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Beata SEREMAK, Kamil PŁAWSKI, Lidia FELSKA-BŁASZCZYK[1], Bogdan LASOTA

HUMAN CHORIONIC GONADOTROPIN (hCG) APPLIED INSTEAD OF FIRST MATING IMPROVES REPRODUCTION IN MINK (*NEOVISON VISON*)

ZASTOSOWANIE GONADOTROPINY KOSMÓWKOWEJ hCG W MIEJSCE PIERWSZEGO KRYCIA NORKI AMERYKAŃSKIEJ (*NEOVISON VISON)* W CELU POPRAWY WYNIKÓW ROZRODU

Department of Biotechnology of Animal Reproduction and Environmental Hygiene
West Pomeranian University of Technology, Szczecin, Poland
[1]Laboratory of Animal Anatomy, West Pomeranian University of Technology, Szczecin, Poland

Streszczenie. Celem badań było określenie, w jaki sposób podanie preparatu hormonalnego, zawierającego gonadotropinę kosmówkową (hCG), w miejsce pierwszego krycia wpłynie na wskaźniki plenności samic norki amerykańskiej (*Neovison vison*). Badania zostały przeprowadzone w dwóch następujących po sobie sezonach rozrodczych. Materiał badawczy stanowiły roczne i dwuletnie samice norki amerykańskiej, o wyrównanej masie ciała, należące do trzech odmian barwnych: szafir, standard brązowy (typu Wild), mahoń. Grupę doświadczalną stanowiły samice, którym podano w formie iniekcji 20 j.m. hCG zamiast pierwszego krycia. Samice kryto ósmego dnia po podaniu preparatu hormonalnego, z kryciem powtórnym następnego dnia według schematu: 8 + 9. Grupę kontrolną stanowiły samice, którym nie podano preparatu hormonalnego. Samice z grupy kontrolnej kryte były w sposób tradycyjny, trzykrotnie, według schematu 1 + 8 + 9 (pierwszy dzień, ósmy dzień, dziewiąty dzień). W wyniku przeprowadzonych badań stwierdzono statystycznie istotne na korzyść grupy doświadczalnej (stymulowanej hormonalnie), różnice w ilości urodzonych i urodzonych żywych młodych w obrębie odmiany barwnej mahoń, w dwóch sezonach rozrodczych, natomiast w przypadku odmian standard brązowy oraz szafir w drugim roku doświadczalnym. Powyższe wyniki świadczą o korzystnym wpływie zastosowanej stymulacji na plenność samic i wskazują na możliwość zastosowania jej w praktyce hodowlanej.

Key words: fertility, hormonal stimulation, reproduction.
Słowa kluczowe: płodność, stymulacja hormonalna, reprodukcja.

INTRODUCTION

Intensive development of fur-bearing animal farming in Poland has led to the situation that mink farming is now the leading branch of agricultural production and Polish farms are one of the largest in the world. Breeder efforts to achieve satisfactory economic outcomes is inherently linked to the necessity of improving reproductive performance (Dziadosz et al. 2010; Seremak et al. 2011), which is an important factor affecting the profitability of breeding and the overall efficiency of the farm.

Corresponding author – Adres do korespondencji: PhD Beata Seremak, Department of Biotechnology of Animal Reproduction and Environmental Hygiene, West Pomeranian University of Technology, Szczecin, Doktora Judyma 6, 71-466 Szczecin, Poland, e-mail: beata.seremak@zut.edu.pl

American mink belongs to mammals with a mono-oestrous reproductive cycle. Oestrus is dependent on the photoperiod, which affects the endocrine function and triggers the onset of the breeding season (Douglas et al. 1998; Felska-Błaszczyk et al. 2013; Skuratko et al. 2013). Under Central European climate conditions, heat in the American mink occurs in March, when the day is 10 hours long. The photoperiod stimulates the gonads on a neurohormonal pathway (retina – pineal gland – pituitary gland) and leads to oocyte maturation (Pilbeam et al. 1979; Wehrenberg et al. 1992). The ova maturate in two to three sub-cycles within the oestrus, hence multiple (2 to 4) matings are possible.

The reproductive capability of female American mink is a complex trait, formed by both genetic and environmental factors (Klotchkov et al. 1998; Kołodziejczyk and Socha 2006; Maciejowski and Jeżewska 1993; Seremak et al. 2011). The sophisticated reproduction process which involves the diapause, large fluctuations in litter sizes (from 1 to 15, or more, kits per litter), and frequent female infertility, all make the mating season the most difficult period of the entire production cycle (Lagerkvist et al. 1993). Therefore, it is advisable to find effective ways to improve reproductive performance, also through biotechnical methods. One such method may be hormonal stimulation (Seremak et al. 2011). Many authors reported positive results of hormonal stimulation of ovulation in mink using chorionic gonadotropin, hCG (Klotchkov and Eryuchenkov 2003; Rietveld 1978) which effectively increased litter sizes. Chorionic gonadotropin can also be used as libido stimulation in male mink, thus improving the efficacy of mating (Lasota et al. 2013). In animal production, the use of hormonal stimulation is a common practice; however, in fur-bearing animals, and especially in mink, such procedures have not been applied, at least on Polish farms.

The aim of the study was to determine how the administration of a hormonal specific containing human chorionic gonadotropin (hCG) instead of the first mating affects the level of selected reproductive parameters in female American mink (*Neovison vison*).

MATERIAL AND METHODS

The experiment was conducted on a mink farm located in northern Poland. The animals were fed semi-liquid feed based on poultry offals and fish, according to the adopted standards. Prior to the breeding season, the animals were arranged in breeding sets, 48 animals each, 40 females and 8 males. The animals were mated within their set throughout the season. The research was conducted in two consecutive breeding seasons. The material consisted of one- and two-year-old female American mink, similar in body weight, belonging to the following colour morphs: Sapphire (S), Standard Brown – Wild type (SBW), and Mahogany (M). The females were assigned to the experimental and the control group (Table 1).

The experimental group (H) included females which were administered an injection of 20 IU hCG in place of the first mating. The females were mated on the eighth day after the administration of the hormone, the mating being repeated the following day, according to the scheme: (8 + 9). The control group (C) were females that had not been given the hormonal preparation. Control females were mated in the conventional way three times, according to scheme 1 + 8 + 9 (the first, the eighth, and the ninth day).

Table 1. The number of animals of each color variety participating in the experiment
Tabela 1. Liczba zwierząt poszczególnych odmian barwnych biorących udział w doświadczeniu

Color variety Odmiana barwna	1st year of experiment 1. rok doświadczenia		2nd year of experiment 2. rok doświadczenia	
	experimental group grupa doświadczalna (H)	control group grupa kontrolna (C)	experimental group grupa doświadczalna (H)	control group grupa kontrolna (C)
Standard Brown – Wild type Standard brązowy – typ wild (SBW)	228	252	189	207
Mahogany Mahoń (M)	255	280	192	196
Sapphire Szafir (S)	228	272	126	165
Total Razem	711	804	507	568

The preparation used for the hormonal stimulation was Chorulon (Farmvet International B.V.). The drug contains human chorionic gonadotropin, hCG, and is intended for veterinary use. Chorulon affects follicular maturation, ovulation and the emergence and development of the corpus luteum.

The resulting data sets were statistically analysed using STATISTICA 10 PL. Statistical analysis was based on descriptive statistics of the studied reproduction parameters, i.e. arithmetic means of the variables and their standard errors. For the number of born and live-born kits, we used the non-parametric Mann-Whitney U test for two independent samples (experimental and control group); for non-whelping females, we used Pearson's chi^2 test.

RESULTS

The percentage of females that did not give birth ranged from 5.81 to 20.40%. In the second year of the experiment, the differences in this parameter between the treatment (H) and control (C) groups, in favour of the previous group, were significant within SBW and Mahogany females. There are also differences in the average litter size at birth, as well as the number of live-born per litter, in favour of the hormonal treatment group (Table 2).

The highest number of born and live-born young in the first year of the experiment was noted for Mahogany females. On average, Mahogany females attained 6.80 born and 6.33 live-born kits per litter. In the control group, which had not received hormone, these values were respectively 5.88 and 5.62 kits. These differences were in each case statistically significant at $p < 0.01$. Within the other colours, the differences in the respective parameters between H and C groups were non-significant. In the case of SBW females, the mean litter size in the experimental group and the control group was 5.48 and 5.16 kits, respectively. In contrast, the average number of live-born per litter was 5.24 in the experimental group and 4.90 in the control group. For Sapphires, the results are as follows: In the experimental group the average number of born was 5.46, and the live-born 5.25; in the control group 5.11 and 5.44, respectively.

Table 2. The mean (and SD) number of born and live-born kits of experimental and control group females by color variety in the first and second year of experiment
Tabela 2. Średnia liczba urodzonych młodych oraz urodzonych żywych młodych dla samic z grup doświadczalnej i kontrolnej u poszczególnych odmian barwnych w pierwszym i drugim roku doświadczenia

Year Rok	Colour variety Odmiana barwna	Group Grupa	Number of whelping females Liczba samic wykoconych	Percentage of non-whelping females Procent samic niewykoconych	Number of kits born Liczba urodzonych norek		Number of live-born kits Liczba norek urodzonych żywych	
				%	mean średnia	SD	mean średnia	SD
1	SBW	H	228	7.89	5.48	0.17	5.24	0.19
		C	252	7.02	5.16	0.16	4.90	0.18
	M	H	255	18.82	6.80 A	0.17	6.33 A	0.18
		C	280	18.82	5.88 B	0.17	5.62 B	0.18
	S	H	228	16.23	5.46	0.19	5.25	0.20
		C	272	17.54	5.44	0.18	5.11	0.19
2	SBW	H	189	9.57 A	6.71 a	0.13	6.08 a	0.12
		C	207	14.53 B	6.34 b	0.15	5.69 b	0.16
	M	H	192	5.81 A	7.41 A	0.12	6.85 A	0.12
		C	196	14.83 B	6.78 B	0.16	6.15 B	0.16
	S	H	126	19.64	6.34 a	0.13	6.05 A	0.14
		C	165	20.40	5.74 b	0.21	5.29 B	0.21

[a, b] Column means with different superscripts differ significantly at $P < 0.05$ – Średnie w kolumnach różnią się istotnie przy $P < 0,05$.
[A, B] Column means with different superscripts differ significantly at $P < 0.01$ – Średnie w kolumnach różnią się istotnie przy $P < 0,01$.

In the second year of the experiment, in each studied colour variety, differences in favour of the treatment group were noted in the number of born and live-born kits.

The highest prolificacy in the second year of research were noted for hormone-treated Mahogany females, with the average of 7.41 and 6.85 born and live-born kits, respectively. In the control group of this variety, the average number of born and live-born kits was much lower, 6.78 and 6.15, respectively, the results being statistically significant at $p < 0.01$. In the remaining colours, the average number of born and live-born kits were lower. In the case of Standard Brown, the number of born kits was 6.71 in the experimental group and 6.34 in the control group; the average number of live-born kits was 6.08 in the experimental group and 5.69 in the control group. For Sapphires, the results were as follows: In the experimental group, the average number of born and live-born kits were 6.34 and 5.74, respectively; in the control group the respective litter sizes were 6.05 and 5.29.

DISCUSSION

First attempts to improve reproductive performance in female mink with injections of progesterone were undertaken by Franklin (1958). Less than ten years later, the results of his work on the use of hCG were published by Holcomb et al. (1962); however, the results were not satisfactory. Positive results of research on female stimulation using hCG were attained by Adams (1981), who suggested the possibility of ovulation induction by application of human chorionic gonadotropin, also in animals that fail to mate. These observations, as the author states, may indicate a behavioural problem rather than a dysfunction at the level of the ovary. This fact has been confirmed by studies of Wehrenberg et al. (1992). These authors also achieved good results in mating-reluctant females by giving eCG and hCG. With a double administration of 100 IU of eCG, the authors attained the largest number of mated and whelping females, as well as the highest average number of young born per litter. Also Klotchkov et al. (1998, 2005) and Klotchkov and Eryuchenkov (2000, 2003) successfully used hCG; administering it to mink, the authors obtained a significant difference ($p < 0.001$) between the treated and untreated groups in the average numbers of born and live-born kits. According to these authors, the average number of born was 5.9 and 5.4, respectively in the treated group and the control; the average number of live-born kits was 5.1 in the treated group and 4.5 in the control group.

The level of prolificacy and fertility of the species is highly varied and this variability is associated with the genotype. As some authors indicate (Felska-Błaszczyk et al. 2010; Møller 2000; Nieminen et al. 2010; Socha et al. 2003), both the length of gestation and the performance of breeding vary depending on the female's colour variety. This fact has also been confirmed by others (Ślaska et al. 2009; Kołodziejczyk and Socha 2011), who indicate that some genes which are involved in the development of new colour varieties are responsible for this. Socha and Markiewicz (2002) argue that the average litter size in Polish mink farms is quite low and ranges from 2.2 to 5.9 individuals. According to other authors, mink litter size remains at a level of 5 kits (Amstislavsky 2000; Persson 2007), although much larger litters may be attained, numbering even 13–15 kits, which may indicate a large, untapped reproductive potential of this species.

CONCLUSION

The study revealed statistically significant differences (in favour of the hormone-stimulated group) in the number of born and live-born kits; within the group of Mahogany females, the differences were significant in both analysed reproductive seasons, whereas for Standard Brown and Sapphires, these were significant in the second year of the experiment. The results indicate beneficial effects of applied stimulation in terms of fertility of female mink and advocate its use in the breeding practice.

REFERENCES

Adams C.E. 1981. Observations on the induction of ovulation and expulsion of uterine eggs in the mink, *Mustela vison*. J. Reprod. Fertil. 63(1), 241–8.

Amstislavsky S., Ternovskaya Y. 2000. Reproduction in mustelids. Anim. Reprod. Sci. 60, 571–81.

Douglas D.A., Houde A., Song J.H., Farookhi R., Concannon P.W., Murphy B.D. 1998. Luteotropic hormone receptors in the ovary of the mink (*Mustela vison*) during delayed implantation and early--postimplantation gestation. Biol. Reprod. 59(3), 571–8.

Dziadosz M., Seremak B., Lasota B., Masłowska A., Mieleńczuk G. 2010. Analysis in some reproduction traits of female mink (*Neovison vison*) depending on the colour varieties and age. Acta Sci. Pol. Zootech. 9(4), 71–80.

Felska-Błaszczyk L., Seremak B., Lasota B., Klecha A. 2013. Extra light during pregnancy improves reproductive performance of mink (*Neovison vison*). Ann. Anim. Sci. 13(4), 797–805.

Felska-Błaszczyk L., Sulik M., Panknin A. 2010. The incidence of barren females of mink (*Mustela vison*) of various colour types in relation to systems and dates of mating. Acta Sci. Pol. Zootech. 9(4), 81–92.

Franklin B.C. 1958. Studies on the effects of progesterone on the physiology of reproduction in the mink, *Mustela vison*. Ohio J. Sci. 58(3), 163–70.

Holcomb L.C., Schaible P.J., Ringer R.K. 1962. The effects of varied lighting regimes on reproduction in mink. Mich. Agric. Exp. Stn. Quart Bull. 44(4), 666–78.

Klotchkov D.V., Alekhina T.A., Trapezov O.V., Petrenko O.I. 2005. Estrous cycle, folliculogenesis, and brain catecholamines after stimulation of the sexual system by choriogonadotropin in female minks selected for behavior. J. Evol. Biochem. Physiol. 41(3), 333–40.

Klotchkov D.V., Eryuchenkov P.A. 2000. Response of the mink reproductive system to hormonal stimulation in october as a prognostic criterion of folluculogenesis and fertility. J. Evol. Biochem. Physiol. 36(2), 170–7.

Klotchkov D.V., Eryuchenkov P.A. 2003. Effects of hCG on folliculogenesis and fecundity in mink (*Mustela vison* Schreb). Theriogenology 60(9), 1583–93.

Klotchkov D.V., Trapezov O.V., Kharlamova A.V. 1998. Folliculogenesis, onset of puberty and fecundity of mink (*Mustela vison* Schreb.) selectively bred for docility or aggressiveness. Theriogenology 49(8), 1545–53.

Kołodziejczyk D., Socha S. 2006. Analysis of factors influencing conformation traits in mink (*Mustela vison* Schreb.) of Standard and Pastel type. Ann. UMCS, Sect. EE Zootechnica 24(57), 409–14.

Kołodziejczyk D., Socha S. 2011. Analysis of effectiveness of breeding work and estimation of genetic and phenotypic trends for reproductive traits in American mink. Ann. Anim. Sci. 11(2), 273–282.

Lagerkvist G., Johansson K., Lundeheim N. 1993. Selection for litter size, body weight, and pelt quality in mink (*Mustela vison*): experimental design and direct response of each trait. J. Anim. Sci. 71, 3261–3261.

Lasota B., Masłowska A., Felska-Błaszczyk L., Dziadosz M., Seremak B., Skuratko A. 2013. Stimulatory effect of hCG on male American mink (*Neovison vison*) in the breeding season. Ann. Anim. Sci. 13(3), 563–70.

Maciejowski J., Jeżewska G. 1993. Genetic grounds of fur animals breeding. Zesz. Nauk. Prz. Hod. 12, 5–12.

Møller SH. 2000. A decision support tool for litter size management in mink, based on a regional farm reproduction database. Scientifur 24(3), 183–92.

Nieminen P., Pölönen I., Mustonen A.-M. 2010. Increased reproductive success in the white American mink (*Neovison vison*) with chronic dietary β-sitosterol supplement. Anim. Reprod. Sci. 119(3), 287–92.

Persson S. 2007. The Mink (*Mustela vison*) as an indicator of environmental reproductive toxicity. Swed. Univ. Agricult. Sci. 50, 1–23.

Pilbeam T.E., Concannon P.W., Travis H.F. 1979. The annual reproductive cycle of mink (*Mustela vison*). J. Anim. Sci. 48(3), 578–84.

Rietveld A.A. 1978. Three years of practical application of hCG at Northwood fur Farms Inc. Scientifur 2, 27–33.

Seremak B., Dziadosz M., Felska-Błaszczyk L., Lasota B., Pławski K., Masłowska A. 2011. A novel arrangement of breeding sets has a positive effect on some reproductive parameters in females of the American mink (*Neovison vison*). Acta Sci. Pol. Zootech. 10(4), 105–14.

Seremak B., Masłowska A., Dziadosz M., Lasota B., Kominiak M. 2010. Influence of hormonal stimulation of White Hedlund female mink which were not mated in appointed term on reproduction performance. Acta Sci. Pol. Zootech. 9(4), 225–30.

Skuratko A., Lasota B., Felska-Błaszczyk L. 2013. Estriol – alternative pregnancy diagnosis marker in the mink? Folia Pomer. Univ. Technol. Stetin., Ser. Agric., Aliment., Piscet., Zootech. 307(28), 91–98.

Ślaska B., Rozempolska-Rucińska I., Jeżewska-Witkowska G. 2009. Variation in some reproductive traits of mink (*Neovison vison*) according to their coat colour. Ann. Anim. Sci. 9, 287–97.

Socha S., Markiewicz D., Wojewódzka A. 2003. Prolificacy of selected color varieties of farm mink (*Mustela vison* Sch.). Zesz. Nauk. Prz. Hod. 68(6), 79–86.

Socha S., Markiewicz D. 2002. Effect of mating and whelping dates on the number of pups in mink. Electron. J. Pol. Agric. Univ. 5(2), http: //www.ejpau.media.pl/articles/volume5/issue2/animal/art-02.pdf, access: 31.03.2015.

Wehrenberg W.B., Kurt K.J., Hutz R.J. 1992. Effects of equine chorionic gonadotropin on reproductive performance in anestrous mink. J. Anim. Sci. 70(2), 499–502.

Abstract. The aim of the study was to determine how an administration of chorionic gonadotropin (hCG) in place of the first mating would affect breeding performance of female American mink (*Neovison vison*). The experiment was conducted in two consecutive breeding seasons on groups of females composed of one- and two-year old mink, similar in body weight, belonging to three colour morphs: Sapphire, Standard Brown (Wild), and Mahogany. The experimental group were females treated with an injection of 20 IU hCG instead of being subjected to the first mating. The females were first mated on day 8 after the treatment, repeated on the following day (scheme: 8 + 9). The control females were mated in a conventional way, three times (scheme: 1 + 8 + 9). Statistically significant differences were found in litter sizes (both numbers of born and live-born kits) in favour of the hormonally treated females. The Mahogany females, treated with hCG, produced larger litters in both studied seasons, whereas Standard Brown and Sapphire females – in the second year of the experiment. The results indicate a positive effect of hCG stimulation on the fertility of female mink and advocate its use in the breeding practice.

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