OP-1-4

USING A CACO-2 AND THP-1 CO-CULTURE MODEL TO EVALUATE INTESTINAL BARRIER PROTECTIVE PROPERTIES — EFFECTS OF DEXAMETHASONE

https://doi.org/10.15605/jafes.037.AFES.04

Sylvia Riedel,[1,2] Lize Engelbrecht,[3] Pieter Venter,[1] Johan Louw,[1,4] Carmen Pheiffer,[1,2,5] Christo J.F. Muller[1,2,4]

[1]*Biomedical Research and Innovation Platform, South African Medical Research Council, Tygerberg, South Africa*

[2]*Centre for Cardio-metabolic Research in Africa, Division of Medical Physiology, Faculty of Medicine and Health Sciences, Stellenbosch University, Cape Town, South Africa*

[3]*Central Analytical Facilities, Microscopy Unit, Stellenbosch University, Matieland, South Africa*

[4]*Department of Biochemistry and Microbiology, University of Zululand, KwaDlangezwa, South Africa*

[5]*Department of Obstetrics and Gynaecology, Faculty of Health Sciences, University of Pretoria, Pretoria, South Africa*

OBJECTIVES

Increased intestinal permeability or so-called "leaky gut," is considered an early event in the development of obesity and diabetes. It is likely driven by the translocation of bacteria-derived products such as lipopolysaccharide (LPS) from the intestinal lumen into the blood stream leading to low-grade inflammation. A co-culture model of intestinal barrier dysfunction was used to characterize the mechanisms of barrier protection induced by an anti-inflammatory drug.

METHODOLOGY

In co-cultured CACO-2 and THP-1 cells representing the intestinal epithelium and immune cells, respectively, barrier dysfunction was induced using LPS and quantified by measuring trans-epithelial electrical resistance (TEER). Dexamethasone was tested for its barrier-protective properties by application to the apical side. Dexamethasone concentration was quantified on apical and basolateral sides after 24-hour treatment using liquid chromatography/mass spectrometry. Cytokine release was determined using a multiplex chemiluminescence assay (mesoscale discovery). Expression of selected tight junction (TJ) proteins was assessed using immunocytochemical staining and confocal imaging.

RESULTS

Dexamethasone significantly ($p=0.009$) improved TEER by 30%, while 74% of the dexamethasone remained on the apical side and 24% was detected basolaterally. Dexamethasone significantly decreased interleukin (IL)-6 ($p<0.0001$), tumour necrosis factor α ($p<0.0001$) and IL-1β ($p=0.051$) release by THP-1 cells. However, confocal imaging revealed that dexamethasone did not improve the localization of occludin, claudin-2 or zonula occludens-1 when compared to LPS-induced cells.

CONCLUSION

Dexamethasone improved barrier function likely due to its anti-inflammatory effect. However, it could not prevent localization of the pore-forming claudin-2, which may explain the presence of residual barrier dysfunction in this model.