

Comparison of preoperative fluid therapy protocols associated with inhalational or total intravenous anesthesia for anesthetic procedures in dogs with sepsis

Comparação de protocolos de fluidoterapia pré-operatória associados a anestesia inalatória ou total intravenosa para procedimentos anestésicos em cães com sepse

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Abstract

This randomized clinical trial aimed to evaluate different fluid therapy protocols associated with inhalational or total intravenous anesthesia in the cardiorespiratory stability of bitches with sepsis subjected to a surgical procedure to control the infectious focus. Thirty-two bitches diagnosed with pyometra and sepsis and treated at the University Veterinary Hospital between 2018 and 2019 were recruited. After admission, diagnosis, clinical, and laboratory evaluation, patients were randomly distributed into the following groups: propofol 5 (P[5]: preoperative restrictive fluid therapy-5mL/kg/h and intravenous general anesthesia); propofol 10 (P[10]: preoperative liberal fluid therapy-10mL/kg/h and intravenous general anesthesia); and isoflurane 5 (I[5]: preoperative restrictive fluid therapy-5mL/kg/h and inhalational general anesthesia). Lactate on admission (LAC1) and release (LAC2), heart rate (HR), respiratory rate (RR), systolic blood pressure (SBP), rectal temperature (RT), oxygen saturation (SpO₂), and carbon dioxide extraction rate (EtCO₂) were analyzed at PRE, T10, T20, T30, T40, T50, TEXT, and TDIS. Clearance of 20% of lactate occurred in 18 dogs, with the P[10] group displaying the best performance. There was no statistical difference in vasopressor requirements among the groups. Liberal fluid therapy showed greater cardiovascular stability than restrictive therapy in the perioperative period. Regarding general anesthesia, isoflurane showed greater cardiorespiratory stability than propofol during anesthetic maintenance. In conclusion, although the three proposed protocols are safe and there is no difference in their superiority, some observed changes may be relevant and considered when it is possible to individualize the therapy for the patient.

Keywords: bitches, pyometra, cardiovascular stability, organ dysfunction, arterial hypotension.

Resumo

Este ensaio clínico randomizado teve a proposição de avaliar diferentes protocolos de fluidoterapia associados a anestesia inalatória ou total intravenosa, na estabilidade hemodinâmica de cadelas com sepse, submetidas a procedimento cirúrgico, para controle do foco infeccioso. Foram incluídas trinta e duas cadelas, com diagnóstico de piometra e sepse, atendidas em hospital veterinário universitário, no período de 2018 a 2019. Após admissão, diagnóstico, avaliação clínica e laboratoriais, os pacientes foram distribuídos de maneira aleatória nos grupos Propofol 5 (P[5]: fluidoterapia restritiva pré-operatória - 5mL/kg/h e anestesia geral intravenosa), Propofol 10 (P[10]: fluidoterapia liberal pré-operatória - 10mL/kg/h e anestesia geral intravenosa) e Isoflurano 5 (I[5]: fluidoterapia restritiva pré-operatória - 5mL/kg/h e anestesia geral inalatória). Foram analisados lactato na admissão (LAC1) e liberação (LAC2), frequências cardíacas (HR) e respiratória (RR), pressão arterial sistólica (SBP), temperatura retal (RT), saturação de oxigênio (SpO₂) e taxa de extração de dióxido de carbono (EtCO₂) nos seguintes momentos: PRE, T10, T20, T30, T40, T50, TEXT e TDIS. A depuração de 20% do lactato ocorreu em 18 cães, tendo o grupo P[10] o melhor desempenho. Não houve diferença estatística no requerimento de vasopressores entre os grupos. A fluidoterapia liberal apresentou maior estabilidade cardiovascular quando comparado



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com a restritiva no período perioperatório. Com relação a anestesia geral, o isoflurano apresentou maior estabilidade cardiorrespiratória que o propofol na manutenção anestésica. Nós concluímos que apesar de os três protocolos propostos serem seguros e sem diferença em superioridade entre eles, algumas alterações observadas podem ser relevantes e ponderadas quando for possível a individualização da terapêutica no paciente.

Palavras-chave: cadelas, piometra, estabilidade cardiovascular, disfunção orgânica, hipotensão arterial.

Introduction

Sepsis is defined as a potentially fatal organ dysfunction caused by a dysregulated host response to infection (Singer et al., 2016). It is one of the main causes of death in humans, causing damage to health systems and impacting the quality of life of survivors (Fleischmann et al., 2016). In veterinary medicine, owing to the scarcity and conflicting data in the literature, its challenges range from the identification of septic animals to proven effective species-specific treatment (Montealegre & Lyons, 2021).

The swiftness at which sepsis is identified for early institution of treatment is the main factor for reducing mortality (Evans et al., 2021). Fluid replacement, as well as the removal of the infectious focus as soon as possible, is the first line of approach (Evans et al., 2021; Rhodes et al., 2015). In dogs, several diseases that can lead to sepsis require surgical treatment, including pyometra, gastric dilatation-volvulus, gastrointestinal tract perforations, and contaminated wounds (Otto, 2007). As these surgical procedures must be performed on an emergency basis (Evans et al., 2021), the choice of protocols for pre-anesthetic stabilization (including fluid challenge and fluid replacement) and anesthesia techniques is crucial (Noel-Morgan & Muir, 2018; Singer et al., 2016) because excessively low or high volumes of fluids or vasodilatation can impair the already compromised tissue perfusion due to microcirculatory dysfunction (Evans et al., 2021; Grubb et al., 2020).

The present study compared bitches with pyometra and sepsis, two preoperative fluid therapy protocols (5 or 10 mL/kg/h), and their effects on the stability of two anesthetic protocols (inhalational or total intravenous anesthesia). The hypothesis was that both volumes would be well tolerated by dogs, showing no significant differences in cardiorespiratory stability during the perioperative period.

Materials and methods

This randomized clinical trial was conducted after approval by the Research Ethics Committee of the UFRRJ (CEUA 3269190418) from March 2018 to November 2019. The admission criteria were bitches of any age, breed, and body weight, with clinical and ultrasound diagnosis of pyometra, at least one organ dysfunction according to the criteria described in Chart 1, and confirmation of the infectious focus through uterine exudate culture. Female dogs with comorbidities and possible systemic repercussions were excluded. All animals were admitted, evaluated, treated, and followed-up by the same researchers, after owners' consent.

Chart 1. Criteria to identify organ dysfunction in bitches with pyometra at the Veterinary Hospital of Federal Rural University of Rio de Janeiro (UFRRJ, Seropédica, Brazil). Period from March 2018 to November 2019.

Organ Dysfunction	Criteria
Renal	Serum creatinine > 1.7 mg/dL
Cardiovascular	Systolic blood pressure < 90 mmHg
Hemostatic	Platelet counts < 200,000/μL
Neurologic	Glasgow Coma Scale modified for dogs ¹ ≤13 points

¹Andrade et al. (2010).

Admission and stabilization

Data on the history and anamnesis of the bitches were recorded after pyometra diagnosis. A complete physical examination was performed at the time of admission (ADM), in which the

following physiological variables were recorded: heart rate (HR), respiratory rate (RR), capillary refill time (CRT), systolic blood pressure (SBP) via Doppler, rectal temperature (RT), and level of consciousness using the Pediatric Glasgow Coma Scale adapted for dogs (Andrade et al., 2010) (Chart 2). Blood samples were collected from the jugular vein for blood count and serum biochemistry (total bilirubin, creatinine, total protein, and albumin), plasma lactate (LACI- mmol/L), and glucose (GLU- mg/dL). Based on the dog's size, an 18-to 22-G cannula was placed in the cephalic vein. Animals that presented with hypoglycemia (GLU < 80 mg/dL) were immediately treated with 25% glucose solution (1 mL/kg, IV). Dogs with arterial hypotension (SBP < 90 mmHg) were subjected to the fluid challenge protocol with lactated Ringer's solution (LRS), as described in Chart 3. The choice of administering items 1, 2, and 3 was individualized for each patient according to the decision of the attendant. Antibiotic therapy was initiated within the first 30 min with intravenous administration of ceftriaxone (50 mg/kg, Triaxon® - Teuto). Other supportive medications were administered as required by the patient.

Chart 2. The Pediatric Glasgow Coma Scale Modified for Dogs.

INDICATOR	CRITERIA/RESPONSE	SCORE
Eye-opening	Spontaneous	4
	Verbal stimulation/command	3
	Verbal stimulation/command/shouting	3
	Painful stimulus	2
	Without opening	1
Better response to vocalization	Bark/growl	5
	Angry crying	4
	Crying in pain	3
	Yawn to pain	2
	No reply	1
Best motor response	Spontaneous and normal movement	6
	Touch reaction	5
	Reaction to pain	4
	Abnormal flexion - decortication	3
	Abnormal extension - decerebration	2
	None	1
Total		15

Adapted from Andrade et al. (2010).

Chart 3. Fluid challenge protocol for bitches with pyometra and sepsis at the Veterinary Hospital of Federal Rural University of Rio de Janeiro (UFRRJ, Seropédica, Brazil). Period from March 2018 to November 2019.

If SBP < 90 mmHg	1. 10 mL/kg of LRS within 15 min; up to two boluses
	2. Administration of 2 mL/kg 7,0% NaCl solution, IV
	3. Continuous rate infusion (CRT) of 0,5-2mcg/kg/min of Norepinephrine, IV

Treatments

After cardiovascular stabilization (SBP ≥ 90 mmHg), each animal was randomly assigned to a treatment group, associated with a preoperative fluid therapy protocol (5 or 10 mL/kg/h, totaling

two hours of administration) and an anesthetic protocol (isoflurane or propofol anesthesia) to be instituted for the surgical procedure (ovariohysterectomy), as described in Chart 4. During the entire preoperative fluid therapy period, vital parameters were monitored, and adverse effects were recorded.

Chart 4. Preoperative fluid therapy and anesthetic protocol for bitches with pyometra and sepsis at Veterinary Hospital of Federal Rural University of Rio de Janeiro (UFRRJ, Seropédica, Brazil). Period from March 2018 to November 2019.

	Fluid Therapy	Anesthetic Protocol
P[5]	Administration of LRS at a rate of 5 mL/kg/h for 2 hours	Pre-anesthetic medication with 0.3 mg/kg of Morphine (IM); anesthetic induction with 4-6 mg/kg Propofol (IV); and anesthetic maintenance with CRI 0.2-0.6 mg/kg/min of 1% Propofol associated with 5-10 µg/kg/h of Fentanyl.
P[10]	Administration of LRS at a rate of 10 mL/kg/h for 2 hours	Pre-anesthetic medication with 0.3 mg/kg of Morphine (IM); anesthetic induction with 4-6 mg/kg propofol (IV); and anesthetic maintenance with CRI 0.2-0.6 mg/kg/min of 1% Propofol associated with 5-10 µg/kg/h of Fentanyl.
I[5]	Administration of LRS at a rate of 5 mL/kg/h for 2 hours	Pre-anesthetic medication with 0.3 mg/kg of Morphine (IM); anesthetic induction with 4-6 mg/kg Propofol (IV); and anesthetic maintenance with 1.0-1.2 MAC of Isoflurane associated with CRI 5-10 µg/kg/h of Fentanyl.

Perioperative period

After two hours of fluid therapy, all bitches underwent ovariohysterectomy (OH) according to the technique established by Fossum (2014). Before pre-anesthetic medication, the animals were submitted to a second clinical evaluation (HR, RR, SBP, RT, CRT - considered as PRE time); then, 0.3 mg/kg of morphine (Dimorf® - Cristália Ltda.) was administered intramuscularly (IM). Twenty minutes later, the animals were induced with propofol (2-6 mg/kg, Propovan® - Cristália Ltda.) until loss of consciousness and jaw tone; orotracheal intubation was performed, 100% oxygen was administered, and all of the animals were maintained on spontaneous ventilation in a circuit with or without gas rebreathing, depending on the size of the patient. Inhalation anesthesia with isoflurane (Isoforine® - Cristália Ltda.) was performed using a universal vaporizer and monitored using a gas analyzer (LifeWindow 9x®, Digicare Animal Health). Total intravenous anesthesia with propofol was administered using infusion pumps (DigiPump SR31x - Digicare®). All groups underwent continuous infusion of fentanyl (5-10 µg/kg/h, IV - Fentanest® - Cristália Ltda.) and fluid therapy with RLS in a total of 5 mL/kg/h of fluids (including the volumes of continuous infusion drugs). Anesthesia duration was determined from the administration of pre-anesthetic medication to the moment of extubation.

Patient monitoring was continuous using reflex responses and a multiparametric monitor (LifeWindow 9x® - Digicare Animal Health); HR, RR, RT, SpO₂, and EtCO₂ values, as well as SBP by vascular Doppler, were recorded every 10 min until the end of the surgical procedure. During the intraoperative period, if the bitch had an SBP < 90 mmHg for more than three minutes, the anesthetic plan was adjusted if possible. If arterial hypotension persisted, fluid challenge with LRS was instituted, as shown in Chart 3. Adjustments in the rates of propofol or isoflurane, fentanyl, and other adverse effects during the intraoperative period were recorded. At the end of the surgical procedure, uterine exudate samples were collected for microbiological analyses. The extubation time was recorded, and a new clinical evaluation with glucose (GLU2) and lactate (LAC2) measurements was performed 40 min after extubation.

The animals were referred to a veterinary inpatient service after administration of supportive medications as needed, and all bitches were followed up for up to 90 days. Deaths within 14 d of treatment were considered to be sepsis-related. Animals that were euthanized for financial reasons were excluded from the study.

Statistical analysis

Statistical analyses were performed using the SigmaStat 11.0 program. All data were subjected to the Shapiro-Wilk normality test; parametric data were described as the mean and standard deviation ($X \pm SD$), and nonparametric data as median and range ($M[\text{min-max}]$). The admission results (ADM) of patients allocated to groups P[5], P[10], and I[5] were compared using one-way ANOVA followed by the post hoc Student-Newman-Keuls or Kruskal-Wallis tests. To evaluate the effect of fluid challenge (ADM *vs.* PRE), a paired t-test or Wilcoxon test was performed; for the evaluation of parameters over time, repeated one-way ANOVA measures were followed by Student-Newman-Keuls or Friedman test. To assess the effects of non-superiority between the protocols (5 mL/kg/h *vs.* 10 mL/kg/h and isoflurane *vs.* propofol), the P[10] and I[5] groups were compared with the P[5] group (P[5] *vs.* P[10] and P[5] *vs.* I[5]) using the unpaired t-test for parametric data or the Mann-Whitney test for non-parametric data. The significance level was set at $P < 0.05$.

Results

Thirty-two bitches were included in this study, with 11 animals in the P[5] group, 10 in group P[10], and 11 in group I[5]. The average age of the animals was 7 ± 4 years, and their average body weight was 16.0 ± 8.5 kg. Ten breeds were represented, with the most common being mixed breeds ($n=18$), pinschers ($n=3$), poodles ($n=3$), dachshunds ($n=2$), American Pit Bull Terrier ($n=1$), Bulldog ($n=1$), Chow-Chow ($n=1$), Cocker Spaniel ($n=1$), Labrador Retriever ($n=1$), and Yorkshire Terrier ($n=1$). No significant differences were observed in the clinical and laboratory findings among the groups. The median number of organ dysfunctions observed was 2[1-5] dysfunctions, with no statistical difference between the groups regarding the amount or type of dysfunction. The clinical and laboratory findings related to the admission examination are described in Table 1.

Arterial hypotension was identified on admission in 7 bitches (22%), of which 5 (71%) responded positively to the bolus of LRS. Only one animal allocated to the P[5] group and one in the P[10] group required vasopressor administration (norepinephrine) to treat arterial hypotension upon admission. In these animals, continuous infusion of norepinephrine was maintained until discharge (DIS). Regarding the fluid therapy protocol established before the surgical procedure (ADM *vs.* PRE), none of the patients experienced an episode of arterial hypotension or any change worth noting. At the end of 2 h, the average SBP of the P[5] group increased significantly (105 ± 28 *vs.* 126 ± 20 , $p = 0.006$), whereas no significant changes were observed in the other groups. Rectal temperature decreased significantly in P[5] ($38.6 [37.5-39.8]$ *vs.* $37.4 [36.8-39.2]$, $p = 0.014$) and P[10] ($38.9 [36.8-39.9]$ *vs.* $37.5 [36.2-38.1]$, $p < 0.001$). There was no statistical difference between the groups for the other parameters, and all animals were considered clinically stable and suitable for the surgical procedure.

The average anesthesia time was 88 ± 26 min, and the surgery time was 46 ± 17 min, with no significant difference between the groups. Disregarding the two animals from the P[5] and P[10] groups that were already under continuous vasopressor infusion since admission, three animals from the P[5] group presented with an episode of arterial hypotension during the surgical procedure. They were treated effectively with vasopressors after fluid challenge. During anesthesia, the average SBP was significantly lower in all three groups, but statistically and clinically relevant only in P[5], especially 10 min after induction of anesthesia (T10). In the post-extubation assessment (EXT), the average SBP of group I[5] was significantly higher than that of group P[5]. Rectal temperature (RT) was significantly lower in all groups; however, the averages observed in I[5] were significantly higher than those observed in P[5], from the PRE time (before pre-anesthetic medication) until the patients' discharge (DIS) (Table 2).

Considering that approximately 4 h elapsed from the clinical examination at the admission of the patient (when the first plasma lactate concentration LAC1 was measured) to the second measurement of lactate (LAC2), the desired clearance of 20% was achieved in 13 patients (41%):

Table 1. Clinical and laboratory findings of bitches with sepsis submitted to preoperative fluid therapy (5 or 10 mL/kg/h) and total intravenous (P) or inhalational (I) anesthesia for emergency ovariohysterectomy, period from March 2018 to November 2019. Veterinary Hospital of UFRRJ, Seropédica, Brazil.

	All n=32	P[5] n=11	P[10] n=10	I[5] n=11
<i>age (yrs. old)</i>	7 ± 4	7 ± 3	8 ± 4	7 ± 4
<i>body weight (kg)</i>	16.0 ± 8.5	15.0 ± 10.8	15.9 ± 7.1	17.2 ± 7.8
Status at admission				
<i>Systolic blood pressure (mmHg)</i>	119 ± 30	105 ± 28	110 ± 36	127 ± 25
<i>Blood hypotension (<90mmHg)</i>	9 (28%)	5 (45%)	2 (20%)	2 (18%)
<i>Capilar refill time</i>	2 [2-4]	3 [2-4]	2 [2-4]	3 [2-3]
<i>Capilar refill time ≥ 3 sec</i>	10 (31%)	6 (54%)	2 (20%)	2 (18%)
<i>Lactate (mmol/L)</i>	3.2 [2.0-6.8]	2.9 [2.4-5.4]	3.7 [2.7-4.9]	3.2 [2.0-6.8]
<i>Glucose (mg/dL)</i>	86 [63-156]	84 [63-155]	86 [77-156]	86 [69-142]
<i>Hypoglycemia (<80 mg/dL)</i>	10 (31%)	5 (45%)	2 (20%)	3 (27%)
<i>Rectal temperature (°C)</i>	38.6 [36.7-40.3]	38.6 [37.5-39.8]	38.9 [36.8-39.9]	38.6 [36.7-40.3]
<i>Hypothermia (<38.1°C)</i>	8 (25%)	3 (27%)	3 (30%)	2 (18%)
<i>Hyperthermia (>39.2°C)</i>	10 (31%)	3 (27%)	4 (40%)	3 (27%)
<i>Total protein (ref. 5.4 - 7.5 g/dL)</i>	7.8 ± 1.5	8.3 ± 2.0	7.5 ± 0.8	7.7 ± 1.2
<i>Albumin (ref. 2.3 - 3.1 g/dL)</i>	1.9 ± 0.5	1.8 ± 0.4	2.2 ± 0.5	1.8 ± 0.5
<i>Hypoalbuminemia (< 2.3 g/dL)</i>	27 (84%)	10 (90%)	7 (70%)	10 (90%)
Number of organs dysfunction				
1	10 (31%)	3 (27%)	4 (40%)	3 (27%)
2	10 (31%)	1 (9%)	4 (40%)	5 (45%)
3	7 (22%)	3 (27%)	2 (20%)	2 (18%)
4	3 (9%)	2 (18%)	0	1 (9%)
5	2 (6%)	2 (18%)	0	0
Type of organ dysfunction				
<i>renal</i>	7 (22%)	3 (27%)	3 (30%)	1 (9%)
<i>hepatic</i>	19 (59%)	7 (64%)	5 (50%)	7 (64%)
<i>neurologic</i>	15 (47%)	9 (82%)	2 (20%)	4 (36%)
<i>hemostatic</i>	23 (72%)	8 (73%)	6 (60%)	9 (82%)
<i>cardiovascular</i>	9 (28%)	5 (45%)	2 (20%)	2 (18%)
<i>vasopressor requirement</i>	2 (6%)	1 (9%)	1 (9%)	0
<i>Lactate>2.5 mmol/L</i>	27 (84%)	9 (82%)	10 (100%)	8 (73%)
Changes during anesthesia				
<i>Hypotension episode</i>	5 (16%)	4 (36%)	1 (10%)	0 (0%)
<i>Vasopressor requirement</i>	3 (9%)	3(27%)	0	0
Postoperative evaluation				
<i>Surgery time (min)</i>	46 ± 17	48 ± 20	49 ± 18	41 ± 12
<i>Anesthesia time (min)</i>	88 ± 26	104 ± 27	92 ± 25	69 ± 15
<i>Lactate (mmol/L)</i>	2.9 [1.5-8.3]	3.3 [2.0-8.3]	2.9[2.4-4.1]	2.7 [1.5-4.9]

Table 1. Continued...

	All n=32	P[5] n=11	P[10] n=10	I[5] n=11
Δ lactate (LAC2-LAC1)	-0.45 [-3.0-4.5]	0.2 [-3.0-4.5]	-0.7 [-2.5-0.2]	-0.6 [-2.3-1.6]
Lactate clearance ≥ 20%	13	2	5	6
Vasopressor requirement	4 (12%)	3 (27%)	1 (10%)	0 (0%)
Non-survivor until 14 days	3 (9%)	2 (18%)	0	1 (9%)

Table 2. Cardiorespiratory parameters of bitches with sepsis submitted to preoperative fluid therapy (5 or 10 mL/kg/h) and total intravenous (P) or inhalational (I) anesthesia for emergency ovariohysterectomy, period from March 2018 to November 2019. Veterinary Hospital of UFRRJ, Seropédica, Brazil.

		PRE	T10	T20	T30	T40	T50	EXT	DIS
HR (pm)	P[5]	125 ± 18	115 ± 30	118 ± 20	120 ± 23	116 ± 17	119 ± 16	120 ± 19	108 ± 19
	P[10]	125 ± 24	103 ± 22	112 ± 19	120 ± 20	117 ± 20	115 ± 15	125 ± 23	103 ± 31
	I[5]	119 ± 19	131 ± 21	115 ± 19	111 ± 23	118 ± 24	109 ± 13	119 ± 20	122 ± 24
RR (pm)	P[5]	43 ± 24	20 ± 15 ^{ab}	16 ± 10 ^{ab}	17 ± 10 ^{ab}	14 ± 7 ^{ab}	14 ± 5 ^{ab}	35 ± 18	41 ± 21
	P[10]	37 ± 14	17 ± 11 ^{ab}	16 ± 9 ^{ab}	16 ± 8 ^{ab}	17 ± 9 ^{ab}	17 ± 8 ^{ab}	41 ± 18	33 ± 18
	I[5]	50 ± 18	16 ± 8 ^{ab}	15 ± 4 ^{ab}	12 ± 3 ^{ab}	12 ± 3 ^{ab}	11 ± 5 ^{ab}	39 ± 23	51 ± 28
SBP (mmHg)	P[5]	126 ± 20	87 ± 23^{ab}	110 ± 30	104 ± 25 ^{ab}	102 ± 31 ^{ab}	98 ± 27 ^{ab}	108 ± 23 ^b	127 ± 20
	P[10]	132 ± 22	108 ± 18^{ab*}	106 ± 12 ^{ab}	127 ± 27	108 ± 15 ^{ab}	119 ± 24	127 ± 18	133 ± 17
	I[5]	132 ± 20	103 ± 16^{ab*}	110 ± 16 ^{ab}	110 ± 14 ^{ab}	122 ± 20 ^b	115 ± 20 ^b	132 ± 19*	139 ± 10
RT (°C)	P[5]	37.4 [36.8-39.2] ^b	37.3 [36.0-38.8] ^b	37.1 [36.3-38.4]	36.6 [35.9-38.3]	36.9 [35.5-38.2] ^a	37.3 [35.7-38.5]	36.6 [34.8-39.1]	36.7 [34.5-38.5] ^a
	P[10]	37.5 [36.2-38.1] ^b	37.3 [36.0-37.9]	36.7 [35.9-37.7]	36.6 [35.5-37.4]	36.6 [35.6-37.3]	36.5 [35.3-37.2] ^a	35.9 [35.1-37.0]	36.5 [35.6-37.7] ^a
	I[5]	38.6 [36.7-40.3] ^{b*}	38.2 [36.9-39.7] ^{b*}	38.1 [37.4-39.4] ^{b*}	37.7 [37.2-39.2] [*]	37.7 [37.0-39.0] ^{a*}	37.5 [36.8-38.7] ^{a*}	37.8 [36.8-39.0] ^{a*}	37.5 [36.9-38.2] ^{a*}
SPO ₂ (%)	P[5]	-	93 ± 6	97 ± 2	95 ± 3	96 ± 3	95 ± 3	-	-
	P[10]	-	96 ± 2	97 ± 2	97 ± 2	97 ± 2	97 ± 2	-	-
	I[5]	-	97 ± 2	97 ± 2	98 ± 1	98 ± 1	96 ± 2	-	-
EtCO ₂ (mmHg)	P[5]	-	40 ± 9	45 ± 11	45 ± 11	45 ± 8	43 ± 9	-	-
	P[10]	-	40 ± 9	44 ± 8	46 ± 12	44 ± 11	46 ± 12	-	-
	I[5]	-	35 ± 10	36 ± 11	36 ± 10	35 ± 8	35 ± 7	-	-

*Statistical differences concerning P[5]; ^aStatistical difference concerning PRE time; ^bStatistical difference concerning DIS time.

two bitches (18%) in P[5] (ΔLAC +0.2 [-3.0 to 4.5]), five bitches (50%) in P[10] (ΔLAC -0.7 [-2.5 to 0.2]), and six bitches (60%) in I[5] (ΔLAC -0.6 [-2.3 to 1.6]).

All anesthetic recoveries were considered satisfactory and the animals were considered stable for referral to an inpatient veterinary service. Three (9%) deaths were recorded, two from the P[5] group, which had four and five organ dysfunctions each (deaths within 24 h and 96 h, respectively); among them, only one presented with a vasopressor requirement during the study. The third death was recorded in group I[5], with four organ dysfunctions (death at 7 days) and no vasopressor requirement during the study.

Discussion

This study proposes standard protocols for bitches with pyometra and sepsis to aid in making therapeutic decisions for similar situations. The results showed that even though all three proposed

protocols are safe and with no difference in superiority, some observed changes may be relevant and considered when individualizing therapy. One of the biggest challenges in clinical studies is the standardization of sampling, which allows for comparisons between groups. However, in the present study, morbidity (assessed by the presence of organ dysfunction) did not differ significantly between groups. Due to the lack of a similar SOFA score (Sequential [Sepsis-related] Organ Failure Assessment - Vincent et al., 1996) for dogs, the identification of sepsis was based on the current understanding of its pathophysiology and clinical findings (Singer et al., 2016). Therefore, the bitches were considered septic in the presence of at least one organ dysfunction, supported by confirmed infectious focus through microbiological analyses of the intrauterine exudate.

Although some bitches admitted with initial systolic hypotension were randomly assigned to the P[5] group, in > 75% of these cases, normal arterial blood pressure was restored with fluid challenge. Patients with sepsis often present with arterial hypotension, which can be secondary to relative hypovolemia due to vasodilation or absolute hypovolemia due to inappetence, and gastrointestinal or renal loss (Montealegre & Lyons, 2021). Although initial fluid resuscitation protocol has reached a consensus among scientific studies and guidelines (Evans et al., 2021; Rhodes et al., 2015; Silversides et al., 2019), the dose and rate of fluids remain imprecise for both humans and dogs (Montealegre & Lyons, 2021). The proposed rate (10 mL/kg/15 min, up to two boluses) was well tolerated, safe, and effective in improving arterial blood pressure. This rate is comparable to that reported by Silverstein et al. (2005) and Conti-Patara et al. (2012). It is important to highlight that specific goals (normotensive arterial pressure) such as macrocirculatory parameters and sepsis-induced microcirculatory changes may persist (Montealegre & Lyons, 2021).

Most studies have focused on endothelial glycocalyx lesions and vasoplegia due to sepsis, which can refute fluid therapy benefits, especially at high rates (liberal protocols) (Self et al., 2018; Silversides et al., 2019). From this perspective, a hypertonic saline solution (HS) has been proposed as part of the fluid challenge protocol to avoid volume overload. HS promotes the translocation of extravascular fluids to the intravascular space (Smorenberg et al., 2013; van Haren, 2013) reducing the risk of hyperchloremic acidosis, impaired coagulation, and renal function (Smorenberg et al., 2013). Nevertheless, in two bitches which did not present with cardiovascular improvement after fluid challenge, early norepinephrine administration was established because of the severity of their clinical conditions. This approach is consistent with recent studies that associate the early use of vasopressors with increased survival in patients with septic shock (Permpikul et al., 2019; Self et al., 2018).

For the P[5] group, the significant increase in SBP values after two hours of fluid therapy (105 ± 28 vs. 126 ± 20 mmHg, $p=0.006$) can be associated with the admission status of these patients, whereas most of the bitches showed arterial hypotension but were responsive to fluid challenge. Therefore, this result demonstrates that these animals benefited from the preoperative fluid therapy protocol, surpassing the minimum postulated SBP value (SBP > 90 mmHg) and reaching values close to the physiological value for dogs (SBP \approx 120 mmHg). Although blood pressure monitoring alone may not accurately reflect the tissue perfusion status of patients (Evans et al., 2021; Skouropoulou et al., 2021), in low-resource settings such as veterinary services wards, this associated assessment with other parameters such as CRT can be applied (Davis et al., 2013; Evans et al., 2021; Noel-Morgan & Muir, 2018). Furthermore, it was possible to observe a negative impact of liberal fluid therapy on the body temperature of patients, which was significantly lower in the P[10] group. Although hypothermia in sepsis has been described as an adaptive strategy in the face of a systemic inflammatory response (L'Her et al., 2006; Rumbus et al., 2017), we believe that this may have been exacerbated by the administration of a large volume of fluid at room temperature, associated with tissue exposure inherent to the surgical procedure.

Anesthetic protocols suggested that isoflurane or propofol associated with fentanyl CRI was clinically and cardiorespiratory suitable for ovariohysterectomy in dogs. The stability of the anesthetic plan is directly related to nociceptive and sympathetic stimuli (Brown et al., 2015) and fentanyl, despite its excellent analgesic effect for this type of surgical procedure (Grubb et al., 2020), can promote bradycardia and decreased cardiac output in healthy patients (Lucas et al., 2016), which was not observed at the dose proposed in this study. Respiratory depression is also common when potent opioids, such as fentanyl, are used, as well as when central nervous system depression is induced by general anesthetics (Grubb et al., 2020). Thus, a significant reduction

in RR during anesthesia is justified. In addition, all animals were tachypneic at admission and pre-anesthetic clinical evaluation, possibly because of compensatory mechanisms of sepsis-induced metabolic acidosis (Conti-Patara et al., 2012). Regarding the safety assessment of the anesthetic protocol, we believe that these results are not clinically relevant since capnometry values remained within the normal range in all groups.

Concerning the intraoperative arterial blood pressure, it is possible to infer a direct effect of the preoperative fluid therapy protocol. SBP showed a significant decrease in the first 10 min of anesthesia with propofol CRI (P[5] and P[10]), which was more evident in the group that received fewer fluids (P[5]). In addition, only in P[5] did three bitches have episodes of intraoperative hypotension and vasopressor requirement, which stopped a few minutes after the propofol CRI was discontinued. In healthy patients, both propofol and isoflurane decreased the circulating volume by increasing venous capacitance, without producing significant changes in cardiac output and systemic vascular resistance (Yamazaki et al., 1998). This vasodilation caused by general anesthetics determines relative hypovolemia, which can be very dangerous in septic patients, potentiating microcirculatory and cardiac dysfunction (Noel-Morgan & Muir, 2018). The fluid therapy rate of 10 mL/kg/h seems to have minimized these effects of propofol because in the anesthetic recovery time (EXT), the isoflurane group (I[5]) showed significantly higher SBP values than the P[5] group. Thus, this more liberal fluid protocol may have been able to increase the volume of stressed circulating blood (Noel-Morgan & Muir, 2018) and, consequently, improve SBP stability.

Hyperlactatemia was observed in 84% of bitches at admission, contributing to the understanding, already mentioned in previous paragraphs, that static parameters of macrocirculation do not necessarily reflect the micro-hemodynamic condition and tissue perfusion of the patient (Skouropoulou et al., 2021). Despite all approaches of fluid resuscitation and vasopressor administration, some patients showed an increase in plasma lactate concentration after treatment. This fact is more evident in the group that received the lowest rate of fluid therapy (P[5]), where only 18% of patients reached lactate clearance of more than 20% during the time comprised by the service (approximately 4 h) (Blutinger et al., 2021; Nguyen et al., 2010; Zollo et al., 2019). Negative performance may be associated with insufficient fluid therapy, lower blood pressure stability during the perioperative period, or the direct influence of the two bitches that died, as it has a positive correlation with sepsis mortality (Blutinger et al., 2021; Conti-Patara et al., 2012; Nguyen et al., 2010).

Although clinical studies can provide valuable information as they more accurately reflect the clinical practice scenario, there are several challenges in their execution, some of which should be highlighted. The heterogeneity of the clinical conditions and the reduced number of animals per group may have compromised some statistical interpretations. Additionally, the duration of morbidity and subclinical conditions could not be evaluated. Therefore, we strongly recommend that, based on the assessment of the results discussed here, veterinarians should carefully evaluate their patients and establish individualized procedures for stabilization and management of sepsis in bitches with pyometra.

Conclusion

In conclusion, liberal fluid therapy associated with total intravenous anesthesia with propofol resulted in greater cardiovascular stability and lactate clearance. In patients undergoing restrictive fluid therapy in the preoperative period, isoflurane resulted in better cardiorespiratory stability than propofol for anesthetic maintenance in dogs with sepsis due to pyometra and undergoing an emergency surgical procedure.

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Ethics statement

The study was accomplished after approval by Animal Use Ethics Committee of the Veterinary Institute of the Federal Rural University of Rio de Janeiro with protocol number 3269190418.

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Conflict of interests

All authors declare no conflict of interest.

Authors' contributions

VCTL - Conceptualization, Aquisition Data, Methodology, Formal Analysis, Project Administration, Investigation, Writing, Review & Editing. AJRP - Aquisition Data, Methodology, Formal Analysis, Project Administration, Investigation, Review, Editing & Critical Revision of the Manuscript. MESLF, LCO, ACSC, AFXO and NVS - Methodology, Aquisition Data & Critical Revision of the Manuscript. FFPCB and CDB - Methodology, Formal Analysis, Investigation & Critical Revision of the Manuscript. CMMC - Conceptualization, Aquisition Data, Methodology, Formal Analysis, Project Administration, Investigation & Critical Revision of the Manuscript.

Availability of complementary results

Online repositories: PUBMED: <https://pubmed.ncbi.nlm.nih.gov> SciELO: <https://scielo.org> https://wp.scielo.org/wp-content/uploads/Lista-de-Repositorios-Recomendados_pt.pdf

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