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Gene variant activity is surprisingly variable between tissues

First comprehensive analysis of the "allelome" shows unexpected, tissue-specific differences in gene variant activity

Every tissue has its own pattern of active alleles (the gene variants inherited from the mother or father), a large-scale study led by an international team of scientists at the CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences has found. For the first time, the distribution of all active alleles in different tissues has been investigated. The scientists of the former group of Denise Barlow were able to show that the differential allele activity is regulated by tissue-specific, regulatory DNA elements known as enhancers – a process that could also be involved in many diseases. The results were published in the high-profile open access journal *eLife*.

(Vienna, the 17.08.2017) Every gene in (almost) every cell of the body is present in two variants – so called alleles: one is deriving from the mother, the other one from the father. In most cases both alleles are active and transcribed by the cells into an RNA message. However, for a few genes, only one allele is expressed, while the other one is silenced. The decision whether the maternal or the paternal version is shut down occurs early in embryonic development – one reason, why for long it was thought that the pattern of active alleles is nearly homogeneous in the various tissues of the organism.

The new study (DOI:10.7554/eLife.25125), where CeMM PhD Student Daniel Andergassen is first author (now a PostDoc at Harvard University), uncovers a different picture. By performing the first comprehensive analysis of all active alleles in 23 different tissues and developmental stages of mice, the team of scientists revealed that each tissue showed a specific distribution of active alleles.

For their experiments, the researchers created hybrids of two genetically distinct mouse strains with a fully sequenced genome, allowing gene variants to be clearly assigned to the maternal or paternal allele. To facilitate the analysis, the team developed a user-friendly program called Allelome.PRO, that can easily be applied to similar datasets in mice and other species, a valuable tool for the community to investigate regulation of allele activity. By using this tool to analyze their data the scientists were able to catalogue active alleles in a comprehensive set of mouse tissues, or the mouse "Allelome", and gain an insight into how this differential gene activity is regulated.

The scientists found that both genetic and epigenetic differences between the maternal and paternal allele contributed to the observed tissue-specific activity patterns. "Our results indicate that a large part of those patterns are induced by so-called 'enhancers'", co-senior author Quanah Hudson, now at IMBA (Institute for molecular Biotechnology of the Austrian Academy of Sciences) explains. "Enhancers are DNA regions that are often located at quite some distance from the observed allele, but nevertheless have a direct influence on their activity".

"This study reveals for the first time a comprehensive picture of all active alleles in different tissues – we have uncovered the first complete allelome" Florian Pauler, now at ISTA (Institute of Science and Technology Austria) and co-senior author, adds. "This is not only valuable to understand basic biological functions, but will also help investigating diseases that involve defective gene regulators."

Some of the genes that contributed to the tissue-specific activity patterns were located on the X chromosome and escaped so-called "X-chromosome inactivation", where one of the two X chromosomes in females gets shut down. Previously it was reported that around 3% of X-chromosomal genes in mice and 15% in humans escape inactivation. However, this study revealed that mice are more similar to humans than previously thought, with an average of around 10% of active genes escaping X-inactivation per tissue. By examining a broad range of organs the researchers showed that the number of escapers varies dramatically between tissues. Most strikingly, muscle showed a surprisingly high rate of escapers, with over 50% of active genes escaping X chromosome activation, a result that may be relevant to some diseases of the muscle.

Finally, the allelome offers a near complete picture of "genomic imprinting", the process that leads to epigenetic silencing of either the maternal or paternal allele that is initiated by an epigenetic mark placed in either the egg or sperm. Previously, it was reported that approximately 100 genes can be subject to imprinted silencing – but in many cases, the tissue specificity was not known. This study led to the discovery of 18 new imprinted genes, validated some known genes and resolved the disputed status of some others to provide a gold standard list of 93 imprinted genes in mouse. The scientists found that those new genes were located near to other imprinted genes, indicating that they were co–regulated. Interestingly, this study demonstrated that Igfr2, the first imprinted gene discovered by Denise Barlow in 1991, is surrounding by a large cluster of imprinted genes that extend over 10% of the chromosome, making it the largest co-regulated domain in the genome outside of the X chromosome. Fittingly, after her lab found the first imprinted gene, and discovered the first imprinted non-coding RNA shown to control imprinted silencing. Giulio Superti-Furga congratulates Denise Barlow who recently went into retirement to her great scientific achievements and for revealing the full picture of imprinted genes in the mouse.

Attached pictures: 1) Artistic representation of the mouse allelome 2) The lead authors of the study: Quanah Hudson, Daniel Andergassen, Denise Barlow and Florian Pauler (f.l.t.r.)

The study "Mapping the mouse Allelome reveals tissue-specific regulation of allelic expression" was published in *eLife* on August 14, 2017. DOI: DOI:10.7554/eLife.25125

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Denise Barlow joined CeMM in 2003 and is an Honorary Professor at the University of Vienna. She was initially trained as a State Registered Nurse in the UK and afterwards completed her undergraduate studies at Reading University (UK) and did a Ph.D. on the interferon system in early mouse development at Warwick University (UK). Postdoctoral work studying mouse embryonic development followed at ICRF (London, UK) with Dr. Brigid Hogan, and on genome biology at EMBL (Heidelberg, D) with Dr. Hans Lehrach. She has also held group leader positions at the IMP (Vienna, A) and the NKI (Amsterdam, NL). On returning to Austria in 2000, Denise was appointed Head of the Dept. of Developmental Genetics at the Austrian Academy IMB Institute (Salzburg, A), and then returned to Vienna in 2003 as a Principal Investigator with CeMM. One of the Barlow lab's major achievements was the discovery in 1991 of the first imprinted gene in mammals to show parental-specific gene expression. Her group's subsequent identification that epigenetic silencing of this imprinted gene is induced by expression of an unusual long non-coding (lnc) RNA has led them to investigate how lncRNAs act throughout the mouse and human genome as regulators of gene expression in development and disease. The lab uses the model of genomic imprinting to dissect how lncRNAs epigenetically silence genes, and together with high throughput sequencing technology is extending these results into the mouse and human genome with the goal of understanding how lncRNAs play a role in human diseases such as cancer. Denise Barlow retired in 2015 and closed her lab. Denise Barlow is a recipient of numerous awards and honours including the Erwin Schrödinger Award, the EMBL Alumni Association Austrian Chapter Achievement Award Medal and a honorary professorship of genetics at University of Vienna. She has been an EMBO member since 1995.

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minded basic life science research in a large and vibrant hospital environment of outstanding medical tradition and practice. CeMM's research is based on post-genomic technologies and focuses on societally important diseases, such as immune disorders and infections, cancer and metabolic disorders. CeMM operates in a unique mode of super-cooperation, connecting biology with medicine, experiments with computation, discovery with translation, and science with society and the arts. The goal of CeMM is to pioneer the science that nurtures the precise, personalized, predictive and preventive medicine of the future. CeMM trains a modern blend of biomedical scientists and is located at the campus of the General Hospital and the Medical University of Vienna.
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