



ENVIS NEWSLETTER

MICROORGANISMS AND IMPACT ON PUBLIC HEALTH

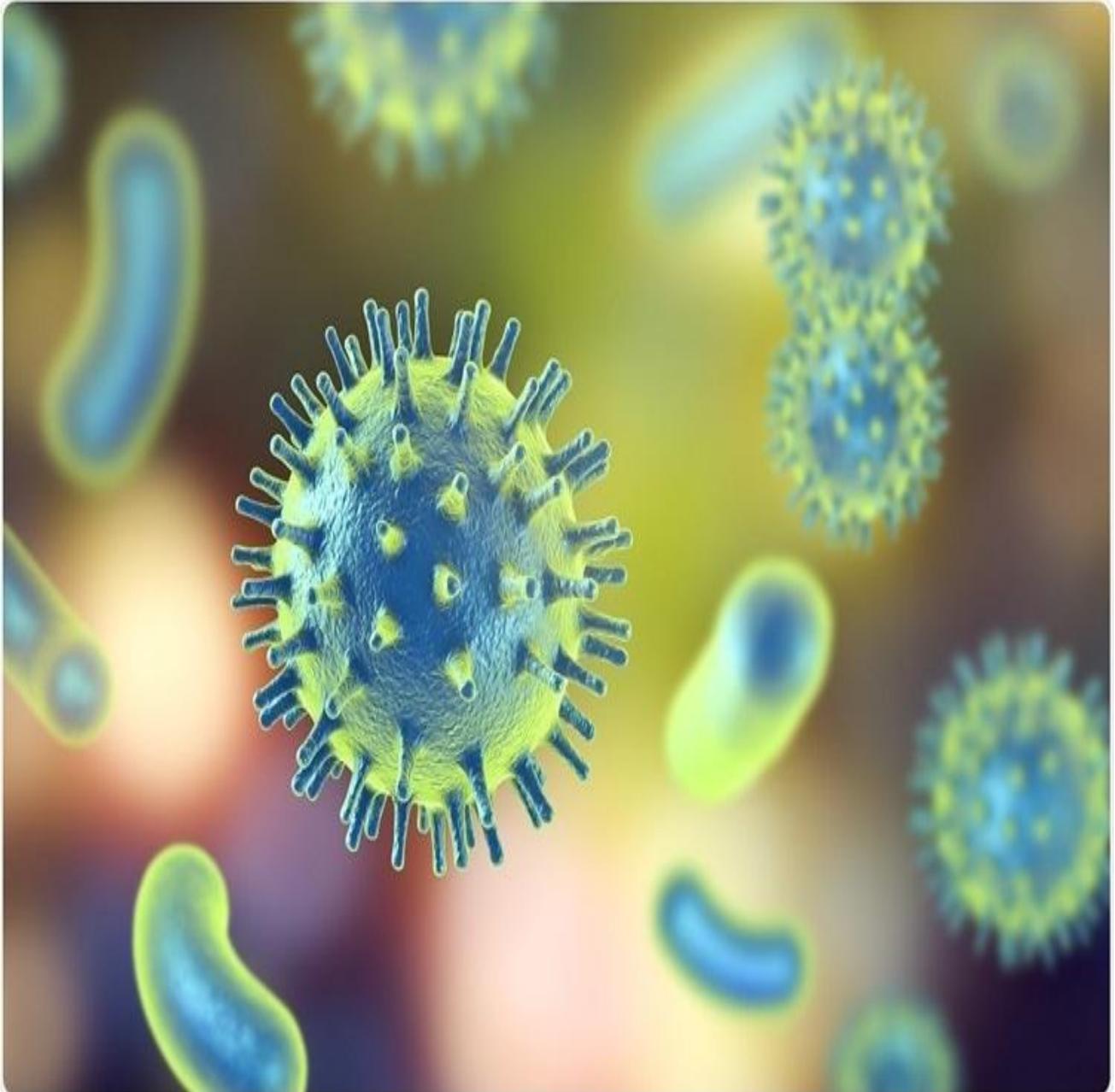
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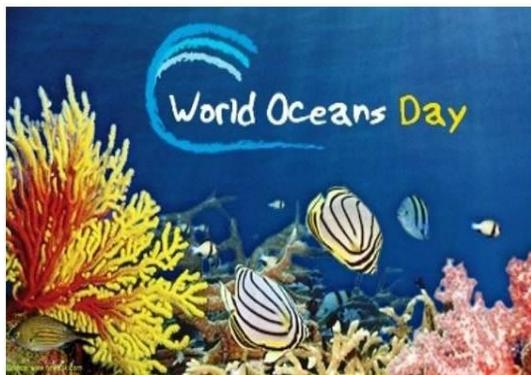
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ENVIS Newsletter on 'Microorganisms and Impact on Public Health', a quarterly publication, brings out original research articles, reviews, reports, research highlights, news-scan etc., related to the thematic area of the ENVIS Centre. In order to disseminate the cutting-edge research findings to user community, ENVIS Centre on Microorganisms and Impact on Public Health invites original research and review articles, notes, research and meeting reports, details of forthcoming conferences / seminars / symposia / trainings / workshops for publication in the newsletter.

The articles and other information should be typed in double space with a maximum of 8 - 10 typed pages. Photographs/line drawings and graphs need to be of good quality with clarity for reproduction in the newsletter. For references and other details, the standard format used in the newsletter may be followed.

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22nd APRIL 2019

From the Coordinator's Desk!

Dear Readers,

Greetings!

Water is the most important natural resource for life. People have been abusing and misusing water since long. Industrial wastes, garbage and other remains of used commodities have been getting dumped in rivers and oceans causing contamination of water. It is also contaminated of toxic chemicals discharged from factories, pesticides used in agriculture and so on. Due to the poor water supply system, contact of sewage with the water supply line contaminates water used for drinking. Contaminated water has become a serious threat to human beings, fish and the animals. The poor and less developed countries are facing greater problem of water pollution as they have dilapidated water treatment and supply system and also lack in proper waste disposal system. The pollution in water could be measured based on chemical and biological indicators. Life is ultimately about choices and so is pollution. We can live with sewage strewn beaches, dead rivers, and fish that are too poisonous to eat. Or we can work together to keep the environment clean so the plants, animals, and people who depend on it remain healthy.

In this context, present issue contains a scientific article on preliminary investigation on quality of potable waters in chennai city during summer 2019 through microbial indicators along with other interesting topics such as effects of intestinal bacteria on blood sugar and lipid levels, use of AI to detect patterns of gut microbes for predicting cholera risk, alteration of skin microbiome due to swimming in ocean, new compounds that control bacterial communications and many more.

Dr. C. Arulvasu

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ENVIS Newsletter Vol.17/ Issue 1/ Jan. - Mar. 2019

MICROBIAL QUALITY OF POTABLE WATERS DURING SUMMER IN CHENNAI

N.K. Udaya Prakash¹, V. Subha Shree², S. Revathy², K. Tara Devi Sharma², K. Mohammed Yasar³, A. Mohamed Azaruddin³, J. Aswin³, D. Santhosh³, S. Aruna Devi⁴, M. Priyadharshini⁴ and S. Bhuvaneshwari^{5*}

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Keywords:

Microbial Quality
Total Plate Count, Coliforms
Vibrio
TCBS agar
Water in Tankers

ABSTRACT

The study was conducted to know the microbial quality of potable waters in Chennai city during summer, 2019 where acute shortage of drinking water is reported. A total of 27 water samples were collected in Chennai from different water sources (well water, bore water, metro water supplied through pipe line and through tankers). Samples were tested for their microbial quality through enumeration of total bacteria, coliform and *Vibrio* as microbial indicators. The bacterial colonies were enumerated by serial dilution method using Nutrient agar, MacConkey agar and TCBS agar for total bacteria, Coliforms and *Vibrio* respectively. An average of 16,177 CFU/ml of total bacteria was recorded from the water samples. An average count of 216.2 CFU/ml and 44.9 CFU/ml of coliforms and vibrio were recorded respectively. Bacteria and coliforms were high in well waters and *Vibrio* was recorded maximum in the water sample supplied through Tankers. The study strongly recommends continuous monitoring of potable waters for microbial presence of drinking water supplied.

Introduction

Water is a vital element for life next to oxygen. Though 71% of earth's surface is covered with water, availability of safe drinking water to human population is becoming scarce. India being second populated country in the world suffers water scarcity because of improper water maintenance, inadequate saving of rain water, urbanization and failing monsoons. India, suffers to an extent where it loses 3.4 million children every year due to unavailability and scarcity of clean and potable drinking water (WHO, 2003). According to UNICEF (2005) assessment, contaminated water leads to 4000 deaths in children each day and states that we can decrease disease burden all over the world by 4% by improving the water quality. Under-five years' age group, Diarrhea is the third most common cause of death in children and contributes to 13% of death, killing 300,000 children each year in India alone. Sadly, Indian population who has access to mobile phones is higher than those who have access to safe drinking water, which is of utmost importance, to maintain the health of the community (Malathy *et.al*, 2017).

Water borne diseases are spread mainly through drinking, washing, bathing in contaminated water or by

eating food which is exposed to contaminated water. The general symptom of water borne diseases includes acute fever, chillness, vomiting, diarrhea, nausea and headache. Waterborne diseases in the household of Chennai were found to be 12.2% (Dhanasekar *et al.*, 2017). The disease risk associated with drinking water in developing countries is due to pathogens, which include viruses, bacteria, and protozoans that spread either via oral or fecal route. *Campylobacter jejuni*, *Microsporidia* sp., *Yersinia enterocolitica*, *Cyclospora* sp., and environmental bacteria like *Mycobacterium* sp., *Aeromonas* sp., *Legionella pneumophila* are serious water borne pathogens causing illness. The continuous disease outbreaks during every year are due to these pathogens spread through water sources and interfuse directly through food and water. The main distribution of many water-borne pathogens varies substantially from one country to another. Some pathogens such as *Vibrio cholerae*, Hepatitis E virus, and the disease *Schistosomiasis* are restricted to certain tropical countries; others, such as *Cryptosporidiosis* and *Campylobacteriosis*, are probably widespread (Devipriya and Kalaivani, 2012).

Contaminated drinking water is believed to be the cause of various diseases which is on raise during summer (Loganathan *et. al.*, 2011). Also, consumption of contaminated water in India has led to frequent outbreaks of waterborne diseases such as cholera, typhoid, and hepatitis

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A and E (Joseph *et. al.*, 2018). The supply of drinking water to residents of the Chennai is controlled by Chennai Metropolitan Water Supply and Sewage Board (Metro Water). As of 2012, it supplies about 830 million liters of water every day through pipelines. Chennai with a population of 8.24 million as per 2011 census, suffers from water stress since, the entire plain is dependent on rainfall. Thus, the study was conducted to know the quality of water samples for the bacterial presence, through counts of total bacteria, coliforms and vibrio in potable waters of Chennai city during summer, 2019 where acute shortage of drinking water is reported.

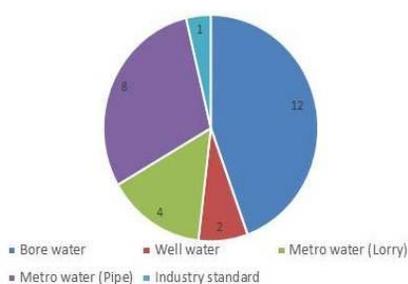
MATERIAL AND METHODS

Collection of water samples

A total of 27 water samples were collected in Chennai from different water sources such as well water, bore water, metro water supplied through pipe line and through tankers (Fig. 1) during the month of May 2019 using sterile containers. The collected water samples were processed immediately for microbial analysis. The sampling sites are shown in Fig. 2.

Fig. 1. Type and number of sample collected in Chennai for enumeration of microbial quality

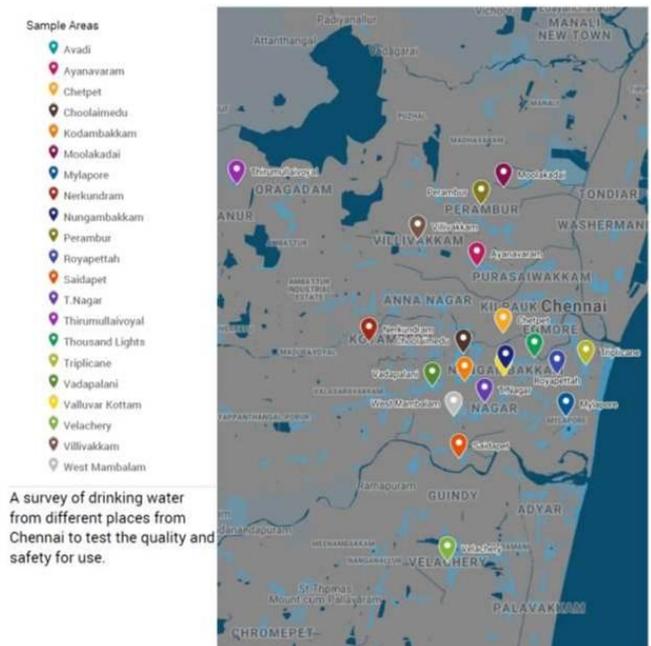
Types of water samples collected in Chennai



Microbial analysis

Water samples were serially diluted to detect the total count of bacteria, coliforms and *Vibrio* present. One ml of water sample was serially diluted to 1:10; 1:100; 1:1000 and further diluted if required using sterile distilled water. One ml of serially diluted sample (1:100), was added with Nutrient agar (Himedia, Bombay) and incubated at $37 \pm 2^\circ\text{C}$ for 24 hours for enumeration of

Fig. 2. Map showing water sampling locations in Chennai.



total bacteria. Similarly, 1 ml of diluted sample (1:10) was mixed with MacConkey Agar (Himedia, Bombay) and incubated at $37 \pm 2^\circ\text{C}$ for 24 hours for the growth and enumeration of coliform bacteria. For the enumeration of *Vibrio*, 1ml of sample was directly added without dilution to a sterile petridish and mixed with Thiosulfate Citrate Bile salts Sucrose agar (TCBS) (Himedia, Bombay) and was incubated at $37 \pm 2^\circ\text{C}$ for 24 hours. The plates were maintained in triplicates and the average values were recorded.

Presentation of data

The data was expressed as colony forming units (CFU) per ml of sample and calculated using the formula given below.

$$\text{Average CFU/ml of water sample} = \frac{\text{Total no. of colonies recorded in all plates}}{\text{No. of replicates}} \times \text{dilution factor}$$

RESULTS

A total of 27 samples were analyzed for presence of total count of bacteria, coliforms and *Vibrio* in potable water collected from different sources during the month of May 2019. An average of 16,177 CFU/ml of total bacteria, 216.2 CFU/ml of coliforms and 44.9 CFU/ml of *Vibrio* were enumerated from samples collected.

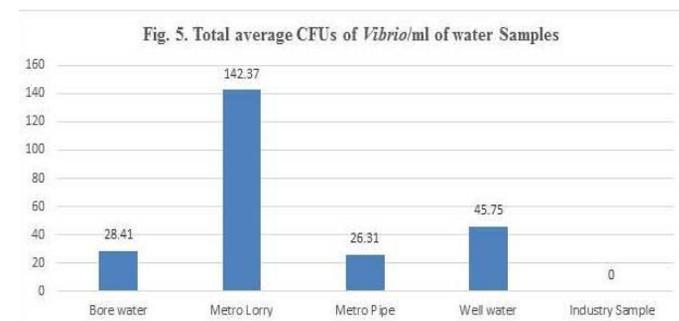
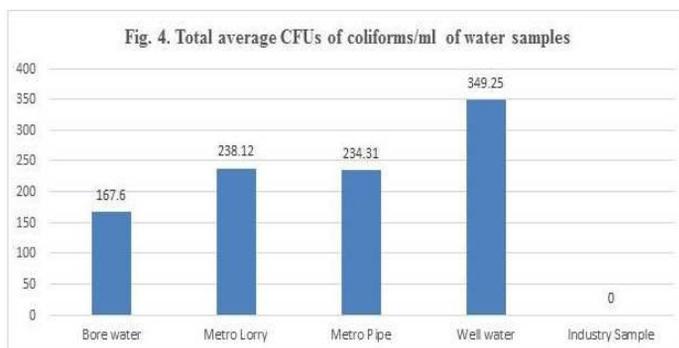
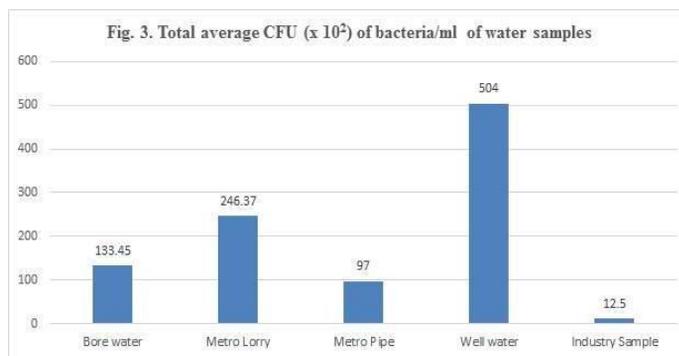
The total bacterial count ranged from minimum of 1250 CFU/ml in Bisleri to maximum of 87900 CFU/ml for well water taken from Royapettah I. Total coliform counts ranged from absence of colonies to 826.5 CFU/ml. The highest

number of coliforms were recorded from the pipe water supplied by Chennai metro in Saidapet II. Similarly, *Vibrio* ranged from 0 to 333 CFU/ml. The high number of *Vibrio* was recorded for the sample collected from Tankers supplied by Chennai metro. The sample number, place of collection, average count of total bacteria, coliforms and *Vibrio* in water samples from different sampling sites in Chennai is provided in Table 1.

Sl. No.	Place of Collection	Total Count (x 10 ²)/ml	Total Coliforms/ml	Total Vibrios/ml	Source
1	Ayanavaram	63.5±0.7	97.5±0.7	22±1.4	Bore water (Ground)
2	Avadi	311.5±0.7	501.5±0.7	62±1.4	Metro water (Lorry)
3	Chetpet	47.5±7.7	34.5±2.1	13.5±3.5	Bore water (Ground)
4	Choolaimedu	21±0	110±0	7.5±0.7	Metro water (Pipe)
5	Kodambakkam I	51±2.8	180.5±3.5	19±2.8	Metro water (Pipe)
6	Kodambakkam II	173±8.4	124±0	11.5±0.7	Metro water (Pipe)
7	Moolakadai	43.5±0.7	70.5±2.1	11.5±0.7	Bore water (Ground)
8	Mylapore	308.5±3.5	82.5±7.7	NIL	Bore water (Ground)
9	Nerkundrum	50.5±2.1	NIL	NIL	Metro water (Pipe)
10	Nungambakkam I	142.5±13.4	215.5±4.9	39.5±2.1	Bore water (Ground)
11	Nungambakkam II	146±1.4	123±0	80±2.8	Metro water (Pipe)
12	Perambur I	330±14.1	65.5±0.7	47.5±0.7	Bore water (Ground)
13	Perambur II	41±0	137.5±7.7	119±1.4	Metro water (Lorry)
14	Royapettah I	879±1.4	94±7.0	NIL	Well water
15	Royapettah II	421±15.5	216.5±9.2	333±18.3	Metro water (Lorry)
16	Saidapet I	23.5±0.7	410.5±0.7	NIL	Bore water (Ground)
17	Saidapet II	268.5±2.1	826.5±7.7	92.5±7.7	Metro water (Pipe)
18	Thousand lights	24±0	123±2.8	NIL	Metro water (Pipe)
19	Thirumullaivoyal	129±4.2	604.5±7.7	91.5±6.3	Well water
20	Triplicane	266.5±0.7	79.5±2.1	11.5±0.7	Bore water (Ground)
21	T. Nagar	159.5±4.9	41.5±0.7	27.5±0.7	Bore water (Ground)
22	Vadapalani	81±0	670±7.0	96.5±0.7	Bore water (Ground)
23	ValluvarKottam	42±0	387.5±3.5	NIL	Metro water (Pipe)
24	Velacherry	53.5±3.5	369.5±10.6	55.5±4.9	Bore water (Ground)
25	Villivakam	66.5±0.7	175±2.8	16±1.4	Bore water (Ground)
26	West Mambalam	212±1.4	97±0	55.5±2.1	Metro water (Lorry)
27	Bisleri	12.5±0.7	NIL	NIL	Industrial Standard

Well water samples had maximum bacterial average of 50400 CFU/ml followed by water supplied through Tankers of Chennai metro containing 24637 CFU/ml. The least count of bacteria was observed in Industry packed Bisleri water (Figure 3). Similarly, maximum counts of coliforms were found in well water followed by water supplied through Tankers and pipes of Chennai metro. (Figure 4). Water supplied through

Tankers of Chennai metro contained maximum *Vibrio*, followed by well water samples (Figure 5).



DISCUSSION

According to the World Health Organisation, diarrheal diseases account for an estimated 4.1% of the total daily global burden and are responsible for the deaths of 1.8 million people every year. It was estimated that 88% of that burden is attributed to unsafe water supply, sanitation and hygiene (Hatami, 2013). Bacterial values of less than 100 CFU/ml were considered to be normal. Values from 100 to 500 CFU/ml is anticipated during seasonal increases or at certain locations in the system (dead end, low residual) which would suggest a need for flushing. In other cases, 5 to 10 fold increases over normal levels were set as a guideline to prompt an investigation (US EPA, 1984). In our study, none of the samples analyzed met this condition of being potable water. Maximum of 87,900 CFU/ml of bacteria was recorded in the well water sample

collected from Royapettah followed 42100 CFU/ml in water supplied through Tankers in the same locality.

Increased number of coliform in water samples will become a reason for epidemic outbreak of typhoid, dysentery, and other complications. The popular coliform, *Salmonella* is known to cause deadly typhoid and gastroenteritis. The group of species of *Salmonella typhi* and *Salmonella paratyphi*, is the cause of the enteric fevers, typhoid and paratyphoid. Another group, consisting of over 2000 serotypes of what is now considered to be one species, *Salmonella enterica*, causes gastroenteritis. In present study, none of the samples except that of Nerkundrum recorded negative for coliforms. The CFU ranged from 34.5/ml in bore water collected from Chetpet and the maximum of 826.5 CFU/ml was recorded from the water supplied through pipeline by Chennai metro. On an average the coliforms was found to be dominant in well water followed by water supplied through Tankers by Chennai metro. The reason for recording more number of coliforms in well water can be attributed to an open area of well and the seepage of nearby drainages, thus contaminating the well water.

A standard potable drinking water should be totally free from coliforms and vibrio population. The genus vibrio comprises of diverse species out of which three species are known to cause disease, *Vibrio cholerae*, the causative agent of cholera, *V. parahaemolyticus* and *V. vulnificus*, which together are responsible for most cases of fatal poisoning (Janelle *et.al.*, 2004). The results obtained from TCBS agar plates showed that, out of 27 samples, seven samples including industrial sample recorded negative and remaining samples showed the presence of vibrios. The presence of vibrio in water is of major concern as there may be an outbreak of cholera at times.

The reason for large bloom of coliforms and vibrios might be due to formation of biofilm layers on tankers, pipelines and in the storage containers. Broken pipes, leaking sewer lines and inadequate maintenance of old pipe networks are the primary causes of contamination. It is noticed that a single lorry supplying contaminated water can easily spread infections in many areas.

The study underlines the importance of following a) Check point for Municipality water Tankers and private to continuously monitor the water turbidity and quality. Apart from microbial contamination, waters should also be checked for total dissolved

solvents and other heavy metals regularly to dispense safe water to public. b) Creation of awareness among rural and urban areas about microbial contamination of water leading to disease. c) Proper treatment and release of effluents and domestic sewage. d) Encouragement of toilet usage and awareness among rural areas to avoid any faecal contamination in potable waters. e) Maintenance of proper infrastructure for harvesting rainwater. f) During rainy seasons and disease outbreaks water should be boiled and filtered before drinking.

Although, the study possesses limited number of samples, we recommend continuous monitoring of potable waters for microbial presence and expanded parameters to monitor presence of *Salmonella*, *Shigella*, *Pseudomonas* and other pathogens. Although, majority of residents purchase packed drinking water (mineral water) for cooking and drinking, this should be kept in mind that whose genuineness again is not monitored (Malathy *et.al* 2017).

Funding

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RESEARCH REPORTS

How intestinal bacteria can affect blood sugar and lipid levels

Intestinal bacteria have attracted recent attention since they were discovered to influence various physiological functions and diseases in humans. Researchers from Kumamoto University in Japan analyzed the influence of changes in intestinal bacteria on sugar and lipid metabolism, and found that secondary bile acids produced by the bacteria can influence blood glucose and lipid concentrations as well as parts of their molecular mechanisms. This result is expected to lead in the treatment of metabolic diseases such as diabetes and dyslipidemia by targeting intestinal bacteria that produce secondary bile acid.

More than 100 trillion bacteria from an estimated 1,000 different species inhabit our intestines. It has been reported that the profiles of intestinal bacteria in obese and non-obese people tend to be different and also involved with the health of the host. The bacteria may affect energy consumption and fat accumulation of host body. In addition, it is known that these bacteria are also associated with lifestyle diseases such as type 2 diabetes, nervous diseases such as autism, and intestinal diseases such as colon cancer.

One factor that alters intestinal bacteria is the administration

of antimicrobials. It is becoming clear that these drugs cause dysbiosis in the qualitative and quantitative balance of bacterial populations in the intestine and have various effects on vital functions. For example, hypoglycemia is a serious, but rare side effect of antibiotics. In fact, some antibiotics, such as gatifloxacin, have been discontinued due to their side effects. Furthermore, taking antibiotics in infancy or childhood has been reported to accelerate weight gain.

Previous research had showed that dysbiosis due to antibiotic administration influences protein expression levels in the liver, an organ responsible for sugar and lipid metabolism. Thus, researchers at Kumamoto University decided to clarify the influence of antibiotic-caused dysbiosis on sugar and lipid metabolism and the mechanism thereof.

A dysbiosis mouse model was prepared by administering antibiotics for 5 days. Compared to non-antibiotic treated mice, the blood glucose levels and lipid (triglyceride) concentrations in the experimental model decreased to 64% and 43% respectively. To assess the mechanisms related to these reductions, researchers focused on secondary bile acids. These acids are metabolites produced by intestinal bacteria that control the liver functions involved in sugar and lipid metabolism.

In the experimental mouse model, intestinal bacteria producing secondary bile acids decreased. Additionally, the concentrations of secondary bile acids (lithocholic and deoxycholic acid) in the mouse liver were reduced to 20% and 0.6% respectively compared to non-antibiotic treated mice. When secondary bile acid is supplemented at the same time as antibiotic administration, blood glucose and blood triglyceride levels were recovered. This result indicates that the secondary bile acid produced by intestinal bacteria affects sugar and lipid metabolism of the host.

Next, the researchers used quantitative proteomics to comprehensively analyze the amount of proteins to assess how secondary bile acids produced by intestinal bacteria influence liver sugar and lipid metabolism. In the livers of the dysbiosis mouse model, the expression levels of proteins involved in glycogen metabolism (storage of sugar) and in the biosynthesis of cholesterol and bile acids were found to have changed. Moreover, the change was restored through supplementation of secondary bile acids.

“Our research shows that enterobacteria and the

secondary bile acids that they produce may be involved in the change of concentration of sugars and lipids in living bodies," said Kumamoto University Professor Sumio Ohtsuki, leader of the study. "He added it is expected that these bacteria will be a future target for the prevention or treatment of metabolic diseases such as diabetes or dyslipidemia."

Source: www.sciencedaily.com

Kitchen towels could contribute to the growth of potential pathogens that cause food poisoning

Researchers from the University of Mauritius have shown that factors such as family size, type of diet, multi-usage of towels, among other factors that impact the growth of pathogens on kitchen towels, potentially causing food poisoning.

49% of the kitchen towels collected in the study had bacterial growth which increased in number with extended family, presence on children and increasing family size. The towels for multipurpose usage (wiping utensils, drying hands, holding hot utensils, wiping/cleaning surfaces) had a higher bacterial count than single-use towels and humid towels showed higher bacterial count than the dry ones. Out of the 49 samples which were positive for bacterial growth, 36.7% grew coliforms, 36.7% *Enterococcus* sp. and 14.3% *S. aureus*.

In this study, they investigated the potential role of kitchen towels in cross-contamination in the kitchen and various factors affecting the microbial profile and load on kitchen towels said Dr. Susheela D. Biranjia-Hurdoyal, Senior Lecturer, Department of Health Sciences, University of Mauritius, lead author on the study.

A total of 100 kitchen towels were collected after one month of use. The researchers cultured the bacteria and identified them by standard biochemical tests. They also determined the bacterial load on the towels.

S. aureus was isolated at a higher rate from families of lower socio-economic status and those with children. The risk of having coliforms (*Escherichia coli*) was higher from humid towels than the dried ones, from multipurpose towels than single-use ones and from families on non-vegetarian diets.

Coliform and *S. aureus* were detected at significantly higher prevalence from families with non-vegetarian diets. *E. coli* is a normal flora of human intestine and it is released in large numbers in human feces. The presence of *E. coli* indicates possible fecal contamination and lack of hygiene practices.

Dr. Biranjia-Hurdoyal said the data indicated that unhygienic practices while handling non-vegetarian food could be common in the kitchen. The presence of potential pathogens from the kitchen towels indicates that they could be responsible for cross-contamination in the kitchen and could lead to food poisoning. "Humid towels and multipurpose usage of kitchen towels should be discouraged. Bigger families with children and elderly members should be especially vigilant to hygiene in the kitchen," she said.

This study was done by Moodelly V as part of an undergraduate project under the supervision of Susheela D Biranjia-Hurdoyal, Senior Lecturer from the Department of Health Sciences, University of Mauritius, Mauritius. The practical component of the work was funded by the University of Mauritius.

Source: www.sciencedaily.com

AI detects patterns of gut microbes for cholera risk A hundred kinds of microbes out of 4,000 determine susceptibility to cholera

Researchers from Duke University, Massachusetts General Hospital and the International Centre for Diarrheal Disease Research in Dhaka, Bangladesh have used machine learning algorithms to spot patterns within communities of bacteria living in the human gut that no human would ever be able to pick out. These patterns could indicate who among the approximately one billion people around the globe at risk of cholera infection will get sick with the diarrheal disease.

"These are patterns that even the most sophisticated scientist couldn't detect by eye," said Lawrence A. David, Ph.D., a senior author of the study and assistant professor of molecular genetics and microbiology at Duke School of Medicine. He added, "While some people are warning about artificial intelligence leading to killer robots, we are showing the positive impact of AI in its potential to overcome disease."

The research was published in the Journal of Infectious Diseases, suggested that a focus on gut microbes may be important for developing improved vaccines and preventive approaches for cholera and other infectious diseases.

"Our study found that this 'predictive microbiota' is as good at predicting who gets ill with cholera as the clinical risk factors that we've known about for decades," said Regina C. LaRocque, M.D., MPH, of the Massachusetts General

Hospital Division of Infectious Diseases, a senior author of the study and assistant professor of medicine at Harvard Medical School. “We've essentially identified a whole new component of cholera risk that we did not know about before.”

Cholera can spread rapidly in areas with unsafe drinking water and inadequate sanitation, causing millions of cases of acute watery diarrhea every year. Despite its global impact, scientists still do not completely understand why some people who come into contact with the cholera bacterium become sick while others do not. Some studies have pinpointed a few risk factors, such as age, blood type and previous infections, but these only partially explain the differences in clinical outcomes after exposure to the pathogen.

In this study, David and LaRocque teamed up with Firdausi Qadri, Ph.D., a leader in cholera vaccine research in Bangladesh, to see whether the gut's trillions of resident bacteria collectively known as the gut microbiota might also play a role in cholera risk.

The researchers collected rectal swab samples from residents of Dhaka who lived in the same household with a patient hospitalized with cholera, and thus were at imminent risk of developing the disease. Of 76 household contacts studied, about a third went on to develop cholera during the follow-up period and about two-thirds remained uninfected.

The researchers profiled the microbiota from the household contacts' rectal swab samples using sequencing technology and then loaded all the data into a computer for analysis. They trained the machine to scan the results from 4000 different bacterial taxa in each of the samples, looking for patterns that distinguished those who got sick from those who didn't. Eventually, the machine hit on a set of 100 microbes associated with susceptibility to cholera.

“Normally, you have to eyeball the data, studying one bacterial species at a time in hopes of finding a signal that is associated with infection,” said Firas S. Midani, a lead author of the study and a graduate student in David's lab. He added “Machines have the ability to look at a hundred species at a time, and amalgamate them into one signal”.

The team showed that the model generated by artificial intelligence could predict illness even better than models previously built by infectious disease experts. The model also suggested hypotheses that might explain why the patterns identified by the computer are associated with disease. For

example, when the researchers picked a bacterial species identified by their model and studied it in the laboratory, they found that the bacteria promoted the growth of cholera in test tubes. Their findings indicate that the composition of the gut microbiota could create an environment that is more or less hospitable to pathogens.

LaRocque said that Scientists have long had a hunch that gut bacteria might affect a person's susceptibility to diarrheal diseases, but the present study is among the first to show this in a real-world setting.

Source: www.sciencedaily.com

ONLINE REPORTS

Turning up the heat on pathogenic bacteria

Pathogenic bacteria come alive at the metabolic level when they enter the warmth of the human gut, firing up genes that encode toxins and other compounds harmful to our bodies. A KAUST-led study shows how a critical bacterial protein senses changes in temperature to slacken DNA strands and boost gene expression in diarrhea-inducing bugs.

“Having determined how these bacteria sense that they are inside humans, we could try to conceive of small molecules to perturb this mechanism,” says research scientist Umar Hameed. “Such compounds would block bacteria from adapting to their environment, which would weaken them and facilitate eliminating them.”

Gut-dwelling bacteria that cause food-borne illnesses, including *Salmonella*, disease-causing strains of *E. coli* and the cholera pathogen *Vibrio cholerae*, all use a protein called histone-like nucleoid-structuring protein (H-NS) to condense their DNA and broadly restrict gene expression. H-NS forms multiunit complexes that help the microbes stay relatively dormant when free-floating in the environment. However, these complexes must break up for the protein to release its grip on DNA, which then allows the bacteria to thrive within a warm-blooded host, explained group leader, Stefan Arold.

Before coming to KAUST, Arold, who has studied H-NS for more than 15 years, previously determined the three-dimensional shape of part of the protein. While he had shown that its complexes collapse at human body temperature, it wasn't clear how that happened.

In his lab at KAUST with colleagues Łukasz and

Mariusz Jaremko, Arold and Hameed have now combined structural and biophysical experiments. This has enabled them to identify the exact part of H-NS, dubbed “site 2” that changes its conformation in response to a temperature rise, prompting the protein complexes to fall apart.

Computer simulations, performed with KAUST colleague Xin Gao and Jianing Li, at the University of Vermont, U.S., helped Arold's team deepen these insights. Ultimately, they demonstrated that the partial disassembly of the H-NS complexes leads the protein to adopt a self-inhibiting form that also blocks its ability to bind and recognize DNA.

Arold and his team now hope to develop drugs that can stabilize the connection between proteins at human body temperature and prevent H-NS complexes from fragmenting. “If we can find compounds that reinforce the structure against heat-induced unfolding,” Arold says, “then bacteria would not express toxins anymore; they would remain inoffensive and be generally weakened inside humans.”

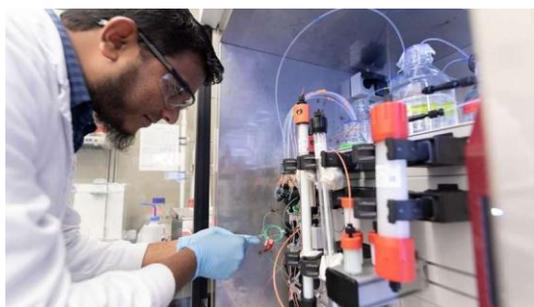


Image: Umar Hameed performs size-exclusion chromatography, an important step in the purification of proteins.

Image credit: King Abdullah University of Science and Technology (KAUST).

Source: www.phys.org

Ocean swimming alters skin microbiome, increasing vulnerability to infection

Swimming in the ocean alters the skin microbiome and may increase the likelihood of infection, according to research presented at ASM Microbe 2019, the annual meeting of the American Society for Microbiology.

Marisa Chattman Nielsen, MS, a Ph.D. student at the University of California, Irvine, and the lead author on the study said that their data demonstrate for the first time that ocean water exposure can alter diversity and composition of human skin microbiome and added that while swimming normal resident bacteria were washed off while ocean bacteria were deposited

onto the skin.

The researchers detected ocean bacteria on all participants after air drying and at six and 24 hours post-swim, but some participants had acquired more ocean bacteria and/or had them persist for longer.

The research was motivated by previous studies which have shown associations between ocean swimming and infections, and by the high prevalence of poor water quality at many beaches, due to wastewater and storm water runoff. Recent research has demonstrated that changes in the microbiome can leave the host susceptible to infection, and influence disease states. Exposure to these waters can cause gastrointestinal and respiratory illness, ear infections, and skin infections.

The investigators sought 9 volunteers at a beach who met criteria of no sunscreen use, infrequent exposure to the ocean, no bathing within the last 12 hours, and no antibiotics during the previous six months. The researchers swabbed the participants on the back of the calf before they entered the water, and again after subjects had air dried completely following a ten-minute swim and at six and 24 hours post swim.

Before swimming, all individuals had different communities from one-another, but after swimming, they all had similar communities on their skin, which were completely different from the “before swim” communities. At six hours post swim, the microbiomes had begun to revert to their pre-swim composition, and at 24 hours, they were far along in that process.

Nielsen found one very interesting finding that *Vibrio* sp. were detected on every participant after swimming in the ocean, and air drying, a genus that includes the bacterium that causes cholera. At six hours post swim, they were still present on most of the volunteers, but by 24 hours, they were present only on one individual. He said while many *Vibrio* are not pathogenic, the fact that they recovered them on the skin after swimming demonstrates that pathogenic *Vibrio* sp. could potentially persist on the skin after swimming. The fraction of *Vibrio* sp. detected on human skin was more than 10 times greater than the fraction in the ocean water sample, suggesting a specific affinity for attachment to human skin.

Nielsen added skin is the body's first line of defense; both physically and immunologically, during exposure to

contaminated water. Recent studies have shown that human skin microbiome plays an important role in immune system function, localized and systemic diseases, and infection. A healthy microbiome protects the host from colonization and infection by opportunistic and pathogenic microbes.



Image credit: CC0 Public Domain

Source: www.phys.org

New compound allows bacterial communication to be controlled by light

Scientists from the University of Groningen have succeeded in incorporating a light-controlled switch into a molecule used by bacteria for quorum sensing a process by which bacteria communicate and subsequently control different cellular processes. With the molecule described, it is possible to either inhibit or stimulate communication. This makes it a very useful tool for further research into bacterial communication and its influence on different genetic pathways. The results were published in the journal *Chem*.

In order to respond to their environment, bacteria 'talk' to each other through a form of chemical communication called quorum sensing. The cells secrete a signal molecule and at the same time monitor its concentration. As more cells secrete the signal molecule, it can exceed a threshold concentration and activate certain genetic pathways, for example, to produce toxins or form a protective biofilm.

Light-sensitive switch

'If we would be able to influence quorum sensing, we might be able to use it to treat serious infections,' says University of Groningen organic chemist Mickel Hansen. He added, it would also be useful to investigate how quorum sensing exactly works. To do this, it would be useful to have a modulator of quorum sensing that could be externally controlled. That is why Hansen and colleagues in the synthetic organic chemistry group led by Professor Ben Feringa set out to build a light-sensitive switch

into a molecule used by bacteria as a signal for quorum sensing.

The molecule is made up of a head and a flexible carbon-based tail, connected via a β -keto-amide linker. The plan was to incorporate a switch into the tail and they had to connect the modified tail to the head via β -keto-amide linkage. However, the synthetic process to obtain this linkage produced a very unstable intermediate, which made it almost impossible to synthesize the molecule.

Library

Building on the extensive experience of the synthetic organic chemistry group at the Stratingh Institute of Chemistry at the University of Groningen, the researchers came up with a solution in the form of a new coupling reaction with a stabilized intermediate. Using this intermediate, they were able to synthesize photoswitchable derivatives in a fast and straightforward way.

Hansen, together with Master's student Jacques Hille, produced a 'library' of 16 different compounds that had the potential to act as agonists or antagonists of quorum sensing. All were fitted with a light-operated switch. All compounds were based on a molecule that is used in one particular quorum sensing system in *Pseudomonas aeruginosa*, which has about five of these quorum sensing systems. In collaboration with molecular biologists from the lab of Professor of Molecular Microbiology Arnold Driessen, also at the University of Groningen, the genes for one of these systems were transferred to an *E. coli* reporter strain, allowing any effect of the newly synthesized compounds to be tested without the interference of other quorum sensing mechanisms.

Toxin production

Bioactivity tests on the compounds obtained showed which parts of the molecule were crucial to controlling quorum sensing. The optimum number of carbon atoms making up the tail appeared to be four. Flipping the switch with light caused the tail to bend. Remarkably, the straight tail had no effect, whereas the bent tail induced the quorum sensing signal. Hansen said 'Overall, it appears that small changes in the molecule can have a large effect on its activity, but we don't yet know exactly why.'

They did find one compound that was able to strongly inhibit the quorum sensing signal and after irradiation with light, leading to the bending of the tail to also strongly

stimulate it. The difference in activity was more than 700-fold, which is huge. Such a large difference to their knowledge, never been shown before for light-switched bioactive molecules. This particular molecule will be a very useful tool for investigating how bacteria communicate. In the study, they showed that they could light-control toxin production in a *Pseudomonas* strain with their switchable modulator. This will be a powerful tool for both clinical and fundamental research into the mechanism of quorum sensing.

Source: www.sciencedaily.com

NEWS

Researchers reveal how bacteria can adapt to resist treatment by antibiotics

New research shows that bacteria produce a specific stress molecule, divide more slowly, and thus save energy when they are exposed to antibiotics. The new knowledge is expected to form the basis for development of a new type of antibiotics.

In a joint collaboration, researchers from Denmark and Switzerland have shown that bacteria produce a specific stress molecule, divide more slowly, and thus save energy when they are exposed to antibiotics. The new knowledge is expected to form the basis for development of a new type of antibiotics.

All free-living organisms are under constant pressure to survive. Darwin dubbed this “survival of the fittest” and thus described how the best adapted species would have most offspring and therefore eventually end up propagating itself.

This fundamental principle is particularly prominent in the world of microorganisms, where free-living bacteria live in a constant fight to be the most well adapted and thus those who divide fastest in any given natural habitat. But when bacteria at the same time are exposed to deadly antibiotics, this fight becomes a question of balancing fitness, i.e. the ability to divide fast, with tolerance towards antibiotics. This amazing adaptability of bacteria is a contributing factor to the severity of infectious diseases in humans, including tuberculosis and severe urinary tract infection, for which the disease often resurfaces after treatment has ended.

In a new research paper, just published in the high-impact journal *Molecular Cell*, researchers from Aarhus

University have collaborated with experts from the University of Copenhagen and the technical university ETH Zürich in Switzerland and taken a close look at how bacteria handle this difficult balancing act. The results show that bacteria very quickly reduce their rate of cell division when exposed to antibiotics in order to maintain the highest possible tolerance, but quickly start growing again when the substances are removed and fitness is the most important factor.

Bacteria save up energy

At the molecular level, the researchers in the group of Asc. Prof. Ditlev Egeskov Brodersen from the Department of Molecular Biology and Genetics at Aarhus University have been able to show that the effect is mediated by an enzyme within the bacteria, capable of saving up molecular energy in the form of constituents of cellular DNA, which can be used for rapid regrowth when the antibiotic treatment is ceased. When the bacteria are exposed to antibiotics, they immediately start breaking down substituents of DNA (the so-called nucleotides), into smaller parts that are then stored in the cell.

The researchers have shown that bacteria produce a specific stress molecule called (p)ppGpp upon exposure to antibiotics that makes the enzyme more active. This thus means that the saving up of energy happens extra fast when the bacteria are exposed to stress. Using an advanced analytical method called x-ray crystallography, the researchers have been able to generate detailed 3D pictures of the enzyme, both in its normal state and when bound to the stress molecule. The results surprisingly show that the enzyme opens up when the stress hormone is present, and thus functions much more efficiently because the nucleotides can more easily access the active site where the breakdown process takes place.

It is expected that the new knowledge about the molecular basis for the reaction of bacteria to antibiotics can be used to develop a whole new type of antibiotics that prevent bacteria from saving up energy and thus adapt to the treatment.

Source: www.sciencedaily.com

Natural environments favor 'good' bacteria

A new study has shown that restoring environments to include a wider range of species can promote ‘good’ bacteria over ‘bad’ with potential benefits for human health.

University of Adelaide researchers report, in the journal *Environment International*, that degraded, low biodiversity land and soils tend to harbour more 'opportunistic' bacteria, while healthy, biodiverse ecosystems favour more stable and specialist bacteria.

They found that the bacterial communities more commonly found in degraded landscapes had "potential pathogenic character," with many in the same genera as prominent disease-causing bacteria *Bacillus*, *Clostridium*, *Enterobacter*, *Legionella* and *Pseudomonas*.

Restoring a more biodiverse ecosystem, however, changed the bacterial composition towards more potentially immune-boosting microbial diversity.

"There is a growing body of evidence associating human health with green space around people's homes, and environmental microbes provide a likely connection between a healthy ecosystem and human health," says lead author, Craig Liddicoat, PhD candidate with the University of Adelaide's School of Biological Sciences and Environment Institute.

However evidence of tangible mechanistic links between human and environmental health is still lacking, although we know that soil microbes are an important part of airborne microbial communities generated from a particular environment.

"We wanted to see if there were particular bacteria that might be representative of ecological restoration, or the change from degraded state to a more biodiverse, natural state."

The researchers analysed soil bacterial communities from a restoration site with a progression of environments from cleared, degraded land to a restored, more biodiverse, natural reference ecosystem. They compared their findings with data from over 200 samples from across Australia which had been assigned as disturbed or natural soils, and found consistent patterns in the proportions of opportunistic versus stable bacteria.

"What this work has shown is the potential for reducing airborne pathogens around us by restoring city environments to be more biodiverse and natural," says senior author Professor Phil Weinstein, Professorial Research Fellow with the School of Biological Sciences.

"While we have not shown a direct link between increased abundance of these particular bacteria with increased human

disease, more opportunistic bacteria in the environment is likely to bring more exposure to airborne pathogens and higher rates of infections in susceptible individuals. In healthy, biodiverse ecosystems this risk is reduced."

The researchers say their study also points to a new way of measuring soil and ecosystem health using groups of bacteria as summary biological indicators.

Source: www.sciencedaily.com

Abstract of Recent Publication

01. Acta Ecologica Sinica, 2019, Vol. 39 (4), Pages: 328 - 333. Evaluating soil biochemical/microbial indices as ecological indicators of different land use/cover in northern Iran. Negar Moghimian, Seyed Mohsen Hosseini, Yahya Kooch, Behrouz Zarei Darki. *Faculty of Natural Resources & Marine Sciences, Tarbiat Modares University, 46417-76489 Noor, Mazandaran, Iran.*

Microbial community structure plays a significant role in the efficiency of dark fermentative hydrogen production using mixed culture. However, the detailed evolutions in microbial community structure during dark fermentation process are still unclear. This study investigated the detailed evolution patterns of microbial community structure during dark fermentation process by high-throughput pyrosequencing. Results showed that microbial community structure changed significantly over time in dark fermentation. Microbial diversity showed a constant decreasing trend during the fermentation process. The analysis of microbial community composition showed that *Clostridium sensu stricto* 1, *Paraclostridium*, *Romboutsia* and *Paeniclostridium*, which were all rarely existed in the inoculum, dramatically became dominant genera in the system after 6 h fermentation, with total relative abundance of more than 99%. This interesting result revealed that how quickly hydrogen-producing genera overwhelmed the microbial community in dark fermentation. Spearman correlation analysis showed that *Clostridium sensu stricto* 1 contributed the most to hydrogen fermentation performances.

Keywords: Biohydrogen production; Dark fermentation; High-throughput pyrosequencing; Microbial community; Evolution pattern.

NATIONAL

Institute of Microbial Technology
<https://www.imtech.res.in>

National Centre for Biological Sciences
<https://www.ncbs.res.in>

Central Electrochemical Research Institute
<https://www.cecri.res.in>

Indian Institute of Soil Sciences
<http://www.iiss.nic.in>

INTERNATIONAL

Biotechnology Innovation Organization
<https://www.bio.org>

Society for General Microbiology
<http://www.socgenmicrobiol.org.uk>

Italian Society of Microbiology
<https://fems-microbiology.org/member-societies/societa-italiana-di-microbiologia/>

Center for Microbial Ecology
<http://www.cme.msu.edu>

EVENTS Conferences / Seminars / Meetings 2019

Advances in Microbiology and Public Health. August 19 - 20, 2019 . **Venue:** Mercure Hotel Wien Westbahnhof Vienna, **Austria.** **Website:** <https://microbiology.euroscicon.com>

CRISPR and beyond: perturbations at scale to understand genomes. September 02 - 04, 2019. **Venue:** Wellcome Genome Campus, **UK.** **Website:** <https://coursesandconferences.wellcomegenomecampus.org/our-events/crispr-and-beyond-2019/>

Plant Genomes in a Changing Environment. October 16 - 18, 2019. **Venue:** Wellcome Genome Campus, **UK.** **Website:** <https://coursesandconferences.wellcomegenomecampus.org/our-events/plant-genomes-2019/>

Annual Biomedical Research Conference For Minority Students (ABRCMS). November 13 - 16, 2019. **Venue:** Anaheim, CA, **USA.** **Website:** <http://www.abrcms.org>

International Day for Biological Diversity – 22nd May, 2019



World Environment Day - 5th June, 2019

