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Brief Communication

Clinical and Blood Biochemical Changes after Administration of Low Doses of Endotoxin in Ewes

Injection of high doses of endotoxins from *Salmonella typhimurium* have been reported to induce endotoxaemia in domestic animals similar to what is seen in cases of sepsis with Gram-negative bacteria (*Fredriksson* 1984, *Aiumlamai et al.* 1990, *Aiumlamai & Kindahl* 1990). A picture of clinical and blood biochemical changes in many disease syndromes such as ruminal acidosis, ruminal stasis and parturient paresis showed similar patterns as during induction of endotoxaemia with high doses of endotoxin, but less pronounced (*Aiumlamai & Kindahl* (in press), *Aiumlamai et al.* (in press a, b)). The resorption of small amount of endotoxin from the gastrointestinal tract during these diseases was detected and discussed to be the cause of or complicate these disease syndromes. The aim of this study was to describe the changes in the ewe after exposure to different low doses of endotoxin.

Eight healthy Swedish peltsheep ewes aged around 2 years, weighing 55–67 kg, were randomly assigned to 2 consecutive treatment groups. Four weeks elapsed before the ewes were used in the next experiment. An endotoxin from *Salmonella typhimurium* was administered intravenously in doses of 100, 50, 25, 10, 5 ng/kg; each dose was repeated in 2 animals. Four ewes were administered with 20 ml of normal saline. The source of endotoxin and preparation procedure was the same as described before (*Fredriksson* 1984). Blood was sampled at –30, –20, –10 min before injection of endotoxin or saline and at 2, 5, 10, 20, 30, 40, 50, 60, 90, 120, 150, 180 min and then every h for 3 more h. The collections were done in heparinized and plain vacutainer tubes. Rectal body temperature (BT) was measured at –30 and –20 min before an injection and every 30 min during 3 h and then every h for 3 more h. Whole blood was collected in EDTA vacutainer tubes for blood morphology at the rime of BT measurement.

Plasma was analysed for the content of 15-ketodihydro-$PGF_{2\alpha}$ (PG) (*Granström & Kindahl* 1982). Whole blood was analysed for total white blood cell number (WBC) and differential count simplified by dividing the cells in polymorphonuclear (PMN) and mononuclear (MN). Serum was analysed for the concentration of calcium (Ca), zinc (Zn) and iron (Fe) according to standard methods validated at the Clinical Chemistry Laboratory, Faculty of Veterinary Medicine, Swedish University of Agricultural Sciences. Serum Ca was not analysed in ewes treated with 5 or 10 ng endotoxin/kg.

The changes in all parameters were evaluated by averaging the data of the samples before treatment in individual animal and compare to the levels after treatment. The difference between the mean of pretreatment values and the treatment values was considered different if the difference was greater than plus or minus 3 standard deviations. Furthermore, at least 2 consecutive samples must be changed in the same way before the change was considered significant.

The changes in all parameters in different doses and saline are presented in Table 1. There is no significant change in any of the parameters of the saline (control) group.

Table 1. A summary of the pathophysiological changes seen in low doses of endotoxin administration in 6 groups of ewes.

	Saline	100 ng	100 ng	50 ng	50 ng	25 ng	25 ng	10 ng	10 ng	5 ng	5 ng
BT	0	+	+	0	+	0	+	+	+	+	+
PG	0	+	+	+	+	+	+	+	+	+	+
WBC	0	–	–	–	–	–	–	–	–/+	–/+	–/+
PMN	0	–/(+)	(–)/+	–/+	–/+	–/+	–/+	–/+	0/+	–/+	–/+
MN	0	–	–	–	–	–	–	–	–	–	–
Ca	0	–	–	–	–	0	0	N	N	N	N
Zn	0	–	–	–	–	–	–	–	0	0	0
Fe	0	–	(–)	–	–	–	–	–	–	–	(–)

0 indicates no change, + indicates a significant increase, – indicates a significant decrease and parenthesis indicates that the change are not significant but have a tendency to change. #/# the first sign indicates whether the change occurred and then followed with the change as indicated by the second sign. N indicates that the samples were not analysed.

Significant increase of body temperature was recorded in all endotoxin doses except for 1 animal in each group of the dose of 50 and 25 ng/kg. Similar changes or tendency to changes were found as in higher doses but presented in a shorter duration and less pronounced (*Aiumlamai & Kindahl* 1990). Prostaglandin $F_{2\alpha}$ metabolite levels significantly increased in all doses. Leukopenia in mononuclear cells and leukocytosis in polymorphonuclear cells after an initial decrease were clearly demonstrated in these low doses and the present findings agree to a previous report (*Yagoda et al.* 1990). This is also in agreement with the changes seen in disease syndromes such as ruminal acidosis, ruminal stasis and parturient paresis (*Aiumlamai & Kindahl* (in press), *Aiumlamai et al.* (in press a, b)). Significant decreases of Zn and Fe were recorded as well as a significant decrease of Ca levels except for the lowest doses of endotoxins. The animals were however followed only during a 6 h period, which might be a too short time for some of these parameters to change. Zinc and Fe seems to be better indicators than Ca to determine endotoxin exposure. Prostaglandin $F_{2\alpha}$ metabolite is a very relevant parameter to indicate exposure to low doses of endotoxin as well as the picture of WBC, particularly the leukocytosis of PMN.

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