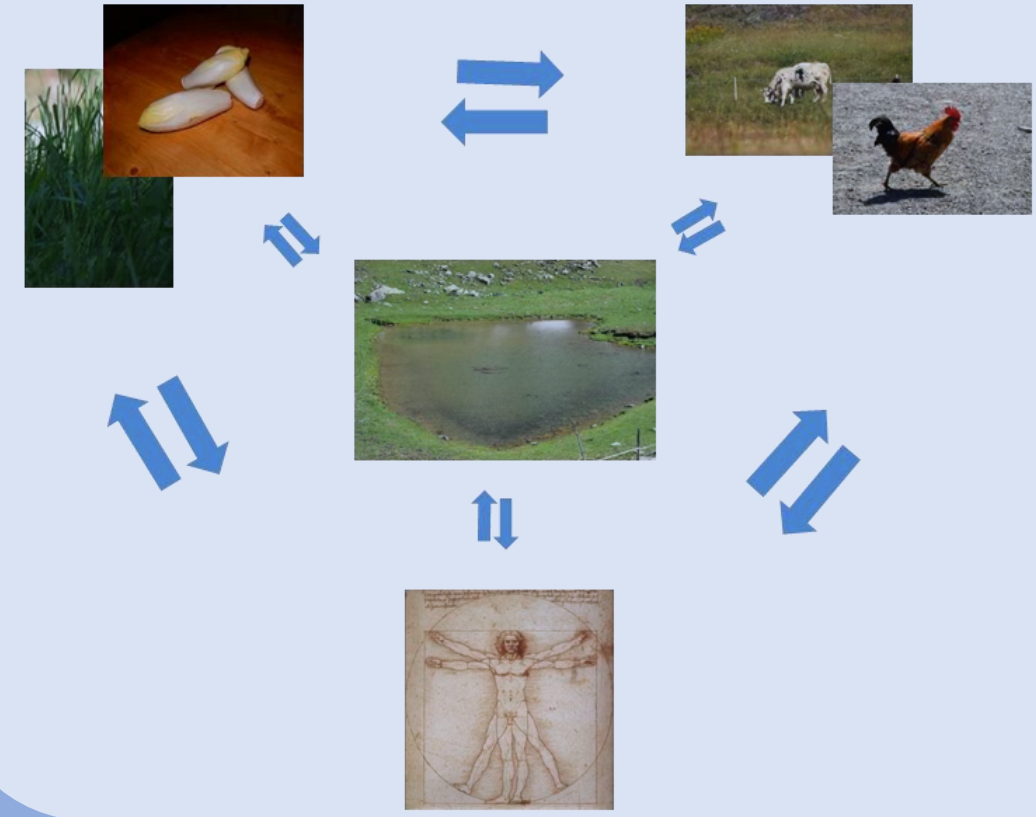


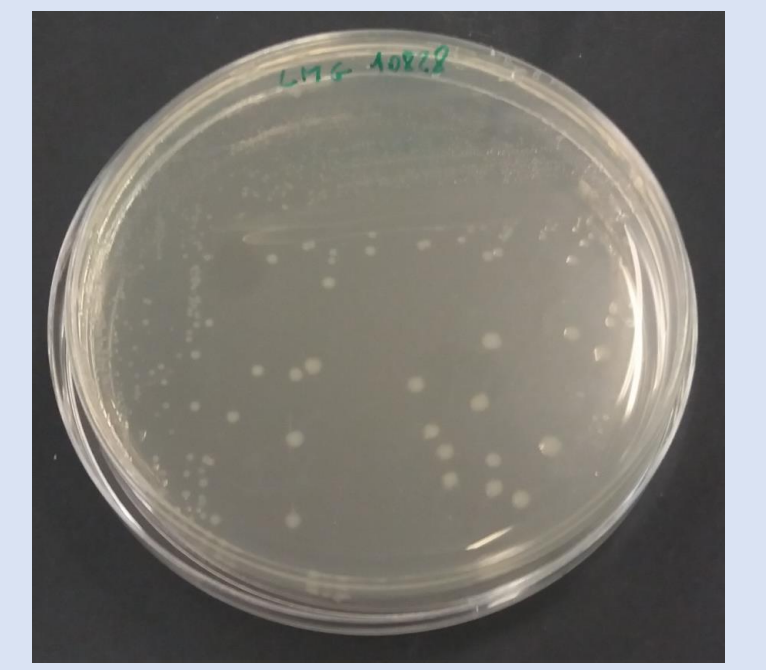
Daide Buzzanca*, Cristian Botta, Luca Coccolin, Valentina Alessandria, Kalliopi Rantsiou

Department of Agricultural, Forest and Food Sciences (DISAFA), University of Turin, Largo Paolo Braccini 2, 10095 Grugliasco (TO). *davide.buzzanca@unito.it



INTRODUCTION

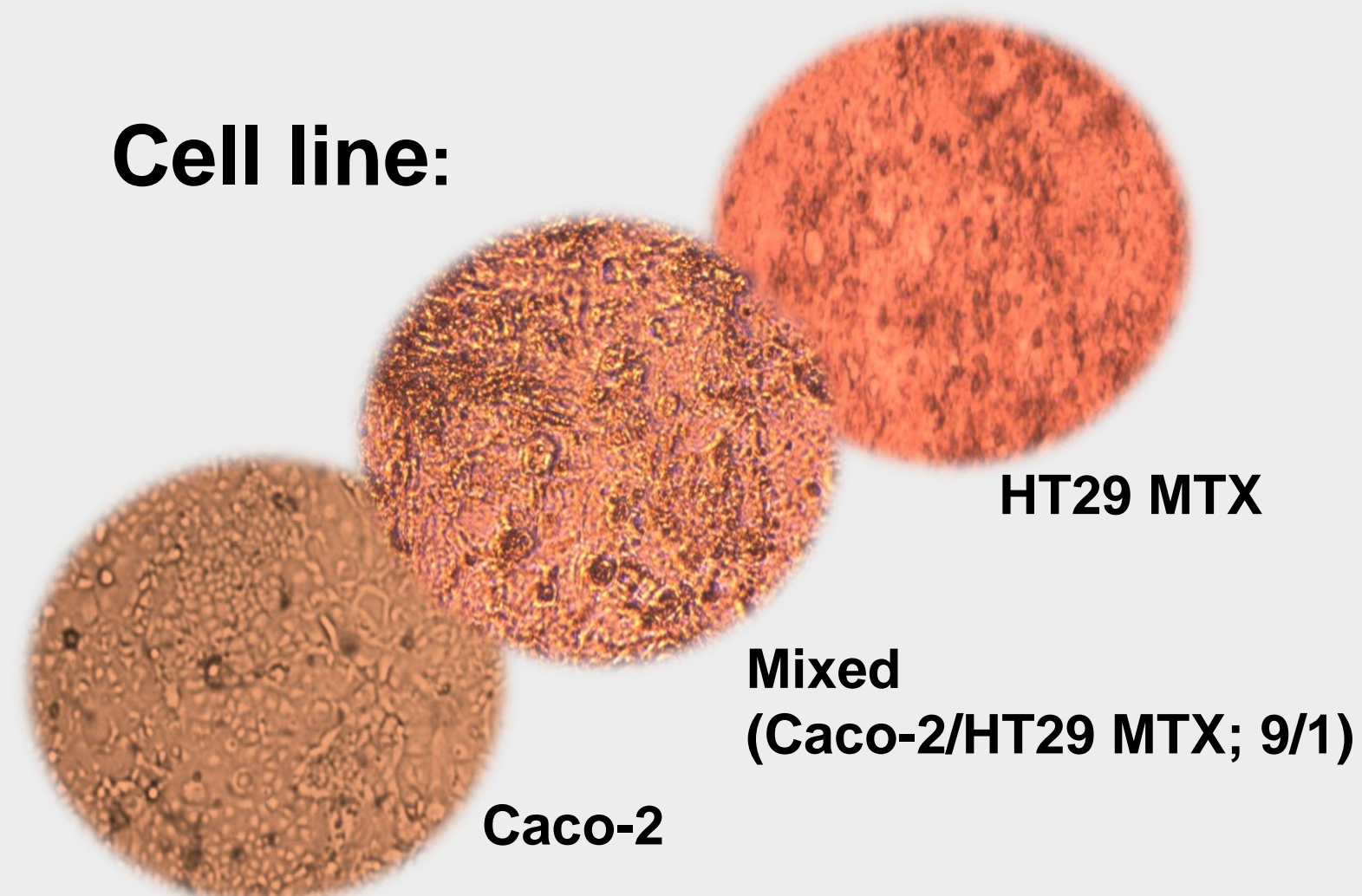
Arcobacter butzleri is an emerging foodborne pathogen of the *Campylobacteraceae* family, often isolated from chicken, pork and bovine meat, which causes different gastrointestinal diseases in human and animals. However, pathogenicity of *A. butzleri* is still underestimated due to a substantial lack of information on its virulence mechanisms, metabolic and genomic features. The aim of this study is an increase, through the use of **gut models** and **molecular biology methods**, the knowledge about *A. butzleri* virulence mechanism.



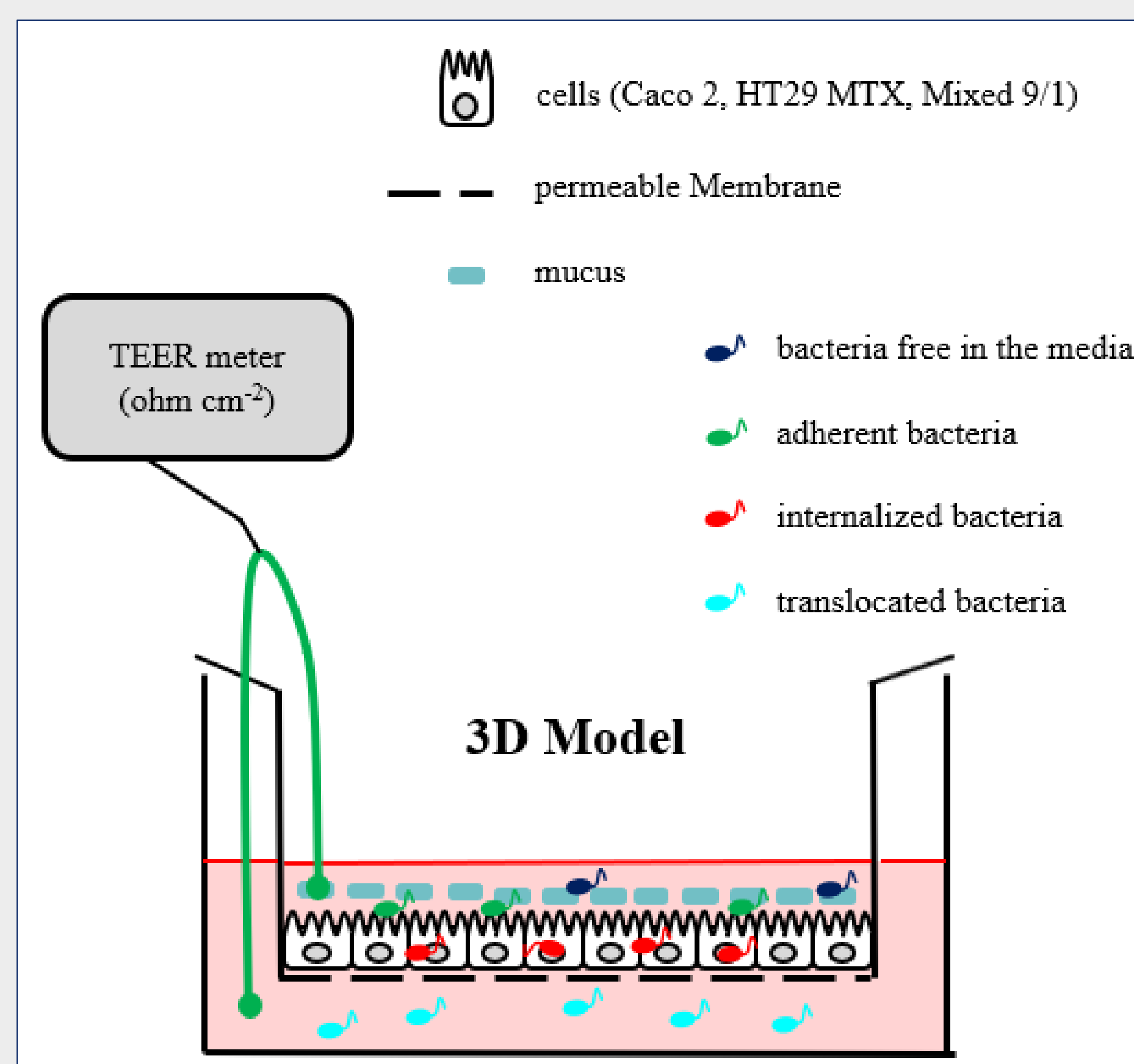
MATERIALS AND METHODS

Strain: *A. butzleri* LMG 10828^T

Cell line:



- Study of:
- ✓ Colonization
 - ✓ Invasion
 - ✓ Intestinal barrier permeability (TEER)
 - ✓ Translocation



... RT-qPCR analysis
focus on *Campylobacter jejuni* related virulence genes present in the genome of *A. butzleri* LMG 10828^T (gene similarity)

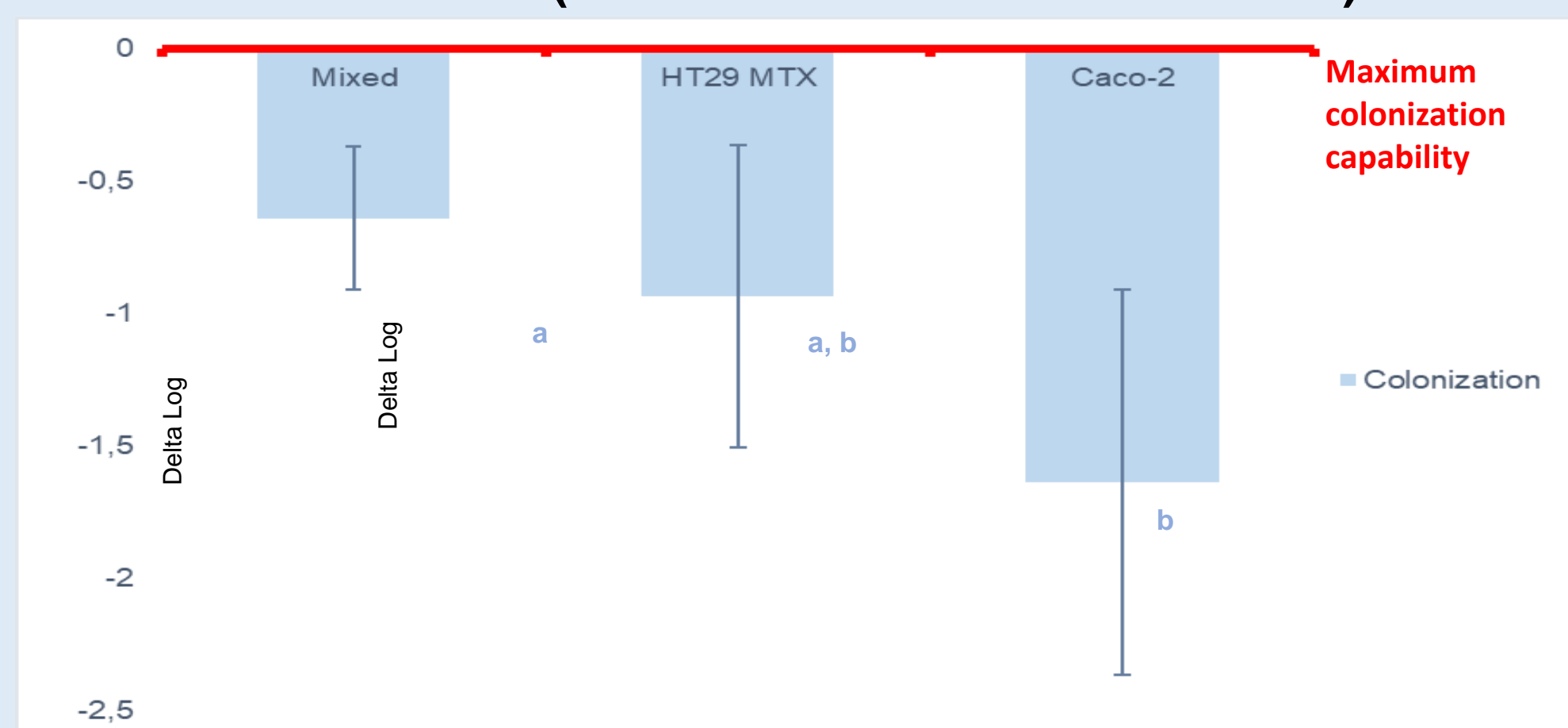
Virulence genes	Function
<i>cadF</i>	Adhesion
<i>cj1349</i>	Adhesion
<i>ciaB</i>	Invasion

In silico primer design
In vitro primer validation during colonization conditions

RESULTS

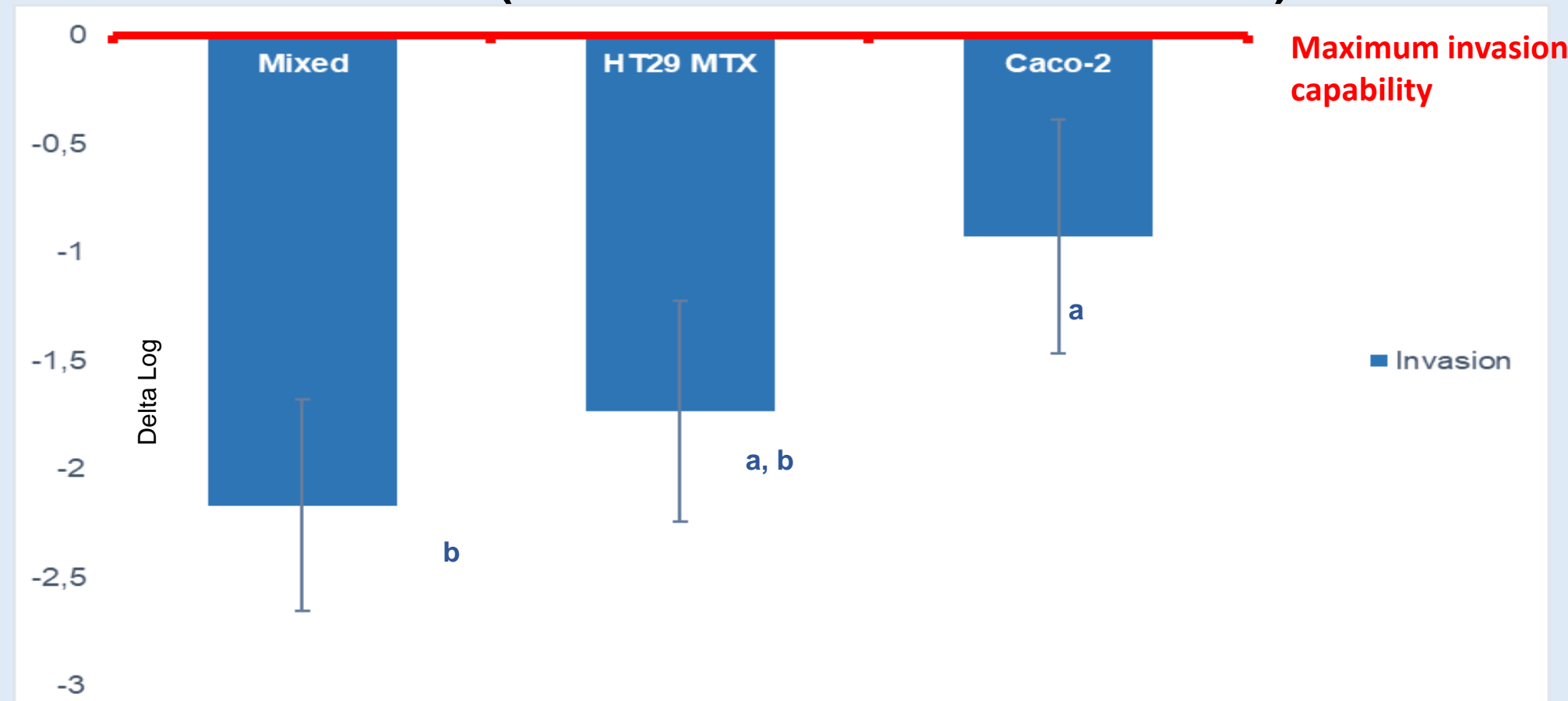
2D MODELS

Colonization (90 minutes after initial inoculum)



Higher colonization on mixed cell lines

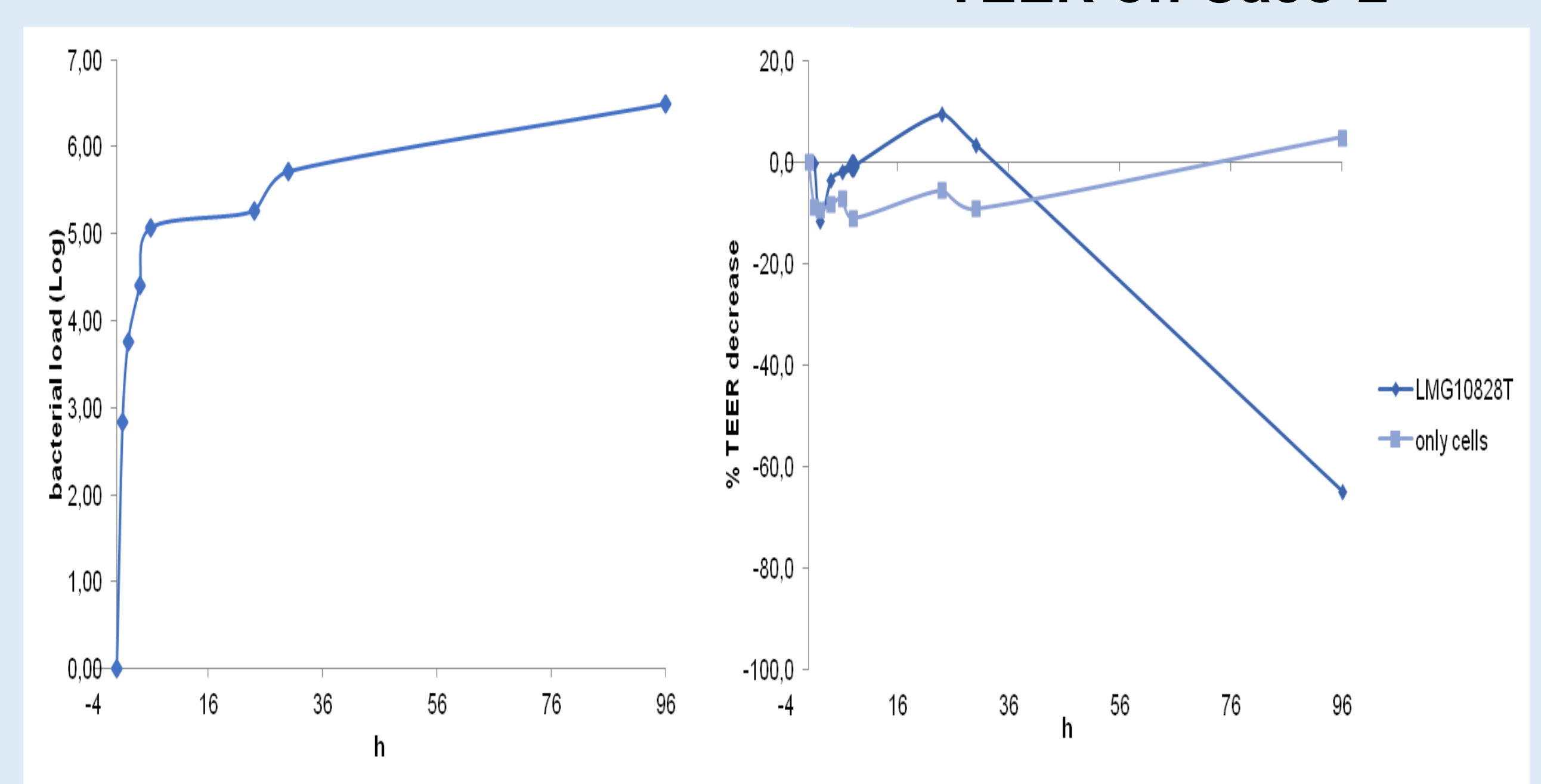
Invasion (90 minutes after initial inoculum)



Higher invasion capability on Caco-2 models compared to Mixed models

3D MODELS

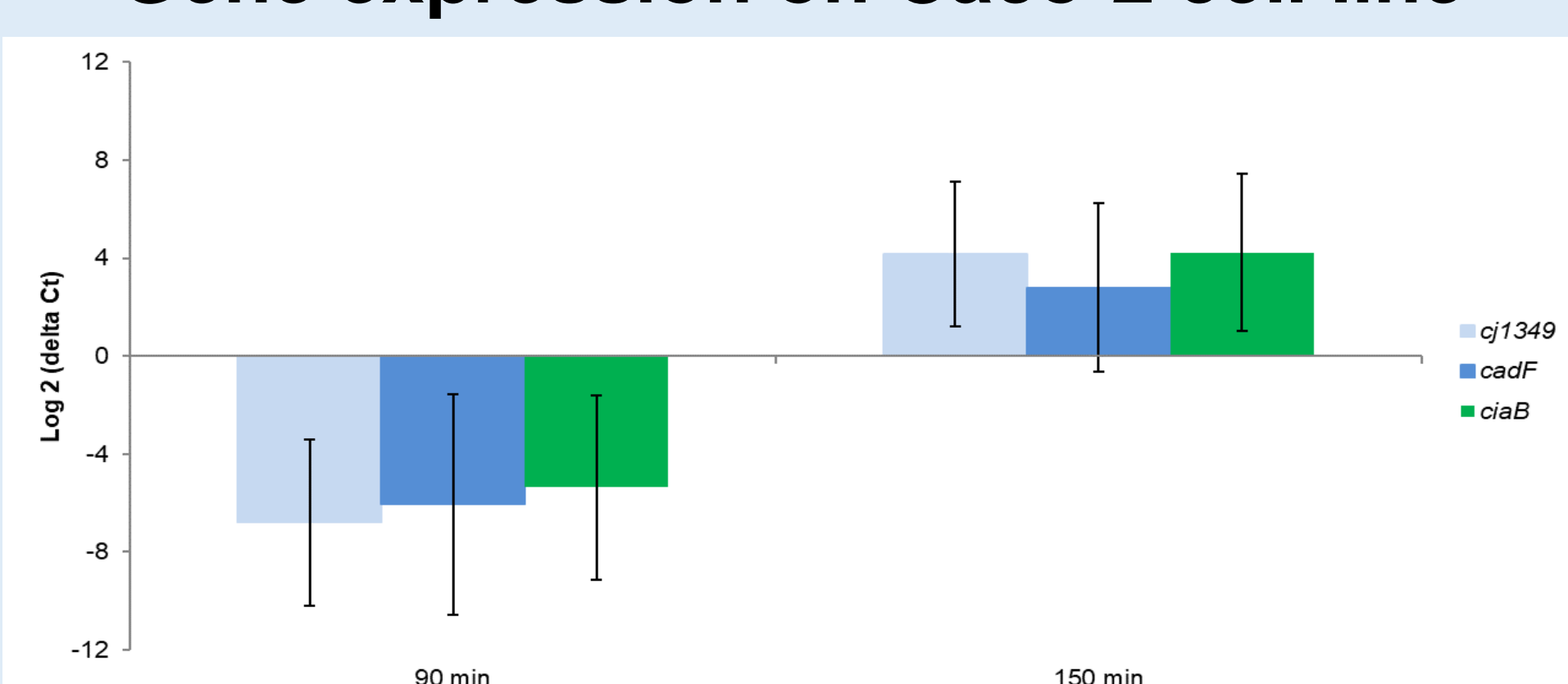
Translocation on Caco-2



All *A. butzleri* cells translocate through the epithelial layer before the first 24 hours

No significant variation of the TEER values were observed within the first 24 hours

Gene expression on Caco-2 cell line



RT-qPCR set-up

Primer3-BLAST design followed by *in vitro* test to confirm absence of non-specifics, appropriate thermal cycle and right target amplification

Gene expression preliminary results

Overexpression of adhesion- and invasion-related genes after 150 minutes from the addition of *A. butzleri* in the models.

CONCLUSIONS

- ✓ Higher colonization in the mixed models
- ✓ Translocation of the bacteria through the layer after 24 hours not linked with TEER decrease
- ✓ Obtainment of a RT-qPCR protocol to study *A. butzleri* virulence gene expression
- ✓ Overexpression of the virulence genes after 150 minutes from the bacteria inoculum

- ✓ The subsequent experiment will deepen the knowledge about gene expression of others genes attributed at the virulence