

## Supporting Information for

Production of Cytotoxic Glidobactins/Luminmycins by *Photorhabdus asymbiotica* in Liquid Media and Live Crickets

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**Table S1.** Summary of antiSMASH analysis of the *P. asymbiotica* ATCC43949 genome. Putative gene clusters for secondary metabolites and their genomic locations are shown. Cluster 15 was identified homologous the genes responsible for glidobactin biosynthesis. NRPS: non-ribosomal peptide synthase, T1PKS: type 1 polyketide synthase.

Gene Cluster	Type of cluster	Location (nt)
1	NRPS-T1PKS	487299-542304
2	NRPS-nucleoside	688894-740731
3	NRPS	968421-1031926
4	T1PKS	1023946-1075950
5	T1PKS	1352218-1399789
6	Phosphoglycolipid	143926-1481759
7	NRPS	1492345 - 1544792
8	NRPS	1645504 - 1695686
9	NRPS	1677807 - 1734065
10	NRPS	2014496 - 2059374
11	NRPS-T1PKS	2456784 - 2519934
12	NRPS-T1PKS	2593887 - 2661296
13	Other	2686922 - 2731935
14	NRPS	2899960 - 2946214
15	T1PKS-NRPS	3035604 - 3103761
16	NRPS	3491508 - 3558715
17	NRPS	3582829 - 3640789
18	NRPS	3915538 - 4000273
19	Butyrolactone	4366935 - 4374226

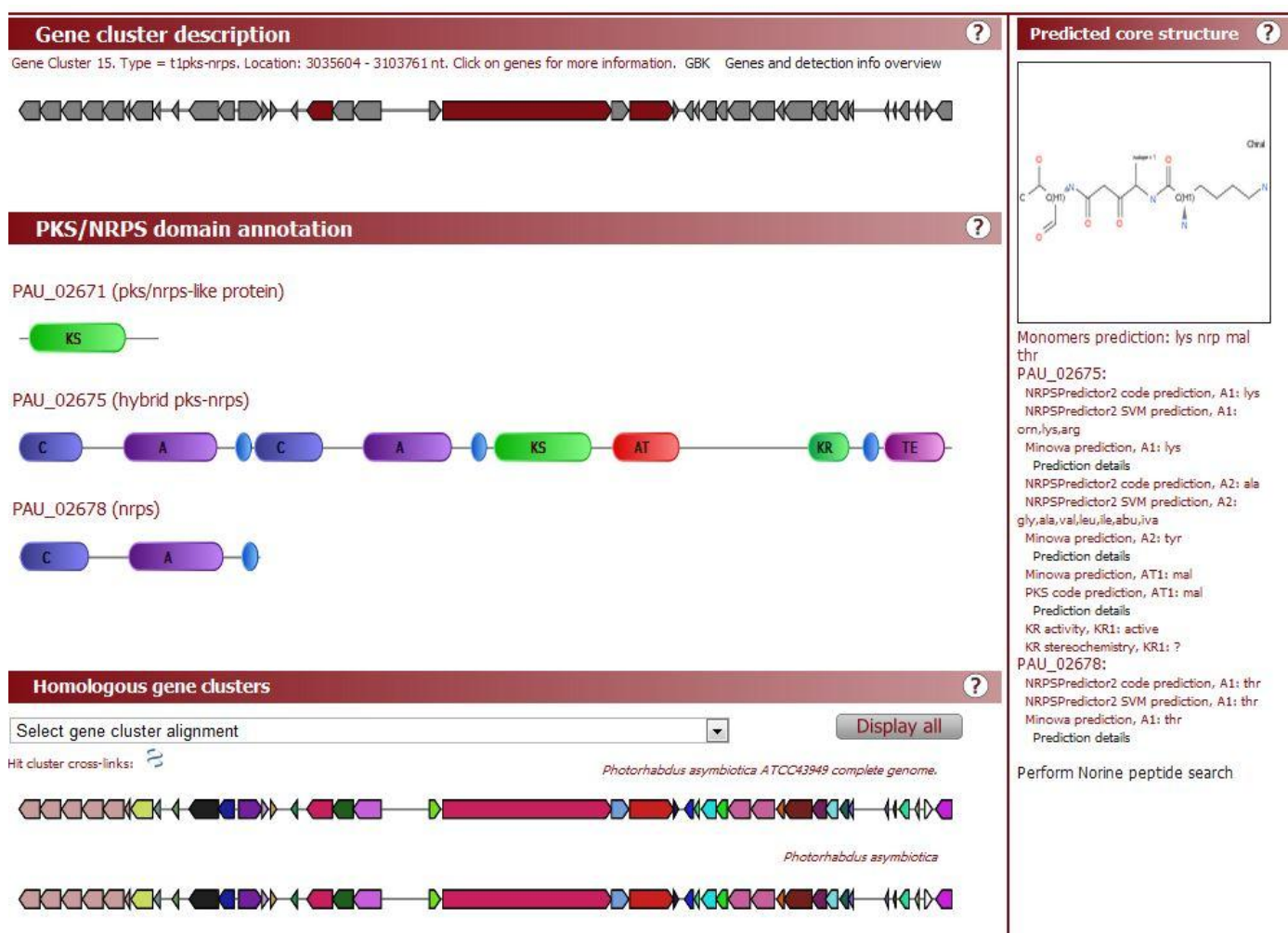
**Table S2.** Summary of media types tested to induce the production of glidobactin/luminmycins.

Name	Formulation	Compounds induced
TSB	Commercially available	-
TSB plus chitin	Commercially available mix at 75% strength with 7.5 g·L <sup>-1</sup> chitin	-
TSB plus soil	Commercially available mix with 10% v/v soil extract added prior to autoclaving	-
TSB plus blood	Commercially available mix with 5% v/v sterile sheep blood added after autoclaving	-
TSB plus bugs	Commercially available mix with 3 ground bugs collected in Norman, OK added to 300 mL of broth prior to autoclaving	-
TSB mimic, less sugar increased tryptone	In g·L <sup>-1</sup> : dextrose ( 1.7), tryptone (27), NaCl (5), K <sub>2</sub> HPO <sub>4</sub> (2.5)	-
TSB mimic, less tryptone, increased sugar	In g·L <sup>-1</sup> : dextrose ( 1.7), tryptone (27), NaCl (5), K <sub>2</sub> HPO <sub>4</sub> (2.5)	-
TSB mimic, glucose	In g·L <sup>-1</sup> : glucose (2.5), tryptone (20), NaCl (5), K <sub>2</sub> HPO <sub>4</sub> (2.5)	-
TSB mimic, ribose	In g·L <sup>-1</sup> : ribose (2.5), tryptone (20), NaCl (5), K <sub>2</sub> HPO <sub>4</sub> (2.5)	-
TSB mimic, sucrose	In g·L <sup>-1</sup> : sucrose (2.5), tryptone (20), NaCl (5), K <sub>2</sub> HPO <sub>4</sub> (2.5)	-
Actinomyces broth	Commercially available	-
Potato dextrose	Commercially available	<b>1</b>
Luria broth	Commercially available	<b>1</b>
Luria broth plus homogenized bugs	Commercially available mix with approximately 10 bugs collected in Norman, OK added to 300 mL of broth and homogenized with mechanical homogenizer prior to autoclaving	<b>1</b>
Defined medium <sup>a</sup>	In g·L <sup>-1</sup> : KH <sub>2</sub> PO <sub>4</sub> (2.0), NH <sub>4</sub> Cl (1.5), MgSO <sub>4</sub> ·7H <sub>2</sub> O (0.5), glycerol (10), myo-inositol (0.4), monosodium L-glutamate monohydrate (5.0), NaF (0.084), FeSO <sub>4</sub> ·7H <sub>2</sub> O (0.025), ZnSO <sub>4</sub> ·7H <sub>2</sub> O (0.01), CoCl <sub>2</sub> ·6H <sub>2</sub> O (0.01), CaCO <sub>3</sub> (0.25), <i>p</i> -aminobenzoate (0.001)	<b>1, 2, 3</b>
Defined medium, honey as carbon source	Same as defined medium but with 10g·L <sup>-1</sup> of honey in place of glycerol	<b>1</b> , (trace amounts of <b>2</b> and <b>3</b> )
Defined medium, sucrose as carbon source	Same as defined medium but with 10g·L <sup>-1</sup> of sucrose in place of glycerol	<b>1</b> , (trace amounts of <b>2</b> and <b>3</b> )
Defined medium, glucose as carbon source	Same as defined medium but with 10g·L <sup>-1</sup> of glucose in place of glycerol	<b>1, 2, 3</b>
Defined medium, 10% v/v	Same as defined medium, with the addition of	<b>1, 2, 3</b>

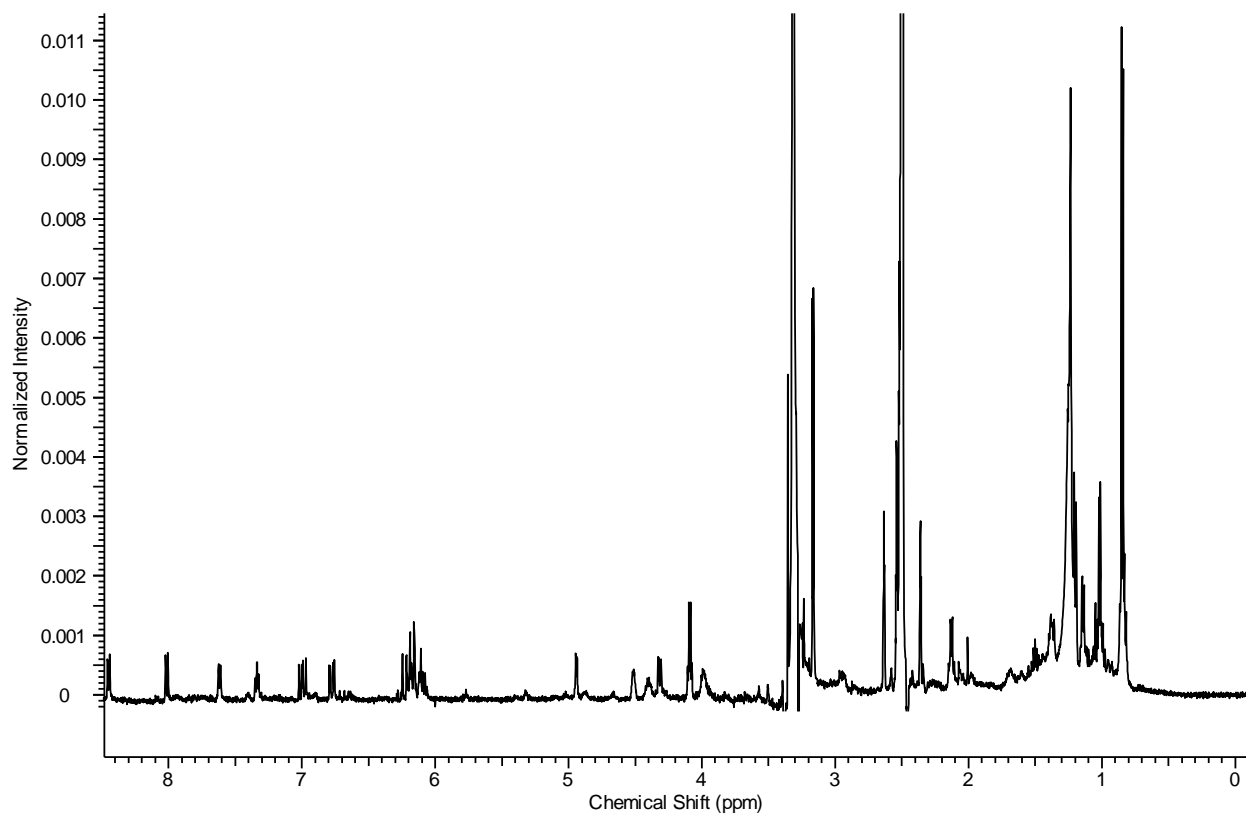
sheep blood	10% v/v of sterile sheep blood added after autoclaving	
0.25x seawater	In g·L <sup>-1</sup> : NH <sub>4</sub> Cl (1.5), glycerol (10), monosodium L-glutamate monohydrate (5), instant ocean (8.8)	-
0.5x seawater	In g·L <sup>-1</sup> : NH <sub>4</sub> Cl (1.5), glycerol (10), monosodium L-glutamate monohydrate (5), instant ocean (17.5)	-
1x seawater	In g·L <sup>-1</sup> : NH <sub>4</sub> Cl (1.5), glycerol (10), monosodium L-glutamate monohydrate (5), instant ocean (35.0)	-
Dulbecco's modified Eagle Medium (DMEM)	Sterile filtered, commercially available	<b>1</b>
DMEM plus 5% fetal bovine serum (FBS)	Sterile, filtered, commercially available DMEM and FBS added at 5% v/v	-
DMEM plus glycerol	In g·L <sup>-1</sup> : glycerol (10). Autoclaved and mixed with sterile, filtered, commercially available DMEM	<b>1</b>
DMEM plus glycerol and 5% FBS	In g·L <sup>-1</sup> : glycerol (10). Autoclaved and mixed with sterile, filtered, commercially available DMEM and 5% v/v FBS	-
Plant food plus glycerol	In g·L <sup>-1</sup> : plant food, commercially available water soluble (1.6), glycerol (10)	<b>1</b>
Plant food plus sucrose	In g·L <sup>-1</sup> : plant food, commercially available water soluble (1.6), sucrose (10)	<b>1</b>
Plant food plus glucose	In g·L <sup>-1</sup> : plant food, commercially available water soluble (1.6), glucose (10)	<b>1</b>
Plant food plus honey	In g·L <sup>-1</sup> : plant food, commercially available water soluble (1.6), honey (10)	<b>1</b>
Plant food plus cholesterol	In g·L <sup>-1</sup> : plant food, commercially available water soluble (1.6), cholesterol (10)	-
Skim milk	In g·L <sup>-1</sup> : skim milk powder (10)	-
Skim milk plus glycerol	In g·L <sup>-1</sup> : skim milk powder (10), glycerol (10)	-

Compound **1** : Glidobactin A; Compound **2**: Luminmycin A; Compound **3**: Luminmycin D;

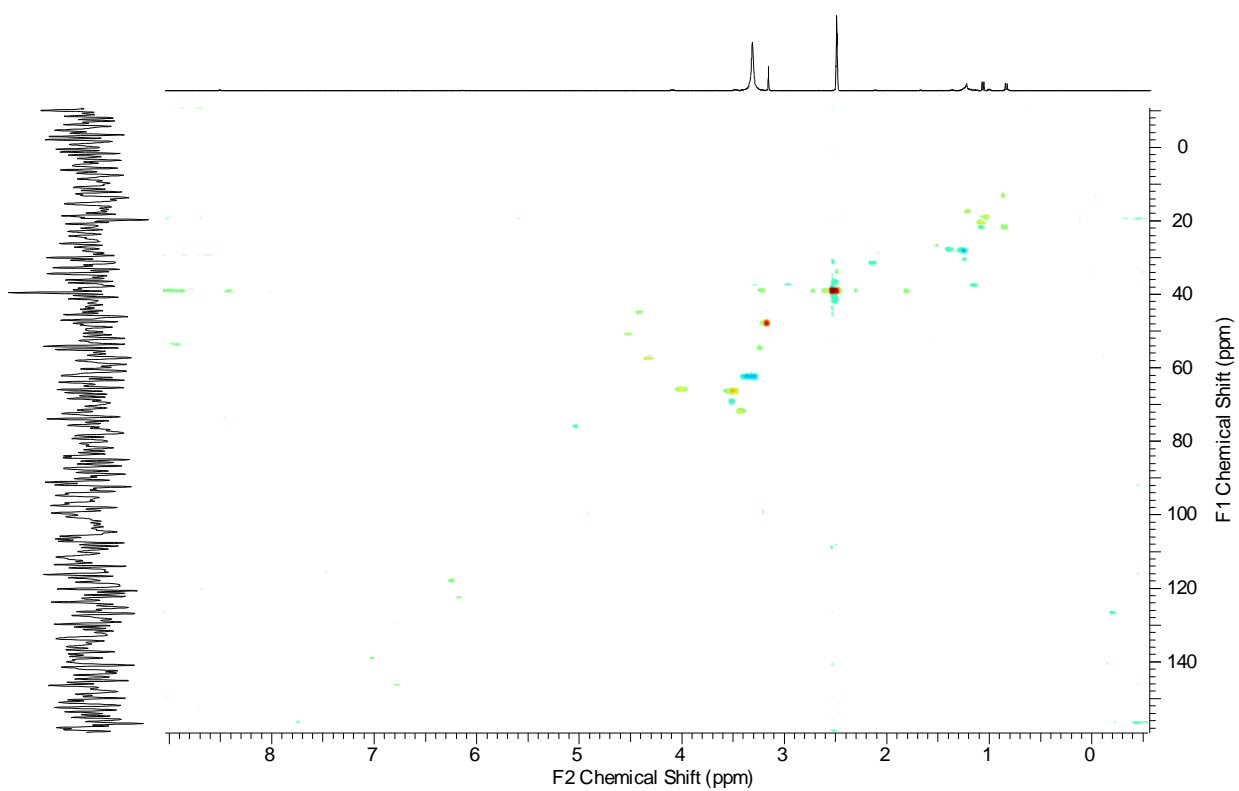
<sup>a</sup> Defined medium described by Rateb *et al. J. Nat. Prod.* **2011**, *74*, 1965-1971.



**Figure S1.** Screen shot of antiSMASH results for secondary metabolite gene cluster 15; the cluster predicted to produce glidobactin-like compounds. KS: ketosynthase, A: AMP binding domain, C: condensation domain, AT: acyl transferase, KR: ketoreductase, TE: thioesterase.

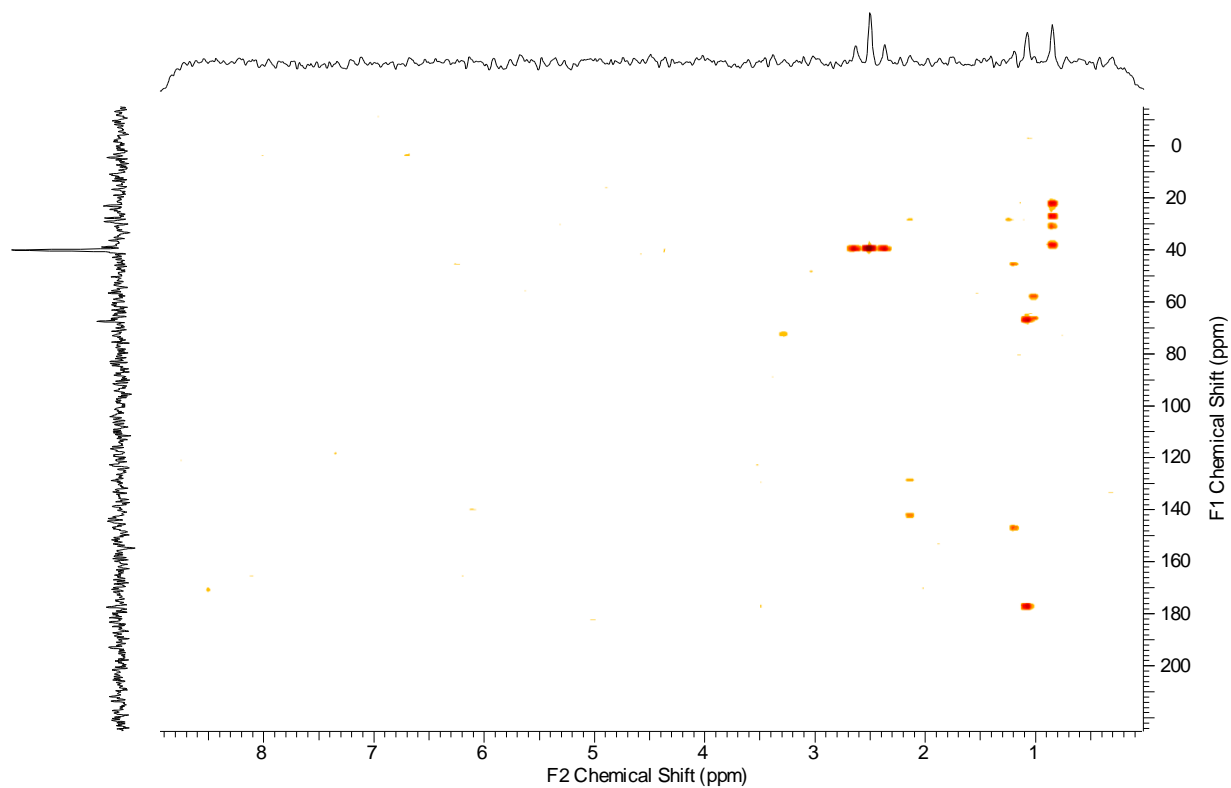


**Figure S2.**  $^1\text{H-NMR}$  (500 MHz,  $\text{DMSO-}d_6$ , 25  $^\circ\text{C}$ ) spectrum of compound **3**.

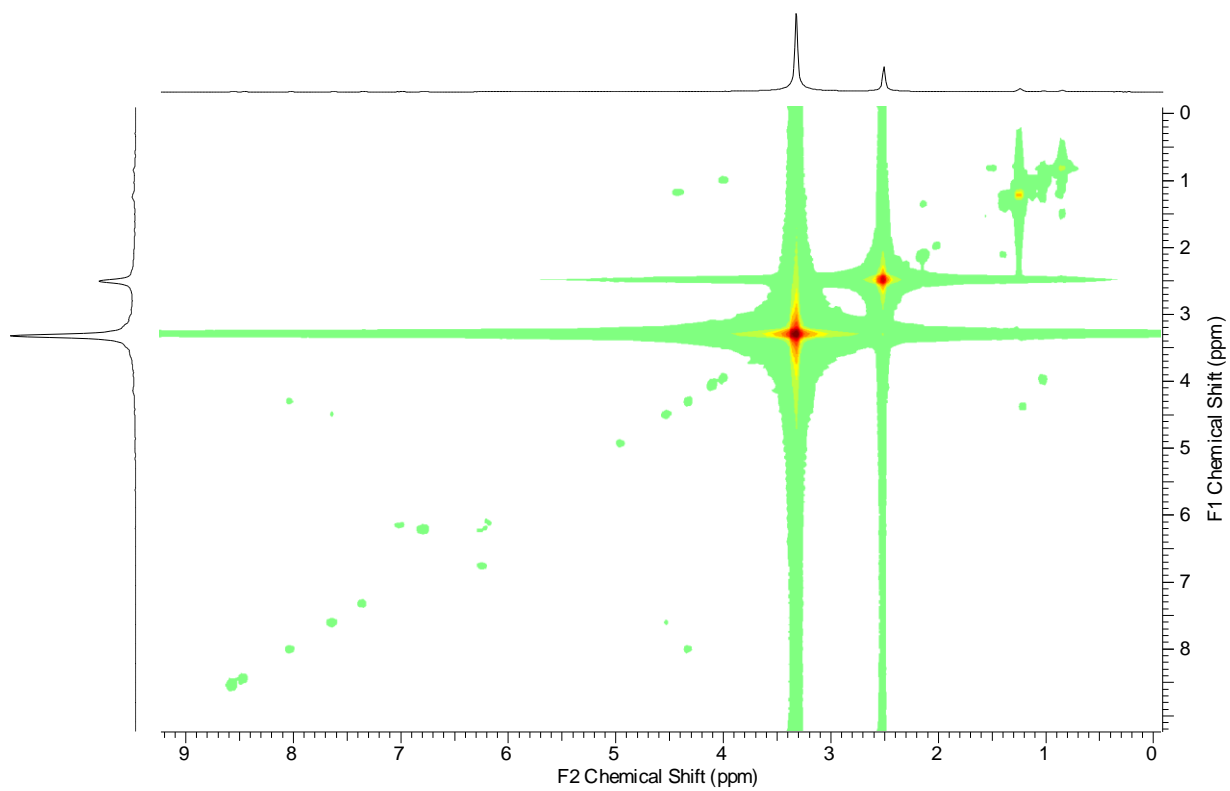


**Figure S3.**  $^1\text{H}$ - $^{13}\text{C}$  HSQC (500 MHz,  $\text{DMSO-}d_6$ , 25  $^\circ\text{C}$ ) spectrum of compound **3**.

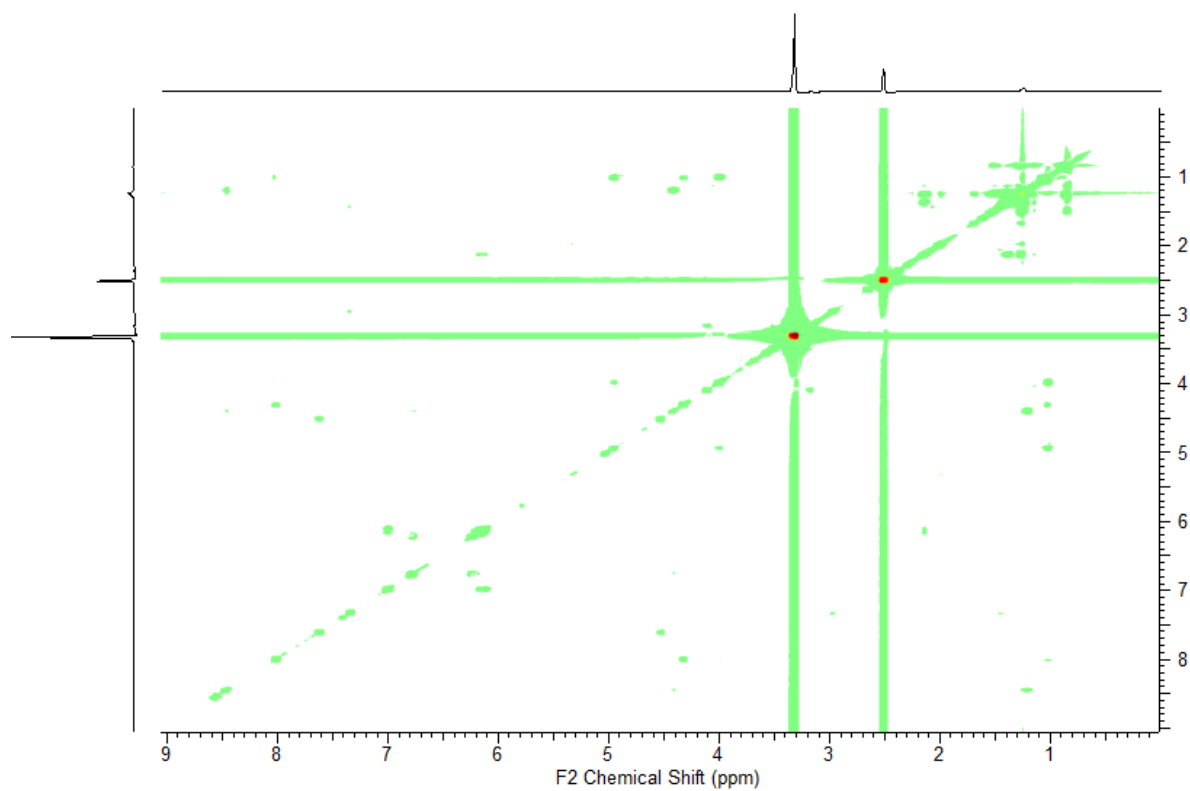




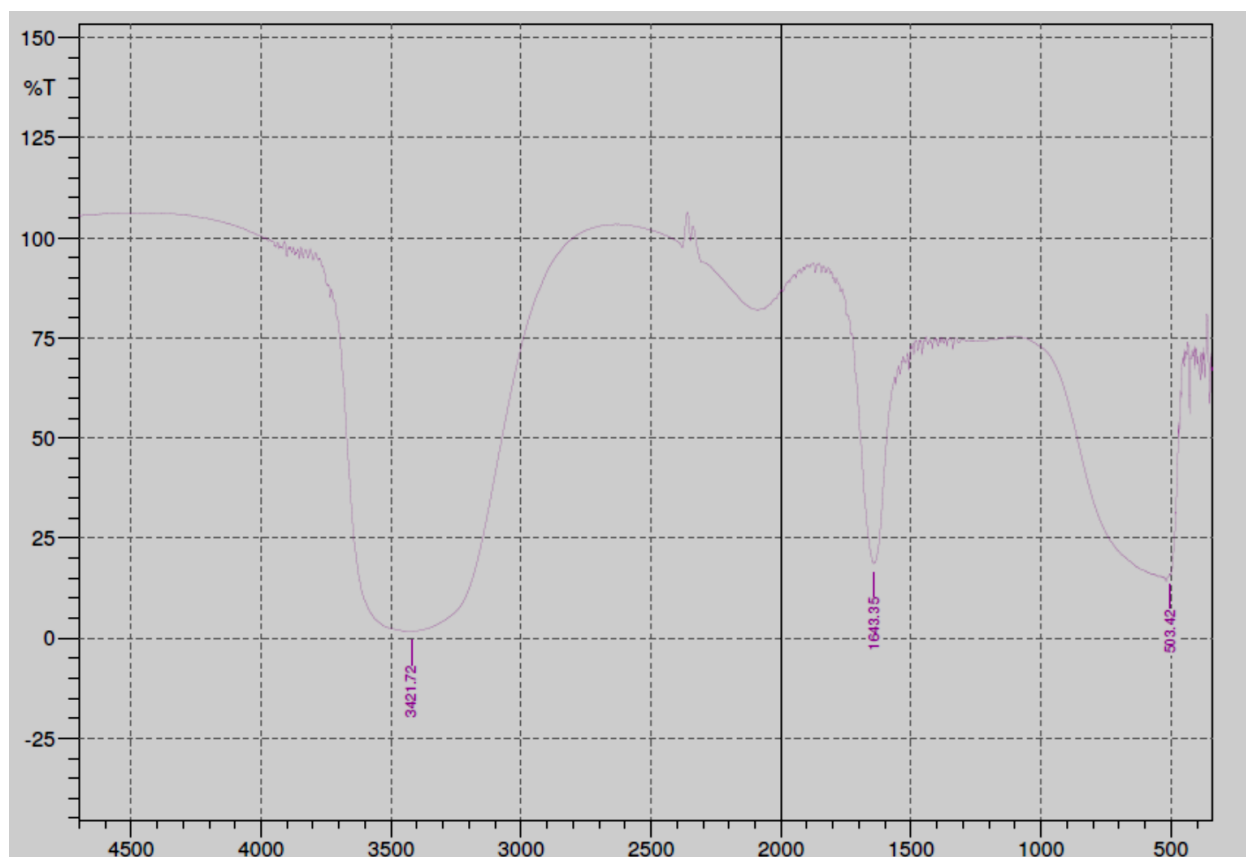
**Figure S4.**  $^1\text{H}$ - $^{13}\text{C}$  gHMBC (500 MHz,  $\text{DMSO-}d_6$ , 25  $^\circ\text{C}$ ) spectrum of compound **3**.



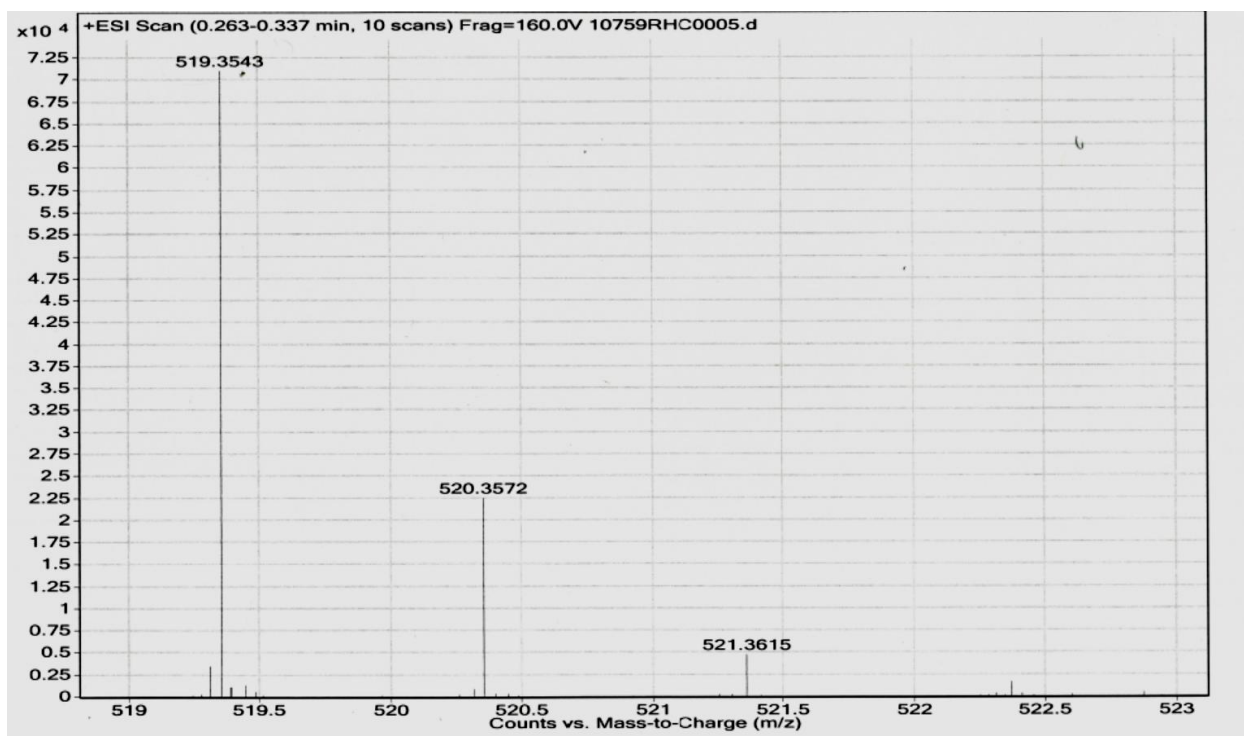
**Figure S5.**  $^1\text{H}$ - $^1\text{H}$  gCOSY (500 MHz,  $\text{DMSO-}d_6$ , 25  $^\circ\text{C}$ ) spectrum of compound **3**.



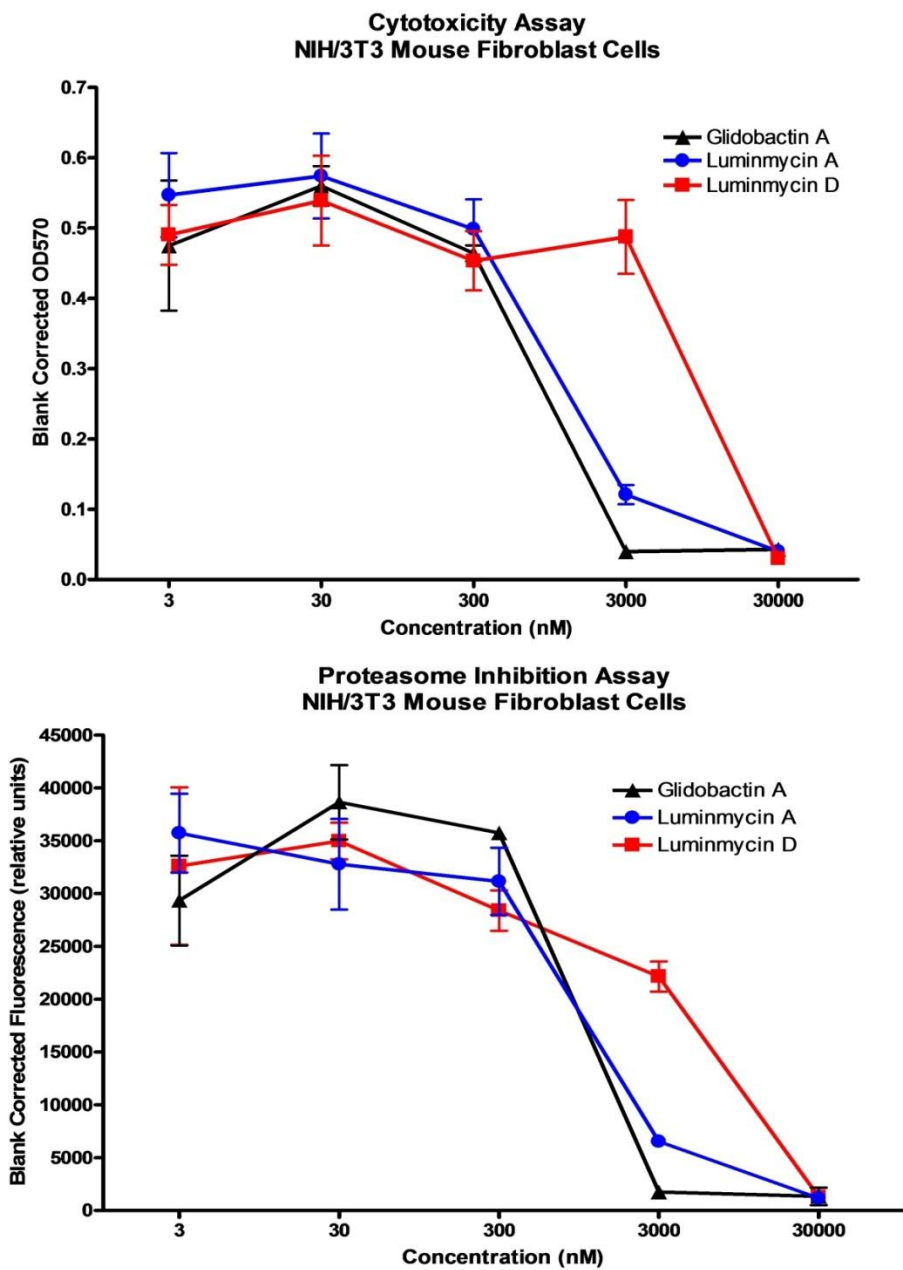
**Figure S6.** <sup>1</sup>H-<sup>1</sup>H tCOSY (500 MHz, DMSO-*d*<sub>6</sub>, 25 °C) spectrum of compound **3**.



**Figure S7.** IR spectrum (thin film, room temperature) of compound **3**.



**Figure S8.** HRESIMS (positive mode) data for compound **3**.



**Figure S9.** *In vitro* cytotoxicity (upper panel) and proteasome inhibition (lower panel) for **1**, **2**, and **3** in normal mouse fibroblast cells.