

# Immunomodulatory effects of dietary supplements on IPEC-J2 cells

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## Organisation

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## SCOPE OF THE METHOD

<b>The Method relates to</b>	Animal health, Human health
<b>The Method is situated in</b>	Basic Research, Translational - Applied Research
<b>Type of method</b>	In vitro - Ex vivo
<b>Species from which cells/tissues/organs are derived</b>	pigs
<b>Type of cells/tissues/organs</b>	IPEC-J2, porcine small intestinal cells

## DESCRIPTION

### Method keywords

immune response

cell culture

ELISA

MTT

citotoxicity

Lps challenge

### **Scientific area keywords**

gut health

immunomodulation

postbiotic

gut barrier

intestine

### **Method description**

Prebiotics are used by the gut microbiota creating metabolites that exert a positive effect on the host (postbiotics). Prebiotics are available on the market for human and animal consumption and may induce differential effects on the host's health and metabolism. They can be tested by in vitro methods to evaluate their capacity to exert a beneficial effect on gut inflammation and barrier by using in vitro assays and cell cultures. IPEC-J2 cells are intestinal epithelial cells non-transformed obtained from the jejunal segment from a newborn and un-suckled piglet. Cells are grown in flat bottom flasks/plates at 37°C and 5% CO<sub>2</sub> humidified atmosphere in complete DMEM/F-12 supplemented with 1% penicillin-streptomycin, 5% fetal bovine serum, L-glutamine, EGF, Insulin, transferrin and selenium. Screening of cytotoxicity of the products of interest by MTT test in a concentration series of the product of interest to establish the dose. Incubation for 24 hours with the chosen concentration of product and parallel LPS challenge. Collection of supernatants and cells for the measurement of the immune mediators released in the medium by the cells, such as cytokines. Collection of cells for RNA extraction and evaluation of gene expression changes induced in the cells by qPCR methods. In parallel, cells can be cultured in snapwell for studying barrier integrity properties by measuring permeability changes and transepithelial resistance in Ussing chambers.

### **Lab equipment**

CO<sub>2</sub> incubator ; Water bath ; Hood (sterile) ; Centrifuge ; Cell counter ; Pipet controller ; Pipettes ; Spectrophotometer ; qPCR ; Ussing chambers.

## **Method status**

Published in peer reviewed journal

## **PROS, CONS & FUTURE POTENTIAL**

### **Advantages**

1. Fast, easy to set up, reproducible, high-throughput screening capacity. 2. Limited ethical and economic constraints. 3. A valuable tool for initial investigation on gut health studies prior to in vivo tests.

### **Challenges**

The gut is a complex tissue where several cell-types constitute the intestinal barrier, this model however only includes intestinal epithelial cells, and thus, it is a much simplified system.

### **Modifications**

The battery of analysis after incubation with the substrates of interest can vary according to the interest of the research. The products of interest have been described as pure prebiotics solutions, however, there are also probiotics and the combination of them as synbiotics that can also be tested. It might be of interest to do an in vitro batch fermentation before incubation with cells to study the postbiotic juice (product of the fermentation of prebiotics/synbiotics in the gut) to screen and evaluate the immunomodulatory effects on intestinal cells. For evaluation of the immune effects of postbiotic juice, before incubation with cells, it should be sterilefiltered.

### **Future & Other applications**

The system could be improved by co-culture in trans-well of intestinal epithelial cells in combination with immune cells.

## **REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION**

### **References**

Uerlings, J., Schroyen, M., Bautil, A., Courtin, C., Richel, A., Sureda, E. A., ... Everaert, N. (2019). In vitro prebiotic potential of agricultural by-products on intestinal

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