CALIFORNIA STATE SCIENCE FAIR
2012 PROJECT SUMMARY

Name(s)

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Project Number

S0507

Project Title

New Mechanism and Neuronal Activation of Brain by Peripheral Leptin: Potential Applications in Obesity

Abstract

Objectives/Goals

Failures to maintain energy balance contributes largely to obesity. The body produces Leptin,a protein in proportion to food intake and stored adipose mass, and acts on brain to maintain energy homeostasis. General understanding about Leptin#s action on brain is that it penetrates BBB. In this study,I hypothesize that CSF is an alternative route for Leptin to act on the brain. My research is to help understand the mechanism of leptin action which should be helpful in developing new drugs to treat obesity.

Methods/Materials

Method:Whole rat brains previously dosed with Leptin used from ongoing institutional research were acquired and 30 microm thick coronal tissue sections were collected in antifreeze solution.Pre-staining was done, and tissues were incubated with goat serum and Triton X. Between each step multiple PBS rinses were performed.Sections were incubated with a pSTAT3 antibody.The sections were incubated in a secondary antibody followed by incubation in ABC solution. Biotinylated tyramide and H202 were added followed by streptavidin FITC conjugate. Sections were rinsed, mounted, and images were acquired with confocal microscopy.

Materials:Rat brains,Tissue slicer,Anti-freeze solution,NaOH,H(2)0(2),Glycine,SDS,Triton X-100,Goat serum,Rabbit anti pSTAT3 antibody,Biotinylated Anti-Rabbit IgG antibody,ABC complex,Biotinylated Tyramine,DAB,Nickel Sulfate,Streptavidin FITC conjugate,Glass Slides,Well Plate,Staining Mesh,Incubator,ProLong antifade,DAPI,and Confocal microscope

Results

Using pSTAT3 as a marker for Leptin activation and by constant repetition, extensive experimental conditions,I was able to see Leptin-neuronal expression throughout all ventricular regions of the brain, thus proving my hypothesis that the CSF is the pathway of Leptin entry and neuronal activation in the brain.

Conclusions/Discussion

Since neurons of the hypothalamus have Leptin receptors and the CSF has significant amounts of Leptin, I hypothesized that I would observe Leptin-neurons along the ventricular regions of the brain by following the pSTAT3 expression via immunohistochemistry.

My research conclusively proved that leptin activates key neurons in the brain via the CSF pathway.This research has significant implications and will alter the focus for future Obesity studies.To the best of my knowledge this is the first report describing CSF as an alternate route of entry for mechanism of action of Leptin.

Summary Statement

This is a novel mechanism of leptin action in the brain using the Cerebrospinal fluid as an alternate route, which activates key neurons in the ventricular regions to maintain energy balance.

Help Received

Used laboratory faciltes at Amylin Pharmaceuticals under the supervision of Dr. Guibao Gu (MD);