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Oral administration of novel alginate enclosed, chitosan coated Fe-bLf loaded ceramic nanocarriers for parasitic infections: A potential alternative medicine

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Background: Lactoferrin (Lf), an iron binding ~80 kDa glycoprotein is a well characterized multifunctional protein found to be present in mammalian milk and in most exocrine secretions. Besides Lf's important physiological roles in the process of iron homeostasis, iron transportation and sequestration, it is well known for its properties such as anti-microbial, anti-viral anti-inflammatory and immunomodulatory functions.

Objectives: Main objective of the study was to develop, characterize and see the bio-distribution of iron saturated lactoferrin protein loaded novel ceramic nanocarriers to deliver orally in Giardia lamblia infected mice.

Results: In our study, we developed the nanoformulation of a novel alginate enclosed, chitosan coated Fe-bLf loaded ceramic nanocarriers (ACSC NCs). Uptakes of these NCs in vitro in human intestinal epithelial CaCo2 cells were analyzed, by measuring the endocytosis and transcytosis. The results show the NCs was having size range of 200nm with spherical morphology. SDS PAGE followed by western blotting, using specific antibodies against bLf confirms the structural integrity of the protein after the nano formulation. Confocal microscopy and flow cytometry qualitatively and quantitatively determines the internalization of rhodamine labeled NCs, upon treating them on to CaCo2 cells. In this study was carried out with the aim to investigate anti-parasitic activities of Fe bLf loaded ACSC NCs in cell based assays and in mice models of Giardia lamblia, a common parasite of children. Initially the experiments were carried out with native Australian bovine lactoferrin (bLf, ~15% saturated with iron). The efficacy of this protein was compared with other forms of Lf: Fe-Lf (100% saturated with iron), Apo-Lf (unsaturated with iron) using different concentrations in comparison to anti-parasitic drug, Metronidazole. The two forms of bovine lactoferrin (bLf)- the apo & native forms showed microbicidal effect on the parasites in vitro and killing was concentration dependent. Apo-bLf showed more inhibitory activity against trophozoites of G. lamblia than native bLf after 12 hrs of incubation with the drug. When the effectiveness of bLf was tested in comparison with metronidazole (40 mM), bLf was found to be more effective in killing the parasites. Fe-bLf loaded ACSC NCs significantly reduced parasitic load in Giardia lamblia infected Balb/c mice. Fe bLf increased the average weight of the spleens of Giardia lamblia infected mice by ~15%, accompanied by a major increase in the numbers of particular leukocyte subsets in the spleen. CD4+, CD8+, NK, IFN-γ+-expressing and dendritic cell numbers in the spleen were significantly ($P<0.001$) increased compared to corresponding cell numbers for mice maintained on the control diet. Fe-bLf loaded ACSC NCs bound to the intestinal epithelium and was preferentially taken up within Peyer's patches. It increased the production of Th1, Th2 and Th17 cytokines within the intestines, including TNF, IFN-γ, IL-18, nitric oxide as well as IL17. Importantly, it restored both red and white peripheral blood cell numbers depleted by anti-parasitic chemotherapy, potentially fortifying the mice against Giardia infections. In summary, Fe-bLf loaded ACSC NCs is a potent natural adjuvant and fortifying agent for augmenting anti-parasitic chemotherapy, but needs to be saturated with iron and administered orally in Fe-bLf loaded ACSC NCs to be effective. Bio-distribution of ACSC NCs was determined my MRI and confirmed by other imaging techniques. Taken together, our results are highly encouraging for the development of nano-therapeutic strategies for anti-parasitic infections.

Conclusions: Taken together, our results are highly encouraging for the development of nano-therapeutic strategies and drug delivery to provide more potent and targeted therapeutic, for gut infections. Fe-bLf loaded ACSC NCs were observed to be more effective as an anti-microbial agent.

Biography

Associate Professor Jagat Kanwar, Group Leader "Nanomedicine-Laboratory of Immunology and Molecular Biomedical Research" working in Deakin University, Australia. He did his PhD in 1993 from PGIMER, Chandigarh, India and worked as a Senior Scientist in The Auckland University, New Zealand for more than 10 years. He has a national and international reputation in investigating fundamental and applied molecular aspects of cancer, microbial infections and chronic inflammation. His research is also focused on miRNA, aptamer, locked nucleic acid (LNA) LNA-modified chimeric aptamers-siRNA conjugates, and immunoliposomes technology and disease targeted drug discovery. His research combines Immunology with state of the art and cutting edge techniques in Molecular Biology, Biochemistry, Nanobiotechnology and visualization to investigate the pathways in which key molecules are regulated in both normal and disease states. He designed nanocarriers for applications in vaccines, immunotherapy, and drug delivery of antigens immunostimulatory ligands to dendritic cells and subsequent stimuli to T- lymphocytes, B-lymphocytes and TH17 cells. His group provides high quality research training and education to undergraduate and postgraduate students which, in turn, strengthen Deakin's strategic research and academic priorities, helping students keep in pace with the emerging concepts of science and technology. His group carry out both academic and commercial research projects and develop new approaches for the diagnosis, treatment, and nanomedicine based new generation delivery systems for the prevention of human diseases like cancer, microbial infections, inflammatory bowel disease (IBD), neurodegenerative, osteoarthritis, cardiovascular and pulmonary diseases. Kanwar's research work generated in total of 12 patent/PCTs with two provisionals in preparation. Five of these patents have been licensed for commercialization to biotech companies Antisoma, NeuronZ, Neuren Pharmaceuticals and Fonterra.

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