Introduction to Exposure Assessment

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GOAL

An exposure assessment is a two part process that includes 1) evaluation of pathways that allow microorganisms to be transported from the source to the point of contact with human beings and 2) estimation of the amount of exposure that is possible between humans and contaminants. Conducting a realistic exposure assessment is important for good risk characterization as well as risk management.

The goal of chapter is to become familiar with fundamental computations for 1) changes in microbial concentrations, e.g. die-off and reduction, 2) exposure doses from different exposure routes, and 3) risk characterization and management.

CHANGE IN MICROBIAL CONCENTRATIONS

Microbial concentrations may vary from the source to the point where a human may contact a pathogen. Many environmental factors, such as temperature, sunlight (ultraviolet (UV) radiation), moisture content, pH, salinity, nutrients, organics, and other chemicals, may affect microbial fate and transportation.

Some microorganisms may grow or survive longer in moisten and nutrient rich environments, e.g. foods and soils. Growth of microorganisms, e.g. *Staphylococcus aureus*, *Salmonella*, *Clostridium perfringens*, *Campylobacter*, *Listeria monocytogenes*, *Vibrio parahaemolyticus*, *Bacillus cereus*, and Entero-pathogenic *Escherichia coli*, cause food poisoning and this is a serious problem in the food industry. Regrowth of microbes, e.g. fecal coliform, *E. coli*, and enterococci in warm environments complicates the use of these microbes as fecal indicators of pollution for recreational water quality assessment. Lower temperatures may support microorganisms to survive longer.

Pathogens may decrease in concentrations due to dilution or natural die-off as they are released into different environments from the original host environments. Sunlight, dry, and low nutrients are major environmental factors that accelerate microbial die-off. Survival rates differ with microorganisms and environmental matrix (water, air, soil, and fomite). In general, microbes survive shorter in seawater compared with freshwater. Microbial inactivation rates in surface water are faster than groundwater.

Transportation of microorganisms can be affected by various environmental conditions, such as distance from the source to the exposure point, flow rate, exchange rate, suspended solid, permeability, and soil sorption. Fate and transportation models for water and air/fomites are discussed in Chapters 7 and 8.

% vs. Log Reduction

Reduction ratios are often expressed in percentage (%) or logarithm ($\log_{10}$ or ln) (Eq. 6.1 and 6.2). It should be noted that given equations can be used to estimate increases in microbial concentrations as well reductions.

$$\text{\% Reduction, } \%R = \left(1 - \frac{M_{final}}{M_{initial}}\right) \cdot 100(\%) \tag{6.1}$$

$$\begin{aligned} \text{Log}_{10}\text{ Reduction, } LR &= -\log_{10}\left(\frac{M_{final}}{M_{initial}}\right) \\ &= \log_{10}(M_{initial}) - \log_{10}(M_{final}) \end{aligned} \tag{6.2}$$

Where *%R* is the % reduction, *LR* is the log10 reduction, $M_{initial}$ is the initial number of microorganisms (typically expressed as CFU or PFU), and M_{final} is the final number of microorganisms (after reduction). In general, % reduction is more commonly used in the public sector while $\log_{10}$ reduction is typically used by engineers. It is useful to be able to convert from % reduction to $\log_{10}$ reduction (Eq. 6.3) or vice-versa (Eq 6.4).

$$\text{\% Reduction, \%R} = \left(1 - 10^{-LR}\right) \cdot 100(\%) \tag{6.3}$$

$$\text{Log}_{10}\text{ Reduction, LR} = -\log_{10}\left(1 - \frac{\%R}{100}\right) \tag{6.4}$$

If the initial concentration of a microorganism and reduction (% or $\log_{10}$) are known, the final concentrations can be calculated using Eqs. 6.5 and 6.6.

$$M_{final} = M_{initial} \cdot \left(1 - \frac{\%R}{100}\right) \tag{6.5}$$

$$M_{final} = M_{initial} \cdot 10^{-LR} \tag{6.6}$$

Inactivation Rate

Prediction of survival of microorganisms is an essential part of exposure assessment. Microorganisms die naturally over time or may be inactivated by UV radiation, the presence of materials toxic to them or other features of the environment. An inactivation rate is often expressed in T90 and T99, which are time required for 90 and 99% reductions. The rate may also be expressed as an inactivation coefficient, or *k*-value, that is a ratio of the initial and final titer in log scale over the survival time. A common unit of k-value is $\log_{10}$ per hour ($\log_{10}$/hr). The *k*-values can be computed based on T90 and T99 (Eq. 6.7) or initial, final concentrations, and survival time are available from the

literature (Eq. 6.8). If you conduct an inactivation study or detailed data are available, the *k*-values can be calculated using all data (Eq. 6.9) via linear regression.

$$k_a = \frac{-\log(1-99/100)}{T99} \tag{6.7}$$

$$k_b = \frac{\log_{10}(M_0)-\log_{10}(M_t)}{T_{survival}} \tag{6.8}$$

k_c = slope (a) of a log-linear equation, $y = ax + b$ (6.9)

In equation 6.9, y is the $\log_{10}$ reduction ($\log_{10}(M_t/M_0)$), x is the monitoring period or survival time (hr), and b is the intercept. Note that assuming k does not vary with time also assume that the features of the environment causing the inactivation (solar radiation, chemical concentrations, etc) also do not vary with time.

Once k-values are obtained, you can predict the microbial concentration at time t (Eqs. 6.10 and 6.11). Predicting microbial concentrations at different times is useful for determining how long it may take to reduce concentrations to a target level (Eq 13).

$$M_t = M_0 \cdot 10^{-kt} \tag{6.10}$$

$$M_t = M_0 \cdot 10^{-(kt+b)} \tag{6.11}$$

$$T_p = -\frac{1}{k} \cdot \log\left(\frac{M_p}{M_0}\right) \tag{6.12}$$

EXAMPLE 6.1 Recovery and Inactivation on fomites

Scenario: 100 µL of titered *Staphylococcus aureus* (3.9 x10^8 colony formed unit (CFU)/mL) solution was inoculated onto 10 cm^2 non-porous surfaces or fomites (aluminum, ceramic, glass, plastic, stainless steel, and wood laminated). Fomite samples were wiped using pre-moistened antistatic wipes and the wipes were placed in 50-mL test tubes contained 10 mL phosphate buffer saline plus 0.01% Tween-80. Test tubes were vortexed and samples were diluted (-3 $\log_{10}$). 100 µL from the diluted samples were dispensed on to Mannitol Salt Agar (MSA) plates. Average colony counts were 317 on the MSA plates from the first wipes just after the inoculation of fomite. Fomites samples were collected for 3 weeks and the results are summarized in the Table 6.1.

Tasks:

- Calculate initial recovery of *S. aureus* from fomites using the wipe method.
- Calculate k-values based on T99, the initial and final concentration over the survival time, and log-linear regression equation.

- Compare measured concentrations with models based on calculated k-values
- Predict how long it would take *S. aureus* level to 1 CFU/cm^2.

Table 6.1 Survival of *S. aureus* on fomites

Survival time (hr)	Average concentration (CFU/10cm^2)
0	(calculate)
22	2.27 x10^6
41	1.17 x10^6
66	5.62 x10^5
115	2.35 x10^4
162	1.65 x10^4
332	5.29 x10^2
522	1.30 x10^1

Solution steps:

1. Calculate the initial recovery using Eq. 1

   $$\%\text{ Initial Recovery, } \%R = \left(1 - \frac{M_{final}}{M_{initial}}\right) \cdot 100(\%)$$

   $M_{inoculated}$ = Titer concentration x inoculated volume on fomites

   $$= \frac{3.9\times10^8 cfu}{mL} \cdot \frac{0.1mL}{10cm^2} = 3.9\times10^7 (cfu/10cm^2)$$

   $$M_{recovered} = \frac{317cfu}{plate} \cdot \frac{plate}{0.1mL} \cdot \frac{10ml}{10cm^2} \cdot 10^3 = 3.17 \text{ x } 10^7 \text{ (CFU/10cm}^2\text{)}$$

   % Initial Recovery = 81.2%

2. Calculate % and Log_{10} reductions using Eqs. 6.1 and 6.2.
3. Make a figure: y-axis is % reduction and x-axis is time.
4. Use the figure and estimate T99 by extrapolation from the best fit curve based on % reductions from 41 to 115 hours. In this case, a power equation fit better than a linear equation (Figure 6.1). T99 = 93.5 hrs

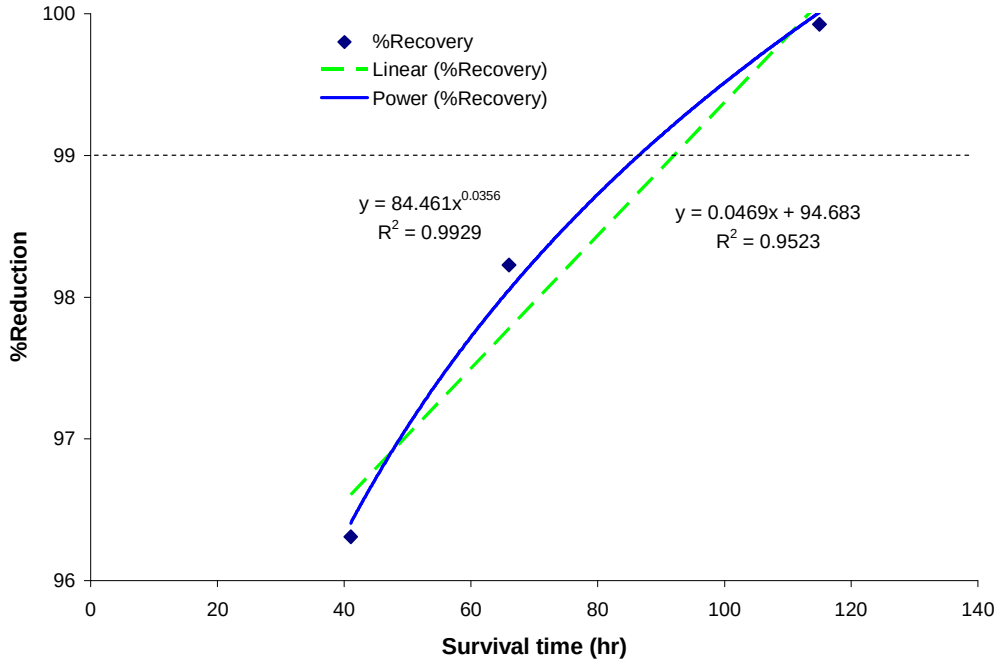

Figure 6.1 Extrapolation of T99

5. Calculate the k-value based on T99 (Eq. 6.7)

$$k_a = \frac{-\log(1-99/100)}{T99} = \frac{-\log(1-99/100)}{93.5hrs} = \boxed{0.0214}$$

6. Calculate k-value only based on the initial and final concentration over time (Eq. 6.8)

$$k_b = \frac{\log_{10}(M_0)-\log_{10}(M_t)}{T_{survival}} = \frac{\log_{10}(3.17\times10^7)-\log_{10}(13)}{522hrs} = \boxed{0.0122}$$

7. Calculate k-value (slope) and intercept of a log-linear regression for reduction ratio over time (Eq. 6.9). Using EXCEL,
   Kc = SLOPE(known_y's,known_x's) = $\boxed{0.0111}$
   Intercept = INTERCEPT(known_y's,known_x's) = 0.986

8. Substitute obtained k-values into the inactivation models (Eqs. 6.10 and 6.11)
   K_a and K_b into $M_t = M_0 \cdot 10^{-kt}$ for
   K_c into $M_t = M_0 \cdot 10^{-(kt+b)}$ for

9. Make a figure (y-axis = *S. aureus* concentration and x-axis = time in hour) that contains measured concentration and predicted concentrations from the models based on three k-values (Figure 6.2).

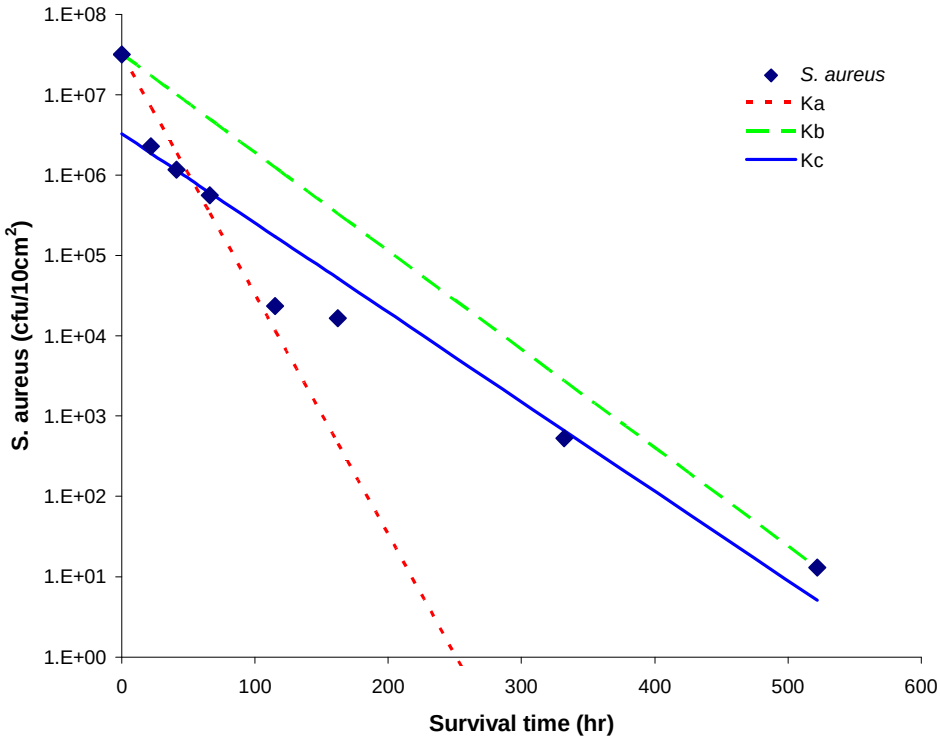

Figure 6.2 *S. aureus* concentration on fomites based on measurements and inactivation models

10. Compute how long it would take *S. aureus* concentration to be 1 CFU/10cm^2 from the last observed level. Since the initial rapid reduction is over at this point, it is unnecessary to use interception. Thus, Eq 6.12 can be used for prediction time using these three k-values.

$$T_p = -\frac{1}{k} \cdot \log\left(\frac{M_p}{M_0}\right) = -\frac{1}{k} \cdot \log\left(\frac{1}{13}\right)$$

2 days based on K_a (T99) and 4 days based on K_b and K_c

Computations can be done using Microsoft EXCEL (Table 6.2)

Table 6.2 Summary computations using EXCEL

	A	B	C	D	E	F	G
1		S. aureus	Reduction		Model		
2	hr	CFU/10cm^2	%	Log_{10}	k_a	k_b	k_c+b
3	0	3.17E+07	=(1-B3/$B$3)*100	=-LOG(B3/$B$3)	=$B$3*10^-($B$17*A3)	3.17E+07	3.27E+06
4	22	2.27E+06	92.84	1.15	6.97E+06	1.71E+07	1.86E+06
5	41	1.17E+06	96.31	1.43	1.88E+06	9.99E+06	1.14E+06
6	66	5.62E+05	98.23	1.75	3.37E+05	4.94E+06	6.03E+05
7	115	2.35E+04	99.93	3.13	1.15E+04	1.24E+06	1.72E+05
8	162	1.65E+04	99.95	3.28	4.54E+02	3.30E+05	5.16E+04
9	332	5.29E+02	99.998	4.78	3.75E-03	2.75E+03	6.63E+02
10	522	1.30E+01	99.99996	6.39	7.82E-09	1.30E+01	5.11E+00
11							
12	Best fit for T99		%R=at^b				
13	%R	a	b	T			
14	99	84.461	0.035	=(A14/B14)^(1/C14)			
15							
16		k-value	intercept				
17	k_a	=(-LOG(1-99/100))/D14					
18	k_b	=(LOG(B3)-LOG(B10))/A10					
19	k_c	=SLOPE(D3:D10,A3:A10)	=INTERCEPT(D3:D10,A3:A10)				
20							
21	How long does it take S. aureus level to be 1CFU/cm^2						
22	from the last observed concentration?						
23		S. aureus					
24	Initial (M_0)	13					
25	Final (M_p)	0.1					
26							
27	Predicted time						
28	Hr	Days	k-value				
29	=-1/B17*LOG($B$25/$B$24)	=A29/24	ka				
30	173	7	kb				
31	190	8	kc				

EXPOSURE DOSE

One of the simplest methods for estimating exposure dose is shown in Eq. 6.13.

$$D = C \cdot V \tag{6.13}$$

where D is the exposure dose, C is the concentration of microorganism, and D is the amount of the containment to which a person is exposed.

Exposure Pathways: Water, Air, Soil, and Food

Common routes of exposure include inhalation, ingestion, and dermal contact. Because dose-response data for microorganisms through dermal exposures are limited, this section primarily focuses on inhalation and ingestion as route of exposure. The amount of air inhaled, water and food ingested by an individual vary with age, sex, health condition, and activities. Personal exposure factors can be obtained from the literature or the US Environmental Protection Agency (US EPA 1991; 1997). Table 6.2 summarizes common exposure pathways and parameters for case scenarios.

Eq. 13 shows how to calculate the exposure dose in the simplest way. It can be rewritten because the amount of contaminants a person contacts depends on the uptake rate and duration of exposure (Eq. 6.14).

$$D = C \cdot R \cdot T \tag{6.14}$$

Where D is the exposure dose expressed as numbers of microorganisms inhaled or ingested (CFU or plaque forming unit (PFU)), C is the concentration of the microorganisms in air, water, food, or soil (CFU or PFU per liter (L) or gram (g)), V is the total volume or mass of air inhaled or water, food, or soil ingested (L^{-1} or g^{-1} or), R is the intake rate (L or g per time, e.g. hour, day, or year), and T is the exposure duration (hr, day, yr).

EXAMPLE 6.2 Ingestion of Water

Scenario: Drinking water may contain low levels (1-20 pfu/1000L) of enteric viruses.

Task: Calculate a conservative annual exposure dose of enteric viruses for an adult and a child through drinking tap water.

Solution steps:

1. Use the upper rage of concentration 20pfu/1000L and 90^{th} percentile daily intake rates; adult 2.3 L/day and a child (1-10 years old) 1.3 L/day (Table 3-1) for making conservative estimates**.**
2. Substitute the given and assumed parameters into the Eq. 6.14.

$$D = C \cdot R \cdot T$$

For an adult, $D = \frac{20\,pfu}{1000L} \cdot \frac{2.3L}{day} \cdot \frac{365days}{yr} =$ 17 pfu/yr

For a child $D = \frac{20\,pfu}{1000L} \cdot \frac{1.3L}{day} \cdot \frac{365days}{yr} =$ 9 pfu/yr

Note: write down all units even if a calculation is easy.

EXAMPLE 6.3 Air

Scenario: Bioaerosols in an office room were collected in AGI-30 impingers, which contained 20 mL of 0.3 mM phosphate buffer water, using a sampling pump with 12.5 L/min flow rate for 10 minutes. 20-mL samples were concentrated to 1 mL using Amicon centrifuge devices. From the concentrated samples, 100 µL were dispensed on TSA agar plates. On average, 10 colonies as total bacteria were observed from the plates after 24 hours incubation at 37ºC. Assume the impinger is 100% efficient in collecting bacteria in the room air.

Task: Estimate how many total bacteria an adult could inhale during a regular work day in the office. Assume that bacteria levels are constant and that air exchange and settling of bacteria are negligible.

Solution steps:

1. Calculate bacterial concentrations in the air

$$= \frac{count(cfu)}{plate} \cdot \frac{plate}{dispense(mL)} \cdot \frac{concentrated(mL)}{sample_{liquid}(mL)} \cdot \frac{sample_{liquid}(mL)}{Flowrate_{air}(L/\min) \cdot time(\min)}$$

$$= \frac{10cfu}{plate} \cdot \frac{plate}{0.1mL} \cdot \frac{1mL}{20mL} \cdot \frac{20mL}{12.5L_{air}/\min \cdot 10\min} = 0.8 \text{ CFU/L air}$$

= total bacteria 800 CFU/m^3 air

2. Since the case scenario was in office, you could assume that the person's job is desk work. The inhalation rate, *R*, for a sedentary adult indoors, from Table 1 is 0.5 m^3/hr. You can assume that the exposure period, *T*, is a typical work day or 8 hours. Substitute the given and assumed parameters into the Eq. 2.

$$D = C \cdot R \cdot T$$

$$D = 800\frac{cfu}{m^3} \cdot 0.5\frac{m^3}{hr} \cdot 8\frac{hr}{day} =$$ total bacteria 3200 CFU/working day

Table 6.3 Exposure pathways and personal exposure factors **1/2**

Exposure pathway	Scenario	Intake	Confidence rating	Reference
Ingestion of potable water	Residential	2 L/day (adult), 1 L/day (child),	Upper percentile	US EPA 1980; 1991a,b
		0.30 L/day (1- 10 yrs old)	Mean	Ershow and Cantor 1989;
		0.97 L/day (11-19 yrs old)	Mean	US EPA 1997
		1.4 L/day (adult)	Mean	
		1.3 L/day (1- 10 yrs old)	90^{th} percentile	
		1.7 L/day (11-19 yrs old)	90^{th} percentile	
		2.3 (adult)	90^{th} percentile	
	Industrial	1 L		US EPA 1991b
Ingestion of water while swimming	Recreational	50 mL/hr and 2.6 hr/swim		Covello and Merkhofer 1993
		20-50 mL/hr		WHO 2003
		37 (0-145) mL/45 min (child)	Mean	Dufour et al. 2006
		16 (0-53) mL/45 min (adult)	Mean	
Ingestion of soil and dust	Residential	100 mg/day (child)	Mean	US EPA 1997
		400 mg/day (child)	Upper percentile	
		50 mg/day (adult)	Mean	
		200 mg/day (child)	Conservative mean	US EPA 1991b
		100 mg/day (adult)		
		10 g/day (Pica child)	Mean	Calabrese et al. 1989
				US EPA 1997
	Industrial	50 mg/day		US EPA 1991b

Table 6.3 Exposure pathways and personal exposure factors **2/2**

Exposure pathway	Scenario	Intake	Confident rating	Reference
Inhalation of contaminants	Residential	20 m^3/day (adult) 12 m^3/day(child)		US EPA 1991b
	Industrial	20 m^3/day		US EPA 1991b
	Long-term	15.2 m^3/day (> 18 yrs old males) 11.3 m^3 /day (> 18 yrs females) 15.3 /day (9-18 yrs males) 12.3 m^3/day (9-18 yrs females) 7.4 /day (0-8 yrs old)	Mean	Layton 1993 US EPA 1997
	Short-term (Indoor)	Activity (Adult and child) Rest: 0.4 and 0.3 m^3/hr Sedentary: 0.5 and 0.4 m^3/hr Light: 1.0 and 1.0 m^3/hr Moderate: 1.6 and 1.2 m^3/hr Heavy: 3.2 and 1.9 m^3/hr	Mean	Layton 1993 US EPA 1997
Consumption of home growth produce	Agricultural	42 g (fruit) 80 g (vegetable)		US EPA 1991b
Consumption of locally caught fish	Recreational	54 g		US EPA 1991b

Exposure Pathway: Fomites

Fomites, which are inanimate objects or surfaces, can be easily contaminated with infectious organisms and thus that may play a key role in disease transmission and in the spread of pathogens (Boone and Gerba 2007). All microorganisms on a fomite do not necessarily transfer to a person's hand when a person contacts the fomite. Similarly, a pathogen may not be transferred from a person's hands to their mouth. The proportion of microorganism transferred affects exposure doses. Rusin et al. (2002) summarized transfer efficiencies of bacteria and phage from fomites to hands and from hand to mouth; these are shown in Table 6.4.

Table 6.4 Transfer efficiency (%) from fomite to hand and from hand to mouth

Organisms	Fomite-to-Hand		Hand-to-Mouth
Micrococcus luteus	Dishcloth	0.04	41
	Sponge	0.03	
	Faucet	40	
	Carrot	0.21	
	Hamburger	0.06	
	Phone receiver	42	
	Laundry 100%cotton	0.13	
	Laundry 50:50 cotton/polyester	0.06	
PRD-1 (bacteriophage)	Dishcloth	0.03	34
	Sponge	0.02	
	Faucet	33	
	Carrot	0.35	
	Hamburger	0.01	
	Phone receiver	66	
	Laundry 100%cotton	0.005	
	Laundry 50:50 cotton/polyester	0.0005	
Serratia rubidea	Dishcloth	0.0045	34
	Sponge	0.0037	
	Faucet	28	
	Carrot	0.12	
	Hamburger	0.002	
	Phone receiver	38	
	Laundry 100%cotton	0.003	
	Laundry 50:50 cotton/polyester	0.0009	

Source: Rusin et al. 2002

EXAMPLE 6.4 Fomite Exposure

Scenario: A person's hands were contaminated with 1 mg of feces after he went to the bathroom. The person used a public phone and 10% of feces were transferred to the phone receiver.

Task: Calculate how many people could ingest at least one microorganism after the person contaminated the phone. Chose one microorganism listed below (Table 6.5) and assume that everyone put his or her hand to the lip (mouth) one time after using the phone.

Table 6.5 Concentration of enteric pathogens in feces

Microorganisms	Concentrations (g^{-1})
Total coliform	10^7-10^9
Fecal coliform	10^6-10^9
Salmonella spp.	10^4-10^{11}
Shigella	10^5-10^9
Ascaris	10^4-10^5
Enteroviruses	10^3-10^{7a}
Rotavirus	10^{10b}
Adenovirus	10^{12b}
Protozoan parasites	10^6-10^7

Source: Feachem et al. 1983
[a]cell culture assays
[b]electron microscopic observation of viral particle

Solution steps:

1. For this example, we will use total coliforms. Use the concentration of total coliform in feces, 10^9/g (Table 1) for this computation. The number of total coliform that could be transferred from the hand to phone were:

   Total coliform = $\frac{10^9}{g} \cdot \frac{g}{10^3 mg} \cdot 1mg \cdot 0.1 =$ $\boxed{10^5 \text{ on a phone}}$

2. Make a table using EXCEL. Explain the equations you put into the EXCEL sheet. The first person could ingest 1.6 x 10^4 (=10^5x0.4x0.4), the 2nd person ingest residual from the first person times 0.16, and so on.

3. 23 people could ingest at least one total coliform by using the contaminated phone.

Computations can be done using Microsoft EXCEL (Table 6.6)

Table 6.6 Summary Computation Table using EXCEL

	A	B	C	D
1	Person	Residual	Hand	Mouth
2	0	100000		
3	1	=B2-C3	=B2*0.4	=C3*0.4
4	2	36000	24000	9600
5	3	21600	14400	5760
6	4	12960	8640	3456
7	5	7776	5184	2074
8	6	4666	3110	1244
9	7	2799	1866	746
10	8	1680	1120	448
11	9	1008	672	269
12	10	605	403	161
13	11	363	242	97
14	12	218	145	58
15	13	131	87	35
16	14	78	52	21
17	15	47	31	13
18	16	28	19	8
19	17	17	11	5
20	18	10	7	3
21	19	6	4	2
22	20	4	2	1
23	21	2	1	1
24	22	1	1	0

RISK CHARACTERIZATION AND MANAGEMENT

Once you estimate exposure dose, the probabilistic infectious risks can be calculated by plugging obtained doses into infectious dose-response models (exponential or beta-Poisson). If a dose-response model for a particular route of exposure is not available for the target microorganism, you may need to use available parameters for other similar microorganisms or another exposure route. For exposure assessment, it is important to state all assumptions and parameters you used for the calculation.

Retrospective Assessment

Exposure can also be approached backward from risk characterization to exposure assessment. For example, if you know an infectious risk, you could estimate the number of microorganisms ingested or concentrations of contaminants. Exponential and beta Poisson models can be rewritten for this approach (Eqs. 6.15 and 6.16) and the corresponding concentration of microorganisms can be estimated using equation 5.

$$D(P_i) = -\frac{\ln(1-P_i)}{r} \tag{6.15}$$

$$D(P_i) = \beta\left((1-P_i)^{-1/\alpha} - 1\right) \tag{6.16}$$

$$C(P_i) = \frac{D(P_i)}{V} \tag{6.17}$$

Where P_i is the probability of infection (ability of the organism to establish and reproduce in the human host), D is the exposure dose expressed as numbers of microorganisms ingested, V is the volume of contaminant intake and α, β, r are constants for specific organism that define the dose-response in an Exponential or beta Poisson model. Another version of the beta-Poisson model frequently used in microbial risk assessment is given in equation 6.18:

$$D(P_i) = \frac{N_{50}\left[(1-P_i)^{-1/\alpha} - 1\right]}{2^{1/\alpha} - 1} \tag{6.18}$$

Where N_{50} is the median infectious dose (the dose at which 50% of the population is infected).

EXAMPLE 6.5 Retrospective Exposure Assessment

Scenario: A local health department reported that 1% of people who purchased fresh vegetable from an organic market infected with *E. coli* O157. Prior studies have shown that the dose-

response data for ingestion exposure to *E. coli* O157 are best fit by the beta-Poisson dose-response model and that the parameters of the model are β=1.78×10^6 and α=0.178.

Task: Estimate the concentration of *E. coli* O157 in the vegetable.

Solution steps:

1. Use the dose-response parameters for pathogenic *E. coli* and plug the parameters to the revised version of Bata-Poisson equation (Eq. 4).
   $D(P_i) = \beta\left((1-P_i)^{-1/\alpha} - 1\right)$
   = 1.78×10^6 × ((1-0.01)^(-1/0.178)-1) = 10^5 of *E. coli* O157 could be ingested
2. Estimate the concentration of *E. coli* O157 in the contaminated vegetable. Assume that people ate 80 g of the vegetable (Table 1). Using Eq. 5,

$$C(P_i) = \frac{D(P_i)}{V} = 10^5/80\text{g} = \boxed{1250/\text{g}}$$

Exposure Reduction

Chlorination and boiling of water and heating of food are practices that have been used to reduce concentrations of microorganisms in food and water for centuries. Despite the introduction of advanced technologies for reduction in pathogens or inactivation of pathogens into water and wastewater treatment plants and the food industry, many outbreaks still occur worldwide. Recently, novel means for air and hand sanitation have become popular means for reducing microbial numbers. QMRA can be a useful tool to examine or estimate the reductions of microorganisms necessary to protect public health.

EXAMPLE 6.6 Hand Wash

Scenario: A graduate student went to a wastewater treatment plant to collect untreated wastewater samples for his research. During sampling, 1 mL of raw sewage splashed onto his bare hands. He washed his hands afterward using an antibiotic soap, which claims to kill 99% of bacteria. The concentrations of microorganisms in the raw sewage are given in Table 6.7.

Tasks:

- Estimate how many fecal coliforms (note that fecal coliforms are a class of "indicator organisms") are still on his hands after washing.
- How many total reductions are required to reduce fecal coliform level to be less than 1 CFU on the hands?

Table 6.7 Microorganism concentrations in untreated municipal wastewater

Microorganisms	Concentrations (100 mL^{-1})
Fecal coliforms	10^5-10^7
Fecal streptococci	10^4-10^6
Clostridium perfringens	10^3-10^5
Salmonella	10^2-10^4
Shigella	1-10^3

Pseudomonas aeruginosa	10^3-10^4
Enteric viruses	10^3-10^4
Helminth ova	1-10^3
Giardia lamblia cysts	10-10^4
Cryptosporidium oocysts	10-10^5
Entamoeba histolytica cysts	10^2-10^5

Source: National Resource Council 1996

Solution steps:

1. Calculate the initial concentration of fecal coliform on the grad student's hands

$$M_{initial} = \frac{10^7\, cfu}{100 mL} \cdot \frac{1 mL}{hands} = 10^5\, cfu / hands$$

2. Calculate the final concentration using Eq. 5

$$M_{final} = M_{initial} \cdot \left(1 - \frac{\%R}{100}\right) = 10^5\, cfu / hands \cdot \left(1 - \frac{99}{100}\right) = \boxed{10^3 CFU/hands}$$

3. Calculate desirable reduction using Eq. 6.1

$$\text{Reduction} > \left(1 - \frac{M_{final}}{M_{initial}}\right) \cdot 100(\%) > \left(1 - \frac{1}{10^5}\right) \cdot 100(\%)$$

$$\boxed{> 99.999\% \text{ or } 5 \log_{10} \text{ reduction}}$$

Effectiveness of microbial reduction varies with the species of microorganism and type of water treatment (Table 6.8 and 6.9). Clearly wastewater treatment plants have lower removals of microorganisms compared with drinking water plants. As a result, high concentrations of microbes may be released into rivers, lakes, and oceans where humans could be exposed to pathogens during recreational activities. Although drinking water plants remove microbes more effectively, treatment may not good enough for certain pathogens or for highly contaminated source waters.

Table 6.8 Typical removal (%) of microorganisms by conventional wastewater treatment

	Primary Treatment	**Secondary Treatment**	
Microorganisms		**Activate Sludge**	**Trickling Filter**
Fecal coliform	<10	0-99	85-99
Salmonella	0-15	70-99	85-99+
Shigella	15	80-90	85-99
Mycobacterium tuberculosis	40-60	5-90	65-99
Entamoeba histolytica	0-50	Limited	Limited
Helminth ova	50-98	Limited	60-75
Enteric viruses	Limited	75-99	0-85

Source: Crook 1992

Table 6.9 Removal (log_{10}) of microorganisms at a reclamation facility in Petersburg, FL

Microorganisms	**Activate Sludge & Clarification**	**Filtration**	**Chlorination**	**Storage**	**Overall**
Total coliforms	1.75	0.51	4.23	0.61	7.10
Fecal coliforms	2.06	0.05	4.95	0.36	7.42
Phage	0.75	3.81	1.03	1.03	6.62
Enterovirus	1.71	0.81	1.45	1.04	5.01
Giardia	1.19	2.00	0.65	0.30	4.14
Cryptosporidium	1.14	1.68	0	0.04	2.86

Source: Rose et al. 1996

EXAMPLE 6.7 Water/Wastewater Treatments
Scenario: A reclamation facility uses raw sewage as a source of water.

Task: How many $\log_{10}$ reductions are required to protect public health? Assume that the sewage contains the following concentrations of pathogens: rotavirus (enteric virus) 10^4/100mL, *Shigella* 10^3/100mL, and *Cryptosporidium* 10^5/100mL.

Solution steps:

1. Calculate a daily acceptable infectious risk. Use US EPA's suggested acceptable annual infection risk for drinking water, which is 1/10,000 (10^{-4}).

$$Pi_{daily} = 1-(1-Pi_{annual})^{1/365} = 2.74 \text{ x } 10^{-7} \qquad (6.19)$$

2. Calculate daily intake doses that may cause 2.74 x 10^{-7} infection based on probabilistic dose-response models (exponential and beta-Poisson)

$$D(P_i) = -\frac{\ln(1-P_i)}{r} \qquad (6.15)$$

$$D(P_i) = \beta\left((1-P_i)^{-1/\alpha} - 1\right) \qquad (6.16)$$

$D(P_i)$ = 4.62 x10^{-7} for rotavirus, 5.61 x10^{-5} for *Shigella*, and 6.52 x10^{-5} for *Cryptosporidium.*

3. Calculate microbial concentrations that meet daily acceptable risk. Assume that people drink 2 L of water daily.

$$C(P_i) = \frac{D(P_i)}{V} \qquad (6.18)$$

$C(P_i)$ = 2.31 x10^{-7} pfu/L $\left(\text{or } \frac{1\text{pfu}}{4.33\times10^6 L} \text{ or } 2.31\times10^{-8} \text{ pfu/100mL}\right)$ for rotavirus, 2.81 x10^{-5} CFU/L $\left(\text{or } \frac{1\text{cfu}}{3.56\times10^4 L} \text{ or } 2.81\times10^{-6} \text{ cfu/100mL}\right)$ for *Shigella*, and 3.26 x10^{-5} oocysts/L $\left(\text{or } \frac{1\text{oocyst}}{3.07\times10^4 L} \text{ or } 3.26\times10^{-6} \text{ oocysts/100mL}\right)$ for *Cryptosporidium.*

4. Calculate the desirable reduction using Eq. 4 (note that concentration may be used instead of number of microorganisms).

Log_{10} reduction = $\log_{10}(C_{source}) - \log_{10}(C(P_i))$
= 12 $\log_{10}$ reduction for rotavirus
= 8.6 $\log_{10}$ reduction for *Shigella*

= 11 $\log_{10}$ reduction for *Cryptosporidium*

Computations can be done using Microsoft EXCEL (Table 6.10)

Table 6.10 Summary Computation Table using EXCEL

	A	B	C	D	E
1			Pathogens		
2		Parameters	Rotavirus	*Shigella*	*Cryptosporidium*
3		Dose-Response			
4		A	0.2531	0.210	
5		B	0.4265	43	
6		R			0.0042
7					
8		Infectious risk, Pi	Exposure dose that may cause infection, D(Pi)		
9	annual	0.0001	=$C$5*((1-B9)^(-1/$C$4)-1)	2.04E-02	=-(LN(1-B9)/$E$6)
10	daily	=1-(1-B9)^(1/365)	4.62E-07	5.59E-05	6.52E-05
11					
12		Water intake (L)	Microbial concentrations that may cause infection, C(Pi)		
13		2	2.31E-07	=D10/B13	3.26E-05
14					
15			Raw sewage concentration, C(source)		
16			1.00E+05	1.00E+04	1.00E+06
17					
18			Log Reduction		
19			=-LOG(C13/C16)	8.6	10

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