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Culturing of Bovine Pre-Compacted Morlae in TCM-199 and Baf in a Standard 5% CO2 Laboratory Incubator and in the Vagina of a Goat Doe

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Abstract : Since more than half a century ago, attempts have been made to culture cells and embryos outside the body (in vitro or ex vivo). This was done with different culture media and in various “incubators”. In the present study two different culture media were used: a standard TCM-199 culture medium and first trimester amniotic fluid (BAF) collected sterilely from pregnant cows after slaughter. Two different culture conditions were also investigated, the standard laboratory CO2 incubator versus culturing bovine embryos in the vagina of a goat doe. Two experiments were done: Firstly the permeability of different receptacles to CO2 gas was analyzed for possible culture in the vagina. Four-well plates and straws were used to incubate TCM-199 and BAF for a period of 120 h in the presence or absence of 5% CO2 gas. The pH values were measured and recorded every 24 h. In the second experiment pre-compacted morula stage bovine embryos were cultured in the above culture media in sealed 0.25 mL straws in a standard laboratory incubator and in the vagina of a goat doe. Evaluation was done on (1) stage of development and (2) number of blastomeres after 96 h of culture. In the first experiment it was shown that the CO2 gas diffused out of the 4-well plate as well as through the wall of the straws in the absence of CO2 gas, while in the presence of CO2 the pH of both media stabilized between 7.3 and 7.5. This meant that the semen straws were permeable to CO2 gas and could therefore be used as receptacles for culturing early stage bovine embryos. In the second experiment no statistical differences ($p>0.05$) were found in the number of pre-compacted bovine embryos that developed to the blastocyst stage, or the hatched blastocyst stage, neither for the culture medium used, or the method of culturing in the two incubators. Neither was there any difference ($p>0.05$) in the number of blastomeres that developed at the blastocyst stage between the two types of incubators. The bovine embryos tended to develop more blastomeres when cultured in BAF than when cultured in TCM-199 in both the standard laboratory incubator and when using the vagina of a goat doe as an incubator.

Keywords : alternative culture, bovine embryos, vagina, bovine amniotic fluid, incubator
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