Supporting Information For:

Discovery and Characterization of Aryl Isonitriles as A New Class of Compounds versus Methicillin- and Vancomycin-resistant *Staphylococcus aureus*

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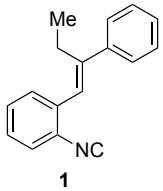
Part I. Synthetic Procedures and Spectra Data

General Methods.

Reactions were performed using standard syringe techniques under argon unless stated otherwise. Starting materials and reagents were used as received from suppliers (Aldrich, Alfa Aeser, Acros). Anhydrous THF was distilled over sodium benzophenone under argon. Acetonitrile (CH_3CN), dichloromethane (CH_2Cl_2), methanol (MeOH), and toluene were purified by passing the previously degassed solvents through activated alumina columns. Flash chromatography was performed using silica gel (230-400 mesh). Thin layer chromatography (TLC) was performed using glass-backed silica plates (Silicycle). NMR spectra were recorded on a *Bruker ARX-300, Bruker ARX-400* spectrometer, *DRX-500* or *AV-500* spectrometer at room temperature. Chemical shifts (in ppm) are given in reference to the solvent signal [^{1}H NMR: $CDCl_3$ (7.26); ^{13}C NMR: $CDCl_3$ (77.2).]. ^{1}H NMR data are reported as follows: chemical shifts (δ ppm), multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, br = broad), coupling constant (Hz), and integration. ^{13}C NMR data are reported in terms of chemical shift and multiplicity. IR data were recorded on a Thermo Nicolet Nexus 470 FTIR.

A Representative Procedure for the Synthesis of Aryl Isonitriles:



1

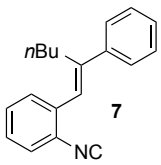
(E)-1-isocyano-2-(2-phenylbut-1-en-1-yl)benzene (1)[1]

To a stirred solution of diisopropyl amine (52 mg, 0.52 mmol) in THF (1.3 ml) was added a solution of *n*-BuLi (2.5 M in hexane, 0.174 ml, 0.43 mmol) dropwise at -78 ^{o}C. After stirring for 5 min, a solution of diethyl (2-isocyanobenzyl)phosphonate **4** (100 mg, 0.395 mmol) in THF (1 ml) was added dropwise

at -78 ^{o}C. The resulting solution was stirred for an additional 30 min and a solution of propiophenone (48 mg, 0.36 mmol) in THF (1 ml) was added dropwise. The reaction was stirred for an additional 30 min at -78 ^{o}C then warmed to room temperature and stirred for 1 h. A saturated aqueous ammonium chloride solution (4 ml) and Et_2O (4 ml) were added. The aqueous layer was extracted with Et_2O (3 x 5 ml) and the combined organic layers were washed with brine (10 ml), dried over anhydrous sodium sulfate and concentrated under reduced pressure. The resulting residue was purified by flash chromatography (CH_2Cl_2/hexane = 1/4) to yield (E)-1-isocyano-2-(2-phenylbut-1-en-1-yl)benzene **1** (50 mg, 60% yield.)

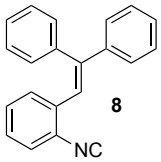
^{1}H NMR (300 MHz, $CDCl_3$) δ 7.35-7.17 (m, 4H), 7.20-6.99 (m, 3H), 6.96 (td, *J* = 7.7, 1.4 Hz, 1H), 6.78 (dd, *J* = 7.9, 1.5 Hz, 1H), 6.63 (s, 1H), 2.62 (qd, *J* = 7.4, 1.5 Hz, 2H), 1.14 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 166.1, 149.1, 140.3, 134.7, 130.2, 128.3 (3C), 128.2, 127.2, 126.7, 126.5, 119.7, 33.0, 12.8. The spectral data matches that reported by Studer and coworkers.[1]

Spectra Data of New Aryl Isonitriles:



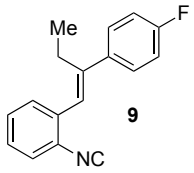
(E)-1-isocyano-2-(2-phenylhex-1-en-1-yl)benzene (7)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.66-7.19 (m, 4H), 7.19-7.04 (m, 3H), 6.95 (td, *J* = 7.7, 1.3 Hz, 1H), 6.75 (d, *J* = 10 Hz, 1H), 6.60 (s, 1H), 2.59 (t, *J* = 7.5 Hz, 2H), 1.32-1.56 (m, 4H), 0.91 (t, *J* = 7 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 165.9, 147.8, 140.3, 134.9, 130.3, 128.5 (2C), 128.4, 128.3, 127.3, 126.8, 126.6, 120.8, 39.8, 30.0, 22.2, 13.9; IR (neat): 2956, 2925, 2854, 2117, 1478, 1448 cm^{-1}; MS (ESI): *m/z* = 284.14 calc. for $C_{19}H_{19}N[M+Na]^+$, found 284.24.



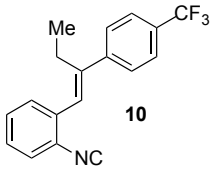
(2-(2-isocyanophenyl)ethene-1,1-diyl)dibenzene (8)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.38-7.28 (m, 8H), 7.16-7.11 (m, 3H), 7.10 (s, 1H), 7.00 (dt, *J* = 8.2, 1.1 Hz, 1H), 6.85 (d, *J* = 8.0 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.5, 146.7, 142.4, 139.5, 134.6, 130.4, 130.2, 128.5, 128.4, 128.3 (3C), 128.1, 127.9, 127.3, 126.9, 122.0; IR (neat) 3059, 3023, 2923, 2853, 2116, 1491, 1473, 1443, 1280, 1114, 1027, 942, 885 cm^{-1}; MS (ESI): *m/z* = 304.11 calc. for $C_{21}H_{15}N[M+Na]^+$, found 304.24.



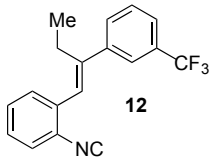
(E)-1-(2-(4-fluorophenyl)but-1-en-1-yl)-2-isocyanobenzene (9)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.30 (dd, *J* = 7.9, 1.3 Hz, 1H), 7.10 (t, *J* = 7.5 Hz, 1H), 7.07-7.04 (m, 2H), 7.00 (t, *J* = 7.8 Hz, 1H), 6.94 (m, 2H), 6.76 (d, *J* = 8.0 Hz, 1H), 6.59 (s, 1H), 2.58 (qd, *J* = 7.4, 1.4 Hz, 2H), 1.11 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.0 162.0 (d, *J* = 244 Hz), 148.0, 136.1 (d, *J* = 4 Hz), 134.6, 130.3, 130.1 (d, *J* = 8 Hz), 128.4, 126.9, 126.6, 125.6, 120.2, 115.4 (d, *J* = 21 Hz), 32.9, 12.8; IR (neat) 2967, 2930, 2117, 1602, 1507, 1222, 1178, 879, 836 cm^{-1}; MS (ESI): *m/z* = 274.10 calc. for $C_{17}H_{14}FN[M+Na]^+$, found 274.16.



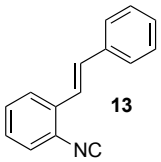
(E)-1-isocyano-2-(2-(4-(trifluoromethyl)phenyl)but-1-en-1-yl)benzene (10)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.51 (d, *J* = 8.0 Hz, 2H), 7.31 (dd, *J* = 8.0, 1.3 Hz, 1H), 7.22 (d, *J* = 8.0 Hz, 2H), 7.12 (td, *J* = 7.7, 1.4 Hz, 1H), 7.01 (td, *J* = 7.7, 1.3 Hz, 1H), 6.73 (d, *J* = 8.1 Hz, 1H), 6.67 (s, 1H), 2.61 (qd, *J* = 7.4, 1.5 Hz, 3H), 1.12 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.1, 147.6, 144.1, 134.1, 130.1, 128.7 (2C), 128.5, 127.2, 126.6, 126.1 (q, *J* = 269 Hz), 125.30, 125.27, 121.1, 32.6, 12.7; IR (neat) 2968, 2931, 2118, 1322, 1164, 1109, 1066, 881, 843 cm^{-1}; MS (ESI): *m/z* = 324.10 calc. for $C_{18}H_{14}F_3N[M+Na]^+$, found 324.08.



(E)-1-isocyano-2-(2-(3-(trifluoromethyl)phenyl)but-1-en-1-yl)benzene (12)

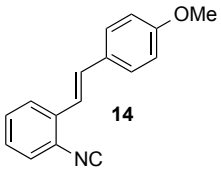
^{1}H NMR (400 MHz, $CDCl_3$) δ 7.49 (dd, *J* = 8.3, 1.1 Hz, 1H), 7.38-7.26 (m, 4H), 7.12 (td, *J* = 7.7, 1.4 Hz, 1H), 7.00 (td, *J* = 7.7, 1.4 Hz, 1H), 6.67-6.72 (m, 2H), 2.63 (qd, *J* = 7.4, 1.5 Hz, 2H), 1.14 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.6, 147.7, 141.3, 134.4, 132.2, 131.1, 130.9, 130.4, 129.1, 128.7, 127.5, 126.9, 126.2 (q, *J* = 267 Hz), 125.4, 124.3, 121.4, 32.6, 12.8; IR (neat) 2969, 2118, 1324, 1202, 1163, 1072, 903, 828 cm^{-1}; MS (ESI): *m/*z = 324.10 calc. for $C_{18}H_{14}F_3N[M+Na]^+$, found 324.08.



(E)-1-isocyano-2-styrylbenzene (13)

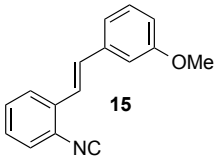
^{1}H NMR (400 MHz, $CDCl_3$) δ 7.75 (dd, *J* = 8.4, 1.3 Hz, 1H), 7.64-7.55 (m, 2H), 7.49-7.13 (m, 8H);

^{13}C NMR (100 MHz, $CDCl_3$) δ 167.0, 136.4, 133.7, 132.7, 129.4, 128.8, 128.6, 128.0, 127.3, 127.0, 125.4, 125.0, 122.2; IR (neat) 3044, 3022, 2119, 1632, 1598, 1480, 1446, 1288, 1263, 1221, 1198, 1092, 959, 875 cm^{-1}; MS (ESI): *m/z* = 228.08 calc. for $C_{15}H_{11}N[M+Na]^+$, found 228.00.



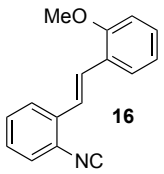
(E)-1-isocyano-2-(4-methoxystyryl)benzene (14)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.71 (dd, *J* = 7.5, 1.2 Hz, 1H), 7.56-7.50 (m, 2H), 7.41-7.36 (m, 2H), 7.27-7.11 (m, 3H), 6.96-6.89 (m, 2H), 3.85 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.7, 160.0, 134.1, 132.2, 129.4, 129.2, 128.4, 127.5, 127.3, 127.0, 125.2, 112.0, 114.3, 55.4; IR (neat) 2924, 2843, 2122, 1633, 1604, 1511, 1481, 1270, 1253, 1172, 1091, 961, 868 cm^{-1}; MS (ESI): *m/z* = 258.09 calc. for $C_{16}H_{13}NO[M+Na]^+$, found 258.10.



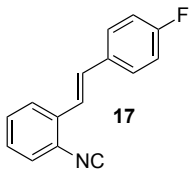
(E)-1-isocyano-2-(3-methoxystyryl)benzene (15)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.73 (d, *J* = 7.9 Hz, 1H), 7.42-7.36 (m, 3H), 7.32-7.26 (m, 2H), 7.19 (s, 1H), 7.16 (d, *J* = 7.8 Hz, 1H), 7.10 (m, 1H), 6.88 (dd, *J* = 8.2, 2.4 Hz, 1H), 3.86 (s 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.0, 159.9, 137.8, 133.6, 132.6, 129.8, 129.4, 128.0, 127.3, 125.5, 124.9, 122.5, 119.6, 114.2, 112.2, 55.3; IR (neat) 3072, 3034, 2119, 1607, 1581, 1489, 1482, 1448, 1286, 1239, 1157, 1135, 1093, 941, 873 cm^{-1}; MS (ESI): *m/z* = 258.09 calc. for $C_{16}H_{13}NO[M+Na]^+$, found 258.00.



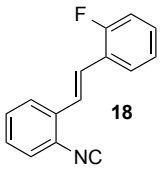
(E)-1-isocyano-2-(2-methoxystyryl)benzene (16)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.80 (d, *J* = 8.0 Hz, 1H); 7.66 (d, *J* = 7.6 Hz, 1H), 7.57 (d, *J* = 16.4 Hz, 1H), 7.46 (d, *J* = 16.5 Hz, 1H), 7.40-7.37 (m, 2H), 7.31 (t, *J* = 8.2 Hz, 1H), 7.24 (d, *J* = 8.0 Hz, 1H), 7.00 (t, *J* = 7.6 Hz, 1H), 6.93 (d, *J* = 8.2 Hz, 1H), 3.91 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.6, 157.2, 134.3, 129.6, 129.2, 127.6, 127.5, 127.1 (2C), 125.4 (2C), 124.8, 122.5, 120.8, 110.9, 55.4; IR (neat) 3033, 2958, 2936, 2834, 2119, 1489, 1463, 1334, 1242, 1191, 1107, 1091, 1031, 992, 965, 879 cm^{-1}; MS (ESI): *m/z* = 258.09 calc. for $C_{16}H_{13}NO[M+Na]^+$, found 258.10.



(E)-1-(4-fluorostyryl)-2-isocyanobenzene (17)

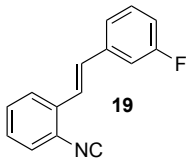
^{1}H NMR (500 MHz, Chloroform-*d*) δ 7.72 (dd, *J* = 8.4, 1.3 Hz, 1H), 7.58-7.52 (m, 2H), 7.40-7.37 (m, 2H), 7.33 (d, *J* = 16.3 Hz, 1H), 7.28 (td, *J* = 7.6, 1.1Hz, 1H), 7.16 (d, *J* = 16.3 Hz, 1H), 7.10-7.06 (m, 2H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.0, 162.9 (d, *J* = 287 Hz), 133.5, 132.6 (d, *J* = 3 Hz), 131.4, 129.4, 128.61, 128.55, 128.0, 127.3, 125.3, 121.9, 115.8 (d, *J* = 22 Hz); IR (neat) 3045, 2926, 2122, 1567, 1508, 1479, 1265, 1232, 1177, 1159, 960, 933, 817; MS (ESI): *m/z* = 224.09 calc. for $C_{15}H_{10}FN[M+H]$, found 224.08.



18

(E)-1-fluoro-2-(2-isocyanostyryl)benzene (18)

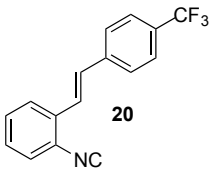
^{1}H NMR (500 MHz, $CDCl_3$) δ 7.77 (dd, *J* = 8.0, 1.3 Hz, 1H), 7.70 (td, *J* = 7.7, 1.7 Hz, 1H), 7.48 (d, *J* = 16.5 Hz, 1H), 7.42-7.37 (m, 3H), 7.30-7.27 (m, 2H), 7.18 (td, *J* = 7.6, 1.2 Hz, 1H), 7.10 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.2, 160.5 (d, *J* = 249 Hz), 133.6, 129.9 (d, *J* = 8 Hz), 129.4, 128.3, 127.2, 127.17 (d, *J* = 3 Hz), 125.5, 124.7 (d, *J* = 3 Hz), 125.0, 124.4, 124.3, 124.1 (d, *J* = 3 Hz), 115.8 (d, *J* = 22 Hz); IR (neat) 3057, 2924, 2122, 1635, 1487, 1476, 1452, 1336, 1283, 123, 1212, 1190, 1090, 960, 868 cm^{-1}; MS (ESI): *m/z* = 246.07 calc. for $C_{15}H_{10}FN[M+Na]^+$, found 246.00.



19

(E)-1-(3-fluorostyryl)-2-isocyanobenzene (19)

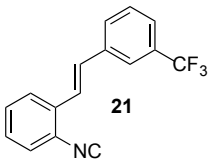
^{1}H NMR (500 MHz, $CDCl_3$) δ 7.76 (dd, *J* = 7.8, 1.5 Hz, 1H), 7.42-7.25 (m, 7H), 7.16 (d, *J* = 16.3 Hz, 1H), 7.01-7.04 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.3, 163.2 (d, *J* = 244 Hz), 138.7 (d, *J* = 7.3 Hz), 133.2, 131.5, 130.3 (d, *J* = 8 Hz), 129.5, 128.4, 127.3, 125.6, 125.1, 123.5, 122.8, 115.4 (d, *J* = 21 Hz), 113.5 (d, *J* = 22 Hz); IR (neat) 3072, 3035, 2122, 1608, 1581, 1482, 1448, 1286, 1238, 1183, 1158, 941, 874, 864, 834; MS (ESI): *m/z* = 246.07 calc. for $C_{15}H_{10}FN$, found 246.00.



20

(E)-1-isocyano-2-(4-(trifluoromethyl)styryl)benzene (20)

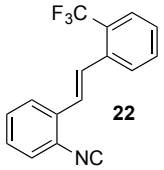
^{1}H NMR (500 MHz, $CDCl_3$) δ 7.75 (dd, *J* = 7.9, 1.3 Hz, 1H), 7.70-7.61 (m, 4H), 7.50 (d, *J* = 16.3 Hz, 1H), 7.42-7.41 (m, 2H), 7.32 (t, *J* = 6.8 Hz, 1H), 7.22 (d, *J* = 16.3 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.5, 139.8, 133.0, 131.0, 130.2 (q, *J* = 32 Hz), 129.5, 128.7, 127.4, 127.1, 125.8 (q, *J* = 4 Hz), 125.6, 125.1, 124.7, 124.1 (q, *J* = 270 Hz); IR (neat) 2924, 2123, 1614, 1483, 1321, 1190, 1155, 1104, 1065, 989, 964, 839 cm^{-1}; MS (ESI): *m/z* = 296.07 calc. for $C_{16}H_{10}FN[M+Na]^+$, found 295.84.



21

(E)-1-isocyano-2-(3-(trifluoromethyl)styryl)benzene (21)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.82-7.77 (m, 3H), 7.61-7.45 (m, 5H), 7.36 (t, *J* = 7.5 Hz, 1H), 7.25 (d, *J* = 16.3 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.3, 137.2, 133.1, 131.3 (q, *J* = 31 Hz), 131.1, 129.6, 129.5, 129.3, 128.6, 127.4, 125.6, 125.02, 124.99, 124.0, 124.0 (q, *J* = 271 Hz), 123.9; IR (neat) 3049, 2118, 1489, 1341, 1324, 1287, 1224, 1163, 1194, 1114, 1093, 998, 983, 962 cm^{-1}; MS (ESI): *m/z* = 296.07 calc. for $C_{16}H_{10}FN[M+Na]^+$, found 296.08.

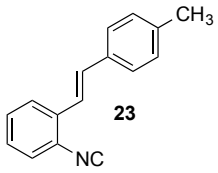


22

(E)-1-isocyano-2-(2-(trifluoromethyl)styryl)benzene (22)

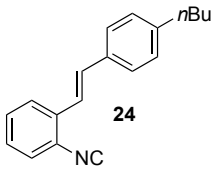
^{1}H NMR (500 MHz, $CDCl_3$) δ 7.86 (d, *J* = 7.8 Hz, 1H), 7.76 (d, *J* = 7.9 Hz, 1H), 7.70 (d, *J* = 7.9 Hz, 1H), 7.61-7.54 (m, 2H), 7.45-7.38 (m, 4H), 7.33 (td, *J* = 8.0, 1.3 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$), δ 167.2, 135.4, 133.2, 132.1, 129.6, 128.7, 128.3, 128.1, 127.9 (q, *J* = 29 Hz), 127.5, 127.3, 126.1,

126.0 (q, J = 5.4 Hz), 125.9, 125.3, 124.3 (q, J = 269 Hz); IR (neat) 3065, 2926, 2125, 1490, 1311, 1291, 1227, 1202, 1060, 1033, 959 cm^{-1}; MS (ESI): m/z = 296.07 calc. for $C_{16}H_{10}F_3N[M+Na]^+$, found 296.16.



(E)-1-isocyano-2-(4-methylstyryl)benzene (23)

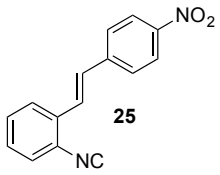
^1H NMR (500 MHz, $CDCl_3$) δ 7.75 (dd, J = 7.6, 1.1 Hz, 1H), 7.48 (d, J = 8.1 Hz, 2H), 7.35-7.41 (m, 3H), 7.26 (td, J = 7.4, 1.2 Hz, 1H), 7.20 (d, J = 7.1 Hz, 2H), 7.18 (d, J = 16.0 Hz, 1H), 2.38 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.8, 138.7, 133.9, 133.6, 132.6, 129.5, 129.4, 127.7, 127.3, 126.9, 125.3, 124.8, 121.1, 21.3; IR (neat) 3023, 2919, 2115, 1630, 1510, 1478, 1445, 1290, 1110, 959, 839 cm^{-1}; MS (ESI): m/z = 242.09 calc. for $C_{16}H_{13}N[M+Na]^+$, found 242.00.



(E)-1-(4-butylstyryl)-2-isocyanobenzene (24)

^1H NMR (500 MHz, $CDCl_3$) δ 7.73 (d, J = 8.0 Hz, 1H), 7.50 (d, J = 7.9 Hz, 2H), 7.36-7.30 (m, 3H), 7.26 (t, J = 7.4 Hz, 1H), 7.22 (d, J = 7.5 Hz, 2H), 7.19 (d, J = 15.8 Hz, 1H), 2.64 (t, J = 7.8 Hz, 2H), 1.63 (m, 2H), 1.39 (m, 2H), 0.95 (t, J = 7.4 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 166.9, 143.8, 134.0, 133.9, 132.7, 129.4, 128.9, 127.8, 127.3, 127.0, 125.4, 124.8, 121.2, 35.5, 33.6, 22.4, 14.0; IR (neat) 2956, 2928, 2857, 2116, 1736, 1632, 1608, 1480, 1449, 1265, 1241, 1018, 962, 854 cm^{-1}; MS

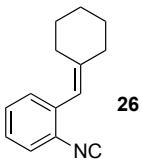
(ESI): *m/z* = 284.14 calc. for $C_{19}H_{19}N[M+Na]^+$, found 284.16.



25

(E)-1-isocyano-2-(4-nitrostyryl)benzene (25)

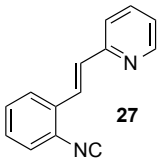
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.25 (d, *J* = 11.7 Hz, 2H), 7.76 (d, *J* = 10.5 Hz, 1H), 7.70 (d, *J* = 11.7 Hz, 2H), 7.56 (d, *J* = 21.8 Hz, 1H), 7.30-7.46 (m, 3H), 7.24 (d, *J* = 21.8 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.8, 147.4, 142.7, 132.5, 130.2, 129.6, 129.2, 127.5 (2C), 126.6, 125.8, 124.2, 123.7; IR (neat) 2923, 2842, 2121, 1632, 1510, 1372, 1340, 1299, 1269, 1253, 1226, 1172, 1108, 1091, 1026, 959, 886, 870; MS (ESI): *m/z* = 273.06 calc. for $C_{15}H_{10}N_2O_2[M+Na]^+$, found 273.12.



26

1-(cyclohexylidenemethyl)-2-isocyanobenzene (26)

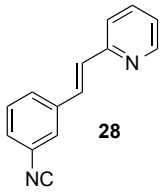
^{1}H NMR (500 MHz, $CDCl_3$) δ 7.36-7.34 (m, 1H), 7.32 (d, *J* = 7.5 Hz, 1H), 7.26 (d, *J* = 7.5 Hz, 1H), 7.27-7.20 (m, 1H), 2.33 (t, *J* = 6.1 Hz, 2H), 2.25 (t, *J* = 5.6 Hz, 2H), 1.70-1.52 (m, 6H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 165.2, 147.2, 135.3, 130.3, 128.6, 126.7, 126.6, 125.8, 116.9, 37.3, 29.8, 28.4, 27.7, 26.4; IR (neat) 2927, 2853, 2118, 1479, 1461, 1445, 1343, 1038, 838 cm^{-1}; MS (ESI): *m/z* = 220.11 calc. for $C_{14}H_{15}N[M+Na]^+$, found 220.08.



27

(*E*)-2-(2-isocyanostyryl)pyridine (27)

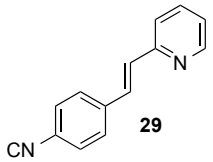
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.65-8.63 (m, 1H), 7.88 (d, *J* = 16.2 Hz, 1H), 7.80-7.76 (m, 1H), 7.71 (td, *J* = 7.7, 1.8 Hz, 1H), 7.53 (dt, *J* = 7.9, 1.1 Hz, 1H), 7.45-7.38 (m, 2H), 7.35-7.28 (m, 2H), 7.21 (ddd, *J* = 7.6, 4.8, 1.1 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 167.6, 154.9, 149.9, 136.6, 133.1, 132.4, 129.5, 128.7, 127.5, 126.3, 126.1, 125.3, 122.8, 122.0; **IR** (neat) 3075, 3042, 2118, 1581, 1560, 1485, 1469, 1453, 1427, 1331, 1303, 1279, 1239, 1209, 1180, 1149, 1091, 1049, 992, 965 [1], 897, 889, 862 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



28

(*E*)-2-(3-isocyanostyryl)pyridine (28)

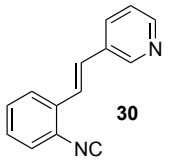
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.62 (ddd, *J* = 4.8, 1.8, 0.9 Hz, 1H), 7.69 (td, *J* = 7.7, 1.8 Hz, 1H), 7.64-7.55 (m, 3H), 7.43-7.35 (m, 2H), 7.32-7.28 (m, 1H), 7.20 (td, 1H, *J* = 4.8, 1.2 Hz), 7.17 (d, *J* = 16.1 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.2, 154.7, 149.9, 138.4, 136.7, 130.4, 130.1, 129.8, 128.0, 127.1, 125.8, 124.5, 122.7 (2 C); **IR** (neat) 3065, 3002, 2925, 2131, 1597, 1583, 1560, 1471, 1441, 1430, 1334, 1302, 1275, 1240, 1209, 1146, 1094, 1083, 978, 951, 892, 863 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



29

(*E*)-2-(4-isocyanostyryl)pyridine (29)

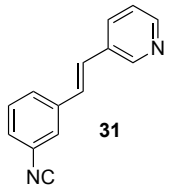
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.62 (ddd, *J* = 4.8, 1.8, 0.9 Hz, 1H), 7.69 (td, *J* = 7.7, 1.8 Hz, 1H), 7.63 (d, *J* = 16.1 Hz, 1H), 7.60-7.57 (m, 2H), 7.40-7.36 (m, 3H), 7.22-7.16 (m, 2H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.8, 154.8, 149.8, 137.9, 136.7, 130.8, 130.1, 127.8 (2 C), 126.8 (2 C), 122.7 (2 C), 122.7; **IR** (neat) 3056, 2922, 2851, 2130, 1583, 1562, 1505, 1468, 1430, 975; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



30

(*E*)-3-(2-isocyanostyryl)pyridine (30)

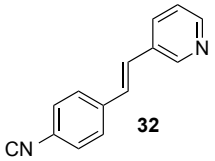
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.74 (d, *J* = 2.3, 1H), 8.55 (dd, *J* = 4.8, 1.6 Hz, 1H), 7.95-7.92 (m, 1H), 7.77-7.73 (m, 1H), 7.46 (d, 1H, *J* = 16.3 Hz), 7.44-7.38 (m, 2H), 7.36-7.29 (m, 2H), 7.18 (d, 1H, *J* = 16.3 Hz); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 167.5, 149.5, 149.2, 133.0, 132.9, 132.1, 129.6, 128.9, 128.7, 127.4, 125.6, 125.1, 124.4, 123.7; **IR** (neat) 3033, 2119, 1583, 1565, 1485, 1450, 1423, 1278, 1228, 1184, 1161, 1091, 1043, 1022, 962, 909 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



31

(*E*)-3-(3-isocyanostyryl)pyridine (31)

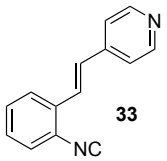
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.74 (dd, *J* = 2.3, 0.9 Hz, 1H), 8.53 (dd, *J* = 4.7, 1.6 Hz, 1H), 7.84 (ddd, *J* = 8.0, 2.3, 1.6 Hz, 1H), 7.56-7.52 (m, 2H), 7.44-7.38 (m, 1H), 7.34-7.28 (m, 2H), 7.11 (s, 2H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.4, 149.3, 148.7, 138.3, 132.9, 132.2, 129.9, 128.5, 127.5, 127.4, 127.2, 125.7, 124.2, 123.7; **IR** (neat) 3027, 2124, 1598, 1581, 1567, 1481, 1435, 1411, 1273, 1182, 1024, 966, 885 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



32

(*E*)-3-(4-isocyanostyryl)pyridine (32)

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.73 (d, *J* = 2.2 Hz, 1H), 8.53 (dd, *J* = 4.8, 1.6 Hz, 1H), 7.83 (dt, *J* = 8.0, 2.0 Hz, 1H), 7.57-7.51 (m, 2H), 7.41-7.36 (m, 2H), 7.31 (ddd, *J* = 8.0, 4.8, 0.9 Hz, 1H), 7.11 (d, *J* = 4.3 Hz, 2H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.9, 149.2, 148.7, 137.9, 132.9, 132.2, 128.9, 127.4 (2 C), 127.3, 126.9 (2 C), 123.6; **IR** (neat) 3028, 2920, 2850, 2127, 1644, 1600, 1574, 1567, 1503, 1483, 1421, 1409, 1329, 1304, 1252, 1166, 1130, 1100, 1022.75, 964, 942, 865 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.

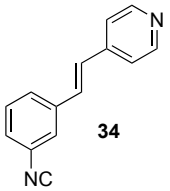


33

(*E*)-4-(2-isocyanostyryl)pyridine (33)

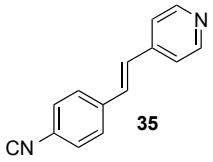
KKB-1-19: **^{1}H NMR** (500 MHz, Benzene-d_6) δ 8.54-8.46 (m, 2H), 7.40 (d, *J* = 16.3 Hz, 1H), 7.01 (dd, *J* = 8.0, 1.3 Hz, 1H), 6.82-6.71 (m, 4H), 6.60 (td, *J* = 7.7, 1.4 Hz, 1H), 6.49 (d, *J* = 16.3 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$**)** δ 167.8, 150.4 (2 C), 143.6, 132.5, 130.1, 129.6, 129.2, 127.5, 126.6, 125.9, 125.3, 121.1 (2 C); **IR** (neat) 3052, 2922, 2852, 2122, 1592, 1549, 1495, 1479, 1451, 1413, 1309, 1274,

1243, 1213, 1090, 991, 966, 958, 880 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



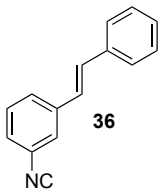
(*E*)-4-(3-isocyanostyryl)pyridine (34)

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.64-8.58 (m, 2H), 7.58-7.52 (m, 2H), 7.42 (t, *J* = 8.1 Hz, 1H), 7.38-7.35 (m, 2H), 7.34-7.31 (m, 1H) 7.23 (d, *J* = 16.3 Hz, 1H), 7.05 (d, *J* = 16.3 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.7, 150.4 (2 C), 143.7, 137.8, 130.8, 129.9, 128.4, 127.8, 127.3, 126.2, 124.5, 121.0 (2 C); **IR** (neat) 3056, 3029, 2922, 2128, 1591, 1549, 1494, 1478, 1450, 1415, 1242, 1217, 1174, 991, 971, 922, 893, 874, 859 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



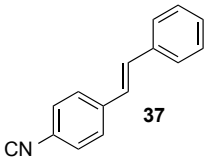
(*E*)-4-(4-isocyanostyryl)pyridine (35)

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.64-8.59 (m, 2H), 7.60-7.53 (m, 2H), 7.42-7.35 (m, 4H), 7.26 (d, 1H, *J* = 16.3 Hz), 7.04 (d, *J* = 16.3 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 165.3, 150.4 (2 C), 143.8, 137.3, 131.1, 128.4, 127.8 (2 C), 126.9 (2 C), 126.2, 121.0 (2 C); **IR** (neat) 3031, 2923, 2118, 1582, 1559, 1504, 1486, 1469, 1453, 1427, 1331, 1302, 1279, 1239, 1209, 1180, 1148, 1091, 992, 962, 942, 898, 858 cm^{-1}; **MS** (GC-MS) *m/z* = 207.08 calc. for $C_{14}H_{10}N_2[M+H]^+$, found 207.1.



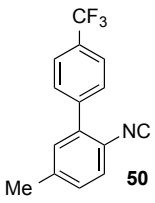
(*E*)-1-isocyano-3-styrylbenzene (36)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.54-7.50 (m, 4H), 7.41-7.35 (m, 3H), 7.34-7.28 (m, 1H), 7.26-7.23 (m, 1H), 7.14 (d, *J* = 16.3 Hz, 1H), 7.04 (d, *J* = 16.3 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.1, 139.0, 136.4, 131.1, 129.7, 128.8 (2 C), 128.4, 127.4, 127.1, 126.8 (2 C), 126.4, 125.1, 124.0; **IR** (neat) 3024, 2922, 2126, 1597, 1578, 1496, 1485, 1472, 1449, 1267, 1226, 1168, 1145, 1081, 1074, 965, 911, 884 cm^{-1}; **MS** (GC-MS) *m/z* = 206.09 calc. for $C_{15}H_{11}N[M+H]^+$, found 206.1.



(*E*)-1-isocyano-4-styrylbenzene (37)

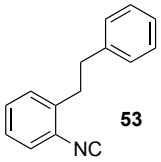
^{1}H NMR (500 MHz, $CDCl_3$) δ 7.54-7.49 (m, 4H), 7.41-7.35 (m, 4H), 7.33-7.28 (m, 1H), 7.16 (d, *J* = 16.3 Hz, 1H), 7.08 (d, *J* = 16.3 Hz, 1H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 164.5, 138.6, 136.5, 131.1 (2 C), 128.8 (2 C), 128.4 (2 C), 127.2 (2 C), 126.8 (3 C), 125.2; **IR** (neat) 3023, 2920, 2850, 2123, 1633, 1576, 1503, 1448, 1417, 1335, 1305, 1220, 1198, 1160, 1107, 1073, 967, 950, 918, 866 cm^{-1}; **MS** (GC-MS) *m/z* = 206.09 calc. for $C_{15}H_{11}N[M+H]^+$, found 206.1.



2-isocyano-5-methyl-4'-(trifluoromethyl)-1,1'-biphenyl (50)

Prepared according to a procedure reported by Studer[2] from 2-bromo-4-methylanaline and 4-(trifluoromethyl)phenylboronic acid.

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.77 (d, *J* = 8.1 Hz, 2H), 7.66 (d, *J* = 8.1 Hz, 2H), 7.44 (d, *J* = 8.6 Hz, 1H), 7.26-7.27 (m, 2H), 2.47 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) 166.8, 140.9, 140.4, 137.3, 131.1, 130.4 (q, *J* = 32 Hz), 129.8, 129.6, 127.9, 125.7, 124.2 (q, *J* = 270 Hz), 122.3, 21.3; IR (cm^{-1}): 2922, 2125, 1620, 1571, 1493, 1396, 1328, 1198, 1179, 1155, 1110, 965, 952, 897, 880, 845; MS (ESI): *m/z* = 262.09 calc. for $C_{15}H_{10}F_3N[M+H]^+$, found 262.2.



53

1-isocyano-2-phenethylbenzene (53)

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.37 (dd, *J* = 7.8, 1.4 Hz, 1H), 7.33-7.28 (m, 3H), 7.27-7.19 (m, 5H), 3.07 (dd, *J* = 8.9, 7.6 Hz, 2H), 2.95 (dd, *J* = 8.9, 5.6 Hz, 2H); **^{13}C NMR** (125 MHz, $CDCl_3$) δ 166.1, 140.8, 138.2, 130.0, 129.4, 128.6 (2 C), 128.5 (2 C), 127.1, 126.9, 126.2, 126.1, 36.0, 34.7; **IR** (neat) 3063, 3028, 2926, 2855, 2119, 1603, 1496, 1487, 1453 cm^{-1}; **MS** (GC-MS) *m/z* = 208.11 calc. for $C_{15}H_{13}N[M+H]^+$, found 208.1.

Compounds **46**[2], **49**[2] and **51**[2] were prepared according to the literature procedure and their spectral data match with the reported ones. Compounds **5**[1], **6**[1], **11**[1], **42**[3], and **43**[4] were prepared according to the representative procedure aforementioned and their spectral data match with the reported ones.

Part II. Biological Materials and Evaluation Methods

Bacterial strains and reagents. Clinical isolates of MRSA, vancomycin-intermediate *S. aureus* (VISA), and vancomycin-resistant *S. aureus* (VRSA) were obtained through the Network of Antimicrobial Resistance in *Staphylococcus aureus* (NARSA) program (Table 1). Vancomycin hydrochloride (Gold Biotechnology, St. Louis, MO, USA) and linezolid (Chem-Impex International, Inc., Wood Dale, IL, USA) powders were purchased commercially and dissolved in DMSO to prepare a stock 10 mM solution.

Assessment of antimicrobial activity of the isonitrile compounds against multidrug-resistant S. aureus strains. The minimum inhibitory concentration (MIC) of each compound and linezolid was determined against eight different strains of MRSA, VISA, and VRSA using a modified version of the broth microdilution method, outlined by the CLSI.[5] The same analysis was performed with vancomycin against the VISA and VRSA strains tested. A bacterial suspension (~1×10^5 CFU/mL) was prepared in Tryptic soy broth (TSB) and then transferred to a microtiter plate. Each agent tested was added (in triplicate) to wells in the first row of the plate and then serially diluted downward. Plates were incubated at 37 ºC for 18-20 h before the MIC was determined as the lowest concentration of each test agent where bacterial growth was not visible.

Toxicity analysis of selected isonitrile compounds tested against mammalian cells. Selected isonitrile compounds were assayed at concentrations of 16 μM, 32 μM, 64 μM, and 128 μM against a murine macrophage (J774) cell line to asses if the compounds exhibited toxicity to mammalian cells *in vitro*. Cells were cultured in Dulbeco's modified Eagle's medium (Sigma-Aldrich, St. Louis, MO, USA) with

10% fetal bovine serum (USA Scientific, Inc.) at 37 ºC with 5% CO_2. Controls received DMSO alone at a concentration equal to that in drug-treated cell samples. The cells were incubated with each compound (in triplicate) in a 96-well tissue-culture plate at 37 ºC and 5% CO_2 for 2 h prior to addition of the assay reagent MTS 3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl) -2-(4-sulfophenyl)-2H-tetrazolium) (Promega, Madison, WI, USA). Absorbance readings (at OD_{490}) were taken using a kinetic microplate reader (Molecular Devices, Sunnyvale, CA, USA). The quantity of viable cells after treatment with each compound was expressed as a percentage of the viability of DMSO-treated control cells (average of triplicate wells ± standard deviation). Statistical analysis was performed (comparing cells treated with compound versus cells treated with DMSO) using the paired t-test ($P < 0.05$) utilizing Microsoft EXCEL software.

*Caco-2 permeability analysis of compound **13***. The ability of compound **13**, ranitidine (low permeability control), warfarin (high permeability control), and talinolol (P-glycoprotein efflux substrate), to effectively permeate across a biological membrane was assessed using a Caco-2 cell monolayer, as described elsewhere.[6] The amount of permeation was determined both from the apical (A) to basolateral (B) direction and the basolateral (B) to apical (A) direction. Data for apparent permeability (P*app*) and the efflux ratio (R_E) were determined as explained elsewhere.[6] An $R_E > 2$ indicates the test agent may be a potential substrate for P-glycoprotein or other active efflux transporters.

Kinetic solubility screen. A kinetic solubility analysis of compound **13**, reserpine, tamoxifen, and verapamil was performed as has been described elsewhere.[6] The solubility limit (in μM) reported is the

maximum concentration of each test agent where turbidity was not observed. Values below 1 μM indicate compound is insoluble, values between 1 to 100 μM indicate partial aqueous solubility, and values above 100 μM indicate test agent is fully soluble.

Metabolic stability analysis using pooled human liver microsomes. To analyze the stability of compound **13** to metabolic processes in the liver, this compound was incubated in duplicate with pooled human liver microsomes at 37 ºC (for 60 min), using a similar protocol described elsewhere, with two modifications.[7,8] First, the reaction mixture utilized 0.3 mg/mL microsomal protein. Additionally, samples were collected after 0 and 60 min and analyzed accordingly. Data are reported as % remaining by dividing by the time zero concentration value.

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