MODULATION OF INNATE AND ACQUIRED IMMUNE RESPONSE BY LEUKOTRIENES AND PROSTAGLANDINS

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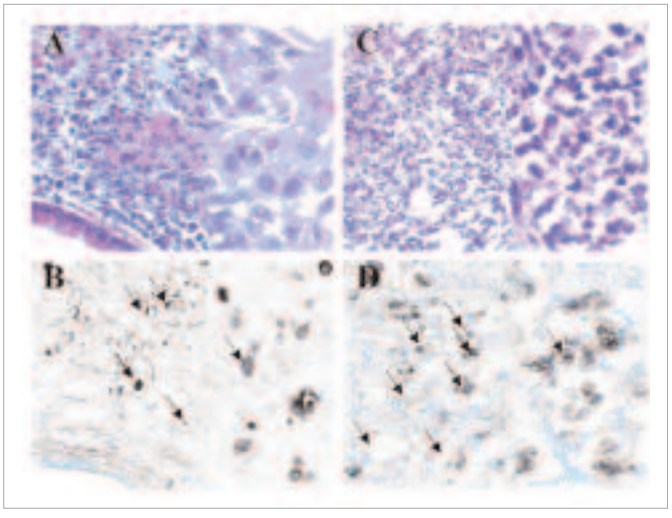
Several cells, in response to various stimuli, produce arachidonic acid (AA) metabolites. AA released can either be metabolized by cyclooxygenase enzymes 1 and 2 (COX1 and COX2) or by one of the lipoxygenases (LO), among them 5-lipoxygenase (5-LO), originating prostaglandins (PGs) and leukotrienes, respectively. PGs induce pain, vasodilatation, increase in vascular permeability, fever, and regulate the production of cytokines and the inflammatory process. On the other hand, some cytokines are able to regulate the production of PGs, and this magnifies the importance of studies involving these mediators and cytokines, which play a role in innate and acquired immune response. Leukotrienes induce leukocyte recruitment, plasma extravasation, mucus secretion, vascular relaxation, vasoconstriction and bronchoconstriction in different inflammatory processes. Moreover, leukotrienes play an important role in modulating the synthesis and release of inflammatory cytokines, immune response, also participating in cellular activation and/or production of antibodies. These lipid mediators also modulate microorganism phagocytosis by immune system cells. We have demonstrated that leukotrienes are involved in the systemic eosinophilia in response to infestation by *Toxocara canis*, but do not modulate expression of some adhesion; recruitment of leukocytes in response to intraperitonial inoculation of *Histoplasma capsulatum* (*Br. J. Pharmacol.*, 1999); and leukocyte recruitment in response to inoculation of venom from the wasp *Polybia paulista*. We have studied the participation of leukotrienes in lung infection by *H. capsulatum* and obtained novel and extremely relevant results. We observed that inhibition of the release of leukotrienes results in death of 100% of animals infected by *H. capsulatum* and this phenomenon seems to be related to the intense inflammatory reaction in the lungs and the accentuated proliferation of the fungus. We have also demonstrated that leukotrienes regulate the release of inflammatory cytokines such as IL-1, IL-6, KC and TNFa, and some chemokines, such as MCP-1, RANTES, MIP-1, MIP-2, IP-10, and MCP-1. Moreover, we have demonstrated that leukotrienes are essential for the release of cytokines in the immune response, such as IL-2, IL-12, and IFNy, as well as nitric oxide. In order to extend the observations on the role of AA metabolites as immunoregulators, we aim to investigate the participation of leukotrienes and/or prostaglandins (i) in the protection induced by an exoantigen from *H. capsulatum*; (ii) in infection by *Strongyloides venezuelensis*; (iii) in the release of cytokines and production of immunoglobulins in toxocariasis; (iv) in infection by *Mycobacterium tuberculosis*; (v) in oral tolerance. In addition, one of our goals is to investigate the use of these metabolites as adjuvants and immunomodulators in the treatment of various infectious diseases. There are no reports so far on experiments where leukotrienes and prostaglandins have been administered *in vivo* with these objectives. In this project we also aim to develop microspheres and nanoparticles containing leukotrienes and/or prostaglandins for *in vivo* administration.

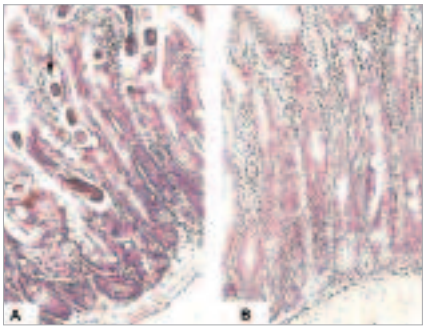


Increased leukocyte recruitment and fungal burden in the lung parenchyma resulting from endogenous leukotriene inhibition using MK886. Representative lung sections from mice infected with H. capsulatum *(Hc) and receiving p.o. water (A) or MK 886 daily. (C) H&E staining for leukocytes; (A and C) and GMS staining (B and D) for yeast cells (black arrow). Magnifications: ?360 (A and B) and ?900 (C and D). (Medeiros et al., 2004).*

SUMMARY OF RESULTS TO DATE AND PERSPECTIVES

Leukotrienes (LTs) comprise a family of lipid products of the 5-lipoxygenase (5-LO) pathway of arachidonic acid metabolism. The two principal bioactive classes of LTs are LTB4 and cysteinyl LTs (cysLTs) which are well known for their participation in inflammatory diseases and in protective immune responses in infection.

Using pharmacological tools (compound Mk886) to inhibit LT synthesis or mice with deficiency of 5-LO-/- enzyme (5-LO-/-), we have demonstrated that these lipid mediators are crucial to effective host defense against *Histoplasma capsulatum*, *Toxocara canis*, *Strongyloides venezuelensis* and *Mycobacterium tuberculosis*. Partial or total absence of LT had an important impact on cell recruitments and in cytokines, nitric oxide (NO) and antibodies productions, resulting in a defective immune response and in an increase of pathogen replication, and worsening the infections.



Histopathology of duodenum from S. venezuelensis*-infected 5-LO-/- (A) or Wild Type (B) mice. The arrows indicate sections of adult worms. The tissues were collected 14 days post infection and stained with H&E. Magnification of panels x100. (Machado et al, 2005)*

Based in ours results we have proposed the use of LTs as adjuvant or immunomodulators to control the immune responses in infectious diseases. We have developed a new LTB4-based pharmacological formulation in PLGA micro spheres (MS-LTB4) as a controlled release system to study the effects of LTs in in vitro and in vivo experimental models. We have demonstrated that MS-LTB4 formulations protects LTB4 from degradation and assure the delivery of the lipid mediator to the site of infection, inducing increased in cell recruitment, phagocytosis, and NO and cytokines productions. Moreover, MS-LTB4 injection protected animals against lethal infection with *H. capsulatum*, suggesting that the use of micro particulate system can be an advantageous tool to *in vivo* delivery of LTs.

With this project we intend to understand better the role of LTs in the immune response during infectious diseases and to develop controlled release formulations containing LTs to be used as therapeutic co-adjuvants for the treatment of infectious diseases.

MAIN PUBLICATIONS

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