IN THE NEWS

Controlling genetic bias

Sir John Sulston, Nobel-Prize winning leader of the Human Genome Project, has proposed that tough laws be introduced to prevent genetic discrimination.

Professor Sulston, from the Sanger Institute, and his co-authors, Professors John Harris and Simona Giordana, both at Manchester University, UK, circulated their proposal to the UK's Human Genetics Commission (HGC) in May 2004.

"What we have to establish, right across the board, is the right for people to be treated equally, regardless of their genetic make-up. We can't keep fudging the issue", said Professor Sulston (*The Guardian*).

Sulston's main concern is that the information generated from genetic tests could be misused (*Reuters*). "People are right to be leery about having genetic tests until we have solid laws in place to protect their rights", he said (*The Daily Times, Pakistan*).

It is still unclear whether the HGC, which advises the UK government on genetics, will back this proposal from three of its members. However, the proposal has already received some support from one prominent UK non-governmental organization.

"If you have a genetic test that predicts you might get ill in the future, currently your employer could refuse you a job or restrict your pension rights. That's something that should definitely be prevented by legislation", said Helen Wallace, deputy director of Genewatch (*The Guardian*).

Predictably, insurers were less enthusiastic about a new law. "It is contrary to the way insurance operates to the benefit of everybody", said Malcolm Tarling of the Association of British Insurers.

However, Sulston is quite clear about what is required: "We need to get something on the books in the next five years", he said (*The Guardian*).

Nick Campbell

GENE EXPRESSION

A disruptive influence

There are LINE-1 (L1) retrotransposons in most mammalian genes, but these are mainly found in the introns, so it was assumed that they rarely influenced gene expression. However, new data show that these elements of selfish DNA can in fact affect transcription of endogenous genes. So, rather than just being quiet non-contributors, these new findings paint a picture of these elements as the disruptive naughty schoolchildren of the genomic classroom.

Despite L1's abundance in mammalian genomes, L1 trancripts have been difficult to detect. Given the succesful expression of L1 in non-mammalian systems, this difficulty indicated that there might be a mammal-specific mechanism for suppressing L1 transcription. To investigate this possibility, Jeff Han and colleagues fused one of the two human L1 open reading frames (ORF2) to a GFP ORF and then measured levels of RNA expression relative to a control *lacZ*/GFP fusion. They found that the ORF2 sequences caused a large decrease in gene expression, whether they were in sense (GFPORF2) or antisense (GFPORF2AS) orientation.

Interestingly, the authors did find a difference between sense and antisense constructs in the way that they achieved this transcriptional downregulation. Cloning and sequencing of the abundant lower molecular mass species that is expressed when ORF2 is in the antisense orientation indicated that premature polyadenylation accounted for the shortfall in full-length transcripts. By contrast, premature polyadenylation only accounted for 15% of the decrease in full-length transcripts from GFPORF2.

Measurement of the RNA half life and real-time PCR showed that an increase in RNA degradation could not account for the reduction in GFPORF2 full-length transcripts. Neither did ORF2 inhibit transcriptional initiation, because a nuclear run-on assay indicated that the polymerase density in the early region of GFPORF2 transcripts was equivalent to that of the *lacZ* control. However, the same assay revealed that polymerase density decreased as it was assayed further along the ORF2 sequence: so, ORF2 seems to suppress gene expression by inhibiting transcriptional elongation.

DEVELOPMENTAL BIOLOGY

Talking about regeneration

Our germ line is the closest we get to immortality, as, for the time being at least, eggs and sperm are our only lasting biological legacies. Two research groups now describe a gene (*Zfp145*) that the stem cells of the testes need to renew themselves, and thereby to ensure a plentiful sperm supply. The *Zfp145* gene is the first of its kind to be characterized in mammals — thanks partly to a mutant mouse that cropped up 50 years ago.

The luxoid (lu) mouse arose in 1950 and was noted for its male-fertility defects. When Robert Braun and colleagues re-examined this phenotype, they found that young, homozygous mutant animals have few sperm and that a gradual loss of germ-line stem cells causes their testes to progressively degenerate. The effect is specific to germ cells, as *lu*/*lu* germ cells cannot colonize the testes of a host that lacks these cells, but *lu*/*lu* somatic tissue supports spermatogenesis from normal transplanted germ cells. Fine recombination mapping of the mutation on chromosome 9, plus some informed guesswork, led the authors to a candidate gene for the luxoid mutant. *Zfp145*, which encodes the PLZF transcriptional repressor, has impeccable credentials as a candidate: as Pier Paolo Pandolfi and colleagues previously showed, males that are mutant for *Zfp145* are sterile; in addition, PLZF is expressed in the testicular stem cells and in the undifferentiated sperm precursors alongside a known stem-cell marker. Importantly, luxoid animals have a nonsense mutation in *Zfp145*, which consequently is not expressed.

Pandolfi and colleagues found similar characteristics in *Zfp145*$^{-/-}$ testes while they were investigating the known involvement of this gene in leukaemogenesis. They showed that mutant testes degenerate not because of low sperm production but, conversely, through sperm over-proliferation followed by germ-cell exhaustion and abnormal checkpoint activation in differentiating cells, resulting in cell death.

Although the two studies do not always agree, they converge on the

Han and colleagues followed up their experiments with a neat bit of bioinformatics. They compared the amount of L1 sequence in the genes with the highest and lowest levels of expression in humans. The results were striking: highly expressed genes contained small amounts of L1 sequence, whereas genes with low levels of expression contained large amounts of L1 sequence. These patterns hold even when differences in total intron content or isochore location are accounted for.

The dovetailing of the authors' experimental and bioinformatic data provides extremely strong circumstantial evidence that L1 insertions decrease gene expression *in vivo*. Moreover, this new work tallies with previous findings that gene expression is suppressed when full-length L1 sequences are inserted into introns.

The authors also raise the intriguing possibility that the mammalian genome might have co-opted transcriptional suppression of L1 (which probably first evolved to prevent excessive mutagenic retrotransposition) as a mechanism for fine-tuning the relative levels of expression of different genes. If this model is correct, perhaps L1 is better viewed as more of a genomic classroom assistant than a problematic pupil?

Nick Campbell

References and links

ORIGINAL RESEARCH PAPER Han, J. S. *et al.* Transcriptional disruption by the L1 retrotransposon and implications for mammalian transcriptomes. *Nature* **429**, 268–274 (2004)

FURTHER READING Han, J. S. & Boeke, J. D. A highly active synthetic mammalian retrotransposon. *Nature* **429**, 314–318 (2004)

WEB SITE

Jef Boeke's laboratory: http://www.bs.jhmi.edu/MBG/boekelab/boeke_lab_homepage

idea that PLZF promotes stem-cell maintenance, possibly through chromatin remodelling. *Zfp145*$^{-/-}$ germ cells would be channelled down the differentiation pathway at the expense of proliferating. Indeed, Pandolfi and co-authors show that *Zfp145*$^{-/-}$ cells do badly at repopulating testes that lack stem cells, probably because so few undifferentiated cells are present.

Given the roles of PLZF in limb development, acute-promyelocytic leukaemia and germ stem-cell differentiation, this protein provides an interesting link between development, cancer biology and stem-cell maintenance.

Tanita Casci

References and links

ORIGINAL RESEARCH PAPERS Buaas, F. W. *et al.* Plzf is required in adult germ cells for stem cell self-renewal. *Nature Genet.* **36**, 647–652 (2004) | Costoya, J. A., Hobbs, R. M. *et al.* Essential role of Plzf in maintenance of spermatogonial stem cells. *Nature Genet.* **36**, 653–659 (2004)

IN BRIEF

EVOLUTION

Noise minimization in eukaryotic gene expression.

Fraser, H. B. *et al. PloS Biol.* 27 Apr 2004 (doi:10.1371/journal.pbio.0020137)

Until now, the functional and evolutionary significance of stochastic ('noisy') fluctuations in the rates of protein production was a mystery. Michael Eisen and colleagues have developed a model of stochastic gene expression in yeast to test their hypothesis that genes that encode subunits of multiprotein complexes or that cause lethality when deleted would be sensitive to such variation. Their estimates of the noise in protein production for most yeast genes confirm this hypothesis and indicate that natural selection minimizes noise in gene expression.

CANCER GENETICS

Impact of the KU80 pathway on NHEJ-induced genome rearrangements on mammalian cells.

Guirouilh-Barbat, J. *et al. Mol. Cell* **14**, 611–623 (2004)

The genomes of cancer cells are often unstable and undergo rearrangements. Non-homologous end joining (NHEJ), a process that joins broken ends of double-strand breaks (DSBs) in DNA, is generally associated with maintaining mammalian genome stability. Josée Guirouilh-Barbat and colleagues now show that NHEJ might occur in two-thirds of DSB repairs. Surprisingly, their data indicate that this pathway often incorporates broken DNA into new locations and so accounts for many genome rearrangements that are seen in cancer cells.

AGEING

Sirt1 promotes fat mobilization in white adipocytes by repressing PPAR-γ.

Picard, F. *et al. Nature* 2 June 2004 (doi:10.1038/nature02583)

Premature ageing in mice expressing defective mitochondrial DNA polymerase.

Trifunovic, A. *et al. Nature* **429**, 417–423 (2004)

Two studies provide separate molecular explanations for the ageing process in mammals. In yeast, the *SIR2* gene is the link between calorie restriction and an extended lifespan. Leonard Guarente and colleagues have now investigated how *Sirt1* (sirtuin 1), the mammalian version of *SIR2*, mediates the same effect in the mouse. Fat reduction extends the mouse lifespan, and the authors have shown that this is caused by the ability of SIRT1 to promote fat mobilization in adipocytes by repressing the fat regulator PPAR-γ during food deprivation. Nils-Göran Larsson and colleagues focussed instead on the mitochondrial DNA (mtDNA), which is known to accumulate mutations with age. Mice that were defective for the mtDNA polymerase PolgA — because they expressed a nuclear knock-in mutation that abolished the proofreading ability of the polymerase — accumulated 3–5 times the number of point mutations, aged prematurely and developed age-related features, such as baldness and reduced fertility. This clever experiment supports the view that mtDNA mutations are the cause rather than the consequence of ageing.