International Journal of Agriculture and Biosciences 2026 15(1): 290-299.
https://doi.org/10.47278/journal.ijab/2025.208

RESEARCH ARTICLE
eISSN: 2306-3599; pISSN: 2305-6622

# Integrated Approach to Post-Surgical Care in Canine Pyometra: Evaluating Histological Changes, Blood Parameters, IL-6, and SDMA Biomarkers

Ayra Detsrisuwan [1], Dutruthai Bunpeng [2], Kannika Na Lampang [3], Phongsakorn Chuammitri [3], Purin Lophaisankit [3] and Worapat Prachasilchai [3,4,*]

[1]Faculty of Veterinary Medicine, Kasetsart University, Bangkok 10900, Thailand
[2]Hang Dong Animal Hospital, Soi 8, Nong Kwai, Hang Dong District, Chiang Mai 50230, Thailand
[3]Faculty of Veterinary Medicine, Chiang Mai University, Chiang Mai 50100, Thailand
[4]Research Center in Bioresources for Agriculture, Industry and Medicine, Chiang Mai University, Chiang Mai 50200, Thailand
*Corresponding author: worapat.p@cmu.ac.th, liverpoolpat@yahoo.com

## ABSTRACT

Pyometra is a common issue and can progress into many conditions due to late diagnosis and improper post-operative care. Therefore, the research team conducted this study to discuss the post-operative management of pyometra in dogs using blood profiles, Interleukin-6 (IL-6), and Symmetric dimethylarginine (SDMA), collected from a total of 15 dogs. In a healthy control group (n=10), histological changes can be seen in 5 different levels, including normal, cystic endometrial hyperplasia (CEH), CEH with mild endometritis, mild endometritis, and chronic endometritis. In a group of dogs with pyometra (n=5), two levels of uterine changes were observed: cystic CEH with mild endometritis, and endometrial hemorrhage. The results of the microscopic examination relate to changes in other systemic parameters, including the blood profile, IL-6, and SDMA. The result of the blood profile showed an increase in average white blood cells and total protein at both pre-and post-operation. However, there were no significant differences in IL-6 between pre-and post-operation in both groups of dogs (P=0.5, P=0.19). The pre- and post-operative SDMA levels were not significantly different in pyometra dogs (P=0.58), but significantly different in healthy dogs (P=0.007). Additionally, the results of bacterial culture and drug administration, antibiotics used at the hospital are not effective against some bacteria found in the uterine body - *Enterobacter cloacae* ssp. *cloacae* and *Klebsiella pneumoniae* ssp. *pneumoniae*. Owing to the results of the mentioned parameters, bacterial culture, drug sensitivity, and histological examination should be performed to help design the treatment strategy and antibiotics used, to manage post-surgical care for both healthy and pyometra dogs properly.

**Keywords:** CEH, Endometritis, IL-6, Pyometra, SDMA.

**Article History**
Article # 25-479
Received: 19-Aug-25
Revised: 15-Oct-25
Accepted: 04-Nov-25
Online First: 07-Dec-25

## INTRODUCTION

At present, pyometra, a uterine infection with pus, is a prevalent issue in dogs in Thailand, occurring in both stray and owned dogs (Rungphattanachaikul et al., 2021). The condition can result from various factors, such as contraceptive injections or factors inherent to the dog itself which are commonly seen in cystic endometrial pyometra or cystic endometrial hyperplasia (CEH) (Smith, 2006; Hagman, 2012; Hagman, 2017). Pyometra is often diagnosed late due to its unclear clinical symptoms and the absence of specific diagnostic methods, leading to delayed treatment for these dogs. This delay poses a risk of various complications, including uterine enlargement, uterine rupture, peritonitis, bloodstream infections, and sudden kidney failure (Xavier et al., 2023). These complications further reduce the survival rate of dogs even after undergoing ovariohysterectomy (OVH), a surgical procedure to remove the uterus and ovaries. Therefore, inadequate post-operative management may exacerbate the severity of these complications or prolong the recovery period for dogs (Liao et al., 2020; Vukmer et al., 2021).

**Cite this Article as:** Detsrisuwan A, Bunpeng D, Lampang KN, Chuammitri P, Lophaisankit P and Prachasilchai W, 2026. Integrated approach to post-surgical care in canine pyometra: evaluating histological changes, blood parameters, IL-6 and SDMA biomarkers. International Journal of Agriculture and Biosciences 15(1): 290-299. https://doi.org/10.47278/journal.ijab/2025.208

A Publication of Unique Scientific Publishers

The research team undertook this study to elucidate post-operative management strategies for pyometra in dogs, specifically focusing on hematological parameters, including interleukin-6 (IL-6) and systemic dimethylarginine (SDMA). Additionally, the study aims to investigate the effects of post-operative treatment following OVH in dogs with pyometra. The research comprises two studies: the first involves comparing blood profiles, circulating IL-6, and SDMA levels in dogs before and after OVH treatment for pyometra. The second study compares these parameters in dogs that underwent OVH treatment, distinguishing between those with pyometra and normal dogs. Parameters examined include vital signs, hematological values, bacterial cultures, antimicrobial susceptibility, and biomarkers (IL-6 and SDMA), linked explicitly to inflammatory, infectious, and renal damage processes. These were assessed through dot blot and IDEXX SDMA testing methods. Additionally, histopathological examination of the uterine tissues was conducted to indicate the severity of pyometra. Clinical data were collected through a cohort study involving dogs receiving treatment at the Small Animal Hospital, Chiang Mai University. The study included 15 dogs with confirmed pyometra and a case-control group of 15 dogs, analyzed using paired t-tests for statistical evaluation.

## MATERIALS & METHODS

Blood samples were collected from all 15 dogs and categorized into two groups: the pyometra group and the healthy control group, which consisted of dogs undergoing elective spaying at the Small Animal Hospital, Chiang Mai University. Samples were obtained via the cephalic vein. The first collection tube contained 3mL of blood in ethylenediaminetetraacetic acid (EDTA) for complete blood count (CBC), blood chemistry, and SDMA analysis (Sysmex DX-3010, Sysmex Corporation, Kobe, Japan). The second tube contained 1mL of blood for serum IL-6 analysis. In total, 4mL of blood was collected from each dog. For IL-6 analysis, the blood was centrifuged to separate plasma, which was then transferred to Eppendorf tubes and stored at −20ºC until analysis. IL-6 levels were determined using the dot blot analysis method. After surgery, uterine body samples measuring 1 × 1 × 1 cm (width × height × length) were collected using a biopsy technique. The samples were fixed in 10% neutral-buffered formalin for histopathological examination. The remaining uterine tissue was submitted to the Animal Disease Diagnosis and Translational Technology Center, Chiang Mai University, for bacterial culture and antimicrobial susceptibility testing using the VITEK® 2 system (bioMérieux, Marcy-l'Étoile, France). Clinical data for both groups of dogs were collected, including sex, age, breed, feeding and care practices, and defecation habits. Information on abnormalities was recorded, encompassing the type of abnormality, date of onset, duration, and progression. Vital signs were assessed, including mental status, pulse rate, heart rate, heart sounds, respiratory rate, respiratory sounds, hydration status, mucous membrane condition, body temperature, and pain score (0–4 scale, Colorado State University). Measurements were obtained at two time points: before surgery and 7 days postoperatively. Postoperative complications were also evaluated on day 7$^{th}$, including wound infection, hypothermia, fever, and abdominal pain on palpation. Post-operative management data will be collected at the Small Animal Hospital, Chiang Mai University, including the type, size, quantity, and frequency of antibiotics and anti-inflammatory drugs, as well as the type, duration, and rate of fluid administration. Data on surgical wound management and the frequency of wound cleaning will also be recorded.

For the dot blot protocol, 15 plasma samples will be thawed at room temperature, after which 20–30µg of each sample will be diluted with phosphate buffer saline (PBS) at a 1:1 ratio. The mixture will be vortexed and then incubated at 95°C for 5min. For the preparation of the Immun-Blot® polymer polyvinylidene difluoride (PVDF) membrane and Whatman filter paper, the PVDF membrane will be soaked in methanol for 2min, rinsed in TBST solution for 3min, and the Whatman filter paper will be soaked in tris-buffered saline with tween-20 (TBST) solution. The soaked PVDF membrane will then be placed directly on the soaked Whatman filter paper. PVDF membranes were soaked in methanol for 2min, rinsed in TBST solution for 3min, and then placed onto Whatman filter paper pre-soaked in TBST. Protein samples were applied 3 µL per spot to the membrane and allowed to air-dry at room temperature for 15min, followed by washing in TBST for 5min. The primary antibody used was purified anti-mouse IL-6 monoclonal antibody. Prior to use, the antibody was equilibrated to room temperature for 10min. The membrane was blocked in 5% bovine serum albumin (BSA) in TBST (blocking buffer) for 30min at room temperature, followed by washing with TBST for 3min. The primary antibody was diluted in blocking buffer to the desired concentration and incubated with the membrane either overnight at 4°C or for 2 hours at room temperature. The membrane was then washed with TBST for 3min. The secondary antibody was equilibrated to room temperature for 10min prior to use and prepared according to the manufacturer's instructions. It was diluted in blocking buffer at a ratio of 1:2,000–1:10,000, incubated at room temperature for 45min, and then washed three times with TBST, 3min per wash. The 3,3′-diaminodbenzidine (DAB) substrate solution was prepared by mixing the membrane with the DAB substrate solution and incubating for 5min at room temperature in the dark. Upon completion of staining, images were captured, and the IL-6 levels were quantified using the Image Studio Lite program (LI-COR Biosciences, Lincoln, Nebraska, USA). The analysis involved measuring the grayscale intensity of the dot blot spots corresponding to each plasma sample, with IL-6 intensity normalized to the total protein intensity.

All samples were stained with ponceau red. For analysis, the relative intensity value was calculated as the ratio of IL-6 intensity to total protein intensity (relative intensity = IL-6 intensity / total protein intensity). The relative intensities of all 15 samples were plotted and statistically analyzed using a paired t-test to compare IL-6

levels in dogs undergoing OVH before and after surgery, as well as to compare IL-6 values between dogs with purulent endometritis and healthy controls. For the measurement of SDMA, 1 mL of blood was collected into an EDTA tube and transported to the Medical Examination Unit, Faculty of Veterinary Medicine, Chiang Mai University. SDMA levels were measured using the IDEXX SDMA assay (IDEXX Laboratories, Wesbrook, Maine, USA). For histopathological examination of uterine tissue, paraffin-embedded tissue blocks were sectioned using a microtome, with the cutting thickness set to 4 µm. Sections were stained with hematoxylin and eosin (H&E) following standard protocols. Briefly, paraffin was removed, and the sections were stained with hematoxylin for 15min, rinsed in water for 5min, counterstained with eosin for 1min, and then coverslipped. The prepared slides were examined under a light microscope.

For bacterial culture, specimens were inoculated onto chocolate agar (CA), blood agar (BA), modified Thayer–Martin (MTM) agar, and MacConkey (MAC) agar, in that order. Culturing was initiated immediately or within 20min of specimen collection. Following incubation, slides were labeled and subjected to Gram staining to assess bacterial morphology and quantity, the number of white blood cells, and the presence of clue cells, thereby aiding in the preliminary differentiation of pathogens from normal flora. Petri dishes were incubated at 35–37 °C in an atmosphere containing 5% carbon dioxide ($CO_2$), or in a candle jar with a water-moistened cotton pad to maintain humidity. After 18–24 hours of incubation, culture plates on CA, BA, MTM, and MAC media were examined. If no bacterial growth was observed, incubation continued for up to 72 hours. In cases where Gram staining revealed coccobacilli arranged in a "school-of-fish" pattern, incubation was extended to 5 days.

For specimens in LIM broth, subculturing was performed onto BA, followed by incubation at 35–37°C in an atmosphere containing 5% $CO_2$ or in a candle jar with a water-moistened cotton pad to maintain humidity. If no bacterial growth was observed, incubation was continued for an additional 48 hours, for a total of 72 hours. Pathogens were examined, classified, and subjected to antimicrobial susceptibility testing (Research Institute Public Health Science, 2018).

Data were analyzed using the Wilcoxon signed-rank test and the Mann–Whitney U test to assess IL-6 and SDMA levels, given the small sample size and non-normal data distribution. These nonparametric tests were employed to compare IL-6 and SDMA concentrations before and after surgery. Additional parameters, including hematologic values and antibiotic usage, were analyzed separately. Descriptive statistics were used to summarize and interpret the study's findings.

## RESULTS

Histopathological examination of 15 dogs, as summarized in Table 1, revealed that among 10 clinically healthy dogs undergoing sterilization surgery, pathological changes in the uterine body were detected in five cases. These included cystic endometrial hyperplasia (CEH) in two dogs, chronic metritis in two dogs, mild endometritis in four dogs, CEH with mild endometritis in one dog, and no pathological abnormalities (normal uterus) in one dog. In the pyometra group (n = 5), two types of pathological changes in the uterine body were identified: endometrial hemorrhage in one case, and CEH accompanied by mild endometritis in four cases.

**Table 1:** The histopathological of uterus in dogs (n = 15)

| Pathological change of uterus | Healthy dogs (n = 10) | Pyometra dogs (n = 5) |
|---|---|---|
| Normal | 1 | - |
| Mild endometritis | 4 | - |
| Chronic endometritis | 2 | - |
| Endometrial hemorrhage | - | 1 |
| CEH | 2 | - |
| CEH & mild endometritis | 1 | 4 |

In the mild endometritis group, histopathological examination revealed degeneration of the endometrial glands and hemorrhage. Edema and mild infiltration of lymphocytes and neutrophils were present within the endometrial layer. Additionally, the subserosal layer exhibited edema and red blood cell accumulation (Fig. 1). In the chronic endometritis group, an increased number of endometrial glands, hemorrhage, and marked infiltration of lymphocytes and macrophages were observed. The subserosal layer demonstrated vascular dilation and perivascular edema (Fig. 2). In cases of endometrial hemorrhage, prominent red blood cell accumulation and interstitial edema were present within the endometrium. Glandular hyperplasia and mild infiltration of neutrophils and lymphocytes were also noted, along with mild vascular hypertrophy (Fig. 3). In CEH group, there was marked thickening of the endometrial layer and extensive cyst formation. Proliferation of endometrial glands along the uterine wall was evident, accompanied by interstitial fibrosis and mild lymphocytic infiltration (Fig. 4).

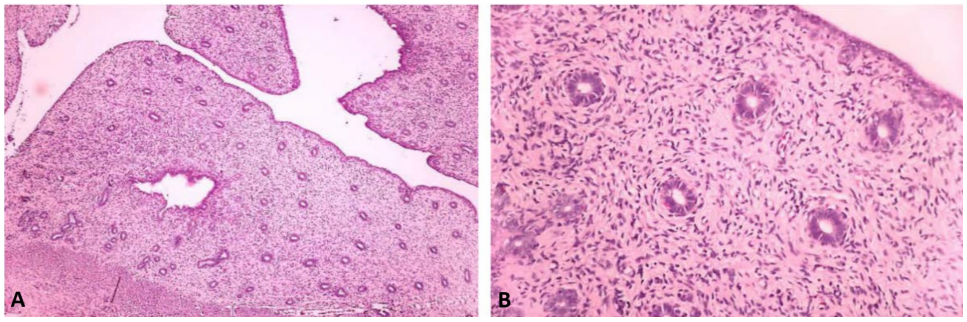


**Fig. 1:** Mild endometritis, dog, uterine body, 10x (A), 40x (B).

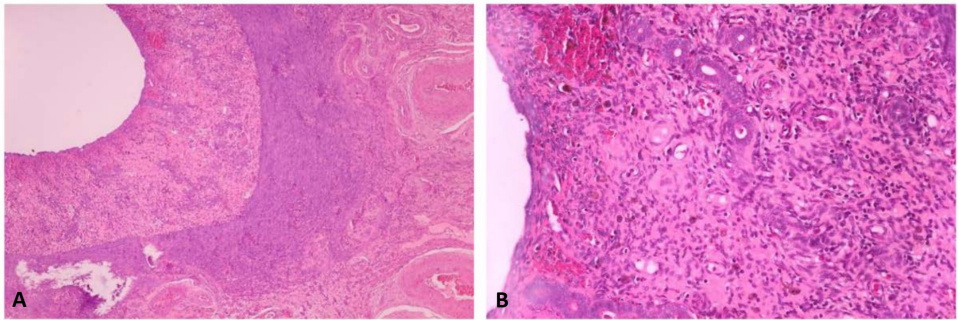


**Fig. 2:** Chronic endometritis, dog, uterine horn, 10x (A), 40x (B).

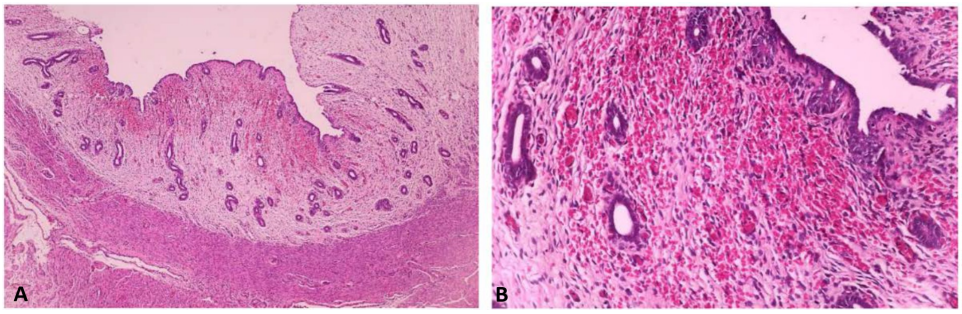


**Fig. 3:** Endometrium hemorrhage, dog, uterine body, 10x (A), 40x (B).

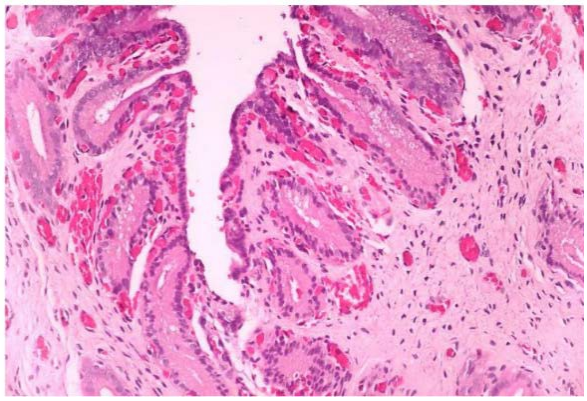

**Fig. 4:** Cystic endometrial hyperplasia, dog, uterine body: 40x.

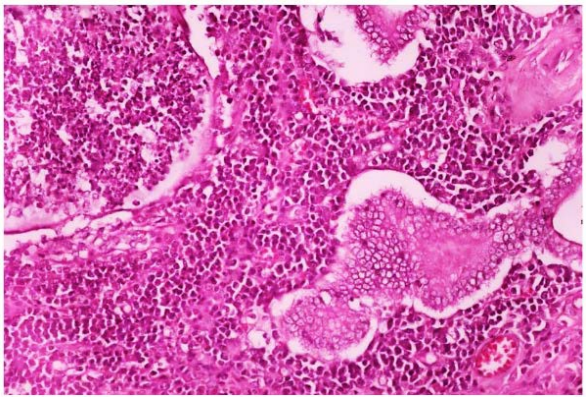

**Fig. 5:** CEH & mild endometritis, dog, uterine body: 40x.

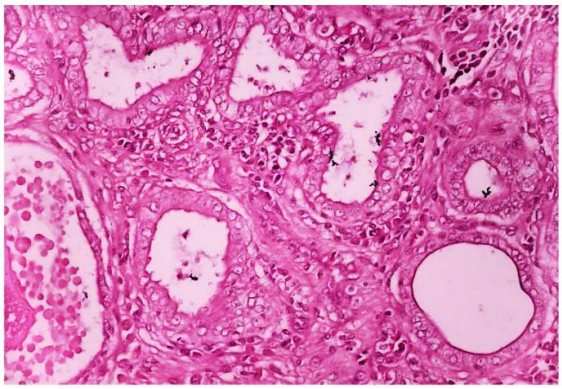

**Fig. 6:** CEH & mild endometritis, dog, uterine body: 40x.

In the group with both CEH and endometritis, numerous cystic formations and marked proliferation of endometrial glands were observed. Neutrophils infiltrated the lumina of the day endometrial glands. Additional findings included interstitial fibrosis, edema, hemorrhage, and the presence of plasma cells and lymphocytes. Vascular hypertrophy was also evident (Fig. 5 and 6).

Bacterial culture of uterine samples from the 15 dogs revealed no bacterial growth in any of the 10 clinically normal dogs. In contrast, bacteria were isolated from the uteri of all five dogs with endometritis. The identified pathogens included *Citrobacter koseri*, *Escherichia coli*, *Enterobacter cloacae* subsp. *cloacae*, *Klebsiella pneumoniae* subsp. *pneumoniae*, and *Streptococcus canis* (Table 2).

**Table 2:** The Bacterial culture of the uterus and drug susceptibility used in the Small Animal Hospital, Chiang Mai University

| | Amoxycillin | Amoxycillin-Clavulanic acid | Cephazolin |
|---|---|---|---|
| *Citrobacter koseri* | - | S | S |
| *Escherichia coli* | I | S | S |
| *Enterobacter cloacae* ssp. cloacae | R | R | R |
| *Klebsiella pneumoniae* ssp. pneumoniae | R | S | S |
| *Streptococcus canis* | - | - | S |

Note: R = resistant, I = intermediate susceptibility, S = sensitive.

The antibiotics listed in Table 2 are those routinely used in Small Animal Hospitals at Chiang Mai University. Most bacterial isolates were susceptible to the antibiotics available in the hospital. However, some isolates exhibited

resistance, including *Enterobacter cloacae* subsp. *cloacae*, which was resistant to amoxicillin, amoxicillin/clavulanic acid, and cefazolin, and *Klebsiella pneumoniae* subsp. *pneumoniae*, which was resistant to amoxicillin. *Escherichia coli* demonstrated intermediate susceptibility to amoxicillin. Based on the results of the Wilcoxon signed-rank test comparing the relative intensity of IL-6 before surgery and 7 days after surgery in all dogs undergoing OVH (n = 15), dogs were categorized into two groups: those with purulent uterine inflammation (pyometra) and clinically healthy dogs undergoing elective sterilization. Among the dogs with uterine inflammation, the mean relative intensity of IL-6 before sterilization was 0.34 (mean = 0.34; min = 0.20; max = 0.67; SD = 0.18).

Seven days after surgery, the mean relative intensity of IL-6 in dogs with purulent uterine inflammation was 0.31 (mean = 0.31; min = 0.17; max = 0.61; SD = 0.18), with no significant difference compared with pre-surgical values (P = 0.50). In clinically healthy dogs undergoing elective sterilization, the mean relative intensity of IL-6 before surgery was 0.08 (mean = 0.08; min = 0.01; max = 0.25; SD = 0.09), and 0.07 after 7 days (mean = 0.07; min = 0.01; max = 0.23; SD = 0.08). No significant difference was observed between pre- and post-surgical values in this group (P = 0.19) (Fig. 7). When comparing pre-surgical IL-6 relative intensity between dogs with purulent endometritis and healthy controls using the Mann–Whitney U test, values in the purulent endometritis group were significantly higher than those in the healthy group (P = 0.005) (Fig. 8). Similarly, post-surgical IL-6 relative intensity remained significantly higher in dogs with purulent endometritis compared with healthy dogs (P = 0.005) (Fig. 9).

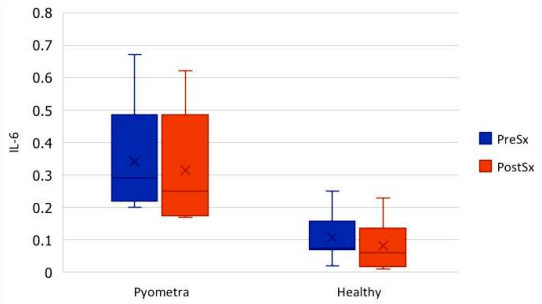


**Fig. 7:** The relative intensity of IL-6 in healthy and pyometra dog before and 7 days after surgery.

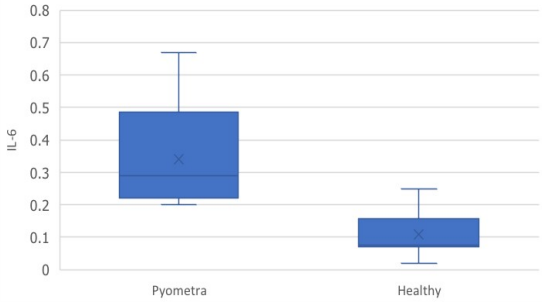


**Fig. 8:** The relative intensity of IL-6 in healthy and pyometra dog before surgery.

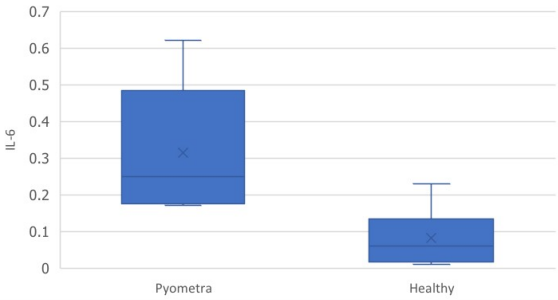


**Fig. 9:** The relative intensity of IL-6 in healthy and pyometra dog before and after surgery.

Complete blood count results are presented in Table 2. The 15 dogs were grouped according to histopathological diagnosis. Healthy dogs were classified into five categories: normal uterus (Normal), CEH with mild endometritis, CEH, mild endometritis, and chronic endometritis. Dogs in the normal (n = 1), CEH (n = 2), and CEH with mild endometritis (n = 1) groups exhibited only slight alterations in hematologic values.

In the mild endometritis group (n = 4), pre-surgical CBC results showed elevated white blood cell (WBC) counts, lymphocytes, and segmented neutrophils compared with reference values. Mean values were 19.02±6.41 $\times10^3$/μL, 14.15±6.37 $\times10^3$/μL, and 3.07±1.02 $\times10^3$/μL, respectively. Following surgery, the mean WBC count and segmented neutrophil count decreased to within the normal range. However, the mean lymphocyte count increased compared with pre-surgical values, with a mean of 4.87±0.69 $\times10^3$/μL.

In the chronic endometritis group (n = 2), pre-surgical results indicated elevated WBC counts, segmented neutrophils, and lymphocytes, with mean values of 20.94±6.28 $\times10^3$/μL, 14.69±7.94 $\times10^3$/μL, and 3.30±2.51 $\times10^3$/μL, respectively. Post-surgical results showed that mean WBC and lymphocyte counts remained above the normal range (14.23±7.18 $\times10^3$/μL and 3.64±0.85 $\times10^3$/μL, respectively), whereas the mean segmented neutrophil count decreased to within the reference range.

Healthy dogs in the normal (n = 1) and CEH (n = 2) groups showed no changes in blood chemistry values before and after surgery (Table 3). In the CEH with mild endometritis group (n = 1), total protein was elevated after surgery (8.3g/dL), while albumin concentrations were above the reference range both before and after surgery (3.9g/dL and 3.4g/dL, respectively) (Table 4).

In the mild endometritis (n = 4) and chronic endometritis (n = 2) groups, mean total protein concentrations were above the reference range both before and after surgery, with lower mean values observed post-surgery. Additionally, in the chronic endometritis group, mean albumin concentration post-surgery was elevated (3.25±0.21g/dL).

In the pyometra group, dogs with CEH and endometritis had mean total protein concentrations higher than the reference range both before and after surgery (9.43±1.46g/dL and 9.33±1.16g/dL, respectively). Dogs with endometrial hemorrhage also exhibited albumin concentrations above the reference range before and after surgery (3.5g/dL and 3.2g/dL, respectively).

**Table 3:** The mean and standard deviation of complete blood count from 15 dogs before and after surgery

| Group | | Hct | Hgb | RBCs | MCV | MCH | MCHC | WBCs | Neu b | Neu s | Lym | Mon | Eos | Bas | Plt |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | % | g/dL | $10^6$/µL | fl | pg | g/dL | $10^3$/µL | $10^3$/µL | $10^3$/µL | $10^3$/µL | $10^3$/µL | $10^3$/µL | $10^3$/µL | $10^3$/µL |
| Normal (n = 1) | Pre | 42 | 14.5 | 6.09 | 69.7 | 23.8 | 34.2 | 10.71 | 0 | 6.9 | 2.44 | 0.88 | 0.49 | - | 559 |
| | Post | 40 | 13.3 | 5.88 | 67.9 | 22.6 | 33.3 | 9.88 | 0 | 8.38 | 0.74 | 0.57 | 0.18 | 0.01 | 275 |
| CEH (n = 3) | Pre | 52.67±6.66 | 17.73±2.37 | 7.56±1 | 69.93±1.6 | 23.47±1.12 | 33.53±0.85 | 13.89±1.64 | 0±0 | 8.65±1.64 | 3.46±1.78 | 0.95±0.51 | 0.8±0.18 | 0.03±0.01 | 374.67±191.69 |
| | Post | 44.33±4.73 | 15.2±1.78 | 6.27±0.6 | 70.9±1.68 | 24.23±0.84 | 34.13±0.45 | 10.22±0.5 | 0±0 | 6.25±0.71 | 2.41±0.66 | 1.06±0.3 | 0.93±0.45 | 0.1 | 339.67±67.9 |
| Mild endometritis (n = 4) | Pre | 37±8.29 | 12.13±2.63 | 5.44±0.95 | 67.98±4.91 | 22.2±1.19 | 32.68±1.42 | 19.02±6.41 | 0.12±0.24 | 14.15±6.37 | 3.07±1.02 | 0.95±0.44 | 0.71±0.21 | 0.03±0.04 | 237.5±47.12 |
| | Post | 43±0 | 13.5±0.17 | 6.31±0.17 | 68.07±1.42 | 21.27±0.32 | 31.1±0.35 | 12.95±1.67 | 0±0 | 6.73±1.01 | 4.87±0.69 | 0.29±0.29 | 1.01±0.36 | 0.04±0.03 | 299±35.59 |
| Chronic endometritis (n = 2) | Pre | 45±7.07 | 15.25±2.19 | 6.52±0.91 | 68.5±1.56 | 23.4±0.14 | 34.15±0.64 | 20.94±6.28 | 0.51±0.72 | 14.69±7.94 | 3.3±2.51 | 0.88±0.17 | 1.59±0.26 | 0.01±0.01 | 300.5±9.19 |
| | Post | 49±8.49 | 16.2±2.83 | 6.97±1.22 | 69.95±0.35 | 23.2±0 | 33.3±0.14 | 14.23±7.18 | 0±0 | 8.51±4.88 | 3.64±0.85 | 0.74±0.86 | 1.31±0.6 | 0.02±0.02 | 353±5.66 |
| CEH + mild endometritis (n = 4) | Pre | 40±9.06 | 13.55±2.79 | 6.19±1.43 | 64.65±2.54 | 22±1.4 | 34.03±1.62 | 20.83±15.53 | 1.97±2.28 | 15.26±12.99 | 2.23±1.18 | 0.7±0.13 | 0.88±0.39 | 0.01±0 | 212.75±117.03 |
| | Post | 37±12.91 | 12.2±4.59 | 5.89±2.04 | 49.98±28.79 | 20.6±1.97 | 32.95±1.17 | 16.84±11.01 | 1.32±2.64 | 12.16±7.87 | 1.41±0.41 | 1.06±0.89 | 0.86±0.49 | 0.02±0.01 | 257.25±86.86 |
| Endometrial hemorrhage (n = 1) | Pre | 47 | 15.6 | 6.49 | 72.5 | 24 | 33.2 | 19.56 | 0 | 8.5 | 5.63 | 0.86 | 4.44 | 0.13 | 225 |
| | Post | 46 | 15.1 | 6.29 | 72.4 | 24 | 33.2 | 16.03 | 0 | 6.25 | 6.33 | 0.78 | 2.61 | 0.06 | 297 |

Note: Hct = hematocrit, Hgb = hemoglobin, RBCs = red blood cells, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, WBCs = white blood cells, Neu b = band neutrophil, Neu s = segmented neutrophil, Lym = lymphocyte, Mon = monocyte, Eos = eosinophil, Bas = basophil, Plt = platelet.

**Table 4:** The mean and standard deviation of blood chemistry from 15 dogs before and after surgery

| Group | | BUN | Crea | ALT | ALP | TP | Alb |
|---|---|---|---|---|---|---|---|
| | | mg/dL | mg/dL | U/L | U/L | g/dL | g/dL |
| Normal (n = 1) | Pre | 13.4 | 0.81 | 39 | 29 | 6.9 | 3.8 |
| | Post | 10.7 | 1.25 | 22 | 36 | 7.3 | 3.4 |
| CEH (n = 3) | Pre | 16.3 ± 3.4 | 1.16 ± 0.41 | 35.33 ± 18.23 | 39.67 ± 22.81 | 7.13 ± 0.35 | 3.5 ± 0.36 |
| | Post | 20.17 ± 3.44 | 1.13 ± 0.27 | 29.67 ± 10.6 | 53.67 ± 50.36 | 7.27 ± 0.96 | 2.97 ± 0.38 |
| Mild endometritis | Pre | 13.85 ± 3.5 | 1.04 ± 0.28 | 28.75 ± 7.14 | 79 ± 20.54 | 8.58 ± 0.56 | 2.83 ± 0.53 |
| (n = 4) | Post | 17.15 ± 4.37 | 1.07 ± 0.06 | 28 ± 5.48 | 62.5 ± 18.65 | 8.15 ± 0.93 | 2.98 ± 0.17 |
| Chronic endometritis | Pre | 17.9 ± 1.98 | 1.01 ± 0.07 | 25.5 ± 9.19 | 50 ± 7.07 | 8.35 ± 0.64 | 2.95 ± 0.07 |
| (n = 2) | Post | 19.85 ± 8.41 | 1.04 ± 0.02 | 16.5 ± 0.71 | 28.5 ± 17.68 | 8.3 ± 0.42 | 3.25 ± 0.21 |
| CEH + mild endometritis | Pre | 16.98 ± 8.54 | 1.09 ± 0.24 | 38.25 ± 22.65 | 95.75 ± 36.1 | 9.43 ± 1.46 | 2.73 ± 0.17 |
| (n = 4) | Post | 22.33 ± 9.84 | 1.26 ± 0.06 | 49.75 ± 26.7 | 52.25 ± 56.81 | 9.33 ± 1.16 | 2.8 ± 0.42 |
| Endometrial hemorrhage | Pre | 21 | 1.55 | 38 | 9 | 7.4 | 3.5 |
| (n = 1) | Post | 23.4 | 1.37 | 35 | 89 | 6.6 | 3.2 |

Note: BUN = blood urea nitrogen, Crea = creatinine, ALT = alanine transaminase, ALP = alkaline phosphatase, TP = total protein, Alb = albumin

**Table 5:** The recommended antibiotic for *Enterobacter cloacae* ssp. *cloacae* and *Klebsiella pneumonia* ssp. *pneumonia* by Center of Veterinary Medical Diagnostic and Animal Health Innovation, Chiang Mai University

| Microorganism | Recommended antibiotic |
|---|---|
| *Enterobacter cloacae* ssp *.cloacae* | Sulfamethoxazole/Trimethoprim dose 30 mg/kg q8h PO |
| | Cephalexin dose 22 mg/kg q12h PO |
| | Enrofloxacin dose 5 mg/kg q24h PO |
| | Marbofloxacin dose 5.5 mg/kg q24h PO |
| | Pradofloxacin dose 3 mg/kg q24h PO |
| | Cefovecin dose 8 mg/kg q7-14days SC |
| | Amikacin dose 15-20 mg/kg q24h SC |
| *Klebsiella pneumonia* ssp *.pneumonia* | Sulfamethoxazole/Trimethoprim dose 30 mg/kg q8h PO |
| | Cephalexin dose 22 mg/kg q12h PO |
| | Enrofloxacin dose 5 mg/kg q24h PO |
| | Marbofloxacin dose 5.5 mg/kg q24h PO |
| | Pradofloxacin dose 3 mg/kg q24h PO |
| | Cefovecin dose 8 mg/kg q7-14 days SC |
| | Amikacin dose 15-20 mg/kg q24h SC |

The results of the comparison of SDMA values from a total of 15 dogs were analyzed using a paired *t*-test. The dogs were divided into two groups: (1) dogs with purulent uterine inflammation (pyometra) and (2) clinically healthy dogs presented for elective sterilization, with SDMA measured both before and 7 days after surgery.

In the pyometra group, the mean preoperative SDMA value was 18.33µg/dL (mean = 18.33, min = 13, max = 24, SD = 5.5). Seven days postoperatively, the mean SDMA value was 18.66µg/dL (mean = 18.66, min = 12, max = 31, SD = 10.7). No statistically significant difference was observed between pre- and postoperative values ($P$ = 0.58).

In the healthy control group, the mean preoperative SDMA value was 11.75µg/dL (mean = 11.75, min = 9, max = 15, SD = 2.75), whereas the mean postoperative value at 7 days was 15.75µg/dL (mean = 15.75, min = 13, max = 19, SD = 2.75). SDMA levels were significantly higher postoperatively compared to preoperative values ($P$ = 0.007) (Fig. 10).

A comparison of preoperative SDMA levels between dogs with purulent endometritis and healthy controls was performed using the Mann–Whitney *U* test. Dogs with purulent endometritis exhibited significantly higher SDMA values than healthy dogs ($P$ = 0.034) (Fig. 11). In contrast, when comparing the relative intensity of IL-6 levels after surgery between the two groups, no statistically significant difference was detected ($P$ = 0.5) (Fig. 12).

## DISCUSSION

Although no significant changes in IL-6 levels were observed in either dogs with purulent endometritis (P = 0.43) or healthy dogs (P = 0.22) when comparing pre-surgery and 7 days post-surgery values, these results differ

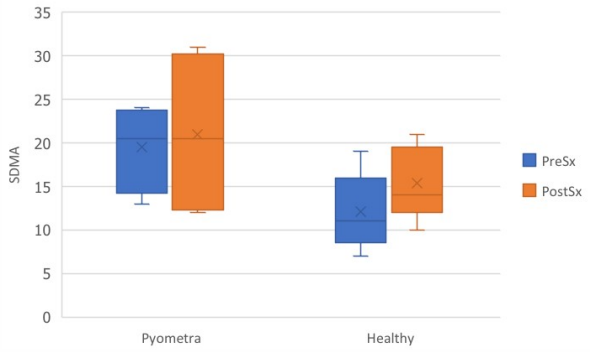


**Fig. 10:** The SDMA levels in healthy and pyometra dog before and 7 days after surgery.

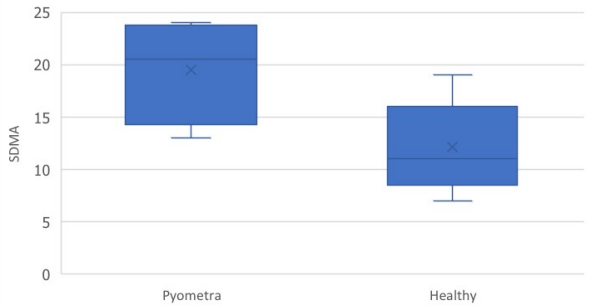


**Fig. 11:** The SDMA levels in healthy and pyometra dog before surgery.

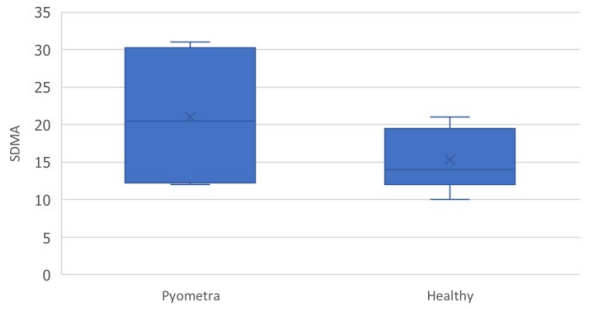


**Fig. 12:** The SDMA levels in healthy and pyometra dog after surgery.

from those reported by Dąbrowski et al. (2015b). In their study, dogs with pyometra exhibited a significant postoperative decrease in IL-6, while healthy dogs showed an increase on day 3 followed by a decrease on day 10 (Dąbrowski et al., 2015b). Recent comprehensive studies have demonstrated that IL-6, along with other inflammatory mediators such as high-mobility group box 1 (HMGB1) and procalcitonin (PCT), significantly decreases following surgical removal of the inflamed uterus, indicating their utility as biomarkers for clinical recovery (Ahn et al., 2021). Furthermore, IL-6 has been identified as one of the most favorable biomolecular markers for predicting endometritis in bitches, with an area under the curve (AUC) of 0.67 in receiver operating characteristic analysis (Schüttler & Neumann, 2015; García Mitacek et al., 2023). When comparing IL-6 levels within individual dogs, a tendency toward decreased IL-6 at day 7 post-surgery was noted. The variability in results between studies may stem from differences in the severity of suppurative endometritis, the clinical condition of the animals, and postoperative care some dogs in this study were not hospitalized post-surgery, potentially leading to discrepancies in owner-administered medications and aftercare (Turkki et al., 2023). Additional factors contributing to variability include the timing of peak inflammatory responses, as IL-6 is recognized as a major proinflammatory cytokine released during the early phase of endotoxemia, and peak concentrations may not have been persistent or reached in cases that did not develop severe sepsis (Karlsson et al., 2012).

Histopathological changes were present even in clinically healthy dogs (n = 10), including conditions such as CEH, mild endometritis, and chronic endometritis all of which were associated with elevated CBC, blood chemistry, and IL-6 levels. For example, even before surgery, healthy dogs with mild and chronic endometritis had elevated WBC counts; however, only the mild endometritis group normalized within 7 days post-surgery. In chronic endometritis cases, WBC and lymphocyte counts remained elevated postoperatively, indicating that subclinical uterine inflammation may persist. This implies that "healthy" dogs without signs of clinical infection may harbor latent endometrial inflammation (Xavier et al., 2023) and may be at risk for future purulent cervicitis; thus, vigilant postoperative monitoring is warranted (Heiene et al., 2007; Woźna-Wysocka et al., 2021). Recent molecular studies support this finding, demonstrating that inflammatory genes including cyclooxygenase 1 (COX1), IL-6, and IL-8 show significantly higher mRNA expression in bitches with endometritis compared to those with normal endometrium, even in the absence of cystic endometrial hyperplasia (García Mitacek et al., 2023). In contrast, dogs with purulent endometritis exhibited histopathological patterns of CEH with mild endometritis or endometrial hemorrhage. These dogs maintained elevated WBC counts both before and after surgery, suggesting that standard postoperative management may be insufficient to fully control inflammation and infection (Turkki et al., 2023). These findings align with those reported by Jitpean et al. (2014), wherein dogs with pyometra demonstrated persistent leukocytosis, neutrophilia with left shift, and monocytosis until at least three days post-surgery (Dąbrowski et al., 2015b).

Recent literature further emphasizes the importance of controlling postoperative inflammation in pyometra cases. Studies utilizing serial biomarker analysis have shown that inflammatory markers including C-reactive protein (CRP) and serum amyloid A (SAA) typically normalize within 7 to 14 days following treatment of infectious diseases, with serial measurements potentially aiding in the identification of disease resolution (Goggs et al., 2022). However, CRP concentrations can increase 16- to 45-fold within 24 to 48 hours following surgical procedures, particularly in cases involving significant surgical trauma, making it a reliable indicator of postsurgical inflammation (Christensen et al., 2015). Importantly, CRP levels measured 4 days postoperatively have been shown to be significantly higher in dogs with incision site infections compared to uninfected animals, with affected dogs demonstrating mean CRP

concentrations of 296.6 mg/L versus approximately 100 mg/L in the uninfected cohort (Christensen et al., 2015). Hematobiochemical parameters, including WBC count, total protein, blood urea nitrogen (BUN), and creatinine, have been reported to gradually improve following ovariohysterectomy; however, in dogs with pyometra, most values remained above the reference range at 15 days post-surgery (Dąbrowski et al., 2015a; Hokamp & Nabity, 2016). Moreover, a recent review on diagnostic biomarkers for pyometra indicated that sustained elevation of systemic inflammatory markers is associated with prolonged hospitalization and poorer prognosis (Jitpean et al., 2014; Singh et al., 2020). Advanced biomarkers of endothelial activation, including vascular endothelial growth factor (VEGF), hyaluronan, and plasminogen activator inhibitor 1 (PAI-1), are significantly elevated in dogs with sepsis and organ dysfunction, suggesting that endothelial activation occurs in naturally occurring sepsis and may contribute to organ dysfunction (Gaudette et al., 2023). These findings highlight the need for optimized postoperative strategies aimed at reducing inflammation and improving recovery outcomes.

The blood chemistry findings revealed that healthy dogs with mild endometritis, those with chronic endometritis, and dogs with purulent endometritis in the CEH with mild endometritis group exhibited mean total protein levels above the normal range both before and after surgery. This aligns with patient histories and histopathological findings. Elevated total protein likely reflects dehydration, chronic inflammation, and/or infection conditions that stimulate globulin synthesis and thus raise overall protein levels. Although mean blood urea nitrogen (BUN) and creatinine remained within normal limits across all groups, two dogs in the purulent endometritis group displayed postoperative elevations in BUN beyond normal limits while creatinine remained unchanged. Notably, these two dogs also exhibited sustained increases in SDMA levels from normal pre-surgery values to 28 and 31μg/dL at day 7 well above the canine reference range (0–14μg/dL). This highlights the potential risk of postoperative acute kidney injury in inflamed uterus cases and underscores the need for close renal monitoring, as SDMA may detect kidney dysfunction earlier than traditional markers (Andrade et al., 2023). SDMA is considered a more sensitive and earlier marker of declining glomerular filtration rate (GFR) compared to serum creatinine, as it increases as early as 25% loss of kidney function, while creatinine cannot identify kidney issues until almost 75% of kidney function is lost (Hall et al., 2016). Furthermore, SDMA is less impacted by extrarenal factors than creatinine, including body condition, advanced age, and disease state, making it particularly valuable for assessing kidney function in animals with inflammatory conditions (Hall et al., 2016).

In statistical comparisons, SDMA levels in dogs with purulent endometritis did not differ significantly pre- versus post-surgery ($P = 0.58$); however, in healthy dogs, postoperative SDMA levels were significantly higher ($P = 0.007$). Intriguingly, all four healthy dogs with elevated postoperative SDMA belonged to the same owner, suggesting potential shared factors such as hydration status, medication practices, or postoperative care differences. The clinical significance of SDMA elevation extends beyond simple renal function assessment, as studies in dogs with chronic kidney disease have demonstrated that SDMA correlates better with renal function than serum creatinine and shows measurable clearance during dialytic treatment, with a 48-hour rebound effect of 25% between sessions (Le Sueur et al., 2019). Microbiological culture results indicated that commonly used antibiotics in small animal hospitals such as amoxicillin, amoxicillin/clavulanic acid, and cefazolin may not effectively target uterine pathogens like *Enterobacter cloacae* and *Klebsiella pneumoniae* (Table 5). The organisms isolated in this study are consistent with those previously reported in cases of canine purulent endometritis (Santana and Santos 2021; Xavier et al., 2023; Ylhäinen et al., 2023). Recent molecular characterization studies have demonstrated that these bacterial pathogens are associated with differential expression of inflammatory cytokines, with IL-6 and IL-8 showing particularly high upregulation in infected endometrial tissue (Sasidharan et al., 2023).

Additional evidence from recent literature supports the clinical significance of these findings. Advanced renal markers, including SDMA and urinary gamma-glutamyl transferase (uGGT), have demonstrated superior sensitivity to creatinine in detecting early renal impairment in dogs with pyometra. In Brazil, the study involving 14 dogs showed that SDMA (ranging 17.7–26.5 μg/dL), uGGT, and urinary protein–creatinine ratio (uPCR) was elevated pre- and shortly post-ovariohysterectomy, while creatinine and urea remained relatively stable emphasizing the value of early biomarkers in monitoring renal injury (Andrade et al., 2023). Moreover, a 2025 prospective study on postoperative acute kidney injury (AKI) in healthy dogs indicated that even elective surgeries can lead to renal impairment in a small percentage (~3%) of cases, with surgery duration being a significant risk factor (Muñoz-Blanco & Salazar, 2025). This underscores the importance of vigilant perioperative monitoring particularly in patients with pre-existing inflammatory conditions like pyometra. Contemporary research has also identified procalcitonin (PCT) as a valuable prognostic biomarker in dogs with systemic inflammatory response syndrome (SIRS), with significantly higher concentrations observed in deceased animals compared to survivors, and positive correlations with pro-inflammatory cytokines IL-6 and tumor necrosis factor-α (Chadorneshin et al., 2023). These findings collectively emphasize the need for comprehensive postoperative monitoring strategies that incorporate multiple biomarkers to optimize patient outcomes and enable early detection of complications.

**Conclusion**

Nevertheless, the relatively small sample size in this study limits the generalizability of the findings. This constraint was primarily due to the restricted timeframe and stringent inclusion criteria applied by the research team. Future studies incorporating larger cohorts are

warranted to elucidate more robust associations between inflammatory mediators, hematological responses, and histopathological alterations. Specifically, investigating the relationships among IL-6, leukocyte counts, and uterine tissue changes may advance our understanding of disease mechanisms and provide prognostic indicators. Furthermore, longitudinal monitoring of IL-6 in conjunction with SDMA and leukocyte profiles could offer valuable tools for assessing disease severity, progression, and postoperative outcomes in dogs with pyometra. The methodological approach for cytokine quantification also warrants refinement. In the present study, circulating IL-6 was measured using a dot blot protocol, which provides relative intensity values derived from grayscale imaging normalized to total protein staining (Ponceau Red). While this method is cost-effective, its semi-quantitative nature may limit accuracy. By contrast, enzyme-linked immunosorbent assay (ELISA) detects IL-6 via antigen–antibody binding and signal amplification, offering higher sensitivity and quantitative reliability. Although ELISA entails higher operational costs, it may provide superior accuracy and reproducibility in future investigations, thereby strengthening the reliability of biomarker-based monitoring in canine purulent endometritis.

## DECLARATIONS

**Funding:** This study was financial supported by the Faculty of Veterinary Medicine, Chiang Mai University.

**Acknowledgement:** We extend our sincere gratitude to the Faculty of Veterinary Medicine, Chiang Mai University, THAILAND and the staff of the Small Animal Hospital for their invaluable assistance.

**Conflict of Interest:** The authors declare no conflicts of interest

**Data Availability:** All data supporting the findings of this study are available from the corresponding author upon reasonable request.

**Ethics Statement:** Ethical approval was obtained from the Ethical Review Committee for Human Research under reference number HS2/2565. The animal study design and experimental procedures were approved by the Animal Care and Use Committee, Faculty of Veterinary Medicine, Chiang Mai University (FVM-ACUC), under reference number S13/2565. Written informed consent was obtained from all participants for the publication of their anonymized information.

**Author's Contribution:** Conceptualization, P.C., & W.P.; methodology, P.C., & W.P.; software, A.D., D.B., K.N.L., P.C., P.L. & W.P.; validation, K.N.L., P.C., P.L. & W.P.; formal analysis, K.N.L., P.C., P.L. & W.P.; investigation, A.D., D.B., K.N.L., P.C., P.L. & W.P.; resources, A.D., D.B., K.N.L., P.C., P.L. & W.P.; data curation, A.D., D.B., K.N.L., P.C., P.L. & W.P.; writing—original draft preparation, A.D., D.B., P.C., P.L. & W.P.; writing—review and editing, A.D., D.B., K.N.L., P.C., P.L. & W.P.; visualization, K.N.L., P.C., P.L. & W.P.; supervision, K.N.L., P.C. & W.P.; project administration, P.L. & W.P. All authors have read and agreed to the published version of the manuscript.

**Generative AI Statement:** The authors declare that no Gen AI/DeepSeek was used in the writing/creation of this manuscript.

**Publisher's Note:** All claims stated in this article are exclusively those of the authors and do not necessarily represent those of their affiliated organizations or those of the publisher, the editors, and the reviewers. Any product that may be evaluated/assessed in this article or claimed by its manufacturer is not guaranteed or endorsed by the publisher/editors.

## REFERENCES

Ahn, S., Bae, H., Kim, J., Kim, S., Park, J., Kim, S.K., Jung, D.I., & Yu, D. (2021). Comparison of clinical and inflammatory parameters in dogs with pyometra before and after ovariohysterectomy. *Canadian Journal of Veterinary Research*, *85*(4), 271–278.

Andrade, M.B., Oliveira, R.S., & Silveira, C.M. (2023). Evaluation of renal biomarkers in dogs with pyometra before and after ovariohysterectomy. *Brazilian Journal of Veterinary Research*, *43*(2), 110–118.

Chadorneshin, J.R., Khaksar, E., Sharif, M.T., & Jahandideh, A. (2023). The prognostic value of procalcitonin in critically ill cases of systemic inflammatory response syndrome in dogs. *Comparative Clinical Pathology*, *32*(1), 91–97. https://doi.org/10.1007/s00580-022-03417-2.

Christensen, M.B., Eriksen, T., & Kjelgaard-Hansen, M. (2015). C-reactive protein: Quantitative marker of surgical trauma and post-surgical complications in dogs—a systematic review. *Acta Veterinaria Scandinavica*, *57*, 71. https://doi.org/10.1186/s13028-015-0164-5.

Dąbrowski, R., Szczubiał, M., Kostro, K., Wawron, W., Ceron, J.J., & Tvarijonaviciute, A. (2015a). Serum insulin-like growth factor-1 and C-reactive protein concentrations before and after ovariohysterectomy in bitches with pyometra. *Theriogenology*, *83*(4), 474–477. https://doi.org/10.1016/j.theriogenology.2014.09.024.

Dąbrowski, R., Pastor, J., Szczubiał, M., Piech, T., Bochniarz, M., Wawron, W., & Tvarijonaviciute, A. (2015b). Serum IL-6 and IL-10 concentrations in bitches with pyometra undergoing ovariohysterectomy. *Acta Veterinaria Scandinavica*, *57*(61), 1–4. https://doi.org/10.1186/s13028-015-0153-8.

García Mitacek, M.C., Praderio, R.G., Stornelli, M.C., De La Sota, R.L., & Stornelli, M.A. (2023). Endometrial mRNA expression of key inflammatory genes and progesterone receptor in uterine tissue of bitches with endometritis. *Veterinarski Arhiv*, *93*(6), 683–694.

Gaudette, S., Hughes, D., & Boller, M. (2023). Biomarkers of endothelial activation and inflammation in dogs with naturally occurring sepsis and organ dysfunction. *Frontiers in Veterinary Science*, *10*, 1127099. https://doi.org/10.3389/fvets.2023.1127099.

Goggs, R., Robbins, S.N., LaLonde-Paul, D.M., & Menard, J.M. (2022). Serial analysis of blood biomarker concentrations in dogs with pneumonia, septic peritonitis, and pyometra. *Journal of Veterinary Internal Medicine*, *36*(2), 708–716. https://doi.org/10.1111/jvim.16374.

Hagman, R. (2012). Clinical and molecular characteristics of pyometra in female dogs. *Reproduction in Domestic Animals*, *47*(Suppl. 6), 323–325. https://doi.org/10.1111/rda.12031.

Hagman, R. (2017). Canine pyometra: What is new? *Reproduction in Domestic Animals*, *52*(Suppl. 2), 288–292. https://doi.org/10.1111/rda.12843.

Hall, J.A., Yerramilli, M., Obare, E., Yerramilli, M., Almes, K., & Jewell, D.E. (2016). Serum concentrations of symmetric dimethylarginine and creatinine in dogs with naturally occurring chronic kidney disease. *Journal of Veterinary Internal Medicine*, *30*(3), 794–802. https://doi.org/10.1111/jvim.13942.

Heiene, R., Kristiansen, V., Teige, J., & Jansen, J.H. (2007). Renal histomorphology in dogs with pyometra and control dogs, and long-term clinical outcome with respect to signs of kidney disease. *Acta Veterinaria Scandinavica*, *49*(1), 13. https://doi.org/10.1186/1751-

0147-49-13.

Hokamp, J.A., & Nabity, M.B. (2016). Renal biomarkers in domestic species. *Veterinary Clinical Pathology*, *45*(1), 28–56. https://doi.org/10.1111/vcp.12333.

Jitpean, S., Ström-Holst, B., Emanuelson, U., Höglund, O., Pettersson, A., Alneryd-Bull, C., & Hagman, R. (2014). Outcome of pyometra in female dogs and predictors of peritonitis and prolonged postoperative hospitalization in surgically treated cases. *BMC Veterinary Research*, *10*, 6. https://doi.org/10.1186/1746-6148-10-6.

Karlsson, I., Hagman, R., Johannisson, A., Wang, L., Karlstam, E., & Wernersson, S. (2012). Cytokines as immunological markers for systemic inflammation in dogs with pyometra. *Reproduction in Domestic Animals*, *47*(Suppl. 6), 337–341. https://doi.org/10.1111/rda.12034.

Le Sueur, A.N.V., Geraldes, S.S., Melchert, A., Takahira, R.K., Coyne, M., Murphy, R., Szlosek, D., & Guimarães-Okamoto, P.T.C. (2019). Symmetric dimethylarginine concentrations in dogs with International Renal Interest Society stage 4 chronic kidney disease undergoing intermittent hemodialysis. *Journal of Veterinary Internal Medicine*, *33*(6), 2635–2643. https://doi.org/10.1111/jvim.15612.

Liao, A.T., Huang, W.H., & Wang, S.L. (2020). Bacterial isolation and antibiotic selection after ovariohysterectomy of canine pyometra: A retrospective study of 55 cases. *Taiwan Veterinary Journal*, *46*(2–3), 1–8. https://doi.org/10.1142/S1682648520500067.

Muñoz-Blanco, L., & Salazar, V. (2025). Incidence of postoperative acute kidney injury in dogs without pre-existing renal disease. *Frontiers in Veterinary Science*, *12*, 1483101. https://doi.org/10.3389/fvets.2025.1483101.

Research Institute Public Health Science (2018). Research Institute Public Health Science, Department of Medical Sciences, Ministry of Public Health Thai-US Cooperation Center Public Health. *Bacteria and fungi operating manual for regional hospitals and general hospitals*. http://nih.dmsc.moph.go.th/data/data/61/LO_manual.pdf.

Rungphattanachaikul, S., Akatvipat, A., Chia, M.P.C., Na Lampang, K., & Sthitmatee, N. (2021). A retrospective study of suspected pyometra causing systemic illness in 348 dogs. *Veterinary Integrative Sciences*, *19*(2), 141–152. https://doi.org/10.12982/VIS.2021.013.

Santana, C.H., & Santos, R.L. (2021). Canine pyometra: An update and revision of diagnostic terminology. *Brazilian Journal of Veterinary Pathology*, *14*(1), 1–8. https://doi.org/10.24070/bjvp.1983-0246.v14i1p1-8.

Sasidharan, J.K., Patra, M.K., Khan, J.A., Singh, A.K., Karikalan, M., De, U.K., Saxena, A.C., Dubal, Z.B., Singh, S.K., Kumar, H., & Krishnaswamy, N. (2023). Differential expression of inflammatory cytokines, prostaglandin synthases, and secretory leukocyte protease inhibitor in the endometrium and circulation in different graded CEH-pyometra in bitches. *Theriogenology*, *197*, 139–149. https://doi.org/10.1016/j.theriogenology.2022.11.017.

Schüttler, J., & Neumann, S. (2015). Interleukin-6 as a prognostic marker in dogs in an intensive care unit. *Veterinary Clinical Pathology*, *44*(2), 223–238. https://doi.org/10.1111/vcp.12255.

Singh, L.K., Patra, M.K., Mishra, G.K., Saxena, A.C., De, U.K., Singh, S.K., Kumar, H., & Narayanan, K. (2020). Prospects of diagnostic and prognostic biomarkers of pyometra in canine. *Asian Pacific Journal of Reproduction*, *9*(4), 166–173. https://doi.org/10.4103/2305-0500.288584.

Smith, F.O. (2006). Canine pyometra. *Theriogenology*, *66*(3), 610–612. https://doi.org/10.1016/j.theriogenology.2006.04.023.

Turkki, O.M., Sunesson, K.W., & den Hertog, E., & Varjonen, K. (2023). Postoperative complications and antibiotic use in dogs with pyometra: A retrospective review of 140 cases (2019). *Acta Veterinaria Scandinavica*, *65*(1), 11. https://doi.org/10.1186/s13028-023-00670-5.

Vukmer, N.A., Urrego, H., & Dizon, A.M. (2021). Medical management of pyometra in the delayed postoperative period. *Case Reports in Obstetrics and Gynecology*, *2021*, 7995348. https://doi.org/10.1155/2021/7995348.

Woźna-Wysocka, M., Rybska, M., Błaszak, B., Jaśkowski, B.M., Kulus, M., & Jaśkowski, J.M. (2021). Morphological changes in bitch endometrium affected by cystic endometrial hyperplasia–pyometra complex: The value of histopathological examination. *BMC Veterinary Research*, *17*(1), 174. https://doi.org/10.1186/s12917-021-02875-0.

Xavier, R.G.C., Santana, C.H., Castro, Y.G., Souza, T.G.V., Amarante, V.S., Santos, R.L., & Silva, R.O.S. (2023). Canine pyometra: A short review of current advances. In J. Dorado (Ed.), *Animals (Basel)*, *13*(21), 3310. https://doi.org/10.3390/ani13213310.

Ylhäinen, A., Mölsä, S., Grönthal, T., Junnila, J., Rantala, M., Laitinen-Vapaavuori, O., & Thomson, K. (2023). A double-blinded randomized placebo-controlled non-inferiority trial protocol for postoperative infections associated with canine pyometra. *BMC Veterinary Research*, *19*, 77. https://doi.org/10.1186/s12917-023-03629-w.