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Evaluation of Microglial Action in a JSK Treated Paraquat Rat Model of Parkinson's Disease

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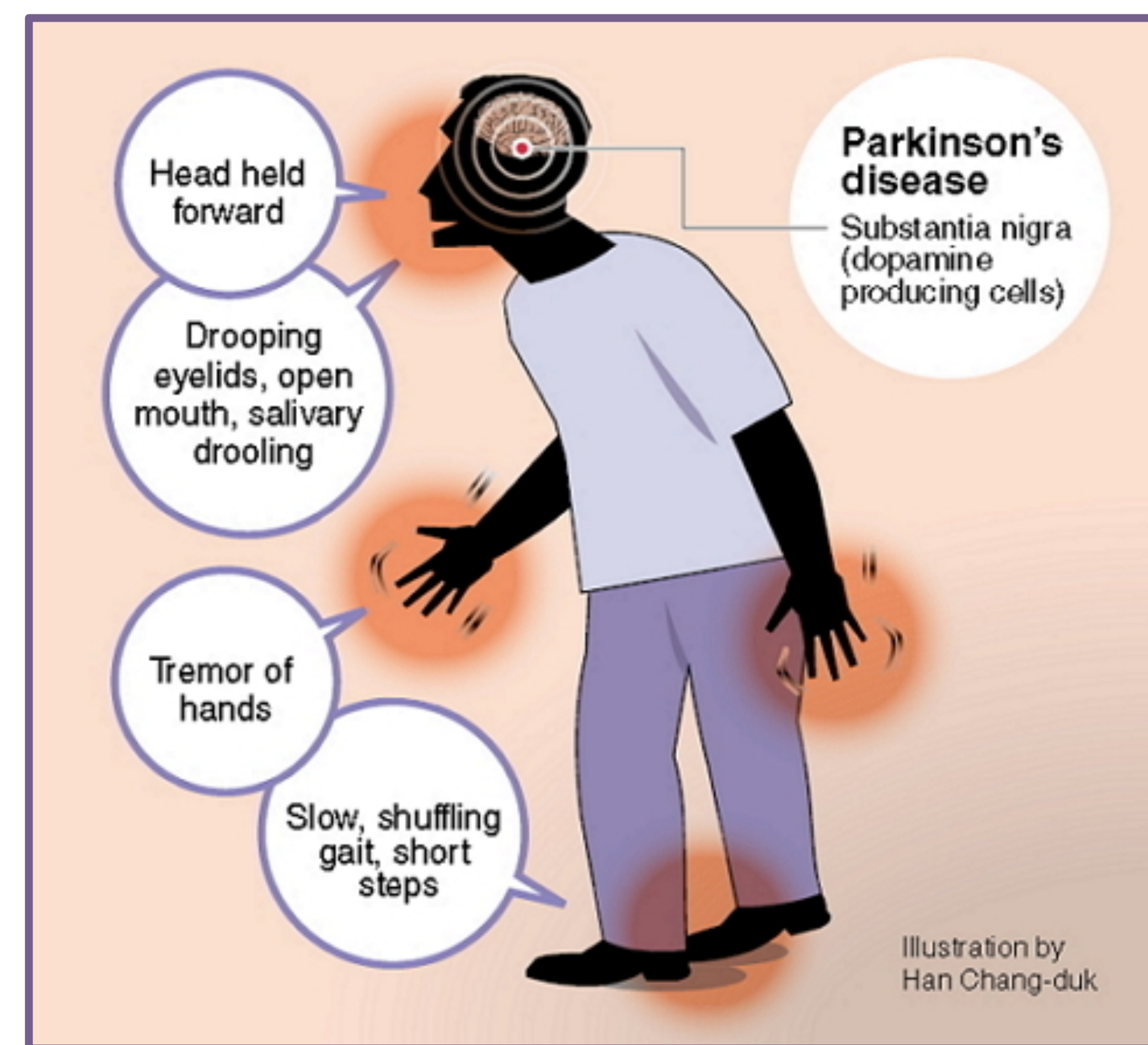
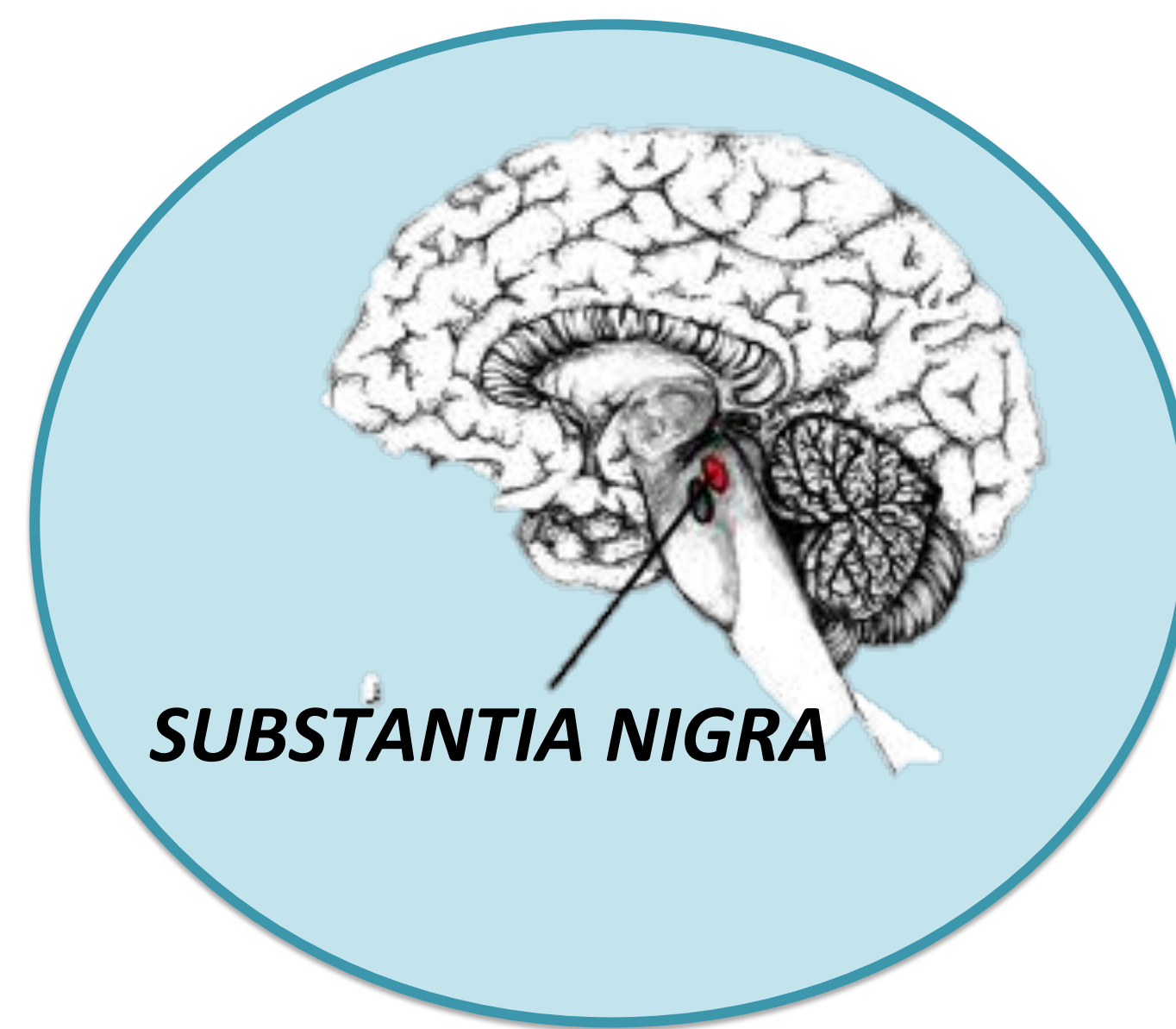
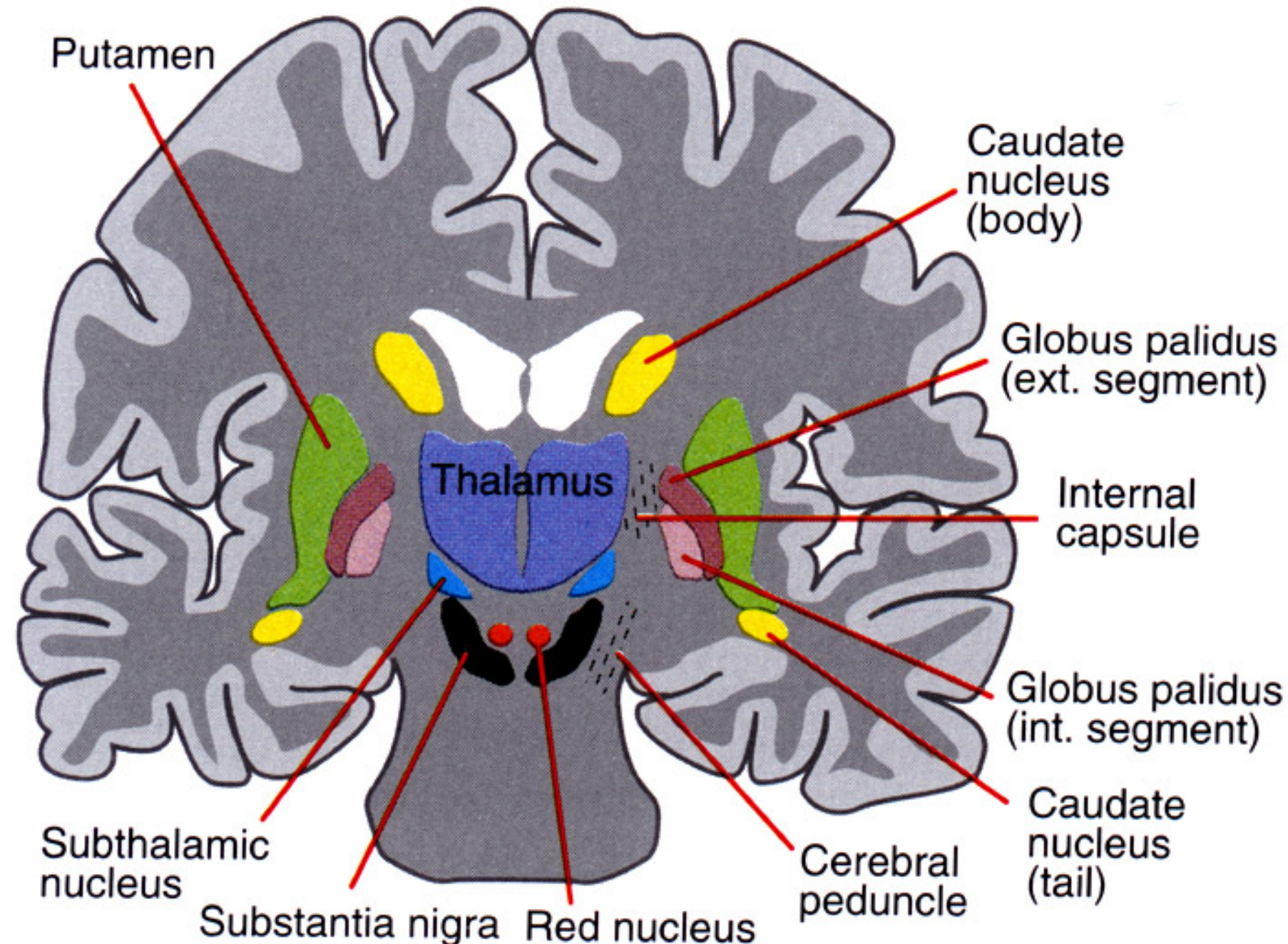
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EVALUATION OF ASTROCYTE ACTION IN A JSK TREATED PARAQUAT RAT MODEL OF PARKINSON'S DISEASE



INTRODUCTION

Parkinson's Disease (PD) is the second most common neurodegenerative disorder, affecting 7 to 10 million individuals worldwide.^[1] This chronic, progressive disease is associated with the degeneration of dopaminergic neurons in the substantia nigra pars compacta region of the basal ganglia. This structure gives rise to motor, associative and limbic circuits, which incorporate the thalamus and cortices.^[2]



PD impacts all three aforementioned circuits leading to symptoms such as resting tremors on one side of the body, hyposmia (reduced ability to smell), muscle rigidity, bradykinesia which is slowed motion and a limited range of movement, as well as mood and REM behaviour disorders.^[1]

Approximately 90% of PD cases are sporadic^[3] and oxidative stress has been implicated as a key player in these situations. However, other known and unknown factors are assumed to be involved and the overall pathophysiology of the disease is not well understood. Hence our lab began to consider the idea of a more holistic and natural approach to treating PD.

Chinese culture has utilized natural herbal remedies since ancient times and several concoctions have proved beneficial for treating illness. In 2013, a novel herbal formulation known as Ji-Su-Kang (JSK) showed positive results in healing damage due to acute spinal cord injury (SCI) in rats, and proved beneficial for a few PD patients.^[4] Therefore, this proposal suggests the use of JSK treatment in a paraquat rat model of Parkinson's Disease.



OBJECTIVES

- Understand if JSK can provide neuroprotection
- What mechanisms are employed by JSK if it possesses neuroprotective activity

Astrocytes are the cells of interest in this project as they are activated in response to inflammation, which is a primal immune response of the body. Activated by the lack of neurotransmitter binding or the presence of abnormal molecules, astrocytes release neurotrophic factors. Glial cell-derived neurotrophic factor (GDNF) and brain cell-derived neurotrophic factor (BDNF), are two important astrocyte molecules responsible for reducing inflammation and protecting neurons.^[5]

Ultimately, the goal of this study is to assess the role of JSK in astrocyte activation using a prophylactic paraquat rat model of Parkinson's Disease.

REFERENCES

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- 2) Galvan, A., & Wichmann, T. (n.d.). Pathophysiology of Parkinsonism. Retrieved March 18, 2015, from <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2467461/>
- 3) Bonifati. (2012) Parkinsonism Relat Disord. Jan;18 Suppl 1:S4-6
- 4) Su, C., Zhang, D., Truong, J., Jiang, C., Lee, S., Jarouche, M., ... & Jiang, S. (2013). Effects of a novel herbal formulation JSK on acute spinal cord injury in rats. *Restorative neurology and neuroscience*, 31(5), 597-617.
- 5) Microglia. (2011, January 1). Retrieved March 17, 2015, from <http://www.networkglia.eu/en/microglia>

METHODOLOGY

18 male rats were divided into three groups and subjected to a prophylactic methodology. Prophylactic studies aim to quantify the maximum neuroprotective capacity of the compounds of interest, by starting treatment prior to inducing disease. Neurotoxin-induced PD is commonly used in laboratory rat models and the four most frequently used parkinsonian neurotoxins are 6-OHDA, MPTP, rotenone and paraquat (used in this study).

GROUP 1

The first group received regular jello (no JSK added) and saline injections. This group allowed any effects of regular jello to be determined and functioned as a control for the stress of injections.

GROUP 2

This second group received regular jello and paraquat injections, acting as a control to assess maximum damage due to the neurotoxin.

GROUP 3

The third group to which JSK supplemented jello and paraquat injections were administered, functioned as the primary experimental group for which JSK neuroprotection was evaluated.



- Strawberry jello is used as a vehicle to administer 0.15g of JSK daily to Group 1
- 22mL of jello are added to each well of the icecube tray, followed by JSK addition
- Groups 2 and 3 receive jello without JSK supplementation

WEEKS 1-4

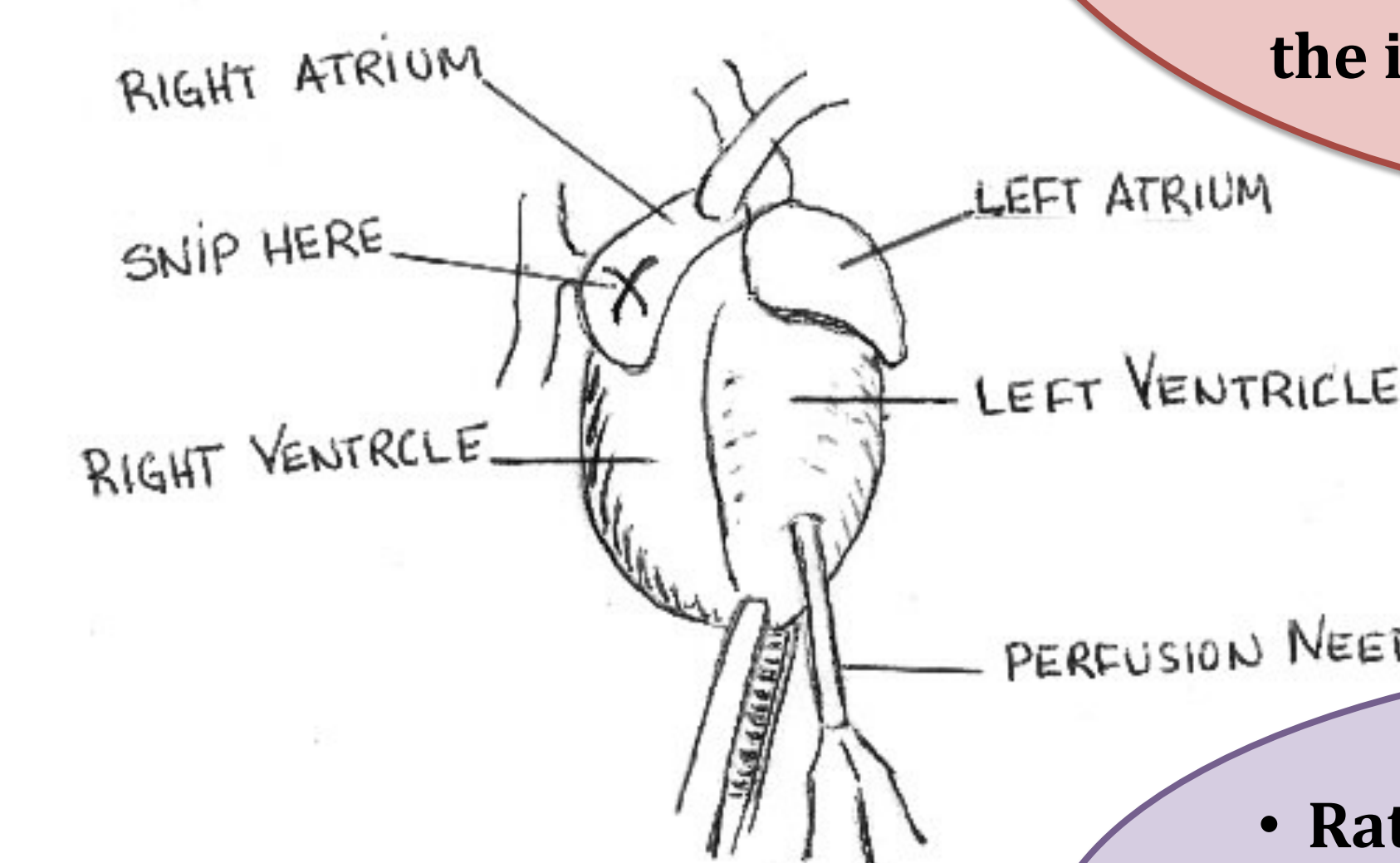
The key contents of the JSK formulation: Ginseng, Chuanxiong Rhi-zoma (chuan xiong), Glycyrrhizae Radix (gan cao), Paeoniae Alba Radix (bai shao) and Cinnamomi Cortex (rou gui). The final product was purchased in powdered form and made into a paste.

- 1 intraperitoneal paraquat or saline injection every 5 days
- Dose is 10mg/kg
- Total of 5 injections are given and treatment continues throughout the injection period

WEEKS 5-7



- Treatment continues

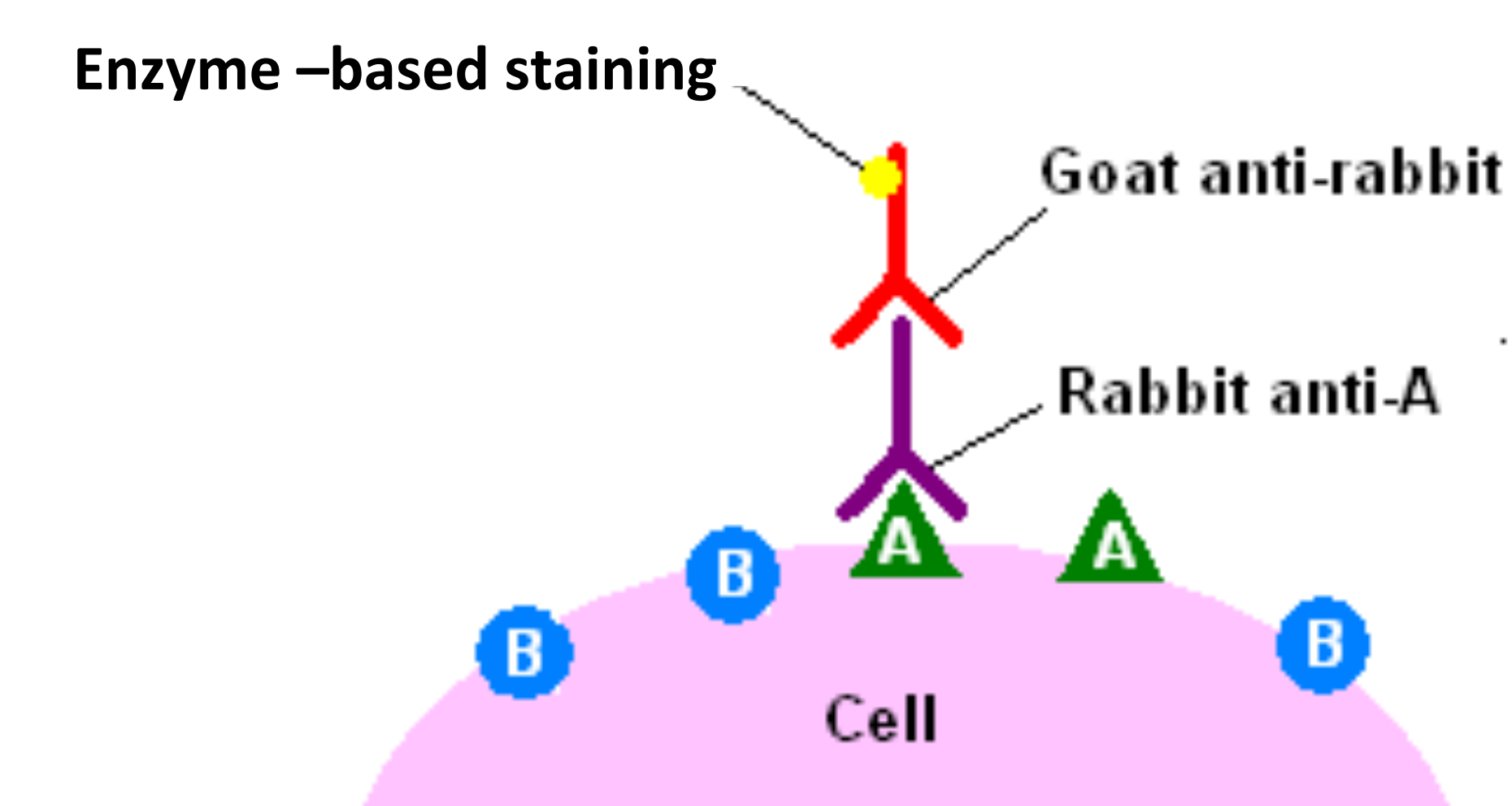


WEEKS 8-11

- Rats are sacrificed in the perfusion process → brains obtained
- Brains are preserved in formaldehyde



PERFUSION



SECTION

- Substantia nigra region of the brain is sectioned and subjected to immunohistochemistry
- Immunohistochemistry with anti-GFAP, anti-BDNF, anti-GDNF antibodies
- Analyze stained brains under microscope

