Effects of whole-grain and sugar content in infant cereals on gut microbiota at weaning: a randomized trial

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Figure legends

Figure S1. Rarefaction curves obtained for each of the sequenced samples.
Figure S2. Principal Coordinate Analysis for all samples visualized by time (baseline i.e. 0 weeks vs. 7 weeks).
Figure S3. Principal Coordinate Analysis for samples at baseline (0 weeks) visualized by intervention group. 0-WG: 100% refined flour infant cereals with a high sugar content, 50-WG: 50% whole grain infant cereals with a low sugar content.
Figure S4. Principal Coordinate Analysis for all samples visualized by time (baseline i.e. 0 weeks vs. 7 weeks) and intervention group. 0-WG: 100% refined flour infant cereals with a high sugar content, 50-WG: 50% whole grain infant cereals with a low sugar content.
Figure S5: Correlations between microbial, anthropometric, demographic, and dietary variables. Pearson correlations are expressed for all variables, significant variables are highlighted in red (negatively correlated) or blue (positive correlated) and corrected using the Benjamini–Hochberg procedure. (A) all infants at baseline i.e., 0 weeks (B) 0-WG after 7 weeks of intervention (C) 50-WG after 7 weeks of intervention. 0-WG: 100% refined flour infant cereals with a high sugar content, 50-WG: 50% whole grain infant cereals with a low sugar content.
### Tables

**Supplemental material**

Supplemental table 1. CONSORT 2010 checklist of information to include when reporting a randomized trial

| Section/Topic                  | Item No | Checklist item                                                                 | Reported on page No |
|--------------------------------|---------|----------------------------------------------------------------------------------|---------------------|
| **Title and abstract**         | 1a      | Identification as a randomised trial in the title                               | 1                   |
|                                | 1b      | Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts) | 1                   |
| **Introduction**               |         |                                                                                  |                     |
| Background and objectives      | 2a      | Scientific background and explanation of rationale                              | 1                   |
|                                | 2b      | Specific objectives or hypotheses                                               | 1                   |
| **Methods**                    |         |                                                                                  |                     |
| Trial design                   | 3a      | Description of trial design (such as parallel, factorial) including allocation ratio | 3 (+reported elsewhere) |
|                                | 3b      | Important changes to methods after trial commencement (such as eligibility criteria), with reasons | Not applicable |
| Participants                   | 4a      | Eligibility criteria for participants                                           | 3 (+reported elsewhere) |
|                                | 4b      | Settings and locations where the data were collected                            | 3                   |
| Interventions                  | 5       | The interventions for each group with sufficient details to allow replication, including how and when they were actually administered | 3+6 (Table 1)       |
| Outcomes                       | 6a      | Completely defined pre-specified primary and secondary outcome measures, including how and when they were assessed | 3                   |
|                                | 6b      | Any changes to trial outcomes after the trial commenced, with reasons           | Not applicable      |
| Sample size | 7a | How sample size was determined | 3 (+reported elsewhere) |
|-------------|----|-------------------------------|------------------------|
|             | 7b | When applicable, explanation of any interim analyses and stopping guidelines | Not applicable |

**Randomisation:**
- **Sequence generation**
  - 8a Method used to generate the random allocation sequence | 3 |
  - 8b Type of randomisation; details of any restriction (such as blocking and block size) | 3 |

**Allocation concealment mechanism**
- 9 Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned | 3 |

**Implementation**
- 10 Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions | 3 |

**Blinding**
- 11a If done, who was blinded after assignment to interventions (for example, participants, care providers, those assessing outcomes) and how | 3 |
- 11b If relevant, description of the similarity of interventions | Not applicable |

**Statistical methods**
- 12a Statistical methods used to compare groups for primary and secondary outcomes | 3+4+5 |
- 12b Methods for additional analyses, such as subgroup analyses and adjusted analyses | 3+4+5 |

**Results**
- **Participant flow (a diagram is strongly recommended)**
  - 13a For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome | 5 + Figure 1 |
  - 13b For each group, losses and exclusions after randomisation, together with reasons | 5 + Figure 1 |

- **Recruitment**
  - 14a Dates defining the periods of recruitment and follow-up | 5 |
  - 14b Why the trial ended or was stopped | Not applicable |

- **Baseline data**
  - 15 A table showing baseline demographic and clinical characteristics for each group | 7 (Table 2) |

- **Numbers analysed**
  - 16 For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups | Not applicable (no missing variables for the final subset) |

- **Outcomes and estimation**
  - 17a For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval) | 8+9 (Table 4 & 5) |
  - 17b For binary outcomes, presentation of both absolute and relative effect sizes is recommended | 8+9 (Table 4 & 5) |
| Section                  | Page | Description                                                                                                                                                                                                 | Page(s) |
|-------------------------|------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------|
| Ancillary analyses      | 18   | Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory analyses.                                                                 | 5-11    |
| Harms                   | 19   | All important harms or unintended effects in each group (for specific guidance see CONSORT for harms).                                                                                                | Not applicable |
| Discussion              |      |                                                                                                                                                                                                             |         |
| Limitations             | 20   | Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses.                                                                                          | 13      |
| Generalisability        | 21   | Generalisability (external validity, applicability) of the trial findings.                                                                                                                                    | 13      |
| Interpretation          | 22   | Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence.                                                                                               | 11-14   |
| Other information       |      |                                                                                                                                                                                                             |         |
| Registration            | 23   | Registration number and name of trial registry.                                                                                                                                                             | 14      |
| Protocol                | 24   | Where the full trial protocol can be accessed, if available.                                                                                                                                               | 2 (ref 30) |
| Funding                 | 25   | Sources of funding and other support (such as supply of drugs), role of funders.                                                                                                                             | 14      |

CONSORT checklist derived from [www.consort-statement.org](http://www.consort-statement.org)
Supplemental table 2. Frequency of infections and complications

| Study visit week | Intervention group | 0-WG | 50-WG | p-value |
|------------------|--------------------|------|-------|---------|
|                  | n=18               | n=25 |       |         |
| Study visit week 1 |                   |      |       |         |
| No episodes      | 14                 | 22   | 0.405 |
| Infections, n   |                    |      |       |         |
| - Upper respiratory tract infections | 3     | 2    |       |
| - Other infections including chicken pox | 0     | 0    |       |
| Study visit week 4 |                   |      |       |         |
| No episodes      | 16                 | 21   | 0.454 |
| Infections, n   |                    |      |       |         |
| - Upper respiratory tract infections | 2     | 2    |       |
| - Lower respiratory tract infections | 0     | 2    |       |
| Study visit week 7 |                   |      |       |         |
| No episodes      | 15                 | 18   | 0.419 |
| Infections, n   |                    |      |       |         |
| - Upper respiratory tract infections | 1     | 4    |       |
| - Other infections including chicken pox | 0     | 2    |       |
| - Gastrointestinal infections | 2     | 0    |       |
| - Non-infectious gastrointestinal symptoms (e.g. constipation, vomiting) | 0     | 1    |       |

Data are presented as absolute count, 0-WG: 100% refined flour infant cereals with a high sugar content, 50-WG: 50% whole grain infant cereals with a low sugar content. Data were analyzed using independent t-tests.
Supplemental table 3. Frequency of treatments

| Study visit week | 0-WG n=18 | 50-WG n=25 | p-value |
|------------------|-----------|------------|---------|
| 1                |           |            |         |
| No treatments    | 16        | 22         | 0.929   |
| Treatments, n    |           |            |         |
| - Antihistamines, anti-inflammatory, and bronchodilators | 2 | 3 |         |
| Study visit week 4 |           |            |         |
| No treatments    | 16        | 21         | 0.301   |
| Treatments, n    |           |            |         |
| - Antihistamines, anti-inflammatory, and bronchodilators | 1 | 4 |         |
| - Antipyretics and analgesics | 1 | 0 |         |
| Study visit week 7 |           |            |         |
| No treatments    | 16        | 17         | 0.417   |
| Treatments, n    |           |            |         |
| - Antihistamines, anti-inflammatory, and bronchodilators | 1 | 3 |         |
| - Antipyretics and analgesics | 0 | 2 |         |
| - Probiotics     | 1         | 0          |         |
| - Laxatives      | 0         | 1          |         |
| - Inhaled or topical corticosteroids | 0 | 1 |         |

Data are presented as absolute count, 0-WG: 100% refined flour infant cereals with a high sugar content, 50-WG: 50% whole grain infant cereals with a low sugar content. Data were analyzed using independent t-tests.