

**A DOSE-RESPONSE STUDY OF IVERMECTIN  
IN A BOVINE IN VITRO PRODUCTION SYSTEM**

B. Avery and M. Schmidt.

Department of Reproduction, Royal Veterinary and Agricultural University,  
Bülowsvej 13, DK-1870 Frederiksberg C, Denmark

Recent data have suggested that use of the antiparasitic drug, Ivermectin, might have a delayed adverse effect on fertility in cattle. Our aim was to perform a dose-response study in order to test whether oocytes and embryos exposed to Ivermectin, *in vitro*, would be affected as judged by cleavage rates at Day 2, blastocyst rates at Day 8 post-insemination, and by embryo morphology. Slaughterhouse oocytes from animals not treated with Ivermectin were matured (IVM) for 24 h in TCM-199 medium with 10% estrous cow serum and Ivermectin. Ivermectin was not present during insemination (IVF). The embryos were then co-cultured (IVC) in Menezo-B2 medium with 10% ECS and bovine oviduct cells in the presence of Ivermectin for 8 d. The Ivermectin stock solution was dissolved in DMSO. Two controls were included in each experiment: with the same concentration of DMSO as in the Ivermectin group, and without DMSO. All cultures were performed without oil in 0.5 ml of medium at 39°C and 5% CO<sub>2</sub> in air. The tested Ivermectin concentrations were 0.2, 2, 20 and 100 µg/ml, with 0.2 µg/ml being in the same order of magnitude as reported for therapeutic plasma and tissue concentrations.

Table 1. Effect of Ivermectin on development of bovine IVM oocytes and IVF/IVC embryos

Group	Development	Ivermectin	DMSO-control	Control
0.2 µg/ml	Cleavage	76% (111/146)	76% (32/42)	83% (33/40)
	Blastocysts	21% (31/146)	19% (8/42)	30% (12/40)
2 µg/ml	Cleavage	63% (30/48)	78% (50/64)	77% (37/48)
	Blastocysts	19% (9/48)	22% (14/64)	23% (11/48)
20 µg/ml	Cleavage	60% (145/243) <sup>B</sup>	74% (78/105) <sup>A</sup>	84% (88/105) <sup>A</sup>
	Blastocysts	7% (13/189) <sup>*</sup>	13% (14/105)	14% (15/105)
100 µg/ml	Cleavage	0% (0/111) <sup>C</sup>	68% (70/103) <sup>A</sup>	72% (49/68) <sup>A</sup>
	Blastocyst	0% (0/111)	0% (0/103)	10% (7/68)

<sup>A,B,C</sup>Different letters within columns denote significant differences (<sup>A,B</sup>P<0.01; <sup>A,C</sup>P<0.0001)

<sup>\*</sup>: 54 ova lost, one well contaminated

Table 1 shows no effect of Ivermectin at 0.2 µg/ml. A slight decrease in the cleavage rates was noted at 2 and 20 µg/ml. This was only significant at 20 µg/ml, a 100 X therapeutic concentration, which will probably not occur during treatment. No morphological differences between Ivermectin treated and control embryos were seen; at Day 8 all blastocysts were either expanded or hatched. At 100 µg/ml Ivermectin, a pronounced toxic effect was seen. In conclusion no pronounced effect of Ivermectin in doses up to 20 µg/ml could be demonstrated for IVM oocytes or IVF/IVC embryos. Supported by the Animal Biotechnology Research Centre.