Clinical Infectious Diseases

INVITED ARTICLE

CLINICAL PRACTICE: Ellie J. C. Goldstein, Section Editor

Probiotics as Antifungals in Mucosal Candidiasis

Victor H. Matsubara,[1,2,3] H. M. H. N. Bandara,[1] Marcia P. A. Mayer,[2] and Lakshman P. Samaranayake[1]

[1]School of Dentistry, University of Queensland, Brisbane, Australia; [2]Department of Microbiology, Institute of Biomedical Sciences, and [3]Department of Prosthodontics, School of Dentistry, University of São Paulo, Brazil

Candida is an opportunistic pathogen that causes mucosal and deep systemic candidiasis. The emergence of drug resistance and the side effects of currently available antifungals have restricted their use as long-term prophylactic agents for candidal infections. Given this scenario, probiotics have been suggested as a useful alternative for the management of candidiasis. We analyzed the available data on the efficacy of probiotics in candidal colonization of host surfaces. A number of well-controlled studies indicate that probiotics, particularly lactobacilli, suppress *Candida* growth and biofilm development in vitro. A few clinical trials have also shown the beneficial effects of probiotics in reducing oral, vaginal, and enteric colonization by *Candida*; alleviation of clinical signs and symptoms; and, in some cases, reducing the incidence of invasive fungal infection in critically ill patients. Probiotics may serve in the future as a worthy ally in the battle against chronic mucosal candidal infections.

Keywords. probiotics; *Candida*; candidiasis; *Lactobacillus*.

The high prevalence of human immunodeficiency virus (HIV) infection and other immunocompromising conditions globally has resulted in resurgence of *Candida* infections. These infections may be present on mucosal surfaces, including the oral cavity, oropharynx, esophagus, and vagina, as well as systemically [1].

Healthy individuals may also be the target of *Candida* infections, as this fungus is a commensal organism in human mucosal surfaces, inhabiting one-half of the human populace as an opportunist pathogen of the gastrointestinal and urogenital tracts [2]. When adverse conditions supervene, particularly in debilitated individuals, *Candida* is capable of causing superficial as well as deep invasive candidiasis, including fungemias. These diseases are essentially caused by candidal biofilms attached to body surfaces, as opposed to the planktonic form of the yeast, which exists in the suspended phase. *Candida albicans* is the most common *Candida* species inhabiting the mucosal surfaces both in health and disease, whereas other *Candida* species such as *C. tropicalis*, *C. guilliermondii*, *C. krusei*, and *C. glabrata* are less frequently isolated.

A range of adverse factors predisposes an individual to local or systemic candidal infection. The critical factors that precipitate systemic infections include the very low-birth-weight neonates [3] and immunosuppression as in HIV disease, or radiation and cytotoxic therapy [4]. Perturbation of mucosal ecosystem or marked changes in the microbial ecosystems due to antibiotics or corticosteroids, hypoendocrine states (eg, hypothyroidism, Addison disease, and diabetes mellitus), blood disorders such as acute leukemia, xerostomia due to irradiation or Sjogren syndrome, and ill-fitting appliances are predisposing factors for localized candidal infections either in healthy or diseased states [4]. Thus, *Candida* is considered to be an opportunistic pathogen, causing "diseases of the diseased."

The aim of this review was to explore critically the available in vitro and in vivo data on the efficacy of probiotic therapy in managing mucosal candidiasis. For this purpose, a critical review of the literature was conducted to select pertinent articles published in the English literature from 2000 to 2015. An electronic search was performed in Medline using the following terms: "probiotic or *Lactobacillus*" AND "*Candida* or candidiasis" to garner clinical evidence, and "probiotic or *Lactobacillus*" AND "*Candida*" for the in vitro studies. Only clinical trials assessing *Candida* infection in the oral cavity, urogenital tract, and gastrointestinal tract were included.

In the following sections, we provide an overview of *Candida* infections, a summary of probiotics, in vitro and in vivo evidence of the antifungal effects of probiotics and their possible mechanisms of action, and the safety and risks of probiotic therapy.

CANDIDA INFECTIONS

Oral Candidiasis

Oral candidiasis can manifest in a variety of clinical guises. The classic triad of oral candidiasis is the pseudomembranous, the erythematous (atrophic), and the hyperplastic variants [4].

In addition, there are a number of other *Candida*-associated lesions where the etiology is multifactorial. These diseases include *Candida*-associated denture stomatitis, angular cheilitis or angular stomatitis, median rhomboid glossitis, and the newly described linear gingival erythema, the microbial etiology of which is still poorly understood [4].

Received 25 November 2015; accepted 20 January 2016; published online 29 January 2016.
Correspondence: L. P. Samaranayake, University of Queensland Oral Health Centre, 288 Herston Road, Brisbane, QLD 4006, Australia (l.samaranayake@uq.edu.au).

Clinical Infectious Diseases® 2016;62(9):1143–53

DOI: 10.1093/cid/ciw038

Extraoral and Systemic *Candida* Infections

Vulvovaginal *Candida* infection (VVC) is the second most common cause of vaginitis after bacterial vaginosis. Transmission of this yeast from the vagina to the mouths of newborns during birth is a major portal of oral infections in newborns, leading to the development of thrush [2].

Candida inhabits the gastrointestinal (GI) tract in almost all humans, and most of the infections involving *Candida* are endogenously acquired from the GI tract. *Candida* can translocate into the bloodstream through the intact gastrointestinal mucosa and spread to visceral organs, leading to systemic candidiasis, especially in critically ill patients [3]. Disruption of normal physiological barriers, such as gastric acidity and perturbations of the indigenous microflora of the colon, facilitate *Candida* overgrowth.

Within the GI tract, the most common site of infection is the esophagus. *Candida* may be associated with gastric ulcers as an opportunistic pathogen that delays ulcer healing and aggravates the disease [5].

Management of Candidiasis

For some decades, systemic antifungal agents have been successfully used to prevent mucosal as well as invasive fungal infections. However, due to the drug side effects (nausea, vomiting, and diarrhea), and potential emergence of resistant strains, antifungal prophylaxis has not been totally successful.

The commonly used antifungals are the polyenes (nystatin and amphotericin B) and azoles (fluconazole, itraconazole, voriconazole). Interestingly, the biofilm phase of *Candida* is much more resistant to all these antifungals compared with their planktonic counterparts [6]. The limited spectrum and toxicity of available antifungals and the gradual emergence of resistance to these drugs are a concern; thus, alternative therapies are urgently warranted.

PROBIOTICS

The use of probiotic bacteria against microbial infections has emerged as an alternative therapeutic technique for *Candida* infections in view of the limitations of the currently available antimicrobials.

Probiotics are defined as live microorganisms that, when administered or consumed in adequate quantities, confer health benefits on the host. Bacteria belonging to the genera *Lactobacillus* and *Bifidobacterium* and, to a lesser extent, *Enterococcus*, *Streptococcus*, *and Saccharomyces* have often been used as probiotics in food supplements for a considerable period of time [7].

A safe probiotic needs to be of human origin, devoid of intrinsic and transmissible antibiotic resistance genes. The functional requirements of a probiotic include acid and bile tolerances, adequate adherence and colonization on epithelial surfaces, immunostimulation, and antagonistic activity against pathogens [7].

Therapeutic Potential of Probiotics

In therapeutic terms, probiotics are known to reduce *Candida* infections in different organ systems of the human body, and are generally considered to be beneficial for overall health. For instance, probiotics can combat diarrhea (mainly in children), and relieve lactose intolerance and symptoms of inflammatory bowel diseases [7]. Additionally, probiotic bacteria have been investigated for their potential for preventing cancers such as colorectal cancer [8], regulating blood pressure [9], and suppressing cholesterol levels [10]. The combination of probiotics with traditional treatment options are thought to generate better outcomes and disease resolution in different loci, with only a marginal increase in the treatment cost [7, 11, 12].

Probiotics as an Antimicrobial

Organisms of the genus *Lactobacillus* have been traditionally used as probiotics for decades, and they are deemed worthy as an alternative biological approach to combat bacterial and fungal pathogens in the oral cavity, GI tract, and urogenital system [1, 3, 11, 13–21]. It is noteworthy that the antimicrobial effect of probiotic bacteria is strain-specific; hence, the selection of probiotics for therapeutic purposes should be targeted for specific pathogens and their beneficial effects cannot be generalized [14]. In addition, there are reports of putative antiviral effects of probiotics, mainly against respiratory viral pathogens, in people of all ages [22].

IN VITRO EVIDENCE OF THE ANTIFUNGAL EFFECTS OF PROBIOTICS

A number of in vitro studies have demonstrated the antifungal effect of polymicrobial combinations of probiotics against human *C. albicans* isolates from the oral cavity, GI tract, and genitourinary tract [13, 14, 23–31]. Table 1 illustrates the variety and the extent of bacterial strains used to evaluate the candidicidal activity of probiotic bacteria, beginning this millennium.

The probiotic bacteria that have been investigated against *Candida* species to date include *Streptococcus salivarius* K12 [23], *Lactobacillus rhamnosus* GR-1, *Lactobacillus reuteri* RC-14 [25], and also clinical isolates of *Lactobacillus* [27, 30].

Antimicrobial activity of lactobacilli is generally well known. Studies using antagonism in agar diffusion assays have demonstrated that *Lactobacillus* species inhibit the growth of both gram-positive and gram-negative pathogens (eg, *Streptococcus mutans* and *Escherichia coli*, respectively) [13, 14], in addition to *Candida* species [13, 14, 25, 27, 30, 32]. *Candida albicans* was found to be more susceptible to the antifungal effect of *Lactobacillus* than *C. tropicalis* [27]. Moreover, probiotic bacteria and their supernatant also exhibited growth inhibitory activities against *C. glabrata* [32]. The production of hydrogen peroxide by the probiotics that antagonize candidal growth was a notable phenomenon observed in a number of these studies [27, 30].

Hyphae formation and adhesion assays were used to evaluate the effect of *Saccharomyces boulardii* [26] and *S. salivarius* [23]

Table 1. In Vitro Investigations on the Antifungal Effects of Probiotics

Reference	Probiotics	Pathogen	Method	Results	Comments
Strus et al, 2005 [27]	14 different strains: *Lactobacillus fermentum*, *Lactobacillus rhamnosus*, *Lactobacillus plantarum*, *Lactobacillus acidophilus*	*Candida albicans*, *Candida pseudotropicalis*	Antagonism on agar plates	- All probiotics inhibited the growth of *C. albicans* to a certain degree - Most lactobacilli were able at least slightly inhibit the growth of *C. pseudotropicalis*	Anticandidal activity related to H_2O_2 production and alternative mechanism.
Thein et al, 2006 [28]	*L. acidophilus*, *Actinomyces israelii*, *Prevotella nigrescens*, *Porphyromonas gingivalis*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Streptococcus mutans*, and *Streptococcus intermedius*	*C. albicans* 2560 g	Biofilm assay (scanning electron microscopy)	- 48 h co-culture: All bacteria, except *S. mutans* and *S. intermedius*, reduced viable yeasts cells - *C. albicans* biofilm + *P. aeruginosa*: reduced hyphal growth compared with bacteria-free biofilm	Bacteria modulate *C. albicans* biofilm formation in mixed species co-cultures and affected the morphogenesis of the yeast.
Hasslöf et al, 2010 [14]	*L. plantarum* 299v, *L. plantarum* 931, *L. rhamnosus* GG ATCC 53103, *L. rhamnosus* LB21, and *Lactobacillus paracasei*	MS: - Reference strains: *S. mutans* NCTC 10449, *S. mutans* Ingbritt, and *Streptococcus sobrinus* OMZ176 - Clinical isolates: *S. mutans* P1:27 and *S. mutans* P2:29 *C. albicans*: - Reference strains: *C. albicans* ATCC 28366, *C. albicans* ATCC 10231 - Clinical isolates: *C. albicans* 1957, *C. albicans* 3339 and *C. albicans* GDM8	Agar overlay interference tests. Four concentrations of probiotics were tested (10^9, 10^7, 10^5, and 10^3 CFU/mL)	MS: - 10^9 to 10^5 CFU/mL: all lactobacilli strains inhibited the growth of the MS strains completely (except *L. acidophilus* La5) - 10^3 CFU/mL: only *L. plantarum* 299v and *L. plantarum* 931 displayed a total growth inhibition for all MS. *L. rhamnosus* GG ATCC 53103 inhibited the growth slightly for 3 MS *C. albicans*: - 10^9 and 10^7 CFU/mL: all lactobacilli except *L. acidophilus* La5 and *Lactobacillus reuteri* PTA 5289 inhibited all *Candida* strains completely - 10^5 CFU/mL: *L. rhamnosus* strains, *L. paracasei*, and *L. reuteri* PTA 5289 displayed a slight inhibition. *L. acidophilus* La5 showed no inhibition. *L. plantarum* and *L. reuteri* ATCC 55730 executed a total inhibition - 10^3 CFU/mL: No inhibition was recorded except for the *L plantarum* strains	*L. acidophilus* La5: weaker inhibition capacity in comparison with the other probiotic strains ($P < .05$). All the tested *Lactobacillus* strains reduced *Candida* growth, but the effect was generally weaker than for MS.
Murzyn et al, 2010 [26]	*Saccharomyces boulardii*	*C. albicans* SC5314	- Hyphae formation assay - Adhesion assay - Gene expression assay	- Active compounds of probiotic yeast reduced *Candida* virulence factors (hyphae formation, cell adhesion, and biofilm formation) - Yeast extract and capric acid reduced expression of HWP1, INO1, and CSH1 genes in *C. albicans* cells	Capric acid was the main compound affecting hyphae formation, *Candida* adhesion, and biofilm formation.
Ishijima et al, 2012 [23]	*Streptococcus salivarius* K12	*C. albicans* (clinical isolate)	- Germ tube formation and mycelial growth of *C. albicans* (adherence to plastic substratum) - Deferred streak assay	- *S. salivarius* reduced adherence of mycelial form to plastic substratum, increased the number of planktonic *Candida* cells in culture medium, but did not inhibit *C. albicans* strain - Probiotic bacteria preferentially bound to hyphae	*S. salivarius* K12 was not directly fungicidal, but appeared to inhibit *Candida* adhesion to the substratum.

Table 1 continued.

Reference	Probiotics	Pathogen	Method	Results	Comments
Köhler et al, 2012 [25]	*L. rhamnosus* GR-1 and *L. reuteri* RC-14 *Lactobacillus johnsonii* PV016 and *Staphylococcus aureus* ATCC 25923 (controls)	*C. albicans* SC5314.	Antagonism on agar plates and in broth cultures	- 48 h: *Lactobacillus* GR-1 and RC-1 showed visible zones of fungal growth inhibition around them - *L. johnsonii*: very weak inhibition zone - *S. aureus*: no inhibition zones - *C. albicans* growth was suppressed at low pH by the *Lactobacillus* culture supernatants - Probiotics inhibited genes associated with biofilm formation	Lactic acid at low pH environment: major role in fungal growth inhibition. Glucose or other nutrient exhaustion was not a likely cause for fungal inhibition. H_2O_2 production may be an anti-*Candida* factor.
Coman et al, 2014 [13]	*L. rhamnosus* IMC 501 *L. paracasei* IMC 502 Combination of both (SYNBIO)	- Gram-positive, gram-negative bacteria - *C. albicans*, *Candida glabrata*, *Candida krusei*, *Candida parapsilosis*, and *Candida tropicalis*	- Modified cross-streak method - Radial streak method - Agar well diffusion method	*L. rhamnosus* - Inhibitory activity against both gram-positive and gram-negative bacteria, and against 2 *C. albicans* strains (ATCC 10261 and ISS7) *L. paracasei* - Inhibitory effect on gram-positive and gram-negative bacteria, especially *S. aureus* and *Proteus mirabilis*. All *Candida* spp were inhibited except *C. glabrata* and *C. tropicalis* SYNBIO- Inhibitory activity against most of the bacteria and fungi strains, especially *C. albicans* and *C. krusei*	*L. paracasei* IMC 502: higher activity toward all the pathogens, especially *Candida* strains; strong inhibition registered for SYNBIO.
Verdenelli et al, 2014 [30]	*L. paracasei* subsp *paracasei*, *L. plantarum*, *L. fermentum*, *L. rhamnosus* IMC 501, and *L. paracasei* IMC 502	*C. albicans*, *C. glabrata*, *C. krusei*, *C. parapsilosis*, *C. tropicalis* (clinical isolates)	- Agar well diffusion assay and radial method - Coaggregation assay	- Well diffusion assay: no inhibition against *Candida* spp - Radial method: All lactobacilli had the capacity to inhibit *Candida* in different degrees	Inhibition and coaggregation ability vary according to the *Lactobacillus* strain and the pathogen involved
Kheradmand et al, 2014 [24]	*L. plantarum* (ATCC 8014) and *L. johnsonii* (clinical isolate) enriched or not with SeNPs	*C. albicans* (ATCC 14053)	- Conventional hole-plate diffusion method and time-kill assay using probiotic supernatant (grown with or without selenium dioxide) - Time-kill assay using probiotic cell suspension (grown with or without selenium dioxide)	Conventional hole-plate diffusion: - *L. plantarum* and *L. johnsonii* supernatant grown with selenium dioxide showed potent anti-*Candida* activity. - No antifungal effect was observed with supernatant without selenium Time-kill assay: - No viable *C. albicans* was present after 4 h incubation with culture supernatants grown with selenium dioxide - Viable *C. albicans* cells were present even after 24 h incubation with culture supernatants grown without selenium dioxide After 0.5 h, *Lactobacillus* strains without SeNPs decreased the viability of *C. albicans* by approximately 10-fold. SeNP-enriched species decreased 1000-fold	Direct antifungal effect was observed when selenium-enriched *Lactobacillus* spp were co-cultured with *C. albicans*. The strong inhibition of *C. albicans* by supernatant of selenium-enriched *Lactobacillus* spp indicated the release of potent exometabolites.

Table 1 continued.

Reference	Probiotics	Pathogen	Method	Results	Comments
Ujaoney et al, 2014 [29]	*L. acidophilus*, *L. rhamnosus*, *L. salivarius*, *Bifidobacterium bifidum*, *Streptococcus thermophilus*, *Bifidobacterium infantis*, *Lactobacillus* GG, and *Bacillus coagulans* BC30	*C. albicans* 10341	Biofilm assay on denture strips using bacterial suspensions and probiotic supernatants (XTT quantification)	Probiotics' supernatant provided a stronger and significant inhibitory effect on biofilm formation than their bacterial counterparts	Depletion of nutrients in the culture media by overgrowth of the probiotic bacteria may inhibit fungal growth.
Vilela et al, 2015 [31]	*L. acidophilus* ATCC 4356	*C. albicans* ATCC 18804	Biofilm assay and *C. albicans* filamentation assay using light microscope	- *L. acidophilus* culture filtrate reduced the growth of *C. albicans* cells by 45.1% - Less hyphae formation in the presence of *L. acidophilus* cells or culture filtrate	*L. acidophilus* produced substances with anti-*Candida* activity, presenting an indirect effect on *Candida*.
Chew et al, 2015 [32]	*L. rhamnosus* GR-1 and *L. reuteri* RC-14	*C. glabrata* ATCC 2001 and clinical isolates	- Spot overlay assay - Plate-based microtiter assay - *Candida* viability assay using confocal laser scanning microscopy - Aggregation assay - MATH assay	- Probiotic strains exhibit growth inhibitory activities (bacterial cells and supernatant) and candidicidal activity against *C. glabrata* - Both probiotic strains exhibited strong autoaggregation and coaggregation in the presence of *Candida*	Lactobacilli may prevent *C. glabrata* colonization through the formation of aggregates. Reduction of pH plays role on the antifungal effect of probiotic, but not H_2O_2. Other inhibitory mechanisms or pathways may be involved.

Abbreviations: CFU, colony-forming units; H_2O_2, hydrogen peroxide; MATH, microbial adhesion to hydrocarbons; MS, mutans streptococci; SeNPs, selenium dioxide nanoparticles; XTT, tetrazolium salt.

on *C. albicans*. *Saccharomyces boulardii* appears to secrete an active compound that inhibits filamentation of *C. albicans* and its mycelial development, a crucial virulence attribute of this fungal pathogen. *Streptococcus salivarius* K12 was not directly fungicidal, but appeared to inhibit *Candida* adhesion to the substratum and increase the planktonic cells in culture medium [23].

The effect of probiotics may be time dependent. Using a time-kill assay, some investigators have attempted to reinforce the probiotic effect of bacteria by supplementing the medium with chemical adjuvants, such as selenium. The latter is an essential micromineral that regulates metabolism and is known to reinforce immunity. Selenium nanoparticle-enriched *Lactobacillus plantarum* and *Lactobacillus johnsonii* cells and supernatant have shown higher antifungal activity against *C. albicans* than controls devoid of the nanoparticles [24]. These data, yet to be confirmed, exemplify how probiotics could be synergized and deserve further study.

Experiments on the effect of probiotics on *Candida* biofilms, as opposed to their suspended planktonic phase, provide another fascinating glimpse of how probiotics behave [28, 29, 31]. It has been shown that a number of bacteria can interfere with the biofilm growth by reducing hyphal development [28, 31], a result akin to that described above [26]. Ujaoney et al [29] reported that the probiotic cell-free supernatant had a strong and significant inhibitory effect on biofilm development on denture acrylic strips than the bacteria per se, indicating that the inhibitory agent is an exometabolite secreted into the medium.

Chew et al [32], using confocal laser scanning microscopy, also demonstrated the candidicidal effect of planktonic lactobacilli and their supernatant against *C. glabrata*, another common fungal pathogen.

As summarized in Table 1, there is now a convincing body of in vitro data to indicate the antifungal effect of probiotics against *Candida* species. The challenge now is to clarify the mechanisms involved and harness these in further translational work. Investigations of the molecular mechanisms underlying the probiotic effect using gene expression and related technology are likely to yield interesting data in this regard.

IN VIVO EVIDENCE OF THE ANTIFUNGAL EFFECTS OF PROBIOTICS

As opposed to the in vitro studies reported above, a number of in vivo studies have also been performed over the past decade or so to substantiate the antifungal activity of probiotics in humans (Table 2). The oral cavity, GI tract, and urogenital tract have been the major loci of investigation, as these sites are susceptible to *Candida* infections.

Oral Cavity

Despite the high prevalence of oral candidal infections in predisposed populations the world over, and the recalcitrance and

Table 2. Clinical Investigations on the Antifungal Effects of Probiotics in the Oral Cavity, Urogenital Tract, and Gastrointestinal Tract of Humans

Reference	Site of Action	Probiotic	Pathogen	Method	Results	Comments
Hatakka et al, 2007 [15]	Oral cavity	*Lactobacillus rhamnosus* GG (ATCC 53103), *L. rhamnosus* LC705, *Propionibacterium freudenreichii* subsp *shermanii* JS	*Candida albicans*, *Candida glabrata*, *Candida krusei*, and *Candida tropicalis*	- RCT: 276 elderly people - Probiotic therapy: daily consumption of 50 g of probiotic cheese or control cheese for 16 wk - Community periodontal index and mucosal lesions were recorded. Sampling for oral yeasts was undertaken	- Prevalence of yeast in saliva decreased in the probiotic group from 30% to 21% (32% reduction), and increased in the control group from 28% to 34% - Probiotic intervention reduced the risk of high yeast counts by 75%	Probiotic reduced the prevalence of hyposalivation; No adverse events were observed.
Mendonça et al, 2012 [39]	Oral cavity	*Lactobacillus casei* and *Bifidobacterium breve*	*C. albicans*, *C. tropicalis*, *Candida guilliermondii*, *C. glabrata*, *Candida lipolytica*, *C. krusei*, *Candida kefyr*, and *Candida parapsilosis*	- 42 women (aged ≥65 y) - Probiotic therapy: 3 times weekly for 30 d - Saliva sample collection for *Candida* cell quantification (CFU counting) and IgA analysis (ELISA)	- Reduction of *Candida* prevalence from 92.9% to 85.7% - Increase of anti-*Candida* IgA levels	*C. albicans* was the most frequently species isolated before and after probiotic consumption.
Sutula et al, 2012 [33]	Oral cavity	*L. casei* Shirota	*Candida* spp *Streptococcus mutans* and gram-negative anaerobic species	- 7 healthy complete denture wearers (aged ≥55 y) - Probiotic therapy: once daily, with the denture in position, for 28 d - Samples of saliva, tongue, and denture biofilm were collected	- No effect of probiotic on occurrence and viability of *Candida* - No significant change in the viability of *Streptococcus mutans* and gram-negative anaerobes	Small sample group (n = 7) completed the study protocol.
Sutula et al, 2013 [34]	Oral cavity	*L. casei* Shirota	- *Candida* spp - Gram-negative anaerobic species	- 21 healthy dentate subjects (aged 18–45 y) - Probiotic therapy: once daily for 28 d - Saliva and tongue-coating samples were collected; Morning breath samples were obtained using portable sulphide monitors	- *Lactobacillus* level in saliva was increased during probiotic consumption period - *Candida* and anaerobic species levels were unaffected by the therapy - Morning breath scores measured were not significantly affected	Confirmation of the temporary and intake-dependent presence of *Lactobacillus*.
Li et al, 2014 [11]	Oral cavity	*Lactobacillus bulgaricus*, *Bifidobacterium longum*, and *Streptococcus thermophilus*	*Candida* spp	- RCT: 65 patients with *Candida*-associated stomatitis - Probiotic therapy: antifungal alone (sodium bicarbonate solution + nystatin paste) or associated with probiotic, 3 times daily for 4 wk - Parameters of hyperemia, visual analogue scale scores, culture of saliva, and lingual dorsum swab were assessed	Detection rate of *Candida* spp was reduced in the probiotic group; Significant relief of clinical signs and symptoms after probiotic administration	No adverse events were observed.
Ishikawa et al, 2015 [16]	Oral cavity	*L. rhamnosus* HS111, *Lactobacillus acidophillus* HS101, and *Bifidobacterium bifidum*	*Candida* spp	- RCT: 55 denture wearers harboring *Candida* spp with no clinical symptoms of oral candidiasis - Probiotic therapy: once daily for 5 wk (probiotic or placebo) - Palatal swab sample for *Candida* cell quantification and identification	- Significant reduction of *Candida* infection after probiotic administration - *C. albicans* was the most prevalent species before and after the probiotic therapy	Reduction of *Candida* infection was independent of initial *Candida* level, colonizing species, or age of denture.

Table 2 continued.

Reference	Site of Action	Probiotic	Pathogen	Method	Results	Comments
Kraft-Bodi et al, 2015 [17]	Oral cavity	*Lactobacillus reuteri* DSM 17938 and *L. reuteri* ATCC PTA 5289	*Candida* spp	- RCT: 215 elderly people (aged 60–102 y) - Probiotic therapy: twice daily for 12 wk (placebo or probiotic) - Prevalence and amount of *Candida* growth (saliva and plaque samples), oral hygiene, and gingival inflammation were assessed	- Significant reduction of *Candida* cells in saliva and plaque after probiotic administration - No differences in the levels of supragingival plaque or bleeding on probing were observed	"Strong taste" of the tablets and gastric upset were compliances reported in both control and experimental groups.
Pirotta et al, 2004 [38]	Urogenital tract	*L. rhamnosus* and *B. longum* (oral powder); *L. rhamnosus*, *Lactobacillus delbrueckii*, *L. acidophilus*, and *Streptococcus thermophiles* (vaginal pessary)	*Candida* spp	- RCT: 235 nonpregnant women (aged 18–50 y) with a short course of oral antibiotic administration - Probiotic therapy: powder administration twice daily and pessary once daily, for 6 d of antibiotic course and 4 d after - Vaginal swab was collected for *Candida* identification - Identification of symptomatic VVC	The use of oral or vaginal forms of probiotic bacteria could not prevent postantibiotic vulvovaginitis	10 d of probiotic therapy may be insufficient time for the occurrence of beneficial effects against *Candida* spp in the vagina.
Martinez et al, 2009 [12]	Urogenital tract	*L. rhamnosus* GR-1 and *L. reuteri* RC-14	*Candida* spp	- RCT: 55 women diagnosed with VVC - Probiotic therapy: single dose of fluconazole plus probiotic or placebo once daily for 4 wk - Vaginal swab was collected for *Candida* identification Clinical evaluation to detect signs and symptoms of VVC	- Probiotic significant reduced vaginal discharge, itching and/or burning vaginal feeling, dyspareunia, and/or dysuria - Probiotic reduced the presence of *Candida* spp	Mild adverse effects were reported, but could not be definitely associated with probiotic administration.
Vicariotto et al, 2012 [2]	Urogenital tract	*Lactobacillus fermentum* LF10 and *L. acidophilus* LA02 (arabinogalactan and fructooligosaccharides as prebiotics)	*Candida* spp	- Thirty female patients (aged 23–64 y) - Probiotic therapy: once daily for 7 d, then once every 3 d for additional 3 wk. In the following month, once weekly - Vaginal swabs were collected for yeast identification	- Probiotic significantly solved *Candida* yeast symptoms in 86.6% of patients after 28 d - At the end of the second month, recurrences were recorded in 11.5% of patients	Probiotic may establish and maintain a protective barrier effect against vaginal *Candida*.
Hu et al, 2013 [1]	Urogenital tract and oral cavity	*Bifidobacterium* and *Lactobacillus* (DanActive or YoPlus yogurt)	*Candida* spp	- 24 women (17 HIV infected, 7 HIV uninfected); - Probiotic therapy: 15 d consuming each yogurt. 30-day washout period between the 2 yogurt consumption periods - Oral and vaginal culture swabs were collected	- Less fungal colonization among women was observed after probiotic consumption - HIV-infected women had significantly lower vaginal fungal colonization after DanActive yogurt consumption	Reduced oral fungal colonization was observed in HIV-infected women consuming probiotic yogurts, but not statistically significant.
Kovachev et al, 2015 [35]	Urogenital tract	*L. acidophilus*, *L. rhamnosus*, *L. delbrueckii* subsp *bulgaricus*, and *S. thermophiles*	*C. albicans*	- 436 women (aged 17–50 y) with *C. albicans* vaginal infection - Probiotic therapy: antifungals alone (fluconazole and fenticonazole) or associated with vaginal probiotic agent - Clinical and microbiological tests were performed	- Probiotic reduced clinical complaints - Probiotic therapy improved the investigated parameters: vaginal fluorine, vaginal tissue changes, and pH	Local application of probiotics may improve the efficacy of conventional antifungals and prevent relapse.
Manzoni et al, 2006 [19]	Gastrointestinal tract	*L. casei* subsp *rhamnosus*	*Candida* spp	- RCT: 80 preterm neonates with a very low birth weight - Probiotic therapy: human milk alone or added with probiotic, for up to 6 wk - Samples from oropharyngeal, stool, gastric aspirate, and rectal specimens were collected to assess fungal colonization in the GI tract	- Human milk supplemented with probiotic reduced significantly the incidence of *Candida* enteric colonization - Probiotic reduced significantly the numbers of fungal isolates	Probiotic reduced incidence and intensity of enteric colonization by *Candida* spp; no adverse events were observed.

Table 2 continued.

Reference	Site of Action	Probiotic	Pathogen	Method	Results	Comments
Romeo et al, 2011 [21]	Gastrointestinal tract	*L. reuteri* (ATCC 55730) and *L. rhamnosus* (ATCC 53103)	*Candida* spp	- 249 preterms neonates with a birth weight <2500 g and a gestational age <37 wk - Probiotic therapy: breast milk or formula milk alone, or supplemented with 1 probiotic, for up to 6 wk - Clinical evaluations were performed. Stool samples, oropharyngeal, and gastric aspirate specimens were collected for *Candida* detection. Blood cultures and Platelia *Candida* test were conducted for the diagnosis of invasive candidiasis	- Probiotic reduced significantly *Candida* stool colonization - *L. reuteri* group had a significant higher reduction in gastrointestinal symptoms than the *L. rhamnosus* and control groups - Probiotics reduced the incidence of abnormal neurological outcome	Probiotics may prevent gastrointestinal colonization by *Candida*, protect from late-onset sepsis, and reduce abnormal neurological outcomes in preterms.
Demirel et al, 2013 [3]	Gastrointestinal tract.	*Saccharomyces boulardii*	*Candida* spp	- 181 preterm neonates with a gestational age ≤32 wk and birth weight ≤1500 g - Probiotic therapy: nystatin suspension every 8 h, or breast milk or formula supplemented with probiotic once daily - Samples of blood, urine, and cerebrospinal fluid were collected to identify invasive fungal infection Skin, stool, or rectal cultures were obtained for *Candida* detection	- *Candida* colonization of the skin and stool were similar between probiotic and nystatin groups - Clinical sepsis and number of sepsis attacks were significantly lower in the probiotic group	Prophylactic *S. boulardii* and nystatin were equally effective in reducing candidal colonization and invasive fungal infection.
Kumar et al, 2013 [18]	Gastrointestinal tract	*L. acidophillus*, *L. rhamnosus*, *B. longum*, *B. bifidum*, *S. boulardii*, and *Saccharomyces thermophilus*	*Candida* spp	- RCT: 150 children (aged 3 mo–12 y) on broad-spectrum antibiotics for at least 48 h - Probiotic therapy: probiotic or placebo twice daily for 7 d - Rectal swab, samples of urine and blood were collected for *Candida* detection	- Probiotic therapy avoided a significant increase in the number of patients colonized by *Candida* spp - Probiotic significantly reduced the presence of *Candida* in the urine, but not in the blood	Probiotics may be an alternative strategy to reduce *Candida* infection in GI tract and urine in children receiving broad-spectrum antibiotics.
Roy et al, 2014 [20]	Gastrointestinal tract	*L. acidophilus*, *Bifidobacterium lactis*, *B. longum*, and *B. bifidum*	*Candida* spp	- RCT: 112 preterm neonates (gestational age <37 wk and birth weight <2500 g) - Probiotic therapy: breast milk supplemented with probiotic or placebo, twice daily for up to 6 wk - Clinical evaluations were performed. Stool samples and gastric aspirate specimens were collected for *Candida* detection. Blood cultures and Platelia *Candida* test were conducted for the diagnosis of invasive candidiasis	- Probiotic therapy reduced the duration of hospitalization and stool fungal colonization - Fungal infection was significant less in the probiotic group - Full feed establishment was earlier in probiotic group.	Probiotics may reduce enteral fungal colonization and reduce invasive fungal sepsis in low–birth-weight neonates.

Abbreviations: CFU, colony-forming units; ELISA, enzyme-linked immunosorbent assay; GI, gastrointestinal; HIV, human immunodeficiency virus; IgA, immunoglobulin A; RCT, randomized controlled trial; VVC, vulvovaginal candidiasis.

chronicity of these diseases, there are only a few in vivo studies evaluating the effect of probiotics on suppressing oral candidiasis. These indicate that probiotics may be a useful adjunct in the battle against oral candidiasis, especially as a prophylactic agent in immunocompetent individuals.

The elderly are a group particularly susceptible to oral candidiasis even in health, due to the prosthesis (dentures) they frequently wear and hyposalivation. Their weakened immune status may favor the recurrence of candidiasis. Two research groups have shown that the daily consumption of lactobacilli-laced cheese [15] or lozenges [17] significantly reduces the high yeast counts in saliva and biofilms in the elderly. Because biofilms on oral prosthetic devices act as potent reservoirs of the yeast, the mechanical removal of biofilms associated with the regular use of probiotics that reduce the oral burden of *Candida* could play a major role in preventing oral candidiasis in denture wearers. Interestingly, one study reported increased salivary flow as a salutary accompaniment to probiotic administration [15].

As mentioned, full denture wearers suffer frequently from *Candida*-associated denture stomatitis [4], which lowers the quality of life. Ishikawa et al [16] have reported that a probiotic product, when regularly placed on the palatal surface of maxillary dentures, reduced oral candidal burden in healthy denture wearers. These preliminary data imply that multispecies probiotics, together with good denture hygiene, may help suppress recurrence of these chronic infections [11].

Commercial food products with probiotics are common worldwide. A widely available probiotic-laced drink containing *Lactobacillus casei* and *Bifidobacterium breve* was able to reduce the prevalence of oral *Candida* in healthy individuals [39]. A significant increase in anti-*Candida* immunoglobulin A levels was associated with probiotic consumption [39]. In contrast, the identical product did not significantly affect the oral candidal colonization in complete denture wearers [33] and in healthy dentate people [34], after 4 weeks of administration. The lower dose of probiotic intake and the small number of individuals included in the latter studies may explain these divergent observations.

Urogenital Tract

Chronic VVC is a widely prevalent disease and impacts the life quality of thousands of women the world over. Although standard antifungals are effective, there is no alternative approach for suppressing these recalcitrant infections. Several groups have therefore evaluated the efficacy of probiotics in the treatment and prophylaxis of VVC [1, 2, 12, 35].

Two studies conducted on healthy women have reported that the coadministration of probiotics with standard antifungal therapy (fluconazole) was more effective in reducing symptoms of VVC, including vaginal discharge, pruritus vulvae, vulvar and vaginal erythema, dyspareunia, and dysuria compared with a group treated with antifungals alone [12, 35]. Clinical improvement was also observed after local administration of a commercial slow-release probiotic product alone, without an antifungal agent, in healthy women with recurrent VVC [2]. Similarly, in a study conducted in immunocompromised women, who are highly susceptible to recurrent and complicated VVC infection, probiotic yogurt consumption led to a decreased frequency of infection [1].

In contrast, another well-controlled study reported that probiotic bacteria taken both orally and locally were unable to prevent postantibiotic VVC in immunocompetent individuals who took oral antibiotics [38]. Qualitative and quantitative differences in the probiotic strains, as well as the period of probiotic administration, are likely to be the reasons for the divergent results between the foregoing studies.

Gastrointestinal Tract

Candida species are common inhabitants of the GI tract of humans. Perturbation of the local microbiome, however, leads to dysbiosis within this ecosystem, leading to candidal overgrowth and possible invasive infections, especially in infants [21].

Hence, immunocompromised children, especially preterm neonates with low birth weight, have been the target population of a number of studies evaluating the efficacy of probiotics against candidal colonization of the GI tract [3, 19–21]. Within this population, most researchers have reported a significant reduction in the incidence and intensity of enteric candidal colonization with probiotic-laced human milk, administered either with or without concurrent antifungals [19–21]. Important secondary effects of the probiotics observed in these studies include reduction of sepsis episodes [3], early establishment of full feeding associated with reduction in the duration of hospitalization [20], and the decrease in the incidence of abnormal neurological outcomes associated with late-onset sepsis [21].

Broad-spectrum antibiotics are notorious for their ability to cause GI tract dysbiosis and candidiasis [18, 38]. In immunocompetent children who had received broad-spectrum antibiotics, probiotic therapy led to a reduction of gastrointestinal candidal colonization as well as candiduria—a surrogate marker of invasive fungal infection [18].

POSSIBLE MECHANISMS OF ACTION OF PROBIOTICS

Clearly, the major attribute of probiotics appears to be the restoration of a natural healthy microbiome in a given habitat, turning it from a catastrophic, disease-inducing, dysbiotic microbiota to a healthy, symbiotic, stable equilibrium. A number of hypotheses, most unproven as yet, has been proposed for the genesis of this well-balanced state from disease to health. Probiotics may compete for nutrients and receptors on the cell surfaces with the pathogenic microorganisms, thus preventing their adhesion and colonization on the mucosal surfaces [2, 29]. Co- and auto-aggregation of probiotics with the formation of a critical

mass required for a healthy biofilm development may act as a protective lining against pathogenic infection [30]. Apart from the above, the production of biosurfactants that interfere with microbial adhesion and desorption [37], the release of exometabolites such as lactic, acetic, and capric acid, and the production of bacteriocins and hydrogen peroxide (H_2O_2) are other possible attributes postulated as mechanisms for probiotic activity [24–26]. Despite such in vitro data on the inhibitory effect of probiotic products on yeasts, the direct effect of probiotics on mucosal candidiasis is yet be shown in a laboratory environment mimicking the oral cavity, vagina, or GI tract.

The host response to probiotics is likely to play an important role in probiotic-mediated microbiome effects. The modulation of both innate and adaptive immune systems is probably associated with alteration of the cytokine profile and *Candida* recognition by epithelial and immune defense cells [28, 36, 40]. Evidence to imply probiotic interference with these host defense factors during candidal infestation is still needed.

With respect to candidal infection, probiotics were found to reduce filamentation and biofilm development in *C. albicans*, 2 key virulence attributes of this fungus [25, 28]. As the yeast form of *Candida*, as opposed to the hyphal form, is more susceptible to phagocytosis [40], probiotics appear to assist the host combat the pathogen more effectively by suppressing filamentation. Despite the evidence that probiotic bacteria may affect the expression of genes associated with biofilm formation and filamentation of *Candida* species [25], the mechanisms by which probiotics affect these yeasts attributes are still unclear.

Administration of probiotics in tandem with antifungal drugs synergizes clearance of *Candida* [11, 12, 35]. Apart from the obvious antifungal effect of the drug, the role of the probiotic under these conditions remains to be elucidated. The increased expression of stress-related genes and decreased expression of genes involved in drug resistance in *Candida*, promoted by the probiotics, would possibly increase the fungus's susceptibility to the antifungal agent administered [25].

SAFETY AND RISKS OF PROBIOTIC THERAPY

A range of bacteria has been utilized as probiotics in humans, depending on the pathological condition. None of the clinical studies mentioned above have reported adverse effects directly related to probiotics, suggesting their safety. Nevertheless, the safety, efficacy, and functionality of probiotic bacteria should be tested in healthy as well as in compromised individuals prior to their administration as therapeutic agents.

FUTURE PERSPECTIVES AND CONCLUSIONS

Clinical studies indicate that probiotics may reduce *Candida* colonization on human mucosal surfaces, relieve signs and symptoms of fungal infection, and enhance the antifungal effect of conventional therapy, implying that probiotics have the potential to sustain a healthy mucosal microbiota by acting both as prophylactic and adjunctive therapy against candidiasis. In vitro studies indicate that the antifungal effect of probiotics is likely to be due to their interference with *Candida* biofilm development and hyphal differentiation. However, it is premature to designate probiotics as an alternative to antifungals as yet, due to the paucity of available clinical trials. In particular, case-control clinical trials with adequate patient numbers are warranted, not only to ascertain the activity of the probiotic formulations, but also to evaluate their dosage, administration schedules, side effects, and biodynamics in humans. As with any formulations of live organisms, other concerns that need further investigation include the potential for selection of resistant strains, mutability, and tolerability on prolonged use, as well as pathogenic potential in immunocompromised patients. Given these caveats, probiotics may serve in the future as worthy allies in the battle against chronic mucosal fungal infections.

Note

Potential conflicts of interest. All authors: No reported conflicts. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Hu H, Merenstein DJ, Wang C, et al. Impact of eating probiotic yogurt on colonization by *Candida* species of the oral and vaginal mucosa in HIV-infected and HIV-uninfected women. Mycopathologia **2013**; 176:175–81.
2. Vicariotto F, Del Piano M, Mogna L, Mogna G. Effectiveness of the association of 2 probiotic strains formulated in a slow release vaginal product, in women affected by vulvovaginal candidiasis: a pilot study. J Clin Gastroenterol **2012**; 46(suppl): S73–80.
3. Demirel G, Celik IH, Erdeve O, Saygan S, Dilmen U, Canpolat FE. Prophylactic *Saccharomyces boulardii* versus nystatin for the prevention of fungal colonization and invasive fungal infection in premature infants. Eur J Pediatr **2013**; 172:1321–6.
4. Samaranayake LP. Superficial oral fungal infections. Curr Opin Dent **1991**; 1:415–22.
5. Brzozowski T, Zwolinska-Wcislo M, Konturek PC, et al. Influence of gastric colonization with *Candida albicans* on ulcer healing in rats: effect of ranitidine, aspirin and probiotic therapy. Scand J Gastroenterol **2005**; 40:286–96.
6. Sardi JC, Scorzoni L, Bernardi T, Fusco-Almeida AM, Mendes Giannini MJ. *Candida* species: current epidemiology, pathogenicity, biofilm formation, natural antifungal products and new therapeutic options. J Med Microbiol **2013**; 62(pt 1):10–24.
7. Saarela M, Mogensen G, Fonden R, Matto J, Mattila-Sandholm T. Probiotic bacteria: safety, functional and technological properties. J Biotechnol **2000**; 84:197–215.
8. Shida K, Nomoto K. Probiotics as efficient immunopotentiators: translational role in cancer prevention. Indian J Med Res **2013**; 138:808–14.
9. Khalesi S, Sun J, Buys N, Jayasinghe R. Effect of probiotics on blood pressure: a systematic review and meta-analysis of randomized, controlled trials. Hypertension **2014**; 64:897–903.
10. Jones ML, Tomaro-Duchesneau C, Martoni CJ, Prakash S. Cholesterol lowering with bile salt hydrolase-active probiotic bacteria, mechanism of action, clinical evidence, and future direction for heart health applications. Expert Opin Biol Ther **2013**; 13:631–42.
11. Li D, Li Q, Liu C, et al. Efficacy and safety of probiotics in the treatment of *Candida*-associated stomatitis. Mycoses **2014**; 57:141–6.
12. Martinez RC, Franceschini SA, Patta MC, et al. Improved treatment of vulvovaginal candidiasis with fluconazole plus probiotic *Lactobacillus rhamnosus* GR-1 and *Lactobacillus reuteri* RC-14. Lett Appl Microbiol **2009**; 48:269–74.
13. Coman MM, Verdenelli MC, Cecchini C, et al. In vitro evaluation of antimicrobial activity of *Lactobacillus rhamnosus* IMC 501®, *Lactobacillus paracasei* IMC 502® and SYNBIO® against pathogens. J Appl Microbiol **2014**; 117:518–27.
14. Hasslöf P, Hedberg M, Twetman S, Stecksen-Blicks C. Growth inhibition of oral mutans streptococci and candida by commercial probiotic lactobacilli—an in vitro study. BMC Oral Health **2010**; 10:18.

15. Hatakka K, Ahola AJ, Yli-Knuuttila H, et al. Probiotics reduce the prevalence of oral *Candida* in the elderly—a randomized controlled trial. J Dent Res **2007**; 86:125–30.
16. Ishikawa KH, Mayer MP, Miyazima TY, et al. A multispecies probiotic reduces oral *Candida* colonization in denture wearers. J Prosthodont **2014**; 24:194–9.
17. Kraft-Bodi E, Jorgensen MR, Keller MK, Kragelund C, Twetman S. Effect of probiotic bacteria on oral *Candida* in frail elderly. J Dent Res **2015**; 94:181S–6.
18. Kumar S, Bansal A, Chakrabarti A, Singhi S. Evaluation of efficacy of probiotics in prevention of *Candida* colonization in a PICU-a randomized controlled trial. Crit Care Med **2013**; 41:565–72.
19. Manzoni P, Mostert M, Leonessa ML, et al. Oral supplementation with *Lactobacillus casei* subspecies *rhamnosus* prevents enteric colonization by *Candida* species in preterm neonates: a randomized study. Clin Infect Dis **2006**; 42:1735–42.
20. Roy A, Chaudhuri J, Sarkar D, Ghosh P, Chakraborty S. Role of enteric supplementation of probiotics on late-onset sepsis by *Candida* species in preterm low birth weight neonates: a randomized, double blind, placebo-controlled trial. N Am J Med Sci **2014**; 6:50–7.
21. Romeo MG, Romeo DM, Trovato L, et al. Role of probiotics in the prevention of the enteric colonization by *Candida* in preterm newborns: incidence of late-onset sepsis and neurological outcome. J Perinatol **2011**; 31:63–9.
22. Lehtoranta L, Pitkaranta A, Korpela R. Probiotics in respiratory virus infections. Eur J Clin Microbiol Infect Dis **2014**; 33:1289–302.
23. Ishijima SA, Hayama K, Burton JP, et al. Effect of *Streptococcus salivarius* K12 on the in vitro growth of *Candida albicans* and its protective effect in an oral candidiasis model. Appl Environ Microbiol **2012**; 78:2190–9.
24. Kheradmand E, Rafii F, Yazdi MH, Sepahi AA, Shahverdi AR, Oveisi MR. The antimicrobial effects of selenium nanoparticle-enriched probiotics and their fermented broth against *Candida albicans*. Daru **2014**; 22:48.
25. Köhler GA, Assefa S, Reid G. Probiotic interference of *Lactobacillus rhamnosus* GR-1 and *Lactobacillus reuteri* RC-14 with the opportunistic fungal pathogen *Candida albicans*. Infect Dis Obstet Gynecol **2012**; 2012:636474.
26. Murzyn A, Krasowska A, Stefanowicz P, Dziadkowiec D, Lukaszewicz M. Capric acid secreted by *S. boulardii* inhibits *C. albicans* filamentous growth, adhesion and biofilm formation. PLoS One **2010**; 5:e12050.
27. Strus M, Kucharska A, Kukla G, Brzychczy-Wloch M, Maresz K, Heczko PB. The in vitro activity of vaginal *Lactobacillus* with probiotic properties against *Candida*. Infect Dis Obstet Gynecol **2005**; 13:69–75.
28. Thein ZM, Samaranayake YH, Samaranayake LP. Effect of oral bacteria on growth and survival of *Candida albicans* biofilms. Arch Oral Biol **2006**; 51:672–80.
29. Ujaoney S, Chandra J, Faddoul F, et al. In vitro effect of over-the-counter probiotics on the ability of *Candida albicans* to form biofilm on denture strips. J Dent Hyg **2014**; 88:183–9.
30. Verdenelli MC, Coman MM, Cecchini C, Silvi S, Orpianesi C, Cresci A. Evaluation of antipathogenic activity and adherence properties of human *Lactobacillus* strains for vaginal formulations. J Appl Microbiol **2014**; 116:1297–307.
31. Vilela SF, Barbosa JO, Rossoni RD, et al. Lactobacillus acidophilus ATCC 4356 inhibits biofilm formation by *C. albicans* and attenuates the experimental candidiasis in *Galleria mellonella*. Virulence **2015**; 6:29–39.
32. Chew SY, Cheah YK, Seow HF, Sandai D, Than LT. Probiotic *Lactobacillus rhamnosus* GR-1 and *Lactobacillus reuteri* RC-14 exhibit strong antifungal effects against vulvovaginal candidiasis-causing *Candida glabrata* isolates. J Appl Microbiol **2015**; 118:1180–90.
33. Sutula J, Coulthwaite L, Thomas L, Verran J. The effect of a commercial probiotic drink on oral microbiota in healthy complete denture wearers. Microb Ecol Health Dis **2012**; 23.
34. Sutula J, Coulthwaite LA, Thomas LV, Verran J. The effect of a commercial probiotic drink containing *Lactobacillus casei* strain Shirota on oral health in healthy dentate people. Microb Ecol Health Dis **2013**; 24.
35. Kovachev SM, Vatcheva-Dobrevska RS. Local probiotic therapy for vaginal *Candida albicans* infections. Probiotics Antimicrob Proteins **2015**; 7:38–44.
36. Wagner RD, Johnson SJ. Probiotic lactobacillus and estrogen effects on vaginal epithelial gene expression responses to *Candida albicans*. J Biomed Sci **2012**; 19:58.
37. Zakaria Gomaa E. Antimicrobial and anti-adhesive properties of biosurfactant produced by lactobacilli isolates, biofilm formation and aggregation ability. J Gen Appl Microbiol **2013**; 59:425–36.
38. Pirotta M, Gunn J, Chondros P, et al. Effect of *Lactobacillus* in preventing post-antibiotic vulvovaginal candidiasis: a randomised controlled trial. BMJ **2004**; 329:548.
39. Mendonça FH, Santos SS, Faria Ida S, Goncalves e Silva CR, Jorge AO, Leao MV. Effects of probiotic bacteria on *Candida* presence and IgA anti-*Candida* in the oral cavity of elderly. Braz Dent J **2012**; 23:534–8.
40. Gantner BN, Simmons RM, Underhill DM. Dectin-1 mediates macrophage recognition of *Candida albicans* yeast but not filaments. EMBO J **2005**; 24:1277–86.