





## Draft Whole-Genome Sequences of Five Klebsiella pneumoniae Isolates from the Subantarctic Islands of New Zealand

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ABSTRACT Klebsiella pneumoniae is a Gram-negative bacterium that can be found in the environment, as well as on mucosal surfaces of humans and animals. Here, we report the genome sequence of five K. pneumoniae isolates from substrate samples and bird feces collected in the Subantarctic Islands of New Zealand.

Plebsiella pneumoniae is an opportunistic pathogen causing nosocomial and community-acquired infections (1, 2). Over the past few decades, hypervirulent strains causing primary liver abscesses and septicemia have increasingly been documented (3, 4). Most hypervirulent strains have a hypermucoviscous phenotype (positive string test) and the rmpA and rmpA2 genes (1, 3, 4). Hypervirulent strains have also been described in animals (5-7).

Hypervirulent K. pneumoniae infection has been reported as a cause of mortality events in New Zealand sea lion (NZSL) pups on Enderby Island, New Zealand, one of the Subantarctic Auckland Islands (7, 8). Annually, a large number of pups die from K. pneumoniae infection in the summer birthing season; however, reservoirs have not been investigated. Environmental substrates, birds that live on the island, and NZSL adults are possible reservoirs of this infection.

The use of animals and sample collection were completed under permit 39915-FAU from the New Zealand Department of Conservation and Massey University Animal Ethics Committee (approval 14/114). In this study, five K. pneumoniae isolates from cloacal swabs or voided feces from subantarctic skuas (n=2) and a yellow-eyed penguin (n = 1) and substrate samples (water; n = 2) (using CHROMagar Orientation) were whole-genome sequenced. The NucleoSpin soil kit (Macherey-Nagel, GmbH & Co. KG, Germany) was used to extract genome-quality DNA from a single colony cultured on agar, which was sent to New Zealand Genomics Limited (Massey Genome Service, Massey University, Palmerston North, New Zealand). A fragment library was prepared using an Illumina TruSeq DNA library preparation kit v1 (Illumina, Inc., Scorsby, Victoria, Australia). Paired-end reads (2 imes 250 bp) were obtained from a MiSeq instrument (Illumina, Inc., San Diego, CA). The five isolates from this study were de novo assembled using SPAdes v3.10 (in the "careful" mode) (9). The contigs of each isolate produced from SPAdes was annotated by Prokka v1.1.2, with default parameters (10). The sequence types (ST) and serotypes of bacterial isolates were determined using the Bacterial Isolate Genome Sequence Database (BIGSdb) servers (http://bigsdb.pasteur .fr/klebsiella/klebsiella.html). Virulence genes were identified by mapping the reads of each isolate, along with 1,000-bp flanks on either side of the virulence gene sequence, using Bowtie 2 in both the "-very-sensitive-local" and "-very-sensitive" modes for local and global read mapping, respectively. Genome sizes, numbers of contigs, ST, sources, serotypes, and virulence genes are summarized in Table 1.

The virulence genes found in environmental and bird isolates were consistent with

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TABLE 1 Descriptions of K. pneumoniae strains sequenced, their genomic characteristics, and associated virulence factors

	GenBank					String Length No. of Fold GC	Length	No. of	Fold	gc	
Isolate	accession no.	accession no. SRA accession no. Source, location	Source, location	Serotype	ST	ST test result (bp)		contigs	coverage	content (%)	contigs coverage content (%) Virulence genes
E14_15_17Sa	E14_15_17Sa QVNJ00000000 SRR7699496	SRR7699496	Subantarctic skua, <sup>a</sup>	Non-K1/1K2 2843	2843	ı	5,644,626 116	116	87	57.1	wabG, uge, irp2, iucD, iutA, mrkD
E14_15_42Sa	E14_15_42Sa QVNI000000000 SRR7699497	SRR7699497	Enderby Island Subantarctic skua, <sup>a</sup>	2	98	+	5,327,210 94	94	93	57.5	rmpA, wabG, uge, iroN, irp2, ybtS, mrkD
E14_15_53Ma	E14_15_53Ma QVNH00000000 SRR7699498	SRR7699498	Enderby Island Yellow-eyed penguin, <sup>a</sup>	S	98	+	5,351,349 91	91	89	57.5	rmpA, wabG, uge, iroN, irp2, ybtS, mrkD
E13_14_10sub	E13_14_10sub QVNG00000000 SRR7699499	SRR7699499	Enderby Island Water, Enderby Island K2	Ş	98	+	5,332,787 85	85	86	57.5	rmpA, wabG, uge, iroN, irp2, ybtS, mrkD
C14_15_17suk	214_15_17sub QVNF00000000 SRR7699495	SRR7699495	Water, Campbell Island K2	S	+ 98	+	5,323,133 99	66	94	57.5	rmpA, wabG, uge, iroN, ybtS, mrkD
<sup>a</sup> This animal wa	as apparently healthy	ı. K. pneumoniae was isc	<sup>2</sup> This animal was apparently healthy. <i>R. pneumoniae</i> was isolated from cloacal swabs or voided feces from nonclinical, live animals.	voided feces fro	om nonc	linical, live an	ıimals.				

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those found in other studies on clinical human isolates (11), suggesting that the isolates have pathogenic potential. This further suggests that the environment and birds may be possible reservoirs of this pathogen. The ST of the environmental isolates from this study was different from that in previous reports on other environmental isolates (12), suggesting genetic diversity of this bacterium in the environment (13).

These are the first whole-genome sequences of *K. pneumoniae* isolated from birds and environmental substrate samples from the Subantarctic Islands. The data from this study will provide information on genomic relationships between isolates from animals and environmental isolates in the Subantarctic and help to inform future research on the role of *K. pneumoniae* in harsh non-human-dominated island environments.

**Data availability.** The whole-genome shotgun sequences described here have been deposited in DDBJ/ENA/GenBank under the accession numbers listed in Table 1. Raw sequence reads have been deposited in the NCBI Sequence Read Archive under the accession numbers listed in Table 1.

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