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Pseudomonas and *Pedobacter* isolates from King George Island inhibited the growth of foodborne pathogens

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Abstract: This report describes the isolation and characterization of bacterial isolates that produce anti-microbial compounds from one of the South Shetland Islands, King George Island, Antarctica. Of a total 2465 bacterial isolates recovered from the soil samples, six (BG5, MTC3, WEK1, WEA1, MA2 and CG21) demonstrated inhibitory effects on the growth of one or more Gram-negative or Gram-positive indicator foodborne pathogens (*i.e. Escherichia coli* 0157:H7, *Salmonella* spp., *Klebsiella pneumoniae*, *Enterobacter cloacae*, *Vibrio parahaemolyticus* and *Bacillus cereus*). Upon examination of their 16S rRNA sequences and biochemical profiles, the six Antarctic bacterial isolates were identified as Gram-negative *Pedobacter cryoconitis* (BG5), *Pseudomonas migulae* (WEK1), *P. corrugata* (WEA1) and *Pseudomonas* spp. (MTC3, MA2, and CG21). While inhibitors produced by strains BG5, MTC3 and CG21 were sensitive to protease treatment, those produced by strains WEK1, WEA1, and MA2 were insensitive to catalase, lipase, α-amylase, and protease enzymes. In addtion, the six Antarctic bacterial isolates appeared to be resistant to multiple antibiotics.

Introduction

Antarctica is the most pristine continent on Earth with a surface area of 14 million km^2. The temperature in Antarctica is extremely low throughout the year ex-

cept during the summer months where the ice on the soils is subjected to thawing (Convey *et al.* 2008). Due to the harsh conditions, microorganisms living in the continent and the neighboring islands have acquired unique adaptation strategies to survive in the extreme environment. In order to gain competitive advantage some microorganisms produce extracellular antimicrobial compounds to inhibit the growth of their competitors (Russell 2006; Lo Giudice *et al.* 2007a). These antimicrobial compounds may have medical applications.

Over the last few decades microorganisms from around the world are being harnessed for antimicrobial compounds. However, the antimicrobial activities of microorganisms from the Antarctic, especially the bacteria, have only been investigated recently. Terrestrial (Moncheva *et al.* 2002; Nediakova and Naidenova 2004; O'Brien *et al.* 2004; Biondi *et al.* 2008) and marine bacteria (De Souza *et al.* 2006; Lo Giudice *et al.* 2007a, b) from several locations in the Antarctic are found to produce a variety of antimicrobial compounds such as antibiotics and bacteriocins (O'Brien *et al.* 2004; Biondi *et al.* 2008). These reports indicated that there are substantial numbers of novel bacteria with antimicrobial activities in the Antarctic. This opens up hope for the discovery of new classes of antibiotices considering that there are only few that are found elsewhere over the past decades (Cassell and Mekalanos 2001). Nevertheless, the geographical locations covered in Antarctic are limited. For example, there is little information pertaining to antimicrobial compounds of bacteria from other locations in Antarctica such as the South Shetland Islands. Hence, this project was conducted (i) to estimate the population of cultivable bacteria with antimicrobial activity from the soil samples collected from King George Island, Antarctic, and (ii) to characterize the bacteria with antimicrobial activity and the antimicrobial compounds they produce.

Materials and methods

Isolation of Antarctic bacteria. — Soil samples were collected from King George Island (62°09'30.0" S, 58°56'15.2" W), one of the South Shetland Islands during the 43rd Scientific Antarctic Expedition organized by the Instituto Antártico Chileno (INACH) in 2007. Samples were collected using sterilized spatula and stored in sterilized containers at -20°C for 10 to 14 days. Isolation of the Antarctic bacteria was performed using several growth media namely, Tryptic Soy agar (TSA) (Difco), Luria-Bertani agar (LBA), Nutrient agar and R2A agar (Difco). One gram of the soil sample was inoculated into 10 ml of sterile distilled water or potassium phosphate buffer (pH 7.2), serially diluted to 100 times and plated on the agar medium. The agar plates were incubated at 20°C between two to ten days for the recovery of the Antarctic bacteria.

Detection of antimicrobial activity. — Antimicrobial compound production was determined using deferred antagonism procedure (Kekessy and Piguet 1970).

Antarctic bacteria colonies on the agar plate were overlaid with 10 ml molten nutrient agar (1.3% nutrient broth and 0.7% agar) containing one of the indicator bacteria. Zones of clearing around the Antarctic bacteria after 2 days of incubation indicated the presence of inhibitors. Indicator bacteria used were foodborne pathogens namely: *Escherichia coli* 0157:H7, *E. coli* V517, *E. coli* 0125, *Salmonella enterica* serovar Typhimurium (S. Tm 13), *S. enterica* serovar Typhi (S. Ty 10), *S. biafra* (S. Bi 8), *S. braenderup* (S. Br. D), *Klebsiella pneumoniae* 14x, *Enterobacter cloacae* 22x, *Vibrio parahaemolyticus* 1808, *V. parahaemolyticus* 1896, *V. parahaemolyticus* 2053, *V. parahaemolyticus* 2341 and *Bacillus cereus* K3. These foodborne pathogens were provided by Professor Son Radu, Universiti Putra Malaysia, Malaysia.

Sensitivity of the antimicrobial agents to enzymes. — A series of enzymes namely: protease (EC 3.4.24.31) (Sigma), catalase (EC 1.11.1.6) (Sigma), lipase (EC 3.1.1.3) (Sigma), α-amylase (EC 3.2.1.1) (Sigma), were used to determine the properties of the antimicrobial agents produced by the Antarctic bacteria. All the enzymes were prepared at a concentration of 25 mg ml^{-1}, according to the manufacturer's instructions. Deferred antagonism assays were performed according to O'Brien *et al.* (2004). The assay plate was incubated at 20°C for 12 hours.

Identification of the Antarctic bacteria. — Genomic DNA was extracted using the DNeasy Blood and Tissue Kit (Qiagen) according to the manufacturer's instruction. PCR amplification of 16S rDNA was performed using the primers, BSF8/20 5'-aga-gtt-tga-tcc-tgg-ctc-ag-3' and BSR1541/20 5'-aag-gag-gtg-atc-cag-ccg-ca-3' (Wilmotte *et al.* 1993). The PCR conditions were 96°C for 2 min, followed by 30 cycles at 96°C for 30 sec, 52°C for 40 sec, 72°C for 40 sec and a final extension step at 72°C for 10 min. The 16S rDNA was sequenced using the Automated Biosystems Sequencer AB3100, and the sequence was analyzed using the BLASTn software (Altschul *et al.* 1997).

Biochemical profiles of the Antarctic bacteria. — Catalase activity was tested according to the methods described by Lee *et al.* (2000), while the presence of oxidase activity was tested according to the manufacturer's instructions (bioMerieux, Marcy-l'Etoile, France). Biochemical profiles of the bacteria were determined using the API 20NE strips (bioMerieux). The incubation times of the API strips were between 4–5 days at 20°C. Motility test was conducted according to the methods described by Tittsler and Sandholzer (1936).

Antibiotic susceptibility and resistance tests. — Antibiotics susceptibility test for the Antarctic bacterial isolates were performed by the disc diffusion method on tryptic soy agar medium. The antimicrobial agent tested included ampicillin (10 µg), ceftazidime (30 µg), imipenem (10 µg), ciprofloxacin (5 µg), chloramphenicol (30 µg), gentamicin (10 µg), kanamycin sulfate (30 µg), streptomycin (10 µg), tetracycline hydrochloride (30 µg), polymyxin B sulfate (300 units), erythromycin (15 µg), clindamycin (2 µg), lincomycin (10 µg), novobiocin (5 µg), and vancomycin (30 µg).

Results

Host spectrum of inhibitor-producing Antarctic bacteria. — A total of 2465 Antarctic bacterial isolates were picked and screened for antimicrobial activity against a series of foodborne pathogens. Six isolates, BG5, MTC3, WEK1, WEA1, MA2 and CG21 were found to inhibit four or more pathogen strains out of the 14 tested (Table 1). Isolates BG5 and MTC 3 inhibited the growth of 8 and 6 strains of pathogens respectively. Isolates WEK1 and WEA1 inhibited the growth of 5 strains of pathogens while isolates MA2 and CG21 inhibited the growth of 4 strains of pathogens.

Antarctic bacterial isolates WEK1, WEA1 and MA2 inhibited the growth of Gram-negative pathogens *V. parahaemolyticus* (1808, 2053 and 2341) and Gram-positive pathogen *B. cereus* (Table 1), but did not inhibit the growth of *E. coli*, *Salmonella* spp., *K. pneumoniae* or *E. cloacae*. In contrast, isolates BG5, MTC3 and CG21 inhibited the growth of *E. coli* (O157:H7, V517 and 0125) and *E. cloacae* but not *V. parahaemolyticus* (1808, 1896, 2053 and 2341) (Table 1). Bacterial isolate BG5 inhibited the growth of *S. enterica* serovar Typhimurium, *S. enterica* serovar Typhi, *K. pneumoniae* and *B. cereus*, while isolate MTC3 inhibited the growth of *S. enterica* serovar Typhimurium and *K. pneumoniae* but not *B. cereus*. None of the antimicrobial compounds produced by the six Antarctic bacterial isolates inhibited the growth of pathogens *S. biafra* and *S. braenderup*.

Partial characterization of the antimicrobial compounds. — The properties of the antimicrobial compounds of the 6 bacterial isolates BG5, MTC3, WEK1, WEA1, MA2 and CG21 were partially resolved by testing the sensitivities of these compounds towards several enzymes (data not shown). None of the

Table 1

Identification of antimicrobial producers against the foodborne pathogens, *E. coli* O157:H7, *E. coli* V517, *E. coli* 0125, *S. enterica* serovar Typhimurium (S. Tm 13), *S. enterica* serovar Typhi (S. Ty 10), *S. biafra* (S. Bi 8), *S. braenderup* (S. Br D), *K. pneumoniae* 14x, *E. cloacae* 22x, *V. parahaemolyticus* 1808, *V. parahaemolyticus* 1896, *V. parahaemolyticus* 2053, *V. parahaemolyticus* 2341 and *B. cereus* K3. Size of the inhibition zone of the foodborne pathogen (mm); (–), no inhibition zone

Antarctic bacterial isolates	*E. coli*			*Salmonella* spp.				*K. pneumonia*	*E. cloacae*	*V. parahaemolyticus*				*B. cereus*
	O157:H7	V517	0125	S. Tm 13	S. Ty 10	S. Bi 8	S. Br D	14X	22X	1808	1896	2053	2341	K3
BG5	23	19	18	4	9	–	–	15	27	–	–	–	–	12
MTC3	16	5	16	15	–	–	–	5	8	–	–	–	–	–
WEK1	–	–	–	–	–	–	–	–	–	12	17	15	14	21
WEA1	–	–	–	–	–	–	–	–	–	10	15	18	15	18
MA2	–	–	–	–	–	–	–	–	–	12	–	19	14	17
CG21	11	16	18	–	–	–	–	–	4	–	–	–	–	–

antimicrobial compounds produced by the six bacterial isolates were sensitive to treatment with catalase, lipase, or α-amylase. However, the inhibitors produced by the isolates BG5, MTC3 and CG21 were sensitive to protease while those produced by the isolates WEK1, WEA1 and MA2 were not. Interestingly, the inhibitors of the six Antarctic bacterial isolates against the pathogens were affected at elevated temperature. Inhibitory effect against the pathogens was detected at 20°C but not when the Antarctic bacteria were incubated at 30°C.

Identification of the Antarctic bacteria. — The 16S rDNA sequence (approximately 1.5 kb) alignment analysis of the six Antarctic bacterial isolates revealed that five isolates MTC3, WEK1, WEA1, MA2 and CG21 had high similarity to the genera: *Pseudomonas* from the phylum of γ-Proteobacteria (Table 2). Bacterial isolates MTC3, WEA1 and WEK1 had the highest similarity to the *Pseudomonas* sp. DhA-91 (99%), *Pseudomonas corrugata* (99%) and *Pseudomonas migulae* (98%) respectively. MA2 and CG21 had the highest similarity to the *Pseudomonas* sp. DM2 with 97% and 98% similarity respectively. The other isolate, BG5 was similar to the *Pedobacter cryoconitis* (99%) from the phylum of Bacteroidetes (Table 2). The 16S rDNA sequences of the six bacterial isolates were deposited in the GenBank and assigned to accession numbers EU637886, EU637887, EU547450, EU547451, EU908689 and EU908688 for isolates BG5, MTC3, WEA1, WEK1, MA2 and CG21 respectively.

Table 2

The 16S rDNA sequence affiliation of the six Antarctic bacterial isolates to their nearest phylogenetic neighbors

Phylum	Strains	Gene bank accession number	Nearest taxonomic neighbor/ accession number	Identities	Similarity (%)
Bacteroidetes	BG5	EU637886	*Pedobacter cryoconitis*/AJ438170	1492/1495	99
γ-Proteobacteria	MTC3	EU637887	*Pseudomonas* sp. DhA-91/AF177916	1521/1531	99
γ-Proteobacteria	WEA1	EU547450	*Pseudomonas corrugata*/AF348508	1516/1542	99
γ-Proteobacteria	WEK1	EU547451	*Pseudomonas migulae*/AF074383	1507/1530	98
γ-Proteobacteria	MA2	EU908689	*Pseudomonas* sp. DM2/FJ517635	1496/1531	97
γ-Proteobacteria	CG21	EU908688	*Pseudomonas* sp. DM2/FJ517635	1510/1520	98

Some of the 2465 Antarctic bacterial isolates, that had pigmented colony but did not exhibit any inhibitory effect against the foodborne pathogens tested, were also identified. Although, these isolates did not inhibit the test pathogens used in this study they might have an effect on other pathogens, because antibiotic production was linked to pigmentation of the bacterial strains (Choi *et al.* 2001; Lo Giudice *et al.* 2007a). They were from the genera: *Arthrobacter* (10 isolates), *Aeromicrobium* (2 isolates), *Brevundimonas* (2 isolates), *Cryobacterium* (4 isolates), *Dyadobacte*r (2 isolates), *Flavobacterium* (2 isolates), *Methylibium* (1 isolate), *Rhodococcus* (7 isolates) and *Sphingomonas* (2 isolates) (16S rDNA sequence data not shown).

Characterization of the Antarctic bacteria. — All the 6 isolates BG5, MTC3, WEK1, WEA1, MA2 and CG21 were Gram-negative bacteria. They did not produce hydrogen sulfide or indole (Table 3); nor did they reduce nitrate, ferment glucose, assimilate adipic acid, trisodium citrate or phenylacetic acid. They were divided into two groups based on their characteristics.

Group one consisted of *Pseudomonas* spp. WEK1, WEA1 and MA2. They were motile, Gram-negative and grew on MacConkey agar medium. However,

Table 3

Morphologies and biochemical profiles of inhibitor-producing bacterial isolates: +, positive; –, negative; ±, variable

Characteristics	BG5	MTC3	WEK1	WEA1	MA2	CG21
Cell morphology	Spirillum	Rod	Spirillum	Spirillum	Rod	Diplococcus
Gram stain	–	–	–	–	–	–
Motility	–	–	+	+	+	–
Growth on MacConkey agar	–	–	+	+	+	–
H_2S production	–	–	–	–	–	–
Acetoin production	+	+	–	–	+	+
Catalase	+	+	+	+	+	+
Reduction of nitrate	–	–	–	–	–	–
Indole production	–	–	–	–	–	–
Fermentation of glucose	–	–	–	–	–	–
Enzyme tests						
Arginine dihydrolase	–	–	+	+	+	–
Urease	–	–	+	+	+	–
β-glucosidase (Esculin)	+	+	+	–	+	+
Protease (Gelatin)	+	+	+	+	+	+
β-galactosidase (PNPG)	+	+	–	–	–	+
Cytochrome oxidase	–	–	+	+	+	–
Carbon sources						
D-Glucose	±	±	±	±	+	±
L-Arabinose	–	±	±	±	+	±
D-Mannose	±	±	±	±	±	±
D-Mannitol	–	–	+	+	+	–
N-acetyl-glucosamine	±	±	+	+	+	–
D-Maltose	+	+	–	–	–	±
Potassium gluconate	–	–	±	±	+	–
Capric acid	–	–	±	–	+	–
Adipic acid	–	–	–	–	–	–
Malic acid	–	–	–	±	±	–
Trisodium citrate	–	–	–	–	–	–
Phenylacetic acid	–	–	–	–	–	–

they differed slightly in their morphologies and biochemical profiles. For example, isolate WEA1 was able to assimilate malic acid but not capric acid, and gave negative reaction to esculin (Table 3). In contrast, isolate WEK1 was able to assimilate capric acid but not malic acid, and gave positive reaction to esculin. Unlike WEA1 or WEK1, isolate MA1 was able to assimilate capric and malic acids, and gave positive reaction to esculin. Additionally, isolates WEK1 and WEA1 were spiral shaped while MA2 was rod shaped.

Pseudomonas spp. MTC3 and CG21, and *Pedobacter* sp. BG5 were assigned to another group. They were non-motile and did not grow on MacConkey agar medium. The three isolates had similar biochemical profile, except that isolates MTC3 and CG21 assimilated L-arabinose while the isolate BG5 did not (Table 3). Additionally, MTC3 and BG5 assimilated *N*-acetyl-glucosamine but CG21 did not. Isolates MTC3, CG21 and BG5 had rod, diplococcus and spiral shapes respectively.

Antibiotics susceptibility. — The results of the antibiotic susceptibility test for the six bacterial isolates BG5, MTC3, WEK1, WEA1, MA2 and CG21 are shown in Table 4. Interestingly, all the six isolates were resistant to ampicillin and vancomycin but were susceptible to imipenem, ciprofloxacin and tetracycline. Isolates WEK1, WEA1, MA2 and CG21 were resistant to 7 out of the 15 antibiotics tested while isolates BG5 and MTC3 were resistant to 9 out of the 15 antibiotics tested.

Table 4

Antibiotics resistance profiles of the six bacterial isolates WEA1, WEK1, BG5, MTC3, CG21 and MA2. R, resistant; S, susceptible

Antibiotics (concentration)	BG5	MTC3	WEA1	WEK1	MA2	CG21
Ampicillin (10 µg)	R	R	R	R	R	R
Ceftazidime (30 µg)	R	R	S	S	S	R
Imipenem (10 µg)	S	S	S	S	S	S
Ciprofloxacin (5 µg)	S	S	S	S	S	S
Chloramphenicol (30 µg)	R	R	R	R	R	S
Gentamicin (10 µg)	R	R	S	S	S	R
Kanamycin sulfate (30 µg)	R	R	S	S	S	R
Streptomycin (10 µg)	R	R	S	S	S	R
Tetracycline hydrochloride (30 µg)	S	S	S	S	S	S
Polymyxin B sulfate (300 units)	R	R	S	S	S	R
Erythromycin (15 µg)	S	S	R	R	R	S
Clindamycin (2 µg)	S	S	R	R	R	S
Lincomycin (10 µg)	S	S	R	R	R	S
Novobiocin (5 µg)	R	R	R	R	R	S
Vancomycin (30 µg)	R	R	R	R	R	R

Discussion

Six out of 2465 isolates inhibited the growth of one or more foodborne pathogens. Hence, the frequency of isolation of inhibitor producers in this study was 0.24 % (6/2465 isolates) which was comparable to bacteria with antimicrobial compounds reported by Lo Giudice *et al.* (2007b) of 0.29% (13/4496 colonies) from Antarctic marine samples. However, the numbers were lower than those reported by O'Brien *et al.* (2004) and Lo Giudice *et al.* (2007a) of 3.8% (22/580 colonies) from the east Antarctic terrestrial samples. In general, these observations suggest that the frequency of isolation of inhibitor producers from Antarctic environment using non-selective media ranged from 0.2% (this work) to 3.8% (O'Brien *et al.* 2004; Lo Giudice *et al.* 2007a).

The antimicrobial compounds from bacterial isolates MTC3 and BG5 had the broadest spectrum among the six isolates, inhibiting the growth of pathogens of four genera (*Escherichia*, *Salmonella*, *Klebsiella* and *Enterobacter*) and five genera (*Escherichia*, *Salmonella*, *Klebsiella*, *Enterobacter* and *Bacillus*) respectively (Table 1). On the other hand, antimicrobial compounds of bacterial isolates WEK1, WEA1, MA2 and CG21 had narrow spectrum. They inhibited the growth of pathogens of two genera only.

All the six Antarctic isolates inhibited the growth of Gram-negative bacteria including pathogens from the genera *Vibrio*, *Klebsiella* and *Enterobacter* that had never been tested on Antarctic bacteria before. In contrast, most of the Antarctic bacteria reported by O'Brien *et al.* (2004) inhibited only Gram-positive bacteria. Additionally, four isolates BG5, WEK1, WEA1 and MA2 also inhibited the growth of the Gram-positive foodborne pathogens and these findings were consistent with those reported by Lo Giudice *et al.* (2007b) although some of the indicator strains used were different.

The results from the enzyme sensitivity tests indicated that the active moieties of the antimicrobial compounds from the six isolates were not sensitive to catalase, lipase and α-amylase and therefore did not contain any hydrogen peroxide, lipid, or glycan. The inhibitors of the bacterial isolates BG5, MTC3 and CG21 were sensitive to protease and elevated temperature, suggesting that their inhibitors might be proteinaceous in nature and could be bacteriocins which were active only at an optimal temperature (Leroy and De Vuyst 1999; Keren *et al.* 2004). Inhibitors of the isolates WEK1, WEA1 and MA2 were not affected by protease treatment and therefore were not likely to be bacteriocins, but they were inactivated at elevated temperature. The reasons for the inactivation were unclear, and would only be known when the inhibitory compounds from those bacterial isolates were purified and characterized.

The six Antarctic bacterial isolates were identified based on the alignments of their 16S rDNA sequences to those of known bacteria in the genebank to find their nearest taxonomic neighbor (Table 2). Isolates WEA1, WEK1 and BG5 were

likely to be *P. corrugata*, *P. migulae* and *Pedobacter cryoconitis*, respectively. The other isolates MTC3, MA2 and CG21 were closest to the *Pseudomonas* spp. and could be new pseudomonad species. These three isolates were likely to be different species because of their different morphology, chemical and pathogen inhibitory profiles. Isolates MTC3 and MA2 were rods while CG21 was a diplococcus. Isolate MA2 was motile, grew on MacConkey agar medium, and inhibited the growth of *V. parahaemolyticus* and *B. cereus*. In contrast, isolates MTC3 and CG21 were non-motile, did not grow on MacConkey agar medium and inhibited the growth of *E. coli* and *E. cloacae*. However, isolate MTC3 was different from CG21 because of its ability to assimilate *N*-acetyl-glucosamine and inhibited an additional pathogen, the *S. enterica* serovar Typhimurium.

The five *Pseudomonas* spp., MTC3, WEK1, WEA1, MA2 and CG21 were from the phylum γ-Proteobacteria which was well known to produce bioactive compounds. Sutherland *et al.* (1985) and Grgurina *et al.* (2005) had isolated mupirocin and syringopeptin from γ-Proteobacteria that showed inhibitory activity against pathogens such as *Haemophilus influenzae*, *Neisseria gonorrhoeae*, *Mycobacterium smegmatis* and *Staphylococcus aureus*. In contrast, *Pedobacter* sp. including isolate BG5 was among the few members from the Bacteroidetes phylum that produced antimicrobial compounds. To our knowledge, this is the first time that *Pedobacter* sp. is reported to produce inhibitors against human pathogens.

The six isolates BG5, MTC3, WEK1, WEA1, MA2 and CG21 were found to be resistant to a series of antibiotics, a phenomenon which was also observed by Kobori *et al.* (1984) and Lo Giudice *et al.* (2007a). The ability of the six Antarctic bacterial isolates in this study to resist to ampicillin suggested that they probably produce β-lactamase to degrade the ampicillin in the agar medium. Their resistance to other β-lactam antibiotics, such as ceftazidime (extended cephalosporins), indicated that the Antarctic bacteria probably had broad spectrum β-lactamases. Nevertheless, additional test on other β-lactams is required to confirm this. The six bacterial isolates in this study were resistant to at least 7 types of common antibiotics. Similar trends were reported by Siebert *et al.* (1996). They found that some of the bacteria from Antarctic sandstone in McMurdo Valley were resistant to one or more antibiotics such as streptomycin, chloramphenicol, erythromycin, ampicillin, cycloserine and tetracycline. Multiple antibiotic resistant strains of environmental bacteria were also found in (i) the river and bay of Tillamork, Oregon (Kelch and Lee 1978), (ii) the ice core from the Greenland (Miteva *et al.* 2004), and the Arctic permafrost subsoil in Siberia (Mindlin *et al.* 2008). These observations implied that the environmental bacteria were probably natural reservoirs of antibiotic resistant genes.

In this study, six or 0.24% of the 2465 bacterial isolates were found to produce antimicrobial compounds against 13 out of the 14 indicator foodborne pathogens. Five of the isolates were from *Pseudomonas* spp. and one from *Pedobacter* sp. Although bacterial isolates from other genera such as *Arthrobacter*, *Aeromicrobium*,

Brevundimonas, *Cryobacterium*, *Dyadobacter*, *Flavobacterium*, *Methylibium*, *Rhodococcus* and *Sphingomonas* were also found in the same soil samples. They did not produce any antimicrobial compounds against any of the 14 indicator pathogens but they were pigmented, and pigmented bacteria had been linked to the production of antibiotics (Shivaji *et al.* 1989; Shivaji *et al.* 1994; Chattopadhyay *et al.* 1997; Choi *et al.* 2001; O'Brien *et al.* 2004; Lo Giudice *et al.* 2007a). Probably these isolates produced antimicrobial compounds with narrow spectrum that targeted pathogens other than those used in this study.

The results from this study showed that the Antarctic bacteria produced antimicrobial compounds, either of broad or narrow spectrum, that targeted a wide range of pathogens. Apart from that, the six Antarctic bacterial isolates were resistant to multiple antibiotics suggesting that these Antarctic bacteria are potential sources of genes encoding for both antimicrobial compounds and resistance to antibiotics for medical applications. From the ecological point of view these two capabilities probably provided the competitive edge to the Antarctic bacteria to survive in the harsh environment (Russell 2006; Lo Giudice *et al.* 2007a), while bacteria lacking such capabilities would be suppressed.

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