GLYCOXIDATIVE STRESS IN AGING AND PATHOLOGY

*Claudia **Borşa**[1], Daniela **Grădinaru**[2], Cristina **Ionescu**[1], Cătălina Monica **Pena**[1], Gabriel Ioan **Prada**[1,3]*

[1] "Ana Aslan" National Institute of Gerontology and Geriatrics, Bucharest, Romania
[2] "Carol Davila" University of Medicine and Pharmacy, Faculty of Pharmacy, Department of Biochemistry, Bucharest, Romania
[3] "Carol Davila" University of Medicine and Pharmacy, Faculty of Medicine, Department of Geriatric Medicine, Bucharest, Romania

Corresponding author: Claudia Borsa, cldbrs@yahoo.co.uk

Abstract. The glycoxidative stress is involved in aging and age-related pathologies. The advanced glycation end products (AGEs) and the activation of AGEs – receptor for AGEs (RAGE) axis may lead to functional decline in aging and to the onset and development of age-related diseases: metabolic, cardiovascular and neurodegenerative diseases. This minireview presents the AGEs involvement in aging and pathology, their mechanisms of action as well as the potential therapeutic strategies to counteract their deleterious effects. Systemic AGEs levels, biomarker of glycoxidative stress, could become a marker of overall health status. Inhibition of AGEs generation, AGEs crosslink formation and AGEs-RAGE axis activation could represent useful approaches in development of alternative, complementary or novel therapies in healthy aging and active longevity.
Key words: advanced glycation end products, receptor for advanced glycation end products, aging

STRESUL GLICOXIDATIV ÎN ÎMBĂTRÂNIRE ŞI PATOLOGIE

*Claudia **Borşa**[1], Daniela **Grădinaru**[2], Cristina **Ionescu**[1], Cătălina Monica **Pena**[1], Gabriel Ioan **Prada**[1,3]*

[1]Institutul Naţional de Gerontologie şi Geriatrie "Ana Aslan", Bucureşti, România
[2] Universitatea de Medicină şi Farmacie "Carol Davila", Facultatea de Farmacie, Bucureşti, România
[3]Universitatea de Medicina si Farmacie "Carol Davila", Facultatea de Medicină, Bucureşti, România

Autor corespondent: Claudia Borşa, cldbrs@yahoo.co.uk

Rezumat. Stresul glicoxidativ este implicat în îmbătrânire şi patologiile dependenţe de vârstă. Produsii finali de glicare avansată (AGEs) şi activarea axei AGEs- receptorul pentru AGEs (RAGE) pot conduce la declinul funcţional în îmbătrânire şi la apariţia şi dezvoltarea bolilor dependenţe de vârstă: metabolice, cardiovasculare şi neurodegenerative. Acest minireview prezintă implicarea AGEs în îmbătrânire şi patologie, mecanismele lor de acţiune precum şi potenţialele strategii terapeutice de a contracara efectele lor dăunătoare. Nivelele sistemice ale AGEs, biomarker al stresului glicoxidativ, ar putea deveni un marker al statusului de sănătate total. Inhibarea generării AGEs, a formării legăturilor încrucişate ale AGEs şi a activării axei AGEs-RAGE ar putea reprezenta cai utile în dezvoltarea terapiilor alternative, complementare sau noi în îmbătrânirea sănătoasă şi longevitatea activă.
Cuvinte cheie: produşii finali de glicare avansată, receptorul produşilor finali de glicare avansată, îmbătrânire

INTRODUCTION

The glycoxidative stress plays a key role in aging and age-associated or age-related pathologies, acting together with oxidative and nitrosative stress. The major consequences of glycoxidative stress include damage to proteins, lipids and DNA, dysfunction of cellular homeostasis and accumulation of damaged molecules. The end products of glycoxidation processes, named advanced glycation end products (AGEs) are age-accumulated, being also involved in the onset and progression of various age-associated diseases, such as: metabolic, cardiovascular or neurodegenerative diseases [1-5].

It has been documented that AGEs may have negative effects on vessels by modifying collagen and other proteins from vascular wall, rising vascular rigidity, thus contributing to the onset and progression of cardiovascular diseases. Also, AGEs may modify lipids and proteins from lipoproteins, especially low density lipoproteins (LDL), enhancing LDL capture in subendothelial compartments [6].

Experimental and clinical studies have underlined the AGEs involvement in oxidative stress enhancing, endothelial dysfunction, cardiovascular and degenerative diseases and even in rising the cardiovascular mortality risk [7, 8].

The specific consequences of glycoxidative stress and AGEs accumulation are direct related to the signal transduction receptor specific for glycoxidation compounds named advanced glycation end products receptor (RAGE). These consequences involve the activation and upregulation of RAGE and RAGE-dependent cellular signal pathways, the sustaining and intensification of pro-inflammatory, pro-trombotic and atherosclerotic processes, the enhancement of oxidative, metabolic and nitrosative stress, cellular stress maintaining, cellular and tisular dysfunctions, damages and modifications specifically to human aging, especially to vascular aging [3, 4, 8].

The AGEs-RAGE interraction contributes to free radicals generation, pro-inflammatory cytokines release, modifications of extracellular matrix or hormons action. Numerous studies have underlined that the inhibition of AGEs generation, AGEs tissular accumulation and of glycoxidative stress may conduct to the elimination or amelioration of their damaging effects [6, 9].

By binding to the specific transmembranar receptor RAGE, the AGEs are involved in various pro-oxidant or pro-inflammatory pathways activated by the nuclear factor NF-kB. The AGEs-RAGE axis contributes to cellular signaling either on NF-kB pathway, or by activation of extracellular signal-regulated kinases (ERK), named ERK1 and ERK2 [3, 6, 9].

The inhibition of AGEs production and AGEs-RAGE axis activation has been shown to prevent the pathogenesis of age-associated diseases in human. The therapeutic advances in this field have resulted in several agents that may prevent the adverse effects of glycoxidative stress [6, 9].

This minireview presents the AGEs involvement in aging and pathology, their mechanisms of action as well as the potential therapeutic strategies to counteract their deleterious effects.

Clinical studies on glycoxidative stress, aging and age-related pathology

The accumulation of AGEs and their crosslinks with long-lived proteins during aging may contribute to the age-related decline of the functioning of cells, tissues and organs in normal aging. Also, AGEs may activate RAGE signaling pathways leading to enhancement of oxidative stress and inflammation conducting to endothelial and vascular dysfunctions. On the other hand the oxidative stress could enhance glycoxidative stress by rising AGEs production. The important studied

biomarkers of glycoxidative stress are represented by: N-ε-carboxy-ethyl-lysine (CEL), N-ε-carboxy-methyl-lysine (CML), imidazolone, methyl-glyoxal-lysine dimer (MOLD), glyoxal-lysine dimer (GOLD), pyrraline, glucosepane and pentosidine, or fluorescent AGEs [2, 3, 6, 9].

Recent clinical studies, presented in table I, have underlined the associations of AGEs with oxidative stress, inflammation, cardiovascular risk and even cardiovascular mortality, in healthy elderly [1, 2, 5, 7, 10, 11-13]. Moreover, AGEs levels were related to cognitive decline in elderly with and without age-associated diseases [14].

Many clinical studies (Tab. I) have also linked AGEs to cardiovascular disease (CVD), particularly in elderly patients with impaired glucose metabolism or metabolic syndrome [11, 12, 14-18]. Thus, AGEs levels were associated with metabolic, endothelial, oxidative stress or cardiovascular disease markers in elderly with IFG, T2DM and metabolic syndrome. These interrelations were also underlined in our recent researches (Tab. I). We pointed out significant positive correlations between systemic AGEs levels with oxidized low density lipoproteins (oxLDL), nitric oxide metabolic pathways products (NOx), insulin resistance (IR), interleukin-6 (IL-6), atherogenic and CVD risk markers in elderly with impaired glucose metabolism and metabolic syndrome [15, 16, 18].

The AGEs-RAGE axis activation leads to initiation of intracellular signaling, production of proinflammatory cytokines and free radicals. These processes may be limited by action of soluble RAGE (sRAGE), which counteracts the deleterious effects of glycoxidative stress by binding AGEs [3, 6, 9]. Our studies pointed out elevated levels of AGEs and AGEs/sRAGE ratio in elderly with T2DM and their relationships with metabolic, oxidative and cardiovascular markers (table I). Thus, our studies underlined that the AGEs/sRAGE ratio may be a more reliable marker of cumulative effects of AGEs, sRAGE and AGEs-RAGE axis [18]. All these studies underline the fact that AGEs levels, biomarker of glycoxidative stress could become a marker of overall health. Future researches are needed to well establish this fact.

Tab. I Clinical studies underlining the glycoxidative stress associations with oxidative, inflammatory and cardiovascular disease markers in healthy aging and age-associated diseases

Subjects	**Glycoxidative stress associations**	**References**
Healthy elderly	AGEs accumulate with age, AGEs ≈ OS and inflammatory markers (CRP)	Uribarri et al. 2005, 2007 [1,2]
Healthy adults and elderly	AGEs ≈ > OS, IR, inflammation in aging	Uribarri et al. 2007 [2] Meigs et al. 2007 [10]
Healthy elderly	CML ≈ CV mortality	Semba et al. 2009 [5]
Women with and without diabetes	↑ AGEs predict CV and coronary mortality	Kilhovd et al. 2005, 2007 [11]
Disabled older women	CML ≈ CV mortality	Semba et al. 2009a [12]
Older adults	CML ≈ increased risk of CV events; CML ≈ age, albuminuria, BP, IR	Kiser et al. 2014 [13]
Young, adults and elderly	Cardiac and systemic CML ≈ age, DM, CHD	Hu et al. 2014 [7]
Elderly with and without T2DM	↑ AGEs ≈ ↑ cognitive decline	Yaffe et al. 2011 [14]
Elderly with IFG and T2DM	↑ AGEs; AGEs ≈ FG, HbA1c, IR, oxLDL, NOx, CVD markers	Gradinaru et al. 2013 [15]

Elderly with metabolic syndrome	↑ AGEs; AGEs ≈ adiponectin	Gradinaru et al. 2016 [16]
Adults and elderly	AGEs ≈ endothelial dysfunction (FMD, NIV)	Kajikava et al. 2015 [17]
Elderly with T2DM	↑ AGEs; ↑ AGEs/sRAGE AGEs/sRAGE ≈ FG, Ai, IL-6, CVD markers	Borsa et al. 2017 [18]

≈, associated with
↑, higher values
AGEs, advanced glycation end products; Ai, atherogenic index; BP, blood pressure; CHD, coronary heart disease; CML, carboxy-methyl-lysine; CRP, C-reactive protein; CV, cardiovascular; CVD, cardiovascular disease; DM, diabetes mellitus; FG, fasting glucose; FMD, flow-mediated vasodilation, HbA1c, hemoglobin A1c; IL-6, interleukin -6; IR, insulin resistance; oxLDL, oxidized low density lipoproteins; NIV, nitroglycerine-induced vasodilation, NOx, nitric oxide metabolic pathways products; OS, oxidative stress; sRAGE, soluble receptor for advanced glycation end products

Molecular mechanisms of glycoxidative stress in aging and pathology

The glycoxidative stress activates important signalling pathways leading to enhance oxidative stress and inflammation. The cellular AGEs generation and accumulation are age-related and AGEs levels represent a reliable biomarker of "in vivo" aging. Two pathways are involved in AGEs contribution to aging and age-related disorders: AGEs receptor-independent pathway and AGEs receptor-dependent pathway. The AGEs receptor-independent pathway consists in accumulation of AGEs and AGEs crosslinks with long-live proteins, like is: haemoglobin, albumin and collagen; altering their structure, conformation, properties and reactivity. The AGEs receptor-dependent pathway activates signalling pathways for inducing oxidative stress and inflammation [2, 3, 8].

The AGEs-RAGE axis activation may increase ROS production and oxidative stress by two pathways: activation of mitochondrial electron transport chain and nicotinamide adenine dinucleotide phosphate (NADPH) oxidase. Also, the AGEs-RAGE axis activation and downstream signalling lead to induction of inflammatory mediators, such as: tumor necrosis factor-alpha (TNF-a), interleukin-1 (IL-1), interleukin-6 (IL-6), C-reactive protein (CRP); as well as activation of genes for inducible nitric oxide synthase (iNOS). The AGEs-RAGE axis contributes to cellular signaling either on NF-kB pathway, or by activation of extracellular signal-regulated kinases (ERK), named ERK1 and ERK2. AGEs- and RAGE-mediated ROS production and enhanced oxidative stress are involved in endothelial dysfunction, vascular stiffness, metabolic, cardiovascular and neurodegenerative diseases [3, 6, 9].

The RAGE-dependent signalling may be counteracted by advanced glycation receptor 1 (AGER 1) and soluble forms of RAGE, which bind AGEs, inactivate AGEs-RAGE axis, diminish cellular signalling related to oxidative stress, inflammation and RAGE-mediated pathogenesis [6].

Strategies targeting glycoxidative stress – related damages

Strategies for prevention and treatment of human AGEs-related disorders have been conducted on three directions:

1. inhibition of AGEs generation
2. degradation of AGEs and AGEs crosslinks with proteins
3. competitive inhibition of AGEs-RAGE axis activation

Experimental and clinical studies have underlined many agents that may counteract the damages induced by glycoxidative stress. These agents are classified as inhibitors of AGEs generation, breakers of AGEs crosslinks or antagonists of AGEs-RAGE signaling [4, 6, 9]. Some

representative agents, their actions and biological effects are presented in Tab. II.

The inhibitors of AGEs formation act either by targeting different inducers (ROS, metal ions) or intermediate products, especially reactive carbonyl species (RCS). In this category are included RCS quenchers, metal ion chelators, antioxidants or xenobiotics [6, 19-25].

The AGEs crosslings breakers have also the potential to effectively reduce AGEs products and act to lower the cardiovascular complication associated with aging and age-related diseases. Alagebrium and related compounds including: ALT-711, ALT-462, ALT-486, ALT-946 act as breakers of AGEs crosslinks with proteins [26-29].

RAGE activation and RAGE-mediated signal transduction are involved in aging and pathogenic mechanisms of age-related diseases. The AGEs-RAGE axis represents an important drug target and inhibition of axis activation exerts beneficial effects in various pathologies [6, 9].

The molecular strategies for inhibition the RAGE activation [30] consist in:

- inactivation of RAGE ligands by RAGE soluble forms
- inactivation of RAGE by antagonists or antibodies
- down-regulation of RAGE expression
- inhibition of RAGE signal transduction

Suppression of RAGE activation could be useful in preventing and slowing aging and age-related pathologies. Potential compounds for RAGE inhibition include:

- inhibitors of RAGE expression
- RAGE antagonists
- blockers of RAGE intracellular signaling
- enhancers of sRAGE and esRAGE production
- sRAGE supplementation

The antagonists of AGEs-RAGE signaling are small molecules that act by blocking RAGE receptor activation, RAGE dependent molecular processes and inhibition of RAGE ligand- triggered signal transduction. Recently, clinical trials studies have tested RAGE antagonists which may block the binding of ligand to extracellular domain of RAGE, such as azeliragone or compound FPS-ZM127. Also, soluble RAGE-type molecules are under study [31].

Recent studies have underlined the important role of exogenous-derived AGEs in rising glycoxidative stress. Thus, the restriction of dietary AGEs may lead to a fall in systemic AGEs and a decrease in biomarkers of oxidative stress, inflammation, endothelial dysfunction and insulin resistance in aging and age-related pathologies [6].

Tab. II Therapeutic agents for glycoxidative stress-related pathology

Compounds	**Actions**	**Effects**	**References**
AGEs inhibitors			
amonoguanidine	traps aldehyde group and reactive carbonyl intermediates	reduces nephropathy and retinopathy	Brownlee et al. 1994 [19]
piridoxamine	traps aldehyde group and reactive carbonyl intermediates, scavenges ROS, inhibits post-Amadori stages of AGEs formation	reduces diabetes and non-diabetes nephropathy and retinopathy	Degehhardt et al. 2002 [20] Stitt et al. 2002 [21] Voziyan et al. 2005 [25]
benfotiamine	reduces AGEs content in renal cells; reduces AGEs effects on endothelial functions	improves microalbuminuria; improves endothelial functions	Babaci-Jadidi et al. 2003 [23] Stirban et al. 2006 [6]

valsartan	reduces plasma and urinary pentosidine and plasma CML, scavenges ROS, chelation of transition metals	antihypertensive, antioxidant and anti- glycoxidation effects	Monacelli et al. 2006 [24]
polyphenols	chelation of transition metals, antioxidant activity	antioxidant effects, reduce albumin glycoxidation	Sadowska-Bartosz et al. 2014 [25]
AGEs breakers			
ALT-711 (dimethyl-3-phenyl-thiazolium chloride; N-phenocylthiazolium; N-phenocyl-4,5-dimethyl-thiazolium	breaks AGEs crosslinks via thiazolium structure	reduces cardiovascular complications in aging and diabetes; rises skin hydration; improves arterial compliance and cardiac function in elderly	Vasan et al. 2003 [26] Bakris et al. 2004 27] Monnier et al. 2006 [28] Little et al. 2005 [29]
Antagonists of AGEs-RAGE signalling			
Small molecules inhibitors of RAGE signalling	bind to ctRAGE; interfere to ctRAGE-DIAPH1 complex	inhibition of ctRAGE interaction with DIAPH1; inhibition of RAGE dependent molecular processes; inhibition of RAGE ligand- triggered signal transduction	Manigrasso et al. 2016 [31]

AGEs, advanced glycation end products; CML, carboxy-methyl-lysine; ROS, reactive oxygen species; RAGE, receptor for advanced glycation end products, ctRAGE, RAGE cytoplasmic tail; DIAPH1, mammalian diaphanous 1

CONCLUSIONS

Glycoxidative stress affecting all long-lived proteins and cell types leads to enhancement of oxidative stress, inflammation, endothelial and vascular dysfunctions.

Systemic AGEs levels, biomarker of glycoxidative stress, are associated with age and age-related diseases and could become a marker of overall health status.

Receptor-independent and RAGE-dependent mechanisms mediates the AGEs biological effects, especially ROS generation and stimulation of inflammatory pathways.

Inhibition of AGEs generation, AGEs crosslink formation and AGEs-RAGE axis activation could represent useful approaches in development of alternative, complementary or novel therapies in healthy aging and active longevity or age-related diseases.

Conflicts of interest

The authors declare no conflicts of interest.

REFERENCES

[1] Uribarri J, Cai W, Sandu O, et al. *Diet-derived advanced glycation end products are major contributors to the body's AGE pool and induce inflammation in healthy subjects.* Ann. N Y Acad. Sci. 2005, 1043, 461-466.

[2] Uribarri J, Cai W, Peppa M, et al. *Circulating glycotoxins and dietary advanced glycation end products: two links to inflammatory response, oxidative stress, and aging.* J. Gerontol. A Biol Sci Med Sci. 2007, 62, 427-433.

[3] Simm A, Wagner J, Gursinsky, et al. *Advanced glycation endproducts: A biomarker for age as an outcome predictor after cardiac surgery?* Experimental Gerontology 2007, 42, 668–675.

[4] Yan SF, Ramasamy R, Schmidt AM. *Soluble RAGE: therapy and biomarker in unraveling the RAGE axis in chronic disease and aging.* Biochem Pharmacol. 2010, 79, 1379-86.

[5] Semba RD, Ferrucci L, Sun K, et al. *Advanced glycation end products and their circulating receptors predict cardiovascular disease mortality in older community-dwelling women.* Aging clinical and experimental research 2009, 21, 182–190.
[6] Stirban A, Gawlowski T, Roden M. *Vascular effects of advanced glycation endproducts: Clinical effects and molecular mechanisms*. Molecular Metabolism 2014, 3, 94–108.
[7] Hu S, He W, Liu Z, et al. *Glycoxidation product N-carboxymethyl-lisine in cardiac tissue with age, diabetes mellitus and coronary heart disease*. Tokoku J. Exp. Med 2013, 230, 25-32.
[8] Peppa M, Uribarri J, Vlassara H. *Aging and glycoxidant stress.* Hormones 2008, 7, 2, 123-132.
[9] Byun K, Yoo YC, Lee MJ, et al. *Advanced glycation end-products produced systemically and by macrophages: A common contributor to inflammation and degenerative diseases.* Pharmacology & Therapeutics 2017, 177, 44–55.
[10] Meigs JB, Larson MG, Fox CS, et al. *Association of oxidative stress, insulin resistance, and diabetes risk phenotypes: the Framingham Offspring Study.* Diabetes Care 2007, 30, 2529-2535.
[11] Kilhovd BK, Juutilainen A, Lehto S, et al. *Increased serum levels of advanced glycation end products predict total, cardiovascular and coronary mortality in women with type 2 diabetes: A population-based 18 year follow-up study.* Diabetologia 2007, 50, 1409–1417.
[12] Semba RD, Bandinelli S, Sun K, et al. *Plasma carboxymethyl-lysine, an advanced glycation end product, and all-cause and cardiovascular disease mortality in older community-dwelling adults*. J. Am. Geriatr. Soc 2009, 57, 1874–1880.
[13] Kizer JR, Benkeser D, Arnold AM, et al. *Advanced glycation/glycoxidation end product carboxymethyl-lysine and incidence of coronary heart disease and stroke in older adults.* Atherosclerosis 2014, 235,1, 116–121.
[14] Yaffe K, Lindquist K, Schwartz AV, et al. *Advanced glycation end product level, diabetes, and accelerated cognitive aging.* Neurology 2011, 77, 1351–1356.
[15] Gradinaru D, Borsa C, Ionescu C, et al. *Advanced oxidative and glycoxidative protein damage markers in the elderly with type 2 diabetes*. Journal of Proteomics 2013, 92, 313-22.
[16] Gradinaru D, Margina D, Borsa C, et al. *Adiponectin: possible link between metabolic stress and oxidative stress in the elderly.* Aging Clin. Exp. Res 2017, 29, 4, 621-629.
[17] Kajikawa M, Nakashima A, Fujimura N, et al. *Ratio of serum levels of AGEs to soluble form of RAGE is a predictor of endothelial function.* Diabetes Care 2015, 38,119–125.
[18] Borsa C, Gradinaru D, Margina D, et al. *Receptor of advanced glycation end products and cardiovascular risk in elderly with type 2 diabetes mellitus*. J. Biological Research 2017, 90, 2, 6462. DOI: 10.4081/jbr:2017.6462
[19] Brownlee M, Vlassara H, Kooney A, et al. *Aminoguanidine prevents diabetes-induced arterial wall protein cross-linking.* Science 1986, 232, 1629-1632.
[20] Degenhardt TP, Alderson NL, Arrington DD, et al. *Pyridoxamine inhibits early renal disease and dyslipidemia in the streptozotocin-diabetic rat.* Kidney Int 2002, 61, 939-950.
[21] Stitt A, Gardiner TA, Alderson NL, et al. *The AGE inhibitor pyridoxamine inhibits development of retinopathy in experimental diabetes.* Diabetes 2002, 51, 2826-2832.
[22] Voziyan PA, Hudson BG. *Pyridoxamine: the many virtues of a maillard reaction inhibitor.* Ann. NY Acad. Sci 2005, 1043, 807-816.
[23] Babaei-Jadidi R, Karachalias N, Ahmed N, et al. *Prevention of incipient diabetic nephropathy by high-dose thiamine and benfotiamine.* Diabetes 2003, 52, 2110-2120.
[24] Monacelli F, Poggi A, Storace D, et al. *Effects of valsartan therapy on protein glycoxidation* 2006, 55, 1619-24.
[25] Sadowska-Bartosz I, Galiniak S, Bartosz G. *Polyphenols protect against protein glycoxidation.* Free Radic. Biol. Med. 2014, 75, Suppl 1, S47.
[26] Vasan S, Foiles P, Founds H. *Therapeutic potential of breakers of advanced glycation end product-protein crosslinks.* Arch. Biochem. Biophys 2003, 419, 89-96.
[27] Bakris GL, Bank AJ, Kass DA, et al. *Advanced glycation end-product cross-link breakers. A novel approach to cardiovascular pathologies related to the aging process*. Am. J. Hypertens 2004, 17, 23S-30S.
[28] Monnier VM, Sell DR. *Prevention and repair of protein damage by the Maillard reaction in vivo.* Rejuvenation Res 2006, 9, 264-73.
[29] Little WC, Zile MR, Kitzman DW, et al. *The effect of alagebrium chloride (ALT-711), a novel glucose cross-link breaker, in the treatment of elderly patients with diastolic heart failure*. J. Card. Fail 2005, 11,191–195.
[30] Aldini G, Vistoli G, Stefek M, et al. *Molecular strategies to prevent, inhibit, and degrade advanced glycoxidation and advanced lipoxidation end products.* Free Radical Research 2013, 47, 1, 93-137.
[31] Manigrasso MB, Pan J, Rai V, et al. *Small molecule inhibition of ligand-stimulated RAGE-DIAPH1 signal transduction*. Scientific Reports 2016, 6, 22450. DOI: 10.1038/srep22450