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Antifungal and antioxidant activity of fatty acid methyl esters from vegetable oils

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ABSTRACT

Fatty acid methyl esters (FAMEs) were obtained from vegetable oils of soybean, corn and sunflower. The current study was focused on evaluating the antifungal activity of FAMEs mainly against *Paracoccidioides* spp., as well as testing the interaction of these compounds with commercial antifungal drugs and also their antioxidant potential. FAMEs presented small IC_{50} values (1.86-9.42 µg/mL). All three FAMEs tested showed antifungal activity against isolates of *Paracoccidioides* spp. with MIC values ranging from 15.6-500 µg/mL. Sunflower FAMEs exhibited antifungal activity that extended also to other genera, with an MIC of 15.6 µg/mL against *Candida glabrata* and *C. krusei* and 31.2 µg/mL against *C. parapsilosis*. FAMEs exhibited a synergetic effect with itraconazole. The antifungal activity of the FAMEs against isolates of *Paracoccidioides* spp. is likely due to the presence of methyl linoleate, the major compound present in all three FAMEs. The results obtained indicate the potential of FAMEs as sources for antifungal and antioxidant activity.

Key words: antifungal, antioxidant, fatty acid methyl esters, vegetable oils.

INTRODUCTION

Antioxidant compounds are capable of inhibiting or delaying lipid oxidation, which is associated with the appearance of degenerative or chronic diseases. Thus, antioxidants act to prevent or avoid incidence of cancer, diabetes, atherosclerosis, coronary heart disease and the aging process (Rufino et al. 2011). Natural substances with known antioxidant potential are phenolic compounds (flavonoids and phenolic acids), vitamins A, C, E and carotenoids which, in different proportions and quantities, can be obtained from foods of plant origin, such as vegetables, fruits, teas, herbs, beans and oil seeds (Ara and Nur 2009, Arnao et al. 2001). Several studies have suggested that the development of chronic or degenerative diseases can be decreased with the regular consumption of vegetables or fruits (Chu et al. 2002). Therefore, in recent years, the search for natural antioxidants, especially of plant origin, has greatly increased (Lima et al. 2010).

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Paracoccidioidomycosis (PCM) is a systemic mycosis caused by fungi of the genus *Paracoccidioides*, and two species are known, *P. brasiliensis* and *P. lutzii*. PCM is a disease limited to Latin America and is a common cause of deaths from fungal diseases. PCM is the eighth most important cause of mortality from chronic infectious diseases reaching rates of 1.65 deaths per 10^6 habitants (Bocca et al. 2013, Coutinho et al. 2002, Prado et al. 2009). There are some drugs available to treat PCM; itraconazole therapy is the first choice to control the mild to moderate clinical forms but this therapy is not easily available in most of the endemic regions. Consequently, sulfamethoxazole-trimethoprim (SMX-THT) is a useful option; this drug is freely distributed by the Brazilian Ministry of Health. The main disadvantage of SMX-THT is the need for long-term treatment (more than 12 months) in moderate and severe cases which can lead patients to abandon treatment (Brummer et al. 1993, Paniago et al. 2003, Travassos et al. 2008). Amphotericin B therapy is the best choice for severe cases of PCM, but toxicity, mainly nephrotoxicity, is related to this drug which will sometimes require discontinuation of this therapy (de Oliveira et al. 2015, Ferreira 2009, Shikanai-Yasuda 2015). Relapses, a common event in PCM patients, associated with toxicity represent an unresolved problem in the conventional therapeutic approach (Travassos et al. 2008, Travassos and Taborda 2012). Due to these facts it is necessary to research new drugs that are safer, more effective and cheaper and with shorter periods of therapy for the treatment of PCM.

Fatty acids are the most abundant component of oils, the most commonly found being stearic, palmitic, oleic, linoleic and linolenic acids (Cabral 2005). The antibacterial and antifungal properties of vegetable oils are reported in the literature, these being especially attributed to the presence of fatty acids (Desbois and Smith 2010, Erdemoglu and Kusmenoglu 2003). However, some works done with methyl esters showed their potential as antifungals (Abdelillah et al. 2013, Agoramoorthy et al. 2007, Chandrasekaran et al. 2011, Golebiowski et al. 2013, Lima et al. 2011), but these works are still scarce, so it is important to test these compounds. Fatty acid methyl esters (FAMEs) are obtained from vegetable oils. This work is focused on evaluating the antifungal activity of FAMEs from vegetable oils against *Paracoccidioides* spp., and also antioxidant potential by the scavenging effect on 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals.

MATERIALS AND METHODS

CHEMICALS

Soybean, corn and sunflower vegetable oils were obtained from ABC Industry, Trade SA and Caramuru Foods SA (Brazil), respectively. All PA and HPLC grade reagents used were purchased from Vetec (Brazil) and Sigma (St. Louis, USA), respectively. Amphotericin B, 2,6-bis(1,1-dimethylethyl)-4-methylphenol (BHT), DPPH, 2',7'-dichlorofluorescein diacetate, ascorbic acid (AA), methyl palmitate, methyl stearate, methyl oleate, methyl linoleate, methyl linolenate, palmitic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, synthetic medium RPMI and morpholine propanesulfonic acid (MOPS) from Sigma-Aldrich (St. Louis, MO, USA). Dihidrorhodamine 123 was obtained from Invitrogen (USA). Sabouraud dextrose agar was purchased from Oxoid (Basingstoke, UK), SMX-THT from Roche (Rio de Janeiro, Brazil) and fluconazole from Pfizer Pharmaceutical (USA).

PREPARATION OF METHYL ESTERS

Vegetable oils (1 g) were refluxed with 1.0 mol/L methanolic sodium hydroxide solution for 30 min and then extracted with ethyl ether. The aqueous phase was acidified with 1.0 mol/L hydrochloric acid solution and extracted with ethyl ether. The

organic phase was dissolved in hexane and then refluxed with 2% v/v sulfuric acid methanolic solution for 60 min. After extraction and solvent elimination FAMEs were obtained (Lima et al. 2011).

ANALYSIS OF METHYL ESTERS BY GAS CHROMATOGRAPHY–MASS SPECTROMETRY (GC-MS)

Analysis of FAMEs by GC-MS was performed on Shimadzu CG-MS QP5050A apparatus, using impact electron at 1.2 kV, helium as the carrier gas and a Supelco PTE-5 column (30 m × 0.25 mm, df 0.25 μm). The temperature was maintained at 120 °C and programmed to 300 °C with increments of 5 °C/min; injection and detector temperatures were 230 °C. The split ratio was 1/10. The mass range was 40–500 *m/z* and flow rate was 2 mL/min. FAMEs were identified by using the NIST 2.0 Library database search and by comparison of their retention times with those of standards.

DPPH RADICAL-SCAVENGING ASSAY

The radical-scavenging ability of FAMEs was analyzed compared to standards BHT and AA based on reaction with DPPH radicals. The DPPH method was adapted for use with microplates (Araújo et al. 2013). A solution of DPPH (0.002% w/v) was prepared in 80% methanol. Exactly 75 μL of sample or standard (1, 10, 100, 250 and 500 μg/mL) was added to 150 μL of DPPH, and the microplate was then covered and left in the dark (once) at room temperature (25 °C); 80% methanol was used for baseline correction. The absorbance was measured in a spectrophotometer (BioTek Power Wave XS2/US) at 517 nm, after 30 min. Scavenging ability was calculated by the following equation (Burda and Oleszek 2001): , where $Abs_{control}$ = absorbance of DPPH radicals and Abs_{sample} = absorbance of samples or standards + DPPH. The antioxidant activity was expressed as IC_{50} (concentration of samples necessary to inhibit by 50% the formation of DPPH radicals, in μg/mL). Probit analysis (Finney 1980) was used to calculate the IC_{50} values. All assays were performed in triplicate.

CULTURE AND MAINTENANCE OF THE FUNGAL ISOLATES

In this study we used eight isolates of *Paracoccidioides brasiliensis* representing the three phylogenetic species (PS1, PS2 and PS3) and three isolates of *P. lutzii*, members of the collection of the Microorganism–Host Interaction Laboratory, Biological Science Institute of Universidade Federal de Minas Gerais (UFMG). *Paracoccidioides* spp. were maintained in YPD (yeast, peptone and dextrose) and subcultures were performed after 7 days of growth at 37 °C. *Candida albicans* ATCC 18804, *C. glabrata* ATCC 2001, *C. krusei* ATCC 200298, *C. parapsilosis* ATCC 22019, *C. tropicalis* ATCC 22019, *Cryptococcus gattii* ATCC 24065 and *C. neoformans* ATCC 24067 were used in the biological assays. The species of *Candida* and *Cryptococcus* were stored frozen at −80 °C.

The inoculum of *Paracoccidioides* was done according Hahn and Hamdan (2000). The resulting suspensions were diluted in RPMI 1640 (1:10) to obtain the final inoculum with 1–5 × 10^5 yeasts/mL (Cruz et al. 2013). For *Candida* spp. and *Cryptococcus* spp., the inoculum was obtained with a final concentration of 0.5–2.5 × 10^3 cells/mL for susceptibility testing (CLSI 2008).

ANTIFUNGAL ACTIVITY

The antifungal activity was evaluated by the minimal inhibitory concentration (MIC) in accordance with the guidelines from the CLSI M27-A3 document (CLSI 2008). The MIC for *Paracoccidioides* was performed according to the method described by Johann et al. (2010). FAMEs were tested in the range of 2000–7.81 μg/mL, amphotericin B was tested in the range of 1–0.008 μg/mL and SMX-THT (4.49 × 10^{-3} to 2.3 μmol/mL) was tested in

the range of 600–4.6 μg/mL. The MIC values correspond to the lowest concentrations that did not allow for the detection of any visual fungal growth. All assays were performed in triplicate and repeated at least once.

ASSESSMENT OF DRUG INTERACTIONS

The assessment of drug interactions of FAMEs and positive controls (amphotericin B, SMX-THT and itraconazole) were performed according to the described methods (Pyun and Shin 2006, White et al. 1996), using eight serial dilutions of each compound. These dilutions were added to 96-well plates in several combinations of concentrations of the two compounds tested. After that, a suspension of Pb-18, done according to the MIC test, was added to all wells of the plate and cultured for 14 days. The fractional inhibitory concentration (FIC) was obtained by dividing the MIC of the positive control in the presence of FAMEs by the MIC of the positive control alone. The FIC of FAMEs was calculated in the same way. The FIC index (FICI) was obtained by adding both FICs. Synergistic activity was observed when the FICI was ≤ 0.5, an indifferent effect in the range 0.5 < FICI < 2.0 and an antagonistic effect when the FICI was > 2.0.

CELLULAR REACTIVE OXYGEN SPECIES (ROS)

The *P. brasiliensis* Pb-18 was cultured in YPD agar during days at 37 ºC. Microplate wells received 100 μL inoculum containing 1 x 10^5 cfu/mL in RPMI medium without phenol red, and then the probes were added. Briefly, the probe for reactive oxygen species (ROS) (2',7'-dichlorofluorescein diacetate) and reactive nitrogen species (RNS) (dihidrorhodamine 123) were prepared in methanol and PBS, respectively, at 100 μM and 20 μL were distributed to each well for a final concentration of 10 μM. Hydrogen peroxide (4M) was included as a control. The drug treatment solutions were evaluated at the respective MIC, in a final volume of 200 μL. The plates were incubated at 37 ºC in the dark. After 120 hour, the fluorescence was measured with a fluorometer (Biotek Synergy 2 SL Luminescence Microplate Reader/US) using excitation at 485 nm and emission wavelengths of 530 nm.

STATISTICAL ANALYSIS

Student's t test was utilized to evaluate the statistical difference between the control group and the group exposed to FAMEs. To evaluate the statistical difference between the three FAMEs tested we used the Newman–Keuls test and ANOVA (1 way) to cellular ROS. A P value < 0.05 was considered statistically significant. The analyses were performed using Prisma 5.0 software.

RESULTS AND DISCUSSION

ANTIOXIDANT ACTIVITY

In this study, FAMEs obtained from vegetable oils were investigated by GC-MS. The analysis by GC-MS revealed a high percentage of unsaturated methyl esters (83.95–88.33%) as compared to saturated methyl esters (11.67–16.05%). In samples, methyl oleate and methyl linoleate were detected as the major unsaturated methyl esters. Methyl palmitate was the most abundant among the saturated methyl esters, followed by methyl stearate (Table I).

The scavenging effect on DPPH radicals of FAMEs from vegetable oils is presented in Figure 1. FAMEs, BHT and AA revealed an inhibitory effect in a concentration-dependent manner. FAMEs were more effective in scavenging DPPH radicals than AA at a concentration of 1 μg/mL and BHT at concentrations of 1 and 10 μg/mL. FAMEs showed IC_{50} values of 1.86, 9.42 and 3.33 μg/mL for soybean, corn and sunflower oils, respectively, lower than BHT (16.36 μg/mL; $P < 0.05$), but higher than AA (1.62 μg/mL; $P < 0.05$). FAMEs obtained from *Annona cornifolia* seeds also presented

TABLE I
Composition and percentages of fatty acid methyl esters (FAMEs).

Peak No.	Fatty acids methyl esters	Soybean	Corn	Sunflower	*Annona cornifolia*[1]
1	Methyl palmitate	12.71	12.92	6.99	16.9
2	Methyl stearate	3.34	2.03	4.68	5.6
3	Methyl oleate	28.28	36.67	22.40	51.5
4	Methyl linoleate	55.67	48.38	65.93	19.1
5	Methyl myristate	-	-	-	0.2
6	Methyl linolenate	-	-	-	0.8
Total		100.00	100.00	100.00	94.1

[1]Lima et al. 2011.

antioxidant activity, with IC_{50} = 3.83 µg/mL (Lima et al. 2012). Soybean and sunflower FAMEs were more active than the FAMEs of *Annona cornifolia*.

ANTIFUNGAL ACTIVITY

The antifungal activity of FAMEs obtained from vegetable oils was evaluated against 18 fungal strains of clinical interest (Table II). Results showed that all FAMEs tested showed antifungal activity against the isolates of *Paracoccidioides* tested, with values between 15.6 and 500 µg/mL. *Paracoccidioides brasiliensis* Penguin showed more susceptibility to soybean and sunflower FAMEs with an MIC value of 15.6 µg/mL, but this isolate was less susceptible to corn FAMEs (MIC = 500 µg/mL). For *P. lutzii*, isolate 1578 was most susceptible to the FAMEs tested in the present study. Moreover, FAMEs (soybean, corn and sunflower) showed better activity than SMX-THT (P = 0.0001) against the isolates of *P. brasiliensis* Pb-18, Pb-608, Pb-1017, Pb-9673 and *P. lutzii* 01. SMX-THT is used in the treatment of PCM, mainly in endemic regions where other therapy (such as itraconazole) is not easily available; consequently, therapy consisting of SMX-THT is a useful option (de Oliveira et al. 2015, Shikanai-Yasuda 2015). The difference in antifungal activity between different FAMEs, against 11 isolates of *Paracoccidioides* spp., is not statistically significant ($P < 0.05$). MIC values found for SMX-THT and amphotericin B against the isolates of *Paracoccidioides* were statistically significant when compared with FAMEs (P = 0.0001).

It is interesting to note that when we tested the susceptibility of other fungus genera we observed that only sunflower FAMEs showed activity under the conditions tested in the present study (Table II). This antifungal activity (sunflower FAMEs) was more pronounced against *C. krusei*, *C. glabrata* and *C. parapsilosis* with MIC values of 15.6, 15.6 and 31.2 µg/mL, respectively.

Few works reporting the biological activity of FAMEs against pathogenic fungi are found in the literature. Twelve clinical strains of the fungus *P. brasiliensis* were susceptible to FAMEs from seeds of *A. cornifolia*, with MICs in the range between 3.4 and 55.5 µg/mL (Lima et al. 2011). The FAME extract of *Excoecaria agallocha* presented antifungal activity against *Candida albicans*, *C. krusei*, *C. parapsilosis* and *Candida tropicalis* (Agoramoorthy et al. 2007).

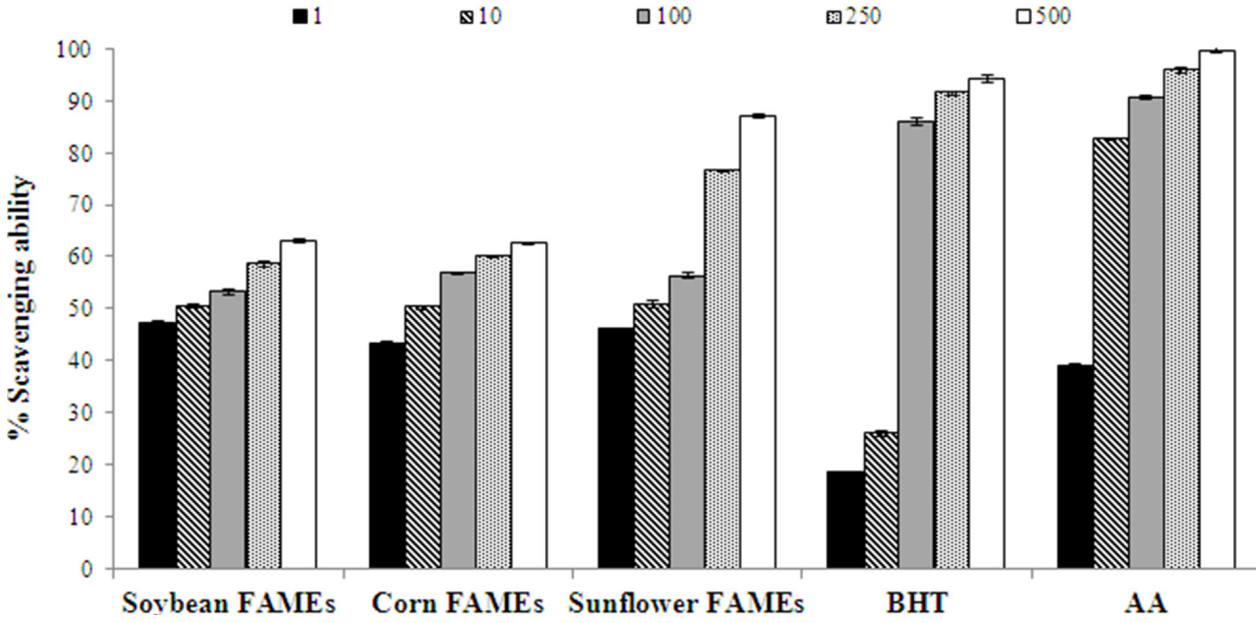

Figure 1 - DPPH radical-scavenging ability of fatty acid methyl esters (FAMEs) at five different concentrations (1, 10, 100, 250 and 500 μg/mL). Values represent averages ± standard deviations for triplicate experiments. BHT: 2,6-bis(1,1-dimethylethyl)-4-methylphenol; AA: ascorbic acid.

Golebiowiski et al. (2013) reported the antifungal activity of FAMEs from *Calliphora vomitoria* against entomopathogenic fungi, protecting this ectoparasite against entomopathogenic microorganisms present in its habitat. Furthermore, these authors showed that when individual FAMEs were tested they presented weak antifungal activity, but the mixture of FAMEs found in living forms of *C. vomitoria* was more active. These experiments are in agreement with those of the present study where the mixture of FAMEs presented antifungal activity.

In the present work we also tested the antifungal activity of standards of the methyl esters which were present in the FAMEs (Table III). Of these methyl esters tested only methyl linoleate showed antifungal activity against *P. brasiliensis* (Pb-18), with an MIC value of 62.5 μg/mL (Table III). It is interesting to note that sunflower FAMEs had a higher percentage of methyl linoleate (65.93%) and also presented better antifungal activity, with 54.54% of isolates of *Paracoccidioides* tested with an MIC value ≤ 62.5 μg/mL. The other two FAMEs tested also followed this pattern; soybean FAMEs containing 55.67% methyl linoleate showed antifungal activity with an MIC value ≤ 62.5 μg/mL for 36.36% of isolates of *Paracoccidioides* tested and corn FAMEs (48.38% methyl linoleate) with 27.27% of isolates tested with an MIC value ≤ 62.5 μg/mL. Probably, methyl linoleate is responsible for the antifungal activity of the FAMEs studied, because it is also present in a greater concentration in all the FAMEs tested (Table I). In present work we also observed the production of reactive oxygen (ROS) by methyl linoleate, sunflower and corn FAMEs (Figure 2) with production significantly compared to the control group (*P. brasiliensis* 18 without treatment) ($P < 0.05$). Methyl linoleate is able to produce hydroperoxide (Yamamoto et al. 1984) and probably this property is responsible for the antifungal activity of this compound in the present work. Therefore the ROS production could be related with antifungal activity of sunflower and corn FAMEs and methyl linoleate compound

TABLE II

Minimal inhibitory concentration (MIC) of FAMEs against 18 clinically important fungi.

Fungi	Soybean FAMEs	Corn FAMEs	Sunflower FAMEs	Amphotericin B	SMX-THT
	MIC µg/mL	MIC µg/mL	MIC µg/mL	MIC µg/mL	MIC µg/mL
Candida albicans	≥ 2000	≥ 2000	500	0.25	-
Candida glabrata	≥ 2000	≥ 2000	15.6	0.125	-
Candida krusei	250	≥ 2000	15.6	0.5	-
Candida parapsilosis	≥ 2000	≥ 2000	31.2	0.5	-
Candida tropicalis	≥ 2000	≥ 2000	≥ 2000	1.0	-
Cryptococcus gattii	≥ 2000	2000	125	1.0	-
Cryptococcus neoformans	≥ 2000	≥ 2000	1000	1.0	-
*Paracoccidioides brasiliensis P*18	125	125	62.5	0.25	300
Paracoccidioides brasiliensis 470	125	250	125	0.125	75
Paracoccidioides brasiliensis EPM83	125	250	62.5	0.125	150
Paracoccidioides brasiliensis 608	31.2	62.5	62.5	0.015	300
Paracoccidioides brasiliensis AP	125	125	125	0.125	75
Paracoccidioides brasiliensis 1017	125	125	62.5	0.062	150
Paracoccidioides brasiliensis Pinguim	15.6	500	31.2	0.125	300
Paracoccidioides brasiliensis 9673	62.5	62.5	62.5	0.062	300
Paracoccidioides lutzii 01	125	250	250	0.125	300
Paracoccidioides lutzii ED01	125	250	250	0.031	75
Paracoccidioides lutzii 1578	62.5	62.5	125	0.062	75

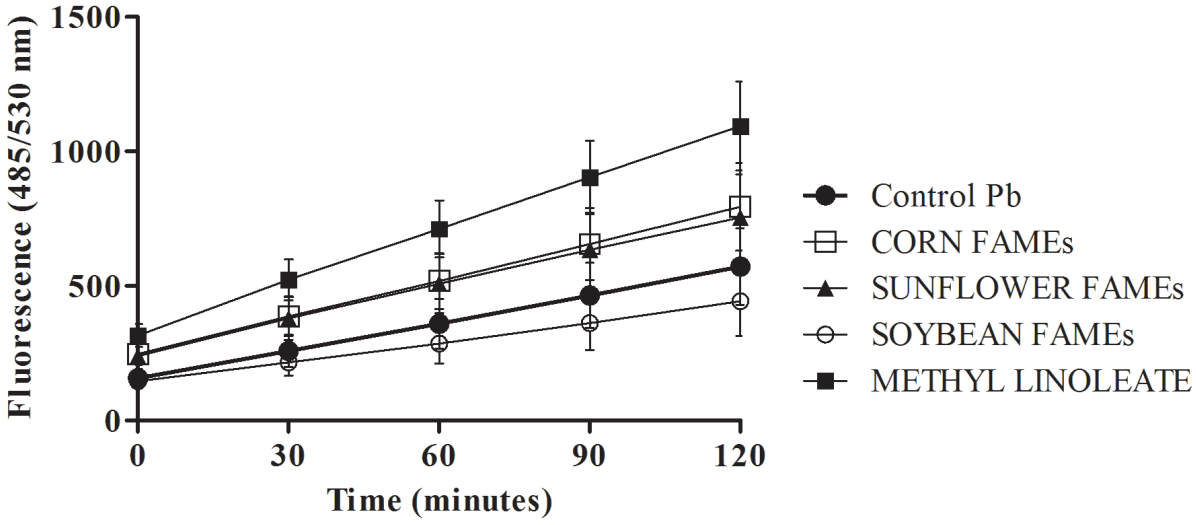

Figure 2 - Production of reactive oxygen species (ROS) in *Paracoccidioides brasiliensis* after treatment with FAMEs and methyl linoleate. Treatments with Corn and Sunflower FAMEs increased ROS production significantly compared to the control group ($P < 0.05$) One-way analysis of variance (ANOVA) and Kruskal-Wallis multiple-comparison tests.

TABLE III
Minimal inhibitory concentration (MIC) of standards of fatty acids and methyl esters against important fungi.

Sample	*C. neoformans* ATCC 24067	*C. gatti* ATCC 24065	*C. albicans* ATCC 18804	*P. brasiliensis* Pb18
	MIC µg/mL	MIC µg/mL	MIC µg/mL	MIC µg/mL
Palmitic acid	≥ 2000	≥ 2000	≥ 2000	≥ 2000
Stearic acid	≥ 2000	≥ 2000	≥ 2000	≥ 2000
Oleic acid	≥ 2000	≥ 2000	≥ 2000	250
Linoleic acid	≥ 2000	≥ 2000	≥ 2000	7.8
Methyl palmitate	≥ 2000	≥ 2000	≥ 2000	≥ 2000
Methyl stearate	≥ 2000	≥ 2000	≥ 2000	≥ 2000
Methyl oleate	≥ 2000	≥ 2000	≥ 2000	≥ 2000
Methyl linoleate	≥ 2000	≥ 2000	≥ 2000	62.5
Amphotericin B	1.0	1.0	0.25	0.25
SMX-THT	-	-	-	300

present in these FAMEs could be responsible for ROS production.

Methyl linoleate has already been tested against inhibition of the *in vitro* proliferation of human tumor cell lines; this compound showed antiproliferative activity with an IC_{50} of 250 µg/mL, but not induction of differentiation. This is an interesting finding, because 5-fluorouracil is also a known antitumor compound that does not induce differentiation and is currently used as an antifungal (Lampronti et al. 2003).

In the present work we also tested the antifungal activity of standards of fatty acids (Table III) because these compounds were present in the oils (sunflower, soybean and corn oils) that originated the FAMEs. Although two of these compounds (oleic acid and linoleic acid) exhibited interesting antifungal activity against Pb-18, the oils did not present activity (data not shown).

To explore the possibility of developing more powerful combination therapies of FAMEs with other antifungal drugs, a checkerboard micro-titer test was carried out. FAMEs showed an antagonistic effect with amphotericin B and an indifferent effect with SMX-THT (Table IV). FAMEs showed a synergetic effect with itraconazole. This illustrates one important result, because it could lead to the development of a new combination therapy using lower concentrations of itraconazole in a shorter treatment time, which would reduce the side effects of patients. Itraconazole interferes in ergosterol synthesis, leading to cell membrane perturbation (Odds et al. 2003). Alteration in membrane permeability could allow increased concentrations of FAMEs to penetrate the cell, resulting in cell death.

CONCLUSIONS

In the search for new drugs with antifungal and antioxidant properties, FAMEs (obtained from vegetable oils of soybean, corn and sunflower) might be good candidates: these esters presented better radical-scavenging activity and IC_{50} values than BHT, a commercial antioxidant. All three FAMEs tested showed antifungal activity against isolates of *Paracoccidioides* spp. with MIC values ranging from 15.6–250 µg/mL. Sunflower

TABLE IV
Fractional inhibitory concentration (FIC) and FIC index (FICI) of FAMEs against *Paracoccidioides brasiliensis* isolate Pb-18.

Compounds	MIC in combination µg/mL	MIC alone µg/mL	FIC	FICI
1. Soybean FAMEs	3.9	125	0.031	4.0312
2. Amphotericin B	1.0	0.25	4	
1. Soybean FAMEs	3.9	125	0.0312	0.5312
2. SMX-THT	150	300	0.5	
1. Soybean FAMEs	3.9	125	0.0312	0.0462
2. Itraconazole	0.0009	0.06	0.0150	
1. Corn FAMEs	3.9	125	0.0312	8.0312
2. Amphotericin B	2	0.25	8	
1. Corn FAMEs	3.9	125	0.0312	0.5312
2. SMX-THT	150	300	0.5	
1. Corn FAMEs	3.9	125	0.0312	0.0612
2. Itraconazole	0.0018	0.06	0.0300	
1. Sunflower FAMEs	3.9	62.5	0.0624	4.0624
2. Amphotericin B	1	0.25	4	
1. Sunflower FAMEs	3.9	62.5	0.0624	0.5624
2. SMX-THT	150	300	0.5	
1. Sunflower FAMEs	3.9	62.5	0.0624	0.0774
2. Itraconazole	0.0009	0.06	0.0150	

FAMEs also exhibited antifungal activity that extended to other genera, with an MIC of 15.6 µg/mL against *Candida glabrata* and *C. krusei* and 31.2 µg/mL against *C. parapsilosis*. FAMEs exhibited a synergetic effect with itraconazole. The antifungal activity of the FAMEs against isolates of *Paracoccidioides* spp. is likely due to the presence of methyl linoleate, the major compound present in all three FAMEs. This work provided knowledge, for the first time, of the antioxidant and antifungal activity of FAMEs obtained from vegetable oils and indicates the potential of FAMEs as a source of antifungal and antioxidant activity.

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