Prenatal study of the Gli family proteins in the cerebellar development under weaver condition

SERGI ALACID ROMERO, Bachelor’s Degree in Genetics
Facultat de Biociències, Universitat Autònoma de Barcelona

INTRODUCTION
Mice carrying the weaver (wv) mutation suffer from a wide range of symptoms, and all of them result from the death of several cell types in the Central Nervous System (CNS). How the wv allele affects the development and so the phenotype isn’t yet clear. One of the most relevant pathways during cerebellum development is Sonic Hedgehog (Shh), which regulates gene expression through the Gli transcription factor family. These proteins are deeply involved in granule-cell (GC) proliferation, and since their depletion is a major feature of the weaver condition, the study of the prenatal expression of the Gli family could shed some light upon the mechanisms that link the weaver genotype to its phenotype.

The weaver mutation and condition

Girk2
953G>A
Gly153Ser

Cerebellar abnormalities:
Structural deficiencies
Depletion of GCs, PCs and deep cerebellar nuclei neurons

Wild type
Sigal sections of wt and wv cerebellums (Martí J. et al, 2007)

Weaver

Key steps in the prenatal cerebellar development mediated by Gls

1. Patterning and growth of the dorsal mes/r1
   - Gli3
   - E7.5 - E11.5

2. Proliferation of GCP’s in the EGL and foliation complexity
   - Gli2 (6 Gli1 lesser effects)
   - E17.5 - P0 (and beyond)

OBJECTIVES/WORKING HYPOTHESIS
The main objective of this study is to check whether the expression of the Gli proteins in weaver mice is correct or not. More specific goals would be to inquire into the specific effect of each Gli member to the weaver phenotype, by searching expression disturbances at specific stages:
- Gli1 and Gli2 during GCP proliferation in the EGL (E17.5 - P0)
- Gli 3 during the patterning and growth of the dorsal mes/r1 (E7.5 - E11.5)

The working hypothesis of this study claims that Gli transcription factor proteins are indeed affected by the weaver condition during embryonic development and thus, they contribute to the resulting phenotype.

METHODOLOGY

Mutant stock maintenance
First heterozygous mutant parents from Jackson Laboratories ME, USA

Genotyping DNA from tail tissue allows us to characterize and select desired mice embryos

Selected wv/wv and +/- mice grow until desired stage of development

Histology, Immunohistochemistry and RNA in-situ hybridization

Fluorescent microscopy and analysis

Quantitative & Semiquantitative wv/wv vs +/-

EXPECTED RESULTS/DISCUSSION
We expect to confirm our working hypothesis and find significant differences between the expression of Gli1, Gli2 and/or Gli3 from weaver to wild-type mice, at least at some stage of the cerebellar development.

If that’s the case, much speculation on how Girk2 affects Gli expression could be made, based on an hypothetically impaired Shh signaling from PCs to GCs:
- Constitutively activated Girk2 changes PCs’ efferent target microenvironment.
- Constitutively activated Girk2 interferes with the expression of GDNF and/or with the appearance of their receptors in PCs.

It is worth noting that poor research has been done in the context of prenatal life in weaver mice, and thus new discoveries could be done in that field. Furthermore, since weaver serves as an animal model for the cerebellar ataxias, findings in their embryonic life development could help to early diagnose and treat the hereditary cases.

REFERENCES