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Alzheimer's disease: Vaccination strategies using antibodies against ß-amyloid plaques: a cure?

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Aaron completed this review as part of a paper in the second semester in second year (2007). It has personal significance as he explains below.

"I have always been interested in the brain and how this one single organ ultimately makes us who we are, my fascination in Alzheimer's disease stems from ever since my grandma was diagnosed with it in 2001. Since then I have observed the progression of the disease on her, her long term memories have started to deteriorate recently as she is at the stage where she is unable to recognise people and thus my interest in the pathogenesis of the disease. As there is still no cure for AD, this is an area where I have always tried to keep up to date with, and this hence provided me with the starting point for my review."

Abstract

The utilisation of concepts of immunotherapy on ß-amyloid plaques involved in Alzheimer's disease has been a focus of major research over the recent years. Using original articles of studies conducted in this area, this paper will examine the feasibility and some of the key milestones that have been critical to this newly expanding area of research, in the bid to offer hope of finding a cure for some of the millions of Alzheimer sufferers worldwide.

Introduction

Alzheimer's disease (AD) is a neurodegenerative disorder which presents with a progressive loss in cognitive function, impairments in memory and eventual loss of learned motor skills leading to death. Pathological hallmarks that are commonly seen in AD affected brains include loss of neurons, formation of ß-amyloid in neuritic plaques (containing abnormal insoluble aggregates of the ß-amyloid peptide) and neurofibrillary tangles[1]. These plaques are often found in large amounts in the association and limbic cortices, and are usually associated with activated microglia and astrocytes, adjacent to the core and surrounding the plaque respectively. In particular, the more hydrophobic species of ß-amyloid, ending at amino acid 42, $Aß_{42}$, predominates in these plaques, prompting current research to focus on this particular species[1].

As there is no cure for AD, the majority of treatment at the present time as part of the New Zealand guidelines focuses on cholinesterase inhibitors that aim at prolonging the action of Acetylcholine in the brain at the synapses. This thereby provides only temporary alleviation of symptoms, slowing the rate of decline; such treatments include Rivastigmine (Exelon) and Donepezil (Aricept)[2,3].

In New Zealand today, approximately 38,000 people are affected with dementia, and of that approximately 50-60% constitutes those affected by AD[4]. Although the cause of the disease is, at present, still unknown, a number of risk factors have been identified for this debilitating disease; gender, Apolipoprotein E genetic status, those suffering from Down syndrome and long term occupational exposure to solvents being a few, but the most crucial and notable being age[2]. It is a disease that predominantly affects those over the age of 65, with the risk of being diagnosed with dementia beginning at approximately 1% at the age of 60-64, rising 1-2% a year until it approaches approximately 30-50% by the age of 85. As the generation of post war baby boomers succeed the current elderly population, the proportion of those aged 65+ in New Zealand have been projected to increase to approximately 25% in 2050, almost double that of the current number[2,4]. It can hence be deduced that Alzheimer's disease will take on a significant burden with respects to New Zealand's health care system in the upcoming decades, calling forth the need for potential interventions to alleviate this impending burden.

The ß-amyloid hypothesis - the basis of current research

The ß-amyloid hypothesis establishes that the ß-amyloid protein (Aß) plays a causative role in AD, due to its neurotoxic properties ultimately causing neuronal degeneration. This is supported by the genotype-phenotype observation in the rare, autosomal dominant familial form of AD, in which mutations in the Amyloid Precursor Protein (APP), Presenilin 1 and Presenilin 2 (enzymes which are involved in the cleavage and processing of APP) lead to an increase in the production of the hydrophobic-42 amino acid form of Aß that predominate in plaques seen in the disease[1]. A study[5] conducted in 2005 also stands to support the claim of the significant role of Aß in AD by attempting to explain the mechanism by which ß-amyloid immunotherapy would be beneficial. Using transgenic mice models, it was observed that both passive and active immunisation resulted in the lowering of Aß levels, providing protection against the loss of synaptophysin (a presynatic vesicle protein found in the hippocampus and association cortices). This also prevented synaptic degeneration, a crucial process believed to be related to AD associated cognitive decline[5], evidence which consequently strengthens the ß-amyloid hypothesis.

Immunotherapy in transgenic mice models - the beginning of an era

Any assessment of immunotherapy in general necessitates the distinction between two crucial concepts; active and passive immunisation. The former involves immunisation with a specific antigen, in this case Aß peptide, which consequently results in an immune response, directed towards the antigen administered, from the host. This compares to the concept of passive immunisation whereby immune-mediated clearance of Aß is promoted

through the regular administration of anti-Aß antibodies.

The possibility of immunotherapy stemmed from the work conducted by Schenk et al. involving $Aß_{42}$ immunisation of both young and old PDAPP transgenic mice models. It was observed that mice which were immunised at the age of 6 weeks did not develop ß-amyloid plaque formation, astrogliosis or neuritic dystrophy[6], essentially preventing the development of the symptoms of Alzheimer's despite having a predisposition. It was also noted that treatment of the older mice (immunised at 11 months) had a lesser extent of the neuropathological hallmarks that accompanied AD. In addition, immunoglobulins (IgG) were also discovered around plaques following the use of immunohistochemistry[6], effectively sparking a new wave of interest as to the possibility of a cure of AD via this pathway.

Clinical trials of Aß immunisation

The success in stimulating the clearance of ß-amyloid plaques in these animal transgenic models, without any reported adverse effects, thus prompted the commencement of clinical trials using an aggregated $Aß_{1-42}$ peptide (AN-1792) combined with QS-21 as an adjuvant. After establishing the safety and tolerability of the vaccine with phase I trials without any significant problems, phase IIA for the same vaccine was launched involving 372 mild-to-moderate AD patients. This trial was also aimed to test the pilot efficacy in addition to the safety and tolerability of the vaccine, given at 0, 1, 3, 6, 9 and 10 months. This was however prematurely halted when 18 of the 298 participants who were given the intramuscular injection of AN1792 (6%) were diagnosed with aseptic meningoencephalitis (not observed in the 72 controls who received saline solution)[7]. Although it was eventually concluded that postvaccination meningoencephalitis occurred without any relation to the AN1792 vaccine itself and the presence of positive antibody titres, it was identified that microglial activation as well as a T cell mediated response could have been responsible[7,8], leading to a shift of focus towards the need to develop safer vaccines that avoided this pro-inflammatory response to evade such adverse associations in future clinical trials.

Another key study[9] was also conducted assessing the effect of antibodies in slowing the progression of AD. This involved a cohort of 28 patients from the original trial conducted by Orgozozo and colleagues, who, over a period of a year, received a prime and a booster immunisation of aggregated $Aß_{42}$. Results of the study showed that 20 of the patients who subsequently generated antibodies against Aß had a considerably slower rate of decline in cognitive function as assessed by a number of mental function tests, such as the Mini Mental State Examination (MMSE), when compared to patients in the placebo group. Although these results are promising, a number of limitations existed in this study. The sample size may not be large enough or representative if the data were to be extrapolated to the general population thereby requiring caution if this was to be done. Another concern raised was that the original trial also encountered early termination, after only 19.7% of participants in total had developed an antibody response, consequently calling for the need for future trials to be of a longer duration to in order to fully assess the efficacy of the vaccine.

The development and progress of a safer vaccine - Th1 vs. Th2 immune responses

The assumption of the involvement of T-cell mediated response via the Th1 pro-inflammatory pathway causing meningoencephalitis has also shifted the focus towards the creation of a safer vaccine ideally aimed at avoiding this pro inflammatory pathway and utilising a Th2 response involving antibody production. Supporting this statement were results from a crucial study conducted in 2003[8], examining the effects of neurotoxicity associated inflammation. These results found nitric oxide release, mediated by microglia (resident immune cells of the CNS), by Aß-reactive Th1 cells contributed to AD neurotoxicity, nonetheless this toxic effect was observed to be counterbalanced by activity of the Th2 cells[8]. This issue has been addressed in a number of studies, one of these being a key trial conducted employing the use of an adenovirus vector encoding 11 tandem repeats of $Aß_{1-6}$ (a more robust immunogen than $Aß_{1-42}$ vaccines)[10]. An immune response polarised towards an anti-inflammatory Th2 type response was able to be observed with immunoglobulin isotyping via this vaccine, with much higher titres being observed when co-administered with additional adenovirus vector encoding granulocyte-macrophage colony stimulating factor (GM-CSF) that served to increase its immunogenecity[10]. This has ultimately allowed for the option of a safer alternative to peptide-based vaccines but nonetheless, as with all other research conducted with animal models, caution is necessary before proceeding into clinical trials.

The development and progress of a safer vaccine - routes of administration and adjuvant toxicity

Apart from problems regarding Th1 immune responses, concerns have also been raised with regards to the toxicity of adjuvants prompting further exploration. In order to address both of these issues of adjuvant toxicity, a study was conducted by Asuni and colleagues using nonfibrillogenic, non-toxic Aß homologous peptides (K6Aß 1-30-NH2) and alum adjuvant (which had lowered toxicity) given by active immunisation. Results provided strong evidence promoting its use as a decrease in total ß-amyloid deposit burden was still seen in addition to the fact that neither alteration in vascular Aß nor increase in cerebral bleeding occurred[11]. Accordingly, this could possibly thus be another safer alternative when compared to the usual peptide-based vaccines.

Other concerns in relation to routes of administration being associated with microhaemorrhages observed in some studies utilising Aß immunotherapy have also been raised. It is important to note that in the majority of cases, Aß deposition not only occurs in the brain parenchyma, but also in the cerebral vasculature; known as cerebral amyloid angiopathy (CAA)[12]. In a study conducted by Ranke et. al., age dependant accumulation of Aß in isolated cerebral vessels have been shown to occur in PDAPP transgenic mice; it's composition being quite similar to that seen in sporadic AD in humans. The same study then examined administration of various types of monoclonal antibodies in the same models via passive immunisation, with results showing that N-terminal antibodies 3D6 and 10D5 were able to bind to their epitopes in the Aß peptide regardless of aggregational state; this was ultimately associated with increased microhaemorrhages and exarcebation of CAA already present[12].

Increased microhaemorrhages have also recently been shown to be associated with active immunisation by Wilcock and colleagues as part of a study investigating Aß vaccinations in comparison to non steroidal anti-inflammatory drug treatment and its effects on Aß deposition in the brain. In this case however, transgenic APP + PS1 mice were used and active immunisation performed using $Aß_{1-42}$. Although results showed a decrease in Aß depositon, increased levels of vascular amyloid and hence CAA as well as microhaemorrhages were also noted[13]. Both these studies consequently raises questions requiring the need for further experimentation with routes of administration of vaccines (both passive and active) in the attempts to avert such adverse effects; nonetheless, comparisons between both are difficult due to the different methodologies used. Moreover, it is also essential to remember that besides the varying routes of administration, combinations of adjuvants and the type of vaccine used would also ultimately play a role in the overall outcome and the interplay of negative side effects which are to be avoided, and as research continues to progress for a potential cure, all of these need to be taken into account.

Conclusion

With the world's continued aging population, the idea of using Aß immunotherapy in preventing the formation of plaques and also promoting the clearance of plaques from the brain is highly appealing, having the potential as a cure for this debilitating disease affecting many worldwide. Despite the fact that clinical trials have been halted and concerns raised, research in this area on transgenic animal models look promising with advances being made in reducing the AD related pathology of Aß plaques, as well as the creation of other safer alternatives besides the usual peptide

based vaccines. It is worth noting however, that current research relying on transgenic animal models may not be truly reflective of its efficacy in humans, as mouse Aß is only approximately 96% homologous to human Aß[14], thereby calling the need for the use and creation of better models whenever possible, which would undoubtedly aid clinical trials in the future. Further clinical trials should also proceed with caution as future directions in this area head towards the investigation of different routes of administration as well as experimentation with active and passive immunisation agents in the attempts to discover a safe vaccine for use in humans. Investigations to unravel the mystery of the mechanisms involved in the functioning of Aß immunotherapy may also be another area of development, in order to fully understand the pathways involved and to help minimise unwanted adverse effects such as inflammation and mircrohaemorrhages with use of such vaccines.

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