

INTRODUCTION

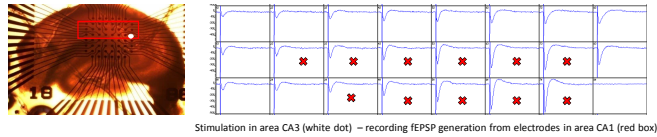
While Huntington's Disease (HD) has classically been considered a disease primarily of the basal ganglia, it is becoming widely accepted that other brain regions, in particular the cortex and hippocampus, are affected. In HD mouse models such as the R6/2 (mhtt exon 1 over-expression model), or the SH4H and Hdh150 models, hippocampal synaptic plasticity is markedly decreased, implying that synaptic transmission deficits are a common hallmark of mhtt expression that may contribute to disease pathophysiology prior to any over neuronal brain atrophy.

We have developed an *in vitro* assay designed around Multi-Electrode Array (MEA) technology to rapidly profile synaptic deficits in hippocampal slices from HD transgenic mouse models. The development of a specialized chip layout to accommodate 3 slices recorded in parallel on one MEA recording station allows a throughput rate of ~ 75 slices per week / operator. R6/2 mice have been characterized for the emergence of synaptic deficits in hippocampal CA3-CA1 transmission at increasing ages (at 6, 8 and 10 weeks) and compared to their WT littermates. Deficits can be seen as early as 6 weeks of age, and involve modest enhancement of basal synaptic transmission and impaired paired-pulse facilitation (PPF), consistent with an increased glutamate release probability from the CA3-CA1 synapses. In addition, the amplitude of Long Term Potentiation (LTP) is significantly decreased at all ages studied.

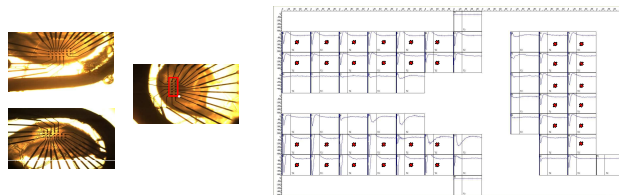
We have initiated a screening strategy in order to identify compounds which may rescue these deficits and thus may be of interest pursuing *in vivo* efficacy models. The achievable throughput is ~ 4 compounds per week (with n = 9 slices per treatment group). We present here some evidence that both BDNF and FGF-2 are capable of differentially rescuing some of the R6/2 synaptic deficits.

MATERIAL & METHODS

Conventional MEA recording of a hippocampal slice on a 60 electrode (8x8) chip



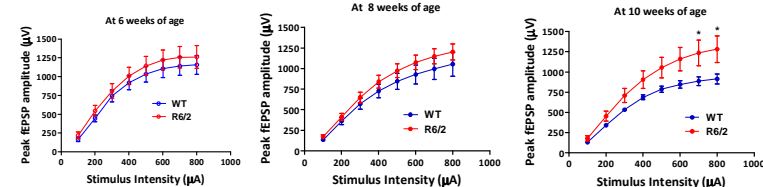
Design of a MEA chip to accommodate 3 hippocampal slices for simultaneous recording



Chips were designed so that electrodes provide adequate space to place 3 slices in one chip with 20 electrodes available for recording from each slice. fEPSPs were recorded in area CA1 following stimulation of the Schaeffer Collaterals with a single electrode underlying the CA3-CA1 border. The electrodes could be further categorized into those underlying the stratum radiatum, or the stratum pyramidale of CA1.

RESULTS

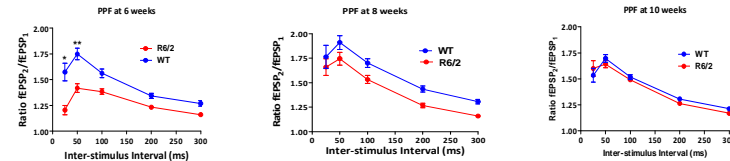
Assay 1 – Input-Output Characteristics to monitor basal synaptic transmission



With age, there is a progressive enhancement of basal synaptic transmission in R6/2 hippocampal slices relative to WT littermates. Table shows the significance value for the increase as assessed by a 2-way ANOVA. Here, the I-O characteristics have been further subdivided as from the stratum radiatum or stratum pyramidale layer.

RESULTS CONTINUED

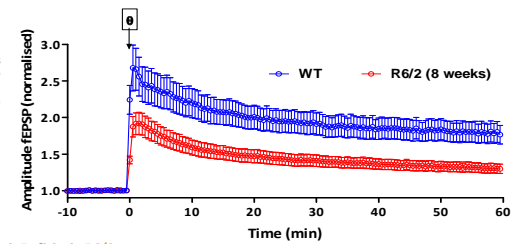
Assay 2 – Paired-Pulse Facilitation



A paired-pulse facilitation protocol at varying inter-stimulus intervals shows robust reduction in PPF values at 6 and 8 weeks, but not at 10 weeks.

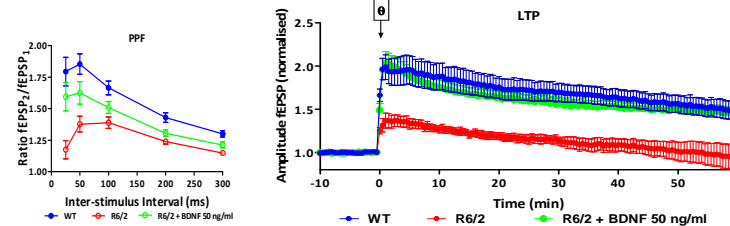
Assay 3 – Long Term Potentiation

Long Term Potentiation was elicited following theta burst stimulation (θ) in R6/2 and WT littermate slices at 6, 8 and 10 weeks of age. Significant deficits in LTP were recorded in R6/2 slices at all age groups interrogated, with no apparent temporal exacerbation of the deficits at the ages recorded. The data shown here is from R6/2 slices at 8 weeks of age. R6/2 animals aged to 7 - 8 weeks were subsequently used for all further screens.



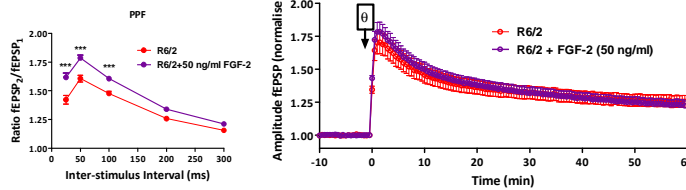
Evaluation of Compounds to Rescue Synaptic Deficits in R6/2

BDNF (50 ng/ml)



In agreement with Simmons et al, 2007, 2 nM BDNF is capable of restoring the LTP deficits in R6/2 slices. Significant rescue of PPF values was also noted.

FGF-2 (50 ng/ml)



FGF-2 reduced synaptic transmission in the R6/2 slice, rescuing the reduction in PPF, but failed to reverse the LTP deficits.

CONCLUSION

Synaptic profiling of transgenic murine models of HD has been evaluated as a potential screening strategy to profile compounds prior to *in vivo* efficacy testing. The increased throughput afforded by novel MEA chip design suggests that this may be a useful tool to probe mechanism of action and efficacy of compounds against an early signature of network dysfunction which likely contributes to HD pathophysiology.