GENE TARGETING IN CATTLE—A GENETIC ISSUE WITH AN EPIGENETIC FACET

Zhongde Wang
Dept. of Epigenetics and Development
Hematech, INC
A division of Kirin Pharmaceutical
Sioux Falls, SD, 57106

ABSTRACT

Due to the lack of embryonic stem cells from farm animals, gene targeting in these species is a challenge. Particularly, there was no method available for knocking out a transcriptionally silent gene or for performing double targeting to produce homozygotes in these species. By using animal cloning (somatic cell chromatin transfer), we developed a sequential gene targeting system in using primary bovine fibroblast cells to knock out both alleles of a silent gene encoding immunoglobulin-mu (IGHM). Furthermore, we also sequentially knocked out both alleles of the prion gene encoding bovine prion protein (PRNP) in the IGHM targeted cell line and produced doubly homozygous knockout calves. Since such sequential gene targeting strategy requires multiple rounds of animal cloning, adequately reprogramming the epigenome of the targeted cells by the oocytes became an important issue for consideration. This presentation will discuss our sequential gene strategy in bovine and some of the issues involved in epigenetic reprogramming.