Available online at www.sciencedirect.com
SCIENCE DIRECT®
Theriogenology
Theriogenology 66 (2006) 122–125
www.journals.elsevierhealth.com/periodicals/the

# Artificial insemination in domestic cats (*Felis catus*)

Toshihiko Tsutsui *

*Department of Reproduction, Nippon Veterinary and Animal Science University 7-1, Kyonan-cho, 1 chome, Musashino-shi, Tokyo 180-8602, Japan*

**Abstract**

Artificial insemination (AI) in cats represents an important technique for increasing the contribution of genetically valuable individuals in specific populations, whether they be highly pedigreed purebred cats, medically important laboratory cats or endangered non-domestic cats. Semen is collected using electrical stimulation, with an artificial vagina or from intact or excised cauda epididymis. Sperm samples can be used for AI immediately after collection, after temporary storage above 0 °C or after cryopreservation. There have been three and five reports on intravaginal and intrauterine insemination, respectively, and one report on tubal insemination with fresh semen. In studies using fresh semen, it was reported that conception rates of 50% or higher were obtained by intravaginal insemination with 10–50 $\times$ $10^6$ spermatozoa, while, in another report, the conception rate was 78% after AI with 80 $\times$ $10^6$ spermatozoa. After intrauterine insemination, conception rates following deposition of 6.2 $\times$ $10^6$ and 8 $\times$ $10^6$ spermatozoa were reported to be 50 and 80%, respectively. With tubal insemination, the conception rate was 43% when 4 $\times$ $10^6$ spermatozoa were used, showing that the number of spermatozoa required to obtain a satisfactory conception rate was similar to that of cats inseminated directly into the uterus. When frozen semen was used for intravaginal insemination the conception rate was rather low, but intrauterine insemination with 50 $\times$ $10^6$ frozen/thawed spermatozoa resulted in a conception rate of 57%. Furthermore, in one report, conception was obtained by intrauterine insemination of frozen epididymal spermatozoa. Overall, there have been few reports on artificial insemination in cats. The results obtained to date show considerable variation, both within and among laboratories depending upon the type and number of spermatozoa used and the site of sperm deposition. Undoubtedly, future studies will identify the major factors required to consistently obtain reliable conception rates, so that AI can become a practical technique for enhancing the production of desirable genotypes, both for laboratory and conservation purposes.


*Keywords:* Artificial insemination; Cat; Deposition site; Semen

## 1. Introduction

### 1.1. Background

In cats, artificial insemination (AI) may be necessary when natural mating is not successful, or when the male and female are housed at separate locations. The technique is also potentially applicable to aiding in the conservation of rare felids on the verge of extinction [1]. During normal mating, cat semen is deposited intravaginally; however, knowledge on semen storage sites after natural mating is limited. Using AI, three sperm deposition sites – intravaginal, intrauterine and intratubal – are possible. The number of spermatozoa necessary for conception may vary according to the insemination site. Both spermatozoa can be inseminated either immediately after recovery, after cryopreservation or after temporary storage at above 0 °C, although there are no reports yet available using the latter method. Furthermore, results may depend on whether or

* Tel.: +81 422 39 7340; fax: +81 422 39 7340.
*E-mail address:* tsutsui@nvau.ac.jp.

0093-691X/$ – see front matter © 2006 Elsevier Inc. All rights reserved.
doi:10.1016/j.theriogenology.2006.03.015

not the female experienced a natural estrus at the time of AI or whether estrus was induced by treatment with gonadotropic hormones.

In the first report on AI in cats by Sojka et al. [2], freshly collected spermatozoa were deposited intravaginally. Since then, AI of fresh as well as frozen spermatozoa, into the vagina, uterus and uterine tube has been reported. Cat semen has been collected both by electroejaculation and voluntary ejaculation using an artificial vagina. Spermatozoa can be recovered from the epididymides either by percutaneous epididymal sperm aspiration (PESA) or by transmigration from the excised cauda epididymides into surrounding medium. These collection methods may affect fertility in different ways. The purpose of the present paper is to review the background and current status of artificial insemination in cats, including factors influencing the success rate, with a particular focus on site of sperm deposition in the reproductive tract.

## 2. Intravaginal artificial insemination

In cats, intravaginal AI is performed deep into the vagina using a fine needle, either without [2] or with anesthesia [3,4], at various intervals after administration of hCG for induction of ovulation. Including the initial report in 1970 [2], of a total of three reports on intravaginal AI in cats, fresh spermatozoa were used in two [2,3] and frozen spermatozoa were used in the third [4].

### 2.1. Fresh spermatozoa

In both studies using fresh semen, ovulation was induced by administering hCG to females in natural estrus. In the first study on AI in cats that resulted in conception [2], fertilization was achieved in one of three females after deposition of $1.25 \times 10^6$ spermatozoa, but not with $0.5 \times 10^6$ spermatozoa. A conception rate of 54% (14/26) was obtained after insemination with 5–$50 \times 10^6$ spermatozoa and two to four offspring were born per litter. In the same report [2], a preliminary trial was done to determine if a second AI would result in a higher pregnancy rate. Eight females were inseminated with $5 \times 10^6$ spermatozoa at the time of the first hCG treatment (50 IU) and again 24 h later, at which time a lower level of hCG (10 IU) was given. Six pregnancies (75%) were produced in this follow-up trial, which should have been interpreted as encouraging evidence for further studies on intravaginal AI in cats. However, although more than three decades have elapsed since the initial report on intravaginal AI, there have only been two additional reports in domestic cats. The second and only other study in which fresh spermatozoa were used [3] was not published until 30 years after the first report [2]. In the recent study, a single AI was performed at 15, 20 or 30 h after hCG administration. The conception rate following AI was 1/16 (6%) with $20 \times 10^6$ spermatozoa, 6/18 (34%) with $40 \times 10^6$ spermatozoa and 7/9 (78%) with $80 \times 10^6$ spermatozoa. There was no correlation between the fertilization rate and the time from hCG administration to AI. Three or four kittens were born per litter regardless of the number of spermatozoa inseminated. Even though Sojka et al. [2] and Tanaka et al. [3] used similar methods for AI, the results of the latter study indicated that higher sperm numbers were required to achieve a satisfactory pregnancy rate as compared to that of the original report, $80 \times 10^6$ spermatozoa versus 5–$50 \times 10^6$ spermatozoa, respectively. Also, as mentioned earlier, another difference between the two studies was that anesthesia was not used in the original report [2], while the cats in the second study were anesthetized for AI [3]

### 2.2. Frozen spermatozoa

In the first report of AI in cats with frozen semen [4], the samples were frozen by pelleting on blocks of dry ice before storage in liquid nitrogen. Upon thawing, 50–$100 \times 10^6$ motile spermatozoa were intravaginally inseminated into anesthetized females on the second and third days of estrus using a 16 g lavage needle (9 cm long) attached to a 1 ml syringe. Some females were inseminated during a natural estrus after induction of ovulation with hCG and other females were given FSH and hCG to induce estrus and ovulation, respectively, before AI. Pregnancy was established after AI in 6 of 56 (11%) cats, 4 of which were from the natural estrus group and 2 were from the induced estrus group. A total of 12 kittens were born, with litter sizes ranging from 1 to 4 kittens.

## 3. Intrauterine artificial insemination

Intrauterine insemination in cats has been performed both by laparoscopy [5] and mid-line laparotomy [6–8]. There have been four studies on intrauterine AI in cats, three using fresh semen (Howard et al. [5], Tsutsui et al. [6,7]) and one using frozen spermatozoa (Tsutsui et al. [8]).

### 3.1. Fresh spermatozoa

In the study by Howard et al. [5], estrus was induced by administration of 100 IU eCG, followed by induction