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Hormonal stimulation of American mink (*Neovison vison*) females during mating improves reproduction parameters

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Abstract: Hormonal stimulation of American mink (Neovison vison) females during mating improves reproduction parameters. The aim of the study was to determine the effect of hormonal stimulation during the mating season on reproduction performance in American mink females and a selection of both the hormonal formulation and its dosage. The study involved one-year-old females of American mink (Neovison vison) of the pearl color morph. The females received one of two hormonal formulations, F1 or F2, with a single injection administered 24 h before planned mating. The formulations contained: (F1) a pituitary gonadotropin releasing hormone analogue, and (F2) freeze-dried, crystalline serum gonadotropin (PMSG) of a strong FSH- and LH-like effect. Each formulation was administered in three dosage levels. The hormonal stimulation applied 24 h before the planned mating significantly reduced the number of sterile females, also brought a significant increase in the average litter size, from 1.05 to 1.35 kits, and in the average number of live births, from 1.03 to 1.98. Of the two formulations tested, the analogue of pituitary gonadotropin releasing hormone (F1) at a dose of 36 IU proved to be the most effective; its application allowed attaining the highest reproductive parameters. Using hormonal stimulation of the female mink during the mating can be effectively put into practice on the farm and may in consequence improve the profitability of the production cycle.

Key words: American mink, reproduction, hormonal stimulation

INTRODUCTION

Some of the most important elements affecting the efficiency of animal farming, irrespective of the species, include prolificacy, fertility and reproductive performance. In the mink, these traits are highly variable and depending on the individual genotype. Many authors (Franklin 1958, Møller 2000, Bielański et al. 2003, Socha et al. 2003, Święcicka 2004, Kołodziejczyk and Socha 2006, Ślaska et al. 2009, Felska-Błaszczyk et al. 2010, Nieminen et al. 2010) have observed that both gestational length and the outcomes of whelping and nursing vary greatly depending on the color variety of females. As soon as in 1968, this fact was reported by Bowness (1968), who found that new color varieties of mink females had on average 1.15-day longer gestation length in relation to primary-color varieties. Reproduction in mink is a complex, multidimensional trait, and its monoestrous character results in a fact that the breeder only once a year is able to carry out mating and obtain offspring. Taking into account

the above aspects and the fact that, as indicated by Kołodziejczyk and Socha (2006), reproduction is the most difficult step in the whole mink production cycle (Lagerkvist et al. 1993), it seems appropriate to strive to find effective ways to improve its performance.

For many years, research has been carried out on improving fertility in mink. One of the factors used to improve fertility mink is, for example, extending the light regime during their pregnancy (Felska-Błaszczyk et al. 2013), which shortens gestation length and increase fertility. Another factor that may boost the breeding success may be hormonal stimulation of both males and females applied prior to and during the heat. Lasota et al. (2013) administered chorionic gonadotropin, hCG, to American mink males 24 h prior to mating, which increased libido in males and, in consequence, improved conception rates. In experiments by Klotchkov and Eryuchenkov (2000, 2003), hCG was administered to mink females during the pro-estrus, which significantly improved their fertility and fecundity; a higher number of mature graffian follicles were observed in the treated females as compared with the control. On mink farms in Poland, no hormonal treatment, in order to improve the fertility of the females, has been applied during mating so far.

Therefore, the aim of this study was to determine the effect of hormonal stimulation during mating on the reproductive performance of female mink and to select both formulation and its dosage.

MATERIAL AND METHODS

The experiment was carried out on a mink farm located in western Poland during the breeding season. Animals were kept on a farm in multipurpose, double-row sheds, in Danish-type wire cages. Feeding followed best feeding standards and the semi-liquid feed was based on chicken and fish.

The material comprised one-year-old female American mink (*Neovison vison*) of the pearl color morph. The females were treated with a single injection, applied 24 h prior to mating, of one of the following hormonal formulations:

- F1, a pituitary gonadotropin releasing hormone analogue;
- F2, freeze-dried, crystalline serum gonadotropin (PMSG) of a strong FSH-like effect and additional LH--like effect.

The animals were mated according to the scheme 1 + 7 + 8, the numbers being the subsequent days in heat, starting between 5 and 10 March. Due to varied effects of each of the hormones, we applied different doses of the formulations. Table 1 presents six treatment groups (plus control) which were formed for the experiment. The control group was composed of randomly selected females of the same age, which were mated in a similar way as the treatment females.

The following parameters were studied: percent of barren females, mean litter size, mean number of live-born per litter, total gestation length (from the first mating date to delivery).

group				
Hormonal formula- tion	Hor- monal group	Group	Total number of females	Injection dose (IU)
Formula- tion 1	F1	Α	26	36
	F1	В	23	32
	F1	С	30	28
Formula- tion 2	F2	Α	20	9
	F2	В	35	8
	F2	С	16	7
Control (C)	control	control	71	_

TABLE 1. Doses of the hormonal treatment by group

Statistical analysis of data was carried out using the Statistica 10 PL package. The following descriptive statistics were included: arithmetic mean and standard deviation (SD). Testing the differences was accomplished with the non-parametric Mann–Whitney U test for two independent samples (treatment and control groups) and the Kruskal–Wallis test for two or more variables (to compare more than two samples).

RESULTS

In the experiment, we applied three doses of either of the tested hormonal formulations. The analysis showed that the lowest percentage of barren females (7.69%) was found in the group of females that had been administered F1 at the dose A, i.e. 36 IU. In the remaining groups, the percentage was higher, from 8.7%, after application of F1 at the B dose of 32 IU, to as high as nearly 19%, after application of F2 at the dose C, 7 IU (Table 2).

The analysis of female reproduction performance revealed that the lowest rate of barren females, slightly exceeding 10%, was observed in the group that

TABLE 2. Percentage of barren mink females in relation to different hormonal formulations and their doses

Factor	Level of factor	Number of whelping females	Number of barren females	Total number of females	Percentage of barren females
Control (C)		61	10	71	14.08
Hormone	Formulation 1 (F1)	71	8	79	10.13
	Formulation 2 (F2)	62	9	71	12.68
Dose	Formulation 1 36IU (F1, group A)	24	2	26	7.69
	Formulation 1 32IU (F1, group B)	21	2	23	8.70
	Formulation 1 28IU (F1, group C)	26	4	30	13.33
	Formulation 2 9IU (F2, group A)	18	2	20	10.00
	Formulation 2 8IU (F2, group B)	31	4	35	11.43
	Formulation 2 7IU (F2, group C)	13	3	16	18.75

had received F1, whereas the highest rate was found in the control group, in which the value exceeded 14%.

As far as litter sizes are concerned (Table 3), the largest litters, reaching 7.69 kits, were produced by females treated with F1. The value differed significantly (at P < 0.01) from those attained in the control group (6.34).

Like in the studies on fertility, the application of the two formulations, F1 and F2, in three different doses revealed superiourity of the formulation F1 at the dose A. The mean numbers of total born and live-born kits per litter, 8.37 and 7.29, respectively, observed in females treated with the F1 at the dose A, were significantly (P < 0.01) higher in relation

TABLE 3. Mean litter size at birth and live-born kits in relation to different hormonal formulations and their doses

Factor	Level of factor	Total born kits			Live-born kits		
racioi		n	AVG	SD	п	AVG	SD
Control (C)		61	6.34 ^{Aa}	1.9	61	4.92 ^{ABa}	2.62
Hormone	Formulation 1 (F1)	71	7.69 ^A	1.82	71	6.90 ^A	1.99
	Formulation 2 (F2)	62	7.39ª	2.59	62	6.5 ^B	2.49
Dose	Formulation 1 36IU (F1, group A)	24	8.37 ^A	1.58	24	7.29 ^A	1.94
	Formulation 1 32IU (F1, group B)	21	7.63	1.98	21	6.84	1.64
	Formulation 1 28IU (F1, group C)	26	7.14	1.76	26	6.61ª	2.23
	Formulation 2 9IU (F2, group A)	18	7.06	2.96	18	6.17	2.77
	Formulation 2 8IU (F2, group B)	31	7.39	2.32	31	6.52	2.11
	Formulation 2 7IU (F2, group C)	13	7.85	2.79	13	6.92	3.04

Values in the same row marked with the same letters in columns differ significantly ^{AA, BB} at $P \leq 0.01$; ^{aa} at $P \leq 0.05$.

A similar relationship was observed with in the average number of live-born litter size in the first year of the study. Here, too, the highest value of this parameter was attained by females that 24 h before mating had received the formulation F1, 6.9 live-born kits per female on average. This result differed significantly (P < 0.01) from the control group, with 4.92 kits per female. to the control, in which the respective values were 6.34 and 4.92.

Given the type of the administered hormone formulation, the longest average length of gestation (54.82 days) was observed in the group of females treated with an injection of formulation F2. This result was significantly (P < 0.01) higher than the value of said characteristic (52.26 days) attained by the females of the control group (Table 4). Gestation

lengths in both treated and control females ranged between 45 and 55 days, and obtained litter sizes, both in terms of total born and live-born kits were satisfactory.

TABLE 4. Gestation length in relation to different hormonal formulations and their doses

Factor	Level of	Gestation length		
Factor	factor	п	AVG	SD
Control (C)		61	52.26 ^{AB}	3.20
Hormone	Formulation 1 (F1)	71	53.61	4.01
	Formulation 2 (F2)	62	54.82 ^A	4.33
Dose	Formulation 1 36IU (F1, group A)	24	54.04	4.24
	Formulation 1 32IU (F1, group B)	21	51.95ª	2.17
	Formulation 1 28IU (F1, group C)	26	54.36	4.53
	Formulation 2 9IU (F2, group A)	18	54.22	4.19
	Formulation 2 8IU (F2, group B)	31	55.48 ^{Ba}	4.95
	Formulation 2 7IU (F2, group C)	13	54.08	2.60

Values in the same row marked with the same letters differ significantly ^{AA, BB} at $P \leq 0.01$; ^{aa} at $P \leq 0.05$.

An analysis of the effect of treatment on gestation length reveales that the shortest gestations (51.95 days) were characteristic of females receiving F1B, whereas the longest (55.48 days) was observed in those treated with F2B, with the differences significant at P < 0.05.

DISCUSSION

Franklin (1958) was probably the first who attempted to improve American mink females reproduction performance by injectable progesterone. Less than a decade later, Holcomb (1967) published the results of his experiments on an application of hCG, which were, however, unsatisfactory. According to the latter author, the control females - which did not receive hCG after mating – whelped in higher numbers and produced larger litters. The author also noticed that most treated females lost their litters within two days from parturition, which probably resulted from lack of lactation in the studied dams.

Positive results of an experiment on female hCG stimulation were attained by Adams (1981), who suggests the possibility of hCG-induced ovulation also in females that failed to mate. These observations, according to the author, may have resulted from a behavioural problem rather than ovary-level malfunctioning. This was later confirmed by Wehrenberg et al. (1992), who attained positive outcomes of eCG and hCG administration to females avoiding mating; by administering a dose of 100 IU of eCG twice, the authors obtained the highest number of pregnant and whelping females, as well as the highest mean litter size. Also Klotchkov and Eryuchenkov (2000, 2003) and Klotchkov et al. (2005) successfully applied hCG.

Application of GnRH, on the other hand, was sudied by, among others, Douglas et al. (1994) and Bäcklin et al.

(1997). Seremak et al. (2010) studied the effects of a synthetic analogue of the hormone in females that were unmated before 19 March. The authors suggest that the effects could be positive, as the fertility after hormonal treatment was at a level of 46.6%, as compared to control (42%).

Desirable outcomes of another stimulus, medroxyprogesterone acetate (MPA), were reported by Concannon et al. (1980). The authors observed that embryonic implantation in the uterine wall of exogenous progesterone-treated female mink took place sooner, and, additionally, the females produced larger litters. Similar studies on skunk, however, did not bring good results (Mead et al. 1981). Also the effects of inhibin (Ireland et al. 1992) and phytoestrogens (Ryökkynen et al. 2005) were studied. Murphy (1983) observed that hormonally treated American mink females had the diapause by 10 days shorter.

CONCLUSIONS

The use of hormonal stimulation 24 h before the planned mating significantly reduced the number of barren females, also resulted in a significant increase in litter sizes, both in the average number of total born and live born kits, from 1.05 to 1.35 and from 1.03 to 1.98 kits, respectively. Of the two formulations tested, the analogue of pituitary gonadotropin releasing hormone (F1) at a dose of 36 IU (A) proved to be the most effective; after its application, the females demonstrated the best reproductive parameters, without gestation length reduced.

Hormonal stimulation of female mink during matings can be successfully applied in the production on the farm, eventually contributing to the success and increased profitability of the production cycle.

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Streszczenie: Hormonalna stymulacja samic norki amerykańskiej (Neovison vison) w trakcie kryć źródłem poprawy parametrów reprodukcyjnych. Celem podjętych badań było określenie wpływu stymulacji hormonalnej na wyniki rozrodu samic norki amerykańskiej poprzez wybór preparatu i zastosowanej dawki. Materiał do badań stanowiły jednoroczne samice norki amerykańskiej (Neovison vison) odmiany barwnej perła. Samicom podawano w postaci jednorazowej iniekcji na 24 h przed planowanym kryciem dwa preparaty hormonalne: (P1) - analog hormonu uwalniającego gonadotropinę przysadkową, (P2) - liofilizowana, krystaliczna substancja zawierającą surowiczą gonadotropinę (PMSG), wykazująca silne działanie głównie o charakterze FSH oraz dodatkowo LH. W doświadczeniu zastosowano trzy różne dawki każdego z dwóch testowanych preparatów hormonalnych. Zastosowanie stymulacji hormonalnej na 24 h przed planowanym kryciem znacząco wpłynęło na spadek liczby jałowych samic, przyniosło także istotny wzrost średniej liczby urodzonych od 1,05 do 1,35 oraz średniej liczby żywo urodzonych od 1,03 do 1,98 norcząt w grupach doświadczalnych. Spośród

testowanych dwóch preparatów hormonalnych najskuteczniejszy okazał się preparat P1 w dawce (36 IU), po zastosowaniu którego uzyskano największe z analizowanych parametry rozrodu. Zastosowanie stymulacji hormonalnej samic norki amerykańskiej podczas okresu kryć z powodzeniem można wprowadzić do praktyki hodowlanej w cyklu produkcyjnym na fermie, a w konsekwencji może to realnie wpłynąć na sukces i opłacalność prowadzonej hodowli.

Słowa kluczowe: norka amerykańska, rozród, stymulacja hormonalna

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