

## Spatial and temporal variation of phytoplankton in hot spring of Atri, Odisha, India

A.Dash<sup>1</sup>, S.S. Panda<sup>3\*</sup>, S.K. Palita<sup>2</sup>, H.K. Patra<sup>1</sup> and N.K. Dhal<sup>3</sup>

<sup>1</sup>P.G Department of Botany, Utkal University, Bhubaneswar, India.

<sup>2</sup>Dept. of Zoology, Kendrapara Autonomous College, Kendrapara, India.

<sup>3</sup>Institute of Minerals and Materials Technology (IMMT), Bhubaneswar, India

### Abstract

Seasonal dynamics of phytoplanktons along with various physicochemical parameters were recorded in the hot spring of Atri, Odisha during the year 2010-2011. The study was carried out to ascertain the phytoplanktons diversity in relation to the changing physico-chemical parameters in an extreme condition of the hot spring on seasonal basis. Total twenty eight genera having forty two species of phytoplanktons were recorded under the following classes, viz. Chlorophyceae, Cyanophyceae Bacillariophyceae and Euglenophyceae. Chlorophyceae was found to be dominating class contributing 40% of the total. The highest numbers of phytoplankton species were recorded during summer season with an average of 11, 3,000 cells/liter. Dominant species identified were *Anabaena aequalis*, *Closterium ehrenbergii* Meneghini ex Ralfs, *Eudorina* sp., *Merismopedia punctata* Meyen, *Microcystis aeruginosa* (Kützing) Kützing, *Navicula membranacea* Cleve, *Oscillatoria* sp., *Pediastrum simplex* Meyen, *Scenedesmus quadricauda* Chodat. Correlation studies of phytoplankton with physicochemical variables indicate a significant positive correlation with chlorophyll-a, alkalinity and nitrate concentration at  $p \leq 0.01$  and  $p \leq 0.05$ .

**Keywords:** Correlation studies, Correlation studies, hot spring, physicochemical parameters, Seasonal dynamics

### INTRODUCTION

Phytoplanktons are the principal components of any water body; the tolerance limits of these organisms to diverse stresses assume tremendous relevance from the ecological standpoint. It is one of the initial biological components from which the energy is transferred to higher organisms through food chain [1-6]. They also play important roles in the material circulation and energy flow in the aquatic ecosystem. Phytoplankton abundance and composition in an aquatic ecosystem are regulated by various abiotic or environmental factors such as pH, light, temperature, salinity, turbidity and nutrients [7-9]. According to Ramakrishnan *et al.*, 97.8% of the variations in phytoplankton density of the freshwater pond were influenced by physico-chemical factors [10]. Besides, their importance as the primary producers in food webs and ensuing ecological balance, phytoplankton are useful indicators of water quality [11]. Species diversity responds to changes in particular to stresses and limiting factors, thus reflecting many interactions which may characterize communities. Changes in any environmental factor will consequently change diversity.

Hot spring is a place where warm or hot ground water escapes from the earth on a regular basis for at least a predictable period and is significantly above the ambient ground temperature. The water released from a hot spring is heated by geothermal energy. Geothermal energy is high heat energy stored in some

favorable geological structures within the top few kilometers of the earth's crust which can be used as energy source. The distinguishing features of these specialized habitats their elevated temperature, depressed dissolved oxygen level and high radioactivity [12, 13]. Extensive studies were carried on the diversity of phytoplankton in thermal springs of U.S.A, Eastern and Western Europe, Japan, Israel, New Zealand, Africa, Germany, Thailand, and Australia [14-16]. But very little work has been carried out in India and especially in the hot springs of Odisha by Adhikary & Sahu and Dash *et al.* [17-19]. The aim of the present study was to determine the taxonomic composition of the phytoplanktons and their abundance for delineation of their temporal pattern, and to study their basic ecological pattern during an annual cycle in the natural hot spring at Atri of Odisha during the year 2010-2011.

### MATERIALS AND METHODS

#### Study area

Atri (20°15'N and 85°30'E in 20°15'N and 85°30'E) is situated in the eastern part of Odisha, in a small village at Baghmari under Khurda district, at a distance of 43 kms from Bhubaneswar. The thermal spring, Atri has circular main tank of 161" diameter and 168" depth (artificially constructed) from where gases escape from the bottom in the form of bubbles. It has a rocky bottom but the water depth is very deep (139"). The water being extremely hot near the origin is channelized into three nearby cemented bathing tanks called overflows where people take their dip. The present investigation was carried out in main tank and its three overflows. Figure 1 shows the schematic diagram of the study site and its overflows path.

Received: July 11, 2012; Revised: Sept 10, 2012; Accepted: Dec 28, 2012.

\*Corresponding Author

S.S. Panda

P.G Department of Botany, Utkal University, Bhubaneswar, India.

Tel: +91-09437155933

Email: [swati.sucharita8@gmail.com](mailto:swati.sucharita8@gmail.com)

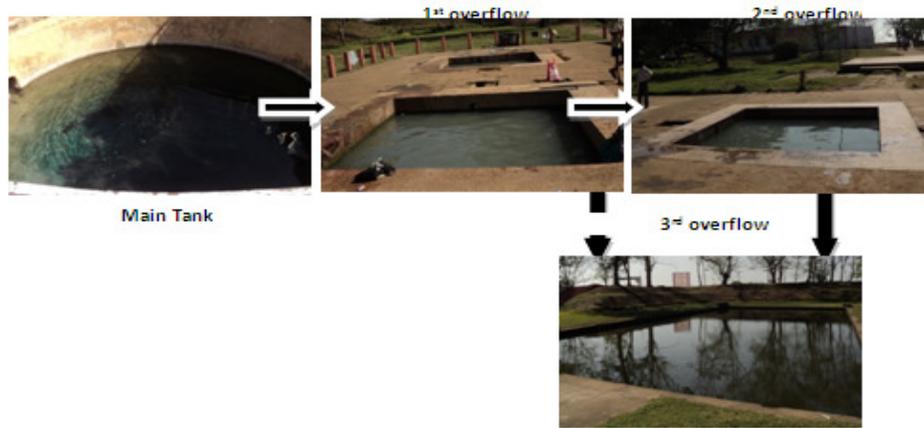


Fig 1. Locations of the overflows of Hot Springs at Atri from the main tank *Sampling and analysis*

The phytoplankton samples were collected from the surface with the help of Niskin water sampler. The sample were preserved with 4% formalin and stored in the laboratory prior to qualitative and quantitative analysis.

Surface water samples were also collected for various physical and chemical analysis following standard procedures and methods as outlined in standard methods [20]. Surface water temperature, pH and EC were measured *in situ* by using WTW kit. Salinity, turbidity, total chloride, fluoride, sulphate, phosphate, nitrates, calcium, magnesium, total iron was measured by standard methods of APHA.

The samples for the analysis of nutrients and chlorophyll-a were preserved in ice box and brought to the laboratory and analyzed immediately. The water samples were filtered through GF/C filter paper and the filtrates were taken for the analysis of nutrients and the residue for chlorophyll-a analysis. The concentration of chlorophyll-a was estimated by the method of Strickland and Parsons [21]. The nutrient contents (nitrate, sulphate, phosphate) were determined in duplicate samples by UV- Visible spectrophotometer (Perkin-Elmer No. Lambda 35) [22].

**Phytoplankton Analysis**

For quantitative phytoplankton analysis, two liters of sample were left to settle for one week and analyzed by a using Sedgewick-Rafter cell counter. An Olympus BX-50 and CK-40 model were used for examination. The identification of the species and their taxonomic categories were given according to Subramanyan and Desikachari

[23, 24].

**Statistical Analysis**

Relationships among variables were explored using Pearson Correlation coefficients. Statistical correlations were done using the program SPSS 13.0 and included all seasonal data during the year 2010-11.

**RESULT AND DISCUSSION**

All the biological and physicochemical parameters studied showed noticeable seasonal as well as spatial variations, which may be attributed to the local climatic conditions and water exchange mechanism between main tank and its overflows.

Temperature is an important factor which regulates the biogeochemical activities in the aquatic environment. During the investigating period, the water temperature varies from 25.4 to 56.8 °C and remains fairly high at the main tank which is the source point of the hot spring and decreases subsequently in the overflows. It was observed that the periodic fluctuation of the temperature of the spring was significant and minor variations may be due to the change in climatic temperature. Highest water temperature was always recorded in the main tank in all the three seasons (Figure 2). One of the noticeable features of the study was the temperature remained high during rainy season. In an aquatic system the water temperature may be regarded as an important factor in maintaining the periodicity and abundance of biological communities [25].

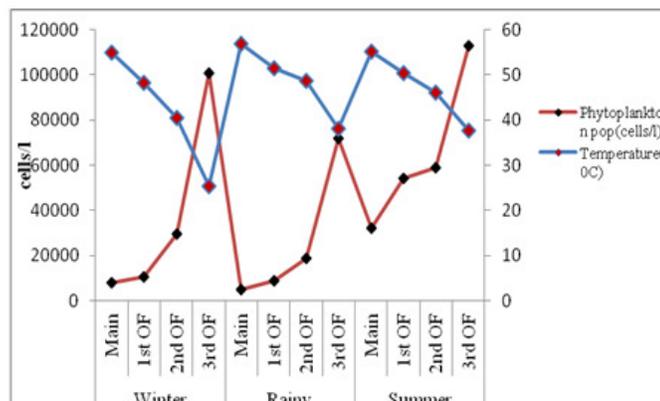


Fig 2. Distribution of phytoplankton species and temperature at different sampling sites (Main Tank- MT, Overflow-OF) on seasonal basis

The pH of water was alkaline in nature and varied from 8.5 to 8.9. High pH value may be due to low water level and high photosynthesis of phytoplanktons resulting in high production of free carbon dioxide and approaching towards alkalinity [26, 27]. Electrical conductivity (EC) was found to be good indicator of the water quality [28, 29]. In the present study, electrical conductivity varied from 670 to 1078  $\mu\text{S}/\text{cm}$  and found lowest in 3<sup>rd</sup> overflow in the rainy season and highest in 1<sup>st</sup> overflow during winter season. This might be due to high anthropogenic activities.

The alkalinity is the main physical parameter that can be attributed to the phytoplankton diversity [30, 31]. It ranged from 15.00 to 48 mg/l and highest value was recorded in 2<sup>nd</sup> and 3<sup>rd</sup> overflow during winter season. According to Klein, Shrivastava and Patil, the alkalinity is directly related to the abundance of phytoplankton since they dissociates bicarbonate into carbonates and carbon dioxide which leads to the increase in alkalinity [32, 33]. The value of hardness ranged from 32 to 54 mg/l. According to Shrivastava and Patil, in water hardness the cations like calcium, magnesium, iron, and manganese contribute to the total water.

Nutrients are essential for survival, reproduction and growth of phytoplankton in an aquatic environment. Phosphate ( $\text{PO}_4^{3-}$ ) is one

of the limiting factors for the productivity of phytoplankton. The phosphate concentration ranged from 0.01 to 0.09 mg/l and highest was recorded in 2<sup>nd</sup> overflow which may be attributed due to the anthropogenic activities. Nitrate ( $\text{NO}_3$ ) is one of the most important indicators of water pollution of water because it is the end product of the aerobic decomposition of organic nitrogenous matters. The concentration of nitrate (0.32 mg/l) was found highest in the 2<sup>nd</sup> overflow during winter season. The concentration of sulphate ( $\text{SO}_4^{2-}$ ) ranged between 27.8 to 36 mg/l during summer season in the second overflow.

The chlorophyll-a concentration ranged from 0.45 to 5.52  $\text{mg}/\text{m}^3$  and highest concentration of chl-a was recorded in the 3<sup>rd</sup> overflow during summer season. The high value of chlorophyll-a concentration may be due to high phytoplankton distribution during the particular site and period.

Phytoplankton plays an important role as primary producers, forming the base of the food chain. Analysis of phytoplankton community composition showed that there were forty species of 3 different algal class identified: 43% of Chlorophyceae, 35 % of Cyanophyceae and 22 % of Bacillariophyceae (Table 1 & Figure 3).

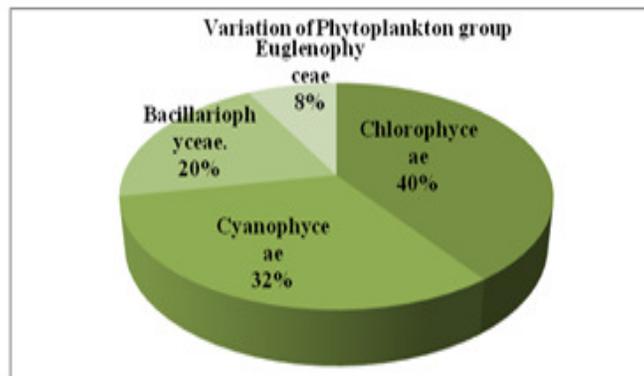


Fig 3. Variation of phytoplankton group in the year 2010-11

Results showed that the low density of phytoplankton distribution in the main tank was attributed to the high water temperature (Figure 4). The maximum phytoplankton species availability was observed in the summer season in all the sampling locations (Main tank and its

three overflows) as shown in Figure 4. This might be due to the moderate temperature and availability of bright light accelerating the rate of photosynthesis.

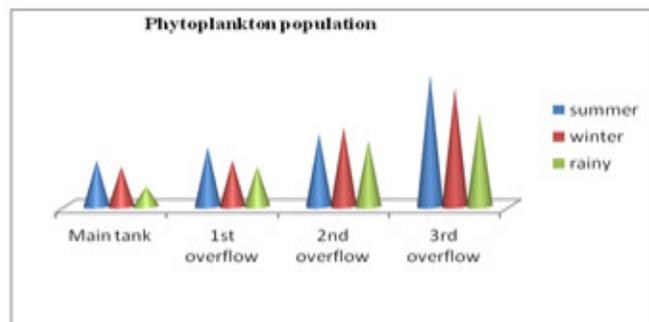


Fig 4. Distribution of phytoplankton species in main tank and its overflows on seasonal basis.

In the main tank the phytoplankton population was very low which might be due the extreme adverse climate and less anthropogenic activities. The highest number of phytoplankton population was recorded in 3<sup>rd</sup> overflow due to high anthropogenic and domestic activities. The diversity of the phytoplankton and its abundance differed in different study sites during the period. In this study it is demonstrated that the phytoplankton species composition and diversity changes in relation to various physico-chemical

parameters. High species diversity coincided with significantly higher relative abundance of chlorophyceae. Dominant species recorded were *Anabaena aequalis*, *Closterium ehrenbergii*, *Eudorina sp.*, *Merismopedia punctata*. *Microcystis aeruginosa*, *Navicula membranacea*, *Oscillatoria sp.*, *Pediastrum simplex*, *Scenedesmus quadricauda*. The seasonal occurrence of individual species in each study site was shown in Table 1.

Table 1. Phytoplankton species in different study sites in summer, rainy and winter

Phytoplankton Species	Summer				Rainy				Winter			
	MT	1 <sup>st</sup> OF	2 <sup>nd</sup> OF	3 <sup>rd</sup> OF	MT	1 <sup>st</sup> OF	2 <sup>nd</sup> OF	3 <sup>rd</sup> OF	MT	1 <sup>st</sup> OF	2 <sup>nd</sup> OF	3 <sup>rd</sup> OF
<i>Anabaena aequalis</i> Borge	-	-	-	+	-	+	-	+	+	-	-	+
<i>Anacystis sp.</i>	-	+	-	+	+	-	-	+	-	+	-	+
<i>Ankistrodesmus falcatus</i> (Corda) Ralfs	-	-	+	+	-	+	-	+	+	-	+	+
<i>Ankistrodesmus sp.</i>	-	-	+	-	-	-	+	+	-	-	+	-
<i>Aphanizomenon flos-aquae</i> Ralfs ex Bomet & Flahault	-	+	-	-	-	-	+	-	-	+	-	-
<i>Chlamydomonas sp.</i>	+	+	+	+	-	+	-	-	-	+	+	+
<i>Chlorellavulgaris</i> Beyerinck (Beijenck)	+	+	+	+	+	+	+	+	+	+	+	+
<i>Chroococcus sp.</i>	+	+	+	+	+	+	+	+	+	+	+	+
<i>Closterium ehrenbergii</i> Meneghini ex Ralfs	+	+	+	-	+	+	+	+	+	+	+	-
<i>Closterium sp.</i>	+	+	+	+	+	+	+	-	-	+	+	+
<i>Cosmarium sp.</i>	-	-	+	+	+	+	+	+	+	-	+	+
<i>Cyclotella sp.</i>	-	+	+	+	-	-	+	+	-	+	+	+
<i>Cymbella lunata</i> W.Smith in Greville	-	+	-	+	+	+	+	+	+	+	-	+
<i>Cymbella prostrata</i> (Berkeley) Cleve	-	-	-	-	-	+	-	+	-	-	-	-
<i>Cymbella sp.</i>	-	-	+	-	-	-	-	-	-	-	+	-
<i>Diatoma elongatum</i> (Lyngbye) C.Agardh	+	+	+	+	-	-	+	-	+	+	+	+
<i>Eudorina sp.</i>	+	+	+	+	-	+	+	+	-	+	+	+
<i>Euglena sp.</i>	-	-	-	+	-	+	-	-	-	-	+	+
<i>Merismopedia elegans</i> G.M.Smith	+	+	+	+	+	+	+	+	+	+	+	+
<i>Merismopedia punctata</i> Meyen	+	-	+	+	+	+	+	+	+	-	+	+
<i>Microcystis aeruginosa</i> (Kützing) Kützing	-	+	-	+	-	-	+	+	-	+	-	+
<i>Microcystis flosaquae</i> (Wittrock) Kirchner	-	-	-	+	+	+	-	+	-	-	-	+
<i>Navicula gracilis</i> Ehrenberg	+	+	+	-	+	-	-	+	+	+	+	-
<i>Navicula lanceolata</i> (C.Agardh) Kützing	+	+	+	-	-	+	+	-	+	+	+	-
<i>Navicula salinarum</i> Grunow	-	-	-	-	-	+	+	-	+	-	-	-
<i>Navicula sp.</i>	+	+	+	+	+	-	-	+	+	+	+	+
<i>Oscillatoria hormagonia</i>	+	+	+	+	+	+	+	+	+	+	+	+
<i>Oscillatoria sp.</i>	+	+	+	-	+	+	+	+	+	+	+	-
<i>Pediastrum simplex</i> Meyen	+	+	+	+	+	+	+	-	+	+	+	+
<i>Pediastrum sp.</i>	-	-	+	-	+	+	+	+	-	-	+	-
<i>Phacus sp.</i>	-	-	-	-	-	+	-	-	-	-	+	+
<i>Scenedesmus acuminatus</i> (Lagerheim) Chodat	-	+	-	+	-	-	+	-	-	+	-	+
<i>Scenedesmus eornis</i> (Ehrenberg) Chodat	-	-	-	+	-	+	-	+	-	-	-	+
<i>Scenedesmus quadricauda</i> Chodat	-	+	-	-	+	-	-	+	-	+	-	-
<i>Spirulina major</i> Kützing ex Gomont	+	+	+	+	+	+	-	-	+	+	+	+
<i>Staurastrum sp.</i>	-	-	+	-	-	+	+	+	-	-	+	-
<i>Surirella sp.</i>	+	+	+	+	+	-	+	-	+	+	+	+
<i>Synechococcus sp.</i>	+	+	+	+	+	+	+	+	-	+	+	+
<i>Synedra sp.</i>	-	-	-	+	-	+	+	+	+	-	-	+
<i>Synedra ulna</i> (Nitzsch) Ehrenberg	-	+	+	-	-	-	-	+	-	+	+	-
<i>Tetraëdron minimum</i> (A.Braun) Hansgig	+	+	+	+	+	+	+	-	+	+	+	+

(Main Tank- MT, Overflow-OF)  
Presence (+), Absence(-)

Table 2. Correlation coefficient matrix between various physico-chemical and biological characteristics in different sampling sites.

Variables	Phytoplankton	Chlorophyll	WT(°C)	pH	EC	Alkalinity	Turbidity	chloride	Flouride	Sulphate	phosphate	Nitrate	Calcium	Magnesium	Iron
Phytoplankton	1														
Chlorophyll	.851**	1													
WT(°C)	-.798**	-.556**	1												
pH	-.544**	-.406*	.393*	1											
EC	.160	.182	-.264	.228	1										
Alkalinity	.352*	.295	-.624**	-.172	.060	1									
Turbidity	.224	.522	.219	-.072	-.036	-.089	1								
chloride	.035	.294	.424*	.250	.061	-.461*	.684**	1							
Flouride	.160	.505**	.374*	-.189	-.182	-.207	.749**	.693**	1						
Sulphate	.255	.374*	-.043	.068	.500**	.022	.378*	.401*	.271	1					
phosphate	.313	.200	-.490**	-.288	-.118	.480**	-.094	-.581**	-.210	-.013	1				
Nitrate	.347*	.459*	-.289	-.115	.075	-.060	.385*	.341	.148	-.084	.017	1			
calcium	.241	.134	-.643**	-.176	-.088	.488**	-.447*	-.584**	-.503**	-.216	.542*	.193	1		
Magnesium	-.295	-.044	.576**	.558**	.025	-.373*	.623**	.724**	.407*	.349*	-	.192	-.601**	1	
Iron	.201	.516**	-.110	-.339	-.327	.305	.319	.010	.491**	-.057	.418*	.225	.400*	-.204	1

\*\* Correlation is significant at the 0.01 level (1-tailed).  
 \* Correlation is significant at the 0.05 level (1-tailed).

The phytoplankton population has showed a positive correlation with the concentration of chlorophyll-a, alkalinity and nitrate at concentration at  $p \leq 0.01$  and  $p \leq 0.05$ , respectively (Table 2). The correlation matrix shows a negative relationship of phytoplankton population with water temperature and pH at  $p \leq 0.05$ .

**ACKNOWLEDGEMENT**

The authors are thankful to Prof. B.K Mishra, Director, Institute of Minerals and Materials Technology and Head of the Botany Department, Utkal University, Bhubaneswar for providing facilities to carry out the work.

**REFERENCES**

[1] Rajesh, K.M., G. Gowda and R.M. Mendon.2002. Primary Productivity of the brackish water impoundments along Nethravathi estuary, Mangalore in relation to some physico-chemical parameters, *Fish Technology*.39: 85-87.

[2] Ananthan, G., P. Sampathkumar, P. Soundarapandian and L. Kannan. 2004. Observation on environmental characteristics of Ariyankuppam estuary and Verampattinam coast of Pondicherry. *J. Aqua. Biol.* 19:67-72.

[3] Tiwari, A. and S.V.S. Chauhan. 2006. Seasonal phytoplanktonic diversity of Kitham Lake, Agra, *J. Environ. Biol.* 27: 35-38.

[4] Sridhar, R., T. Thangaradjou, S.S. Kumar and L. Kannan. 2006. Water quality and phytoplankton characteristics in the Palk bay, southeast coast of India, *J. Environ. Biol.* 27: 561-566.

[5] Shashi, T.R., B.R. Kiran, E.T. Puttaiah, Y. Shivaraj and K.M. Mahadenvan. 2008. Phytoplankton as index of water quality with reference to industrial pollution, *J. Environ. Biol.* 29: 233-236.

[6] Saravanakumar, A., M. Rajkumar, G.A. Thivakaran and J.S. Serebiah. 2008. Abundance and seasonal variations of phytoplankton in the creek waters of western mangrove of Kachchh-Gujarat, *J. Environ. Biol.* 29: 271-274.

[7] Buzzi, F. 2002. Phytoplankton assemblages in two sub-basins of Lake Como, *J. Limnol.* 61: 117-128.

[8] Lewis, M.W. 2000. Basis of the protection and management of tropical lakes and Reservoirs, *Research and Management.* 5: 35-48.

[9] Sin, Y., L.R. Wetzel and C.I. Anderson. 1999. Spatial and temporal characteristics of nutrient and phytoplankton dynamics in the York River Estuary, Virginia, Analyses of long-term data, *Estuaries.* 22: 260-275.

[10] Ramakrishnan, N., P. Hariparasath and S.K. Sampath. 2002. Algal diversity in three freshwater pond ecosystems at Tiruvannamalai, Tamil Nadu, India. In: Symposium on Conservation, Restoration and Management of Aquatic Ecosystems. IISc Campus, Bangalore.

[11] Rey, P.A., J.C. Taylor, A. Laas, L. Hensburg and A.Vosloo. 2004. Determining the possible application value of diatoms as indicators of general water quality a comparison with SASS 5, *Water S.A.*30: 325-332.

[12] Andrews, J.N. 1991. Radioactivity and dissolved gases in the thermal waters of Bath. In: KELLAWAY, G.A. (Ed.), *Hot Springs of Bath.* Bath City Council, Bath. 157-170.

[13] Edmunds, W.M. and D.L. Miles. 1991. The geochemistry of Bath thermal waters. In: Kellaway, G.A. (Ed.), *Hot Springs of Bath.* Bath City Council, Bath.143-156.

[14] Castenholz, R.W. 1970. Laboratory culture of thermophillic cyanophytes. *Schweiz.Z. Hydrol.* 32: 538-551.

[15] Brock, T.D. and M.L. Brock. 1969. Recovery of a hot spring community from a catastrophe. *J. Phycol.*5:75-77.

[16] Stockner, J.G. 1968. Algal growth and primary productivity in a thermal stream. *J. Fish. Res. Board. Can.* 25:2037-2058.

[17] Adhikary, S.P., and J.Sahu.1987. Limnology of thermal springs of Orissa. *J. Bombay Natural History Soc.* 84: 497-503.

[18] Dash, A., S.K. Palita, N.K. Dhal, S.S. Panda, S. Srichandan, H.K. Patra.2011. Phytoplankton and zooplanktons in Hot water

- Spring of Atri, Odisha – A Preliminary Report. Biotechnology and Stresses Management H.K Patra, Eds 41-46.
- [19] Dash, A., S.S. Panda, S.K. Palita, H.K. Patra, N.K. Dhal. 2012. Effect of abiotic stresses on phytoplankton diversity in Hot Sulphur Spring, Atri, Odisha. *Plants and Stress Management* H.K. Patra, Eds. 47-55.
- [20] APHA. 2005. Standard methods for examination of water and wastewater. 21st Edn. APHA, AWWA, WPCF, Washington DC, USA.
- [21] Strickland, J.D.H. and T.R. Parsons. 1972. A Practical Handbook of Seawater Analysis. 2nd ed., Bull. Fish. Res. Bd. Can. No. 167: 310.
- [22] Grasshoff, K., M. Ehrhardt and K. Kremling. 1999. Methods of Seawater Analysis (Verlag Chemie Weinheim, Germany) 159-226.
- [23] Subramanyan, R.A. 1946. Systematic account of marine plankton diatoms of Madras Coast, Proc, Indian Acad. Sci., 24: 5-197.
- [24] Desikachari, T.V. 1986. Atlas of Diatoms, (Madras Science Foundation), Madras.
- [25] Zafar, A.R. 1967. On ecology of algae in certain fish ponds of Hyderabad, India-III. The periodicity. *Hydrobiol.* 30: 96-112.
- [26] Trivedy, R.K. 1989. Limnology of freshwater pond in Mangalore. National Symposium on Advances in limnology conservation of endangered fish species. Oct. 23-25. Srinagar Garhwal.
- [27] Shiddamallayya, N. and M. Pratima. 2008. Impact of domestic sewage on fresh water body. *J. Environ. Biol.*, 29: 303-308.
- [28] Abbassi, S.A., D.S. Arya, A.S. Hameed and N. Abbassi. 1996. Water quality of a typical; river of Kerala, Punnurpuzha. *Pollut. Res.* 15:163-166.
- [29] Gaikwad, S.R., K.N. Ingle and S.R. Thorat. 2008. Study of zooplankton patten and resting egg diversity of recently dried waterbodies in north Maharashtra region. *J. Environ. Biol.* 29: 353-356.
- [30] Kouwenberg, J.H.M. 1993. Copepod distribution in relation to seasonal hydrographic and spatial structure in the north-western Mediterranean (Gulf du Lion) *Estuar. Coastal Shelf Sci.* 38: 69-90.
- [31] Ramaiah, N and V. Nair. 1997. Distribution and abundance of Copepods in the pollution gradient zones of Bombay harbour, Thane, Creek-basin creek, West coast of India, *Ind.J.Mar.Sci.* 26: 20-25.
- [32] Klein, L. 1959. River Pollution Chemical Analysis. I. Causes and Effects. Butterworth and company, London. I: 29-31.
- [33] Shrivastava, V.S. and P.R. Patil. 2002. Tapti River water pollution by industrial wastes: A statistical approach. *Nat. Environ. Pollut. Tech.* 1: 279-283.