MND Australia
International Research Update

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A new year heralds new links between MND and brain plasticity

We might think that, as adults, there is not a lot of growth or change occurring in our bodies, except for the changes that occur with ageing. In particular, it is a common belief that the adult brain doesn't undergo much growth and positive change as we get older. On the other hand, you may have also heard that our brains are "plastic", meaning they do indeed have the ability to form new neural connections throughout adult life, which is important for learning and memory.

Although MND is mainly a disorder of the motor system, MND researchers are beginning to find hints that the molecules that regulate neuroplasticity and learning are affected and involved in motor neurone (MN) dysfunction. In this report, we will find out more about this research, plus other studies that have set 2016 off to a flying start and bring promise of much exciting MND research to come this year.

Motor neurones make a fuss about neuroplasticity and change

MND is a very complicated disorder, even compared to other states of injury and diseases of the brain and spinal cord. Highlighting its complexity is a study carried out by Sighild Lemarchant and her team in Kuopio, Finland. They investigated a protein called ADAMTS-4 that is specialised in altering the tissue that surrounds and supports neural networks. Through this tissue alteration, it mediates neuroplasticity during development and ageing. ADAMTS-4 is important for neuronal repair after spinal cord injury, however its functions in other diseases of the brain and spinal cord have largely remained unknown.
That is, until Sighild and her team found that the levels of ADAMTS-4 in the spinal cord of SOD1 MND model mice were reduced at disease end-stage compared to unaffected, healthy littermates. What is particularly interesting in their findings is that increasing the levels of ADAMTS-4 in the mice did nothing to improve the disease state; in fact it worsened their condition and accelerated neuromuscular dysfunctions. These results suggest that the reduction in the levels and activity of ADAMTS-4 in MND may be a strategy used by MNs to mitigate whatever harm is actually caused by this protein in these tissues. So despite being beneficial in other injury and disease states of the spinal cord, ADAMTS-4 interferes negatively in the complicated condition that is MND. Exactly *how* it causes harm is a question researchers will need to address next.

Stars, motor neurones and neuroplasticity

Brain plasticity, or neuroplasticity, refers to the ability of the brain to re-wire itself and effectively promote life-long learning. The human brain is made up of billions of neurones that connect to each other in varying networks and patterns to form literally trillions of neural connections. These are astounding numbers; there are about 100 times more neural connections in one human brain than there are stars in the largest galaxies.
These neural connections can change over our whole lifetime. Connections that are not used enough begin to weaken, while new connections are made every time we learn something new and form new memories. This extraordinary ability is regulated by complicated networks of molecules that we are beginning to discover more about. MNs are the largest cells in our bodies, and the ones we are born with are the ones we have for life. This is in contrast to the other types of neurones that we know to be subject to neuroplasticity as both the cells themselves and their connections with other neurones change throughout our lifetime. Thus it will be particularly interesting to investigate and understand the roles of these neuroplasticity-regulating molecular networks in the disease-causing changes that occur in MNs.

MND Research Shorts

- *Targeting single aspects of MND has so far proved to be ineffective, highlighting its complex nature. To try to overcome this, researchers in Israel have developed a "cocktail" treatment to target three molecular pathways involved in the excitotoxicity and oxidative stress that occurs in MND. Impressively, by increasing the activity of key genes in MND model mice, they were able to prolong survival.*
- *Mutations in a protein called FUS are responsible for some of the most aggressive, juvenile-onset forms of MND. For years, researchers have had divergent opinions about how mutant FUS causes disease; is it because of loss of function of the protein, or gain of some toxic function? Now, researchers in New York have new evidence to show that mutant FUS acquires toxic functionality.*
- *Researchers in Japan have discovered that one of the MND-causing mutations of SOD1, H43R, causes complete reversal of the normal antioxidant activity of SOD1. They managed to pinpoint the exact location in the mutation that obtains the "pro-oxidant" activity and causes its damaging effects. This is expected to become an important target for MND treatment.*
- *High levels of interleukin (IL)-18, a protein involved in inflammation, have been found in the serum of sporadic MND patients in several studies. But for the first time, researchers in Belgium and Canada have detected high levels of IL-18 in patients' brains. Further implicating IL-18 these researchers detected another three inflammatory proteins (RIPK3, NLRP3 and caspase-1) in the brain.*

A tiny protein makes a big change

Nature is full of tiny miracles. For instance, we have all those little pollinating insects to thank for the ongoing existence of natural plant communities and the crops we consume. A group of scientists in Australia have discovered a new little miracle in the form of a tiny protein called c29 that has the ability to promote MN health. Dusan Matusica and his colleagues were interested in the p75 neurotrophin receptor ($p75^{NTR}$), a protein that sits on the surface of neurones and detects neurotrophins that regulate neurone function, development and survival. $p75^{NTR}$ helps regulate programmed cell death following disease or trauma. This group derived c29 from $p75^{NTR}$ to test if this tiny protein could actually inhibit MN death caused by $p75^{NTR}$. They cut off the axons of MNs, simulating the dying off process that occurs in MND, and applied c29 to these deteriorating cells. This resulted in decreased cell death, and systemic treatment of c29 in SOD1 MND model mice delayed disease onset and increased the survival rate of spinal cord MNs. Dusan found that the ability of c29 to encourage MN survival depended on the presence of brain-derived neurotrophic factor (BDNF), a protein that helps regulate neuroplasticity, learning and memory. In complete contrast to Sighild Lemarchant's discovery about ADAMTS-4, BDNF and the mechanisms by which it regulates neuroplasticity are somehow very important for MN survival during disease.

Engineering small RNA molecules may help mitigate TDP-43-mediated MND

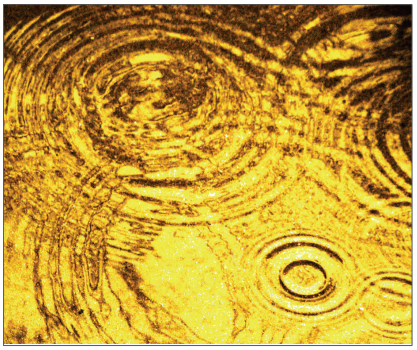
There are several steps in the process that creates proteins from their encoded genes in a person's DNA. Because gene mutations can lead to the production of disease-causing proteins, an approach scientists can use to prevent disease is to interfere in one of these steps and stop the process. RNA interference (RNAi) is a naturally occurring phenomenon in cells in which the messenger RNA (mRNA) molecules that mediate protein production from DNA are targeted by small interfering RNA molecules (siRNAs) and broken apart by the RNA Induced Silencing Complex (RISC). RNAi therapy has been successful in treating some forms of cancer, HIV, immunological disorders and neurological disorders.

Vishwambhar Vishnu Bhandare and Amutha Ramaswamy in Pondicherry, India, wanted to use an RNAi approach to combat MND-causing mutations in the gene encoding TDP-43. Most of these mutations are located in one end of the protein in a region that is rich with glycine (glycine-rich region; GRR). Glycine is the smallest of the 20 different amino acids that make up our proteins, and its high density in part of TDP-43 causes the protein to have a characteristic lack of structure in this region, which occurs in other proteins that also contain a GRR. Vishwambhar and Amutha went about designing possible siRNAs to target the GRR mutants using specialised computational tools. After analysing all aspects of siRNA structure and ability to efficiently target mutant TDP-43 mRNAs they designed very strong candidate siRNAs. However these siRNAs now remain to be experimentally tested to confirm their ability to reduce the production of different TDP-43 mutants and prevent TDP-43-mediated MND.

A small RNA molecule completes a vicious inflammatory cycle

A major contributor to MN death in MND is MN inflammation, caused in large part by activation of a type of neurone support cell called microglia. Associated with this activation is the nuclear factor kappa B (NF-κB) pathway. This pathway regulates a variety of biological processes including immunity and stress responses and thus is very important. However, it becomes harmful when its activity is abnormally sustained. Chiara Parisi and colleagues in Rome, Italy, carried out a study into the molecular basis of the over-active NF-κB pathway with the aim of identifying a way to normalise its activity. A known key protein involved in terminating the NF-κB pathway is the ubiquitin-editing enzyme A20. Chiara and her team investigated A20 more closely and uncovered its involvement in a pathological circuit that sustained microglial activation, inflammation and MN toxicity. This circuit involves a small molecule called miR-125b that is produced by activated microglia. MiR-125b suppresses A20, thereby further activating the NF-κB pathway. Completing this vicious cycle, the suppression of A20 in turn strengthens and prolongs the activity of a protein called the P2X7 receptor, which then activates NF-κB in microglia.

The ripple-on effects of these different molecules on the over-activation of microglia had very harmful consequences for MNs. Chiara then moved on to demonstrate that miR-125b inhibition restored A20 levels and promoted MN survival, shining a spotlight on miR-125b as key mediator of microglia dynamics in MND.



Worms say "cheese!" to accelerate MND research

Many biological studies can be performed using cell-based systems, particularly time-resolved imaging of the health of cells and the molecular events going on inside them. However, the study of complex human diseases, particularly neurodegenerative diseases, usually requires long-term studies in animal models. One of the most useful model organisms used by researchers is *C. elegans*, a species of worm. The ability to use *C. elegans* in automated imaging systems is thus a highly desirable goal for many researchers. Matteo Cornaglia and fellow researchers in Lausanne, Switzerland, have been working on achieving this goal. And their work has, excitingly, come to fruition. They have developed a new multi-functional platform that enables automated worm isolation and culture and long-term high-resolution imaging. Matteo and his team used this system to observe the process of protein aggregation, a universal feature of MND, inside each individual worm and were able to locate the specific tissues in which this took place. This is unprecedented imaging resolution, and is anticipated to be of benefit in screenings for pharmacological compounds that will help treat people with MND.