Summary: 2nd Automated Analysis of Ageing Workshop

October 25-26th 2018
CRG Barcelona, Spain
Charles Darwin room – Ground level – PRBB Building

Zachary Pincus kicked off the workshop describing the C. elegans culture system he developed to support the observation of freely moving, individual worms through their lives. Briefly, this system enclosures single worms in droplets of bacteria between a PEG surface and a thin PDMS layer. Zach mentioned that the speed and accuracy of autofocus could be improved by adding beads to each chamber—beads too large to be swallowed by C. elegans but small enough to provide a good subject for focusing. In his second talk Zach discussed his group's ongoing work into miRNA as predictors of longevity in *C. elegans*, as well as the relationship between behavioral and morphometric changes ("healthspan") with lifespan across different mutant strains.

Benjamin Towbin spoke about his work studying evolutionary trade-offs in aging. He described his system involving GFP tagged ribosomes to characterize the relationship between early-life ribosome biogenesis and development and fertility.

Andre Brown spoke about the worm tracking system developed by his group and his approach for making and interpreting high-dimensional measurements of worm behavior – posture, aggregation and swarming. For these behavioral assays, the preparation time per is approximately equal to the length of an imaging run, meaning that even in large screens robotic arms are not necessary, as personnel are not limited by using a single microscope setup. Multiple microscopes are key to throughput. In his second talk, he focused on approaches for studying traits that depend on many genetic loci.

David Weinkove spoke about his automated measurements of healthspan, and his system's application for performing drug screens. David announced a start-up company, Magnitude Biosciences.

Julian Ceron described his new approach to CRISPR- Nested CRISPR for introducing mutations in *C. elegans* genome. By adopting a nested GFP integration strategy, much higher integration rates can be obtained compared to other methods. Julian's group's method is competitive with the new overhang-based CRISPR method developed by the Mello lab, with the major difference being that Julian's method does not require large oligos.

Stephen Banse described his new microfluidic systems for performing stress assays on flatbed scanners. He described the CITP consortium's recent progress, with a focus on sources of variability in experiments. Remarkably, plate-to-plate differences in lifespan within a single experiment appear to be consistently larger than between-lab differences in lifespan. It would appear something important varies between individual plates in a single run. This result agrees with many other attendee's experience.

Nicholas Stroustrup discussed a new version of the lifespan machine. Marquee features include improved, fully-automated death time calling, a 4x decrease in image file storage requirements, and image analysis runs on institutional high-performance computing clusters. The image capture server now runs on small ~$100 "Raspberry Pi" computers. New labs, instead of having to set up and configure the system themselves, the idea is that we simply mail them an SD card with everything pre-configured.

Ben Lehner presented his published work looking at the effect of mechanical constriction on the position of cells during the early development of C. elegans embryos. A particular cell moves at a specific time during development to compensate for early squishing. This squishing is likely physiologically relevant—mimicking the compression of embryos in the *C. elegans* uterus.

Thomas Wilhelm described his screen for RNAi constructs that extend lifespan when applied very late in life. *bec-2* appears to be very interesting in this regard. Notably, his screen involved culturing *C. elegans* on a fine mesh, allowing him to serially dunk populations into media containing different RNAi constructs.

Samantha Edwards described the H2020 *C. elegans* healthspan project, and her ongoing work studying neuropeptides in aging. She noted that users should be careful working with some neuron-specific SID-1 strains, in which SID-1 appears to be expressed in more tissues than previously recognized.

In the final discussion, all groups expressed a frustration with the apparent plate-to-plate and replicate-to-replicate variability in Caenorhabditis lifespan experiments. The issue appears to something inherent to the culturing conditions we use and independent of that approach to measurement, so this is not an issue that automated imaging can fix. Bacteria is an obvious suspect, and many groups are exploring different strategies for inactivating *E coli*. A bacteria-free approach involving liposomes was discussed, but apparently, the prep is not easy enough yet to scale up. Obviously, any group have a large incentive to be conservative and stick to existing protocols. Yet, plate-to-plate variability seems to be making everyone's research substantially harder and more time consuming.

Tim Etheridge was unable to attend due to the recent Soyuz Rocket crash.