SUPPLEMENTARY MATERIAL

Antibacterial and antifungal properties of guanylhydrazones

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ANALYTICAL AND SPECTRAL DATA OF THE COMPOUNDS

Compounds were analyzed for purity (HPLC) using a Agilent 1200 HPLC system equipped with Quat Pump (G1311B), Injector (G1329B) 1260 ALS, TCC 1260 (G1316A) and Detector 1260 DAD VL+ (G1315C). HPLC analysis was performed in the diverse systems:

Method A

Zorbax Eclipse Plus C18 4.6 x 150mm, 1.8µ, S.N. USWKY01594 was used as the stationary phase. Eluent was made from the following solvents: 0.2% formic acid in water (A) and acetonitrile (B). The analysis were performed at the UV max of the compounds to maximize selectivity. Compounds were dissolved in methanol, final concentrations were ~ 1 mg/mL. Flow rate was 0.5 mL/min.

Compounds **22**, **23**, **24** were eluted using gradient protocol: 0 – 0.5 min 95% A, 0.5 - 3 min 95% A→ 5% A, 3 - 13 min 5% A, 13 – 14 min 5% A→ 95% A, 14 – 16 min 95% A.

Method B

Zorbax Eclipse Plus C18 4.6 x 150mm, 1.8µ, S.N. USWKY01594 was used as the stationary phase. Eluent was made from the following solvents: 0.2% formic acid in water (A) and methanol (B). The analysis were performed at the UV max of the compounds to maximize

selectivity. Compounds were dissolved in methanol, final concentrations were ~ 1 mg/mL. Flow rate was 0.5 mL/min.

Compounds **22**, **23**, **24** were eluted using gradient protocol: 0 – 0.5 min 95% A, 0.5 - 3 min 95% A→ 5% A, 3 - 13 min 5% A, 13 – 14 min 5% A→ 95% A, 14 – 16 min 95% A.

Method C

Zorbax Eclipse Plus C18 2.1 x 100mm, 1.8µ, was used as the stationary phase. Eluent was made from the following solvents: 0.2% formic acid in water (A) and acetonitrile (B). The analysis were performed at the UV max of the compounds to maximize selectivity. Compounds were dissolved in methanol, final concentrations were ~ 1 mg/mL. Flow rate was 0.2 mL/min.

Compounds **18**, **19**, **20**, **21**, **25**, **26** and **27** were eluted using gradient protocol: 0 – 0.5 min 95% A, 0.5 - 3 min 95% A→ 5% A, 3 - 13 min 5% A, 13 – 14 min 5% A→ 95% A, 14 – 16 min 95% A.

Method D

Zorbax Eclipse Plus C18 2.1 x 100mm, 1.8µ, was used as the stationary phase. Eluent was made from the following solvents: 0.2% formic acid in water (A) and methanol (B). The analysis were performed at the UV max of the compounds to maximize selectivity. Compounds were dissolved in methanol, final concentrations were ~ 1 mg/mL. Flow rate was 0.2 mL/min.

Compounds **18**, **19**, **20**, **21**, **25**, **26** and **27** were eluted using gradient protocol: 0 – 0.5 min 95% A, 0.5 - 3 min 95% A→ 5% A, 3 - 13 min 5% A, 13 – 14 min 5% A→ 95% A, 14 – 16 min 95% A.

5-(4-Methylphenyl)furan-2-carbaldehyde (4)[1]

Orange amorphous powder; m.p. = 49-51 °C. IR (ATR): 3308w, 3128w, 3026w, 2915w, 2859w, 2824w, 1657s, 1607m, 1528m, 1482m, 1416w, 1387w, 1291w, 1255m, 1203w, 1118w, 1028m, 964w, 921w, 822w cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ 9.63 (s, 1H), 7.72 (d, *J* = 8.2, 2H), 7.31 (d, *J* = 3.7, 1H), 7.27 (s, 1H), 7.23 (s, 1H), 6.79 (d, *J* = 3.7, 1H), 2.39 (s, 3H) ppm. GC/MS (*m/z* (%)): 186.0 ($[M]^+$, 100), 129.0 (70).

5-(4-Bromophenyl)furan-2-carbaldehyde (5)[2]

Orange amorphous powder; m.p. = 151-152 °C. ^1H NMR (500 MHz, $CDCl_3$): δ 9.66 (s, 1H), 7.72-7.65 (m, 2H), 7.61-7.55 (m, 2H), 7.31 (d, *J* = 3.7 Hz, 1H), 6.84 (d, *J* = 3.7 Hz, 1H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.22, 158.21, 152.17, 132.20, 127.88, 126.69, 123.93, 123.39, 108.04 ppm. GC/MS (*m/z* (%)): 251.9 ($[M]^+$, 100), 192.9 (40).

5-(4-Fluorophenyl)furan-2-carbaldehyde (6)[1]

Yellow solid; m.p. = 70-71 ºC. IR (ATR): 3315m, 3135m, 3103m, 2918m, 2850m, 1666s, 1602s, 1567m, 1482s, 1420s, 1392m, 1356m, 1304w, 1286m, 1254m, 1227s, 1157m, 1102m, 1065w, 1024m, 966m, 922m, 889w, 834m, 813s cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ 9.65 (s, 1H), 7.86-7.82 (m, 1H), 7.81-7.77 (m, 1H), 7.32 (d, *J* = 3.8 Hz, 1H), 7.20-7.09 (m, 2H), 6.79 (d, *J* = 3.7 Hz, 1H) ppm. GC/MS (*m/z* (%)): 190.0 ($[M]^+$, 100).

5-(4-Methoxyphenyl)furan-2-carbaldehyde (7)[1]

Orange oil. IR (ATR): 3318w, 3214.8w, 3118w, 3004w, 1937m, 2838m, 2733w, 2552w, 1733w, 1668s, 1609s, 1530wm 1481s, 1389m, 1296m, 1254s, 1177m, 1114m, 1065wm 1026m, 967m, 921w, 835m cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ 9.60 (s, 1H), 7.77 (d, *J* = 9.0 Hz, 2H), 7.31 (d, *J* = 4.3 Hz, 1H), 6.96 (d, *J* = 9.0 Hz, 2H), 6.72 (d, *J* = 3.7 Hz, 1H), 3.86 (s, 3H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 176.85, 160.88, 159.81, 151.59, 126.96, 124.19, 114.40, 106.28, 95.75, 55.33 ppm. GC/MS (*m/z* (%)): 202.0 ($[M]^+$, 100), 187.0 (40), 145.0 (40).

4-Bromo-5-phenylthiophene-2-carbaldehyde (10)[3]

Yellow solid; m.p. = 57-60 °C. IR (ATR): 3310w, 3082w, 3053w, 3026w, 2845w, 1678s, 1645s, 1519w, 1449m, 1430m,1394w, 1309w, 1226m, 1122w, 1031w, 997w, 966w, 915w, 842w, 755w cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ = 9.86 (s, 1H), 7.72 (s, 1H), 7.70-7.67 (m, 2H), 7.50-7.45 (m, 3H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 181.8, 148.1, 141.3, 139.8, 131.8, 129.7, 129.0, 128.8, 108.8 ppm. GC/MS (*m/z* (%)): 267.9 $[M]^+$.

4-Bromo-5-phenyl-2-furaldehyde (11)[4]

Dark oil. IR (ATR): 3341w, 3132w, 2834w, 1682s, 1566w, 1521w, 1476m, 1284w, 1140w cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ 9.64 (s, 1H), 8.15-8.05 (m, 2H), 7.52-7.41 (m, 3H), 7.34 (s, 1H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 176.97, 153.96, 150.46, 130.04, 128.74, 128.17, 126.76, 125.72, 98.23 ppm. GC/MS (*m/z* (%)) : 249.9 $[M]^+$.

4,5-Diphenylthiophene-2-carbaldehyde (12)

Yellow oil. IR (ATR): 2919m, 2851m, 1734w, 1657s, 1542w, 1452w, 1427m, 1253w, 1165m, 1106w,1071w cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ 9.92 (s, 1H), 7.80 (s, 1H), 7.34-7.29 (m, 8H), 7.28-7.25 (m, 2H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 182.83, 148.74, 141.41, 139.55, 139.08, 135.16, 133.10, 129.19, 128.97, 128.87, 128.72, 128.64, 127.65 ppm. GC/MS (*m/z* (%)): 264.0 ($[M]^+$, 100), 235.0 (50).

4-(4-Fluorophenyl)-5-phenylthiophene-2-carbaldehyde (13)

Yellow oil. IR (ATR): 3318w, 3058w, 2926w, 2819w, 1670s, 1604w, 1543w, 1507m, 1434m, 1259w, 1226m, 1175m, 1159w, 1109w, 1072w cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$): δ 9.91 (s,

1H), 7.76 (s, 1H), 7.37-7.28 (m, 5H), 7.25-7.20 (m, 2H), 7.04-7.97 (m, 2H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 182.72, 162.27 (d, *J* = 245.5 Hz), 148.69, 141.46, 138.72, 138.42, 132.89, 131.15 (d, *J* = 2.7 Hz), 130.63 (d, *J* = 7.1), 129.15, 128.97, 128.81, 115.66 (d, *J* = 21.7 Hz) ppm. GC/MS (*m/z* (%)): 282.0 ($[M]^+$, 100), 253.0 (30).

2-(4-Bromo-5-phenyl-2-furyl)-1,3-dioxolane (14)

Yellow oil. ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.98-7.94 (m, 2H), 7.44-7.39 (m, 2H), 7.35-7.31 (m, 1H), 6.57 (s, 1H), 5.96 (s, 1H), 4.17-4.09 (m, 2H), 4.07-3.95 (m, 2H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 150.46, 149.25, 129.41, 128.42, 128.23, 125.72, 114.51, 97.43, 96.12, 65.18 ppm.

4-Fluoro-5-phenyl-2-furaldehyde (15)

Yellow solid. IR (ATR): 3188w, 3115w, 3067w, 2847w, 1683s, 1607m, 1528m, 1435m, 1314m, 1164w,1132w, 964w cm^{-1}. ^{1}H NMR (500 MHz, $CDCl_3$): δ 9.61 (s, 1H), 7.88-7.85 (m, 2H), 7.50-7.46 (m, 2H), 7.43-7.38 (m, 1H), 7.20-7.19 (m, 1H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 177.60, 149.49 (d, *J* = 255.5 Hz), 147.46 (d, *J* = 6.4 Hz), 142.68, 142.52, 129.50, 129.00, 127.20 (d, *J* = 4.5 Hz), 125.05 (d, *J* = 5.4 Hz) ppm. GC/MS (*m/z* (%)) : 190.0 $[M]^+$.

(4-Nitro-5-phenyl-2-thienyl)methylene diacetate (16)

Yellow oil. IR (ATR): 3108w, 3063w, 3025w, 2937w, 1769s, 1558m, 1528s, 1505m, 1372m, 1338m,1225s, 1193s, 1129m, 1070w, 1006m cm^{-1}. ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.81 (s, 1H), 7.78 (s, 1H), 7.50-7.42 (m, 5H), 2.17 (s, 6H) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ 168.24, 146.35, 141.73, 136.14, 129.93, 129.89, 129.54, 128.50, 124.20, 85.35, 20.61 ppm. (+)ESI-HRMS (*m/z):* $[M + Na]^+$ 358.03576 (error 0.5 ppm).

4-Nitro-5-phenylthiophene-2-carbaldehyde (17)

GC/MS (*m/z* (%)): 233.0 $[M]^+$.

(2E)-2-{[5-(4-methylphenyl)furan-2-yl]methylidene}hydrazinecarboximidamide hydrochloride (18)

Yellow solid; m.p. = 84-87 ^{o}C. IR (ATR): 3573w, 3433m, 3281s, 3217s, 3111s, 3001m, 1682s, 1635s, 1602s, 1528m, 1492m, 1426m, 1372w, 1333w, 1296w, 1268w, 1194w, 1139m, 1027m, 966w, 937m, 822w cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.00 (s, 1H), 7.68 (d, *J* = 8.2 Hz, 2H), 7.24 (d, *J* = 8.0 Hz, 2H), 7.00 (d, *J* = 3.5 Hz, 1H), 6.86 (d, *J* = 3.5 Hz, 1H), 2.36 (s, 3H) ppm. ^{13}C NMR (125 MHz, CD_3OD): δ 158.31, 156.92, 149.11, 139.95, 138.73, 130.58, 128.45, 125.50, 118.46, 108.01, 21.35 ppm. (+)ESI-HRMS (*m/z*): $[M+H]^+$ 243.12366 (error -1.53). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-{[5-(4-methoxyphenyl)furan-2-yl]methylidene}hydrazinecarboximidamide hydrochloride (19)*

Yellow solid; m.p. = 174-177 ºC. IR (ATR): 3405s, 3167m, 2932m, 2863m, 1676.4s, 1635s, 1494s, 1441m, 1294m, 1251m, 1175m, 1114w, 1020m, 967w, 923w, 831m cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 7.97 (s, 1H), 7.71 (d, *J* = 8.6 Hz, 2H), 6.96 – 6.95 (m, 3H), 6.75 (d, *J* = 3.3 Hz, 1H), 3.80 (s, 3H) ppm. ^{13}C NMR (125 MHz, D_2O): δ 161.43, 158.08, 156.65, 148.54, 138.52, 126.86, 123.72, 118.48, 115.16, 106.90, 55.62 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 259.11951 (error 2.15 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-{[5-(4-fluorophenyl)furan-2-yl]methylidene}hydrazinecarboximidamide hydrochloride (20)*

Orange solid; m.p. = 197-199 ºC IR (ATR): 3060m, 2998m, 2927m, 2857m, 2775m, 1668s, 1615s, 1531s, 1484s, 1445s, 1334m, 1304m, 1270m, 1214s, 1158s, 1136s, 1019m, 924m, 836m cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.01 (s, 1H), 7.85-7.82 (m, 2H), 7.18-7.15 (m, 2H), 7.02 (d, *J* = 3.6 Hz, 1H), 6.91 (d, *J* = 3.6 Hz, 1H) ppm. ^{13}C NMR (125 MHz, D_2O): δ 164.25 (d, *J* = 246.3 Hz), 157.01, 156.96, 149.53, 138.58, 127.70 (d, *J* = 3.5 Hz), 127.61 (d, *J* = 8.1 Hz), 118.33, 116.88 (d, *J* = 21.6 Hz), 108.53 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 307.01822 (error -2.22 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-{[5-(4-bromophenyl)furan-2-yl]methylidene}hydrazinecarboximidamide hydrochloride (21)*

Orange solid; m.p. = 99-101 ºC. IR (ATR): 3590m, 3320s, 1689s, 1638s, 1476m, 1406w, 1338w, 1272w, 1206w, 1155m, 1073w, 1032w, 1007w, 972w, 827w cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.00 (s, 1H), 7.74-7.72 (m, 2H), 7.59-7.57 (m, 2H), 7.03 (d, *J* = 3.6 Hz, 1H), 6.98 (d, *J* = 3.6 Hz, 1H) ppm. ^{13}C NMR (125 MHz, D_2O): δ 156.98, 156.73, 149.85, 138.49, 133.15, 130.21, 127.13, 123.34, 118.22, 109.39 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 307.01822 (error -2.22 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-[(4-fluoro-5-phenyl-2-furyl)methylene]hydrazinecarboximidamide hydrochloride (22)*

Yellow solid; m.p. = 97-101 ºC. IR (ATR): 3408s, 1694w, 1631m, 1493w, 1432w, 1168w cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 7.95 (s, 1H), 7.79-7.76 (m, 2H), 7.48-7.44 (m, 2H), 7.37-7.32 (m, 1H), 7.08 (s, 1H) ppm. ^{13}C NMR (125 MHz, CD_3OD) : δ 157.06, 151.29 (d, *J* = 252.0 Hz), 146.69, 146.62, 138.26 (d, *J* = 2.7 Hz), 130.07, 129.47, 129.26, 125.13 (d, *J* = 4.5 Hz), 107.69 (d, *J* = 20.7 Hz) ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 247.09872 (error -0.98 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-[(4-bromo-5-phenyl-2-thienyl)methylene]hydrazinecarboximidamide hydrochloride (23)*

Yellow solid; m.p. = 186-190 ºC. IR (ATR): 3391s, 2508s, 1679m, 1623s, 1530w, 1456w, 1300w, 1248w, 1149w cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.24 (s, 1H), 7.70-7.65 (m, 2H),

7.50-7.41 (m, 4H) ppm. ^{13}C NMR (125 MHz, CD_3OD) : δ 157.05, 143.04, 142.92, 138.40, 136.44, 133.77, 130.34, 130.11, 129.98, 108.91 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 322.99557 (error -1.50 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-[(4-bromo-5-phenyl-2-furyl)methylene]hydrazinecarboximidamide hydrochloride (24)*

Yellow solid; m.p. = 99-102 ºC. IR (ATR): 3361s, 3168s, 2878m, 1682s, 1629s, 1479w, 1445w, 1340w, 1249w, 1153w, 1073w, 1024w, 981w, 955w, 926w, 813w cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.07–8.04 (m, 2H), 7.99 (s, 1H), 7.51–7.46 (m, 2H), 7.45–7.40 (m, 1H), 7.16 (s, 1H) ppm. ^{13}C NMR (125 MHz, CD_3OD) : δ 157.06, 152.32, 149.22, 137.71, 130.25, 129.79, 127.15, 127.68, 120.65, 98.98 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 307.01797 (error -3.03 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-[(4-nitro-5-phenyl-2-thienyl)methylene]hydrazinecarboximidamide hydrochloride (25)*

Yellow solid; m.p. = 198-202 ºC. IR (ATR): 3391s, 3275s, 1691s, 1662s, 1621s, 1543m, 1521m, 1398w, 1331m, 1157w, 1015w cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.29 (s, 1H), 7.99 (s, 1H), 7.55-7.45 (m, 5H) ppm. ^{13}C NMR (125 MHz, CD_3OD): δ 156.95, 148.49, 143.99, 142.18, 137.08, 131.69, 131.15, 130.56, 129.98, 128.38 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 209.07008 (error -1.85 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-[(4,5-diphenyl-2-thienyl)methylene]hydrazinecarboximidamide hydrochloride (26)*

Yellow solid; m.p. = 208-210 ºC. IR (ATR): 3271s, 3169s, 2324m, 1680s, 1620s, 1258m, 1492m, 1423m, 1314m, 1272m, 1233m, 1195m, 1106m, 1071m, 1012m cm^{-1}. . ^{1}H NMR (500 MHz, CD_3OD): δ 8.31 (s, 1H), 7.50 (s, 1H), 7.32 – 7.23 (m, 10H) ppm. ^{13}C NMR (125 MHz, CD_3OD): δ 156.81, 143.96, 143.57, 140.25, 137.34, 137.03, 135.74, 135.00, 130.23, 130.04, 129.73, 129.60, 129.37, 128.48 ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 321.11597 (error -2.73 ppm). The compound was >95% pure based on HPLC purity analysis.

*(2*E*)-2-{[4-(4-fluorophenyl)-5-phenyl-2-thienyl]methylene}hydrazinecarboximidamide hydrochloride (27)*

Yellow solid; m.p. = 102-108 ºC. IR (ATR): 3158s, 1674s, 1622s, 1508s, 1435m, 1255m, 1193m, 1157m cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD): δ 8.29 (s, 1H), 7.50 (s, 1H), 7.31–7.23 (m, 7H), 7.05–7.00 (m, 2H) ppm. ^{13}C NMR (125 MHz, CD_3OD): δ 163.61 (d, J = 243.6 Hz), 156.84, 143.87, 143.65, 139.11, 137.47, 135.54, 134.85, 133.24 (d, J = 3.6 Hz), 131.94 (d, J = 8.1 Hz), 130.25, 129.84, 129.50, 116.38 (d, J = 21.6 Hz) ppm. (+)ESI-HRMS *m/z*: $[M + H]^+$ 339.10657 (error -2.52 ppm). The compound was >95% pure based on HPLC purity analysis.

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