Water quality in urban slums of Andhra Pradesh- Pilot Study of Attagutta

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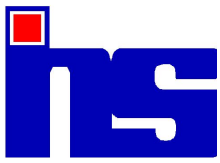

THE INSTITUTE OF HEALTH SYSTEMS

Water quality in urban slums of Andhra Pradesh- Pilot Study of Attagutta

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Water Quality in Urban Slums: Pilot study in Addagutta, Hyderabad

I. Background:

The quality of drinking water is a vital element of public health and well-being. Poor quality drinking water and inadequate sanitation are among the world's major preventable causes of early mortality. Contaminated water is an important cause of diarrhoeal diseases which is responsible for about 19% of death among 'children under five' in the developing countries (WHO, 1997). According to World Health Organization estimates, diarrhoeal diseases kill around 2.5 million people globally each year (WHO, 1997). The provision of an adequate supply of safe water was one of the eight components of primary health care identified by the International Conference on Primary Health Care in Alma-Ata in 1978 (WHO, 1978). In most countries the principal risks to human health associated with consumption of polluted water are microbiological in nature. It is therefore imperative that quality of water supplied for drinking purposes be continuously monitored, at the minimum, for indicators of faecal pollution, turbidity, and residual chlorine (if water is disinfected with chlorine) (WHO, 1997).

Epidemics of waterborne diseases are more common in slum areas. Poverty, poor sanitation, lack of sufficient and good quality drinking water, malnutrition, crowded living, lack of access to health care, poor hygienic practices etc., contribute to perpetuation of waterborne diseases in this area. Ensuring adequate and safe supply of water in slum areas, therefore plays a crucial role in interrupting this vicious cycle of waterborne disease epidemics. A key preventive measure is therefore to periodically check water quality and conduct sanitary surveys in slum areas.

In Hyderabad, The Hyderabad Metropolitan Water Supply and Sewerage Board (HMWSSB), provides water supply that caters to the drinking water needs of about 55.33 lakh persons, including those living in about 800 slums. The HMWSSB has in-house testing facilities. Other additional facilities in Hyderabad include the Institute of Preventive Medicine (IPM), Rural Water Supply (RWS) as well as a few private laboratories. However, existing mechanisms for drinking water quality testing may not be enough to meet the challenge of such a large city. Although laboratories in the public sector are open to people, they are not fully utilized due to various factors such as non-availability of well designed water sample collection kit, lack of awareness, accessibility etc. One alternative to improve water quality monitoring in the city, would be to expand the water testing facilities in the public sector. This is not likely to be cost effective considering past experience. The establishment cost per sample collected

through these mechanisms is usually very high. Hence it would be desirable to explore the possibility of a Public Private Partnership (P3) model to monitor water quality in the city. As a first step, the Board is exploring the feasibility of assessing water quality and associated risks in high priority areas such as slums.

II. Objectives of the Study:

The main objectives of the study are:-

1. To expand water testing capacity in Hyderabad and to supplement testing done by the HMWSSB in high priority areas such as slums
2. To identify risks associated with spread of waterborne diseases in slum areas of Hyderabad
3. To systematize drinking water quality monitoring based on a Public Private Partnership model and to develop the required data and experience for planning and implementation of similar projects

III.Partnering Agencies

The Hyderabad Metropolitan Water Supply and Sewerage Board and the Institute of Health Systems will be the key partners representing the public and private side of the partnership, respectively.

A. The Hyderabad Metropolitan Water Supply and Sewerage Board (HMWSSB)

Established in 1989, the HMWSSB aims to provide citizens of Hyderabad with a regular supply of drinking water of the highest quality at an affordable cost. The Board is supplying and maintaining supply of potable water and sewerage system in the twin cities of Hyderabad and Secunderabad covering MCH limits; Cantonment area, Industrial areas and surrounding Municipalities. The coverage area of the Board includes about 800 slums. In order to provide clean, potable water, the HMWSSB has adopted a mechanism of both internal and external monitoring of the distribution network. Internal monitoring is carried out within the Board by the Board's staff. Where as the external monitoring is done in correlation with outside agencies like Institute of Preventive Medicine (I.P.M). Currently the HMWSSB daily check the residual chlorine in the service reservoirs and the distribution system. Twenty (20) sample takers regularly monitor the water quality in the allotted 20 zones in Hyderabad and Secunderabad. Daily the sample takers and the field staff of the O&M Divisions check a total of 1200-1500

points in the distribution network. About 300-400 points are examined daily by QAT wing. Samples collected for bacteriological quality assessment are analysed in the QAT labs and also at IPM. Besides piped water supply, borewells are also assessed for bacteriological quality and remedial measures suggested wherever pollution is detected.

B. The Institute of Health Systems

The Institute of Health Systems (IHS) Hyderabad, established in 1990 is a premier public health research and training institution. The IHS mission is to generate knowledge, gather evidence and groom skills to improve the efficiency, quality and equity of health system. The Institute strives to build local capacity and global knowledge base for public health and socioeconomic development. IHS activities fall into (a) research and consultancy, (b) training (c) health informatics and (d) public services. The Institute is equipped with a core group of multidisciplinary and interdisciplinary full time faculty. These include people from various backgrounds including Community Health, Nutrition, Health Economics, Social Sciences, Health Care Management, Environmental Sciences, Laboratory Sciences, Biotechnology, and Computer Applications.

In pursuance of its public service, teaching and research goals, the Institute has set up a Public Health Laboratory. It is well equipped for testing potability of water. A key innovation by the laboratory is the design of Water Sample Collection Kit. Pre-sterilized bottles are packed in polythene bags along with Water Sample Collection guides, Sample Collection Record, and a carry bag to easily transport the sample to the laboratory. These bottles will be available from the IHS Front Office, round-the-clock. A catalogue of services giving details about various tests of water quality has been developed to help consumers decide on the test they need the most in a given situation. A consumers guide to collection of water samples from eateries, shops and establishments has been developed. Another guideline for collection of samples by households is also available

IV.Site Description:

The metro water board authorities have given permission to IHS for conducting a pilot study on water quality in urban slum of Addagutta. The site was selected as survey site as there was a death in a dwelling near to section -C of Addagutta in the recent past and the media attributed it to the gastroenteritis. The Addagutta slum is under 10th ward, Block IV of Municipal Corporation of Hyderabad (MCH). It is divided into five sections viz., A, B, C, D

and Vadderbasthi. There are about 5,000 dwellings with about 35,000 residents in the slum. The main source of drinking water in Addagutta slum is the water supplied by HMWSSB which can be categorized as under:-

i. Public Stand Posts or PSPs
ii. Pit Taps or PTs
iii. House Taps or HTs and
iv. Metro Water Tanker or MWT.

All the residents are supplied water either through PSPs, PTs or HTs, except the people residing in huts adjoining to Section- C, for whom the water is supplied trough water tanker. There are, twenty seven PSPs out of which three are dismantled, nine PTs out of which one is dismantled and about 2,400 HTs. Water supply is provided on alternate days in each section at fixed timings, usually for about 3 hours. The following table shows the numbers of taps that are present and functional in the Addagutta area at the time of survey.

Table 1 : No. of Functional Taps in Addagutta**.**

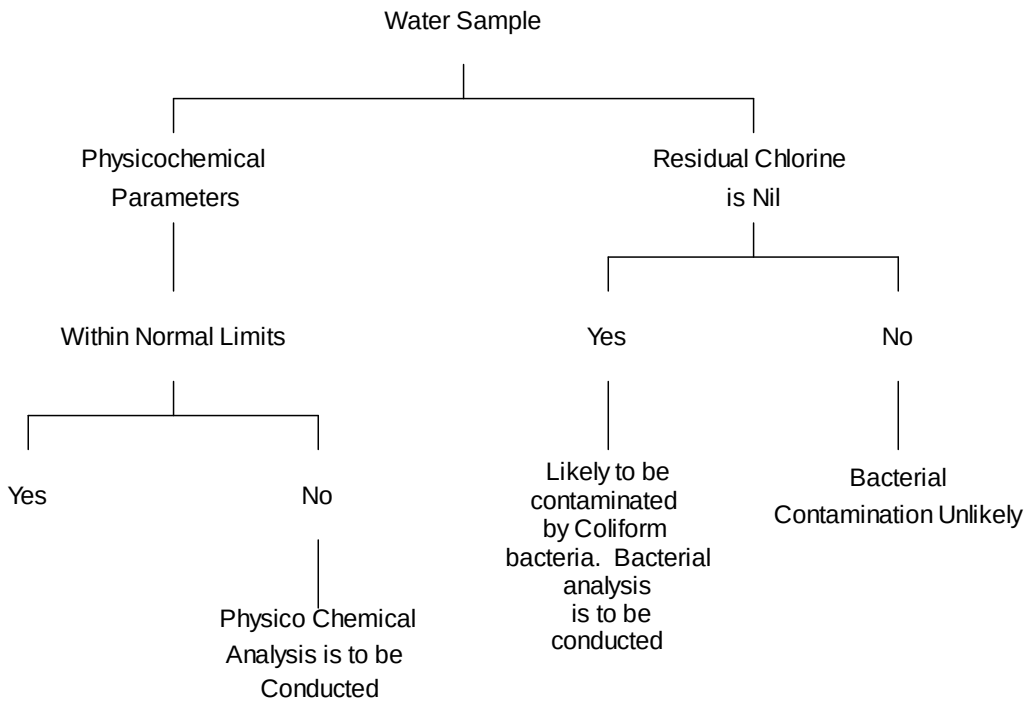
Tap type	Total No of Taps	Dismantled	Functional Taps
PSPs	27	3	24
PTs	9	1	8
HTs	2,400	NA	2,400
Total	2,436	4	2,432

V. Methodology

A. Sample collection for water quality analysis

Water samples are collected as per the algorithm in Figure -1. The quality of water is tested based on three parameters, (a) Physical (b) Chemical and (c) Bacteriological. All samples were analyzed by the IHS Water Quality Testing Laboratory (WQT Lab). Samples for Laboratory testing were collected in IHS Water Sample Collection Bottles.

Fig: 1: Algorithm followed for conducting water analysis



Physicochemical Analysis:

The physicochemical analysis will be conducted at the WQT Lab, if the general appearance, odour and turbidity of the water sample is found to be objectionable.

As the physical and chemical parameters do not vary from tap to tap, two water samples were collected; one at random from a Public Stand Post (PSP) and the other from a House Tap (HT) where water appearance was found to be objectionable . Details of all the tests conducted for physical and chemical parameters are enclosed in as Annexure -I.

Bacteriological Analysis:

The water samples for bacteriological analysis were collected directly from the tap (Direct) or from the stored water (Stored), if the supply was not present on the day of the visit, for the survey. [Stored water means the water stored by the household either in vessels, pots or plastic buckets.]

The prerequisite for conducting the bacteriological analysis is residual chlorine (RC) test. If the residual chlorine levels are within normal limits (0.2 - 0.5 PPM), the water sample is unlikely to be contaminated with bacteria and hence considered bacteriologically satisfactory for

drinking purpose. If the residual chlorine levels are less than normal limits, the water sample is likely to be bacteriologically contaminated and hence bacteriological analysis is done. Details of all the tests conducted for bacteriological analysis are enclosed in as Annexure-II.

For conducting the residual chlorine tests water samples were collected from (a) all public stand posts (b) all pit taps (c) house taps of first, middle and last house in each street (d) if the selected public stand posts and pit taps did not have water supply on the day of survey, a stored water sample was collected.

B. Sanitary Survey of Taps:

A sanitary survey is an 'on-site inspection and evaluation' by qualified individuals of all conditions, devices and practices in the water supply system that pose an actual or potential danger to the health and well-being of the consumer.

The sanitary survey form (Annexure 1II) was designed by taking ready reference from 'A Manual on Control of Gastroenteritis With Special Reference to Andhra Pradesh', India (Mahapatra & Samatha, 2001), which was adopted from Guidelines for Drinking Water Quality (WHO, 1997). The sanitary survey was conducted for the same PSPs, PTs and HTs that were selected for the water quality testing. The assessment of risk was done based on a score for each question. If the risk was present, score of 1 and if the risk was absent a score of 0 was assigned.

VI. Results

A. Test for Residual Chlorine

The following table shows the number of samples collected for RC test.

Table 2. Distribution of Samples Collected

Tap Type	Functional Taps	No. Of samples collected		
		Direct Samples	Stored samples	**Total**
PSPs	24	24	5*	**29**
PTs	8	8	3*	**11**
HTs	2,400	82	2	**84**
MWT	NA	1	2	**3**
ColnTotal	2,432	115	12	**127**

* Both direct and stored samples were collected from five PSPs and three Pts on two different days.

Total of 127 samples were conducted for residual chlorine tests, out of which 29 were from PSPs, eleven were from PTs and 84 were from HTs and three from MWTs. All the

residual chlorine tests were done at the site. The following table shows the number of residual nil water samples in Addagutta area.

Table- 3. Results of Residual Chlorine Test

Direct/ Stored	Source					Residual Chlorine Nil Samples				
	PSPs	PTs	HTs	MWT	**Total**	PSPs	PTs	HTs	MWT	**Total**
Direct	24	8	82	1	**115**	0	0	3	0	**3**
Stored	5	3	2	2	**12**	5	1	0	2	**8**
Total	29	11	84	3	**127**	5	1	3	2	**11**

The water samples with nil RC were carried to IHS WQT Lab using the pre-sterilized water collection bottles within 6 hrs. from collection time. A total of 11 samples were having nil RC and hence collected for bacteriological analysis.

B. Physico Chemical Analysis

Out of the two selected samples for physicochemical analysis, the sample collected from PSP was found to be satisfactory for drinking purpose. The other sample collected from HT was aesthetically unsatisfactory. It was muddy coloured and turbidity levels were 28 NTU as against the permissible limits of 10 NTU of maximum.

Chemical analysis of both these samples showed the results to be within normal limits the physico-chemical analysis reports of the two samples is enclosed with this report..

C. Bacteriological Analysis

The 11 samples with nil residual chlorine were analyzed for bacterial presence. Out of this, eight were stored samples (including two metro water tanker samples) and three were direct metro water tap samples. The following table shows the number of samples collected from different sources and number of samples positive for bacteria from each source.

Table -4: Distribution of Bacteriologically Contaminated Samples

Direct / Stored	Source of samples with Nil RC					Bacteria Positive				
	PSPs	PTs	HTs	MWT	**Total**	PSPs	PTs	HTs	MWT	**Total**
Direct	0	0	3	0	**3**	0	0	3	0	**3**
Stored	5	1	0	2	**8**	5	1	0	2	**8**
Total	5	1	3	2	**11**	5	1	3	2	**11**

Three direct metro water samples collected from 3 different house taps (HT-5, HT-36 and HT-45) were found positive for bacteria. Eight stored metro water samples consisting of

five collected from Public Stand Posts (PSP-14, PSP-16, PSP-6, PSP-8, PSP-9), one sample from Pit tap (PT-4) and two from Metro Water Tanker stored in Huts opposite to "C" Section (MWT-1, MWT-2) were found positive for bacteria. Reports of each of the 11 samples are enclosed.

D. Sanitation Survey

As there were eight items in the survey form, the maximum possible risk score was eight while the minimum could be zero for any PSP/PT/HT. The risk scores were classified into four categories as under:-

i. *If the score is greater than or equal to six - Very High Risk*
ii. *If the score is between 4 and 5 - High Risk*
iii. *If the score is between 2 and 3 - Intermediate Risk*
iv. *If the score is 1 - Low Risk.*

The risk score indicates the likelihood of contamination of water, especially during monsoons and epidemics. The following table shows the distribution of risk categories for PSPs, PTs and HTs, in Addagutta area.

TABLE-5 : Distribution of Risk Categories for PSPs, PTs and HTs in Addagutta

Risk category	Score	PSPs	%	PTs	%	HTs	%
Very High	>= 6	3	11.11	3	37.5	4	4.76
High	4 - 5	4	14.81	2	25	6	7.14
Intermediate	2 - 3	8	29.63	0	0	6	7.14
Low	1	9	33.33	3	37.5	68	80.95
Total		24	100	8	100	84	100

About 26%, 55% and 12% of the PSPs, PTs, and HTs is under the high risk category for water contamination.

VII. Analysis and Recommendations:

1. Overall, water supplied by the Metro Board to Attagutta slum is physically, chemically and bacteriologically satisfactory for drinking purpose. A total of 127 water samples were tested, of which 112 were direct samples from source of water supply and 12 were stored samples. All direct samples except 3 taken from house taps had adequate chlorination. All the 3 samples were found to be bacteriologically contaminated. The house taps may be distant endpoints in the distribution system. Therefore necessary

steps may be taken to ensure that adequate residual chlorine levels is maintained through out the distribution system.

2. Of the 12 stored water samples taken, 8 were found to be bacteriologically unsatisfactory for consumption. This indicate that though water supplied at a PSP or PT may be micro biologically safe, it may become contaminated due to unhygienic transportation, storage and/or practices.

3. Though water supplied through PSPs and PTs was found to be microbiologically safe for consumption, there exists a risk of contamination especially during monsoons and epidemics. About 62% of the PTs are in the high risk category. Pit taps are more vulnerable to contamination than others as the taps were not surrounded by cement compound and there was water stagnation around the taps and open defecation near the PTs. 26% of the PSPs are under high risk category due to presence of drainage pipes near them, open defecation with in 30 feet and leakage of taps.

4. 12% of the HTs are in the high risk category, mainly due to the direct tapping of water from the metro water pipe line through plastic pipes, without valve. And these are near to open defecation and drainage lines.

5. Adequate attention to minimizing these risk factors, by the Board and other public health authorities can reduce risk of future contamination of water. High level of contamination in stored water samples make it evident that hygienic behaviour and practices play a key role in drinking water quality.

6. It may be noted that the study was carried out with the knowledge and help of Board personnel in the area. There exists a possibility that extra attention was paid to ensure high levels of chlorination during the field study. This may have altered the normal water quality scenario in this area and affected the findings of the study.

7. The methodology, scope and findings of the study may be assessed and possibility of undertaking similar studies in urban slums of Hyderabad may be explored

VIII.References:

i. Guidelines for drinking- water quality, Surveillance and control of community supplies, Second edition, Vol.3, WHO1997.

ii. Guidelines for drinking- water quality, Health criteria and other supporting information, Second edition, Vol.2,WHO1997.

iii. Guidelines for drinking- water quality, Recommendations, Second edition, Vol.1, WHO1997

iv. Standard Methods for the Examination of Water & Waste Water, 18th edition,1992, Arnold E. Grenberg, Lenore S. Clesceri, Andrew D. Eaton.

Annexure - I

I. Physical parameters:

A. Colour:

Colour is measured by visual comparison of the sample with Platinum Cobalt Standards. Standards having 5,10,15,20,25,50 Hazen units. Standards are prepared by diluting 0.5, 1.0,1.5, 2.0, 2.5 & 5 ml respectively of standard Chloroplatinate solution and make up to 50 ml with distilled water. 50 ml of test sample is taken in Nessler's tube and compared with standards. Maximum permissible limits for colour in the absence of alternate source is 25 hazen units. If colour is morethan maximum permissible limits it is Aesthetically not acceptable.

B. Turbidity:

Turbidity is measured by Nephelometric method. Turbidity measurement is based on the comparison of the intensity of light scattered by the sample under defined conditions with the intensity of light scattered by standard reference suspension under the same conditions. Turbidity is measured in Nephelometric Turbidity Units (NTU). First the Turbditimeter is set with Standard Hexamethelene Tetramine solution whose turbdity is 40 NTU, then the sample is taken and its turbidity is measured in turbidimeter. Maximum permissible limits for turbidity in the absence of alternate source is 10 NTU. If turbidity is morethan maximum permissible limits it is objectionable from the point appearance.

C. Odour:

Odour like taste, depends on contact of a stimulating substance with the appropriate human receptor cell. Odour is determined by Threshold odor test. If the sample is with odour than it is aesthetically not acceptable.

D. Conductivity:

Conductivity is a measure of the ability of an aqueous solution to carry an electric current. The Electrical conductivity provides useful estimate of the dissolved solids in water. Electrical conductivity is the measure of the mineral constituents dissolved in water. The electrical conductivity determination of sample is made by using a conductivity cell with platinum electrodes in conjunction with a Wheatstone bridge. Maximum permissible limits for conductivity in the absence of alternated source is 1500 μSiemens.

E. P^H:

P^H value is the logarithm of reciprocal of Hydrogen ion activity in moles per litre. pH of the sample is measured by P^H meter. Procedure includes standardization of the pH meter by using buffer solution of known P^H , adjust the temperature correction and measure the pH of the sample. Normal range for P^H is 6.5 to 8.5. Beyond the normal limits the water will affect the mucus membrane.

I. Chemical parameters:

A. Alkalinity:

Alkalinity is determined by titration with standard solution of strong acid to certain end point as given by the indicator solution. Phenolthalein is satisfactory for the first point contributed by hydroxide and carbonate. Methyl orange is used for the second point contributed by the bicarbonates. 50 ml of sample is taken, add two drops of Phenolthalein indicator, if pink colour appears titrate with N/50 H_2So_4 till colour disappears. Note the reading. Add two drops of Methyl orange indicator, titrate against acid till the yellow colour turns to light orange. Take down the final reading. Maximum permissible limits for alkalinity in the absence of alternated source is 600 mg/litre.

B. Total Hardness:

It is a titrimetric method based on reaction of Calcium and Magnesium salts with Ethelene diamine tetra acetic acid. Take 25 ml of sample make upto 50ml with distilled water, add 4 drops of Eriochrome Black T indicator solution, add 1ml of ammonia buffer, titrate immediately with EDTA till a blue colour is obtained. Note the final reading. Maximum permissible limits for Total Hardness in the absence of alternate source is 600 mg/litre. Beyond these limits the water leads to cardiovascular disease.

C. Calcium:

Take 25ml of sample dilute to 100ml. Add 2ml of 1N NaoH solution and sufficient Murexide powder to produce marked colour. Titrate against EDTA till a purple colour is obtained. Note the final reading. Maximum permissible limits for Calcium in the absence of alternated source is 200 mg/litre.

D. Magnesium:

Amount of Magnesium present in the sample = Total hardness- Calcium levels gives amount of Magnesium in mg/litre. Maximum permissible limits for Magnesium in the absence of

alternate source is 100 mg/litre. Beyond the maximum permissible limits Magnesium in combination with Sulphates causes laxative effect.

E. Chlorides:

Take 100 ml of sample or aliquot sample diluted to 100ml. Add 1ml of Potassium Chromate indicator. Titrate against Standard Silver Nitrate solution till brown colour appears and persists. Note the reading .Titrate 100 ml of distilled water in the same manner as Blank. Note the reading. Maximum permissible limits for Chloride in the absence of alternate source is 1500 mg/litre. Beyond this limit Water is salty in taste and affects palatability.

F. Ammonia:

The sample is buffered and distilled. The ammonia in the distillate or in the sample is treated with Nesslers reagent and the colour developed is matched with that of standard Ammonia solution. Take 50 ml of sample and add a pinch of Rochelle salt. Add 1 ml of Nesslers reagent. If yellow colour appears compare the colour with standards of Ammonia. In drinking water ammonia should not be present. Presence of ammonia in water promotes the growth of organisms, growth of algae.

G. Nitrite:

Nitrite is determined through formation of a reddish purple azo dye produced at P^H 2 to 2.5 by coupling diazotised sulphanilic acid with N-(Napthyl)-Ethyelene diamine dihydrochloride. Take 50ml of sample, add 1ml of Sulphanillic acid solution and mix thoroughly and then add 1ml of Alpha Napthol amine hydrochloride solution. Mix well and allow to stand for 10mts. Compare the colour with the standards obtained in the same way. Nitrite should not be present in drinking water. Presence of Nitrite in water leads to formation of a compound named Nitrosamine which is Carcinogenic.

H. Nitrate:

Evaporate 50 ml of sample in a water bath. Dissolve the residue in 2 ml Phenol Di Sulphonic Acid solution. Rub thoroughly with glass rod to dissolve the residue. Add 20ml distilled water and add KOH solution till an yellow colour develops. Make up to 50ml with distilled water. Compare the colour with Standards prepared simultaneously. Maximum permissible limits for Nitrate in the absence of alternated source is 10 mg/litre as N. Beyond these limits leads to Infantile Methemoglobinemia in children and irritation of mucous membrane in adults.

I. Sulphate:

Take 20 ml of the sample. Add 1ml of 1:9 Hcl and 1ml of Conditioning reagent. Mix well for 30 seconds. Prepare 1ml , 2ml, and 8ml of standards and make upto 20 ml with distilled water. Add 1ml of 1:9 Hcl and 1ml of Conditioning reagent. Mix well for 30 seconds. Standardize the turbidimeter with Standards and not down the NTU for sample. Maximum permissible limits for Sulphates in the absence of alternate source is 400 mg/litre. Beyond these limits the water leads to gastrointestinal initiation, laxative effect. Anaerobic condition releases hydrogen sulphide.

J. Flouride:

Take 50ml or aliquot sample and make upto 100ml in a 100ml Nesslers tube. Take standard solutions of Fluoride by diluting 2, 4, 6, 8, 10 and 12ml of Standard NaF solution in a Nesslers tube and make upto 100 ml with distilled water. Mix well and add 5ml of Acid reagent with a bulb pipette. Mix well and stand exactly for one hour and take the readings by comparing colors with that of standards under a white surface. Maximum permissible limits for Fluoride in the absence of alternate source is 1.5 mg /l itre. Excess Fluoride levels leads to Dental caries in children and Skeletal fluorosis in adults.

Annexure - II

Bacteriological Analysis

A. Residual Chlorine:

The RC test is conducted to know the presence of residual chlorine levels in the water sample and this is a spontaneous test. The R.C Levels are checked immediately by adding a drop of Orthotouledene to the samples. If the sample is found Nil for R.C, the sample will be collected in pre- sterilized bottle after filling the particulars provided on the bottle (Sample collection date, brief identification of the source) and will be carried for other tests

B. Microscopic Examination:

This test is done to identify the presence of microorganisms (bacteria, algae, fungi or protozoan parasites) in a given sample. A drop of sample is taken on a clean glass slide and observed under microscope. After examination, if the sample is clear it is reported as (**NAF**) Nothing Abnormal Found. If the sample is unclear then organism present is identified and reported.

C. Most Probable Number from serial dilution:

This test is done particularly for low concentrations, only viable organisms are enumerated by the MPN determination which reveals presence of colifrom bacteria.

For each sample to be tested

1. 3x10ml sample added to 10ml Double strength MacConkeys broth
2. 3X1ml sample added to 10ml single strength MacConkeys broth
3. 3X0.1ml sample added to 10ml single strength MacConkeys broth

This test is done in three successive steps i.e.,

1) **Presumptive Test-** Inoculation of MacConkeys broth in all the 3 sets of tubes, which are kept for incubation for 24 hours. If the test is positive there is gas production (fermentation in the tubes), MPN count is taken through Mc Crady's table. The positive tubes are incubated for another 24 hours.
2) **Confirmed Test-** If there are positive tubes after 48 hours the MPN count of the positive tubes is taken from Mc Crady's table, which gives the most probable number. The fermented tubes are taken and a drop of culture is streaked on Eosin Methylene Blue (EMB) agar (Selective media for E.coli). which is incubated for 24 hours. If green metallic sheen appears then the sample is said to be positive for E.coli, if pink color mucoid colonies are formed it is positive for some other coliform bacteria.

3) **Completed Test-** To distinguish between E.Coli and other coliforms Biochemical tests are performed, which are known as IMVIC (I-Indole, M-methyl red, V-Voges proskauer and C-Citrate) tests.

   i) From the EMB agar the colonies are selected and inoculated in peptone water, MRVP and Simmons citrate agar. Suitable reagent is added to all the tubes.

   ii) The results of MPN test are interpreted based on following Mc Cradys table.

MPN Table

3 of 10ml Each	3 of 1.0ml Each	3 of 0.1ml Each	MPN of Coliform Bacteria	95%Confidence Limit	
				Low	Upper
0	0	1	3	0.5	9
0	1	0	3	0.5	13
1	0	0	4	0.5	20
1	0	1	7	1	21
1	1	0	7	1	23
1	1	1	11	3	36
1	2	0	11	3	36
2	0	0	9	1	36
2	0	1	14	3	37
2	1	0	15	3	44
2	1	1	20	7	89
2	2	0	21	4	47
2	2	1	28	10	149
3	0	0	23	4	120
3	0	1	39	7	130
3	0	2	64	15	379
0	1	0	43	7	210
3	1	1	75	14	230
3	1	2	120	30	380
3	2	0	93	15	380
3	2	1	150	30	440
3	2	2	210	35	470
3	3	0	240	36	1,300
3	3	1	460	71	2,400
3	3	2	1,100	150	4,800
3	3	3	1,609	-	-
5	5	5	2,400	-	-

Standard Plate reading for E.coli: 1 plus =2 , 2 plus =5, 3 plus =9.

Biochemical Reactions: ***IMVIC Tests***: The organism or coliform bacteria present is identified by this test. Indole reagent -Kovac's Reagent : Cherry red ring is formed if positive; Methyl red-Methyl red indicator solution: Pink/red color precipitate is formed,if positive; Voges Proausker-Barrit's Reagent A and B: Pink color is formed if positive; Citrate- Indicator is bromothymol blue: Green color turns to blue if positive.

Name of the Organism	Indole	Methyl red	Voges proskauer	Citrate
Escherichia. coli (E.Coli I)	+	+	-	-
Escherichia coli (E.Coli II)	-	+	-	-
Citrobacter. freundii I	-	+	-	+
Citrobacter. freundii II	+	+	-	+
Klebsiella. aerogenesI	-	-	+	+
Klebsiella. aerogenesII	+	-	+	+
Irregular II	-	+	-	-
Irregular VI	-	-	+	+

D. Spread Plate Technique for Salmonella.

One ml of sample is added to selective media of Deoxycholate Citrate agar. Using spread plate technique, and incubated for 24 hours. If black colonies are observed, the sample is positive for Salmonella.

E. Standard Plate Count:

Plate counts are useful in determining the efficiency of operations for removing or destroying organism, E.g, Filtration, Chlorination. Colony counts are performed after plating aliquots of the water sample.

15 ml agar tubes are kept in boiling water until the media gets completely melt and then cooled to 45-50^{0}C. One ml sample is taken on sterile petridish and the agar from the tube is poured onto the plate containing the sample. The plate is shaken gently in so that the media and sample get mixed up thoroughly. After the media gets solidified, the plate is incubated for 24 hours and the number of colonies present are counted with colony counter.

Annexure-III

Form: WS-3

Sanitary survey of piped water supply system (PWS)

Date of visit:

Water sample taken?

I. Identification:

1. Local name and description of the source:

2. Address:

II. Assessment of risk (If present score=1, else score=0)

Sanitary survey question and source characteristic	Score	Remark
1 Is the supply intermittent?		
2 Are there any leaks in the distribution system?		
3 Is the area around the tapstand unclean?		
4 Does water accumulate near the tapstand?		
5 Are there human excreta within 30 ft of the tapstand?		
6 Is there any sewerage pipe line passing very close to the water pipe line?		
7 Is the plinth of the tapstand cracked or eroded?		
8 Does the tap leak?		
Total score for risk assessment		

III.General comments:

Name, designation and signature of surveyor

Significant persons present during the inspection:

Name & address	Designation / role	Signature