2021 CILIATE MOLECULAR BIOLOGY MEETING July 19-22, 2021 (on Zoom)

Organizers (Ciliate Advisory Board):

Mireille Betermier (mireille.betermier@i2bc.paris-saclay.fr), Paris-Saclay Univ., France Marcella Cervantes (marcella.cervantes@gmail.com), Albion College, USA Jacek Gaertig (jgaertig@uga.edu), University of Georgia, USA Sabrice Guerrier (SGUERRIER@Rollins.edu), Rollins College, USA Chad Pearson (CHAD.PEARSON@CUANSCHUTZ.EDU), Univ. of Colorado Anschutz, USA Sean Taverna (staverna@jhmi.edu), John Hopkins University, USA

ZOOM CONNECTION:

Join Zoom Meeting

https://jhjhm.zoom.us/j/99389037092?pwd=ckVFQXIEZFZLYTJVZzdLdFM1VHdjQT09

Meeting ID: 993 8903 7092 Passcode: 637522

POSTER VIEW:

Please use this link to asynchronously view posters. All posters should be online starting on July 16, 2021 but there are already a few posters up.

https://posters.com/posters/cmbc/posters.cgi/Login/current

NOTE: Sychronous poster sessions will occur in Zoom breakout rooms established during the meeting.

EASTERN DAYLIGHT TIME (EDT)

| Monday July 19, 2021 | |
|------------------------------|--|
| Session 1 10:00AM-11:00AM | Chair: Mireille Betermier (mireille.betermier@i2bc.paris-saclay.fr), I2BC-CNRS, Paris-Saclay Univ., France |
| 10:00AM | Julien Bischerour (julien.bischerour@i2bc.paris-saclay.fr) I2BC-CNRS, France "Coupling of Pgm with Ku DNA repair protein ensures accurate genome wide DNA rearrangement in Paramecium tetraurelia" |
| 10:15AM | Salman Shehzada (salman.shehzada@igh.cnrs.fr), IGH, CNRS, France "A SUMO E3 ligase is required for scnRNA selection and DNA elimination in Tetrahymena" |

other HP1-like proteins for the heterochromatin establishment and how Hpl8p facilitates DNA elimination.

Poster 1.10

Using Tetrahymena to study the molecular mechanisms of the contractile vacuole <u>Cheng, Chao-Yin</u>*, cycheng@uchicago.com; Turkewitz, Aaron, apturkew@uchicago.edu

The University of Chicago, USA

The contractile vacuole (CV) is an osmoregulatory organelle that periodically and regularly expels water from the cell. CVs, which are sometimes described as cellular kidneys, are widely distributed in protists but poorly understood at the level of assembly, maintenance, and the mechanisms that underlie cyclic contraction. We propose to explore CV mechanisms by using T. thermophila, and have initially focused on a small set of genes that are important for CV function. VPS8D encodes a subunit of a CORVET tether complex that is strongly localized to CVs. VPS8D knockdown mutants show drastically reduced survival after osmotic shock, consistent with an osmoregulatory function. Live cell imaging of cells expressing endogenously mNeon-tagged Vps8dp reveals features of the organization and dynamics of CVs, and provides evidence that Vps8dp accumulates at sites of membrane fusion. A second gene is DOP1, the T thermophila ortholog of a protein involved in endosomal remodeling in yeast and mammals. Previous studies showed that DOP1 deletion in T. thermophila results in changes in CV morphology and osmotic hypersensitivity. Dop1p shows tight localization to CVs, but in a pattern distinct from Vps8dp. Ongoing experiments aim to elucidate how VPS8D and DOP1 contribute to membrane trafficking that underlies CV function. Additional CV-related genes are being sought using both proteomic approaches and transcriptional profiling.

Poster 1.11

Cryo-electron tomography of Drosophila sperm flagella

<u>**Gui, Long**</u>*, long.gui@utsouthwestern.edu; Fu, Gang, gang.fu@utsouthwestern.edu; Zhao, Yanhe, yanhe.zhao@utsouthwestern.edu; Ugrankar, Rupali, rupali.ugrankar@utsouthwestern.edu; Henne, Mike, mike.henne@utsouthwestern.edu; Nicastro, Daniela, daniela.nicastro@utsouthwestern.edu *University of Texas Southwestern Medical Center, USA*

The axoneme, which is longitudinally composed of 96 nm repeated units, is the core structure of cilia. Drosophila has unusually long sperm (~1.8 mm) but the high-resolution structure of sperm axoneme remains unknown. Here, using cryo-electron tomography and sub-tomogram averaging we characterized the native 3D structure of Drosophila sperm axoneme, which shares conserved regulatory complexes but also possesses Drosophila-specific features. Structural analyses of the Drosophila ASP-RNAi mutant versus wild-type reveal that depletion of ASP resulted in assembly defects of the outer dynein arms (ODA) and thus lead to immobile sperm and male sterile. Our results suggest Droposhila sperm as a novel model system to study the structure of function of ciliary proteins.

Poster 1.12

Variety of sexual processes and the new case of self-fertilization in Paramecium <u>Maksim Melekhin</u> (1,2), maksim.s.melekhin@gmail.com*; Irina Nekrasova (1), neirina@yandex.ru; Alexey Potekhin (1,2), loxodes@list.ru

Sex has arisen together with eukaryotic cell and since that time living organisms, especially protists, developed an uncountable variety of sexual systems. Even within some small taxa diversity of sexual systems may be amazing. Paramecium is a wellknown ciliate, and cytology and genetics of its sexual processes are well studied for several species. Conjugation between cells of complementary mating types is a traditional way of sexual genetic exchange, some species also have self-fertilization known as autogamy, and sometimes also intrastrain conjugation (selfing) occurs. Selfing and autogamy may take place in different Paramecium species scattered along the genus phylogenetic tree. However, for non-model species, the biggest challenge is an experimental conjugation assay design. Paramecium putrinum belongs to Helianter subgenus, phylogenetically basal to other paramecia, and it has a system of multiple mating types (Jankowski, 1969). Working with more than twenty P. putrinum strains we managed to clarify intraspecies phylogenetic relationships and to establish the mating protocol. We found that specifically for P. putrinum autogamy may be induced by initial mating reaction, and we described cytological features of both conjugation and autogamy in P. putrinum, being significantly different from other Paramecium species. It is the first case of autogamy in Helianter subgenus, which may help to understand genetics and evolution of sexual processes in Paramecium.

Poster 1.13

The MAC genome of Blepharisma stoltei

<u>Singh, Minakshi</u>, minakshi.singh@tuebingen.mpg.de*; Seah, Kwee Boon Brandon, kb.seah@tuebingen.mpg.de; Emmerich, Christiane,

christiane.emmerich@tuebingen.mpg.de; Singh, Aditi, aditi.singh@tuebingen.mpg.de; Ortenzi, Claudio, claudio.ortenzi@unimc.it; Buonanno, Federico,

federico.buonanno@unimc.it; Sugiura, Mayumi, msugi@cc.nara-wu.ac.jp; Harumoto, Terue, harumoto@cc.nara-wu.ac.jp; Swart, Estienne,

estienne.swart@tuebingen.mpg.de;

Max Planck Institute for Developmental Biology, Germany

Blepharisma is a distinctive genus of ciliates known for the pink, light-sensitive pigment.. Blepharisma belongs to class Heterotrichea, which together with class Kayorelictea, represent the earliest diverging pair of ciliate lineages. Among ciliates, Blepharisma is of special interest due to its alternative pathways of macronuclear development. Here we present the first investigations of genomic and transcriptomic resources for Blepharisma stoltei. With the benefit of long read sequencing we find that, like Paramecium bursaria, the Blepharisma macronuclear genome is organized as mini-chromosomes. Like Stentor coeruleus, Blepharisma has tiny introns, 15 or 16 nt – the shortest known spliceosomal introns. Using HMMER searches of PFAM, we found several genes with transposase domains in the MAC genome, among which were homologs of known ciliate transposases. We report our analyses of possible functionality/catalytic potential of these transposases, and their expression during development. We also report our analyses of additional genes involved in small RNA production. These analyses show that Blepharisma may serve as a very useful model for genome reorganization, complementing those in model ciliates.