

C 427017: MAJOR BIOLOGICAL SCIENCE

KEY WORD: BOVINE EMBRYO/ SEX DETERMINATION/ IN VITRO / PCR

SUNUNTHA SAKTHAWEEKULKIT : SEX DETERMINATION OF IN VITRO FERTILIZED BOVINE EMBRYOS BY POLYMERASE CHAIN REACTION.

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Accuracy of polymerase chain reaction technique in determining sex of bovine embryo was studied. Embryos obtained from in vitro fertilization were cultured in modified Hamster Culture Medium-3 (mHECM-3) without glutamine. Effect of both non-essential (NEA) and essential (EA) amino acids, glucose and phosphate on the development of embryos through blastocyst stage during 72-192 hour after fertilization was also investigated. In vitro fertilized and developed 2- to \geq 7-cell embryos were randomly distributed to culture in mHECM-3 with the above mentioned substances added. Results indicated that medium with NEA+EA added supported the development of bovine embryos significantly higher ($p > 0.05$) than medium containing NEA or EA alone, with the number of total blastocysts $34(20.6 \pm 11.2)$ and expanded blastocysts $28(17.2 \pm 10.2)$ against [$24(14.3 \pm 12.0)$, $14(8.4 \pm 12.1)$] and [$14(8.3 \pm 11.2)$, $10(6.3 \pm 10.9)$] respectively. Increasing of glucose concentration added to medium containing NEA+EA tend to decrease both the number of total blastocysts and expanded blastocysts. Adding of glucose+phosphate in the medium did not increase the support of embryonic development either.

Sex determination of bovine embryos by means of DNA amplification through polymerase chain reaction technique was able to identify 49 out of 50 embryos (98%) studied, of which, 21 embryos (42%) were male and 28 embryos (56%) were female. One of these embryos did not show DNA band. The ability to identify sex of embryos clearly and quickly by this technique before transfer of embryos to suitable recipients will be of great advantage in planning sex of calves one would like to obtain. This technique may be applied to breeding and development of economic species in the future.

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