

**P027** Phosphorylation of the V-ATPase subunit C by protein kinase A

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V-ATPases are regulated by the reversible disassembly  $V_1V_O$  holoenzyme into a cytosolic  $V_1$  complex and a membrane bound  $V_O$  complex. The signalling cascades triggering these events are largely unknown. We report that the  $V_1$  subunit C of the tobacco hornworm *Manduca sexta* interacts with protein kinase A (PKA) and is the only V-ATPase subunit that is phosphorylated by PKA. Subunit C can be phosphorylated as single polypeptide as well as a part of the  $V_1$  complex, but not as a part of the  $V_1V_O$  holoenzyme. Both the phosphorylated and the unphosphorylated form of subunit C are able to reassociate with the  $V_1$  complex from which subunit C had been removed before. Using salivary gland extracts of the blowfly *Calliphora vicina* in which V-ATPase reassembly and activity is regulated by the neurohormone serotonin via PKA, we show that 8-CPT-cAMP causes phosphorylation of subunit C. Similarly, incubation of intact salivary glands with 8-CPT-cAMP or serotonin leads to the phosphorylation of subunit C but this is abolished by the PKA inhibitor H-89. These data suggest that subunit C binds to and serves as a substrate for PKA and that this phosphorylation may be a regulatory switch for the formation of the active  $V_1V_O$  holoenzyme.