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Antimicrobial activity of two Antarctic *Streptomyces* strains

Chen ZiAng[1], Teoh Chui Peng[1], Fan Hui Yin[2], Lavin Paris[3] and Wong Clemente Michael Vui Ling[1]*

[1]Biotechnology Research Institute, Universiti Malaysia Sabah, 88400 Kota Kinabalu, Sabah, Malaysia.
[2]Food Safety and Security Research Group, Faculty of Food Science and Nutrition, Universiti Malaysia Sabah, Jalan UMS, 88400 Kota Kinabalu, Sabah, Malaysia.
[3]Departamento de Biotecnologia, Facultad de Ciencias del Mar y Recursos Biologicos, Universidad de Antofagasta, 601 Avenida Angamos, Antofagasta 1270300, Chile.
Email: michaelw@ums.edu.my

ABSTRACT

Aims: The search for new antibiotics is an ongoing effort and has expanded to pristine niche areas in the Antarctic in recent years due to the emergence of multi-drug resistant pathogens that outpaced the discovery of new antibiotics. We have recently isolated two new actinomycetes strains, INACH3013a and INACH3013b, which displayed antimicrobial properties from soil samples collected from Ardley Island, Antarctica. Hence, an investigation was carried out to identify them and to characterise the antimicrobial compounds produced.
Methodology and results: Strains, INACH3013a and INACH3013b were identified based on their 16S rDNA sequence alignment to those in the GenBank. The results showed that strain INACH3013a was closest to *Streptomyces* spp. while strain INACH3013b was closest to *Streptomyces corallincola* and *Streptomyces bullii*. The extracellular compounds they produced were extracted using various solvents and the extracted compounds were tested against the test pathogens. The dichloromethane extracts from strains, INACH3013a and INACH3013b inhibited mainly Gram-positive pathogens that include *Listeria monocytogenes*, *Staphylococcus aureus*, *Staphylococcus haemolyticus*, *Staphylococcus equorum*, *Bacillus cereus* K3 and *Enterococcus faecalis* while extracts from strain INACH3013b also inhibited a Gram-negative pathogen, *Klebsiella pneumonia* 14x. Predominantly non-polar constituents seem responsible for antibacterial effects, with dichloromethane extracts proving most efficacious, followed by chloroform and ethyl acetate.
Conclusion, significance and impact of study: The research highlights the potential of *Streptomyces* spp. INACH3013a and INACH3013b as a source of potential novel antibiotics. This research explores Antarctic *Streptomyces* strains' antimicrobial capabilities, enabling the potential for the discovery of novel antibiotics and revealing how these compounds may have helped them to compete and survive in nutrient-deficient Antarctic niches.

Keywords: Actinobacteria, antarctic bacteria, antimicrobial compounds, bioactive compounds, *Streptomyces*

INTRODUCTION

In the late 1960s and early 1970s, significant advances in the development of antibiotics to combat pathogenic diseases led to the mistaken belief that the war between infectious diseases and humans was over. However, currently, medical centres are experiencing a rise in antimicrobial resistance of pathogenic bacteria and the susceptibility of pathogens against antibiotics is steadily decreasing.

Antimicrobial resistance (AMR) is a widespread issue affecting clinical practice and public health worldwide. It presents challenges in infection prevention and treatment, making it more expensive and challenging. AMR affects individual patient outcomes and healthcare systems' sustainability. To address this complex issue, a global consensus for coordinated action is needed. The complexity of addressing AMR has led to its classification as a "super-wicked problem." This research aims to explore the consequences of AMR, its impact on clinical practice and the need for a united global response to combat this public health crisis (IUFoST, 2023).

Food poisoning has been caused by the emergence of food-borne microbial pathogens, which pose a serious threat to human health (Haryani *et al.*, 2007). Furthermore, the threat of bioterrorism posed by multidrug-resistant pathogens such as Anthrax and Cholera emphasises the importance of continuing to invest in antimicrobial research (Gilligan, 2002; Holden *et al.*, 2004). Another factor is the downtrend of antibiotic research and development even though there is an increase in pathogen resistance and the fact that no new classes of antibiotics have been developed since 1963. Although two new antibiotics with narrow-spectrum drugs, linezolid and daptomycin, were approved, there have been no new structural classes of antibiotics introduced into the market since 1963, when quinolone, nalidixic acid, were approved (Carpenter and Chambers, 2004;

***Corresponding author**

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Hancock, 2007). After 40 years of research, a new class of antimicrobial agents, oxazolidinones, was introduced in 1999. As a result, new classes of antibiotics with novel mechanisms of action against novel antibacterial targets are required. To slow the development of antibiotic resistance, these antibiotics should be free of cross-resistance to previously existing antibiotics (Labischinski, 2001). Without innovative public policy and additional financial support, there were fewer and fewer antibiotics available to treat the growing number of drug-resistant and dangerous microbes threatening the global community.

Over half a century, interest in glacier microflora has grown due to the discovery of novel bioactive compounds, particularly antibiotics and enzymes, active in low temperatures. This has led to the isolation of new Actinomyces species, such as *Friedmanella antarctica*, *Friedmanella lacustris*, *Nocardiopsis aurantiacus* and *Modestobacterium multiseptatum.* Some studies revealed that a significant proportion of the actinomycete isolates had elevated levels of proteolytic, cellulase and chitinase activity. Additionally, the presence of antibacterial activity was noted (Nedialkova and Naidenova, 2005). This highlights potential avenues for further development in local microflora research.

The global and ongoing search for new antimicrobial agents has met with little success. Hence, the search for antimicrobial agents has now been expanded by scientists to include pristine environments like Antarctica, which is believed to harbour novel microorganisms with novel antimicrobial agents or compounds (Wong *et al.*, 2011). Due to its unique geographical, climatic and environmental characteristics, Antarctica has formed a special habitat of dryness, extreme cold and strong radiation, which has created the novelty and diversity of polar microorganisms. Antarctica is not only an important resource treasure house for discovering new species of microorganisms but also a resource treasure house for discovering new drug lead compounds and other active substances, which makes it a good test material for the study of cryogenic biology and an important potential source of new active substances. Because of their unique originality, novelty and diversity, polar microorganisms are well-equipped with these factors, which provide new exploration ideas for the screening and development of new lead compounds and new drugs.

Since the discovery of penicillin in 1929, more than one hundred clinically used antibiotics, anticancer and antiviral drugs have been developed from microorganisms. The novelty of polar microbial resources includes both metabolite novelty and function novelty. In the future, the emphasis of research should be increased on the production of naturally active drugs by polar microorganisms, to discover new lead compounds with novel structures.

Actinobacteria phyla are abundant in various natural environments, including terrestrial, aquatic and extreme regions, and the most interesting genus is the *Streptomyces*. The ability of the genus *Streptomyces* to synthesise numerous biologically active secondary metabolites, including more than 80% of all known antibiotics, antitumor agents and growth effectors, among other compounds, has drawn a significant amount of interest. As a result, the actinobacterial genus *Streptomyces* is regarded as the main source of useful drugs that cannot be matched by any other organisms (Genilloud, 2017; Newman and Cragg, 2020). The lack of prospects in discovering new antibiotics around many countries has prompted scientists to explore unusual and pristine environments, such as Antarctica. Bruntner *et al.* (2005) have discovered a novel angucyclinone antibiotic, frigocyclinone from Antarctic *Streptomyces greseus*. More recently, Lavin *et al.* (2016) and Astudillo-Barraza *et al.* (2023) reported the isolation of a novel Antarctic *Streptomyces fildesensis.* It has been found to produce compounds with antimicrobial activities as well as antiproliferative activity against human tumour cell lines such as MCF-7, PC-3, HT-29 and non-tumoral colon epithelial cells (CoN) (Astudillo-Barraza *et al.*, 2023). They concluded that those compounds are likely to be novel based on the results of their detailed analysis. Hence, efforts were made in this study to isolate more Antarctic *Streptomyces* or actinobacteria and analyse their bioactive compounds.

MATERIALS AND METHODS

Bacterial isolation

Soil samples were collected aseptically from Ardley Island, Antarctica and stored at -20 °C until use. To isolate bacteria, several media were utilised, including Tryptic Soy Agar (TSA) (Difco, USA), Luria-Bertani agar (Difco, USA), Nutrient agar (NA) (Oxoid, United Kingdom), R2A agar (Difco, USA), GYM Streptomyces medium (glucose 4 g, yeast extract 4 g, malt extract 10 g, $CaCO_3$ 2 g, agar 12 g mix with distilled water 1000 mL and Mueller-Hinton agar (Difco, BD, USA) (Shivaji *et al.*, 1989). One g of the soil sample was dispensed in 9 mL of sterile distilled water. The suspension was then serially diluted to achieve 10-fold and 100-fold dilution factors, and 100 µL of suspension with 100-fold dilution factors was disseminated with a sterile hockey stick on the agar media. The agar plates were incubated at 20 °C for two to ten days. The number of colonies was counted, and their physical traits were noted. Olympus U-CMAD3 T7 light microscopes were used to visualise the selected bacteria.

Antimicrobial compound extraction from the culture medium

The bacteria samples were grown in oat milk broth (2% oat, 1% glycerol mixed with 1000 mL distilled water) and incubated at 28 °C for 3 days. The cell-free media were recovered from the oat milk broth culture by centrifugation at 8,500× *g* for 15 min. It was used for bioactive compound extraction after filtering through 0.2 µm syringe filters. The cell-free media were first extracted with dichloromethane (DCM) (Fisher Scientific, USA); while the remaining aqueous layer was extracted with

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chloroform (ChemAR®, Systerm, Malaysia) and ethyl acetate. Each solvent extraction procedure was performed in a separatory funnel by manually shaking for 20 min. The aqueous layer was re-extracted with the same solvent again (Schneider *et al.*, 2021) The organic phase obtained from the first and second extractions was combined before being subjected to rotary evaporation and lyophilization. The final product was dissolved in 50% ethanol. The fractions obtained were assayed for bacterial inhibition activity.

Disc diffusion test

The National Committee for Clinical Laboratory Standards (CLSI, 2002) recommends the Kirby-Bauer method for antimicrobial susceptibility testing. Mueller-Hinton agar (Difco, BD, USA) medium was used for the disc diffusion test. Prepare media according to the manufacturer's instructions and autoclave. Cool the medium in an oven at 60 °C and pour it into plastic Petri dishes with a diameter of 100 mm. Agar media solidified at room temperature and stored at 4 °C for no more than one week.

Two actinomycete strains isolated from Antarctic soils were used in the antimicrobial activity screening (Table 1). The compounds extracted from the two actinomyces were tested against 14 test pathogens namely, Gram-positive bacteria, *Listeria monocytogenes*, *Staphylococcus aureus*, *Staphylococcus haemolyticus*, *Staphylococcus equorum*, *Bacillus cereus* K3, *Enterococcus faecalis* and Gram-negative bacteria, *Klebsiella pneumonia* 14x. *Salmonella typhimurium* TM13, *Salmonella* Newport, *Salmonella paratyphi* Ty10, *Salmonella braenderup*, *Enterobacter cloaca*e 22x, *Escherichia coli* 0157:H7 were kindly provided Professor Dr. Son Radu, Universiti Putra Malaysia.

A sterile cotton swab was taken out of a portion of the pathogen to be tested and single colonies of respective pathogens were individually suspended in Mueller-Hinton broth media. Each resulting cell suspension was then meticulously transferred and swabbed onto separate Mueller-Hinton agar plates, ensuring uniform and comprehensive coverage of the agar surface. The swabbing procedure was repeated for each pathogen and extract type to ensure consistent distribution. Within 15 min of inoculation, sterile antimicrobial paper discs were gently placed onto the surface of each agar plate. A volume of 10 µL (10 mg/mL) of the specific extracted compound and 50% ethanol as the control was carefully aliquoted onto each paper disc. Subsequently, the agar plates were individually incubated at 37 °C overnight to facilitate the growth and interaction of the respective pathogens with their corresponding compounds.

Genomic DNA extraction

The bacteria were grown in oat milk broth medium (2% oat, 1% glycerol mixed with 1000 mL distilled water) and incubated at 28 °C for 3 days. The cells were harvested for genomic DNA extraction by centrifugation at 8,000× *g* for 5 min. The genomic DNA from the cells was extracted using Qiagen DNeasy Blood and tissue kit according to the manufacturer's instructions. A 1.2% agarose gel was prepared, and 1 µL of the extracted genomic DNAs was loaded into the wells. Gel electrophoresis was performed with a 1 kb ladder (Promega, USA) as the DNA marker and stained ethidium bromide. The quantity and quality of extracted DNA were assessed using a ThermoScientific NanoDrop 2000 UV/Vis spectrophotometer (ThermoScientific, Wilmington, Delaware, USA).

PCR amplification of the bacterial 16S rDNA

The PCR amplification was performed in a 30 µL final reaction volume consisting of 1× GoTaq PCR buffer, 0.2 mM dNTPs, 1.5 mM $MgCl_2$, 10 pmol/µL forward and reverse primers, 1U of Taq polymerase and 1 µL of DNA sample. The 16S rDNA gene amplification was carried out using primers, BSF-8/20 (5'-AGA GTT TGA TCC TGG CTC AG-3') as the forward primer and BSR-1541/20 (5'-AAG GAG GTG ATC CAG CCG CA-3) as the reverse primer. The PCR conditions were pre-denaturation at 95 °C for 5 min, denaturation at 95 °C for 1 min, annealing at 50 °C for 30 sec and extension at 72 °C for 1 min for 30 cycles followed by a final extension at 72 °C for 5 min. The PCR products were analysed on a 1% agarose gel and stained with ethidium bromide. The PCR products were purified according to the manufacturer's instructions by using a Qiagen MinElute ® PCR purification kit (QIAGEN, Germany).

16S rDNA sequencing and analysis

The purified PCR was sequenced using a Sanger ABI3730XL sequencer (Applied Biosystem, USA). The basic local alignment search tool (BLASTn; http://www.ncbi.nlm.nih.gov/blast/Blast.cgi) was used to align the 16s rDNA sequence to those in the NCBI GenBank database. The phylogenetic tree was constructed by aligning sequences to the nearest *Streptomyces* species using the ClustalW programme in the MEGA11 software. The phylogenetic analysis and determination of evolutionary distances were conducted using the maximum-likelihood approach with 1,000 bootstrap replicates. The model used for this analysis was the Tamura-Nei (TN93) with gamma-distribution (G) and accounting for invariant sites (I) in MEGA11 software (http://www.megasoftware.net/index.html).

RESULTS AND DISCUSSION

There are between 80 and 105 colonies each on the TSA, LB, NA, R2A agar and GYM *Streptomyces* agar media. Out of that, only two colonies appeared on the *Streptomyces* agar medium to be actinomyces with the fungus-like branched networks of hyphae morphologies on the GYM *streptomyces* agar medium (Figure 1A and 1C). Their morphologies under the light microscope are shown in Figures 1B and 1D) which indicated that they

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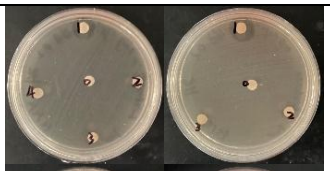
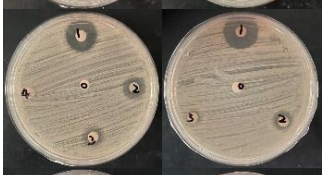
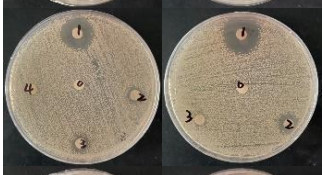
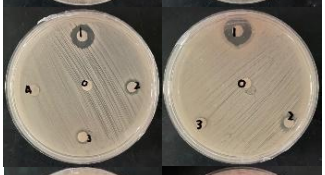
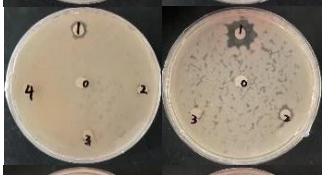
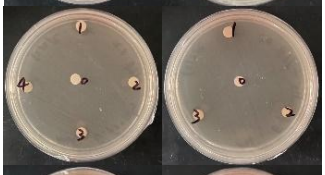
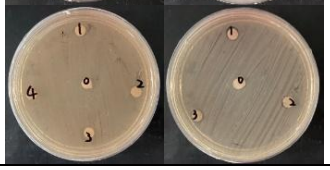
Table 1: The diameter inhibition zones (cm) by compounds extracted from strains INACH3013a and INACH3013b using different solvents ("/" = No inhibition) effectiveness of INACH3013a and INACH3013b extracts against *E. faecalis* and *K. pneumonia* 14x is only marginal.

Test pathogen	Gram property		Image	INACH3013a	INACH3013b
L. monocytogenes	Positive	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		1.2 cm / /	1.4 cm / /
S. haenolyticus	Positive	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		1.7 cm 1.3 cm 1 cm	2 cm 1 cm /
S. aureus	Positive	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		1.8 cm 0.9 cm 0.8 cm	2 cm 1.1 cm /
S. equorum	Positive	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		1.3 cm 0.9 cm 0.8 cm	1.6 cm 0.9 cm 0.7 cm
B. cereus K3	Positive	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		0.9 cm 0.7 cm /	1.6 cm 1 cm /
E. faecalis	Positive	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		0.9 cm / /	0.7 cm / /
K. pneumonia 14x	Negative	1. Dichloromethane 2. Chloroform 3. Ethyl acetate		/ / /	0.8 cm / 0.7 cm

were Gram-positive and had elongated cells that branch into filaments or hyphae. They were further identified based on their 16S rDNA sequence alignment to actinobacteria in the NCBI GenBank database.

The 16S rDNA sequences of strains, INACH3013a and INACH3013b have homology of higher than 98% to a *Streptomyces* sp. and clustered together with several *Streptomyces* spp. (Figure 1). While strain INACH3013a was closest to other *Streptomyces* spp., strain INACH3013b clustered together with *Streptomyces corallincola* and *Streptomyces bullii* (Figure 2) suggesting that strains, INACH3013a and INACH3013b were different species. This is further confirmed by their different colony morphology on the agar plate and under the light microscope (Figure 1A and 1C). The subsequent characteristics of the antimicrobial compounds have also indicated that what they produced differs slightly from each other.

Subsequently, the compounds of strains, INACH3013a and INACH3013b were extracted using various solvents and the results of the inhibitory tests showed that dichloromethane extracts had the broadest antimicrobial compound-containing extracts, followed by chloroform and ethyl acetate (Figure 3 and Table 1). None of the butanol extracts that were recovered had any inhibitory effects on any of the test pathogens (Figure 3)

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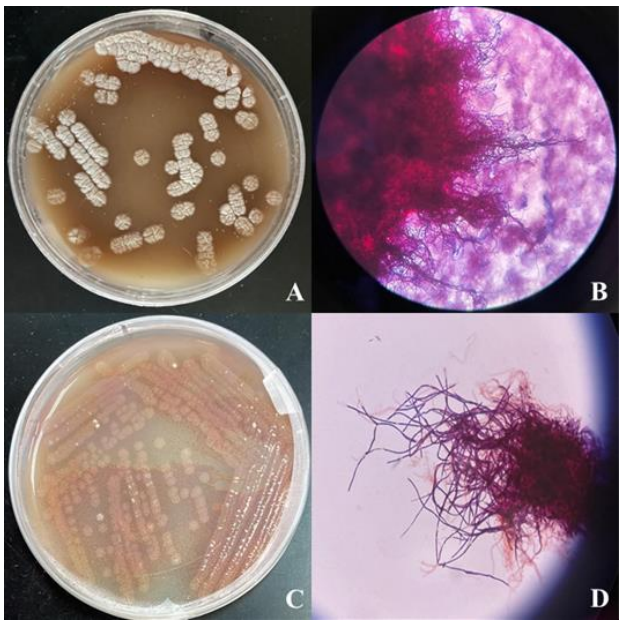

Figure 1: A) INACH3013a colony morphologies on a GYM streptomyces agar medium, B) INACH3013a cell morphology, C) INACH3013b colony morphology on a GYM streptomyces agar medium, D) INACH3013b cell morphologies.

and therefore, not presented in Table 1. It is plausible that the predominant antibacterial constituents are mostly composed of non-polar molecules.

Out of a total of 14 test pathogens, the extracts from INACH3013a and INACH3013b using different solvents inhibited mainly the seven Gram-positive bacteria namely, *L. monocytogenes*, *S. haemolyticus*, *S. aureus*, *S. equorum*, *B. cereus* K3, *E. faecalis* and a Gram-negative bacterium, *K. pneumonia* 14x. The extracts failed to inhibit other test pathogens including *S. typhimurium* TM13, *S. newport*, *S. paratyphi* Ty10, *S. braenderup*, *Enterobacter cloacae* 22x and *E. coli* 0157:H7.

The dichloromethane extracted from strains, INACH3013a and INACH3013b inhibited all the eight pathogens, except INACH3013a extract on *K. pneumonia* 14x. The only Gram-negative, *K. pneumonia* 14x pathogen, was inhibited by compounds extracted from strain INACH3013b but not those from strain INACH3013a. This provides additional evidence that the two strains could be distinct in that certain antimicrobial compounds produced by the strain INACH3013b were not produced by strain INACH3013a. However, we do not rule out the possibility that both strains produce the same compounds but at different concentrations.

In terms of pathogen classification, it is evident that both INACH3013a and INACH3013b strains have significant inhibitory properties against *L. monocytogenes*, *S. haemolyticus*, *S. aureus* and *S. equorum*, with

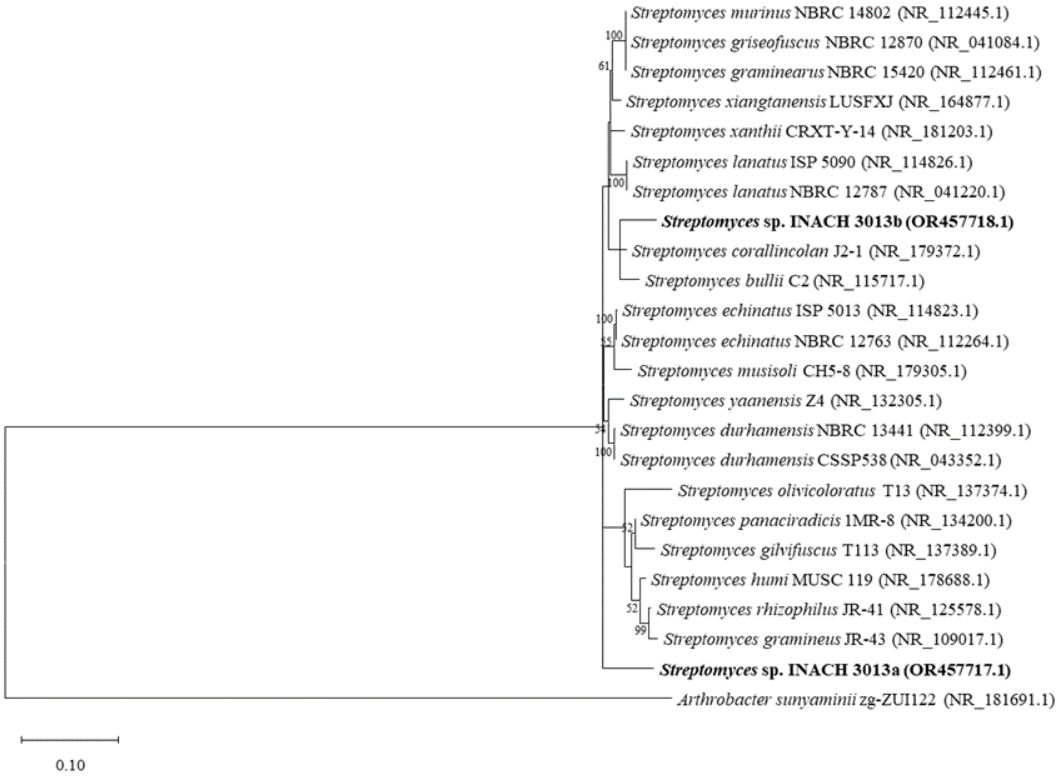

Figure 2: Phylogenetic tree of strains, INACH3013a and INACH3013b constructed based on their 16S rDNA sequences to indicate their relatedness to another *Streptomyces* spp. *Athrobacter sunyaminii* zq-ZUI122 served as the outgroup.

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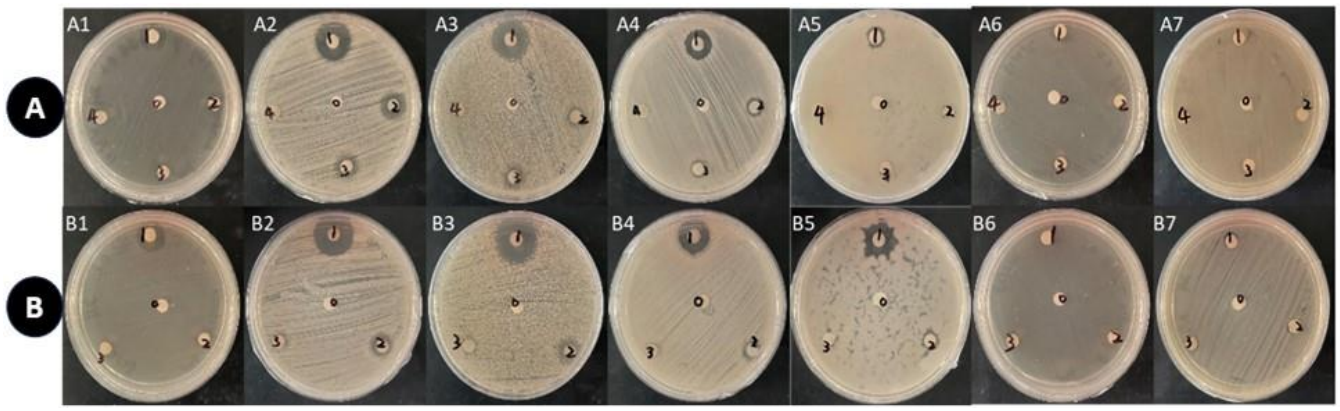

Figure 3: A) INACH3013a extract filtered with a 0.2 µm filter, B) INACH3013b extract filtered with a 0.2 µm filter. The test pathogen in A1 to A7, B1 to B7: *L. monocytogenes, S. haemolyticus, S. aureus, S. equorum, B. cereus* K3*, E. faecalis*, *K. pneumonia* 14x. The extract solvent follows the number inside the plate: 1, Dichloromethane; 2, Chloroform; 3, Ethyl acetate; 4, Butanol; 0, 50% Ethanol (Control). The Petri dish's diameter is 9 cm.

inhibitory zones exceeding 1 cm in diameter. The inhibition range of the compounds recovered using dichloromethane was more than 1.5 cm, particularly for *S. haemolyticus* and *S. aureus*. For *B. cereus* K3, INACH3013b extract still provided a significant inhibitory effect compared to INACH3013a which suppressed slightly less. However, the Actinomycetes represent a promising avenue for the exploration of novel bioactive metabolites, while the likelihood of uncovering new compounds is diminishing due to the intensive investigation of standard species. According to recent research, there is compelling evidence indicating that uncommon actinomycetes, such as those from the less explored Antarctic possess significant promise in the production of novel antibiotics (Lazzarini *et al.*, 2001). According to reports, novel antibiotics with specialised properties have been discovered from actinomycetes residing under unique environmental circumstances (Zakharova *et al.*, 2003).

The antimicrobial compounds of *S. corallincola* and *S. bullii* which are the closest to strain INACH3013b have very limited information in published journals. *S. corallincola* is a new species that was recently isolated from the coral, *Favites pentagona* from Rayong Province, Thailand (Buangrab *et al.*, 2022). Meanwhile, *S. bullii* sp. was isolated from a hyper-arid Atacama Desert soil (Santhanam *et al.*, 2013) and has been shown to produce angucyclinones and anticancer compounds (Kim *et al.*, 2020). Due to the limited information on its closest species, strain INACH3013b serves as a valuable source of antimicrobial compounds of the future. This suggests that the compounds from this strain could be novel and warranted further investigation. Similarly, strain INACH3013a, which does not appear to be related to any specific known *Streptomyces* species, has a significant potential to yield novel antibacterial compounds.

The extracts from strains, INACH3013a and INACH3013b had notable antimicrobial activity against a range of pathogens, even at low doses. The observed phenomenon has a notable inhibitory and bactericidal impact, particularly against Gram-positive bacterial strains. This study establishes a theoretical framework for the potential advancement of novel antibiotics discovery. However, further investigations are required to explore the mechanisms underlying their antibacterial effects so their novelties can be established. On another note, this study found that the ability to produce a broad spectrum of antimicrobial compounds probably gives *Streptomyces* an advantage in competing and surviving in nutrient-deficient habitats in the Antarctic, similar to that of other bacteria reported by Wong *et al.* (2011). In the future, it can be tested whether the two *Streptomyces* are also resistant to antibiotics like those reported by Wong *et al.* (2011) and Tam *et al.* (2015).

CONCLUSION

The study explores the antimicrobial properties of extracts from *Streptomyces* spp., INACH3013a and INACH3013b, derived from Antarctic soil samples. The extracts, which were extracted using dichloromethane, showed significant inhibitory effects against Gram-positive bacterial strains. The highest potency was found in dichloromethane, followed by chloroform and ethyl acetate. The study highlights the potential of *Streptomyces* spp. in developing innovative antibiotics, especially compared to conventional species. Further research is needed to fully explore these strains' capabilities.

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REFERENCES

Astudillo-Barraza, D., Oses, R., Henríquez-Castillo, C., Wong, C. M. V. L., Pérez-Donoso, J. M., Purcarea, C. *et al.* (2023). Apoptotic induction in human cancer cell lines by antimicrobial compounds from Antarctic *Streptomyces fildesensis* (INACH3013). *Fermentation* **9(2), 129.**

Bruntner, C., Binder, T., Pathom-aree, W., Goodfellow, M., Bull, A. T., Potterat, O. *et al.* (2005). Frigocyclinone, a novel angucyclinone antibiotic produced by a *Streptomyces* griseus strain from Antarctica. *The Journal of Antibiotics* **58, 346-349.**

Buangrab, K., Sutthacheep, M., Yeemin, T., Harunari, E., Igarashi, Y., Sripreechasak, P. *et al.* (2022). *Streptomyces corallincola* and *Kineosporia corallincola* sp. nov., two new coral-derived marine actinobacteria. *International Journal of Systematic and Evolutionary Microbiology* **72(2), 005249.**

Carpenter, C. F. and Chambers, H. F. (2004). Daptomycin: Another novel agent for treating infections due to drug-resistant Gram-positive pathogens. *Clinical Infectious Diseases* **38(7), 994-1000.**

CLSI, Clinical and Laboratory Standards Institute. (2002). National Committee for Clinical Laboratory Standards. Performance Standards for Antimicrobial Disc Susceptibility Tests. Clinical and Laboratory Standards Institute, Wayne, PA.

Genilloud, O. (2017). Actinomycetes: Still a source of novel antibiotics. *Natural Product Reports* **34(10), 1203-1232.**

Gilligan, C. A. (2002). An epidemiological framework for disease management. *Advances in Botanical Research* **38, 1-64.**

Hancock, R. E. W. (2007). The end of an era? *Nature Reviews Drug Discovery* **6(1), 28-28.**

Haryani, Y., Noorzaleha, A., Fatimah, A., Noorjahan, B., Patrick, G., Shamsinar, A. *et al.* (2007). Incidence of *Klebsiella pneumonia* in street foods sold in Malaysia and their characterization by antibiotic resistance, plasmid profiling, and RAPD-PCR analysis. *Food Control* **18(7), 847-853.**

Holden, M. T. G., Titball, R. W., Peacock, S. J., Cerdeño-Tárraga, A. M., Atkins, T., Crossman, L. C. *et al.* (2004). Genomic plasticity of the causative agent of melioidosis, *Burkholderia pseudomallei*. *Proceedings of the National Academy of Sciences* **101(39), 14240-14245.**

IUFoST, International Union of Food Science and Technology. (2023). Antimicrobial Resistance: A Food Safety Concern - New Scientific Information Bulletin (SIB) released. IUFoST: https://iufost.org/news/antimicrobial-resistance-food-safety-concern-new-scientific-information-bulletin-sib-released [Retrieved on 1 September 2023].

Kim, J. W., Kwon, Y., Bang, S., Kwon, H. E., Park, S., Lee, Y. *et al.* (2020). Unusual bridged angucyclinones and potent anticancer compounds from *Streptomyces bulli* GJA1. *Organic and Biomolecular Chemistry* **18(41), 8443-8449.**

Labischinski, H. (2001). New antibiotics. *International Journal of Medical Microbiology* **291(5), 317-318.**

Lavin, P. L., Yong, S. T., Wong, C. M. and De Stefano, M. (2016). Isolation and characterization of Antarctic psychrotroph *Streptomyces* sp. strain INACH3013. *Antarctic Science* **28(6), 433-442.**

Lazzarini, A., Cavaletti, L., Toppo, G. and Marinelli, F. (2001). Rare genera of actinomycetes as potential producers of new antibiotics. *Antonie van Leeuwenhoek* **78(3), 399-405.**

Nedialkova, D. and Naidenova, M. (2005). Screening the antimicrobial activity of actinomycetes strains isolated from Antarctica. *Journal of Culture Collections* **4(1), 29-35.**

Newman, D. J. and Cragg, G. M. (2020). Natural products as sources of new drugs over the nearly four decades from 01/1981 to 09/2019. *Journal of Natural Products* **83(3), 770-803.**

Santhanam, R., Rong, X., Huang, Y., Andrews, B. A., Asenjo, J. A. and Goodfellow, M. (2013). *Streptomyces bullii* sp. nov., isolated from a hyper-arid Atacama Desert soil. *Antonie van Leeuwenhoek* **103, 367-373.**

Schneider, Y. K., Jørgensen, S. M., Andersen, J. H. and Hansen, E. H. (2021). Qualitative and quantitative comparison of liquid-liquid phase extraction using ethyl acetate and liquid-solid phase extraction using poly-benzyl-resin for natural products. *Applied Sciences* **11(21), 10241.**

Shivaji, S., Rao, N. S., Saisree, L., Sheth, V., Reddy, G. S. and Bhargava, P. M. (1989). Isolation and identification of *Pseudomonas* spp. from Schirmacher Oasis, Antarctica. *Applied and Environmental Microbiology* **55(3), 767-770.**

Tam, H. K., Wong, C. M. V. L., Yong, S. T., Blamey, J. and González, M. (2015). Multiple antibiotic-resistant bacteria from the maritime Antarctic. *Polar Biology* **38, 1129-1141.**

Wong, C. M. V. L., Tam, H. K., Alias, S. A., González, M., González-Rocha, G. and Domínguez-Yévenes, M. (2011). *Pseudomonas* and *Pedobacter* isolates from King George Island inhibited the growth of foodborne pathogens. *Polish Polar Research* **32(1), 3-14.**

Zakharova, O. S., Zenova, G. M. and Zvyagintsev, D. G. (2003). Some approaches to the selective isolation of actinomycetes of the genus *Actinomadura* from soil. *Microbiology* **72, 110-113.**

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