6	27.1 \pm 6.2	1999 \pm 364.2
7.	91.1 \pm 23.9	25.0 \pm 3.7	24.6 \pm 4.9	1942 \pm 349.6

Table 3. Level of hormones of thyroid gland in plasma during the experiment (n=52)

Blood sample	T3 ng/ml	T4 ng/ml
1.	0.95±0.19	106.4±54.9
2.	0.85±0.19	119.3±51.1
3.	0.85±0.28	113.8±53.9
4.	0.84±0.23	126.1±46.8
5.	0.71±0.61	112.0±52.2
6.	0.82±0.23	133.0±56.7
7.	0.99±0.27	78.7±24.9

Table 4. Andrological parameters (n=16)

TESTICLES			TESTOSTERONE		SEMEN	
A weight (g)	B volume (cm ³)	C widht (cm)	D after GnRH (nmol/l)	E increase	F average quant. (ml)	G no.cells 10 ⁹ /ml
766.8±115.7	715.7±111.4	7.5±0.6	21.6±8.9	13.0±7.6	4.9±0.7	1.4±0.3
CORRELATION COEFFICIENTS						
A - D	0.59 ***					
A - E	0.37 NS					
B - D	0.63 ***					
B - E	0.44 NS					
D - G	0.40 **					
E - F	0.06 NS					
E - G	0.47 *					

*** p<0.001 ** p<0.01 p<0.05 NS p>0.05