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New 3- and 4-hydroxyfuranones as anti-oxidants and anti-inflammatory agents

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Abstract—Two series of new furanones substituted by methylsulfonylphenyl or methylsulfamidophenyl moieties were found to protect against oxidation damage by inhibiting or quenching free radicals and reactive oxygen species in in vitro experiments. The effect on lipid peroxidation was also examined. In addition, we investigated the activity of products in two models of inflammation: phorbol ester-induced ear edema in mice and carrageenan-induced paw edema in rat. The most powerful compounds and with reducing activity against DPPH (IC_{50} = 1779 and 57 μM, respectively), superoxide anion quenching capacity (IC_{50} = 511 and 49 μM, respectively), lipid peroxidation inhibitory effect and anti-inflammatory properties (about 50–65% inhibition of edema at 200 mg/kg ip in both tests used) were selected for further pharmacological and toxicological tests because of their attractive profile for the treatment of inflammatory diseases.

1. Introduction

Free radicals are species of chemical entities, that are energically unstable and highly reactive. The occurrence of reactive oxygen species is a characteristic of normal aerobic organisms. They are generated by-products of multiple metabolic functions and are thus implicated in tissue damage seen in various pathological events such as atherosclerosis, myocardial infarction, asthma, cancer, liver disease, neurologic trauma…[1,2] Under normal conditions, aerobic organisms have developed anti-oxidant defences to protect themselves against free radicals: enzymes such as superoxide dismutase and glutathione peroxidase, and anti-oxidants and radical scavengers such as α-tocopherol (vitamin E), β-carotene, glutathione and ascorbic acid (vitamin C).[3] Ascorbate is one of the most potent naturally occurring anti-oxidants because it works directly by reaction with aqueous peroxyl radicals and indirectly by restoring the anti-oxidant properties of fat-soluble vitamin E.[4] Furthermore, it plays an important part in the prevention of a large number of chronic diseases.[5,6] However, the low solubility of ascorbic acid in lipophilic environments, as well as its susceptibility to thermal and oxidative degeneration, has led to interest in the synthesis of derivatives with better lipophilicity and increased stability.[7–17] The recent efforts devoted to the development of new ascorbic acid analogues have resulted in obtaining anti-oxidant,[18–22] antitumoural[23] and potentially anti-inflammatory[19,24] agents.

Concerning inflammation, free radicals are well known to play an important part in the inflammatory process. In the cyclooxygenase pathway, oxygen radicals are liberated during the conversion from PGG_2 to PGH_2,[25,26] and different reports have suggested that an enhancement of free radical generation may contribute to the development of inflammatory processes[27] and particularly to the pathogenesis of airway hyperactivity in asthma.[28,29] In these conditions, the use of drugs with both scavenging activity and anti-inflammatory properties may be of benefit in the treatment of airway inflammation and bronchial hyper-responsiveness.

In connection to our previous works[19,20] on the synthesis of lactone derivatives **1**, **2**, **3** (Chart 1) related to

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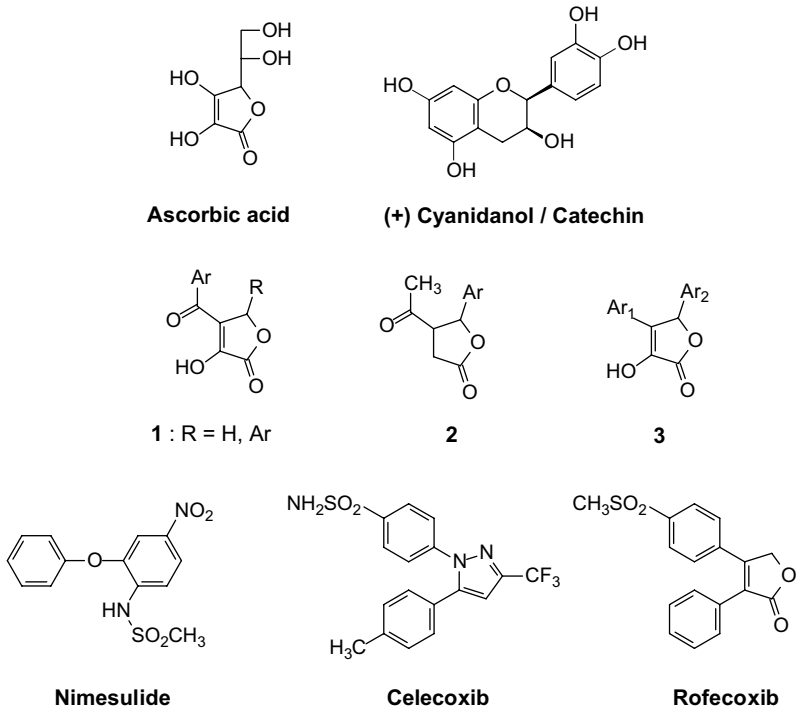


Chart 1.

ascorbic acid, we have found out that some 4,5-diaryl-3-hydroxy-2(5*H*)-furanones **3** possess significant anti-inflammatory and anti-oxidant activities. These results prompted us to design and synthesize new furanones substituted by methylsulfonylphenyl or methylsulfamidophenyl moieties typical of potent non-steroidal anti-inflammatory drugs (NSAIDs) such as rofecoxib nimesulide and celecoxib (Chart 1).

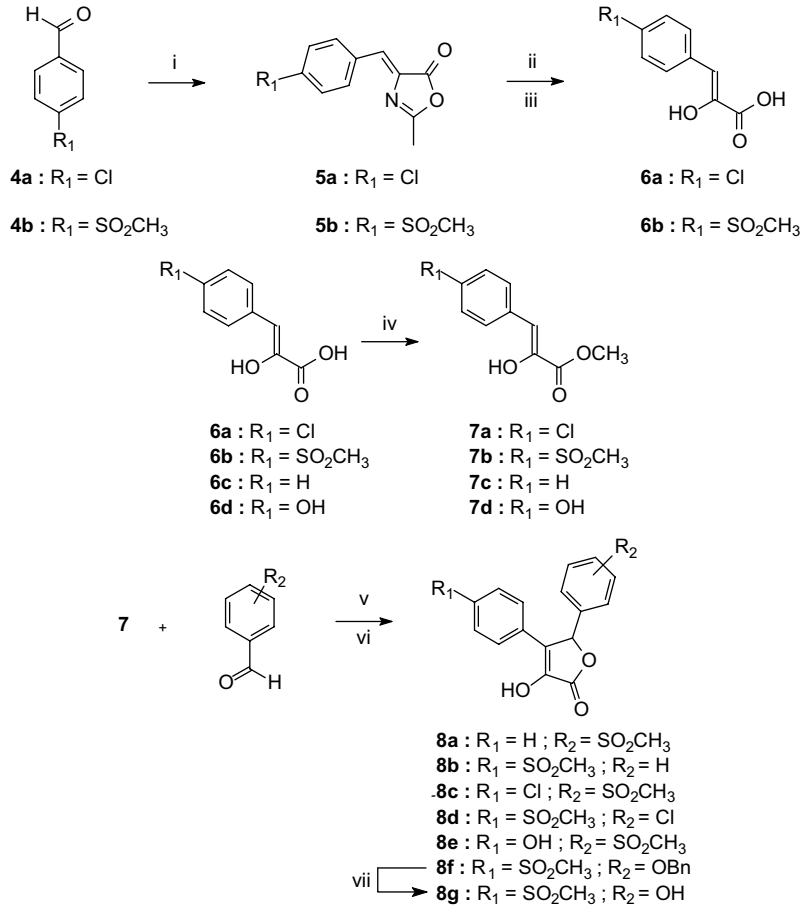
Thus, the anti-oxidant effects of these new derivatives have been carried out in vitro by means of three tests: measurement of the reducing activity on the stable 2,2-diphenyl-1-picrylhydrazyl radical (DPPH), superoxide anion scavenging and lipid peroxidation tests. Furthermore, anti-inflammatory activities have been studied by use of two tests: phorbol 12-myristate 13-acetate (TPA)-induced ear edema test on the mouse and a model of carrageenan-induced paw edema on the rat.

2. Chemistry

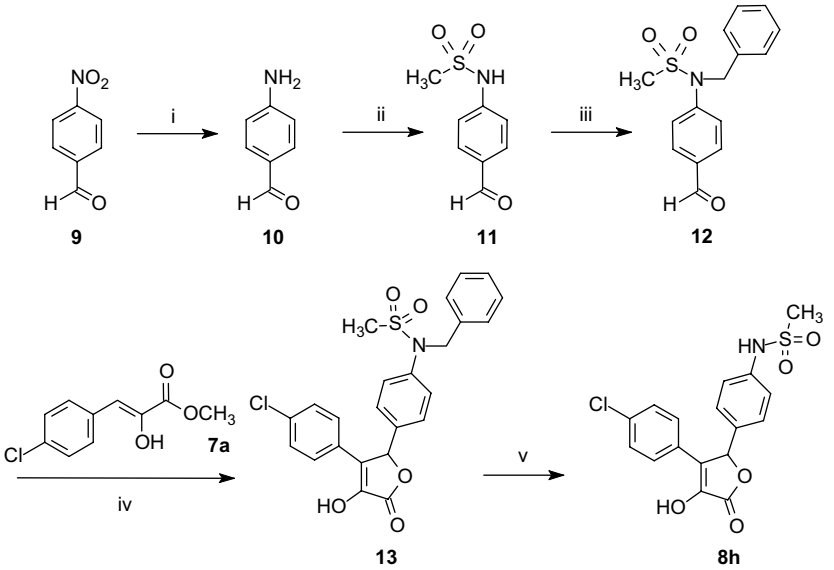
For the synthesis of compounds **8** possessing methylsulfonyl substituents on the phenyl rings, we adopted the synthetic methods described in a previous communication.[19] Preparation of 4,5-diaryl-3-hydroxyfuranones **8a**–**g** was obtained by lactonization after aldol condensation of methyl arylpyruvates **7** with appropriate benzaldehydes in the presence of 1,5-diazabicyclo[5.4.0]undecene (DBU) in dimethylformamide,[30–32] as shown in Scheme 1. The requisite α-ketoesters **7a**,**b** were readily accessible from the commercially available benzaldehydes **4a**,**b** through the three-step sequence depicted in Scheme 1 via azlactones **5a**,**b**. Thus, the condensation of the appropriately substituted benzaldehydes with *N*-acetylglycine and sodium acetate in refluxing acetic anhydride[33] furnished the crystalline oxazolones **5a**,**b** in high yield. The oxazolones were converted to the corresponding phenylpyruvic acids **6a**,**b** through a two-step hydrolysis with aqueous sodium hydroxide followed by aqueous hydrochloric acid. Methyl arylpyruvates **7** were prepared by methylating arylpyruvic acids **6** using DBU (1 equiv) and MeI (5 equiv) in DMF according to the procedure described by Namiki et al.[30]

In a similar way, the synthesis of derivative **8h** (Scheme 2) was achieved by the same addition/cyclization process starting from the pyruvate **7a** and the sulfonamido aldehyde **12**. This intermediate **12** was obtained from the commercially available 4-nitrobenzaldehyde **9** in three steps comprising the formation of amine **10** according to the method of Bellamy,[34] the sulfonamide **11** and final benzylation. As mentioned above, the addition of **12** to the appropriate pyruvate yielded furanone **13**. Final debenzylation was achieved through hydrogenolysis in the presence of Pd/C to give the target furanone **8h**.

The various 4-hydroxy-2(5*H*)-furanones **18** were synthesized via alkaline cyclization of ethoxycarbonylmethyl phenylacetates **17** using sodium hydride in dry THF (Scheme 3). Intermediates **17** were obtained by the condensation of a phenylacetic acid derivative **14** with a suitable α-bromo carboxylic ester **16** in the presence of triethylamine. Bromo compounds **16** were prepared in two steps starting from phenylacetic acids **14a**–**c** after esterification to **15** followed by bromina-



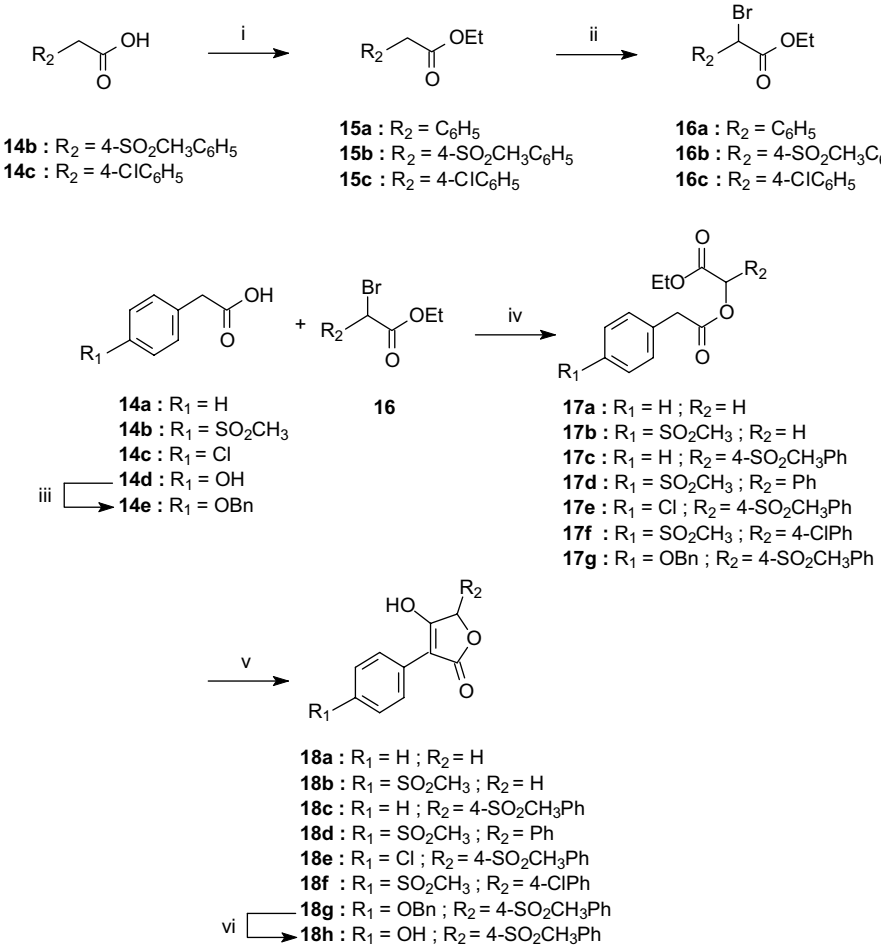
Scheme 1. Reagents and conditions: (i) *N*-acetylglycine, Ac_2O, AcONa, 100 °C; (ii) NaOH; (iii) HCl, reflux; (iv) DBU, CH_3I, DMF, 0 °C; (v) DBU, DMF, 0 °C; (vi) HCl; (vii) Pd/C, H_2, THF/EtOH.



Scheme 2. Reagents and conditions: (i) $SnCl_2$, EtOH; (ii) CH_3SO_2Cl, pyridine; (iii) BnCl, NEt_3, DMF, 80 °C; (iv) DBU, DMF, 0 °C; (v) Pd/C, H_2, THF/EtOH.

tion. All phenylacetic acids **14** were commercially available except **14e**, which was obtained by benzylation of **14d**.[35]

The structural determination and purity of furanones **8** and **18** were achieved by IR, ^1H and ^{13}C NMR. All compounds were obtained as racemates.



Scheme 3. Reagents and conditions: (i) DBU, CH_3CH_2I, DMF, 0 °C; (ii) NBS, $(PhCOO)_2$, CCl_4, reflux; (iii) BnCl, K_2CO_3, KI, DMF, reflux; (iv) NEt_3, THF, reflux; (v) NaH, THF; (vi) Pd/C, H_2, THF/EtOH.

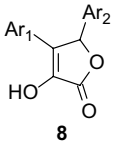
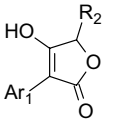
3. Results and discussion

3.1. In vitro anti-oxidant activities

As anti-oxidant action is considered to be a complex process, which may include preventing of both the formation or scavenging of free radicals, it was of interest to investigate the interaction of the synthesized compounds with the stable free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH). This test is representative of the capacity of compounds to scavenge free radicals independently from any enzymatic activity. With IC_{50} values ranging from 9.2 to 1779 μM (Table 1), all derivatives were found to interact with DPPH but the variant degree of anti-oxidant effect shown by the products cannot be directly related to their structure. The position of hydroxy substituent on furanone ring (**8** vs **18**) significantly affect reducing activity on DPPH, the 4-position being preferred. In series **8**, it appeared that the 4-methylsulfonylphenyl group was more favourable to activity when attached to the phenyl nucleus in the 5-position of the olide ring (**8a**,**c**,**e** vs **8b**,**d**,**g**), which was not so evident in the series **18** (**18c**,**e** vs **18d**,**f**). Nevertheless, a 4-hydroxyphenyl moiety as the Ar_1 substituent furnished in both series the most efficient compounds **8e** (IC_{50} = 20.1 μM) and **18h** (IC_{50} = 9.2 μM), displaying the importance of a phenol group for anti-oxidant activity. Finally, compared with conventional anti-oxidants, compounds **8e** and **18h** showed activity, respectively, equipotent or two times superior to vitamin C (IC_{50} = 21.0 μM) and vitamin E (IC_{50} = 22.1 μM), but less important than (+)-catechin, which is a flavonoid, a class of molecules well known to have radical-scavenging properties.[36,37]

The first product of the univalent reduction of oxygen is the superoxide anion $O_2^{\cdot -}$, which is generated in many biological processes.[38,39] As a result, compounds able to scavenge $O_2^{\cdot -}$ could be protecting agents against cellular injury during the reperfusion of ischemic tissues. Therefore, the superoxide anion scavenging activity of furanones **8** and **18** was explored using a non-enzymatic biological generator of superoxide anion radical, for instance, the phenazine methosulfate NADH system.[40] This test was limited by the solubility of products in aqueous solution. In the superoxide anion scavenging test (Table 1), the presence of a hydroxyl group in the 4-position on the olide ring is favourable to activity compared with the 3-position (**18** vs **8**). Compounds bearing a 4-methylsulfonylphenyl group as Ar_1 (**8b**,**d**,**g**) were more active, or as active as, the analogues **8a**,**c**,**e** in contrast to the observations made in the previous test.

Table 1. Anti-oxidant activities of derivatives **8** and **18**

	Compd	Ar_1	Ar_2 or R_2	Reducing activity on DPPH IC_{50} (μM)	Superoxide anion scavenging activity IC_{50} (μM)	Lipid peroxidation IC_{50} (μM) or % of inhibition
 8	**8a**	C_6H_5	4-$SO_2CH_3C_6H_4$	530 ± 25	780 ± 127	22 ± 5[a]
	8b	4-$SO_2CH_3C_6H_4$	C_6H_5	1570 ± 169	589 ± 78	21 ± 3[a]
	8c	4-ClC_6H_4	4-$SO_2CH_3C_6H_4$	383 ± 27	160 ± 8	NT
	8d	4-$SO_2CH_3C_6H_4$	4-ClC_6H_4	1014 ± 80	122 ± 7	37 ± 8[a]
	8e	4-OHC_6H_4	4-$SO_2CH_3C_6H_4$	20.1 ± 0.6	488 ± 19	91.9 ± 2.3
	8g	4-$SO_2CH_3C_6H_4$	4-OHC_6H_4	1779 ± 161	511 ± 76	123 ± 29
	8h	4-ClC_6H_4	4-$NHSO_2CH_3C_6H_4$	700 ± 40	NT	NT
 18	**18a**	C_6H_5	H	87 ± 2	18 ± 15[b]	21 ± 4[a]
	18b	4-$SO_2CH_3C_6H_4$	H	238 ± 6	1628 ± 199	30 ± 5[b]
	18c	C_6H_5	4-$SO_2CH_3C_6H_4$	119 ± 18	242 ± 13	16 ± 4[a]
	18d	4-$SO_2CH_3C_6H_4$	C_6H_5	212 ± 3	97 ± 4	3 ± 1[a]
	18e	4-ClC_6H_4	4-$SO_2CH_3C_6H_4$	146 ± 4	61 ± 31	32 ± 4[b]
	18f	4-$SO_2CH_3C_6H_4$	4-ClC_6H_4	57 ± 1	49 ± 6	22 ± 5[b]
	18h	4-OHC_6H_4	4-$SO_2CH_3C_6H_4$	9.2 ± 0.4	236 ± 55	169 ± 57
	Ascorbic acid			21.0 ± 0.4	24.0 ± 1.5[c]	Pro-oxidant
	Vitamin E			22.1 ± 2.2	NT	NT
	(+)Catechin			7.5 ± 0.3	176 ± 10	13.8 ± 3

NT: not tested.
[a] % inhibition at 0.25 mg/mL.
[b] % inhibition at 0.5 mg/mL.
[c] % inhibition at 1 mg/mL.

This time the same analysis can be made with the series **18** (**18d**,**f** vs **18c**,**e**). Except for compounds **18a** and **18b**, all tested 4-hydroxyfuranones **18** showed a noteworthy superoxide anion scavenging activity with IC_{50} values ranging from 49 to 242 μM. The presence of a phenyl nucleus in the 5-position on the olide appeared to be essential for the efficiency of furanones **18** (**18a**,**b** vs **18c**–**h**). In addition, compounds **18d**,**e**,**f** displayed a better activity than previously reported 3-hydroxyfuranones **3**[19] and (+)-catechin with IC_{50} values of, respectively, 97, 61 and 49 μM, and even better than ascorbic acid with only 24% inhibition at a concentration of 1 mg/mL. Vitamin E was not tested due to its insolubility in aqueous medium.

In order to further investigate the radical-trapping anti-oxidant behaviour of compounds **8** and **18**, and so their ability to prevent continued allylic hydrogen abstraction from lipids, their effects were evaluated on the Fe^{2+}/ADP-ascorbate induced lipid peroxidation of rat liver microsomes. The autoxidation of Fe^{2+}/ADP causes a massive lipid peroxidation in liver microsomes, which can be followed by the formation of malondialdehyde. Only derivatives **8e**,**g** and **18h** produced a noteworthy activity while other compounds were almost ineffective or could not be tested due to their poor solubility in the biological reaction mixture (**8c**,**h**). In this test, as previously reported,[41–43] vitamin C underwent an autoxidative destruction in the presence of iron initiating lipid peroxidation. However, this 'prooxidant' activity in vitro has obviously no significance in vivo and ascorbic acid works as an anti-oxidant under most circumstances.[42,43] All compounds are less effective than (+)-catechin, which is an efficient chain-breaking anti-oxidant with a good anti-oxidant activity in microsomal lipid peroxidation.[44,45]

Considering their activity in the three tests used, only derivatives **8e**,**g** and **18f**,**h** with multiple mechanisms of protective action appeared useful to minimize tissue injury in human disease. Indeed, they were able to scavenge free radicals and/or to trap $O_2^{\cdot -}$, preventing its conversion into H_2O_2. Furthermore, they could contribute to reducing the devastating effects of lipid peroxidation, taking into account their ability to scavenge peroxyl and/or hydroxyl radicals (except **18f**).

3.2. In vivo anti-inflammatory activities

Reactive oxygen species are well known to be critical mediators of inflammation and tissue damage.[46–48] Therefore, anti-oxidants could be beneficial in the treatment of some acute and chronic inflammatory diseases. Furthermore, in addition to a well recognized inhibitory effect on the cyclooxygenase and lipoxygenase activities, NSAIDs have been reported to affect a variety of other processes and above all to possess anti-oxidant properties.[49–53] By the structural modifications of previously described furanones[19] consequent to the incorporation of sulfonyl and methylsulfamido moieties present in nimesulide and coxibs for instance (Chart 1), we aimed at enhancing anti-inflammatory properties in compounds **8** and/or **18**, which were thus investigated in this area.

First, behavioural effects and intraperitoneal acute toxicities of compounds **8** and **18** were examined in mice. Neither of the tested drugs produced significant behavioural effects, even at doses up to 800 mg/kg ip and all the animals were still alive after a one week observation period.

With regard to the evaluation of furanones **8** and **18** as anti-inflammatory compounds, they were first adminis-

Table 2. Anti-inflammatory activity of derivatives **8** and **18**

Compound	TPA-induced ear edema test % inhibition of edema at		Carrageenan-induced paw edema test % inhibition of edema at
	100 mg/kg ip	200 mg/kg ip	200 mg/kg ip
8a	40.4 ± 11.1[a]	88.0 ± 5.5[b]	45.1 ± 6.1[a]
8b	39.2 ± 6.2[b]	38.3 ± 8.0[a]	NT
8c	42.4 ± 11.0[a]	88.1 ± 4.1[b]	61.7 ± 3.5[b]
8d	57.0 ± 6.1[b]	81.0 ± 8.4[b]	49.7 ± 6.2[a]
8e	7.8 ± 7.4 (NS)	12.1 ± 8.6 (NS)	NT
8g	40.2 ± 9.1[a]	65.7 ± 7.3[b]	64.1 ± 11.1[b]
8h	1.0 ± 7.8 (NS)	3.5 ± 2.9 (NS)	NT
18a	33.9 ± 8.0[a]	61.2 ± 9.1[b]	29.1 ± 9.0[a]
18b	7.3 ± 5.1 (NS)	9.5 ± 6.7 (NS)	NT
18c	11.1 ± 8.1 (NS)	13.4 ± 9.8 (NS)	NT
18d	10.1 ± 8.6 (NS)	15.6 ± 6.2 (NS)	NT
18e	12.2 ± 7.5 (NS)	16.2 ± 8.4 (NS)	NT
18f	47.3 ± 11.8[a]	78.4 ± 6.3[b]	50.8 ± 1.5[b]
18h	20.3 ± 5.2[a]	46.4 ± 8.3[b]	NT
Rofecoxib	12.4 ± 9.5 (NS)	14.8 ± 7.1 (NS)	52.3 ± 6.1[b,c]
Ketorolac	10.6 ± 8.2 (NS)	14.6 ± 6.4 (NS)	53.3 ± 9.1[b,c]
Dexamethasone	93.3 ± 2.0[b,d]	NT	76.5 ± 5.4[b,d]

NT: not tested. NS: not significant.
[a] $0.001 < p < 0.05$.
[b] $p < 0.001$.
[c] Tested at 15 mg/kg ip.
[d] Tested at 1 mg/kg ip.

tered ip at doses of 100 and 200 mg/kg in the phorbol 12-myristate 13-acetate (TPA)-induced ear edema test (Table 2). At 100 mg/kg ip, only compounds **8a**,**c**,**d**,**g** and **18f** revealed a noteworthy activity and produced more than 40% inhibitory effects. Anti-inflammatory effects were dose-dependent since **8a**,**c**,**d**,**g** and **18a**,**f** were particularly effective in reducing TPA inflammation at 200 mg/kg with more than 60% activity. However, the influence of substituents on anti-inflammatory properties did not clearly appear: it seemed that a hydroxy group on aromatic ring Ar_1 was not absolutely necessary, as shown by the lower activity of **8e** versus **8a** and **8c**. Furthermore, the position of the methylsulfonylphenyl moiety on the furan skeleton of **8** did not significantly affect activity in this test. Contrary to compound **8c** showing a good anti-inflammatory efficiency with 88% of inhibition at 200 mg/kg ip in the TPA test, furanone **8h**, bearing a 4-methylsulfamido group on Ar_2, revealed no activity, suggesting that this moiety is not favourable to anti-inflammatory activity compared to a methylsulfonyl group. Surprisingly, among furanones **18**, the 4-methylsulfonyl group on Ar_1 and 4-chloro substituent on Ar_2 provided the most efficient derivative **18f**, while the potency of the same substituted furanones on Ar_1 (**18b**,**d**) was dramatically reduced in comparison with that of parent compound **18a**. Clearly, 3-hydroxyfuranones **8** were more active in diminishing TPA inflammation than 4-hydroxyfuranones **18**. Consistent with the literature data,[54] classical NSAIDs rofecoxib and ketorolac failed to produce any significant effect after ip administration, since the maximum reduction of ear thickness was only 13.8% at 100 mg/kg when steroid dexamethasone proved a 93.3% inhibition at 1 mg/kg ip. Indeed, it has been well established by now that TPA produces a long-lasting edema that is associated with a marked influx of neutrophils along with the predominant formation of leukotriene LTB4.[55,56] So it seems quite natural that, in this model, cycloxygenase inhibitors show lower anti-inflammatory activity than dexamethasone, which is well known for inhibiting phospholipase A2 and so leukotriene release.

The in vivo anti-inflammatory efficiency of furanones **8a**,**c**,**d**,**g** and **18a**,**f** was further completed using the functional model of carrageenan-induced paw edema in the rat, a test particularly efficient in detecting compounds whose activity is the result of the inhibition of prostaglandin amplification.[57] Except with the derivative **18a**, all tested drugs were indeed effective at 200 mg/kg ip. Furanones **8c**,**g** and **18f** produced 50–65% of anti-inflammatory activity and were equipotent to NSAID administered at the low dose of 15 mg/kg ip.

So, we can see that some furanones (**8a**,**c**,**d**,**g** and **18f**) were effective in these two models and these results suggest that their anti-inflammatory effects may be due, like steroids, to the inhibition of both cycloxygenase and 5-lipoxygenase pathways.

4. Conclusion

Although it was difficult, conflicting even with these series of furanones, to gather in the same structure anti-oxidant and anti-inflammatory properties, appropriate modifications of previous derivatives **3** led to active products in both areas. However, regarding all the data only furanones **8g** and **18f** showed significant and potent biological effects in the different tests used, associated to a large therapeutic index in view of their LD_{50} superior to 800 mg/kg ip. The presence of two aromatic nuclei on the olide ring confers a high lipophilicity to the mole-

cules, which may contribute to their effective access, retention and interaction with biological membranes. If we extrapolate these in vitro results to an in vivo situation, we can assume that **8g** and **18f** can interfere at distinct levels in the radical chain reaction, thus exerting a synergistic effect in mitigating tissue damage that occurs during inflammatory disease. Thus **8g** and **18f** were selected for further pharmacological and toxicological tests since they have an attractive profile as anti-oxidant and anti-inflammatory agents.

5. Experimental

5.1. General chemical methods

All starting materials used for synthesis were reagent-grade from Acros (Noisy-le-Grand, France). Reagents for biochemical tests were purchased from Sigma (Montluçon, France). Melting points were determined on a Reichert apparatus (Isi, Paris, France) and were uncorrected. Column chromatography was carried out on SDS silica gel 60 (70–200 mesh). The infrared (IR) spectra were recorded with a FTIR-Nicolet Impact 410 spectrophotometer (Thermo Optek, Montigny le Bretonneux, France). The proton and carbon nuclear magnetic resonance (^{1}H NMR and ^{13}C NMR) spectra were recorded on a Brüker AC-400 (400 MHz) spectrometer (Brüker, Wissembourg, France). Chemical shifts are reported in parts per million (δ ppm) relative to tetramethylsilane used as an internal standard. The abbreviations used for signal patterns are: br s, broad singlet; s, singlet; d, doublet; m, multiplet. Mass spectral analyses were performed on a Hewlett-Packard 5985B or 5989A instrument. The ultra violet (UV) measurements were carried out with a DU-70 spectrometer (Beckman Instruments, Fullerton, USA).

5.2. General procedure for preparation of 4-benzylidene-2-methyl-5-oxazolones (5)

The general procedure of the oxazolone preparation was adapted from the literature procedure.[33] A solution of the appropriate benzaldehyde (8.14 mmol) was dissolved in 8 mL of acetic anhydride. *N*-Acetylglycine (0.95 g, 8.14 mmol) was added, followed by sodium acetate (0.67 g, 8.14 mmol), and the mixture was heated to 100 °C in an oil bath for 4–5 h. The reaction mixture was allowed to slowly cool overnight. Meanwhile, the azlactone precipitated as a yellow/orange solid, which was isolated by filtration. Washing with cold diethyl ether afforded quantitative azlactone of suitable purity for use in subsequent syntheses.

5.2.1. 4-(4-Chloro)benzylidene-2-methyl-5-oxazolone (5a).[58] Yield 98%.

5.2.2. 4-(4-Methylsulfonyl)benzylidene-2-methyl-5-oxazolone (5b). Yield 95%; IR (KBr) ν 1805, 1302, 1146 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 8.27 (d, 2H, H_{Ar}), 8.01 (d, 2H, H_{Ar}), 7.14 (s, 1H, CH), 3.09 (s, 3H, SO_2CH_3), 2.46 (s, 3H, CH_3).

5.3. General procedure for preparation of phenylpyruvic acids (6)

The general procedure was adapted from the literature procedure.[33] A suspension of azlactone **5** (7.54 mmol) in 12 mL of 1.0 N sodium hydroxide solution was heated at 90 °C until homogeneous. The solution was acidified to pH 1–2 with a hydrochloric acid solution 3 N and a precipitate was formed. Concentrated hydrochloric acid (2 mL) was added, followed by water (15 mL) and the mixture was heated to reflux for 6 h. The mixture was cooled in an ice cold bath, and the precipitate thus formed was isolated by filtration. Washing with water and drying under reduced pressure produced phenylpyruvic acids **6** of suitable purity for use in subsequent operations.

5.3.1. 3-(4-Chlorophenyl)-2-oxo-propionic acid (6a). Yield 43%; mp 188 °C (lit., 182 °C);[59] IR (KBr) ν 3465, 3200–2500, 1664 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 13.34 (br s, 1H, OH), 9.51 (br s, 1H, OH), 7.81 (d, 2H, H_{Ar}), 7.41 (d, 2H, H_{Ar}), 6.44 (s, 1H, CH); ^{13}C NMR (DMSO-d_6) δ 166.3, 142.6, 134.0, 131.5, 130.9, 120.4, 108.3.

5.3.2. 3-(4-Methylsulfonylphenyl)-2-oxo-propionic acid (6b). Yield 50%; mp 249–250 °C; IR (KBr) ν 3445, 3200–2400, 1684, 1304, 1149 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 13.60 (br s, 1H, OH), 9.96 (br s, 1H, OH), 8.02 (d, 2H, H_{Ar}), 7.91 (d, 2H, H_{Ar}), 6.51 (s, 1H, CH), 3.23 (s, 3H, CH_3); ^{13}C NMR (DMSO-d_6) δ 165.9, 144.6, 140.3, 138.4, 129.5, 127.0, 107.4, 43.6.

5.4. General procedure for preparation of 4,5-diaryl-3-hydroxy-2(5*H*)-furanones (8)

DBU (0.80 mL, 5.35 mmol) was added under stirring to a cold (0 °C) solution of a mixture of methyl 2-oxo-3-phenylpropionate **7** (5.27 mmol) and appropriate benzaldehyde (5.27 mmol) in dry DMF (24 mL). The mixture was stirred for 3–5 h at 0 °C and then poured into a mixture of ethyl acetate (10 mL) and HCl 1 M (50 mL). The organic layer was separated and the aqueous layer was extracted with AcOEt. The organic layers were combined, washed with water, dried over Na_2SO_4 and evaporated in vacuo. The oily residue was then purified either by column chromatography on silica gel or by precipitation in water, filtration and crystallization from ethyl ether.

5.4.1. 3-Hydroxy-5-(4-methylsulfonylphenyl)-4-phenyl-2(5*H*)-furanone (8a). This was prepared from methyl 2-oxo-3-phenylpropionate **7c** and 4-methylsulfonylbenzaldehyde **4b**. Precipitation in water and crystallization from ethyl ether led to compound **8a** in 73% yield; mp 215 °C; IR (KBr) ν 3314, 1762, 1292, 1143 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 11.27 (br s, 1H, OH), 7.94 (d, 2H, H_{Ar}), 7.72 (d, 2H, H_{Ar}), 7.65 (d, 2H, H_{Ar}), 7.38–7.26 (m, 3H, H_{Ar}), 6.75 (s, 1H, H-5), 3.21 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.9, 142.2, 141.6, 139.1, 130.2, 128.9, 128.7, 127.9, 127.8, 127.4, 78.6, 43.4; EI-MS *m*/*z* 330 [M]$^+$.

5.4.2. 3-Hydroxy-4-(4-methylsulfonylphenyl)-5-phenyl-2(5*H*)-furanone (8b). This was prepared from methyl

2-oxo-3-(4-methylsulfonylphenyl)propionate **7b** and benzaldehyde. Purification by column chromatography on silica gel, eluting with EtOAc/petroleum benzine (5/5), produced compound **8b** in 56% yield; mp 192–193 °C; IR (KBr) ν 3278, 1740, 1311, 1149 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.77 (br s, 1H, OH), 7.93 (d, 2H, H_{Ar}), 7.87 (d, 2H, H_{Ar}), 7.48–7.39 (m, 5H, H_{Ar}), 6.68 (s, 1H, H-5), 3.21 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.6, 141.1, 139.7, 136.0, 135.3, 130.2, 129.1, 127.9, 127.8, 127.1, 125.5, 79.6, 43.3; EI-MS *m/z* 330 $[M]^+$.

5.4.3. 4-(4-Chlorophenyl)-3-hydroxy-5-(4-methylsulfonylphenyl)-2(5*H*)-furanone (8c). This was prepared from methyl 2-oxo-3-(4-chlorophenyl)propionate **7a** and 4-methylsulfonylbenzaldehyde **4b**. Precipitation in water and crystallization from ethyl ether led to compound **8c** in 66% yield; mp 260 °C; IR (KBr) ν 3306, 1743, 1312, 1150 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.49 (s, 1H, OH), 7.97 (d, 2H, H_{Ar}), 7.74 (d, 2H, H_{Ar}), 7.67 (d, 2H, H_{Ar}), 7.47 (d, 2H, H_{Ar}), 6.77 (s, 1H, H-5), 3.25 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.6, 141.8, 141.6, 139.5, 133.0, 129.0, 128.9, 128.7, 127.7, 126.4, 78.3, 43.3; EI-MS *m/z* 364 $[M]^+$.

5.4.4. 5-(4-Chlorophenyl)-3-hydroxy-4-(4-methylsulfonylphenyl)-2(5*H*)-furanone (8d). This was prepared from methyl 2-oxo-3-(4-methylsulfonylphenyl)propionate **7b** and 4-chlorobenzaldehyde **4a**. Purification by column chromatography on silica gel, eluting with EtOAc/petroleum benzine (5/5), gave compound **8d** in 78% yield; mp 199 °C; IR (KBr) ν 3466, 1757, 1308, 1151 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.83 (br s, 1H, OH), 7.93 (d, 2H, H_{Ar}), 7.86 (d, 2H, H_{Ar}), 7.52–7.47 (m, 4H, H_{Ar}), 6.71 (s, 1H, H-5), 3.22 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.5, 141.3, 139.7, 135.1, 135.0, 134.1, 129.9, 129.1, 127.7, 127.2, 125.2, 78.7, 43.3; EI-MS *m/z* 364 $[M]^+$.

5.4.5. 3-Hydroxy-4-(4-hydroxyphenyl)-5-(4-methylsulfonylphenyl)-2(5*H*)-furanone (8e). This was prepared from methyl 2-oxo-3-(4-hydroxyphenyl)propionate **7d** and 4-methylsulfonylbenzaldehyde **4b**. Purification by column chromatography, eluting with EtOAc/petroleum benzine (5/5), gave compound **8e** in 47% yield; mp 241–242 °C; IR (KBr) ν 3390, 3340, 1712, 1314, 1155 cm^{-1}; ^1H NMR (DMSO-d_6) δ 10.81 (s, 1H, OH), 9.87 (s, 1H, OH), 7.98 (d, 2H, H_{Ar}), 7.71 (d, 2H, H_{Ar}), 7.52 (d, 2H, H_{Ar}), 6.78 (d, 2H, H_{Ar}), 6.67 (s, 1H, H-5), 3.25 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 169.1, 157.9, 142.5, 141.4, 136.7, 129.1, 129.0, 128.8, 127.7, 121.1, 115.5, 78.4, 43.3; EI-MS *m/z* 346 $[M]^+$.

5.4.6. 5-(4-Benzyloxyphenyl)-3-hydroxy-4-(4-methylsulfonylphenyl)-2(5*H*)-furanone (8f). This was prepared from methyl 2-oxo-3-(4-methylsulfonylphenyl)propionate **7b** and 4-benzyloxybenzaldehyde.[60] Purification by column chromatography, eluting with a gradient of EtOAc/cyclohexane, gave compound **8f** in 62% yield; mp 154 °C; IR (KBr) ν 3181, 1760, 1296, 1142 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.69 (br s, 1H, OH), 7.86 (m, 4H, H_{Ar}), 7.40–7.37 (m, 7H, H_{Ar}), 7.01 (d, 2H, H_{Ar}), 6.59 (s, 1H, H-5), 5.06 (s, 2H, PhCH_2O), 3.19 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.7, 159.1, 141.1, 139.6, 136.8, 135.4, 129.4, 128.4, 128.0, 127.9, 127.8, 127.1, 125.3, 115.2, 79.4, 68.3, 43.3.

5.4.7. 3-Hydroxy-5-(4-hydroxyphenyl)-4-(4-methylsulfonylphenyl)-2(5*H*)-furanone (8g). Pd/C (10%, 0.30 g) was added to 5-(4-benzyloxyphenyl)-3-hydroxy-4-(4-methylsulfonylphenyl)-2(5*H*)-furanone **8f** (1.37 g, 3.14 mmol) dissolved in THF/EtOH (10/30 mL). The reaction mixture was then submitted to a hydrogen atmosphere for 7 h. After filtration of the catalyst and evaporation of the solvent, crystallization from ethyl ether produced 0.96 g (88%) of **8g**; mp 230 °C; IR (KBr) ν 3271, 1734, 1312, 1148 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.64 (br s, 1H, OH), 9.71 (br s, 1H, OH), 7.90 (d, 2H, H_{Ar}), 7.82 (d, 2H, H_{Ar}), 7.22 (d, 2H, H_{Ar}), 6.76 (d, 2H, H_{Ar}), 6.52 (s, 1H, H-5), 3.18 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.7, 158.4, 141.1, 139.6, 135.5, 129.5, 127.8, 127.1, 126.0, 125.3, 115.8, 79.8, 43.3; EI-MS *m/z* 346 $[M]^+$.

5.4.8. *N*-(4-Formylphenyl)methylsulfonamide (11). Methanesulfonyl chloride (2 mL, 25.8 mmol) was added to a 0 °C solution of **10** (3 g, 24.8 mmol) in pyridine (30 mL) and the reaction solution was allowed to remain at this temperature under stirring for 3 h. Most of the pyridine was evaporated under reduced pressure and the concentrated solution was diluted in ethyl acetate. Dilute HCl (1 N) was added, and the organic layer was separated, washed with water, then brine, dried over Na_2SO_4 and evaporated in vacuo. Crystallization from diethyl ether produced 2.68 g (54%) of the title compound; IR (KBr) ν 3242, 1674, 1328, 1145 cm^{-1}; ^1H NMR (DMSO-d_6) δ 10.53 (s, 1H, NH), 9.92 (s, 1H, CHO), 7.91 (d, 2H, H_{Ar}), 7.39 (d, 2H, H_{Ar}), 3.18 (s, 3H, SO_2CH_3).

5.4.9. *N*-Benzyl-*N*-(4-formylphenyl)-methylsulfonamide (12). *N*-(4-Formylphenyl)methylsulfonamide **11** (2 g, 10 mmol) was dissolved in DMF (20 mL), triethylamine (1.2 mL, 12.1 mmol) was added and the mixture was stirred at room temperature for 30 min. Benzyl chloride (1.21 mL, 10.5 mmol) was then added dropwise and the reaction solution heated for 15 h. The mixture was diluted with 40 mL of 1 N sodium hydroxide solution, and extracted with ethyl acetate. The organic layers were combined, dried over Na_2SO_4 and concentrated in vacuo. The residue was chromatographed on silica gel. Elution with AcOEt/cyclohexane (5/5) provided compound **12** in 41% yield; mp 81–82 °C; IR (KBr) ν 1703, 1338, 1149 cm^{-1}; ^1H NMR (DMSO-d_6) δ 9.96 (s, 1H, CHO), 7.90 (d, 2H, H_{Ar}), 7.65 (d, 2H, H_{Ar}), 7.35–7.20 (m, 5H, H_{Ar}), 5.03 (s, 2H, CH_2), 3.22 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ **192.1, 144.6, 136.3, 134.2, 130.1, 128.4, 127.8, 127.7, 127.4, 52.7, 37.9.**

5.4.10. 5-[4-(*N*-Benzyl-methylsulfonamido)phenyl]-4-(4-chlorophenyl)-3-hydroxy-2(5*H*)-furanone (13). This was prepared from methyl 3-(4-chlorophenyl)-2-oxo-propionate **7a** and *N*-benzyl-*N*-(4-formylphenyl)-methylsulfonamide **12**. Crystallization from ethyl ether gave compound **13** in 69% yield; mp 187 °C; IR (KBr) ν 1757, 1633, 1351, 1156 cm^{-1}; ^1H NMR (DMSO-d_6) δ

11.40 (br s, 1H, OH), 7.60–7.23 (m, 13H, H_{Ar}), 6.57 (s, 1H, H-5), 4.88 (s, 2H, CH_2), 3.11 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 168.7, 140.2, 139.4, 136.5, 135.2, 132.8, 129.2, 128.9, 128.7, 128.6, 128.3, 127.9, 127.3, 126.3, 78.8, 53.4, 37.4.

5.4.11. 4-(4-Chlorophenyl)-3-hydroxy-5-[4-(methylsulfonamido)phenyl]-2(5*H*)-furanone (8h). Pd/C (10%, 0.20 g) was added to a solution of 5-[4-(*N*-benzyl-methylsulfonamido)phenyl]-4-(4-chlorophenyl)-3-hydroxy-2(5*H*)-furanone **13** (0.90 g, 1.91 mmol) in THF/EtOH (5/10 mL). The reaction mixture was then submitted to a hydrogen atmosphere for 12 h. After filtration of the catalyst and evaporation of the solvent, the concentrate was then purified by column chromatography on silica gel with EtOAc/petroleum benzine (4/6) as developing solvent. Crystallization from diisopropyl ether gave compound **8h** in 55% yield; mp 200 °C; IR (KBr) ν 3303, 1742, 1320, 1151 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.08 (br s, 1H, OH), 9.94 (br s, 1H, NH), 7.64 (d, 2H, H_{Ar}), 7.40–7.27 (m, 4H, H_{Ar}), 7.21 (d, 2H, H_{Ar}), 6.56 (s, 1H, H-5), 3.03 (s, 3H, CH_3); ^{13}C NMR (DMSO-d_6) δ 169.1, 139.3, 138.8, 131.5, 130.5, 129.0, 128.5, 127.7, 127.4, 119.4, 79.3, 39.6; EI-MS *m*/*z* 379 $[M]^+$.

5.5. General procedure for synthesis of compounds (15b,c)

DBU (3.5 mL; 23.4 mmol) and ethyl iodide (3.8 mL; 47.5 mmol) were added to a solution of the appropriate phenylacetic acid (23.3 mmol) in dry DMF (80 mL) under stirring at 0 °C, and the mixture was stirred for 3 h at the same temperature, then poured into dilute HCl 3 N. The product was extracted with ethylacetate and the organic extract was washed successively with water, a saturated aqueous Na_2CO_3 solution, brine and dried over Na_2SO_4. Concentration under reduced pressure gave ester **15** of suitable purity for use in subsequent synthetic operations.

5.5.1. Ethyl 4-methylsulfonylphenylacetate (15b). Yield 81%; mp 80 °C (lit.,[61] 79–80 °C); IR (KBr) ν 1727, 1297, 1149 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.87 (d, 2H, H_{Ar}), 7.48 (d, 2H, H_{Ar}), 4.15 (q, 2H, CH_2CH$_3$), 3.70 (s, 2H, CH_2), 3.03 (s, 3H, SO_2CH_3), 1.24 (t, 3H, CH$_2$CH_3); ^{13}C NMR ($CDCl_3$) δ 170.4, 140.4, 139.3, 130.4, 127.6, 61.3, 44.5, 41.1, 14.1.

5.5.2. Ethyl 4-chlorophenylacetate (15c)[62]. Yield 99%.

5.6. General procedure for synthesis of α-bromo 4-substituted-phenylacetate (16)

The synthesis of ethyl α-bromophenylacetate **16a** and its purification were performed as described in the literature[63] and the procedure used to obtain compounds **16b** and **16c**[64] was derived from it, therefore we only include a brief description of the synthesis. The general procedure is as follows: a solution of the appropriate ethyl phenylacetate **15**, *N*-bromosuccinimide (1 equiv) and benzoyl peroxide (catalytic amount) in CCl_4 was heated under reflux for several hours. The reaction solution was cooled to room temperature and filtered free of succinimide. The succinimide was washed with CCl_4 and these extracts were combined with the filtrate before being evaporated in vacuo. The concentrate was then purified by means of a silica gel chromatography to give the desired compound.

5.6.1. Ethyl α-bromo-(4-methylsulfonylphenyl)acetate (16b). Elution with EtOAc/cyclohexane (5/5) gave compound **16b** as an oil in 63% yield; IR (NaCl) ν 1736, 1309, 1150 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.93 (d, 2H, H_{Ar}), 7.76 (d, 2H, H_{Ar}), 5.37 (s, 1H, CH), 4.24 (q, 2H, CH_2CH$_3$), 3.05 (s, 3H, SO_2CH_3), 1.28 (t, 3H, CH$_2$CH_3); ^{13}C NMR ($CDCl_3$) δ **167.6, 141.8, 141.1, 129.8, 127.9, 63.0, 44.9, 44.5, 13.9.**

5.7. General procedure of synthesis of compounds (17)

Triethylamine (1 equiv) was added to a solution of the appropriate phenylacetic acid **14** in dry THF and stirring was maintained for 15 min. The appropriate ethyl bromoacetate **16** was added and the solution refluxed for several hours. After cooling, water was added to dissolve the precipitate and the mixture was extracted with ethyl acetate. The organic layers were combined, successively washed with HCl 3 M twice, water, a saturated solution of $NaHCO_3$ (two times) and brine. Then the solution was dried over Na_2SO_4 and concentrated under reduced pressure. The residue was then purified to give the desired compound **17**.

5.7.1. Ethoxycarbonylmethyl phenylacetate (17a). This was prepared from phenylacetic acid **14a** (3 g, 22.0 mmol) and ethylbromoacetate (2.44 mL, 21.9 mmol) in dry THF (30 mL). The oily residue was then purified by column chromatography on silica gel, eluting with EtOAc/cyclohexane (2/8) and compound **17a** was obtained as an oil in 83% yield; IR (KBr) ν 1747 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.33–7.29 (m, 5H, H_{Ar}), 4.60 (s, 2H, OCH_2CO), 4.18 (q, 2H, CH$_3$CH_2), 3.73 (s, 2H, PhCH_2CO), 1.23 (t, 3H, CH_3CH$_2$); ^{13}C NMR ($CDCl_3$) δ 171.4, 168.1, 133.9, 129.8, 129.1, 127.7, 61.9, 61.5, 41.2, 14.5.

5.7.2. Ethoxycarbonylmethyl 4-methylsulfonylphenylacetate (17b). This was prepared from 4-methylsulfonylphenylacetic acid **14b** (2.5 g, 11.7 mmol) and ethylbromoacetate (1.29 mL, 11.6 mmol) in dry THF (25 mL). After evaporation, compound **17b** was obtained as a white powder in 75% yield; mp 71 °C; IR (KBr) ν 1757, 1745, 1288, 1149 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.93 (d, 2H, H_{Ar}), 7.61 (d, 2H, H_{Ar}), 4.75 (s, 2H, OCH$_2$CO), 4.16 (q, 2H, CH_2CH$_3$), 4.00 (s, 2H, PhCH$_2$CO), 3.25 (s, 3H, SO_2CH_3), 1.21 (t, 3H, CH$_2$CH_3); ^{13}C NMR ($CDCl_3$) δ 170.2, 167.6, 140.0, 139.6, 130.5, 127.1, 61.2, 61.0, 43.6, 39.5, 14.0.

5.7.3. Ethoxycarbonyl-α-(4-methylsulfonylphenyl)methylphenylacetate (17c). This was prepared from phenylacetic acid **14a** (1.5 g, 11.0 mmol) and ethyl α-bromo-(4-methylsulfonylphenyl)acetate **16b** (3 g, 9.34 mmol) in dry THF (15 mL). Purification by column chromatography, eluting with EtOAc/petroleum benzine (3/7), produced compound **17c** as an oil in 63% yield; IR (KBr) ν

1744, 1313, 1151 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.94 (d, 2H, H_{Ar}), 7.64 (d, 2H, H_{Ar}), 7.35–7.26 (m, 5H, H_{Ar}), 6.02 (s, 1H, CH), 4.14 (m, 2H, CH_2CH_3), 3.80 (s, 2H, $PhCH_2CO$), 3.04 (s, 3H, SO_2CH_3), 1.18 (t, 3H, CH_2CH_3); ^{13}C NMR ($CDCl_3$) δ 170.5, 167.6, 141.1, 139.6, 133.0, 129.3, 128.6, 128.2, 127.8, 127.3, 73.8, 62.2, 44.3, 40.8, 13.9.

5.7.4. Ethoxycarbonyl-α-phenylmethyl 4-methylsulfonylphenylacetate (17d). This was prepared from 4-methylsulfonylphenylacetic acid **14b** (1.5 g, 7.00 mmol) and ethyl α-bromophenylacetate **16a** (1.7 g, 6.99 mmol) in dry THF (15 mL). Purification by column chromatography, eluting with EtOAc/cyclohexane (3/7), gave compound **17d** in 28% yield; mp 101–102 °C; IR (KBr) ν 1750, 1740, 1289, 1149 cm^{-1}; ^1H NMR (DMSO-d_6) δ 7.96 (d, 2H, H_{Ar}), 7.66 (d, 2H, H_{Ar}), 7.55–7.45 (m, 5H, H_{Ar}), 6.03 (s, 1H, CH), 4.15 (m, 2H, CH_2CH_3), 4.05 (s, 2H, $PhCH_2CO$), 3.26 (s, 3H, SO_2CH_3), 1.23 (t, 3H, CH_2CH_3); ^{13}C NMR (DMSO-d_6) δ 170.2, 168.4, 139.9, 139.6, 133.6, 130.6, 129.4, 128.9, 127.7, 127.1, 74.6, 61.4, 43.6, 39.5, 13.9.

5.7.5. Ethoxycarbonyl-α-(4-methylsulfonylphenyl)methyl 4-chlorophenylacetate (17e). This was prepared from 4-chlorophenylacetic acid **14c** (0.80 g, 4.69 mmol) and ethyl α-bromo-(4-methylsulfonylphenyl)acetate **16b** (1.5 g, 4.67 mmol) in dry THF (10 mL). Purification by column chromatography, eluting with EtOAc/petroleum benzine (3/7), led to compound **17e** in 82% yield; mp 90–93 °C; IR (KBr) ν 1758, 1746, 1314, 1154 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.96 (d, 2H, H_{Ar}), 7.66 (d, 2H, H_{Ar}), 7.23–7.33 (m, 4H, H_{Ar}), 6.01 (s, 1H, CH), 4.15 (q, 2H, CH_2CH_3), 3.78 (s, 2H, $PhCH_2O$), 3.06 (s, 3H, SO_2CH_3), 1.20 (t, 3H, CH_2CH_3); ^{13}C NMR ($CDCl_3$) δ 170.4, 167.8, 141.5, 139.8, 133.6, 131.8, 131.0, 129.1, 128.5, 128.1, 74.2, 62.6, 44.6, 40.3, 14.2.

5.7.6. Ethoxycarbonyl-α-(4-chlorophenyl)methyl 4-methylsulfonylphenylacetate (17f). This was prepared from 4-methylsulfonylphenylacetic acid **14b** (2.35 g, 11.0 mmol) and ethyl α-bromo-(4-chlorophenyl)acetate **16c** (3.22 g, 11.6 mmol) in dry THF (25 mL). Purification by column chromatography, eluting with EtOAc/cyclohexane (4/6), gave compound **17f** in 77% yield; mp 94 °C; IR (KBr) ν 1736, 1303, 1147 cm^{-1}; ^1H NMR (DMSO-d_6) δ 7.92 (d, 2H, H_{Ar}), 7.60 (d, 2H, H_{Ar}), 7.52 (m, 4H, H_{Ar}), 6.05 (s, 1H, CH), 4.11 (q, 2H, CH_2CH_3), 4.03 (s, 2H, $PhCH_2CO$), 3.23 (s, 3H, SO_2CH_3), 1.09 (t, 3H, CH_2CH_3); ^{13}C NMR (DMSO-d_6) δ 170.0, 168.1, 139.8, 139.6, 134.1, 132.5, 130.5, 129.5, 128.9, 127.0, 73.7, 61.5, 43.5, 39.4, 13.8.

5.7.7. Ethoxycarbonyl-α-(4-methylsulfonylphenyl)methyl 4-benzyloxyphenylacetate (17g). This was prepared from 4-benzyloxyphenylacetic acid **14e** (1.36 g, 5.60 mmol) and ethyl α-bromo-(4-methylsulfonylphenyl)acetate **16b** (1.80 g, 5.60 mmol) in dry THF (15 mL). Purification by column chromatography, eluting with EtOAc/petroleum benzine (3/7), provided compound **17g** as an oil in 88% yield; IR (KBr) ν 1742, 1304, 1145 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.94 (d, 2H, H_{Ar}), 7.65 (d, 2H, H_{Ar}), 7.40–7.32 (m, 5H, H_{Ar}), 7.22 (d, 2H, H_{Ar}), 6.94 (d, 2H, H_{Ar}), 6.00 (s, 1H, CH), 5.05 (s, 2H, $PhCH_2O$), 4.11 (q, 2H, CH_2CH_3), 3.74 (s, 2H, $PhCH_2CO$), 3.03 (s, 3H, SO_2CH_3), 1.18 (t, 3H, CH_2CH_3); ^{13}C NMR ($CDCl_3$) δ 171.0, 167.9, 158.4, 141.5, 140.0, 137.2, 130.7, 128.8, 128.5, 128.2, 128.1, 127.7, 125.6, 115.3, 74.1, 70.2, 62.4, 44.6, 40.2, 14.2.

5.8. General procedure for synthesis of 5-substituted 3-aryl-4-hydroxy-2(5*H*)-furanones (18)

A dropwise solution of compound **17** in dry THF was added to a suspension of NaH (60% dispersion in mineral oil, 1.1 equiv) in dry THF in an ice cold bath and the stirring was maintained at room temperature for several hours. Water was added and the solution was extracted twice with ethyl ether. The aqueous phase was cooled to 0 °C and then acidified with hydrochloric acid (3 M) to give a solid precipitate. Filtration and washing with water or extraction furnished an analytical sample of the furanones.

5.8.1. 4-Hydroxy-3-phenyl-2(5*H*)-furanone (18a). [65] This was prepared from ethoxycarbonylmethyl phenylacetate **17a**. Crystallization from diisopropyl ether led to compound **18a** in 25% yield.

5.8.2. 4-Hydroxy-3-(4-methylsulfonylphenyl)-2(5*H*)-furanone (18b). This was prepared from ethoxycarbonylmethyl 4-methylsulfonylphenylacetate **17b**. Crystallization from ethyl ether led to compound **18b** in 35% yield; mp 240 °C; IR (KBr) ν 1707, 1655, 1308, 1153 cm^{-1}; ^1H NMR (DMSO-d_6) δ 8.24 (d, 2H, H_{Ar}), 7.95 (d, 2H, H_{Ar}), 4.86 (s, 2H, H-5), 3.23 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 178.1, 172.5, 137.7, 136.1, 126.9, 126.2, 95.8, 66.3, 43.7; EI-MS m/z 254 $[M]^+$.

5.8.3. 4-Hydroxy-5-(4-methylsulfonylphenyl)-3-phenyl-2(5*H*)-furanone (18c). This was prepared from ethoxycarbonyl-α-(4-methylsulfonylphenyl)methyl phenylacetate **17c**. The white precipitate thus formed was filtered and the aqueous phase was extracted with ethyl ether (three times). The organic layers were combined, dried over Na_2SO_4 and evaporated in vacuo. Crystallization from ethyl ether led to compound **18c** in 23% yield; mp 188 °C; IR (KBr) ν 1718, 1647, 1310, 1154 cm^{-1}; ^1H NMR (DMSO-d_6) δ 8.05 (d, 2H, H_{Ar}), 7.96 (m, 2H, H_{Ar}), 7.73 (d, 2H, H_{Ar}), 7.45 (t, 2H, H_{Ar}), 7.31 (t, 1H, H_{Ar}), 6.13 (s, 1H, H-5), 3.28 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 175.0, 171.9, 141.6, 140.8, 130.1, 129.0, 128.1, 127.6, 126.9, 126.8, 98.6, 78.3, 43.5; EI-MS m/z 330 $[M]^+$

5.8.4. 4-Hydroxy-3-(4-methylsulfonylphenyl)-5-phenyl-2(5*H*)-furanone (18d). This was prepared from ethoxycarbonyl-α-phenylmethyl 4-methylsulfonylphenylacetate **17d**. Crystallization from ethyl ether led to compound **18d** in 55% yield; mp 226 °C; IR (KBr) ν 1737, 1660, 1305, 1149 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.90 (br s, 1H, OH), 8.31 (d, 2H, H_{Ar}), 7.99 (d, 2H, H_{Ar}), 7.48–7.44 (m, 5H, H_{Ar}), 5.99 (s, 1H, H-5), 3.24 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) δ 178.6, 171.8, 137.8, 136.2, 135.0, 129.4, 128.9, 128.1, 126.9, 126.6, 96.5, 78.6, 43.7; EI-MS m/z 330 $[M]^+$.

5.8.5. 3-(4-Chlorophenyl)-4-hydroxy-5-(4-methylsulfonylphenyl)-2(5*H*)-furanone (18e). This was prepared from ethoxycarbonyl-α-(4-methylsulfonylphenyl)methyl 4-chlorophenylacetate **17e**. Crystallization from diisopropyl ether led to compound **18e** in 51% yield; mp 122–125 °C; IR (KBr) *ν* 1728, 1667, 1298, 1152 cm^{-1}; ^1H NMR (DMSO-d_6) *δ* 8.20–7.98 (m, 4H, H_{Ar}), 7.67 (d, 2H, H_{Ar}), 7.46 (d, 2H, H_{Ar}), 6.25 (s, 1H, H-5), 3.25 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) *δ* 176.0, 171.9, 141.6, 140.7, 131.0, 130.3, 129.3, 129.1, 128.3, 127.6, 97.2, 77.5, 43.5; EI-MS *m/z* 364 $[M]^+$.

5.8.6. 5-(4-Chlorophenyl)-4-hydroxy-3-(4-methylsulfonylphenyl)-2(5*H*)-furanone (18f). This was prepared from ethoxycarbonyl-α-(4-chlorophenyl)methyl 4-methylsulfonylphenylacetate **17f**. Crystallization from diisopropyl ether led to compound **18f** in 51% yield; mp 262 °C; IR (KBr) *ν* 3019, 1707, 1651, 1309, 1151 cm^{-1}; ^1H NMR (DMSO-d_6) *δ* 11.86 (br s, 1H, OH), 8.28 (d, 2H, H_{Ar}), 7.96 (d, 2H, H_{Ar}), 7.52 (d, 2H, H_{Ar}), 7.44 (d, 2H, H_{Ar}), 5.99 (s, 1H, H-5), 3.22 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) *δ* 178.6, 171.8, 137.8, 136.1, 134.1, 130.0, 128.9, 126.9, 126.6, 96.4, 77.8, 43.7; EI-MS *m/z* 364 $[M]^+$.

5.8.7. 3-(4-Benzyloxyphenyl)-4-hydroxy-5-(4-methylsulfonylphenyl)-2(5*H*)-furanone (18g). This was prepared from ethoxycarbonyl-α-(4-methylsulfonylphenyl)methyl 4-benzyloxyphenylacetate **17g**. Crystallization from diisopropyl ether led to compound **18g** in 66% yield; mp 169 °C; IR (KBr) *ν* 3032, 1714, 1637, 1302, 1148 cm^{-1}; ^1H NMR (DMSO-d_6) *δ* 8.02 (d, 2H, H_{Ar}), 7.89 (d, 2H, H_{Ar}), 7.68 (d, 2H, H_{Ar}), 7.46–7.34 (m, 5H, H_{Ar}), 7.09 (d, 2H, H_{Ar}), 6.08 (s, 1H, H-5), 5.15 (s, 2H, $PhCH_2O$), 3.26 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) *δ* 173.5, 172.1, 157.1, 141.6, 140.9, 137.1, 129.0, 128.5, 128.2, 127.8, 127.7, 127.6, 122.6, 114.6, 98.5, 77.3, 69.2, 43.5.

5.8.8. 3-(4-Hydroxyphenyl)-4-hydroxy-5-(4-methylsulfonylphenyl)-2(5*H*)-furanone (18h). Pd/C (10%, 80 mg) was added to 3-(4-benzyloxyphenyl)-4-hydroxy-5-(4-methylsulfonylphenyl)-2(5*H*)-furanone **18g** (313 mg, 0.72 mmol) dissolved in THF/EtOH (5/10 mL) and submitted to a hydrogen atmosphere for 4 h. After filtration of the catalyst and evaporation of the solvent, the residue was purified by column chromatography on silica gel, eluting with EtOAc/cyclohexane (8/2) and compound **18h** was obtained in 42% yield; IR (KBr) *ν* 3326, 1719, 1669, 1302, 1144 cm^{-1}; ^1H NMR (DMSO-d_6) *δ* 9.51 (br s, 1H, OH), 8.00 (d, 2H, H_{Ar}), 7.75 (d, 2H, H_{Ar}), 7.65 (d, 2H, H_{Ar}), 6.80 (d, 2H, H_{Ar}), 6.04 (s, 1H, H-5), 3.25 (s, 3H, SO_2CH_3); ^{13}C NMR (DMSO-d_6) *δ* 173.2, 172.6, 156.7, 141.9, 141.4, 129.3, 128.7, 128.0, 121.1, 115.4, 99.4, 77.6, 43.9; EI-MS *m/z* 346 $[M]^+$.

5.9. Anti-oxidant assays

5.9.1. Measurement of the reducing activity against the stable radical DPPH. A solution of 4 mL of DPPH (10^{-4} M, 2 mg) in EtOH (50 mL) was added to 1 mL of a solution of the test compound in DMF, to prepare a 5×10^{-3} M sample solution. The absorbance at 517 nm was measured every 5 min for 3 h. The absorbance of the control sample without test compound was measured simultaneously. The difference in absorbance between the control and the test compound was taken as the reducing activity.

5.9.2. Superoxide anion scavenging assay. The technique of Slater and Eakins[40] based on the interactions of NADH, phenazine methosulfate (PMS), molecular oxygen and nitro blue tetrazolium (NBT) was used to evaluate the superoxide anion scavenging. The NADH/PMS/O_2/NBT system involves the intermediate formation of the superoxide anion radical ($O_2^{\cdot -}$) from the interaction of reduced PMS with O_2; the superoxide anion radical then reduces NBT to the highly coloured formazan. The reaction can be followed by measuring the absorbance of the formazan at 578 nm. The incubation mixture contained disodium hydrogen phosphate buffer (200 μL, 19 mM, pH = 7.4), PMS (200 μL, 10.8 μM), NBT (200 μL, 172 μM) and NADH + H^+ (200 μL, 360 μM). In order to assess their activity, the compounds were dissolved in the buffer at different concentrations and 200 μL of buffer in the reaction mixture were replaced by 200 μL of these solutions of compounds. Meanwhile, blanks were realized by replacing 200 μL of NADH + H^+ by 200 μL of buffer. Each test was performed after a 5 min incubation.

5.9.3. Lipid peroxidation assay. Iron-dependent peroxidation of male rat liver microsomes was tested as previously described.[66] Liver microsomes were prepared according to the technique of May and McCay.[67] Lipid peroxidation was initiated by the ADP-Fe^{2+}/ascorbate system,[68] which produced hydroxyl radical (·OH). Incubations of 200 μL microsomal fractions (1 mg protein/mL) were carried out at 37 °C with 200 μL potassium phosphate buffer (0.1 M, pH = 7.4), and 200 μL of ADP solution in buffer (2 mM), 200 μL of aqueous $FeSO_4$ solution (16 μM). Lipid peroxidation was initiated by adding a 200 μL aqueous ascorbate solution (0.5 mM) to the above incubation mixture. The extent of polyunsaturated fatty acid peroxidation was measured spectrophotometrically by the rate of malondialdehyde formed after a 90 min microsomial incubation. The reaction was quenched by the addition of 0.5 mL of 20% trichloroacetic acid; after centrifugation, the supernatant fraction was collected and then boiled with 0.5 mL of 0.67% thiobarbituric acid solution for 20 min. After cooling, samples were diluted with 1 mL of distilled water and extinction was read at 535 nm. In order to assess their activity, the compounds were dissolved into disodium hydrogen phosphate buffer (19 mM, pH = 7.4) at different concentrations and 200 μL of buffer in the reaction mixture was replaced by 200 μL of the solution of compounds. In parallel, blanks were realized by replacing 200 μL of ascorbate by 200 μL of buffer.

5.10. Pharmacological assays

All tests were performed in accordance with the recommendations and policies of the International Association for the Study of Pain (IASP).[69]

5.10.1. Acute toxicity in mice. The compounds were administered intraperitoneally as suspensions in saline (0.9% NaCl) at doses of 200, 400, 600 and 800 mg/kg. Swiss male mice purchased from Elevage Depre (Saint-Doulchard, France) weighing 18–22 g were used. Mice were kept in groups of 10 in a temperature controlled room with a 12 h light/dark cycle. Food and water were available ad libitum during the time of the experiment. The animals were observed for 8 days in order to detect any sign of toxicity.

5.10.2. TPA-induced ear edema. Male Swiss mice (20–22 g) were used (Elevage Depre, Saint-Doulchard, France). The anti-inflammatory activity of the studied drugs was evaluated according to the procedure of Carlson et al.[54] Mice were divided into groups of 10. Phorbol 12-myristate 13-acetate (TPA) (Sigma–Aldrich, Saint-Quentin Fallavier, France) was dissolved in 80% aqueous ethanol at concentration of 250 μg/mL. Drugs or saline were given intraperitoneally (0.25 mL/20 g). Thirty minutes after an intraperitoneal drug administration or 1 h after an oral drug administration, TPA was applied topically by an automatic pipette to the right ear (10 μL) and left ear only received the vehicle (10 μL of ethanol/water 80/20). Inflammation was allowed to develop for 3 h 30 min after TPA. Edema measurements were taken with an Oditest Calipers (Oditest S 5010-Kreoplin, Gmbh, Germany). The thickness of the right and left ears was measured in units of 0.01 mm. Ear edema was calculated by substracting the thickness of the left ear (vehicle control) from the right one (treated ear).

5.10.3. Carrageenan-induced paw edema in rat. Male Sprague–Dawley rats (120–140 g) were used (Elevage Depre, Saint-Doulchard, France). The anti-inflammatory activity of the studied drugs was evaluated according to the procedure of Winter et al.[57] Rats were divided into groups of six and treated intraperitoneally (0.25 mL/100 g) with compounds or saline. Thirty minutes after intraperitoneal drug administration or 1 h after oral drug administration, 0.05 mL of a w/v 1% suspension of carrageenan (Sigma–Aldrich, Saint-Quentin Fallavier, France) in deionized water was given by intraplantar injection into the right paw. The animals were returned to the cage and 3 h later the edema was measured with a plethysmometer (V.G.O. Basile-Apelex 05.7150). The edema means of the compound-treated groups were compared to the vehicle to give the percentage of inhibition of paw swelling.

5.10.4. Data analysis. Statistical analysis of the results was performed using the method of Schwartz.[70] Data were analyzed by using the Student's *t*-test. All values were expressed as mean ± SD.

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