ISSN : 0378 4568

anveṣak

A bi-annual journal

VOL. 51, NO.1 (IX) January - June 2021

Sardar Patel Institute of Economic and Social Research
Thaltej Road, Ahmedabad - 380 054. India

SARDAR PATEL INSTITUTE OF
ECONOMIC AND SOCIAL RESEARCH

anveṣak

A bi-annual journal

CERTIFICATE OF PUBLICATION

This is to certify that the paper entitled

STUDY ON OCCURRENCE AND DISTRIBUTION OF FECAL INDICATOR BACTERIA IN COASTAL REGIONS OF PALGHAR , NORTH WEST COAST OF MAHARASHTRA

Authored by

Shailaja.P.Palan
Research Scholar, Shri Jagdishprasad Jhabarmal Tibrewala University, Jhunjhunu, Rajasthan, India

vol. **51** No. **01(IX)**

in

Anvesak A bi–annual Journal

UGC Care Group - 1

ISSN : 0378 - 4568

January – June 2021

Editor
Anvesak
A bi–lingual Journal

STUDY ON OCCURRENCE AND DISTRIBUTION OF FECAL INDICATOR BACTERIA IN COASTAL REGIONS OF PALGHAR , NORTH WEST COAST OF MAHARASHTRA

Shailaja.P.Palan Research Scholar, Shri Jagdishprasad Jhabarmal Tibrewala University, Jhunjhunu, Rajasthan, India
Arun Kumar Assistant Professor, Department of Microbiology, Shri Jagdishprasad Jhabarmal Tibrewala University, Jhunjhunu, Rajasthan, India
Shilpa.M. Gharat Assistant professor, Department of Biotechnology, Sonopant Dandekar college,Palghar, Maharashtra, India

Abstract
The population density, occurrence and distribution of bacterial indicators of fecal contamination were studied in the coastal waters and intertidal sediments at three sites during three seasons in Palghar taluka, North west coast of Maharashtra. In the study fecal indicator bacteria viz total coliform, fecal indicator bacteria and total enterococcal count was found to be highest in the creek than at the inshore sites and furthermore in sediments than in the overlying water column. Monsoon was found to contribute towards elevated levels of faecal indicator bacteria in water column whereas was found to be least during pre-monsoon. Annual abundance of FIB and in exceedance to standard permissible limits indicates the fecal contamination at Dandi creek and Navapur coast as resultant effects of anthropogenic activities along the coastal region.

Key words: Faecal indicator bacteria, coastal water, intertidal sediments, Palghar taluka.

Introduction:
As one of the largest aquatic ecosystem, marine ecosystem provides 90% of habitable space on earth. It encompasses varied near shore and off shore systems which further characterize the biological community present in that environment. Acting as epicenter for many man made activities like industrialization, aquaculture and recreation has exposed coastal environments to various sources of contamination. One of the prime causative agent of coastal pollution reaching coastal waters are fecal contamination through rivers, open sewers, open defecation along shorelines (Richa S. et al., 2018) and from fore shore sands (Whitman and Nevers, 2003). Fecal contamination leads to release of organisms not indigenous to aquatic system resulting in prevalence of both bacteria and viruses that are pathogenic to humans. Sewage is identified as major source of fecal contamination in aquatic systems and results in deleterious effect on water quality. It is also the major source of pathogens responsible for many waterborne epidemics and associated mortality around the world (WHO, 2011). Fecal contamination from both human and animal origin are exposed to coastal regions but it is well documented that human health risk from human fecal contamination is more in comparison to animal feces (Tyagi et al., 2006). Epidemiological studies underline the increasing evidence of gastro-intestinal illness, sinus infection, ear infection and wound infection in humans when in contact with faecally contaminated waters.
In an ecosystem, the underlying disturbances can well be represented by indicator organisms. Indicators that are sensitive, reliable, responsive and that can be scientifically proven are of prime importance. Monitoring microbial indicator organisms or microbiological quality of the water depicts the health status of the site and extent of pollution (Kumarasamy et al., 2009). Release of sewage both directly and indirectly can affect the microbial population in which it is released hence it is crucial to have indicator system to quickly reflect the quality status of underlying water system. Microbiologically indicator organisms of fecal origin are used to ascertain the quality of water. For assessment of water quality, monitoring group of indicator organisms than single indicator organism can be used as a better predictive tool for presence of certain pathogens which are of human health concern (Tyagi et al., 2006). As per WHO (2003), faecal indicator bacteria are the indicators of faecal contamination. Faecal indicator bacteria are group of bacteria that inhabit GI tract of warm

blooded animals. One of the reasons for monitoring faecal indicator bacteria in waters instead of pathogens is the ease with which it can be detected and its presence as substitute for pathogenic organisms that are responsible for microbial pollution (Abdelzaher et al., 2010). Further since levels of pathogens are either, too low in concentration in environmental samples; require technical skill and hence requires expensive detection methods (Korajkic et al., 2018) and also are lesser adapted to aquatic environmental stressors (Hassard et al., 2016). Most commonly used fecal indicator bacteria (FIB) are Coliform group of bacteria and its subsets that includes total coliform, faecal coliform, *Escherichia coli* and fecal streptococci. These pollution indicator bacteria are studied as proxy for occurrence of pathogenic bacteria (Halliday and Gast, 2011).

Conventionally total coliform count is used as indicator of possible fecal contamination from sewage and thus, is of sanitary significance. Subsets of total coliform are considered to be more definitive of homoeothermic fecal contamination and referred to as fecal coliform or also known as thermotolerant coliform which is found to correlate well with fecal contamination with warm blooded animals. But since total and fecal coliform, are present in non-fecal sources and have lesser survival rate in water body (Tyagi et al., 2006) hence the occurrence of fecal coliform in relation to fecal contamination must be referred with appropriate correlation. Alternative to coliform another group of bacteria used as fecal indicator bacteria, a subset of fecal streptococci is 'Enterococci'. Enterococci spp is abundant in human feces is found in the range of 10^4- 10^6 /gm of fecal matter (Boehm and Sassoubre, 2014). It is more widely accepted indicators over total coliforms as requires easy detection methods, persists longer and in detectable concentration even on dilution (Tyagi et al., 2006) and correlates better with pathogens found in sewage(Jin et al., 2018). As per USEPA standards in recreational marine waters permissible levels of enterococci was not more than 104 CFU/100ml in single sample and geometric mean of 35 MPN/ 100ml and for Fecal coliform is less than 200 CFU/100ml. In India, as per Centre for pollution control board (CPCB) standards for designated best use, seawater is classified fit for commercial fishing, contact recreation and bathing activities if fecal coliforms levels are less than 100CFU/100ml.

Several studies in coastal regions like by Vignesh et al., (2014) in beaches of Tamil nadu, Banoo et al., (2014) at Odisha coast, Bharathi et al., (2018) along Ennore coast, Rajendran et al (2018) at Tuticorin coast , by Kutty and Sebastian, (2013), in Varkala beach in Kerala, in coastal waters of southern Kerala by Robin et al., (2012), in North west coast of India by Fulke et al., (2019) found fecal indicator bacteria to be more prevalent along anthropogenically influenced near shoreline regions of both east and west coast of India. No such study has been undertaken to assess the microbial pollution level in coastal waters of Palghar taluka, northwest coast of Maharashtra which has undergone rapid urbanization and industrialization in last decade. Present study will thus help in generating a baseline data on annual occurrence, abundance and distribution of fecal indicator bacteria in coastal environments of Palghar region.

Materials and method:

Study site:

Palghar taluka is a coastal town located in newly formed Palghar district as on 1st August 2014.This town is located in northwest part of Maharashtra with Arabian Sea forming its western boundary. Further the coastal region enclosing Palghar taluka harbors many small ports, recreational beaches, and industrial zones. The microbiological characteristics of the coastal zones at 3 different locations along the Palghar coast were undertaken in the study. Site I Kelwa beach located at 19°36'39.6"N 72°43'46.8"E is a stretch of 8 km, an increasingly popular recreational beach in Palghar taluka visited by tourist and locals during weekends. Site II was Navapur coast located at 19°47'14.1"N 72°40'54.2"E and harbours a submarine outfall which is used for disposal of 25MLD of treated combined domestic and industrial effluents generated from human settlements along the coast and from the nearby town of Boisar. The coast is also exposed to open defecation which is still a practice among the localities residing along the coast. Boisar is home to largest Maharashtra industrial development corporation which is home to many small, medium and large scale textile, Pharmaceutical, bulk drug production units. But from past decade the manifold increase in the effluent generated due to rapid industrialization and urbanization in and around vicinity of Boisar is estimated to be around ~3-4times (NIO,2018).Combined increase in domestic and industrial effluent increases the risk of deterioration experienced by the coast. Site III was Dandi creek located at 19°47'59.8"N 72°41'19.0"E which extends from Dandi village and flows in westward direction towards Arabian sea and finally opens into the Navapur coast. It is a 10 km long creek with very narrow mouth region and towards the upper stretch receives freshwater from tributaries of Surya river. It is a minor creek with poor flushing and receives large volume of partially treated effluent and untreated domestic sewage from nearby settlements find their way into this creek. The creek is also used by localities for fishing.

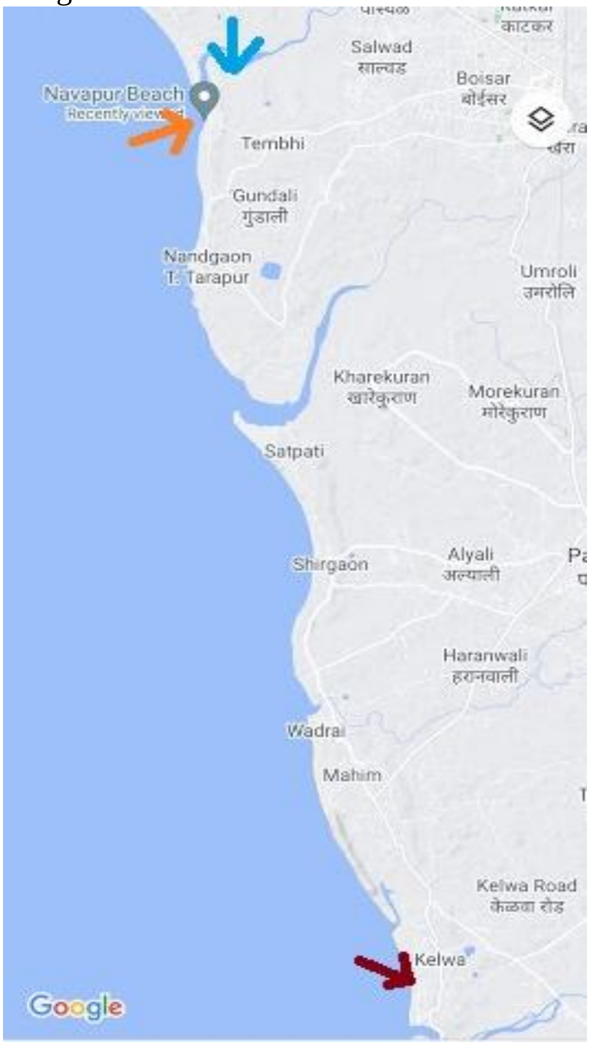

Fig:1: Three Sampling sites Dandi creek Navapur coast Kelwa coast

Sampling method:

In the present study, sampling was carried out from November 2019 to October 2020 covering three different season as post-monsoon from November to February, pre-monsoon from March to June and Monsoon from July to October. Sampling sites and frequency of sampling was found to be in accordance with guidelines of water quality monitoring of surface water by CPCB, 2008

Monthly one litre of surface water at the depth of 0-15cm and 250gm of intertidal sediments were collected in sterile polyethylene bottles from three regions at each site separated ~200 meters by grab sampling. Samples were collected and stored in ice box at 4 °C and transported to the laboratory within 6 hrs for analysis. Total of 36 water samples and 36 intertidal sediments were collected at each site for microbiological analysis.

Enumeration of Fecal indicator bacteria:

Total coliform count, faecal coliform and total enterococcal count was determined from both surface water and intertidal sediments by Membrane filtration technique as per the standard methods for examination of water and waste water by APHA, 2012. Depending on turbidity of sample either direct water sample or diluted water samples were filtered for analysis. For sediment sample, 10gm of sediment sample was suspended in 100 ml of sterile saline and mixed vigorously for 30 seconds and allowed to stand for 5 minutes. The supernatant was used for further analysis. The samples were appropriately diluted and 10 ml of sample were filtered in duplicates. Membrane filter was aseptically transferred on appropriate selective media at selective temperature for growth of the bacteria. Red or dark red mucoid colonies with golden metallic sheen on m-Endo agar and further confirmed for gas production in st. lauryl tryptose broth at 37°C for 24 hrs was counted as confirmed coliform. Blue coloured colony on st. m- FC agar with production of gas in lauryl tryptose broth is counted on incubation at 44.5° C for 24 hrs was enumerated as confirmed faecal coliform count. Red and dark red colonies on m –enterococcus agar on incubation at 37°C for 24 hrs, confirmed further by showing growth in St Brain heart infusion broth incubated at 44.5° C and in St BHI broth containing NaCl at 37°C were enumerated as Total enterococcal count. The counts obtained were interpreted as CFU/ml in water samples and CFU/gm in sediment samples.

Results and Discussion:

As individual indicator organisms may not completely represent the source hence instead of detecting individual indicator organisms monitoring group of indicator organisms can be used as predictive tool for presence of pathogens (Tyagi et al., 2006). Fecal indicator bacteria include total coliform (TC), fecal coliform (FC), and total Enterococcal count (TEC). In the present study, occurrence, abundance and distribution of TC, FC and TEC were determined in both surface water and intertidal sediments. Monthly variation in total coliform, fecal coliform, and fecal streptococci count was determined at each site along coastal waters and intertidal sediments annually from the period of November 2019 to October 2020.The mean average count obtained in three different seasons is summarized in (Table.1)

Microbiological count in Surface water:

The total coliform count enumerated at 3 sites under the study annually from the period of November 2019-October 2020 ranged from $0.063x10^2$–$0.08x10^2$ CFU/ml, $27.6x10^2$ – $249x10^2$ CFU/ml and 72.1 $x10^2$– 385 $x10^2$ CFU/ml at Kelwa coast, Navapur coast and Dandi creek respectively.

At Kelwa coast no FC count was recorded in surface water throughout the study period. At Navapur coast and Dandi creek the fecal coliform count ranged from 7.06×10^2 - 63.4×10^2 CFU/ml and 17.8×10^2 - 184 $\times10^2$ CFU/ml respectively. A spike in FC count was noted during mid pre-monsoon in May 2020. Total Enterococcal count was not detected in surface water throughout the study period at Kelwa coast whereas it ranged from 4.08 $\times10^2$- 42.5×10^2 CFU/ml at Navapur coast and between 8.9×10^2 - 48.4×10^2 CFU/ml at Dandi creek.

Seasonally the abundance of all fecal indicator bacteria i.e Total coliform, Fecal coliform and total enterococcal count was found to vary at all sites. The average mean count recorded showed higher abundance during monsoon followed by post monsoon and least during premonsoon. The relative rise in the TC, FC and TE count in coastal waters during monsoon was noted to be ~ 5-6 fold at Navapur coast and only marginally at Dandi creek whereas at Kelwa coast only Total coliform was recorded with marginal rise during monsoon and fecal coliforms and enterococci were absent annually. Between sites, the levels of TC, FC and TE counts were found to be highest at Dandi creek annually and lowest at Kelwa coast (Fig 2). During the study period a sporadic rise was noted in all indicator bacterial counts during premonsoon in May 2020 at Dandi creek .

In Intertidal Sediments:

The prevalence of Total coliform, fecal coliform and Total enterococcal count in intertidal sediment was studied and its load when determined annually noted Total coliform count to range from 0.48 $x10^2$ - 1.82 $x10^2$ CFU/gm, 15.58 $x10^4$ - 71.6 $x10^4$ CFU/gm, 19.26 $x10^4$ - 125.2 $x10^4$ CFU/gm at

Kelwa coast, Navapur and Dandi creek respectively. The abundance of fecal coliform count in intertidal sediments ranged from 0.1 $\times 10^2$- 0.5 $\times 10^2$ CFU/gm, 0.94 $\times 10^4$ - 16.36 $\times 10^4$ CFU/gm, 6.2 $\times 10^4$ - 26.4$\times 10^4$ CFU/gm at Kelwa, Navapur coast and Dandi creek respectively. Whereas the levels of Total enterococcal count ranged from 3.68 $\times 10^4$ - 19.26 $\times 10^4$ CFU/gm, 4.8 $\times 10^4$ - 28.86 $\times 10^4$ CFU/gm at Navapur coast and Dandi creek respectively .But was not detectable annually in intertidal sediments at Kelwa coast.

Seasonally, the abundance of all fecal indicator bacteria in intertidal sediments recorded was highest during post monsoon and lowest during monsoon at all the three sites under study. Further among sites The population density of all fecal indicator bacteria in intertidal sediments was highest at Dandi creek followed by at Navapur coast where it varied by 1-2 fold and least at Kelwa coast that varied in abundance by 3-4 order of magnitude (Fig 3).

The FIB counts recorded in both surface water and intertidal sediments among the sites the distribution and abundance of FIB in intertidal sediments was found to be higher by 2- 4 order of magnitude in comparison to surface water except for Total enterococcal count at Kelwa coast which was found to be annually undetectable both in water and intertidal sediments .

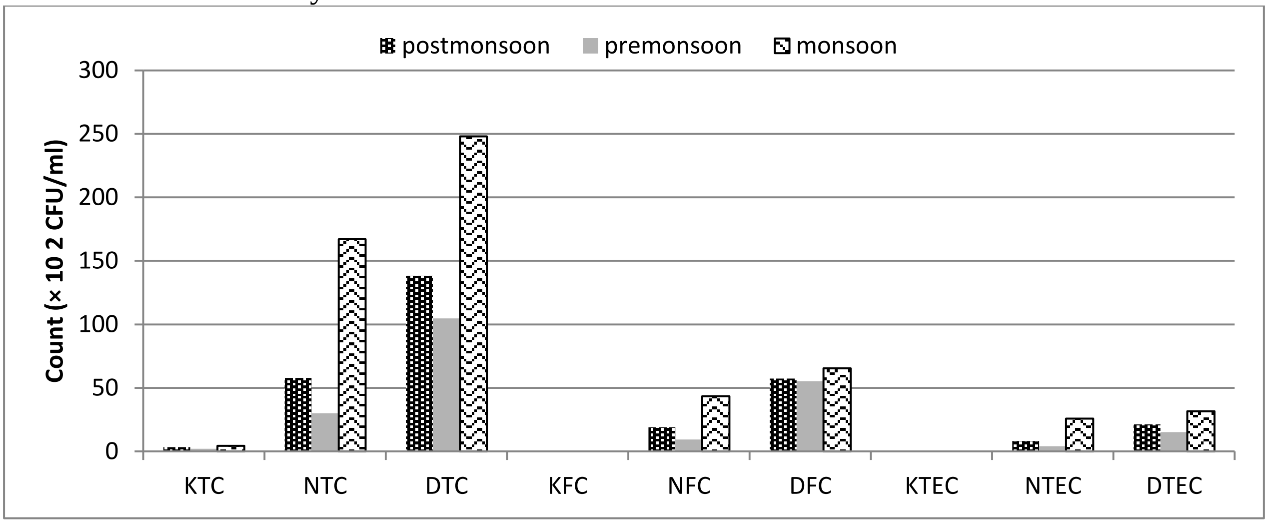

Fig:2 Seasonal variation in FIB levels in coastal water at different sites

K: Kelwa, N: Navapur, D: Dandi TC: total coliform, FC: Faecal coliform, TEC: Total enterococcal count

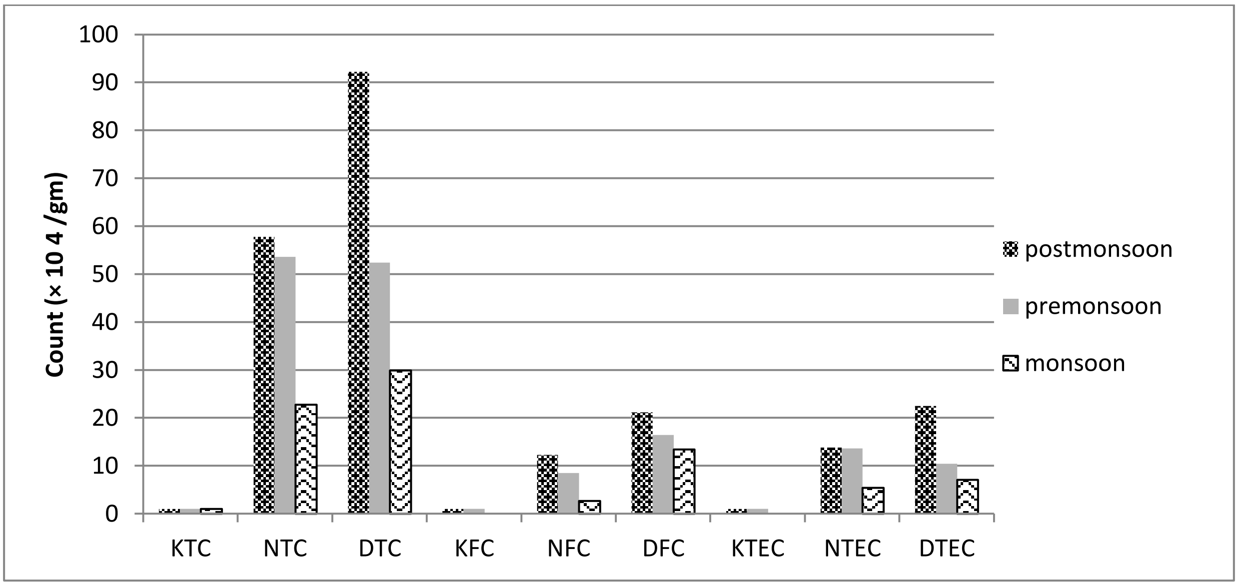

Fig:3 Seasonal variation in FIB levels in intertidal sediments at different sites

K: Kelwa, N: Navapur, D: Dandi TC: total coliform, FC: Faecal coliform, TEC: Total enterococcal count

Table 1:Seasonal variation in mean faecal indicator bacterial count in water and intertidal sediments at different sites.

Sites	Kelwa			Navapur			Dandi		
Water ($\times 10^2$ CFU/ml)	**PM**	**PRM**	**MON**	**PM**	**PRM**	**MON**	**PM**	**PRM**	**MON**
Total coliform	**0.32 (±0.18)**	**0.08 (±0.02)**	**0.36 (±0.18)**	**57.68 (±13.9)**	**30.1 (±1.7)**	**166.9 (± 88.4**	**138.2 (±41.8)**	**104.6 (±33.9)**	**248.2 (±133.8)**
Fecal coliform	**0.0**	**0.0**	**0.0**	**18.94 (±**	**9.4 (± 1.6)**	**43.4 (±19.4)**	**57.4 (±11.1)**	**55.4 (±23.5)**	**65.4 (±31.8)**
Total enterococcal	**0.0**	**0.0**	**0.0**	**8.12 (±3.03**	**4.2 (±0.3**	**25.9 (±15.6**	**21.2 (±9.5**	**2.4 (±1.0)**	**31.6 (±14.2)**
Sediment ($\times 10^4$ CFU/gm)	**PM**	**PRM**	**MON**	**PM**	**PRM**	**MON**	**PM**	**PRM**	**MON**
Total coliform	**0.01 (±0.005)**	**0.01 (±0.0)**	**0.007 (±0.002)**	**57.8 (±9.7)**	**53.6 (±12.4)**	**22.8 (±8.8)**	**92.2 (±33.3)**	**52.4 (±12.7)**	**29.9 (±12.2)**
Fecal coliform	**0.003 (±0.002**	**0.0**	**0.001 (±0.0)**	**12.3 (±3.1)**	**8.5 (±2.8)**	**2.7 (±2.2**	**21.2 (±7.2)**	**16.4 (±6.0)**	**13.4 (±8.1)**
Total enterococcal	**0.0**	**0.0**	**0.0**	**13.8 (±4.0)**	**13.6(±1.8)**	**5.4 (±2.9)**	**22.5 (±6.0)**	**10.4 (±8.6)**	**7.1 (±2.5)**

Discussion :

In the present investigation, to detect the fecal pollution in coastal environments of Palghar taluka, the annual load of fecal indicator organisms in surface water and intertidal sediments encompassing TC, FC and TEC were carried out. Further seasonal variation in abundance and distribution of FIB in different habitats were determined and were found to be prevalent in both surface and intertidal sediments in varying concentration.

In comparison to fecal coliform and Total enterococcal count the total coliform count in the study was found to be abundant which may be due to the fact that coliform group of bacteria are present in many environmental samples like soil, aquatic system. E.coli is a subset of fecal coliform are found in the foreshore sands that act as its reservoirs further acting as a non- point source of indicator in coastal water (Whitman and Nevers, 2003; Shibata et al., 2004). Fecal coliform count recorded in the study ranged from 1.34 – 184 × 10^2 CFU/ml in water and 0.001 - 26.4× 10^4 CFU/gm in sediments were found to be beyond this permissible range given by CPCB even at site recording lowest count (i.e Kelwa coast). The fecal coliform count recorded in the present study was found to be higher than recorded at Paradip port (Banoo et al., 2014), Tapi estuary (Borade et al., 2014), and fecally polluted sites at Toothukudi coast (Sugumar et al., 2008) but lower than at Ennore creek (Bharathi et al., 2018) Chapora bay in both water and sediments (Sangodkar et al., 2020). As per WHO, (2003) and USEPA guidelines for water quality standards, Enterococcus spp are adjudged as better correlated faecal indicator in marine water used to assess the microbiological quality. A geometric mean count of 35CFU/100ml is considered quantitatively as safer limit of Enetrococci spp by USEPA. No such limit is specified by CPCB standards. Levels of Enterococcal count were found to be beyond permissible limits both Navapur coast and Dandi. Levels of Enterococcal counts were found to be higher in sediments than in overlying water as opposed to fecal coliform count which remained similar in both water and sediment. Similar observation was noted by Alm et al., (2003) and concluded *Enterococci* being more prone to sunlight inactivation than *E.coli* in water and further also resistant to attack by bacteriophages and protozoans in sand. Meena et al., (2015) established fecally contaminated coastal sites of Port blair to harbour Enterococcal spp of human origin. Similarly both water and sediments along the coastal outfall at Brazil was noted to harbour different enterococcal species which were further also found to be antibiotic resistant (Carvalho et al., 2014)

Further the sediments harboured more coliform counts than the waters. Halliday and Gast (2011) documented mesocomal studies based on loss rate and growth of culturable cells of fecal origin (both E.coli and Enterococci) to persist longer in wets sands than in water. This is because sediments provide buffered temperature, more nutrients, protection from harsh environmental factors (Daveis et al., 1995) and protection from biofilm attached on sediment particles (Priester et al., 2007). Among the sites, highest density of FIB was noted at Dandi creek followed by Navapur coast, and least at Kelwa coast both in water and sediments. Reduced count at Kelwa coast may also be due to this recreation site being closed for major part during the study period due to global COVID-19 pandemic condition. Though seasonal variation was evident in mean counts of FIB, but at Dandi creek the abundance of indicator bacteria remained elevated annually in comparison to other sites with only marginal rise in count during monsoon. This may be due to cumulative effect of both industrial effluents and sewage from nearby human settlements carried by riverine source showing continual anthropogenic influence at Dandi creek. Similar observation was noted by Rodrigues at al., (2011) in estuarine environment along west coast of India as a common practice of direct discharge of untreated effluents and septic wastes into estuaries and at Tapi estuary by Maloo et al., 2014 due to industrial and domestic discharge. Also Dandi creek receives fresh water from tributaries of Surya river leading to variation in salinity and thus a favourable condition for prevalence and proliferation as Lipp et al., (2001) also underscored region with lesser salinity and high septic systems to harbour higher fecal indicator density.

Abundance of FIB at Navapur coast was relatively lower in comparison than at Dandi creek but higher than recorded Kelwa coast in the study. This could be attributed as Navapur coast is located downstream to Dandi creek and receives seaward influx during low tide from the creek resulting in mixing of effluents in creek water with shoreline, marine outfall disseminating treated and partially treated domestic and industrial effluents and due to open defecation prevalent on the shorelines. Similar finding by Sangodkar et al., (2020) detected even higher incidence then present findings, of indicator and pathogenic bacteria (ND-10^3 CFU/ml) in water and sediment (ND – 10^2 CFU/gm) at inner estuarine zone due to anthropogenically impacted riverine source loading in the estuary. Richa et al., (2018) noted beaches of Goa exposed to open defecation to harbour higher fecal coliform counts

The abundance of fecal indicator bacteria was also noted to vary seasonally in the study. Monsoon recorded higher population density of FIB in surface water whereas it was noted to be high in sediment during post monsoon. Monsoon associated elevated FIB levels have been reported by Fulke et al., (2019) and Borade et al., (2014) attributing role of rainfall associated runoff from both land based and riverine sources to contribute to the higher counts. Rainfall, coastal upwelling and resuspension of sediments leading to 80% of faecal coliforms and enterococcal count in water column are some of the contributing factors for elevated levels of faecal indicator bacteria in water column (Nallathambi et al., 2001, Byappanhalli et al., 2012, Rani et al., 2018).Continual persistence of indicator organisms is indication of contamination from land run off, direct wastewater inputs and fecal matter from human and animal sources to which the coast is exposed to (Anderson et al., 2005).

Hence in terms of fecal pollution faecal indicator bacteria was transiently observed at Kelwa coast but the study finds perpetual occurrence and abundance of faecal indicator bacteria at Navapur coast and Dandi creek in water and further higher in sediments. The creek habitat to be most impacted by fecal contamination then the inshore sites in the present study.

Conclusion:

The population density and distribution of faecal indicator bacteria was annually quantitated to assess the microbiological quality of coastal region of Palghar taluka,Maharashtra. Fecal indicator bacteria (FIB) in terms of Total coliforms (TC), fecal coliforms (FC), and total Enterococcal count (TEC) were studied. Levels of all three indicator bacterial population were noted to be higher at Dandi creek. The abundance and distribution of faecal indicator bacteria was found to vary seasonally and indicate the domestic discharges, open defecation and land run offs to contaminate the Navapur coast and Dandi creek. Higher abundance in intertidal sediments by 2 to 3 order of magnitude advocates the sand to act as reservoir for FIB which additionally indicates the continual external inputs at the sites leading to deterioration of the coastal water and the life forms coming in contact with it. Study thus helps in generating a baseline data for microbiological characteristics which can be further used to investigate the pollution status in terms of pathogens, identify the sources of contamination and undertake remedial measures for maintaining the coastal health.

Acknowledgement:

The first author is thankful to Dr. Kiran Save, I/C Principal and management of S.D.S.M. College,Palghar-401404 (M.S.) India, for providing the facilities and encouragement for conducting this study

References:

1. Abdelzaher, A. M., Wright, M. E., Ortega, C., Solo-Gabriele, H. M., Miller, G., Elmir, S., & Palmer, C. J. (2010). Presence of pathogens and indicator microbes at a non-point source subtropical recreational marine beach. Applied and environmental microbiology, 76(3), 724-732.
2. Alm, E. W., Burke, J., & Spain, A. (2003). Fecal indicator bacteria are abundant in wet sand at freshwater beaches. *Water research*, *37*(16), 3978-3982.
3. Anderson, K. L., Whitlock, J. E., & Harwood, V. J. (2005). Persistence and differential survival of fecal indicator bacteria in subtropical waters and sediments. Applied and environmental microbiology, 71(6), 3041-3048.
4. APHA, AWWA, WEF. (2012). 22nd edition, Standard Methods for examination of water and wastewater. Washington: American Public Health Association.
5. Banoo, S., Kar, R. N., & Panda, C. R. (2014). Seasonal variation and distribution of sewage pollution indicator and human pathogenic bacteria along Odisha coast. Indian journal of Marine Sciences, 43(5): 859-869.
6. Bharathi, M. D., Sundaramoorthy, S., Patra, S., Madeswaran, P., &Sundaramanickam, A. (2018). Seasonal and spatial distribution of heterotrophic bacteria in relation to physico-chemical properties along Ennore coastal waters. Indian Journal of Marine Sciences, 47(3), 587-597.
7. Boehm, A. B., & Sassoubre, L. M. (2014). Enterococci as indicators of environmental fecal contamination. In Enterococci: from commensals to leading causes of drug resistant infection.
8. Borade, S., Dhawde, R., Maloo, A., Gajbhiye, S. N., & Dastager, S. G. (2014). Occurrence and seasonal variation in distribution of fecal indicator bacteria in Tapi estuary along the West coast of India. Indian Journal of Marine Sciences. 43(3), 340-347.
9. Byappanhalli Narayanan, R. M., Sharmila, K. J., &Dharanirajan, K. (2016). Evaluation of marine water quality–a case study between cuddalore and Pondicherry coast, India. Indian journal of geomarine sciences ,45(4), 517-532
10. Carvalho, E. M., Costa, R. A., Ara&ujo, A. J., Carvalho, F. A. C., Pereira, S. P., Sousa, O. V., & Vieira, R. H. (2014). Multiple antibiotic-resistance of Enterococcus isolated from coastal water near an outfall in Brazil. *African Journal of Microbiology Research*, *8*(17), 1825-1831.
11. Davies, C. M., Long, J. A., Donald, M., & Ashbolt, N. J. (1995). Survival of fecal microorganisms in marine and freshwater sediments. Applied and Environmental Microbiology, 61(5): 1888-1896.
12. Fulke, A.B., D'Souza, E., Maloo, A., Ram, A., Mulani, N. and Majithiya, D., 2019. Determination of spatio-temporal influences on the distribution of fecal indicator organisms along the north-west coast of India. Indian journal of Geo Marine Sciences. Vol. 48 (05), pp. 698-706.
13. Halliday, E., & Gast, R. J. (2011). Bacteria in beach sands: an emerging challenge in protecting coastal water quality and bather health. Environmental science & technology, 45(2): 370-379.

14. Hassard, F., Gwyther, C. L., Farkas, K., Andrews, A., Jones, V., Cox, B., and Malham, S. K. (2016). Abundance and distribution of enteric bacteria and viruses in coastal and estuarine sediments—a review. Frontiers in microbiology, 7, 1692.
https://www.ncbi.nlm.nih.gov/sites/books/NBK190421/ (Acessed on October 2020)
15. Korajkic A., McMinn B.R. and Harwood V.J., (2018), Relationships between microbial indicators and pathogens in Recreational Water Settings, International Journal of Environmental Research and Public Health, 15(12), 1-39.
16. Kumarasamy P., Vignesh S., James A., Muthukumar, K & Rajendran A., (2009). Enumeration and identification of Pathogenic Pollution Indicators in Cauvery River, South India, Research Journal of Microbiology. 4(12), 540-549.
17. Kutty, S. N. and Sebastian C.D., (2019) Microbial pollution assessment in major tourist beaches of Kerala. Online International Interdisciplinary Research Journal, {Bi-Monthly}, ISSN 2249-9598, 9 (4),29-34
18. Lipp, E. K., Kurz, R., Vincent, R., Rodriguez-Palacios, C., Farrah, S. R., & Rose, J. B. (2001). The effects of seasonal variability and weather on microbial fecal pollution and enteric pathogens in a subtropical estuary. Estuaries, 24(2):266-276
19. Meena, B., Anburajan, L., Sathish, T., Raghavan, R. V., Jha, D. K., Venkateshwaran, P., and Kirubagaran, R. (2015). Enterococcus species diversity and molecular characterization of biomarker genes in Enterococcus faecalis in Port Blair Bay, Andaman and Nicobar Islands, India. Marine Pollution Bulletin, 94(1-2), 217-227.
20. Nallathambi, T., Eashwar, M., and Kuberaraj, K. (2002). Abundance of indicator and general heterotrophic bacteria in Port Blair bay, Andamans. Indian Journal of Marine Sciences, 31(1), 65-68.
21. Priester, J. H., Horst, A. M., Van De Werfhorst, L. C., Saleta, J. L., Mertes, L. A., & Holden, P. A. (2007). Enhanced visualization of microbial biofilms by staining and environmental scanning electron microscopy. Journal of microbiological methods, 68(3), 577-587
22. Richa, S., Singh, A. P., & Gaurav, B. (2018). Microbiological and physico-chemical study in coastal water of Port Goa, India. Trends in Biosciences, 11(16), 2608-2619.
23. Robin, R. S., Vardhan, K. V., Muduli, P. R., Srinivasan, M., &Balasubramanian, T. (2012). Preponderance of enteric pathogens along the coastal waters of Southern Kerala, Southwest coast of India. Marine Science, 2(1), 6-11
24. Sangodkar, N., Gonsalves, M.J., Shanbhag, Y., Rayadurga, A.S. and Nazareth, D., (2020). Prevalence of indicator and potential pathogenic bacterial groups in the Chapora bay-estuarine system, Goa, central west coast of India. Environmental Monitoring and Assessment, 192(6), 397-397.
25. Sugumar, G., Chrisolite, B., Velayutham, P., Selvan, A., & Ramesh, U. (2008). Occurrence and seasonal variation of bacterial indicators of faecal pollution along Thoothukudi Coast, Tamil Nadu. Journal of environmental biology, 29(3), 387.
26. Tyagi, V. K., Chopra, A. K., Kazmi, A. A., & Kumar, A. (2006). Alternative microbial indicators of faecal pollution: current perspective. Journal of Environmental Health Science & Engineering, 3(3), 205-216.
27. Vignesh, S., Dahms, H.U., Emmanuel, K.V., Gokul, M.S., Muthukumar, K., Kim, B.R. and James, R.A., (2014). Physicochemical parameters aid microbial community? A case study from marine recreational beaches, Southern India. Environmental monitoring and assessment, 186(3), 1875-1887
28. Whitman, R. L., & Nevers, M. B. (2003). Foreshore sand as a source of Escherichia coli in nearshore water of a Lake Michigan beach. Applied and environmental microbiology, 69(9), 5555-5562.
29. WHO (2003).Coastal and Fresh Waters. In Guidelines for Safe Recreational Water Environments; World Health Organization: Geneva, Switzerland,; Volume 1.
30. World Health Organization. (2011). Microbial fact sheets. World health organization guidelines for drinking-water quality (WHO GDWQ), 271-273.