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NOVEL 10-SUBSTITUTED ANTIPSORIATIC ANTHRONES AS INHIBITORS OF EPIDERMAL 12-LIPOXYGENASE AND LIPID PEROXIDATION IN MEMBRANES

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Abstract—The ability of novel 10-substituted anthrones to inhibit 12-lipoxygenase (12-LO) in mouse epidermal homogenate and lipid peroxidation in both bovine brain phospholipid liposomes and erythrocyte ghosts was investigated, and compared with their ability to inhibit 5-lipoxygenase (5-LO) in bovine leukocytes. The compounds were fairly potent inhibitors of epidermal 12-LO, in addition to their strong inhibitory effects against leukocyte 5-LO. Although the antipsoriatic drug, anthralin, predominantly inhibited epidermal 12-LO, the novel derivatives were more selective 5-LO inhibitors. Compounds with free phenolic groups in the attached aromatic ring were also potent inhibitors of nonenzymatic lipid peroxidation in both sources of lipid substrate. This property was not correlated with their ability to inhibit the 5- and 12-LO pathways, suggesting that their mechanism of 5-/12-LO inhibition is not simply due to scavenging of peroxyl radicals generated at the active site of the enzymes. The compounds are dual-purpose inhibitors and may play a protective role against oxidative damage to psoriatic skin, in addition to their antiinflammatory 5-LO and 12-LO inhibitory properties.

Key words: anthralin; antipsoriatics; dithranol; lipid peroxidation; 12-lipoxygenase; psoriasis

Anthralin is an antipsoriatic anthrone that is widely used because of its reliability and effectiveness. One of its adverse effects is irritation of the skin surrounding treated psoriatic plaques [1]. Several lines of evidence derived from chemical and biological studies have revealed that anthralin can generate free radicals [2–4] and active oxygen species [5–7] during its autoxidation process. These species have been implicated in a wide variety of oxidative effects affecting cellular DNA, enzymes, lipids, and cell membranes [8–10]. With the realization that these biological targets are all influenced by antipsoriatic anthrones, it has been suggested that active oxygen species play a crucial role in the mode of action and manifestation of adverse side effects of these drugs [11].

In an attempt to produce agents with diminished or no irritancy as clinical alternatives to anthralin, we have developed a novel series of 10-substituted derivatives [12]. These agents show substantially reduced hydroxyl radical formation, and several compounds are approximately 100-fold more potent in inhibiting 5-LO† in bovine PMNL than anthralin [12]. LO products are lipid mediators with an important role in inflammatory disease states, such as psoriasis [13]. The regulation of the abnormal arachidonic acid metabolism has become a prime target for pharmacological intervention in psoriasis [14] because increased concentrations of leukotrienes and HETEs have been measured in psoriatic skin [15–17].

Although the literature on leukotriene antagonists and 5-LO inhibitors is vast, not many compounds have been tested for 12-LO inhibitory activity. However, 12-HETE is the major lipoxygenase product found in the skin [18] and has been demonstrated to be present in high levels in psoriatic epidermis [17, 19, 20]. In fact, the characterization of LO activity in human epidermis indicates that germinal layer keratinocytes contain a highly active 12-LO that is selectively expressed at a higher level during psoriatic inflammation [21]. 12-HETE stimulates keratinocyte proliferation and induces histological changes characteristic of psoriasis [22–24]. Studies on the evaluation of 12-LO inhibitors have identified platelets as the source of the enzyme in most cases [25, 26]. Observations that potent inhibitors of epidermal 12-LO did not inhibit platelet-derived 12-LO suggest the existence of two distinct enzymes [27, 28]. With psoriasis as target for therapy, potential 12-LO inhibitors have to be evaluated using epidermal 12-LO [29].

In this study we used epidermal strips from mouse skin to investigate the effects of some selected novel 10-substituted analogs [12] of the antipsoriatic drug anthralin on 12-LO activity. We also examined the capability of these compounds to inhibit nonenzymatic lipid peroxidation in bovine brain phospholipid liposomes and human erythrocyte ghosts cells. Figure 1 presents the structures of the compounds examined in this study.

MATERIALS AND METHODS

Reagents

All anthrone derivatives were prepared as described [12], 2,2′-azo-bis-(2-amidinopropane) hydrochloride was from Polysciences (Eppelheim, Germany). 12(*R*)- and 12(*S*)-HETE were purchased from Sigma (Deisen-

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† Abbreviations: AAPH, 2,2′-azo-bis-(2-amidinopropane) hydrochloride; BHT, 2,6-di-tert-butyl-4-methylphenol; DMSO, dimethylsulfoxide; HETE, hydroxyeicosatetraenoic acid; LO, lipoxygenase; MDA, malondialdehyde; MHA, 1-hydroxy-8-methoxy-9,10-anthracenedione; NDGA, nordihydroguaiaretic acid; PBS, phosphate buffered saline; PMNL, polymorphonuclear leukocytes; TBA, 2-thiobarbituric acid.

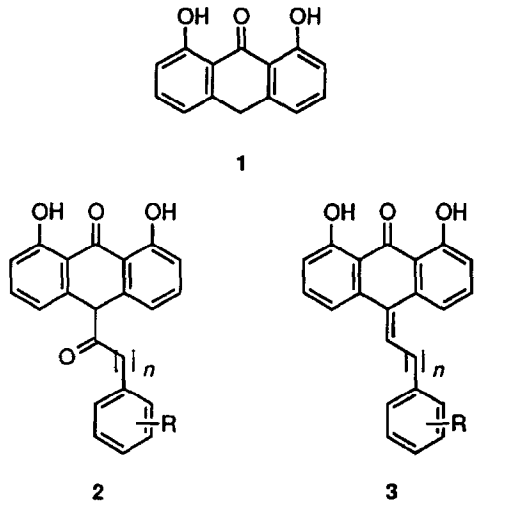

Fig. 1. Structures of anthralin (1), 10-ω-phenylacyl-1,8-dihydroxy-9(10*H*)-anthracenones (2) and 10-ω-phenylalkylidene-1,8-dihydroxy-9(10*H*)-anthracenones (3). R and *n* are defined in Table 1.

hofen, Germany). All other reagents were of the highest grades available from Sigma or from Merck (Darmstadt, Germany). UV spectroscopy was run on a Kontron 810 spectrophotometer.

Preparation of epidermal homogenates

Two days before use, female NMRI mice (Charles River, Sulzfeld, Germany) were shaved of dorsal hair with clippers. The animals were killed by cervical dislocation. The full-thickness skin was cut off, freed from blood vessels, glands, and hair and kept at 0°C. The epidermis was separated from the dermal tissue by scraping with a razor blade, and the epidermal pieces were sucked off. The epidermis was weighed, suspended in PBS, pH 7.4, (50 mg wet weight/mL) and homogenized in an ultra-turrax for 15 sec at 4°C. The homogenate was filtered through glasswool and kept at 0°C. Protein content of the epidermal homogenate was determined by the method of Bradford [30].

Epidermal 12-lipoxygenase assay

The epidermal homogenate (50 mg wet weight/mL) was allowed to warm to 37°C. It was then preincubated with 10 μL of anthrone derivative at the desired concentrations in DMSO or vehicle control (DMSO at final concentration of 0.4%) for 5 min at 37°C in a shaking water bath. Calcium chloride and arachidonic acid (final concentrations 2 mM and 5 μM, respectively) were added and the incubation conducted for 10 min at 37°C. Formation of 12-HETE was terminated by the addition of 3.0 mL MeOH/CH_3CN (1 + 1) containing NDGA and MHA as a chromatographic marker (final concentrations 0.3 and 30 μM, respectively). The incubation mixture was kept in an ice bath for 20 min and then centrifuged at 5000 *g* for 20 min at 0°C. Pertinent controls were performed to determine the original amounts of 12-HETE already present in the homogenate. The supernatant was diluted with 5 mL of water and passed through a prewashed octadecylsilane reversed phase cartridge (Baker) that had been washed with 5 mL MeOH and 5 mL water. The material was eluted with 3 mL MeOH, diluted with 3 mL water and subjected to reversed phase HPLC analysis performed on a 250 × 4 mm column (4 × 4 mm precolumn) packed with LiChrospher 100 RP18 (5-μm particles; Merck, Darmstadt, Germany). The isocratic elution conditions of 12-HETE were methanol/acetonitrile/water (35 + 35 + 30, vol), plus 0.1 vol % acetic acid, pH 5.5, flow rate 1.0 mL/min (Kontron 420 pump), monitored at 232 nm with a Kontron 735 LC UV detector. Data were recorded on a MacLab data acquisition system (WissTech, Germany) and analysis was performed with the software Peaks on an Apple Macintosh Quadra computer. Integrated areas of the peaks were compared to the internal standard and to external standards of authentic samples. Molar absorption coefficients given by Samuelsson *et al.* [31] were used for calculations; % inhibition of the formation of 12-HETE by epidermal homogenates was calculated by comparing the test compound ($N = 3$, SD < 10%) with control activity ($N = 8$, SD < 5%).

Stereochemical analysis by chiral phase chromatography

12-HETE obtained from *in vitro* incubation experiments was purified by isocratic reversed-phase HPLC as described above. This was, in turn, analyzed by chiral phase HPLC [32] on a Chiracel OB 250 × 4.6 mm column (Daicel, Tokyo, Japan). The isocratic elution conditions were hexane/2-propanol/methanol/water (99.1 + 0.68 + 0.1 + 0.03, vol), flow rate 1.0 mL/min (SP 8700 Thermo Separation Products, Darmstadt, Germany), monitored at 235 nm with a Knauer UV detector (Berlin, Germany). The retention times for 12-(*R*)- and 12-(*S*)-HETE were 24 and 35 min, respectively.

Isolation of phospholipids from bovine brain and preparation of liposomes

Phospholipids were prepared by the method of Gutteridge [33] as previously described [9].

Assay of lipid peroxidation

The assay was performed as recently described [9]. The following reagents were added to glass tubes in the order and at the final concentrations stated and preincubated for 3 min: 0.3 mL KH_2PO_4-KOH buffer, pH 7.4 (30 mM), 0.29 mL H_2O (double distilled), 0.2 mL liposomes (1 mg/mL), and 0.01 mL of anthrone derivative (variable concentrations). Appropriate blanks and controls with the vehicle (acetone) were conducted. The final reaction volume was standardized to 0.9 mL (when necessary, the volume of H_2O added was reduced). Then 0.1 mL AAPH (10 mM) was added and the reaction mixture incubated for 1 hr at 37°C in a shaking water bath. The reaction was terminated by the addition of BHT (10 μL, 20% w/v), 0.5 mL of 25% (w/v) HCl, and 0.5 mL of 1% (w/v in 0.05 N NaOH) TBA. Subsequently, the sample was heated at 100°C for 15 min and then cooled in an ice bath (5 min). Then, 2.0 mL of 1-butanol were added and the mixture vigorously shaken in a vortex mixer (Heidolph) for 15 sec. The organic layer was separated by centrifugation at 1500 *g* (15 min), and the absorbance at 532 nm was measured against 1-butanol. Calibration was performed using a malondialdehyde standard prepared by hydrolysis of 1,1,3,3,-tetraethoxypropane [34]. TBA-reactive material was expressed in terms of nmoles MDA per mg phospholipid (mean values ± SD, $N \geq 3$). IC_{50} values for inhibition of lipid peroxidation were derived by interpolation of a log concentration *vs* inhibition plot using 6 concentrations of

the compound, spanning the 50% inhibition point. All experiments were run in triplicate.

Preparation of erythrocyte ghosts

Resealed ghosts were prepared essentially as described by Dodge *et al.* [35]. Human blood of healthy volunteers was centrifuged at 5000 *g* for 5 min. The plasma and buffy coat were removed by aspiration. The erythrocytes were isolated by washing 3 times with a three-fold volume of a potassium citrate solution (280 mM, pH 7.2), centrifuging the cells at 5000 *g* for 5 min and carefully removing the supernatant. The resulting erythrocyte suspension was cooled (0°C), pipetted into centrifuge tubes, and treated with 5 times the volume of the hemolyzing solution consisting of citric acid (3.8 mM) and trimagnesium dicitrate (2 mM). The contents were mixed and centrifuged at 30,000 *g* and 0°C. The pellet was resuspended in the hemolyzing solution, and the procedure repeated until the supernatant was colorless. The isolated erythrocyte ghosts were then suspended in an isotonic solution of sodium citrate and stored below 4°C.

The anthrone derivatives (0.01 mL, variable concentrations) and the erythrocyte ghost suspension (0.9 mL) were preincubated at 37°C for 3 min. Then, 0.1 mL of AAPH (100 mM) was added and the reaction mixture incubated at 37°C for 3 hr, and the assay was performed as described above.

RESULTS

Inhibition of epidermal 12-lipoxygenase

To demonstrate the inhibitory effects of anthrones on epidermal 12-LO activity, we used homogenates of mouse epidermis. 12-HETE concentration was measured by reversed phase HPLC. The elution pattern of the main LO product was consistent with that of an authentic sample of 12-HETE. The absolute stereochemistry of the mouse epidermal 12-HETE was determined to confirm the presence of 12(*S*)-LO activity. Therefore, 12-HETE from *in vitro* incubation experiments was isolated and purified by reversed-phase HPLC and then subjected to chiral-phase HPLC. Comparison with authentic, stereochemically pure 12-HETE enantiomeric standards demonstrated that the 12-HETE present was the 12(*S*) isomer, clearly ruling out the presence of the 12(*R*) isomer. This excludes oxidation of arachidonic acid by a nonspecific free radical mechanism that gives racemic 12-HETE.

Anthralin (**1**), 10-phenylacyl (**2**), and 10-phenylalkylidene (**3**) substituted analogs were added to the mouse epidermal homogenate at micromolar concentrations, in the range 0–30 μM. The influence of these compounds on the production of 12-HETE is shown in Table 1. Incubation of mouse epidermal homogenates with anthralin resulted in a concentration-dependent inhibition of 12-HETE synthesis (Fig. 2) and gave an IC_{50} value of 9 μM. In contrast to Bedord's report [36], its inactive metabolite danthron (1,8-dihydroxy-9,10-anthracenedione) did not inhibit 12-HETE production at doses up to 30 μM (data not shown). On the other hand, with the exception of compound **3d** the novel 10-substituted derivatives of anthralin were all inhibitors of epidermal 12-LO, the catechol derivative **3b** of the phenylalkylidene series being the most effective with an IC_{50} value of 2 μM (Table 1). When the phenylalkylidene substituent of compound **3b** was replaced with a related catecholic phenylacyl substituent, as in **2c**, inhibitory activity against 12-LO was also observed (IC_{50} = 15 μM), but less than that of **3b**. In the phenylacyl series, the monophenolic compound **2b** was more effective than the catechol and pyrogallol derivatives **2c** and **2d**, respectively, suggesting that the number of phenolic hydroxyl groups is not important for 12-LO inhibition. Unexpectedly, a phenolic group in the terminating aromatic ring of the C-10 substituent of anthralin may not be necessary for 12-LO inhibitory activity because the nonphenolic derivatives **2e** and **2f** and the methylated analog of **2b** (i.e. compound **2a**) also exhibited anti-12-LO activity against the epidermal enzyme.

Table 1. Inhibition of mouse epidermal 12-LO, bovine PMNL 5-LO, lipid peroxidation in phospholipid liposomes, and lipid peroxidation in erythrocyte ghosts by 10-phenylacyl-(**2**), 10-phenylalkylidene-1,8-dihydroxy-9(10*H*)-anthracenones (**3**), and standard drugs

cpd*	*n*	R	12-LO IC_{50}, μM†	5-LO IC_{50}, μM‡	Selectivity§	LPO (PL) IC_{50}, μM‖	LPO (EG) IC_{50}, μM‖
2a	1	4-OCH_3	12	0.5	24	27	34
2b	1	4-OH	6	14	0.4	15	28
2c	1	3,4-$(OH)_2$	15	11	1.4	6	15
2d	1	3,4,5-$(OH)_3$	13	0.3	43	3	7
2e	2	H	10	0.5	20	30	25
2f	3	H	20	0.3	67	42	23
3a	0	4-OH	35	4	8.8	7	5
3b	0	3,4-$(OH)_2$	2	0.5	4	3	5
3c	0	3,4,5-$(OH)_3$	12	0.4	30	7	2
3d	2	H	>30	6	>5	29	7
Anthralin			9	37	0.2	79	10
NDGA			21	0.4	53	2	3

* The general formulas of the compounds are given in Fig. 1.

† Inhibition of 12-HETE biosynthesis in mouse epidermal homogenates. Inhibition was significantly different with respect to that of the control (DMSO), $N = 3$ or more, $P > 0.05$.

‡ The IC_{50} data for inhibition of 5-HETE and LTB_4 biosynthesis in bovine PMNL come from ref. [12].

§ Ratio of IC_{50} for 12-LO inhibition to that of 5-LO inhibition; ratio >1 indicates selective 5-LO inhibition.

‖ Inhibition of lipid peroxidation in bovine brain phospholipid liposomes (PL) and human erythrocyte ghosts (EG) stimulated by AAPH, $N = 3$ or more, $P > 0.05$; the IC_{50} values were obtained from at least 6 concentrations of the compounds.

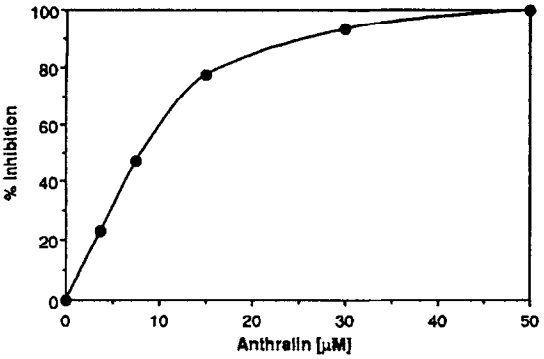

Fig. 2. Concentration-dependent inhibition of mouse epidermal 12-lipoxygenase by anthralin. Results are expressed as mean values (SD < 10%) of at least 3 tests at each concentration.

The relationship between the inhibitory effects of the anthrones against epidermal 12-LO and 5-LO in PMNL was then investigated. Figure 3A shows that there was no correlation between the effects of these novel anthrones in the two kinds of assays ($r = 0.04$). Furthermore, the ratio of the IC_{50} for 12-LO inhibition to that of 5-LO inhibition was taken as a parameter for selectivity for 12-LO inhibition (Table 1). In general, the novel derivatives were better inhibitors of 5-LO than of 12-LO, and compound **2b** was a more selective 12-LO inhibitor. Furthermore, the antioxidant NDGA was a selective inhibitor of 5-LO. By contrast, the antipsoriatic drug anthralin showed selectivity for epidermal 12-LO versus PMNL 5-LO with a factor of 4 in favor of 12-LO. Moreover, although anthralin is a powerful inhibitor of epidermal 12-LO, we observed much weaker inhibitory activity ($IC_{50} \approx 30$ μM) against the bovine platelet derived enzyme (data not shown).

Inhibition of lipid peroxidation in bovine brain phospholipid liposomes

The inhibitory effect on lipid peroxidation of the anthrones was evaluated with bovine brain phospholipid liposomes, which provide an ideal model system for lipid peroxidation studies [33]. Table 1 shows the results obtained in this system. The effects of anthralin and the standard compound NDGA are also shown in Table 1. All 10-substituted analogs of anthralin inhibited the release of TBA-reactive material significantly with IC_{50} values between 42 and 3 μM. Inhibition of AAPH-induced lipid peroxidation was strongly dependent on the presence of hydroxyl groups on the phenyl moiety of the 10-substituent, reflecting the antioxidant activity of the phenolic hydroxyl groups. On the other hand, inhibition of lipid peroxidation was independent of the nature of the chain linking the anthrone nucleus and the phenyl ring terminus. Relatively strong anti-lipid peroxidation activities were observed for compounds of both the phenylacyl and the phenylalkylidene series. Compounds **2d** and **3b** with IC_{50} values of 3 μM were almost equipotent

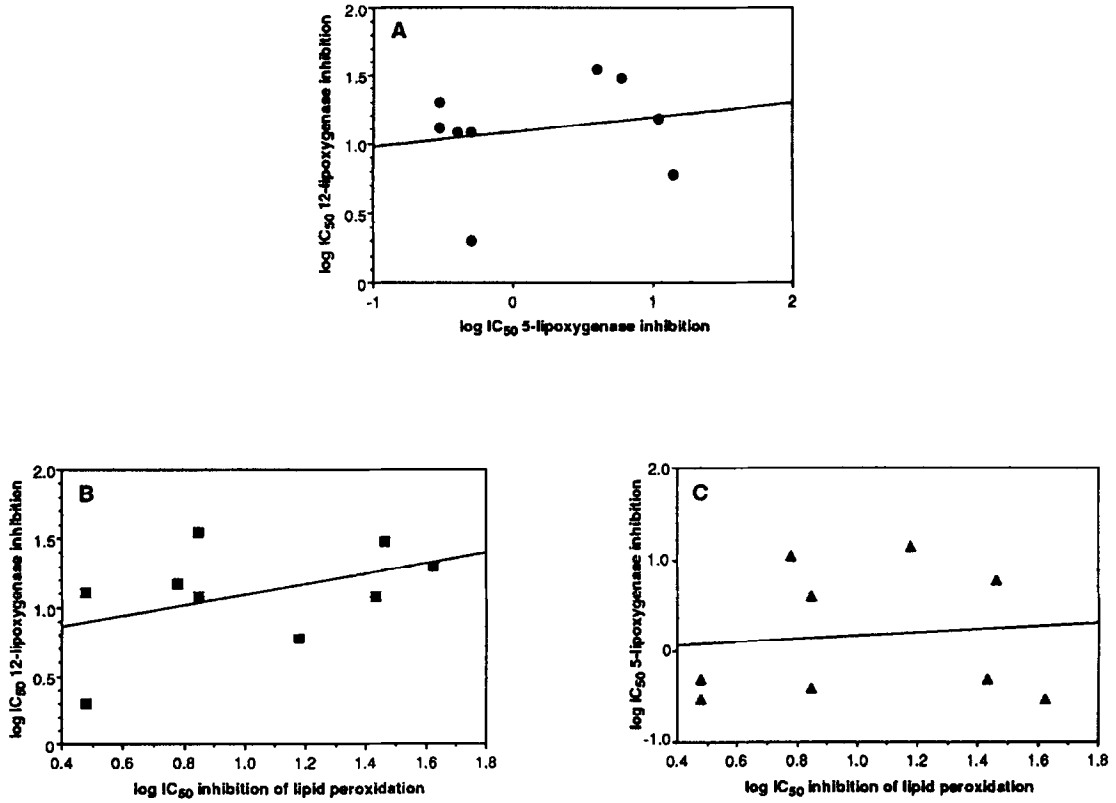

Fig. 3. Correlation plots of the inhibitory effects of anthrone derivatives against epidermal 12-LO compared with their inhibitory effects against PMNL 5-LO (A), and inhibitory effects against lipid peroxidation in phospholipid liposomes compared with their inhibitory effects against epidermal 12-LO (B), and PMNL 5-LO (C). The activities are shown on logarithmic scales in terms of the IC_{50} values (μM).

with the standard NDGA (IC_{50} = 2 μM). All novel compounds were far more powerful inhibitors of peroxidation than anthralin, which actually stimulated lipid peroxidation in this system [9].

Next, the antioxidant activities of the compounds as measured in the liposomal system and their ability to inhibit either 12-LO or 5-LO were compared, because inhibition of LO was expected to correlate with the potency of anti-lipid peroxidation activity [37]. Once again, however, a relationship was not found. Neither plots of the log IC_{50} for inhibition of nonenzymatic lipid peroxidation versus log IC_{50} for 12-LO inhibition (Fig. 3B) nor those versus the log IC_{50} for 5-LO inhibition (Fig. 3C) supported any relationship (r = 0.19 and 0.01, respectively).

Inhibition of lipid peroxidation in erythrocyte ghosts

Several anthrone derivatives also inhibit lipid peroxidation in erythrocyte ghosts. On the basis of its relative simplicity, the erythrocyte was selected as a source material for study of anthrone-induced damage to membrane substances. Erythrocyte ghosts undergo rapid lipid peroxidation when incubated in the presence of the radical initiator AAPH. The protecting activity of both the 10-phenylacyl and the 10-phenylalkylidene substituted derivatives increased with the number of free phenolic groups in the attached aromatic ring, the pyrogallol derivatives **2d** and **3c** being the most effective representatives of each series (Table 1). Furthermore, the protection by the 10-phenylalkylidene derivatives was better than that of the 10-phenylacyl analogs. In general, the activities of the compounds as inhibitors of lipid peroxidation in erythrocyte ghosts roughly correlated with their corresponding activities in phospholipid liposomes. Surprisingly, although not being protective in the latter system, anthralin itself turned out to be an inhibitor in erythrocyte ghosts.

DISCUSSION

Because leukotriene B_4 is an important mediator of psoriasis, there has been considerable interest in the development of 5-LO inhibitors [29]. The compounds tested in the present study are derived from 2 chemical series of antipsoriatic anthrones with improved 5-LO inhibitory activity as compared to that of the commonly used anthralin [12]. However, 12-HETE has also been reported to be involved in the disease process. Accordingly, compounds that inhibit both 5- and 12-LO may be superior to individual enzyme inhibitors. The novel anthrones at 2–20 μM concentrations also effectively inhibited the production of 12-HETE in epidermal homogenates of mouse skin. It was anticipated that partially blocking the C-10 position of anthralin might provide agents with diminished redox activity [12]. This has been documented for compounds that lack phenolic groups in the attached aromatic ring [12]. In agreement with their inhibitory effects against 5-LO [12], the inhibitory action against epidermal 12-LO was not necessarily dependent on the presence of phenolic groups (e.g. compounds **2a** and **2e** were also potent inhibitors of 12-LO). However, it is suggested that the compounds possessing phenolic hydroxyl groups exhibited better 12-LO inhibitory activity.

Surprisingly, while the antipsoriatic drug anthralin showed selectivity for epidermal 12-LO versus both platelet 12-LO and leukocyte 5-LO (ratios of IC_{50} values are 0.2 and 0.18, respectively), most of our novel compounds were more selective 5-LO inhibitors. Nevertheless, their inhibitory effects against epidermal 12-LO were in the same order of magnitude as that of anthralin.

An unexpected observation of anthrone derivatives with phenolic hydroxyl groups in the attached aromatic ring was the enhanced formation of hydroxyl radicals as compared to nonphenolic analogs [12]. Even though these compounds exhibited prooxidant action against nonlipid molecules, such as deoxyribose [12], the results of the present study show that phenolic compounds such as **2c, 2d, 3a–c** all protect against lipid peroxidation. Accordingly, their antioxidant properties predominate in membrane lipids, as demonstrated in erythrocyte ghosts and phospholipid bilayers. This is of great importance, because psoriatic lesions are characterized by infiltration of PMNL [38]. The production of oxygen radicals by PMNL and macrophages leads to injury to the tissue and inflammation [39, 40], and a similar role of active oxygen species has been identified in psoriasis [41, 42]. Furthermore, it has been reported that superoxide anion production in fibroblasts obtained from involved and uninvolved areas of skin of psoriatic patients is increased [43]. Psoriatic epidermis has been shown to have increased activity of xanthine oxidase, an enzyme capable of generating superoxide radical [44]. When generated in excess, active oxygen species can peroxidize the lipid-rich membranes of the skin, a tissue that is particularly vulnerable to the effects of these species [45]. The increased exposure of the skin to prooxidants, together with an insufficient capacity of the antioxidative system to respond, plays an important role in the induction of psoriasis by exogenous factors [46]. Accordingly, compounds that inhibit initiation or propagation of these oxidative processes may prove beneficial in psoriasis.

It has been suggested that iron is responsible for the catalysis of lipid peroxidation [47]; it is also important for the LO reaction. Lipoxygenases contain a non-heme iron that is converted to the ferric form upon activation [48–50]. It may be expected that, similar to the oxidation process of anthralin, electron transfer from the anthralin anion [5] to the active ferric form of LO results in an inactivated enzyme (ferrous form), according to one proposed mechanism [51]. The so-called redox-active inhibitors are thought to exert their action by this mechanism [52]. Furthermore, the biosynthesis of LO products is a radical-based oxidation [53]. Consequently, many inhibitors often act by formation of a lower energy radical, thus, interfering with the intended pathway of the reaction. Moreover, the first step in the LO reaction may be thought of as an enzymatically assisted lipid peroxidation [54]. Accordingly, many LO inhibitors also inhibit lipid peroxidation [37, 55–57], acting by scavenging chain-propagating peroxyl free radicals [58]. Hence, it is widely assumed that LO inhibition by antioxidants is due to scavenging of similar radicals that are generated within the active site of the enzyme [59, 60]. However, we did not observe any correlation between either 12-LO or 5-LO inhibition and the antioxidant activity against lipid peroxidation. This suggests a more selective mechanism for enzyme inhibition, rather than a nonspecific redox process or antioxidant effect. This is also supported by the lack of correlation between 12-LO and 5-LO inhibitory effects. Thus, other factors, such as an appropriate geometry of the molecules when bound to

the active site of the enzymes, probably contribute to the interaction of the compounds with the enzymes.

Taken together, the data of this study illustrate that the selected 10-substituted anthrones are fairly potent inhibitors of epidermal 12-LO, in addition to exercising a strong inhibitory effect against PMNL 5-LO. Furthermore, the antioxidant properties of the compounds alone are not a prerequisite for potent inhibition of 12- and 5-lipoxygenase. Nevertheless, our finding that many 10-substituted anthrones inhibit lipid peroxidation both in model membranes and in erythrocyte ghosts suggests that these compounds may play a protective role against oxidative damage to psoriatic skin, in addition to their antiinflammatory 12-LO and 5-LO properties. These dual-purpose inhibitors have the combined inhibitory action against both enzymatic (12-LO and 5-LO) and non-enzymatic lipid peroxidation and may have a greater impact on the therapeutic need than individual enzyme inhibitors or antioxidants.

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