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Evaluation of propolis and its subproduct as an inhibitor of growth and biofilm formation in vaginal yeast from pregnant women

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Objectives The treatment of vulvovaginal candidiasis (VVC) is still unsatisfactory, especially in pregnant women, being promising to the utilization of alternative therapies. Propolis extract solution (PES) has demonstrated antifungal efficacy and low toxicity. In addition, the subproduct of propolis extract solution (SPES) is produced during the process of preparing PES and is usually discarded, but can still submit substances responsible for biological effects, such as the polyphenols, responsible for the therapeutic activity of propolis. SPES have not been investigated or used as an antimicrobial agent. Thus, the objective of the present study was to investigate the effect of PES and SPES on *Candida* spp. isolated from the vaginal material of pregnant women.
Methods Vaginal samples from 291 pregnant women were collected and cultivated for yeasts, which were identified by the classical method and performing susceptibility tests against PES, SPES and conventional antifungal agents. The anti-biofilm effect and cytotoxicity tests of the PES and SPES were evaluated.
Results In 38.48% (112/291) of culture was positive for *Candida* species. There were patients with two different species, being a total of 115 yeasts (82.61% *C. albicans*; 6.08% *C. glabrata*; 5.22% *C. tropicalis*; 5.22% *C. parapsilosis* and 0.87% *C. krusei*). PES and SPES were effective, even against isolates resistant to conventional antifungal (Table 1) and reduced about 25% *C. tropicalis* biofilm, besides presenting its low toxicity in the concentrations of fungicides.
Conclusion Thus, in addition to the PES, SPES can also be a promising alternative treatment, especially in this population.

Table 1. Minimum inhibitory concentration (MIC) values (μg ml^{-1}) to amphotericin B, nystatin, fluconazole, PES and SPES.

Species (n)	MIC[d] (μg/mL)			
antifungal agente	MIC Range	MIC 50[a]	MIC 90[b]	Mean
***C. albicans* (95)**				
Amphotericin B	0.25 - 1.0	0.5	1.0	0.5
Nystatin	0.125 - 0.5	0.125	0.25	0.125
Fluconazole	0.125 - 4.0	0.125	0.125	0.125
PES[c]	68.35 – 1093.75	136.71	273.43	206.50
SPES[c]	22.29 – 356.71	40.58	22.29	59.36
***C. glabrata* (7)**				
Amphotericin B	0.5 - 2.0	1.0	2.0	1.0
Nystatin	0.06 - 0.125	0.125	0.25	0.125
Fluconazole	0.125 - 4	1.0	4.0	1.0
PES[c]	68.35 – 536.87	68.35	546.87	146.47
SPES[c]	11.14 – 89.17	44.58	89.17	38.21
***C. tropicalis* (6)**				
Amphotericin B	0.5 - 1.0	0.5	1.0	0.5
Nystatin	0.125	0.125	0.125	0.125
Fluconazole	0.125 - 1.0	0.125	1.0	0.125
PES[c]	136.72-1093.75	546.87	1093.75	455.72
SPES[c]	22.29 – 178.35	89.17	178.35	89.17
***C. parapsilosis* (6)**				
Amphotericin B	0.5 - 1.0	0.5	1.0	0.5
Nystatin	0.125	0.125	0.125	0.125
Fluconazole	0.125	0.125	0.125	0.125
PES[c]	273.47–1093.75	273.43	1093.75	410.15
SPES[c]	89.17 – 178.35	89.17	178.35	104.03
***C.krusei* (1)**				
Amphotericin B	1.0	-	-	-
Nystatin	0.125	-	-	-
Fluconazole	16.0	-	-	-
PES[c]	136.71	-	-	-
SPES[c]	22.29	-	-	-

[a] the lowest concentration of drug that was able to inhibit 50% of isolates of each type.
[b] a lowest concentration of drug that was able to inhibit 90% of isolates of each species
[c] PES e SPES values in μg/mL of total phenol content in gallic acid
[d] MFC test presented the same value as the one found for the MIC