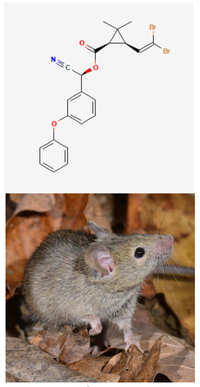
Developmental pyrethroid exposure disrupts molecular pathways for circadian rhythms and MAP kinase in mouse brain

Jennifer H. Nguyen[1], Melissa A. Curtis[1], Ali S. Imami[1], William G. Ryan[1], Khaled Alganem[1], Kari L. Neifer[1], Nilanjana Saferin[1], Charlotte Nawor[2], Brian P. Kistler[2], Gary W. Miller[3], Rammohan Shukla[1], Rob McCullumsmith[1], James Burkett[1]

[1]University of Toledo College of Medicine, Department of Neurosciences; [2]Department of Medicine; [3]Columbia University

COLLEGE OF MEDICINE AND LIFE SCIENCES
THE UNIVERSITY OF TOLEDO

INTRODUCTION



- Neurodevelopmental disorders (NDDs) are lifelong, incurable brain disorders with few biomarkers and few treatments[1]. The incidence of NDDs is rapidly rising, with 17% of children in the US now affected[2].
- Developmental pyrethroid exposure **(DPE)** results in an increase in dopamine transporter that directly causes an ADHD-like behavioral phenotype in mouse[3].
- **Two recent epidemiological studies** have linked pyrethroid pesticides with autism risk[4,5].

Key experimental question:

- Does developmental exposure to pyrethroids cause **brain-wide molecular changes**

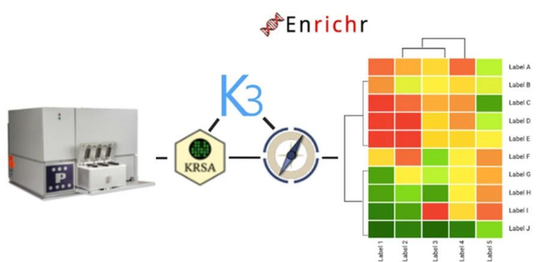
EXPERIMENTAL DESIGN

Pesticide exposure

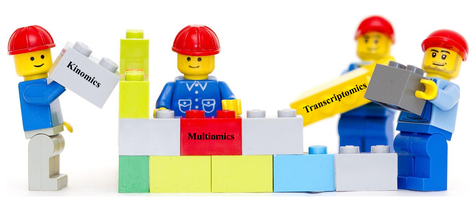
Transcriptome

24-Hour Mobility

Kinome



Multiomics



RESULTS

FIGURE 1: DPE causes transcriptional changes to clock genes

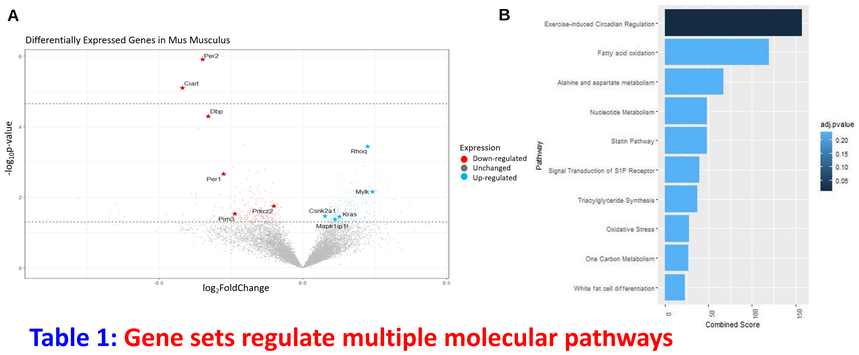


Table 1: Gene sets regulate multiple molecular pathways

A CHEA3 Analyses

Rank	TF	Score
1	SOX18	37.33
2	FOSB	41.67
3	PRRX1	46
4	ZNF524	51.33
5	MYC	57.33
6	ZNF326	67
7	JUN	70.33
8	TWIST1	74
9	NR4A1	79.5
10	ZNF672	83.67

B KEA3 Analyses

Rank	Protein	Mean rank
1	CSNK1D	33.1
2	CSNK1E	33.91
3	PRKDC	35.91
4	MAPK1	44.45
5	EGFR	45.73
6	ATM	47.82
7	SRC	51.18
8	CSNK2A1	55.09
9	CSNK1A1	55.64
10	MASTL	57.75

Key: Regulates
- synaptic plasticity
- adult neurogenesis
- circadian rhythm

FIGURE 2: DPE increases activity in kinases regulating synaptic plasticity

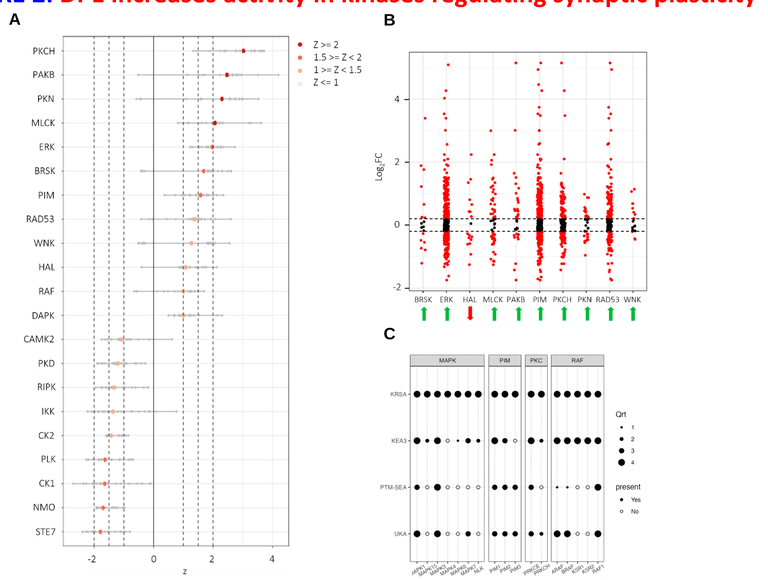


FIGURE 3: DPE causes multi-modal changes in molecular pathways

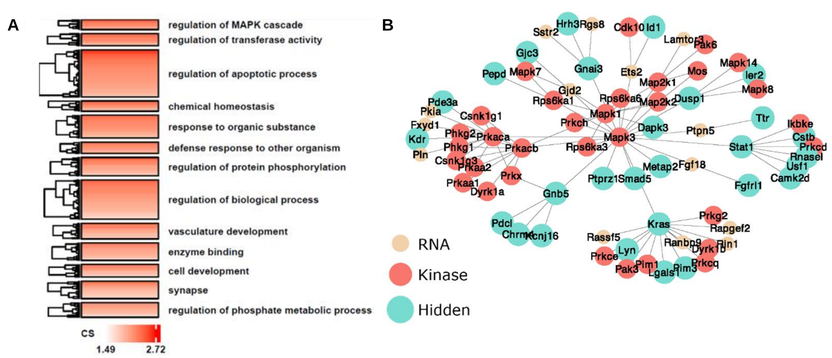


FIGURE 4: DPE causes changes at the metabolome level

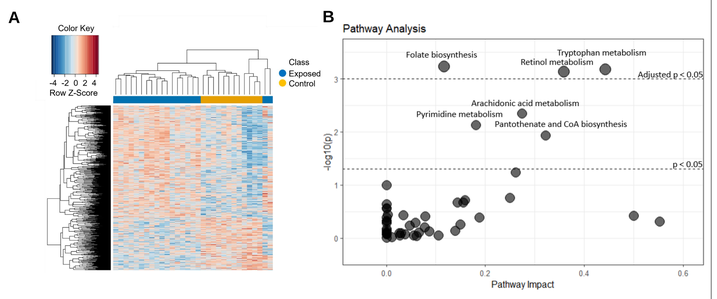
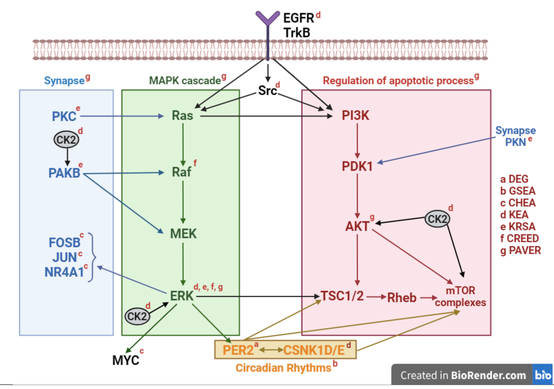


FIGURE 5: Multi-modal changes in circadian rhythms, MAPK, growth/apoptosis, and synapse function



CONCLUSIONS

DPE may alter circadian rhythm

- Genetic results from transcriptome in mouse show significant disruptions in two CLOCK genes and a pattern of changes in genes of interest concentrating in circadian rhythm gene sets.

DPE may alter synaptic plasticity

- Disruptions in synaptic plasticity and changes in dendritic spines have been implicated in the etiology of autism.
- All seven kinases with increased kinase activity in DPE mice have roles in synaptic plasticity, and synapse function was identified as a significant cluster in the multi-omics network.
- This broad increase in kinase activity may reflect a biophenotype of dendritic spine overgrowth and/or decreased synaptic pruning, as is seen in autistic patients and some mouse models.

DPE may alter folate biosynthesis, retinol metabolism, and trypophan metabolism

- Metabolomic results showed DPE mice had altered folate biosynthesis, a process that is particularly critical during neurodevelopment for preventing neural tube defects. DPE mice also had altered retinol and tryptophan metabolism, two processes related to circadian rhythms

Multi-modal changes in MAPK and mTOR cascades

- The largest identified multi-modal gene cluster in our data was for the regulation of apoptotic processes, which directly affects adult neurogenesis, in part through the mTOR pathway.

FUNDING

Funding was provided by the National Institute of Environmental Health Sciences to JB (K99ES027869, R00ES027869); the deArce-Koch Memorial Endowment Fund to JB; and a research gift from the Promedica Foundation to the University of Toledo College of Medicine.

REFERENCES

(1) Silverman, J. L. et al. Reconsidering animal models used to study autism spectrum disorder: Current state and optimizing future. Genes Brain Behav 21, e12803 (2022). https://doi.org:10.1111/gbb.12803
(2) Cogswell, M. E. et al. Health Needs and Use of Services Among Children with Developmental Disabilities - United States, 2014-2018. MMWR Morb Mortal Wkly Rep 71, 453-458 (2022). https://doi.org:10.15585/mmwr.mm7112a3
(3) Richardson, J. R., Taylor, M. M., Shalat, S. L., Guillot, T. S., 3rd, Caudle, W. M., Hossain, M. M., . . . Miller, G. W. Developmental pesticide exposure reproduces features of attention deficit hyperactivity disorder. FASEB J 29, 1960-1972, doi:10.1096/fj.14-260901 (2015). PMC PMC4415012
(4) Shelton, J. F., Geraghty, E. M., Tancredi, D. J., Delwiche, L. D., Schmidt, R. J., Ritz, B., . . . Hertz-Picciotto, I. Neurodevelopmental disorders and prenatal residential proximity to agricultural pesticides: the CHARGE study. Environ Health Perspect 122, 1103-1109, doi:10.1289/ehp.1307044 (2014). PMC PMC4181917
(5) Hicks SD, Wang M, Fry K, Doraiswamy V, Wohlford EM. Neurodevelopmental Delay Diagnosis Rates Are Increased in a Region with Aerial Pesticide Application. Front Pediatr. 2017 May 24;5:116. doi: 10.3389/fped.2017.00116.