Synthesis of 5,7-Dihydrodibenzo[*b*,*f*][1,7]naphthyridine-6,12-dione, an Unexpected Isolate from *Isatis tintoria*

by **Herbert M. Riepl*** and **Melanie Kellermann**

Institute of Resource and Energy Technology, Technical University of Munich, Petersgasse 18, DE-94315 Straubing
(phone: +49-9421-187120; fax: +49-9421-187111; e-mail: herbert.riepl@wzw.tum.de)

The dye plant *Isatis tinctoria* yields a number of heterocyclic compounds with interesting anti-inflammatory and cytotoxic properties, formed mainly in an unknown manner by post-harvest treatment. A synthesis of the incidently isolated 5,7-dihydrodibenzo[*b*,*f*][1,7]naphthyridine-6,12-dione (**4a**) is presented. Starting from different 1,2-diarylhydrazines, adducts **11** with acetylenedicarboxylates (= but-2-ynedioates) are thermally treated (*Scheme*). In a *Fischer*-type rearrangement, 3-(arylamino)quinolinecarboxylic acids **9** are obtained, which can be cyclized under *Friedel–Crafts* conditions to yield a number of analoga **4** of the title compound.

1. Introduction. – *Isatis tinctoria* (Brassicaceae), a plant native to the temperate climate of Europe and Asia, is one of the sources of the indigo dye since its discovery more than four or five thousand years ago. The dye is formed from some indole glycosides present in the cells [1][2]. Some other plants like *Indigofera* species (Leguminosae), *Wrightia tinctoria* (Apocynaceae), *Polygonum tinctorium* (Polygonaceae), or *Baphicacanthes cusia* (Acanthaceae), all native to South East Asia, also produce these precursors. Preparations from the plant as well as the dye itself have been and are used as a phytopharmaceutical. *Quing Dai* [3], a preparation of the Chinese Traditional Medicine from various indigo-yielding plants, is mentioned as a cure against malaria, various inflammations, namely gingival, jaundice, and 'cancer'. With respect of the last topic, one of the dyes present, indirubin (**1**; *Fig. 1*) and much more interesting, the nonnaturally occurring indirubinsulfonic acid, has been found as an effective inhibitor of cyclin-dependent kinases [4] and thus establishes the tradition to be right. In Europe, there are hints on a use against 'allergy' in the folk medicine. As

1 **2**

Fig. 1. *Major compounds of pharmaceutical value from* Isatis tinctoria *besides indigo: indirubin* (**1**) *and tryptanthrin* (**2**)

© 2009 Verlag Helvetica Chimica Acta AG, Zürich

early as 1951, *De Landsheere* observed a decreasing effect on the blood-cell count of eosinophiles of experimental animals after administration of indirubin (**1**) [5].

Besides these dyes, various other heterocycles have been isolated, *e.g.*, tryptanthrin (**2**), with interesting anti-inflammatory properties, and some more complex derivatives [6][7].

2. Results and Discussion. – Considering the interesting medical properties of *I. tinctoria*, the properties and synthesis of a new heterocycle will be reported here. During an attempt to obtain quingdaone from an European *Isatis* spp. – a molecule first isolated by *Li* [8] from Chinese *I. tinctoria* – we have collected a fraction from which a tiny amount of a highly insoluble material separated. Having the same relative molecular mass as indigo or indirubin, *i.e.*, corresponding to the formula $C_{16}H_{10}O_2N_2$, but having different mass spectra and being a dull yellow substance with λ_{max} (DMF; log ε) 389 (0.43), 369 (0.41), 352 (0.23), 333 (0.39), and 319 (0.25) nm, this unknown compound is different from both dyes or even from isoindigo.

First, the substance was suspected to be epindolidione[1]) (**3**; *Fig. 2*), known from the industrial production of indigo as a minor component [9], or an isomeric ketenaminal [10]. But the ^{1}H- and ^{13}C-NMR spectra (*Table*) revealed a structure with eight recordable H-atoms, thus rendering impossible all symmetric structures. The complicated C,H-COSY plot was consistent with a highly conjugated aromatic ring system, featuring, *e.g.*, three ^{13}C-NMR signals connected with one ^{1}H-NMR signal at δ 7.41, representing three H-atoms of two different rings (*Table*, *cf. Footnotes b* and *c*). Full HBMC assignment of the NMR data was possible only for a corresponding quaternary *N*-methyl salt, reported elsewhere.

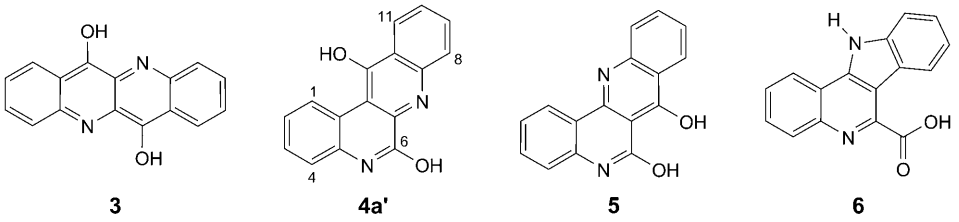


Fig. 2. *Possible dibenzonapthyridindione isomers from natural and synthetic sources: Epindolidione* (**3**), *a minor impurity in the technical indigo synthesis, and dibenzo[*b,h*][1,6]naphthyridine-6,7-diol* (**5**), *and 11*H*-indolo[3,2-*c*]quinoline-6-carboxylic acid* (**6**) *from* Tabernanthe iboga

The UV/VIS spectra of the unknown structure gave a hint to a six-membered-ring system rather than to an indoxyl- or an indolone-like indigoid system, thus a structure such as **4a**, **4a′**, **5**, or **6** was proposed for this natural product (*Figs. 2* and *3*); however, tautomeric forms could not be established up to now.

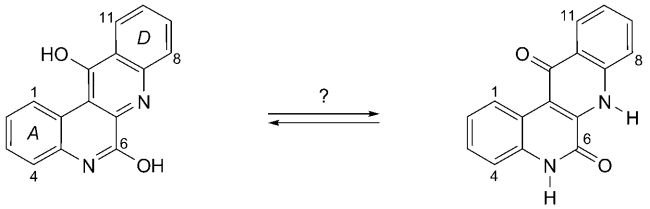
Unfortunately, the substance **4a** or **4a′** was found only once by us in *I. tinctoria* plant material. The lack of crystals suitable for X-ray analysis made necessary a synthetic effort to reveal the correct structure of this substance by comparison. The possibility of

[1]) λ_{max} (DMF; log ε) 444 (0.22), 419 (0.185), 398 (0.033), 334 (0.072), and 274 (1.13) nm.

Table. *^{1}H- and ^{13}C-NMR Data* (H,H- and C,H-COSY) *of 5,7-Dihydrodibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4a**/**4a′**; see *Fig. 3*). In (D_6)DMSO; δ in ppm.

^{13}C-NMR		^{1}H-NMR	
		H–C(1) to H–C(4) (ring *A*)	H–C(8) to H–C(11) (ring *D*)
177.86	(C(12)=O)		
157.28	(C(6)=O)		
138.85	(*s*[a]))		
134.63		7.76	
133.09	(*s*[a]))		
127.72		7.41[b])[c])	
126.18	(*s*[a]))		
126.16	(*s*[a]))		
125.78			9.73
125.16		8.33	
123.92			7.41[b])[c])
123.27			7.35
119.48		8.11	
118.04	(*s*[a]))		
115.65			7.41[b])[c])
114.96	(*s*[a]))		

[a]) Quaternary C-atoms. [b]) Tentative assignment to either ring *A* or *D*. [c]) Signal belonging to both rings *A* and *D* (not resolved in (D_6)DMSO.



4a' **4a**

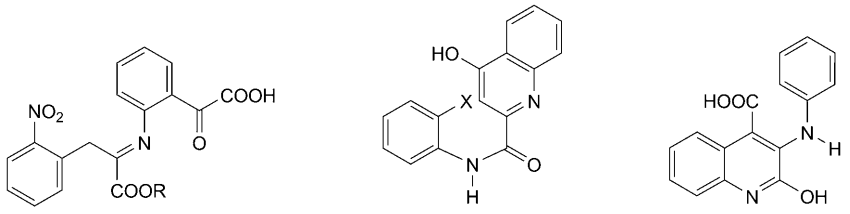
Fig. 3. *Dibenzo[*b,f*][1,7]naphthyridine-6,12-diol* (**4a**) *and 5,7-Dihydrodibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4a′**) *as possible tautomers*

a whole set of isomeric structures for which very few data were available rendered insecure a purely NMR-based approach. However, some isomeric systems could be ruled out based on a HR-MS evidence, such as **5** [11] or **6**. Moreover, the ethyl ester of **6** was synthesized as published [12]; the free acid **6** easily lost CO_2 in the mass spectrometer under various ionization conditions.

The alternative structure proposal **4a** or **4a′** proved to be the correct ones. This class of heterocycles is not very common in the chemical literature. Some related 6-phenyldibenzonaphthyridine derivatives [13] are reported, as well as various chloro derivatives or a 6-deoxy variant [14].

Retrosynthetic analysis suggests as starting material for the synthesis of **4a** a condensation product **7** from isatin (=1*H*-indole-2,3-dione) with 2-nitrophenylpyruvic

acid (= 3-(2-nitrophenyl)-2-oxopropanoic acid) (*Fig. 4*), which should be subjected to reductive amidation of the nitro group. However, different attempts to synthesize **7** did not yield any isolable product. Even by means of Pd-catalyzed C–C coupling of the *N*-(2-bromophenyl)-substituted amide **8** of kynureninic acid (=4-hydroxyquinoline-2-carboxylic acid), no trace of **4a** was obtained.



7 **8** **9**

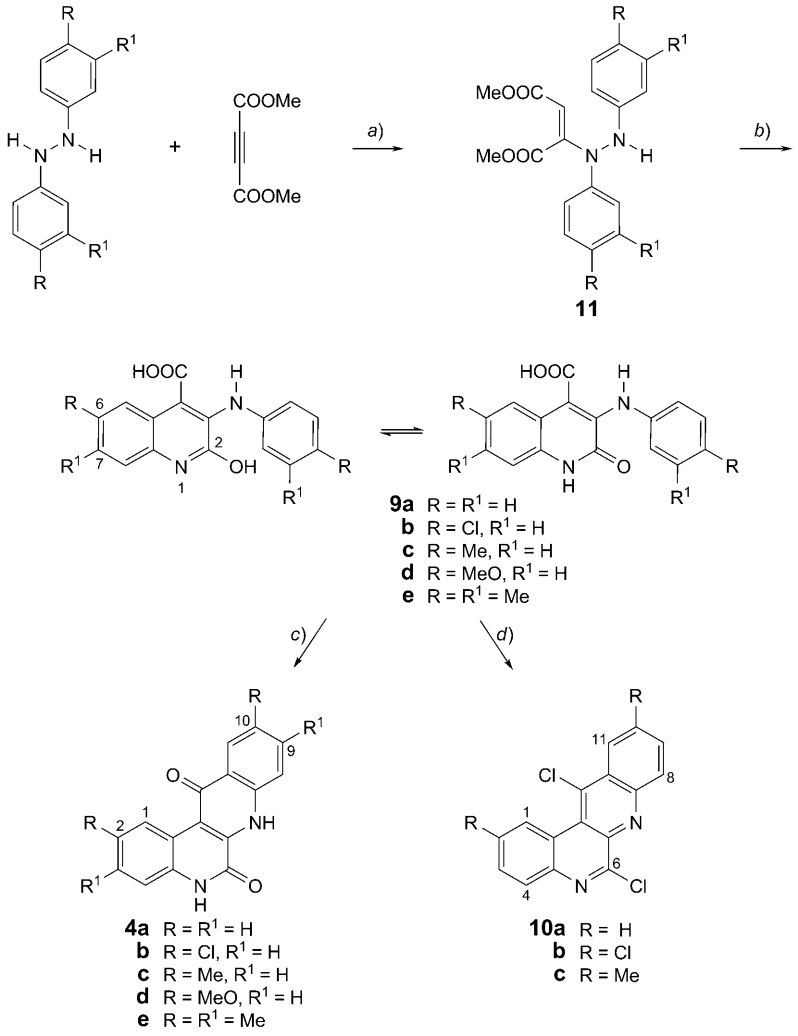
Fig. 4. *Potential starting materials* **7**–**9** *from retrosynthetic designs for substance* **4**. The synthesis of **7** seems not to be feasible; **8** was obtained in 90% yield but did not react further; derivatives of **9** could be cyclized (see *Scheme*).

Finally, it was shown that under certain circumstances, 3-(arylamino)-2-hydroxyquinoline-4-carboxylic acids **9a**–**9e** could be cyclized in a *Friedel–Crafts*-like manner to give **4a**–**4e** (*Scheme*). Provided the reaction was performed in suspension, yields were good, otherwise decarboxylation occurred quickly; this was ascertained by use of PCl_3 as a medium which has the additional advantage of a low boiling point. The use of $POCl_3$ also promoted cyclization but yielded the chloro derivatives **10a**–**10c** (*Scheme*), as expected. Access to the 3-(arylamino)-2-hydroxyquinoline-4-carboxylic acids **9a**–**9d** was easy; they are the products of a *Fischer*-type rearrangement of diesters **11**, which are adducts of symmetric 1,2-diarylhydrazines with acetylenedicarboxylic acid dimethyl ester (=dimethyl but-2-ynedioate) [15]. With nonsymmetric 1,2-diarylhydrazines, this reaction gave mixtures whose purification was difficult. Generally, even for the reported case, the success of this reaction depended very much on the quality of the 1,2-diarylhydrazines. In our hands, all commercially available diarylhydrazines failed to give the products **11** completely. The addition worked best with 1,2-diarylhydrazines obtained by reduction from the corresponding azobenzenes with Zn powder in AcOH under moderate conditions in the absence of air [16].

For the time being, the synthesis of additional analogues **4** according to the protocol described above is confined since further 3-(arylamino)-2-hydroxyquinoline-4-carboxylic acids **9** could not be synthesized by use of this reaction.

For instance, 1,2-(4-bromophenyl)hydrazine did not give useful products. A small yield of 3-[(3,4-dimethylphenyl)amino]-1,2-dihydro-6,7-dimethyl-2-oxoquinoline-4-carboxylic acid (**9e**) was obtained from 1,2-bis(3,4-dimethylphenyl)hydrazine with dimethyl acetylenedicarboxylate; its low yield was due to a 18% admixture of the regioisomeric 3-[(3,4-dimethylphenyl)amino]-1,2-dihydro-5,6-dimethyl-2-oxoquinoline-4-carboxylic acid, which had to be removed by fractional crystallization. The major crystallizate was pure and characterized by its ^{1}H-NMR spectrum to be **9e** (four clear *s* for the Me groups) Acid **9e** was cyclized according to the general protocol to

Scheme. *Successful Protocol for the Synthesis of Compounds of Type* **4**



a) Addition in MeOH, reflux. *b*) 1. *Fischer*-type rearrangement in pyridine or picoline (=2-methylpyridine), reflux; 2. 15% aq. NaOH soln. *c*) *Friedel–Crafts* cyclization with $AlCl_3/PCl_3$ or *d*) $POCl_3$/reflux.

yield two regioisomers **4e** and **4e′** of a tetramethyl derivative in a nearly 1:1 ratio. In case of shorter reaction times, the isomer ratio **4e**/**4e′** was 3:1 or better whereby the isomers could be distinguished by their ^{1}H-NMR signals in (D_6)DMSO (minor isomer **4e′** with two *d*/*s* systems for the aromatic H-atoms clearly separated from **4e** with four *s* where one *s* coincides with a *s* of **4e′**). The regioisomers **4e** and **4e′** could not be separated sufficiently by crystallization.

The above described synthesis supplied us with a useful number of analogues **4** for an ongoing clarification of their possible cytotoxic activities and of the speculations about the bioformation of **4a**, an isolate which must be considered as an interesting artefact until another isolation is successfully achieved. Indigo is a product of the hydrolysis at basic pH in the dyeing broth and enzyme-free oxidative dimerization of indoxyl (=1*H*-indol-3-ol, mainly from isatan B or indican which is the β-D-glucoside of 1*H*-indol-3-ol). It occurs in very small concentrations in the fresh, intact plant. The formation of indirubin (**1**) or other substances may be as well a nonenzymatic condensation of indoxyl with isatin (=1*H*-indole-2,3-dione), which is present in *Isatis* species [17]. The marked influence of post-harvest treatment on the synthesis of tryptanthrin (**2**) in the extracts of *Isatis* – it is formally condensed in this manner from isatine and anthranilic acid (=2-aminobenzoic acid) under less known circumstances – was already a matter of concern [18].

Excellent support from the instrumental analytics by *G. Krutsch*, *R. Dumitrescu*, and *U. Ammari* is given special attention here. We are indebted for financial support of the project by *G. Klinge*, the *Klinge Pharma Holding GmbH & Co. KG.*

Experimental Part

General. UV/VIS Spectra: *Analytik Jena-Specord 210*; λ_{max} (log ε) in nm. IR Spectra: *Perkin-Elmer 481*; in KBr pellets, $\tilde{\nu}$ in cm^{-1}. NMR Spectra: *Bruker AMX-400*; δ in ppm, *J* in Hz. MS: *Finnigan MAT 70*, EI at 70 eV, CI in isobutane; in *m*/*z* (rel. %). Elemental analyses were obtained from the Mikroanalytisches Labor, Institut für Anorganische Chemie, Technische Universität München.

Synthesis of **11** *and* **9**. Dimethyl acetylenedicarboxylate (=dimethyl but-2-ynedioate) was purchased from *Sigma Aldrich*. A limited number of 3-(arylamino)-1,2-dihydro-2-oxoquinoline-4-carboxylic acid methyl esters could be prepared according to a known procedure [15] with freshly prepared 1,2-diarylhydrazines. Shortly, the hydrazine and dimethyl (or diethyl) acetylenedicarboxylate were used in equimolar amounts and boiled in MeOH. After crystallization, the adduct **11** was heated in a base, pyridine or picoline. The 3-(arylamino)-1,2-dihydro-2-oxoquinoline-4-carboxylic acid **9** was obtained by alkaline treatment of the respective methyl ester (*ca.* 2 g) by refluxing in 15% aq. NaOH soln (50 ml) mixed with MeOH (3–4 ml) for 45 min. Insoluble materials were removed by filtration, and the filtrate was then acidified with dil. aq. HCl soln. The solid acid was isolated by suction and dried in the air.

Friedel–Crafts *Cyclization* **9** → **4**: *General Procedure.* Exemplified by 6-chloro-3-[(4-chlorophenyl)-amino]-1,2-dihydro-2-oxoquinoline-4-carboxylic acid (**9b**): A suspension of dry **9b** (0.340 g, 0.001 mol) in PCl_3 (10 ml) was stirred for 30 min. Then PCl_5 (0.2 g) was added, and the mixture was heated to reflux. After 1 h, coarse-grained anh. $AlCl_3$ (0.36 g; *Fluka*) was added while continuing the reflux for additional 4 to 5 h. The fine suspension turned redish, and an oil separated which was scratched from the walls from time to time to ensure complete mixture with the rest of the soln. Finally, the solvent mixture was decanted from the red oil, and ice (20 g) was added. By continued stirring, the oil turned into a yellow powder: 0.276 g (83%) of **4b**. Recrystallization was achieved from boiling DMF (or dimethylacetamide or DMSO).

1,2-Dihydro-2-oxo-3-(phenylamino)quinoline-4-carboxylic Acid (**9a**). Yield 59%. Yellow needles (MeOH). M.p. 253°. IR: 3379*m* (br.), 2985*w*, 2957*w*, 2887*w*, 2853*w*, 1689*m*, 1655v*s*, 1560*s*, 1519*s*, 1427*m*, 1375*m*, 1313*w*, 905*w*, 755*m*, 704*m*. ^{1}H-NMR ((D$_6$)DMSO): 12.25 (*s*, 1 H); 7.89 (*s*, 1 H); 7.63 (*d*, *J* = 7.82, 1 H); 7.37–7.32 (*m*, 2 H); 7.19–7.15 (*m*, 3 H); 6.96 (*d*, *J* = 7.82, 2 H); 6.87 (*t*, *J* = 7.82, 1 H). ^{13}C-NMR ((D$_6$)DMSO): 167.0 (COOH); 158.8 (CONH); 142.7; 133.7; 130.7; 128.5; 127.1; 123.6; 122.5; 121.2; 120.7; 118.6; 117.4; 115.3. HR-CI-MS: 282 (18), 281 (100), 280 (38, $[M+1]^+$), 263 (7), 237 (17), 236 (10). Anal. calc. for $C_{16}H_{12}N_2O_3$ (280.29): C 68.57, H 4.32, N 9.99; found: C 68.43, H 4.17, N 9.95.

*5,7-Dihydrodibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4a**). Yield 89%. Orange powder (dimethylacetamide). M.p. >400°. UV/VIS (DMF): 414 (0.05), 389 (0.43), 369 (0.41), 352 (0.23), 333 (0.39), 319

(0.25). IR: 3034*s* (br.), 1626*m*, 1603v*s*, 1542v*s*, 1499*w*, 1482*m*, 1442*w*, 1421*m*, 1396*m*, 1324*w*, 1295*m*, 1151*w*, 1117*w*, 1043*w*, 1020*w*, 956*w*, 888*w*, 854*w*, 777*w*, 753*s*, 721*w*, 701*w*, 677*w*, 628*w*, 599*m*, 572*w*, 538*w*, 515*w*, 470*w*. ^{1}H-NMR ($CDCl_3$): 12.53 (*s*, 1 H); 12.24 (*s*, 1 H); 9.73 (*d*, *J* = 7.8, 1 H); 8.30 (*d*, *J* = 7.8, 1 H); 8.12 (*d*, *J* = 8.8, 1 H); 7.76 (*t*, *J* = 7.8, 1 H); 7.46–7.39 (*m*, *J* = 7.1, 3 H); 7.31 (*t*, *J* = 7.3, 1 H). ^{13}C-NMR ((D_6)DMSO): 177.9; 157.3; 138.9; 134.6; 133.1; 127.7; 126.2; 126.2; 125.8; 125.2; 123.9; 123.3; 119.5; 118.0; 115.7; 115.0. HR-CI-MS: 263 (52), 262 (100), 244 (3). Anal. calc. for $C_{16}H_{10}N_2O_2$ (262.27): C 73.27, H 3.84, N 10.68; found: C 73.8, H 3.76, N 10.64.

*6,12-Dichlorodibenzo[*b,f*][1,7]naphthyridine* (**10a**). Yield 90%. Tan solid (dimethylacetamide). M.p. >350°. IR: 1674*w*, 1614*w*, 1581*w*, 1541*w*, 1479*m*, 1459*m*, 1398*m*, 1332*w*, 1291*s*, 1254*w*, 1199*w*, 1136*w*, 1054*m*, 1032*m*, 1011*w*, 903*w*, 860*w*, 822*s*, 752v*s*, 650w. ^{1}H-NMR ((D_6)DMSO): 9.77 (*d*, *J* = 7.83, 1 H); 8.69 (*d*, *J* = 8.8, 1 H); 8.41 (*d*, *J* = 7.83, 1 H); 8.12–8.10 (*m*, 2 H); 8.04 (*t*, *J* = 7.82, 1 H); 7.94–7.88 (*m*, 2 H). HR-CI-MS: 302 (17), 301 (47), 300 (74), 299 (73), 298 (100), 263 (19), 228 (18).

6-Chloro-3-[(4-chlorophenyl)amino]-1,2-dihydro-2-oxoquinoline-4-carboxylic Acid (**9b**). Yield 82%. Bright yellow needles (MeOH). M.p. 315°. IR: 3024*s* (br.) 1663v*s*, 1637v*s*, 1538*s*, 1544v*s*, 1490*s*, 1421*s*, 1383*s*, 1321*s*, 1286*s*, 1235v*s*, 1086*m*, 1009*w*, 903*s*, 886*s*, 847*m*, 820*s*, 807*m*, 777*w*, 746*w*, 729*s*, 720*w*, 697*m*. ^{1}H-NMR ((D_6)DMSO): 12.38 (*s*, 1 H); 8.28 (*s*, 1 H); 7.63 (*s*, 1 H); 7.36 (*d*, *J* = 9.8, 1 H); 7.30 (*d*, *J* = 9.8, 1 H); 7.22 (*d*, *J* = 7.3, 2 H); 6.98 (*d*, *J* = 7.3, 2 H). ^{13}C-NMR ((D_6)DMSO): 167.13 (CO); 158.68 (CONH); 141.3; 132.8; 132.3; 128.7 (C_p); 127.0; 126.8; 125.9; 122.6; 121.5 (C(6)); 119.2; 117.5; 116.9. HR-CI-MS (methyl ester): 364 (67), 362 (100), 332 (17), 330 (25), 304 (31), 302 (49), 295 (23), 267 (24), 239 (19), 170 (34). Anal. calc. for $C_{16}H_{10}Cl_2N_2O_3$ (349.18): C 55.04, H 2.89, Cl 20.31, N 8.02; found: C 55.11, H 2.95, Cl 20.46, N 8.01.

*2,10-Dichloro-5,7-dihydrodibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4b**). Yield 83%. Dull yellow powder (dimethylacetamide). M.p. >400°. IR: 3162*m*, 3032*m*, 1670v*s*, 1594v*s*, 1560v*s*, 1534v*s*, 1481*m*, 1457*m*, 1417*m*, 1391*w*, 1378*w*, 1338*w*, 1276*w*, 1231*w*, 1177*w*, 1143*w*, 1078*w*, 899*w*, 862*w*, 831*m*, 799*w*, 766*w*, 727*s*, 588*m*, 571*m*. ^{1}H-NMR ((D_6)DMSO): 12.57 (*s*, 1 H); 12.43 (*s*, 1 H); 9.63 (*s*, 1 H); 8.12 (*s*, 1 H); 8.08 (*d*, *J* = 8.8,1 H); 7.74 (*d*, *J* = 7.85, 1 H); 7.43 (*d*, *J* = 7.85, 1 H); 7.34 (*d*, *J* = 8.8, 1 H). ^{1}H-NMR ($CDCl_3$): 9.27 (*d*, *J* = 2.25, 1 H); 7.8 (*d*, *J* = 2.27, 1 H); 7.58 (*d*, *J* = 8.8, 1 H); 7.11 (*dd*, *J* = 2.25, 8.8, 1 H); 6.98 (*d*, *J* = 8.8, 1 H); 6.84 (*dd*, *J* = 2.27, 8.8, 1 H). HR-CI-MS: 332 (12), 331 (4), 330 (19), 308 (11), 307 (15), 306 (70), 305 (40), 304 (100), 303 (29).

*2,6,10,12-Tetrachlorodibenzo[*b,f*][1,7]naphthyridine* (**10b**). Yield 83%. Yellow powder (dimethylacetamide). M.p. >400°. IR: 1603*m*, 1579*m*, 1543*m*, 1458*m*, 1393*m*, 1314*s*, 1235*w*, 1173*w*, 1139*w*, 1100*m*, 1085*m*, 1045*s*, 939*m*, 868*m*, 828v*s*, 787*w*, 731*w*, 688*m*, 665*w*, 589*w*, 561*m*. ^{1}H-NMR ($CDCl_3$): 9.75 (*s*, 1 H); 8.63 (*s*, 1 H); 8.39 (*d*, *J* = 9.8, 1 H); 8.075 (*d*, *J* = 8.6, 1 H); 7.9 (*d*, *J* = 9.8, 1 H); 7.78 (*d*, *J* = 8.6, 1 H). HR-CI-MS: 372 (10), 371 (8), 370 (48), 369 (21), 368 (100), 367 (15), 366 (72), 333 (11), 332 (4), 331 (12), 298 (17), 297 (6), 296 (27), 262 (2), 261 (7).

1,2-Dihydro-6-methyl-3-[(4-methylphenyl)amino]-2-oxoquinoline-4-carboxylic Acid (**9c**). Yield 79%. Citron-yellow needles (MeOH). M.p. 229°. IR: 3385*s* (br.), 2875*m*, 1687*s*, 1658v*s*, 1565*s*, 1523*s*, 1472*w*, 1405*w*, 1368*w*, 1313*w*, 1258*m*, 1236*w*, 1187*w*, 945*w*, 898*w*, 859*w*, 809*s*, 774*w*, 684*w*, 614*w*, 530*w*. ^{1}H-NMR ((D_6)DMSO): 12.15 (*s*, 1 H); 7.4 (*s*, 1 H); 7.21 (*d*, *J* = 8.8, 1 H); 7.15 (*d*, *J* = 8.8, 1 H); 7.0 (*d*, *J* = 7.8, 2 H); 6.88 (*d*, *J* = 7.8, 2 H); 2.3 (*s*, 3 H); 2.21 (*s*, 3 H). ^{1}H-NMR (ethyl ester, $CDCl_3$): 7.52 (*s*, 1 H); 7.4 (*s*, 1 H); 7.28 (*d*, *J* = 8.05, 1 H); 7.11–7.09 (*m*, 3 H); 7.03 (*d*, *J* = 8.05, 2 H); 3.46 (*q*, *J* = 7.02, 2 H); 2.36 (*s*, 3 H); 2.31 (*s*, 3 H); 1.05 (*t*, *J* = 7.03, 3 H). ^{13}C-NMR (ethyl ester, $CDCl_3$): 166.4 (COO); 160.0 (CON); 138.3; 133.6; 133.2; 132.8; 129.7; 127.7; 123.0; 121.5; 118.7; 115.9; 112.9; 61.0 (Me*C*H$_2$); 21.3 (*Me*Ar); 20.8 (*Me*Ar); 13.8 (*Me*CH$_2$). HR-CI-MS: 337 (25), 336 (100), 290 (15), 275 (11), 263 (11), 262 (43), 261 (10).

*5,7-Dihydro-2,10-dimethyldibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4c**). Yield 70%. Yellow powder (dimethylacetamide). M.p. >400°. UV/VIS: 325 (0.33), 339 (0.51), 358 (0.3), 375 (0.53), 396 (0.55), 415 (sh, 0.06). IR: 3160 (br.), 3024*m*, 2924*m*, 1670v*s*, 1602v*s*, 1558*s*, 1537*s*, 1504*s*, 1453*w*, 1421*m*, 1386*m*, 1342*w*, 1322*w*, 1291*m*, 1259*w*, 1209*w*, 1195*w*, 1160*w*, 1039*w*, 993*w*, 903*w*, 874*w*, 822*m*, 770*m*, 710*m*, 670*w*, 622*w*, 603*m*, 576*w*, 551*w*, 484*w*, 472*w*. ^{1}H-NMR ((D_6)DMSO): 12.42 (*s*, 1 H); 12.15 (*s*, 1 H); 9.58 (*s*, 1 H); 8.07 (*s*, 1 H); 8.01 (*d*, *J* = 7.8, 1 H); 7.58 (*d*, *J* = 8.8, 1 H); 7.3–7.24 (*m*, 2 H); 2.45 (*s*, 3 H); 2.41 (*s*, 3 H). HR-CI-MS: 292 (8), 291 (50), 290 (100). Anal. calc. for $C_{18}H_{14}N_2O_2$ (290.31): C 74.47, H 4.86, N 9.65; found: C 74.36, H 4.71, N 9.59.

*2,10-Dimethyl-6,12-dichlorodibenzo[*b,f*][1,7]naphthyridine* (**10c**). Yield 70%. White needles (dimethylacetamide). M.p. >400°. UV/VIS (DMF): 325 (0.22), 339 (0.35), 357 (0.2), 375 (0.31), 395 (0.32). IR: 1692*s*, 1614*s*, 1581*m*, 1547*w*, 1477*s*, 1397*m*, 1324*w*, 1311*s*, 1181*w*, 1165*w*, 1140*w*, 1115*w*, 1061*w*, 979*w*, 873*w*, 831*s*, 812*s*, 729*m*, 679*w*, 626*w*, 588*m*, 528*w*, 464*w*. ^{1}H-NMR ((D$_6$)DMSO): 9.5 (*s*, 1 H); 8.44 (*s*, 1 H); 8.3 (*d*, *J* = 8.57, 1 H); 7.99 (*d*, *J* = 8.57, 1 H); 7.95 (*d*, *J* = 8.6, 1 H); 7.74 (*d*, *J* = 8.6, 1 H); 2.69 (*s*, 3 H); 2.64 (*s*, 3 H).

1,2-Dihydro-6-methoxy-3-[(4-methoxyphenyl)amino]-2-oxoquinoline-4-carboxylic Acid (**9d**). Yield 64%. Citron-yellow needles (MeOH). M.p. 140° (dec.). IR: 3341*s*, 2932*m*, 2833*m*, 1675*s*, 1660 (sh), 1615*s*, 1588*m*, 1555*w*, 1512v*s*, 1454*w*, 1411*w*, 1366*m*, 1321*m*, 1295*s*, 1245*s*, 1219*s*, 1190*m*, 1169*m*, 1129*w*, 1035*m*, 904*w*, 842*m*, 826*w*, 808*w*, 781*w*, 729*w*, 710*w*, 678*w*, 610*m*, 544*w*. ^{1}H-NMR ((D$_6$)DMSO): 12.12 (*s*, 1 H); 7.81 (*s*, 1 H); 7.23 (*d*, *J* = 8.8, 1 H); 7.1 (*s*, 1 H); 6.99–6.94 (*m*, 3 H); 6.79 (*d*, *J* = 8.8, 2 H); 3.7 (*s*, 6 H). ^{13}C-NMR ((D$_6$)DMSO): 167.38; 158.06; 156.0; 155.13; 154.8; 134.7; 133.0; 126.8; 122.6; 118.9; 116.4; 113.8; 105.7; 55.23. HR-CI-MS (methyl ester): 355 (1), 354 (1), 353 (7), 338 (4), 189 (11), 188 (100), 150 (12), 109 (15). Anal. calc. for $C_{18}H_{16}N_2O_5$ (340.34): C 63.53, H 4.74, N 8.23; found: C 63.41, H 4.56, N 8.18.

*5,7-Dihydro-2,10-dimethoxydibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4d**). Yield 63%. Tan solid. M.p. 140° (dec.). ^{1}H-NMR ((D$_6$)DMSO): 12.38 (*s*, 1 H); 12.29 (*s*, 1 H); 9.45 (*s*, 1 H); 8.08 (*d*, *J* = 9.78, 1 H); 7.7 (*s*, 1 H); 7.42 (*d*, *J* = 8.8, 1 H); 7.32 (*d*, *J* = 8.8, 1 H); 7.07 (*d*, *J* = 9.78, 1 H); 3.88 (*s*, 3 H); 3.83 (*s*, 3 H). ^{13}C-NMR ((D$_6$)DMSO): 176.7; 156.5; 155.9; 155.0; 133.8; 133.4; 127.0; 126.05; 123.7; 121.2; 118.8; 116.3; 115.2; 113.3; 108.8; 104.3; 55.4; 55.2.

3-[(3,4-Dimethylphenyl)amino]-1,2-dihydro-6,7-dimethyl-2-oxoquinoline-4-carboxylic Acid (**9e**). Following the procedure reported in [15], a bright yellow powder was obtained in 78% yield, consisting of 3-[(3,4-dimethylphenyl)amino]-1,2-dihydro-6,7-dimethyl-2-oxoquinoline-4-carboxylic acid methyl ester and *ca.* 18% of 3-[(3,4-dimethylphenyl)amino]-2-hydroxy-5,6-dimethylquinoline-4-carboxylic acid methyl ester. This ester mixture was saponified with base and the resulting acid mixture precipitated by acid. Part of the acid mixture (5 g) was recrystallized with MeOH (30 ml) by heating and cooling to r.t. The crystals from three such recrystallizations were pooled and recrystallized again in MeOH to yield pure **9e** in 33% yield. Bright yellow solid. M.p. 278°. IR: 3379*s*, 2919*m*, 1682*s*, 1642*s*, 1609*w*, 1583*w*, 1547*s*, 1514*s*, 1447*m*, 1405*m*, 1359*w*, 1320*m*, 1263*w*, 1241*m*, 1216*m*, 1193*m*, 1118*w*, 969*w*, 864*w*, 820*w*, 788*w*, 752*w*, 733*m*. ^{1}H-NMR ((D$_6$)DMSO): 12.07 (*s*, 1 H); 7.56 (*s*, 1 H); 7.36 (*s*, 1 H); 7.08 (*s*, 1 H); 6.91 (*d*, *J* = 7.2, 1 H); 6.75 (*s*, 1 H); 6.7 (*d*, *J* = 7.2, 1 H); 2.24 (*s*, 3 H); 2.21 (*s*, 3 H); 2.1 (*s*, 3 H); 2.09 (*s*, 3 H). ^{13}C-NMR ((D$_6$)DMSO): 167.35; 158.85; 140.3; 135.98; 135.76; 131.73; 130.79; 130.24; 129.54; 128.94; 123.64; 120.07; 119.14; 116.5; 115.74; 115.62; 19.57; 19.54; 19.35; 18.68. HR-CI-MS: 338 (19.5), 337 (100), 319 (19), 293 (21), 150.0 (19). Anal. calc. for $C_{20}H_{20}N_2O_3$ (336.4): C 71.41, H 5.99, N 8.33; found: C 70.36, H 5.99, N 8.21.

*5,7-Dihydro-2,3,9,10-tetramethyldibenzo[*b,f*][1,7]naphthyridine-6,12-dione*/*5,7-Dihydro-2,3,10,11-tetramethyldibenzo[*b,f*][1,7]naphthyridine-6,12-dione* (**4e**/**4e′**). According to the general protocol, a suspension of pure **9e** (1.0 g, 0.001 mol) was stirred in PCl_3/PCl_5/$AlCl_3$ and worked up after 1.5 h refluxing time as indicated. Yellow solid (68%). After recrystallization from DMF, a powder was obtained consisting of **4e**/**4e′** 3 : 1 (two sets of ^{1}H-NMR signals). ^{1}H-NMR ((D$_6$)DMSO): **4e**: 12.29 (*s*, 1 H); 11.71 (*s*, 1 H); 9.35 (*s*, 1 H); 8.01 (*s*, 1 H); 7.83 (*s*, 1 H); 7.13 (*s*, 1 H); 2.5–2.3 (*m*, 12 H); **4e′**: 12.32 (*s*, 1 H); 11.93 (*s*, 1 H); 9.5 (*s*, 1 H); 7.72 (*d*, *J* = 7.28, 1 H); 7.44 (*d*, *J* = 7.28, 1 H); 7.13 (*s*, 1 H); 2.5–2.3 (*m*, 12 H).

REFERENCES

[1] T. Kokobun, J. Edmonds, P. John, *Phytochemistry* **1997**, *49*, 79.

[2] K. G. Gilber, D. J. Hill, C. Crespo, A. Mas, M. Lewis, D. T. Cooke, *Phytochem. Anal.* **2000**, *11*, 18.

[3] W. Tang, G. Eisenbrand, 'Chinese Drugs of Plant Origin', Verlag Springer, Berlin – Heidelberg, 1992, p. 805.

[4] D. Marko, S. Schaetzle, A. Friedel, A. Genzlinger, H. Zankl, L. Meijer, G. Eisenbrand, *Br. J. Cancer* **2001**, *84*, 283.

[5] B. C. De Landsheere, *Experientia* **1951**, *7*, 307.

[6] X. Wu, G. Qin, K. K. Cheung, K. F. Cheng, *Tetrahedron* **1997**, *53*, 13323.
[7] P. Molina, A. Tarraga, A. Gonzales-Tejero, I. Rioja, A. Ubeda, M. C. Terenico, M. J. Alcaraz, *J. Nat. Prod*. **2001**, *64*, 1297.
[8] Q. H. Li, *Acta Bot. Sin*. **1987**, *29*, 67.
[9] E. E. Jaffe, H. Matrick, *J. Org. Chem*. **1968**, *33*, 4004.
[10] R. Gompper, R. Kunz, *Chem. Ber*. **1965**, *98*, 1391.
[11] M. W. Partridge, J. M. Sprake, H. J. Vipond, *J. Chem. Soc., C* **1966**, 1245.
[12] H. B. MacPhillamy, R. L. Dziemian, R. A. Lucas, M. E. Kuehne, *J. Am. Chem. Soc.* **1957**, *80*, 2172.
[13] M. Colonna, *Gazz. Chim. Ital*. **1949**, *78*, 503.
[14] R. Anet, *Can. J. Chem*. **1959**, *37*, 43.
[15] E. H. Huntress, J. Bornstein, W. H. Hearon, *J. Am. Chem. Soc.* **1956**, *78*, 2225.
[16] E. Enders, 'Methoden der Organischen Chemie', 4th edn., Ed. E. Müller, Thieme-Verlag, Stuttgart, 1967, Vol. X/2, p. 706ff.
[17] Y. Z. Guo, F. K. Chen, *Chin. Tradit. Herb. Drugs* **1986**, *17*, 8.
[18] C. Oberthür, M. Hamburger, *Planta Med.* **2004**, *70*, 642; C. Oberthür, H. Graf, M. Hamburger, *Phytochemistry* **2004**, *65*, 3261.

Received August 29, 2008