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Vapor-induced transfer of bacteria in the absence of mechanical disturbances

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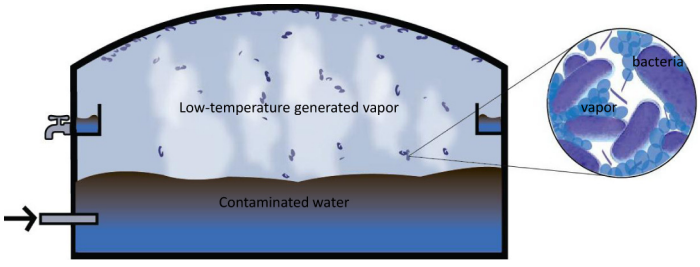
HIGHLIGHTS

- Study is first to investigate the possibility of transfer of bacteria through vapor.
- Bacteria exhibited transfer in the absence of mechanical disturbances in reactors.
- Gram positive smaller bacteria transferred more than gram negative larger bacteria.
- Transfer probability increases at optimal growth temperature of mesophilic bacteria.
- Salinity lowers bacterial survival and has synergistic effect with temperature.

GRAPHICAL ABSTRACT



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ABSTRACT

Transfer of bacteria through water vapor generated at moderate temperatures (30–50 °C) in passive solar stills, has scarcely been reported. The objective of this research was to investigate whether bacteria in highly humid atmospheres can get transferred through water vapor in the absence of other transfer media to find their way to the distillate. To achieve this objective, passive solar reactors were chosen as the medium for experimentation, and distillation experiments were conducted by spiking a pure bacterial culture (*Escherichia coli*, *Klebsiella pneumonia* or *Enterococcus faecalis*) in low mineralized water vs. highly mineralized water in the dark under moderate temperatures ranges (30–35 °C, 40–45 °C and 50–55 °C). Results showed that bacteria indeed get transferred with the vapor in stills when not exposed to solar U.V. radiation. The trends observed were adequately explained by a zero-modified Hurdle–Poisson model. The numbers of cultivable bacterial colonies transferred were bacterial size, water type and temperature dependent with highest transfers occurring in *E. faecalis* > *E. coli* > *K. pneumonia* at the 40 °C range in low mineralized water. Proper management strategies are recommended to achieve complete disinfection in solar stills.

1. Introduction

Many studies have been reported on the physical and operational aspects of solar stills with most reporting on methods which may be introduced in order to enhance the productivity of stills [1–9], while very few discussed the quality of the distillate

Abbreviations: Mdn, Median; IQR, Inter-quartile range; CI, confidence interval; CFU/100 ml, colony forming units per 100 ml of water; AIC, Akaike Information Criterion.

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resulting from these stills. In this context, contradictory results have been shown to exist in that some studies indicated that the distillate produced by a solar still was totally disinfected and fit for direct consumption [10–15] while others have reported bacteriological contamination to exist in the distillate [16–20]. Some of these studies have attributed contamination to the possibility that salinity and growth of algae and other micro-flora in the water to have affected the disinfection process in solar stills and sheltered microbial contaminants from UV radiation [18]. Others have hypothesized that bubbles, created by the turbulence of adding water to the basin, could have burst and formed droplets on the glass cover which contaminated the distillate [17]. Balladin et al. attributed their distillate bacterial findings to airborne microorganism contamination and low chloride concentration [16]; Malaeb on the other hand suspected cross contamination [19]. These inconsistent results deem to shed doubts as to the health safety of distillates resulting from solar stills.

The authors who reported the presence of bacterial contamination have not confirmed nor substantiated the sources of such contamination and the issue was left without plausible explanation. In the absence of justifications for the reasons leading to distillate contamination, the present study is initiated to explore the possible transfer and dispersal of bacteria from contaminated water subjected to moderate solar-induced temperatures via the generated vapor as a transport medium in the absence of mechanical disturbances. The study will be limited to determining any physical transfer and the related impact of some operating parameters rather than investigating the dynamics of the transfer. The results of the study should help in determining the impact of such a transfer if present and the related dangers in disease transmission and possibly help shape the management strategies of solar stills specifically and similar technologies that depend on water vapor for water treatment and reuse.

2. Materials and methods

2.1. Solar stills

Four small single-basin solar reactors were constructed at the American University of Beirut (AUB) from materials purchased locally. Each reactor mainly consists of a circular basin, a hemispherical dome and a distillate collector (Fig. 1). The basin has a diameter of 23.5 cm, an area of 433.5 cm^2 and a working capacity of 500 ml of water while each dome has a diameter of 24 cm, a volume of 3620 cm^3 and a surface area of 905 cm^2. The basin can be detached from the rest of the reactor to add or remove the contaminated water without contaminating the rest of the reactor.

2.2. Test organisms and growth conditions

Three mesophilic bacterial strains commonly found in contaminated water sources were chosen in this study: *Escherichia coli* ATCC 25922, *Klebsiella pneumoniae* ATCC 13883, and *Enterococcus faecalis* ATCC 51922. Pure cultures were acquired on inoculated agar plates from the AUB Medical Center – Beirut, Lebanon. The cultures were preserved by freezing at −80 °C. All reagents, labware and materials used in preserving the pure cultures were sterilized by autoclaving. To prepare the stock for each type of bacteria, the frozen culture was inoculated into 100 ml nutrient broth and incubated at 37 °C for 18 h to reach stationary phase. The phase of bacterial growth was determined spectrophotometrically (*E. coli*: A_{600}, 1.5; *E. faecalis*: A_{600}, 1.1; *K. pneumoniae*: A_{600}, 1.3) (DR/4000 spectrophotometer, HACH, USA). The inoculated broth was then centrifuged (Sorvall ST16, Thermo Scientific, USA) three times at 10,000 rpm for 20 min to ensure the complete removal of the growth medium and re-suspended into sterile Milli-Q grade water to form the stock solution with an approximate concentration of 10^8 CFU/ml. All types of bacteria were harvested in the same manner, with differences in the growth media used. MacConkey agar was obtained from HiMedia Laboratories (Mumbai) and used for *E. coli* and *K. pneumoniae*. Columbia Colistin Naladixic Acid (CNA) agar was obtained from Deben Diagnostics (UK) and used for *E. faecalis*; and nutrient broth from BD Difco (USA) was used for all three bacterial types. This method allowed for the detection of viable, cultivable bacteria only.

2.3. Disinfection procedure

The disinfection of the reactors was insured by a rigorous cleaning procedure of the reactors with lab-grade detergent, 70% ethanol, Milli-Q grade water then subjecting the reactors to ozonation (Microzone 300, Clear Water Tech, LLC) and artificial 254 nm U.V. light (Philips, Holland) for 30 min. Selection of the control still among the four operating stills was done in a sequential manner to check for any possible carry over contamination from the previous experiment. If the control showed any contamination, the data was discarded, the system re-disinfected and the experiment repeated.

2.4. Enumeration technique

The Miles and Misra drop count technique [21] was used for serially diluting the stock solution after which bacterial counts were calculated by averaging the number of colonies, multiplying by 50 and dividing by the dilution factor to get the number of CFU/ml. The sampling water had a bacterial population of approximately

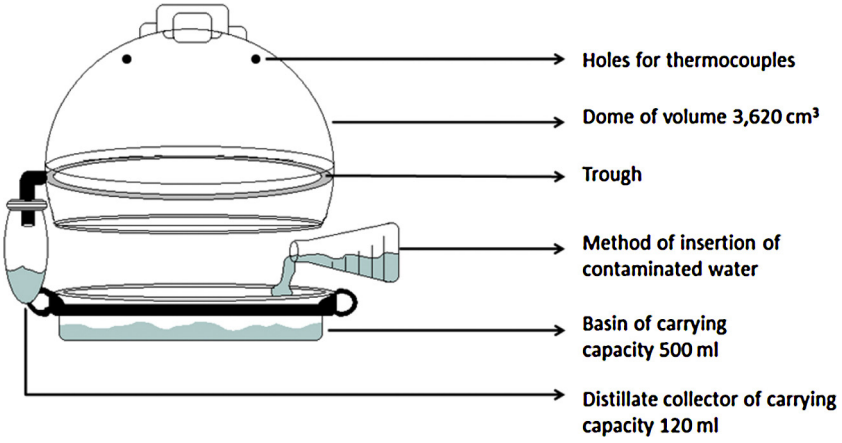

Fig. 1. Schematic of fabricated hemispherical solar stills.

10^6 CFU/ml and was prepared in a batch mode before each experiment then divided and added equally into each of the three stills while the fourth still was kept as control. Prior to each experiment, swab tests were taken from inside the dome and distillate collector of each reactor to ensure the absence of contamination in the stills. Following the termination of each experiment, which was determined to be 24 h, two grab samples were taken from the distillate collector and remaining water in the basin. Samples from solar experiments were immediately shielded from sunlight and processed directly to avoid photoinactivation. Processing of samples was performed using membrane filtration on cellulose filters (47 mm diameter, 0.4 μm pore size), followed by plating on MacConkey agar for *E. coli* and *K. pneumoniae* or Columbia CNA agar with 5% sheep blood for *E. faecalis*. As for checking for bacterial transfer to the dome of the distillation units, swab tests were taken from each reactor.

2.5. Distillation under dark/thermally heated atmosphere

Reactors in thermally-heated atmospheres were placed in a Thermo Scientific 1300 Series bio-safety cabinet to eliminate any environmental contamination. Heat was provided through heating mantles (Electrothermal, London) fitted with temperature regulators to maintain constant temperatures. Light interference was eliminated by covering the glass of the cabinet with a black cloth.

Water temperature and humid air temperature variations inside the reactors were measured by two pre-sterilized K-type thermocouples inserted at the top of each reactor. Thermocouples were attached to a Data Acquisition Module (Cole Parmer, Illinois) connected to a PC and recorded by LabView (National Instruments, Austin) at 30-min intervals for continual reading of temperature (Fig. 2). The tests were conducted by adding to the reactors a single species of bacteria of the same concentration and operated in parallel under the same parametric conditions. Three reactors with identical setups were run. Two samples from the distillate and two samples from the basin were taken per reactor. A flow diagram of the experiments is presented in Fig. 3 while experimental blocks are presented in Table 1.

Fig. 2. Setup of thermal and solar experiments.

2.6. Solar distillation under natural sunlight

For comparative purposes, six solar distillation experiments with the three bacterial strains where each strain was tested separately, were carried out on a building roof at AUB (latitude: N33°54′; longitude: E35°29′; altitude: 29 m) (Fig. 2). The solar experiments were conducted for the synergistic role of temperature and U.V. radiation on the inactivation of bacteria. Two solar experiments were conducted on *E. coli*, two using tap water and two sea water, in the months of September and October with a maximum solar irradiance of 840 W/m^2 and average of 460 W/m^2. Water temperatures are recorded in Table 2. Similarly, two solar experiments were conducted on *E. faecalis* in April, 2013 under sunny conditions with a maximum solar irradiance of 830 W/m^2 and average irradiance of 456 W/m^2. As for *K. pneumoniae*, two solar experiments were conducted in April and September under sunny conditions with

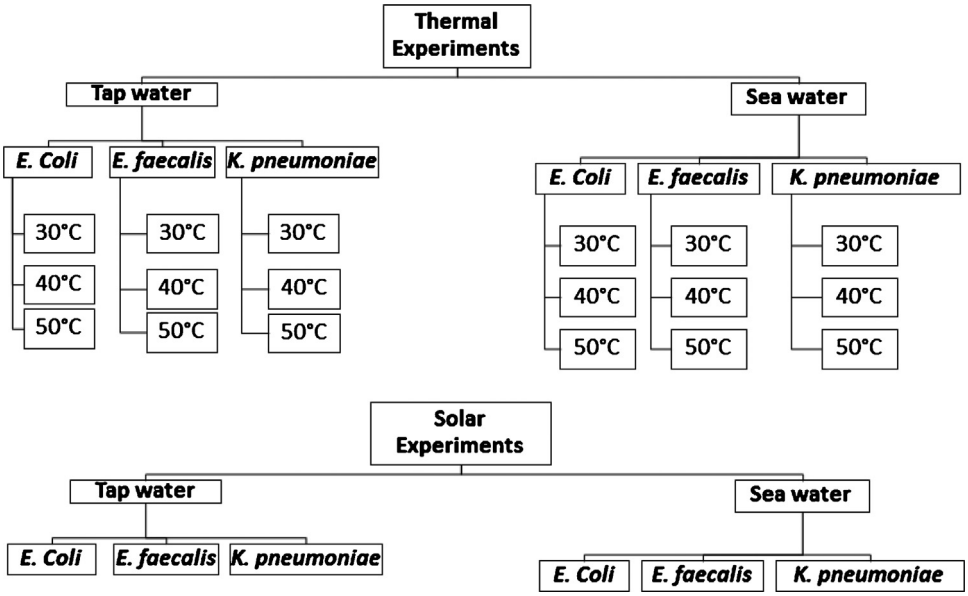

Fig. 3. Flow diagram of experiments.

Table 1
Experimental blocks for thermal experiments.

Experimental unit	Bacteria type	Water type	Water temperature (°C)	Replication
1	*E. faecalis*	Tap	30	6
2		Sea	30	6
3		Tap	40	6
4		Sea	40	6
5		Tap	50	6
6		Sea	50	6
7	*E. coli*	Tap	30	6
8		Sea	30	3
9		Tap	40	6
10		Sea	40	3
11		Tap	50	3
12		Sea	50	3
13	*K. pneumoniae*	Tap	30	9
14		Sea	30	3
15		Tap	40	6
16		Sea	40	5
17		Tap	50	3
18		Sea	50	3

a maximum and average solar irradiance of 837 and 459 W/m^2, respectively. Solar irradiance was measured with a portable CM3 pyranometer (Kipp and Zonen, The Netherlands) measuring global solar radiation in the spectral range of 305–2800 nm during the experiments.

2.7. Sampling procedure

Sea water, collected from the AUB Beach, was sampled in batch mode to ensure consistency throughout the experiments. Prior to use, samples were autoclaved. Chlorinated tap water samples were collected from the laboratory where the samples were boiled and cooled to remove any residual free chlorine. Water quality parameters were analyzed based on standard methods [22] and presented in Table 3.

2.8. Modeling

To explore the data, the Kruskal–Wallis (KW) one-way analysis of variance by rank test, the non-parametric analogue of a one-way ANOVA, was used to assess the statistical significance of bacterial transfers as a function of the single effect of bacterial type, water temperature, and the reactor water type. The Mann–Whitney (MW) post-hoc test was also conducted whenever the KW test indicated statistically significant difference between at least two groups.

As for predicting bacterial transfers, Poisson generalized linear models were chosen since they are used to model positively right-skewed count data. However, adopting a Poisson model to predict bacterial transfer in reactors can result in over-predicting or under-predicting the number of no transfers (0 CFU). As a result, a flexible two-part zero-modified Poisson–Hurdle model was used [23,24].

The Hurdle model for bacterial transfer data was fit using the pscl package [25,26] in the statistical software R [27,28]. The Hurdle model allows for zero-deflation, whenever the observed number of zeros is fewer than expected. The model structure is expressed as follows:

$$Pr(Y_i = y_i) = \begin{cases} 1 - \pi(z_i) & \text{when } Y_i = 0;\ 00\pi \le 1 \\ \pi(z_i)\dfrac{\lambda_i^{y_i} e^{-\lambda_i}}{y_i!(1 - e^{-\lambda_i})} & \mu > 0 \text{ and } y \text{ is a position integer greater than zero} \end{cases} \quad (1)$$

where π is the probability of a non-zero response (i.e. crossing the hurdle $\Pr(Y_i > 0)$) and λ is the mean of the Poisson model. Note that when $(1 - \pi)$ is less than $e^{-\lambda}$, the Poisson–Hurdle model results in zero deflation. A logistic regression is used to model $\pi(z_i)$ according to Eq. (3). Additionally, a log link is used to model the mean of the Poisson model as shown in Eq. (4). Note that x_i and z_i are $(n \times p)$ vectors of predictor variables and β^1 and β are $(p \times 1)$ regression coefficient vectors for the Hurdle and Poisson model respectively. The x_i and z_i need not be different.

$$\text{logit}(\pi(z_i)) = \ln\left(\frac{\pi(z_i)}{1 - \pi(z_i)}\right) = z_i'\beta^1 \quad (2)$$

$$\log(\lambda_i) = x_i'\beta \quad (3)$$

The response variable (Y_i) was defined as the count of the number of bacteria that were transferred in the reactor. The covariates of interest were the type of bacteria, the temperature at which transfer occurred, and the type of water (tap versus seawater). A

Table 2
Experimental blocks for solar experiments.

Experimental unit	Bacteria type	Water type	Water temp$_{Average}$ (°C)	Water temp$_{Max}$ (°C)
1	*E. faecalis*	Tap	32.6	38.9
2		Sea	32.1	38.3
3	*E. coli*	Tap	37.8	49.6
4		Sea	36.2	47.8
7	*K. pneumoniae*	Tap	38.0	49.8
8		Sea	32.5	38.5

Table 3
Quality parameters of water used in stills.

	Conductivity (μS/cm[a])	TDS (mg/l at 25 °C)	pH (at25 °C)	TSS (mg/l)	Turbidity (NTU[b])
Tap water	<1500	563 ± 250	7.42 ± 0.2	0.13	0.66 ± 0.4
Sea water	>15,000	29,000 ± 250	7.86 ± 0.2	35.8	0.84 ± 0.4

Note: results are the average of 4 samples taken throughout the study period.
[a] μS/cm: microsiemens per centimeter.
[b] NTU: nephelometric turbidity units.

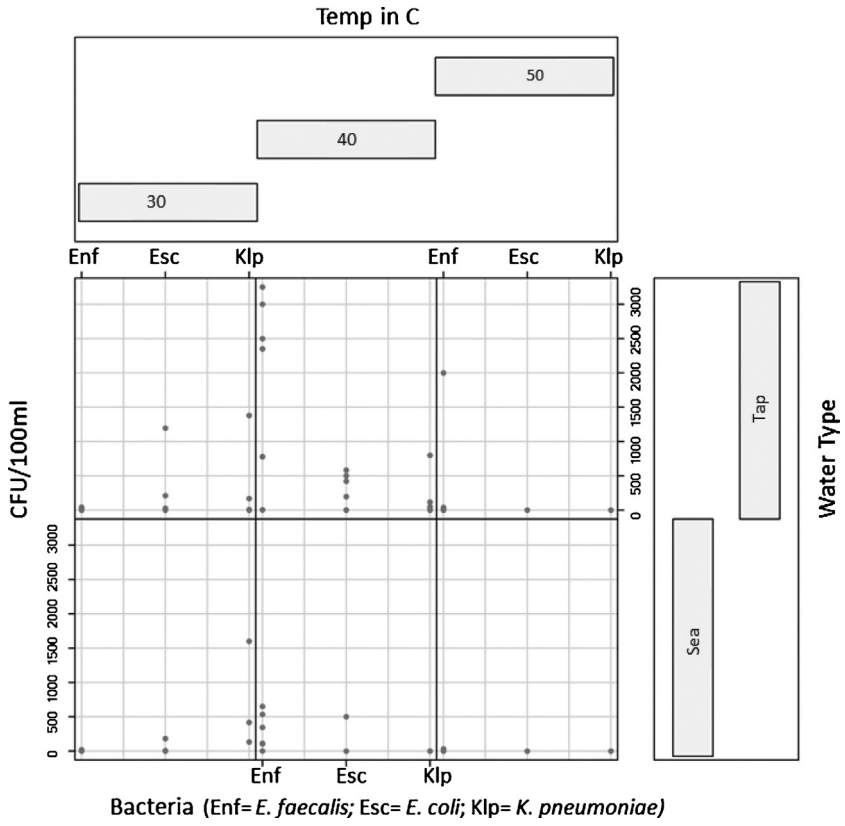

Fig. 4. Bacterial transfer measures in CFU/100 ml as a function of bacterial type, water type, and still temperature.

step-wise AIC based analysis was conducted to choose the predictor variables for both the binomial Hurdle model and the truncated Poisson model. It was found that temperature and the type of bacteria were the best predictors for predicting the possibility of transfer. Once transfer occurred, the number of bacteria transferred was best predicted by the type of bacteria, water temperature, and type of water used. As such Eqs. can be expressed as:

$$\text{logit}(\pi(z_i)) = \beta^1_{E.\,faecealis} + \beta^1_{E.\,coli} + \beta^1_{K.\,pneumoniae} + \beta^1_{40} \times I(\text{Temp} = 40\,^\circ\text{C}) \quad (4)$$

$$\log(\lambda_i) = \beta_{E.\,faecealis} + \beta_{E.\,coli} + \beta_{K.\,pneumoniae} + \beta_{40} \times I(\text{Temp} = 40\,^\circ\text{C}) + \beta_{50} \times I(\text{Temp} = 50\,^\circ\text{C}) + \beta_{\text{Tap}} \times I(\text{Water} = \text{Tap}) \quad (5)$$

3. Results and discussion

3.1. Exploratory data analysis

The solar reactor efficiency under natural sunlight was calculated based on the method adopted in [13] to be 14.5%.

$$E = \frac{Q \times h_{\text{fg}}}{A \times I} = \frac{0.02 \times 2.26 \times 10^6}{0.0434 \times 7.2 \times 101^6} = 0.145 \quad (6)$$

where E is the volumetric efficiency; Q is the average daily distilled water output under natural sunlight (l); h_{fg} is the latent heat of vaporization of water, equal to 2.26 MJ/l; A is the water surface area in the still (m^2); and I is the average daily solar insolation in Lebanon, equal to 7.2 MJ/m^2 [29].

The rates of evaporation in thermal experiments were an average of 18 ml of distillate collected when stills were kept at 30 °C, 95 ml at 40 °C, and 120 ml at 50 °C.

The effects of water temperature, water type, and bacterial type on bacterial transfer in solar reactors were assessed. A total of 18 tests were conducted. Each test had between 3 and 9 replications as shown in Table 1. The number of bacteria in the distillate was measured following the termination of each experimental unit, which was 24 hours.

It was found that bacterial transfer showed variability as a function of the physical conditions of the reactor and the bacterial culture used (Fig. 4). Bacterial transfer data are clearly not normally distributed, positively skewed, and lack equal variances across the different groups (Fig. 5).

Results showed that the number of cultivable bacterial colonies transferred in thermal experiments, expressed in CFU/100 ml, from the basin water to the distillate was highest in *E. faecalis* (Mdn = 21.1, IQR = 4.2–160.6) compared to *K. pneumoniae* (Mdn = 0, IQR = 0–50) and *E. coli* (Mdn = 1.25, IQR = 0–200) as shown in Fig. 6. The KW test results indicate that there is a significant difference (p-value = 0.043) in the number of cultivable bacterial colonies transferred as a function of bacterial type. The results from the MW post-hoc test indicate that the difference between *K. pneumoniae* and *E. faecalis* is statistically significant (p-value = 0.05). The other pairwise comparisons between the bacterial groups are not significant at the 95% CI.

With respect to the effect of reactor temperature, the KW test results indicated that the numbers of bacteria transferred were highly affected by reactor temperatures (p-value = 0.001394). MW test results indicated that the only statistically significant difference was between reactors heated to 40 °C and 50 °C (p-value = 0.0018). No statistically significant differences were observed between transfers occurring at 30 and 40 °C or between

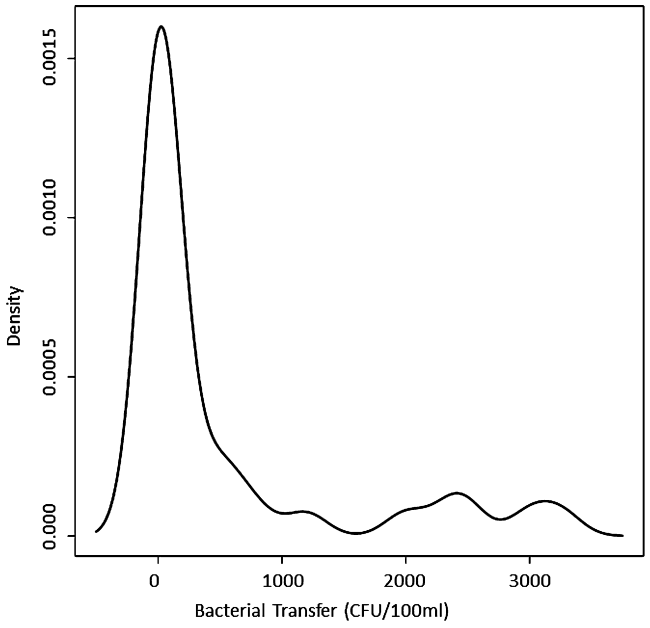

Fig. 5. Probability density function (pdf) plot of bacterial transfer across all bacterial types, still temperatures, and still water type.

30 and 50 °C. The highest number of colonies transferred originated from tap water (Mdn = 15.2, IQR = 0–203.3), as opposed to colonies transferred from sea water (Mdn = 1.68, IQR = 0–86.1). However, the effect of water type on bacterial transfer did not show statistical significance at the 95% CI ($p = 0.1057$).

3.2. Hurdle–Poisson model

The probability of observing bacterial transfer in a reactor was found to be dependent on the type of bacteria and water temperature (95% CI; p-value < 0.05). Meanwhile, the type of water used in the reactor was not found to be a significant factor (95% CI; p-value = 0.088) in predicting bacterial transfer. As such, it was dropped from the final model. Similarly, the probabilities of transfer at 30 and 50 °C were not significantly different at the 95% CI and as such were combined in the model and predicted by a single term. Table 4 compares the predicted probability of bacterial transfer to the observed probability as a function of temperature and bacterial type. The predicted probabilities are very close to those observed, indicating that the model is adequately capturing the transfer process (Fig. 6). *E. faecalis* has the highest probability of transfer of the three bacterial types at a given water temperature. Additionally,

Table 4
Predicted and observed probability of bacterial transfer as a function of bacterial type and water temperature.

Bacterial type	Temperature (in °C)	Predicted bacterial transfer probability	Observed bacterial transfer probability
Enterococcus faecalis	40	0.90	1.00
	30 and 50	0.72	0.67
Escherichia coli	40	0.73	0.67
	30 and 50	0.43	0.47
Klebsiella pneumoniae	40	0.60	0.55
	30 and 50	0.30	0.29

the transfer probability is higher at 40 °C as compared to transfer rates at 30 and 50 °C, irrespective of bacterial type.

Once transfer occurs, the number of bacteria in the distillate can be predicted by the truncated Poisson model nested within the Hurdle model. The predicted transfers of bacteria are shown in Table 3, which summarizes the predicted count data as compared to the observed counts in reactors that experienced transfer.

The effects of the water type, water temperature, and the bacterial type were all found to be significant at the 95% confidence level (p-values on all model coefficients were <0.05). Additionally, the model results indicated that there was a significant interaction between the effects of the water type and the effects of water temperatures (p-values < 0.05). Reactors filled with tap water were predicted to have the higher bacterial counts across all bacterial types, irrespective of the water temperature. The lowest predicted number of bacteria transferred occurs when the reactors were filled with seawater and heated to 50 °C. Yet, the effect of temperature on the number of bacteria transferred was found to be a function of salinity. Seawater reactors heated to 40 °C had on average slightly lower bacterial counts as compared to their counterparts that were heated to 30 °C. The ratio between seawater reactors heated to 40 °C and those heated to 30 °C was 0.85. In freshwater reactors, the opposite effect was observed; reactors heated to 40 °C tended to have on average 3.3 times more bacteria than the freshwater reactors that were heated to 30 °C. When the seawater reactors were heated to 50 °C, the bacterial counts were predicted to be much lower than those observed under 30 °C; the ratio was predicted to be 0.04. Nevertheless, the model results indicated that the effect of heating the water to 50 °C was dampened when the reactors were filled with freshwater; bacterial counts were predicted on average to be almost identical (ratio = 0.98) to the counts predicted for freshwater reactors heated to 30 °C. This indicates that the effect of the combined stress of using high salinity water that is heated to elevated temperatures is larger than the effect of any of the two stressors alone (Table 5).

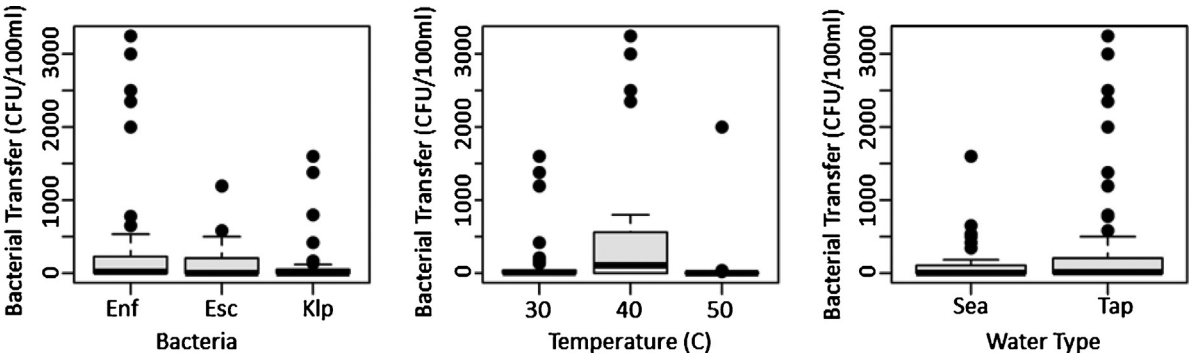

Fig. 6. Boxplot of the bacterial transfer data by different groups. Solid black lines show medians; gray boxes show IQR; extreme values within 1.5 times the IQR are the ends of the lines; observations more than 1.5 times the IQR from the median are plotted individually as black point.

Table 5
Mean predicted transfers of bacteria by the Hurdle–Poisson model.

Bacteria	Water temperature (°C)	Water type	Mean predicted transfer (CFU/ml)	Median observed (CFU/ml)	Range observed (CFU/ml)
Enterococcus faecalis	30	Tap	431.2	37.5	10.5–40.7
		Seawater	388.5	13.9	7.7–20.0
	40	Tap	1427.3	2425.0	5.0–3250.0
		Seawater	328.8	228.8	1.7–650.0
	50	Tap	420.6	34.0	9.7–2000.0
		Seawater	14.3	6.0	5.0–32.5
Escherichia coli	30	Tap	159.3	26.9	0.8–1195.0
		Seawater	143.6	93.9	6.8–181.0
	40	Tap	527.5	420.8	1.7–583.3
		Seawater	121.5	500.0	500.0–500.0
	50	Tap	155.4	NA	NA
		Seawater	5.3	NA	NA
Klebsiella pneumoniae	30	Tap	204.2	167.5	8.0–1379.31
		Seawater	184.0	416.7	133.3–1600.0
	40	Tap	675.8	50.0	8.8–800.0
		Seawater	155.7	1.7	1.7–1.7
	50	Tap	199. 2	NA	NA
		Seawater	6.8	NA	NA

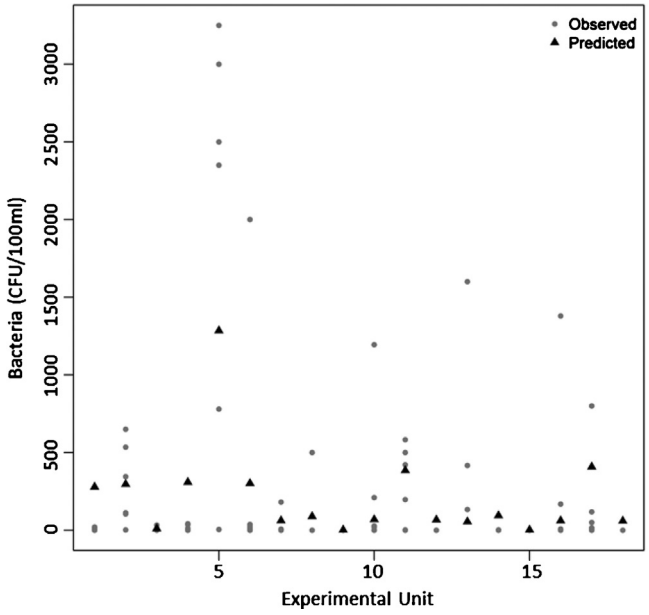

Fig. 7. Plot showing mean predicted bacterial counts by Hurdle–Poisson model.

Similar to the results in the overdispersed Poisson model, the Hurdle model results indicate that bacterial type is a major factor influencing the bacterial count in the distillate. The highest rates of transfer were observed for *E. faecalis*, while the counts of *E. coli* and *K. pneumoniae* were largely very similar. Model predicted mean bacterial counts as a function of the experimental units are shown in Table 5 and illustrated in Fig. 7. The results indicate that the model tends to capture the main trends in the experimental data and fits the data adequately well, given the large variability in the observed data. The model residuals were checked for any biases; but none were found as all residuals were centered on zero.

3.3. Thermal experiments

Thermal experiments highlighted the ability of bacteria, in the absence of solar U.V. radiation, to transfer from water to air under moderate temperatures. Swabs tests did not show any bacterial presence under all temperature ranges for the three bacterial species. Membrane filtration tests conducted on the remaining water in the basin of the reactor showed slight bacterial reduction for *E. coli* and *K. pneumonia* (0.7–2 $\log_{10}$ reduction) as compared to the original contamination levels when kept at temperatures not exceeding 45 °C. When heated to 50–55 °C, levels of the two bacterial species were significantly reduced and most often eliminated in the basin (5.5–6 $\log_{10}$ reduction). This was in accordance with the findings of Padia et al. [30]. At lower temperatures, results were consistent with those reported by Rojko [31], who reported that *E. coli* concentrations were not significantly reduced at 46 °C. Note that Pope et al. [32], who kept *E. coli* from different water sources in storage at 35 °C for 24 hours, reported variable results ranging from no significant reduction to a 95.51% reduction in cultivable colonies as tested by the membrane filtration method. Given these results it deemed expected that transfer at the 30–35 °C and 40–45 °C ranges would be higher than the transfer at higher temperatures as the experimental results have shown.

When *E. faecalis* were heated to temperatures ranging between 30 and 35 °C, microbial counts in the basin, after 24 hours, did not show any reduction. A slight reduction (1–2 $\log_{10}$ reduction) was observed at 40–45 °C. At 50 °C, a significant reduction (4.6–5.5 $\log_{10}$ reduction) was observed in the basin; yet *E. faecalis* were not completely eliminated. This was in accordance with studies that have reported *E. faecalis* to be more resistant to heat treatment than other mesophilic bacteria like *E. coli* and *K. pneumoniae* and that the stationary phase cells of *E. faecalis* can survive high pasteurization temperatures of up to 65 °C [33].

The study findings show that bacteria indeed have a mode of transfer via water vapor which promoted transfer to the distillate. The current results concur with the findings of Bonheyo et al. [34], who reported that low numbers of bacteria could be transported on water vapor across hot springs releasing steam. However, bacterial transfer in that study could have been the result of mechanical transfer mechanisms such as wind currents and cannot be definitely concluded to be the direct action of water vapor as the current study has concluded.

No study up to our knowledge has investigated whether vapor was actually capable of transporting microbial particles in solar stills. It could be explained that, in the absence of any mechanical disturbances, thermal convection currents in the water generated upon heating initiate the aerosolization process of bacteria to the water vapor particles.

3.4. Solar experiments

Results showed that, under sunlight, a starting contamination range of 10^6 CFU/ml was totally eliminated in the distillate as well

as the brine in the basins of the three bacterial species. Transfer of bacteria could be taking place under sunlight, however, transferred bacteria could be inactive and hence unable to be harvested on growth media. Since dark/thermally-driven experiments showed transfer, it can be assumed that the total absence of transfer in solar experiments is due to the synergistic effect of thermal and optical processes. McGuigan et al. [35] reported that thermal inactivation starts playing a significant role in bacterial inactivation at water temperatures exceeding 45 °C under sunny weather conditions. However, current results showed that optical inactivation could be sufficient alone even at temperatures as low as 38 °C when strong sunshine is available. This corroborates studies [10–15] who reported total bacterial inactivation in passive solar reactors under strong sunshine. However, these studies, as well as the present study, only tested inactivation under favorable sunny weather conditions. Differences in weather conditions, along with other factors like variations in latitude and season, could explain why some studies reported less than complete bacterial inactivation [16,17,20] while others reported total bacterial elimination under sunlight.

4. Conclusions

The present research targeted the transfer of bacteria to the distillate by three passive single basin solar reactors in the presence and absence of solar U.V. radiation. Experiments were repeated with changes in the bacterial species, water type and water temperatures. Results showed that under natural sunlight conditions, the synergistic effect of solar U.V. radiation and temperature plays an effective role in inactivating the three fecal bacterial species *E. coli*, *E. faecalis* and *K. pneumoniae* after the evaporation/condensation cycles within the solar reactors while transfer of bacteria to the distillate was recorded in thermally heated reactors in the absence of U.V. radiation.

The trends observed in the experimental reactors that were not exposed to solar radiation were adequately explained by a two-part zero-modified HP model. A number of significant findings can be deduced from the HP model:

- Low-temperature evaporation (30–50 °C) in the absence of mechanical disturbances and UV radiation could lead to the transfer of bacteria through vapor from contaminated raw water in the evaporation basin to the distillate in a solar still.
- The only plausible medium of transfer of bacteria into the distillate of the solar still is the vapor.
- Bacterial transfer was significantly affected by bacterial type with gram positive, smaller bacteria (*E. faecalis*) transferring on average more than gram negative, larger bacteria (*E. coli* and *K. pneumoniae*).
- As the temperature in the water approaches the optimal growth temperature of mesophilic bacteria, i.e. 37 °C, the probability of transfer increases. Hence, transfer at the 40 °C range was found to be significantly higher than transfer at other ranges.
- Salinity in seawater significantly lowers bacterial counts in the distillate and plays a synergistic interaction effect with increased temperature on bacteria.

Based on the findings of the study, the following recommendations can be drawn for the management and use of solar stills:

- To ensure that complete inactivation of bacteria in the solar still is achieved, all parts of the still should be exposed to solar radiation and introduction of the raw water to the still should be performed during sunshine hours to give ample exposure of the added water to UV radiation.
- Complete disinfection of solar still distillate should not be preassumed and post disinfection of the distillate should invariably be carried before ingestion.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jhazmat.2014.08.003.

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