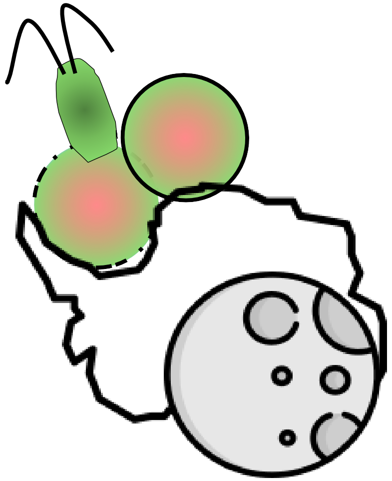
Snow Algae Plasticity and Metabolic Shifts Under Simulated Lunar Photoperiod and Gravity Conditions



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MELiSSA Conference 7-9th October 2025

Culture Collection of algae & protozoa

SAMS

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Introduction – Snow algae are extremophiles

- Cryophilic and cryotolerant microalgae
- Can survive freezing temperatures
- Low nutrient disposition
- Freeze-thaw cycles
- Extended darkness adaptation – winter dormancy
- Hight light irradiances – up to ~2000 PAR (µmol m^{-2} s^{-1}) in the Austral summer

Unique plasticity of Snow Algae

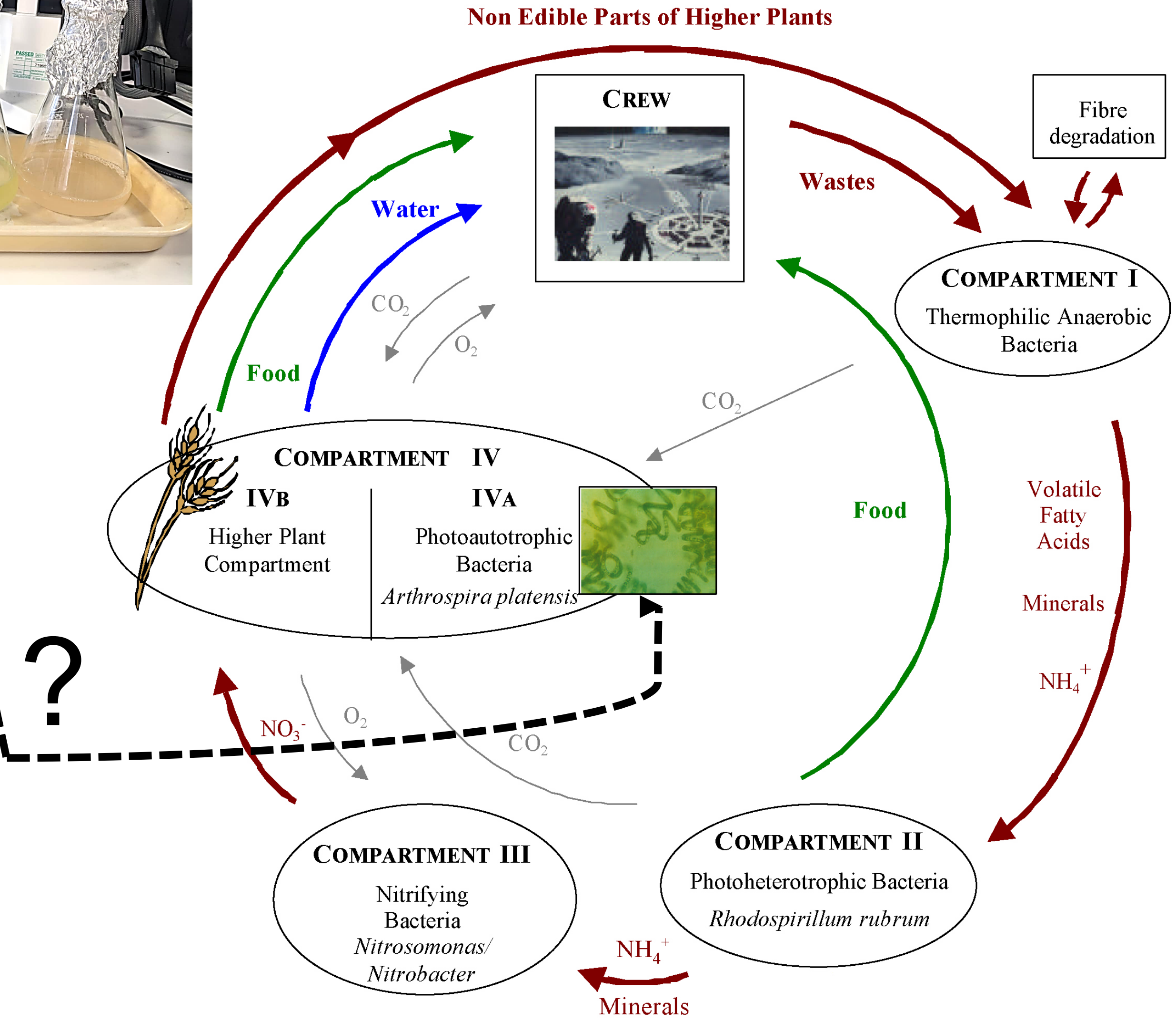
Snow algae blooms – Antarctica. Photo credit M. Davey

Introduction – Potential of snow algae in BLSS

Snow Algae Function

- Cryoprotection & Stress Tolerance
- Nutrient & Biomass Supply
- Gas exchange
- Radiation & UV Protection
- Energy Storage

Photo. Matt Davey



Credit: ESA, MELiSSA project.

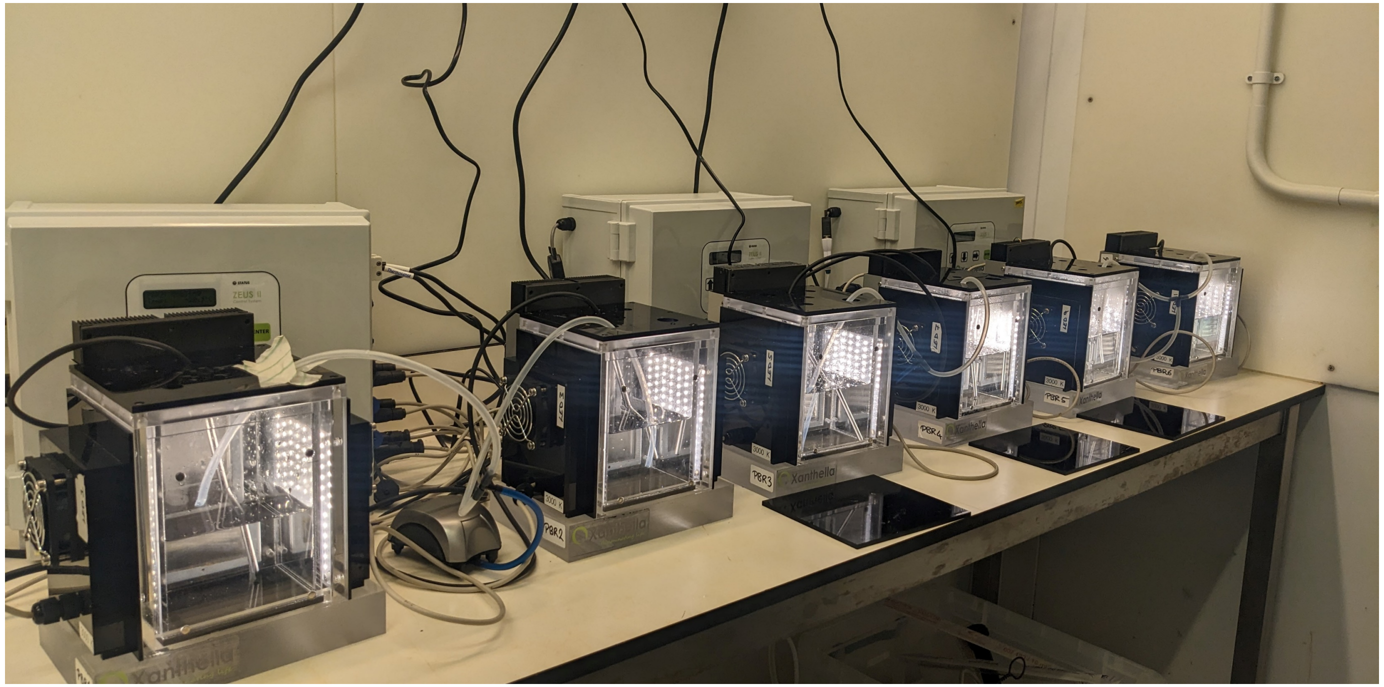
Research Questions

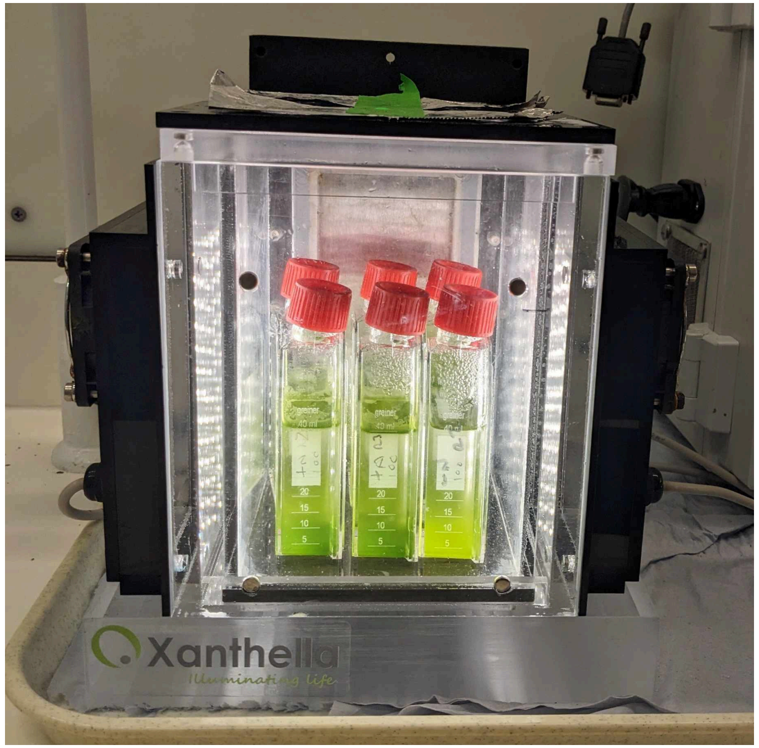
- How snow algae adapt to a simulated lunar photoperiod in conditions
- Snow algae axenically *vs* xenically
- How do they respond to simulated microgravity and partial gravity
- Which metabolic traits underpin their survival and plasticity

Simulated lunar photoperiods

Chlorominima sp. and *Limnomonas* sp. **growth and metabolic shifts under Lunar Light Cycles**

- Simulated Lunar photoperiods → 14:14:14 days (light: dark: light)
- Cultivated in Axenic (only one microorganism) and Xenic (containing other microorganisms) conditions
- 100PAR 4°C, 55mL flasks
- Growth and metabolite content
- Assessing stability, contamination risk, and microbial interactions



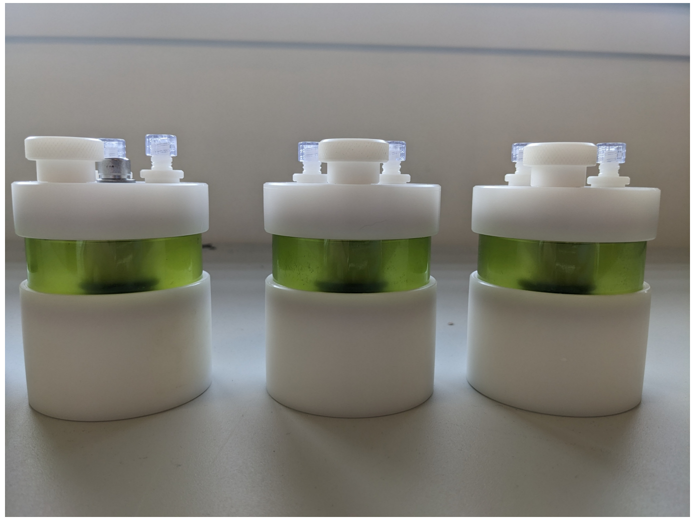


Simulated microgravity and partial gravity

Chlorominima sp. and *Limnomonas sp.* **growth and metabolic shifts under altered gravity**

- Microgravity 0.003 – 0.00006 g
- Partial gravity (Lunar g)   ~0.16 g
- Earth gravity controls 1g

- 16:8h (light: dark), 55mL SLTV
- 100PAR 4°C, 21 days
- End point growth and metabolite content

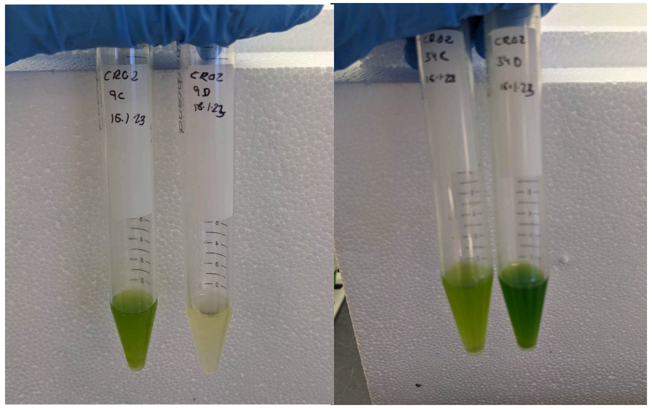


Workflow

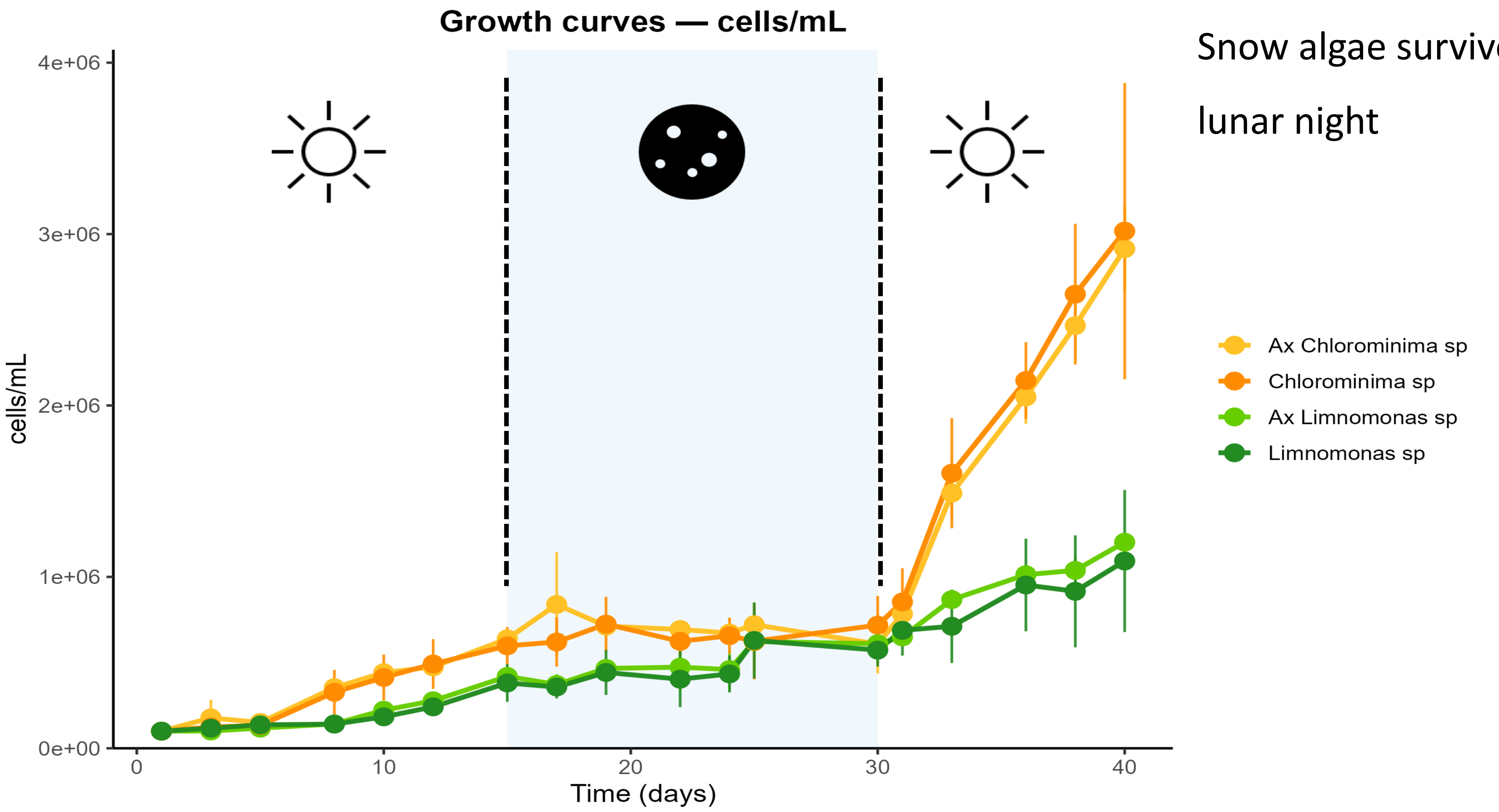
Evaluation

Growth → OD600nm OD750nm
Coulter counter – cells/mL
Microscopic check → ANOVA and Multivariate Data Analysis. MetaboanalysR

Metabolomics → Metabolite & Pigment extractions
MSTFA derviatizations → Semi-targeted metabolomics GC-MS



Simulated lunar photoperiods - Results

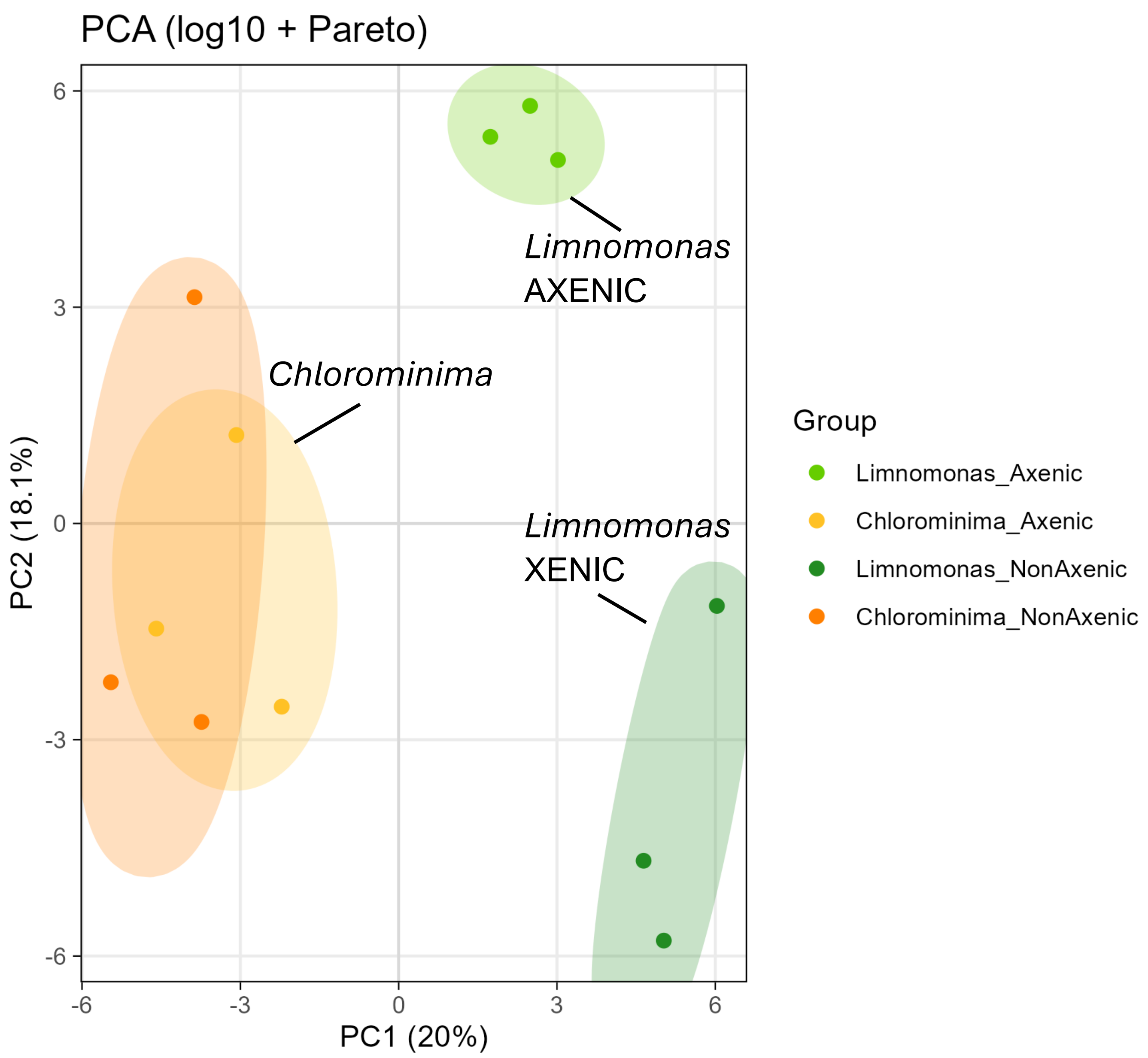


Snow algae survive with extended lunar night

Mean proportion of algal growth over time (in days) across four experimental conditions. The shaded blue region highlights the dark phase (15–30 days). Values are mean ±SD (n = 3, sum of replicates)

Simulated lunar photoperiods - Results

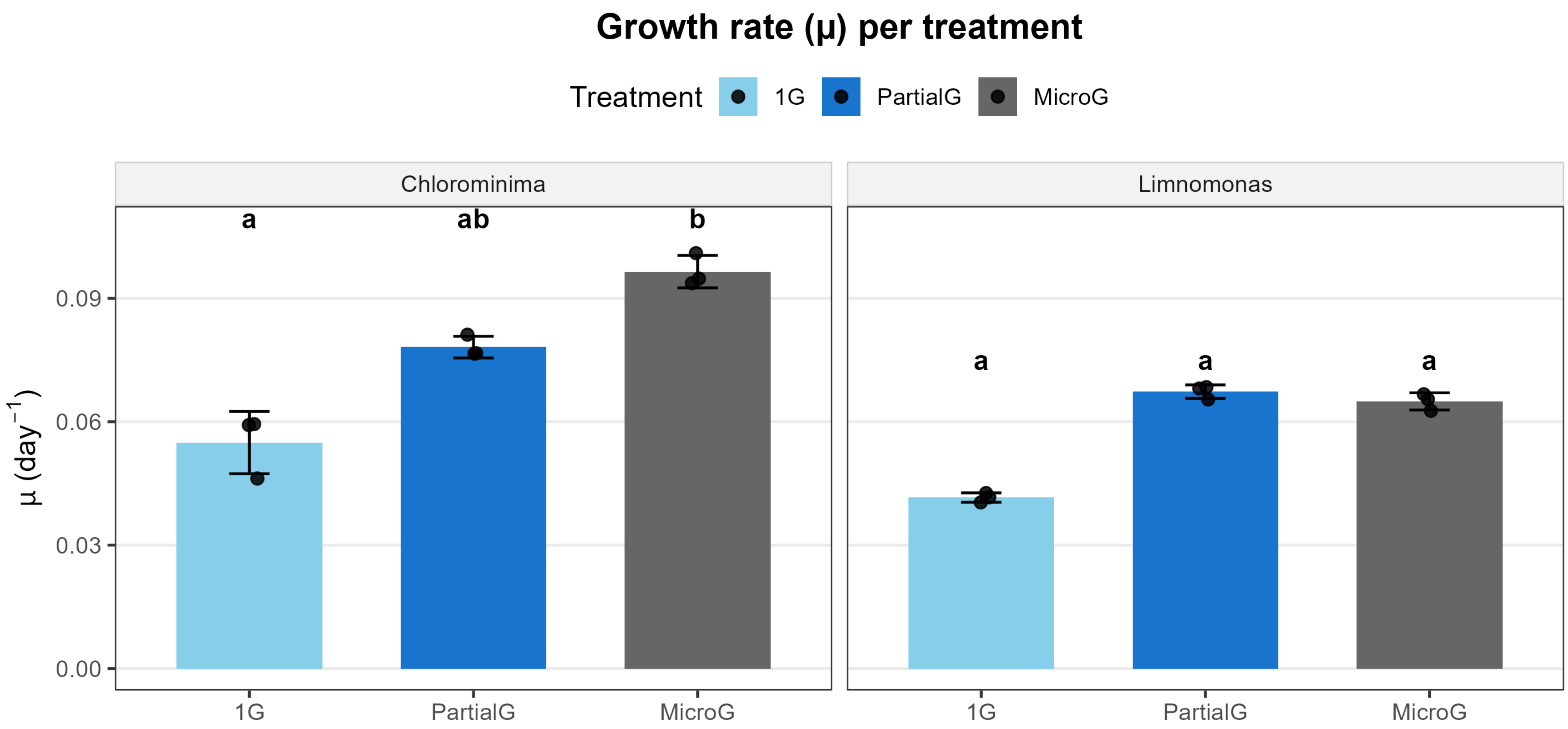
Snow algae show distinct metabolic fingerprints across species & culture types



PCA scores of GC-MS peak intensities of light orange (circle n = 3), dark orange (circle n = 3), light green (circle n = 3) and dark green (circle n = 3) snow algae isolates in different culturing conditions. Also illustrated are the 95% confidence intervals for each group.

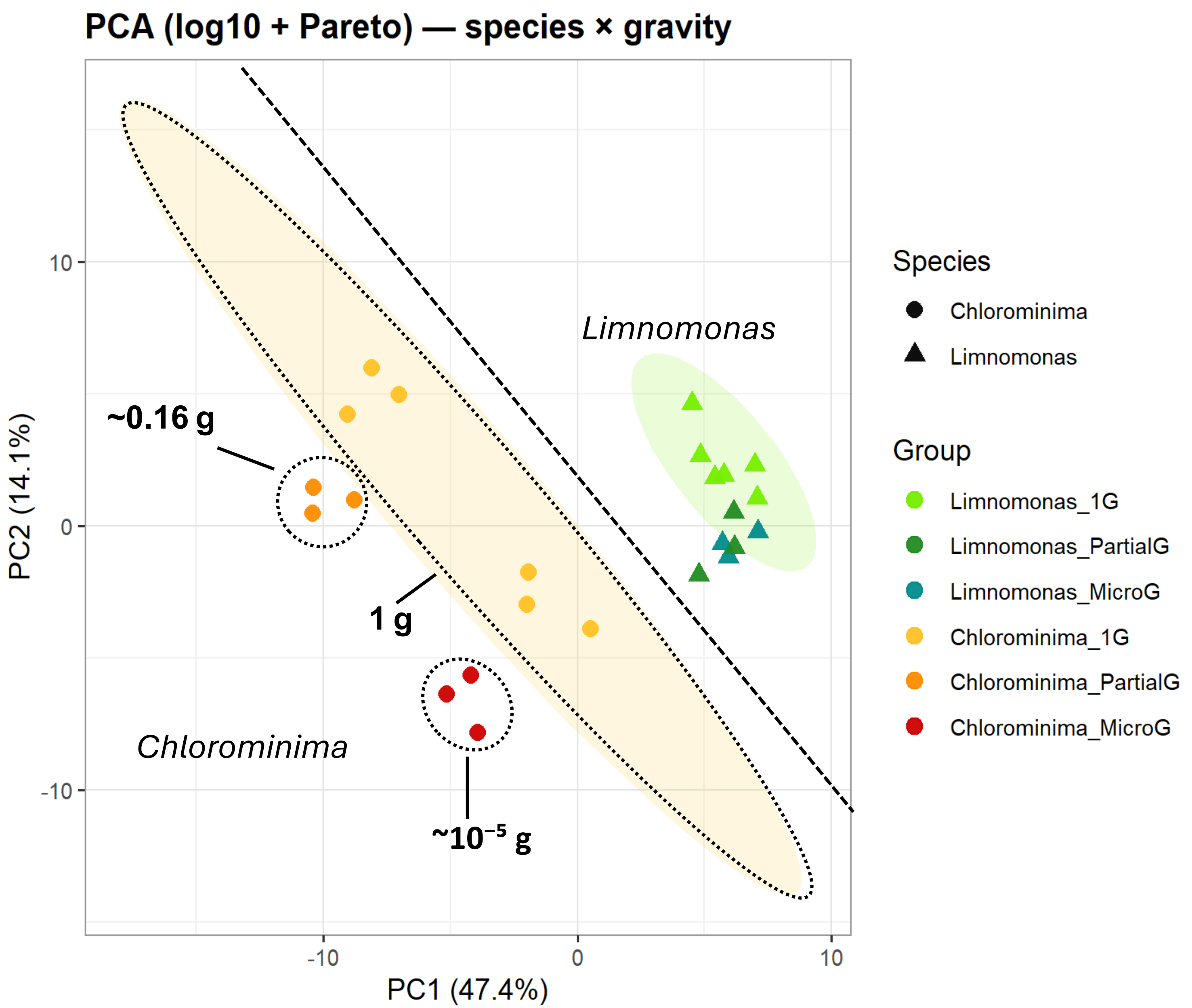
Simulated microgravity and partial gravity - Results

Chlorominima grows more under simulated microgravity, but *Limnomonas* shows little change.



Specific growth rates (u, day-[1]) of *Chlorominima sp* and *Limnomonas sp* cultivated under Earth gravity (1g), lunar partial gravity (~0.16g), and microgravity (10^{-3}-10^{-6}g). Values are mean ±SD from three biological replicates.

Simulated microgravity and partial gravity - Results



Chlorominima shows strong metabolic shifts, *Limnomonas* maintains a conservative profile

PCA of GC-MS peak intensities red(circle n = 3), light orange (circle n = 3), dark orange (circle n = 3), blue (circle n = 3), light green (circle n = 3) and dark green (circle n = 3) snow algae isolates in different gravity conditions. Also illustrated are the 95% confidence intervals for each group

Conclusions

- Both species survive simulated lunar light cycles and altered gravity → ***Robust survival traits for space environments***
- Species strategies differ
  - *Chlorominima = Metabolically plastic under altered gravity*
  - *Limnomonas = Metabolically plastic to biological context*
- Microbial context matters!

  Axenic = Reliability & Control

  Xenic = Resilience & Microbial interactions

- For BLSS: Metabolic Plasticity supports snow algae resilience and system stability during lunar nights and reduced gravity
- Next step → Strain choice and community design

Thank you!

***Get in touch*:**

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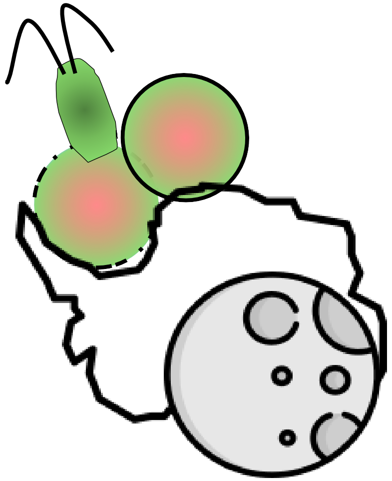

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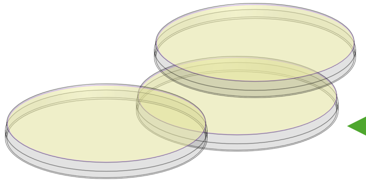
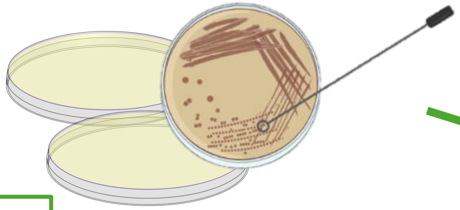
Methods

Preliminary work

Snow algae isolation using antibiotic treatments and 1NBBM Agar (1.5%)





Re-streak several times

Unikaryotic agar plates produced

Scale up to liquid culture for experiments

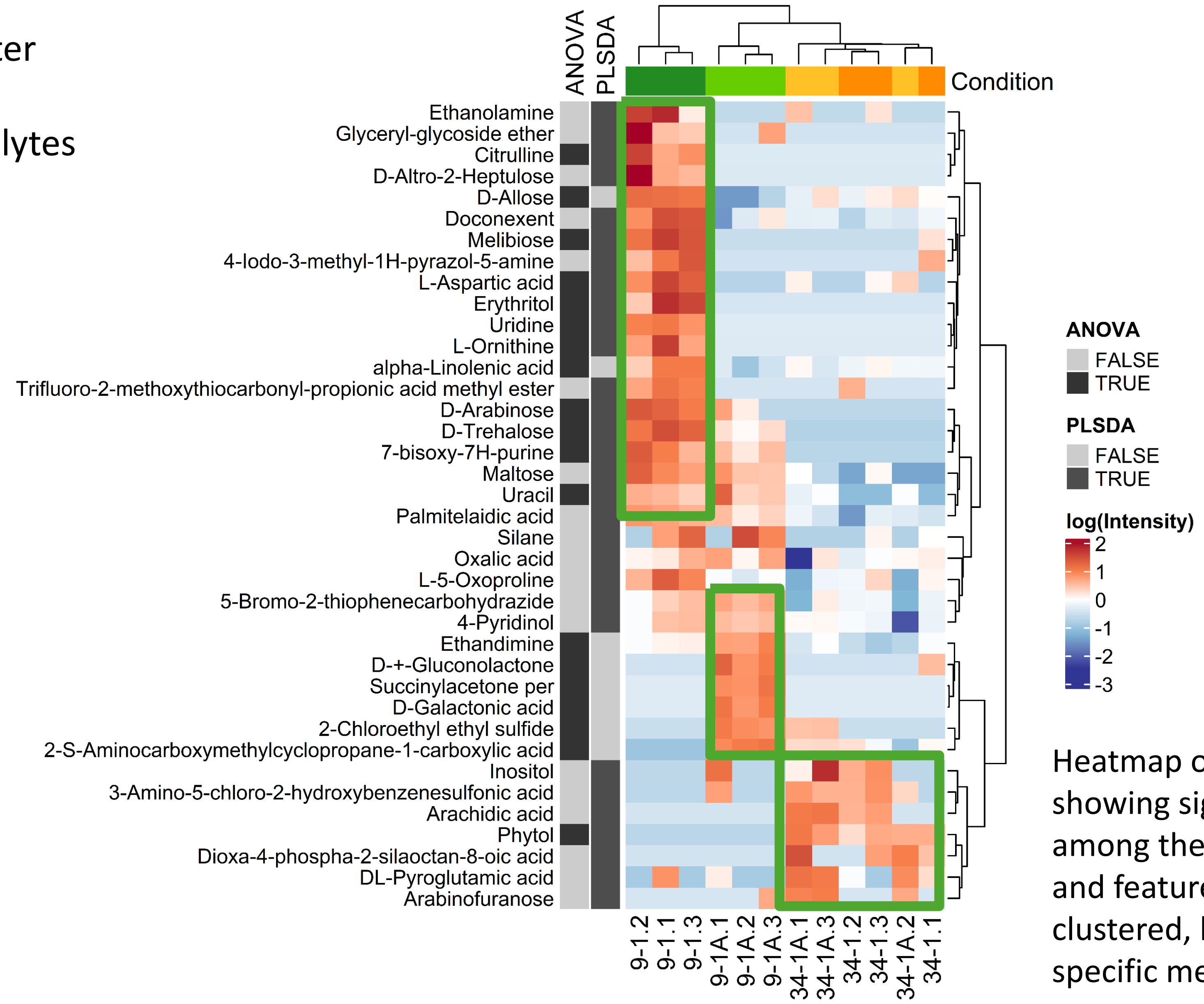
Stock cultures are maintained at

- 16:8h (light: dark)
- 100PAR 4°C
- 1NBBM + AB
- Subcultured every 3-6 weeks

Simulated lunar photoperiods - Results

Intra and interspecific – Cluster patterns:

- Sugars, AAs, PUFAS, osmolytes
- Sugar acids, redox stress, metabolic imbalance
- Phytol, lipids, inositol, glutathione related



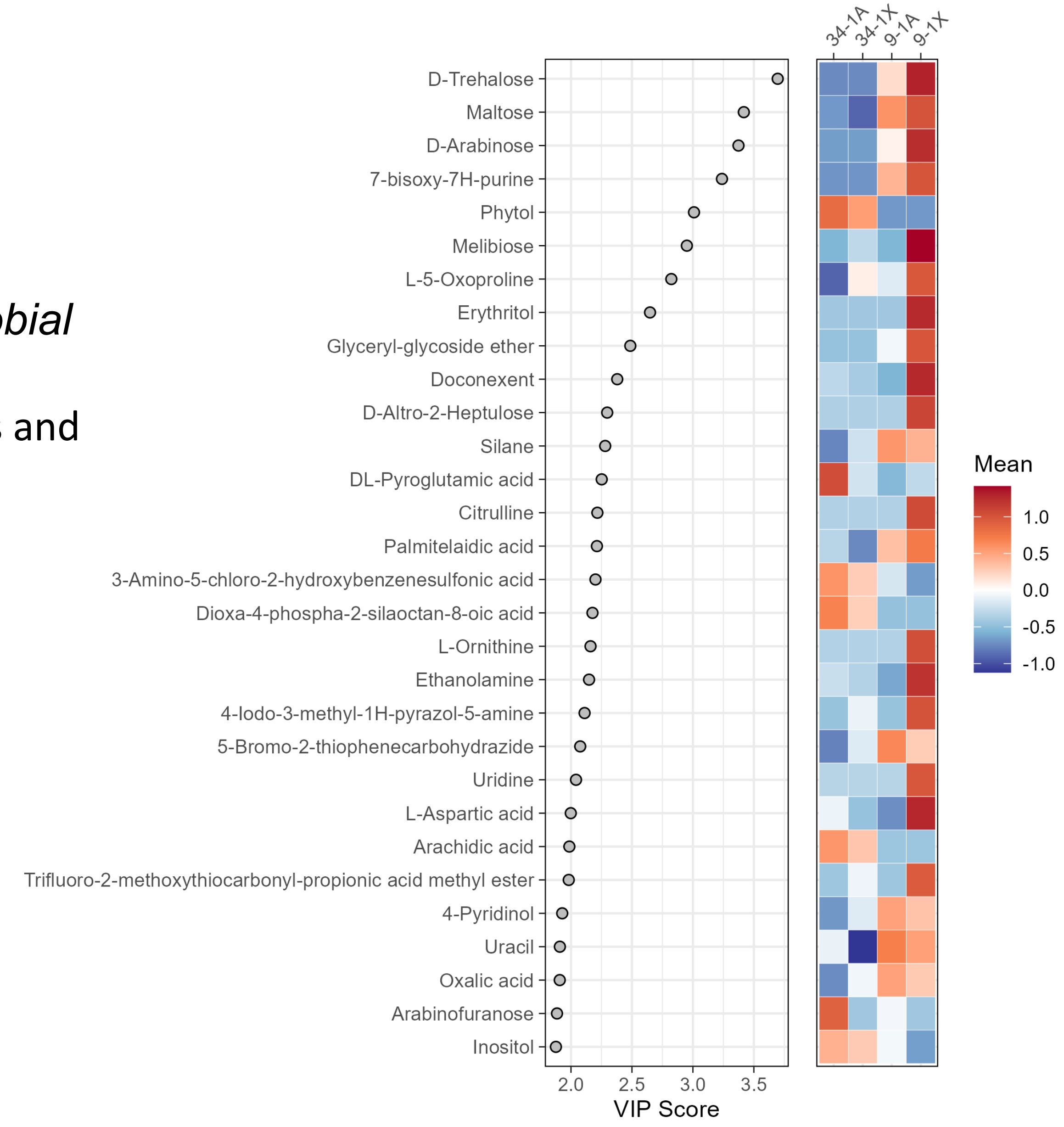
Heatmap of the top 30 metabolites showing significant differences among the four groups. Samples and features are hierarchically clustered, highlighting group-specific metabolic patterns.

Simulated lunar photoperiods - Results

- Sugars like:

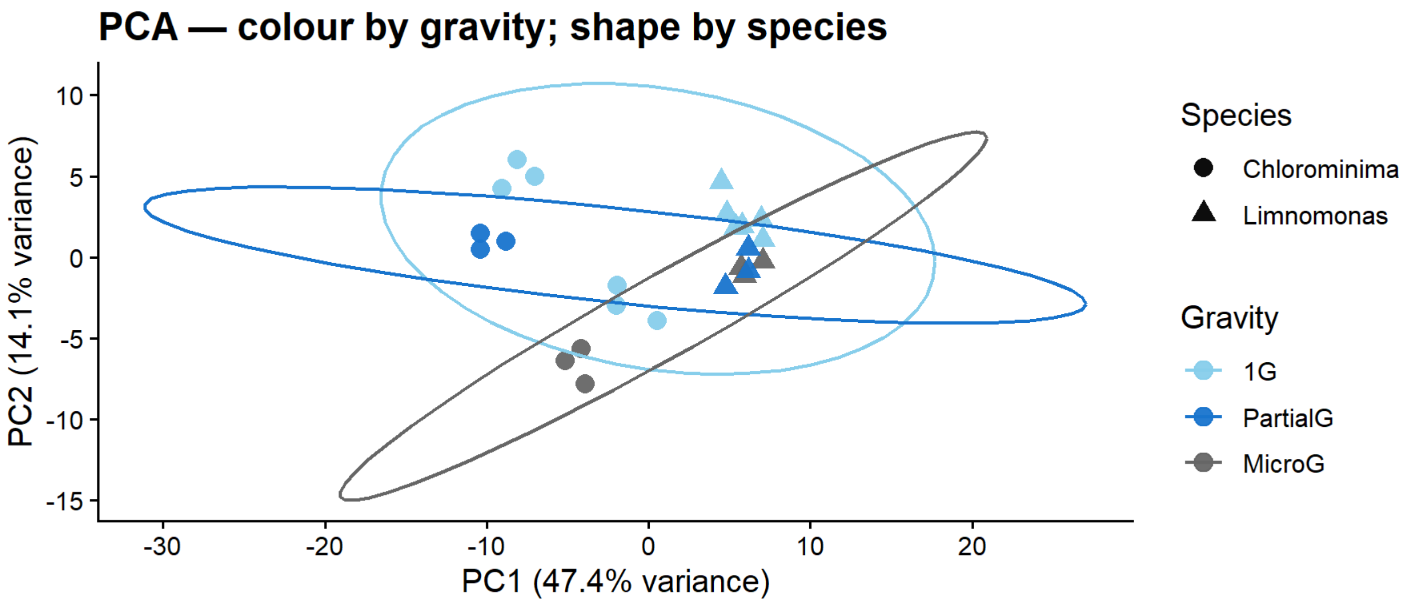
Trehalose (an antifreeze),
Maltose
D-arabinose rank highest

- Key discriminators under different *light and microbial* conditions.
- Stress-related: osmolytes and or energy resources



VIP (Variable Importance in Projection) score plot of the most important discriminant metabolites by PLS-DA. The relative abundance of each metabolite was indicated with a colour code scaled from blue (low) to red (high). A high VIP score indicates a high impact of the metabolite as a discriminant feature among the sample groups.

Simulated microgravity and partial gravity - Results



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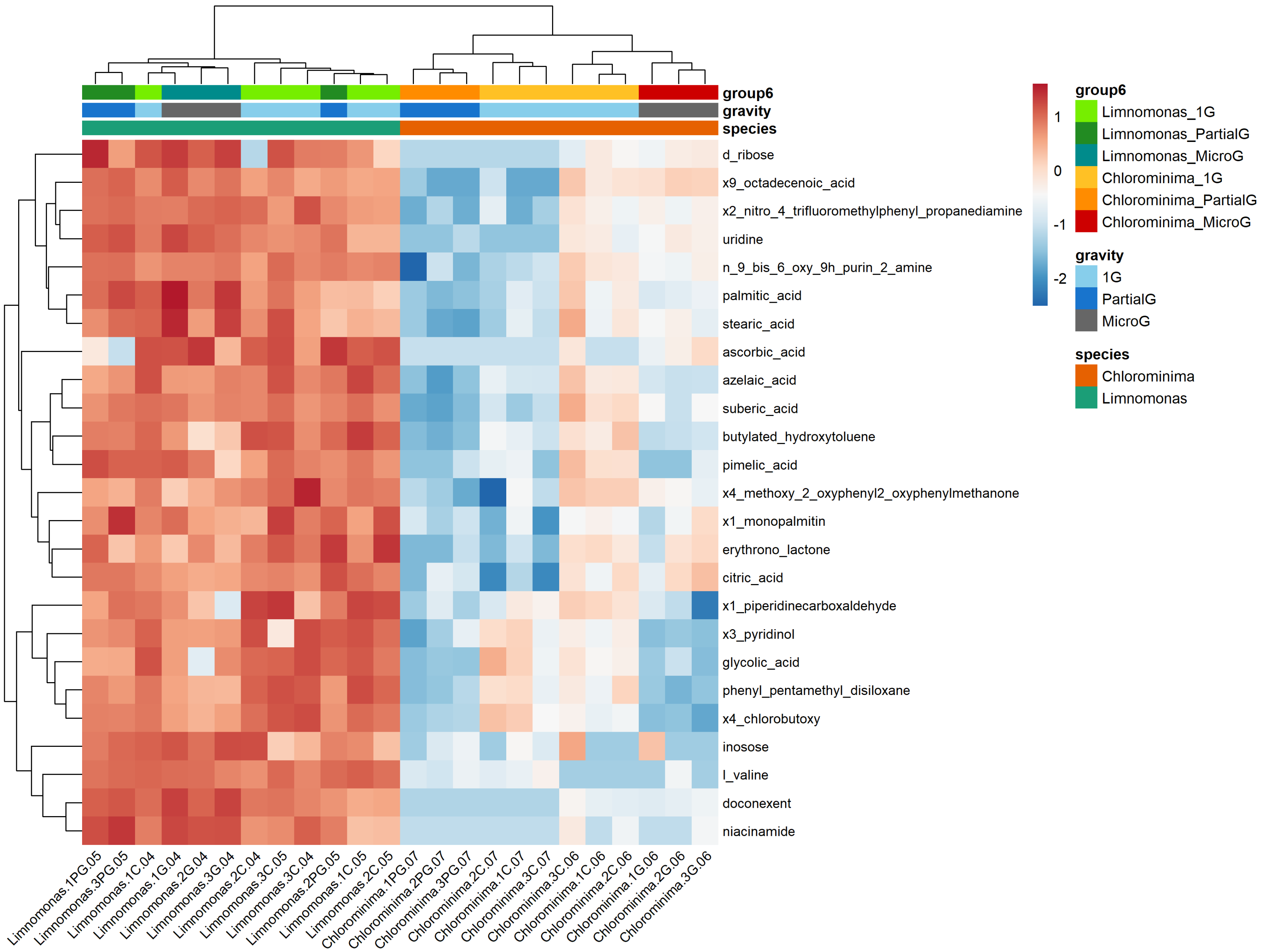

Simulated microgravity and partial gravity - Results