Reactivation and functional activity of neurons in cultured postmortem brain tissue slices.
R.W.H. Verwer and D.F. Swaab
Netherlands Institute for Neuroscience, Meibergdreef 47, 1105 BA Amsterdam.
r.verwer@nin.knaw.nl

Background and research question
Our aim is to study the cellular and molecular aspects of AD using human brain tissue in order to look for methods to reactivate neurons. To this end we have developed a culture system of post-mortem brain tissue from adult control and AD patients. Many cells in this tissue stay alive for long periods and can be manipulated experimentally.

Methods and tissues used
Postmortem brain tissue from the precentral gyrus of either controls or patients suffering from Alzheimer’s disease was obtained at rapid autopsies (< 8 hr postmortem delay) performed by the Netherlands Brain Bank (NBB). Tissue slices (200-300 μm thick) were cultured in serum free medium. Viability of neurons in the slices was assessed using calcein-AM/ethidium bromide. Routine immunostaining included βA4, AT-8, NeuN, GFAP and HLA. For co-culture experiments rat embryonic neural stem cells were used. Also, pharmacological intervention with cell cycle compounds has been applied.

Results and conclusion
We have shown for the first time that neurons in brain tissue from deceased AD patients can be induced to express adeno-associated viral vector mediated transgenes while they were surrounded by massive Alzheimer pathology (plaques and tangles), even if the neurons contained pretangles. However, cells in cultured postmortem brain slices from Alzheimer patients were on average less viable than those from normal aged subjects. This lower viability was correlated with the presence of local AD pathology. However, we have shown that embryonic neural stem cells release factors that improve the viability of neurons in post-mortem slices from Alzheimer patients and normal aged subjects. Local pathology did not influence the extent of the improvement of viability. Thus, with slices from some AD patients co-culturing resulted in the same final viability as control subjects, whereas in tissue from other AD patients viability was less improved. This suggests that there may be differences among AD patients that determine their potential to respond to survival promoting factors. We have used q-PCR to investigate changes in gene expression patterns that might help explain the observed changes in neuronal viability. Currently, we are analyzing the data from these experiments.