DIFFERENT RECUMBENCIES FOR GESTATIONAL ULTRASOUND IN RABBITS: FIRST RESULTS

Kassy Gomes da SILVA*[1], Angelo SIGNORI[1], Ubirajara Iobe TASQUETI[1], Leandro Batista COSTA[1], Cristina Santos SOTOMAIOR[1]

*corresponding author: kgomes13@hotmail.com
[1]Pontifical Catholic University of Paraná, Curitiba, Paraná, Brazil

Resumo: O decúbito dorsal é o mais utilizado para ultrassonografia gestacional de coelhas. A realização do exame com a coelha em estação pode diminuir o estresse da contenção. O objetivo foi comparar o decúbito dorsal (DR), lateral direito (RLR), lateral esquerdo (LLR) e esternal (SR) no exame ultrassonográfico gestacional em coelhas. Cada decúbito foi utilizado aos 7, 14, 21 e 28 dias pós-cobertura em 18 coelhas. Avaliou-se o diagnóstico gestacional (sensibilidade, especificidade, valor preditivo positivo- VPP, valor preditivo negativo- VPN e acurácia), a contagem de vesículas embrionárias/fetos e o tempo de exame para cada decúbito. No dia 7, o DR apresentou maior sensibilidade (81,3%). Nos dias 14, 21 e 28, a sensibilidade e especificidade foram de 100%, sem diferença entre os decúbitos. O número de láparos foi subestimado nos DR, RLR e LLR, e superestimado no SR, com o maior percentual de acerto (38%) no DR. O SR proporcionou maior tempo de exame aos 7 dias, porém sem diferença entre os decúbitos nos demais dias. O decúbito esternal pode ser utilizado como alternativa aos decúbitos dorsal e lateral para diagnóstico de gestação em coelhas a partir dos 14 dias pós-cobertura. O uso da ultrassonografia para contagem de láparos não foi eficiente em coelhas.

Keywords: accuracy, diagnostic, *Oryctolagus cuniculus*, sensitivity, specificity

Introduction

Diagnostic ultrasonography uses high frequency sound waves targeted to the region of interest, whose returning echoes are analyzed by the computer to produce

high resolution transverse images of organs, tissues and blood flow (Mattoon and Nyland, 2015). In veterinary medicine, ultrasonography has been widely used to diagnose pregnancy in various species, such as bitches and cats (Mattoon and Nyland, 2015). In rabbits, the application of ultrasound in reproduction, especially in females, has been described in recent years (Gutierrez and Zamora, 2004; Idris et al., 2016).

The gestational evaluation of small animals, such as dogs and cats, is done by placing the transducer in ventral medial line, both in the lateral and dorsal decubitus, but the latter is the most convenient (Mattoon and Nyland, 2015). This is also the decubitus commonly used for this evaluation in rabbits (Ypsilantis and Saratsis, 1999; Gutierrez and Zamora, 2004; Ajadi et al., 2015; Idris et al., 2016).

Thus, currently, the dorsal recumbency is the standard position for gestational ultrasonography of rabbits. However, an auxiliary is required to keep the rabbit in the dorsal recumbency throughout the assessment. Even if the instructions for restrainement are easy, there should always be a person trained to perform such a task, to avoid injury both to the helper and the examiner, and especially to avoid injuring the animal. This also applies to the use of lateral recumbency.

One of the reasons for the use of both recumbencies (dorsal and lateral) is the ease in locating the embryonic structures and their evaluation, since the transducer scan area is larger (Ypsilantis and Saratsis, 1999; Idris et al., 2016). In addition, examination of the entire abdominal cavity can also be done more quickly and efficiently.

One option is to perform the sternal recumbent examination, which would be the most natural of the recumbencies for the rabbit, besides the fact that the lateral access, allowed by this recumbency, may be enough for the evaluation of a pregnant uterus. However, no studies were found that describe the use of this technique for gestational evaluation in rabbits.

The aim of this study was to compare the gestational ultrasonographic examination of rabbits in the dorsal, lateral (right and left) and sternal recumbencies, observing whether the sternal recumbency can be included as a position for this type of evaluation.

Material e methods

The study was approved by the Ethics Committee on Animal Use of Pontifical Catholic University of Paraná (PUCPR) under number 0937. The experiment was carried out in the rabbitry of the Gralha Azul Experimental Farm, located in Fazenda Rio Grande, Paraná, from November/ 2017 to March/2018.

Eighteen adult New Zealand white rabbits does were used, 13 pregnant and 5 non-pregnant, housed individually in suspended wire cages, with automatic drinking and manual feeder. Feed, hay and water were supplied *ad libitum* for all rabbits. Natural mating was performed in day 0 of experiment.

Each female was evaluated on days 7, 14, 21 and 28 post-mating, in the dorsal (DR), right lateral (RLR), left lateral (LLR) and sternal (SR) recumbencies. The preparation was: for DR, the animal remained with the dorsal region in contact with the padded foam and restrained by an auxiliary. Before the examination, the ventral abdominal region was clipped; for RLR and LLR, the animal remained with the lateral (left / right) of the body in contact with the padded foam and restrained by an auxiliary. The same DR trichotomy was used; for SR, the animal remained on the padded foam without manual restraint, in natural station position. Before the examination, the abdomen was clipped on both sides, which was initiated from the last rib, following caudally to the coxal tuberosity, parallel to the lumbar spine. When necessary, an auxiliary restrained the animal by supporting the dorsal region over the shoulder blades toward the padded foam.

The acoustic gel was applied to the abdominal area immediately preceding the beginning of the examination. All examinations were performed with a

microconvex (4-8.5 MHz) transducer (Ecovet3, Chison, China). Ultrasound examination and evaluation of the images obtained were performed by a single ultrasonography technician.

The data evaluated were: pregnancy diagnosis (positive or negative), vesicles / fetuses count (days 14, 21 and 28) and time of examination. The diagnosis of pregnancy was considered positive when it was possible to observe the uterus (day 7), the embryonic vesicles (day 14) or fetuses (days 21 and 28). For counting, each embryonic vesicle or fetus was considered as an individual and the total number was compared to the number of births born.

For the diagnosis of pregnancy, sensitivity, specificity, positive predictive value, negative predictive value and accuracy (correct classification) were calculated according to Kawamura (2002). The Fisher exact test was used for the number of embryonic vesicles / fetuses. For the examination time, the data were submitted to analysis of variance (ANOVA), the means being compared by the Tukey test (days 14, 21 and 28) and the Bonferroni test (day 7), considering the value of significance of 5% for all tests. The software used was Statgraphics® Centurion XVII, version 17.2.05.

Results and discussion

On the day 7, the sensitivity varied from 73.3% to 81.3%, with the DR presenting the highest value (Table 1). The specificity was 33.3% for RLR and SR, and 100% for DR and LLR. The PPV was 100% for DR and LLR, and 84.6% and 69.2% for RLR and SR, respectively. The dorsal recumbency presented the best accuracy (83.3%) on day 7 among the evaluated recumbencies. Inaba et al. (1986) obtained an accuracy of 64% with the use of dorsal recumbency at 7 days, with similar values in the sternal (61.1%) and right lateral (66.7%) of the current study. The percentages of sensitivity, specificity, PPV, NPV and accuracy were the same among the recumbencies on days 14, 21 and 28 (Table 1), so that the choice of

recumbency to be used during ultrasonographic examination is indifferent from day 14 to the parameter of pregnancy diagnosis.

Table 1. Sensitivity (S), specificity (E), positive predictive value (PPV), negative predictive value (NPV) and accuracy between different recumbency and different times for gestational evaluation in rabbits

Day 7	Recumbency	S (%)	E (%)	PPV (%)	NPV (%)	Accuracy[e] (%)
(n=18)	DR[a]	81,3	100,0	100,0	40,0	83,3
	RLR[b]	73,3	33,3	84,6	20,0	66,7
	LLR[c]	76,5	100,0	100,0	20,0	77,8
	SR[d]	75,0	33,3	69,2	40,0	61,1
Days 14, 21 e 28 (n=18)	DR	100,0	100,0	100,0	100,0	100,0
	RLR	100,0	100,0	100,0	100,0	100,0
	LLR	100,0	100,0	100,0	100,0	100,0
	SR	100,0	100,0	100,0	100,0	100,0

[a] dorsal recumbency; [b] right lateral recumbency; [c] left lateral recumbency; [d] sternal recumbency; [e] (true positive+ true negative)/n

The mean number of kits born was 6.6 ± 2.6 kits. The correct rate for counting the number of embryonic vesicles on day 14 was greater ($p < 0.05$) in DR (38%), in LLR (23%) and in SR (15%) than in RLR, in which there was no success (0%). There was no difference ($p > 0.05$) between the recumbencies on days 21 and 28.

In general, the number of kits was underestimated in DR, RLR and LLR, and overestimated in SR. The number of kits was underestimated with a mean of -1.6 kits on day 28 in 53.8% in DR (Table 2). In the sternal recumbency, there was an overestimate, an average, of +1.6 kits in 69.2% of the exams on day 14. On day 28, the mean of underestimated kits was -2.7 kits in 46.2% of the exams in this recumbency.

Table 2. Percentage of pregnancies with underestimate (%Under), overestimated (%Over), number of underestimated kits (NUnder) and number of overestimated kits (NOver) between different positions used during gestational ultrasonography in rabbits (n = 13)

Position		%Under	%Over	NUnder	NOver
DR[a]	Day 14	46,2	15,4	-1,8	+4,0
	Day 21	38,5	38,5	-2,3	+2,5
	Day 28	53,8	23,1	-1,6	+2,0
RLR[b]	Day 14	76,9	23,1	-1,5	+1,3
	Day 21	30,8	15,4	-2,3	+2,5
	Day 28	53,8	38,5	-2,0	+1,6
LLR[c]	Day 14	53,8	23,1	-1,9	+2,3
	Day 21	46,2	23,1	-2,0	+1,3
	Day 28	46,2	23,1	-2,3	+2,3
SR[d]	Day 14	15,4	69,2	-2,5	+1,6
	Day 21	38,5	46,2	-2,0	+1,3
	Day 28	46,2	38,5	-2,7	+1,4

[a] dorsal recumbency; [b] right lateral recumbency; [c] left lateral recumbency; [d] sternal recumbency;

The mean time to the 7-day examination was greater ($p < 0.05$) in the SR (138 ± 77s) than in the DR (50 ± 34s), RLR (60 ± 32s) and LLR (42 ± 17s). The mean of the other days evaluated were: day 14 (54 ± 17s), day 21 (41 ± 14s) and day 28 (48 ± 14s), with no difference ($p > 0.05$) between the positions evaluated at these times.

Conclusion

For pregnancy diagnosis in rabbits at 7 days post-mating, dorsal recumbency was the best position for ultrasound examination. The sternal recumbency can be used as an alternative to dorsal and lateral positions for diagnosis of pregnancy in rabbits from 14 days post-mating. The use of ultrasonography for the counting of kits was not efficient in the studied positions.

Acknowledgments

We thank the Pontifical Catholic University of Paraná for the PIBIC scholarship.

References

Gutierrez, H.E.; Zamora, F.M.M. 2004. Ultrasonography study of rabbits pregnancy. Pg. 276-280. In: Proceedings of the 8th World Rabbit Congress, México.

Idris, S.Y.; Audu, H.A.; Lawal, M.; Ibinaiye, P.O.; Fadason, S.T.; Muazu, B.N.; Echekwu, O.W. 2016. Sonographic diagnosis of pregnancy and study of gestational changes in rabbit does. Nigerian Veterinary Journal 37(3): 133-139.

Inaba, T.; Mori, J.; Torii, R. 1986. Use of echography in rabbits for pregnancy diagnosis. Nihon Juigaku Zasshi 48(5): 1003-1006.

Kawamura, T. 2002. Interpretação de um teste sob visão epidemiológica: Eficiência de um teste. Arquivos Brasileiros de Cardiologia 79(4): 437-441.

Mattoon, J.S. e Nyland, T.G. 2015. Small animal diagnostic ultrasound. 3rd ed. Elsevier, Missouri.

Ypsilantis, P.; Saratis, P. 1999. Early pregnancy diagnosis in the rabbit real time ultrasonography. World Rabbit Science 7: 95-99.