A biomimetic glyceride prodrug approach to promote drug delivery to the lymphatics, associated immune cells and tissues

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Background: The lymphatic system comprises a network of vessels, nodes and tissues distributed throughout the body that is surrounded by adipose tissue and has three main physiological functions: the preservation of fluid balance, dietary lipid absorption and regulation of immunity (Trevaskis, Kaminskas & Porter, 2015). In recent years there has been increasing studies demonstrating the significant role of the lymphatics, associated immune cells and adipose tissue, in modulating the pathogenesis of diseases such as cancer, inflammatory conditions, metabolic disease, and infections such as HIV (Trevaskis, Kaminskas & Porter, 2015). The current treatments for these diseases are limited in efficacy and are associated with life limiting off-target toxicities. Site-specific drug delivery to targets associated with the lymphatics has the potential to transform the treatment of these diseases by enhancing drug efficacy and facilitating the avoidance of off-target toxicities. The majority of orally administered small molecule drugs, however, access the lymphatics in limited quantities as they are absorbed and transported from the intestine *via* the blood circulation.

Aim: To determine the potential to enhance drug delivery to the lymphatics, associated immune cells and adipose tissue by linking the drug to a glyceride backbone to form a glyceride prodrug that integrates into the physiological process of dietary triglyceride absorption

Methods: The model immunosuppressant, mycophenolic acid (MPA) was esterified at the 2 position of a 1,3-diglyceride to form the prodrug 2-MPA-TG (1,3-dipalmitoyl-2-mycophenoloyl glycerol). All experiments were approved by the local Animal Ethics Committee and included full anaesthesia. 2-MPA-TG or MPA (as a control) were administered into the intestine of Sprague-Dawley rats or orally to greyhound dogs. Intestinal lymph fluid, lymph nodes and adipose tissues at different sites in the body were then collected at set time points. Lymphocytes were separated from the lymph *via* centrifugation. Sample concentrations of drug and prodrug were analysed *via* LC-MS/MS.

Results: The prodrug 2-MPA-TG efficiently incorporated into the absorption pathway for dietary triglyceride thereby markedly increasing intestinal lymph transport of total MPA related derivatives (90-fold in rats and 288-fold in dogs) and lymph lymphocyte concentrations of active MPA (19 fold in dogs) when compared to administration of the parent drug MPA. The extent of lymphatic transport of 2-MPA-TG was significantly greater in the dogs (with 36.4 % of the dose recovered in lymph) when compared to rats (13.4 % of the dose) (data from Han *et al.*, 2014). The prodrug 2-MPA-TG also facilitated a significant increase in the accumulation of active free MPA in local mesenteric lymph nodes in rats which appeared to occur due to direct entry of MPA into the mesenteric lymph nodes *via* the afferent lymph draining the small intestine. In support of this suggestion the accumulation of free MPA was greater and occurred at earlier time points in the superior mesenteric lymph node which receives lymph from the upper small intestine, when compared to the inferior mesenteric lymph nodes which receive lymph from lower intestinal regions, likely reflecting the pattern of triglyceride and prodrug absorption along the intestine. In contrast, 2-MPA-TG did not promote drug targeting to peripheral lymph nodes or any adipose tissue depot. The low and fluctuating accumulation of MPA at these sites reflected the plasma concentrations of MPA suggesting that MPA accessed these sites from the blood circulation.

Conclusion: Linking drugs to a glyceride backbone to form glyceride prodrugs can facilitate drug integration into the physiological process of dietary triglyceride absorption *via* the lymphatics. This increases drug exposure to the lymphatics, associated immune cells and tissues, thereby representing a promising strategy to enhance the treatment of diseases involving the lymphatics and associated tissues such as cancer, inflammatory conditions, metabolic disease, and infections such as HIV.

Trevaskis NL, Kaminskas LM, Porter CJH. (2015) *Nature Rev Drug Discov* In press.

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