

Identification of Solar-Powered N₂-Fixing Cyanobacteria from Native Ecosystems for Bio-Nitrogen Fertilizer Application in South Dakota

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2018-05-22 2:31 PM
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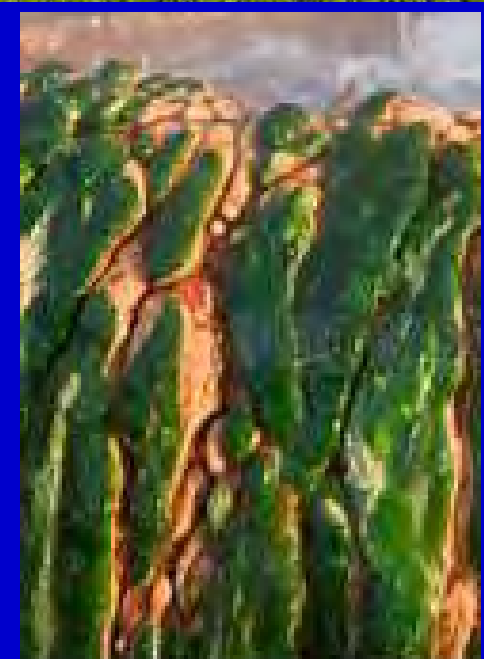
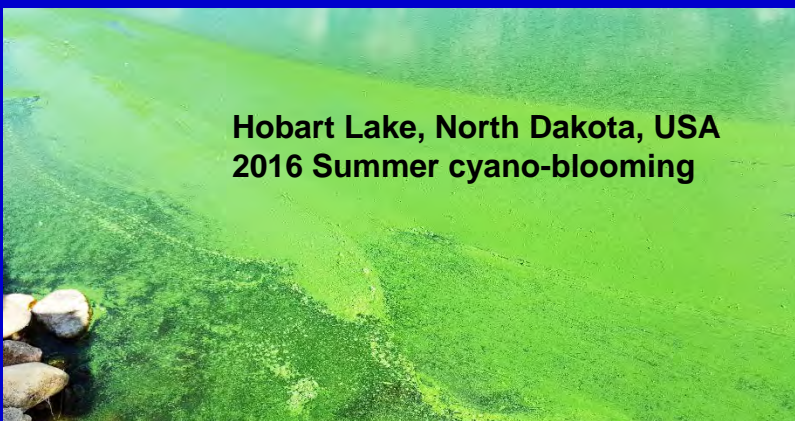


Solar-powered N₂-fixing Cyanobacteria Can Fix >10M tons of N annually to support annual biomass production by native grasses/forests

Unique Solar-powered, N₂ & CO₂-Fixing Biochem Pathways

Cyanobacteria can fix $\approx 50\%$ of 258 billions tons of CO₂ annually, **also fix >10M tons of N annually**
(258,000,000,000 tons CO₂ on earth are fixed biologically annually)

All life forms on Earth are relying on these Two Most Fundamental Biochem Pathways



Sunlight and N₂ gas are the most abundant resource around us

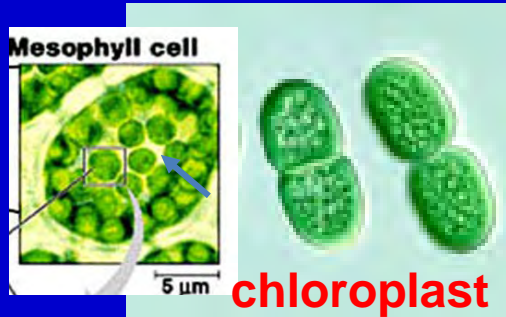
Dinitrogen (N₂ gas) as a resource is 2,000-fold more abundant (78%) than CO₂ (0.04%) in the atmosphere and can be converted into numerous of high-demand chemicals by N₂-fixing cyanobacteria.

Although we are surrounded by this vital resource such as N₂ gas and solar energy, but we cannot easily use it since they are in the wrong form.

Fortunately, the uniquely, solar-powered N₂-fixing cyanobacteria can help us to use these two unlimited natural resources.

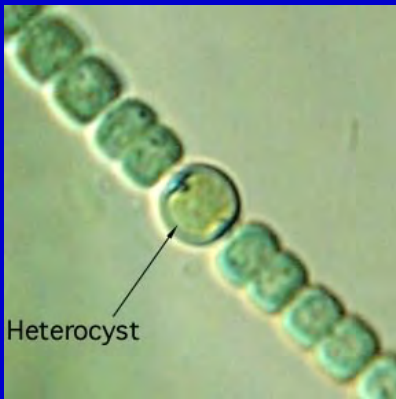
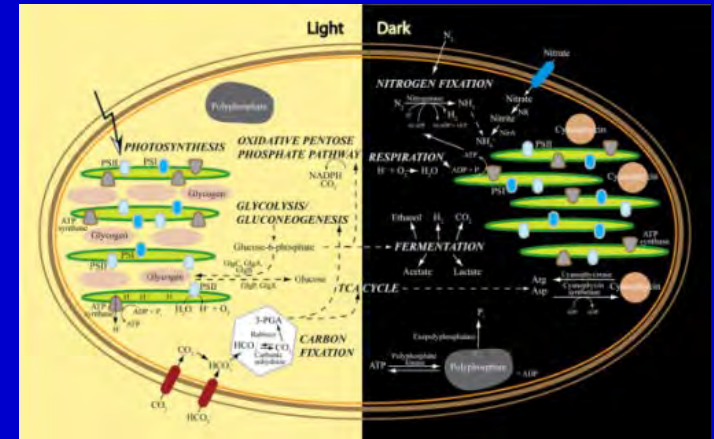
How Can They Do So?

Two Major Groups of Solar-Powered N₂-fixing Cyanobacteria



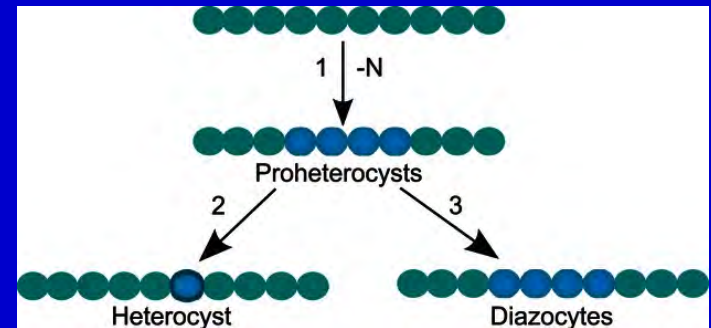
Temporarily

By timing and
circadian
rhythm



Spatially

Specially
differentiated
cells

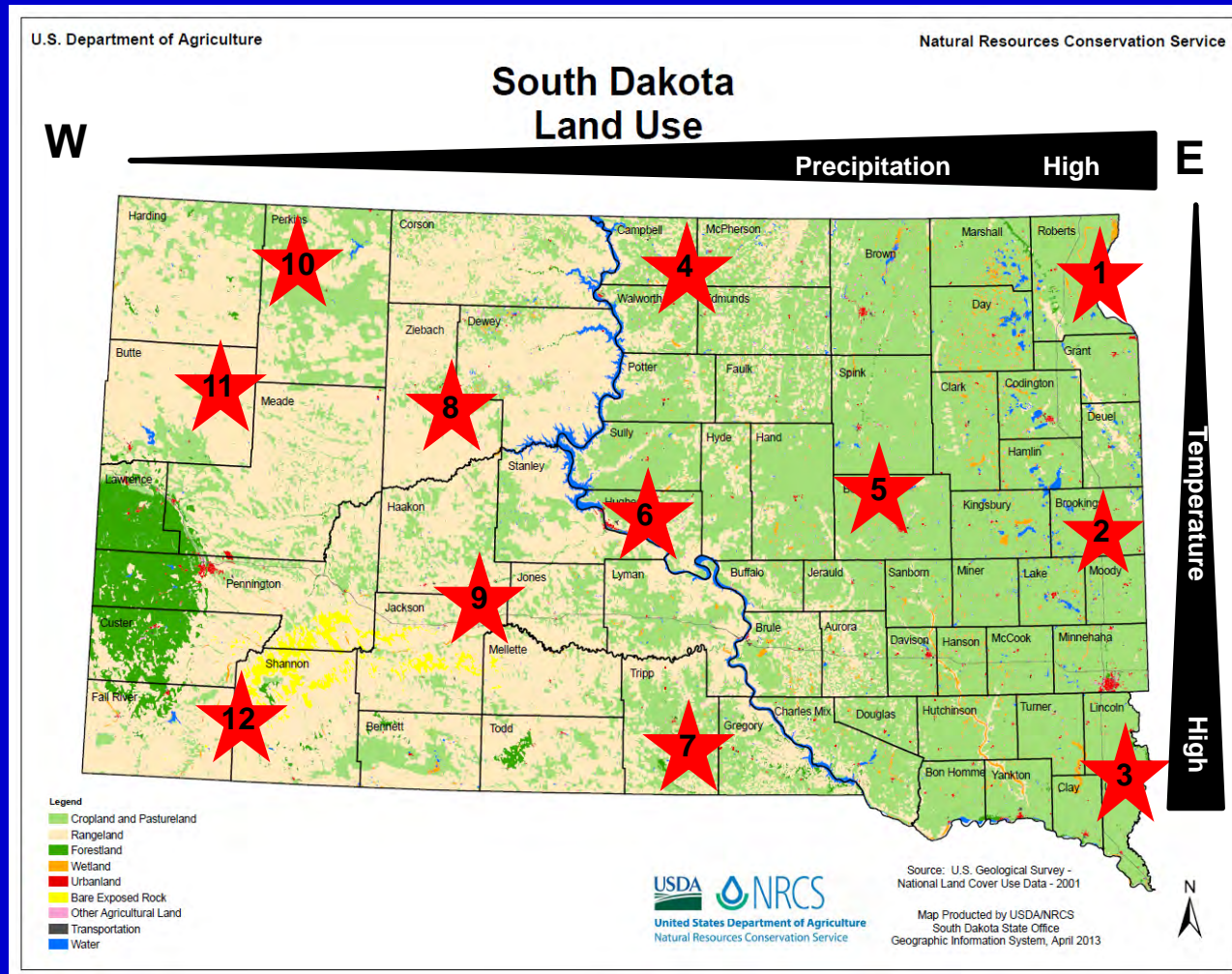


Project Objectives

- ❖ Isolation of N_2 -fixing cyanos from native ecosystems in SD (Y1-Y2)
- ❖ Quantifying their capability of nitrogen fixation (Y3-Y4).
- ❖ Inoculate the best isolates with plants in a sterilized agar-plate and Qatar sands to test its impact on plant growth (Y4).
- ❖ Then move to a sterilized soil and actual soil in greenhouse trials (Y5)
- ❖ Finally field trials (Y6).
- ❖ Commercial production of solar-powered nitrogen-fixing cyanos for **bio-N** fertilizer (Y7-Y10).

Identify N₂-Fixing Cyanobacteria in Topsoil of SD

The Best Organisms Under the Sun



3 Native soil



3 No Till Corn soil



3 Conventional Tillage soil

Fig. 2. Distribution of topsoil sampling sites at SD



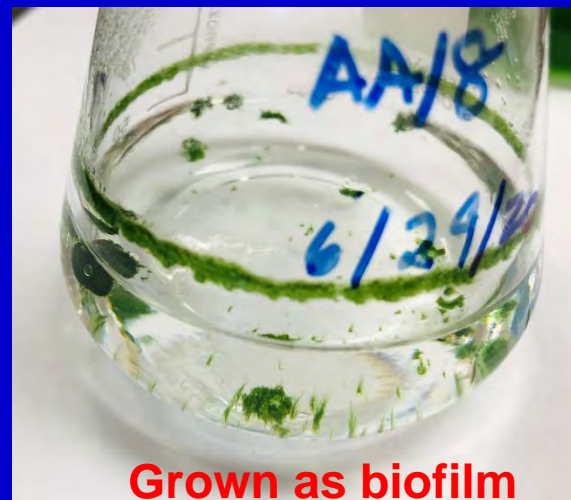
**>1000 agar-plates were used to screen for
N₂-fixing cyanobacteria**



Results Summary

- 1) From the 244 topsoil samples, we have isolated **at least 15 new potential N₂-fixing cyanobacterial strains** that are able to reproducibly grow well on BG11₀ agar plates (without any fixed nitrogen), suggesting that these 15 strains can fix nitrogen using solar energy.
- 1) Interestingly, we noticed that 15 potential N₂-fixing cyanobacteria seems exclusively found in **either No-till crop soils or native ecosystem**, Not in tilled crop soils.
- 2) Microscopy verified that eight of them are filamentous cyanobacteria, and seven of them are unicellular cyanobacteria/microalgae.
- 3) Acetylene reduction assay confirmed at least **5 strains (all filamentous)** have great nitrogenase activity.
- 4) The other **10 potential N₂-fixing cyanobacteria or microalgae** strains are currently under purification to test for nitrogenase activity

BLN-C1 B, N-



Grown as biofilm

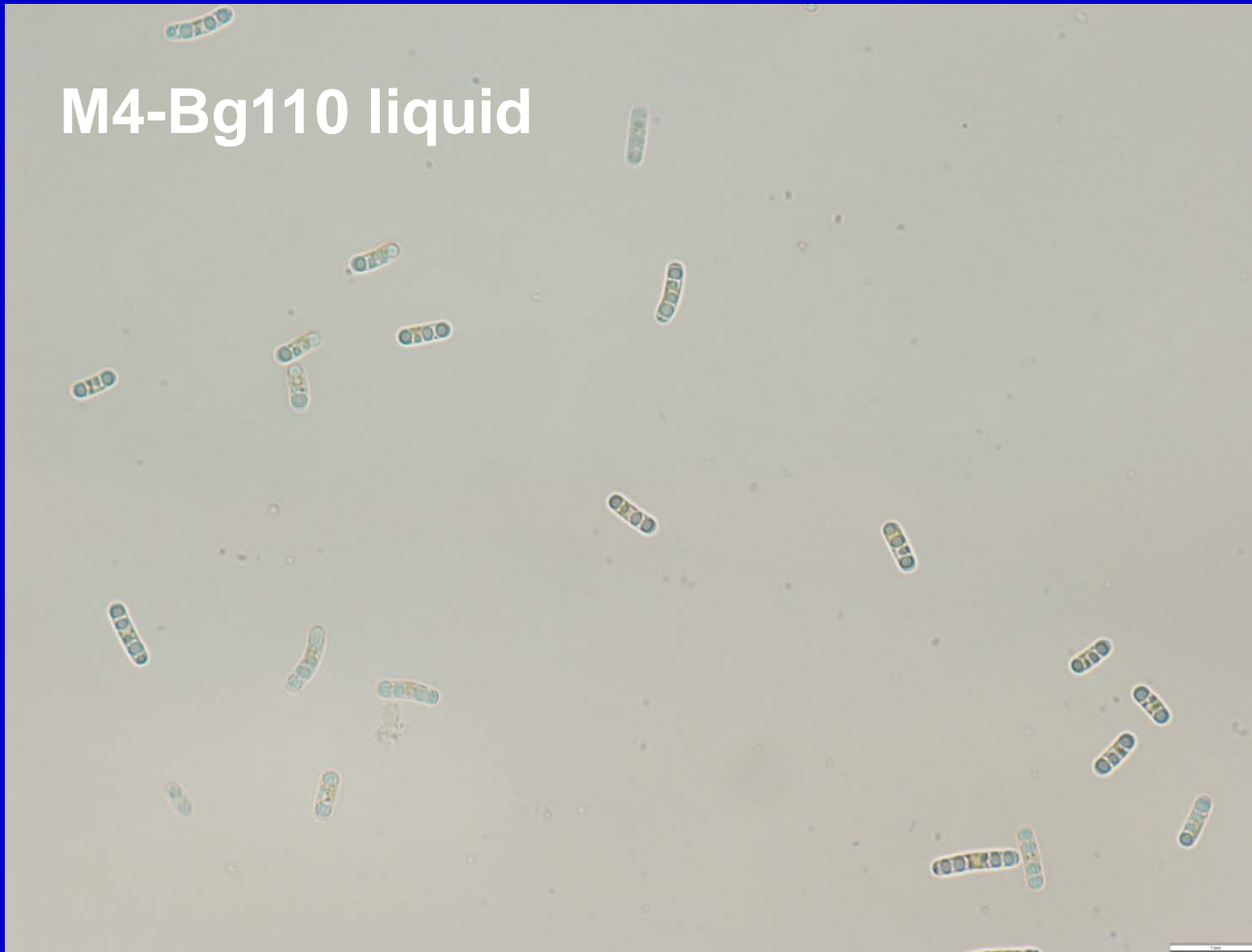


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BLN-C1B was isolated from this mega-drought condition



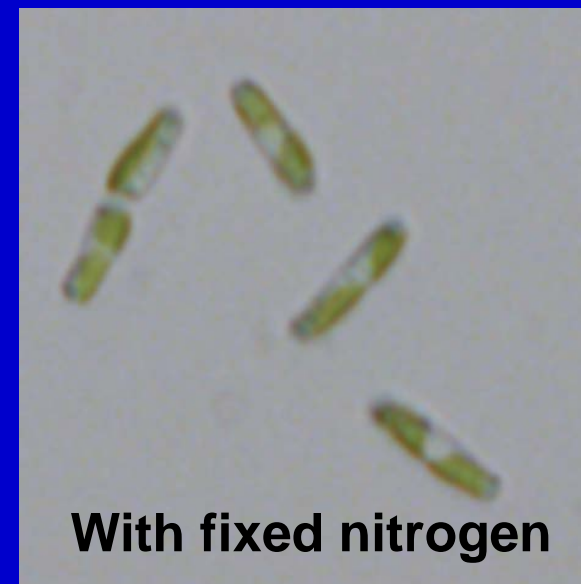
M4-Bg110 liquid



M4-BG110 plate



Without fixed nitrogen



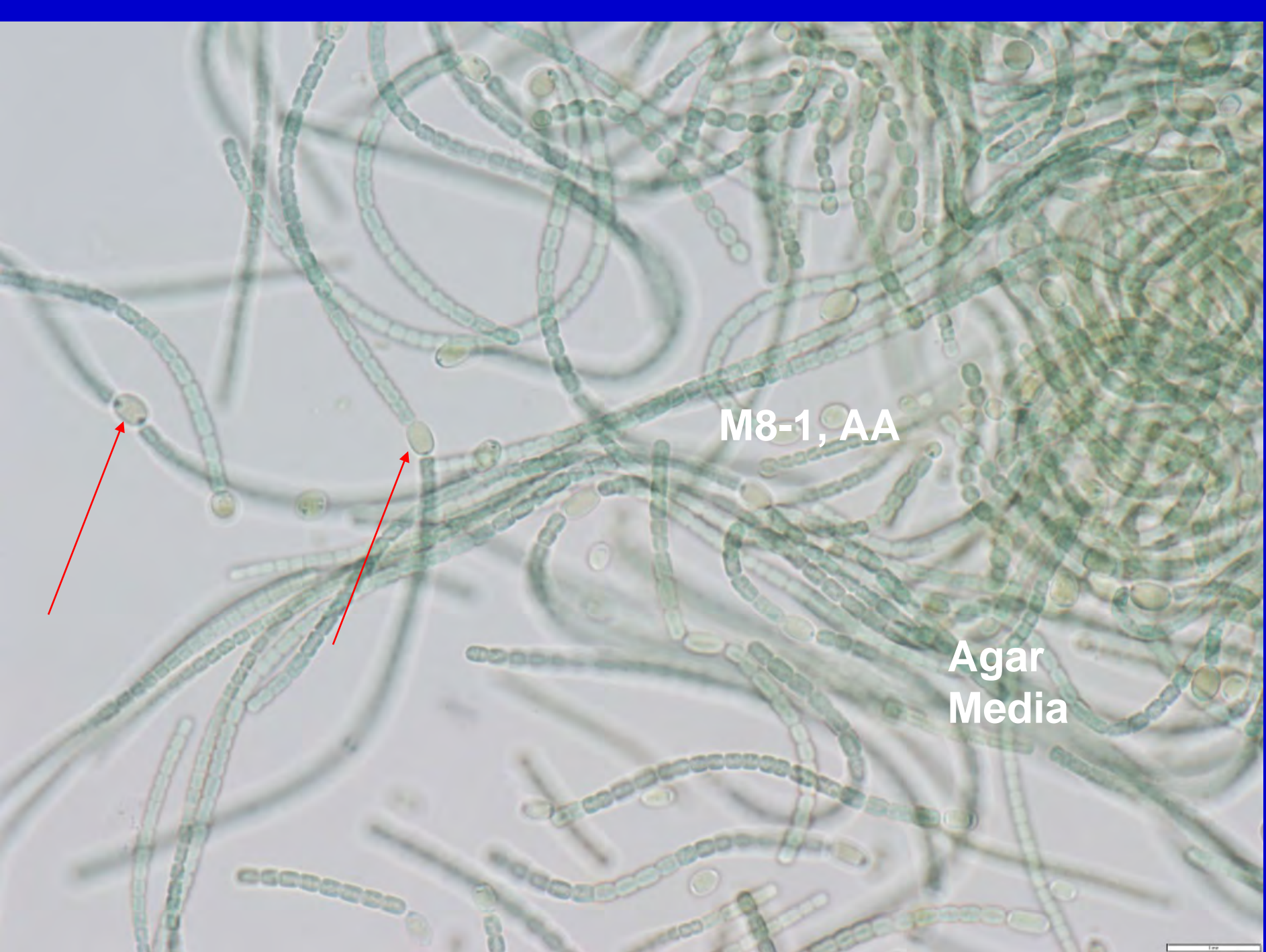
With fixed nitrogen

SDH-2B.1

N-

40x





M8-1, AA

Agar
Media



M10, BG110-L

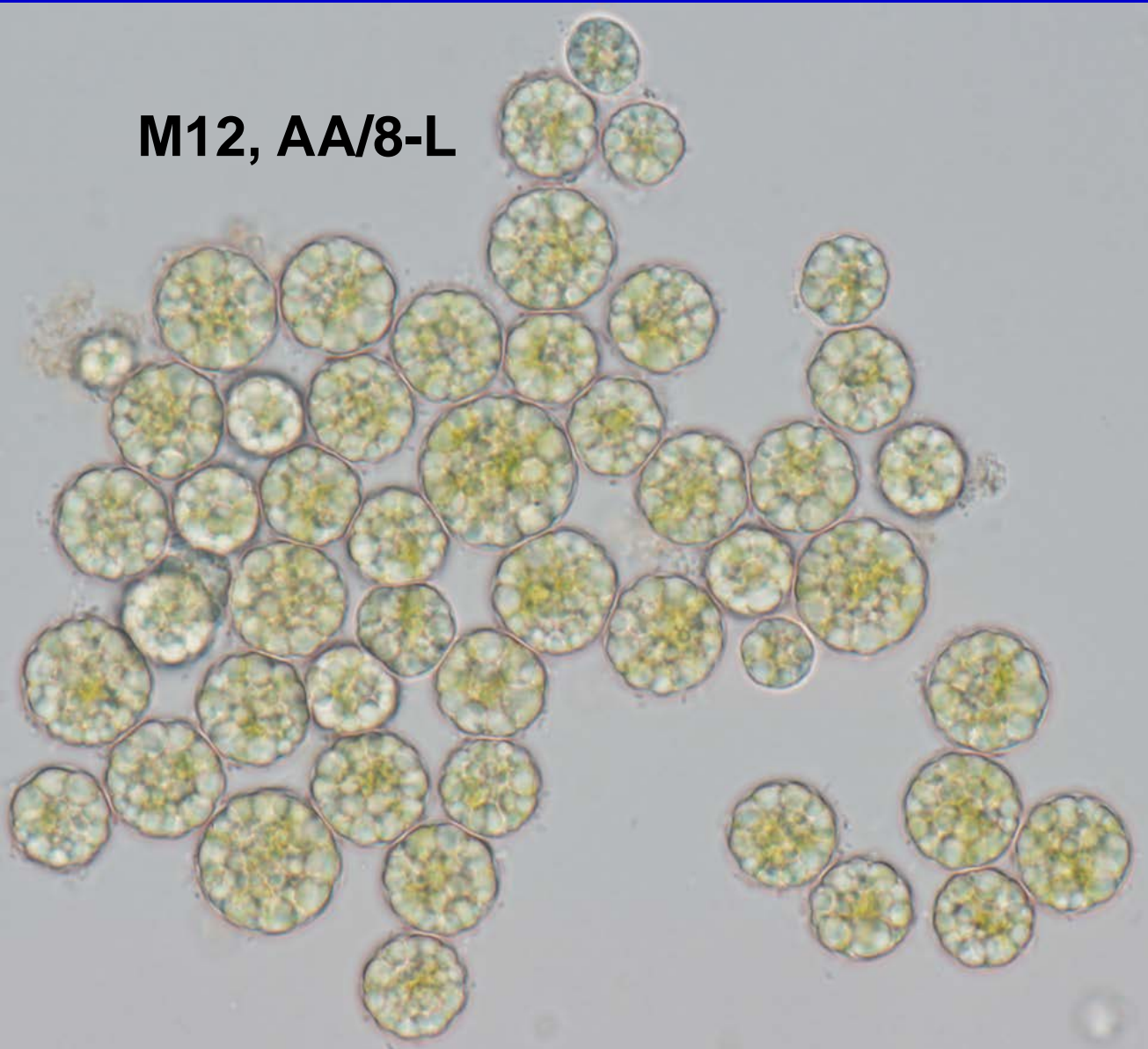
This micrograph shows a sparse population of M10 cells in BG110-L medium. The cells are visible as individual, oval-shaped structures with a distinct green internal structure and a thin, light-colored outer membrane. They are distributed across the field of view.



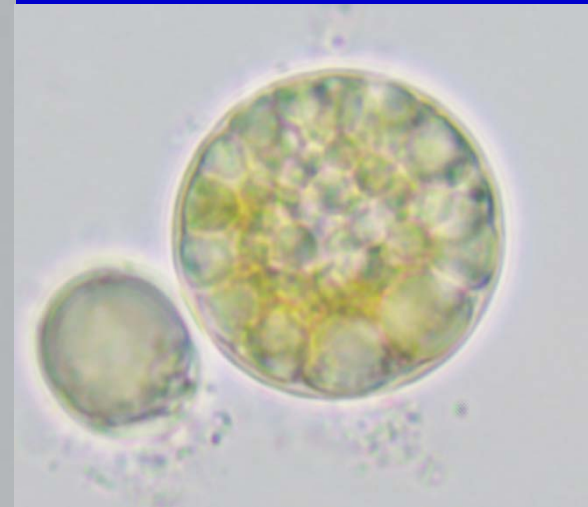
M10, BG110-Solid

This micrograph shows a dense, confluent mass of M10 cells in BG110-Solid medium. The cells are tightly packed together, forming a large, irregular cluster. The overall color of the mass is a vibrant yellow-green. A small white scale bar is visible in the bottom right corner of the image.

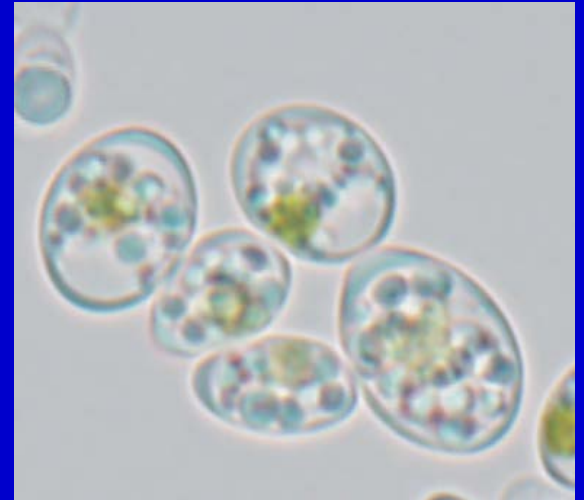
M12, AA/8-L

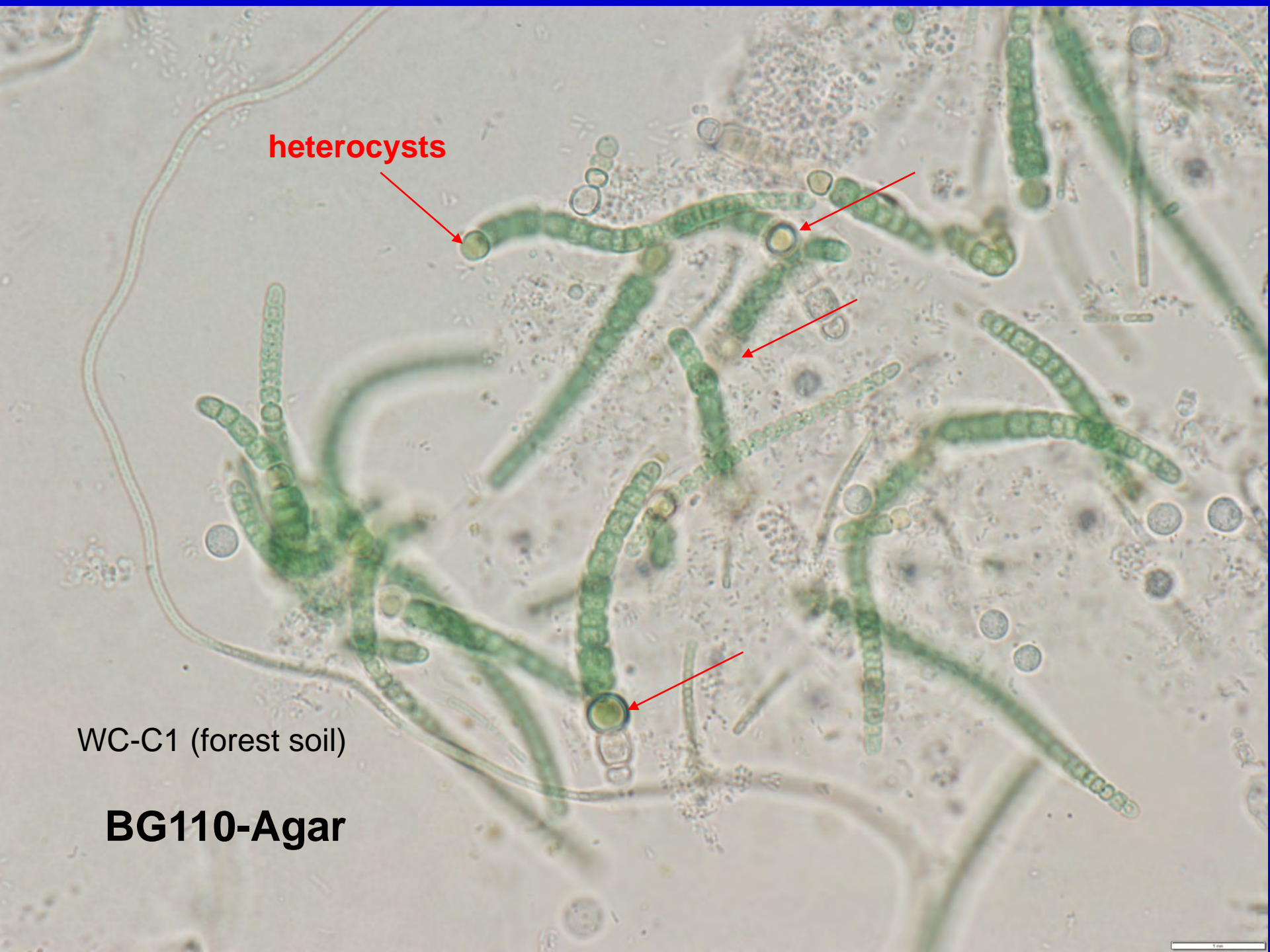


M12, AA



Xu81, native grasslands, AA solid






heterocysts

WC-C1 (forest soil)

BG110-Agar



A light micrograph showing numerous long, thin, filamentous cyanobacteria (CSP-C2) and several smaller, spherical cyanobacteria (BG110-Agr). The filaments are composed of individual cells, some showing internal structure. The background is a light, slightly textured surface.

**CSP-C2,
BG110-Agr**

Thank you so much for funding this pioneer research!

We wholeheartedly plead your continued supports on my next proposal!



Native grasslands

