Supplemental Experimental

Cytotoxicity Assay

Cytotoxicity of LMW and HMW fractions of all foods samples was determined by measuring the release of LDH with the CytoTox 96 Assay (Promega) according to the manufacturer's instruction. LDH-release was calculated as percentage of LDH released in the culture media of total LDH inside and outside cells.

Legend to supplemental Figure

Supplemental Fig. 1

Cytotoxicity of HMW and LMW fractions from Japanese soy sauce, coffee, and red wine. After a 5 h preincubation in Dulbecco's modified Eagle's medium supplemented with 0.1% fetal bovine serum, rat C6 glioma cells that had been stably transformed with an expression plasmid containing human full-length RAGE cDNA and with a firefly luciferase reporter gene under the control of the NF-κB promoter were stimulated by AGE-BSA and food-derived fractions (A, 1.0 mg/mL; B, 0.5 mg/mL) for 24 h. After 24 h stimulation, the media and the lysates were assayed for the released and total LDH activity. AGE-BSA, 50 μg/mL glyceraldehyde-derived AGE-BSA; BSA, 50 μg/mL non-glycated BSA.

Supplemental Fig. 2

Effects of mixtures of HMW and LMW fractions from Japanese soy sauce, coffee, and red wine on RAGE signaling. RAGE signaling was assayed with human RAGE-expressing, NF-κB-promoter-luciferase reporter gene-carrying rat C6 glioma cells as described in the Experimental section. (A) Equal amounts (0.5 mg/mL each) HMW and LMW fractions from soy sauce, coffee

and red wine were combined and used for the assay. (B) Soy sauce-derived HMW and LMW fractions were combined at the indicated ratio and used for the assay. AGE-BSA, 50 μg/mL glyceraldehyde-derived AGE-BSA; BSA, 50 μg/mL non-glycated BSA. #, $p < 0.01$ (*vs.* BSA);**, $p < 0.01$ (*vs.* AGE-BSA) (n = 3).