ISOLATION AND CULTIVATION OF ALGAE

Aim: Isolation and cultivation of algae from a given sample (water or soil).

Algae exist in environments ranging from oceans, rivers, and lakes to ponds, brackish waters, and even snow. Algae are usually green, but they can be found in a variety of different colors. The media used for the isolation and cultivation of algae, such as chlorophyll-bearing organisms, are capable of manufacturing their own carbohydrate requirements through the process of photosynthesis.

Principle

The principles behind the isolation and cultivation of algae and *Spirulina*, *Chlorella*, and *Scytonemia*, are grounded in microbiology, environmental science, and ecological understanding. These processes are designed to achieve two primary objectives: isolation, which separates the target microorganisms from complex microbial populations, and cultivation, which provides controlled conditions for their growth and reproduction.

Isolation aims to obtain pure cultures of the specific algae or cyanobacteria by separating them from mixed microbial communities. The key principle is the use of dilution techniques to reduce population density, followed by plating onto suitable agar-based media. This separation allows for the selection of individual colonies or cells with the desired characteristics, ensuring a pure culture. The cultivation principle revolves around mimicking the natural habitat of these microorganisms in a controlled environment. This involves providing a culture medium rich in essential nutrients (e.g., nitrogen, phosphorus), simulating the ecological conditions where these microorganisms thrive. Environmental factors such as light, temperature, and aeration are regulated to facilitate photosynthesis, metabolic processes, and overall growth.

Requirement:

- 1. Water sample: Soil /Freshwater sample (Pond/Lake /other freshwater water bodies)
- 2. Culture medium: Sterile Beneck's broth flasks (250 ml capacity) medium-200 ml
- 3. Erlenmeyer flasks (250 ml capacity)-2 nos.
- 4. Light source (Natural/ artificial lighting) for photosynthesis.

Beneck's Broth#:	KNO_3	0.2 g
	$MgSO_4$	0.2 g
	K_2HPO_4	0.2 g
	CaCO ₃	0.1 g
	FeCl ₃ (1%)	2 drops
	Distilled water	1 liter
	рН	7.5
	*Agar	20 g (*Only in Beneck's agar medium)

^{*}Deshmukh, A. M. (2007). Handbook of media stains and reagents in microbiology. Oxford Book Company.

Procedure:

- 1. Prepare Sterile Beneck's broth medium, transfer 100 ml into 250 ml Erlenmeyer flasks, plug, and autoclave them.
- 2. When the broth is cool, add 1g or 1ml of soil/water to each flask, shake well, and incubate the flask at 25-30°C (room temperature-RT) for 15 days under natural/artificial lighting for photosynthesis.
- 3. After incubation, algal colonies will appear in the flasks in the broth.
- 4. Prepare Beneck's agar medium by adding 2% agar to Beneck's broth and autoclaving as described above.
- 5. To obtain pure cultures, transfer a few cells of algal colonies with the help of the nichrome wire loop onto the Beneck's agar medium in petri dishes.
- 6. Incubate the plate at 25-30°C (RT) for a week under natural/artificial lighting illumination.

Observation:

Expected Observation:

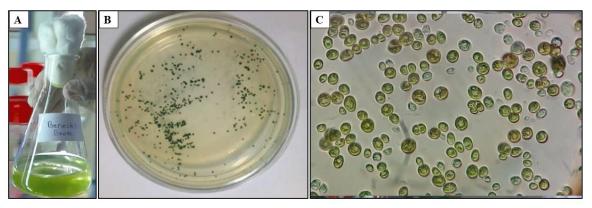


Fig.1 Isolation and cultivation of algae; A) Green color in Beneck's broth due to algal growth. B) Beneck's agar plate showing green algal colonies. C) Microscopic view (100X) indicating green algal cells.

- Green coloration in Beneck's broth (Fig.1A) indicates the growth of algae.
- Algal colonies observed on Beneck's agar plate appear as green clusters (Fig. 1B).
- Microscopic examination at 100X magnification reveals individual algal cells showing characteristic green pigmentation (Fig. 1C).

Result: Algal colonies were observed on Beneck's agar; however, algal cultivation was confirmed by microscopy.

Result interpretation: Based on the above result, it is concluded that the green color observed in Beneck's broth is due to the cultivation of algae from the given water sample.