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In vitro **analysis of epithelial tolerability and anti-*Candida* effect of a new lactic acid-based vaginal gel formulation**

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INTRODUCTION. Vulvovaginal candidiasis (VVC) is the most prevalent vaginal infection in adult women. It is mainly caused by *Candida albicans*, and it affects 75% of healthy women at least once during their reproductive age; 5-10% of such women have recurrent episodes (RVVC), with more of 4 episodes of acute VVC per year. Symptoms of VVC include itching, burning, swelling and redness of the vaginal mucosa with white vaginal discharge. The urinary system can also be affected, with pain and burning when urinating. This condition seriously damages the well-being and the life quality of the affected women. Since *Candida* is a commensal fungus of the vaginal mucosa of healthy women, the main question is how the fungus can switch from harmless component of the vaginal microbiota to virulent pathogen. In this work we analyzed the capacity of lactic acid-based vaginal gel formulation Respecta® Balance Gel (RBG) to counteract *C. albicans* virulence after epithelial cells infection *in vitro*.
MATERIALS AND METHODS. For the establishment of the *in vitro* infection model, we used a monolayer of the A-431 vaginal epithelial cell line and two different strains of *C. albicans* (strain SC5314 and the bioluminescent strain gLUC59). Dose-dependent experiments were performed to test the epithelial tolerability to RBG (IHS srl, Biofarma Group) by monitoring lactate-dehydrogenase (LDH) release from damaged cells. The capacity of RGB to counteract *Candida*-induced epithelial damage were analysed by monitoring LDH release from cells. Fungal growth and adhesion capacity during vaginal epithelial cells infection in the presence of RGB were evaluated by quantify the Relative Luminescent Units (RLU) and CFU counts, respectively.
RESULTS. Our results show that, at dilution 1:150, RGB is well tolerated by the vaginal epithelium and consequently we used this dose for the subsequent experiments. RBG was able to significantly reduce (by 65%) *C. albicans*-induced damage of vaginal epithelial cells. This effect was accompanied with the capacity of RGB to significantly reduce *Candida* adhesion to the epithelium (adhesion reduction by 34%). Intriguingly, no inhibition of fungal growth was observed after 24h of infection in the presence of RGB in our experimental conditions.
DISCUSSION AND CONCLUSIONS. Our results show that RGB significantly reduce *C. albicans*-induced damage of vaginal epithelial cells. One of the mechanisms underlying this effect is the inhibition of *C. albicans* adhesion to the vaginal epithelial cells, which may prevent *Candida* from penetrating and damaging epithelial cells, hence counteract *Candida* virulence. Collectively our preliminary results suggest that RBG can strengthen the VVC therapy favoring the establishment of an ecosystem that prevent *Candida* virulence.