

Harnessing Filamentous Fungi for the Extraction of Industrial and Agricultural Chemical Elements from Planetary Regolith Simulants

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BACKGROUND

Long-term exploration and settlements of the Moon and Mars relies on *in situ resource utilization (ISRU)*, reducing dependence on Earth resources and lowering mission costs [1, 2].

Space biomining uses microorganisms to extract metals and nutrients from the regolith for potential construction usage. The ESA-led *Microbial Consortia for Space Biomining* project combines bioleaching fungi with oxygen-producing bacteria to model sustainable ISRU strategies [3]

Fungal biomining shows strong potential: *Penicillium simplicissimum* can tolerate 60% (w/v) lunar regolith and release magnesium, calcium, iron and aluminium under low-energy conditions [1]. Beyond metal recovery, filamentous fungi solubilize **plant-essential nutrients** (Mg, Ca, K, P, N) via organic acids and siderophores, enhancing regolith bioavailability for **space agriculture**.

Biotechnology also supports **circular life support**, enabling waste recycling and adaptability to different mission classes [2]. While bacteria such as *Shewanella oneidensis* can mobilize iron even under perchlorate stress [4] fungi offer advantages including stress tolerance, hyphal networks and effective chelation. These traits make them key candidates for **off-Earth crop production**.

PROJECT AIM

Over the next year, our aim is to **test and optimise the bioleaching potential of filamentous fungi** to release both construction-relevant metals and plant nutrients (e.g. N, P, K) from planetary regolith simulants. We will focus on:

- LHS-1E engineering-grade lunar highland simulant [Space Resources Technology]
- MMS-1 Mars regolith simulant [The Martian Garden]

EXPERIMENTAL DESIGN

The project is split into **TWO** phases:

PHASE 1 – Growth Characterisation

Fungi → *Aspergillus niger*, *A. oryzae*, *Rhizopus oligosporus*.

Substrates → LHS-1E (lunar highland simulant), MMS-1 (Mars simulant), sand (-ve control), all mixed with 2% agarose; agarose (2%, -ve control), PDA (positive control).

Measurements:

- Biomass (dry weight, protein content)
- Radial Growth (colony expansion)

Dilution Gradients → determine tolerance thresholds & optimal conditions.

Aims & Hypotheses:

- Growth performance on different substrates will vary by substrate type & concentration.
- *A. niger* expected to show superior growth due to metabolic versatility [5, 6]

PHASE 2 – Elemental Bioleaching

Workflow → Leachate collection → filtration/centrifugation → HNO₃ acidification → optional solid digestion

Analytical Methods → ICP-MS / ICP-OES / AAS
Measure solubilized elements → Fe, Mg, Ca, P, K, Na, trace metals

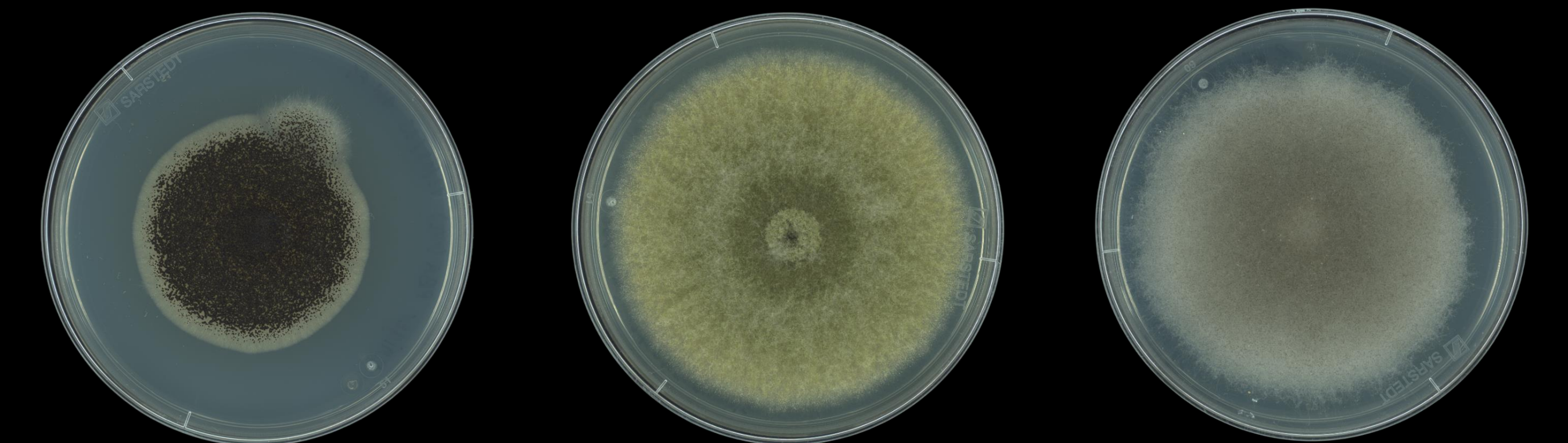
Aims & Hypotheses:

- Compare fungal bioleaching across substrates and conditions to assess ISRU potential for construction and agriculture.
- Fungal species differ in elemental leaching efficiency and spectrum, with *A. niger* being highest overall (strong acid producer).
- Regolith type influences the diversity & concentration of released elements

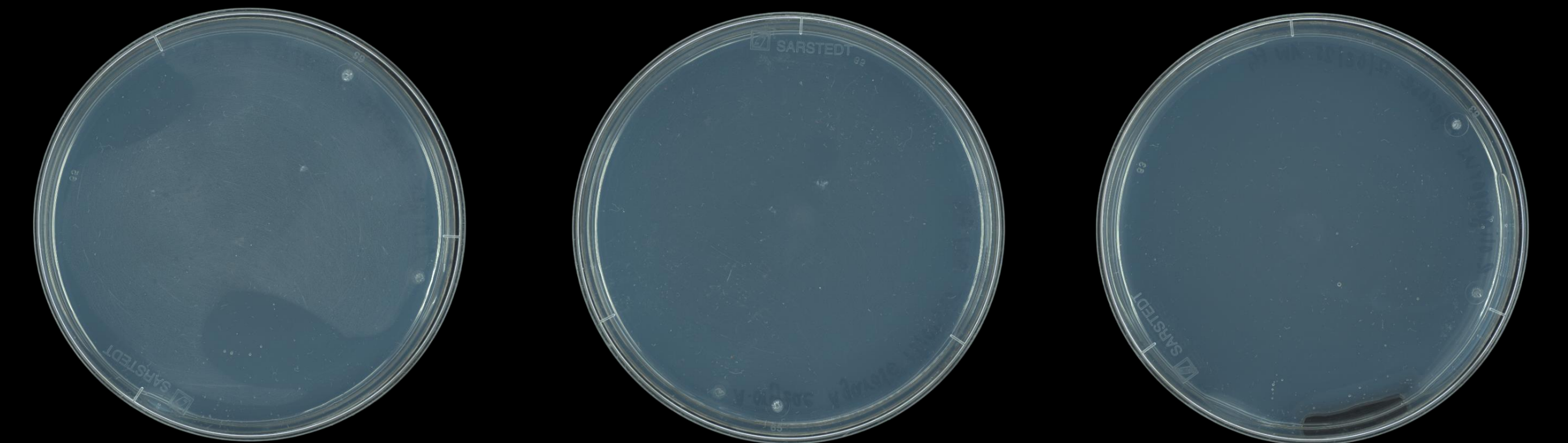
PRELIMINARY

RESULTS

Positive Control: PDA (Potato Dextrose Agarose)



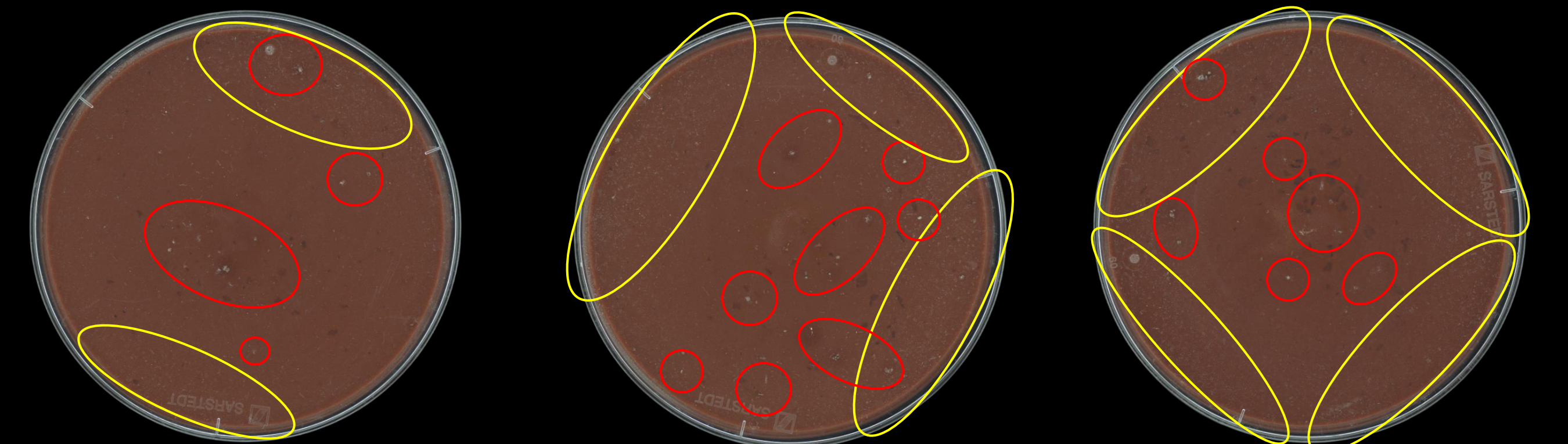
Negative Control: Agarose Gel



Treatment 1: Sand (Earth analogue)



Treatment 2: MMS-1 (Mars regolith simulant)



Treatment 3: LHS-1E (engineering-grade lunar regolith simulant)



KEY:

(LEFT) - *Aspergillus niger*
(MIDDLE) - *Aspergillus oryzae*
(RIGHT) - *Rhizopus oligosporus*
Red – Definite fungi colonies/growth
Yellow – Faint areas of fungi growth
Purple – Inconclusive/Unknown Growth

CONCLUSIONS

- PDA:** All fungi grew as expected. *A. niger* appeared reduced compared to *A. oryzae* and *R. oligosporus*, most likely due to a dilution error rather than biological limitation.
- Agarose:** No growth observed, confirming that nutrient availability in the treatments alone determined fungal survival.
- Sand:** *A. niger* and *R. oligosporus* showed no growth. *A. oryzae* displayed abnormal colony morphology, inconsistent with standard identification, possibly indicating contamination.
- MMS-1 :** All fungi exhibited sporadic, patchy growth. *A. oryzae* grew most extensively, *A. niger* the least, and *R. oligosporus* was intermediate. Colonies were largely white and diffuse, consistent with vegetative hyphal expansion but limited sporulation.
- LHS-1E :** All three fungi grew sporadically. *A. oryzae* and *A. niger* showed comparable colony presence, while *R. oligosporus* grew least. Darker, denser patches suggested more mature colony establishment and possible early sporulation, indicating marginally greater support for fungal biomass compared with MMS-1.

FUTURE DIRECTIONS

- Revisit Phase 1:** Formally characterise fungal growth on each treatment to establish baseline growth profiles.
- Expand fungal library:** Test additional candidate fungi to build a databank of useful species and define their potential applications (e.g., biomining, nutrient recycling, biopolymer production).
- Nutrient supplementation:** Test whether addition of simple carbon/nitrogen sources enhances growth consistency and biomass yield on regolith simulants.
- Biomining potential (Phase 2):** Assess whether fungi can solubilise or mobilise useful elements (e.g., Fe, Mg, Ca, trace metals) from regoliths under controlled conditions.
- Bioprocess optimisation:** Explore alternative growth strategies (e.g., co-culturing fungi with bacteria or cyanobacteria) to improve nutrient cycling in resource-limited substrates.
- Space agriculture implications:** Investigate filamentous fungi potential for biowaste recycling and soil conditioning in closed-loop extraterrestrial life-support systems.

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