

Repurposing Diabetes Drugs to Treat Insulin Resistance in Alzheimer's Disease

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Abstract

Alzheimer's disease is a neurodegenerative condition which results in a significant decline in cognitive status. Novel treatment approaches for Alzheimer's are sorely needed, as current medications for the disease offer only marginal clinical benefit. Research has discovered a connection between the pathology of Alzheimer's and Type 2 Diabetes, two serious and seemingly unrelated disorders. Clinical studies have shown that Alzheimer's disease is associated with brain insulin resistance similar to the pathology of Type 2 Diabetes. This observation has led to the notion that drugs developed for the treatment of Type 2 Diabetes may be beneficial in modifying the cognitive function and pathophysiology of individuals suffering from Alzheimer's disease. This paper offers a comprehensive review of the clinical studies demonstrating the potential of using diabetes medications as an effective therapeutic method for the prevention and treatment of Alzheimer's disease. Special focus is given towards the metabolic hormones insulin, amylin and leptin.

Introduction

Alzheimer's and Type 2 Diabetes are two of the most common diseases afflicting the elderly population today. A 2016 Report by the Alzheimer's Association found that of the 5.4 million Americans with Alzheimer's, an estimated 5.2 million people are age 65 and older (Alzheimer's Association, 2014). Similarly, the American Diabetes Association reported that in 2012, of the 29.1 million Americans with Diabetes, an estimated 11.8 million people were age 65 and older (American Diabetes Association, 2012). The two conditions have traditionally been treated as independent disorders. However, recent studies linking Alzheimer's to Type 2 Diabetes have led to the proposal that Alzheimer be referred to as "Type 3 Diabetes". Discoveries of common pathological mechanisms between these two diseases have given rise to novel clinical trials incorporating diabetes therapies to treat Alzheimer's disease.

Methods

A variety of literary reviews and research papers on the subject were collected through use of the PubMed database. Keywords such as Alzheimer's disease, insulin resistance and Type 2 Diabetes were used to search for relevant material. Access to PubMed was provided by the Touro College online library system.

Pathophysiology of Alzheimer's and Type 2 Diabetes

Alzheimer's disease is a neurodegenerative condition, which results in nerve cell death and tissue loss throughout the brain (Li, Z et al., 2015). Scans of brains of individuals suffering from Alzheimer's demonstrate severe shrinkage of the hippocampus and cerebral cortex, as well as the enlargement of the ventricles (Querfurth et al., 2010).

The pathophysiology of Alzheimer's disease is described by the amyloid cascade hypothesis. Cleavage of amyloid precursor protein (APP) leads to the formation of the protein amyloid beta. Excessive cleavage of APP coupled with inefficient removal of amyloid beta can lead to the formation of amyloid beta plaques in the brain. These plaques damage and destroy

brain cells by blocking cell-to-cell signaling at synapses (Reitz, 2012). Amyloid beta plaques have also been shown to cause apoptosis. The presence of amyloid plaques in the brain generates the production of harmful oxidative free radicals which in turn activates the c-Jun N-terminal kinase (JNK) pathway. The JNK pathway stimulates the transcription of several key target genes, including the death inducer Fas ligand. The binding of Fas ligand to its receptor Fas then induces a cascade of events that lead to caspase activation and ultimately neuronal cell death. (Yoshiyuki et al., 2001).

The tau protein also plays an important role in Alzheimer's disease (Lasagna-Reeves CA et al., 2012). This protein is integral to maintenance of internal support and transport systems between brain cells. Hyperphosphorylation, which is the addition of phosphate to too many amino acids, leads to the collapse of tau proteins into twisted strands referred to as neurofibrillary tangles (Jack et al., 2013). Without the support of the tau protein, the transport system in the brain collapses. Consequently, essential nutrients are unable to reach brain cells and cell death ensues (Gong and Iqbal, 2008).

Diabetes is a metabolic disorder which develops when the body does not make enough insulin or is unable to use insulin effectively. (Yarchoan and Arnold, 2014). If left untreated, diabetes can cause many complications, including kidney failure, diabetic ketoacidosis, heart disease and stroke (Schnell et al., 2016). Type 2 Diabetes is characterized by an insulin resistant state which is most commonly caused by obesity (Smith and Kahn, 2016). In insulin resistance, muscle, fat, and liver cells do not respond properly to insulin and thus cannot easily absorb glucose from the bloodstream. As a result, the body needs higher levels of insulin to help glucose enter cells. The pancreatic beta cells initially increase their insulin output but fail over time to keep up with the body's increased demands for insulin. Type 2 diabetes develops when insulin production is inadequate to overcome insulin resistance, resulting in the drastic rise in blood glucose levels (Yarchoan and Arnold, 2014).

Discovery of Insulin Resistance in Alzheimer's Disease

Insulin receptors are widely distributed in brain regions known to be involved in memory function, including the hippocampus, cerebral cortex and cerebellum (Werther et al., 2015). Clinical studies have found evidence for central insulin resistance in Alzheimer's brains. Post-mortem studies of brain tissues from people with Alzheimer's disease have discovered extensive abnormalities in insulin and insulin-like growth factor signaling mechanisms in the brain (Steen et al., 2005). A study of post-mortem human hippocampus Alzheimer's tissue was done by exposing the tissue to different concentrations of insulin. This allowed researchers to study the activation of insulin pathways in the brain tissue of people with Alzheimer's compared to the brain tissue of those with normal cognitive function. In normal brains, activation of the insulin receptor initiated a signaling cascade which led to the production of many downstream insulin signaling proteins. The blunted insulin response observed in the Alzheimer's brain tissues was similar to the insulin resistance observed in Type 2 diabetes peripheral tissues (Talbot et al., 2012).

The formation of amyloid beta plaques has been implicated as a possible impetus for the removal of insulin receptors in brain cells. Studies of Alzheimer's brains have demonstrated that the binding of amyloid beta oligomers to hippocampal neurons triggers the removal of dendritic insulin receptor substrates from the outer membrane of the cell (Xie et al., 2002). These studies prove that elevated amyloid beta levels induce the removal of cell surface insulin receptors, thereby furthering insulin resistance.

A molecular model has been proposed to explain how the amyloid beta plaques found in Alzheimer's disease promote insulin resistance (Dineley et al., 2014). This model is based on the inflammatory pathway observed in Type 2 Diabetes, where inflammatory cytokines play an important role in establishing insulin resistance. Signaling from the inflammatory cytokine Tumor Necrosis Factor (TNF) stimulates JNK (Hoeks et al., 2012). Activation of the JNK pathway by TNF results in serine phosphorylation of the insulin receptor (Zhao et al., 2008). In order to be activated, phosphorylation of the insulin receptor must occur at a tyrosine residue. Addition of the phosphate group at the serine amino acid in individuals with Type 2 Diabetes results in inhibition of the insulin receptor (Bomfim et al., 2012).

In a similar fashion, inflammatory cytokines induced by the presence of amyloid beta in the brain can lead to insulin resistance in Alzheimer's. Recent work suggests that soluble misfolded amyloid beta can induce inflammatory cytokines through an inflammatory pathway known as NF- κ B-inducing kinase (NIK). The resulting inflammatory state induces insulin resistance through feedback inhibition of the insulin receptor (Carrero et al., 2012).

In this case the many signaling chemicals produced by the inflammatory pathway stop the activity of the insulin receptor (Talbot et al., 2012).

Insulin resistance may promote Alzheimer pathology through several mechanisms. Firstly, abnormal insulin signaling may promote amyloid beta and hyperphosphorylated tau formation through the kinase GSK-3. The activity of GSK-3 is normally regulated through inhibition by the protein AKT, which is an important kinase in the insulin signaling cascade referred to as the IRS-1 –AKT pathway (Yarchoan and Arnold, 2014). Therefore, dysfunctional insulin signaling caused by disturbances in the IRS-1 –AKT pathway leads to increased GSK-3 activity. Studies of GSK-3 found that its activity increases tau protein phosphorylation (Li, X et al., 2006) and that it is involved in amyloid beta production (Takashima, 2006).

Abnormal insulin signaling also interferes with memory and learning pathways. Insulin is a direct regulator of the ERK/MAP kinase pathway. This pathway is essential to the induction of longer term potentiation (LTP) and memory consolidation in the hippocampus (Winder 1999). LTP is responsible for the maintenance of long term memory, which is the ability to recall episodes which are not part of the immediate past (Kelleher et al., 2004). Disruptions of LTP and memory consolidation contribute to the impaired cognitive function typified by Alzheimer's disease (Dineley et al., 2014).

Discussion: New Role for Diabetes Drugs as a Treatment for Alzheimer's

Given the role of insulin resistance and deficiency in the pathogenesis of Alzheimer's disease, it is possible that a drug currently prescribed for Type 2 Diabetes may also be useful for Alzheimer's. Various diabetes drugs are under study to test for their potential activity in Alzheimer's. This section provides an overview of the various clinical studies which assess the potential prospects of using the diabetes drugs insulin and amylin, as well as the weight loss hormone leptin, to treat Alzheimer's.

Insulin

The administration of exogenous insulin has predictably become a prime focus of the effort to treat insulin signaling dysfunction in Alzheimer's. Clinical trials have demonstrated that insulin administration acutely affects behavior and cognitive performance in both healthy individuals and those suffering from Alzheimer's. A 2001 study assessed the effects of peripherally administered insulin infusion in non-impaired individuals (Kern et al., 2001). Two groups were infused with insulin for 6 hours, one at a high rate and the other at a low rate. Any effects of the insulin administration on blood glucose concentrations were counteracted by constant glucose infusions. Comparison of the results

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of the high rate insulin and low rate insulin treatment groups indicated an improvement in the cognitive function of the high rate insulin group. Subjects exposed to higher insulin infusion rates demonstrated changes in auditory-evoked potentials, had enhanced memory as evidenced by improved word recall and improved cognitive flexibility as measured by the Stroop test. Similar benefits of peripheral insulin administration on cognitive function were observed in individuals with Alzheimer's. In a 1999 study, Alzheimer's patients showed improved story recall and attention during insulin infusion relative to saline infusion (Craft et al., 2012).

Despite the results of these clinical trials, there are two main concerns that must be addressed with regards to utilizing peripheral insulin administration as a treatment for Alzheimer's. Firstly, peripherally administered insulin cannot bypass the blood brain barrier in order to enter the central nervous system (CNS) and is therefore unable to affect and improve brain function. Secondly, peripheral insulin infusions can induce hypoglycemia. Hypoglycemia is a condition characterized by abnormally low blood glucose which can lead to a seizure or unconsciousness if left untreated. Although hypoglycemia was mitigated by simultaneous glucose infusions in both studies involving peripheral insulin administration, this solution is highly impractical outside of a research setting (Yarchoan and Arnold, 2014).

In light of these concerns, intranasal insulin is considered to be the best method for insulin delivery in Alzheimer clinical trials. Insulin that is delivered nasally bypasses the blood brain barrier and is rapidly delivered into the cerebrospinal fluid from where it can easily enter the CNS (Mao et al., 2016). Additionally, because intranasal insulin is preferentially delivered to the CNS, it is possible to achieve clinically relevant insulin concentrations in the CNS without causing systemic hypoglycemia (Yarchoan and Arnold, 2014).

Pilot clinical trials using intranasal insulin have had successful results. A 2008 clinical trial reported that delivery of intranasal insulin for 21 days improved story recall, attention and caregiver-rated functional status in cognitively impaired subjects or individuals with Alzheimer's (Reger et al., 2008). In another clinical trial subjects given insulin spray over a placebo demonstrated improved delayed memory and cognitive function (Craft et al., 2012).

One study sought to illustrate the underlying mechanisms through which intranasal insulin ameliorates Alzheimer's pathology (Mao et al., 2016). APP/PS1 mice possessing the pathological features of Alzheimer's disease received intranasal insulin treatments for a total of 6 weeks, while a control group received saline treatments. Tissue samples were harvested from both groups upon the conclusion of the treatment period.

In order to determine whether intranasal insulin enhances brain insulin signaling, the respective levels of key components of the insulin signaling pathway, such as the insulin receptor beta-subunit (IRB) and protein B kinase (AKT), were measured through Western Blot analysis. Tissue from healthy wild type mice was also analyzed to serve as a reference marker of normal levels. The total levels of IRB and AKT were significantly decreased in the saline treated APP/PS1 mice compared with wild-type controls. However, the levels of IRB and AKT in the mice which received intranasal treatment were closer that of the wild-type mice, indicating that intranasal insulin treatment can partially protect APP/PS1 mice from brain insulin signaling deficits.

Intranasal insulin was also shown to reduce the activation of JNK in APP/PS1 mice. Activation of the JNK pathway results in serine phosphorylation of the insulin receptor and induces apoptosis. The level of phosphorylation of JNK was significantly increased in the hippocampus of saline-treated mice compared with wild-type controls, signifying that intranasal insulin reduces JNK activation.

Immunohistochemical analysis of the tissue samples measured the amounts of amyloid beta plaque deposits in the brains of the APP/PS1 mice. The number of amyloid plaques in the APP/PS1 insulin-treated mice was significantly reduced in both the hippocampus and cortex compared to the saline control group. It was also discovered that the area of amyloid beta plaques was significantly decreased in both the hippocampus and cortex of the insulin treated mice. Additionally, quantitative analysis revealed substantially reduced soluble amyloid beta oligomers in insulin-treated APP/PS1 mice. This revelation is especially significant considering that soluble amyloid beta oligomers are considered the most neurotoxic form of amyloid beta. Enhanced neurogenesis was also observed in the insulin-treated APP/PS1 mice. It was shown that intranasal insulin significantly increased levels of DCX, a marker of neurogenesis. The overall conclusions of the study were that intranasal insulin treatment improves cognitive deficits, ameliorates defective brain insulin signaling, strongly reduces amyloid beta plaque formation, inhibits JNK activation and promotes neurogenesis in APP/PS1 mice.

Despite the successful results observed in studies involving intranasal insulin treatment, there is concern that chronic hyperinsulinemic conditions in the brain may actually promote brain insulin resistance. Excessive exposure to insulin in mice has been shown to lead to abnormal phosphorylation of key components of the insulin signaling pathway, such as AKT and GSK-3, in a manner consistent with insulin resistance (Kim et al., 2011). It may therefore be beneficial to explore avenues of diabetes treatments which restore the byproducts of insulin signaling without directly affecting insulin levels.

Amylin

Amylin is a metabolic hormone which is co-secreted with insulin by pancreatic beta cells (Adler et al., 2014). Amylin's systemic effects in diabetic patients include the lowering of blood glucose levels through delayed gastric emptying, increased satiety, and decreased secretion of glucagon, which is an antagonist of insulin (Yarchoan and Arnold, 2014).

Amylin's signaling activity has the potential to alleviate the detrimental effects of insulin resistance in Alzheimer's disease. Amylin binds to independent receptors in the brain to activate signaling pathways that converge with insulin signaling. Amylin activates the production of the protein AKT, which is needed for the proper regulation of GSK-3, a protein which can lead to increased production of amyloid beta plaques and hyper phosphorylated tau (Moon et al., 2011). Amylin is also a known modulator of the ERK signaling cascade, a pathway significant in the maintenance of long term memory and memory consolidation (Adler et al., 2014). An advantage of using amylin as a medication is that it poses no risk of hyperinsulinemia, as it can activate the insulin pathway without interfering with the concentration of insulin in the body (Yarchoan and Arnold, 2014).

One study investigated the potential outcomes of using amylin as a treatment for Alzheimer's. The first part of the study compared plasma human amylin levels between individuals with Alzheimer's or mild cognitive impairment and individuals with no cognitive impairments (Adler et al., 2014). The results showed significantly lower amylin levels among subject with Alzheimer's and mild cognitive impairments compared to individuals with no cognitive deficits. With this correlation between amylin levels and cognitive function established, a follow-up study was conducted in order to investigate the effects of amylin administration on Alzheimer's pathology. A senescence-accelerated prone (SAMP8) mouse was selected as a model of Alzheimer's related dementia, because it displays multiple features known to occur early in the pathogenesis of Alzheimer's including cortical atrophy, amyloid beta alterations, tau phosphorylation and severe deficits of learning and memory. The SAMP8 mice were treated with either amylin or saline infusions for a total of 5 weeks. Due to the tendency of amylin to aggregate into brain plaques in its natural form, a soluble analog of amylin called pramlintide was used in the study.

The object recognition test was performed on the mice during the last week of the 5-week treatment period in order to assess the effects of amylin on cognitive function. The object recognition test is a behavioral assay that is based upon the natural tendency of mice to investigate a novel object instead of a familiar one, as well as their innate tendency to restart exploring

when they are presented with a novel environment. The mice were placed in an open field box filled with different objects of various shapes and sizes. After a series of trials, during which the mice habituated to the configuration and properties of the different objects, some of the objects were replaced with new ones to evaluate novel object recognition. The pramlintide-treated SAMP8 mice spent a greater proportion of time exploring the novel objects as compared with the familiar objects, whereas the saline-treated SAMP8 mice did not differ in time spent with the novel and familiar objects. The behavior of the pramlintide-treated mice signified a marked improvement in their cognitive function.

Hippocampal tissue samples were harvested from the SAMP8 mice in order to assess the effect of amylin on oxidative stress, an important pathologic feature of Alzheimer's. The protein levels found in the pramlintide-treated SAMP8 mice indicated a decrease in the molecular markers associated with oxidative stress and neuro-inflammation. The pramlintide-treated SAMP8 mice had significantly decreased expression of the protein HO-1 in the hippocampus compared with saline-treated mice. HO-1 is a cellular stress protein that is activated during high oxidative stress and inflammatory states, and is also known to be increased in the cerebral cortex of Alzheimer's brains. The pramlintide-treated SAMP8 mice also had decreased levels of the lipid peroxidation adduct HNE and the enzyme COX-2. HNE is a protein that is known to be an early and abundant cellular stress marker in Alzheimer's brains, while COX-2 is a classic marker of inflammation which is increased in Alzheimer's brains.

The pramlintide-treated SAMP8 mice expressed high amount of proteins associated with synaptic activity and dendritic growth. Amylin was found to increase expression of hippocampal synapsin I, a protein located in neuronal synaptic vesicles that is involved in synapse formation, neurotransmitter release, and learning and memory. A robust increase in CDK5 was also observed in the hippocampus of the pramlintide-treated SAMP8 mice. CDK5 is a kinase which plays an intimate role in synaptic plasticity, learning and memory in adult brains.

The overall conclusions of the study were that chronic infusion of amylin in SAMP8 mice was found to improve memory performance in object recognition tests, increase neural synaptic activity and decrease inflammatory markers in the hippocampus. Amylin treatment improved both the cognitive status and Alzheimer's pathology features of the SAMP8 mice.

Another study also assessed the results of amyloid treatment on behavioral impairment and brain amyloid pathology in mouse models of Alzheimer's disease (Zhu et al., 2015). The

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study utilized 5XFAD Alzheimer mice which exhibit significant neuron loss. The 5XFAD mice were treated with intraperitoneal injections of either pramlintide or saline once daily for 10 weeks.

The 5XFAD mice were tested for improvements in cognitive functions by going through a Morris water maze. The Morris water maze test is used to determine hippocampal spatial memory deficits. The test consisted of placing the rodent in a circular tank filled with cloudy water, which is used to motivate the animal to escape the water by swimming to a hidden platform located in one quadrant of the pool. Over several days the rodent learnt to find the hidden platform by using spatial cues, such as posters or taped objects strategically placed on the walls outside of the water maze, in the testing room. The pramlintide treatment improved the performance of the mice in the Morris water maze test, reducing the time necessary for memory acquisition and retention during maze training as compared to the saline-treated control group. The improved performance of the pramlintide-treated groups over the saline-treated groups in the Morris water maze test demonstrated that peripheral treatments with pramlintide improves learning and memory in the 5XFAD mice.

Pramlintide treatment was shown to have an effect on amyloid beta levels, one of the prime pathological hallmarks of Alzheimer's. Immunoassays of brain tissue from the 5XFAD mice revealed that pramlintide-treated 5XFAD mice experience a reduction in both size and intensity of amyloid beta plaques in the cortex, hippocampus and thalamus and had decreased numbers of amyloid beta plaques in all areas of the brain with the exception of the hippocampus. Furthermore, comparison of amyloid beta serum levels from before and after the intraperitoneal pramlintide infusion revealed a significant increase in serum amyloid beta after the infusion. These results indicate that pramlintide enhances the removal of amyloid beta from the brain and its transfer into the blood.

The successful outcomes of these separate studies assessing the effect of pramlintide treatment on different Alzheimer's mouse models indicate that amylin has potential to become a promising new avenue for the treatment of Alzheimer's. Amylin was shown to improve cognitive function, reduce oxidative stress markers, increase synaptic activity and enhance clearance of amyloid beta from the brain.

Leptin

Leptin is a chemical produced by fat tissues which activates the central nervous system to regulate food consumption and energy expenditure (Oomura et al., 2006). Although leptin has traditionally been studied in the context of obesity, recent studies

have examined its neurological effects. Leptin receptors are highly expressed in areas of the brain that are involved in learning and memory, such as the hippocampus (Li, X-L et al., 2002).

A study was designed in order to assess leptin's role in the regulation of hippocampal functions and the control of learning and memory processes (Li, X-L et al., 2002). The study focused on leptin receptor-deficient mice. The behavioral and molecular data obtained from the leptin receptor-deficient mice was compared against a positive leptin receptor control group. Both groups of mice were put through the Morris water maze task. The leptin receptor-deficient mice swam greater distance than their positive controls before they found and climbed onto the hidden platform. When the platform was removed, the leptin receptor-deficient mice crossed the original platform location fewer times than the positive control group. The hippocampus was removed from the leptin receptor-deficient rodents and the control group. Electrophysiological analysis of the hippocampal tissue of the leptin receptor-deficient mice showed impairments of long term potentiation (LTP) and long term depression (LTD). The decreased cognitive behavior and impaired LTP and LTD processing observed in the leptin receptor-deficient mice support the conclusion that leptin enhances LTP and regulates mechanism involved in both learning and memory

Leptin also appears to regulate a number of defining features of Alzheimer's disease. AMP- dependent kinase (AMPK) is known to regulate glycogen synthase kinase-3B (GSK-3B), a kinase which is crucial in the regulation of tau phosphorylation (Nikolaos et al., 2009). It has been shown that leptin directly activates AMPK and thereby possesses the ability to modulate tau phosphorylation (Yu et al., 2004). Leptin also facilitates the uptake of amyloid beta complexes via its regulation of the lipoprotein receptor-like protein (Fewlass et al., 2004). Thus, leptin's activity directly affects the regulation of amyloid beta uptake and tau phosphorylation, two of the impaired pathophysiological features of Alzheimer's disease.

One study assessed the effects of leptin treatment on CRND8 Alzheimer's mouse models (Greco et al., 2010). The CRND8 mice received daily treatments of leptin for a total of 8 weeks with a control group receiving saline infusions. Leptin-treated mice spent statistically more time with the novel object compared to the saline-treated control group during the object recognition test, indicating an improvement in working memory performance after leptin treatment. Analysis of brain tissue from the CRND8 mice revealed that the leptin-treated group had reduced amyloid beta levels in both brain and serum. Staining of the brain tissue for amyloid fibers showed a significant decrease in amyloid burden in the hippocampus of the leptin

treated CRND8 mice, which was associated with a decrease in the average size of amyloid plaques. There was no significant increase in the levels of the inflammatory molecule C-reactive protein, tumor necrosis factor or cortisol in the plasma of the leptin-treated group compared to the saline-treated group, indicating that leptin does not induce an inflammatory reaction. The results of the study fully support the ability of leptin to ameliorate Alzheimer's like pathological pathways, strengthening leptin's potential of becoming a novel therapeutic treatment for Alzheimer's disease.

Synergistic Effects of Amylin and Leptin

Leptin and amylin activate overlapping signaling cascades and ultimately converge on the insulin signaling pathway by activating AKT and increasing insulin sensitivity (Yarchoan and Arnold, 2014). Recent studies have indicated that leptin and amylin signaling appear to have synergistic properties.

One study profiled hypothalamic neurons in order to determine the effects of amylin and leptin on hypothalamic activity (Li, Z et al., 2015). It was discovered that hypothalamic expression of lapp, a precursor to amylin, was markedly decreased in mice with mutations in the gene regulating the production of amylin, but was normalized after infusions of leptin. The decrease of amylin expression in mice that had mutated leptin genes showed that hypothalamic amylin is a neuropeptide that is leptin regulated. Additionally, AC187, an amylin antagonist, was found to blunt the activity of leptin and decreased its effects on neurons in the hypothalamus. The presence of the amylin antagonist significantly inhibited the effects of leptin on both leptin depolarizing and hyperpolarizing neurons. The ability of an amylin antagonist to blunt the response of leptin suggests that amylin can modulate leptin's effects. Leptin and amylin were also found to have synergistic effects on hypothalamic neurons. Patch clamp recordings demonstrated that the presence of either leptin or amylin elicited similar excitatory and inhibitory effects on hypothalamic neurons. Leptin excited 65% and inhibited 35% of the neurons, while amylin excited 62% and inhibited 38%. The significant correlation between the effects of individual neurons exposed to both treatments indicates that amylin depolarizes the same neurons that are depolarized by leptin and hyperpolarizes the same neurons that are hyperpolarized by leptin. This suggests that the response of the neuron would be amplified upon simultaneous presentation of amylin and leptin.

The synergistic potential of these two treatments has been explored with regards to treating obesity. Stand-alone obesity treatments have proven unsuccessful because diet-induced obese (DIO) rats and obese humans are only minimally responsive to even high pharmacological doses. Nonetheless, amylin possesses the ability to heighten leptin's effects. For this reason,

one study found that doses of exogenous leptin that was highly effective in lean rats had minimal effects on weight or food intake in DIO rats. However, the results of the study showed that administration of amylin together with leptin resulted in a synergistic, fat-specific reduction in body weight in two independent experiments (Roth et al., 2008).

In light of this discovery, a clinical trial was designed in order to evaluate the weight-lowering effect of combined amylin/leptin (using pramlintide/metreleptin) treatment in human obesity. A 24-week, randomized, double-blind, study was conducted in obese or overweight subjects. Subjects receiving pramlintide/metreleptin lost almost 13% of their initial body weight over 24 weeks, compared with only ~8% in subjects receiving either pramlintide or metreleptin. Towards the end of the study, weight loss plateaued in subjects treated with monotherapy, but not in subjects treated with the combination. The overall results of the study showed that weight loss caused by the combination of leptin and amylin in humans was greater than the additive weight loss of each drug used alone (Ravussin et al., 2009).

Thus far, there have been no studies to examine the efficacy of combination therapy of leptin and amylin to treat Alzheimer's. The successful results observed in weight loss clinical trials suggest that even greater improvements in memory in Alzheimer's patients may be possible by using amylin and leptin as a combined therapy.

Conclusion

The discovery of insulin resistance in Alzheimer's has given way to a growing interest in restoring insulin signaling in Alzheimer's with therapeutic agents originally developed for the treatment of Type 2 Diabetes. Intranasal insulin, amylin and leptin are examples of hormones typically associated with obesity and diabetes which have shown promise for treating Alzheimer's disease. An advantage of using amylin and leptin as medications for Alzheimer's is that unlike with insulin, treatment using these hormones pose no risk of inducing hyperinsulinemia, as they activate the products of the insulin signaling pathway without interfering with the concentration of insulin in the body. Furthermore, weight loss studies have discovered that amylin and leptin are synergistic substances which produce significantly enhanced results when used in combination. This unique synergy suggests that achieving even greater improvements in memory may be possible by using amylin and leptin as a combined therapy for Alzheimer's. Further research will need to take place before the proposed combined therapy can be tested in a clinical trial and ultimately be distributed pharmaceutically.

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