AUTHORS:
Kojo S. Acquah[1,2]
David W. Gammon[1]
Denzil R. Beukes[3]

AFFILIATIONS:
[1]Department of Chemistry, University of Cape Town, Cape Town, South Africa
[2]Division of Medicinal Chemistry, Department of Pharmaceutical Sciences, University of Connecticut, Storrs, Connecticut, USA
[3]School of Pharmacy, University of the Western Cape, Cape Town, South Africa

CORRESPONDENCE TO:
Denzil Beukes

EMAIL:
dbeukes@uwc.ac.za

DATES:
Received: 06 Feb. 2022
Revised: 30 Aug. 2023
Accepted: 11 Sep. 2023
Published: 30 Jan. 2024

HOW TO CITE:
Acquah KS, Gammon DW, Beukes DR. South African actinobacteria: A treasure trove of novel bioactive metabolites for drug discovery. S Afr J Sci. 2024;120(1/2), Art. #13211. https://doi.org/10.17159/sajs.2024/13211

ARTICLE INCLUDES:
☒ Peer review
☐ Supplementary material

DATA AVAILABILITY:
☐ Open data set
☒ All data included
☐ On request from author(s)
☐ Not available
☐ Not applicable

EDITORS:
Priscilla Baker
Amanda-Lee Manicum

KEYWORDS:
Actinomycetes, filamentous bacteria, microbial biodiversity, natural products, biological activity

FUNDING:
South African National Research Foundation (grant no. 138000)

South African actinobacteria: A treasure trove of novel bioactive metabolites for drug discovery

Although South Africa is known as one of the most biodiverse countries in the world, based on its unique plants and animals, microorganisms have received much less attention. Microorganisms in general and actinobacteria in particular are an underexplored source of new medicines. Recent studies have demonstrated the presence of diverse cultivable actinobacteria from various biomes. However, investigations of the natural product diversity associated with these microorganisms are lacking. We hereby present a review of natural products isolated from South African actinobacteria together with their biological activities. Many of these natural products are structurally novel and include compounds belonging to the following classes: anthraquinones, isoflavonoids, ketolides, macrolides, macrolactams, tripeptides and depsipeptides. They show a wide range of biological activities including antibacterial, antifungal, cytotoxic and antitumour activities.

Significance:

- This review highlights the importance of actinobacteria in the discovery of new medicines and summarises the state-of-the-art on their research in South Africa.
- We reveal a gap in the exploitation of this resource and emphasise the opportunities for multidisciplinary research.

Introduction

Natural products from plants, invertebrates and microorganisms have played an important role in the development of new medicines and agrochemicals.[1] Microbial natural products, in particular, offer significant advantages over natural products produced by macroorganisms. These advantages include a reduced impact on the environment (ecosystems), and hence reduced competition with food crops for arable land, as well as relative ease of production and manipulation of biosynthetic pathways to produce novel compounds for commercial exploitation.

As one of the most biodiverse countries in the world[2], South Africa has a rich tradition of natural products research.[3] However, the main focus of these endeavours has been on plant natural products, and more recently marine natural products[4,5], with much less attention on microbial natural products.

One of the most important sources of medicinally important natural products are the actinobacteria. Actinobacteria, also known as actinomycetes, are filamentous Gram-positive bacteria with high guanine and cytosine (G+C) content in their DNA. The phylum Actinobacteria comprises 20 orders and more than 50 families.[6] This bacterial phylum is widely distributed in terrestrial and both fresh and marine aquatic environments. Some can thrive under extreme conditions such as hyperaridity, high salinity, cold, high pressure, low pH (acidic) and heavy metal contaminated ecosystems.[7] Actinobacteria are either free living, such as soil-dwelling bacteria, or living in association with other organisms, like the plant commensals or those living in and/or on the surfaces of animals like ants, termites and marine invertebrates.[8-10] Some actinobacteria are also plant and animal pathogens[11], while others find use in agriculture, biotechnology, and medicine. In agriculture, they are saprophytic and break down dead plant and animal remains, hastening decomposition.[8] They also aid in nitrogen fixation and are known to produce plant growth promoters, insecticides, herbicides and fungicides.[9] Natural products produced by actinobacteria are structurally diverse and have shown diverse biological activities, including antioxidant, antimalarial, anthelmintic, antifungal, enzyme inhibitory, antibacterial, anticancer, immunosuppressive and cardiovascular properties.[9,12,13] Antibiotics are the largest class of drugs discovered from actinomycetes, as they produce about 70% of all naturally derived antibiotics currently in clinical use.[9] Most of these antibiotics were discovered during the 'golden era' of antibiotic drug discovery and include the aminoglycosides, β-lactams, glycopeptides, macrolides, rifamycins and tetracyclines.[12]

In this review, we discuss the natural products produced by actinobacterial strains isolated from South African environments. The many excellent studies focusing only on the distribution and description of new actinobacteria species, as well as those only reporting on the biological activity of crude extracts and the discovery of enzymes fall outside the scope of this review.

Natural products from South African actinobacteria

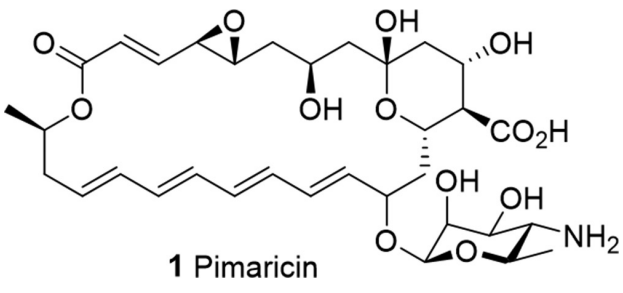
Several novel actinomycete strains have been isolated from South African soils, flora and fauna, in both terrestrial and marine environments, and have been shown to contain bio-active secondary metabolites (Table 1). These strains include species of the ubiquitous *Streptomyces* genus and the less isolated rare genera *Actinomadura, Actinosynnema, Amycolatopsis, Gordonia, Kribbella, Nocardia, Nonomuraea, Rhodococcus, Streptosporangium, Saccharopolyspora* and *Tsukamurella*.[14-22]

Streptomyces

The first report of a South African actinomycete-derived secondary metabolite was a tetraene macrolide natamycin **1** (Figure 1) (also known as pimaricin, natacyn, tennecetin and E235) which was patented in 1955 for its antifungal activity.[23,24] This antibiotic was first purified in 1955 from the extract of the culture broth of *Streptomyces natalensis*,

Table 1: Secondary metabolites from South African actinomycetes and their biological activities. Structures of compounds **1–122** are shown in the text.

Compounds	Name	Structural class	Bioactivity	Producing species	Discovery tool	Reference
1	Natamycin	Tetraene macrolide	Antifungal	*Streptomyces natalensis*		23-32
2–10	Altromycins	New anthraquinone pluramycin type	Antibacterial, antitumour and cytotoxicity	Strain AB 1246E-26	Antibiotic screening	33-37
11–51	Platensimycin, platencin, their analogues and unrelated compounds	New ketolides	Antibacterial	*Streptomyces platensis* strain MA7327	Antisense screening	38-48
52–60	Natalamycin A, reblastatin, geldanamycin and its derivatives	Ansamycin macrolides	Antifungal	*Streptomyces* strain M56		49
61	17-Hydroxycyclooctatin		Cytotoxicity	*Streptomyces* sp. M56	Liquid chromatography–mass spectrometry (LCMS) based	50
62–75	Termisoflavones A–D and other isoflavonoids	Isoflavonoids		*Streptomyces* sp. RB1	Antibiotic screening	51,52
76	1-*O*-(2-aminobenzoyl)-α-L-rhamnopyranoside (ABR)		Cytotoxicity	*Streptomyces* sp. RB1	Bioactivity	53
77–80	Dentigerumycin, Dentigerumycin C–D	Cyclic depsipeptides	Antifungal	*Streptomyces* sp. M41	Bioactivity and LCMS based	54
81–83	Krisynomycins	Cyclic nonribosomal peptides	Antibacterial	*Streptomyces canus* strain CA-091830	Bioactivity and LCMS based	55,56
84–89	Rubterolones A–F	Tropolone alkaloids	Antifungal	*Actinomadura* sp. 5-2	Bioactivity and LCMS based	57
90–92	Natalenamides A–C	Cyclic tripeptides	Cytotoxicity	*Actinomadura* sp. RB99	LCMS based	58
93–107	Polychlorinated and polybrominated analogues of daidzein and genistein	Isoflavonoids	Antibacterial and antifungal	*Actinomadura* sp. RB99	LCMS based	59
108	Fridamycin A	Type II polyketide synthase-derived	Antibiotic and antitumour	*Actinomadura* sp. RB99	LCMS based	60
109–112	Macrotermycins A–D	Glycosylated polyketide macrolactams	Antifungal	*Amycolatopsis* sp. M39	Bioactivity and LCMS based	61
113–122	Speibonoxamine, dehydroxylated desferrioxamine analogues and diketopiperazines	Desferrioxamine		*Kribbella speibonae* strain SK5	Bioactivity and LCMS based	62,63



1 Pimaricin

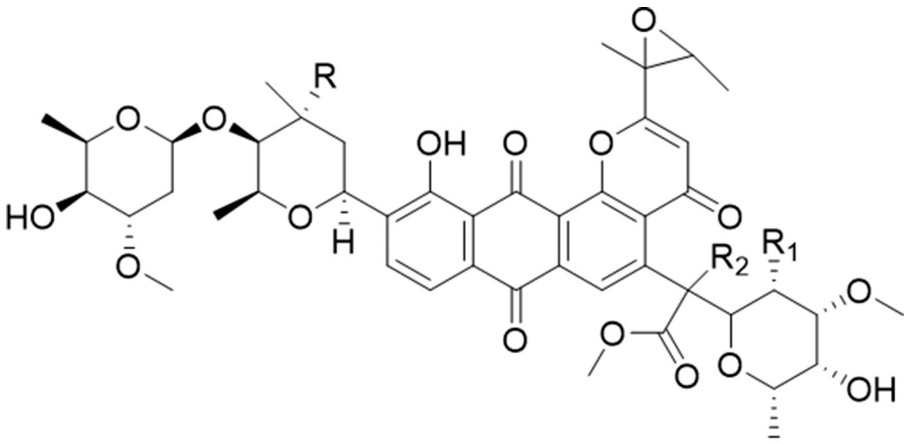
Figure 1: Natamycin **1** from *Streptomyces natalensis*.

isolated from a soil sample collected from Pietermaritzburg (giving rise to its original name, pimaricin) in the KwaZulu-Natal Province, South Africa.[24] Natamycin **1** was later also isolated from *Streptomyces gilvosporeus, Streptomyces chattanogenesis* and *Streptomyces lydicus*.[25-27] It is active against a variety of saprophytic and parasitic fungi and is therefore used commercially as a preservative.[28] It acts by binding to ergosterol in the fungal cell wall, thereby inhibiting fungal growth.[29,30] Natamycin **1** therefore has a wide spectrum of antifungal activity and minimal toxicity to mammalian cells. It also displays in vitro activity against numerous protozoa including *Trypanosoma* and *Acanthamoeba*.[31,32] Clinically, natamycin **1** is used to treat keratitis, and especially that caused by *Aspergillus fumigatus, Candida albicans* and *Acanthamoeba* sp.[28,32] It is also used to treat fungal infections caused by *Cephalosporium, Fusarium* and *Penicillium*, and has shown activity against *Alternaria, Colletotrichum, Curvularia, Lasiodiplodia, Scedosporium* and *Trichophyton*.[28] It is a commercial food additive which has been used for about half a century to prevent fungal growth on foods such as cheese, sausages, yoghurt, fruits, meats, baked confectioneries and beverages.[28]

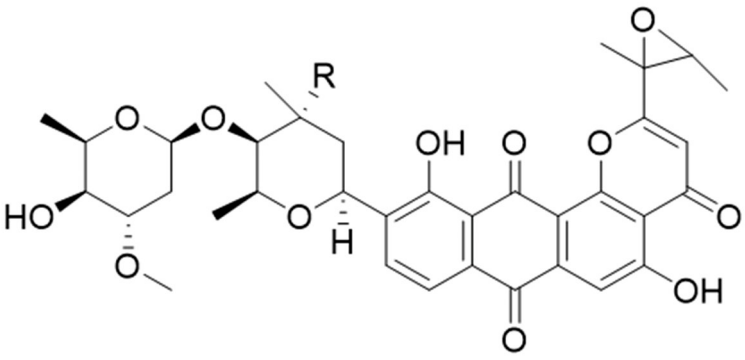
A soil microbe antibiotic screening programme led to the isolation of actinomycete strain AB 1246E-26 from South African bushveld soil.[33] Although the genus of this actinomycete strain was not determined, preliminary characterisation narrowed the taxonomic assignment to either *Nocardia* or the defunct *Micropolyspora*.[33] Strain AB 1246E-26 showed activity against the antibiotic-sensitive strain of *Pseudomonas aeruginosa* K799/61 among other *P. aeruginosa* strains.[33,34] The organic extract of the whole fermentation broth of strain AB 1246E-26 was

subjected to a bioactivity guided isolation protocol to yield the novel anthraquinone-derived class of antibiotics called altromycins.[34,35] The altromycins A-I **2–10** (Figure 2) are nine closely related members of the pluramycin class of compounds with a single epoxide substituent, an amino-disaccharide and/or a 6-deoxy-3-O-methylaltrose attached to the conjugated ring systems of an anthraquinone-γ-pyrone core.[34-36] Altromycin B **3** was screened against 30 bacterial strains including *Staphylococcus aureus, Staphylococcus epidermidis, Micrococcus luteus, Enterococcus hirae, Streptococcus bovis, Streptococcus agalactiae, Streptococcus pyogenes, Escherichia coli, Enterobacter aerogenes, Klebsiella pneumoniae, Providencia stuartii, Pseudomonas aeruginosa, Pseudomonas cepacia* and *Acinetobacter* sp. clinical isolates.[33] Altromycin B **3** exhibited potent antibacterial activity against the clinical isolates of *Staphylococcus* and *Streptococcus* with a minimum inhibitory concentration (MIC) range of 0.39–3.12 μg/mL and 0.2–3.12 μg/mL, respectively, but displayed moderate to weak antibacterial activity against Gram-negative bacteria with an MIC range of 25 to >100 μg/mL.[33] The altromycins also showed cytotoxic activity against various cancer and tumour cell lines including cervical cancer (HeLa), human lung cancer (A549), colon tumour (HCT-8), murine leukemia cell (P388) lines and ovarian sarcoma (M5076).[37]

In their search for antibiotics that inhibit fatty acid biosynthesis in bacteria, researchers at Merck discovered the novel broad-spectrum antibiotic platensimycin **11** (Figure 3) from *Streptomyces platensis* strain MA7327, which was originally isolated from a soil sample collected in the Eastern Cape Province of South Africa.[38] An organic extract of the fermentation broth of *S. platensis* strain MA7327 was subjected to a unique antisense differential sensitivity whole-cell two-plate agar diffusion bioassay-guided fractionation process to yield platensimycin **11**.[38] Platensimycin **11** consists of a 3-amino-2,4-dihydroxybenzoic acid tethered via an amide bond to a C-17 tetracyclic enone which includes a bridge-head oxygen.[38,39] Another closely related compound, platencin **12** (Figure 3), was produced by *Streptomyces platensis* strain MA7339 using the same bioassay-guided fractionation procedure, although its biosynthetic gene cluster was also identified in strain MA7327.[40] Several other analogues **13–49** (Figure 3) – with modifications on or loss of the aromatic ring, modifications on the terpenoid and anilide moieties and a change in the length of the enone acid portion of platensimycin and platencin – have also been isolated from strain MA7327.[41-48] Compounds **50** and **51** (Figure 3), which are structurally different from the platensimycin group of compounds, were also isolated from strain MA7327.[41,42] Furthermore, other glycosylated analogues of platensimycin **11** and platencin **12** have



2 R = NHMe, R_1 = R_2 = OH, Altromycin A
3 R = $N(Me)_2$, R_1 = R_2 = OH, Altromycin B
4 R = NHMe, R_1 = H, R_2 = OH, Altromycin C
5 R = $N(Me)_2$, R_1 = H, R_2 = OH, Altromycin D
6 R = NHMe, R_1 = OH, R_2 = H, Altromycin E
7 R = $N(Me)_2$, R_1 = OH, R_2 = H, Altromycin F
8 R = NH_2, R_1 = OH, R_2 = OH, Altromycin G



9 R = NHMe, Altromycin H
10 R = $N(Me)_2$, Altromycin I

Figure 2: Altromycins A-I **2–10** from South African actinomycete strain AB 1246E-26.

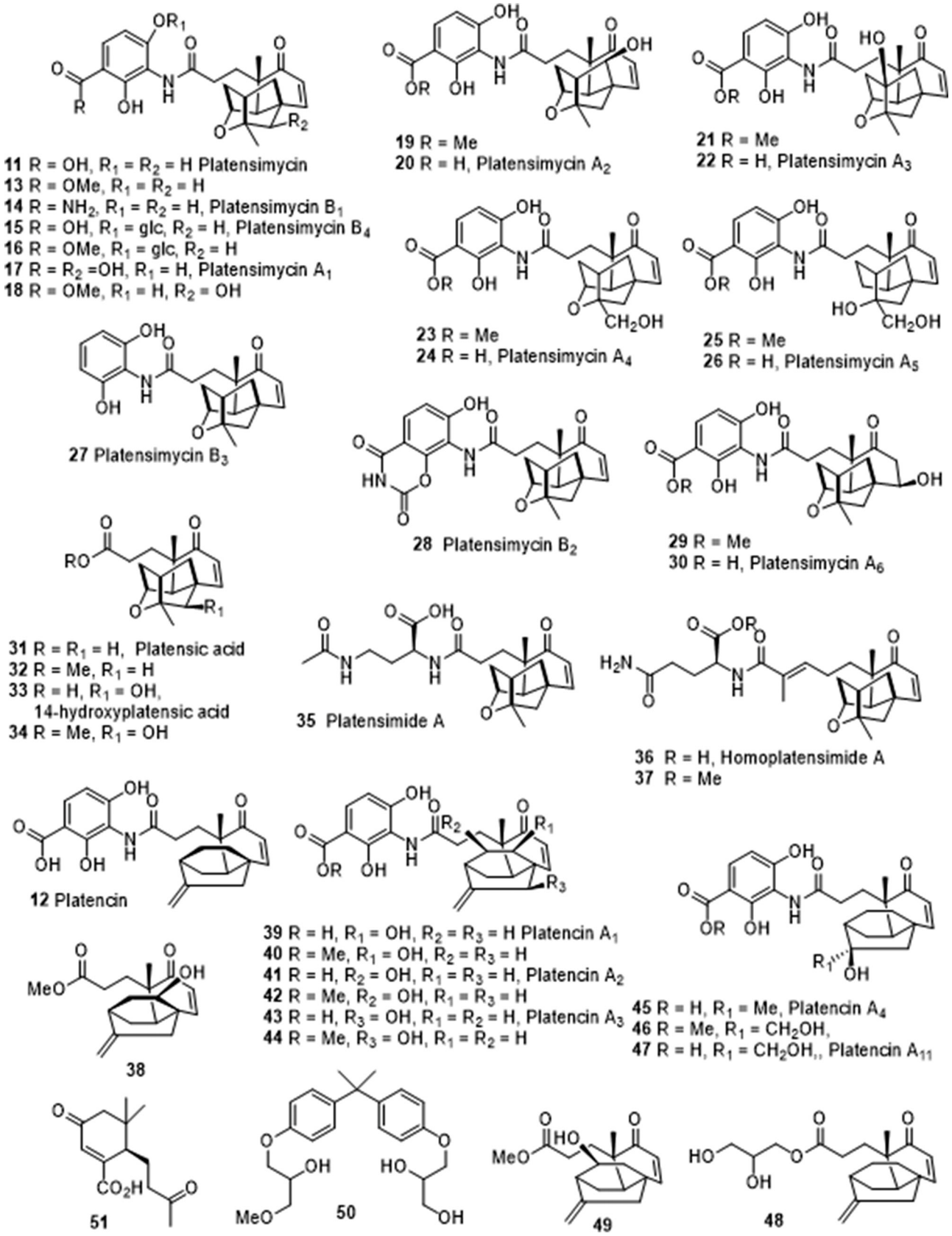


Figure 3: Secondary metabolites **11**–**51** from South African actinomycetes *Streptomyces platensis*.

been produced by an engineered mutant strain *S. platensis* SB12600.[40] Platensimycin **11** selectively inhibits the elongation-condensing enzyme FabF of the bacterial fatty acid synthesis pathway, while platencin **12** equally inhibits both the initiation condensing (FabH) and elongation (FabF) enzymes.[39] Platensimycin, platencin and their analogues have shown potent in vitro activity against both cell-free and whole-cell systems including methicillin-resistant *Staphylococcus aureus*, vancomycin-resistant *Enterococcus* and *Mycobacterium tuberculosis*.[39]

Although the isolated analogues and synthesised ones did not exhibit improved activity compared with compounds **11** and **12**, they provide important structure–activity relationship information to determine the pharmacophore of **11** and **12**.

In their quest to isolate biologically active metabolites from termite-associated actinobacteria, Kim et al. discovered that *Streptomyces* strain M56, isolated from the fungal comb of a South African *Macrotermes natalensis* Mn802 colony, exhibited potent broad-spectrum antifungal activity.[49] Bioassay-guided isolation resulted in the purification of the novel fused bicyclic ansa macrolide natalamycin A **52** (Figure 4) alongside other ansa macrolides including reblastatin **53**, geldanamycin **54** and its derivates, 17-O-demethyl-geldanamycin **55**, 19-S-methylgeldanamycin **56**, 17-amino-17-demethoxy-geldanamycin **57**, methyl geldanamycinate derivative **58**, 17-amino-17-demethoxy-methyl geldanamycinate **59** and 19-[(1′S,4′R)-4′-hydroxy-1′-methoxy-2′-oxopentyl]geldanamycin **60**.[49] Although the fractions that yielded the isolated compounds showed strong antifungal activity against some strains, the natalamycins exhibited weak or no activity against *Saccharomyces cerevisiae* and other fungal isolates.[49] An untargeted dereplication of the liquid chromatography–mass spectrometry (LCMS) data of the methanol extract of *Streptomyces* strain M56 signified the presence of new metabolites.[50] The methanol extract of a large-scale culture of strain M56 was subjected to several chromatographic methods to yield the fused 5-8-5 tricyclic diterpene 17-hydroxycyclooctatin **61** (Figure 4).[50] Compound **61** is a potential ERα antagonist and exhibited weak cytotoxicity activity against MCF-7 human breast cancer cell lines with an IC_{50} value of 566.95 ± 0.48 μM.[50]

Further studies on the metabolites of the actinobacteria associated with the fungus-growing South African termite *M. natalensis* led to the isolation of *Streptomyces* strain RB1 which exhibited antibacterial activity against *Staphylococcus aureus* and *Candida albicans*.[51] Fractionation of the methanol (MeOH) extract of strain RB1 yielded the new isoflavonoid glycosides, termisoflavone A-C **62–64.** and other isoflavonoids **66–70**, **72–74** (Figure 5).[51] The isolated compounds showed no antifungal or antibacterial activity when screened against *C. albicans, C. neoformans, S. aureus*, and *E. coli,* but compounds **69** and **73** ameliorated cisplatin-induced kidney cell damage.[51] Further investigation of the MeOH extract of strain RB1 using LCMS- and NMR-based dereplication strategies led to the identification and subsequent isolation of another new isoflavonoid glycoside, termisoflavone D **65**, together with the known isoflavonoids **66**, **67**, **69**, **71–73** and **75** (Figure 5).[52] Isoflavonoid **69** displayed activity against glutamate-induced HT22 cells by preventing accumulation of intracellular reactive oxygen species.[52] Another study exploring the termite associated actinobacteria for reno- and kidney-protective drug discovery found that the MeOH extract of *Streptomyces* sp. RB1 exhibited a protective effect against cisplatin-induced cytotoxicity.[53] A bioassay (LLC-PK1 cells)-guided isolation process yielded the renoprotective 1-O-(2-aminobenzoyl)-α-L-rhamnopyranoside (ABR) **76** (Figure 5).[53]

Analysing the chemical and metabolomic profiles of actinobacteria derived from termite nests with an unbiased high-throughput high-performance liquid chromatography–high-resolution mass spectrometry based dereplication strategy revealed that *Streptomyces* sp. M41 isolated from the South African termite *M. natalensis* produces new complex nonribosomal peptide polyketide synthase (NRPS/PKS) hybrid compounds.[54] Chromatographic purification of a large-scale culture of strain M41 interestingly yielded new analogues (two linear **77, 78** and one cyclic **79**) of the cyclic depsipeptide dentigerumycin **80** (Figure 6).[54]

The South African soil actinomycete, *Streptomyces canus* strain CA-091830, is a producer of the cyclic depsipeptides krisynomycins A–C **81–83** (Figure 7).[55,56] Krisynomycin A was initially isolated based on a screening project with the aim of identifying and isolating imipenem potentiators against methicillin-resistant *Staphylococcus aureus* (MRSA).[55] Further investigation led to the isolation of Krisynomycin B and C, which are chlorinated analogues of Krisynomycin A.[56] Although compounds **81–83** showed weak activity against MRSA, the activity was improved when they were tested in combination with sub-lethal concentrations of imipenem.[56]

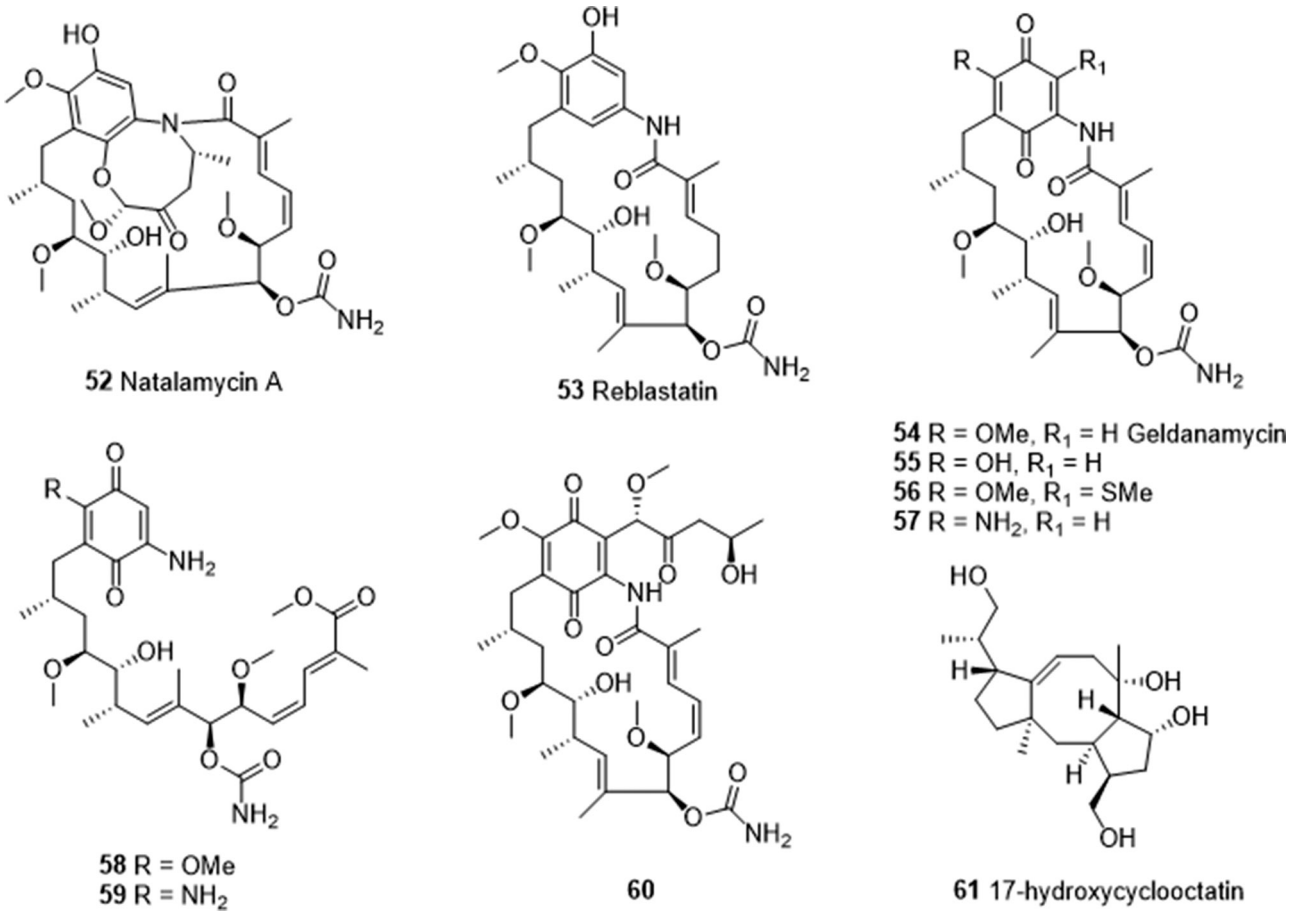


Figure 4: Secondary metabolites **52–61** from South African actinomycetes *Streptomyces* strain M56.

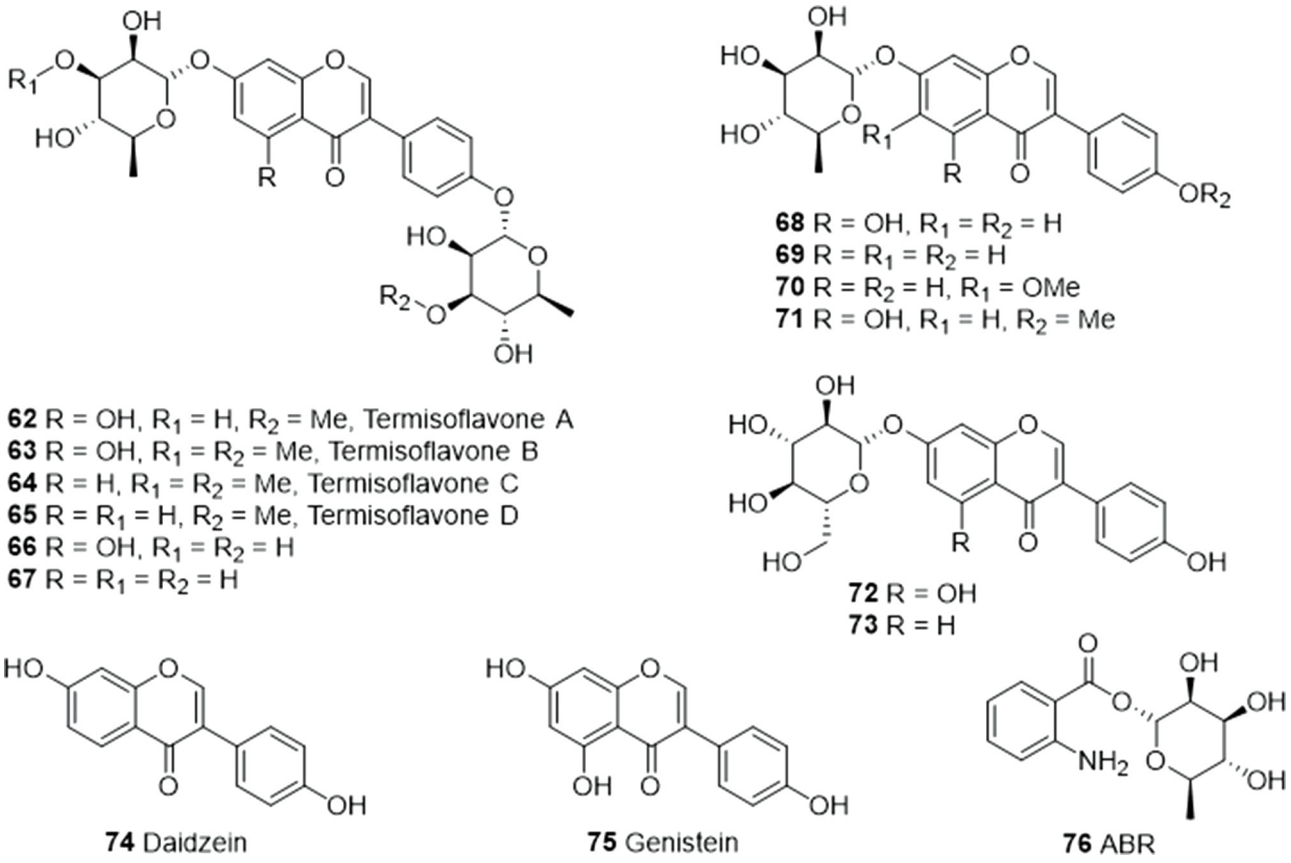


Figure 5: Secondary metabolites **62–76** from South African actinomycetes *Streptomyces* strain RB1.

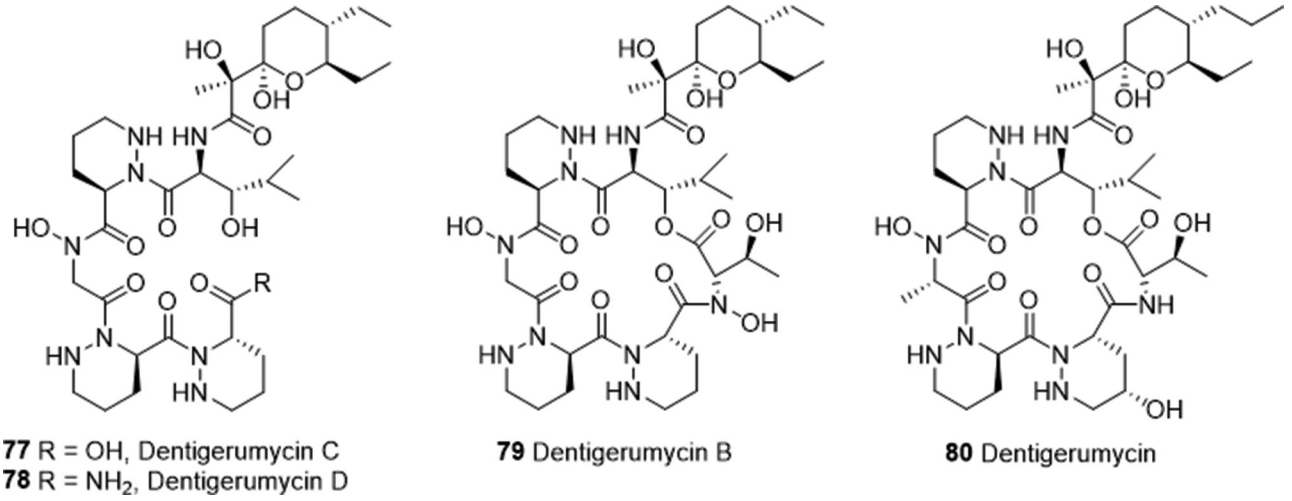


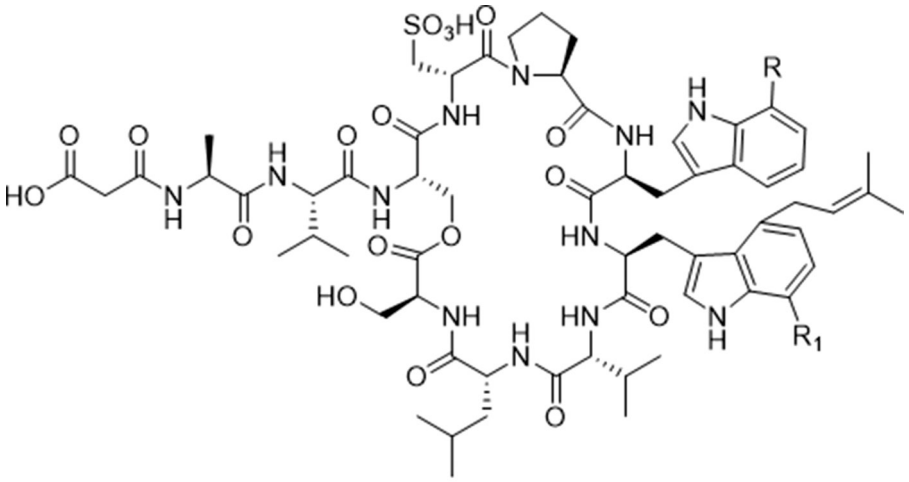
Figure 6: Dentigerumycins A–D **77–80** from South African actinomycetes *Streptomyces* sp. M41.

Rare actinomycete strains

The rare actinomycete *Actinomadura* sp. 5-2, which was recovered from the gut of the fungus-growing termite *M. natalensis*, produced novel, highly substituted tropolone alkaloids, rubterolones A–F **84–89** (Figure 8).[57] These compounds were detected by both bioactivity and high-resolution mass spectrometry based dereplication techniques, and subsequently isolated from the organic extract of a culture of strain 5-2.[57] Curiously, compounds **84–89** did not show any significant antifungal activity.[57]

The crude extract of another *Actinomadura* isolate, strain RB99, isolated from the surface of the termite *M. natalensis*, was analysed by liquid chromatography (LC)/ultraviolet (UV)/mass spectrometry (MS) and shown to produce new compounds.[58] A spectrometry guided isolation led to the discovery of three new cyclic tripeptides named natalenamides A–C **90–92** (Figure 9).[58] The isolated compounds exhibited weak cytotoxicity when screened against HepG2 and HeLa/A549 cells.[58] Compound **92** showed significant activity against IBMX-mediated melanin synthesis in a dose-dependent manner.[58]

Analyses of the high-resolution tandem mass spectrometry (HR-MS^2) data of the MeOH extract of strain RB99 and further exploration of the HR-MS^2 data on the Global Natural Product Social (GNPS) molecular networking platform showed that strain RB99 produces polyhalogenated isoflavonoids.[59] Seoung et al. proved that *Actinomadura* sp. RB99 can bio-transform the plant-based daidzein **74** and genistein **75** isoflavonoids contained in the ISP2 growth medium to polyhalogenated derivatives.[59] Optimisation of the growth



81 R = R_1 =H, Krisynomycin A
82 R = Cl, R_1 = H, Krisynomycin B
83 R = R_1 = Cl, Krisynomycin C

Figure 7: Krisynomycins **81–83** from South African actinomycetes *Streptomyces canus* strain CA-091830.

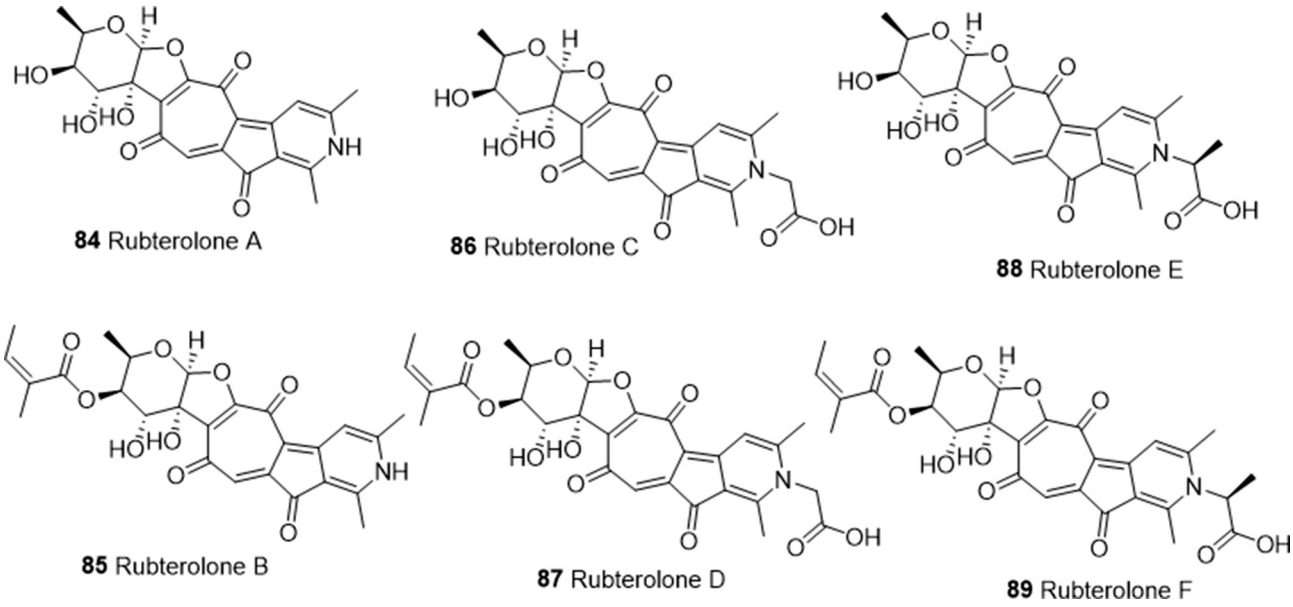


Figure 8: Rubterolones A–F (**84–89**) from South African actinomycetes *Actinomadura* sp. 5-2.

medium (ISP2 augmented with NaCl or KBr) led to the production and subsequent MS-guided purification of eight polychlorinated analogues **93–100** (Figure 9), of which six were new, and seven novel polybrominated analogues **101–107** (Figure 9), of daidzein and genistein.[59] The isolated chlorinated analogues did not exhibit any antibacterial or antifungal activities against *E. coli, S. aureus, S. epidermidis* and *C. albicans,* but the brominated analogues **101** and **105** were active against *Helicobacter pylori*.[59] Additional analysis of the LCMS data of the MeOH extract of strain RB99 led to the detection and isolation of the antibiotic and antitumour agent fridamycin A **108** (Figure 9), which is a type II polyketide.[60] Fridamycin A **108** showed good antidiabetic properties in 3T3-L1 adipocytes and could serve as a promising lead for type 2 diabetes drug discovery.[60]

Metabolomic and bioactivity profiling of termite-associated actinomycetes led to the detection and subsequent isolation of four new 20-membered glycosylated polyketide macrolactams, named macrotermycins A–D **109–112** (Figure 10), from the organic extract of the rare actinomycete *Amycolatopsis* sp. M39.[61] Strain M39 was also isolated from the termite *M. natalensis* and its organic crude extract exhibited a unique metabolomic profile and was active against the termite fungal garden competitor *Pseudoxylaria* spp.[61] Only compounds **109** and **111** were active against *Pseudoxylaria* sp.[61]

A rare actinomycete, *Kribbella speibonae* strain SK5, isolated from a soil sample collected from Stellenbosch in the Western Cape Province of South Africa, displayed strong antimycobacterial activity against *Mycobacterium aurum* strain A+.[62] Chemical and metabolomic profiling of an organic extract of a liquid culture of this strain showed that it is a prolific producer of hydroxamate siderophores, including new dehydroxylated desferrioxamine analogues and diketopiperazines (DKP).[63] Two new dehydroxylated desferrioxamines, speibonoxamine **113** and desoxy-desferrioxamine D1 **114** (Figure 11), alongside already reported desferrioxamines **115–118** and a DKP **119** (Figure 11), were subsequently isolated from an organic extract of a liquid culture of strain SK5.[63] The plausible structures of three new dehydroxylated analogues **120–122** were determined by the GNPS molecular network and MS/MS fragmentation analyses.[63]

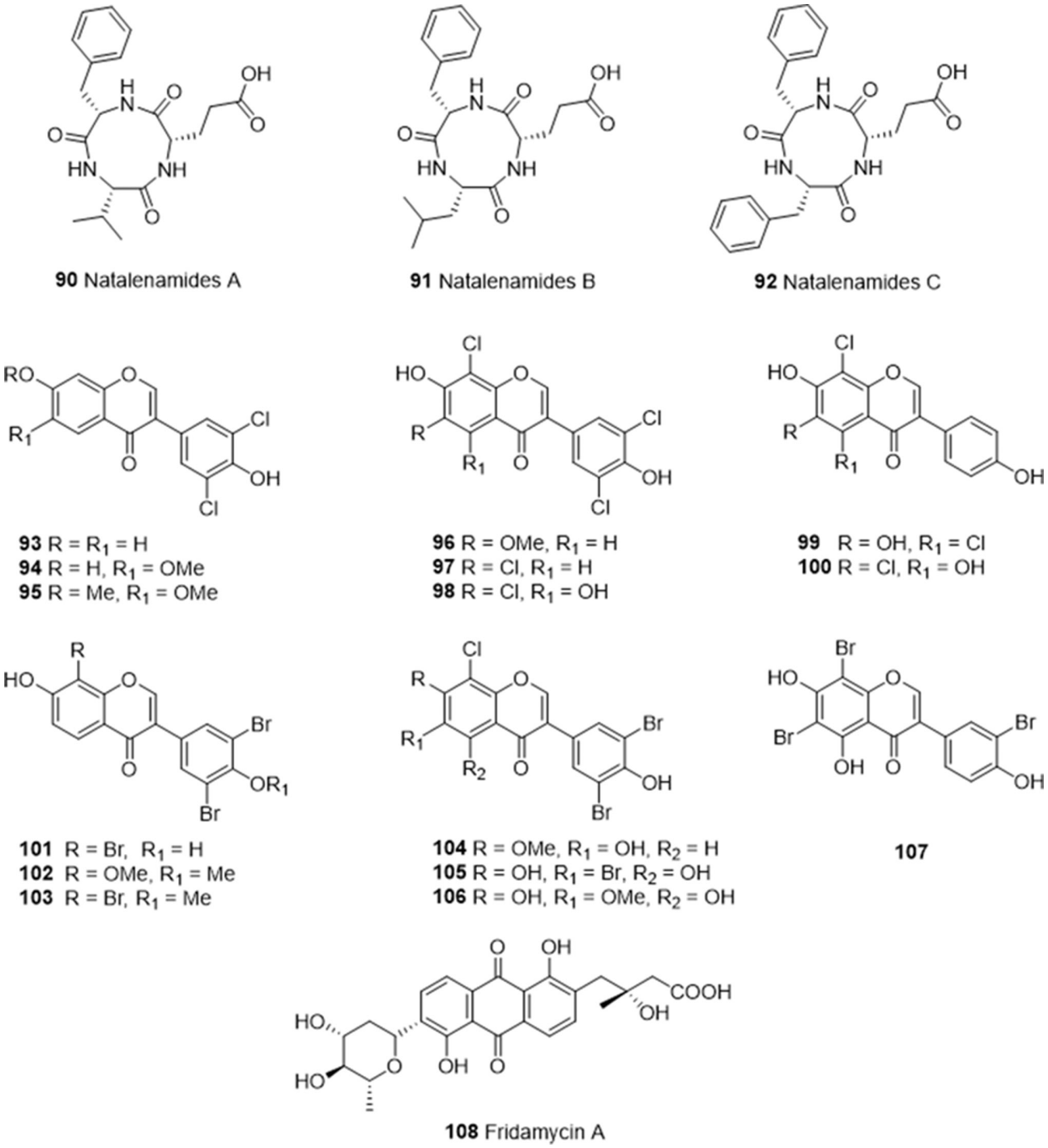


Figure 9: Secondary metabolites from South African actinomycetes **90–108.**

Conclusions and future prospects

Biodiversity is "more than just legs and leaves"[64] and South Africa's microbial biodiversity presents a tremendous opportunity for the natural products chemist and those interested in drug discovery. In this review, we have described 122 compounds and shown that South African actinobacteria are prolific producers of novel, bioactive metabolites. Interestingly, the first compound described from a South African actinomycete, natamycin, is also the only one that has made it to the clinic. Other compounds, such as platensimycin and geldanamycin, have shown promise but either lack efficacy in humans or showed toxic side effects which prevented their development as drugs. South African researchers interested in natural products based drug discovery face the same challenges as elsewhere in the world. These challenges include the significant cost of drug development, re-isolation of previously reported compounds, and lack of interest in natural products for drug development. Nevertheless, the compounds reviewed here present only the tip of the iceberg and many more species remain to be discovered and studied for natural product production. Furthermore, with innovations and technological advancement in purification, structure elucidation, chemical biology, genome sequencing and mining, dereplication, and bioinformatic, cheminformatic and metabolomic tools like the GNPS molecular networking, microbial natural product drug discovery in South Africa shows great potential. It is worth mentioning that, apart from our research on the chemistry of the metabolites of the South African rare actinomycete *Kribbella speibonae* strain SK5, all the research on South African actinomycetes reported here was done by research

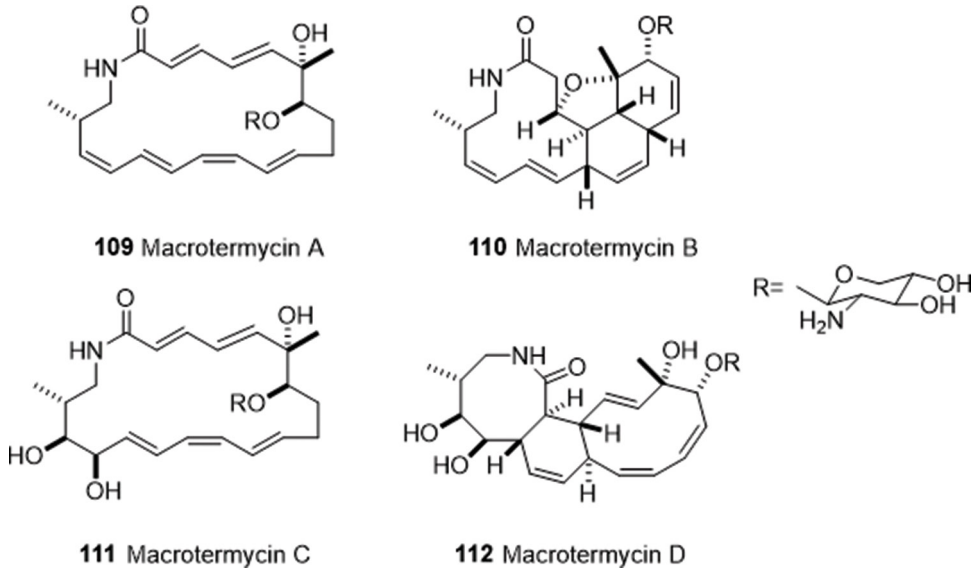


Figure 10: Macrotermycins A–D **109–112** from South African actinomycetes *Amycolatopsis* sp. M39.

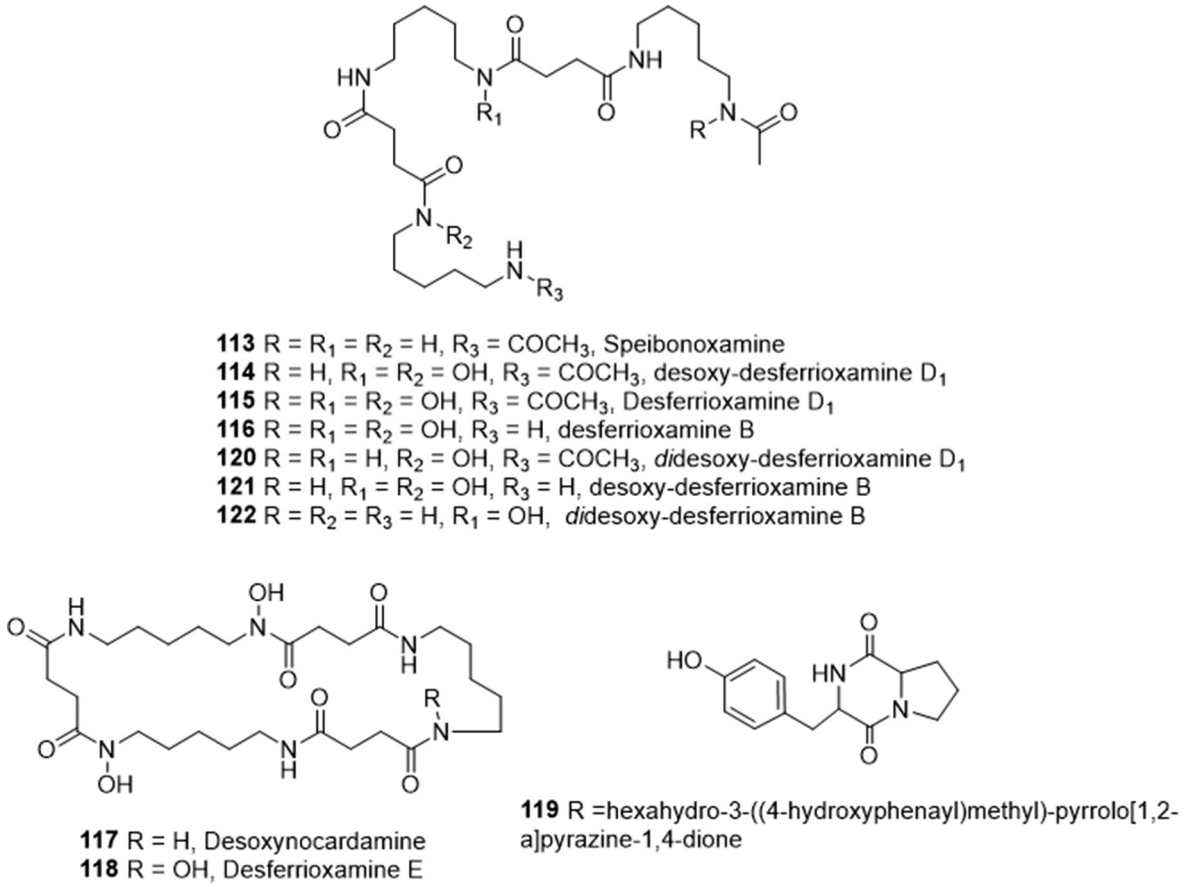


Figure 11: Secondary metabolites **113–122** from the South African *Kribbella speibonae* strain SK5.

groups based outside of South Africa. This represents a challenge and an opportunity for closer collaboration between South African researchers (microbiologists, pharmacologists and chemists) in order to fully explore the opportunities presented by South African microbial biodiversity.

Acknowledgements

We gratefully acknowledge financial support from the National Research Foundation of South Africa (grant no. 138000).

Competing interests

We have no competing interests to declare.

Authors' contributions

K.S.A: Conceptualisation; data collection; writing – the initial draft. D.W.G: Conceptualisation; student supervision; writing – revisions. D.R.B: Conceptualisation; data collection; student supervision; writing – revisions.

References

1. Newman DJ, Cragg GM. Natural products as sources of new drugs over the nearly four decades from 01/1981 to 09/2019. J Nat Prod. 2020;83(3):770–803. https://doi.org/10.1021/acs.jnatprod.9b01285
2. UN Environment Programme World Conservation Monitoring Centre (UNEP-WCMC). Biodiversity A-Z. Cambridge, UK: UNEP-WCMC; 2014. Available from: https://www.biodiversitya-z.org/content/megadiverse-countries
3. Drewes SE. Natural products research in South Africa: 1890–2010. S Afr J Sci. 2012;108(5/6), Art. #765. https://doi.org/10.4102/sajs.v108i5/6.765
4. Davies-Coleman MT. Natural products research in South Africa: End of an era on land or the beginning of an endless opportunity in the sea? S Afr J Chem. 2010;63:105–113.
5. Davies-Coleman MT, Veale CG. Recent advances in drug discovery from South African marine invertebrates. Mar Drugs. 2015;13(10):6366–6383. https://doi.org/10.3390/md13106366
6. Parte AC, Sardá Carbasse J, Meier-Kolthoff JP, Reimer LC, Göker M. List of prokaryotic names with standing in nomenclature (LPSN) moves to the DSMZ. Int J Syst Evol Microbiol. 2020;70:5607–5612. https://doi.org/10.1099/ijsem.0.004332
7. Merino N, Aronson HS, Bojanova DP, Feyhl-Buska J, Wong ML, Zhang S, et al. Living at the extremes: Extremophiles and the limits of life in a planetary context. Front Microbiol. 2019;10, Art. #780. https://doi.org/10.3389/fmicb.2019.00780
8. Mayfield CI, Williams ST, Ruddick SM, Hatfield HL. Studies on the ecology of actinomycetes in soil IV. Observations on the form and growth of *Streptomycetes* in soil. Soil Biol Biochem. 1972;4(1):79–91. https://doi.org/10.1016/0038-0717(72)90045-4
9. Barka EA, Vatsa P, Sanchez L, Gaveau-Vaillant N, Jacquard C, Klenk HP, et al. Taxonomy, physiology, and natural products of Actinobacteria. Microbiol Mol Biol Rev. 2016;80(1):143. https://doi.org/10.1128/MMBR.00019-15
10. Benndorf R, Martin K, Küfner M, de Beer ZW, Vollmers J, Kaster AK, et al. *Actinomadura rubteroloni* sp. nov. and *Actinomadura macrotermitis* sp. nov., isolated from the gut of the fungus growing-termite *Macrotermes natalensis*. Int J Syst Evol. 2020;70(10):5255–5262. https://doi.org/10.1099/ijsem.0.004403
11. Salwan R, Sharma V. The role of actinobacteria in the production of industrial enzymes. In: Singh BP, Gupta VK, Passari AK, editors. New and future developments in microbial biotechnology and Bioengineering. Amsterdam: Elsevier; 2018. p. 165–177. https://doi.org/10.1016/B978-0-444-63994-3.00011-4
12. Genilloud O. Actinomycetes: Still a source of novel antibiotics. Nat Prod Rep. 2017;34(10):1203–1232. https://doi.org/10.1039/C7NP00026J
13. Taechowisan T, Chaisaeng S, Phutdhawong WS. Antibacterial, antioxidant and anticancer activities of biphenyls from *Streptomyces* sp. BO-07: An endophyte in *Boesenbergia rotunda* (L.) Mansf A. Food Agric Immunol. 2017;28(6):1330–1346. https://doi.org/10.1080/09540105.2017.1339669
14. Rohland J, Meyers PR. *Streptomyces fractus* sp. nov., a novel streptomycete isolated from the gut of a South African termite. Antonie van Leeuwenhoek. 2015;107(5):1127–1134. https://doi.org/10.1007/s10482-015-0404-8
15. Cook AE, Le Roes M, Meyers PR. *Actinomadura napierensis* sp. nov., isolated from soil in South Africa. Int J Syst Evol. 2005;55(2):703–706. https://doi.org/10.1099/ijs.0.63359-0
16. Everest GJ, Le Roes-Hill M, Rohland J, Enslin S, Meyers PR. *Amycolatopsis roodepoortensis* sp. nov. and *Amycolatopsis speibonae* sp. nov.: Antibiotic-producing actinobacteria isolated from South African soils. J Antibiot. 2014;67(12):813–818. https://doi.org/10.1038/ja.2014.79
17. Everest GJ, Meyers PR. *Kribbella hippodromi* sp. nov., isolated from soil from a racecourse in South Africa. Int J Syst Evol Microbiol. 2008;58(2):443–446. https://doi.org/10.1099/ijs.0.65278-0
18. Le Roes M, Goodwin CM, Meyers PR. *Gordonia lacunae* sp. nov., isolated from an estuary. Syst Appl Microbiol. 2008;31(1):17–23. https://doi.org/10.1016/j.syapm.2007.10.001
19. Benndorf R, Schwitalla JW, Martin K, De Beer ZW, Vollmers J, Kaster AK, et al. *Nocardia macrotermitis* sp. nov. and *Nocardia aurantia* sp. nov., isolated from the gut of the fungus-growing termite *Macrotermes natalensis*. Int J Syst Evol Microbiol. 2020;70(10):5226–5234. https://doi.org/10.1099/ijsem.0.004398
20. Le Roes M, Meyers PR. *Nonomuraea candida* sp. nov., a new species from South African soil. Antonie van Leeuwenhoek. 2008;93(1):133–139. https://doi.org/10.1007/s10482-007-9187-x
21. Sibanda T, Mabinya LV, Mazomba N, Akinpelu DA, Bernard K, Olaniran AO, et al. Antibiotic producing potentials of three freshwater actinomycetes isolated from the Eastern Cape Province of South Africa. Int J Mol Sci. 2010;11(7):2612–2623. https://doi.org/10.3390/ijms11072612
22. Le Roes-Hill M, Durrell K, Prins A, Meyers PR. *Streptosporangium minutum* sp. nov., isolated from garden soil exposed to microwave radiation. J Antibiot. 2018;71(6):564–574. https://doi.org/10.1038/s41429-018-0036-0
23. Struyk AP, Waisvisz JM. (Gist-Brocades, Delft): Pimaricin and process of producing same. US-Patent. 1955;3:892.
24. Struyk AP, Drost G, Haisvisz JM, Van Eek T, Hoogerheide JC. Pimaricin, a new antifungal antibiotic. Antibiot Annu. 1957/1958;5:878–885.
25. Chen GQ, Lu FP, Du LX. Natamycin production by *Streptomyces gilvosporeus* based on statistical optimization. J Agric Food Chem. 2008;56(13):5057–5061. https://doi.org/10.1021/jf800479u
26. Du YL, Chen SF, Cheng LY, Shen XL, Tian Y, Li YQ. Identification of a novel *Streptomyces chattanoogensis* L10 and enhancing its natamycin production by overexpressing positive regulator ScnRII. J Microbiol. 2009;47(4):506–513. https://doi.org/10.1007/s12275-009-0014-0
27. Lu CG, Liu WC, Qiu JY, Wang HM, Liu T, Liu DW. Identification of an antifungal metabolite produced by a potential biocontrol actinomyces strain A01. Braz J Microbiol. 2008;39(4):701–707. https://doi.org/10.1590/S1517-83822008000400020
28. Aparicio JF, Barreales EG, Payero TD, Vicente CM, De Pedro A, Santos-Aberturas J. Biotechnological production and application of the antibiotic pimaricin: Biosynthesis and its regulation. Appl Microbiol Biotechnol. 2016;100(1):61–78. https://doi.org/10.1007/s00253-015-7077-0
29. Te Welscher YM, Jones L, Van Leeuwen MR, Dijksterhuis J, De Kruijff B, Eitzen G, et al. Natamycin inhibits vacuole fusion at the priming phase via a specific interaction with ergosterol. Antimicrob Agents Chemother. 2010;54(6):2618–2625. https://doi.org/10.1128/AAC.01794-09
30. Te Welscher YM, Hendrik H, Balagué MM, Souza CM, Riezman H, De Kruijff B, et al. Natamycin blocks fungal growth by binding specifically to ergosterol without permeabilizing the membrane. J Biol Chem. 2008;283(10):6393–6401. https://doi.org/10.1074/jbc.M707821200
31. Rolón M, Seco EM, Vega C, Nogal JJ, Escario JA, Gómez-Barrio A, et al. Selective activity of polyene macrolides produced by genetically modified *Streptomyces on Trypanosoma cruzi*. Int J Antimicrob Agents. 2006;28(2):104–109. https://doi.org/10.1016/j.ijantimicag.2006.02.025
32. Sunada A, Kimura K, Nishi I, Toyokawa M, Ueda A, Sakata T, et al. In vitro evaluations of topical agents to treat *Acanthamoeba keratitis*. Ophthalmology. 2014;121(10):2059–2065. https://doi.org/10.1016/j.ophtha.2014.04.013
33. Jackson M, Karwowski JP, Theriault RJ, Hardy DJ, Swanson SJ, Barlow GJ, et al. Altromycins, novel pluramycin-like antibiotics I. Taxonomy of the producing organism, fermentation and antibacterial activity. J Antibiot. 1990;43(3):223–228. https://doi.org/10.7164/antibiotics.43.223
34. Brill GM, Mcalpine JB, Whittern DN, Buko AM. Altromycins, novel pluramycin-like antibiotics II. Isolation and elucidation of structure. J Antibiot. 1990;43(3):229–237. https://doi.org/10.7164/antibiotics.43.229
35. Brill GM, Jackson M, Whittern DN, Buko AM, Hill P, Chen RH, et al. Altromycins E, F, G, H and I; additional novel components of the altromycin complex. J Antibiot. 1994;47(10):1160–1164. https://doi.org/10.7164/antibiotics.47.1160
36. Hansen M, Hurley L. Altromycin B threads the DNA helix interacting with both the major and the minor grooves to position itself for site-directed alkylation and guanine N7. J Am Chem Soc. 1995;117(9):2421–2429. https://doi.org/10.1021/ja00114a006
37. McAlpine JB, Karwowski JP, Jackson M, Brill GM, Kadam S, Shen L, et al. Altromycins: A new family of antitumor antibiotics-discovery and biological evaluation. In: Valeriote FA, Corbett TH, Baker LH, editors. Anticancer drug discovery and development: Natural products and new molecular models. Boston, MA: Springer; 1994. p. 95–117. https://doi.org/10.1007/978-1-4615-2610-0_6

38. Singh SB, Jayasuriya H, Ondeyka JG, Herath KB, Zhang C, Zink DL, et al. Isolation, structure, and absolute stereochemistry of platensimycin, a broad spectrum antibiotic discovered using an antisense differential sensitivity strategy. J Am Chem Soc. 2006;128(36):11916–11920. https://doi.org/10.1021/ja062232p

39. Martens E, Demain AL. Platensimycin and platencin: Promising antibiotics for future application in human medicine. J Antibiot. 2011;64(11):705–710. https://doi.org/10.1038/ja.2011.80

40. Yu Z, Rateb ME, Smanski MJ, Peterson RM, Shen B. Isolation and structural elucidation of glucoside congeners of platencin from *Streptomyces platensis* SB12600. J Antibiot. 2013;66(5):291–294. https://doi.org/10.1038/ja.2013.1

41. Zhang C, Ondeyka J, Herath K, Jayasuriya H, Guan Z, Zink DL, et al. Platensimycin and platencin congeners from *Streptomyces platensis*. J Nat Prod. 2011;74(3):329–340. https://doi.org/10.1021/np100635f

42. Zhang C, Ondeyka J, Dietrich L, Gailliot FP, Hesse M, Lester M, et al. Isolation, structure and biological activities of platencin A2-A4 from *Streptomyces platensis*. Bioorg Med Chem. 2010;18(7):2602–2610. https://doi.org/10.1016/j.bmc.2010.02.030

43. Herath KB, Zhang C, Jayasuriya H, Ondeyka JG, Zink DL, Burgess B, et al. Structure and semisynthesis of platensimide A, produced by *Streptomyces platensis*. Org Lett. 2008;10(9):1699–1702. https://doi.org/10.1021/ol900695v

44. Singh SB, Jayasuriya H, Herath KB, Zhang C, Ondeyka JG, Zink DL, et al. Isolation, enzyme-bound structure, and activity of platensimycin A1 from *Streptomyces platensis*. Tetrahedron Lett. 2009;50(37):5182–5185. https://doi.org/10.1016/j.tetlet.2009.06.118

45. Zhang C, Ondeyka J, Zink DL, Burgess B, Wang J, Singh SB. Isolation, structure and fatty acid synthesis inhibitory activities of platensimycin B1-B3 from *Streptomyces platensis*. Chem Comm. 2008(40):5034–5036. https://doi.org/10.1039/b810113b

46. Zhang C, Ondeyka J, Guan Z, Dietrich L, Burgess B, Wang J, et al. Isolation, structure and biological activities of platensimycin B4 from *Streptomyces platensis*. J Antibiot. 2009;62(12):699–702. https://doi.org/10.1038/ja.2009.106

47. Singh SB, Ondeyka JG, Herath KB, Zhang C, Jayasuriya H, Zink DL, et al. Isolation, enzyme-bound structure and antibacterial activity of platencin A1 from *Streptomyces platensis*. Bioorg Med Chem Lett. 2009;19(16):4756–4759. https://doi.org/10.1016/j.bmcl.2009.06.061

48. Jayasuriya H, Herath KB, Ondeyka JG, Zink DL, Burgess B, Wang J, et al. Structure of homoplatensimide A: A potential key biosynthetic intermediate of platensimycin isolated from *Streptomyces platensis*. Tetrahedron Lett. 2008;49(22):3648–3651. https://doi.org/10.1016/j.tetlet.2008.03.155

49. Kim KH, Ramadhar TR, Beemelmanns C, Cao S, Poulsen M, Currie CR, et al. Natalamycin A, an ansamycin from a termite-associated *Streptomyces* sp. Chem Sci. 2014;5(11):4333-–4338. https://doi.org/10.1039/C4SC01136H

50. Lee SR, Lee D, Park M, Lee JC, Park HJ, Kang KS, et al. Absolute configuration and corrected NMR assignment of 17-hydroxycyclooctatin, a fused 5-8-5 tricyclic diterpene. J Nat Prod. 2020;83(2):354–361. https://doi.org/10.1021/acs.jnatprod.9b00837

51. Kang HR, Lee D, Benndorf R, Jung WH, Beemelmanns C, Kang KS, et al. Termisoflavones A-C, isoflavonoid glycosides from termite-associated *Streptomyces* sp. RB1. J Nat Prod. 2016;79(12):3072–3078. https://doi.org/10.1021/acs.jnatprod.6b00738

52. Lee SR, Song JH, Song JH, Ko HJ, Baek JY, Trinh TA, et al. Chemical identification of isoflavonoids from a termite-associated *Streptomyces* sp. RB1 and their neuroprotective effects in murine hippocampal HT22 cell line. Int J Mol Sci. 2018;19(9), Art. #2640. https://doi.org/10.3390/ijms19092640

53. Lee D, Kang KS, Lee HJ, Kim KH. Chemical characterization of a renoprotective metabolite from termite-associated *Streptomyces* sp. RB1 against cisplatin-induced cytotoxicity. Int J Mol Sci. 2018;19(1), Art. #174. https://doi.org/10.3390/ijms19010174

54. Wyche TP, Ruzzini AC, Beemelmanns C, Kim KH, Klassen JL, Cao S, et al. Linear peptides are the major products of a biosynthetic pathway that encodes for cyclic depsipeptides. Org Lett. 2017;19(7):1772–1775. https://doi.org/10.1021/acs.orglett.7b00545

55. Therien AG, Huber JL, Wilson KE, Beaulieu P, Caron A, Claveau D, et al. Broadening the spectrum of β-lactam antibiotics through inhibition of signal peptidase type I. Antimicrob Agents Chemother. 2012;56(9):4662–4670. https://doi.org/10.1128/AAC.00726-12

56. Pérez-Bonilla M, Oves-Costales D, González I, De la Cruz M, Martin J, Vicente F, et al. Krisynomycins, imipenem potentiators against methicillin-resistant *Staphylococcus aureus*, produced by *Streptomyces canus*. J Nat Prod. 2020;83(9):2597–2606. https://doi.org/10.1021/acs.jnatprod.0c00294

57. Guo H, Benndorf R, Leichnitz D, Klassen JL, Vollmers J, Görls H, et al. Isolation, biosynthesis and chemical modifications of rubterolones A-F: Rare tropolone alkaloids from *Actinomadura* sp. 5-2. Chem Eur J 2017;23:9338–9345. https://doi.org/10.1002/chem.201701005

58. Lee SR, Lee D, Yu JS, Benndorf R, Lee S, Lee DS, et al. Natalenamides A-C, cyclic tripeptides from the termite-associated *Actinomadura* sp. RB99. Molecules. 2018;23(11), Art. #3003. https://doi.org/10.3390/molecules23113003

59. Rak Lee S, Schalk F, Schwitalla JW, Benndorf R, Vollmers J, Kaster AK, et al. Polyhalogenation of isoflavonoids by the termite-associated *Actinomadura* sp. RB99. J Nat Prod. 2020;83(10):3102–3110. https://doi.org/10.1021/acs.jnatprod.0c00676

60. Yoon SY, Lee SR, Hwang JY, Benndorf R, Beemelmanns C, Chung SJ, et al. Fridamycin A, a microbial natural product, stimulates glucose uptake without inducing *adipogenesis*. Nutrients. 2019;11(4), Art. #765. https://doi.org/10.3390/nu11040765

61. Beemelmanns C, Ramadhar TR, Kim KH, Klassen JL, Cao S, Wyche TP, et al. Macrotermycins A-D, glycosylated macrolactams from a termite-associated *Amycolatopsis* sp. M39. Org Lett. 2017;19(5):1000–1003. https://doi.org/10.1021/acs.orglett.6b03831

62. Curtis SM, Norton I, Everest GJ, Pelser JG, De Kock MC, Meyers PR. Development of a *Kribbella*-specific isolation medium and description of *Kribbella capetownensis* sp. nov. and *Kribbella speibonae* sp. nov., isolated from soil. Antonie van Leeuwenhoek. 2020;113(5):617–628. https://doi.org/10.1007/s10482-019-01365-6

63. Acquah KS, Beukes DR, Warner DF, Meyers PR, Sunassee SN, Maglangit F, et al. Novel South African rare actinomycete *Kribbella speibonae* strain SK5: A prolific producer of hydroxamate siderophores including new dehydroxylated congeners. Molecules. 2020;25(13), Art. #2979. https://doi.org/10.3390/molecules25132979

64. Cowan DA, Valverde A, Wingfield MJ, Rybicki EP, Tuffin MI. Biodiversity: So much more than legs and leaves. S Afr J Sci. 2013;109(11/12), Art. #a0037. https://doi.org/10.1590/sajs.2013/a0037