ABSTRACTS

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Hypoluteoidism in the bitch - Investigation of endocrine and immunological parameters

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OBJECTIVES AND METHODS: Embryonic as well as fetal loss, abortion and shortening of the estrous cycle length may be associated with insufficient secretion of progesterone by the canine corpora lutea. The aim of the study was to investigate the concentration of progesterone and prolactin in suspected hypoluteoid (shortening of the estrous cycle length, failure of the previous pregnancies) and control bitches during pregnancy and the nonpregnant cycle. Moreover, canine antibodies against progesterone were analysed due to a possible connection between embryonic loss associated with hormonal changes related to a hormone antibody response. Twenty-seven German Shepherd dogs as well as Rottweiler dogs (n=4), Bernese mountain dogs (n=2), Airedale Terrier (n=2), Rhodesian Ridgeback dogs (n=2) und one Border Collie were included in the study. All bitches were privately owned and the owners agreed to the use of the serum and data of reproduction. Serum samples (5 times between days 12 and 40) from 20 suspected hypoluteoid bitches and 18 control subjects were examined. Both groups were assigned to pregnant and nonpregnant animals by ultrasound examination (between days 21-25 p. ov. with Logiq 5, General Electric Company). Progesterone was measured with an Enzyme-linked fluorescent assay (miniVIDAS®, bioMerieux, France) and prolactin with an Enzyme-linked immuno assay (Demeditec Diagnostics GmbH, Germany). Relaxin concentrations were determined by a modified Enzymeimmunoassay (EIA) system as described by Einspanier et al. An ELISA detecting anti-progesterone-antibodies was developed at the Institute of Veterinary-Biochemistry, Freie Universität Berlin. The assay development included the testing of different horseradish peroxidase-conjugated antibodies (IgG, IgG, IgM) as well as the determination of their optimal dilution. In this context three different types of microtiter plates, two coating-buffer solutions, five blocking solutions and three different sources of substrate were compared with each other.

RESULTS: The results show that the nonpregnant, suspected hypoluteoid bitches had significantly lower progesterone concentrations than the control animals. It was inescapable to substitute the pregnant suspected hypoluteoid bitches with exogenic progesterone (Utrogestan®, 5mg/kg bw t.i.d.) therefore the measured values are affected by oral supplementation. Nevertheless mean progesterone concentrations of the suspected hypoluteoid pregnant bitches were lower than the control group by trend. Prolactin concentrations were significantly lower in German Shepherd Dogs compared with all dogs of other breeds in all groups. They also showed significantly lower concentrations in the pregnant control group compared with the suspected hypoluteoid group. Relaxin concentrations were similar in both pregnant groups but significantly higher from day 29 p. ov. compared with the nonpregnant groups.

IgG, IgM and IgE antibodies against progesterone were found in the serum of bitches. To examine whether antibodies to progesterone are specific or cross-reactive to other steroids, a competition ELISA by adding progesterone, estradiol, testosterone and hydrocortisone in the liquid phase was performed. Results showed that only the addition of progesterone adsorbed the IgM and IgE antibodies most intensely. Preliminary results indicate constant levels of antibodies in the individual bitches, irrespective of the point of measurement.

CONCLUSION: The results demonstrate that a frequent measurement of progesterone is necessary to determine the onset of a persistent progesterone decline also in the early luteal phase. In contrast to former investigations (2) prolactin concentrations were significant higher in the pregnant suspected hypoluteoid bitches matched with the control group. This rise could be a reaction of the progesterone withdrawal as a consequence of the pituitary regulation. In addition to that hyperprolactinaemia has been causally related with luteal phase defects and recurrent spontaneous abortion at the woman (3). Antibodies against progesterone and estrogen were found in the sera of women with autoimmune progesterone dermatitis. This appears as a rare cyclic reaction to progesterone producing during the luteal phase of woman’s menstrual cycle with a variety of presentations including asthma, erythema multiforme and infertility (4). It is discussed that progesterone and their metabolites may act as antigens after binding to blood proteins such as albumin or globulin and promote type 2 helper cell development, thereby regulating antibody synthesis and allergy (5). Our results demonstrated the evidence of antibodies to progesterone in the canine serum too. Longterm investigations to determine a possible connection between the decline of progesterone in suspected hypoluteoid bitches associated with high levels of IgG, IgM and IgE are still in progress.


