MIMOVAX

SIXTH FRAMEWORK PROGRAMME

Project No.:
LSHB-CT-2006-037702

Project acronym:
MimoVax

Project title:

Alzheimer´s disease-treatment targeting truncated Aβ40/42 by active immunisation

Instrument: Specific Targeted Research Project

Thematic Priority: **LSH-2005-1.2.4-7**

Publishable final activity report

Period covered: from 01/10/2006 to 31/12/2011

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Duration: 51 months

Project coordinator name: Frank Mattner
Project coordinator organisation name: AFFIRIS

1.1. Section 1 - Project execution

Background:

Alzheimer's disease (AD) is the most common form of dementia in humans. At present, no effective treatment is available to stop the progressive neuro-degeneration and associated cognitive decline in AD patients.
AD is caused by the abnormal deposition of Beta–Amyloide (BA) plaques associated with extensive neuronal loss in the brain of patients. Main constituent of BA are protein fragments, so called peptides of 40-42 amino acids. These fragments derive from the Amyloid Precursor Protein (APP), which is expressed on various cell types in the nervous system. In humans the majority of amyloid plaque material is formed by truncated and modified Aβ40/42 derivatives. Aβ peptides are considered to be directly involved in the pathogenesis and progression of AD. Consequently, reduction of Aβ burden in the brain is predicted to slow down or halt disease progression and could also stop cognitive decline in AD patients.
Immunotherapeutic treatment targeting Aβ led to amyloid plaque reduction and had beneficial impact on disease progression in animal models. However, the first phase II clinical vaccination trial in AD patients using full length Aβ42 as antigen had to be discontinued due to severe neuroinflammatory side effects including brain infiltration by autoreactive T-cells.
Similar to full length Aβ, truncated and modified Aβ peptides also seem to be involved in the pathogenesis and progression of AD. Thus they are also a suitable point of attack for novel treatment strategies, but no relevant development programme has been started to date using immunotherapy to reduce the burden of full length and modified Aβ peptides.

Objectives

The MimoVax project is a specific targeted research project (STREP) for development and optimisation of a novel treatment to stop the progression of AD. The project aims at developing a vaccine against modified forms of BA. The immune system of AD patients will be activated to attack and remove BA and such fight the cause of the disease directly.
The novel AFFITOME technology presented in the MimoVax project has been developed to create antigens mimicking the structure of neo-epitopes[1] which do not contain sequences of the native Aβ peptide. A Mimotope-based AD vaccine would induce antibody responses **exclusively reacting with the pathological truncated and modified forms of the Aβ** molecules but not with parental structures like APP[2].
The first step in this project was the **identification and evaluation of novel mimotopes** mimicking epitopes present on different Aβ derivatives. Subsequently, these novel vaccine candidates were **tested for efficacy** of altering disease progression **in animal models of AD**. This evaluation includes analysis of **AD-pathology** in the brain as well as the testing of **memory and learning** in these animals by cognitive tests in vivo. The successful candidate is currently tested for **tolerability and safety** in a **phase 1 clinical trial in AD patients**.
In addition **new diagnostic methods** are being developed in the course of the MimoVax project in order to monitor treatment efficacy in vivo in animal models and to foster development of novel tracers for **diagnosis and monitoring of AD** in patients.

Thus the goal of the MimoVax project is the development of a **safe and efficacious Alzheimer vaccine** which is predicted to **avoid the development of immunological complications** due to autoreactive T-cells. The development of such **innovative AD vaccines targeting truncated and modified forms of Aβ** could therefore be a safe treatment regimen to efficiently fight AD in patients and could limit the severe personal and economic impact of AD on patients, their families and society.

[1] The epitope is the part of the antigen that is recognised by the immune system

[2] Amyloid precursor protein: a neuronal protein

Contractors involved and co-ordinator contact details

Specific Targeted Research Projects such as MimoVax are specifically designed to reinforce the scientific and technological knowledge and to foster the validation and exploitation of innovative solutions of European SMEs. The MimoVax project presented here is coordinated by an SME (AFFiRiS) with support by a dedicated management partner (Biolution). The project is conducted by 5 SMEs and 2 academic institutions from Spain, Germany and Austria. The project has been originally planned for three years and has been prolonged to another fifteen months by the EU to fulfil the clinical objectives leading to an overall project duration of 51 months. The project is receiving Euro 2.4 million in EU funding.

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Work performed and results achieved so far and expected results

The work performed within the MimoVax project in the three reporting periods is attributed to preclinical lead identification and evaluation by screening for novel antibodies and respective AFFITOPES as well as the in vitro and in vivo evaluation of identified vaccine candidates. In addition, the core work package of the MimoVax project is the subsequent first in human clinical trial of the successfully identified MimoVax vaccine candidate AD03 in a phase 1 clinical trial in early AD patients.
Within the initial reporting period the consortium could identify novel monoclonal antibodies and their respective AFFITOPES. Within the second reporting period the consortium could subsequently demonstrate that the novel MimoVax AD vaccine is able to reduce amyloid plaque load and alleviate the pathologic hallmarks in the brain of animal models for AD. In addition, immunisation with the selected candidate for later use in clinical trials, also led to improved spatial memory and learning in transgenic animals. Based on the successful POC experiments the consortium also initiated and demonstrated the safety of the MimoVax vaccine by extensive testing of the vaccine stability and by toxicologic analyses in animals. Thus the MimoVax preclinical development programme has been successfully completed in period two of the MimoVax project.
In reporting period three the consortium took the next step in product development and initiated the preparation of documents required for application for the clinical trial. Subsequently, the consortium was able to receive the permission to perform this important test in human AD patients in Austria and consequently started the active clinical trial for the evaluation of safety and tolerability of this novel vaccine in early AD patients in November 2010. By the end of the reporting period the first patients

have been successfully vaccinated within the study. Therefore, the trial is expected to be completed by the end of 2011.

The expected end result of the MimoVax project, which will be achieved outside of the project time frame, should thus be the demonstration of safety of such an innovative AD vaccine targeting Aβ in patients suffering from AD and could therefore open doors to the development of a safe treatment regimen in further clinical testing (clinical phase II).
In summary, vaccines based on AFFITOPE peptides mimicking neo-epitopes which are derived from the highly pathological and abundant Amyloid-β derivatives, provide an innovative, inexpensive and efficient way to treat this wide spread disease with its severe personal and economic impact on patients, their families and society. Successful implementation of the MimoVax vaccine would thus significantly reduce the high costs associated with this disease, which will soon exceed 4 billion € annually.

Intention for using and disseminating the knowledge (project logo and website)

Following these results, the Coordinator has encouraged all participants to actively disseminate their project aims and results to colleagues, stake-holders and the broader public. Public awareness of MimoVax has been addressed by a website highlighting principles and advances of the project. This web page www.mimovax.eu is hosted by the Coordinator (AFFiRiS) and P9 (biolution). The multimedia-based content (including new media such as video clips) is regularly checked, updated and includes key background information on the topic of AD, in keeping with all the rules, laws and ethical considerations required for a sensitive health topic. All content adheres as far as possible to the recommendations of the American Medical Association for health information websites (http://www.ama-assn.org/ama/pub/category/1905.html) and is presented in an accessible form and is of particular use to AD patients and their families. The entire website (including the video clips) is available in three languages (English, German, Spanish). Given there is already a considerable amount of information on AD in existence on the web, the MimoVax website has been hyperlinked to other reputable information-providing websites concerned with AD research and care as well as social platforms.

The Consortium also takes measures to provide MimoVax exposure to the public through conventional PR media work, which will complement the public access website, particularly for those that do not have internet access.

The scientific community has a vested interest in keeping up with MimoVax progress and results, to build on the body of knowledge that currently exists. This was and will be achieved through conventional scientific dissemination means, i.e. peer reviewed publications and presentations at international conferences.

A project logo has been created to identify the project and is used together with the contract number LSHB-CT-2006-037702 and the FP6 logo, on any printed and electronic issues for the communications to the European Commission and for any other official contact.

1.2. Section 2 – Dissemination and use

No original Mimovax research results have been published yet. Currently one peer reviewed publication featuring findings of MimoVax has been published by AFFiRiS (a review article):
Hum Vaccin. 2010 Nov 1;6(11):64-68;
AFFITOME® technology in neurodegenerative diseases: The doubling advantage; Schneeberger A, Mandler M, Mattner F, Schmidt W
All authors are employees of AFFiRiS AG; PDF is attached to the document.

Additionaly publications are currently in preparation by AFFiRiS and TUM and are scheduled for submission in 2011 (outside of the project duration). No additional publications are expected from partners UNIMAR, EE, JSW or piCHEM regarding the MimoVax project.

Published patentfamilies (AFFiRiS):

A951/2008: Austrian patent application and related applications

A952/2008: Austrian patent application and related applications

Human Vaccines 6:11, 1-5; November 2010; © 2010 Landes Bioscience

AFFITOME® technology in neurodegenerative diseases

The doubling advantage

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Key words: alzheimer, parkinson, neurodegenerative diseases, vaccine, immunotherapy, disease modification

Abbreviations: Aa, amino acid; Aβ, amyloid β; Ab, antibody; Ag, antigen; AD, alzheimer's disease; AE, adverse event; APP, amyloid precursor protein; BBB, blood brain barrier; CMI, cell mediated immunity; FDA, federal drug agency; LB, lewy body; ME, meningoencephalitis; MBP, myelin basic protein; NINCDS/ADRDA, national institute of neurological and communicative disorders and stroke and the alzheimer's disease and related disorders association criteria; TH1, T cells of the helper phenotype 1; POC, proof of concept; SUSAR, suspected unexpected serious adverse reaction; IFN, Interferon; Ig, Immunoglobulin

Neurodegenerative diseases are still an area of unmet medical need. This is in contrast to our increasing knowledge on their pathology (e.g., Alzheimer's-(AD), Parkinson's (PD) disease). They are driven by the cerebral accumulation and aggregation of specific proteins (e.g., β-amyloid and hyperphosphorylated tau in the case of AD) in defined brain regions and, as a consequence, death of neurons. Accordingly, removal of given protein aggregates is expected to modify the course of the respective neurodegenerative disease. This has been convincingly demonstrated in animal models of human diseases. However, not every technology that can be used and proves successful in animal models can be translated to the human situation. As highlighted by recent progress in the field of AD research, specific immunotherapy is a viable option in this regard. Given the fact that the aggregates are composed of self-proteins, immunotherapeutic approaches have to consider the issue of potential autoimmunity. This is especially true in case of vaccines. An innovative solution to this problem is offered by the so called AFFITOME® technology, which relies on the use of "doubles" of native molecules, functionally mimotopes or AFFITOPES® if identified by AFFiRiS, as the antigenic vaccine component.

Introduction

The concept of immunotherapy of neurodegenerative diseases was introduced by Dale Schenk and colleagues in 1999 when they used an Aβ vaccine to tackle AD.[1] Their vaccine was composed of Aβ itself and the immunological adjuvant QS21. Results obtained in mice transgenic for human APP, a model for AD, showed that its administration was capable of reducing cerebral Aβ load of vaccinated, transgenic mice, to ameliorate their Aβ-induced neuropathology and, as a result, to improve their cognitive function. The effects appeared to be mediated by vaccination-induced, Aβ-specific antibodies (Abs). Thus, the observed vaccine activity was consistent with a disease-modifying effect, that is, a novel therapeutic quality. Importantly, these results were consequently corroborated by several other investigators.[2-5]

As a next step, this vaccine (termed AN1792) was taken to the clinic. During the clinical development program assessing AN1792 its formulation was changed to include the stabilizer PS-80, known to shift the quality of the immune response induced towards a T helper 1 (TH1) phenotype (IFNγ high, CMI high, IgG1 high). This change in the vaccine's formulation was associated with the occurrence of cases of meningoencephalitis (ME). In total, 6% of vaccine recipients were found to develop and suffer from ME[6] which led to study halt. Research into its pathogenesis identified Aβ-specific, TH1-type CD4+ T lymphocytes as the most likely cause. This notion was not only supported by the results of the investigation but also highly plausible in light of studies governing other disease entities. In particular, research into the origin of multiple sclerosis identified myelin basic protein (MBP)-specific T cells as mediators of the inflammatory processes running in the brain of affected individuals and resulting in neurological deficits.[7] Of note, while the Aβ-specific TH1 type T cells were essential for ME to occur, they were not sufficient. This is based on the difference in the frequencies of vaccinees developing Aβ-specific IgG Abs and ME, which amounts to 22 versus 6 percent. As the Ab response critically depends on specific TH cells, the discrepancy might be explained by a cofactor. This would be in analogy to animal models, where, despite the activation of MBP-specific TH1 cells, encephalitis is only seen upon coadministration of pertussis toxin, an agent known to open the blood brain barrier (BBB).

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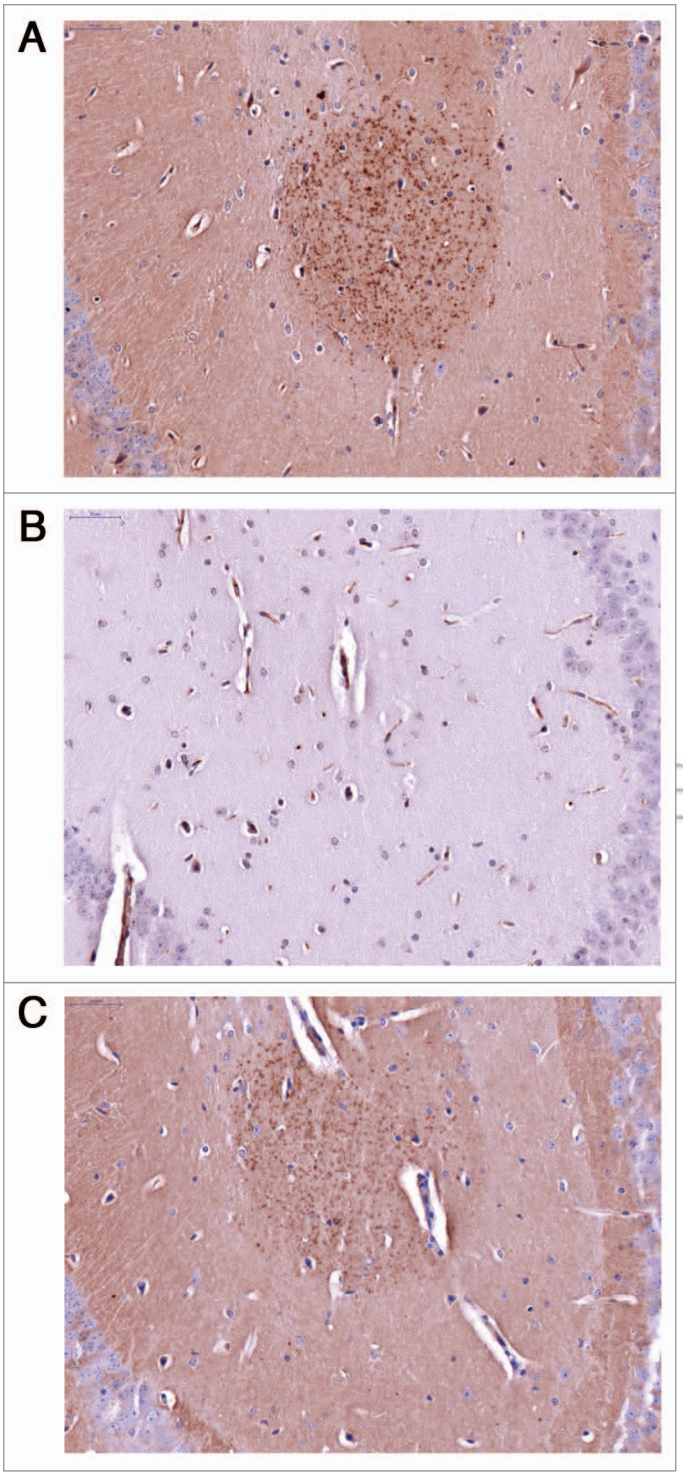

Figure 1. Antibodies elicited by an α-syn vaccine react with human α-syn in a specific manner. Brain slides of human α-syn transgenic mice were exposed to sera of an AFFITOPE-immunized wild-type mouse in the absence (A) or presence of competing peptide (B). Within the hippocampal area a distinct and specific staining pattern is observed with the AFFITOPE-induced serum. This is comparable to the staining pattern obtained with a commercially available human α-syn-specific Ab (C).

The above examples impressively illustrate a general principle. Cellular immune responses directed towards cerebral molecules potentially result in autoimmunological damage and destruction of target-expressing brain cells, especially in case of a TH1 type of response and irrespective of whether it is induced artificially or develops "endogenously".

Despite the fact that, on average, only 2 rather than the projected 6 vaccinations were applied, vaccination with AN1792 generated evidence for the proposed activity. There was evidence for an amelioration of the amyloid driven pathology underlying the disease as well as for a clinical benefit.[8-12] More recently, by relating neuropathological findings with the clinical course in a small cohort of AD patients having received AN1792, Nicoll and colleagues questioned whether isolated reduction of cerebral Aβ could translate into a clinical benefit.[13]

The other issue to be considered when developing vaccines for the treatment of neurodegenerative diseases is the induction of humoral autoimmunity. In recent years, there is an increasing body of literature demonstrating the existence of Ab-mediated autoimmune disorders of the central nervous system (CNS) (review in ref. 14). This field of research originates out of the effort of elucidating the nature of paraneoplastic syndromes of the CNS. Defined neurological entities in patients with certain types of cancer could be attributed to the presence of (high titer) Abs specific for molecules expressed in the respective brain areas. While the field is still evolving, as up to date only a limited set of such Abs and the corresponding clinical syndromes have been identified, it is becoming clear, that the respective syndromes can occur in the absence of cancer but in an idiopathic, autoantibody-mediated manner. Of importance in the context of this discussion, the molecular targets of these Abs include both intracellular- and cell-surface/synaptic proteins. Examples of the former group are cancer-related onconeuronal Abs to SOX and Zic families of proteins as well as Abs that are associated with CNS syndromes in a nonparaneoplastic fashion such as those directed against glutamic acid decarboxylase (GAD). The latter encompass synaptic proteins like voltage gated synaptic channel (VGKC) related protein, the NMDA-, AMPA-, $GABA_B$-and glycine receptor. In conclusion, these syndromes are rare but exist and are the result of a biological principle that needs to be taken into consideration when developing immunotherapies targeting CNS diseases.

Implications for the Design of Vaccines Targeting Self-proteins

Autoimmunity is the major side effect of vaccines addressing self-proteins. This is not only demonstrated by the AN1792 example and the paraneoplastic/autoimmune CNS syndromes but also by the experience with human cancer vaccines.[15] Other potential side effects include, but are not limited to, toxicities associated with specific vaccine components used.

Obviously, prevention of self-protein-specific cellular autoimmunity requires the activity of a given vaccine to be mediated by the Ab component of the immune response induced. Thus, a potentially successful vaccine has to meet a series of criteria.

First, its antigenic component must not activate T cells but serve as a B cell antigen. Therefore, it has to obey the biological principles that govern antigen (Ag) recognition by T lymphocytes, which include the following. The Ag needs to be presented by antigen-presenting cells on their MHC molecules. Binding to them requires the peptide Ags to (i) have a certain length (8–9 aa in case of MHC class I) and (ii) to exhibit defined aa residues in so called anchor positions.[16] In essence, the antigenic epitope has to be short enough to prevent T cell activation but long enough to serve as an antibody epitope. However, the generation of an IgG type Ab response is dependent on T helper (TH) cells. This is thus the second task; the vaccine must contain (an) epitope(s) capable of activating TH cells. These must not be related to the Ag of interest as this could result in cellular autoimmunity. Ideally, they have to be strong TH epitopes and known to be incapable of activating Abs that crossreact with molecules/structures found in humans. Finally, to fulfil their task, the protein which provides T help needs to be physically linked to the B cell epitope driving the Ab response.

Tailoring the Ab response of a vaccine targeting a self protein is the true challenge. It needs to tackle the desired structure without attacking related ones. This requires fulfilling two criteria at a time. One, the Abs raised need to bind the defined target, which is crucial for the vaccine's intended activity. Two, they must however not bind to related or homologous structures, which is an essential safety feature.

The AFFiRiS solution to this is based on the proprietary AFFITOME® technology.[17] It aims at targeting a neo-epitope, a structure specific for the form of the self protein that drives the pathology of a given disease. This is accomplished by using AFFITIOPES, (short) peptides mimicking (parts of) the native sequence/structure of the neo-epitope. AFFITOPES are identified with a so called "selecting antibody". It is this Ab's features that are going to be translated to the humoral immune response elicited by the given AFFITOPE. To this end, the pool of all possible peptides of a predefined length is screened for those binding to the selecting Ab. The binding ones will have imprinted the specific features of the selecting Ab. Upon immunization with theses AFFITOPE candidates, the imprinted features are passed onto the resulting Abs. Candidates become true AFFITOPES if they are immunogenic, transfer the features of the selecting Ab and are successful in POC studies.

Thus, AFFITOPES differ from the native amino acid sequence of the target they mimic. As selected candidates do also not exhibit sequence identity with other human proteins, they are "foreign" to the human immune system. This endows the technology with a conceptual advantage. Immune responses are more readily elicited towards foreign- as compared to self-proteins as the organism protects "self" from being destructed by an immune attack via "central" and "peripheral" tolerance mechanisms.[18] This allows AFFiRiS to use aluminium hydroxide, the agent first approved as immunological adjuvant for human use and, thus, exhibiting an excellent safety profile.

Alzheimer's: Targeting the unmodified N-terminus of Aβ. This approach is based on the notion that cerebral accumulation of Aβ species is central to AD pathogenesis. AFFiRiS has chosen the free N-terminus of Aβ as suitable neo-epitope based on the following facts. The free N-terminus is readily accessible for Abs on all identified toxic Aβ species, in particular Aβ oligomers. However, this target structure does not exist within APP, the Aβ precursor molecule. Technically, this is accomplished by using an Ab that specifically recognizes the free N-terminus of Aβ as the AFFITOPE selecting Ab. This resulted in the identification of a series of "N-terminus AD AFFITOPES". The set identified and patented most likely encompasses all peptides sharing this feature. Most of them could be synthesized and were found to be immunogenic in mice. Two of them (AD01 and AD02) were developed through POC studies. Both exhibited disease modifying potential in the sense that their administration to APP transgenic mice reduced their cerebral Aβ load, improved their AD-like neuropathology and cognitive function (M. Mandler, manuscript in preparation). Toxicity studies in rats and guinea pigs (their Aβ sequence is identical to the human one) revealed an excellent safety profile for both, AD01 and AD02.

The strategy behind clinical testing of AFFITOPES AD01 & AD02. According to current thinking, agents with Aβ-lowering activity possess disease-modifying potential. Thus, the general strategy has to consider relevant clinical parameters and biomarkers over a sufficient time period using appropriate statistical tools.

As a first step, safety and toxicity of AFFITOPE AD01 and AD02 had to be addressed. The rationale behind the respective trials (AFFiRiS001 examined AFFITOPE AD01, AFFiRiS002 AFFITOPE AD02) was based on the following facts. AFFITOPES AD01 and AD02 were designed to preclude cellular as well as humoral autoimmunity and proved to be safe in animal experiments. True toxicity testing of peptide-based vaccines has to consider immunotoxicity which requires (i) vaccine administration in a way that allows for the elicitation of an immune response and (ii) application of the vaccine to patients, as only they are exhibiting the targeted structure, i.e., Aβ, in sufficient amounts.

Based on the thinking outlined above, both studies were designed as two-arm, parallel group studies in which participants received the AFFITOPE/carrier conjugate either on itself or adjuvanted with aluminium. Both studies were directed to patients with mild to moderate AD (MMSE 16–26). Having passed the screening visit, participants received 4 vaccinations at 4-week intervals. The final visit was performed 8 weeks after the last immunization. Primary objective of either trial was the evaluation of the vaccines' safety and tolerability. Secondary objective was the exploratory assessment of the vaccines' immunological and clinical activity.

Safety measures built into the trial included the availability of a caregiver, stepwise enrolment, emergency plans, frequent patient contacts and regular review of the data by an external and independent data safety monitoring board.

The phase I results supported the safety concept realized in the first AD AFFITOPE vaccines. For each of the two studies, 24 patients with mild to moderate AD were recruited. Both study groups were divided into two subgroups of 12 patients who received the vaccine with or without adjuvant. All 48 patients were able to complete the study without any problems, with the

data demonstrating acceptable tolerability of vaccine candidates AD01 and AD02. All AEs were of mild to moderate degree, two thirds were found to be local reactions. None of the 48 participants in the studies showed any indications of ME or other autoimmune reactions. Thus, both vaccine candidates met the primary phase I endpoints.

Clinical development of AFFITOPE AD01&AD02—the next steps. Based on the first interim analysis of the secondary endpoints at the 6 month time point, AFFiRiS decided to focus its Alzheimer's vaccine program on one candidate. The vaccine candidate AD02 is planned to enter a multicenter, international phase II study involving >400 early AD patients. Furthermore, the AD02 patients from the completed phase I study were offered a booster vaccination within a separate clinical trial.

Targeting the truncated, pyroglutamate-modified N-terminus of Aβ. Aβ comes in several forms, besides the classical Aβ1-40- and Aβ1-42 molecules, there exist N-terminally truncated and pyroglutamate-modified Aβ versions. The latter receive increasing attention due to the facts (i) that they are the major component of human amyloid plaques (60–80% of the amyloid) (ii) that they are more amyloidogenic than their non-modified counterparts (longer half-life, their high hydrophobicity substantially increases the aggregation rate),[19-21] and (iii) that they are found early on during the disease process.[22-24] Moreover, their occurrence appears to be specifically linked to the disease. Together, these features render the N-terminally truncated and modified Aβ versions excellent neo-epitopes. Accordingly, AFFiRiS selected corresponding AFFITOPES and applied them to the routine preclinical screening program. One of them, called AD03 (MimoVax), which fulfilled all predefined criteria is planned to enter phase I clinical testing in 2010. This highlights the potency of the AFFITOME® technology which allows AFFiRiS to translate new research data into a novel vaccine candidate within a narrow time frame.

Opening new indications: Parkinson's disease. PD is a neurodegenerative disorder clinically characterized by tremor, rigidity, bradykinesia and cognitive impairment in a subset of patients. In western countries, it affects up to 2% of the population above 60 years of age. Current treatment options include medications such as L-Dopa and dopamine agonists, anticholinergic drugs as well as inhibitors of monoamine oxidase or a surgical intervention termed deep brain stimulation. However, all currently available treatment options are of symptomatic benefit only, a drug with disease modifying properties is still lacking.

Lewy bodies (LBs) and Lewy neurites, found in various cortical and subcortical areas of the brain, are neuropathological hallmarks of the disease. They are associated with a progressive loss of neurons, especially in movement control areas including the substantia nigra. The pathology appears to be most abundant in regions containing high numbers of dopaminergic neurons or neuronal projections. The major constituent of LBs is the synaptic protein α-synuclein (α-syn), a naturally unfolded protein of 140 aa. Although the exact mechanism(s) of α-syn toxicity remain(s) to be determined, recent results suggest α-syn oligomers to play a critical role in the processes that lead to neurodegeneration.[25] Hence, reduction of α-syn deposition and oligomerization is expected to exhibit a disease-modifying effect. Experimental evidence supporting this notion was provided by recent work of Masliah and colleagues. They treated mice overexpressing the human α-syn gene with an α-syn-based vaccine. Mice developing high affinity Abs were found to have reduced amounts of aggregated α-syn both within their cell bodies and at the synapses. Importantly, the reduction of α-syn oligomers was found to correlate inversely with the extent of neurodegeneration. Furthermore, elicited Abs appeared to react with abnormal forms of α-syn located within the neuronal membrane and promoted their degradation, probably via lysosomal pathways.[26,27] These investigators obtained similar effects when using passive immunotherapy with an α-syn-specific Ab.

While the above studies underline the value of immunotherapy, they do not allow for a full safety evaluation. The biggest threat of α-syn immunotherapy concerns crossreaction with other members of the synuclein family. The family consists of three members, α-syn, β-syn and γ-syn, which do not only share extensive sequence homology but also a similar expression pattern. They are thought to act as chaperones thereby regulating vesicle transport and synaptic plasticity. Of note, α-syn knock-out mice are viable and show no overt neuronal phenotype indicating that α-syn function can be replaced by other synucleins. β-syn shows the highest overlap of expression with α-syn. It appears to counteract most of the toxic effects of α-syn. Its neuroprotective properties are in part mediated by AKT signalling. In addition, β-syn has been shown to prevent aggregation and oxidation of α-syn.

Obviously, immunotherapeutic approaches need to respect these relations, in particular a vaccine targeting α-syn must not attack β-syn. This task can be mastered by the AFFITOME® technology (**Fig. 1**). Consequently, the AFFiRiS PD drug development program focuses on AFFITOPES eliciting Abs recognizing α-syn while sparing β-syn. It delivered a series of candidates. One AFFITOPE, termed PD01, was found to exhibit disease-modifying activity in POC studies in animal models of the disease. This stresses the importance and the general applicability of the neo-epitope concept inherent to the technology.

Conclusion

Misfolded proteins are critical for the pathology of various neurodegenerative entities. This knowledge opens new and potentially disease-modifying treatment options. Key to the safety of immunological treatment approaches is the avoidance of crossreaction that is the effector mechanisms induced have to react exclusively with the pathologic forms of the proteins involved. The AFFITOME® technology, which relies on "doubles", so called AFFITOPES®, rather than on original protein sequences, is capable of delivering vaccine candidates fulfilling this essential criterion. The most promising ones, of those developed for AD and PD, are currently being tested in clinical trials.

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