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CYTOGENETIC AND BIOTECHNOLOGICAL ASSESSMENT OF STALLIONS OF TRAKEN AND HANNOVER STUD BREEDS

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The comparative cytogenetic and biotechnological evaluation of the inspected Traken and Hannover stallions by their general chromosomal instability and by the quantitative and qualitative parameters of sperm after thawing have been conducted. The general chromosomal instability of the investigated stallions of Hannover breed was 4,9%, the biotechnological fitness of sperm was 76,19% and the biotechnological fitness of sperm in the stallions of Traken breed was 72,73 and their chromosomal instability was 5,91%. The presence of pair and circular aberrations also influences the biotechnological fitness of sperm of the inspected stallion.

Key words: *stallion, sperm, cytogenetics, biotechnology, selection.*

Statement of the problem. The necessity of the use of genetic evaluation including the cytogenetic one in Ukraine is regulated by the Law of Ukraine “ Breeding policy in animal husbandry” and by the “Regulations of the procedure of genetic evaluation of the origin and manifestation of anomalies in stud animals”. In accordance with the above legislative statements all stud animals must be put on a genetic trial: horses, cattle, pigs, rabbits, poultry and even fish [1]. In this connection special attention should be paid to the cytogenetic and biotechnological evaluation of the breeding stallions to improve the performances of horse reproduction. When the sperm of the above stallion is used in the system of artificial insemination of mares the risk of the chromosome anomaly distribution increases greatly that can lead to great economic losses.

Analysis of main investigations and publications that initiated the problem solution. The selection in horse breeding has been based for a long time on the evaluation of the parameters of sperm quality and on the results of their use in artificial insemination [5]. It has been carried out without any cytological tests that has led to the accumulation of chromosomal defects in the populations and it has reduced the reproductive function of the stud stallion and mares and as a result it has reduced the output of foals to the critical level. [2]. The development of chromosomes in farm animals [2,3] and the

understanding of the role of cytogenetics in the solution of the selectional problems in animal husbandry [1] gave the possibility to introduce the cytogenetic methods into horse breeding.

Aim and task of investigation. The aim of the study was to carry out the cytogenetic and biotechnological estimation of the stud stallions of Traken and Hannover breeds by their general chromosomal instability and by the quantitative and qualitative parameters of sperm after thawing.

The tasks of the study were to make the assessment of the sperm quality after thawing in stud stallions of Traken and Hannover breeds and to make the cytogenetic evaluation of the stallions under investigation by their structural aberrations of chromosomes.

Methods of investigation. Three stud stallions of Hannover breed, one stallion of Traken breed of Kharkiv stud-farm, Kharkiv region and three stallions of Traken breed of Chutivskyy stud-farm "Traken", Poltava region have been taken for the experiment. The stallion semen collection and cryoconservation have been carried out by Kharkiv technology that was developed by the department of animal reproduction biotechnology of the Institute of animal science, National Academy of Agricultural Science of Ukraine [3]. The preparations of stallion chromosomes have been made by common methods [4]. During the analysis of the chromosomal preparations from every stallion the general chromosomal instability; the general amount of aberrations; the number of aberrations per a hundred of cells; absolute and relative number of single, pair and circular aberrations have been registered. [5]. Statistical processing of the results of have been done with the use of common methods [6].

Results of investigations. The evaluation of the quality of the deconserved semen of Traken and Hannover stallions of breeds has been carried out (Table 1). Among the stallions of Hannover breed Sandros-Diamond had the best semen fit for breeding that has been proved by the highest percent of cryoresistant semen (by 14,29 % more than Monaco and by 57,14 % more than Marcusee). The highest sperm motility after deconservation was in Sandros-Diamond and Marcusee, it was 1,17 points as high as Monaco. Sandros-Diamond had the best sperm viability that is by 0,85 hours more than Monaco and by 1,6 hour ($p < 0,05$) than Marcusee. Monaco had the highest rate of sperm duration, it was by 1,3 % higher than Sandros-Diamond and by 8,49 % than Marcusee.

1. Parameters of sperm after thawing in stud stallion of Hannover and Traken breeds

Stud stallion	Amount of sperm	Amount of cryoconserved semen	Cryoresistance, %	Indices of sperm after thawing (M±m)		
				Sperm motility, points	Sperm viability at 37°C, hours	Sperm durability, %
Hannover breed						
Sandros-Diamond	7	7	100,00	3,50 ±0,22	4,10 ±0,33	53,86 ±1,62
Monaco	7	6	85,71	3,50 ±0,16	3,25 ±0,28	55,16 ±1,74

Marcusee	7	3	42,86	2,33±0,17* *	2,50 ±0,29*	46,67 ±3,33
Total	21	16	76,19	3,23±0,18	3,39 ±0,24	52,70 ±1,45
Traken breed						
Tron	7	6	85,71	3,17 ±0,19	2,94 ±0,10	58,33 ±3,38
Peon	5	3	60,00	2,20 ±0,34	2,50 ±0,39	39,38 ±6,88
Arlean	5	4	80,00	2,40 ±0,51	2,10 ±0,46	43,66 ±8,57
Prospect	5	3	60,00	2,50 ±0,27	2,10 ±0,43	44,34 ±3,51
Total	22	16	72,73	2,67 ±0,17	2,50 ±0,16	48,41±3,00

Note: * - $p < 0,05$, ** - $p < 0,01$.

Among the stud stallions of Traken breed the highest percent of biotechnological fitness of semen was revealed in Tron that was by 5,71 % higher than in Arlean and by 25,71 % higher than in Peon and Prospect. The highest motility of sperm after thawing was in Tron that was by 0,97 points more than in Peon and by 0,77 points more than in Arlean and by 0,67 points more than in Prospect. Tron had the highest index of sperm viability that was by 0,44 hours more than Peon had, by 0,84 hours more than Arlean and Prospect had. Sperm durability rate was the best in Tron: that was by 18,95 % higher than in Peon, by 14,67 % higher than in Arlean and by 13,99 % more than the sperm durability in Prospect.

The next stage of the investigation was to make the cytogenetic evaluation of the stallions under investigation by their structural aberrations of chromosomes. The results of the cytogenetic evaluation of the stud stallions of Hannover and Traken breeds by their structural aberrations of chromosomes are presented in Table 2.

It can be seen from Table 2 that Marcusee and Prospect had the greatest amount of metaphases with aberrations that was 1 phase more than Arlean had, by 0,5 metaphase more than Peon and Tron had, by 2 metaphases more than Sandros-Diamond and Monaco had ($p < 0,05$). The stud stallions Sandros-Diamond and Monaco showed the standard level of general chromosomal instability. The stud stallions Marcusee, Tron, Peon, Arlean and Prospect had the higher level of general chromosomal instability by 2,05 %, 1,5 %, 0,9 %, 0,5 % and by 0,75 %, respectively.

Sandros-Diamond had the lowest index of general quantity of aberrations, by 1 aberration less than Monaco had, by 5 aberrations less ($p < 0,05$) than Marcusee and Arlean had, by 2 aberrations less than Tron had, by 4,5 aberrations less ($p < 0,05$) than Peon had, by 4,5 aberrations less ($p < 0,05$) as compared to Prospect. In the calculation per 100 cells Sandros-Diamond had the lowest quantity of aberrations that was by 1,95($p < 0,05$), 8,45; 3,7; 7,55; 8,85 less than Monaco, Marcusee, Tron, Peon and Arlean ($p < 0,01$) had, respectively and by 5,5 aberrations more than Prospect had ($p < 0,05$).

The quantity of single aberrations among stud stallions of Hannover and Traken breeds was at the similar level. Sandros – Diamond had the highest percent of single aberrations that was by 33,3 %, 85,4

% ($p < 0,001$), 53,35 % , 61,9 % ($p < 0,01$), 70,85 % ($p < 0,01$) and by 61,9 % ($p < 0,01$) more than Monaco, Marcusee, Tron, Peon, Arlean and Prospect had, respectively.

The amount of pair aberrations among the stud stallions of Hannover and Traken breeds was not more than two; pair aberrations were not revealed only in Sandro – Diamond. Monaco had the highest percent of pair aberrations that was more than Marcusee, Tron, Peon, Arlean and Prospect had by 16,65%, 6,65% , 3,5% , 12,45% and by 2,35% respectively.

2. Structural aberrations of chromosomes of stud stallions of Hannover and Traken breeds ($M \pm m$)

Stud stallion	Number of metaphases	Metaphases with aberrations <i>n</i>	Aberrations		per 100 cells	single	pair	circular		<i>n</i>	%	<i>n</i>	%
			total	%				<i>n</i>	%				
			Hannover breed										
Sandro-Diamond	105	2,00 $\pm 0,00$	3,82 $\pm 0,18$	2,00 $\pm 0,00$	3,80 $\pm 0,20$	2,00 $\pm 0,00$	100,0 $\pm 0,00$	0,00 $\pm 0,00$	0,00 0	0,00 0	0,00 0	0,00 0	0,00 0
Monaco	105	2,00 $\pm 0,00$	3,82 $\pm 0,18$	3,00 $\pm 0,00$	5,75 $\pm 0,25^*$	2,00 $\pm 0,00$	66,70 $\pm 0,00$	1,00 $\pm 0,00$	33,3 0 $\pm 0,0$ 0	0,00 0 $\pm 0,0$ 0	0,00 0 $\pm 0,0$ 0	0,00 0 $\pm 0,0$ 0	0,00 0 $\pm 0,0$ 0
Marcusee	115	4,00 $\pm 1,00$	7,05 $\pm 2,05$	7,00 $\pm 1,00$ *	12,25 $\pm 2,25$	1,00 $\pm 0,00$	14,60 $\pm 2,10^{***}$	1,00 $\pm 1,41$	16,6 5 $\pm 23,$ 6	5,00 0 $\pm 2,0$ 0	68,7 5 $\pm 18,$ 8	5 0 $\pm 18,$ 8	5 0 $\pm 18,$ 8
Total	325	2,67 $\pm 0,49$	4,90 $\pm 0,86$	4,00 $\pm 1,00$	7,27 $\pm 1,72$	1,67 $\pm 0,21$	60,43 $\pm 15,73$	0,67 $\pm 0,47$	16,65 $\pm 10,$ 53	1,67 $\pm 2,0$ 3	22,9 2 $\pm 26,$ 47	2 0 $\pm 26,$ 47	2 0 $\pm 26,$ 47
Traken breed													
Tron	110	3,50 $\pm 0,50$	6,50 $\pm 1,50$	4,00 $\pm 1,00$	7,50 $\pm 2,50$	2,00 $\pm 1,00$	46,65 $\pm 13,4$	1,00 $\pm 0,00$	26,6 5 $\pm 6,6$ 5	1,00 0 $\pm 0,0$ 0	26,6 5 $\pm 6,6$ 5	1,00 0 $\pm 0,0$ 0	26,6 5 $\pm 6,6$ 5
Peon	119	3,50 $\pm 0,50$	5,90 $\pm 0,10^{**}$	6,50 $\pm 0,50$ *	11,35 $\pm 2,65$	2,50 $\pm 0,50$	38,10 $\pm 4,80^{**}$	2,00 $\pm 1,00$	29,8 0 $\pm 13,$ 1	2,00 0 $\pm 1,0$ 0	32,1 5 $\pm 17,$ 9	2,00 0 $\pm 1,0$ 0	32,1 5 $\pm 17,$ 9
Arlean	110	3,00 $\pm 0,00$	5,50 $\pm 0,50$	7,00 $\pm 1,00$ *	12,65 $\pm 0,65^{**}$	2,00 $\pm 0,00$	29,15 $\pm 4,15^{**}$	1,50 $\pm 0,50$	20,8 5 $\pm 4,1$ 5	3,50 0 $\pm 0,5$ 0	50,0 0 $\pm 0,0$ 0	3,50 0 $\pm 0,5$ 0	50,0 0 $\pm 0,0$ 0
Prospect	140	4,00 $\pm 0,00^*$	5,75 $\pm 0,25^*$	6,50 $\pm 0,50$ *	9,30 $\pm 1,10^*$	2,50 $\pm 0,50$	38,10 $\pm 4,80^{**}$	2,00 $\pm 0,00$	30,9 5 $\pm 2,3$ 5	2,00 0 $\pm 0,0$ 0	30,9 5 $\pm 2,3$ 5	2,00 0 $\pm 0,0$ 0	30,9 5 $\pm 2,3$ 5
Total	479	3,50 $\pm 0,19$	5,91 $\pm 0,33$	6,00 $\pm 0,53$	10,20 $\pm 1,04$	2,25 $\pm 0,25$	38,00 $\pm 3,75$	1,63 $\pm 0,26$	27,06 $\pm 3,2$ 7	2,13 0 $\pm 0,4$ 0	34,9 4 $\pm 4,9$ 6	2,13 0 $\pm 0,4$ 0	34,9 4 $\pm 4,9$ 6

Note. * - $p < 0,05$; ** - $p < 0,01$; *** - $p < 0,001$

No circular aberrations have been found out in Sandros – Diamond. and Monaco. Marcusee had the largest number of circular aberrations that was by 4; 3; 3 and 1,5 aberrations more than Tron, Peon, Prospect and Arlean had, respectively. Marcusee had the highest percent of circular aberrations that was by 42,1 % , 36,6 % , 37,8 % and by 18,75 % more than Tron, Peon, Prospect and Arlean had, respectively.

Conclusion:

1. It has been found out that the inspected stallions of Hannover breed had the average level of general chromosomal instability – 4,9 % and they had the average biotechnological fitness of sperm - 76,19 %. The stallions of Traken breed had the higher level of general chromosomal instability –5,91 % and the level of the biotechnological fitness of sperm was 72,73 %.

2. It has been proved that the relative amount of single aberrations in the inspected stallions of Hannover breed was higher by 22,43%, the number of circular aberrations was less by 10,41 and the quantity of circular aberrations was by 11,82% less than in Traken stallions.

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