Author: **Dr Catherine Blizzard**
Qualification: **PhD**
Institution: **University of Tasmania, Menzies Institute for Medical Research**

Title of Project: **Dendritic spine alterations in TDP-43 aggregated Frontotemporal dementia: a novel therapeutic target**

Frontotemporal dementia (FTD) has a prevalence second only to Alzheimer's disease persons under the age of 65. And yet, relatively little is known about how pathogenic events lead to cognitive decline. FTD is caused by frontotemporal lobar degeneration (FTLD) that can be pathologically characterised by the cytoplasmic accumulation of aggregated proteins. One such protein is the transactive response DNA-binding protein 43 (TDP-43).

There is increasing evidence that early synaptic alterations play a key role in many neurodegenerative diseases. Recent studies have shown that, in addition to its' localisation to the nucleus and pathologically in the cytoplasm, TDP-43 is also localised within somatodendritic spines of hippocampal neurons, where it has been shown to act as an upstream regulator of spine turnover and homeostasis. These observations, in conjunction with our preliminary evidence of synaptic scaffolding proteins being misprocessed in the TDP-43 A315T mouse model of FTLD, open up a new potential route by which TDP-43 can affect neuronal function and conversely be implicated in the pathogenesis of neurodegenerative diseases. Therefor we are investigating the novel hypothesis 'mutant TDP-43 results in synaptic pathology, leading to impaired neuroplasticity and ALS'. To achieve this we have created a novel transgenic mouse cross, using the TDP-43^{A315T} mouse model of FTD, and yellow fluorescent protein (YFP) mice. These mice ubiquitously express YFP in subsets of motor neurons in the cortex, making them an ideal model to study synapse morphology.

With this transgenic mouse we have created a complete tissue bank of YFP crossed TDP-43^{A315T} positive and control brains over a time-course of disease and are currently characterising levels of synaptic proteins and post-synaptic spine densities

and morphology. Immunohistochemistry directed at endogenous YFP and synaptic markers was performed on tissue from TDP-43^{A315T}mice crossed with YFP-H mice (P30 n = 5, P90 n =5) and in YFP-H litter matched controls (P30 n = 5, P90 n = 5). Confocal microscopy with subsequent image analysis was used to quantify YFP-H expressing layer V pyramidal neurons, pre-synaptic punctum and basal and apical dendritic spines in the motor and somatosensory cortices. There was no significant difference in the number of YFP-expressing pyramidal neurons in either the motor or somatosensory cortices between TDP-43^{A315T} x YFP-H mice and YFP-H controls at 30 days ($p > 0.05$, two-way ANOVA with Bonferroni post-hoc test). There was however, a significant reduction in pyramidal neurons of the motor cortex in TDP-43^{A315T} x YFP-H mice compared to controls at 90 days, and in comparison to the motor cortex of TDP-43^{A315T} x YFP-H mice at 30 days. No significant changes were identified in pre-synaptic punctum levels at either time point ($p > 0.05$, two ANOVA with Bonferroni post-hoc text). At 30 days, there were no significant differences in spine density between TDP-43^{A315T} x YFP-H mice and controls; however, in TDP-43^{A315T} x YFP-H mice at 90 days there were significant reductions in the spine density of motor basal and apical dendrites, and of somatosensory apical dendrites, in comparison to YFP-H controls (Figure 1). Furthermore we are currently in the process of establishing the 2PLSM live imaging platform in this novel mouse cross that will enable us to determine a precise time-course of synaptic alterations, pin-pointing the earliest synaptic events occurring prior to cell loss.

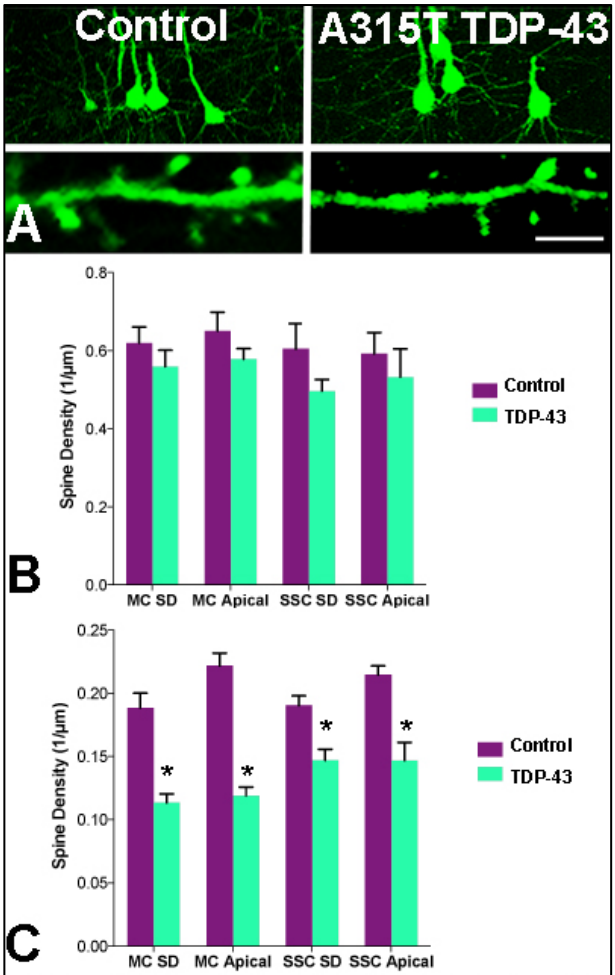

Figure 1. A. Day 90 Control and A315T TDP-43 YFP-H MC. B. No difference in spine density at day 30. C. Spine densities was significantly reduced in somatodendritic (SD) and apical dendrites in the TDP-43 mice, in comparison to control at day 90. Scale bar = A&B 200µm, insets 10µm

Our findings, which form the backbone of a 2016 NHMRC project grant application, suggest post-synaptic dysfunction may be an early-occurring event in mutated TDP-43 pathology, occurring prior to neuronal loss. Understanding the role that TDP-43 plays in synaptic dysfunction may reveal new therapeutic windows for intervention in TDP-43 proteinopathies.