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# STUDIES OF THE EFFECT OF TEMPERATURE VARIATIONS, DIFFERENT LIGHT INTENSITIES AND SOME NUTRIENTS ON GROWTH AND REPRODUCTION OF PEDIASTRUM BORYANUM.

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## ABSTRACT

The main object of this work is to study the effects of temperature variations, varying light intensities, and some nitrogenous substances (nitrate, ammonium salts, proteins and peptones) and non-nitrogenous substances (dextrose, sucrose and phosphate) on growth and reproduction of **Pediastrum boryanum.** The organism was cultured under controlled conditions of light and temperature in the laboratory using Schrieber and Chodat medium. The results showed that higher temperatures above 30° C have lethal effects on the alge, while Temperatures below 30° C favoured growth and asexual reproduction of the alga. Temperatures below 20° C favoured sexual reproduction. Higher light intensities (3,229 to 2,153 Lux) accelerated the rate of growth and spore formations. At low light (below 1,076 Lux) fertilisation was accelerated and occaisonal zygospores were found. Nitrate and phosphate affected the growth of the alga but when the amount of phosphate or nitrate was very little, they limited its growth. Provided that all other factors were found in excess, it was the phosphate concentration that affected growth. Addition of bacteria-free dextrose to the culture media stimulated **Pediastrum** cells to grow fast for about 6-10 days after which growth was diminshed. Cells fed with proteins continued growth and remained healthy, while those fed with sucrose ceased growth in about 7 days, Cultures provided with excess dextrose and less peptone accelerated reproduction much nore than growth.

## INTRODUCTION

**Pediastrum boryanum** is a member of phytoplankton suspended in the water of the covered drainage channel Ghait el-Nakhl and sometimes deposited on the bottom mud. Knowledge of the plankton algae is becoming of increasing practical importance in view of the need to develop and maintain adequate supplies of water of suitable quality for domestic and industrial use.

Day length and temperature, particularly the former, seem also to be important and the highest number of **Pediastrum boryanum** occurs during the prolonged periods of bright dry weather (i.e. during summer period). In late summer the diatoms become predominent forming vast floating areas, thus suppressing the number of phytoplanktons. The culture solutions used were water enriched with nitrogenous and non-nitrogenous compounds. The former included, sodium nitrate, proteins and peptones while the latter comprised dextrose, sucrose, phosphate and acetate concentrations.

**Pediastrum boryanum** is regarded as a constituent of the phytoplankton which comprises numbers of chlorophyta, bacillariophyta, flagellates and cyanobacteria. The latter comprises member with heterocysts such as **Nostoc** and other without heterocysts such as **Oscillatoria** and **Lyngbya**. The cyanobacteria deviate from other phytoplankton in the following:

- Firstly, their possession of gas vacuoles imparts a positive buoyancy which enables them to take up a favourable position in the water column.

- Secondly, the fixation of atmospheric nitrogen by the heterocystous species is likely to promote their success in nitrogen limited environment and may be the main reason for their prominence during the late summer.

- Thirdly evidence is accumulating of a significance role for extracellular products of high biological activity which may act either growth-promoting properties such as the chelation of metals or perhaps by suppressing the growth of other organisms.

## MATERIAL AND METHODS

The alga was separated from the rest of phytoplankton microscopically and thoroughly washed and cleaned from contaminants by shaking against glass beeds and water. Changing of the water was repeated till it became clear, but this does not mean that the alga was completely freed from bacteria.

Living material is to be preferred, but as **Pediastrum boryanum** is too delicate, it is as well to fix part of the sample immediately on collection. As a general preservative, Lugol's iodine is recommended. This is a saturated aqueous solution of potassium iodide, to which is added crystalline iodine to saturate it again. From this stock solution, only a few drops are needed, enough to colour the water sample a pale brown. After a few weeks or months of storage, the sample may become decolourized, when more iodine should be added.

## Experimentation

In order to procure an accurate way for testing the growth and reproduction of Pediastrum boryanum under controlled conditions several experiments were conducted for this purpose. Three sets of experiments were carried out to trace the importance of temperature variations, changes in light intensities and different concentrations of specific nutrient nitrogenous and non-nitrogenous substances on growth and reproduction of the alga. This included experiments which were carried out each at certain temperature starting from 10°C to 50°C. Four thermostats with glass botoms, each containing 6 culture bottles were used.

There are gas wash bottles, each fitted with a glass stopper through which pass 2 glass tubes one short and the other long ending into a disc of scintered glass. The bottles were put in 2 rows and connected together by means of rubber tubings of 50 mm in diameter and each row was attached to a manifold about 90 cm in length and 2.5 cm in diameter. The 2 mainfolds of each thermostat were attached together by means of a pressure rubber tubing. This was again attached to another similar one from the other thermostat through a T-tube by another pressure rubber tabing and connected to a cylinder fitted with compressed air. Two fluorescent lamps each one meter in length were used as a source of light during this experiment. The light intensity was about 1614 Lux as measured by photometer. The lamps were usually on, from about 9 amto 5 pm. The aim of putting them off during the night was to furnish more or less similar conditions to what takes place in nature. A constant current of air was allowed to pass through these bottles from a cylinder of compressed air at a rate of about half a litre per hour.

## Light Intensity Variations

The same experiments were repeated using different light intensities, while the other conditions were kept constant.

## Different Specific Nutrients

These were nitrogenous and non-nitrogenous substances added to the culture media.

# RESULTS AND DISCUSSION

This drainage channel with neutral or slightly alkaline water, generally rich in nutrients, with sediments composed of silt is regarded as a unique habitat. It has in addition to Pediastrum boryanum a rich phytoplankton of flagellates, chlorophyta, bacillariophyta and cyanobacteria. Oscillatoria, Nostoc, Scenedesmus and diatoms form the majority of phytoplankton. It appears that these genera are resistant to grazing by the soil protozoa and some rotifers and probably pass through the gut in a viable state. In

common with most photosynthetic plankton members of cyanobacteria with the exception of Oscillatoria proved to be low light organisms i.e. flourish well under reduced light intensities (1076-Zero Lux). This agrees with the findings of Mirose (1954), Haigh and Beavers (1964), and Chung and Watenabe (1982). Laboratory studies are misleading, since strains which form large colonies in nature dissociate into single cells in culture Cyanobacteria deviate from other phytoplanktons in the following:

Firstly, their possession of gas vacuoles imparts a positive buoyancy which enables them to take up a favourable position in the water column.

Secondly, the fixation of atomspheric $N_2$ by the heterocystous species is likely to promote their success in $N_2$ limitted environments and may be the main reason for their prominance during late summer. Higher temperatures above 30°C have lethal effects on growth of the alga indicated by the decrease in dry weight and reproduction. This might be due to the partly deplation of reserve foods and an increase in respiration.

This agrees with the finding of Marre (1962). Still temperature from 25°C to 30°C favours the growth, development of spores and various metabolic processes. Germination of Zoospores was observed and the life cycle traced. The optimum temperature exerting its influence on the dry weight of the alga is 25°C. Temperatures below 20°C favours the production of gametes i.e. sexual reproduction. The gametes were fast moving and positively phototactic but their flagella were not rendered visible. According to Atkins (1945) gametophytes of Undaria pinnatifida can survive within temperature range of 1°C to 22.5°C. At 30°C they die within 2-3 days. According to him the most suitable temperatures for growth and maturation of the gametophytes lie between 10°C and 20°C (especially between 15°C and 20°C). Fritsch (1952) concluded detailed investigations of the effect of temperature on monospore formation in conchocelis phase of Porphyra, and studied in culture experiments the formation of gametangia in Halicystis parvula at different temperatures and found that the average number of gametangia increases with rise of temperature from 16°C to 23°C, then started to decrease with further rise in temperature. High light intensities (3229 - 2153 Lux) accelerated the rate of growth of the alga and in about 2 weeks the cells doubled their size. The spores when shed showed a high degree of motility. Fertilization was frequent and it was proved experimentally that when the nutrients were found in excess the fertility was directly dependent on light intensity, (Ehleringer and Werk, 1986). The zygotes germinated rapidly without passing a period of rest. The gametes when shed were plugged in to a mucilgenous mass in a more or less coiled state, but under such light intensities they changed from the coiled inactive form to the uncoiled and more active one in few seconds.

The gametes then seemed to escape strong illumination which when prolonged proved to be detrimental to them and this may explain the huge masses of dead ones under such conditions. At moderate light intensities the alga showed a luxuriant growth. The cell walls were rather thin and the cells remained healthy till the end of the experiment. Fertilization was possible and development of zygotes was quite normal. At low light intensities (1076-Zero Lux), the rate of growth was greatly reduced and the colour of cells was less intensive. The nuclei became quite visible and attained a relatively large size. The act of fertilization was enhanced and numerous zygotes were formed. After about one week no zygotes were seen to grow, though some of them appeared quite healthy. The optimum growth of Pediastrum boryanum took place at about 0.54 gm. atom/litre phosphate, when the amounts of phosphate and nitrate were very little they limited the plant growth. The rate of absorption of nitrate from the culture solutions by the alga was independent of the concentration of phosphate in the culture solution but increased as the concentration of nitrate in the medium was increased.

The addition of nitrate and phosphate salts in moderate concentrations to the culture solutions resulted in high rates of vegetative growth as well as in high reproductive affinities (Cooper 1982.

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