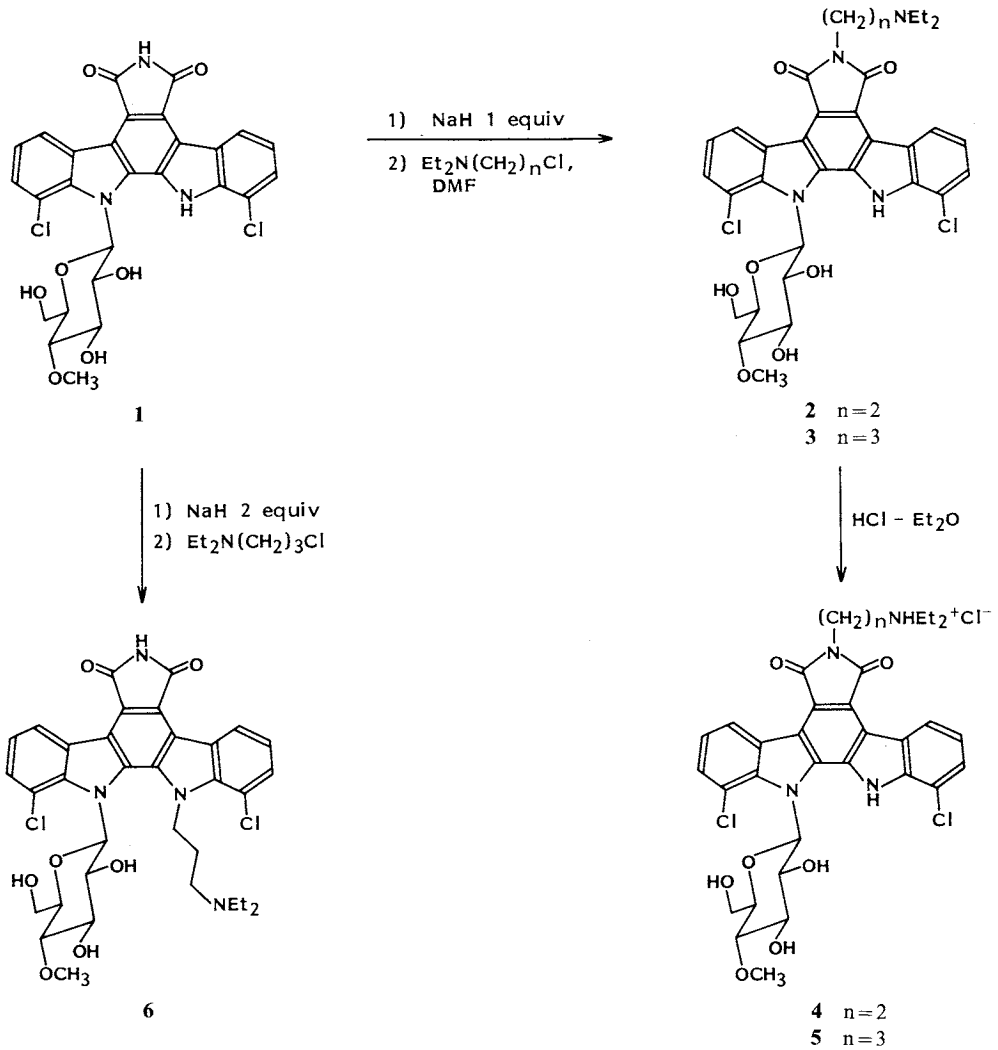
WATER SOLUBLE DERIVATIVES OF REBECCAMYCIN

Sir:

Recently we reported isolation and synthesis of rebeccamycin (**1**) as a new chemotype of anticancer agent[1,2]. More recently, it was reported that structurally related staurosporin[3] and K-252a[4] are potent inhibitors of protein kinase C. Other related compounds include a series of disaccharide derivatives designated AT2433 which are active against P388 leukemia[5]. Although rebeccamycin exhibits significant *in vivo* antitumor activity against P388 leukemia and B16 melanoma, its limited solubility in aqueous media posed some problems in further evaluation[6]. In order to find more water-soluble analogues we have carried out structural modifications of rebeccamycin and in this communication we report some of the promising analogues.

To impart water solubility, effort was made to introduce an aminoalkyl group to the molecule. This substructure is often incorporated into a parent drug to improve physico-chemical properties of the drug[7]. The site of attachment was controlled by employing conditions for alkylating an imide nitrogen. Thus, when rebeccamycin was treated with 1.1 equiv of NaH in DMF at room temperature followed by addition of 2-diethylaminoethyl chloride, N-6 alkylated product was obtained in 66% yield. (For identifying the various positions the Chemical Abstruct numbering system is used. The

Fig. 1 Synthesis of aminoalkyl derivatives of rebeccamycin.



Chemical Abstract nomenclature for rebeccamycin is 1,11-dichloro-12,13-dihydro-12-(4-*O*-methyl-β-D-glucopyranosyl)-5*H*-indolo[2,3-*a*]pyrrolo[3,4-*c*]carbazole-5,7(6*H*)-dione.) Similarly, when 3-diethylaminopropyl chloride was used, compound **3** was obtained in 28% yield. The corresponding HCl salts (**4** and **5**) were obtained in 90% yield by addition of 1 equiv of ethanolic HCl solution to a THF solution of **2** or **3** at 0°C. When rebeccamycin was treated with slightly more than 2 equiv of NaH in DMF and then with 1 equiv of 3-diethylaminopropyl chloride, N-13 alkylated product (**6**) was obtained in 34% yield. Apparently, the dianion was formed in this case and the more reactive indolyl anion reacted with the aminoalkyl chloride to give the observed product.

These salts indeed possess increased water-solubility. For example, compound **4** is soluble at a rate of 4 mg per ml of water whereas rebeccamycin is essentially insoluble (<1 μg/ml). The antitumor activity of the free bases and salts against P388 leukemia is shown in Table 1. The activity of free bases **2** and **3** indicates that incorporation of an aminoalkyl group at N-6 did not reduce the antileukemic activity since rebeccamycin under the same treatment schedule generally gives T/C values 132~155% at 16~512 mg/kg/injection[6]. Furthermore, their salts can achieve practically the same T/C values at lower doses. More significantly, while it was difficult to show activity of rebeccamycin itself by an iv route against iv implanted P388 leukemia (unpublished data), compound **4** showed activity reaching a T/C of 150% at 96 mg/kg (Table 1). This hydrochloride salt was active also against ip implanted B16 melanoma yielding a T/C of 191% at 18 mg/kg/injection. Rebeccamycin under the same treatment schedule gave a T/C 175% at 256 mg/kg/injection[6]. This corresponds to an increase of potency approximately 14 times. These new derivatives of rebeccamycin, therefore, appear to possess the desired solubility, and antitumor activities. They and other related salts are currently under further evaluation.

Table 1. Antitumor activity of rebeccamycin analogues against P388 lymphocytic leukemia[a].

Compound	Dose (mg/kg)	Route/schedule	T/C (%)
2	50	ip, 1×[b]	167
	25		133
3	50	ip, 1×	152
4	24	ip, 1×	133
	18		139
	96	iv, 1×[c]	150
	64		133
5	8	ip, 1×	139
	6		144
6	50	ip, 1×	164
	25		155
	12.5		141

[a] P388 leukemia, 10^6 cells, were implanted ip on day 0 into CDF_1 mice.
[b] Administered ip on day 1.
[c] P388 leukemia, 10^6 cells, were implanted iv into CDF_1 mice and the drug was administered iv on day 1.

Table 2. Antitumor activity of compound **4** against B16 melanoma[a].

Compound	Dose (mg/kg)	Route/schedule	T/C (%)
4	18	ip, qd 1-9	191
	12		183
	6		183

[a] B16 melanoma, 0.5 ml of a 10% tumor brei, was inoculated ip on day 0 into BDF_1 mice.
[b] Administered ip daily for 9 days starting day 1.

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