

INTRAMUSCULAR BOVINE PREADIPOCYTE *IN VITRO* CULTURE MODEL



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INTRODUCTION

- Developing a greater understanding of bovine adipocyte biology may lead to devise strategies that increase fat deposition in intramuscular depots in order to improve beef quality. Nevertheless, the mechanisms regulating adipogenesis in ruminants are poorly understood.
- Primary preadipocyte in vitro systems are very useful models to study the adipocyte biology of meat animals under controlled conditions, avoiding extensive use of live animals.

OBJECTIVES

- The main objective of this work was to develop an in vitro cell culture system using a chemically defined medium to differentiate primary intramuscular bovine preadipocytes obtained from the local cattle breed Pirenaica, characterized by low marbling deposition. The system will be further used to study the molecular mechanisms of adipogenesis.

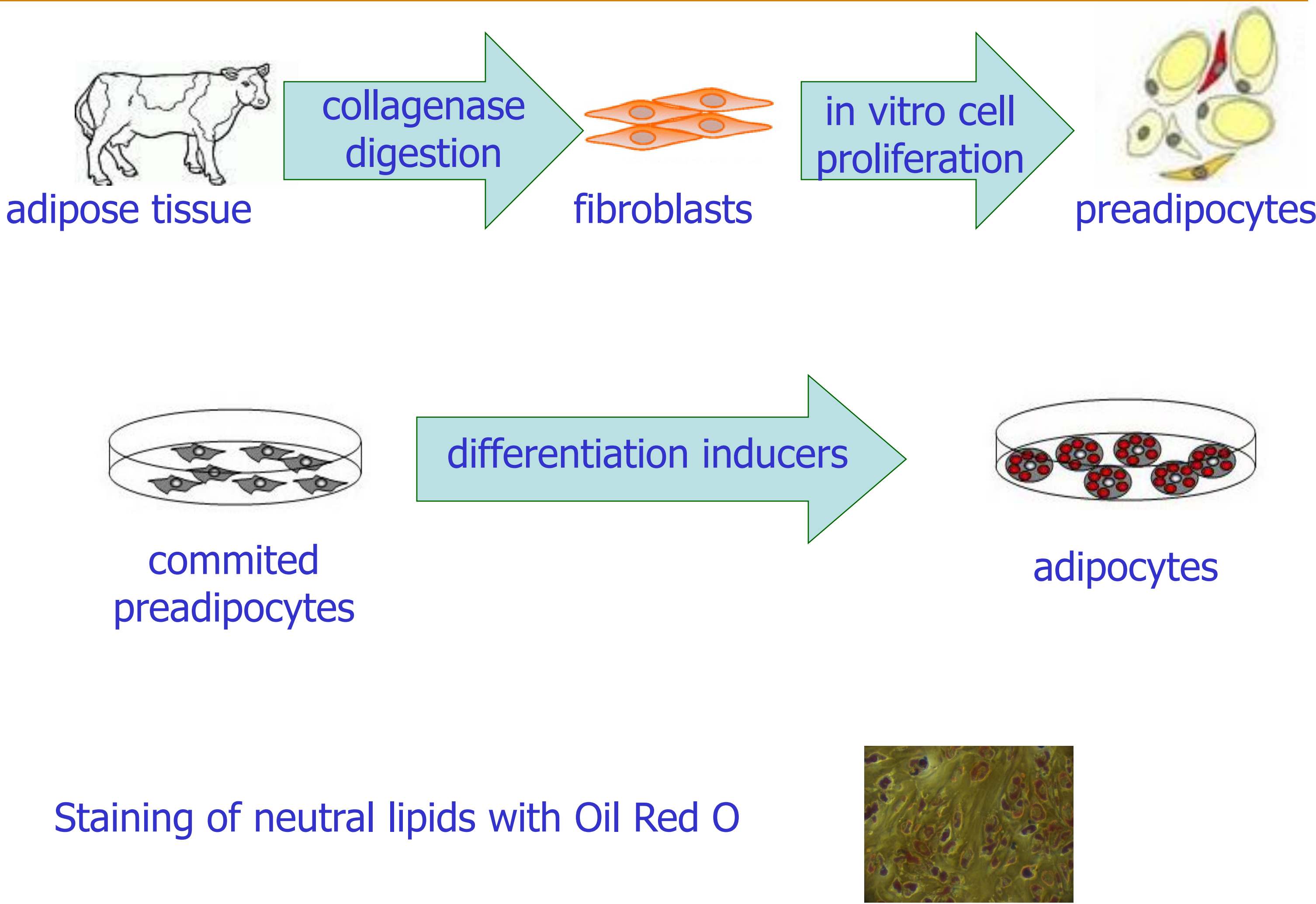
METHODS

- Primary preadipocytes isolation and in vitro culture

- Induction of preadipocytes differentiation

- Basal Medium (BM): DMEM/F12 + Antibiotics + [1.6 mg/ml Insulin¹ or 0,5%(v/v) ITS²; 10 nM Dexamethasone; 0,5 mM Isometilbutilxantine; 0,12 % w/v Bovine serum albumin; 1 mM Octanoate]
- Other additions to Basal Medium assayed: 10 µM Rosiglitazone (Ros); 10 µM Troglitazone (Tro); 0,5 µM Biotine (Bio); 200 mM Oleic Acid; 0,55mg/ml transferrin; 0,5 µg/mL selenium

- Assessment of differentiation



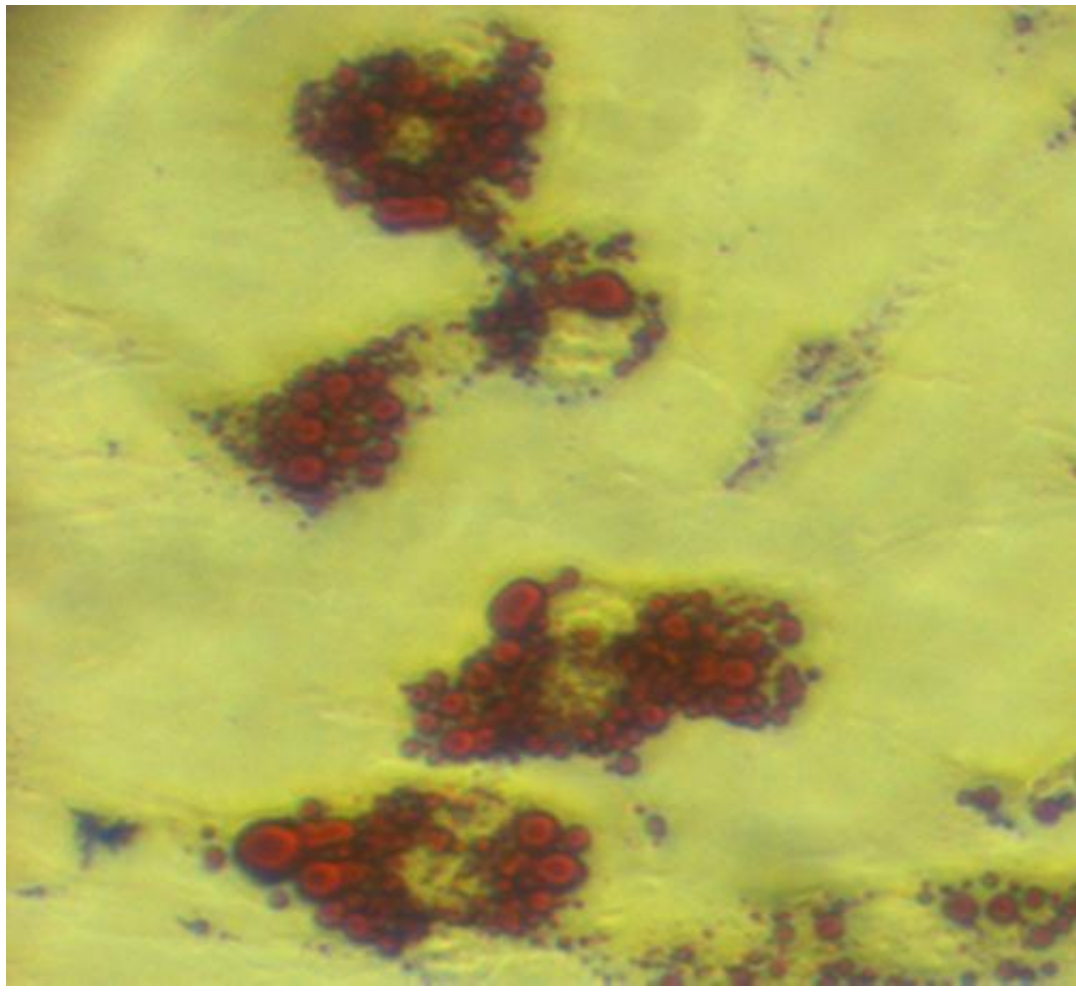
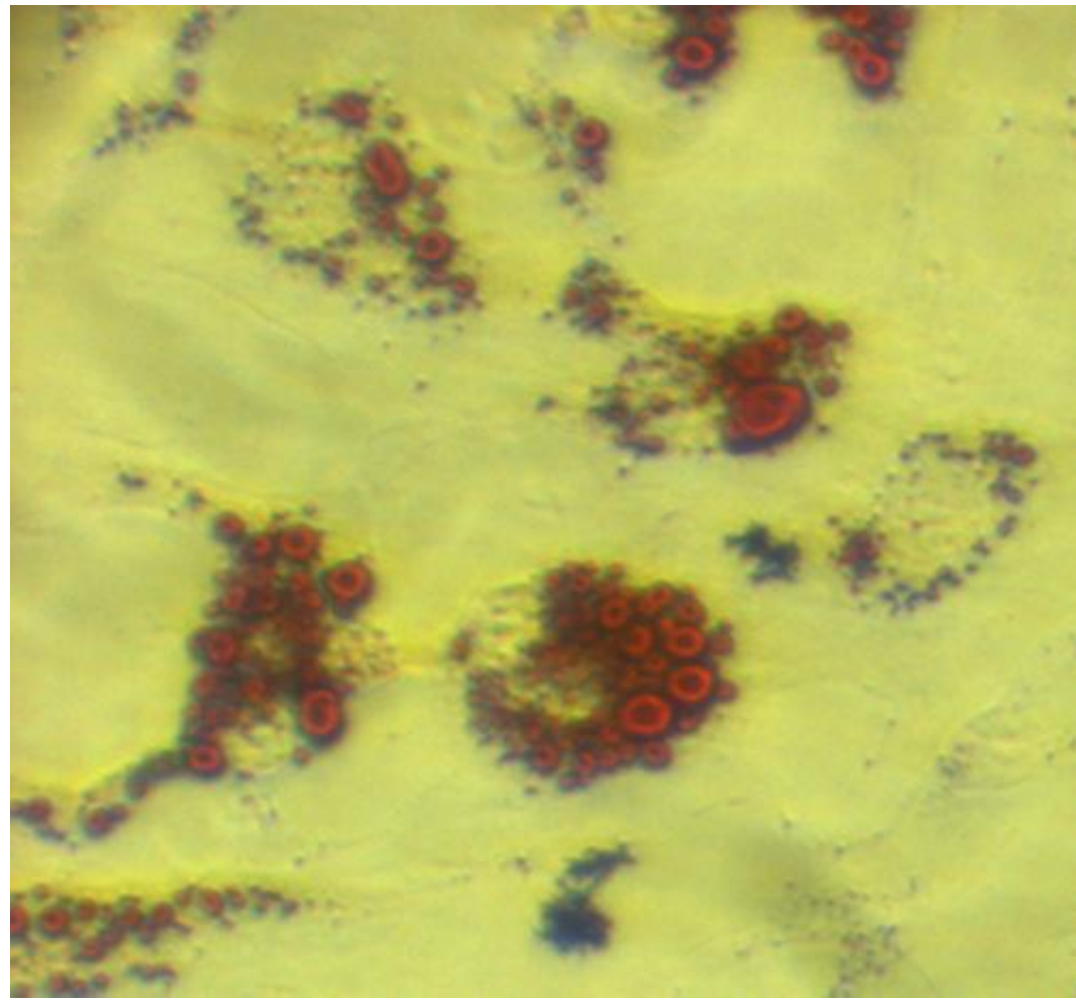
RESULTS

- Both subcutaneous and intramuscular stromovascular cells were obtained, grown to confluence and challenged with different combinations of compounds in order to get preadipocyte differentiation.

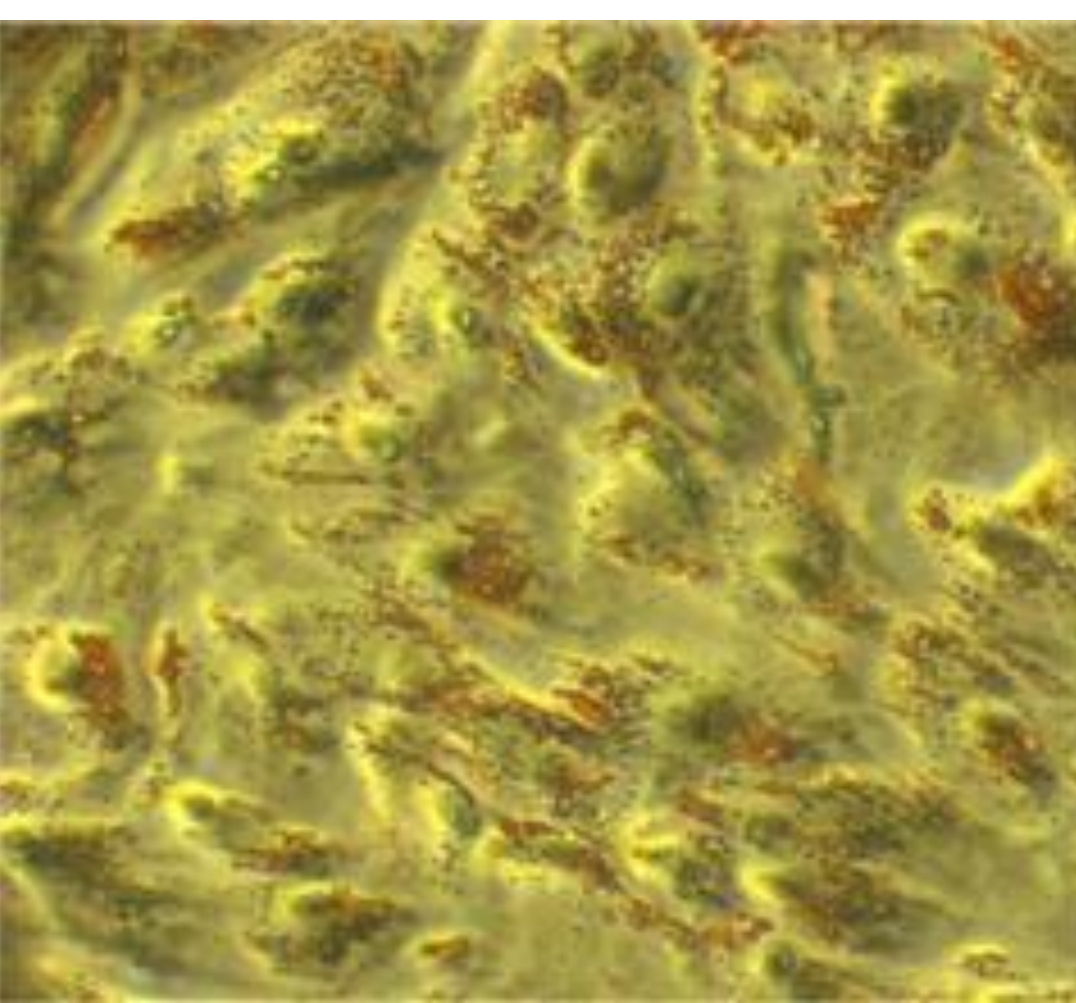
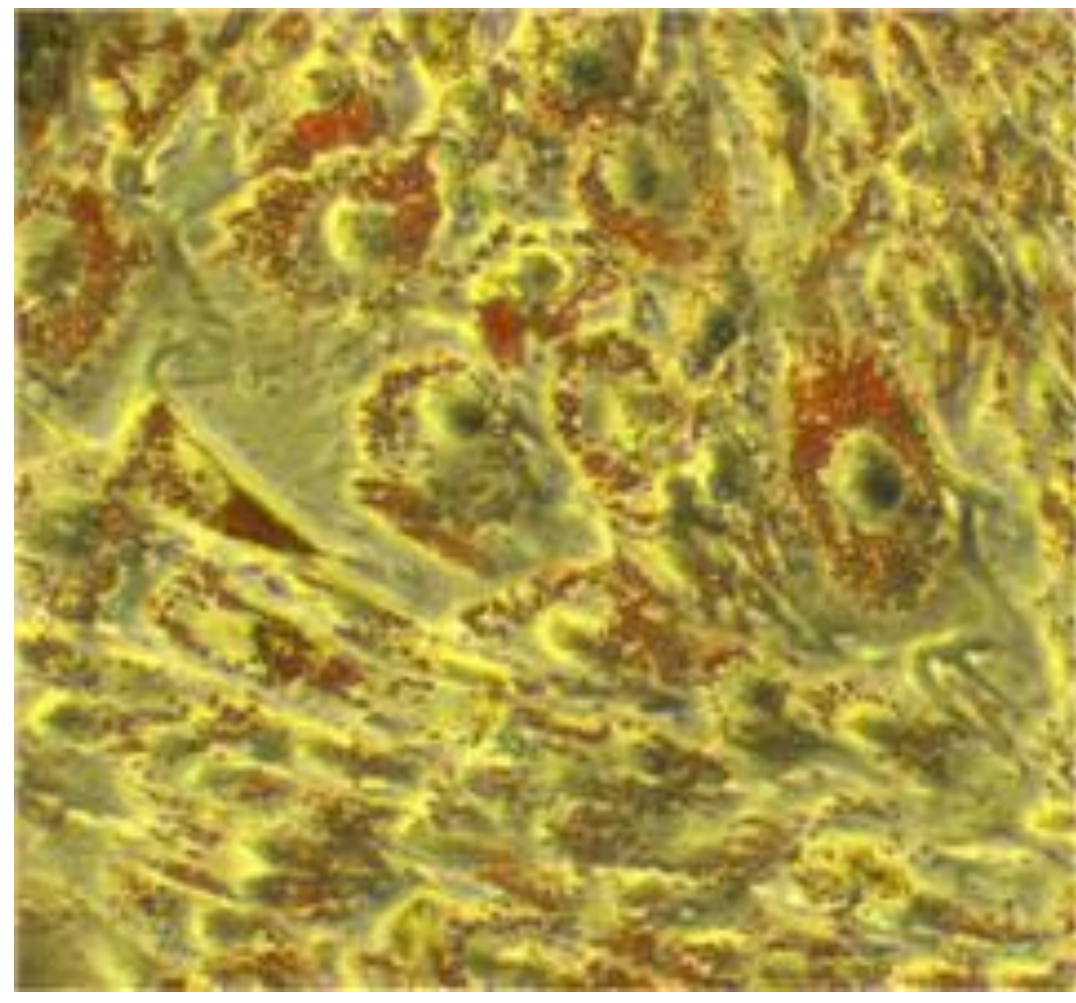
SUBCUTANEOUS

INTRAMUSCULAR

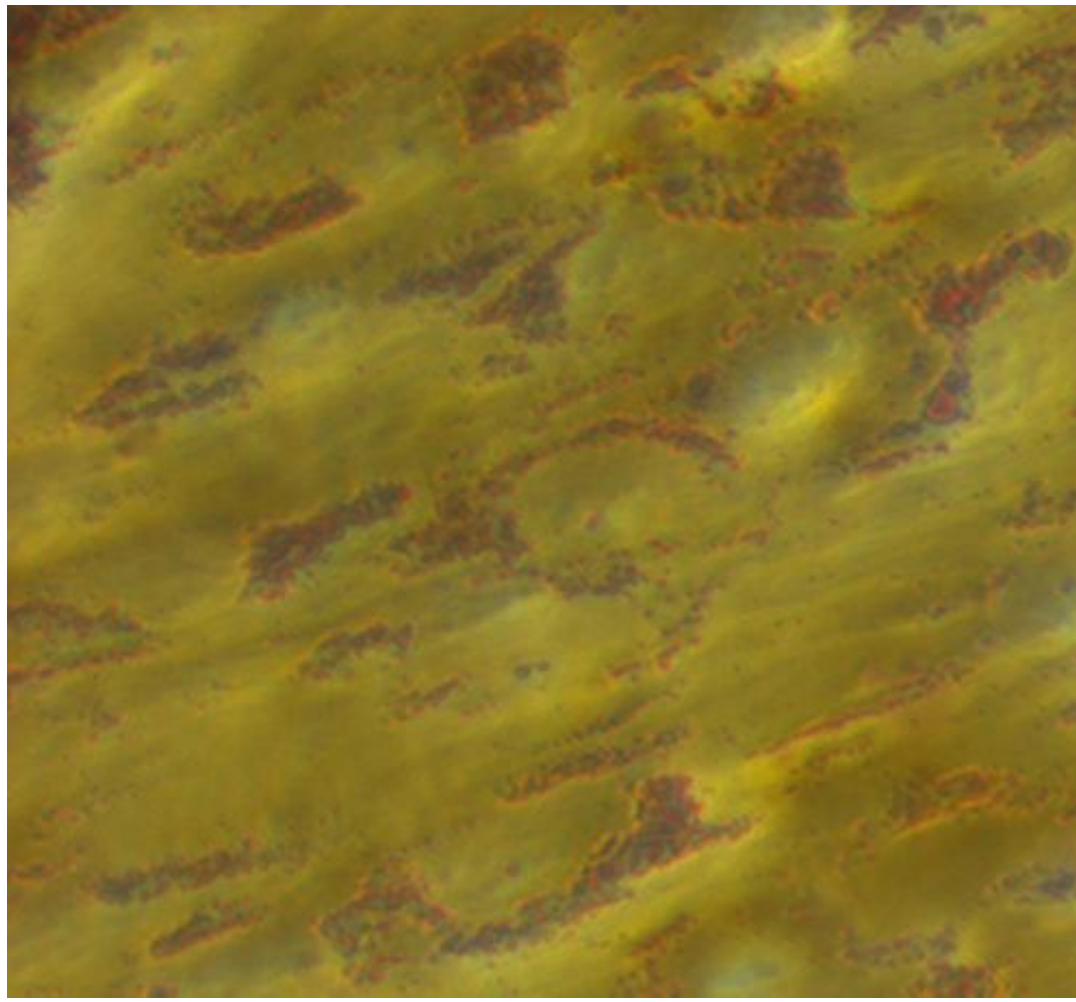
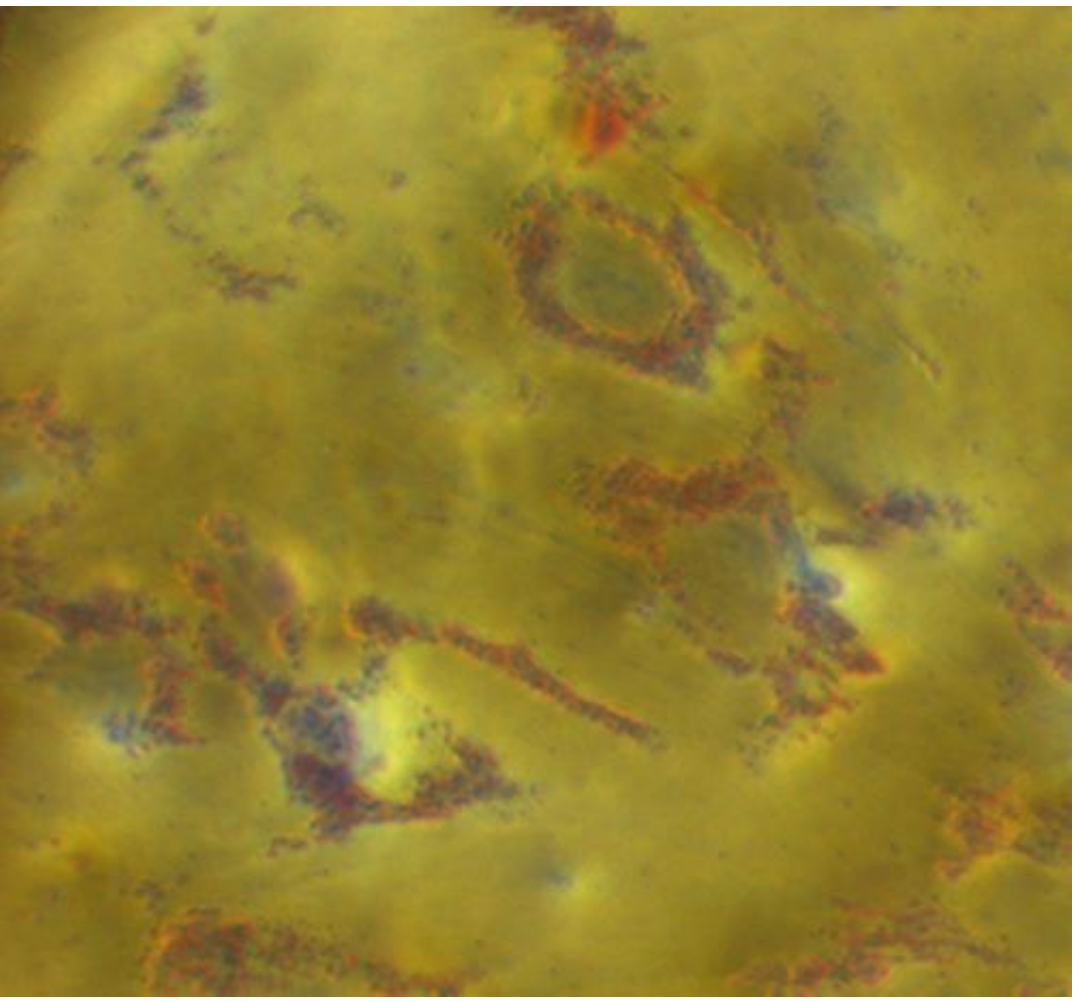
BM²
+ Bio
+ Ros



BM²
+ Tro



BM¹
+ Bio
+ Ros



BM²
+ Tro

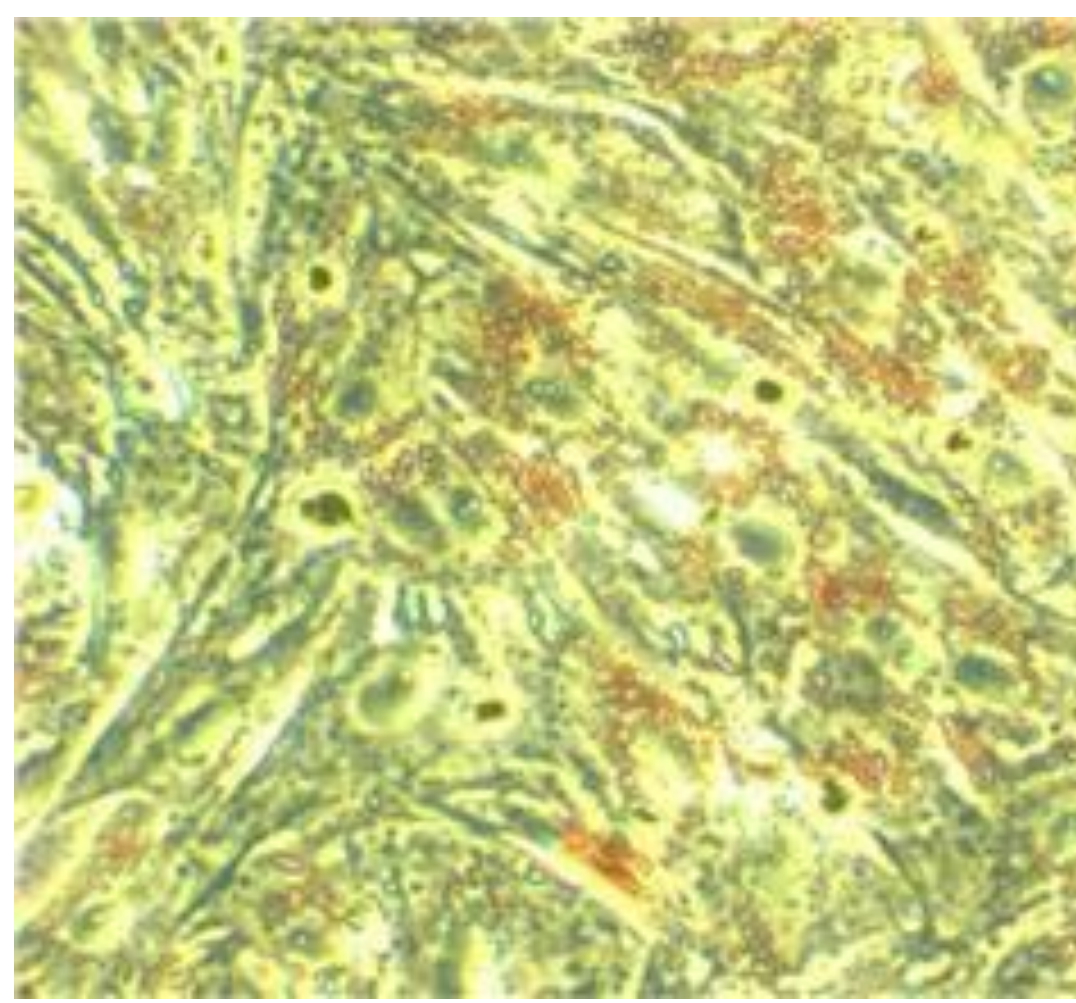
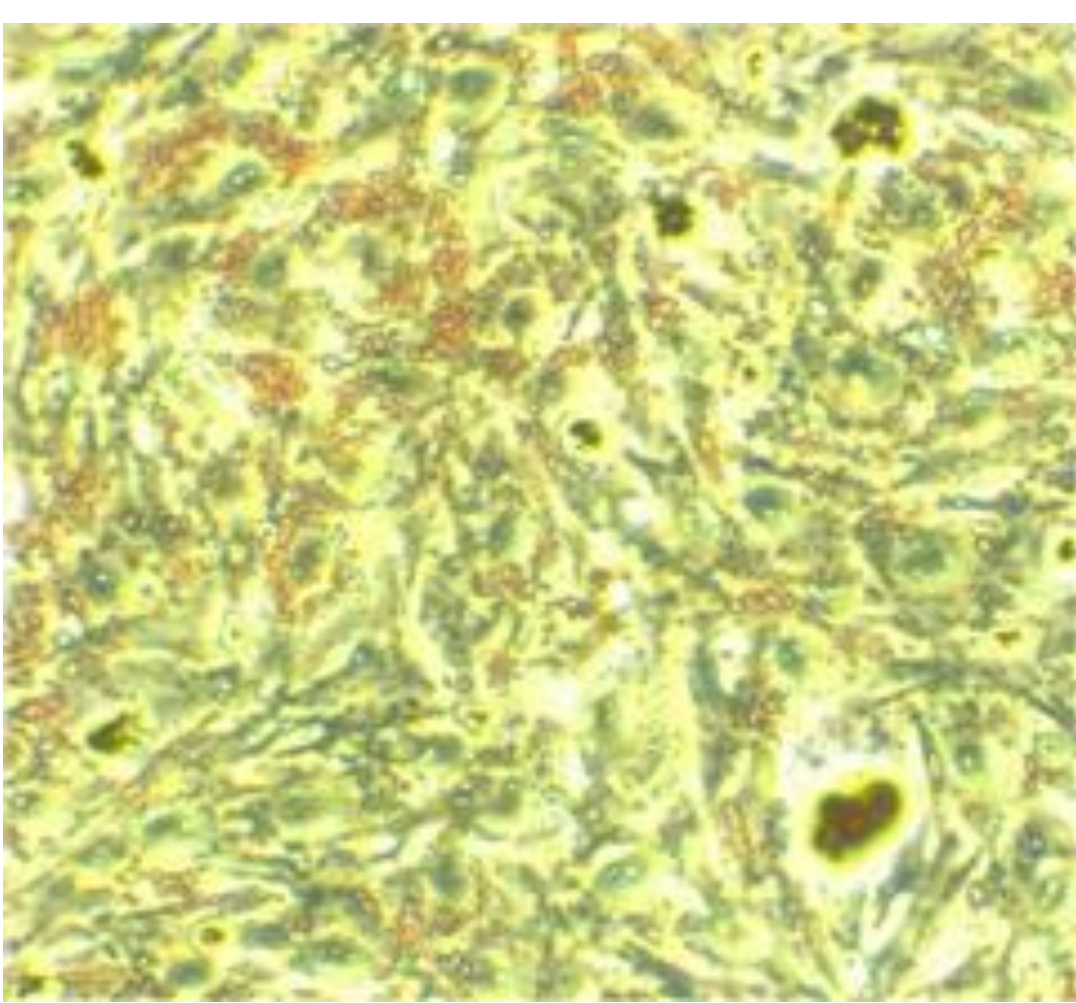


Figure 1.- Microphotographs (x100) of subcutaneous (SC) and intramuscular (IM) preadipocytes differentiating after induction with two identified combinations of compounds that achieved preadipocyte differentiation.

CONCLUSIONS

- Two combination of compounds that achieved the differentiation of IM and SC bovine preadipocytes were identified, both of them requiring the presence of a PPARγ agonist (rosiglitazone or troglitazone). Instead, the standar methods to differentiate preadipocytes did not work with those type of cells obtained from low marbling deposition cattle as the Pirenaica steers.
- SC bovine preadipocytes differentiate to a higher extent than cells from IM depot under the same conditions.