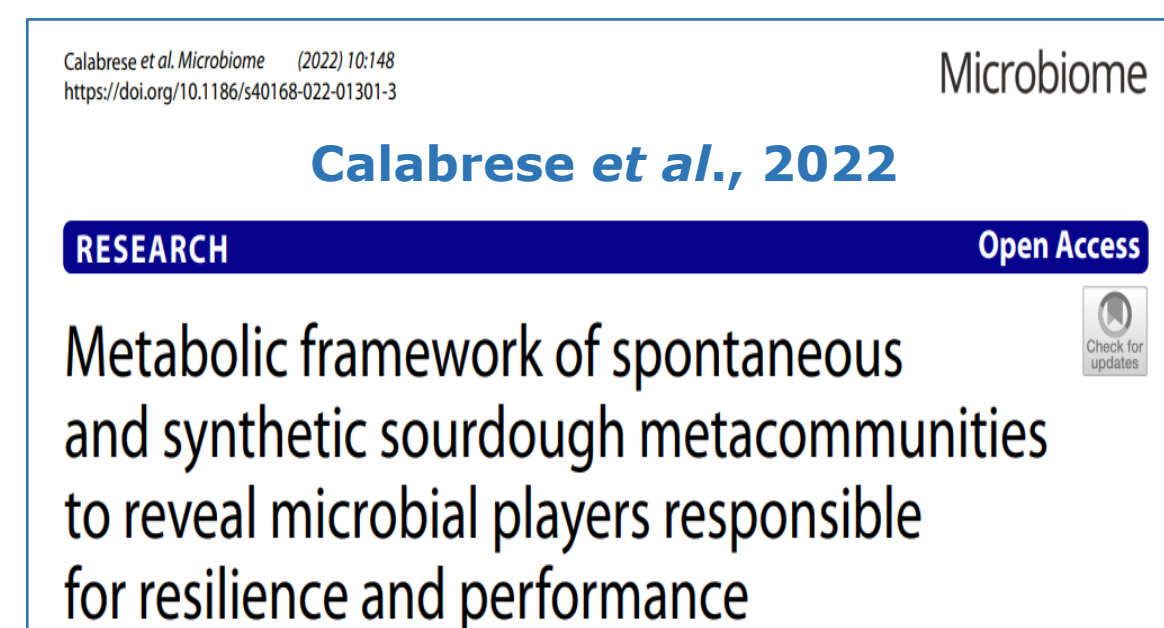


Validation of Sourdough Key Players using De Novo Synthetic Microbial Communities

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1 BACKGROUND



Sourdough is a tractable fermented ecosystem where key microbial players ensure its performance and resilience. Synthetic communities of representative strains offer a more sustainable alternative to traditional starters, for leavened baked goods (Nikoloudaki *et al.*, 2024; Calabrese *et al.*, 2022).

Is the theory and applicative output of 'SDGlobal' (robust sourdough SMC) reproducible by changing dominants and subdominants at species/strain level?

2 MATERIALS & METHODS

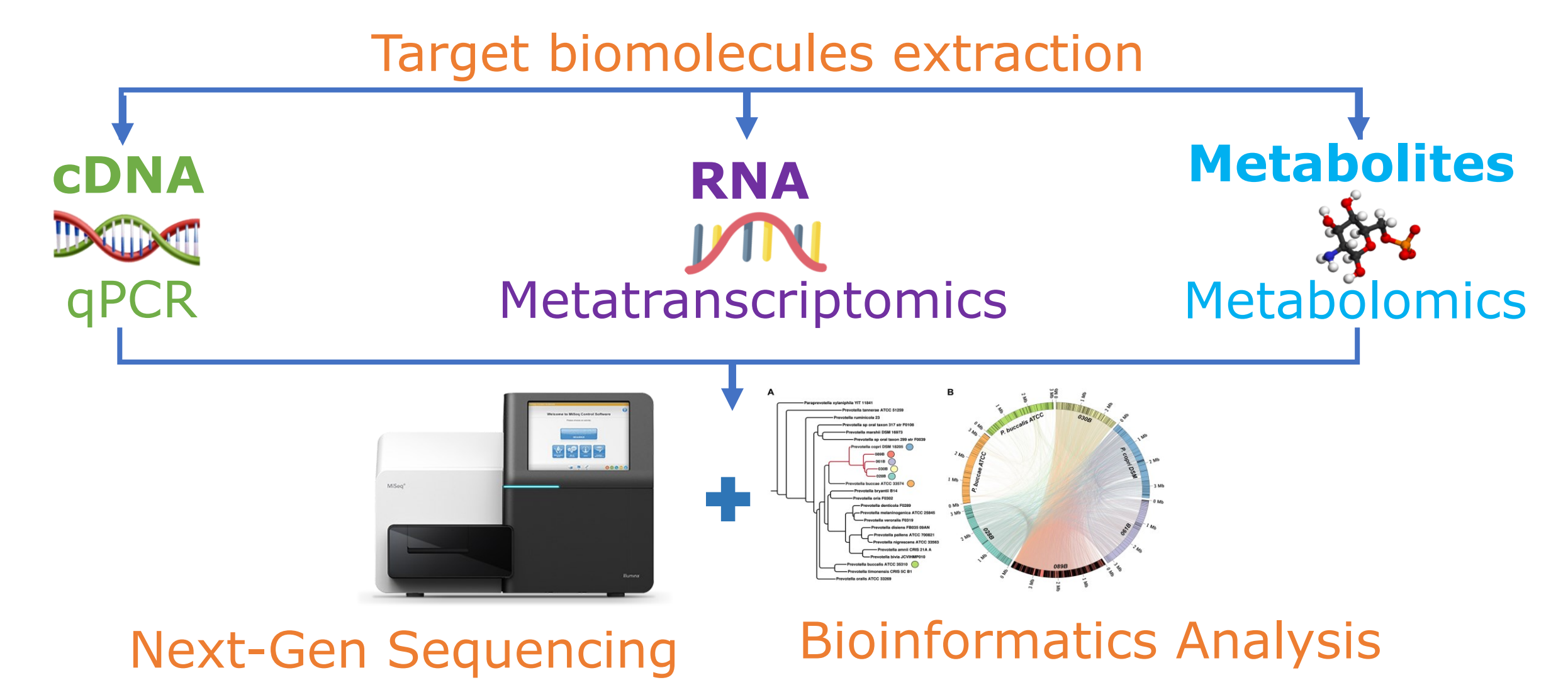
6 strains each: 2 core dominant LAB, 2 sub-dominant LAB, 1 satellite member & 1 core dominant yeast

20 New SMCs Construction

In wheat flour hydrolysate (at mid-exponential (M) & beginning of stationary (BS) phases)

Confirmed by Sanger sequencing

Defined by Growth Kinetics (30 °C)



3 PRELIMINARY RESULTS

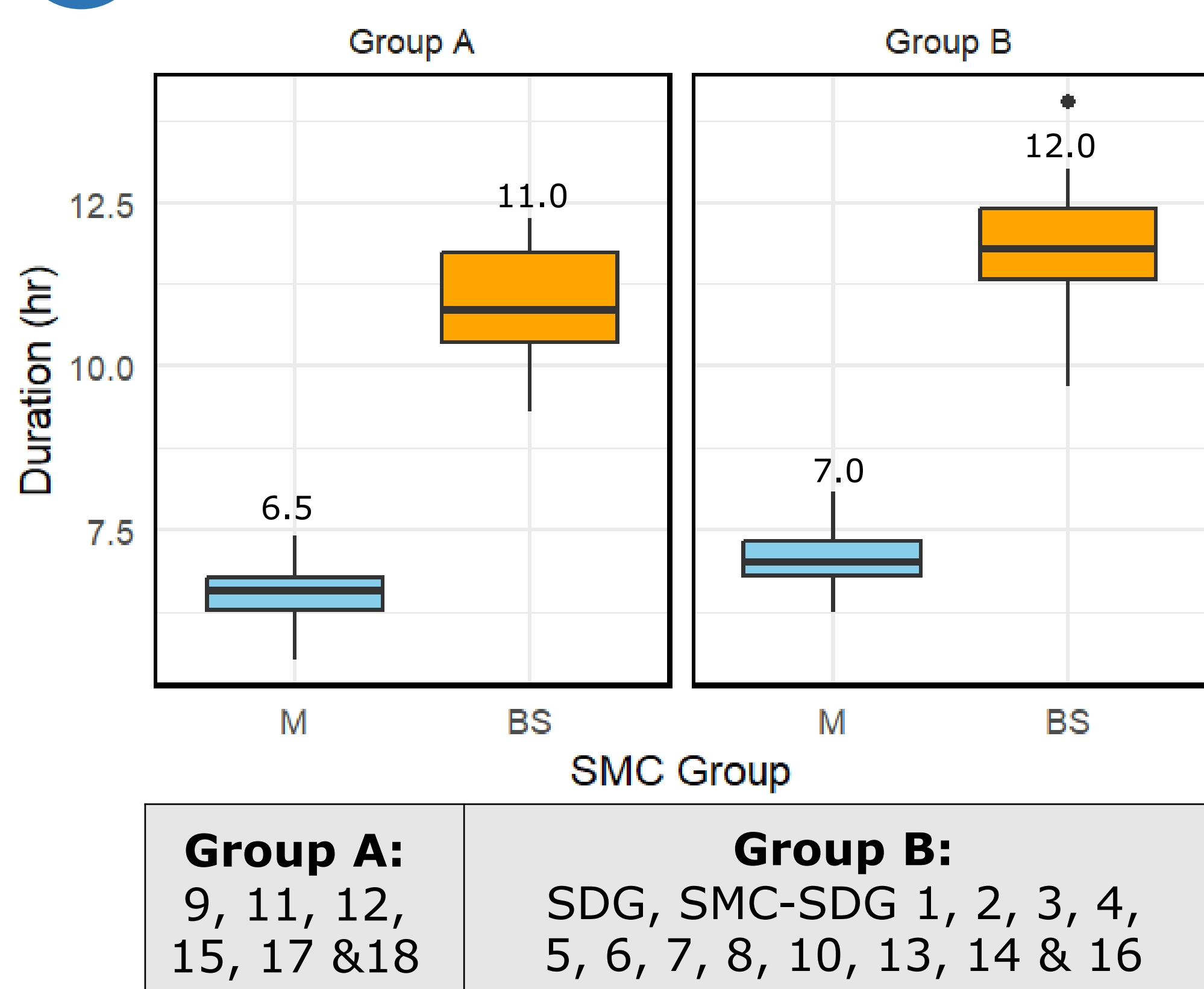


Fig. 1: SMCs Fermentation time for the M & BS phases

Corroborates observations by Calabrese *et al.* (2022), validating the reproducibility and utility of SDGlobal as a reliable control.

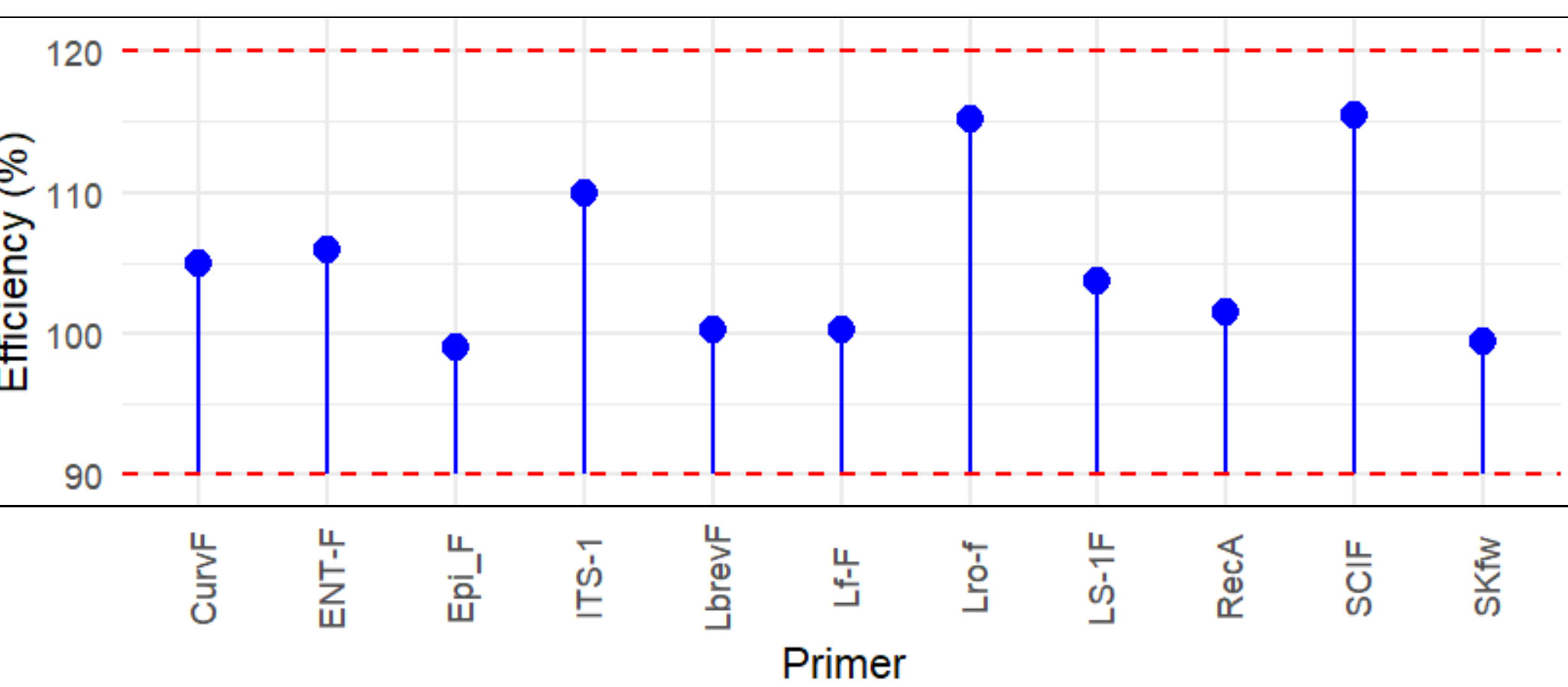


Fig. 2: qPCR efficiencies of the species-specific primers

Primer efficiencies were within the acceptable range of 90-120% (Broeders *et al.*, 2014).

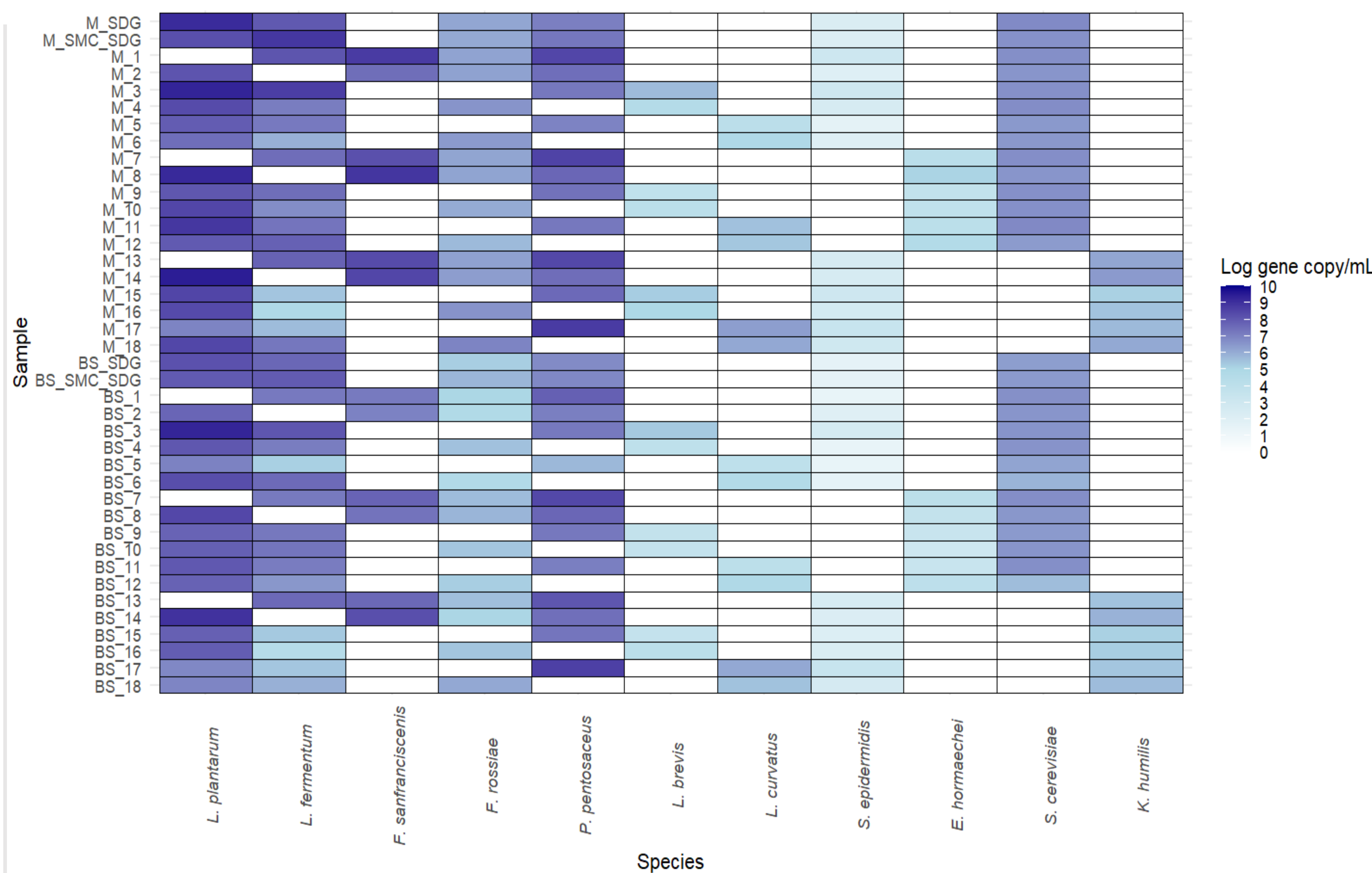


Fig. 3: Heatmap of SMCs absolute abundances at M and BS, expressed in log gene copy/mL

As expected, *Lactiplantibacillus plantarum*, *Limosilactobacillus fermentum*, and *Fructilactobacillus sanfranciscensis* dominated the SMCs. However, *Pediococcus pentosaceus* was more abundant than previously reported. Microbial abundances were higher in the mid-exponential phase (M) compared to the beginning of the stationary phase (BS), suggesting a decline in microbial activity as fermentation progressed (Calabrese *et al.*, 2022). Despite the depletion of individual species, the overall microbial abundance remained relatively stable, indicating potential metabolic redundancy.

4 CONCLUSION & FUTURE WORK

The new SMCs have demonstrated consistent performance in line with the theoretical framework of SDGlobal. Ongoing bioinformatics analyses of metatranscriptomics and metabolomics data will provide insights into their metabolic redundancy and impact on sourdough stability and performance. The subsequent studies include competition experiments and *in-situ* analyses.

5 REFERENCES

- Nikoloudaki O, Aheto F, Di Cagno R, & Gobbetti M. (2024). Synthetic microbial communities: A gateway to understanding resistance, resilience, and functionality in spontaneously fermented food microbiomes. *Food Research International* **192**: 114780.
- Broeders S, Huber I, Grohmann L, Berben G, Taverniers I, Morisset D (2014). Guidelines for validation of qPCR methods. *Trends Food Sci. Technol.* **37**(2): 115-126.
- Calabrese FM, Ameer H, Nikoloudaki O, Celano G, Vacca M, Junior WJ, Gobbetti M (2022). Metabolic framework of spontaneous and synthetic sourdough metacommunities to reveal microbial players responsible for resilience and performance. *Microbiome* **10**: 148.