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### **Dietary consumption of black raspberries or their anthocyanin constituents alters innate immune cell trafficking in esophageal cancer**

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Freeze-dried black raspberries (BRB), their component anthocyanins (ACs), and a metabolite of BRB ACs, protocatechuic acid (PCA), inhibit the development of esophagus cancer in rats induced by the carcinogen, N-nitrosomethyl benzyl amine (NMBA). All three components reduce inflammation in the esophagus and in plasma. The present study determined the relation of changes in inflammatory markers to infiltration of innate immune cells into NMBA-treated esophagus. Rats were injected with NMBA (0.35 mg/kg) for five weeks while on control diet. Following NMBA treatment, rats were fed diets containing 6.1% BRB powder, an AC-rich fraction of BRBs (3.8  $\mu$ moles/g diet), or 500 ppm PCA. At weeks 15, 25 and 35, inflammatory biomarker expression in the plasma and esophagus was quantified and infiltration of immune cells in the esophagus was examined. At all three time points, BRB, AC, and PCA similarly affected cytokine production in the esophagus and plasma of NMBA-treated rats, relative to the NMBA-only control. These included decreased expression of the pro-inflammatory cytokine IL1 $\beta$ , and increased the expression of the anti-inflammatory cytokine IL10. Moreover, all three diets also increased the expression of IL12, a cytokine that activates both cytolytic NK and CD8+ T cells. Additionally, the three diets also decreased infiltration of both macrophages and neutrophils into the esophagus. Overall, our results suggest that another mechanism by which BRBs, ACs, and PCA inhibit NMBA-induced esophageal tumorigenesis is by altering cytokine expression and innate immune cell-traffic into tumor tissues.

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