A mucin-enriched fermentation model to assess prebiotic potential of new indigestible carbohydrates

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Context

- Prebiotics for gut health
  - New indigestible carbohydrates ➔ Prebiotic potential ????
  - In vivo trials = substrate and time consuming
  - In vitro screening of new indigestible carbohydrates
- In vitro models
  - Different models : simple to complex, cheap to expensive
  - Simple batch fermentation model

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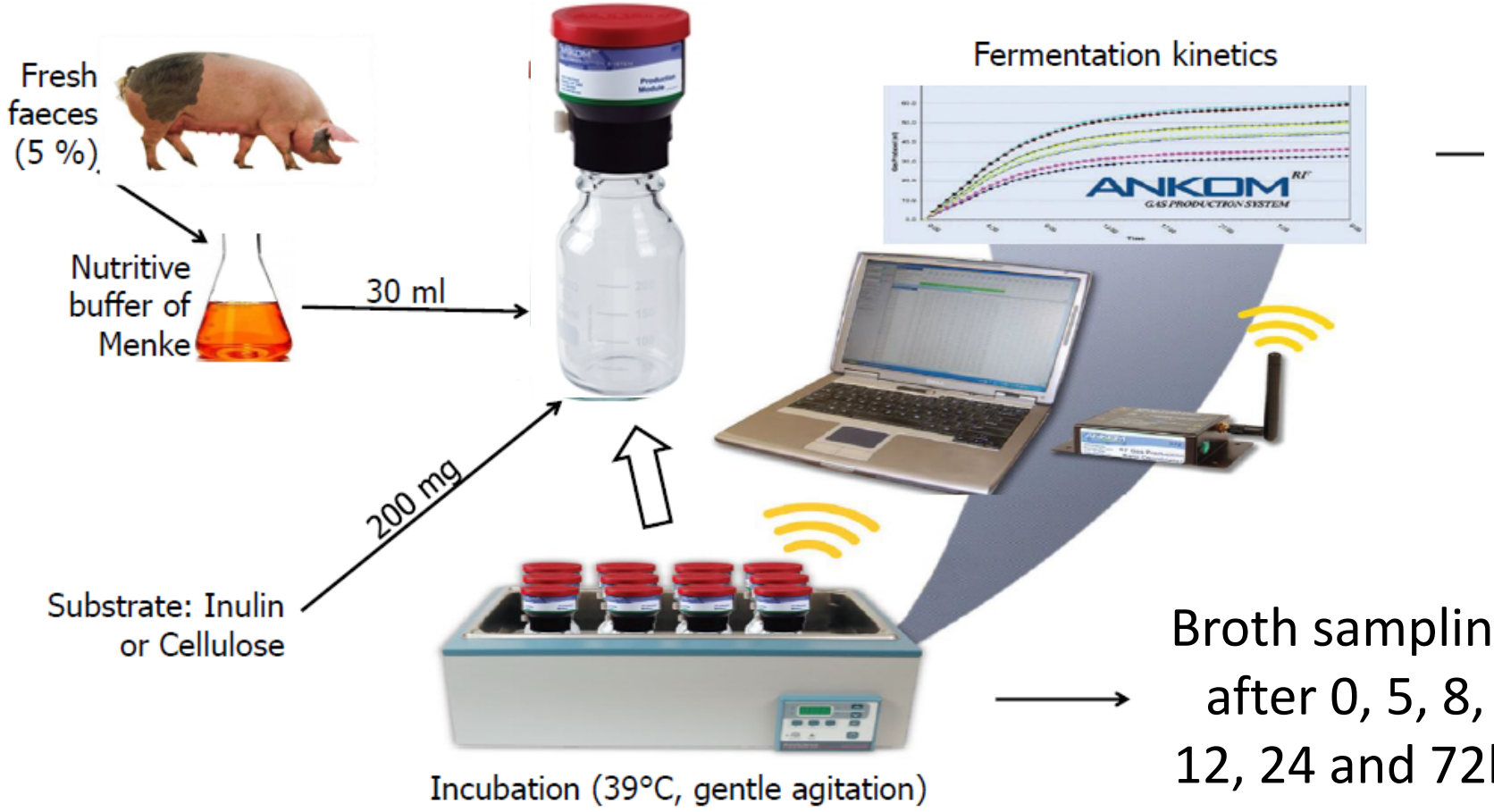
  - Simple batch fermentation model
    - Screening

Context

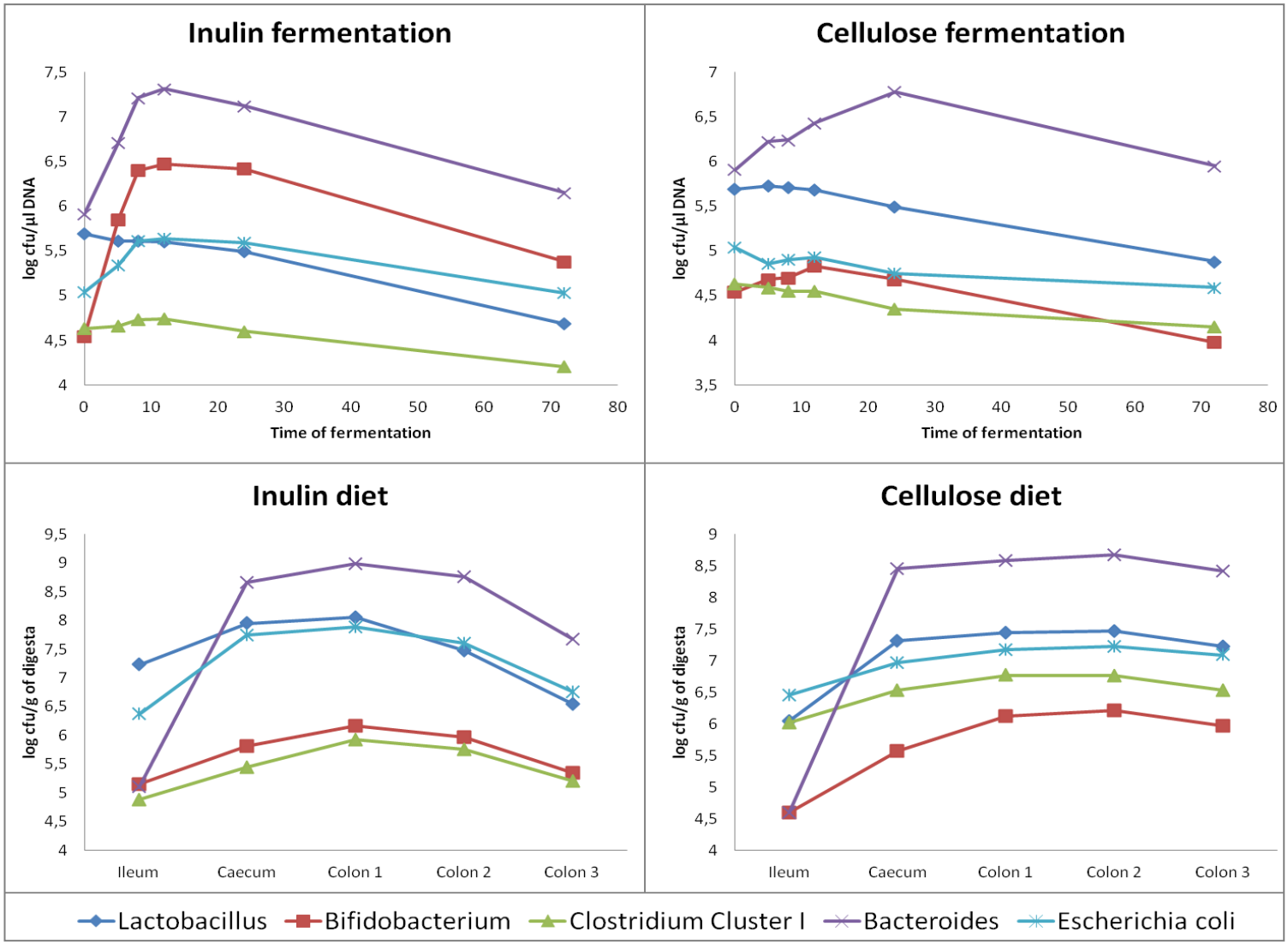
- In vivo validation
  - In vivo trial
    - 3 groups of 8 growing pigs
    - Treatments : (1) Synthetic diet + 5 % Cellulose
      (2) Synthetic diet + 5 % Inulin
      (3) Synthetic diet + 2,5 % Cellulose + 2,5 % Inulin
    - After 15 and 18 days : faeces for in vitro trial
    - After 21 days : slaughtering (digestive tract content sampling)
  - In vitro trial
    - On days 15 and 18 with faeces of the in vivo trial (inoculum)
    - 2 substrates tested : Inulin and Cellulose
    - Fermentation broth collected after 0, 5, 8, 12, 24 and 72h


Fresh faeces (5 %)
Nutritive buffer of Menke
30 ml
200 mg
Substrate: Inulin or Cellulose
Incubation (39°C, gentle agitation)
Fermentation kinetics
ANKOM
GAS PRODUCTION SYSTEM
Broth sampling after 0, 5, 8, 12, 24 and 72h

Context

In *vitro*

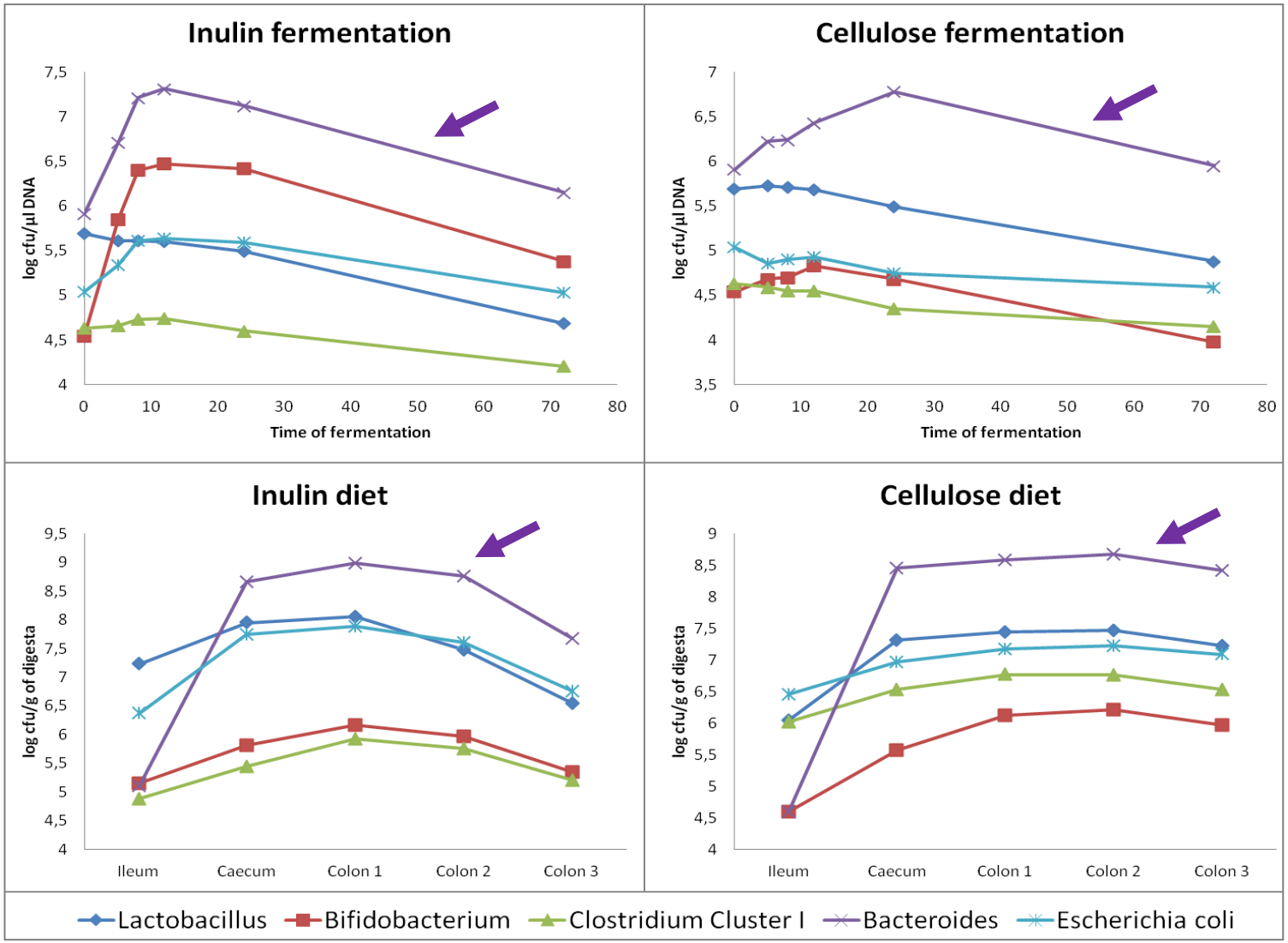
In *vivo*



Context

In *vitro*

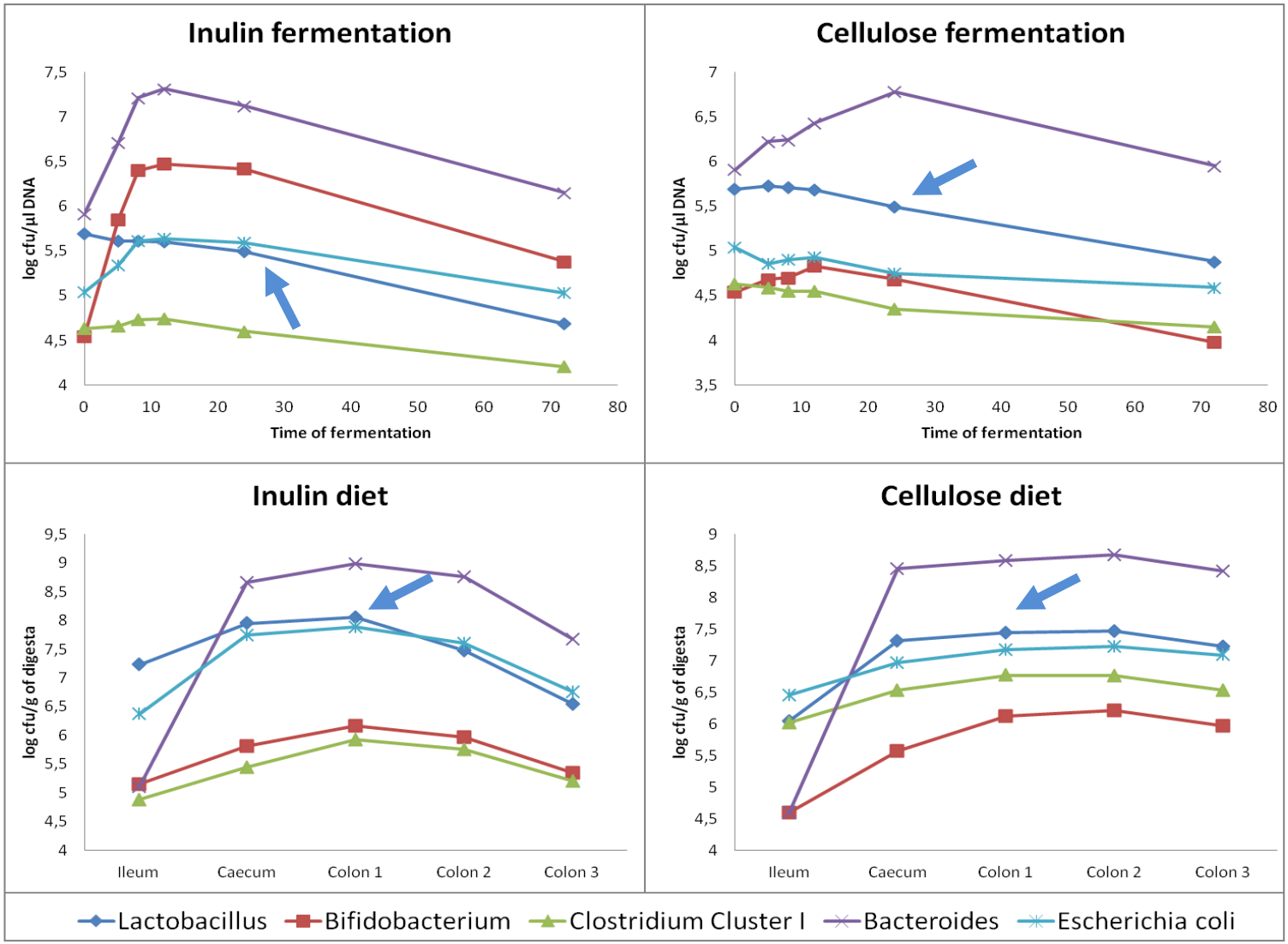
In *vivo*



Context

In *vitro*

In *vivo*



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- In vitro models
  - Different models : simple to complex, cheap to expensive
  - Simple batch fermentation model
    - Screening
- In vivo validation
    - Inulin and cellulose fermentation

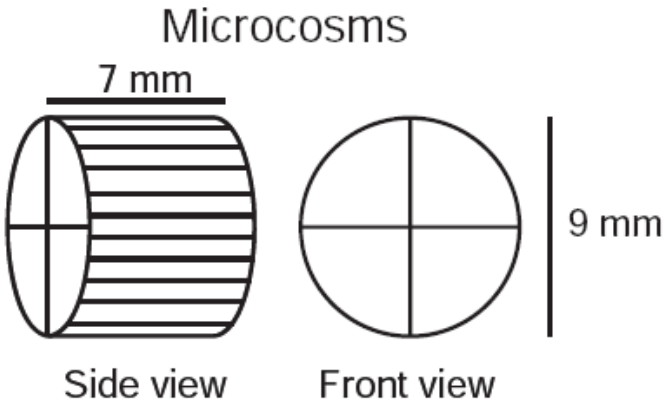
➔ **Descrepancies between in vivo and in vitro observations**

Materials and methods

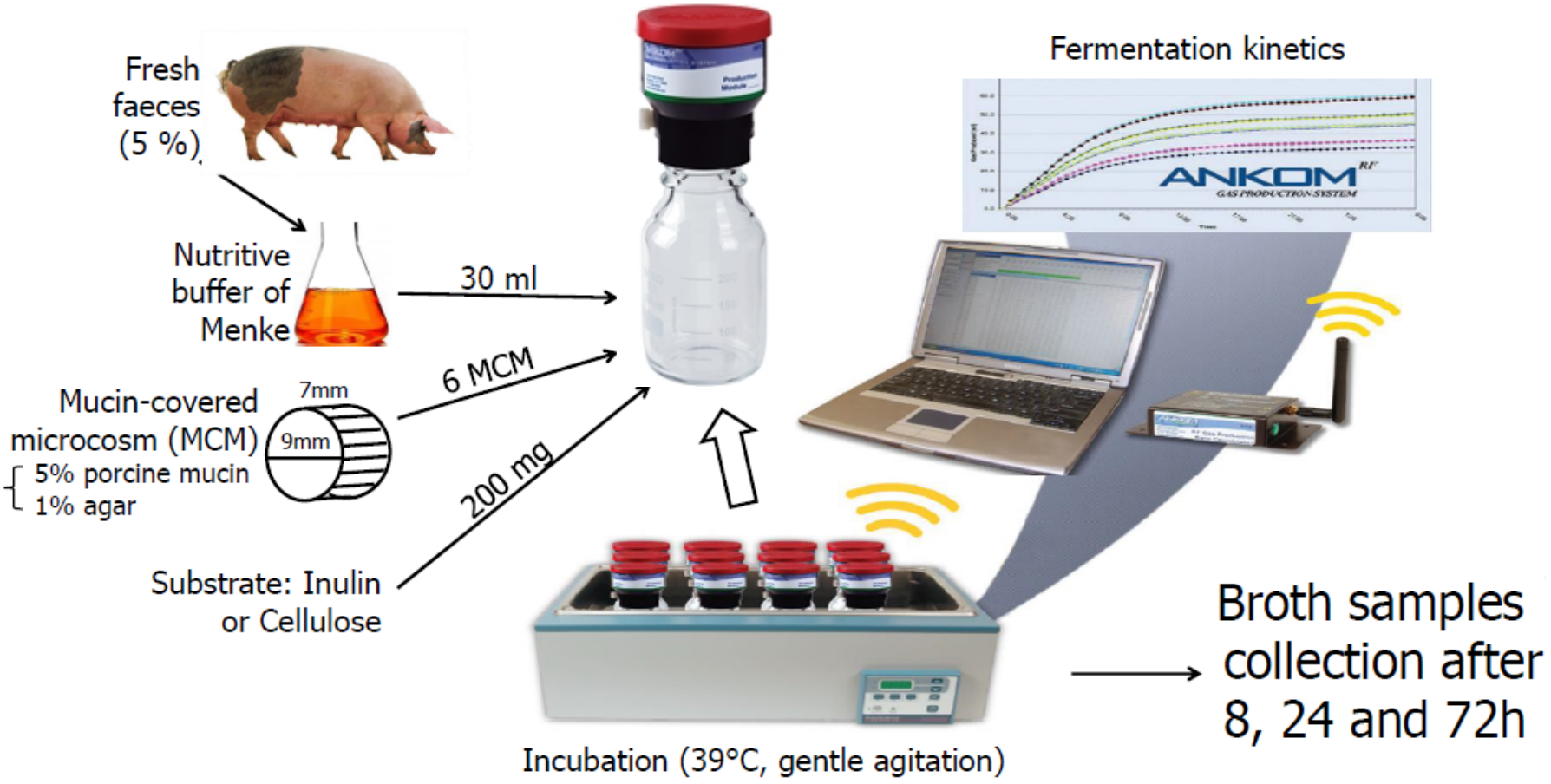
- Mucus Carriers (microcosms)

  (According to Van den Abbeele *et al*., 2012. Microbial Biotechnology 5, 106-115)

  - Coated with 1 % agar and 5 % pig stomach mucin (Sigma)

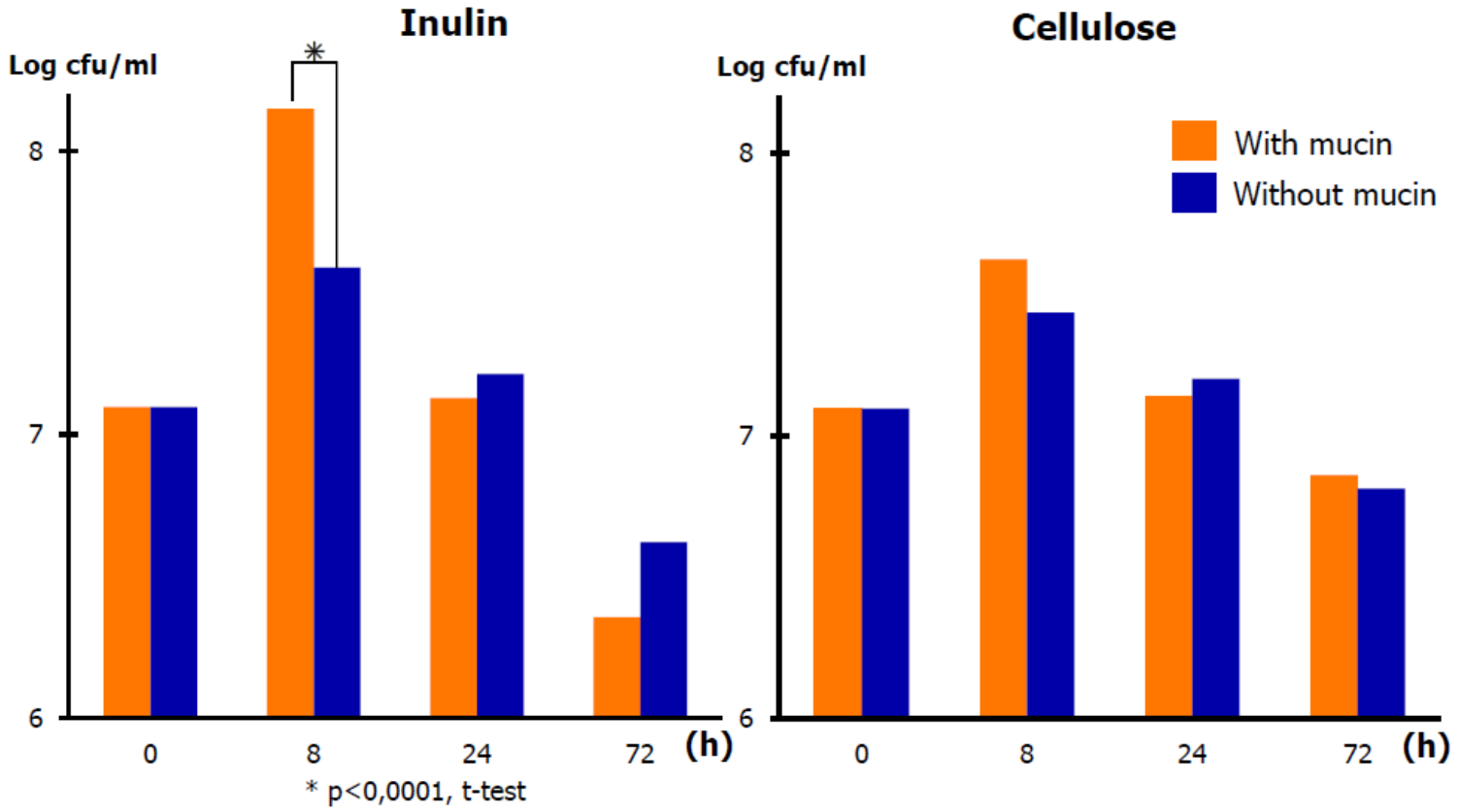


Material and Methods



Results

- qPCR : Lactobacillus in fermentation broth with or without mucus

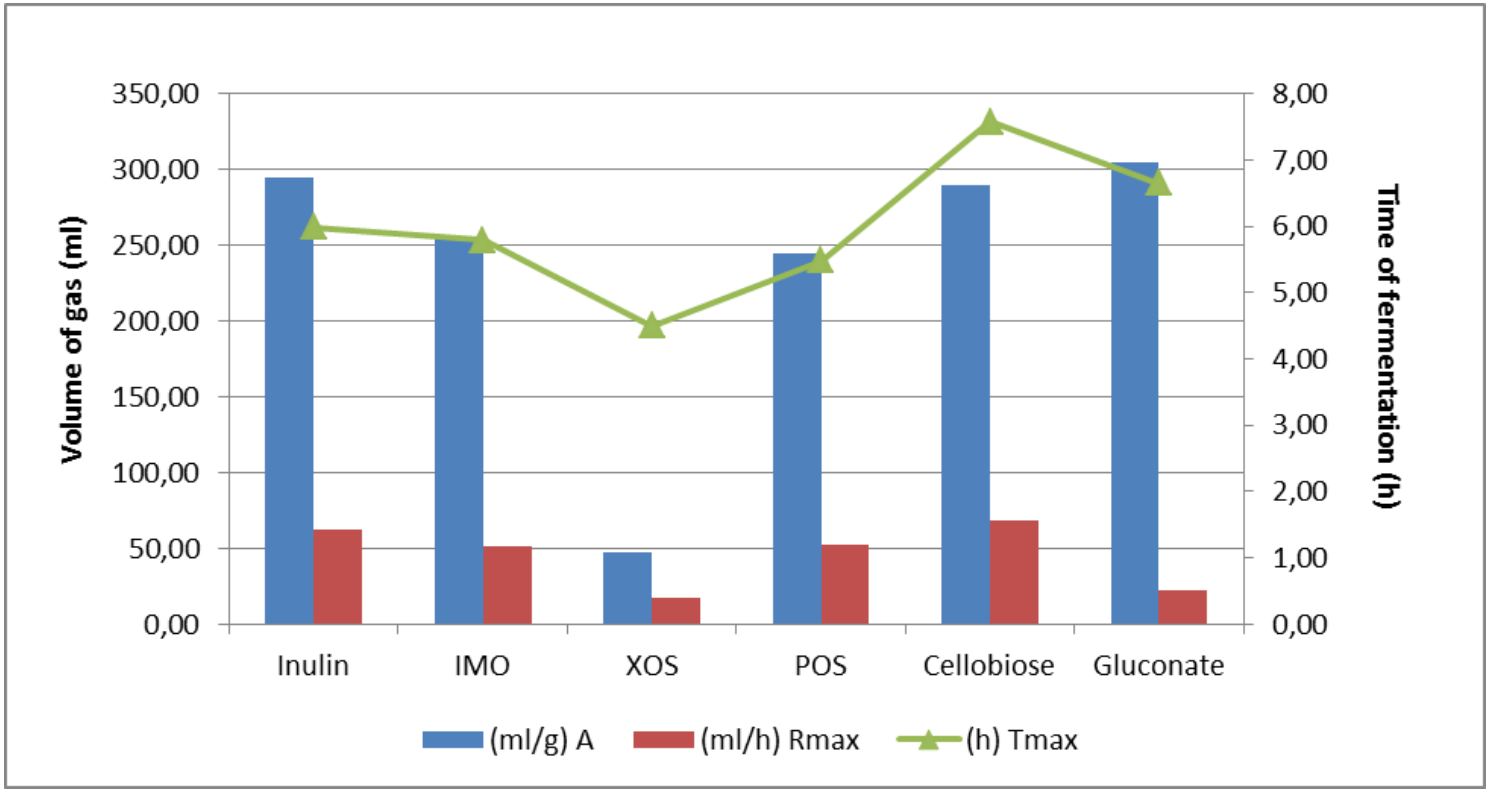


Materials and methods

- In vitro CHO Screening
  - Inulin (positive control)
  - IMO
  - POS
  - Gluconic acid
  - Cellobiose
  - XOS
- Analyses
  - Fermentation kinetics (48h)
  - Fermentation broth after 0, 8 and 48h
    - qPCR : Lactobacillus, Bifidobacteria, Colostridium Cl 1, Bacteroïdes
    - HPLC : SCFA (Acetate, Propionate, Butyrate) and BCFA

Results

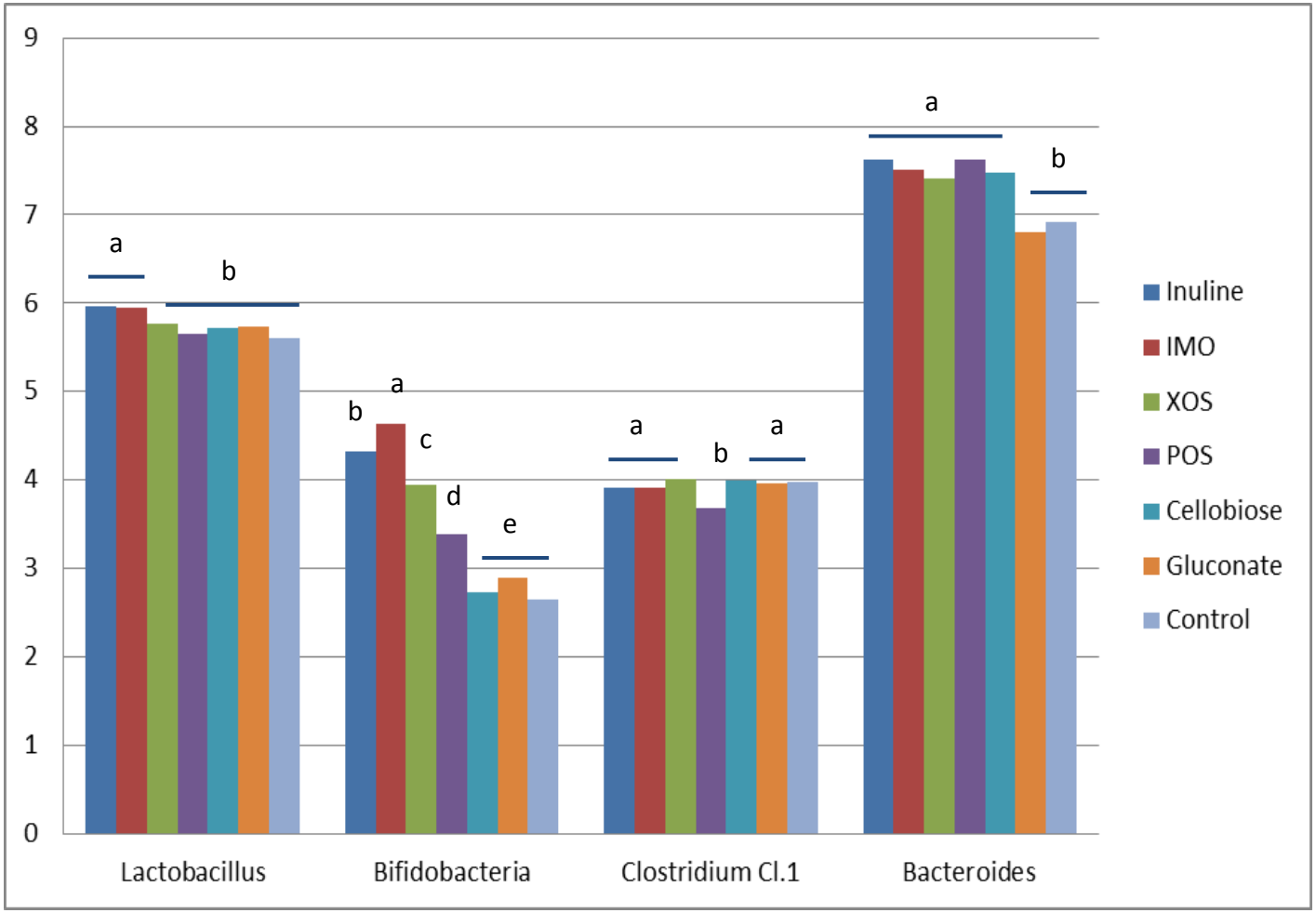
- Fermentation Kinetics (Groot et al. 1996)



A = gas production/g of substrate, Rmax = max rate of gas production/h, Tmax = Time at which Rmax is reached

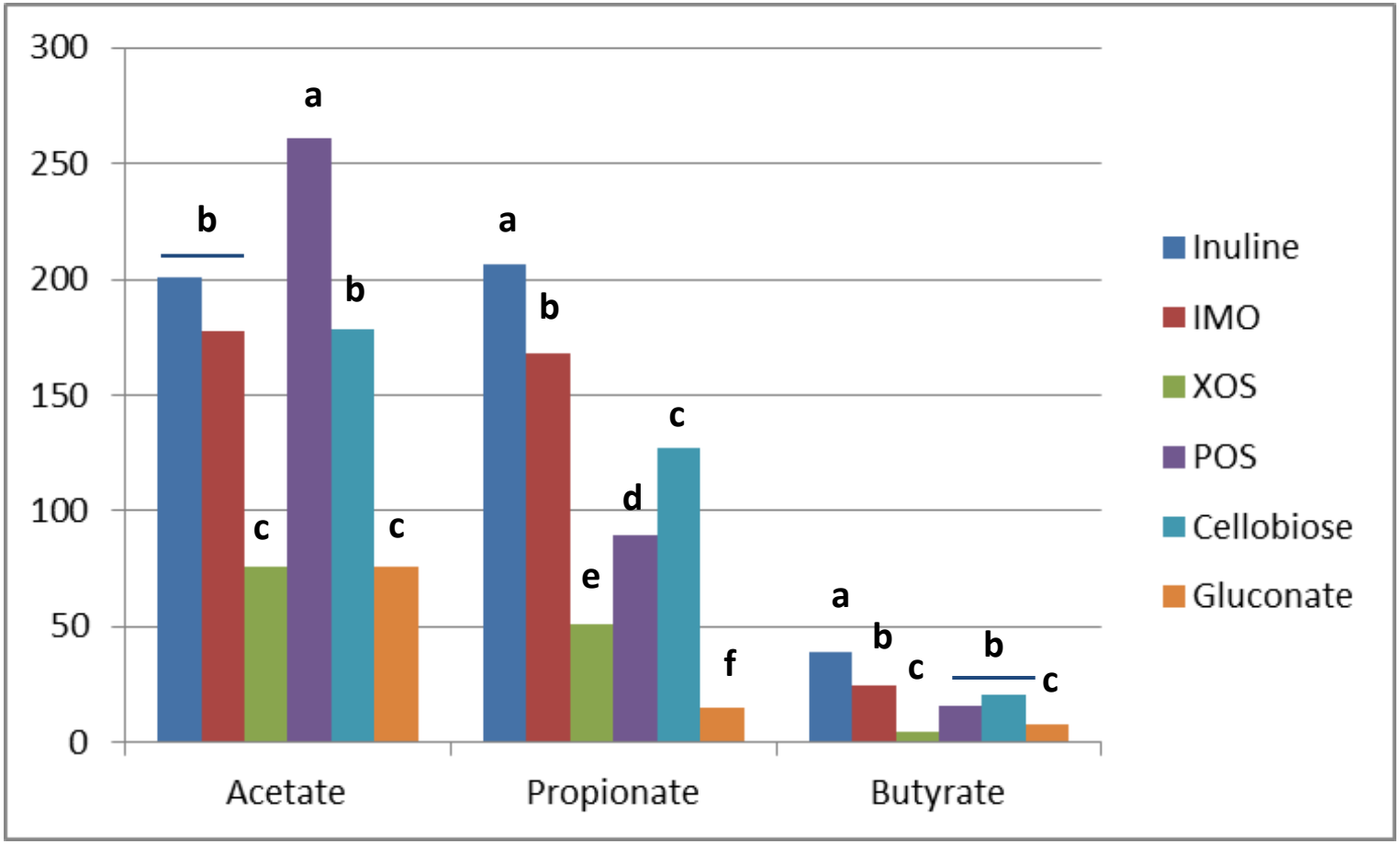
Results

- Microflora



Results

- SCFA



Conclusions

- IMO shows the best prebiotic potential with inulin
  - Increase of Lactobacillus and Bifidobacteria
  - High butyrate production
- POS shows an inhibitive effect on clostridium Cl 1
- Cellobiose shows an interesting ratio of butyrate

➔ Selection of IMO for in vivo studies

➔ Test combinations of different CHO

Thank you for your attention

- Acknowledgments for financial support
  - Service Public of Wallonia SPW – DGO6 through project HYDRASANTE
  - National Fund for Scientific Research and Wallonie Brussels International for scientific stays
- Acknowledgments
  - E. Bera and S. Mabille for technical help

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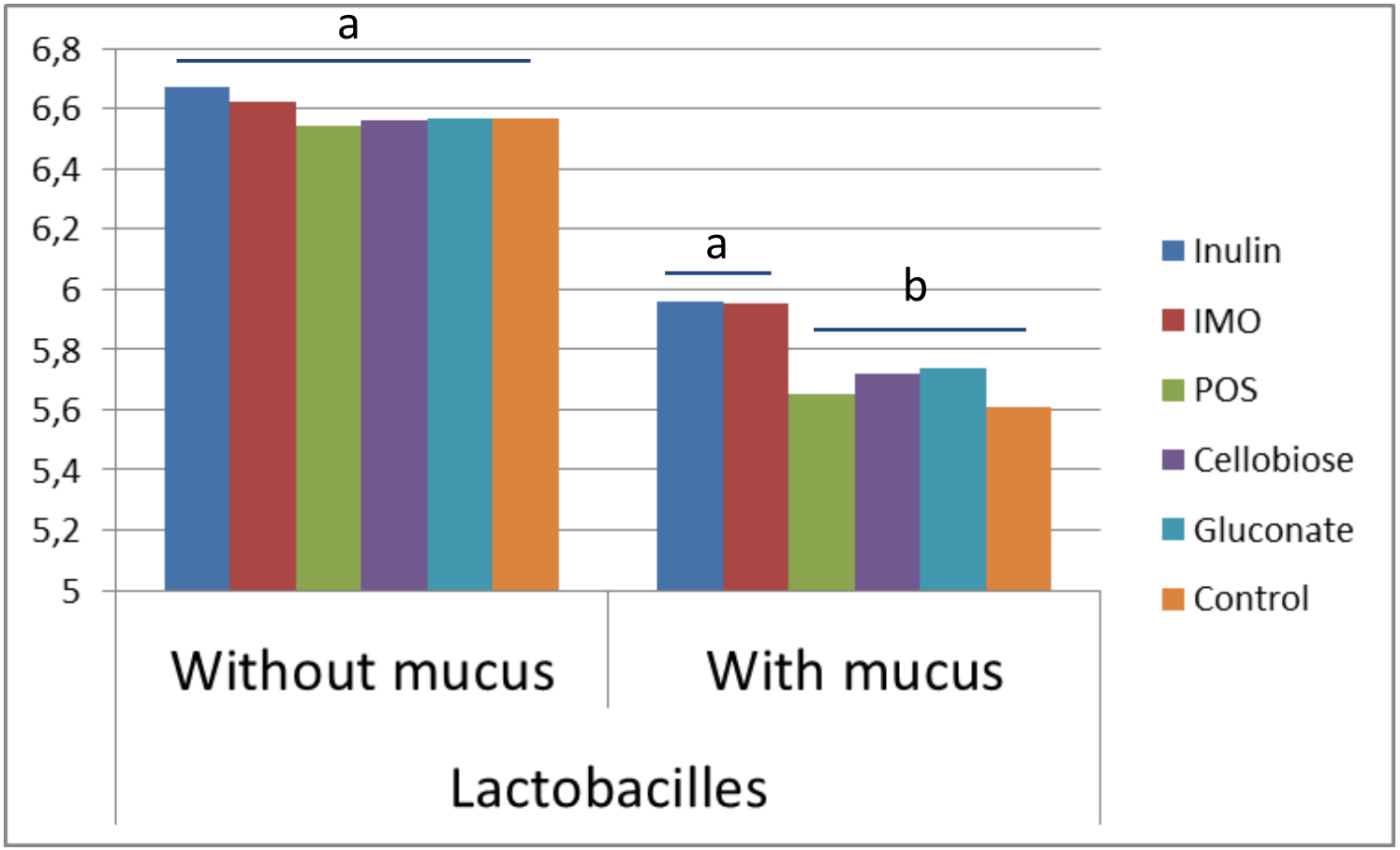
Results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6,2
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5,6
5,4
5,2
5
Without mucus
With mucus
Lactobacilles
Inulin
IMO
POS
Cellobiose
Gluconate
Control