CoMiniGut - a small volume in vitro colon model for the screening of gut microbial fermentation processes.

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The paper by Wiese and co-workers describes the use of a pH-controlled faecal batch fermentation system; CoMiniGut. The system has been used to evaluate the influence of inulin, lactulose, 3'SL and 6'SL on the faecal microbiota composition and metabolic activity. Faecal samples from two healthy adults and two healthy infants were used.

The paper thus describes two topics; the CoMiniGut and the *in vitro* effect of the tested substrates on the faecal microbiota.

The CoMiniGut is a small volume pH-controlled faecal batch fermentation system. It would have been good if the authors had explored the potential of the system more. In the discussion (lines 377-380) the potential of the system is mentioned; samples can be drawn at different time points and the system can be used as a fed-batch system. None of this is, however, explored. Especially the sampling at different time points would show value as it avoids the complexity of needing a multi-chamber system.

The authors might also like to discuss the pro's and con's of using frozen faecal stocks instead of fresh faeces (lines382-383)

On what basis was the medium that was used (lines 174-177) composed; please explain the reasoning behind it.

The fermentations of the test substrates indicate interindividual variation; Figure 3 gives some indication on variation between the replicate simulations but this could have been indicated more also in figures 4 and 5; if not in the figures, then mentioning some measure of variation in the figure legends or the text.

The paper indicates different fermentation patterns between the different tested HMO's. This is an interesting and relevant finding.

The paper should maybe focus more the fermentation results and describe the CoMiniGut in a separate paper, exploring it's potential such as the fed-batch option and especially the possibility to do time series.

Minor points:

As far as I can judge, the English language is of sufficient quality.

The system is pH-controlled with 1M NaOH; does this cause volume change? the working volume of the system is 5ml; how many ml's or μ l of NaOH are added as a ballpark? line 153

The infant donors; were they exclusively breast-fed? Lines 164-165

Lines 180-185 what was the purity of the used test components; were they essentially free (or low content) of mono and di-saccharides; e.g. lactose in the HMO's of the lactitol.

Line 178 Please spell out 'MES'

Line 180 what is the manufacturer of the inulin?

Line 348 bifidobacteria is not with a capital 'b'

Line 419 bifidobacterial is not with a capital 'b'

Line 415 Bifidobacteri<u>um</u> bifidum

Line 416 Bifidobacterium faecale? please check the spelling of the species

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Figure 5 the text above the Figure is referring to adult donors while the Figure legend (lines 594-596) correctly refers to babies.

Figure 6 consider using a log scale? The Figure mentions 3FL while the text above the figure and the Figure legend refer to 6'FL while materials and methods (line 183) refer to 3FL. What is it?