

P030 Zinc transport across SH-SY5Y plasma membranes
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Zinc (Zn^{2+}) may play a key role in the aetiology of Alzheimer's disease (AD). Amyloid precursor protein and $A\beta$ bind Zn^{2+} , which can precipitate $A\beta$. Zn^{2+} is highly concentrated in senile plaques and amyloid angiopathy in AD. Zn^{2+} is transported into synaptic vesicles at some glutamatergic nerve terminals by the vesicular membrane transporter, ZnT3. Levels of other members of the same transporter family (ZnT1, 4 and 6) are increased in AD. Although Zn^{2+} moves across the neuronal plasma membrane, the specific transporters responsible are not identified. We have investigated molecular aspects of plasma membrane Zn^{2+} transport in human neuroblastoma SH-SY5Y cells.

SH-SY5Y cells were incubated with $^{67}Zn^{2+}$, and isotope uptake quantified using inductively coupled plasma mass spectrometry (ICPMS). Expression of seven potential Zn^{2+} transporters, and five potential reference genes, was studied using quantitative real-time reverse transcription PCR (qPCR). Zn^{2+} transporter expression levels were decreased using siRNAs (Ambion), and the effect on SH-SY5Y $^{67}Zn^{2+}$ uptake investigated.

SH-SY5Y cells imported $^{67}Zn^{2+}$ from the medium, and they expressed a number of potential Zn^{2+} transporters. qPCR analysis identified the two most stable reference genes for gene expression data normalisation. Transporter expression was successfully knocked-down (> 70%), enabling us to determine their potential roles in SH-SY5Y plasma membrane $^{67}Zn^{2+}$ transport.