

Essential oil and plant extract of oregano as agents influencing the virulence factors of *Candida albicans*

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Abstract

Candida albicans, a polymorphic yeast, is a physiological component of the human and animal commensal microbiome. It is an etiological factor of candidiasis, which is treated by azole antifungals. Growing resistance to azoles is a reason to look for other alternative treatment options. The pharmacotherapeutic use of plant extracts and essential oils has become increasingly important. In our experiment, *C. albicans* showed susceptibility to four observed plant extracts and essential oils from peppermint (*Mentha piperita*), thyme (*Thymus vulgaris*), sage (*Salvia officinalis*), and oregano (*Origanum vulgare*). Oregano plant extract and essential oil showed the highest antifungal activity, at MIC values of 4.9 mg/mL and 0.4 mg/mL respectively. Therefore, it was subjected to further research on the influence of virulence factors – biofilm formation, extracellular phospholipase production and germ tube formation. Oregano plant extract and essential oil showed an inhibitory effect on the observed *C. albicans* virulence factors at relatively low concentrations. The extract inhibited the adherence of cells at MIC 12.5 mg/mL and essential oil at MIC 0.25 mg/mL. Degradation of the formed biofilm was detected at MIC 14.1 mg/mL for plant extract and at MIC 0.4 mg/mL for essential oil. Extracellular phospholipase production was most effectively inhibited by the essential oil. In particular, the number of isolates with intensive extracellular phospholipase production decreased significantly. Of the 12 isolates intensively producing extracellular phospholipase, only 1 isolate (4.5%) retained intense production. Essential oil caused up to a 100 % reduction in germ tubes formation and plant extract reduced their formation depending on the concentration as follows: 2.6% (0.8 mg/mL), 21.2 % (6.25 mg/mL), and 64.5 % (12.5 mg/mL) compared to the control.

Keywords: biofilm, *Candida albicans*, essential oils, extracellular phospholipase, germ tubes, plant extracts



Introduction

Candida albicans (*C. albicans*) is frequently found as a part of the microbiota of healthy humans and animals. It can be found in the intestinal tract and oral cavity of healthy individuals and is the predominant causative agent of candidiasis, which is the most common opportunistic yeast infection. From animals it is mainly a cow, horse, pig, cat, dog and bird that are susceptible to *Candida* infections (Edelman et al. 2005, Seyedmousavi et al. 2018). The importance of yeast commensalism in the microbiota of a healthy individual lies in the protection of mucous membranes against the overgrowth of pathogenic microorganisms, as well as in the support of the immune system (Kumamoto et al. 2020). Azole antifungals, particularly fluconazole and clotrimazole are preferred in the therapy of mycotic infections, due to their low toxicity and availability in several dosage forms (oral tablets, vaginal creams, or tablets). In addition, repeated use of antifungals may lead to an increase in the pathogen's drug resistance. Therefore, it is necessary to look for new safer antifungal compounds, but the specifics of their inhibitory action on *C. albicans* are not yet known (Pristov et al. 2019). The pathogenicity of *C. albicans* is based on virulence factors. Important for virulence is the conversion of yeast to hyphae (dimorphism). The hyphal forms cause infection compared to the yeast forms, which ensure the spread of yeast. The basic virulence factor of *C. albicans* is biofilm production when the yeast colonizes the surface. The production of hydrolases by pathogen hyphae also plays a key role in cell virulence (Martins et al. 2014).

Plant extracts and essential oils have long been used in the treatment of various diseases. Among the *Plantae* kingdom, the *Lamiaceae* family is a rich source of plants used in cosmetics, aromatherapy, and medicine. The *Lamiaceae* family includes several plant species that have a rich spectrum of medicinal properties from anti-inflammatory through antioxidant, antimicrobial, antiseptic, antiviral to anti-proliferative effects. Such species include *Rosmarinus officinalis*, *Mentha piperita*, *Lavandula angustifolia*, *Salvia officinalis*, *Melissa officinalis*, *Ocimum basilicum*, *Origanum vulgare* and *Hyssopus officinalis* (Raut and Karuppayil 2014, Nagy et al. 2017).

Materials and Methods

Clinical isolates

A total of 25 clinical isolates of *C. albicans* were provided by Department of Medical and Clinical Microbiology (Louis Pasteur University Hospital,

Košice, Slovakia). These were obtained from randomly selected human patients of different ages and both sexes with suspected candidiasis of respiratory tract (14 men and 7 women), intestinal tract (1 man), and vagina (3 women). When providing samples, all privacy protection rules were maintained. Since all data were anonymized, and only the gender and place of sample collection are known, the Ethics Agreement was not required. The presence of *Candida albicans* infection was subsequently confirmed at the named institute. Isolates of *Candida albicans* were identified phenotypically by using HiChrome Candida Differential Agar (Hi Media Laboratories, Mumbai, India) and API ID 32 (bioMérieux, France). As the reference strain, *C. albicans* ATCC 10231 (Czech Collection of Microorganisms, Brno, Czech Republic) was used. Growth conditions were as follows: 35°C on Sabouraud dextrose agar for 24 hours.

Essential oils and plant extracts

Four essential oils (EOs) and plant extracts (PE) of the *Lamiaceae* family herbs were used: peppermint (*Mentha piperita*), sage (*Salvia officinalis*), oregano (*Origanum vulgare*), and thyme (*Thymus vulgaris*) (Calendula company, Nová Lúbovňa, Slovakia). Individual certified EOs were obtained from different parts of the plant such as herb (oregano, thyme and sage) and leaves (peppermint). Components of EOs were detected by gas chromatography (Table 1). Stock solutions of individual EOs were prepared as an emulsion formed by gum arabic constituting 30 % of essential oil. Each of the EOs and PEs was tested in the concentration range from 200 mg/mL to 0.4 mg/mL. Sabouraud-dextrose broth (Hi Media Laboratories Pvt. Ltd., Mumbai, India) enriched with 10 mM glucose (SG) was used as a solvent.

Determination of minimal inhibitory concentration

The standard microdilution method CLSI M27-A3 (2017) with some modifications was used to test the susceptibility of *C. albicans* to selected EOs and PEs. Inoculum (10^3 CFU/mL), from 25 *C. albicans* clinical isolates and reference strain ATCC 10231, were prepared in SG. After 24 hours of incubation at 35°C, the minimum inhibitory concentrations (MICs) were read based on the absence of yeast growth. A 0.15% aqueous solution of resazurin (10 µL) was used to better visualize the results. In further experiments (biofilm formation, extracellular phospholipase production, germ tube test), the EO and PE that showed the best inhibitory effect on yeast growth were used.

Table 1. Compounds of essential oils.

Essential oil – plant species	Main content substances
Oregano (<i>Origanum vulgare</i>)	carvacrol (85.0±3%)
Peppermint (<i>Mentha piperita</i>)	menthol (39.0±1%), menthone (24±0.5%)
Thyme (<i>Thymus vulgaris</i>)	ρ-cymene (40.0±3%), thymol (32.0±2%)
Sage (<i>Salvia officinalis</i>)	1,8-cineole (30.0±1%), thujone (3.0±0.2%), borneol (3.0±0.2%)

Table 2. Classification of biofilm producers.

Biofilm production	Interpretation criteria
none (0)	$OD \leq OD_c$
weak (+)	$OD_c < OD \leq 2xOD_c$
medium (++)	$2xOD_c < OD \leq 4xOD_c$
intensive (+++)	$4xOD_c < OD$

Abbreviations: OD – optical density of sample = average absorbance value of the three measurements; OD_c – cut-off value = OD value of the negative control from the three measurements + 3 x SD (standard deviation) of the negative control

Table 3. Interpretation of phospholipase production by *Candida albicans*.

Production of EPL	Pz Index
none	1
weak	0.99 – 0.90
moderate	0.89 – 0.80
medium	0.79 – 0.70
intensive	< 0.70

Abbreviations: EPL – extracellular phospholipase; Pz – precipitation zone

Anti-biofilm activity of oregano essential oil and plant extract against *C. albicans* clinical isolates

The tested strains were classified into four groups of biofilm producers according to the interpretation criteria in Table 2 (Ruchi et al. 2015). This was performed according to the method described by Jin et al. (2003).

The biofilm-forming strains were subsequently used to test the inhibitory effect of EOs on adhered cells (0-hour biofilm) and on mature biofilm (48-hour biofilm) which was carried out by the method according to Jin et al. (2003) with minor modifications. *Origanum vulgare* extract and EO were tested at a concentration range of 50-0.1 mg/mL and 0.5-0.01 mg/mL respectively, prepared by binary dilution. Ten microliters of resazurin aqueous solution (0.15 %) were used to better visualize the results.

Production of extracellular phospholipase – Egg yolk agar method

To perform the Egg yolk method (Ellepola et al. 2016), a specific Egg yolk agar (13 g SDA, 11.7 g NaCl, 0.11 g CaCl₂, 10% egg yolk emulsion) was pre-

pared. The cell suspension of each strain was prepared in sterile phosphate-buffered saline (PBS) using a spectrophotometer (520 nm, optical density of 1.5). A total of 15 µL were applied on the agar medium in Petri dishes, in duplicate. The plates were incubated at 35°C for 7 days. Extracellular phospholipase (EPL) production was manifested by the formation of a white, dense zone around the colony formed. The activity of EPL production was evaluated as precipitation zone index (Pz):

The Pz was considered positive when a precipitation zone was visible around the colony growth. The value of EPL production was determined by the ratio of the diameter of the colony + the precipitation zone, to the diameter of the colony (Ellepola et al. 2016). Table 3 presents interpretation criteria for the evaluation of EPL activity based on the obtained Pz index. To test the effect of *Origanum vulgare* extract and essential oil on EPL formation, a solution at a concentration of 2 x MIC was added to the suspension of each yeast strain. After 30 minutes of exposure at 35°C, the cells were washed twice with PBS (10 min, 3,000 RPM) to remove residual agents.

Table 4. Interpretative criteria for evaluating the intensity of germ tubes formation.

Intensity of germ tubes production	n
weak (+)	< 25 %
moderate (++)	25 – 50 %
medium (+++)	51 – 75 %
intensive (++++)	76 – 100 %

Abbreviation: n – number of cells forming germ tubes

Table 5. Statistical analysis of MIC (mg/mL) of PEs acting on *Candida albicans* planktonic cells.

Plant extract	min. – max.	x	SD	Mo	Me	MIC50	MIC90
<i>Origanum vulgare</i>	0.4 – 12.5	4.9 ^b	2.69	6.25	6.25	6.25	6.25
<i>Mentha piperita</i>	50 – 100	52 ^a	10	50	50	50	50
<i>Thymus vulgaris</i>	50 – 200	60 ^a	32.27	50	50	50	50
<i>Salvia officinalis</i>	50 – 200	122 ^c	45.83	100	100	100	200

Abbreviations: min. – max. – minimum and maximum MIC value (mg/mL); x – average; SD – standard deviation; Mo – mode; Me – median; MIC50/MIC90 – minimum inhibitory concentration inhibiting 50% or 90% of the total number of isolates; average MIC values with different superscripts are statistically different ***p<0.0001

Table 6. Statistical analysis of MIC (mg/mL) of EOs acting on *Candida albicans* planktonic cells.

Essential oil	min. – max.	x	SD	Mo	Me	MIC50	MIC90
<i>Origanum vulgare</i>	0.4	0.4 ^a	0	0.4	0.4	0.4	0.4
<i>Mentha piperita</i>	0.4	0.4 ^a	0	0.4	0.4	0.4	0.4
<i>Thymus vulgaris</i>	0.4	0.4 ^a	0	0.4	0.4	0.4	0.4
<i>Salvia officinalis</i>	0.4 – 3.13	0.7 ^b	0.58	0.4	0.4	0.4	0.8

Abbreviations: min.-max. – minimum and maximum MIC value (mg/mL); x – average; SD – standard deviation; Mo – mode; Me – median; MIC50/MIC90 – minimum inhibitory concentration inhibiting 50% or 90% of the total number of isolates; average MIC values with different superscripts are statistically different **p<0.001

Germ tube test

The germ tubes formation of 25 clinical isolates was quantified based on the Germ tube test according to the study by Mattei et al. (2013). A few colonies were suspended in 1 mL of sheep serum and incubated for 2 hours at 35°C. For each isolate, the intensity of germ tube formation was determined microscopically (magnification 400x) (Table 4).

The effect of oregano EO and PE on germ tube formation was investigated on selected *C. albicans* strains that formed germ tubes from the moderate to the most intensive. The cell suspensions were prepared from clinical isolates and the reference strain, similarly to the EPL formation assay. *C. albicans* (ATCC 10231) served as a positive control and *C. tropicalis* (ATCC 13803), which does not produce germ tubes, was the negative control. The reduction of germ tube formation after EO/PE exposure was calculated according to the formula, where “sample” means the production of germ

tubes after EO/PE exposure and “control” means the production of germ tubes without EO/PE exposure:

Statistical analysis

The obtained results were evaluated by using MS Excel statistical functions: mean (AVERAGE), standard deviation (SD), mode (Mo) and median (Me). The susceptibility of *C. albicans* to EO and PE was assessed by the statistical program GraphPad Prism 5.0 (GraphPad software Inc. CA, USA) using a one-way ANOVA test, Tukey’s Multiple Comparison Test.

Results

MICs of essential oils and extracts

The antifungal effect of the tested PEs and EOs on *C. albicans* is evaluated in Table 5. Of all tested extracts, the lowest mean of MIC (4.9±2.69 mg/mL)

Table 7. The intensity of biofilm production.

Biofilm production	n (%)
none (0)	1 (4 %)
weak (+)	23 (92 %)
moderate (++)	1 (4 %)
intensive (+++)	0

Explanations: n - number of isolates; ODC = 0.0067

Table 8. Statistical evaluation of the effect *Origanum vulgare* EO and PE on 0-h and 48-h biofilm (mg/mL).

Parameters	0 h-biofilm		48 h-biofilm	
	PE	EO	PE	EO
min. – max.	12.5	0.25	12.5 – 25	0.25 – 0.5
x	12.5	0.25	14.1	0.4
SD	0	0	4.2	0.11
Mo	12.5	0.25	12.5	0.5
Me	12.5	0.25	12.5	0.5
MIC50	12.5	0.25	12.5	0.5
MIC90	12.5	0.25	25	0.5

Abbreviations: min.-max. – minimum and maximum MIC value (mg/mL); x –average; SD – standard deviation; Mo – mode; Me – median; MIC50/MIC90 – minimum inhibitory concentration inhibiting 50% or 90% of the total number of isolates

was found for the *Origanum vulgare* extract. Higher MIC values were recorded for the extracts of *Mentha piperita* (52±10 mg/mL), *Thymus vulgaris* (60±32.27 mg/mL), and *Salvia officinalis* (122±45.83 mg/mL). The MIC50 and MIC90 values for the most effective *Origanum vulgare* extract were 6.25 mg/mL, while values for the *Mentha piperita* and *Thymus vulgaris* extracts were 50 mg/mL. The MIC50 of *Salvia officinalis* PE was 100 mg/mL, while the MIC90 reached 200 mg/mL.

The MIC values of the tested EOs were significantly lower (MIC range: 0.4 mg/mL – 3.13 mg/mL) when compared to PEs (Table 6). Out of the four tested EOs, *Salvia officinalis* showed the weakest effect (MIC = 0.7±0.58 mg/mL, MIC90 = 0.8 mg/mL, MIC50 = 0.4 mg/mL). The other three EOs showed MIC of 0.4 mg/mL. The same value (0.4 mg/mL) was found for both MIC50 and MIC90. Based on these results, only the most effective PE and the corresponding EO were selected for further experiments, specifically PE and EO of *Origanum vulgare*.

Anti-biofilm activity of oregano essential oil and plant extract

The intensity of biofilm production by the tested *C. albicans* strains is documented in Table 7. Twenty-three isolates (92%) showed weak biofilm formation. Moderate biofilm production was detected in one

isolate (4%) and one isolate did not form a biofilm. For further study of the anti-biofilm effect of *Origanum vulgare* EO and extract, 24 isolates (forming the biofilm weakly and moderately) were selected.

Table 8 evaluates the inhibition of biofilm formation in the adherence phase (0-hour biofilm) and the disintegration of mature biofilm (48-hour biofilm) after exposure to oregano EO and PE. The same MIC (12.5 mg/mL) of PE was effective on the 0-h biofilm in all isolates and therefore this value was also found for MIC50 and MIC90. In all isolates, EO showed an inhibitory effect on the 0-h biofilm at MIC of 0.25 mg/mL. Degradation of the 48-hour biofilm by EO was achieved at the MICs ranging from 0.25 mg/mL to 0.5 mg/mL and PE from 12.5 to 25 mg/mL.

Production of extracellular phospholipase

In this sub-study, the determination of potential EPL production was first performed, in all 25 clinical isolates and in the reference strain of *C. albicans*. As shown in Table 9, intense EPL production was found in 12 isolates (48%), medium in 4 strains (16%) and moderate one in 6 isolates (24%). Twenty-two phospholipase-producing isolates were selected for the following experiment in which the inhibitory effect of *Origanum vulgare* EO (2xMIC) on EPL formation was observed.

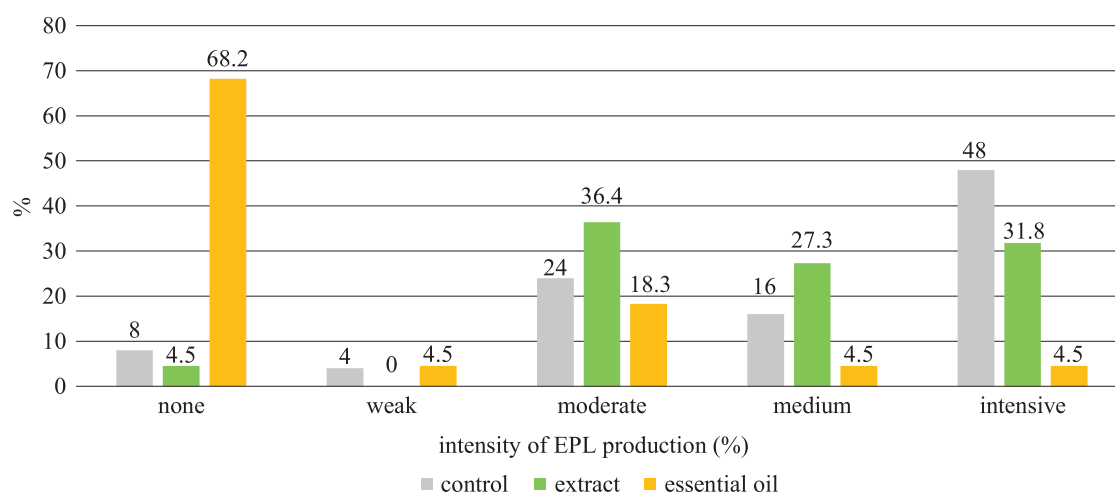


Fig. 1. Phospholipase production by *Candida albicans* clinical isolates after oregano extract and essential oil exposure.

Table 9. Evaluation of phospholipase activity of *Candida albicans* clinical isolates.

Production of EPL	Control	PE	EO
	n	n	n
none	2 (8 %)	1 (4.5 %)	15 (68.2 %)
weak	1 (4 %)	0	1 (4.5 %)
moderate	6 (24 %)	8 (36.4 %)	4 (18.3 %)
medium	4 (16 %)	6 (27.3 %)	1 (4.5 %)
intensive	12 (48 %)	7 (31.8 %)	1 (4.5 %)

Abbreviation: n – number of isolates

Table 10. Statistical evaluation of the Pz index and reduction of EPL formation.

Parameter	Control	PE	EO
min.-max.	0.45 – 1	0.5 – 1	0.59 – 1
x	0.71	0.75	0.94
SD	0.16	0.12	0.11
Reduction (%)	–	5.6	32.4

Abbreviations: min.-max. – minimum and maximum value of the Pz index; x – average of the index Pz; SD – standard deviation

Figure 1 illustrates the inhibitory effect of PE and EO on EPL production. A notable decrease in EPL production was observed after exposure of the cells to EO, the extract appeared to be less effective.

The better efficiency of the EO, in the reduction of EPL formation was confirmed by the average Pz index evaluated in Table 10. In both cases, after exposure of cells to the PE, the reduction in EPL formation reached 5.6%, with a Pz index value of 0.75 ± 0.12 , and after exposure to EO, the reduction in EPL production was 32.4%, with increasing Pz index (0.94 ± 0.11), compared to the control.

Microscopic quantification of germ tubes of *C. albicans* (germ tube test)

Preliminary examination confirmed the formation of germ tubes in all 25 clinical isolates of varying intensity (Table 11).

Isolates with intensive and medium germ tubes formation (48.0 %) were chosen to study the inhibitory effect on germ tubes formation after oregano EO and PE exposure. After exposure to a concentration of 2x MIC (0.8 mg/mL; 6.25 mg/mL; 12.5 mg/mL), a concentration-depending reduction in germ cell production was observed (Table 12). The highest reduction, compared to the control, was recorded for PE

Table 11. Evaluation of germinating hyphae formation of 25 *Candida albicans* isolates.

Intensity	n (%)
weak (+)	5 (20.0 %)
moderate (++)	8 (32 %)
medium (+++)	6 (24 %)
intensive (++++)	6 (24 %)

Table 12. Inhibition of the germ tubes formation by *Origanum vulgare* PE in three concentrations.

	Control	PE (0.8 mg/ml)	Control	PE (6.25 mg/ml)	Control	PE (12.5 mg/ml)
n/N	215/241	218/251	55/64	44/65	524/584	143/450
%	89.2	86.9	85.9	67.7	89.7	31.8
Reduction (%)	-	2.6	-	21.2	-	64.5

Abbreviations: n – number of cells forming germ tubes; N – total number of cells

Table 13. Evaluation of the influence of *Origanum vulgare* EO on the germ tube formation.

	Control	EO (0.8 mg/mL)
n/N	794/889	0/649
%	89.3	0
Reduction (%)	-	100

Abbreviations: n – number of cells forming germ tubes; N – total number of cells

at a concentration of 12.5 mg/mL (64.5 %). The lowest reduction was found at a concentration of 0.8 mg/mL (2.6 %).

In contrast to the extract, EO was able to completely inhibit the formation of germinating hyphae, and the reduction reached 100 % compared to the control (Table 13).

Discussion

One of the reasons for the increased incidence of candidiasis is the growing resistance of yeast to conventional antifungals, due to their frequent and repeated use. This leads to the constant search for new strategic approaches in the therapy and prophylaxis of candidiasis. The pathogenicity of yeast is conditioned by virulence factors, so their study represents an opportunity to prevent the development of candidiasis and its transmission (Rossoni et al. 2013).

Biofilm plays a key role in the development of *C. albicans* infection when planktonic cells adhere to an abiotic or biotic surface and their hyphal growth begins. The formed biofilm reduces the susceptibility of the pathogen to antifungals. This fact is conditioned by several mechanisms. The biofilm consists of a thinner layer of yeast cells at the bottom and a thicker com-

pact layer of hyphae. As soon as the cell density increases, the pathogen's resistance to antifungals also increases. An essential component of the biofilm is the extracellular matrix, which the cells themselves produce. It reduces the diffusion of administered drug to the target. In some yeasts, the mutation can alter the ergosterol synthesis, especially the ERG11 gene mutation, making azoles ineffective. Efflux pumps in yeast cell membranes also provide biofilm resistance (Silva et al. 2013).

Our pilot study on field isolates of *Candida albicans* was focused on the effect of plant substances on virulence factors – biofilm formation and extracellular phospholipase production and was performed at the phenotypic level. In our experiment, of the 25 clinical isolates tested, biofilm formation of weak or moderate intensity was observed in 23 strains (92%), in contrast to the study by Vitális et al. (2020), where 52% of isolates formed biofilm slightly to intensively. However, Mohandas and Ballal (2011) noted weak biofilm formation in *C. albicans*, similar to our study.

Origanum vulgare EO inhibited the adherence phase of biofilm formation at a lower concentration (0.25 mg/mL) when compared to its inhibitory effect on planktonic cells (0.4 mg/mL) and biofilm (0.4 mg/mL). The cause may be a range of tested concentrations.

The anti-biofilm effect is probably due to the content of carvacrol and thymol as the main components of EO. This fact is supported also by the study by Doke et al. (2014), which reports the anti-biofilm activity of monoterpenes carvacrol and thymol at concentrations of 0.25 and 1 mg/mL for the adherence phase and 1-2 mg/mL for mature biofilm.

There are only a few studies aimed at the anti-biofilm effect of plant extracts, but according to the experiment performed by Lee et al. (2018) the anti-biofilm effect of apigenin, a substance found in *Origanum vulgare*, was observed. This flavonoid can change the permeability of the pathogen's membranes, which also reduces the biofilm mass.

In addition to biofilm, the production of EPL, which acts on host cell membranes, particularly glycerophospholipids, contributes to yeast virulence. Isolates from patients have been found to have higher EPL formation than isolates from non-healthy patients (Mohandas and Ballal 2011).

The intensity of phospholipase activity is assessed on the base of the precipitation zone (complex of calcium with fatty acids) formed around the colony after 7 days of incubation. A complex of calcium with fatty acids arises from the action of phospholipase on phospholipids present in the egg yolk medium (de Souza et al. 2015). Of the isolates tested, up to 92% (23 isolates) were EPL producers, which is similar to the study by Fule et al. (2015), where 81.08% were EPL active of which 56.66% showed intense production.

In our experiment, almost the same percentage of isolates (56.5%) from phospholipase-producing isolates (23 isolates) formed EPL intensively. Brondani et al. (2018) consider *Origanum vulgare* EO to be suitable for reducing EPL production, but with prolonged exposure the anti-enzymatic activity decreases. EO reached a lower Pz index value, but up to 68.2 % of isolates did not form EPL after its exposure. EPL production is concentrated in hyphal apices, indicating a correlation between phospholipase activity and germ tube formation in *C. albicans*. Mattei et al. (2013) found that 95% of tested isolates formed germ tubes (GT). In our experiment, the tested strains showed 100% formation of GT, while 48 % of isolates were evaluated as isolates with moderate and intense formation. After one-hour exposure to the extract, a concentration-dependent reduction in the GT formation was observed.

With increasing concentration of EO, the reduction also increased. *Origanum vulgare* EO caused a reduction of up to 100 %, indicating that it is a suitable agent to prevent the development of infection at a concentration of 0.8 mg/mL. Pozzatti et al. (2010) report further EOs (thyme, cinnamon, ginger, and basil) with the ability to inhibit germ tube formation and found

the inhibitory activity for *Origanum vulgare* EO at a concentration of 0.05-0.2 mg/mL.

Conclusion

The pathogenicity of *Candida albicans* is conditioned by virulence factors, and therefore the study of their influence represents an opportunity to prevent candidiasis. In recent years, the pharmacotherapeutic use of essential oils or plant extracts has become increasingly important. A suitable subject of research is the family *Lamiaceae*, which includes several plants with the potential to make a positive contribution to the treatment of candidiasis. As the results of our study show, the action of selected *Lamiaceae* essential oils had the ability to inhibit the growth of *C. albicans* planktonic cells, in addition, *Origanum vulgare* EO or PE inhibit the yeast adhesion, disintegrate biofilm, and reduce the phospholipase and germ tubes formation. Knowledge of the effect of EOs and PEs on *C. albicans* virulence provides new opportunities for the discovery of antifungal drugs by counteracting the difficulties or failures of conventional *Candida* infections therapy.

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