

# Healthy gut ecosystems

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## Organisation

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**Country** Belgium

**Geographical Area** Flemish Region

## 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

## DESCRIPTION

### Method keywords

gut  
bioreactor  
ex vivo  
gut microbiota  
cecum

### Scientific area keywords

microbiology

antimicrobial resistance

probiotics

prebiotics

digestion

## **Method description**

With a one vessel fermentor (BioFlo 110, New Brunswick), an *in vitro* simulation of a certain part of the gastrointestinal tract (e.g. cecum, colon) is performed. In this fermentation system, physiological gut parameters (pH, temperature) are kept constant, while chemico-physical characteristics of the gut chime are simulated by adding pork mucin and fibre to the fermentation medium. For a first screening, the fermentation system can be performed in short time (24-48 h) batch mode. The system is inoculated with cecal or fecal material from animals or humans, and can be inoculated with a pathogen like Salmonella. Batch fermentations are monitored by periodic sampling on a quantitative level by dilution plating on selective media for certain bacterial groups (e.g. lactic acid bacteria, bifidobacteria, Enterobacteriaceae, clostridia) as well as for Salmonella, and on a qualitative level for the total microbiota by cultivation independent 16S rDNA based metagenomics; production of short chain fatty acids is measured by GC. From these batch fermentations, for more detailed investigation long-term (up to 2 weeks) a continuous fermentation system is used which simulates even better the gastrointestinal tract of monogastrics like the pig because it takes into account the retention time (or conversely flow of intestinal content) in a certain part of the gut. A continuous fermentation is started in batch mode as described above after which fresh medium is added with a peristaltic pump to the fermentor vessel at a fixed dilution speed and fermented medium is wasted with the same speed to maintain the medium volume in the vessel. After a few days, a steady state is reached and bacteriological and fatty acid parameters can be monitored over several days (or weeks) as described above. All fermentations (batch and continuous) are performed at least twice and for every condition or component, a blank fermentation is included.

## **Lab equipment**

1 liter bioreactors with peristaltic pumps for in and outflow of medium ;

L2 lab ;

Incubators ;  
-80°C freezer ;  
UPLC-MS/MS.

### **Method status**

History of use  
Internally validated  
Published in peer reviewed journal

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

### **Advantages**

No animal experiments are needed to investigate the gut microbiota ;  
Ease of sampling and reproducible conditions can be executed ;  
Two modes of operation are possible depending on the time and resources available :  
1/ batch fermentation for screening is possible in a few days ;  
2/ continuous fermentation is possible for more in depth studies, taking a few weeks.

### **Challenges**

The *ex vivo* system only simulates the gut luminal microbial ecosystem without an animal or human host component (except for addition of porcine mucin to the medium) like gut epithelial cells and host immune system.

### **Modifications**

Addition of mucin beads is possible for simulation of attached bacteria to the mucin layer.

### **Future & Other applications**

Applications in biofilm research are possible, both in the gut ecosystem as on inert surfaces.

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

## References

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## Associated documents

[Campy-allicine.pdf](#)

[Campy-Enterococcus screening.pdf](#)

[ESBL conjugatie.PDF](#)

[Salmonella-in vitro.pdf](#)

[Selection and transfer of an IncI1-tet\(A\) plasmid.pdf](#)

[cross contamination doxycycline ex vivo model.pdf](#)

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