Actin strange

Jo Whelan, freelance writer

Actin, an abundant protein that forms part of the cytoskeleton, could be the key to a potential new class of antiviral drugs. Researchers at the Brookhaven National Laboratory (http://www.bnl.gov) have discovered that actin can act as a co-factor for viral enzymes that break down the cytoskeleton and enable new virus particles to escape from the infected cell.

Many viruses exploit their host's cytoskeletal proteins during infection, and actin is known to have various roles in the infection process of several types of human virus. These include the adenoviruses, which cause respiratory and gastrointestinal infections and conjunctivitis. The maturation of newly synthesized adenovirus particles is completed by a viral protease called adenovirus proteinase (AVP) [1]. AVP also cleaves cytokeratin 18, a host cytoskeletal protein, at two consensus cleavage sequences, causing the cytokeratin network to fall apart [2].

However, a puzzle remained. Cytokeratin 18 is cleaved by AVP in the cytoplasm. Brookhaven's Walter Mangel and colleagues had shown that AVP is synthesized in an inactive form [3] and then migrates to the nucleus, where it is activated by two viral co-factors inside nascent virions. Cytokeratin 18 can not be cleaved by AVP *in vitro*, but the cleavage can take place in the cytoplasm of HeLa cells, in the absence of other viral proteins [4]. Therefore, the search was on for a co-factor that could account for the activity of AVP in the cytoplasm.

Actin as co-factor

When Mangel presented his work on AVP at a seminar, Clarence Schutt from Princeton University (http://www.princeton.edu) pointed out that one of the viral co-factors had an amino acid sequence similar to that of actin. Specifically, the C terminus of the actin molecule has a region that is homologous to the viral peptide pVIc, a known co-factor of AVP. In a highly specific fluorogenic assay, actin stimulated the activity of AVP in a concentration-dependent manner, just as pVIc does. They showed that AVP alone did not cleave cytokeratin 18 *in vitro*, but cleavage did take place when actin was added [4]. AVP was shown to bind to the C-terminus of actin and the resulting complex was able to cleave cytoskeletal proteins, including actin itself. 'The really interesting finding is that actin is a co-factor for its own destruction,' says Mangel.

Work is now under way to crystallize the actin–AVP complex and to determine its atomic structure. 'Once we have the three-dimensional structure we will know the exact points at which actin binds, and then we can use structure-based drug design [5] to find a molecule that interacts with the actin binding site on AVP,' says Mangel. 'Finding a suitable molecule in a database should be easy.' Actin has many physiological roles, so to avoid side-effects any drug will need to interact with AVP, which is specific to the virus, rather than with actin itself.

Novel antiviral approach

Most antiviral drugs target the active sites of viral enzymes. Targeting a co-factor binding site is a novel approach [6]. 'If you can stop actin from binding to AVP, you may have a new kind of antiviral agent,' says Mangel. 'If the virus cannot lyse the host cell, the new virions cannot escape and the infection is aborted.' The expectation is that this is applicable to other viruses, not just adenoviruses. 'All viruses have to do something to actin, and this is a new way that may be used by other viruses,' Mangel added. However, the group intends to characterize the system fully in adenoviruses before moving on to test other viral families. There are numerous animal models of adenovirus infection in which potential new antiviral agents could be tested.

Targeting a co-factor binding site could prove to have advantages in terms of viral resistance. 'One of the binding sites that actin utilizes on the protease is the peptide co-factor binding site. So if you were to prevent actin binding by mutation, you might also inactivate the protease because the peptide co-factor could not bind,' explained Mangel. 'If that were the case, then it would be less likely that resistance would arise.'

'Blocking a co-factor would seem to give us more possibilities for counteracting viral infections,' says Jadwiga Chroboczek, Head of the Molecular and Structural Virology Laboratory in the Institut of Structural Biology (CEA/CNRS; http://www.ibs.fr) in Grenoble, France. 'For example, you can imagine a situation where a direct enzyme inhibitor is insoluble, or can not pass the cell membrane, or can not reach a particular tissue, but the inhibitor of a co-factor can.' She adds: 'This work expands our knowledge about viral strategies and will help us to ask detailed questions concerning the molecular mechanisms of interaction of other human viruses with actin'.

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Big gene banks: nuggets for drug discovery or fool's gold?

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Do gene-mapping studies that look for complex traits among human populations hold promise for identifying drug targets? The best known of these studies, undertaken in Finland and Iceland, have certainly deepened our scientific understanding of the causes of disease. However, some experts outside the industry are now questioning whether the payoff in treatment will ultimately justify the expense.

'Everyone in the world is trying to find genes and appropriate them to complex diseases, and then do epidemiology on cohorts to find differences between biological and environmental components,' says Claude Laberge, Director of the Quebec Network of Applied Genetic Medicine (http://www.rmga.qc.ca/default.htm), which recently issued its *Principles on Human Genetic Research Involving Populations*. 'This is good for finding genes,' he says, 'but afterwards someone must put the findings to use in the general population. He continues, 'Just where will this new knowledge be important? In prevention, promotion of treatment, organizing access to genetic services, or to customize or personalize a type of treatment?' The answer is unclear.

Benefits of large-scale studies

Large-scale population studies are numerous, from those involving the Amish and Mennonites, to the Pima Indians of Arizona, the Sardinians and the Bedouins. Those conducted in Finland and Iceland are perhaps the most substantial examples. With stable cultures and demographic registries, they approximate the usefulness of inbred animal strains to genetic research. For example, Leena Peltonen a human geneticist of the University of California, Los Angeles (http://www.ucla.edu), and the University of Helsinki (http://www.helsinki.fi/english), notes that these populations have a high prevalence for certain diseases, more inbreeding (the best conditions for mapping recessive genes), a more uniform genetic background and good genealogical records.

Certain hallmarks of Finland's demographic history – few original founders, subsequent isolation, rapid expansion and bottlenecks that contribute to genetic drift – have helped to create a unique catalogue of genetic diseases. Finnish records include a large proportion of the total population of 5.1 million.

Iceland, founded by a small number of ninth-century settlers from Norway, Ireland and Scotland, has experienced little immigration over the past 11 centuries. Its 1000-year file of genealogical records provides unique sources for tracking hereditary diseases.

Mapping gene location

The private Reykjavik company, deCode Genetics (http://www.decodegenetics.com/), makes the most of these historical records. The Icelandic government has licensed deCode to build and operate a database based on a detailed genetic study of the population.

In recent months, deCode scientists have announced their mapping of the locations of errant genes for numerous

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