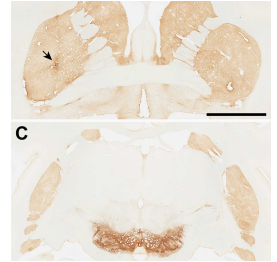


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Protocol for "quantification of the nigrostriatal system in non-human primate brain sections"

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Protocol status: Working

We use this protocol and it's working

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Disclaimer

Nothing to be disclosed

Abstract

Here we describe the method conducted to estimate (i) number of TH+ neurons in the substantia nigra pars compacta by taking advantage of a dedicated algorithm (Aiforia) and (ii) optical density of TH innervation in the putamen nucleus.

These two levels of analysis are required to properly estimate the potential damage of the nigrostriatal system in non-human primates, bot at origin (substantia nigra pars compacta) as well as at destination (putamen nucleus).

Image Attribution

Jose L. Lanciego

Quantification of TH+ neurons in the substantia nigra of non-human primates"

- 1 Scan sections to be analysed in the Aperio CS scanner at a magnification of 20x.
- 2 Upload digitalized sections to the Aiforia cloud (www.aiforia.com).
- 3 Create a dedicated project for analysis.
- 4 Select up to 3-4 sections for training the algorithm, these sections showing different morphologies and intensity stains of tyrosine hydroxylase (TH).
- 5 Train the model algorithm to analyze potential errors.
- 6 Activate the annotation assistant tool to retrieve additional beneficial inputs that can be added by the platform itself.
- 7 Repeat steps 5 to 7 while increasing the number of annotations and iterations (> 1,000) until the model algorithm has learned to properly identify every single neuron showing TH stain.
- 8 Release the model algorithm.
- 9 Use the drawing tool to delineate the boundaries of the substantia nigra where TH+ neurons need to be quantified in every single section of the project.
- 10 Export obtained raw data to an Excel spreadsheet for statistical analysis.

Quantification of optical density of TH innervation in the putamen of non-human primates

- 11 Scan sections to be analyzed in the Aperio CS scanner at a magnification of 20x



- 12 For each section, the boundaries of the putamen nucleus (pre- and post-commissural putamen) are delineated and the optical density of TH immunoperoxidase stain measured with Fiji Image J software.
- 13 Convert obtained values for optical densities to a logarithmic scale (Ruifrok and Johnston, 2011).
- 14 Export obtained raw data values to an Excel spreadsheet for statistical analysis.

Protocol references

Ruifrok,

A.C., and Johnston, D.A. 2011. Quantification of histochemical staining by color deconvolution. *Anal. Quant. Cytol. Histol.* 23, 291-299