IJAE
Vol. 117, n. 2 (Supplement): 128, 2012
ITALIAN JOURNAL OF ANATOMY AND EMBRYOLOGY

Morphometrical evaluation of neurodegeneration: an integrated multiparametric approach using whole slide imaging

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Assessing degree of neurodegeneration and eventually identifying a compound related neuroprotection can be much more arduous than one might imagine: number of animals, number of samples/animal, experimental design and statistics, together with the choice of proper IHC markers and the technology whereby they are evaluated are prominent factors in order to obtain reliable results. In acute models, histopathological measurements are an essential part of the experimental procedure; here we describe an integrated (qualitative/quantitative) histological assessment to be applied for the evaluation of neuroprotection in acute models of Huntington's disease.

We used whole slide imaging to validate a rat model of Huntington disease obtained by intrastriatal viral vector delivery of mutant huntingtin (Htt). Recombinant Adeno Associated Viral (AAV) vectors have been used successfully to transfer genes in a variety of tissues, including the brain, in adult animals [1]. Here we used rAAV9, charged with Exon 1 Htt carrying 17 and 138 CAG repeats. AAV9-Ex1-GFP-Q138 injection induced the formation of GFP positive Htt aggregates in the entire striatal area, increased GFAP and microglial activation with respect to Q17 injected striatum.

NeuN, ChAT, GFAP, OX42 immunohistochemistry and GFP epifluorescence were evaluated contemporaneously to qualitatively evaluate the degree of induced lesions; qualitative evaluation allowed to exclude animals that have not responded to rAAV9 infection. The remaining selected animals were used for a multivariate statistical analysis based on whole slide imaging of immunohistochemically stained sections.

We believe this approach increases results reliability when evaluating animal models of neurodegeneration.

References

[1] Tenenbaum L, Chtarto A, Lehtonen E, Velu T, Brotchi J, Levivier M (Recombinant AAV-mediated gene delivery to the central nervous system. J Gene Med 6 Suppl 1:S212-S222.2004).

Keywords: Whole slide imaging, immunohistochemistry, Huntington's disease, animal model.