

Radioimmunoassay of Pregnancy-Associated Glycoprotein 1 (PAG-1) isolated from Zebu (*Bos Indicus*) Placenta: Preliminary Results

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Introduction

- ⚡ *Bos indicus* is used interchangeably with the term zebu which originates from the Tibetan word Zen or Zeba, which means "the hump of the camel".
- ⚡ Zebu cattle is an important source of animal protein in many countries, being more effective in extracting nutrients from low quality roughages and having a remarkable ability to tolerate high temperatures
- ⚡ Reproductive differences between domestic (*Bos taurus*) and zebu cattle have been described. Zebu cattle reach puberty later (16 to 40 months of age) than *Bos taurus* x *Bos indicus* crossbreeds or purebred taurine cattle. Different authors reported also longer pregnancy lengths in some breeds of zebu cattle as well as in crossbreed *Bos taurus* x *Bos indicus*
- ⚡ Till now, in zebu cattle, no specific studies were made in order to describe placental production of hormones and pregnancy-associated proteins during normal and/or pathologic gestations
- ⚡ Production of new reagents isolated from zebu placenta can be useful for the development of homologous radioimmunoassay (RIA) systems. In veterinary practice, these RIA may be used for both pregnancy diagnosis and follow-up of fetal well-being

Aim

The aim of this study was to analyze the characteristics of a new preparation of zebu PAG-1 in the development of a new homologous PAG radioimmunoassay system

Material and Method ~1

Preparation of antigen



Placenta

Extraction

Acid precipitation

Ammonium sulfate precipitation (0-40% and 40-80% A.S.)

Anion exchange chromatography (DEAE Sephadex A25 column)

Cation exchange chromatography (CM Ceramic column)

Lyophilization

Immunization of rabbits

Sousa et al, 2000b

Material and Method ~2

Radiolabelling and antiserum production

- ✓ Purified zebu PAG-1 (0.04 M NaCl fraction of DEAE Sephadex A25 column, CM ceramic column peak XI) was radiolabelled by lactoperoxidase method
- ✓ Antiserum against zebu PAG-1 was raised in rabbits (250 µg of lyophilized antigen were injected at 2-week intervals)
- ✓ First bleeding was made 1-week after the third immunization

Results and Discussion

- ✓ In the presence of antibody in excess, 73.6% of labeled zebu PAG-1 was bound
- ✓ The optimal dilution of the antiserum issued from the first bleeding (1-week after the third injection of antigen) was 1:100 000
- ✓ These first results indicate that new reagents prepared from zebu placenta are now available for the development of a homologous RIA system in this species

Perspectives

- ⚡ Further investigations are in progress to produce new reagents for the development of a highly sensitive radioimmunoassay method for PAG detection in peripheral blood of pregnant zebu females
- ⚡ This new system could be used in experimental and/or farm conditions in order to improve the knowledge about the endocrine physiology of zebu females

General References

Mukasa-Mugerwa E. ILCA Monograph 6. ILCA, Ethiopia.

Sousa, NM et al. (2000a). 16th Scientific Meeting AETE., p.212-213.

Sousa, NM et al. (2000b). 27th Conference of the IETS (abstract accepted).

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