Pavlovian Society
Annual Meeting, 2022

September 29 – October 1, 2022
Saint Kate Hotel, Milwaukee, WI

Overview

Thur	6:00–10:00 PM	Opening Reception The Arc Hors d'Oeuvres & Cash Bar
Fri	7:30–8:20	Breakfast
	8:20–12:10	Morning Sessions
	12:10–1:50	Lunch (Exec Committee Meeting)
	1:50–5:45	Afternoon Sessions
	5:45–7:15	Posters & Cash Bar
Sat	7:30–8:25	Breakfast
	8:25–11:50	Morning Sessions
	11:50–2:00	Lunch (WIL Luncheon)
	2:00–6:00	Afternoon Sessions
	6:00–7:30	Posters & Cash Bar
	7:30–9:00	Banquet

Program

Friday (September 30)

7:30–8:20 **Breakfast** (Simone Foyer)
All talks in Simone 2, 3, & 4
[V] Virtual presentation

8:20–8:30 **Karyn Frick** (*U Wisconsin-Milwaukee*)
Welcome

The Society thanks the following for their support: Plexon; Univesity of Wisconsin-Milwaukee Department of Psychology; University of Wisonsin-Milwaukee College of Letters & Science; Medical College of Wisconsin

8:30–9:00 **Natalie Tronson** (*U Michigan*) Past President Lecture: Neuroimmune activation, memory deficits and the risk of cognitive decline in the age of COVID-19

9:00–10:30 **Symposium 1: Honoring the scientific contributions and legacy of Dr. Nadia Chaudhri (Sydney Trask** (*Purdue U*) **& Susan Sangha** (*Indiana U School of Medicine*)**, Chairs)**

* **Shaun Khoo**[V] (*Concordia U*) Pavlovian conditioned approach after extended training: What's influencing sign- and goal-tracking?" (virtual)

* **Marie Monfils** (*U Texas Austin*) Last call for alcohol

* **Milan Valyear** (*McGill U*) Phenotypic differences in what is learned about discrete alcohol cues and contexts

* **Patricia Janak** (*Johns Hopkins U*) Complexities in neural mechanisms for contextual gating of cue-elicited behavior

10:30–10:50 **Coffee Break**

10:50–12:20 **Symposium 2: The effect of stress on appetitive learning and memory (Jaqueline Giovanniello** *(UCLA)***, Chair)**

* **Christian Bravo-Rivera** (*U Puerto Rico School of Medicine*) Neural circuits mediating reward approach and punishment avoidance conflict

* **Shannon Gourley** (*Emory U*) Durable consequences of early-life isolation on goal-seeking behavior

* **Barbara Knowlton** (*UCLA*) Early life stress, habitual responding, and substance use in young adults

* **Jacqueline Giovanniello** (*UCLA*) Opposing amygdala-striatal pathways enable chronic stress to promote habit formation

12:20–1:50 **Lunch (on your own)**
Executive Committee Meeting (Lyrical Boardroom)

1:50–2:20 **Susan Sangha** (*Indiana U School of Medicine*) Women in Learning Lecture: Using learned safety cues to map the behavioral and circuit mechanisms of fear regulation

2:20–2:40 **Christopher Olsen** (*Medical College of Wisconsin*) Prefrontal cortex drug seeking ensembles: Necessity, specificity, and modulation by injury

2:40–3:00 **Jeffrey Lopez-Rojas** (*U Wisconsin-Milwaukee*) Lateral entorhinal cortical input to the hippocampus: hearing the news from a faraway place

3:00–3:20 **Coffee Break**

3:20–4:50 **Symposium 3: Socially connected: How social dynamics and experiences shape behavior and the brain (Moriel Zelikowsky** *(U Utah)***, Chair)**
* **Moriel Zelikowsky** (*U Utah*) Communication and courtship following social isolation
* **Matthew Lovett-Barron** (*UC San Diego*) Neurobiology of collective behavior in schooling fish
* **Ann Clemens** (*U Edinburgh*) Neural circuits of kinship behaviour
* **Regina Sullivan**^{V} (*Nathan Kline Inst & NYU Langone Medical Center*) Infant social trauma, not asocial trauma, targets the amygdala to produce deficits in fear and social behavior

4:50–5:45 **Breakout Discussion Groups (with Snacks)**
Janine Kwapis *(Penn State)* **& Susan Sangha** *(Indiana U School of Medicine)* Funding disparities among women and under-represented minority populations (Method)
Olga Lipatova *(Christopher Newport U)* **& Matthew Campolattaro** *(Christopher Newport U)* Pavlovian research at primarily undergraduate institutions: What, how, and why? (Dada 1)
Nancy Smith *(UCLA)***, Daniel Weatherill** *(UCLA)***, & Andrew Wikenheiser** *(UCLA)* Leveraging machine learning to discover meaningful patterns of animal behavior (Dada 2)
Michael Drew *(U Texas Austin)***, Abha Rajbhandari** *(Icahn School of Medicine, Mt Sinai)***, Andrew Poulos** *(SUNY Albany)***, Kenji Nishimura** *(U Texas at Austin)* Fear sensitization: What is it? How can we study it? What does it tell us about disease? (Deep Image)

5:45–7:15 **Posters and Cash Bar** (Simone 1 & Simone Foyer)

7:15 **Dinner (on your own)**

Saturday (October 1)

7:30–8:25 **Breakfast** (Simone Foyer)

8:25–8:30 **Karyn Frick** *(U Wisconsin-Milwaukee)* Welcome

8:30–10:00 **Symposium 4: The neuropeptide PACAP at the intersection of stress, memory, and behavior (Marieke Gilmartin** *(Marquette U)* **& Abha Rajbhandari** *(Icahn School of Medicine, Mt Sinai)***, Chairs)**
* **Sayamwong (Jom) Hammack** (*U Vermont*) The role of central pituitary adenylate cyclase activating polypeptide (PACAP) signaling in stress and emotion
* **Briana Chen** (*Columbia U*) The VPAC2 receptor mediates fear learning in a sex-specific manner
* **Jessica Barson** (*Drexel U*) Pituitary adenylate cyclase-activating polypeptide (PACAP) in the thalamic paraventricular thalamus and binge-like ethanol drinking
* **Abha Rajbhandari** (*Icahn School of Medicine, Mt Sinai*) Role of PACAP and PAC1 in stress via the brain and body axis

10:00–10:20 **Coffee Break**

10:20–11:50 **Symposium 5: The generalization of learning (Ekaterina Likhtik** *(Hunter College, CUNY)* **& Nesha Burghardt** *(Hunter College, CUNY)***, Chairs)**
* **Jelena Radulovic** (*Albert Einstein College of Medicine*) Retrieval-based generalization of aversive conditioning
* **Larry Zweifel** (*U Washington*) The role of dopamine in threat generalization
* **Maria Geffen** (*U Pennsylvania*) A cortico-thalamic circuit for learning generalization

* **Joseph Dunsmoor** (*U Texas Austin*) Latent associative structures facilitate higher-order transfer of learned fear

11:50–2:00 **Lunch / Women in Learning Luncheon**
Deer Camp MKE, 1023 Old World Third Street (See map on last page)

2:00–2:20 **Polymnia Georgiou**[V] *(U Wisconsin-Milwaukee)* Assessing the role of neuroinflammation in ethanol withdrawal induced memory deficits

2:20–2:40 **Timothy Jarome** *(Virgina Polytechnic Institute & State U)* The role of sex in the necessity for diverse polyubiquitination modifications in memory formation

2:40–4:10 **Symposium 6: Neuroimmune interactions with stress, learning, and memory (Nicole Ferrara** *(Rosalind Franklin U)***, Chair)**

* **Ruth Barrientos** (*The Ohio State U*) High-fat diet impairs long-term memory and synaptic plasticity in aged rats via neuroinflammatory mechanisms

* **Erica Glasper** (*The Ohio State U*) Early-life rearing influences sex differences in social learning and neuroimmune function

* **Amiel Rosenkranz** (*Chicago Medical School / RFUMS*) Effects of mild inflammation on amygdala and social function

* **Eric Wohleb** (*U Cincinnati*) Microglial P2RY12 mediates chronic stress-induced synaptic deficits and behavioral consequences

4:10–4:30 **Coffee Break**

4:30–6:00 **Symposium 7: Transitions from goals to habits: Identifying the what, when and how (Sharlen Moore** *(Johns Hopkins U)***, Chair)**

* **Talia Lerner** (*Northwestern U*) Dopamine circuits for habit formation

* **Rachel Smith** (*Texas A&M U*) Investigating the link between habits and punishment-resistant cocaine seeking

* **Neil Garrett**[V] (*U East Anglia* & Princeton U) Model based habits

* **Sharlen Moore** (*Johns Hopkins U*) Sudden transitions from goal-directed to habitual behavior during sensorimotor learning in mice

6:00–7:30 **Posters and Cash Bar** (Simone 1 & Foyer)

7:30–9:00 **Banquet, Awards, & Closing**
Simone 2, 3, & 4

Posters

In Simone 1 & Simone Foyer. Generally alphabetical by first author's last name, with A–K on Friday and L–Z on Saturday, except in the case of special requests by authors.

Friday

1. **Balderston, NL** (*U PENN*) Using contextual threat to investigate the interactions between anxiety and cognition

2. **Balsam PD, Mallea J** (*Barnard College & Columbia U*) The effect of rate and probability of reward on the appearance and strength of Sign Tracking Behavior

3. **Beamish SB, Gross KS, Anderson MA, Helmstetter FJ, Frick KM** (*U Wisconsin-Milwaukee*) Sex differences in training-induced protein degradation in the dorsal hippocampus of male and female mice

4. **Blair RS, Cain EC, Ports E, Nagaya N** (*Texas A&M U*) The effects of intra-BNST androgen metabolite on Pavlovian fear conditioning

5. **Bonanno GR, Met Hoxha E, Ferrara NC, Trask S** (*Purdue U*) Context fear memory can be reduced through memory updating with a weak shock

6. **Bonsib AG, Taibl EG, Totis JE, Petersen AR, Kochli DE.** (*Washington College*) Developmental and adult isolation housing reduces goal-directed behavior but promotes cue-directed behavior

7. **Broomer MC, Bouton ME** (*U Vermont*) Renewal, spontaneous recovery, and reacquisition after punishment and extinction

8. **Carroll E, Love J, Conoscenti M, Covington, Zelikowsky M** (*U of Utah School of Medicine*) The lateral preoptic area Controls the effects of social isolation on mouse courtship behavior and song

9. **Denholtz LE**[1,2]**, Liu J**[3]**, Nahmoud I**[4]**, Casaccia P**[3]**, Likhtik E**[1,2] ([1]*CUNY;* [2]*Hunter College, CUNY;* [3]*CUNY;* [4]*Wayne State U School of Medicine*) Safety learning mediated changes in myelination at the medial prefrontal cortex

10. **Dhillon PS, Halcomb CJ, Philipp TR, Cox JH, Vanderhoof SO, Jasnow AM** (*U South Carolina School of Medicine*) Sex differences in the acquisition and extinction of platform mediated avoidance: Role of glucocorticoid receptors

11. **DiFazio LE, Jia R, Greer Z, Sharpe MJ** (*UCLA*) The temporal dynamics of encoding fear memories in the BLA, and how they change following experience with rewards

12. **Hoang IB, Munier JJ, Greer Z, Millard SJ, DiFazio LE, Wassum KM, Izquierdo A, Sharpe MJ** (*UCLA*) Methamphetamine and midbrain-hypothalamic control of cue-guided behavior

13. **Ferrara NC, Trask S, Ritger AC, Padival M, Rosenkranz JA** (*Rosalind Franklin U, Purdue U*) Brief isolation alters maturing cortical-amygdala circuits supporting social preference in adults and adolescents

14. **Fleischer AW**[1], **Ramirez S**[2], **Frick KM**[1] ([1]*U of Wisconsin-Milwaukee;* [2]*Boston U*) Tet-tagging as a method to test the estrogenic Two-Step Wiring Hypothesis

15. **Franco CY, Knowlton BJ** (*UCLA*) Childhood racial discrimination effects on habitual responding and disordered eating

16. **Fukunaga Y, Gonzalez SE, Lock E, Schulman E, Cazares VA** (*Williams College*) Exposure of conditioned stimuli in multiple contexts enhances fear extinction memories

17. **Garcia AK, Kendall RK, Wikenheiser AM** (*UCLA*) Methamphetamine reduces the duration of foraging bouts in rats performing a naturalistic decision-making task

18. **Guerra DP, Moscarello JM** (*Texas A&M U*) The bed nucleus of the stria terminalis mediates the expression of avoidant behavior in male rats

19. **Alwood MR, Moscarello JM** (*Texas A&M U*) Chemogenetic inactivation of the dorsal hippocampus facilitates avoidance learning in female but not male rats

20. **Halcomb CJ, Vanderhoof SO, Mott DD, Jasnow AM** (*U South Carolina School of Medicine*) Basolateral amygdala inputs to the anterior cingulate cortex regulate fear learning that promotes generalized fear

21. **Hart E, Gardner MP, Schoenbaum G** (*NIDA IRP*) Anterior cingulate neurons signal neutral cue pairings during sensory preconditioning

22. **Alcalá JA**[1], **Prados J**[2], **Urcelay GP**[3] ([1]*Complutense U Madrid;* [2]*U Derby;* [3]*U Nottingham*) Dimensional training attenuates overshadowing

23. **Herrera E**[1], **Austen JM**[2], **Urcelay GP**[2] ([1]*Bournemouth U;* [2]*U Nottingham*) The effects of goal–landmark distance on overshadowing: a replication in humans of Goodyear & Kamil (2004)

24. **Urcelay GP**[1], **Hulley T**[1], **Alcalá JA**[2] ([1]*U Nottingham;* [2]*Complutense U Madrid*) Uncertainty increases generalization of human predictive learning

25. **Hodebourg R**[1], **Meyerink ME**[2], **Crow AD**[1], **Reichel CM**[1], **Kalivas PW**[1], **Garcia-Keller C**[2] ([1]*Medical U South Carolina;* [2]*Medical College of Wisconsin*) Cannabinoid use is enhanced by stress and changes conditioned stress responses

26. **Hu S, George A, Packard K, Song M, Kissi J, Nguyen-Lopez O, Su A, Tesone E, Opendak M** (*Kennedy Krieger Institute & Johns Hopkins School of Medicine*) Functional ontogeny and social control over the developing lateral habenula

27. **Jaiyesimi A, Lohr C, Banerjee A, Waddell J** (*U Maryland School of Medicine*) A single dose of psilocybin facilitates extinction of fear in male rats

28. **Jin B, DeNardo L** (*UCLA*) Brain-wide mapping of amnesic and persistent fear memory circuits throughout development

29. **Kaneko R, Mallea J, Balsam PD** (*Columbia U*) Impact of temporal uncertainty about when a CS is presented on sign tracking behavior

30. **Kaplan K, Hunsberger H** (*Rosalind Franklin U*) Benzodiazepines impair contextual fear memory

31. **Keiser AA, Dong T, Kramár EA, Butler C, Matheos DP, Tong L, Berchtold NC, Chen S, Samad M, Magnan C, Beardwood J, Shanur S, Baldi, P, Cotman CW, Wood MA** (*UC Irvine*) Exercise parameters that open a 'molecular memory window' for cognitive enhancement shine light on key memory mechanism in the adult, aging, and Alzheimer's Disease brain

32. **Kurtoglu B**[1], **Estes M**[1], **Laskowski L**[1], **Windsor BW**[1], **Davidson R**[1], **Van Newenhizen E**[1], **Okunseri T**[2], **Mantych M**[3], **Spring M**[4], **Wheeler DS**[3], **Wheeler RA**[3], **Hearing MC**[3], **Mantsch JR**[1] ([1]*Medical College of Wisconsin;* [2]*Yale U;* [3]*Marquette U;* [4]*Dartmouth College*) Effects of chronic stress and corticosterone on Pavlovian conditioned approach behavior and prefrontal cortical function in rats

33. **Kutlu MG, Tat J, Zachry J, Calipari ES** (*Vanderbilt U*) Accumbal dopamine response to expected aversive outcomes mediates the expression of conditioned behavior

34. **Lamb JH, Furtak SC** (*CSU Sacramento*) Chemogenetic silencing of the perirhinal cortex attenuates fear extinction learning to a discontinuous visual conditioned stimulus

35. **Laughlin LC, Samels SB, Moloney DM, Andrade E, Sears RM, Cain CK** (*NYU; NKI*) Counterconditioning of response-produced safety signals is highly context-dependent in female rats

36. **Le QE, Hereford D, Borkar CD, Fadok JP** (*Tulane U*) Dynamics of defensive behavior using Pavlovian fear conditioning with a serial compound stimulus

37. **Lee J, Hanif S, Aubry A, Burghardt NS** (*CUNY*) The contribution of associative learning to stress-induced social avoidance behavior

38. **López-Moraga A, Luyten L, Beckers T** (*KU Leuven*) Sex differences in generalization of an auditory fear memory

39. **Mehrzad Z, Cherukupalli C, Prasad S, Campese VD** (*U Evansville*) Sexually dimorphic effects of reinforcement schedules on active avoidance

40. **Tashjian SM, Mobbs D** (*Caltech*) Adaptive safety coding

41. **Wilson WJ** (*Albion College, Frontière Astrophotography*) Learning continues post-retirement: Trying hard to make this relevant to the Society

Saturday

1. **Machado GDB, Fleischer AW, Schnitzler AL, Frick, KM** (*U Wisconsin-Milwaukee*) G-coupled protein estrogen receptor (GPER) agonism in the dorsal hippocampus enhances memory consolidation in gonadectomized male mice

2. **Magalhaes G, Burnell H, Subedi S, Jabri S, Ganiyu O, Meyer H** (*Boston U*) Sex differences in safety learning are present in adults but not adolescents

3. **McKinney A, Davis I, Pickens C** (*Kansas State U*) Adolescent exposure to amphetamine did not impair later devaluation in adulthood in tasks that do or do not allow for compensation between strategies

4. **Hougham AL, Kim J, Pickens CL, Wang C** (*Kansas State U*) Short term withdrawal from repeated fentanyl injections leads to impairment in the expression of goal-directed action in a Pavlovian goal-tracking task

5. **Weaver C, Pajser A, Fisher H, Pickens C** (*Kansas State U*) Repeated exposure to amphetamine does not lead to long-term alterations in omission contingency learning

6. **Mondello JE, Chang CW, Trott JM, Anaya A, Solorio S, Tran L, Fanselow MS** (*UCLA*) Stress enhanced fear learning enhances excitatory synaptic transmission in basolateral amygdala neurons

7. **Moore E, Harris H, Slover W, Lipatova O, Campolattaro M** (*Christopher Newport U*) Generalization of associative responding between tone-off auditory cues

8. **Mueller DM, Giglio EM, Chen C, Grissom NM** (*U Minnesota*) Explore-exploit state governs the spatial configuration of touch actions in a mouse bandit decision making task

9. **Olvera ME, Roberto RU, Cervantes MC, Monfils M-H, Gonzales RA, Lee HJ** (*U Texas Austin; U Missouri*) Conditioned responses to cues associated With alcohol availability in Rats with a history of ethanol dependence

10. **Pahua AE, King C, Davison T, Mali I, Payne B, Plakke B** (*Kansas State U*) Impact of exercise on cognitive performance in a rodent model of autism

11. **Peterson S, Chavira J, Maheras A, Garcia-Arango A, Seamans E, Keiflin R** (*UC Santa Barbara*) Role of the orbitofrontal cortex and dorsal hippocampus in the expression and inferred generalization of contextual rules for reward prediction

12. **Pierce-Messick ZJ, Corbit LH** (*U Toronto*) The influence of extinguishing the context-response operandum pairing on goal-directed behaviour

13. **Plakke B, King C, Davison T, Maze T, Mali I, Payne M, Bossmann S** (*Kansas State U*) Enlarged frontal cortices during adolescence impacts cognitive performance in female autism spectrum modeled rats

14. **Raskin MR, Malone CA, Hilz EN, Shumake J, Lee HJ, Monfils MH** (*U Texas Austin*) CO2 reactivity predicts spontaneous recovery of conditioned food seeking in rats

15. **Rojas GR, Grissom NM** (*U Minnesota*) Choice inflexibility is observed in male 16p11.2 hemideletion mice during acquisition of delay but not probability discounting

16. **Sarka BC, Liu S, Liu QS, Stucky CL, Olsen, CM** (*Medical College of Wisconsin*) Measuring the effect

of neuropathic pain on drug-sSeeking ensembles in the dmPFC

17. **Schwabe MR, Fleischer AW, Kuehn RK, Beaty HA, Milkie EM, Schnitzler AL, Chaudhury S, Donaldson WA, Sem DS, York JM, LaDu MJ, Frick KM** (*U Wisconsin-Milwaukee; Marquette U; Concordia U; U Wisconsin; U Illinois Chicago*) Effects of a novel estrogen receptor beta agonist and APOE genotype on synaptic markers of memory in a mouse model of Alzheimer's disease

18. **Seese S, Tinsley CE, Hixon JG, Monfils MH** (*U Texas Austin; OHSU*) Conspecific interactions predict social transmission of fear in female rats

19. **Shipman ML, Chen SE-S, Desilets GL, Corbit LH** (*U Toronto*) Microglial activation in the DMS impairs goal-directed control

20. **Liu S, Olsen C** (*Medical College of Wisconsin*) Investigation of the necessity and specificity of the dmPFC cocaine seeking ensemble

21. **Smies CW, Bellfy L, Wright DS, Bennetts SS, Urban MW, Brunswick CA, Kwapis JL** (*Penn State*) Epigenetic mechanisms supporting competition in reconsolidation-based memory updating

22. **Bellfy L, Smies CW, Bodinayake KK, Bernhardt AR, Stuart EM, Wright DS, Lo C-Y, Boyd HM, Kwapis JL** (*Penn State*) Hippocampal Per1 may contribute to time-of-day effects on memory consolidation

23. **Brunswick CA, Baldwin DJ, Bodinayake KK, Bellfy L*, McKenna AR, Stuart EM, Murakami S, Smies CW*, Kwapis JL** (*Penn State; *Inimitable, according to Brunswick*) Age-related deficits in spatial memory due to epigenetic dysregulation of Per1 within the retrosplenial cortex but not the suprachiasmatic nucleus

24. **Sood A, Richard JM** (*U Minnesota*) Investigating ventral pallidal encoding of expected outcome value

25. **Staffeld J** (*Eastern Michigan U; U Michigan*) Pavlovian Conditioned Avoidance and its Correlation to Pavlovian Conditioned Approach

26. **Stidham N, Russo-Savage L, Giddings E, Brabec JL, Lara M, Laprade KA, Bonney EA, Stafford, JM** (*U Vermont*) A method for producing high, volitional opioid use during gestation reveals developmental outcomes in offspring that can be mitigated by pharmacological intervention

27. **Su CJ, Fukunaga Y, Cazares VA** (*Williams College*) No effects of partial reinforcement on fear extinction learning in mice

28. **Swarovski MS, Lemmon D, Romero N, Conoscenti M, Brigidi S, Zelikowsky M** (*U Utah*) CA1 molecular signatures underlying context fear and fear renewal after extinction

29. **Conoscenti MA, Sattler KP, Kennedy A, Zelikowsky M** (*U Utah; Northwestern U*) Social isolation and footshock stress produce aggression that is behaviorally and biologically distinct

30. **Sattler K, Miller R, Zelikowsky M** (*U Utah*) The Role of the ventral hippocampus in trauma-induced aggression and enhanced fear

31. **Gagon J, Gatlin RE, Zelikowsky M** (*U Utah*) Control of isolation-induced aggression through activation of medial prefrontal cortex Ppramidal neurons

32. **Gatlin, RE, Gagon J, Winebrenner C, Zelikowsky M** (*U Utah*) The Role of the mPFC/LS Tachykinin 2 system in isolation induced aggression

33. **Grammer AJ, Zelikowsky M** (*U Utah*) The BNST to NAc: a neural circuit for isolation induced social anxiety

34. **Taylor DL, Zelikowsky M** (*U Utah*) The effect of social instability on behavior and the brain

35. **Thomas CMP, Bouton ME, Green JT** (*U Vermont*) Inactivation of the prelimbic cortex eliminates ABA renewal in a stress acquisition context

36. **Vasudevan K, Ramanathan KR, Vierkant V, Maren S** (*Texas A&M U*) Nucleus reuniens inactivation does not impair consolidation or reconsolidation of fear extinction

37. **Vercammen L, Beckers T, Luyten L, Vervliet B** (*KU Leuven*) The rewarding properties of safety signals as established by a two-way active avoidance task in rats

38. **Wachter SL, Parker KL, Freeman JH** (*U Iowa*) Disconnection of cerebellar communication with the prefrontal cortex causes deficits in executive function in rats

39. **Wang Z, Moore S, Sun R, Lee A, Zhu Z, Charles A, Kuchibhotla K** (*Johns Hopkins U*) Slow or sudden: revealing naturalistic transitions to habitual behavior during learning

Abstracts

In alphabetical order by first author's last name.

Alcalá JA[1], Prados J[2], Urcelay GP[3] ([1]*Faculty of Psychology, Complutense U of Madrid, Spain;* [2]*School of Psychology, U of Derby, UK;* [3]*School of Psychology, U of Nottingham, UK.*) Dimensional training attenuates overshadowing ABSTRACT: Prior experience with elemental or configural discriminations shapes how agents learn subsequent information: prior training that encourages elemental processing promotes competition between events, while prior configural training tends to attenuate competition, and sometimes results in facilitation. We sought to assess whether configural and elemental pretraining modulated subsequent overshadowing when using two separable dimensions (i.e., colors and symbols) in a predictive learning task. Furthermore, we used post-test questionnaires asking whether participants used either only one or both dimensions during training and assessed if this influenced the magnitude of overshadowing. Across 3 experiments, prior experience with a configural discrimination (i.e., biconditional discrimination) did not influence the magnitude of overshadowing compared to control groups. However, attended dimensions were less prone to overshadowing (Experiments 1 and 2), and Experiment 3 revealed that prior elemental training (making one dimension relevant) attenuated subsequent overshadowing of that dimension. Hence, preferred or trained dimensions influenced the size of overshadowing. These results are discussed in light of attentional and configural theories of associative learning. SUPPORT: UK ESRC Grant (ES/R011494/2) awarded to GPU and JP

Alwood MR, Moscarello JM (*Texas A&M U*) Chemogenetic inactivation of the dorsal hippocampus facilitates avoidance learning in female but not male rats ABSTRACT: Compulsive avoidance behavior is a hallmark symptom of post-traumatic stress disorder (PTSD; American Psychiatric Association, 2013). This avoidance of traumatic reminders can interfere with extinction-based therapies, which are the gold-standard treatment for PTSD. For this reason, understanding the neural mechanisms which underlie avoidance behavior is of clear clinical concern. In a preliminary experiment our group sought to examine the role of dorsal hippocampus in the acquisition and retention of avoidance behavior. Using the excitatory hM3Dq DREADD, we inhibited the dorsal hippocampus of male and female rats by activating dlx+ GABAergic interneurons (Krueger et al., 2020). Rats then underwent 10 days of unsignaled (Sidman) active avoidance training, in which shuttling in response to regularly delivered shocks (escape) leads to a safety period that can be extended if the subject shuttles again before the safety period ends (avoidance). IP injections of CNO were administered prior to days 2 and 8 of training to assess the effects of dHPC inactivation on the acquisition and retention of avoidance learning. Typically, animals begin with high numbers of escape responses that decrease as the animals learn to avoid. Although no differences emerged on the day of injection, female dlx-hM3Dq rats demonstrated elevated avoidance responding on the days following both injections when compared with GFP controls. In contrast, no differences were observed between dlx-hM3Dq and GFP groups in male rats. This suggests that dHPC opposes avoidance in females specifically. Given that the previous literature demonstrates a role for dHPC in contextual fear memory in male rodents, it is unlikely that disruption of context-shock associations memory is responsible for the effects we report here. Instead, our results may indicate that hippocampal memory biases females toward reactive responses (escape) and away from proactive responses (avoidance) in female rats, though further work is needed to confirm this hypothesis.

Balderston, NL (*U PENN*) Using contextual threat to investigate the interactions between anxiety and cognition ABSTRACT: Context conditioning has been extensively used as a model system to understand how declarative memories are formed. A variant of this procedure, threat of unpredictable shock (i.e. instructed context conditioning), has also been used to model the hyperarousal symptoms experienced by individuals suffering from PTSD and other anxiety disorders. In a typical experiment, subjects are instructed that there are safe contexts where they will not receive shock and threat contexts where they can receive shocks at any time. Because very few shocks are needed to sustain the perception of the instructed threat, this robust within-subjects comparison can be paired with various cognitive and behavioral tasks. In my research, I use threat of unpredictable shock as a passive contextual manipulation, and then study the effect of this manipulation on behavioral, cognitive, and neural outcomes. I will present data from several lines of research using psychophysiology, cognitive testing, neuroimaging, and neuromodulation. I will highlight the following basic findings: 1) unpredictable shock threat reliably increases arousal, which can be measured through potentiation of the acoustic startle reflex, 2) unpredictable threat increases the excitability and connectivity of the parietal cortex, potentially leading an increase in distractor susceptibility, 3) targeting connectivity hubs within the parietal cortex with inhibitory transcranial magnetic stimulation reduces acoustic startle potentiation. Together, these results suggest that connectivity hubs in the parietal cortex may be key for the hyperarousal experienced by anxiety patients, and targeting these hubs may be a potential avenue for future treatments for anxiety disorders. SUPPORT: This work is supported in part by a NARSAD Young Investigator Grant from the Brain & Behavior Research Foundation (NLB: 2021), and a career development grant from the National Institutes of Health: K01MH121777.

Balsam PD, Mallea J (*Barnard College and Columbia U*) The effect of rate and probability of reward on the appear-

ance and strength of Sign Tracking Behavior ABSTRACT: When distinctive stimuli are used as cues predictive of reward, we often observe two distinctive behavioral phenotypes in animals' anticipatory behavior: subjects will orient, approach, and interact with the cue (i.e., sign-tracking) or the location where the reward is expected (i.e., goal-tracking). Previous studies have shown higher levels of sign-tracking under uncertain conditions, like probabilistic reinforcement. However, changes in probability are often confounded with changes in reinforcement rate. In two experiments in mice, we analyzed the effect of probabilistic or continuous reinforcement when it's either accompanied or not by differences in reinforcement rate (Experiment 1), and over different CS durations (Experiment 2). The response rate to both the cue and reward port, the temporal distribution of responses during a trial, as well as the point in which Sign Tracking behavior becomes predominant were analyzed. Results showed higher levels of sign-tracking behavior under probabilistic reinforcement only when probability and rate were confounded. Similarly, animals reinforced at lower rate started allocating the majority of their responses to the cue earlier in training. Despite this, the temporal organization of both behavioral phenotypes and the overall rate of responding was similar in all groups. Together, these experiments showed that reward rate is a better predictor of the appearance of sign tracking behavior than probability, and that sign tracking may be mediated by similar underlying processes than other forms of anticipatory behavior. SUPPORT: This study was supported by National Institute of Mental Health Grant R01MH068073 to Peter Balsam. Jorge Mallea was funded by Fulbright and Conicyt (Fulbright-Conicyt Bio Scholarship #56160012).

Barrientos, RM, Butler, MJ, Gonzalez-Olmo, BM, Muscat, SM, Mackey-Alfonso, S (*Institute for Behavioral Medicine Research, Department of Psychiatry and Behavioral Health, Department of Neuroscience, Chronic Brain Injury Program, The Ohio State U, Columbus, OH, USA*) High-fat diet impairs long-term memory and synaptic plasticity in aged rats via neuroinflammatory mechanisms. ABSTRACT: Sensitized microglia are a hallmark of the aging brain, and are key contributors to exaggerated neuroinflammatory responses to peripheral insults. My talk will focus on high-fat diet (HFD) as a peripheral insult with robust effects on brain health in aged rats. Short-term consumption of a HFD rich in saturated fats is sufficient to produce exaggerated neuroinflammation in the hippocampus and amygdala of the aged, but not young adult rat brain, and this is accompanied by profound impairments in hippocampal- and amygdalar-dependent long-term memory. Blocking the receptor for the proinflammatory cytokine IL1 beta centrally, prevents these memory impairments. We now have data indicating that synaptic plasticity (LTP) is potently degraded in aged HFD-fed rats and that this too can be prevented with an IL1 receptor antagonist suggesting that neuroinflammation is a key mediator of these cognitive and synaptic impairments. Our recent data suggest that palmitic acid, a prominent proinflammatory fatty acid component of HFD, may be a critical signaling molecule to microglia. My talk will include new data and a discussion on the role of the classical complement cascade leading to indiscriminate synaptic engulfment by palmitic acid-activated microglia. SUPPORT: NIH grants AG028271 & AG067061

Barson JR (*Drexel U College of Medicine, Department of Neurobiology and Anatomy, Philadelphia, PA 19129 USA*) Pituitary adenylate cyclase-activating polypeptide (PACAP) in the thalamic paraventricular thalamus and binge-like ethanol drinking ABSTRACT: The paraventricular nucleus of the thalamus (PVT) is a major node of the limbic system and expresses a number of neuropeptides through which it may make its contributions to motivated and affective behavior. We have found in both rats and mice that one neuropeptide that is densely expressed in the PVT is pituitary adenylate cyclase-activating polypeptide (PACAP). While PACAP is associated with both motivated and stress-related behaviors, most studies focus on the PACAP-38 isoform, which comprises the major portion of PACAP in the brain. Notably, our results show that it is PACAP-27 that is present in a high percentage of cells in the PVT, with females having more PACAP in the PVT than males. Moreover, levels of PACAP-27 in individual cells of the PVT are elevated at the end of an ethanol binge. To determine the effect of this rise in PACAP, we injected an AAV into the PVT of rats to increase local expression of PACAP and, in addition to finding that this significantly and selectively elevated levels of PACAP-27 in the PVT, we observed that it significantly suppressed binge-like ethanol drinking, with few accompanying changes in stress-related behaviors in the light-dark box, novelty-suppressed feeding test, and forced swim text. Next, we found that the PVT sends major PACAP-27+ projections to the nucleus accumbens shell, among other limbic regions. In male and female rats, we found with microinjections into the nucleus accumbens shell that PACAP-27 but not PACAP-38 dose-dependently reduced binge-like ethanol but not sucrose drinking. Moreover, while injection of PACAP-38 into the shell reduced exploratory behavior in a light-dark box, we found no such effects from similar injections of PACAP-27. All together, these results suggest that PACAP-27 acts as an endogenous negative feedback signal in the PVT to curb binge-like ethanol drinking, and that it has minimal effects on stress-related behavior. SUPPORT: This research was supported by NIH Grants R01AA028218 and R00AA021782.

Beamish SB, Gross KS, Anderson MA, Helmstetter FJ, Frick KM (*U of Wisconsin-Milwaukee*) Sex differences in training-induced protein degradation in the dorsal hippocampus of male and female mice ABSTRACT: The ubiquitin proteasome system (UPS) targets substrate proteins to the

26S proteasome for degradation by tagging them with ubiquitin. UPS activity in the dorsal hippocampus (DH) is necessary for multiple types of memory, including object memory, in male rodents. However, sex differences in DH UPS activation after fear conditioning suggest that other forms of learning may also differentially regulate DH UPS activity in males and females. Here, we examined markers of UPS activity in the synaptic and cytoplasmic fractions of dorsal hippocampus (DH) tissue collected 1 h following object training in adult male and female mice. Mice were first handled for 30 s/d for 3 d. Mice were then habituated in an empty testing arena for 5 min/d for 2 d. During training, mice accumulated 30 s of exploration with two identical objects and DH tissue was collected 1 h after completion of training. In males, training increased phosphorylation of proteasomal subunit Rpt6, 20S proteasome activity, and amount of postsynaptic protein PSD-95 in the DH synaptic fraction. In females, training did not affect measures of UPS or synaptic activity the DH synaptic fraction, but instead increased Rpt6 phosphorylation in the DH cytoplasmic fraction. Levels of K48 polyubiquitination were not affected by training in either sex, nor were levels of phosphorylated CaMKII and PKA, both of which regulate proteasome activity. Overall, training-induced UPS activity was greater in males than females and greater in synaptic fractions than in cytosol. These data suggest that object training drives sex-specific alterations in UPS activity across subcellular compartments in the DH. SUPPORT: This work was supported by R01MH107886 awarded to K.M.F. and F31MH118782 to K.S.G.

Bellfy L, Smies CW, Bodinayake KK, Bernhardt AR, Stuart EM, Wright DS, Lo C-Y, Boyd HM, Kwapis JL (*Pennsylvania State U*) Hippocampal Per1 may contribute to time-of-day effects on memory consolidation ABSTRACT: Many biological processes are influenced by the circadian system, including memory performance. Behavioral paradigms, such as the dorsal hippocampus (DH)-dependent paradigm Object Location Memory (OLM), typically show that memory is better during the day compared to the night. In this paradigm, mice learn the location of two identical objects during a training session. During the test session 24 hours later, one of the objects is moved to a novel location. Memory is measured as the time the mouse spends investigating the object in the novel location compared to that in the familiar location. We used OLM to answer the outstanding question of what is driving this time-of-day effect on memory performance. We hypothesized that circadian rhythm genes may regulate the consolidation process across the day/night cycle contributing to the time-of-day effect. To test our hypothesis, we first tested long-term memory performance across the day/night cycle and found better memory performance during the day than the night. Next, we tested short-term memory and found that mice were able to acquire the memory at similar levels during the day and the night, suggesting that nighttime acquisition is intact despite the reduced long-term memory. We then assessed memory retrieval across the day/night cycle. Specifically, we trained mice at the peak and trough of memory, ZT5 and ZT17 respectively, but tested them 36 hours later, so mice that were trained during the day were tested at night and vice versa. We found that the time of memory acquisition, rather than the time of retrieval, was the driving factor determining whether memory was intact; mice that were trained during the day were able to retrieve the memory at night whereas the night-trained mice showed poor memory retrieval when tested during either the day or nighttime. Together, these results demonstrate that nighttime memory deficits are likely due to impaired consolidation. As consolidation is known to be transcription-dependent, we performed RNA-sequencing to identify learning-induced gene changes over the day/night cycle. Notably, circadian rhythm genes were commonly upregulated in response to learning during the day but not the night, including the circadian gene, Period1 (Per1). When hippocampal Per1 expression was assessed, we found that it oscillated in tandem with memory performance, consistent with a potential role in regulating memory across the day/night cycle. In conclusion, memory consolidation oscillates across the 24h day and may be regulated in part by hippocampal Per1 expression.

Blair RS, Cain EC, Ports E, Nagaya N (*Psychological & Brain Sci., Inst. For Neuroscience., Texas A&M Univ.*) The effects of intra-BNST androgen metabolite on Pavlovian fear conditioning ABSTRACT: Sex steroid hormones and their neuroactive metabolites can modulate fear and anxiety behavior in humans and rodents. We have previously shown that the progesterone metabolite allopregnanolone (ALLO) can influence the expression contextual fear when infused into the bed nucleus of the stria terminalis (BNST) of male rats. Similarly, the testosterone metabolite, 3a-androstanediol (3a-diol), has been implicated in fear and anxiety. Here, we explore the role of intra-BNST infusion of 3a-diol on the acquisition and expression of Pavlovian fear conditioning in adult male rats. We infused 3a-diol (7.3 ug/uL) or vehicle (VEH; 30% 2-hydroxypropyl-B-cyclodextrin) into the BNST 10 min prior to a conditioning session consisting of 5 tone (CS; 2 kHz, 10 s, 80 dB)-footshock (2 s, 1 mA) pairings. On subsequent days, fear retention was tested by separate exposures to context (10 min) and cue (4 CS-alone trials in a novel context) after infusion of either the same or different drug (3a-diol or VEH). Acquisition of conditioned fear was facilitated by pre-training infusions of 3a-diol; however, expression of contextual and cued fear was similar regardless of drug infusion. These effects were not state-dependent. It is possible that endogenous neurosteroids competed with the ability of exogenous 3a-diol to confer state-dependence. Therefore, we are currently exploring whether reducing endogenous neurosteroids within the BNST with finasteride, a

neurosteroid synthesis inhibitor, will reveal 3a-diol effects on performance. Preliminary data suggest that under these conditions, intra-BNST 3a-diol reduces the expression of contextual freezing, similar to ALLO. Work in progress will determine if the anxiolytic effect of 3a-diol in finasteride-treated rats is similarly state-dependent. SUPPORT: College of Liberal Arts, Texas A&M U

Bollinger JL, Dadosky, DT, Flurer JK, Rainer IL, Woodburn SC, Wohleb ES (*U of Cincinnati*) Microglial P2RY12 mediates chronic stress-induced synaptic deficits and behavioral consequences ABSTRACT: Chronic unpredictable stress (CUS) drives microglia-mediated neuronal remodeling and synapse loss in the prefrontal cortex (PFC), contributing to deficits in cognition and behavior. However, it remains unclear what mechanisms guide microglia-neuron interaction in stress. Evidence indicates that neuronal activity-dependent purinergic signaling directs microglial processes and synaptic engagement via P2Y12, a purinergic receptor exclusively expressed by microglia in the brain. Stress alters excitatory neurotransmission in the PFC, thus we aimed to determine if P2Y12 signaling promotes functional changes in microglia in chronic stress. Here we used genetic ablation of P2Y12 (P2ry12-/-) or pharmacological blockade (clopidogrel, ticagrelor) to examine the role of purinergic signaling in stress-induced microglia-neuron interaction. Multiple behavioral, physiological, and cytometric endpoints were analyzed. Deletion of P2Y12 led to a number of fundamental alterations in the PFC, including heightened microglial number and increased dendritic spine density. Blocking P2Y12 shifted surface levels of microglial CX3CR1, CSF1R, and CD11b, suggesting changes in synaptic engagement and phagocytosis in the PFC. In line with this, pharmacological blockade of P2Y12 prevented CUS-induced increases in the proportion of microglia with neuronal inclusions, limited dendritic spine loss in the PFC, and prevented alterations in stress coping behavior and working memory function. Overall, these findings indicate that microglial P2Y12 is a critical mediator of stress-induced synapse loss in the PFC and subsequent behavioral deficits. SUPPORT: This work was supported by the National Institute of Mental Health (F32MH123051, JLB; R01MH123545, ESW) and the U of Cincinnati.

Bonanno GR, Met Hoxha E, Ferrara NC, Trask S (*Purdue U*) Context fear memory can be reduced through memory updating with a weak shock ABSTRACT: Recent work has demonstrated that fear memories established by pairing a context with a footshock can be reliably reduced by subsequent pairings of that context with a weaker shock. This procedure shares similarities with extinction learning in that both involve extended time in the conditioning chamber following training and reduce freezing to that context. However, rather than relying on the creation of new inhibitory memory acquired in extinction, this weak shock exposure is hypothesized to engage reconsolidation-like processes that weaken the original memory. The present experiments were therefore designed to directly compare the 10 weak-shock procedure to extinction. Male and female Long Evans rats were first fear conditioned with 5 presentations of a strong (1.0 mA) footshock. The next day, animals were placed back in the acquisition context and divided into three groups: weak shock, extinction, or no exposure. The weak shock group received 10 0.3 mA footshocks, the extinction group received an equivalent amount of context exposure but no footshocks (i.e., extinction), and the no exposure group remained in their homecage. On the final day, all animals were placed back into the conditioning chamber and behavior was assessed for 10 minutes without any footshocks. We first demonstrated that both weak shock exposure and extinction result in decreased freezing to the context relative to the no exposure group. We next found that conditioning with the weak shock is not enough to form a context-shock association on its own, suggesting that the 10 weak-shock procedure does not create a new memory. We then demonstrated that weak shock exposure in a new context can still reduce freezing elicited by the training context, suggesting that it reduces responding through a different process than extinction which does not transcend context. Together, these results suggest that weak shock procedure does not rely entirely on the creation of a new inhibitory memory like that created in extinction and instead might alter the original representation of the shock to reduce fear responding.

Bonsib AG, Taibl EG, Totis JE, Petersen AR, Kochli DE (*Washington College*) Developmental and adult isolation housing reduces goal-directed behavior but promotes cue-directed behavior ABSTRACT: Pavlovian Lever Autoshaping (PLA) is a common method for assessing individual differences in addiction vulnerability. In this procedure, brief lever insertion predicts non-contingent delivery of a food pellet. Sign-trackers preferentially interact with the lever, while goal-trackers preferentially interact with the foodcup. Sign-trackers display a variety of "addiction-vulnerable" behaviors such as poor behavioral flexibility, while goal-trackers are sensitive to outcome value. To promote motivation during PLA, rats commonly undergo mild food restriction that requires isolation housing—stressful living conditions for social species. The present work examines the contribution of housing conditions to PLA, a Morris Water Maze (MWM) dual solution task, and a "drinking in the dark" ethanol self-administration and quinine adulteration task. Male and female rats are assigned to one of three housing conditions at weaning: Enriched (four rats to a large cage with enrichment objects), Single (singly-housed shoebox cage with no enrichment), and Raised Enriched (raised under Enriched conditions, but transitioned to Single conditions at eight weeks). The Raised Enriched condition models a "typical" experiment in which rats are raised in groups but

isolated seven days prior to the start of experimentation. We found that housing conditions influence PLA performance. Enriched rats engage in more goal-directed behavior while Single and Raised Enriched rats engage in more cue-directed behavior. Similarly, Raised Enriched rats favor a habit-like "response" strategy in the MWM. Results suggest that measures of individual differences in goal- vs. cue-directed behavior are highly sensitive to stressors such as housing conditions; this should be carefully considered when designing experiments.

Broomer MC, Bouton ME (*U of Vermont*) Renewal, spontaneous recovery, and reacquisition after punishment and extinction ABSTRACT: Punishment and extinction are both effective methods of weakening instrumental responding and may involve similar learning mechanisms. We compared three recovery (relapse) effects—renewal, spontaneous recovery, and reacquisition—following either punishment or extinction of an instrumental response. In Experiment 1, both punished and extinguished responses renewed to similar degrees following a context change at test (ABA renewal). In Experiment 2, responding spontaneously recovered to similar degrees seven days following punishment or extinction. In Experiment 3, responding was rapidly reacquired when the response was reinforced again following extinction but not following punishment, as predicted by the idea that the reinforcer delivered in reacquisition is part of the context of punishment, but not extinction. The results collectively suggest that both punishment and extinction produce similar context-dependent retroactive interference effects. More broadly, they also suggest that punished and extinguished responses may be equally likely to return following a change of context despite the intuition that punishment may provide a more extreme and effective means of suppressing behavior. To our knowledge, this is the first direct behavioral comparison of response recovery after punishment and extinction within individual experiments.

Brunswick CA, Baldwin DJ, Bodinayake KK, Bellfy L, McKenna AR, Stuart EM, Murakami S, Smies CW, Kwapis JL (*Penn State U*) Age-related deficits in spatial memory due to epigenetic dysregulation of Per1 within the retrosplenial cortex but not the suprachiasmatic nucleus ABSTRACT: Aging is associated with deficits in both memory function and circadian rhythm, but the exact relationship between these impairments remains elusive. Growing evidence suggests that these changes might both be due to epigenetic dysregulation of clock genes. Although clock genes are best known for their roles in maintaining circadian rhythms in the suprachiasmatic nucleus (SCN), but they are also expressed ubiquitously. Prior work from our group suggests that the activity of clock gene Period1 (Per1) within the retrosplenial cortex (RSC) might gate the formation of spatial memory. Here, we investigate if age-related changes in Per1 expression within the RSC might contribute to deficits in spatial memory. We trained two cohorts of young (7-weeks) and aging (18-months) C57BL/6J mice in a spatial learning task at six different timepoints (Zeitgeber Times: ZT1, ZT5, ZT9, ZT13, ZT17, ZT21). One cohort was sacrificed 1 hour after training to investigate learning-induced changes in gene expression while the other was tested 24 hours later to examine memory performance. We found that relative increases in Per1 expression are induced by learning in both the RSC and the SCN (regardless of animal age), although absolute levels of Per1 are lower in aging animals. In the RSC, this Per1 induction fluctuated with time-of-day and was greatest during the day (lights on), coinciding with peak memory performance and suggesting a link between Per1 expression here and the formation of spatial memory. Notably, there was no effect of time-of-day on learning-induced Per1 expression in the SCN. To investigate a causative role of Per1 in memory performance, we locally downregulated Per1 expression in the RSC of young mice, which impairs memory performance. Likewise, local upregulation of Per1 in the RSC of aging mice is sufficient to rescue memory formation. Notably, manipulating Per1 expression in the SCN of young mice has no effect on memory performance. In sum, these results expand the known roles of clock genes beyond the SCN and suggest that Per1 expression within the RSC is responsible for linking memory performance to time-of-day. We suggest that brain-wide epigenetic dysregulation of Per1 associated with aging contributes to both age-related memory deficits and disruptions in circadian rhythm. Local dysregulation of Per1 within the RSC (and other memory structures, like the dorsal hippocampus) likely contributes to memory impairments, while dysregulation within the SCN causes circadian disruptions.

Carroll E, Love J, Conoscenti M, Covington A, Zelikowsky M (*U of Utah School of Medicine*) The lateral preoptic area controls the effects of social isolation on mouse courtship behavior and song ABSTRACT: Social communication is a vital component of courtship behavior across species. Through a series of behavioral experiments and acoustic recordings, we have discovered that chronic social isolation (SI) significantly alters mouse mating behavior and associated ultrasonic vocalizations (USVs). Detailed analysis of audio recordings collected from male mice during a homecage mating assay with a female conspecific reveal significant differences in USV production patterns, where isolated mice produce longer song syllables at a lower frequency with a smaller frequency range and fewer frequency jumps than group housed males. Importantly, we found that isolation negatively impacted behavioral chains of events such that Markovian principles were followed in group housed mice but were not present in isolated mice. Moreover, we found a strong correlation between female defensive behaviors and male mating attempts for group housed mice, but this relationship was not present for isolated mice, suggesting

that isolation disrupts the relationship between male mounting behavior and female receptivity. This was supported by follow-up experiments demonstrating that females preferred interacting with group housed males vs. isolated males in a modified 3-chamber social interaction assay. We complemented these behavioral and acoustic approaches with neural circuit analyses. Isolated males with altered USVs showed increased cfos expression in the lateral preoptic nucleus (LPO), a region previously implicated in mouse USVs more generally. To further probe the role of the LPO in SI-mouse song, mice were injected with fluorescently encoded calcium indicators and implanted with GRIN lenses to assess the in vivo activity of neurons in the LPO during mating behavior and USVs. Initial results revealed correlated activity of neurons in the LPO during SI-mouse USVs. Collectively, our findings indicate that SI negatively modulates mouse courtship behaviors and song, and that this modulation is encoded by unique ensembles of neurons in the LPO.

Chen BK, Shah A, Shin G, Hunsberger HC, Denny CA (*Department of Psychiatry, Columbia U Irving Medical Center (CUIMC); Division of Systems Neuroscience, Research Foundation for Mental Hygiene, Inc. (RFMH) / New York State Psychiatric Institute (NYSPI); Doctoral Program in Neurobiology and Behavior (NB&B), Columbia U; Boston U; Center for Neurodegenerative Diseases and Therapeutics, Rosalind Franklin U of Medicine and Science / The Chicago Medical School*) The VPAC2 receptor mediates resilience to stress in female, but not male mice ABSTRACT: Pituitary adenylate cyclase activating peptide (PACAP) is a neuropeptide that has previously been implicated in the pathophysiology of post-traumatic stress disorder (PTSD). Although PACAP binds to the vasoactive intestinal peptide receptor 2 (VPAC2) it is still unknown whether VPAC2 plays a role in the psychopathology of stress-induced psychiatric disease. Here, we hypothesized that administration of the pharmacological antagonist of VPAC2, Bay 55-9837, prevent a wide variety of stress-induced fear, behavioral despair, and anxiety-like behaviors. A single injection of saline, (R,S)-ketamine, or Bay 55-9837 was administered before or after contextual fear conditioning (CFC) stress in male and female 129S6/SvEv mice (n = 5-18 mice per group). Drug efficacy was assayed using the forced swim test (FST), elevated plus maze (EPM), open field (OF), novelty-suppressed feeding (NSF), contextual fear discrimination (CFD), and Piezo sleep boxes. Brain-wide VPAC2 expression was assayed using immunohistochemistry. Activating VPAC2 prior to stress attenuated learned fear, reduced behavioral despair, suppressed hyponeophagia, and facilitated CFD in female, but not male mice. Prophylactic Bay 55-9837 administration protected against stress-induced changes in sleep/wake cycles in female mice. Administration of Bay 55-9837 after stress reduced behavioral despair and hyponeophagia in both sexes. CFD learning in female mice upregulated VPAC2 expression in hippocampal CA3 and the agranular insular cortex. Our data indicate that agonism of the VPAC2 receptor using the peptide agonist Bay 55-9837 suppresses fear behavior and facilitates CFD learning in female, but not male mice. Overall, these results suggest that VPAC2 receptor activation critically modulates fear learning and retrieval in a sex-specific manner. Overall, our results suggest that VPAC2 may be a novel, female-specific target for preventing and treating stress-induced psychiatric disorders. SUPPORT: This work was supported by an NIMH F31MH121023 to BKC, an NIMH T32MH126036 to AS, an NIA K99AG059952 to HCH, and an NICHD R01HD101402 to CAD.

Conoscenti MA, Sattler KP, Kennedy A, Zelikowsky M (*U of Utah, Northwestern U*) Social isolation and footshock stress produce aggression that is behaviorally and biologically distinct. ABSTRACT: Social interaction is central to mammalian life. Inter-animal aggression represents one of the chief forms of social interaction across species. Importantly, aggression serves to establish stable social hierarchies, secure mates, and defend limited resources. While aggression is highly adaptive, it can also be deleterious when elicited inappropriately or in excess, as observed following extreme stress. While recent work has begun to elucidate the neural circuit mechanisms underlying aggression, little work has examined the mechanisms of violence produced by stress, and virtually no work has compared the effects of various stressors on aggression. Here, we test the hypothesis that aggression induced by social isolation differs behaviorally and biologically from that induced by footshock. C57Bl6/N mice were exposed to three weeks of chronic social isolation stress (SIS) or an acute footshock stressor (FS; 10, 1mA shocks randomly distributed across a 60-minute session). Mice were tested for an array of social behaviors, including aggression, using the resident intruder assay. We found that SIS and FS mice approached the intruder from different directions prior to attack and directed aggression towards different parts of the intruder's body. Subsequent analyses using our machine vision action recognition system (MARS) and unsupervised computational approaches were able to decode an animal's condition based off of investigatory behavior prior to attack. These behavioral differences suggest that FS-induced aggression may be mediated by a dissociable biological mechanism from SIS aggression. We hypothesized that the bed nucleus of the stria terminalis (dBNST) may play a selective role in FS- but not SIS-induced aggression, due to its canonical role in anxiety, sensitivity to footshock, and mediating role in an array of social behaviors. To test this, we chemogenetically inhibited dBNST using hM4Di DREADDs and found that FS, but not SIS aggression was reduced. We next employed a combined RNAscope + retrograde tracing approach to genetically identify the population of dBNST cells which are activated following FS-aggression and that send projections to the ventrolateral sub-

division of the ventromedial hypothalamus (VMHvl), a region implicated in the control of mouse aggression. We are further dissecting the necessity and sufficiency of genetically defined dBNST-VMHvl neurons in FS-aggression using cre-dependent chemogenetic manipulations during the resident intruder assay. Taken together, our findings suggest that exposure to footshock results in a unique aggression phenotype that may be mediated by unique biological circuits at the intersection of stress and aggression.

Denholtz LE[1,2], Liu J[3], Nahmoud I [4], Casaccia P[3], Likhtik E[1,2] (*[1]Biology Program, The Graduate Center, CUNY, New York, NY; [2]Biological Sciences, Hunter College, CUNY, New York, NY; [3]Neuroscience, Advanced Science Research Center, CUNY, New York, NY; [4]Wayne State U School of Medicine*) Safety learning mediated changes in myelination at the medial prefrontal cortex. ABSTRACT: Anxiety disorders are characterized by fear generalization, resulting in inappropriate fear responses to non-threatening cues. Safety training establishes a salient association between a cue and safety and was shown to enhance discrimination between novel non-threat and threat, suggesting that it is an effective approach to combat excessive fear expression. However, it is unknown how safety training mediates its protective effects. We used RNA sequencing analysis of the mPFC in mice after discrimination learning post safety- or auditory fear- conditioning. We found that safety training upregulated myelin-associated transcripts in the mPFC compared to fear conditioning, suggesting that safety training affects the process of adult myelination in the mPFC. To understand the short- and longer-term effects of safety conditioning on myelination, we next asked whether safety learning also impacted myelin proteins and myelinated tracts. Immunohistochemistry revealed upregulation of myelin basic protein (MBP) following remote (21 days) memory recall of both learned fear and safety cues in the prelimbic and infralimbic cortices of the mPFC. We also found that recent recall (24h) was associated with increased oligodendrocyte progenitor proliferation, and remote recall (21 days) was associated with mature oligodendrocytes, suggestive of enhanced differentiation. Using immunostaining with paranodal markers to measure internodal segment length, we found higher internodal segment length in the mPFC after remote cued-safety recall relative to cued-fear recall. Collectively, these results suggest distinct effects of safety versus cued fear learning on myelin. Given that myelination optimizes signal propagation, these results suggest that safety training driven myelin plasticity may enhance communication between the mPFC and other structures, such as the basolateral amygdala and hippocampus, during threat-safety discrimination. Future work will elucidate how myelination affects circuit communication in these pathways during safety learning and retrieval.

Dhillon PS, Halcomb CJ, Philipp TR, Cox JH, Vanderhoof SO, Jasnow AM (*U of South Carolina School of Medicine*) Sex differences in the acquisition and extinction of platform mediated avoidance: Role of glucocorticoid receptors ABSTRACT: Women are more likely to develop an anxiety or stress-related disorder compared to men, but core mechanisms for this sex bias are still not yet fully defined. In addition, while avoidance is a major diagnostic criterion for anxiety disorders, the neurobiological mechanisms of persistent avoidance are not clear. Rodent models of active avoidance have recently been used to understand the acquisition, maintenance and extinction of avoidance associated with anxiety disorders. Here, we used platform mediated avoidance to investigate learning and extinction differences between male and female C57BL/6 mice. Mice were trained using 5 tone-shock pairings each day for three consecutive days. An exclusion criterion was established whereby on the third training day, if mice did not avoid 3 or more shocks they were excluded from further analysis. We found a robust sex difference in the acquisition and extinction retention of platform avoidance, whereas all mice extinguished avoidance initially. Female mice displayed deficits in the acquisition and extinction memory of avoidance compared to males, but there were no differences observed in freezing between the sexes. Next, we trained males using a 10-day training procedure to investigate the impact of increased training sessions on extinction retention. Under these training conditions, males displayed similar deficits to females in the retention of avoidance extinction. Next, we investigated the role of glucocorticoid receptors (GR) in mediating persistent avoidance. The GR antagonist, Mifepristone, was administered during platform avoidance training in males and females. Here we found that mifepristone reduced tone-elicited freezing, but not avoidance in males, whereas it significantly reduced avoidance in females suggesting GR activation contributes to impairments in extinction retention of cue-mediated avoidance in females.

DiFazio LE, Jia R, Greer Z, Sharpe MJ (*UCLA*) The temporal dynamics of encoding fear memories in the BLA, and how they change following experience with rewards ABSTRACT: Decades of research has shown that lesions or inactivation of the basolateral amygdala (BLA) disrupts the acquisition and storage of memories during Pavlovian fear learning (i.e. where a tone is paired with a shock; Cousens & Otto, 1998, Behavioral Neuroscience; Maren, Aharonov & Fanselow, 1996, Behavioral Neuroscience; Phillips & LeDoux, 1992, Behavioral Neuroscience). As a result, current models of fear learning posit that information about the stimuli and aversive event arrive in the BLA and become an associative fear memory (Maren & Quirk, 2004, Nature Reviews Neuroscience; Pitkänen, Savander & LeDoux, 1997, Trends in Neurosciences). Optogenetics allows us to investigate the temporal dynamics of the BLA's involvement in fear learning in a way that was not previously possible. We investigated the effect of optogenetic inhibition of glutamatergic

neurons in the BLA at different time-points during Pavlovian fear conditioning. We found that inhibition of BLA neurons during the tone, but not the shock, disrupted learning. This demonstrates that the BLA is necessary for learning about the shock-predictive tone, but processing of the shock itself remains intact when BLA activity is inhibited. Following recent evidence that the lateral hypothalamus becomes necessary for fear learning following experience with reward learning (Sharpe et al., 2021, Nature Neuroscience), we investigated how reward learning experience might impact the role of the BLA in fear learning. In a new experiment, one group of rats learned a light-food association, while the "naïve" control group had the same experience with the light alone. Then, in a different context, these rats experienced Pavlovian fear conditioning. Here, we inhibited pyramidal neurons in the BLA during the tone. We found that BLA inhibition disrupted fear memories in naïve rats, replicating our prior result. However, in rats with prior reward learning experience, BLA inhibition did not affect fear memories. These results show that reward learning shifts the fear circuit towards the lateral hypothalamus and away from the BLA.

Dunsmoor JE, Cooper SE (*U of Texas at Austin*) Latent associative structures facilitates transfer of learned fear ABSTRACT: Animals have a remarkable ability to rapidly learn about and remember signals of threat in the environment, and then generalize this learning to a host of related stimuli. One facet of threat generalization, which has received limited attention in contemporary neuroscience, involves incorporating associative knowledge structures to infer threat of novel stimuli. In a paradigm known as sensory preconditioning, participants form a latent association between two stimuli in the absence of reinforcement. If one of these stimuli is later predictive of an aversive outcome, the pre-associated stimulus can acquire the capacity to elicit a defensive conditioned response—despite never having been directly linked to an aversive experience. Neuroscience research in rodents shows that this form of fear generalization requires the hippocampus and orbitofrontal cortex. The neurobehavioral mechanisms of sensory preconditioning in humans is unclear, but insights could offer clues on how fear learning broadly generalizes to a host of seemingly inconsequential stimuli following highly aversive events (e.g., trauma). Here, we used a novel sensory preconditioning task during fMRI in healthy adults to investigate how latent associative networks facilitate the transfer of learned fear. During functional MRI, participants first learned that a category of images (preconditioned stimulus, PS; animals or tool, counterbalanced) predicted a picture of a square, while another PS category (tools or animals, respectively) predicted a circle. Next, one shape (conditioned stimulus, CS+) was paired with an electrical stimulation to the wrist (unconditioned stimulus, US), while the other shape (CS-) was unpaired. Subjects then viewed novel category exemplars (PS+) from the category originally paired with the CS+ and exemplars (PS-) from the category originally paired with the CS-. Subjects returned 24 hours later for a surprise recognition memory test of PS+/PS- exemplars encoded before and after threat conditioning the previous day. Participants acquired conditioned learning to the CS+, expressed behaviorally through skin conductance responses and expressed neurally through differential (CS+ > CS-) activity in the thalamus, insula, and dorsal anterior cingulate cortex (all fMRI activity $p < .001$ corrected for multiple comparisons at $p < .05$). A region of interest analysis of the transfer test revealed greater differential hippocampal and orbitofrontal activity to the PS + > PS- ($p < .001$, two-tailed t-test). Finally, participants had a selective 24-hour memory bias for PS+ > PS- items encoded prior to fear conditioning ($p = .02$, two-tailed t-test). These results provide neurobehavioral evidence for how latent associative structures support the transfer of learned fear to stimuli that indirectly acquired emotional value. This extends evidence from rodent neuroscience on the role of the hippocampus and OFC in sensory preconditioning, and offers insights into key neurocircuity implicated in the higher order transfer of emotional learning. Sensory preconditioning may be a valid but underappreciated model for widespread and idiosyncratic nature of fear generalization following highly aversive events, such as trauma.

Farrell K, Navabpour S, McFadden T, Musaus M, Auerbach A, Ray WK, Helm RF, Jarome TJ (*Virginia Polytechnic Institute and State U*) The role of sex in the necessity for diverse polyubiquitin modifications in memory formation ABSTRACT: The ubiquitin-proteasome system (UPS) controls the majority of protein degradation in cells via the small modifier ubiquitin. Multiple ubiquitin molecules can bind a target substrate and form polyubiquitin chains at 8 different linkage sites, the most abundant of which are linked at lysine-48 (K48) or lysine-63 (K63), the former of which is preferentially targeted for degradation by the proteasome while the latter is thought to be independent of the degradation process. Over the last decade, numerous studies have demonstrated the necessity of UPS-mediated protein degradation for memory formation in the brain, focusing the K48 polyubiquitin mark. However, the role of degradation-independent ubiquitin signaling, such as K63 polyubiquitination, during memory formation remains unknown. Furthermore, the literature has focused almost exclusively on male rodents, leaving unanswered questions about the importance of degradation-dependent and independent ubiquitin signaling for memory formation in females. Recently, we reported the first evidence demonstrating that while male and female rats both need K48 polyubiquitin-mediated protein degradation to form contextual fear memories in the amygdala the protein targets vary widely across sexes, suggesting unique functional roles for protein degradation to form the same memory. Conversely, in the present study, we present the

first evidence that degradation-independent K63 polyubiquitination is selectively involved in fear memory formation in females. Using a K63-specific Tandem Ubiquitin Binding Entity (TUBE) with liquid chromatography mass spectrometry (LC/MS), we identified 13 proteins that gained a K63 polyubiquitin mark in the female amygdala following fear conditioning, including a major proteasome subunit, with no proteins identified in males, suggesting that changes in K63 polyubiquitination may be specific to females following fear conditioning. Using a novel CRISPR-dCas13b RNA-editing approach to make site-specific modifications on the major ubiquitin coding gene, Ubc, at the nucleotide sequence coding for K63, we found that K63 polyubiquitination was necessary for contextual fear memory formation in the female, but not male, amygdala. Consistent with our proteomic data, loss of K63 polyubiquitination in females resulted in a reduction in learning-related increases in proteasome activity in the amygdala. This suggests that proteasome-independent K63 polyubiquitination is involved the regulation of the protein degradation process in the female amygdala during fear memory formation. Together, these findings provide the first evidence of a novel, sex-selective role for K63 polyubiquitination in the regulation of proteasome activity in the female amygdala during fear memory formation.

Ferrara NC, Trask S, Ritger AC, Padival M, Rosenkranz JA (*Rosalind Franklin U, Purdue U*) Brief isolation alters maturing cortical-amygdala circuits supporting social preference in adults and adolescents ABSTRACT: Several neuropsychiatric disorders are characterized by changes in social behavior, and disorder diagnosis is high during adolescence. The social environment both contributes to and protects against disorder diagnosis, highlighting a need to understand the interactions between the social environment and behavior during development. The amygdala and its cortical inputs mature throughout adolescence, and amygdala abnormalities have been implicated in a number of behaviors that change over the course of development, including social behavior. The anterior cingulate cortex (ACC) and the basolateral amygdala (BLA) are critical for social behavior and are sensitive to shifts in the social environment. ACC inputs to the BLA are critical for social learning in adults, and may therefore play an important role in the developmental transitions in behavior critical for social memory and sensitivity to the social environment. Here, we investigated the impact of brief social isolation (2hrs) on social behavior and social preference as well as the neural processes sensitive to isolation. We found that brief isolation facilitated sociability in both adults and adolescents that was dependent on ACC activity during isolation. However, the influence of isolation had opposing effects on novel partner preference, often used as a measure of social memory, between age groups. In adolescents, the same isolation that facilitated social behavior also increased novel partner preference but reduced this preference in isolated adults. We next investigated the neural processes sensitive to isolation across ages. We found isolation increased ACC activity, including ACC neurons projecting to the BLA, in both adults and adolescents. This same isolation increased ACC driven changes in BLA activity, but in a frequency dependent manner that differed between ages. This suggests that developmental differences in ACC-driven BLA activity following isolation may underlie changes in age-specific social behaviors that differentially influence social preference. Collectively, this work provides insight to the maturation of cortico-BLA circuitry that contribute to shifts in social behavior and memory. SUPPORT: MH118237(JAR), F32MH122092 (NCF).

Fleischer AW, Ramirez S, Frick KM (*U of Wisconsin-Milwaukee (Fleischer, Frick), Boston U (Ramirez)*) Tet-tagging as a method to test the estrogenic Two-Step Wiring Hypothesis ABSTRACT: Estrogens promote spatial and object identity memory consolidation by acting as neuromodulators in brain regions such as the dorsal hippocampus (DH). In object memory tasks, object exploration triggers neuronal activity in brain regions such as the dorsal hippocampus (DH), an area critical for the consolidation of memories for the location and identities of objects. This increased activity generates new contacts between neurons to form a memory engram. However, if the magnitude and duration of this stimulus is insufficient, the resulting neural contacts subsequently fade and do not support intact recall of the learned information. The potent estrogen, 17β-estradiol (E2), facilitates the consolidation of spatial and object recognition memories via a wide array of molecular mechanisms that trigger dendritic spinogenesis and increase synaptic plasticity. The concordance of these stimuli – that is, learning- and E2-induced cellular activation – has been proposed to give rise to superior recall abilities by recruiting more cells into the memory engram, as well as strengthen their connections, than either stimulus alone. This process has been termed the Two-Step Wiring Hypothesis by Srivastava et al. (2008, 2012). However, which cells are activated by each stimulus is still under investigation. Tet-Tag viruses allow tracking and identification of individual cells recruited into a memory engram with precise temporal control, which might be utilized to further test the Two-Step Wiring Hypothesis. Here, we tested the extent to which Tet-Tagging could be used to identify neurons activated by training and/or E2 treatment in ovariectomized female mice at different delays following surgical recovery. Activity in tagged cells was indicated by viral expression of GFP protein and colocalization with the cellular activity marker c-fos. We expected that viral expression would be evident within 1 week of surgery and hypothesized that training + E2 treatment would provide the greatest number of tagged c-fos+ cells as compared to vehicle-treated and homecage controls. Female C57BL/6 mice were ovariectomized, infused bilaterally with a 1:1 viral mixture

of AAV9-c-fos-tTa and AAV9-TRE-EGFP into the DH, and implanted with bilateral guide cannulae aimed at the DH. Mice were given 1 or 2 weeks to recover from surgery prior to behavioral training. Mice were kept on doxycycline to repress the production of GFP protein until 2 days prior to object training, in which mice explored 2 identical objects for an accumulated 30 s. Upon completion of object exploration, mice were infused into the DH with either vehicle or E2 and perfused 90 minutes later. Homecage controls were handled for 30 s and perfused 90 minutes later. Brains were sectioned at 30 μm and immunohistochemically labeled for c-fos, GFP, and DAPI. Finally, the dorsal dentate gyrus and dorsal CA1 of each section (n = 4-6/brain) were quantified bilaterally for c-fos, GFP, or DAPI, as well as their overlap. Preliminary results suggest that viral uptake is sufficient by 1 week following surgical procedures, and that training and infusion of E2 upregulate GFP and c-fos in the DH. These data support the potential use of Tet-Tagging as a method to investigate the Two-Step Wiring Hypothesis, and subsequent research will utilize this technique to probe for the identities of different neural and glial cells involved in estrogenic memory enhancement. SUPPORT: Supported by National Institutes of Health grants R01MH107886, R15GM118304, and R01MH117964, a UWM Discovery and Innovation Grant (101x418), and a Distinguished Graduate Student Fellowship to AWF. Miranda Schwabe and Dr. Lisa Taxier also provided invaluable technical and conceptual advice for this work.

Franco CY, Knowlton BJ (*UCLA*) Childhood racial discrimination effects on habitual responding and disordered eating ABSTRACT: Previous studies have shown that childhood experiences with trauma and abuse facilitate habitual responding, potentially predisposing individuals to developing repetitive maladaptive behaviors. However, such examinations have not accounted for other types of early trauma like childhood exposure to racial or ethnic discrimination, nor have they expanded to examine real-world maladaptive outcomes in relation to laboratory-derived measures of habitual responding frequency. As such, we examined how childhood racial/ethnic discrimination (CRED) influenced habitual responding and determined whether these two factors predicted disordered eating, alcohol use, and drug use behaviors. Forty young adults (20.15 ± 1.49 years old; 30 female, 10 male) completed a noise avoidance task, during which they viewed abstract stimuli and pressed an associated key to avoid a screaming sound delivered via earphones, followed by a devaluation test to examine avoidance habits. Habitual responding was calculated as the number of responses made to the now devalued stimulus at test. Participants also completed questionnaires examining childhood racial/ethnic discrimination, disordered eating behaviors, alcohol and drug use, and other neuropsychological variables. Results from linear regression analyses revealed that childhood exposure to threat or aggression due to racial/ethnic background was a positive predictor of habitual responding (b = 6.362, p = 0.035), consistent with effects of other types of early life stressors on habit responding. Results further revealed that frequency of habitual responding, alongside other neuropsychological variables like trait anxiety and depression, significantly negatively predicted frequency of restraint or avoidance of eating (b = -0.178, p = 0.031) and eating concern (b = -0.092, p = 0.030). Neither CRED nor habitual responding significantly predicted alcohol or drug use (ps > 0.228), nor were significant gender differences found on any of the measures reported here (ps > 0.174). These results support extant literature showing that a variety of early life stressors predict degree of habit responding in young human samples. They also indicate that laboratory derived measures of habitual responding, in conjunction with other neuropsychological variables, can predict real-world health behaviors like disordered eating.

Fukunaga Y, Gonzalez SE, Lock E, Schulman E, Cazares VA (*Psychology Department and Neuroscience Program, Williams College*) Exposure of conditioned stimuli in multiple contexts enhances fear extinction memories ABSTRACT: Fear extinction is context-specific; thus if an extinguished conditioned stimulus (CS), such as a tone, is experienced in any context except for the one in which it was extinguished, the CS will re-elicit the fear response, a phenomenon termed fear renewal. We have previously shown that fear extinction training in multiple novel contexts overcomes fear renewal, enhances fear extinction learning, and improves extinction recall 7 days later. The present study extends these findings to report that multiple context fear extinction (MCFE) suppresses spontaneous recovery (when tested 30 days after extinction). We also report that contextual novelty in MCFE is not necessary since exposure to multiple familiar contexts still enhances fear extinction memories. Furthermore, we show that enhancement of fear extinction in the MCFE paradigm requires the hippocampus since chemogenetic activation or inhibition of hippocampal cells impairs fear extinction recall in animals that were trained in multiple contexts, but not those trained repeatedly in a single context. Finally, to ascertain differences in neural activation between multiple- and single-context fear extinction on day 2 and day 3 of training, we labeled Fos-expressing neurons using TRAP (Targeted Recombination in Active Populations) and immunohistochemistry. Altogether, our results suggest that hippocampal cell activation is necessary for generalization and consolidation of contextually distinct extinction memories. SUPPORT: We thank VAC Lab members & alumni, Jack Snyder (animal care staff), Roffe Fellowship, the U.S.-Japan Council Watanabe Scholarship, the Wilmers Research & Travel Fellowship and all the individuals who supported our research.

Gagon J, Gatlin RE, Zelikowsky M (*U of Utah*) Control of isolation-induced aggression through activation of me-

dial prefrontal cortex pyramidal neurons ABSTRACT: The effects of social isolation are deeply felt throughout many populations. One example is prison inmates who have been placed in solitary confinement are more likely to experience suicidal ideology, irritability, and aggression. Furthermore, mouse models have shown social isolation in mice increases aggression in comparison to group-housed animals. Previous work has established that mPFC pyramidal neurons project to multiple subcortical aggression centers, and that activation of such neurons in mPFC reduces aggression. Despite this, no work has focused on how this population of neurons may regulate social isolation-induced aggression nor have these neurons been studied in females. Here, we expand on previous work by examining whether activation of mPFC pyramidal neurons reduces aggression in socially isolated female mice. To this end, we infused a virus encoding the excitatory DREADD, hM3D, fused to mCherry, under control of the CaMKII promoter into the mPFC of female C57Bl6/N mice (N=8). A virus expressing mCherry without the hM3D DREADD under the control of the CaMKII promoter was used as a control (N=9). Following surgery, all mice were socially isolated for 4 weeks to induce aggression. After isolation, a within subject's design was used to examine the impact of DREADD-mediated activation on aggression. Each animal was tested twice on the resident intruder assay- once in which the DREADD ligand, Descholorclozapine (DCZ) administered via i.p, injection and once with vehicle- 48 hours apart. The viral conditions were counterbalanced such that half the animals in each viral condition received DCZ on the first test day and half received an injection of the vehicle. Activation of mPFC pyramidal neurons significantly decreased aggression in isolated mice (Repeated Measures ANOVA, $p<.05$). These results support the hypothesis that the mPFC plays an inhibitory role in aggressive behavior, and mPFC pyramidal neurons are essential to this inhibitory control. SUPPORT: U of Utah Office of Undergraduate Research, U of Utah Department of Neurobiology

Garcia AK, Kendall RK, Wikenheiser AM (*U of California, Los Angeles*) Methamphetamine reduces the duration of foraging bouts in rats performing a naturalistic decision-making task ABSTRACT: Knowing when to abandon a depleting food source is critical for efficient foraging. Persisting for too long with a dwindling resource may mean missing out on better opportunities elsewhere, while abandoning locations too readily may mean spending too much time searching for better options. One prominent theoretical model proposes that tonic dopamine levels encode the average rate of reward in an environment, a critical determinant of how long to persist with a food source at hand. Here, we test whether increasing dopamine levels via administration of methamphetamine decreases persistence with potential food sources, in line with theoretical models of dopamine function. Rats performed a spatial foraging task in which they earned food from two distinct foraging patches. Within each patch, the rate of food delivery decreased over time following an experimenter-controlled schedule. The travel cost of switching between patches was manipulated by imposing a delay between leaving one patch and entering another. Before each session, rats were administered methamphetamine (1 mg/kg) or a control saline injection. Our main behavioral measure of interest was how long rats persisted on visits to each foraging patch. Consistent with theoretical models of foraging behavior, rats remained longer in patches with slower depletion rates, and remained longer in all patches when the travel cost was high. Methamphetamine caused a significant reduction in patch residence duration relative to saline sessions. Importantly, methamphetamine did not alter rats' sensitivity to travel cost or patch depletion rate. However, methamphetamine significantly reduced rats' consumption of food pellets, and strongly affected the way rats moved. Compared to saline injections, methamphetamine first decreased, but over sessions came to increase, the amount of time that rats spent stationary or moving at very slow speeds. Methamphetamine increased rats' running speed during bouts of movement and increased thigmotaxis. Together, these data are consistent with a role for dopamine in encoding average rate of reward, but more specific manipulations will be necessary to disentangle the motivational, motor, and decision making functions of dopamine release.

Garrett N, Allan SM, Daw ND (*U of East Anglia*) Model based habits ABSTRACT: A core idea across psychological and neural theories is that the brain simplifies laborious computations using habits. A well-developed version of this idea comes from associative learning accounts and concerns the trade-off between habits (or stimulus-response associations) and goal directed actions. Reinforcement learning operationalises these two behaviours as arising from two distinct systems for learning - model free (MF) and model based (MB) respectively. Here we present a new approach to MB in which agents are given the capacity to maintain separate sets of their environment's dynamics (formally a Markov Decision Process, MDP). Which MDP to update and use for planning is modelled as a process of Latent Causal Inference (LCI). We test the theory in a series of simulations that examine whether this exclusively MB learner can generate the devaluation insensitive decisions interpreted as a hallmark of habits (i.e., arising from MF), in a classic instrumental reinforcer devaluation procedure widely studied in rodents. Our simulations reveal that segregating MDP learning during acquisition (during which agents leverpress to obtain food) and devaluation (during which food is devalued via pairing it with illness) phases into different latent causes, prevents devaluation generalizing to a subsequent lever test (an instrumental probe used to assess habit formation). This gives rise to insensitivity to reinforcer devaluation (persistent lever pressing in the lever test) despite the model omitting any habit-

ual (MF) component from the model. "Ersatz habits" (just like laboratory ones) emerge after overtraining, interact with contextual cues and show preserved sensitivity to reinforcer devaluation in a consumption test (a standard control). While these results do not rule out habit formation per se, they highlight the need for caution in using devaluation procedures to rule them in (or out) and offer a new perspective on the neurocomputational substrates of repetitive harmful behaviours prevalent in clinical pathologies such as OCD and addiction.

Gatlin, RE, Gagon J, Winebrenner C, Zelikowsky M (*Neuroscience Graduate Program; Department of Neurobiology; U of Utah*) The role of the mPFC/LS Tachykinin 2 system in isolation induced aggression ABSTRACT: Chronically isolated mice show altered behaviors, including increased aggression, however the neural circuitry underlying this aggression is poorly understood. Both the medial prefrontal cortex (mPFC) and lateral septum (LS) are strongly involved in social and emotional behaviors, including aggression, however much of this work lacks cell-type specificity. Here, we propose a role for neurokinin-3 receptor (Nk3R)-expressing neurons in the zone where dorsal mPFC transitions to anterior LS (mPFC/LS) in modulating isolation-induced aggression. Previous work has established a role for the Tachykinin 2 (Tac2) neuropeptide system in isolation-induced alterations to behavior. Tac2 encodes the neuropeptide Neurokinin B, which binds to the Neurokinin 3 receptor (Nk3R), and while it is known that both genes are expressed both in the mPFC and LS, there has been no work performed to understand the role of this neuropeptide system in these regions in mediating isolation induced aggression. Previous work suggests that Nk3R-expressing neurons in cortex are pyramidal neurons and when activated, mPFC pyramidal neurons reduce aggression. Additionally, activation of specific LS to the ventrolateral subdivision of the ventromedial hypothalamus (VMHvl) reduces aggression, yet it is not known what specific cell types comprise this projection. Thus, to begin to understand the role of the mPFC/LS Tac2/Nk3R system, we used a viral-mediated chemogenetic approach in Nk3R-Cre mice to test whether activation of mPFC/LS Nk3R+ cells was sufficient to block isolation-induced aggression. Following surgeries to express either Cre-dependent hM3D DREADD fused to mCherry or mCherry only (controls) in mPFC/LS, mice were single housed for 4 weeks, then tested on the Resident Intruder assay twice. Ten minutes before the assay, mice were given an i.p. injection of either the DREADD-ligand, Deschloroclozapine (DCZ; 1/*mu*g/g), or a vehicle solution. On the second test day 48hrs later, the animals received the opposite solution (order counterbalanced). Activity levels (total distance traveled) were assessed through automated tracking (Noldus Ethovision) during the three-minute baseline phase before the introduction of the intruder on both test days. Aggression (number of attack bouts and total time engaged in attack) towards the Balb/c intruder was then hand-scored by a blinded experimenter (Noldus Observer). DREADD-mediation activation of Nk3R+ neurons significantly reduced both the time animals spent attacking as well as the number of attack bouts. Importantly, neither activation of these Nk3R-neurons nor DCZ alone altered activity levels. These findings highlight a fundamental, previously unknown role for Tac2/Nk3R signaling in mPFC/LS in the control of isolation-induced aggression. SUPPORT: 1F31MH131359-01; Whitehall Foundation

Georgiou P, Sklirou M, Zanos P, Kalk N, Lingford-Hughes A, Wells LA, Bailey AD (*Department of Psychology, U of Wisconsin-Milwaukee; Department of Biological Sciences, U of Cyprus*) Assessing the role of neuroinflammation in ethanol withdrawal induced memory deficits ABSTRACT: Chronic ethanol use and withdrawal is well known to cause cognitive and motor impairment; however, the neurobiological mechanisms driving those behavioral changes are still not clear. Emerging evidence suggests a role of microglia activation (i.e. neuro-inflammation) in the mechanisms underlying neuropsychiatric disorders. Here, we present novel data illustrating the effect of chronic ethanol use and of acute, medium and long-term withdrawal on a cerebral neuroinflammation marker and its association with memory impairments. 18kDa translocator protein (TSPO) is sparsely expressed in the brain parenchyma, and resting glia, but richly expressed in activated glial cells. We also assessed the effect of chronic ethanol use and of acute, medium- and long-term abstinence on mGlu5 glutamate receptors which is also known to be associated with neuroinflammation and alcohol addiction. C57Bl/6J mice were treated with an escalating dose of ethanol-containing diet or isocaloric replacement diet for ten days. Different mice cohorts were then let to spontaneously withdraw for one, four or seven days and underwent behavioral assessments of cognition and coordination, using the novel object recognition (NOR) and Rota-Rod, respectively. Chronic ethanol treatment had a negative impact on motor coordination which persisted for one day post-withdrawal from ethanol but recovered after 4 days withdrawal. Ethanol treatment did not have a significant effect on memory as evaluated by NOR; however, ethanol-withdrawal resulted in a reduction of memory with a lack of object recognition on withdrawal days 1 and 7. Chronic ethanol treatment and one day post-ethanol withdrawal lead to a transient significant increase in TSPO binding in several brain regions. Moreover, an overall group increase in mGlu5 binding was observed in several key brain regions of alcohol-treated mice compared to controls. Significant negative correlations were identified between binding levels and memory scores in several brain regions in ethanol treated and abstinent mice. The results of this study indicate neuroinflammation as a possible mechanism driving cognitive and behavioral changes observed in alcohol dependent individu-

als undergoing withdrawal.

Giovanniello J, Paredes N, Uwadia H, Oregwam C, Sehgal M, Nnamdi G, Malvaez M, Adhikari A, Silva A, Wassum K (*Department of Psychology, U of California Los Angeles*) Opposing amygdala-striatal pathways enable chronic stress to hasten habit formation ABSTRACT: When making decisions, we prospectively evaluate all of our potential actions and their consequences before executing a behavior. This allows us to adapt when situations change but it is also cognitively taxing. We also use habits – a more resource-efficient but inflexible strategy in which behaviors are executed without thought of their consequences based on past success. Balance between goal-directed and habitual behavioral control strategies permits adaptive and efficient behavior. However, overreliance on habits is an endophenotype of many psychiatric conditions. Stress is a major contributing factor to these conditions and can tip the balance of behavioral control towards habit. The neural mechanisms that allow this to happen are unclear. Thus, the goal of this project is to uncover the brain processes by which stress hastens habit formation. We focused on amygdala-striatal projections because the dorsomedial striatum (DMS) is critical for controlling goal-directed learning and the amygdala is a known stress hub also implicated in behavioral control. While direct excitatory projections from the basolateral amygdala (BLA) to the DMS have long been known to exist, recent evidence has revealed the central amygdala (CeA) also directly projects to the dorsal striatum. We identified that these inhibitory CeA projections primarily target the DMS. To examine whether and how BLA->DMS and CeA->DMS projections mediate the influence of stress on habit formation, we coupled a task to model chronic stress-induced acceleration of habit learning in mice with pathway-specific fiber photometry calcium imaging or chemogenetic manipulations. We found that BLA->DMS and CeA->DMS have opposing activity patterns during instrumental learning that are oppositely modulated by chronic stress. We also found that these amygdala-striatal projections differentially regulate habit learning. CeA->DMS activity opposes goal-directed learning and permits stress to promote habits. Conversely, BLA->DMS activity opposes the influence of stress on habits and is sufficient to restore normal goal-directed behavioral control. In addition to establishing a function for the newly-discovered CeA-DMS pathway, these data indicate that amygdala-striatal projections have opposing function in goal-directed and habit learning and are differentially regulated by stress to promote habit. This has important implications for conditions influenced by stress and characterized by maladaptive habits. SUPPORT: This work was funded by NIH R01DA046679 (KW), NIH T32DA024635 (JG), NIH F32DA056201 (JG), A.P. Giannini Fellowship (JG), and NIH TL4GM118977 (NP).

Glasper ER, Walker SL, Beyene R (*The Ohio State U Wexner Medical Center, Department of Neuroscience and Institute for Behavioral Medicine Research, Columbus, OH 43210, USA*) Early-life rearing influences sex differences in social learning and neuroimmune function. ABSTRACT: Adverse early-life experiences, like parental neglect, increase susceptibility to anxiety disorder development that often accompanies social avoidance behaviors and neuroinflammation. Social avoidance involves reduced time engaging with others and the inability to initiate interactions. This may be due to reduced social motivation (i.e., a desire to interact) or a result of distress experienced during social interactions that is learned over time. In rodents, social approach is reduced by social stressors and is accompanied by increased social vigilance (i.e., looking at, but not approaching, an unfamiliar individual from a safe distance). In humans, reduced social approach along with increased social vigilance may predict the development of social anxiety disorders and neuroimmune dysfunction. This knowledge greatly stems from studies investigating uniparental rodents, like Rattus and Mus, that do not engage in biparental care. In biparental species, paternal care significantly improves offspring survival and development. Our knowledge of paternal contributions to mental health disorder prevalence is understudied due to the rare expression of biparental care in mammals. Thus, our understanding of the consequences of paternal absence on offspring social behavior and neuroimmune functioning remains unexplored. Using the biparental California mouse (Peromyscus californicus), we examined to what extent neonatal paternal deprivation induces social impairments and pro-inflammatory cytokine production in adulthood during a low-risk social interaction. Adult biparentally-reared and paternally-deprived (permanent removal of the father 24h after offspring birth) California mice were assessed for sociability, preference for social novelty, social anxiety, and social vigilance within a 3-chambered social interaction apparatus in response to same-sex conspecifics. Region-specific pro-inflammatory cytokine concentrations were determined via Luminex multianalyte analyses. Paternal deprivation impaired social motivation and increased social anxiety-like behavior in males, but not females. This paternal deprivation effect on social behavior was associated with more hypothalamic interleukin-1/*beta* protein concentration in males. A simple linear regression was used to predict social vigilance behavior during social motivation testing on Day 1. Social vigilance explained a significant amount of variance in time spent interacting with the unfamiliar conspecific in paternally-deprived males and females. The regression indicated that an increase in social vigilance corresponded with less time in the chamber with the unfamiliar same-sex conspecific. This relationship was absent in control-reared adult offspring. During social anxiety testing, less social vigilance was observed in paternal deprivation-reared males, but not females; however, social

vigilance no longer explained the variance observed in social anxiety-like behavior. These data suggest the adverse early-life experience of paternal deprivation may disrupt social anxiety-like behavior in males, but not in females. This impairment in social behavior was observed after repeat exposure to a non-threatening, same-sex conspecific, suggesting that social learning may also be impaired in paternally-deprived males. Disruptions in neuroimmune signaling in brain areas involved in social processing may underlie these disruptions in social behaviors.

Gourley SL (*Emory U*) Adolescent social experience shapes adult prefrontal cortex structure and function ABSTRACT: During adolescence, the prefrontal cortex (PFC) undergoes dramatic reorganization as individuals transition toward adult behaviors and cognition. PFC development is profoundly influenced by the social environment, disruptions to which may prime the emergence of psychopathology across the lifespan. Here, we socially isolated mice during adolescence which produced a bias toward inflexible habits and anhedonic-like behavior in adulthood despite normalization of the social environment. This was accompanied by dendritic spine over-abundance and aberrant neuronal hyper-excitability in the ventromedial PFC (vmPFC), which was both necessary for the expression of isolation-induced habits and sufficient to trigger behavioral inflexibility in socially reared controls. Further, adolescent isolation durably enhanced ROCK2-LIMK-cofilin cytoskeletal regulatory pathway signaling, while shRNA-mediated Rock2 silencing in the vmPFC during adolescence, but not in adulthood, blocked the emergence of isolation-induced habits and reward-related deficits alike. Altogether, our findings reveal an adolescent critical period during which social experiences facilitate prefrontal cortical maturation.

Grammer AJ, Zelikowsky M (*U of Utah*) The BNST to NAc: a neural circuit for isolation induced social anxiety. ABSTRACT: Prolonged social isolation has been shown to promote a host of deleterious behaviors including increased social anxiety; however, the neural mechanisms underlying this effect remains unknown. The nucleus accumbens (NAc) is integral to social behavior and has been implicated in social approach and avoidance. Alternatively, the dorsal bed nucleus of the stria terminalis (dBNST) is known for its role in mediating anxiety and social isolation stress. Thus, these regions represent prime candidates for the control of isolation-induced social anxiety. Although the dBNST is known to project to the NAc, this pathway has remained relatively unexplored both anatomically and functionally. Combining retrograde tracing and in situ hybridization (Multiplex Fluorescent RNAscope), we found that a majority of neurons projecting from the dBNST to the NAc are GABAergic and express mRNA for a variety of stress related neuropeptides such as Corticotropin Releasing Hormone (CRH), Tachykinin2 (Tac2), and Oxytocin Receptor (OTR). In addition, using a chemogenetic loss-of-function approach, we found that the NAc is required for social approach in a three-chamber sociability assay. Collectively, these findings support the hypothesis that this pathway allows for the synchronization of social behavior and anxiety. To further investigate more complex social anxiety-like behavior in mice, I developed a new assay termed Selective Access to Unrestricted Social Interaction (SAUSI), in which mice can freely interact with a conspecific while maintaining control over the choice to interact. The SAUSI apparatus consists of two chambers connected by a long, narrow tube. Experimental mice are habituated to the tube and trained to use it as a passageway for access to either chamber. Conspecifics, however, are deterred from using the tube via operant training. Using in-depth hand scoring as well as automated machine-learning approaches, we found that four weeks of social isolation stress increased both approach and escape anticipation, as well as altered complex social behaviors measured during free interaction. Using this assay, I am currently investigating the contribution of specific, genetically-defined dBNST to NAc projection cells in isolation induced social anxiety-like behavior. Collectively, this project reveals the stress-sensitivity of an understudied pathway from the dBNST to NAc and explores a novel role for this circuit in isolation induced social anxiety-like behavior. SUPPORT: NSF GRFP

Guerra DP, Moscarello JM (*Texas A&M U*) The bed nucleus of the stria terminalis mediates the expression of avoidant behavior in male rats ABSTRACT: Avoidant behavior is a unifying symptom of many anxiety disorders. Our study aims to elucidate its underlying neural circuitry. We report on a series of experiments that used a signaled active avoidance (SAA) procedure to model avoidant behavior in male rats. The SAA procedure is composed of two phases, the subject first acquires an association between a tone (CS) and a shock (US) and then learns to perform an avoidance response during the CS to prevent US delivery. As the avoidance response is acquired, the US becomes less frequent and goes from being a certain/imminent threat to a possible/distal threat. Although the bed nucleus of the stria terminalis (BNST) has been shown to mediate defensive responses to possible/distal threats, its role in SAA has yet to be determined. We used a chemogenetic approach to test the hypothesis that BNST is necessary for the expression of the avoidance response. Male rats received intra-BNST infusions of AAV containing the gene construct for either the inhibitory hM4Di DREAD, or GFP. After four weeks of recovery, animals received four days of SAA followed by two additional days of SAA training preceded by counterbalanced IP injections of either the DREADD ligand CNO (clozapine N-oxide) or vehicle. We found that CNO decreased avoidance responses in hM4Di subjects but not in GFP controls, demonstrating that the BNST is necessary for the avoidance response. Because some subjects showed hM4Di expres-

sion in the medial septum (MS), we followed up with an anatomical control experiment specifically targeting this region. However, we found no evidence of a role for MS, indicating that the effects observed in our initial experiment were BNST-specific. We then compared chemogenetic activation (hM3Dq) and inhibition (hM4Di) of BNST in a test of avoidance under extinction conditions. This test consisted of 10 CSs that did not inactivate upon the expression of shuttling and no USs were delivered if the animal failed to avoid. Because stimuli were delivered in an invariant way that was not contingent upon the subjects' response, this design allowed us to test all groups under common conditions. After AAV infusions and recovery, subjects received six days of avoidance training followed by the test under extinction conditions preceded by CNO 24 hours later. We found that CNO decreased avoidance in hM4Di subjects and increased avoidance in hM3Dq subjects relative to GFP and vehicle controls, confirming that BNST is not only necessary for normal levels of avoidance but is also sufficient to potentiate the response. These results demonstrate that BNST, a region known to mediate behavioral defense against low imminence threats, is a crucial substrate of avoidant behavior.

Halcomb CJ, Vanderhoof SO, Mott DD, Jasnow AM (*U of South Carolina School of Medicine*) Basolateral amygdala inputs to the anterior cingulate cortex regulate fear learning that promotes generalized fear ABSTRACT: A common symptom of nearly all anxiety disorders as well as trauma- and stress-related disorders is the overgeneralization of fear. This is characterized by the expression of fear in new or ambiguous environments and can disrupt normative functioning. We previously showed the anterior cingulate cortex (ACC) and its projections to the basolateral amygdala (BLA) control fear responses to novel contexts in which the threat of an aversive event is uncertain. However, the role of the ACC in context fear learning is not well understood. Here, we used pharmacological inactivation, NMDA receptor blockade, and circuit specific manipulations of the ACC to determine its role in context fear learning. Mice were also evaluated for anxiety-like behavior in open field and elevated plus maze to assess associations between fear and anxiety-like behavior. We found pharmacological inactivation or NMDA receptor blockade in the ACC during context fear learning eliminates fear when mice are tested in a novel context but does not alter fear to the training context. We then investigated the role of BLA inputs to the ACC during fear learning to determine circuit mechanisms involved in ACC encoding during context fear learning. We used a chemogenetic, intersectional approach to specifically inactivate BLA-to-ACC projections during context fear learning. Our data show chemogenetic silencing of BLA-to-ACC projections during context fear learning eliminates fear to a novel context but leaves fear to the training context fully intact. These results suggest a critical role for the BLA-ACC circuit is to encode highly salient experiences that shape subsequent behavior to new environments in which threat is uncertain.

Hammack SE, Boucher MN, Aktar M, Moriarty SK, Fontaine NR, May V (*Department of Psychological Science, U of Vermont*) The role of central pituitary adenylate cyclase activating polypeptide (PACAP) signaling in stress and emotion. ABSTRACT: Exposure to stressful stimuli has been argued to play an important role in the etiology of anxiety disorders. Consistent with this role, increases in anxiety-like behavior are often observed in rodents repeatedly exposed to environmental stressors. Several brain nuclei have been implicated in coordinating the autonomic, endocrine and behavioral response to stressor exposure. We have argued that pituitary adenylate cyclase-activating peptide (PACAP) activation and PAC1 receptor signaling in the bed nucleus of the stria terminalis (BNST) mediate many of the consequences of exposure to chronic stress. Moreover, a critical source of afferent PACAP to the BNST originates in the lateral parabrachial nucleus (LPBn), and activation of LPBn PACAP afferents in the BNST enhances anxiety-like behavior, which can be blocked by PACAP receptor antagonism. Here we review this current work along with our prior work implementing molecular and pharmacological approaches to demonstrate that PACAP and PAC1 receptor signaling in several brain regions are critical for many of the consequences of stressor exposure. This work complements the work of several other groups suggesting that central PACAP systems are critical for stress responding and stress-related psychopathology. SUPPORT: Supported by grant MH97988 from the National Institutes of Health

Hart E, Gardner MP, Schoenbaum G (*NIDA IRP*) Anterior cingulate neurons signal neutral cue pairings during sensory preconditioning ABSTRACT: Of all frontocortical subregions, the anterior cingulate cortex (ACC) has perhaps the most overlapping theories of function. Recording studies in rats, humans, and other primates have reported diverse neural responses that support many theories, yet nearly all these studies have in common tasks in which one event reliably predicts another. This leaves open the possibility that ACC represents associative pairing of events, independent of their overt biological significance. Sensory preconditioning provides an opportunity to test this. In the first phase, preconditioning, value-neutral sensory stimuli are paired (A/B). To test whether this was learned, subjects are given standard conditioning during which one of the previously neutral sensory cues is paired with a biologically meaningful outcome (B/outcome). During the final probe test, the neutral cue which was never paired with a biologically meaningful outcome is presented alone (A/) and will elicit a conditional response, suggesting that subjects had learned the associative structure during preconditioning and use that knowledge to infer presentation of the biologically relevant outcome (A/B/outcome). Inference-based responding demon-

strates a fundamental property of model-based reasoning and requires learning of the associations between neutral stimuli before rewards are introduced. ACC neurons developed firing patterns that reflected the learning of sensory associations during preconditioning, even though no rewards were present. The strength of these correlates predicted rats' ability to later mobilize and use that associative information during the probe test. These results demonstrate that clear biological significance is not necessary to produce correlates of learning in ACC. SUPPORT: Fi2 GM 133534 (EH), ZIA DA000587 (GS)

Herrera E[1], Austen JM[2], Urcelay GP[2] (*[1]Department of Psychology, Bournemouth U, UK; [2]School of Psychology, U of Nottingham, UK.*) The effects of goal-landmark distance on overshadowing: a replication in humans of Goodyear & Kamil (2004) ABSTRACT: Spatial cognition has seen a debate between associative theories that predict competition between landmarks and geometry, and modular theories that anticipate absence of competition. Results from numerous labs have documented (or not) said competition, but little is known about the variables that determine whether competition is observed in the spatial domain. It has been suggested that spatial contiguity could be a determinant of competition between events. In these experiments, we aimed to replicate, in humans, findings by Goodyear & Kamil in birds (Clark's nutcrackers). Participants were recruited through Prolific. During training, they had to find a hidden goal using 4 landmarks arranged in the shape of a cross, in the presence of orientation cues. In Group Close, landmarks were placed at 10, 30, 50, and 70 virtual units (VUs) from the goal, whereas in Group Intermediate the distances were 30, 50, 70 and 90 VUs. Finally, in Group Distal landmarks were placed at 50, 70, 90 and 110 VUs. Following 16 training trials, all participants were tested individually with each of the 4 landmarks trained. Of interest was how well participants did when tested with landmarks 50 and 70, which were common across all 3 groups. Consistent with the results in birds, we observed better performance in Group Distal than in Group Close, suggesting that overshadowing was greater in Group Close and thus dependent on the spatial contiguity between landmarks and the goal. Landmarks near the goal overshadow landmarks far from the goal, but the opposite is not true. A second experiment, in which landmarks and orientation cues were misaligned in order to prevent the use of a straightforward solution to the task, showed similar results. The results are discussed in terms of a modification of Pearce's configural model. SUPPORT: Funded by UK ESRC Grant (ES/R011494/2) awarded to GPU and JP

Hoang IB, Munier JJ, Greer Z, Millard SJ, DiFazio LE, Wassum KM, Izquierdo A, Sharpe MJ (*U of California, Los Angeles*) Methamphetamine and midbrain-hypothalamic control of cue-guided behavior ABSTRACT: Through life experiences, we learn to associate certain cues with particular outcomes. These cue-outcome memories help to inform our everyday decisions, such as making dinner plans or picking our next snack at the grocery store. Previously, we have shown that GABAergic neurons in the lateral hypothalamus (LH) are critical for learning about cue reward associations (Sharpe et al., 2017, Current Biology; Sharpe et al., 2021, Nature Neuroscience). Here, we tested the nature of the reward learning that taking place in LH GABA neurons, the wider neural circuits that facilitate this learning in LH, and how drug exposure may impact this process. First, we show that LH encodes sensory-specific (or model-based) associations between cues and rewards. We next demonstrated that these associations are driven by input from dopamine neurons in the ventral tegmental area (VTA). Finally, we found that rats with a history of methamphetamine self-administration showed an enhancement in the control that reward-paired cues have over behavior in a very selective manner, consistent with the nature of learning taking place in hypothalamic-midbrain circuits. Further analyses showed this effect was positively correlated with prior drug intake and a physiological strengthening of hypothalamic-midbrain circuits. Altogether, these data suggest that the LH accumulates sensory-specific cue-reward memories by virtue of dopaminergic input from the VTA and that this process is strengthened with drug exposure to produce enhancements in the control that reward-paired cues have over behavior in substance use disorders. SUPPORT: UCLA Graduate Division (GRM and GSRM) and NIDA 5T32DA024635-14 (Edythe London)

Hodebourg R[1], Meyerink ME[2], Crow AD[1], Reichel CM[1], Kalivas PW[1], Garcia-Keller C[2] (*[1]Department of Neuroscience, Medical U of South Carolina, Charleston, SC, USA. [2]Department of Pharmacology and Toxicology, Medical College of Wisconsin, Milwaukee, Wisconsin, USA*) Cannabinoid use is enhanced by stress and changes conditioned stress responses ABSTRACT: Individuals diagnosed with post-traumatic stress disorder (PTSD) are often comorbid for substance use disorders. Cannabis is widely used by PSTD patients, and the literature is mixed on whether cannabis use ameliorates or exacerbates patient responses to stress associated conditioned stimuli (stress-CS). We determined if cannabis use affects responsivity to stress-CS in rats receiving 2 h stress in the presence of an odor stress-CS. Three weeks after acute stress, rats self-administered cannabinoids (delta9-tetrahydrocannabinol + cannabidiol; THC + CBD) for 15 days, and the stressed males consumed more THC + CBD than sham males. We then used the stress-CS or a novel odor (stress-NS) to reinstate THC + CBD seeking. Surprisingly, the stress-NS reinstated THC + CBD seeking, an effect blocked by N-acetylcysteine. Moreover, the stress-CS inhibited THC + CBD-CS induced reinstatement. To determine if the unexpected effects of stress-NS and -CS resulted from THC + CBD altering conditioned stress, the

effect of THC + CBD use on stress-NS/CS-induced coping behaviors and spine morphology was quantified. In THC + CBD-treated rats, stress-NS increased active coping (burying). Conversely, stress-CS reduced active coping and increased passive coping (immobility) and other behavioral parameters associated with stress responses, including self-grooming and defecation. Transient spine head expansion in nucleus accumbens core is necessary for cue-induced drug seeking, and THC + CBD self-administration prevented the increase in head diameter by stress-CS in control rats. These data show THC + CBD self-administration altered the salience of environmental cues, causing neutral cues to promote active behavior (drug seeking and burying) and stress-CS to switch from active to passive behavior (inhibiting drug seeking and immobilization). We hypothesize that cannabis may exacerbate conditioned stress responses.

Hougham AL, Kim J, Pickens CL, Wang C (*Kansas State U*) Short term withdrawal from repeated fentanyl injections leads to impairment in the expression of goal-directed action in a Pavlovian goal-tracking task ABSTRACT: Previous research has shown that the orbitofrontal cortex is needed at the time of test for proper goal-directed action in Pavlovian goal-tracking devaluation tasks, in which a cue predicts an outcome that has had its value reduced. Other research suggests that orbitofrontal cortex function may be impaired during early withdrawal from repeated opiate exposure. However, to our knowledge, there has been no prior assessment of devaluation behavior during early withdrawal from repeated opiate exposure. Here, we trained rats on a Pavlovian goal-tracking task in which two separate lights predicted different foods, and an anticipatory approach to the site of food delivery prior to food delivery was used as the measure of conditioning. After training was completed, rats received twice daily injections of 10 ug/kg injections of fentanyl or saline (5 hours apart each day). 48 h after the last injection, the rats were sated on one of the pellets and tested for responding to each of the two lights. After the first test, the rats received retraining on each of the two light-food associations and then received a second fentanyl or saline exposure regimen and received testing (with satiation on the other food pellet) 48 h after the end of the second regimen. We found that 48-h withdrawal from fentanyl impaired devaluation in this Pavlovian goal-tracking task. During my poster, I will discuss the significance of these findings and what this may mean for the ability to maintain adequate goal-directed action during early opiate withdrawal. SUPPORT: Partially supported by the Cognitive and Neurobiological Approaches to Plasticity (CNAP) Center of Biomedical Research Excellence (COBRE) of the National Institutes of Health under grant number P20GM113109.

Hu S, George A, Packard K, Song M, Kissi J, Nguyen-Lopez O, Su A, Tesone E, Opendak M (*Kennedy Krieger Institute & Johns Hopkins School of Medicine*) Functional ontogeny and social control over the developing lateral habenula ABSTRACT: Early life attachment to a caregiver sculpts the brain and guides adaptive behaviors. Therefore, trauma during infancy, especially in the form of caregiver-child maltreatment, can create long-lasting consequences to threat processing and social behavior. Recent work has highlighted the mesolimbic dopamine system as a critical interface between early care quality and maternal regulation of the infant, but specific mechanisms remain unclear. Here, we assessed how care quality impacted maternal regulation of core nodes of the mesolimbic dopamine circuit, including the lateral/medial habenula and ventral tegmental area (VTA). To modulate maternal care quality, we employed the Scarcity-Adversity model of low bedding (SAM-LB). Following control or SAM-LB rearing from PN8-12, rat pups underwent odor-shock conditioning with or without maternal presence at postnatal day 18 (PN18, before weaning) and postnatal day 28 (PN28, after weaning). Brains were removed and analyzed with autoradiography. Separately, habenulae were dissected at PN18/PN28 and assayed for levels of CaMKII/*beta* and GABAB receptor. In control-reared pups, we observed that maternal presence suppressed VTA and lateral habenula engagement, as measured through 2-deoxyglucose metabolism. The effect of maternal presence was abolished in adversity-reared pups, with early rearing significantly altering patterns of functional connectivity within this circuit. We observed an increase in CaMKII/*beta* expression, a marker for excitation, and a decrease in GABAB receptor, a marker for inhibition, in PN28 pups compared to PN18 pups, regardless of rearing condition. Together, this suggests that the molecular profile and functional importance of the habenula changes as early development progresses. These findings indicate that lasting social impairments following early adversity may be due to impaired maternal regulation of the mesolimbic dopamine circuit and that the habenula may be a target for early intervention.

Jaiyesimi A, Lohr C, Banerjee A, Waddell J (*U of Maryland, School of Medicine, Baltimore MD*) A single dose of psilocybin facilitates extinction of fear in male rats ABSTRACT: The 5-HT2A receptor agonist psilocybin can induce robust spinogenesis, increase BDNF release and produce long-term benefit in several psychiatric disorders, including cancer-related depression and anxiety, major depressive disorder, nicotine addiction, and alcoholism. Modulation of the serotonergic system can facilitate extinction of fear and render extinction generalizable beyond the extinction context. Our study examined the effect of psilocybin during extinction of fear training as measured by fear-potentiated startle. Male and female adult Sprague Dawley rats received 10 pairings of a light conditioned stimulus (CS) that overlapped and coterminated with a mild foot-shock (US). Acquisition of conditioned fear was determined

by assessment of the fear-potentiated startle reflex. The day following the acquisition test, rats were treated with either psilocybin (2 mg/kg) or saline thirty minutes before the initiation of extinction training. Extinction training consisted of 30 presentations of the light alone, with an average ITI of 3 minutes. Males treated with psilocybin did not express fear to the CS when tested the day after extinction, while saline treated males did exhibit fear. Surprisingly, females did not exhibit fear regardless of their treatment condition. Analysis of parvalbumin cells in the infralimbic cortex found that fewer of these neurons were ensheathed by perineuronal nets 6 hours after extinction. Western blotting found an increase in BNDF at this timepoint, and a decrease in parvalbumin in the same animals in the prefrontal cortices. These effects were not observed in females. Overall, our results suggest that psilocybin might open a critical window of enhanced plasticity similar to that observed by others in male rats after 2 weeks of fluoxetine administration. SUPPORT: This study was supported by Mydecine Innovations, Inc.

Janak PH (*Dept. of Psychological and Brain Sciences; Dept. of Neuroscience; Johns Hopkins U*) Complexities in neural mechanisms for contextual gating of cue-elicited behavior ABSTRACT: Cues can facilitate reward seeking and acquisition, but their ability to do so may vary, depending upon the context in which they are experienced. Context is defined here as the external signal or signals that provide current information regarding reward predictability of a given cue. Here, we use multiple behavioral designs to probe the dependence of contextual gating on hippocampus and related circuitry under conditions in which context-gated cue meaning changes daily, or even trial-by-trial. I will discuss findings of distinct contributions of dorsal and ventral hippocampus to cue-elicited behavior that is gated by contextual cues, as well as evidence for broader roles of ventral hippocampus in hierarchical learning. SUPPORT: NIH/NIAAA and NIH/NIDA

Jin B, DeNardo L (*UCLA*) Brain-wide mapping of amnesic and persistent fear memory circuits throughout development ABSTRACT: Infantile amnesia describes a process wherein episodic memories formed early in life are rapidly forgotten. On the other hand, memories formed in older animals can last a lifetime. Although the neural substrates of memory have been well studied in adults, how memories are stored and retrieved during development remains poorly understood. Here, we performed a brain-wide screen to identify developmental changes in memory networks. We used TRAP2 mice (Targeted Recombination in Active Populations) in combination with brain clearing and light sheet fluorescence microscopy to compare neuronal populations and networks activated by recent (1 day) fear memory retrieval at infant (P17), juvenile (P25) or adult (P60) stages. While adults had more activated neurons in the prefrontal cortex, and anterior thalamic nuclei, juveniles had more activated cells in the hypothalamus and midbrain during fear memory retrieval. Network analyses revealed that the functional organization of memory networks was also developmentally regulated. Adult memory networks included a highly interconnected set of cortical regions. Of these, the retrosplenial cortex (RSP), a key memory center, was absent from memory networks in juvenile animals. To further understand how activity in these regions related to memory retrieval, we examined correlations between TRAPed cell numbers and freezing levels of individual animals. Regions that were highly correlated with freezing differed by age. Notably, RSP was highly salient for freezing at P25, but significantly anti-correlated with freezing at P60. To further examine how RSP memory functions change with age, we looked at the extent to which neurons activated during recent memory retrieval (1d) are reactivated during later memory retrieval (7d) at different ages. Adults had higher reactivation rates in RSP compared to younger groups. Together these data reveal specific changes in the activity of brain regions and circuits that coincide with the developmental transition from amnesic to persistent memories. RSP, which has not been studied in the context of infantile amnesia, may be key for this switch. In ongoing work, we are using in vivo chemogenetic manipulations to identify the how the memory functions of the RSP change across development.

Jo YS, Zweifel LS (*U of Washington*) Dopaminergic circuits underlying threat generalization ABSTRACT: Dopamine neurons of the ventral tegmental area play an important role in the regulation of discriminatory threat learning during Pavlovian conditioning. We have found that dopamine neurons encoded the uncertainty associated with threat generalization through their projections to the central nucleus of the amygdala. In the work presented here, I will discuss unpublished data demonstrating the role of dopamine signaling in regulating the encoding of conditioned stimulus responses in the central nucleus during period of increased uncertainty of unconditioned stimulus presentation. I will also discuss how enhancing dopamine neuron activation with an appetitive conditioned stimulus presentation can both prevent and reverse generalized fear responses. Collectively, this work provides a link between the interactions of appetitive and aversive stimuli at the level of a mesoamygdala circuit.

Kaneko R, Mallea J, Balsam PD (*Columbia U*) Impact of temporal uncertainty about when a CS is presented on sign tracking behavior ABSTRACT: Previous studies demonstrated that sign tracking increases when a conditioned stimulus (CS) signals uncertain outcomes. Variable's reward magnitude or probabilistic reinforcement increase sign tracking (Anselme et al., 2013). We wondered if uncertainty about when a CS would be presented might also enhance sign tracking. We investigated whether variability in the inter-trial interval (ITI) affects the acquisition/extinction sign tracking.

We trained four groups of mice that differed according to the duration of the ITI and whether it was of fixed or variable duration to associate a lever extension CS with a reward. Two groups had 72s ITIs and the remaining two had 144s ITIs. One of the groups at each duration experienced fixed durations and the other group experienced variable durations with the same mean. Each trial consisted of the extension of a lever for 16s followed by 5s access to a drop of evaporated milk. Statistical tests revealed that neither ITI duration nor variability affected goal tracking (head entry). In contrast, groups exposed to a variable ITI showed significantly more lever pressing than those exposed to a fixed ITI. Finally, only ITI variability had a significant effect on sign tracking score [LP-HE/(LP+HE)]). To see if the greater sign tracking induced by ITI variability during acquisition was due to learning or performance, we conducted a test in extinction. Half the subjects from each group were extinguished with either a 108 s fixed or 108s variable ITI duration. As in acquisition, each lever extension lasted for 16s but no reward was presented. Both goal tracking and sign tracking declined during extinction. The variability of the ITI during extinction had no impact on performance. Subjects who were trained with variable ITIs during acquisition continued to show significantly more lever pressing in extinction regardless of whether the testing ITI was fixed or variable. During the extinction test subjects originally trained with a fixed ITI made significantly more head entries than subjects originally trained with variable ITIs. Thus in the extinction test, subjects originally trained with the variable ITI showed more sign tracking while subjects trained with a fixed ITI showed more goal tracking. Our results show that temporal uncertainty about when the CS will appear during acquisition leads to higher levels of sign tracking and this difference is the result of learning that takes place during initial acquisition. Thus temporal uncertainty, like uncertainty about reward, leads to higher lever pressing suggesting that many kinds of uncertainty contribute to the development of sign tracking. Additionally, these effects of initial learning may endure even when environmental conditions change.

Kaplan K, Hunsberger H (*Rosalind Franklin U*) Benzodiazepines impair contextual fear memory ABSTRACT: Benzodiazepines are anxiolytic drugs that are currently prescribed to patients suffering from anxiety and panic disorders. More specifically, these drugs are often prescribed to combat the anxiety associated with post-traumatic stress disorder (PTSD). However, Benzodiazepines are known to interrupt anterograde memory formation or the formation of new memories. Although benzodiazepines have been in use for over 60 years, the affected brain regions, and neuronal mechanisms responsible for this detrimental side-effect are largely unknown. To analyze the effects of Benzodiazepines on long term memory, ArcCreERT2 x eYFP mice were injected with Alprazolam 30 minutes before being exposed to a 3-shock contextual fear conditioning procedure. This novel ArcCreERT2 x eYFP mouse model allows us to tag neuronal ensembles that are active during an experience or memory by using Arc as an immediate early gene (IEG) promotor. We then re-exposed mice to the same context 5 days later and euthanized mice 1 hr after exposure to examine retrieval cell activation using a second IEG, cFOS. This method allows us to determine which brain regions undergo changes after benzodiazepine injection and how these cellular changes relate to freezing behavior (a proxy for memory). We found that male and female mice injected with Alprazolam exhibited increased freezing behavior during training compared to controls. Interestingly, freezing behavior was significantly lower in female treatment mice during re-exposure compared to controls. These results suggest that Alprazolam impairs either the encoding or retrieval of a fear memory. We are currently analyzing hippocampal and amygdala regions to determine whether cells active during encoding or retrieval are altered. Completion of this project will help us to better understand the long-term memory deficits associated with benzodiazepines. We also have the potential to discover novel brain regions involved in these side effects and therefore offer alternative treatment options. Our future studies will examine the impact of chronic use of benzodiazepines on aging and Alzheimer's disease. SUPPORT: Dr. Holly Hunsberger

Keiser AA, Dong T, Kramár EA, Butler C, Matheos DP, Tong L, Berchtold NC, Chen S, Samad M, Magnan C, Beardwood J, Shanur S, Baldi, P, Cotman CW, Wood MA (*U of California, Irvine*) Exercise parameters that open a 'molecular memory window' for cognitive enhancement shine light on key memory mechanism in the adult, aging, and Alzheimer's Disease brain ABSTRACT: The ability to learn, consolidate and retrieve information is critical for everyday survival and this ability begins to decline with normal aging and is severely exacerbated by Alzheimer's Disease (AD). Basic research and clinical trials have universally demonstrated the benefits of exercise for cognitive function including enhancements in long-term memory formation and synaptic plasticity in addition to alleviating cognitive impairments associated with normal aging and AD, however, the mechanism by which exercise leads to cognitive enhancement is unclear. Here, we apply exercise as an approach to unlock a novel understanding of the molecular mechanisms that drive memory consolidation by utilizing specific exercise parameters that enhance cognitive benefits. Employing an intermittent exercise protocol, we demonstrate that 14 days of voluntary wheel-running facilitates hippocampus-dependent memory and synaptic plasticity in adult mice, effects which can be maintained and re-engaged with brief 2-day re-introduction to exercise following a sedentary delay. To identify novel mechanisms that drive memory consolidation, we utilized an unbiased RNA-sequencing approach to uncover genes in the dorsal hippocampus that are differ-

entially expressed under conditions where exercise benefits are maintained throughout sedentary delay periods and enable the formation of long-term memory and synaptic plasticity. We identify a gene coding for a type 1 receptor for the TGF-/*beta* family of signaling molecules, Acvr1c as one of few genes (including Bdnf) showing up-regulation in the hippocampus under exercise conditions that enable the formation of long-term memory and synaptic plasticity. Utilizing viral manipulations in the adult hippocampus to disrupt and over-express Acvr1c, we identify Acvr1c as a key bi-directional regulator and driver of hippocampus-dependent long-term memory and synaptic plasticity. We find Acvr1c levels decrease in the hippocampus with age in C57Bl/6J and 5xFAD female and male mice and demonstrate that Acvr1c over-expression ameliorates age and AD-associated impairments in memory and synaptic plasticity. These data suggest that promoting ACVR1C through exercise or pharmacological intervention may protect against age and AD-associated cognitive impairment, providing a potentially powerful and novel disease modifying treatment strategy for AD. SUPPORT: This work was funded by the US National Institute on Aging (K99 AG078501 to AAK, R01 AG051807 to MAW and CWC, R21 AG067613 to MAW and AO and F32 AG071209 to AAK), the US National Institute on Drug Abuse (R01 DA047981 to MKL and MAW and R01 DA047441 to MAW and GSL), the US National Institute of General Medical Sciences (R01 GM123558 to PB).

Khoo SY, Uhrig A, Samaha AN, Chaudhri N (*Concordia U/U of Montreal*) Pavlovian conditioned approach after extended training: What's influencing sign- and goal-tracking? ABSTRACT: A key question in the Chaudhri laboratory was the factors that underlie changes in incentive salience attribution after extended training. Dopamine is known to be involved in inventive salience, where conditioned cues acquire motivational value. However, the role of dopamine might change with training. We studied dopamine D1- and D2-like receptor antagonists on the expression of sign- and goal-tracking conditioned responses after extended Pavlovian conditioned approach training. We also tested if amphetamine-induced psychomotor sensitization and vendor breeding colony influence acquisition of sign-tracking after extended training. In experiment 1, male Long-Evans rats received 20 PCA sessions in which one lever (CS+, 10 s) predicted 0.2 ml sucrose (10 %, w/v) delivery and the other lever (CS-) did not. SCH-23390 (D1-like antagonist) or eticlopride (D2-like antagonist) were administered before non-reinforced behavioural tests at doses of 0, 0.01, and 0.1 mg/kg (s.c.). We found that both doses of SCH-23390 reduced sign- and goal-tracking, but also reduced locomotor behaviour. A low dose of eticlopride (0.01 mg/kg) selectively reduced goal-tracking, without affecting sign-tracking or locomotor behaviour. In experiment 2, rats received vehicle or 2 mg/kg amphetamine (i.p.) for 7 days (n = 12/group). Ten days later, they received 16 PCA training sessions. Although amphetamine produced psychomotor sensitization, this did not affect the acquisition of sign- or goal-tracking. In experiment 3, 40 rats from two breeding colonies (K72 n = 20, R06 n = 20) were trained in a single lever design (Paired n = 15; Unpaired = 5) for 11 days. Here, K72 rats had higher Pavlovian Conditioned Approach scores than R06 rats. Together, these results show that D2-like receptors and the genetic and environmental factors associated with different vendor facilities affect incentive salience, while amphetamine-induced psychomotor sensitization did not. SUPPORT: This research was supported by grants from the Canadian Institutes of Health Research (CIHR; MOP-137030; NC), the Natural Sciences and Engineering Research Council (NSERC; RGPIN-2017-04802, NC), Fonds de la recherche du Québec – Santé (Chercheur-Boursier, NC) and Concordia U (Center for Studies in Behavioral Neurobiology, Nadia Chaudhri). SYK was supported by a Concordia Horizon Postdoctoral Fellowship and a postdoctoral fellowship from the Fonds de la recherche du Québec – Santé (FRQS; 270051 and 306413). AU was supported by a Concordia Undergraduate Student Research Award. ANS was supported by a salary grant from the FRQS (Award ID: 28988).

Knowlton, BJ, Franco, CY (*UCLA*) Substance use correlates with habitual responding in a probabilistic classification task ABSTRACT: The transition from action to habit may be a fundamental component of the development of substance abuse disorder. Here, we used a laboratory measure of human habit learning to examine its relationship with reported substance use. Undergraduate participants (N=96) performed a probabilistic classification task (the Weather Prediction Task) in which combinations of cues are probabilistically associated with one of two outcomes (sun or rain). On each trial, participants received feedback as to whether their response was correct or not. This task can be learned in a declarative, model-based manner, or in a more habitual, stimulus-response manner. After 100 trials, the probabilities were reversed, and we conceptualized habitual behavior as perseverating responses based on the old probabilities. While initial performance did not correlate with measures of reported substance use, we found a significant correlation between habitual behavior and both reported drug use (r=.25) and alcohol use (r=.34) that was robust to removal of outliers and low performers on initial learning. These results suggest that habitual behavior, resulting in a reduced ability to flexibly modify associations as measured in a laboratory task, may be related to real-world health behaviors. Furthermore, multiple regression analysis controlling for measures of subjective socio-economic status and symptoms of depression and anxiety identified reported childhood emotional neglect as a predictor of increased habitual behavior, consistent with findings that early life stress, particularly a

history of neglect, may predispose individuals to increased habitual responding. These results support the idea that an increase in habit behavior may contribute to the well-known link between early life stress and substance abuse in adulthood. SUPPORT: NIH/NIDA 5R01DA045716-03 (Knowlton, PI), NSF Graduate Research Fellowship (Franco)

Kurtoglu B[1], Estes M[1], Laskowski L[1], Windsor BW[1], Davidson R[1], Van Newenhizen E[1], Okunseri T[2], Mantych M[3], Spring M[4], Wheeler DS[3], Wheeler RA[3], Hearing MC[3], Mantsch JR[1] (*[1]Medical College of Wisconsin; [2]Yale U; [3]Marquette U; [4]Dartmouth College*) Effects of chronic stress and corticosterone on Pavlovian conditioned approach behavior and prefrontal cortical function in rats ABSTRACT: Pavlovian conditioned approach behavior in rats can be studied using an autoshaping protocol in which the presentation of a CS+ (a retractable lever presented for 10 sec) immediately precedes the delivery of a sucrose pellet into a food receptacle. When the CS+ is presented in this design, rats display sign tracking (i.e., behavior directed at the CS+ indicated by lever pressing) or goal tracking (i.e., behavior directed at the food receptacle) prior to food delivery. This phenomenon makes autoshaping useful for studying the neural pathways that regulate motivated approach behavior, as some experiences may alter approach in a way that aligns with pathology. For example, we previously found that chronic unpredictable stress (CUS) reduces CS+ directed behavior and impairs the associated activity of a pathway from the prelimbic prefrontal cortex (PL-PFC) to the nucleus accumbens (NAc) core. As previously reported, 14 days of CUS reduced sign-tracking (CS+ directed) behavior when measured several days after the last stressor, an effect associated with increased head entry (goal-directed) behavior. By contrast, when measured during a period of ongoing CUS (days 10-21), CUS reduced CS+ directed behavior without altering head entries. Whole cell recording of retrobead-label layer 5/6 PL-PFC pyramidal neurons (PNs) projecting to the NAc core following CUS demonstrated reduced mEPSC frequency and amplitude and increased mIPSC frequency and amplitude compared to controls. This stress effect was mimicked by chronic corticosterone (CORT) treatment. Implantation of a CORT pellet reduced CS+ directed behavior without affecting head entries. Preliminary data indicate that the same treatment reduced spine density in di-OIistically labeled PL-PFC PNs, while CORT delivery in the drinking water produced CUS-like effects on mEPSCs and mIPSCs in these neurons. It has been previously reported in mice that CUS produces CORT-dependent dysfunction of the PFC via increased expression of REDD1 (regulated in development and DNA damage responses-1; aka DDIT4, RTP801, Dig2) and disruption of mTORC1 (mechanistic target of rapamycin complex 1) signaling. Moreover, REDD1 is increased post-mortem in the PFC of individuals diagnosed with major depressive disorder. As in mice, we find CUS produces time-dependent increases in PL-PFC REDD1 expression and disrupts mTORC1 as demonstrated by reduced phosphorylation of Raptor, a key component of the TORC1 complex. However, viral overexpression of REDD1 in PL-PFC PNs failed to reproduce the effects of CUS or CORT on autoshaping, resulting in heightened CS+ directed behavior. Finally, we observed sex differences in the effects of CUS on autoshaping behavior. While males and females showed similar reductions in CS+ directed behavior after CUS, in contrast to males, females do not show increases in head entries (i.e., goal-directed behavior). Overall, these findings suggest that stress, via CORT, alters PL-PFC function and motivated behavior in a sex-dependent manner and have relevance for understanding stress-related disorders such as depression. SUPPORT: Charles E. Kubly Mental Health Research Center

Kutlu MG, Tat J, Zachry J, Calipari ES (*Vanderbilt U*) Accumbal dopamine response to expected aversive outcomes mediates the expression of conditioned behavior ABSTRACT: Dopamine release in the nucleus accumbens (NAc) has been causally linked to adaptive aversive learning and its dysregulation is a core phenotypic characteristic of anxiety and stress disorders. Thus, understanding the role of dopamine in aversive learning is important for both understanding neuromodulatory signaling in the brain as well as how its dysregulation is important in psychiatric disease states. Using optical approaches to directly record and manipulate dopamine release in the NAc in awake and behaving animals, we show that dopamine responses evoked at the time of omitted aversive outcomes are causal to the expression of conditioned responses. The magnitude of the dopamine response at the time of an omitted footshock scaled positively with the prediction of the aversive outcome on that trial; however, this effect was only apparent at the time of the omitted outcome, but not in response to the predictive cue itself. Importantly, we show, via optogenetic manipulation of this signal, that dopamine in this context is not transmitting an error-based signal, as enhancing this signal disrupted learning rather than enhancing it. Finally, using the KCS model, we showed that these effects can be explained by dopamine signaling the perceived saliency of predicted aversive events, but not prediction errors.Together, we show that NAc core dopamine responses to expected but omitted aversive stimuli causally determine associative learning for aversive stimuli in mice. These results add to the growing literature supporting the dopaminergic encoding of perceived saliency by dopamine in the NAc. Our conclusions regarding the dopaminergic information encoding in associative learning also have important clinical implications for anxiety and stress disorders. In sum, these results have far-reaching implications for the theory of learning and memory, the understanding of the mesolimbic neurocircuitry, and the psychopathology of anxiety and stress disorders.

Lamb JH, Furtak SC (*California State U, Sacramento*) Chemogenetic silencing of the perirhinal cortex attenuates fear extinction learning to a discontinuous visual conditioned stimulus. ABSTRACT: The perirhinal cortex (PER) is an important structure in memory processing and formation. Over the past few decades, it has been debated whether the PER plays a role in perceptual processing, mnemonic function, or both. One theory posits that the PER supports stimulus unitization, a process in which the PER collates stimulus characteristics across modalities, time or features into a single stimulus representation, and functions to incorporate and update emotional or motivational attributes of the unitized stimulus. Several lines of research manipulating perirhinal function during Pavlovian fear conditioning have supported this theory. Here, in the first study to chemogenetically silence the PER during fear learning, we use designer receptors exclusively activated by designer drugs (DREADDs) to suppress cell activity during extinction training. Sprague-Dawley rats were handled for 5 days and then underwent surgery injecting the PER bilaterally with pAAV8-CaMKIIa-hM4D(Gi)-mCherry DREADD from a lateral approach. Three weeks following surgery, rats were put through a Pavlovian fear extinction paradigm. The first day, rats were habituated to the context used throughout the paradigm. On day two, rats were fear conditioned using 5 paired presentations of a brief foot shock unconditioned stimulus (US) and a discontinuous light conditioned stimulus (CS). On the third day, half of animals received intraperitoneal injections of clozapine N-oxide (CNO), while the other half received injections of the dimethyl sulfoxide (DMSO) vehicle 45 mins prior to fear extinction training. Extinction training consisted of 20 CS-alone presentations. On the final day of the paradigm, animals were returned to context for extinction retrieval and received an additional 15 CS-alone presentations. Results demonstrate an attenuated extinction response within the CNO group compared to the DMSO group indicated by elevated freezing levels in the CNO group during fear extinction, when the PER was chemogenetically silenced. In addition, the following day during extinction retrieval, the CNO group continued to display higher levels of freezing compared to the DMSO group, indicating an impairment of extinction memory. These results further support the involvement of the PER in fear extinction to discontinuous CS, and the stimulus unitization hypothesis. SUPPORT: NSF Grant IOS 175111

Laughlin LC, Samels SB, Moloney DM, Andrade E, Sears RM, Cain CK (*Child & Adolescent Psychiatry at NYU, Emotional Brain Institute at NKI*) Counterconditioning of response-produced safety signals is highly context-dependent in female rats ABSTRACT: In signaled active avoidance (SigAA), rats are presented with a warning stimulus (WS; e.g. sound) that predicts an aversive unconditioned stimulus (US; e.g. footshock). Performance of the avoidance response (AR) terminates the WS, prevents the US, and produces new feedback stimuli. Failure to perform the AR results in WS-US pairings, transforming the WS into a conditioned threat. The reinforcement mechanism in SigAA is unknown, though prominent models suggest negative reinforcement is key (WS-termination or US-omission). Another possibility is that positive reinforcement contributes to instrumental avoidance, via response-produced stimuli that become safety signals due to negative correlations with the WS and US. To examine this, we designed a 2-way shuttlebox SigAA task with an explicit feedback stimulus (5s tone) and an outcome-devaluation procedure involving Pavlovian counterconditioning of the feedback-tone in separate chambers (40 tones & shocks, paired or unpaired). Final tests involved 15 WS presentations in the shuttleboxes (no WS-termination, shock or feedback tones). In male rats, avoidance behavior after moderate training (5 sessions) was strongly suppressed by devaluation and freezing to the WS reemerged. These effects were absent after 20 sessions of training, consistent with appetitive studies showing that overtraining results in habitual behavior that is no longer dependent on outcome value. Subsequent tests confirmed that devaluation was effective in male rats, who displayed high freezing to the feedback-tone in multiple contexts (counterconditioning boxes, shuttleboxes, and a novel open field). A different profile was observed in female rats using identical procedures. Devaluation effects were absent after both moderate training and overtraining. Females showed high freezing to the feedback-tone in the counterconditioning boxes, confirming that devaluation was effective. However, very low freezing to the same cue was observed in the shuttleboxes and open field, suggesting that expression of this learning is context-dependent. To circumvent this context-dependence of devaluation, counterconditioning was next conducted in the shuttleboxes with the door blocked, followed by avoidance testing with the door open (females only). Devaluation effects were again absent. Subsequent tests in the shuttleboxes revealed high freezing to the devalued tone only when the door was blocked. In the final experiments, avoidance behavior collapsed when training, devaluation and testing all occurred in the shuttleboxes with the door open. Taken together, these data suggest that response-produced safety signals contribute to positive reinforcement of goal-directed avoidance in male rats. The unusually strong context-dependence of counterconditioning in females means that other methods like contingency degradation may be necessary to explore mechanisms of goal-directed vs. habitual avoidance in females. This novel context effect may also help explain why anxiety disorders characterized by avoidance are more prevalent in females: devaluation of an avoidance outcome may not weaken goal-directed avoidance in other contexts. SUPPORT: R01MH114931 to CKC

Le QE, Hereford D, Borkar CD, Fadok JP (*Tulane U*) Dynamics of defensive behavior using Pavlovian fear conditioning with a serial compound stimulus ABSTRACT: Defensive responses are varied and complex, and action selection is contingent upon threat intensity, proximity, and context of exposure. Based on these factors, we developed a Pavlovian fear conditioning paradigm that elicits clear transitions between conditioned freezing and flight behavior within individual subjects. This model employs higher intensity footshocks and a greater number of pairings between the conditioned and unconditioned stimulus. Additionally, this paradigm utilizes serial presentation of pure tone and white noise (WN) auditory stimuli as the conditioned stimulus. Following conditioning, mice exhibit freezing behavior in response to the tone stimulus, and flight responses (darting, jumping) during the WN stimulus. Our goal with this paradigm is to better understand how neuronal circuits control freezing and flight and to test for potential sex differences in the expression of complex behavioral responses to threat. Here, we use markerless pose estimation together with supervised behavioral classifiers to analyze the details of defensive ethograms of male and female C57Bl/6J mice in the standard paradigm (paired group) versus two control conditions (unpaired and shock-only). We also report behavioral changes that occur over two extended extinction sessions. We analyze freezing, flight, jumps, speed , darting, and tail rattling behaviors to assess changes in each group's ethogram. During the second day of conditioning, the paired group displayed greater freezing to tone compared to the unpaired group, and paired mice displayed characteristic jumping and darting behavior to WN in patterns unseen in unpaired or shock-only mice. During extinction, paired mice displayed higher freezing to both tone and WN compared to unpaired and shock-only mice. While both paired and unpaired mice displayed reductions in tone freezing over the extinction session, only paired mice showed reduced flight and increased freezing to WN from early to late extinction. Changes in freezing to tone or WN were not seen in shock-only mice across extinction. Within early extinction, only paired mice displayed significant jumping behavior to WN. Further in-depth analysis of behaviors, including speed changes and tail rattling, across the entire paradigm is in progress. Overall, our results elucidate distinct behavioral response profiles to threat cues and context based on conditioning parameters. SUPPORT: This work was supported by the Louisiana Board of Regents through the Board of Regents Support Fund (LEQSF(2018-21)-RD-A-17) and the National Institute of Mental Health of the National Institutes of Health under award number R01MH122561

Lee J, Hanif S, Aubry A, Burghardt NS (*Psychology Program, Behavioral and Cognitive Neuroscience, The Graduate Center, CUNY, New York, NY*) The contribution of associative learning to stress-induced social avoidance behavior ABSTRACT: Chronic social defeat stress (CSDS) leads to social avoidance, which is often interpreted as depressive-like behavior. This is supported by the finding that stressed C57/BL6J mice avoid CD-1 mice, the strain used to induce CSDS, and mice of the same strain (C57/BL6J). However, we find that stressed 129Sv/Ev mice only avoid CD-1 mice and not a conspecific, indicating that motivation to socialize is intact. We hypothesize that in 129Sv/Ev mice, fear conditioning occurs during each social defeat session such that the CD-1 serves as a conditioned stimulus and the CD-1 attack serves as an unconditioned stimulus, leading to the formation of a fear memory that is retrieved during the social interaction test. If true, then social avoidance should be subject to extinction and second-order fear conditioning, both of which are commonly tested following traditional models of Pavlovian fear conditioning. We tested extinction of social avoidance by exposing defeated mice to 5 social interaction tests/day for 7 consecutive days. A novel CD-1 or 129Sv/Ev mouse was used as the social target for each test. Consistent with extinction learning, we found that interaction time with the CD-1 significantly increased across experimental days (n=10; Day 1 vs. Day 7, $p< 0.01$). In contrast, interaction time remained high throughout testing when the social target was a 129Sv/Ev mouse (n=9; Day 1 vs. Day 7, $p= 0.62$). In a follow-up experiment, we found that defeated mice avoid a Swiss Webster mouse, but not the scent of a CD-1 (n=20; $p< 0.05$), indicating that threat was associated with specific visual cues (color) during CSDS. We next tested whether pairing an aggressive CD-1 with a tone (4Khz, 70dB) produces second-order conditioning. On each day of defeat, the last 10 seconds of a tone overlapped with the introduction of a CD-1. At test, the same tone was played when mice entered the interaction zone of an empty wire-mesh enclosure. Defeated mice (n=14) spent significantly less time with the tone-paired cup than non-defeated controls (n=16), indicating that CSDS can elicit avoidance behavior in the absence of a social target ($p< 0.05$). When the social target is present, avoidance of a CD-1 upregulated c-Fos in the basolateral nucleus of the amygdala (defeated n=5, non-defeated n=5; $p< 0.001$), consistent with the known role of this region in retrieval of a conditioned fear memory. Collectively, these results indicate that CSDS may be useful for studying the formation of fear memories involving a social cue. SUPPORT: R21MH114182 and the PSC-CUNY Awards program

Lerner TN (*Northwestern Feinberg School of Medicine*) Dopamine circuit for habit formation ABSTRACT: The basal ganglia operate largely in closed parallel loops, including an associative circuit for goal-directed behavior originating from the dorsomedial striatum (DMS) and a somatosensory circuit important for habit formation originating from the dorsolateral striatum (DLS). An exception to this parallel circuit organization has been proposed to explain how infor-

mation might be transferred from DMS to DLS during habit formation. The "ascending spiral hypothesis" proposes that DMS disinhibits dopamine signaling in DLS through a trisynaptic, open-loop striatonigrostriatal circuit. My lab has recently used transsynaptic and intersectional genetic tools to investigate both closed- and open-loop striatonigrostriatal circuits. We found strong evidence for closed loops, which would allow striatal subregions to regulate their own dopamine release. We also found evidence for functional synapses in open loops. However, these synapses are unable to modulate tonic dopamine neuron firing, questioning the prominence of their role in mediating crosstalk between striatal subregions. I will discuss our recent findings as well as efforts to elucidate the function of open loops, including the ascending spiral, and to reformulate the ascending spiral hypothesis in light of new evidence.

Liu S, Olsen C (*Medical College of Wisconsin*) Investigation of the necessity and specificity of the dmPFC cocaine seeking ensemble ABSTRACT: Cocaine use disorder is a chronic and relapsing neuropsychiatric disorder characterized by a strong propensity for relapse upon re-exposure to a previously cocaine-associated environment. The dorsal medial prefrontal cortex (dmPFC) is a critical node in the mesocorticolimbic system related to cue-induced cocaine craving and seeking. There is evidence that learned associations between cues and drug seeking behavior are encoded by specific ensemble of neurons sparsely scattered throughout the dmPFC. Thus, we explored the necessity and specificity of the cocaine seeking ensemble in the dmPFC and hypothesized that inhibition of dmPFC cocaine seeking ensembles inhibits cocaine seeking memory retrieval, and these ensembles are not involved in recall of conditioned fear which is also mediated by the dmPFC. We tested this hypothesis by co-injection of viruses expressing TRE-Cre and a cre-dependent inhibitory PSAM-GlyR into the dmPFC of male and female mice to enable "tagging" of ensemble neurons with an inhibitory chemogenetic receptor. After stereotaxic and jugular catheterization surgery, mice were trained to self-administer cocaine (0.5 mg/kg) for 14 days. After 7 days forced abstinence, a 2-hour drug seeking session was performed and the ensemble was tagged. After another 14 days abstinence, mice received ligand (uPSEM792s) 30 minutes before the second drug seeking session for activation of the chemogenetic receptors. 3 days after the second seeking session, mice received tone-shock associative learning in the fear conditioning training, and a context test and cued test were performed 24 hours after the training. The uPSEM792s ligand was also given 30 minutes before each test. We compared these two seeking sessions and found that chemogenetic inhibition of the dmPFC cocaine seeking ensemble suppressed cocaine seeking. We also quantified the freezing effects during the fear conditioning tests and found that suppression of the cocaine seeking ensemble did not affect fear memory retrieval. These results indicated that the dmPFC cocaine seeking ensemble is necessary for context- and cue-induced seeking memory retrieval, but these ensemble neurons are only specific to cocaine seeking, no effect on fear conditioning memory retrieval. SUPPORT: NIH R01 Grant DA042792

López-Moraga A, Luyten L, Beckers T (*KU Leuven, Leuven Brain Institute*) Sex differences in generalization of an auditory fear memory ABSTRACT: Pervasive avoidance is a core symptom of anxiety-related disorders. Investigating its interaction with or causal influence on other fear learning mechanisms involved in the development of maladaptive fear is important for improving our understanding of the basis of anxiety disorders. It has been reported that the higher the intensity of the unconditional stimulus (US) or the higher an individual's state anxiety during fear learning, the more conditioned fear will generalize. Avoidance responses limit the confrontation with a US and, thus, hinder habituation to it. Therefore, avoidance could increase the aversiveness of the US. We hypothesize that avoidance may thus amplify later generalization of the fear memory. Using a platform-mediated avoidance procedure, we assessed the effect of avoidance on generalization of conditioned fear responding in male and female rats. Rats (n=36) were divided into avoiders, non-avoiders and yoked-avoider animals. Avoiders had the possibility to avoid the tone(CS)-signaled US by stepping onto a platform, while yoked-avoiders (without a platform) received the same tone-shock pairings as the avoider they were yoked to. Non-avoiders had no platform to avoid the shocks and received all tone-shock pairings. Animals went through three days of avoidance training followed by two days of generalization testing during which the platform was absent and two new tones of increasing dissimilarity to the CS (GS1 and GS2) were presented. Next, a generalization session was conducted with the platform present and two new generalization tones (GS3 and GS4). We found significant sex differences during the generalization session in the discrimination index of freezing between CS and GS1 ($p=0.02$) and a significant sex by tone interaction in suppression of bar pressing ($p=0.01$). Finally, during the generalization session with platform we found a sex by group by tone interaction for avoidance ($p<0.01$). These results illustrate the potential effect of avoidance on subsequent generalization responses and suggest sex differences in generalization of an auditory fear memory. SUPPORT: We acknowledge the support of KU Leuven Research Grant 3H190245 and FWO PhD fellowship 11K3821N.

Lopez-Rojas J, Lofaro O, de Solis CA, Leroy F, Kandel ER, Siegelbaum SA (*UWM, Columbia U*) Lateral entorhinal cortical input to the hippocampus: hearing the news from a faraway place ABSTRACT: Identifying and adequately reacting to social signals is critical for survival. Yet, social cognition is compromised in neuropsychiatric and neurodegen-

erative disorders. The hippocampus is essential for forming and storing declarative memories, including social memory, the ability of an animal to recognize and remember a conspecific. To serve this mnemonic function, the hippocampus relies on a variety of external inputs. Despite recent advances in our understanding of the brain circuits supporting social memory, little is known about the role of the entorhinal cortex, which provides the primary source of multimodal sensory input from neocortex to hippocampus. Our data show that crucial multisensory information from lateral entorhinal cortex (LEC) reaches dorsal CA2 during social exploration and silencing the lateral entorhinal input to dorsal CA2 impairs social recognition memory. Moreover, optical recordings of neural activity in awake behaving animals show that LEC LII neurons contain sufficient information to decode a conspecific's identity and degree of familiarity. These experiments contribute to our understanding of the network mechanisms at the core of social recognition memory and could open the door to therapeutic interventions in schizophrenia and other brain disorders.

Machado GDB, Fleischer AW, Schnitzler AL, Frick KM (*Department of Psychology, U of Wisconsin-Milwaukee, WI, USA*) G-coupled protein estrogen receptor (GPER) agonism in the dorsal hippocampus enhances memory consolidation in gonadectomized male mice ABSTRACT: Estrogens are cholesterol-derived hormones that play crucial physiological and pathological roles in both sexes and across the lifespan. Many research groups have replicated the beneficial roles of estradiol (E2), the most potent estrogen, in memory consolidation in the past decades, even though some mechanisms are still unclear. The rapid effects of E2 in memory formation are attributed to its binding to different estrogen receptors (ER), notably the intracellular receptors ER-alfa and ER-beta, as well as the membrane ER called G protein-coupled estrogen receptor (GPER). Previous work from our laboratory demonstrated that acute post-training infusion of E2 into the dorsal hippocampus (DH) of ovariectomized female mice enhances object recognition and spatial memory consolidation via activation of ER-alfa and ER-beta, and downstream ERK signaling (Boulware et al., 2013). Although E2 has similarly beneficial effects on memory consolidation in male mice, these effects do not depend on ERK signaling (Koss et al., 2018), suggesting sex differences in the molecular mechanisms through which E2 consolidates object memories. We have also shown that post-training DH infusion of the GPER agonist G-1 enhanced memory consolidation in ovariectomized female mice in a manner dependent on JNK/ATF2 signaling and actin polymerization (Kim et al., 2016, 2019). However, we have not yet investigated whether GPER activation in the DH will similarly influence memory consolidation in males. Thus, the present study examined effects of bilateral DH G-1 infusion on memory consolidation in gonadectomized male mice. Our preliminary data show that immediate post-training bilateral DH infusion of G-1 enhances consolidation in object placement and object recognition tasks, as previously demonstrated in female mice. Ongoing work will evaluate molecular pathways activated by GPER agonism in male mice. These data will provide important new insights about the role of GPER in modulating memory consolidation and into possible sex differences in the molecular mechanisms through which GPER regulates memory. Thus, this work may open new avenues for sex-specific treatment of memory-related disorders. SUPPORT: Supported by R01MH107886 to KMF, a Dr. Robert Cialdini and Bobette Gordon Graduate Fellowship to GDBM, a Distinguished Graduate Student Fellowship to AWF, and a Support for Undergraduate Research Fellowship (SURF) to ALS.

Magalhaes G, Burnell H, Subedi S, Jabri S, Ganiyu O, Meyer H (*Boston U*) Sex differences in safety learning are present in adults but not adolescents ABSTRACT: Difficulty discriminating between threat and safety cues is a hallmark symptom of many anxiety disorders. The risk is greater for females, who are twice as likely as males to develop anxiety disorders following trauma, and adolescents, with diagnoses of anxiety disorders peaking around 13 years. While growing evidence suggests that sex differences in fear responding are apparent in adults, it remains unclear when these differences emerge throughout development and what behavioral factors may drive them. Using adult (postnatal day/PND 70) and adolescent (PND 30) mice of both sexes, we set out to study age and sex differences in discriminative learning using a series of safety cues and fear cues. Following this period, animals underwent a test session including a summation (fear cue paired with safety cue) along with the introduction of a novel cue presented alone or in compound with the fear cue to assess fear inhibition and fear generalization, respectively. Our results revealed that mice of both ages and both sexes froze more during fear cues than safety cues, though adult male mice showed greater discrimination between fear and safety cues than adult female mice. In contrast, no significant sex differences were observed during adolescence. A summation test revealed that while all mice show fear inhibition specific to the safety cue, the magnitude of inhibition is greater within adult males than adult females, again with no sex differences during adolescence. Both sexes of adolescent and adult mice displayed strong fear generalization to a novel cue. Our data indicate that sex differences in safety learning emerge during adulthood and that while male and female mice perform similarly during discrimination learning, females show deficits in discriminating between safety and fear cues. Ongoing data collection will consider immediate early gene expression alongside fear or safety in both ages and sexes. Together, these findings illustrate the potential to improve clinical approaches to anxiety disorders by informing how treatments could be altered. While simi-

lar treatments may work for both sexes during adolescence, treatments for adults may potentially benefit from being tailored by sex. SUPPORT: NIMH R00MH119320

McKinney A, Davis I, Pickens C (*Kansas State U*) Adolescent exposure to amphetamine did not impair later devaluation in adulthood in tasks that do or do not allow for compensation between strategies ABSTRACT: Many previous studies have shown that exposure to psychostimulants can lead to impaired devaluation in laboratory animals, but the evidence on devaluation impairments in human drug users is mixed. While there are several possible explanations for this mixed pattern in humans, we conducted a study to examine one possible explanation: compensation through the use of alternative strategies to maintain goal-directed action. Most laboratory studies of devaluation in rodents allow for only one strategy to guide goal-directed action, either cue-outcome associations (usually using cuelights) or response-outcome associations (usually using levers in a specific location). While this is useful for isolating the neural substrates of one strategy, it prevents animals from compensation for the loss of one strategy by switching to a different strategy. We used a task in which responses to two specific lever-light compounds earned different luation in laboratory animals, but the evidence on devaluation impairments in human drug users is mixed. While there are several possible explanations for this mixed pattern in humans, we conducted a study to examine one possible explanation: compensation through the use of alternative strategies to maintain goal-directed action. Most laboratory studies of devaluation in rodents allow for only one strategy to guide goal-directed action, either cue-outcome associations (usually using cuelights) or response-outcome associations (usually using levers in a specific location). While this is useful for isolating the neural substrates of one strategy, it prevents animals from compensation for the loss of one strategy by switching to a different strategy. We used a task in which responses to two specific lever-light compounds earned different outcomes during training. After training, rats were sated on one of the pellets before a devaluation test in which the lever-light compounds were congruent with those in training (so rats could compensate for loss of one strategy by switching to the other strategy) or incongruent (so compensation was prevented). We gave male and female rats repeated exposure to amphetamine during adolescence using a regimen previously shown to lead to altered function of the prelimbic cortex during adulthood (3 mg/kg injections every other day from post-natal day 27-45), as prelimbic cortex is putatively a critical brain area for the response-outcome strategy used in many devaluation tasks. To our surprise, this regimen did not lead to impaired devaluation in either version of the test. During my poster, I will discuss possible causes of this null effect.

Mehrzad Z, Cherukupalli C, Prasad S, Campese VD (*Department of Psychology & Behavioral Sciences, U of Evansville, Evansville IN, 47722*) Sexually dimorphic effects of reinforcement schedules on active avoidance ABSTRACT: Prior work has shown that like extinction, signaled active avoidance (AA) learning is also contextually specific. On the other hand, probing memory for avoidance using a transfer test approach demonstrates that even when AA responding is not available outside of the avoidance context, freezing does not renew. To expand the scope of analyses comparing reductions in conditioned freezing brought about by either extinction or avoidance to one another, two separate studies using rats were run. In the first study, the effects of various extinction approaches on established signaled AA behavior were measured. In the other, an incrementing reinforcement schedule approach to AA training was explored for the purpose of transfer testing. The findings from these studies suggest that while AA may be sensitive to extinction manipulations, interpretations of these results are obscured by the confounding factors inherent in standard avoidance protocols. In contrast, using a traditional reinforcement approach to AA and conducting transfer tests with previously trained stimuli may offer a more systematic way of evaluating contingency effects on AA generalization across contexts in future studies. For instance, the study using this approach found that male and female rats performed at different asymptotic levels of AA and that the transition between reinforcement schedules negatively impacted males more than females. Using transfer tests showed that moving from a continuous reinforcement schedule to a fixed ratio 2 also impaired performance in male, but not female subjects. Given the stimulus exposure confounds in signaled avoidance, such conclusions are more difficult to draw.

Mondello JE, Chang CW, Trott JM, Anaya A, Solorio S, Tran L, Fanselow MS (*UCLA*) Stress Enhanced Fear Learning enhances excitatory synaptic transmission in Basolateral Amygdala neurons ABSTRACT: Post-traumatic stress disorder (PTSD) is marked by maladaptive learning processes that lead to inappropriately exaggerated fear responses following a traumatic experience. In order to recapitulate some of the symptomology of PTSD, our laboratory developed a rodent stress model, termed Stress Enhanced Fear Learning. In this stress model, a single traumatic stressor (10 footshocks across 60 min) leads to heightened fear learning and defensive behaviors. Our lab and others have shown that stress leads to hyperexcitability and neural plasticity in the basolateral amygdala (BLA), leading to an enhanced propensity for future associative learning. In the present set of studies, we further tested the functional impact of traumatic stress on BLA synapses. We conducted whole-cell voltage clamp recordings of BLA neurons in acute brain slices from adult male and female C57BL/6J mice in order to compare miniature excitatory postsynaptic currents (mEPSCs) between animals that received traumatic stress or no stress. While there was no difference in the amplitude of the mEPSCs,

there was increased frequency of mEPSCs in the neurons of stressed mice. Next, we utilized a novel activity-dependent labeling AAV (AAV9-RAM-d2TTA:TRE-hM4Di-mCherry-WPREgamma, "RAM" virus) that fluorescently tags neurons that have expressed c-fos and/or Npas4, immediate-early genes associated with neuronal activity. This custom viral vector allows for robust and temporally specific labeling and inhibition of previously activated neurons, i.e. "engram" cells. BLA neurons were tagged during traumatic stress in male and female mice and then whole-cell voltage clamp recordings were performed one day after the stress. We confirmed in stressed mice that there was increased mEPSC frequency, but not amplitude, in tagged neurons vs. untagged neurons. Additionally, we confirmed that clozapine-n-oxide successfully reduces neuronal excitability in tagged BLA cells. Taken together, these data suggest that trauma promotes excitatory input onto BLA neurons, likely through either increased presynaptic neurotransmitter release and/or number of synapses. Our findings additionally provide partial validation of the novel RAM virus and indicate we can further dissect functional difference between different neuronal populations based on their prior activity history. SUPPORT: Funded by NIMH R01-MH115678, NIDA T32-DA024635, NIDA F31-DA054792

Monfils, MH (*U of Texas at Austin*) Last call for alcohol! ABSTRACT: Dr. Nadia Chaudhri was an esteemed colleague and researcher who contributed greatly to our understanding of Pavlovian alcohol conditioning. From 2014 to 2019, my colleagues at University of Texas at Austin and I collaborated with Nadia. In this talk, I will showcase some of the work we did together, and highlight the continued impact on the field.

Moore E, Harris H, Slover W, Lipatova O, Campolattaro M (*Christopher Newport U*) Generalization of associative responding between tone-off auditory cues ABSTRACT: Generalization is the ability to apply what has been learned in the past to similar situations in the present and future. The ability to generalize demonstrates that learned behavior is flexible, however it tends to occur more readily when situations are similar. Generalization studies often use conditions that involve turning on a CS (e.g., turning on a tone). We wanted to know how readily generalization occurs in circumstances when the CSs are turn-off cues. To accomplish this goal, we used delay eyeblink conditioning procedures to train rats to associate the absence of background tone (2-kHz or 8-kHz 500-msec tone-off CS, counterbalanced) with a 25-msec periorbital electrical stimulation US (1-2 mA). We subsequently tested for immediate generalization of eyeblink CRs. To do this, the background tone frequency was changed (e.g., 2-kHz to 8-kHz) and tone-off CS-alone presentations were given. We found that rats did not immediately generalize when the background tone frequencies were changed, even though the CS presented during this and the previous training sessions were identical (i.e., a brief period of no sound). Next, we tested for generalization by pairing the absence of the new tone frequency with the US, and found that faster conditioning occurred during these sessions relative to initial training. The lack of immediate generalization suggests that rats learned about the frequency of the background tone during initial training, and the finding that rapid learning occurred during the subsequent training sessions showed that rats can generalize and flexibly apply learned associations across training conditions with tone-off CSs.

Moore S[1], Wang Z[1], Sun R[2], Lee A[1], Zhu Z[1], Charles A[2], Kuchibhotla KV[1] ([1]*Department of Psychological and Brain Sciences, Krieger School of Arts and Sciences, Johns Hopkins U;* [2]*Department of Biomedical Engineering, Whiting School of Engineering, Johns Hopkins U*) Slow or sudden: revealing naturalistic transitions to habitual behavior during learning ABSTRACT: Animals use different decision processes to efficiently adapt to complex environments. When operating in a goal-directed mode, animals deliberate amongst alternatives based on their motivation. As the statistics of the environment become predictable, animals can shift to a more automatic habit mode. The transition from goal-directed to habitual decision-making during learning is thought to be gradual, yet permanent. Current approaches for distinguishing between the two decision processes require discrete 'test' sessions that preclude assessment of the nature, timing and properties of transitions. Here, we devised a naturalistic devaluation approach to assess the underlying decision mode en passant, without discrete 'test' sessions. Mice motivated by a taste preference for normal water (provided during the task) versus mice with ad libitum access to water doped with citric acid initially exhibited naturalistic fluctuations in the cue-driven response rate during discriminative instrumental learning. Then, abruptly and overnight, this variability ceased, and mice transitioned to a high and stable response rate. To test whether habit-like transitions were smooth or state-based fluctuations we applied a hidden Markov model to our dataset and found that during the goal-directed phase, distinct states best described action rate fluctuations, while post-transition, the behavior abruptly switched to a single state. The sudden transitions were also accompanied by signatures of automaticity in lick microstructures. Surprisingly, some animals reverted to goal-directed mode after several sessions in habit mode suggesting that transitions to habitual decision-making are not intrinsically permanent. Ongoing work aims to isolate pupil-based biomarkers of goal-directed and habitual behavior and to identify the role of the dorsolateral striatum (DLS) in guiding these abrupt transitions. Thus, naturalistic devaluation provides a powerful en passant approach to study habit formation and shows that transitions to habitual decision-making are strikingly abrupt, but also reversible.

Mueller DM, Giglio EM, Chen C, Grissom NM (*U of*

Minnesota) Explore-exploit state governs the spatial configuration of touch actions in a mouse bandit decision making task ABSTRACT: In bandit decision making tasks, the challenge of sampling between options versus settling on a currently best option is better known as the explore-exploit tradeoff. Across species, there is substantial evidence that explore and exploit can be defined as neurobehavioral states using a hidden markov model (HMM) approach. Using a restless bandit task, in which the reward probability of each choice changes randomly and independently across trials, we see that animals enter self-initiated periods of exploration and exit these to begin exploiting an option. In mice, the use of touchscreen chambers allows us to record precise locations for each mouse touch, allowing us to consider detailed information about how decisions translate into physical motion. Transitioning between explore and exploit states could be considered as an online change in cognitive flexibility, which may be reflected in motor and behavioral flexibility. We took advantage of the data on touch locations to test whether individual trials labeled as exploit by our HMM are accompanied by more stereotyped motor behaviors in choice selection than the same choices during explore states. Thirty-two 129/b6j F1 mice (16 male and 16 female) were tested on restless bandit schedules. We find that successive touches to the same choice are further apart while an animal is in an explore state than in an exploit state, suggesting greater motor stereotypy when exploiting an option. Male mice tended to have a wider range of nosepoke coordinates than female mice do across states, suggesting different levels of coordination between motor and cognitive systems across sexes. This novel analysis has the potential to allow all labs using touchscreens to investigate how stereotyped motor behaviors may be captured in response data and reflect hidden contributions to decision flexibility.

Olsen C, Liu S, Sarka B, Nawarawong N (*Dept. of Pharmacology, Neuroscience Research Center, Medical College of Wisconsin*) Prefrontal cortex drug seeking ensembles: necessity, specificity, and modulation by injury ABSTRACT: Neuronal ensembles are groups of neurons that are that are important in memory recall. Drug seeking and extinction have been shown to be modulated by ensembles in the medial prefrontal cortex (mPFC). We have found that reactivation of an ensemble in the dorsomedial prefrontal cortex (dmPFC) is correlated with persistence of drug and non-drug reward seeking. Here, we expanded this work by testing the hypothesis that inhibition of the dmPFC drug seeking ensemble would suppress persistence of drug seeking without affecting other behaviors. We also tested the hypothesis that an injury producing neuropathic pain during abstinence would increase opioid seeking and selectively increase intrinsic excitability of dmPFC ensemble cells. Chemogenetic inhibition of a dmPFC cocaine seeking ensemble suppressed persistence of cocaine seeking without affecting other tested behaviors, supporting our hypothesis that the ensemble was necessary and specific for cocaine seeking. Neuropathic pain induced during abstinence from oxycodone self-administration produced sex-specific increases in drug seeking and dmPFC intrinsic excitability. In conclusion, the dmPFC cocaine seeking ensemble was necessary and specific for cocaine seeking in mice of both sexes. Neuropathic pain during oxycodone abstinence produced sex-specific increases in oxycodone seeking and dmPFC excitability, although this change in excitability was not selective to ensemble neurons. SUPPORT: NIDA, Advancing a Healthier Wisconsin

Olvera ME, Roberto RU, Cervantes MC, Monfils M-H, Gonzales RA, Lee HJ (*Department of Psychology, The U of Texas at Austin, Austin, Department of Psychological Sciences, The U of Missouri, Division of Pharmacology & Toxicology, The U of Texas at Austin, Austin*) Conditioned responses to cues associated with alcohol availability in rats with a history of ethanol dependence ABSTRACT: Environmental cues present during alcohol consumption can become conditioned stimuli that trigger alcohol-seeking behavior. Significant knowledge about cue-driven alcohol-seeking behavior and the underlying neural mechanisms are based on animal models exposed to moderate levels of alcohol, including those used in our own studies (Cofresi et al., 2017, 2018a,b, 2019). However, there is limited understanding of acquisition and extinction of Pavlovian conditioned responses in rat models that mimic human conditions of problematic alcohol use and dependence. Evidence suggests that physical dependence on alcohol can lead to impaired extinction (Gass et al., 2014) and enhanced reinstatement (Liu & Weiss, 2002) of operant behavior (but see Carpio et al., 2022). These changes in behavior produced by physical dependence on alcohol suggest that Pavlovian conditioning processes also may be altered. However, to our knowledge, this has not yet been specifically tested. The current study aims to characterize acquisition of conditioned responses to alcohol-associated cues in rats with a history of alcohol dependence. After intermittent access to unsweetened alcohol in home-cage, male rats were exposed to ethanol vapor (daily 14-hr exposure for 10 days). Then, the rats received Pavlovian conditioning in which a visual cue (light) predicted 10-sec access to unsweetened alcohol. Over the course of light-alcohol pairings, the rats developed conditioned alcohol-seeking behavior in the presence of light and increased consumption of alcohol during the 10-sec access. Overall, the rats previously exposed to the ethanol vapor acquired comparable conditioned responses as the control rats that were not exposed to ethanol vapor. Interestingly, the rats previously exposed to the ethanol vapor displayed slightly greater conditioned alcohol-seeking behavior at the beginning of the light onset but both groups showed comparable consummatory behavior when alcohol became available.

Consequently, we plan to further study the degree of cue-alcohol association and its influence on relapse-like behavior in rodent models of alcohol dependence.

Pahua AE, King C, Davison T, Mali I, Payne B, Plakke B (*Kansas State U*) Impact of exercise on cognitive performance in a rodent model of autism ABSTRACT: Valproic acid (VPA) is frequently prescribed to treat mood swings, migraines and epilepsy. Previous research has indicated an increase in susceptibility to children developing autism spectrum disorder (ASD) when their mothers took VPA during pregnancy. ASD is defined as the expression of restricted, repetitive and stereotyped behaviors (RRBs) and impairments in social communication. Several studies examining the effects of exercise intervention have found that spatial memory, cognitive ability and behavioral outcomes are improved. This study focused on effects of aerobic exercise on attentional set-shifting in the VPA rat model of autism. Pregnant Long-Evans rats received a single dose intraperitoneal injection of either saline or VPA (600 mg/kg valproic acid) on gestational day 12. It was hypothesized that rats who received the exercise intervention would demonstrate improved cognitive performance when compared to the sedentary rats. Using a rodent treadmill, exercise was started on PND 40 and animals ran on a 0% slope for 30 minutes/day, 5 days/week for 4 weeks. Attentional set shifting was conducted in the fourth week of running. All behavioral data collection and MRI brain segmentation was conducted by blind-to-condition researchers. Data was separated by sex based on past findings. The study used N=88 (Male: Exercise Control 9, Sedentary Controls 9, VPA Exercise 12, VPA Sedentary 13; Female: Exercise Control 10, Sedentary Control 9, VPA Exercise 13, VPA Sedentary 13). Preliminary data analysis demonstrates that exercise groups were significantly better at making extra-dimensional shifts of attention compared to sedentary groups. This behavioral data supports the hypothesis that exercise can improve some forms of executive function even in a model of ASD.

Perez-Torres J, Vega-Reyes AG, Bonilla-Gutiérrez MF, Ramirez-Sanchez LJ, Boyle S, Li B, Bravo-Rivera C (*U of Puerto Rico School of Medicine*) Neural circuits mediating reward approach and punishment avoidance conflict ABSTRACT: Reward is often present in risky environments, requiring individuals to weigh the benefits of rewards against the associated risks. There are individuals that are unable to choose an appropriate response during risky reward opportunities and thus exhibit extreme avoidance or risky behaviors that can severely impair quality of life or endanger people. It is therefore necessary to characterize how neurons mediate reward approach and threat avoidance conflict. Here, we adapted the platform-mediated avoidance conflict task (Bravo-Rivera et al 2014; Bravo-Rivera et al 2021), such that water-deprived mice could nose-poke for a light-signaled water reward and avoid a tone-signaled foot-shock by stepping onto a safety platform away from the reward port. Optogenetic activation of GABAergic neurons in the ventral pallidum invigorated reward approach at the expense of receiving shocks. Photometry recordings of glutamatergic neurons in the ventral pallidum and in the lateral habenula during conflict revealed that these structures promote avoidance and become inhibited during conflicted reward approach. These results suggest that a pallidal-habenula circuit mediate motivational conflict. We also compared behavioral conflict in male and female mice. Interestingly, females stepped on the platform earlier than males after tone onset and took longer to leave the platform after tone offset. Males received more shocks than females (5 vs 2 out of 20) and received more water reward (759 ul vs 609 ul) than females by the end conflict training. Moreover, females exhibited more tone-induced freezing (33% vs 15% of tone duration) and exhibited more frequent darting (73% vs 51% of trials) than males. These results suggest that females exhibit more avoidance behavior and less reward approach than males in the face of approach/avoidance conflict. SUPPORT: MH123495, MH058883, NS115917, MH108924

Peterson S, Chavira J, Maheras A, Garcia-Arango A, Seamans E, Keiflin R (*UC, Santa Barbara*) Role of the orbitofrontal cortex and dorsal hippocampus in the expression and inferred generalization of contextual rules for reward prediction ABSTRACT: Contextual control over cue-evoked appetitive behavior is essential for adaptive behavior. The lateral orbitofrontal (OFC) cortex and the dorsal hippocampus (dHipp) harbor neurons that encode reward cues in a context-dependent manner. However, the causal role of these brain regions in learning and enforcing the contextual control over cue-evoked reward seeking remains uncertain. Here we used chemogenetic silencing to interrogate the necessity of the OFC and the dHipp in the expression and inferred generalization of contextual rules for cue-evoked reward seeking. Rats were trained in either a context-dependent or a simple Pavlovian discrimination task. In the context-dependent discrimination task, a visual contextual cue (A) informed the predictive status of two brief auditory cues (X and Y), so that one cue was rewarded only in the presence of A, while the other cue was rewarded only in its absence (A:X+ / X- / A:Y- / Y+). In the simple discrimination task, only one cue was rewarded regardless of context (A:X+ / X+ / A:Y- / A:Y-). For each training condition, rats were made to express the inhibitory DREADD hM4di or the control transgene mCherry in the lateral OFC or the dHipp. Silencing the OFC (by systemic injection of the DREADD ligand CNO) disrupted context-dependent but not simple discrimination performance. Silencing the dHipp had no effect in either task. To explore the role of the OFC and dHipp in the acquisition of contextual control, we then leveraged the predictions of computational models. Several models predict that animals who previously learned a contextual rule for reward

prediction should be prone to expend this rule over new associations. In line with this prediction, we observed that rats previously trained in the context-dependent discrimination task spontaneously inferred that any new reward cue obeyed a contextual rule, even in absence of explicit information to confirm this belief. This rapid acquisition of an inferred context-dependent discrimination was abolished by silencing the OFC or the dHipp. In contrast, animals previously trained in a simple discrimination task learned a new association in a context-independent manner, and OFC or dHipp silencing had minimal effects on this process. These results indicate that the orbitofrontal cortex and the dorsal hippocampus both contribute, in different ways, to the contextualization of associative predictions. Specifically, the orbitofrontal cortex is necessary for both the expression and the rapid acquisition of new (inferred) contextual rules. In contrast, the role of the dorsal hippocampus appears limited to the rapid acquisition of contextual rules but the not the behavioral expression of those rules.

Pierce-Messick ZJ, Corbit LH (*U of Toronto*) The influence of extinguishing the context-response operandum pairing on goal-directed behaviour ABSTRACT: Goal-directed behaviour is that which is sensitive to the dynamic value of the outcome that it produces, as well as to the contingency between the action and outcome. Alternatively, habitual control is not sensitive to such things, and is considered to be performed automatically based on previously established stimulus-response (S-R) associations. However, the stimulus and response that form the S-R association underlying habitual control are often ill-defined. I will describe experiments that aimed at 1) identifying one possible stimulus involved in S-R learning underlying habitual control, and then 2) modifying the relationship of this stimulus with the response in order to promote goal-directed control at the expense of S-R control. An incidental stimulus present during free-operant conditioning is the chamber or context. Thus, the training context (involving the physical chamber itself, as well as the illuminated house-light) was considered the dominant stimulus exerting S-R control over behaviour, and the response operandum itself (a lever that was previously trained to yield reward pellets) was considered the response involved in the S-R association. We hypothesized that extinguishing the relationship between the context (S) and the operandum (R) would weaken the S-R relationship and promote goal-directed behaviour in a devaluation test utilizing sensory specific satiety. Rats were trained to perform a single response using variable interval schedules reported to encourage habit learning (Dickinson et al., 1983). After confirming poor goal-directed control during a devaluation test, animals were separated into two groups. Both groups were retrained according to a VI-60 schedule the day before each devaluation test. In order to extinguish the association between the context and response, one group was exposed to the instrumental context with the house-light illuminated for 30 mins before the retraining program was started, at which point the lever was presented. The other group was placed into the training boxes and the program was started immediately. Results from the devaluation tests suggest that goal-directed control was promoted in the group that was explicitly exposed to the context without the response operandum present. While sensitivity to outcome devaluation provides evidence of goal-directed control, insensitivity, while indicating lack of goal-directed control, provides little satisfying detail about the nature of the presumed habit or how it is controlled. These findings and related experiments could help scientists better understand and specifically define the conditions necessary for habitual control.

Plakke B, King C, Davison T, Maze T, Mali I, Payne M, Bossmann S (*Kansas State U, Dept Psych Sci*) Enlarged frontal cortices during adolescence impacts cognitive performance in female autism spectrum modeled rats ABSTRACT: People with ASD (autism spectrum disorder) have a unique developmental trajectory with overgrowth occurring in the frontal cortex during adolescence. This study used the valproic acid (VPA) model to induce ASD-like symptoms in rodents. Prior studies have demonstrated that VPA animals are impaired on executive function, paralleling results in humans with ASD. Pregnant dams were injected with 600 mg/kg VPA or saline on gestational age 12. To control for the litter effect, one male and one female pup per litter were assigned to an experimental condition. 30 rats performed a set-shifting task (9 male controls, 7 VPA males, 9 control females, 7 VPA females) and then brains were imaged with MRI 3D scans on postnatal day 40. Data analysis compared male and female data separately based on past findings (McKinnell et al.,2020). Researchers that conducted behavior and volumetric segmentation for brain regions were blind-to-condition. The VPA adolescent female rats were impaired on the set-shifting task and had enlarged frontal cortices compared to control females. In addition, adolescent VPA females with enlarged frontal cortices performed the worst of all groups across the entire task. These results mirrors those observed in females with ASD, suggesting that ASD-like symptom progression in the VPA model is similar to that observed in humans. These novel findings highlight the importance of studying the brain at different developmental stages and implicate overgrowth of the anterior cingulate cortex are impacting executive functions in ASD. SUPPORT: the National Institute of General Medical Science GM113109

Rajbhandari AK, Shetty S, Duesman S, Das A, Ogale N, Rajbhandari P, Rajbhandari P (*Icahn School of Medicine at Mount Sinai*) Role of PACAP and PAC1 in stress-related fear and cardiorespiratory functions ABSTRACT: Stress related conditions like post-traumatic stress disorder (PTSD) are increasingly being recognized as brain and body disorder with alterations in fear expression and car-

diorespiratory functions. PTSD is involves exaggerated activation of the sympathetic system associated with enhanced arousal and fear, and worsened cardiorespiratory outcomes. For homeostatic balance of behavioral and other functions, the sympathetic and parasympathetic systems need to be in a balanced state. Approaches that increase parasympathetic tone such as vagus nerve stimulation have been shown to enhance extinction of fear but the precise biological mechanisms are not clear. We have shown that the nodose ganglion of the vagus nerve expresses high levels the neuropeptide PACAP, which innervates the nucleus of the solitary tract (NTS). A relatively less studied role of parasympathetic regulation in behavior and cardiorespiratory functions involves the nucleus ambiguus (NAmb) to the heart pathway, which also has high PACAPergic expression. Human and animal studies have linked PACAP/PAC1 to PTSD diagnosis and symptom severity and regulation of sympathetic/parasympathetic functions. PAC1 receptors are associated with changes in both fear and cardiorespiratory functions. However, the role of PACAP/PAC1 via the parasympathetic pathways in regulation of behavioral and cardiorespiratory functions has not been studied together. We hypothesized that modulation of PACAP either in the nodose to brainstem pathway or the NAmb to heart would increase parasympathetic tone resulting in decreased fear, anxiety, and cardiorespiratory outcomes. We injected excitatory or inhibitory DREADDs in either the nodose ganglion or NAmb of mice that express Cre-recombinase in PACAPergic cells and then measured fear using stress-enhanced fear learning and modified open field. Telemetry measurements were carried out during the freezing measurements or during anxiety test. Chemogenetic stimulation of nodose to NTS pathway via injections of clozapine-N-oxide decreased anxiety and cardiorespiratory tone. NAmb to heart chemogenetic activation did not alter fear expression, but inhibition of this pathway led to enhanced fear in unstressed mice. Taken together, our results indicate that PACAP/PAC1 is an important neuropeptidergic system in the brain parasympathetic node that can regulate stress-associated behavioral and cardiorespiratory functions. SUPPORT: Brain and Behavior Research Foundation, National institute of Diabetes and Kidney Disease, Friedman Brain Institute, Akira Arimura, WhiteHall Foundation

Raskin MR, Malone CA, Hilz EN, Shumake J, Lee HJ, Monfils MH (*Institute for Neuroscience & Dept. of Psychology, UT Austin*) CO2 reactivity predicts spontaneous recovery of conditioned food seeking in rats ABSTRACT: Extinction of fear and reward memories are subject to the same return of behavior phenomena and have overlap in neural circuitry (Millan et al., 2011; Peters et al., 2009). Extinction learning underlies exposure therapy, which is one of the best available treatments for anxiety-related disorders and addiction; however, our ability to predict whether an individual will respond to extinction/exposure remains limited. Determining, pre-treatment, whether an individual is a good candidate for exposure would be helpful to patients and clinicians alike. Orexin neurons, which originate in the lateral hypothalamus (LH), have been implicated in the extinction of fear and reward memories. While orexin cannot be directly measured, our lab and others have found that orexin neurons in the LH are activated by exposure to carbon dioxide (CO2) (Johnson et al., 2012; Monfils et al., 2019). We recently found that in rats, behavioral reactivity to a CO2 challenge explained a significant portion of the variance in long-term fear extinction memory (Monfils et al., 2019). Specifically, less CO2 reactivity (determined by measures such as ambulation during flush-out) predicted better long-term fear extinction memory and fewer activated orexin neurons in the LH. As such, CO2 reactivity can potentially be used as a proxy for orexin activation to predict fear extinction memory. CO2 challenge is safe for use in humans, and CO2 reactivity was found to be predictive of PTSD symptom development in war veterans (Telch et al., 2012). Here, we tested the hypothesis that CO2 reactivity would be predictive of long-term appetitive extinction memory. Rats underwent a CO2 challenge, followed by Pavlovian light-food conditioning, extinction, and a long-term memory test. Using the best subset approach to linear regression, we identified ambulation and rearing during CO2 flush-out as the strongest predictors in our model, together explaining 23% of the variance in spontaneous recovery of conditioned food seeking. Specifically, ambulation during flush-out was positively correlated with spontaneous recovery of conditioned food seeking. This suggests that the relationship between appetitive extinction and CO2 reactivity is the opposite of what we previously found with fear extinction, which was negatively correlated with ambulation during flush-out; yet, both results are consistent with work showing that orexin blockade attenuates extinction of food seeking behavior, but facilitates fear extinction (Flores et al., 2014; Keefer et al., 2016). Future analyses will determine whether conditioned orienting phenotype (known to moderate appetitive extinction memory (Olshavsky et al., 2013)) moderates the relationship between CO2 reactivity and spontaneous recovery of conditioned food seeking.

Ren LY, Cicvaric A, Zhang H, Aa Meyer M, Guedea AL, Gao P, Petrovic Z, Sun X, Lin Y, Radulovic J. (*Albert einstein College of Medicine, Northwestern U, Aarhus U*) Retrieval-based generalization of aversive conditioning ABSTRACT: Generalization, the process of applying knowledge acquired in one context to other contexts, often drives the expression of similar behaviors in related situations. At the cellular level, generalization is thought to depend on the activity of overlapping neurons that represent shared features between contexts (general representations). Using contextual fear conditioning in mice, we demonstrate that generalization can also occur in response to stress and

result from reactivation of specific, rather than general context representations. We found that generalization emerges during memory retrieval, along with stress-induced abnormalities of septohippocampal oscillatory activity and acetylcholine release, which are typically found in negative affective states. In hippocampal neurons that represent aversive memories and drive generalization, cholinergic septohippocampal afferents contributed to a unique reactivation pattern of cFos, Npas4, and repressor element-1 silencing transcription factor (REST). Together, these findings suggest that generalization can be triggered by perceptually dissimilar but valence-congruent memories of specific aversive experiences. Through promoting the reactivation of such memories and their interference with ongoing behavior, abnormal cholinergic signaling could underlie maladaptive cognitive and behavioral generalization linked to negative affective states.

Rojas, GR, Grissom, NM (*U of Minnesota*) Choice inflexibility is observed in male 16p11.2 hemideletion mice during acquisition of delay but not probability discounting. ABSTRACT: Delay discounting and probability discounting tasks are thought to reflect different aspects of impulsive choice. The amount of discounting reflects the individuals' value assessment of the reward. Steep delay discounting is thought to reflect an inability to wait for a larger reward and seek immediate gratification. Neurodevelopmental disorders like ADHD or ASD have been linked to impulsive choice under delay discounting. We had mice with hemideletion of 7qF3 - a model of 16p11.2 hemideletion in humans which is associated to neurodevelopmental disorders - undergo delay discounting and probability discounting in order to determine if mice were more sensitive to a temporal cost or uncertainty. Our lab has previously demonstrated mice can acquire both tasks in sequence. We had mice experience both "Worsening" (i.e. increasing delay cost or uncertainty within a session) and "Improving" (i.e. decreasing delay cost or uncertainty within a session) versions of delay and probability discounting to determine if mice were sensitive to task order changes. We found male 16p11.2 hemideletion mice were more willing to endure long delays in order to receive large rewards. This is in contrast to probability discounting where they were less willing to endure risk of loss of a reward. Contrary to a hypothesis of impulsive choice, male 16p11.2 hemideletion mice repeatedly sought out large rewards. Our data suggests choice history is important when assessing decision-making in neurodevelopmental disorders. SUPPORT: NIMH R01 MH123661, NIMH P50 MH119569

Rosenkranz JA (*Chicago Medical School/RFUMS*) Effects of mild inflammation on amygdala and social function. ABSTRACT: Mild peripheral inflammation is caused by a wide range of health conditions and is associated with higher risk of depression and anxiety. Activity in the amygdala is sensitive to peripheral inflammation in humans, and amygdala hyperactivation has been associated with anxiety. Peripheral inflammation can impact forebrain regions that modulate learning and emotion, but much of the prior work examines high levels of inflammation that can produce sickness syndrome (lethargy, anorexia, fever), which interferes with expression of many behaviors. In a series of studies we tested whether mild peripheral inflammation impacts amygdala activity and amygdala-related behaviors independent of sickness syndrome. We focused on social behaviors that rely on a balance of approach-avoidance that is highly sensitive to peripheral inflammation. Using in vivo electrophysiology we found that low dose IL-1b increases basolateral amygdala neuronal activity and shifts microglia towards an activated profile. In parallel experiments, low dose IL-1b decreased interaction in open field social tests. To further clarify effects on BLA function, we measured effects on cued fear conditioning and found that IL-1b had small effects on conditioned fear expression and acquisition of extinction. However, prolonged inflammation produced a unique effect that included more subtle changes in social habituation and basolateral amygdala activity. These studies begin to uncover how mild peripheral inflammation can impact social behaviors, and point to a potential substrate for this effect.

Sangha S (*Indiana U School of Medicine*) Using learned safety cues to map the behavioral and circuit mechanisms of fear regulation ABSTRACT: Stressful events can have lasting and impactful effects on behavior, especially by disrupting normal regulation of fear and reward processing. Posttraumatic stress disorder (PTSD) has become a growing public health concern, with a lifetime prevalence of 6.4% of the US population, and a high rate of comorbidity with additional psychiatric disorders. Among these, alcohol abuse or dependence is prevalent in 41.8% of people with PTSD. Increasing evidence suggests PTSD and chronic alcohol use may alter the ability to tell the difference between different stimuli, in particular where PTSD disrupts the ability to use signals that indicate safety, and overgeneralize the impact of fear signals. Our lab uses a unique behavioral task that allows us to examine stimulus discrimination alongside regulation of fear and reward processing behaviors. We have discovered sex differences in these behaviors under both stress and stress-free conditions. This task includes different stimuli that are paired with natural reward (sucrose), an aversive stimulus (footshock), or no outcome ("S-"). When the S- is presented along with the fear stimulus, but without footshock (FS-), male rats show significantly reduced fear expression while females do not. Prior stress in males dampens this fear reduction during FS-, suggesting stress impairs safety learning in males. Both sexes reduce sucrose seeking when the S- is presented with the reward stimulus but without sucrose (RS-), thus females can use S- to alter responding, just not for fear cues. Our work shows the ability of unstressed males to significantly downregulate fear in the presence of S-

(FS- cue) is dependent on the infralimbic region of the medial prefrontal cortex (IL), specifically the neurons projecting to the central amygdala (CeA), but not basolateral amygdala (BLA). We also show increased neural activity in the IL during the FS-, consistent with other works on fear extinction and latent inhibition of fear, suggesting a specific role of the IL in downregulating fear across several types of inhibitory memories.

Sarka BC, Liu S, Liu QS, Stucky CL, Olsen, CM (*Medical College of Wisconsin*) Measuring the effect of neuropathic pain on drug-seeking ensembles in the dmPFC ABSTRACT: Approximately 50 million Americans suffer from chronic pain, and opioids are commonly prescribed for such individuals. Unfortunately, nearly a quarter of chronic pain patients have reported misusing their prescription. We are investigating the effect of chronic pain on drug-seeking behavior at the neuronal level. Repeated drug-seeking is associated with reactivation of an ensemble of neurons sparsely scattered throughout the dorsomedial prefrontal cortex (dmPFC). Prior research has demonstrated that chronic pain increases intrinsic excitability of dmPFC neurons, which may increase the likelihood of reactivation during drug seeking. We tested the hypothesis that chronic pain would increase oxycodone seeking behavior, and that the pain state would differentially increase intrinsic excitability in dmPFC drug seeking ensemble neurons.

TetTag mice self-administered intravenous oxycodone. After 7 days forced abstinence, a drug seeking session (extinction conditions) was performed and the ensemble was tagged. Mice received spared nerve injury (SNI) to induce chronic pain during the period between a first and second seeking session, and we measured persistence of seeking between the two sessions to determine if the SNI exacerbates seeking. Following the second seeking session we performed electrophysiology on individual neurons within the dmPFC to assess intrinsic excitability of the drug-seeking ensemble and non-ensemble neurons. We found significant sex differences in the effect of SNI on oxycodone seeking and electrophysiology, such that the induction of chronic pain can modulate seeking behavior in mice that have previously self-administered oxycodone prior to injury. SUPPORT: Qingsong Liu and Shuai Liu for their help with electrophysiology. Cheryl Stucky & lab for their help with pain models and pain assays.

Sattler K, Miller R, Zelikowsky M (*U of Utah*) The role of the ventral hippocampus in trauma-induced aggression and enhanced fear ABSTRACT: A single, acute traumatic experience can result in a host of negative behavioral effects, such as increased aggression and exaggerated fear responses to mild stressors. Despite the large body of research on the neurobiology of trauma, we nevertheless have a poor understanding of how the brain encodes trauma-associated changes in behavior. The ventral hippocampus (VH) is well-suited for processing the multi-modal behavioral effects of trauma, as it receives inputs from sensory integration sites and sends output to regions involved in social and emotional behavior. Importantly, the VH projects to both the basolateral amygdala (BLA), a central node for fear, and the ventromedial hypothalamus (VMH), which has been implicated in aggression. Thus, we hypothesized that the effects of trauma to enhance both fear and aggression are controlled by projection neurons in the VH. To test this hypothesis, mice were injected with a virus encoding hM4D into the VH to allow for chemogenetic silencing of the VH during the resident intruder assay. We show that the VH is necessary for trauma-enhanced aggression. To further dissect the involvement of neuronal ensembles in the VH to encode the effects of trauma on both enhanced fear and enhanced aggression, we tested whether ensembles activated by each phenotype have unique projection profiles using tract tracing and immunohistochemistry. Collectively, our findings suggest a role for the VH as a central hub underlying trauma-altered social behaviors and provide insight into how experiencing a traumatic event can lead to diverse behavioral changes.

Schwabe MR, Fleischer AW, Kuehn RK, Beaty HA, Milkie EM, Schnitzler AL, Chaudhury S, Donaldson WA, Sem DS, York JM, LaDu MJ, Frick KM (*U of Wisconsin-Milwaukee, Marquette U, Concordia U Wisconsin, U of Illinois at Chicago*) Effects of a novel estrogen receptor beta agonist and APOE genotype on synaptic markers of memory in a mouse model of Alzheimer's disease. ABSTRACT: Among the hallmark pathologies of Alzheimer's disease (AD) is significant synapse loss. The e4 allele of apolipoprotein E (APOE4) increases synapse loss in AD patients. While females are at a greater risk for AD compared to males, female APOE4 carriers have the greatest risk for AD. Estrogen therapy increases synaptic proteins and dendritic spine density, potentially preventing or reversing AD-induced synaptic changes. However, estrogen treatment is associated with increased risks of cancer due to activation of estrogen receptor a (ERa). Thus, selective ERβ agonists may be safer alternative therapies. To study interactions between APOE genotype and estrogen treatment in AD, we used mice EFAD mice (5xFAD+/-/APOE+/+), specifically E3/3FAD, E3/4FAD and E4/4FAD. E4/4FAD mice have decreased expression of the synaptic proteins synaptophysin and PSD-95, increased GFAP, and reduced basal dendritic spine density in the mPFC and CA1 region of the dorsal hippocampus relative to E3/3FAD mice (Taxier et al., 2022). Dorsal hippocampal infusion of 17b-estradiol (E2) increases apical CA1 dendritic spine density in E3/3FAD and E3/4FAD females, but not E4/4FAD females (Taxier et al., under review). Thus, the goal here is to determine the extent to which a novel highly selective ER/*beta* agonist, EGX-358, increases expression of synaptic markers associated with memory in E3/3FAD and E3/4FAD mice. E3/3FAD and E3/4FAD mice

were ovariectomized at 5 months of age and then treated orally with vehicle (1% DMSO) or EGX358 (10 mg/kg/day) via hydrogel for 9 weeks. Mice were tested for spatial and object memory, anxiety-like behavior, and vasomotor symptoms following injection of the tachykinin receptor 3 agonist senktide, with testing concluding 2 weeks prior to tissue collection. On the day of collection, brains were removed, flash frozen, and hemisected 1-3 hours after a final EGX358 treatment. Half the brain was Golgi stained for measurement of apical and basal dendritic spine densities on DH and PL/IL mPFC pyramidal neurons. The remaining half was flash frozen for Western blot analysis of the DH and mPFC. We found that EGX-358 enhanced object recognition memory in both E3/3 and E3/4 mice compared to vehicle. Preliminary analyses of brain tissue suggest no effects of EGX358 or APOE genotype on DH expression of PSD95 or synaptophysin; ongoing analyses are assessing other proteins associated with synaptic plasticity including phosphorylated CREB, GFAP, and Iba-1. Dendritic spine analyses are also in process and should provide insights into whether ER/*beta* agonism interacts with APOE genotype to impact synaptic morphology in a background of AD-like pathology. SUPPORT: This project was supported by a UW System Regent Scholar Award to KMF and 2R15GM118304-02 to DSS, KMF, and WAD. The Frick lab was also supported by R01MH107886 to KMF, the UWM College of Letters & Science, and the UWM Office of Undergraduate Research. Additional funding for the LaDu lab was provided by R01AG056472, R01AG057008, UH2/3NS100127, R56AG058655, philanthropic support from Lou and Christine Friedrich, and UIC institutional funds.

Seese S, Tinsley CE, Hixon JG, Monfils MH (*U of Texas at Austin; OHSU*) Conspecific interactions predict social transmission of fear in female rats ABSTRACT: Social transmission of fear occurs when an individual learns to display a fear response to a previously neutral stimulus by observing or interacting with a conspecific during fear memory retrieval. The conditions under which fear can be learned socially in rats have received attention in recent years. These investigations provide evidence that a variety of social factors modulate the effectiveness of social transmission of information. One such modulator, social rank, has been predominantly studied in males. Specifically, dominance hierarchy was previously found to impact fear conditioning by proxy. Here, we aimed to investigate if social roles in females also have an influence on social transmission of information. In-line with previous findings in males, we find that social interactions in the home cage can describe the social relationship between related female rats in the laboratory and that these relationships predict the degree of fear acquired by-proxy. We also find that female social roles within the home cage may be related to estrous cycle such that more dominant females have longer periods of estrus. This could suggest that greater dominance is associated with greater sexual receptivity.

Shipman ML, Chen SE-S, Desilets GL, Corbit LH (*U of Toronto*) Microglial activation in the DMS impairs goal-directed control ABSTRACT: Recent work in our lab has shown that history of obesogenic diet results in an impairment in goal-directed responding for other reinforcers. While we were able to ameliorate this deficit with manipulation of metabotropic glutamate II receptors within the dorsomedial striatum (DMS), obesogenic diet has been shown to cause numerous changes in the body and brain that might dysregulate glutamatergic systems, including central inflammation. We investigated if lipopolysaccharide (LPS), a drug that induces a strong immune reaction and activation of microglia, would be sufficient to mimic the effects of obesogenic diet and impair goal-directed responding if microglial activation was constrained to the DMS. We found that LPS infused directly into the DMS prior to acquisition sessions was sufficient to disrupt goal-directed responding at test. Immunohistological analyses confirmed an increase in activated microglia at the time that animals underwent operant training. In experiment 1, rats were trained on two levers with a common outcome on a RR1 schedule, followed by three days of RR5 training. They then received three days of bilateral DMS LPS infusions (.1ug/.5ul) followed one hour later by RR10 training with specific outcomes (grain pellets and chocolate pellets) for each lever. Training was spaced by three days due to lingering LPS effects. Rats were then tested with specific satiation as a means of outcome devaluation and responding was examined in an extinction test with both levers. Following a day of retraining, rats were tested with the other outcome devalued. A probe experiment also examined LPS at time of test 1 hour, 1.5 hours, and 3 hours following infusion and found no effect on reinforcer devaluation, implicating a specific role for activated microglia on mechanisms associated with acquisition. A follow up experiment (Experiment 2) is currently determining if minocycline (an antibiotic that prevents microglial activation) in drinking water is sufficient to prevent the disruption to goal-directed control caused by LPS. We hypothesize that minocycline will prevent the LPS-induced impairment in goal-directed control by preventing microglial activation.

Smies CW, Bellfy L, Wright DS, Bennetts SS, Urban MW, Brunswick CA, Kwapis JL (*Penn State*) Epigenetic mechanisms supporting competition in reconsolidation-based memory updating ABSTRACT: Memories are plastic to allow for modification of recorded experiences. During memory consolidation and after memory retrieval, memories become labile, particularly when new information is learned or is incorporated into an already existing memory. Many of the molecular mechanisms underlying memory consolidation have been heavily explored, but the mechanisms unique to reconsolidation-dependent memory updat-

ing are not as well understood. One potential mechanism that may be important for memory consolidation and updating is epigenetic modifications, which change gene expression by modulating chromatin structure. Histone acetylation, a major epigenetic modifier that helps establish a permissive chromatin structure, is modified via the competing actions of histone acetyltransferases (HATs) and histone deacetylases (HDACs). HDAC3, an enzyme that blocks acetylation, functions as a molecular brake pad during memory formation (McQuown et al., 2012); HDAC3 inhibition during memory formation can transform a subthreshold learning event into one that produces robust and persistent long-term memory (McQuown et al., 2012 & Kwapis et al., 2018). However, the role of HDAC3 in reconsolidation-dependent memory updating is unknown. Here we show that systemic administration of the HDAC3 inhibitor RGFP966 improves aging-induced impairments in spatial memory updating in the Objects in Updated Locations paradigm (OUL; reviewed in Wright et al., 2020). Surprisingly, we found that when young animals are systemically administered RGFP966 following an update session, an impairment for the original memory emerges, suggesting that the original and updated information compete for behavioral expression and strengthening the updated memory occurred at the expense of the original information. Next, we used RGFP966 to systemically inhibit HDAC3 immediately after different phases of OUL to test whether strengthening or weakening the original or updated memory can affect this competition process. Together, the current studies demonstrate that HDAC3 plays a key role in both memory formation and reconsolidation-memory updating and suggest that the original and updated information compete for behavioral expression.

Smith RJ (*Department of Psychological and Brain Sciences, Institute for Neuroscience, Texas A&M U*) Investigating the link between habits and punishment-resistant cocaine seeking ABSTRACT: Drug addiction is characterized by compulsive use despite negative consequences. Many have theorized that this type of compulsive behavior is associated with habits. My lab is investigating the relationship between habits and compulsive behavior using an animal model of punishment resistance, in which a subset of rats continue to self-administer cocaine despite footshock consequences. Before and after exposure to footshock punishment during self-administration, we evaluated whether rats' cocaine seeking was goal-directed or habitual using a novel outcome devaluation procedure we developed for intravenous cocaine. We found that habitual cocaine seeking before punishment testing did not lead to increased punishment resistance. However, punishment resistance was associated with habitual cocaine seeking after punishment testing, and conversely, punishment sensitivity was associated with goal-directed cocaine seeking after punishment testing. In other words, rats changed their response strategy in the face of footshock punishment, such that some transitioned from goal-directed to habitual behavior, and others from habitual to goal-directed. These data indicate that transitions between goals and habits are bidirectional, and that transitions can be triggered by changes in the expected outcome, including new aversive consequences. SUPPORT: Funded by NIDA

Sood A, Richard JM (*Univ. of Minnesota*) Investigating ventral pallidal encoding of expected outcome value ABSTRACT: Goal-directed behavior relies on accurate mental representations of outcomes and their expected value. Disruptions in goal-directed decision-making are a core component of several neuropsychiatric conditions, including addiction. An understanding of the neural mechanisms underlying expected value encoding within the brain reward centers is key for developing effective long-term treatments for addiction. The Ventral Pallidum (VP) is a basal forebrain region that is important for motivation and reward processing. Cue-evoked activity in some VP neurons appears to encode the expected value of rewards. Yet, the impact of outcome (reward) devaluation on these neural responses to cues has not been previously assessed. Outcome devaluation, in which the value of the expected outcome is diminished, is a classic test of goal-directed behavior that can be used to test whether these neural responses to cues reflect expected value, or merely other factors that correlate with expected value. In this study, we used in-vivo electrophysiology to record from single units in the VP of adult rats responding to Pavlovian reward cues before and after reward devaluation. Adult rats (n= 5, 3 males, 2 females) were trained with an auditory conditioned stimulus (CS+) that signaled the delivery of a 10% sucrose reward and a control cue (CS-) that never predicted reward. They then underwent reward devaluation via sensory specific satiety where they were allowed free access to either sucrose (thereby altering its expected value) or a control substance (maltodextrin). We recorded from the VP of these rats during training and reward devaluation sessions. As expected, we found that many VP neurons (65%) were excited by the CS+, and that these neurons were more responsive to the CS+ than the CS-. Additionally, VP responses to the CS+ were reduced after free consumption of sucrose. Interestingly, free access to maltodextrin also reduced VP responses to the CS+, suggesting that these changes in VP responding were not due to the encoding of the sucrose reward specifically. Future experiments will aim to investigate whether VP neurons respond more selectively to other forms of devaluation. Overall, this study will enhance our understanding of how VP neurons encode changes in the expected value or rewarding outcomes.

Staffeld J (*Eastern Michigan University; University of Michigan*) Pavlovian Conditioned Avoidance and its Correlation to Pavlovian Conditioned Approach ABSTRACT: Various studies have used Pavlovian conditioned approach

(PCAp) to identify learning differences in addiction models. Sign-tracking, meaning conditioned approach towards an appetitive cue, indicates the cue has acquired incentive value. In contrast, conditioned approach towards the reward itself (i.e. goal-tracking), indicates the cue has acquired predictive but not incentive value. It is possible that propensity to ST may correlate with a tendency to attribute motivational value to aversive cues as well. However, no task has yet been developed to distinguish predictive from motivational value in aversive learning. To this end, we developed a new Pavlovian conditioned avoidance (PCAv) task. First, 36 male Sprague Dawley rats were trained to use a platform to escape a shock predicted by a tone. They then learned to associate a lever with footshock. Finally, rats faced direct conflict in which the platform was placed near the lever. Movement away from the lever was interpreted as a fear response to the lever itself, whereas movement toward the lever and onto the platform was interpreted as using the lever as a predictor of shock. Preliminary results have suggested no statistically significant correlation between PCAp phenotypes and PCAv behaviors, though more detailed analysis is ongoing.

Stidham N, Russo-Savage L, Giddings E, Brabec JL, Lara M, Laprade KA, Bonney EA, Stafford, JM (*U of Vermont; Department of Neurological Sciences U of Vermont; Department of Obstetrics, Gynecology and Reproductive Sciences*) A method for producing high, volitional opioid use during gestation reveals developmental outcomes in offspring that can be mitigated by pharmacological intervention ABSTRACT: Opioid use disorder (OUD) affected nearly 2.7 million Americans in 2020, exacting tremendous physical and mental health cost to the afflicted individuals and society as a whole. Particularly vulnerable to the life-long consequences of OUD are infants delivered to mothers with OUD, a population that has increased over four-fold in the past few years. Here we aimed to develop a novel translation model that overcomes challenges in identify the long-term molecular, neurobiological and behavioral consequences of prenatal opioid exposure. To achieve this, we first built and adapted a device (Abraham et al., 2019) that produces high, volitional oral consumption of the commonly abused opioid oxycodone as well as a vehicle control directly in the home cage. Because these devices allow for the precise monitoring of intake over time, we were able to examine consumption patterns during opioid initiation, the transition to experienced opioid use and finally how opioid consumption patterns in the dams changed from the pre-conception, gestation and weaning phases of the experiment. Second, we included a group that modelled medication for OUD (MOUD) during gestation using the frequently prescribed treatment, buprenorphine. Including this MOUD model group allowed us to assess whether treatment minimized adverse outcomes relative to the oxycodone group. Our first observation was that prenatal opioid exposure had a negative impact on sensorimotor milestones and accelerated weight gain through the pre-weaning period. In contrast, a variety of behaviors such as sociability and contextual fear conditional were largely unaffected suggesting that the impact of prenatal opioid exposure does not have a broad impact on cognitive function. Interestingly, transitioning dams to from oxycodone to buprenorphine around placental development diminished the impact of gestational opioid use on a number of developmental measures. Our results expand a growing number of preclinical and clinical studies on prenatal opioid exposure by creating a more translational model. Our initial studies demonstrate the utility of the model by homing in on specific early developmental features of this disorder that have potential to be targeted by therapy during gestation. Further implications for targeted interventions, additional behavioral outcomes and their neurobiological sequelae will be discussed. SUPPORT: This work was support by a pilot grant from the Northern New England Clinical and Translational Research Network, an initiative created by National Institutes of Health Grant U54 GM115516.

Su CJ, Fukunaga Y, Cazares VA (*Williams College*) No effects of partial reinforcement on fear extinction learning in mice ABSTRACT: Prior studies have investigated a paradoxical finding showing that intermittent CS-US pairings during fear conditioning leads to resistance to extinction learning, relative to a conditioning schedule following a 1:1 CS-US frequency ratio. This effect has been termed the partial reinforcement extinction effect (PREE). It has been largely studied in operant and reward-based learning, but significantly less is known about the PREE in fear learning. Our objective was to investigate the effects of partial reinforcement in fear acquisition, fear memory consolidation, and recall in mice. We investigated whether the effects of partial reinforcement are altered by the number of CS presentations or CS duration. C57BL/6J mice were conditioned to an auditory CS paired with a mild footshock (US) for two days. Following conditioning, extinction was carried out for three consecutive days with one session of 12 unreinforced CS presentations on each day. To assess the effects of partial reinforcement on fear extinction memory, we measured fear recall at 48 hours and 30 days later. Our results demonstrated that partial reinforcement (0.5, CS:US) enhanced fear acquisition and fear retrieval on extinction day 1. However, there were no effects of partial reinforcement on extinction acquisition, consolidation, or recall and no moderating effect of the number of CS presentations or CS duration. Taken together, the present study suggests a small or minimal role of PREE on fear learning in contrast to PREE previously reported in the context of appetitive learning. SUPPORT: We thank Jack Snyder (animal care staff), Allison Davis Research Fellowship, the Wilmers Research & Travel Fellowship, the U.S.-Japan Council Watanabe Endowment Scholarship, and all the individuals who supported our research.

Sullivan, RM (*New York U Langone Medical Center and Nathan Kline Institute*) Infant trauma within social context targets the amygdala to produce deficits in fear and social behavior ABSTRACT: Psychiatric disorders are rooted in early life, although expression is typically delayed until later life. While infant maltreatment is known to be one causal factor, which features of this complex experience are causal remains unknown. For 90min a day from 8-12 days old, rat pups were reared with a maltreating mother (induced by insufficient bedding for nest building) or some feature of maltreatment related to maternal presence (i.e. abusive, control or an anesthetized mother with no maternal behavior or an inanimate object). We paired this with two features of maltreatment, elevated cortisol or 0.5mA shock as pain). Later (13-14 days old), a social behavior test and amygdala/hippocampus anatomy and function were explored. Hippocampus was minimally disrupted in all trauma/stress exposures. All social trauma experiences disrupted amygdala function/anatomy, and stress was required to engage the amygdala and uncover infant social behavior deficits. Circuit assessment showed dopaminergic VTA input to the amygdala was causally related to the social behavior deficits. Overall, these results suggest that the infant amygdala is vulnerable to social trauma and impacts circuits important in later life, although an acute stress can leverage infant social behavior as a biomarker of later-life pathology. SUPPORT: NIHR37HD083217

Swarovski MS, Lemmon D, Romero N, Conoscenti M, Brigidi S, Zelikowsky M (*Department of Neurobiology, U of Utah*) CA1 molecular signatures underlying context fear and fear renewal after extinction. ABSTRACT: The dorsal hippocampus (DH) has been shown to mediate contextual fear conditioning as well as tone fear renewal after extinction. While context conditioning involves the direct learning of a context-shock association, fear renewal requires a modulatory role for context in disambiguating the meaning of a trained tone. Despite these distinct roles for context in fear learning, little is known about how they are differentially encoded at the molecular level. It has been shown that experience drives induction of immediate-early gene (IEG) expression that produces heterogeneity within the same cell type specifically among pyramidal neurons in CA1 of the DH, an area important for spatial encoding. Additionally, inducible transcription factors (ITFs, a subset of IEGs) are thought to promote downstream gene programs that alter cellular functions within the CA1 circuit. We hypothesize that unique gene expression profiles in CA1 cellular ensembles differentially characterize context fear (CF) vs. fear renewal after extinction (FR). We tested this by first subjecting wildtype mice to either CF or FR training. CF mice were shocked (2s, 0.8 mA, 80s ITI) four times in context A, and tested for context freezing 24 hours later in context A. Control mice received the same exposures but without any footshock. FR mice were trained to fear a tone (20s, 80dB) co-terminating with a footshock (2s, 0.8mA) in context A, followed by extinction training in context B (comprised of 20 tones, 80dB, 20s duration, 40 sec ITI) for four days (1 session/day), and renewal testing in context A (4 tone presentations). Renewal control mice were tested for tone fear in context B. Thirty minutes following behavior, mice were sacrificed and brains were flash frozen, sectioned, and processed for highly multiplexed single molecule in situ hybridization (Hi-Plex) using 24 targeted probes against immediate early genes and genes that have been shown to be induced in response to experience or activity. Sections were imaged using 20x stitches of dorsal CA1 and the number of puncta/cell was quantified using Fiji. We identified genes that were induced uniformly across fear learning paradigms, as well as genes that uniquely marked each type of learning. Collectively, these data suggest that although CA1 plays a general role in spatial encoding, there are unique gene-expression signatures which underlie contextual learning that is direct (CF) vs. modulatory (FR).

Tashjian SM, Mobbs D (*Caltech*) Adaptive safety coding ABSTRACT: For humans, the inability to identify safety is a hallmark of anxiety, which is linked to poor health (Felger, 2018) and psychological outcomes (Mennin et al., 2005). Existing computational accounts fail to explain why individuals suffering from anxiety have difficulty identifying safety. This lack of understanding may be due, in part, to existing focus on external threat detection during Pavlovian conditioning. This perspective presumes that deficits in safety recognition are a result of threat overestimation, but fails to consider circumstances in which external threat is accurately identified, but safety is underestimated. Safety computations are hypothesized to reflect independent computations related to the self that mediate threat estimates (Tashjian et al., 2021). The current research tests a novel paradigm that partitions safety recognition into two main evaluative components: external threat and a self-oriented protection. Behavioral data was collected from 100 participants online (ages 18-40) demonstrating more accurate safety predictions when evaluating self-oriented sources of protection. fMRI data collected from a separate sample of 30 participants (ages 20-40) demonstrates dissociable neural systems that adaptively code changing safety contingencies depending on the source of safety. Searchlight results point to a role of the anterior vmPFC in detecting increasing safety, consistent with prior theoretical work (Tashjian et al., 2021). This work has important implications for understanding how different types of safety information are processed at the neural level in cognitive and reactive defensive circuits.

Taylor DL, Zelikowsky M (*U of Utah*) The effect of social instability on behavior and the brain ABSTRACT: Disruptions to social stability, including divorce, entrance into the foster care system, and, most recently, repeated COVID-associated school closures, represents a profound source of

chronic stress. Importantly, social instability has been shown to have long-lasting, negative effects on both socioemotional and physical health, nevertheless, the neurobiology underlying chronic social instability remains largely unknown. To assess the effects of social instability on behavior and the brain, mice (8-10 weeks) were randomly assigned to one of two groups: socially unstable (SiN, n = 9) or stable controls (CTL, n = 9), and were housed in groups of 3. Every 48 hours, mice in the SiN group were re-housed such that none of the cage mates were familiar with each other. The composition of SiN cages was changed 11 times over a 4-week period. Following at least 2 weeks of SiN experience, mice were tested for a variety of social, anxiety, and fear behaviors on assays including the 3-chamber social approach task, the resident intruder test, elevated zero maze, open field test, looming disk, tail suspension, and contextual fear conditioning. We found that one of the strongest effects of SiN was its propensity to enhance contextual fear. Indeed, SiN mice froze significantly more than CTL mice when tested on an 8-minute context fear test. This effect revealed that SiN mice are more susceptible to developing enhanced contextual fear as a result of social instability. We are currently testing the hypothesis that oxytocin signaling across the brain serves as a neural mechanism for the impact of social instability on behavior. Collectively, our findings highlight the importance of everyday social stability to the formation of adaptive fear responses.

Thomas CMP, Bouton ME, Green JT (*U of Vermont*) Inactivation of the prelimbic cortex eliminates ABA renewal in a stress acquisition context ABSTRACT: The prelimbic cortex (PL) is known to mediate the effects of context on instrumental behavior; however, demonstrations of the role of the PL have been primarily limited to physical contexts. We have recently shown a role for PL in supporting other types of contexts, including previous behaviors (Thomas et al., 2020) and satiety state (Thomas et al., in preparation) in guiding instrumental behavior. The current experiment was designed to test the hypothesis that the PL would likewise be necessary for a stress state to provide contextual control of an instrumental behavior. To that end, rats received lever press training sessions shortly after (2-4 min) exposure to a stressor ("context A"). Stressors varied by day (pedestal or oscillation on days 1 and 4, restraint on days 2 and 5, and foot shock on days 3 and 6) to avoid habituation. One stressor, either pedestal or oscillation, was reserved as each animals' future "test stressor" such that it was not associated with acquisition. Subsequently, the response was extinguished in the absence of stressor exposure ("context B"). ABA renewal was observed in control animals following exposure to their "test stressor", replicating Schepers and Bouton (2019) and demonstrating that stress state, rather than a specific stressor, promoted the renewal of the extinguished response. In contrast, inactivation of PL (bilateral infusion of 0.5 uL of 1.0 mM/0.1 mM baclofen/muscimol) eliminated stress state-related ABA renewal. These preliminary results suggest an expansion to the range of acquisition contexts (e.g., physical, stress, etc.) that the PL mediates and support the idea that the PL may act as a hub of context processing, where "context" is defined functionally rather than as simply a physical location.

Tronson, NC (*U Michigan*) - Neuroimmune activation, memory deficits and the risk of cognitive decline in the age of COVID-19 ABSTRACT: COVID-19 has affected more than 540 million individuals worldwide, and up to 40% of survivors experience post-acute covid sequelae (PASC, "long COVID"). The symptoms of long COVID include "brain fog" and cognitive impairments, and mood related symptoms such as depression or anxiety. SARS-COV-2 virus only rarely infects the brain, suggesting that other effects of COVID-19 causes memory impairments. Acute inflammation during illness is known to modulate memory, cognition, and mood; and many different illnesses cause subsequent syndromes including neurodegeneration and neuroinflammation-related damage. In our laboratory, we have demonstrated that effects of lipopolysaccharide and poly-IC- induced inflammation on memory and neuroimmune priming can last months after resolution of immune signaling. Yet, SARS-COV-2 is a single stranded RNA (ssRNA) virus, and triggers innate immune activation via Toll-Like Receptor (TLR) 7 and TLR8, a different pathway than LPS (Toll- like Receptor (TLR)-4) or PolyI:C (TLR3). In this project we examined the hypothesis that TLR7- triggered immune challenge causes lasting changes in the brain that contribute to the cognitive impairments and "brain fog" observed in Long COVID. We used a subchronic immune challenge protocol, previously developed in my laboratory, to determine whether the TLR7 agonist R848 causes memory impairments or anxiety- and depression-like phenotypes that emerge and persist at least in the months after a 2 week immune challenge. We observed a clear dose- response of cytokine and chemokine elevations in the hippocampus of young and mid-aged male and female mice; and mild weight loss to successive doses of R848, particularly in males. Eight weeks after immune challenge, we observed memory impairments in hippocampal- dependent novel object recognition in both sexes, and exaggerated neuroimmune response to subsequent challenge. These effects were not due to lingering elevations in neuroimmune activation or changes in locomotor activity. These findings are an initial step towards understanding how inflammatory sequelae of COVID-19 and other ssRNA viral illnesses contribute to cognitive effects of post-viral syndromes, and may, contribute to increased risk for aging-relate cognitive decline and Alzheimer's Disease in the decades to come. SUPPORT: Michigan Alzheimer's Disease Research Center/Michigan Institute for Clinical and Health Sciences/Claude D. Pepper Older Americans Independence

center Pilot Grant to NCT. Alzheimer's Association-NTF to NCT

Urcelay GP[1], **Hulley T**[1], **Alcalá JA**[2] ([1]*School of Psychology, U of Nottingham, UK;* [2]*Faculty of Psychology, Complutense U of Madrid, Spain*) Uncertainty increases generalization of human predictive learning ABSTRACT: In this experiment, we wanted to assess generalization gradients as a function of outcome probability in human predictive learning. Participants (N=180) were recruited through Prolific and experienced differential conditioning of two Gabor patches (90° and 0°, counterbalanced) with a fictitious outcome (i.e., shock). The 3 groups differed during training in terms of the probability that the CS+ was followed by the outcome (100%, 50%, 25% of CS+ trials). Following 12 training trials with each CS, all participants were tested with the trained stimuli (90° and 0°) and generalization stimuli (GS) at intermediate values (15°, 30°, 45°, 60°, 75°, 105°, 120°, 135°, 150°, 165°). During training, participants in Group 100% learned the discrimination between CS+ and CS- better than Group 50%, which in turn learned it better than Group 25%. The results during training thus scale with the different probabilities that were scheduled for each group. During test, shock expectancy judgements for GS near the CS+ also scaled with training probabilities, in that Group 100% expected the outcome more than Group 50%, and the latter expected it more than Group 25%. However, shock expectancy for GS near the CS- revealed more responding in Group 25% than in Group 100%, a finding that challenges standard associative theories of learning. We will discuss potential explanations for these findings. SUPPORT: Funded by UK ESRC Grant (ES/R011494/2) awarded to GPU and JP

Valyear MD (*McGill U*) Phenotypic differences in what is learned about discrete alcohol cues and contexts ABSTRACT: Environmental stimuli that predict alcohol influence the vigour with which alcohol is pursued. Consider the visual and orosensory properties of a preferred alcoholic beverage that serve as discrete alcohol cues, and the ambience of a regular alcohol-drinking environment like a bar that serves as an alcohol context. Dr. Nadia Chaudhri developed a conditioning procedure to study the influence that discrete alcohol cues have on Pavlovian responding for alcohol when discrete cues are presented in alcohol contexts and neutral contexts. First, rats are familiarized with the taste and pharmacological effects of alcohol (15% ethanol) in their homecage. Then, rats receive Pavlovian conditioning sessions in which a 10 s conditioned stimulus (CS; 15/session) is paired with alcohol delivery (0.2 ml/CS, 3 ml/session) into a fluid port for oral consumption. Pavlovian conditioning sessions are conducted every other day in a unique alcohol context. On days intervening Pavlovian conditioning sessions rats are placed in a distinct neutral context wherein alcohol is never available. After an equal number of sessions in the alcohol and neutral contexts, the CS is presented without alcohol in the alcohol context and the neutral context to test conditioned responding for alcohol. At test, rats generally make significantly more port entries during the CS in the alcohol context than in the neutral context. The elevation in CS port entries that occurs in the alcohol context relative to the neutral context at test can be explained by at least two different learning processes: 1) summation of two independent associations between the discrete alcohol CS, the alcohol context, and alcohol or 2) the alcohol context serving as an occasion setter to modulate the association between the discrete CS and alcohol. Here, we compiled data from various replicates of the Pavlovian conditioning with context alternation procedure and used a hierarchical clustering approach to examine naturalistic groupings of rats based on the number of port entries they made at test during the CS, and between CS presentations (NonCS port entries), in the alcohol and neutral contexts. A notable divergence occurred when three clusters emerged. One cluster, termed Context Learners, showed elevated CS and NonCS port entries in the alcohol context relative to the neutral context, consistent with summation. Another cluster, termed Occasion Learners, showed elevated CS port entries in the alcohol context relative to the neutral context, but similar NonCS port entries in both contexts, consistent with occasion setting. The final group, termed Discrete Learners, showed a similar number of CS and NonCS port entries irrespective of context, consistent with learning the discrete CS-alcohol association. Altogether, rats differ in their propensity to learn about discrete alcohol cues and contexts, or the relationship between discrete cues and alcohol contexts. As such, alcohol contexts, discrete alcohol cues, and their combination may influence behaviour by recruiting distinct neural circuits that subserve separate learning processes across rats. Considering the learning processes that prominently control responding for alcohol in individual people may have implications for devising efficacious treatments for alcohol use disorder. SUPPORT: CIHR, FRQS

Vasudevan K, Ramanathan KR, Vierkant V, Maren S (*Texas A&M U*) Nucleus reuniens inactivation does not impair consolidation or reconsolidation of fear extinction ABSTRACT: Recent data reveal that the thalamic nucleus reuniens (RE) has a critical role in the extinction of conditioned fear. Muscimol (MUS) infusions into the RE impair within-session extinction of conditioned freezing and result in poor long-term extinction memories in rats. Although this suggests that RE inactivation impairs extinction learning, it is also possible that it is involved in the consolidation of extinction memories. To examine this possibility, we examined the effects of RE inactivation on the consolidation and reconsolidation of fear extinction in male and female rats. Twenty-four hours after auditory fear conditioning, rats underwent an extinction procedure (45 CS-alone trials) in a novel context and were infused with saline (SAL) or MUS within minutes of the final extinction trial. Twenty-four hours later, condi-

tioned freezing to the extinguished CS was assessed in the extinction context. Postextinction inactivation of the RE did not affect extinction retrieval. In a second experiment, rats underwent extinction training and, 24 h later, were presented with a single CS to reactivate the extinction memory; rats were infused with SAL or MUS immediately after the reactivation session. Pharmacological inactivation of the RE did not affect conditioned freezing measured in a drug-free retrieval test the following day. Importantly, we found in a subsequent test that MUS infusions immediately before retrieval testing increased conditioned freezing and impaired extinction retrieval, as we have previously reported. These results indicate that although RE inactivation impairs the expression of extinction, it does not impair either the consolidation or reconsolidation of extinction memories. We conclude that the RE may have a critical role in suppressing context-inappropriate fear memories in the extinction context. SUPPORT: This study was supported by National Institutes of Health grants R01MH065961 and R01MH117852.

Vercammen L, Beckers T, Luyten L, Vervliet B (*KU Leuven*) The rewarding properties of safety signals as established by a two-way active avoidance task in rats. ABSTRACT: Safety signals (SSs) are thought to acquire rewarding properties when they are presented contingent with the omission of expected aversive events, as occurs when rats make an avoidance response. However, to what extent SSs are functionally similar to rewards is not entirely clear, given that only a handful of studies assessed the properties of safety signals in the context of avoidance. In the current study, we aimed to probe the rewarding properties of a SS, by evaluating whether rats show approach behavior towards this SS after five days of avoidance training. Twenty-eight male adult Wistar rats were subjected to five consecutive avoidance training sessions, each consisting of 30 pairings of a conditioned stimulus (CS; 3 kHz tone, 75 dB, 20 s) with an unconditioned stimulus (US; 0.6 mA foot shock, 10 s). Rats had the possibility to avoid the foot shocks by shuttling to a neighboring safe compartment during CS presentation, upon which a safety signal was presented (i.e., turning on of the house light for 5 seconds). Twenty four hours after the final avoidance training session, the rats were subjected to a 10-minute compartment-preference (CP) training session, followed by a 5-minute CP test the next day. We found that rats spent significantly more time in the compartment that was paired with the SS, compared to the compartment where no stimulus was present. We observed this during both CP training and CP test. These findings suggest that SSs trigger approach behavior, similar to what has been observed for cues that predict natural rewards, thus suggesting that stimuli that are presented contingent upon an avoidance response acquire rewarding properties.

Wachter SL, Parker KL, Freeman JH (*The U of Iowa*) Disconnection of cerebellar communication with the prefrontal cortex causes deficits in executive function in rats ABSTRACT: Research on humans who have suffered from posterior cerebellar stroke indicates that the cerebellum plays a role in executive functions such as task switching, inhibition, spatial learning, and working memory (Schmahmann, & Sherman, 1998). However, results indicating deficits in executive function and working memory in human patients needs to be interpreted very cautiously due to variability in location and extent of damage. In rats, the posterior lobules of the cerebellum, particularly Crus I/II, have bidirectional communication with forebrain structures (e.g., infralimbic, prelimbic and orbital cortices). Tracing studies indicate that Crus I/II have strong connectivity with the lateral cerebellar nuclei (LCN), indicating that communication from Crus I/II to forebrain structures goes through the LCN. To investigate the role that the cerebellum plays in executive functioning and working memory, we disrupted the communication between Crus I/II and forebrain structures with bilateral electrolytic lesions of the LCN. The rats were then tested in a rule-based category learning task, a feature negative discrimination task, a radial arm maze (RAM) working memory task and a RAM reference memory task. The tasks were selected because they require the prefrontal cortex in rodents. Preliminary data indicate that there are differences in performance in several of the tasks, specifically, the rule-based category learning task, feature negative discrimination task, and the RAM working memory task. In addition, in both RAM tasks, lesioned rats use non-spatial and non-working memory strategies more often than controls.

Wang Z, Moore S, Sun R, Lee A, Zhu Z, Charles A, Kuchibhotla K (*Johns Hopkins U*) Slow or sudden: revealing naturalistic transitions to habitual behavior during learning ABSTRACT: Animals use different decision processes to efficiently adapt to complex environments. When operating in a goal-directed mode, animals deliberate amongst alternatives based on their motivation. As the statistics of the environment become predictable, animals can shift to a more automatic habit mode. The transition from goal-directed to habitual decision-making during learning is thought to be gradual, yet permanent. Current approaches for distinguishing between the two decision processes require discrete 'test' sessions that preclude assessment of the nature, timing and properties of transitions. Here, we devised a naturalistic devaluation approach to assess the underlying decision mode en passant, without discrete 'test' sessions. Mice motivated by a taste preference for normal water (provided during the task) versus mice with ad libitum access to water doped with citric acid initially exhibited naturalistic fluctuations in the cue-driven response rate during discriminative instrumental learning. Then, abruptly and overnight, this variability ceased, and mice transitioned to a high and stable response rate. To test whether habit-like transitions were smooth or state-based fluctuations we applied a hidden Markov model

to our dataset and found that during the goal-directed phase, distinct states best described action rate fluctuations, while post-transition, the behavior abruptly switched to a single state. The sudden transitions were also accompanied by signatures of automaticity in lick microstructures. Surprisingly, some animals reverted to goal-directed mode after several sessions in habit mode suggesting that transitions to habitual decision-making are not intrinsically permanent. Ongoing work aims to isolate pupil-based biomarkers of goal-directed and habitual behavior and to identify the role of the dorsolateral striatum (DLS) in guiding these abrupt transitions. Thus, naturalistic devaluation provides a powerful en passant approach to study habit formation and shows that transitions to habitual decision-making are strikingly abrupt, but also reversible.

Weaver C, Pajser A, Fisher H, Pickens C (*Kansas State U*) Repeated exposure to amphetamine does not lead to long-term alterations in omission contingency learning ABSTRACT: In omission contingency training, rodents learn to suppress their natural tendency to approach or touch a reward-predictive cue (termed "autoshaping" or "sign-tracking" responses) if the approach/touching responses lead to the omission of the reward. Previous research in our laboratory suggested that adolescent access to alcohol (through voluntary drinking) or forced injections of alcohol in both adolescence or adulthood would alter the rate of omission contingency learning. Notably, the pattern with the forced injections of alcohol suggested that there was a biphasic dose-response in which a moderate dose impaired omission contingency learning and a higher dose had no effect. We conducted the current experiment to determine if amphetamine exposure would also alter behavior in the omission contingency task in order to assess the generality of the pattern observed with alcohol. Amphetamine has effects on the dorsal and ventral striatum regions shown to modulate the rate of omission contingency learning. We gave rats once-daily injections of 0, 1, 2, or 4 mg/kg amphetamine for 7 days in adulthood and then trained them in the autoshaping and omission contingency learning tasks starting 2 weeks after the final injection. However, we found no effect of any amphetamine dose on omission contingency learning. During my poster, I will discuss possible causes of this null effect and possible interpretations of why the effects of alcohol might be stronger than those of amphetamine.

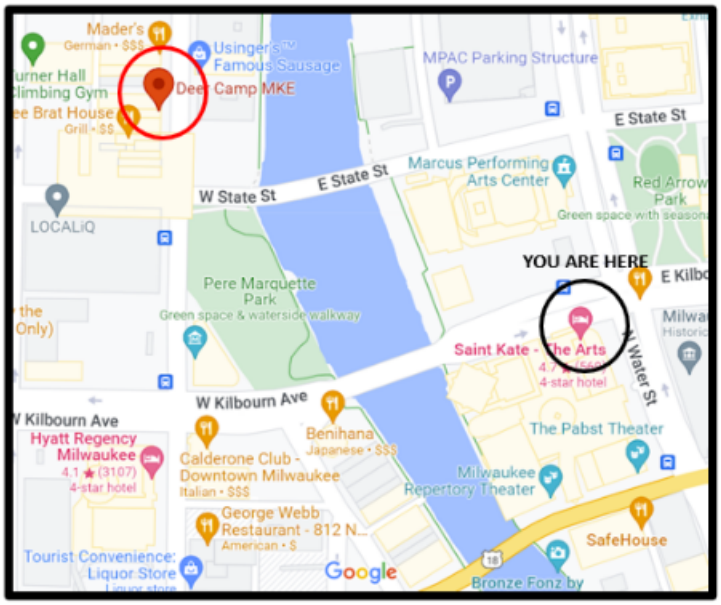
Wilson WJ (*Albion College, Frontière Astrophotography*) Learning continues post-retirement: Trying hard to make this relevant to the Society. ABSTRACT: After retiring from my academic position I planned to continue my photography. COVID shut down what had been my preferred targets (rodeo and live music) so I turned to my life-long interest in astronomy and started shooting astrophotography. Poster presents a brief description of this journey, evidence of improvement with practice, and many recent images. tl;dr: Astrophotography is far different from the photography I had done previously. SUPPORT: My TIAA retirement acct provides funding; Frontière Farm House graciously provides a site for my work.

Wood KC, Angeloni CF, Oxman K, Clopath C, Geffen MN (*U of Pennsylvania*) A cortico-thalamic circuit for auditory associative learning ABSTRACT: Learning to avoid dangerous signals while preserving normal responses to safe stimuli is essential for everyday behavior and survival. Following identical experiences, subjects exhibit fear specificity ranging from high (specializing fear to only the dangerous stimulus) to low (generalizing fear to safe stimuli), yet the neuronal basis of fear specificity remains unknown. Here, we identified the neuronal code that underlies inter-subject variability in fear specificity using longitudinal imaging of neuronal activity before and after differential fear conditioning in the auditory cortex of mice. Neuronal activity prior to, but not after learning predicted the level of specificity following fear conditioning across subjects. Stimulus representation in auditory cortex was reorganized following conditioning. However, the reorganized neuronal activity did not relate to the specificity of learning. These results present a novel neuronal code that determines individual patterns in learning.

WIL Luncheon location

12th Annual Women in Learning Luncheon Directions

Join WIL at Deer Camp MKE from 12 pm – 1:30 pm on Saturday, October 1st with Distinguished Guest Speaker Dr. Susan Sangha.



Deer Camp MKE address: 1023 Old World Third St (AKA North Doctor MLK Jr Drive)

Directions to Deer Camp from the Saint Kate Hotel (5-10 min walk):

1. Head West on E. Kilbourn Ave toward Milwaukee River (350 ft)
2. Turn right onto N. Riverwalk Way (500 ft)
3. Turn left onto E. State St (bridge over Milwaukee River) (0.1 mi)
4. Turn right onto Old World Third St/ N. Doctor MLK Jr Drive (200 ft)
5. Deer Camp will be on your left