Structure−Activity Relationship for Antineoplastic Imidazoacridinones: Synthesis and Antileukemic Activity *in Vivo*

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Synthesis of several new 5-amino-substituted derivatives of 5-amino-6*H*-imidazo[4,5,1-*de*]-acridin-6-one bearing in the benzene ring OH, OCH_3, CH_3, *tert*-butyl, or OCH_2O groups is described. 8-OH-Substituted compounds or double-substituted 7-OH-10-OCH_3 compounds demonstrated potent *in vivo* activity against murine P388 leukemia. The highest activity was exhibited by 5-[[2-[[2-(diethylamino)ethyl]amino]ethyl]amino]-8-hydroxy-6*H*-imidazo[4,5,1-*de*]-acridin-6-one (**4c**).

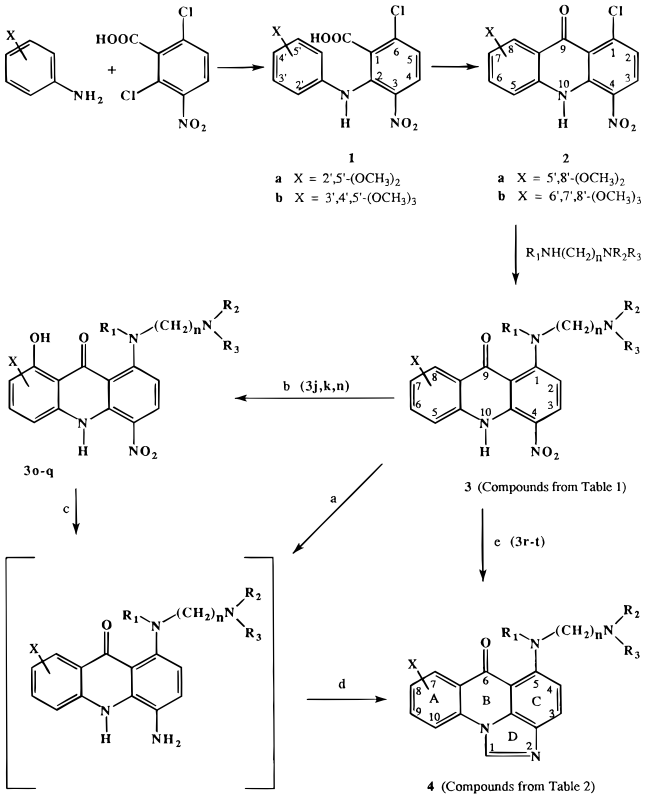
Introduction

In our recent papers derivatives of 5-amino-6*H*-imidazo[4,5,1-*de*]acridin-6-one were reported as a novel class of antineoplastic agents.[1,2] The compounds substituted with a hydroxy group in position 8 were found to be especially active against transplantable murine tumors: leukemia P388, melanoma B-16, and two colon adenocarcinomas, C-26 and C-38.[3] The derivatives also presented significant and diversified cytotoxic activity against 66 human tumor cell lines in the NCI *in vitro* screening system. Several of them passed this screening test and are being considered for further testing on human tumor xerografts in nude mice (unpublished data). The most active derivatives are presently examined extensively in other preclinical tests.

The earlier structure−activity studies revealed that antitumor activity of 5-amino-6*H*-imidazo[4,5,1-*de*]acridin-6-one derivatives substituted in position 8 with a hydroxy or methoxy group is higher than for unsubstituted compounds.[1,2] Structure of the aliphatic side chain seemed to have only secondary influence, providing it was an ethylene- or propylenediamine chain.[2]

The present paper describes the synthesis of new 5-amino-6*H*-imidazo[4,5,1-*de*]acridin-6-one derivatives unsubstituted as well as diversely substituted with hydroxy or methoxy groups in ring A. It was of interest to examine how the oxygen substituents, their location and number, influence biological activity, and for this reason congeners with hydroxy and methoxy groups in positions other than 8, as well as with the position 8 blocked by non-oxygen substituents, were prepared. Compounds possessing, in ring A of imidazoacridinone, hydroxy, methoxy, or methylenedioxy groups in position ortho or para to each other which, by some metabolic transformation, could be transformed into an *o*- or *p*-quinone system could be considered as a special group. Some structure−activity studies concerning the side chain of imidazoacridinones are also performed to learn more about the relevance of side chain structure for biological activity, as well as susceptibility to modification.

Scheme 1[a]



[a] Reagents: (a) and (c) NH_2NH_2/Raney Ni/THF; (b) concentrated HCl/reflux; (d) HCOOH/reflux; (e) HCOOOH/Ni−Al alloy/reflux.

Chemistry

A general route to the target 5-[(aminoalkyl)amino]-6*H*-imidazo[4,5,1-*de*]acridin-6-ones **4** is presented in Scheme 1. The starting substituted 1-chloro-4-nitro-9-(10*H*)acridinones **2** were obtained by cyclization of the corresponding *N*-phenylanthranilic acids **1** according to the previously described methods.[1,4,5] Two new *N*-phenylanthranilic acids, **1a**,**b**, were prepared by condensation of 2,6-dichloro-3-nitrobenzoic acid with a suitable amine in ethyl alcohol in the presence of triethylamine. Condensation of acridinones **2** with an excess of the corresponding amine in *N*,*N*-dimethylformamide provided then the desired nitroacridinones **3a**−**u**. Their physical properties are presented in Table 1.

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Table 1. Physical Properties of the 1-Substituted 4-Nitro-9(10*H*)acridinones **3**

no.	X	*n*	R_1	R_2	R_3	cryst solvent	mp, °C	yield, %	formula[a]
3a	7-OH	5	H	CH_3	CH_3		237−239	80	$C_{20}H_{24}N_4O_4$
3b	H	2	H	H	$CH_2CH_2N(C_2H_5)_2$		180−181	90	$C_{21}H_{27}N_5O_3$
3c	7-OH	2	H	H	$CH_2CH_2N(C_2H_5)_2$		213−215	70	$C_{21}H_{27}N_5O_4$
3d	H	2	CH_3	CH_3	CH_3		133−134	85	$C_{18}H_{20}N_4O_3$
3e	7-OH	2	CH_3	CH_3	CH_3		127−128	60	$C_{18}H_{20}N_4O_4$
3f	7-OH	2	H	$(CH_2)_3CH_3$	$(CH_2)_3CH_3$		200−202	85	$C_{23}H_{30}N_4O_4$
3g	7-OH	2	H	$CH(CH_3)_2$	$CH(CH_3)_2$		243−245	90	$C_{21}H_{26}N_4O_4$
3h	6-OH	2	H	CH_2CH_3	CH_2CH_3	acetone	189−191	80	$C_{19}H_{22}N_4O_4$
3i	5-OCH_3	2	H	CH_2CH_3	CH_2CH_3	DMF−H_2O	166−167	95	$C_{20}H_{24}N_4O_4$
3j	5,8-$(OCH_3)_2$	2	H	CH_2CH_3	CH_2CH_3		161−162	95	$C_{21}H_{26}N_4O_5$
3k	5,8-$(OCH_3)_2$	2	H	CH_3	CH_3		152−153	97	$C_{19}H_{22}N_4O_5$
3l	6,7-$(OCH_3)_2$	2	H	CH_2CH_3	CH_2CH_3		208−210	96	$C_{21}H_{26}N_4O_5$
3m	6,7-$(OCH_3)_2$	3	H	CH_3	CH_3		206	95	$C_{20}H_{24}N_4O_5$
3n	6,7,8-$(OCH_3)_3$	2	H	CH_2CH_3	CH_2CH_3	benzene−hexane	167−168	89	$C_{22}H_{28}N_4O_6$
3o	5-OCH_3,8-OH	2	H	CH_2CH_3	CH_2CH_3	chloroform−hexane	171−172	77	$C_{20}H_{24}N_4O_5$
3p	5-OCH_3,8-OH	2	H	CH_3	CH_3	chloform−hexane	180−181	80	$C_{18}H_{20}N_4O_5$
3q	6,7-$(OCH_3)_2$,8-OH	2	H	CH_2CH_3	CH_2CH_3	chloroform−hexane	204−206	86	$C_{21}H_{26}N_4O_6$
3r	6,7-(-OCH_2O-)	2	H	CH_2CH_3	CH_2CH_3	chloroform	234−235	60	$C_{20}H_{22}N_4O_5$
3s	6,7-(-OCH_2O-)	3	H	CH_3	CH_3	chloroform	240−241	65	$C_{19}H_{20}N_4O_5$
3t	7-CH_3	2	H	CH_2CH_3	CH_2CH_3	benzene−hexane	198−199	91	$C_{20}H_{24}N_4O_3$
3u	7-*tert*-butyl	2	H	CH_2CH_3	CH_2CH_3	hexane	121−122	90	$C_{23}H_{29}N_4O_3$

[a] All compounds were analyzed for C, H, N, and the results are within ±0.4% of the theoretical values.

Table 2. Physical Properties of the Target Imidazoacridinones **4** from Scheme 1 and Their Activities against P388 Leukemia in Mice

									P388 leukemia	
no.	X	*n*	R_1	R_2	R_3	mp, °C[a]	yield, %	formula[b]	opt dose (mg/kg/day)	T/C, %[c]
4a	8-OH	5	H	CH_3	CH_3	237−239	64	$C_{21}H_{24}N_4O_2{\cdot}2HCl{\cdot}H_2O$	70	172
4b	H	2	H	H	$CH_2CH_2N(C_2H_5)_2$	165−167	60	$C_{22}H_{27}N_5O{\cdot}2HCl{\cdot}1.5H_2O$	100	130
4c	8-OH	2	H	H	$CH_2CH_2N(C_2H_5)_2$	150−151	50	$C_{22}H_{27}N_5O_2{\cdot}2HCl{\cdot}H_2O$	100	254
4d	H	2	CH_3	CH_3	CH_3	211−213	55	$C_{19}H_{20}N_4O{\cdot}2HCl$	100	180
4e	8-OH	2	CH_3	CH_3	CH_3	220−223	40	$C_{19}H_{20}N_4O_2{\cdot}2HCl{\cdot}0.5H_2O$	6.25	210
4f	8-OH	2	H	$(CH_2)_3CH_3$	$(CH_2)_3CH_3$	168−170	66	$C_{24}H_{30}N_4O_2{\cdot}2HCl{\cdot}0.5H_2O$	12.5	110
4g	8-OH	2	H	$CH(CH_3)_2$	$CH(CH_3)_2$	204−206	65	$C_{22}H_{26}N_4O_2{\cdot}2HCl$	25	200
4h	9-OH	2	H	CH_2CH_3	CH_2CH_3	180−181	40	$C_{20}H_{22}N_4O_2{\cdot}2HCl{\cdot}H_2O$	25	127
4i	10-OH	2	H	CH_2CH_3	CH_2CH_3	179−182	50	$C_{20}H_{22}N_4O_2{\cdot}2HCl$	25	100
4j	7,10-$(OH)_2$	2	H	CH_2CH_3	CH_2CH_3	250−252	20	$C_{20}H_{22}N_4O_3{\cdot}2HCl$	50	145
4k	10-OCH_3	2	H	CH_2CH_3	CH_2CH_3	201−202	70	$C_{21}H_{24}N_4O_2{\cdot}2HCl$	200	130
4l	7,10-$(OCH_3)_2$	2	H	CH_2CH_3	CH_2CH_3	220−222	20	$C_{22}H_{26}N_4O_3{\cdot}2HCl$	25	115
4m	8,9-$(OCH_3)_2$	2	H	CH_2CH_3	CH_2CH_3	170	60	$C_{22}H_{26}N_4O_3{\cdot}2HCl$	50	110
4n	8,9-$(OCH_3)_2$	3	H	CH_3	CH_3	221−223	40	$C_{21}H_{24}N_4O_3{\cdot}1.5HCl{\cdot}H_2O$	50	115
4o	7-OH,10-OCH_3	2	H	CH_2CH_3	CH_2CH_3	195	20	$C_{21}H_{24}N_4O_3{\cdot}2HCl{\cdot}H_2O$	50	185
4p	7-OH,10-OCH_3	2	H	CH_3	CH_3	122−123	30	$C_{19}H_{20}N_4O_3{\cdot}2HCl{\cdot}H_2O$	7.3	163
4q	7-OH,8,9-$(OCH_3)_2$	2	H	CH_2CH_3	CH_2CH_3	186−188	60	$C_{22}H_{26}N_4O_4{\cdot}1.5HCl{\cdot}H_2O$	100	115
4r	8,9-(-OCH_2O-)	2	H	CH_2CH_3	CH_2CH_3	250−252	56	$C_{21}H_{22}N_4O_3{\cdot}2HCl{\cdot}H_2O$	100	120
4s	8,9-(-OCH_2O-)	3	H	CH_3	CH_3	240−241	60	$C_{20}H_{20}N_4O_3{\cdot}2HCl$	50	122
4t	8-CH_3	2	H	CH_2CH_3	CH_2CH_3	210−212	45	$C_{21}H_{24}N_4O{\cdot}2HCl{\cdot}H_2O$	200	115
4u	8-*tert*-butyl	2	H	CH_2CH_3	CH_2CH_3	142−143	35	$C_{24}H_{30}N_4O{\cdot}2HCl$	25	110

[a] All compounds were crystallized from methanol−ether. [b] Elemental analyses are within ±0.4% of the theoretical values for C, H, N. [c] BDF_1 mice were implanted ip with 10^6 P388 cells on day 0, and each compound was administered ip as aqueous solution on days 1−5. Each group except control consisted of seven mice. The control group consisted of 10 mice. Values of T/C ≥ 125% indicate statistically significant antileukemic activity. For general procedure, see ref 6.

Imidazoacridinones **4** were prepared by hydrazine−Raney nickel reduction of nitroacridinones **3** to the corresponding amino derivatives followed by cyclization with formic acid. The overall process was performed in different pathways. The usual way was reduction with hydrazine−Raney nickel in THF (path a). The reduction products were isolated as the hydrochloride salts and crystallized from methanol−ether. Since the obtained hydrochlorides were rather unstable, they were not characterized but used directly in the cyclization process. Attempts to reduce the methoxy derivatives **3j**,**k**,**n** led to complex mixtures of products. Therefore, they were hydrolyzed, prior to reduction, by refluxing with concentrated hydrochloric acid to give products **3o**−**q** selectively deprotected in position 8 (path b). These compounds were isolated as hydrochlorides, transformed into free bases, and reduced cleanly with hydrazine to give the suitable amino derivatives. All the amino derivatives of compounds **3a**−**j**,**l**,**m**,**o**−**q**,**u** obtained with paths a and b were transformed in high yield (95%) into the corresponding imidazoacridinones **4a**−**h**,**k**−**q**,**u** by refluxing in formic acid. Compounds **3r**−**t** could not be reduced with hydrazine, probably because of their poor solubility in THF or dioxane. They were transformed into the corresponding imidazoacridinones **4r**−**t** by a one-pot reduction−cyclization in 80% formic acid in the presence of nickel aluminum alloy (path e). All imidazoacridinones were isolated as hydrochlorides and crystallized from methanol−ether. Hydroxy derivatives **4i**,**j** were prepared from the corresponding methoxy compounds **4k**,**o** in 50% yield by reaction with boron tribromide−methyl sulfide complex. Yields and physical properties of the target compounds **4** are reported in Table 2. The structures of the

synthesized compounds were established on the basis of NMR studies, using spin-decoupling methods for aromatic proton assignment.

Biological Results and Discussion

The antineoplastic activity of all the synthesized derivatives of 5-[(aminoalkyl)amino]-6*H*-imidazo[4,5,1-*de*]acridin-6-ones **4a**−**u** was tested *in vivo* against murine leukemia P388 (ip/ip: days 1−5). For each compound a full drug dose−response study was carried out, with doses ranging from ineffective to toxic. The optimal dose was defined as that which produced the highest T/C value. The results obtained at the optimal dose, a median from three independent tests, are presented in Table 2. The results, in connection with those published earlier,[1,2] allow to formulate structure−activity relationships for both side chain and ring A modifications of 5-[(aminoalkyl)amino]-6*H*-imidazo-[4,5,1-*de*]acridin-6-one derivatives.

The following generalizations concerning the side chain can be deduced from the data. (1) The distance between the two amino groups does not seem to be important for 8-hydroxyacridinones, as even compound **4a**, which has a long spacer (five methylene units) in the side chain, still possesses significant activity. (2) The proximal amino group does not have to be secondary as its methylation, as in compounds **4d**,**e**, does not spoil activity. (3) Elongation of the side chain by a second ethyleneamino fragment results in very high activity, provided that the 8-OH substituent is present (compound **4c**). In fact, compound **4b**, with identical side chain but lacking in 8-OH group, has only marginal activity (T/C of 130%). (4) The significance of bulkiness of alkyl substituents on the distal amino group is not clear. Dimethyl and diethyl derivatives are active,[2] related di-*n*-butyl compound **4f** is inactive, but, surprisingly, a compound bearing the space-demanding isopropyl substituent (**4g**) is still active. The first two observations are of special interest as they are in opposition to those made for related 5-aminotriazoloacridinones, for which a secondary proximal amino group and ethylene or propylene side chain seem to be obligatory.[7]

Substitution pattern of the A ring of the studied compounds was found to be important. The most essential was the presence of a hydroxy substituent in position 8 (compounds **4a**,**c**,**e**,**g**), and in accordance with the previous results,[2] all the compounds were active. Derivatives having a hydroxy substituent in position 9 (**4h**) or 10 (**4i**) were inactive. It is possible that the 8-hydroxy-substituted compounds are active because, possessing the OH group in para position to the nuclear nitrogen of the acridine system, they could be metabolically transformed into compounds with quinone imine structures, able to form adducts with DNA. Similar mechanism of action was found for 8-hydroxyellipticine, which also has a heterocyclic nitrogen situated in para position relative to the hydroxy substituent.[8] By analogy with ellipticine, it can also be supposed that the activity of 8-unsubstituted compounds relies on initial enzymatic oxidation to the hydroxy derivatives. This supposition is supported by the findings that substitution of the position 8 with another group, methyl (**4t**) or *tert*-butyl (**4u**), leads to inactive compounds, whereas in our previous works we have found that the corresponding 8-hydroxy derivative (compound **4k** of ref 2) and the 8-unsubstituted derivative 5-[[(dimethylamino)-ethyl]amino]imidazo[4,5,1-*de*]acridin-6-one (compound **11** of ref 1) are active.

To explore the influence of other types of quinones on biological activity of imidazoacridinones, compounds with a potential *p*-quinoid system in ring A were synthesized. 7,10-Dihydroxy (**4j**) and 7,10-dimethoxy (**4l**) derivatives were found to be inactive, but surprisingly, 7-hydroxy-10-methoxy compounds **4o**,**p** exhibit significant biological activity. Derivatives bearing a potential *o*-quinoid system, namely, 8,9-dimethoxy (**4m**,**n**), 7-hydroxy-8,9-dimethoxy (**4q**), and 8,9-methylenedioxy (**4r**,**s**) imidazoacridinones, also were found to be inactive. It is worthwhile to note that in mammalian cells the methylenedioxy group could be metabolized to catechol.[9,10]

In conclusion, it can be generalized that the most promising group of active imidazoacridinones is the 8-hydroxy derivatives, which potentially are able to form a quinone imine system which probably resulted in increased ability for covalent binding to DNA. Replacing this potential quinone imine system by potential quinone systems (ortho or para) resulted in diminishment or loss of biological activity.

Experimental Section

^{1}H NMR spectra of the synthesized compounds were obtained with either a Varian VXR-300 or a Varian Gemini 200 MHz spectrometer, using TMS as an internal standard. NMR abbreviations used are as follows: br (broad), s (singlet), d (doublet), t (triplet), qu (quartet), qt (quintet), m (multiplet), and ex (exchangeable with deuterium oxide). *J* values are reported in hertz (Hz). Elemental analyses were performed by the Laboratory of Elemental Analysis of Department of Chemical Sciences, University of Camerino, or by Laboratory of Elemental Analysis, University of Gdansk.

***N*-(2′,5′-Dimethoxyphenyl)-6-chloro-3-nitroanthranilic Acid (1a).** A solution of 11.8 g (0.05 mol) of 2,6-dichloro-3-nitrobenzoic acid in 100 mL of ethanol was added to 15.5 g (0.1 mol) of 2,5-dimethoxyaniline and 15 mL (0.1 mol) of triethylamine. The mixture was refluxed for 100 h under argon. The solvent was evaporated, and the residue was dissolved in 2% aqueous solution of Na_2CO_3, heated to boiling with charcoal, and filtered after cooling. The filtrate was acidified with hydrochloric acid, and the orange precipitate was washed with water, dried, and crystallized from toluene providing 5.34 g (30%) of **1a**, mp 172−174 °C. Anal. ($C_{15}H_{13}ClN_2O_6$) C, H, N.

***N*-(3′,4′,5′-Trimethoxyphenyl)-6-chloro-3-nitroanthranilic Acid (1b).** A mixture of 11.9 g (0.065 mol) of 3,4,5-trimethoxyaniline, 11.8 g (0.05 mol) of 2,6-dichloro-3-nitrobenzoic acid, 30 mL of ethanol, and 10 mL (0.07 mol) of triethylamine was heated under reflux 40 h. To the reaction mixture was added 80 mL of 5 N aqueous solution of NaOH, and ethanol and triethylamine were distilled off. The orange sodium salt precipitate was stirred with 200 mL of benzene, collected, and dried. It was dissolved in 400 mL of water and acidified with hydrochloric acid. The formed precipitate was washed with water and crystallized from acetone−water to give 13.4 g (70%) of red crystals of **1b**, mp 212−214 °C. Anal. ($C_{16}H_{15}ClN_2O_7$) C, H, N.

1-Chloro-4-nitro-5,8-dimethoxy-9(10*H*)-acridinone (2a). A mixture of *N*-(2′,5′-dimethoxyphenyl)-6-chloro-3-nitroanthranilic acid (**1a**) (10 g, 0.028 mol), phosphorus oxychloride (10 mL), *N*,*N*-dimethylaniline (1 mL), and chloroform (50 mL) was refluxed for 1 h. The solvents were evaporated under reduced pressure, and the residue was added to a mixture of acetic acid (50 mL) and water (200 mL) and stirred for 2 h. The formed red precipitate was filtered and crystallized from toluene to give **2a** (6.7 g, 71%), mp 146−148 °C. ^{1}H NMR

(DMSO-d_6): δ 11.8 (1H, s, ex, N**H**), 8.48 (1H, d, J = 8.8, C3-**H**), 7.24 (1H, d, J = 8.8, C2-**H**), 7.11 (1H, d, J = 8.8, C6-**H**), 6.67 (1H, d, J = 8.8, C7-**H**), 4.06 (3H, s, C5-OC**H**$_3$), 3.98 (3H, s, C8-OC**H**$_3$). Anal. ($C_{15}H_{11}ClN_2O_5$) C, H, N.

1-Chloro-4-nitro-6,7,8-trimethoxy-9(10*H*)-acridinone (2b). This compound was obtain in an analogous manner as **2a**. Yield was 85% after crystallization from chloroform−hexane, mp 242−246 °C. ^{1}H NMR (DMSO-d_6): δ 11.30 (1H, s, ex, N**H**), 8.46 (1H, d, J = 8.8, C3-**H**), 7.48 (1H, s, C5-**H**), 7.32 (1H, d, J = 8.8, C2-**H**), 3.92 (3H, s, OC**H**$_3$), 3.84 (3H, s, OC**H**$_3$), 3.74 (3H, s, OC**H**$_3$). Anal. ($C_{16}H_{13}ClN_2O_6$) C, H, N.

General Procedure for the Preparation of 1-[[(Dialkylamino)alkyl]amino]-4-nitro-9(10*H*)-acridinones 3a−n,r−u: 1-[[2-[[2-(Diethylamino)ethyl]amino]ethyl]amino]-4-nitro-9(10*H*)-acridinone (3b). A mixture of 1-chloro-4-nitro-9(10*H*)-acridinone (2.9 g, 0.01 mol), 2-[(diethylamino)ethyl]-ethylenediamine (6.3 g, 0.04 mol), and DMF (15 mL) was stirred and heated at 80 °C for 30 min. Next, a mixture of acetone and water (v/v 8:2, 100 mL) was added, and the mixture was left overnight in a refrigerator. The precipitated solid was filtered off and washed twice with water and methanol to give pure **3c**. ^{1}H NMR (DMSO-d_6): δ 12.90 (1H, s, ex, N10-**H**), 11.90 (1H, t, ex, N**H**-CH$_2$), 8.36 (1H, d, J = 9.9, C3-**H**), 8.20 (1H, d, J = 8.8, C8-**H**), 7.96 (1H, d, C5-**H**), 7.78 (1H, t, C7-**H**), 7.42 (1H, t, C6-**H**), 6.42 (1H, d, J = 9.9, C2-**H**), 3.55 (2H, qu, NHC**H**$_2$CH$_2$), 2.87 (2H, t, NHCH$_2$C**H**$_2$), 2.62 (2H, t, CH$_2$NHC**H**$_2$), 2.45 (6H, m, C**H**$_2$N(C**H**$_2$CH$_3$)$_2$), 0.95 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

1-[[2-(Diethylamino)ethyl]amino]-5,8-dimethoxy-9(10*H*)-acridinone (3j). ^{1}H NMR (DMSO-d_6): δ 12.56 (1H, s, ex, N10-**H**), 11.80 (1H, t, ex, N**H**-CH$_2$), 8.28 (1H, d, J = 9.7, C3-**H**), 7.32 (1H, d, J = 9.0, C6-**H**), 6.76 (1H, d, J = 9.0, C7-**H**), 6.57 (1H, d, J = 9.7, C2-**H**), 4.00 (3H, s, 5-OC**H**$_3$), 3.80 (3H, s, 8-OC**H**$_3$), 3.60 (2H, qu, NHC**H**$_2$CH$_2$), 2.80 (2H, t, NHCH$_2$C**H**$_2$), 2.70 (4H, qu, N(C**H**$_2$CH$_3$)$_2$), 1.00 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

1-[[2-(Diethylamino)ethyl]amino]-6,7-(methylenedioxy)-9(10*H*)-acridinone (3r). ^{1}H NMR (DMSO-d_6): δ 12.65 (1H, s, ex, N10-**H**), 12.01 (1H, t, ex, N**H**-CH$_2$), 8.42 (1H, d, J = 8.9, C3-**H**), 7.71 (1H, s, C5-**H**), 6.86 (1H, s, C8-**H**), 6.38 (1H, d, J = 8.9, C2-**H**), 6.14 (2H, s, OC**H**$_2$O), 3.52 (2H, qt, NHC**H**$_2$CH$_2$), 2.86 (2H, t, NHCH$_2$C**H**$_2$), 2.68 (4H, qt, N(C**H**$_2$CH$_3$)$_2$), 1.12 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

General Procedure for the Preparation of 1-[[(Dialkylamino)alkyl]amino]-8-hydroxy-4-nitro-9(10*H*)-acridinones 3o-q (Table 1): 1-[[2-(Diethylamino)ethyl]amino]-8-hydroxy-5-methoxy-4-nitro-9(10*H*)-acridinone (3o). A mixture of compound **3j** (2.01 g, 0.05 mol) and concentrated hydrochloric acid (20 mL) was refluxed with stirring for 3 h. The solution was cooled down, and the product was precipitated by addition of acetone (60 mL). The precipitate was filtered off, washed with acetone, dissolved in water, alkalized with concentrated ammonia, and extracted with chloroform. The chloroformic solution was dried ($MgSO_4$) and evaporated to dryness, and the residue was crystallized from chloroform−hexane to give **3o**. ^{1}H NMR (DMSO-d_6): δ 12.70 (1H, s, ex, O**H**), 12.36 (1H, s, ex, N10-**H**), 11.26 (1H, t, ex, N**H**-CH$_2$), 8.40 (1H, d, J = 9.8, C3-**H**), 7.07 (1H, d, J = 8.8, C6-**H**) 6.60 (1H, d, J = 8.8, C7-**H**), 6.30 (1H, d, J = 9.8, C2-**H**), 3.98 (3H, s, 5-OC**H**$_3$), 3.50 (2H, qu, NH-C**H**$_2$CH$_2$), 2.88 (2H, t, NH-CH$_2$C**H**$_2$), 2.70 (4H, qu, N(C**H**$_2$CH$_3$)$_2$, 1.12 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

General Procedure for the Preparation of 5-[[(Dialkylamino)alkyl]amino]-6*H*-imidazo[4,5,1-*de*]acridin-6-ones 4a−h,k−q,u (Table 2): 5-[[2-(Diethylamino)ethyl]-amino]-10-methoxy-6*H*-imidazo[4,5,1-*de*]acridin-6-one (4k). A suspension of **3i** (2.5 g, 0.0065 mol) in THF (50 mL) was stirred for 30 min at room temperature. Next, Raney nickel (1 g) followed by hydrazine hydrate (1.3 mL, 0.026 mol) was slowly added, and the mixture was vigorously stirred for 1 h. The reaction mixture was filtered into THF containing gaseous HCl (100 mL); the precipitated solid was filtered off and crystallized quickly from a mixture methanol−ether. The obtained hydrochloride was dissolved in 95% formic acid (10 mL), and the solution was refluxed with stirring for 20 h. The reaction mixture was evaporated to dryness under reduced pressure, and the resulting oily residue was dissolved in hot methanol and decolorized with charcoal. Addition of ethereal HCl caused precipitation of product (**4k**) as dihydrochloride salt. ^{1}H NMR (free base) ($CDCl_3$): δ 9.13 (1H, t, ex, N**H**-CH$_2$), 9.10 (1H, s, C1-**H**), 8.20 (1H, dd, J_o = 8.1, J_m = 1.2, C7-**H**), 7.97 (1H, d, J = 9.0, C3-**H**), 7.44 (1H, t, J = 8.1, C8-**H**), 7.34 (dd, 1H, J_o = 8.1, J_m = 1.2, C9-**H**), 6.75 (1H, d, J = 9.0, C4-**H**), 4.14 (3H, s, 10-OC**H**$_3$), 3.48 (2H, qu, NHC**H**$_2$CH$_2$), 2.84 (2H, t, NHCH$_2$C**H**$_2$), 2.66 (4H, qu, N(C**H**$_2$CH$_3$)$_2$), 1.11 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

5-[[2-(Diethylamino)ethyl]amino]-7-hydroxy-10-methoxy-6*H*-imidazo[4,5,1-*de*]acridin-6-one (4o). ^{1}H NMR (free base) ($CDCl_3$): δ 13.48 (1H, s, ex, 7-O**H**), 9.08 (1H, s, C1-**H**), 8.82 (1H, t, N**H**-CH$_2$), 7.97 (1H, d, J = 8.8, C3-**H**), 7.31 (1H, t, J = 8.8, C9-**H**), 6.87 (1H, d, J = 8.8, C8-**H**), 6.78 (1H, d, J = 8.8, C4-**H**), 4.06 (3H, s, 10-OC**H**$_3$), 3.68 (2H, qu, NHC**H**$_2$CH$_2$), 2.96 (2H, t, NHCH$_2$C**H**$_2$), 2.80 (4H, qu, N(C**H**$_2$CH$_3$)$_2$), 1.12 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

5-[[2-(Diethylamino)ethyl]amino]-8,9-(methylenedioxy)-6*H*-imidazo[4,5,1-*de*]acridin-6-one (4r). A mixture of **3r** (2 g, 0.005 mol), nickel aluminum alloy (1.5 g), and 80% formic acid (10 mL) was refluxed with stirring for 20 h. The reaction mixture was evaporated under reduced pressure and worked up as described above for **4k**. ^{1}H NMR (free base) ($CDCl_3$): δ 9.06 (1H, ex, N**H**-CH$_2$), 8.44 (1H, s, C1-**H**), 7.98 (1H, d, J = 8.9, C3-**H**), 7.92 (1H, s, C10-**H**), 7.34 (1 H, s, C7-**H**), 6.77 (1H, d, J = 8.9, C4-**H**), 6.20 (2H, s, OC**H**$_2$O), 3.51 (2H, qu, NHC**H**$_2$-CH$_2$), 2.88 (2H, t, NHCH$_2$C**H**$_2$), 2.70 (4H, qu, N(C**H**$_2$CH$_3$)$_2$), 1.15 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

5-[[2-(Diethylamino)ethyl]amino]-10-hydroxy-6*H*-imidazo[4,5,1-*de*]acridin-6-one (4i). To a vigorously stirred solution of **4k** (free base, 1.2 g, 0.0027 mol) in 1,2-dichloroethane (50 mL) was added 1 M solution of BBr_3·Me_2S in dichloromethane (3 mL) slowly at room temperature. The mixture was refluxed with stirring for 100 h, and during this time an additional 6 mL of BBr_3 complex solution was added in two portions. The mixture was cooled to room temperature, the reaction was quenched with ethanol (5 mL), and the mixture stirred for an additional 30 min. The precipitated solid was filtered off, dissolved in hot water, and, after cooling, alkalized using 0.5 N NaOH. The unreacted substrate was separated by extraction with chloroform; the aqueous solution was acidified with diluted hydrochloric acid and neutralized with concentrated ammonia. The precipitated product was filtered, crystallized from methanol−a few drops of concentrated HCl−ether to give hydrochloride of **4i**. ^{1}H NMR (free base) (acetone-d_6): δ 11.25 (1H, s, ex, 10-O**H**), 9.20 (1H, s, C1-**H**), 9.10 (1H, t, ex, N**H**-CH$_2$), 8.01 (1H, d, J = 8.9, C3-**H**), 7.90 (1H, dd, J_o = 6.6, J_m = 2.8, C7-**H**), 7.42 (2H, m, C8-**H**, C9-**H**), 6.77 (1H, d, J = 8.9, C4-**H**), 3.45 (2H, qu, NHC**H**$_2$CH$_2$), 2.78 (2H, t, NHCH$_2$C**H**$_2$), 2.62 (4H, qu, N(C**H**$_2$CH$_3$)$_2$), 1.05 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

5-[[2-(Diethylamino)ethyl]amino]-7,10-dihydroxy-6*H*-imidazo[4,5,1-*de*]acridin-6-one (4j). ^{1}H NMR (free base) (DMSO-d_6): δ 13.48 (1H, s, ex, 7-O**H**), 10.65 (1H, s, ex, 10-O**H**), 9.08 (1H, s, C1-**H**), 8.82 (1H, t, N**H**-CH$_2$), 7.97 (1H, d, J = 8.8, C3-**H**), 7.31 (1H, d, J = 8.8, C9-**H**), 6.87 (1H, d, J = 8.8, C8-**H**), 6.78 (1H, d, J = 8.8, C4-**H**), 3.60 (2H, qu, NHC**H**$_2$CH$_2$), 2.60−2.80 (6H, m, C**H**$_2$N(C**H**$_2$CH$_3$)$_2$), 1.12 (6H, t, N(CH$_2$C**H**$_3$)$_2$).

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