

Microbial risk implications of rainfall-induced runoff events entering a reservoir used as a drinking-water source

R. S. Signor, N. J. Ashbolt and D. J. Roser

ABSTRACT

Waterborne disease outbreaks have been associated with periods of heightened source water pathogen concentrations in treated drinking-water supplies. For their management it is necessary to identify and quantify the impacts of events which lead to adverse concentration fluctuations. The aim of this work was to estimate relative microbial risks to water consumers arising from one such event: rainfall-induced runoff entering a surface drinking-water reservoir in an Australian agricultural catchment. Runoff events are known to influence both the source water entering a reservoir and the ability of a reservoir to act as a barrier to pathogen progression. Hydrograph separation methods were used to distinguish 'runoff event' from baseflow periods and pathogen concentrations of the inflow to the reservoir during runoff and baseflow periods respectively were estimated. Relative impacts of runoff event periods on health risks to consumers were assessed using Quantitative Microbial Risk Assessment principles. Runoff event conditions predominated 14% of the time. The proportions of infections attributable to runoff event periods from *Cryptosporidium*, *Giardia* and *Campylobacter* spp. were 57%, 80%, and 28% respectively. Daily infection risks were greatest in winter months than other seasons primarily due to the higher frequency of runoff events. Results from this and similar analyses, aiming to assess impacts of explicitly identified events on consumer health risks, provides important information for water system risk management, e.g. identifying periods of heightened risk and setting management priorities.

Key words | *Campylobacter*, *Cryptosporidium*, *Giardia*, microbial risk assessment, runoff events

R. S. Signor (corresponding author)
N. J. Ashbolt
D. J. Roser
School of Civil and Environmental
Engineering/Centre for Water and Waste
Technology,
The University of New South Wales,
Sydney NSW 2052,
Australia
Tel: +61(2) 9385 7896
Fax: +61(2) 9313 8624
E-mail: ryan.signor@student.unsw.edu.au

INTRODUCTION

Quantitative Microbial Risk Assessment (QMRA) offers a means to estimate communicable disease/infection risks to consumers of treated drinking-water (e.g. Rose *et al.* 1991; Teunis *et al.* 1997; Crabtree *et al.* 1997; Barbeau & Payment 2000; Teunis & Havelaar 2002; Pouillot 2004). Defining source water quality is an integral part of QMRA. Source water quality fluctuates with time and environmental conditions, and such variability can have a major impact on the long-term health risk estimate (Teunis *et al.* 2004). Particularly, contaminant concentrations in catchment surface waters increase following periods of rainfall

(e.g. Poulton *et al.* 1991; Atherholt *et al.* 1998; Kistemann *et al.* 2002; Signor *et al.* 2005) and epidemiologic studies have linked waterborne disease outbreaks with periods of high precipitation (e.g. Curriero *et al.* 2001; Rose *et al.* 2001; Naumova *et al.* 2005).

Storage reservoirs often serve as one of the first barriers to the transportation of microbial pathogens through a water supply system. Mechanisms include dilution of the incoming flow and provision of storage time allowing particle settling & pathogen decay (Hipsey *et al.* 2005). Governed by diverse environmental factors, the reservoir's

barrier effect varies between systems and with time, e.g. periods of lower sunlight intensity (Sinton *et al.* 1999; Linden *et al.* 2001) and cooler temperature (Buswell *et al.* 1998; Walker & Stedinger *et al.* 1999) may enhance a pathogen's survivability. Following rainfall, the contaminant loading rate into the reservoir may be many orders of magnitude greater than during the preceding dry weather (e.g. Roser & Ashbolt 2004). High inflows can also cause reservoir short-circuiting, where rather than mixing, inflows travel as a constrained plume along the reservoir depths – enhancing the possibility of viable pathogens being transported the length of the reservoir (Hipsey *et al.* 2005).

Mechanistically characterising the pathogen protection offered by reservoir storage processes is complex (Hipsey *et al.* 2005). It has been for QMRA purposes estimated empirically by comparing microbial count data collected at a stream's point of entry into a reservoir with that at a downstream water treatment plant (WTP) off-take (Teunis & Havelaar 1999; Teunis *et al.* 1999). Considering the numerous processes which influence a waterborne pathogen's ability to survive within a reservoir (Hipsey *et al.* 2005), the approach is rudimentary, yet is conceptually sound and suits the QMRA purpose. Not considered in prior empirical applications however is what may cause the reservoir transformation ratio to vary. For example, in a catchment which experiences more frequent rainfall events, cooler temperatures and more cloudy days in winter than in the summer months, intuitively the reservoir may be expected to serve more poorly as a pathogen barrier in the winter. If typical daily pathogen loads entering the reservoir were also greater in winter than summer due to more frequent runoff events, this may enhance the health risks to water consumers over this period. Capturing these links may be important for the QMRA.

The primary aim of this work was to adapt QMRA principles to estimate the impact on health risks arising from rainfall-induced runoff events entering a surface water reservoir for a case-study from southern Australia. A methodology was developed that involved: (i) distinguishing historical reservoir inflow 'runoff events' from 'baseflow' conditions; (ii) quantifying concentrations entering the reservoir under each condition for *Escherichia coli* as well as three reference pathogens (*Cryptosporidium parvum*, *Giardia lamblia*, *Campylobacter jejuni*); (iii) estimating the microbial transportation "barrier effect" of the reservoir; and

(iv) inputting results from the three previous steps into a QMRA model to assess the relative health risks to consumers posed under dry weather and rainfall event conditions.

STUDY AREA

The main study area is a 76-km² catchment draining to a drinking-water reservoir (*ca.* 27 GL capacity) located in the Adelaide Hills of South Australia (Figure 1). The reservoir's total catchment area including the reservoir foreshores is 140 km². The region has a warm temperate climate with mean daily maximum temperatures ranging from 13°C in July to 27°C in January. The mean annual rainfall is 767 mm with heavier rainfall in the winter. Consequently, the average daily flow entering the reservoir was much greater in winter (68 ML in July) than summer (0.87 ML in February) for the period 2000–2004.

Several potential pathogen contaminant sources existed. Most land area was used for grazing (62%) and dairying (24%) with unrestricted stock access along 81% of the primary watercourses. Within the catchment the human population was approximately 500 persons living in 200 dwellings serviced by on-site sewage systems. About 100 dwellings either have a mapped ephemeral watercourse flowing by them or were located on the edge of the reservoir and considered potential contaminant point sources.

DATA

Hydrology data

A stream rating curve and hourly river stage data collected over the period 2000 to 2005 near the main watercourse point of entry into the reservoir (SP1, Figure 1) were obtained from the South Australian Department of Water, Land & Biodiversity Conservation.

Microbiological data

Microbial data collected between 2000 and 2005 from two locations: within the main river 0.5 km upstream of the reservoir inlet (denoted as SP1) and from within the

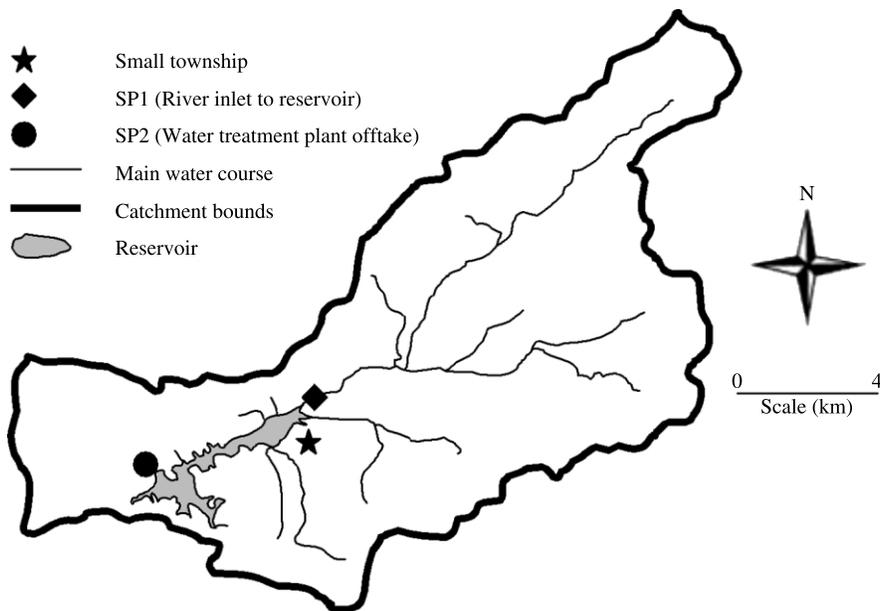


Figure 1 | Study area.

reservoir at the water treatment plant off-take (SP2) (Figure 1) were made available. Water quality analyses were undertaken by the Australian Water Quality Centre's (AWQC's) Analytical Laboratory (Bolivar, South Australia) using their standard procedures for *E. coli*, *Cryptosporidium*, *Giardia*, and *Campylobacter* spp. (AWQC methods 2246-19/2247-50/2248-50, 88-01 and 81-07 respectively). In brief, protozoa were concentrated by flocculation of 10 L samples, immunomagnetic separation and centrifugation, and then counted using USEPA Method 1623 (USEPA 1999). A fluorescently labelled control (either 'EasySeed™' or 'ColorSeed™') of known concentration were used to assess the efficacy of the method for each protozoa assay. *E. coli* was enumerated from either 100 or 1 ml samples using IDEXX Colilert®. *Campylobacter* spp. were assayed by enrichment in 3 × 3 most probable number tubes followed by polymerasechainreaction based confirmation.

DATA ANALYSIS

All numerical operations for this study were performed using *Mathematica*® 5.0.1.0 software (Wolfram Research Inc., Champaign, Illinois, USA).

Defining 'runoff events'

Runoff event periods were defined using a time-step based hydrograph filtering algorithm applied to the flow data to separate the baseflow component of the total flow (Chapman 1999). While there are several variations used by hydrologists for such purposes, the most common is:

$$Q_{s,i} = KQ_{s,i-1} + \frac{(1+K)}{2}(Q_i - Q_{i-1}) \quad (1)$$

where $Q_{s,i}$ is the flowrate runoff component at time step i , Q_i is the total recorded flow at time step i , and K is the baseflow recession constant for the stream. During baseflow conditions (periods of no runoff contribution) the hydrograph recession curve is expressed as:

$$Q_t = Q_0 K^t \quad (2)$$

where Q_t is the total flow at time t , and Q_0 the total initial flowrate. Estimating K requires first identifying periods of baseflow recession (Tallaksen 1995). Ten flow recession periods were identified from the 5-year hydrograph representing all seasons and record years, ranging from 15 to 143 h duration (average = 60.9 h). Such periods were identified as recessive sections of the hydrograph which appeared linear on a semi-logarithmic hydrograph plot (Chapman 1999).

An estimate of the recession constant was made for each period by the method of least squares (Devore 2000). The overall recession constant used was the average of those ten estimates. A ‘runoff event’ was defined as any period of consecutive hourly time steps where the runoff contribution estimated from Equation 1 was $Q_{s,i} > 0.05\text{-m}^3/\text{s}$ which corresponded to a stream runoff component depth of *ca.* 0.1 to 0.2 m when the baseflow component was very low ($Q_i < 0.01\text{-m}^3/\text{s}$), as estimated from the stream rating curve. At other times, the flow condition was defined as ‘baseflow’. The probability of the total flow being in either the runoff or baseflow states was summarised by the parameters A_R and A_B respectively, estimated as the proportion of time the hydrograph corresponded to each state within the period studied.

Organism concentrations entering reservoir

All microbial data collected from SP1 were time-matched with flow data and categorised as either ‘runoff event’ or ‘baseflow’ depending on the corresponding state of the river. When multiple samples were collected over the course of a single runoff event or baseflow period, they were grouped and summarised by a single arithmetic mean pathogen concentration for that period, similar to an Event Mean Concentration (Huber 1993; Signor et al. 2005). Probability density functions were employed to describe the variability of the microbial source water quality data. It was assumed the mean organism concentration Λ of whole runoff events or baseflow periods varied between independent periods according to a probability density function f so $\Lambda \sim f(\lambda; \Theta)$, where Θ is a parameter vector.

Results from each protozoan sample were reported as a set of: the number of (oo)cysts counted in the sample n ; the sample volume v ; the number x of fluorescent (oo)cysts seeded into a parallel sample to assess the recovery fraction; and the number s of seeded (oo)cysts recovered by the same enumeration method from the parallel sample. Random distribution of the (oo)cysts in the water body at the time of sampling was modelled by a Poisson probability distribution. The mixed Poisson likelihood function L_c used to estimate Θ (by maximum likelihood methods) was based on those described by Haas et al. (1999: 195) – also refer to the

bold text in the Addendum for more detail:

$$L_c(\Theta|n_{ij}, v_{ij}, s_{ij}, x_{ij}) = \prod_{j=1}^k \left[\int_0^{\infty} f(\lambda; \Theta) \prod_{i=1}^m \left[\frac{(\lambda v x / s)^n}{n! e^{-\lambda v x / s}} \right]_i \cdot d\lambda \right] \quad (3)$$

where $(n_{ij}, v_{ij}, x_{ij}, s_{ij})$ are the given data from the i th of m samples taken during the j th of k observed independent runoff or baseflow periods and $L(\mathbf{X}|Y)$ is the likelihood function L to estimate parameters \mathbf{X} given dataset Y .

E. coli data were reported as most probable numbers (MPN)/ml or 100 ml and always as a whole integer estimate. *Campylobacter* spp. data were also reported as MPN/100 ml, though as a continuous rather than discrete number. The following likelihood function (Haas et al. 1999: 216, Cook et al. 2000) L_d was optimised to obtain estimates of Θ for the bacteria data:

$$L_d(\Theta|\lambda_j, \lambda_{up,j}, \lambda_{lo,j}, \delta_j) = \prod_{j=1}^k [f(\lambda_j; \Theta)]^{\delta_j} \cdot [F(\lambda_{up,j}; \Theta) - F(\lambda_{lo,j}; \Theta)]^{1-\delta_j} \quad (4)$$

where λ_j is the estimated average organism concentration from m samples taken within the j th of k observed baseflow or runoff periods, $F(\cdot)$ is the probability density’s cumulative function, δ_j is a binary indicator variable set to zero when one or more of m samples taken within the j th period were reported ‘negative’ (*i.e.* reported as ‘below detection limit’), otherwise $\delta_j = 1$, and $\lambda_{up,j}$ and $\lambda_{lo,j}$ are inferred upper and lower bounds of the observed average concentration for the j th period.

For input to Equation 4 λ_j was estimated as follows: Consider z_{ij} as the MPN reported from the i th of m samples taken during the j th of k observed independent runoff or baseflow periods. When all m samples taken within the j th period were positive, $\delta_j = 1$, and:

$$\lambda_j = \left(\sum_{i=1}^m z_{ij} \right) / m$$

Otherwise if one or more of m samples taken within the j th period were ‘negative’ then $\delta_j = 0$, and:

- all $\lambda_{lo,j}$ were estimated from the same equations used for λ_j , but where all ‘negative’ z_{ij} were set to zero;
- all $\lambda_{up,j}$ were estimated from the same equations used for

λ_j , but where all ‘negative’ z_{ij} were set to the reported MPN method detection limits (of 0.3-MPN/100-ml for *Campylobacter* spp. data, and 1 MPN/100-ml for *E. coli*).

The lognormal:

$$P(\Lambda = \lambda) = \frac{e^{-[\ln(\lambda) - \mu]^2 / (2\sigma^2)}}{\sqrt{2\pi}\sigma\lambda}, \text{ NB : } \Lambda > 0; e = 2.718\dots;$$

$$\pi = 3.141\dots; \mu, \sigma \text{ are scale, shape parameters}$$

and the gamma:

$$P(\Lambda = \lambda) = \frac{\lambda^{\alpha-1} e^{-\lambda/\beta}}{\beta^\alpha \Gamma(\alpha)}, \text{ NB : } \Lambda > 0; \Gamma(\cdot) \text{ is the gamma}$$

function α, β are scale, shape parameters

probability density functions were the candidates used to describe the water quality variability (Ott 1995). Numerical optimisation techniques were used to maximise the likelihood functions and estimate the function parameters. The candidate probability function which produced the largest optimised likelihood value was adopted as the representative function (Kappenman 1982). To assess the significance of differences in microbial concentrations between baseflow & runoff event periods overall and between seasons (summer, autumn, winter, spring), a deviance statistic-based likelihood test was used as described by Haas *et al.* (1999: 297). When an event or baseflow period overlapped multiple seasons, it was classed as having occurred within the season that the period began.

Reservoir transformation ratio

The many processes which dictate the reservoir’s ability to act as a barrier against pathogen transport were summarised by a transformation ratio. The reservoir transformation ratio was defined as the ratio of the observed mean micro-organism concentration at SP2 to that at SP1, taking into account the proportion of time that the incoming flows were in baseflow and runoff event modes. An unpaired data model likelihood function L_r was derived (Equation A.1, see Addendum) to estimate ϵ_r , based on models described by Teunis *et al.* (1999). Though *E. coli* data was reported by the laboratory as a whole integer number of organisms, it is a derived MPN rather than an organism count. No additional

information with regard to the MPN derivation was reported. Equation A.1 was applied to *E. coli* data by interpreting each reported MPN at SP1 and SP2 as an organism count in the reported sample volume and assuming the method recovery fraction to be unity. Essentially, this meant random sampling processes were incorporated into the *E. coli* ratio estimate, but not the uncertainty associated with derivation of the MPN. No *Campylobacter* spp. data was available at SP2 and no specific ratio estimate was possible. Numerical integration and optimisation methods were used to estimate ϵ_r by maximising Equation A.1.

The transformation ratio was estimated separately for each season, using season-specific datasets. Hence, the variability of ϵ_r , rather than described by a continuous probability density function, was considered here as a possible variance between seasons of a specific ratio value.

QMRA

Where d was the pathogen dose ingested by a consumer over a daily period:

$$d = [A_B \cdot E(\Lambda_B) + A_R \cdot E(\Lambda_R)] \cdot \epsilon_r \cdot \epsilon'_{\text{WTP}} \cdot V' \quad (5)$$

and V' was an individual’s long-term average daily water consumption, ϵ'_{WTP} was the fraction of pathogen organisms which successfully pass through physico-chemical water treatments, and $E(\Lambda_B)$ & $E(\Lambda_R)$ were the expected mean organism concentrations of flow into the reservoir under baseflow and runoff conditions respectively. Parameters $A_B, A_R, E(\Lambda_B), E(\Lambda_R)$, and ϵ_r (the ‘parameters of interest’) in Equation 5 were estimated as described previously. Other parameters were based on relevant literature (Table 1).

The estimates of d were input to dose–response relationships (Table 2) to estimate long-term average daily probability of infection P_{daily} to consumers. Assuming consumers ingest one independent pathogen dose per day, the probability of infection to a consumer from a series of independent exposures P_{ann} over 365 days is

Table 1 | Literature-based parameters used for QMRA

Parameter	Value	Reference/comments
$\varepsilon'_{\text{WTP}}$:		
<i>Cryptosporidium</i> spp.	6.3×10^{-4}	Based on Hijnen <i>et al.</i> (2005) review of <i>Cryptosporidium</i> oocyst removal by conventional water treatment ^a + assumed 0% reduction due to disinfection ^b (chlorine addition)
<i>Giardia</i> spp.	7.9×10^{-5}	Based on Hijnen <i>et al.</i> (2005) review of <i>Giardia</i> cyst removal by conventional water treatment ^a + assumed 90% reduction due to disinfection ^b (chlorine addition)
<i>Campylobacter</i> spp.	7.9×10^{-6}	Based on Hijnen <i>et al.</i> (2005) review of bacterial organism removal by conventional water treatment ^a + assumed 99.9% reduction due to disinfection ^b (chlorine addition)
V' :	1.1-L	Roseberry & Burmaster (1992)

^aThe source water is treated by coagulation & flocculation, floc removal (by flotation), rapid sand filtration and chlorine disinfection.

^bChlorination provides protection against pathogen transmission though *Cryptosporidium* spp. are quite resistant (Korich *et al.* 1990). *Giardia* spp. (Clark *et al.* 1989) and especially *Campylobacter* spp. (Blaser *et al.* 1986) are more susceptible. Conservative relative density reductions due to chlorination were assumed, which was considered sufficient for modelling purposes.

(Haas *et al.* 1999):

$$P_{\text{ann}} = 1 - (1 - P_{\text{daily}})^{365} \quad (6)$$

The contribution of runoff events and the seasonal differences in health risks to water consumers from this system was assessed using Equations 5 & 6 and the relationships in Table 2 with season-specific estimates of the parameters of interest as inputs.

RESULTS & DISCUSSION

Hydrology & 'runoff event' characterisation

The recession constant estimated for the hydrograph filtering was $K = 0.989 (\pm 0.005)$. 'Runoff event' periods (periods where the total flow and baseflow values deviate)

corresponded well with recorded rainfall in the catchment (Figure 2). The average annual hydraulic load for the studied period was 9099 ML. Fifty eight percent of the annual hydraulic load entered the reservoir in the three winter months, and 40 % was attributable to winter runoff events (Table 3).

Microbial concentrations and loads entering the reservoir

The charts in Figure 3 are a plot of the average reported organism concentration and flowrate for each identified continuous baseflow or runoff event period. Organism concentrations in the stream entering the reservoir were greater during 'runoff event' periods.

A data summary is displayed in Table 4. Differences in concentrations were significant between baseflow and

Table 2 | Dose–response relationships

Pathogen	Dose–response relationship	Reference
<i>Campylobacter</i>	$P_{\text{inf}} = 1 - (1 + d/7.59)^{-0.145}$	Medema <i>et al.</i> (1996)
<i>Giardia</i>	$P_{\text{inf}} = 1 - e^{-d/50.3}$	Teunis <i>et al.</i> (1996)
<i>Cryptosporidium</i>	$P_{\text{inf}} = 1 - e^{-d/238.6}$	Haas <i>et al.</i> (1996)

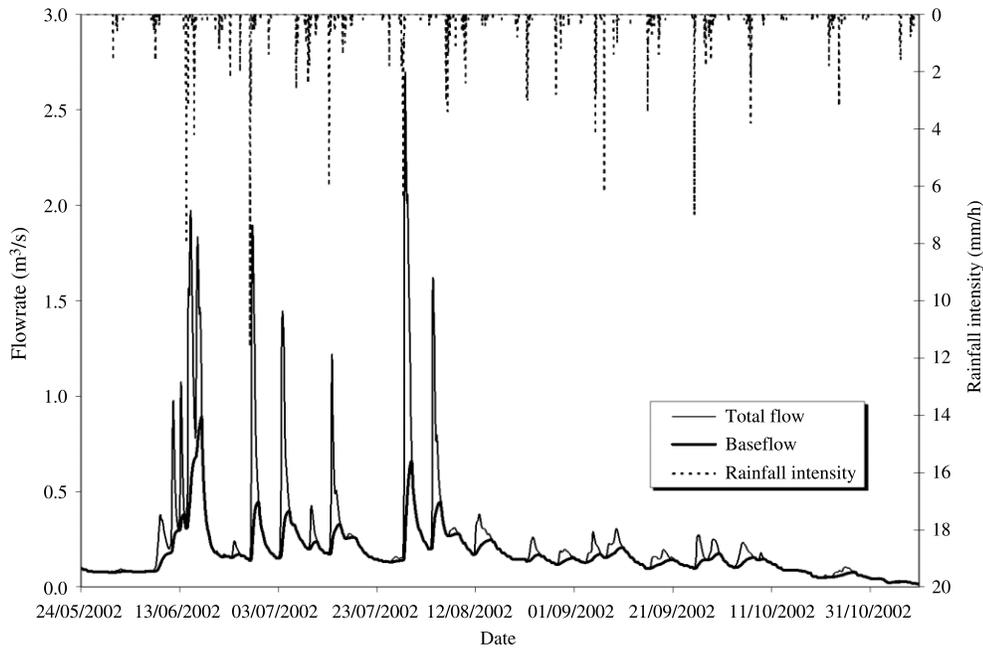


Figure 2 | Separation of baseflow and runoff components of river flow entering.

Table 3 | Characteristics of stream flowrates entering reservoir

Season	State ^a	Avail. ^b		Q (m ³ /s)		Average	Average annual hydraulic load (ML)
		(fraction)	5th %ile	Median	95th %ile		
Sum.	R	0.017	0.017	0.30	0.98	0.33	42
	B	0.983	4×10^{-4}	0.0058	0.065	0.016	122
Aut.	R	0.065	0.070	0.28	1.50	0.54	280
	B	0.935	$< 1 \times 10^{-4}$	0.044	0.14	0.051	375
Win.	R	0.329	0.24	0.95	3.67	1.34	3488
	B	0.671	0.092	0.25	0.77	0.34	1806
Spr.	R	0.160	0.17	0.76	5.57	1.39	1745
	B	0.840	0.015	0.13	0.62	0.19	1241
All	R	0.143	0.16	0.76	3.86	1.23	5555
	B	0.857	0.001	0.055	0.48	0.13	3544
	C	1.000	0.002	0.081	1.38	0.29	9099

^aR = 'runoff event', B = 'baseflow periods', C = 'combined'.

^bAvailability – proportion of time that the stream flowrate corresponds to 'runoff event' and 'baseflow periods' respectively for the season(s) indicated.

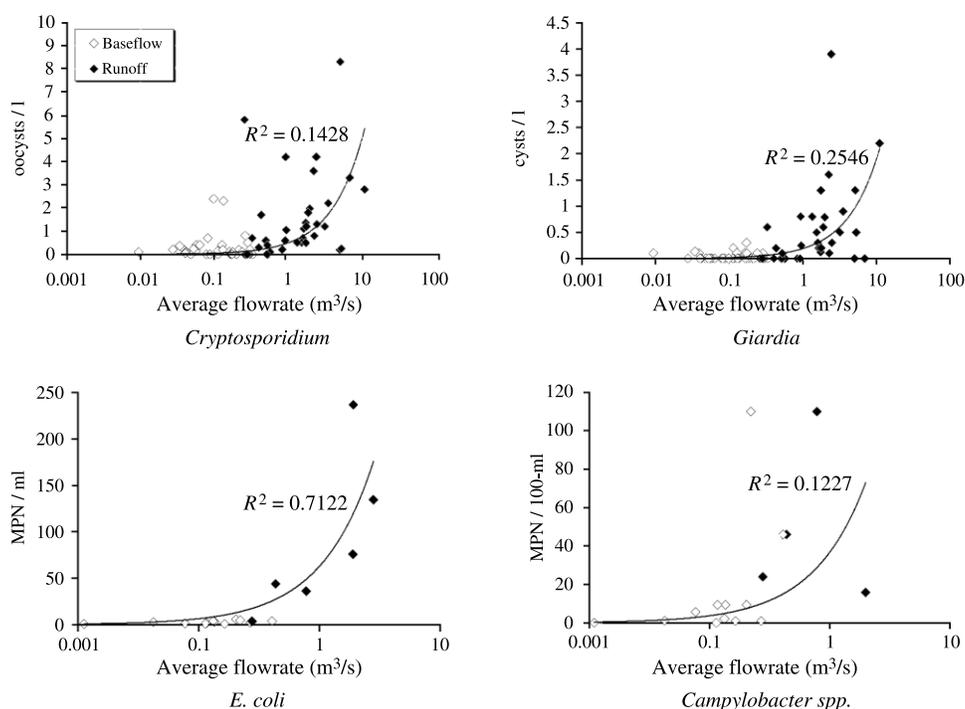


Figure 3 | Average organism densities vs. average flowrate for independent baseflow and runoff event periods with linear trendlines fitted (negative data plotted as zero pathogen density). Protozoa data not adjusted for method recovery.

runoff event data subsets, but not between seasons within the same categories, for *Cryptosporidium* and *Giardia* [p -values < 0.05]. Mean recovery fractions from all samples were high for both analytes (*Cryptosporidium* 51%, *Giardia* 49%), providing a high degree of confidence in the quality of the protozoa data. As the *E. coli* and *Campylobacter* spp. datasets were smaller than those for the protozoa, statistical comparison between seasons was not practical. However runoff event concentrations were significantly greater than the baseflow concentrations [p -values < 0.05]. The observed differences between runoff event and baseflow organism concentrations in the river were consistent with findings from previous studies (e.g. Poulton *et al.* 1991; Atherholt *et al.* 1998; Kistemann *et al.* 2002; Signor *et al.* 2005). Note that although runoff event inflows occurred for just 14% of the time, 51% (*Cryptosporidium* and *Giardia*), 33% (*E. coli*) and 25% (*Campylobacter*) of sampling periods at SP1 occurred during runoff events. The water utility sampling protocol calls for additional water quality sampling to occur following some rainfall event-based trigger, hence the dataset was biased toward event

conditions. As micro-organism concentrations were significantly greater during runoff events, simply fitting all data to a single probability density function would have resulted in an over-estimation of the overall mean pathogen concentration, in turn impacting QMRA results based on these estimates.

The Exceedance Probability charts (Figure 4) show that the selected probability distributions (either lognormal or gamma) provided good fits, all data shown fell within or very near the depicted 90% confidence intervals. The EP plot for the *Campylobacter* spp. highlights an issue regarding the use of very small datasets – although the expected mean *Campylobacter* spp. concentration was greater during runoff periods than baseflow periods, the maximum-likelihood estimate at the 95th and greater percentiles of variability for baseflow concentrations was greater than that for the runoff concentrations. Despite a smaller sample size, the 90% uncertainty interval surrounding the *Campylobacter* spp. runoff estimate was narrower than that for the baseflow estimate. The likely explanation was that the small number of observed *Campylobacter* spp. runoff event

Table 4 | Summary of microbiological data collected at SP1

Season	State ^a	Data Summary Mean ^b	SD ^b	k ^c	# pos. ^d	Maximum	Minimum
<i>Cryptosporidium</i> (oocysts/L)							
Summer	R	–	–	0	–	–	–
	B	0.38	0.25	4	4	0.74	0.20
Autumn	R	4.00	5.73	7	6	16.11	< 0.38
	B	1.18	2.25	8	6	6.67	< 0.082
Winter	R	5.28	8.37	20	18	35.00	< 0.12
	B	0.30	0.45	13	7	1.36	< 0.063
Spring	R	1.39	1.70	9	7	5.16	< 0.16
	B	0.47	0.63	10	6	1.92	< 0.083
<i>Giardia</i> (cysts/L)							
Summer	R	–	–	0	–	–	–
	B	0.061	0.12	4	1	0.24	< 0.082
Autumn	R	0.75	0.93	7	4	2.62	< 0.17
	B	0.072	0.11	8	3	0.29	< 0.063
Winter	R	3.04	8.60	20	15	39.00	< 0.096
	B	0.098	0.18	13	4	0.50	< 0.13
Spring	R	0.27	0.42	9	5	1.14	< 0.14
	B	0.042	0.089	10	2	0.22	< 0.036
<i>E. coli</i> (MPN/ml)							
Summer	R	–	–	0	–	–	–
	B	3.1	0.92	2	2	3.7	2.4
Autumn	R	–	–	0	–	–	–
	B	–	–	0	–	–	–
Winter	R	120	100	3	3	240	44
	B	3.2	2.2	4	4	5.5	0.78
Spring	R	58	68	3	3	130	3.5
	B	1.7	1.2	6	6	3.50	0.54
<i>Campylobacter</i> spp. (MPN/100-ml)							
Summer	R	–	–	0	–	–	–
	B	1.6	0.71	2	2	2.1	1.1
Autumn	R	–	–	0	–	–	–
	B	–	–	0	–	–	–
Winter	R	31	21	2	2	46	15.9
	B	30	53	4	4	110	0.9
Spring	R	67	61	2	2	110	24
	B	12	17	6	4	46	< 0.30

^aR = 'runoff event', B = 'baseflow period'.

^bMean = arithmetic mean, SD = normal standard deviation, of the mean densities from *k* independent baseflow or runoff periods. Each period mean density was estimated by assuming all 'negative' samples had a density of 0-organisms per unit volume. Each protozoa sample was adjusted by multiplying the organism count by the inverse of that sample's recovery fraction.

^cNumber (unitless) of observed baseflow, runoff or combined periods with associated microbiological data.

^dNumber (unitless) of observed baseflow, runoff or combined periods with associated microbiological data for which at least one sample was not 'negative'.

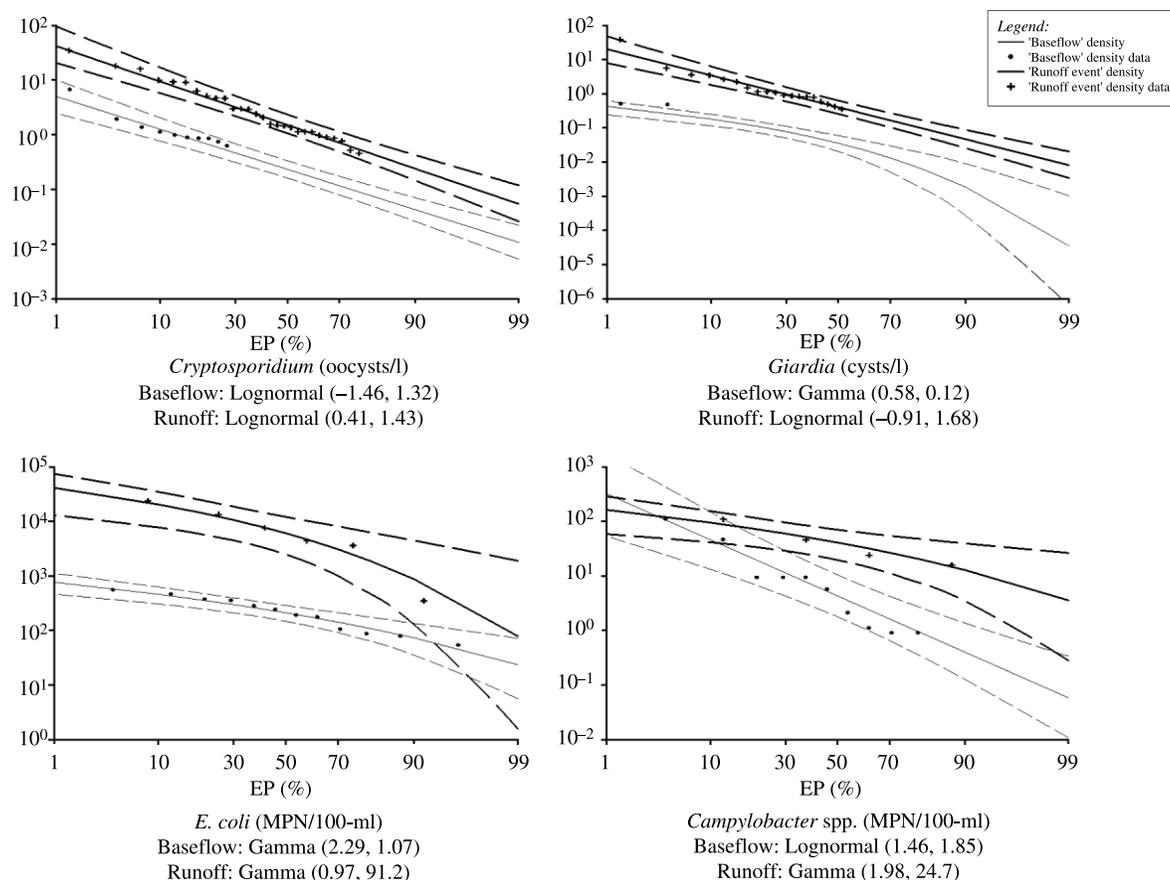


Figure 4 | Exceedance probability charts of mean pathogen densities entering the reservoir at SP1 for independent event/baseflow periods. Solid lines are in the maximum-likelihood variability estimates and correlate to the relevant probability density function (lognormal or gamma) listed below the charts. Dashed lines represent 90% confidence limits derived from Markov-chain Monte Carlo method application (Gilks *et al.* 1996). Only data points with values greater than the highest recorded detection limit and where all samples in the event/baseflow period were positive are plotted.

concentrations ($k = 4$) did not capture the extent of the true variation – an important consideration if the distribution were used in a probabilistic QMRA model, where high percentile values of skewed stochastic inputs heavily influence overall risk estimates (Haas 1997; Teunis *et al.* 2004). More *Campylobacter* spp. data would have benefited this analysis.

Estimates of the total annual microbial load entering the reservoir from the catchment upstream of SP1 under each flow condition and for each season were made using the product of the average flowrate, expected mean organism concentration and the estimated amount of time that the particular flow condition occurred for that season (Figure 5). In excess of 70% of the *Campylobacter* and more than 90% of the *E. coli* and protozoa loads occurred during the runoff events. Forty to sixty

percent of the total micro-organism loading into the reservoir occurred in the three winter months.

Reservoir transformation ratio

E. coli concentrations at SP2 were highest in the winter, and the few observed positive protozoa samples at SP2 occurred in those months (Table 5). Though the reservoir transformation ratio was assessed seasonally, as micro-organism concentrations entering the reservoir under 'baseflow' and 'runoff event' conditions respectively were not significantly different between seasons, all microbial data collected at SP1 were used as inputs to Equation A.1 regardless of the season of interest. However, the A_R & A_B estimates and the SP2 data inputs were season-specific. As no protozoa were detected at SP2 in any

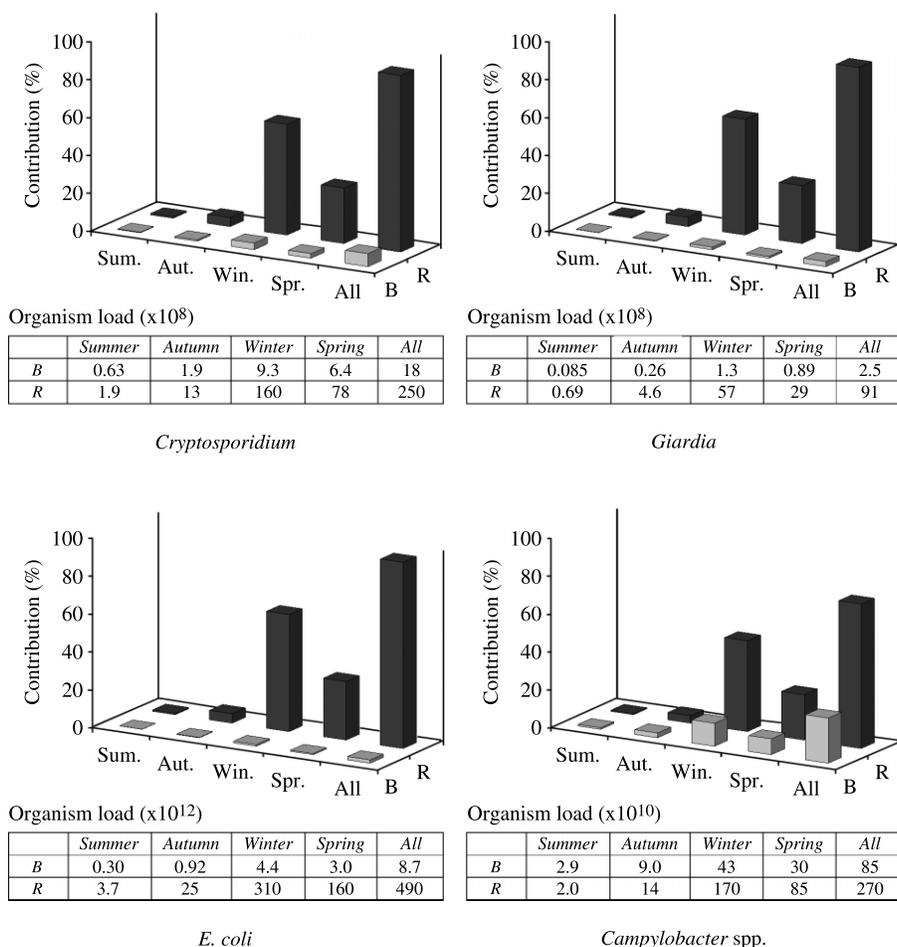


Figure 5 | Season/state breakdown of estimated annual micro-organism loads entering reservoir at SP1. 'B' = baseflow periods, 'R' = runoff event periods, 'All' = all seasons.

season other than the winter, the maximum likelihood protozoa transformation ratio estimates for the other seasons approached zero and were not meaningful estimates, which is a limitation of the approach adopted, and only the winter estimates were considered further.

Best-estimates and 90% confidence intervals for each transformation ratio are summarised in Table 6. There were a greater proportion of *E. coli* detects from samples at SP2 over all seasons (Table 5) and the maximum-likelihood point-estimates of ratios had a range 0.0020–0.0073. It was unanticipated that the seasonal ratio estimates would be so similar, given the seasonal disparity in flow and microbial reservoir loadings. Winter ratio estimates for *Cryptosporidium* (0.021) and *Giardia* (0.045) were of the same order of magnitude. Empirically the reservoir provided about an

order of magnitude greater protection against bacteria than against protozoa. Mechanistic modelling by Hipsey *et al.* (2005) of micro-organism fate and transportation in Australian reservoirs drew similar conclusions, citing the greater susceptibility of bacteria to ultra-violet light inactivation, and proneness to attach to larger settling-sized particles as the major reasons.

Despite the larger datasets, the transformation ratio estimates for protozoa had wider 90% confidence bands than the estimates for *E. coli*, due to a combination of low counts at SP1 and the large number of zero (oo)cyst counts in 10 L samples at SP2. Amongst the *E. coli* seasonal estimates, the autumn and summer ratios had widest uncertainty bands, which corresponded to the seasons with the greatest percentage of negative samples at SP2.

Table 5 | Summary of microbiological concentration data at SP2

Season	Mean ^a	SD ^a	# samples ^b	# positive ^c	Max.	Min.
<i>Cryptosporidium</i> (oocysts/L)						
Sum.	–	–	13	0	<0.29	<0.16
Aut.	–	–	12	0	<0.30	<0.12
Win.	0.025	0.090	15	2	<0.40	<0.12
Spr.	–	–	12	0	<0.33	<0.14
<i>Giardia</i> (cysts/L)						
Sum.	–	–	13	0	<1.00	<0.14
Aut.	–	–	12	0	<0.33	<0.15
Win.	0.018	0.063	15	1	<0.56	<0.11
Spr.	–	–	12	0	<0.48	<0.14
<i>E. coli</i> (organisms/ml)						
Sum.	0.086	0.028	14	9	0.11	<0.01
Aut.	0.016	0.19	15	9	0.61	<0.01
Win.	0.20	0.34	16	13	1.20	<0.01
Spr.	0.051	0.068	14	11	0.24	<0.01

^aMean = arithmetic mean, SD = normal standard deviation. Estimated by assuming all samples with no organisms detected had a density of 0-organisms per unit volume.

^bCount (unitless) of number of samples taken.

^cCount (unitless) of samples from the total number that had positive organism detection.

The methods described to estimate the transformation ratios and the associated outcomes reflected a pragmatic, conceptual approach to work with our specific (small and censored) datasets. Methods based on Teunis *et al.*'s (1999) unpaired data likelihood models were used. In such unpaired data likelihood models, the maximum-likelihood point estimate of the transformation ratio is largely unaffected by the assumed distributional form of the *variability* of the data at SP1 and SP2 (Teunis *et al.* 1999). Hence here the reservoir transformation ratios were defined simply as the ratio of the observed mean micro-organism concentrations at SP2 to those at SP1. Teunis *et al.* (1999) define the transformation ratio slightly differently, primarily because they have used the model to estimate not only the ratio, but also the variable pathogen concentrations of the water prior to a treatment process. Adopting the same approach here would have meant that the data collected at SP2 affected not only the transformation ratio estimates used in the subsequent QMRA, but also the estimates of the baseflow and runoff pathogen concentrations entering the reservoir respectively. As it was not possible to categorise the SP2 data as pertaining wholly to

Table 6 | Seasonal reservoir transformation ratio estimates and confidence intervals

Season	Estimated ϵ_r	90 % confidence interval ^a
<i>Cryptosporidium</i>		
Summer	–	–
Autumn	–	–
Winter	0.022	0.0095, 0.070
Spring	–	–
<i>Giardia</i>		
Summer	–	–
Autumn	–	–
Winter	0.045	0.019, 0.27
Spring	–	–
<i>E. coli</i>		
Summer	0.0034	0.0024, 0.0048
Autumn	0.0073	0.0065, 0.0086
Winter	0.0061	0.0058, 0.0067
Spring	0.0020	0.0016, 0.0024

^aEstimated from application of Markov-chain Monte Carlo methods (Gilks *et al.* 1996).

runoff or baseflow conditions it was our preference to use only SP1 data to characterise inflow concentrations, and SP1 + SP2 data to estimate the transformation ratio only.

QMRA results

For the protozoa, the winter data-derived transformation ratios were assumed to apply for all seasons. For *Campylobacter* spp. the *E. coli* transformation ratios were assumed applicable. Two other qualifications must be emphasised. Firstly, the QMRA results are indicative of the time period over which data was collected only. Secondly, the approach adopted was intended to quantitatively summarise the overall effects of runoff events on overall infection risks to consumers averaged out over a certain time period (either

seasonal or annual). Shorter-term impacts of individual event scenarios were not assessed.

The QMRA results were expressed in two ways (Table 7). The P_{daily} to a consumer is tabulated as the theoretical daily risk to a consumer on days when the source water corresponded wholly to a 'runoff event', a 'baseflow' period, and the overall combined daily infection risk taking into account the seasonal probability of the flow being in either state, respectively. The second way was the expected annual number of infections per population of 10,000 people for each scenario, accounting for the number of days per year that the scenario was expected to occur. The proportion of annual infections attributable to 'runoff event' periods for *Cryptosporidium*, *Giardia* and *Campylobacter* spp. were 57 %, 80 %, and 28 % respectively,

Table 7 | QMRA results

Season	State ^a	N ^b	<i>Cryptosporidium</i> spp.		<i>Giardia</i> spp.		<i>Campylobacter</i> spp.	
			P_{daily}^c	# inf ^d	P_{daily}^c	# inf ^d	P_{daily}^c	# inf ^d
Summer	R	1.5	2.63×10^{-7}	0.0039	1.29×10^{-7}	0.0019	2.75×10^{-7}	0.0041
	B	88.5	3.22×10^{-8}	0.0285	5.47×10^{-9}	0.0048	1.34×10^{-7}	0.1186
	C	90.0	3.61×10^{-8}	0.0325	7.52×10^{-9}	0.0068	1.36×10^{-7}	0.1228
Autumn	R	6.0	2.63×10^{-7}	0.0158	1.29×10^{-7}	0.0078	5.96×10^{-7}	0.0358
	B	86.0	3.22×10^{-8}	0.0277	5.47×10^{-9}	0.0047	2.90×10^{-7}	0.2497
	C	92.0	4.73×10^{-8}	0.0436	1.36×10^{-8}	0.0125	3.10×10^{-7}	0.2855
Winter	R	30.2	2.63×10^{-7}	0.0796	1.29×10^{-7}	0.0390	4.98×10^{-7}	0.1505
	B	61.8	3.22×10^{-8}	0.0199	5.47×10^{-9}	0.0034	2.43×10^{-7}	0.1499
	C	92.0	1.08×10^{-7}	0.0995	4.61×10^{-8}	0.0424	3.27×10^{-7}	0.3004
Spring	R	14.5	2.63×10^{-7}	0.0383	1.29×10^{-7}	0.0188	1.62×10^{-7}	0.0235
	B	76.5	3.22×10^{-8}	0.0246	5.47×10^{-9}	0.0042	7.88×10^{-8}	0.0602
	C	91.0	6.92×10^{-8}	0.0629	2.52×10^{-8}	0.0230	9.20×10^{-8}	0.0837
Annual	R	52.2	2.63×10^{-7}	0.1377	1.29×10^{-7}	0.0675	4.10×10^{-7}	0.2139
	B	312.8	3.22×10^{-8}	0.1008	5.47×10^{-9}	0.0171	1.85×10^{-7}	0.5784
	C	365.0	6.53×10^{-8}	0.2385	2.31×10^{-8}	0.0846	2.17×10^{-7}	0.7923

^aR = 'runoff event', B = 'baseflow', C = 'combined' (runoff event + baseflow).

^bN = number of days *per annum* corresponding to the seasonal/event condition indicated, inferred from A_R and A_B estimates in Table 2 when $K = 0.989$.

^cTheoretical daily risk to a consumer from Equation 5 on days when the source water corresponded wholly to a 'runoff event' (R), a 'baseflow' period (B), or the overall combined daily infection risk taking into account the seasonal probability of the flow being in either state (C), from Equation 5.

^dEstimated number of infections in a population of 10,000 attributed to the indicated seasonal/event condition from Equation 6..

shared among several sporadic event periods which accounted for 14% of the duration of one year. The greater frequency of runoff events resulted in 42% (*Cryptosporidium*), 50% (*Giardia*) and 38% (*Campylobacter* spp.) of the long-term annual risk occurring in the winter months alone, compared to 13%, 8% and 15% in the summer. Though under the described model assumptions *Campylobacter* spp. posed the greatest absolute risk to consumers (0.79 infections per 10,000 people *per annum*, compared to 0.24 and 0.08 for *Cryptosporidium* and *Giardia* respectively), the relative impact of runoff events on protozoa-borne risk appeared more pronounced. Two factors were identified as responsible: (i) the absolute difference in estimated runoff and baseflow mean pathogen concentrations was smaller for *Campylobacter* spp. than other pathogens analysed; and (ii) at low doses, the P_{daily} estimates from the *Campylobacter* spp. dose–response curve was less sensitive to dose fluctuations than the dose–response curves used for protozoa. *Cryptosporidium* and *Giardia* at SP2 that had zero (oo)cyst counts.

Risk management implications

The quantitative results suggest that source water quality entering and within the case study reservoir was poorest in the winter months, stemming from more frequent runoff events. All else being equal, this translated to greater potential infection risks to the receiving population over the same period. Others have demonstrated that pathogen concentrations in surface waters in other regions can have seasonal fluctuations (e.g. Westrell *et al.* 2004) and have described seasonal patterns in waterborne enteric disease occurrence in humans (Lopman *et al.* 2004; Naumova *et al.* 2005). An issue then is whether it is sufficient to adopt an annualised health target alone, or whether to implement additional targets for identified shorter-term periods when the infection risks are greater and there is heightened outbreak potential. Future efforts to set health-targets could explore the merits of setting goals pertaining to both annual and episodic risks.

Results from this and similar QMRAs which aim to assess impacts of specific factors on risks to consumers provide other important management information. For example, having identified that winter coincides with heightened risk periods and that rainfall-induced ‘runoff events’ are primarily

responsible offers at least two possible ways to reduce risks. The utility may compare the economic and risk mitigation viability of adopting enhanced physical treatment methods over the winter period against adopting catchment management initiatives to minimise the amount of contaminants which can potentially be mobilised by rainfall. For the case study, other important information needs were identified – particularly the need for more microbial data (especially *Campylobacter* spp.) to reduce QMRA outcome uncertainties, and larger volume sampling at SP2 for protozoa in order to better quantify health risks from the system.

CONCLUSION

Of more general significance than the quantified results is the following premise: QMRA undertaken with an aim to assess impacts of explicitly identified and defined events on health risks to consumers can provide important information for risk management. QMRA has here provided awareness as to when higher-risk periods occur. Following application of a hydrograph separation technique to define and identify periods of runoff contribution entering a reservoir, QMRA was performed to conceptually quantify the relative health risks to drinking-water consumers emanating from rainfall-induced ‘runoff events’ entering a surface water reservoir. While the details of the approach were developed to suit the characteristics of the case study catchment and the available data, adaptation to other sites with different available data should be possible. Pertaining to the case study, the major proportion of annual infection risk was attributable to runoff event periods. Daily infection risks were greater in winter primarily due to the higher frequency of runoff events over those months. Under the model assumptions, the reservoir’s bacterial concentration transformation ratio did not substantially differ between seasons, abstractly suggesting that the heightened loading during the winter was primarily responsible for the associated higher risks.

ACKNOWLEDGEMENTS

This study has been performed with funding from the Australian Department of Education, Science and Training

as part of the MicroRisk project which is co-funded by the European Commission under the Fifth Framework Programme, Theme 4: “Energy, environment and sustainable development” (contract EVK1-CT-2002-00123). This document does not necessarily represent the opinion of the European Community. Thanks to the South Australian Department of Water, Land & Biodiversity Conservation and South Australia Water for provision of data. The corresponding author would also like to acknowledge support provided by the Australian government, the Centre for Water & Waste Technology, the University of New South Wales.

REFERENCES

- Atherholt, T. B., LeChevallier, M. W., Norton, W. D. & Rosen, J. S. 1998 Effect of rainfall on *Giardia* and *Cryptosporidium*. *J. AWWA* **90**(9), 66–80.
- Barbeau, B. & Payment, P. 2000 Evaluating the risk of infection from the presence of *Giardia* and *Cryptosporidium* in drinking water. *Quantitative Microbiology* **2**, 37–54.
- Bates, B. C. & Campbell, E. P. 2001 A Markov-chain Monte Carlo scheme for parameter estimation and inference in conceptual rainfall-runoff modelling. *Water Resour. Res.* **37**, 937–947.
- Blaser, M. J., Smith, P. F., Wang, W. L. & Hoff, J. C. 1986 Inactivation of *Campylobacter jejuni* by chlorine and monochloramine. *Appl. and Envir. Microbiol.* **51**, 307–311.
- Buswell, C. M., Herlihy, Y. M., Lawrence, L. M., McGuiggan, J. T. M., Marsh, P. D., Keevil, C. W. & Leach, S. A. 1998 Extended survival and persistence of *Campylobacter* spp. in water and aquatic biofilms and their detection by immunofluorescent antibody and rRNA staining. *Appl. and Envir. Microbiol.* **64**(2), 733–741.
- Chapman, T. 1999 A comparison of algorithms for stream flow recession and baseflow separation. *Hydrocarbon Process.* **13**, 701–714.
- Clark, R. M., Read, E. J. & Hoff, J. C. 1989 Analysis of inactivation of *Giardia lamblia* by chlorine. *Hydrological Processes* **115**(1), 80–90.
- Conover, W. J. 1999 *Practical nonparametric statistics*, 3rd ed. Wiley, NY.
- Cook, D., Duckworth W. M., Kaiser, M. S., Meeker, W. Q., Stephenson, W. R. 2000 Principles of maximum likelihood estimation and the analysis of censored data. In: *Beyond traditional statistical methods* Duckworth, W. & Stephenson, W. R. (Eds) Iowa State University, Ames, USA, chapter 2. Accessed 13th March 2006 at: http://www.public.iastate.edu/~stat415/meeker/ml_estimation_chapter.pdf
- Crabtree, K. D., Gerba, C. P., Rose, J. B. & Haas, C. N. 1997 Waterborne adenovirus: a risk assessment. *Water Sci. Technol.* **35**(11–12), 1–6.
- Curriero, F. C., Patz, J. A., Rose, J. B. & Lele, S. 2001 The association between extreme precipitation and waterborne disease outbreaks in the United States, 1948–1994. *Am. J. Pub. Health* **91**(8), 1194–1199.
- Devore, J. L. 2000 *Probability and statistics for engineering and the sciences*, 5th edn. Duxbury-Thomson Learning, Duxbury, UK.
- Efron, B. & Tibshirani, R. J. 1993 An introduction to the bootstrap. *Monographs on statistics and applied probability* 57. Chapman & Hall, NY.
- Gale, P. & Stanfield, G. 2000 Cryptosporidium during a simulated outbreak. *J. AWWA* **92**(9), 105–116.
- Gilks, W. R. S., Richardson, S. & Spiegelhalter, D. J. 1996 *Markov-Chain Monte Carlo in practice*. Chapman & Hall, London, UK.
- Haas, C. N. 1997 Importance of distributional form in characterising inputs to Monte Carlo risk assessments. *Risk. Anal.* **17**(1), 107–113.
- Haas, C. N., Crockett, C. S., Rose, J. B., Gerba, C. P. & Fazil, A. M. 1996 Assessing the risk posed by oocysts in drinking-water. *J. AWWA* **88**(9), 131–136.
- Haas, C. N., Rose, J. B. & Gerber, C. P. 1999 *Quantitative microbial risk assessment*. John Wiley & Sons, Inc., NY.
- Hijnen, W. A. M., Beerendonk, E. Medema, G. J. 2005 Elimination of Micro-organisms by Water Treatment Processes – A Review. Public Report for Kiwa Project #111444.100.003, Kiwa, Nieuwegein, The Netherlands.
- Hipsey, M. R., Brookes, J. D., Antenucci, J. P., Burch, M. D., Regel, R. H., Davies, C., Ashbolt, N. J. Ferguson, C. 2005 *Hydrodynamic distribution of pathogens in lakes and reservoirs*. Report, American Water Works Association Research Foundation, Denver, USA.
- Hollander, M. & Proschan, M. A. 1979 Testing to determine the underlying distribution using randomly censored data. *Biometrics* **35**, 393–401.
- Huber, W. C. 1993 Contaminant transport in surface water. In: Maidment, D. R. (ed.) *Handbook of Hydrology*. McGraw-Hill, Inc., NY, pp. 14.1–14.50.
- Kappenman, R. F. 1982 On a method for selecting a distributional model. *Commun. Stat. Theor. Methods* **11**(6), 663–672.
- Kistemann, T., Claßen, T., Koch, C., Dangendorf, F., Fischeder, R., Gebel, J., Vacata, V. & Exner, M. 2002 Microbial load of drinking water reservoir tributaries during extreme rainfall and runoff. *Appl. and Envir. Microbiol.* **68**(5), 2188–2197.
- Korich, D. G., Mead, J. R., Madore, M. S., Sinclair, N. A. & Sterling, C. R. 1990 Effects of ozone, chlorine dioxide, and monochloramine on *Cryptosporidium parvum* oocyst viability. *Appl. and Envir. Microbiol.* **56**, 1423–1428.
- Linden, K. G., Shin, G. & Sobsey, M. D. 2001 Comparative effectiveness of UV wavelengths for the inactivation of *Cryptosporidium parvum* oocysts in water. *Water Sci. Technol.* **43**(12), 171–174.
- Lopman, B., Vennema, H., Kohli, E., Pothier, P., Sanchez, A., Negrodo, A., Buesa, J., Schreier, E., Reacher, M., Brown, D., Gray, J., Iturriza, M., Gallimore, C., Bottiger, B., Hedlund, K., Torven, M., von Bonsdorff, C., Maunula, L., Poljsjak-Prijatelj, M., Zimek, J., Reuter, G., Szucs, G., Melegh, B., Svensson, L., van Duynhoven, Y. & Koopmans, M. 2004 Increase in viral

- gastroenteritis outbreaks in Europe and epidemic spread of new norovirus variant. *Lancet* **363**(9410), 682–688.
- Macler, B. A. & Regli, S. 1993 Use of microbial risk assessment in setting United States drinking water standards. *Int J. Food Microbiol.* **18**(4), 245–256.
- McCullagh, P. & Nelder, J. A. 1989 *Generalized Linear Models. of Monographs on Statistics and Applied Probability*, (57). Chapman & Hall, London, UK.
- Medema, G. J., Teunis, P. F. M., Havelaar, A. H. & Haas, C. N. 1996 Assessment of the dose–response relationship of *Campylobacter jejuni*. *Int J. Food Microbiol.* **30**, 101–111.
- Naumova, E. N., Christodouleas, J., Hunter, P. R. & Syed, Q. 2005 Effect of precipitation on seasonal variability in cryptosporidiosis recorded by the North West England surveillance system in 1990–1999. *J. Wat. Health* **3**(3), 185–196.
- Ott, W. R. 1995 *Environmental Statistics and Data Analysis*. Lewis Publishers, Boca Raton, FL.
- Payment, P. J., Seimiatycki, J., Richardson, L., Renaud, G., Franco, E. & Prevost, M. 1997 A prospective epidemiological study of gastrointestinal health effects due to the consumption of drinking water. *Int J. Environ. Health Res.* **7**, 5–31.
- Pouillot, R., Beaudou, P., Denis, J.-B. & Derouin, F. 2004 A quantitative risk assessment of waterborne cryptosporidiosis in France using second-order Monte Carlo simulation. *Risk Anal.* **26**(1), 1–17.
- Poulton, M., Colbourn, J. & Dennis, P. J. 1991 Thames Water's experience with *Cryptosporidium*. *Water Sci. Technol.* **24**, 21–26.
- Rose, J. B., Epstein, P. R., Lipp, E. K., Sherman, B. H., Bernard, S. M. & Patz, J. A. 2001 Climate variability and change in the United States: potential impacts of water- and foodborne diseases caused by microbiological agents. *Environ. Health Perspect.*, **109**(2), 211–219.
- Rose, J. B., Haas, C. N. & Regli, S. 1991 Risk assessment and control of waterborne giardiasis. *Am. J. Pub. Health* **81**(6), 709–713.
- Roseberry, A. M. & Burmaster, D. E. 1992 Lognormal distributions for water intake by children and adults. *Risk Anal.* **12**(1), 99–104.
- Roser, D. Ashbolt, N. 2004 *Transforming pathogen water quality data into source water monitoring and control information*. Draft report for cooperative research centre for water quality technology project 2.2.1 - 'Monitoring and Management of surface and subsurface source waters', Centre for Water & Waste Technology, University of New South Wales, Sydney, Australia.
- Signor, R. S., Roser, D. J., Ashbolt, N. J. & Ball, J. E. 2005 Quantifying the impact of runoff events on microbiological contaminant concentrations entering surface drinking source waters. *J. Wat. Health* **3**(4), 453–468.
- Sinton, L. W., Finlay, R. K. & Lynch, P. A. 1999 Sunlight inactivation of fecal bacteriophages and bacteria in sewage-polluted seawater. *Appl. and Environ. Microbiol.* **65**(8), 3605–3613.
- Tallaksen, L. M. 1995 A review of baseflow recession analysis. *J. Hydrol.* **165**, 349–370.
- Teunis, P. F. M. & Havelaar, A. H. 1999 Cryptosporidium in drinking water: evaluation of the ILSI/RSI quantitative risk assessment framework. Report no. 284 550 006, National Institute of Public Health and the Environment, Bilthoven, The Netherlands.
- Teunis, P. F. M. & Havelaar, A. H. 2002 Risk assessment for protozoan parasites. *Int J. Biodeterioration and Biodegradation* **50**, 185–193.
- Teunis, P. F. M., Evers, E. G. & Slob, W. 1999 Analysis of variable fractions resulting from microbial counts. *Q. Microbiology* **1**, 63–88.
- Teunis, P. F. M., Medema, G. J., Kruidenier, L. & Havelaar, A. H. 1997 Assessment of the risk of infection by *Cryptosporidium* or *Giardia* in drinking water from a surface water source. *Water Res.* **31**(6), 1333–1346.
- Teunis, P. F. M., van der Heijden, O. G., van der Giessen, J. W. B. & Havelaar, A. H. 1996 *The dose–response relation in human volunteers for gastro-intestinal pathogens*. Report, National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands.
- Teunis, P., Davison, A. Deere, D. 2004 *Short-term fluctuations in drinking water quality and their significance for public health*. Draft report, World Health Organization, Geneva, Switzerland.
- United States Environmental Protection Agency (USEPA) 1999 Method 1623 -*Cryptosporidium* and *Giardia* in Water by Filtration/IMS/IFA. Office of Water, United States Environment Protection Agency, Washington DC, USA.
- Walker, F. R. & Stedinger, J. R. 1999 Fate and transport model of *Cryptosporidium*. *J. Environ. Engrg.* **125**(4), 325–333.
- Westrell, T., Teunis, P., van den Berg, H., Lodder, W., Katelaars, H., Stenström, T. A. & de Roda Husman, A. M. 2004 Short and long term fluctuations of norovirus concentrations in surface water and their implications for public health. Paper III. In: *Microbial risk assessment and its implications for risk management in urban water systems*, PhD thesis (Westrell, T.) Linköping University, Sweden.
- World Health Organization (WHO) 2004 *Guidelines for drinking water quality*, 3rd edn. WHO, Geneva, Switzerland.
- Zwietering, M. H. & van Gerwen, S. J. C. 2000 Sensitivity analysis in quantitative microbial risk assessment. *Int. J. Food Microbiol.* **58**, 213–221.

First received 1 February 2006; accepted in revised form 27 July 2007

A. ADENDUM (LIKELIHOOD MODEL TO ESTIMATE ϵ_R)

The reservoir transformation ratio was defined as the ratio of the mean observed micro-organism concentration at SP2 to the mean observed micro-organism concentration at SP1, allowing for the proportion of time that the incoming flows were in baseflow and runoff event modes. When each sample result consisted of the number of (oo)cysts counted in the sample n ; the sample volume v ; the number x of fluorescent (oo)cysts seeded into a parallel sample to assess the recovery fraction; and the number s of seeded (oo)cysts recovered by the same enumeration method from the parallel sample, consider a dataset describing microbial water quality entering a reservoir of:

$m_{R,j}$ samples taken during each j th of a total k_R independent runoff event periods-

$(n_{R,ij}, v_{R,ij}, x_{R,ij}, s_{R,ij})$ where $i = 1, 2, 3, \dots, m_{R,j}$ and $j = 1, 2, 3, \dots, k_R$

$m_{B,b}$ samples taken during each b th of a total k_B independent baseflow periods-

$(n_{B,ab}, v_{B,ab}, x_{B,ab}, s_{B,ab})$ where $a = 1, 2, 3, \dots, m_{B,b}$ and $b = 1, 2, 3, \dots, k_B$

and describing microbial quality of water exiting the reservoir at a point downstream:

k_r samples-

$(n_{r,c}, v_{r,c}, x_{r,c}, s_{r,c})$ where $c = 1, 2, 3, \dots, k_r$

The number of organisms present in a sample of volume $v_{R,ij}$ is a random integer described by Poisson processes. The likelihood function to estimate the observed mean concentration of the j th 'runoff event' period $\lambda_{R,j}$ is:

$$L_1(\lambda_{R,j} | n_{R,ij}, v_{R,ij}, s_{R,ij}, x_{R,ij}) = \prod_{i=1}^{m_{R,j}} \left[\frac{(\lambda_{R,j} v_{R,ij} / s_{R,ij})^{n_{R,ij}}}{n_{R,ij}! e^{\lambda_{R,j} v_{R,ij} / s_{R,ij}}} \right]_{ij}$$

When Λ_R is the runoff event pathogen concentration that varies between 'runoff event' periods with observed arithmetic mean value $\bar{\lambda}_R$, then the likelihood function is:

$$L_2(\bar{\lambda}_R | n_{R,ij}, v_{R,ij}, s_{R,ij}, x_{R,ij}) = \prod_{j=1}^{k_R} \left[\bar{\lambda}_R \int_0^{\infty} \prod_{i=1}^{m_{R,j}} \left[\frac{(\lambda_{R,j} v_{R,ij} / s_{R,ij})^{n_{R,ij}}}{n_{R,ij}! e^{\lambda_{R,j} v_{R,ij} / s_{R,ij}}} \right] d\lambda_{R,j} \right]_j$$

Similarly, Λ_B is the baseflow pathogen concentration that varies between independent 'baseflow' periods with observed arithmetic mean $\bar{\lambda}_B$, the likelihood function is:

$$L_3(\bar{\lambda}_B | n_{B,ab}, v_{B,ab}, s_{B,ab}, x_{B,ab}) = \prod_{b=1}^{k_B} \left[\bar{\lambda}_B \int_0^{\infty} \prod_{a=1}^{m_{B,b}} \left[\frac{(\lambda_{B,b} v_{B,ab} / s_{B,ab})^{n_{B,ab}}}{n_{B,ab}! e^{\lambda_{B,b} v_{B,ab} / s_{B,ab}}} \right] d\lambda_{B,b} \right]_b$$

NB: the derivation of Equation 3 was similar, except where probability density functions describing the variability of Λ_R/Λ_B were input instead of constant values $\bar{\lambda}_B$ and $\bar{\lambda}_R$ in the above two equations.

The organism concentration at a point downstream was interpreted as a ratio ϵ_r of the concentrations observed at SP1, accounting for the proportions of time A_R and A_B that runoff and baseflow conditions respectively predominated. It can be inferred that the observed downstream mean organism concentration $\bar{\lambda}_r$ is:

$$\bar{\lambda}_r = \epsilon_r (A_R \bar{\lambda}_R + A_B \bar{\lambda}_B)$$

The organisms counted in a sample taken downstream at any point in time come from a water body where the organisms are randomly (Poisson) distributed - the likelihood function to estimate the downstream concentration λ_r at the time of the c th sample is:

$$L_4(\lambda_{r,c} | n_{r,c}, v_{r,c}, s_{r,c}, x_{r,c}) = \left[\frac{(\lambda_{r,c} v_{r,c} / s_{r,c})^{n_{r,c}}}{n_{r,c}! e^{\lambda_{r,c} v_{r,c} / s_{r,c}}} \right]_c$$

When Λ_r is the overall downstream organism concentration that varied between sampling times with observed mean value $\bar{\lambda}_r = \epsilon_r (A_R \bar{\lambda}_R + A_B \bar{\lambda}_B)$, the contagious likelihood function is:

$$L_5(\epsilon_r, \bar{\lambda}_R, \bar{\lambda}_B | n_{r,c}, v_{r,c}, s_{r,c}, x_{r,c}, A_R, A_B) = \prod_{c=1}^{k_r} \left[\epsilon_r (A_R \bar{\lambda}_R + A_B \bar{\lambda}_B) \int_0^{\infty} \frac{(\lambda_{r,c} v_{r,c} / s_{r,c})^{n_{r,c}}}{n_{r,c}! e^{\lambda_{r,c} v_{r,c} / s_{r,c}}} d\lambda_{r,c} \right]_c$$

The likelihood function L_r to estimate ϵ_r (and the redundant parameters $\bar{\lambda}_R$ & $\bar{\lambda}_B$) is:

$$L_r = L_2.L_3.L_5 \dots \quad (A.1)$$