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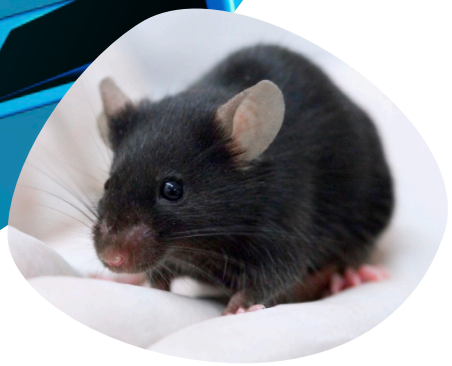

Customer case study

The Jackson Laboratory – mouse genomics

The importance of mouse models

The laboratory mouse is a powerful system for mammalian genetic and biomedical research. Among the many advantages to using the mouse as a model organism, the most important is their striking similarity to humans in anatomy, physiology and genetics. Over 95% of the mouse genome is similar to the human genome, making it particularly applicable to human disease. The physiology shared between mice and humans not only makes the mouse ideal for modelling complex human diseases but also for drug efficacy testing.
The natural variation among inbred strains of mice (species which are nearly identical to each other in genotype due to long inbreeding) provides an essential system to study complex diseases involving the interaction of multiple genes. Inbred strains of animals are frequently used in laboratories for experiments where the reproducibility of conclusions is important and so all the test animals should be as similar as possible.
As many of the genes responsible for numerous complex diseases are shared between mice and humans, research in mice is crucial for the identification of genetic risk factors in the human population. Naturally occurring, spontaneous mutations also often cause afflictions in mice that mimic similar human genetic diseases.

Mouse research has lead to major advances in the ability to treat a number of serious human diseases and conditions. Today, mice provide effective models for conditions such as atherosclerosis, hypertension, diabetes, osteoporosis, glaucoma, neurological and neuromuscular disorders, and cancer, as well as many rare diseases.

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GENOMIC ANALYSIS BY LGC

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Industry challenge: complex human disorders

The ability to directly manipulate the mouse genome provides a powerful tool to model diseases for which the causative gene is known. For example, the manipulation of genes involved in human cancer has allowed for the creation of hundreds of mouse models of cancer, enhancing the search for effective treatments against many different types of neoplasia (new, abnormal growth of tissue).

However, there is an imminent need to improve the genetic understanding of mouse models so that the data and knowledge they generate can be accurately applied to humans.

Researchers are using mouse models to conduct rigorous research into the genetics and pathophysiology of human diseases and other traits determined by either single genes or sets of multiple loci. Two recent advancements have substantially improved these capabilities. First, maps constructed with a high density of simple sequence length polymorphism (SSLP) markers and expressed sequence tag (EST) loci (Dietrich *et al.* 1996; Rowe *et al.* 2003) greatly improved the process of identifying candidate genes. Second, the genome of the completely sequenced strain C57BL/6J (Waterston *et al.* 2002) with sequences from other mouse strains revealed an abundance of single nucleotide polymorphisms (SNPs), one of the most common types of genetic variation.

Technical challenge

SSLP markers allow for the analysis of one locus per experiment, permitting allelic variations of individual loci to be distinguished. As a result, linkage groups between different genetic maps can be identified. The downside is that it is necessary to know the sequence, which is expensive and labour intensive. The great abundance and lower costs of assaying SNPs offers substantial advantages over the use of SSLPs in genetic mapping. SNP markers facilitate genome-wide scans and comparisons even between closely related species and in all possible crosses.

Several groups have described tens of thousands of SNPs each in up to 15 mouse strains (Grupe *et al.* 2001), (Wade *et al.* 2002), (Wiltshire *et al.* 2003) In doing so, these groups have laid the foundation for SNP genotyping in the mouse. The increased interest in SNPs is reflected by the development of a diverse range of SNP genotyping technologies. Mouse genotyping and scanning requires a technology that can provide flexibility as well as high resolution to detect polymorphisms.

Dr. P. Petkov from The Jackson Laboratory comments,

"Our aim is to find technologies that are cost effective, flexible, and easily customisable, and can be employed to scan and genotype mice in order to find practical solutions to improve human health. SSLPs used in genetic mapping are quite expensive and labour-intensive when compared to SNPs."

The solution

To further the genetic mapping of mice, scientists at The Jackson Laboratory identified a robust set of SNP markers, sufficiently polymorphic to perform quantitative genome scans and trait locus (QTL) analyses between nearly any two mouse strains, including virtually all of the inbred and wild-derived inbred strains available from The Jackson Laboratory, and demonstrated their effectiveness.

Following this breakthrough research, a team of scientists led by Dr. P. Petkov has used this technology to investigate genetic recombination during meiosis.

Recombination is an essential part of meiosis (cell division that results in two new cells each with half the chromosome number of the parent). It ensures the correct segregation of chromosomes and provides contact and exchange of genetic material in order to generate genetic diversity between offspring. In humans and mice, recombination events are located at preferential, highly active sites termed hotspots, whose placement and activity are tightly regulated. The researchers were interested to learn more about the factors controlling the location and relative activity of mammalian recombination hotspots.

For this particular study, The Jackson Laboratory required a genotyping technology that would offer accuracy and performance. The research team selected KASP® genotyping technology from LGC due to its flexibility in assay design, excellent SNP genotyping performance and intelligent cost saving.

Dr. P. Petkov comments,

"LGC's KASP genotyping chemistry was our preferred choice because it not only provided cost benefits but it also contributed greatly in achieving flexibility in designing assays required for genotyping SNPs."

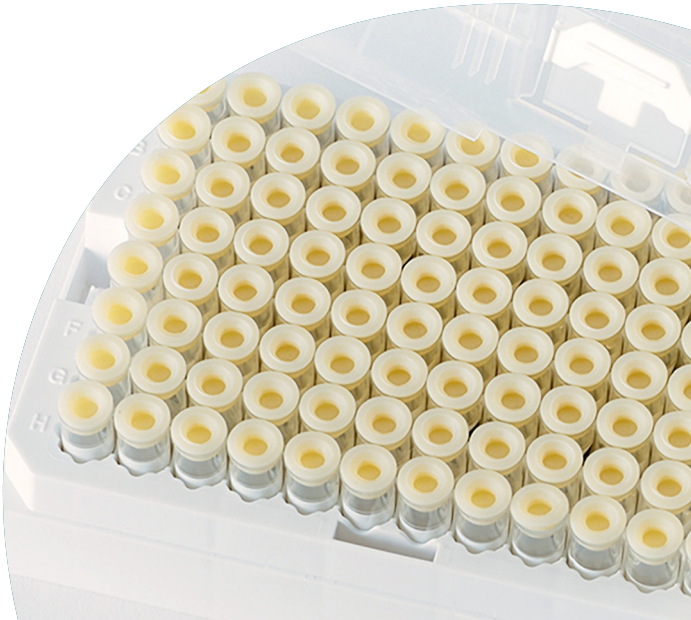
Results

The Jackson Laboratory used LGC's KASP genotyping technology to facilitate a key part of their study. Phenotyped animals from their controlled genetic crosses were genotyped with 165 SNP markers spaced across the genome, at 20-Mb resolution, to provide high-resolution genetic mapping.

The researchers have identified a hotspot-regulating locus gene, Rcr1, which simultaneously controls the locations of multiple hotspots. Subsequently, they identified the gene within this locus, Prdm9, which indicates the existence of a newly emerging class of genes important in recombination.

Dr. P. Petkov explains,

"The advancement in genotyping technology has allowed us to design compatible assays to narrow down the genomic region and identify the gene responsible for hotspot regulation.The discovery of Prdm9 will lead to further improvements in our research to gain insight on its functions. KASP has been a valuable tool in reaching those meaningful conclusions and we are confident that it will continue to support us in better understanding of genetic factors underlying human fertility and evolution."



Why KASP

KASP genotyping chemistry was the preferred choice for The Jackson Laboratory because the technology offers tremendous flexibility to researchers in designing assays that yields a higher success rate. Cost is the major concern for any researcher; LGC's KASP genotyping assay provides a major cost breakthrough due to utilisation of a unique and universal competitive allele-specific PCR. The technology does not require dual-labelled probes and completely automated processes provide maximum levels of accuracy through which the laboratory has achieved noticeable improvements in genome mapping.

Future

There is a need to fully understand the underlying mechanisms of complex human diseases, and it is therefore important to determine the function of genes and the elements that control disease-associated genes throughout the human genome and in the genomes of model organisms like the mouse.

Genetic scanning and monitoring of mouse models using panels of SNP markers is now widely used by researchers to confirm strain identity, monitor genetic quality and elucidate strain relationship. With the use of KASP technology, there is a significant increase in the accuracy of information related to genome scanning as well as mouse genotyping, and greater accessibility now for more researchers to use this technology.

The technology provides a greater degree of freedom in assay design, giving a higher design success rate of >95% for KASP assays. It also gives flexibility in terms of conducting low-, medium- and high-throughput studies and individual repeat assays. In addition to this, the technology supports broad liquid handling, thermal cycling, and fluorescent reader compatibility.

The Jackson Laboratory in a joint effort with LGC has developed 1638 working assays for pre-validated mouse SNPs. These assays provide a powerful tool for the mapping of traits within mouse disease models and breeding populations.

Dr. P. Petkov says,

"Accuracy of the data lays the foundation for correct analysis and conclusions. We achieved 96% of success rate with the use of KASP assays. We have developed more than 2100 working assays in collaboration with LGC and we will continue to work closely to harness the potential of the technology in our research."

References

Dietrich, W. F., *et al.* 1996. A comprehensive genetic map of the mouse genome. *Nature* 380: 149-152.

Grupe, A., *et al.* 2001. In silico mapping of complex disease-related traits in mice. *Science* 292: 1915-1918.

Parvanov, E. D., *et al.* 2009. Trans-regulation of mouse meiotic recombination hotspots by *Rcr1*. PLoS Biol 7: 340-349.

Parvanov, E.D., Petkov, P. M., and Paigen, K. 2010. Prdm9 controls activation of mammalian recombination hotspots. *Science* 327: 835.

Rowe, L.B., *et al.* 2003. The comprehensive mouse radiation hybrid map densely cross-referenced to the recombination map: A tool to support the sequence assemblies. *Genome Res.* 13: 122-133.

Wade, C.M., *et al.* 2002. The mosaic structure of variation in the laboratory mouse genome. *Nature* 420: 574-578.

Waterston, R.H., *et al.* 2002. Initial sequencing and comparative analysis of the mouse genome. *Nature* 420: 520-562.

Wiltshire, T., *et al.* 2003. Genome-wide single-nucleotide polymorphism analysis defines haplotype patterns in mouse. *Proc. Natl. Acad. Sci.* 100: 3380-3385.

The Jackson Laboratory

The Jackson Laboratory is an independent, non-profit organisation focusing on mammalian genetics research to advance human health. The laboratory aims to discover precise genomic solutions for disease and empower the global biomedical community in the shared quest to improve human health.

Using the mouse as a model organism, The Jackson Laboratory conducts research in a range of areas including, but not limited to, aging, bioinformatics, cancer, cardiovascular, genomics, metabolism and neurobiology. The state of the art Genome science services department facilitates SNP-based Genome Scanning Services for researchers that breed animals in their home facility. The service can be used to assess certain strain mixtures, to facilitate marker-assisted breeder selection (for constructing congenic lines), to map new mutations, or to detect/measure recent strain contamination. In addition to this, they offer a Mouse Diversity Genotyping Array service that utilises an innovative genotyping microarray which was designed for high-density, genome-wide profiling of SNPs.

Along with it's research, The Jackson Laboratory provides scientific resources, techniques, software and data to scientists around the world. The Laboratory also breeds and manages colonies of mice to supply other research institutions and laboratories.

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