JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

pubs.acs.org/JAFC

Additive Capacity of [6]-Shogaol and Epicatechin To Trap Methylglyoxal

Qiju Huang,[†,‡] Pei Wang,[‡] Yingdong Zhu,[‡] Lishuang Lv,[*,†] and Shengmin Sang[*,‡]

[†]Department of Food Science and Technology, Nanjing Normal University, No. 122 Ninghai Road, Nanjing, 210097, P. R. China

[‡]Laboratory for Functional Foods and Human Health, Center for Excellence in Post-Harvest Technologies, North Carolina Agricultural and Technical State University, North Carolina Research Campus, 500 Laureate Way, Kannapolis, North Carolina 28081, United States

ABSTRACT: Methylglyoxal (MGO), a reactive dicarbonyl species, is thought to contribute to the development of long-term pathological diabetes as a direct toxin or as an active precursor of advanced glycation end products (AGEs). Trapping MGO by dietary phenols to inhibit the MGO induced AGE formation is an approach for alleviating diabetic complications. The present study investigated whether dietary compounds with different structures and active sites have the additive capacity to trap MGO. Ginger phenolic constituent [6]-shogaol and tea flavonoid (−)-epicatechin were selected and tested under simulated physiological conditions, showing that they additively trapped about 41% MGO at a concentration of 10 μM within 24 h. Furthermore, whether [6]-shogaol and epicatechin can retain their MGO trapping efficacy in vivo or a biotransformation limits their MGO trapping capacity remain virtually unknown. An acute mouse study was carried out by giving a single dose of [6]-shogaol, epicatechin, and the combination of both ([6]-shogaol + epicatechin) through oral gavage. A mono-MGO adduct of [6]-shogaol was identified from [6]-shogaol and [6]-shogaol + epicatechin treated mice, and mono- and di-MGO adducts of epicatechin and its metabolite, 3′-*O*-methyl epicatichin, were detected in urine samples collected from epicatechin and [6]-shogaol + epicatechin treated mice. To our knowledge, this is the first study demonstrating the additive MGO trapping efficacy of [6]-shogaol and epicatechin and that [6]-shogaol and epicatechin retained their MGO trapping capacity in mice.

KEYWORDS: *[6]-shogaol, epicatechin, methylglyoxal, trapping, additive effect*

INTRODUCTION

Increasing evidence indicates that the accumulation of advanced glycation end products (AGEs) are closely linked to the pathogenic pathways between hyperglycemia and diabetes related complications.[1−3] AGEs are a complex and heterogeneous group of compounds derived from nonenzymatic glycation between reducing sugars and amino residues of proteins, lipids, and nucleic acids.[4,5] Among the precursors of AGEs, methylglyoxal (MGO) is regarded as a major and highly reactive α,β-dicarbonyl intermediate in the glycation process, contributing to the formation of both intracellular and extracellular MGO-derived AGEs, such as argpyrimidine, N^{ϵ}-(carboxyethyl)-lysine, MGO lysine dimer, and MGO-derived hydroimidazolone.[6−9]

Scavenging MGO and preventing AGE formation have been investigated as therapeutic strategies to attenuate MGO-related diabetes mellitus.[10] Several pharmacological reagents, such as aminoguanidine,[11−13] pyridoxamine,[14−17] monascin,[18,19] and metformin,[20−24] have been reported to inhibit the formation of AGEs and prevent the deterioration of diabetic complications by scavenging reactive dicarbonyl species, mainly MGO. However, these pharmacological reagents can impose adverse side effect on diabetes patients, which makes it necessary to exploit effective and safe agents to prevent diabetic complications.

Discovery of MGO scavenging reagents from natural foods and beverages for reasons of safety seems feasible, and indeed, many dietary plants and their constituents have been described to have the capacity to alleviate MGO-induced glycation, mainly due to their MGO-scavenging and antioxidant properties. In our previous studies, a number of compounds, including soy isoflavones,[25,26] tea catechins and theaflavins,[27,28] apple polyphenols,[29] ginger gingerols and shogaols,[30] have been tested and determined to have significant MGO scavenging capacities in in vitro and/or in vivo models. Recently, our structure−activity relationship (SAR) study revealed that the A ring is the critical active site in flavonoids that contributes to their MGO trapping efficacy.[31] Furthermore, we investigated the potential additive effects of different flavonoids under simulated physiological conditions, in which we found that phloretin and quercetin can additively trap MGO through the same mechanism.[31]

In contrast to the flavonoids, the MGO trapping mechanisms of ginger phenolics were different. Trapping of MGO by ginger phenolics ([6]-shogaol and [6]-gingerol) involves a classic aldol condensation reaction because of the active protons of the α-carbonyl group in their side chains.[30] This fact naturally leads to the hypothesis that different types of components can additively trap MGO. The daily intake of individual dietary phenols may not be high enough to significantly decrease the levels of reactive MGO in humans. However, the total intake of phenols from a variety of foods can reach the effective doses to trap a significant amount of exogenous and endogenous

Received: June 24, 2017
Revised: August 30, 2017
Accepted: September 3, 2017
Published: September 3, 2017

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362

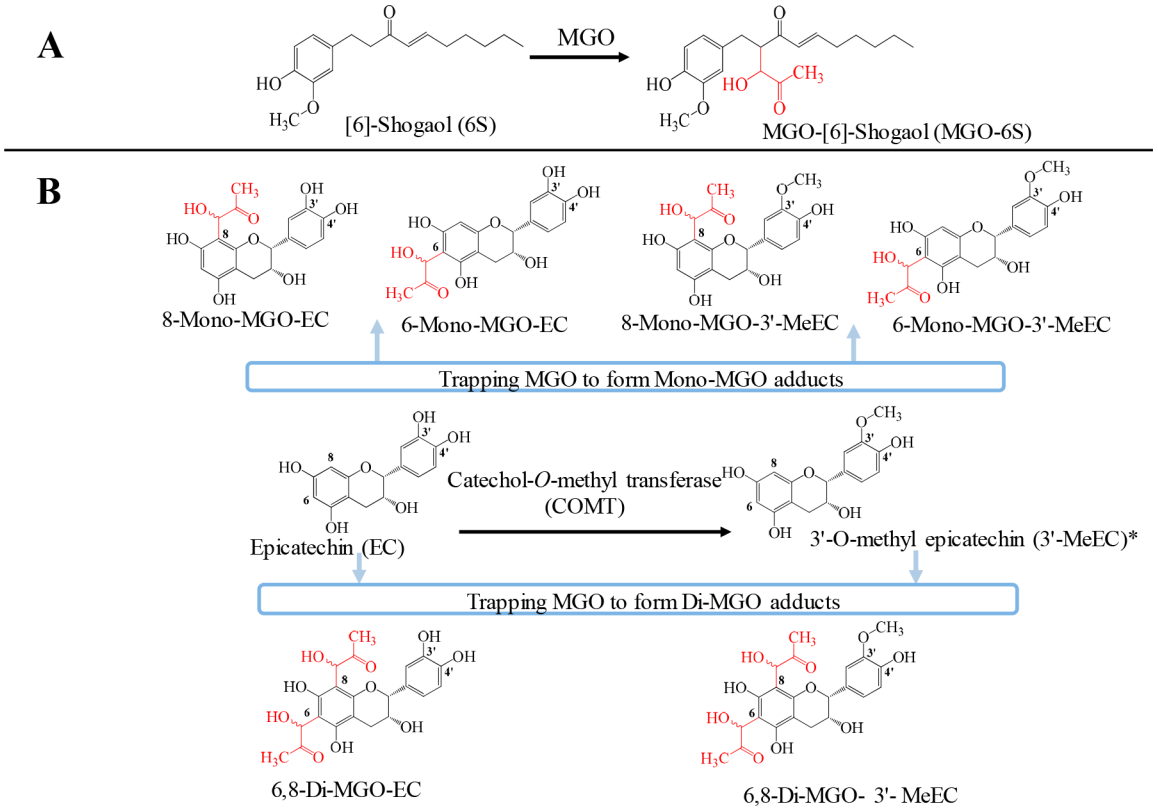

Figure 1. (A) Structures of [6]-shogaol (6S) and its MGO adduct, MGO-[6]-shogaol (MGO-6S); (B) structures of epicatechin (EC) and its methylated metabolites and their MGO adducts.

reactive dicarbonyls. In the present study, we investigated the additive effects of trapping MGO by ginger phenolic constituent [6]-shogaol and tea flavonoid (−)-epicatechin (Figure 1) under an in vitro system. Additionally, whether [6]-shogaol, epicatechin, and their major metabolites preserved their MGO trapping efficacy was also investigated in an in vivo model.

MATERIALS AND METHODS

Materials. [6]-Shogaol was purified from ginger root extract according to our previous published methods.[32] (−)-Epicatechin was purchased from Tokyo Food Techno Co., Ltd. (Shizuoka, Japan). MGO (40% in water), dimethyl sulfoxide (DMSO), and 1,2-diaminobenzene (OPD) were purchased from Sigma (St. Louis, MO). LC-MS grade solvents and other reagents were obtained from Thermo Fisher Scientific (Waltham, MA).

Trapping of MGO by [6]-Shogaol, Epicatechin, and the Combination of [6]-Shogaol and Epicatechin. MGO (60 μM) was incubated with [6]-shogaol (10 μM) and epicatechin (10 μM) or [6]-shogaol + epicatechin (10 μM each) in 100 mM potassium phosphate buffer (PBS), pH 7.4, and shaken for 4 and 24 h at 37 °C. Then, to the reacted mixture (500 μL) was added 2 μL of acetic acid to stop the reaction. The solution incubated for 24 h was used for derivatization of the remaining MGO by adding 50 mM OPD based on our previous method,[27] and the solution incubated for 4 h was used for LC-MS analysis.

Animal Studies. All mouse experiments conformed to a protocol approved by the Institutional Animal Care and Use Committee of the North Carolina Research Campus (no. 16-016). Female CF-1 mice were purchased from Charles River Laboratories, Inc. (Wilmington, MA) and acclimated for at least 1 week before being randomly allocated to different experimental groups. The mice were housed (5 mice/cage) and kept in air-conditioned quarters with a room temperature of 20 ± 2 °C, relative humidity of 50 ± 10%, and a light−dark cycle of 12:12 h (7 am to 7 pm). The mice were allowed free access to water and were fed an AIN-93G diet. Twenty mice were divided into a control group and three MGO and compound-treated groups (5 mice/group), including MGO and [6]-shogaol, MGO and epicatechin, and MGO and [6]-shogaol + epicatechin treated groups, which were kept in metabolic cages. For the MGO and compound-treated groups, each mouse received a single dose of [6]-shogaol, epicatechin, or a combination of [6]-shogaol (200 mg/kg body weight in DMSO) and epicatechin (200 mg/kg body weight in DMSO) via oral gavage. Ten min later, MGO solution (1.0 g/kg body weight in water) was administrated to the mice via oral gavage. The mice in the control group received DMSO followed by water. Twenty-four hours later, mouse urine and fecal samples were collected and stored at −80 °C before analysis.

Sample Preparation. For the preparation of the urine samples, enzymatic deconjugation of the samples was processed as described previously.[25,33] Briefly, 200 μL of urine sample from each group was treated with β-glucuronidase (250 U) and sulfatase (3 U) for 3 h at 37 °C and extracted three times with ethyl acetate. The ethyl acetate fractions were combined and dried under a gentle stream of nitrogen gas and then reconstituted with 200 μL of 80% aqueous methanol with 0.1% acetic acid for further analysis. For the preparation of the fecal samples, 100 mg of feces from each group was mixed with 1 mL of 80% aqueous methanol with 0.1% acetic acid; then samples were homogenized for 90 s by a Bead Ruptor Homogenizer (Omni International, Kennesaw, GA, USA) and then centrifuged at 14 000 rcf for 10 min. The supernatant (200 μL) was collected and diluted 10 times for analysis.

LC-MS Analysis. LC-MS analysis was performed using a Spectra system consisting of an Ultimate 3000 degasser, an Ultimate 3000 RS pump, an Ultimate 3000 RS autosampler, an Ultimate 3000 RS column compartment, and an LTQ Velos Pro ion trap mass spectrometer (Thermo Electron, San Jose, CA, USA) equipped with an electrospray ionization (ESI) interface. Chromatographic separation was performed using a 150 mm × 3.0 mm i.d., 5 μm, Gemini C18

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362

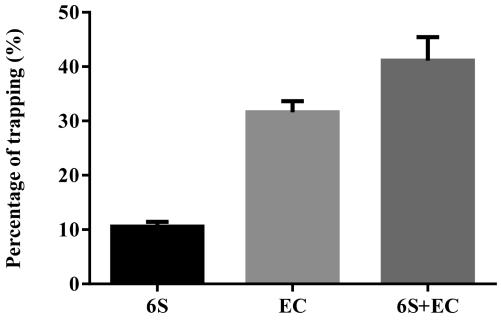

Figure 2. Trapping of MGO (60 μM) by [6]-shogaol (6S), epicatechin (EC), and the combination of 6S + EC (equal molar combination) at 10 μM in phosphate buffer (pH 7.4 and 37 °C) at 24 h. Data are presented as the means ± standard deviation (SD) of three replications.

column, (Phenomenex, Torrance, CA). The mobile phase consisted of 5% aqueous methanol with 0.1% formic acid (mobile phase A) and 95% aqueous methanol with 0.1% formic acid (mobile phase B). For the determination of the level of remaining MGO, the gradient elution was performed for 15 min at a flow rate of 0.2 mL/min using the following gradient: 0 to 2.5 min, 50−90% B, 2.5 to 6 min 90−100% B, and 6 to 12 min held at 100%, and then the column was re-equilibrated with 50% B for 3 min. The analysis of [6]-shogaol and its main MGO adduct was identical to our previous study.[30] For the analysis of epicatechin and its main MGO adducts, the gradient was initiated at 10% B and held constant for 5 min, followed by a linear increase to 55% from 5 to 25 min, to 100% from 25 to 30 min, and then held for 5 min. The column was then re-equilibrated with 10% B for 5 min; the flow rate was increased to 0.3 mL/min, and the injection volume was 10 μL. The negative ion polarity mode was set for an ESI ion source with the voltage on the ESI interface maintained at approximately 3.6 kV. Nitrogen gas was used as the sheath gas at a flow rate of 34 AU and the auxiliary gas at 10 AU. The collision-induced dissociation was conducted with an isolation width of 1.0 Da and a normalized collision energy of 35 for MS/MS analysis. Data were acquired with Xcalibur version 2.0 (Thermo Electron).

RESULTS AND DISCUSSION

Additive MGO-Trapping Effects by [6]-Shogaol and Epicatechin. Our previous investigations demonstrated that different kinds of natural products, such as flavonoids including (−)-epigallocatechin 3-gallate, genistein, phloretin, and quercetin, or [6]-shogaol and [6]-gingerol derived from ginger can effectively trap MGO under physiological conditions (pH 7.4 and 37 °C). Given that these natural products usually coexist in our diet like vegetables and fruits, it is necessary to evaluate the additive MGO-trapping effect of them. Our laboratory found that different flavonoids (phloretin and quercetin) could additively trap MGO, which was attributed to the A ring of flavonoids; MGO can react with the two unsubstituted carbons in the A ring to form mono- and di-MGO adducts.[34] However, there have been no studies reporting the additive effect of dietary compounds with different active sites in trapping MGO. Therefore, this study focused on the additive effect of [6]-shogaol and epicatechin. The formation of MGO adducts of [6]-shogaol is believed to involve a classic aldol condensation, due to the α-carbonyl group in its structure,[35] while

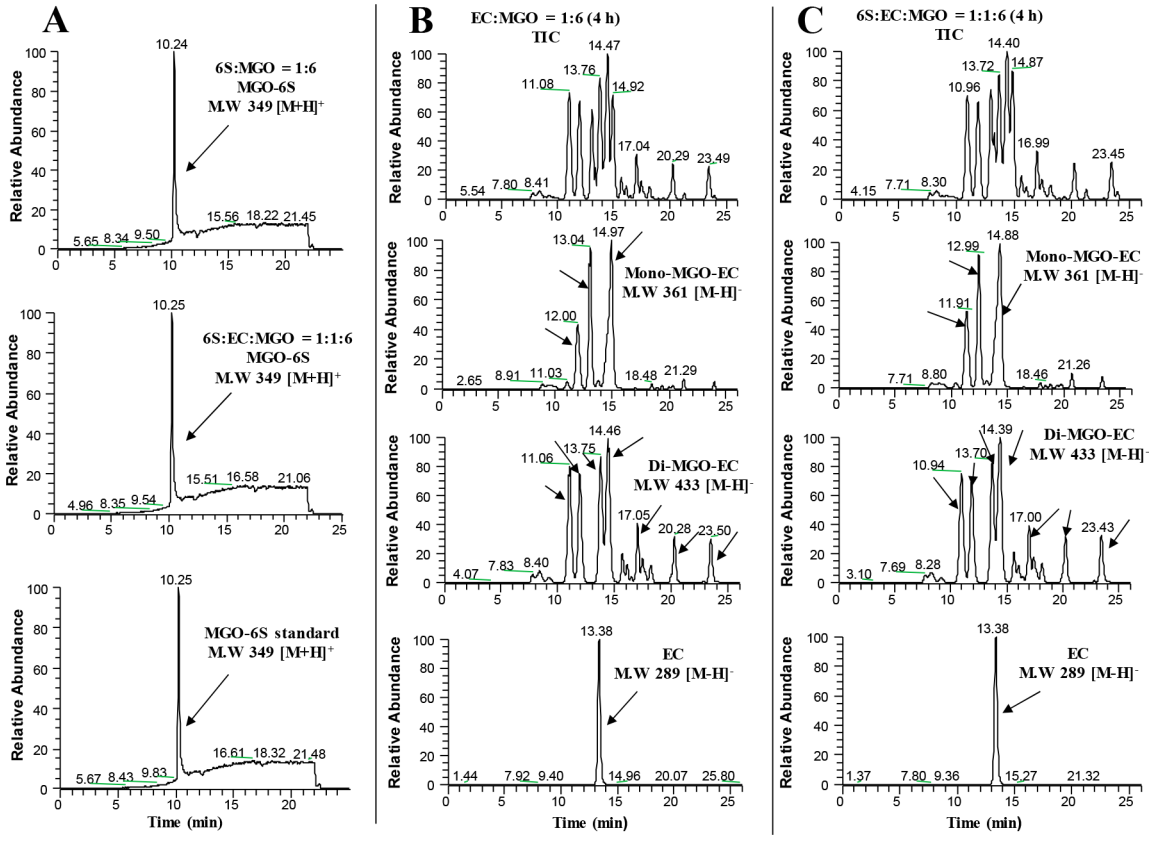

Figure 3. (A) SIM chromatograms of mono-MGO−[6]-shogaol (MGO-6S) adduct in [6]-shogaol−MGO (6S:MGO; 1:6) and [6]-shogaol−epicatechin−MGO (6S−EC−MGO; 1:1:6) incubation systems, as well as MGO-6S standard; (B) LC chromatograms of EC after incubation with MGO (1:6 molar ratio) for 4 h; (C) LC chromatograms of 6S and EC after incubation with MGO (1:1:6 molar ratio) for 4 h. The chromatograms of EC, mono, and di-MGO adducts of epicatechin (mono-MGO-EC and di-MGO-EC) were obtained using a SIM mode.

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362

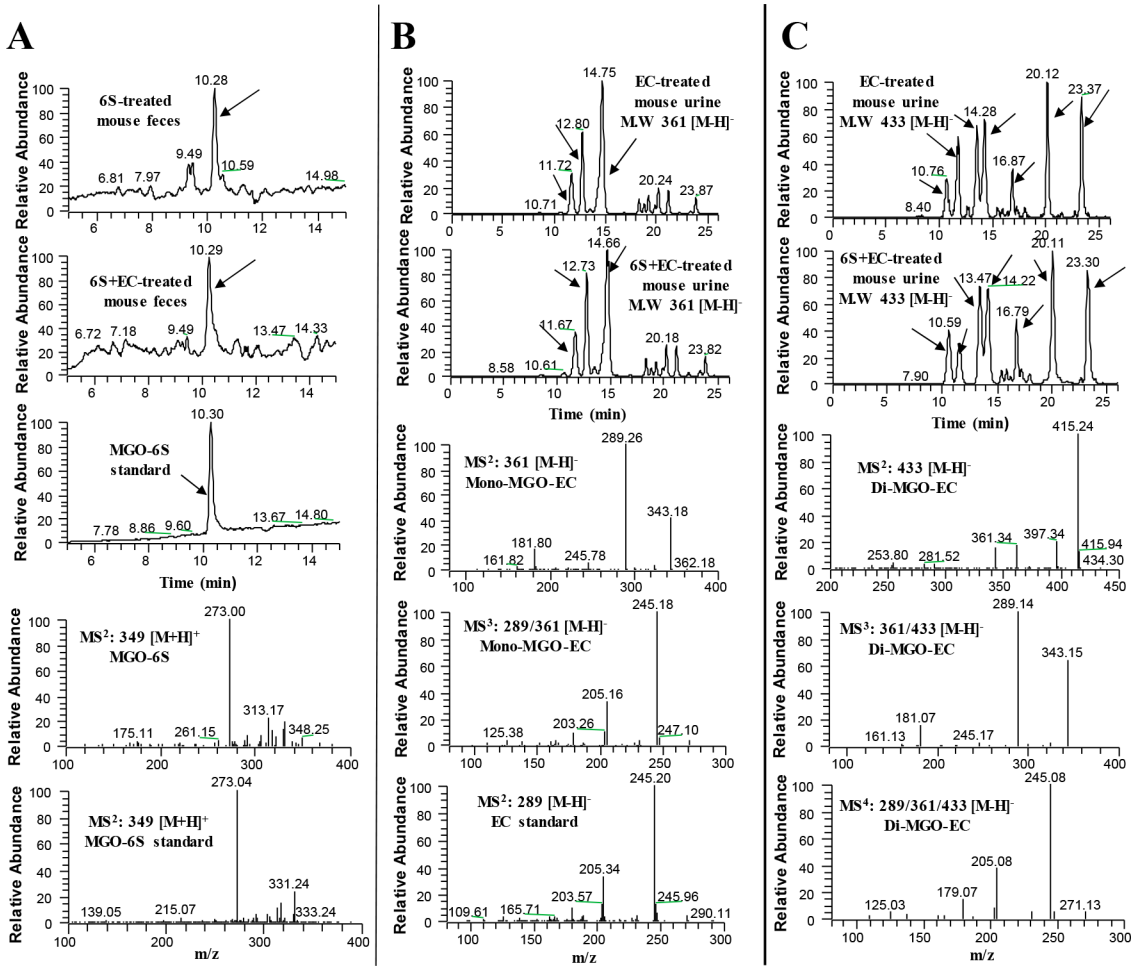

Figure 4. (A) SIM chromatograms and ESI-MS^n (n = 2; negative ion) spectra of mono-MGO−[6]-shogaol (MGO-6S) adduct in 6S and 6S + EC treated mouse feces, as well as MGO-6S standard; (B) SIM chromatogram and ESI-MS^n (n = 2−3; negative ion) spectra of mono-MGO−epicatechin (mono-MGO-EC) in EC and 6S + EC treated mouse urine; (C) SIM chromatogram and ESI-MS^n (n = 2−4; negative ion) spectra of di-MGO−epicatechin (di-MGO-EC) in EC and 6S + EC treated mouse urine.

epicatechin is a kind of flavonoid that C-6 and C-8 of its A ring can be conjugated with MGO.[31]

The MGO trapping capability of [6]-shogaol, epicatechin, and [6]-shogaol + epicatechin was investigated at a concentration of 10 μM with a ratio between each compound and MGO at 1:6 under pH 7.4 at 37 °C. Our results indicated that [6]-shogaol and epicatechin trapped 11 and 30% MGO, respectively, and 41% MGO was trapped by [6]-shogaol + epicatechin over 24 h (Figure 2). Our results indicated that ginger shogaol [6]-shogaol and tea flavonoid epicatechin, which had different MGO trapping mechanisms, can additively trap MGO.

Formation of MGO Adducts of [6]-Shogaol and Epicatechin in Physiological Conditions. To determine the underlying mechanism that [6]-shogaol and epicatechin additively inhibit MGO, the adducts formed in the [6]-shogaol, epicatechin, or [6]-shogaol + epicatechin treated reaction were investigated using LC-MS analysis in selective ion monitoring (SIM) mode (Figure 3). The reaction mixtures of [6]-shogaol and [6]-shogaol + epicatechin with MGO at different ratios (1:6 and 1:1:6, respectively) were collected after being incubated for 4 h. Figure 3A showed one major peak (t_R = 10.2 min, m/z 349 $[M + H]^+$) in both mixtures; the retention time and MS/MS spectrum of this peak were identical to that of the authentic [6]-shogaol−MGO that we synthesized and purified from the reaction mixture of [6]-shogaol and MGO,[30] demonstrating that the formation of [6]-shogaol−MGO (Figure 3A) in [6]-shogaol and epicatechin + [6]-shogaol treated mixtures underwent the same mechanism and formed the same adduct.

With the same ratios, the reaction mixtures of epicatechin and [6]-shogaol + epicatechin with MGO were also collected after 4 h of incubation. As indicated in Figure 3B and C, three major peaks (t_R = 12.0, 13.0, and 14.9 min) presented 72 Da (one molecule of MGO) higher than that of epicatechin (m/z 289 $[M - H]^-$), indicating that these peaks were mono MGO adducts of epicatechin (Figure 3B), while the peaks with the molecular ion at m/z 433 $[M - H]^-$ were 144 Da (two molecules of MGO) higher than that of epicatechin, which demonstrated that these peaks were di-MGO adducts of epicatechin (Figure 3B). In addition, the retention times and MS/MS spectrum of those peaks in epicatechin and [6]-shogaol + epicatechin treated mixtures were almost identical. All of the evidence above further verified that mono and di-MGO adducts of epicatechin were produced in both mixtures.

Identification of MGO Adducts of [6]-Shogaol and Epicatechin in Mice. The in vivo environment is quite different from in vitro, and many factors may influence this trapping reaction, such as pH, oxygen, pressure, and the presence of

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362

other endogenous and exogenous compounds. Thus, whether [6]-shogaol, epicatechin, or their combination could trap MGO in vivo remained unknown. To answer this question, we analyzed the formation of mono- and di-MGO adduct s of [6]-shogaol and epicatechin in urine and fecal samples collected from [6]-shogaol, epicatechin, and [6]-shogaol plus epicatechin treated mice under the same LC-MS conditions used in our in vitro studies.

In the SIM chromatogram of m/z 349 $[M + H]^+$ (molecular ion peak of the mono-MGO adduct of [6]-shogaol), consistent with our in vitro study, only one peak (t_R = 10.2 min) was detected in the mouse fecal samples collected from the acute study (Figure 4A). This peak shared the same retention time and MS/MS spectrum with those of the authentic mono-MGO-[6]-shogaol, which established that this peak is the mono-MGO adduct of [6]-shogaol. The mono- and di-MGO adducts of epicatechin were detected in the urine samples collected in the acute study (Figure 4B and C). Both in epicatechin and [6]-shogaol + epicatechin treated mouse urine, three peaks (t_R = 11.7, 12.8, and 14.7 min) were observed that corresponded to the mono-MGO adducts of epicatechin (molecular ion peak at m/z 361 $[M\text{-}H]^-$), all peaks were 72 Da higher than that of epicatechin (m/z 289 $[M - H]^-$), which indicated that they were mono-MGO adducts of epicatechin. This finding was further confirmed by the observation that these peaks included a fragment ion that lost one MGO unit (m/z 72), and the tandem mass spectrum of this fragment ion (m/z 289 (MS^3 289/361)) was almost identical to that of epicatechin (MS^2 289) (Figure 4B). When selecting m/z 433 as monitoring ion, seven peaks with the same molecular weight with different retention times were detected as shown in Figure 4B and C, those peaks also shared similar tandem mass spectra, including a fragment ion at m/z 361 (MS^3 361/433) which was identical to that of mono-MGO-epicatechin (MS^2 361) (Figure 4C), showing that these peaks were di-MGO adducts of epicatechin, which was further supported by a fragment ion [m/z 289 (MS^4 289/361/433)] lost two MGO units (m/z 144) and was nearly coincident to that of epicatechin (MS^2 289; Figure 4B and C).

Identification of the Metabolites of [6]-Shogaol and Epicatechin and Their MGO Adducts in Mice. Since both [6]-shogaol and epicatechin are extensively metabolized in mice, we searched the metabolites of [6]-shogaol and epicatechin and the MGO adducts of these metabolites in mouse urine samples. We did not find mono- and/or di-MGO adducts of [6]-shogaol metabolites in mouse urine, or at least their quantities were not sufficient enough to be detected under our LC-MS conditions. Therefore, whether the metabolites of [6]-shogaol can trap the MGO effectively in vivo requires future research.

Both in epicatechin and [6]-shogaol + epicatechin treated mouse urine samples, we observed two peaks (m/z 303 $[M - H]^-$) that were 14 mass units higher than that of epicatechin (m/z 289 $[M - H]^-$), which preliminarily indicated that they were methylated metabolites of epicatechin (Figure 5). Previous studies reported that the retention time of 3′-*O*-methyl epicatechin was prior to that of 4′-*O*-methyl epicatechin (with a 4:1 ratio) in the synthesis of 3′-*O*-methyl epicatechin using catechol-*O*-methyl transferase (COMT) (Figure 1B).[36] Therefore, we identified that the major peak (t_R = 19.2 min) as 3′-*O*-methyl epicatechin, while the minor peak (t_R = 20.5 min) was 4′-*O*-methyl epicatechin. Both peaks had similar tandem mass spectra of fragment ions.

It was noticeable that the methylation occurred on the B ring of epicatechin, and C-6 and C-8 of the A ring are the active sites to trap MGO, so the trapping efficacy of epicatechin

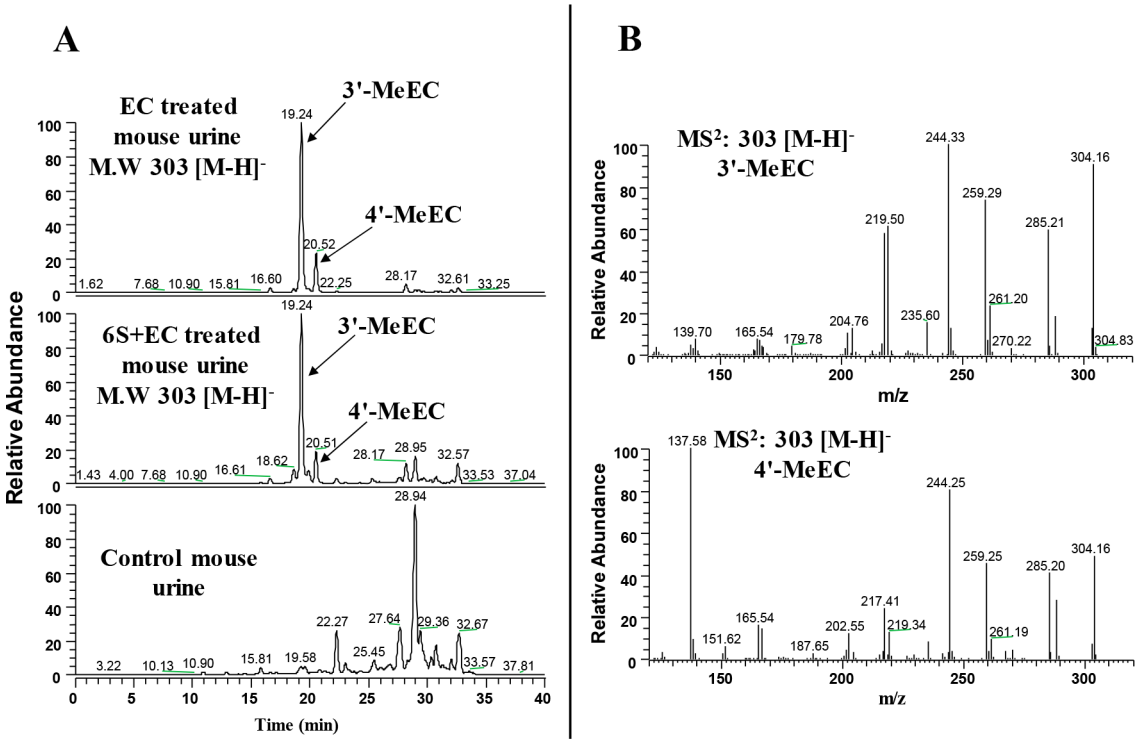

Figure 5. (A) SIM chromatogram of 3′-methyl epicatechin (3′-MeEC) and 4′-methyl epicatechin (4′-MeEC) in EC and 6S + EC, and vehicle treated mouse urine samples. (B) ESI-MS^n (n = 2; negative ion) spectra of 3′-methyl epicatechin (3′-MeEC) and 4′-methyl epicatechin (4′-MeEC).

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362

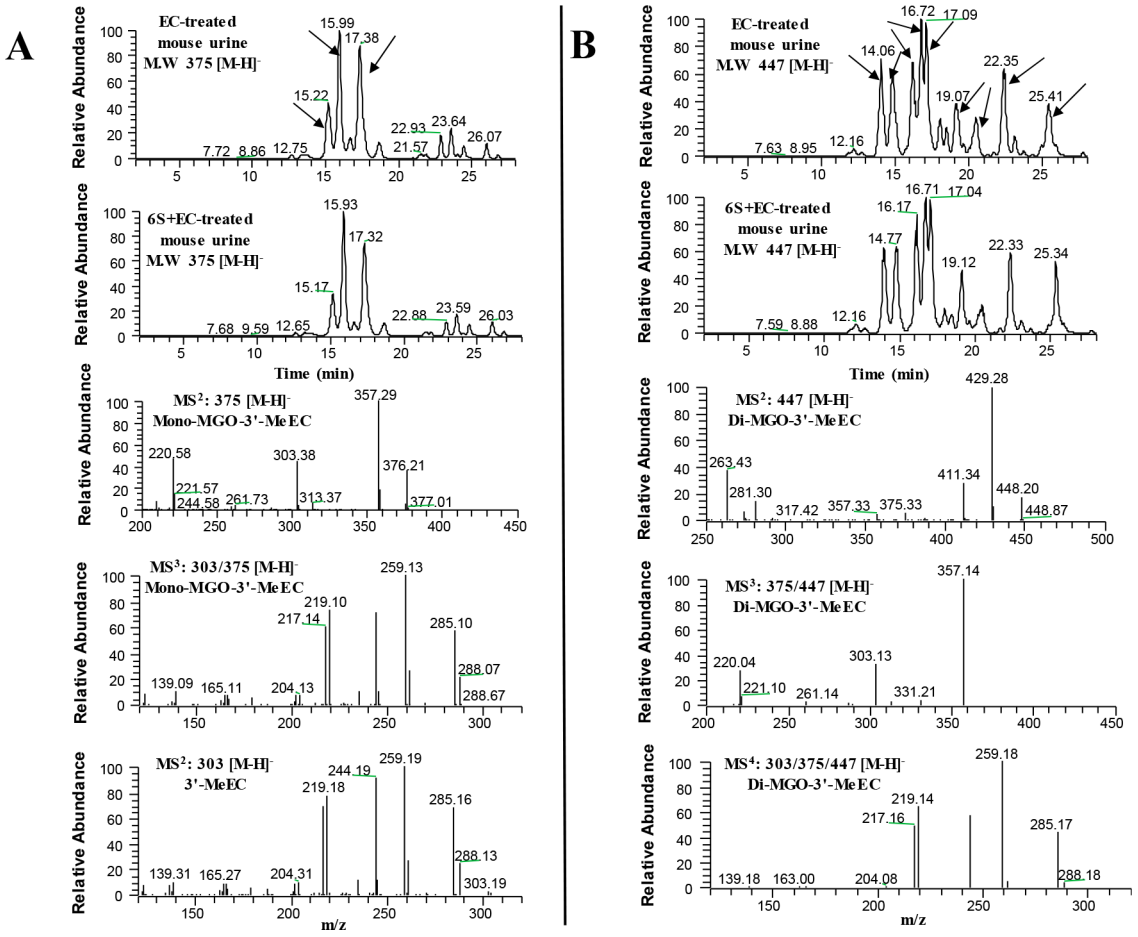

Figure 6. (A) SIM chromatogram and ESI-MS^n (n = 2−3; negative ion) spectra of mono-MGO-3′-methyl epicatechin (mono-MGO-3′-MeEC) in epicatechin (EC) and [6]-shogaol (6S) + epicatechin (EC) treated mouse urine; (B) SIM chromatogram and ESI-MS^n (n = 2−4; negative ion) spectra of di-MGO−3′-methyl epicatechin (di-MGO-3′-MeEC) in EC and 6S + EC treated mouse urine.

metabolites should be preserved. To investigate whether metabolites of epicatechin still have the capability of trapping MGO, epicatechin, and [6]-shogaol + epicatechin treated mouse urine samples were analyzed using LC-MS. In the SIM chromatogram of m/z 375 $[M - H]^-$ (molecular ion peak of the mono-MGO adduct of 3′-*O*-methyl epicatechin), three peaks (t_R = 15.2, 15.9, and 17.3 min) were observed with a fragment ion of m/z 303. The tandem mass spectrum with a fragment ion (MS^3 303/375) was almost identical to that of 3′-*O*-methyl epicatechin (Figure 6), which demonstrated that these three peaks were the mono-MGO adducts of 3′-*O*-methyl epicatechin.

According to a similar approach, di-MGO adducts of 3′-methyl epicatechin were also confirmed by observing the fragment ions that lost one MGO molecule [m/z 375 (MS^3 375/477)] and two MGO molecules [m/z 303 (MS^4 303/375/447)] which corresponded to those of mono-MGO-3′-methyl epicatechin and 3′-methyl epicatechin, respectively (Figure 6). Our results clearly demonstrated that the metabolites of epicatechin still possess the MGO-trapping efficacy.

In summary, this investigation revealed the additive MGO-trapping effect of dietary compounds with different structures, which provides meaningful suggestions about a healthy diet for people to prevent the development of diabetic complications. Additionally, this is the first report to illustrate that epicatechin and its metabolites are still competent to scavenge MGO in vivo; this discovery was consistent with our in vitro results and further confirmed our previous findings that the A ring of flavonoids is the active site mainly responsible for trapping reactive dicarbonyl species.

AUTHOR INFORMATION

Corresponding Authors

*Tel.: 704-250-5710; fax: 704-250-5729; e-mail: ssang@ncat.edu.
*Tel.: +86 25 83598286; fax: +86 25 83598901; e-mail: lishuanglv@126.com.

ORCID

Lishuang Lv: 0000-0003-3731-0136
Shengmin Sang: 0000-0002-5005-3616

Funding

The authors are grateful for financial support from the National Natural Science Foundation of China grant 31571783 to L. Lv. and USDA-NIFA grant 2012-67017-30175 to S. Sang.

Notes

The authors declare no competing financial interest.

REFERENCES

(1) Goh, S.-Y.; Cooper, M. E. The role of advanced glycation end products in progression and complications of diabetes. *J. Clin. Endocrinol. Metab.* **2008**, *93*, 1143−1152.

(2) Singh, V. P.; Bali, A.; Singh, N.; Jaggi, A. S. Advanced glycation end products and diabetic complications. *Korean J. Physiol. Pharmacol.* **2014**, *18*, 1−14.

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362

(3) Vlassara, H.; Striker, G. E. *Advanced glycation endproducts (AGEs) and chronic complications in diabetes* **2016**, 1.
(4) Singh, R.; Barden, A.; Mori, T.; Beilin, L. Advanced glycation end-products: a review. *Diabetologia* **2001**, *44*, 129−146.
(5) Negre-Salvayre, A.; Salvayre, R.; Augé, N.; Pamplona, R.; Portero-Otin, M. Hyperglycemia and glycation in diabetic complications. *Antioxid. Redox Signaling* **2009**, *11*, 3071−3109.
(6) Maessen, D. E.; Stehouwer, C. D.; Schalkwijk, C. G. The role of methylglyoxal and the glyoxalase system in diabetes and other age-related diseases. *Clin. Sci.* **2015**, *128*, 839−861.
(7) Allaman, I.; Bélanger, M.; Magistretti, P. J. Methylglyoxal, the dark side of glycolysis. *Front. Neurosci.* **2015**, *9*, 75.
(8) Angeloni, C.; Zambonin, L.; Hrelia, S. Role of methylglyoxal in Alzheimer's disease. *BioMed Res. Int.* **2014**, *2014*, 1.
(9) Chen, M.; Curtis, T.; Stitt, A. Advanced glycation end products and diabetic retinopathy. *Curr. Med. Chem.* **2013**, *20*, 3234−3240.
(10) Matafome, P.; Rodrigues, T.; Sena, C.; Seiça, R. Methylglyoxal in Metabolic Disorders: Facts, Myths, and Promises. *Med. Res. Rev.* **2017**, *37*, 368−403.
(11) Tóth, A. E.; Tóth, A.; Walter, F. R.; Kiss, L.; Veszelka, S.; Ózsvári, B.; Puskás, L. G.; Heimesaat, M. M.; Dohgu, S.; Kataoka, Y. Compounds blocking methylglyoxal-induced protein modification and brain endothelial injury. *Arch. Med. Res.* **2014**, *45*, 753−764.
(12) Thomas, M.; Baynes, J.; Thorpe, S.; Cooper, M. The role of AGEs and AGE inhibitors in diabetic cardiovascular disease. *Curr. Drug Targets* **2005**, *6*, 453−474.
(13) Tanaka, N.; Yonekura, H.; Yamagishi, S.-i.; Fujimori, H.; Yamamoto, Y.; Yamamoto, H. The receptor for advanced glycation end products is induced by the glycation products themselves and tumor necrosis factor-α through nuclear factor-κB, and by 17β-estradiol through Sp-1 in human vascular endothelial cells. *J. Biol. Chem.* **2000**, *275*, 25781−25790.
(14) Amarnath, V.; Amarnath, K.; Avance, J.; Stec, D. F.; Voziyan, P. 5′-*O*-alkylpyridoxamines: Lipophilic analogues of pyridoxamine are potent scavengers of 1, 2-dicarbonyls. *Chem. Res. Toxicol.* **2015**, *28*, 1469−1475.
(15) Almeida, F.; Santos-Silva, D.; Rodrigues, T.; Matafome, P.; Crisóstomo, J.; Sena, C.; Gonçalves, L.; Seiça, R. Pyridoxamine Reverts Methylglyoxal-induced Impairment of Survival Pathways During Heart Ischemia. *Cardiovasc. Ther.* **2013**, *31*, e79−e85.
(16) Nagaraj, R. H.; Sarkar, P.; Mally, A.; Biemel, K. M.; Lederer, M. O.; Padayatti, P. S. Effect of pyridoxamine on chemical modification of proteins by carbonyls in diabetic rats: characterization of a major product from the reaction of pyridoxamine and methylglyoxal. *Arch. Biochem. Biophys.* **2002**, *402*, 110−119.
(17) Voziyan, P. A.; Metz, T. O.; Baynes, J. W.; Hudson, B. G. A post-Amadori inhibitor pyridoxamine also inhibits chemical modification of proteins by scavenging carbonyl intermediates of carbohydrate and lipid degradation. *J. Biol. Chem.* **2002**, *277*, 3397−3403.
(18) Shi, Y.-C.; Liao, V. H.-C.; Pan, T.-M. Monascin from red mold dioscorea as a novel antidiabetic and antioxidative stress agent in rats and *Caenorhabditis elegans*. *Free Radical Biol. Med.* **2012**, *52*, 109−117.
(19) Hsu, W.-H.; Lee, B.-H.; Chang, Y.-Y.; Hsu, Y.-W.; Pan, T.-M. A novel natural Nrf2 activator with PPARγ-agonist (monascin) attenuates the toxicity of methylglyoxal and hyperglycemia. *Toxicol. Appl. Pharmacol.* **2013**, *272*, 842−851.
(20) Kinsky, O. R.; Hargraves, T. L.; Anumol, T.; Jacobsen, N. E.; Dai, J.; Snyder, S. A.; Monks, T. J.; Lau, S. S. Metformin scavenges methylglyoxal to form a novel imidazolinone metabolite in humans. *Chem. Res. Toxicol.* **2016**, *29*, 227−234.
(21) Kender, Z.; Fleming, T.; Kopf, S.; Torzsa, P.; Grolmusz, V.; Herzig, S.; Schleicher, E.; Racz, K.; Reismann, P.; Nawroth, P. Effect of metformin on methylglyoxal metabolism in patients with type 2 diabetes. *Exp. Clin. Endocrinol. Diabetes* **2014**, *122*, 316−319.
(22) Lu, J.; Ji, J.; Meng, H.; Wang, D.; Jiang, B.; Liu, L.; Randell, E.; Adeli, K.; Meng, Q. H. The protective effect and underlying mechanism of metformin on neointima formation in fructose-induced insulin resistant rats. *Cardiovasc. Diabetol.* **2013**, *12*, 58.
(23) Beisswenger, P.; Ruggiero-Lopez, D. Metformin inhibition of glycation processes. *Diabetes Metab.* **2003**, *29*, 6S95−6S103.
(24) Beisswenger, P. J.; Howell, S. K.; Touchette, A. D.; Lal, S.; Szwergold, B. S. Metformin reduces systemic methylglyoxal levels in type 2 diabetes. *Diabetes* **1999**, *48*, 198−202.
(25) Wang, P.; Chen, H.; Sang, S. Trapping Methylglyoxal by Genistein and Its Metabolites in Mice. *Chem. Res. Toxicol.* **2016**, *29*, 406−414.
(26) Lv, L.; Shao, X.; Chen, H.; Ho, C.-T.; Sang, S. Genistein inhibits advanced glycation end product formation by trapping methylglyoxal. *Chem. Res. Toxicol.* **2011**, *24*, 579−586.
(27) Sang, S.; Shao, X.; Bai, N.; Lo, C.-Y.; Yang, C. S.; Ho, C.-T. Tea polyphenol (−)-epigallocatechin-3-gallate: a new trapping agent of reactive dicarbonyl species. *Chem. Res. Toxicol.* **2007**, *20*, 1862−1870.
(28) Lo, C. Y.; Li, S.; Tan, D.; Pan, M. H.; Sang, S.; Ho, C. T. Trapping reactions of reactive carbonyl species with tea polyphenols in simulated physiological conditions. *Mol. Nutr. Food Res.* **2006**, *50*, 1118−1128.
(29) Shao, X.; Bai, N.; He, K.; Ho, C.-T.; Yang, C. S.; Sang, S. Apple polyphenols, phloretin and phloridzin: new trapping agents of reactive dicarbonyl species. *Chem. Res. Toxicol.* **2008**, *21*, 2042−2050.
(30) Zhu, Y.; Zhao, Y.; Wang, P.; Ahmedna, M.; Sang, S. Bioactive ginger constituents alleviate protein glycation by trapping methylglyoxal. *Chem. Res. Toxicol.* **2015**, *28*, 1842−1849.
(31) Totlani, V. M.; Peterson, D. G. Epicatechin carbonyl-trapping reactions in aqueous Maillard systems: identification and structural elucidation. *J. Agric. Food Chem.* **2006**, *54*, 7311−7318.
(32) Sang, S.; Hong, J.; Wu, H.; Liu, J.; Yang, C. S.; Pan, M. H.; Badmaev, V.; Ho, C. T. Increased growth inhibitory effects on human cancer cells and anti-inflammatory potency of shogaols from Zingiber officinale relative to gingerols. *J. Agric. Food Chem.* **2009**, *57*, 10645−50.
(33) Chen, H.; Lv, L.; Soroka, D.; Warin, R. F.; Parks, T. A.; Hu, Y.; Zhu, Y.; Chen, X.; Sang, S. Metabolism of [6]-shogaol in mice and in cancer cells. *Drug Metab. Dispos.* **2012**, *40*, 742−753.
(34) Shao, X.; Chen, H.; Zhu, Y.; Sedighi, R.; Ho, C.-T.; Sang, S. Essential structural requirements and additive effects for flavonoids to scavenge methylglyoxal. *J. Agric. Food Chem.* **2014**, *62*, 3202−3210.
(35) Zhu, Y.; Zhao, Y.; Wang, P.; Ahmedna, M.; Sang, S. Bioactive ginger constituents alleviate protein glycation by trapping methylglyoxal. *Chem. Res. Toxicol.* **2015**, *28*, 1842−9.
(36) Yang, C. S.; Maliakal, P.; Meng, X. Inhibition of Carcinogenesis by Tea. *Annu. Rev. Pharmacol. Toxicol.* **2002**, *42*, 25−54.

DOI: 10.1021/acs.jafc.7b02917
J. Agric. Food Chem. 2017, 65, 8356−8362