Full Paper

Synthesis, Antiplatelet and Vasorelaxing Activities of Xanthone Derivatives

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A series of ω-aminoalkoxylxanthones was synthesized and tested *in vitro* for their ability to inhibit platelet aggregation and cause vasorelaxing action. Compounds **4**, **5**, **12**, **17**, and **18** showed significant antiplatelet effects on thrombin-, arachidonic acid (AA)-, collagen-, and platelet activating factor (PAF)-induced washed rabbit platelet aggregation and exhibited inhibition of primary and secondary aggregation induced by adenosine-5'-diphosphate (ADP) in human platelet-rich-plasma (PRP). Compounds **4**, **17**, and **18** revealed vasorelaxing activities in rat thoracic aorta. We concluded that these compounds may be developed as new antithrombotic agents.

Keywords: Antiplatelet effect / Vasorelaxing activity / Xanthones derivaties

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Introduction

Xanthonoids isolated from Gentianaceous plants have been shown to be potent inhibitors of platelet aggregation and vasoconstriction [1, 2]. Synthetic xanthone derivatives have shown antiplatelet effect, inhibiting effects on cyclooxygenase, vasorelaxing effect and reduced blood pressure, heart rate, and attenuated isoprenaline-induced tachycardia in rats [3–6]. For the study of structure-activity relationship of various xanthone derivatives, characterizing their mode of action and their design as antithrombotic or antihypertensive agents, we synthesized further ω-aminoalkoxylxanthones.

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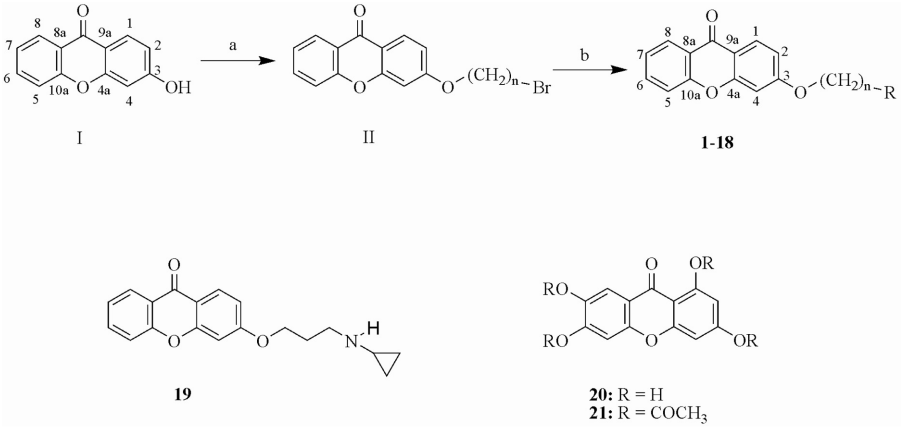
Abbreviations: arachidonic acid (AA); platelet activating factor (PAF)

Chemistry

Compounds **1**–**18** were synthesized (Scheme 1) by the method described in the previous reports [4, 7, 8]. Briefly, compounds **1**–**18** were obtained by the reaction of potassium salts of 3-hydroxyxanthone with 1,ω-dibromoalkane in *tert*-butanol and then aminated with appropriate amines to give the final product. The synthetic compounds were characterized by spectroscopic methods and the spectroscopic data were compared with the data reported in the literature [4, 7, 8].

Biological results and discussion

The antiplatelet effects of **1**–**18** were studied in the aggregation of washed rabbit platelets induced by thrombin, AA, collagen, and PAF. As shown in Table 1, compounds **3**–**6** and **12**–**18** (each at 300 μM) showed potent antiplatelet effects on the aggregation induced by the four agonists used in Table 1. Compound **2** (300 μM) showed potent antiplatelet effects on aggregation induced by AA,



Reagents and conditions: (a) aqueous 20% KOH, *tert*-BuOH, 1,ω-dibromoalkane, reflux; (b) THF or *tert*-BuOH, various amines, aqueous NaOH, stirring, 70°C.

Scheme 1. Synthesis of the compounds **1**–**18**.

Table 1. Effect of xanthone derivatives on the platelet aggregation induced by thrombin, AA, collagen, and PAF.

Compound	Platelet aggregation (%)			
	Thrombin	AA	Collagen	PAF
DMSO (control)	94.8 ± 1.0	92.1 ± 3.1	92.1 ± 0.8	93.8 ± 1.6
1	96.7 ± 3.0	46.8 ± 17.3[a)]	22.9 ± 15.0[c)]	72.1 ± 3.4[c)]
2	91.3 ± 3.0	20.4 ± 7.4[c)]	0.0 ± 0.0[c)]	29.0 ± 4.2[c)]
3	27.3 ± 8.0[c)]	15.3 ± 7.7[c)]	0.0 ± 0.0[c)]	22.7 ± 4.1[c)]
4	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]
5	6.8 ± 5.9[c)]	18.7 ± 7.9[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]
6	10.0 ± 5.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]
7	89.8 ± 2.6	31.1 ± 15.8[b)]	7.9 ± 6.8[c)]	37.7 ± 8.3[c)]
8	95.9 ± 0.6	84.4 ± 6.0	85.7 ± 1.9	89.6 ± 1.8
9	94.6 ± 0.6	87.8 ± 2.4	83.6 ± 3.7	89.9 ± 1.7
10	84.7 ± 0.9	88.2 ± 1.4	81.5 ± 1.8[b)]	87.3 ± 2.0
11	85.7 ± 3.0[a)]	68.0 ± 6.9[c)]	9.2 ± 5.5[c)]	10.4 ± 6.1[c)]
12	34.2 ± 8.5[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	1.9 ± 1.6[c)]
13	27.3 ± 8.0[c)]	15.3 ± 7.7[c)]	0.0 ± 0.0[c)]	22.7 ± 4.1[c)]
14	13.6 ± 5.9[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]
15	28.2 ± 5.8[c)]	0.0 ± 0.0[c)]	12.7 ± 7.2[c)]	0.0 ± 0.0[c)]
16	9.5 ± 4.8[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]
17	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]	0.0 ± 0.0[c)]
18	13.7 ± 8.5[c)]	4.6 ± 4.0[c)]	11.7 ± 5.9[c)]	2.0 ± 1.8[c)]
Acetyl salicylic acid	91.9 ± 2.5	0.0 ± 0.0[c)]	85.4 ± 3.9	90.5 ± 1.2

Platelets were preincubated with **1**–**18** (each at 300 μM), aspirin (50 μM), or DMSO (0.5%, control) at 37°C for 3 min, then thrombin (0.1 units/mL), AA (10 μM), collagen (10 μg/mL), or PAF (2 ng/mL) was added. Percentages of aggregation are presented as means ± S.E.M. (n = 3–7).

[a)] $P < 0.05$.

[b)] $P < 0.01$.

[c)] $P < 0.001$ as compared with the respective control value.

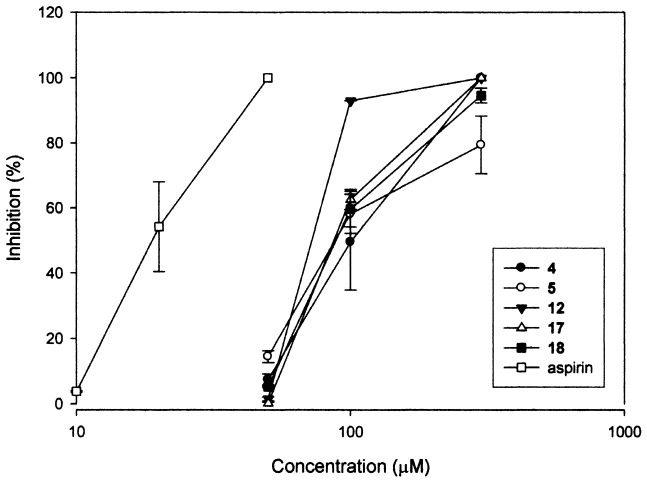


Figure 1. Concentration-dependent inhibitory effect of **4**, **5**, **12**, **17**, **18**, and aspirin on platelet aggregation induced by AA. Washed rabbit platelets were incubated with various concentrations of **4**, **5**, **12**, **17**, **18**, and aspirin or dimethylsulfoxide (0.5%) at 37°C for 3 min, then AA (100 mM) was added to trigger the aggregation. Percent inhibitions are presented as means ± S.E.M. (n = 3–6).

collagen, and PAF. Among them, compounds **4**, **5**, **12**, **17**, and **18** were the most potent inhibitors with minimal effective concentrations around 50 μM and maximal effective concentrations at 300 μM.

As shown in Figs. 1 and 2, the antiplatelet effects of **4**, **5**, **12**, **17**, and **18** on the aggregation induced by AA and **4**, **12**, and **18** on the aggregation induced by PAF were concentration-dependent. In comparison with the data previously reported for 3-[3-(cyclopropylamino)propoxy]xanthone **19**, norathyriol **20**, and norathyriol tetraacetate **21** (Scheme 1) [1, 8], compounds **4**, **5**, **12**, **17**, and **18** had less potent antiplatelet effects than **19** and **21**, when AA was used as the aggregation agonist. This indicated that the increasing carbon number of oxyalkyl side chain or substitution with various amino groups in the oxyalkyla-

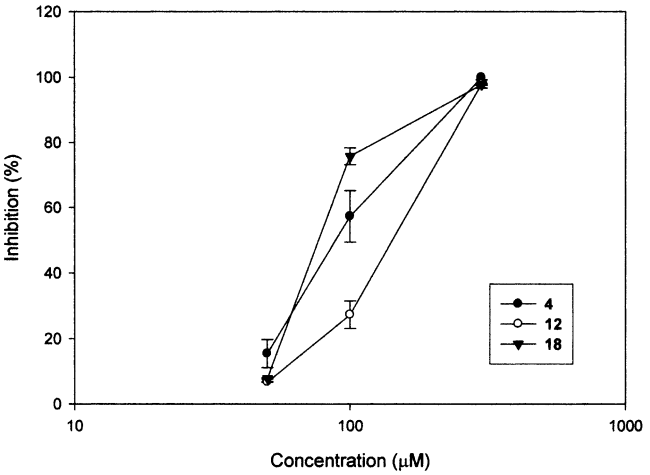


Figure 2. Concentration-dependent inhibitory effect of **4**, **12**, and **18** on platelet aggregation induced by PAF.
Washed rabbit platelets were incubated with various concentrations of **4**, **12**, and **18** or dimethylsulfoxide (0.5%) at 37°C for 3 min and then PAF (2 ng/mL) was added to trigger the aggregation. Percent inhibitions are presented as means ± S.E.M. (n = 3–6).

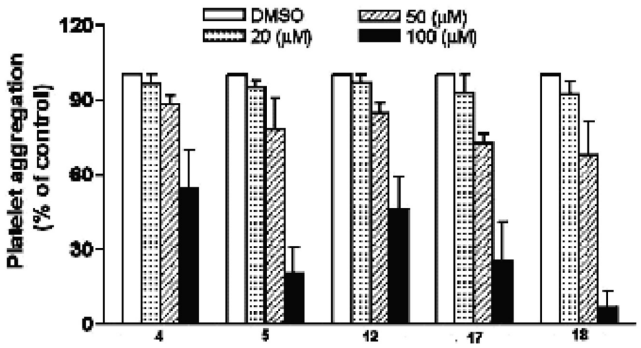


Figure 3. Concentration-dependent inhibitory effect of **4**, **5**, **12**, **17**, and **18** on platelet aggregation induced by ADP in human PrP.
Human PrP was incubated with various concentrations of **4**, **5**, **12**, **17**, and **18** or dimethylsulfoxide (0.5%) for 3 min and then ADP (20 μM) was added to trigger aggregation. Data are presented as means ± S.E.M. (n = 3–6).

mino side chain of **19** did not enhance the antiplatelet effects when AA is used as the aggregation agonist.

The antiplatelet effects of selective compounds **4**, **5**, **12**, **17**, and **18** were also studied on the aggregation of human PrP induced by ADP. As shown in Fig. 3, all these compounds (each at 100 μM) showed a potent antiplatelet effect on ADP-induced aggregation. In human PrP, these compounds prevented secondary aggregation and suppressed the primary aggregation at higher concentrations induced by ADP (for example **5** in Fig. 4). We conclude that their mechanism of action are chiefly due to the inhibition of thromboxane formation and interference in the ADP-receptor interaction [9, 10, 11]. Compounds **4**, **12**, and **18** significantly inhibited platelet aggregation induced by PAF, which did not cause thromboxane formation. This inhibition could be due to the calcium antagonizing effect or inhibitor of intracellular calcium mobilization [12]. Further experiments are needed to elucidate their mechanism of action.

It has been reported that high K^+-induced constrictions in smooth muscle is mediated by an increase in Ca^{2+}

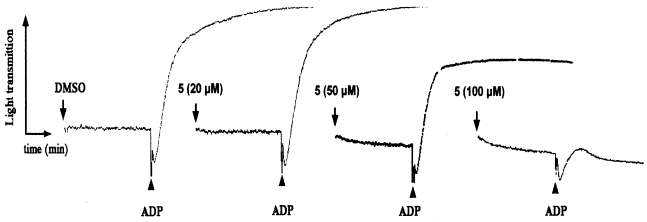


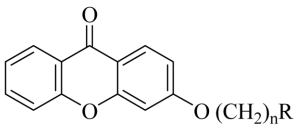
Figure 4. Effect of **5** on the aggregation of human PrP induced by ADP.
Human PrP was pre-incubated with dimethylsulfoxide (0.5%, control) or various concentrations of **5** for 3 min and then ADP (20 μM) was added to trigger aggregation.

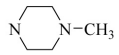
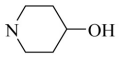
Table 2. Effect of xanthone derivatives on high K^+-, Ca^{2+}-induced, and noradrenaline-induced contraction of rat thoracic aorta.[a)]

Compound (μM)	K^+ (80 mM) + Ca^{2+} (1.9 mM)	Noradrenaline	
		(3 μM) Phasic	(3 μM) Tonic
Control	100 ± 7.2	100 ± 6.7	100 ± 2.3
1 (120)	85.9 ± 3.7	87.1 ± 2.7	100.5 ± 5.1
2 (120)	52.9 ± 8.6[b)]	52.7 ± 5.1[d)]	68.5 ± 1.3[c)]
3 (10)	104.0 ± 2.8	72.1 ± 2.0[b)]	84.3 ± 7.6
4 (120)	33.5 ± 6.0[d)]	8.7 ± 0.3[c)]	10.2 ± 1.3[c)]
5	ND	ND	ND
6 (60)	56.8 ± 3.7[c)]	86.1 ± 1.0	95.5 ± 3.2
7 (60)	105.9 ± 4.2	108.3 ± 5.9	100.0 ± 0.0
8 (40)	79.4 ± 4.7	98.0 ± 6.9	110.2 ± 5.4
9 (40)	71.4 ± 2.6	87.0 ± 1.7	84.6 ± 4.3[b)]
10 (40)	64.9 ± 1.2[b)]	69.1 ± 0.6[b)]	82.7 ± 3.2[d)]
11 (60)	35.0 ± 4.2[d)]	57.1 ± 6.7[b)]	33.3 ± 3.0
12	ND	ND	ND
13 (60)	80.4 ± 1.5	88.1 ± 3.4	88.9 ± 7.9
14 (60)	28.4 ± 1.7[c)]	89.2 ± 2.9	67.5 ± 7.3[b)]
15 (60)	66.2 ± 7.6[b)]	94.2 ± 7.7	88.8 ± 8.0
16 (60)	16.0 ± 0.9[c)]	75.6 ± 1.6[b)]	55.7 ± 3.0[c)]
17 (60)	15.9 ± 2.4[c)]	90.0 ± 1.2	85.8 ± 3.0[b)]
18 (60)	14.4 ± 2.0[c)]	63.8 ± 5.4[b)]	39.0 ± 11.0[d)]

[a)] Rat aorta was preincubated with xanthone derivatives or DMSO (0.1%, control) at 37°C for 15 min, then high K^+ (80 mM) and Ca^{2+} (1.9 mM), or noradrenaline (3 μM) was added. Percentages of the contraction were calculated and presented as means ± S.E.M (n = 3), ND = not determined.
[b)] $P < 0.05$.
[b)] $P < 0.01$.
[c)] $P < 0.001$ as compared with the respective control value.

Table 3. Physical data of xanthone derivatives.



Compound	R	n	Yield (%)	Mp. (°C)	Recrystallization solvent
1	$N(CH_3)_2$	2	71	218–220	$CHCl_3$-MeOH
2	$N(CH_3)_2$	3	74	219–221	$CHCl_3$-MeOH
3	$N(CH_3)_2$	4	75	212–216	$CHCl_3$-MeOH
4	$N(CH_3)_2$	5	68	205–208	$CHCl_3$-MeOH
5	$N(CH_3)_2$	6	62	160–163	$CHCl_3$-MeOH
6	$N(C_2H_5)_2$	4	51	172–173	$CHCl_3$-MeOH
7	$N(CH_3)C_2H_4OH$	4	65	213–214	$CHCl_3$-MeOH
8		4	61	211–214	$CHCl_3$-MeOH
9		4	64	188–190	$CHCl_3$-MeOH
10		4	62	229–232	$CHCl_3$-MeOH
11		4	54	168–171	$CHCl_3$-MeOH
12		4	56	183–185	$CHCl_3$-MeOH
13	NHC_2H_5	4	72	208–212	$CHCl_3$-MeOH
14	NHC_4H_9	4	68	152–154	$CHCl_3$-MeOH
15	NHC_2H_4OH	4	64	86–89	$CHCl_3$-MeOH
16	$NHCH(CH_3)C_2H_5$	3	66	217–218	$CHCl_3$-MeOH
17	$NHCH(CH_3)C_2H_5$	4	69	172–175	MeOH
18	$NHCH(CH_3)C_2H_5$	5	62	118–121	MeOH

influx through voltage-dependent Ca^{2+} channels. Compounds **4**, **14** and **16**–**18** depressed markedly the constrictions induced by Ca^{2+} (1.9 mM) in high-K^+ (80 mM) medium. These compounds may be blocker of voltage-dependent Ca^{2+} channels [2]. The maintenance of tonic constriction in response to noradrenaline results primarily from Ca^{2+} influx through receptor activated Ca^{2+} channels with little requirement for Ca^{2+} influx through voltage dependent Ca^{2+} channels. Compound **4** also depressed markedly the constrictions induced by noradrenaline (3 μM) (Table 2). It revealed that **4** may be also a blocker of receptor-activated Ca^{2+} channels [2]. Further experiments are needed to elucidate their mechanism of action. Compounds **4**, **16**, and **17** possess antiplatelet and vasorelaxing activity. This study further verifies that compounds **4**, **5**, **12**, **17**, and **18** showed significant inhibitory effects on platelet aggregation. These compounds revealed antiplatelet effects on washed rabbit platelets and in human PrP, and vasorelaxing activities in rat thoracic aorta. These dual activities revealed that these compounds can serve as a new antithrombotic agent.

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The authors have declared no conflict of interest.

Experimental

Melting points (uncorrected) were determined with a Yanaco Micro-Melting Point apparatus (Yanaco Seisakusho Co. Ltd, Japan). IR spectra were determined with a Perkin-Elmer system 2000 FTIR spectrophotometer (Perkin Elmer, Norwalk, CT, USA). ^{1}H- (400 MHz) and ^{13}C- (100 MHz) NMR spectra were recorded on a Varian Unity-400 spectrometer (Varian Inc., Palo Alto, CA, USA) and MS spectra were obtained on a JMS-HX-100 mass spectrometer (JEOL, Ltd. Japan). Elemental analyses were performed by Vario EL III (Elementar Analysensysteme GmbH, Hanau, Germany). Chromatography was performed using a flash-column technique on silica gel 60 supplied by E. Merck (Darmstadt, Germany).

Chemistry

*General procedure for the synthesis of compounds **1–5***

To a solution of KOH (1.28 g, 22.6 mmol) in H_2O (1.0 mL) were added *t*-BuOH (30 mL), I (3.72 g, 17.6 mmol) and 1, 2-dibromo-

ethane or 1, 3-dibromopropane or 1, 4-dibromobutane or 1, 5-dibromopentane or 1, 6-dibromohexane (each 45.2 mmol), and the mixture was stirred for 3 h under reflux. The reaction mixture was evaporated and the organic material was extracted with ether. Extracts were washed with H_2O, dried, and evaporated *in vacuo*. The residue was purified by a silica-gel column to yield II (82–85%). A solution of II (0.76 g, 2.2 mmol) and 50% aqueous $(CH_3)_2NH$ (20 mL) in THF (20 mL) was stirred at room temperature for 6 h. The organic material was extracted with ether and the extracts were washed with brine, dried, and evaporated *in vacuo*. The residue dissolved in ether was added ethanolic HCl or HBr (20%, 0.5 mL) to afford various crystals, which were recrystallized from appropriate solvent (Table 1) to obtain **1** (0.50 g, 1.57 mmol), **2** (0.54 g, 1.63 mmol), **3** (0.57 g, 1.65 mmol), **4** (0.54 g, 1.50 mmol), and **5** (0.51 g, 1.36 mmol).

3-(2-(Dimethylamino)ethoxy)xanthone hydrochloride ***1***

Physical data: see Table 3. IR (KBr) 1645, 1618 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 2.93 (s, 6H, Me × 2), 3.56 (t, 2H, *J* = 4.4 Hz, NCH_2), 4.54 (t, 2H, *J* = 4.4 Hz, OCH_2), 6.92 (dd, 1H, *J* = 8.3, 2.4 Hz, H-2), 6.94 (d, 1H, *J* = 2.4 Hz, H-4), 7.32–7.42 (m, 2H, H-6 and H-7), 7.66 (m, 1H, H-5), 8.17 (d, 1 H, *J* = 8.3 Hz, H-1), 8.22 (dd, 1H, *J* = 8.1, 1.6 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 43.6 (Me × 2), 56.3 (NCH_2), 63.1 (OCH_2), 101.3 (C-4), 113.1 (C-2), 116.4 (C-9a), 117.6 (C-5), 121.6 (C-8a), 124.1 (C-7), 126.4 (C-8), 128.5 (C-1), 134.7 (C-6), 156.1 (C-10a), 157.8 (C-4a), 162.4 (C-3), 176.5 (CO). EIMS (70 eV) *m/z* (% rel. int.): 283 $[M]^+$ (0.9), 195 (4), 155 (4), 139 (25), 58 (100). Anal. Calcd. for $C_{17}H_{17}NO_3$. HCl . H_2O: C, 60.40; H, 6.00; N, 4.10. Found: C, 60.51; H, 5.96; N, 4.00.

3-(3-(Dimethylamino)propoxy)xanthone hydrochloride ***2***

Physical data: see Table 3. IR (KBr) 1645, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 2.33 (m, 2H, CH_2), 2.98 (s, 6H, Me × 2), 3.41 (t, 2H, *J* = 6.1 Hz, NCH_2), 4.26 (t, 2H, *J* = 5.8 Hz, OCH_2), 7.01 (dd, 1H, *J* = 8.6, 2.4 Hz, H-2), 7.04 (d, 1H, *J* = 2.4 Hz, H-4), 7.42–7.55 (m, 2H, H-6 and H-7), 7.77 (m, 1H, H-5), 8.13 (d, 1H, *J* = 8.6 Hz, H-1), 8.22 (dd, 1H, *J* = 8.0, 1.6 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 24.1 (CH_2), 42.7 (Me × 2), 56.3 (NCH_2), 65.1 (OCH_2), 100.7 (C-4), 113.3 (C-2), 115.7 (C-9a), 117.6 (C-5), 121.4 (C-8a), 123.9 (C-7), 126.2 (C-8), 128.0 (C-1), 134.5 (C-6), 156.0 (C-10a), 157.9 (C-4a), 163.6 (C-3), 176.6 (CO). EIMS (70 eV) *m/z* (% rel. int.): 297 $[M]^+$ (4) 183 (7), 139 (13), 127 (12), 58 (100). Anal. Calcd. for $C_{18}H_{19}NO_3$. HCl . H_2O: C, 61.40; H, 6.30; N, 4.00. Found: C, 61.38; H, 6.28; N, 3.75.

3-(4-(Dimethylamino)butoxy)xanthone hydrochloride ***3***

Physical data: see Table 3. IR (KBr) 1640, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.88–2.00 (m, 4H, CH_2 × 2), 3.08 (m, 2H, NCH_2), 4.06 (t, 2H, *J* = 5.6 Hz, OCH_2), 6.79 (d, 1H, *J* = 2.4 Hz, H-4), 6.84 (dd, 1H, *J* = 8.7, 2.4 Hz, H-2), 7.29-7.40 (m, 2H, H-6 and H-7), 7.63 (m, 1H, H-5), 8.11 (d, 1H, *J* = 8.7 Hz, H-1), 8.19 (dd, 1H, *J* = 7.9, 1.8 Hz, H-8), ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 21.8 (CH_2), 26.5 (CH_2), 43.1 ($N(CH_3)_2$), 57.9 (NCH_2), 67.9 (OCH_2), 101.2 (C-4), 114.0 (C-2), 116.1 (C-9a), 118.2 (C-5), 122.1 (C-8a), 124.7 (C-7), 126.9 (C-8), 128.6 (C-1), 135.0 (C-6), 155.6 (C-10a), 158.5 (C-4a), 164.6 (C-3), 177.1 (CO). EIMS (70 eV) *m/z* (% rel. int.): 311 $[M]^+$ (4), 236 (3),183 (3), 139 (9), 83 (26), 69 (38), 58 (100). Anal. Calcd. for $C_{19}H_{21}NO_3$. HCl: C, 65.60; H, 6.40; N, 4.00. Found: C, 65.55; H, 6.38; N, 3.95.

3-(5-(Dimethylamino)pentoxy)xanthone hydrochloride ***4***

Physical data: see Table 3. IR (KBr) 1640, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.63 (m, 2H, CH_2), 1.89–2.01 (m, 4H, CH_2 × 2), 2.85 (s, 6H, Me × 2), 3.06 (m, 2H, NCH_2), 4.09 (t, 2H, *J* = 6.3 Hz, OCH_2), 6.85 (d, 1H, *J* = 2.1 Hz, H-4), 6.91 (dd, 1H, *J* = 9.0, 2.1 Hz, H-2), 7.37–7.45 (m, 2H, H-6 and H-7), 8.22 (d, 1H, *J* = 9.0 Hz, H-1), 8.31 (dd, 1H, *J* = 8.0, 1.8 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 23.2 (CH_2), 24.1 (CH_2), 28.3 (CH_2), 42.8 ($N(CH_3)_2$), 57.7 (NCH_2), 67.8 (OCH_2), 100.6 (C-4), 113.2 (C-2), 115.7 (C-9a), 117.6 (C-5), 121.8 (C-8a), 123.8 (C-7), 126.5 (C-8), 128.1 (C-1), 134.3 (C-6), 156.1(C-10a), 157.9 (C-4a), 164.2 (C-3), 176.2 (10). EIMS (70 eV) *m/z* (% rel. int.): 325 $[M]^+$ (1), 183 (3), 155 (4), 139 (6), 114 (37), 58 (100). Anal. Calcd. for $C_{20}H_{23}NO_3$ · HCl · H_2O: C, 63.20; H, 6.90; N, 3.70. Found: C, 63.07; H, 6.89; N, 3.51.

3-(6-(Dimethylamino)hexoxy)xanthone hydrochloride ***5***

Physical data: see Table 3. IR (KBr) 1640, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.54 (m, 4H, CH_2 × 2), 1.88 (m, 4H, CH_2 × 2), 2.78 (s, 6H, Me × 2), 2.97 (m, 2H, NCH_2), 4.08 (t, 1H, *J* = 6.3 Hz, OCH_2), 6.86 (1H, *J* = 2.4 Hz, H-4), 6.89 (dd, 1H, *J* = 8.7, 2.4 Hz, H-2), 7.36–7.45 (m, 2H, H-6 and H-7), 7.69 (m, 1H, H-5), 8.23 (d, 1H, *J* = 8.7 Hz, H-1), 8.32 (dd, 1H, *J* = 8.1, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 24.4 (CH_2), 25.5 (CH_2), 26.4 (CH_2), 28.7 (CH_2), 42.9 ($N(CH_3)_2$), 58.0 (NCH_2), 68.2 (OCH_2), 100.6 (C-4), 113.5 (C-2), 115.7 (C-9a), 117.7 (C-5), 121.9 (C-8a), 123.9 (C-7), 126.6 (C-8), 128.2 (C-1), 134.3 (C-6), 156.2 (C-10a), 158.0 (C-4a), 164.4 (C-3), 176.3 (CO). EIMS (70 eV) *m/z* (% rel. int.): 339 $[M]^+$ (3), 183 (1), 139 (4), 128 (8), 58 (100). Anal. Calcd. for $C_{21}H_{25}NO_3$ · HBr · H_2O: C, 57.50; H, 6.40; N, 3.20. Found: C, 58.86; H, 6.85; N, 3.40.

General procedure for the synthesis of ***6–12***

To a solution of II (0.85 g, 2.47 mmol) in THF (20 mL) was added diethylamine or *N*-methylethanolamine or pyrrolidine or piperidine or *N*-methylpiperazine or morpholine or 4-hydroxy piperidine (each 3.80 mmol) and NaOH (0.1 g, 2.5 mmol) in H_2O (1.0 mL). The reaction mixture was stirred at 70°C for 8 h. The organic material was extracted with ether and the extracts were washed with brine, dried, and evaporated *in vacuo*. To the residue dissolved in ether was added ethanolic HCl or HBr (20%, 0.5 mL) to afford various products which were recrystallized from appropriate solvent (see Table 1) to yield **6** (0.47 g, 1.26 mmol), **7** (0.61 g, 1.61 mmol), **8** (0.56 g, 1.51 mmol), **9** (0.61 g, 1.58 mmol), **10** (0.62 g, 1.53 mmol), **11** (0.52 g, 1.33 mmol), and **12** (0.56 g, 1.38 mmol).

3-(4-(Diethylamino)butoxy)xanthone hydrochloride ***6***

Physical data: see Table 3. IR (KBr) 1640, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.42 (m, 6H, Me × 2), 1.95–2.05 (m, 4H, $CH_2$62), 2.96–3.22 (m, 6H, $N(CH_2)$ 63), 4.12 (t, 1H, *J* = 5.7 Hz, OCH_2), 6.83 (brs, 1H, H-4), 6.87 (dd, 1H, *J* = 9.0 Hz) H-2, 7.34, 7.42 (m, each 1H, H-6 and H-7), 7.67 (m, 1H, H-5), 8.19 (d, 1H, *J* = 9.0 Hz, H-1), 8.27 (d, 1H, *J* = 7.8 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 8.6 (Me), 11.1 (Me), 20.6 (CH_2), 26.3 (CH_2), 42.2 (NCH_2), 46.5 (NCH_2), 51.1(NCH_2), 67.4 (OCH_2), 100.7 (C-4), 113.3 (C-2), 115.9 (C-9a), 117.7 (C-5), 121.8 (C-8a), 123.9 (C-7), 126.5 (C-8), 128.2 (C-1), 134.3 (C-6), 156.1 (C-10a), 157.9 (C-10a), 163.9 (C-3), 176.1 (CO). EIMS (70 eV) *m/z* (% rel. int.): 339 $[M]^+$ (1), 324 (2), 225 (3), 183 (3), 139 (8), 86 (100). Anal. Calcd. for $C_{21}H_{25}NO_3$ · HBr · H_2O: C, 57.50; H, 6.40; N, 3.20. Found: C, 57.57; H, 6.51; N, 3.55.

3-(4-(N-Methyl-N(2-hydroxyethyl)amino)butoxy)xanthone hydrochloride ***7***

Physical data: see Table 3. IR (KBr) 3300, 1645, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.85–2.00 (m, 4H, CH_2 × 2), 2.84 (s,

3H, NMe), 3.17 (m, 4H, $N(CH_2 \times 2)$), 3.89 (t, 2H, *J* = 5.1 Hz, CH_2OH), 4.07 (t, 2H, *J* = 5.7 Hz, OCH_2), 6.81 (d, 1H, *J* = 2.1 Hz, H-4), 6.85 (dd, 1H, *J* =8.7, 2.1 Hz, H-2), 7.30–7.38 (m, 2H, H-6 and H-7), 7.63 (m, 1H, H-5), 8.11 (d, 1H, *J* = 8.7 Hz, H-1), 8.19 (d, 1H, *J* = 8.1 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 20.8 (CH_2), 25.9 (CH_2), 40.5 (NCH_3), 55.7 (NCH_2), 56.3 (NCH_2), 57.8 (CH_2OH), 67.4 (OCH_2), 100.6 (C-4), 115.5 (C-9a), 117.3 (C-5), 121.5 (C-8a), 123.9 (C-7), 126.3 (C-8), 128.0 (C-1), 134.5 (C-6), 156.1 (C-10a), 158.0 (C-4a), 164.1 (C-3), 176.7 (CO). ESIMS *m/z* 342 $[M+1]^+$. HRESIMS: Calcd. for $C_{20}H_{24}NO_4$: 342.1705. Found: 342.1704.

3-(4-Pyrrolidinobutoxy)xanthone hydrochloride ***8***

Physical data: see Table 3. IR (KBr) 1645, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.98 (m, 2H, CH_2), 2.17 (m, 6H, $CH_2 \times 3$), 3.13–3.60 (m, 6H, $N(CH_2 \times 3)$), 4.13 (t, 2H, *J* = 6.0 Hz, OCH_2), 6.85 (d, 1H, *J* = 2.1 Hz, H-4), 6.91 (dd, 1H, *J* = 9.0, 2.4 Hz, H-2), 7.38–7.46 (m, 2H, H-6 and H-7), 7.69 (m, 1H, H-5), 8.23 (d, 1H, *J* = 9.0 Hz, H-1), 8.32 (dd, 1H, *J* = 8.1, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 22.8 (CH_2), 23.3 ($CH_2 \times 2$), 26.3 (CH_2), 53.6 ($N(CH_2)_2$), 55.1 (NCH_2), 67.5 (OCH_2), 100.7 (C-4), 113.4 (C-2), 115.9 (C-9a), 117.7 (C-5), 121.9 (C-8a), 123.9 (C-7), 126.6 (C-8), 128.3 (C-1), 134.3 (C-6), 156.2 (C-10a), 158.0 (C-4a), 163.9 (C-3), 176.2 (CO). EIMS (70 eV) *m/z* (% rel. int.): 337 $[M]^+$ (12). Anal. Calcd. for $C_{21}H_{23}NO_3 \cdot HCl$: C, 67.50; H, 6.50; N, 3.70. Found: C, 67.42; H, 6.18; N, 3.40.

3-(4-Piperidinobutoxy)xanthone hydrochloride ***9***

Physical data: see Table 3. IR (KBr) 1640, 1615 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.87–2.04 (m, 10H, $CH_2 \times 5$), 2.63–350 (m, 4H, $N(CH_2)_2$), 4.80 (t, 2H, *J* = 6.0 Hz, OCH_2), 6.83 (d, 1H, *J* = 2.4 Hz, H-4), 6.86 (dd, 1H, *J* = 8.7, 2.4 Hz, H-2), 7.32–7.41 (m, 2H, H-6 and H-7), 7.64 (m, 1H, H-5), 8.14 (d, 1H, *J* = 9.0 Hz, H-1), 8.22 (dd, 1H, *J* = 7.8, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 20.6 (CH_2), 21.8 (CH_2), 22.5($CH_2 \times 2$), 26.1 (CH_2), 53.1($N(CH_2)_2$), 56.7 (NCH_2), 67.1 (OCH_2), 100.6 (C-4), 113.5 (C-2), 115.6 (C-9a), 117.6 (C-5), 121.5 (C-8a), 123.9 (C-7), 126.3 (C-8), 128.0 (C-1), 134.5 (C-6), 156.1 (C-10a), 158.0 (C-4a), 164.1 (C-3), 176.6 (CO). EIMS (70 eV) *m/z* (% rel. int.): 351 $[M]^+$ (2), 195 (1), 183 (1), 139 (4), 98 (100). Anal. Calcd. for $C_{22}H_{25}NO_3 \cdot HCl \cdot 2H_2O$: C, 62.30; H, 7.10; N, 3.30. Found: C, 62.34; H, 7.51; N, 3.65.

3-(4-(4-Methylpiperazino)butoxy)xanthone hydrochloride ***10***

Physical data: see Table 3. IR (KBr) 1645, 1625 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.87–2.02 (m, 4H, $CH_2 \times 2$), 2.86 (3H, NMe), 3.21 (m, 2H, NCH_2), 3.59–3.76 (m, 8H, $N(CH_2 \times 4)$, 4.05 (t, 2H, *J* = 6.0 Hz, OCH_2), 6.81 (1H, *J* = 2.4 Hz, H-4), 6.85 (dd, 1H, *J* = 8.7, 2.4 Hz, H-2), 7.30–7.38 (m, 2H, H-6 and H-7), 7.62 (m, 1H, H-5), 8.11 (d, 1H, *J* = 8.7 Hz, H-1), 8.18 (dd, 1H, *J* = 8.1, 1.8 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 20.5 (CH_2), 25.8 ($CH_2 \times 2$), 42.5 (NCH_3), 48.0 ($N(CH_2)_2$), 49.6 ($N(CH_2)_2$), 56.5 (NCH_2), 56.5 (NCH_2), 67.1 (OCH_2), 100.6 (C-4), 113.4 (C-2), 115.5 (C-9a), 117.6 (C-5), 121.4 (C-8a), 123.9 (C-7), 126.2 (C-8), 128.0 (C-1), 134.5 (C-6), 156.0 (C-10a), 157.9 (C-4a), 164.0 (C-3), 176.7 (CO). EIMS (70 eV) *m/z* (% rel. int.): 366 $[M]^+$ (12), 310 (3), 183 (3), 113 (100), 70 (52). Anal. Calcd. for $C_{22}H_{26}N_2O_3 \cdot 2HCl$: C, 60.10; H, 6.40; N, 6.40. Found: C, 60.27; H, 6.55; N, 6.05.

3-(4-Morpholinolbutoxy)xanthone hydrochloride ***11***

Physical data: see Table 3. IR (KBr) 1645, 1625 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.88 (m, 4H, $CH_2 \times 2$), 2.94 (m, 6H, $N(CH_2 \times 3)$), 3.92 (m, 4H, $OCH_2 \times 2$), 4.05 (t, 2H, *J* = 5.9 Hz, OCH_2), 6.81 (d, 1H, *J* = 2.0 Hz, H-4), 6.84 (dd, 1H, *J* = 8.7, 2.4 Hz, H-2), 7.26–7.40 (m, 2H, H-6 and H-7), 7.63 (m, 1H, H-5), 8.11 (d, 1H, *J* = 8.7 Hz, H-1), 8.20 (dd, 1H, *J* = 8.1, 1.7 Hz, H-8), ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 20.4 (CH_2), 26.2 (CH_2), 51.9 ($N(CH_2)_2$), 57.4 (NCH_2), 63.6 ($O(CH_2)_2$), 67.4 (OCH_2), 100.7 (C-4), 113.4 (C-2), 115.8 (C-9a), 117.7 (C-5), 121.8 (C-8a), 123.9 (C-7), 126.5 (C-8), 128.3 (C-1), 134.4 (C-6), 156.1 (C-10a), 158.0 (C-4a), 164.0 (C-1), 176.4 (CO). EIMS (70 eV) *m/z* (% rel. int.): 353$[M]^+$ (2), 322 (2), 195 (2), 183 (2), 139 (7), 100 (100). Anal. Calcd. for $C_{21}H_{23}NO_4 \cdot HCl \cdot H_2O$: C, 61.80; H, 6.40; N, 3.40. Found: C, 61.50; H, 6.58; N, 3.51.

3-(4-(4-Hydroxypiperidino)butoxy)xanthoen hydrochloride ***12***

Physical data: see Table 3. IR (KBr) 3400, 1650, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.90–2.33 (m, 8H, $CH_2 \times 4$), 3.11–3.27 (4 m, H, $N(CH_2 \times 2)$, 3.53 (m, 2H, NCH_2), 4.15 (m, 2H, OCH_2), 6.90 (brs, 1H, H-4), 6.94 (d, 1H, *J* = 8.7 Hz, H-2), 7.38-7.48 (m, 2H, H-6 and H-7), 7.74 (m, 1H, H-5), 8.21 (d, 1H, *J* = 8.7 Hz, H-1), 8.29 (dd, 1H, *J* = 8.1, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 20.7 (CH_2), 26.2 (CH_2), 29.4 ($CH_2 \times 2$), 47.5 ($N(CH_2 \times 2)$), 56.9 (NCH_2), 60.0 (CHOH), 67.5 (OCH_2), 100.7 (C-4), 113.6 (C-2), 115.6 (C-9a), 117.7 (C-5), 121.6 (C-8a), 124.0 (C-7), 126.4 (C-8), 128.1 (C-1), 134.6 (C-6), 156.1 (C-10a), 158.0 (C-4a), 164.2 (C-3), 176.7 (CO). EIMS (70 eV) *m/z* (% rel. int.): 367 $[M]^+$ (2), 212 (7), 195 (2), 114 (100). Anal. Calcd. for $C_{22}H_{25}NO_4 \cdot HCl \cdot H_2O$: C, 62.60; H, 6.70; N, 3.30. Found: C, 62.30; H, 6.75; N, 3.16.

General procedure for the synthesis of ***13–18***

To a solution of compound II (1.0 g, 2.38 mmol) in *t*-BuOH (25 mL) was added ethylamine or *n*-butylamine or ethanolamine or *sec*-butylamine (4.0 mmol) and NaOH (0.2 g, 5.0 mmol) in H_2O (1.0 mL). The reaction mixture was stirred at 70°C for 5 h. The organic material was extracted with ether and the extracts were washed with brine, dried, and evaporated *in vacuo*. The residue was purified by column chromatography. The eluate dissolved in ether was added ethanolic HCl or HBr (20%, 0.5 mL) to give various products which recrystallized from appropriate solvent (see Table 1) to afford **13** (0.55 g, 1.62 mmol), **14** (0.53 g, 1.71 mmol), **15** (0.50 g, 1.52 mmol), **16** (0.51 g, 1.57 mmol), **17** (0.56 g, 1.64 mmol), and **18** (0.52 g, 1.48 mmol).

3-(4-(Ethylamino)butoxy)xanthone ***13***

Physical data: see Table 3. IR (KBr) 1650, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm):1.42 (t, 3H, *J* = 7.2 Hz, Me), 2.01 (m, 4H, $CH_2 \times 2$), 3.09 (m, 4H, $N(CH_2 \times 2)$), 4.14 (t, 2H, *J* = 5.4 Hz, OCH_2), 6.88 (d, 1H, *J* = 2.1 Hz, H-4), 6.93(dd, 1H, *J* = 8.7, 2.4 Hz, H-2), 7.37–7.44 (m, 2H, H-6 and H-7), 7.70 (m, 1H, H-5), 8.19 (d, 1H, *J* = 8.7 Hz, H-1), 8.27 (dd, 1H, *J* = 7.8, 1.2 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 10.8 (CH_3), 22.9 (CH_2), 25.9 (CH_2), 42.8 (NCH_2), 46.8 (NCH_2), 67.5 (OCH_2), 100.5 (C-4), 113.5 (C-2), 115.4 (C-9a), 117.6 (C-5), 121.4 (C-8a), 123.4 (C-7), 126.2 (C-8), 127.9 (C-1), 134.5 (C-6), 156.0 (C-10a), 157.9 (C-4a), 164.2 (C-3), 176.7 (CO). EIMS (70 eV) *m/z* (% rel. int.): 311 $[M]^+$ (128). Anal. Calcd. for $C_{19}H_{21}NO_3 \cdot HBr \cdot H_2O$: C, 55.60; H, 5.90; N, 3.40. Found: C, 55.90; H, 5.60; N, 3.55.

3-(4-(n-Butylamino)butoxy)xanthone ***14***

Physical data: see Table 3. IR (KBr) 1645, 1625 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.92 (t, 3H, *J* = 7.2 Hz, Me), 1.39–1.74 (m, 4H, $CH_2 \times 2$), 1.97 (m, 4H, $CH_2 \times 2$), 2.83-2.92 (m, 4H, $N(CH_2) \times 2$), 4.07

(t, 2H, *J* = 6.0 Hz, OCH_2), 6.81 (d, 1H, *J* = 2.4 Hz, H-4), 6.90 (dd, 1H, *J* = 9.0, 2.4 Hz, H-2), 7.31 – 7.42 (m, 2H, H-6 and H-7), 7.66 (m, 1H, H-5), 8.21 (d, 1 H, *J* = 9.0 Hz, H-1), 8.29 (dd, 1H, *J* = 8.1, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 13.7 (Me), 20.3 (CH_2), 24.5 (CH_2), 26.6 (CH_2), 29.7 (CH_2), 48.4 (NCH_2), 48.6 (NCH_2), 67.9 (OCH_2), 100.6 (C-4), 113.4 (C-2), 115.8 (C-9a), 117.6 (C-5), 121.9 (C-8a), 123.8 (C-7), 126.6 (C-8), 128.3 (C-1), 134.2 (C-6), 156.1 (C-10a), 157.9 (C-4a), 164.2 (C-3), 176.2 (CO). EIMS (70 eV) *m/z* (% rel. int.): 339 $[M]^+$ (5). Anal. Calcd. for $C_{21}H_{25}NO_3 \cdot HCl \cdot H_2O$: C, 64.00; H, 7.20; N, 3.60. Found: C, 64.15; H, 7.20; N, 3.49.

3-(4-(2-Hydroxyethyl)amino)butoxy)xanthone ***15***

Physical data: see Table 3. IR (KBr) 3420, 1645, 1625 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 1.78 – 1.94 (m, 4H, $CH_2 \times 2$), 2.79 (m, 4H, $N(CH_2 \times 2)$), 3.71 (t, 2H, *J* = 4.8 Hz, 2H, CH_2OH), 4.14 (t, 2H, *J* = 6.3Hz, OCH_2), 6.92 (d, 1H, *J* = 2.1 Hz, H-4), 6.97 (dd, 1H, *J* = 8.7, 2.4, H-2), 7.40 – 7.50 (m, 2H, H-6 and H-7), 7.74 (t, 1H, *J* = 8.4 Hz, H-5), 8.22 (d, 1H, *J* = 8.7 Hz, H-1), 8.30 (dd, 1H, *J* = 7.8, 1.5, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 25.6 (CH_2), 26.4 (CH_2), 49.5 (CH_2), 50.7 (CH_2), 60.8 (CH_2OH), 68.1 (OCH_2), 100.5 (C-4), 113.2 (C-2), 115.4 (C-9a), 117.6 (C-5), 121.4 (C-8a), 123.4 (C-7), 126.2 (C-8), 128.5 (C-1), 134.5 (C-6), 156.0 (C-10a), 158.0 (C-4a), 164.2 (C-3), 176.8 (CO). ESIMS *m/z* 328 $[M+1]^+$. HRESIMS: Calcd. for $C_{19}H_{22}NO_4$: 328.1549. Found: 328.1548.

3-(3-(sec-Butylamino)propoxy)xanthone ***16***

Physical data: see Table 3. IR (KBr) 1645, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$-CD_3OD) δ (ppm): 0.91 (t, 3H, *J* = 7.4 Hz, Me), 1.28 (d, 3H, *J* = 6.62 Hz, $CHCH_3$), 1.52 (m, 1H, CHH), 1.83 (m, 1H, CHH), 2.27 (m, 2H, CH_2), 3.06 (m, 3H, NCH_2 and $NCH(CH_3)C_2H_5$), 3.23 (m, 1H, 1H, NCH), 4.12 (t, 2H, *J* = 5.7 Hz, OCH_2), 6.83 (d, 1H, *J* = 2.2 Hz, H-4), 6.87 (dd, 1H, *J* = 9.4, 2.4 Hz, H-2), 7.27 – 7.39 (m, 2H, H-6 and H-7), 7.62 (m, 1H, H-5), 8.09 (d, 1H, *J* = 9.4 Hz, H-1), 8.16 (dd, 1H, *J* = 7.9, 1.7 Hz, H-8). ^{13}C-NMR ($CDCl_3$-CD_3OD) δ (ppm): 9.4 (Me), 15.1 (Me), 25.6 ($CH_2 \times 2$), 42.0 (NCH_2), 55.8 (NCH), 65.4 (OCH_2), 100.7 (C-4), 113.4 (C-2), 115.6 (C-9a), 117.6 (C-5), 121.4 (C-8a), 123.9 (C-7), 126.2 (C-8), 128.0 (C-1), 134.5 (C-6), 156.1 (C-10a), 157.9 (C-4a), 163.8 (C-3), 176.7 (CO). EIMS (70 eV) *m/z* (% rel. int.): 325 $[M]^+$ (1). Anal. Calcd. for $C_{20}H_{23}NO_3 \cdot HBr$: C, 59.10; H, 6.00; N, 3.40. Found: C, 59.31; H, 5.94; N, 3.42.

3-(4-(sec-Butyllamino)butoxy)xanthone ***17***

Physical data: see Table 3. IR (KBr) 1645, 1625 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.00 (t, 3H, *J* = 7.2 Hz, Me), 1.48 (d, 3H, *J* = 6.6 Hz, $CHCH_3$), 1.72 – 2.23 (m, 6H, $CH_2 \times 3$), 3.09 (m, 2H, NCH_2), 3.21 (m, 1H, NCH), 4.04 (t, 2H, *J* = 6.0 Hz, OCH_2), 6.75 (d, 1H, *J* = 2.4 Hz, H-4), 6.87 (dd, 1H, *J* = 9.0, 2.4 Hz, H-2), 7.28 – 7.38 (m, 2H, H-6 and H-7), 7.63 (m, 1H, H-5), 8.18 (d, 1H, *J* = 9.0 Hz, H-1), 8.25 (dd, 1H, *J* = 7.8, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 10.2 (Me), 15.5 (Me), 23.0 (CH_2), 25.8 (CH_2), 26.5 (CH_2), 44.3 (NCH_2), 56.1 (NCH), 67.6 (OCH_2), 100.6 (C-4), 113.3 (C-2), 115.8 (C-9a), 117.6 (C-5), 121.8 (C-8a), 123.8 (C-7), 125.5 (C-8), 128.3 (C-1), 134.2 (C-6), 156.0 (C-10a), 157.8 (C-4a), 163.9 (C-3), 176.1 (CO). EIMS (70 eV) *m/z* (% rel. int.): 339 $[M]^+$ (1). Anal. Calcd. for $C_{21}H_{25}NO_3 \cdot HBr$: C, 60.00; H, 6.20; N, 3.30. Found: C, 59.92; H, 6.24; N, 3.30.

3-(5-(sec-Butylamino)pentoxy)xanthone ***18***

Physical data: see Table 3. IR (KBr) 1645, 1620 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.93 (t, 3H, *J* = 7.5 Hz, Me), 1.24 (d, 3H, *J* = 6.4 Hz, $CHCH_3$). 1.56 – 1.82 (m, 8H, $CH_2 \times 4$), 2.81 (m, 3H, NCH_2 and NCH), 4.03 (t, 2H, *J* = 6.3 Hz, OCH_2), 6.79 (d, 1H, *J* = 2.4 Hz, H-4), 6.89 (dd, 1H, *J* = 8.9, 2.7 Hz, H-2), 7.31 – 7.41 (m, 2H, *J* = H-6 and H-7), 7.63 (m, 1H, H-5), 8.19 (d, 1H, *J* = 8.9 Hz, H-1), 8.25 (dd, 1H, *J* = 7.9, 1.5 Hz, H-8). ^{13}C-NMR ($CDCl_3$) δ (ppm): 10.2 (Me), 17.5 (Me), 23.6 (CH_2), 27.6 (CH_2), 27.8 (CH_2), 28.6 (CH_2), 45.7 (NH_2), 55.3 (NCH), 68.2 (OCH_2), 100.6 (C-4), 113.5 (C-2), 115.6 (C-9a), 117.6 (C-5), 121.8 (C-8a), 123.8 (C-7), 126.5 (C-8), 128.1 (C-1), 134.2 (C-6), 156.1 (C-10a), 157.9 (C-4a), 164.4 (C-3), 176.2 (CO). EIMS (70 eV) *m/z* (% rel. int.): 353 $[M]^+$ (2). Anal. Calcd. for $C_{22}H_{27}NO_3 \cdot HBr$: C, 60.80; H, 6.50; N, 3.20. Found: C, 60.72; H, 6.82; N, 3.50.

Biological assays

Platelet aggregation

Washed rabbit platelets were obtained from EDTA-anticoagulated PrP according to procedures described previously. Human PrP was obtained from the supernatant after centrifugation of a 1 : 9 mixture of blood and sodium citrate solution (3.8%). Platelet numbers were counted by using a Coulter Counter (Model ZM; Beckman Coulter, USA) and adjusted to 4.5×10^8 platelets/mL. The platelet pellets were suspended in Tyrode's solution containing (mM): NaCl (136.8), KCl (2.8), $NaHCO_3$ (11.9), $MgCl_2$ (2.1), NaH_2PO_4 (0.33), $CaCl_2$ (1.0), and glucose (11.2) with 0.35% BSA. All glassware was siliconized. PrP or the platelet suspension was stirred at 1200 rev/min 1 min before addition of the aggregation inducer. Aggregation was measured by a turbidimetric method. The absorbance platelet-poor plasma or platelet-free Tyrode's solution was taken as 100% aggregation. The aggregation was measured by means of a Lumi-aggregometer (Chrono-Log Co., USA) connected to dual channel recorders [4].

Aortic contraction

Wistar rats of either sex, 250 – 300 g, were killed by a blow to the head. The thoracic aorta was isolated, excess fat and connective tissue were removed and placed in organ bath containing 5 mL Krebs solution, maintained at 37°C, and bubbled with a 95% O_2 / 5% CO_2 mixture. Two stainless-steel hooks were inserted into the aortic lumen, one was fixed while the other was connected to a transducer. Aorta were equilibrated in the medium for 90 min with three changes of Krebs solution and maintained under an optimal tension of 1 g before specific experimental protocols were initiated. Contractions were recorded isometrically via a force displacement transducer connected to a Gould polygraph (Model 2400; Gould Instruments,USA). The final concentration of DMSO was fixed at 0.1% [4].

Data analysis

Data are presented as means ± S.E.M. One-way analysis was used for multiple comparison, and if significant variation was observed between the treatment groups and the inhibitor-treated groups, they were then compared with the control group by student's t-test. Values of $P < 0.05$ were considered statistically significant.

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