ABSTRACT

All neurons in a human brain are constantly transporting proteins, which is vital for the survival of neurons. A person who has Alzheimer’s disease develops neuronal dysfunction resulting in plaques of Aβ (Amyloid beta), synaptic lesions and neuronal apoptosis. It has been suggested that the plaques containing Aβ accumulate due to abnormalities in the cleavage of Amyloid Precursor Protein (APP) by Presenilin (PSN), which is the catalytic component of the gamma secretes complex. Previously, APP was found to function as a receptor for kinesin-1 mediated transport. Recently, both PSN and GSK-3β have been implicated in kinesin-1 transport. Using Drosophila, as our model system we investigated how PSN and GSK-3β function in axonal transport. In Drosophila, mutations of motor proteins show axonal blocks indicating axonal transport defects. Using this phenotype we first tested over expression of PSN and found no defect in axonal transport. We next expressed active GSK3β, inactive GSK3β, and a kinase dead form of GSK3β. While both active GSK3β and the kinase dead GSK3β did not show blockages, the inactive GSK3β showed blocks. Furthermore, we tested if GSK3β and PSN genetically interact with each other. We found that reduction of PS with over-expression of active GSK3β also resulted in blockages, in contrast to heterozygous reduction of PSN and expression of active sgg alone. These observations indicate that PSN and GSK3β function in axonal transport. Axonal blockages are observed using the synaptic vesicle marker CSP.

GSKβ and PSN have a role in axonal transport.

Figure 1: APP transports a subclass of vesicles that contain PSN (Kamal et al 2001). APP can function as a kinesin-1 receptor during transport (Gunawardena et al 2001). PSN and GSK3β are thought to interact and regulate kinesin-based transport (Pigino et al 2003, Lazarov et al 2007).

Table 1: Quantitative analysis indicates the extent of axonal blocks. Neuronal expression of inactive GSK3β shows a significant amount of blocks which are enhanced by reduction of PSN. This enhancement is much greater than what is observed with reduction of PSN with active GSK3β. N=4 larvae.

Table 2: Quantitative analysis indicates that the extent of axonal blocks in larvae expressing PSN with reduction of kinesin show a significant amount of blocks compared to larvae expressing GSK3β with reduction of PSN. These observations suggest that both PSN and GSK3β genetically interacts with kinesin and may regulate kinesin based axonal transport pathways. N=4 larvae.

Figure 2: Neuronal expression of active GSK3β resulted in axonal blocks. The reduction of PSN with over-expression of active GSK3β and reduction of PSN with over-expression of inactive GSK3β also resulted in axonal blocks, in contrast to heterozygous reduction of PSN and expression of active sgg alone. These observations indicate that PSN and GSK3β function in axonal transport. Axonal blockages are observed using the synaptic vesicle marker CSP.

Figure 3: Genetic reduced of the kinesin with excess wild type GSK3β (sggβ), active GSK3β (sggS9A), and two different isoforms of PSN resulted in blockages, indicating that GSK3β and presenilin interacts with the kinesin motor.

Quantification analysis of the extent of NMJ transport defects in GSKβ and PSN expressing larvae.

Figure 4: Synaptic defects are observed in larval synapses over expressing wild type GSK3β constitutively active GSK3β (sggS9A) and inactive GSK3β (KK83-84141) compared to wild type larvae. All larvae that are over expressing different forms of GSK3β show defects in NMJ development. NMJs were visualized using the presynaptic markers CSP and HRP at muscle segments 6/7.

Future Directions

- Both PSN and GSK3β have a role in axonal transport and both affect the kinesin-1 motor protein.
- PSN and GSK3β genetically interacts with each other for axonal transport.
- GSK3β has a role in NMJ development.

Conclusions

- Both PSN and GSK3β have a role in axonal transport and both affect the kinesin-1 motor protein.
- PSN and GSK3β genetically interacts with each other for axonal transport.
- GSK3β has a role in NMJ development.

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