Pivoting on p53

Helen Dell, BMN News

The tumour-suppressor protein p53 can also regulate lifespan in the mouse, report US researchers. It seems to be the pivot to a molecular see-saw, maintaining the cellular balance between tissue regeneration and tumour suppression, they suggest.

A serendipitous discovery

The discovery of this role for p53 was serendipitous, says Heidi Scrable, Associate Professor of Neuroscience at the University of Virginia in Charlottesville (http://www.virginia.edu). 'We set out to make a certain mouse, and we made another one that turned out to be a lot more interesting,' she said.

Scrable was trying to create a mouse with a p53 gene that could be switched on and off so that she could manipulate tumour formation. But the transgenic mice she produced turned out to be particularly unusual. 'The homozygote animals are about half size, and they stay half size,' she said. 'They are perfect miniatures,' she told *BioMedNet News*. 'All the organs are present.'

This puzzling phenotype was caused by the gene they had added to the mice, says Scrable. This turned out to encode a shortened form of p53 (termed deltaN-p53), and although the mice have wild-type p53, they seem to overexpress the shortened form, she says.

By plotting growth curves for the mice and looking at cells derived from them, they concluded that the mini mice have difficulties with cell proliferation, rather than just having small cells.

'This is what you might expect, given that p53 is a cell-cycle regulator,' said Scrable.

More surprises

But the mice had more surprises in store – they appeared to be aging very quickly. 'We started to see these ancient mice at about seven-months old,' said Scrable. 'Their fur begins to look scruffy, their skeletons lose bone density, and ... they look shaky and just generally awful.'

This gave some clues about what might be going on, says Scrable. 'When we saw an aging phenotype ... together with the size deficits, a really big bell went off and told us to start looking at IGF signaling,' she said.

The insulin-like growth factor (IGF) pathway is the only signalling cascade in embryos that is exclusively devoted to growth, she explains. Other 'growth' pathways have additional functions, such as deciding cell fate, that were not disturbed in the transgenic mice, she says. Moreover, studies in the nematode worm, fruit fly and mice have implicated IGF signalling in lifespan.

In cells from the deltaN-p53 mice, the IGF receptor is greatly upregulated, they report in *Genes and Development* [1]. What is more, treating cells from these mice with IGF increased signalling along the entire IGF pathway. This does not occur in wild-type cells, they say. 'We got increased signalling all the way down the pathway, into the nucleus and to transcription,' noted Scrable.

Upregulation of the IGF receptor causes stimulation of the MAP kinase pathway, and this targets p21 – a cell-cycle inhibitor. So when Scrable's team looked at cells from deltaN-p53 mice, they found elevated activity of the MAP kinase pathway, and much greater levels of p21. 'That's when we found what was causing our growth arrest,' she said.

Premature aging

So the molecular explanation for both small size and rapid aging all hinges on p53, she argues. p53 normally suppresses the IGF receptor, but is unable to do so in the presence of deltaN-p53. Upregulation of the IGF pathway causes the aging phenotype, but also stimulates the p21 pathway, resulting in the cell proliferation phenotype, she suggests.

Tissue regeneration is a prerequisite for the relatively long lives that mammals lead, explains Scrable, and this means that certain cells must proliferate when called on. 'But if the cells can't be called upon because p53 is screwed up, then one of the consequences is premature aging, or at least that is what our model would suggest,' she said.

Conversely, cell proliferation must be controlled to avoid tumours, which is where p21 comes in – again regulated by p53. 'It's kind of a see-saw,' said Scrable. 'A trade-off of lifespan for cancer. What we have been able to provide is a molecular pathway linking these changes to the IGF pathway.'

Interesting links

The work impresses Lawrence Donehower, Professor of Molecular and Cellular Biology at Baylor College of Medicine in Houston, Texas (http://www.bcm.tmc.edu). 'It's a real advance,' he said, 'not only showing the phenomenology, but ... also getting at what's going on at the molecular and cellular level.'

Linking p53 with aging, IGF signalling, and MAP kinase signalling is

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very interesting, he says. But p53 does a lot of other things, he adds. 'I would like to consider that perhaps there are other functions of p53 that are playing a role here as well,' he said. 'I would like to see them address these other pathways.'

Scrable has already begun to look at how the inability to regenerate is affecting different systems in the body, starting with the brain. 'It looks like p53 really is a central regulator of lifespan, but it's going to take us some time to figure out what that means exactly if we go system by system in the animal,' she said. 'We certainly have unexpected results.'

Reference

1 Maier, B. *et al.* (2004) Modulation of mammalian life span by the short isoform of p53. *Genes Dev.* 18, 306–319

Unfolding targets for dengue fever

Sadaf Shadan, BMN News

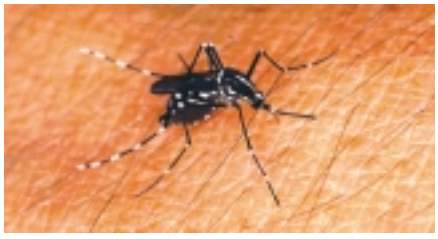
Recent structural data show conformational changes in envelope proteins of the dengue virus during fusion with the host membrane, providing important clues for the development of therapeutic drugs against a number of viral diseases, report researchers.

Fusion proteins

Specific proteins on the membrane of enveloped viruses are categorized into two classes. Class I fusion proteins are found on the surface of HIV and influenza virus among others. These differ structurally from class II proteins, which are found on the membrane of viruses responsible for diseases such dengue fever, hepatitis C, West Nile and yellow fever.

The structural differences between proteins from the two classes have led to the assumption that they also differ mechanistically. The available information on the fusion mechanism of class I proteins with the host plasma membrane has contributed to the development of existing therapies against HIV. Now, ultrastructural analysis of glycoprotein E on the surface of dengue virus provides data that suggest parallel entry mechanisms for viruses with class I and class II fusion proteins [1].

Researchers led by Stephen Harrison, a principal investigator at Harvard Medical School (http://www.hms.harvard.edu), used the crystal structure of the glycoprotein E to study its insertion into the host membrane. From the data obtained using this high-resolution approach, they propose a mechanism for the entry of dengue virus into the host cell.



Dengue virus vector *Aedes albopictus* feeding on a human host. DC/PHIL/James Gathany

Pore formation

According to the model, binding of glycoprotein E to receptors on the host-cell surface and/or exposure to low pH lead to orientational rearrangements in the three domains of this protein. Consequently, a trimer of exposed fusion peptides forms and directly binds to the host-cell membrane. The protein, which is now attached to both viral and host-cell membranes, undergoes further conformational changes pulling the two membranes close together. The resulting membrane fusion leads to the formation of pores and thus the entry of viral genetic material.

In addition to elucidating the entry mechanism of viruses with class II proteins, these data are important for designing drugs against the wide range of diseases caused by this group of viruses, claim the researchers. Dengue fever alone infects 50–100 million people worldwide each year. The most severe form of the disease is dengue hemorrhagic fever, which affects 0.5 million individuals and is life threatening.

Treatment approaches

These latest data highlight several possible approaches to the treatment of dengue fever, says lead author Harrison. 'There would be several targets: a protease, a helicase, and a polymerase,' he said. 'We plan to work on two categories of potential fusion inhibitors. But I believe the first therapeutics are more likely to be enzyme inhibitors.'

Highlighting the broader significance of this study, Harrison added, 'A more important therapeutic target is hepatitis C virus, which may have a similar structure for its fusion protein. We hope to pursue this problem, based on our success with dengue.'

Reference

1 Modis, Y. et al. (2004) Structure of the dengue virus envelope protein after membrane fusion. *Nature* 427, 313–319

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