

# A new Bifidobacteria Expression SysTem (BEST) to produce and deliver heterologous proteins

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# Introduction

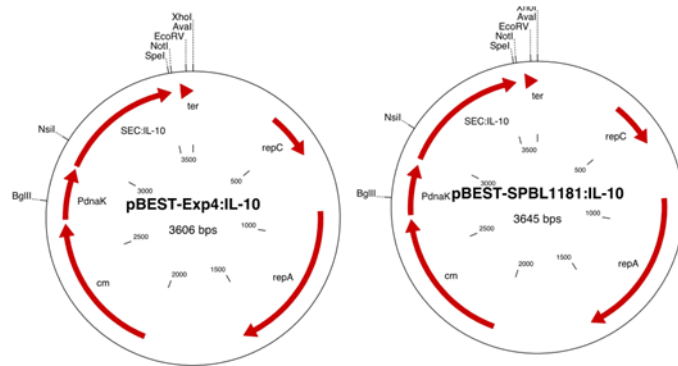
- Bifidobacteria are good candidates to deliver therapeutics molecules at mucosal surface
  - Well recognized probiotics properties for some strains
  - GRAS status (Generally Recognized As Safe)
  - Adapted to the human gut
- But, bifidobacteria are complex to be genetically manipulated
- IL10 cytokine is an anti-inflammatory cytokine successfully expressed in different LAB, with a beneficial effect on the treatment of colitis (Martin *et al*, 2014)

The development of a new Bifidobacteria Expression SysTem (BEST), allowing the production and delivery of IL10 *in-vitro* and tested in a murine model of colitis as a proof of concept

# Development of BEST system for the production and delivery of heterologous proteins by recombinant bifidobacteria

Plasmid vectors:

- pDNA-K, an inducible promoter, stress sensitive
- two different signal peptides :
  - SP<sub>Exp4</sub> from *L.lactis*
  - SP<sub>1181</sub> from *B.longum*

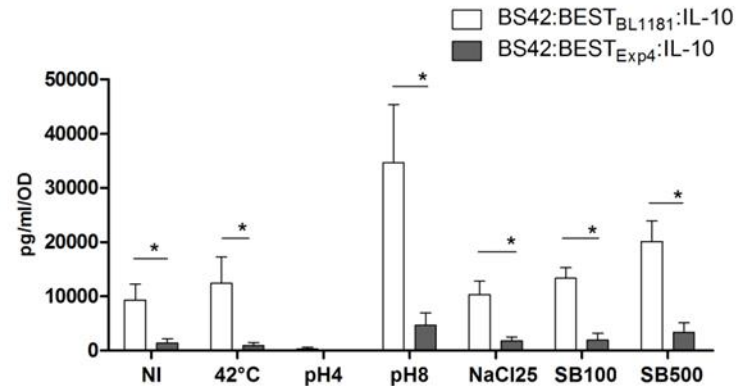


Transformation in 3 different strains of bifidobacteria :

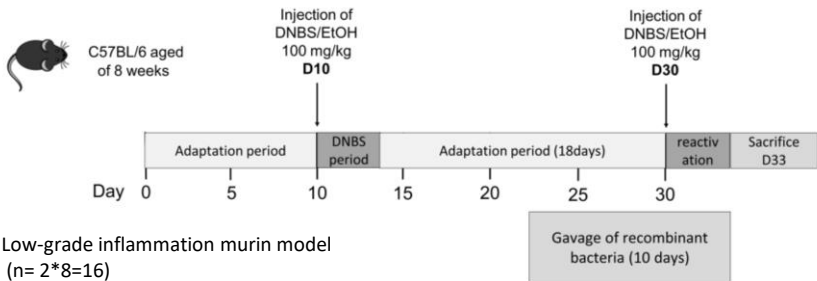
- B.bifidum* (BS42)
- B.breve* (BS56)
- B.longum* (NCC2705)

➔ Selection of BS42 based on previous work (Ménard et al. 2008)

Impact of stress on secretion of IL10



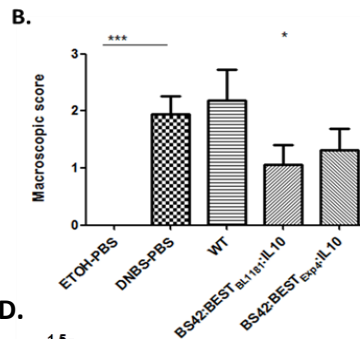
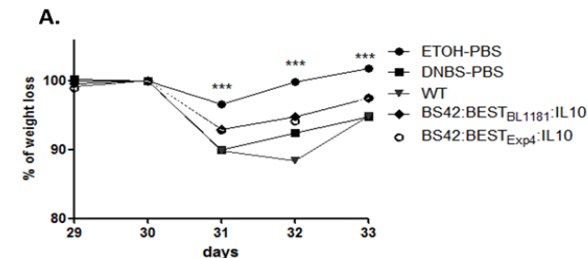
# Impact of IL-10 delivery by recombinant *B. bifidum* using BEST system in a low-grade inflammation murine model



Low-grade inflammation murin model  
(n= 2\*8=16)

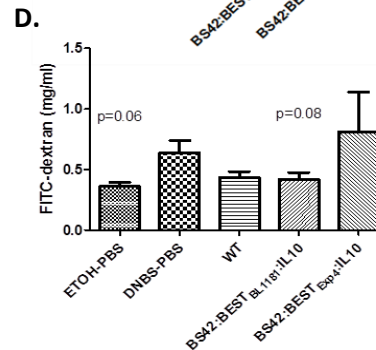
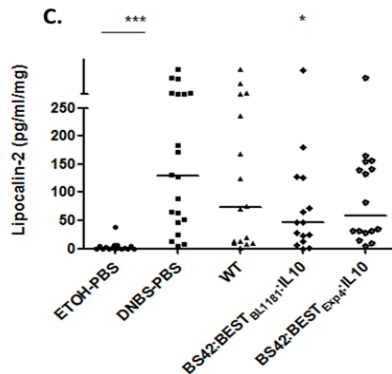
SHAM CTL : Mice receive ETOH and PBS without bacteria  
 Positive controle : Mice receive DNBS and PBS without bacteria  
 Others groups : Mice receive DNBS and PBS with bacteria

Percentage of weight loss



Macroscopic score

Dosage of Lipocalin-2, early marker of inflammation



Evaluation of intestinal permeability by the FITC-dextran dosage

# Conclusion

- BEST system was successfully established in 3 different *Bifidobacterium*
- This inducible system is efficient with a better secretion with the pBEST-BL1181-IL10
- BS42: BEST<sub>BL1181</sub>:IL10 had beneficial effect on colitis model:
  - a significant effect on macroscopic score
  - a significant effect on lipocaline-2
  - tendencies in intestinal gut permeability

**The BEST system is relevant for delivery of therapeutic molecules in the colon**

# Thanks for your attention



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