

Screening and Identification of a Newly Antimicrobial *Streptomyces rehmannaiae* from Rhizosphere of *Rehmannia glutinosa*

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Running title: Screening and Identification of a *Streptomyces rehmannaiae*

Supplementary Materials

Table S1 Fermentation medium used in this study.

Medium	Ingredients and content
GYM I	skim milk powder 0.1%, yellow bean cake flour 2%, sucrose 8%, K ₂ HPO ₄ ·3H ₂ O 0.1%, FeSO ₄ ·7H ₂ O 0.1%, CaCO ₃ 0.3%, pH 7.2-7.4
GYM III	D-glucose 0.4%, yeast extract powder 0.4%, malt extract 1%, CaCO ₃ 0.2%, pH 7.2
ISP 2	yeast extract powder 0.4%, malt extract 1%, D-glucose 0.4%, pH 7.2-7.4
HZ-2	yeast extract powder 0.4%, FeSO ₄ ·7H ₂ O 0.1%, malt extract 0.2%, D-glucose 1%, soluble starch 4%, CaCO ₃ 0.2%, MnCl ₂ ·4H ₂ O 0.1%, pH 7.2-7.4
GY	yeast extract powder 1%, D-glucose 1%, K ₂ HPO ₄ ·3H ₂ O 0.05%, MgSO ₄ ·7H ₂ O 0.05%, pH 7.2-7.4
HZ-1	yeast extract powder 0.4%, maltodextrin 1%, D-glucose 1%, soluble starch 4%, trace element solution 0.1mL/L (FeSO ₄ ·7H ₂ O 0.1%, MnCl ₂ ·4H ₂ O 0.1%, ZnSO ₄ ·7H ₂ O 0.1%), pH 7.2-7.4
MB	D-glucose 1%, soluble starch 0.5%, yeast extract powder 0.2%, NaCl 0.4%, peptone 0.2%, K ₂ HPO ₄ ·3H ₂ O 0.05%, MgSO ₄ ·7H ₂ O 0.05%, CaCO ₃ 0.2%, pH 7.2
H9	soluble starch 2%, cottonseed meal powder 1%, yeast extract powder 0.5%, maltodextrin 2%, malt extract 0.5%, MgSO ₄ ·7H ₂ O 0.2%, NaCl 0.2%, CaCO ₃ 0.2%, pH 7.0-7.2

Table S2. Genes and their position and products associated in nigericin biosynthetic gene cluster.

Identifiers	Position	Product
ABC84455.1	420 - 1232 (-)	NigE
ABC84456.1	1475 - 8722 (+)	NigAI
ABC84457.1	8764 - 15465 (+)	NigAII
ABC84458.1	15507 - 27758 (+)	NigAIII
ABC84459.1	27775 - 39942 (+)	NigAIV
ABC84460.1	39985 - 52122 (+)	NigAV
ABC84461.1	52139 - 57244 (+)	NigAVI
ABC84462.1	57331 - 58197 (-)	NigCII
ABC84463.1	58370 - 59629 (+)	NigD
ABC84464.1	59705 - 60043 (-)	NigAXI
ABC84465.1	60152 - 64051 (-)	NigAX
ABC84466.1	64048 - 65478 (-)	NigCI
ABC84467.1	65513 - 65980 (-)	NigBIII
ABC84468.1	66016 - 66453 (-)	NigBI
ABC84469.1	66450 - 72278 (-)	NigAIX
ABC84470.1	72327 - 78947 (-)	NIGAVIII
ABC84471.1	78995 - 93358 (-)	NigAVII
ABC84472.1	93400 - 93948 (-)	unknown

Figure legends

Figure S1 The deposit certificate of strain Rer75.

Figure S2 Morphological characteristics of aerial mycelia and spores of strain Rer75 using SEM.

Figure S3 Cultural characteristics observed on tested media for strain Rer75.

Figure S4 Phospholipid composition of strain Rer75.

Figure S5 Subsystem statistics of *Streptomyces* sp. Rer75 genome analyzed by RAST.

Figure S6 HPLC of compound **1**.

Figure S7 HRESIMS of compound **1**.

Figure S8 ^1H spectrum of compound **1**.

Figure S9 ^1H NMR of expanded regions at 2–2.7 ppm.

Figure S10 ^1H NMR of expanded regions at 5–7.1 ppm.

Figure S11 ^{13}C spectrum of compound **1**.

Figure S12 Dept 135 spectrum of compound **1**.

Figure S13 HSQC spectrum of compound **1**.

Figure S14 ^1H - ^1H cosy spectrum of compound **1**.

Figure S15 HMBC spectrum of compound **1**.

中国典型培养物保藏中心

用于专利程序的培养物保藏受理通知书 (收据)

地址: 中国, 武汉, 武汉大学 邮编: 430072 电话: (027) 68754052 传真: (027) 68754833 E-mail: cctcc@whu.edu.cn

请求保藏人及其代理人:

请求保藏人: 河南中医药大学 何海荣

专利代理人:

专利申请号:

您(们)提供请求保藏的培养物名称
及注明的鉴别特征:

本保藏中心保藏编号
CCTCC NO: M 20231749

Streptomyces rehmanna Rer75

上述请求保藏的培养物附有

科学描述

提议的分类命名

注: 在框内打 √ 号表示有, 打×号表示没有。

该培养物已于 2023 年 09 月 20 日由本保藏中心收到, 并登记入册。根据您(们)的请求, 由该日起保存三十年, 在期满前收到提供培养物样品的请求后再延续保存五年。

该培养物的存活性本保藏中心于 2023 年 09 月 27 日检测完毕, 结果为存活。

中国典型培养物保藏中心

负责人(签名)



2023 年 09 月 27 日

Figure S1 The deposit certificate of strain Rer75.

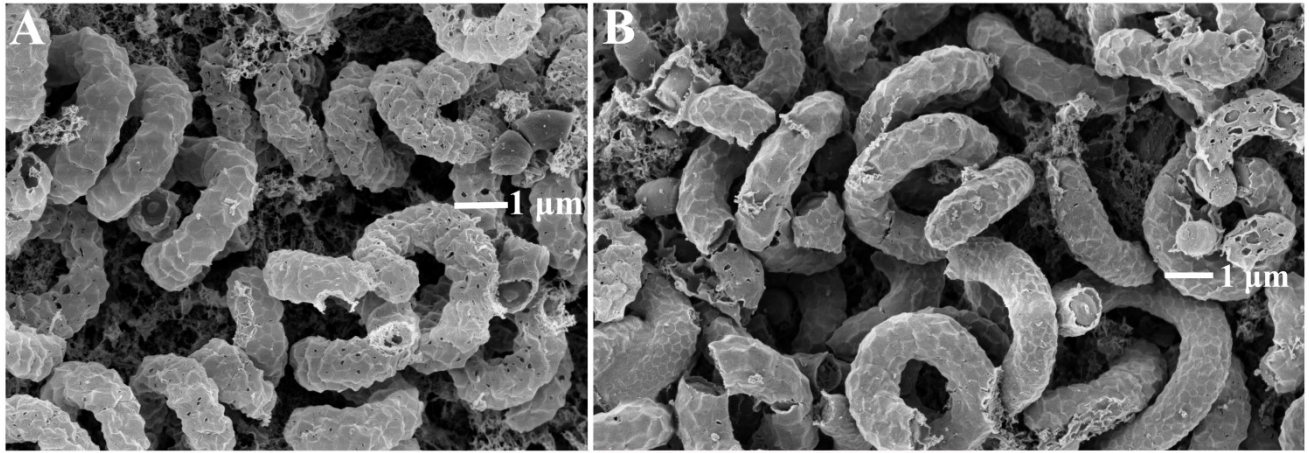


Figure S2 Morphological characteristics of aerial mycelia and spores of strain Rer75 using SEM.



Figure S3 Cultural characteristics observed on tested media for strain Rer75.

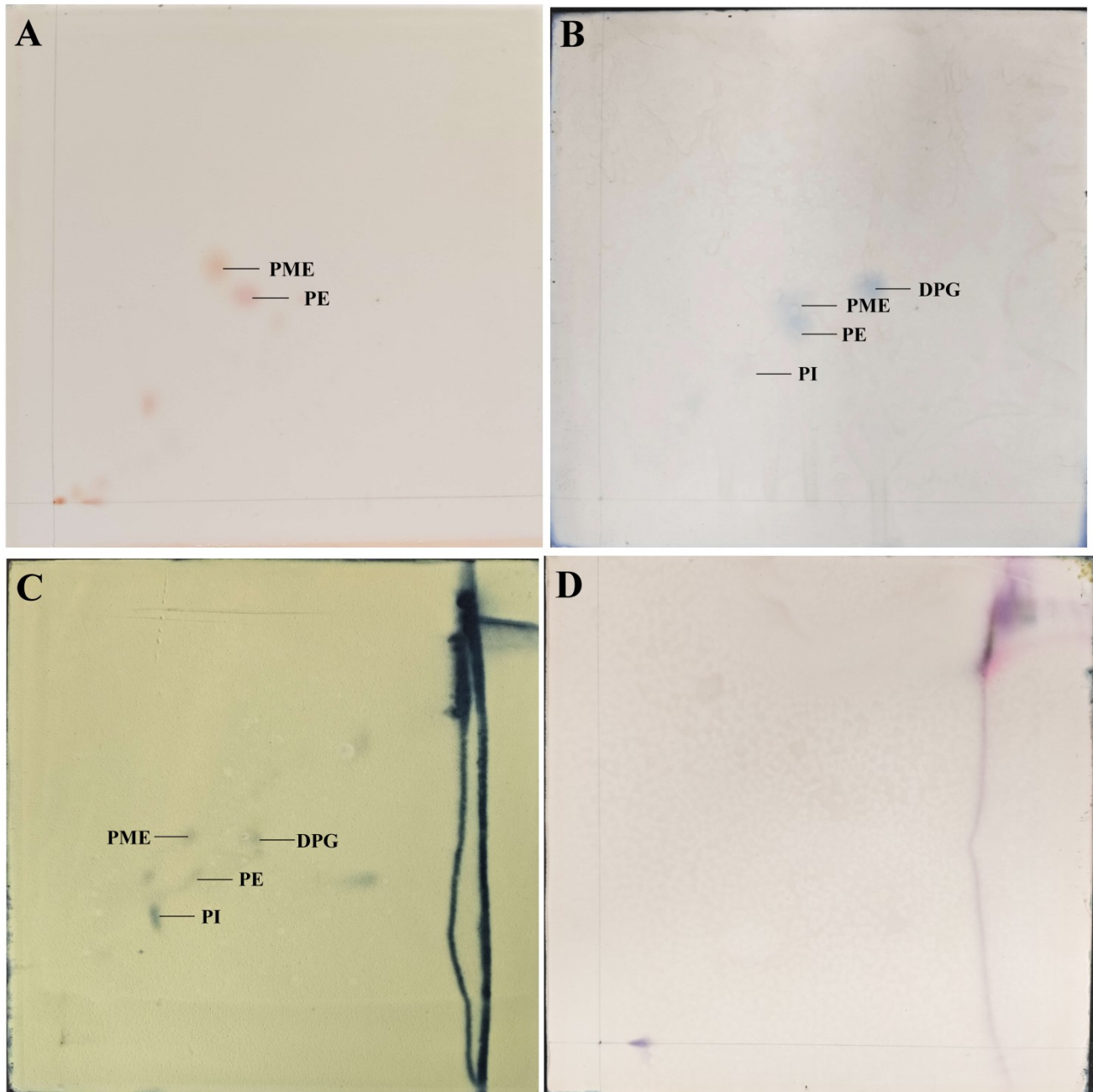
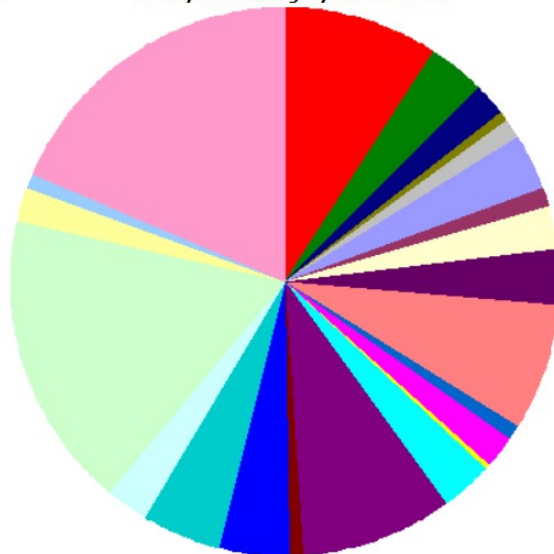


Figure S4 Phospholipid composition of strain Rer75

Subsystem Coverage



Subsystem Category Distribution



Subsystem Feature Counts

- ☒ Cofactors, Vitamins, Prosthetic Groups, Pigments (428)
- ☒ Cell Wall and Capsule (158)
- ☒ Virulence, Disease and Defense (90)
- ☒ Potassium metabolism (21)
- ☒ Photosynthesis (0)
- ☒ Miscellaneous (56)
- ☒ Phages, Prophages, Transposable elements, Plasmids (4)
- ☒ Membrane Transport (146)
- ☒ Iron acquisition and metabolism (56)
- ☒ RNA Metabolism (117)
- ☒ Nucleosides and Nucleotides (149)
- ☒ Protein Metabolism (345)
- ☒ Cell Division and Cell Cycle (34)
- ☒ Motility and Chemotaxis (12)
- ☒ Regulation and Cell signaling (92)
- ☒ Secondary Metabolism (20)
- ☒ DNA Metabolism (141)
- ☒ Fatty Acids, Lipids, and Isoprenoids (405)
- ☒ Nitrogen Metabolism (38)
- ☒ Dormancy and Sporulation (10)
- ☒ Respiration (194)
- ☒ Stress Response (219)
- ☒ Metabolism of Aromatic Compounds (114)
- ☒ Amino Acids and Derivatives (803)
- ☒ Sulfur Metabolism (87)
- ☒ Phosphorus Metabolism (46)
- ☒ Carbohydrates (822)

Figure S5 Subsystem statistics of *Streptomyces* sp. Rer75 genome analyzed by RAST.

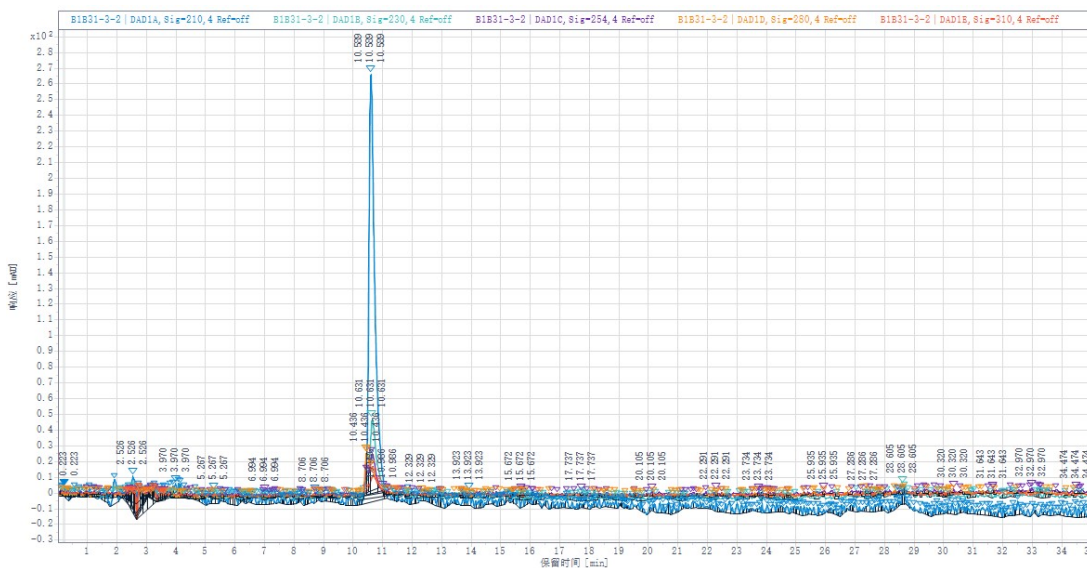


Figure S6 HPLC of Compound 1.

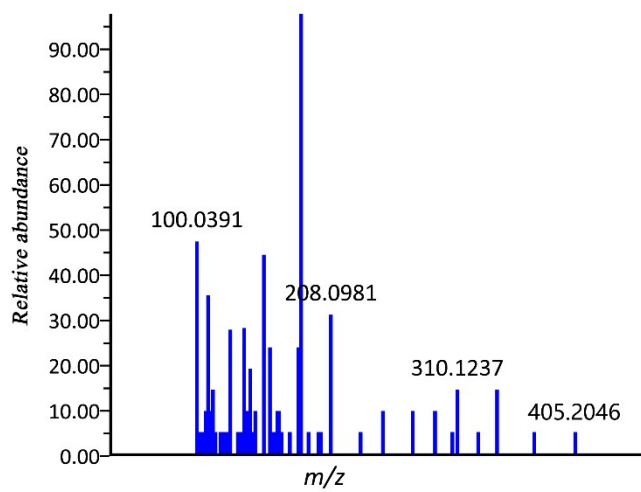
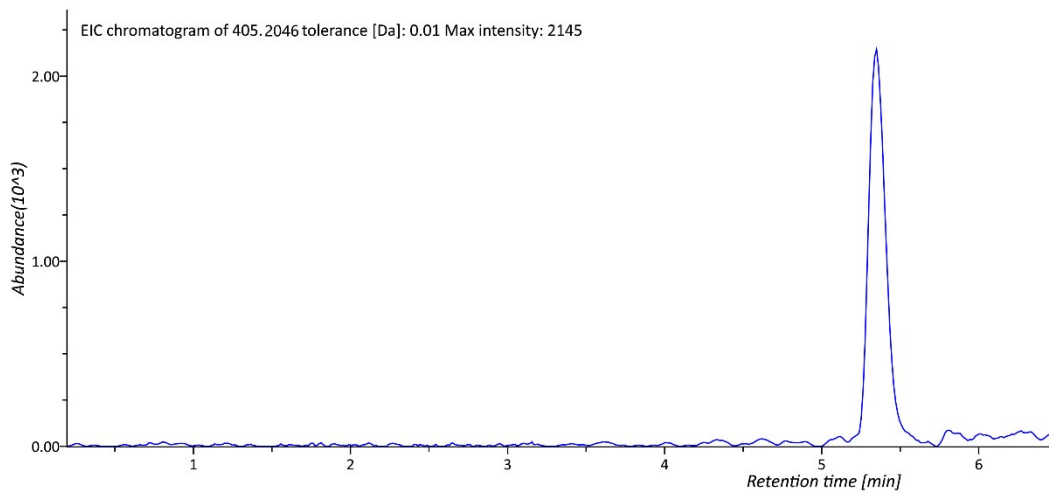


Figure S7 HRESIMS of compound **1**.

HJR-A1-10.1.fid
HJR-A1-10
MEOD

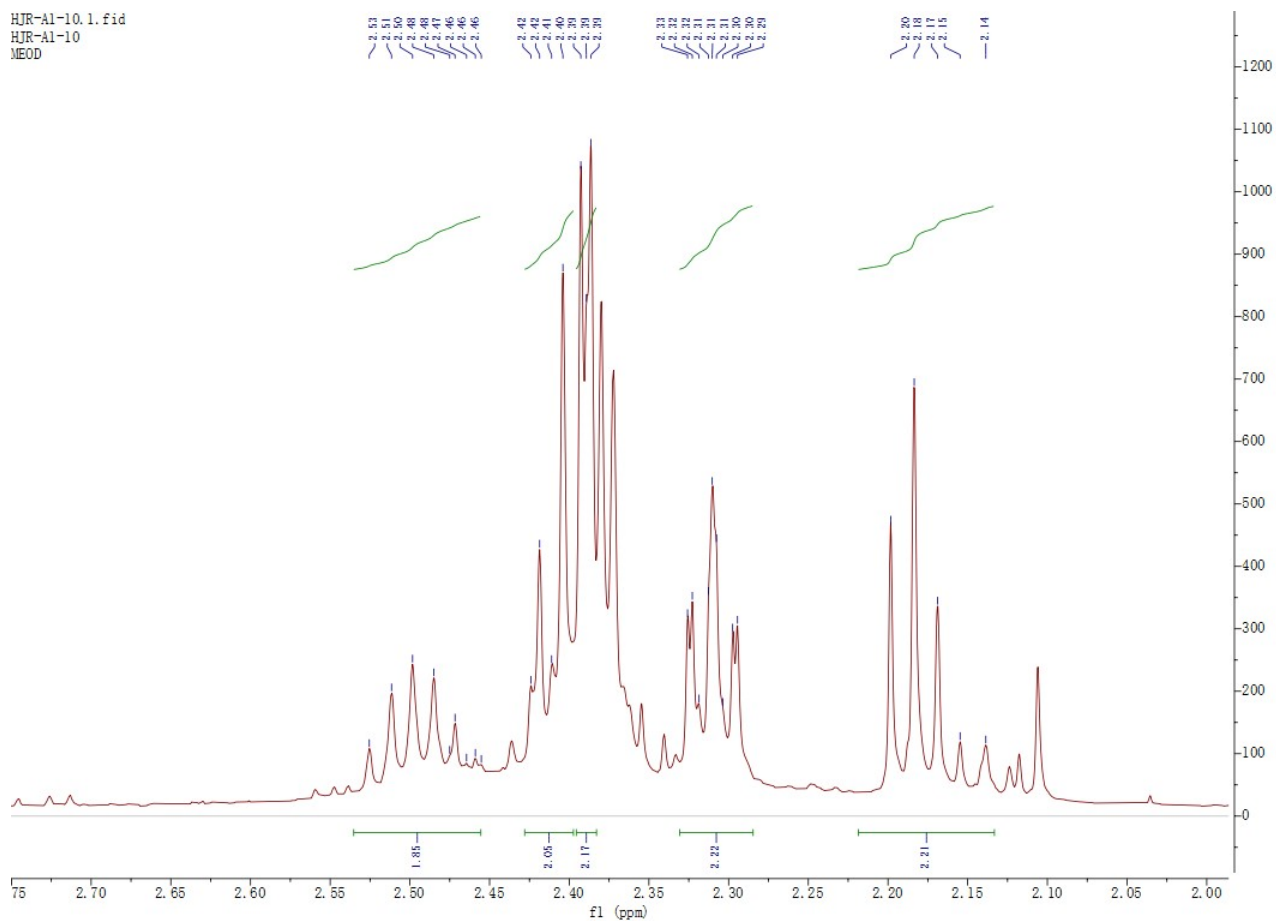


Figure S9 ^1H NMR of expanded regions at 2–2.7 ppm.

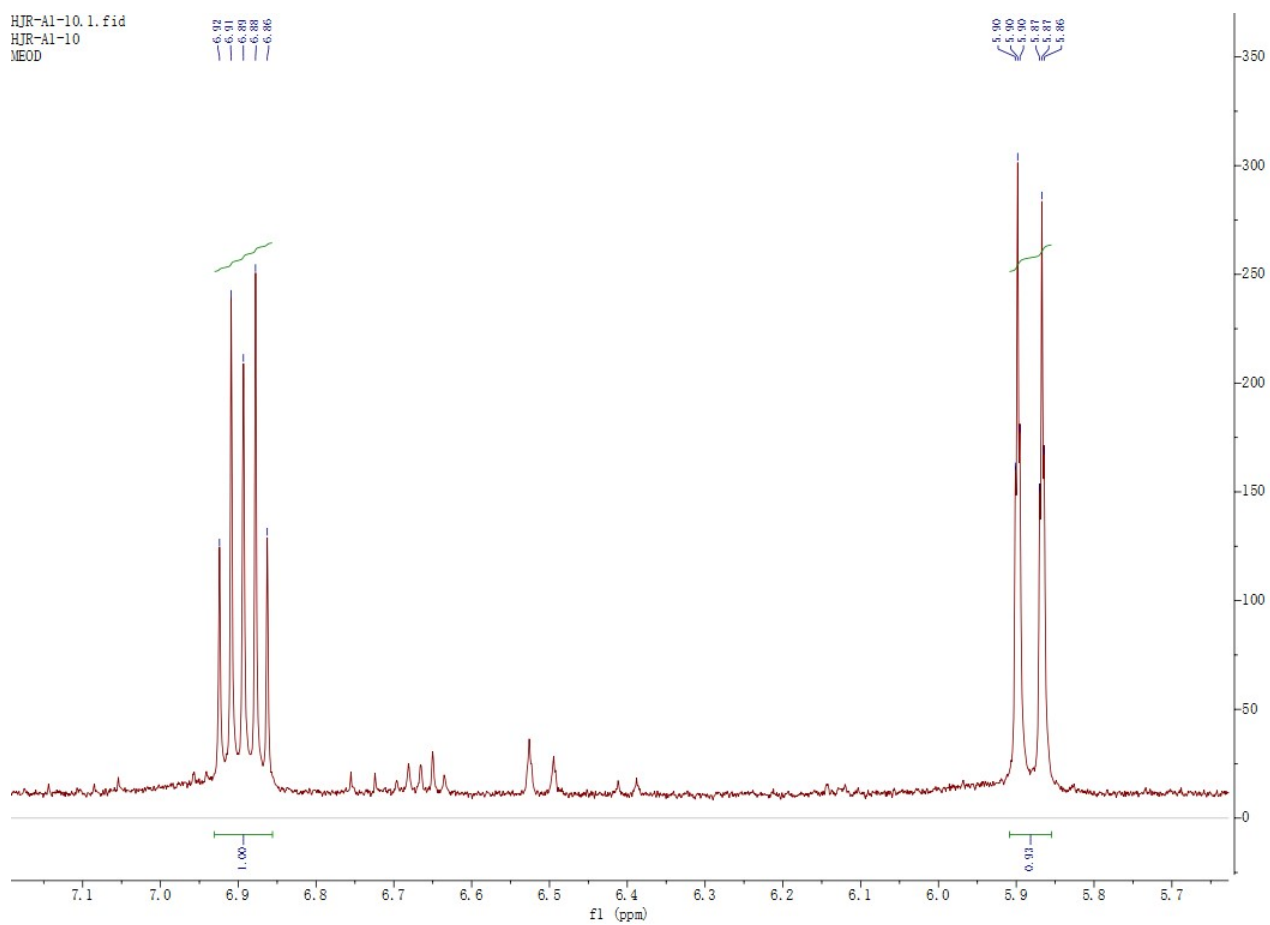


Figure S10 ^1H NMR of expanded regions at 5–7.1 ppm.

HJR-A1-10.3.fid
HJR-A1-10 C
MEOD

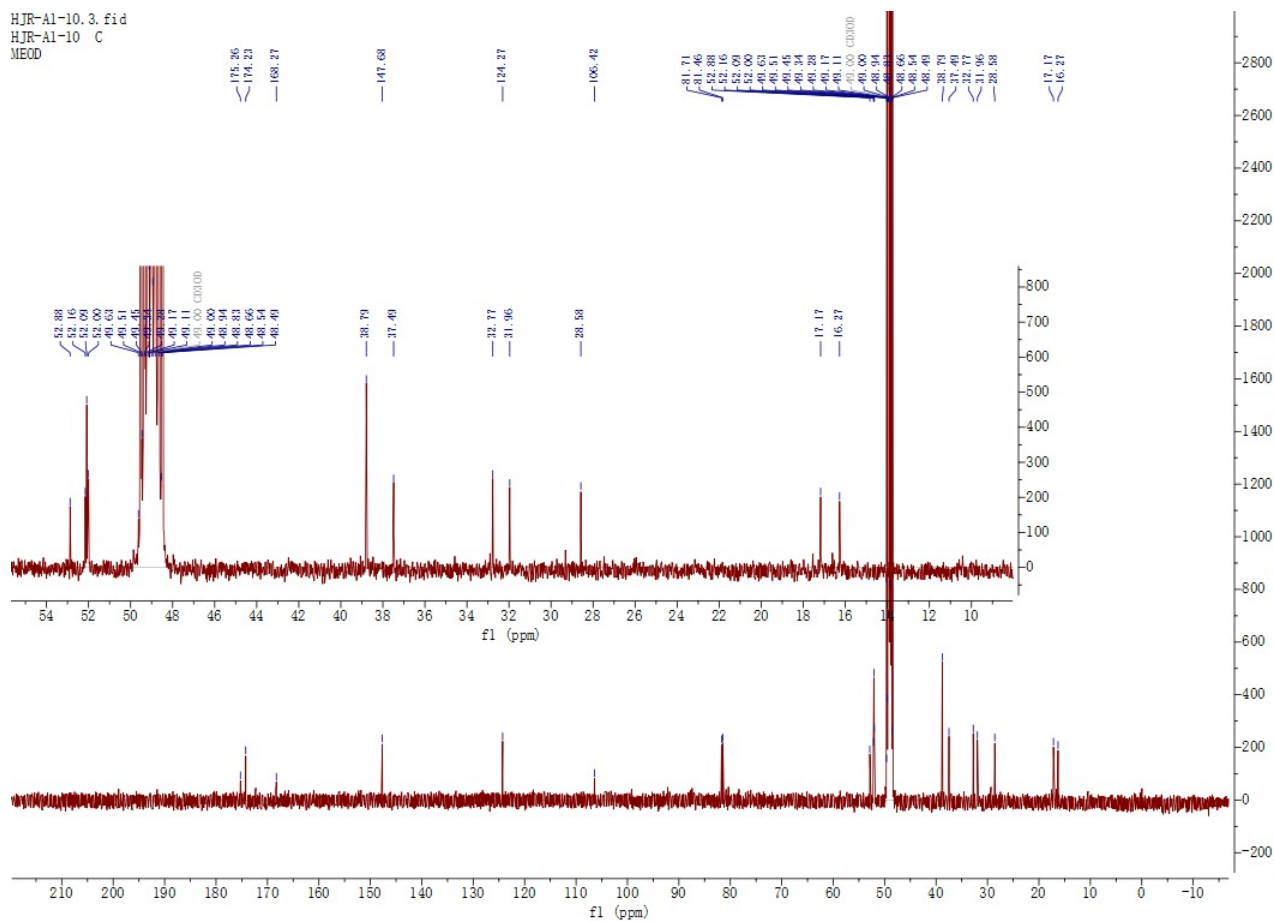


Figure S11 ¹³C spectrum of compound 1.

HJR-Al-10.4.fid
HJR-Al-10 dept135
meod

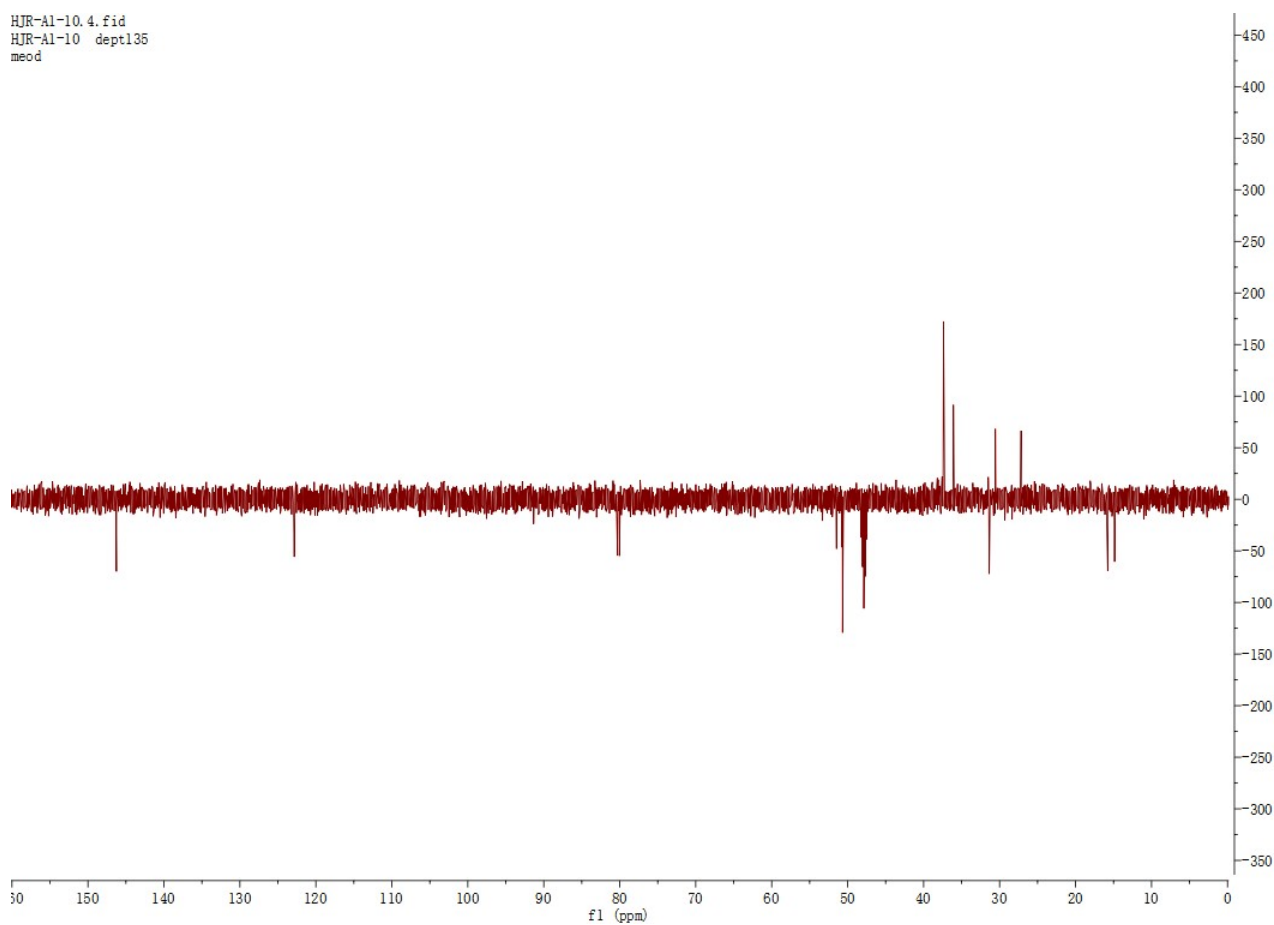


Figure S12 Dept 135 spectrum of compound **1**.

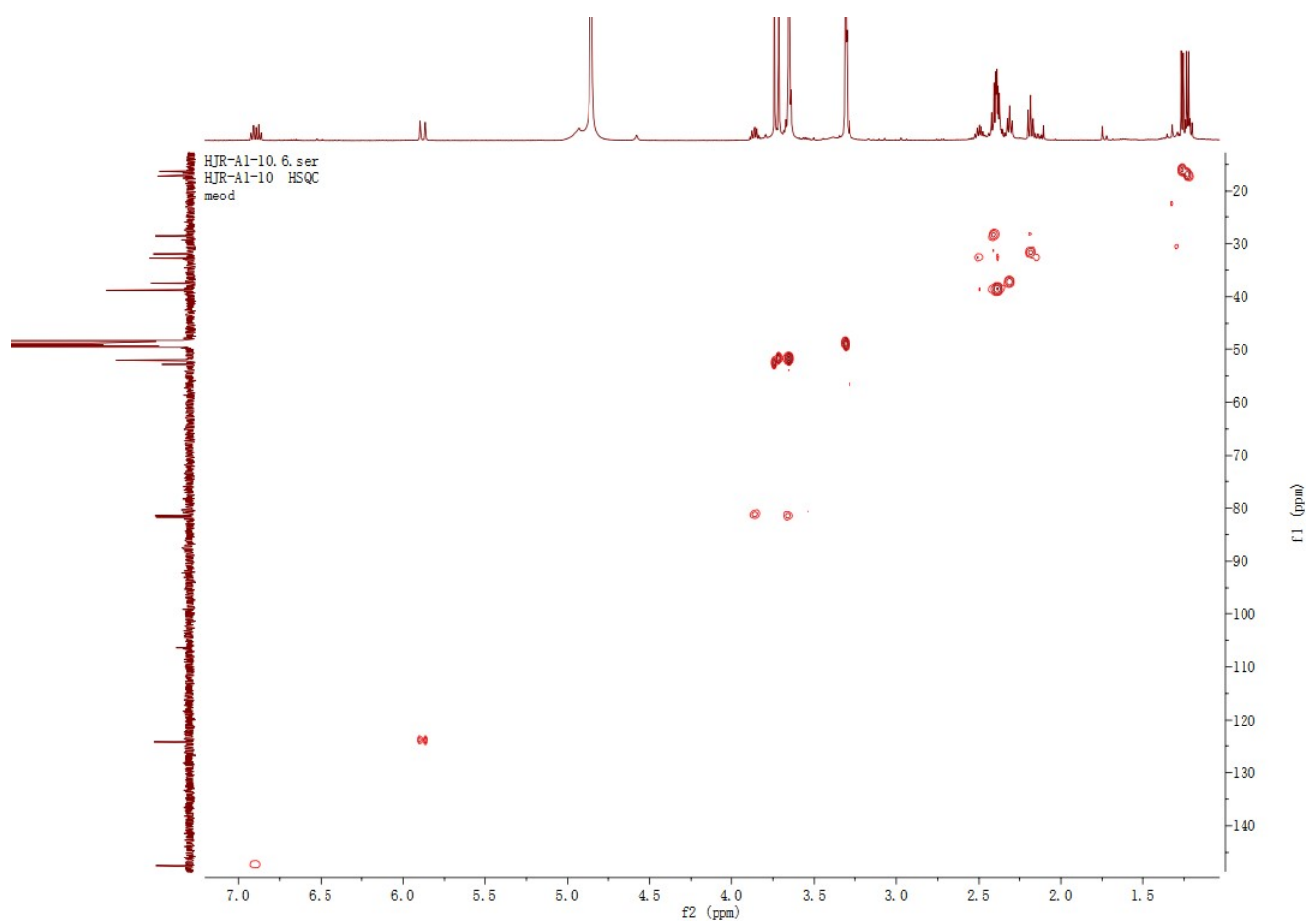


Figure S13 HSQC spectrum of compound **1**.

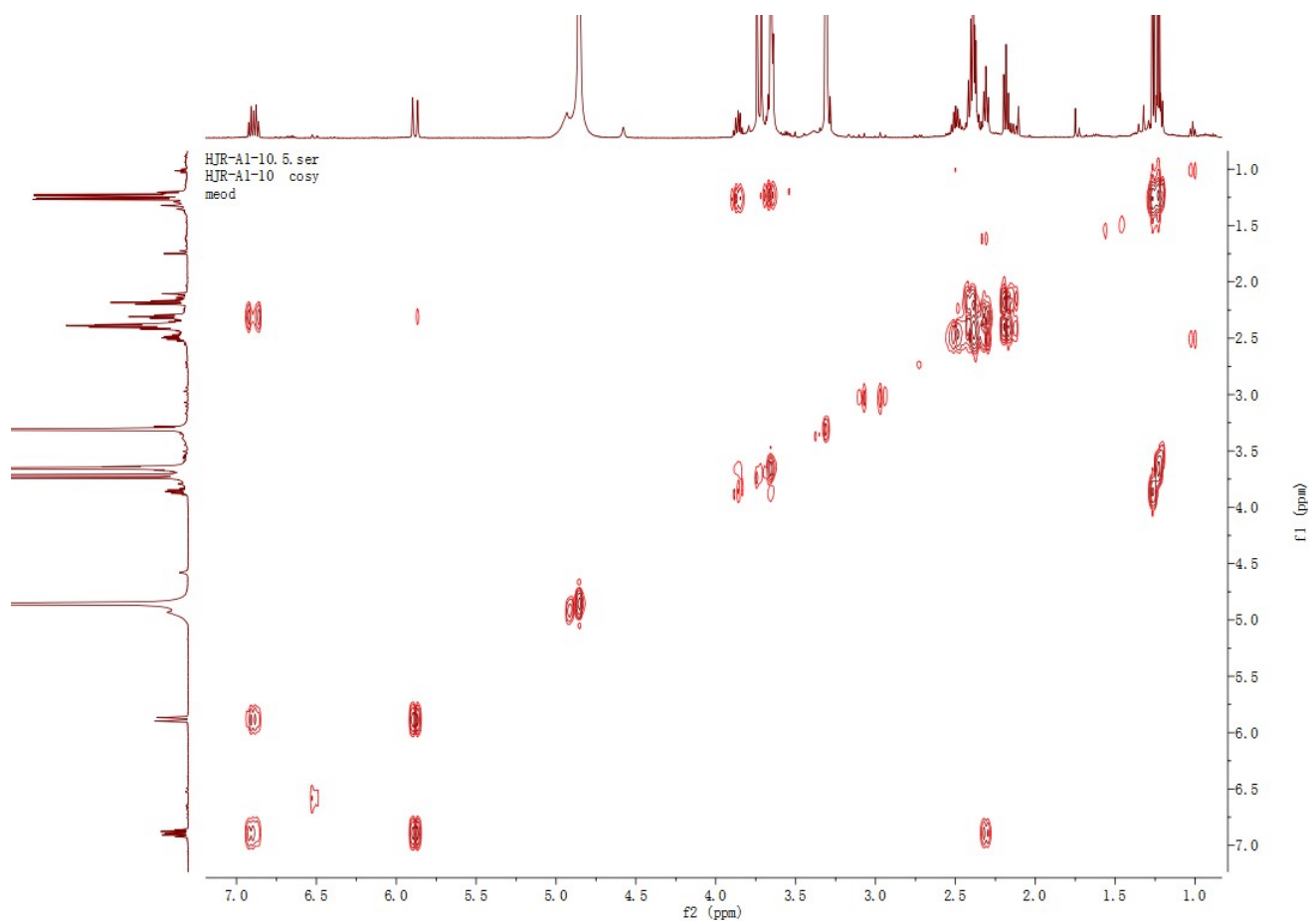


Figure S14 ^1H - ^1H cosy spectrum of compound **1**.

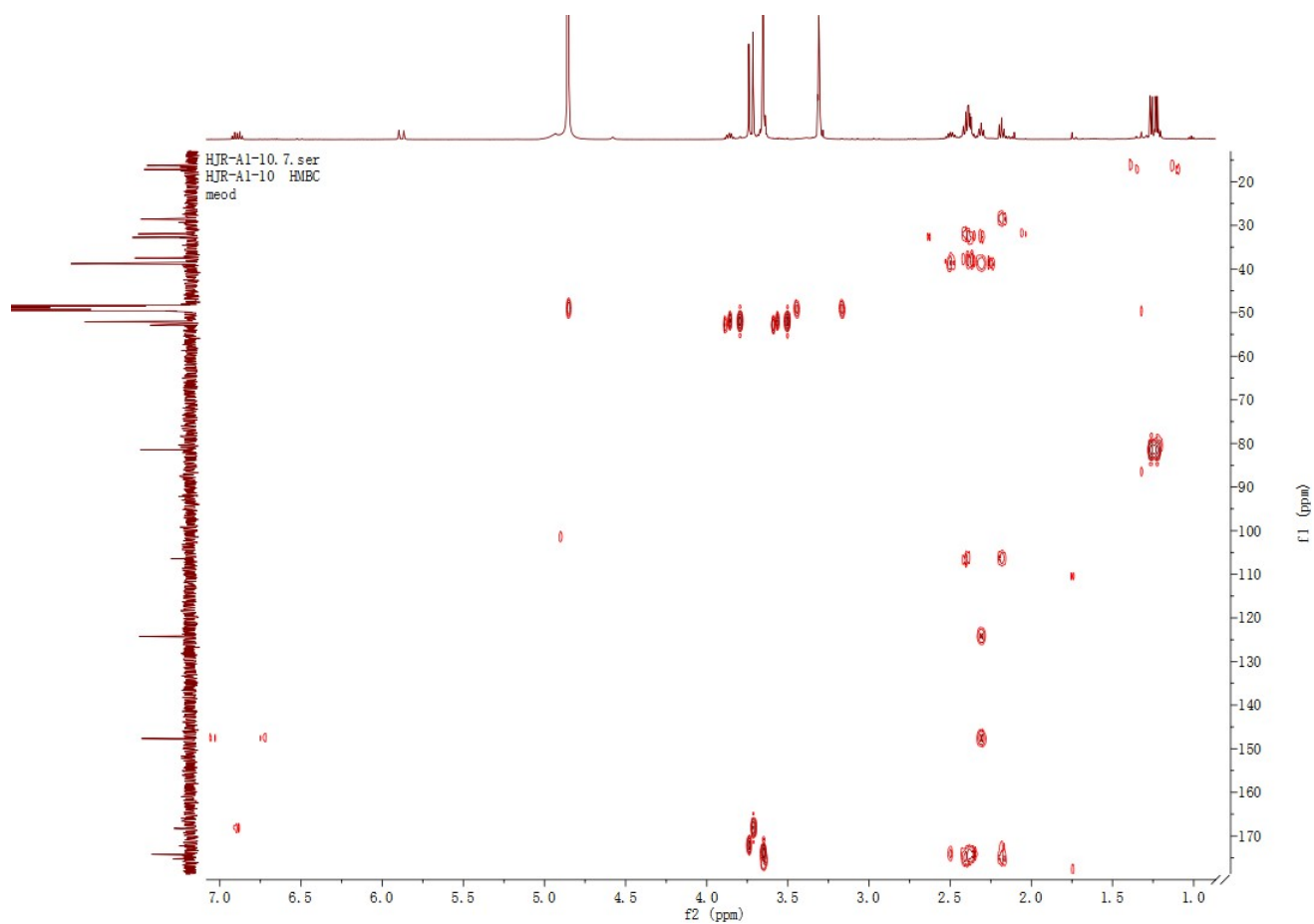


Figure S15 HMBC spectrum of compound **1**.