Synthesis of a 6*H*-Pyrazolo[4,5,1-*de*]acridin-6-one Derivative: A Useful Intermediate of Antitumour Agents

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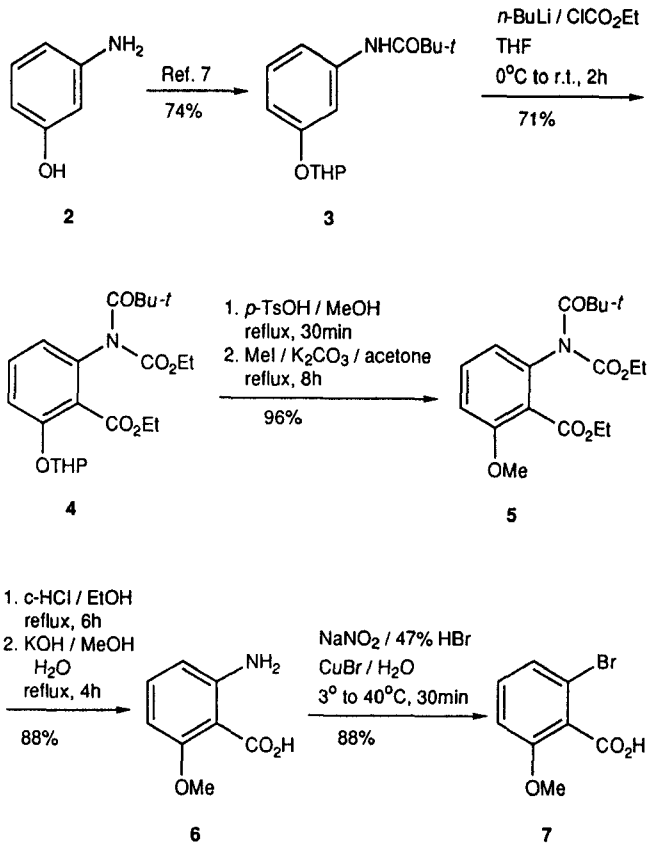
A 6*H*-pyrazolo[4,5,1-*de*]acridin-6-one derivative, a useful intermediate of antitumor agents, was prepared by a facile synthetic route from 2-bromobenzoic acid and 6-nitroindazole involving a halogenocopper(I)-catalyzed Ullmann coupling reaction and Friedel–Crafts cyclization.

We have investigated novel coplanar chromophores for DNA intercalating antitumor agents and 6*H*-pyrazolo[4,5,1-*de*]acridin-6-ones, which are abbreviated as pyrazoloacridones, show intercalation activity in an ethidium-binding assay.[1] We report here a new synthetic pathway to 5-bromo-7-methoxy-2-methylpyrazoloacridone (**1a**), which is a key intermediate of pyrazoloacridone antitumor agents.

Unsubstituted pyrazoloacridone has already attracted attention as a dyestuff and was synthesized in 1964[3] from a phenylhydrazone derivative of methyl 2,4-dinitrophenylglyoxylate, which was prepared[2] by the diazotization of anthranilic acid followed by coupling with methyl 2,4-dinitrophenylacetate. In this method, all the substituents were removed at the later stage of the synthesis. From the view point of antitumor activities, we needed pyrazoloacridones having a 7-hydroxy substituent and side chains at the C-2 and/or C-5 positions of pyrazoloacridones. Moreover, the synthetic route should be convenient and readily amenable to large scale work for further pharmaceutical and clinical studies. Our synthetic strategy is to build the pyrazoloacridone skeleton by Ullmann coupling reaction of 2-bromo-6-methoxybenzoic acid and indazole, followed by Friedel–Crafts cyclization. For the indazole we could make use of the known 3-methyl-6-nitroindazole (**8**), since the 3-methyl group, instead of the carboxy group, would be stable under a high temperature in the cyclization step and its hydrogen could be converted to the bromine by bromination with *N*-bromosuccinimide.

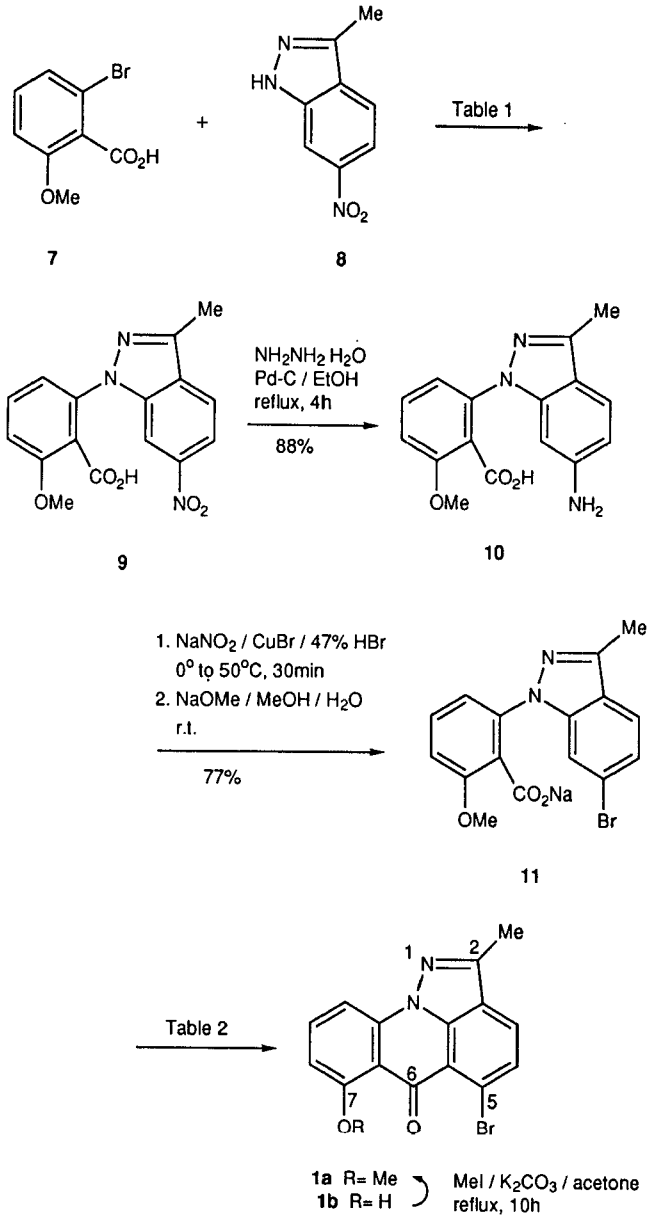
Although a synthesis of 2-bromo-6-methoxybenzoic acid (**7**) from *m*-dinitrobenzene has been reported,[4] it suffered from a poor overall yield (less than 10%). We required large quantities of the 2-amino-6-methoxybenzoic acid (**6**), because it could be readily converted to the corresponding bromide **7** by the Sandmeyer reaction. Paquette and co-workers described a route to 2-amino-6-methoxybenzoic acid (**6**) from *m*-dinitrobenzene[5] but the yield was also poor (17%). An improved method from 2,6-dinitrobenzoic acid was reported in 45% yield,[6] unfortunately the overall yield dropped to 27% if the preparation of the 2,6-dinitrobenzoic acid from the commercially available 2,6-dinitrotoluene was included. Thus, we attempted a more efficient synthesis from another commercially available material, 3-hydroxyaniline (**2**) (Scheme 1). The hydroxy and amino groups were protected by dihydropyran and pivaloyl chloride, respectively, by the method described already.[7] Then, ethoxycarbonyl groups were introduced both to amide nitrogen and aromatic carbon by lithiation with 2 equivalents of butyllithium followed by substitution with ethyl chloroformate. After the tetrahydropyranyl group was cleaved, the resultant phenol was converted to the methyl ether **5**. Since the basic deprotection of the amino and carboxyl groups needed very severe conditions (refluxing in 10 N aqueous sodium hydroxide), stepwise deprotections were adopted under mild acidic and subsequent basic conditions. After the pivaloyl group was removed first by acid, 3 N potassium hydroxide in aqueous methanol was enough to hydrolyse the ethyl ester. The overall yield of **7** from 3-hydroxyaniline (**2**) was 39%.



Scheme 1

3-Methyl-6-nitroindazole (**8**) was obtained readily from 2-ethyl-5-nitroaniline[8] by the method published already.[9] Ullmann coupling reaction of the bromide **7** and the indazole **8** proceeded with copper(II) oxide in nitrobenzene, however, the reaction temperature needed was more than 160 °C. This temperature is not suitable to a large scale synthesis. In order to lower the temperature, we screened several halogenocopper(I) compounds as cata-

lysts and found the reaction proceeded smoothly with them in dimethylformamide at 100 °C (Table 1). Copper(I) iodide was the most effective and the reaction was complete in 4 hours, although at this temperature the reaction did not proceed with copper(II) oxide. The nitro group of **9** was reduced to the amine **10** by a treatment with hydrazine and palladium-on-charcoal, then **10** was converted to the bromide **11** by Sandmeyer reaction (Scheme 2). Friedel–Crafts cyclization (Table 2) with both sulfuric acid and trifluoromethanesulfonic acid were effective at 100 °C. No reactions were observed at 150 °C with polyphosphoric acid (PPA). When trifluoromethanesulfonic acid was used, the reaction proceeded cleanly and the yield was higher than in the case of sulfuric acid. At this stage the methyl ether at C-7 was partially cleaved, but it was readily regenerated by reaction with methyl iodide and potassium carbonate.



Scheme 2

In summary, we have developed a facile synthetic pathway to a pyrazoloacridone derivative in large scale, starting from 2-bromobenzoic acid and indazole using the Ullmann coupling reaction, followed by a Friedel–Crafts cyclization under mild conditions.

Table 1. Copper-Catalyzed Coupling Reaction of **7** and **8**

Catalyst[a]	Solvent	Conditions		Yield[b] (%)
		Time (h)	Temp. (°C)	
CuI	DMF	4	100	85
CuBr	DMF	7.5	100	71.8
CuCl	DMF	7.5	100	66.1
CuCl	$PhNO_2$	7.5	100	34.1
CuO	DMF	18	100	no reaction
CuO	$PhNO_2$	4.5	160	78

[a] 9 mol% of each catalyst is used.
[b] Isolated yield of **9**.

Table 2. Cyclization of **11**

Acid	Conditions		Yield[a] (%)	Ratio[b] **1a** : **1b**
	Time (h)	Temp. (°C)		
H_2SO_4	5	100	42.7	90.2 : 6.6
CF_3SO_3H	4.5	100	94.4	58.4 : 32.7
PPA	4	150	no reaction	

[a] Isolated yield of a mixture of **1a** and **1b**.
[b] Ratios are determined by HPLC analysis.

Melting points were determined with a Yanagimoto hot-stage microscope and are uncorrected. ^{1}H NMR spectra were recorded on a Bruker AC300 spectrometer with TMS as an internal standard. IR spectra were recorded on a Shimadzu FTIR-4300 spectrophotometer. Mass spectra were recorded on a Hitachi M-80B mass spectrometer. All reagents and solvents were of commercial quality. Microanalytical results (C,H,N) of all new compounds are within ± 0.4% of the theoretical values and were determined on a Yanaco MT-3 CHN apparatus.

The protected hydroxyaniline **3**[7] and the indazole **8**[9] were prepared according to the reported procedures.

Ethyl 2-(*N*-Ethoxycarbonyl-*N*-pivaloyl)amino-6-(tetrahydropyran-2-yloxy)benzoate (4):

To a solution of **3**[7] (131 g, 0.47 mol) in THF (1.2 L) was added dropwise 15% BuLi in hexane (616 mL, 0.99 mol) below 0 °C and the mixture was further stirred at the same temperature for 4 h. To this solution was added dropwise ethyl chloroformate (90.6 mL, 0.95 mol) below 0 °C, the mixture was stirred at r.t. for 2 h, then poured into ice-cooled water, and partitioned. The organic layer was washed with sat. $NaHCO_3$, brine, and dried ($MgSO_4$). The solvent was evaporated and the residue was recrystallized from CH_2Cl_2/hexane affording **4** as colorless prisms; yield: 142 g (71%); mp 82–83 °C.

^{1}H NMR ($CDCl_3$/TMS): δ = 1.19 (t, 3H, J = 7.0 Hz), 1.31–1.41 (m, 12H), 1.55–1.94 (m, 6H), 3.60 (br, 1H), 3.92 (dt, 1H, J = 11.1, 3.0 Hz), 4.16 (q, 2H, J = 7.0 Hz), 4.26–4.40 (m, 2H), 5.48 (br, 1H), 6.76 (dd, 1H, J = 7.8, 0.9 Hz), 7.20 (dd, 1H, J = 8.4, 0.9 Hz), 7.32 (dd, 1H, J = 8.4, 7.8 Hz).

IR (KBr): ν = 1740, 1715, 1600, 1460 cm^{-1}.

CI-MS: m/z (%) = 422 ($M^+ + 1$, 4), 338 (70), 292 (100), 254 (33).

Ethyl 2-(*N*-Ethoxycarbonyl-*N*-pivaloyl)amino-6-methoxybenzoate (5):

The ester **4** (451 g, 1.07 mol) was dissolved in MeOH (1 L) at 50 °C and cooled to r.t. To this solution was added *p*-TsOH · H_2O (9.5 g, 0.05 mol) and the mixture was stirred at r.t. for 30 min. After evaporation of the solvent, EtOAc and H_2O were added to the residue and partitioned. The organic layer was washed with sat. aq $NaHCO_3$, brine, dried ($MgSO_4$), concentrated and the residue was dissolved in acetone (1.2 L). To the solution were added K_2CO_3 (276 g, 2.00 mol), MeI (311 mL, 500 mol), and the mixture was refluxed for 8 h. After cooling to r.t., the mixture was filtered, the filtrate was concentrated, and the residue was dissolved in $CHCl_3$. The solution was washed with H_2O, brine, dried ($MgSO_4$), and concentrated. The residue was recrystallized from MeOH/H_2O to afford **5** as colorless plates; yield: 362 g (96%); mp 64–65 °C.

^1H NMR ($CDCl_3$/TMS): δ = 1.17 (t, 3H, J = 7.0 Hz), 1.31 (t, 3H, J = 7.0 Hz), 1.32 (s, 9H), 3.82 (s, 3H), 4.14 (q, 2H, J = 7.0 Hz), 4.30 (q, 2H, J = 7.0 Hz), 6.72 (dd, 1H, J = 8.1, 0.9 Hz), 6.91 (d, 1H, J = 8.4 Hz), 7.35 (dd, 1H, J = 8.4, 8.1 Hz).

IR (KBr): ν = 1740, 1710, 1605, 1475 cm^{-1}.

EI-MS: m/z (%) = 351 (M^+, 8), 267 (76), 221 (33), 57 (100).

2-Amino-6-methoxybenzoic Acid (6):

To a stirred ethanolic (1 L) solution of **5** (359 g, 1.02 mol) was added conc. HCl (330 mL) and the mixture was refluxed for 6 h. The solvent was evaporated until the volume of the residue was about 0.5 L, then to the residue was added MeOH (800 mL), 13 N aq KOH (330 mL), and the mixture was refluxed for **4** h. After evaporation of MeOH, the pH of the solution was adjusted to 5.6 by adding conc. HCl, then CH_2Cl_2 (500 mL) was added and the pH of the stirring mixture was adjusted to 2.5 by adding conc. HCl. To the mixture were added CH_2Cl_2 and H_2O and partitioned. The aqeous layer was extracted with CH_2Cl_2, the combined organic layers were dried ($MgSO_4$) and concentrated. The residue was recrystallized from *i*-PrOH affording **6** as colorless prisms; yield: 151 g (88%); mp 88–89 °C (Lit.[5] mp 71–75 °C, Lit.[6] mp 84–85 °C).

^1H NMR ($CDCl_3$/TMS): δ = 3.93 (s, 3H), 6.19 (dd, 1H, J = 8.1, 0.9 Hz), 6.20–6.36 (br, 2H), 6.34 (dd, 1H, J = 8.4, 0.9 Hz), 7.16 (dd, 1H, J = 8.4, 8.1 Hz), 11.32 (br, 1H).

IR (KBr) ν = 3450, 3350, 3200, 1690, 1620, 1560, 1475, 1445 cm^{-1}.

EI-MS: m/z (%) = 167 (M^+, 57), 149 (100), 122 (37), 107 (43).

2-Bromo-6-methoxybenzoic Acid (7):

A mixture of **6** (650 g, 3.89 mol), H_2O (1 L), and 47% aq HBr (1.56 L) was heated at 55 °C until the solid was dissolved. Then the solution was cooled to −8 °C and to the solution were simultaneously added aq $NaNO_2$ (7.8 M, 500 mL, 3.9 mol) and ice (700 g) portionwise below 3 °C. The obtained orange solution was added to a warm (40 °C) mixture of CuBr (308 g, 2.15 mol), 47% aq HBr (310 mL), and $CHCl_3$ (1.9 L), the stirring was continued at 40 °C for 30 min and the mixture was partitioned. The aqueous layer was extracted with $CHCl_3$ and the combined organic layers were washed with 0.5 N HCl, dried ($MgSO_4$), concentrated and the solid residue was washed with toluene; yield: 789 g (88%); mp 124–127 °C (Lit.[4] mp 126–128 °C).

^1H NMR ($CDCl_3$/TMS): δ = 3.86 (s, 3H), 6.89 (dd, 1H, J = 8.1, 0.9 Hz), 7.17 (dd, 1H, J = 8.1, 0.9 Hz), 7.23 (dd, 1H, J = 8.1, 8.1 Hz), 8.6 (br, 1H).

IR (KBr): ν = 3450, 1720, 1640, 1590, 1575, 1470, 1430 cm^{-1}.

EI-MS: m/z (%) = 232/230 (M^+, 98/100), 215/213 (73/78), 202/200 (29/35), 185/183 (94/88).

1-(2-Carboxy-3-methoxy)phenyl-3-methyl-6-nitro-1*H*-indazole (9):

A mixture of the bromide **7** (509 g, 2.20 mol), indazole **8**[9] (410 g, 2.31 mol), K_2CO_3 (334 g, 2.42 mol), and CuI (37.7 g, 0.20 mol) were stirred in DMF (6.6 L) at 100 °C for 6 h. After cooling, the precipitated solid was filtered and to the filtrate was added H_2O (6 L), the mixture was warmed to 70 °C, and dissolved. At this temperature the solution was treated with activated carbon for 15 min. After filtration, conc. HCl (330 mL) was added and the mixture was cooled to afford **9** as yellow needles (634 g, 84%); mp 220–221 °C.

^1H NMR (DMSO-d_6/TMS): δ = 2.62 (s, 3H), 3.90 (s, 3H), 7.27 (d, 1H, J = 8.1 Hz), 7.30 (d, 1H, J = 8.4 Hz), 7.65 (dd, 1H, J = 8.4, 8.2 Hz), 8.08 (d, 1H, J = 8.9 Hz), 8.03 (d, 1H, J = 8.7 Hz), 8.23 (s, 1H).

IR (KBr): ν = 3400, 1720, 1690, 1585, 1520, 1470, 1435 cm^{-1}.

EI-MS: m/z (%) = 327 (M^+, 41), 283 (100), 237 (25).

6-Amino-1-(2-carboxy-3-methoxy)phenyl-3-methyl-1*H*-indazole (10):

To a stirred mixture of **9** (579 g, 1.77 mol), 10% Pd/C (wet, 50% H_2O, 29 g), and EtOH (5.8 L) was added $N_2H_4 \cdot H_2O$ (290 mL, 5.98 mol) at 50 °C, and the mixture was refluxed for 2 h. Ater filtration of the precipitate, the solvent was evaporated. To the residue was added H_2O (2.5 L) for dissolving the partially crystallized solid, and 47% aq HBr (ca. 220 mL) until the pH of the solution was 3 to afford **10** as colorless prisms; yield: 633 g (88%); mp 213–214 °C.

^1H NMR (DMSO-d_6/TMS): δ = 2.36 (s, 3H), 3.85 (s, 3H), 6.53 (s, 1H), 6.54 (d, 1H, J = 9.0 Hz), 7.10 (d, 2H, J = 8.2 Hz), 7.37 (d, 1H, J = 9.0 Hz), 7.51 (t, 1H, J = 8.2 Hz).

IR (KBr): ν = 3390, 3300, 1730, 1625, 1590, 1515, 1480, 1440, 1410 cm^{-1}.

EI-MS: m/z (%): = 297 (M^+), 253 (100), 223 (30).

6-Bromo-1-(2-carboxy-3-methoxy)phenyl-3-methyl-1*H*-indazole Sodium Salt (11):

The amine **10** (409 g, 1.38 mol) was dissolved in MeOH (4 L) and 47% aq HBr (1.6 L). To the solution cooled at −10 °C, was added $NaNO_2$ (95.9 g, 1.39 mol) in H_2O (200 mL), then the mixture was added slowly to a warmed (50 °C) suspension of CuBr (107 g, 0.74 mol) in 47% aq HBr (230 mL) and stirred at r.t. for 1 h. After evaporation of MeOH, $CHCl_3$ and H_2O were added to the residue. The mixture was partitioned and the organic layer was dried ($MgSO_4$), treated with the activated carbon, and concentrated. To the residue were added MeOH (1 L), methanolic NaOMe (28%, 350 mL), and H_2O (200 mL) to afford the sodium salt **11** as a colorless solid; yield: 418 g (77%); mp > 300 °C.

^1H NMR (DMSO-d_6/TMS): δ = 2.50 (s, 3H), 3.77 (s, 3H), 6.73 (d, 1H, J = 7.4 Hz), 7.02 (d, 1H, J = 7.8 Hz), 7.18–7.21 (m, 2H), 7.64 (d, 1H, J = 8.3 Hz), 7.67 (s, 1H).

IR (KBr): ν = 3400, 1610, 1515, 1470, 1435, 1405 cm^{-1}.

5-Bromo-7-methoxy-2-methyl-6*H*-pyrazolo[4,5,1-*de*]acridin-6-one (1a):

The indazole **11** (5.0 g, 13.1 mmol) was heated in CF_3SO_3H (75 mL) at 100 °C for 4 h, cooled and the solution was poured into a mixture of $CHCl_3$ and H_2O and partitioned. The H_2O layer, which contained some precipitate, was extracted with $CHCl_3$ and the combined organic layer was washed with H_2O and concentrated. To the residue was added *i*-PrOH to afford a yellow solid (4.5 g, 6:4 mixture of **1a** and **1b**).[10] A mixture of this solid (4.5 g), MeI (100 mL, 1.6 mol), and K_2CO_3 (2 g, 14 mmol) was refluxed in acetone (2 L) for 10 h. After filtration of the precipitate, the filtrate was concentrated and the residue was dissolved in $CHCl_3$, then the solution was washed with brine, dried ($MgSO_4$), and concentrated. The residue was recrystallized from CCl_4 affording **1a** as yellow needles; yield: 4.2 g (94%); mp 220–221 °C.

^1H NMR ($CDCl_3$/TMS): δ = 2.65 (s, 3H), 3.97 (s, 3H), 6.80 (dd, 1H, J = 8.3, 1.0 Hz), 7.59 (d, 1H, J = 8.2 Hz), 7.60 (t, 1H, J = 8.3 Hz), 7.68 (d, 1H, J = 8.2 Hz), 7.74 (dd, 1H, J = 8.3, 1.0 Hz).

IR (KBr): ν = 1660, 1605, 1520 cm^{-1}.

EI-MS: m/z (%) = 344/342 (M^+, 99/100), 315/313 (43/56), 298/296 (47/47).

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