Cranberry Phytochemicals Inhibit Glycation of Human Hemoglobin

and Serum Albumin by Scavenging Reactive Carbonyls

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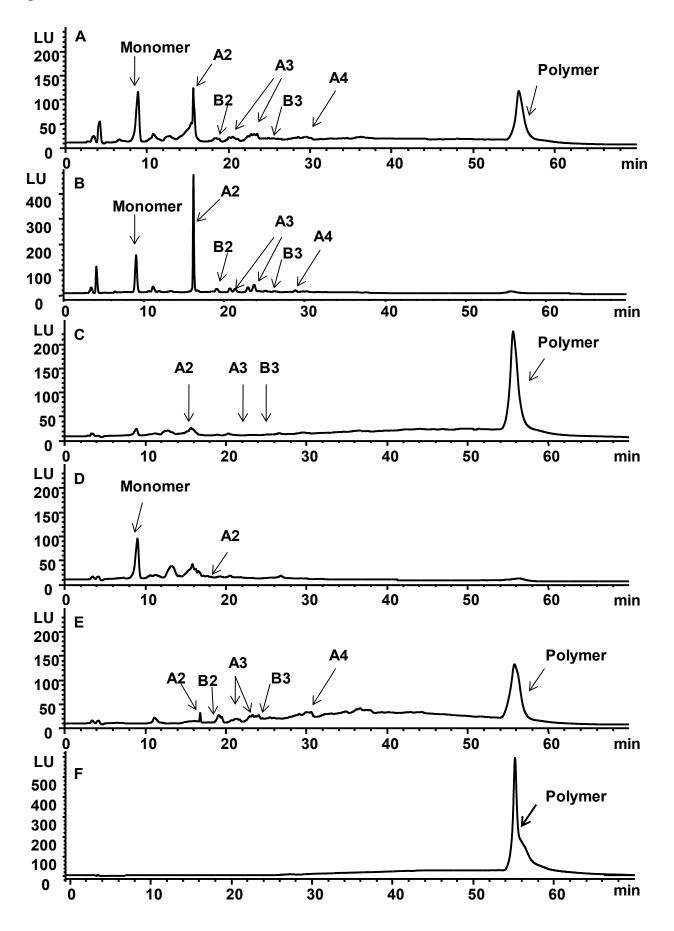
Supplemental data:

HPLC-MS analysis of cranberry phytochemical extract and fractions: Five miligrams of each cranberry fraction was dissolved in 1mL methanol. All the test solutions were centrifuged at 13 300 rpm for 5 minutes before injection. Twenty µL of cranberry phytochemical extract, water fraction, water fraction-I, II, and III, 10 µL of ethyl acetate fraction were injected for HPLC analysis (Agilent Technologies, Palo Alto, CA). Compound separation was carried on a Luna Silica column (250 × 4.6 mm, 5µm, Phenomenex, Torrance, CA). Mobile phases were composed of (A) methylene chloride/methanol/acetic acid/water (82:14:2:2, v/v/v) and (B) methanol/acetic acid/ water (96:2:2, v/v/v). The flow rate was set as 1 mL/min. The gradient for elution was: 0-20 min, 0.0-11.7% B linear; 20-50 min, 11.7-25.6% B linear; 50–55 min, 25.6–87.8% B linear; 55–65 min, 87.8% B isocratic; 65–70 min, 87.8–0.0% B linear; followed by 5 min of re-equilibration. Procyanidins were detected using fluorescent detection (Excitation 231 nm, Emission 320 nm). Electrospray mass spectrometry was performed with a HCT mass spectrometer (Bruker Daltonics, Billerica, MA) in the negative ion mode. The experimental conditions for the mass spectrometer were as follows: nebulizer, 50 psi; dry gas, 10.0 L/min; dry temperature, 350 °C; smart parameter setting (SPS), compound stability, 50%; trap drive level, 110%; ion trap, scan from m/z 150 to 2000. The most abundant ions in full scan mass spectra were isolated and its product ion spectra were recorded.

Supplementary Figure Legends

Figure S1. Chromatograms of procyanidins in cranberry phytochemical extract (A), ethyl acetate fraction (B), water fraction (C), water fraction I (D), II (E), and III (F) using fluorescence detection (excitation 231 nm and emission 320 nm). Identification was performed using HPLC-MS/MS ; B2 and B3 are B-type procyanidin dimers and trimers; A2,A3 and A4 are A-type procyanidin dimers, trimers and tetramers, respectively.

Figure S1



Supplemental Table S1: Correlation coefficients (r) between phytochemical contents and antiglycation activities of cranberry extracts and fractions.

	Total phenolic content	Total procyanidin content	Total anthocyanin content	HSA-MGO assay, EC50	HAS-Glucose assay, EC50	HbA1c levels
Total phenolic content	1.00	0.97	-0.66	-0.71	-0.72	-0.47
Total procyanidin content		1.00	-0.68	-0.81	-0.84	-0.54
Total anthocyanin content			1.00	0.82	0.75	0.35
HSA-MGO assay, EC ₅₀				1.00	0.99	0.26
HAS-Glucose assay, EC ₅₀					1.00	0.31
HbA1c levels (%)						1.00