Recent Progress in Advanced Glycation and Diabetic Vascular Disease: Role of Advanced Glycation End Product Receptors

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Advanced glycosylation end products (AGEs) form principally from the rearrangement of early glycation products, i.e., Amadori products, which produce a class of stable moieties that possess distinctive chemical cross-linking and biological properties. It has been generally believed that proteins with half-lives of longer than a few weeks are most susceptible to advanced glycosylation and that the highest levels of AGEs occur on proteins that comprise the long-lived structural components of connective tissue matrix and basement membrane. ***Diabetes*** **45 (Suppl. 3):S65–S66, 1996**

Of interest has been the recent observation that advanced glycosylation end products (AGEs) can occur on short-lived proteins, on lipid constituents, and on nucleic acids. This is most apparent under conditions of high AGE accumulation, such as diabetes and renal failure, and results from the fact that AGE formation proceeds through a succession of reactive intermediates that can bind indiscriminately to amino groups present on diverse bystander proteins. Reactive intermediates can also be released from naturally degraded tissue AGE, only to bind again on other substances if not cleared via the kidney, forming second-generation AGEs.

Cell surface receptors that are specific for the recognition and degradation of AGE-modified proteins also have been identified on circulating monocytes, lymphocytes, endothelial and renal mesangial cells, and other cellular systems to partake in both normal tissue remodeling and tissue damage.

In this synoptic review, we highlight selected recent studies indicating that AGEs may alter the structural and functional properties of proteins and may contribute to many of the pathophysiological changes that accompany longstanding diabetes.

The late rearrangements of the covalent nonenzymatic modification of proteins by glucose, called AGEs, have been shown to accumulate in diabetic and aging tissues (1). In serum, AGE levels correlate inversely with renal function and are found to be markedly elevated in diabetic patients with end-stage renal disease (ESRD) (2). Numerous studies have indicated that reactive AGEs can directly alter the physical and structural properties of the extracellular matrix, for instance, by inducing collagen cross-linking, basement membrane thickening, and covalent trapping of plasma proteins such as LDL and IgG (1,2). In addition, AGEs are known to elicit a wide range of cell-mediated responses, thereby indirectly mediating phenotypic changes leading to vascular dysfunction, matrix expansion, and atherosclerosis and glomerulosclerosis (1). These cellular responses are thought to be largely induced through an AGE-specific cell-surface receptor (AGE-R) complex (3,4), which has been identified on many cell types, including premyeloid cells, lymphoid cells, monocyte/macrophages, mesangial and endothelial cells, smooth muscle cells, and fibroblasts (1,17). Interaction of AGE-modified proteins with the AGE-R complex serves not only to degrade AGE proteins but also to induce the synthesis and release of cytokines (5) and growth factors (6,7), suggesting a dual purpose: disposal of senescent AGE-modified molecules and initiation of tissue repair and protein turnover. A novel regulatory pathway linked to extracellular matrix (ECM) synthesis and secretion has been suggested to be an important function of a novel AGE-binding receptor system identified on murine and human mesangial cells (8,9). Exposure of cultured mouse mesangial cells to AGE-bovine serum albumin results in a AGE receptor–mediated upregulation of mRNA and protein secretion of matrix proteins, such as fibronectin α1 type IV collagen and laminin A, B1, and B2 (8,9). The collagen α1IV mRNA increase is mediated, at least in culture, by platelet-derived growth factor (PDGF).

Direct in vivo evidence on the pathogenic influence of AGEs, independent of diabetes, was obtained recently. Studies in normal rats and rabbits, for example, showed that the short-term administration of in vitro prepared AGE-albumin can produce vascular defects similar to those associated with experimental diabetes, such as vascular leakage and mononuclear cell extravasation, as well as unresponsiveness to vasodilatory agents (10). In a subsequent study, the direct contribution of brief AGE injection on molecular events involved in mesangial cell responsiveness, such as the induction of ECM or growth factor genes, was tested (11). A predominant increase in α1IV collagen mRNA (by 1.7-fold) and in laminin B1 mRNA (by 2.2-fold) was observed only in mice receiving AGE-modified mouse serum albumin, whereas no changes were observed in mice cotreated with the AGE inhibitor aminoguanidine (11). Although in this study PDGF-B mRNA was not found to be increased, the mRNA of another growth-promoting molecule, transforming growth factor (TGF)-β1 was significantly increased (1.5-fold) in response to AGE-treated mice; again in AGE + aminogua-

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AGE, advanced glycosylation end product; AGE-R, AGE-specific cell-surface receptor; Apo, apolipoprotein; ECM, extracellular matrix; ESRD, end-stage renal disease; LMW-AGE, low molecular weight AGE; TGF, transforming growth factor.

nidine-treated mice, TGF-β mRNA remained normal. Prolonged exposure to AGEs induced glomerular hypertrophy, basement membrane thickening, and glomerulosclerosis, along with significant proteinuria/albuminuria (12). Cotreatment with aminoguanidine markedly limited both structural and functional defects, further strengthening and expanding the evidence that AGEs can influence independently glomerular structure and function.

Based on earlier studies, it is believed that tissue macrophages, via specific AGE receptors, constitute the principal mechanism for AGE-modified tissue catabolism. Intracellular digestion of AGEs is followed by the release of small soluble AGE peptides, which when combined with extracellular proteolysis of matrix components gives rise to a class of circulating low molecular weight substances likely to contain variable amounts of AGE moieties, depending on the underlying tissue levels and the activity of AGE removal systems. These degradation products of AGE-modified proteins (mostly low–molecular weight AGE-rich peptides [LMW-AGE]) are presumably released into the circulation to be cleared by the kidneys (2,13).

With a sensitive AGE-specific immunoassay (14), serum LMW-AGE levels have been found to correlate with renal function (2,13). In normal human control subjects, LMW-AGE clearance was estimated to be 0.72 ml/min (13). While diabetic individuals with normal glomerular filtration rates can clear AGE peptides at the same rate, progressive loss of kidney function correlates with increasing circulating LMW-AGE levels, up to eightfold in diabetic patients with ESRD requiring dialysis (2,13). Furthermore, all current modes of hemodialysis or peritoneal dialysis are apparently inefficient in removing AGE peptides, compared with creatinine, leaving renal transplantation as the only existing effective treatment (2,13).

Diabetic patients with ESRD are known to be particularly susceptible to cardiovascular complications due to accelerated atherosclerosis. Thus, the pronounced increase in serum AGEs observed in diabetic anephric patients, while indicative of inefficient clearance by current dialysis modalities, also has raised the possibility that uncleared recirculating AGEs can participate in undesired interactions with the vascular tissues and plasma lipoproteins, potentially accelerating ongoing pathological changes.

The toxic potential of the LMW-AGE–rich substances was suggested in studies showing that LMW-AGE peptides, when isolated from human serum, readily react and bind with collagen or plasma LDL in vitro, producing AGE collagen and AGE-LDL (13,15). This was consistent with the marked elevations in serum AGE-apolipoprotein (Apo) B that were found in diabetic and nondiabetic patients with impaired renal function (15). This extent of AGE modification of Apo B cannot be attributed to ambient glucose alone. On the contrary, the data are consistent with the disproportionally increased LMW-AGE found in the serum of both diabetic and nondiabetic uremic patients (2,13). It has been determined further that such AGE modification of LDL can result in a delayed clearance by the normal LDL receptor (16).

Thus, it can be concluded that not only glucose-derived AGE proteins but also their endogenously produced reactive derivatives, AGE peptides, can markedly exacerbate cellular and extracellular matrix protein dysfunction via AGE-receptor pathways. These responses may be accelerated in individuals who are genetically susceptible to diabetic renal and extrarenal disease. After kidney damage has occurred, renal insufficiency may serve to accelerate extrarenal vascular pathological changes by promoting AGE toxicity.

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