INHIBITORY ACTIVITY OF XANTHONE DERIVATIVES ISOLATED FROM SOME GUTTIFERAEOUS PLANTS AGAINST DNA TOPOISOMERASES I AND II

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A xanthone derivative, subelliptenone F, and the related compounds showed an intensive inhibitory effect against topoisomerases I and II in *in vitro* experiments. These xanthones are prospective lead compounds for anticancer drugs.

KEY WORDS topoisomerase I; topoisomerase II; Guttiferae; xanthone; subelliptenone F; benzophenone

Two major topoisomerases (Topo), types I and II, play a critical cellular role in alterations of the topological state of DNA such as supercoiling, knotting, and catenation. Topo I becomes covalently attached to the DNA with reversible breakage of one DNA strand, thereby effecting DNA relaxation. Topo II, which breaks both strands of duplex DNA, can relax supercoiled DNA and resolve knotted or catenated DNA rings.[1] The prime importance of these enzymes makes them critical targets for the action of a wide variety of anticancer drugs. Camptothecin and etoposide analogs have been applied clinically, but their severe side effects remain a serious problem. The development of a new class of inhibitors of Topo is thus awaited. In the development of new drugs, natural products obtained from plants are sometimes useful directly or serve as starting material for semisynthesized active agents. Furthermore, naturally occurring compounds can supply suitable leads for the subsequent design of structurally related molecules that are more active or less toxic. In our search for physiologically active principles in Guttiferaeous plants, we report here on the inhibitory effects of xanthone and benzophenone derivatives against Topo I and II in *in vitro* experiments.

Xanthones (**1**-**23**) isolated from *Garcinia dioica*[2] (**1** and **2**), *G. mangostana*[3] (**3**-**5**), *G. subelliptica*[4-6] (**6**-**12**), *Calophyllum inophyllum*[7-10] (**13**-**22**), and *Mammea acuminata*[11] (**23**) and two benzophenones (**24** and **25**) from *G. subelliptica* and *G. purpurea*[12] were examined in the present screening test and their structures are shown in Chart 1. The inhibitory activity of **1**-**25** against Topo types I and II was estimated according to the method of Tsutsui *et al.*[13] The first screening was performed at concentrations of 10, 100, and 500 μg/ml against Topo I and II. The compounds that were effective at 100 μg/ml were further subjected to determination of the inhibitory effect at 1, 10, 25, 50, and 100 μg/ml. The IC_{50} values observed at less than 300 μg/ml are listed in Table 1. In the inhibitory screening for Topo I, subelliptenone F (**10**) showed the most potent activity and was effective at 30 μg/ml. The IC_{50} value of the control, camptothecin, was 0.87 μg/ml.[14] On the other hand, in the screening for Topo II, an intensive inhibitory effect of subelliptenone F (**10**) was also observed at less than 1 μg/ml concentration. Xanthone **5** was active at 5 μg/ml. The IC_{50} values of **1**, **3**, **11**, **24**, and **25** ranged from 30 to 55 μg/ml. The inhibitory activities of these seven compounds

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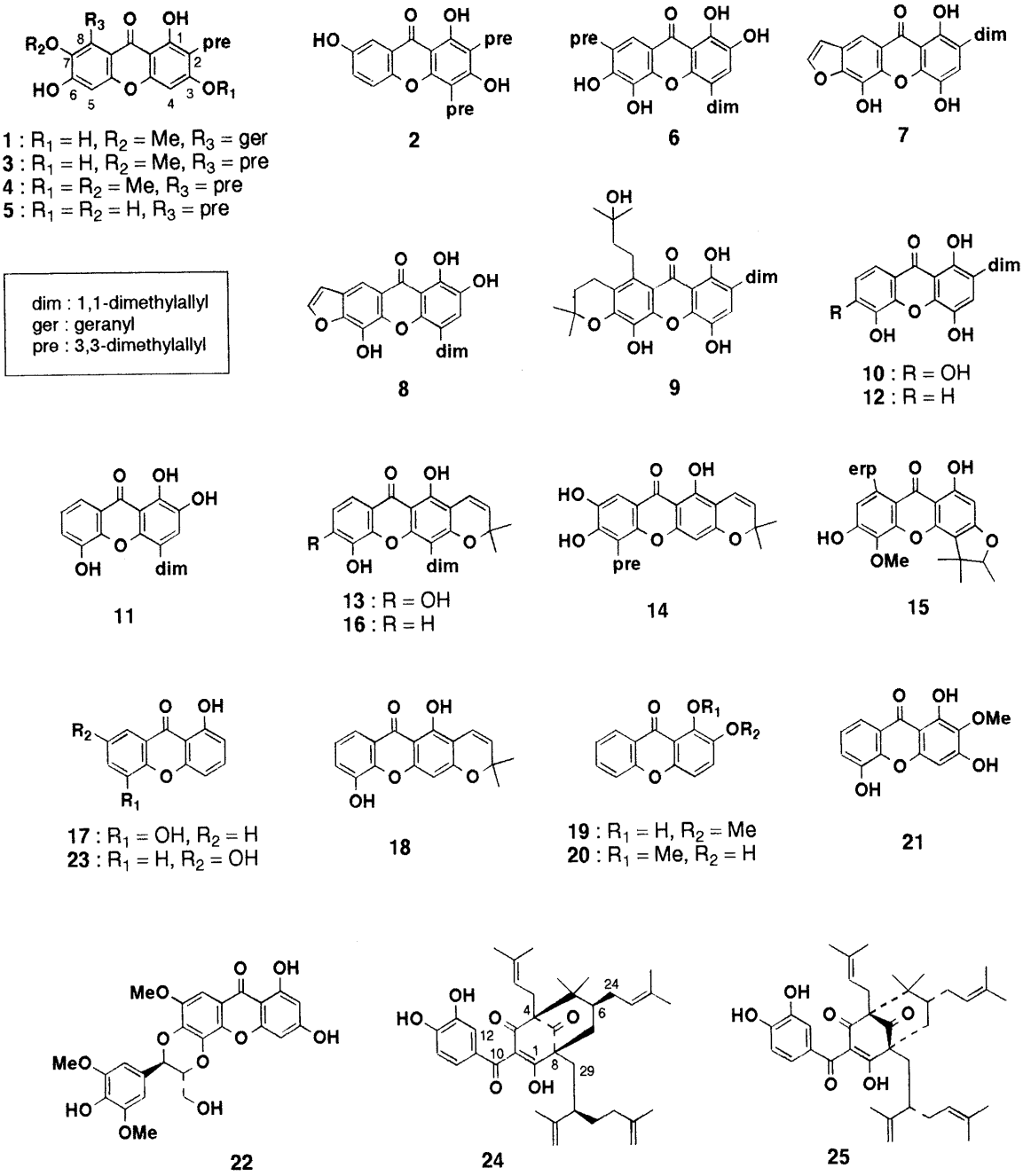

Chart 1. Structures of **1-25**

Table 1. IC_{50} Value (μg/ml) for Compounds **1-25** and Etoposide against Topoisomerases I and II

Compound	1	3	4	5	7	8	9	10	11	12	22	24	25	Etoposide
IC_{50} (Topo I)	60	55	150	39	400	290	400	30	300	290	300	33	43	-
IC_{50} (Topo II)	39	38	80	5	110	330	210	<1	26	>500	200	40	55	70

were stronger than that of the control, etoposide (70 μg/ml). No enzymatic selectivity of the xanthones was observed except for **11**, which inhibited Topo II more intensively than Topo I.

In Topo II inhibitory screening, xanthones **7**, **9**, **10**, and **12** have a common identical partial structure [1,4,5-trihydroxy-2-(1,1-dimethylallyl)xanthone] (Chart 1). The structural differences were substitutional patterns at C-6 and C-8. In the case of subelliptenone F (**10**), a hydroxyl group is located at C-6, but no substitutional groups are present at C-7 and C-8. The absence of a hydroxyl group at C-6 in **10** such as **12** caused remarkable diminution of the inhibitory activity. Although **7** and **9** have an oxygen function at C-6, the substitution of the hydrogen atom with an alkyl chain resulted in the loss of activity. On the basis of these findings, the presence of a free hydroxyl group at C-6 in a 1,4,5-trihydroxy-2-(1,1-dimethylallyl)xanthone appears to be essential to the inhibitory activity. Among xanthones **1**, **3**, **4**, **5**, and **14**, which have the same oxidative pattern (1,3,6,7-tetraoxygenated xanthone) (Chart 1), only γ-mangostin (**5**) showed significantly strong activity. The inhibitory effect was generally reduced with the decrease in the number of free hydroxyl groups in a xanthone skeleton.

To reduce the toxicity and side effects of anticancer agents, the development of a new class of compounds is urgently needed. In the present study, a potent inhibitory effect against Topo I and II in *in vitro* experiments was confirmed for the first time in phenolic compounds, particularly in xanthone derivatives.

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