TITLE: Evaluation of anti-*Candida* activity of clinical strains of *Lactobacillus*: identification of strains with probiotic potential

AUTHORS: SANTOS, J.D.; ROSSONI, R.D.; BARROS, P.P.; ALVARENGA, J.A.; VELLOSO, M.S.; RIBEIRO, F.C.; JORGE, A.O.C.; JUNQUEIRA, J.C.

INSTITUTION: Institute of Science and Technology, UNESP - Univ. Estadual Paulista, São José dos Campos, SP, Brazil.

ABSTRACT:
Candida albicans is an opportunistic pathogen that can cause severe and recurrent infections in the mucosa, but also fatal systemic infections. Recently, probiotics bacteria have been studied as a potential method to prevent opportunistic infectious diseases due to their ability to inhibit the virulence of pathogens. In this context, the objective of this study was to isolate and identify *Lactobacillus* strains from caries-free subjects and to select the best strains with antifungal potential on *C. albicans* biofilm using the CFU count assay. After that, among different identified strains of *Lactobacillus*, we verified that *L. paracasei* 28.4, *L. rhamnosus* 5.2 and *L. fermentum* 20.4 strain had the greatest ability to affect the biofilm formation of *C. albicans*. *In vitro* analysis investigated the effects of these strains of *Lactobacillus* on *C. albicans* biofilm formation by crystal violet and scanning electron microscopy assays. Moreover, the expression of adhesion (ALS3 and HWP1) and transcriptional regulatory (EFG1 and CPH1) genes was determined by quantitative real-time PCR assay. In the violet crystal assay, biofilms associated with *Lactobacillus* or its supernatants obtained lower biomass compared to biofilms formed only by *C. albicans*. For SEM analysis, it was verified an intimate relationship between microorganisms and lower hyphal formation in *Lactobacillus*-associated biofilms compared to control biofilms. All of the *C. albicans* genes analyzed were significantly downregulated in association with *Lactobacillus* compared to the control group achieving 100-fold decrease for ALS3, 333-fold decrease for HWP1, 5-fold decrease for CPH1 and 6-fold decrease for EFG1. We concluded that most of clinical isolates of *Lactobacillus* have some degree of CFU reduction of *C. albicans*. *L. fermentum* 20.4, *L. paracasei* 28.4 and *L. rhamnosus* 5.2 have potential to be used as probiotics in the oral cavity because they affect the biofilm of *C. albicans* by downregulating expression of ALS3, HWP1, CPH1 and EFG1 genes.

KEYWORDS: *Candida albicans;* probiotic; *Lactobacillus*; gene expression.

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