Anti-MRSA Activity of Actinomycin X\textsubscript{2} and Collismycin A Produced by \textit{Streptomyces globisporus} WA5-2-37 From the Intestinal Tract of American Cockroach (\textit{Periplaneta americana})

Zhiyu Chen\textsuperscript{1,2}, Peiyu Ou\textsuperscript{1,2}, Lingyan Liu\textsuperscript{1,2} and Xiaobao Jin\textsuperscript{1,2*}

\textsuperscript{1} School of Life Sciences and Biopharmaceutics, Guangdong Pharmaceutical University, Guangzhou, China, \textsuperscript{2} Guangdong Provincial Key Laboratory of Pharmaceutical Bioactive Substances, Guangdong Pharmaceutical University, Guangzhou, China

INTRODUCTION

\textit{Streptomyces globisporus} has been isolated from soil, plants, and so on (Jizba et al., 1991; Li et al., 2012), but rarely from insects. Previous literatures also showed that \textit{S. globisporus} was able to produce a wide variety of metabolites with antibacterial, antifungal, and antitumor activities, such as globimycin, radamycin, methylsulfomycin, etc. (Li X. et al., 2016; Kaweewan et al., 2017; Kemung et al., 2018). Therefore, \textit{S. globisporus} are considered as an economically and biotechnologically useful microorganism due to its diversity and proven ability to produce bioactive compounds.
However, it has not reported that *S. globisporus* could produce any bioactive compounds with anti-MRSA activity.

Cockroach is one of the most diverse and abundant pests all over the world (Hassan, 2017). *Periplaneta americana*, a largest species of common cockroach, has been extensively studied for its certain special substances and defense mechanisms to resist environmental threats (Li N. et al., 2016). Moreover, *P. americana* is a well-known medicinal insect in China with excellent therapeutic effects on wound repair and ulcer (Owen and Bouquillon, 1992; Song et al., 2017). But only few chemical investigations were reported on the title insect, especially the metabolites of the symbiotic microorganism of *P. americana*.

In this study, 159 strains were isolated from the intestinal tract of *P. americana* and 1 *S. globisporus* strain, WA5-2-37, showed great activity against MRSA ATCC 43300. Herein, this study aimed to determine the identity of the anti-MRSA compounds produced by *S. globisporus* strain WA5-2-37 and described the anti-MRSA action of the compounds.

**MATERIALS AND METHODS**

**Microorganisms, Chemicals, and Media**

The strain of bacteria was tested: MRSA ATCC 43300 which were obtained from Guangdong culture collection center. The genomic DNA isolation kit was purchased from Sangon Biotech. Microbiological media were purchased from Guangdong Huankai Microbial Sci. & Tech. Co., Ltd. High-performance liquid chromatography (HPLC) grade methanol was purchased from Honeywell. Solvents used for extraction and column chromatography were of analytical grade which were obtained from Guangdong oryx Sci-Tech Co., Ltd.

**Primary Anti-MRSA Activity Assay**

Primary screening for anti-MRSA activity was performed by growing cultures on Luria-Bertan (LB) agar plates swabbed with the MRSA ATCC 43300 suspension (1–5 × 10⁸ CFU/mL). 90 µL of the ethyl acetate extract obtained from each tested strain was added to an oxford cup on the LB agar plates and the incubated at 37°C for 12 h (Song et al., 2018). Vancomycin was used as positive control with the concentration of 100 µg/mL and the experiment was performed in triplicate. The diameter of the inhibition zone was recorded the anti-MRSA activity of each tested strain expressed millimeter.

**Isolation and Identification of *S. globisporus* Strain WA5-2-37**

The *P. americana* insects in this study were reared and provided by Guangdong Provincial Center for Disease Control and Prevention. The insects were firstly paralyzed at 4°C and washed alternately with water and 75% ethanol 3 times before dissection. Then the intestinal tract of *P. Americana* was dissected and removed with a sterile scalpel. The intestinal tract samples were grinded with sterile water and diluted to 1 × 10⁻³ g/mL, 2 × 10⁻³ g/mL, and 4 × 10⁻³ g/mL. Then the samples were coated on plate medium and cultured at 28°C for 7 days. Finally, the colony was purified and re-cultured, and a single colony was obtained according to the method of reference (Cook et al., 2007; Wang et al., 2014).

*S. globisporus* strain WA5-2-37 was inoculated on Gause’s synthetic agar no. 1 plates and incubated at 28°C for 4 days, during which time the culture had entered the logarithmic growth phase. The cultural and morphological characteristics of *S. globisporus* strain WA5-2-37 on Gause’s synthetic agar were tentatively identified based on the published descriptions of various Streptomyces species (Shirling and Gottlieb, 1968a,b, 1969, 1972). Whole genomic DNA of *S. globisporus* strain WA5-2-37 was extracted using the GenElute Bacterial Genomic DNA Kit (Sigma) according to manufacturer’s instructions. The 16S rDNA gene was then amplified by PCR, using universal bacterial primer set 27F (5′-AGA TTG TCA TGG TCA AG-3′) and 1492R (5′-TAG ACC TGG TAG CCA GTT ACC ACT T-3′) (Hamedi et al., 2013). The PCR was performed in a total volume of 25 µL containing 1 µL DNA template, 1 µL upstream primer (Invitrogen), 1 µL downstream primer (Invitrogen), 12.5 µL PCR Premix (Invitrogen) and 9.5 µL ddH₂O. The PCR cycle consisted of a 94°C pre-denaturation for 4 min, 35 cycles of three steps (94°C denaturation for 30 s, 55°C for annealing for 30 s, 72°C extension for 30 s), and 72°C post-cycle extension for 5 min. After that, PCR products were verified by electrophoresis on a 1.5% agarose gel and was sequenced by Beijing Genomics Institute (Beijing, China). Sequences analysis of 16S rDNA was then conducted using BLAST hosted by National Center for Biotechnology Information (NCBI) website1. Sequences were aligned with Clustal software and MEGA 7.0 was used to carry out a phylogenetic tree of the alignment using the maximum likelihood method (Kumar et al., 2008; Tamura et al., 2011).

**Fermentation, Extraction, and Isolation**

Single colonies of *S. globisporus* strain WA5-2-37 were inoculated into a 250 mL flask containing 100 mL of ISP-1 medium as seed and then incubated on a rotary shaker at 28°C and 150 rpm for 2 days, which was used as seed culture. From this, 5% of this seed was transferred to 500 mL flask containing 300 mL of ISP-2 medium as production flasks and incubated at 28°C and 150 rpm for 7 days. The total 40 L of fermentation were extracted with 12 major fractions (A–L). Every fraction was used for testing for anti-MRSA action of the compounds.

**Spectroscopic Analysis**

The active and pure fraction was analyzed by analytical HPLC using a YMC-Pack ODS-AQ C18 column (4.5 mm × 250 mm, YMC, Japan) with a photodiode array detector (PAD). The

---

1https://www.ncbi.nlm.nih.gov/
proton and carbon nuclear magnetic resonance (NMR) spectra were recorded at 600 MHz using a Brucker AVANCE III 600M spectrometer (Brucker) with tetramethysilane (TMS) as an internal reference and dimethyl sulfoxide (DMSO) as solvent.

**Minimum Inhibitory Concentration Testing**

The minimum inhibitory concentration (MIC) for purified compounds against the MRSA were determined by the tube-dilution method using individually pack-aged, flat bottomed, 96-well microtiter plates (Liu et al., 2017). The tested bacterial strain was cultivated in Mueller-Hinton Broth at 37°C until the cellular density reached approximately $1 \sim 5 \times 10^5$ CFU/mL. Each of the tested compounds and drugs were dissolved in DMSO and then diluted with sterile broth by the twofold dilution method. The final concentrations of each sample in the wells were 512, 256, 128, 64, 32, 16, 8, 4, 2, 1, 0.5, 0.25, and 0.125 µg/mL. Vancomycin was used as positive control and ampicillin was used as a control to prove that the tested bacteria were MRSA. A serial dilution of compounds were performed in the microplates and incubated at 37°C for 12 h. All the experiment was performed in triplicate and the last tube with no growth of microorganism was recorded as the MIC value expressed in µg/mL.

**Scanning Electron Microscopy Analysis**

The morphological changes of the tested MRSA were determined by scanning electron microscopy (SEM). The tested bacterial suspension ($1 \sim 5 \times 10^5$ CFU/mL) were treated with 4× the MIC of the compound for 12 h at 37°C. Cells were then placed onto a glass coverslip and fixed with 2.5% glutaraldehyde at 4°C overnight. Fixed samples were washed with 1× phosphate buffered saline (PBS) three times for 20 min each and dehydrated in increasing concentrations of ethanol (20, 40, 60, 80, and 100%). The coverslips were finally dried then analyzed by Quanta FEG 200 FESEM at an accelerating voltage of 2–19 kV under standard operating conditions.
GenBank Submission

The partial sequence of the 16S rDNA gene of isolate WA5-2-37 was submitted to NCBI GenBank under accession number MH304280.

RESULTS

Screening of the Strains With Anti-MRSA Activity and Identification of Strain WA5-2-37

The screening for anti-MRSA activity showed the strain WA5-2-37 had great activity against MRSA ATCC 43300 (Figure 1A). Based on the cultural and morphological characteristics showed in Figures 1B–D, strain WA5-2-37 belonged to genus Streptomyces. The strain appeared as leathery colonies with abundant spores (powder) on the Gause’s synthetic agar no. 1 plates. The Gram staining result showed strain WA5-2-37 was the Gram-positive bacterium. The morphological characteristics were observed under SEM and the result showed that the spore of strain WA5-2-37 were found to be oval to round in shape. The phylogenetic analyses (Figure 2) based on 16S DNA sequences and the taxonomic position of strain WA5-2-37 in the phylogenetic tree indicated that strain WA5-2-37 was most similar to S. globisporus NBRC 12867 (AB184203). Based on these result, strain WA5-2-37 was preliminary confirmed as S. globisporus.

Isolation and Purification of the Anti-MRSA Compounds

Strain WA5-2-37 was cultured by the shake-flask method, after which the culture broth (40 L) was then centrifuged at 4000 rpm at 4°C cell-free supernatants were extracted with an equal volume of ethyl acetate. The organic layer was then concentrated in vacuo using a rotary evaporator. After that, the crude extract (12.0 g) of strain WA5-2-37 was subjected to bioactivity guided isolation and purification. Twelve different major fractions (A–L) were obtained after eluting by gradient elution with an increasing polarity of ethyl acetate (1–100%) and hexane. Each fraction was then concentrated and tested for activity against MRSA. The fraction A and D showed the most potent activity against MRSA ATCC 43300. The fraction A were separated further on Spehadox LH-20 with a solvent gradient of 100% methanol and purified by semi-preparative HPLC with a solvent gradient of 80% methanol.
FIGURE 3 | Structure identification of the purified compound 1. (A) Molecular structure of the compound. (B) Mass spectrometry. (C) Proton ($^1$H) nuclear magnetic resonance. (D) Carbon ($^{13}$C) nuclear magnetic resonance spectrum of the compound 1 in DMOS-6d.

FIGURE 4 | Structure identification of the purified compound 2. (A) Molecular structure of the compound. (B) Mass spectrometry. (C) Proton ($^1$H) nuclear magnetic resonance. (D) Carbon ($^{13}$C) nuclear magnetic resonance spectrum of the compound 2 in DMOS-6d.
The fraction D were purified by semi-preparative HPLC with a solvent gradient of 70% methanol. There two pure compounds were obtained finally.

**Spectroscopic Analysis**

The structures of two pure compounds were elucidated by high resolution electrospray ionization mass spectrometry (HRESIMS) and NMR analysis as well as by comparison with the previously reported MS and NMR data. HRESIMS of pure compounds showed molecular ion peaks at \( m/z \) 1291.59686 [M + Na]\(^+\) for compound 1 (Figure 3B) and 276.07993 [M + H]\(^+\) for compound 2 (Figure 4B). The \(^1\)H and \(^{13}\)C NMR spectra of pure compounds showed a great similarity to the previously reported data (Figures 3C,D, 4C,D). Therefore, the structures of compounds 1 and 2 were confirmed to be actinomycin X\(_2\) and collismycin A (Figures 3A, 4A).

**Anti-MRSA Activity of the Isolated Compounds**

As the results showed in Table 1, compounds 1 and 2 showed significant activity against MRSA ATCC 43300 with MIC values of 0.25 and 8 \( \mu \)g/mL, which was higher than vancomycin (0.125 \( \mu \)g/mL). Moreover, ampicillin showed no inhibitory activity against MRSA, indicating that the strain was MRSA. The SEM analysis showed the cell membrane of MRSA were destroyed after treating actinomycin X\(_2\) and collismycin A (Figure 5).

**DISCUSSION**

Methicillin-resistant *S. aureus* is recognized as one of the pathogens that causes hospital- and community-acquired infections worldwide (Sit et al., 2017). MRSA is highly prevalent in hospitals worldwide, especially in Asia (Stefani et al., 2012). Moreover, the outcome of invasive MRSA infection is poor with high 90-day mortality rates from bacteremia (Nickerson et al., 2009; Bal et al., 2017). Therefore, there is an emerging demand for discovery of novel, safe and potent bioactive compounds with anti-MRSA activity (Uchida et al., 2014). Therefore, more and more research efforts have focused on the exploration of microorganism to discover bioactive secondary metabolites (Gos et al., 2017; Sadaf et al., 2018; Lee et al., 2018). But with decreased of the identification of natural products from traditional sources, scientists have turned their eyes to the microorganism from new sources such as insects (Beemelmanns et al., 2017; Ma et al., 2017). Insects, as the most abundant species in nature, can adapt and live in a variety of environments. Moreover, insects contain many types of symbiotic microorganisms in their bodies that produce a variety of bioactive substances (Beemelmanns et al., 2016; Angela, 2018). Therefore, the microbes isolated from insects can be a new source to identify bioactive substances.

In this study, we carried out taxonomic identification of strain WA5-2-37 from the intestinal tract of *P. americana* and isolated and characterized the bioactive metabolites produced by this strain. Strain WA5-2-37 was identified as *S. globisporus* based on morphological characteristics and molecular identification. WA5-2-37 is the first reported strain of *S. globisporus* found in the intestinal tract of *P. americana*. To the best of our knowledge, *S. globisporus* was reported capable of producing a variety of bioactive substances (Chen et al., 2010; Seo et al., 2001), whereas strain WA5-2-37 is the first reported naturally occurring strain of *S. globisporus* with anti-MRSA activity. Without any optimization of the culture conditions, strain WA5-2-37 showed better activity against MRSA ATCC 43300 than other strains in this study. Therefore, optimization of the culture conditions and gene regulation was expected to

---

**TABLE 1 |** The anti-MRSA activity of compounds isolated from strain WA5-2-37 (MIC, \( \mu \)g/mL, \( n = 3 \)).

| Tested strain                  | Compound 1 | Compound 2 | Vancomycin | Ampicillin |
|-------------------------------|------------|------------|------------|------------|
| Methicillin-resistant Staphylococcus aureus ATCC 43300 | 0.25       | 8          | 0.125      | –          |

\( \sim \), no activity was detected.

---

**FIGURE 5 |** Scanning electron microscopy (SEM) images of pure compounds treated and untreated MRSA (50,000×). (A) Untreated MRSA. (B) Actinomycin X\(_2\) treated MRSA. (C) Collismycin A treated MRSA.
be done to exploit the full potential for discovery of biologically active substances of this strain.

Actinomycin X₂ and collisymycin A are potent bioactive substances that can be used in the treatment of several human diseases, such as cancer, bacterial infection, and so on (Reich, 1963; Chen et al., 2012). Both of these two compounds can be produced by many Streptomyces sp. (Xiong et al., 2008; Kawatani et al., 2016), but it is the first reported that S. globisporus can produce actinomycin X₂ and collisymycin A. Moreover, the current studies showed that collisymycin A was almost discovered from plants, marine, or soil sources, but never found in insects (Vior et al., 2014; Lee et al., 2017). Herein, this literature is the first reported that a S. globisporus isolated from the intestinal tract of P. americana with the ability to produce actinomycin X₂ and collisymycin A. In this study, actinomycin X₂ and collisymycin A showed great activity against MRSA ATCC 43300 with the MIC values of 0.25 and 8 µg/mL, and both of them were harmful to the cell membrane of MRSA. However, the specific mechanisms of the anti-MRSA of actinomycin X₂ and collisymycin A were unclear and need to be studied in further research.

**REFERENCES**

Angela, E. D. (2018). Omics and the metabolic function of insect-microbial symbioses. *Curr. Opin. Insect. Sci.* 29, 1–6. doi: 10.1016/j.cois.2018.05.012

Bal, A. M., David, M. Z., Garau, J., Gottlieb, T., Mazzei, T., Scaglione, F., et al. (2017). Future trends in the treatment of meticillin-resistant Staphylococcus aureus (MRSA) infection: an in-depth review of newer antibactericactive against an enduring pathogen. *J. Glob. Antimicrob. Resist.* 10, 295–303. doi: 10.1016/j.jgar.2017.05.019

Beemelmanns, C., Guo, H., Rischer, M., and Poulsen, M. (2016). Natural products from microbes associated with insects. *Beilstein. J. Org. Chem.* 12, 314–327. doi: 10.3762/bjoc.12.34

Beemelmanns, C., Ramadhar, T. R., Kim, K. H., Klassen, J. L., Cao, S., Wyche, T. P., et al. (2017). Macrotetmycins A-D, glycosylated macroolactams from a termite-associated *Amycolatopsis* sp. M39. *Org. Lett.* 19, 1000–1003. doi: 10.1021/acs. orglett.6b03831

Chen, C., Song, F., Wang, Q., Abdel-Mageed, W. M., Guo, H., Fu, C., et al. (2012). A marine-derived *Streptomyces* sp. MS494 produces high yield of actinomycin X2and actinomycin D with potent anti-tuberculosis activity. *Appl. Microbiol. Biotechnol.* 95:199927. doi: 10.1007/s00253-012-4079-z

Chen, Y., Yin, M., Horxman, G. P., Huang, S., and Shen, B. (2010). Manipulation of pathway regulation in *Streptomyces globisporus* for overproduction of the endene antitumor antibiotic C-1027. *J. Antibiot.* 63, 482–485. doi: 10.1038/ja.2010.55

Cook, D. M., DeCrescenzo, H. E., Upchurch, R., and Peterson, J. B. (2007). Isolation of polymer-degrading bacteria and characterization of the hindgut bacterial community from the detritus-feeding larvae of Tipula abdominalis (*Diptera: Tipulidae*). *Appl. Environ. Microbiol.* 73, 5683–5686. doi: 10.1128/AEM.00213-07

Esposito, M., Nothias, L. F., Nedev, H., Gallard, J. F., Leyssen, P., Retailleau, P., et al. (2016). *Euphorbia dendroides* latex as a source of jatrophone esters: isolation, structural analysis, conformational study, and Anti-CHIKV activity. *J. Nat. Prod.* 79, 2873–2882. doi: 10.1021/acs.jnatprod.6b00644

Gos, F., Savi, D. C., Shaaban, K. A., Thorson, J. S., Aluizio, R., Possiede, Y. M., et al. (2016). Antibacterial activity of endophytic actinomycetes isolated from the medicinal plant vochysia divergens (Pantanal, Brazil). *Front. Microbiol.* 8:1642. doi: 10.3389/fmicb.2017.01642

Hamed, I., Mohammadipanah, F., and Ventosa, A. (2013). Systematic and biotechnological aspects of halophilic and halotolerant actinomycetes. *Extremophiles* 17, 1–13. doi: 10.1007/s00792-012-0493-5

Hassan, N. (2017). Infestation of cockroaches (*Insecta: Blattaria*) in the human dwelling environments: a systematic review and meta-analysis. *Acta Trop.* 167, 86–98. doi: 10.1016/j.actatropica.2016.12.019

Jiha, J., Sedmera, P., Zima, J., Beran, M., Blumauerová, M., Kandybin, N. V., et al. (1991). Macrotetrolide antibiotics produced by *Streptomyces globisporus*. *Folia. Microbiol.* 36, 437–443. doi: 10.1007/BF028, 84062.

Kawatani, M., Muroi, M., Wada, A., Inoue, G., Futamura, Y., Aono, H., et al. (2016). Proteomic profiling reveals that collisymycin A is an iron chelator. *Sci. Rep.* 6:38385. doi: 10.1038/srep38385

Kaweewan, I., Komaki, H., Hemmi, H., and Kodani, S. (2017). Isolation and structure determination of a new thiopelptide glyobicymic from *Streptomyces globisporus* subsp. globisporus based on genome mining. *Tetrahedron. Lett.* 4, 409–414. doi: 10.1016/j.tetlet.2017.12.056

Kemung, H. M., Tan, I. T., Khan, T. M., Chan, K. G., Pusparajah, P., Goh, B. H., et al. (2018). *Streptomyces* as a prominent resource of future Anti-MRSA Drugs. *Front. Microbiol.* 9:2221. doi: 10.3389/fmicb.2018.02221

Kumar, S., Nei, M., Dudley, J., and Tamura, K. (2008). MEGA: a biologist-centric software for evolutionary analysis of DNA and protein sequences. *Brief. Bioinform.* 9, 299–206. doi: 10.1093/bib/bbn017

Lee, J. H., Kim, E., Choi, H., and Lee, J. (2017). Collisymycin C from the micrornese marine bacterium *Streptomyces* sp. MC025 inhibits staphylococcus aureus biofilm formation. *Mar. Drugs.* 15:e387. doi: 10.3390/md15123087

Lee, I. H., Chan, K. G., Stach, J., Wellington, E., and Goh, B. H. (2018). Editorial: the search for biological active agent(s) from actinobacteria. *Front. Microbiol.* 9:824. doi: 10.3389/fmicb.2018.00824

Li, N., Lu, R., Yu, Y., Lu, Y., Huang, L., Jin, J., et al. (2016). Protective effect of Periplaneta americana extract in ulcerative colitis rats induced by dinitrochlorobenzene and acetic acid. *Pharm. Biol.* 54, 2560–2567. doi: 10.3109/13880290.2016.1170862

Li, Q., Ning, P., Zheng, L., Huang, J., Li, G., and Hsiang, T. (2012). Effects of volatile substances of *Streptomyces globisporus* JK-1 on control of Botrytis cinerar on tomato fruit. *Biol. Control. 61*, 113–120. doi: 10.1016/j.biocontrol.2011.10.014

Li, X., Lei, X., Zhang, C., Jiang, Z., Shi, Y., Wang, S., et al. (2016). Complete genome sequence of *Streptomyces globisporus* C-1027, the producer of an endene antibiotic lidamicyn. *J. Biotechnol.* 222, 9–10. doi: 10.1016/j.jbiotec.2016.02.004

Liu, F., Chen, G., Zhang, L. H., Liu, B., Chen, S., Hua, H., et al. (2017). Isolation and capacity building project of Guangdong Province (Nos. PJ03-11. 17, 1–13. doi: 10.1007/s00792-012-0493-5

DATA AVAILABILITY STATEMENT

The datasets generated for this study can be found in the NCBI GenBank under accession number MH304280.

**AUTHOR CONTRIBUTIONS**

XJ and ZC conceived and designed the experiments and wrote the manuscript. ZC, PO, and LL performed the experiments and analyzed the data. All authors read and approved the manuscript.

**FUNDING**

This work was funded by the Public Welfare Research and Capacity Building Project of Guangdong Province (Nos. 2016A030303059 and 2017A020211008). The Key Projects of Basic Research and Applied Basic Research of Guangdong Province Normal University (No. 2018KZDXM041).
Nickerson, E. K., Hongswan, M., Limmathurosakul, D., Wuthiekaun, V., Shah, K. R., Srismang, P., et al. (2009). Staphylococcus aureus bacteraemia in a tropical setting: patient outcome and impact of antibiotic resistance. *Plos. One* 4:e4308. doi: 10.1371/journal.pone.0004308

Owen, M. D., and Bouquillon, A. I. (1992). The synthesis of (I-DOPA) in the cerebral ganglia of the cockroach, *Periplaneta americana* L. *Insect. Biochem. Molec.* 22, 193–198. doi: 10.1016/0965-1748(92)90159-C

Reich, E. (1963). Biochemistry of actinomycin. *Cancer. Res.* 23, 1428–1441.

Sadaf, M., Bilal, H. A., Uzair, B., and Rashda, A. (2018). Natural products as reservoirs of novel therapeutic agents. *EXCLI. J.* 17, 420–451. doi: 10.17179/excl2018–1174

Seo, H. J., Shimonishi, T., Ohmiya, K., and Hayashi, K. (2001). Characterization of N-acetylmuramidase M-1 of *Streptomyces globisporus* produced by *Escherichia coli* BL21(DE3)pLysS. *J. Biosci. Bioeng* 92, 472–474. doi: 10.1016/s1389-1723(01)80299-1

Shirling, E. B., and Gottlieb, D. (1968a). Cooperative description of type culture of *Streptomyces*. III. Additional species descriptions from first and second studies. *Int. J. Syst. Bacteriol.* 18, 279–392. doi: 10.1099/00207713-19-4-391

Shirling, E. B., and Gottlieb, D. (1968b). Cooperative description of type cultures of *Streptomyces*. IV. Species descriptions from the second, third and fourth studies. *Int. J. Syst. Bacteriol.* 18, 69–189. doi: 10.1099/00207713-19-4-391

Shirling, E. B., and Gottlieb, D. (1969). Cooperative description of type culture of *Streptomyces*. III. Additional species descriptions from first and second studies. *Int. J. Syst. Bacteriol.* 19, 391–512. doi: 10.1099/00207713-19-4-391

Song, Q., Gou, Q., Xie, Y., Zhang, Z., and Fu, C. (2017). *Periplaneta americana* extracts promote skin wound healing via nuclear factor kappa B canonical pathway and extracellular signal-regulated kinase signaling. *evid.-based. Compl. Alt.* 2017, 5821706. doi: 10.1155/2017/5821706

Song, T., Xi, Y., and Du, J. (2018). Antibacterial hydrogels incorporated with poly(glutamic acid)-based vesicles. *Acta. Polym. Sin.* 1, 119–128. doi: 10.17777/j.issn1000-3304.2018.17229

Stefani, S., Chung, D. R., Lindsay, J. A., Friedrich, A. W., Kearns, A. M., Westh, H., et al. (2012). Methicillin-resistant *Staphylococcus aureus* (MRSA): global epidemiology and harmonisation of typing methods. *Int. J. Antimicrob. Agents.* 39, 273–282. doi: 10.1016/j.ijantimicag.2011.09.030

Tamura, K., Dudley, J., Nei, M., and Kumar, S. (2011). MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. *Mol. Biol. Evol.* 24, 1596–1599. doi: 10.1093/molbev/msr121

Uchida, R., Hanaki, H., Matsu, H., Hamamoto, H., Sekimizu, K., Iwatsuki, M., et al. (2014). In vitro and in vivo anti-MRSA activities of nosokomycins. *Drug. Discov. Ther.* 8, 249–254. doi: 10.5582/dtt.2014.01050

Vior, N. M., Ólano, C., García, I., Méndez, C., and Salas, J. A. (2014). Collidimycin A biosynthesis in *Streptomyces* sp. CS4 is regulated by iron levels through two pathway-specific regulators. *Microbiology* 160, 467–478. doi: 10.1099/mic.0.075218-0

Wang, H., Jin, L., Peng, T., Zhang, H., Chen, Q., and Hua, Y. (2014). Identification of cultivable bacteria in the intestinal tract of *Bactrocera dorsalis* from three different populations and determination of their attractive potential. *Pest. Manag. Sci.* 70, 80–87. doi: 10.1002/ps.3528

Xiong, Z. Q., Tu, X. R., and Tu, G. Q. (2008). Optimization of medium composition for actinomycin X2 production by *Streptomyces* spp JAU4234 using response surface methodology. *J. Ind. Microbiol. Biotechnol.* 35, 729–734. doi: 10.1007/s10295-008-0339-4

**Conflict of Interest:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Chen, Ou, Liu and Jin. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.