

Gene Expression Profiling to Discover Genes Controlling Feed Intake in Dairy Cattle

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An experiment was conducted with five Holstein cattle fitted with duodenal cannulae to identify and characterize gastrointestinal genes that are differentially expressed during conditions of hunger or satiety. Intestinal tissue samples were obtained from the duodenum, approximately 50-cm distal of the abomasal sphincter, using an endoscopy technique, at five time points during fasting (48 h) and five time points during refeeding (24 h). Gastrointestinal genes that were differentially expressed over the course of fasting and refeeding were identified using gene expression profiling technology (differential display using adapter-specific oligonucleotide primers with 3'-end restriction enzyme fragments of cDNA). Validation of the procedure was based on theoretical prediction of the differential display primer pair that would target sequences corresponding to the bovine cholecystokinin (CCK) gene; we are the first group to have cloned the bovine CCK gene. The primers produced a cDNA of the predicted size, which was identified based on differential expression as detected by polyacrylamide gel electrophoresis and autoradiography. The cDNA was isolated, cloned, sequenced, and confirmed to correspond to the sequence of the bovine CCK gene. The same pattern of differential gene expression was also confirmed in a ribonuclease protection assay using antisense riboprobes generated from the expressed gene (CCK) clone.

The method that we have developed for gene expression profiling produces highly consistent and reproducible gene expression patterns. It also minimizes the redundancy of expressed genes, which has allowed us to develop a unique collection of cDNA fragments (expressed gene clones) corresponding to candidate genes that exhibit differential expression in our model of fasting and refeeding. Our study demonstrates the validity and effectiveness of gene expression profiling technology as a systematic approach for the discovery of novel genes that encode proteins with key roles in regulating hunger and satiety in dairy cattle.

Implications of the Research for the Dairy Industry: The development of these leading-edge techniques in genetic technology was accomplished through efforts to continue advancing the molecular component of our dairy research program. In addition to providing new knowledge about regulation of feed intake in dairy cattle, another important outcome of this advance is the creation of new opportunity for novel approaches to address and provide solutions to current issues facing the industry.