

## Revelation of Non-putrefying Property of Ganga Water

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**Abstract:** During the Water Quality Studies from Patna (25°37'N 85°24'E) to Bhagalpur (25°22'N 87°42'E), it was observed that the Dissolved Oxygen (D.O.) ranged from 7.2 to 11.5 mg/l which was exceedingly high. The B.O.D. at the outfall of sewage discharge was 5.9 mg/l which decreased after d/s 10 km to 1.9 mg/l, whereas, D.O. increased from 7.0 to 9.0 mg/l suggesting very high rate of re-aeration removing the putrefiable organic matter suggesting self-purification. As it is well known that putrefaction can take place by anaerobic bacteria, the high D.O. content of Ganga water is toxic and lethal for the anaerobic bacteria leading to non-putrefaction of Ganga water. Spectrophotometric studies have shown that the aerated Ganga water is denser than the deoxygenated water at 244 nm. Our studies reveal the cause of non-putrefaction of Ganga water for the first time based on experimental studies.

**Key words:** Putrefaction, Ganga water, Dissolved oxygen, Anaerobic bacteria

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### I. Introduction:

River water tends to putrefy when the lack of oxygen promotes the growth of anaerobic bacteria that gives water a distinct smell and stale taste. The water of Ganga though considered one of the dirtiest, does not putrefy over a long period of storage. In fact, British Physician, Nelson (2008) [1] observed that Ganga water taken from Hoogly River (one of the dirtiest) by returning ships to England remained fresh throughout the voyage. Non-putrefaction property of Ganga water was the reason East India Company used water from Ganges for drinking purposes on their 3-month long voyage to England because it stayed "Sweet & Fresh". Hindus have always believed that "Ganga Jal" to be pure, pious and drinkable. Much reverence is given to Ganga water during Hindu rituals from birth to death. Hankin (1896) [2] reported that cholera microbes (*Vibrio cholerae*) died within 3 hours in Ganga water but on the contrary, the same bacteria continued to live in distilled water even after 72 hours. However, Hankin (1896) [2] observed that Ganga water lost the disinfecting property after it had been boiled. He concluded that some volatile material was responsible for the death of the microbes. Similarly, Felix d' Herelle (1927) [3] was amazed to find no microbes at all in the Ganga water collected just below few feet the floating bodies of people who died of cholera and dysentery. The presence of bacteriophages was considered to be the reason behind this quality and purity.

Bhargava (1987) [4a] reported that the "Ganga Jal" does not putrefy even after prolonged periods of storage because of Ganga water's high content of D.O., extraordinarily high rate of reaeration, long retention abilities and very fast assimilation of putrefiable organic matter that has been discharged into the river Ganga. Ganga's self-purifying property leads to O<sub>2</sub> levels 25 times higher than any other river of the world [4b]. But Bhargava (1983) [4b] did not assign any valid reasons for non-putrefaction. In our laboratory, we have also observed that retention capacity as well as very fast assimilation of added putrefiable organic matter of the Ganga water.

Putrefaction can be termed as decomposition of proteins and eventual breakdown of cohesiveness between the tissues and liquefaction of most organs. This is caused by decomposition of the organic matter by the anaerobic bacteria which causes the release of gases that infiltrate the body's tissues which leads to deterioration of tissues and organs.

It is well known that putrefaction can only take place by anaerobic bacteria and O<sub>2</sub> is toxic and lethal for the anaerobic bacteria. This is precisely the reason why cholera microbes died in Ganga water and survived in distilled water. The anaerobic bacteria cannot tolerate O<sub>2</sub> because they utilize metabolic schemes built around enzymes that react with oxidants. The reliance upon low potential flavoproteins for anaerobic respiration probably causes substantial superoxide and hydrogen peroxide to be produced when anaerobes are exposed to dissolved oxygen.

Oxygen Toxicity:

Anaerobes lack certain enzymes that are essential for bacteria to survive in the presence of oxygen. During growth and metabolism oxygen reduction products are generated within the organisms. One oxygen reduction product is superoxide anion produced by univalent reduction of O<sub>2</sub>:  $O_2 \rightarrow O_2^-$

It is generated during the interaction of molecular oxygen with various cellular constituents like flavoproteins, quinines, thiols and iron-sulphur proteins. It causes intracellular damage by an unknown mechanism but is capable of taking part in number of destructive reactions potentially lethal to cell. Moreover, the products of the secondary reaction may amplify the toxicity by Haber- Weiss Reaction [5]. A subsequent reaction between superoxide anion and the hydroxyl radical produce singlet oxygen, is very reactive and even damages the cells also. An enzyme super oxide dismutase (SOD) is possessed by the aerobic bacteria in order to destroy the oxygen free radicals, thus preventing the destructive action of the superoxide anion.



As anaerobes lack this enzyme and as such they cannot tolerate the toxic effects of the  $O_2$  and hence do not survive in presence of  $O_2$ .

Therefore, it was considered of interest to study the role of D.O. (volatile material) which could reveal the mystery of non-putrefying property of Ganga water.

## II. Materials and Methods:

Spectral Analysis: Light absorption of dissolved oxygen and deoxygenated Ganga water were also studied to see whether Ganga water with dissolved  $O_2$  was denser than deoxygenated Ganga water or not. This was studied by UV- absorption spectra measurement at 1 nm intervals in the region of 200- 350 nm by double scanning using quartz cell in Shimadzu UV- Vis Spectrophotometer, Model- UV-1800 containing samples of normal Ganga water, saturated with oxygen Ganga water and deoxygenated Ganga water by boiling. It was observed that molecular  $O_2$  dissolved in water absorbs more light than deoxygenated sample at 244 nm making it denser (Fig. 2).

Dissolved Oxygen: Dissolved oxygen was measured by Winkler's method and BOD was measured according to the APHA Standard Methods, 2017 [6].

Total Coliform and Fecal Coliform: These were measured according to APHA Standard Methods, 2017 [6].

## III. Results:

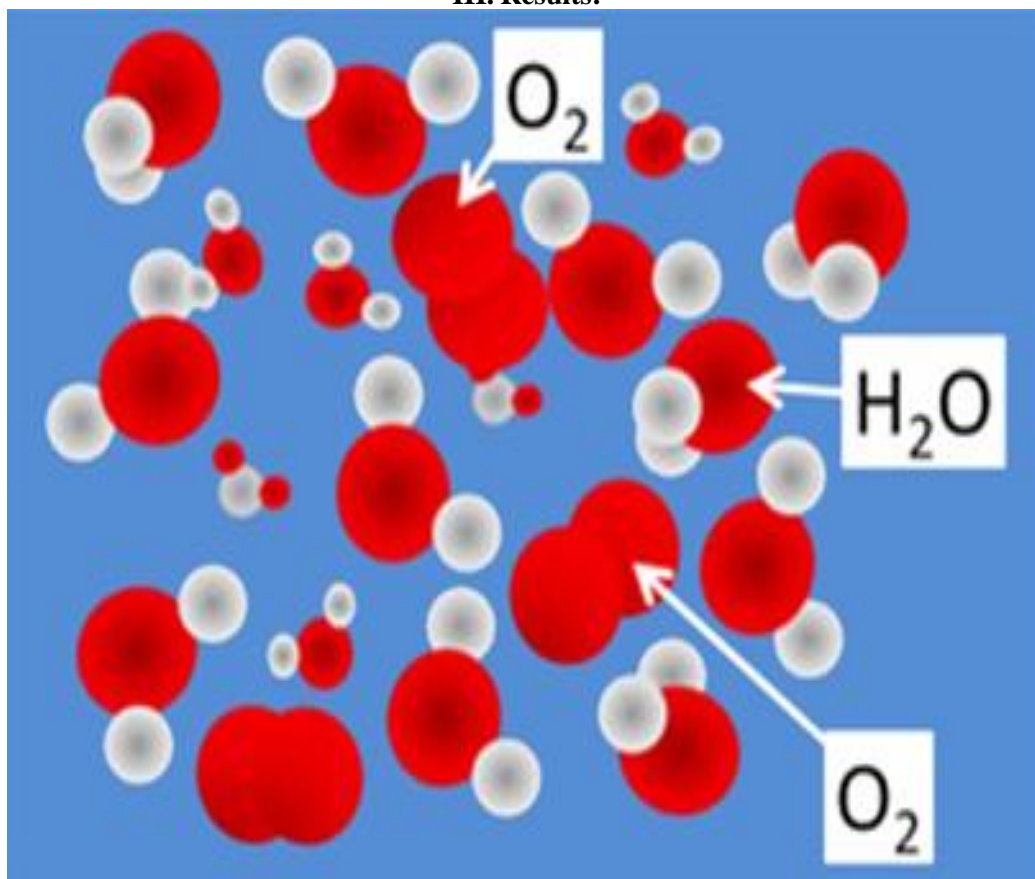


Fig. 1: The molecular representation of trapped  $O_2$  in the intermolecular spaces of water molecules [7]

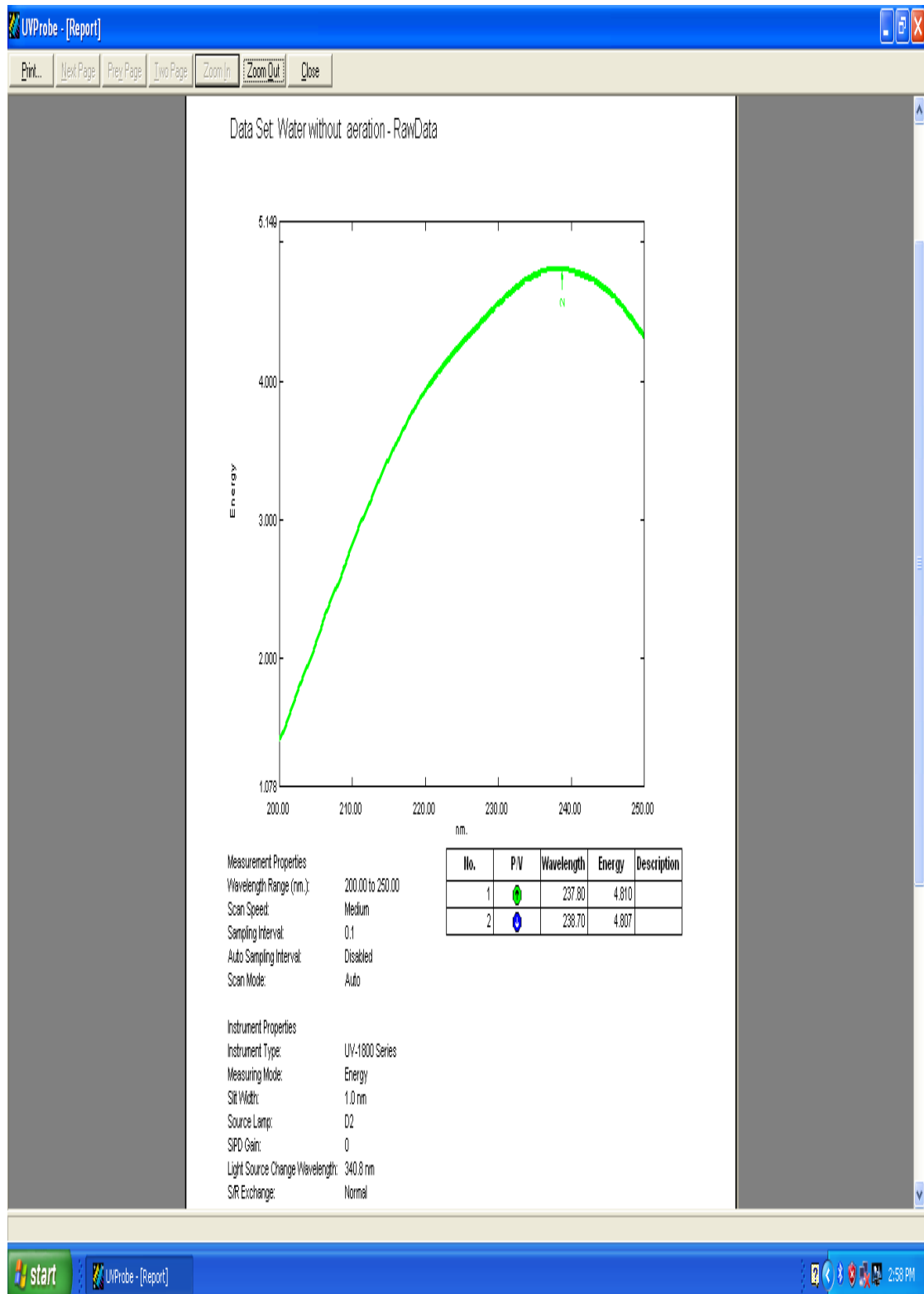


Fig. 2(a): Showing Spectral analysis of Ganga water without aeration

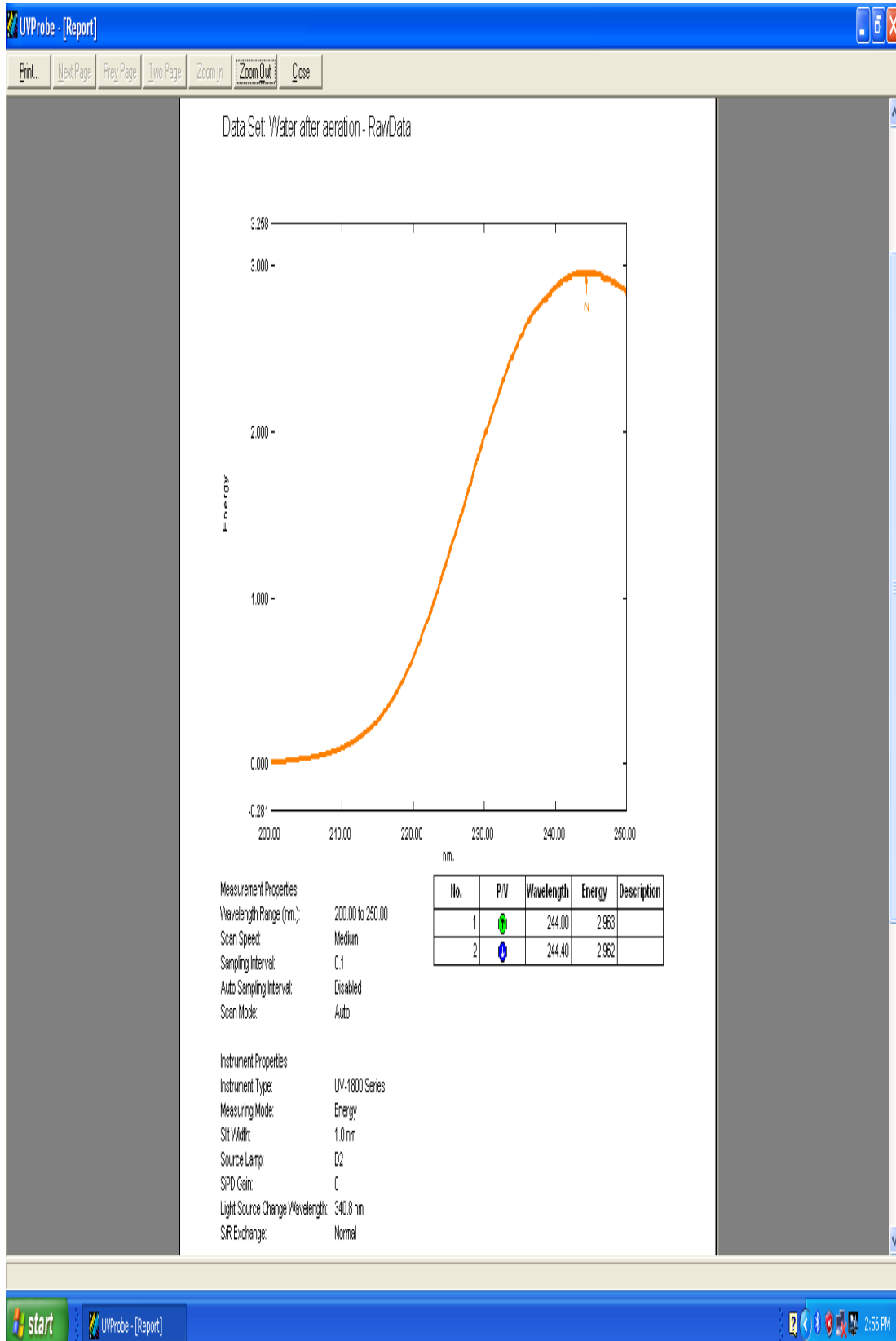


Fig. 2(b): Showing Spectral analysis of Ganga water after aeration

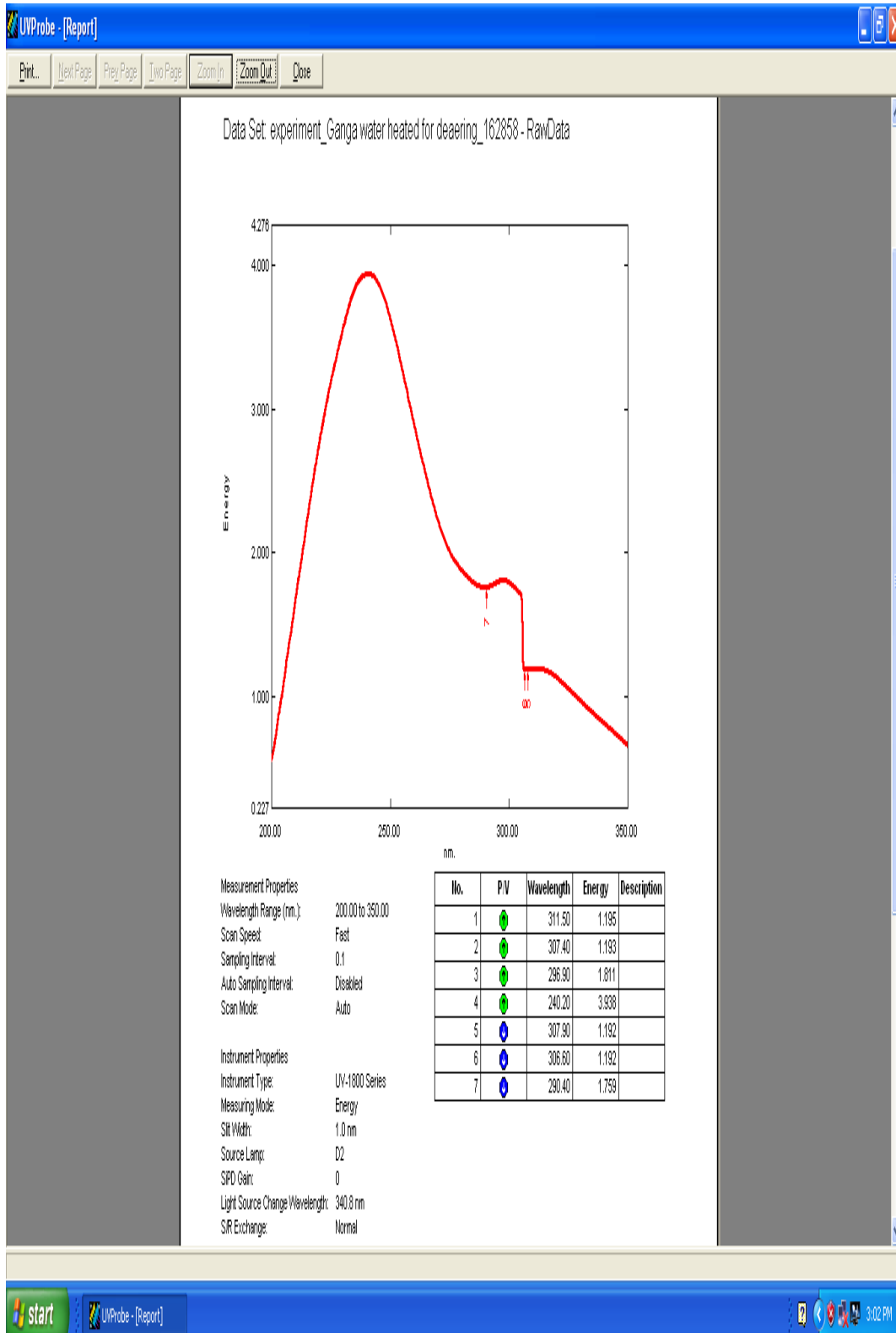
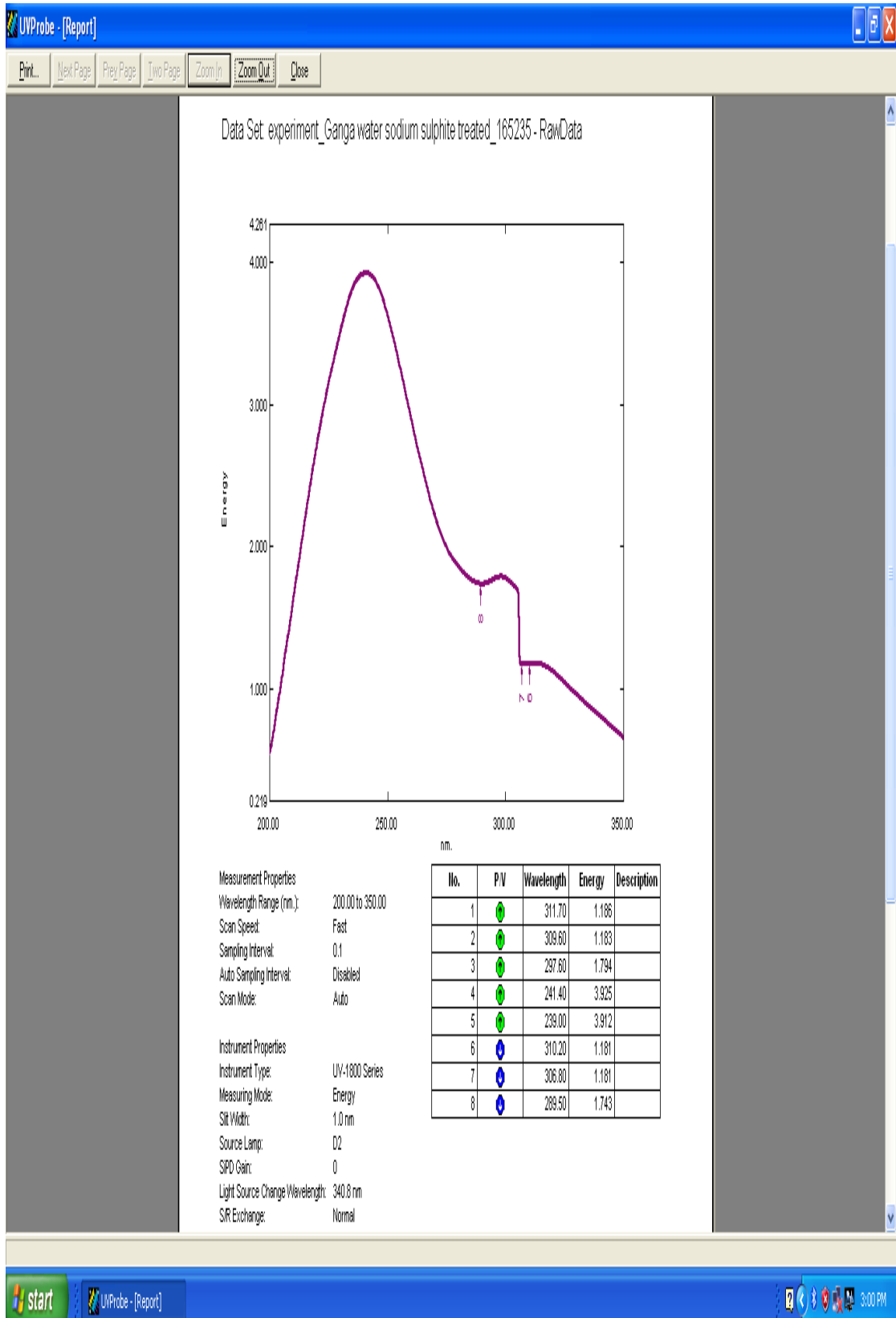


Fig. 2(c): Showing Spectral analysis of Ganga water after deaerating by boiling



**Fig. 2(d): Showing Spectral analysis of Ganga water after deaerating by treating with sodium sulphite**  
**Fig. 2(a), (b), (c) and (d): Showing Spectral analysis of the Ganga water at different states of aeration.**

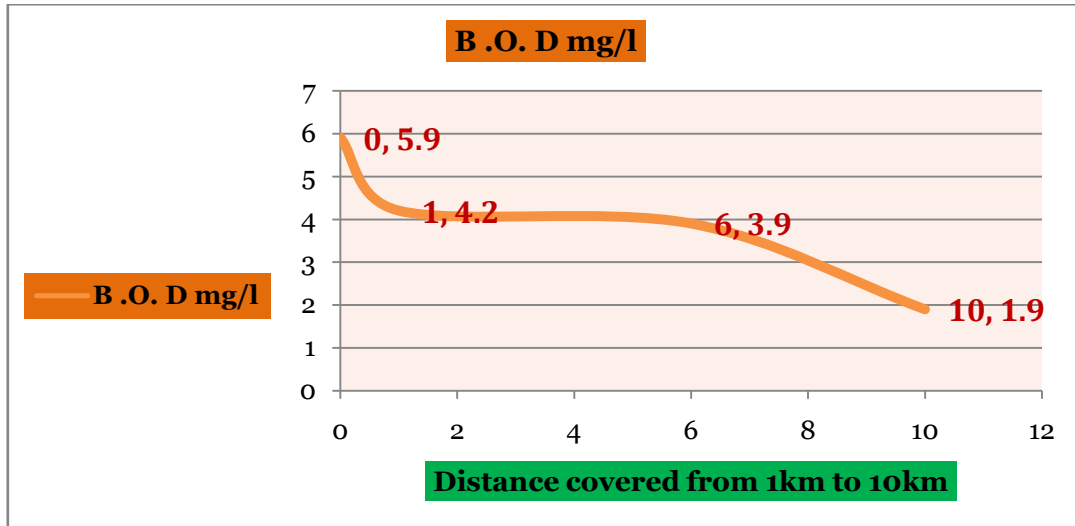


Fig. 3: Variation in D.O. with respect to Distance

Sample ID	Type	Ex	Conc	WL238.0	WL244.0	WL237.7	WL238.7	Comments
1	water without aeration	Unknown	*****	0.002	0.002	0.002	0.002	
2	water after aeration	Unknown	*****	0.040	0.037	0.040	0.039	
3	G water Na <sub>2</sub> SO <sub>3</sub>	Unknown	*****	-0.927	-0.866	-0.856	-0.856	
4	G water direct heat	Unknown	*****	-0.035	-0.035	-0.035	-0.035	
5								
6								
7								

Water without aeration= Ganga water without aeration.  
 Water after aeration= Ganga water after aeration,  
**G Water Na<sub>2</sub>SO<sub>3</sub>= Ganga water treated with sodium sulphite**  
 G water direct heat= Ganga water boiled after aeration

Table 1: Absorbance table of Ganga water with and without aeration

Distance	B.O.D. mg/l	D.O. mg/l
0	5.9	7.0
1	4.2	8.3
6	3.9	8.2
10	1.9	9.0

Table: 2 Table showing distance from the Sewage discharge point and their corresponding BOD and DO of the Ganga Water

Name of Place	Co-ordinates	TC (MPN/100ml)	Count	FC (MPN/100ml)	Count
Krishna Ghat	25°62'20.48"N, 85°16'88.61"E	24000		9000	
Gandhi Ghat	25°62'21.00"N, 85°16'88.90"E	16000		5000	
GaiGhat	22°61'67.59"N, 85°20'51.54"E	9200		3500	
Malsalami (AdrakGhat)	25°59'70.68"N, 85°24'12.09"E	5400		1700	

Table: 3 Table showing the variation in TC and FC at designated locations as a result of increase in D.O. in Ganga water

Type of Sample	Name of Sample Water		
	Ganga water	Distilled water	Tap water
Normal	8.0	7.0	4.0
Aerated	9.4	8.0	8.2
Boiled at 100°C after aeration	2.5	1.6	0.8
Added NaCl at concentration 1g/l after aeration	5.1	4.3	6.0

Tab. 4 Table showing changes in D.O. content of different water samples in different conditions

#### IV. Discussion:

It has been mentioned earlier by Hankin (1896) [2] that when cholera microbes were put in Ganga water, it died within 3 hours and remained alive in distilled water for 72 hours. But the disinfecting property of Ganga water was lost when it was boiled. The reason assigned by Hankin (1896) [2] that there was a volatile material responsible for killing the cholera microbes.

It is very surprising to note that why Hankin (1896) [2] did not assign the reason of dissolved oxygen (D.O.) responsible for the killing of the cholera microbes instead of volatile material. Preliminary studies in the laboratory have shown that D.O. of Ganga water during the pre-monsoon season ranges from 7.2 to 11.5 mg/l which corroborates the findings of Bhargava (1987) [4a] and NEERI's Report (2017) [8]. Methanogenic microorganisms have been categorized as the strictest anaerobes known, with little or no tolerance to O<sub>2</sub> [9, 10]. Inhibitory characteristics of free oxygen towards strict anaerobes present in the anaerobic digesters have been investigated by a number of authors [11, 12, 13]. It is well known that putrefaction can only take place by anaerobic bacteria. This is precisely the reason why cholera bacteria died in Ganga water having high D.O. content and survived in distilled water. The Ganga water lost its disinfecting property when the Ganga water was boiled due to which the dissolved oxygen was lost. The D.O. refers to the free O<sub>2</sub> molecules rather than O atoms bound to H- atoms in the water molecules since D.O. is not bound to H atoms in water molecules. On the molecular scale, D.O. molecules can be illustrated as fitting in space between adjacent water molecules i.e. in the intermolecular spaces (Fig. 1).

We have also studied in our laboratory the spectrophotometric analysis of Ganga water, boiled Ganga water and after the addition of sodium sulphite to make it deoxygenated. It was observed that molecular oxygen dissolved in water absorbed more light than deoxygenated sample suggesting that normal Ganga water was denser because of the entrapped dissolved O<sub>2</sub> (Fig. 2). Similar findings have been reported by Heidit and Ekstrom (1957) [15].

Fig. 3 shows that in about 10 km from the outfall of sewage discharge, the BOD drops from 5.9 to 1.9 mg/l and DO increases from 7.0 to 9.0 mg/l. corresponding to the water temperature which shows unique resilience of Ganga water. Similarly, we observed that T.C. and F.C. count was 24000 and 9000 MPN/100ml at u/s Krishna Ghat (25°62'20.48"N, 85°16'88.61"E), 16000 and 5000 MPN/100 ml at u/s Gandhi Ghat (25°62'21.00"N, 85°16'88.90"E) respectively. At d/s Gai-Ghat (22°61'67.59"N, 85°20'51.54"E) T.C. and F.C. reduced to 9200 and 3500 MPN/100 ml respectively which further reduced to 5400 and 1700 MPN/100ml respectively at Malsalami (AdrakGhat) (25°59'70.68"N, 85°24'12.09"E) (Tab. 3). This is due to very rapid high rate of reaeration (Fig. 3). This reduction was quite pronounced due to normal dying out and increasing D.O. content at Malsalami (AdrakGhat). It is known that in most rivers organic materials exhausts the available O<sub>2</sub> and starts putrefying. The self- purification capacity has been also reported by Bhargava (1983) [4b]. Fig. 3 also shows the rapid decline of BOD till 1 km and thereafter slight increase till 6 km. This increase in BOD was due to several small drains discharging sewage into Ganga water. Thereafter, a further rapid decline in BOD till 10 km where small drains were not found till 10 km, shows a rapid re-aeration and self-purification.

Field observations were further confirmed in our laboratory by saturating D.O. by aeration whereby F.C. reduced to 330 from 1400 MPN/100ml. This reduction was quite pronounced due to high D.O. content.

The molecular structure of D.O. refers to free O<sub>2</sub> molecules rather than oxygen atoms bound to hydrogen atoms in water molecules since D.O. is not bound to water molecules (Fig.1). On an average, each water molecule is coordinated by 4 other water molecules (Wilson, 2010) [14].

We have investigated the D.O. content of Ganga water and also after successive boiling and observed that the D.O. depleted markedly. On an average each water molecule is coordinated by 4 other water molecules. When water boils, all H- bonds are broken. The energy involved in breaking the H- bonds accounts for the high heat of vapourization [14]. When we added sodium chloride, the D.O. also depleted. The depletion of D.O. after adding sodium chloride was due to the displacement of D.O. entrapped into the intermolecular spaces of water molecules with the help of ionic charges of Na<sup>+</sup>, Cl<sup>-</sup> and H<sub>2</sub>O. It is known that the capacity to hold D.O. in water decreases as salinity increases. This is due to the more effective competition between salt for more intermolecular spaces because of their ionic nature and the dissolved oxygen. Similar findings have been reported by Wilson (2010) [14].



During Spectrophotometric study of the Ganga water, it was found that the energy level in the absorption spectra changed in the UV region from 200nm to 250 nm. However the trend continued till 340 nm but for precise reasons the spectral analysis was focused on the wavelength range of 200 to 250 nm as such, more number of peaks appeared in this region. At 244 nm a new peak appeared in the spectral scan of aerated Ganga water which was absent in de-aerated Ganga water. So it is assumed that dissolved oxygen in water shows absorption near the UV region of light. To justify the findings, four closely placed wavelengths were selected to find the absorption of aerated, de-aerated by Na<sub>2</sub>SO<sub>3</sub> and by boiling and normal Ganga water. Fig. 2, Tab. 2 show that molecular O<sub>2</sub> dissolved in water absorb more light than deoxygenated sample at 244 nm and at wavelengths close to it making it denser. It is quite plausible that water with D.O. would be denser than deoxygenated water because D.O. absorb more light than deoxygenated water due to the variable amount of atmospheric O<sub>2</sub> dissolved in water. Similar reports have been made by Heidt and Ekstrom (1957) [15].

There are various reports of some authors giving the antimicrobial properties of water and they are the following:

1. Phytochemicals: There are certain weeds which produce some phytochemicals which kill the bacteria in Ganga water [16, 17, 18]. These bioactive compounds could destroy the bacteria by inhibiting the cell wall synthesis, DNA replication, protein synthesis and altering the intermediary metabolic activities. Some of the algae which have antimicrobial activity contain steroids, alkaloids, phenols and flavonoids.
2. Radioactive substance present in the silt of Ganga water: Frochlich and Walling (2002) [19] and Abbas and Subramanian (1980) [20] have radioactive substances in the silt of Ganga water which emit radons killing the bacteria present in Ganga water.
3. Presence of Bacteriophages: Ganga water has variety of bacteriophages that have specific anti-bacterial activity against pathogens. A French microbiologist D' Herella (1927) [3] established bacteriophages was the cause of antimicrobial property. Khairnar (2018) [21] reported the presence of bacteriophages at the mouth of Gamukh, the origin of Ganga. The antimicrobial activity of river Ganga was assessed by analyzing the water quality in which E. coli was the baseline. It was seen that antimicrobial activity was slightly pronounced. A thorough understanding of the kind of activity and the causative factor requires further experiments to cover multiple aspects of this framework (NEERI Report, 2017) [8].

## V. Conclusion:

It is concluded that the high D.O. content (volatile material) reported by Hankin, (1896) [2] of Ganga water is toxic and lethal for anaerobic bacteria responsible for putrefaction, thus giving a new concept for non-putrefying property of Ganga water. However, this needs to be confirmed by further studying the antioxidant enzymes.

In view of the above, present authors of this paper do not contradict or deny the above 3 factors proposed by different authors but gives another factor for non-putrefying property of Ganga water.

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