

Elimination of microbes from different drinking water sources of Visakhapatnam using potassium permanganate: dose based disinfection approach

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ABSTRACT

Potassium permanganate has been used since ages for disinfection of water. It is known for oxidation of cell membrane of micro organisms. But the pink colour prevailing after use makes people reject the water on aesthetic grounds. Water samples from bore wells and tap water from public places were collected from different areas of Visakhapatnam. Physio-chemical and microbial analysis analyses of the water samples were carried out. A dose based permanganate treatment of water depending upon number of colony forming units and species of microorganism present in the water was carried out [1,2,5].

Keywords: Potassium permanganate; Bergy's Manual; AWWA; B.I.S.

1. INTRODUCTION

Water sanitation is crucial for health therefore it is necessary to evaluate the microbial quality of drinking water. Chlorination, which is generally employed for disinfection of water leads to the formation of THM's (Tri Halo Methane) compound which include chloroform, bromoform etc. People generally use cloth filtration which removes particulate matter but does little to eradicate microbes from water.

In some places alum is used which only coagulates the suspended matter resulting in less number of bacterial colonies but complete eradication of microbial population is not achieved by any of these processes. Potassium permanganate has been used since ages for disinfection of water.

It is known for its oxidation of cell membrane of micro organisms. But the pink colour prevailing after use makes people reject the water on aesthetic grounds. Previous research show that high dose of permanganate are required for complete disinfection of water.

A dose of 20 mg/L and contact times of 24 hours were necessary to deactivate many pathogens and at that concentration the water turned an objectionable pink color.

Hazen and Sawyer (1992) [5] reported that complete removal of coliform bacteria was accomplished at doses of 1, 2, 3, 4, 5, and 6 mg/L.

Present study was conducted in different parts of Visakhapatnam city, a Northern District of Andhra Pradesh. Water samples from different bore wells in residential areas and tap water from public places were collected. Physio-chemical and microbial analysis analyses of the water samples were carried out.

The areas included in our study are inhabited by heterogeneous strata of society which lack basic awareness about water sanitation. We have made an attempt to carry out a dose based permanganate treatment of water depending upon number of colony forming units and species of microorganism present in the water.

Yet restricting the amount of Mn within the permissible limit for drinking water as prescribed by B.I.S (extended upto 0.5 mg/l).

2. MATERIALS & METHODS

Analytical grade chemicals were purchased from Merck, Himedia Laboratories Pvt. Ltd., SRL Chemical Pvt. Ltd., Qualigens Chemicals.

2. 1. Biochemical Characterization

Isolation of bacteria and their characterization was performed as per standard procedures (Bergy's Manual for Classification of Bacteria) [7-13].

2. 2. Chemical Parameters

The water samples were analyzed for Dissolved Oxygen, Residual Chlorine, Nitrite, Ammonia, Sulphides & Sulphates all the test were performed as per standard protocol described by APHA [16].

2. 3. Methodology

Bore well and tap water samples were collected in sterile PET bottles from different residential areas interspersed with slums and storage tanks in public places, respectively.

The bottles were rinsed with the sample water before collection and were then tightly sealed with a parafilm to prevent secondary contamination and were safely transported to the University's Lab for microbial characterization and other selected physicochemical parameters.

The water is then transferred to sterilized one liter glass bottles and then to each bottle Potassium Permanganate was added the dose was varied at 0.5 mg/l, 1 mg/L and 2 mg/L concentration.

The bacterial count of the water samples was carried out after a period of 24 hrs-72 hrs. The samples were screened for microbial growth up to 72 hrs. The isolates were then subjected to biochemical characterization like catalase, indole, methyl red, voges proskauer, citrate and motility test .

3. RESULTS

Table 1. Bacterial Density and the number of selected isolates from drinking water samples.

Sample	Cfu/ 50 µl before treatment with KMnO ₄	Selected isolates	Cfu/ 50 µl after treatment with KMnO ₄ Dose: (0.5 mg/l)			Cfu/ 50 µl after treatment with KMnO ₄ Dose: (1 mg/l)			Cfu/ 50 µl after treatment with KMnO ₄ Dose: (2 mg/l)		
			12 Hrs	36 Hrs	72 Hrs	12 Hrs	36 Hrs	72 Hrs	12 Hrs	36 Hrs	72 Hrs
GW1	80	2	80	80	80	60	60	40	-	-	-
TP1	200	4	200	200	190	160	160	120	30	-	-
GW2	70	2	60	60	60	50	50	40	-	-	-
TP2	46	2	40	40	40	30	26	10	-	-	-
TP3	68	3	50	50	50	40	30	20	-	-	-
GW3	100	4	100	100	100	80	68	54	-	-	-
TP4	96	2	90	96	96	78	60	40	-	-	-

* GW – Ground Water, TP - Tap Water

Analyzing a total of seven samples collected from different water sources of Visakhapatnam city yielded 19 isolates were selected for further characterization. The samples with high cfu were subjected to serial dilution (Ten fold) and then they were again subjected to biochemical characterization which allows the accurate identification of the bacterial fauna. The cfu was found out to be maximum in case of TP1 even at the KMnO₄ dosage of 0.5 mg/l and the minimal growth was observed with the dosage of 2 mg/l with the contact period of 72 hrs.

Table 2. Gram Staining.

Sample	Gram Staining
GW1	Gram Positive Cocci
TP1	Gram Negative Cocci
GW2	Gram Positive Cocci
GW2	Gram Negative Cocci
TP2 (Dilution 10 ⁻⁴)	Gram Positive Bacilli
TP-3	Gram Negative Bacilli
TP-3 (Dilution 10 ⁻⁴)	Gram Negative Bacilli

GW-3 (Dilution 10-4)	Gram Negative Bacilli
TP-4	Gram Negative Bacilli
TP-4 (Dilution 10-3)	Gram Negative Cocci
TP-4 (Dilution 10-4)	Gram Positive Bacilli
TP-4 (Dilution 10-5)	Gram Negative Cocci

* GW – Ground Water, TP - Tap Water

After screening the isolates from different water sources the samples were subjected to Gram staining to establish the shape and the type of microbial species which could possibly be present in the water samples [7,8].

Table 3. Morphological and biochemical observations of the different isolates from different water sources.

Sample	Form	Surface	Colour	Elevation	Opacity	Catalase Test	Indole Test	Methyl Red Test	Voges-Proskauer Test	Citrate Test	Motility	Microbe Identified
GW1	Circular	Smooth	Cream	Raised	Opaque	-	-	-	+	+	-	May be Klebsiella Sps., E.coli
TP1	Circular	Smooth	Yellow	Convex	Opaque	+	+	+	-	+	+	May be Streptococcus Sps., Klebsiella sps., Fugus
GW2	Circular	Shiny	White	Raised	Opaque	-	-	-	+	+	-	May be Klebsiella Sps.

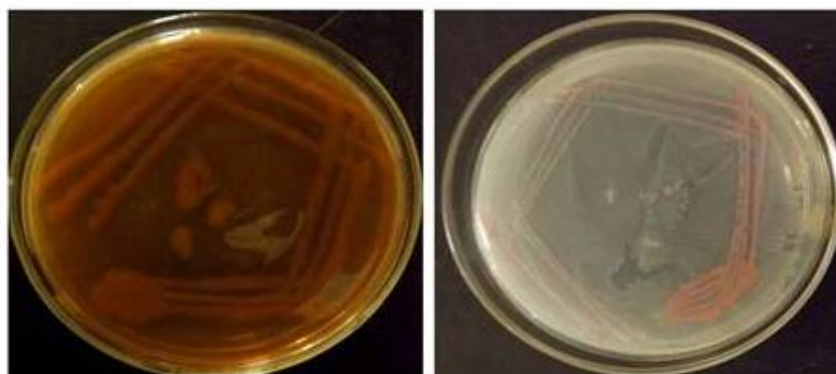
TP-4 (Dilution 10-4)	TP-4 (Dilution 10-3)	TP-4	GW-3 (Dilution 10-4)	TP-3 (Dilution 10-4)	TP-3	TP2 (Dilution 10-4)	GW2
Circular	Circular	Circular	Circular	Circular	Circular	Circular	Circular
Smooth	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth
Cream	Cream	Yellow	Yellow	Cream	Cream	Yellow	Colourless
Convex	Convex	Convex	Convex	Convex	Convex	Raised	Raised
Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque
'	'	'	'	'	'	'	'
+	+	+	+	+	+	'	'
+	'	'	+	+	+	'	+
'	'	'	'	'	'	'	+
'	'	'	'	'	+	+	+
'	'	'	'	+	+	'	'
May be Klebsiella Sps., E.coli, Streptococcus Sps.	May be Klebsiella Sps.	May be Klebsiella Sps.	May be Klebsiella Sps., E.Coli.	May be Klebsiella Sps., E.Coli.	May be Streptococcus Sps., Klebsiella sps., E.Coli.	May be Klebsiella Sps.	May be Streptococcus Sps., Klebsiella sps.

TP-4 (Dilution 10-5)	Circular	Smooth	Cream	Convex	Opaque	-	+	+	-	-	-	May be Klebsiella Sps.
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* GW – Ground Water, TP - Tap Water

After Gram staining was performed the samples were then subjected to various biochemical test and were scrutinized and thereby finding the genus of the microbes present in TP2 sample the growth of fungus was established by plating the selected isolate in Potato Dextrose Agar along with Rifamycin (30 mg/l) to eliminate the bacterial growth in the media with the above biochemical characterization the microbes that are identified are E.Coli, Streptococcus Sps., Klebsiella Sps., Fugus.

The presence of E.coli is confirmed by MPN test, Streptococcus sps., was identified by its growth on specific media Blood agar.



**Streptococcus Sps.
grown on blood agar plate.**

**Fungus grown on Potato Dextrose
Agar(PDA).**

Table 4. Physico - Chemical analysis of Water Samples.

Sample	Residual Chlorine	Dissolved Oxygen	Nitrate	Ammonia	Sulphide	Sulphate
GW1	-	-	0.03 mg/L	-	1.6 mg/l	20 mg/L
TP1	-	4.9 mg/L	0.04 mg/L	0.3 mg/L	-	0.2 mg/L
GW2	-	-	0.06 mg/L	-	-	11.6 mg/L
TP2	-	5.6 mg/L	0.03 mg/L	-	1.6 mg/l	-
TP3	-	-	-	0.5 mg/L	-	-

GW3	-	-	1.7 mg/L	0.2 mg/L	-	13 mg/L
TP4	-	4.6 mg/L	-	0.2 mg/L	-	-

* GW – Ground Water, TP - Tap Water

After biochemical characterization and physicochemical analysis of water samples was done to identify an appropriate and apt dose of Permanganate to kill the microbes and also making the water available for human consumption the dosage was set within the permissible limit of B.I.S.

4. CONCLUSION

The bacterial colonies were reduced from 200 to 30 with a minimum dose of KMnO_4 at 2 mg/l. After treatment, the amount of manganese was found to be within the permissible limit. Awareness about water sanitation among masses is poor, dose based disinfection of water, depending on the bacterial count and species present, using a common and cheap chemical like KMnO_4 can be demonstrated at local level without compromising with aesthetic value [1-4].



**Before treatment After treatment at 1 mg/l After treatment at 2mg/l dose
dose**

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