

**D-7** Immnoreactivity of Two Neuropeptides, Locustatachykinin and Allatostatin, in the Wax Moth Brain and Retrocerebral Complex

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Antisera raised against locustatachykinin I (LomTK I), one of the myotropic peptides that have been isolated from the locust brain and corpora cardiaca, and allatostatin I originally isolated from the cockroach, were used for immunocytochemical detection of neurons in the brain and retrocerebral complex (corpora cardiaca-corpora allata, CC-CA) in the seventh (or last) instar larva of the wax moth *Galleria mellonella*. Most of LomTK I-immunoreactive cells showed strongly immunoreactivity in the pars intercerebralis of the brain with axons leading to the corpus allatum, whereas allatostatin I-immunoreactivity was distributed in specific neurosecretory cells of the brain and corpus allatum. Especially, other LomTK I-immunoreactive cells were found in the tritocerebrum. The widespread distribution of LomTK and allatostatin immunoreactivities suggests that these neuropeptides were synthesized in the brain, transported and stored in the corpus allatum, and finally released into the hemolymph. Therefore, these two kinds of neuropeptides act as neurohormones in the wax moth, suggesting that these hormones have connection with larval metamorphosis.

**D-8** Enhanced expression of insulin mRNA by GLUT2 cDNA in the human pancreatic islet  $\beta$ -cell lines, COBE18.2 and N2

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We have introduced the gene GLUT2 encoding the facilitated glucose transporter GLUT2 into the COBE18.2 and N2 cells to assess its impact on glucose-stimulated insulin release and glucose metabolism. The chromosomal localization of incorporated GLUT2 cDNA was detected by FISH, and expression of GLUT2 was detected by various means. Western blot analysis showed an expression of 54kDa GLUT2 protein in transfected cells, and electron microscopy showed ultrastructural changes of the cellular activity, including nucleolus and polysomes, after transfection. Stable transfection with human low affinity GLUT2 glucose transporter cDNA revealed a significant improvement in glucose-stimulus expression of insulin mRNA. RT-PCR analysis revealed that 16.7mM glucose induce in expression of the insulin gene in the GLUT2-transfected cell clone COG1 COG2, NG1, NG2, and NG3. This results showed GLUT2 transfected cells recovered insulin transcription responsiveness to high glucose concentration. The experiments demonstrate that an increased glucose uptake via a low-affinity glucose transporter is an important factor for the induction of insulin gene expression .