INTRODUCTION

Huntington’s disease (HD) is a hereditary neurodegenerative disease, which causes dysfunction in both cognitive and behavioral motor systems. It is brought about by genetic mutations of the CAG trinucleotide codon on the huntingtin gene. CAG repeats in excess of the normal 37-39 base pairs result in HD.

The striatum, the major input region of the basal ganglia, is integral to the production of motor movement. The striatum is a key region of interest in HD research, as striatal activity is disrupted in the early stages of HD and in the later stages of the disease the striatum undergoes significant cell loss. Syntphin-1A (STX1A) is a membrane protein involved in neurotransmitter release (Figure 1). Mutant huntingtin appears to interfere with the normal function of STX1A and may disrupt neurotransmitter release in HD (Figure 1b, Swney et al., 2003).

METHODS

- All mice, (R6/2, R6/2 STX1A -/+ and wild type littermates) were tested between 12.5 and 18.5 weeks of age. Animals were provided by the Ellerby lab at the Buck Institute for Research on Aging.
- During microdialysis the target area was perfused with aCSF during baseline collections and high potassium (10mM) aCSF for the remainder of the experiment.
- Post-dialysis, tissue microprobes were taken from the cortex, striatum, and cerebellum of the opposite hemisphere.
- Microdialysis and microprobe samples were analyzed using HPLC-ED and OPF/immunoassay pro-column derivatization.
- All animals were treated according to guidelines of Central Michigan University’s Institutional Animal Care and Use Committee (IACUC).

RESULTS

- Glutamate main effect of time (p<0.001) and group difference (p=0.047; B).
- Glutamate showed a main effect of time (p<0.001) and group difference (p=0.047; B); GABA main effect of time (p<0.001) and group difference (p=0.014; C). Post-hoc WT and STX1A -/+ (p=0.043) STX1A values showing greater percent change across normalized collections. [*p=0.05]

CONCLUSION

These results suggest that HD and STX1A-/- mutations influence both basal and stimulated levels of glutamate, glutamine, and GABA in the striatum and that the combination of the mutations produces a distinct physiological profile by comparison to WT. There were several distinctions in microdialysis results between the WT compared to STX1A-/- and WT compared to R6/2 STX1A -/- which suggest that the STX1A mutation did modify neurotransmitter release within the striatum.

After stimulation, percent change values for glutamate were similar across genotypes. Both the R6/2 STX1A -/- and STX1A -/- groups had significantly higher nanomolar values of glutamate at baseline and under stimulated conditions. Thus, it is possible that the similarity of percent change across genotypes is due to a ceiling effect in the R6/2 STX1A -/- and STX1A -/- groups.

Microdialysis analysis revealed significant group differences for all three amino acids in the striatum, which suggests a distinct genotype effect on tissue levels of these neurotransmitters. The R6/2 group had the highest mean values compared to all other genotypes, which suggests an increase in the production and/or storage of glutamate, glutamine, and GABA in the context of HD. Although only striatal glutamine is significantly different between R6/2 and R6/2 STX1A -/-, the overall trend suggests that the STX1A mutation brings R6/2 to tissue levels of glutamine, glutamate, and GABA closer to WT within the striatum.

This exploration was stimulated by the observed increase in motor symptoms in the R6/2 animal as a result of the STX1A mutation. Although our procedures did not directly stimulate neurotransmitter release in the striatum, it did not drive striatal activity in the way a rotated task would. Furthermore, our animals were mostly sedentary during microdialysis which may explain the lack of distinction between R6/2 and R6/2 STX1A -/- animals.

The relationship between STX1A-/- and mHtt represents one way in which the HD mutation directly influences regulation of neurotransmitter release and levels of intracellular calcium. The R6/2, R6/2 STX1A -/- and STX1A -/- animals all present with similar total amounts of all three neurotransmitters measured in microdialysis yet the STX1A mutation alone is not sufficient to produce motor deficits. It is possible that the STX1A -/- animals were able to compensate for the so that even though their physiology is altered by comparison to WT, their motor behavior is not.

REFERENCES


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