Prof. Beka Solomon's Research Interests

My scientific interest is focused on investigation of biorecognition in general and on the nature of the interactions of antibodies and their corresponding antigens in particular. In my research work I am involved in the elucidation of the mode of action of monoclonal antibodies (mAbs) on the structure and function of their corresponding antigens in in vitro and in vivo environments. Antibodies were found to act as reporting probes for the detection of conformational changes induced in various protein antigens by environmental factors, as well as playing active roles in inducing changes and rearrangements in the antigen molecule. The following topics are being investigated:

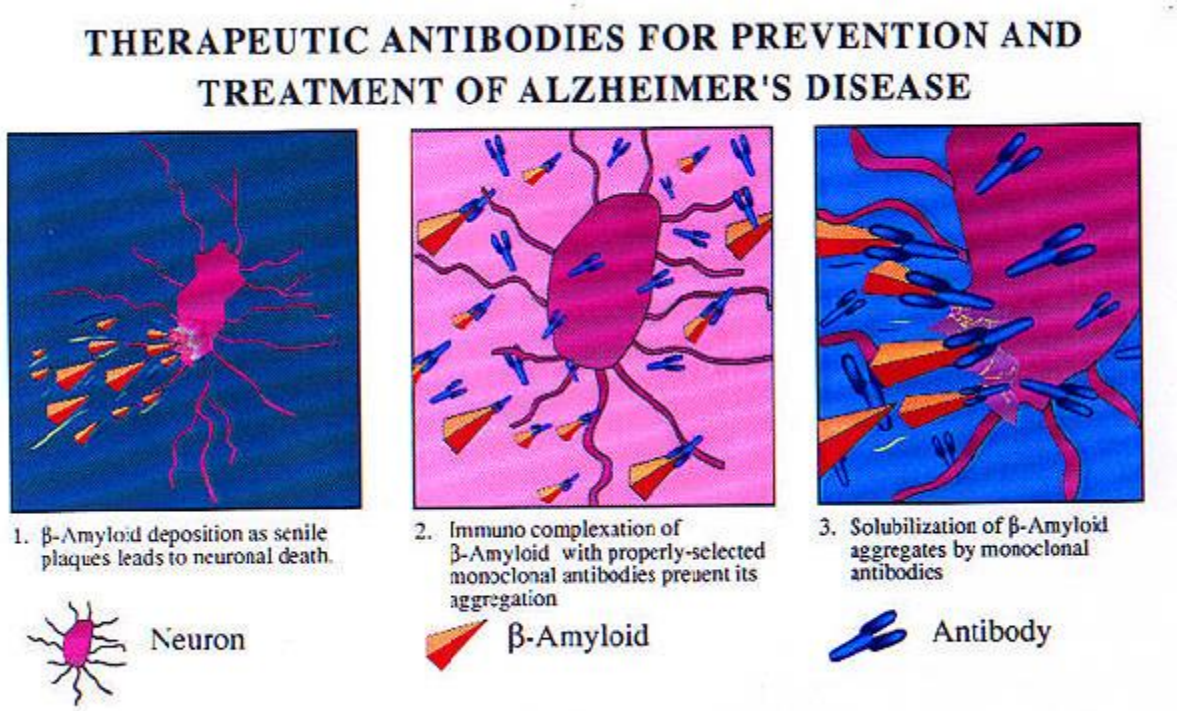
a. detection of conformation changes in the protein antigen by their mAbs.
b. stabilization of the antigen structure by mAbs.
c. Chaperone characteristics of mAbs leading to the refolding of the respective antigen.
d. Prevention of aggregation and solubilization of previously formed protein aggregates.

In addition, I am involved in the design and production of novel antibodies with the desired combination of binding specificity and biological properties.

My group prepared monoclonal antibodies towards several proteins, such as carboxypeptidase A, Che Y, hexosaminidase A, horseradish peroxidase, p53 protein, calmodulin and against aluminium ions, and recently against Alzheimer¹s ß-amyloid peptide and prion peptides 106-126, in order to study conformational changes occurring in these proteins when exposed to various working conditions. The monoclonal antibodies were used as probes to assay conformation changes of interacting epitopes in these target proteins, induced by temperature, pH, proteolytic cleavage or metal interactions. The availability of the mAbs enabled me and my collaborators to develop new immobilization techniques yielding fully active immobilized Ab-carrier conjugates, which led to novel techniques for enzyme immobilization and stabilization. The immobilized mAbs permitted the development of novel immunoaffinity techniques and original immunoassay techniques. A microalbuminuria assay based on the above techniques was especially appreciated and received an Award for Excellence in a Meeting of Clinical Biochemistry held in Japan in 1991.

Protein aggregation plays an important role in various human diseases, such as Down¹s syndrome, Alzheimer¹s disease, diabetes and/or cataracts, and in many other so-called amyloidosis disorders. In order to reduce or eliminate the extent of pathological protein depositions my group focused on the development of potent and selective inhibitors of aggregate formation. We commenced our studies by using carboxypeptidase A (CPA) as a model system and a large panel of mAbs prepared against it. We found that appropriate mAbs interact at strategic sites where protein unfolding is initiated, thereby stabilizing the protein and suppressing further aggregation. These studies were extended on the suppression of Alzheimer ß-amyloid peptide by immunocomplexation with highly specific monoclonal antibodies raised against the peptide. The amyloid ß-peptide is a main component of the senile plaques amyloid found in the brain tissues of Alzheimer¹s diseased patients. Amyloid stability, even under harsh conditions, was one of the unsurmountable problems in the initial characterization of its constituents.

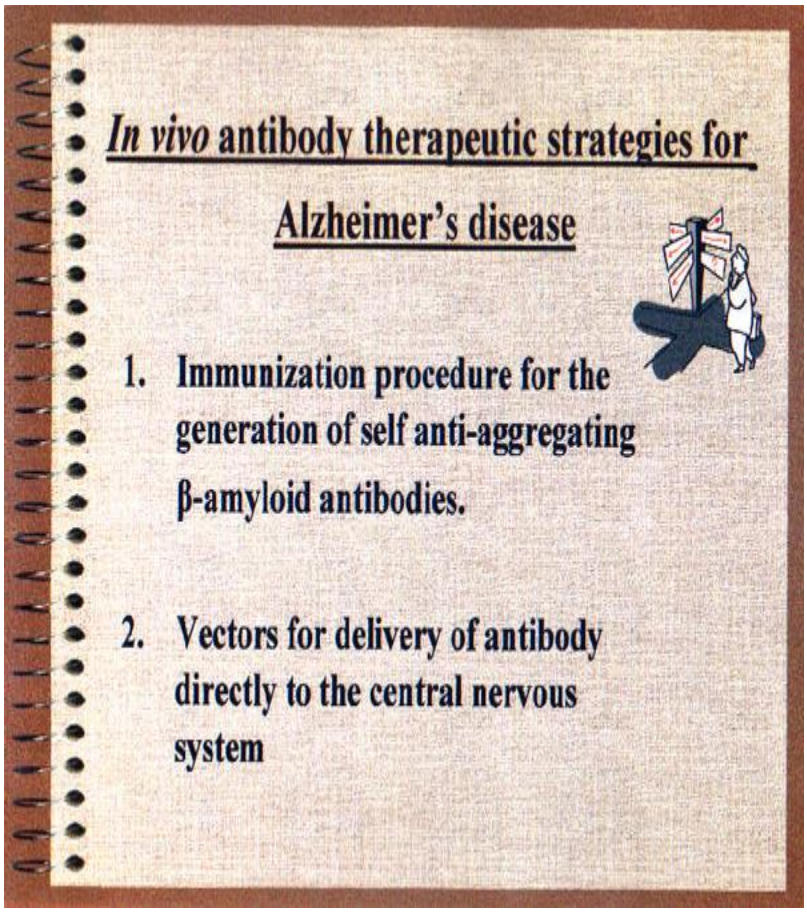
We recently found that selected mAbs against ß-amyloid peptide can solubilize preformed ß-amyloid aggregated filaments by reversal of the ß-sheet insoluble conformation into a corresponding soluble random coil. Such Œchaperone-like¹ properties of mAbs were found to be related to so-called anti-aggregating epitopes on each antigen. We found that mAbs directed to these regions are able to interfer with the dynamics and rearrangement of whole molecules.



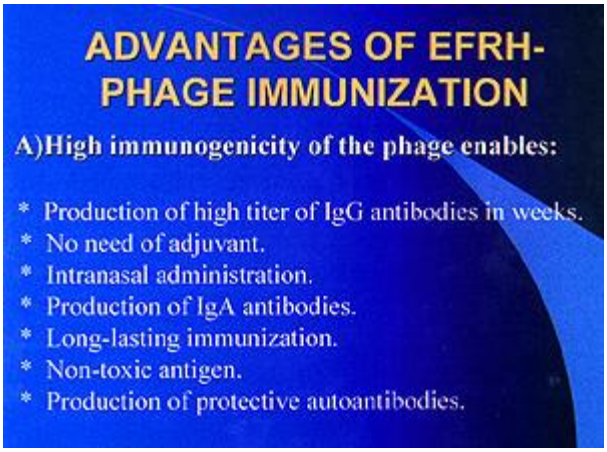
We propose the use of such small epitopes, which exhibit high immunogenicity and belong to the immunodominant region of the antigen, to elicit antibodies with anti-aggregating properties. Encouraging data on the prevention and suppression of aggregation of ß-amyloid peptide and especially on the solubilization of preformed amyloid fibrils in conjunction with recent advances in antibody engineering raise the possibility that the anti-aggregating antibodies could prove feasible for the treatment of Alzheimer¹s disease. New research strategies are developed in our lab for preparation and brain targeting of anti-ß-amyloid antibodies towards prevention and/or treatment of Alzheimer¹s disease. The two main approaches described below for the delivery of such antibodies to the brain are dependent on the permeability of the blood-brain-barrier (BBB) to antibody molecules.

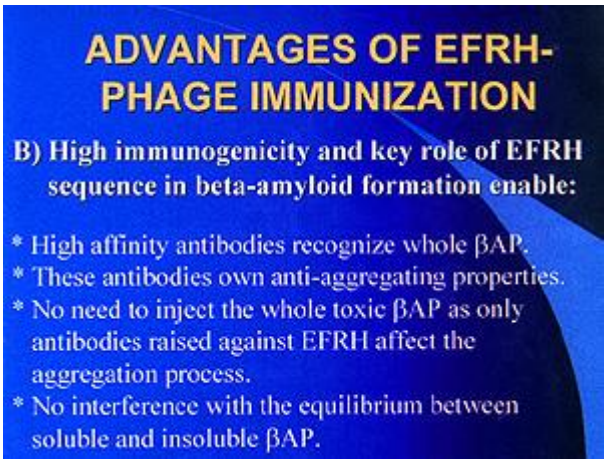
1. Brain delivery of anti-ß-amyloid antibodies obtained by EFRH-phage immunization

Filamentous phages, which are excellent vehicles for the expression and presentation of foreign peptides, were found to induce a strong immunological response to all the phage proteins after in vivo administration. The efficacy of this procedure is directly dependent on the immunogenicity of the peptide displayed. Due to the high antigenicity of the EFRH peptide, which belongs to the immunodominant region of ßAP, high affinity (IgG) antibodies were obtained after a very short immunization time (one week) compared to protein carrier immunization (one year). Immunogenicity of the phage enables administration via intranasal route without any use of adjuvant. Intranasal administration appears to be a more effective route of



mice immunization in terms of timing and reproducibility of response.





2. Intranasal delivery of engineered antibodies

In preliminary experiments we found that vectors carrying single-chain Fv (ScFv) antibodies may be delivered directly to the CNS by intranasal administration via olfactory receptor neurons. Their axons traverse the cribriform plate and project to the first synapse of the olfactory pathway in the olfactory bulb of the brain. They form a highway by which viruses or other transported substances may gain access to the CNS. To our knowledge, this is the first attempt to deliver antibodies to the CNS.

ß-Amyloid is only one example of at least 15 different polypeptides known to cause in vivo different forms of pathological amyloidosis via their deposition in particular organs and tissues as insoluble protein fibrils. The prevention of aggregation and solubilizations of other peptides that form amyloid in tissues, such as amylin, serum amyloid A, prion protein, are being investigated at present. Since properly selected mAbs can be prepared against virtually any antigen, their immunocomplexation may provide a general and convenient method for the stabilization of the soluble physiological conformation of the above proteins without affecting their biological properties.

Our recent findings on the chaperone-like activity of mAbs in stabilization, refolding and solubilization of the already aggregated antigens, as well as the introduction of antibody engineering techniques, open up new possibilities for the potential use of mAbs in immunotherapy of Alzheimer¹s disease. The development of therapeutic antibodies for use as a future vaccine in Alzheimer¹s disease and/or other amyloidogenic diseases, such as prion disease, are the subject of the Master of Science and Ph.D. theses of students in my lab.

We have started to apply our novel procedures for the production of anti-aggregating antibodies by active and passive immunization, as well as by direct delivery to the brain avoiding the blood brain barrier in the Alzheimer¹s disease model of transgenic mice.

We recently commenced another project based on a search for biological markers in body fluids from Alzheimer¹s disease and control patients to enable evaluation of the pathophysiological hypothesis of this disease, taking into consideration the fact that a defect in the transduction system might play a central role in the pathogenesis of Alzheimer¹s disease. An early detection test for Alzheimer¹s disease is not yet available and there is an urgent need to develop technologies for its production, as wel as the accurate detection of AD. Our laboratories will study the metabolism of ßAPP in the human circulation. Such studies are aimed at elucidating environmental or nutritional factors which may alter the levels of ßAPP production and determine whether abnormalities of ßAPP metabolism evident in the brain in AD are reflected in the blood.