

Gharanei S, Zatyka M, Astuti D, Fenton S, Sik A, Nagy Z, Barrett T. Vacuolar-type H⁺-ATPase V1A subunit is a molecular partner of Wolfram syndrome 1 (WFS1) protein, which regulates its stability and expression. *Human Molecular Genetics* 2012

Lay Summary of the research publication in Human Molecular Genetics

What is already known in this subject

Wolfram syndrome is a very rare childhood disease affecting about 1 in 500,000 children in the UK. It causes diabetes, blindness, deafness and slowly degenerating brain disease. There is no cure and no treatment to stop or slow down the course of the disease. It is caused by mistakes in the gene that encodes Wolfram protein, a vitally important protein for protecting cells from stress. It was thought that the only mechanism for cells dying was through too much cell stress that could not be resolved. However, recently, a paper from Japan showed that in insulin secreting cells in the pancreas, the packets of insulin that were being made by the cells, were not being acidified properly and so could not release their cargo of insulin from the cell membrane into the bloodstream. This suggested that there might be another mechanism of disease that had been overlooked.

What we did in our paper

Firstly we wanted to create a good working cell model of Wolfram syndrome: we used childhood brain cancer cells that had been removed at operation. This is a common practice as it gives us a source of human cells that we can study. We removed most of the healthy Wolfram protein out of these cells. We then tested the cells for markers of cell stress; we found that the cells behaved exactly as we would expect in Wolfram patients. We therefore think that we have a good cell model with which to study Wolfram syndrome.

Secondly, we discovered that the Wolfram protein interacts with an enzyme that acidifies protein packets, such as those carrying insulin or neurotransmitters. We think we have found the mechanism by which the packets of insulin made by pancreas cells are not getting acidified properly, so can't release their cargo of insulin into the bloodstream. We think that the same process is happening with packets of some neurotransmitters in the brain, so contributing to the brain disease.

Thirdly, we discovered that in Wolfram protein depleted cells, there was a reduction in the amount of another protein, p21^{cip}. This is a special protein that works when cells divide, to keep them alive. Interestingly, cells that managed to maintain reasonable levels of p21^{cip}, despite low levels of Wolfram protein, did manage to survive much better than cells with low levels of Wolfram protein and p21^{cip} protein.

What this paper adds

We have shown why in Wolfram syndrome, cells cannot release their cargoes of proteins into the bloodstream. It is because the Wolfram protein has to interact with an enzyme that makes the inside of the protein packets acid, without which they cannot dock with the cell membrane. This is important as it suggests that in Wolfram syndrome, disease is caused not just by cell stress, but also

by failure to acidify protein packets. This is a potential link between the hormone problems and the brain problems in Wolfram. The enzyme that acidifies protein packets, the 'proton pump', is itself a drug target and we can try to find medicines that may help to restore or replace its action.

In addition we have identified p21^{cip} as a drug target. There are many chemicals and medicines that increase the expression of p21^{cip} that could potentially protect Wolfram syndrome cells from death.

Implications for children affected by the disease

We are urgently undertaking a drug screening programme to identify drugs and compounds that increase the expression of our target, p21^{cip} using our Wolfram protein depleted cell models. We need to find a drug that keeps Wolfram syndrome cells alive for longer, without being toxic to cells. We are starting with drugs that already have a licence for treatment in other unrelated conditions. If we can find such a drug, we will then be in a position to develop a randomised clinical trial for effectiveness in children with Wolfram syndrome. I am hopeful that we may be able to start such a trial in the next 5 years.