

EFFECTS OF BLOOD STORAGE IN GASOMETRIC ANALYSIS IN ARTERIAL BLOOD OF RABBITS¹

EFEITO DA ESTOCAGEM SANGÜÍNEA NA ANÁLISE GASOMÉTRICA DE SANGUE ARTERIAL DE COELHOS

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Objective: assess the effect of blood storage in gasometric values of arterial blood samples of rabbits submitted to controlled hemorrhagic shock. **Method:** fourteen male California rabbits were used for the present study, weighting 2000-2500 grams. Each animal was submitted to catheterization of right carotid artery, with posterior placement of a polyethylene catheter. It was obtained 1 ml of arterial blood from each rabbit and analyzed in three different periods: T0 – immediately; T30 – 30 minutes after withdrawal; T60 – 60 minutes after withdrawal. The samples were kept in thermal insulating recipient, between 2-6°C. Values of pH, PaCO₂, PaO₂, SatO₂, HCO₃, SBE, sodium, potassium and glyceimic concentrations were compared. **Results:** the variation of obtained means related to pH, PaCO₂, SatO₂, HCO₃, SBE, sodium, potassium and glyceimic concentrations didn't have statistically significant difference (p<0,05), demonstrating that the storage was efficient for these parameters, but there was variation in PaO₂ when compared T0 to T60 (p=0,04). **Conclusion:** storage for 30 and 60 minutes didn't present statistically significant difference, except for PaO₂ after 60 minutes.

Keywords: Blood storage, gasometric analysis, rabbits.

INTRODUCTION

At clinical practice, the arterial gasometry is the main laboratorial exam to be chosen to evaluate the alterations of pulmonary gas exchanges of patients with cardiopulmonary dysfunction because they represent the best indicator for diagnosis of

acid-base imbalance and allows the appropriate and immediate indication of reposition therapy, reaching the clinical control of the patient.^{1,2}

However, the occurrence of important errors during collection, conservation and blood analysis may cause damages to the results, affecting the success of the established treatment.¹

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Many factors have been cited as capable to influence negatively on gasometric values, such as: the storage temperature, storage time and the syringe material which is, in general, plastic,³ being the storage time the most important factor to influence on the gas analysis, according to the literature, but the conclusions of several researches are conflicting, including the experimental studies.⁴ However, there is already the recognition of its influence.²

To assess the effect of blood storage period in the gasometric values of the arterial blood sample of rabbits submitted to controlled hemorrhagic shock, using a clinically-relevant experimental model for catheterization of right carotid artery and jugular vein.

METHODS:

This study was performed at the Multidisciplinary Unit of Experimental Medicine, Surgery Department, Federal University of Pará (UFPA) - Brazil, and done in accordance with guidelines established by the Institutional Animal Care and Use Committee at the State University of Pará. Fourteen male *California* rabbits (2500-3000g), all proceeded from Zootecny Center of Rural Federal University of Amazon (UFRA), were used in all experiments, which were kept in, adjusted conditions of light and temperature, receiving standard diet *ad libitum*, except for 6 h before surgery when there was access to water only.

All the animals were submitted to anesthetic and monitorization procedures previously to the surgery that was made for the same team, during the entire study, and were submitted to surgery in the same period of the day, avoiding any influence that the difference of the period could bring to the results. Rabbits were anesthetized with an intramuscular solution of ketamine (80mg/kg), xylazine (10mg/kg) and atropine (0.05 mg/kg). Oxygen was managed in the concentration of 2 L/min, using a plastic mask that was adapted to the

face of the animal, protecting it partially. The characterization of the anesthetic plan was evaluated using parameters, such as: mustache and ears movements after stimulation (sedation); absence of tail and legs' retraction after digital pressure (surgical anesthesia). Our preliminary studies had determined these procedures settings to maintain physiological blood gas concentration prior to the onset of hemorrhage.

After anesthetic induction, it was made depilation of the neck, followed by apprehension of members in extension with plastic wires. Using aseptic technique, the right carotid artery was then dissected with placement of a polyethylene catheter (4mm gauge) for withdrawal of an arterial blood sample. Following a 5 min stabilization period, baseline (BL) measurements of mean arterial pressure (MAP), heart rate (HR), respiratory rate (RR) and rectal temperature (RT) were recorded, and, with utilization of a plastic syringe (Becton Dickinson - BD), 1 ml of blood sample was collected for analysis of the gasometric parameters, electrolytes (Na⁺, K⁺), glycemia, which were performed in three moments: immediately after the withdrawal, after a period of 30 min (T30) and 60 min (T60). The samples T30 and T60 were stored in a thermal insulating recipient with temperature between 2-6 °C and the analysis were performed in Rapidlab 348 blood gas analyzer.

Comparison of parameters between each of the three different periods of the experiment was performed using one-way analysis of variance (ANOVA) and Tukey test. Differences were considered significant at $p < 0.05$. All results are expressed as mean \pm standard error of mean (SEM).

RESULTS

The obtained results are on the table 1, presented as mean \pm standard error of mean, with their respective p-values:

Table 1: Gasometric values in the three periods of analysis.

METABOLIC PARAMETERS	T0	T30	T60	P-value
Arterial pH	7.387 ± 0.07	7.367 ± 0.06	7.344 ± 0.06	0.3695
PaCO ₂ (mmHg)	46.9 ± 3.34	49.0 ± 2.97	48.9 ± 3.20	0.9153
PaO ₂ (mmHg)	394.3 ± 14.99	350.6 ± 18.34	303.9 ± 18.20	0.0400
SatO ₂	98.07 ± 0.04	98.00 ± 0.75	94.63 ± 0.01	0.5599
HCO ₃ (mmol/L)	20.61 ± 2,60	21.5 ± 1,70	25.42 ± 1,64	0,5640
Base deficit (mmol/L)	1.7 ± 1.21	2.6 ± 0.51	1.5 ± 0.84	0.7641
Na ⁺ (mEq/L)	138.181 ± 1.00	138 ± 0.50	138.222 ± 0.51	0.9828
K ⁺ (mEq/L)	3.39 ± 0.19	3.64 ± 0.08	3.72 ± 0.14	0.3728
Glycemia	242.11 ± 14.13	251.50 ± 11.67	243.00 ± 11.75	0.9029

T0: immediate analysis after withdrawal; T30: analysis after 30 minutes; T60: analysis after 60 minutes.

It was observed significant difference only related to PO₂ when T0 was compared to T60 (p = 0,04).

DISCUSSION

On the literature, several studies about blood storage for gasometric analysis searched for appropriate methods in order to evaluate the effects of time storage, but there is still controversy among them. According to the National Committee for Clinical Laboratory Standarts⁵, the samples collected in plastic syringes shall not pass 30 minutes to be analyzed, being kept in room temperature because it is believed that, when conserved between 0-4°C in this type of syringes, occurs the contraction of plastic molecules, opening larger pores for oxygen to diffuse through, but not large enough for the larger carbon dioxide molecule.⁶

On the other hand, the cooling of the samples, using ice, is considered a good process in order to induce the retardation of cellular metabolism because it causes minor oxygen consumption and carbon dioxide production by leukocytes and erythrocytes, but it is not perfect.^{1,6}

Thus, as it was observed in the present study, there was no alteration in the pH values of samples when T0, T30 and T60 were

compared. Knowles et al⁶ concluded that there was no difference in T30, while Deane *et al*⁴ and Beaulieu *et al*⁷ didn't find pH alteration in T60.

According to Knowles et al⁶, the gasometric analysis of arterial blood samples in plastic syringes evidenced significant increase in PO₂ when compared the analyzed samples in T0 to T30, not presenting differences between the temperature conservation (0-4°C or 22°C). Beaulieu et al⁷ and Mahoney et al⁸ also found increase in PO₂ in iced samples that were analyzed after 30 minutes, presenting statistical difference. Deane et al⁴ evidenced PO₂ elevation in samples after 60 minutes, suggesting that the oxygen diffusion through the plastic material is the main factor of this alteration.⁷

Smeenck et al³ found significant PO₂ reductions in T30 and T60. This last finding is according to the present study.

The present research didn't show PCO₂ alteration in any period (T0, T30 or T60), according to the literature.^{4,6,7} The explanation is that carbon dioxide is four times more soluble than oxygen in polypropylene syringes and, being iced, its solubility raise, limiting the efflux of carbon dioxide.⁸

In relation to oxygen saturation, the present study didn't demonstrate changes with statistical significance, according to Yen et al.²

SBE, HCO₃, sodium, potassium and glycemic concentration values didn't show alterations with significant difference. However, their analyses were not evaluated in any other research in the literature. Therefore, the authors of the present study suggest that other studies should be done in order to determine the effect of time conservation on their values.

CONCLUSION

In summary, the results indicate that the samples in plastic syringes, between 2-6°C, must be analyzed in the first 30 minutes after the withdrawal without having damages in the gasometric parameters, including PO₂.

SUMÁRIO

EFEITO DA ESTOCAGEM SANGÜÍNEA NA ANÁLISE GASOMÉTRICA DE SANGUE ARTERIAL DE COELHOS

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Objetivo: analisar o efeito da estocagem sanguínea na análise gasométrica de sangue arterial de coelhos. **Método:** foram utilizados 14 coelhos California adultos, machos, com peso compreendido entre 2000 a 2500 gramas. Cada animal foi submetido à cateterização da artéria carótida para posterior obtenção de amostra sanguínea. Foi colhido, em seringas plásticas, 1 ml de sangue total de cada coelho e dosado em três tempos distintos: T0 – amostra no tempo considerado zero; T30 – amostra no tempo 30 minutos; T60 – amostra no tempo 60 minutos. As amostras foram estocadas em recipiente isolante térmico, com temperatura mantida entre 2-6 °C. Foram comparados os valores de pH, PaCO₂, PaO₂, HCO₃, SatO₂, SBE, concentrações de Na⁺ e K⁺ e glicemia. **Resultados:** a variação das médias obtidas em relação ao pH, PaCO₂, SatO₂, HCO₃, SBE, concentrações de Na⁺ e K⁺ e glicemia não alcançou significância estatística (p>0,05), demonstrando que a estocagem foi eficaz para esses parâmetros, no entanto na PaO₂ houve variação do grupo T60 para o grupo T0, com p = 0.04. **Conclusão:** a estocagem por 30 e 60 minutos não apresentou diferença estatisticamente significativa, exceto a PaO₂ após 60 minutos.

Palavras-chave: estocagem, sanguínea, análise gasométrica, coelhos.

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