11/6/67

~~**~~ * ~~Idea~~ *

1) Get dividing neurons in vitro by fusing (cell hybrids) neurons + growing cell line via UV inactivated Sendai or Parainfluenza type 3 virus.

Need purified fusing factor from virus

a) Cloning in vivo
 1) Ascites ~~tumor~~ + Neuron this in peritoneal cavity
 2) Same solid
 3) In vitro

b) Assay or select for line until can ~~[illegible]~~
 1) ~~Still synthesize~~ secrete neurohormones. function still intact. [illegible]

 2) ~~Still react~~ to neurohormones. Receptor function intact

Ultimate 3) Still [illegible] 1 cell type or 2 cell types mixed

\+ 2) Use UV inactivated Sendai to fuse presynaptic ~~isolated [illegible]~~ terminals. Get functional circuits. Build diff kinds of circuits ~~[illegible]~~ mixing diff purified preps.

3) ~~Isolate~~ 1 kind of neuron (preparation). Fuse cells (very, very).
 1) ~~Look~~ for very large cells (syncytium). [illegible] or
 2) What is maximum size of syncytium possible.

3) ~~Selection technique~~: Neural Function
~~No selection, Many clones~~
1) Produce ~~neurohormone.~~ Excrete
1) A Choline, Have Chol. Esterase in agar. (~~Make black mpt of cholinest Acetate~~). Dye, Turns color due to acid production when Ach hydrolyzed

2) Form Synapses. — Morphology

3) Uptake Neurohormone
a) Hormone impregnated

4) Replica plate (or use 2 thin agar plts) Sensitivity to inhibitors of neurons. 1) Tetrodotoxin
2) Eserine

Use filter paper under agar. Remove paper after clones grown. Paper will pick up diffused hormones.

5) Stain part of culture — Neuron stain.

3) Make colored Neurons for use in tracing connections. Fuse with a pigmented cell line. Retinal pigment cells in culture.
Melanoma.

a) Use analogical technique sort cells very dilute sol. contrast flow apparatus which will get insanity

4) Red Blood Cells (– nucleus but large) + Neurons.

4) Have 1) micro electrodes, attached sites in large wounds + Neurons + Virus.
2) Dye in small coating wounds. Uptake of dye.
) Method for fixing cells to ~~stated~~ sites which are monitored.

5) Try Embryonic Neuroblasts + in vitro Cell line + Virus