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Escherichia coli survival in waters: Temperature dependence

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ABSTRACT

Knowing the survival rates of water-borne *Escherichia coli* is important in evaluating microbial contamination and making appropriate management decisions. *E. coli* survival rates are dependent on temperature, a dependency that is routinely expressed using an analogue of the Q_{10} model. This suggestion was made 34 years ago based on 20 survival curves taken from published literature, but has not been revisited since then. The objective of this study was to re-evaluate the accuracy of the Q_{10} equation, utilizing data accumulated since 1978. We assembled a database of 450 *E. coli* survival datasets from 70 peer-reviewed papers. We then focused on the 170 curves taken from experiments that were performed in the laboratory under dark conditions to exclude the effects of sunlight and other field factors that could cause additional variability in results. All datasets were tabulated dependencies "log concentration vs. time." There were three major patterns of inactivation: about half of the datasets had a section of fast log-linear inactivation followed by a section of slow log-linear inactivation; about a quarter of the datasets had a lag period followed by log-linear inactivation; and the remaining quarter were approximately linear throughout. First-order inactivation rate constants were calculated from the linear sections of all survival curves and the data grouped by water sources, including waters of agricultural origin, pristine water sources, groundwater and wells, lakes and reservoirs, rivers and streams, estuaries and seawater, and wastewater. Dependency of *E. coli* inactivation rates on temperature varied among the water sources. There was a significant difference in inactivation rate values at the reference temperature between rivers and agricultural waters, wastewaters and agricultural waters, rivers and lakes, and wastewater and lakes. At specific sites, the Q_{10} equation was more accurate in rivers and coastal waters than in lakes making the value of the Q_{10} coefficient appear to be site-specific. Results of this work indicate possible sources of uncertainty to be accounted for in watershed-scale microbial water quality modeling.

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1. Introduction

Escherichia coli is commonly used to indicate the presence of fecal contamination in water and, by extension, the possible presence of pathogenic microorganisms. Although the vast majority of *E. coli* strains are non-pathogenic, a strong correlation has been observed between elevated levels of *E. coli* in recreational water and occurrences of gastrointestinal disease (U.S. EPA, 1986). Surface waters and, to some extent, groundwater serve as both habitats and dissemination pathways for

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bacteria. Therefore, knowing *E. coli* survival rates is important for assessing the severity of contamination that has occurred and making appropriate management decisions. Existing fate and transport predictive models, such as SWAT (Gassman et al., 2007) and HSPF (Bicknell et al., 1997) account for *E. coli* survival by estimating survival rates.

E. coli survival rates are known to be dependent on temperature (Faust et al., 1975; Vasconcelos and Swartz, 1976; Flint, 1987; Craig et al., 2004; Jamieson et al., 2004). A multitude of other environmental factors including pH (Presser et al., 1998), salinity (Bordalo et al., 2002), predation (McCambridge and McMeekin, 1980), streambed resuspension (Harmel et al., 2010), and sunlight intensity (Davies and Evison, 1991; Sinton et al., 2002) have also been found to affect *E. coli* survival in waters; temperature, however, is usually viewed as the major factor (Mancini, 1978; Flint, 1987).

The Q_{10} equation is often used to express the dependence of biological rates on temperature (Reyes et al., 2008). Applying this equation usually assumes applicability of the Chick law, i.e., the first-order exponential die-off model (Chick, 1908). The Q_{10} temperature coefficient is used to describe fractional change in a biological rate in accordance with a 10 °C change in temperature, since a given increment of temperature change affects a reaction rate by a constant factor (Johnson and Thornley, 1985). An analogue of the Q_{10} equation was used by Mancini (1978) to obtain a single value for the temperature coefficient, based on 20 published papers documenting coliform mortality rates at various temperatures in various surface waters. Since then, many *E. coli* survival studies have been performed; however, information from these studies has not been analyzed using the Q_{10} equation or the analogue proposed in the Mancini (1978) model. The latter is being used in microbiological water quality modeling (e.g. Parajuli, 2007; Coffey et al., 2010) without modifications.

The objective of this study was to evaluate the accuracy of the Q_{10} equation as a model for the dependence of *E. coli* survival on temperature in waters from various sources, utilizing the majority of published data.

2. Materials and methods

2.1. Experimental data

Data was extracted from 70 peer-reviewed papers on *E. coli* survival in water from various sources to assemble a database of 450 survival datasets in coordinates "time vs. log concentration." Additional database information corresponding to each dataset included author name(s), year of publication, *E. coli* strain (if provided), water source name as given by the authors, approximate inoculation concentration (if provided), temperature, enumeration technique, and duration of the experiment. The general characterization of the database is provided in the Supplemental data.

In this study we focused on results of laboratory experiments performed in the dark to exclude effects of sunlight and variations in predation and nutrient contents. This criterion provided a database of almost 170 datasets, which was over eight times larger than the one used by Mancini (1978). *E. coli* enumeration was done using growth media; therefore, the decrease in *E. coli* concentration is referred to below as inactivation rather than as die-off. The database was categorized by groups – those containing waters of agricultural origin (runoff, effluent ponds, catfish ponds), pristine water sources (caves, springs), groundwater and wells, lakes and reservoirs, rivers and streams, estuaries and seawater, and wastewater.

2.2. The Q_{10} equation and related models of temperature dependencies of bacteria die-off and inactivation in waters

Temperature dependencies were established based on the first-order inactivation rates k_c, as defined by the Chick equation (Chick, 1908):

$$C = C_0 e^{-k_c t} \tag{1}$$

which, after taking logarithms, becomes

$$\ln C = \ln C_0 - k_c t \tag{2}$$

Here and below, t is time (days). We used decimal logarithms to convert (2) into

$$\log C = \log C_0 - kt \tag{3}$$

so the rate constant k would be related to k_c simply as $k = k_c \times \log(e) = k_c \times 0.4342$.

In biological studies, the relationship between k and temperature is most commonly defined via the Q_{10} temperature coefficient (Behradek, 1930) as

$$\frac{k}{k_*} = Q_{10}^{(T_C - T_{C,*})/10} \tag{4}$$

here, Q_{10} is a measure of the rate of change in the inactivation rate as a consequence of increases in temperature by 10 °C increments, where $T_{C,*}$ is the reference temperature, °C, k_* is the inactivation rate at the reference temperature, k is the inactivation rate at Celsius temperature T_C, and k and k_* have the same units, which are day^{-1}.

Water quality models use a slightly different formulation of the temperature dependence in the form proposed by Mancini (1978):

$$\frac{k}{k_*} = \theta^{T_C - T_{C,*}} \tag{5}$$

where T_C is Celsius temperature, $T_{C,*}$ is the reference temperature (often taken equal to 20 °C), k_* is the first-order inactivation constant at temperature $T_{C,*}$, and θ is the temperature sensitivity parameter.

It is easy to see that parameters θ and Q_{10} are related:

$$\theta = Q_{10}^{1/10} \tag{6}$$

The Arrhenius equation is used to describe the dependence of first-order reaction rates on temperature in the form

$$k = Ae^{(-E_a/RT)} \tag{7}$$

where A is the constant pre-factor, E_a is the activation energy, R is the gas constant, T is the temperature in Kelvin, and k is the inactivation rate at the temperature T °K. Both (4) and (5) are approximations of (7) (see Appendix A).

Several authors have reported decimation time, or decimal reduction time, t_{90}, i.e., time needed to diminish the initial

population by tenfold, at different temperatures. As shown in Appendix A, if (1) and (4) are applicable, the logarithm of the decimation time should depend on temperature linearly and the slope of this dependence is related to Q_{10}.

Values of Q_{10} and k_* were converted by logarithmic transformation from (4) to the form

$$\log k = \log k_* + \frac{1}{10}\log Q_{10}(T_C - T_{C,*}) \quad (8)$$

and by applying the nonlinear regression fitting algorithm in the SigmaPlot 11 software (Systat Software, San Jose, CA) to the experimental dependencies of k on temperature. The value of $T_{C,*}$ was set to 20 °C, and the nonlinear fit was used to estimate both average and standard errors of Q_{10} and k_{20}. Hypotheses about equality of average values of Q_{10} between different data groups were tested, assuming normal distribution of the estimated average and using the t-test with squared standard errors as estimates of variance of the averages with the TINV function in MS Excel. Significance was assessed at the 0.05 level for two-tailed distributions.

2.3. *Determination of the inactivation rates*

Each survival dataset was digitized from its original publication with the mapping software Surfer8 (Golden Software, Golden, CO). The digitized data were transferred to SigmaPlot 11 (Systat Software, San Jose, CA) for plotting and analysis.

Inspection of the datasets showed generally four different ways that the dependencies of logarithms of concentrations on time were shaped. Fig. 1 shows the schematics we adopted to characterize these inactivation patterns. Type 1 refers to data that are approximately linear throughout the whole range of observation times. Type 2 refers to data exhibiting a fast decrease in population until the break time, t_b, after which the slope drastically decreases or becomes close to zero. The data collected after t_b is referred to as the "tail." Type 3 refers to data exhibiting an approximately linear dependence of Log C on time after some t_0 substantially greater than 0. The term "shoulder" describes the part of the dataset between experiment start time and t_0. Finally, Type 4 refers to data with a combination of "shoulder" and "tail" characteristics. The first-order inactivation rate constants were calculated from all data for datasets of Type 1, from data between start time and t_b for the datasets of Type 2, from data after t_0 for Type 3, and for data between t_0 and t_b for Type 4.

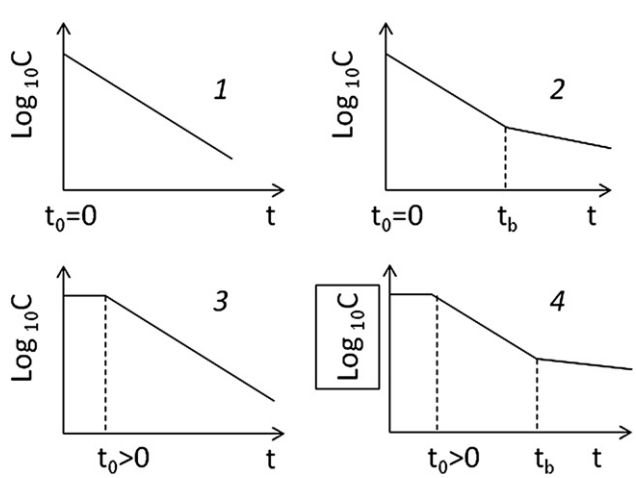

Fig. 1 – Patterns found in data on *E. coli* inactivation in waters.

Values for time zero (t_0) and break time (t_b) had to be determined to establish the range of points that made up the approximately linear sections in datasets to be used in computing inactivation rates. To find these values, we applied the method of sequential linear fitting (Dathe et al., 2001). To find the breakpoint t_b for datasets of Type 2, the determination coefficient R^2 of the linear regression 't – log C' was initially computed for the first three experimental points, then for four points, then for five points, etc., until all points were used in computation. The set of points with the highest value of the determination coefficient R^2 was used to determine k. Similarly, to find the t_0 on datasets of Type 3, we computed determination coefficients of the linear regression, 't – log C,' and time beginning from the last three points, including more and more points toward the start time. The point where the determination coefficient reached maximum, t_0, indicated the starting point for the dataset section from which the k value was found for Type 3 datasets. The method of sequential linear fitting was not applicable to all points in observation for the Type 4 datasets because of the presence of a shoulder and a tail, so visual inspection was applied to select t_0. All points occurring before t_0 were eliminated and sequential linear fitting was applied to these shortened datasets to determine t_b for Type 4 datasets.

An example of applying this algorithm for a Type 2 dataset is shown in Fig. 2. The maximum determination coefficient is achieved when correlation is computed between log C and time for points 1–5. The graph shows that the linear fit seems

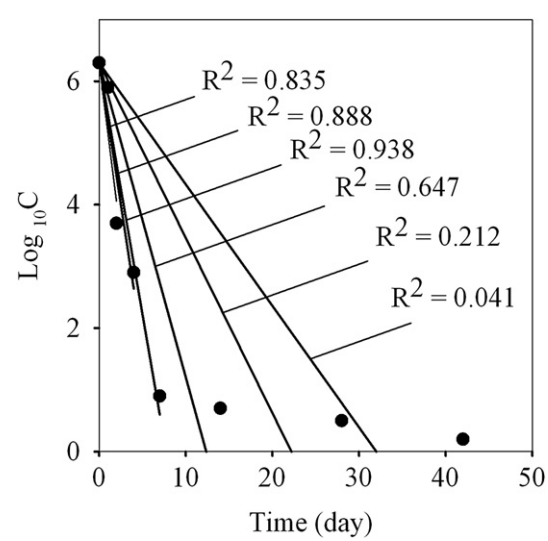

Fig. 2 – An illustration of the sequential linear fitting used to find the first-order die-off section of a type 2 dataset by selecting the number of points included in the linear regression computation that yields the greatest value for the determination coefficient R^2. In this case, the first 5 points provide the best fit since $R^2 = 0.938$, and points from first through fifth constitute the approximately linear section.

to be applicable for the first five points, but deterioration of the fit is encountered as more than five points are included in the regression; this deterioration is reflected by the decrease in R^2.

3. Results

3.1. Occurrence of different inactivation patterns

The observed frequencies of the inactivation patterns were as follows: Type 2 was found most often – in 46% of all cases; Type 1 and Type 3 were encountered 25% and 27% of the time, respectively; and Type 4 was found in 2% of all cases. The prevalence of Type 2 is notable.

Initial concentrations and linearity ranges in the experiments are shown in Fig. 3. The logarithms of initial concentrations expressed in CFU/ml, MPN/ml, or cells/ml were most often between 5.5 and 8. Ranges in logarithms of concentrations for the approximately linear section of inactivation in datasets were most often between 2 and 6. A weak relationship was observed between initial concentrations and concentrations in the end of the linear section – the determination coefficient of the linear regression was only 0.22. Subsetting data of Type 2 resulted in distributions of start concentrations and linearity ranges (Fig. 3b, d) that were similar to distributions observed for the whole database (Fig. 3a, c).

The determination coefficients and root-mean-square-errors obtained from all points in observations of Type 2, 3, and 4 datasets were compared to linear fits of the approximately linear sections of those datasets (Fig. 4). Linear fit of all points in observations of Type 2, 3, and 4 were, on average, $R^2 = 0.628$ and RMSE = 0.952. Linear fit of the approximately linear sections of those datasets provided much better accuracy: values of R^2 and RMSE were 0.901 and 0.439, respectively.

3.2. Temperature effect on inactivation rates for the broad groups of datasets

Inactivation rate constants were very small in pristine waters; moderate in agricultural waters and lake waters; moderate-to-large in estuary waters and groundwater; and large in river water and wastewater (Fig. 5). Results of the regression analysis are presented in Table 1. The values of Q_{10} varied from a minimum of 1.36 ± 0.24 in wastewater to a maximum of 2.07 ± 0.19 in pristine water. Fig. 5 and Table 1 show that the Q_{10} equation can simulate the trends in survival rate constants as the temperature changes, although R^2 values are low.

There were no significant differences ($p \leq 0.05$, two-tailed t-test) between estimates of Q_{10} for different groups. On the contrary, however, significant differences were encountered between k_{20} values of pristine water and all other water sources. Additionally, there was a significant difference in k_{20} values between rivers and agricultural waters, wastewaters and agricultural waters, rivers and lakes, and wastewater and lakes.

3.3. Site-specific applications of the Q_{10} equation

Inactivation rates at three or more temperatures were studied at some sites. Applicability of the Q_{10} equation at those sites is illustrated in Fig. 6 and complementary Table 2. The Q_{10} equation fit site-specific data better than data from broad groups. Results of the regression analysis for site-specific cases are presented in Table 2. Changes in inactivation rates with temperature at specific sites generally fit well with the

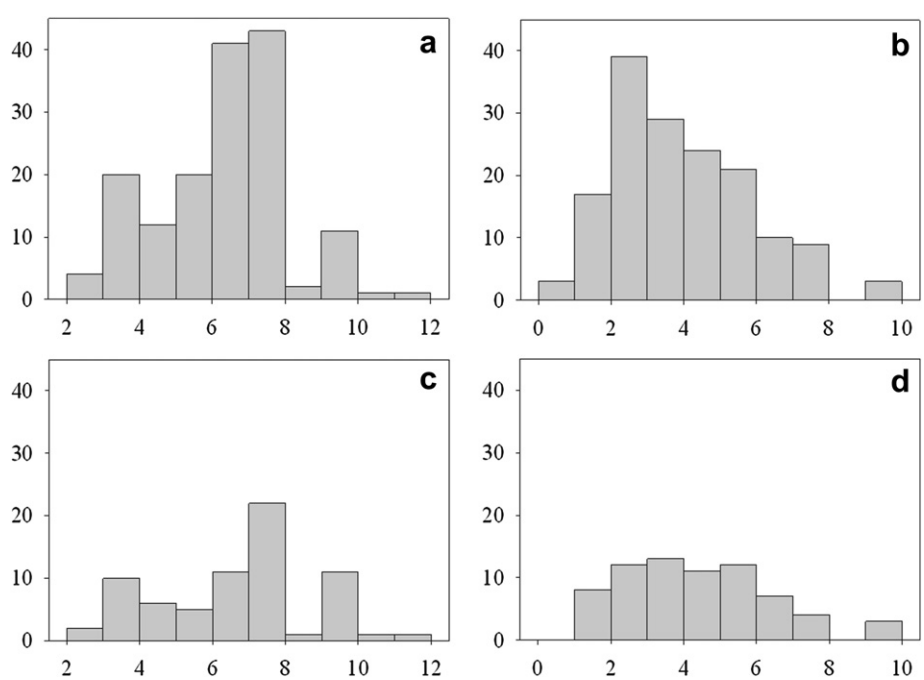

Fig. 3 – Frequency distributions of initial concentrations and ranges of inactivation in *E. coli* survival experiments: (a) initial concentrations for all experiments, (b) range of concentrations for all experiments; (c) Initial concentrations for datasets with the type 2 inactivation pattern; (d) Concentration ranges along the first linear section for the type 2 inactivation pattern.

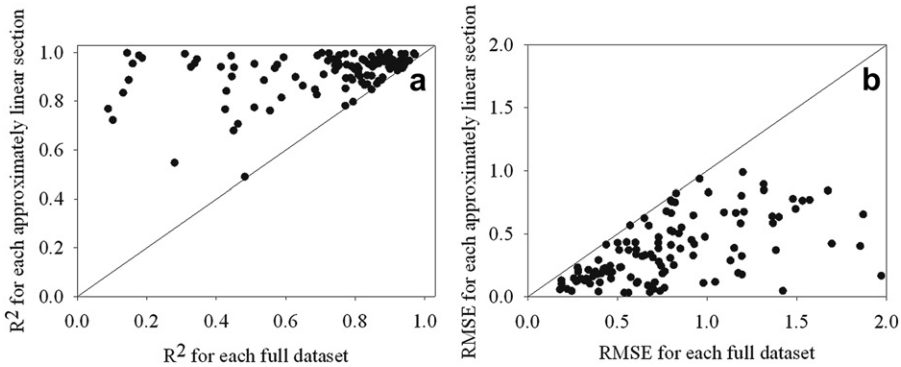

Fig. 4 – Accuracy of linear approximation of all points in observation for type 2, 3, and 4 inactivation datasets as opposed to linear approximation only of the approximately linear sections in these datasets: (a) determination coefficients of linear regressions and (b) root-mean-squared errors.

Q_{10} equation, however, the value of Q_{10} constants appeared to be site-specific. The Q_{10} values varied widely in each category of waters, and were mostly smaller than 2. Values of k_{20} were substantially smaller in lakes than in rivers and coastal water.

4. Discussion

Type 2 inactivation patterns, representing fast inactivation followed by slow inactivation, were the most prevalent in our dataset. Mancini (1978) made a similar observation on coliform inactivation, reporting that mortality rates commonly decreased as population decreased. Type 2 patterns may originate from the fact that when protozoa coexist with various sources of prey, active predation is exercised over the most abundant population and others remain as alternative prey (Danso and Alexander, 1975). As the *E. coli* population is depleted, it ceases to be the preferred prey and its inactivation slows. If that is mostly true, there could be dependence between the initial concentration and concentration at the end of the fast inactivation stage. We have not observed such dependence in our database (data not shown), but this could be caused by the general heterogeneity of data. Another explanation for change in the inactivation rate is that different predator groups prey on coliforms during different periods within the inactivation observation time (Brettar and Höfle, 1992; McCambridge and McMeekin, 1980). Factors other than predation can also be responsible for the inactivation rate change. In particular, Easton et al. (2005) proposed that the pattern of leveling off toward some equilibrium level with

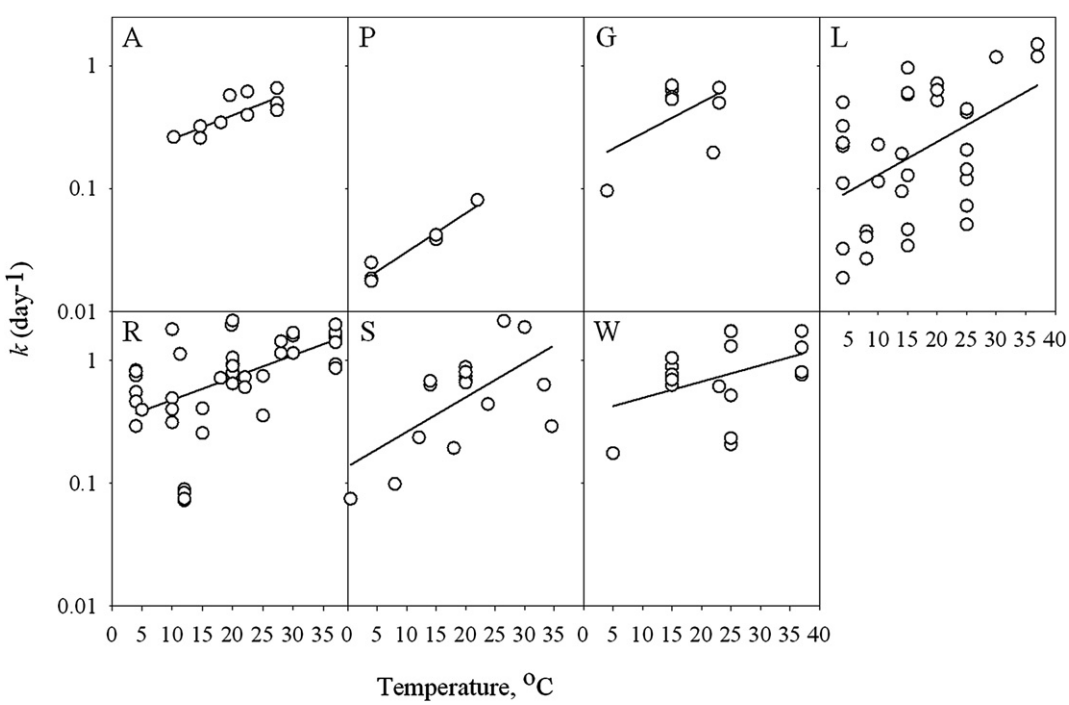

Fig. 5 – Dependencies of *E. coli* inactivation rates on temperature, grouped by water types; A, agricultural waters; P, pristine waters; G, groundwater; L, lakes and reservoirs; R, rivers and streams; S, seawater and estuaries; W, wastewater.

Table 1 – Regression analysis of datasets for different water sources.

Water source	Q_{10}	k_{20}, day^{-1}	R^2
Agricultural	1.548 ± 0.161	0.388 ± 0.024	0.640
Pristine	2.066 ± 0.190	0.063 ± 0.007	0.939
Groundwater	1.783 ± 0.702	0.504 ± 0.136	0.265
Lake	1.863 ± 0.375	0.241 ± 0.050	0.236
River	1.520 ± 0.154	0.725 ± 0.078	0.271
Sea, coastal	1.912 ± 0.411	0.501 ± 0.094	0.393
Wastewater	1.358 ± 0.238	0.672 ± 0.114	0.160

The "±" sign separates estimates of the average value of a parameter and its standard error.

increasing time may have two possible reasons: (a) rapid rate inactivation continues until the carrying capacity of the environment is reached, and populations are then maintained at that level because they are supported by the nutrients present; or (b) these studies may show evidence of quorum-sensing by microorganisms, i.e., the microorganisms are preprogrammed to die-off by two or three log units and maintain that level. Genetic differences can be also manifested during the inactivation observation time, such that subpopulations with different survival capabilities dominate the total generic *E. coli* population during different periods of the total period of observations (Velz, 1984). Finally, bacterial prey may also persist due to the development of predation-resistant organisms and evolution of avoidance strategies (McCambridge and McMeekin, 1980).

The substantial presence of Type 3 inactivation patterns also can be interpreted as the result of interactions between *E. coli* and predator populations. Barcina et al. (1986a) observed shoulders on *E. coli* survival graphs until the number of predators reaches maximum, and afterward the *E. coli* population and heterotrophic community started to decrease. The shoulder, or lag phase in this case, was interpreted as a result of a dynamic balance between growth and death of the *E. coli* population due to predator grazing. Another reason for the appearance of the observed, apparent lag period could be the presence of non-viable *E. coli* cells in the inoculum used. This could explain Barcina et al.'s (1986a) observation that the count of *E. coli* remained constant while the heterotrophic community and the protozoa population increased. We note that patterns found in wastewater datasets mostly were of Type 3 in our database. The growth of enteric bacteria with adequate nutrients present at increasing temperatures has been noted (Lessard and Sieburth, 1983) and related to improved *E. coli* survival (Ravva and Korn, 2007).

Grouping of waters in this work followed earlier observations that *E. coli* inactivation patterns within the same category of waters were similar; for example, the closeness of the *E. coli* inactivation rates for estuaries was noted by Faust et al. (1975). We found, however, that *E. coli* survival rates within a category of waters were highly variable at the same temperature (Fig. 3); variations in biological and physical survival factors could be the reason. The microbial ecosystem composition can affect *E. coli* survival. Also, the nutritional value of added *E. coli* for eukaryotic predators is higher than wild or indigenous prey (Barcina et al., 1986a) and predators such as ciliates, for example, can select prey in terms of its nutritional value (Dive, 1973). Noble et al. (2004) compared *E. coli* inactivation rates in mesooligotrophic and eutrophic waters and noted that enzymatic degradation, secondary production, and grazing occurred at lower rates in meso-oligotrophic waters, which may have caused lower rates of inactivation or removal of allochthonous bacteria. The indirect effect of water biota on *E. coli* survival was demonstrated by Gray (1975) who suggested that the numbers of *E. coli* in natural waters may sometimes be determined by the content of dissolved carbon dioxide, which itself is affected by the volume of photosynthesizing green cells. Sampson et al. (2006) reported that the presence of sand in a lake water microcosm increased the time of *E. coli* survival. They suggested that the presence of solid particulate material or green algae could affect *E. coli* survival in microcosms by providing protection or a site of higher nutrient concentrations. Although temperature-dependence on *E. coli* survival rates from the same water sources varied, substantial

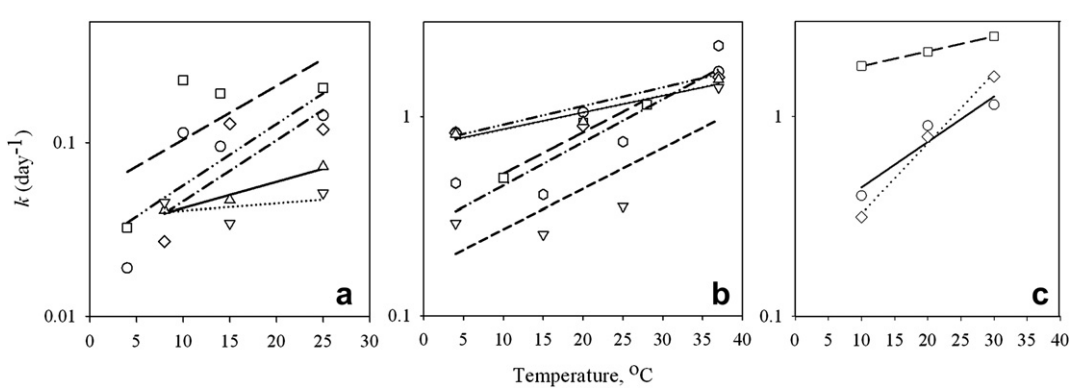

Fig. 6 – *E. coli* inactivation rates at three or more temperatures at specific sites; (a) lakes and reservoirs: Lake Winnebago, WI, USA (with sand) (○--); Lake Winnebago, WI, USA (no sand) (□--); Lake Jackson, GA, USA (◇--); Lake Herrick, GA, USA (△–); Reservoir, Griffin, GA, USA (▽---). (b) Rivers: Butron River, Spain (□--); Missouri River, MO, USA (CFU) (○--); Missouri River, MO, USA (MPN estimate) (◇---); Missouri River, USA (DVC) (△–); River Sowe, UK (above effluent outfall) (○--); River Sowe, UK (below effluent outfall) (▽---). (c) Coastal waters: Onkaparinga Estuary, Australia (○–); Henley Beach, Australia (□--); Port Adelaide, Australia (◇---).

Table 2 – Regression analysis results for datasets for site-specific studies.

Water source	Q_{10}	k_{20}, day^{-1}	R^2	Reference
Lakes				
Lake Winnebago, WI, USA (with sand)	2.261 ± 1.030	0.128 ± 0.060	0.616	Sampson et al., 2006
Lake Winnebago, WI, USA (no sand)	2.043 ± 1.124	0.212 ± 0.119	0.457	Sampson et al., 2006
Lake Jackson, GA, USA	2.247 ± 1.430	0.103 ± 0.053	0.618	Wang and Doyle, 1998
Lake Herrick, GA, USA	1.415 ± 0.100	0.056 ± 0.003	0.960	Wang and Doyle, 1998
Reservoir Griffin, GA, USA	1.101 ± 0.239	0.045 ± 0.008	0.163	Wang and Doyle, 1998
Rivers				
Butron River, Spain	1.612 ± 0.172	0.834 ± 0.066	0.953	Barcina et al., 1986b
Missouri River, MO, USA[a]	1.241 ± 0.047	1.131 ± 0.057	0.971	Bogosian et al., 1996
Missouri River, MO, USA[b]	1.217 ± 0.099	1.049 ± 0.115	0.854	Bogosian et al., 1996
Missouri River, MO, USA[c]	1.213 ± 0.070	1.051 ± 0.081	0.919	Bogosian et al., 1996
River Sowe, UK (above effluent outfall)	1.646 ± 0.288	0.734 ± 0.159	0.802	Flint, 1987
River Sowe, UK (below effluent outfall)	1.600 ± 0.342	0.434 ± 0.113	0.707	Flint, 1987
Coastal waters				
Onkaparing Estuary, Australia	1.691 ± 0.278	0.746 ± 0.100	0.911	Craig et al., 2004
Henley Beach, Australia	1.188 ± 0.007	2.124 ± 0.009	0.999	Craig et al., 2004
Port Adelaide, Australia	2.253 ± 0.150	0.795 ± 0.040	0.993	Craig et al., 2004

The "±" sign separates estimates of the average value of a parameter and its standard error.
a CFU/ml on eosin methylene blue agar.
b Cells/ml with MPN estimate using lysogeny broth (LB) medium.
c Cells/ml with direct viable count.

differences between groups could still be observed (Table 1) – inactivation in rivers and wastewater was faster than in lakes and clean water sources.

Bacteria enumeration techniques differed among the experiments cited in this work. The MPN estimates were determined with both Colilert and Colisure media, while CFU counts were performed using one of the following: EMB agar, LB agar, mFC agar, lauryl sulphate agar, oxoid nutrient agar, lactose agar, blood heart infusion agar, MacConkey agar, Endo agar, and Chromcult agar. Available nutrients differ for each medium which might affect the detection capacity of *E. coli*, especially in non-pure cultures where *E. coli* must compete with other microbes for nutrients. *E. coli* survival rates in Missouri river water (Table 2) obtained with three different media were not statistically different ($p < 0.05$), however, this result cannot be generalized. The selection of the enumeration medium significantly affected the estimated survival rates of *Listeria innocua* due to the sensitivity of stressed bacteria to selective compounds (Miller et al., 2009). If this is also applicable to *E. coli*, differences in enumeration methods could influence the observed variability we have seen in *E. coli* survival rates in experiments from the same categories of waters.

Site-specific Q_{10} equations were more accurate than Q_{10} equations for broad water categories in terms of the R^2 values in regressions (compare Tables 1 and 2). The site-specific Q_{10} constants had $R^2 > 0.7$ for rivers and coastal waters, whereas R^2 values were much smaller with lakes and reservoirs data (Table 2). The Q_{10} or the Arrhenius equation with a single set of parameters may not necessarily be applicable over the wide range of temperatures from 4 or 5 °C to 35 or 37 °C that is usually considered. *E. coli* inactivation is a result of the complex interaction of factors, with the influence of each generally having its own temperature-dependence. Grazing by zooplankton and microplankton, toxic algal products and other toxins, along with various chemical and physical factors are commonly mentioned as reasons for decreased *E. coli* numbers in untreated surface waters (Klein and Alexander, 1986; Amy and Hiatt, 1989; Barcina et al., 1997). Barcina et al. (1986a) noted that besides an intrinsic influence of the temperature on the death rate of *E. coli*, there is an indirect effect of temperature on *E. coli* die-off through natural microflora of the water. McCambridge and McMeekin (1980) showed that bacterial decline depended on the presence of both bacterial and protozoan predators, the latter having a temperature optimum of 15–20 °C and the former becoming more important as the incubation temperature increased. Enzinger and Cooper (1976) suggested that eukaryotic organisms (protozoan predators) are more effective in removing *E. coli* than prokaryotic organisms. Besides, different groups of predators often prey on coliforms during different periods within the inactivation observation time (Brettar and Höfle, 1992) and the dependencies of those groups on temperature may not coincide. Also, *E. coli* metabolism can undergo qualitative changes as temperature decreases. In cold, low temperature environments, *E. coli* metabolism is drastically slowed and their affinity for substrate becomes lower. When the temperature becomes low enough (around 5 °C) *E. coli* can be driven into a viable but not culturable (VBNC) state, which allows continuous survival without the ability to divide (Garcia-Armizen and Servais, 2004; Na et al., 2006). In the non-culturable state, the cells may not be available for enumeration with growth media, but they can be resuscitated to regain that ability when the environmental stress is alleviated (Wang and Doyle, 1998). It is possible that the Q_{10} equation is only applicable to simulate *E. coli* survival in an optimum window of temperatures starting above and ending below stressful conditions.

Values of Q_{10} temperature coefficients reported for most biological reactions commonly range between 2 and 3, meaning that the reaction rate doubles or triples for every

10 °C increase (Reyes et al., 2008). Our results show values of Q_{10} mostly less than 2 (Tables 1 and 2) and as small as 1.1, indicating very low sensitivity to temperature. Applying Eq. (A5) to the data of Allwood et al. (2003), Barcina et al. (1986a), and Craig et al. (2004) on *E. coli* inactivation in dechlorinated tap water, river water, and recreational coastal water, respectively, resulted in values of $Q_{10} = 1.31$, 1.98, and 2.01, which are substantially smaller than the values shown in Table 2. The sensitivity of the survival rates to temperature becomes relatively low at values of Q_{10} close to 1. Such low sensitivity has been observed in experiments with an absence of microflora (Korhonen and Martikainen, 1991). The temperature sensitivity of *E. coli* die-off in natural conditions, and therefore values of Q_{10}, might also be affected by light–dark diurnal variations. This effect could be elucidated in light–dark variation-based *E. coli* survival studies that are currently very rare but would be of special interest.

The sensitivity of *E. coli* survival to temperature has been shown to have implications for management decisions. Sampson et al. (2006) noted that the observed dependence of *E. coli* survival to temperature challenges the beliefs held by many recreational water managers that *E. coli* will not survive as long at colder temperatures as in warmer conditions. Vasconcelos and Swartz (1976) stated that since the survival potential of *E. coli* and other coliforms can be extended by low water temperatures, the discontinuance of wastewater disinfection during the winter months seems inadvisable for many cold coastal regions. Harmel et al. (2010) observed much lower *E. coli* concentrations in a river than in small streams and agricultural waters across its watershed, suggesting faster *E. coli* die-off in the river than other categories of waters (analogous to what can be seen in Fig. 3 and Table 1). The possibility of scale-dependence in microorganism survival in waters needs to be researched with reference to watershed-scale bacterial transport modeling.

The frequent occurrence of Type 2 and 3 inactivation patterns observed in our database supports the conclusions of previous reviews (Benham et al., 2006; Pachepsky et al., 2006), namely, that applicability of the one-parametric Chick law (1) to actual data on microorganism survival is limited, and more flexible models may be needed. The linear approximation of "log $C - t$" dependencies appears to be inaccurate in many cases (Fig. 4); therefore, dataset truncation that corresponds to the approximately linear sections of Type 2, 3, and 4 patterns more accurately represents the *E. coli* inactivation rate. A variety of models, including expressions developed by Veltz, Weibull, Gompertz, Frost and Streeter, among others, have been suggested as better mathematical expressions to fit *E. coli* inactivation data in which gradual decrease in die-off rate is observed (Easton et al., 2005; Corradini and Peleg, 2004). We note however, that we have observed mostly well-expressed piecewise-linear Type 2 patterns, while smooth, noticeably downward, concave curves with well defined non-linearity were relatively rare. Current predictive models used to support management decisions, such as SWAT and HSPF, consider only Type 1 inactivation patterns (Benham et al., 2006). Therefore, these models may substantially underestimate or overestimate the in-stream microorganism inactivation when the Type 1 is not applicable – or in almost three-quarters of cases based on frequencies found in this work.

5. Conclusions

Inspection of multiple datasets on *E. coli* inactivation in natural waters in laboratory experiments showed there are three general patterns of *E. coli* inactivation in waters. In two of them, which occurred almost three-quarters of the time, *E. coli* inactivation does not follow the first-order decay equation, and dependence of logarithm of concentration on time is not approximately linear, but rather piecewise-linear with, usually, two linearity sections. The uncertainty in inactivation shapes presents a source of uncertainty that must be acknowledged and dealt with explicitly in pathogen and indicator fate and transport models.

Applied to data from the first linearity section of inactivation, the Q_{10} equation can be used to model the dependence of *E. coli* inactivation rates on temperature in various sources of water. Differences in *E. coli* inactivation rates among broad groups of waters can be seen, however, the Q_{10} equation is more accurate when water from the same source is tested at different temperatures.

This work can be extended to test the applicability of the Q_{10} equation on *E. coli* inactivation in waters exposed to time-variable temperature and illumination conditions, as well as for *E. coli* inactivation in soils and organic waste. Preliminary research shows that data on *Salmonella* and enterococci can be collected and processed in a manner similar to this work. Collecting a broader database will be helpful because better guidance can be given in calibrating fate and transport modeling systems used to make environmental management decisions.

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Appendix A

Relationships between models of microbial survival rate dependence on temperature

From the Arrhenius equation (7), the ratio of rate constants at temperatures T and T_* is

$$\begin{aligned}\frac{k}{k_*} &= \exp\left[-\frac{E_a}{R}\left(\frac{1}{T}-\frac{1}{T_*}\right)\right]\\ &= \exp\left[-\frac{E_a}{R}\left(\frac{1}{273.15+T_C}-\frac{1}{273.15+T_{C,*}}\right)\right]\\ &= \exp\left[\frac{E_a}{R}\,\frac{(T_C-T_{C,*})}{(273.15+t)(273.15+t_*)}\right]\\ &= \left\{\exp\left[\frac{1}{R\times 273.15^2}\,\frac{E_a}{\left(1+\frac{T_C}{273.15}\right)\left(1+\frac{T_{C,*}}{273.15}\right)}\right]\right\}^{t-t_*}\end{aligned} \tag{A1}$$

where T_C is the Celsius temperature. If terms $T_C/273.15$ and $T_{C,*}/273.15$ are small compared to 1, which is commonly the case, then the last equation reduces to

$$\frac{k}{k_*} = \left\{ \exp\left[\frac{E_a}{R \times 273.15^2}\right] \right\}^{T_C - T_{C,*}} \tag{A2}$$

Comparison of Eqs. (A1) and (4) shows that they are identical provided

$$Q_{10} = \exp\left[\frac{10E_a}{R \times \times 273.15^2}\right] \tag{A3}$$

The value of the gas constant is $R = 8.31$ J/(mol K), and values of the inactivation energy for Q_{10} equal to 1.5, 2, and 2.5 are 2.5×10^4 J/mol, 4.3×10^4 J/mol, and 5.7×10^4 J/mol, respectively.

Eq. (3) with k defined from (4) is

$$\log C = \log C_0 - k_* Q_{10}^{(T_C - T_{C,*})/10} t \tag{A4}$$

Decimation results in $\log C_0 - \log C = 1$ and therefore

$$t_{90} = k_* Q_{10}^{-(T_C - T_{C,*})/10} \tag{A5}$$

Therefore, linear regression

$$\log t_{90} = A - BT_C \tag{A6}$$

has the slope B related to Q_{10} as

$$Q_{10} = 10^{10B} \tag{A7}$$

Appendix B. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.watres.2012.10.027.

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