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Short communication

2-Arylalkyl-substituted anthracenones as inhibitors of 12-lipoxygenase enzymes. 1. Structure–activity relationships of the terminal aryl ring

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Abstract – A series of 2-arylmethyl-substituted anthracenones were synthesized and tested as inhibitors of three types of 12-lipoxygenase isoforms in epidermal homogenate of mice, bovine platelets and porcine leucocytes. Their inhibitory activities were compared with those to inhibit the 5-lipoxygenase enzyme in bovine leucocytes. Structure–activity relationships are described with particular emphasis on modifications of the terminal aryl nucleus. The ability of the compounds to selectively inhibit the 12-lipoxygenase enzymes was dependent on a high overall lipophilicity of the inhibitor, whereas compounds with decreased lipophilicity were also inhibitors of the 5-LO enzyme. Among the more lipophilic inhibitors, the unsubstituted 2-phenylmethyl analogue **6a** as well as the carboxylic acid ester **6q** appeared to be selective inhibitors of platelet-type 12-LO isoform. © 2001 Éditions scientifiques et médicales Elsevier SAS

anthracenone / 12(*S*)-HETE / lipophilicity / 5-lipoxygenase / 12-lipoxygenase

1. Introduction

Lipoxygenases are a superfamily of enzymes that catalyse the transfer of molecular oxygen into arachidonic acid to yield hydroperoxy fatty acids, which are further metabolised to hydroxy derivatives as end products [1]. These enzymes show not only marked stereospecificity but also differ in the position of dioxygenation in arachidonic acid, and the currently used nomenclature is based on this specificity of the enzyme acting on its substrate [2]. Lipoxygenase proteins have a catalytical domain containing a non-heme iron, and oxidation to the active ferric form is required for catalysis [1].

The physiological role of individual mammalian lipoxygenases is uncertain aside from 5-lipoxygenase, which is the key enzyme of the biosynthesis of leucotrienes [3]. Inhibitors of the 5-lipoxygenase enzyme are used in the therapy of asthma [3] and could provide useful therapy in inflammatory skin diseases such as psoriasis [4, 5]. Different isoforms of 12-lipoxygenase have been described. Based upon biochemical and immunological criteria, 12-lipoxygenases have been characterized into platelet-type (*p*12-LO) and leucocyte-type (*l*12-LO) categories [6]. Furthermore, mouse epidermal lipoxy-genase (*e*12-LO) represents a third isoform that is more related to *p*12-LO than to *l*12-LO [7, 8]. Formation of the 12(*S*)-hydroxyeicosatetraenoic acid (12(*S*)-HETE) enantiomer can be accounted for by these isoforms [9–12]. Recently, *R*-lipoxygenases have also been identified [13, 14]. However, for the 12-LO isoforms, their biological roles are far from clear. Increased 12-LO activity has been demonstrated to induce apoptosis in fibroblasts [15], and its metabolite 12(*S*)-HETE has been reported to be critically involved in cancer metastasis [16] and hyperproliferative skin diseases such as psoriasis [4].

Abbreviations: 12(*S*)-HETE, 12(*S*)-hydroxyeicosatetraenoic acid; LO, lipoxygenase.

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In earlier reports on structure–activity relationships of the anthracenone class of antipsoriatic compounds, we have established 10-substituted derivatives of anthralin (**1**, *figure 1*) such as **2** as inhibitors of 5-lipoxygenase [17–19]. In this paper, we report the synthesis of 2-arylmethyl analogues and the assessment of their inhibitory activities against three types of 12-lipoxygenase isoforms and 12(*S*)-HETE biosynthesis, with particular emphasis on modifications of the terminal aryl nucleus.

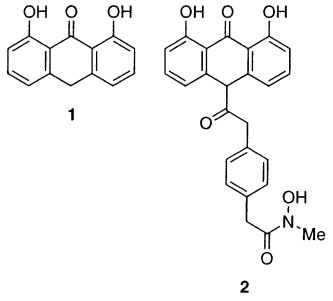


Fig. 1. Structures of anthralin (**1**) and a 10-substituted derivative (**2**).

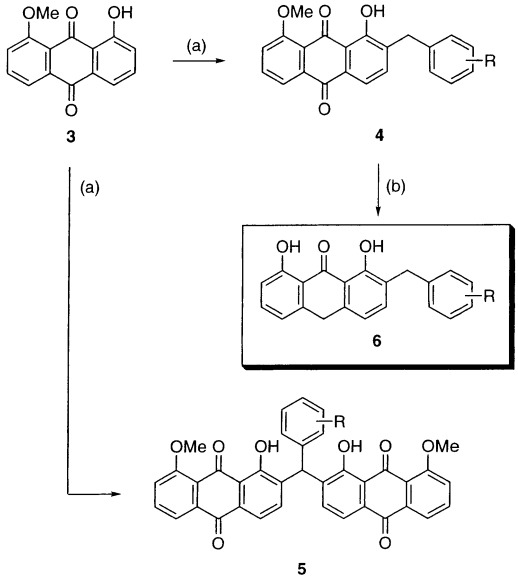


Fig. 2. Reagents: (a) $Na_2S_2O_4$, NaOH, R-PhCHO (X-CHO), N_2, 90 °C; (b) $SnCl_2$, HCl, glacial HOAc, 118 °C. R or X are defined in *table I*.

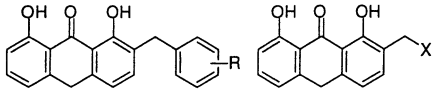
2. Chemistry

As hydroxyanthracenones are chemically unstable under alkaline conditions, introduction of the 2-arylmethyl functionality onto the anthracenone nucleus was achieved at the anthracenedione stage. Accordingly, the requisite 2-substituted anthracenedione precursors were obtained by Marschalk reaction [20] from 1-hydroxy-8-methoxy-9,10-anthracenedione (**3**) [21] as the key intermediate (*figure 2*). Alkylation of **3** with benzaldehydes provided the 2-benzylated anthracenediones **4a**–**4r**. In some cases, only moderate yields were obtained as a consequence of dimer formation. Dimerisation products similar to the methylene-bridged bisanthraquinone such as **5b**, which could be isolated in up to 20% yield, have also been observed in the reaction of other anthracenediones with formaldehyde [22]. The formation of these unpleasant side products could be reduced by addition of a 5-fold excess of the benzaldehydes. On the other hand, alkylation of **3** proceeded cleanly with naphthaldehydes and thiophene-2-carboxaldehyde to give anthracenediones **4w**–**4y**. Finally, reduction of **4a**–**4y** with $SnCl_2$–HCl proceeded with concomitant ether cleavage of the 8-methoxy group, which was selective for those compounds having an ether function in the 2-substituent (**6i**–**6p**), to produce the desired anthracenones **6a**–**6y**.

3. Lipoxygenase inhibition

Although the literature of 5-LO inhibitors is vast [3, 23], not many compounds have been tested for 12-LO inhibitory activity. However, 12-HETE is the major lipoxygenase product found in the skin [24] and has been demonstrated to be present in high levels in psoriatic epidermis [25]. Furthermore, the characterization of LO activity in human epidermis indicates that germinal layer keratinocytes contain a highly active 12-LO which is selectively expressed at a higher level during psoriatic inflammation [11]. 12-HETE stimulates keratinocyte proliferation and induces histological changes characteristic of psoriasis [26–28]. Based on this information, we investigated structure–activity relationships of the 2-substituted anthracenones by evaluating their ability to inhibit the 12-LO pathway in epidermal homogenate of mice [29]. In order to explore their selectivity, we also evaluated their inhibitory action against 12-LO isoen-

Table I. Inhibition of mouse epidermal 12-LO, bovine platelet 12-LO, porcine leucocyte 12-LO, and bovine leucocyte 5-LO by 2-arylmethyl-1,8-dihydroxy-9(10*H*)-anthracenones.



6a–6v 6w–6y

Cpd	R or X	log *P* [a]	*e*12-LO [b] IC_{50} (μM)	*p*12-LO [c] IC_{50} (μM)	*l*12-LO [d] IC_{50} (μM)	5-LO [e] IC_{50} (μM)
6a	H	5.62	>30	13	>30	>30
6b	4-Ph	6.52	>30	>30	23	>30
6c	4-Me	6.04	30	21	28	>30
6d	4-CF_3	5.92	>30	12	16	>30
6e	4-F	6.03	9	9	18	>30
6f	4-Cl	5.98	9	14	26	>30
6g	4-Br	6.35	19	12	27	>30
6h	4-CN	4.31	12	14	8	>30
6i	3-OPh	6.35	16	12	>30	>30
6j	4-O*n*-Pr	6.06	>30	38	18	>30
6k	4-OEt	5.80	>30	32	15	>30
6l	4-OMe	5.62	30	11	22	>30
6m	3,4-$(OMe)_2$	5.04	30	14	11	>30
6n	2,4-$(OMe)_2$	5.79	24	12	14	>30
6o	2,5-$(OMe)_2$	5.72	>30	8	16	>30
6p	3,4-OCH_2O	4.51	>30	19	12	>30
6q	4-CO_2Me	5.34	>30	9	>30	>30
6r	4-CO_2H	4.10	11	13	14	2
6s	4-OH	4.66	10	14	13	7
6t	3,4-$(OH)_2$	4.01	10	8	4	4
6u	2,4-$(OH)_2$	3.68	10	8	7	4
6v	2,5-$(OH)_2$	3.33	5	7	6	0.9
6w	1-naphthyl	6.96	>30	>30	>30	>30
6x	2-naphthyl	7.07	>30	>30	>30	>30
6y	2-thienyl	5.33	30	>30	16	26
anthralin	–	4.23	9	9	9	37
NDGA [f]	–	–	21	12	24	0.5

[a] Experimentally determined partition coefficient.
[b] Inhibition of 12(*S*)-HETE biosynthesis in mouse epidermal homogenates, [c]bovine platelets, and [d]porcine leucocytes.
[e] Inhibition of LTB_4 biosynthesis in bovine leucocytes. For each value of the LO assays, inhibition was significantly different with respect to that of the control ($n = 3$ or more, S.D. $\leq 10\%$, $P < 0.01$).
[f] NDGA was used as the standard inhibitor.

zymes in bovine platelets and porcine leucocytes as well as their ability to inhibit 5-LO in bovine leucocytes.

4. Results and discussion

The structures and enzyme inhibitory activities of the novel anthracenones are listed in *table I*. As IC_{50} values for lipoxygenase inhibition are dependent on cell density [30] and may vary with the assay performed, parent anthralin (**1**) and the general lipoxygenase inhibitor nordihydroguaiaretic acid (NDGA) were used as reference compounds. Anthralin was only a moderate inhibitor of 5-LO in bovine leucocytes and an equally potent inhibitor of the 12-LO isoforms, whereas NDGA appeared to be a more selective inhibitor of 5-LO. The IC_{50} values against the 12-LO enzymes reported here are not as low as those reported in other studies [31, 32], although the values are not strictly comparable because of differences in the assay conditions.

As compared to anthralin, introduction of a 2-benzyl substituent particularly decreased potency against

*e*12-LO and *l*12-LO. Accordingly, unsubstituted **6a** was found to be a selective inhibitor of the 12-LO platelet-type. The effect of various substituents on the terminal aromatic ring was then investigated. The addition of a *para*-carboxylic acid ester (**6q**) substituent also yielded a *p*12-LO selective inhibitor with slightly improved inhibitory action. Halogen (**6e–6g**) and cyano (**6h**) substituents gave compounds with activity for all 12-LO isoforms. Compounds having ether groups, as in **6i–6p**, showed more or less inhibitory action against 12-LO, but did not display specificity for a single 12-LO isoform. A striking feature was the observation that any substitution of the aromatic ring gave compounds of general selective 12-LO inhibitory action as opposed to 5-LO, as long as the overall lipophilicity of the inhibitor exhibited a log *P* value of considerably higher than 4. The monophenol **6s** was an exception. On the other hand, compounds with decreased lipophilicity were also inhibitors of the 5-LO enzyme.

The results in *table I* further reflect the importance of acidic groups such as carboxyl or hydroxyl in the terminal phenyl ring for additional inhibition of 5-LO. The carboxylic acid derivative **6r** was a more potent inhibitor of 5-LO than the 12-LO enzymes. Also, substitution on the terminal phenyl ring by hydroxyl groups produced activity against the 5-LO enzyme. Since phenols are known to react with the iron centre of the enzyme, it is not surprising that compounds **6s–6v** are inhibitors of both 5-LO and the 12-LO isoforms. It may be expected that electron transfer from these phenols to the catalytically active ferric form of the lipoxygenase enzymes results in an inactivated enzyme in the ferrous form. Even though there are potent inhibitors among the phenols such as **6v**, these agents would be poor choices as potentially selective inhibitors.

A terminal naphthyl ring, as in compounds **6w** and **6x**, did not substitute for the phenyl group without loss of activity. This may be due to their exceptionally high lipophilicities with log *P* values of about 7. Finally, substitution of the terminal phenyl ring by a thienyl group (**6y**) resulted in a moderate inhibitor of the leucocyte-type 12-LO.

Even though specific 12-LO inhibition would be useful for differentiating the physiological roles of the 12-LO isoenzymes in disease states such as psoriasis and cancer, no specific inhibitor of high potency against 12-LO could be identified that would serve as a lead compound for drug development.

5. Conclusions

In conclusion, the ability to selectively inhibit the 12-LO enzymes as opposed to 5-LO was quite dependent on the lipophilicity of the compounds. Unsubstituted 2-phenylmethyl analogue **6a** as well as the carboxylic acid ester **6q** were identified as selective inhibitors of the platelet-type 12-LO isoform. However, none of the 2-substituted anthracenones was able to discriminate between the epidermal-type and the other 12-LO isoforms.

6. Experimental protocols

6.1. Chemistry

6.1.1. General

For analytical instruments and methods see Ref. [17].

6.1.2. General procedure for the preparation of 2-arylmethyl-1-hydroxy-8-methoxy-9,10-anthracenediones by Marschalk alkylation

*6.1.2.1. 2-[(4-Biphenyl)methyl]-1-hydroxy-8-methoxy-9,10-anthracenedione (**4b**)*

To a solution of 1.5% NaOH in water (150 mL) was added **3** [21] (2.54 g, 10.0 mmol) and a solution of $Na_2S_2O_4$ (3.0 g, 17.3 mmol) in water (15 mL) at 40 °C under N_2. The solution was heated to 50 °C for 15 min. 4-Biphenylaldehyde (9.05 g, 50 mmol) was added dropwise over 30 min at 70 °C, and the reaction mixture was stirred for 2 h. It was stirred for an additional 12 h at 90 °C, then cooled to room temperature, aerated for 30 min, and acidified with 6 N HCl. The orange suspension thus obtained was extracted with CH_2Cl_2 (3×100 mL), and the organic phase was washed with water and dried over Na_2SO_4. The solvent was evaporated, and the residue was purified by column chromatography (SiO_2–CH_2Cl_2) to provide orange–yellow crystals; 13% yield; m.p. 176–178 °C; ^{1}H-NMR ($CDCl_3$) δ 13.42 (s, 1H), 7.97–7.26 (m, 14H), 4.15 (s, 2H), 4.07 (s, 3H); FTIR 3432 (OH), 1669 (CO), 1632 cm^{-1} (CO···OH). Anal.: $C_{28}H_{20}O_4$ (C, H).

*6.1.2.2. 2-[(4-Fluorophenyl)methyl]-1-hydroxy-8-methoxy-9,10-anthracenedione (**4e**)*

The title compound was obtained from **3** and 4-fluorobenzaldehyde as described for **4b** to provide orange crystals; 23% yield; m.p. 193–194 °C; ^{1}H-NMR

($CDCl_3$) δ 13.35 (s, 1H), 7.95–6.85 (m, 9H), 4.05 (s, 2H), 4.00 (s, 3H); FTIR 3432 (OH), 1699 (CO), 1634 cm^{-1} (CO···OH); MS $m/z = 362$ (100, M^+). Anal.: $C_{22}H_{15}FO_4$ (C, H).

*6.1.2.3. 2-[(4-Bromophenyl)methyl]-1-hydroxy-8-methoxy-9,10-anthracenedione (**4g**)*

The title compound was obtained from **3** and 4-bromobenzaldehyde as described for **4b** to provide orange crystals; 12% yield; m.p. 214–215 °C; ^{1}H-NMR ($CDCl_3$) δ 13.40 (s, 1H), 8.00–7.10 (m, 9H), 4.05 (s, 2H), 4.00 (s, 3H); FTIR 3433 (OH), 1699 (CO), 1634 cm^{-1} (CO···OH). Anal.: $C_{22}H_{15}BrO_4$ (C, H).

*6.1.2.4. 4-[(9,10-Dihydro-1-hydroxy-8-methoxy-9,10-dioxo-2-anthracenyl)methyl]benzonitrile (**4h**)*

The title compound was obtained from **3** and 4-cyanobenzaldehyde as described for **4b** to provide yellow crystals; 24% yield; m.p. 252–253 °C; ^{1}H-NMR ($CDCl_3$) δ 13.40 (s, 1H), 7.98–7.26 (m, 9H), 4.15 (s, 2H), 4.08 (s, 3H); FTIR 3433 (OH), 2229 (CN), 1669 (CO), 1632 cm^{-1} (CO···OH). Anal.: $C_{23}H_{15}NO_4$ (C, H, N).

*6.1.2.5. 1-Hydroxy-8-methoxy-2-[(4-propoxyphenyl)-methyl]-9,10-anthracenedione (**4j**)*

The title compound was obtained from **3** and 4-propoxybenzaldehyde as described for **4b** to provide yellow crystals; 8% yield; m.p. 166–169 °C; ^{1}H-NMR ($CDCl_3$) δ 13.42 (s, 1H), 7.97–6.83 (m, 9H), 4.15 (s, 2H), 4.05 (s, 3H), 3.90 (t, $J = 6.5$ Hz, 2H), 1.88 (sext, $J = 6.5$ Hz, 2H), 1.03 (t, $J = 6.5$ Hz, 3H); FTIR 3432 (OH), 1669 (CO), 1631 cm^{-1} (CO···OH). Anal.: $C_{25}H_{22}O_5$ (C, H).

*6.1.2.6. 2-[(4-Ethoxyphenyl)methyl]-1-hydroxy-8-methoxy-9,10-anthracenedione (**4k**)*

The title compound was obtained from **3** and 4-ethoxybenzaldehyde as described for **4b** to provide orange crystals; 24% yield; m.p. 153–154 °C; ^{1}H-NMR ($CDCl_3$) δ 13.39 (s, 1H), 7.97–6.81 (m, 9H), 4.15 (s, 2H), 4.08 (s, 3H), 4.01 (q, $J = 7$ Hz, 2H), 1.40 (t, $J = 7$ Hz, 3H); FTIR 3450 (OH), 1665 (CO), 1631 cm^{-1} (CO···OH). Anal.: $C_{24}H_{20}O_5$ (C, H).

*6.1.2.7. 1-Hydroxy-8-methoxy-2-[(3,4-methylenedioxyphenyl)methyl]-9,10-anthracenedione (**4p**)*

The title compound was obtained from **3** and 3,4-methylenedioxybenzaldehyde as described for **4b** to provide yellow crystals; 46% yield; m.p. 196–197 °C; ^{1}H-NMR ($CDCl_3$) δ 13.38 (s, 1H), 7.95–7.25 (m, 8H), 5.90 (s, 2H), 4.10 (s, 2H), 4.05 (s, 3H); FTIR 3427 (OH), 1667 (CO), 1634 cm^{-1} (CO···OH).); MS $m/z = 388$ (100, M^+). Anal.: $C_{23}H_{16}O_6$ (C, H).

*6.1.2.8. 1-Hydroxy-8-methoxy-2-[(1-naphthyl)methyl]-9,10-anthracenedione (**4w**)*

The title compound was obtained from **3** and 1-naphthaldehyde as described for **4b** to provide orange–yellow crystals; 46% yield; m.p. 219–222 °C; ^{1}H-NMR ($CDCl_3$) δ 13.58 (s, 1H), 7.97–7.09 (m, 12H), 4.56 (s, 2H), 4.09 (s, 3H); FTIR 3422 (OH), 1667 (CO), 1627 cm^{-1} (CO···OH). Anal.: $C_{26}H_{18}O_4$ (C, H).

*6.1.2.9. 1-Hydroxy-8-methoxy-2-[(2-naphthyl)methyl]-9,10-anthracenedione (**4x**)*

The title compound was obtained from **3** and 2-naphthaldehyde as described for **4b** to provide orange crystals; 52% yield; m.p. 162–163 °C; ^{1}H-NMR ($CDCl_3$) δ 13.44 (s, 1H), 7.98–7.26 (m, 12H), 4.27 (s, 2H), 4.07 (s, 3H); FTIR 3449 (OH), 1667 (CO), 1627 cm^{-1} (CO···OH). Anal.: $C_{26}H_{18}O_4$ (C, H).

*6.1.2.10. 1-Hydroxy-8-methoxy-2-(2-thenyl)-9,10-anthracenedione (**4y**)*

The title compound was obtained from **3** and thiophene-2-carboxaldehyde as described for **4b** to provide yellow crystals; 40% yield; m.p. 169–171 °C; ^{1}H-NMR (DMSO-d_6) δ 13.43 (s, 1H), 7.90–6.94 (m, 8H), 4.24 (s, 2H), 3.98 (s, 3H); FTIR 3444 (OH), 1667 (CO), 1636 cm^{-1} (CO···OH). Anal.: $C_{20}H_{14}O_4S$ (C, H).

*6.1.2.11. 1-(4-Biphenyl)-1,1-bis-(1-hydroxy-8-methoxy-9,10-dioxo-2-anthracenyl)methane (**5b**)*

The title compound was obtained as described for **4b**. However, a different molar ratio of **3** (2.54 g, 10.0 mmol) and 4-biphenylaldehyde (0.91 g, 5.0 mmol) and double the volume of the solvent was used. **5b** was obtained as orange crystals; 6% yield; m.p. 213–216 °C; ^{1}H-NMR ($CDCl_3$) δ 13.44 (s, 2H), 7.97–7.21 (m, 19H), 6.48 (s, 1H), 4.04 (s, 6H); FTIR 3432 (OH), 1669 (CO), 1632 cm^{-1} (CO···OH); MS $m/z = 672$ (90, M^+), 420 (100). Anal.: $C_{43}H_{28}O_8$ (C, H).

6.1.3. General procedure for the reduction of 9,10-anthracenediones to 9(10H)-anthracenones [33]

*6.1.3.1. 2-[(4-Biphenyl)methyl]-1,8-dihydroxy-9(10H)-anthracenone (**6b**)*

To a solution of **4b** (0.35 g, 1.0 mmol) in glacial HOAc (20 mL) heated to reflux was added, dropwise

over 3 h, a solution of 40% $SnCl_2$ in 37% HCl (10 mL). The solution was then cooled, and the resulting crystals were collected by filtration. Purification by chromatography (SiO_2–CH_2Cl_2) provided yellow crystals; 24% yield; m.p. 178–181 °C; ^{1}H-NMR ($CDCl_3$) δ 12.68 (s, 1H), 12.34 (s, 1H), 7.97–7.26 (m, 14H), 4.34 (s, 2H), 4.12 (s, 2H); FTIR 3442 (OH), 1615 cm^{-1} (CO···OH). Anal.: $C_{27}H_{20}O_3$ (C, H).

*6.1.3.2. 2-[(4-Fluorophenyl)methyl]-1,8-dihydroxy-9(10H)-anthracenone (**6e**)*

The title compound was obtained from **4e** as described for **6b** to provide yellow crystals; 95% yield; m.p. 185–186 °C; ^{1}H-NMR ($CDCl_3$) δ 12.66 (s, 1H), 12.28 (s, 1H), 7.50–6.78 (m, 9H), 4.30 (s, 2H), 4.00 (s, 2H); FTIR 3448 (OH), 1615 cm^{-1} (CO···OH). Anal.: $C_{21}H_{15}FO_3$ (C, H).

*6.1.3.3. 2-[(4-Bromophenyl)methyl]-1,8-dihydroxy-9(10H)-anthracenone (**6g**)*

The title compound was obtained from **4g** as described for **6b** to provide yellow crystals; 87% yield; m.p. 171–172 °C; ^{1}H-NMR ($CDCl_3$) δ 12.62 (s, 1H), 12.22 (s, 1H), 7.45–6.75 (m, 9H), 4.25 (s, 2H), 3.93 (s, 2H); FTIR 3448 (OH), 1618 cm^{-1} (CO···OH). Anal.: $C_{21}H_{15}BrO_3$ (C, H).

*6.1.3.4. 4-[(9,10-Dihydro-1,8-dihydroxy-9-oxo-2-anthracenyl)methyl]benzonitrile (**6h**)*

The title compound was obtained from **4h** as described for **6b** to provide pale yellow crystals; 42% yield; m.p. 193–196 C; ^{1}H-NMR ($CDCl_3$) δ 12.67 (s, 1H), 12.23 (s, 1H), 7.59–6.85 (m, 9H), 4.34 (s, 2H), 4.08 (s, 2H); FTIR 3435 (OH), 2231 (CN), 1615 cm^{-1} (CO···OH). Anal.: $C_{22}H_{15}NO_3$ (C, H, N).

*6.1.3.5. 1,8-Dihydroxy-2-[(4-propoxyphenyl)methyl]-9(10H)-anthracenone (**6j**)*

The title compound was obtained from **4j** as described for **6b** to provide yellow crystals; 42% yield; m.p. 184–186 °C; ^{1}H-NMR ($CDCl_3$) δ 12.71 (s, 1H), 12.34 (s, 1H), 7.97–7.26 (m, 9H), 4.37 (s, 2H), 4.04 (s, 2H), 3.93 (t, $J = 6.5$ Hz, 2H), 1.87 (sext, $J = 6.5$ Hz, 2H), 1.05 (t, $J = 6.5$ Hz, 3H); FTIR 3448 (OH), 1618 cm^{-1} (CO···OH). Anal.: $C_{24}H_{22}O_4$ (C, H).

*6.1.3.6. 2-[(4-Ethoxyphenyl)methyl]-1,8-dihydroxy-9(10H)-anthracenone (**6k**)*

The title compound was obtained from **4k** as described for **6b** to provide yellow crystals; 66% yield; m.p. 169–170 °C; ^{1}H-NMR ($CDCl_3$) δ 12.63 (s, 1H), 12.31 (s, 1H), 7.97–6.80 (m, 9H), 4.31 (s, 2H), 4.15 (s, 2H), 4.01 (q, $J = 7$ Hz, 2H), 1.39 (t, $J = 7$ Hz, 3H); FTIR 3448 (OH), 1621 cm^{-1} (CO···OH). Anal.: $C_{23}H_{20}O_4$ (C, H).

*6.1.3.7. 1,8-Dihydroxy-2-[(3,4-methylenedioxyphenyl)-methyl]-9(10H)-anthracenone (**6p**)*

The title compound was obtained from **4p** as described for **6b** to provide pale yellow crystals; 37% yield; m.p. 164–167 °C; ^{1}H-NMR ($CDCl_3$) δ 12.64 (s, 1H), 12.30 (s, 1H), 7.51–6.74 (m, 8H), 5.91 (s, 2H), 4.31 (s, 2H), 3.94 (s, 2H); FTIR 3440 (OH), 1618 cm^{-1} (CO···OH); MS $m/z = 360$ (100, M$^+$). Anal.: $C_{22}H_{16}O_5$ (C, H).

*6.1.3.8. 1,8-Dihydroxy-2-[(1-naphthyl)methyl]-9(10H)-anthracenone (**6w**)*

The title compound was obtained from **4w** as described for **6b** to provide pale yellow crystals; 42% yield; m.p. 192–193 °C; ^{1}H-NMR ($CDCl_3$) δ 12.82 (s, 1H), 12.35 (s, 1H), 8.01–6.69 (m, 12H), 4.48 (s, 2H), 4.29 (s, 2H); FTIR 3440 (OH), 1618 cm^{-1} (CO···OH). Anal.: $C_{25}H_{18}O_3$ (C, H).

*6.1.3.9. 1,8-Dihydroxy-2-[(2-naphthyl)methyl]-9(10H)-anthracenone (**6x**)*

The title compound was obtained from **4x** as described for **6b** to provide pale yellow crystals; 47% yield; m.p. 166–168 °C; ^{1}H-NMR ($CDCl_3$) δ 12.69 (s, 1H), 12.31 (s, 1H), 7.81–6.80 (m, 12H), 4.31 (s, 2H), 4.20 (s, 2H); FTIR 3441 (OH), 1617 cm^{-1} (CO···OH). Anal.: $C_{25}H_{18}O_3$ (C, H).

*6.1.3.10. 1,8-Dihydroxy-2-(2-thenyl)-9(10H)-anthracenone (**6y**)*

The title compound was obtained from **4y** as described for **6b** to provide yellow crystals; 36% yield; m.p. 156–158 °C; ^{1}H-NMR ($CDCl_3$) δ 12.67 (s, 1H), 12.29 (s, 1H), 7.51–6.83 (m, 8H), 4.32 (s, 2H), 4.22 (s, 2H); FTIR 3443 (OH), 1619 cm^{-1} (CO···OH). Anal.: $C_{19}H_{14}O_3S$ (C, H).

Anthracenones **6a**, **6c**, **6d**, **6f**, **6i**, **6l**–**6o**, and **6q**–**6v** were prepared as described [34].

6.2. Biological assay methods

Epidermal homogenates from NMRI mice were prepared as described [29], platelets were obtained from sodium EDTA-anticoagulated bovine blood and were

suspended at a concentration of 4×10^7 cells mL^{-1}, and porcine leucocytes were prepared from porcine blood in a similar fashion as described [35] and were suspended at a concentration of 1×10^6 cells mL^{-1}. The 12-LO [29] and the 5-LO [35] assays were performed essentially as described previously in full detail. 12(*S*)-HETE was analysed by chiral phase chromatography as described [29]. Inhibition was calculated by the comparison of the mean values of test compound ($n=3$) with control ($n=6$–8) activity: (1−test compound/control)×100; S.D. ≤ 10%. Inhibition was statistically significant compared to that of the control (Student's *t*-test; $P<0.01$). Each IC_{50} value was derived by interpolation of a log inhibitor concentration versus response plot using four or more concentrations of the compound, spanning the 50% inhibition point.

6.3. Lipophilicity

Determination of log *P* values was performed by a standard reversed-phase HPLC procedure as described [35].

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