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Sequential Expression of let-7 and E93 during the Larva to Pupa Transition in *Drosophila*

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Whether transitioning from larva to pupa, or child to adolescent, animals proceed through distinct stages during their development. How such transitions are specified is poorly understood. In many animal forms, the transition from juvenile to adult development is controlled by accumulation of the let-7 micro-RNA. In *Drosophila*, adult development also requires the transcription factor E93 which is expressed specifically at the pupal stage. To determine the relationship between let-7 and E93, I have focused on the mushroom bodies (MBs) of the brain. MBs are generated by neuroblasts, stem cells that divide to produce neurons throughout larval and pupal development. Neurons generated at the pupal stage produce distinct adult lobes of the mushroom body. In pupae, E93 is expressed in a ring of neurons immediately surrounding each neuroblast, whereas let-7 is expressed in an adjacent outer ring of neurons. Two models could explain these expression patterns: 1) neurons express E93 soon after birth, but then transition to expressing let-7 as they become displaced from the neuroblast by later-born neurons; or 2) neuroblasts generate let-7-expressing neurons early in metamorphosis, and then switch to generating distinct E93-expressing neurons. These models were distinguished by permanently marking cells that have expressed E93 at any time in their developmental history. The experiment was to use E93-Gal4 to drive expression of yeast Flippase which then permanently activated GFP expression by the excision of a cassette containing a transcriptional stop. The results support model 2; green fluorescence was restricted to E93-Gal4-expressing neurons, with no fluorescence seen in let-7 expressing neurons. These observations indicate that the transition from larval to adult development in the mushroom body occurs in two steps, with neuroblasts first generating let-7-expressing neurons, followed by the production of E93-expressing neurons. Experiments are under way to determine the fates of each of these neuronal types.