

EAAP 2024

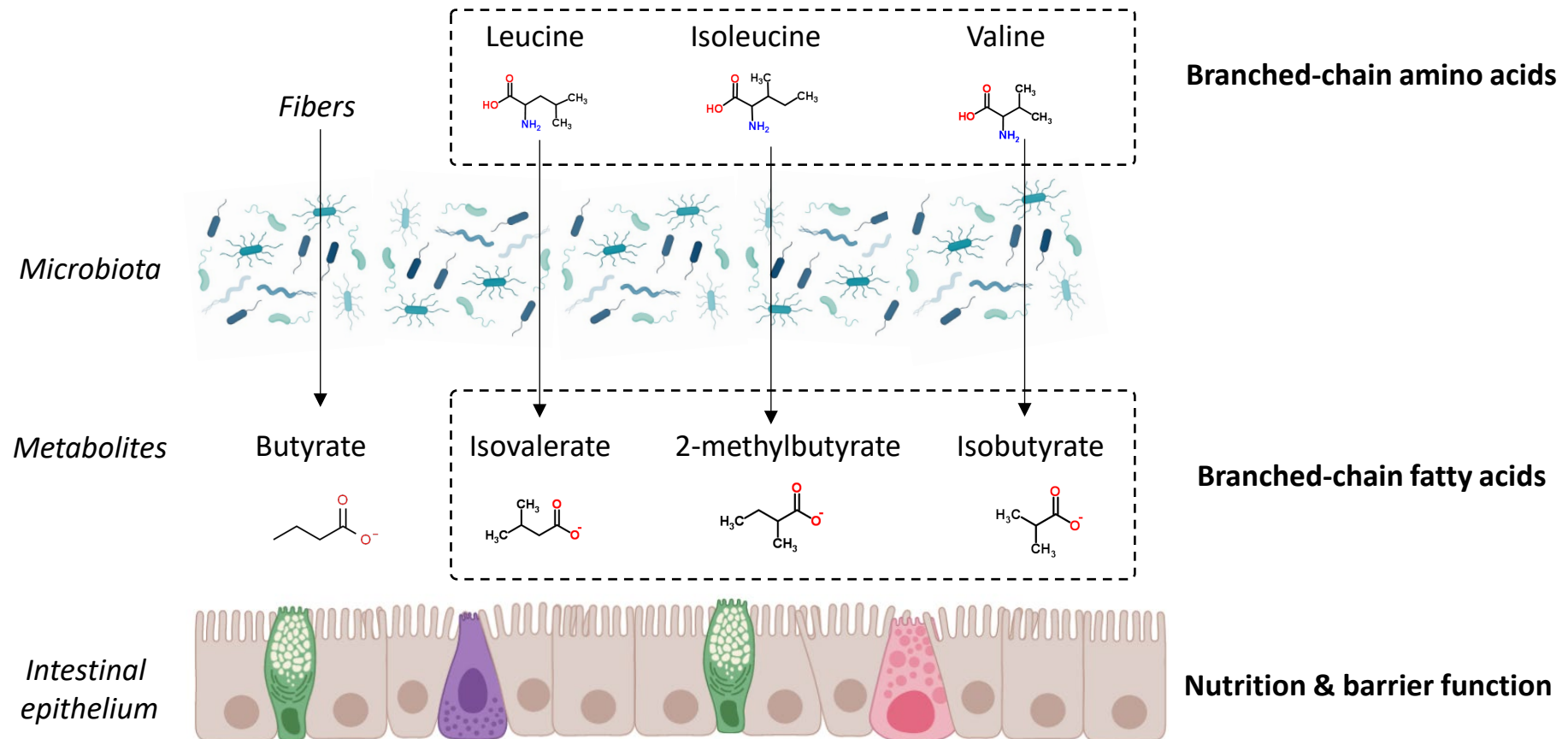
➤ The leucine-derived gut microbiota metabolite isovalerate enhances the epithelial barrier function in cell monolayers derived from pig ileum organoids

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2 - Eurolysine Avril, Paris, France.

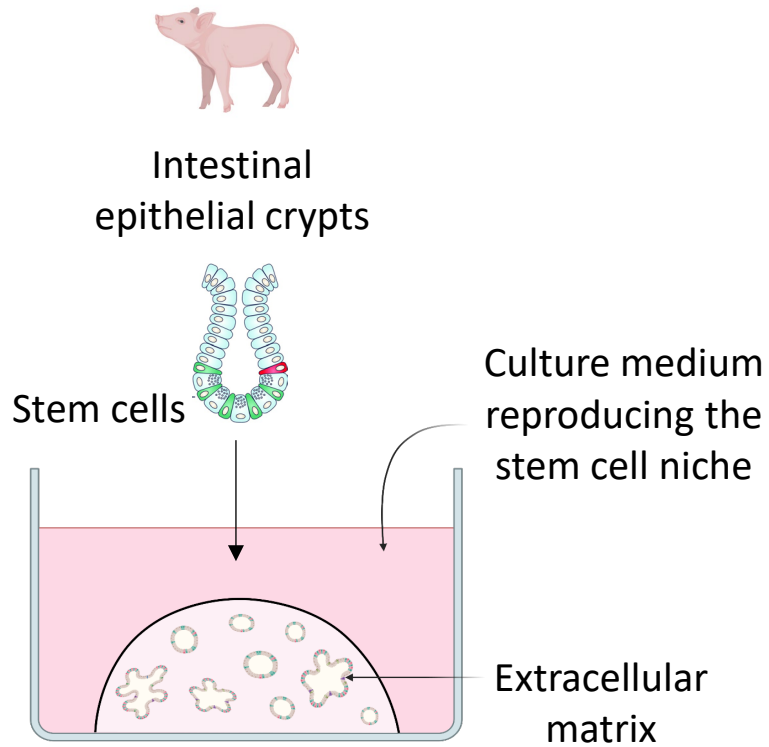
➤ Targeting microbiota-derived metabolites to improve gut health



What are the effects of branched-chain fatty acids on the pig intestinal epithelium?



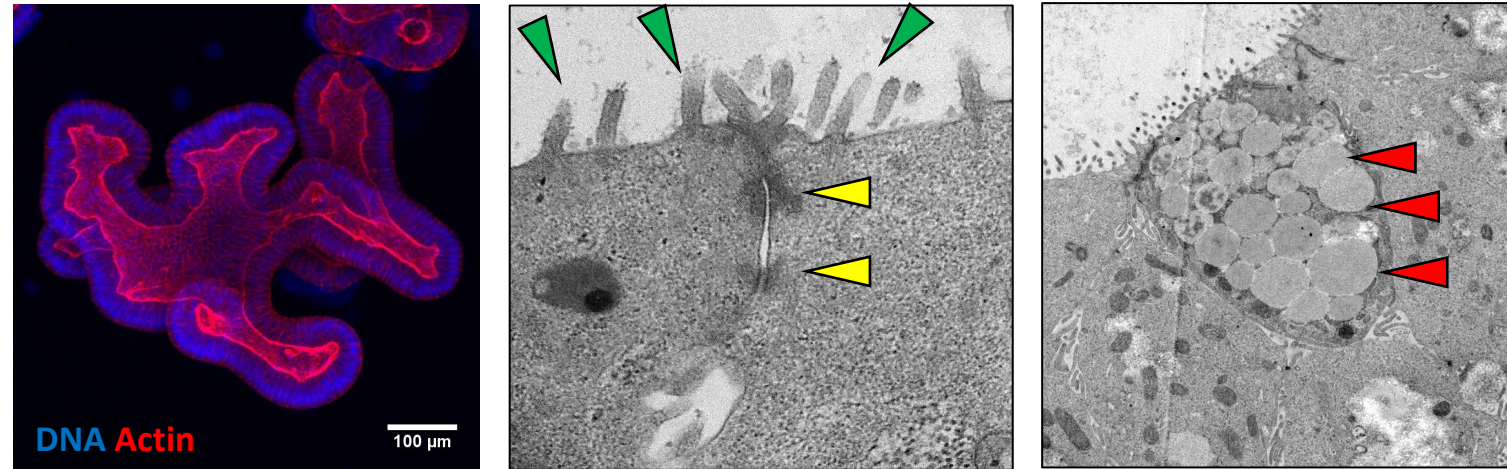
➤ Pig intestinal organoids



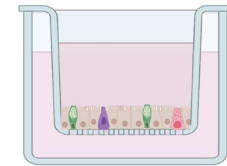
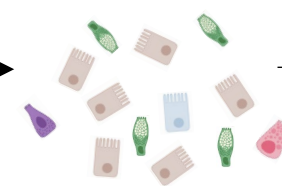
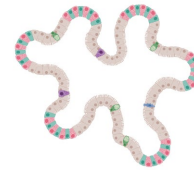
Mussard et al., Front. Cell. Dev. Biol., 2022

Mussard et al., JOVE, 2023

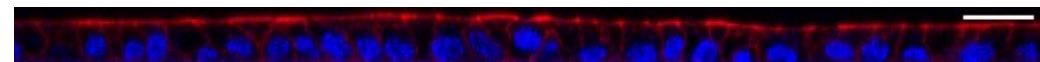
Beaumont et al., Vet Res, 2021



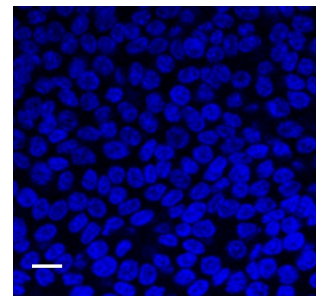
3D
organoids



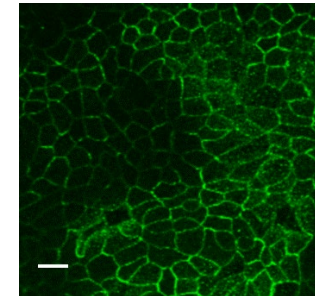
Monolayers



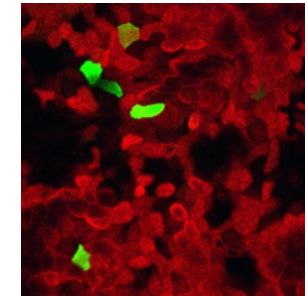
ADN
Actine



ADN



Occludine



WGA UEA1

Biology

Culture of Piglet Intestinal 3D Organoids from Cryopreserved Epithelial Crypts and Establishment of Cell Monolayers

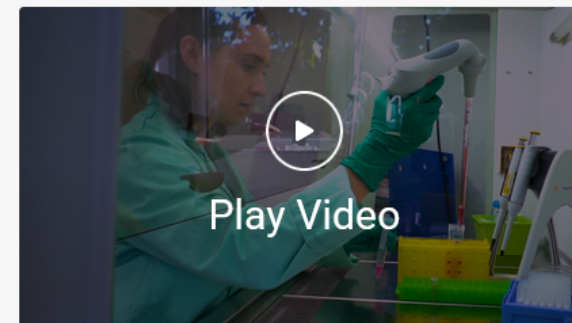
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Summary

Here, we describe a protocol to culture pig intestinal 3D organoids from cryopreserved epithelial crypts. We also describe a method to establish cell monolayers derived from 3D organoids, allowing access to the apical side of epithelial cells.



Play Video



PDF

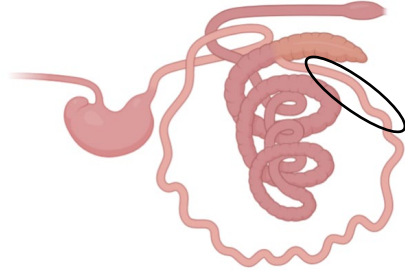
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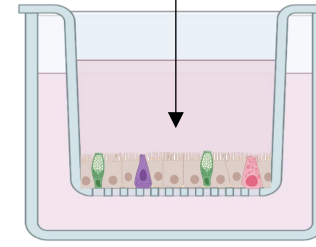
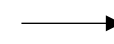
➤ Effects of microbiota metabolites



Ileum



3D organoids

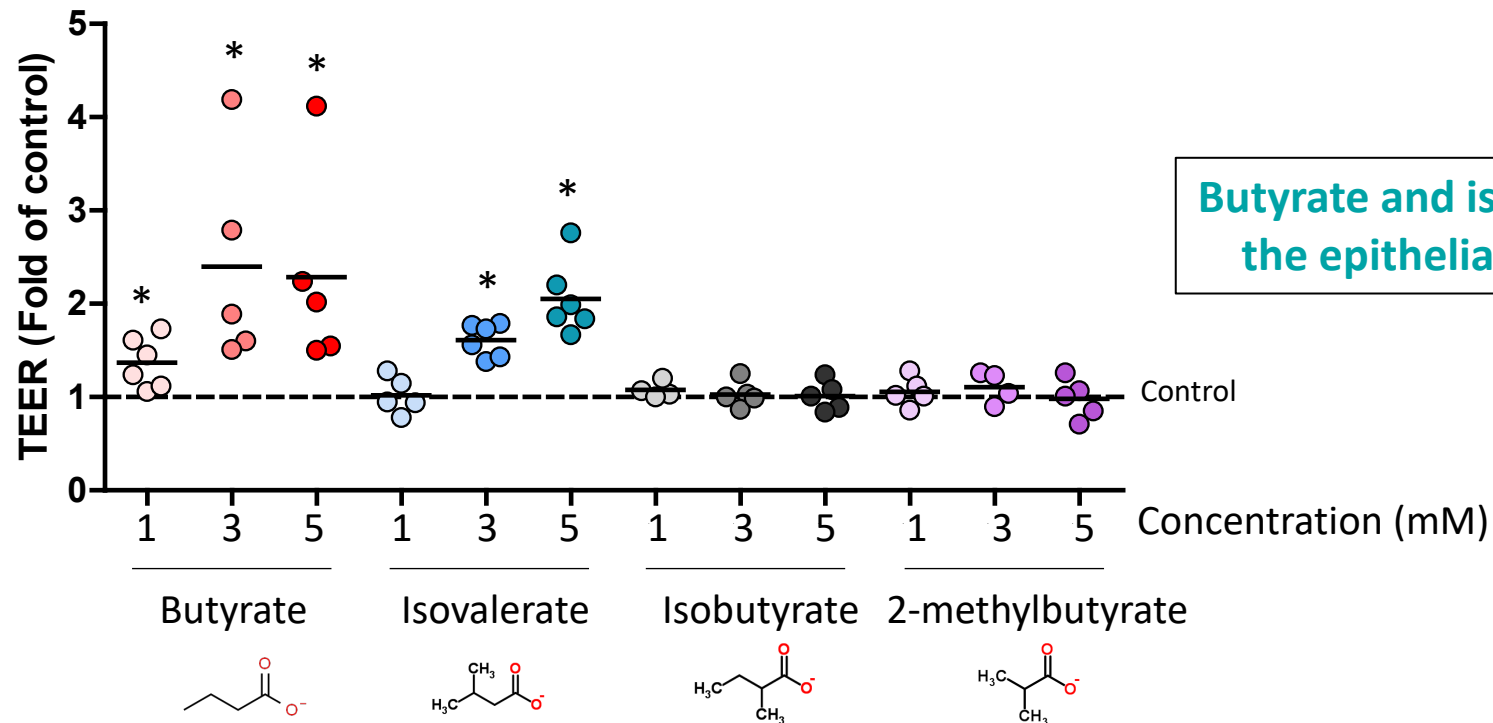


Monolayers

- PBS (negative control)
- Butyrate (1 - 3 - 5 mM)
- Isovalerate (1 - 3 - 5 mM)
- Isobutyrate (1 - 3 - 5 mM)
- 2-methylbutyrate (1 - 3 - 5 mM)

Epithelial barrier function

Transepithelial electrical resistance (TEER)



Butyrate and isovalerate enhance the epithelial barrier function

*: P<0.05 vs control group

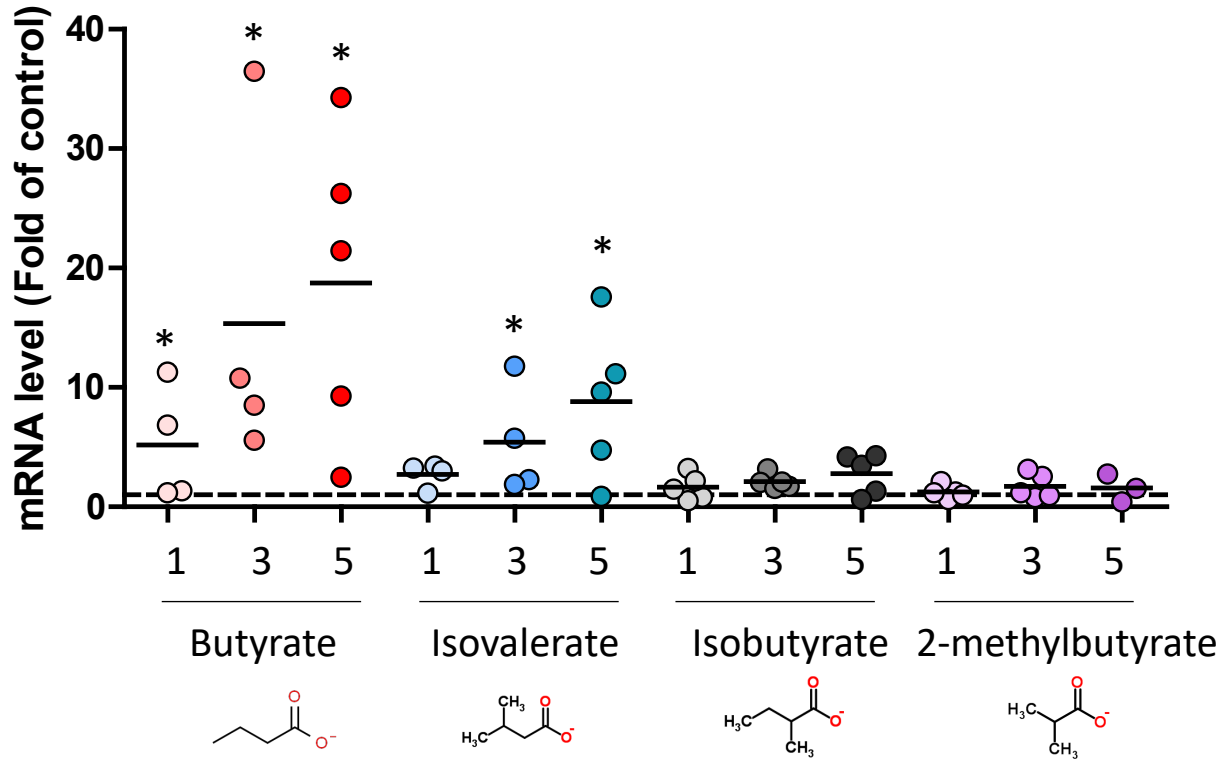
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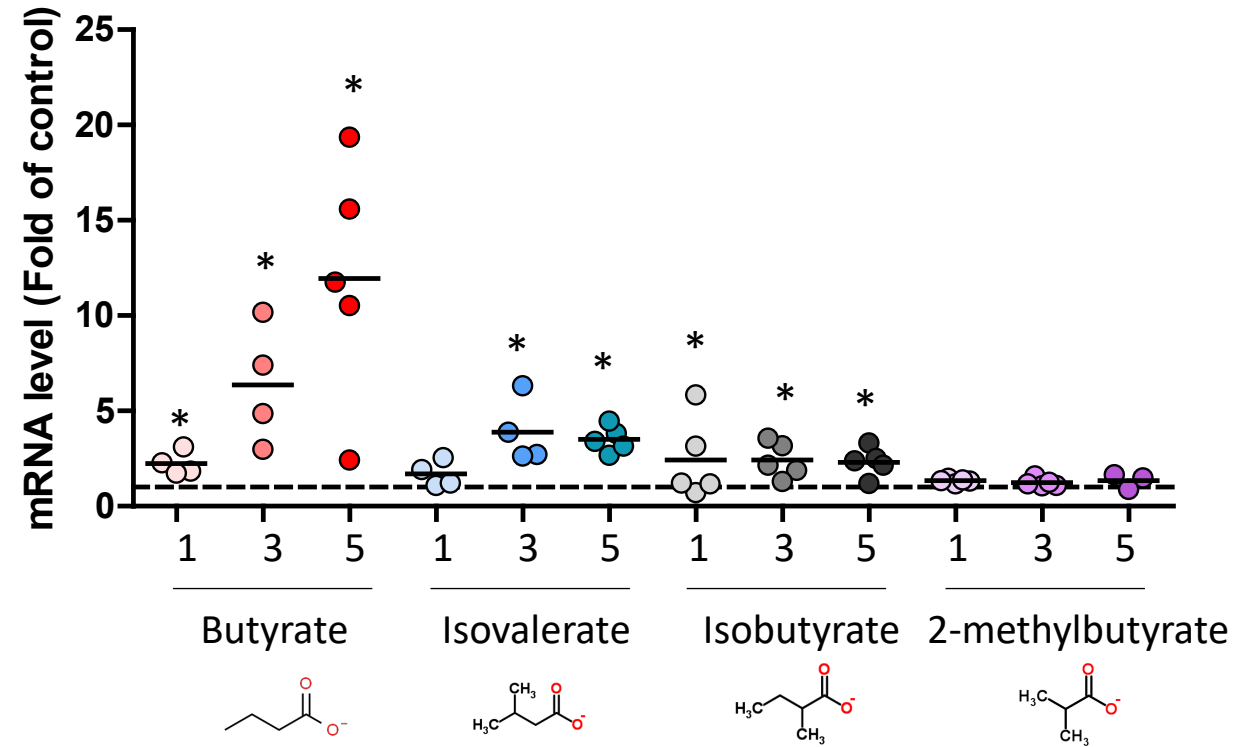
➤ Antimicrobial defenses

qPCR

SLPI (antimicrobial peptide)



CXCL8 (cytokine)



*: P<0.05 vs control group

INRAE

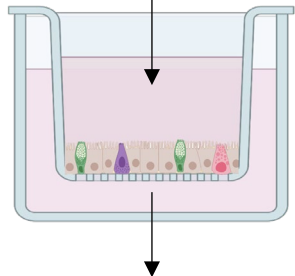
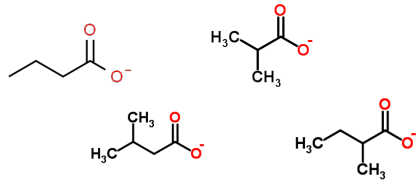
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Butyrate and isovalerate induce the expression of genes involved in epithelial defenses

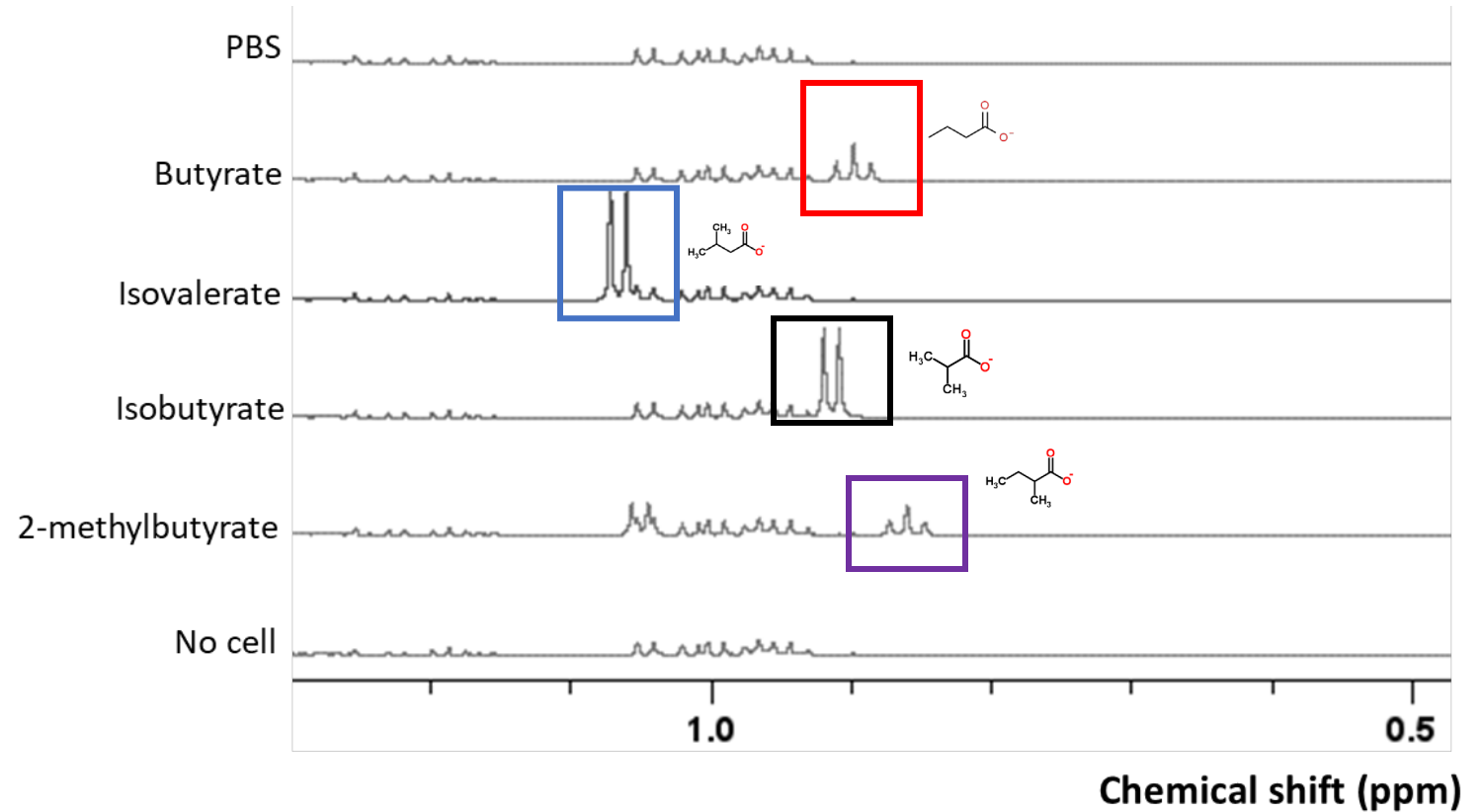
➤ Transepithelial transport of bacterial metabolites

Metabolites (5 mM)



Basal medium

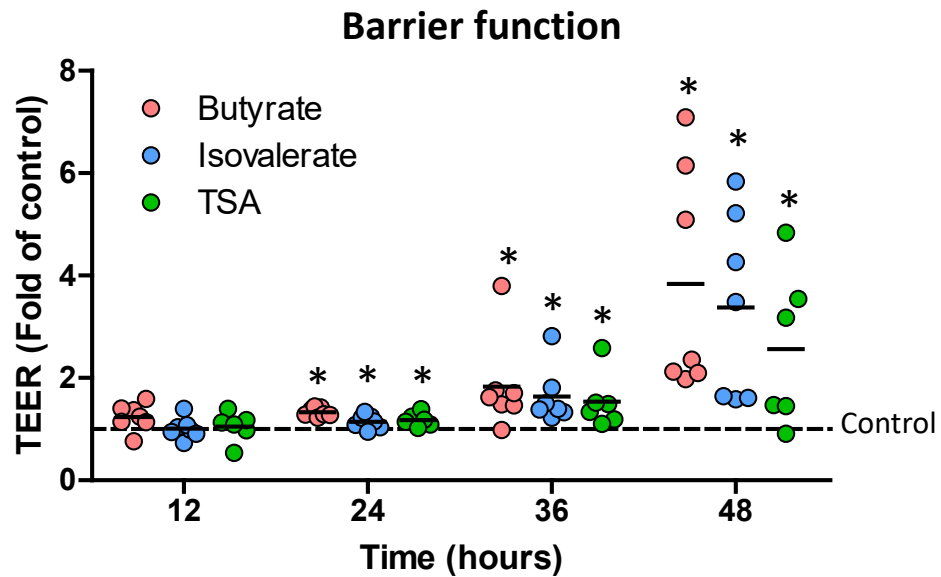
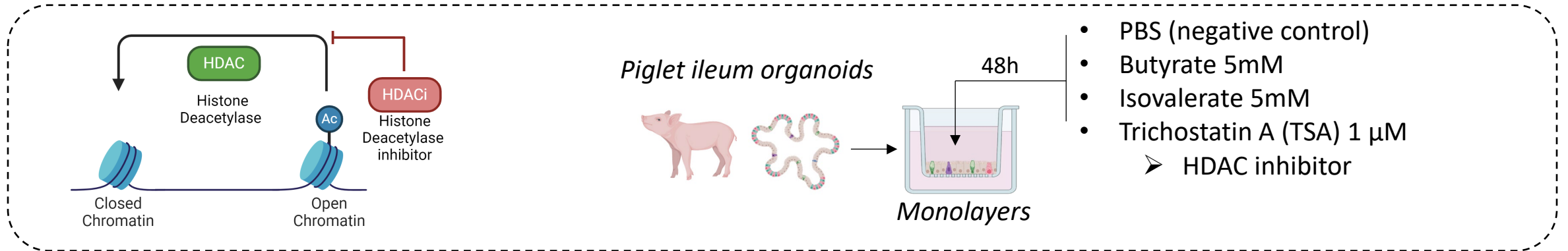
*Nuclear magnetic resonance (NMR)
spectroscopy*



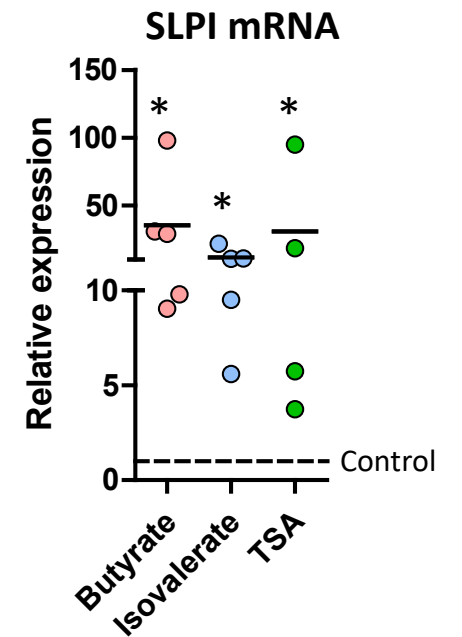
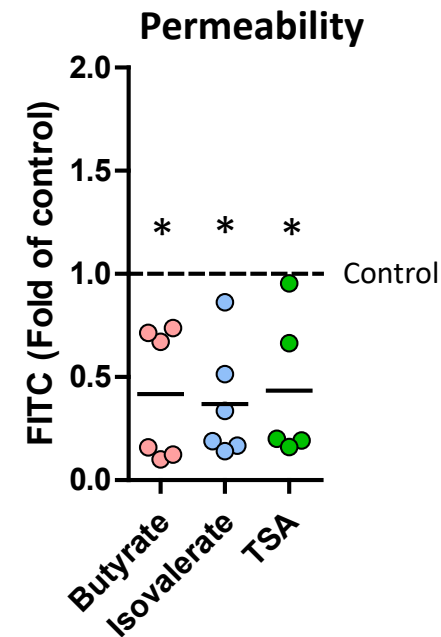
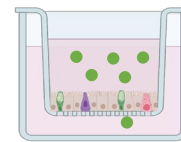
Butyrate and branched-chain fatty acids cross the pig intestinal epithelium *in vitro*

Transepithelial transport is not specific of butyrate and isovalerate that enhance the barrier function

➤ Mode of action: epigenetic effect?



FITC-dextran
4 kDa



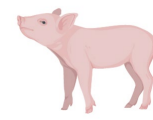
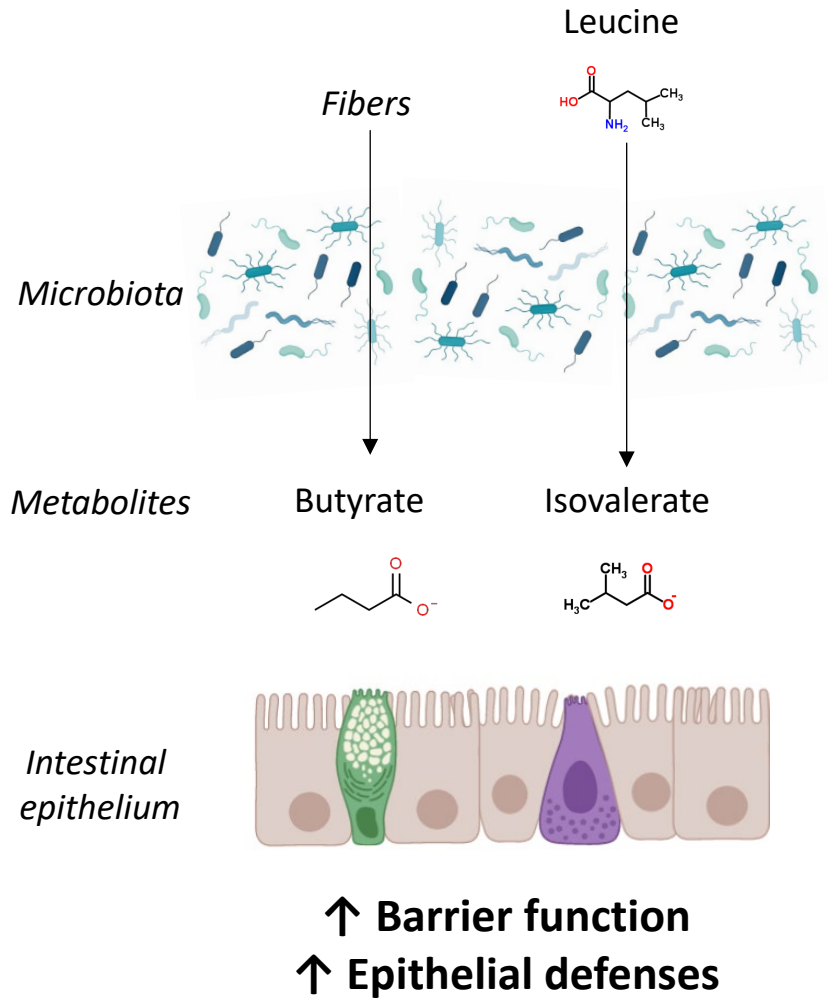
*: $P < 0.05$ vs control group

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HDAC inhibition may be involved in the effects of butyrate and isovalerate on the epithelial barrier

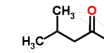
> Conclusion



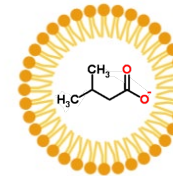
Isovalerate-based feed additives to improve piglet gut health?

Postbiotic

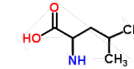
Isovalerate



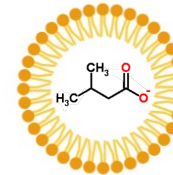
Protected isovalerate



Leucine



Protected leucine



Prebiotic

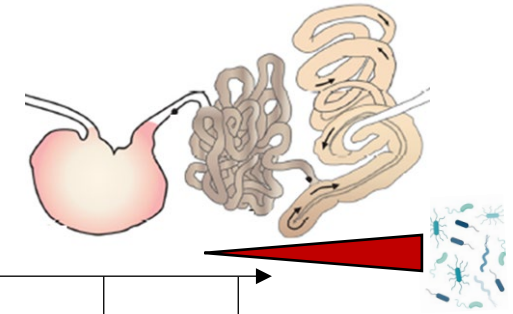
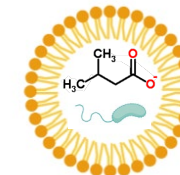
Probiotic

Isovalerate producing bacteria



Symbiotic

Probiotic + Leucine



Absorption

Selective nourishing of gut microbiota with amino acids: A novel prebiotic approach?

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Conny Turni⁴, Joris Michiels⁵ and
Tristan Chalvon-Demersay^{3*}

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➤ Acknowledgments



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