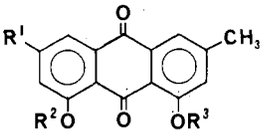
Novel Type of Potential Anticancer Agents Derived from Chrysophanol and Emodin. Some Structure–Activity Relationship Studies[1]

Sir:

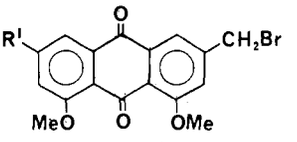
Certain antitumor intercalating agents, e.g., ellipticine,[2] amsacrines,[3] and the anthracycline antibiotics,[4] have been the subject of a variety of types of structural modification directed toward improving their therapeutic potential. Preliminary screening of a number of synthetic analogues of these drugs unfortunately shows that there is no straightforward structure–activity relationship between intercalating potency and anticancer activity. These results seem to suggest that though intercalation may be an important condition, it may not be a sufficient one for such drugs to exert anticancer activity. It may be necessary for these agents to bind tightly with DNA to disrupt the DNA function eventually. This hypothesis may be supported by a report on the mechanism of nonintercalating anticancer antibiotic CC1065[5] that it binds to the minor groove of DNA and slowly alkylates DNA with simultaneous opening of the cyclopropane ring of CC1065.

We linked the nitrogen mustard moiety to the methyl side chain of chrysophanol (**1a**) and emodin (**1b**)[6] (Figure 1), natural anthraquinones with little anticancer activity, at the methyl side chain in order to examine if such modified molecules might intercalate into and then alkylate DNA. Methylation of 1 with Me_2SO_4/Me_2CO in DMF[7] afforded the corresponding per-*O*-methylated 3-methylanthraquinones **2**, which were treated with 1,3-dibromo-5,5-dimethylhydantoin in CCl_4 in the presence of dibenzoyl peroxide[8] to give the monobromides **3** as the major products along with small amounts of the dibromides **4**, which were readily separated on a silica gel column. Treatment of **3** with diethylamine or bis(β-hydroxyethyl)amine afforded the corresponding alkylamino derivatives **5** and **6**. Chlorination of the β-hydroxyethylamines **6a**-I and **6b**-I with $SOCl_2$ in Et_2O gave 3-[bis(β-chloroethyl)amino]-1,8-di-*O*-methylchrysophanol (**7a**-I) and 6-[bis(β-chloroethyl)amino]-1,3,8-tri-*O*-methylemodin (7b-I), respectively. O-Demethylation of **7a,b**-I was achieved stepwise in HBr/AcOH: at room temperature, only one peri methyl group was removed, giving rise to the mixtures of 1-*O*-methyl and 8-*O*-methylanthraquinones **7a**-II and **7b**-II, whereas at reflux temperature both methoxy groups were hydrolyzed to give the corresponding 3-modified chrysophanol and emodin **7a**-III and **7b**-III. This stepwise process not only gave the 1,8-dihydroxy derivatives **7a,b**-III but also provided the 1- and 8-mono-*O*-methyl compounds, which are important for structure–activity relationship studies.

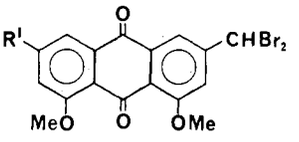


1a R¹, R², R³ = H
1b R¹ = OH, R², R³ = H

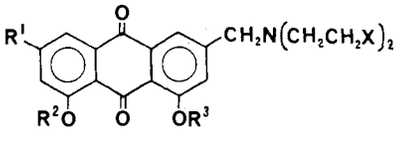
2a R¹ = H, R², R³ = Me
2b R¹ = OMe, R², R³ = Me



3a R¹ = H
3b R¹ = OMe



4a R¹ = H
4b R¹ = OMe



5a-I R¹ = H, R², R³ = Me, X = H
5a-II R¹ = H, R² = Me or H, R³ = H or Me, X = H
5a-III R¹, R², R³ = H, X = H

5b-I R¹ = OMe, R², R³ = Me, X = H
5b-III R¹ = OMe, R², R³ = H, X = H

6a-I R¹, R² = Me, R³ = H, X = OH
6a-II R¹ = H, R² = Me or H, R³ = H or Me, X = OH
6a-III R¹, R², R³ = H, X = OH

6b-I R¹ = OMe, R², R³ = Me, X = OH
6b-III R¹ = OMe, R², R³ = H, X = OH

7a-I R¹ = H, R², R³ = Me, X = Cl
7a-II R¹ = H, R² = Me or H, R³ = H or Me, X = Cl
7a-III R¹, R², R³ = H, X = Cl

7b-I R¹ = OMe, R², R³ = Me, X = Cl
7b-III R¹ = OMe, R², R³ = H, X = Cl

Figure 1.

It appears that the results of preliminary bioassay of **5**–**7** for their inhibitory activity against L1210 mouse leukemia cells in tissue culture[9] (Table I) are consistent with our

(1) This investigation was supported in part by funds from the National Cancer Institute, NIH, USDHHS, Grant No. CA-08748 and CA-18856.
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(3) Denny, W. A.; Cain, B. F.; Atwell, G. J.; Hantsch, C.; Panthananickal, A.; Leo, A. *J. Med. Chem.* **1982**, *25*, 276.
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(8) Cava, M. P.; Ahmed, Z.; Benfaremo, N.; Murphy, R. A.; O'Malley, G. J. *Tetrahedron* **1984**, *40*, 4767.
(9) Personal communication from Dr. J. Kapuscinski, who studied the interaction of these compounds with DNA spectrophotometrically. Full details of synthetic procedures and results of spectrometric, biological, and biochemical studies will be published together.

0022-2623/88/1831-0283$01.50/0

Table I. Antileukemic Activity of Derivatives of Chrysophanol and Emodin Bearing Alkylating Potential

compd	X	R^1	R^2	R^3	L1210: ID_{50}, μM
5a-I	H	H	Me	Me	128.3
5a-II	H	H	H (or Me)	Me (or H)	11.9
5a-III	H	H	H	H	2.75
6a-I	OH	H	Me	Me	>50
6a-II	OH	H	H (or Me)	Me (or H)	23.4
6a-III	OH	H	H	H	5.92
7a-I	Cl	H	Me	Me	12.5
7a-II	Cl	H	H (or Me)	Me (or H)	1.40
7a-III	Cl	H	H	H	0.13
5b-I	H	OMe	Me	Me	6.91
5b-III	H	OMe	H	H	1.16
6b-I	OH	OMe	Me	Me	>27.6
6b-III	OH	OMe	H	H	13.7
7b-I	Cl	OMe	Me	Me	2.66
7b-II	Cl	OMe	H (or Me)	Me (or H)	1.75
7b-III	Cl	OMe	H	H	0.023
chrysophanol (1a)					>393
emodin (1b)					13.3

hypothesis. The presence of both the potent intercalating and alkylating functionalities in the molecule (e.g., **7a,b**-III) maximizes the activity. Lack of one of these functionalities (e.g., **7a,b**-I, which lack potent intercalating capability due to the presence of two bulky methoxy groups, or **1a,b**, which lack the alkylating potential) greatly reduced the cytotoxicity. The activity of 1- or 8-mono-*O*-methylanthraquinones **7a,b**-II falls between that of the 1,8-dihydroxy (**7**-III) and 1,8-dimethoxy (**7**-I) analogues. It is also interesting to note that the diethylamino derivatives **5a,b**-III exhibited significant activity whereas the 1,8-di-O-methylated analogues **5a,b**-I showed little cytotoxicity. Due to the presence of basic nitrogen in the molecule, **5a,b**-III can interact more strongly with DNA than the corresponding unmodified natural products, **1a,b**.[9] The 1,8-di-O-methylated derivatives did not interact with DNA most probably due to the presence of two bulky methoxy groups.[9] A very preliminary in vivo test using BDF mice with transplanted L1210/0 leukemia and using a procedure previously described[10] showed that **7a**-III produced an increase in life span of 29% at 25 mg/kg per day × 5.

Studies on the synthesis and biological activities of potential intercalators with alkylating capability are currently under way, and a report on further details will be forthcoming.

(10) A modification of the technique of Fischer (*Ann. N.Y. Acad. Sci.* **1958**, *76*, 673) was employed for cell culture studies. Burchenal, J. H.; Chou, T.-C.; Lokys, L.; Smith, R. S.; Watanabe, K. A.; Su, T.-L.; Fox, J. J. *Cancer Res.* **1982**, *42*, 2598.

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Received September 14, 1987

Articles

Renin Inhibitors. Design and Synthesis of a New Class of Conformationally Restricted Analogues of Angiotensinogen

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Molecular modeling methods have been used to design a novel series of conformationally constrained cyclic peptide inhibitors of human renin. Three goals were defined: enhanced inhibitory potency, high specificity for renin, and increased metabolic stability. Three cyclic compounds were synthesized with ring sizes 10, 12, and 14, based upon a linear hexapeptide inhibitor with a reduced amide replacing the scissile bond at the active site. When tested, the 14-membered-ring compound was as potent an inhibitor of human renin as the parent while the 12-membered-ring compound was 6-fold more potent than the parent against mouse renin. However, the 10-membered-ring compound was inactive against both renins. The lack of potency of the 10-membered compound was explained by using NMR and molecular modeling techniques. It forms another conformation in solution that is inconsistent with binding at the active site. The cyclic compounds did not inhibit either pepsin or cathepsin D significantly. The cyclic modification rendered these inhibitors significantly resistant to cleavage by chymotrypsin and thus prevented loss of activity by this enzyme. Thus, the goals of enhanced inhibitory potency, high specificity, and metabolic stability were achieved in the series of compounds.

The aspartic proteinase renin selectively cleaves the protein substrate angiotensinogen to produce a decapeptide angiotensin I, which is converted by the angiotensin converting enzyme to the potent pressor peptide angiotensin II. Inhibitors of angiotensin converting enzyme have proved to be important modulators of blood pressure.[1,2] Renin inhibitors should exhibit similar antihy-

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0022-2623/88/1831-0284$01.50/0