24th International

C. elegans

Conference

June 24-28, 2023 | Glasgow, Scotland

ABSTRACT BOOK

GENETICS GSA G3 Genes | Genomes | Genetics

Mutations in the GBA gene in humans are a common risk factor for Parkinson's disease. Therefore, we are generating a quadruple deletion of the four GBA orthologs in *C. elegans*, three of which are upregulated by *B. subtilis*. Furthermore, we are also testing deletions of all 26 upregulated genes for their involvement in inhibiting α-synuclein aggregation.

In a parallel and complementary approach, we are working on uncovering the bacterial signals and metabolites that trigger the protective effect (see poster by Deep Prakash *et al*).

References

Goya M.E. *et al.*, Probiotic *Bacillus subtilis* Protects against α-Synuclein Aggregation in *C. elegans*. Cell reports 30, 367-380.e367. (2020).

Van Ham T.J. *et al.*, *C. elegans* model identifies genetic modifiers of α-synuclein inclusion formation during aging. PLoS genetics 4, e1000027. (2008)

998C **The neuropeptide receptor *npr-38* regulates avoidance and recovery sleep** Emily Le[1], Caroline Curtin[1], Madison Honer[2], Teagan McCarthy[1], Jonathan Fingerut[1], Matthew Nelson[1]1Biology, Saint Joseph's University, [2]Lewis Katz School of Medicine

C. elegans demonstrates protective avoidance behavior in the face of threats. When exposure to these noxious stressors is unavoidable, cellular damage and injury is incurred. Stress-induced sleep promotes recovery by redistributing resources towards cellular repair. How *C. elegans* coordinates a state of heightened arousal (i.e. avoidance) with that of recovery sleep is unclear. We have identified *npr-38*, a G-coupled protein neuropeptide receptor, as a key component of these dynamics. Loss-of-function of *npr-38* disrupts avoidance and reduces stress-induced sleep. *npr-38* mutants also show reduced activity outside of sleep, evident by reduced spontaneous movement and arousal threshold, but move normally when stimulated with harsh touch. Using cell-specific rescue of *npr-38* and degradation of AID-tagged NPR-38 protein, we identified the ADL sensory neurons as the primary site of action for avoidance and sleep, and the RIS and DVA for arousal. Genetic ablation of the ADL neurons increases sleep, while optogenetic activation reduces it. Multi-copy over-expression of *npr-38* shortens the avoidance phase, resulting in early sleep and premature waking. While the ADL neurons are glutamatergic, we identified *nlp-50* and *flp-25* neuropeptides as being wake-promoting and likely released from the ADL. Based on our data and the connectivity, we propose a model in which the ADL neurons inhibit sleep neurons (i.e., ALA and RIS) to promote heightened arousal during the avoidance phase. *npr-38* then inactivates the ADL neurons to ensure that stress-induced sleep is initiated and terminated at the appropriate times.

999V **Single-cell profiling reveals striking diversity within adult *C. elegans* motor neurons and new functions for a terminal selector gene** Jayson J Smith[1], Seth R Taylor[2], David M Miller[2], Paschalis Kratsios[1]1Department of Neurobiology, University of Chicago, [2]Department of Cell and Developmental Biology, Vanderbilt University

To reveal gene regulatory mechanisms that define neuronal identity and maintenance, we used single-cell RNA-sequencing (scRNA-seq) to profile motor neurons in the adult *C. elegans* ventral nerve cord. We find that eight motor neuron (MN) classes, previously defined by anatomical criteria, subdivide into 29 distinct subclasses delineated by unique Hox and neuropeptide expression codes. Profiling of adult cholinergic MN subclasses lacking the terminal selector *unc-3* (Collier/Olf/EBF), ortholog of the neurodevelopmental syndrome-causing gene *EBF3*, revealed differential responses; eleven subclasses collapse into seven groups, each acquiring new, distinct molecular features, such as alternative neurotransmitter identities. Intriguingly, loss of *unc-3* in cholinergic MNs also disrupts the transcriptomes of post-synaptic GABAergic neurons. Electron microscopy reconstruction suggests that altered gene expression in GABA MNs perturbs connectivity. Thus, unbiased molecular profiling at single-cell resolution uncovered striking diversity within adult MNs, as well as cell-autonomous and indirect mechanisms through which a terminal selector gene sculpts neuronal identity.

1000V ***Caenorhabditis elegans* betaine-sensitive nicotinic receptors: molecular function and physiological roles** Ornella Turani, Guillermina Hernando, Noelia Rodriguez Araujo, Cecilia Bouzat‌I NIBIBB-CONICET-UNS

Caenorhabditis elegans possesses an extensive and diverse family of nicotinic receptors (nAChR), many of which remained uncharacterized. nAChRs are involved in worm locomotion and are targets of anthelmintic drugs. Parasitic nematodes have acquired resistance to most anthelmintic drugs, thus generating problems in human and animal health. Because of this, the identification of novel drugs and targets is required. The potent nematocidal drug monepantel (MNP), which belongs to the recently discovered class of compounds amino-acetonitrile derivatives (AADs), has been shown to target ACR-23 nAChR. ACR-23, whose endogenous agonist is betaine (BE), is a poorly characterized nAChR present in body-wall muscle and mechanosensory neurons of nematodes. Since it is not conserved in vertebrates, ACR-23 is an interesting pharmacological target for anthelmintic drugs. Our goal is to decipher ACR-23 molecular function and its potential as a novel anthelmintic drug target. By performing locomotion assays with wild-type adult worms we showed that exogenous BE significantly increased worm motility. This effect was not observed in *acr-23* mutants, indicating that the hypermotility is mediated by ACR-23. The exposure of worms to MNP produced the opposite effect, resulting in reduced motility as a function of concentration (EC_{50} = 50 µM). MNP induced spastic

paralysis and inhibited egg hatching, indicating important anthelmintic ability. Locomotion assays with mutant worms demonstrated that MNP-induced paralysis is mediated by ACR-23 and DEG-3/DES-2, a nAChR present in sensory neurons involved in nociception and chemotaxis. By patch-clamp recordings from cultured *C. elegans* L1 muscle cells, we described for the first time the properties of BE-elicited single-channel and macroscopic currents. Our study provides novel information aiming at the elucidation of the molecular function and pharmacology of the nAChR family. It also contributes to the understanding of the molecular basis of anthelmintic action, which paves the way for the development of novel drugs.

1001V **Major sperm proteins expressed in ADL chemosensory neurons require the NRDE-3 somatic nuclear RNAi pathway** Maria C. Ow[1], Abdul Rouf Dar[2], Rebecca A. Butcher[2], Sarah E. Hall[1,11]Syracuse University, [2]University of Florida

Environmental conditions experienced early in the life of an animal can result in gene expression changes during adulthood. We have previously shown that *C. elegans* animals that experienced the developmentally arrested and stress resistant dauer stage (postdauers) retain a cellular memory of early-life stress that manifests during adulthood as genome-wide changes in gene expression, chromatin states, and altered life history traits. One consequence of developmental reprogramming in *C. elegans* postdauer adults is the downregulation of expression of a TRPV channel gene, *osm-9*, in the ADL chemosensory neurons which results in the reduced avoidance to a pheromone component, ascr#3. This reduction in ascr#3 avoidance requires the somatic nuclear RNAi pathway.

To investigate the role of the somatic nuclear RNAi pathway in regulating the developmental reprogramming of *osm-9* in ADL due to early-life stress, we profiled the mRNA transcriptome of control and postdauer ADL in wild-type and *nrde-3* mutant adults. We find that the wild-type ADL transcriptome expresses germline-expressed genes. NRDE-3, the effector of the somatic nuclear RNAi pathway, plays a critical role in regulating the expression of germline-expressed genes in ADL neurons, such as major sperm proteins (MSPs) genes, even under non-stressful growth conditions. Loss of MSPs function, through mutation of the *gsp-3* and *gsp-4* sperm-specific PP1 phosphatases, results in the abrogation of ascr#3 avoidance and aberrant olfactory behavior. We also show that an Argonaute pseudogene, *y49f6a.1* (*wago-11*), is expressed in ADL and is required for ADL chemosensory function. Overall, our results suggest that small RNAs and reproductive genes program the ADL mRNA transcriptome during their developmental history and highlight a nexus between neuronal and reproductive networks in calibrating animal neuroplasticity.

1002V **ALK/SCD-2-dependent expression of DAF-7 from the ASJ neurons couples bacterial food ingestion to foraging state dynamics in *C. elegans*** Sonia Boor[1,2], Joshua Meisel[3], Dennis Kim[11]Boston Children's Hospital, [2]Biology, MIT, [3]MGH

Animal internal state is modulated by nutrient intake, resulting in behavioral responses to changing food conditions. Whereas neuronal circuitry responsive to changes in food availability has been increasingly characterized, less is known about the role for changes in neuronal gene expression in shaping internal state. *C. elegans* exhibits a food-dependent two-state foraging behavior that alternates between exploration and exploitation behavioral states known as roaming and dwelling. Here, we identified a role for the *C. elegans* ortholog of Anaplastic Lymphoma Kinase (ALK), SCD-2, in the regulation of a neuroendocrine gene expression loop that couples the ingestion of bacterial food to the dynamics of foraging behavior. We showed that ALK/SCD-2 controls the expression of the neuronal TGF-beta, DAF-7, in the ASJ chemosensory neurons, which we found to be inhibited by the ingestion of bacterial food. In turn, we determined that DAF-7 expression from the ASJ neurons promotes roaming state behavior by extending the duration of the roaming period. Our data reveal a pivotal role for ALK/SCD-2 regulation of food-dependent, dynamic DAF-7 expression that functions in a physiological positive-feedback loop that facilitates behavioral state changes in response to changing food conditions.

1003V **Neuronal FMRFamide neuropeptide signaling controls the activation of the head mesodermal cell (hmc) during a rhythmic behavior in *C. elegans*** Mingxi Hu, Ukjin Choi, Derek SieburthZilkha Neurogenetic Institute

FMRFamides are an evolutionarily conserved family of neuropeptides that are highly expressed in the nervous system and play important roles in behavior, energy balance and reproduction. Here, we show that FMRFamide signaling is critical for the anterior body wall muscle contraction (aBoc) step of the defecation motor program (DMP), and functions by controlling the generation of calcium responses in a single cell of previously unknown function, hmc. FLP-22 is released from a bifunctional motor neuron AVL in response to pacemaker signaling and activates the G protein coupled receptor (GPCR), FRPR-17, in hmc. FRPR-17 activates a G alpha s-protein kinase A (PKA) signaling cascade in hmc, leading to the generation of a single large calcium transient in hmc every 50 seconds that occurs in phase with AVL activation and aBoc. Genetic ablation of hmc results in missing aBoc steps during the DMP. Similarly, *flp-22* or *frpr-17* mutations leads to missing aBocs and to the near absence of calcium transients in hmc, and expression of FRPR-17 selectively in hmc restores normal aBoc and hmc calcium transient frequency in *frpr-17* mutants. hmc itself is not contractile but is functionally coupled to neck muscles through gap junctions composed of UNC-9/innexin. aBoc and hmc activation are inhibited by signaling from a second FMRFamide-like neuropeptide, FLP-9, which functions through its GPCR, FRPR-21, in hmc. Overexpressing FLP-9 eliminates aBoc and hmc activation, whereas *flp-9* or *frpr-21* mutations restore aBoc