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How Effective Are the Potential Treatments for Alzheimer's Disease, How Do They Work, and Is a Cure in Sight?

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ABSTRACT

This literally overview looks into the effectiveness of the current treatments available to patients suffering from Alzheimer's disease. There are many factors linked to the pathogenesis of Alzheimer's disease (AD). Ultimately, AD is caused by the selective loss of neurons in regions of the brain such as the hippocampus.

The disease process is 'a kind of reversal of ageing', with the sufferer losing management function, the ability to make judgements, evaluate courses of action and their consequences, and undertake multifaceted tasks. The need for a cure is apparent. Nonetheless, current treatments are not effective enough for sustained treatment of Alzheimer's.

The article explores the differences in function between acetyl-cholinesterase inhibitor, NMDA receptor antagonist, γ-secretase modulator, anti-Aß antibody and insulin therapy in relation to Alzheimer's. It has become evident that as of yet, we have been unable to arrive at a certain cure for this disease. However, we are now able to prescribe to patient's drugs that can aid in making their quality of life as an Alzheimer's patient and the families of the unfortunate more bearable. Some treatments can help slow the onset of the adverse effects of Alzheimer's, if spotted early; but not stop them.

It is hopeful to think that a cure is near. It's currently unlikely as we still do not fully understand which elements trigger the disease, and which causes the most damage.

Keywords - *Alzheimer's disease; Acetylcholinesterase; Parkinson's disease; Amyloid; Rosiglitazone.*

INTRODUCTION

Alzheimer's disease (AD) is a neurodegenerative disorder and the most common cause of dementia.[1] Progressive loss of memory and cognitive function are hallmarks of Alzheimer's[2], which eventually results in behavioural and personality changes. The patient may be apathetic, socially withdrawn, or disoriented in familiar environments, and his inhibitions may become so reduced that he acts bizarrely and inappropriately in public. The disease process is 'a kind of reversal of ageing.[3] with the sufferer losing executive function, the ability to make decisions, evaluate courses of action and their consequences, and undertake complex tasks.[4] The extensive damage to the frontal lobes of the brain, thought to be responsible for complex thought and reasoning is an underlying cause of Alzheimer's, and this atrophy results in the devastating final stages of Alzheimer's: the patient becomes bed-bound, unable to communicate, and unable to eat or drink due to the loss of coordination of muscles that control swallowing.[5] Death results after a period of up to ten years from diagnosis.

There are many factors linked to the pathogenesis of Alzheimer's disease. Ultimately, AD is caused by the selective loss of neurons in regions of the brain such as the hippocampus (implicated in memory) and the cerebral cortex (responsible for reasoning, memory, and language). Neurons do not get replaced because they remain in the G0 phase[6] and do not undergo mitosis, therefore, a neuron must live as long as its host.[7] It is for this reason that the adult brain cannot repair itself, although it does compensate for damage by making new connections between remaining neurons.[8]

There is some controversy over which pathway triggers the series of events that lead to neuronal death, and which factor is the most important in the pathogenesis of AD.

The amyloid cascade hypothesis is one of the leading theories. Senile plaques were first observed by Alois Alzheimer in the brain of a 51 year old woman who had died after progressive dementia. These plaques are composed of a dense core of Amyloid-beta (Aß) protein, and their frequent appearance in the brains of AD sufferers led to the conclusion that the insoluble aggregates caused the disease.[9] Another anomaly observed in the AD brain is the neurofibrillary tangle (NFT) composed of tau protein. Tau is a microtubule-associated protein which promotes tubulin polymerisation and stabilises microtubules; however, when tau is hyperphosphorylated, its tertiary structure is altered and it no longer binds to microtubules.[10] Hyperphosphorylated tau is the major component of the paired helical filaments which make up NFTs, and these NFTs result in the impaired functioning of microtubules. Microtubules provide structural support to a cell, along with routes for nutrient transport, so it is theorised that the build up of tau tangles causes neuronal death by stopping the transmission of signals and the transportation of vital

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molecules.[11] The amyloid cascade hypothesis states that the accumulation of Aß plaques sets in motion a series of events (such as tau hyperphosphorylation) that lead to neuronal death. Studies suggest that every five years after the age 65, the risk of developing AD doubles.[12]

The need for a cure is apparent, but in the case of AD, the definition of 'cure' is ambiguous. Because Alzheimer's often occurs late in life, it may not be necessary to halt the progression altogether. Delaying the onset by just five years could reduce the number of people with AD by 50% by 2050.[13] Therefore, in assessing the effectiveness of a treatment for AD, therapies that slow the course of the disease must also be considered. Nevertheless, the definition of 'cure' remains as a treatment which stops the progression of the disease by interfering with its mechanism and any therapy which fails to do this is only palliative. The term 'effectiveness' can also be used to distinguish between treatments that only give symptomatic relief, and those that have the potential to stop or reverse AD; for example, can the treatment delay the onset of AD by more than five years? Can the treatment normalise Aß and phosphorylated tau levels? And can it stop neuronal death, or even replenish lost nerve cells? Questions of this sort will be consulted when trying to ascertain the effectiveness of a drug or therapy.

Current Treatments

One of the first breakthroughs in AD treatment came in the 1970s when David Bowen discovered that the activity of acetyltransferase, the enzyme which catalyses acetylcholine synthesis was reduced in the brains of AD sufferers. Ten years previously it had been demonstrated that dopamine deficiency causes Parkinson's disease, so it was hypothesised that AD occurs as a result of low concentrations of acetylcholine and a resulting dysfunction of cholinergic neurons.[14] It is now well established that the cholinergic system has a role in AD: for instance, it is known that anticholinergic agents cause attention and memory deficits, and that the cholinergic system modulates memory and learning. Post-mortem studies also show that the degree of cognitive impairment in Alzheimer's patients correlates well with cholinergic abnormalities.[15] These observations led to the development of the 'cholinergic enhancement treatment approach', and the most commonly used drugs of this family are acetylcholinesterase inhibitors (AChEIs) (Table 1).

Acetylcholine (ACh) is an ester of choline and acetic acid with the structural formula $CH_3COOCH_2CH_2N^+(CH_3)_3$ (Figure 1).

It is a neurotransmitter released by cholinergic neurons. When a nerve signal arrives at the axon of a neuron, vesicles containing ACh fuse with the plasma membrane, causing its release into the synaptic cleft, where the neurotransmitter will bind to post-synaptic receptors and activate a response.[16] When ACh binds, the receptor channel opens, causing an influx of Na^+ into the cell and an efflux of K^+ ions.[17]

Table 1: Current treatment of AD

Treatment	How it works	Pros	Cons
Acetylcholinesterase inhibitor	Inhibits the action of the enzyme which breaks down neurotransmitter Acetylcholine.	Can give some symptomatic relief.	Expensive and the benefits are modest: only effective in half of treated patients. Some side effects. Does not slow disease progression.
NMDA Receptor antagonist	Protects against neuronal excitotoxic death from over-activation of the NMDA receptor by glutamate.	Can reduce neuronal death. Cheaper, more efficacious, and has fewer side effects than AChEIs.	Does not interfere with senile plaques or neurofibrillary tangles; more symptomatic than disease modifying.
γ-secretase modulator	Reduces the activity of an enzyme which cleaves APP to make the toxic Aß peptide.	Reduces concentration of Aß.	Recent trials have shown such drugs to be ineffective in halting disease progression.
Anti-Aß antibody	The antibodies attach to Aß in the brain and the immune system clears it, breaking down senile plaques.	Reduces concentration of Aß, clears senile plaques.	Recent trials have shown that this approach may not work. Dangerous side effects have been associated with these drugs.
Insulin therapy	Insulin blocks the toxic binding of ADDLs, decreases the intraneuronal concentration of Aß, and reduces tau hyperphosphorylation.	Interacts with both Aß and tau. Can completely reverse tau hyperphosphorylation, sparing microtubules from dissociation. Blocks the toxic effects of ADDLs, potentially preventing neuronal death.	Increases extracellular concentration of Aß. As yet untested in Alzheimer's, and could prove as disappointing as the Aß vaccine. Insulin-sensitizing drugs can have dangerous side effects.

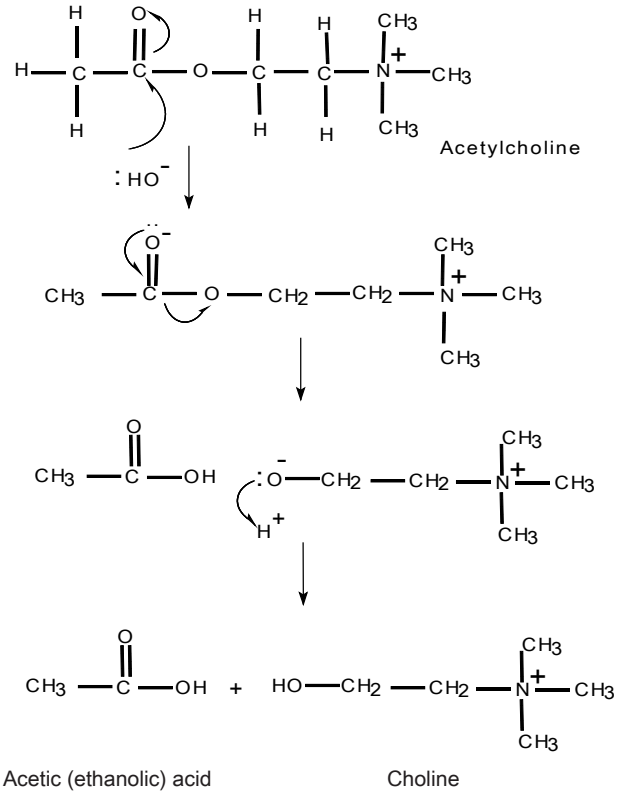

Figure1: The mechanism for the hydrolytic deactivation of acetylcholine.

This activity is terminated when ACh is broken down into acetic acid and choline by ester hydrolysis[18] *via* the Acetylcholinesterase enzyme (AChE). Acetylcholinesterase inhibitors inhibit AChE, and work under the assumption that sparing ACh from hydrolytic deactivation will increase its concentration in the synapse and therefore improve cognitive function.

There are currently four AChEIs approved by the FDA for the treatment of AD, and of these, tacrine (tetrahydroaminoacridine) was the first, in the early 1990s. Tacrine is a reversible non-competitive AChEI with a half life of 2-4 hr (the half life of a drug is the time taken for the blood concentration of the drug to decrease by half). However, tacrine is very toxic, and produces serious side effects including nausea, abdominal distress, tachycardia, and liver damage[19], necessitating a search for second generation AChEIs with fewer side effects and longer half lives. Aricept (donepezil hydrochloride) and Exelon (rivastigmine) were discovered in 1997, closely followed by Reminyl (galantamine) in 2000.

Galantamine is a competitive AChEI with a long half life of 7 hr and good bioavailability after oral administration. Trials have shown that treatment with 30-50mg daily for 13 weeks results in significant improvement in cognitive performance without the liver toxicity associated with Tacrine.[20] Galantamine is an interesting case because it has a second mode of attack, it also modulates cholinergic receptors on neurons to increase ACh release[21] due to its allosteric potentiating ligand properties[22] (essentially it activates cholinergic receptors by changing their tertiary structure). Phenserine is another AChEI with a secondary proposed mechanism of action. In animal models this compound is reported to inhibit the formation of Aß, and this anti Aß-activity could alter the actual disease course. Two recent trials show conflicting results: a phase III trial showed no significant difference between phenserine and placebo, but in a later phase IIb trial the drug reduced Aß levels in the cerebrospinal fluid as well as the formation of Aß plaques.[12]

The use of AChEIs has been controversial because the 'treatments are expensive and the benefits of these drugs are moderate at best. They certainly do not represent a miracle cure'.[23] The inhibition of AChE does not modify the progression of AD or prevent neuronal death, and at best it only treats symptoms. AChEIs are only effective in 50% of AD patients, and one third of those treated will experience deterioration over the first six months.[24] As Dr Bernhardt MD, a neurologist from the University of Maryland says: 'if you put a patient on [an AChEI] and took the same patient in a parallel universe without the drug, you might see a difference, but not much'.[25] Along with the side effects that accompany this treatment (nausea, vomiting, dizziness, diarrhoea, and anorexia), these factors make AChEIs ineffective for AD, but other cholinergic enhancement techniques look more hopeful. More specific approaches, such as targeting cholinergic receptors directly, may be efficacious. ACh interacts with both nicotinic and muscarinic receptors, which require nicotine and muscarine respectively to be allosterically activated in order to accommodate Ach.[26] Chemicals which can imitate ACh by binding to nicotinic or muscarinic receptors and evoking a response (cholinergic agonists) can improve the attention and cognition of AD patients.[27] Nicotine itself has been shown to have significant effects on the mood of AD patients[28] not observed in non-sufferers, but its toxic effects limit its safety and relevance. However, several muscarinic agonists (molecules which activate muscarinic receptors) based on the structure of ACh have been synthesised. Many are of little therapeutic value because they cause a wide range of responses (oxotremorine causes tremors similar to those in Parkinson's[13], but xanomeline offers a possibility for the future. It is a selective muscarinic agonist which is safe, efficacious, and superior to placebo. Studies have shown that it promotes 'robust improvement' in verbal learning and short term memory formation[28], but due to its unfortunate gastrointestinal side effects, there is a high drop-out rate in clinical trials.[29] Whether these drugs will prove to be more effective than AChEIs remains to be seen, but the cholinergic route cannot represent a cure for AD.

The only drug approved for the treatment of moderate to severe Alzheimer's is Memantine, which works on the N-methyl-D-aspartate (NMDA) receptor of the central nervous system and modulates synaptic plasticity. Psychologist Donald Hebb first proposed the idea that a memory is formed when two neurons interact in a way that strengthens future signalling through the synapse in

1949.[30] The NMDA receptor (a protein complex found in the plasma membrane of post synaptic neurons) plays a pivotal role in synaptic plasticity, as it detects when two neurons fire simultaneously, hence strengthening the connection between the pair. It has been demonstrated that mice lacking NMDA receptors in the hippocampus show memory deficits, but when the production of NMDA receptors is increased, mice learn faster and preserve memories for a longer time.[30] The NMDA receptor is a voltage and ligand-gated receptor that allows Ca^{2+} and Na^{+} to flow into a neuron and K^{+} to flow out when open. Under resting conditions when membrane potential is approximately -80mV, Mg^{2+} blocks the ion channel (Figure 2).

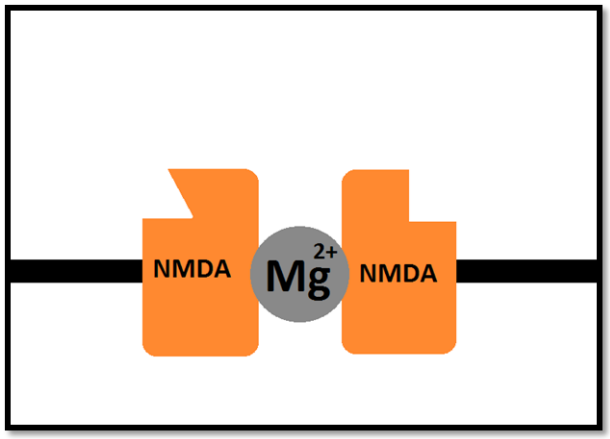

Figure 2: The unopened NMDA receptor at -80mV with Mg^{2+} ion blocking channel.

The receptor requires two molecules of co-agonists glutamate (an amino acid and the major excitatory neurotransmitter in the brain)[13] and glycine to bind, along with Mg^{2+} dissociation caused by a wave of membrane depolarisation, in order to open.[31] The influx of Ca^{2+} ions results in the activation of signalling pathways responsible for long-term cellular changes (Figure 3).

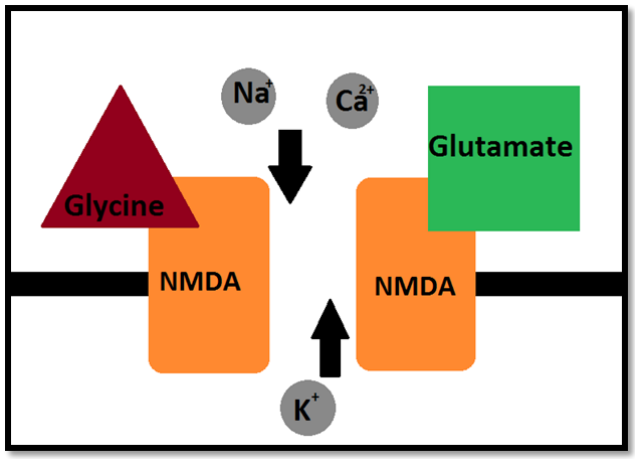

Figure 3: The opened NMDA receptor after membrane depolarisation, Mg^{2+} dissociation, and binding of coagonists glycine and glutamate. Sodium and calcium ions flow into the cell, potassium flows out.

Glutamergic neurotransmission is an event that occurs as a result of a pre synaptic release of glutamate into the synaptic cleft, where it can then binds to the NMDA receptor and provokes a response. This glutamergic system has been implicated in memory and learning, but high levels of glutamate can also be neurotoxic[32], and may be involved in AD pathogenesis *via* excitotoxicity: a chronic overstimulation of NMDA receptors which leads to excessive Ca^{2+} influx and consequential osmotic disturbances and inappropriate activation of enzymatic pathways, ultimately resulting in neuronal death.[33] This can result in a chain reaction when dying neurons release huge quantities of glutamate that stimulate apoptosis in other cells.[25] Therefore, antagonising NMDA receptors offers a legitimate approach for treating AD; however, because glutamate can both enhance learning and cause excitotoxicity, any such treatment has to be carefully balanced. For example, although high affinity NMDA receptor antagonists powerfully block the ion channel and give excellent protection from excitotoxic cell death, they are accompanied by severe side effects such as memory loss, hallucinations, and ataxia (the gross loss of muscle coordination).[34] These side effects are due to the high affinity, slow unblocking rate, and insensitivity to membrane potential changes that characterise these drugs, which prevent the normal levels of NMDA receptor activity required for learning and memory formation.[35]
On the other hand, lower affinity NMDA receptor antagonists can selectively allow normal receptor function while inhibiting over activation. Memantine is one such drug has low to moderate affinity, uncompetitive and voltage-sensitive binding properties, and a fast unblocking rate.[35] The Memantine molecule essentially plays the same role as Mg^{2+} ions in blocking the ion channel, but binds with a higher affinity than Mg^{2} (Figure 4).

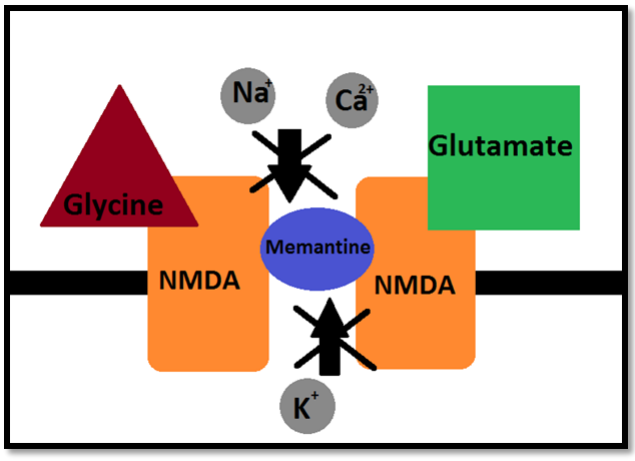

Figure 4: Memantine acting as a NMDA receptor antagonist. It blocks the ion channel, stopping the toxic influx of calcium ions.

It effectively protects against excitotoxic cell death without the side effects of higher affinity NMDA receptor antagonists. The efficacy and safety of Memantine have been demonstrated in several clinical trials. A 28 week trial

in 2003 showed that the side effects caused by Memantine occurred at similar rates in patients treated with placebo, and when the trial ended, patients taken off Memantine treatment showed significant functional and cognitive decline.[36] Another 24 week US trial in 2004 compared the effects of donepezil and Memantine versus Donepezil and placebo. It was found that the combination of Memantine and Donepezil was advantageous, and patients given Memantine showed less functional and global decline than placebo.[37]

Memantine therapy significantly reduces care dependence in severely demented patients[38], and in combination with AChEIs it slows the rate of decline on key measures of cognition, global functioning, (a measure of the social, occupational, and psychological functioning of adults) and basic activities of daily living, with cumulative effects over time.[39] Memantine use also correlates to a reduction in patient management costs for society and the caregiver[40] and it costs on average 20% less than AChEIs.

Despite its efficacy in delaying the onset of AD, Memantine does not interfere with the build up of Aß plaques or tau tangles that appear to drive the disease progression[41], and so while it plays a more neuroprotective role than AChEIs, its effects are more symptomatic than disease-modifying. Altering cholinergic or glutamergic neurotransmission is not enough to alleviate AD; current treatments do not offer a cure, and while they are reasonably effective as palliative solutions, they represent a dead end in research. Drugs that interact more directly with disease progression are required in order to combat Alzheimer's, and there are many new treatments in the works that aim to do just this.

Drugs under development

The majority of research over the last ten years has focused primarily on disrupting the aggregation of Aß, as it is believed that this event is central in AD pathogenesis. Amyloid is the term used to describe an array of fibrillar aggregates that have a ß-pleated-sheet tertiary structure and share certain characteristics. Amyloid-beta is generated from the processing of amyloid precursor protein (APP), a type I transmembrane glycoprotein between 696-770 amino acids long that is naturally produced by many cells and tissues in the body.[42] It runs through the plasma membrane with a long extracellular N-terminal (NH_2) and a shorter cytoplasmic C-terminal (COOH) domain. APP is processed by three proteases, two of which are putative enzymes: α- and γ-secretase are assumed to exist, and there is good evidence that they do, but they have yet to be isolated. There are two main ways in which APP is cut, and the predominant pathway does not lead to Aß production. This non-amyloidogenic pathway first involves the action of α-secretase, which cleaves APP between positions 16-17, leading to the formation of soluble αAPPs and the harmless C83 fragment (an 83-residue C-terminal peptide that remains attached to the membrane and is later degraded by γ-secretase)[43] (Figure 5).

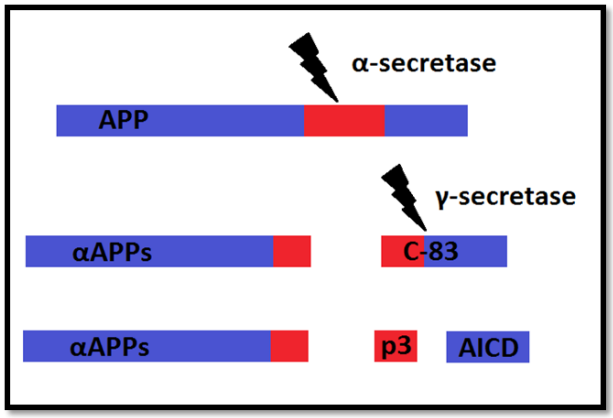

Figure 5: The non-amyloidogenic cleavage of APP, resulting in the formation of harmless fragments.

The second (amyloidogenic) pathway is initiated by ß-secretase, a membrane-anchored protease which cleaves APP to produce soluble ßAPPs and a 99-residue C-terminal fragment that remains membrane-bound and is known as the C99-ßAPP fragment. This product then becomes the substrate for γ-secretase, which generates two Aß peptides – either $Aß_{1-40}$ (40 amino acids long) or $Aß_{1-42}$ (42 amino acids long)[44] (Figure 6).

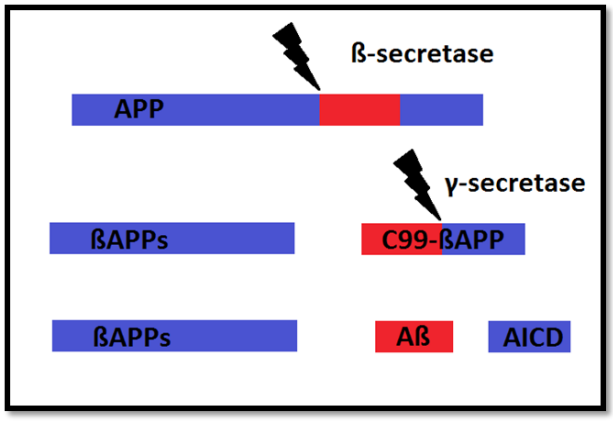

Figure 6: The amyloidogenic cleavage of APP, resulting in the toxic Aß peptide which forms senile plaques.

Ten percent of Aß is the 42-residue variety which forms more rapidly, kills neurons at lower concentrations, and is one hundred times more toxic than $Aß_{1-40}$.[45] $Aß_{1-42}$ disrupts Ca^{2+} regulation, damages mitochondria leading to ROS release[42], and over-activates microglia (the immune cells of the CNS), causing an inflammatory response[46] which escalates damage and can lead to neuronal death.

There is increasing evidence that soluble, globular Aß complexes (amyloid-derived diffusible ligands or ADDLs) and not Aß plaques are the molecular pathogens in AD. Clinical studies show a weak correlation between plaque number and neuron loss[47], whereas soluble Aß levels match more strongly with AD symptoms: seventy times as many ADDLs are present in the brains of Alzheimer's patients than non-sufferers.[48] Behavioural changes occur long before Aß plaque appearance, so it is likely that ADDLs trigger the disease, and plaques

develop and cause damage later on. Researchers believe that the overproduction and decreased clearance of $A\beta_{1-42}$ results in oligimerization, a process in which Aß links in globules of 12-24 molecules, forming ADDLs. These highly toxic ADDLs then bind with high affinity to neuronal receptors, causing memory impairment, synaptic loss, and dementia.[49] Even at highly dilute concentrations, ADDLs interfere with long-term potentiation[50], a process essential for learning and memory. Therefore, it may be more beneficial to direct Aß therapy specifically at ADDLs in the early stages of Alzheimer's.

Regardless of the key amyloid species involved, the basic strategies for combating Aß are essentially the same, and fall into three groups: secretase modulators that inhibit Aß production, immunotherapy which removes Aß once it has already been formed, and treatments that aim to nullify the neurotoxicity of Aß. Many pharmaceutical companies are developing small molecule inhibitors of ß- and γ-secretase that block the ability of these enzymes to cut APP in a way that releases Aß peptides.[51] ß-secretase is a difficult target because it has a large active site, necessitating the use of large molecule inhibitors which are poorly bioavaliable and cannot cross the blood-brain barrier (BBB).[52] However, γ-secretase is a more tenable target. Initially, compounds such as LY-411-575 seemed promising, but at the dose at which Aß was inhibited, the drug had deleterious effects on lymphocyte development, with altered B lymphocyte maturation being observed. In the intestine, LY-411-575 treatment increased goblet cell number and radically altered tissue structure.[53] These undesirable effects are due to the non-APP substrates of γ-secretase. Inhibitors of this enzyme have been shown to block a vital step in Notch processing (Notch is a receptor protein involved in cell-fate decisions during development), so targeting γ-secretase for AD treatment may come with the risks of toxicity caused by reduced Notch signalling.[54] However, it is possible for γ-secretase inhibitors to reduce Aß levels without inducing Notch-mediated toxicity.

For example, a compound known as BMS-299897 has been demonstrated to be fifteen times more effective at preventing cleavage of APP than of Notch in vitro, and in transgenic mice it showed dose dependent reductions of Aß in the brain, cerebrospinal fluid, and plasma. Unlike other γ-secretase inhibitors, BMS-299897 is not associated with a change in lymphocyte or intestinal goblet cell maturation[55], and it looks to be a good candidate for clinical trial.

An alternative to secretase inhibition an approach with limited safety and efficacy is secretase modulation: changing but not blocking the action of enzymes to produce a reduction in the pathogenic isoform $A\beta_{1-42}$ and preferential formation of lower molecular types of Aß.[56] Current research is focused on non-steroidal anti inflammatory drugs, such as Ibuprofen and Curcuminoids[57], which have been shown to lower Aß levels, possibly by means of allosteric interaction with γ-secretase.[52] Unfortunately, a recent trial of tarenflurbil, a modulator of γ-secretase activity, gave discouraging results. Tarenflurbil prevents learning and memory deficits, and reduces $A\beta_{1-42}$ brain concentration in mice[58], and an earlier phase II trial of 210 AD patients who were given either 800mg of Tarenflurbil or placebo showed that those treated with Tarenflurbil had lower rates of decline in global function.[59] Despite these positive results, the phase III randomized, double-blind and placebo-controlled trial which included 1,684 subjects with AD showed that Tarenflurbil had no beneficial effect on primary or secondary outcomes.[60] It could have been the case that even 800mg of Tarenflurbil twice daily was too low a dose, but it is more likely that the reduction of Aß is not enough to prevent neurodegeneration. Perhaps Aß is not the main toxic agent of AD, or perhaps the cascade of damage becomes independent of amyloid burden once a chain of events has been set off by $A\beta_{1-42}$.[61] Either way, this trial raises doubts about the potential of secretase modulation as a therapeutic for Alzheimer's, but these problems may be overcome once these drugs come out of their infancy and become more selective in their inhibition.

A second strategy involves removing the aggregates of Aß by training the immune system to destroy the misfolded proteins after they appear. Vaccines have been used to induce the body to produce antibodies that bind to amyloid and transport it from the brain. It was Dale Schenk and his colleagues at Elan Pharmaceuticals who first demonstrated that active immunization exposing the body to attenuated pathogenic antigens so that it can generate antibodies to fight a future infection with a synthetic version of Aß prevented the development of plaques and reduced the extent and development of AD pathology in APP-transgenic mice.[62] This discovery led to a phase I trial of 80 AD patients who received up to four injections of the AN-1792 vaccine over a period of twenty four weeks. Positive antibody response was observed in almost 60% of treated patients[63], and so a phase II trial was quickly arranged. Unfortunately, the first phase II trial in 2002[64] was suddenly terminated because 6% of the treated sufferers developed meningoencephalitis, acute inflammation of the brain and the membranes which envelop the CNS. There is some dispute over the exact way in which the AN-1792 vaccine worked; at first it was believed that the antibodies formed crossed the blood-brain barrier to form a complex with Aß, triggering microglia to destroy senile plaques. This theory seemed acceptable because the inflammation caused was most likely a result of T cell and microglial over-activation. However, antibodies are typically too large to cross the blood-brain barrier [65], with only 0.05% of antibodies in the blood found in the cerebrospinal fluid too few to activate microglia. The second theory offered was that the AN-1792 antibodies bound to Aß in the blood, causing the blood to act as a sink for the protein, pulling it from the brain and shifting the equilibrium in favour of the soluble protein. Evidence seems to support this idea: after immunization with AN-1792, mice had one thousand times more Aß in their blood, and antibody-amyloid complexes were found in their spleens[66], indicating that the amyloid was being

processed by the immune system. After the failed trial, Elan and Wyeth redesigned the vaccine, and it is currently being tested in nine phase III trials.[56]

Recent trials have focused on humanized monoclonal anti-Aß antibodies instead, aiming to reduce inflammation with passive immunization. Bapineuzumab is a monoclonal antibody directed against the N-terminus of Aß. In preclinical studies with mice, the antibody bound to senile plaques, reduced amyloid burden, and reversed memory deficits.[67] However, a recent phase II trial of Bapineuzumab in which 234 AD patients were randomly assigned either placebo or Bapineuzumab showed no significant differences in efficacy, although Aß burden was reduced by Bapineuzumab.[68] Because of the varying doses administered, and the small sample size, the study was not considered statistically precise enough to support or condemn the efficacy of Bapineuzumab, so the drug has been moved into phase III trials. Other groups argue that the use of antibodies that break up senile plaques could actually exacerbate the symptoms of AD, because the resulting release of soluble Aß would inadvertently increase ADDL levels.[69] Therefore, more specific ADDL-targeting antibodies could prove more effective. The monoclonal antibody NAB61 is capable of causing learning and memory improvement in mice[70], and it is hypothesised that such anti-ADDL antibodies bind to neurons and block the inhibition of long-term potentiation.[71] These drugs also have the potential to reduce tau phosphorylation, showing great promise as future treatments because of their dual attack on AD pathology.

Another line of reasoning for therapeutics was first investigated because of observations that showed a clear link between Alzheimer's and Diabetes Mellitus (DM). Type 2 diabetics have a significantly increased risk of dementia[72], and The Rotterdam study, a large population based study in 1996 showed that, 22.3% of dementia patients have DM[73], and AD may be more frequent in elderly diabetics. A follow up three years later revealed that having DM can almost double the risk of developing Alzheimer's.[74] Clinical studies have also shown that AD sufferers have higher fasting plasma concentrations and lower cerebrospinal fluid concentrations of insulin[75] results that are indicative of insulin resistance. There is a possible cause-effect link between decreased insulin plasma concentrations and the decline of cognitive function, indicating that insulin plays a key role in the regulation of brain activity, and might have a role in AD.[76]

Insulin and insulin receptors (IRs) are present throughout the brain[77], and insulin is actively transported across the blood-brain barrier. IRs are distributed unevenly, and are more abundant in specific brain regions that control high cognition[78], such as the hippocampus and cortex, so they have been implicated in learning and memory formation.[79] These transmembrane receptors are located at the synapse where they regulate neurotransmitter release and play a potential role in synaptic plasticity. Disrupting IR function by the use of streptozotocin injections causes cognitive impairment in rats[80], but insulin injection improves performance[81], further strengthening the link between insulin and dementia.

There is also evidence that insulin directly affects two of the pathological hallmarks of AD: Aß accumulation, and tau phosphorylation. Aß and insulin share a number of properties. Both proteins are amyloidogenic, and because they are both substrates for insulin-degrading enzyme (IDE), they share a common sequence recognition motif, or length of amino acids that interacts with receptors.[84, 85] Aß is also capable of binding to IRs, and it is likely that this binding involves residues 16-25 of Aß, which are identical to residues 21-30 of the ß-chain of insulin the very section involved in insulin binding.[84]

Insulin can increase extracellular Aß accumulation in two ways: by promoting its secretion through increased trafficking of Aß and APP from the Golgi apparatus to the plasma membrane, and by inhibiting its degradation *via* IDE. IDE is the major protease involved in the breakdown of Aß, but insulin directly competes with Aß, thus hastening its accretion into senile plaques.[85] These facts seem contradictory: insulin leads to more extracellular Aß but also improves cognitive function. However, in accelerating Aß secretion, insulin reduces the concentration of intraneuronal Aß, the accumulation of which is thought to trigger the AD disease process, as intracellular beta-amyloid precedes both senile plaques and neurofibrillary tangles.[86] Therefore, the increased secretion of this peptide could actually be beneficial for AD. Insulin and Aß are also linked because they compete for binding to IRs. Both $Aß_{1-40}$ and $Aß_{1-42}$ reduce insulin binding to IRs because they are direct and competitive inhibitors of insulin binding and action; their inhibition has been shown to be specific too, as the reverse sequence $Aß_{40-1}$ does not reduce insulin binding.[84] This inhibition is relieved by increasing insulin levels by infusion, and this approach has been shown to improve memory in AD patients.[87] Because insulin is active as a monomer (although it is stored as a hexamer), it is likely that the species of Aß which bind to IRs are ADDLs, which have been shown to induce a loss of IRs from neuronal dendrites.[88] ADDL binding to synapses triggers oxidative stress, loss of synaptic spines, and the loss of receptors crucial to plasticity and memory, but insulin is able to block ADDL toxicity by preventing its binding and the loss of IRs and synapses which it induces.[89]

The insulin signalling pathway also has a profound effect on tau metabolism. The binding of tau to microtubules is vital for stabilisation, and is regulated through the action of several protein kinases which phosphorylate (add phosphate groups to) tau.[76] When tau is hyperphosphorylated, its affinity for microtubules is reduced, and it becomes the major component of neurofibrillary tangles (NFTs); it is believed that this event is critical in the pathogenesis of AD, as cognitive defects do not usually occur until NFTs develop[90], and the number of NFTs correlates more strongly with the severity of the disease than senile plaques.[91]

Glycogen synthase kinase-3ß (or GSK-3ß) is one kinase that phosphorylates tau in vitro. Its activity is down-

regulated by both insulin and insulin-like growth factor 1 (IGF-1) through the activation of the PI3-kinase pathway. Over-expression of GSK-3ß has been shown to reduce the binding of tau to microtubules[92], but insulin and IGF-1 inhibit GSK-3ß, indirectly dephosphorylating tau and increasing microtubule binding. Another study showed that inducing insulin deficiency using Streptozotocin a drug which is used to produce diabetes causes massive hyperphosphorylation of tau after forty days, but treatment with insulin completely prevented phosphorylation, indicating that changes in tau are attributable to decreased insulin levels in AD patients.[93]

These effects of insulin make a case for the use of anti-diabetic drugs for the treatment of Alzheimer's. Rosiglitazone, an insulin-sensitizing drug, enhances the neuroprotective properties of insulin *in vitro*.[89] A placebo-controlled double-blind trial in which thirty patients were randomly assigned a six month course of either rosiglitazone or placebo found that patients receiving the drug had better recall and attention[94], so it would be prudent to test rosiglitazone further in a larger study. However, rosiglitazone was recently pulled from the UK market because it is associated with myocardial infarction.[95] Another option is troglitazone, an anti-diabetes drug which doubles up as a PPAR-γ receptor agonist. This receptor is involved in insulin sensitizing and inflammation, and when activated by troglitazone, it suppresses inflammation and neurotoxicity caused by Aß by inhibiting stimulation of neuronal-death pathways and microglia.[96] A recent trial with troglitazone showed decreased tau phosphorylation after just twenty four hr, along with modulation of Aß production.[97] Clearly, treatment with insulin or anti-DM drugs in the early stages of Alzheimer's is a therapeutic option which should be urgently studied, as such treatment could prevent the intraneuronal build up of Aß which may trigger the disease, as well as combating the critical phosphorylation of tau, potentially preventing the disease altogether.

CONCLUSION

Clearly, current treatments are not effective enough for sustained treatment of Alzheimer's disease. Acetylcholinesterase inhibitors only provide temporary relief, because they do not completely stop the breakdown of acetylcholine. Therefore, their symptomatic benefits only last as long as the neurotransmitter does. NMDA receptor antagonists may play a more neuroprotective role, and Memantine has been shown to be far more effective than AChEIs, but neither of these approved drugs interferes with amyloid-beta or tau pathology. Although they can slow disease progression in some cases marginally at best current treatments cannot stop or reverse it, and drugs that can do the latter are need for Alzheimer's: drugs that have curative potential.

The treatments that are in development seem to be more efficacious, but actually carry with them some serious side effects, as well as doubts about whether they will even work *in vivo*. Secretase modulators do not seem to be a viable tool because secretases catalyse a wide range of reactions therefore, they may not be safe. Recent trials with γ-secretase modulators have given discouraging results, demonstrating that these drugs are not as efficacious as hoped. The Aß vaccine has also had issues from the start, with dangerous side effects and limited success in clinical trials. Passive immunization seems to be the most elegant approach to AD training the body to remove Aß (as either aggregates or ADDLs) and thus immunizing the patient for an extended period of time. However, if Aß is not the key pathogenic species, then neither of these approaches would work. This is where insulin therapy comes in to play. Insulin regulates both Aß and tau, and has been shown to improve memory in AD patients, prompting some to dub the disease type 3 or brain diabetes. However, this route has yet to be tested rigorously in clinical trials, and it may fall through, as did the Elan AN-1792 vaccine.

Of the three research options presented here, I believe that insulin therapy holds the most promise because of its dual mode of action. Very few treatments work on tau, but this protein may have to be the next target for treatment in the light of recent results indicating that efforts to reduce Aß levels are worthless after the peptide sets off an irreversible cascade of events. There is a great deal of activity in the AD research community, which makes the outlook for a cure somewhat more hopeful, but with a disease as complex as Alzheimer's it could be a long time before effective treatment is found. A cure is currently unlikely because we still do not fully understand which species triggers the disease, and which causes the most damage; as Kenneth Kaitin, head of the Tufts Centre for Study of Drug Development says: 'companies are shooting in the dark'.[98] Perhaps the best approach for AD will prove to be a combination therapy similar to that used for AIDS or leprosy a cocktail of different drugs, each designed to target a different potential cause. In the meantime however, Alzheimer's patients can only be provided with palliative care, so the importance of further research into a permanent cure cannot be overemphasised.

REFERENCES

[1] MacInnes N (2008) '*What is Alzheimer's?*' [Internet], London, Alzheimer's Society. Available at: <http://www.alzheimers.org.uk/site/scripts/documents_info.php?categoryID=200171&documentID=100&gclid=CPPVxsqbvqMCFRf92Aod61iheA> [Accessed 16 August 2010].

[2] Gull D (2010) Alzheimer's Disease, *Biological Sciences Review* **22**(4), 34.

[3] Voss S (2002) The Winter Years: Understanding Ageing, *Psychology Review* **8**(3), 28.

[4] Gull D (2010) Alzheimer's Disease, *Biological Sciences Review* **22**(4), 35.

[5] Gull, D (2010) Alzheimer's Disease, *Biological Sciences Review* **22**(4), 36.

[6] Zhu X, Raina A, Smith M (1999) Cell Cycle Events in Neurons: Proliferation or Death?, *The American Journal of Pathology* **155**(2), 327-329.

[7] Deretic V, Klionsky D (2008) How Cells Clean House, *Scientific American* **298**(5), 52.

[8] Kempermann G, Gage F (1999) New Nerve Cells for the Adult Brain, *Scientific American* **280**(5), 38.

[9] Tuite M, Staniforth G (2009) Protein Misfolding Diseases, *Biological Sciences Review* **22**(1), 22.

[10] Hong M, Lee V (1997) Insulin and Insulin-like Growth Factor-1 Regulate Tau Phosphorylation in Cultured Human Neurons, *The Journal of Biological Chemistry* **272** (31), 19547.

[11] St. George-Hyslop P (2000) Piecing Together Alzheimer's, *Scientific American* **283**(6), 58-59.

[12] Todd A, Moore A, Ashton M, Van S (2010) Current Research and Development of Treatments for Alzheimer's Disease, *The Pharmaceutical Journal* **284**(7599), 429.

[13] Palmer A (2007) Drugs for Dementia, *Education In Chemistry* **44**(1), 20.

[14] Francis P, Palmer A, Sims N, Bowen D, Davison A, Esiri M, Neary D, Snowden J, Wilcock G (1985) *The New England Journal of Medicine* **313**(1), 7.

[15] Khachaturian Z, Radebaugh T (ed.) (1996) Alzheimer's Disease: Cause(s), Diagnosis, Treatment, and Care, *CRC Press*, p. 258.

[16] Powers 'Acetylcholine' [Internet], Illinois, Chemistry Explained. Available at: <http://www.chemistryexplained.com/A-Ar/Acetylcholine.html> [Accessed 18 August 2010].

[17] Farlow M (2004) NMDA Receptor Antagonists: A New Therapeutic Approach for Alzheimer's Disease. *Geriatrics* **59** (6), 28.

[18] Mann J (2006) Medicinal Compounds, *Education In Chemistry* **43**(5), 118.

[19] Khachaturian Z, Radebaugh T (ed.) (1996) Alzheimer's Disease: Cause(s), Diagnosis, Treatment, and Care, *CRC Press*, pp. 260-261.

[20] Khachaturian Z, Radebaugh T (ed.) (1996) Alzheimer's Disease: Cause(s), Diagnosis, Treatment, and Care, *CRC Press*, pp. 259-260.

[21] Woodruff-Pak, D, Vogel R, Wenk, G (2001) Galantamine: effect on nicotinic receptor binding, acetylcholinesterase inhibition, and learning [Abstract only], *Proceedings of the National Academy of Sciences of the United States of America* **90**(4).

[22] Samochocki M, Höffle A, Fehrenbacher A, Jostock R, Ludwig J, Christner C, Radina M , Zerlin M, Ullmer C, Pereira E, Lübbert H, Albuquerque E, Maelicke A (2003) Galantamine Is an Allosterically Potentiating Ligand of Neuronal Nicotinic but Not of Muscarinic Acetylcholine Receptors, *The Journal of Pharacology and Expermental Therapeutics* **305**(3), 1024-1036.

[23] Gull D (2010) Alzheimer's Disease, *Biological Sciences Review* **22**(4), 37.

[24] Farlow M (2004) NMDA Receptor Antagonists: A New Therapeutic Approach for Alzheimer's Disease, *Geriatrics* **59**(6), 22.

[25] Shurkin J (2009) Decoding Dementia, *Scientific American Mind* **20**(6), 61.

[26] Roberts K (2000) Alzheimer's Disease: Forget the Past, Look to the Future, *Education in Chemistry* **37**(3), 3.

[27] Palmer A (2007) Drugs for Dementia, *Education In Chemistry* **44**(1), 15.

[28] Khachaturian Z, Radebaugh T (ed.) (1996) Alzheimer's Disease: Cause(s), Diagnosis, Treatment, and Care, *CRC Press*, pp. 260.

[29] Mirza R, Peters D, Sparks R (2003) Xanomeline and the antipsychotic potential of muscarinic receptor subtype selective agonists [Abstract only], *CNS Drug Reviews* **9**(2).

[30] Tsien J (2007) Memory Code, *Scientific American* **297**(1), 41.

[31] Farlow, M (2004) NMDA Receptor Antagonists: A New Therapeutic Approach for Alzheimer's Disease, *Geriatrics* **59**(6), 29.

[32] Khachaturian Z, Radebaugh T (ed.) (1996) Alzheimer's Disease: Cause(s), Diagnosis, Treatment, and Care, *CRC Press*, pp. 266.

[33] Doble, A (1999) The role of excitotoxicity in neurodegenerative disease: implications for therapy [Abstract only], *Pharmacology and Therapeutics* **81**(3).

[34] Farlow M (2004) NMDA Receptor Antagonists: A New Therapeutic Approach for Alzheimer's Disease, *Geriatrics* **59**(6), 24-25.

[35] Farlow M (2004) NMDA Receptor Antagonists: A New Therapeutic Approach for Alzheimer's Disease, *Geriatrics* **59**(6), 25.

[36] Reisberg B, Doody R, Stöffler A, Schmitt F, Ferris F (2003) Memantine in Moderate-to-Severe Alzheimer's Disease, *New England Journal of Medicine* **348**(14), 1333-1341.

[37] Tariot P, Farlow M, Grossberg G, Graham S, McDonald S, Gergel I (2004) Memantine treatment in patients with moderate to severe Alzheimer disease already receiving donepezil: a randomized controlled trial [Abstract only], *The Journal of the American Medical Association* **291**(3).

[38] Winblad B, Poritis N (1999) Memantine in severe dementia: results of the 9M-Best Study (Benefit and efficacy in severely demented patients during treatment with Memantine) [Abstract only], *International Journal of Geriatric Psychiatry* **14**(2).

[39] Rountree S, Chan W, Pavlik V, Darby E, Siddiqui S, Doody R (2009) Persistent Treatment With Cholinesterase Inhibitors and/or Memantine Slows Clinical Progression of Alzheimer Disease, *Alzheimer's Research* and *Therapy* **1**(2), 7.

[40] Farlow M (2004) NMDA Receptor Antagonists: A New Therapeutic Approach for Alzheimer's Disease, *Geriatrics* **59** (6), 26.

[41] Stix G (2010) Alzheimer's: Forestalling the Darkness, *Scientific American* **302**(6), 35.

[42] St. George-Hyslop P (2000) Piecing Together Alzheimer's, *Scientific American* **283**(6), 56.

[43] Gasparini L, Netzer W, Greengard P, Xu H (2002) Does Insulin Dysfunction Play a Role in Alzheimer's Disease?, *Trends in Pharmacological Sciences* **23**(6), 289.

[44] Estrada L, Soto C (2007) Disrupting ß-Amyloid Aggregation for Alzheimer Disease Treatment, *Current Topics in Medicinal Chemistry* **7**(1), 116.

[45] Hoshi M, Sato M, Matsumoto S, Noguchi A, Yasutake K, Natsuko Y (2003) Spherical aggregates of ß-amyloid (amylospheroid) show high neurotoxicity and activate tau protein kinase I/glycogen synthase kinase-3ß, *Proceedings of the National Academy of Sciences* **100**(11), 6370-6375.

[46] Krause D, Müller N (2010) Neuroinflammation, Microglia and Implications for Anti-Inflammatory Treatment in Alzheimer's Disease. *International Journal of Alzheimer's Disease* pp. 1-9.

[47] Terry R, Masliah E, Salmon D, Butters N, DeTeresa R, Hill R, Hansen L, Katzman R (1991) Physical basis of cognitive alterations in Alzheimer's disease: synapse loss is the major correlate of cognitive impairment [Abstract only], *Annals of Neurology* **30**(4).

[48] Freeman K (2003) ADDLs: A New Explanation for Alzheimer Disease?, *Environmental Health Perspectives* **111** (15), 810.

[49] Catalano S, Dodson E, Henze D, Joyce J, Krafft G, Kinney G (2006) The Role of Amyloid-Beta Derived Diffusible Ligands (ADDLs) in Alzheimer's Disease, *Current Topics in Medicinal Chemistry* **6**(6), 598.

[50] Northwestern University (1998) Toxic Protein That Interferes With Brain Signals May Trigger Onset Of Alzheimer's Disease. *Science Daily.* Available at:<http://www.sciencedaily.com/releases/1998/05/980528065052.htm> [August 21 2010].

[51] Stix G (2010) Alzheimer's: Forestalling the Darkness, *Scientific American* **302**(6), 38.

[52] Catalano S, Dodson E, Henze D, Joyce J, Krafft G, Kinney G (2006) the Role of Amyloid-Beta Derived Diffusible Ligands (ADDLs) in Alzheimer's Disease, *Current Topics in Medicinal Chemistry* **6**(6) 602.

[53] Wong G, Manfra D, Poulet F, Zhang Q, Josien H, Bara T, Engstrom L, Pinzon-Ortiz M, Fine J, Lee H, Zhang L, Higgins G, Parker E (2003) Chronic Treatment with the γ-Secretase Inhibitor LY-411,575 Inhibits β-Amyloid Peptide Production and Alters Lymphopoiesis and Intestinal Cell Differentiation [Abstract only], *The Journal of Biological Chemistry* **279**(13).

[54] De Strooper B, Annaert W, Cupers P, Saftig P, Craessaerts K, Mumm J, Schroeter E, Schrijvers V, Wolfe M, Ray W, Goate A, Kopan R (1999) A presenilin-1-dependent gamma-secretase-like protease mediates release of Notch intracellular domain [Abstract only], *Nature* **398**(6727).

[55] Barten D, Guss V, Corsa J, Loo A, Hansel S, Zheng M, Munoz B, Srinivasan K, Wang B, Robertson B, Polson C, Wang J, Roberts S, Hendrick J, Anderson J, Loy J, Denton R, Verdoorn T, Smith D, Felsenstein K (2005) Dynamics of β-Amyloid Reductions in Brain, Cerebrospinal Fluid, and Plasma of β-Amyloid Precursor Protein Transgenic Mice Treated with a γ-Secretase Inhibitor, *The Journal of Pharmacology and Experimental Therapeutics* **312**(2), 635-643.

[56] Todd A, Moore A, Ashton M, Van S (2010) Current Research and Development of Treatments for Alzheimer's Disease, *The Pharmaceutical Journal* **284**(7599), 430.

[57] Stix G (2007) Spice Healer, *Scientific American* **296**(2), 54-57.

[58] Kukar T, Prescott S, Ericksen J, Holloway V, Murphy M, Koo E, Golde T, Nicolle M (2007) Chronic administration of R-flurbiprofen attenuates learning impairments in transgenic amyloid precursor protein mice [Abstract only], *BMC Neuroscience* **8**(54).

[59] Wilcock G, Black S, Hendrix S, Zavitz K, Swabb E, Laughlin M (2008) Efficacy and safety of tarenflurbil in mild to moderate Alzheimer's disease: a randomised phase II trial [Abstract only], *Lancet Neurology* **7**(6).

[60] Green R, Schneider L, Amato D, Beelen A, Wilcock G, Swabb E, Zavitz K (2009) Effect of tarenflurbil on cognitive decline and activities of daily living in patients with mild Alzheimer disease: a randomized controlled trial [Abstract only], *The Journal of the American Medical Association* **302**(23).

[61] Prins N, Visser P, Scheltens P (2010) Can Novel Therapeutics Halt the Amyloid Cascade?, *Alzheimer's Research* and *Therapy* **2**(2), 5.

[62] Schenk D, Barbour R, Dunn W, Gordon G, Grajeda H, Guido T, Hu K, Huang J, Johnson-Wood K, Khan K, Kholodenko D, Lee M, Liao Z, Lieberburg I, Motter R, Mutter L, Soriano F, Shopp G, Vasquez N, Vandevert C, Walker S, Wogulis M, Yednock T, Games D, Seubert P (1998) Immunization With Amyloid-ß Attenuates Alzheimer-Disease-Like Pathology in the PDAPP Mouse [Abstract only], *Nature* **400**, 6740.

[63] Bayer A, Bullock R, Jones R, Wilkinson D, Paterson K, Jenkins L, Millais S, Donoghue S (2005) Evaluation of the Safety and Immunogenicity of Synthetic Abeta42 (AN1792) in Patients with AD [Abstract only], *Neurology* **64**(1).

[64] Orgogozo J, Gilman S, Dartigues J, Laurent B, Puel M, Kirby L, Jouanny P, Dubois B, Eisner L, Flitman S, Michel B, Boada M, Frank A, Hock C (2003) Subacute Meningoencephalitis in a Subset of Patients with AD After Abeta42 Immunization [Abstract only], *Neurology* **61**(1).

[65] Ferber D (2007) Bridging the Blood-Brain Barrier: New Methods Improve the Odds of Getting Drugs to the Brain Cells That Need Them, *PLoS Biology* **5**(6), 1191-1194.

[66] Martindale D (2002) Peeling Plaque, *Scientific American* **286**(5), 12-13.

[67] Bard F, Cannon C, Barbour R, Burke R, Games D, Grajeda H, Guido T, Hu K, Huang J, Johnson-Wood K, Khan K, Kholodenko D, Lee M, Lieberburg I, Motter R, Nguygen M, Soriano F, Vasquez N, Weiss K, Welch B, Seubert P, Schenk D, Yednock T (2000) Peripherally administered antibodies against amyloid beta-peptide enter the central nervous system and reduce pathology in a mouse model of Alzheimer disease [Abstract only], *Nature Medicine* **6**(8).

[68] Salloway S, Sperling R, Gilman S, Fox N, Blennow K, Raskind M, Sabbagh M, Honig L, Doody R, van Dyck C, Mulnard R, Barakos J, Gregg K, Liu E, Lieberburg I, Schenk D, Black R, Grundman M (2000) A Phase 2 Multiple Ascending Dose Trial of Bapineuzumab in Mild to Moderate Alzheimer Disease [Abstract only], *Neurology* **73**(24).

[69] Catalano S, Dodson E, Henze D, Joyce J, Krafft G, Kinney G (2006) The Role of Amyloid-Beta Derived Diffusible Ligands (ADDLs) in Alzheimer's Disease, *Current Topics in Medicinal Chemistry* **6**(6), 603.

[70] Ma Q, Lim G, Harris-White M, Yang F, Ambegaokar S, Ubeda O, Glabe C, Teter B, Frautschy S, Cole G (2006) Antibodies Against Beta-Amyloid Reduce Abeta Oligomers, Glycogen Synthase Kinase-3Beta Activation and Tau Phosphorylation in Vivo and in Vitro [Abstract only], *Journal of Neuroscience Research* **83**(3).

[71] Barghorn S, Nimmrich V, Striebinger A, Krantz C, Keller P, Janson B, Bahr M, Schmidt M, Bitner R, Harlan J, Barlow E (2005) Globular Amyloid ß-Peptide$_{1-42}$ Oligomer - a Homogenous and Stable Neuropathological Protein in Alzheimer's Disease, *Journal of Neurochemistry* **95**(3), 834-847.

[72] Leibson C, Rocca W, Hanson A, Cha R, Kokmen E, O'Brien C, Palumbo P (1997) Risk of Dementia Among Persons with Diabetes Mellitus: A Population-Based Cohort Study, *American Journal of Epidemiology* **145**(4), 301-308.

[73] Ott A, Stolk R, Hofman A, van Harskamp F, Grobbee D, Breteler M (1996) Association of Diabetes Mellitus and Dementia: The Rotterdam Study [Abstract only], *Diabetologia* **39**(11).

[74] Ott A, Stolk R, van Harskamp F, Pols H, Hofman A, Breteler M (1999) Diabetes Mellitus and the Risk of Dementia: The Rotterdam Study [Abstract only], *Neurology* **53**(9).

[75] Craft S, Peskind E, Schwartz M, Schellenberg G, Rasking M, Porte D (1998) Cerebrospinal Fluid and Plasma Insulin Levels in Alzheimer's Disease: Relationship to Severity of Dementia and Apolipoprotein E Genotype [Abstract only], *Neurology* **50**(1).

[76] Gasparini L, Netzer W, Greengard P, Xu H (2002) Does Insulin Dysfunction Play a Role in Alzheimer's Disease? *Trends in Pharmacological Sciences* **23**(6), 290.

[77] Schulingkamp R, Pagano T, Hung D, Raffa R (2000) Insulin Receptors and Insulin Action in the Brain: Review and Clinical Implications [Abstract only], *Neuroscience and Behavioural Reviews* **24**(8).

[78] Zhao W, Alkon D (2001) Role of Insulin and Insulin Receptor in Learning and Memory [Abstract only], *Molecular and Cellular Endocrinology* **177**(1-2).

[79] Zhao W, Chen H, Xu H, Moore E, Meiri N, Quon M, Alkon D (1999) Brain Insulin Receptors and Spatial Memory. Correlated Changes in Gene Expression, Tyrosine Phosphorylation, and Signalling Molecules in the Hippocampus of Water Maze Trained Rats, *The Journal of Biological Chemistry* **274**(49), 34893-34902.

[80] Lannert H, Hoyer S (1998) Intracerebroventricular Administration of Streptozotocin Causes Long-Term Diminutions in Learning and Memory Abilities and in Cerebral Energy Metabolism in Adult Rats [Abstract only], *Behavioural Neuroscience* **112**(5).

[81] Park C, Seeley R, Craft S, Woods S (2000) Intracerebroventricular Insulin Enhances Memory in a Passive-Avoidance Task [Abstract only], *Physiology and Behaviour* **68**(4).

[82] Kurochkin I (1998) Amyloidogenic Determinant as a Substrate Recognition Motif of Insulin-Degrading Enzyme [Abstract only], *FEBS Letters* **427**(2).

[83] Turnell W, Finch J (1992) Binding of the Dye Congo Red to the Amyloid Protein Pig Insulin Reveals a Novel Homology Amongst Amyloid-Forming Peptide Sequences [Abstract only], *The Journal of Molecular Biology* **227**(4).

[84] Xie L, Helmerhorst E, Taddei K, Plewright B, Van Bronswijk W, Martins R (2002) Alzheimer's ß-Amyloid Peptides Compete for Insulin Binding to the Insulin Receptor, *The Journal of Neuroscience* **22**(10),1-5.

[85] Gasparini L, Gouras G, Wang R, Gross R, Beal M, Greengard P, Xu H (2001) Stimulation of Beta-Amyloid Precursor Protein Trafficking by Insulin Reduces Intraneuronal Beta-Amyloid and Requires Mitogen-Activated Protein Kinase Signalling [Abstract only], *The Journal of Neuroscience* **21**(8).

[86] Gouras G, Tsai J, Naslund J, Vincent B, Edgar M, Checler F, Greenfield J, Haroutunian V, Buxbaum J, Xu H, Greengard P, Relkin N (2000) Intraneuronal Abeat42 Accumulation in Human Brain [Abstract only], *The American Journal of Pathology* **156**(1).

[87] Craft S, Asthana S, Newcomer J, Wilkinson C, Matos I, Baker L, Cherrier M, Lofgreen C, Latendresse S, Petrova A, Plymate S, Rasking M, Grimwood K, Veith R (1999) Enhancement of Memory in Alzheimer Disease with Insulin and Somatostatin but Not Glucose [Abstract only], *Archives of General Psychiatry* **56**(12).

[88] Zhao W, De Felice F, Fernandez S, Chen H, Lambert M, Quon M, Krafft G, Klein W (2008) Amyloid Beta Oligomers Induce Impairment of Neuronal Insulin Receptors [Abstract only], *The FASEB Journal* **22**(1).

[89] De Felice F, Vieira M, Bomfim T, Decker H, Velasco P, Lambert M ,Viola K, Zhao W, Ferreira S, Klein W (2009) Protection of Synapses Against Alzheimer's-linked Toxins: Insulin Signalling Prevents the Pathogenic Binding of Abeta Oligomers, *Proceedings of the National Academy of Sciences of the United States of America* **106**(6), 1972-1973.

[90] McKee A, Kosik K, Kowall N (1991) Neuritic Pathology and Dementia in Alzheimer's Disease [Abstract only], *Annals of Neurology* **30**(2).

[91] Arriagada, P, Growdon, J, Hedley-Whyte, E, Hyman, B (March 1992) Neurofibrillary Tangles but Not Senile Plaques Parallel Duration and Severity of Alzheimer's Disease [Abstract only], *Neurology* **42**(3pt1).

[92] Hong M, Lee V (1997) Insulin and Insulin-like Growth Factor-1 Regulate Tau Phosphorylation in Cultured Human Neurons, *The Journal of Biological Chemistry* **272**(31), 19551.

[93] Planel E, Tatebayashi Y, Miyasaka T, Liu L, Wang L, Herman M, Yu W, Luchsinger J, Wadzinski B, Duff K, Takashima A (2007) Insulin Dysfunction Induces in Vivo Tau Hyperphosphorylation Through Distinct Mechanisms, *The Journal of Neuroscience* **27**(50),13635-13648.

[94] Watson G, Cholerton B, Reger M, Baker L, Plymate S, Asthana S, Fishel M, Kulstad J, Green P, Cook D, Kahn S, Keeling M, Craft S (2005) Preserved Cognition in Patients with Early Alzheimer Disease and Amnestic Mild Cognitive Impairment During Treatment with Rosiglitazone: a Preliminary Study [Abstract only], *The American Journal of Geriatric Psychiatry* **13**(11).

[95] Mitchell D (2010) *'FDA Places Restrictions on Rosiglitazone Because of Safety Concerns'* [Internet], Kansas, American Academy of Family Physicians. Available at: <http://www.aafp.org/online/en/home/publications/news/news-now/health-of-the public/20100924 avandiarestrictions.html> [Accessed 26th September 2010].

[96] Combs C, Jonhson D, Karlo J, Cannady S, Landreth G (2000) Inflammatory Mechanisms in Alzheimer's Disease: Inhibition of Beta-Amyloid-Stimulated Proinflammatory Responses and Neurotoxicity by PPARgamma Agonists [Abstract only], *The Journal of Neuroscience* **20**(2).

[97] D'Abramo C, Ricciarelli R, Pronzato M, Davies P (2006) Troglitazone, a Peroxisome Proliferator-Activated Receptor-Gamma Agonist, Decreases Tau Phosphorylation in CHOtau4R Cells [Abstract only], *Journal of Neurochemistry* **98**(4)

[98] Yahoo! Finance (2010) *'Lilly's Alzheimer Flop Prods Collaboration.'* Yahoo! Available at: <http://finance.yahoo.com/news/Lillys-Alzheimer-Flop-Prods-ibd-2269153187.html?x=0 [Accessed 6th September 2010].