Supporting Information

The Glycopeptide Antitumor Antibiotic Zorbamycin from *Streptomyces flavoviridis* ATCC21892: Strain Improvement and Structure Elucidation[⊥]

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[⊥] Dedicated to the late Dr. Kenneth L. Rinehart, of the University of Illinois at Urbana-Champaign, for his pioneering work on bioactive natural products.

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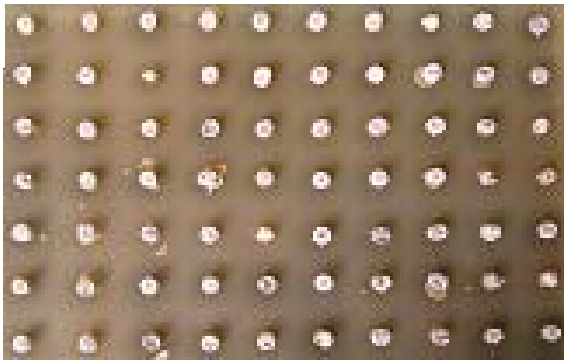
Table S1. UV-irradiation Time and *S. flavoviridis* ATCC21892 Spore Viability.

A spore stock solution (1 mL with ~ 10^3 spores) of *S. flavoviridis* ATCC21892 was exposed to UV irradiation (254 nm) at a distance of 12 cm for different time periods according to a standard literature protocol.[1] The colonized spores (living spores) were counted after incubation at 30 ^{o}C for 10 days. The most appropriate irradiation time was determined to be 4 to 5 min by the viability rate of 0.1% to 1%.

irradiation time (min)	colony number	viability (%)
0	1000	100
0.5	1000	100
1	700	70
2	140	14
3	19	1.9
4	8	0.8
5	4	0.4
10	0	0

Figure S1. Screening for high-producing strains of **1** upon UV irradiation of the wild-type *S. flavoviridis* ATCC21892 strain by bioassays against *M. smegmatis*. (I). Colonies showing different spore colors and morphology after UV irradiation were picked and inoculated into 96-well plates on agar medium (1.5% glucose, 1.5% starch, 2.0% soybean meal, 0.5% yeast extract, 0.25% NaCl, 0.32% $CaCO_3$, 0.0005% $CuSO_4 \cdot 5H_2O$, 0.0005% $MnCl_2 \cdot 4H_2O$, 0.005% $ZnSO_4 \cdot 7H_2O$, pH 7.4, and 0.7% agar) and then incubated for 8 days. (II) Each of the agar blocks from the 96-well plates was placed on a bioassay plate pre-seeded with *M. smegmatis* to screen for high-producers of **1** with the wild-type *S. flavoviridis* ATCC21892 strain as a control. (1). The wild-type *S flavoviridis* ATCC21892 strain and (2) the improved *S. flavoviridis* SB9000 strain.

I



II

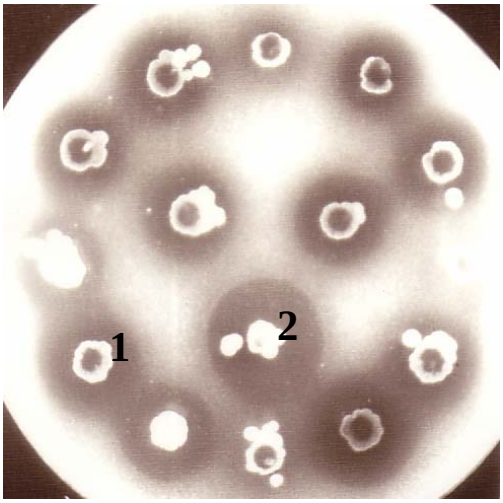
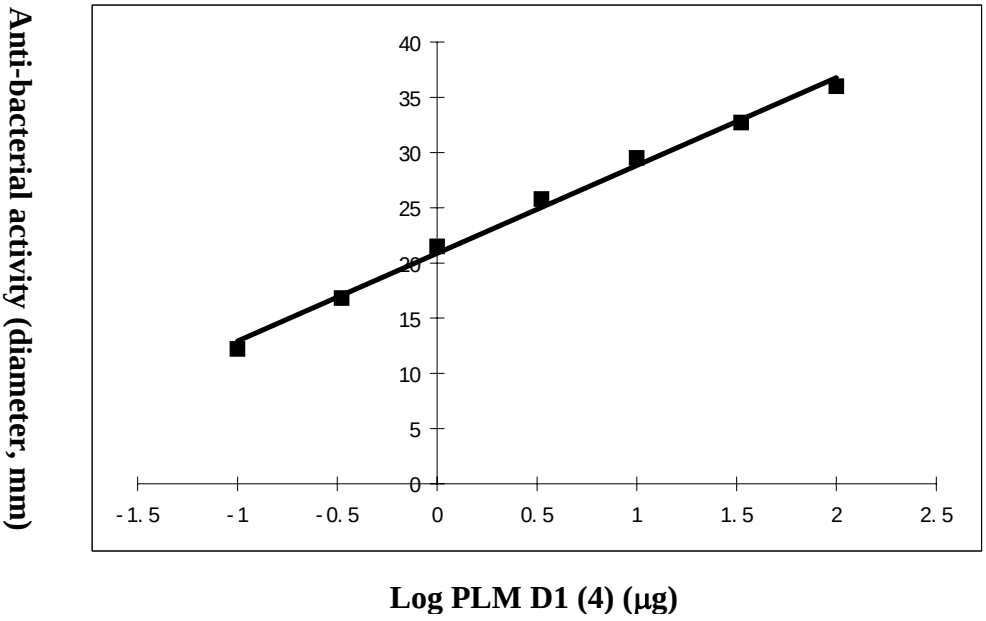


Figure S2. The standard curve for **4** against *M. smegmatis* by the paper disk bioassay.



References and Notes

(1) Kieser, T.; Bibb, M. J.; Buttner, M. J.; Chater, K. F.; Hopwood, D. A. *Practical Streptomyces Genetics;* The John Innes Foundation: Norwich, UK; 2000, pp 102-103.

(2) Okuda, T.; Awataguchi, S. Jpn. Tokkyo Koho, JP 47002557, 1972.

(3) The optimized medium for zorbamycin production was derived from the original production medium in reference 2 by (i) increasing the concentrations of $CuSO_4 \cdot 5H_2O$ from 0.0005% to 0.01% and of $ZnSO_4 \cdot 7H_2O$ from 0.005% to 0.05% and (ii) eliminating $MnCl_2 \cdot 7H_2O$ completely.