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Assessment of Wastewater Disinfection efficiency by modified SODIS technique and its reuse potential

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Abstract: *The present study has been attempted to adopt the potential of modified SODIS technique for the assessment of (i) the wastewater disinfection efficiency (ii) the optimum time duration required for wastewater disinfection from pathogenic microbial contamination, and (iii) application potential of the disinfected wastewater for the germination of wheat seed. The results revealed that the light intensity and temperature dependent 100% decrement of heterotrophic, E coli, Salmonella sp. and Enterobacteriaceae bacterial population. The seed germination study showed that germination rate varies from 90 to 100% from 1to 4h, whereas vigour index varies from 1158.3 to 1596. Relative root growth and chlorophyll content varies from 116.28 to 133.67 and 0.259 to 0.448mg/ GFW. This study significantly explored the excellent use of modified SODIS method in wastewater disinfection by a box type solar cooker and its significant reuse. Before application, its disinfection and treatment is necessary. Therefore, it can be concluded that modified SODIS method involves low cost, environment friendly method for wastewater disinfection that uses solar energy to kill/destroy the pathogenic microbial load and various toxic pollutants, which make it safe for reuse.*

Keywords: *SODIS, Wastewater, Solar cooker, Vigour index, Pathogenic*

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1. Introduction:

Wastewater is one of the important store houses/sources of various nutrients (carbon, nitrogen and phosphorus, *etc*.). In addition to important source of nutrients and pollutants, wastewater is considered as a habitat for vast array of water borne pathogenic microorganisms including bacteria, viruses and protozoa. Discharge of pathogenic microorganisms containing untreated wastewater into environment may threaten public health (Zhou et al., 2007; Dababneh et al., 2012; Pandey et al., 2014). Therefore, it is advisable to treat the wastewater before discharging into the environment and removal techniques for various kinds of pollutants to protect the environment and adverse impacts. In order to use the wastewater for agriculture, aquaculture or any other purposes, disinfection is necessary to avoid the adverse impacts of pathogenic microorganisms. Various types of disinfection techniques (Dejung et al., 2007) have been introduced in this respect, but most of them are quite expensive and complicated to operate (Altherr et al., 2006). Therefore, development of an effective, eco-friendly disinfection system with a low infrastructural and installation costs, easy to use and handling and little maintenance (Queluz and Sánchez-Román, 2014).

Solar disinfection (SODIS) technique has been widely used in the water treatment purpose. Most literature mainly reported the disinfection of drinking water through SODIS technique (McGuigan et al., 2012; Bitew et al., 2018) and lesser attention has given for application of SODIS in case of wastewater treatment. Consequently, the wide studies concerning the extensive application of SODIS technique in optimizing various parameters required in treating the wastewater have not been performed so far as well as the question, whether the SODIS can disinfect the pathogenic microbial contamination in wastewater has still remain unclear. Therefore, the present study has been aimed to utilize the potential of modified SODIS technique for the assessment of (i) the wastewater disinfection efficiency (ii) the optimum time duration required for wastewater disinfection from pathogenic microbial contamination, and(iii) application potential of the disinfected wastewater for the germination of wheat seed.

2. Materials and methods

2.1. *Disinfection Experiment*

2.1.1. *Experimental design*

The experiment was conducted using solar disinfection system SDS, Geetanjali Solar Enterprise. The solar cooker employed as most effective and SDS is the popular box type appliances chosen for the present disinfection study. Four black circular aluminium pots (2 L) of SDS (solar cooker) were employed as experimental vesicle to carry out the disinfection

experiment. The experiments were carried out at the outdoor of the Laboratory, Department of Ecological Studies, University of Kalyani, Kalyani. The six experiment were performed during six months (one experiment in each month) from January, 2018 to June, 2018 to determine the effect of seasonal variation of solar intensity on the performance efficiency of SDS in wastewater disinfection. The wastewater (5.0 L) sample (pH-6.8, PO_4-P- 2.011 mg/L, NO_3-N- 0.788 mg/L, NO_2-N- 0.416 mg/L, NH_4-N- 2.107mg/L) was collected from Kalyani sewage treatment plant, Kalyani, India (22.9751° N, 88.4345° E), filtered to remove the large particles and divided into two parts. One part of the sample was used as initial sample to record the initial reading and other part of the sample was used for treatment. The aluminium pots (2 L) were filled by filtered sample at the rate of 1.0 L/pot. The four sets of water filled aluminium pots were placed within the SDS at 10.00 am and each set of pot was removed from the SDS at every 0,1, 2, 3 and 4 h interval and cooled to bring the normal temperature for analysis the samples.

2.1.2. *Solar disinfection system*

The solar cooker used in the experiments was made with aluminum, with a toughened glass, processed for the method of thermal aspersion in a steel matrix, with the objective of increasing its energy efficiency as shown in(Fig.1.1). The solar cooker used as SDS in study consists of the following specifications:

i	Body dimension (L×W×H)	: 23×23×"
ii	Reflector material	: Mirror glass, 4 mm
iii	Plane glass	: 4 mm
iv	Insulation materials	: Glass wool
v	04Nos.Aluminium container covered with toughened glass	: 4 mm
vi	Inbuilt analog	: Temperature meter
	Approx. weight	: 4 kg

Fig.1.1. Box type solar cooker(a) Reflector, (b) Double glazing, (c) Aluminium container, (d) Temperature meter of the different parts in SDS

2.1.3. *Culture media and glass wares*

Cultured media used in this study was chromocult agar (MERCK)). The following are composition of the chromocult agar media was mentioned in general methodology section.

All glass wares and sample bottles were soaked in diluted HCl solution for 12 h, washed and then rinsed several times with distilled water. Double distilled and milli Q waters were used for the preparation of reagent solutions.

2.1.4. *Temperature and light intensity measurement*

Temperature (ºC) inside the solar cooker and light intensity were measured at regular intervals of 0, 1, 2, 3 and 4 h periods using thermometer and LUX meter respectively throughout the experimentation.

2.1.5. *Microbial examination*

The samples were collected from the experimental pots. All samples were at first sequentially diluted with sterile buffered water using three dilution factors (10^1 – 10^3) based on previous analyses of similar samples. Then the aliquots were examined for microorganisms using membrane filtration methods (APHA, 2012). This method is the basis of the widely used membrane filtration methods for detecting indicator bacteria, including total and fecal coliforms, enterococci and *Clostridium perfringes* (Eaton et al., 1998). The method used microporous membranes are typically composed of cellulose esters. The sample (mL)

was filtered through the membrane. The cells recovered on a membrane filter were directly cultured on chromo cult agar medium to determine total coliforms and *E. coli*. The plates were incubated at 37°C for 24hour. Each analytical batch included blanks and positive controls. After incubation, the plates were manually enumerated. All results were volume normalized to give concentrations in colony forming units (CFU) per 100 ml.

The nutrient parameters (Phosphate-P, Nitrate-N, Nitrite-N and Ammonium-N) were measured at regular intervals during this experimental period using the method APHA (2012).

After the experiment, the treated wastewater was used for the germination of wheat seed (*Triticum aestivum)*.

2.1.6. *Seed Germination Experiment*

Seed germination study was performed using the domestic wastewater collected from modified SODIS disinfection experiment at 0, 1, 2, 3 and 4 h periods to assess whether the treated/disinfected water has the seed germination ability without pathogenic infection. Three hundred seeds of wheat (*Triticum aestivum*) were surface-sterilized with 2% sodium hypochlorite solution for 1 min and washed 5 times in single distilled water followed by air-drying (Miche and Balandreau, 2001). All dried sterilized seeds were randomly divided into five groups with three replicate (20 x 3 x 5 = 300). Five groups were treated by 5 mL wastewater collected at 0, 1, 2, 3 and 4 h periods of disinfection experiment for 10 min and then air-dried. The triplicate sets of fifteen healthy air-dried seeds of each group were placed on sterilized Petri dishes (9 cm) containing two layers of moistened filter paper. A triplicates sets of fifteen healthy seeds were treated with sterilized distilled water, air dried and placed on sterilized Petri dishes (9 cm) containing two layers of moistened filter paper was maintained as control group. All the Petri dishes were incubated in a light incubator.

The seed germination rate, root length, shoot length, fresh weight, dry weight and chlorophyll content of plants (Arnon, 1949) were measured at seventh and fifteenth day of experimentation (Tian et al., 2014). Samples were dried in an oven at 60°C for 48 h to measure the dry weight. The number of leaves of each plant was counted after harvesting. The germination rate, relative seed germination rate, relative root growth, germination index and vigour index were calculated according to the following equations (Islam et al., 2016):

$$\text{Germination rate (\%)} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds}} \times 100 \quad (1)$$

$$\text{Relative seed germination rate (\%)} = \frac{\text{Number of seeds germinated in control}}{\text{Number of seeds germinated in sample}} \times 100 \quad (2)$$

$$\text{Relative root growth (\%)} = \frac{\text{Average root length in sample}}{\text{Average root length in control}} \times 100 \quad (3)$$

$$\text{Germination Index (\%)} = \frac{\text{Relative shoot growth}}{\text{Relative root growth}} \times 100 \quad (4)$$

$$\text{Vigour index} = \% \text{ of germination} \times \text{total plant length} \quad (5)$$

2.1.6.1. *Chlorophyll content measurement*

Chlorophyll content of the leaf samples was analyzed using Arnon's method (1949). The fresh leaves (0.2 g) were ground in mortar and pestle with 10 mL of 80% acetone. The homogenate obtained was centrifuged at 6000 rpm for 5 min. The supernatant was used for chlorophyll estimation using spectrophotometer (Shimadzu, UV-Vis Spectrophotometer) at 663, 645 and 652 nm wavelengths. Chlorophyll content of leaves was further calculated from the following equations:

$$\text{Chlorophyll a (mg g}^{-1}\text{ FW)}: (12.7x\ O.D_{663} - 2.69\ x\ O.D_{645}) \times \frac{v}{1000} \times W \quad (6)$$

$$\text{Chlorophyll b (mg g}^{-1}\text{ FW)}: (22.9\ x\ O.D_{645} - 4.68x\ O.D_{663}) \times \frac{v}{1000} \times W \quad (7)$$

$$\text{Total chlorophyll (mg g}^{-1}\text{ FW)}: (O.D_{652} \times \frac{1000}{34.5} \times \frac{v}{1000} \times W) \quad (8)$$

Here, FW stands for fresh weight; V is the final volume of 80% acetone; W is the weight of leaves

3. Results and Discussions

3.1. *Disinfection Experiment*

3.1.1. *Light intensity and temperature*

The light intensity and temperature were ranged from 0.84 to 4.58µmol/s/m^{2}and 45 to 115°C, respectively. The light intensities increased from 0.84 to 3.86, 3.61 to 4.51, 3.36 to 4.45, 4.28 to 4.58, 4.19 to 4.58 and 3.13 to 4.58 58µmol/s/m^2 in January, February, March, April, May and June, respectively as well as temperatures were increased from 45 to 100, 45 to 100, 60 to 110, 65 to 110, 70 to 115 and 45 to 100°C in January, February, March, April, May and June, respectively. The average light intensity and temperature were maximum (light intensity 4.28 µmol/s/m^2 and temperature 70°C) in April and May month, respectively as well as minimum

(light intensity 0.84µmol/s/m^2 and temperature 45°C)in January and February month, respectively (Fig.1.2).

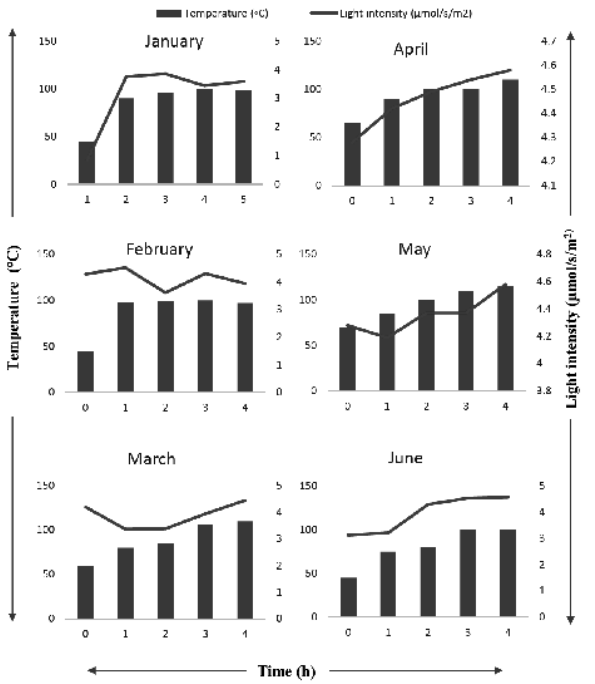

Fig: 1.2 Variation in temperature and light intensity with time progressed in different months

3. 1.2. ***Microbial population***

The change of microbial populations with time, temperature and light intensity was varied in different months (from January to June), which was graphically represented in Figures 1.3, 1.4, 1.5 and 1.6.

3.1.2.1.*Heterotrophic bacteria*

The heterotrophic bacterial population varied from 60 – 5.08 × 10^3/mL in all the months (Fig.1.3). The bacterial population was decreased with increasing time and temperature in all the months studied (Fig.1.3). The decreasing rates of heterotrophic bacterial population were 87%, 94%, 99%, 99%, 99% and 99% in January, February, March, April, May and June,

respectively at 4 h- period of experimentation. At 1 h period, the decrease in heterotrophic bacterial population was maximum (99%) in April and minimum in January (87%) months.

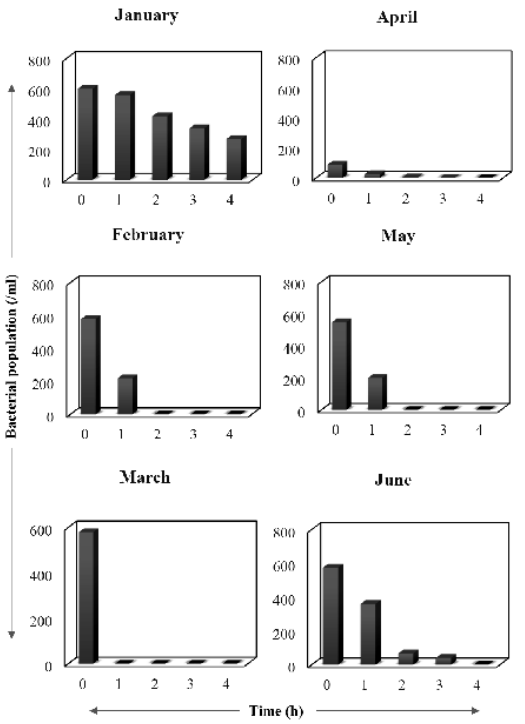

Fig.1.3. Change of heterotrophic bacterial population with time, temperature and light intensity in different months (from month of January to June)

In the month of January, the initial temperature was 45˚C, which increased to 100˚C within 3h and decreased to 98˚C thereafter. Likewise, light intensity increased from 0.84µmol/s/m²to 3.86 µmol/s/m² and then decreased thereafter to 3.59µmol/s/m². The percentage of reduction in bacterial population was 87% showing direct relationship with time. There was very little variation of temperature between January and February but light intensity was quite high 4.28µmol/s/m² at the time of initial temperature 45˚C. As the experiment proceeds little fluctuation in light intensity was observed but temperature increased constantly upto 100˚C with time and at 4 h, there was sudden drop in temperature (97˚C) and 94%bacterial population reduction was achieved in February. In the month of March and April, the temperature at

initiation period of the experiment was 60˚C and 65˚C, which was quite higher from the previous two months and it increased to 110˚C with time and the light intensity during the experiment was also high resulting in 99.9% and 99.7% reduction in bacterial population in the month of March and April, respectively. In the month of May, initial temperature was high 70˚C, which reached to 110˚C within 3h. Similarly, light intensity increased from 4.28to 4.58μmol/s/m^2resulting in 99.7% reduction in bacterial population. In the month of June, the initial temperature of 45˚C was reached to 80˚C within 2 h as the experiment proceeds and reached to 100˚C within 3h from the initiation time and remains stable upto next 4h, whereas, light intensity keeps on increasing with time but the bacterial population achieves 99% reduction at the end of the experiment.

3.1.2.2. *Escherichia coli*

The number of *E. coli* bacteria ranged from 0.6 – 0.5 x 10^3/mL in all the months (Fig.1.4). The bacterial number was decreased with increasing time and temperature in all the months studied (Fig.1.4). The decreasing rates of coliform bacteria were 55%, 100%, 100%, 100%, 100% and 100% in January, February, March, April, May and June, respectively at 4 h periods of study. At 1 h period, *E. coli* population decreased in maximum (100%) in March and minimum in January (55%) months. The 100% decrement of *E. coli* population was found in February to June except January.

Figure 1.4 clearly disclosed that *E.coli* population was decreased with time, temperature as well as with the light intensity. In the month of January, 55% reduction in bacterial population was observed which was related with temperature (r=-0.74695) as well as with time (r=0.801).

Similarly, in the month of February 100% reduction was observed within 4 h period, although very little fluctuation in temperature was observed between January and February. The temperature (60˚C) and light intensity (4.2 μmol/s/m^2) was quite high at initiation time of experiment in the month of March, which further increased as the time progressed and reached to 110˚C and 4.45 μmol/s/m^2 within 4 h resulting in 100% destruction of coliform (*E. coli*). In the month of April and May, a similar trend was observed in *E.coli* population. The temperature was increased from 65 to 110˚C and 70to 115˚C within 4h resulting in 100% reduction of *E. coli* in April and May, respectively. Light intensity of April and May increased from 4.28to 4.58μmol/s/m^2. In the month of June 100% reduction in *E.coli* was observed within 3 h exposure of wastewater to solar radiation when light intensity and temperature reached3.13 to 4.58 μmol/s/m^2 and45 to 100˚C within 4 h. Thus, temperature, time and light intensity are the important parameters responsible for destruction of bacterial population. The

bacterial inactivation rate in wastewater sample is proportional to the intensity of sunlight and atmospheric temperature (Acra et al., 1990). In this month temperature ranges between 45 and100˚C which was enough for destroying bacteria as temperature above 45˚C completely inactivates the bacteria present in water (Dunlop et al., 2011). The rate of bacterial inactivation for sample temperatures ranging from 12 to 40°C, when the water temperature was increased to 50°C, the same fraction of the initial population of *E. coli* was inactivated (Caslake et al., 2004).

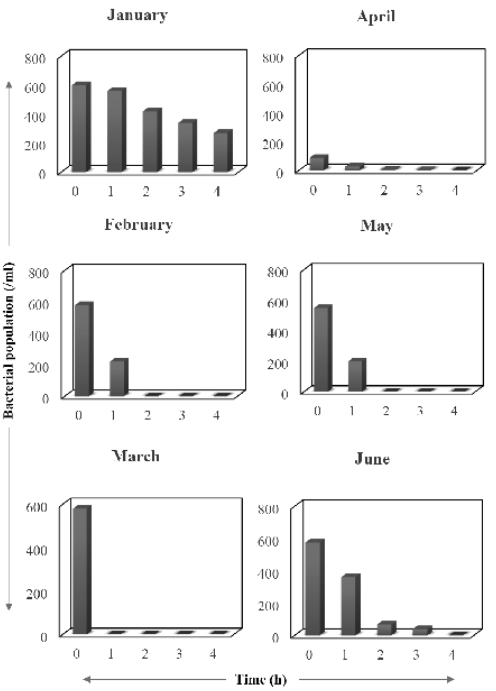

Fig.1.4. Change of *E Coli* population with time, temperature and light intensity in different months (from month of January to June)

3.1.2.2 Salmonella sp.

The population of *Salmonella* sp. varied from 0.6 – 0.5 x 10^3/mL in all months (Fig.1.5). The bacterial population was decreased with increasing time and temperature in all the months studied (Fig.1.5). The decreasing rates of *Salmonella* sp. population were 66%, 100%, 70%, 100%, 100% and 100% in January, February, March, April, May and June, respectively at 4 h period of study. The *Salmonella* sp. was not found in February, March, April, May and June at 4 h periods of experimentations, respectively.

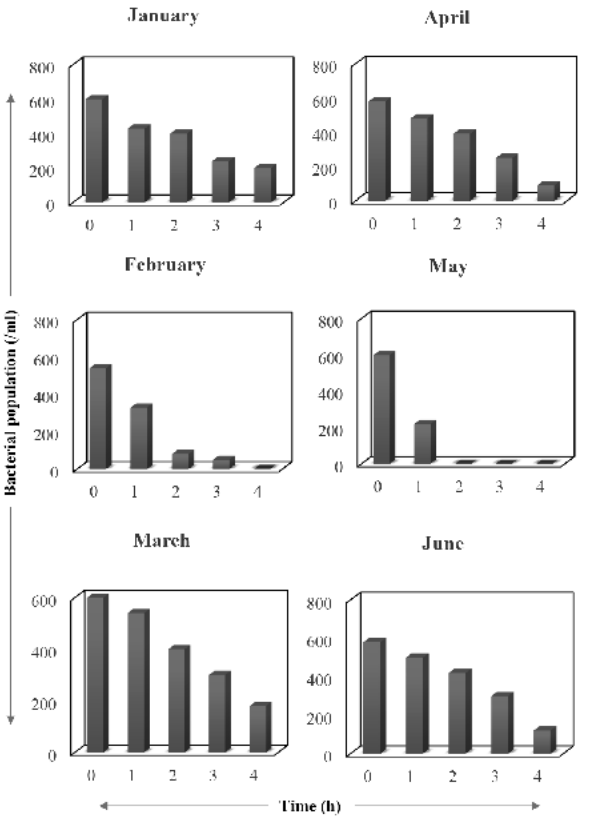

Fig.1.5. Graphical representation of *Salmonella* population in various months

In the month of January the initial temperature and light intensity were 45˚C and0.84 µmol/s/m^2 which increased to 100˚C and 3.44 µmol/s/m^2 within 3 h period of experiment, respectively that reduced 66.7%*Salmonella*. A sharp temperature increment in the month of February resulted in 100% killing of bacterial population. In the month of March, April and May, the initial temperature inside the box type solar cooker was increased from 60to 110˚C, 65 to 70˚C and 110 to 115˚C,respectively within 4h which was resulted in 100% destruction of bacteria. Likewise, in the month of June, the initial temperature and light intensity were increased from 45˚C and 3.13µmol/s/m^2 to 80˚C and 4.3 µmol/s/m^{2}results, respectively. Due to lower temperature, the reduction of *Salmonella* population (88%) was lower compared to other months within 4 h period

of experimentation. Sunlight destroyed biomolecules and UV-A (wavelength of 320–400 nm) was absorbed by DNA and reactive oxygen species (ROSs) produced by solar radiation in water or wastewater. UV-A and UV-B from sunlight and the reactive molecules kill microorganisms, such as *Pseudomonasaeruginosa*, *Escherichia coli*, *Salmonella*and *ShigellaFlexneri* (Polo-López et al., 2011; Giannakis et al., 2015; Lawrie et al., 2015).

3.1.2.3. Enterobacteriaceae group

The *Enterobacteriaceae* bacterial population varied from 0.6 – 0.5 x 10^3/mL in all the months (Fig.1.6). The bacterial population was decreased with increasing time and temperature in all the months studied (Fig.1.6). The decreasing rates of Enterobacteriaceae population were 73%, 62%, 100%, 100%, 100% and 100% in January, February, March, April, May and June, respectively at 4 h period of study. The maximum and minimum reduction in bacterial number was found in March (100%) and June (100%), respectively.

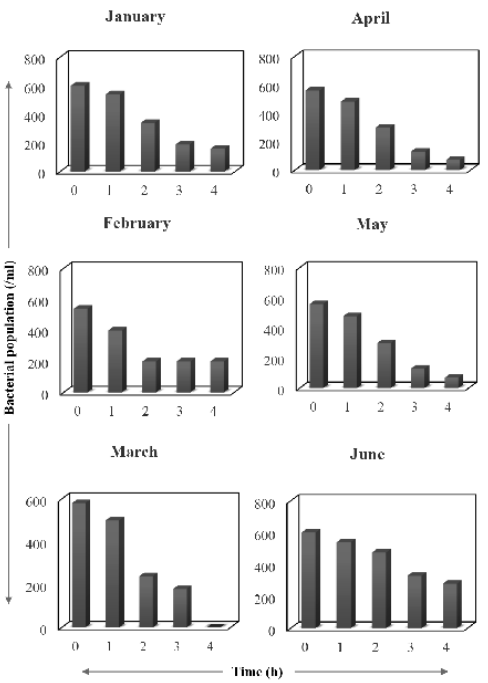

Fig.1.6. Graphical representation of Enterobacteriacae group in various months

In the month of January and February, the initial temperature inside the box type solar cooker was 45°C which increased to 100°C within 3 h, whereas no appreciable decrement of *Enterobacteriaceae* bacterial population (73%) was found, although the light intensity increased from 0.84 to3.59 μmol/s/m^2. In February, light intensity decreased from 4.28μmol/s/m^2 to 3.94μmol/s/m^2 resulting in lower (63%) destruction in bacterial population. The initial temperature inside the box type solar cooker were higher 60, 65 and 70°C, respectively in the month of March, April and May which increased to 110, 110 and 115°C within 4 h period. The 100% bacterial reduction was found in March, except other five months. Within next 2 h this temperature and light intensity increases to 100°C and 4.58μmol/s/m^2 resulting in 53% of bacterial population. Bacterial inactivation by solar radiation was completely depends on light intensity, solar exposure time, availability of oxygen and temperature (Reed, 2004; Berney et al., 2006). In the present study temperature reached to 193°C after 4 h which was directly proportional to reduction in bacterial load (99% heterotrophic bacteria and 100% coliform group), similar to the study of Oates et al., (2003). In the present study at the time of initiation temperature inside the solar cooker was 65°C which increases to 110°C after 4 h which is quite sufficient in reducing bacterial load from wastewater samples. From the above figure it was clearly observed that with increase in time temperature increased which caused decrement of bacterial populations present in waste water sample. In the present study temperature lies between 70 and 115°C which was quite high for reduction of microbial load, because temperature varied between 20 and 40°C, the inactivation rate of fecal coliforms remains constant (Wegelinet al., 1994). Above temperatures of 50°C, the microbial inactivation is enhanced through the synergistic effects of UV and temperature (Navantoff et al., 2008; Bitton, 2011; Giannakiset al., 2014). In case of solar disinfection of water the main mechanism involves inactivation of microorganisms by UV radiation is caused by damage to the genetic material (DNA / RNA) (Hijnen et al., 2006). UV-C radiation is responsible for *Klebsiella* species destruction as it damages their genetic material caused by thymine dimerization which blocks nucleic acid replication thereby inactivates the cell (Bitton, 2011). Mc Cambridge, (1981) described the susceptibility of bacteria to light induced degradation varied among different organisms which was represented as *Klebsiella pneumoniae>E. coli>Salmonella typhimurium*, *Streptococcus faecium*, *Enterobacter aerogenes*, *Erwinia herbicola*. The 100% decrement of *Enterobacteriaceae* bacterial population signified that the disinfection of wastewater from *Enterobacteriaceae* bacteria can be done by SODIS techniques.

Correlation between the light intensity and temperature with heterotrophic, *E coli*, *Salmonella* sp. and *Enterobacteriaceae* bacterial population showed the negative relationship

(Table-1.1). The above results clearly revealed that heterotrophic bacterial population is significantly decreased with increasing light intensity and temperature within the 4 h period of experimentation. The light intensity and temperature dependent 100% decrement of heterotrophic, *E Coli*, *Salmonella* sp. and *Enterobacteriaceae* bacterial population indicated that the wastewater can be treated by SODIS techniques.

Table 1.1: Correlation between different month, bacteria, light intensity and temperature

Month	Bacteria	Light intensity (μmol/s/m^2)	Temperature (°C)
January	Heterotrophic bacteria	– 0.61386	– 0.75487
	E.coli	– 0.57905	– 0.74695
	Salmonella sp.	– 0.71567	– 0.85866
	Enterobacteriaceae	– 0.58566	– 0.76435
February	Heterotrophic bacteria	0.481719	– 0.79166
	E.coli	0.479471	– 0.93015
	Salmonella sp.	0.540531	– 0.83208
	Enterobacteriaceae	0.579424	– 0.83742
March	Heterotrophic bacteria	0.662161	– 0.84009
	E.coli	0.239787	– 0.77272
	Salmonella sp.	0.443933	– 0.95381
	Enterobacteriaceae	0.550978	– 0.92819
April	Heterotrophic bacteria	– 0.95781	– 0.97459
	E.coli	– 0.86234	– 0.91132
	Salmonella sp.	– 0.94066	– 0.86702
	Enterobacteriaceae	– 0.93674	– 0.88066
May	Heterotrophic bacteria	– 0.6457	– 0.96251
	E.coli	– 0.30111	– 0.78536
	Salmonella sp.	– 0.55258	– 0.93021
	Enterobacteriaceae	– 0.40928	– 0.85803

June	Heterotrophic bacteria	– 0.89551	– 0.96722
	E.coli	– 0.95965	– 0.90537
	Salmonella sp.	– 0.86825	– 0.90038
	Enterobacteriaceae	– 0.87895	– 0.99166

The results revealed that average temperature and percentage of bacterial reduction was highest in the May (Fig.1.7) compared to that of the remaining months.

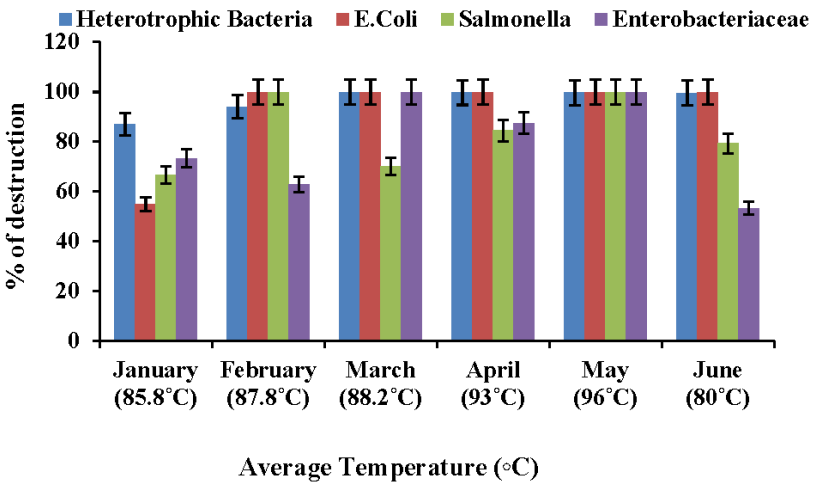

Fig.1.7. Average percentage of bacterial reduction in average temperature of different months.

4. Nutrient parameters

The concentration of the Phosphate-P, Nitrite-N, Nitrate-N and Ammonium-N were varied from 1.732 to 1.678, 1.178 to 1.164, 0.833 to 0.819 and 10.732 to 10.717 mg/L in the month of January, in February Phosphate varied from 1.933 to 1.919, 1.065 to 1.054, 0.629 to 0.617, 14.222 to 14.209 mg/L, March varied from 2.375 to 2.363, 0.985 to 0.945, 1.016 to 1.076, 13.330 to 12.979 mg/L, April were varied from 2.276 to 2.267 0.945 to 0.936, 1.076 to 1.070, 12.979 to 12.968 mg/L, May were varied from 1.990 to 1.977, 0.900 to 0.882, 1.414 to 1.404, 11.878 to 11.857 mg/L and June were varied from 1.957 to 1.945, 0.799 to 0.786, 1.386 to 1.372, 11.663 to 11.649 respectively (Table 2). In the month of January, the reduction percentage of Phosphate-P, Nitrate-N, Nitrite-N and Ammonium-N were 0.031%, 1.19%, 1.68% and 0.14%,

respectively within 4 h period of experimentation; in February the reduction percentages were0.72%, 1.03%, 1.90% and 0.009%respectively; in March 0.50%,1.005%, 0.88% and 0.09% respectively; in April were 0.39%,0.95%, 0.55% and 0.08% reduction were found. During the month of May and June, the reduction percentages were 0.65 and 0.61%, respectively reduction in Phosphate-P, 2.4 and 1.62% in Nitrate-N, 0.92 and 1% in Nitrite-N and 0.18 and 0.12% reduction in Ammonium-N were found. The results did not show any drastic changes in nutrient content of wastewater. The differences in percentage of reduction may be attributed to the process of sedimentation as solar radiation does not have any impact on this percentage of reduction.

5.Seed Germination Experiment

To conduct seed germination experiment the wastewater characteristic is essential before its exposure to solar radiation and after disinfection for its subsequent application.

5.1. *Wastewater Characteristics*

The nutrient parameters of wastewater before and after disinfection were analyzed using standard method (APHA, 2012) before its application for germinating wheat seeds (Table 1.2):

Table 1.2: Characteristics of wastewater before and after disinfection

Parameters	Initial	After 1h	After 2h	After 3h	After 4h
Phosphate-P (mg/L)	1.935	1.927	1.924	1.919	1.916
Ammonium-N (mg/L)	11.580	11.575	11.572	11.767	11.763
Nitrite-N (mg/L)	1.409	1.389	1.385	1.380	1.377
Nitrate-N (mg/L)	0.831	0.811	0.807	0.802	0.797

5.2.*Seed germination*

Root length of germinated plants ranged from 4.22cm in 1h, 4.30cm in 2h, 4.5cm in 3h and higher growth of 5cm was found in seed exposed in treated water of 4 h. Similarly, the shoot length of germinated plants ranged from 3.55cm in 1h, 3.68cm in 2h, 3.96 cm in 3h and 4.54cm in 4h (Fig.1.8). The seed germination rate varied from 90 to 100% from 1h to 4h, whereas vigour index varied from 1158.3 to 1596. Relative root growth and chlorophyll content varied from 116.28 to 133.67 and 0.259 to 0.448mg/GFW(Table 1.3).

Table 1.3: Variation of Mean ± SE values of different plant growth parameters at different time intervals during the seed germination study.

Parameters	Raw Wastewater (Control)		Disinfected wastewater							
			After 1hour		After 2hour		After 3 hour		After 4 hour	
	7 days	15 days	7 days	15 days	7 days	15 days	7 days	15 days	7 days	15 days
Root Length (cm)	1.80 ±0.001	3.86 ±0.001	3.10 ±0.001	4.22 ±0.001	3.36 ±0.001	4.30 ±0.001	3.18 ±0.001	4.5 ±0.001	3.42 ±0.001	5 ±0.001
Shoot Length (cm)	1.68 ±0.001	3.79 ±0.001	2 ±0.001	3.55 ±0.001	2.28 ±0.001	3.68 ±0.001	2.98 ±0.001	3.96 ±0.001	3 ±0.001	4.54 ±0.001
Seed Germination Rate (%)	---	85	---	90	---	100	---	100	---	100
Relative Seed Germination Rate	---	85	---	90	---	100	---	100	---	100
Vigour index	---	945.62	---	1158.3	---	1362.5	---	1411.5	---	1596
Relative Root Growth	---	89.91	---	116.28	---	116.92	---	114.06	---	133.67
Germination Index	---	94.53	---	77.39	---	85.53	---	87.67	---	90.44
Chlorophyll (mg/GFW)	---	0.198	---	0.259	---	0.340	---	0.344	---	0.448

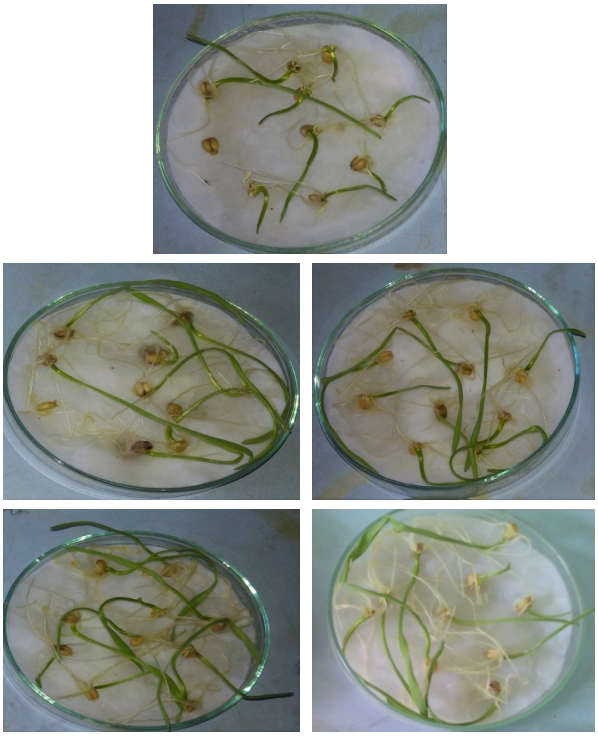

Fig.1.8. Seed germination using raw and disinfected wastewater

From the above result it was observed that germination occurred in all the treatments but the treatment with 4 h disinfected water showed the maximum growth compared to other treatments. The growth of plants was higher in the treated water compared to the plants grown in untreated water (Control).The higher growth in the plants exposed to the treated water was may be due to the lower stress factors (such as, lower microbial load, microbial extracellular products, toxic substances, etc.), where as adverse environmental conditions in control is responsible for lower plant growth. The present study revealed that raw wastewater affects seed germination due to production of various enzymes (Shukla and Pandey, 1991) or the seeds undergo physiological stress due to high salinity (Rao and Nanda Kumar, 1983) or due to excess quantities of micronutrients, heavy metals and toxic chemicals (Dollar et al., 1972; Indra and Sivaji, 2006) present. Moreover raw wastewater contains large amount of microbial and the use of such water for agricultural purpose is quite harmful (Mouhanni et al., 2011) which not only suppress the germination rate but also affects the human beings if it was consumed.

This study significantly explored the excellent use of modified SODIS method in wastewater disinfection by a box type solar cooker and its significant reuse. In many places wastewater is very frequently used for various agricultural purposes because of high nutrient contents that enhances crop productivity. Therefore, it can be concluded that modified SODIS method is low cost, environment friendly method for wastewater disinfection that uses solar energy to kill/destroy the pathogenic microbial load and various toxic pollutants, which make it safe for reuse by maintaining the EPA standards.

References

1. Acra. A., Jurdi. M., Mu'allem. H., Karahagopian. Y., Raffoul. Z. (1990), Water disinfection by solar radiation. Assessment and application.IDRC-TS66e. International Development Research Centre, Ottawa, Canada.
2. Altherr. A.M., Mosler. H.J., Tobias R., Butera. F. (2006),Attitudinal and Relational Factors Predicting the Use of Solar Water Disinfection: A Field Study in Nicaragua. Health Education & Behavior, Vol. XX (X), 1-14.
3. American Public Health Association (APHA) (1995), Standard Methods for the Examination for Water and Wastewater (19th edition). Byrd Prepess Springfield, Washington.
4. Arnon. D.I. (1949). Copper enzymes in isolated chloroplast. Polyphenoloxidase in Beta vulgaris. Plant Physiology, 24, 1-15.
5. Bitew. B.D., Gete. Y.K., Biks. G.A., Adafrie. A.A. (2018). The effect of SODIS water treatment intervention at the household level in reducing diarrheal incidence among children under 5 years of age: a cluster randomized controlled trial in Dabat district, northwest Ethiopia. Research, 19,412.
6. Berney. M., Weilenmann.H.U., Simonetti. A., Egli. T. (2006), Efficacy of solar disinfection of Escherichia coli, Shigella flexneri, Salmonella Typhimurium and Vibrio cholera. Journal of Applied Microbiology, 101(4), 828–836.
7. Bitton. C. (2011), Wastewater Microbiology, 4th edition, Wiley-Blackwell.
8. Caslake. L. F., Connolly. D.J., Menon. V., Duncason. C.M., Rojas. R., Tavakoli. J. (2004),Disinfection of Contaminated Water by Using Solar Irradiation. Applied Environmental Microbiology,70(2),1145–1150.
9. Dababneh. B.F., Shquirat. W.D., Abbassi. B.E. (2012), Coliform specific solar disinfection of treated wastewater. Polish Journal of Environmental Studies, 21(6), 1577-1581.

10. Dejung. S., Wegelin. M., Fentes. I., Amanza. G., Jarro. R., Navarro. L., Arias. G., Urquieta. E., Torrico. A., Fenandez. W., Iriarte. M., Birrer. C., Stahel. Wa. (2007), Effect of Solar Water Disinfection (SODIS)on model microorganisms under improved and field SODIS conditions. Journal of Water Supply Research and Technology, 56(4), 245.
11. Dollar. S.G., Bovlet. I.R., Keenev. A.D. (1972), Paper mill sludge disposal on soils. Effect of the yield and mineral nutrition of oats.Journal of Environmental Quality, 1, 405-409.
12. Dunlop. P.S.M., Ciavola. M., Rizzo. L., Byrne. J.A. (2011), Inactivation and injury assessment of Escherichia coli during solar and photocatalytic disinfection in LDPE bags. Chemosphere**,** 85, 1160-1166.
13. Giannakis. S., Darakas. E., Escalas-Canellas. A., Pulgarinc. C. (2014), The antagonistic and synergistic effects of temperature during solar disinfection of synthetic secondary effluent. Journal of Photochemistry and Photobiology A: Chemistry, 280, 14-26.
14. Hijnen. W.A.M., Beerendonk. E.F., Medema. G.J. (2006), Inactivation credit of UV radiation for viruses, bacteria and protozoan (oo)cycts in water: a review. Water Research, 40, 3-22.
15. Indra. V., Sivaji. S. (2006), Metals and organic components of sewage and sludges. Journal of Environmental Biology, 27, 723-725.
16. Islam. S., Akanda. M.A., Prova. A., Islam. M.T., Hossain. M.M. (2016), Isolation and identification of plant growth promoting rhizobacteria from cucumber rhizosphere and their effect on plant growth promotion and disease suppression. Frontiers in Microbiology, 6, 1360.
17. Lawrie. K., Mills. A., Figueredo-Fernández. M., Gutiérrez-alfaro. S., Manzano. M., Saladin. (2015),UV dosimetry for solar water disinfection (SODIS) carried out in different plastic bottles and bags. Sensor Actuator B Chemistry, 208, 608-615.
18. McGuigan. K.J., Conroy. R.M., Mosler. H.J., Preez. Md., Jaswa. E.U., Fernandez-Ibanez. P. (2012), Solar water disinfection (SODIS): A review from bench-top to roof-top. Journal of Hazardous Materials, 235-236, 29-46.
19. Miche. L., Balandreau. J. (2001). Effects of rice seed surface sterilization with hypochlorite on inoculated *Burkholderia vietnamiensis*. Applied Environmental Microbiology, 67(7), 3046-3052.
20. Mouhanni. H., Bendou. A., Er-Raki. S. (2011), Disinfection of Treated Wastewater

and its Reuse in the Irrigation of Golf Grass: The Case of Plant M'zar Agadir-Morocco. Water, 3, 1128-1138.

21. Navntoft. C., Ubomba-Jaswa. E., McGuigan. K.G., Fernández-Ibáñez. P. (2008), Effectiveness of solar disinfection using batch reactors with non-imaging aluminium reflectors under real conditions: natural well-water and solar light. Journal of Photochemistry and Photobiology B, 93 (3), 155–161.
22. Oates, P.M., Shanahan. P., Polz. M.F. (2003),Solar disinfection (SODIS): simulation of solar radiation for global assessment and application for point-of-use water treatment in Haiti. Water Research, 37,47–54.
23. Pandey. P.K., Kass. P.H., Soupir. M.L., Biswas. S., Singh. V.P. (2014), Contamination of water resources by pathogenic bacteria. AMB Express, 4(51), 2-16.
24. Polo-López. M.I., Fernández-Ibáñez. P., Ubomba-Jaswa. E., Navntoft. C., García-Fernández.I., Dunlop. P.S.M., Schmid. M., Byrne. J.A., Mcguigan. K.G. (2011),*Elimination of water pathogens with solar using an automated sequential batch CPC reactor.*Journal of Hazardous Materials, 196, 16-21.
25. Quelz. J.G.T., Sanchez-Roman. R.M. (2014),Efficiency of domestic wastewater solar disinfection in reactors with different colors.Water Utility Journal, 7, 35-44.
26. Rao. M.G., Nandakumar. N.V. (1983), Impact of effluent on seed germinability and chlorophyll content in *Cicer arietinum*. Pollution Research, 2, 33-37.
27. Reed. R.H. (2004),The inactivation of microbes by sunlight: solar disinfection as a water treatment process. Advances in Applied Microbiology, 54, 333–365.
28. Shukla. N., Pandey. G.S. (1991), Oxalic acid manufacturing plant waste waters. Effect on germination and seedling height in selected cereals. Journal of Environmental Biology, 12, 149-151.
29. Tian. Y., Guan. B., Zhou. D., Yu. J., Li. G., Lou. Y. (2014), Responses of seed germination, seedling growth and seed yield traits to seed pretreatment in maize (*Zea mays* L.). The Scientific World Journal, 2014, 1-8.
30. Wegelin. M., Canonica. S., Mechsner. K., Persaro. F., Metzler. A. (1994), Solar Water Disinfection: Scope of the process and analysis of radiation experiments. Journal of Water Science Research and Technology, 43(3), 154-169.
31. Zhou. C.E., Smith. J., Lam. M., Zemla. A., Dyer. M.D., Slezak. T., Mvir.D.B. (2007), A microbial database of protein toxins, virulence factors and antibiotic resistance genes for bio-defence applications. Nucleic Acids Research, 35(Database issue), D391–394.