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Application of palladium-catalyzed carboxyl anhydride-boronic acid cross coupling in the synthesis of novel bile acids analogs with modified side chains

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ABSTRACT

Palladium-catalyzed cross coupling of 4-methoxycarbonyl phenyboronic acid with acetylated bile acids in which the carboxyl functions was activated by formation of a mixed anhydride with pivalic anhydride afforded the cross coupled compounds, which were converted in novel side chain modified bile acids by one pot carbonyl reduction/removal of the protecting acetyl groups by Wolff–Kishner reduction. Unambiguous assignments of the NMR signals and crystal characterization of the heretofore unknown compounds are provided.

1. Introduction

Bile acids constitute a subfamily of steroids that has been the focus of intensive attention for nearly a century [1,2]. Some members of this family have been employed as starting materials in the synthesis bioactive steroids as sex hormones [3], brassinosteroids and their analogues [4,5], Gram-negative bacteria sensitizers and steroid antibiotics [6–8]. Bile acids have also played a major role in supramolecular chemistry serving as building blocks in the synthesis of ion receptors [9–11], molecular scaffolds [12], cholaphanes [13,14], cyclopeptides and cyclocholates [15–18], as well as dendrimers [19], gelators [20–22], and surfactants amongst others [23–25].

In general the above-mentioned applications imply the introduction of non-natural functionality in both, the steroid framework and the side chain. Palladium catalyzed reactions are useful tools in steroid total synthesis and for the introduction of structural modifications in the skeleton and/or the side chain of this family of compounds [26]. We have recently employed a previously reported palladium-catalyzed cross coupling between *in situ*-generated carboxyl anhydrides and phenylboronic acid [27–28] as a convenient way to obtain 24-phenyl-24-oxo steroids derived from the acetates of different naturally occurring bile acids [29]. Herein we describe the application of this methodology to synthesis of novel bile acids in which the side chain has been enlarged by attaching a 4-carboxyphenyl moiety at position C-24.

2. Experimental

The starting acetylated bile acids **1a–c** [29] were prepared following the standard Ac_2O/pyr/DMAP procedure. Reactions were monitored by TLC on ALUGRAM® SIL G/UV254 plates from MACHEREY-NAGEL. Chromatographic plates were sprayed with a 1% solution of vanillin in 50% $HClO_4$ and heated until color developed. Melting points were measured on a Melt-Temp II apparatus. Mass spectra were registered in a Thermo-Electron and Jeol-SX102A spectrometers. NMR spectra were recorded in $CDCl_3$ or DMSO-d_6 solutions in a Varian INOVA 400 spectrometer using the solvents signal as references. NMR signals assignments were carried out with the aid of a combination of 1D and 2D NMR techniques that included ^1H, ^{13}C, COSY, Nuclear Overhauser Effect Spectroscopy (NOESY), Heteronuclear Single Quantum Correlation (HSQC) and Heteronuclear Multiple Bond Correlation (HMBC) [30]. Mono-crystal X-ray diffraction of compound **3c** corroborated the obtained structure [31].

2.1. General procedure for palladium-catalyzed carboxyl anhydride-boronic acid cross coupling

$Pd(AcO)_2$ (10.05 mg, 0.045 mmol), the acetylated bile acid **1a–c** (1 mmol), pivalic anhydride (418.25 mg, 0.375 mL, 2.25 mmol in

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THF 4 mL), water (67.5 mg, 0.067 mL, 3.75 mmol), 4-methoxycarbonylphenylboronic acid (323.95 mg, 1.8 mmol in THF, 2 mL) and tris(4-methoxyphenyl)phosphine (37.05 mg, 0.105 mmol) were mixed in this order under sonication and the flask was purge 3 times with Ar. The resulting mixture was stirred at 60 °C for 24 h and the solvent was evaporated. Purification in a chromatographic column packed with silica gel employing a gradient of hexane/ethyl acetate as eluent afforded the corresponding coupling product.

2.1.1. 3α-Acetoxy-24-(4′-methoxycarbonylphenyl)-5β-cholan-24-one (**2a**)

Yield 343.5 mg, 64%. Mp 198.9–200.5 °C (*from CH_2Cl_2/hexane*). ^{1}H NMR ($CDCl_3$, 400 MHz) δ ppm: 8.11 (*d*, *J* = 8.5 Hz, 2H, H-3′), 7.99 (*d*, *J* = 8.4 Hz, 2H, H-2′), 4.71 (tt, *J* = 11.3, 4.8 Hz, 1H, H-3β), 3.94 (s, 3H, OCH_3), 3.01 (ddd, *J* = 16.1, 10.0, 4.8 Hz, 1H, H-24a), 2.90 (ddd, *J* = 16.4, 9.4, 5.8 Hz, 1H, H-24b), 2.02 (d, *J* = 1.4 Hz, 3H, CH_3 acetyl), 0.97 (*d*, *J* = 6.3 Hz, 3H, H-21), 0.92 (*d*, *J* = 1.4 Hz, 3H, H-19), 0.65 (s, 3H, H-18). ^{13}C NMR (100.52 MHz) δ ppm: 35.0 C-1, 26.3 C-2, 74.4 C-3, 32.3 C-4, 41.8 C-5, 27.0 C-6, 26.6 C-7, 35.8 C-8, 40.4 C-9, 34.6 C-10, 20.8 C-11, 40.1 C-12, 42.7 C-13, 56.5 C-14, 24.2 C-15, 28.3 C-16, 56.1 C-17, 12.0 C-18, 23.3 C-19, 35.5 C-20, 18.5 C-21, 30.2 C-22, 35.9 C-23, 200.4 C-24, 133.6 C-1′, 127.9 C-2′, 129.8 C-3′, 140.3 C-4′, 166.3 $\underline{C}OOCH_3$ (C-5′), 52.4 $COO\underline{C}H_3$, 21.5 CH_3 acetyl, 170.6 C=O acetyl. MS (EI, 70 Ev), *m*/*z* (%): 537 (0.06, MH^+), 536 (0.12, M^+), 315 (20), 299 (13), 298 (27), 283 (17), 257 (28), 256 (26), 255 (94), 230 (11), 229 (13), 217 (11), 216 (18), 215 (44), 203 (11), 201 (19), 191 (13), 179 (12), 178 (49), 175 (12), 173 (12), 163 (100), 161 (28), 159 (20), 149 (15), 147 (38), 145 (18), 135 (32), 134 (15), 133 (22), 131 (12), 121 (22), 120 (12), 119 (21), 109 (20), 108 (19), 107 (34), 106 (14), 105 (33), 104 (12), 97 (11), 95 (31), 94 (13), 93 (38), 91 (29), 83 (16), 81 (35), 79 (28), 77 (17), 71 (10), 69 (19), 67 (23), 57 (17), 55 (30), 43 (31), 41 (18), 40 (11).

2.1.2. 3α,6α-Diacetoxy-24-(4-methoxycarbonyl-phenyl)-5β-cholan-24-one (**2b**)

Yield 445.7 mg, 75%. Mp 146.7–148.3 °C (from benzene). ^{1}H NMR ($CDCl_3$, 400 MHz) δ ppm: 8.15–8.08 (m, 2H, H-3′), 8.03–7.94 (m, 2H, H-2′), 5.14 (dt, *J* = 12.3, 4.8 Hz, 1H, H-6β), 4.69 (ddd, *J* = 16.0, 11.2, 4.7 Hz, 1H, H-3β), 3.94 (s, 1H, OCH_3), 3.00 (ddd, *J* = 15.0, 10.0, 5.0 Hz, 1H, H23a), 2.95–2.85 (m, 1H, H-23b), 2.03 (s, 3H, CH_3 acetyl), 2.00 (s, 3H, CH_3 acetyl), 0.97 (*d*, *J* = 5.5 Hz, 3H, H-21), 0.96 (s, 3H, H-19), 0.65 (s, 3H, H-18). ^{13}C NMR (100.52 MHz) δ ppm: 35.0 C-1, 26.4 C-2, 73.6 C-3, 26.2 C-4, 45.3 C-5, 70.9 C-6, 31.3 C-7, 34.6 C-8, 39.9 C-9, 36.0 C-10, 20.7 C-11, 39.9 C-12, 42.9 C-13, 56.1 C-14, 24.1 C-15, 28.2 C-16, 56.1 C-17, 12.0 C-18, 23.2 C-19, 35.5 C-20, 18.6 C-21, 30.2 C-22, 35.9 C-23, 200.3 C-24, 133.7 C-1′, 127.9 C-2′, 129.8 C-3′, 140.3 C-4′, 166.2 $\underline{C}OOCH_3$, 52.4 $COO\underline{C}H_3$, 21.4, 21.4 CH_3 acetyl, 170.4, 170.4 C=O acetyl. MS (EI, 70 Ev), *m*/*z* (%): 594 (0.5, M^+), 475 (15, MH^+-2xCH_3COOH), 474 (34, M^+-2xCH_3COOH), 459 (14), 448 (10), 374 (18), 373 (70), 356 (18), 313 (14), 298 (16), 297 (68), 296 (40), 281 (10), 256 (18), 255 (79), 254 (13), 253 (19), 229 (13), 228 (31), 227 (13), 219 (13), 215 (25), 214 (20), 213(46), 201 (10), 199 (15), 191 (10), 185 (15), 179 (14), 178 (91), 173 (17), 171 (12), 164 (11), 163 (100), 161 (13), 160 (12), 159 (33), 157 (10), 149 (10), 147 (20), 145 (30), 143 (10), 135 (17), 133 (20), 131 (15), 121 (20), 120 (14), 119 (16), 109 (14), 107 (26), 105 (22), 95 (34), 93 (24), 91 (12), 83 (10), 81 (30), 79 (11), 69 (14), 55 (16), 43 (25).

2.1.3. 3α,7α-Diacetoxy-24-(4′-methoxycarbonylphenyl)-5β-cholan-24-one (**2c**)

Yield 407.6 mg, 69%. Mp. 134.7–135.9 °C (*from CH_2Cl_2/hexane*). ^{1}H NMR ($CDCl_3$, 400 MHz) δ ppm: 8.11 (d, *J* = 8.7 Hz, 2H, H-3′), 7.99 (*d*, *J* = 8.7 Hz, 2H, H-2′), 4.87 (q, *J* = 3.3 Hz, 1H, H-7β), 4.58 (tt, *J* = 11.4, 4.5 Hz, 1H, H-3β), 3.94 (s, 3H OCH_3), 3.20–2.62 (m, 2H, H-23), 2.04 (s, 3H, CH_3 acetyl), 2.02 (s, 3H CH_3 acetyl), 0.98 (*d*, *J* = 6.3 Hz, 3H, H-21), 0.93 (s, 3H, H-19), 0.66 (s, 3H, H-21). ^{13}C NMR (100.52 MHz) δ ppm: 34.6 C-1, 26.8 C-2, 74.2 C-3, 32.9 C-4, 40.9 C-5, 31.3 C-6, 71.2 C-7, 37.9 C-8, 34.1 C-9, 34.8 C-10, 20.7 C-11, 39.5 C-12, 42.7 C-13, 50.4 C-14, 23.6 C-15, 28.1 C-16, 55.9 C-17, 11.7 C-18, 22.7 C-19, 35.5 C-20, 18.6 C-21, 30.2 C-22, 35.9 C-23, 200.3 C-24, 133.7 C-1′, 127.9 C-2′, 129.8 C-3′, 140.3 C-4′, 166.2 $\underline{C}OOCH_3$ (C-5′), 52.4 $COO\underline{C}H_3$, 21.5, 21.6 CH_3 acetyl, 170.4, 170.6 C=O acetyl. MS (EI, 70 Ev), *m*/*z* (%): 595 (0.014, MH^+), 594 (0.06, M^+), 475 (11), 474 (30), 459 (24), 315 (11), 313 (17), 297 (20), 296 (20), 256 (15), 255 (67), 254 (12), 253 (39), 229 (14), 228 (16), 227 (11), 219 (12), 215 (10), 214 (11), 213 (33), 201 (26), 199 (16), 191 (13), 185 (14), 178 (24), 173 (15), 171 (15), 164 (11), 163 (100), 161 (14), 160 (11), 159 (30), 157 (17), 155 (12), 147 (23), 145 (29), 143 (17), 135 (25), 134 (14), 133 (22), 131 (24), 129 (11), 121 (15), 120 (11), 119 (24), 117 (12), 107 (21), 106 (12), 105 (40), 95 (18), 94 (14), 93 (21), 91 (25), 81 (18), 79 (17), 77 (10), 60 (14), 45 (14), 43 (19).

2.2. General procedure for Wolff-Kishner reduction

A solution of KOH (336 mg, 6 mmol) in water (1.5 mL) was added to a suspension of the steroid **2a–c** (1 mmol) in ethylene glycol (16 mL) and the mixture was stirred at 120 °C for 1 h. Hydrazine hydrate solution (3.2 mL) was added to the cooled mixture and the resulting solution was stirred at 120 °C for 1 h followed by addition of a solution of KOH (526 mg) in water (2.6 mL) and the mixture was distilled until the temperature raised to 198 °C. The mixture was refluxed for 4 h, cooled in an ice bath, acidified to pH 2 with 10% HCl and extracted with diethyl ether (2 × 30 mL). The organic layer was washed with water (2 × 20 mL), dried (anh. Na_2SO_4) and evaporated. Purification in a chromatographic column packed with silica gel employing ethyl acetate/methanol 95/5 as eluent afforded the desired modified bile acid.

Table 1
Crystal data and structure refinement for compound **3c**.

Compound 3c	Parameters
Empirical formula	$C_{31}H_{46}O_4$
Formula weight	482.68
Temperature	130 (2) K
Wavelength	1.54184 Å
Crystal system	Monoclinic
Space group	$P2_1$
Unit cell dimensions	*a* = 12.423 (4) Å *b* = 10.3324 (14) Å *c* = 21.061 (7) Å β = 100.21 (3)°
Volume	2660.6 (13) Å^3
Z	4
Density (calculated)	1.205 Mg/m^3
Absorption coefficient	0.606 mm^{-1}
F(000)	1056
Crystal size	0.480 × 0.140 × 0.037 mm^3
Theta range for data collection	3.615–73.866°.
Index ranges	−15<=*h*<=14, −12<=*k*<=8, −25<=*l*<=24
Reflections collected	9804
Independent reflections	7555 [*R*(int) = 0.0682]
Completeness to theta = 67.684°	99.8%
Refinement method	Full-matrix least-squares on F^2
Data/restraints/parameters	7555/7/655
Goodness-of-fit on F^2	1.001
Final *R* indices [*I* > 2sigma(I)]	*R*1 = 0.0578, w*R*2 = 0.1278
R indices (all data)	*R*1 = 0.0876, w*R*2 = 0.1555
Absolute structure parameter	−0.1 (4)
Largest diff. peak and hole	0.240 and −0.273 e Å^{-3}