

Supplementary Information

Identification and Characterisation of the Nybomycin Gene Cluster from the Marine Strain *Streptomyces albus* subsp. *chlorinus* NRRL B-24108

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Tables

Table S1. ¹H-NMR (500 MHz, d-TFA) data for isolated and standard nybomycin.

| Isolated nybomycin | | Standard nybomycin | |
|--------------------|---------------------|--------------------|---------------------|
| Position | δ_{H} | Position | δ_{H} |
| 2 | 6.77 s | 2 | 6.77 s |
| 5 | 7.09 s | 5 | 7.09 s |
| 6' | 2.81 s | 6' | 2.81 s |
| 7 | 7.67 s | 7 | 7.67 s |
| 8' | 5.52 s | 8' | 5.52 s |
| 9 | 8.10 s | 9 | 8.10 s |
| 11' | 4.47 s | 11' | 4.47 s |

Table S2. Bacterial strains and BACs used in this work.

| Bacterial strain | Features | Reference/Source |
|--|---|-------------------------|
| <i>Streptomyces albus</i> subsp. <i>chlorinus</i> NRRL B-24108 | <i>S. albus</i> subspecies strain harboring nybomycin biosynthetic gene cluster | [1] |
| <i>Streptomyces albus</i> Del14 | Wild-type strain | [2] |
| <i>Streptomyces albus</i> 4N24 | <i>S. albus</i> strain with BAC 4N24 insertion | This work |
| <i>Streptomyces albus</i> 4M14 | <i>S. albus</i> strain with BAC 4M14 insertion | This work |
| <i>Streptomyces albus</i> 6M11 | <i>S. albus</i> strain with BAC 6M11 insertion | This work |
| <i>Streptomyces lividans</i> TK24 | Wild-type strain | [3] |
| <i>Streptomyces lividans</i> 4N24 | <i>S. lividans</i> strain with BAC 4N24 insertion | This work |
| <i>Escherichia coli</i> ET12567 pUB307 | Donor strain for intergeneric conjugation | [4] |
| <i>Escherichia coli</i> DH10 β | General cloning strain | [5] |
| BACs | | |
| pSMART | AmR; BAC vector | Lucigen (USA) |
| 4N24/4M14/6M11 | BACs containing full or partial nybomycin gene cluster | This work |

Figures

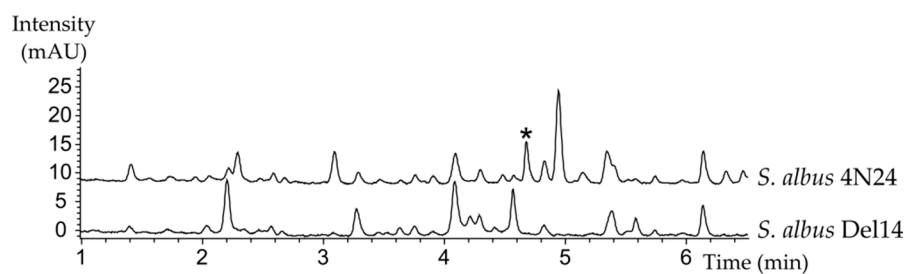


Figure S1. UV chromatograms of crude extracts from *S. albus* 4N24 and *S. albus* Del14. The new peak found in *S. albus* 4N24 crude extract is indicated with an asterisk (*). The profiles correspond to wavelength 285 nm.

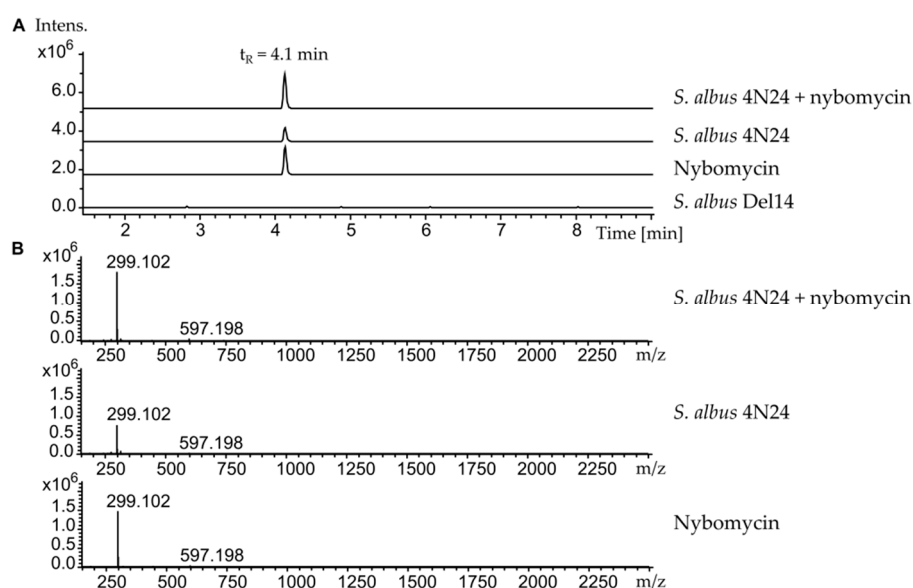


Figure S2. Comparison of LC-MS chromatograms at $t_R = 4.1$ min of *Streptomyces albus* 4N24 crude extract and a nybomycin standard. (A) Extracted ion chromatograms (299.10 ± 0.1 Da) of crude extract from *S. albus* 4N24 supplemented with 0.05 mg/ml of pure nybomycin, crude extract from *S. albus* 4N24 broth culture, a 0.05 mg/ml nybomycin solution in methanol, and crude extract from *S. albus* Del14 broth culture. (B) Mass spectra associated to the peak at $t_R = 4.1$ min from the three upper chromatograms displayed in (A).

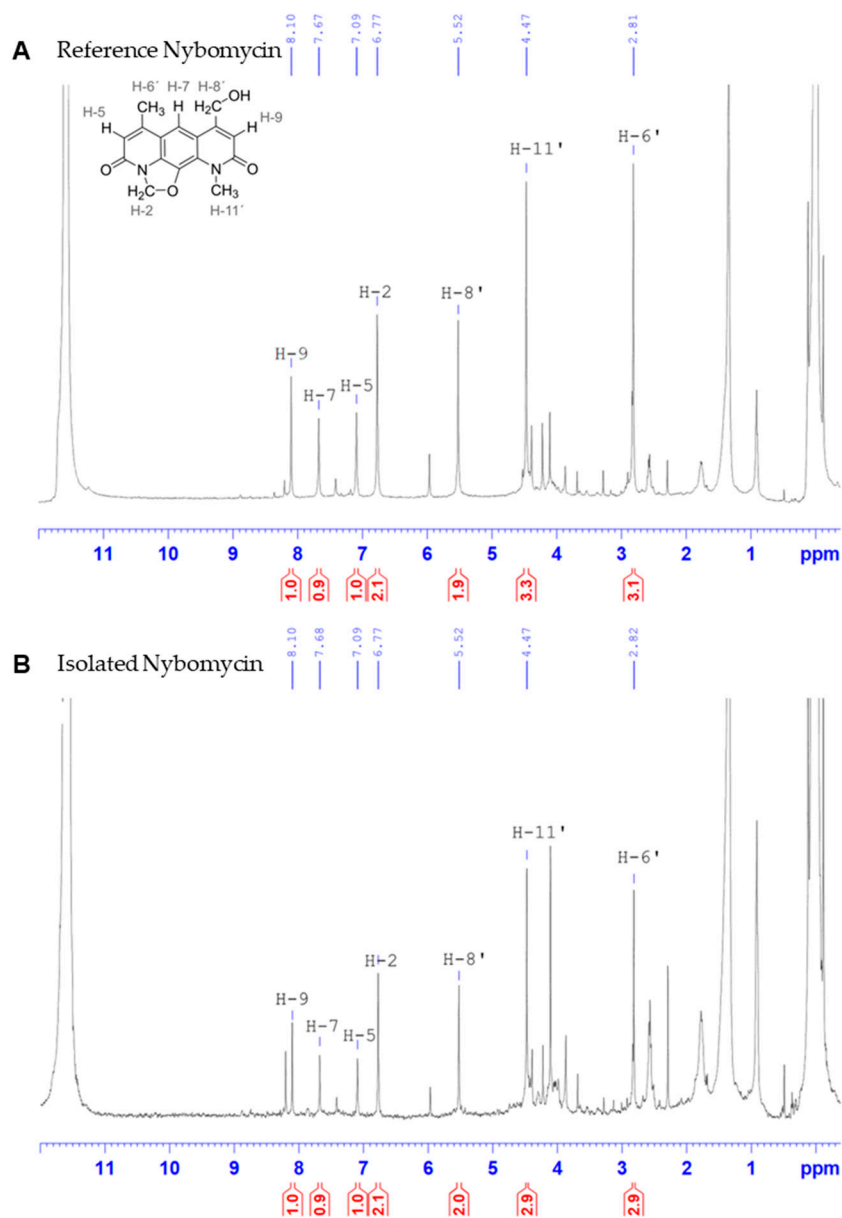


Figure S3. $^1\text{H-NMR}$ (500 MHz, d-TFA) spectra of a nybomycin standard and nybomycin isolated from *S. albus* 4N24. $^1\text{H-NMR}$ spectra of nybomycin standard (A) and the isolated nybomycin (B) are identical.

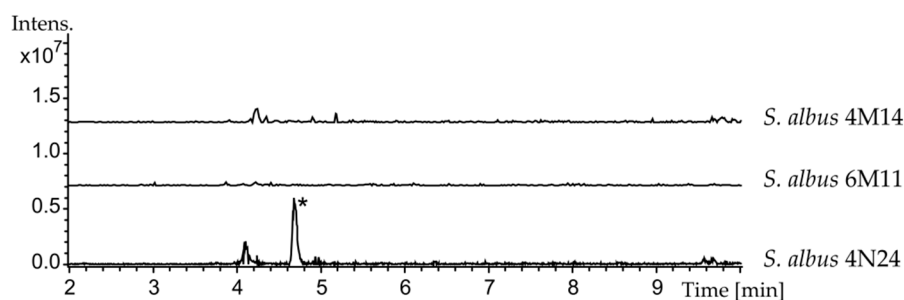


Figure S4. Extracted ion chromatograms of *S. albus* 4M14, *S. albus* 6M11, and *S. albus* 4N24 crude extracts. Nybomycin appears only in *S. albus* 4N24 crude extract, which shows a peak at $t_R = 4.7$ min (indicated with an asterisk) comprising an $[M+H]^+$ of 299.102 m/z (mass spectrum not shown).

References

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