Neuroendocrine systems in arthropods are in large part peptidergic. Not surprisingly, neuropeptide hormones are involved in the regulation of a plethora of physiological processes and behaviour. Most of these peptide hormones are produced by secretory neurons of the CNS, and are stored and released from neurohemal organs. We data-mined the 12 *Drosophila* genomes for homologs of neuropeptide genes identified in *Drosophila melanogaster*, and predicted the neuropeptidome. In five species covering main phylogenetic lines of *Drosophila*, we then biochemically profiled the peptide hormone content of the major neurohemal organs and the epitracheal glands by MALDI-TOF mass spectrometry. Our results suggest that all *Drosophila* species have an identical neuropeptidome and peptide hormone complement-the same (homolog) peptide hormones are stored in the respective neurohemal organs and the epitracheal glands in all investigated species. This infers that the last common ancestor of *Drosophila* already had a set of neuropeptides and peptide hormones identical to that of modern fruitflies. This evolutionary stability of peptide hormone systems over more than 50 million years is remarkable, since drosophilid flies have adapted to different environments which likely require various adaptations of physiological processes that are under peptidergic regulation.

VI.59

Acquisition of electrical properties in motoneurons is specified by a combinatorial code of transcription factors

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Specification of motoneurons is orchestrated by a combinatorial code of homeodomain containing transcription factors. In the *Drosophila* larval ventral nerve cord, expression of Even-skipped (Eve) specifies axon pathfinding and synapse formation of dorsally projecting motoneurons (aCC/RP2). By comparison, ventrally projecting motoneurons, RPs 1, 3, 4 and 5 (the RPs), are specified by the LIM-homeodomain transcription factors Islet, Lim3 and dHb9. In addition, we show that the two motoneuron populations can also be distinguished by their electrophysiological properties. Our results indicate that the acquisition of these distinct electrical properties utilises a similar combinatorial code of transcription factors. Thus, overexpression of Eve is sufficient to alter electrical properties of aCC/RP2. Furthermore, we demonstrate that ectopic expression of Eve in the RPs and ectopic expression of Islet and dHb9 in aCC/RP2 are similarly able to alter the electrical properties and membrane excitability of these motoneurons.

Using DamID we have identified candidate target genes to which Eve can bind. A number of these candidates could potentially alter the electrical properties of motoneurons. Here, we present verification data for several of the Eve target genes and describe their contribution to electrical properties of motoneurons.

VI.60

Structure and Development of the Central Complex

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The distinct structure of the Central Complex (CC) spanning the midline is one of the most prominent features of the Drosophila brain. The CC is highly conserved across insect species and is involved in multimodal information processing and coordination of locomotion. Although analyses have been performed on the organisation of this structure in the adult brain, few investigations have addressed its ontogeny. Characterisation of the development and neuroarchitecture of this structure is critical for elucidation of function. By employing a combination of immunohistochemical techniques, enhancer trap technology and mutant analysis, we have investigated the structure and development of the CC. We isolated 16 enhancer trap lines that showed expression in the CC. Analysis using these and three reporter lines revealed a distinct genetic subdivision of neurons and isolation of several isomorphic neuron sets in the adult brain. These lines were subsequently used to assess the development of several identified CC neurons. We determined that the CC was established by P48. We found that sets of neurons developed incrementally over this period starting with the HFS. From this study we have gained further insight into CC structure and information flow and have determined a timeline of development for several major CC neuron types. In addition, we have characterised a set of enhancer trap lines that will valuable in future developmental, structural and behavioural studies.

VII.2

Selective attention gates visual information processing in *Drosophila*

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Selective attention is a very important question in neuroscience and cognitive sciences, but its mechanisms are not well understood. Here we present a direct proof that *Drosophila* uses selective attention mechanisms in its visual processing. Our proof was obtained by recording and analysing the brain activity of a flying *Drosophila* for its quantifiable viewing choices.

- The key of our success was the development of a new technology that enables reliable recordings of neural activity (action potentials and field potentials, LFPs) from the lobula plates (LPs) of behaving *Drosophila*; previously thought almost impossible by our peers.
- At the same time, we devised a novel paradigm, which greatly amplifies the optomotor behaviour of a fly facing competing visual inputs. In our flight simulator, a flying tethered fly has to choose between two moving scenes (right and left) that compete for its interest.
- By simultaneously measuring its optomotor behaviour and neural activity in its LPs, we show that the fly brain boosts the neural output of the eye in the attended side and suppresses the output of the opposite eye. These results provide direct evidence that *Drosophila* uses selective attention to gate the flow of information from its eyes.

VII.4

Mushroom body miniature B implicates the nuclear pore complex in mushroom body development and classical conditioning

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The brain's intricate neuronal circuitry has provided the framework for a variety of complex behaviors seen throughout the animal kingdom. Mushroom bodies (MBs) have been implicated in most insects as centers of sensory integration and association. We have shown that flies with a mutation in the gene *mushroom body miniature B* (*mbmB*) display significant MB phenotypes including a reduction in calyx volume, fewer cells and axon guidance defects. Behaviorally, *mbmB* has learning and long-term memory defects. Sterility and reduced MB volume in homozygous *mbmB* females were indistinguishable by genetic recombination. Using sterility, we mapped *mbmB* by complementation to 30F4-31A2. Sequencing revealed that *mbmB* mutants have a premature stop codon in the *Pendulin* (*Pen*) gene, encoding *Drosophila Importin alpha2*, a central component of the nuclear pore complex (NPC). The NPC has been implicated in axon guidance, neuronal injury response, synaptic plasticity, cell proliferation and apoptosis. We have examined PEN protein levels in adult heads. PEN is found throughout neuronal cell bodies in the brain. We are now working to rescue mutant *mbmB* behavioral and developmental phenotypes with targeted *Pen* transgene constructs and analyzing the cellular location of common learning and memory genes. Our detailed molecular characterization of *mbmB* provides strong evidence that trafficking across the nuclear membrane is critical for normal MB development, learning and long term memory.

VII.5

Does natural variation in a cGMP-dependent protein kinase (PKG) gene (*foraging*) underlie an adaptive response of memory to environmental heterogeneity?

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Natural allelic variation in the *foraging (for)* gene encodes a cGMP-dependent protein kinase (PKG) that contributes to differences in learning and memory in larval and adult *Drosophila melanogaster*. These allelic variants are pleiotropic for differences in food-related locomotion in larvae and adults and likely have fitness consequences. We investigated if there was a relationship between learning, memory and the adaptive response to environmental heterogeneity in these natural variants. We tested their responses to environmental variation using both reversal learning and multiple stimulus learning in a Pavlovian paradigm. We predicted that the rover strain (for^R) would place greater emphasis on their most recent experience compared to sitter (for^s) and sitter mutants on a rover genetic background (for^{s2}) since they are more likely to encounter environmental variation in nature. As expected, the for^R strain displayed a greater ability to reverse their response to the stimulus-shock pairing compared to for^s and for^{s2}. When flies were asked to learn multiple stimulus-odour pairings, the rover strain placed more emphasis on the last conditioning while decreasing the strength of the first, whereas the sitter strains placed equal emphasis on both. These data, considered alongside other known *for* pleiotropies, suggest that variation in *for* influences an adaptive response to environmental heterogeneity which may