The Synthesis of Pyranoacridinone Inhibitors of Protein Tyrosine Kinases

Paul W. Groundwater* and Kevin R. H. Solomons
School of Chemistry and Applied Chemistry, University of Wales College of Cardiff, PO Box 912, Cardiff CF1 3TB, UK

7-Oxo-5,6,7,8-tetrahydroflavone **10** reacts with anthranilonitrile and ethyl anthranilate to give the corresponding enamines, **11** and **13**. These enamines undergo base-catalysed cyclization to pyrano[2,3-*a*]acridin-4-ones, **12** and **14**, which undergo oxidation to the fully aromatic systems, **4** and **5**. Biological testing of some of these fused heterocyclic systems shows them to have potential in cancer chemotherapy as inhibitors of growth factor-mediated cell proliferation.

Protein tyrosine kinases (PTKs), enzymes which catalyse the transfer of phosphate from a donor such as ATP to the tyrosine residue of a peptide substrate, form an integral part of the cell surface receptors of several growth factors including epidermal growth factor (EGF), platelet-derived growth factor (PDGF) and insulin. PTKs are also the products of several oncogenes and the cellular genes from which they originate (proto-oncogenes).[1] Binding of the appropriate growth factor to the extracellular domain of the receptor results in receptor activation and leads, ultimately, to cell proliferation *via* a cascade of protein tyrosine kinases.[2] These enzymes thus play a crucial role in the general functioning and development of human cells.

However, in a number of diseases, including breast cancer, ovarian cancer, gastric cancer, atherosclerosis and psoriasis,[3] the cells have lost the ability to regulate the activity of PTKs and these enzymes become continuously activated resulting in the uncontrolled proliferation characteristic of these diseases.

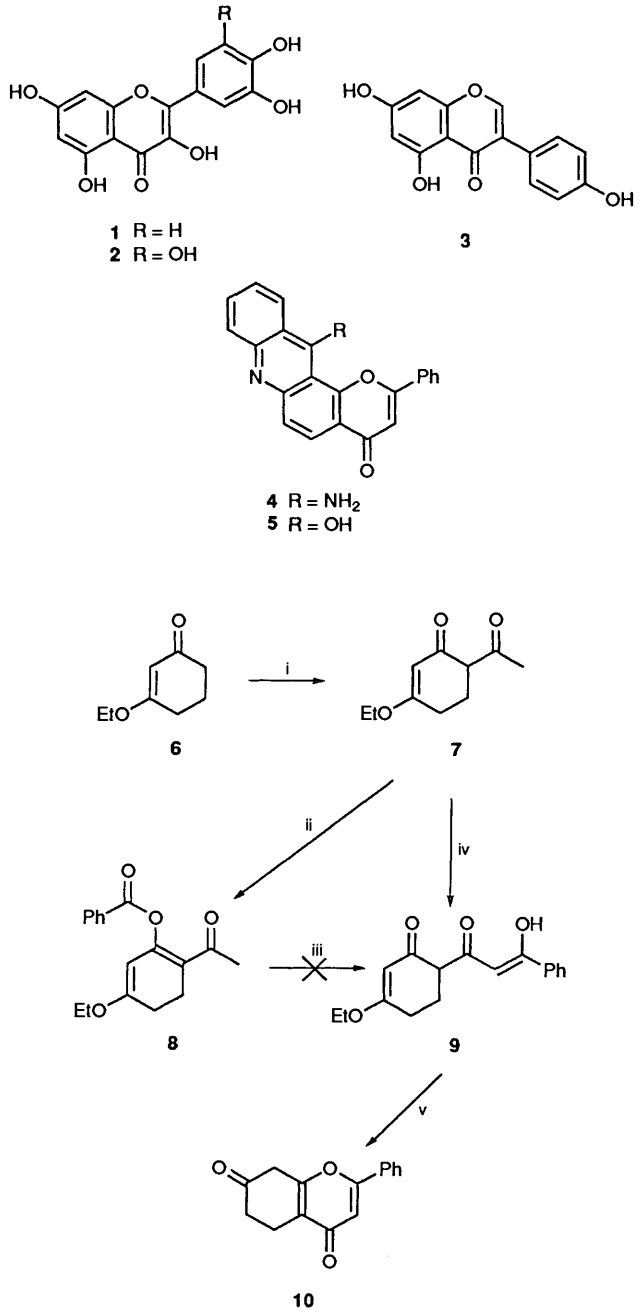
The significance of PTKs in the regulation of cell growth makes them important targets for pharmaceutical intervention. Specific inhibitors of PTKs have potential as chemotherapeutic agents and as molecular tools for defining the signalling pathways in which the PTKs are involved.

Amongst the known inhibitors of the PTKs are the flavones, *e.g.* quercetin **1**[4] and myricetin **2**,[5] and the isoflavones, *e.g.* genistein **3**.[6] Myricetin **2** has been shown to inhibit the PTK activity of the oncogene product pp130fbs by competition with ATP but inhibits the PTK activity of the insulin receptor non-competitively to ATP. Hidaka *et al.*[5] have attempted to explain these observations in terms of a flavone binding site at, or near, the ATP binding site of the oncogene-encoded PTKs but remote from the ATP binding site of the insulin receptor.

As part of a project aimed at synthesizing novel inhibitors of the protein tyrosine kinases we have synthesized 12-amino-2-phenylpyrano[2,3-*a*]acridin-4-one **4** and 12-hydroxy-2-phenyl-pyrano[2,3-*a*]acridin-4-one **5** which incorporate structural elements of both the phosphate donor and the flavones. It was hoped that these species would bind at the flavone/ATP binding site of the oncogene-encoded PTKs specifically and so be specific inhibitors of these PTKs.

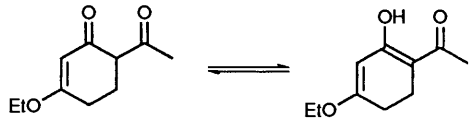
Results and Discussion

The synthesis of the pyranoacridinones, **4** and **5**, was achieved *via* the base-catalysed cyclization of the cyano- and ester-substituted enamines, **11** and **13**, using a modification of the method of Strekowski *et al.*[7] The synthesis of 7-oxo-5,6,7,8-tetrahydroflavone **10**, which was condensed with anthranilonitrile or ethyl anthranilate to give the enamines, is outlined in Scheme 1. Anion formation at the most acidic C-2 position of cyclohexane-1,3-dione was prevented by preparation of 3-



Scheme 1 Synthesis of 7-oxo-5,6,7,8-tetrahydroflavone **10**. *Reagents:* i, NaH, then EtOAc; ii, PhCOCl, pyridine; iii, KOH, pyridine; iv, $NaNH_2$, liq. NH_3, $PhCO_2Me$; v, AcOH, H_2SO_4.

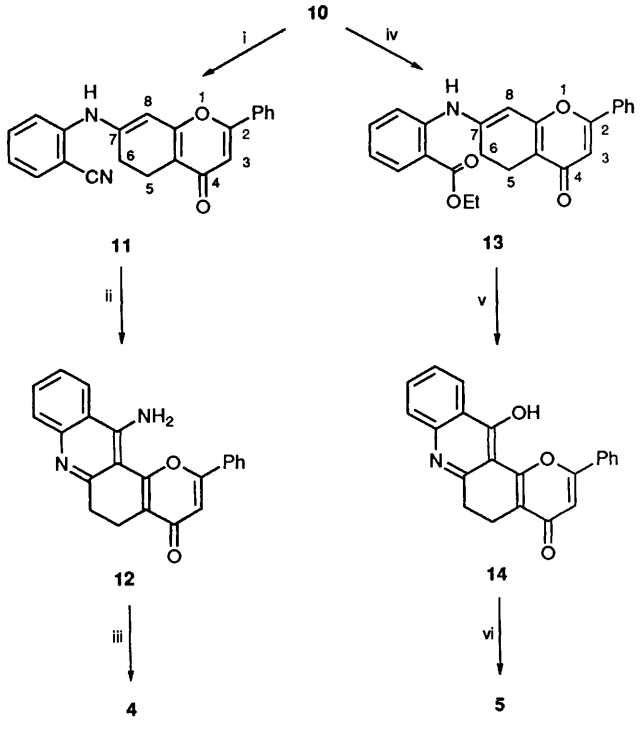
ethoxycyclohex-2-enone **6**.[8] Formation of the anion at the 6-position of this enol ether followed by coupling with ethyl acetate gave 6-acetyl-3-ethoxycyclohex-2-enone **7**, the ^{1}H NMR spectrum of which shows a mixture of the keto **7a** and enol **7b**



7a **7b**

tautomers in the ratio 2:1. *O*-Acylation of the diketone **7**, with benzoyl chloride in pyridine, gave the ester **8** but the Baker–Venkataraman[9] rearrangement of this ester to the triketone **9** was unsuccessful. 1-(4′-Ethoxy-2′-oxocyclohex-3′-enyl)-3-phenylpropane-1,3-dione **9** was, however, obtained directly from the diketone **7** *via* acylation of the diketone dianion with methyl benzoate. Acylation occurred at the least stabilized, most reactive terminal position rather than at the most stabilized, least reactive 6-position. The ^{1}H NMR spectrum of the triketone **9** indicates that, in solution, it exists predominantly (> 95%) as the enol tautomer **9** shown in Scheme 1. Finally, the 7-oxo-5,6,7,8-tetrahydroflavone **10** was prepared by the method of Baker[9] involving acid-catalysed cyclodehydration of the triketone.

Condensation of **10** with the primary aromatic amines was accomplished under the standard conditions for azeotropic removal of water (Scheme 2). Analysis of the ^{1}H NMR spectra

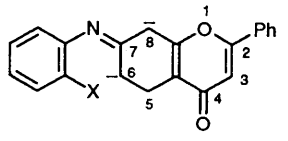


Scheme 2 Synthesis of the pyrano[2,3-*a*]acridin-4-ones **4** and **5**. *Reagents:* i, 2-$H_2NC_6H_4CN$, toluene-*p*-sulfonic acid, $PhCH_3$; ii, $NaNH_2$, liq. NH_3; iii, MnO_2, $PhCH_3$; iv, 2-$H_2NC_6H_4CO_2Et$, toluene-*p*-sulfonic acid, $PhCH_3$; v, $NaNH_2$, 15-crown-5, 1,2-DME; vi, $Hg(OAc)_2$, DMSO.

of the products indicates that they exist primarily as the enamine (vinylogous amide) tautomer. It was expected, by analogy with the formation of the triketone **9**, that formation of the dianion of the enamines **11a** or **13a** would lead to cyclization from the least stabilized 6-position rather than from the most stabilized 8-position. However, even under conditions conducive to dianion formation, the base-catalysed intramolecular cyclization of the enamines onto the cyano **11** or ester **13** groups gave only the products, **12** and **14**, of the cyclization from the anion at the 8-position, with no evidence for the cyclization of the less stabilized 6-anion. Oxidation of the cyclized products with manganese dioxide or mercuric acetate gave the aromatic amino- and hydroxy-substituted pyrano[2,3-*a*]acridin-4-ones **4** and **5**.

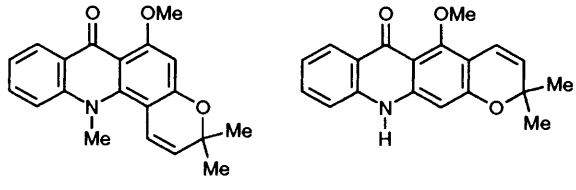
Table 1 Antiproliferative activity of tyrosine kinase inhibitors against DHER cells stimulated by epidermal growth factor (EGF) or calf-serum (CS)

	IC_{50} (μmol dm^{-3})	
Inhibitor	EGF-dependent proliferation	CS-dependent proliferation
4	1.9	⩾10
10	3.1	⩾20
12	5.8	Not determined
15	3.6	⩾32
16	6.5	Not determined



11a X = CN
13a X = CO_2Et

12-Amino-2-phenylpyrano[2,3-*a*]acridin-4-one (APPA) **4** and its dihydro derivative **12** were tested in a biological assay[10] for inhibition of growth factor-mediated cell proliferation along with 7-oxo-5,6,7,8-tetrahydroflavone **10** and some naturally occurring pyranoacridines, acronycine **15** and norisoacronycine **16**[11] (see Table 1). DHER cells, NIH-3T3 cells which over-



15 **16**

express the EGF receptor, can be stimulated to proliferate by either EGF or calf-serum. The inhibition of this proliferation can be assayed by monitoring of (^{3}H-Me)thymidine uptake by cells treated with the inhibitor and comparing this with that of the control cells. The data in Table 1 shows that APPA **4** is a selective inhibitor of the EGF-dependent proliferation of these cells with an IC_{50} of 1.9 μmol compared to that for inhibition of the calf-serum-dependent proliferation of ⩾ 10 μmol.

In another assay breast carcinoma cells were treated with APPA **4** at a concentration of 10 μmol and it was found that after 3 and 6 h the phosphotyrosine levels in the EGF receptor were reduced to approximately 50% of the control and that after 24 h the phosphotyrosine levels had been reduced to approximately 20% of the control. In addition, the entire pattern of EGF-stimulated phosphotyrosine proteins in the cells had been significantly decreased.

Experimental

M.p.s were determined on a Kofler hot-stage and are uncorrected. Elemental analyses were performed on a Perkin-

Elmer 240B. IR spectra were recorded on a Perkin-Elmer 1600 series FTIR spectrophotometer using sodium chloride plates. UV spectra were obtained on a Perkin-Elmer Lambda 2 spectrophotometer. ^{1}H NMR and ^{13}C NMR spectra were acquired on a Bruker WM360 spectrometer at 360 and 90 MHz respectively. ^{1}H NMR coupling constants are given in Hz and all chemical shifts are relative to an internal standard of tetramethylsilane. Low resolution electron impact mass spectra were obtained on a Varian CH5-D spectrometer (Cardiff) and high resolution EI spectra on a VG ZAB-E spectrometer (SERC Mass Spectrometry Service Centre, Swansea). Thin layer chromatography was performed on Merck silica gel $60F_{254}$ and dry-column flash chromatography on Merck silica gel 60H. 1,2-Dimethoxyethane (1,2-DME), diethyl ether (referred to as ether) and tetrahydrofuran were dried from sodium/benzophenone. 3-Ethoxycyclohex-2-enone **6** was prepared by the method of Gannon and House.[8]

Preparation of 6-*Acetyl*-3-*ethoxycyclohex*-2-*enone* **7**.—To a stirred suspension of sodium hydride (60% dispersion in mineral oil; 45.39 g, 1.135 mol) and 15-crown-5 ether (40 drops) in dry 1,2-DME (250 cm^3), under a nitrogen atmosphere, was added a solution of 3-ethoxycyclohex-2-enone **6** (52.95 g, 0.378 mol) in dry 1,2-DME (50 cm^3). The mixture was stirred for 20 min at 0 °C and then ethyl acetate (185 cm^3, 1.896 mol) was added slowly to it over 10 min. The ice-bath was removed and the reaction mixture was heated slowly to reflux over 30 min and maintained at reflux for a further 30 min. After the mixture had cooled, aqueous ammonium chloride (10% w/v; 400 cm^3) was added to it and the organic layer was separated and the aqueous layer extracted with ethyl acetate (3 × 100 cm^3). The combined organic layers were washed with water (2 × 200 cm^3), dried (Na_2SO_4) and evaporated under reduced pressure. The residual oil separated into two layers; the lower layer was distilled to yield a yellow oil, 6-*acetyl*-3-*ethoxycyclohex*-2-*enone* **7** (41.89 g, 61%), b.p. 110–112 °C at 0.07 mmHg (Found: C, 65.8; H, 7.7. $C_{10}H_{14}O_3$ requires C, 65.9; H, 7.7%); ν_{max}(film)/cm^{-1} 1714 (C=O), 1648 and 1603; δ_H($CDCl_3$) 1.35 (2 H, t, *J* 7, CH_2CH_3 keto form), 1.36 (1 H, t, *J* 7, CH_2CH_3 enol form), 2.01 (1 H, s, COMe enol form), 2.26 (2 H, s, COMe keto form), 1.96–2.06 (1 H, m, 5-H), 2.28–2.42 (2 H, m, 4-H, 5-H), 2.50–2.68 (1 H, m, 4-H), 3.39 (0.67 H, t, *J* 6, 6-H keto form), 3.90 (1.33 H, q, *J* 7, CH_2CH_3 keto form), 3.93 (0.67 H, q, *J* 7, CH_2CH_3 enol form), 5.27 (0.33 H, s, 2-H enol form), 5.35 (0.67 H, s, 2-H keto form) and 15.97 (0.33 H, s, OH enol form); δ_C($CDCl_3$) 13.82 (CH_3), 13.85 (CH_3), 19.53 (CH_3), 21.68 (CH_2), 22.54 (CH_2), 26.92 (CH_2), 28.32 (CH_2), 29.61 (CH_3), 58.84 (CH), 64.14 (CH_2), 64.28 (CH_2), 99.47 (CH), 101.45 (CH), 101.86 (quat.), 174.42 (quat.), 175.62 (quat.), 177.97 (quat.), 190.18 (quat.), 194.68 (quat.) and 205.12 (quat.); *m*/*z* 182 (M$^+$, 68%), 139 (100), 122 (34), 112 (52), 111 (61), 105 (62) and 84 (73).

1-(4′-*Ethoxy*-2′-*oxocyclohex*-3′-*enyl*)-3-*phenylpropane*-1,3-*dione* **9**.—To a stirred suspension of sodium amide (12.64 g, 0.324 mol) in liquid ammonia (500 cm^3) was added the diketone **7** (29.50 g, 0.162 mol) in dry ether (50 cm^3). After the mixture had been stirred for 1 h, methyl benzoate (11.02 g, 0.081 mol) in dry ether (20 cm^3) was added dropwise to it over 7 min. Further additions were made sequentially as follows: after 1 h, sodium amide (6.32 g, 0.162 mol); after 30 min, methyl benzoate (5.51 g, 0.040 mol); after 30 min, sodium amide (3.16 g, 0.081 mol); after 30 min, methyl benzoate (5.51 g, 0.040 mol). The reaction mixture was then stirred until the ammonia had evaporated. Aqueous acetic acid (10% v/v; 500 cm^3) was added to quench the reaction, after which the acid was neutralised with saturated aqueous sodium hydrogen carbonate, and the aqueous phase extracted with ethyl acetate (200 cm^3) and separated. The aqueous layer was further extracted with ethyl acetate (3 × 100 cm^3) and the combined organic extracts were washed with water (100 cm^3), dried ($MgSO_4$) and evaporated under reduced pressure. The residue was dissolved in hot aqueous ethanol (60% v/v) and the solution left overnight at −8 °C to crystallize. The solid was filtered off and recrystallized from ether to yield yellow crystals, 1-(4′-*ethoxy*-2′-*oxocyclohex*-3′-*enyl*)-3-*phenylpropane*-1,3-*dione* **9** (20.33 g, 44%), m.p. 82–86 °C (Found: C, 70.3; H, 6.3. $C_{17}H_{18}O_4 \cdot \frac{1}{4}H_2O$ requires C, 70.2; H, 6.4%); ν_{max}(Nujol)/cm^{-1} 1652 (C=O) and 1605; λ_{max}(MeOH)/nm 252 (ε/dm^3 mol^{-1} cm^{-1} 20 300) and 319 (17 200); δ_H($CDCl_3$) 1.37 (3 H, t, *J* 7, CH_2CH_3), 2.12–2.23 (1 H, m, 6′-H), 2.36–2.43 (2 H, m, 5′-H, 6′-H), 2.65–2.75 (1 H, m, 5′-H), 3.41 (1 H, t, *J* 6, 1′-H), 3.93 (2 H, q, *J* 7, CH_2CH_3), 5.45 (1 H, s, 3′-H), 6.36 (1 H, s, 2-H), 7.39–7.55 (3 H, m, 3″-H, 4″-H, 5″-H), 7.81–7.90 (1 H, s, 2″-H, 6″-H) and 15.86 (1 H, br s, OH); δ_C($CDCl_3$) 14.05 (CH_3), 23.76 (CH_2), 27.22 (CH_2), 55.47 (CH), 64.47 (CH_2), 96.53 (CH), 102.40 (CH), 126.97 (2 × CH), 128.50 (2 × CH), 132.27 (CH), 134.11 (quat.), 178.19 (quat.), 181.01 (quat.), 194.88 (quat.) and 195.10 (quat.); *m*/*z* 286 (M$^+$, 27%), 167 (22), 156 (100), 147 (25), 140 (59), 139 (22), 127 (29), 112 (58), 105 (64), 84 (40) and 77 (38).

7-*Oxo*-5,6,7,8-*tetrahydroflavone* **10**.—A solution of the triketone **9** (10.00 g, 35.0 × 10^{-3} mol) in glacial acetic acid (150 cm^3) and concentrated sulfuric acid (0.5 cm^3) was refluxed for 30 min. On cooling, the reaction mixture was poured onto ice, neutralized with saturated aqueous sodium hydrogen carbonate, and extracted with ethyl acetate (3 × 200 cm^3). The combined organic extracts were washed with water (2 × 200 cm^3), dried ($MgSO_4$) and evaporated to dryness. Flash chromatography of the residue on silica gel with ethyl acetate–light petroleum (b.p. 40–60 °C) (80:20) to ethanol–ethyl acetate (10:90) as eluent yielded the *title compound* **10** (5.45 g, 65%). Recrystallization of this from ethyl acetate gave light brown needles, m.p. 172 °C (Found: C, 75.1; H, 5.1. $C_{15}H_{12}O_3$ requires C, 75.0; H, 5.0%); ν_{max}(Nujol)/cm^{-1} 1715 (C=O), 1659 (C=O) and 1615; λ_{max}(MeOH)/nm 272 (ε/dm^3 mol^{-1} cm^{-1} 23 200); δ_H($CDCl_3$) 2.65 (2 H, t, *J* 7, 5-H), 2.94 (2 H, t, *J* 7, 6-H), 3.57 (2 H, s, 8-H), 6.88 (1 H, s, 3-H), 7.47–7.55 (3 H, m, 3′-H, 4′-H, 5′-H) and 7.73–7.80 (2 H, m, 2′-H, 6′-H); δ_C($CDCl_3$) 18.46 (CH_2), 37.94 (CH_2), 41.01 (CH_2), 109.97 (CH), 121.85 (quat.), 125.60 (2 × CH), 128.98 (2 × CH), 130.91 (quat.), 131.35 (quat.), 158.29 (quat.), 163.33 (quat.), 177.92 (quat.) and 204.13 (quat.); *m*/*z* 240 (M$^+$, 95%), 212 (58), 211 (100), 184 (15), 105 (13), 102 (16) and 77 (20).

7-(2″-*Cyanoanilino*)-5,6-*dihydroflavone* **11**.—A solution of the flavone **10** (2.00 g, 8.33 × 10^{-3} mol), anthranilonitrile (0.99 g, 8.39 × 10^{-3} mol) and toluene-*p*-sulfonic acid (100 mg, 5.26 × 10^{-4} mol) in toluene (150 cm^3) was refluxed for 18 h with the azeotropic removal of water. On cooling, the reaction mixture was evaporated to dryness. Flash chromatography of the residue on silica gel yielded, with ethyl acetate–light petroleum (b.p. 40–60 °C) (80:20 to 100:0) as the eluent, the flavone **10** (0.62 g, 31%) and with ethanol–ethyl acetate (7.5:92.5 to 12.5:87.5) as the eluent the *title compound* **11** (1.85 g, 65%). A small amount was recrystallized from aqueous ethanol to give yellow needles (the dihydrate), m.p. 152–154 °C (Found: C, 70.3; H, 5.3; N, 7.5. $C_{22}H_{16}N_2O_2 \cdot 2H_2O$ requires C, 70.2; H, 5.4; N, 7.4%); ν_{max}(Nujol)/cm^{-1} 3201 (NH), 2230 (CN) and 1644 (C=O); λ_{max}(MeOH)/nm 274 (ε/dm^3 mol^{-1} cm^{-1} 24 100) and 381 (21 200); δ_H($CDCl_3$) 2.67 (2 H, t, *J* 8, 5-H), 2.92 (2 H, t, *J* 8, 6-H), 5.78 (1 H, s, 8-H), 6.28 (1 H, br s, NH), 6.73 (1 H, s, 3-H), 7.22 (1 H, t, *J* 7, 4″-H), 7.43–7.48 (3 H, m, 3′-H, 4′-H, 5′-H), 7.61 (1 H, t, *J* 7, 5″-H), 7.64–7.69 (2 H, m, 3″-H, 6″-H) and 7.72–7.77 (2 H, m, 2′-H, 6′-H); δ_H([$^2H_6$]-DMSO) 3.37 (4 H, s, 5-H, 6-H), 5.32 (1 H, s, 8-H), 6.75 (1 H, s, 3-H), 7.41 (1 H, t, *J* 8, 4″-H), 7.45–7.50 (3 H, m, 3′-H, 4′-H, 5′-H), 7.59 (1

H, d, *J* 8, 6″-H), 7.77 (1 H, t, *J* 8, 5″-H), 7.84–7.88 (2 H, m, 2′-H, 6′-H), 7.91 (1 H, d, *J* 8, 3″-H) and 9.08 (1 H, s, NH); δ_C([$^2H_6$]-DMSO) 18.46 (CH_2), 28.53 (CH_2), 92.11 (CH), 105.47 (quat.), 110.77 (CH), 112.13 (quat.), 116.50 (quat.), 122.89 (CH), 124.25 (CH), 125.62 (2 × CH), 128.93 (2 × CH), 130.76 (CH), 131.90 (quat.), 133.62 (CH), 133.99 (CH), 142.48 (quat.), 149.68 (quat.), 161.17 (quat.), 162.67 (quat.) and 177.37 (quat.); *m*/*z* 340 (M^+, 92%), 339 (100), 212 (10) and 102 (10).

7-(2″-*Ethoxycarbonylanilino*)-5,6-*dihydroflavone* **13**.—This compound was prepared in the same manner as **11** from the flavone **10** (0.5 g, 2.08 × 10^{-3} mol), ethyl anthranilate (0.69 g, 4.17 × 10^{-3} mol) and toluene-*p*-sulfonic acid (50 mg, 2.63 × 10^{-4} mol) in toluene (70 cm^3). Flash chromatography of the residue on silica gel yielded, with ethyl acetate–light petroleum (b.p. 40–60 °C) (90:10 to 100:0) as the eluent, flavone **10** (0.17 g, 34%) and with ethanol–ethyl acetate (5:95 to 7.5:92.5) as the eluent the *title compound* **13** (0.38 g, 47%). A small amount was recrystallized from toluene to give a yellow solid, m.p. 156–158 °C (Found: C, 74.3; H, 5.5; N, 3.8. $C_{24}H_{21}NO_4$ requires C, 74.5; H, 5.5; N, 3.6%); ν_{max}(Nujol)/cm^{-1} 3440 (NH), 1689 (C=O), 1660 (C=O) and 1622; λ_{max}(MeOH)/nm 277 (ε/dm^3 mol^{-1} cm^{-1} 20 500) and 393 (29 300); δ_H($CDCl_3$) 1.43 (3 H, t, *J* 7, CH_2CH_3), 2.64 (2 H, t, *J* 8, 5-H), 2.89 (2 H, t, *J* 8, 6-H), 4.39 (2 H, q, *J* 7, CH_2CH_3), 6.04 (1 H, s, 8-H), 6.71 (1 H, s, 3-H), 7.05 (1 H, t, *J* 8, 4″-H), 7.43–7.48 (3 H, m, 3′-H, 4′-H, 5′-H), 7.56 (1 H, t, *J* 8, 5″-H), 7.64 (1 H, d, *J* 8, 6″-H), 7.72–7.80 (2 H, m, 2′-H, 6′-H), 8.07 (1 H, d, *J* 8, 3″-H) and 9.53 (1 H, s, NH); δ_C([$^2H_6$]-DMSO) 14.31 (CH_3), 18.49 (CH_2), 29.46 (CH_2), 61.45 (CH_2), 91.91 (CH), 110.75 (CH), 111.89 (quat.), 116.92 (quat.), 120.02 (CH), 121.35 (CH), 125.61 (2 × CH), 128.90 (2 × CH), 130.62 (CH), 131.85 (CH), 132.12 (quat.), 133.95 (CH), 143.32 (quat.), 149.11 (quat.), 160.94 (quat), 163.23 (quat.), 167.94 (quat.) and 177.27 (quat.); *m*/*z* 387 (M^+, 100%), 358 (68), 340 (47), 312 (25), 238 (26), 223 (25), 222 (26) and 156 (43).

12-*Amino*-2-*phenyl*-5,6-*dihydropyrano*[2,3-a]*acridin*-4-*one* **12**.—To a stirred suspension of sodium amide (150 mg, 3.84 × 10^{-3} mol) in liquid ammonia (50 cm^3) at −78 °C was added dropwise the flavone **11** (620 mg, 1.82 × 10^{-3} mol) in dry THF (10 cm^3) over 3 min. An immediate colour change from yellow to red was observed. The reaction mixture was allowed to warm to room temperature over 4 h until all the ammonia had evaporated. Dry 1,2-DME (50 cm^3) was added to the mixture which was then refluxed for 17 h; at this point TLC indicated that no starting material was present. On cooling, the reaction was treated with aqueous ammonium chloride (10% w/v; 50 cm^3) and the organic layer was separated; the aqueous layer was then extracted with ethyl acetate (4 × 100 cm^3). The combined extracts were washed with water (100 cm^3), dried ($MgSO_4$) and evaporated under reduced pressure. Flash chromatography of the residue on silica gel with ethanol–ethyl acetate (10:90 to 20:80) as eluent yielded the *title compound* **12** (180 mg, 29%). Brown needles crystallized from a NMR sample in [$^2H_6$]-DMSO, m.p. 299–302 °C [Found: C, 68.0; H, 5.5; N, 6.6. $C_{22}H_{16}N_2O_2$·$(CD_3)_2SO$ requires C, 67.9; H, 5.2; N, 6.6%]; ν_{max}(Nujol)/cm^{-1} 3485 (NH), 1641, 1611 and 1568; δ_H($CDCl_3$) 2.94 (2 H, t, *J* 7, 5-H), 3.14 (2 H, t, *J* 7, 6-H), 6.18 (2 H, br s, exchanges with D_2O, NH_2), 6.82 (1 H, s, 3-H), 7.49 (1 H, t, *J* 7.5, 9 H or 10 H), 7.54–7.60 (3 H, m, 3′-H, 4′-H, 5′-H), 7.71 (1 H, t, *J* 7.5, 9-H or 10-H), 7.81–7.88 (3 H, m, 2′-H, 6′-H, 8-H or 11-H) and 7.95 (1 H, d, *J* 7.5, 8-H or 11-H); δ_H([$^2H_6$]-DMSO) 2.71 (2 H, t, *J* 7, 5-H), 2.93 (2 H, t, *J* 7, 6-H), 7.04 (1 H, s, 3-H), 7.47 (1 H, t, *J* 7, 9-H or 10-H), 7.50 (2 H, s, exchanges with D_2O, NH_2), 7.56–7.62 (3 H, m, 3′-H, 4′-H, 5′-H), 7.69 (1 H, t, *J* 7, 9-H or 10-H), 7.76 (1 H, d, *J* 7, 7-H or 11-H), 8.02–8.08 (2 H, m, 2′-H, 6′-H) and 8.34 (1 H, d, *J* 7, 7-H or 11-H); δ_C([$^2H_6$]-DMSO) 18.06 (CH_2), 32.13 (CH_2), 101.22 (quat.), 110.12 (quat.), 118.56 (quat.), 119.57 (CH), 122.91 (CH), 124.65 (CH), 125.73 (2 × CH), 128.54 (CH), 129.25 (2 × CH), 130.74 (CH), 131.31 (CH), 131.87 (quat.), 147.66 (quat.), 148.58 (quat.), 159.97 (quat.), 160.12 (quat.), 161.39 (quat.) and 176.80 (quat.); *m*/*z* 340 (M^+, 83%), 339 (100), 329 (43), 237 (7), 181 (8) and 156 (10).

12-*Hydroxy*-2-*phenyl*-5,6-*dihydropyrano*[2,3-a]*acridin*-4-*one* **14**.—To a stirred suspension of sodium amide (280 mg, 7.17 × 10^{-3} mol) in dry 1,2-DME (50 cm^3) at room temperature was added dropwise flavone **13** (920 mg, 2.38 × 10^{-3} mol) in dry 1,2-DME (50 cm^3). The reaction mixture was then refluxed for 2.5 h. On cooling, the mixture was treated with aqueous ammonium chloride (10% w/v; 100 cm^3), the organic layer separated and the aqueous layer extracted with ethanol–chloroform (1:1; 5 × 100 cm^3). The combined extracts were washed with water (2 × 100 cm^3), dried ($MgSO_4$), and evaporated under reduced pressure. Flash chromatography of the residue on silica gel with ethanol–ethyl acetate (10:90 to 30:70) as eluent yielded the *title compound* **14** (521 mg, 64%). Recrystallization of this from aqueous ethanol (75% v/v) gave a yellow solid (the monohydrate), m.p. > 360 °C (Found: C, 73.5; H, 4.6; N, 4.1. $C_{22}H_{15}NO_3$·$1H_2O$ requires C, 73.5; H, 4.8; N, 3.9%); ν_{max}(Nujol)/cm^{-1} 3210 (OH) and 1616; λ_{max}(MeOH)/nm 231 (ε/dm^3 mol^{-1} cm^{-1} 20 150), 267 (23 800) and 356 (14 400); δ_H([$^2H_6$]-DMSO) 2.76 (2 H, t, *J* 8, 5-H), 3.03 (2 H, t, *J* 8, 6-H), 7.05 (1 H, s, 3-H), 7.45 (1 H, t, *J* 7.5, 9-H or 10-H), 7.56–7.62 (3 H, m, 3′-H, 4′-H, 5′-H), 7.68 (1 H, d, *J* 7.5, 8-H or 11-H), 7.75 (1 H, t, *J* 7.5, 9-H or 10-H), 8.29 (1 H, d, *J* 7.5, 8-H or 11-H), 8.31–8.35 (2 H, m, 2′-H, 6′-H) and 12.63 (1 H, br s, OH); δ_C([$^2H_6$]-DMSO) 17.07 (CH_2), 26.41 (CH_2), 107.01 (quat.), 109.24 (CH), 115.71 (quat.), 118.46 (CH), 124.61 (CH), 125.87 (CH), 126.01 (2 × CH), 126.51 (quat.), 129.12 (2 × CH), 131.24 (CH), 131.51 (quat.), 132.38 (CH), 138.46 (quat.), 155.34 (quat.), 159.22 (quat.), 160.85 (quat.), 172.07 (quat.) and 176.60 (quat.); *m*/*z* 341 (M^+, 62%), 340 (100) and 157 (11).

Compound **14** was also prepared in 59% overall yield (compared with 30% for two steps) from the flavone **10** (1 g, 4.167 × 10^{-3} mol) and ethyl anthranilate (2 equiv.) without isolating the enamine intermediate. In this case the solvent was evaporated from the enamine after refluxing in toluene (150 cm^3) for 4 h with toluene-*p*-sulfonic acid (100 mg); the crude enamine was dried *in vacuo*, dissolved in dry 1,2-DME (50 cm^3) and added to $NaNH_2$ (3 equiv.) in dry 1,2-DME (50 cm^3) with a catalytic amount of 15-crown-5 ether (10 drops). After refluxing for 1 h the reaction mixture was worked-up as above.

12-*Amino*-2-*phenylpyrano*[2,3-a]*acridin*-4-*one* **4**.—A stirred mixture of the acridinone **12** (50 mg, 1.469 × 10^{-4} mol) and manganese(IV) oxide (1.05 g; activated, brown) was refluxed in toluene (100 cm^3) for 2 h. The reaction mixture was filtered whilst hot and the residual solid washed with hot ethanol. The solvents were evaporated under reduced pressure and the resulting solid recrystallized from toluene–light petroleum (b.p. 40–60 °C) to give the *title compound* **4** as a yellow solid (36 mg, 72%), m.p. 310–312 °C (Found: M^+, 338.1055. $C_{22}H_{14}N_2O_2$ requires *M*, 338.1055); ν_{max}(Nujol)/cm^{-1} 3512, 3335 (NH_2), 1634 and 1596; λ_{max}(MeOH)/nm 260 (ε/dm^3 mol^{-1} cm^{-1} 44 700) and 377 (10 900); δ_H([$^2H_6$]-DMSO) 7.15 (1 H, s, 3-H), 7.52 (1 H, t, *J* 8, 9-H or 10-H), 7.66–7.70 (3 H, m, 3′-H, 4′-H, 5′-H), 7.73 (1 H, d, *J* 8.5, 5-H or 6-H), 7.82 (1 H, t, *J* 8, 9-H or 10-H), 7.92 (1 H, d, *J* 8, 7-H or 11-H), 8.10 (1 H, d, *J* 8.5, 5-H or 6-H), 8.12–8.16 (2 H, m, 2′-H, 6′-H), 8.39 (2 H, br s, NH_2) and 8.53 (1 H, d, *J* 8, 7-H or 11-H); *m*/*z* 339 (27%), 338 (M^+, 100), 236 (60), 180 (21), 155 (19), 153 (15) and 105 (13).

12-*Hydroxy*-2-*phenylpyrano*[2,3-a]*acridin*-4-*one* **5**.—A stirred mixture of the acridinone **14** (500 mg, 1.46 × 10^{-3} mol) and mercury(II) acetate (510 mg, 1.60 × 10^{-3} mol) was refluxed in DMSO (10 cm^3) for 24 h. The DMSO was removed under reduced pressure and flash chromatography with ethyl acetate–ethanol (100:0 to 0:100) yielded the *title compound* **5** (254 mg, 51%), m.p. > 360 °C (Found: M^+, 339.0895. $C_{22}H_{13}NO_3$ requires *M*, 339.0895); ν_{max}(Nujol)/cm^{-1} 1626; λ_{max}(MeOH)/nm 240 (ε/dm^3 mol^{-1} cm^{-1} 14 400), 269 (15 000), 345 (6000) and 388 (4600); δ_H([$^2H_6$]-DMSO) 7.27 (1 H, s, 3-H), 7.42 (1 H, t, *J* 7.5, 9-H or 10-H), 7.58 (1 H, d, *J* 8.5, 5-H or 6-H), 7.65 (1 H, d, *J* 7.5, 8-H or 11-H), 7.65–7.72 (3 H, m, 3′-H, 4′-H, 5′-H), 7.83 (1 H, t, *J* 7.5, 9-H or 10-H), 8.28 (1 H, d, *J* 8.5, 5-H or 6-H), 8.38 (1 H, d, *J* 7.5, 8-H or 11-H), 8.57 (2 H, d, *J* 7.5, 2′-H, 6′-H) and 12.31 (1 H, s, OH); *m*/*z* 340 (25%), 339 (M^+, 100), 311 (40), 156 (22), 153 (27) and 105 (14).

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