Aberrant *Bdnf* DNA Methylation in the Brain is Mirrored in the Blood Following Early-Life Stress

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Introduction

- Epigenetic alterations brought about by early-life stress may be associated with behavioral and neurological problems later in life.[1,2,3,4]
- As the human brain is difficult to access in living subjects, epigenetic changes in blood are often used as a proxy for that of the brain, where neurological disorders presumably originate.

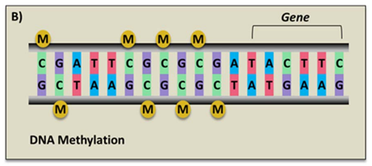

Figure 1. DNA methylation occurs when a methyl group binds to the DNA sequence, typically to a cytosine[5]

- Literature examining whether epigenetic changes in the blood mimic that of the brain is mixed, with some studies finding correlations between blood and brain DNA methylation[6,7,8] and others finding differences in methylation patterns between the two tissues.[9,10]
- Previously, our lab has found a significant increase in DNA methylation at exon IX but not exon IV of the *Bdnf* gene in the prefrontal cortex (PFC) of infant rodents who were exposed to early-life stress.[11,12] The human literature has also found increases in *Bdnf* methylation in blood in adults who experienced child maltreatment.[13,14,15]
- The current study examines how early-life stress impacts DNA methylation of *Bdnf* exons IV and IX in the PFC and in blood of infant rats.

Methods

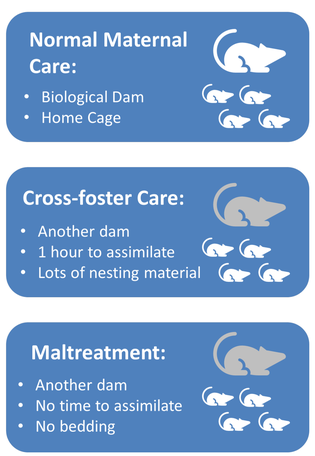

Figure 2. Scarcity-adversity rodent model of low nesting resources outside the home cage

- From PN1 to PN7 for 30 minutes each day, Long-Evans rats were exposed to an early-life stress model (Fig. 2) previously used by our lab.[11,12]
- Whole blood and PFC tissue were extracted on PN8 for subsequent DNA methylation analysis.
- A subset of rodents underwent a saline flush perfusion to rid the brain of any blood that could be altering methylation results.
- DNA extractions and bisulfite conversions were performed with Qiagen AllPrep DNA/RNA (PFC)/Qiagen DNeasy Blood and Tissue and Qiagen Epitect Bisulfite kits, respectively.
- Methylation-specific real-time polymerase chain reactions (MSP) were performed using primers specific for *Bdnf* exons IV and IX.

Results

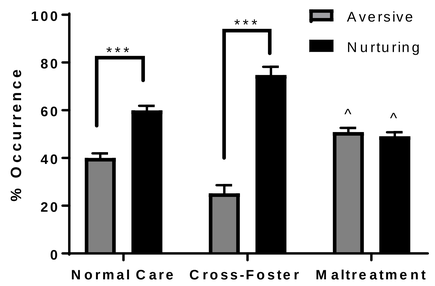

Figure 3. Percent occurrence of aversive and nurturing caregiving behaviors in each infant condition (n=6-7 litters; ***p<0.001, ^p<0.05 versus normal and cross-foster care controls; error bars represent SEM)

(Cross-foster behaviors were significantly different from normal care behaviors)

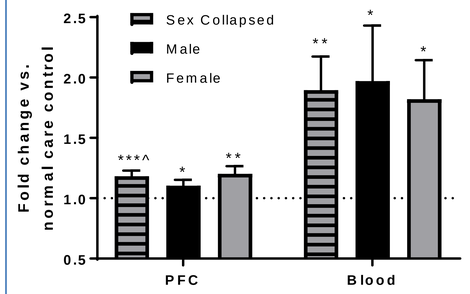

Figure 4. *Bdnf* exon IV DNA methylation in the PFC and in blood in maltreated animals collapsed across perfusion groups (n=12-21 per group; *p<0.05 versus normal care controls; **p<0.01 versus normal care controls; ***p<0.001 versus normal care controls; ^p<0.1 versus cross-foster care controls; error bars represent SEM)

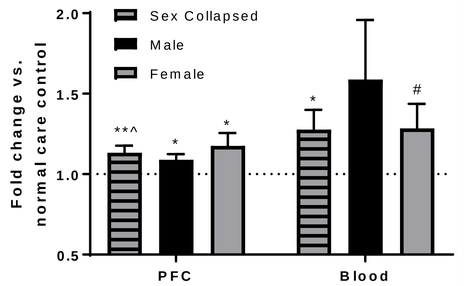

Figure 5. DNA methylation in the PFC and in blood in maltreated animals at exon IX collapsed across perfusion groups (n=12-21 per group; *p<0.05 versus normal care controls; **p<0.01 versus normal care controls; #p<0.1 versus normal care controls; ^p<0.1 versus cross-foster care controls; error bars represent SEM)

Conclusions and Future Directions

- PFC and blood tissue show similar increases in *Bdnf* DNA methylation in the maltreatment group compared to normal care controls at both exons IV and IX.
- These results suggest that blood may indeed be a reliable proxy for *Bdnf* DNA methylation changes in the brain.
- This is a promising finding for the human literature that relies on using blood to investigate neurological disorders, many of which may be influenced by early-life stress.
- There was no main effect of perfusion/no perfusion, suggesting that blood in the brain does not interfere with brain DNA methylation results.
- An increase in DNA methylation at exon IV in infancy was a novel finding for our lab, but in line with the adult literature in both rodents and humans.[11,13,14,15]
- Future studies should investigate correlations between blood and PFC CpG sites within exons IV and IX of *Bdnf*.

References and Acknowledgements

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