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Comparative study of efficacy on mifepristone and intravaginal PGF2α in pyometra in bitches

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Abstract
Retrospective data was collected and analysed to record the influence of various factors on incidence of canine pyometra at VCC, C.V.Sc, Rajendranagar, Hyderabad from January 2015 to December 2021 while, the present study focused on efficacy of the treatment with Mifepristone and PGF2α by intravaginally with help of a uterine catheter. Six reproductively sound bitches were considered as control group (Group I), 18 pyometra affected bitches were allotted randomly into 3 groups of 6 each and were treated with different protocols. Group II was treated with Mifepristone @ 2.5 mg/kg body weight per oral route for 5 days. Group III was treated with Mifepristone @ 2.5 mg/kg body weight p.o along with Cloprostenol intravaginal route @ 25 µg in small breed and 50 µg in large breeds. Group IV was treated with Mifepristone @ 2.5 mg/kg body weight along with Lutalyse @ 2.5 mg intravaginal route. Treatment was given until disappearance of clinical signs and confirmed by ultrasonography. Blood samples were collected on day 0, 7 and 14 to record the progress of treatment. Anemia, leukocytosis and neutrophilia were prominent in the haematological analysis while elevated ALP, BUN and creatinine were significant (p<0.05) in serum biochemical profile of dogs affected with pyometra. Ultrasonography was found as an accurate aid in the quantitative and qualitative evaluation in canine pyometra. Salivary arborisation changes was noticed on day 0, 7 and 14 where in partial or no crystallization was observed on day 0, however, crystallization was noticed on day 7 and 14. No observed side effect by the use of intravaginal administration of PGF2α. The efficacy in group II, III and IV were 83.33%, 100% and 100% respectively. In conclusion, combination protocols can be considered a better treatment choice as it resulted in complete evacuation of uterine pus with less side effects and cured the pyometra condition in bitch.

Keywords: Canine pyometra, mifepristone, intravaginal administration of prostaglandin, salivary arborisation

1. Introduction
Pyometra is accumulation of purulent material within the uterine lumen (Greco and Davidson, 2017). It is a common reproductive disorder of intact, diestral bitch affecting one fourth of all female dogs before the age of ten (Kumari Baithalu *et al*., 2010) [6] that often leads to loss of breeding potential and is also life threatening (Silva *et al*., 2020) [15]. Approximately 60% of bitches with pyometra suffer life-threatening organ dysfunction due to sepsis (Hagman, 2018) [4]. The mortality is assessed as 4% despite the modern treatment, which is mainly due to impairment of kidneys and septicemia (Prasad *et al*., 2017) [10]. Renal failure due to pyometra (10.0%) is highest in middle age groups (50.0%) and lowest in younger dogs (10.0%) (Tufani *et al*., 2015). High progesterone levels are critical (Santana and Santos, 2021) while, higher levels of IGF – 1 located in surrounding epithelial cells of the endometrium also play an important role in development of CEH-Pyometra complex in dogs (Kumari Baithalu *et al*., 2010) [6]. It is mostly associated with other uterine pathological conditions such as hydrometra, mucometra and chronic endometritis (Kumar and Saxena, 2018) [5]. Mucometra, hematometra or hydrometra are classified as sterile and the character of intrauterine fluids differs from pyometra where the fluid is seromucous, bloody or serous respectively and is mostly associated with CEH (Pretzer, 2008) [11]. As the use of prostaglandins by subcutaneous route showed side effects, an attempt was made to observe the effect of prostaglandins by intravaginal administration with help of uterine catheter.

2. Materials and Methods
Retrospective analysis of data collected since 2015 to 2021 to record the incidence of pyometra in bitches. A total of 24 bitches that attended to Department of Veterinary Gynaecology & Obstetrics, College of Veterinary Science, Rajendranagar, Hyderabad during

the period 2021-2022 were considered for this study. Six healthy and eighteen pyometra affected bitches were grouped as follows. Group I bitches were reproductively sound and considered as control group which remained healthy throughout the treatment period. Group II bitches were treated with administration of Mifepristone @ 2.5 mg/kg body weight once daily per oral route until complete cessation of pus discharge from vagina. Group III bitches were treated with a combination of Mifepristone @ 2.5 mg/kg body weight per oral for 5 days along with synthetic PGF2α, Cloprostenol sodium @ 25 µg/day intravaginal route with help of uterine catheter until complete cessation of pus. Group IV bitches were treated with a combination of Mifepristone @ 2.5 mg/kg body weight per oral for 5 days along with natural PGF2α, Lutalyse @2.5mg/day intravaginal route through catheter until complete cessation of pus discharge from vagina. Blood samples were collected using sterile disposable syringes from all the bitches on day 0, 7 and 14 on peripheral vein puncture (either cephalic or saphenous vein) in 4 ml BD vacutainers containing EDTA as anticoagulant (Levaram Life sciences Pvt. Ltd. India) in the respective groups. The samples were processed within 2–3 hours after collection for the hematological parameters *viz.* haemoglobin (Hb), Total Leukocyte Count (TLC) and Neutrophil count (%) performed on the day of blood collection as per standard technique. The serum was separated from blood samples collected in BD vacutainers without anticoagulant (Levaram Life sciences Pvt. Ltd. India) by centrifugation at 2000 rpm for 5 minutes and collected into serum collection vials. Sera samples were subjected to estimation of Alanine Amino Transaminase (ALT), Aspartate Transaminase (AST), Alkaline Phosphatase (ALP), Blood Urea Nitrogen (BUN) and creatinine as per standard technique on the same day of collection. Saliva was collected on day 0, 7 &14 to notice changes along course of treatment. Antibiotics Marbofloxacin @ 2.5 mg/kg body weight and Amoxicillin and Pottasium Clavulanate @ 20 mg/kg body weight per oral and advised to continue for 2 weeks in pyometra affected groups. Supportive therapy was given in the form of intravenous fluids, vitamin B-complex with liver extract, Hematinic and Metoclopramide hydrochloride.

3. Result and Discussion

3.1 Retrospective analysis

Data collected since 2015 to 2021 revealed an incidence of 15.28% as pyometra, 0.37% as mucometra, 0.06% as hematometra and 0.018% as hydrometra. As the type of fluid cannot be differentiated the incidence of mucometra, hematometra and hydrometra is not much known and is found accidentally during spaying of bitches. Based on patency 78% were diagnosed as open whereas 22% as closed pyometra with a mean age of 6.98±1.5 years mainly affected middle aged bitches. Most common breed presented was Labrador with 33.34% may be due to breed popularity in the location as per data pursued.

3.2 Haematological parameters

Hb, TLC and neutrophil percent are considered as one of the best indicators (Shah *et al*., 2017) [14]. Hb was significantly decreased (p<0.05) indicating anemic condition in affected bitches which is in accordance with Samantha *et al*. (2018) [12] and Ucmak *et al*. (2021). Contrarily, Pati *et al*. (2021) [9] witnessed non- significant decrease in Hb levels. Occurence of anemia could be by erythrocyte diapedesis into lumen of uterus and decreased erythropoiesis in the bone marrow as a result of chronic inflammatory disease (Thangmani *et al*., 2018 and Ucmak *et al*., 2021) and degree of involvement of infection. TLC and Neutrophils were significantly (p<0.05) increased in pyometra affected bitches when compared with control group. It is in accordance with Samantha *et al*. (2018) [12] that leukocytosis was characteristic feature in almost all affected bitches and may be attributed to the uterine defense mechanism to microorganisms. However, the values decreased to normal physiological values significantly by the end of treatment (Table 1).

3.3 Biochemical parameters

ALT and AST levels in serum before treatment did not differ significantly from control group bitches suggesting lack of liver and hepatocellular damage which is in accordance with Shah *et al*. (2017) [14] who reported normal physiological values. Serum ALP levels increased significantly (p<0.05) in affected bitches when compared to control group which may be due to toxemia that is in accordance with Samantha *et al*. (2018) [12] and Llazani *et al*. (2021) [7]. BUN and serum creatinine levels were significantly increased (Table 2) when compared to control group and was high in closed pyometra cases than in open pyometra cases due to the bacterial endotoxins effect. Verstegen *et al*. (2008) [21] reported that serum BUN and creatinine concentrations were not usually elevated, unless pre –renal azotemia develops due to dehydration. Contrarily, Hagman *et al*. (2009) [3] reported normal BUN and creatinine values.

3.4 Changes in Salivary arborisation and ultrasonography during treatment

Sample was taken by imprinting a frosted slide over tongue of animal, later air dried and examined. On day 0 less or no pattern of crystallization was observed indicating low amount of estrogen (diestrus phase or metestrus phase). It is in accordance with (Pardo-Carmona *et al*., 2010 and Skliarov, 2021) [17] based on phenomenon of crystallization due to increased estrogen in the stage of proestrus-estrus. Ultrasonography was done to measure the diameter of uterus on day 0 and day 14. Except one bitch in group II all bitches responded to treatment.

3.5 Efficacy of different treatment protocols

Efficacy was measured in terms of cessation of discharge, disappearance of clinical signs and changes in hematological and biochemical parameters, ultrasonography. The efficacy rate of Group II, Group III and Group IV were 83.33%, 100% and 100% respectively. Closed pyometra opened after 36-48 hours of onset of mifepristone therapy as evidenced by onset of vulvar discharge. The removal of inhibitory effect of progesterone on uterine contractions by mifepristone have opened cervical canal while ecbolic effect in uterine evacuation by promoting contractions (Shah *et al*., 2016) [13] In Group II, 2 bitches showed side effects vomition and 1 bitch showed retention of pus within the uterus at the end of treatment. No side effects were observed by administration of prostaglandin by intravaginal route with help of uterine catheter which is in accordance with Gabor *et al*. (1999) [1]. Addition of Mifepristone improved the recovery rate in pyometra affected bitches (Singh *et al*., 2019) [16].

Table 1: Haematological parameters

Parameter studied			Groups	Day 0		Day 7		Day 14	
Haemoglobin (g/dl)			Group I	15.75±0.96 a		15.75±0.96	a	15.75±0.96	
			Group II	11.20±0.40	b,x	12.57±0.43	b,x	14.07±0.96 y	
			Group III	11.48±0.66	b,x	13.08±0.66	b,x	14.25±0.40 y	
			Group IV	11.30±0.38	b,x	12.22±0.42	b,x	14.15±0.34 y	
Total (103/µl)	Leucocyte	count	Group I	13.67±0.98	a	13.67±0.98	a	13.67±0.98	a
			Group II	35.74±3.70	b,x	24.17±1.50	c,y	19.47±0.90 b,y	
			Group III	35.15±3.40	b,x	19.28±1.12	b,y	15.57±1.09 a,c,y	
			Group IV	35.18±3.20	b,x	23.35±1.12	b,c,y	18.38±0.35 b,c,y	
Neutrophils (%)			Group I	69.50±2.60	a	69.50±2.60	a	69.50±2.60	
			Group II	86.57±0.68	b,x	78.70±2.22	b,x	73.32± 2.67	y
			Group III	86.15±0.98	b,x	76.60±1.70	a,y	71.58 ±1.61	y
			Group IV	86.80±1.10	b,x	77.60±1.79	a,y	72.20 ±1.51	y

Table 2: Serum biochemical parameters

Parameters studied	Groups	Day 0	Day 7	Day 14
Aspartate Trasaminase (U/L)	Group I	50.78±5.93	50.78±5.93	50.78±5.93
	Group II	57.18±8.12	54.23±8.2	52.73±7.5
	Group III	56.95±4.8	53.95±5.2	51.45±5.3
	Group IV	56.30±2.3	53.42±3.5	51.25±4.17
Alanine Transaminase(U/L)	Group I	39.12±6.9	39.12±6.9	39.12±6.9
	Group II	35.75±1.7	37.92±1.9	38.92±1.6
	Group III	35.27±1.5	38.43±1.5	40.43±1.9
	Group IV	36.50±2.3	38.02±1.66	39.52±1.08
Alkaline Phosphatase(U/L)	Group I	67.35±14.06 a	67.35±14.06	67.35±14.06
	Group II	210±36.80 b,x	87.63±11.86 y	68.80±14.00 y
	Group III	211.8±14.43b,x	83.75±7.18 y	66.25±1.91y
	Group IV	209.1±34.04b,x	85.77±4.15 y	67.93±3.93 y
Blood urea nitrogen (mg/dl)	Group I	20.00±1.51a	20.00±1.51	20.00±1.51
	Group II	32.50±2.20b,x	23.50±1.78y	20.00±1.06y
	Group III	33.17±1.53b,x	20.17±1.35y	18.00±1.77y
	Group IV	32.67±1.53b,x	21.8±1.53y	19.33±0.95y
Creatinine (mg/dl)	Group I	1.01±0.11a	1.01±0.11	1.01±0.11
	Group II	1.93±0.15b,x	1.40±0.17x	1.05±0.09y
	Group III	2.03±0.99b,x	1.78±0.12y	0.93±0.04y
	Group IV	1.86±0.98b,x	1.33±0.12x	0.95±0.08y

Values are Mean±SE (n=6); One way ANOVA. Mean with different superscripts in a column: a, b, c, d and row: x, y differs significantly (p<0.05).

4. Conclusion

In the present study, combination protocols used in group III and IV were found advantageous over group II where Mifepristone was used alone with no side effects by use of intravaginal administration of prostaglandins. Ultrasonography and haemato-biochemical changes in correlation with stage of estrus was useful in diagnosis of pyometra in canines.

5. References

1. Gabor G, Siver L, Szenci O. Intravaginal prostaglandin f 2α for the treatment of metritis and pyometra in the bitch. Acta Veterinaria Hungarica. 1999;47(1):103-108.
2. Greco DS, Davidson AP. Blackwell's Five-Minute Veterinary Consult Clinical Companion: Small Animal Endocrinology and Reproduction. John Wiley and Sons, 2017.
3. Hagman R, Reezigt BJ, Bergström Ledin H and Karlstam E. Blood lactate levels in 31 female dogs with pyometra. Acta Veterinaria Scandinavica. 2009;51(1):1-9.
4. Hagman R. Pyometra in small animals. Veterinary Clinics: Small Animal Practice. 2018;48(4):639-661.
5. Kumar A, Saxena A. Canine pyometra: Current perspectives on causes and management: A review. The Indian Journal of Veterinary Sciences and Biotechnology. 2018;14(01):52-56.
6. Kumari Baithalu R, Maharana BR, Mishra C, Sarangi L, Samal L. Canine pyometra. Veterinary World. 2010;3(7):340.
7. Llazani M, Qoku A, Dhaskali L. Laboratory Findings, Vaginal Cytology and Histopathology in Bitches with Cystic Endometrial Hyperplasia–Pyometra Complex. European Journal of Biology and Biotechnology. 2021;2(3):61-63.
8. Pardo-Carmona B, Moyano MR, Fernández-Palacios R, Pérez-Marín CC. Saliva crystallisation as a means of determining optimal mating time in bitches. Journal of Small Animal Practice. 2010;51(8):437-442.
9. Pati P, Mishra PC, Patra BK. Pyometra in bitches: A haematological, biochemical and ultrasonographical study. The Pharma Innovation Journal. 2021;10(1):98-101.
10. Prasad VD, Kumar PR, Sreenu M. Pyometra in bitches: A review of literature. Research & Reviews: Journal of Veterinary Science and Technology. 2017;6(2):12-20.
11. Pretzer SD. Clinical presentation of canine pyometra and mucometra: A review. Theriogenology. 2008;70(3):359-363.
12. Samantha G, Sarath T, Monica G, Arunmozhi N, Sridevi P, Joseph C. Ultrasonographic and haemato-biochemical

evaluation of bitches affected with cystic endometrial hyperplasia-pyometra complex. International Journal of Current Microbiology and Applied Science. 2018;7(6):2327-2338.
13. Shah MA, Pande N, Shah IA, Agrawal R, Sharma U, Ghuman SPS. Treatment of pyometra in female dogs using prostaglandin f2α±antiprogestin (mifepristone). The Indian Journal of Animal Reproduction. 2016;37(1):23-36.
14. Shah SA, Sood NK, Wani BM, Rather MA, Beigh AB, Amin U. Haemato-biochemical studies in canine pyometra. Journal of Pharmacognosy and Phytochemistry. 2017;6(4):14-17.
15. Silva E, Silva MF, Henriques S, Diniz P, Carneiro C, Lopes-da-Costa L, Mateus L. The Role of Uteropathogenic *Escherichia coli* in the Development of Canine Pyometra. Advances in Animal Health, Medicine and Production, 2020, 510-527.
16. Singh LK, Patra MK, Mishra GK, Linda N, Saxena AC, De UK, *et al*. Comparative efficacy of antiprogestin and dopamine agonist in the treatment of canine pyometra. The Indian Journal of Animal Reproduction. 2019;40(2):28-31.
17. Skliarov PM, Holubiev OV, Bilyi DD, Roman LH, Lieshchova MO, Bobrytska OM. Efficiency test microscope Arbor Elite for determining the fertile period of bitches. Scientific Messenger of LNU of Veterinary Medicine and Biotechnologies. Series: Veterinary Sciences. 2021;23(101):55-60.
18. Thangamani A, Srinivas M, Chandra Prasad B. Pyometra in bitches: A critical analysis. International Journal of Science, Environment and Technology. 2018;7(3):1072-1078.
19. Tufani NA, Singh JL, Kumar M, Gupta D, Shekhar P, Rajora VS. Renal failure in Indian dogs: An epidemiological study. Indian Journal of Veterinary Medicine. 2015;35(1):7-11.
20. Uçmak ZG, Kurban İ, Uçmak M. Assessment of Hematological Parameters and Uterine Hemodynamic Indices in Bitches with Pyometra. Acta Scientiae Veterinariae. 2021;49:1796.
21. Verstegen J, Dhaliwal G, Verstegen-Onclin K. Mucometra, cystic endometrial hyperplasia, and pyometra in the bitch: Advances in treatment and assessment of future reproductive success. Theriogenology. 2008;70(3):364-374.