Summary

One of the most important post-translational modification of proteins is the non-enzymatic attachment of reducing sugars. Subsequent oxidations, dehydrations and rearrangements produce a heterogenous group of heterocyclic, coloured and fluorescent compounds termed "advanced glycation endproducts" (AGEs). In the course of their formation, free radicals and other reactive intermediates are created. AGE-modified proteins are resistant to proteases, their formation is irreversible. They accumulate on long-lived proteins with slow turn-over, e.g. on collagen or eye lens cristallin and on pathological protein deposits, e.g. in Alzheimer´s diease (AD). Accumulation of AGEs also occurs in the diabetic kidney and in the pathogenesis of dialysis related amyloidosis (DRA) and is discussed to be of importance in the etiology of AD.
The pathology of Alzheimer´s disease involves accumulation of intra and extracellular protein aggregates like senile plaques and tangles. Further hallmarks are a reduction of glucose metabolism in the affected brain areas, together with signs for an acute phase response and for oxidative stress.

In vitro experiments showed, that AGEs accelerate the crosslinking of βA4, the major component of senile plaques in AD. Since glycation of the peptide monomer is the first step of this reaction, this points to a participation of glycation in plaque-formation in AD. The formation of covalently crosslinked hight-weight βA4 oligomers is further accelerated by micromolar amounts of copper and iron ions. Formation of these AGE-crosslinks can be inhibited by agents which are able of capping amino-groups, by redox-inaktive metall chelators and by antioxidants, suggesting that these drugs may have the potential to slow down the formation of insoluble protein deposits *in vivo*.

AGEs have a direct toxic effect on BHK 21 fibroblasts and human SH-SY5Y neuroblastoma cells, leading to inhibition of mitochondrial respiration, membrane leakage and subsequent cell death in a dose dependant manner. AGE-modification renders proteins cytotoxic, the toxicity of a protein increases with the total AGE-content and depends from the modified protein. The LD_{50} of a model-AGE (between 110µM and 160µM for BSA-AGE) can be correlated with the *in vitro* radical production by the modified protein.
On the level of signal transduction, AGEs induce the activation of the nuclear transcription factor κB as a stress response in neuroblastoma cells. AGEs enhance the formation of intracellular lipid-peroxidation products as markers for oxidative stress.
AGE-toxicity is mediated by ROS and oxidative stress since various antioxidants were able to attenuate AGE-induced cell death. The receptor for AGEs (RAGE) appeared to be involved in

AGE-toxicity as well, because blocking of RAGE with neutralizing antibodies increased the percentage of vital cells after AGE- treatment.
The AGE-induced cell death shows signs of an initial apoptotic, final necrotic pathway. Though the percentage of annexin positive, apoptotic cells is slighly increased in cell cultures treated with sublethal amounts of AGEs and cytochrome c is released in the cytoplasma as a proapoptotic signal, no activation of caspase-3 was found.

Methylglyoxal, an AGE-precursor, is directly or indirectly cytotoxic, too. In contrast to AGE-modified proteins, most of the tested antioxidants were unable to inhibit or attenuate cell death suggesting other signalling pathways and other cellular targets of the substance. Scavenging of the reactive carbonyl groups of methylglyoxal completely inhibited toxicity.

AGE-induced stress leads to an imbalance in the cellular redox-status, recognizable by a shift in the GSH / GSSG ratio and a depletion of intracellular glutathion. This is accompanied by changes in the cells glucose metabolism and impairment of energy production.
During the first 6 hours of AGE-stress glucose uptake of the cells was increased up to 3-fold. After this time point almost no further uptake of glucose from the medium was detectable but hight amounts of lactate were released. The ATP content of the cells was reduced up to 50%. Administration of antioxidants together with or before administration of AGEs normalized ATP-levels as well as glucose uptake and lactate release, with (R+) α-lipoic acid being the most effective substance.

Interaction of AGEs with cells has been shown to cause oxidative stress, not only by receptor mediated pathways but also by production of free radicals by chemical oxidation and degradation of AGEs. This causes metabolic dysfunctions, energy depletion and impairment of the cells antioxidative defense. In the AD brain, this may lead to neurodegeneration and cell death, suggesting a role of AGEs in the pathogenesis of this disease.
The results encourage the use of membrane permeable antioxidants in novel treatment strategies of AD. AGE-inhibitors may represent an interesting approach to slow down plaque formation.