

Building a cattle DNA database in Japan



Over the last decade, the Livestock Improvement Association of Japan (LIAJ) has developed a DNA and beef sample database capable of parentage discrimination analysis for pedigree registration and individual identification of every head of cattle in Japan. To handle the large number of samples involved, scientists at LIAJ have chosen Tecan's Freedom EVO® 200 workstation, with barcoded sample identification, for the automated extraction of DNA.

Since its establishment in 1965, LIAJ has developed its activities, with the improvement of cattle as the primary objective. Today, the association continues to work on livestock improvement, using various methodologies to support the production of superior breeding stock and to develop efficient methods for the individual identification of animals. The Maebashi Institute of Animal Science, LIAJ's livestock improvement technology laboratory in Gunma, Japan, was opened in 1977 to carry out blood typing and artificial fertilization research in cattle. The institute has since expanded to handle embryo transplantation, sperm sorting, as well as all DNA-based testing such as parentage testing, testing for hereditary diseases and individual identification. In addition to the various inspection businesses, the Maebashi Institute is involved in research work, including quantitative trait loci (QTL) analysis of Japanese varieties of cattle using DNA markers, in collaboration with other research institutions.

As a technical scientist in the Division of Animal Genetics at the Maebashi Institute, Dr Kazuhito Kurogi is in charge of the individual identification and parentage discrimination analysis. Dr Kurogi explained: "In Japan, a blood typing-based cattle identification database was created in the 1970s, which LIAJ took over from the Japan Holstein Registration Association in 1978. The work to create a DNA-based database for cattle

started in 1997 and we started DNA-based parentage testing in July 2000. By October 2004, DNA-based parentage testing completely superseded the blood typing system and the DNA database is now fully operational, with all testing carried out using DNA."

The cattle DNA database is used for various objectives like monitoring registration and parentage testing, verifying biotechnology techniques such as artificial fertilization and embryo transplantation, and for meat traceability.

Meat traceability is one of several recently implemented government initiatives to win back consumer confidence following the discovery of a bovine spongiform encephalopathy (BSE)-infected cow in Japan in 2001. All steps in the movement of beef in Japan are subject to traceability testing, from birth to slaughter, storage,

and sale at supermarkets, butchers, and even steakhouses and specialist restaurants which handle cuts of beef for final preparation at the diners' table. The new traceability legislation has been actively enforced since December 2004.

Dr Kurogi elaborated: "In Japan, each animal is identified by a unique 10-digit number. The cattle wear ear tags bearing this number, which is also used as a key for DNA testing. Each pack of dressed meat is labeled with the same 10-digit number, and anybody can find out the past history of each animal from birth to slaughter using this number. This information is readily available to consumers through in-store computers as well as on the internet through the National Livestock Breeding Center's homepage."

"The Ministry of Agriculture, Forestry and Fisheries of Japan has entrusted LIAJ with the task of storing beef samples taken after slaughter, as well as individual identification testing of the stored samples and samples collected from retailers. By carrying out the DNA testing we verify that the traceability system is functioning precisely as specified. For this testing, the cattle DNA analysis technology is essential for individual identification."



The Division of Animal Genetics at the Maebashi Institute of Animal Science, LIAJ.

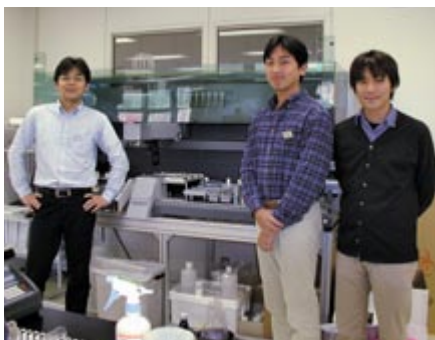


In response to the recent and rapid increase in the number of samples handled for registration and testing, in 2004 Dr Kurogi decided to automate the DNA extraction process using Tecan's Freedom EVO 200 workstation with an integrated robotic manipulator (RoMa) arm, 8-channel liquid handling arm (LiHa) and a PosID™ barcode reader. Dr Kurogi uses the Freedom EVO with its integrated Te-MagS™ magnetic bead separation module to extract and purify DNA from hair root, blood and tissue samples. "We operate the Freedom EVO all day, almost every day. Our extraction program takes approximately two hours to process one plate of 96 samples, so it is possible to handle approximately four plates – 384 samples – in a working day. It is also possible to operate the Freedom EVO unattended so, when we need to process more than four plates in a day, the workstation is run overnight. At present, we are extracting DNA from approximately 75,000 samples in a year, and the cattle DNA database is updated daily."

"We have optimized and standardized our programs so that we can extract clean DNA from blood and hair roots using essentially the same method. It is very easy to use and all operations can be carried out by one person, simply by pressing buttons on a PC. We just line up the samples in the morning and leave the workstation to run for the whole day, only stopping briefly to make slight adjustments to the program if different sample types are being processed. We are now making efforts to shorten the process time by simplifying the program without compromising the quality of results."

"The Freedom EVO has saved labor and time by automating the DNA extraction procedures and the complete barcode management is extremely useful for us, eliminating human error. In a testing facility such as ours, the thoroughness of sample management is critical so the PosID barcode identification is particularly important."

The flexibility of the Freedom EVO is allowing the scientists to automate other laboratory tasks. Dr Kurogi concluded: "Currently, we are continuing to transfer the tasks which were previously performed manually to Tecan's workstation. The possibility to expand the application in response to increased workload in the future, of course, is another important characteristic that attracted us to the Freedom EVO."



Dr Kazuhito Kurogi, Dr Tomohito Ito and Mr Yusuke Kozono with the Freedom EVO workstation.



The Freedom EVO workstation at the Maebashi Institute of Animal Science, LIAJ.