Proceedings of the British Pharmacological Society at http://www.pA2online.org/abstracts/Vol11Issue3abst152P.pdf

Effect of sulforaphane on cellular interactions with in the inflamed cerebral microvasculature

Scarlett Gillespie[1], Laura Gray[2], Felicity Gavins[1]. [1]Imperial College, London, UK, [2]Louisiana State University, London, USA

Introduction: Inflammation within the brain is known to underlie the driving pathology of several diseases of the CNS. It is known that during an inflammatory response there is an increase in leukocyte, platelet and endothelial interactions within the blood and at the vessel wall [1, 2]. Sulforaphane (SFN) is a phytochemical found in broccoli that is known to have anti-inflammatory effects on several cell types via induction of Nrf2[3]. Visualisation of SFN's effects of cellular interactions at the blood brain barrier has not been previously published.

Aim: Investigate the effect of SFN on cellular interactions in cerebral pial vessels following exposure to an endotoxaemic challenge (LPS; 0.5mg/kg).

Methods: Male, C57BL6 mice 21-30g were injected with lipopolysaccharide (*i.p.*, 0.5mg/kg 0111:B4, LPS, 4hrs) ± SFN (*i.p.*, 5-50mg/kg, at -24hr) followed by drilling of a cranial window and exposure of the parietal cortex. Cellular interactions in pial vessels were visualised via injection of Rhodamine 6G (100µl/0.02%) using fluorescent intravital microscopy. All experiments were conducted in accordance with appropriate Home Office licencing according to ASPA 1986.

Results: Following endotoxaemic challenge (*i.p.*, 0.5mg/kg LPS) mice showed little leukocyte-endothelial (L-E) interactions within the pial vessels 1 or 2hrs post challenged. However, at 4hrs post injection there was a significant increase in the number of cells that were adherent to the vessel wall when compared to mice undergoing saline treatment (728 *vs.*11 cells/min/mm^3 respectively). There was also a trend towards an increase in the number of cells rolling through the vessels (cells/min/mm^3), and a decrease in the rolling velocity (µm/sec) of these cells. These experiments were then repeated in the presence of SFN. Treatment with SFN at 5mg/kg for 24hr (666 cells/min/mm^3) was not seen to affect the L-E interactions within the vessels visualise; however, when the dose of SFN was increased to 50mg/kg the number of cells adherent within pial vessels was seen to significantly decrease compared to those receiving vehicle treatment (corn oil; 371 vs. 711 cells/min/mm^3).The rolling cell velocity of mice treated with SFN was not altered when compared to vehicle treated mice, however, the number of cells rolling through the pial vessels following SFN 50mg/kg/LPS 0.5mg/kg was significantly increased compared vehicle treated mice (335 vs. 127 cells/min/mm^3). (n=3/8, p<0.05 using ANOVA + Bonferroni's Multiple Comparison Test)

Conclusion: These results an increase in L-E interactions within the brain following a peripheral challenge with LPS. Furthermore, they make evident the ability of SFN to alter these inflammatory interactions in a dose dependent manner. This work highlights the potential for SFN to alter the cellular interactions present within vascular inflammatory interactions within the brain.

References:

Gillespie S, Gavins FN (2013). Phytochemicals: countering risk factors and pathological responses associated with ischaemia reperfusion injury. *Pharmacol Ther* **138**(1)**:** 38-45.

Ishikawa M, Cooper D, Arumugam TV, Zhang JH, Nanda A, Granger DN (2004). Platelet-leukocyte-endothelial cell interactions after middle cerebral artery occlusion and reperfusion. *J Cereb Blood Flow Metab* **24**(8)**:** 907-915.

Zarbock A, Polanowska-Grabowska RK, Ley K (2007). Platelet-neutrophil-interactions: linking hemostasis and inflammation. *Blood Rev* **21**(2)**:** 99-111.