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Histopathologic examination of the genital tract in rabbits treated once or twice with a slow-release deslorelin implant for reversible suppression of ovarian function

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ABSTRACT

A total of 13 rabbits were treated with a subcutaneous deslorelin long-term release implant (4.7 mg) to study the effect on ovarian function and histologic features of the uterus. Seven rabbits (group 1) were implanted with a slow-release deslorelin implant before onset of puberty for 273 days as a part of a previous study. After resumption of ovarian function had been confirmed, they were implanted again at the age of 430 days. Six adult rabbits (>177 days old; group 2) were implanted with a slow-release deslorelin implant for 273 days. Ovarian function before, during, and after treatment with the implant was assessed by measuring serum progesterone levels 10 days after a challenge injection of a short-acting GnRH (0.8 μg buserelin intramuscularly) on progesterone levels in peripheral blood. Values more than 4 ng/mL progesterone were considered to verify ovarian function. Animals in group 1 underwent ovariohysterectomy during the second treatment with the implant and the uteri, and ovaries were subjected to histopathologic examination. Endometrial hyperplasia and endometritis were observed in 5 of 7 animals. Nonatretic and atretic follicles at different developmental stages, but no active corpora lutea, were present in the ovaries. Ovariohysterectomy of group 2 animals was performed 2 to 12 months after implant removal. The histopathologic examination of the uterus and ovary of four animals neutered during induced pseudopregnancy showed no signs of uterine disorders. In two animals undergoing ovariohysterectomy 12 months after implant removal, endometritis was present. Their ovaries contained follicles at different developmental stages and corpora albicantia. Reversible suppression of ovarian function can be achieved in female rabbits by the use of GnRH slow-release implants administered before or after puberty. The findings of endometrial hyperplasia and endometritis in seven out of 13 rabbits treated once or twice with the implant may indicate that the development of age-related pathologies of the uterus cannot be prevented by the suppression of ovarian function with a long-acting GnRH implant.

1. Introduction

The dekapeptide hormone GnRH is the primary releasing hormone of reproduction and an important part of the hypothalamic–pituitary–gonadal axis [1]. Suppression of sexual function with subcutaneous GnRH slow-release

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analog implants could be achieved in several species [2–4], and a deslorelin-containing implant is approved for suppression of sexual function in the adult male dog in several countries. In a case report, the treatment of a rabbit buck with a 4.7-mg deslorelin implant led to downregulation of testosterone and reduction of testicular size [5]. Another study could not confirm this observation and concluded that the treatment is not suitable for hormonal castration [6]. In prepubertal female rabbits, ovarian function could be suppressed for at least 273 days. In five of seven animals, the occurrence of puberty could be delayed by the treatment [7]. To the best of our knowledge, no study concerning the use of slow-release analog implants in adult female rabbits has been published yet.

Because of the high incidence of diseases of the reproductive tract [8–10], preventive surgical neutering of the female rabbit is recommended [9,11,12]. The procedure has the disadvantage of possible surgery-related complications [13] and a high anesthetic risk [14].

Serum progesterone concentrations less than 2 ng/mL are within the basal range [15]. The female rabbit is a reflex ovulator [16] and ovulation can be induced by mating, similar stimuli, and different drugs. Administration of 0.8 μg buserelin, a GnRH analog, induced ovulation in 87.9% of cases [17]. If the ovulated oocytes are not fertilized, a pseudopregnancy with elevated progesterone concentrations for 17 [18] to 18 [19] days will be established [11], during which the endometrium undergoes morphologic changes with six distinct phases. The priming phase is followed by a proliferative phase, in which the epithelium becomes pseudostratified and the glands proliferate. After mucosal folding, secretory activity, and cell fusion to multinucleated cells, the fifth phase with maximum progesterone levels takes place from the eighth to the 13th day. Nearly all cells are multinucleated, except occasional ciliated cells, and extensive ciliation is possible. In the last period, the endometrium degenerates and is sloughed off [20].

The aim of the study was to examine the effect of a 4.7-mg deslorelin slow-release implant on ovarian function in cycling rabbits. The hypothesis of this study was that ovulation and pseudopregnancy could not be induced under implant exposure. Additionally, the genital tract of rabbits treated once or twice with the implant was examined by histologic examination.

2. Materials and methods

Animal experimentation was performed in the Clinic of Small Animal Surgery and Reproduction, Center for Clinical Veterinary Medicine, Faculty of Veterinary Medicine, Ludwig Maximilian University Munich, Germany, and approved by the local authority (Gz. 55.2-1-54–2532–41–11; Kreisverwaltungsreferat, Munich, Germany).

2.1. Animals and animal housing

Thirteen cycling female Zika hybrid rabbits were used. They were housed in groups of two to four animals. No artificial lightening was provided. Hay and water were available ad libitum. Additionally, each animal received 25 g of pelleted food (Canin Kombo, Asamhof, Kissing, Germany) and a constant amount of vegetables per day.

2.2. Experimental design and blood sampling

The study was designed as a monocentric, randomized study. Before the treatment, a complete blood count and a serum analysis were performed. The general condition of all animals was assessed daily. A thorough clinical examination of each animal was performed twice weekly, and the animals were observed for mounting behavior for 1 hour. The color of the vulva was recorded, and the perivulvar region was examined for the presence of vaginal discharge as a possible sign of metropathies. After the end of the study, all animals were passed on to new owners. All of them requested neutering, and thus, 13 genital tracts could be submitted for histopathologic examination.

The animals were randomly divided into different groups. Group 1 consisted of seven adult animals (animal nos 1–7), which had been treated as prepubertal animals with a 4.7-mg deslorelin slow-release implant for 273 days in a previous study (Fig. 1). There are no data concerning the expected duration of deslorelin release in female rabbits so far. The deslorelin implant used in this study is licensed for the induction of temporary infertility in healthy, entire, sexually mature male dogs, and infertility is achieved from 6 weeks up to at least 6 months after initial treatment [21]; 105 days after the first implant had been removed, the animals from group 1 were treated with another 4.7-mg deslorelin slow-release implant at the age of 430 days (Day 0); 10 days after implant insertion, serum progesterone values were determined to screen for implant-induced ovulation and pseudopregnancy; 73 days (animal no. 1) or 273 days (animal nos 2–7) after implant insertion, ovariohysterectomy was performed at the age of 503 (animal no. 1) and 703 days (animal nos 2–7) under the

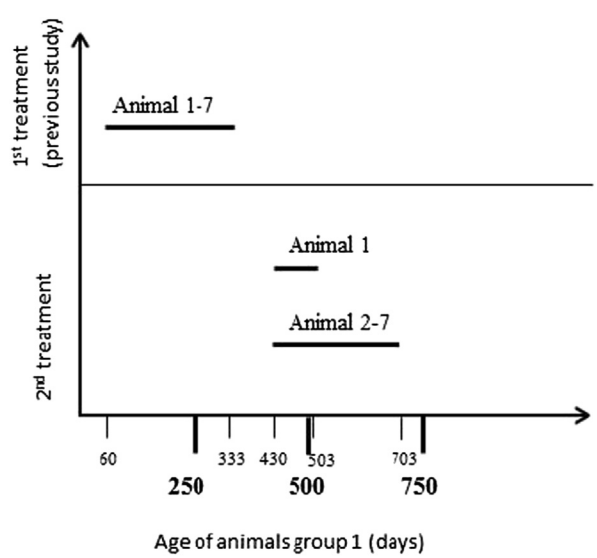

Fig. 1. Periods of treatment with the GnRH slow-release analog implant (bars) (group 1).