

Pathogen reduction requirements for direct potable reuse - Supplementary Materials

1 Methods

2
3 Additional information about methods used to develop and implement the model is provided below.

6 Dose-Response Models

7
8 The norovirus dose-response model published by Teunis et al. (2008) estimates probabilities of
9 infection and illness as functions of dose. Fit parameters for the combined inocula dataset
10 (8f11a+8f11b) were used, making no assumptions about the aggregation state of the virus particles.
11 One of the fit parameters provided by Teunis et al. (2008) exceeds the limits of this model;
12 therefore, the Pfaff transformation was used as a very close approximation (assuming all doses
13 $\leq 33,323$). The probability of norovirus infection per dose (p_{inf_NV} ; $\text{person}^{-1} \text{day}^{-1}$) was estimated as
14

$$p_{inf_NV} = 1 - \left[{}_2F_1(\beta_{NV}, \frac{\lambda_{NV}(1 - a_{NV})}{a_{NV}}, \alpha_{NV} + \beta_{NV}; a_{NV}) \left(\frac{1}{1 - a_{NV}} \right)^{-\left(\frac{\lambda_{NV}(1 - a_{NV})}{a_{NV}} \right)} \right], \quad [1]$$

15
16 where ${}_2F_1$ is a hypergeometric function, λ_{NV} is the dose of norovirus (number of organisms), α_{NV} and
17 β_{NV} are fit parameters and a_{NV} represents the fit parameter of the (logarithmic series) aggregate size
18 distribution. The conditional probability of illness in infected subjects ($p_{ill_inf_NV}$) was modeled
19 following Teunis et al. (2008) as
20

$$p_{ill_inf_NV} = 1 - (1 + \eta_{NV}\lambda_{NV})^{-r_NV} \quad [2]$$

21
22 where η_{NV} and r_nv are model parameters described in Teunis et al. (1999). The probability of illness
23 per dose (p_{ill_NV}) was defined as
24

$$p_{ill_NV} = p_{inf_NV} p_{ill_inf_NV} \quad [3]$$

25
26 and using Eqs. [3, 4 and 5] the tolerable dose of norovirus, λ_{NV} , was determined.

27
28 For giardia, Teunis et al. (1996) fitted the exponential dose-response model, using
29

$$p_{inf_G} = 1 - \exp(-r_G\lambda_G), \quad [4]$$

30
31 to the original data published by Rendtorff (1954) where r_G is an infectivity parameter (interpreted
32 as the probability for one organism to initiate infection) and λ_G is the dose of giardia (number of
33 organisms) consumed. The mechanism of giardia pathogenicity and host responses to infection
34 remain unclear (Roxström-Lindquist et al., 2006), although it has been widely reported that a high
35 proportion of giardia infections are asymptomatic; even the original study found no evidence of
36 illness that could be connected to ingestion of giardia cysts (Rendtorff, 1954). The reported
37 proportions of asymptomatic cases are highly variable: two community-wide studies reported 0.19
38 (Birkhead and Vogt, 1989) and 0.76 (Lopez et al., 1980) and studies of adults found 0.07 (Hoque et
39 al., 2002) and 0.30 (Yakoob et al., 2010). Therefore, a Uniform distribution was used to represent the
40 proportion of infections that result in illness. The tolerable daily probability of infection (p_{inf_G}) was
41 estimated as
42

$$p_{inf_G} = \frac{p_{ill_G}}{(inf:ill)}, \quad [5]$$

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44 where *inf:ill* is the proportion of infections that are symptomatic (illness). Eq. [4] was then used to
45 solve for the tolerable dose of giardia, λ_G .

46

47 The probability of *Campylobacter* infection per dose (p_{inf_C} ; person⁻¹ day⁻¹) was estimated as

48

$$p_{inf_C} = 1 - {}_1F_1(\alpha, \alpha + \beta; -\lambda_C), \quad [6]$$

49

50 where ${}_1F_1$ is a hypergeometric function, λ_C is the dose of *Campylobacter* (number of organisms) and
51 α and β are fit parameters. The conditional probability of illness in infected subjects ($p_{ill_inf_C}$) was
52 found to be dose-dependent and was modeled following Teunis et al. (2005) as

53

$$p_{ill_inf_C} = 1 - (1 + \eta_C \lambda_C)^{-r_C} \quad [7]$$

54

55 where η_C and r_C are model parameters described in Teunis et al. (1999). We have assumed the
56 values of η_C and r_C were incorrectly reported in Teunis et al. (2005) such that the published value of
57 η_C is actually r_C . The probability of illness per dose (p_{ill_C}) was defined as

58

$$p_{ill_C} = p_{inf_C} p_{ill_inf_C} \quad [8]$$

59

60 and using Eqs. [8, 9 and 10] the tolerable dose of *Campylobacter*, λ_C , was determined.

61

62

63 **Estimates of municipal sewage**

64

65 Measurements of norovirus in municipal wastewater are scarce which can be explained, at least in
66 part, by the methodological challenges related to the detection of norovirus (Haramoto et al., 2006;
67 Katayama et al., 2008; La Rosa et al., 2010; Ottoson et al., 2006a; Ottoson et al., 2006b). Only two
68 studies reported recovery efficiencies for norovirus detection (Haramoto et al., 2006; Katayama et
69 al., 2008); therefore, a Mixture distribution, incorporating both studies with equal weighting, was
70 used assuming that norovirus concentrations are similar across populations with high living
71 standards. *Giardia lamblia* cyst numbers were surveyed in raw sewage from three sewage treatment
72 plants over a 6 to 12 month period (Van Den Akker et al., 2011). Concentration values, corrected for
73 recovery efficiency, were similar across all three sewage treatment plants with a mean of 2.5 log₁₀
74 cysts L⁻¹. The log₁₀ mean and standard deviation were used to define a Normal distribution (by
75 definition the antilog is Lognormal¹) for each sewage treatment plant and the concentration of
76 giardia in raw sewage was represented by a Mixture distribution of random values drawn from the
77 three Normal distributions with equal weighting. There was limited information on *Campylobacter*
78 concentrations although there is a reference in the Australian Guidelines for Water Recycling
79 (NRMCC et al., 2006a) to unpublished research (10² to 10⁵ cfu L⁻¹ in raw sewage, 95th percentile
80 7x10³). To represent station conditions, the Guideline values (95th and estimates of 1st and 2nd
81 percentiles) were used to estimate a Lognormal distribution.

82

83

¹ Technically, the definition uses the natural logarithm, but data were provided in base 10; it was assumed the definition still applies.

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84 Table S.1 Published values of daily per capita drinking water consumption (L person⁻¹ day⁻¹).

Country	Mean	Standard Deviation	Description	Reference
USA	1.098	0.922 (estimated)	adults 20 years and older	(USEPA, 2004)
USA	1.48	0.984 (estimated)	adults 25-54 years old	(USEPA, 2006)
Canada	1.2	0.8	data from 7 cross-sectional studies	(Roche et al., 2012)
USA	1.3	1.17	Lognormal distribution	(Schijven et al., 2011)
Sweden	0.873	0.541	Lognormal distribution	(Åstrom et al., 2007)
France	1.760	0.001715		(Hunter et al., 2011)

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87 Table S.2 Comparison of methods used to estimate required log₁₀ reductions for potable reuse of municipal sewage.

Model Parameters	Australian Guidelines: Augmentation of Drinking Water Supplies ^a	WHO: Guidelines for Drinking Water Quality ^b	Model (estimated municipal sewage)
Model type	Deterministic	Deterministic	Stochastic
Reference pathogens	RV, Cr, Cb	RV, Cr, Cb	NV, G, Cb
Pathogen concentration (# L ⁻¹)	95 th percentile values in raw sewage RV: 8.00x10 ³ = adenovirus concentration from Virginia Pipeline Scheme, SA (unpublished) Cr: 2.00x10 ³ ; Cb: 7.00x10 ³	River Water RV: 10 Cr: 10 Cb: 100	Mixture distribution for raw sewage NV: 3.12x10 ⁶ (mean), 1.02x10 ⁷ (95 th percentile) ^c G: 2.51x10 ³ (mean), 9.04x10 ³ (95 th percentile) Cb: 1.90x10 ³ (mean), 7.19x10 ³ (95 th percentile)
Dose-response model	RV: simplified approx. Beta-Poisson Cr: Exponential (r=0.059) Cb: simplified approx. Beta-Poisson	RV: Beta-Poisson (α=0.2531, β=0.4265) Cr: Exponential (r=0.00467) Cb: Exponential (r=0.019)	NV: full Beta-Poisson (hypergeometric) G: Exponential (r=Triangular) Cb: full Beta-Poisson (hypergeometric)
Disease burden (DALYs case ⁻¹)	RV: 1.3x10 ⁻² ; Cr: 1.5x10 ⁻³ ; Cb: 4.6x10 ⁻³	RV: 1.4x10 ⁻² Cr: 1.5x10 ⁻³ Cb: 4.6x10 ⁻³	NV: Uniform(3.71x10 ⁻⁴ , 6.23x10 ⁻³) ~ 3.30x10 ⁻³ (mean) G: Uniform(2.10x10 ⁻³ , 2.68x10 ⁻³) ~ 2.39x10 ⁻³ (mean) Cb: Uniform(4.60x10 ⁻³ , 4.10x10 ⁻²) ~ 2.28x10 ⁻² (mean)
Susceptibility fraction	RV: 0.06 (population <5 years) Cr and Cb: 1	RV: 0.06 Cr and Cb: 1	NV: Uniform(0.8, 1.0) G and Cb: 1
Ratio of infection to illness	RV: 0.88 Cr: 0.70 Cb: 0.30	RV: 0.5 Cr: 0.7 Cb: 0.3	NV: non-linear dose-response model G: Uniform(0.24, 0.93) ~ 0.58 (mean) Cb: non-linear dose-response model
Daily per capita drinking water (L)	2	1	Lognormal(3, 1) – truncated at 2 and 6
Required log ₁₀ reduction	RV: 9.5 Cr: 8 Cb: 8.1	RV: 5.96 Cr: 5.89 Cb: 5.98	NV: 6.9 (95 th percentile) G: 8.0 (95 th percentile) Cb: 7.4 (95 th percentile)

88 ^aGuidelines refer to Phase I Guidelines for many of the parameter values (NRMCC et al., 2006).

89 ^b(WHO, 2011)

90 ^cMunicipal treatment plants were different sizes. In Haramoto et al. (2006) the WWTP serves a population of ~63,000 and treats 28,000m³ of sewage per day. In Katayama et al. (2008), samples were collected from 6 WWTPs that , ranging in size from 63,000 to 770,000 people served and average daily treated volume of 28,000 to 571,000 m³ per day.

93 Note: Cb=*Campylobacter*, Cr=cryptosporidium, G=Giardia, NV=norovirus, RV=rotavirus

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94 Table S.3. Stepwise results from reverse QMRA for required log₁₀ reduction (LRV) of pathogens in sewage for potable reuse of treated wastewater. Values
 95 reported as 50th[5th, 95th] percentiles.

Model Parameter	Norovirus	Giardia	<i>Campylobacter</i>
Health target (DALYs person ⁻¹ year ⁻¹)	10 ⁻⁶	10 ⁻⁶	10 ⁻⁶
Sewage conc'n – municipal (# L ⁻¹)	1.7x10 ⁶ [3.2x10 ⁵ , 1.0x10 ⁷]	6.5x10 ² [4.2x10 ¹ , 9.0x10 ³]	6.7x10 ² [6.3x10 ¹ , 7.2x10 ³]
Sewage conc'n – Davis outbreak (# L ⁻¹)	5.0x10 ¹¹ [6.5x10 ¹⁰ , 1.4x10 ¹²]	9.7x10 ⁵ [7.5x10 ⁵ , 1.4x10 ⁶]	1.2x10 ⁸ [9.6x10 ⁶ , 4.9x10 ⁸]
Sewage conc'n – Melbourne outbreak (# L ⁻¹)	2.6x10 ¹⁰ [3.4x10 ⁹ , 7.2x10 ¹⁰]	n/a	n/a
Tolerable annual probability of illness	3.4x10 ⁻⁴ [1.9x10 ⁻⁴ , 1.7x10 ⁻³]	4.2x10 ⁻⁴ [3.8x10 ⁻⁴ , 4.7x10 ⁻⁴]	4.4x10 ⁻⁵ [2.6x10 ⁻⁵ , 1.6x10 ⁻⁴]
Tolerable daily probability of illness	3.8x10 ⁻⁶ [1.8x10 ⁻⁶ , 1.9x10 ⁻⁵]	4.6x10 ⁻⁶ [3.4x10 ⁻⁶ , 6.6x10 ⁻⁶]	5.0x10 ⁻⁷ [2.5x10 ⁻⁷ , 1.8x10 ⁻⁶]
Tolerable daily probability of infection	4.6x10 ⁻³ [3.2x10 ⁻³ , 1.0x10 ⁻²]	8.2x10 ⁻⁶ [4.5x10 ⁻⁶ , 1.8x10 ⁻⁵]	6.2x10 ⁻⁴ [4.4x10 ⁻⁴ , 1.2x10 ⁻³]
Tolerable daily dose (#)	3.8[2.6, 8.5]	3.3x10 ⁻⁴ [1.3x10 ⁻⁴ , 1.0x10 ⁻³]	9.2x10 ⁻⁴ [6.7x10 ⁻⁴ , 1.7x10 ⁻³]
Tolerable drinking water conc'n (# L ⁻¹)	1.3[0.7, 3.0]	1.1x10 ⁻⁴ [4.0x10 ⁻⁵ , 3.6x10 ⁻⁴]	3.1x10 ⁻⁴ [1.7x10 ⁻⁴ , 6.3x10 ⁻⁴]
Required LRV – municipal	6.1[5.3, 6.9]	6.8[5.5, 8.0]	6.3[5.3, 7.4]
Required LRV – outbreak	11.6[10.6, 12.1]	10.0[9.4, 10.4]	11.6[10.5, 12.3]
Required LRV – Melbourne outbreak	10.3[9.4, 10.8]	n/a	n/a
Required LRV – Guideline values	9.5	8.0	8.1

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97 Table S.4 Estimated required protozoan log₁₀ reduction values (LRVs) for stepwise methodological
 98 changes from the Guideline method (NRMCC et al., 2008) to a deterministic approximation of the
 99 model using municipal sewage concentrations.

Step	RV	Model Input Parameters ^a						
		<i>V</i>	<i>c</i>	<i>B</i>	<i>S_f</i>	<i>inf:ill</i>	d-r	<i>n</i>
1.	.0	2	2000	1.5x10 ⁻³	1	0.70	Cr ^b	365
2.	.6	2	9.04x10 ³ (95 th G)	1.5x10 ⁻³	1	0.70	Cr ^b	365
3.	.3	4.8 (95 th)	2000	1.5x10 ⁻³	1	0.70	Cr ^b	365
4.	.9	2	9.04x10 ³ (95 th G)	2.7x10 ⁻³ (95 th)	1	G ^c	G ^c	365
5.	.2	4.8 (95 th)	9.04x10 ³ (95 th G)	2.7x10 ⁻³ (95 th)	1	G ^c	G ^c	365
6.	.4	2	9.04x10 ³ (95 th G)	2.7x10 ⁻³ (95 th)	1	G ^c	G ^c	118 (95 th AAD)
7.	.8	4.8 (95 th)	9.04x10 ³ (95 th G)	2.7x10 ⁻³ (95 th)	1	G ^c	G ^c	118 (95 th AAD)

100 ^aModel input parameters: *V* = daily water consumption (L person⁻¹), *c* = sewage pathogen
 101 concentration (# L⁻¹), *B* = disease burden (DALYs case⁻¹), *S_f* = susceptibility fraction, *inf:ill* = ratio of
 102 infection to illness, d-r = dose-response model, *n* = days of exposure.

103 ^bexponential dose-response model; r=5.9x10⁻²

104 ^cgiardia exponential dose-response model: use 95th values of r (0.0468) and Inf:ill (0.8954).

105

106

107 The Guidelines (NRMCC et al., 2008) recommend a minimum cryptosporidium log₁₀ reduction (LRV)
 108 of 8.0 for the production of drinking water from sewage while the full stochastic model, using
 109 municipal sewage concentration, obtained the same value for giardia. To compare these two
 110 methods, sequential steps in methodology from the Guideline method (Step 1) to a deterministic
 111 approximation of the model method (Step 7, using 95th percentile values of all input distributions)
 112 are reported. The difference in LRVs between Steps 1 and 2 shows the effect of using Australian
 113 giardia concentrations (8.0 to 8.6). The difference between Steps 2 and 4 shows the slight increase in
 114 LRV due to the giardia dose-response model (8.6 to 8.9). The difference between Steps 4 and 5
 115 shows the impact of using the higher drinking water volume (8.9 to 9.2) and the difference between
 116 Steps 5 and 7 shows the impact of a shorter exposure period (9.2 to 8.8). Comparing the 95th
 117 percentile of the full stochastic model (8.0) with a deterministic approximation of the method (Step
 118 7; 8.8), the difference is moderate.

119

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120 Table S.5 Estimated required bacterial log₁₀ reduction values (LRVs) for stepwise methodological
 121 changes from the Guideline method (NRMMC et al., 2008) to a deterministic approximation of the
 122 model using municipal sewage concentrations.

Step	RV	Model Input Parameters ^a						
		<i>V</i>	<i>c</i>	<i>B</i>	<i>S_f</i>	Inf:ill	d-r	<i>n</i>
1.	.1	2	7000	4.6x10 ⁻³	1	0.30	Cb ^b	365
2.	.5	4.8 (95 th)	7000	4.6x10 ⁻³	1	0.30	Cb ^b	365
3.	.6	2	7000	3.9x10 ⁻² (95 th)	1	Cb ^c	Cb ^c	365
4.	.0	4.8 (95 th)	7000	3.9x10 ⁻² (95 th)	1	Cb ^c	Cb ^c	365
5.	.4	2	7000	3.9x10 ⁻² (95 th)	1	Cb ^c	Cb ^c	118 (95 th)
6.	.7	4.8 (95 th)	7000	3.9x10 ⁻² (95 th)	1	Cb ^c	Cb ^c	118 (95 th)

123 ^aModel input parameters: *V* = daily water consumption (L person⁻¹), *c* = sewage pathogen
 124 concentration (# L⁻¹), *B* = disease burden (DALYs case⁻¹), *S_f* = susceptibility fraction, Inf:Ill = ratio of
 125 infection to illness, d-r = dose-response model, *n* = days of exposure.

126 ^bsimplified approximate Beta-Poisson; alpha=0.145, beta=7.58

127 ^cfull Beta-Poisson

128

129 The Guidelines (NRMMC et al., 2008) recommend a minimum *Campylobacter* log₁₀ reduction (LRV)
 130 of 8.1 for the production of drinking water from sewage while the full stochastic model, using
 131 municipal sewage concentrations, determined a 95th percentile LRV of 7.4. To compare these two
 132 methods, sequential steps in methodology from the Guideline method (Step 1) to a deterministic
 133 approximation of the model (Step 6, using 95th percentile values of all input distributions) are
 134 reported. The difference between Steps 1 and 2 shows the impact of using the higher drinking water
 135 volume (8.1 to 8.5). The difference between Steps 1 and 3 shows the reduction in LRV due to the full
 136 *Campylobacter* dose-response model (8.1 to 7.6) and a further reduction is shown with the
 137 implementation of the (shorter) summer exposure period (Steps 4 and 6; LRVs of 8.0 and 7.7).
 138 Comparing the 95th percentile of the full stochastic model (7.4) with a deterministic approximation
 139 of the method (Step 6; 7.7), the difference is small.

140

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141 Table S.6 Published maximum pathogen concentrations in raw sewage.

142

Country	Peak value	Units	Account for recovery ^a	%	Reference
NOROVIRUS					
Brazil	~5x10 ⁴	Genomic copies L ⁻¹	γ/a		(Victoria et al., 2010)
Finland	10 ⁶	PCR units L ⁻¹	γ/a		(Von Bonsdorff et al., 2002)
France	1x10 ⁹ (NV GI)	Genomic copies L ⁻¹	γ/a		(Da Silva et al., 2007)
Germany	9.7*10 ⁵	Genomic equivalents L ⁻¹	γ/a		(Pusch et al., 2005)
Italy	5.7x10 ⁸	GC/L (have assumed error in paper)	γ/a		(La Rosa et al., 2010)
Japan	1.9x10 ⁷ total NV (I+II)	copies L ⁻¹	yes		(Haramoto et al., 2006)
Japan	6.6x10 ⁶ total NV (I+II)	monthly mean RT-PCR units L ⁻¹	yes		(Katayama et al., 2008)
Netherlands	8.5x10 ⁵	PDU L ⁻¹	γ/a		(Lodder and De Roda Husman, 2005)
Netherlands	10 ⁶	PCR detectable units L ⁻¹	γ/a		(Van Den Berg et al., 2005)
Singapore	1x10 ⁷ (NV GI)	Genomic copies mL ⁻¹	γ/a		(Aw and Gin, 2010)
Sweden	3.65	log ₁₀ MPN PCR units L ⁻¹	γ/a		(Ottoson et al., 2006b)
Sweden	4.5 x10 ³	# L ⁻¹	γ/a		(Ottoson et al., 2006a)
Sweden	1x10 ⁷ (NV GII)	Genomic copies L ⁻¹	γ/a		(Nordgren et al., 2009)
UK	1.8x10 ⁷	cDNA copies L ⁻¹	γ/a		(Laverick et al., 2004)

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145 Table S.6 Published maximum pathogen concentrations in raw sewage - continued.

146

Country	Peak value	Units	Account for recovery ^a	% Reference
GIARDIA				
Australia	$>5.0 \times 10^2$	cysts L ⁻¹	r/a	(Wohlsen and Katouli, 2006)
Canada	2.1×10^2	cysts L ⁻¹	r/a	(Chauret et al., 1999)
Japan	3.9×10^3	cysts L ⁻¹	yes	(Oda et al., 2005)
Netherlands	2.6×10^3	cysts L ⁻¹	yes	(Medema and Schijven, 2001)
Spain	1.4×10^4	cysts L ⁻¹	yes	(Castro-Hermida et al., 2010)
Spain	8.31×10^3	cysts L ⁻¹	r/a	(Castro-Hermida et al., 2008)
Sweden	5.72×10^4	cysts L ⁻¹	yes	(Ottoson et al., 2006b)
Sweden	1.77×10^4	cysts L ⁻¹	yes	(Ottoson et al., 2006a)
USA	1.4×10^4	cysts L ⁻¹	r/a	(Gassmann and Schwartzbrod, 1991)
USA	1.3×10^2	cysts L ⁻¹	r/a	(Rose et al., 1996)
USA	1.4×10^4	cysts L ⁻¹	r/a	(Sykora et al., 1991)
Australia	~900	cysts L ⁻¹	yes	(Van Den Akker et al., 2011)

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149 Table S.6 Published maximum pathogen concentrations in raw sewage - continued.

150

Country	Peak value	Units	Account for recovery ^a	% Reference
CAMPYLOBACTER				
Germany	>1x10 ⁷	CFU L ⁻¹	10	(Rechenburg and Kistemann, 2009)
Italy	10 ⁵	CFU L ⁻¹	10	(Stellacci et al., 2010)
Baltic Sea	1.1x10 ⁶	CFU L ⁻¹	10	(Holler, 1988)
Netherlands (combined sewers)	2.4x10 ⁴	CFU L ⁻¹	10	(ten Veldhuis et al., 2010)
Germany	10 ⁴	CFU L ⁻¹	10	(Stelzer, 1991)
USA	6.2x10 ⁷	CFU L ⁻¹	1/a	(Hellein et al., 2011)
Spain	1.5x10 ⁵	MPN L ⁻¹	10	(Rodríguez and Araujo, 2010)
Switzerland	2.3x10 ⁶	cells L ⁻¹	10	(Rinsoz et al., 2009)
France	3x10 ⁶	genes L ⁻¹	10	(Wéry et al., 2008)
UK	4.6x10 ⁵	MPN L ⁻¹		(Arimi et al., 1988)

151 ^a n/a = not stated, unclear

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