

D-41 Effect of 5-Azacytidine on Gene Expression and Development of Preimplantation Mouse Embryos

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Methylation of DNA appears to be an important regulation mechanism for sequential gene expression during embryonic development. The effects of cytidine analog 5-azaC, a demethylating agent, on embryonic development, and on the expression pattern of mRNAs for DNA-cytosine methyltransferase (DCMT), hypoxanthine phosphoribosyl transferase (HPRT), and insulin-like growth factor I receptor (IGF-IR) using the method of RT-PCR were studied in preimplantation mouse embryos. The temporal pattern of DCMT and IGF-IR mRNA showed a gradual decrease from GV stage to 8-cell stage, followed by an increase upto blastocyst stage. The high level of HPRT mRNA in GV oocytes was temporally decreased at one-cell stage, thereafter increased from 2-cell to blastocyst stage. However, the expression pattern of corresponding gene in the embryos treated with 5-azaC was not different in comparison with control. At varying concentrations (5-100 μ M) of 5-azaC, the embryos underwent mitotic arrest dose-dependently at all stages of embryos. In conclusion, the mitotic arrest by 5-azaC may be resulted from perturbation of other gene expression rather than covalent binding of DCMT.

D-42 Effect of cAMP on Connexin43 (Cx43) Transcription and Gap Junctional Communication in Preimplantation Mouse Embryo

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The present study was undertaken to investigate the temporal expression of Cx43, and the role of cAMP on Cx43 transcription and gap junctional communication in preimplantation mouse embryo. Cx43 mRNA was first detected at 2-cell stage and increased upto blastocyst stage. The amount of Cx43 mRNA was not altered significantly by treatment with H8, Rp-cAMP, or 8-Br-cAMP for 12 hr. Embryos treated with 50 μ M H8 or Rp-cAMP showed delay in fluorescent dye transfer and some of the embryos treated with H8 failed to transfer LY to all cells within 10 min. Dye transfer was speed up by 8-Br-cAMP. When embryos at late 8-cell stage were treated with 50 μ M H8 or 2mM Rp-cAMP, the number of embryos that developed blastocyst stage was decreased according to the culture time. Temporary effect of 8-Br-cAMP made any difference in development beyond compaction and blastocyst formation. In conclusion, Cx43 mRNA expressed after 2-cell stage may be not modulated by cAMP and cAMP-dependent protein kinase A(PKA), but communication through gap junction consisted of Cx43 may be regulated.