Septal glucagon-like peptide 1 receptors (GLP-1Rs) mediate the behavioural effects of cocaine

A.E. Harasta,[1] M. Schneider,[2] R. Spanagel,[2] G.D. Housley[1] and M. Klugmann,[1] [1]Translational Neuroscience Facility, Department of Physiology, School of Medical Sciences, University of New South Wales, NSW 2052, Australia and [2]Central Department of Mental Health, Psychopharmacology, University of Heidelberg, Mannheim, Germany.

Background: By activating neuronal circuitries of the reward system addictive agents cause brain alterations due to their reinforcing properties. The shift from controlled to compulsive drug use is still not fully understood, but the lateral septum (LS) seems to play a key role in the contextual reinstatement of drug-seeking. Anatomically, the LS is located in the subcortical forebrain and is positioned as an essential nodal point for integrating cognitive with affective information and relying it to directly control behavioural responses appropriate to the environmentally stimuli (Sheehan, 2004). The molecular mechanisms underlying neuroplastic changes in LS neurons are unknown. The glucagon-like peptide 1 receptor (GLP-1R), a G_s-protein-coupled receptor, is expressed in various CNS regions including the LS. GLP-1R signalling in other brain regions protects neurons from excitotoxicity, and enhances learning (During *et al.*, 2003). GLP-1R agonists protect glutamate-induced apoptosis in dissociated hippocampal neurons and lead to a transient elevation of intracellular Ca^{2+} (Gilman *et al.*, 2003). The endogenous agonist glucagon-like peptide 1 (GLP-1) regulates satiety and acts as a neurotransmitter produced in the nucleus of the solitary tract (NTS). Based on beneficial effects of GLP-1R stimulation in the pancreas, the long-lasting exogenous GLP-1R agonist exendin-4 (Ex4) has been approved for treatment of type-2 diabetes (T2DM). GLP-1R agonists can cross the blood brain barrier but it is not known if systemic administration activates septal GLP-1Rs. Central stimulation of GLP-1R suppresses food reward by interacting on the mesolimbic system (Dickson *et al.*, 2012).

Hypothesis: GLP-1R signalling in the LS might impact on drug-induced neuroplasticity and behavioural outputs.

Methods and Results: To determine the expression of GLP-1R in the rodent brain we performed *in situ* hybridization studies and confirmed most abundant expression of GLP-1R mRNA in the LS. Systemic administration of the receptor agonists (Ex4, 2ug/kg, i.p.) triggers neuronal activity in the LS determined by c-fos-immunohistochemistry. We then addressed the role of central GLP-1R in the locomotor sensitization to cocaine, a non-contingent model to study enduring neuroplasticity in addiction. Locomotor sensitization to cocaine (10mg/kg, i.p.), develops over 5 d, followed by 7 d of cocaine withdrawal after which the animals are tested for sensitization expression by a final cocaine administration. Pharmacological blunting of GLP-1R signalling in C57/Bl6 mice, using the specific receptor antagonist, Exendin9-39 (150ug/ kg, i.p., 30 min prior to cocaine administration, n=7), attenuates the expression of locomotor responses to cocaine. In line with that, development and expression of sensitization is attenuated in GLP-1R-deficient GLP-1R KO mice (n=12). We complemented GLP-1R specifically in the LS of adult GLP-1R KO mice using neurotropic adeno-associated virus (AAV)-mediated GLP-1R gene transfer. AAV1/2-GLP-1R (3.8 × 1010 vector genomes in 1µl, at 200 nl/min; microprocessor controlled mini-pump) was delivered bilaterally into the LS (+0.5 mm AP; ±0.3mm ML; −2.6 mm DV from bregma) of anaesthetized (isoflurane 5% induction; 1% maintenance) GLP-1R KO mice (n=12). As controls, titre-matched AAV-EGFP vector was injected into the LS of wildtype (n=12) or KO animals (n=12). 3 wk post surgery, when AAV-mediated transgene expression peaked to remain at stable levels, animals were subjected to the sensitization paradigm. Animals receiving AAV-EGFP were indistinguishable from naïve subjects. Importantly, AAV-GLP-1R-injected KO animals showed a wildtype-like performance highlighting the LS as a central brain region for GLP-1R function *in vivo*.

Conclusion: Our data suggest a pivotal role of GLP-1R signalling for drug-induced neuroplasticity in the LS.

Dickson SL, Shirazi RH, Hansson C, Bergquist F, Nissbrandt H, Skibicka KP. (2012) The glucagon-like peptide 1 (GLP-1) analogue, exendin-4, decreases the rewarding value of food: a new role for mesolimbic GLP-1 receptors. *Journal of Neuroscience* **32:** 4812-4820.

During MJ, Cao L, Zuzga DS, Francis JS, Fitzsimons HL, Jiao X, Bland RJ, Klugmann M, Banks WA, Drucker DJ, Haile CN. (2003) Glucagon-like peptide-1 receptor is involved in learning and neuroprotection. *Nature Medicine* **9:** 1173-1179.

Gilman CP, Perry T, Furukawa K, Grieg NH, Egan JM, Mattson MP. (2003) Glucagon-like peptide 1 modulates calcium responses to glutamate and membrane depolarization in hippocampal neurons. *Journal of Neurochemistry* **87:** 1137-1144.

Sheehan, T. (2004) Regulation of affect by the lateral septum: implications for neuropsychiatry. *Brain Research Reviews* **46:** 71-117.