Chapter-XI

General Discussion
General Discussion

Of all the planet's renewable resources, water has a unique place. It is essential for sustaining all forms of life, for food production, and also for economic development. Water often is called "the universal solvent" because of its extraordinary ability to dissolve a broad range of substances. In fact, it dissolves more substances in greater quantities than any other liquid (Driscoll, 1989).

Many seasonal studies on fresh waters have only documented environmental (physico-chemical) parameters, but there are no studies comparing the environmental (physico-chemical) parameters of the ground water from six different sampling sites from Agricultural, Domestic and Industrial areas of Mysore district, Karnataka State, India as carried out in this study. Therefore, an attempt has been made here, for the first time, to compare and contrast the environmental (physico-chemical) parameters in the ground waters of Agricultural, Domestic and industrial areas of Mysore district, Karnataka State, India. The results described in the Chapter 3 showed that, most of the environmental (physico-chemical) parameters in the ground water of Industrial area were comparatively more when compared to the ground water of Domestic and Agricultural areas during study period, which is mainly due to mixing of industrial and sewage effluents in ground water of Industrial area. Similarly, it is the first comprehensive ecological study of micro-biota of the ground water from six different sampling sites from Agricultural, Domestic and Industrial areas. An attempt has been made here for the first time to compare and contrast the abundance of heterotrophic bacterioplankton in the ground waters of Agricultural, Domestic and industrial areas. The results described in the Chapter 4 showed that the ground water of Agricultural
and Industrial areas were comparatively less favorable habitat for the abundance of heterotrophic bacteria as compared to the ground water of Domestic area. The evidence for this conclusion was that, mean values of abundance of heterotrophic Free Living Bacteria (FLB), Particle Bound Bacteria (PBB) and Total Bacteria (TB) were high in the ground water of Domestic area when compared to Agricultural and Industrial areas, both of which were almost similar to each other in having less abundance of FLB, PBB and TB. High bacterial abundance in the ground water of Domestic area when compared to other areas in this study, may be due to the less depth of the ground water in the study area, where the accumulated material is more due to less effectiveness of filtration process to remove them through soil layers as reported by Strayer (1994) who studied limits to biological distributions in groundwater ecology or may be due to the availability of nutrients (Wright and Coffin, 1983 and 1984; Shiah, 1993; Wommack and Colwel, 2000) and also due to the effect of the distance of this sampling site from the pollutants source. However, less bacterial abundance in Industrial and Agricultural areas may be due to the location of these sampling sites far-away from the pollutants source - similar findings were reported by Marxsen (1981), Harvey et al., (1984), Harvey and George (1987) and Godsy et al., (1992). The increased bacterial abundance during summer season may be due to the increased temperature - similar investigation was reported by Jonas and Tuttle (1990).

The abundance of Colony Forming Units (CFUs) were found more in the ground water of Domestic area followed by Industrial area, suggesting probably that the natural habitat in both of these areas, is not favorable for normal replication of microbes. Interestingly, no seasonal variations were observed in CFUs of the ground water in all sampling sites during study period.
There was no growth of CCFUs in the ground water from Agricultural, Domestic and Industrial areas during study period. Which may be due to the non acidic condition of these ground waters similar findings have been reported by Goulder (1988) on Epilithic bacteria in an acid and a calcareous head stream; Simon and Jones (1992) study on the absence of bacteria from acid waters in Northwest England; Yamakanamardi (1995) on Microbial ecology of three contrasting lowland water courses in Northeast England and Harsha et al., (2006b) on Temporal variations in the abundance of heterotrophic bacteria in the river Cauvery and its down stream tributaries, Karnataka, India and Mahadaevaswamy et al., (2006) on microbial ecology of main river Cauvery and its four important upstream tributaries in Mysore district of Karnataka state.

In this investigation, as much as 2.79% (in Agricultural area) and 3.60% (in Industrial area) of total bacteria could be grown on the artificial nutrient agar media. Whereas, the bacterial communities of ground water from Domestic area were found growing more (4.69 %) in the artificial nutrient agar media. It is noteworthy that the lowest mean CFUs as % of AODCs (1.85%) recorded in the ground water of Agricultural area and the highest value of 6.44% recorded in the ground water of Domestic area were the lowest and the highest recorded values of CFUs as % of AODCs during study period.

The abundance of TB and FLB were affected by more number of environmental (physico-chemical) parameters and the extent to which they affected was also more, when compared to the abundance of PBB. Furthermore, 9% variations in the concentration of CFUs was due to Alkalinity in the ground water of Agricultural area, 13% due to SO$_4$ (+) in the ground water of Domestic area and no environmental (physico-chemical) parameters showed correlation and regression with
CFUs in the ground water of Industrial area. There was no growth of CCFUs in any of the ground water from six sampling sites from Agricultural, Domestic and Industrial areas, in both First and second year of the study. Furthermore, there was no environmental (physico-chemical) parameters showed correlation and regression with CFUs as % AODCs in the ground waters of Agricultural Domestic and Industrial areas during study period. The entire data given in tables 5.4-5.6 suggest that several key environmental (physico-chemical) parameters were potentially responsible for some of the variations in the bacterial abundance.

Apart from this, the other environmental (physico-chemical) parameters such as F pH, L pH, Turbidity, CO₂, Chloride, TASA, Sulphate, Hardness, Calcium, magnesium, Alkalinity, TDS, TS, TSS, BOD, Conductivity, air and water Temperature, Rainfall and Nitrate also entered in the regression equation and thus participated in deciding the abundance of bacterioplankton. Similar observations were reported by Anesio et al., (1997) on influence of the hydrological cycle on the Bacterioplankton of an impacted clear water Amazonian lake, Mohamed et al., (1998) on phosphorus limitation of heterotrophic biofilms from the Fraser river, British Columbia and the effect of pulp mill effluent; Kirschner et al., (1999) on Material fluxes through the prokaryotic compartment in a eutrophic backwater branch of river Danube; Mitsuru et al., (2000) on seasonal variation of two different heterotrophic bacterial assemblages, in sub arctic coastal seawater; Lindstrom, (2001) on investigating influential factors on Bacterioplankton community composition: results from a field study of five mesotrophic lakes; Heidelberg et al. (2002) on Seasonality of Chesapeake Bay and Castillo et al. (2004) on Seasonal and interannual variation of bacterial production in lowland rivers of the Orinoco basin. However, this study revealed that, more number of factors in the ground water of Agricultural and
Domestic areas and less number of factors affected the bacterial abundance in the ground water of Industrial area. Hence, it can be concluded that abundance of bacterioplankton in all ground water studied may have been largely controlled by environmental (physico-chemical) parameters, which is in agreement with other such studies on ground water, temperate streams, rivers and coastal waters - on origins, characterization and dynamics of suspended bacteria in two chalk streams (Baker and Farr, 1977) on seasonal variations in heterotrophic activity and population density of planktonic bacteria in a clean river (Goulder, 1980) on seasonal variations in abundance and heterotrophic activity of suspended bacteria in two lowland rivers; (Goulder, 1986) on Untersuchungen zur Okologie der Bakterien in der flissenden Welle; (Marxsen, 1980b) on Bacterial biomass and bacterial uptake of glucose in polluted and unpolluted ground water of sandy and gravelly deposits; (Marxsen, 1981) on the effect of environmental factors on the suspended bacteria in Welsh river Dee and on The populations, characterization and activity of suspended bacteria in the Welsh river Dee; (Nuttall, 1982a and 1982b) on survival strategies of bacteria in the natural environment; (Roszak et al., 1987) on Microbial investigations in rivers VII. Seasonal variations of bacterial numbers and activity in eutrophied rivers of Northern Germany; (Gocke and Rheinheimer, 1988) on Inorganic phosphorus stimulation of bacterioplankton production in a mesoeutrophic lake; (Toolan et al., 1991) on Temporal variability of attached and free-living bacteria on coastal waters; (Unanue et al., 1992) on groundwater microbiology ;(Madsen and Ghiorse,1993) on Activity and abundance of bacterioplankton in three diverse lowland water courses; (Yamakanamardi and Goulder, 1995) on Phosphorus limitation of heterotrophic biofilms from the Fraser river, British Columbia and the effect of pulp mill effluent; (Mohamed et al., 1998) on Distribution and composition of microbial populations in a
landfill leakage contaminated aquifer Grindsted, Denmark; (Ludvigsen et al., 1999) on diversity and activity of microorganisms in subsurface igneous rock aquifers of the Fennoscandian Shield; (Pedersen, 2001- In: Subsurface Microbiology and Biochemistry) on Seasonal and interannual variations of bacterial production in lowland rivers of the Orinoco basin; (Castillo et al., 2004). All these studies suggested that, seasonal variations in abundance of bacterioplankton are substantially dependent on environmental (physico-chemical) parameters.

The size of bacteria is an important trait in the predator-prey relationship of aquatic bacteria and bacterivorous protists. As grazing by bacterivorous protists (flagellates and ciliates) is size selective (González et al., 1990; Šimek and Chrzanowski, 1992). Thus, small and large bacteria may have a refuge from protistan grazing. Filament formation or permanent filamentous growth is one highly effective, size dependent grazing defense mechanism of aquatic bacteria (Hahn et al., 1999 and 2000). Experimental field studies have demonstrated that natural bacterial community with respect to cell size was mainly dependent on physical and chemical factors, such as temperature, substrate availability and nutrient supply and protistan grazing pressure (Hahn and Höfle, 1999). These are related to cell volume and biomass, although not linearly. Cell breadths were not measured, hence calculation of biovolume and biomass was not possible. In this study bacterial cell size (mean length) and size category distribution was measured over a period of two years (Feb. 2005 – Jan. 2007). Bacterial cell size varies either inversely or independently of trophic conditions (Van ES and Meyer-Reil, 1982; Bird and Kalf, 1984). Based on recent intercamparision of cell size measurements it would appear that much of the reported variations in size is related to methodology which varies among investigators (Cole et al., 1993). Nevertheless cell lengths were likely to be related to physiological
state of cells. During present investigation surprisingly men length of both FLB and PBB were similar in all the ground waters studied.

Temporal variations in the mean cell length of both FLB in the ground water of Agricultural area fluctuated during both first and second year of study, in Domestic area this variations was only in the second year of study, whereas, in Industrial area it was only in the in the second year of study, thus variations may be due to the variations in food supply and the grazing pressure (Wright, 1988; Callieri and Heinimaa, 1997). The examination of correlation between mean cell length of FLB and PBB with environmental (Physico-chemical) parameters showed the presence of many correlations suggested that, the greater availability of nutrients and organic substrates such as Chloride, TASA and Calcium contributed by anthropogenic inputs like discharge of untreated effluent and domestic sewage for growth and reduced predation on larger cells as reported by Cole et al., (1993); Callieri and Heinimaa, (1997); Schauer et al., (2005).

The regression analysis reveled that as much as 12% of variations in the mean length of FLB was due to NO₃ (+) in Agricultural area and 9% due to Total Solids (+) in Domestic area. However, the 8 % variations in bacterial cell size of FLB were due to Mg (+) in Industrial area (Table 6.11). Whereas, 13 % variations in mean length of PBB were due to SO₄ (+) in Agricultural area and 10% of variations because of TASA (+) in Domestic area. Furthermore, 10 % of variations were due to PO₄ (+) in Industrial area. This kind of relation between mean length and environmental (physico-chemical) parameters suggest that the, Temperature, Calcium, SO₄, Chloride, SO₄, TASA, Total Solids, NO₃ and BOD were directly responsible for the observed changes. This was in agreement with Jugnia et al., (2000) in a flooded Sep reservoir of France. The other reason for the such dependence of bacterial cell size
may be due to substrate availability, nutrients, because bacterial growth in terms of
size probably maintained at a maximum level by a density dependent factors such as
carbon or other nutrients as proposed by Gasol and Vaque (1993) on Lack of coupling
between heterotrophic nanoflagellates and bacteria: Jonathan et. al (1993) and
Ekebom (1999). The obvious conclusion is that, though the bacterial mean cell length
was similar in all samples, yet different sets of environmental (physico-chemical)
parameters were apparently controlling cell-size in different ground water studied in
this investigation. Similar observations were made for the hypertrophic Humboldt
Lake and oligotrophic Redberry lake in Sasktchewan, Canada (Tumber et al., 1993),
in recently flooded oligo-mesotrophic reservoir, France (Jugnia et al., 2000) and in
sediments of Botany bay in Sydney, Australia which is fed by two rivers, the Cooks
river and the Georges river (Lee and Patterson, 2002).

The specific growth rate (SGR), determined by dividing the production
between the biomass, is an indicator of the capacity of the bacterial population to
replace its biomass (Irberri et al., 1990). Heterotrophic bacteria are thought to be
superior competitors for P to phytoplankton in a variety of pelagic environments,
mainly in oligotrophic ones (Cotner and Biddanda, 2002; Šimek et al., 2006). In
planktonic systems, algae (Bird and Kalff, 1984; Cole et al., 1988), inorganic
nutrients (Currie, 1990; Le et al., 1994), allochthonous dissolved organic C (DOC)
(Findlay et al., 1991; del Giorgio el al., 1996) and temperature (White et al., (1991),
Shiah and Ducklow, (1994); Felip et al., (1996); Yamakanamardi and Goulder,
(1999); Rier and Stevenson, (2001) have all been identified as factors that influence
the biomass, productivity and growth of bacteria. In general, SGR of bacteria (k) in all
the ground water samples fluctuated over a two year period, but was largely not
related to bacterial parameters. This suggested that the temporal changes in abundance
of bacterioplankton in these ground water samples were not primarily caused by underlying variations in water quality or stress levels which influence k. The temporal changes in abundance might be more related to other parameters, grazing pressure of microzooplankton or other water quality parameters. High bacterial growth during summer season when compared to winter season suggested that, the substrate supply might be the major factor regulating bacterial growth (Shiah and Ducklow, 1994) or may be due to the effect of water temperature which is more in summer season when compared to winter and rainy seasons same result was reported by (Jonas and Tuttle 1990). In conclusion, the lack of interrelationship between SGR and other bacterial parameters suggested that the other constraints like resource limitation coupled with grazer mortality, predators-prey interactions (Chrzanowski et al., 1995; Chrzanowski and Grover, 2001) might have played dominant role in controlling bacterial growth rate (Carlsson and Caron, 2001). According to the regression analysis SGR tended to respond more strongly towards environmental (physico-chemical) parameters (e.g. water Temperature, PO₄ and TS) than other bacterial parameters which might have caused physiological stress, which in turn regulated the SGR in all ground water samples studied. This study suggested that the SGR is potentially useful in detection of inhibition of bacterioplankton, and potential loss of biopurification capacity, brought about by adverse water quality which may be related to natural process or to toxic pollution (Yamakanamardi and Goulder, 1999) and the effect of nutrients on SGR same results were reported by Toolan et al., (1991); Coveney and Wetzel (1992); Le et al., (1994) and Elser et al., (1995).

The two most useful divisions of the Coliforms group of bacteria are the “Total Coliforms (TC) group” and the “Fecal Coliforms (FC)” a sub-group of Coliforms (Geldrech, 1967). The TC group is defined as “all the aerobic and
facultative anaerobic, gram negative, non-spore-forming, rod-shaped bacteria which ferment lactose with gas formulation within 48 hours at 35°C; whereas, FC subgroup as all those Coliforms bacteria "which ferment lactose with gas formation within 24 hours at 44-45°C". The lowest mean TC bacteria 2.58 colonies per 100 ml recorded in the ground water of Industrial area and the highest value of 13.77 colonies per 100 ml recorded in Domestic area were the lowest and the highest recorded values of TC during study period. The increased quantity of wastewater in the Domestic area, may probably be the reason for the increased number of TC in the ground water of Domestic area when compared to Agricultural and Industrial areas, which is in agreement with findings of EEA (1999) and USEPA (2000).

The presence of FC implies the potential presence of microorganisms that are pathogenic to humans. Fecal Coliforms bacteria have a strong correlation with fecal contamination of water from warm-blooded animals. If 1 Fecal Coliforms per 100 mL of water is detected, the water is considered unsafe to ingest (Greenberg et al., 1992; USEPA, 1998). This indicates that the ground water of Agricultural and Domestic areas are not safe for human consumption, as in these ground waters there were more than 1 FC per 100 ml.

There were more significant correlation between TC and FC bacteria in the ground waters of Agricultural and Domestic area, similar results were reported by Eckner (1998) and Noble et al., (2003). However, the was no growth of FC bacteria the ground water of Industrial area. However, there was growth of TC only, which indicates that, there is no fecal contamination and the presence of TC in the study area was from other sources like soil and vegetation (Greenberg et al., 1992).

It can be concluded that Coliforms bacteria in all samples may have been controlled by some environmental (physico-chemical) parameters, which is in
agreement with studies on TC and FC in natural spring water as related to recreational mountain areas by Youn and Peter (2005), Changes in chemical and physical properties of stream water across an urban-rural gradient in western Georgia by Jon et al., (2005) that suggested, seasonal variations in Coliforms bacteria is substantially dependent on some environmental (physico-chemical) parameters.

Trace elements in subsurface environments come from natural and anthropogenic sources. The weathering of minerals is one of the major natural sources. Anthropogenic sources include fertilizers, industrial effluent (Chi-Man Leung and Jiu Jimmy Jiao, 2006). As far as this author knows, there are no studies available on aquatic metals from ground water of Agricultural, Domestic and Industrial areas of Mysore district, Karnataka State, India.

The concentrations of aquatic metals were within the permissible limits as suggested by WHO (1993) in all ground waters from Agricultural, Domestic and Industrial areas studied. However there were some aquatic metals (Pb, Zn and Ni) not detected in all sampling sites.

It can be concluded that microbial (bacterial) parameters in all samples may have been largely controlled by few aquatic metals which is in agreement with studies of Marxen (1988) who studied on investigation into the number of respiring bacteria in groundwater from sandy and gravelly deposits; Balkwill (1989) on Numbers, diversity, and morphological characteristics of aerobic, chemoheterotrophic bacteria in deep subsurface sediments from a site in South Carolina; Bengtsson (1989) on Growth and metabolic flexibility in groundwater bacteria; Toolan et al., (1991) on Inorganic phosphorus stimulation of bacterioplankton production in a meso eutrophic lake.; Morris and Lewis (1992) on Nutrient limitation of bacterioplankton growth in Lake Dillon, Colorado; Chapelle (1993) on groundwater microbiology and

Geographical Information System (GIS) can be defined as: An organized collection of computer hardware, software, geographic data, and personnel designed to efficiently capture, store, update, manipulate, analyse, and display all forms of geographically referenced information (ESRI, 1996). GIS technology integrates common database operations such as query and statistical analyses with the unique visualisation and geographic analysis, benefits offered by maps and spatial databases. These abilities distinguish GIS from other information systems and make it a valuable tool for explaining events, predicting outcomes, and planning strategies.

GIS has increasingly been used for water and environmental problems such as water quality prediction (Golojuch, 1994), water management (Tremblay, 1994 and Barbera et al., 1994) and impacts to and vulnerability of groundwater resources (Civita and De Maio, 1997; Navulur and Engel, 1997; Yang et al., 1999b; Ducci, 1999). As for as this author knows, there are no studies available on comparative studies of water quality from ground water of Agricultural, Domestic and Industrial areas using GIS application.

Generally, water quality of the ground water of the study areas can be represented in thematic maps using GIS tools with different colours which is indicated the
concentration of the studied parameters. From the legend (index) of each map we can get clear picture on the status of studied parameters which may be within the permissible limit of water quality standards or beyond that, therefore these types of maps can be successfully used in quality monitoring programmes.

Next Chapter XII includes all the research literatures cited in this thesis followed by list of publications (in Appendix -I) arising out of this thesis.