Using an identical canine factor IX expression cassette, they cross-packaged the genome into virions generated from five AAV serotypes and injected them into the muscles of immunodeficient mice and FIX knockout mice. Surprisingly, while the time-to-onset of detectable serum levels appeared the same for all serotypes, types 1, 3 and 5 produced 100–1000-fold more canine factor IX than type 2. 'Simplistically, we think that this could be because more muscle cells are capable of taking up the virus in types 1, 3 and 5,' says Walsh. This would directly result in increased protein leaching into the bloodstream. Furthermore, using green-fluorescent-protein genes in the expression cassette, they found that AAV type 2 could only attach to the slow-twitch muscle fibres, whereas types 1, 3 and 5 also attached to fast-twitch fibres. The team is now repeating the experiments in haemophiliac dogs and primates to ensure there are no key differences in muscle composition between mice and other animals.

Future studies

The results of these studies will be used as a benchmark to determine whether a clinical trial is warranted. Studies will be needed to show that the gene persists in making proteins at high levels (so far, results in mice look promising after six months) and that there are no obvious side effects of the introduced virus or muscle damage when tested in larger animals.

'The mice in the experiment were immunosuppressed to avoid the production of antibodies against the virus or protein. If we move into clinical trials, this will be a major hurdle to overcome,' says Walsh. If ultimately successful, Walsh believes the delivery system could be used for other diseases that require an increase in circulating levels of a protein. For example, in cancer, he suggests the method could be used to produce anti-angiogenesis proteins.

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Flower power: fact or fiction?

Joanna Owens, Joanna.Owens@current-trends.com

Key researchers from both sides of the Atlantic have called for the urgent revision of the regulations of herbal medicines. These remarks were made during a recent seminar in London (UK), sponsored by the Royal Society of Chemistry (London, UK) and the American Chemical Society (Washington, DC, USA).

The dietary supplements market has been one of the fastest growing areas in the healthcare arena in recent years, particularly in the USA where 50% of the population regularly take supplements ranging from multivitamins for general wellbeing to alternative herbal remedies for chronic and serious disease. However, for the majority of these products there is little, if any, scientific evidence of efficacy. This, together with unregulated advertising and media coverage, is a potential risk to the public who might be led to believe that herbal remedies are as effective and safe as conventional medicine. Furthermore, the lack of a regulatory system for these supplements means that the quality and purity of the extract, its toxicity and side effects (e.g. drug–drug interactions, risk to pregnancy) and potential for overdose present a significant risk to the public.

The need for evidence

Edzard Ernst (Chair in Complementary Medicine, Exeter University, UK) presented data from clinical trials that have demonstrated the efficacy of several herbal extracts, such as ginkgo biloba, which has been shown to be beneficial in delaying the chemical deterioration of dementia[1]. Ernst also highlighted the potential problem of drug–drug interactions in patients taking herbal

supplements. For example, it has been reported that St John's wort causes induction of hepatic P450 enzymes, which might reduce plasma levels of concomitantly taken medication[2]. Such interactions are of particular concern because patients often do not inform their doctor that they are taking supplements.

Ernst also discussed how negative data obtained by herbal extract manufacturers might be withheld, leading to

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inaccurate reports of a supplement's safety and efficacy. He emphasized the need for extensive randomized clinical trials, similar to those already published for St John's wort and ginkgo biloba, which are necessary to distinguish between genuinely beneficial herbal supplements and those that have little medical value and pose significant toxicological risk.

A view from the USA

Varro Tyler (Dean and Distinguished Professor Emeritis, School of Pharmacy, Purdue University, West Lafayette, IN, USA) discussed how herbal remedies are regulated as dietary supplements rather than medicines in the USA. Manufacturers of herbal products in the USA are not allowed to claim that a product has a therapeutic effect. Moreover, they must state that any effect that the product might have has not been proven scientifically. The nutritional supplement market in the USA is swamped with products that vary greatly in quality, and a recent trend is the marketing of nutraceuticals, which are expensive foods containing only a small amount of herb extract.

Since its initial boom, the supplements market in the USA has been in decline (by 11.9% from 1998–mid-2000), which is thought to be because of reported side effects, drug–drug interactions, variable quality and limited evidence of efficacy. Tyler described how the lack of patent protection for supplements discourages clinical studies, and how only a few products have been adequately researched; the remainder is based on 'borrowed science'. This leads to a plethora of herbal products that are similar but inferior to well-researched products, which consumers find unsatisfactory, thus causing a decline in sales. Tyler suggested that a potential solution to this would be to approve herbal remedies as over-the-counter or prescription drugs with quality assurance.

Regulating the remedies

Michael Baker (Director of Legal and Regulatory Affairs, Proprietary Association of Great Britain, London, UK) discussed the existing regulations for herbal remedies in the UK, and what options were available for improving these. In the UK, a recent House of Lords report (http://www.parliament.the-stationery-office.co.uk) has outlined recommendations concerning the regulation of herbal remedies, the possibility of setting up centres of excellence for complementary medicine research and the provision of information sources and training for the public and healthcare professionals, respectively. With increased funding and research training, combined with effective regulation, herbal remedies could become an invaluable and safe addition to conventional medicine.

Acknowledgement

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A budding role for the UPR

Sharon Dorrell, Freelance writer

Activation of a stress signal transduction pathway – the unfolded protein response (UPR) – could play an important role in nitrogen-sensing in budding yeast. It could also be a weak point for attack by antifungal agents. Martin Schröder and colleagues at the Howard Hughes Medical Institute (Ann Arbor, MI, USA) suggest that the UPR is activated when there is a rich source of nitrogen and translation rates are high[1]. The UPR is responsible for transcriptional induction of 381 open reading frames in response to endoplasmic reticulum (ER) stress, including genes for ER-resident chaperones and ER-associated protein degrading machinery[1]. The team therefore proposes that high translation rates cause a build-up of unfolded polypeptide chains in the ER and UPR activation induces chaperones that promote protein folding and enable it to catch up with the supply of unfolded proteins in the ER. When nitrogen levels drop, translation rates decrease and the UPR is downregulated.

A protective mechanism

Down-regulation of the UPR also stimulates yeast to switch from normal, vegetative growth to either meiosis and asci formation (sporulation) or to filamentous pseudohyphal growth (the pathogenic form of many of these organisms)[1]. By switching to filamentous pseudohyphal growth, yeast can move and forage for a new nitrogen supply[2]. 'When the ER is stressed and contains unfolded proteins, the UPR has a protective function, and strains that can activate

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