Case Study: Models in Neurobiology (Part 1)

Methods in Neurobiology

Overview

In this assignment you will set up a model to answer a specific biological question and explain why this model is more appropriate than others. This a 20-point assignment.

Instructions

1. Pick a biology topic. It does not have to be new for you. You can select a topic you have experience with from your personal or professional life, you are interested in, or have strong feelings about.
2. Pick a biology question related to this topic, such as how you would continue the research on this topic.
3. Build and propose a model among those presented in Module 2 (primary or secondary cell lines, tissue slices in culture, iPSCs) that will help to study and solve your biological question. Explain why you picked this model, what you are trying to achieve and what this model will mean for your research.
4. Your work should be around 1 page max (without citations). Include citations in your background paragraph (same format as in scientific articles. Only the web link or Doi number is not sufficient).
5. It is NOT ok to copy from a published article, but it is ok to take inspiration from it. For example, you can use the same topic but change the aim or apply their model to another topic. Make sure though that your work is substantially different from that paper.

Example One

Question/Biological Problem (10 points)
Amyotrophic lateral sclerosis (ALS) is a fatal adult-onset neurodegenerative disorder characterized by the progressive loss of motor neurons in the brain, brainstem, and spinal cord, which culminates in paralysis and death within a few years of diagnosis[1]. New technologies for gene mapping have enabled the identification of nearly 30 genes in ALS pathogenesis. The first gene discovered associated with familiar forms of ALS is SOD1[2]. To date, over 180 different mutations have been described in SOD1 gene whose function is still unknown. One of the earliest and most common mutations discovered is a glycine to alanine substitution in position 93 of the protein.

AIM: To build a model to mimic how the mutant form of SOD1, SOD1 G93A, can cause cellular death and impact normal function of the endogenous, wild-type form of SOD1.

Research Model and Plan (10 points)

- Through transgenesis, a DNA vector carrying a mutant copy of human SOD1 G93A gene will be inserted stably into human neuroblastoma SH-SY5Y cells, an immortalized cell line commonly used to study brain disorders. After transgenesis, cells will be selected using appropriate selection markers (such as gene products to antibiotics resistance). Within the same DNA vector, the SOD1 mutant will be tagged at the C-terminal with a fluorescent protein such as GFP to facilitate identification of cell clones in live cell imaging. Thus, this line will become a stable cell line for mutant SOD1 expression and can be used to study how mutant SOD1 impacts the function of the endogenous wild-type form of the gene.
- This cell line is the most appropriate model compared to primary cell cultures because immortalized cell lines can be cultured indefinitely and do not require repeated human or animal sampling.
- Compared to brain slices that require more advanced/complicated techniques, it is easier to express exogenous genes in cell lines.

1) Beckman JS, Estévez AG, Crow JP, Barbeito L. Superoxide dismutase and the death of motoneurons in ALS. Trends Neurosci. 2001 Nov;24(11 Suppl):S15-20. doi: 10.1016/s0166-2236(00)01981-0.
2) Sau D, De Biasi S, Vitellaro-Zuccarello L, Riso P, Guarnieri S, Porrini M, Simeoni S, Crippa V, Onesto E, Palazzolo I, Rusmini P, Bolzoni E, Bendotti C, Poletti A. Mutation of SOD1 in ALS: a gain of a loss of function. *Human Molecular Genetics*, Volume 16, Issue 13, 1 July 2007, Pages 1604–1618. doi:10.1093/hmg/ddm110

Example Two

Question/Biological Problem (10 points)
Recently, the efficacy of antidepressant agents has been a matter of controversy. The selective serotonin reuptake inhibitors (SSRIs), such as citalopram and fluoxetine (Prozac), are currently considered to be a first-line therapeutic tool for the treatment of depression, even though less than 40% of treated patients respond to this type of antidepressant[1]. SSRIs are thought to moderate depressive symptoms by mainly enhancing the availability of synaptic serotonin (5-HT). However, several findings support the contention that certain SSRIs can block other monoamine transporters, such as those implicated in noradrenergic transmission.

AIM: Build a model to test how SSRIs such as fluoxetine influence the noradrenergic system.

Research Plan/Model (10 points)

- To test how SSRIs such as fluoxetine can influence the noradrenergic system, I need to build a model that can mimic the cytoarchitecture of the noradrenergic circuits. Acute brain stem slices obtained from wild-type mice are a model rich in noradrenergic terminals since the locus coeruleus, which is in the pons area of the brain stem, is one of the main sites of the noradrenaline innervation.
- Fresh brain stem slices of about 250 μm of thickness maintained in complete cell media, will be treated with increasing concentration of fluoxetine and spontaneous firing rate of noradrenergic neurons will be measured through electric recordings. If SSRIs influence noradrenergic transmission, a decrease activation of noradrenergic neurons should be recorded.
- I chose this model because brain slices provide the same cyto-organization of the tissue/organ from which they are derived, compared to dissociated cells in cultures and it is easier to selectively record firing potentials by noradrenergic neurons.
- Brain Stem dissociated cultured neurons do not retain tissue cytoarchitecture and present a larger variability in terms of type of neurons.

[1]Celada P, Puig M, Amargós-Bosch M, Adell A, Artigas F. The therapeutic role of 5-HT1A and 5-HT2A receptors in depression. *J Psychiatry Neurosci*. 2004;29(4):252-265.