

December 2009

## MND Research Shorts

- Although exposure to electromagnetic fields, such as power lines, had previously been hypothesised to be a risk factor for MND, researchers from France found no detrimental effects of electromagnetic radiation on MND mice.
- Researchers from France have shown that there is evidence of a small increase in the number of enterovirus infections in MND cases. They found that 14% of MND patients in three French MND centres showed evidence of enterovirus infection compared to 7% of the normal population.
- It has been hypothesised that severe infections may effect MND progression. Researchers in Germany recently infected mutant SOD1 MND mice with the bacteria that causes Strep throat a number of times. These infections had no adverse effects on disease progression.
- Researchers from Chicago, USA have found that the muscle fibres of SOD1 MND mice display localised loss of calcium due to stress near the neuromuscular junction (NMJ). They suggest that MND may be a consequence of excess release of calcium.

## New TDP-43 MND mouse; little feet, but a big step forward in MND research

Before a drug can enter clinical trials it generally must go through rigorous pre-clinical trials in the test tube and in animal models of disease. Therefore it is vital to have animal models that reflect the human disease to test these potential drugs on. There have been many mouse models of MND made using various mutations in the SOD1 gene associated with some familial forms. However, although these mice have given us a wealth of valuable information on how the disease may develop in humans, we have not seen this progress into much needed effective treatments. Some would argue that since new genetic mutations in human genes, such as that encoding TDP-43, have now been found to cause MND the time is now right to create new mouse models that may reflect sporadic MND more closely. It comes as no surprise then that a group of researchers from the Hope Center for Neurological Diseases, Washington, USA has made such a mouse. The researchers headed by Dr Robert H. Baloha, published their findings in the Proceedings of the National Academy of Sciences. This mouse model has some features similar to that of human MND, such as protein deposits and specific cell death of motor neurones. This is an exciting step forward and adds a powerful tool to the fight against MND.



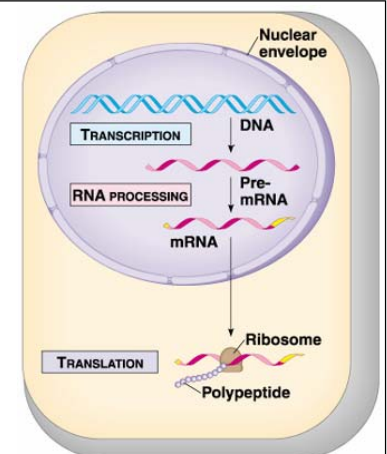
## TDP-43 pile up in MND

Like a production line producing cars that don't work, motor neurones in MND produce proteins, including TDP-43, that pile up and accumulate at the end of the production line. These piles of junk proteins are a common feature of sporadic and familial forms of MND. A recent study conducted in Japan suggests that while researchers are looking for mistakes in the production of proteins it may be mistakes made earlier in the design of these molecules that cause the pile-ups. RNA is a molecule that is a messenger that carries the information from the blueprint in the gene to the machinery that will construct the protein product. Messenger RNA starts off its life as a full-length version of the original blueprint, however, it needs to be cropped or "spliced" in order to remove unnecessary information. This study headed by Yoshinori Nishimoto suggests that in the test tube, messenger RNA may not be "spliced" properly in the presence of mutant TDP-43.

In a direct test of the theory that the protein production line is faulty at the level between blueprint and production line a research group led by Professor John Ravits in Seattle, USA has examined the spinal cords of sporadic MND tissue donors. In a world first Professor Ravits'

## What is RNA?

RNA is very similar in structure to DNA, however, it differs in some very important ways. Most importantly RNA is a much shorter molecule, generally one gene in length and is single stranded while DNA contains thousands of genes and is double stranded. Messenger RNA is a "template" of the DNA that encodes a particular gene. However, there are generally modifications required before it can be used to create a protein. Once the messenger RNA has been modified it is fed into protein construction machinery (called a ribosome) and the code is used to build a string of amino acids called a polypeptide.



team have shown that messenger RNA molecules from hundreds of different human genes are incorrectly "spliced". What this means is that proteins will be assembled from the wrong plans. So they are much more likely to either accumulate into piles of junk or not be produced at all. These errors were found to be specific to the area in and immediately around the

motor neurones. Since these errors are found in motor neurones but not other distant parts of the spinal cord it may be a vital clue in solving the MND puzzle.

Linking sporadic MND with SOD1 familial MND is the work of Professor Michael Strong's laboratory in Ontario, Canada who have just shown that mutant SOD1 associated with some familial cases of MND can bind to RNA and stop it from interacting with the protein production machinery. This may mean that SOD1 may directly interact with some kinds of RNA slowing production of important proteins required for healthy neuron functioning.

### **Molecular housekeepers keeping things tidy**

There are resident proteins in cells whose job it is to make sure that junk proteins do not pile up. These proteins are called molecular chaperones, they can tidy up the cell by recognising and removing damaged proteins. There are many of these chaperones inside cells. It makes sense then that these chaperone proteins may be involved in MND since all forms of MND are associated with accumulations of proteins. Work coming out of Dr Julie Atkin's laboratory at the University of Melbourne suggests that a protein called PDI may be an important chaperone for mutant SOD1. Increasing the levels of PDI actually diminishes the amount of SOD1 accumulated in the cell and protects cells from mutant SOD1 toxicity. Interestingly, PDI is altered in sporadic MND patients as well as mutant SOD1 mice to a form that doesn't work as well at preventing the SOD1 pile up. The researchers also showed that small molecule drugs could be used to mimic PDI and provide similar effects. In addition, researchers from Yale, USA show that a drug called Nogo-A can increase the amount of PDI available and prolong the life of mutant SOD1 MND mice.

### **Immune involvement in MND**

Motor neurone death in MND is associated with an inflammatory response. This recruitment of immune cells is a necessary part of the "clean up" of dying cells. Without this immune response dying neurones would become toxic to neighbouring cells. However, the immune response can become toxic to the very cells it is protecting, and prolonged inflammation in the brain can be detrimental. It is known that MND patients have an increased level of an immune signalling molecule called MCP-1. This, like the bat signal that attracts Gotham's defender, attracts immune cells to the source of the signal. A study recently published by Pamela Shaw's research group at the University of Sheffield,



UK has shown that a particular immune cell in the brain, microglia, can express 3 times as much of this MCP-1 signal when it carries the mutation in SOD1 in the test tube. This may mean that people carrying the mutation are more likely to have an inappropriately increased immune response to certain insults.

Out of a collaboration of Professor Don Cleveland and Professor Beroslav Zlokovic's laboratories in the USA has come research that suggests that treatment with activated protein C therapy can substantially prolong the life span of mutant SOD1 MND mice. The researchers say that this treatment works at the most fundamental level by reducing the levels of the SOD1 in motor neurones and importantly by acting on the brains immune cells. The treatment was able to lower immune signalling of MCP-1 and thus lowered the amount of inflammation in the brain. This drug is already in use for other purposes and so is a promising lead for mutant SOD1 carriers.

Although we just looked at the over-active immune system being harmful, some researchers think that recruiting certain arms of the immune system may prove beneficial. A research team led by Yuanjin Zhang in Beijing, China has used G-CSF (an specific immune stimulator) in a clinical trial of 13 MND patients. The study shows that there are mild side effects to the treatment such as fever or infection-like symptoms. The report also suggests that the patients did not decline faster after the treatment, in fact they may have declined slower over the 6 months after treatment. Caution must be used when interpreting data such as this, since not all patients have a consistent linear progression. Further clinical trials are required to definitively demonstrate any positive outcomes.

### **Flicking Genetic switches in MND**

Just like flicking a light switch genes can be switched on and off by a process known as methylation. This is a useful thing in the body because there are genes that need to be switched on in liver cells that are not required in brain cells and vice versa. However, this switching on and off of genes can go wrong. Until now the possibility that genes had been turned off or on incorrectly by methylation had not been tested in humans. Work coming out of Roger Pamphlett's laboratory at the University of Sydney led by Julia Morahan did just that. They examined methylation in normal brain tissue across the whole genome. The study revealed a number of differences between sporadic MND patients and the normal population. However, the researchers did not find a pattern common to all MND patients. Although specific genes were not commonly altered in their methylation, the researchers identified groups of genes that were more likely to be switched off or on inappropriately. These include genes involved in calcium homeostasis, excitotoxicity and oxidative stress, all of which have been implicated in MND.

