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Presentation Time: 2:00 – 2:20 p.m.
(15 min presentation; 5 min for Q&A)

Title: Drugs *for* the Brain *from* the Brain: CNS Glycopeptide Drugs from Peptide Hormones

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Peptide neurotransmitters related to enkephalins, dynorphins, angiotensin, secretins, and other peptide hormones, both cyclic and linear, have been converted to carbohydrate-bearing O-linked glycopeptide drug candidates with enhanced stability *in vivo* and increased BBB penetration. Short glycopeptides (5–7 residues) have been designed which produce mu opioid agonism, delta opioid agonism, or synergistic mu + delta opioid agonism. By linking helical amphipathic *"addresses"* to these opioid *"messages"* it was possible to enhance their anti-nociceptive effects *in vivo* in rodents. Glycosylated angiotensin analogues with neuroprotective activity (*e.g.* PNA5 from ProNeurogen) show promise for clinical application for treatment of post-surgical dementia. Even larger pituitary adenylate cyclase-activating peptide (PACAP) show promise in the treatment of neurodegeneration. These glycosylated hormones were evaluated for their ability to stimulate cAMP production *in vitro* using individual CHO cell lines expressing PAC1, VPAC1, and VPAC2. LC-MS[3] was used to test for *in vitro* stability and blood-brain barrier (BBB) penetration using microdialysis. Using mild progressive unilateral 6-hydroxydopamine (6-OHDA) lesions in rat, we were able to "flip" the normal hemi-parkinsonized PD model to provide a read out of damage in the presence (15 mg/kg, *i.p.*, 2 doses, 6 hrs pre- and 48 hrs post-6-OHDA injection) of a PACAP lactoside **2LS98LAC**. The glyco-PACAP treated rats were compared to untreated (n=8/group). The cumulative amphetamine-induced rotations at 2 and 4 weeks post-lesion were reduced compared to the vehicle control group (2-tail *t*-test, *p*=0.04). Unbiased stereology of dopaminergic (DA) neurons in the substantia nigra (n=8/group) showed in the control group a 25% decrease on the lesioned vs the intact side, while there was no significant difference between intact vs lesioned side in the PACAP-treated animals.

Additional studies showed that **2LS98LAC** can prevent brain damage in mice that were subjected to transient Medial Cerebral Artery Occlusion (tMCAO) for 60 minutes followed by 23 hours of reperfusion. Both infarct volume and edema were reduced after 24 hrs. Rotarod performance and neurological defect scores were also significantly enhanced in these mice (n=6, **p<0.01).

These results and others suggest that glycosylation of endogenous peptide hormones shows great promise as a general approach for the design of CNS active drugs to treat a variety of conditions. Molecular weight (MW) does not appear to affect BBB penetration rates, at least in the range of MW's examined so far, 550—3,500 Daltons. We hypothesize that this ability to penetrate the BBB is due to the ability of the glycopeptides to adopt conformations that render them either highly water soluble or highly amphipathic structures that associate strongly with biological membranes, a feature we have dubbed *"biousian behavior."*