



## Effect of the estrous cycle on the oxidative burst activity of blood polymorphonuclear leukocytes in cows

A. Chaveiro\*, R. Agrícola, I. Carvalhais, M. Faheem and F. Moreira da Silva

Department of Agrarian Sciences - Animal Reproduction - University of the Azores, 9701-851, Terra-Chã, Portugal  
CITA-A, Centro de Investigação e Tecnologias Agrárias do Açores, Largo da Igreja, 9701-851 Terra Chã, Portugal  
\*antoniochaveiro@uac.pt

### Introduction

Blood polymorphonuclear leukocyte (PMN) oxidative burst activity, plasma cortisol levels, and the total and differential white blood cells counts (WBC) of six cyclic dairy cows were evaluated

Throughout the cycle the various parts of the female reproductive tract undergoes changes brought about the action of the pituitary and ovarian hormones. The higher susceptibility to disease is well-documented in the periparturient period in cattle and supports the idea of a relation between the reproductive hormonal oscillations and immunoactivity. Indeed, PMN function varies due to reproductive stages, concomitant with variations in steroid hormone concentrations.

### Objectives

The present study was designed to evaluate whether the PMN's oxidative burst in dairy cows could be altered during the oestrous cycle. Plasma cortisol levels, total white blood cells (WBC) and their differentiation were also evaluated during the same period.

### Materials and Methods

#### Animals

- 6 clinically healthy multiparous lactating Holstein Friesian cows

#### Blood collection

- Blood (10ml) was aseptically collected from the external jugular vein in vacuum tubes (L.D.M France).

#### Polymorphonuclear leukocyte oxidative burst activity measurement by Flow Cytometry

- Evaluated by flow cytometry, measuring the intracellular oxidation of 2',7'-dichlorofluorescein diacetate (DCFH) to 2',7'-dichlorofluorescein (DCF) by  $H_2O_2$ -production. Results were presented as the mean fluorescence intensity (MFI) of DCF~.

#### Cortisol and progesterone measurements

- Plasma cortisol levels were determined using a fluorimetric method (Delfia Cortisol Kit, Wallac, Turku, Finland). Plasma progesterone was measured using a solid-phase radioimmunoassay test (Diagnostic Products Corp., Los Angeles, USA)

### Results

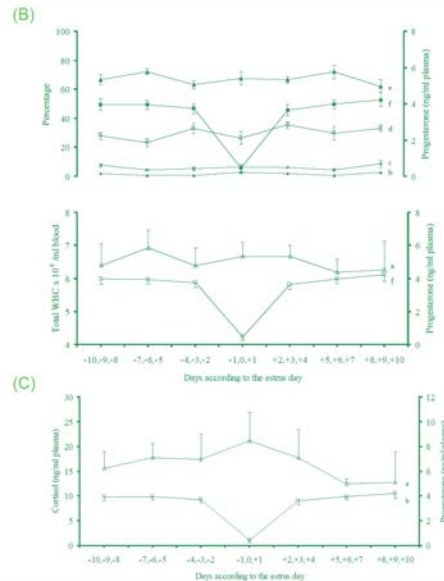
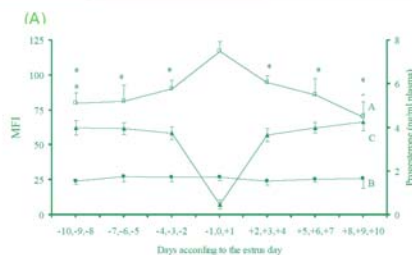


Figure 1- (A) Variation of PMN's oxidative burst according to the estrous cycle of six cows. A, PMA-activated PMN, B, resting PMN, C, Plasma progesterone levels. (B) Total (a) WBC count during the estrous cycle of six cows, and the respective differentiation (%): monocytes (b), eosinophils (c), neutrophils (d), lymphocytes (e) and plasma progesterone levels (f). (C) Variation of (a) plasma cortisol levels and (b) plasma progesterone levels during the estrous cycle of six cows as evaluated by a fluorimetric method. Each value represents the means±SEM. Day 0 represents the estrus day. \*, P<0.05; \*\*, P<0.01

### Conclusions

- The maximum PMN's burst activity (MFI = 117.2±7.4) occurred during the oestrus period (days -1, 0, +1).
- No statistical differences in the total and differential WBC counts were observed during the whole period.
- The maximum value of cortisol was reached during the oestrus period, [21.54 ng/ml plasma (±3.97)] followed by a decrease during the luteal phase of the oestrous cycle with a minimum [12.7 ng/ml plasma (±0.84)] reached on the days +5,+6,+7.
- The oestrous cycle alters, directly or indirectly, bovine PMN function, altering thus the normal immune activity of the animals. Because estradiol 17  $\beta$  and progesterone are present concurrently in bovine blood in varying concentrations, the ratio of these two hormones may be important determining factor in the effect on the immune system.

### Acknowledgements

This work was supported by the Luso-American Foundation. (FLAD), grant number 322-5/04. CITA-A is fully acknowledged.