HUMAN GENETICS

Mitochondrial variation linked to type 2 diabetes

Recent years have seen the emergence of circumstantial evidence for a contribution of sequence variation in the mitochondrial genome to the risk for several metabolic disorders, including type 2 diabetes. A new study provides the first direct evidence that links mitochondrial genomic variation to inherited variation in the risk for type 2 diabetes in rats.

Proving a direct link between mitochondrial sequence variation and disease is not easy. For example, although maternal inheritance might be suggestive, other phenomena such as maternal environmental effects or imprinting can have the same effect. And separating the effects of mitochondrial and nuclear sequence variation has not been easy either. But this is exactly what Pravenec *et al.* achieved. As a result of several rounds of backcrossing of highly inbred spontaneously hypertensive rats with another highly inbred strain, the Brown Norway rat, the authors created conplastic strains that had two different mitochondrial genomes on a virtually identical nuclear genetic background. The spontaneously hypertensive rat strains were chosen because, in the past, this genetic background proved useful for detecting the effects of DNA variants on glucose and lipid metabolism.

Careful measurements of glucose and insulin levels, skeletal muscle glycogen content and ATP levels revealed significant differences between these strains. Sequence analysis of the two mitochondrial genomes identified a number of polymorphisms, including coding SNPs in several genes that encode proteins involved in important mitochondrial functions, such as oxidative phosphorylation. Comparing the coding SNPs with the corresponding residues in several vertebrate mitochondrial genomes revealed that they are relatively highly conserved; analysis of mitochondrial enzyme activities in these strains confirmed their functional significance. Although the relative contributions of each of the mitochondrial DNA (mtDNA) variants on glucose metabolism remains to

GENOME EVOLUTION

A lateral take on eukaryotic inheritance

A new study of *Wolbachia*, an abundant intracellular bacterium that infects more than one-fifth of insects, as well as other eukaryotes, challenges our conventional view of genome evolution. Scientists at the University of Rochester, USA, and the J. Craig Venter Institute, USA, led by Jack Werren (Rochester), report that genes from this bacterium — and in some cases almost its entire genome — have found their way into the nuclear DNA of its eukaryotic hosts, where they are stably inherited.

Prokaryotes are notorious for being generous with their DNA, which is exchanged with relative ease between cells and species by lateral gene transfer. But multicellular eukaryotes have always been more conservative, relying largely on mutations to acquire new gene functions. The fact that *Wolbachia* inhabits the germ line of its eukaryotic hosts and is transmitted (maternally) by them led these authors to enquire whether any endosymbiont DNA has been transferred to the host genome where it would be inherited from generation to generation.

The team compared the genome sequence of *Wolbachia pipientis* to those of 26 arthropod and nematode hosts for which whole-genome sequences are available. They found that DNA ranging from short sequences of under 500 base pairs through to nearly its entire 1 Mb genome are present in eight (~31%) of the hosts, including insects and nematodes. Sequence matches between *Wolbachia* and the candidate host genomes were experimentally verified by PCR in five hosts. The case of the tropical fruitfly *Drosophila ananassae* was the most extreme, with almost the entire *W. pipientis* genome having been inserted into an autosome of the host.

The cosy relationship between the DNA of the endosymbiont and that of its hosts was not merely transient; when the host organisms were 'cured' of the endosymbiont (by treatment with antibiotics) *Wolbachia* DNA was still inherited like 'normal' genes in the insect genome. Not only is it inherited, but 2% of *Wolbachia* genes were expressed in cured fruitflies. Whether this transcription is meaningful is difficult to tell, but at least one mRNA is capped by the eukaryotic transcription machinery.

> "…these findings open up the possibility that eukaryotic organisms might derive some new gene functions by picking them up directly from their intracellular inhabitants."

Apart from the relevance of this gene mobility for studies of evolution, its occurrence is an eye opener for sequencers of eukaryotic genomes, who often dismissed the presence of bacterial sequences as contamination.

Overall, these findings open up the possibility that eukaryotic organisms might derive some new gene functions by picking them up directly from their intracellular inhabitants. It will be interesting to find out why the bacterial sequences are allowed into the genome in the first place. Their presence raises questions about the function and possible selective advantages to the host — finding out whether the transcribed genes are functional would be a first step in addressing this matter. And *Wolbachia* genes might not be alone in delving into the host genome, given the number of other bacteria residing in invertebrate germline cells that could be involved in similar activities.

Tanita Casci

ORIGINAL RESEARCH PAPER
Dunning Hotopp, J. C., Clark, M. E. *et al.* Widespread lateral gene transfer from intracellular bacteria to multicellular eukaryotes. *Science* 30 August 2007 (doi 10.1126/science.1142490)

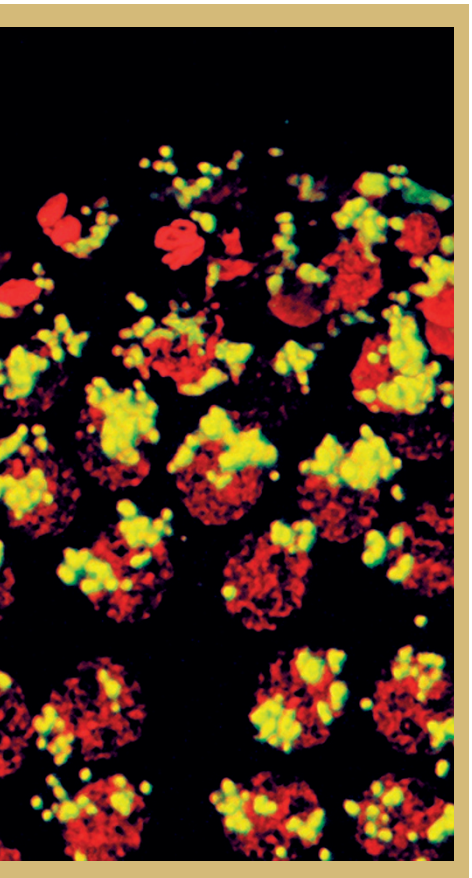
WEB SITE
Jack Werren's laboratory homepage:
http://www.rochester.edu/college/bio/labs/WerrenLab/

be determined, these results provide a number of plausible links between mtDNA variation and functional differences that might underlie the condition.

Not only does this work demonstrate that naturally occurring variation in mitochondrial genomic sequence can ultimately lead to pathogenesis underlying common diseases, it also establishes an important animal model for studying the contribution of mtDNA variation to glucose metabolism.

Magdalena Skipper

ORIGINAL RESEARCH PAPER Pravenec, M. *et al.* Direct link of mitochondrial genome variation to risk factors for type 2 diabetes in conplastic strains. *Genome Res.* 14 August 2007 (doi:10.1101/gr.6548207)
FURTHER READING Frayling, M. Genome-wide association studies provide new insights into type 2 diabetes aetiology. *Nature Rev. Genet.* **8**, 657–662 (2007) | Taylor, R. W. & Turnbull, D. M. Mitochondrial DNA mutations in human disease. *Nature Rev. Genet.* **6**, 389–402 (2005)



Wolbachia (yellow) associated with nuclei (red) in the posterior end of a *Drosophila simulans* embryo (germline pole cell nuclei at the top of the image and somatic nuclei at the bottom). Image courtesy of M.Clark, University of Rochester, New York, USA.

GENETIC INSTABILITY

Genomic instability links diet to cancer

A genetic screen in yeast has found a surprising link between vitamin B6 deficiency and increased genome instability, a hallmark of cancerous cells. This study gives new insights into the causes of genetic aberrations, and provides a possible mechanistic explanation for epidemiological evidence that suggests a link between micronutrient deficiencies and cancer risk.

When faithful DNA replication is disrupted, genetic lesions are created. If the DNA-repair pathways that correct these lesions are faulty, illegitimate repair can result in gross chromosomal rearrangements (GCRs) such as translocations, amplifications, inversions and deletions. In particular, break-induced replication (BIR), a type of homologous recombination repair, is thought to be a major mechanism by which GCRs occur.

Kanellis and colleagues engineered a *Saccharomyces cerevisiae* strain that includes a GCR reporter located at a chromosomal position at which rearrangements take place by BIR. In a genome-wide screen, the authors picked out *BUD16* as a potent suppressor of GCR, and alignment studies revealed that this gene encodes a putative pyridoxal kinase (Pdxk), an enzyme that is crucial for the metabolism of vitamin B6 to produce pyridoxal 5′ phosphate (PLP), the biologically active form. Strikingly, *bud16*Δ mutant cells have a 124-fold increased GCR rate compared with wild-type cells, coinciding with a reduction in PLP levels to 1.8%.

Rad52 is essential for homologous recombination and hence DNA repair. *bud16*Δ *rad52*Δ double mutants showed synthetic sickness and poor viability, suggesting that *bud16*Δ cells have high levels of DNA disruption during replication, and rely on Rad52-mediated repair for survival. Furthermore, fluorescence microscopy showed that, after budding, Rad52 foci (or 'repair centres') were present in 57–75% of *bud16*Δ cells, compared with 2–21% in wild-type cells.

But do these symptoms of replication stress necessarily result from PLP depletion? By interfering with components of the PLP pathway, the authors generated a similar GCR phenotype to that of *bud16*Δ cells. Similar results were seen in mammalian cells: addition of a vitamin B6 analogue that inhibits PDXK in human cells (thereby reducing PLP levels) resulted in the induction of DNA lesions and activation of the DNA-damage response.

PLP is an essential cofactor in dTMP synthesis pathways; might this be the mechanism by which it prevents DNA lesions? *bud16*Δ cells have significantly higher uracil levels in their DNA compared with wild-type cells. It seems likely that the *bud16*Δ mutation, by disrupting Pdxk's role in nucleotide synthesis, causes an increase in dUMP pools, promoting incorporation of dUTP into DNA during replication. Uracil-excision processes might then promote the occurrence of lesions, leading to chromosomal instability.

Interestingly, the authors go on to propose that the cellular response to low PLP could actually represent a defense mechanism against cancer — depletion of metabolites resulting from overproliferation of cancerous cells might be sensed by the cells as replication stress, activating damage-response pathways to bring about senescence.

Carrie Patis

ORIGINAL RESEARCH PAPER Kanellis, P. *et al.* A screen for suppressors of gross chromosomal rearrangements identifies a conserved role for PLP in preventing DNA lesions. *PLoS Genet.* **3**, e134 (2007)