Cyanidin-3-O-glucoside exhibits anti-inflammatory properties and improves intestinal epithelial barrier integrity in Caco-2 cells exposed to TNF-α

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The intestinal mucosal barrier plays an important role in the body's protection against luminal pathogens and antigenic molecules; intercellular tight junctions (TJs), mainly composed of cytoplasmic proteins (including zona occludens proteins) and two distinct transmembrane proteins (occludin and claudin), are the key structures responsible for intestinal epithelial barrier integrity (Turner, 2009). A dysregulation of one of this components, resulting in a paracellular permeability alteration, can lead to severe intestinal disorders, including inflammatory bowel diseases (IBDs), characterized by symptoms such as weight loss, diarrhoea, rectal bleeding, abdominal pain, fever and anemia (Cao *et al*, 2013). TNF-α-induced increase in intestinal epithelial TJ permeability has been proposed to be an important proinflammatory mechanism contributing to intestinal inflammation in IBD (Ye *et al*, 2006). Although the molecular mechanism involved in intestinal barrier dysfunction caused by proinflammatory cytokines is still unclear and it represents a research focus in pathogenesis of IBD, it has been believed that NF-kB plays a very important role in the proinflammatory cytokines-induced intestinal barrier disruption and in a downregulation of TJ proteins expression (Ma *et al*, 2004). Recent studies support beneficial effects of anthocyanins, a class of flavonoid compounds widely distributed in Mediterranean diet, in various chronic inflammatory diseases, such as IBDs.
The aim of this work was to investigate the protective effect exerted by Cyanidin-3-O-glucoside (C3G) pretreatment on TNF-α induced alteration in intestinal epithelial permeability and the intracellular mechanisms involved in it, by using an in vitro intestinal epithelial system consisting of filter grown Caco-2 monolayers. Caco-2 cells exposure to TNF-α for 6 h activated IKK/NF-kB proinflammatory pathway, and induced COX-2 and IL6 expression. Cells pretreatment for 24h with C3G (20 and 40 μM) was effective in preventing TNF-α-induced changes. Furthermore, C3G was able to improve intracellular redox status altered by TNF-α by increasing cellular GSH levels and antioxidant power. Finally, TNF-α exposure for 6h altered Caco-2 cells barrier permeability and integrity, and C3G pretreatment prevented the decrease in epithelial barrier integrity.
In conclusion, C3G showed anti-inflammatory properties, through the modulation of NF-kB pathway, and improved intestinal epithelial barrier integrity in Caco-2 cells challenged with TNF-α. These data suggest that anthocyanins could contribute, as complementary approaches to the conventional already existing therapeutic strategies (i.e. non-steroidal anti-inflammatory drugs), to the management of IBDs (Romier *et al*, 2008).

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