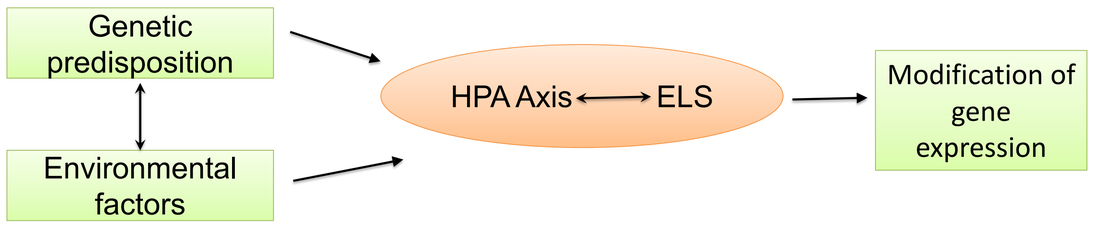
Psychiatric risk gene *Cacna1c* and early life stress: potential gene-environment interaction?

Moon A, Haan N, Brydges N, Thomas K, Hall J
Neuroscience and Mental Health Research Institute, Cardiff University, UK

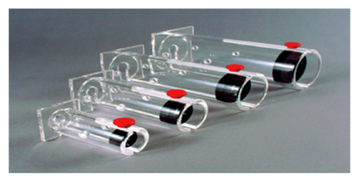
Introduction

- **Early life stress (ELS)** is highly associated with **development of psychopathology and mood disorders** in adulthood [1]
- Environmental factors can interact with genes, e.g. environment can lead to significant risk gene expression changes or a genetic factor can influence sensitivity to a particular environment
- Genetic studies have identified variation in the gene *calcium voltage-gated channel subunit alpha1C* **(CACNA1C)** to increase risk for several psychiatric disorders [2].

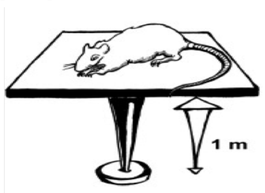


- *CACNA1C* encodes an **alpha-1 subunit of voltage-gated calcium channels** (VTCCs), which mediate calcium influx into cells.
- In humans**, SNPs in *CACNA1C*** has been shown to interact with adult trauma to predict depression [3].
- In rodents, acute and chronic stress in rats causes an increase in VTCCs in the hippocampus, cortex and basolateral amygdala and Cacna1c+/- mice have increased susceptibility to chronic social defeat stress [4].
- **Aim: To investigate the expression of Cacna1c following prepubertal stress (PPS)**

PND 25: Forced Swim
(10 minutes, water temperature 25ºC)



PND 26: Restraint tube
(3 x 30 minutes, separated by 30 minute breaks)



PND 27: Elevated platform
(3 x 30 minutes, separated by 60 minute break)

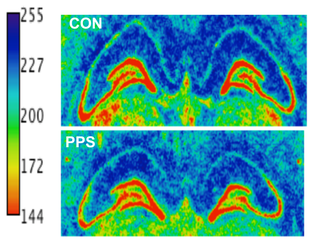
Methods

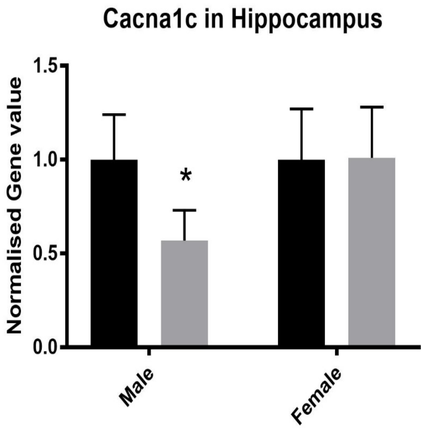
- PPS was conducted on rats from **PND25-27** and mRNA analysed by **qPCR and ISH**. Protein analysis was done by **Western Blot**.
- Cacna1c+/- rats and PPS rats where subject to **trace fear conditioning**. Rats were conditioned with 0.5mA footshock. Bursts of 75-86db white noise formed the CS, and CS-US were separated by an interval.. All animals were presented with 10 CS-US pairings. Recalls took place 24 and 48 hours later.

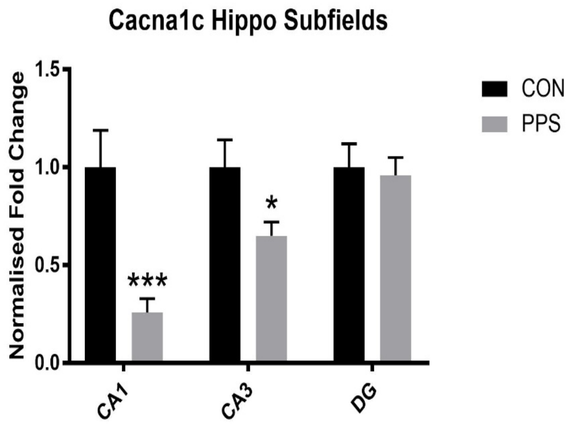
PPS in rats results in a downregulation of Cacna1c in the hippocampus in a sex specific manner

Cacna1c mRNA expression in the whole hippocampus was significantly reduced by 43% in male rats following PPS in comparison to control rats ($F(1, 17) = 6.69$, $p = 0.019$). However, no differences were observed in female rats, suggesting a sex specific effect of stress on *Cacna1c* expression.

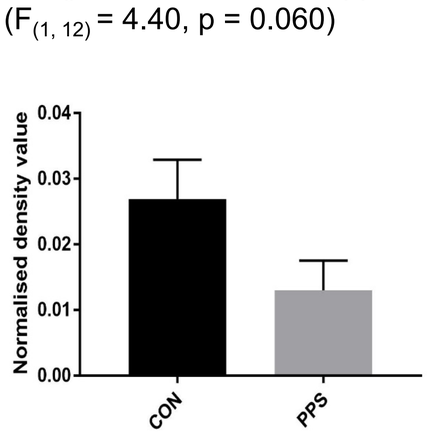
A more specific analysis of the hippocampus revealed that this Cacna1c decrease was specific to CA1 ($F_{(1,17)} = 14.22$, $p = 0.002$) and CA3 ($F_{(1,17)} = 4.99$, $p = 0.040$), with no difference of expression within the dentate gyrus.

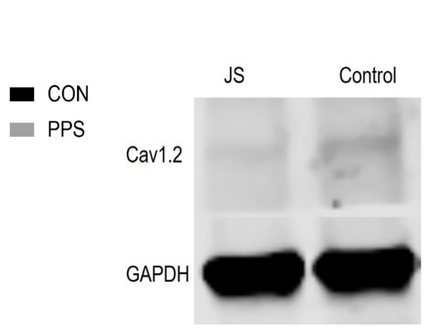


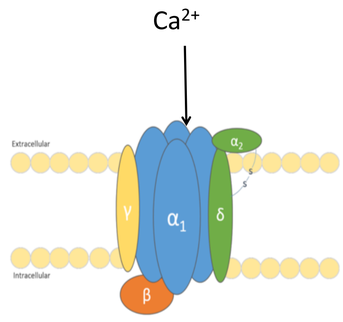




Cacna1c encodes Cav1.2 protein, a subunit within the L-type calcium channel. Cav1.2 trended to being decreased in the hippocampus of PPS male rats in comparison to non-stressed littermates ($F_{(1, 12)} = 4.40$, $p = 0.060$)



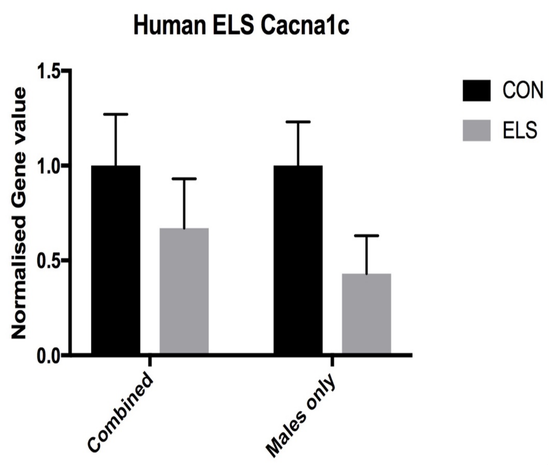




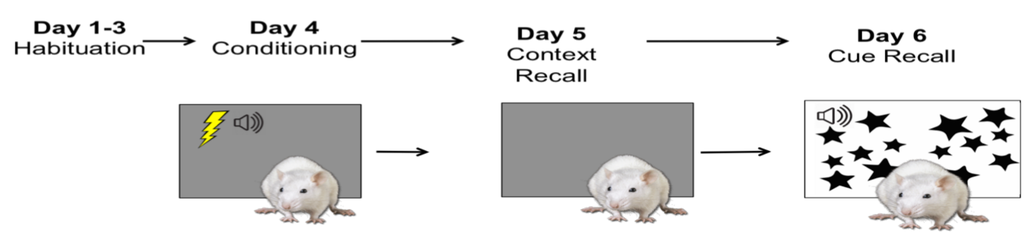
Cacna1c mRNA expression is reduced in humans stratified by early life stress, subject to sex

CACNA1C mRNA expression was investigated in post-mortem hippocampal tissue from subjects who had experienced early life stress (ELS) compared to subjects with no childhood trauma.

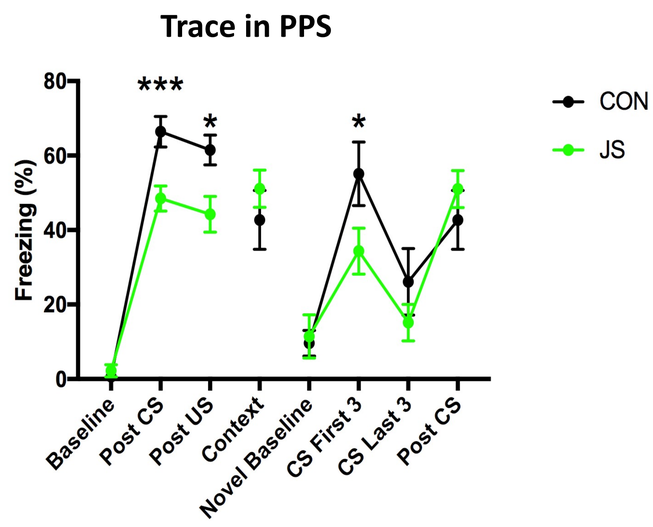
There was a trend to a decrease in *CACNA1C* mRNA expression ($F_{(1, 12)} = 3.26$, $p = 0.098$) in males who had suffered ELS, but when males and females are combined, there was no significant expression difference.



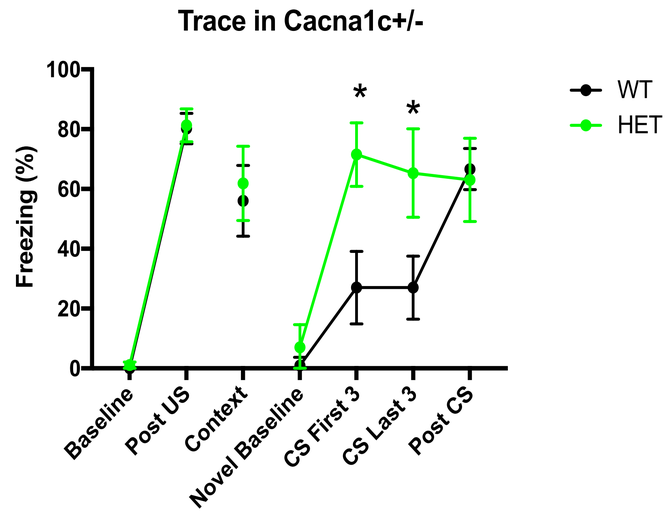
PPS and Cacna1c heterozygosity both result in trace auditory fear conditioning impairments



In trace conditioning, there is a 30 sec gap between white noise offset (CS) and the footshock (US); learning the trace association is both hippocampal and neurogenesis dependent.



PPS animals trained in a trace fear conditioning paradigm showed decreased fear memory encoding during conditioning ($F_{(1,11)} = 10.901$. $p = 0.007$) and during cued recall ($p = 0.047$)



Cacna1c+/- rats also showed deficits in the trace conditioned paradigm; they showed increased fear responses during cued recall, specifically during CS presentation ($p = 0.029$).

Conclusions

- Male rats who has been subject to PPS have decreased Cacna1c mRNA in the hippocampus, specifically the CA1 and CA3. Female rats have intact Cacna1c expression.
- Male rats also trend to a decreased Cav1.2 protein level within the hippocampus
- Human males who had suffered ELS also show a decreased Cacna1c mRNA
- Both PPS and *Cacna1c*$^{+/-}$ rats show deficits in trace auditory fear conditioning, however PPS rats show decreased fear responses whereas *Cacna1c*$^{+/-}$ rats show increased fear responses. .This suggests that impairments in stress pathways and *Cacna1c* affects hippocampal-dependent tasks

References

[1] Carr et al (2013) *Journal of Nervous and Mental Disease* 201:12, [2] PGC (2013) *The Lancet* 381:9875, [3] Dedic et al (2018) *Molecular Psychiatry* 23:3, [4] Mamczarz et al (1997) *Pol J Pharmacol* 49:6