

Effect of Nitrate on the Colony Morphology of *Gloeocapsa* (*Gloeotheca*) sp.

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Nitrate seems to influence the nature of the extracellular investments and thereby the colony morphology of *Gloeocapsa*. By suitable manipulation of the nitrate content of the medium, it was possible to obtain a homogenous suspension. The advantages of a homogenous single-celled culture of *Gloeocapsa* in physiological, biochemical and genetic studies, need hardly be over emphasised.

Key Words : Cellular investments, Colony morphology, *Gloeocapsa* Mucilage, Nitrate effect, Sheath

Introduction

Variations in the colony morphology of *Gloeocapsa* have frequently been observed in culture (Brand 1900 Crow, 1922, Hollerbach, 1938 Jaag, 1940 Padmaja, 1972). Colony morphology is a direct reflection of variations in the nature of the extracellular investments. The extracellular investments of *Gloeocapsa* sp. can be grouped as: (a) the "sheath", with or without laminations, and is usually visible without staining, and (b) the mucilage, which is an amorphous shroud surrounding the colonies and variable in consistency (Martin & Wyatt 1974, Bold & Wynne 1978). With the change in the nitrate supply in the medium, the extracellular investments were altered and thus the colony morphology. This was exploited to produce homogenous single cell suspension

of *Gloeocapsa* which is very useful for physiological studies.

Materials and Methods

The Alga: *Gloeocapsa* (*Gloeotheca*) sp., strain LB 795 of the Indiana University Culture Collection, was obtained from the culture collection centre of the Centre of Advanced Study in Botany, University of Madras (MUBL No. 93).

Cultural Conditions: The Culture was maintained in Allen and Arnon's (1955) medium amended with 20 mM KNO₃ and trace elements manganese, boron, zinc, molybdenum and copper at the concentrations as used by Fogg (1949). The culture was maintained at 25±1°C and illuminated with

cool-white fluorescent lamps at an intensity of 500 lux and 12:12 LD regime.

Variations in KNO_3 Concentration: *Gloeocapsa* sp., was grown as batch cultures in basal medium amended with varying amounts of KNO_3 ranging from zero to 20 mM (cf. figure 2a). The size of the colony, the number of cells per colony, the relative number of unicellular to multicellular colonies, the presence/absence of sheath and mucilage and also their nature were observed microscopically.

Gradual depletion of KNO_3 : *Gloeocapsa* sp., was also grown in basal medium amended with 20 mM KNO_3 contained in the growth chamber of a semi-continuous culture apparatus (figure 1). After 15 days growth, one-half of the culture in the growth chamber

was drained off and then made up to the original volume with N-free medium from the reservoir. This process was repeated every 5th day, and finally, after the tenth replacement, nitrate was not detectable. The culture was examined microscopically every time it was drained off the chamber and any change in external investments, colony morphology etc. was recorded.

Results and discussion

In *Gloeocapsa*, when mucilage is copious, the colonies clump together forming large masses. Conversely, as mucilage gets reduced, the size and number of cells per colony gets progressively diminished with a corresponding increase in the number of single cells.

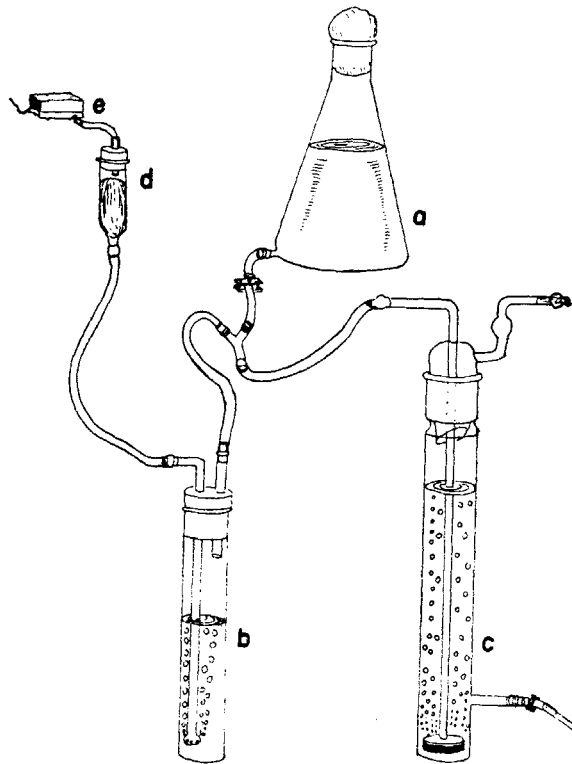


Figure 1 Semi-continuous culture apparatus. a, Reservoir; b, Humidifying chamber; c, Growth chamber; d, Cotton filter; e, Air pump

The latter culture yields a homogenous suspension.

The culture showed varied responses when grown in batch culture with different concentrations of nitrate (figure 2a). Increase in mucilage content was noticed with an increase in nitrate content of the medium. Changes in other features like the presence/absence of sheath, number of cells per colony, number of single cells etc., were at random and not related to the nitrate concentration. This experiment did not yield a homogenous culture as both single cells as well as small and large-sized colonies were present in all concentrations of nitrate.

Nitrate *per se* had some role in altering the nature of the extracellular investments of *Gloeocapsa*. An attempt was then made to gradually reduce the nitrate content of the medium and monitor changes induced therein in the semi-continuous culture condition rather than batch culture.

Growth of *Gloeocapsa* as a semi-continuous culture along with the gradual dilution of the nitrate content resulted in an uniform suspension predominantly of single cells (figure 2b). This was due to the separation of the cells in a colony into a number of individual cells associated with the gradual reduction of mucilage and its complete absence during the final dilutions.

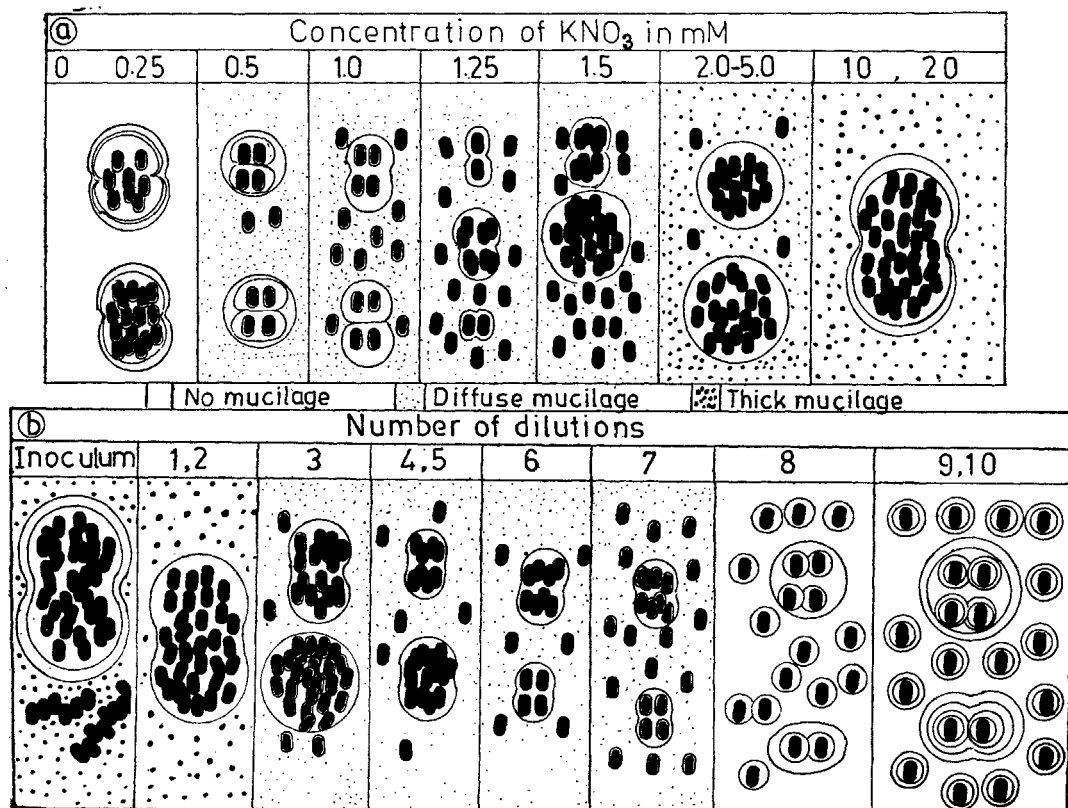


Figure 2 Schematic representation of the appearance of the culture of *Gloeocapsa* sp., grown as (a) batch culture with various concentrations of KNO_3 , and (b) semi-continuous culture (See text for details).

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