Protein-bound *Vaccinium* fruit polyphenols decrease IgE binding to peanut allergens and RBL-2H3 mast cell degranulation *in vitro*

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Electronic Supplementary Information

RBL-2H3 mast cell degranulation upon co-exposure with plant extracts alone

The method used is described in sections 2.8 – 2.10 of the original research article. Polyphenol concentrations tested were the same as present in the peanut protein-cranberry or blueberry polyphenol complexes containing 5 - 40% polyphenols.
Fig. 1S. Histamine (A and B) and β-hexosaminidase (C and D) release following cranberry (A and C) or blueberry pomace extract (B and D) co-exposure with DNP-BSA in RBL-2H3 cells sensitized with anti-DNP IgE and ionomycin. Polyphenol concentrations tested were the same as present in the peanut protein-cranberry or blueberry polyphenol complexes containing 5 - 40% polyphenols. Controls (treatment 0 µg mL\(^{-1}\)) were cells which only received ionomycin and PBS 1x. Data shown are means of six replicates with SE; values with different letters are significantly different at p<0.05. DNP, dinitrophenyl; BSA, bovine serum albumin, PBS; phosphate buffered saline.
Measurement of cell viability using the XTT assay

RBL-2H3 cells in Eagle’s minimum essential medium (EMEM; Gibco, ThermoFisher Scientific, Waltham, MA, USA) and 15% FBS (fetal bovine serum) in EMEM (v/v) (Gibco, ThermoFisher Scientific, Waltham, MA, USA) were seeded at a concentration of $2 \times 10^5$ cells/well in a 24-well plate. The cells were incubated in a humidified incubator ($37 ^\circ C; 5\% CO_2$) for at least 24 h. The cells were serum starved in 1% FBS containing EMEM (v/v) and were treated with peanut protein-polyphenol complexes (containing 5-40% polyphenols) and incubated for 3 h in a humidified incubator ($37 ^\circ C; 5\% CO_2$). The effect of polyphenols alone on cell viability was also tested in a separate experiment. After incubation, the toxicity was assessed using XTT (2,3-Bis-(2-Methoxy-4-Nitro-5-Sulfophenyl)-2H-Tetrazolium-5-Carboxanilid) reagent as per manufacturer’s protocol (ATCC, Manassas, VA, USA). Two hundred µL of activated XTT solution was added to each well and subsequently incubated for an additional 2 h at $37 ^\circ C$ and 5% CO$_2$. The plate was then removed from the incubator and immediately measured for absorbance at 475 nm using a Synergy 2 microplate reader (BioTek, Winooski, VT, USA).
**Fig. 2S.** Cell viability (%) following cranberry (A) or blueberry (B) pomace extract co-exposure with DNP-BSA in RBL-2H3 cells sensitized with anti-DNP IgE and ionomycin. Polyphenol concentrations tested were the same as present in the peanut protein-cranberry or blueberry polyphenol complexes containing 5 - 40% polyphenols. Controls (treatment 0) were cells which only received ionomycin and PBS 1x. Cell viability (%) following peanut protein- cranberry (C) or blueberry (D) polyphenol complex (5-40% polyphenols) or unmodified peanut flour (PN) co-exposure with DNP-BSA in RBL-2H3 cells sensitized with anti-DNP IgE and ionomycin. Controls were cells which only received PBS 1x (blank). Data shown are means of six replicates with SE; values with different letters are significantly different at p<0.05. DNP, dinitrophenyl; BSA, bovine serum albumin, PBS; phosphate buffered saline.