Introduction
Alzheimer disease (AD) is a progressive neurodegenerative disorder, characterized by the formation of amyloid peptides (Aβ) and their deposition in the brain as senile plaques (1). The amyloid peptides Aβ40 and Aβ42, and especially their oligomeric aggregates, are believed to play a crucial and early role in the pathogenesis of AD by causing neurotoxicity (2), development of neurofibrillary tangles (3), impairment of long-term potentiation (LTP) (4), and age-related cognitive deficits (5).

Strategies to treat AD are aimed to prevent the formation of Aβ peptides and their deposition in the brain. Therefore, β- and γ-secretases that generate Aβ peptides by sequential cleavage of the amyloid precursor protein (APP) are obvious and main targets for the development of specific inhibitors (6).

Alternatively, increasing α-secretase activity in the brain provides an attractive strategy, since proteolysis of APP within the Aβ sequence precludes the formation of Aβ peptides (7, 8, 9). In addition, α-secretase cleavage of APP releases its N-terminal extracellular domain of about 110 kDa referred to as APPαx, which has neurotrophic and neuroprotective properties (10, 11) and enhances LTP (12). In behavioral paradigms, APPαx was demonstrated to enhance memory in normal and amnesic mice (13). In this respect it is interesting to note that a reduction of APPαx is evident in the cerebrospinal fluid of AD patients (14, 15).

Studies with neuronal cell lines and hippocampal neurons have shown exogenous APPαx, after binding to a yet unknown receptor, to activate several signal pathways which regulate calcium homeostasis. Furthermore, it was demonstrated that APPαx neutralizes pro-apoptotic factors, has neuroprotective properties and promotes neurite outgrowth, but the cellular target proteins of the APPαx action are present at unknown.

Moreover, in double-transgenic ADAM10 x APP mice the amount of the C-terminal APP fragment generated by α-secretase is increased (19). This membrane-bound fragment is a substrate for subsequent γ-secretase cleavage that releases APPαx and the APP intracellular domain (AICD) which might serve as a potential transcriptional modulator. APPαx itself modulates the cytosolic calcium concentration, neutralizes pro-apoptotic factors, has neuroprotective properties and promotes neurite outgrowth, but the cellular target proteins of the APPαx action are present at unknown.

By overexpression of ADAM10 in the APPγ/717 mice background we observed significant improvements in cognitive abilities (Fig. 2). In comparison to APPγ/717 mice, long-term potentiation, learning and memory were improved in double-transgenic ADAM10 x APPγ/717 animals (19). In contrast, a neuron-specific knock-out of the presenilin 1 γ-secretase prevented amyloid plaque formation, but did not improve cognitive deficits of APPγ/717 mice (20). Therefore, the beneficial effect of increased ADAM10 activity, including cognitive improvements, must be attributed to the combined effects of decreased levels of toxic Aβ peptides and increased amounts of neuroprotective APPαx. In this respect it is important to note that exogenously applied APPαx has been shown to enhance LTP in hippocampal slices (12), and in behavioral paradigms, intracerebroventricularly administered APPαx enhanced memory in normal and amnesic mice (13).
Disease-oriented Genome Networks

Fig 2: Acquisition of spatial learning in the Morris water-maze hidden-platform task. Learning deficit in APP(V717I) transgenic mice was ameliorated in double-transgenic ADAM10-mo*APP(V717I) mice. Lines represent mean ± SEM for 9–10 mice per group. *P < 0.05, **P < 0.01, APP(V717I) vs. control; §P < 0.05 APP(V717I) vs. ADAM10-mo x APP(V717I) by Student’s t test.

Outlook
The project is aimed to identify genes mediating neuroprotective and neurotrophic functions of the α-secretase ADAM10. Activation of these genes thus might prevent Alzheimer disease and other neurodegenerative disorders. Differentially expressed genes will be identified by applying Affymetrix mouse genome microarray analysis using brain samples of single-transgenic ADAM10, and dominant-negative ADAM10 overexpressing FVB/N mice. In these studies, non-transgenic FVB/N mice represent the control group. A direct target for APPα secretase action might be a putative APPα receptor which may be upregulated in brains with very low amounts of APPα, e.g. in dominant-negative ADAM10-expressing mice. Other direct target genes for the action of ADAM10 may comprise putative ADAM10 substrates such as the neuronal adhesion molecule L1. These might be upregulated during increased processing by ADAM10. Indirect target genes include those of cellular signaling molecules.

After identification of differentially expressed genes, the outcome of the microarray analysis shall be validated by quantitative real time RT-PCR and Western Blots on mouse brain samples. Finally, putative ADAM10 target genes shall be knocked-down by small interfering RNAs in established cultured cell lines, and analyzed with regard to anti-apoptotic and anti-neurotropic properties. In addition, the function of putative ADAM10 target genes will be analyzed after overexpression. New putative ADAM10 substrates can be validated by the use of specific ADAM10 inhibitors and by in vitro cleavage assays.

As the γ-secretase released C-terminus of APP (AICD) is a potential transcription factor, double-transgenic mice expressing either ADAM10 x APP or dominant-negative ADAM10 x APP will be analyzed for differentially expressed genes as described above. In ADAM10 x APP mice the amount of the C-terminal fragment of APP generated by the α-secretase is two-fold increased. Therefore, more AICD is likely to be released to modulated gene expression.