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Original article

Dietary intake of advanced glycation endproducts is associated with higher levels of advanced glycation endproducts in plasma and urine: The CODAM study

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SUMMARY

Background & aims: Advanced glycation endproducts (AGEs) are formed by the reaction between reducing sugars and proteins. AGEs in the body have been associated with several age-related diseases. High-heat treated and most processed foods are rich in AGEs. The aim of our study was to investigate whether dietary AGEs, are associated with plasma and urinary AGE levels.

Methods: In 450 participants of the Cohort on Diabetes and Atherosclerosis Maastricht study (CODAM study) we measured plasma and urine concentrations of the AGEs Nε-(carboxymethyl)lysine (CML), Nε-(1-carboxyethyl)lysine (CEL) and Nδ-(5-hydro-5-methyl-4-imidazolon-2-yl)-ornithine (MG-H1) using UPLC-MS/MS. We also estimated dietary intake of CML, CEL and MG-H1 with the use of a dietary AGE database and a food frequency questionnaire (FFQ). We used linear regression to investigate the association between standardized dietary AGE intake and standardized plasma or urinary AGE levels, after adjustment for age, sex, glucose metabolism status, waist circumference, kidney function, energy- and macro-nutrient intake, smoking status, physical activity, alcohol intake, LDL-cholesterol and markers of oxidative stress.

Results: We found that higher intake of dietary CML, CEL and MG-H1 was associated with significantly higher levels of free plasma and urinary CML, CEL and MG-H1 (βCML = 0.253 (95% CI 0.086; 0.415), βCEL = 0.194 (95% CI 0.040; 0.339), βMG-H1 = 0.223 (95% CI 0.069; 0.373) for plasma and βCML = 0.223 (95% CI 0.049; 0.393), βCEL = 0.180 (95% CI 0.019; 0.332), βMG-H1 = 0.196 (95% CI 0.037; 0.349) for urine, respectively). In addition, we observed non-significant associations of dietary AGEs with their corresponding protein bound plasma AGEs.

Conclusion: We demonstrate that higher intake of dietary AGEs is associated with higher levels of AGEs in plasma and urine. Our findings may have important implications for those who ingest a diet rich in AGEs.

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1. Introduction

Advanced glycation endproducts (AGEs) are a large and heterogeneous family of sugar-modified proteins, which have been linked to a plethora of age-related diseases including diabetes, atherosclerosis, chronic obstructive pulmonary disease and Alzheimer's disease [1–3].

In 1912, the French chemist Louis Camille Maillard discovered the reaction between reducing sugars and free amino acids on proteins [4], leading to the formation of AGEs. At body temperature,

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the Maillard reaction takes place at a very slow pace. In contrast, in foods which had a prolonged exposure to high heat AGEs were formed rapidly [5]. The consumption of processed foods that were cooked at high temperatures has increased over the past decades [6]. As a consequence, the exposure to dietary AGEs have also increased and may be a risk factor for chronic diseases [7]. Indeed, recent studies have shown associations of dietary AGEs with insulin sensitivity [8], abdominal obesity and hypertriglyceridemia [9] and with poorer memory in Alzheimer's disease [10]. AGEs in the body may contribute to development of age-related diseases through several mechanisms, such as interaction with the receptor for AGEs (RAGE) [11] and crosslinking on long-lived proteins [12]. However, whether dietary AGEs are substantially absorbed in the digestive tract and released into the circulation to contribute to any of these effects remains unclear.

The studies so far addressing whether high intake of dietary AGEs lead to increased plasma and urinary AGEs have not been conclusive [13–22], because most of these studies have been small and have not taken into account confounding factors such as kidney function or dietary energy intake, which may explain the associations [19,20]. Moreover, different analytical techniques to analyze AGE content in food and plasma samples were used in these studies, which may have led to differences in AGE concentrations [21,22].

Taken these considerations into account, the aim of the current study was to establish whether higher intake of dietary AGEs are associated with higher levels of plasma and urinary AGEs. In a previous study, we developed a new dietary AGE database of three major AGEs Nε-(carboxymethyl)lysine (CML), Nε-(1-carboxyethyl) lysine (CEL) and Nδ-(5-hydro-5-methyl-4-imidazolon-2-yl)-ornithine (MG-H1), as analyzed in 190 specific food items, based on a state-of-the art ultra-performance liquid chromatography tandem mass spectrometry (UPLC-MS/MS) technique [23]. In the present study, we linked information on AGE-content of these food items to a food frequency questionnaire (FFQ) and calculated the consumption of AGEs in a cohort of 465 participants. We subsequently examined the association of dietary AGE intake with three major plasma and urinary AGEs (CML CEL and MG-H1), adjusting for major potential confounders.

2. Materials and methods

2.1. Study population

Cross-sectional analyses were performed on data from the Cohort on Diabetes and Atherosclerosis Maastricht study (CODAM), which includes 574 individuals with an elevated risk for T2DM and cardiovascular disease and described in detail elsewhere [24]. Participants were instructed to withhold their lipid-lowering medication for a fortnight prior the first visit, and not to consume any alcoholic drinks, coffee and/or tea, not to smoke, and withhold all other medication the day before. The habitual dietary intake over the last twelve months of all participants was established by a self-administered food frequency questionnaire (FFQ) which queried 194 foods [25].

Individuals were excluded if they did not qualify to report the FFQ successfully (n = 56, i.e. more than 10% items missing on the FFQ). Participants who reported an energy intake outside the range of 800–4200 kcal/day for men and 600–3500 kcal/day for women were also excluded (n = 6). Due to sample availability, 450 participants were used for statistical analysis (Fig. 1).

Fasting and 2-h postload glucose concentrations were used to classify the study participants' glucose metabolism status (GMS), described in details elsewhere [26]. Questionnaires were used to assess smoking behavior (never, ever, or current smoker) and use of medication (lipid-, glucose-, and blood pressure-lowering medication). Plasma creatinine levels were measured with the Jaffe diagnostic test (Roche Diagnostics, Mannheim, Germany), and the estimated glomerular filtration rate (eGFR) was calculated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation [27]. Plasma malondialdehyde (MDA) levels were measured with a reagent kit for high-performance liquid chromatography analyses (Chromsystems Instruments and Chemicals, Munich, Germany), and total antioxidative status (TAS) was measured in serum with an enzymatic kit (Randox Diagnostics, County Antrim, U.K.). LDL cholesterol was calculated with the Friedewald formula [28], after measurement of fasting total cholesterol, HDL-cholesterol and triglyceride levels.

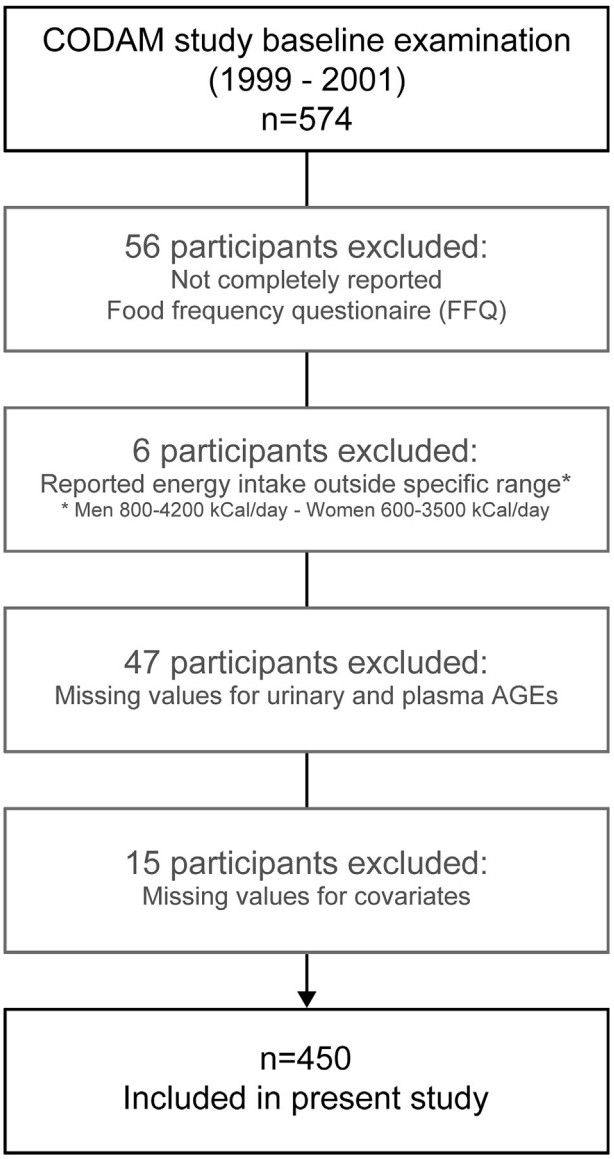

Fig. 1. Flowchart for the exclusion of participants in the current CODAM study.

All subjects gave written informed consent. The study was approved by the local Medical Ethical Committee of the University of Maastricht and University Hospital Maastricht.

2.2. Food, plasma and urinary AGE quantification

Protein-bound AGEs in individual food items and in plasma, and free AGEs in plasma and in urine were analyzed as described in

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