DNA Aβ42 Vaccination as Possible Alternative Immunotherapy for Alzheimer Disease

Immunotherapy for Alzheimer disease was advanced in reports[1,2] showing that the use of Aβ42 peptide vaccination in a transgenic mouse model of Alzheimer disease significantly reduced the Aβ42 plaque count, significantly reduced levels of gliosis, and significantly improved behavior. Based on the positive experimental animal model data, a human Aβ42 peptide immunization clinical trial [ie, the AN1792(QS-21)-201 trial][3] for patients with Alzheimer disease was conducted; however, it was stopped owing to significant adverse effects in 6% of immunized patients. Subsequent studies showed that there were significant reductions in Aβ42-containing plaque in the brain but that the patients continued to become more demented.[4] For patients with mild Alzheimer disease, the use of passive immunotherapy with the humanized monoclonal antibody bapineuzumab resulted in no significant clinical benefit, but the use of passive immunotherapy with the humanized monoclonal antibody solanezumab resulted in some memory and behavioral benefits (*New York Times*, July 24, 2012). Intravenous immunoglobulins, which probably contain anti-Aβ antibodies, have also been used to treat patients with Alzheimer disease, and their use has resulted in clinical stabilization.[5]

Two new passive immunotherapy studies are now moving forward to determine whether therapy introduced earlier in the disease process will prove to be beneficial. The Dominantly Inherited Alzheimer Network will use 2 monoclonal antibodies (gantenerumab and solanezumab) and a β-secretase inhibitor, and the Alzheimer Preventive Initiative will study the monoclonal antibody crenezumab in patients with autosomal dominant presenilin 1 mutations. Patients who are carriers of the presenilin 1 mutation and who are asymptomatic will be selected, with therapy being initiated at least 15 years before the mean age at onset of disease in their families. This early-onset date was determined from cerebrospinal fluid studies of patients in the Dominantly Inherited Alzheimer Network who showed reductions of Aβ levels in cerebrospinal fluid samples as early as 25 years before the mean age at onset of dementia in their families.[6] It is hoped that patients beginning immunotherapy as early as 15 years before dementia begins in their families will avoid the secondary downstream independent pathologies of Alzheimer disease that can continue even if the level of Aβ is successfully reduced, leading to progressive disease with neuronal loss, gliosis, and dementia unresponsive to immunotherapy in the later stages of disease.

Antiamyloid immunotherapy can lead to inflammation with edema, demyelination, infarction, and microbleeds, as occurred in the patients in the Aβ42 peptide study [ie, the AN1792(QS-21)-201 trial].[3] We have used a DNA-based immunization approach in which the immunization agent is DNA encoding the Aβ1-42 trimer (3 copies of Aβ1-42), which produces a noninflammatory immune response.[7-9] DNA immunization using the gene-gun delivery into the skin results in a strongly polarized immune response that greatly differs from a peptide-generated immune response.[10,11] Aβ42 DNA vaccination via the gene gun generates a T_H2 cellular noninflammatory immune response. We have also shown that in vitro T-cell proliferation in response to Aβ1-42 peptide restimulation was absent in full-length DNA trimer (3 copies of the Aβ42 DNA) immunized mice, thus indicating the safety of this approach.[10-12]

Our report on the gene-gun–delivered DNA Aβ42 immunization, with a constitutive heterologous promoter that induced a good antibody response against Aβ42 in Balb/cJ mice, was the first to show that it is possible to use this method as an alternative to Aβ42 peptide immunization. With the same plasmid system, we further demonstrated that prophylactic DNA Aβ42 immunization in APPswe/PSEN 1δE9 transgenic mice reduced the brain Aβ42 plaque load by 42% and, furthermore, that DNA immunization with this human Aβ42 sequence also lead to good antibody production in a monkey that we had tested.[8,9] The humoral response to DNA Aβ1-42 immunization was substantially improved using a binary Gal4/UAS system in combination with a novel Aβ1-42 trimer construct. Aβ42 trimer DNA vaccination of wild-type mice resulted in greatly increased antibody levels showing a polarized T_H2 bias (IgG1 antibodies only) with no associated T-cell response, whereas a mixed T_H1/T_H2 (IgG1/IgG2a antibodies) immune response was found in Aβ42 peptide immunized mice, which were analyzed in parallel with the production of IFN-γ and IL-17 indicative of a potentially inflammatory cellular immune response.[10-12]

To further increase the level of antibody response with DNA immunization, we have developed a prime-boost protocol. The first immunizations initiate the immune response, and the following immunizations lead to an expansion of antigen-specific cells, with the selection of antibody-producing cells that have high antigen avidity and that lead to a boost of the specific response. We have analyzed 2 differ-

ent boost regimens: the peptide boost of a DNA-primed immune response and the DNA boost of a peptide-primed immune response. To our knowledge, this is the first time that a DNA boost was shown to have such a strong effect on the developing immune response. We were able to increase the antibody levels in plasma to 250 μg/mL with the peptide-primed/DNA boost method. Furthermore, peptide-primed/DNA boost immunization did not result in Aβ42-specific T-cell proliferation in these mice, nor did it lead to the production of inflammatory cytokines. Thus, the combination of peptide-primed/DNA boost immunization with our second-generation binary Gal4/Aβ42 trimer vaccine resulted in high levels of anti-Aβ42 antibodies that are polarized to a noninflammatory T_H2 cellular immune response with no T-cell proliferation and with no production of inflammatory cytokines. As therapy, it has the potential to delay or prevent Alzheimer disease if administered early in the disease process.[13]

If passive immunotherapy is shown to provide clinical benefit by reducing Aβ plaque levels, is nontoxic, and delays or prevents the development of Alzheimer disease, then interest in active DNA Aβ42 vaccination therapy will increase because DNA vaccination is less expensive, possibly longer lasting, and easier to administer to large populations at risk for Alzheimer disease than passive immunotherapy, which requires expensive humanized anti-Aβ42 monoclonal antibodies. Using cerebrospinal fluid biomarkers and brain amyloid imaging, it is now possible to identify asymptomatic persons at risk for Alzheimer disease, and these at-risk persons offer us the best opportunity to study how passive immunotherapy delays or prevents the disease from occurring. Thus, active vaccination with the DNA Aβ42 vaccine may become the method that will provide clinical benefit.[14,15]

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