New Zealand Journal of Marine and Freshwater Research

ISSN: 0028-8330 (Print) 1175-8805 (Online) Journal homepage: http://www.tandfonline.com/loi/tnzm20

Faecal indicators and pathogens in selected New Zealand waterfowl

EM Moriarty , N Karki , M Mackenzie , LW Sinton , DR Wood & BJ Gilpin

To cite this article: EM Moriarty , N Karki , M Mackenzie , LW Sinton , DR Wood & BJ Gilpin (2011) Faecal indicators and pathogens in selected New Zealand waterfowl, New Zealand Journal of Marine and Freshwater Research, 45:4, 679-688, DOI: 10.1080/00288330.2011.578653

To link to this article: http://dx.doi.org/10.1080/00288330.2011.578653

New Zealand Journal of Marine and Freshwater Research
Vol. 45, No. 4, December 2011, 679–688

Taylor & Francis
Taylor & Francis Group

Faecal indicators and pathogens in selected New Zealand waterfowl

EM Moriarty*, N Karki, M Mackenzie, LW Sinton, DR Wood and BJ Gilpin

Christchurch Science Centre, Institute of Environmental Science and Research (ESR) Ltd, Christchurch New Zealand

(*Received 17 November 2010; final version received 1 April 2011*)

Freshly excreted droppings from Canada geese ($n = 80$), black swans ($n = 80$), ducks ($n = 80$) and gulls ($n = 80$) were collected from sites around New Zealand. The droppings were enumerated for *Escherichia coli*, enterococci and *Salmonella* spp., and for the presence/absence of *Cryptosporidium* spp. Overall prevalence of *E. coli* and enterococci in samples was 95% and 94%, respectively. *Cryptosporidium* spp. was detected in 2% of the samples, whereas no *Salmonella* spp. were detected in the survey. Preliminary estimates of daily microbial outputs suggest that ducks will produce the highest loadings of *E. coli* and enterococci per bird, whereas Canada geese will produce the highest loadings of *Campylobacter* spp. per bird. This study provides the first set of indicator and pathogen counts for one of the largest sources of diffuse faecal contamination of natural waters in New Zealand.

Keywords: waterfowl; *E. coli*; enterococci; *Campylobacter*; *Salmonella*; *Cryptosporidium*; water quality

Introduction

Concern has arisen over the contribution of waterfowl to the microbial loadings of surface waters, and their consequent impacts on bathing water quality (Wither et al. 2005) and industries such as shellfish harvesting (Albarnaz et al. 2007). Waterfowl, such as mallard ducks (*Anas platyrhynchos*), Canada geese (*Branta canadensis*), black swans (*Cygnus atratus*), and species of gull are abundant in New Zealand. Mallard duck numbers are estimated at 4.5 million (Fish and Game NZ 2009), and Canada geese and black swan numbers are each estimated at less than 100,000 (Heather and Robertson 2005), but there are no published national totals of gull numbers. These birds live on and near coastlines, estuaries, rivers, streams, wetlands and lakes, and they are also found on and around waste stabilisation ponds.

Waterfowl harbour a range of potentially pathogenic microorganisms (Waldenstrom et al. 2002; Nielsen et al. 2004), and as such, are important reservoirs of nonpoint sources of faecal contamination. Overseas studies have identified a range of potentially zoonotic pathogens in gull faeces, including *Salmonella* spp., *Campylobacter coli* and *Campylobacter jejuni* (Quessy and Messier 1992; Moore et al. 2002). *Cryptosporidium* spp. has been frequently isolated from the faeces of Canada geese (Jellison et al. 2004; Kassa et al. 2004; Zhou et al. 2004), along with bacterial pathogens, including *Campylobacter* spp. and *Salmonella* spp. (Pacha et al. 1988; Wahlstrom et al. 2003). There appears to be little published information on the microbial loading from black swans, although one study noted the presence of *Cryptosporidium* spp. in the faeces of the birds at a zoo (Rohela et al. 2005).

*Corresponding author. Email: elaine.moriarty@esr.cri.nz

ISSN 0028-8330 print/ISSN 1175-8805 online

http://dx.doi.org/10.1080/00288330.2011.578653
http://www.tandfonline.com

Apart from one study of ducks, which identified the presence of potentially pathogenic bacteria, including *C. jejuni*, in their faeces (Murphy et al. 2005), there is little published information on the microbial loading of waterfowl in New Zealand. However, their likely contribution to microbial contamination of recreational and shellfishing waters is recognised in the New Zealand Ministry for the Environment and Ministry for Health guidelines (Ministry for the Environment 2003).

This paper describes a survey of the faeces of several waterfowl species in New Zealand—Canada geese, black swans, gulls and ducks—to evaluate their potential contribution to microbial pollution of surface waters. Faecal concentrations of the bacterial indicators *Escherichia coli* and enterococci, and the pathogens *Campylobacter* spp. and *Salmonella* spp. were determined, as well as the presence of *Cryptosporidium* spp. These data will inform scientists modelling wildfowl versus other sources of faecal and pathogen contamination.

Material and methods

Sampling sites and sample collection

Freshly deposited faecal droppings ($n = 80$) from each of the four waterfowl groups—Canada goose, duck, gull and black swan—were collected in sterile faecal specimen containers. Samples were returned to the laboratory in a chilled, darkened, insulated container for analysis within 24 h. Samples were collected from four locations in NZ: Auckland, Hamilton, Farewell Spit and Christchurch.

Microbial analysis

Laboratory methods

Depending on the size of the faecal matter available for collection, a portion of the sample, ranging from 1 to 25 g, was added to a sterile filter stomacher bag containing buffered peptone water (Oxoid, UK), to create a 10-fold dilution. The mixture was homogenised using a Bag Mixer (Interscience, Ontario, Canada). An aliquot (20 ml) was removed and 1 ml of this sub-sample was used to create a series of 10-fold dilutions in 9 ml of peptone water (0.1%) (Fort Richards, NZ). This procedure was repeated for all samples.

Enterococci/E. coli

Enterococci and *E. coli* were enumerated using the Enterolert® and Colilert® systems, respectively (Idexx Laboratories Inc., Westbrook, ME, US), according to the manufacturer's instructions, with the modification of an elevated incubation temperature (44.5 °C) for faecal coliform detection (Yakub et al. 2002). The limit of detection for the Enterolert® and Colilert® systems was determined to be 10 colony-forming units/g of faecal sample.

Campylobacter *spp.*

Campylobacter spp. were enumerated by a 3×5 MPN procedure in 30-ml volumes of Exeter Broth and incubated at 42 °C for 48 h under microaerophilic conditions (10% CO_2). MPN tubes were plated onto m-Exeter agar (Fort Richards, Auckland, New Zealand) and incubated at 37 °C for a minimum of 4 h, followed by 42 °C for 44 h under microaerophilic conditions (10% CO_2). The dilutions used for the MPN series differed according to the expected concentration of *Campylobacter* spp. in the samples. Despite this, a number of samples ($n = 35$) were above the limit of detection for *Campylobacter* spp. A multiplex PCR was used to identify the isolates as *C. jejuni*, *C. coli* or thermotolerant *Campylobacter*, as previously described (Wong et al. 2004; Moriarty et al. 2008). Isolates that were classified as thermotolerant by multiplex PCR were further analysed by real-time PCR for the detection of *C. lari* (Chaban et al. 2009).

Salmonella *spp.*

Salmonella spp. were enumerated using a 3×3 MPN procedure giving limits of detection from 4 to 1.1×10^3 MPN *Salmonella* spp. per gram of faeces. One-millilitre samples were dispensed into 9-ml vials of Selenite Cystine broth (Fort Richards, Auckland, New Zealand), incubated at 35 °C for 24 h, plated onto Hektoen and XLD agar (Fort Richards, Auckland, New Zealand), and incubated at 35 °C for 24 h. All suspect colonies were re-streaked to purify them, and biochemical and latex agglutination tests were carried out to confirm the presence of *Salmonella*.

Cryptosporidium *spp.*

Faecal samples were analysed for *Cryptosporidium* spp. according to the method of Ng et al. (2006), which employs both a freeze-thaw step to release the DNA from robust oocysts and QIAmp DNA Stool Kit to clean up the DNA (QIAGEN, Hilden, Germany). The samples were tested only for the presence/absence of *Cryptosporidium* spp.

Estimation of microbial loadings

Preliminary estimates were made of the likely daily microbial outputs of the waterfowl using published information on daily faecal outputs (Geldreich 1966; Wood & Trust 1972; Hussong et al. 1979; Mitchell & Wass 1995), and the microbial concentrations recorded in this study. These estimates are given in Table 1.

Statistical analysis

Values below the limit of detection were assumed to be zero, whereas those above it were assigned a value equal to the limit of detection of that particular dilution series. All counts were expressed as arithmetic means, based on 80 samples from each bird type. Arithmetic means were calculated in order to provide estimates of the daily microbial output per bird (McBride 2005). XLSTAT version 2008.4.02 was used to calculate inferential statistics such as Spearman rank correlations.

Results

The arithmetic mean counts per gram (wet weight) and prevalence of *E. coli*, enterococci, *Salmonella* spp., *Campylobacter* spp. and *Cryptosporidium* spp. determined for each type of bird are presented in Fig. 1 and Table 1. The principal findings were the presence of *E. coli* in over 94% of samples and enterococci in over 79% of the samples. *Salmonella* spp. was not detected in any of the samples, and *Campylobacter* spp. ranged in prevalence from 29% in ducks to 59% in gulls. *Cryptosporidium* spp. was present in the faeces of four Canada geese, two black swans and one duck.

Of the 176 faecal samples that were positive for *Campylobacter* spp., all were identified as *C. jejuni,* with the exception of nine isolates from gulls, which were identified as *C. lari.*

A wide range of counts of *E. coli*, enterococci and *Campylobacter* spp. was observed in individual bird faeces ranging from <10 g^{-1} to $>10^5$ g^{-1} for *Campylobacter* and $>10^9$ g^{-1} for *E. coli* and enterococci (Fig. 1). The ranking for the highest average concentration of indicator organisms (*E. coli* and enterococci) in the faeces of the wildfowl was ducks > gulls > black swans > Canada geese. This order changed for *Campylobacter* spp. with the highest average concentration of the pathogen recorded in Canada geese, followed by gull, black swan and duck.

Spearman rank correlation of individual droppings (Table 4) suggest that there is strong evidence that *E. coli* is correlated with both enterococci ($P < 0.0001$) and *Campylobacter* ($P = 0.038$), though overall there is no significant association between enterococci and *Campylobacter*. The association is a moderately strong positive ($r_s = 0.489$) between *E. coli* and enterococci and a weekly positive ($r_s = 0.116$) between *E. coli* and *Campylobacter*.

Table 1 Counts and prevalence of key enteric microorganisms in freshly-deposited faecal samples from selected New Zealand waterfowl, and (where applicable) estimated daily microbial output per bird, based on previously reported daily faecal outputs.

Waterfowl	Microorganism[a]	Mean count (g^{-1} wet weight)	Prevalence (%)	Mean daily faecal output (g)	Estimated daily microbial output per bird
Black swan	*E. coli*	1.91×10^6	94	418 (Mitchell & Wass 1995)	7.98×10^8
	Enterococci	1.10×10^6	79		4.59×10^8
	Campylobacter spp.	2.04×10^2	45		8.53×10^4
	Salmonella spp.	0	0		0
	Cryptosporidium spp.	(+)	2.5		–
Canada goose	*E. coli*	3.61×10^4	95	250 (Hussong et al. 1979)	9.03×10^6
	Enterococci	2.50×10^4	98		6.25×10^6
	Campylobacter spp.	4.84×10^3	40		1.21×10^6
	Salmonella spp.	0	0		0
	Cryptosporidium spp.	(+)	5.0		–
Duck	*E. coli*	9.46×10^7	95	336 (Geldreich 1966)	3.18×10^{10}
	Enterococci	1.01×10^8	100		3.39×10^{10}
	Campylobacter spp.	5.92×10^1	29		1.99×10^4
	Salmonella spp.	0	0		0
	Cryptosporidium spp.	(+)	1.3		–
Gull	*E. coli*	1.87×10^7	96	50 (Wood & Trust 1972)	9.35×10^8
	Enterococci	8.90×10^6	99		4.45×10^8
	Campylobacter spp.	7.66×10^2	59		3.83×10^4
	Salmonella spp.	0	0		0
	Cryptosporidium spp.	(−)	0		–

[a]All counts are arithmetic means, based on 80 samples from each bird type. For *Cryptosporidium* spp., the samples were tested for presence/absence only. For calculation of the arithmetic means, all values above (>) and below (<) the limits of detection were, respectively, assigned a value equal to the limit of detection, and assumed to be zero.

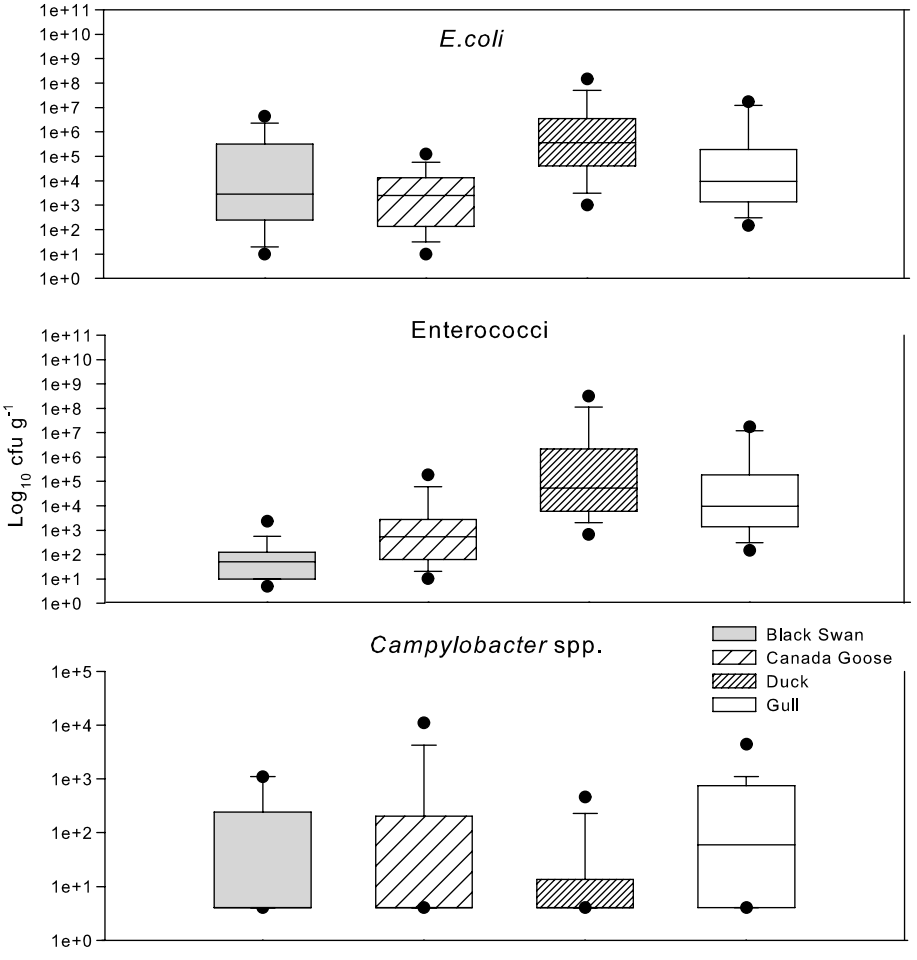

Figure 1 Variability associated with faecal samples from each of the wildfowl sampled. The horizontal line represents the median, boxes are the 25th to 75th percentile, whiskers 10th and 90th percentile and points are the 5th and 95th percentile values. The maximum value is unknown for *Campylobacter* spp. as 35 samples exceeded the detection threshold of the MPN procedure.

Discussion

This paper provides the first comprehensive set of data on the incidence of microbial indicators and selected pathogens in the faeces of common waterfowl in New Zealand. The study highlights the role birds play in polluting waters and recreational areas. The high variability in the concentrations of indicator and pathogens in their faeces presents difficulties in assessing the extent of their role in dissemination of waterborne disease.

The prevalence of *E. coli* was high ($\geq$94%) in all the waterfowl droppings analysed in this study. The highest mean concentration of *E. coli* was recorded in duck faeces, followed by lower levels in gull faeces (almost 10-fold lower), black swan faeces (almost 100-fold lower) and Canada geese faeces (almost 10,000-fold lower). The mean *E. coli* values are similar to those previously published (Wood & Trust 1972; Alderisio & DeLuca 1999; Fogarty et al. 2003; Lévesque et al.

Table 2 Prevalence and concentration of indicator bacteria in avian faeces from literature.

Bird	Microorganism	Conc./Prevalence	Reference
Canada goose	*E.coli*	63%	Fallacara et al. 2001
Canada goose	*E.coli*	2–94%	Kullas et al. 2002
Canada goose	*E.coli*	3.6×10^5/74.6%	Middleton & Ambrose 2005
Canada goose	Faecal coliform	1.5×10^4	Alderisio & DeLuca 1999
Canada goose	Faecal coliform	3.6×10^4	Hussong et al. 1979
Canada goose	enterococci	7.3×10^5	Middleton & Ambrose 2005
Duck	*E.coli*	89%	Fallacara et al. 2001
Duck	*E.coli*	$\geq 1.0 \times 10^5$	Murphy et al. 2005
Gull	*E.coli*	$< 1.0 \times 10^5 - 10^9$	Fogarty et al. 2003
Gull	Faecal coliform	3.69×10^8	Alderisio & DeLuca 1999
Gull	Faecal coliform	$1.2 \times 10^6 - > 1.1 \times 10^{10}$	Lévesque et al. 2000
Gull	Faecal coliform	1.7×10^6	Wood & Trust 1972
Gull	Enterococcus faecalis	0–38%	Kuntz et al. 2004
Gull	enterococci	1.8×10^5	Wood & Trust 1972
Gulls	Faecal streptococcus	2.2×10^4–1.5×10^6	Pourcher et al. 1991

2000). Although the black swan is widespread in both Australia and New Zealand, literature searches did not reveal any studies on *E. coli* prevalence or counts for this bird.

While almost all the faeces from ducks, gulls and Canada geese contained enterococci, this organism was recovered from only 79% of the black swan faeces. Enterococci counts were similar to the *E. coli* counts, with the same pattern of highest counts in duck faeces, and the lowest counts in Canada geese, the latter being four orders of magnitude lower. There are few published studies of enterococci counts or incidences in waterfowl (Table 2).

Direct comparisons of the current data with previously published data are hindered by methodological differences used in each of these studies to enumerate the organisms. The data from our study and those previously reported (Table 2 and 3) clearly demonstrate

Table 3 Prevalence and concentration of microorganisms in avian faeces from literature.

Bird	Microorganism	Conc./ prevalence	Reference
Canada goose	*Cryptosporidium*	23.4%	Zhou et al. 2004
Canada goose	*Salmonella*	0%	Hussong et al. 1979
Duck	*C. jejuni*	45%	Luechtefeld et al. 1980
Gull	*Cryptosporidium*	0	Moore et al. 2002
Gull	*Salmonella*	6.3%	Ferns & Mudge 2000
Gulls	*Salmonella*	51%	Sixl et al. 1997
Gulls	*Campylobacter* spp.	$4.9 \times 10^1 - 1.7 \times 10^5$	Fenlon et al. 1982
Gulls	*Campylobacter* spp.	3.0×10^3–1.2×10^7	Lévesque et al. 2000
Gulls		14%	Moore et al. 2002
Free living waterfowl	*Campylobacter* spp.	50%	Fallacara et al. 2001
Migratory birds	*Campylobacter* spp.	5%	Pacha et al. 1988
Swan	*Salmonella*	0%	Hussong et al. 1979
Wild birds	*Campylobacter* spp.	12.5	French et al. 2009

Table 4 Spearman Rank correlation coefficients (r_s) between microbial species.

	Spearman Rank correlation coefficients (r_s) between microbial species			
Bird species	*E. coli*–*Campylobacter*	Enterococci– *Campylobacter*	*E. coli*–Enterococci	*n*
Gull	0.118	0.134	**0.447**	80
Duck	**0.366**	**0.439**	**0.540**	80
Canada goose	**0.245**	0.017	**0.403**	80
Black swan	0.130	−0.041	0.018	80
All birds	**0.116**	0.049	**0.489**	320

Values in bold are significantly different from 0 with a significance level alpha = 0.05.

that there is a large variability in concentration of these indicator bacteria in the faeces of individual wildfowl types.

Campylobacter spp. prevalence ranged from 29–59%, with a ranking of gulls > black swans > Canada geese > ducks. The highest average concentration was found in Canada geese, followed by gulls, black swans and ducks. Despite the use of an MPN system, which can measure up to 10^5 g^{-1} (i.e. spanning up to five dilutions), a large number of samples (35/320) in our study contained concentrations of *Campylobacter* spp. in excess of our upper limit of detection.

The reasonably high prevalence (28.8–45.0%) and concentrations (up to 10^5 g^{-1}) of *Campylobacter* spp. in waterfowl faeces, suggest that waterfowl may constitute significant reservoirs for this organism. Previous studies (Table 3) have documented a range of prevalence of *Campylobacter* spp. in the faeces of waterfowl. However, the proportion of zoonotic strains and, thereby, the avian contribution to campylobacteriosis in New Zealand, is not yet fully understood. The genetic fingerprinting techniques of Pulse Field Gel Electrophoresis (PFGE) and Multi Locus Sequence Typing (MLST) were carried out on *C. jejuni* isolated from wild bird faeces in children's playgrounds in New Zealand. The results of these molecular typing techniques revealed profiles for the *C. jejuni* isolates, which were indistinguishable from previous human cases in New Zealand (French et al. 2009). This suggests that wild birds may shed strains of *Campylobacter* spp., which are associated with human illness.

Concentrations of *Campylobacter* spp. reported in the literature in avian faeces vary widely (Table 3). Waldenstrom et al. (2002) noted that the prevalence of *Campylobacter* spp. infection in wild birds seems to be linked to various ecological and phylogenetic factors, with considerable variation in carriage rates among different taxa and species.

The Spearman correlation analysis (Table 4) confirms that in freshly deposited faeces, *E. coil* is a reasonable indicator for the level of enteroccoci. The presence of fresh avian faecal material is a good indicator that *Campylobacter* spp may be present. The relative concentrations of *E. coli* and *Campylobacter* spp. in the wildfowl samples assayed in our survey also enable preliminary calculations on microbial impact of waterfowl faeces on water quality. If all the *Campylobacter* from these wildfowl are assumed to be equally infective to humans, then one implication of this study is that in water contaminated with wildfowl faeces, assessments of water quality using *E. coli* or enterococci need to take account of particular wildfowl present to evaluate the risk of *Campylobacter* infection. For example, our results suggest that water containing 1000 *E. coli* per 100 ml, may only contain between 0.1 and 0.001 *Campylobacter* per 100 ml water if the faeces are from black swans, gulls or ducks, but it will contain more than 100 *Campylobacter* per 100 ml water if all the *E. coli* present are from Canada geese. This is because of the higher concentration of

Campylobacter spp. per gram in Canada goose faeces, compared with the other wildfowl sampled in our study. The absence of *Salmonella* spp. in the faeces of the waterfowl surveyed in our study concurs with the findings of Hussong et al. (1979), although highly variable concentrations of *Salmonella* spp. have been reported for gulls (Table 3).

Cryptosporidium spp. was present in the faeces of four Canada geese, two black swans and one duck. As speciation was not carried out, their potential health impacts cannot be determined. Similarly, Moore et al. (2002) did not isolate *Cryptosporidium* spp. from 205 gull faecal specimens. In contrast, Zhou et al. (2004) found that 23.4% of Canada goose faecal samples contained *Cryptosporidium* spp., which is considerably higher than the prevalence determined in our study. However, only 10.2% of the isolates were potentially zoonotic strains (*C. parvum*/*C. hominis*), suggesting that Canada geese play a minor role in the animal to human transmission.

The daily faecal output of various bird species has been measured by a number of researchers. Daily wet weight faecal outputs have been estimated at 50 g for gulls (Wood & Trust 1972), 250 g for Canada geese (Hussong et al. 1979), 418 g for black swan (Mitchell & Wass 1995) and 336 g for ducks (Geldreich 1966). These data allow preliminary estimates to be made of the likely daily microbial outputs of the selected wildfowl (Table 1). However, their relative contributions to the microbial pollution of a water body will obviously depend on the sizes of the bird populations in a particular region, and their overall proximities to the waters.

The data from this survey, together with the results of an associated study of the survival of enteric indicators and pathogens in Canada goose faeces, will be used contribute to the knowledge of microbial carriage in waterfowl. It will assist modellers in determining the size and wide variability in concentration of key enteric microbes in waterfowl in New Zealand.

Acknowledgements

The authors wish to thank Mr Robert Van Duivenboden and Mrs Fariba Nourozi for collecting samples. Dr Andrea Donnsion, AgResearch, and Dr Hilary Michie, ESR, provided valuable review comments. This research was funded from the New Zealand Public Good Science Fund and Envirolink TSDC30, administered by the Foundation of Research Science and Technology.

References

Albarnaz JD, Toso J, Correa AA, Simoes CuM, Barardi C.lR 2007. Relationship between the contamination of gulls (*Larus dominicanus*) and oysters (*Crassostrea gigas*) with Salmonella serovar *Typhimurium* by PCR-RFLP. International Journal of Environmental Health Research 17: 133–140.

Alderisio KA, DeLuca N 1999. Seasonal enumeration of fecal coliform bacteria from the feces of ring-billed gulls (*Larus delawarensis*) and Canada geese (*Branta canadensis*). Applied and Environmental Microbiology 65: 5628–5630.

Chaban B, Musil KM, Himsworth CG, Hill JE 2009. Development of cpn60-based real-time quantitative PCR assays for the detection of 14 *Campylobacter* species and application to screening of canine fecal samples. Applied and Environmental Microbiology 75: 3055–3061.

Fallacara DM, Monahan CM, Morishita TY, Wack RF 2001. Fecal shedding and antimicrobial susceptibility of selected bacterial pathogens and a survey of intestinal parasites in free-living waterfowl. Avian Diseases 45: 128–135.

Fenlon DR, Reid TMS, Porter IA 1982. Birds as a source of *Campylobacter* infections. In: Newel DJ ed. Campylobacter: epidemiology, pathogenesis and biochemistry. Lancaster, MTP Press Limited.

Ferns PN, Mudge GP 2000. Abundance, diet and *Salmonella* contamination of gulls feeding at sewage outfalls. Water Research 34: 2653–2660.

Fish and Game New Zealand 2009. Hunting in New Zealand. http://wwwfishandgameorgnz/Site/HuntingNZ/Huntingducksgeeseandswansaspx 2009.

Fogarty LR, Haack SK, Wolcott MJ, Whitman RL 2003. Abundance and characteristics of the recreational water quality indicator bacteria *Escherichia coli* and enterococci in gull faeces. Journal of Applied Microbiology 94: 865–878.

French NP, Midwinter A, Holland B, Collins-Emerson J, Pattison R, Colles F, Carter P 2009.

Molecular epidemiology of *Campylobacter jejuni* isolates from wild-bird fecal material in children's playgrounds. Applied and Environmental Microbiology 75: 779–783.

Geldreich EE 1966. Sanitary significance of faecal coliforms in the environment: U.S Department of the Interior, Federal Water Pollution Control Administration.

Heather B, Robertson H 2005. The field guide to the birds of New Zealand. Auckland, Viking Books.

Hussong D, Damare JM, Limpert RJ, Sladen WJ, Weiner RM, Colwell RR 1979. Microbial impact of Canada geese (*Branta canadensis*) and whistling swans (*Cygnus columbianus columbianus*) on aquatic ecosystems. Applied and Environmental Microbiology 37: 14–20.

Jellison KL, Distel DL, Hemond HF, Schauer DB 2004. Phylogenetic analysis of the hypervariable region of the 18S rRNA gene of *Cryptosporidium* oocysts in feces of Canada geese (*Branta canadensis*): evidence for five novel genotypes. Applied and Environmental Microbiology 70: 452–458.

Kassa H, Harrington BJ, Bisesi MS 2004. Cryptosporidiosis: a brief literature review and update regarding *Cryptosporidium* in feces of Canada geese (*Branta canadensis*). Journal of Environmental Health 66: 45.

Kullas H, Coles M, Rhyan J, Clark L 2002. Prevalence of *Escherichia coli* serogroups and human virulence factors in faeces of urban Canada geese (*Branta canadensis*). International Journal of Environmental Health Research 12: 153–162.

Kuntz RL, Hartel PG, Rodgers K, Segars WI 2004. Presence of *Enterococcus faecalis* in broiler litter and wild bird feces for bacterial source tracking. Water Research 38: 3551–3557.

Lévesque B, Brousseau P, Bernier F, Dewailly É, Joly J 2000. Study of the bacterial content of ring-billed gull droppings in relation to recreational water quality Water Research 34: 1089–1096.

Luechtefeld NA, Blaser MJ, Reller LB, Wang WL 1980. Isolation of *Campylobacter* fetus subsp. *jejuni* from migratory waterfowl. Journal of Clinical Microbiology 12: 406–408.

McBride GB 2005. Using statistical methods for water quality management: issues, problems and solutions. Hoboken, NJ, John Wiley.

Middleton JH, Ambrose A 2005. Enumeration and antibiotic resistance patterns of fecal indicator organisms isolated from migratory Canada geese (*Branta canadensis*). Journal of Wildlife Diseases 41: 334–341.

Ministry for the Environment 2003. Microbiological water quality guidelines for marine and freshwater recreational areas. Ministry for the Environment and Ministry of Health.

Mitchell SF, Wass RT 1995. Food consumption and faecal deposition of plant nutrients by black swans (*Cygnus arratus* Latham) in a shallow New Zealand lake. Hydrobiological Journal 29: 189–197.

Moore JE, Gilpin D, Crothers E, Canney A, Kaneko A, Matsuda M 2002. Occurrence of *Campylobacter* spp., *Cryptosporidium* spp. in gulls (Larus spp.). Vector Borne and Zoonotic Diseases 2: 111–114.

Moriarty EM, Sinton LW, Mackenzie ML, Karki N, Wood DR 2008. A survey of enteric bacteria and protozoans in fresh bovine faeces on New Zealand dairy farms. Journal of Applied Microbiology 105: 2015–2025.

Murphy J, Devane ML, Robson B, Gilpin BJ 2005. Genotypic characterization of bacteria cultured from duck faeces. Journal of Applied Microbiology 99: 301–309.

Ng J, Pavlasek I, Ryan U 2006. Identification of novel *Cryptosporidium* genotypes from avian hosts. Applied and Environmental Microbiology 72: 7548–7553.

Nielsen EM, Skov MN, Madsen JJ, Lodal J, Jespersen JB, Baggesen DL 2004. Verocytotoxin-producing *Escherichia coli* in wild birds and rodents in close proximity to farms. Applied and Environmental Microbiology 70: 6944–6947.

Pacha RE, Clark GW, Williams EA, Carter AM 1988. Migratory birds of central Washington as reservoirs of *Campylobacter jejuni*. Canadian Journal of Microbiology 34: 80–82.

Pourcher AM, Devriese LA, Hernandez JF, Delattre JM 1991. Enumeration by a miniaturized method of *Escherichia coli, Streptococcus bovis* and enterococci as indicators of the origin of faecal pollution of waters. Journal of Applied Bacteriology 70: 525–530.

Quessy S, Messier S 1992. Prevalence of *Salmonella* spp., *Campylobacter* spp, *Listeria* spp. in ring-billed gulls (*Larus delawarensis*). Journal of Wildlife Diseases 28: 526–531.

Rohela M, Lim YA, Jamaiah I, Khadijah PY, Laang ST, Nazri MH, Nurulhuda Z 2005. Occurrence of *Cryptosporidium* oocysts in wrinkled hornbill and other birds in the Kuala Lumpur National Zoo. Southeast Asian Journal of Tropical Medicine and Public Health 36 Suppl 4: 34–40.

Sixl W, Karpiskova R, Hubalek Z, Halouzka J, Mikulaskova M, Salava J 1997. *Campylobacter* spp., *Salmonella* spp. in black-headed gulls (*Larus ridibundus*). Central European Journal of Public Health 5: 24–26.

Wahlstrom H, Tysen E, Olsson Engvall E, Brandstrom B, Eriksson E, Morner T, Vagsholm I 2003. Survey of *Campylobacter* species, VTEC O157 and *Salmonella* species in Swedish wildlife. Veterinary Record 153: 74–80.

Waldenstrom J, Broman T, Carlsson I, Hasselquist D, Achterberg RP, Wagenaar JA, Olsen B 2002. Prevalence of *Campylobacter jejuni, Campylobacter lari*, and *Campylobacter coli* in different ecological guilds and taxa of migrating birds. Applied and Environmental Microbiology 68: 5911–5917.

Wither A, Rehfisch M, Austin G 2005. The impact of bird populations on the microbiological quality of bathing waters. Water Science and Technology 51: 199–207.

Wong T, Devane ML, Hudson JA, Scholes P, Savill MG, Klena JD 2004. Validation of a PCR method for *Campylobacter* detection on poultry packs. British Food Journal 106: 642–650.

Wood AJ, Trust TJ 1972. Some qualitative and quantitative aspects of the intestinal microflora of the glaucous-winged gull (*Larus glaucescens*). Canadian Journal of Microbiology 18: 1577–1583.

Yakub GP, Castric DA, Stadterman-Knauer KL, Tobin MJ, Blazina M, Heineman TN, Yee GY, Frazier L 2002. Evaluation of Colilert and Enterolert defined substrate methodology for wastewater applications. Water Environment Research 74: 131–135.

Zhou L, Kassa H, Tischler ML, Xiao L 2004. Host-adapted *Cryptosporidium* spp. in Canada geese (*Branta canadensis*). Applied and Environmental Microbiology 70: 4211–4215.