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EFFECTS OF PROSTAGLANDIN E_2 , PROSTAGLANDIN $F_{2\alpha}$ AND ENDOTOXIN ON PLASMA CALCIUM LEVELS IN THE GOAT*

By

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FREDRIKSSON, GUNNAR, HANS KINDAHL and LARS-ERIC EDQVIST: *Effects of prostaglandin E_2 , prostaglandin $F_{2\alpha}$ and endotoxin on plasma calcium levels in the goat.* Acta vet. scand. 1984, 25, 378—384. — Intravenous administration of 2.6—3.3 $\mu\text{g/kg}$ of an endotoxin from *Salmonella typhimurium* to goats caused a marked drop in plasma Ca levels associated with an increase of prostaglandin synthesis and release measured as peripheral plasma levels of 15-keto-13,14-dihydro-PGF $_{2\alpha}$. This is one of the main metabolites of PGF $_{2\alpha}$, but also PGE $_{2\alpha}$ is partly metabolised to this compound. The infusion of 10 mg of PGF $_{2\alpha}$ lowered plasma Ca levels. Ten mg of PGE $_2$ did not change Ca concentrations significantly.

PGE $_2$; PGF $_{2\alpha}$; Ca.

Intravenous administration of endotoxins to goats has a luteolytic effect mediated by the synthesis and release of PGF $_{2\alpha}$ (Fredriksson *et al.* 1985). In cattle endotoxins are reported to produce hypocalcaemia (Griel *et al.* 1973, Fredriksson 1984). Luthman *et al.* (1983) found that i.v. administration of PGE $_2$ to cows after parturition produces a transient decrease in peripheral plasma levels of Ca. We wanted to further investigate the influence of endotoxins and i.v. administered PGF $_{2\alpha}$ and PGE $_2$ on plasma Ca levels in goats.

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MATERIAL AND METHODS

Five Swedish Landrace goats were used in the first experiment when 2.6–3.3 $\mu\text{g/kg}$ (100 μg per animal) of an endotoxin from *Salmonella typhimurium*, prepared by a method described by *Svenson & Lindberg* (1978) was administered into the jugular vein. Peripheral Ca and prostaglandin metabolite levels were followed with frequent blood sampling during the following 8 days (hourly during the first 30 h, then 5 times daily). In the second experiment 4 goats received 10 mg of $\text{PGF}_{2\alpha}$ (crystalline, Upjohn Company) and on another occasion 10 mg PGE_2 (Prostin E_2 , Upjohn Company). At both occasions the prostaglandin was dissolved in 1 % ethanol in saline and given during 1 h. Blood samples were collected at 15, 10, 5 min before and at 15, 30, 45, 60, 90, 120, 150, 240, 300, 360, 420 min and 24 h after the start of the infusion. The plasma was frozen immediately at -20° and then analysed for the content of 15-keto-13,14-dihydro- $\text{PGF}_{2\alpha}$ (*Kindahl et al.* 1976), progesterone (*Bošu et al.* 1976) and Ca (*Trudeau & Freier* 1967). The pre-experimental mean values for Ca of each animal were based on analysis of 20 plasma samples taken at different times.

RESULTS AND DISCUSSION

After i.v. administration of endotoxin Ca levels decreased approximately 20 % below pre-experimental concentrations

Figure 1. Percentage change in plasma Ca concentration after administration of endotoxin 2.6–3.3 $\mu\text{g/kg}$. Mean levels of 5 goats $\pm$ s.e.m. Zero for Ca is 2.56 mmol/l. Solid arrow indicates the time of endotoxin injection into the jugular vein.

Figure 2. Peripheral blood plasma levels of 15-keto-13,14-dihydro-PGF_{2α} (●—●) and plasma calcium concentrations (O---O) in one goat after i.v. administration of 2.6 μg/kg of endotoxin.

during the first 5–10 h (Figs. 1 and 2). After 2–3 days Ca slowly increased and reached initial levels after 6–8 days. The 15-keto-13,14-dihydro-PGF_{2α} metabolite levels were elevated during approximately 20 h after the administration of endotoxins (Fig. 2). When PGF_{2α} was given i.v. a slight, temporary decrease was seen in Ca levels during and shortly after the infusion (Fig. 3, upper panel). In the case of PGE₂ no obvious changes were found during the infusion (Fig. 3, lower panel).

These results differ from those reported in the cow, in which PGE₂ infusion is more effective in lowering circulating Ca than is PGF_{2α} (Luthman et al. 1983). This may be explained by differences between the two species in the metabolism of PGF_{2α} and PGE₂ (Kumlin et al. 1984). The infusion both of PGF_{2α} and PGE₂ gave rise to elevations in the 15-keto-13,14-dihydro-PGF_{2α} metabolite; the response of the former was however approximately 15 times larger (Fig. 4). It is known that PGE₂ is partly metabolised by a 9-keto-reductase and further to 15-keto-13,14-dihydro-PGF_{2α}. Other metabolites of PGE₂ are difficult to measure

Figure 3. Percentage change in plasma Ca concentration after intravenous infusion of 10 mg of $\text{PGF}_{2\alpha}$ (upper panel) and 10 mg of PGE_2 (lower panel). Mean level of 4 goats $\pm$ s.e.m. Zero for Ca is 2.39 mmol/l. Horizontal black bar indicates the time of infusion.

(Granström *et al.* 1982). The prostaglandin response following endotoxin infusion is probably very complex involving not only $\text{PGF}_{2\alpha}$ and PGE_2 , but also other products of the arachidonic acid cascade are formed which may influence Ca levels. It is known that many of the clinical signs observed after administration of endotoxin e.g. increased heart rate and diarrhoea are also seen after administration of prostaglandins of the E-family (Nakano 1973, Robert 1976).

Figure 4. Geometrical mean $\pm$ s.e.m. of peripheral blood plasma levels of 15-keto-13,14-dihydro-PGF_{2 α} after i.v. infusion of 10 mg of PGF_{2 α} (dotted line) and 10 mg of PGE₂ (dashed line). Note the logarithmic scale on the y-axis. Horizontal black bar indicates the time of infusion. Levels < 200 pmol/l are excluded from the figure.

When judging the response of plasma Ca to endotoxin administration in relation to the response after PGE₂ and PGF_{2 α} administration it should be emphasized that 15-keto-13,14-dihydro-PGF_{2 α} levels were elevated for about 20 h following administration of endotoxin (Fig. 2) and only approximately 3 and 6 h following PGE₂ and PGF_{2 α} infusions, respectively (Fig. 4).

All goats receiving endotoxin released enough prostaglandin to act luteolytically. In the case of PGF_{2 α} infusion progesterone values decreased by about 50 % in all animals after 3–5 h. In no case was luteolysis induced and the length of the oestrous

cycle did not change. No clear effect on progesterone was seen following PGE_2 infusion. Neither luteolytic nor luteotropic effects of the PGE_2 infusions were monitored and the length of the oestrous cycles was normal.

In conclusion this study demonstrates that i.v. administration of endotoxins to goats result in a marked elevation of prostaglandin synthesis and release and is associated with a drop in Ca levels. The prostaglandin response is probably very complex in nature. The infusion of $PGF_{2\alpha}$ but not PGE_2 also lowers plasma Ca levels.

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REFERENCES

- Bosu, W. T. K., L.-E. Edqvist, P. Lindberg, K. Martinsson & E. D. B. Johansson: The effect of various dosages of lynesterol on the plasma level of oestrogens and progesterone during the menstrual cycle in the rhesus monkey. *Contraception* 1976, 13, 677—684.
- Fredriksson, G., H. Kindahl & L.-E. Edqvist: Endotoxin induced prostaglandin release and corpus luteum function in goats. *Anim. Reprod.* 1985, 8, in press.
- Fredriksson, G.: Some reproductive and clinical aspects of endotoxins in cows with special emphasis on the role of prostaglandins. *Acta vet. scand.* 1984, 25, in press.
- Granström, E., F. A. Fitzpatrick & H. Kindahl: Radioimmunologic determination of 15-keto-13,14-dihydro- PGE_2 : A method for its stable degradation product, 11-deoxy-15-keto-13,14-dihydro-11 β , 16 ξ -cyclo- PGE_2 . *Methods in Enzymology* 1982, 86, 306—320.
- Griel, L. C. Jr., A. Zarkower & R. J. Eberhart: Clinical and clinicopathological effects of *Escherichia coli* endotoxin in mature cattle. *Canad. J. comp. Med.* 1975, 39, 1—6.
- Kindahl, H., L.-E. Edqvist, E. Granström & A. Bane: The release of prostaglandin $F_{2\alpha}$ as reflected by 15-keto-13,14-dihydroprostaglandin $F_{2\alpha}$ in the peripheral circulation during normal luteolysis in heifers. *Prostaglandins* 1976, 11, 871—878.
- Kumlin, M., G. Fredriksson, H. Kindahl & E. Granström: Metabolism of $PGF_{2\alpha}$ in goat and cattle: The use of two different PGF metabolites as parameters of PGF production. Poster Winter Prostaglandin Conference, Keystone, Colorado, USA, March 20—24, 1984.

- Luthman, J., S. O. Jacobsson & H. Kindahl*: The influence of prostaglandins on calcium homeostasis in dairy cows. *Zbl. Vet. Med.* 1983, 30, 1—9.
- Nakano, J.*: Cardiovascular actions. In: Ramwell, P. W. (ed): *The Prostaglandins*, vol. 1. Plenum Press, New York-London 1973, pp. 238—316.
- Roberts, A.*: Antisecretory, antiulcer, cytoprotective and diarrheogenic properties of prostaglandins. *Advances in Prostaglandin and Thromboxane Research* 1976, 2, 507—520.
- Svenson, S. B. & A. A. Lindberg*: Immunochemistry of Salmonella O-antigens: preparations of an octasaccharide-bovine serum albumin immunogen representative of Salmonella serogroup B O-antigen and characterization of the antibody response. *J. Immunol.* 1978, 120, 1750—1757.
- Trudeau, D. L. & E. F. Freier*: Determination of calcium in urine and serum by atomic absorption spectrophotometry (AAS). *Clin. Chem.* 1967, 13, 101—114.

SAMMANFATTNING

Effekter av PGE₂, PGF_{2α} och endotoxin på plasma Ca-nivåerna hos get.

Ett endotoxin från *Salmonella typhimurium* gavs iv till getter i doser om 2,6—3,3 µg/kg. Plasma Ca-nivåerna sjönk med ca 20 % under de första 5—10 timmarna för att efter 2—3 dagar långsamt öka och nå ursprungskoncentrationer efter 7—8 dagar. Höga nivåer av 15-keto-13,14-dihydro-PGF_{2α} uppmättes under ca 20 timmar efter injektionen. Detta är en av huvudmetaboliterna till PGF_{2α}, men också PGE₂ metaboliseras i mindre omfattning till denna förening. När 10 mg PGF_{2α} gavs iv under en timme erhöles en övergående sänkning av Ca-nivåerna. Då PGE₂ gavs på motsvarande sätt påverkades inte plasma Ca-koncentrationerna påtagligt.

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