Soluble Receptor for Advanced Glycation End-products (sRAGE) and Colorectal Cancer Risk: A Case–Control Study Nested within a European Prospective Cohort

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ABSTRACT

Background: Overexpression of the receptor for advanced glycation end-product (RAGE) has been associated with chronic inflammation, which in turn has been associated with increased colorectal cancer risk. Soluble RAGE (sRAGE) competes with RAGE to bind its ligands, thus potentially preventing RAGE-induced inflammation.

Methods: To investigate whether sRAGE and related genetic variants are associated with colorectal cancer risk, we conducted a nested case–control study in the European Prospective Investigation into Cancer and Nutrition (EPIC). Plasma sRAGE concentrations were measured by ELISA in 1,361 colorectal cancer matched case–control sets. Twenty-four SNPs encoded in the genes associated with sRAGE concentrations were available for 1,985 colorectal cancer cases and 2,220 controls. Multivariable adjusted ORs and 95% confidence intervals (CIs) were computed using conditional and unconditional logistic regression for colorectal cancer risk and circulating sRAGE and SNPs, respectively.

Results: Higher sRAGE concentrations were inversely associated with colorectal cancer ($OR_{Q5vs.Q1}$, 0.77; 95% CI, 0.59–1.00). Sex-specific analyses revealed that the observed inverse risk association was restricted to men ($OR_{Q5vs.Q1}$, 0.63; 95% CI, 0.42–0.94), whereas no association was observed in women ($OR_{Q5vs.Q1}$, 1.00; 95% CI, 0.68–1.48; $P_{heterogeneity}$ for sex = 0.006). Participants carrying minor allele of rs653765 (promoter region of *ADAM10*) had lower colorectal cancer risk (C vs. T, OR, 0.90; 95% CI, 0.82–0.99).

Conclusions: Prediagnostic sRAGE concentrations were inversely associated with colorectal cancer risk in men, but not in women. An SNP located within *ADAM10* gene, pertaining to RAGE shedding, was associated with colorectal cancer risk.

Impact: Further studies are needed to confirm our observed sex difference in the association and better explore the potential involvement of genetic variants of sRAGE in colorectal cancer development.

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Introduction

Advanced glycation end-products (AGE) are a heterogeneous group of molecules formed by nonenzymatic reactions between reducing sugars and proteins, lipids, or nucleic acids (1). AGEs are produced endogenously, but diet and lifestyle are likely the largest contributors to the overall AGEs pool particularly from high-temperature processed food products, which contain high amounts of AGEs and/or their precursors (2–4). Glycated proteins tend to become dysfunctional and agglutinate with other reacting molecules to create cross-links and aggregates which can accumulate within diverse tissues in the body (5). The accumulation of AGEs throughout the life course is thought to contribute to intracellular signaling alterations, chronic low-level inflammation, and a decrease in tissue functionality (6).

AGEs are recognized by a multi-ligand cell surface protein receptor, known as the receptor for AGE (RAGE). RAGE consists of an extracellular N-terminal, a transmembrane helix, and an intracellular C-terminal tail (7). RAGE is expressed at low levels in most tissue types, except the lungs in which the expression is generally high (8). Overexpression of RAGE and its high activity have been demonstrated in various cancers including in the colon, breast, brain, prostate, and in the ovaries (9). Binding of AGEs to their receptor triggers a signaling cascade leading to intracellular inflammation with activation of NF-κB, increased secretion of cytokines and chemokines, and elevated production of reactive oxygen and nitrogen species (10).

Soluble RAGE (sRAGE) is a free circulating isoform of RAGE that also binds AGEs and acts as a decoy for RAGE. In contrast to RAGE, binding of AGEs to sRAGE does not induce inflammation and oxidative stress (8). Although the concentration of sRAGE is likely insufficient to bind all circulating AGEs (11), higher sRAGE levels had been associated with low inflammation and lower risk of several chronic diseases, including cancers (12). The variability in sRAGE concentrations is considerably affected by a combination of genetic and environmental factors (13). sRAGE levels have been reported to be elevated in women versus men, younger versus older individuals, and individuals with normal weight versus individuals with overweight and obesity (14–17). Furthermore, genetic determinants of sRAGE expression have also been identified and include SNPs located within advanced glycosylation end-product specific receptor (*AGER*), a disintegrin and metalloproteinase domain 10 (*ADAM10*), glyoxalase I (*GLO1*), and ring finger protein 5 (*RNF5*) genes (17–21).

We hypothesized that higher circulating sRAGE levels are inversely associated with colorectal cancer development. Previously, only two prospective studies have investigated the association, and showed an inverse association of high sRAGE concentrations with colorectal cancer risk among Finnish male smokers (22) and women with overweight and obesity (23). However, there are sparse data from other prospective studies, and there is a need to carefully investigate possible differences in the association by sex or lifestyle factors. To address these gaps, we studied the association between prediagnostic levels of circulating sRAGE and risk of colorectal cancer in a large, multinational European prospective cohort. We also investigated whether SNPs, reported to be related to sRAGE levels or RAGE function, are associated with colorectal cancer risk.

Materials and Methods

Study population and data collection

We used a case–control design nested within the European Prospective Investigation into Cancer and Nutrition (EPIC) cohort. EPIC is an ongoing multicenter prospective cohort with 521,324 participants (70% women) recruited from 23 study centers located in 10 European countries (Denmark, France, Germany, Greece, Italy, the Netherlands, Norway, Spain, Sweden, and the United Kingdom). The rationale and methods of the EPIC study, including information on the recruitment of the participants as well as data collection, have been described previously (24). Participants provided written informed consent before joining the EPIC study. Participant's health history, anthropometry, and socio-demographic and standardized lifestyle variables, including education, smoking, and physical activity, were collected by questionnaire at baseline, prior to disease onset or diagnosis. Physical activity was based on the Cambridge physical activity index: inactive (sedentary job and no recreational activity), moderately inactive (sedentary job with <0.5 hours recreational activity per day/or standing job with no recreational activity), moderately active (sedentary job with 0.5–1 hour recreational activity per day/or standing job with 0.5 hours recreational activity per day/or physical job with no recreational activity), or active (sedentary job with >1 hour recreational activity per day/or standing job with >0.5 hours recreational activity per day/or physical job with at least some recreational activity/or heavy manual job; ref. 25). Dietary intake was assessed at recruitment by validated center-specific questionnaires. At each of the study centers, blood samples were drawn at recruitment (∼80% of participants provided blood samples) and stored in liquid nitrogen (−196°C, liquid nitrogen) at the International Agency for Research on Cancer (Lyon, France) biobank, or in local biobanks (at −150°C in nitrogen vapor in Denmark and −80°C freezers at Malmö and Umeå centers in Sweden; ref. 24).

Follow-up for cancer incidence and vital status

Vital status follow-up (98.4% complete) was collected by record linkage with regional and/or national mortality registries in all countries, except Germany and Greece, and the Italian center of Naples, where data are collected actively. Incident cancer cases were identified through record linkage with regional cancer registries or using a combination of methods, including health insurance records, cancer and pathology registries, and active follow-up through participants and their relatives. Colorectal cancer cases were eligible if they were

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Note: Supplementary data for this article are available at Cancer Epidemiology, Biomarkers & Prevention Online (http://cebp.aacrjournals.org/).

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first incident and histologically confirmed. Cases were defined using the International Classification of Diseases for Oncology. Colon cancers were defined as tumors that occurred in the cecum, appendix, ascending colon, hepatic flexure, transverse colon, splenic flexure, descending and sigmoid colon (C18.0–C18.7), and overlapping and or unspecified origin tumors (C18.8 and C18.9). Rectal cancers were defined as tumors that occurred at the recto-sigmoid junction (C19) or rectum (C20). Cancers of the anal canal were excluded.

Case–control design

From baseline onwards, 1,413 first incident colorectal cancer cases with available blood samples were identified (until June 2003 as endpoint) among all the 2,476 colorectal cancer cases ascertained (**Fig. 1**). For each identified case, one control was matched by incidence density sampling from all cohort members alive and cancer free at the time of diagnosis of the index case. Cases and controls were matched by age (±1 year), sex, center, and blood collection details, including time (±3 hours) and fasting prevenepuncture (<3, 3–6, and >6 hours); and additionally among women only, by menopausal status (pre-, peri-, and postmenopausal) and hormone replacement therapy use at the time of blood collection (yes/no). After exclusion of participants with incomplete matched case sets ($n = 16$), those with extreme sRAGE levels ($n = 3$ controls and one case with sRAGE concentrations unusually high, i.e., >mean + 4 SD), and 32 cases and matched controls from Greece due to unforeseen data restriction issues, 1,361 cases and 1,361 matched controls were included in the sRAGE analysis. Among EPIC participants, 4,487 participants (until December 2012 as endpoint, 2,148 colorectal cancer cases and 2,339 matched controls) have been genotyped previously. After exclusion of 100 colorectal cancer cases and 100 matched controls from Greece and 82 participants with missing lifestyle variable, 1,985 colorectal cancer cases and 2,220 matched controls were included in the genetic analysis. Among the participants who have been genotyped, 972 colorectal cancer cases and 767 noncases overlapped with case–control sets in whom sRAGE measurements were conducted.

Laboratory analyses

Circulating sRAGE concentrations were measured in citrated plasma samples by ELISA (Quantikine, R&D Systems), following the manufacturer's instructions. Previous studies have reported that sRAGE is stable in plasma over a long period of time (26). Analyses were run with case–control sets randomized across batches ($n = 40$ batches, with an average of 35 case–control pairs analyzed per batch). Intra- and interbatch coefficients of variation (CV) were assessed by measuring three different samples used as quality controls in duplicate in each. Mean intra- and interbatch CVs were 1.25% and 6.0%, respectively. C-reactive protein (CRP) concentrations were determined using a high-sensitivity assay (Beckman-Coulter).

DNA genotyping and genetic variants selection

DNA was extracted from buffy coats from citrated blood samples at the Center for Inherited Disease Research (Johns Hopkins University, Baltimore, MD) using the HumanOmniExpressExome-8v1–2 array as described elsewhere (27). All SNPs met criteria for quality control for genotyping call rate (above 95%). Candidate SNPs selected for our study were those previously associated with sRAGE levels. Most of these SNPs appear to be located within the *AGER* gene, with rs2070600 being the most important and explaining 22% of the variability in sRAGE concentrations in Caucasians (17). In addition to *AGER*, three additional genes that contain SNPs associated with sRAGE were identified: *RNF5*, a neighboring gene which encodes for RAGE (28), *ADAM10*, which encodes for metalloproteinases involved in the shedding of RAGE ectodomain to form sRAGE (29), and *GLO1*, which encodes for glyoxalase enzyme responsible to metabolize methylglyoxal and prevent aberrant AGEs formation (30). The main SNPs were from *AGER* (rs2070600, rs1800625, rs1800624, rs184003, and rs2854050), *ADAM10* (rs653765), and *RNF5* (rs9469089; refs. 17–21, 31–38). We additionally considered less-studied SNPs located within *AGER* (rs1035798, rs1800684, rs3131300, rs3134940, rs2269422, rs2853807, rs9391855, and rs17846798), *ADAM10* (rs514049), *RNF5* (rs57409105, rs41268928, and rs17493811), and *GLO1* (rs4746, rs1130534, rs1049346, rs6932648, and rs10484854).

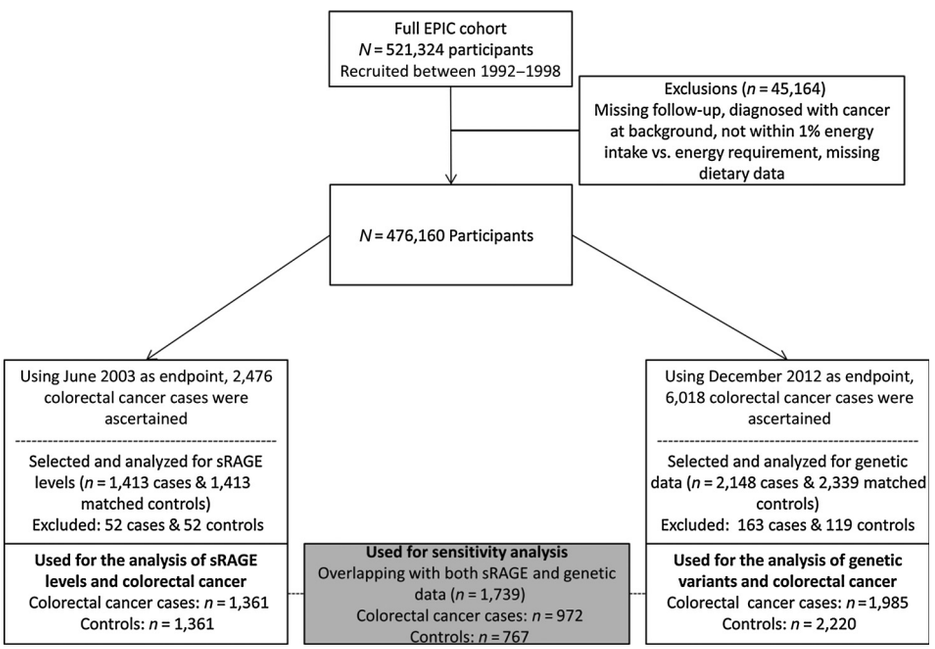

Figure 1.
sRAGE and genetic data available within EPIC. Two endpoints were used for our data, the first ended in June 2003 and included 1,361 colorectal cancer cases and 1,361 matched controls for the analysis of sRAGE concentrations. December 2012 was considered for the second endpoint, with 1,985 samples of colorectal cancer cases and 2,220 controls analyzed for genetic data. The overlapping between the two samples was used for sensitivity analysis.

The choice of this supplementary group of SNPs was based on the potential influence and interactions they may have in modulating sRAGE levels directly, or through AGEs (13, 17, 21, 31, 39–41).

Genotype distributions were in Hardy–Weinberg equilibrium (cutoff $P = 1 \times 10^{-3}$) for all the SNPs considered, with the exception of rs6932648, which was consequently excluded from the analysis. The selected SNPs and their characteristics are detailed in Supplementary Table S1. To select the independent variants, linkage disequilibrium (LD) pruning (LD ≤ 1%) was performed using NCI LDlink tools (https://ldlink.nci.nih.gov). We found the following independent variants (highly correlated variants are in brackets): rs2070600 (rs41268928, rs9391855, and rs2854050), rs1800625 (rs3131300 and rs3134940), rs1800624 (rs17846798), rs4746 (rs1130534 and rs10484854), rs17846798 (rs57409105), rs9469089, rs1800684, rs2269422, rs2853807, rs1049346, rs17493811, and rs653765 (rs514049). A flowchart outlining the selection of the independent SNPs is detailed in Supplementary Fig. S1.

Among the 767 control subjects who had both sRAGE and genetic data available, we assessed the association between the independent genetic variants and log-transformed sRAGE levels using linear regression models (Supplementary Table S2). The SNPs in the following genes were significantly associated with sRAGE levels: *AGER* (rs2070600 and rs1800625), *RNF5* (rs9469089), and *GLO1* (rs4746). Although rs653765 (*ADAM10*) was not associated with sRAGE levels, we decided to conserve it in our analysis for two main reasons: first, as a major variant of metalloproteinases, which are involved in the shedding of the ectodomain of RAGE to produce sRAGE; and second, this variant was previously associated with sRAGE levels in other populations (21). Overall, five SNPs (rs2070600, rs1800625, rs9469089, rs4746, and rs653765) were examined for the association with colorectal cancer risk.

Statistical analysis

Case–control differences in baseline characteristics were evaluated using Student paired *t* test and Wilcoxon signed-rank test for continuous variables and Kruskal–Wallis test for categorical variables. Spearman rank correlation was used to correlate sRAGE levels to anthropometry, dietary intakes, and other biomarkers. We divided sRAGE concentrations into quintiles based on the distribution in the control group. Conditional logistic regression was used to compute ORs and 95% confidence intervals (CI) for the associations between circulating levels of sRAGE and colorectal cancer risk. We ran two different models by including for each successive model additional adjustment variables incrementally. Model 1 (crude) was conditioned on the matching factors. Model 2 was additionally adjusted for body mass index (BMI), height, education (none, primary, technical and professional, secondary, and higher), physical activity (inactive, moderately inactive, moderately active, and active), smoking status, duration, and intensity (never; cigarettes/day 1–≤15, 16–≤25, and >26; and former smokers ≤10, 11–≤20, >20 years, and occasional), dietary energy, and intakes of alcohol, red and processed meat, dietary fiber, and dairy products. Dietary factors included as adjustment factors have been previously associated with colorectal cancer and/or sRAGE levels (42). *P* values for the linear trend (P_{trend}) were obtained by including the median value of each quintile as a continuous variable in the model. We also examined sRAGE levels as a continuous variable, per SD increment.

Stratified analyses were performed by anatomic subsites (colon vs. rectal cancers and proximal colon vs. distal colon cancers), sex (men vs. women), age groups (<50, ≥50–<55, ≥55–<60, ≥60–<65, and ≥65), smoking status (never, former, and ever), alcohol intake (tertiles), physical activity (inactive, moderately inactive, moderately active, and active), and BMI (<25, ≥25–<30, and ≥30 kg/m^2); and below or above sex-specific recommended cutoffs for waist circumference (WC; men, 94 cm and women, 80 cm) and waist-to-hip ratio (WHR; men, 0.90 and women, 0.85), and in women by menopausal status (pre-, post-, and perimenopause). The cutoffs for WC and WHR were based on the World Health Organization's definitions of central adiposity in European men and women (43). Additional stratified analyses were conducted for CRP (tertiles) as a marker of inflammation. $P_{heterogeneity}$ was calculated using the Wald test. For subgroup analyses by anthropometric measures, individual models were run for BMI, WC, and WHR in men and women separately (model 2 without BMI). In sensitivity analyses, we excluded cases diagnosed during the first 2 years of follow-up and rerun the analyses.

We assessed the association between the genetic variants and colorectal cancer risk using data of all participants genotyped in EPIC study to increase the statistical power of the analysis. The associations between the five independent genetic variants and colorectal cancer risk were assessed by unconditional logistic regression models. Two models were run, an unadjusted model and a multivariable adjusted model, adjusted for sex, age, BMI, smoking status, alcohol, and country. Additive (major allele = 0, heterozygous = 1, and minor allele = 2), dominant (major allele = 0 and heterozygous + minor allele = 1), and recessive models (major allele + heterozygous = 0 and minor allele = 1) were run for the genetic variants. In sensitivity analyses, we analyzed the participants with overlapping genetic and sRAGE concentrations data. All the statistical analyses were performed using Stata 14.0 (StataCorp). $P < 0.05$ was considered statistically significant.

Data sharing statement

For information on how to submit an application for gaining access to EPIC data and/or biospecimens, please follow the instructions at http://epic.iarc.fr/access/index.php.

Results

Baseline characteristics and sRAGE levels in cases and controls are presented in **Table 1**. Compared with controls, colorectal cancer cases had higher BMI, WC, WHR, and CRP concentrations, and consumed more alcohol and less dairy products and fruit and vegetables. sRAGE concentrations were slightly lower in colorectal cancer cases than controls (1,086 vs. 1,130 pg/mL), but this was mainly observed among men (982 vs. 1,066 pg/mL in male cases vs. controls, respectively); whereas among women, sRAGE was 1,185 pg/mL in cases and 1,191 pg/mL in controls. BMI, WC, WHR, and alcohol intake were all negatively correlated with sRAGE levels, whereas sugar and confectionaries, fruit and vegetable, and cereals intake showed positive correlations (Supplementary Table S3). Women with higher sRAGE levels had lower CRP concentrations (Spearman rho = −0.156; $P = 0.004$).

sRAGE and colorectal cancer risk

sRAGE concentrations were inversely associated with colorectal cancer risk in multivariable adjusted analyses (OR comparing the highest with the lowest quintile, $OR_{Q5vs.Q1}$, 0.75; 95% CI, 0.58–0.98; $P_{trend} = 0.035$; **Table 2**). Subgroup analyses by sex showed an inverse risk association for men ($OR_{Q5vs.Q1}$, 0.63; 95% CI, 0.42–0.94; $P_{trend} = 0.001$), but not in women ($OR_{Q5vs.Q1}$, 0.94; 95% CI, 0.63–1.38; $P_{trend} =$

Table 1. Selected baseline demographic and lifestyle characteristics of study participants by colorectal cancer status, EPIC study 1992–2012.

	Cases (n = 1,361)	Controls (n = 1,361)	P^a
Women, %	51.5	51.7	
Age, years, mean ± SD	58.4 ± 7.35	58.3 ± 7.38	0.877
Anthropometry, mean ± SD			
BMI, kg/m^2	26.7 ± 4.25	26.2 ± 3.74	0.004
WC, cm	90.4 ± 13.0	88.3 ± 12.1	<0.001
WHR	0.88 ± 0.10	0.87 ± 0.10	0.001
Lifestyle variables, *n* (%)			
Smoking status and intensity			
Never	514 (37.9)	542 (39.8)	0.703
Current, 1–≤15 cig/day	129 (9.51)	139 (10.2)	
Current, 16–≤25 cig/day	87 (6.40)	94 (6.91)	
Current, >26 cig/day	20 (1.47)	23 (1.69)	
Former, quit ≤10 years	139 (10.3)	129 (9.48)	
Former, quit 11–≤20 years	144 (10.6)	123 (9.04)	
Former, quit >20 years	166 (12.2)	177 (13.0)	
Current, pipe/cigar/occasional	125 (9.22)	102 (7.49)	
Physical activity			
Inactive	343 (25.4)	307 (22.6)	0.057
Moderately inactive	439 (32.4)	446 (32.3)	
Moderately active	307 (22.7)	282 (20.8)	
Active	264 (19.5)	321 (23.7)	
Highest education level attained			
None	68 (5.01)	66 (4.85)	0.275
Primary school completed	453 (33.4)	490 (36.0)	
Technical/professional school	324 (23.9)	343 (25.2)	
Secondary school	217 (16.0)	184 (13.5)	
Higher education	247 (18.2)	244 (17.9)	
Dietary intake, mean (SD)			
Energy, Kcal/day	2,124 ± 620	2,127 ± 609	0.764
Alcohol, g/day	17.0 ± 22.1	15.4 ± 19.7	0.040
Red and processed meats, g/day	87.6 ± 53.1	85.1 ± 52.0	0.215
Fruits and vegetables, g/day	396 ± 233	421 ± 248	0.007
Cereals, g/day	216 ± 121	216 ± 119	0.941
Dairy products, g/day	331 ± 251	351 ± 244	0.042
Fish, g/day	28.2 ± 28.8	29.6 ± 30.6	0.226
Sugar and confectionaries, g/day	48.7 ± 66.6	48.7 ± 68.9	0.995
Fat, g/day	28.3 ± 15.6	27.9 ± 16.0	0.536
Protein, g/day	89.3 ± 27.9	90.3 ± 27.5	0.337
Biomarkers			
CRP, ng/mL[b]	4,013 ± 6,011	3,433 ± 5,607	0.026
sRAGE levels, mean ± SD, pg/mL			
All participants	1,086 ± 469	1,130 ± 470	0.015
Men	982 ± 431	1,066 ± 438	<0.001
Women	1,185 ± 483	1,191 ± 490	0.831

Note: Frequencies may not add up to 100% due to missing data.
[a]Student paired *t* test and Wilcoxon signed-rank test for continuous variables and Kruskal–Wallis test for categorical variables.
[b]CRP was available for 1,103 cases and 925 controls.

0.754; $P_{heterogeneity}$ = 0.006). In men, sRAGE concentration was associated with a lower risk of both colon cancer (OR per SD increment, OR, 0.84; 95% CI, 0.70–0.99) and rectal cancer (OR, 0.80; 95% CI, 0.64–0.99), with no heterogeneity across anatomic subsites ($P_{heterogeneity}$ = 0.607; **Table 3**). The magnitude of the inverse association appeared stronger for distal colon cancer (OR, 0.61; 95% CI, 0.44–0.84) compared with proximal cancer (OR, 0.94; 95% CI, 0.69–1.29), but no heterogeneity was observed ($P_{heterogeneity}$ = 0.671). In women, no association was found between sRAGE concentration and colon cancer (OR, 0.99; 95% CI, 0.85–1.15) or rectal cancer (OR, 1.06; 95% CI, 0.86–1.32). Stratified analyses by age groups, BMI categories, WC and WHR cutoffs, and smoking status showed no significant differences across strata (**Fig. 2**). Women in higher CRP tertiles tended to have higher colorectal cancer risk associated with sRAGE ($P_{heterogeneity}$ across = 0.011; **Fig. 2**).

Analyses of genetic variants

Table 4 presents the association of the genetic variants with colorectal cancer risk. While comparing minor allele versus major allele, rs1800625 (*AGER*, G vs. A, OR, 1.15; 95% CI, 1.02–1.29) was associated with an increased risk of colorectal cancer, whereas rs653765 (*ADAM10*, C vs. T, OR, 0.88; 95% CI, 0.80–0.97)

Table 2. ORs and 95% CIs for colorectal cancer risk associated with circulating sRAGE (quintiles and continuous), EPIC study 1992–2012.

	Quintiles of sRAGE (cutoff points, in pg/mL)[a]							
	Quintile 1 (<754)	Quintile 2 (754–<941)	Quintile 3 (941–<1,157)	Quintile 4 (1,157–<1,440)	Quintile 5 (≥1,440)	P_{trend}	Continuous, per SD	Continuous, per SD[b]
All participants								
Cases/controls	344/273	258/272	272/271	239/272	248/273		1,361/1,361	1,101/1,101
Model 1[c]	1.00 (Ref.)	0.74 (0.58–0.94)	0.77 (0.61–0.98)	0.64 (0.50–0.83)	0.69 (0.54–0.89)	0.002	0.90 (0.83–0.97)	0.91 (0.82–1.00)
Model 2[d]	1.00 (Ref.)	0.75 (0.60–0.96)	0.83 (0.65–1.07)	0.69 (0.53–0.90)	0.75 (0.58–0.98)	0.035	0.93 (0.85–1.01)	0.92 (0.83–1.02)
Men								
Cases/controls	222/156	146/138	121/140	85/124	83/99		657/657	521/521
Model 1[c,e]	1.00 (Ref.)	0.77 (0.56–1.05)	0.62 (0.46–0.87)	0.46 (0.32–0.65)	0.57 (0.39–0.82)	<0.001	0.81 (0.72–091)	0.77 (0.65–0.91)
Model 2[d,e]	1.00 (Ref.)	0.79 (0.57–1.09)	0.62 (0.44–0.87)	0.49 (0.33–0.72)	0.63 (0.42–0.94)	0.001	0.84 (0.74–0.96)	0.75 (0.63–0.90)
Women								
Cases/controls	122/117	115/134	151/131	152/148	164/174		704/704	580/580
Model 1[c,e]	1.00 (Ref.)	0.77 (0.53–1.12)	1.04 (0.73–1.50)	0.93 (0.65–1.35)	0.90 (0.63–1.35)	0.967	0.99 (0.88–1.10)	1.00 (0.88–1.13)
Model 2[d,e]	1.00 (Ref.)	0.77 (0.52–1.15)	1.16 (0.79–1.70)	1.03 (0.70–1.53)	0.94 (0.63–1.38)	0.754	1.00 (0.89–1.13)	1.02 (0.89–1.16)

[a]Quintiles (in pg/mL) were created on the basis of the distribution of sRAGE in the control group. All the models were run using conditional logistic regression.
[b]Analysis excluding cases that occurred within 2 years of follow-up.
[c]Model 1 was conditioned on the matching factors.
[d]Model 2 is model 1, further adjusted for BMI (continuous); height (continuous); education (none, primary, technical and professional, secondary, and higher education); physical activity (inactive, moderately inactive, moderately active, and active); smoking status, duration, and intensity (never, 1–≤15 cigarettes/day, 16–≤25 cigarettes/day, >26 cigarettes/day, former smokers who quit ≤10 years, former smokers who quit 11–≤20 years, former smokers who quit >20 years, current pipe-cigar, and occasional smokers); dietary energy (continuous); and intakes of alcohol, red and processed meat, dietary fiber, and dairy products (all as continuous variables).
[e]Heterogeneity by sex for sRAGE and colorectal cancer risk association was statistically significant for the two models ($P_{heterogeneity} = 0.005$, and 0.006 for models 1 and 2, respectively).

was associated with a lower colorectal cancer risk, in univariate models. After multivariate adjustments, the association remained statistically significant for rs653765 (*ADAM10*, C vs. T, OR, 0.90; 95% CI, 0.82–0.99), but not for rs1800625 (*AGER*, G vs. A, OR, 1.11; 95% CI, 0.99–1.25).

Sensitivity analysis

Exclusion of the cases that occurred within the first 2 years of follow-up did not change the associations between sRAGE concentrations and colorectal cancer (**Table 1**). The associations between SNPs and colorectal cancer in participants with

Table 3. ORs and 95% CIs for risk of colorectal cancer anatomic subsites associated with circulating sRAGE (continuous, per SD), EPIC study 1992–2012.

	Colon cancer			
	All colon	Proximal colon	Distal colon	Rectal cancer
All participants				
Cases/controls[a]	854/854	372/372	414/414	502/502
OR (95% CI)[b]	0.94 (0.84–1.04)	0.92 (0.77–1.10)	0.88 (0.75–1.03)	0.90 (0.78–1.05)
Men				
Cases/controls[a]	388/388	160/160	191/191	270/270
OR (95% CI)[b,c]	0.84 (0.70–0.99)	0.94 (0.69–1.29)	0.61 (0.44–0.84)	0.80 (0.64–0.99)
Women				
Cases/controls[a]	466/466	212/212	223/223	232/232
OR (95% CI)[b,c]	0.99 (0.85–1.15)	0.85 (0.64–1.13)	1.05 (0.83–1.31)	1.06 (0.86–1.32)

[a]Some colorectal cancers cases were not included in the analysis as they were overlapping (five were neither colon nor rectal tumors, 68 were neither proximal nor distal colon tumors).
[b]Conditional logistic regression models conditioned on matching factors and adjusted for BMI (continuous); height (continuous); education (none, primary, technical and professional, secondary, and higher education); physical activity (inactive, moderately inactive, moderately active, and active); smoking status, duration, and intensity (never, 1–≤15 cigarettes/day, 16–≤25 cigarettes/day, >26 cigarettes/day, former smokers who quit ≤10 years, former smokers who quit 11–≤20 years, former smokers who quit >20 years, current pipe-cigar, and occasional smokers); dietary energy (continuous); and intakes of alcohol, red and processed meat, dietary fiber, and dairy products (all as continuous variables).
[c]$P_{heterogeneity}$ values for colon cancer versus rectal cancer were 0.607, 0.091, and 0.291 for all the participants, men, and women, respectively. $P_{heterogeneity}$ values for proximal colon cancer versus distal colon cancer were 0.307, 0.671, and 0.870 for all the participants, men, and women, respectively. $P_{heterogeneity}$ values by sex were 0.042, 0.832, 0.004, and 0.063 for all colon cancer, proximal colon cancer, distal colon cancer, and rectal cancer, respectively.

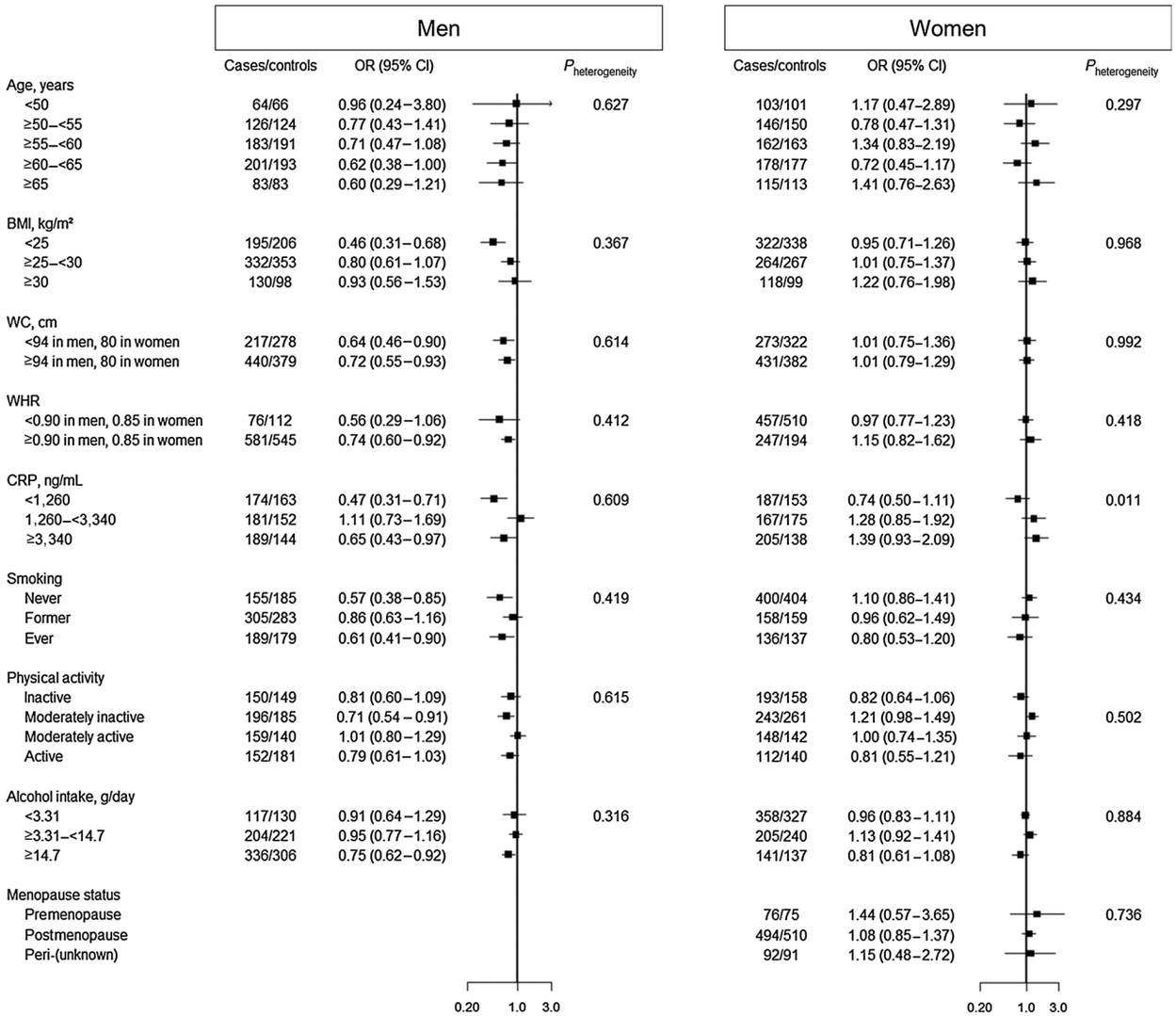

Figure 2.

Multivariable adjusted OR and 95% CI of the associations between RAGE level and colorectal cancer, stratified by lifestyle, obesity, CRP, and menopause status. Multivariable adjusted OR and 95% CI were computed for the stratified analysis. All the analyses were conditional logistic regression models conditioned on matching factors and adjusted for BMI, education, physical activity, smoking status, dietary energy, and intakes of alcohol, red and processed meat, dietary fiber, and dairy products. The analyses stratified by BMI, physical activity, smoking, and alcohol were not adjusted for their respective variables.

overlapping genetic and sRAGE data showed similar, but no statistically significant associations for rs653765 (*ADAM10*, OR, 0.90; 95% CI, 0.78–1.05) or rs1800625 (*AGER*, G vs. A, OR, 1.00; 95% CI, 0.83–1.19; Supplementary Table S4).

Discussion

In this large, case–control study nested within a European prospective cohort, we found that prediagnostic circulating sRAGE levels were inversely associated with colorectal cancer risk in men, but not in women. The associations observed between sRAGE concentration and colorectal cancer did not vary by age or by lifestyle factors, including obesity and smoking status, suggesting that sex is the main effect modifier in the association between sRAGE levels and colorectal cancer. With respect to the SNP analyses, we found that the minor allele of rs653765 (*ADAM10*) was inversely associated with risk of colorectal cancer, whereas an increased risk was suggested for rs1800625 (*AGER*). However, we did not observe the association between rs653765 and levels of sRAGE.

RAGE is a pattern recognition receptor that recognizes multiple ligands such as S100, high mobility group box 1 protein (HMGB1), and amyloid-β peptide, in addition to the AGEs (44). RAGE is overexpressed in several diseases of the colon, including inflammatory bowel diseases (45). RAGE action in colon tissues may participate in colorectal cancer tumor initiation, progression, and invasion (46–48). sRAGE, by acting as a decoy of RAGE, binds to AGEs in the circulation and clears them by decreasing interaction with full-length cell surface RAGE. The evidence from mouse studies shows that injection of

Table 4. ORs and 95% CIs for colorectal cancer risk associated with SNPs associated with sRAGE levels, EPIC study 1992–2012.

SNP	Cases	Controls	OR (95% CI)[a]	*P*[b]	OR (95% CI)[c]	*P*[b]
rs2070600 (*AGER*)						
CC	1,836	2,048	1.00 (ref.)		1.00 (ref.)	
CT	148	164	1.01 (0.80–1.27)	0.955	1.06 (0.84–1.35)	0.608
TT	1	8	0.14 (0.02–1.12)	0.063	0.17 (0.02–1.36)	0.095
T vs. C	1,985	2,220	0.93 (0.75–1.16)	0.519	0.99 (0.79–1.24)	0.906
CT+TT vs. CC	1,985	2,220	0.97 (0.77–1.21)	0.768	1.03 (0.81–1.30)	0.835
TT vs. CT+CC	1,985	2,220	0.14 (0.02–1.12)	0.063	0.17 (0.02–1.35)	0.094
rs1800625 (*AGER*)						
AA	1,350	1,584	1.00 (ref.)		1.00 (ref.)	
AG	574	578	1.17 (1.02–1.34)	0.028	1.13 (0.98–1.3)	0.084
GG	61	58	1.23 (0.86–1.78)	0.261	1.17 (0.81–1.7)	0.397
G vs. A	2,135	2,331	1.15 (1.02–1.29)	0.020	1.11 (0.99–1.25)	0.071
AG+GG vs. AA	2,135	2,331	1.17 (1.03–1.34)	0.019	1.13 (0.99–1.3)	0.067
GG vs. AG+AA	2,135	2,331	1.18 (0.82–1.7)	0.369	1.13 (0.78–1.64)	0.513
rs9469089 (*RNF5*)						
GG	1,408	1,619	1.00 (ref.)		1.00 (ref.)	
GC	532	548	1.12 (0.97–1.28)	0.121	1.14 (0.99–1.31)	0.070
CC	45	53	0.98 (0.65–1.46)	0.907	0.99 (0.65–1.49)	0.948
C vs. G	1,985	2,220	1.08 (0.95–1.21)	0.231	1.09 (0.97–1.23)	0.152
GC+CC vs. GG	1,985	2,220	1.10 (0.96–1.26)	0.150	1.13 (0.98–1.29)	0.089
CC vs. GC+GG	1,985	2,220	0.95 (0.63–1.42)	0.796	0.95 (0.63–1.43)	0.813
rs4746 (*GLO1*)						
TT	651	724	1.00 (ref.)		1.00 (ref.)	
TG	965	1,034	1.04 (0.90–1.19)	0.596	1.03 (0.9–1.19)	0.645
GG	369	462	0.89 (0.75–1.06)	0.179	0.89 (0.75–1.06)	0.192
G vs. T	1,985	2,220	0.95 (0.88–1.04)	0.275	0.95 (0.88–1.04)	0.282
TG+GG vs. TT	1,985	2,220	0.99 (0.87–1.13)	0.899	0.99 (0.87–1.13)	0.870
GG vs. TG+ TT	1,985	2,220	0.87 (0.75–1.01)	0.071	0.87 (0.75–1.02)	0.084
rs653765 (*ADAM10*)						
TT	1,076	1,125	1.00 (ref.)		1.00 (ref.)	
TC	757	887	0.89 (0.79–1.01)	0.081	0.90 (0.79–1.02)	0.098
CC	152	208	0.76 (0.61–0.96)	0.019	0.83 (0.66–1.04)	0.109
C vs. T	1,985	2,220	0.88 (0.80–0.97)	0.008	0.90 (0.82–0.99)	0.038
TC+CC vs. TT	1,985	2,220	0.87 (0.77–0.98)	0.022	0.88 (0.78–1.00)	0.051
CC vs. TC+TT	1,985	2,220	0.80 (0.64–1.00)	0.048	0.87 (0.70–1.09)	0.219

[a]Crude model (unadjusted).
[b]*P* values were calculated by considering genetic variant as continuous.
[c]Adjusted for sex, country, age (1-year categories), BMI (continuous), smoking status (never, former, and current), and alcohol intake (continuous).

sRAGE is associated with a reduction in the expression of inflammatory mediators, such as TNFα (49). Evidence from case–control studies also shows that elevated sRAGE levels are associated with a lower risk of several cancers, including liver (50) and pancreatic cancer (51). This suggests that higher concentrations of sRAGE are protective against AGE-induced inflammation, which is involved in the etiology of various chronic diseases, such as diabetes and cancers, but the mechanisms need further exploration.

The underlying reasons for the observed difference between men and women in the association between sRAGE concentration and colorectal cancer risk are unclear. Several previously published studies that compared sRAGE levels between men and women suggest higher circulating levels in women (14, 15, 17), which we also observed in our study. One explanation of the sex difference in sRAGE levels may be that estrogens stimulate sRAGE expression and production (52). Estrogens have also been reported to reduce AGEs production and AGE-related inflammation (53). In our study, women with higher sRAGE levels had lower CRP concentrations (Spearman rho = −0.156; $P = 0.004$) and lower colorectal cancer risk, suggesting that sRAGE may possibly reduce colorectal cancer risk in women, by mitigating overall inflammation. However, analysis by menopausal status showed no differences across strata in our study population. Our findings suggest that additional studies are needed to understand the physiologic sex differences in sRAGE levels and how they may translate into the differential colorectal cancer risk associations that we have observed in this study.

Interestingly, the two previous publications on sRAGE and colorectal cancer in prospective cohorts have been conducted in men (22) and in women (23) only. The Alpha-Tocopherol, Beta-Carotene Cancer Prevention study reported high serum sRAGE levels to be associated with low colorectal cancer risk in Finnish male smokers (22). We expanded this observation by showing that such an inverse association was also observed in male never smokers. We expected to observe a greater reduction in colorectal cancer risk in nonsmokers compared with smokers, but our findings did not differ by smoking status. Smoking may be a source of AGEs exposure (2), but the magnitude of the contribution of smoking to overall AGEs exposures remains to be explored. sRAGE levels have been reported to be higher, lower, or unchanged in smokers compared with nonsmokers (54–56). It is still unknown whether smoking could induce an adaptive

mechanism of sRAGE synthesis to cope with sustained formation of AGEs from glycotoxins contained in cigarettes. In a previous nested case–control study on a subsample of 1,249 postmenopausal women in the Women's Health Initiative study, higher sRAGE levels were observed to be associated with lower colorectal cancer risk in individuals with overweight and obesity, but not among normal weight postmenopausal women (23). Overall, our findings showed that sRAGE levels were associated with an inverse risk of colorectal cancer only in men, with no difference in magnitude across smoking status or any other lifestyle factor was observed.

We found that rs653765 located within *ADAM10* (C vs. T) was associated with lower risk for colorectal cancer. However, rs653765 (*ADAM10*) was not associated with sRAGE levels in our study, in contrast to previous studies in which the minor allele of rs653765 was associated with lower sRAGE levels (21). Another SNP, rs1800625, located in the promoter region of *AGER* is involved in the initiation of the production of the RAGE or its isomers (39). Xu and colleagues (57) reported in a meta-analysis of 18 case–control genetic studies that the recessive model of rs1800625 was associated with an increase of overall cancer risk while analyzing case–controls studies of 6,246 cases of renal, lung, breast, cervical, liver, oral, breast, and colorectal cancers. Although our findings with genetic variants are intriguing, they may be attributed to the diversity of functions associated with *AGER* and *ADAM10* genes. The production of sRAGE through the shedding of RAGE is dependent on ADAM10 levels. Thus, the overexpression of *AGER* coupled with lower ADAM10 activity will result in higher transmembrane RAGE and lower circulating sRAGE levels. This suggests that the interactions between AGER and ADAM10 may provide a better understanding of the genetic implications of RAGE and sRAGE in colorectal cancer development. In addition, the associations observed with the genetic data could be explained by other functions of the SNPs examined, particularly in the case of ADAM10 when considering its multiple actions, such as the formation of amyloid inclusions and the cleavage of a range of proteins (58). We did not observe a significant association between rs2070600 (*AGER*) and colorectal cancer, albeit our study showed that the major allele (C allele) of this SNP associates with higher sRAGE levels. A meta-analysis of 15 case–control studies showed that homozygous minor allele of this SNP was associated with an increased risk of all cancers (59). The absence of association of this SNP with colorectal cancer may be due to low statistical power, particularly as carriers of the minor allele are rare. Additional studies, using genetic data from larger research consortia, are needed to explore the link between the expression of *AGER, ADAM10*, and *RNF5* genes, and levels of sRAGE and colorectal cancer initiation and development.

The strengths of our study include the large number of cases and controls, the prospective design, and the availability of dietary and lifestyle factors and genetic variants. Our study was, however, limited by the fact that we did not differentiate between endogenous secretory RAGE (esRAGE) and proteolytically cleaved RAGE (cRAGE), the two components of sRAGE. esRAGE is formed by alternative splicing of RAGE mRNA and cRAGE is produced by the shedding of the ectodomain of RAGE by metalloproteinases located at the surface of the cells. esRAGE is stable throughout the life course, whereas cRAGE levels vary with age and environmental factors (60). Because we have measured the total pool of plasma sRAGE we, therefore, cannot discern whether the different variants of sRAGE have specific and potentially opposite associations with study outcomes. Although the variability of cRAGE makes it a poor biomarker for a prospective study, cRAGE levels data would have permitted us to explore the association between SNPs from the *ADAM10* gene, levels of cRAGE, and colorectal cancer risk. Our study was also limited by the fact that lifestyle factors and blood samples were collected at the recruitment, and may not necessarily reflect changes over years. Moreover, we cannot rule out residual confounding or unmeasured confounders, such as lifetime history of anti-inflammatory medication use.

In conclusion, we observed that prediagnostic circulating sRAGE levels were inversely associated with colorectal cancer risk in men, but not among women. We also found that the minor allele of rs653765 (*ADAM10*) was inversely associated with colorectal cancer risk. Additional studies are, however, required to further investigate how genetic variation and sex may affect sRAGE levels or modify its association with colorectal cancer risk.

Authors' Disclosures

No disclosures were reported.

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References

1. Gugliucci A. Formation of fructose-mediated advanced glycation end products and their roles in metabolic and inflammatory diseases. Adv Nutr 2017;8:54–62.
2. Cerami C, Founds H, Nicholl I, Mitsuhashi T, Giordano D, Vanpatten S, et al. Tobacco smoke is a source of toxic reactive glycation products. Proc Natl Acad Sci U S A 1997;94:13915–20.
3. Scheijen J, Clevers E, Engelen L, Dagnelie PC, Brouns F, Stehouwer CDA, et al. Analysis of advanced glycation endproducts in selected food items by ultra-performance liquid chromatography tandem mass spectrometry: presentation of a dietary AGE database. Food Chem 2016;190:1145–50.
4. Takeuchi M, Takino J-I, Furuno S, Shirai H, Kawakami M, Muramatsu M, et al. Assessment of the concentrations of various advanced glycation end-products in beverages and foods that are commonly consumed in Japan. PLoS One 2015;10:e0118652.
5. Rabbani N, Thornalley PJ. Advanced glycation end products in the pathogenesis of chronic kidney disease. Kidney Int 2018;93:803–13.
6. Semba RD, Nicklett EJ, Ferrucci L. Does accumulation of advanced glycation end products contribute to the aging phenotype? J Gerontol A Biol Sci Med Sci 2010;65:963–75.
7. Ott C, Jacobs K, Haucke E, Navarrete Santos A, Grune T, Simm A. Role of advanced glycation end products in cellular signaling. Redox Biol 2014;2:411–29.
8. Riehl A, Nemeth J, Angel P, Hess J. The receptor RAGE: bridging inflammation and cancer. Cell Commun Signal 2009;7:12.
9. Hsieh HL, Schafer BW, Sasaki N, Heizmann CW. Expression analysis of S100 proteins and RAGE in human tumors using tissue microarrays. Biochem Biophys Res Commun 2003;307:375–81.
10. Turner DP. Advanced glycation end-products: a biological consequence of lifestyle contributing to cancer disparity. Cancer Res 2015;75:1925–9.
11. Yamagishi S, Matsui T. Soluble form of a receptor for advanced glycation end products (sRAGE) as a biomarker. Front Biosci 2010;2:1184–95.
12. Ramasamy R, Yan SF, Schmidt AM. RAGE: therapeutic target and biomarker of the inflammatory response–the evidence mounts. J Leukoc Biol 2009;86:505–12.
13. Duan Z, Chen G, Chen L, Stolzenberg-Solomon R, Weinstein SJ, Mannisto S, et al. Determinants of concentrations of N(epsilon)-carboxymethyl-lysine and soluble receptor for advanced glycation end products and their associations with risk of pancreatic cancer. Int J Mol Epidemiol Genet 2014;5:152–63.
14. Prakash J, Pichchadze G, Trofimov S, Livshits G. Age and genetic determinants of variation of circulating levels of the receptor for advanced glycation end products (RAGE) in the general human population. Mech Ageing Dev 2015;145:18–25.
15. Norata GD, Garlaschelli K, Grigore L, Tibolla G, Raselli S, Redaelli L, et al. Circulating soluble receptor for advanced glycation end products is inversely associated with body mass index and waist/hip ratio in the general population. Nutr Metab Cardiovasc Dis 2009;19:129–34.
16. Moriya S, Yamazaki M, Murakami H, Maruyama K, Uchiyama S. Two soluble isoforms of receptors for advanced glycation end products (RAGE) in carotid atherosclerosis: the difference of soluble and endogenous secretory RAGE. J Stroke Cerebrovasc Dis 2014;23:2540–6.
17. Maruthur NM, Li M, Halushka MK, Astor BC, Pankow JS, Boerwinkle E, et al. Genetics of plasma soluble receptor for advanced glycation end-products and cardiovascular outcomes in a community-based population: results from the Atherosclerosis Risk in Communities study. PLoS One 2015;10:e0128452.
18. Salonen KM, Ryhanen SJ, Forbes JM, Harkonen T, Ilonen J, Laine AP, et al. Circulating concentrations of soluble receptor for AGE are associated with age and AGER gene polymorphisms in children with newly diagnosed type 1 diabetes. Diabetes Care 2014;37:1975–81.
19. Gaens KH, Ferreira I, van der Kallen CJ, van Greevenbroek MM, Blaak EE, Feskens EJ, et al. Association of polymorphism in the receptor for advanced glycation end products (RAGE) gene with circulating RAGE levels. J Clin Endocrinol Metab 2009;94:5174–80.
20. Lim SC, Dorajoo R, Zhang X, Wang L, Ang SF, Tan CSH, et al. Genetic variants in the receptor for advanced glycation end products (RAGE) gene were associated with circulating soluble RAGE level but not with renal function among Asians with type 2 diabetes: a genome-wide association study. Nephrol Dial Transplant 2017;32:1697–704.
21. Huang WH, Chen W, Jiang LY, Yang YX, Yao LF, Li KS. Influence of ADAM10 polymorphisms on plasma level of soluble receptor for advanced glycation end products and the association with Alzheimer's disease risk. Front Genet 2018;9:540.
22. Jiao L, Taylor PR, Weinstein SJ, Graubard BI, Virtamo J, Albanes D, et al. Advanced glycation end products, soluble receptor for advanced glycation end products, and risk of colorectal cancer. Cancer Epidemiol Biomarkers Prev 2011;20:1430–8.
23. Chen L, Duan Z, Tinker L, Sangi-Haghpeykar H, Strickler H, Ho GY, et al. A prospective study of soluble receptor for advanced glycation end-products and colorectal cancer risk in postmenopausal women. Cancer Epidemiol 2016;42:115–23.
24. Riboli E, Hunt KJ, Slimani N, Ferrari P, Norat T, Fahey M, et al. European Prospective Investigation into Cancer and Nutrition (EPIC): study populations and data collection. Public Health Nutr 2002;5:1113–24.
25. Wareham NJ, Jakes RW, Rennie KL, Mitchell J, Hennings S, Day NE. Validity and repeatability of the EPIC-Norfolk Physical Activity Questionnaire. Int J Epidemiol 2002;31:168–74.
26. Wu F, Afanasyeva Y, Zeleniuch-Jacquotte A, Zhang J, Schmidt AM, Chen Y. Temporal reliability of serum soluble and endogenous secretory receptors for advanced glycation end-products (sRAGE and esRAGE) in healthy women. Cancer Causes Control 2018;29:901–5.
27. Huyghe JR, Bien SA, Harrison TA, Kang HM, Chen S, Schmit SL, et al. Discovery of common and rare genetic risk variants for colorectal cancer. Nat Genet 2019;51:76–87.

28. Sessa L, Gatti E, Zeni F, Antonelli A, Catucci A, Koch M, et al. The receptor for advanced glycation end-products (RAGE) is only present in mammals, and belongs to a family of cell adhesion molecules (CAMs). PLoS One 2014;9:e86903.
29. Raucci A, Cugusi S, Antonelli A, Barabino SM, Monti L, Bierhaus A, et al. A soluble form of the receptor for advanced glycation endproducts (RAGE) is produced by proteolytic cleavage of the membrane-bound form by the sheddase a disintegrin and metalloprotease 10 (ADAM10). FASEB J 2008;22:3716–27.
30. Schalkwijk CG, Stehouwer CDA. Methylglyoxal, a highly reactive dicarbonyl compound, in diabetes, its vascular complications, and other age-related diseases. Physiol Rev 2020;100:407–61.
31. Chocholaty M, Jachymova M, Schmidt M, Havlova K, Krepelova A, Zima T, et al. Polymorphisms of the receptor for advanced glycation end-products and glyoxalase I in patients with renal cancer. Tumour Biol 2015;36:2121–6.
32. Krechler T, Jachymova M, Mestek O, Zak A, Zima T, Kalousova M. Soluble receptor for advanced glycation end-products (sRAGE) and polymorphisms of RAGE and glyoxalase I genes in patients with pancreas cancer. Clin Biochem 2010;43:882–6.
33. Li T, Qin W, Liu Y, Li S, Qin X, Liu Z. Effect of RAGE gene polymorphisms and circulating sRAGE levels on susceptibility to gastric cancer: a case-control study. Cancer Cell Int 2017;17:19.
34. Adams JN, Raffield LM, Martelle SE, Freedman BI, Langefeld CD, Carr JJ, et al. Genetic analysis of advanced glycation end products in the DHS MIND study. Gene 2016;584:173–9.
35. Daborg J, von Otter M, Sjolander A, Nilsson S, Minthon L, Gustafson DR, et al. Association of the RAGE G82S polymorphism with Alzheimer's disease. J Neural Transm 2010;117:861–7.
36. Yamaguchi K, Iwamoto H, Horimasu Y, Ohshimo S, Fujitaka K, Hamada H, et al. AGER gene polymorphisms and soluble receptor for advanced glycation end product in patients with idiopathic pulmonary fibrosis. Respirology 2017;22:965–71.
37. Miyashita M, Watanabe T, Ichikawa T, Toriumi K, Horiuchi Y, Kobori A, et al. The regulation of soluble receptor for AGEs contributes to carbonyl stress in schizophrenia. Biochem Biophys Res Commun 2016;479:447–52.
38. Peculis R, Konrade I, Skapare E, Fridmanis D, Nikitina-Zake L, Lejnieks A, et al. Identification of glyoxalase 1 polymorphisms associated with enzyme activity. Gene 2013;515:140–3.
39. Serveaux-Dancer M, Jabaudon M, Creveaux I, Belville C, Blondonnet R, Gross C, et al. Pathological implications of receptor for advanced glycation end-product (AGER) gene polymorphism. Dis Markers 2019;2019:2067353.
40. Jabaudon M, Berthelin P, Pranal T, Roszyk L, Godet T, Faure JS, et al. Receptor for advanced glycation end-products and ARDS prediction: a multicentre observational study. Sci Rep 2018;8:2603.
41. Kalousova M, Jachymova M, Germanova A, Kubena AA, Tesar V, Zima T. Genetic predisposition to advanced glycation end products toxicity is related to prognosis of chronic hemodialysis patients. Kidney Blood Press Res 2010;33:30–6.
42. Ivancovsky-Wajcman D, Zelber-Sagi S, Fliss Isakov N, Webb M, Zemel M, Shibolet O, et al. Serum soluble receptor for AGE (sRAGE) levels are associated with unhealthy lifestyle and nonalcoholic fatty liver disease. Clin Transl Gastroenterol 2019;10:1–10.
43. WHO. Waist circumference and waist–hip ratio: report of a WHO expert consultation. Geneva, Switzerland: World Health Organization; 2008; p. 39.
44. Leclerc E, Fritz G, Vetter SW, Heizmann CW. Binding of S100 proteins to RAGE: an update. Biochim Biophys Acta 2009;1793:993–1007.
45. Ciccocioppo R, Vanoli A, Klersy C, Imbesi V, Boccaccio V, Manca R, et al. Role of the advanced glycation end products receptor in Crohn's disease inflammation. World J Gastroenterol 2013;19:8269–81.
46. Kuniyasu H, Chihara Y, Kondo H. Differential effects between amphoterin and advanced glycation end products on colon cancer cells. Int J Cancer 2003;104:722–7.
47. Sakellariou S, Fragkou P, Levidou G, Gargalionis AN, Piperi C, Dalagiorgou G, et al. Clinical significance of AGE-RAGE axis in colorectal cancer: associations with glyoxalase-I, adiponectin receptor expression and prognosis. BMC Cancer 2016;16:174.
48. Yilmaz Y, Yonal O, Eren F, Atug O, Hamzaoglu HO. Serum levels of soluble receptor for advanced glycation endproducts (sRAGE) are higher in ulcerative colitis and correlate with disease activity. J Crohns Colitis 2011;5:402–6.
49. Chen Y, Yan SS, Colgan J, Zhang HP, Luban J, Schmidt AM, et al. Blockade of late stages of autoimmune diabetes by inhibition of the receptor for advanced glycation end products. J Immunol 2004;173:1399–405.
50. Moy KA, Jiao L, Freedman ND, Weinstein SJ, Sinha R, Virtamo J, et al. Soluble receptor for advanced glycation end products and risk of liver cancer. Hepatology 2013;57:2338–45.
51. White DL, Hoogeveen RC, Chen L, Richardson P, Ravishankar M, Shah P, et al. A prospective study of soluble receptor for advanced glycation end products and adipokines in association with pancreatic cancer in postmenopausal women. Cancer Med 2018;7:2180–91.
52. Mukherjee TK, Reynolds PR, Hoidal JR. Differential effect of estrogen receptor alpha and beta agonists on the receptor for advanced glycation end product expression in human microvascular endothelial cells. Biochim Biophys Acta 2005;1745:300–9.
53. Lin J, Zuo G-Y, Xu Y, Xiong J, Zheng Z, Wang S, et al. Estrogen reduces advanced glycation end products induced HUVEC inflammation via NF-kappa B pathway. Lat Am J Pharm 2014;33:93–100.
54. Pouwels SD, Klont F, Kwiatkowski M, Wiersma VR, Faiz A, van den Berge M, et al. Reply to Biswas: acute and chronic effects of cigarette smoking on sRAGE. Am J Respir Crit Care Med 2019;199:806–7.
55. Pouwels SD, Klont F, Kwiatkowski M, Wiersma VR, Faiz A, van den Berge M, et al. Cigarette smoking acutely decreases serum levels of the chronic obstructive pulmonary disease biomarker sRAGE. Am J Respir Crit Care Med 2018;198:1456–8.
56. Biswas SK. Acute and chronic effects of cigarette smoking on sRAGE. Am J Respir Crit Care Med 2019;199:805.
57. Xu Y, Lu Z, Shen N, Wang X. Association of RAGE rs1800625 polymorphism and cancer risk: a meta-analysis of 18 case-control studies. Med Sci Monit 2019;25:7026–34.
58. Przemyslaw L, Boguslaw HA, Elzbieta S, Malgorzata SM. ADAM and ADAMTS family proteins and their role in the colorectal cancer etiopathogenesis. BMB Rep 2013;46:139–50.
59. Huang Q, Mi J, Wang X, Liu F, Wang D, Yan D, et al. Genetically lowered concentrations of circulating sRAGE might cause an increased risk of cancer: meta-analysis using Mendelian randomization. J Int Med Res 2016;44:179–91.
60. Rebholz CM, Astor BC, Grams ME, Halushka MK, Lazo M, Hoogeveen RC, et al. Association of plasma levels of soluble receptor for advanced glycation end products and risk of kidney disease: the Atherosclerosis Risk in Communities study. Nephrol Dial Transplant 2015;30:77–83.