

Review

Implication of coliforms as a major public health problem in Nigeria

Akyala Ishaku A.^{1,2}, Olufemi Ajumobi¹ and Adebola Olayinka¹

¹Nigeria Field Laboratory Epidemiology Training Program, Abuja- Nigeria.

²Microbiology Unit, Department of Biological Sciences, Nasarawa State University, Keffi, Nigeria.

Accepted 26 November, 2013

Water, the essence of life, is threatened by bacterial contamination. Coliform count is the major tool to determine the bacteriological quality of water. The determination is quite easy and informative. The different methodologies are employed depending on suitability by maximum probable number (MPN) which is the most accepted. The environment conditions like sunlight, water salinity, temperature etc. provide simple concepts to justify the coliform counts at various places. Faecal coliforms are discussed here with special emphasis as these which are very significant indicators of faecal contamination. Though uncomplicated, coliform counts also determine framing policies for safe and healthy living. However, caution has to be taken while interpreting the coliform data. This paper aims to present the best for understanding the coliform data and interpreting them in a justifiable way.

Key words: Enterobacteriaceae, total coliforms, faecal coliform, *Escherichia coli*, *Enterococci*, *Streptococcus*, maximum probable number.

INTRODUCTION

Water has endless uses namely drinking, industrial, livestock, irrigation, aesthetics, boating, swimming, and fishing and so on. However, this elixir of life is being threatened by various pollutions but mainly the bacteriological pollution of water is a serious problem. Considering the bacteriological problems of water, what comes to our mind is the word 'coliform'. Since public and environmental health protection demands safe drinking water (free from pathogenic bacteria) therefore coliforms are major concern. Coliforms are single celled bacteria, classified as total and faecal coliform, where faecal coliforms are supposed to be more severe indicator of water pollution. Coliform bacteria form a part of the Enterobacteriaceae family (Kilb et al., 2003) which can also be naturally found in soil. However, faecal coliforms strictly live the gastrointestinal tract of warm-blooded animals and so originate from animal and human faecal discharges. *Escherichia coli* is a member of faecal

coliform group and *E. coli* is a specific indicator of faecal pollution (Rompre' et al., 2002). Detection of disease-causing bacteria and other pathogens in water is expensive and may pose potential health hazards. Further, testing for pathogens requires large volumes of water, and the pathogens may be difficult to grow in the laboratory and isolate. However, this problem can be easily solved by testing water for faecal coliforms especially *E. coli* as because they generally live longer than pathogens and are easy to culture in a laboratory than pathogens.

DEFINITIONS OF COLIFORMS

In standard method for the examination of water and wastewater (APPHA, 2005), coliform group members are described as:

1. All aerobic and facultative anaerobic, non-spore forming,

Gram-negative, rod-shaped bacteria that ferment lactose with gas and acid formation at 35°C within 48 h or;

2. All aerobic and numerous facultative anaerobic, Gram-negative, non-spore-forming, rod-shaped bacteria that grow as red colonies with a metallic sheen at 35°C within 24 h on an endo-type medium containing lactose.

The description of the coliform group has now included other characteristic, such as b-D-galactosidase-positive reactions (APPHA, 2005). The search for b-galactoside positive and b-galactoside-permease-positive organisms also permit a confirmation step for lactose fermentation, when the multi-tube fermentation method is used. The cytochrome-oxidase test is also used as a confirmation test to eliminate some bacteria of the *Aeromonas* or *Pseudomonas* genera that would ferment lactose.

The definition of coliform bacteria varies country wise slightly or on the organization in charge of the microbiological monitoring regulations. In Canada, the definition is the same as in the US, in some countries in Europe, the definition varies. For example, the French Standardization Association (1990) defines total coliforms (TC) as: "rod-shaped, non-spore-forming, Gram-negative, oxidase-negative, aerobic or facultative anaerobic bacteria that are able to grow in the presence of bile salts or other replacement surface active agents having an analogous growth inhibitory effect and that ferment lactose with gas and acid or aldehyde production within 48 h at 37 = 1°C. AFNOR (1990) defines other coliform groups, together with the thermo tolerant coliforms (also called faecal coliform, FC) and, more specifically, *E. coli* as thermo tolerant coliforms which have the same fermentation properties as total coliforms (TC) but at a temperature of 44 = 0.5°C. *E. coli* produces in-dole from tryptophan at a temperature of 44 = 0.5°C, gives a positive result methyl red test, is incapable to produce acetyl methyl cabinol and does not use citrate as its sole carbon source".

The faecal coliform group includes all of the rod-shaped non-spore forming bacteria, gram-negative, lactose-fermenting in 24 h at 44.5°C, and which can grow with or without oxygen. Another type of faecal bacteria is faecal streptococcus which is normally present in large numbers in the intestinal tracts of warm-blooded animals other than humans.

ENVIRONMENTAL SIGNIFICANCE

Total coliform is abundant in the soil. Coliform are found in natural environments, of earthy origin, but drinking water is not a natural environment for them. Their presence does not necessarily imply contamination from wastewater nor the presence of other sanitation based health risks but does indicate the need for an analysis of all water system facilities and their operations to decide the route of organisms entering the water system. Public notice to water system users is required since properly

constructed and maintained water should not have total coliform. Monitoring for organisms other than coliforms is also recommended by various estuarine waters (sometimes in legislation) for example, enterococci, faecal streptococci, salmonella, entero-viruses, etc. However, these recommendations and legal requirements usually apply only to bathing, recreational area or to shellfish zones.

The coliform include the following genus: *Escherichia*, *Klebsiella*, *Enterobacter*, *Citrobacter*, *Yersinia*, *Serratia*, *Hafinia*, *Pantoea*, *Kluyvera*, *Cedecea*, *Ewingella*, *Moelleralla*, *Lectercia*, *Rahnella*, *Yokenella* (Topley, 1997; Ballows, 1992). Coliforms such as *Citrobacter*, *Enterobacter* and *Klebsiella* species can also be found in natural environments such as soil, vegetation, or surface waters, where their presence is not necessarily related to faecal contamination (Leclerc et al., 2001). Faecal coliform is a subgroup of the total coliform group (American Public Health Association (APHA), 2005). Faecal coliform bacteria normally originate in the intestines of mammals, as discussed. They have a comparatively short life span compared to other coliform bacteria. Their occurrence could be related to improper disposal of sanitary waste. Immediate public notice and a boil order to the users (within 24 h) are required due to the higher likelihood of disease organisms also being present in water. Dominant in the area are *Escherichia* and *Enterococci* (Stevens et al., 2003).

E. coli is the main bacterium within the thermo tolerant coliform group, present in large numbers in feces at concentrations of about 10⁹ bacteria per gram of faecal matter (Brenner et al., 1982). It does not multiply appreciably in the environment (Edberg et al., 2000), whereas other members of these bacteria are found naturally in water, soil and vegetation (Parch and Malheur, 2012). Also, these are universally present in large numbers in sewage but do not grow in natural waters (Environment Agency, 2002). Town (2001) reported a strong positive correlation between faecal coliform and *E. coli* bacteria. When concentrations of faecal coliform bacteria are elevated, concentrations of *E. coli* bacteria are elevated too. Compared to other faecal coliform, they have a relatively short life span. Their presence indicates a strong probability that human or animal wastes are entering the water system.

E. coli is considered to be most sensitive to environmental stresses. Its survival time in the environment is dependent on many factors, such as temperature, exposure to sunlight (UV rays), presence and types of other micro flora, and the physico-chemical characteristics of water involved (for example, groundwater, surface water, or treated distribution water). In general terms, *E. coli* survives for about 4 to 12 weeks in water containing a moderate amount of micro flora at a temperature of 15 to 18°C (Edberg et al., 2000). Regrowth of *E. coli* in water distribution systems is not a concern, since *E. coli* rarely grows outside the human or animal gut (Geld, 1996).

So far, the Guidelines for Canadian Recreational Water Quality (Health and Welfare, Canada, 1996) have suggested *E. coli* as the best indicator of faecal contamination from warm-blooded animals in freshwaters whereas the enterococci group is for marine waters (Nail, 2004). Generally, for water examination purposes, enterococci can be regarded as indicators of faecal pollution, although some can rarely originate from other environment.

Enterococci have a number of advantages as indicators over total coliforms and even *E. coli*, as they have been known to survive longer (Meters et al., 1974). Despite being less numerous than faecal coliforms and *E. coli* in human feces (Fleche et al., 1983), they are still abundant enough to be detected after significant dilution. There is a concern that enterococci are a diverse group of bacteria, and that the group contains species that are environmental and their presence in water is not necessarily indicative of faecal pollution. This concern is driven by the problems associated with the use of total coliforms as an indicator of faecal pollution. An early research report by Geld (1970) indicated that *Enterococcus faecalis* vary liquefactions was common in good quality water and its importance was not clearly considered if recovered in waters in concentrations of less than 100 organisms/100 mile however, more recent research on the relevance of faecal streptococci as indicators of pollution showed that the majority of enterococci (84%) isolated from a variety of polluted water sources were “true faecal species” (Pinto et al., 1999).

SETTING WATER QUALITY GOALS

As per Central Pollution Control Board (CPCB), an apex body in the field of water quality management, India, the term quality must be considered relative to the anticipated use of water. From the user's point of view, the term “water quality” is defined as “those physical, chemical or biological characteristics of water by which the user evaluates the acceptability of water” (CPCB, 2008). The water supply must be pure, wholesome, and potable. Therefore, for setting water quality objectives of a water body, it is essential to identify the uses of water in that water body. CPCB has developed a concept of “designated best use”. According to which, out of several uses a particular water body is put to, the use which demands highest quality of water is called its “designated best use”, and consequently the water body is designated. For each of these five “designated best uses”, the CPCB has identified water quality requirements in terms of few chemical characteristics, known as primary water quality criteria. The “designated best uses” along with respective water quality criteria is given in Table 1. For aquaculture and cooling, the coliforms are not considered as there is no direct damage found till now. The CPCB, in collaboration with the concerned state Pollution Control Boards,

has classified all the water bodies including coastal water in country according to their “designated best use”.

RISK TO HUMAN HEALTH

Most people are concerned about the health risk that coliform may pose. People exposed to coliform contaminated water may exhibit fever, diarrhea and abdominal cramps, chest pain, or hepatitis. During bathing, exposure to coliforms may cause urinary tract infection. While *E. coli* by itself is not generally dangerous, other pathogens of faecal origin that are health threats include *Salmonella*, *shield*, and *Pseudomonas aeruginosa*. Non-bacterial pathogens that may be present with faecal material include protozoans, such as *Cryptosporidium*, *Giardia* and viruses. Vero cytotoxic *E. coli* (Parch and Malheur, 2012). The vero-cytotoxin/shiga toxin producing *E. coli* (VTEC/STEC) group has over 200 different serotypes, including the highly pathogenic enterohaemorrhagic *E. coli* (EHEC) with *E. coli* O157:H7 the most significant serotype that causes hemorrhagic colitis with bloody diarrhea and haemolytic uraemic syndrome better known as HUS (Bolton et al., 2009; WHO, 2004). There are also other pathogens, such as: Enterotoxigenic *E. coli* (ETEC), enteropathogenic *E. coli* (EPEC), enteroinvasive *E. coli* (EIEC), enteroaggregative *E. coli* (EAEC), and diffusely adherent *E. coli* (DAEC), whose spread occurs mostly through the human faecal-oral route (Bolton et al., 2009). Several authors have reported waterborne disease outbreaks in water meeting the coliform regulations (Gofti et al., 1999).

LABORATORY METHODS FOR TOTAL COLIFORM DETECTION

All method of total coliform identification requires culturing of the sample in the presence of a special media. The culturing process requires approximately one to three days for the coliform to grow before interpreting the bacterial data. There are mainly three laboratory procedures that are majorly used to detect coliform in a water sample. However, there are many other sophisticated methods which have come up in the recent years.

Multiple tubes

This method was developed in the early 1900s. It uses some test tubes and measures the amount of gas production in another small tube called Durham's tube during 48 h of incubation. Results are reported in terms of most probable number of organisms (MPN) per 100 milliliters of sample. Lactose and lauryl tryptose broths are used as presumptive media, but Seidler et al. (1981) and Evans et al. (1981) have observed interference of

Table 1. Use based classification of surface water in Nigeria (NAFDAC, 2008).

Designated-best-use	Class of water	Criteria
Drinking Water Source without conventional treatment but after disinfection	A	-Total coliforms organism MPN/100 ml shall be 50 or less -pH between 6.5 and 8.5 -Dissolved oxygen 6 mg/1 or more -Biochemical oxygen demand 5 days 20°C 2 mg/1 or less
Outdoor bathing (organised)	B	-Total coliforms organism MPN/100 ml shall be 500 or less -pH between 6.5 and 8.5 -Dissolved oxygen 5 mg/1 or more -Biochemical oxygen demand 5 days 20°C 3 mg/1 or less
Drinking water source after conventional treatment and disinfection	C	-Total coliforms organism MPN/100 ml shall be 50 or less -pH between 6 to 9 -Dissolved Oxygen 4mg/1 or more -Biochemical oxygen demand 5 days 20°C 3mg/1 or less
Propagation of wild life and Fisheries	D	-pH between 6.5 and 8.5 -Dissolved oxygen 4mg/1 or more -Free ammonia (as N) 1.2 mg/1 or less
Irrigation, industrial cooling, controlled waste disposal	E	-pH between 6.0 and 8.5 -Electrical Conductivity at 25°C micro mhos/cm Max.2250 -Sodium absorption ratio Max. 26 -BoronMax. 2 mg/1

non-coliform bacteria, using lactose broth. A1 broth is used to detect faecal coliforms. The tubes with a positive presumptive reaction are then subjected to a confirmatory test. This number is a statistical estimate of the mean number of coliforms in the sample. As a result, this technique is a semi-quantitative enumeration of coliforms. This is reliable, easy to implement and requires only basic microbiological training apart from being relatively economical. This method suffers from lower precision in the estimation and depends on the number of tubes used for the analysis. The method is very tiresome, time-consuming and labor intensive since many dilutions have to be processed for each water sample. Significant numbers of glassware are used and laboratory cleanup is required.

Membrane filter (MF) method

This method came up in early 1950s. It filters organisms from the water through a sterile filter with a 0.45 mm pore size which retains bacteria and then incubates the initial parent organisms on the filter paper to produce visible colonies. A minimum of 22 h incubation time is required. Results are recognized as "count" of colony forming units (CFUs) per 100 milliliters. Many media and incubation conditions for the MF method have been tested for

optimal recovery of coliforms from water samples (Rice et al., 1987). Among these, the most extensively used method for drinking water analysis are the m-Endo-type media in North American (American Public Health Association (APHA), 2005) and the Tergitol-TTC medium in Europe (Association Francaise de Normalisation (AFNOR), 1990). Coliform bacteria form red colonies with a metallic gloss on an endo-type medium (incubation 24 h at 35°C for TC) or yellow-orange colonies on Tergitol-TTC media (incubation 24 and 48 h at 37 and 44°C for TC and FC, respectively). Other media, like MacConkey agar and the Teepol, have been used in South Africa and Britain. However, comparisons have shown that m-Endo agar yields higher counts than MacConkey or Teepol agar (Grabow and du Preez, 1979). The chromo cult agar has been found to be an alternative to MacConkey agars.

To enumerate FC, the APHA (2005) proposed that filters be incubated on an enriched lactose medium (m-FC) at a temperature of 44.5°C for 24 h. Due to the elevated incubation temperature and the addition of rosolic acid salt reagent, few non-faecal coliform colonies may develop on the m-FC medium (APHA, 2005). Dark blue colonies confirm faecal coliform's presence. Additionally, typical colonies with shining may be produced occasionally by non-coliform bacteria and dark red or nucleated colonies without sheen may occasionally be coliforms. Coliform verification is therefore recommended

recommended for both types of colonies (APHA, 2005). Some improvements in the method have increased detection of injured coliform bacteria, including the development of m-T7 medium formulated specifically for the recovery of stressed coliforms in drinking water (LeChevalier et al., 1983).

Evaluation on routine drinking (Meters et al., 1986) and surface (Freier and Hartman, 1987) water samples showed higher coliform recovery on the m-T7 medium as compared with that on the m-endo medium. However, m-T7 may not be as efficient when stressing agents other than chlorine are involved. Rice et al. (1987) achieved no significant difference in coliform recovery on m-T7 compared with m-Endo LES from mono-chlorinated samples. Adams et al. (1989) found that the m-T7 medium performed no better than the medium in enumerating *E. coli* and *C. freundii* cells exposed to ozone. This method is much simpler than MPN, less labor intensive and requires less clean up of glassware. However, it cannot be used on muddy water. The presence of high numbers of background heterotrophic bacteria has been reported to decrease coliform recovery by MF (Clark, 1980; Burlingame et al., 1984).

Too much crowding of colonies on m-Endo media has been linked with a reduction in coliform colonies producing the metallic shine (Hsu and Williams, 1982). The principal concern about MF is its incapability to recover stressed or injured coliforms. A number of chemical and physical factors involved in drinking water treatment, like disinfection, can cause sub lethal injury to coliform bacteria, resulting in injured cells which fail to develop colonies on a selective medium. Exposure of bacteria to chlorines like products may also result in enhanced sensitivity bile salts or replacement of surface-active agents (sodium deoxycholate or Tergitol 7) contained in some selective media (Rompre et al., 2001).

MMO chromogenic fluorogenic method

This method was developed in the late 1980s. It comprises of culturing the coliforms in the sample bottle. An incubation time of 18 to 28 h is required. The yellow color indicates the presence of total coliform and florescent condition under black light indicates *E. coli*. Results are stated as the presence/absence of coliform organisms per 100 milliliters. Non-coliform organisms are not produced, this being an advantage. The enzyme substrate, for example o-nitrophenyl-b-D-galactopyranoside (ONPG), β -Galactosidase assay (CPRG), and 4-methylumbelliferyl-b-D-glucuronide (MUG) are organism specific and where they are not, the target organism is selected for by suppressing the competing. The target population is characterized by enzyme systems that metabolize the substrate to release the chromogen/fluorogen. This results in a colour change in the medium and/or fluorescence detected under long wave UV

radiation. The most important food pathogens can be screened using chromogenic/fluorogenic media in a wide variety of food samples like *Salmonella*, *Camphylobacter*, *Listeria*, *Listeria monocytogenes*, *S. aureus*, Coliforms, *E. coli* as well as specific target organisms such as *E. coli* O157.

ANOMALY IN COLIFORM DATA

Sometimes the estimation of coliform does not lead to proper understanding of the situation. This may be due to the following reasons. When the non-coliforms are present in high numbers, (more than 200 colony forming units (CFUs) in a 100 milliliter sample), it inhibits the growth of coliforms. Coliform counts for total and faecal can vary greatly throughout the stretch of an estuary-mainly due to the dilution of freshwater with seawater which continuously changes as a result of tidal fluctuations. In an inadequately filtered well, bacteria are expected to be present. Organisms that enter a well can be there one day and die off before a second sample is taken a few days or a week later. Therefore, one may fall sick but the cause may not be detected. Variation in methods of analysis can lead to variable counts. Some bacterial tests use a filtration step while others do not. Each test uses a different media to incubate the organisms. Sometimes the bacteria themselves are counted while in other cases enzyme byproducts are measured. Some methods better detect stressed coliform species while others do not. Fully representative samples are hard to obtain since bacteria often combine together in clumps in pipes and in the sample container. Thus, in cases where there are few organisms, they may not be evenly distributed in the water. Due to high salinity the coliform count may be much below the permissible limits. However, this condition does not allow the water quality to be drinkable

COLIFORMS' ENTRY TO WATER SYSTEM

1. Open defecation in the catchments area release the human waste to the water body which then meets the water through surface runoff. Animal feces also contribute in the similar way. Dellile (1987) found a strong positive correlation between penguin population and bacterial numbers in the sea water adjacent to the rockeries and also a decline in bacteria numbers with distance from store. This finding supports the correlation between cattle feces and coliforms. Thus, runoff from cattle feedlots, hog farms, dairies, and barnyards that have poor animal keeping practices where waste is not properly disposed contribute a lot.
2. Domestic sewage can be the dominant source of faecal microorganisms in the marine environment and have a significant environmental impact (Lenihan et al.,

1990).

3. Discharges from illegal or leaky sanitary sewer connections, poorly functioning septic systems, wastewater treatment plant effluent are potent contributors. Bacteria are much more abundant in soils than in water.

4. Storms flows containing high amounts of sediment are often related to high concentrations of pathogenic bacteria (Marino and Gannon, 1991). The bacteria can attach to sediment particles to escape invertebrate predators (Murdoch and Cheo, 1996). Fast-running water can carry more sediment, so higher levels of bacteria can occur during high runoff. During storm flow, a strong positive correlation has been established between faecal coliform and *E. coli* bacteria (Town, 2001).

5. Bacteria washed into the ground by rainfall or snow-melt are usually filtered out as water seeps through the soil, so properly constructed water wells do not typically harbor coliform bacteria. However, fractured bedrock aquifers close to the surface are the exceptions, nevertheless, coliform bacteria can persist within slime formed by naturally occurring ground water microorganisms.

6. The slime (or biofilm) clings to the well screen, casing, drop pipe, and pump. Bacteria can enter into a new well during construction and can remain if the water system is not thoroughly disinfected and flushed. Well construction defects such as insufficient well casing depth, improper sealing of the space between the well casing and the borehole, corroded or cracked well casing, and poor well seals or caps can allow sewage, surface water, or insects to carry coliform bacteria into the well. Unplugged abandoned wells can also carry coliform bacteria into deeper aquifers. Opening at the top of the well; rusty or damage well casing; unprotected suction line; buried wellhead; and, nearness of a well to septic tanks, drain fields, sewers, kitchen sinks, drains, animal feedlots, abandoned wells, and surface water enhance the problem. Cross-connections with wastewater plumbing can also introduce coliform bacteria into the water supply. Sometimes water sources are contaminated by coliforms existing on biofilms predominantly *Citrobacter species* (kilb et al., 2003) harbored on rubber-coated valves in the water treatment units.

7. The increase in the number of industrial farms, without soil nearby, represents an opportunity to reuse their residues for agricultural purposes, as a source of nutrients and organic matter (Rufete et al., 2006) which often contributes faecal coliforms to soil and then ultimately to water.

FAVOURABLE FACTORS FOR GROWTH

1. Water depth can influence the effectiveness of solar radiation in faecal coliform inactivation (Sinton et al., 1994). Action spectra for *E. coli* show that UVB radiation has the greatest bactericidal effect (Webb and Brown, 1976), but UVA may be more vital in the marine background, as it penetrates the water column to a greater

depth (Davis-Colley et al., 1994).

2. The radiation further produces heat which again has a significant effect on coliforms. Bacteria grow faster at higher temperatures. The growth rate slows drastically at very low temperatures (Smith et al., 1994).

3. Research suggests that particles as small as 11 nm naturally occurring in surface water are able to harbor indigenous coliform bacteria and *E. coli*, subsequently offering protection from UV light at a wavelength of 254 nm and up to a dose of 40 mJ/cm² (Cantwell and Hofmann, 2008). This phenomenon has been observed in water with turbidities as low as 0.8 NTU.

4. High concentration of dissolved oxygen boost microbial inactivation as seen in the Antarctic (Hughes, 2003). Further, temperature and salinity play important roles in regulating the concentration of oxygen found in seawater, when oxygen is present, photochemical damage to *E. coli* enhances, particularly in the presence of UVA (Sinton et al., 1994). The combination of UV and oxygen allows the formation of highly reactive free radicals (including singlet oxygen, hydroperoxyl, and hydroxyl groups), which cause cellular damage to the coliforms (Vincent and Neale, 2000). A weak negative correlation was found between dissolved oxygen and concentration of faecal coliform bacteria and *E. coli* (Hughes, 2003).

5. Stream flow often causes dilution of sewage and other wastes. It also dilutes freshwater, further reducing the coliform count (Hughes, 2003).

6. Algal blooms act as shields and reduce the penetration of solar radiation into the water column (Hader et al., 1998).

7. Sea ice thickness and physical properties, together with the snow that collects on its surface, can result in the reduction of solar radiation input into the water column (Belzile et al., 2000).

8. Salinity can affect faecal bacterial viability with high or rapidly changing salt concentrations increasing the cell inactivation (Anderson et al., 1979). The input of freshwater from iceberg melt, snowmelt from the shore, and sewage waste contributed to the low salinity in colder areas (Hughes, 2003). Seasonal factors can affect seawater salinity such as glacial melt and can reduce salinity. In summer, salinity around a piece of melting glacier ice can vary between almost freshwater and > 30‰ salinity (Hudier and Ingram, 1994), while in winter, salt released during sea ice formation can increase sea water salinity (Golden, 2001). Coliform mortality may be greater than before by quick and sudden changes in osmotic stress caused by passing through seawater with spatially variable salinity.

RECOMMENDATIONS

If coliform bacteria are present, the source of the problem should be identified. Re-sampling from several locations within the water system is helpful. The entire water system

may need to be thoroughly flushed and disinfected before a negative bacteria sample can be withdrawn. Sometimes it is necessary to repeat the disinfection process. Proper changes or repairs should be made in the well. After the defects are corrected, the whole water system should be disinfected and the water re-examined before drinking. Many removal and disinfection procedures have been developed to control coliforms. Fluidized sand bio filters have been effectively used to remove total coliform bacteria (Davidson et al., 2008). An overall reduction of total and faecal coliforms in activated sludge system has also been found to be significant (Kazmi et al., 2008). Further, an interrelationship of biological oxygen demand (BOD) and suspended solids (SS) has been found with coliforms which suggest that improvement of the microbiological quality of wastewater could be linked with the removal of SS. Therefore, SS can serve as a regulatory tool in lieu of a clear coliforms standard.

Photo catalysis (TiO₂) has recently emerged as an alternative technology for bacteria inactivation (McLoughlin et al., 2004). Some simple approaches may be boiling the water. Chlorine (as gas or hypochlorites), chlorine dioxide, ozone and UV radiation are common tools for disinfection of drinking water (Rizzo, 2009). A very important remedy is to use bacteriophage to remove the coliforms. This is the most natural way. Ultimately, personal hygiene has no alternative. Washing thoroughly with soap after contact with contamination can prove to be effectively safe. The information on coliforms helps the water quality managers and planners to set water quality targets and identify needs and priority for water quality restoration programs for various water bodies in the country. The famous Ganga Action Plan and subsequently the National River Action Plan are results of such exercise (Central Pollution Control Board (CPCB), 2008).

REFERENCES

- Adams JC, Lytle MS, Dickman DG, Foster DH, Connell JP, Bressler WR (1989). comparison of method for enumeration of selected coliforms exposed to ozone. *Appl. Environ. Microbiol.* 55:33-35.
- Association Francaise de Normalisation (1990). *Eauxme 'thodes d'essais, Recueil de Norms Francaises*, 4th ed. La De' fense, Paris.
- Anderson IC, Rodes MW, Kator HI (1979). Sublethal stress in *Escherichia coli*: a function of salinity. *Appl. Environ. Microbiol.* 38:1147-1152.
- APHA (American Public Health Association), AWWA (American Water Works Association, WEF (2005). *Standard Methods for the Examination of Water and Wastewater*. 21th edn. Washington, DC.
- Ballows A (1992). *The Prokaryotes*, 2nd ed. Springer Verlag, New York.
- Belzile C, Johnnessen SC, Gosselin M, Demers S, Miller WL (2000). Ultraviolet attenuation by dissolved and particulate constituents of first-year ice during late spring in an *Arctic polynya*. *Limnol. Oceanogr.* 45:1265-1273.
- Bolton DJ, Duffy G, O'Neil CJ, Baylis CL, Tozzoli R, Moraboto S, Wasteson Y, Lofdahl S (2009). Epidemiology and Transmission of Pathogenic *Escherichia Coli*. Co-ordination Action FOOD-CT-2006-036256, Ashtown Food Research Centre, Teagasc, Dublin, Ireland.
- Brenner DJ, McWhorter AC, Knutson JK, Steigerwalt AG (1982). *Escherichia vulneris*: a new species of Enterobacteriaceae associated with human wounds. *J. Clin. Microbiol.* 15:1133-1140.
- Burlinggame GA, McElhancy J, Bennett M, Pipes WO (1984). Bacteria interference with coliform colony sheen production on membrane filters. *Appl. Environ. Microbiol.* 47:56-60.
- Cantwell RE, Hofmann R (2008). Inactivation of indigenous coliform bacteria in unfiltered surface water by ultraviolet light. *Water Res.* 42:2729-2735.
- Central Pollution Control Board (2008). *Guidelines for Water Quality Management*. Parivesh Bhawan, East Arjun Nagar, Delhi.
- Clark JA (1980). The influence of increasing numbers of non-indicator organisms by the membrane filter and presence-absence test. *Can. J. microbial.* 26:827.
- Davidson J, Helwig N, Summerfelt ST (2008). Fluidized sand biofilters used to remove ammonia, biochemical oxygen demand, total coliform bacteria, and suspended solids from an intensive aquaculture effluent. *Aquacult. Eng.* 39:6-15.
- Davies-Colley RJ, Bell RG, Donnison AM (1994). Sunlight inactivation of Enterococci and faecal coliforms in sewage effluent diluted in seawater. *Appl. Environ. Microbiol.* 60:2049-2058.
- Dellile D (1987). Spatial distribution of coastal Antarctic seawater bacteria: relationship with Avifauna. *Polar Biol.* 8:55-60.
- Ederberg SC, Rice EW, Karlin RJ, Allen MJ (2000). *Escherichia coli*: the best biological drinking water indicator for public health protection. *J. Appl. Microbiol.* 88:106S-116S.
- Environment Agency (2002). *The Microbiology of Drinking Water, Part 1-Water Quality and Public Health, methods for the Examination of Water and Associated Materials*, Bristol.
- Evens TM, Waarvick CE, Seidler RJ, LeChevallier MW (1981). Failure of the most probable number technique to detect coliforms in drinking water and raw water supplies. *Appl. Environ. Microbiol.* 41:130-138.
- Freier TA, Hartman PA (1987). Improved membrane filtration media for enumeration of total coliforms and *Escherichia coli* from sewage and surface waters. *Appl. Environ. Microbiol.* 53:1246-1250.
- Geld REE (1996). *Microbial quality of water supply in distribution systems. Biological profits in drinking water* CRC Press, Lewis Publishers. pp. 293-367.
- Geld REE (1970). Applying bacteriological parameters to recreational water quality. *J. Amer. Water Works Assoc.* 62:113-120.
- Gofti L, Zmirou D, Murandi FS, Hartemann P, Peleton JL (1999). Waterborne microbiological risk assessment: a state of the art and perspectives. *Rev. epidemiol. Sante' Publis* 47:61-75.
- Golden KM (2001). Brine percolation and the transport properties of sea ice'. *Ann. Glacial.* 33:28-36.
- Grabow WOK, DU Preez M (1979). Comparison of m-Endo LES, MacConkey, and teepol media for membrane filtration counting of total coliform bacteria in water. *Appl. Environ. Microbiol.* 38:351-358.
- Hader DO, Kumar HD, Smith RC, Worrest RC (1998). Effects on aquatic ecosystems. *J. Photochem. Photobiol. B.* 46:53-68.
- Health and Welfare, Canada (1992). *Guidelines for Canadian Recreational Water Quality*, Government Publishing Centre, Ottawa.
- Hsu SC, Williams TJ (1982). Evaluation of factors affecting the membrane filter technique for testing drinking water. *Appl. Environ. Microbiol.* 44:453-460.
- Hudier E, Ingram G (1994). Small-scale melt processes governing the flushing of nutrients from a first-year sea ice, Hudson Bay, Canada. *Oceanol. Acta.* 17:397-403.