3. Effect of SO$_2$ at different pH on

Spirogra setiformis.
INTRODUCTION

A beginning of the study of pond algae in relation to both the chemical and meteorological factors seemed to have been made in 1924 by Atkins and Harries. Birge and Juday, 1911; Anderson and Walker, 1920; Birge and Juday, 1922; Daily, 1938; Flint, 1938; Couch, 1941; Lund, 1942; and Rodhe, 1948 studied algae of lakes in relation to pH. It was studied by many scientists that pH is very much related to the periodicity of algae. That means a particular species requires a particular pH for every physical and physiological activities leading to growth, reproduction etc. (Vepenskaja, 1924; Howland, 1931; Daily, 1938; Hustedtta, 1938 and Jorgencen, 1948). Thus it can be suggested that pH above or below '7', may be toxic to the organism, which the present study elucidates. But in view of the complex of interrelation of physical, chemical and biological factors, such data can not be regarded as final (Chu, 1942-43), it is therefore described that the conclusions arrived through field studies should be verified through culture studies (Pear-sall, 1932). It may be recognised at the same time that the culture observation in themselves are totally reliable (Lund, 1947), because of the obvious environmental difference between the natural conditions and the cultural condition.

As a pond ecosystem contains various phytoplankton as well as zooplankton and even large fishes, there is every possibility of changing in pH values of water. This may be mainly due to their photosynthetic and respiratory activities. Thus all living organisms in the pond might be facing different pH conditions. It was also put forth by several scientist that air in towns contains SO₂ gases which infact is highly soluble in H₂O and is much heavier than air. Therefore it might be
receiving by many waterbodies like ponds, lakes, rivers etc. with rain as well as runoff water affect all aquatic organisms. This may be either toxic or beneficial, which also may change pH of water bodies. Thus creative interest has been made in this study to know the effect of SO$_2$ in terms of pH values on _S. setiformis_. 
**MATERIALS AND METHODS**

*Spirulina setiformis*, a fresh water green algae was collected from the ponds at fish farm Sambalpur. Healthy and matured specimen were isolated as described in previous chapter (Chapter \(\text{A}\)).

Ambient pond water was boiled, cooled and filtered to make it free from micro-organisms debris. Then it was taken in 4 glass beakers (curing). Out of four, one was treated as normal. Remaining three beakers with ambient pond water were taken and \(\text{SO}_2\) gas was allowed to mix thoroughly with the said water till the change of pH to 2, 4 and 6 respectively. pH value was recorded with the help of high ranged pH metre (electronics) as well as pH paper (BDH).

The healthy and adult specimens were transferred to each beaker and were kept for three hours under laboratory condition.

The specimens were taken after 1h, 2h and 3h interval from the experimental beaker along with normal specimen for scientific study under compound microscope and microphotographs were taken to record histomorphological changes if any.
OBSERVATIONS

Fig. 14a shows normal *spirogyra setiformis* filament having pH value of 7.5.

The filament taken after being treated at pH 2 showed drastic damage of the cell and its components. At 1h exposure timing certain cells were found to be affected showing shifting of whole chloroplast aside of cells, complete disruption in cellular components, pyrenoid bodies were indistinguishable and disintegration of chloroplast with chlorophyll pigment.

In certain cells of the filamentous algae though the spiral arrangement remained undisturbed brownish colouration was well marked at the peripheral portions of total number of pyrenoid bodies (Fig. 16d).

When the specimens (*spirogyra setiformis*) was exposed for 2 hour duration into the toxicant, certain positive effects were observed. However after 3h all the cells were found to be uniformly but adversely affected. The effect was evident which showed solubilization of the chloroplast indicated by green mass inside the cell. Partition wall between two cells were found to be slightly ruptured in certain cells (Fig. 16b).

At pH-4 of the treated solution *S. setiformis* showed less effect than was observed at pH-2 with 1h exposure dissolution of the chloroplast was seen. However very few cells remained unaffected. Spiral arrangement of filaments were found to be normal (Fig. 16c). At same pH with 2h and 3h timing exposure the adverse effect was a must with peripheral mobilization. Even in some cells normal round shape of pyrenoid bodies
Fig 16. Effect of $SO_2$ at different pH on *Spirogyra setiformis*
(f) pH 6 3h  
(e) pH 6 1h  
(d) pH 4 3h  

Fig. 16. Effect of $SO_2$ at different pH on *Spirogyra setiformis*
were changed. These may be divided into smaller units. Chloroplast bands were invisible. Position of the chloroplast was further localised at the central core of the cell (Fig. 16, c).

At pH-6 with 1h exposure spirally arranged chloroplast was disturbed a little with slight mobilization (Fig. 16, e). On increase of exposure timing up to 3h duration, the normal green colour of the chloroplast filament was changed into brownish with little contractions of the whole chloroplast material. Pyrenoid bodies were also affected (Fig. 16, f). However the effect at 3h was less.
DISCUSSION

Sulphur dioxide is a highly poisonous, colourless gas, much heavier than air and readily soluble in water. It is present in the air of industrial area to a harmful extent and pertains suffocating as well as pinching odour. It is used as a bleaching agent. Thus the property itself proves to be toxic to living organisms. The remarkable property of SO$_2$ is its solubility in water and changes the pH of the medium. This in turn effects the organisms readily (Robertson, 1958).

Benerjee, 1967 and Agarwal, 1980 opine that pH between 6.5 to 7.5 is favourable for productivity, which is correlated in the present study because below the pH range of 2 and 6 filamentous cells are affected particularly showing damage to the chloroplast as well chlorophyll pigment (The seed of photosynthesis). Scientist other than Bayer et al, 1963; Bayer, 1964 and Benerjee, 1967 have also worked on pH in relation to organism. The decolourisation of chloroplast may be due to bleaching action of SO$_2$ on _S. setiformis_ which corroborates the findings of Gilbert, 1951 and Mitra, 1966. SO$_2$ when comes in contact with water unstable H$_2$SO$_3$ (sulphurous acid) was formed which may bring about a drastic damage to the cell organelles. At pH-2, toxicity gets higher may be due to higher SO$_2$ concentration.

At higher pH value of 6, minimum effect was observed in _S. setiformis_. Thus it proves that this species can thrive well at high pH (i.e. in alkaline medium. This is further correlated in the present study because _S. setiformis_ was largely found in the pond, where pH in water recorded was between the range of 7 to 8.5. At pH-6 pyrenoid bodies are badly affected indicated by dark brownish ring at peripheral region. It appears that SO$_2$ exhibits high affinity towards starch (i.e.
pyrenoid is the site of starch formation. Vashista, 1983). In the present experiment, toxicity of SO$_2$ was observed to increase with the decrease of pH value of the medium. This shows that acidic medium has a lethal effect on _S. setiformis_. SO$_2$ is found toxic to organisms because of the fact that it interacts with water to form H$_2$SO$_3$, which creates a toxic condition for the organisms. The solubilization of the chloroplast was also observed at low pH. It may be due to the action of sulphurous acid on the organism. In some filaments, it was observed that except pyrenoid, rest of the chloroplast band was found to be disintegrated. This shows that pyrenoids which are in close contact with each other, are more susceptible to low pH than rest of the filament. The filament in control sample having pH 7.5 on exposure to 3h timing imparts no effect. Thus it can be concluded that low pH shows acute toxicity on _S. setiformis_. Whereas comparatively low alkalinity values are considered to be safe for the organisms.

According to Gonzalves and Joshi (1946) low pH is always associated with high CO$_2$ concentration in a medium which may be considered as one of the plausible reasons for the damage of a living cell.

The above laboratory observation shows quite clearly that though _S. setiformis_ can flourish well in little alkaline medium, a little acidic medium is also not very much dangerous to the said algae. Whereas as strictly acidic medium has been proved toxic to _S. setiformis_.

PART IV (B)

Physiological Changes

1. Effect of certain heavy metal pollutants, Insecticides and Aromatic oils on the Primary Productivity of Spirogyra setiformis.