Original Article

Failure to conceive in deslorelin-induced oestrous bitches with regard to removal of hormone implants after ovulation

Suppawiwat Ponglowhapan[1*] Atid Suthamnatpong[1] Achiraya Khamanarong[1]

Ekkapol Akaraphutoporn[1] Suphanut Kulabsri[1] Uttra Jamikorn[2]

Abstract

Removal of a GnRH agonist deslorelin implant either before or during ovulation in deslorelin-induced oestrous bitches is suggested because prolonged administration of the slow-release agonist may compromise luteal function secondary to pituitary down-regulation. This study aimed at investigating the effect of 4.7 mg deslorelin (Suprelorin®) on oestrous induction, ovulation and conception rate after removing the implant 72-96 hours post-ovulation to ensure that the entire ovulation process was complete. Ovulation began when serum progesterone reached 5-6 ng/mL. The implants were inserted subcutaneously in the umbilical area in 5 healthy, intact anoestrous beagles (1.5-3 years). Time of implantation was considered as Day 0, and time elapsed from Day 0 to oestrous induction was calculated. In all bitches, vaginal cytology was abruptly changed on Day 3, and oestrous signs were observed on Day 5 post-implantation. Ovulation occurred in all treated bitches on Days 11.4±0.9 (ranging from 11-13 days) post-implantation. Transcervical artificial inseminations (TCAI) with chilled semen (>75% sperm motility) using Scandinavian catheter were performed on the 2nd and 4th day post-ovulation. Pregnancy was confirmed on Days 32-35 post-TCAI by transabdominal ultrasonography and serum relaxin test (WITNESS® Relaxin). No foetuses were detected and the relaxin tests were negative (pregnancy rate = 0%). Of all bitches, progesterone levels were within a normal range throughout the dioestrus period (62 days). The unsuccessful pregnancy outcome was possibly related to the delayed removal of the implants. Further study should be conducted to investigate physiological changes in follicular development, ovulatory process, oocyte quality, oviduct and uterine environment of oestrous-induced bitches in relation to the time of implant removal.

Keywords: deslorelin, dog, oestrous induction, pregnancy

[1]*Department of Obstetrics, Gynaecology and Reproduction,* [2]*Department of Animal Husbandry, Faculty of Veterinary Science, Chulalongkorn University, Henri Dunant Rd., Bangkok 10330, Thailand*

***Correspondence**: sponglowhapan@gmail.com*

Introduction

A non-seasonal monoestrous reproductive pattern of domestic bitches is unique among other domestic animals. The follicular phase and spontaneous ovulations are followed by a luteal phase usually lasting for about 2 months, regardless of pregnancy. Thereafter, a period of anoestrus with duration of 2 to 10 months follows each oestrous cycle, making a range of inter-oestrous interval in the bitch being between 4 and 14 months (Christie and Bell, 1971). Considering the unpredictable length of anoestrus, induction of fertile oestrus is of particular interest to be able to plan breeding management and to time whelping to commercialize puppies in a certain period of the year. In addition, clinical importance of oestrous induction in bitch is the treatment of primary or secondary anoestrus (Kutzler et al., 2009). Many hormonal manipulations to induce canine oestrous cycle have been reported including sex steroid hormone (synthetic estrogens), exogenous gonadotropins (FSH, LH, hCG, PMSG and human menopausal gonadotropin), gonadotropin releasing hormone (GnRH) and its analogues (lutrelin, buserelin, fertirelin, leupprolide and deslorelin), dopamine agonists (bromocriptine and cabergoline) and opioid antagonists (naloxone) (for review; Kutzler, 2007). However, the results vary greatly in the efficacy of inducing oestrus and in the fertility of the induced oestrus. At present, there are no veterinary medicinal products licensed for the induction of oestrus in dog.

Deslorelin was first introduced to the field of small animal reproduction as a novel contraceptive approach in dogs (Trigg et al., 2000). Interestingly, instead of oestrous suppression, the subcutaneous administration of deslorelin implant induces oestrus in all treated bitches (Wright et al., 2001). After that, deslorelin has been researched for the purpose of oestrous induction (Volkmann et al., 2006a; Kutzler et al., 2009; Walter et al., 2011; Fontaine et al., 2011; Wolf et al., 2012; von Heimendahl and Miller, 2012), and the results were satisfactory. Pregnancy rates following deslorelin implants vary between studies, ranging between 25 and 68.8% depending on (i) the stage of oestrous cycle (dioestrus, early anoestrus or late anoestrus) of treated bitches and (ii) the time of implant removal in relation to ovulation (Volkmann et al., 2006a; Kutzler et al., 2009; Walter et al., 2011; Fontaine et al., 2011; von Heimendahl and Miller, 2012). The necessity of implant removal to avoid subsequent premature luteal regression and suppression of serum progesterone concentrations during the second half of the luteal phase caused by gonadotropin insufficiency (Volkmann et al., 2006b) remains unclear because some bitches were able to whelp successfully although the implant had not been removed (Maenhoudt et al., 2012). Nonetheless, if the implant was not removed, the percentage of late abortion was up to 65% (Kutzler et al., 2009). Time of implant removal varied among studies; at the time of clinical heats before ovulation, i.e. serum progesterone levels first exceeded 1.5 ng/mL (Kutzler et al., 2009), as soon as bloody vaginal discharge was observed (Walter et al., 2011) or on the day of ovulation (Fontaine et al., 2011; von Heimendahl and Miller, 2012). Because a complete ovulation period in a bitch lasts approximately 48-72 hours, removal of the implants after the entire process of ovulation is complete may improve pregnancy rate and litter size of bitches. Litter size has been reported to be smaller in deslorelin-induced oestrus than natural litters (Wolf et al., 2012).

The aims of this study were to investigate the pregnancy outcome in deslorelin-induced oestrous bitches with regard to removal of hormone implant 72-96 hours post-ovulation and to monitor changes in genital appearances, vaginal cytology and serum plasma progesterone levels after deslorelin implantation.

Materials and Methods

***Animals*:** Five healthy, nulliparous beagle bitches, 1.5-3 years old and weighing between 10.5-13 kg, were enrolled in this study. The bitches were considered anoestrus when their serum progesterone levels were below 1 ng/mL and the vaginal cytology showed <10% superficial cells (Kutzler et al., 2009). However, the duration of anoestrus was unknown. This study was conducted under the stipulations of University Animal Care and Use Committee (Protocol No. 1431070).

***Induction of oestrus*:** Deslorelin implants of 4.7 mg (Suprelorin®, Virbac, Carros, France) were inserted subcutaneously in the post umbilical in all anoestrous bitches on the same day. Time of implantation was considered as Day 0, and time elapsed from Day 0 to oestrous induction was calculated. The implant was removed 72-96 hours after ovulation. After local anaesthesia (0.2 mL of 2% lidocaine; subcutaneous injection), a small incision (0.5 cm long) was made using a scalpel blade on the administration site. The animals were observed for any adverse side effect on pathological changes in the ovary and uterus for 6 months after implant removal.

***Genital examination and serum progesterone measurement*:** Following implant insertion, the bitches were examined daily for vulvar swelling and sanguineous vaginal discharge. The onset of vaginal bleeding and discharge appearance were observed and classified as serosanguineous, bloody, serous, mucous and milky. Exfoliative vaginal cytology was performed once before and daily post-implantation until the onset of cytologic dioestrus (<70% superficial cells) (Vokmann et al., 2006a). Smears were stained using Diff-quick solution. Two hundred cells were evaluated and classified as parabasal, intermediate and superficial cell types (Christie et al., 1972), and the percentage of cells was calculated. Blood samples were collected and circulating progesterone concentrations were measured by chemiluminescence immunoassay (Architect ci 16200, Abbott Diagnostics, IL, USA) once before deslorelin administration, daily during peri-ovulatory period (>60% superficial cells until progesterone levels exceeded 10 ng/mL) and every 3-4 days throughout dioestrus. Intra- and inter-assay coefficients of variation were 12.0% and 14.0%, respectively. The detection limit of serum progesterone was 0.1 ng/mL.

***Ovulation timing*:** Based on the serum progesterone assay, the day of initial ovulation was determined when serum progesterone levels reached 5-6 ng/mL (Fontaine et al., 2011). Time (days) from implantation to initial ovulation was recorded.

***Breeding management*:** Semen was collected from 4 fertile beagles and pooled before preservation at 5°C. Sperm quality was examined each time before use. Only good semen samples with >75% sperm motility were inseminated. Transcervical artificial inseminations (TCAI) using the Scandinavian catheter was done twice on Days 2 and 4 post-ovulation. All bitches were inseminated with $150\text{-}200\times10^6$ progressively motile spermatozoa that were chilled for 1-3 days (Ponglowhapan et al., 2004).

***Pregnancy diagnosis*:** Examinations on all inseminated bitches were carried out 32-35 days after first TCAI using transabdominal ultrasound scan and relaxin test kit (WITNESS® Relaxin, Zoetis Ltd., NJ, USA).

***Data analysis*:** Data were analysed using descriptive statistic. Chronological changes in morphologic vaginal epithelial cells and progesterone concentrations were plotted and showed in the results.

Results

Vaginal cytology and discharge: All treated bitches well responded to the deslorelin administration by showing oestrous signs. External genital appearances indicated that the onset of mild vulvar swelling and a vaginal haemorrhage were first observed on Days 3 and 5, respectively. Vaginal discharge was observed for 5-12 days. Abrupt changes in the exfoliative vaginal cytology (approximately 20% intermediate cells) were observed on Day 3 following implantation. The percentage of superficial cells increased after Day 4 (>50%), continued increasing until Day 15 (>80%) and subsequently decreased on Day 16 (Figure 1).

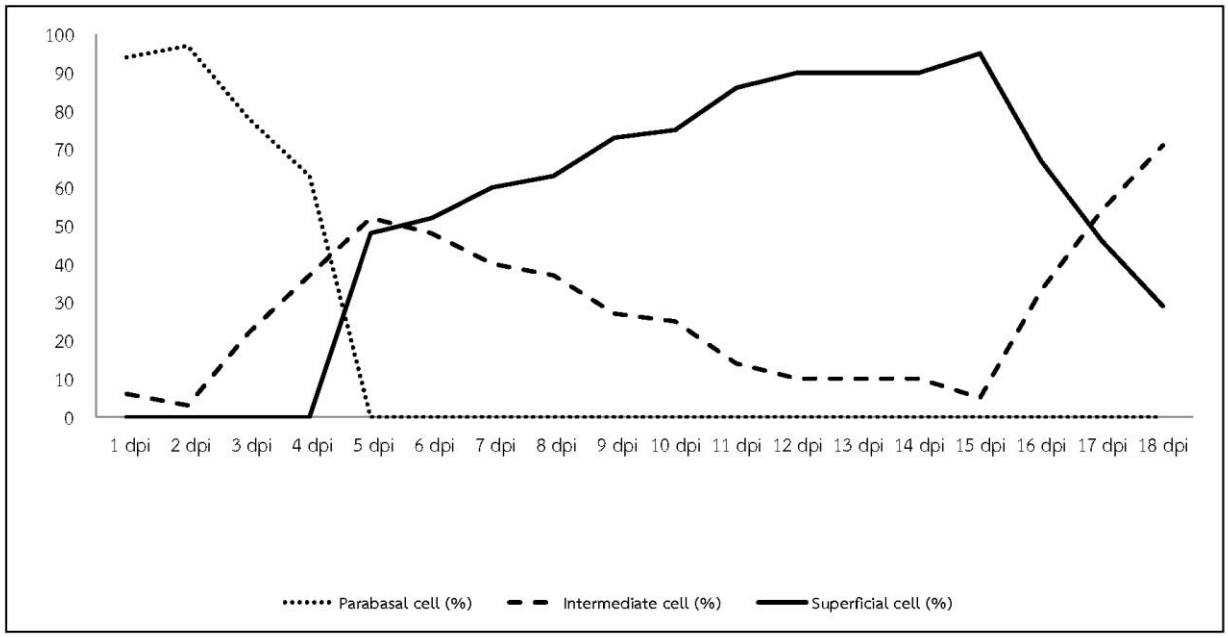

Figure 1 Changes in the mean percentage of vaginal cytology (parabasal cells, intermediate cells and superficial cells) over time post-implantation (dpi; day post-implantation) in deslorelin-treated bitches (n=5)

Ovulation and serum progesterone levels: All oestrous-induced bitches ovulated. The mean interval (mean±SD) between insertions of the implant and ovulation as determined by serum progesterone concentrations being between 5 and 6 ng/mL was 11.2±1.09 days or 8.2±1.09 days after the onset of detected oestrus. The time interval where mean progesterone concentrations were between 4 and 10 ng/mL (ovulation period) lasted for 2 days, between Days 10 and Day 12 post-implantation (Figure 2). Mean progesterone levels increasingly went up to 42.57 ng/mL on Day 29. On Day 32 onwards, the progesterone concentrations gradually declined and reached 1 ng/mL on Day 62. The mean (±SD) of progesterone concentrations is demonstrated in Figure 2, and the progesterone levels of each dog are shown in Figure 3.

Pregnancy outcomes: None of the implanted bitches gave positive results on relaxin test kit and there were no evidences of gestational sacs or embryonic structures found on the ultrasound scan on Days 32-35 after the first TCAI or Day 48 after implantation. In addition, there were no abnormal echogenic structures found in the ovaries and uterine horns of all treated bitches.

Discussion

The results obtained in this study clearly reveal the good efficacy of deslorelin implant at a dosage of 4.7 mg per dog in the induction of oestrus and ovulation in anoestrous bitches, which is in agreement with previous studies performed in a group of experimental beagle bitches (Walter et al., 2011) or in clinical trials where the population size was larger with

a wide variety of dog breeds (Fontaine et al., 2011; von Heimendahl and Miller, 2012). All bitches in our study were in oestrus as indicated by the external genital appearances and vaginal cytology. These clinically significant findings indicate that deslorelin administration is a highly reliable method for oestrous induction in anoestrous bitches, compared to the efficacy of other regimens (Kutzler, 2007).

After implant insertion, changes in genital appearance as well as vaginal cytology were very consistent in all 5 bitches; vulvar swelling was noticeable on Day 3, increased intermediate cells (approximately 20%) were found on Days 3-4 and approximately 2 days later the presence of vaginal bleeding together with abrupt increases in superficial cells (>50%) were observed. In this study, we were able to monitor daily changes in the morphologic appearance of vaginal epithelial cells before and after deslorelin implantation throughout the proestrous and oestrous stages of the cycle. The alteration in cellular pattern of exfoliative vaginal cytology found in our study was similar to natural oestrus (Phemister et al., 1973; Concannon, 2011). Ovulation occurred in all induced-oestrous bitches beginning on Day 10 or Day 11 after treatment and, based on the progesterone levels of 4-10 ng/mL, the ovulation took about 2 days. During ovulation, morphologic appearance of vaginal epithelial cells exhibited around 80% superficial cells and vaginal discharge became clearer. Taken together, these findings imply that the follicular development, the synthesis of oestrogen and ovulation can be induced by the administration of GnRH agonist deslorelin implants in the same chronological events as for natural oestrous cycle.

In bitch, considerable variation in the time at which ovulations occur in relation to the onset of vulvar swelling or vaginal bleeding of early proestrus has been documented (England et al., 1989). In a group of 50 bitches presented for breeding management, ovulation during natural heat took place approximately 12 days after the onset of proestrus and the range was reported between 7 and 22 days (Johnston and Root, 1995). Of induced-oestrous bitches, if the animals were treated with deslorelin implants during anoestrous stage, ovulation was observed 7.5 days (6-9) (Fontaine et al., 2011), 8.2 days (4-15) (Walter et al., 2011), 9.2 days (4-16) (von Heimendahl and Miller, 2012) and, in our study, 8.2 days (7-9) after the onset of detected oestrus. These data indicate that the time of ovulation induced by subcutaneous deslorelin implantation is fairly constant and slightly earlier than that of natural oestrous cycle. Furthermore, ovulation was observed in 81.3-100% of deslorelin-treated bitches (Fontaine et al., 2011; Walter et al., 2011; von Heimendahl and Miller, 2012). Anovulatory oestrus may occur if bitches are treated with deslorelin during early anoestrus or early removal of the implant prior to ovulation period (Fontaine and Fontbonne, 2011), which is not the case in our study.

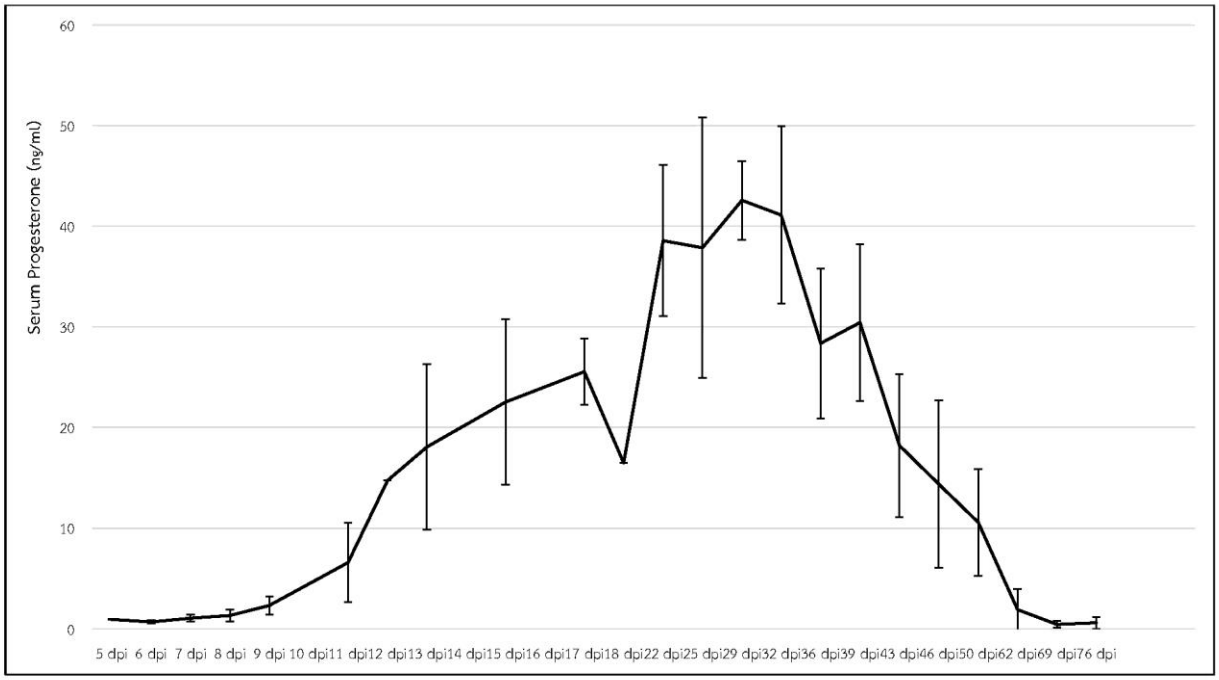

Figure 2 Mean (±SD) serum progesterone concentrations (ng/mL) over time post-implantation (dpi; day post-implantation) in deslorelin-treated bitches (n=5). Implants were removed 72-96 hours after ovulation.

Pregnancy rate was reported to be between 25 and 78% depending on the time of deslorelin treatment during anoestrus (early or late stage) (Fontaine et al., 2011). In this study, the duration of anoestrus was unknown. It is difficult to explain unsuccessful pregnancies in deslorelin-induced oestrous bitches with regard to removal of the implant 72-96 hours after ovulation because (i) all treated bitches showed normal oestrous signs as monitored by genital appearances and vaginal cytology, (ii) ovulation occurred in all treated bitches as indicated by progesterone levels, (iii) timing of ovulation was closely monitored and inseminations were performed during the fertile period, (iv) the quality of chilled semen had been

examined and only sperm with good progressive motility were used for TCAI, and (v), most importantly, serum progesterone concentrations following ovulation and throughout dioestrus were within normal reference ranges (Thuróczy et al., 2016; Thejll Kirchhoff and Goericke-Pesch, 2016).

Insufficient luteal function resulting in pregnancy loss is considered when serum progesterone is <5 ng/mL on 28-35 days of pregnancy and continues to decline (Root Kustritz, 2001). During this critical period, progesterone concentrations were found to be >10 ng/mL in all deslerelin-treated bitches. Therefore, suspected luteal insufficiency, so-called hypoluteoidism, is unlikely to be an explanation in the present study. Incomplete ovulation may be another explanation for the unsuccessful pregnancy observed in the present study; however, this phenomenon usually happens during pubertal oestrus (Wildt et al., 1981). Delayed removal of deslorelin implant 72-96 hours post-ovulation may be associated with infertility or failure to conceive. Further studies should be conducted to investigate physiological changes in the follicular development, ovulatory process, oocyte quality, oviduct and uterine environment of oestrous-induced bitches in relation to time of the implant removal.

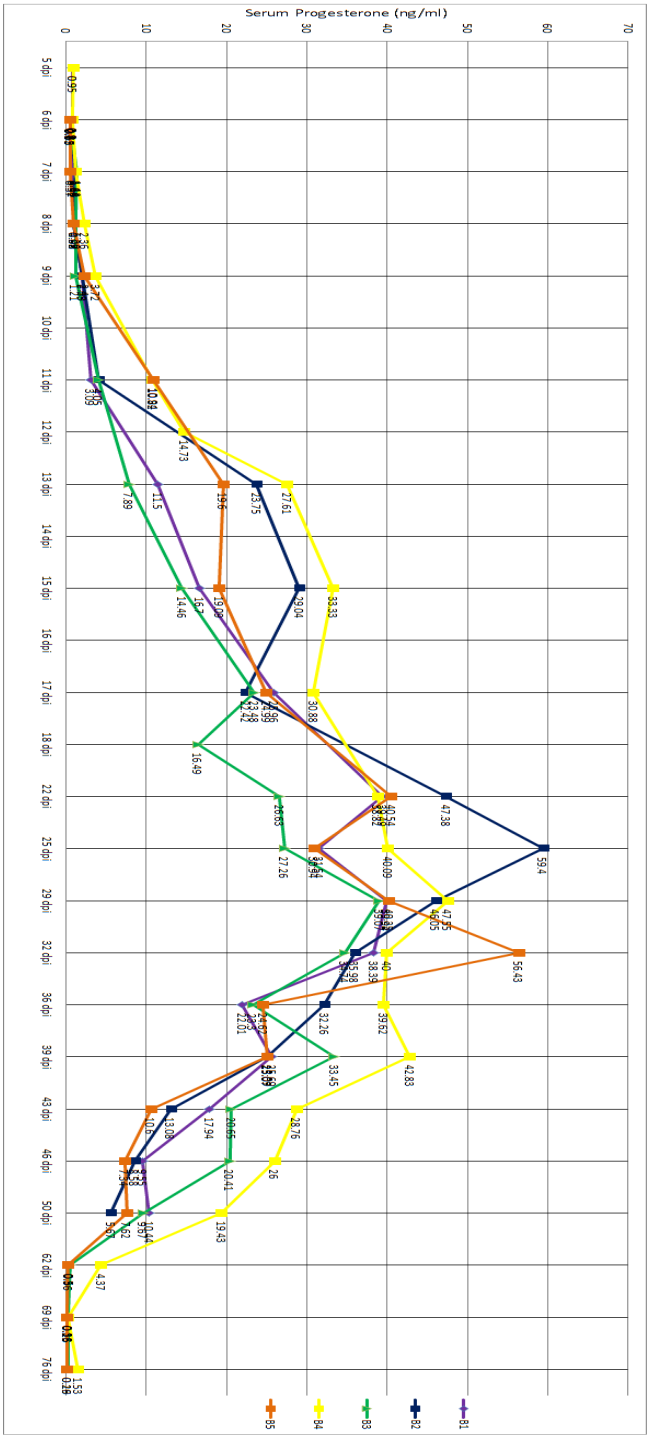

Figure 3 Serum progesterone concentrations (ng/mL) over time post-implantation in 5 deslorelin-treated bitches (B1-B5) (dpi; day post-implantation). Implants were removed 72-96 hours after ovulation.

Although no adverse health effects were seen in this study, they have been described in other deslorelin-treated bitches (Palm and Reichler, 2010; Arlt et al., 2011; Fontaine and Fontbonne, 2011). Prolonged bloody vaginal discharge or persistent heat and subsequent pyometra especially in elderly dogs were mainly observed. Few individuals were affected by urinary incontinence and coat change. In conclusion, the efficacy of subcutaneous administration of the potent GnRH agonist deslorelin formulated in a slow-release implant was proved to be a highly reliable method for the induction of oestrus and ovulation in anoestrous bitches. Delayed removal of deslorelin implant post-ovulation (72-96 hours) may be associated with failure to conceive, which requires further investigation.

Acknowledgements

This study was financially supported by the Faculty of Veterinary Science, Chulalongkorn University. We thank Research Unit of Obstetrics and Reproduction in Animals for providing the chemicals and technical assistance.

References

Arlt SP, Spankowsky S and Heuwieser W 2011. Follicular cysts and prolonged oestrus in a female dog after administration of a deslorelin implant. N Z Vet J. 59(2): 87-91.

Christie DW and Bell ET 1971. Endocrinology of the oestros cycle in the bitch. J Small Anim Pract. 12(7): 383-389.

Christie DW, Bailey JB and Bell ET 1972. Classification of cell types in vaginal smears during the canine oestrous cycle. Br Vet J. 128(6): 301-310.

Concannon PW 2011. Reproductive cycles of the domestic bitch. Anim Reprod Sci. 124(3-4): 200-210.

England GC, Allen WE and Blythe SA 1989. Variability of the time of calculated LH release in 218 canine pregnancies. Vet Rec. 125(25): 624-625.

Fontaine E and Fontbonne A 2011. Clinical use of GnRH agonists in canine and feline species. Reprod Domest Anim. 46(2): 344-353.

Fontaine E, Mir F, Vannier F, Gérardin A, Albouy M, Navarro C and Fontbonne A 2011. Induction of fertile oestrus in the bitch using Deslorelin, a GnRH agonist. Theriogenology. 76(8): 1561-1566.

Johnston SD and Root MV 1995. Serum progesterone timing of ovulation in the bitch. In: Proceedings of the annual meeting of the society for Therigenology, San Antonio, TX, p. 195-203.

Kutzler MA 2007. Estrus induction and synchronization in canids and felids. Theriogenology. 68(3): 354-374.

Kutzler M, Lamb SV and Volkmann D 2009. Comparison between vestibular and subcutaneous insertion of deslorelin implants for oestrus induction in bitches. Reprod Domest Anim. 44 (Suppl 2): 83-86.

Maenhoudt C, Santos NR, Fontaine E, Mir F, Reynaud K, Navarro C and Fontbonne A 2012. Results of GnRH agonist implants in oestrous induction and oestrous suppression in bitches and queens. Reprod Domest Anim. 47 (Suppl 6): 393-397.

Palm J and Reichler IM 2010. Effectiveness of deslorelin acetate for the suppression of heat in the bitch. In: Proc 7th EVSSAR congress. Louvain la Neuve, Belgium. p 87.

Phemister RD, Holst PA, Spano JS and Hopwood ML 1973. Time of ovulation in the beagle bitch. Biol Reprod. 8(1): 74-82.

Ponglowhapan S, Essén-Gustavsson B and Linde Forsberg C 2004. Influence of glucose and fructose in the extender during long-term storage of chilled canine semen. Theriogenology. 62(8): 1498-1517.

Root Kustritz MV 2001. Use of supplemental progesterone in the management of canine pregnancy. In: Concannon PW, England GCW, Verstegen J (eds), Recent advances in small animal reproduction. International Veterinary Information Service (http:www.ivis.org), Ithaca.

Thejll Kirchhoff K and Goericke-Pesch S 2016. Changes in serum progesterone concentrations in Bernese mountain dogs and Cavalier King Charles Spaniels during pregnancy. Theriogenology. 86(7): 1850-1855.

Thuróczy J, Müller L, Kollár E and Balogh L 2016. Thyroxin and progesterone concentrations in pregnant, nonpregnant bitches, and bitches during abortion. Theriogenology. 85(6): 1186-1191.

Trigg TE, Wright Pj, Armour AF, Williamson PR, Junaidi A and Martin GB 2000. Long-term reversible desexing of male dogs and oestrus postponement of bitches using a GnRH analogue implant. In: Proc Adv Dog Cat Exotic Carniv Reprod. p 21.

Volkmann DH, Kutzler MA, Wheeler R and Krekeler N 2006a. The use of deslorelin implants for the synchronization of estrous in diestrous bitches. Theriogenology. 66(6-7): 1497-1501.

Volkmann DH, Kutzler MA, Wheeler R, Krekeler N, Klewitz J and Lamb SV 2006b. Failure of hCG to support luteal function in bitches after estrus induction using deslorelin implants. Theriogenology. 66(6-7): 1502-1506.

von Heimendahl A and Miller C 2012. Clinical evaluation of deslorelin to induce oestrus, ovulation and pregnancy in the bitch. Reprod Domest Anim. 47 Suppl 6: 398-399.

Walter B, Otzdorff C, Brugger N and Braun J 2011. Estrus induction in Beagle bitches with the GnRH-agonist implant containing 4.7 mg Deslorelin. Theriogenology. 75(6): 1125-1129.

Wildt DE, Seager SW and Chakraborty PK 1981. Behavioral, ovarian and endocrine relationships in the pubertal bitch. J Anim Sci. 53: 182-191.

Wolf T, Meyer H and Kutzler M 2012. Litter size response to oestrous induction with deslorelin (Ovuplant®) in dogs. Reprod Domest Anim. 47 (Suppl 6): 387-388.

Wright PJ, Verstegen JP, Onclin K, Jöchle W, Armour AF, Martin GB and Trigg TE 2001. Suppression of the oestrous responses of bitches to the GnRH analogue deslorelin by progestin. J Reprod Fertil Suppl. 57: 263-268.

บทคัดย่อ

การผสมไม่ติดในสุนัขที่เหนี่ยวนำการเป็นสัดด้วยฮอร์โมนเดสโรเลอริน และถอดฮอร์โมนออกหลังการตกไข่

ศุภวิวัธน์ พงษ์เลาหพันธุ์ [1*] อธิศ สุธัมนาถพงษ์[1] อชิรญา ขมะณะรงค์[1]
เอกพล อัครพุทธิพร[1] ศุภณัฐ กุหลาบศรี[1] อุตรา จามีกร[2]

การเหนี่ยวนำการเป็นสัดในสุนัขเพศเมียด้วยการฝังฮอร์โมนจีเอ็นอาร์เอชอะโกนิส เดสโลเรอลิน มีคำแนะนำว่าควรเอาเม็ดฮอร์โมนสังเคราะห์ออกเมื่อสุนัขแสดงอาการเป็นสัดในช่วงก่อนหรือระหว่างตกไข่ เนื่องจากหากไม่นำออกจะทำให้เกิดการกดการทำงานของเนื้อเยื่อลูเตียลและส่งผลต่อการลดลงของระดับโปรเจสเตอโรนเมื่อสุนัขตั้งท้อง การศึกษานี้มีวัตถุประสงค์เพื่อศึกษาผลของการฝังเดสโรเรอลินขนาด 4.7 มิลลิกรัมใต้ผิวหนังต่อการเหนี่ยวนำการเป็นสัด การตกไข่ และการตั้งท้อง หากนำเม็ดฮอร์โมนออกภายหลังการตกไข่เสร็จสิ้นสมบูรณ์ (72-96 ชั่วโมง) โดยการตกไข่เริ่มเกิดขึ้นเมื่อระดับซีรั่มโปรเจสเตอโรนอยู่ที่ 5-6 นาโนกรัมต่อมิลลิลิตร ทำการฝังฮอร์โมนบริเวณสะดือของสุนัขบีเกิ้ล 5 ตัว อายุ 1.5-3 ปี ซึ่งอยู่ในระยะแอนเอสตรัส ภายหลังการฝังฮอร์โมนสุนัขทุกตัวมีอาการเป็นสัดอย่างชัดเจน เซลล์เยื่อบุช่องคลอดเปลี่ยนแปลงในวันที่ 3 และปากช่องคลอดบวมแดง มีของเหลวปนเลือดออกจากช่องคลอดในวันที่ 5 หลังฝังฮอร์โมน ไข่ตกวันที่ 11.4±0.9 (ช่วง 11-13 วัน) ทำการผสมเทียมโดยการปล่อยน้ำเชื้อในมดลูกในวันที่ 2 และ 4 หลังไข่ตก ตรวจการตั้งท้องในวันที่ 32-35 หลังผสมด้วยชุดตรวจรีแลกซินในกระแสเลือดและอัลตร้าซาวน์มดลูก พบว่าสุนัขทั้ง 5 ตัวไม่ตั้งท้อง ระดับโปรเจสเตอโรนในกระแสเลือดอยู่ในช่วงค่าปกติของระยะไดเอสตรัสตลอดระยะเวลา 62 วันหลังฝังฮอร์โมน การผสมไม่ติดในการศึกษานี้อาจเป็นผลมาจากช่วงเวลาที่นำฮอร์โมนออกไม่เหมาะสม ในอนาคตควรมีการศึกษาต่อไปถึงการเปลี่ยนแปลงทางสรีรวิทยาของการตกไข่ คุณภาพของไข่ที่ตก การเปลี่ยนแปลงของท่อนำไข่และมดลูก ซึ่งอาจมีความสัมพันธ์กับช่วงเวลาต่าง ๆ ที่นำเม็ดฮอร์โมนออก

คำสำคัญ: เดสโลเรอลิน สุนัข การเหนี่ยวนำการเป็นสัด การตั้งท้อง

[1]ภาควิชาสูติศาสตร์ เธนุเวชวิทยา และวิทยาการสืบพันธุ์, [2]ภาควิชาสัตวบาล, คณะสัตวแพทยศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย ปทุมวัน กรุงเทพฯ 10330 ประเทศไทย

***ผู้รับผิดชอบบทความ** E-mail: sponglowhapan@gmail.com*