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Diagnostic and prognostic value of C-reactive protein and lactate dehydrogenase in canine pyometra

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Abstract

Pyometra is one of the most common and life-threatening reproductive disorders in intact bitches, characterized by uterine infection and systemic inflammatory response. The present study was undertaken to evaluate the diagnostic and prognostic significance of C-reactive protein (CRP) and lactate dehydrogenase (LDH) in canine pyometra, along with associated biochemical alterations. A total of 24 bitches were included in the study, comprising 12 healthy controls and 12 pyometra-affected cases. Clinical evaluation, biochemical analysis, and ultrasonography were performed for diagnosis and monitoring. Blood samples were collected at the day of presentation, before surgery, and at the time of suture removal following ovariohysterectomy. Pyometra incidence was recorded at 24.58%, with higher occurrence in older (>7 years), nulliparous, and Labrador bitches. Biochemical analysis indicated elevated hepatic enzymes and renal parameters. CRP levels were markedly elevated in affected bitches from date of presentation (DOP) (98.69 ± 6.54 mg/L) to before surgery (BFS) (100.42 ± 5.92) compared to controls (4.82 ± 0.87 mg/L) and declined significantly following treatment (8.29 ± 0.53 mg/L). Whereas LDH (U/L) levels were significantly decreased from DOP (820.75 ± 90.55 U/L) to BFS (367.43 ± 172.57) and showed progressive reduction post-treatment (220.79 ± 15.47 U/L). The study concludes that CRP is a sensitive biomarker for systemic inflammation and therapeutic monitoring, while LDH serves as an indicator of tissue damage. These biomarkers, in combination with routine parameters, provide valuable tools for diagnosis, prognosis, and monitoring of canine pyometra.

Keywords: Canine pyometra, C-reactive protein, lactate dehydrogenase, biomarkers

Introduction

Canine pyometra is a common, hormonally mediated, and potentially fatal reproductive disorder characterized by the accumulation of purulent material within the uterine lumen. It predominantly affects intact, middle-aged to older bitches during the diestrus phase under the influence of progesterone (Smith, 2006; Hagman, 2023) ^[1, 2]. It develops typically between 4 weeks and 4 months after onset of estrus (Nelson and Feldman, 1986; Smith, 2006) ^[3, 1]. Epidemiological studies indicate that nearly 25% of intact bitches develop pyometra by 10 years of age (Egenvall *et al.*, 2001; Pretzer, 2008) ^[5, 6]. The disease is associated with systemic complications such as endotoxemia, septicemia, renal dysfunction, and hepatic impairment, which contribute to its high morbidity and mortality (Fieni, 2006) ^[7]. So far ovariohysterectomy is the best prophylactic and therapeutic method for pyometra in canines (Wheaton *et al.* 1989) ^[8] Surgical ovariohysterectomy remains the treatment of choice, although medical management may be considered in selected cases (Bartoskova *et al.*, 2007; Hagman, 2022) ^[9, 10].

Biochemical alterations commonly observed in pyometra reflect systemic inflammatory response and organ dysfunction secondary to endotoxin release (Nelson and Feldman, 1986; Johnston *et al.*, 2001) ^[3, 4]. In recent years, there has been increasing interest in the use of biomarkers such as C-reactive protein (CRP) and lactate dehydrogenase (LDH) for the diagnosis and monitoring of inflammatory and tissue-damaging conditions. CRP is a major acute-phase protein in dogs, rapidly increasing in response to inflammation, infection, and

tissue injury (Yamamoto *et al.*, 1994)^[12]. LDH, a cytoplasmic enzyme present in multiple tissues, is released into circulation following cellular damage and serves as a non-specific indicator of tissue injury (Fransson, 2003)^[14].

Despite advancements in diagnostic techniques, the prognostic value of these biomarkers in canine pyometra remains under investigation. Therefore, the present study was undertaken to evaluate the diagnostic and prognostic significance of CRP and LDH, along with biochemical parameters, in pyometra-affected bitches

Materials and Methods

The present study was conducted on 24 bitches presented to the Department of Veterinary Gynaecology and Obstetrics, NTR College of Veterinary Science, Gannavaram, Andhra Pradesh. The animals were divided into two groups: Group I (n=12) consisting of clinically healthy bitches, and Group II (n=12) comprising bitches diagnosed with cystic endometrial hyperplasia-pyometra complex.

Diagnosis was based on clinical history, physical examination, ultrasonographic findings, and biochemical analysis. Clinical parameters such as rectal temperature, heart rate, respiratory rate, and presence of vaginal discharge were recorded. Blood samples were collected aseptically from the cephalic vein at three time points: day of presentation (DOP), before surgery (BFS) and day of suture removal (12–14 days post-ovariohysterectomy). Serum biochemical parameters including ALT, AST, ALP, BUN, creatinine, CRP, and LDH were estimated using standard protocols.

Statistical analysis was performed using one-way ANOVA followed by Tukey's and Duncan's post hoc tests. A significance level of $P < 0.05$ was considered statistically significant.

Results and Discussion

Out of 484 bitches presented, 119 were diagnosed with pyometra, indicating an overall incidence of 24.58%. This highlights pyometra as a major reproductive disorder in bitches, consistent with earlier reports (Hagman, 2004; Smith, 2006)^[15, 1]. Other gynaecological conditions observed included pregnancy diagnosis (32.02%), breeding advice (24.79%), dystocia (6.81%), mismating (4.13%) and miscellaneous conditions (7.64%), reflecting increased clinical attention toward reproductive management.

The mean age of affected bitches was 7.15 ± 0.35 years (range: 2–18 years), with peak incidence in the 6–9 years age group (31.93%), followed by 3–6 years (26.05%). Lower occurrence was recorded in older age groups. This pattern confirms that middle-aged bitches are more susceptible, likely due to repeated progesterone exposure leading to cystic endometrial hyperplasia and uterine infection (Dow, 1959; Hagman, 2004)^[16, 15]. Breed-wise, Labradors (19.32%) showed the highest occurrence, followed by Pomeranian/Spitz (10.90%), Shih Tzu (9.83%) and Golden Retriever (7.56%). This variation may be influenced by breed popularity and hospital presentation trends, although breed predisposition has been suggested (Egenvall *et al.*, 2001)^[5]. Parity analysis revealed a higher occurrence in nulliparous bitches (67.22%), followed by primiparous (26.89%) and pluriparous (5.88%). This supports previous findings that absence of pregnancy increases the risk of pyometra due to prolonged hormonal exposure (Johnston *et al.*, 2001)^[11].

Based on cervical status, open cervix pyometra (89.07%) was more common than closed cervix (10.92%), likely due to easier clinical detection through vaginal discharge, as reported

earlier (Hagman, 2018)^[17]. However, closed cervix cases remain clinically significant due to their association with severe systemic complications.

Significant elevation of hepatic enzymes (ALT, AST, ALP) indicates hepatocellular damage and cholestasis associated with endotoxemia. Increased BUN and creatinine levels reflect renal impairment due to reduced perfusion and immune complex deposition (Fieni, 2006)^[7] (Table:1)

A highly significant elevation ($P < 0.01$) in serum hepatic enzymes was observed in pyometra-affected bitches, as evidenced by increased alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels compared to the control group. The mean ALT concentration was markedly higher during the preoperative period (46.26 ± 0.52 U/L) than in controls (27.32 ± 1.75 U/L), and although a slight decline was noted by the day of suture removal (40.35 ± 4.12 U/L), the values remained significantly elevated, indicating sustained hepatic stress. Similarly, AST levels showed a significant rise ($P < 0.01$) at the day of presentation (63.35 ± 6.86 U/L) and prior to surgery (65.22 ± 6.85 U/L), followed by a significant reduction at the time of suture removal (40.34 ± 3.28 U/L); however, these values continued to be higher than those of the control group. The persistent elevation of both ALT and AST suggests hepatocellular damage associated with systemic endotoxemia and inflammatory processes in pyometra, while the gradual decline post-surgery reflects progressive hepatic recovery following therapeutic intervention.

A marked elevation in alkaline phosphatase (ALP) levels was recorded in pyometra cases, with values reaching 216.58 ± 61.33 U/L at presentation and remaining significantly elevated throughout the study period ($P < 0.05$ – 0.01). This increase may be attributed to cholestasis and hepatobiliary dysfunction associated with systemic infection.

Renal parameters also showed significant alterations. Blood urea nitrogen (BUN) levels were significantly increased ($P < 0.05$ – 0.01) in affected bitches during the preoperative period (52.13 ± 5.20 mg/dl) compared to controls (20.13 ± 1.11 mg/dl), with a gradual decline observed postoperatively (45.36 ± 3.75 mg/dl).

Serum creatinine levels exhibited a highly significant increase ($P < 0.01$) at presentation (2.13 ± 0.24 mg/dl) and before surgery (1.81 ± 0.20 mg/dl), followed by a significant reduction (1.10 ± 0.19 mg/dl) at the time of suture removal, approaching near-normal values. The decline in these parameters post-surgery confirms the effectiveness of ovariohysterectomy in eliminating infection and restoring organ function and clinical recovery.

The mean C-reactive protein (CRP) concentration in the control group (Group I) was 4.82 ± 0.87 mg/L. In contrast, pyometra-affected bitches (Group II) exhibited a highly significant ($P < 0.01$) elevation in CRP levels at the day of presentation (98.69 ± 6.54 mg/L) and before surgery (100.42 ± 5.92 mg/L) compared to the control group. However, a marked decline in CRP concentration was observed on the day of suture removal (8.29 ± 0.53 mg/L), although values remained significantly higher than controls. (Fig.1)

Within Group II, CRP levels recorded at the day of presentation and before surgery did not differ significantly from each other, indicating sustained systemic inflammatory response during the preoperative period. In contrast, a significant reduction was noted postoperatively, suggesting resolution of inflammation following ovariohysterectomy. CRP levels were markedly elevated in pyometra cases, confirming severe systemic inflammation. The significant

reduction following treatment aligns with previous studies (Dabrowski *et al.*, 2013; Jitpean *et al.*, 2014)^[1, 18], elevated CRP may be attributed to increased synthesis of acute phase proteins in response to uterine infection and endotoxemia as were reported by Ahlen (2017)^[19] and Bostrom (2018)^[20] highlighting its value as a sensitive marker for disease monitoring.

Although CRP levels showed a declining trend following surgical intervention, they did not return completely to baseline levels by the time of suture removal. This may be due to persistent inflammatory responses induced by surgical trauma and tissue healing. Gogoi *et al.* (2022)^[21] and Hadzimusic *et al.* (2024) similarly reported sustained elevation of CRP during the postoperative period, attributing it to increased production of proinflammatory cytokines following ovariohysterectomy.

Furthermore, the absence of a significant difference between CRP levels at presentation and before surgery in the present study suggests that the inflammatory status remains relatively stable until surgical intervention. This observation aligns with findings of Saraswat *et al.* (2023)^[23] who reported persistently elevated CRP concentrations in untreated pyometra cases.

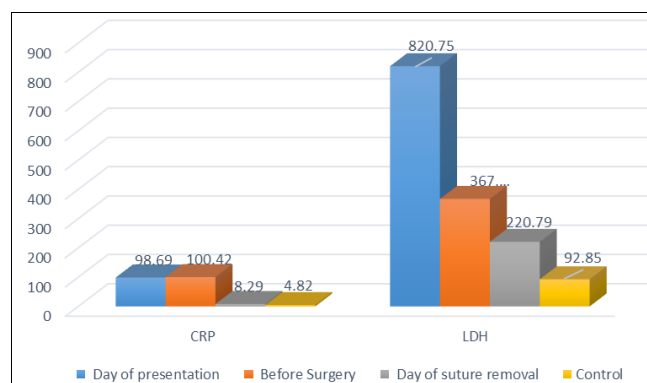


Fig 1: Histogram showing serum biomarkers CRP, LDH in healthy control and pyometra affected bitches

The mean lactate dehydrogenase (LDH) activity in the control group (Group I) was 92.85 ± 19.58 U/L. In contrast, pyometra-affected bitches (Group II) exhibited a highly significant ($P < 0.01$) elevation in LDH levels at the day of presentation (820.75 ± 90.55 U/L) compared to the control group. A progressive decline in LDH activity was observed during the course of treatment, with values decreasing to

367.43 ± 172.57 U/L before surgery and further to 220.79 ± 15.47 U/L on the day of suture removal. Although postoperative LDH levels showed a significant reduction ($P < 0.01$) compared to the day of presentation, they remained elevated relative to control values.

Within Group II, LDH activity at the day of presentation was significantly higher than both preoperative and postoperative values, whereas no significant difference was observed between the before surgery and day of suture removal stages, indicating gradual resolution of tissue damage following therapeutic intervention.

The present findings are in agreement with earlier reports. Kayamz *et al.* (1999) documented markedly elevated LDH concentrations (512 ± 271 U/L) in bitches with CEH-pyometra complex. Similarly, Yoon *et al.* (2017)^[25] reported increased LDH levels in affected dogs, with values exceeding the normal reference range. The marked elevation of LDH observed in the present study may be attributed to extensive tissue damage and cellular leakage associated with uterine infection, endotoxemia, and systemic inflammatory response in pyometra. LDH is a cytoplasmic enzyme present in multiple tissues, including muscle, liver, erythrocytes, kidneys, and pancreas, and its elevation reflects generalized cellular injury rather than organ-specific damage.

Furthermore, Fransson (2003)^[14] suggested that increased LDH activity in pyometra is more likely associated with muscle breakdown than hepatocellular injury. This is supported by the fact that LDH elevation may occur even when liver-specific enzymes remain within normal limits, indicating that the increase is primarily due to systemic effects such as toxemia, cellular hypoxia, and inflammatory tissue damage.

The declining trend in LDH levels following treatment observed in the present study is consistent with the findings of Hadzimusic *et al.* (2024) who reported significantly elevated LDH activity at the onset of disease followed by a reduction during subsequent days, reflecting recovery and reduction in tissue injury. The postoperative decrease in LDH in the present study further supports the effectiveness of ovariohysterectomy in eliminating the source of infection and reducing systemic inflammation. Elevated LDH levels indicate extensive tissue damage and cellular leakage. The gradual decline post-treatment reflects recovery and reduced cellular injury. However, due to its non-specific nature, LDH should be interpreted alongside other parameters (Fransson, 2003)^[14].

Table 1: Serum biomarkers (MEAN $\pm$ S.E) in healthy control group and pyometra affected bitches (pre-operative and post-operatively)

S. No.	Parameter	Control (Group I)	Pyometra (Group II)		
			Day of presentation (DOP)	Before surgery (BFS)	Day of suture removal
1	ALT (U/L)	27.32 ± 1.75	39.78 ± 8.39^a	$46.26 \pm 0.52^{a**}$	$40.35 \pm 4.12^{a**}$
2	AST (U/L)	27.32 ± 1.83	$63.35 \pm 6.86^{a**}$	$65.22 \pm 6.85^{a**}$	$40.34 \pm 3.28^{b**}$
3	ALP (U/L)	46.78 ± 4.38	$216.58 \pm 61.33^{a*}$	$152.83 \pm 12.21^{a**}$	$190.72 \pm 13.02^{a**}$
4	BUN (mg/dl)	20.13 ± 1.11	$53.76 \pm 12.17^{a*}$	$52.13 \pm 5.20^{a**}$	$45.36 \pm 3.75^{a**}$
5	Creatinine (mg/dl)	0.87 ± 0.04	$2.13 \pm 0.24^{a**}$	$1.81 \pm 0.20^{ab**}$	1.10 ± 0.19^b
6	CRP (mg/L)	4.82 ± 0.87	$98.69 \pm 6.54^{a**}$	$100.42 \pm 5.92^{a**}$	$8.29 \pm 0.53^{b**}$
7	LDH (U/L)	92.85 ± 19.58	$820.75 \pm 90.55^{a**}$	367.43 ± 172.57^b	$220.79 \pm 15.47^{b**}$

Different superscripts (a,b) within row indicate significant difference (** $P < 0.01$, * $P < 0.05$)

However, LDH is a nonspecific biomarker and may be elevated in various pathological conditions, as reported by Marcanto *et al.*, (2009) Therefore, its interpretation should be made in conjunction with other clinical and biochemical parameters.

Conclusion

The CRP concentration was found to vary according to the intensity of inflammatory activity (Enginler *et al.*, 2014)^[27] and hence CRP could be used as an end-point marker of treatment efficiency (Hagman, 2014)^[18]. The kinetic

properties of canine CRP during an acute phase response are characterized by a low concentration at normal homeostasis (Yamamoto *et al.*, 1994; Otabe *et al.*, 1998)^[12, 29], a short lag-phase after the inflammatory stimulus of around four hours (Higgins *et al.*, 2003)^[30], reaching a peak in concentration after approximately 24 h and a quick normalization after the end of inflammatory stimulus (Yamamoto *et al.*, 1992)^[31]. The findings of the present study revealed that estimation of serum CRP concentration could be utilised as an effective tool in the evaluation of treatment progress of canine pyometra.

The present study demonstrates that canine pyometra is associated with significant hematological and biochemical alterations reflecting systemic inflammation, hepatic dysfunction, and renal impairment. Among the biomarkers evaluated, C-reactive protein proved to be a highly sensitive indicator of inflammatory status and therapeutic response, while lactate dehydrogenase reflected the extent of tissue damage. The significant decline in CRP and LDH levels following ovariohysterectomy correlated well with clinical recovery, emphasizing their prognostic value. Therefore, the combined assessment of CRP, LDH, and routine hematobiochemical parameters provides a reliable approach for diagnosis, prognosis, and monitoring of treatment in canine pyometra.

Conflict of Interest

Not available

Financial Support

Not available

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