This hasn't been evaluated, said Virgin, but he added: 'It could have been overlooked.'

The complement system has been studied extensively in pox and herpes viruses, according to Peter Lachmann from the UK's Centre for Veterinary Science in Cambridge (http://www.vet.cam.ac.uk). 'What seems to be new… is the biological effect on latency in C3 knockout mice, which looks interesting,' he commented. 'Mostly, complement-deficient humans do not have problems with pox viruses or herpes viruses,' he added. This is probably because the main immune control of such viruses is through T cells, and the effect of complement is, as suggested by its name, complementary.

Other evasionary tactics

Recent research has shown that susceptibility to the pathogen *Borellia bergdorferi*, which causes Lyme disease, also involves complement. According to Klaus Kurtenbach at Imperial College of Science, Technology and Medicine (http://www.ic.ac.uk), the interaction of Lyme disease spirochetes with host complement is 'a new framework that has only recently been recognized.'

Spirochetes in the gut of the tick that transmits Lyme disease interact with complement, he explains. *Borrelia* express proteins that specifically bind the complement-regulator factor H of animals that the ticks feed upon, and spirochetes that coat themselves with host complement regulators are protected against lysis. 'This is not exactly mimicry, rather deflection of innate immunity,' Kurtenbach said.

The next step in herpes virus research is to elucidate the molecular mechanism by which the γHV68 protein mimic regulates complement, said Virgin. He added, 'This viral protein is the only one of its class that regulates complement in both mouse and humans.' This unique characteristic could make it useful for further studies of complement, he says.

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Do all paths lead to DARPP-32?

Vida Foubister, freelance writer

A central brain chemical involved in signaling pathways that range from the psychostimulant action of caffeine to the biochemical effects of the antidepressant Prozac has now been linked to a psychiatric illness. Patients with schizophrenia were found to have significantly reduced levels of DARPP-32 in the dorsolateral prefrontal cortex, an area of the brain that is compromized in schizophrenia. This is the first time that DARPP-32 has been implicated in a disease, and suggests that this brain processor might be associated with other psychiatric or neurological disorders.

The biochemistry of DARPP-32

Dopamine- and cyclic AMP-regulated phosphoprotein of relative molecular mass 32,000 (DARPP-32) has a key role in many neurotransmitter pathways throughout the brain [1]. It has been shown to be involved in controlling receptors, ion channels and other physiological factors including the brain's response to drugs of abuse, such as cocaine, opiates and nicotine. A recent study has also shown that the stimula tory effect of caffeine on motor activity is reduced in laboratory mice that have been genetically altered to lack the DARPP-32 protein [2].

DARPP-32 is reciprocally regulated by the two neurotransmitters that are most often implicated in schizophrenia – dopamine and glutamate (Fig. 1). Dopamine activates DARPP-32 through the D1 receptor pathway and disables DARPP-32 through the D2 receptor. Glutamate, acting through the *N*-methyl-D-aspartate receptor, renders DARPP-32 inactive.

It was this association that gave William E. Bunney Jr, Professor of Psychiatry at the University of California at Irvine (http://www.uci.edu/), and his colleagues the impetus to look at the levels of this brain chemical in patients with schizophrenia. 'DARPP-32 appeared to be sitting at a really key control spot in this metabolic pathway,' he explains.

Their analysis of 14 deceased schizophrenia patients found a selective decrease of DARPP-32 protein levels in the dorsolateral prefrontal cortex circuit, a region of the brain that is important for thinking and mood [3]. It is also a circuit

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that is known to have abnormalities in patients with schizophrenia. 'This is not just any region, this is a highly compromized critical area of the brain,' Bunney said.

Causative or compensatory

Although the study is attracting interest, it remains unknown whether the reduction in DARPP-32 is a cause of schizophrenia or a compensatory adaptation to the disease. Either way, Richard E. Chipkin, a neuropharmacologist and CEO of Psychiatric Genomics (http://www.psygenomics.com/), said the finding has implications for drug discovery. 'It's probably just as important to know if DARPP-32 is involved with the compensatory changes because that might be important in a treatment. If the body is making changes in DARPP-32 to try and fix itself, then you might want to do the same thing.'

The researchers also left open the question of whether DARPP-32 was in an activated, phosphorylated state or inactive, dephosphorylated state. Therefore, it is unclear whether there is an abnormal component based on phosphorylation.

Anissa Abi-Dargham, Associate Professor in Psychiatry and Radiology at Columbia University (http://www.columbia.edu), would like to see the results replicated. 'Post-mortem studies are technically very difficult,' she observes. A second step, which has its own technical challenges, would be to radiolabel DARPP-32 and perform brain-imaging studies on living patients.

Implications for other diseases

Schizophrenia affects 1% of the population worldwide. Although the exact cause of the disease remains unknown, it is thought to be triggered by the interaction of genetic and environmental factors. The symptoms vary in severity and can include social problems, hallucinations, delusions, paranoia, reduced emotional responses and disorganized behaviour.

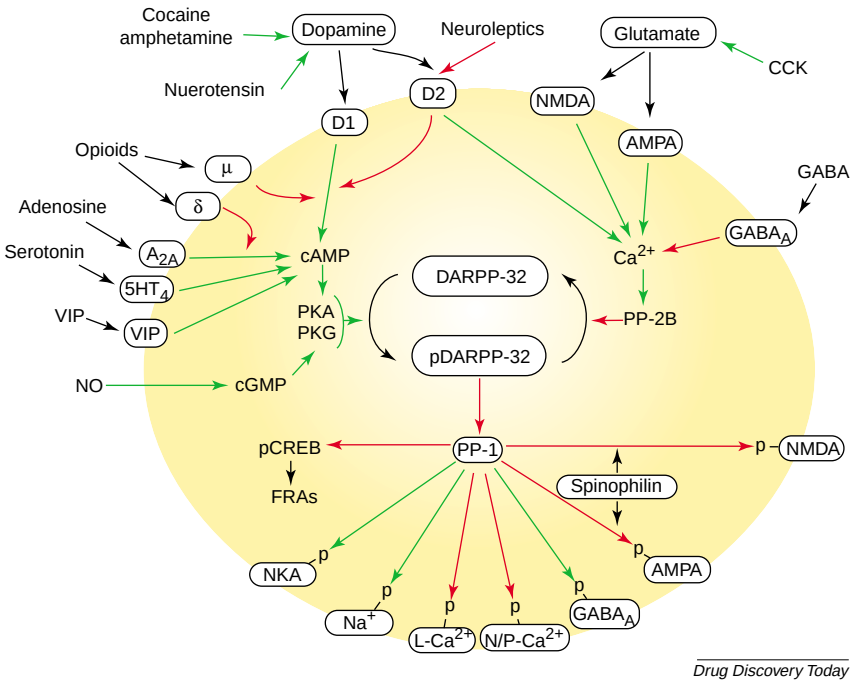

Figure 1. Central role of DARPP-32 as a molecular integrator of dopaminergic and glutamatergic signaling. Abbreviations: A_{2A}, adenosine 2A; AMPA, L-α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid; cAMP, cyclic adenosine monophospate; pCREB, cAMP response binding protein; CCK, cholacystokinin; cGMP, cyclic guanosine monophosphate; GABA, γ-aminobutyric acid; L-, N- and P-type Ca^{2+}, ion channels; NKA, neurokinin A; NMDA, *N*-methyl-D-aspartate; NO, nitric oxide; PKA, protein kinase A; PKG, protein kinase G; PP2B, protein phosphatase 2B (calcineurin); $5HT_4$, serotonin 4; VIP, vasoactive intestinal polypeptide. Reproduced, with permission, from Ref. [3].

Given the pivotal role of DARPP-32 in the brain, it is likely to be implicated in other diseases. Paul Greengard, Professor of Molecular and Cellular Neuroscience at Rockefeller University (http://www.rockefeller.edu) who won the Nobel Prize for Medicine in 2000 for his work on DARPP-32, has begun looking at the regulation and levels of this chemical in parts of the brain that are thought to be associated with neurologic or psychiatric disorders. 'When we discovered DARPP-32 many years ago, we thought that its main role was involved in signaling in the dopamine pathway,' said Greengard. 'But now it is clear that it is involved in signaling in many neurotransmitter pathways, and it has functions throughout the brain.'

More specifically, two recent studies suggest that DARPP-32 might have an important role in depression. Researchers found that fluoxetine (Prozac) no longer has its beneficial behavioural effects in DARPP-32 knockout mice, indicating a crucial role for this phosphoprotein in mediating the actions of serotonin [4,5].

Based on new data that suggests bipolar disease and schizophrenia share some pathophysiological abnormalities, Bunney is interested in looking at DARPP-32 levels in patients with bipolar disease. It is also possible that DARPP-32 abnormalities might be found in other illnesses that have altered dopaminergic signaling pathways, such as Parkinson's disease and cocaine addiction.

Drug discovery in this area, however, is not likely to be simple. Many drugs that are typically used to regulate protein levels are unable to cross the blood–brain barrier, rendering them useless in the CNS. Instead, scientists need

to understand fully the pathways that DARPP-32 is involved in before they can determine where to intervene therapeutically. 'You would have to find some way to regulate what it does, either its level of phosphorylation or some effect downstream from there,' Chipkin said.

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New rodent models gnawing at the black box of ALS

Kathryn Senior, freelance writer

Despite intensive research, potential therapies prove elusive for the fatal disease amyotrophic lateral sclerosis (ALS). To encourage aggressive research towards a cure, the ALS patients' group in the USA set up the Lou Gehrig Challenge in May 2000. One result is a new transgenic rat model of the disease, which became available to laboratories worldwide in August 2002.

David Howland of Wyeth Research (http://www.wyeth.com) has developed transgenic rats that, like the transgenic mice already used extensively in ALS research, overexpress Cu^{2+}–Zn^{2+} superoxide dismutase 1 (SOD1) [1]. Some inherited cases of human ALS are a result of defects in this gene. The transgenic rats and mice show remarkably similar signs and symptoms, making them valuable research tools.

Worldwide, about 120,000 individuals develop ALS (also called motor neuron disease or MND) each year, typically active adults between 40 and 70. Most die within 2–5 years of diagnosis. Currently just one drug, Rilutek, has FDA approval for ALS, but this extends life by an average of only three months. The new rat model should enable research to progress more quickly toward new stem cell therapies as well as new drugs, because the transgenic rat, larger and longer-lived than the mouse, should enable experimental approaches that were impossible before.

Recent progress using transgenic mice

Meanwhile, the mouse model has been providing useful insights. It figured in a study published in September 2002, which sheds light on the origins of the progressive motor-neuron degeneration typical of ALS. The results suggest that exposure to free radicals disturbs membrane lipid metabolism in motor neurons, causing cellular accumulation of ceramides and cholesterol esters. This eventually pushes the cell into apoptosis.

'By blocking sphingolipid synthesis, the first step in this altered pathway, it may be possible to prevent ceramide accumulation and so protect motor neurons against death induced by oxidative stress,' commented senior author Mark Mattson of the Gerontology Research Center at the US National Institute on Aging (http://www.grc.nia.nih.gov/).

Mattson and colleagues found that sphingomyelin, ceramides and cholesterol esters were all elevated in spinal cords from presymptomatic and symptomatic Cu^{2+}–Zn^{2+}-SOD mice, and also found evidence for the same altered lipid metabolism in spinal cords from ALS patients. Cells cultured from spinal cord tissue from these mice responded to experimentally induced oxidative stress by increasing ceramide and cholesterol ester production, sensitizing the motor neurons to apoptosis.

'This makes sense since in normal cells, ceramides play an important role in apoptosis and they have been implicated in the deaths of neurons that occur in ischemic stroke and Parkinson's disease,' Mattson declared [2].

ISP1 as a potential therapy

A drug that hinders sphingolipid production brought this abnormal lipid metabolism cascade to an abrupt halt. 'Blocking *de novo* sphingolipid synthesis using myriocin (ISP1), an inhibitor of serine palmitolytransferase (Fig. 1), prevents increased ceramide synthesis and protects cells from apoptosis,' said Mattson. Work is under way to establish whether ISP1 can delay the onset of motor neuron degeneration and extend lifespan in Cu^{2+}–Zn^{2+}-SOD mice with ALS. The team plans to test whether changes in dietary fatty acids and cholesterol affect ALS development in mice, and (collaborating with epidemiologists) whether risk of ALS is affected by blood levels of sphingolipid and cholesterol-binding proteins.

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