Clinical evaluation of the use of aglepristone, with or without cloprostenol, to treat cystic endometrial hyperplasia-pyometra complex in bitches

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Abstract

The aim of the study was to evaluate the efficacy of aglepristone (10 mg/kg on days 1, 2 and 8) for the treatment of metritis or pyometra in bitches (n = 67) either alone for cases of metritis (n = 15), or in cases of pyometra (n = 52) with (n = 32) or without (n = 20) the addition of low doses (1 μg/kg) of cloprostenol for 5 days (days 3–7). Examinations performed on day 90, in addition to days 8, 14 and 28, determined that treatments had been curative in the long term in 54/67 bitches (80.6%). Bitches in whom pyometra did not resolve, were given additional aglepristone on day 14 (n = 38) and day 28 (n = 20). Aglepristone alone was curative in 15/15 bitches with metritis. In 17/17 bitches with closed pyometra, cervical opening occurred within 48 h of aglepristone administration. Amongst the 52 bitches with open (n = 35) or closed (n = 17) pyometra, the additional treatment with cloprostenol from days 3 to 7, significantly improved the overall success rate at day 90, which was 27/32 (84.4%), compared to 12/20 (60.0%) in bitches without cloprostenol (P < 0.05). The leucocyte count and plasma progesterone concentrations significantly decreased over the course of treatment. Thirteen of 15 bitches in whom plasma progesterone concentrations were initially low (<3.18 nmol/L) were cured. The recurrence rate after 12 and 24 months was 13.0% (3/23) and 19.0% (4/21), respectively.

Keywords: Pyometra; Metritis; Aglepristone; Cloprostenol; Bitch

1. Introduction

Cystic endometrial hyperplasia-pyometra complex or metritis/pyometra is a very common disease in bitches over 8 years of age. Depending on the degree of cervical opening, metritis/pyometra may cause inflammation of the endometrium with an intrauterine accumulation of purulent and/or hemorrhagic secretions, similar vaginal discharge, and loss of general condition. Without treatment, the infection is fatal. The disease generally occurs after the bitch has been in estrus (generally during metestrous [1]). Currently, it is generally accepted that the disease is very closely linked to hormonal imbalance in which progesterone plays a major role [2].

Surgical treatment, i.e. ovariohysterectomy, is generally performed if the bitch can tolerate the anesthetic and the owner can bear the expense. Surgery is a radical form of treatment, preventing any further reproduction. Because of the luteolytic and uterotonic properties of prostaglandin-F2 alpha (PGF), the repeated administration of PGF has been used to treat pyometra. The treatment consists of one or more injections daily over a minimum period of 5 days [3,4]. To avoid the risk of serious side effects, prostaglandins are usually only recommended for use in young bitches with no liver or

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kidney dysfunction or cardiac disease. Some authors also suggested that the cervix must be open and the uterus non-hypertrophied [5]. The duration of treatment and the onset of side effects related to the action of the prostaglandins on smooth muscle (diarrhea, salivation and vomiting) make this a demanding course of treatment, which usually requires hospitalization of the bitch.

Given the major role of progesterone in inducing cystic endometrial hyperplasia-pyometra complex, antiprogestins such as aglepristone could be expected to be useful in a medical treatment protocol. Aglepristone has been shown to effectively suppress the biological action of progesterone during gestation, interrupting gestation [6–8], causing cervical opening, and inducing parturition [9]. The molecule has been effectively used in the treatment of uterine infections associated with elevated plasma progesterone concentrations (n = 6, 7 and 31) [1,10,11]. Recent work has described the use of aglepristone to treat pyometra in bitches, in terms of the short or medium term result after the initial acute recovery (n = 52) [12], and including its combined use with cloprostenol (n = 15) [13]. However, to be suitable for medical use by practitioners, a treatment needs to be efficient, fast, and prescribed on the basis of immediate clinical findings, without the need for lengthy laboratory testing.

The aim of this clinical study was to evaluate the efficacy and safety of the treatment of metritis/pyometra in bitches by the administration of aglepristone in bitches with various forms of infectious uterine diseases, that were presented to the clinics at the Veterinary School in Nantes, and to evaluate it in comparison with the administration of combined aglepristone and low dose cloprostenol. The latter treatment strategy was designed to improve outcomes and hasten recovery without the side effects usually induced by prostaglandins in bitches with open or closed pyometra.

2. Materials and methods

2.1. Animals

This study involved 67 bitches of different breeds, aged 8.5 ± 3.2 years, with “open” or “closed” forms of metritis or cystic endometrial hyperplasia-pyometra complex. The diagnosis was confirmed by the observation of purulent or hemorrhagic vaginal discharge associated in some, but not all, cases with enlargement of the uterine lumen detected using uterine ultrasonography. Irrespective of their general health, all bitches treated had a serum concentration of urea and creatinine of less than 0.6 g/L and 10 mg/L, respectively.

2.2. Procedure

The bitches were allocated into three groups based on clinical observation: Group 1, metritis (putrid vaginal discharge with no enlargement of the uterine lumen detected on uterine ultrasonography, n = 15); Group 2, open pyometra (putrid vaginal discharge with enlargement of the uterine lumen, n = 35) and Group 3, closed pyometra (enlargement of the uterine lumen without vaginal discharge, n = 17).

Treatment varied, depending on the clinical situation, but all the bitches received one subcutaneous injection of 10 mg/kg body weight of aglepristone (Alizine®; Virbac, Carros, France) once daily on days 1, 2 and 8. This was the only treatment administered in bitches with metritis (Group 1). In addition to this treatment, two-thirds of each group of the bitches with open (Group 2, n = 10/17) or closed (Group 3, n = 21/35) pyometra were given daily subcutaneous injections from days 3 to 7, with 1 μg/kg of cloprostenol (Estrumate®; Schering-Plough, Levallois-Perret, France). Bitches with hyperthermia or dehydration and all bitches with closed pyometra received additional treatment, i.e. intravenous infusion of ringer lactate at day 1 and 24 mg/kg/day of amoxicillin-clavulanic acid from days 1 to 5 (Synulox®; Pfizer, Paris, France).

The bitches from all three groups were checked on days 14, 28 and 90. Bitches in whom pyometra did not resolve, were given additional aglepristone on day 14 (n = 38) and day 28 (n = 20). The efficacy criteria were recovery of good general health, absence of uterine lumen enlargement and absence of vaginal discharge. At the beginning of the trial and at various stages during the study, the bitches underwent a general clinical examination, gynecological examination and an ultrasound examination of the uterus (Aloka Echo Camera SSD-500 with 5 MHz transducer; Aloka, Cergy St. Christophe, France). A blood sample was also taken from the jugular vein. Mean white and red blood cell counts were measured using an impedance counter (MS9; Melet Schloesing Laboratoire, Cergy-Pointoise, France). An automated biochemistry analyzer (BP144; Ph Diagnostics, Montpellier, France) was used to determine the plasma concentrations of urea and creatinine via photospectrometry. Peripheral progesterone concentrations were measured using a commercial RIA kit (Diria-progk kit from Diasorin, Antony, France). The assay sensitivity was 0.3 nmol/L and assay specificity was 97.5%.
2.3. Statistical evaluation

Qualitative results, such as the success of the treatment, were reported with a confidence interval of 95%. Quantitative results were reported as means ± S.D. Data were analyzed by Statview (SAS Institute Inc., Cary, NC, USA). Treatment efficacy and the clinical response of metritis/pyometra were compared using a Chi-square test. A Student-Fisher test was used to compare the mean decrease in the diameter of the uterine lumen, the mean leucocyte count and the mean peripheral plasma progesterone concentration between treatments at various times during the experiment. Values were considered statistically significant when \( P < 0.05 \).

3. Results

When measuring efficacy at day 90, 54/67 bitches were cured (i.e. 80.6%, with a confidence interval range of 71.1–90.1%). The overall recovery rate was lower \( (P < 0.001) \) at day 14 (24/68, 35.3%) than at day 28 (46/67, 68.6%) and day 90 (54/67, 80.6%). The effectiveness of the treatment was not dependent on the clinical form of uterine disease (Table 1).

In bitches with metritis, treatment with aglepristone alone gave a high cure rate, with clinical resolution in over half of the bitches (9/15) within 2 weeks and within 4 weeks for the remainder with, one exception. The bitch cured at day 90 had only a slight serous discharge on day 28. This was visible by the clinician during the genital exam but not by the owner.

Clinically, the administration of aglepristone in cases of pyometra led to changes in the nature of the vaginal discharge, which changed from purulent, to mucous-like, before finally becoming serous, with a concurrent decrease in volume. The last clinical sign recorded was self-cleaning of the vulval area by the bitch. For all bitches with closed pyometra, cervical opening was induced following the first two aglepristone treatments (18/18). The mean time to cervical opening was 25.8 ± 12.2 h. The shortest time was 4 h after the first administration of aglepristone; however, cervical opening was complete within 48 h in all of the bitches. Cervical opening induced enabled the evacuation of large volumes of purulent discharge, which was associated with an immediate improvement in general condition and, in most cases, with increased appetite.

For bitches with open or closed pyometra, additional treatment with cloprostenol from days 3 to 7 improved the overall success rate at day 90 \( (P < 0.05); 27/32 \) (84.4%) with cloprostenol versus 12/20 (60.0%) without cloprostenol (Table 2). Recovery was accompanied by a gradual decrease in the diameter of the uterine lumen, which became non-detectable ultrasonographically. The mean decrease in the diameter of the uterine lumen over time depended on the treatment used (Fig. 1) and was significantly greater between days 8 and 14 in bitches receiving the combined aglepristone–cloprostenol treatment than in those treated with aglepristone alone (84.8 ± 20.7% versus 64.6 ± 41.0%).

The general condition of bitches suffering from metritis was not altered; it remained good throughout the trial. Bitches suffering from open or closed pyometra presented were depressed and anorexic in 41/53 (77.3%) and 34/53 (64.1%) cases, respectively. In all animals treated, there was a marked improvement in this criterion by the second day of treatment. On day 8, moderate lethargy was observed in 9 bitches (17.3%) and 14 bitches (26.9%) showed loss of appetite. On day 15, all bitches still receiving treatment were in good general condition, with only one showing any loss of appetite. No side effects were observed after treatment with cloprostenol in 15/33 bitches (45.5%). In the remainder, nausea was the most commonly observed side effect (12/33), and vomiting occurred in six bitches.

None of the bitches had a noticeable change in red blood cell count. The mean white blood cell counts in bitches suffering from metritis were normal and, remained lower \( (P < 0.05) \) than the counts in bitches with open or closed pyometra (Fig. 2) throughout the trial. In bitches recovering from pyometra, a marked decrease in leucocytosis was observed over the course of treatment. This mean decrease was significant on day

Table 1

<table>
<thead>
<tr>
<th>Number of bitches cured, at various stages in the trial, based on the form of uterine disease</th>
<th>Day 14</th>
<th>Day 28</th>
<th>Day 90</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metritis (%)</td>
<td>9/15</td>
<td>14/15</td>
<td>15/15</td>
</tr>
<tr>
<td>Open pyometra (%)</td>
<td>13/35</td>
<td>20/35</td>
<td>26/35</td>
</tr>
<tr>
<td>Closed pyometra (%)</td>
<td>2/18</td>
<td>12/17</td>
<td>13/17</td>
</tr>
</tbody>
</table>

Table 2

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Day 14</th>
<th>Day 28</th>
<th>Day 90</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aglepristone (%)</td>
<td>3/20 (15.0)</td>
<td>9/20 (45.3)</td>
<td>12/20 (60.0)</td>
</tr>
<tr>
<td>Aglepristone + cloprostenol (%)</td>
<td>12/33 (36.4)</td>
<td>23/32 (71.9)</td>
<td>27/32 (84.4)</td>
</tr>
</tbody>
</table>

Difference in cure rate between the two treatments on day 90 \( (P < 0.05) \).
Fig. 1. Mean (±S.E.M.) rates of decreases in the diameter of the uterine lumen at various stages in the trial, in bitches with open or closed pyometra (Group 1 + Group 2) cured by the administration of aglepristone alone (n = 12), in comparison with those receiving a combination of aglepristone + cloprostenol (n = 27). (*) Differences (P < 0.05).

Fig. 2. Mean (±S.E.M.) concentration of leucocytes at various stages in the trial, in bitches with metritis (n = 15), open pyometra (n = 26) or closed pyometra (n = 13) cured by treatment with aglepristone or aglepristone + cloprostenol. For each day, the mean white blood cell count was significantly lower in cases of metritis than in cases of open or closed pyometra. In bitches with closed pyometra, the white blood cell count was lower at day 14 (P < 0.01) and at day 28 (P < 0.001) than at day 0.
Table 3  
Means (±S.D.) plasma progesterone concentrations (nmol/L) in bitches with open or closed pyometra (Group 1 + Group 2) treated with aglepristone alone, compared to those treated with a combination of aglepristone + cloprostenol

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Day 1</th>
<th>Day 8</th>
<th>Day 14</th>
<th>Day 28</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aglepristone (n = 20)</td>
<td>12.8 ± 14.4a</td>
<td>5.6 ± 6.6b</td>
<td>4.8 ± 5.1b</td>
<td>3.3 ± 4.2b</td>
</tr>
<tr>
<td>Aglepristone + cloprostenol (n = 32)</td>
<td>19.3 ± 11.6a</td>
<td>15.08 ± 10.5c</td>
<td>4.7 ± 1.1b</td>
<td>3.96 ± 2.8b</td>
</tr>
</tbody>
</table>

Within a row, values with different superscript letters (a and b) differ (P < 0.05); within a column, values with different superscript letters (x and y) differ (P < 0.05).

15 for bitches with closed pyometra (Fig. 2). Among the 67 treated bitches, on day 1, 15 had plasma progesterone concentrations below 3.18 nmol/L. Treatment was successful in all of them, except for two bitches with open pyometra. For bitches with open or closed pyometra treated with cloprostenol (n = 32), there was a significant decrease in mean peripheral plasma progesterone concentration at day 8 (12.8 ± 14.4 nmol/L versus 5.6 ± 6.6 nmol/L on days 1 versus 8, respectively). However, in bitches treated without cloprostenol (n = 20), no significant decrease in mean plasma progesterone concentration was observed until day 14 (19.3 ± 11.6 nmol/L versus 4.7 ± 1.1 nmol/L on days 1 versus 14; Table 3).

Among the 13 failures using this treatment, one bitch did not present for follow-up and recovery could not be confirmed. One bitch with open pyometra treated with aglepristone died on day 5 in the owner’s house, and another treated with aglepristone and cloprostenol was euthanized in a private clinic on day 15 due to declining health. Ten bitches underwent an ovariohysterectomy. In this last group, six had ovarian cysts. After recovery, 23 bitches underwent follow-up visits over the 24 months following the beginning of treatment. Three of them developed pyometra between 7 and 12 months later (3/23, 13.0%) and another developed it at 19 months (4/21, 19.0%). Two of them received an additional, successful course of treatment (aglepristone + cloprostenol) and no pyometra was observed at the next cycle. We have kept in contact with the owners of three of the bitches used in the trial for some considerable time. They were treated more than 6 years ago and have come into estrus normally, with no recurrence of uterine disease. Five bitches were mated at the first estrus after recovery; four became pregnant.

4. Discussion

The use of aglepristone alone cured 27/35 (77.1%) bitches in 90 days. These results are equivalent to those reported by Hoffman et al. [11] (25/31, 80.6% at day 30) and by Trasch et al. [12] (44/52, 84.6% at day 90). The number of animals treated is insufficient to show any difference in efficacy depending on clinical type (metritis, open pyometra and closed pyometra). This study, however, demonstrated the satisfactory results obtained from the use of aglepristone in bitches suffering from metritis (recovery rate, 15/15). The use of aglepristone is also of particular interest in the treatment of closed pyometra, since it induces cervical opening, and the subsequent evacuation of purulent discharge with a marked improvement in the animal’s general condition. These results demonstrated that surgery is no longer the only treatment for closed pyometra. Moreover, even if surgery has been planned, it is possible to recommend medication as a first-line treatment (aglepristone and rehydration) to improve the general health of the bitch and minimize surgical risk. In this study, the cervix of all the bitches with closed pyometra opened within 48 h. In a study of nine bitches with closed pyometra, Wehrend et al. [14] observed cervical opening within 72 h and the same marked improvement in the animal’s general condition.

In bitches with open or closed pyometra, the repeated administration of low dose cloprostenol significantly increased the success rate at day 90 (27/32, 84.4%). The uterotonic action of cloprostenol lead to a significantly faster decrease in the diameter of the uterine lumen and its luteolytic action caused a faster drop in mean plasma progesterone concentration. This decline in progesterone concentration was observed in 15/15). The use of aglepristone is also of particular interest in the treatment of closed pyometra, since it induces cervical opening, and the subsequent evacuation of purulent discharge with a marked improvement in the animal’s general condition. These results demonstrated that surgery is no longer the only treatment for closed pyometra. Moreover, even if surgery has been planned, it is possible to recommend medication as a first-line treatment (aglepristone and rehydration) to improve the general health of the bitch and minimize surgical risk. In this study, the cervix of all the bitches with closed pyometra opened within 48 h. In a study of nine bitches with closed pyometra, Wehrend et al. [14] observed cervical opening within 72 h and the same marked improvement in the animal’s general condition.

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This particular sensitivity to low doses of cloprostenol can be explained by the poor general condition of the bitches. Nevertheless, the owners did not make any comments.

Among the treated bitches, 17 had basal progesterone concentrations. Cases of pyometra were observed at all stages of the reproductive cycle in a previous study [1]. The effects of aglepristone in pyometra have been studied in diestrus bitches selected with a progesterone concentration of more than 3.2 μm/L [1,10,11]. In the present study 15/17 bitches with a low progesterone concentration were cured after the administration of aglepristone. Similar results were observed in another report [13]. This seems to confirm that, in the pathogenesis of pyometra in bitches, the action of progesterone on the uterus is dependent on the number of progesterone receptors or their specific sensitivity, and not on the plasma progesterone concentration of this hormone.

The recurrence rate of pyometra after treatment differed from results reported in other studies. We did not observe any recurrence between days 30 and 90, and the cure rate was higher, whereas Trasch et al. [12] reported a recurrence rate of 9.8% (4/41) for the same interval; this may be due to the length of treatment and the repeated administration of aglepristone until day 28 if needed (whereas Trasch only administered aglepristone on days 1, 2 and 7). The recurrence rate from 12 to 14 months after treatment was 18.9% (4/37) for Trasch et al. [12], 9% (1/11) for Hoffmann et al. [11] and 21.4% (3/14) for Gobello et al. [13]. The mean age of the bitches (8 years) may explain the recurrence of pyometra in the 24 months following the end of treatment, since age is a major epidemiological factor in the onset of pyometra in bitches. However, treatment failure or recurrence of the disease is closely linked to the presence of ovarian cysts, which maintain a permanent endocrine imbalance. Ovarian cysts were observed in 6 of 10 bitches who underwent ovariohysterectomy due to the failure of medical treatment.

Conservative treatment with prostaglandins, which is recommended for bitches with metritis or pyometra with mild dilatation of the uterine lumen, produces a cure rate of 83–100%, with recurrence rates varying from 10 to 40%. The treatment, which requires several administrations, produces discomfort and is often associated with side effects such as hypersalivation, vomiting and diarrhea [3,4].

In conclusion, treatment with aglepristone alone was a safe and effective treatment for metritis, and an effective means of inducing cervical opening in cases of closed pyometra. The combination of aglepristone and cloprostenol was more effective in the medical treatment of open and closed pyometra than aglepristone alone. Nevertheless, in all cases, we only recommend such treatment in bitches with no liver or kidney dysfunction, and advise close clinical monitoring throughout the entire course of treatment.

References


Follow-up Examinations of Bitches after Conservative Treatment of Pyometra with the Antigestagen Aglepristone

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With 2 figures and 2 tables

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Summary

The aim of this study was to determine the therapeutic success of the medical treatment of canine pyometra with the antigestagen aglepristone and to document the recurrence rate in relation to the time interval after treatment with antigestagens. In 48 (92.8%) of the 52 treated bitches, healing could be achieved within the first 3 weeks after the treatment had been started. One bitch died as a result of renal insufficiency; in three bitches there was no emptying of the uterus, so ovariohysterectomy became necessary. In these three patients, ovarian and endometrial cysts were present. Forty-one bitches could be followed up for 3 months. Four animals developed a recurrence (9.8%). In three bitches ovarian cysts and cystic endometrial hyperlasia could be found intra operationem. The development of 37 bitches could be followed for at least 1 year. Seven animals developed a pyometra again (18.9%). Two received a repeated treatment with aglepristone and have been free from recurrence for over 12 months. In 37 animals data on the subsequent sex cycles are available. In 22 bitches next heat started at the expected time, in seven animals heat started too early. In eight bitches the period of anoestrus was prolonged. Five of the six bred bitches delivered at least one litter. The presented data show that treatment of pyometra by aglepristone results in a high healing rate. The recurrence rate can be minimized by the selection of bitches without ovarian cysts and cystic endometrial hyperlasia.

Introduction

Pyometra has so far mostly been treated by ovariohysterectomy. The advantage of this treatment is the exclusion of the risk of recurrence. However, surgical treatment has its limits where the risk of surgery is increased or in case where the owner refuses ovariohysterectomy. For this reason, different strategies of conservative treatment have been developed. The advantages of a non-surgical intervention are that the ability to breed can be maintained and that the risks of anaesthesia and surgery are excluded.

Different procedures of conservative pyometra treatment have been described. One of those is the single systemic and local use of antibiotics (Querol, 1981; Threlfall, 1995). A significant number of cases treated as above and transferred afterwards, show, however, that this treatment frequently did not result in a healing but a protraction of the disease.

Another medical treatment is the parenteral application of prostaglandin F2α. Even single cases of shock and death have been reported (Berkolt, 1997). In order to minimize these risks, low concentrations are administered with a high application frequency (Hubler et al., 1991; Nolte et al., 1993). This regime clearly restricts the practicability of the proposed treatment protocols in practice, particularly in outpatients. Fieni et al. (2001a) treated bitches with the closed and open form of pyometra with a combination of aglepristone and cloprostenol successfully, but 68% of the bitches showed side-effects like vomiting.

The effects of oestrogen administration such as cervix relaxation and opening, as well as the increase of uterus contractility justify theoretically the use of this hormone in the treatment of pyometra. However, its mediocre therapeutic success and a high potential of undesired side-effects make its use rather questionable (Watson, 1942; Legendre, 1976; Kraft and Kuffer, 1995).

The use of oxytocin and secale alkaloids was also discussed (Hardy and Osborne, 1974; Funkquist et al., 1983). The therapeutic effect has not been proven to date. All forms of medical treatment presented so far show clear limitations in their usage and go along with undesired effects. Regarding the recurrence rates of the different therapeutic regimens, there are either no data available, or, with regard to prostaglandin treatment, the published data in literature vary strongly (Johnston et al., 2001).

With the introduction of antigestagen-based drugs in veterinary medicine new therapeutic agents for the treatment of progesterone-dependent disorders have become available. After the demonstration of the antigestagenic effect of this group of substances in the bitch (Hoffmann and Gerres, 1989), the field of indication of this pharmaceutical agent in gynaecology and obstetrics of small animals has been constantly enlarged. The successful use of this medication could be shown not only in the treatment of canine and feline pyometra (Blendinger et al., 1997; Breitkopf et al., 1997; Hecker et al., 2000; Hoffmann et al., 2000; Fieni et al., 2001a), but also in the prevention of nidation (Hubler and Arnold, 2000), in the induction of abortion (Blendinger et al., 1994), in the treatment of fibroadenomatosis (Wehrend et al., 2001) and in the induction of labour (Riesenbeck et al., 1999; Fieni et al., 2001b).

In order to assess the use of antigestagens in the treatment of pyometra compared with other therapeutic procedures, it is necessary to know the healing and recurrence rate after the use of antigestagens. So far, the only information available is based only on a few number of animals and does not allow a differentiated examination of the recurrence rate in different time intervals following successful treatment for more than...
The data of 52 bitches of different breeds who suffered from pyometra and were treated with aglepristone were evaluated. All bitches underwent a general and gynaecological examination including ultrasonography (5 MHz convex probe, Kretz SA 9900; Kretz, Zipf, Austria) and differential blood count (CELL - DYN 3500; Abbott Diagnostika GmbH, Wiesbaden-Delkenheim, Germany). To determine urea and creatinine concentrations a photometric procedure (Reflotron®; Boehringer, Mannheim, Germany) was used.

The inclusion criteria for this study were:
1 Filled uterus detectable by ultrasonography (Fig. 1)
2 Putrid vaginal discharge at the day of presentation (open pyometra) or onset of putrid vaginal discharge within the first 24 h after first application (closed pyometra)
3 Poor general condition, as well as increased renal parameters (normal values: urea > 8.3 mmol/l, creatinine > 106 μmol/l; Kraft and Dürr, 1999) did not count as exclusion criterion. Only bitches with an uterus rupture were excluded from conservative treatment.

The antigestagenic agent aglepristone was licensed in France in 1996 in form of the veterinary drug Alicine® (Virbac, Carros, France) for the prevention of nidation and abortion induction in the dog. The treatment protocol for subcutaneous application of 10 mg/kg body weight on day 1, two and seven was applied (Hoffmann et al., 2000). All animals received antibiotic treatment for at least 7 days. Amoxicillin combined with clavulanic acid (Synulox®; Pfizer, Karlsruhe, Germany) or enrofloxacin (Baytril®; Bayer, Leverkusen, Germany) in dosages according to the manufacturer’s instructions were used.

The bitches underwent outpatient treatments, except for three bitches. The reasons for inpatient treatment were that two of these had increased renal parameters and required infusion therapy, and the other one was in an extremely bad condition.

The maximum total diameter of the filled uterus was 15.3 cm. In seven bitches the heat was regularly suppressed by progestins (n = 7, 13.5%). In three animals, nidation had been prevented by oestrogens prior to the date of the consultation (two times 2 weeks earlier, once 8 weeks earlier) because of unwanted breeding (n = 3, 5.8%).

Findings upon admittance
Twenty-two bitches were in good general condition, 25 animals showed slight impairments. Five of the bitches were in a poor general condition, three of them were not able to get up. The rectally measured inner body temperature of 21 animals was above 39.0°C, that of the others was within the reference range. In 44 animals an open form of pyometra was detected. In eight patients, a closed pyometra was diagnosed.

The maximum total diameter of the filled uterus was between 0.6 and 6.3 cm.

The blood count of 27 bitches showed leucocytosis (leucocytes > 12 g/l), in three animals leucopaenia could be noticed (leucocytes < 5 g/l). Urea and creatinine were increased in two animals (bitch 1: urea 34.0 mmol/l, creatinine 257 μmol/l; bitch 2: urea 15.2 mmol/l, creatinine 183 μmol/l).

Short-term success up to day 21 after treatment
Fifty-one of the 52 treated bitches showed an obvious improvement in the general state of health during the first two days after starting the treatment. This could be seen in an increase of food ingestion and a more joyful behaviour. One of the two bitches who had been presented with increased renal values died of acute renal failure during the second day of
treatment, despite immediate admission to a clinic and the start of infusion therapy. In the second bitch, a lasting improvement of the renal parameters under infusion therapy with isotonic 0.9% sodium chloride solution (sodium chloride solution ad usum veterinarium, Selectavet, Weyau-Holzolling, Germany) could be recorded within 3 days. Up to day 7 the inner body temperature returned to normal in all patients. A decline of the initial leucocytosis could be observed in all affected animals until the fourth consultation. A distinctive increase of vaginal discharge could be detected in all bitches within the first 24 h after application of aglepristone. The uterus lumen of 48 animals showed a distinctive reduction up to day 7. The ultrasonographic finding of the uterus was inconspicuous in these animals upon the fourth consultation. Ultrasonographic examination of three patients showed neither an increase in a reduction of the uterus lumen nor an increase in vaginal discharge within the first few days. In these animals ovariohysterectomy was performed. Cystic changes of the ovaries and cystic endometrial hyperplasia could be detected intra operationem (Fig. 2). One of the bitches was oestrogen-pretreated, two animals were in the phase of dioestrus.

In 48 of the 52 bitches, a clinical healing of the pyometra could be achieved. This corresponds to a short-term success rate of 92.3% (Table 2).

**Medium-term success up to month 3 after treatment**

The general and reproduction health of 41 bitches could be followed (Table 2). Four of these animals developed a recurrence within 3 months (recurrence rate of 9.8%). In these animals an ovariohysterectomy was performed. In three cases, a cystic change of the ovaries and the endometrium could be detected. None of the bitches was pretreated with hormones. The remaining 37 bitches did not show any abnormalities or signs of a new metropathy during this period of time.

**Long-term success during, at least, 1 year after treatment**

The further course could be observed in 37 bitches. Seven of these animals developed a pyometra within 1 year (recurrence rate 18.9%). In five of these animals, ovariohysterectomy was performed by general veterinary practitioners. For this reason, the findings of the ovaries and the uterus are unknown. In two animals, the treatment with aglepristone was successfully repeated; they did not show new signs of an uteropathy over a period of, at least, 12 months (Table 2).

Data on the further reproductive behaviour of totally 37 bitches are available. In 22 bitches, the next heat started at the expected time. The course of the heat in these animals was normal. Five of these animals developed a pyometra in the stage of dioestrus following oestrus. Seven animals showed premature signs of subsequent heat. One of this bitches developed a pyometra 8 weeks after oestrous behaviour. In eight bitches the period of anoestrus was prolonged. In one bitch the following heat was delayed by 6 months so that the length of the interoestrus interval was 12 months. However, a regulation to the normal cycle could be detected in seven animals until the second heat after treatment. One bitch with delayed interoestrous interval developed a pyometra. Six of the bitches were bred during the time until the end of the observation period, five delivered at least one litter of whelps.

**Table 2. Recurrence and healing rates of bitches with pyometra up to 21 days, 3 month and at least 1 year after medical treatment with aglepristone**

<table>
<thead>
<tr>
<th>Time Period</th>
<th>Total no. of bitches</th>
<th>n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Short-term up to day 21</td>
<td>52</td>
<td>100.0</td>
</tr>
<tr>
<td>Total no. of bitches</td>
<td></td>
<td>52 (100.0)</td>
</tr>
<tr>
<td>No emptying of the uterus</td>
<td>3</td>
<td>5.8</td>
</tr>
<tr>
<td>Initial emptying of the uterus</td>
<td>49</td>
<td>94.2</td>
</tr>
<tr>
<td>Healing rate</td>
<td>48</td>
<td>92.3</td>
</tr>
<tr>
<td>Medium-term up to month 3</td>
<td>41</td>
<td>100.0</td>
</tr>
<tr>
<td>Total no. of bitches</td>
<td></td>
<td>41 (100.0)</td>
</tr>
<tr>
<td>Recurrence rate</td>
<td>4</td>
<td>9.8</td>
</tr>
<tr>
<td>Long-term, at least 1 year</td>
<td>37</td>
<td>100.0</td>
</tr>
<tr>
<td>Total no. of bitches</td>
<td></td>
<td>37 (100.0)</td>
</tr>
<tr>
<td>Recurrence rate</td>
<td>7</td>
<td>18.9</td>
</tr>
</tbody>
</table>

**Discussion**

The aim of this clinical study was to evaluate the success of the treatment of canine pyometra by the antigestagen aglepristone and to document the lasting therapeutic success in different time stages.

The information obtained by anamnesis and the clinical findings reflect the already known data from literature (Blendinger and Bostedt, 1991; Berchtold, 1997). Therefore, it can be assumed that the population examined in this study is representative of bitches diseased with pyometra.

The fact that a conservative treatment of the closed form of pyometra can be successfully carried out, in contradiction to the recommendations made by Johnston et al. (2001), seems to be noteworthy. In seven of the eight animals diseased with this form of pyometra, the opening of the cervix and thus an emptying of the uterus could be induced. Only one bitch had to undergo surgery because no vaginal discharge occurred. None of the seven successfully treated bitches developed a recurrence. These data confirm with Hoffmann et al. (2000) and...
Fieni et al. (2001a), who have been shown that bitches with a closed form of pyometra can be treated successfully.

In 98.1% of the treated bitches, an improvement of the general condition developed within the first two days after the first application. In 94.2% of the cases an initial induction of the emptying of the uterus could be realized within 12–24 h, along with an increase in the vaginal discharge.

A normalization of leucocytosis could be realized in each case during the following 21 days after beginning of treatment. In none of the animals an increase in renal parameters or undesired effects were observed.

With a short-term healing success rate of 92.3% the use of aglepristone is certainly justifiable, at least in order to gain the possibility of postponing surgery in the case of a poor general health condition and associated increased surgical and anaesthetic risk. For medium and long-term success, however, it is important that the participation of relatively or absolutely increased oestrogen values can be excluded. In case of an absolutely or relatively increased oestrogen concentration, such as in relation to the ovarian cysts, a successful treatment cannot be expected. Lemmer (1999) also indicated already that a conservative treatment of pyometra is not successful in the presence of ovarian cysts.

In most cases of recurrence, the presence of cystic changes of the ovaries and the endometrium could be detected. Unfortunately, it was not possible to examine the individual plasma hormone concentrations. It is concluded that the recurrence rate can be minimized by the selection of bitches without ovarian cysts and cystic endometrial hyperplasia. One possibility of detecting oestrogen influence without determining the exact oestrogen value is the performance of an exfoliative vaginal cytology (Tammer et al., 1994). Ultrasonographic diagnosis of ovarian cysts is also possible in some cases. But it is not possible to differentiate between oestrogen-producing ovarian cysts, clinically irrelevant paraovarian cysts and corpora lutea with a fluid filled cavity by ultrasonography (Wehrend and Bostedt, 2002).

To compare the healing and recurrence rate in the present study with the results of other authors it might be helpful to calculate the final recurrence rate from our data. In our opinion this give false information because not all of the animals could be followed up for the same period of time.

There is a significantly higher number of recurrences after aglepristone treatment in the present study in comparison with the study by Hoffmann et al. (2000). In this study, 21 of 22 successfully treated bitches could be further examined during a period of 14 months on average. Potential recurrence in three of these animals could not be commented on, as they were killed due to another clinical picture. In seven animals, no repeated heat could be detected within the observation period. In only one case of the residual 11 animals, purulent vaginal discharge occurred again. Two of the bitches were bred; one of them had a miscarriage for unknown reasons during the last quarter of pregnancy. Unfortunately, the data do not show during which period of time recurrence took place. Only a total average observation period of 14 months and no individual courses of time are given. Fieni et al. (2001a) treated bitches with the closed and open form of pyometra with a combination of aglepristone and cloprostenol in comparison with the treatment with only aglepristone. The global success rate on day 90 after first treatment was 87% with cloprostenol and aglepristone versus 63% with aglepristone alone. Nevertheless, they used very small doses of cloprostenol, 68% of the bitches showed side-effects like vomiting. The authors gave no clear information about the recurrence rate after day 90 following the first treatment.

Johnston et al. (2001) gave an overview of healing success and recurrence rates after conservative treatment of pyometra with progestagens. However, the underlying clinical studies included only low number of bitches. The therapeutic success varies between 100% (n = 10) and 46% (n = 15); all known side-effects such as restlessness, hypersalivation, vomiting and diarrhoea were described in these studies. The recurrence rates are 10% on average. There are regularly no data described on the times of the recurrence of the disease. Only Meyers-Wallen et al. (1989) offer more details on the number of recurrences. They describe a therapeutic success in all of the 10 treated animals. In 40% of the bitches under follow-up examinations a recurrence could be observed within 1 year, in 77% of the bitches pyometra symptoms recurred within 27 months.

Before starting antigestagen treatment, the advantages and disadvantages of the treatment have to be discussed with the owner. Due to the distinctive improvement of the general condition a trial of the treatment can be justified, because a surgery at a later time, after health condition has improved, is still possible. The good conception rate of the subsequent breedings allow the use of antigestagens especially in breeding bitches; however, the results regarding further fertility have to be secured by further examinations in a higher number of bitches.

References


A study of two protocols combining aglepristone and cloprostenol to treat open cervix pyometra in the bitch

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Abstract

To compare the efficacy and safety of two protocols using a combination of aglepristone and cloprostenol for the treatment of open cervix pyometra in the bitch and to describe the progesterone (P₄) serum profiles before and during treatments, 15 bitches were randomly allocated into two treatment groups: I (n = 8): aglepristone was administered at 10 mg/kg, s.c., on Days 1, 3, 8, and 15 (if not cured), combined with cloprostenol at the dose of 1 μg/kg, s.c., on Days 3 and 8, and II (n = 7): received the same treatment with aglepristone as Treatment I but cloprostenol on Days 3, 5, 8 10, 12, and 15 (if not cured). Before the beginning of the treatments and then on Days 8, 15, and 29 all bitches were evaluated for clinical signs, side effects, hemogram, serum P₄ concentrations, and uterus diameters. Bitches in both treatment groups, with (n = 6) or without (n = 9; ≥1.2 ng/ml) initial basal P₄ serum concentrations, achieved treatment success without side effects and no significant differences, either on Day 15 (6/8 for Treatment I and 4/7 for Treatment II) or on Day 29 (2/8 for Treatment I and 3/7 for Treatment II). In both treatments groups, clinical signs, blood parameters, and uterine diameters improved to normal values throughout the experiments. A significant interaction between day and treatment was found for percentage change in P₄ when all bitches were considered together. Redevelopment of pyometra in the next estrous cycle occurred in 20% of the bitches. One nonrecurrent bitch was mated and whelped a normal litter. It is concluded that these two combined protocols proved to be efficient and safe in reversing clinical signs of open cervix pyometra independently of initial P₄ concentrations and

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that the number of cloprostenol administrations seemed to have an effect on P_4 serum changes throughout treatments.

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Keywords: Pyometra; Bitch; Aglepristone; Cloprostenol

1. Introduction

The cystic endometrial hyperplasia—pyometra complex (CEH-P) is a common progesterone (P_4)-dependent disease of the genital tract that appears clinically either in the diestrous or anestrous period of the canine estrous cycle [1–4]. Medical treatment of CEH-P is usually required for bitches intended for breeding. Either prostaglandins (PG) or antiprogestins have been proved to be effective for this purpose [3,5–8]. Prostaglandins increase myometrial contractions, may enhance cervical relaxation and have a luteolytic effect after Day 5 of diestrus, decreasing serum P_4 concentrations [2]. Cloprostenol sodium is a synthetic PG F2 alpha analogue that has a luteolytic effect and potent uterotonic activity [2,8].

Antiprogestins are synthetic steroids which bind with great affinity to P_4 receptors without any effects of P_4 [9]. Aglepristone (RU 534) is an antiprogestin, recently marketed for veterinary use, which competes for uterine receptors with a fixating rate three-fold that of P_4 in the bitch [10]. In human beings, a combination of antiprogestin (mifepristone, RU 486) and PGs have been successfully used to induce abortion [11]. A combination of PG and aplepristone has shown the best results for the treatment of canine CEH-P as recently reported in two studies [12,13]. In the first study, an improvement in success rate of 22 and 32% was found on Days 14 and 28, respectively, when compared with antiprogestin-treated bitches [12]. Progesterone serum concentrations during these combined protocols, although potentially useful to describe their mechanism of action, have not been reported. Moreover, further work is necessary to determine the optimum administration of these combined treatments.

Therefore, the objective of this study was two-fold: to compare the clinical efficacy and safety of two different administration intervals for cloprostenol combined with aplepristone for the treatment of open cervix CEH-P, and to describe the P_4 serum profiles before and during treatment in this species.

2. Materials and methods

2.1. Animals

Fifteen mixed and purebred bitches, ranging from 16 months to 15 years of age, weighing 4–50 kg, with open cervix CEH-P (defined as an enlarged fluid-filled uterus and vaginal discharge) were recruited and included in this study. Diagnosis of CEH-P was confirmed by routine uterine ultrasound findings [14]. All the bitches had serum urea and creatinine <60 and 1.5 mg/dl, respectively, after restoration of normal hydration.
2.2. Procedure

The bitches were randomly allocated to one of the following groups: Treatment I \((n = 8)\): aglepristone (Alizine®), Virbac, Carros, France) was administered at 10 mg/kg, s.c., on Days 1, 3, 8, and 15 (if not cured) and cloprostenol (Estrumate®, Schering Plough, Bs. As, Argentina) far from feeding time was administered at the dose of 1 μg/kg, s.c., on Days 3 and 8; or Treatment II \((n = 7)\): the bitches received the same treatment with aglepristone as for Treatment I, but the cloprostenol (same dose) was administered in the same way on Days 3, 5, 8, 10, 12, and 15 (if not cured). Day 1 was considered as the day of presentation of the bitch.

A combination of amoxycillin–clavulanate at 12.5 mg/kg (bid p.o., Clavamox®, Pfizer, Bs. As, Argentina) and supportive hydration were administered during both therapeutic protocols. Before the beginning of the treatments on Day 1 and then on Days 3, 8, 15, and 29 (if not cured), all bitches were evaluated for body weight and temperature, hydration, anorexia, polyuria/polydipsia, uterus total, and lumen diameters assessed by ultrasonography (Pie Medical S100, 5 MHz transducer, Maastricht, The Netherlands), and vulvar discharge. On Days 1, 8, 15, and 29 (if not cured), blood samples were taken for hemogram and serum P₄ concentrations (Coat-A-Count, DPC®, Los Angeles, CA) determinations. Animals were observed for possible side effects during the treatments. All the bitches were followed up to their next estrous cycle.

2.3. Statistical analyses

Categorical data for the frequency of bitches achieving clinical success (defined as recovery to general health and ultrasonographic observation of normal uterus) and side effects either for Treatments I and II, or for bitches with initial basal or nonbasal P₄ serum concentrations were analyzed by PROC FREQ [15] on Days 15 or 29 (bitches not cured on Day 15). For this purpose, P₄ serum concentrations on Day 1 were categorized as basal or nonbasal (< or ≥1.2 ng/ml, respectively).

Percent change of serum P₄ concentrations ((final value [Days 8, 15, or 29]–initial value [Day 1]/initial value) × 100) was analyzed by least-squares analysis of variance using the General Linear Model procedure PROC GLM [15]. The mathematical model included the main effects of treatment (I or II) and day (8, 15, or 29) and the treatment by day interaction. Descriptive statistics of all parameters assessed were analyzed by PROC MEANS [15] and expressed as LSM ± S.E.M. The level of significance was set at \(P < 0.05\).

3. Results

All bitches in Treatments I (8/8) and II (7/7) achieved treatment success either on Day 15 (6/8 for Treatment I and 4/7 for Treatment II) or 29 (2/8 for Group I and 3/7 for Treatment II) of the protocols. No significant differences in achieving success was found between treatments or initial P₄ concentrations either on Day 15 or 29. None of the bitches showed either systemic or local side effects in relation to the treatment (0/15).
Vulvar discharge was increased in all the bitches within the 24–48 h after the first administration of aglepristone with an improvement in general health condition. Body temperature, hydration, appetite, and polyuria/polydipsia began to improve markedly to normalcy from Day 3 in both groups. Hemogram parameters had a clear tendency toward normal values at the end of the treatments, being the white blood cells within physiological range at that time (Fig. 1). Uterine diameters diminished to normal size and the lumen was undetectable or without contents on Days 15 or 29 in both groups (Fig. 2).

Progesterone serum concentrations before and during Treatments I and II in bitches either with basal or nonbasal initial P₄ are represented in Fig. 3. Progesterone showed a decreasing tendency in three of the four subgroups.

A significant interaction between day and treatment was found for P₄ percentage change when all the bitches were considered together and when only the bitches with nonbasal initial P₄ concentrations (n = 9) were analyzed (P > 0.05). The number of bitches (n = 6) with basal initial P₄ concentrations was statistically insufficient to be analyzed.

Redevelopment of CEH-P in the next estrous cycle occurred in 3/15 old bitches (7–15 years old); two of these had had more than two previous episodes of CEH-P. One bitch was ovariohysterectomized before her next cycle. Nonrecurrence in the next cycle occurred in 11/15 bitches; four of these were treated with amoxicillin during the next open cervix period (proestrus and estrus). One (2 years old) of these four bitches was mated and whelped a normal litter.

Fig. 1. Least square means of the white blood cells (WBC) of 15 bitches suffering cystic endometrial hyperplasia—pyometra complex treated with two combined protocols of PG and aglepristone before and during treatments. No significant differences in WBC were found between treatment, so they were represented together. Bars on symbols represent the corresponding S.E.M.
Fig. 2. Least square means of uterine total and lumen diameters assessed by ultrasonography of the same animals ($n=15$) before and during the treatments. Bars on symbols represent the corresponding S.E.M.

Fig. 3. Least square means of serum $P_4$ concentrations of the same animals ($n=15$) divided into four subgroups according to treatment (I or II) and initial (Day 1) $P_4$ concentrations (basal or nonbasal, $<1.2$ ng/ml, respectively) before and during treatment. Bars on symbols represent the corresponding S.E.M.
4. Discussion

The proportion of bitches with initial basal P₄ concentrations in the present trial was higher than that reported in two previous studies [3,12]. Although this finding was probably incidental, it does confirm that this complex can become clinically evident either during diestrus or anestrus [1,6].

In line with a recent study but in contradiction with a previous one, in which only aglepristone was used, no relationship between initial basal P₄ concentrations and treatment success was found in this study [12,16].

Some bitches were cured without achieving basal P₄ serum concentrations at the end of the treatments. Moreover, pregnancy termination with aglepristone in the bitch occurs in the presence of high P₄ serum concentrations [17]. These findings seem to confirm that antiprogestins decrease intrauterine P₄ through the number or sensitivity of P₄ receptors and not through the serum concentrations of this hormone [12]. The initial serum rise (on Day 8) of a nonbasal group could be explained by the blocking of uterine receptors and the consequent elevation of P₄ in blood. Ecbolic PG effect could have also contributed to success in these cases.

More difficult to explain is the clinical improvement in bitches with basal P₄ concentrations throughout treatments. Although the uterotonic effect of PG could have caused the success in these cases, clinical improvement and the increase in the vaginal discharge was seen before the first treatment with PG on Day 3.

Although in the present study, cloprostenol was administered at 48 h or longer intervals, the results on Day 15 (6/8 and 4/7 for Treatments I and II, respectively) were better than those reported in another study (50%), using the same drug and doses on a daily basis [12]. In the current study, based on two treatment protocols for cloprostenol, the number of treatments did not seem to be crucial in the resolution of the clinical cases, although a larger number of treated bitches would be necessary to either confirm or reject this initial finding. Conversely, the number of PG treatments seemed to influence P₄ serum changes during the different days of the study.

In disagreement with two recent studies [2,12], but in line with a previous report in which only aglepristone was used [5], no side effects were reported in this study. The administration of a low dose of a potent PG at a time far from feeding could have accounted for these differences. Moreover, all the bitches were in a better condition on the day of the first cloprostenol injection (Day 3) than at the beginning of the treatments.

In this study, recurrence rate was higher than in a similar previous study in which no redevelopment appeared in 12 bitches that were followed up to the next estrous cycle [12], and also higher than in one study in which only aglepristone was used [16]. The higher recurrence rate may be due to the advanced age of most of the bitches used in the present study. Conversely, the present results were in the lower limit range of recurrence for bitches treated only with PG (10–77%) [6,7]. These findings suggest that the uterotonic effect of PG does not reverse CEH and many dogs may have a decrease in clinical signs towards subclinical levels which then becomes undetectable. We hypothesized that antiprogestin treatment might better manage endometrial abnormalities depending on the depth of inflammatory infiltration measured using Dow’s classification [18]. Conversely, a study in
which histological examination of uteri was carried out revealed no differences between 6 days-antiprogestin-treated and control bitches [3].

Consistent with a previous report [5], fertility after treatment did not seem to be affected by endometrial pathology in the young bitch that was mated. This young bitch was one of the most representative of the population for which these combined protocols are mainly intended in clinical practice. Further studies including larger number of bitches of different ages, with or with out past episodes of CEH-P are required to evaluate fertility after treatment.

We concluded that these two combined protocols proved to be efficient and safe in reversing clinical signs and abnormal uterine ultrasonographic findings in bitches suffering CEH-P, independently of initial P4 serum concentrations. Also, the number of PG administrations seemed to have an effect on P4 serum concentrations throughout treatments. Further work with a larger number of bitches is necessary to confirm these initial findings.

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References


ABSTRACTS

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&

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"Reproductive biology and medicine of domestic and exotic carnivores"

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DIAGNOSIS OF ENDOMETRITIS IN THE INFERTILE BITCH: A NEW APPROACH

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Introduction - Very few reports have been made about endometritis in the bitch; its relationship with failure to conceive remains unclear. It may be due to the difficulty to collect uterine samples for further investigations. Today, transcervical catheterization by vaginal endoscopy allows us to evaluate the endometrium in infertile bitches. Diagnosis criteria were determined according to previous studies on uterine cytology and bacteriology[1-3]. The aim of our study was to test the efficiency of this technique to diagnose endometritis in the bitch and furthermore, to evaluate the incidence of endometritis within infertile bitches.

Material and methods - 26 bitches presented for infertility in Alfort Veterinary College were included in this study. Classical infertility investigations were not indicative: time of mating had been correctly determined by progesterone assays, male used had sired successfully, clinical examination and genital ultrasonography revealed no abnormalities. A vaginal endoscopy was performed and presence of vaginitis and cervical discharge was evaluated. In all bitches, a transcervical catheterization was performed using a human ureteral catheter (Ureteral CRU® ch.6 223602). Flushing of the uterine lumen was realized with sterile saline fluid (NaCl 0.9%, 2mL/10 kg instilled then reabsorbed) and collected samples were used for uterine cytology and aerobic bacteriology (Amies agar gel with charcoal). The normal leucocyte score, which reflected absolute and relative numbers of leucocytes versus endometrial cells, was defined by Watts [3]. All cytologies exceeding normal scores were considered as endometritis.

If cytology pointed out an inflammatory state correlated to bacterial heavy growth, the bitch was considered to suffer from infectious endometritis. A cytologic inflammatory state of the uterus in the absence of bacterial growth was considered to be a non infectious endometritis.

Results - Fourteen different breeds were concerned, from Shi-Tsu (5kg) to Mastiff (96kg). Bitches were mainly large and giant breeds and were aged 1 to 7 years (mean 4 years/SD=1.5). Uterine investigations were realized in dioestrus (21 bitches), anoestrus (4) and pro-oestrus (1). 10/26 bitches suffered from endometritis. Among them, seven suffered from infectious endometritis. Three bitches had a bacterial heavy growth without cytologic abnormalities and we considered that it was a sign of contamination by the normal vaginal flora. In 4/10 bitches suffering from endometritis, a cervical discharge was observed by vaginal endoscopy. Surprisingly, another bitch negative towards cytological and bacteriological criteria was found to show also a purulent cervical discharge. A clear cervical inflammation was also observed in 3/7 bitches suffering from infectious endometritis. Signs of vaginitis were visualised in two bitches, both of them suffering from infectious (1/10) and non infectious (1/10) endometritis. One bitch suffering from infectious endometritis presented signs of vaginitis, cervicitis and cervical discharge. Infectious endometritis was diagnosed during dioestrus (6/7) and pro-oestrus (1/7). Non infectious endometritis was diagnosed in dioestrus (2/3) and anoestrus (1/3). Four bitches for which endoscopy was performed in dioestrus encountered pyometra after flushing, two of which were not initially suffering from endometritis. Five bitches suffering from endometritis were further bred after treatment combining antibiotics +/- aglepristone (Alizine ®) and they all went pregnant. 4/5 bitches whelped normally and 1/5 had a premature parturition at 59 days of pregnancy.
**Discussion** - In our study, endometritis seemed to have in most cases an infectious origin (70% of affected bitches), but these results may be underestimated, as some other pathogens (anaerobic bacteria, mycoplasms, fungi), were not searched for. Dioestrus seems to be the best period to diagnose endometritis. However, the endometrium impregnated with progesterone is more sensitive and despite all precautions, this could explain that we got induced pyometra after endoscopy. Early anoestrus may be a more adequate period to perform those investigations, as progesterone impregnation is over. One bitch showed a purulent cervical discharge without a positive endometrial cytology. Our diagnosis technique lacks comparison with histology, which should be done to ensure a more accurate diagnosis. Indeed, impact of endometritis is underestimated. Endometritis, in our opinion, should be investigated in each unexplained case of infertility in bitches. The technic used here seems reliable: all bitches treated were bred successfully whereas previous infertility treatments had not succeeded. Defining more accurate criteria will improve the efficiency of this non invasive technique that could help to treat unexplained infertility cases.

**References**

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MEDICAL TREATMENT OF THE CANINE PYOMETRA

KONRAD BLENDINGER

Three conditions are grouped to the Cystic-Endometrial-Hyperplasia (CEH)-Pyometra-Complex: 1. Cystic endometrial hyperplasia (enlargement of the uterine glands forming cysts which may be as small as 1 mm or as large as 1 or more cm). 2. Endometritis (infection of the uterus with uterine intraluminal diameter of less than 1 cm). and 3. Pyometra (infection of the uterus with uterine intraluminal diameter of more than 1 cm). Most of the cases with infection are combined with a varying degree of CEH (cystic endometrial hyperplasia). The three diseases or mixed cases may be presented with the same symptoms and the appearance of the diseased uterus may vary dramatically.

In one study, the clinical signs are vaginal discharge (85%), lethargy to depression (65), anorexia to inappetence (43%), polyuria and/or polydipsia (28%), vomiting (15%), nocturia (5%), diarrhea (5%), abdominal enlargement (5%). Other studies find polyuria/polydipsia and vomiting somewhat more frequent. The bitches are presented at different stages of the oestrous cycle. Diestrous (58%), anoestrous (19%), signs of oestrus (6%), abnormal cycle (6%), pretreated with progestagens (12%), pretreated with estrogens (3%).

Most of the pyometric bitches are under the influence of progesterone, which is one reason for the disease considered to be hormonally (progestagen)-induced.

Is pyometra an emergency?

Each case of uterine disease in the bitch has primary to be considered as an emergency because of its potential of uterine rupture and death from septicaemia. Nevertheless, there are bitches with pyometra that get rid of all the symptoms without any treatment and show the signs again during next diestrous – probably with a worse outcome (cyclic appearance of pyometra).

Influences that shape the uterus:

The shape of the uterus varies dramatically under diseased conditions. Different influences act on both the endometrium and myometrium. Progesterone: inflow of the cervix, stimulation of secretory cells, proliferation, increase of bacterial adherence to the endometrium. Estrogens: Proliferation, vascularisation. Prostaglandins: Enhancement and part of an inflammatory reaction. Bacteria (with or without toxins): Local cell irritation on endometrium and myometrium, uterine ulceration, uterine rupture. In case of pyometra, there are endometrial prostaglandin-metabolites detectable, in case of mucometra, they are not detectable (Hagman, 2005).

Assessment of an individual case:

If medical treatment is considered in an individual case, more information is needed than if ovariohysterectomy should be performed. The core assessment is: General examination, diameter of uterine lumen, uterine content (pus, mucus, blood, retained fetal membranes, retained fetus). Complete blood count (or at least HTC, RBC, WBC, BUN, Creat). If medical treatment is considered, additional information is necessary. Close history of previous reproductive cycles, “closed” or “open” cervix, hormonal state (at least progesterone and estrogens), thickness of the uterine wall. To assess the estrogen influence to the tissue, a simple vaginal smear provides good information. Progesterone may be estimated by history of the reproductive cycle or an inhouse semiquantitative ELISA – test, or by accurate laboratory measurement.

The Cystic Hyperplasia of the Endometrium (CHE) represents a hormone-mediated gradual change of the uterine lining. Its presence predisposes the uterus to infection.

Pyometra is an infection overlying CHE with subsequent formation of intrauterine purulent fluid. Nulliparous animals may be predisposed. Animals with a history of false pregnancy are not predisposed. Diagnosis requires demonstration of uterine enlargement and presence of purulent intrauterine fluid. Neutrophilia with a regenerative left shift and monocytosis are often present. Secundary renal disease may occur subsequent to endotoxemia (E. coli). Ovariectomy is the treatment of choice in most cases. Medical treatment may be attempted in some special conditions.

Indications for medical treatment of canine pyometra:

Preclusion of uterine rupture or secondary renal disease.

Intention of further breeding.

Preparation of patients with high anesthetic risk to surgery.

Breeds with a high risk of urinary incontinence after OHE.

The owner absolutely refuses surgery.

It is important for the pet owner, to know that medical treatment is not the cheap way to deal with the disease. It may be even more expensive than surgery.

Drugs for medical treatment of canine pyometra:

1. Antibiotics - indicated in every case with presence of bacteria - even in the absence of bacteria (e.g. mucometra) because of the risk of superinfection.

- G- and G+ efficacy
- according resitogram as soon as possible
- Amoxicillin/Cloxacillin
- Quinolones (Enrofloxacine/Marbofloxacine)
- Cefalosporins

2. Prostaglandin F2alpha

The effects of prostaglandin F2alpha on the female reproductive system include contraction of the myometrium (immediate effect), luteolysis (delayed effect), relaxation of the uterine cervix (least consistent effect). The corpus luteum in the bitch is refractory to PGE action during the first 1-2 weeks. After 1-2 weeks, it takes about 5 days until luteolysis is achieved.

There are several side effects / reactions to prostaglandin F2alpha in the dog:

<table>
<thead>
<tr>
<th>Reaction</th>
<th>(dose: 0.1 mg/kg nat. PGF2alpha)</th>
<th>Percent of Dogs (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Restlessness</td>
<td></td>
<td>85</td>
</tr>
<tr>
<td>Pacing</td>
<td></td>
<td>85</td>
</tr>
<tr>
<td>Hypersalivation</td>
<td></td>
<td>82</td>
</tr>
<tr>
<td>Panting</td>
<td></td>
<td>79</td>
</tr>
<tr>
<td>Vomiting</td>
<td></td>
<td>73</td>
</tr>
<tr>
<td>Abdominal pain or cramping</td>
<td></td>
<td>61</td>
</tr>
<tr>
<td>Tachycardia</td>
<td></td>
<td>55</td>
</tr>
<tr>
<td>Fever</td>
<td></td>
<td>33</td>
</tr>
<tr>
<td>Defecation</td>
<td></td>
<td>30</td>
</tr>
<tr>
<td>Uterine evacuation</td>
<td></td>
<td>30</td>
</tr>
</tbody>
</table>

The side effects are dose dependent and decrease / disappear with repeated administrations.
Dosage for medical treatment of pyometra:
- Natural PGF2α (dinoprost):
  - 0.02 mg/kg SC, 2–3 times/day duration: 7–10 days until uterine content has evacuated
or
- 0.1 mg/kg once on day 1
- 0.2 mg/kg on day 2
- 0.25 mg/kg on day 3–7
- Synthetic PGF2α (cloprostenol)
  - Longer duration of action
  - 0.001 – 0.002 mg/kg SC once daily

3. Antiprogestin
Agleriprostone is commercially available in several countries. It is a competitive antagonist of progesterone on the receptor level thus overcoming most clinical effects in the presence of proges
terone: The uterine cervix consistently opens (day 1–3 after beginning of treatment). Antiprogestins stop the progesterative inhibitory effect to myometrial contractions, they stop the prog
estative immunosuppressive effect. It is important to be aware, that antiprogestins have no intrinsic luteolytic effect.

Dosage for medical treatment of pyometra:
- Agleriprost 10 mg/kg SC day 1, day 2, day 7
- Probably repeated injections once a week as long as natural progesterone is present / active.

4. Dopaminergic drugs / Antiprolatins
Antiprolatins initiate luteolysis (delayed action, comparable to the luteolytic effect of prostaglandins) and stop prolactin produc
tion (IT present).
Dosage
- Cabergoline 0.005 mg/kg PO once daily for 7–10 days or some times longer. It should be administered as long as uterine content has evacuated.

5. Supportive therapy
In general, the treatment of bitches with pyometra should be initiated according general condition and results of CBC:
- IV fluids (Lactated Ringer’s sol / Glucose 5%)
- H2-blocker: Metoclopramid (in case of gastric injury / vomiting)
- Antiinflammatory drugs (NSAIDs)
Close observation of the bitch during the first days of treatment has to be ensured.

Monitoring:
- Repeated general condition (once or twice daily)
- Repeated ultrasound – decrease of uterine content (every 2 – 3 days)
- Repeated complete blood count (every 1 – 2 days)
- Repeated intravenous fluids as necessary

Results from several studies

Prostaglandins:
Natural prostaglandin was given on day 1: 0.1 mg/kg SC; day 2: 0.2 mg/kg, day 3–7: 0.25 mg/kg. Antibiotics 14 d; Re-evaluation at days 7 and 14; Re-treatment in case of purulent vaginal dis
crage persists, fever, increased WBC, or fluid-filled uterus pers
ists.

Results in open-cervix pyometra:
- Complete resolution of their uterine infection in 153 of 163 cases (94%)
- Whelped litter: 98% (of the 153)
- 1/4 had more than one litter
- Breeding in first cycle after treatment is strongly recom
mended because of the risk of recurrence due to uterine desease, because pregnant bitches may be less suscepti
ble to infection and because there is no benefit from skip
ping a cycle.

Results in closed-cervix pyometra:
These were clinically more severe cases. Complete resolution of their uterine infection occurred in 16 of 53 cases (31%), 12 of 16 required 2 series of treatment. All (1) 16 successful treated bitches subsequently whelped a healthy litter.

Antiprogestins:
Relapses after medical treatment of pyometra using antiproges
tins:
Trach et al. 2003 18.9% (7 of 37) after one year
Gobello et al. 2003 29% (3 of 15)
Fieni 2006 19% (4 of 21) after 24 months
Corrada et al. 2006 20.7% (6 of 29)

Antiprogestin + prostaglandin (in this study)

Pregnancies after medical treatment of pyometra using antiprogestins:
Hoffmann et al. 2002 2 5 pregnancies of 6 mated
Gobello et al. 2003 One recurrent was successfully mated
Fieni 2006 One of two mated bitches had a litter

Summary:
Relapse rate of at least 20% during the first two years after medical treatment
Antiprogestins are capable to open the cervix in case of clausure by progesterone activity
Prostaglandin F2α is a valuable drug for open-cervix pyometra
Slowly luteolysis may be induced by prolanct antagonist agents

Conclusion:
Ovariotomy is the therapy of choice for canine pyome
tra because of the high relapse rate in medical treatment.
It is not recommended to treat bitches with uterine ruptures and/or renal diseases
Medical treatment has it’s best outcome in young bitches with open-cervix pyometra
The use of antiprogestins is a good choice in closed cervix situ
ations

Bitches with ovarian cysts and irregular heats are hardly treated successfully

References:
2) Trach K, Wehrand A, Bostedt H (2003): Follow up examina
drostenol to treat open cervix pyometra in the bitch. Theriogenology
Pyometra is a common disease in bitches over eight years of age. The disease generally occurs during the di-oestrous phase of the reproductive cycle (Blendiger et al 1997). Currently, it is generally accepted that cystic endometrial hyperplasia (CEH)-pyometra complex is very closely associated with increased circulating blood progesterone concentrations (Noakes et al 2001). Progesterone promotes multiplication of the glandular cells in the uterus, which may result in cystic endometrial hyperplasia. Progesterone also depresses local mechanisms that clear bacteria from the genital tract. Infections with *E. coli*, ascending the urogenital tract, from the bowel, have been identified as the most common bacterial component of the canine pyometra complex (Hagman & Kunh, 2002, Wadas et al 1996).

Pyometra is associated with inflammation of the endometrium with the accumulation of purulent and/or haemorrhagic secretions in the uterus. Depending on the degree of cervical opening, a vaginal discharge may be present. Without treatment the infection has high morbidity and mortality due to endotoxaemia and septicaemia (Hagman et al 2006).

Surgical treatment of pyometra by ovariohysterectomy is curative. Anaesthetic and surgical risk may be increased due to the systemic effects of pyometra and these must be addressed before, during and after surgery.

Due to the luteolytic and uterotonic properties of prostaglandin-F2 alpha (PGF₂α), the repeated administration of PGF₂α has been used to treat pyometra (Gilbert et al 1989, Meyers et al 1986). To avoid the risk of serious side effects, prostaglandins are only recommended for use in young bitches with no liver, kidney or cardiovascular disease. The duration of treatment and the onset of side effects related to the action of the prostaglandins on smooth muscle, causing diarrhoea, salivation and vomiting, do not make it an ideal single modality treatment option for pyometra.

Given the major role of progesterone in inducing CEH-pyometra complex, anti-progesterins such as aglepristone¹ could be expected to be useful in a medical treatment protocol. Aglepristone is a potent progesterone antagonist, with an affinity for progesterone receptors in vitro three times greater than that of progesterone. Aglepristone binds to uterine progesterone receptors without producing the biological effects of progesterone. Aglepristone has been shown to effectively suppress the biological action of progesterone during gestation, interrupting gestation (Fieni et al 1996, Galac et al 2000, Fieni et al 2001a), and causing cervical opening and inducing parturition (Fieni et al 2001b). The molecule has been effectively used in the treatment of uterine infections associated with the presence of a high level of plasma progesterone (Blendiger et al 1997, Breitkopf et al 1997, Hoffman et al 2001). Recent work has described the use of aglepristone to treat pyometra, for a short or medium term after the initial acute recovery (Trasch et al 2003), including its combined use with cloprostenol (Gobello et al 2003).

In a recent large clinical study (Fieni, 2006) the use of aglepristone, alone and in combination with low doses of cloprostenol, was evaluated in the treatment of metritis-pyometra. Sixty-seven bitches, aged 8.2 ± 3.5 years, with a uterine infection were accordingly allocated into three groups: group 1 – metritis (n=15); group 2 – open pyometra (n=35); and group 3 – closed pyometra (n=17). All bitches were injected subcutaneously three times with 10mg/kg of aglepristone on Days 1, 2 and 8. It was the only treatment administered to bitches with metritis. In addition to this treatment, half the bitches with open and closed pyometra were injected daily from Day 3 to Day 7 with 1μg/kg cloprostenol² subcutaneously. All pyometra bitches were concurrently treated with amoxicillin/clavulanic acid antibiotics³. All bitches were

¹Alizin, Virbac
²Estrumate, Schering-Plough Animal Health
³Synulox, Pfizer
closely monitored with regular clinical and gynaecological examinations and full blood counts. The efficacy criteria were assessed as recovery of general health, lack of vulvar discharge and lack of uterine lumen enlargement. If necessary an additional treatment with aglepristone was administered on Days 14 and 28. The final examination was on Day 90.

At Day 90, 80.6% (54/67) of the bitches were cured including all of the bitches with metritis (15/15), receiving only aglepristone; the closed pyometra cases (17/17) had cervical opening within 48 hours of receiving aglepristone. In the bitches with open or closed pyometra, additional treatment with cloprostenol from Days 3 to 7 significantly improved the global success rate at Day 90 to 84.4% (27/32) compared to 60% (12/20) in bitches without cloprostenol (p<0.05).

It was concluded that aglepristone was effective and safe to treat metritis and an effective means of inducing cervical opening in cases of closed pyometra. The combination of aglepristone and cloprostenol was more effective in the medical treatment of open and closed pyometra than aglepristone alone.

A review of records between 2001 and 2006 from Applecross Veterinary Hospital showed 56 cases (11 cats and 45 dogs) where aglepristone protocols had been used to treat pyometra. Five protocols had been used in treatment: a) Single – single subcutaneous injection of aglepristone 10mg/kg on Day 1; b) Standard – subcutaneous injections of aglepristone 10mg/kg on Days 1 and 2; c) Extended – subcutaneous injections of aglepristone 10 mg/kg on Days 1, 2 and 8; d) Standard + Cloprostenol – Standard protocol with daily subcutaneous cloprostenol 1µg/kg starting between Days 4 and 8 and continuing for a maximum of eight injections; and e) Extended + Cloprostenol – Extended protocol with daily subcutaneous cloprostenol 1µg/kg starting between Days 4 and 8 and continuing for a maximum of eight injections. A successful outcome was based on resolution of clinical signs by Day 30 after treatment with or without ultrasound confirmation of a normal uterus post-treatment. All animals received concurrent medical therapy as appropriate including but not limited to antibiotics, intravenous fluids and nutritional support. The results are summarised in Tables 1 and 2.

For all treatment protocols 84.9% (28/33) and 75% (9/12) of bitches, with open and closed cervix pyometra respectively, and 100% (11/11) of queens irrespective of cervical patency had successful outcomes. The numbers in each group are insufficient for meaningful statistical comparison. In successfully treated bitches 18.9% (7/37) delivered pups at a subsequent oestrus after treatment and 10.8% (4/37) had recurrent pyometra at the next diestrus. In successfully-treated queens 54.5 % (6/11) delivered kittens at a subsequent oestrus after treatment and one had recurrent pyometra five months after initial treatment. The remaining 26 bitches and four queens have either not been mated, not conceived, undergone elective ovariohysterectomy or been lost to follow-up.

The reasons for failure of aglepristone protocols to achieve more successful outcomes in the bitch include the presence of focal multi-locular pyometra, client reluctance to pursue medical therapy and rapid clinical deterioration of systemic clinical status, e.g. endotoxaemia requiring ovariohysterectomy. The bitches that underwent ovariohysterectomy within 48 hours of starting treatment did not indicate failure of aglepristone per se but reflected an inability to medically stabilise the patient until the aglepristone effect was expected.

The conservative treatment of pyometra with anti-progestins (such as aglepristone) will not replace ovariohysterectomy, particularly when medical stabilisation of the patient is unlikely in the first 24 hours. However, it is a valuable alternative for the treatment of bitches used for breeding purposes, for dogs with risk of anaesthesia and for those dogs where, for whatever reasons, the owners object to ovariohysterectomy.

### REFERENCES:


FIENI, F (2006) Clinical evaluation of the use of aglepristone with

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### TABLE 1: Number (%) of bitches and queens that achieved successful outcomes based on aglepristone treatment protocols at Applecross Veterinary Hospital (2001-2006).

<table>
<thead>
<tr>
<th>Species</th>
<th>Single</th>
<th>Standard</th>
<th>Standard + PG</th>
<th>Extended</th>
<th>Extended + PG</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bitches</td>
<td>3/5 (60.0)</td>
<td>27/31 (87.1)</td>
<td>2/3 (66.7)</td>
<td>4/5 (80.0)</td>
<td>1/1 (100.0)</td>
</tr>
<tr>
<td>Queens</td>
<td>2/2 (100.0)</td>
<td>8/8 (100)</td>
<td>(0)</td>
<td>1/1 (100.0)</td>
<td>(0)</td>
</tr>
</tbody>
</table>

### TABLE 2: Number (%) of bitches and queens that achieved successful outcomes in response to aglepristone treatment protocols based on cases from Applecross Veterinary Hospital (2001-2006).

<table>
<thead>
<tr>
<th>Species</th>
<th>Open pyometra</th>
<th>Closed pyometra</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bitches</td>
<td>28/33 (84.9)</td>
<td>9/12 (75.0)</td>
</tr>
<tr>
<td>Queens</td>
<td>10/10 (100)</td>
<td>1/1 (100)</td>
</tr>
</tbody>
</table>
Clinical evaluation of the use of aglepristone, with or without cloprostenol, to treat cystic endometrial hyperplasia-pyometra complex in bitches

F. Fieni*

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Abstract

The aim of the study was to evaluate the efficacy of aglepristone (10 mg/kg on days 1, 2 and 8) for the treatment of metritis or pyometra in bitches \( n = 67 \) either alone for cases of metritis \( n = 15 \), or in cases of pyometra \( n = 52 \) with \( n = 32 \) or without \( n = 20 \) the addition of low doses (1 \( \mu \)g/kg) of cloprostenol for 5 days (days 3–7). Examinations performed on day 90, in addition to days 8, 14 and 28, determined that treatments had been curative in the long term in 54/67 bitches (80.6%). Bitches in whom pyometra did not resolve, were given additional aglepristone on day 14 \( n = 38 \) and day 28 \( n = 20 \). Aglepristone alone was curative in 15/15 bitches with metritis. In 17/17 bitches with closed pyometra, cervical opening occurred within 48 h of aglepristone administration. Amongst the 52 bitches with open \( n = 35 \) or closed \( n = 17 \) pyometra, the additional treatment with cloprostenol from days 3 to 7, significantly improved the overall success rate at day 90, which was 27/32 (84.4%), compared to 12/20 (60.0%) in bitches without cloprostenol \( P < 0.05 \). The leucocyte count and plasma progesterone concentrations significantly decreased over the course of treatment. Thirteen of 15 bitches in whom plasma progesterone concentrations were initially low (<3.18 nmol/L) were cured. The recurrence rate after 12 and 24 months was 13.0% (3/23) and 19.0% (4/21), respectively.

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Keywords: Pyometra; Metritis; Aglepristone; Cloprostenol; Bitch

1. Introduction

Cystic endometrial hyperplasia-pyometra complex or metritis/pyometra is a very common disease in bitches over 8 years of age. Depending on the degree of cervical opening, metritis/pyometra may cause inflammation of the endometrium with an intrauterine accumulation of purulent and/or hemorrhagic secretions, similar vaginal discharge, and loss of general condition. Without treatment, the infection is fatal. The disease generally occurs after the bitch has been in estrus (generally during metestrus [1]). Currently, it is generally accepted that the disease is very closely linked to hormonal imbalance in which progesterone plays a major role [2].

Surgical treatment, i.e. ovariohysterectomy, is generally performed if the bitch can tolerate the anesthetic and the owner can bear the expense. Surgery is a radical form of treatment, preventing any further reproduction. Because of the luteolytic and uterotonic properties of prostaglandin-F2 alpha (PGF), the repeated administration of PGF has been used to treat pyometra. The treatment consists of one or more injections daily over a minimum period of 5 days [3,4]. To avoid the risk of serious side effects, prostaglandins are usually only recommended for use in young bitches with no liver or
kidney dysfunction or cardiac disease. Some authors also suggested that the cervix must be open and the uterus non-hypertrophied [5]. The duration of treatment and the onset of side effects related to the action of the prostaglandins on smooth muscle (diarrhea, salivation and vomiting) make this a demanding course of treatment, which usually requires hospitalization of the bitch.

Given the major role of progesterone in inducing cystic endometrial hyperplasia-pyometra complex, antiprogestins such as aglepristone could be expected to be useful in a medical treatment protocol. Aglepristone has been shown to effectively suppress the biological action of progesterone during gestation, interrupting gestation [6–8], causing cervical opening, and inducing parturition [9]. The molecule has been effectively used in the treatment of uterine infections associated with elevated plasma progesterone concentrations (n = 6, 7 and 31) [1,10,11]. Recent work has described the use of aglepristone to treat pyometra in bitches, in terms of the short or medium term result after the initial acute recovery (n = 52) [12], and including its combined use with cloprostenol (n = 15) [13]. However, to be suitable for medical use by practitioners, a treatment needs to be efficient, fast, and prescribed on the basis of immediate clinical findings, without the need for lengthy laboratory testing.

The aim of this clinical study was to evaluate the efficacy and safety of the treatment of metritis/pyometra in bitches by the administration of aglepristone in bitches with various forms of infectious uterine diseases, that were presented to the clinics at the Veterinary School in Nantes, and to evaluate it in comparison with the administration of combined aglepristone and low dose cloprostenol. The latter treatment strategy was designed to improve outcomes and hasten recovery without the side effects usually induced by prostaglandins in bitches with open or closed pyometra.

2. Materials and methods

2.1. Animals

This study involved 67 bitches of different breeds, aged 8.5 ± 3.2 years, with “open” or “closed” forms of metritis or cystic endometrial hyperplasia-pyometra complex. The diagnosis was confirmed by the observation of purulent or hemorrhagic vaginal discharge associated in some, but not all, cases with enlargement of the uterine lumen detected using uterine ultrasonography. Irrespective of their general health, all bitches treated had a serum concentration of urea and creatinine of less than 0.6 g/L and 10 mg/L, respectively.

2.2. Procedure

The bitches were allocated into three groups based on clinical observation: Group 1, metritis (putrid vaginal discharge with no enlargement of the uterine lumen detected on uterine ultrasonography, n = 15); Group 2, open pyometra (putrid vaginal discharge with enlargement of the uterine lumen, n = 35) and Group 3, closed pyometra (enlargement of the uterine lumen without vaginal discharge, n = 17).

Treatment varied, depending on the clinical situation, but all the bitches received one subcutaneous injection of 10 mg/kg body weight of aglepristone (Alizine®; Virbac, Carros, France) once daily on days 1, 2 and 8. This was the only treatment administered in bitches with metritis (Group 1). In addition to this treatment, two-thirds of each group of the bitches with open (Group 2, n = 10/17) or closed (Group 3, n = 21/35) pyometra were given daily subcutaneous injections from days 3 to 7, with 1 µg/kg of cloprostenol (Estrumate®; Schering-Plough, Levallois-Perret, France). Bitches with hyperthermia or dehydration and all bitches with closed pyometra received additional treatment, i.e. intravenous infusion of ringer lactate at day 1 and 24 mg/kg/day of amoxicillin-clavulanic acid from days 1 to 5 (Synulox®; Pfizer, Paris, France).

The bitches from all three groups were checked on days 14, 28 and 90. Bitches in whom pyometra did not resolve, were given additional aglepristone on day 14 (n = 38) and day 28 (n = 20). The efficacy criteria were recovery of good general health, absence of uterine lumen enlargement and absence of vaginal discharge. At the beginning of the trial and at various stages during the study, the bitches underwent a general clinical examination, gynecological examination and an ultrasound examination of the uterus (Aloka Echo Camera SSD-500 with 5 MHz transducer; Aloka, Cergy St. Christophe, France). A blood sample was also taken from the jugular vein. Mean white and red blood cell counts were measured using an impedance counter (MS9; Melet Schloesing Laboratoire, Cergy-Pointoise, France). An automated biochemistry analyzer (BPI44; Ph Diagnostics, Montpellier, France) was used to determine the plasma concentrations of urea and creatinine via photospectrometry. Peripheral progesterone concentrations were measured using a commercial RIA kit (Diria-progk kit from Diasorin, Antony, France). The assay sensitivity was 0.3 nmol/L and assay specificity was 97.5%.
2.3. Statistical evaluation

Qualitative results, such as the success of the treatment, were reported with a confidence interval of 95%. Quantitative results were reported as means ± S.D. Data were analyzed by Statview (SAS Institute Inc., Cary, NC, USA). Treatment efficacy and the clinical response of metritis/pyometra were compared using a Chi-square test. A Student-Fisher test was used to compare the mean decrease in the diameter of the uterine lumen, the mean leucocyte count and the mean peripheral plasma progesterone concentration between treatments at various times during the experiment. Values were considered statistically significant when $P < 0.05$.

3. Results

When measuring efficacy at day 90, 54/67 bitches were cured (i.e. 80.6%, with a confidence interval range of 71.1–90.1%). The overall recovery rate was lower ($P < 0.001$) at day 14 (24/68, 35.3%) than at day 28 (46/67, 68.6%) and day 90 (54/67, 80.6%). The effectiveness of the treatment was not dependent on the clinical form of uterine disease (Table 1).

In bitches with metritis, treatment with aglepristone alone gave a high cure rate, with clinical resolution in over half of the bitches (9/15) within 2 weeks and within 4 weeks for the remainder with, one exception. The bitch cured at day 90 had only a slight serous discharge on day 28. This was visible by the clinician during the genital exam but not by the owner.

Clinically, the administration of aglepristone in cases of pyometra led to changes in the nature of the vaginal discharge, which changed from purulent, to mucous-like, before finally becoming serous, with a concurrent decrease in volume. The last clinical sign recorded was self-cleaning of the vulval area by the bitch. For all bitches with closed pyometra, cervical opening was induced following the first two aglepristone treatments (18/18). The mean time to cervical opening was 25.8 ± 12.2 h. The shortest time was 4 h after the first administration of aglepristone; however, cervical opening was complete within 48 h in all of the bitches. Cervical opening induced enabled the evacuation of large volumes of purulent discharge, which was associated with an immediate improvement in general condition and, in most cases, with increased appetite.

For bitches with open or closed pyometra, additional treatment with cloprostenol from days 3 to 7 improved the overall success rate at day 90 ($P < 0.05$); 27/32 (84.4%) with cloprostenol versus 12/20 (60.0%) without cloprostenol (Table 2). Recovery was accompanied by a gradual decrease in the diameter of the uterine lumen, which became non-detectable ultrasonographically. The mean decrease in the diameter of the uterine lumen over time depended on the treatment used (Fig. 1) and was significantly greater between days 8 and 14 in bitches receiving the combined aglepristone–cloprostenol treatment than in those treated with aglepristone alone (84.8 ± 20.7% versus 64.6 ± 41.0%).

The general condition of bitches suffering from metritis was not altered; it remained good throughout the trial. Bitches suffering from open or closed pyometra presented were depressed and anorexic in 41/53 (77.3%) and 34/53 (64.1%) cases, respectively. In all animals treated, there was a marked improvement in this criterion by the second day of treatment. On day 8, moderate lethargy was observed in 9 bitches (17.3%) and 14 bitches (26.9%) showed loss of appetite. On day 15, all bitches still receiving treatment were in good general condition, with only one showing any loss of appetite. No side effects were observed after treatment with cloprostenol in 15/33 bitches (45.5%). In the remainder, nausea was the most commonly observed side effect (12/33), and vomiting occurred in six bitches.

None of the bitches had a noticeable change in red blood cell count. The mean white blood cell counts in bitches suffering from metritis were normal and, remained lower ($P < 0.05$) than the counts in bitches with open or closed pyometra (Fig. 2) throughout the trial. In bitches recovering from pyometra, a marked decrease in leucocytosis was observed over the course of treatment. This mean decrease was significant on day

<table>
<thead>
<tr>
<th>Table 1</th>
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<tbody>
<tr>
<td>Number of bitches cured, at various stages in the trial, based on the form of uterine disease</td>
</tr>
<tr>
<td>Treatment</td>
</tr>
<tr>
<td>Metritis (%)</td>
</tr>
<tr>
<td>Open pyometra (%)</td>
</tr>
<tr>
<td>Closed pyometra (%)</td>
</tr>
</tbody>
</table>

| Difference in cure rate between the two treatments on day 90 ($P < 0.05$). |
Fig. 1. Mean (±S.E.M.) rates of decreases in the diameter of the uterine lumen at various stages in the trial, in bitches with open or closed pyometra (Group 1 + Group 2) cured by the administration of aglepristone alone (n = 12), in comparison with those receiving a combination of aglepristone + cloprostenol (n = 27). (*Differences (P < 0.05).

Fig. 2. Mean (±S.E.M.) concentration of leucocytes at various stages in the trial, in bitches with metritis (n = 15), open pyometra (n = 26) or closed pyometra (n = 13) cured by treatment with aglepristone or aglepristone + cloprostenol. For each day, the mean white blood cell count was significantly lower in cases of metritis than in cases of open or closed pyometra. In bitches with closed pyometra, the white blood cell count was lower at day 14 (P < 0.01) and at day 28 (P < 0.001) than at day 0.
15 for bitches with closed pyometra (Fig. 2). Among the 67 treated bitches, on day 1, 15 had plasma progesterone concentrations below 3.18 nmol/L. Treatment was successful in all of them, except for two bitches with open pyometra. For bitches with open or closed pyometra treated with cloprostenol \((n = 32)\), there was a significant decrease in mean peripheral plasma progesterone concentration at day 8 (12.8 ± 14.4 nmol/L versus 5.6 ± 6.6 nmol/L on days 1 versus 8, respectively). However, in bitches treated without cloprostenol \((n = 20)\), no significant decrease in mean plasma progesterone concentration was observed until day 14 (19.3 ± 11.6 nmol/L versus 4.7 ± 1.1 nmol/L on days 1 versus 14; Table 3).

Among the 13 failures using this treatment, one bitch did not present for follow-up and recovery could not be confirmed. One bitch with open pyometra treated with aglepristone died on day 5 in the owner’s house, and another treated with aglepristone and cloprostenol was euthanized in a private clinic on day 15 due to declining health. Ten bitches underwent an ovariohysterectomy. In this last group, six had ovarian cysts. After recovery, 23 bitches underwent follow-up visits over the 24 months following the beginning of treatment. Three of them developed pyometra between 7 and 12 months later (3/23, 13.0%) and another developed it at 19 months (4/21, 19.0%). Two of them received an additional, successful course of treatment (aglepristone + cloprostenol) and no pyometra was observed at the next cycle. We have kept in contact with the owners of three of the bitches used in the trial for some considerable time. They were treated more than 6 years ago and have come into estrus normally, with no recurrence of uterine disease. Five bitches were mated at the first estrus after recovery; four became pregnant.

### 4. Discussion

The use of aglepristone alone cured 27/35 (77.1%) bitches in 90 days. These results are equivalent to those reported by Hoffmann et al. [11] (25/31, 80.6% at day 30) and by Trasch et al. [12] (44/52, 84.6% at day 90). The number of animals treated is insufficient to show any difference in efficacy depending on clinical type (metritis, open pyometra and closed pyometra). This study, however, demonstrated the satisfactory results obtained from the use of aglepristone in bitches suffering from metritis (recovery rate, 15/15). The use of aglepristone is also of particular interest in the treatment of closed pyometra, since it induces cervical opening, and the subsequent evacuation of purulent discharge with a marked improvement in the animal’s general condition. These results demonstrated that surgery is no longer the only treatment for closed pyometra. Moreover, even if surgery has been planned, it is possible to recommend medication as a first-line treatment (aglepristone and rehydration) to improve the general health of the bitch and minimize surgical risk. In this study, the cervix of all the bitches with closed pyometra opened within 48 h. In a study of nine bitches with closed pyometra, Wehrend et al. [14] observed cervical opening within 72 h and the same marked improvement in the animal’s general condition.

In bitches with open or closed pyometra, the repeated administration of low dose cloprostenol significantly increased the success rate at day 90 (27/32, 84.4%). The uterotonic action of cloprostenol lead to a significantly faster decrease in the diameter of the uterine lumen and its luteolytic action caused a faster drop in mean plasma progesterone concentration. This decline in progesterone was commonly observed in bitches treated for pyometra with cloprostenol alone [15], or with cloprostenol associated with aglepristone [13]. In the present study, a decrease in progesterone concentrations was also observed in bitches treated with aglepristone only, but at a later stage. Because of the anti-progesterone properties of aglepristone, this luteolysis does not seem to modify short-term effectiveness. Between 30 and 90 days, we did not note any correlation between blood progesterone concentration and success rate. The low dose of cloprostenol given (1 µg/kg) does not usually cause any adverse reactions [16–18]. Contrary to the results of a similar study [13], 56% of bitches treated had side effects, usually vomiting.

Table 3

Means (±S.D.) plasma progesterone concentrations (nmol/L) in bitches with open or closed pyometra (Group 1 + Group 2) treated with aglepristone alone, compared to those treated with a combination of aglepristone + cloprostenol.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Day 1</th>
<th>Day 8 (*)</th>
<th>Day 14 (*)</th>
<th>Day 28 (*)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aglepristone ((n = 20))</td>
<td>12.8 ± 14.4</td>
<td>5.6 ± 6.6b</td>
<td>4.8 ± 5.1b</td>
<td>3.3 ± 4.2b</td>
</tr>
<tr>
<td>Aglepristone + cloprostenol ((n = 32))</td>
<td>19.3 ± 11.6</td>
<td>15.08 ± 10.5y</td>
<td>4.7 ± 1.1b</td>
<td>3.96 ± 2.8b</td>
</tr>
</tbody>
</table>

Within a row, values with different superscript letters (a and b) differ \((P < 0.05)\); within a column, values with different superscript letters (x and y) differ \((P < 0.05)\).
This particular sensitivity to low doses of cloprostenol can be explained by the poor general condition of the bitches. Nevertheless, the owners did not make any comments.

Among the treated bitches, 17 had basal progesterone concentrations. Cases of pyometra were observed in all stages of the reproductive cycle in a previous study [1]. The effects of aglepristone in pyometra have been studied in diestrous bitches selected with a progesterone concentration of more than 3.2 μm/L [1,10,11]. In the present study 15/17 bitches with a low progesterone concentration were cured after the administration of aglepristone. Similar results were observed in another report [13]. This seems to confirm that, in the pathogenesis of pyometra in bitches, the action of progesterone on the uterus is dependent on the number of progesterone receptors or their specific sensitivity, and not on the plasma progesterone concentration of this hormone.

The recurrence rate of pyometra after treatment differed from results reported in other studies. We did not observe any recurrence between days 30 and 90, and the cure rate was higher, whereas Trasch et al. [12] reported a recurrence rate of 9.8% (4/41) for the same interval; this may be due to the length of treatment and the repeated administration of aglepristone until day 28 if needed (whereas Trasch only administered aglepristone on days 1, 2 and 7). The recurrence rate from 12 to 14 months after treatment was 18.9% (4/37) for Trasch et al. [12], 9% (1/11) for Hoffmann et al. [11] and 21.4% (3/14) for Gobello et al. [13]. The mean age of the bitches (8 years) may explain the recurrence of pyometra in the 24 months following the end of treatment, since age is a major epidemiological factor in the onset of pyometra in bitches. However, treatment failure or recurrence of the disease is closely linked to the presence of ovarian cysts, which maintain a permanent endocrine imbalance. Ovarian cysts were observed in 6 of 10 bitches who underwent ovariohysterectomy due to the failure of medical treatment.

Conservative treatment with prostaglandins, which is recommended for bitches with metritis or pyometra with mild dilatation of the uterine lumen, produces a cure rate of 83–100%, with recurrence rates varying from 10 to 40%. The treatment, which requires several administrations, produces discomfort and is often associated with side effects such as hypersalivation, vomiting and diarrhea [3,4].

In conclusion, treatment with aglepristone alone was a safe and effective treatment for metritis, and an effective means of inducing cervical opening in cases of closed pyometra. The combination of aglepristone and cloprostenol was more effective in the medical treatment of open and closed pyometra than aglepristone alone. Nevertheless, in all cases, we only recommend such treatment in bitches with no liver or kidney dysfunction, and advise close clinical monitoring throughout the entire course of treatment.

References


