Inactivation of Escherichia Coli in Water Using UV Light in Continuous Flow Reactor

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\textbf{Abstract}— In rare progress countries, paucity of healthy drinking water is one of the most challenging health problems. One of the common and most severe pollution of water resources is biological contamination. These deaths are mainly due to gastrointestinal infections from Escherichia coli species. In this research work an attempt was done to do disinfection of water by UV- light in continuous flow reactor. Physico-chemical parameters viz. turbidity (INTU to 6 NTU), pH (6 to 8), temperature (23.9°C to 25.4 °C), total hardness (9.9mg/l to 234mg/l) and bacteriological parameter E.coli (92 to 2400MPN/100ml) were investigated before and after disinfection which is in permissible limits as per IS:10500 (2012) standards except E.coli concentration . As the water flow through the continuous flow reactor, bacteria were exposed to 240nm to 280nm ultraviolet light radiation which causes damage to genetic molecules. First the disinfection was done with uncovered experimental set-up having pair of 6 watts UV light used for microbial inactivation i.e. E.coli, secondly same experimental set-up was modified by covering it with reflecting mirrors i.e. covered experimental set-up to disinfect the water, thirdly same experimental set-up was modified by increasing the length and decreasing the height of experimental set-up and lastly same experimental set-up was modified by increasing the intensity by introducing another pair of 11 watts UV light. The complete experimental set-up runs on three flowrates viz. 100ml/min, 55ml/min, 40ml/min. The uncovered experimental set-up was running at different flow rates does not show any inactivation of E.coli. The modified covered experimental Set-up running at different flow rates show 33% inactivation of E.coli. The modified experimental set-up with increasing length running at different flow rates show 90% (1 log reduction) inactivation of E.coli. The modified experimental set-up with increasing intensity running in all three flowrates showed complete 3 log reduction inactivation of E.coli.

\textbf{Keywords}: Continuous flow reactor, Disinfection, E coli, Flowrates, Ultraviolet radiation

I. INTRODUCTION

Conventional methods of disinfection of water are not so effective and there are problems associated with the usage of very expensive instruments and chemicals (M. Lotierzo,2002) . Diarrheal illnesses are one of the leading causes of morbidity and mortality in developing countries (WHO, 2002). In many analyses of interventions to reduce diarrhoea, improved water quality is shown to have a lower effect than other interventions such as sanitation and hygiene. The contamination of surface and groundwater drinking water sources by pathogens arises from the ingress of residues derived from the faeces and urine of infected humans and animals. As such reservoirs of infection exist in all human and animal populations, the risk of water contamination is always there. Thirty percent of the world’s population lacks access to clean water and consumption of contaminated water contributes to 1.6 million deaths per year (AR Mesadghinia,2009) . A number of point-of-use technologies have been evaluated including boiling, biosand filtration, chlorination, chlorination plus flocculation and ceramic filters. Disinfection using ultraviolet radiation in the UV-C range may be a more favourable option for many applications. The key factor of a UV treatment system is the UV dosage which can be measured from the known UV intensity, exposure time and water flow rate. It does not utilize chemicals and disinfects at much higher rates than SODIS which utilizes temperature and radiation in the UV-A range. UV disinfection is a well-established disinfection technology that has been used in centralized water and wastewater facilities in developed countries for decades. UV radiation inactivates bacteria, viruses, and protozoa, with the benefits of no taste and odour issues, no known disinfection by products (DBPs), no danger of overdosing, relatively fast treatment rates compared to sand and ceramic filters, and low-maintenance requirements. Over the last ten years, small UV systems have become available, including commercially available household systems and the low-cost, locally manufactured UV-tube system that have become an appropriate treatment option for developing communities (Amanda Chau,2008). Limited research has been conducted on the effectiveness of UV-LAMPS, UV-LEDs, for water disinfection. Most of the data available are for Lamps that emit light in the UV-A range (320–400 nm), which is less efficient at disinfection than light in the germicidal range of UV-C (200–280 nm) since it is poorly absorbed by DNA.

II. RESEARCH OBJECTIVES

The goal of this research work was to evaluate the efficacy of ultraviolet light technology to remove the bacteria viz. E.coli from lake water and to improve the public health especially in rural communities in a sustainable, environmentally responsible manner.

Specifically, this research evaluated the use of UV lights at 265nm for inactivation of E.coli in water through the following objectives: (1) To evaluate the performance of continuous flow reactor through analysis of raw and treated water samples. (2) To check the efficacy of removal of E.coli in continuous flow reactor at different flow rates.

III. WHAT IS UV LIGHT:

The sun emits energy over a broad spectrum of wavelengths, visible light that you see, infrared radiation that you feel as heat, and ultraviolet (UV) radiation that you can’t see or feel. UV radiation has a shorter wavelength and higher energy than visible light (Lucinda Hazell,2012). It affects human health both positively and negatively. Short exposure to UVB radiation generates vitamin D, but can also lead to sunburn...
depending on an individual’s skin type. Ultraviolet radiation is an invisible light emitted from the sun. Over 100 years ago European scientists from different countries discovered the top surface of lake-water was sterile when exposed to sunlight. Investigation led to the discovery of Ultraviolet light and to the invention of UV bulbs. Ultraviolet (UV) light is situated in the electro-magnetic spectrum between X-rays and visible light. UV light is split into four main categories, UV-A, UV-B, UV-C and Vacuum UV. The area between 240 and 280 nanometres (nm) is UV-C, commonly known as germicidal light (K.M. Johnson, 2010).

Fig. 1: Electromagnetic spectrum (2, 3)

A. UV Light Generation:

Generation of UV light is similar to the generation of light in a fluorescent lamp. In general, a UV lamp contains an inert gas (e.g., argon) and a small amount of liquid mercury. When a voltage is applied to the lamp, some of the liquid mercury vaporizes. Free electrons and ions then collide with the gaseous mercury atoms, “exciting” the mercury atoms into a higher energy state. Excited mercury atoms have a tendency to return to their ground, or normal, energy state by discharging energy. The energy discharged is in the form of UV light. Mercury is advantageous for UV disinfection applications because it emits light in the germicidal wavelength range (200 – 300 nm) (H.B. Wright, 2008). The UV light produced depends on the concentration of mercury atoms in the UV lamp, which is directly related to the mercury vapor pressure. Low pressure mercury vapor produces monochromatic (light at primarily one wavelength) UV light at a wavelength of 253.7 nm. Higher pressure mercury vapor produces UV light at several wavelengths (polychromatic) (Clement Solomon, 1998).

IV. UV DISINFECTION REACTION

The degree to which the destruction or inactivation of microorganisms occurs by UV radiation is directly related to the UV dose. The UV dosage is calculated as:

\[ D = I \times t \]

Where: \( D \) = UV Dose, mWxs/cm²
\( I \) = Intensity, mW/cm²
\( t \) = Exposure time

Research indicates that when microorganisms are exposed to UV radiation, a constant fraction of the living population is inactivated during each progressive increment in time (Yu. Bilenko, 2010). This dose-response relationship for germicidal effect indicates that high intensity UV energy over a short period of time would provide the same kill as lower intensity UV energy at a proportionally longer period of time. The UV dose required for effective inactivation is determined by site-specific data relating to the water quality and log removal required. Based on first order kinetics, the survival of microorganisms can be calculated as a function of dose and contact time (White, 1992; USEPA, 1996). For high removals, the remaining concentration of organisms appears to be solely related to the dose and water quality, and not dependent on the initial microorganism density. Tchobanoglous (1997) suggested the following relationship between coliform survival and UV dose:

\[ N = f \times D^n \]

Where: \( N \) = Effluent coliform density, /100mL
\( D \) = UV dose, mWxs/cm²
\( n \) = Empirical coefficient related to dose
\( f \) = Empirical water quality factor.

The empirical water quality factor reflects the presence of particles, colour, etc. in the water. For water treatment, the water quality factor is expected to be a function of turbidity and transmittance (Sneha J, 2007).

V. INACTIVATION KINETICS OF MICRO-ORGANISMS:

Inactivation by UV is based on the damage caused to the nucleic acids (DNA/RNA) of the cell or virus (Brahmi Maanouer, 2013). Primarily the formation of pyrimidine dimers, but also of other photoproducts of nucleic acids and nucleic acid lesions, inhibit replication and transcription and hence, prevent the cell or virus from multiplying. The UV absorbance of DNA peaks around 260 nm; at lower and higher wavelengths the absorbance decreases. Below 230nm the absorbance increases again. Most studies used low-pressure mercury lamps with a major wavelength output (85%) at 254nm. The UV sensitivity of the selected microorganisms is described by the parameters of the inactivation kinetics. Inactivation is defined as the reduction of the concentration of culturable micro-organisms N due to the exposure to a concentration disinfectant C during a specific contact time t. The inactivation kinetic for chemical disinfectants is most commonly described by the first-order disinfection model of Chick (1908) and Watson (1908) and the same model can be applied for UV disinfection. The inactivation of microorganisms is usually described by the log inactivation of N. Based on the first-order model, the linear relationship between log inactivation and the UV dose or fluence is described by:

\[ 10\log(N_t / N) = -k \times \text{fluence} \]

Where, \( N_t \) is the microbial concentration after contact time t. Fluence is the product of the UV fluence rate (mW/cm²) and the exposure t (mWs/cm² • m3/cm²).

<table>
<thead>
<tr>
<th>Micro-organisms</th>
<th>Dosage of Ultraviolet radiation (UV dose) in mWsec / cm² needed to kill the selected micro-organisms</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacteria</td>
<td>90% (1 log reduction)</td>
</tr>
<tr>
<td>Bacillus subtilis</td>
<td>11</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>5.8</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
<td>6.6</td>
</tr>
</tbody>
</table>

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Pseudomonas fluorescens 3.5 6.6
Salmonella enteritidis 4 7.6
Salmonella paratyphi - Enteric fever 3.2 6.1
Salmonella typhosa - Typhoid fever 2.1 4.1
Salmonella typhimurium 8 15.2
Sarcina lutea 19.7 26.4
Shigella dysenteriae – Dysentery 2.2 4.2
Shigella flexneri – Dysentery 1.7 3.4
Shigella paradysenteriae 1.68 3.4
Staphylococcus aureus 2.6 6.6
Vibrio comma – Cholera 3.4 6.5
Virus 90% (1 log reduction) 99% (2 log reduction)
Bacteriophage - E. Coli 2.6 6.6
Infectious Hepatitis 5.8 8
Poliovirus – Poliomyelitis 3.15 6.6

Table 1: Shows UV dosage for destroying micro-organisms (3)

VI. METHODOLOGY
Water from an established Ambazari lake, Hingna road, Nagpur, Maharashtra, India, (21.1287° N, 79.0405° E) has been collected for the research work. The lake water is supplied to the 13.5 MLD water treatment plant in MIDC, Hingna to the industrial area. The characterization of raw water i.e. lake water was determined for different seasons of ten day’s intervals by standard testing equipments and methods as per APHA standards. In characterization of raw water turbidity, pH, temperature and hardness was determined before and after disinfection and the results so obtained was compared with the drinking water standard IS:10500 2012. The physico-chemical parameters viz. turbidity, pH, temperature and hardness were in permissible limit except bacteriological parameter viz. E.coli. Initially E.coli concentration of raw water sample has been tested by a standard multiple tube dilution (MTD) test as per APHA standards, the water has been passed to all the continuous flow reactor with the help of peristaltic pump, the water is collected at the outlet of the continuous flow reactor and tested for the final concentration of E.coli by MTD test. The complete set-up of UV light system is show in fig 6.1.

VII. EXPERIMENTAL SET-UPS
A low pressure (LP) Philips UV lamps of 6 and 11 watts is housed in an continuous flow reactor connected in parallel on direct power supply. The lamp is of 15cm long and 1.5cm diameter. In this project peristaltic pump is used to pass the water through the continuous flow reactor at same flowrate. The continuous flow reactor has been modified three times to achieve the maximum inactivation of E.coli. The experimental set-up contained a uncovered glass box model of size 25cmX10cmX10cm having two 6 watts low pressure UV germicidal lamp connected in parallel as showed in fig 7.1, which emit monochromatic wavelength. The lamp is attached to the upper cover of the box with distance of lamp from the water level was 7cm inside the continuous flow reactor.

Fig. 6.1: Set-up of uv light system.

As the result is not satisfactory, the same experimental set-up glass box of size 25cmX10cmX10cm has been modified by providing reflecting mirror from inside the box to the whole area i.e. covered experimental set-up, as showed in fig 7.2, provided with the same pair of UV lights of 6 watts. The distance of light from the water level is 7cm.

Fig. 7.1: Uncovered experimental set-up.

Fig. 7.2: Modified covered experimental set-up.

As only few removal of E.coli has been observed, a new experimental set-up has been fabricated with increasing length and decreasing height of set-up . The new modified experimental set-up is of size 32cmX10cmX5cm with reflective glasses from inside to the whole area as showed in fig. 7.3, provided with a pair of 6 watts UV light. The distance
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VIII. RESULTS AND DISCUSSION:
The characterization results of water sample i.e. turbidity, pH, temperature and total hardness obtained within permissible limits as per IS: 10500, 2012 and hence pre-treatment were not given to raw water. Multiple Tube Dilution (MTD) test was performed for finding E.coli in raw water and after disinfection for duration of 1st November 2014 to 31st March 2015.

Fig. 8.1: E.coli concentration from uncovered experimental set-up for the month of November 2015.

Fig. 8.1 showed the result from the uncovered experimental set-up is not satisfactory as there was no inactivation of E.coli. Most of the radiation from UV light gets refracted outside the experimental set-up. Therefore the experimental set-up was modified to increase the efficacy of inactivation of E.coli.

Fig. 8.2: E.coli concentration from covered experimental set-up for the month of December.

Fig. 8.2 showed the result from the covered experimental set-up shows some inactivation of E.coli at the flow rate of 40ml/min. The inactivation of E.coli comes down to 1600 MPN from 2400 MPN of raw water sample. The result does not show any inactivation for the flow rate of 100ml/min and 55ml/min because of less detention time. The radiation in covered experimental set-up were reflected back because of reflective mirrors, which increases the radiation and temperature inside the covered experimental set-up.

Fig. 8.3: E.coli concentration from modified increased length experimental set-up for the month of January.

In fig 8.3, the modified experimental set-up with increasing length and decreasing the height shows good result with an 1 log reduction of inactivation of E.coli. Due to increasing the length of experimental set-up, the detention time increases and the water is subjected to longer time for inactivation, the height of the continuous flow reactor is decreased by 5cm, so that the radiation penetrate more in the water and increase the efficiency of inactivation.

Fig. 8.4: E.coli concentration from modified increased intensity experimental set-up for the month of February.
Fig 8.4 showed the removal of 3 log reduction of E.coli for all the flowrates viz. 100,55,40 ml/min. Introducing another pair of UV light of 11 W increases the intensity in continuous flow reactor, this inactivates the E.coli at much higher rate.

IX. CONCLUSIONS
This work showed that the UV disinfection by the UV light can be used for disinfection of water. The continuous flow reactor performed well and disinfected 99.99% of E.coli under visible light irradiation at a optimum flow rate of 153ml/min. Turbidity influence the disinfection efficiency of UV light and slightly decreases after disinfection. The variation of pH does not affect the disinfection by UV light. The pH slightly increases after disinfection. The temperature affects the inactivation efficiency and increasing the temperature will increase inactivation efficiency. High concentration of salts in water affect the disinfection, however hardness slightly decreases after disinfection due to dissociation.

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