A membrane trafficking protein may be involved in combating dementia

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INTRODUCTION

Alzheimer's disease (AD) and Dementia with Lewy Bodies (DLB) are the two most common dementias, known to result from the accumulation of 'sticky' toxic proteins in neurones.

RAB39B is a protein involved in **membrane trafficking** - the transport of molecules in neurones (1). Previous genetic studies have shown that mutation (removal) of this protein causes neurodegeneration and DLB pathology (2), possibly as a result of failure to maintain effective membrane trafficking.

This project sought to understand the role of **non-mutated** RAB39B in dementia instead.

AIMS

1) To determine if **overall RAB39B levels** are changed in non-genetic variants of AD and DLB
2) To determine if RAB39B **distribution in the cell** is different in healthy and diagnosed cases
3) To **histologically visualise** RAB39B in the brain

METHODS

1) 9 control, 10 DLB and 12 AD cases of frozen post-mortem **temporal lobe tissue** were solubilised and run on a Western blot which was stained with antibodies to show RAB39B.
2) The same brain tissue samples were separated into two sub-cellular fractions by centrifugation: cytoplasmic, containing inactive RAB39B, and **membrane,** containing active RABB39B. The fractions were run on a Western blot.
3) Sections of AD post-mortem temporal lobe tissue were used to search for co-localisation of RAB39B and amyloid plaques (AD pathology) in the brain using immunofluorescence and confocal microscopy.

RESULTS

1)

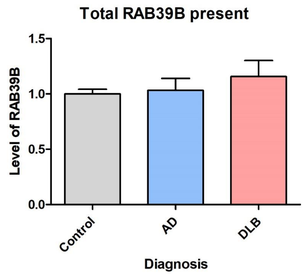

Figure 1. Total RAB39B levels obtained by measuring the amount of RAB39B present in each brain tissue sample of control, AD and DLB cases.

Established that there is **no overall difference in RAB39B levels** between healthy, AD and DLB cases.

2) Separating the sub-cellular components of the brain tissue and analysing the fractions produced on a Western blot showed **no statistically significant difference in RAB39B levels between the cytoplasm and membrane.**

However, RAB39B appeared to decrease in the cytoplasmic fraction and **increase in the membrane** fraction, where it is present in its **active form**.

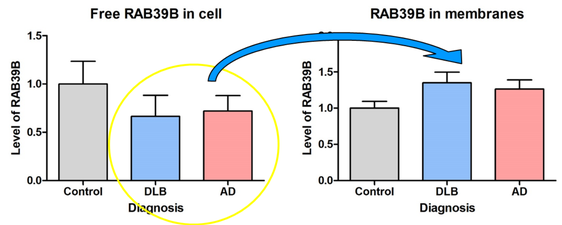

Figure 2. As RAB39B levels decrease in the cytoplasm, they increase in the membrane. This correlation was found to be statistically significant ($p<0.05$).

Furthermore, the increase in membrane (active) RAB39B levels was found to correlate with the severity of AD pathology.

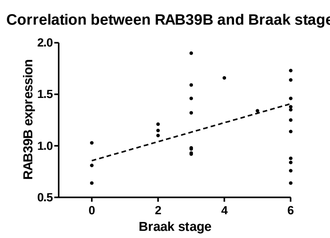

Figure 3. Active RAB39B levels increase with higher Braak stages of Alzheimer's disease pathology, $p<0.05$, $R=0.40$.

3)

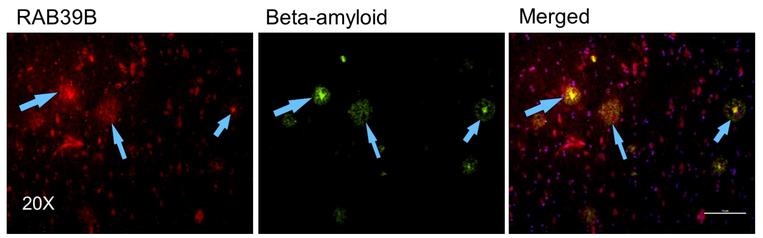

Figure 4. Certain cases of AD presented RAB39B and amyloid plaques co-localising.

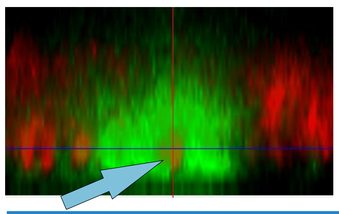

Figure 5. A cross-section of an amyloid plaque (green) with RAB39B (red) incorporated into its core in an AD case.

DISCUSSION

The found trend for decreased cytoplasmic (free) RAB39B and increased membrane RAB39B in AD and DLB (*Figure 2*) may suggest that **this protein is recruited to the membrane where it possesses its active form and potential protective properties against toxic protein aggregates**. This is supported by the increasing levels of active RAB39B with increasing AD severity (Figure 3).

Furthermore, we found that in certain cases of AD, not only does **RAB39B co-localise with amyloid plaques** (protein aggregates seen in AD) but is also incorporated into them (*Figure 4,5*). This might be a pathological mechanism of AD that could be inactivating the proposed protective properties of RAB39B, and therefore further aggravating pathology.

Studying membrane trafficking proteins such as RAB39B could reveal other defensive mechanisms that the cell employs to combat neurodegeneration, which could ultimately lead to the development of treatment.

ACKNOWLEDGEMENTS

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