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Full Papers

In Vitro Evaluation of Antifungal Properties of Phenylpropanoids and Related Compounds Acting Against Dermatophytes

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Thirty-four arylpropanoids and related compounds were evaluated in vitro for antifungal properties. Among them, 22 phenyl-, 4 naphthyl-, and 4 phenanthrylpropanoids; naphthalene; phenanthrene; and 2-chloro-1-hexyl-1-propanone were tested against dermatophytes by the agar dilution method. α-Halopropiophenones exhibited a broad spectrum of activities against *Microsporum canis*, *Microsporum gypseum*, *Trichophyton mentagrophytes*, *Trichophyton rubrum*, and *Epidermophyton floccosum*, with MIC values between 0.5 and >50 μg/mL. Keto, alcohol, and α-haloketo propyl derivatives of naphthalene and phenanthrene also showed very good activity against all dermatophytes tested, clearly showing that in these series, a halogen atom is not necessary for activity. Phenanthryl derivatives were more active (MICs, 3−20 μg/mL) than naphthyl ones (MICs, 3−50 μg/mL). A structure−activity relationship study was carried out and aided in establishing the structural requirements of arylpropanoids for antifungal activities. Because dermatophytes are a group of fungi that characteristically infect the keratinized areas of the body, these new series of antifungal compounds open the possibility of discovering new topical antifungal drugs for the treatment of dermatomycoses, which are frequently very difficult to eradicate.

During the past two decades, the incidence of fungal infections, especially involving immunocompromised patients, has increased dramatically.[1] In particular, some forms of dermatomycoses are the cause of a great morbidity in patients receiving antineoplastic chemotherapy, undergoing organ transplants, or suffering from AIDS. These infections are produced by dermatophytes, a group of fungi that characteristically infect the keratinized areas of the body. Although imidazole compounds such as clotrimazole, miconazole, and econazole have proven to be effective for the treatment of dermatomycoses, these infections are frequently very difficult to eradicate,[2] and more effective new topical antifungal agents are still needed.

In the course of our screening program for antifungal activity, we reported that 8.O.4′-neolignans (a small group among the great structural variety of neolignans) possess moderate but significant antifungal activity against dermatophytes.[3]

In addition, other types of neolignans and structurally related lignans showed antifungal activities.[4−6] Considering that lignans and neolignans are plant compounds formed by two C-6−C-3 units[7] and that dimerization of phenylpropanoids through dehydrogenation produces the skeleton of all natural and synthetic lignans known to date, we decided to carry out a systematic study of the antifungal properties of their phenylpropanoid moieties.

Studies have reported contradictory evidence of antimicrobial activities of some phenylpropanoids. Zemek et al.,[8] for example, reported that isoeugenol exhibits an inhibitory effect on the growth of bacteria, yeasts, yeast-like organ-

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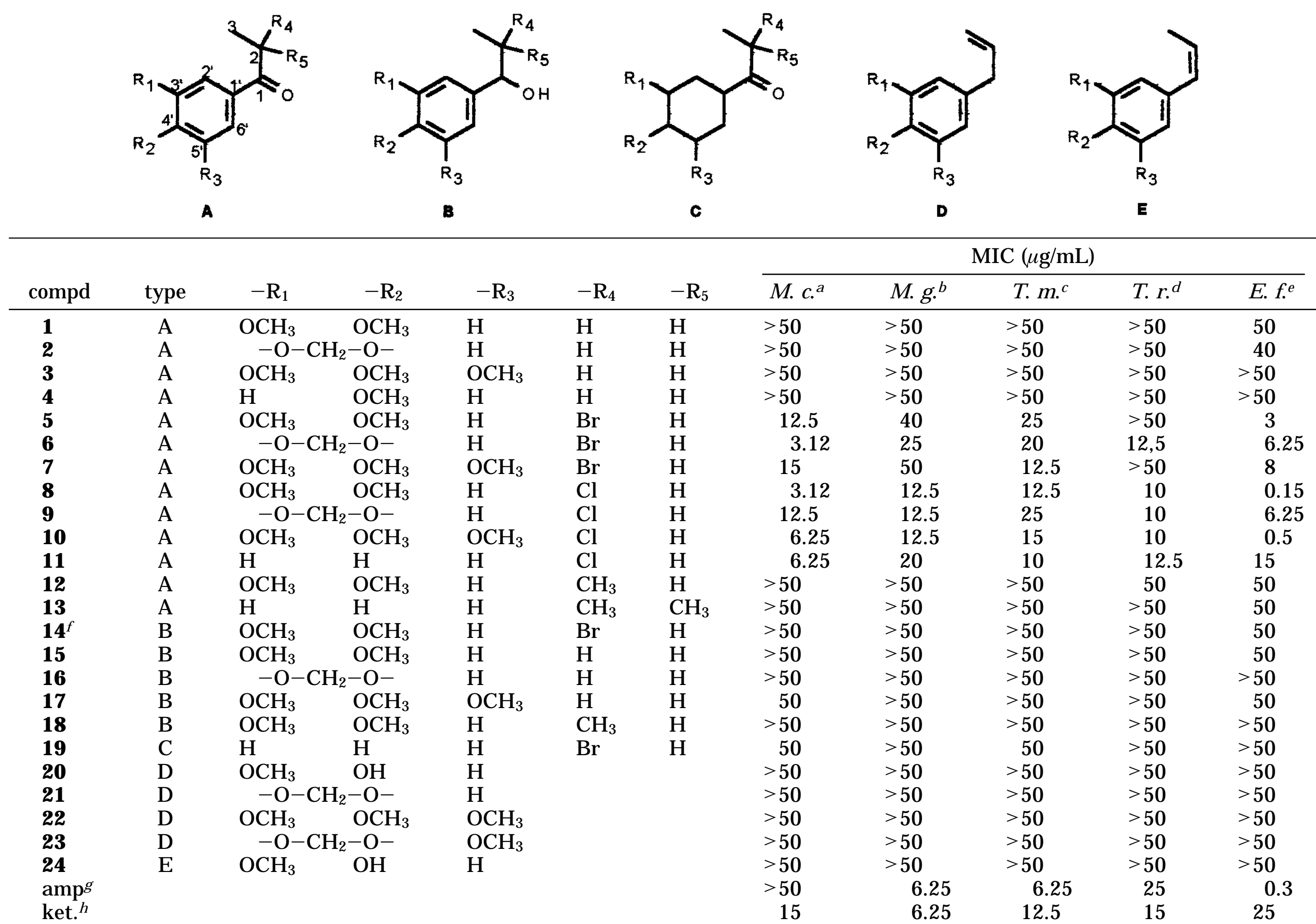
Table 1. *In Vitro* Antifungal Activities of Phenylpropanoids and a Nonaromatic Analogs



compd	type	$-R_1$	$-R_2$	$-R_3$	$-R_4$	$-R_5$	MIC (μg/mL) *M. c.*[a]	*M. g.*[b]	*T. m.*[c]	*T. r.*[d]	*E. f.*[e]
1	A	OCH_3	OCH_3	H	H	H	>50	>50	>50	>50	50
2	A	$-O-CH_2-O-$		H	H	H	>50	>50	>50	>50	40
3	A	OCH_3	OCH_3	OCH_3	H	H	>50	>50	>50	>50	>50
4	A	H	OCH_3	H	H	H	>50	>50	>50	>50	>50
5	A	OCH_3	OCH_3	H	Br	H	12.5	40	25	>50	3
6	A	$-O-CH_2-O-$		H	Br	H	3.12	25	20	12,5	6.25
7	A	OCH_3	OCH_3	OCH_3	Br	H	15	50	12.5	>50	8
8	A	OCH_3	OCH_3	H	Cl	H	3.12	12.5	12.5	10	0.15
9	A	$-O-CH_2-O-$		H	Cl	H	12.5	12.5	25	10	6.25
10	A	OCH_3	OCH_3	OCH_3	Cl	H	6.25	12.5	15	10	0.5
11	A	H	H	H	Cl	H	6.25	20	10	12.5	15
12	A	OCH_3	OCH_3	H	CH_3	H	>50	>50	>50	50	50
13	A	H	H	H	CH_3	CH_3	>50	>50	>50	>50	50
14[f]	B	OCH_3	OCH_3	H	Br	H	>50	>50	>50	>50	50
15	B	OCH_3	OCH_3	H	H	H	>50	>50	>50	>50	50
16	B	$-O-CH_2-O-$		H	H	H	>50	>50	>50	>50	>50
17	B	OCH_3	OCH_3	OCH_3	H	H	50	>50	>50	>50	50
18	B	OCH_3	OCH_3	H	CH_3	H	>50	>50	>50	>50	>50
19	C	H	H	H	Br	H	50	>50	50	>50	>50
20	D	OCH_3	OH	H			>50	>50	>50	>50	>50
21	D	$-O-CH_2-O-$		H			>50	>50	>50	>50	>50
22	D	OCH_3	OCH_3	OCH_3			>50	>50	>50	>50	>50
23	D	$-O-CH_2-O-$		OCH_3			>50	>50	>50	>50	>50
24	E	OCH_3	OH	H			>50	>50	>50	>50	>50
amp[g]							>50	6.25	6.25	25	0.3
ket.[h]							15	6.25	12.5	15	25

[a] *M. canis* C 112. [b] *M. gypseum* C 115. [c] *T. mentagrophytes* ATCC 9972. [d] *T. rubrum* C 113. [e] *E. floccosum* C 114. [f] *threo* isomer. [g] Amp. = amphotericin B. [h] Ket. = ketoconazole.

isms, and molds twice as effective as its dimer, which consists of two isoeugenol units. Contrary to this, Hattori et al.[9] reported that dimeric phenylpropanoids show stronger antimicrobial action than the corresponding monomer unit. On the other hand, Himejima et al.[10] reported that safrole, methyleugenol, eugenol, and anethole were inactive against *Saccharomyces cerevisiae*, *Candida utilis*, *Pityrosporum ovale*, and *Penicillum chrysogenum* at concentrations up to 100 μg/mL.

A more systematic investigation of the antifungal activities of phenylpropanoids utilizing a large number of compounds seems in order and will provide information on the role that phenylpropanoids play in the antifungal activity of their dimers against human pathogenic and opportunistic fungi and will establish whether the antifungal activity reported for 8.O.4′-neolignans can be ascribed to their phenylpropanoid moieties. In addition, this systematic study could aid in the development of new and more potent topical antifungal agents.

We describe here the antifungal properties of phenylpropanoids **1**−**18**, **20**−**24**, and hexylpropanoid **19** against dermatophytes. In addition, results obtained with naphthalene, phenanthrene, naphthyl- and phenanthryl-propanoids **25**−**32**, and a structure−activity relationship (SAR) study considering all the evaluated compounds are also reported.

Among phenylpropanoids tested, aromatic ether **12** was isolated from *Wedelia forsteriana*[11] (Compositae); eugenol **20** was isolated from *Myristica fragrans* (Myristicaceae), *Cinnamomum zeylanicum* (Lauraceae), and *Eugenia caryophillus*[12] (Myrtaceae); methyleugenol **21**, 5′-methoxyeugenol **22**, elemicin **23**, and isoeugenol **24** were isolated from *M. fragrans*;[13] and compounds **22** and **23** were isolated from *Virola surinamensis*[14] (Myristicaceae) and *Diphlolophium buchanani*[15] (Umbelliferae), respectively. Compounds **1**−**3**, **5**−**11**, and **14**−**19** are synthetic analogues. Compounds **4** and **13** are commercial samples (Aldrich Chemical Co). To the best of our knowledge, compounds **9**, **10**, **15**, **18**, and **29**−**32** have not been reported previously in the literature.

In this study, agar dilution assays[16] were used to determine the minimal inhibitory concentrations (MICs) of compounds by using a panel of human pathogenic fungi consisting of yeasts as well as dermatophytes.

Results and Discussion

To carry out the antifungal evaluation, concentrations of arylpropanoids up to 50 μg/mL were incorporated into growth media according to reported procedures.[3] Compounds producing no inhibition of fungal growth at that level were considered inactive.

The agar dilution method showed that none of the compounds tested possessed any activity against the yeasts *Candida albicans*, *S. cerevisiae*, or *Cryptococcus neoformans* or the filamentous fungi *Aspergillus niger*, *Aspergillus fumigatus*, or *Aspergillus flavus* (results not shown). In contrast, different results were obtained for compounds of the series against dermatophytes, with some compounds displaying strong activities. These results are shown in Tables 1 and 2. All of the dermatophytes tested were

Table 2. In Vitro Antifungal Activities of Naphthyl- and Phenanthrylpropanoids; Naphthalene and Phenanthrene

G H

J K

compd	type	R	MIC(μg/mL)				
			M. c.[a]	*M. g.*[b]	*T. m.*[c]	*T. r.*[d]	*E. f.*[e]
25	G	H	50	40	25	40	40
26	G	CH_3	40	25	25	>50	40
27	H	H	50	40	25	40	40
28	H	CH_3	25	25	12.5	12.5	25
29	J	H	3	6.25	3	6.25	20
30	J	CH_3	20	20	6.25	20	20
31	K	CH_3	6.25	6.25	6.25	10	10
32	J	Cl	3	10	3	1.5	15
napht.[f]			>50	>50	>50	>50	>50
phen.[g]			>50	>50	>50	>50	>50
amp.[h]			>50	6.25	6.25	25	0.3
ket.[i]			15	6.25	12.5	15	25

[a] *M. canis* C112. [b] *M.* gypseum C115. [c] *T.* mentagrophytes ATCC 9972. [d] *T.* rubrum C113. [e] *E.* floccosum C114. [f] Napht. = naphthalene. [g] Phen. = phenanthrene. [h] Amp. = amphotericin B. [i] Ket. = ketoconazole.

inhibited at 50 μg/mL and most often at lower concentrations.

Among the phenylpropanoids tested, compounds **1**−**3**, **15**−**17**, and **20**−**22**, constitutive moieties of 8.O.4′-neolignans reported previously,[3] and compounds **4** and **12**−**14**, were all inactive. However, it is interesting to note that phenylpropanoids **5**−**11** showed strong antifungal activities comparable to those of amphotericin B and ketoconazole (Table 1). Compounds **6** and **8**−**11** inhibited all tested dermatophytes with MICs ranging between 0.5 and 25 μg/mL, and compounds **5** and **7** were active against all dermatophytes except *Tricophyton rubrum*, with MICs ranging between 3 and 50 μg/mL. The analysis of active structures reveals that all of them possess a halogen in C-2 and a keto group in C-1, and bromine derivatives **5**−**7** were less active (MICs between 3 and >50 μg/mL) than chlorine derivatives **8**−**10** (MICs between 0.5 and 25 μg/mL). By comparing these results with those obtained with compounds **1**−**4**, **12**, and **13** (MICs = 40 μg/mL against all dermatophytes tested), which have only a keto group joined to a benzene ring in their structures, it would appear that the presence of a halogen atom at C-2 seems to be crucial to produce the biological response. Nevertheless, the lack of activity found for *threo*-bromohydrin **14** indicates that the presence of a halogen group is necessary but not sufficient for antifungal activity of phenylpropanoids.

To evaluate if a halogen in a [$-COCH(CH_3)X$] system is actually responsible for the observed activity, we tested compound **19**. The fact that it showed no activity against all tested dermatophytes also suggests that this system alone is not enough to produce the antifungal response.

To determine if the halogen atom effects the antifungal response through electronic or steric factors, we compared the antifungal activities of compounds **8** and **12** (the last one obtained through a bioisosteric substitution of chlorine by a methyl group in compound **8**). Compound **12** was inactive, suggesting that the biological response is governed by electronic rather than steric factors.

Because α-haloketones **5**−**11**, which have substantial intrinsic chemical and biological reactivity,[17] were the only phenylpropanoids active against dermatophytes, we synthesized and tested structures **25**−**31** (Table 2). These compounds do not possess α-halo groups, and benzene rings were replaced by two different extended π-systems (naphthalene and phenanthrene).

The replacement of the benzene ring with or without substituents (compounds **1**−**4**, **12**, and **15**−**18**) by naphthalene and phenanthrene rings (compounds **25**−**28** and **29**−**31**) showed that an increase in the number of rings respective to benzene results in better activity. The comparison of MICs of keto compounds **12**, **26**, and **30** illustrates this point well. Among naphthyl and phenanthryl derivatives, results showed that compounds **25**−**31** inhibited all dermatophytes tested with MICs between 3 and 50 μg/mL, phenanthrene derivatives **29**−**31** being the most active ones (MICs 3−20 μg/mL).

It is interesting to note, that whereas the presence of both halogen and keto groups were essential for antifungal activities in the phenylpropanoid series, the same seems not to be true for naphthyl and phenanthryl compounds. Keto compounds **25**, **26**, **29**, and **30** and alcohol derivatives **27**, **28**, and **31** all possess antifungal activities. In addition, it is noteworthy that even though α-halo ketone **32** displays strong antifungal activity, ketone **29**, which does not possess a halogen atom in its structure, shows similar antifungal properties. In addition, although the bioisosteric substitution of the chlorine atom by a methyl group (compounds **32** vs **30**) results in a decrease in antifungal activity (compare MICs of both compounds), compound **30** does show some antifungal properties, in some cases similar to or better than the antifungal agents amphotericin or ketoconazole.

To test the actual role of naphthyl or phenanthryl structures in the antifungal activities of their propyl derivatives, naphthalene and phenanthrene were evaluated with the agar dilution method. Results indicated that these compounds were completely devoid of activity, clearly showing that the presence of naphthyl or phenanthryl framework alone is not enough to produce activity.

Regarding fungi tested with phenanthrene derivatives, *Trichophyton mentagrophytes* was the most susceptible species (4 MICs ≤ 6.25 μg/mL) followed by *Microsporum canis* (3 MICs ≤ 20 μg/mL).

According to the results obtained here, we could not attribute to phenylpropanoid moieties the activities reported previously for 8.O.4′-neolignans. In addition we found that among phenylpropanoids tested, only α-haloketoderivatives possess strong antifungal activities against dermatophytes. Because they are known reactive compounds, in some cases vesicant or lachrymatory,[17,18] they are not suitable for developing new antifungal topical compounds. Nevertheless, when we investigated the effect of replacing the phenyl group by naphthyl or phenanthryl structures in the monomers, we found a novel series of arylpropanoids not possessing halogen in their structures (compounds **25**−**31**), with a strong antifungal effect against dermatophytes. Among them, structures **29**−**31** displayed antifungal behavior similar to or better than those of amphotericin B or ketoconazole, and therefore they could

be leads for the development of new topical antifungal agents. In addition, results reported here open the possibility of synthesizing new dimers that may have better antifungal properties than the previously reported antifungal 8.O.4′-neolignans.

Experimental Section

General Experimental Procedures. Melting points were obtained on an electrothermal apparatus and are uncorrected. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker model AC 200 spectrometer in $CDCl_3$ solutions. Carbon chemical shifts are expressed in the δ scale in parts per million, using $CDCl_3$ as a reference signal at 76.9. J values are given in Hertz. Elemental analyses were carried out at Atlantic Microlab, Inc. (Norcross, GA), and all compounds submitted for testing had analytical results within ±0.4% of the theoretical values. Reversed-phase HPLC was performed on a Beckman chromatograph (model 332) equipped with an UV detector of 254 nm, on a C_{18} ODS2 analytical column (0.46 × 25 cm) using MeOH−H_2O 70:30 and MeCN−H_2O 60:40 as the eluents, with a flow rate of 1 mL/min. Preparative TLC was done on Si gel 60 F_{254} 1 mm. IR spectra were measured with a Bruker IFS 25 FT IR spectrophotometer. MS were measured on a MS Shimadzu QP-5000 and a ZAB−SEQ4F70 spectrometer.

Test Compounds. Phenylpropanoids **1**, **5**, and **8**,[19,20] **2**,[20−22] **3**, **7**, and **17**,[14−23] **7**,[22,24] **11**,[18] **12**,[25−28] *threo*-bromohydrin **14**,[29] alcohol **16**[3], hexylpropanoid **19**,[30] and compounds **22**,[14,25] **27**,[31,32] **26**,[33] and **28**[33] were synthesized following general methods described previously.[18−30] Their ^{1}H NMR, ^{13}C NMR, IR, mp, and MS spectra were all in agreement with previously reported data.[18−33] Compounds **4**, **13**, **20**, **21**, **23**, and **24** were commercial samples.

2-Methyl-1-(3′,4′-dimethoxyphenyl)-1-propanol (18). Compound **18** was prepared through a Grignard reaction[14] with 3,4-dimethoxybenzaldehyde (Aldrich) (539.5 mg, 3.25 mmol) in anhydrous Et_2O (5 mL) and a 2.0 M solution of isopropylmagesium chloride (Aldrich) (6.7 mL, 13.34 mmol). The mixture was stirred for 8 h, then cooled in an ice bath and poured into a cold saturated ammonium chloride solution. The ethereal layer was washed with aqueous 1% NaOH and brine and thoroughly washed with 0.1 N HCl and H_2O, dried (Na_2SO_4), and concentrated under reduced pressure. The residue was purified by column chromatography on Si gel 60H using hexane−EtOAc (9:1) as the eluent; 491.7 mg (2.34 mmol, 72% yield) of 2-methyl-1-(3′,4′-dimethoxyphenyl)-1-propanol (**18**) was obtained as white crystals: mp 67−68 °C; IR (KBr) ν_{max} 2970, 2920, 2890, 1605 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 0.75 (3H, d, J = 6.8 Hz, H-3), 0.98 (3H, d, J = 6.8 Hz, H-3), 1.90 (1H, m, H-2), 2.04 (1H, brs, C-1, −OH), 3.83−3.84 (2 × OCH_3), 4.23 (1H, d, J = 7.1 Hz, H-1), 6.78−6.84 (3H, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 18.4 and 18.9 (q, C-3 and 2-methyl), 35.1 (d, C-2), 55.7 (q, −OCH_3 on C-3′ and C-4′), 79.8 (d, C-1), 109.2 (d, C-2′), 110.4 (d, C-5′), 118.7 (d, C-6′), 136.2 (s, C-1′), 148.0 (s, C-4′), 148.6 (s, C-3′); EIMS *m/z* [M$^+$] 210 (63), 168 (100), 139 (92), 108 (72), 77 (22) 71 (45), 65 (18); HREIMS *m/z* 210.1260 (calcd for $C_{12}H_{18}O_3$, 210.1256); *anal.* C 68.53%, H 8.68%, calcd for $C_{12}H_{18}O_3$, C 68.53%, H 8.63%.

α-Chloropropiophenones. A chlorination procedure suitable for preparing compounds **9**, **10,** and **32** was made following the methodology described by Kosower et al.[18] It is illustrated for the case of α-chloro-1-(3′,4′-methylenedioxyphenyl)-1-propanone (**9**).

α-Chloro-1-(3′,4′-methylenedioxyphenyl)-1-propanone (9). Piperonyl propiophenone **2** (320.5 mg, 1.8 mmol) was added to a mixture of copper (II) chloride dihydrate (523 mg, 3.10 mmol), lithium chloride (90 mg, 2.1 mmol), and DMF (7 mL) and heated for 6 h to 80 °C. α-Chloro-1-(3′,4′-methylenedioxyphenyl)-1-propanone (**9**) (252.3 mg, 1.19 mmol, 66% yield) was obtained after column chromatography purification (Si gel 60 H, eluent $CHCl_3$) as a white solid: mp 32.5−33 °C; IR (KBr) ν_{max} 3010, 2940, 1695 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.71 (3H, d, J = 6 Hz, H-3), 5.16 (1H, q, J = 6 Hz, H-2), 6.05 (2H, s, −OCH_2O−), 6.87−7.63 (ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 19.9 (q, C-3), 52.5 (d, C-2), 101.9 (t, −OCH_2O−), 107.9 (d, C-2′), 108.6 (d, C-5′), 125.6 (d, C-6′), 128.5 (s, C-1′), 148.2 (s, C-3′), 152.2 (s, C-4′), 191.8 (s, C-1); EIMS *m/z* [M$^+$] 212 (17), 149 (100), 121 (50), 91 (20), 65 (35); HREIMS *m/z* 212.0243 (calcd for $C_{10}H_9ClO_3$, 212.0240); HPLC (MeOH−H_2O) t_R = 8.68 min (99.8%); HPLC (MeCN−H_2O) t_R = 7.82 min (99.7%).

α-Chloro-1-(3′,4′,5′-trimethoxyphenyl)-1-propanone (10). Compound **10** was obtained following the same method used for compound **9** with ketone **3** (255.4 mg, 1.14 mmol), copper(II) chloride dihydrate (340 mg, 2 mmol), lithium chloride (50.4 mg, 1.2 mmol, and DMF (6 mL). α-Chloro-1-(3′,4′,5′-trimethoxyphenyl)-1-propanone (**10**) (196 mg, 0.76 mmol, 67% yield) was obtained as white crystals (hexane): mp 71.5−72.5 °C; IR (KBr) ν_{max} 2941, 1685, 1128 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.70 (3H, d, J = 6.6 Hz, H-9), 3.88−3.90 (3 × OCH_3), 5.19 (1H, q, J = 6.6 Hz, H-8), 7.25 (2H, s, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 19.9 (q, C-3), 52.4 (d, C-2), 56.2 (q, −OCH_3 on C-3′ and C-5′), 60.8 (q, −OCH_3 on C-4′), 106.6 (d, C-2′, C-6′), 129.0 (s, C-1′), 143.2 (s, C-4′), 153.0 (s, C-3′, C-5′), 192.3 (s, C-1); EIMS *m/z* [M$^+$] 258 (9), 195 (100), 167 (18), 152 (21), 109 (19), 77 (26), 66 (26); HREIMS *m/z* 258.0654 (calcd for $C_{12}H_{15}O_4Cl$, 258.0659); *anal.* C 55.78%, H 5.86%, Cl 13.52%, calcd for $C_{12}H_{15}O_4Cl$, C 55.80%, H 5.86%, Cl 13.55%.

1-(3′,4′-Dimethoxyphenyl)-1-propanol (15). An ethereal solution of ketone **1** (428 mg, 2.2 mmol) was gradually added to a stirred suspension of $LiAlH_4$ (456 mg, 12 mmol) in dry Et_2O (36 mL). After addition was complete, the mixture was refluxed for 8 hs. Excess $LiAlH_4$ was carefully destroyed by addition of EtOAc. The mixture was extracted with Et_2O (2 × 50 mL). The combined Et_2O extracts were washed with 10% HCl, saturated aqueous NaCl and H_2O, dried (Na_2SO_4), decanted, and evaporated under vacuum, yielding 356 mg (1.82 mmol, 83%) of 1-(3′,4′-dimethoxyphenyl)-1-propanol (**15**) as a colorless oil; IR (film) ν_{max} 3482, 2968, 2874, 1594 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 0.89 (3H, t, J = 7.3 Hz, H-3), 1.78 (2H, m, H-2), 1.96 (1H, brs, C-1, −OH), 3.86−3.87 (2 × OCH_3), 4.51 (1H, t, J = 6 Hz, H-1), 6.83−6.87 (m, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) d 10.1 (q, C-3), 31.7 (t, C-2), 55.6 and 55.7 (q, −OCH_3 on C-3′ and C-4′), 75.7 (d, C-1), 108.8 (d, C-2′), 110.7 (d, C-5′), 118.1 (d, C-6′), 137.1 (s, C-1′), 148.2 (s, C-4′), 148.8 (s, C-3′); EIMS *m/z* [M$^+$] 196 (19), 167 (80), 139 (100), 124 (52), 95(30), 77 (65), 53 (69), 39 (80); HREIMS *m/z* 196.1093 (calcd for $C_{11}H_{16}O_3$, 196.1099); HPLC (MeOH−H_2O) t_R = 4.57 min (100%); HPLC (MeCN−H_2O) t_R = 3.92 min (98.4%).

1-(9′-Phenanthryl)-2-methyl-1-propanol (31). Compound **31** was obtained from 9-phenanthraldehyde (Sigma) (383.2 mg, 1.86 mmol) and a 2.0 M solution of isopropyl Mg chloride (Aldrich) (5 mL, 10 mmol), following the procedure described for compound **18**, as a white solid (386.8 mg, 1.54 mmol, 83% yield): mp 106−107 °C; IR (KBr) ν_{max} 3589, 3577, 3064, 2956, 1473, 1010 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.00 (6H, d, J = 6 Hz, H-3), 2.05 (1H, d, H-1, J = 4 Hz), 2.35 (1H, m, H-2), 5.22 (1H, brs, 1H, −OH), 7.26−8.76 (m, 9 H, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 16.9 and 20.2 (q, C-3 and 2-methyl), 33.7 (d, C-2), 76.5 (d, C-1), 122.3, 123.2, 124.0, 124.2, 126.0, 126.3, 126.4, 126.5, 128.6 (d, C 1′-8′, C-10′), 129.7, 129.8, 130.6, 131.2, 137.6 (s, C-4′a, 4′b, 8′a, 9′, 10′a); EIMS *m/z* [M$^+$] 250 (22), 207 (100), 179 (55), 69 (20), 41 (30); HREIMS *m/z* 250.1429 (calcd for $C_{18}H_{18}O$, 250.1358); *anal.* C 86.28%, H 7.27%, calcd for $C_{18}H_{18}O$, C 86.35%, H 7.25%.

1-(9′-Phenanthryl)-1-propanone (29). Compound **29** was obtained by oxidation of 1-(9′-phenanthryl)-1-propanol (330.5 mg, 1.4 mmol) (obtained in turn by a Grignard reaction of 9-phenanthraldehyde and ethyl Mg bromide) with Jones' reagent as a white solid (320.7 mg, 1.37 mmol, 98% yield): mp 55.5−56.5 °C IR (KBr) ν_{max} 2980, 2870, 1602 cm^{-1}, ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.33 (3H, t, J = 6 Hz, H-3), 3.13 (2H, q, H-2, J = 6 Hz), 7.63−8.65 (9 H, m, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 6.6 (q, C-3), 35.4 (t, C-2), 122.6, 122.8, 126.5, 127.0, 127.3, 127.4, 128.5, 128.6, 129.5 (d, C 1′-8′, C-10′), 128.3, 130.0, 130.7, 131.6, 135.5 (s, C-4′a, 4′b, 8′a, 9′, 10′a), 205.4 (s, C-1); EIMS *m/z* [M$^+$] 234 (32), 205 (100), 177 (56), 151 (10), 88 (32), 69 (78); HREIMS *m/z* 234.1123 (calcd for $C_{17}H_{14}O$, 234.1045); *anal.* C 87.05%, H 6.05%, calcd for $C_{17}H_{14}O$, C 87.14%, H 6.03%.

1-(9′-Phenanthryl)-2-methyl-1-propanone (30). Compound **30** was obtained by oxidizing the secondary alcohol **31** (200 mg, 0.8 mmol) with Jones' reagent (188.5 mg, 0.76 mmol, 95% yield) as a white solid: mp 47−48 °C; IR (KBr) ν_{max} 2970, 2920, 2890, 1605 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.26 (6H, d, J = 6 Hz, H-3), 3.60 (1H, m, H-2), 7.26−8.75 (m, 9 H, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 18.5 (q, C-3 and 2-methyl), 39.7 (d, C-2), 122.5, 122.7, 126.3, 126.9, 127.0, 127.1, 127.3, 128.2, 129.3 (d, C 1′-8′, C-10′), 128.7, 130.0, 130.6, 131.3, 136.1 (s, C-4′a,4′b, 8′a, 9′, 10′a), 209.0 (s, C-1); EIMS m/z [M$^+$] 248 (20), 205 (100), 177 (49), 151 (9), 88 (13), 69 (78); HRFABMS m/z 249.1274 (calcd for $C_{18}H_{17}O$ [M+H]$^+$, 249.1279); *anal.* C 86.82%, H 6.57%, calcd for $C_{18}H_{16}O$, C 87.05%, H 6.50%.

1-(9′-Phenanthryl)-2-chloro-1-propanone (32). Compound **32** was prepared from ketone **29** (200 mg, 0.81 mmol), Cu(II) chloride dihydrate (236.5 mg, 1.4 mmol), lithium chloride (46.2 mg, 1.1 mmol), and DMF (3 mL), following the procedure described previously for compound **9**. Compound **32** (187.6 mg, 0.7 mmol, 87% yield) was obtained as a white solid: IR (KBr) ν_{max} 3010, 2940, 1695 cm^{-1}; ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.64 (3H, d, 3H, J = 7 Hz, H-3), 5.41 (1H, q, J = 7 Hz, H-2), 7.70−8.72 (9 H, m, H-1′-7′, 10′, ArH); ^{13}C NMR ($CDCl_3$, 50 MHz) δ 20.3 (q, C-3), 56.4 (d, C-2), 122.7−122.8, 126.3, 127.2, 127.3, 127.6, 128.6, 129.0, 129.7 (d, C 1′-8′, C-10′), 130.2, 131.1, 132.2, 133.8, 134.0 (s, C-4′a,4′b, 8′a, 9′, 10′a), 197.33 (s, C-1); EIMS m/z 268 (32), 205 (100), 177 (56), 90 (31); HREIMS m/z 268.0729 (calcd for $C_{17}H_{13}OCl$ 268.0655); *anal.* C %, H %, calcd for $C_{17}H_{13}OCl$; C 76.10%, H 4.89%, Cl 13.04%; HPLC analysis, the observed t_R values for compound **32** in two different solvent systems ($MeOH-H_2O$, 70:30 and CH_3CN-H_2O 60:40) were 5.38 min (100%) and 4.56 min (98.7%), respectively.

Microorganisms and Media. The following microorganisms used for the fungistatic evaluation were purchased from American Type Culture Collection (Rockville, MD): *C. albicans* ATCC 10231, *S. cerevisiae* ATCC 9763, *C. neoformans* ATCC 32264, *A. flavus* ATCC 9170, *A. fumigatus* ATCC 26934, and *A. niger* ATCC 9029. Strains were grown on Sabouraud chloramphenicol agar slants for 48 h at 30 °C. Cell suspensions in sterile distilled water were adjusted to give a final concentration of 106 viable yeast cells/mL.[34] Dermatophytes: *M. canis* C 112, *T. rubrum* C 113, *E. floccosum* C 114, and *M. gypseum* C 115 are clinical isolates and were kindly provided by CEREMIC, Centro de Referencia Micológica, Facultad de Ciencias Bioquímicas y Farmacéuticas (Suipacha 531, 2000 Rosario, Argentina). *T. mentagrophytes* was ATCC 9972. Organisms were maintained on slopes of Sabouraud dextrose agar (SDA, Oxoid) and subcultured every 15 days to prevent pleomorphic transformations. Spore suspensions were obtained according to reported procedures[34] and adjusted to 10^6 spores with colony-forming ability per mL.

Antifungal Assays. The fungistatic activity of phenylpropanoids was evaluated with the agar dilution method by using Sabouraud chloramphenicol agar for both yeast and dermatophyte species. The assay was carried out in 96-well microtiter plates. Stock solutions of phenylpropanoids in DMSO were diluted to give serial two-fold dilutions that were added to each medium resulting in concentrations ranging from 0.10 to 50 μg/mL. The final concentration of DMSO in the assay did not exceed 2%. Using a micropipet, an inoculum of 5 μL of the yeast cell or spore suspensions was added to each Sabouraud chloramphenicol agar well. The antifungal agents ketoconazole (Janssen Pharmaceutica) and amphotericin B (Sigma Chemical Co) were included in the assay as positive controls. Drug-free solution was also used as a blank control. The plates were incubated 24, 48, or 72 h at 30 °C (according to the control fungus growth) up to 15 days for dermatophyte strains. MIC was defined as the lowest compound concentration showing no visible fungal growth after incubation time.

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