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With the recent surge in identification of genes responsible for various familial forms of MND, the task at hand has become putting all the pieces together to build a picture of what precisely causes neurones to die in MND. Thus, finding common biological pathways across the various forms of sporadic and familial forms of MND is vital. One feature of MND that is common across the various forms is called *ER stress*. Dr Julie Atkin and co-workers at the Howard Florey Institute have been pioneers of this exciting hypothesis. ER stress does not refer to the stress found in the *Emergency Room* but rather in a compartment found inside motor neurons. ER stands for endoplasmic reticulum and is in fact a vital 'organelle' for all human cells. There have been many studies published on the role of ER stress in MND in the last few months.

In this report we will look at these studies, and some of the other MND research going on around the world.

What is the ER?

The *endoplasmic reticulum* or *ER* is an organelle that creates a separate compartment of the cell using the physical boundary of a membrane. Cells have compartments because certain jobs are performed better in separate spaces. As we would keep production of a flammable liquid away from a job that required flames, the cell also separates specific tasks.

The ER has a number of functions in the cell. Firstly, the ER actively participates in the production, packaging and transport of proteins around the cell and contributes to the release of proteins from the cell. In fact, the ER is very important to neurones since it produces the enzymes that guide production of neurotransmitters that control signal between neurones, and the 'receptors' that act as antenna to receive incoming messages.



What is ER stress?

There are processes that are in place to respond to the level of unfinished proteins built up in the ER. If the protein build up reaches a toxic level, genetic switches are thrown that start the production of specific genes to combat the 'stress'. If the ER senses excessive or prolonged 'stress' it can trigger the cell to self-destruct as a means of ridding the body of dysfunctional cells.

ER stress in motor neurones in sporadic MND

The ER is a factory for producing proteins destined for various organelles in the cell or for export outside of the cell. When there is an accumulation of proteins that are not ready to 'export', the cell switches on a stress response in order to fix the problem. Work that has come out of the Howard Florey Institute has indicated that sporadic MND patients have ER stress response switched on in their spinal cord. To further investigate this, scientists in Japan have recently published work looking at the ER using an electron microscope. They show, what they hypothesise to be, proteins accumulating in the ER and a range of other ER alterations providing the first evidence that the ER in motor neurones is out of shape.

Further work on ER stress has come out of Umea in Sweden where researchers have been analysing the proteins found associated with the SOD1 deposits in mutant SOD1 MND mice. They found that only half of the accumulated protein was actually mutant SOD1 and the other half of the junk was proteins that had unrelated functions. Such as the scaffolding that creates the shape of the cell or molecular chaperone proteins that are supposed to stop proteins accumulating. Of interest is a number of proteins that originate in the ER. This finding further supports the role of the ER in MND and raises the question: how and why is a protein from one compartment interacting with those of another?

MND and a genetic association with ER stress

Failure to restore balance to the ER is associated with human disease. To identify the underlying changes in gene expression in response to ER stress, researchers from Philadelphia, USA examined ER stress in human cells in the laboratory. The scientists found genes that were already known to play a role in the ER-stress response and uncovered several thousand genes that were not previously known to be involved. The genes encoding angiogenin, optineurin and sentaxin are of interest as mutations in these genes are now associated with various forms of MND. This finding is important because it shows that these genes are associated with ER stress and strongly implicate ER stress in these forms of MND. It may be possible that other mutated genes that cause MND may trigger ER stress without being a part of the process itself. If ER stress is a common feature of MND then it may be used to fight against it. If motor neurones have a problem because the ER stress response is constantly switched on, the next step is to try to find a way of switching it off.

TDP-43 lost its keys?

TDP-43 accumulation is a hallmark of sporadic MND. TDP-43 is normally predominantly located in the cell compartment that houses the DNA, the nucleus. However, during MND it is found outside the nucleus where it accumulates. The reason that this occurs is not known. One possibility is that the mechanism that transports TDP-43 back into the nucleus is faulty. Just like a key would get us back into a locked room, so too proteins need specific “keys” or “passwords” in order to pass through the gate of the nucleus. Researchers from London have explored the factors that regulate the transport of TDP-43 and found that the key that TDP-43 uses to get back into the nucleus (called CAS) is deficient in MND patients. The researchers propose that this knowledge could be used to target TDP-43 nuclear transport as a possible treatment.



Doubt over some mutations of SOD1 and their involvement in MND

There are 153 mutations in the SOD1 gene that have been claimed to be associated with MND in an autosomal dominant or autosomal recessive pattern. Research that has come out of Umea, Sweden has cast some doubt on two of the 153 reported mutations. The authors report on four MND pedigrees from Finland, France, Germany and Sweden that had been reported to have either the D90A or E100K SOD1 mutation. They found that some members of the families were affected by MND but did not carry the SOD1 mutation. This may mean that these reported mutations do not cause MND. So just as the changing definition of a planet meant that Pluto was crossed off the planet list, the D90A and E100K SOD1 mutations may have to be re-evaluated as a cause for MND.

FUS junk pile in MND

As with most biological molecules, proteins are being made, used, and disposed of all the time in neurones. Tight regulation of this process means that if a protein is damaged in any way it is disposed of rapidly. A characteristic trait of motor neurones in MND is that proteins accumulate into deposits. These deposits are large ‘junk piles’ that the cell has not been able to eliminate. Many forms of MND are associated with deposits of the protein made from instructions of the TDP-43 gene, while some familial MND cases are associated with deposits of protein made from the mutant SOD1 gene. Researchers in Japan have shown that mutations in the



FUS gene mean that the protein it encodes accumulates and is found in deposits in motor neurones but also in non-neuronal brain cells called astrocytes. This suggests that protein accumulation is a common characteristic of all forms of MND. This knowledge may one day lead to identifying a therapeutic target.

New genetic mutation associated with MND

The genetic mutations associated with most cases of familial MND remain unknown. Researchers from Japan have shown that there are mutations in the gene encoding optineurin (OPTN) in some patients with MND. The optineurin protein has been suggested to have many functions ranging from inflammation induction to shuttling large molecules around the cell. Laboratory experiments showed that the toxicity of OPTN mutations may result from deregulation of a switch that can activate specific genes or as a consequence of accumulation of the optineurin protein into protein ‘junkpiles’ known as deposits. Interestingly, the researchers discovered that optineurin could be found in protein deposits along with TDP-43 and SOD1 in sporadic and SOD1 associated MND respectively.



MND Research Shorts

- Work coming out of Korea suggests that acupuncture or electro-acupuncture slowed MND symptoms in SOD1 MND mice.
- Researchers in Italy have observed that an unusual number of professional soccer players are diagnosed with MND. They found that Italian professional soccer players have higher levels of inflammatory signalling molecules than the normal population.
- Further work in Italy has examined the exposure of MND patients and controls to strenuous physical activity or trauma. The researchers found that MND patients had more exposure to work related and sports related physical activity, but found there was no difference in exposure to trauma.
- Researchers in Washington have called for the use of cannabis for MND patients. They say that cannabis is a powerful antioxidative and anti-inflammatory agent and has neuroprotective effects. It also has properties applicable to symptom management of MND, including analgesia, muscle relaxation, bronchodilation, saliva reduction, appetite stimulation and sleep induction.