The Hypotensive Effect and Antifungal Activity of 3,3′-Dihydroxy-α,β-diethyldiphenylethane

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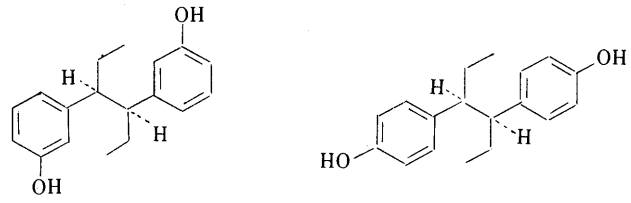
3,3′-Dihydroxy-α,β-diethyldiphenylethane (I), an isomer of hexestrol (II), was found to have a strong hypotensive effect on rats and antifungal activity against some pathogenic fungi. The hypotensive effect of I (−100.0±21.0 mmHg, 10 mg/kg, i.v.) was stronger than that of II (−40.0±2.60 mmHg, i.v.). Unlike II and other oxystilbene-related compounds already tested, I showed prolonged hypotensive action at the dose of 10 mg/kg, and the blood pressure was not restored to the original level within 20 min. I also showed antifungal activity against all fungi tested (minimal inhibitory concentration: 10—120 μg/ml). It should be emphasized that by shifting the phenolic hydroxyl group to *m*-position, the biological activities were greatly increased.

Keywords 3,3′-dihydroxy-α,β-diethyldiphenylethane; 3,3′-dihydroxy-α,β-diethylstilbene; hexestrol; oxystilbene-related compound; diethylstilbestrol; hypotensive effect; antifungal activity; phenolic hydroxyl group

So far, hexestrol (II, Chart 1),[1,2] diethylstilbestrol[2-4] and 3,3′-dihydroxy-α,β-diethylstilbene[4,5] have already been reported to show the following biological activities by the authors: antifungal activity, phytogrowth-inhibitory activity, coronary vasodilator action on isolated guinea-pig heart and hypotensive effect on rats. Among these compounds, 3,3′-dihydroxy-α,β-diethylstilbene has no hormonal side effect.[6] Recently, we reported that 3,3′-dihydroxy-α,β-diethyldiphenylethane (I, Chart 1), an isomer of II, had coronary vasodilator action,[7] antifungal activity on plant-pathogenic fungi and phytogrowth-inhibitory activity.[8] In particular, the inhibitory activity of I on plant growth was as strong as that of sodium 2,4-dichlorophenoxyacetate used as a standard. However, there has been no report on the hypotensive effect of I, or on the antifungal activity of I against pathogenic fungi except for plant-pathogenic fungi.

In this report, the antifungal activity against nine kinds of pathogenic fungi and the hypotensive effect on rats of I are described in comparison with those of II.[1,2]



3,3′-dihydroxy-α,β-diethyldiphenylethane (I) hexestrol (II)

Chart 1

TABLE I. Effect of I on Blood Pressure in Rats

Compound	Dose (mg/kg)	Mean arterial blood pressure (mmHg)
I	2.5	−28.3±12.30
	5.0	−64.2± 7.60
	10.0	−100.0±21.00
II[1]	10.0	−40.0± 2.60

Each value represents the mean±S.D. of 3 rats. Route: Intravenous injection.

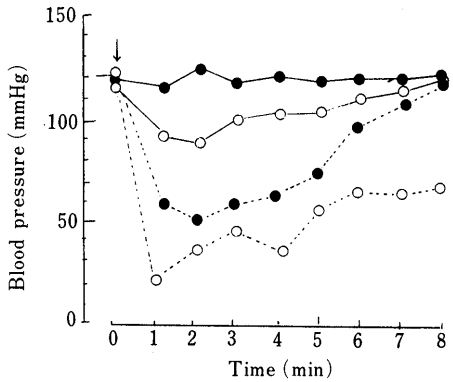


Fig. 1. Effect of I on Blood Pressure in Rats

○---○, 10 mg/kg; ●---●, 5 mg/kg; ○—○, 2.5 mg/kg; ●—●, 5% acacia.
Each value represents the mean of 3 rats. Body weight: 310—385 g. Arrow: injection of 2.5, 5.0 or 10.0 mg/kg of I.

Materials and Methods

Chemical 3,3′-Dihydroxy-α,β-diethyldiphenylethane (I) was synthesized by the reduction of 3,3′-dihydroxy-α,β-diethylstibene[9] (I: mp 100—101 °C (dec.)).

Animals The animals used for the experiments on hypotensive effect were male Wistar strain rats weighing 310—385 g.

Fungi Nine kinds of fungi listed in Table II were used for the antifungal activity test.

Biological Activity Tests 1) Measurement of Blood Pressure: Systemic blood pressure was measured with a pressure transducer (Nihon Kohden Kogyo Co., Ltd., MPV-0.5, PM-85) following cannulation of the carotid artery in rats under anesthesia with sodium pentobarbital (40 mg/kg, i.p.). Compound I was suspended in 5% acacia and administered *via* the femoral vein. It was shown that 5% acacia had no effect on blood pressure. The blood pressure prior to drug administration was 129.0±14.6 mmHg ($n=9$).

2) Antifungal Activity: Antifungal activity test was carried out by the agar dilution method. The media used were as follows: potato sucrose agar, Sabouraud glucose agar or potato dextrose agar. The test fungi were applied to media containing various concentrations of I. The plates were incubated at 27 °C for 7 d and the growth was observed with the naked eye.

Results and Discussion

Effect of 3,3′-Dihydroxy-α,β-diethyldiphenylethane (I) on Blood Pressure in Rats The effect of I on blood pressure in rats was investigated and compared with that of hexestrol II,[1] an isomer of I. The results are summarized in Table I. Compound I showed strong hypotensive activity. The activity of I (−100.0±21.0 mmHg, 10 mg/kg, i.v.) was much stronger than that of II (−40.0±2.60 mmHg, 10 mg/kg, i.v.).[1] As shown in Fig. 1, at the dose of 2.5 and

5.0 mg/kg, the hypotensive actions of I were transient and the blood pressure recovered to the original level within 8 min. The pattern of the time course of I was similar to those of II,[1] diethylstilbestrol,[4] 3,3′-dihydroxy-α,β-diethylstilbene,[4] 3,3′,4,5′-tetrahydroxystilbene,[10] 3,3′,4,5′-tetrahydroxybibenzyl[10] and 3,4-*O*-isopropylidene-3,3′,4,5′-tetrahydroxystilbene.[11] On the other hand, unlike other oxystilbene-related compounds, I showed prolonged hypotensive action at the dose of 10 mg/kg and the blood pressure did not recover to the original level within 20 min. This prolonged hypotensive effect of I is noteworthy. As regards the mechanism of hypotensive action of oxystilbene-related compounds, Saruta *et al.*[12] reported that diethylstilbestrol acts directly on the vascular bed and attenuates vascular response to norepinephrine. However, the precise mechanisms of the hypotensive action of oxystilbene-related compounds have not yet been clarified. Considering that, as shown in our recent report,[7] I had strong relaxing action on the contraction of guinea-pig mesenteric artery due to KCl and norepinephrine at the low concentration of 10^{-5}—10^{-6} M, the mechanism of the hypotensive action is of considerable interest. Further studies on the hypotensive effect of many oxystilbene-related compounds, together with the mechanisms involved, are in progress.

Antifungal Activity of 3,3′-Dihydroxy-α,β-diethyldiphenylethane (I) As mentioned above, I has already been reported to show antifungal activity against plant-pathogenic fungi. However, no work has been done on the antifungal activity against other pathogenic fungi. Thus, the antifungal activity of I was examined with nine kinds of pathogenic fungi and compared with that of hexestrol (II),[8] an isomer of I. As shown in Table II, unlike II, compound I showed antifungal activity toward all fungi tested (minimal inhibitory concentration (MIC: 10—120 μg/ml). The results indicate that two phenolic hydroxyl groups attached to the *m*-position are favorable for the antifungal activity of diethylstilbestrol-related compounds. From the standpoint of the structure–activity relationship, such a broad antifungal spectrum of I is of considerable interest in comparison with those of II and diethylstilbestrol.[2] We showed in our previous paper that I had antifungal activity against plant-pathogenic fungi.[8] Furthermore, 3,3′,4,5′-tetrahydroxystilbene[13] and 3,3′,4,5′-tetrahydroxybibenzyl,[13] a constituent of *Cassia garrettiana*, and its derivative, 3,4-*O*-isopropylidene-3,3′,4,5′-tetrahydroxystilbene,[11,14] showed the same activities.

TABLE II. Antifungal Activity of I

Fungus	Medium	Antifungal activity (MIC: μg/ml)	
		I	II
Trichophyton rubrum IFO-5811	A	20.0	14.0
Trichophyton mentagrophytes IFO-5467	A	30.0	16.0
Penicillium notatum IFO-4046	B	10.0	1000.0
Penicillium thomii IFO-7002	B	20.0	1000.0
Penicillium citrinum IFO-6026	B	50.0	1000.0
Aspergillus niger IFO-4414	B	20.0	1000.0
Trichoderma longibrachiatum IFO-4847	B	10.0	1000.0
Mucor racemosus IFO-4581	B	40.0	1000.0
Candida albicans IAM-4966	C	120.0	1000.0

Culture conditions: 27 °C, 7 d. Media: A, potato dextrose agar; B, potato sucrose agar; C, Sabouraud glucose agar. Method: agar dilution method.

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References and Notes

1) Y. Inamori, T. Kobayashi, M. Ogawa and H. Tsujibo, *Chem. Pharm. Bull.*, **36**, 815 (1988).
2) Y. Inamori, M. Kubo, M. Ogawa, K. Moriwaki, H. Tsujibo, K. Baba and M. Kozawa, *Chem. Pharm. Bull.*, **33**, 4478 (1985).
3) Y. Inamori, M. Kubo, M. Ogawa, K. Moriwaki, H. Tsujibo, K. Baba and M. Kozawa, *Chem. Pharm. Bull.*, **33**, 420 (1985).
4) Y. Inamori, M. Kubo, M. Ogawa, H. Tsujibo, Y. Miki and S. Takemura, *Chem. Pharm. Bull.*, **35**, 3502 (1987).
5) Y. Inamori, M. Kubo, H. Tsujibo, M. Ogawa, Y. Saito, Y. Miki and S. Takemura, *Chem. Pharm. Bull.*, **35**, 887 (1987).
6) D. Adam and H. Schonenberger, *Arzneim.-Forsch.*, **16**, 738 (1966).
7) Y. Inamori, H. Tsujibo, H. Hachiken, Y. Miki and S. Takemura, *Chem. Pharm. Bull.*, **36**, 1252 (1988).
8) Y. Inamori, H. Tsujibo, M. Ogawa, Y. Miki and S. Takemura, *Chem. Pharm. Bull.*, **36**, 3173 (1988).
9) E. C. Dodds, L. Goldberg, W. Lawson and R. Robinson, *Nature* (London), **141**, 247 (1938).
10) Y. Inamori, M. Kubo, Y. Kato, M. Yasuda, K. Baba and M. Kozawa, *Yakugaku Zasshi*, **104**, 819 (1984).
11) Y. Inamori, M. Kubo, Y. Kato, M. Yasuda, H. Tsujibo, K. Baba and M. Kozawa, *Chem. Pharm. Bull.*, **33**, 2904 (1985).
12) K. Kondo, T. Okuno, T. Eguchi, T. Yasui, H. Suzuki, S. Nagahama and T. Saruta, *Endocrinol. Jpn.*, **27**, 307 (1980).
13) Y. Inamori, Y. Kato, M. Kubo, M. Yasuda, K. Baba and M. Kozawa, *Chem. Pharm. Bull.*, **32**, 213 (1984).
14) Y. Inamori, M. Kubo, Y. Kato, M. Yasuda, K. Baba and M. Kozawa, *Chem. Pharm. Bull.*, **32**, 801 (1984).