POS-TUE-001

THE INITIAL AXON OUTGROWTH FROM THE OLFACTORY EPITHELIUM

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The olfactory system provides an outstanding model that allows for the understanding of the mechanisms that drive neurodevelopment and axon-glia interactions. This system is unique because new neurons are constantly generated from stem cells that line the basal layer of the olfactory epithelium. New axons then extend from the epithelium into central nervous system where they terminate. The glia of the olfactory system, the olfactory ensheathing cells (OECs), are thought to be essential for the regenerative capacity of the olfactory system. However the initial outgrowth of axons and the interactions with OECs during early development are poorly understood. To visualise olfactory axons in early development we used OMP-ZsGreen transgenic mice at the ages E10.25 to E13 (n=3 at each age). The bright fluorescence of the ZsGreen enabled us to view growth cone morphology and track the trajectory of the axons as they exited the basal layer of the olfactory epithelium and projected into the central nervous system. Using the ZsGreen axons, combined with a more sensitive immunohistochemistry protocol, we have identified that olfactory sensory neurons first arise at E10.25 and their axons penetrate the telencephalon at E11.0. At E10.75 we have also identified the presence of dendrites projecting from the olfactory neurons. OECs migrate ahead of the axons and establish the pathway through which the axons extend which can be seen from as early as E11. These results demonstrate that the establishment of the olfactory nerve pathway is dependent on the migration of OECs and that olfactory axons penetrate the olfactory bulb earlier than previously thought.

POS-TUE-002

ALTERING DOPAMINE ONTOGENY IN DROSOPHILA MELANOGASTER INCREASES VISUAL RESPONSIVENESS IN ADULT MALES

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Purpose: Epidemiological evidence indicates that schizophrenia is a neurodevelopmental disorder. At a neurochemical level, it would appear that there are also underlying abnormalities in dopamine (DA) signaling in the brain as a result. Therefore the aim of this research, was to use the invertebrate model *Drosophila melanogaster*, which provides genetic tools to exquisitely control spatial and temporal expression of genes, to investigate the effects of transiently altering DA activity during early fly development and measure the effects of such alterations on visual behaviour of adult flies. **Methods:** DA signaling was manipulated to either attenuate or potentiate DA release during four critical developmental epochs in fly brain development. DA release was controlled using the temperature-sensitive trans-genes UAS-*Shibire*^{ts} and UAS-TRPA1 coupled to a tyrosine hydroxylase Gal4 driver line (TH-Gal4). Determining alterations in visual behaviour was conducted with a visual choice maze that segregated fly populations according to their level of responsiveness to a moving grating, expressed as an Optomotor Index (OI). **Results:** Decreasing DA release, at any of the four developmental epochs, did not significantly alter visual responsiveness in male or female adult flies. However, increasing DA activity in male adult flies, during the third developmental epoch (5-7.5 days) significantly increased optomotor response (OI=1.24 ± 0.11, N=580 compared to both controls TH/+ (t = 7.024, p < 0.0001) and TRPA/+ (t = 6.994, p <0.0001), but not in females. Furthermore, increasing DA activity in male flies, during developmental epoch 7.5-10 days also significantly increased optomotor response (OI=1.35 ± 0.176, N=580), compared to both controls TH/+ (t = 6.96, p < 0.0001) and TRPA/+ (t = 5.710, p <0.0001), but not in females. **Conclusion:** We propose that increased optomotor responsiveness in flies reflects a failure in the ability to suppress a visual reflex. Our findings point to later stages in male fly brain development as a key epoch for establishing the dopaminergic circuitry necessary for attention-like processes such as stimulus suppression. Our results suggest that *Drosophila* may be an efficient model system to study underlying developmental or functional correlates observed in neurodevelopmental disorders that compromise attention, such as schizophrenia.

POS-TUE-003

OVEREXPRESSION OF TEN-M3 IN THE RETINA OF THE WALLABY M*ACROPUS EUGENII* ALTERS THE TOPOGRAPHY OF IPSILATERALLY-PROJECTING RETINAL AXONS

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Purpose: Ten-m3 is a transmembrane glycoprotein expressed in a decreasing ventrodorsal retinal gradient and a decreasing mediolateral gradient in the superior colliculus (SC) of the wallaby (*Macropus eugenii*). Here we examine its distribution in the dorsal lateral geniculate nucleus (dLGN) and assess the effects of its localised overexpression in the retina on the development of retinal projections. **Methods:** The expression of mRNA for Ten-m3 in the dLGN was investigated at early developmental stages by in situ hybridisation (n=4). Ten-m3 was overexpressed on postnatal day 18-22 by in vivo electroporation of a DNA construct encoding Ten-m3/green fluorescent protein (GFP) in localised regions of the retina (n=7). A control group was electroporated with a construct encoding GFP (n=5). Animals survived for 18-22 days before retinal projections were traced by an intravitreal injection of horseradish peroxidase. **Results:** In the SC there was an increase in ipsilateral projections to medial and rostral regions when Ten-m3 was overexpressed in the retina. In the dLGN there was a high dorsal to low ventral gradient of expression, corresponding topographically to its retinal gradient. The effect on ipsilateral projections was dependent on the retinal site of overexpression. An expansion was observed when Ten-m3 was overexpressed in ventronasal retina (n=3). Overexpression in dorsonasal retina produced no change (n=4). **Conclusion**: Overexpression of Ten-m3 alters the distribution of ipsilateral projections to the dLGN and SC. This complements data from Ten-m3 knockout mice and supports a key role for Ten-m3 in the control of ipsilateral retinal mapping in the brain.

POS-TUE-004

SEIZURE-RELATED GENE 6: A MODULATOR OF EXCITATORY SYNAPSE DEVELOPMENT

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Seziure-related gene 6 (Sez-6) is required for normal dendritic arbor development of cortical pyramidal neurons. While neurons lacking Sez-6 display excessive branching, these branches appear to be less able to support synapse development as excitatory synapses in the adult cortical neuropil are reduced by around 30%. The structure of Sez-6 also suggests a role in synapse biology. All three isoforms contain protein interaction domains (CUB and SCR) which are involved in neurotransmitter receptor trafficking and gating. **Purpose:** These experiments were aimed at investigating the role of Sez-6 in excitatory synapse development. **Methods:** Synaptogenesis and trafficking assays were performed in cultured neurons. Synapse development was monitored in Sez-6 null and control cortical neurons at a series of time-points (up to 18 days *in vitro*) by fixing and immunostaining for the vesicular glutamate transporter (VGLUT1; a presynaptic marker for cortical excitatory synapses) and post-synaptic density 95 (PSD-95). Colocalized regions (puncta size 0.8 - 4.0 μm^2) were scored. Antibody-feeding with the anti-Sez6 antibody was used to assess Sez-6 trafficking. **Results:** By immunoelectron microscopy, Sez-6 is detected in dendrites and dendritic spines. Trafficking experiments revealed that Sez-6 is trafficked from the cell-surface in recycling endosomes in an activity-enhanced manner. Excitatory synapses were observed to develop at a similarly low rate in Sez-6 wild-type (WT) and knockout (KO) neurons over the first week in culture. By 14 DIV, however, Sez-6 KO neurons exhibited significantly more synapses than WT neurons. Interestingly, this situation was reversed by 18 DIV. Neurons treated with secreted Sez-6 exhibited an increase in excitatory synapse number (50%, P<0.01) and colocalized puncta size (13%, P<0.01). **Conclusion:** Sez-6 is not required for the initial stages of excitatory synaptogenesis. In contrast, the maintenance of newly-formed synapses is enhanced by Sez-6, supporting a role in synapse maturation.