The identification of protein tyrosine phosphatase, non-receptor type 1 (Ptp1b) in hippocampal modulation of food anticipatory behavior.

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Introduction

Activity based anorexia (ABA) is a rodent model of hyperactivity evoked by food restriction. The food restriction evoked hyperactivity can be expressed as food anticipatory activity (FAA). The paradoxical hyperactivity during food restriction is reminiscent of that observed in patients suffering from eating disorders. Unraveling the genetic basis of this phenotype could help us create better treatments for eating disorder patients. In this study we aimed at finding and testing a novel candidate gene for food restriction evoked high activity observed in ABA.

Methods and results

1. Mice of chromosome substitution strains (CSS) (created using host strain expressing FAA (C57BL/6J) and a donor strain lacking FAA (A/J)) were tested in ABA model. Chr 2 was pointed as containing a genetic region influencing FAA.
2. QTL analysis on F2 population (CSS#2 x C57BL/6J) revealed a region on Chr2 associated with FAA. Microarray study [Affymetrix 430A 2.0] (to identify genes differentially expressed in hippocampus of mice from F2 population selected for high or low FAA) pointed to a candidate gene: Ptpn1, located within the QTL region on Chr2.
3. Involvement of *Ptpn1* in FAA was tested functionally with the use of shRNA gene knock down (KD); delivered in AAV virus [pAAV-shbase containing GFP cassette, titer = 7.8×10^{12} genomic copies /ml]:
   a. virus spread was assessed with anti-GFP ISH
   b. IHC for NeuN and ISH for mir124 showed that there is no neuronal damage,
   c. gene knockdown was confirmed using RA ISH,
   d. FAA levels of *Ptpn1* KD mice are lower than FAA levels of control virus injected mice,
   e. *Ptpn1* expression in DG correlates with FAA level.

Conclusions

Ptpn1 is a novel candidate gene, which expression in the DG is influencing levels of food anticipatory activity in a mouse model of eating disorders.

1

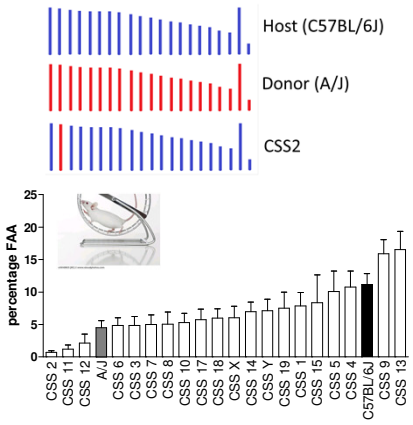

Fig. 1. CSS mice with an A/J chromosome on the C57BL6/J background were tested in an ABA model. CSS2 had significantly lower FAA levels than the host strain (C57BL/6J).

2

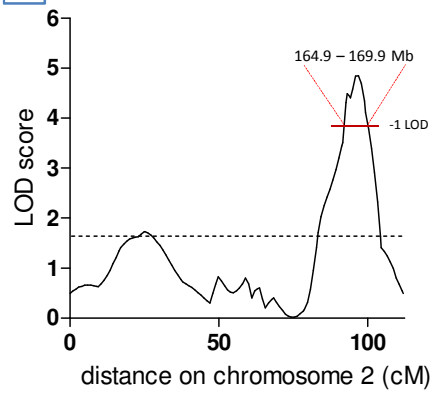

Fig. 2. F2 generation of a cross between C57BL/6J and CSS2 strain was tested in ABA model. QTL analysis showed a locus on mouse chromosome 2 associated with FAA [MQM-MapQTL procedure: significance threshold = 1.65; the peak LOD score = 4.85; CI (minus 1 LOD) = 164.9 – 169.9 Mb.

3

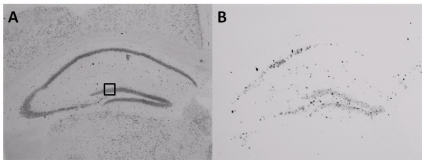

Fig. 3a. AAV virus spread in hippocampus. A) Nissl staining, B) GFP ISH. The black square in figure A indicates the injection site.

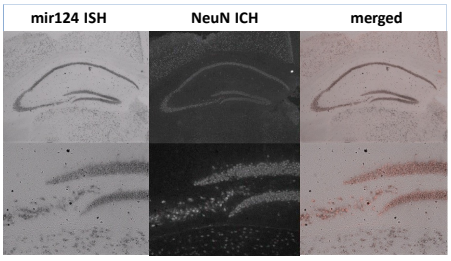

Fig. 3b. AAV virus injection in hippocampus caused no detectable neuronal damage.

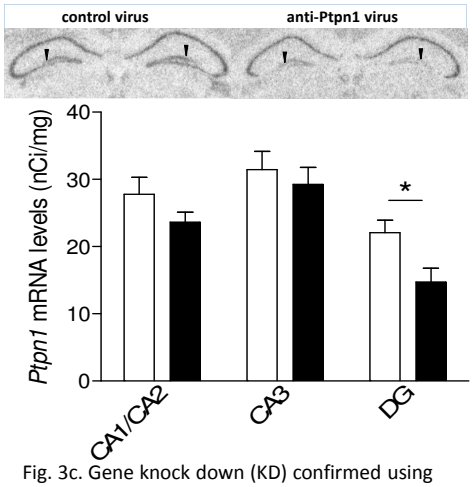

Fig. 3c. Gene knock down (KD) confirmed using radioactive (RA) ISH. Upper panel: example photos of RA ISH; arrow heads indicate injection sites. Lower panel: quantification of the KD.

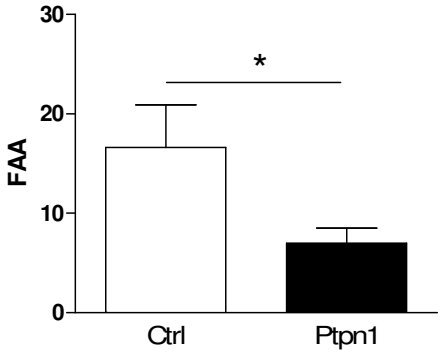

Fig. 3d. FAA is diminished in C57BL/6J mice injected with anti-Ptpn1 shRNA virus in the dentate gyrus.

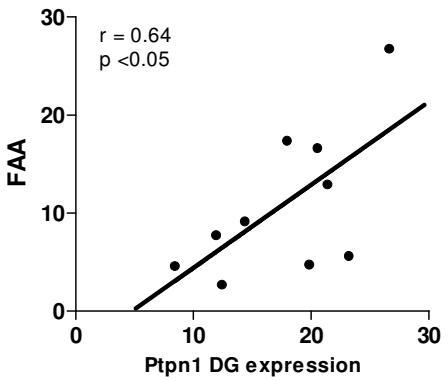

Fig. 3e. The expression of *Ptpn1* in the dentate gyrus (DG) of the hippocampus correlates with the levels of FAA.

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