

Table S1. Details of the *Nocardia* isolates.

Local Isolates/ID	Closest Relative ^a	Percent Identity (%)	Phage Susceptibility ^b	Collection Location ^c	Month of Collection
USC-21006	MH261196.1 <i>Nocardia nova</i> strain MGA115	98.44%	–	Mooloolaba	February
USC-21010	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.78%	–	Mooloolaba	February
USC-21011	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.70%	–	Mooloolaba	February
USC-21012	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.71%	–	Mooloolaba	February
USC-21016	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.71%	–	Cotton Tree	March
USC-21017	MH261196.1 <i>Nocardia nova</i> strain MGA115	98.43%	–	Cotton Tree	March
USC-21018	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.57%	±	Cotton Tree	March
USC-21021	KJ571085.1 <i>Nocardia grenadensis</i> strain T7-8	97.73%	++	Cotton Tree	March
USC-21022	MH261196.1 <i>Nocardia nova</i> strain MGA115	97.78%	–	Cotton Tree	March
USC-21024 DSM 111829*	MH261196.1 <i>Nocardia nova</i> strain MGA115	98.97%	–	Mooloolaba	February
USC-21025	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.42%	–	Mooloolaba	February
USC-21026	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.71%	–	Mooloolaba	February
USC-21027	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.78%	–	Mooloolaba	February
USC-21028	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.63%	–	Mooloolaba	February
USC-21029	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.06%	–	Mooloolaba	February
USC-21030	MH261196.1 <i>Nocardia nova</i> strain MGA115	98.21%	–	Cotton Tree	March
USC-21032	MH261196.1 <i>Nocardia nova</i> strain MGA115	98.49%	–	Cotton Tree	March
USC-21034	MH261196.1 <i>Nocardia nova</i> strain MGA115	98.97%	–	Mooloolaba	February
USC-21035	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.56%	–	Mooloolaba	March
USC-21036	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.64%	–	Mooloolaba	February
USC-21037	KY454539.1 <i>Nocardia testacea</i> strain F-161197	98.15%	+++	Cotton Tree	March
USC-21038	MH261196.1 <i>Nocardia nova</i> strain MGA115	99.64%	–	Mooloolaba	February
USC-21039 DSM 111830*	KM978823.1 <i>Nocardia higoensis</i> strain A6	97.01%	–	Mooloolaba	February
USC-21040	MH261196.1 <i>Nocardia nova</i> strain MGA115	97.14%	–	Mooloolaba	October

USC-21042	NR_149226.1 <i>Nocardia rayongensis</i> strain RY45-3	96.79%	+++	Maroochydore	October
USC-21043	HM584914.1 <i>Nocardia nova</i> strain CBU 09/875	97.99%	–	Maroochydore	October
USC-21044 DSM 111630*	NR_117402 <i>Nocardia niigatensis</i> strain W8186	97.84%	++	Maroochydore	October
USC-21046 DSM 111727*	NR_109056.1 <i>Nocardia goodfellowii</i> strain A2012	93.03%	++	Maroochydore	October
USC-21047	MK302233.1 <i>Nocardia tengchongensis</i> strain JBRI-MO-0018	97.19%	–	Maroochydore	October
USC-21048 DSM 111726*	NR_117320.1 <i>Nocardia elegans</i>	96.50%	–	Maroochydore	October
USC-21049	MN620411.1 <i>Nocardia</i> sp. strain 19D1V24	97.26%	–	Maroochydore	October
USC-21050	NR_117322.1 <i>Nocardia flavorosea</i>	96.80%	–	Maroochydore	October

^aClosest relative based on 16s rRNA gene sequence similarity. ^b +++: Highly susceptible (complete lysis), ++: Susceptible (complete partial lysis), +: Moderately susceptible (lysis and single plaques), ±: Low susceptibility (Lysis but regrowth of the host), –: Not susceptible. ^c Marine/beach locations situated on the Sunshine Coast, QLD, Australia. *:Leibniz Institute DSMZ – German Collection of Micro-organisms and Cell Cultures (DSMZ) strain numbers for 5 deposited isolates which are publicly available.

Table S2. Antibiotic susceptibility testing results.

ZONE DIAMETER (mm) /ANTIBIOTIC SUSCEPTIBILITY									
*Interpretation threshold (mm) as described by Lebeaux, D., Bergeron, E., Berthet, J., Djadi-Prat, J., Mouniee, D., Boiron, P., Lortholary, O. and Rodriguez-Nava, V., 2019. Antibiotic susceptibility testing and species identification of <i>Nocardia</i> isolates: a retrospective analysis of data from a French expert laboratory, 2010–2015. <i>Clinical Microbiology and Infection</i> , 25(4), 489–495.									
STRAIN ID	Ampicillin (10µg), ≥21 (S)*, <16 (R)*	Imipemen (10µg), ≥24 (S)*, <17 (R)*	Cefotaxime (30µg), ≥26 (S)*, <23 (R)*	Ceftriaxone (30µg), ≥26 (S)*, <23 (R)*	Tobramycin (10µg), ≥18 (S)*, <16 (R)*	Amikacin (30µg), ≥17 (S)*, <15 (R)*	Minocycline (30µg), (30IU, ≥26 (S)*, <23 (R)*	Erythromycin (15µg), 15IU, ≥22 (S)*, <17 (R)*	Trimethoprim/sul phamethoxazole (1.25/23.75 µg) ≥16 (S)*, <10 (R)*
USC-21006	28 (S)	60 (S)	0 (R)	10 (R)	12 (R)	58 (S)	30 (S)	40 (S)	20 (S)
USC-21010	34 (S)	68 (S)	0 (R)	0 (R)	12 (R)	60 (S)	0 (R)	44 (S)	0 (R)¥
USC-21011	30 (S)	60 (S)	0 (R)	10 (R)	14 (R)	60 (S)	52 (S)	48 (S)	0 (R)¥
USC-21012	32 (S)	60 (S)	0 (R)	12 (R)	14 (R)	50 (S)	42 (S)	42 (S)	0 (R)¥
USC-21016	22 (S)	56 (S)	0 (R)	0 (R)	10 (R)	40 (S)	22 (S)	46 (S)	28 (S)
USC-21017	26 (S)	56 (S)	8 (R)	0 (R)	17 (I)	54 (S)	30 (S)	44 (S)	40 (S)
USC-21018	36 (S)	54 (S)	16 (R)	0 (R)	10 (R)	24 (S)	32 (S)	42 (S)	30 (S)
USC-21021	30 (S)	42 (S)	34 (S)	38 (S)	44 (S)	44 (S)	46 (S)	20 (I)	50 (S)
USC-21022	24 (S)	54 (S)	0 (R)	0 (R)	14 (R)	39 (S)	34 (S)	40 (S)	36 (S)
USC-21024	32 (S)	60 (S)	0 (R)	0 (R)	16 (I)	62 (S)	50 (S)	40 (S)	0 (R)¥
USC-21025	28 (S)	60 (S)	0 (R)	0 (R)	0 (R)	66 (S)	30 (S)	40 (S)	0 (R)¥
USC-21026	14 (R)	52 (S)	0 (R)	0 (R)	0 (R)	58 (S)	26 (S)	46 (S)	0 (R)¥
USC-21027	32 (S)	62 (S)	0 (R)	0 (R)	8 (R)	60 (S)	30 (S)	44 (S)	0 (R)¥
USC-21028	20 (I)	52 (S)	0 (R)	0 (R)	10 (R)	58 (S)	28 (S)	46 (S)	0 (R)¥
USC-21029	20 (I)	64 (S)	0 (R)	12 (R)	10 (R)	56 (S)	24 (I)	50 (S)	0 (R)¥
USC-21030	34 (S)	68 (S)	0 (R)	50 (S)	8 (R)	42 (S)	26 (S)	48 (S)	0 (R)¥
USC-21032	20 (I)	64 (S)	0 (R)	20 (R)	12 (R)	50 (S)	30 (S)	40 (S)	20 (S)
USC-21034	16 (I)	56 (S)	0 (R)	0 (R)	14 (R)	48 (S)	30 (S)	28 (S)	0 (R)¥
USC-21035	30 (S)	70 (S)	0 (R)	0 (R)	14 (R)	50 (S)	36 (S)	52 (S)	0 (R)¥
USC-21036	16 (I)	42 (S)	0 (R)	0 (R)	34 (S)	32 (S)	36 (S)	44 (S)	40 (S)

USC-21037	28 (S)	50 (S)	36 (S)	32 (S)	60 (S)	60 (S)	30 (S)	10 (R)	60 (S)
USC-21038	26 (S)	60 (S)	0 (R)	0 (R)	8 (R)	52 (S)	32 (S)	46 (S)	0 (R)¥
USC-21039	32 (S)	42 (S)	22 (R)	30 (S)	32 (S)	40 (S)	42 (S)	16 (R)	42 (S)
USC-21040	24 (S)	42 (S)	0 (R)	20 (R)	14 (R)	40 (S)	20 (R)	50 (S)	30 (S)
USC-21042	62 (S)	64 (S)	40 (S)	39 (S)	62 (S)	70 (S)	60 (S)	50 (S)	30 (S)
USC-21043	10 (R)	48 (S)	0 (R)	0 (R)	0 (R)	60 (S)	30 (S)	40 (S)	0 (R)¥
USC-21044	0 (R)	0 (R)	0 (R)	0 (R)	38 (S)	40 (S)	30 (S)	24 (S)	34 (S)
USC-21046	24 (S)	42 (S)	30 (S)	32 (S)	50 (S)	48 (S)	40 (S)	48 (S)	40 (S)
USC-21047	0 (R)	0 (R)	0 (R)	0 (R)	40 (S)	38 (S)	38 (S)	40 (S)	38 (S)
USC-21048	24 (S)	56 (S)	10 (R)	26 (S)	40 (S)	50 (S)	40 (S)	28 (S)	0 (R)¥
USC-21049	20 (I)	64 (S)	0 (R)	0 (R)	14 (R)	44 (S)	30 (S)	34 (S)	0 (R)¥
USC-21050	14 (R)	46 (S)	32 (S)	39 (S)	34 (S)	56 (S)	48 (S)	24 (S)	44 (S)

S, susceptible. I, Intermediate. R, resistant. Footnote: S, susceptible. I, Intermediate. R, resistant, ¥: inhibition zone is <10mm for Trimethoprim/sulphamethoxazole (1.25/23.75 µg) and an E-test strip is required to be subsequently performed as per Lebeaux, D., Bergeron, E., Berthet, J., Djadi-Prat, J., Mouniee, D., Boiron, P., Lortholary, O. and Rodriguez-Nava, V., 2019. Antibiotic susceptibility testing and species identification of *Nocardia* isolates: a retrospective analysis of data from a French expert laboratory, 2010–2015. *Clinical Microbiology and Infection*, 25(4), 489–495.).

Table S3. Differences in the adhesion capability of the 32 *Nocardia* isolates and control strains (HMLN-1 and 73-89) evaluated by microscopic method.

Isolates & Strains	Average No. of Adhering Bacteria/Cell ^a	Average No. of Adhering Bacteria /Cell ^a	Percentages of Cells Showing Adhering Bacteria ^b	Adherence Pattern Phenotype ^c
USC-21006	4.3 ± 0.3	4.3 ± 2.9	22.0	DA
USC-21010	1.4 ± 0.4	1.4 ± 3.4	5.0	DA
USC-21011	0.1 ± 0.0	0.1 ± 0.4	0.7	DA
USC-21012	0.1 ± 0.0	0.1 ± 0.3	0.7	DA
USC-21016	0.2 ± 0.1	0.2 ± 0.7	1.3	DA
USC-21017	4.3 ± 0.8	4.3 ± 7.2	9.3	DA
USC-21018	3.4 ± 0.4	3.4 ± 3.7	15.0	DA
USC-21021	5.3 ± 0.6	5.3 ± 5.6	15.3	DA
USC-21022	0.3 ± 0.1	0.3 ± 0.9	2.3	DA
USC-21024	0.1 ± 0.1	0.1 ± 0.6	0.3	DA
USC-21025	10.8 ± 0.8	10.8 ± 6.5	31.0	LA
USC-21026	0.5 ± 0.1	0.5 ± 1.2	4.3	DA
USC-21027	0.5 ± 0.1	0.5 ± 1.1	5.3	DA
USC-21028	1.3 ± 0.3	1.3 ± 3.0	5.3	DA
USC-21029	0.7 ± 0.2	0.7 ± 1.6	4.7	DA
USC-21030	8.9 ± 0.5	8.9 ± 4.0	52.7	DA
USC-21032	0.3 ± 0.1	0.3 ± 0.8	3.3	DA
USC-21034	15.5 ± 0.8	15.5 ± 6.6	99.3	DA
USC-21035	20.0 ± 0.1	20.0 ± 0.9	60.7	DA
USC-21036	6.5 ± 0.8	6.5 ± 6.6	16.3	LA
USC-21037	0.1 ± 0.1	0.1 ± 0.5	1.7	DA
USC-21038	16.5 ± 1.0	16.5 ± 9.0	21.7	DA
USC-21039	4.6 ± 0.5	4.6 ± 4.2	18.0	DA
USC-21040	0.3 ± 0.1	0.3 ± 1.1	1.7	DA
USC-21042	5.4 ± 0.6	5.4 ± 5.5	16.7	DA
USC-21043	0.2 ± 0.1	0.2 ± 0.8	1.3	DA
USC-21044	16.9 ± 0.7	16.9 ± 6.2	88.7	DA
USC-21046	4.7 ± 0.6	4.7 ± 5.0	15.0	DA
USC-21047	0.5 ± 0.2	0.5 ± 1.3	4.7	DA
USC-21048	5.0 ± 0.6	5.0 ± 5.3	16.0	DA
USC-21049	16.5 ± 1.0	16.5 ± 9.0	94.7	DA
USC-21050	19.4 ± 0.9	19.4 ± 7.7	98.3	DA
HMLN-1	21.8 ± 0.6	21.8 ± 5.1	90.0	DA
73-89	21.0 ± 0.4	21.0 ± 3.4	85.7	DA

Data shown are from triplicate experiments. Difference in the degrees of adhesion was found to be significant ($P < 0.0001$). ^aBacterial adhesion determined by enumerating the number of bacterial cells per 25 randomly selected Calu-3 cells in triplicates. ^bMean Percentage calculated by identifying bacterial cells displaying adherence to 100 randomly selected Calu-3 cells. ^cDA: diffuse; LA: Localized.

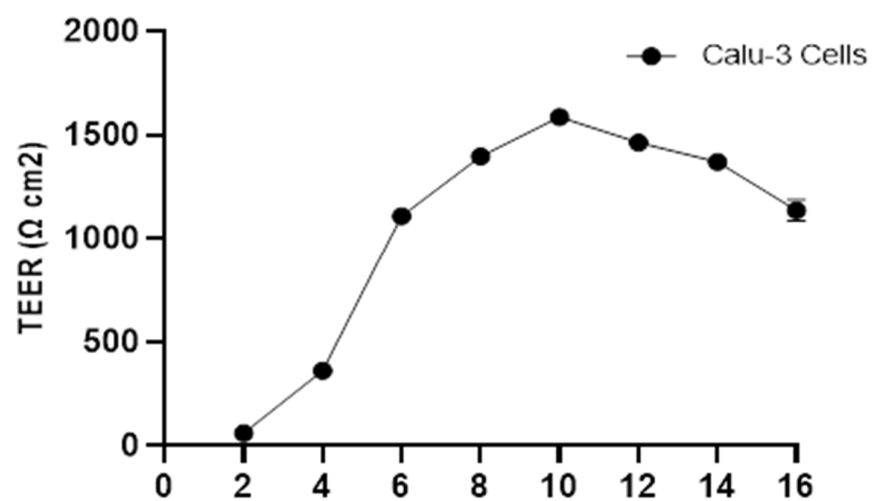


Figure S1. Change in TEER values of calu-3 cells following 16 days post seeding. Data shown are from triplicate experiments and plotted as the mean \pm SEM. Maximum TEER value (1588 Ω cm²) obtained on day 10 post seeding.