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Biochemical Perimeters Measurements of Algae Extracted from Galtaji Pond Jaipur

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Abstract: The growth and biochemical content of *Chlorella vulgaris* in batch culture during an interval of two weeks are evaluated in the present investigation. With fresh weight increasing from 250 mg to 490 mg and dry weight from 9.3 mg to 18.6 mg, biomass exhibited exponential growth, indicating ideal culture conditions for cell proliferation. Significant metabolic improvements were identified by biochemical analyses: total soluble sugars increased by 34% (2.727 to 3.667 mg), starch by 32% (3.550 to 4.673 mg), lipids by 24% (6.317 to 7.860 mg), and proteins by 20% (15.69 to 18.925 mg), indicating enhanced nitrogen assimilation, anabolic metabolism, and carbon fixation. Improved light-harvesting efficiency and stress tolerance were also indicated by an increase in photosynthetic pigments, such as chlorophyll *a* (33%, 1.033 to 1.372 mg), chlorophyll *b* (23%, 0.493 to 0.608 mg), total chlorophyll (30%, 1.527 to 1.980 mg), and carotenoids (15%, 0.127 to 0.146 mg). These results suggest that *C. vulgaris* biomass and biochemical productivity may be effectively increased in laboratory conditions.

Keywords: *Chlorella vulgaris*, biomass, total soluble sugars, proteins.

I. INTRODUCTION

Algae are photosynthetic organisms that occur in diverse habitats like marine and freshwater to desert sands including hot boiling springs to snow and ice. They occur from unicellular to multi cellular forms. They have various mode of reproduction like vegetative, asexual and sexual.

From the ecological point of view they possess important place in food chain as they manufacture organic food and helps in aquatic habitat (Borah *et al.*, 2020; Lucakova *et al.*, 2021).

The Chlorophyceae family of algae is the most diversified and developed group of all the algae. It is widespread and may be found in a variety of environments. Among them is the green algae *Chlorella vulgaris*, a single-celled member of the phylum Chlorophyta. The most extensively grown eukaryotic green microalgae is *Chlorella*, which finds use in the cosmetics and pharmaceutical industries, as well as in health food and feed supplements. Proteins, lipids, carotenoids, immune-stimulating substances, polysaccharides, vitamins, antioxidants, and minerals are all present (Costa *et al.*, 2019; Sedaghat *et al.*, 2025).

The primary component of algae that includes carbs is polysaccharides, which are made up of a variety of soluble and critically important phytochemicals. Polysaccharide release in the intracellular space that assesses the allelopathic connection pathway, ion exchange desorption processes, and cell defence against extreme stimuli. Algae have enough lipid and fatty acid storage products, metabolites, as membrane component, and source of energy. Algal fatty acid including oils have a broad range of potential uses as they possess uniqueness similar to those of fish and vegetable oils, and can thus be measured as possible replacements for the products of fossils oil.

II. METHODOLOGY

Galtaji is an ancient Hindu pilgrimage about 10 km away from Jaipur, in the Indian state of Rajasthan. Algal samples were collected from Galtaji pond of Jaipur city (Rajasthan) and nearby sites. In a conical flask with 250 mL of medium, an additional 50 mL of culture (inoculums) was added. Three duplicates of each experiment were carried out. Following the initial measurements, observations were made over the course of two weeks. Growth, fresh, and dry weights were among the many factors examined.

Growth followed through optical density, fresh weight and dry weight. OD was recorded with the help of photo colorimeter at 670 nm. For fresh weight and dry weight 50 ml of cultures centrifuged. The alga material was used as the experimental material for further experimentation. Biochemical such as total soluble sugar (Loomis and Shull, 1937), starch (Dubois *et al.*, 1951), lipids (Jayaraman, 1958), proteins (Lowry *et al.*, 1951) and pigments like chlorophyll (Arnon, 1949) and carotenoid (Kirk and Allen 1965) carried out both qualitatively and quantitatively.

III. RESULTS AND DISCUSSIONS

The growth evaluation of *Chlorella vulgaris* in culture media indicates an apparent increase in both fresh weight and dry weight over a two-week period. At first, the dry weight was 9.3 mg and the fresh weight was 250 mg on average. Significant biomass build-up was seen after the first week, as fresh weight increased to an average of 357 mg and dry weight to 13.58 mg. The fresh weight increased to 490 mg and the dry weight to 18.6 mg after the second week. The findings suggest favourable culture conditions that promote biomass synthesis and cell multiplication (Taghavijeloudar *et al.*, 2025). The findings show significant exponential growth in the first week and a slight increase in the second, which is consistent with the growth behaviour of microalgae in batch cultures.

Table-1 Growth evaluation of *Chlorella vulgaris* in culture media

Time	Parameters	Replicate-I	Replicate-II	Average Value
Initial reading	Fresh weight (mg)	252	248	250
	Dry weight (mg)	9.34	9.26	9.3
After I st week reading	Fresh weight (mg)	352	362	357
	Dry weight (mg)	13.28	13.88	13.58
After II nd week reading	Fresh weight (mg)	495	485	490
	Dry weight (mg)	18.98	18.22	18.6

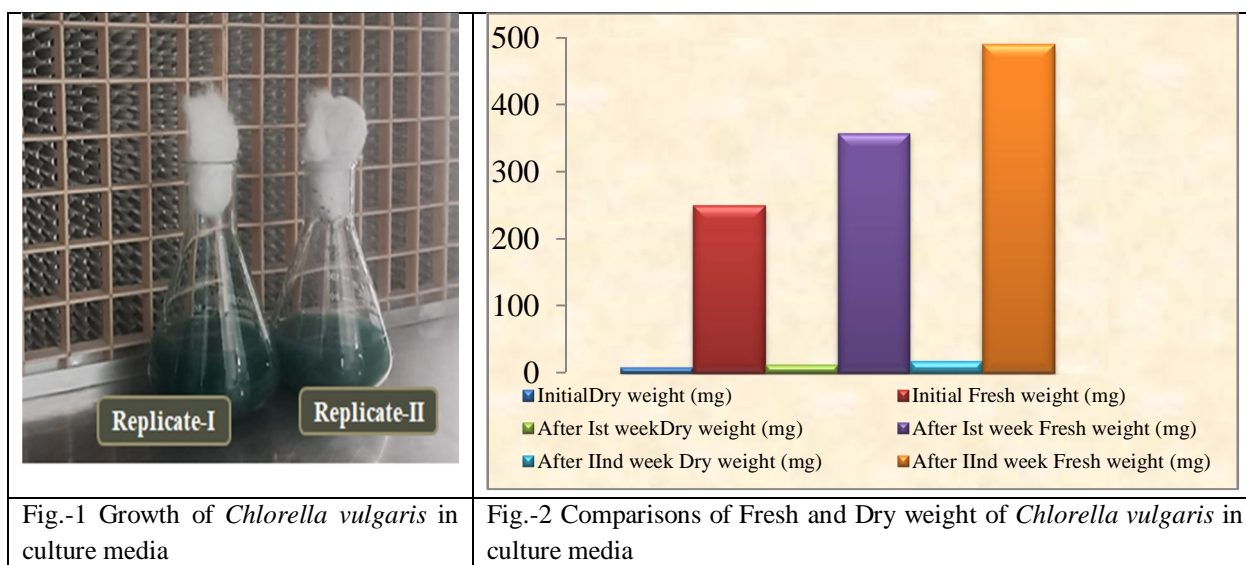


Fig.-1 Growth of *Chlorella vulgaris* in culture media

Fig.-2 Comparisons of Fresh and Dry weight of *Chlorella vulgaris* in culture media

A. Qualitative Estimation

The qualitative biochemical investigation into the sample was carried out to evaluate the presence of important organic macromolecules, including carbohydrates, proteins, and lipids.

Table-2 Qualitative estimation of Bio-chemical parameters

Bio-chemical parameters	Procedure	Presence or Absence
Carbohydrates	Fehling's Test	+
Proteins	Biuret Test	+
Lipid	Formation of soap	+

B. Quantitative Estimation

According to the quantitative evaluation of biochemical parameters, the sample revealed considerable metabolic and physiological improvement from its beginning to the final readings. These findings support the positive effect of the laboratory environment on the growth efficiency and biochemical composition. Total soluble sugar content increased from 2.727 mg at the initial phase to an average of 3.667 mg at the end of the experiment. Accelerated photosynthetic carbohydrate synthesis contributed for this 34% improvement. Soluble sugars are metabolic intermediates and instant energy sources (Yakameran *et al.*, 2024).

Their increase indicates increased carbon fixation and efficient photosynthate distribution throughout the development phase. The concentration of starch increased 32%, from 3.550 mg to 4.673 mg. Starch, being the primary storage carbohydrate in microalgae and plant tissues, accumulates when photosynthetic carbon assimilation exceeds its utilization. The increase indicates that the sample attained a metabolically advantageous state where additional sugars were progressively transformed into starch granules. This also suggests a change to anabolic metabolism and an enhanced ability to store carbohydrates (Javan *et al.*, 2024). The lipid content improved by 24%, increasing from 6.317 mg to 7.860 mg. Lipids are long-term energy stores and crucial parts of cell membranes. Improved nutritional availability, ideal culture conditions, or effective carbon partitioning towards fatty acid production might all be responsible for the increased lipid accumulation. Lipid accumulation is frequently an indicator of physiological health and possible stress adaption in microalgae, which makes the increase significant.

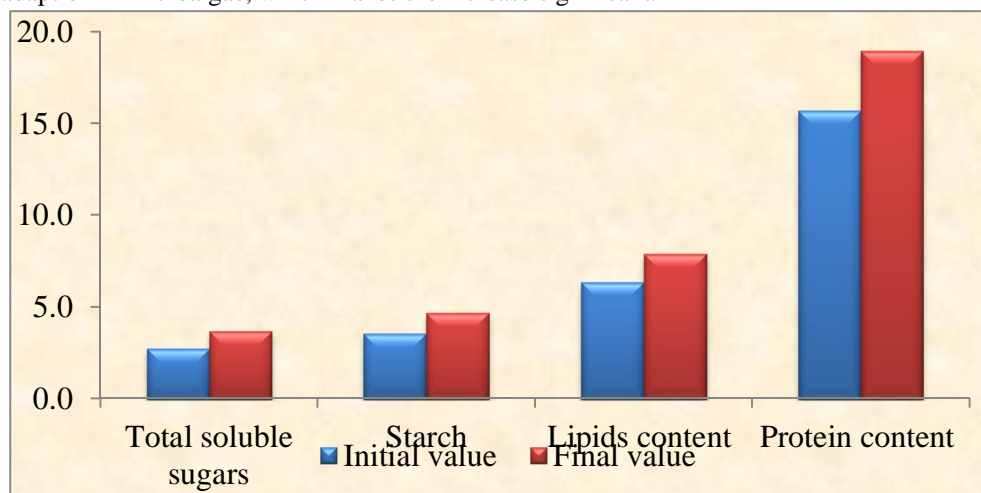


Fig.-3 Comparisons of Bio-chemical Parameters

The content of protein increased by 20%, from amount 15.69 mg to 18.925 mg. Since proteins are structural elements, functional molecules, and enzymes, they are important markers of metabolic activity. The increase suggests active cellular protein production and improved nitrogen uptake. This is a sign of increased enzyme activity, rapid cell proliferation, and enhanced metabolic performance.

C. Photosynthetic Pigments

The amount of chlorophyll-a increased from 1.033 mg to 1.372 mg. Since chlorophyll a is the main pigment involved in photochemical processes, increases in it signify improved photosynthetic rate and enhanced light-capturing efficiency. The amount of chlorophyll-b increased from 0.493 mg to 0.608 mg. Chlorophyll-b is an additional pigment that expands the spectrum of light absorption. The increase is a reflection of the photosynthetic apparatus's improved structure and functionality, especially in the antenna complexes.

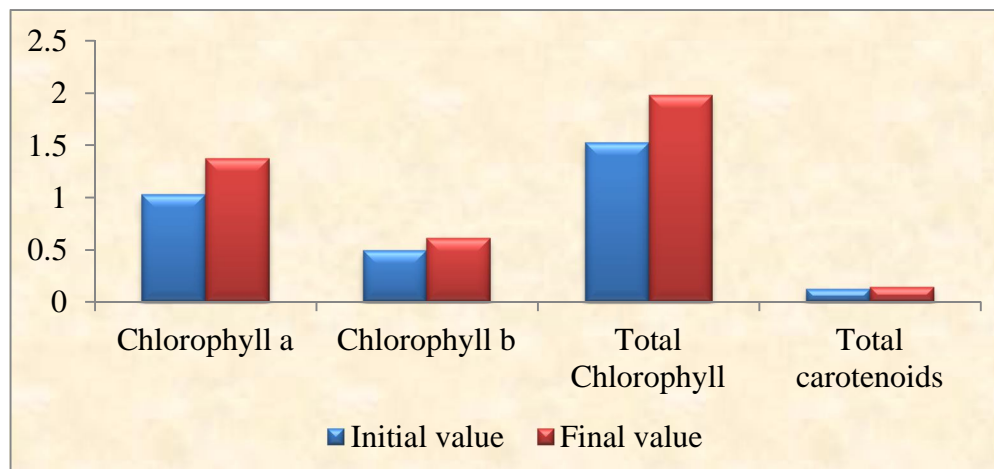


Fig.-4 Comparisons of Photosynthetic Pigments

The total amount of chlorophyll improved by 30%, from 1.527 mg to 1.980 mg. Strong photosynthetic mechanism and active chloroplast formation are strongly indicated by this considerable increase. The total carotenoid content increased by 15%, from 0.127 mg to 0.146 mg. Carotenoids is essential for energy transmission, anti-oxidation, and photo protection. The increase indicates better resistance against oxidative stress and improved chlorophyll–protein complex stabilisation. This increase supports a highly efficient photosynthetic system and is correlated with higher quantities of chlorophyll.

Table-3 Quantitative Estimation of Bio-chemical Parameters and Photosynthetic Pigments

Bio-chemical Parameters	Initial Reading	Final Reading		
		Replicate-I	Replicate-II	Average Value
Total soluble sugars (mg)	2.727 ± 0.3037	3.607 ± 0.2433	3.727 ± 0.3037	3.667
Starch (mg)	3.550 ± 0.2309	4.837 ± 0.1849	4.508 ± 0.2022	4.673
Lipids content (mg)	6.317 ± 0.3283	7.850 ± 0.2179	7.869 ± 0.3159	7.860
Protein content (mg)	15.69 ± 1.157	18.80 ± 0.4359	19.05 ± 0.4404	18.925
Photosynthetic Pigments				
Chlorophyll a	1.033 ± 0.06667	1.383 ± 0.09280	1.360 ± 0.06658	1.372
Chlorophyll b	0.4933 ± 0.04667	0.6133 ± 0.05207	0.6033 ± 0.1362	0.608
Total Chlorophyll	1.527	1.997	1.963	1.980
Total carotenoids	0.1273 ± 0.002906	0.1447 ± 0.001764	0.1467 ± 0.003528	0.146

IV. CONCLUSION

Over the duration of two weeks, the laboratory cultivation of *Chlorella vulgaris* produced significant biomass accumulation and biochemical improvements, highlighting the culture media's suitability for microalgae development. Elevated amounts of soluble sugars, starch, lipids, and proteins, together with exponential increases in fresh and dry weights, demonstrate effective photosynthetic carbon partitioning, anabolic changes, and metabolic activity. Better photosynthetic machinery and resistance to oxidative stress are further confirmed by parallel increases in chlorophyll a, b, total chlorophyll, and carotenoids. These findings point to *C. vulgaris* as a viable option for scalable biomass generation in environmental biotechnology applications like bioremediation or bioenergy, and they are consistent with usual batch culture dynamics.

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