



# Spontaneous ovulation in the cat: incidence among queens presented at a veterinary teaching facility

Journal of Feline Medicine and Surgery  
1–7

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DOI: 10.1177/1098612X241248351

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## Abstract

**Objectives** The queen is recognised as an induced ovulator. Ovulation without male contact is generally regarded as spontaneous. The aim of this study was to provide an estimate of the incidence of spontaneous ovulation in a population of intact queens presented to a veterinary care facility for both reproductive and non-reproductive reasons. The secondary objective was to determine the roles of age, breed, body weight, presence of tom cats or other cycling queens, and physical contact with humans on triggering spontaneous ovulation, along with its implications.

**Methods** Serum samples from post-pubertal intact queens presented between January 2020 and June 2023 to the Veterinary Teaching Hospital of the University of Padova, Italy, were retrieved and assayed for progesterone (P4) levels. Serum P4 above 2.0 ng/ml without a history of male contact was considered as proof of spontaneous ovulation.

**Results** In total, 31 serum samples from 29 intact post-pubertal queens were obtained. Of the 31 samples, 14 had a P4 concentration above 2.0 ng/ml and 9/29 (31.0%) queens ovulated spontaneously. The mean age and weight of the nine spontaneously ovulating queens were  $4.3 \pm 5.7$  years and  $3.7 \pm 0.8$  kg, respectively. One queen ovulated spontaneously at her first heat at 6 months of age, which makes it the earliest spontaneous ovulation reported.

**Conclusions and relevance** As both our findings and previous publications indicate that the incidence of spontaneous ovulation in queens is consistently  $\geq 30\%$ , cats should not be considered strictly induced ovulators, but as a species in which ovulation can be either spontaneous or induced. Since the risk of progesterone-dependent conditions (cystic endometrial hyperplasia – pyometra complex, feline mammary hypertrophy) is increased in these queens, veterinarians should be aware and advise breeders and clients accordingly.

## Plain language summary

Female cats ovulate upon vaginal stimulation exerted by the spikes of the male's penis while mating, which makes them induced ovulators. When ovulation occurs without male contact, it is considered spontaneous. There are several factors that are thought to facilitate this non-induced ovulation, but no consensus on their relevance. The aim of this study was to provide an estimate of the rate of spontaneous ovulation in a population of intact female cats of various breeds presented to a veterinary care facility, as well as the influence of factors such as age, breed, body weight, presence of male cats or other cycling females, and physical contact with humans on triggering spontaneous ovulation. In addition, possible implications arising from progesterone exposure were assessed.

Progesterone was retrospectively assayed in the serum of adult cycling female cats presented to the Veterinary Teaching Hospital of the University of Padova, Italy, between January 2020 and June 2023.

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Values above 2.0 ng/ml without a history of male contact were considered proof of spontaneous ovulation. Out of 29 cats, nine (31%) ovulated spontaneously, with one female having done so at puberty (6 months of age), which makes it the first spontaneous ovulation ever reported in a pubertal queen.

As spontaneous ovulation has been found to occur at a rate of more than 30% both in our and in previous publications on this topic, we propose that cats should be considered both an induced and a spontaneously ovulating species. Since animals that ovulate spontaneously, and therefore experience additional luteal phases, are at a higher risk of developing progesterone-dependent conditions, veterinarians should be aware and advise breeders and clients accordingly.

**Keywords:** Spontaneous ovulation; queen; progesterone; luteal phase

**Accepted:** 3 April 2024

## Introduction

The cat is an induced ovulator, polyoestrus, long-day breeder. Once the breeding season starts – soon after day length increases – queens experience sequential oestrus periods of 5–7 days interspersed with 9-day (range 2–19) interoestrus periods in the absence of ovulation.<sup>1–3</sup> Seasonality is determined by photoperiod and resultant melatonin concentration. An increased number of hours of daylight, associated with lower melatonin release by the pineal gland, triggers the onset of reproductive activity.<sup>4–6</sup> In the northern hemisphere, this starts in January or February, depending on latitude, and cyclic ovarian activity is present until late September to mid-October, when the length of daylight is no longer sufficient, prompting the onset of anoestrus.<sup>3</sup> Currently, because many cats live indoors, artificial lighting impacts on reproductive seasonality, causing some queens to cycle all year long, although most indoor-living queens still display a seasonal anoestrus.<sup>3,7</sup> In addition, Oriental breeds seem to be less sensitive to the photoperiod and may cycle throughout the year.<sup>8</sup>

As in most mammalian species, ovulation must be preceded by a rise in luteinising hormone (LH).<sup>9,10</sup> In the cat, distension of the posterior vagina due to penile intromission<sup>11</sup> causes hypothalamic release of gonadotropin-releasing hormone (GnRH), quickly resulting in an LH surge whose magnitude and duration is proportional to the number of copulations.<sup>9,10,12</sup> Ovulation occurs approximately 24 h after the copulation-induced LH peak,<sup>13</sup> with a noticeable progesterone (P4) output detected from day 3 after mating.<sup>13</sup> A P4 concentration greater than 1.0 ng/ml is regarded as evidence of luteal function in the queen, and therefore as proof of ovulation.<sup>13,14</sup>

Ovulation is defined as spontaneous whenever it occurs without intromission or other vaginal stimuli. Mounting without intromission,<sup>9</sup> stroking of the tail, head or lower back,<sup>2</sup> or simply even the presence of male cats<sup>15</sup> have been reported to increase the rate of spontaneous

ovulation (SO). Feline SO as a field of study is indebted to Dr Dennis Lawler and his collaborators who serendipitously observed evidence of queens ovulating spontaneously while examining their reproductive tracts for other reasons.<sup>16</sup> Since then, several other studies have either specifically studied SO in queens<sup>15,17</sup> or reported queens ovulating spontaneously within the course of feline reproductive studies.<sup>18–21</sup> The rate of SO in these publications is in the range of 35%<sup>16</sup> to 87%.<sup>15</sup> In addition, anecdotal reports of purebred queens having long (several weeks) intervals between consecutive oestrous cycles may suggest the occurrence of a luteal phase following SO.<sup>22</sup>

The aim of the present study was to provide an estimate of the incidence of SO in a normal population of intact queens of various breeds presented to a veterinary care facility for both reproductive and non-reproductive reasons. Furthermore, the roles of age, breed, body weight (BW), presence of tom cats or other cycling queens, and physical contact with humans on triggering SO are discussed.

## Materials and methods

The clinical records database of the Veterinary Teaching Hospital of the University of Padova, Italy, was searched for records of (1) post-pubertal intact queens presented between January 2020 and June 2023, and for which (2) a serum sample was still available in the archive of the Clinical Chemistry Laboratory where biological samples are stored frozen for at least 3 years after the original assay. Queens were considered post-pubertal according to age (2 years or older). If the queen was younger than 2 years, post-pubertal status was ascertained by telephone consultation with the owner and confirmation of the manifestation of at least one heat. Serum samples that satisfied criteria (1) and (2) were retrieved and permission was obtained from the cats' owners for assaying P4 in the stored samples. Serum P4 was assayed using

an automated immunoassay analyser AIA-360 (TOSOH Bioscience). Intra-assay and inter-assay coefficients of variation (CVs) were calculated on one pool of samples analysed 10 times on the same day and twice in five consecutive days, respectively. Owners of queens whose serum was found to have a P4 concentration  $\geq 1.0$  ng/ml were contacted again to obtain information about whether any contact with male or other female cats had occurred. If contact with an intact male cat was ruled out, owners were questioned about the queens' housing and management conditions at the time of blood collection from which the serum was used to assay P4. The presenting complaint and signalment data from queens whose P4 was found to be above 1.0 ng/ml were recovered and tabulated to study whether breed, age and BW played a role in the occurrence of SO. The mean and standard deviation were calculated for the age and BW of the SO queens. Breed, age and BW results from our study were compared with those available in the literature. However, because of different study designs and different definitions of SO between our study and those previously published, no statistical comparison was performed.

Despite the cut-off value of 1.0 ng/ml reported in the literature,<sup>13,14</sup> evidence of SO in our study was determined only when a queen presented serum P4  $> 2.0$  ng/ml without a history of sexual contact with male cats. This threshold was adopted to minimise the possibility of an incorrect definition of SO (with P4 values in the range of 1.0–2.0 ng/ml) due to intra-assay variation.

## Results

Serum samples were obtained from 29 intact post-pubertal queens presented for musculoskeletal/trauma ( $n=4$ ), reproductive ( $n=3$ ), dermatological ( $n=2$ ), internal medicine – hepatic and renal ( $n=2$ ), ophthalmological ( $n=1$ ) or unknown ( $n=1$ ) complaints, or for preoperative or general health check-ups ( $n=6$ ), or with the purpose of starting treatment with megestrol acetate ( $n=6$ ),<sup>23</sup> deslorelin ( $n=5$ ) or melatonin ( $n=1$ ) to control reproduction. Two queens were sampled twice at two different times, and so two serum samples were available for each, making a total of 31 serum samples. Intra- and inter-assay CVs were 5.03% and 3.51%, respectively.

Of the 31 samples, 14 had a P4 concentration above 2.0 ng/ml. However, in only eight queens could sexual contact with a tom cat be ruled out and thus the ovulation considered spontaneous. Two of the SO queens developed pyometra in the subsequent luteal phase. In addition, one other cat (not included in the eight above), from which a serum sample could not be recovered, was diagnosed with feline mammary hypertrophy (FMH). Progestin therapy was ruled out through history and no owner used any progestin-based product (creams or ointments) that could have been percutaneously absorbed or licked by the queen, indicating that an SO took place. The

possibility of vaginal stimulation for this queen was ruled out based on history. Of the 29 queens, nine (31%) were thus considered to have ovulated spontaneously. Three additional queens would have been considered to have ovulated spontaneously had the P4 cut-off been defined at 1.0 ng/ml, which would raise the incidence to 41.4%.

Age, breed, BW, P4 values, date of blood sampling and housing conditions of the nine spontaneously ovulating queens, plus the other three queens with a serum P4 of 1.0–2.0 ng/ml, are displayed in Table 1. The mean age and weight of the nine SO queens were  $4.3 \pm 5.7$  years and  $3.7 \pm 0.9$  kg, respectively. One of the nine queens in our study ovulated at her first heat at 6 months of age, which makes it the earliest SO reported. No specific breed, age or BW influence on SO was evident. Only 3/9 SO queens were individually housed, while the rest (6/9) had some degree of contact, continuous or not, with other cats (neutered males, intact females or in the context of a breeding establishment). The female that developed FMH and one that was presented for pyometra were housed in breeding establishments. Hence, contact with females in heat and olfactory and auditory stimulus from tom cats are possible in these queens.

## Discussion

The aim of the present study was to estimate the incidence of SO – based on the observation of high serum P4 concentrations – in intact queens presented at a veterinary clinic for a variety of different complaints. SO had been either purposely investigated or serendipitously observed in previous publications, in which the presence of corpora lutea on histopathological examination<sup>18,21,24</sup> or serum<sup>15,16,25</sup> or faecal<sup>15,19,20</sup> P4 levels above baseline in non-mated queens was considered proof of SO. Our results (31%) are slightly lower than those previously reported (35–87%).<sup>15–21,24</sup> The queens' cycle phase was unknown before the P4 assay. An unknown number of queens might have been in anoestrus at the time of blood sampling and therefore unable to ovulate, which possibly underestimates the incidence of SO in the intact female cat population. Based on our results, SO may be expected to occur in approximately one-third of all queens (cycling and non-cycling) seen in a veterinary clinic, while this percentage may increase if one considers only cycling queens.

Age is one factor suggested to influence SO.<sup>16,17</sup> The age range in our data is too wide (0.50–16.33 years) to make any statistical comparison. However, one of the queens in our series, Kira (see Table 1), ovulated spontaneously at her first heat at 6 months of age, a finding that has not been reported in earlier studies. The role of age on the incidence of SO in domestic queens has not yet been clarified, although increasing age has been proposed to be associated with an increased SO rate.<sup>16</sup> Fontbonne et al<sup>17</sup> observed SO only in adult queens (age

**Table 1** Identification, age, body weight (BW), breed, housing conditions, progesterone (P4) value, associated pathologies and date of blood sampling of the nine spontaneously ovulating queens (serum P4 >2.0 ng/ml) and the additional three queens with a serum P4 of 1.0–2.0 ng/ml, from the study conducted in the Veterinary Teaching Hospital

Queen	Age (years)	BW (kg)	Breed	Housing	Signalment	P4 value (ng/ml)	Associated pathology	Date of blood sampling
Mel	16.33	2.19	European Shorthair	No contact with cats	Pyometra	3.34	Pyometra	26/03/2022
Josephine	12.83	NA	Maine Coon	No contact with cats	Preoperative health check-up	19.83	None	07/02/2022
Margot	1.00	4.20	European Shorthair	Contact with one neutered male cat	Megestrol acetate study enrolment	2.71	None	31/03/2022
Wendy	0.67	4.10	Maine Coon	Breeding establishment, contact with intact female cats and auditory and olfactory stimulus from tom cats	Pyometra	3.26	Pyometra	21/03/2022
Kira	0.50	4.80	Maine Coon	Breeding establishment, contact with intact female cats and auditory and olfactory stimulus from tom cats	FMH	NA	FMH	NA
Otta	0.83	3.20	European Shorthair	No contact with cats	Megestrol acetate study enrolment	20.80	None	07/02/2023
Kalika*	0.75	3.50	Bengal	Contact with one intact female cat	Megestrol acetate study enrolment	8.15	None	08/02/2023
Beatrice	4.75	NA	Scottish Fold	No contact with male cats	Chronic renal insufficiency	5.69	None	10/05/2021
Thelma	1.08	4.18	European Shorthair	Contact with one intact female cat	Deslorelin study enrolment	2.78	None	19/06/2023
Tebe†	0.50	NA	European Shorthair	No contact with cats	Trauma	1.10	None	17/10/2022
Lulu†	1.25	2.15	Persian	No contact with cats	Dermatological problems	1.04	None	11/05/2022
Aida†	5.17	3.00	Snowshoe	No contact with male cats	Dermatological problems	1.52	None	27/10/2020

\*Queen subjected to vaginal cytology 1 week before

†Queens with serum P4 of 1.0–2.0 ng/ml

FMH = feline mammary hypertrophy; NA = data not available

>3 years) and Binder et al<sup>18</sup> observed a trend of increasing rate of SO in increasing age groups. On the other hand, the populations in the studies by Gudermuth et al<sup>15</sup> and Graham et al<sup>20</sup> were composed of young (mostly 1-year-old) queens, and the SO rate was 87% and 67%, respectively.

The effect of BW on SO was first assessed by Binder et al,<sup>18</sup> who observed a significant difference in the rate of SO among different weight groups. The mean BW (3.7 kg) of the seven SO queens for which BW data were available in our study is in line with the findings of Binder et al<sup>18</sup> (heaviest group 3.3–4.3 kg), which supports an

influencing role of BW on SO. Breed might also act as a confounding variable at this level, since 2/7 queens were Maine Coons, which is a large size breed. In the study by Binder et al,<sup>18</sup> in the heaviest group, more than two-thirds of the queens ovulated spontaneously, which suggests that overnutrition might facilitate SO. Leptin is a hormone produced and released by the adipose tissue and its ability to modulate ovarian function has been demonstrated in sheep.<sup>26</sup> Likewise, leptin might play a role in ovulation in cats.<sup>18</sup> Breed-related differences in size are present in the domestic cat. Therefore, body condition score is a more precise indicator of overnutrition than

BW itself, as the latter is also dependent on the size of the animal (height at withers). The role of leptin in influencing ovulation and even SO should be further investigated in studies using body condition score rather than BW.<sup>18</sup>

Breed influences feline reproductive features such as seasonality,<sup>8</sup> display of heat behaviour,<sup>2,27</sup> pregnancy rate,<sup>28</sup> pregnancy length, litter size and age at puberty.<sup>22</sup> Out of nine SO cats in our study, three were Maine Coons. There is no previous report on the incidence of SO in this breed. However, a study of Maine Coons, Bengals, Norwegian Forest Cats and Persians reported a mean interestrus interval of  $39.3 \pm 17$  days, with no significant difference among breeds.<sup>22</sup> This length indicates that some episodes of SO might have extended the time gap between heats.<sup>22</sup> In addition, Maine Coon was among the breeds with a high incidence of pyometra in Sweden.<sup>29</sup> Fontbonne et al<sup>17</sup> reported a higher frequency of SO in an Oriental breed (Thai). Although the study population was small ( $n=11$ ), these findings suggest a potential breed influence and warrant further investigation, since these animals might face an increased risk of P4-dependent conditions.<sup>8,29</sup>

Interaction between female cats in oestrus and other conspecifics has been proposed to elicit SO, particularly if in the presence of a tom cat. Behaviours like mounting, neck-rubbing and genitalia (self) licking between group-housed female cats may trigger a release of LH,<sup>15</sup> which, depending on individual sensibility, might precipitate ovulation in individual queens. Two-thirds of our SO queens had regular contact with conspecifics. When intact male cats are group-housed with queens, auditory,<sup>20</sup> olfactory, tactile and/or visual stimuli<sup>15</sup> are triggered, which may be responsible for increasing SO rates. Moreover, a significant increase in SO was registered by Gudermuth et al<sup>15</sup> after the introduction of a male cat to the room. However, SO is also observed in individually housed cats.<sup>19,24</sup> Binder et al<sup>18</sup> reported a significantly higher rate of SO in individually housed queens compared with group-housed queens kept with neutered male cats. These findings suggest the possible importance of the neuter status of the tom cat in triggering SO. Non-physical cues (pheromonal) from intact and neutered male cats might be perceived differently by female cats, thus conditioning SO. One of the nine SO queens from our study was housed with a neutered male cat, yet the owner had never observed any reproductive interaction between the cats, even when the queen was in heat. This appears to dismiss the possibility of an induced ovulation (as a result of mating). However, there is still the possibility that the tom cat may have vaginally stimulated the queen without the owner noticing. Nevertheless, we classified this as an SO as both cats had been living with the owner for years and no such behaviour was ever observed. Although visual,

pheromonal or auditory stimuli from intact male cats are not essential for SO, they may facilitate its occurrence. These conditions are often found in breeding establishments, which potentially makes breeding queens more prone to SO.

In our study, 3/9 cats developed P4-dependent conditions: pyometra ( $n=2$ ) and FMH ( $n=1$ ). Cystic endometrial hyperplasia (CEH) – pyometra complex is a hormonally induced disease in which the endometrium displays hyperplasia and cystic dilation followed by bacterial colonisation with subsequent purulent fluid accumulation due to a closed-cervix uterus.<sup>8,30</sup> P4 plays a pivotal role in the development of this condition by causing hyperplasia of endometrial glands, increasing glandular secretory activity, inhibiting leukocyte activity, reducing myometrial contractions and causing cervical closure.<sup>8,30</sup> A recent study<sup>29</sup> suggested that Oriental cat breeds may have an increased incidence of pyometra. Authors substantiate this finding with the longer, almost continuous, reproductive season displayed by queens of these breeds, as well as the higher rate of SO.<sup>8,29</sup> FMH is a benign, hyperplastic fibroglandular proliferation of the mammary glands, without milk production, associated with endogenous or exogenous P4 stimulation, which may occur in queens and tom cats.<sup>31–33</sup> The detailed pathophysiology of FMH is currently not entirely understood. Yet, the role of P4 in the development of the condition has been clearly demonstrated because of its association with high-dose progestin therapy, the disappearance of clinical signs after ovariectomy, the spontaneous resolution in pregnant queens after luteolysis, parturition or abortion, and the positive response to antiprogestin treatment.<sup>31–34</sup> Exposure to luteal phase(s) following SO may increase the risk of developing conditions in which P4 plays a predisposing role, such as CEH–pyometra complex<sup>8,16</sup> and FMH.<sup>31,32</sup> Two of the three queens in our study that developed these conditions were owned by cat breeders. The association between being housed in a breeding establishment and having a higher risk of pyometra or FMH deserves further studies, as other contributing or hindering factors might play a role. For instance, breeding queens tend to have a lower risk of developing pyometra as pregnancy seems to play a protective role, as in bitches.<sup>8,35</sup>

Stress has been proposed to both elicit and inhibit SO.<sup>15,16</sup> P4 has been reported to increase either after adrenocorticotrophic hormone injections and after restraint procedures for blood collection in female cats.<sup>36</sup> To rule out the contribution of handling-related stress, some studies assayed P4 in the faeces rather than in the serum, bypassing the procedure of blood collection.<sup>15,19,20</sup> The fact that SO was diagnosed in a majority of queens based only on faecal P4<sup>15</sup> dismisses the effect of human manipulation on precipitating SO in queens. Another form of stress,

associated with adaptation to a new owner, household or conspecifics, might also have a negative impact on SO, causing a decreased incidence of SO in queens recently housed with other cats.<sup>18</sup>

One of our queens, Kalika (Table 1), was subjected to vaginal cytology 1 week before blood collection. In this procedure, the swab is introduced briefly (less than 5s) into the vagina, taking care to avoid unnecessary stimulation. Likewise, queens that ovulated spontaneously in a previous study<sup>21</sup> underwent vaginal cytology. Although introducing a swab into the vagina is likely to produce vaginal stimulation, no scientific data are available to support that a single insertion might induce ovulation in queens in heat.<sup>21</sup> However, we cannot be completely certain that this procedure did not trigger ovulation, even if care was taken not to cause overstimulation. We have previously attempted (on a clinical patient not included in this study) to induce ovulation by stroking the vagina of a queen in heat with a cotton swab without success. Queens might show different sensitivity thresholds for vaginal stimulation to result in endogenous GnRH release and consequent LH surge. As such, we believe that only queens predisposed to SO might do so when subjected to vaginal cytology.

The present study has some limitations. These include its retrospective nature, memory bias (one owner was asked to report events that took place 2.5 years previously), the fact that P4 was not assayed in replicates but only once for each sample and the possibility that one case of SO was indeed induced by vaginal cytology.

Nonetheless, both our findings and those in previous publications indicate that the incidence of SO in queens is consistently  $\geq 30\%$ , implying that the classification of cats as an induced-ovulating species should probably be changed to induced and spontaneously ovulating species.

## Conclusions

Of the cats presented to our Veterinary Teaching Hospital over a period of 3 years, approximately one-third ovulated spontaneously. The effect of age was not consistent, but there is clearly no minimum age beyond puberty for SO to occur. Although further studies are needed, weight (heavier queens) and breed (Maine Coons) should be regarded as potential predisposing factors for SO. Despite physical and sensory interaction with conspecifics not being essential for queens to ovulate spontaneously, these circumstances tend to facilitate its occurrence. As SO increases the prospect of CEH–pyometra and FMH, owners or breeders of queens that are predisposed or known to ovulate spontaneously should be aware of these risks. Overall, cats should no longer be regarded as a primarily induced-ovulating species but also as spontaneously ovulating, and veterinarians should be aware and advise breeders and clients accordingly.

**Conflict of interest** The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

**Funding** The authors received no financial support for the research, authorship, and/or publication of this article.

**Ethical approval** The work described in this manuscript involved the use of non-experimental (owned or unowned) animals. Established internationally recognised high standards ('best practice') of veterinary clinical care for the individual patient were always followed and/or this work involved the use of cadavers. Ethical approval from a committee was therefore not specifically required for publication in *JFMS*. Although not required, where ethical approval was still obtained, it is stated in the manuscript.

**Informed consent** Informed consent (verbal or written) was obtained from the owner or legal custodian of all animal(s) described in this work (experimental or non-experimental animals, including cadavers) for all procedure(s) undertaken (prospective or retrospective studies). For any animals or people individually identifiable within this publication, informed consent (verbal or written) for their use in the publication was obtained from the people involved.

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