



# Preliminary results of in vitro fertilization for the endangered murciano-levantina bovine breed

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## A B S T R A C T

The Murciano-levantina cattle from the Spanish South-East belongs to a bovine endangered breed under the special protection of FAO (WWL-DAD:3)\*. Their characteristics of rusticity, longevity, docility and disease resistance make interesting the recovery of the breed. The objective of the present work was to determine the efficiency of in vitro bovine embryo production using oocytes collected from post mortem ovaries fertilized with frozen-thaw sperm obtained either from commercial or Murciano-levantina bulls.

matured for 24 h in TCM199 with 10% FBS, 2mM L-glutamine, 0.2 mM sodium pyruvate, 0.3 µg/mL LH and µg/mL FSH. Oocytes presumed matured were fertilized with Percoll-prepared sperm (750.000/mL) in IVF-TALP as described by Parrish et al. (Biol. Reprod 38:1171-1180, 1988). Zygotes (at 18 h) or two-cell stage embryos (at 48 h) were assessed by staining with Hoechst 33342 (µg/mL; 20 min) under an epifluorescence microscopy. The experiment was replicated on 4 occasions.

%, average number of spermatozoa inside the oocyte (S/O), monospermy (MON- %) putative embryos (zygotes with two pronuclei, 2 PN-%) and percentages of cleavage at 48 hours (Table 1) were recorded. The results show that early embryos can be obtained with no difference with the source of sperm used in the monospermy and the cleavage rate between commercial and Murciano-levantina sperm ( $p > 0.05$ ).

\*Lista Mundial de Vigilancia para la Diversidad de los Animales Domésticos (1997). <http://www.fao.org/docrep/V8300S/V8300S00.htm>

Cumulus oocyte complexes were

The results of penetration (PEN-

### In vitro fertilization results with one commercial and one Murciano-levantina bull

	N <sup>1</sup>	PEN <sup>1</sup>	S/O <sup>1</sup>	MON <sup>1</sup>	2PN <sup>1</sup>	N <sup>2</sup>	1CELL <sup>2</sup>	2-4CELLS <sup>2</sup>	>4CELLS <sup>2</sup>
Commercial	79	62.02±5.49	1.38±0.11	77.55±6.02	61.22±7.03	92	55.43±5.21	34.78±4.99	9.78±3.11
Murciano-levantina	76	60.53±5.64	1.48±0.16	76.09±6.35	73.91±6.54	95	53.68±5.14	29.47±4.70	16.8±3.85
P		0.849	0.657	0.868	0.191		0.811	0.440	0.158