



Identification of transgenic cloned dairy goats harboring human lactoferrin and methylation status of the imprinted gene *IGF2R* in their lungs

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ABSTRACT. Dairy goat is a good model for production of transgenic proteins in milk using somatic cell nuclear transfer (SCNT). However, animals produced from SCNT are often associated with lung deficiencies. We recently produced six transgenic cloned dairy goats harboring the human lactoferrin gene, including three live transgenic clones and three deceased transgenic clones that died from respiratory failure during the perinatal period. Imprinted genes are important regulators of lung growth, and may be subjected to faulty reprogramming. In the present study, first, microsatellite analysis, PCR, and DNA sequence identification were conducted to confirm that these three dead kids were genetically identical to the transgenic donor cells. Second, the CpG island methylation profile of the imprinted insulin-like growth factor receptor (*IGF2R*) gene was assessed in the lungs of the three dead transgenic kids and the normally produced kids using bisulfite sequencing PCR. In addition, the relative mRNA level of

IGF2R was also determined by real-time PCR. Results showed that the *IGF2R* gene in the lungs of the dead cloned kids showed abnormal hypermethylation and higher mRNA expression levels than the control, indicating that aberrant DNA methylation reprogramming is one of the important factors in the death of transgenic cloned animals.

Key words: Transgenic cloned dairy goats; Lung; DNA methylation; *IGF2R*; mRNA expression