Proteomic Insights into the Developmental Pathophysiology of Stereotypy

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Background and Rationale

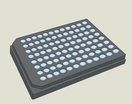
- Stereotypies (repetitive, unvarying, and seemingly goal-less behaviors) are cardinal to several neuropsychiatric and neurodevelopmental disorders[1,2] and are associated with distressing affective states[3].
- Spontaneous stereotypy in animals is a well-validated model for human stereotypy including shared pathophysiology of the pre-motor corticostriatal loop and behavioral deficits[2,3].
- Despite extensive literature examining stereotypy, the underlying developmental pathophysiology is unknown.
- Compulsive behaviors, which involve dysfunction of an adjacent corticostriatal loop[2] are strongly correlated with REDOX imbalance (oxidative stress), and symptom severity is reduced via antioxidant therapy[4].
- Therefore, given the conservation and homologous nature of the corticostriatal loops[5], we hypothesized that REDOX balance would also predict severity of stereotypy.
- Here, we tested if REDOX imbalance, quantified via plasma glutathione (GSH), is predictive of severity of stereotypy.

Does GSH predict severity of stereotypy?

N = 20 C57Bl/6 mice (10 M; 10 F) were acclimated to cages for 7 days.

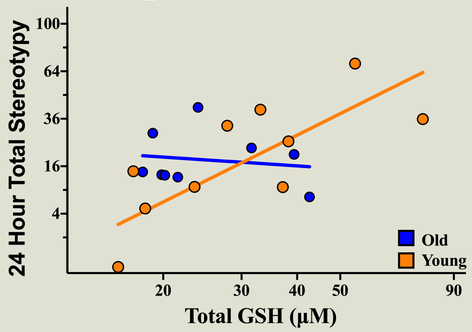
Home-cage behavior was recorded and quantified for severity of stereotypy.

Plasma was collected and stored at -80°C for future analysis.



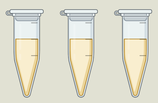
GSH was quantified using a commercially available kit.

GSH predicts severity of stereotypy, but relationship diminishes with age.



Plasma-based total GSH positively predicts severity of stereotypy in younger (T = 3.6467; P = 0.0033), but not older, mice (T = -0.7191; P = 0.4845).

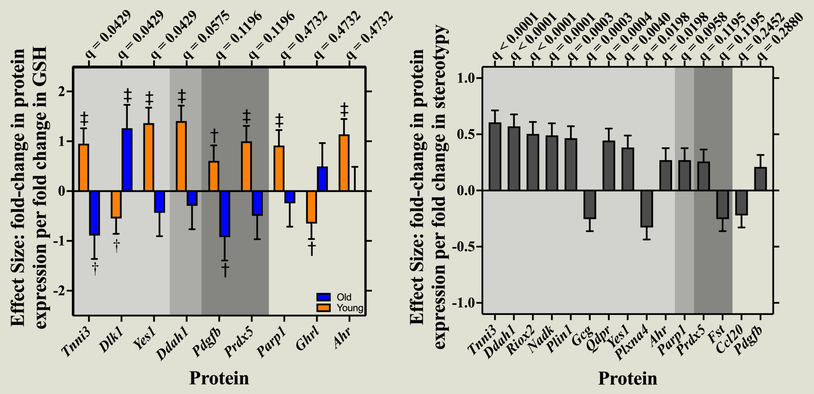
Is there a more specific and sensitive proteomic biomarker profile?



N = 18 plasma samples were sent to Stanford University Human Immune Monitoring Center for proteomic analysis.

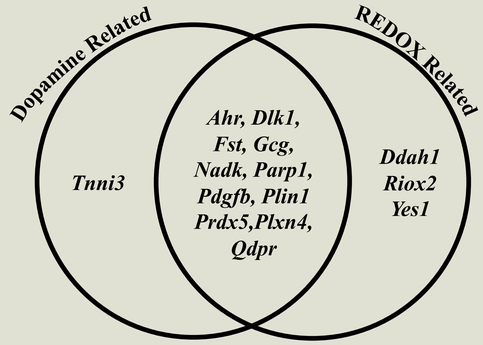
Expression of 96 proteins was quantified in parallel using the OLINK mouse exploratory panel.

Different proteomic signatures correlate with GSH and stereotypy.



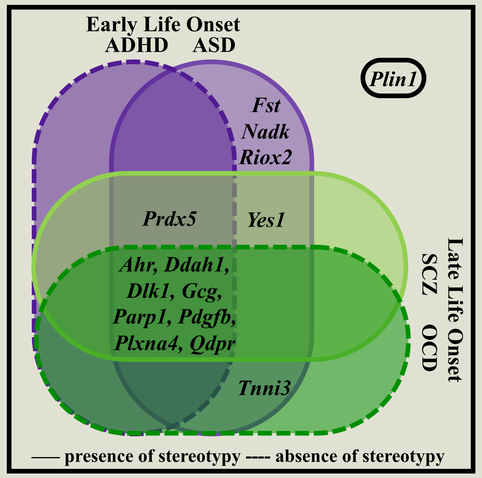
As a function of age, expression of 9 proteins correlate with plasma-based total GSH ($F_{91,1274}$ =1.15; P = 0.0023). Independent of age, expression of 15 proteins correlate with severity of stereotypy ($F_{91,1274}$ = 3.15; $P < 0.0001$). Benjamini-Hochberg (BH) corrected q-values depicted above graphs. Light gray = pass BH correction at q < 0.05; medium gray = pass at q < 0.10; dark gray = pass at q < 0.20.

Proteomic hits are related to dopamine metabolism and/or REDOX processes.



All proteins passed BH correction at q < 0.20 in at least one analysis.

Proteomic hits are validated by association with disorders characterized by stereotypy.



Proteomic biomarkers are highly specific to disorders where stereotypy is a cardinal sign and onset is early in life. Only *Tnni3* and *Plin1* do not fit this pattern.

Conclusions & Future Directions

- Plasma-based total GSH is predictive of severity of stereotypy, but the predictive nature diminishes with age.
- The relationship between GSH and stereotypy in mice parallels the relationship between GSH and compulsive behaviors in mice and humans.
- Proteomic hits tightly correlate with dopamine, REDOX processes, and disorders characterized by stereotypy.
- Together, these results support a REDOX imbalance developmental pathophysiology for stereotypy.

Future Directions:

1. Translate findings to other species (e.g. primates and humans).
2. Determine if antioxidant therapies are effective for stereotypy as they are for compulsive behaviors.

References:

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3) Garner, J.P., Meehan, C. L., Mench, J. A. (2003) *Behav. Brain Res.* 145(1): 125-134
4) Vieiera et al., (2017). *PLOS ONE*. 12(4): e0175222
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Methods figures were produced using BioRender.com

This work was supported by the Stanford University Department of Comparative Medicine and philanthropic gifts to JPG to support animal welfare research.