GLP-1 and GIP analogues as novel treatments for Alzheimer’s and Parkinson’s disease

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**abstract**

Type 2 diabetes is a risk factor for developing chronic neurodegenerative disorders such as Alzheimer’s or Parkinson’s disease. The underlying mechanism appears to be insulin desensitisation in the brain. A range of GLP-1 mimetics and GIP analogues initially designed to treat diabetes protected transgenic animals that model Alzheimer’s disease and toxin based animal models of Parkinson’s disease. Novel dual GLP-1/GIP analogues also show good neuroprotective effects. Based on these findings, first clinical trials have been conducted. In a pilot study in patients with Alzheimer’s disease, the GLP-1 analogue liraglutide showed good protective effects in \(^{18}\)FDG-PET brain imaging. It was found that the disease related decay of brain activity had been completely stopped by the drug. In a pilot study in patients with Parkinson’s disease, the GLP-1 mimetic exendin-4 showed good protection from motor and cognitive impairments. These results demonstrate the potential of developing disease-modifying treatments for Alzheimer’s and Parkinson’s disease.

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1. Diabetes is a risk factor for neurodegenerative disorders

One of the established risk factors for the development of Alzheimer's (AD) or Parkinson's disease (PD) is type II diabetes mellitus (T2DM). In several patient data base analyses, T2DM has been identified as a risk factor for PD, indicating that insulin desensitization in the periphery may be a factor in initiating or accelerating the development of neurodegenerative processes. In AD, several epidemiological studies found a correlation between T2DM and an increased risk of developing AD at a later stage in life. In one investigation, T2DM had been identified as a risk factor that doubled the chance of developing AD. In a longitudinal cohort study that follows up the health status of people over time, glucose intolerance in an oral glucose tolerance test correlated with an increased risk of developing AD in people with significantly elevated blood glucose levels. Other studies arrived at similar conclusions. In Parkinson's disease, T2DM has also been identified as a risk factor. In the basal ganglia, dopaminergic transmission failure, insulin desensitisation and energy depletion had been associated with T2DM.

2. Insulin signaling desensitises in the brain

A key mechanism that appears to link T2DM with neurodegenerative disorders is the loss of insulin signaling in the brain. A biochemical analysis of brain tissue of AD patients showed a clear profile of insulin desensitisation, even in people that were not diabetic. Insulin receptor subunits and IRS1 was found to be hyper-phosphorylated, a biochemical profile also seen in diabetics in the peripheral tissue. In PD, insulin desensitisation was also observed in the key brain area such as the basal ganglia and substantia nigra. Energy utilisation, mitochondrial function, insulin signaling and dopamine transmission was found to be compromised. It is interesting to note that these effects were also found in non-diabetic subjects. This demonstrates that insulin desensitisation is not always dependent on glucose levels. However, patient data showed that a higher percentage of PD patients were diabetic or glucose intolerant compared to age matched controls.

3. Insulin is a key growth factor
Insulin is an important growth factor that is essential for the homeostasis and cell growth and repair in neurons. Counter-intuitively, glucose uptake in neurons is not insulin dependent, with the exception of large neurons that express the GLUT4 subtype 25,26. Hence, the brain had been commonly known as an ‘insulin insensitive’ organ 27. However, insulin and IGF-1 are important growth factors that activate cell growth, cell repair, gene expression, energy utilisation and protein synthesis 28-31. This may explain why insulin desensitisation in the brain increases the risk for developing neurodegenerative disorders such as AD and PD.

4. Treating AD patients with insulin

Just as insulin improves T2DM, treating AD patients with insulin shows improvements in cognition, attention, reducing levels of biomarkers for AD, and normalising brain energy utilisation 32-35. Insulin cannot be given to people who are not diabetic. Delivering insulin by nasal application where it enters the brain more directly can circumvent the problem of inducing hypoglycaemia. Nasal application of insulin improved attention and memory formation even in non-diabetic people 34,36,37. A phase II clinical trial in AD patients showed improved cognition in patients with mild cognitive impairments (MCI). It further improved the amyloid1-40/1-42 ratio in the cerebrospinal fluid and increased brain activation as seen in 18FDG-PET scans which measure brain activity and energy utilisation, and furthermore showed improvement in mental tasks 38-40. However, similar to patients with T2DM, insulin delivery appears to enhance brain insulin desensitisation and worsen cognitive decline 40. For a review, see 41,42.

5. T2DM drugs have neuroprotective properties

Drugs to treat T2DM and that normalise insulin signaling are on the market. These are mimetics of the incretin hormone Glucagon-like peptide 1 (GLP-1) 43,44. GLP-1 is a growth factor of the glucagon family type and has similar properties than insulin has 31. These drugs do not affect blood glucose levels directly and therefore are safe to take by people who are not diabetics 45. The drugs are well received and have a good safety record 46. Several of these drugs can cross the blood-brain barrier, which demonstrates that there is a transporter for GLP-1, similar to other growth factors such as insulin or leptin 25,47-51.
There has been some discussion whether GLP-1 receptors are expressed in neurons. A study that analysed RNA expression of the GLP-1 receptor has demonstrated a wide distribution of GLP-1Rs in the brain, including the cortex, hippocampus, and the substantia nigra; key brain areas in AD and PD disease development. Several antibody-based histological investigations of GLP-1 receptor expression in the brain had been conducted since. However, a study that demonstrated that these antibodies may not be selective for the receptor followed, and a recent analysis of GLP-1R expression in the brain using a transgenic GFP expression reporter mouse strain demonstrated good expression of GLP-1Rs in the cortex, hippocampus area CA3 and dentate gyrus, and others, putting the discussion to rest once and for all.

6. GLP-1 mimetics show effects in animal models of Alzheimer’s disease

In several transgenic mouse models of AD, which expresses the human Swedish mutated form of the amyloid precursor protein (APP) and a mutated human form of presenilin-1 (PS1), both mutations which lead to AD in humans, GLP-1 mimetics were neuroprotective. Liraglutide (Victoza®) is on the market as a treatment for T2DM. Once-daily injections for 8 weeks reduced key parameters such as memory loss, synapse loss, reduced synaptic transmission, chronic inflammation in the brain, and amyloid plaque load in the brain. The same treatment in aged transgenic mice with advanced amyloidosis still showed some protective effects, suggesting that treatment at later disease stages may still have benefits. When treated from an early age onward, the drug did prevent disease progression and has the potential to be used as a prophylactic. The GLP-1 mimetic lixisenatide (Lyxumia®) also had similar neuroprotective effects compared to liraglutide. Liraglutide had clear protective effects in a mouse model of tau phosphorylation and tangle formation, a key biomarker for AD. In the human P301L mutated tau expressing mouse, a model of fronto-temporal lobe dementia and ALS, liraglutide reduced the amount of tangles and hyperphosphorylated tau. In the accelerated senescence SAMP8 mouse model, liraglutide also showed good protective effects on memory formation and neuronal loss. The GLP-1 mimetic exendin-4 (Byetta®, Exenatide®, Bydureon®) also showed good effects in a triple transgenic mouse model of AD.
Exendin-4 showed neuroprotective effects in other animal models of neurodegeneration as well. GLP-1 mimetics furthermore improve neuronal progenitor cell proliferation and neurogenesis in the mouse brain. In mouse models of AD and of diabetes, GLP-1 analogues can increase or normalise neuronal progenitor cell proliferation in the CNS. Testing analogues of the sister incretin Glucose-dependent insulino tropic polypeptide (GIP) also showed good effects in the APP/PS1 mouse model of AD.

7. GLP-1 mimetics show effects in animal models of Parkinson's disease

Exendin-4 has shown good neuroprotective effects in several mouse models of PD. In the 6-hydroxydopamine (6-OHDA) model of PD where dopaminergic neurons are eliminated by 6-OHDA, animals were treated for 3 weeks and showed functional recovery. In the substantia nigra, dopaminergic neurons were partly protected from the toxic effects of 6-OHDA. This result was confirmed in a second study which also used the 6-OHDA lesion technique, and a second technique, the lipopolysaccaride (LPS) induced substantia nigra lesion. Exendin-4 reduced the lesions induced by the toxins. The levels of dopamine measured in the basal ganglia were also increased. The numbers of neurons in the substantia nigra was also higher than in the lesion only group. In a third study, Exendin-4 protected dopaminergic neurons and rescued motor function in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) lesion mouse model of PD.

When comparing the more effective GLP-1 mimetics liraglutide and lixisenatide with exendin-4, it was found that both liraglutide and lixisenatide demonstrated good protective effects while exendin-4 showed only minor protection in the MPTP mouse model of PD. Motor activity was partly rescued, and dopaminergic neurons were protected in the substantia nigra. Expression of the dopamine biomarker tyrosine hydroxylase (TH) was also rescued in the liraglutide and lixisenatide treated mice. Pro-apoptotic cell signaling was reduced, while growth factor signaling was enhanced by both drugs. When testing the sister incretin GIP in the MPTP mouse model, it was found that the long-lasting protease resistant analogue D-Ala2-GIP-glu-PAL showed good protective effects. Motor activity was partly rescued, and the number of dopaminergic neurons in the substantia nigra was increased. Synapse numbers were increased, and the cAMP/PKA/CREB growth factor second messenger pathway was shown to be activated by D-Ala2-GIP-glu-PAL.
New dual GLP-1 and GIP receptor agonists have been developed to treat T2DM. Some have already been tested in clinical trials and show superior performance compared to liraglutide. When testing a novel dual agonist in the MPTP mouse model of PD, it was found that it rescued motor activity, synapse numbers, numbers of dopaminergic neurons in the substantia nigra, and reduced chronic inflammation (see fig. 1). Interestingly, the expression of the neuroprotective growth factor Brain-Derived Neurotropic Factor (BDNF) was enhanced, which can explain some of the neuroprotective effects observed. BDNF has clear protective effects on synaptic activity.

8. Clinical trials

The results obtained in the preclinical studies show an impressive range of neuroprotective effects of GLP-1 and GIP mimetics. As several GLP-1 mimetics are already on the market as treatments for T2DM with a good safety profile, clinical trials have started that investigate the neuroprotective effects of exendin-4 or liraglutide in PD or AD patients.

Parkinson’s disease

A clinical pilot trial of exendin-4 in PD patients has been completed (clinical trials identifier NCT01174810). This ‘proof of concept’ study tested the effects of exendin-4 in a randomised, open label trial in 45 patients. The drug was administered over 12 months, followed by a 2 month wash-out period. In a single-blinded rating of motor activity, clear improvements were found, and cognitive measures were improved in the drug group compared to controls. Exendin-4 treated patients had a mean improvement at 12 months on the MDS-UPDRS of 2.7 points, compared to a mean decline of 2.2 points in control patients (P = 0.037). Importantly, the drug group showed a clear improvement in the Mattis DRS-2 cognitive score, suggesting that exendin-4 has beneficial effects on cognition and memory. The group was re-tested 12 months after the trial had finished, and the clear differences between groups in motor performance and cognitive scores had not changed. This suggests that the group difference is not due to a placebo effect, as 12 months is too long for such subjective effects to last. A phase II trial testing the once-weekly formulation of exendin-4, Bydureon®, has been completed (NCT01971242). The results will be reported shortly and initial observations suggest a good outcome.
A phase II trial testing liraglutide in PD patients is under preparation and will start in July 2016, testing 100 patients in a double blind, placebo controlled design.

**Alzheimer’s disease**

A randomized, double blind clinical trial to assess the safety and efficacy of Exendin-4 treatment in 230 MCI patients (early phase Alzheimer’s disease) is currently ongoing at the NIH/NIA in the USA. This trial is testing the effects of exendin-4 on key parameters such as performance in the Clinical Dementia Rating (CDR) scale sum-of-boxes, the Alzheimer’s Disease Assessment Scale - cognitive sub-scale (ADAS-cog), Behavioral and cognitive performance measures, changes on structural and functional MRI brain imaging, and hormonal and metabolic changes in cerebrospinal fluid and plasma AD biomarkers (ClinicalTrials.gov Identifier: NCT01255163).

A small-scale trial with 34 patients has been completed in Denmark at the University of Aarhus. This double blind, randomized trial tests the effects of liraglutide vs. placebo on MCI patients, using $^{18}$FDG-PET imaging to estimate cortical activity and PIB-PET imaging to measure plaque load. Excitingly, there was a clear effect on brain $^{18}$FDG-PET activity. FDG is a modified glucose molecule, and the uptake correlates well with brain activity, synaptic activity, and disease progression. While the placebo group showed the expected reduction in the $^{18}$FDG-PET signal of up to 20%, the drug group showed no reduction at all and even demonstrated improved signalling in some brain areas (NCT01469351).

A second larger scale phase II clinical trial with liraglutide in 206 MCI patients is currently ongoing in the UK. The trial has a randomised, placebo controlled double blind design and will analyse $^{18}$FDG-PET brain activity, PET inflammation markers (microglia activation), MRI brain scan changes, CSF samples for inflammation markers and amyloid /tau levels, and cognitive tests such as the ADAS Exec score. Patient recruitment is currently ongoing (NCT01843075).

**References:**

GLP-1 and GIP analogues as novel treatments


A novel GLP-1/GIP dual agonist protects the brain in a mouse model of Parkinson's disease

Motor activity is protected from the effects of MPTP

Dopaminergic neurons are protected in the brain

Fig. 1: A novel dual GLP-1 and GIP receptor agonist displays neuroprotective properties in the MPTP mouse model of PD. Motor activity was much reduced by MPTP, and the novel drug rescued this to some extent. In the substantia nigra, dopaminergic neurons that express TH are much reduced in numbers by MPTP, and the novel dual agonist protected neurons to some extent. Shown are histological sample sections; A=saline control, B=drug, C=MPTP, D=MPTP plus drug. Adapted from 86.