

Potential of mammary secretions during pseudopregnancy as colostrum substitutes

M. Abrard, P. Ronsin, H. Mila, S. Chastant-Maillard.

NeoCare, Ecole Nationale Vétérinaire de Toulouse, UMR 1225 INRA/ENVET, 23 chemin des Capelles, 31076 Toulouse Cedex 3, France

s.chastant@envt.fr

Despite transfer of maternal antibodies through colostrum is known to be crucial for puppies survival, no substitute for canine colostrum, efficiently providing homospecific immunoglobulins is currently available [1]. Canine plasma or serum only allow a limited transfer [2]. Mammary secretions produced at the end of diestrus during overt pseudopregnancy potentially represent a source of canine immunoglobulins. The objective of this work was to determine concentrations in immunoglobulins G [IgG] and A [IgA] of mammary secretions produced during pseudopregnancy and to compare with [IgG] and [IgA] of canine colostrum and milk. Mammary secretions were collected by manual milking in 31 bitches with overt pseudopregnancy from secreting mammary glands (one sample per bitch). For five bitches, samples were additionally available for several mammary glands. Were registered on bitches their age (young: below 2 yrs; adult: 2-6 yrs; senior: more than 6 yrs) and their breed size (small: less than 15 kg; medium: 15-25 kg; large: more than 25 kg). The aspect of secretions was qualified as colostrum when opaque and orange yellowish, milky when aqueous and white; or hemorrhagic. In parallel, 9 colostrum samples collected 24 hours after whelping and 9 milk samples collected 14 days after parturition (on 9 different bitches) were used as references. IgG and IgA concentrations were determined in pseudopregnancy, colostrum and milk samples thanks to an ELISA assay specific from canine IgG and IgA (Dog IgG or IgA ELISA Quantitation set; Bethyl Lab, Montgomery, USA). Intra and inter assay coefficients of variation were below 6% for both assays. The influence of age, bitch breed size together with that of aspect and nature of mammary secretions (pseudopregnancy/colostrum/milk) on [IgG] and [IgA] was evaluated thanks to ANOVA. Results are expressed as mean \pm SD. Results. In total, 42 pseudopregnancy secretions have been collected. [IgA] was 11.6 ± 9.9 g/L (min 1.6 ; max 50.3) and [IgG] 24.2 ± 15.5 g/L (min 1.9 ; max 62.7). Correlation between [IgA] and [IgG] was moderate ($r=0.48$, $p=0.015$). [IgA] was significantly influenced by breed size (higher in large bitches) and by the age of the bitch (higher in senior females), but not by the aspect of the secretions. [IgG] was neither influenced by the breed size, the age, nor by the aspect of the secretions. Despite no statistical analysis was possible, a large variability between [IgG] in the secretions from the different mammary glands of a given female was observed (variation factor: 1.6 to 8.8).

[IgA] did not differ significantly between the three types of secretions (10.5 ± 3.5 g/L for colostrums, 7.6 ± 4.2 g/L in milks, $p>0.05$). [IgG] was neither significantly different between pseudopregnancy secretions and colostrums ([IgG] in colostrums: 18.0 ± 12.0 g/L, $p>0.05$), but significantly higher than that in milks (2.0 ± 1.3 g/L, $p<0.0001$). IgG represented $67.8 \pm 16.7\%$ of the total [IgG]+[IgA] pseudopregnancy secretions, vs $46.9 \pm 19.7\%$ in colostrums and $22.0 \pm 14.0\%$ for milks.

This study evidenced that pseudopregnancy secretions were similar to colostrums considering IgG and A concentrations. Comparison with literature indicates that they are close to a colostrum collected 24 hours after whelping [3]. Their high [IgG] makes them potential colostrum substitutes in canine, at least for the immune aspect. Their energetic value remains to be quantified and their efficiency in terms of passive immune transfer would deserve to be evaluated, together with the persistence of [IgG] in pseudopregnancy secretions along with repeated milkings.

[1] Mila et al, 2016 *Reprod Dom Anim* 51 (Suppl. 3): 1-7. [2] Poffenbarger et al, 1991 *Am J Vet Res*;52(8):1221-4. [3] Albaret et al 2016. 8th ISCFR, Paris, France, pp. 23.

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