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Temporal changes in the fermentation characteristics, bacterial community structure and the functionality of the predicted metagenome of a batch fermenter medium containing the upper gastrointestinal enzyme resistant fraction of white sorghum (*Sorghum bicolor* L. Moench)

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Highlights

- Raw and autoclaved-freeze-dried sorghum exhibit similar colonic fermentation trends.
- Abundance of key members of *Lactobacillaceae* is different between the two groups.
- Different temporal organic acid concentrations manifest differences in metabolism.
- Predicted abundance of some carbohydrate metabolic pathways is different in between.
- Cooking-cooling methods can modify the functionality of resistant starch in daily food.

Abstract

Sorghum is a potential prebiotic ascribed to the high native resistant starch (RS) content. Our previous studies on raw sorghum have revealed prominent amino acid fermentation despite the high RS content. Interestingly, autoclaved-freeze-dried sorghum fed rats exhibited beneficial microbial and biochemical profiles. Having a keen interest to reciprocally scrutinize the underlying mechanisms behind these contrasting outcomes, we used an *in vitro* porcine batch fermentation model. The fermentable substrates in raw and autoclaved-freeze-dried (three cycles) sorghum (AC)

temporal trends in fermentation characteristics of autoclaved-frozen (three cycles) sorghum and its raw counterpart, with respect to the changes in the bacterial community structure and the predicted functionality of the metagenome. We hypothesized that autoclaved-freeze-dried sorghum and respective native counterpart might influence different microbial taxa and exhibit different fermentation characteristics under *in vitro* conditions.

Section snippets

Preparation of sorghum enzyme resistant fraction

Upon native RS content determination using Megazyme RS assay (K-RSTAR 08/11, Wicklow, Ireland), sorghum grain sample provided by the U. S. Grains Council (Tokyo, Japan) was subjected to three autoclave-freeze cycles (grains: water = 1:2; 121 °C, 20 min; −30 °C overnight) (Pelpolage et al., 2020). The milled raw sample and freeze-dried autoclaved-frozen sample after the third cycle were subjected to *in vitro* gastrointestinal digestion according to the modified Lebbet method as follows (Karimi et ...

Organic acid production and pH

Temporal production trends of major organic acids were evaluated in the three groups (Fig. 1). Interestingly, there was a soaring ($p < 0.05$) content of lactate in both sorghum groups (RAW and AC) by 6 h compared to CON, where RAW group reported almost 120 μmol/mL of fermentation medium, with 80 and < 5 μmol/mL in AC and CON, respectively. This surge of lactate production in the two sorghum groups was speculated to lead to a drastic ($p < 0.05$) drop in the pH in the medium (Fig. 1), which was...

Conclusions

This study aimed to reciprocally scrutinize the temporal colonic degradation characteristics of raw and autoclaved-freeze-dried sorghum residues after *in vitro* gastrointestinal digestion in a retrospective manner. An early lactate surge (6 h) and gradual increment of SCFAs concomitantly with the diminished lactate content (12–48 h) were characteristic to the two sorghum groups. However, the marked differences in the concentration peaks of lactate between the two groups, suggested potential...

Uncited references

Flint, 2020, Garber et al., 2021, Vince and Burridge, 1980....

CRediT authorship contribution statement

Samanthi W. Pelpolage: Conceptualization, Methodology, Software, Formal analysis, Investigation, Data curation, Writing – original draft, Writing – review & editing, Visualization. **Haruhi Kobayashi:** Formal analysis, Investigation, Data curation, Visualization. **Naoki Fukuma:** Software, Investigation, Resources, Data curation, Writing – review & editing. **Michiyo Hoshizawa:** Conceptualization, Resources, Writing – review & editing. **Tetsuo Hamamoto:** Conceptualization, Resources, Writing – review &...

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper....

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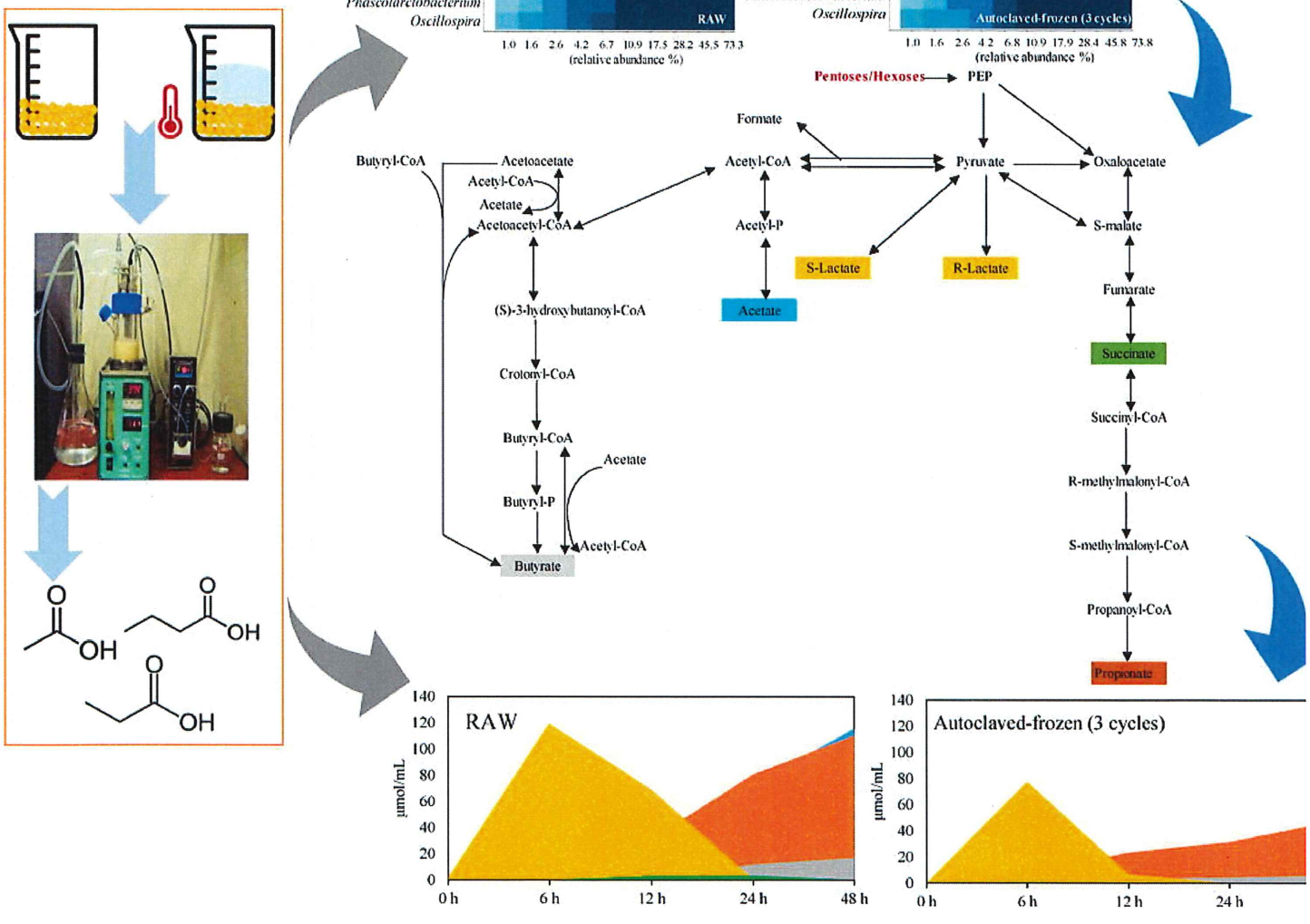
Lactobacillus
Prevotella
Megasphaera
Clostridium
Acidaminococcus
Succinivibrio
Bifidobacterium
Mitsuokella
Phascolarctobacterium
Oscillospira
RAW
1.0 1.6 2.6 4.2 6.7 10.9 17.5 28.2 45.5 73.3
(relative abundance %)
Lactobacillus
Prevotella
Megasphaera
Not Assigned
Acidaminococcus
Succinivibrio
Shutterworthia
Mitsuokella
Phascolarctobacterium
Oscillospira
Autoclaved-frozen (3 cycles)
1.0 1.6 2.6 4.2 6.8 10.9 17.9 28.4 45.8 73.8
(relative abundance %)
Pentoses/Hexoses
PEP
Formate
Pyruvate
Oxaloacetate
Acetyl-CoA
Acetyl-P
Acetate
S-Lactate
R-Lactate
S-malate
Fumarate
Succinate
Succinyl-CoA
R-methylmalonyl-CoA
S-methylmalonyl-CoA
Propanoyl-CoA
Propionate
Butyryl-CoA
Acetoacetate
Acetyl-CoA
Acetate
Acetoacetyl-CoA
(S)-3-hydroxybutanoyl-CoA
Crotonyl-CoA
Butyryl-CoA
Acetate
Butyryl-P
Acetyl-CoA
Butyrate
RAW
µmol/mL
140
120
100
80
60
40
20
0
0 h
6 h
12 h
24 h
48 h
Autoclaved-frozen (3 cycles)
µmol/mL
140
120
100
80
60
40
20
0
0 h
6 h
12 h
24 h