Functional analysis of virulence potential from Gardnerella vaginalis and other anaerobes commonly associated with Bacterial vaginosis

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In the past half century, bacterial vaginosis (BV) has been a controversial topic in medical microbiology, and despite the wealth of information on this topic, the etiological agent has not yet been definitively identified [1]. The first advances on BV pointed Gardnerella vaginalis as the infectious causative agent of BV [2] but soon after it was found that G. vaginalis was also present in healthy women [3]. Additionally, G. vaginalis was not able to cause BV consistently. Furthermore, other microorganisms started to be associated with BV, and this resulted in a shift in the paradigm to that of a multispecies infection. However, epidemiological data revealed inconsistencies with this latter theory [4]. A couple of years ago the first descriptions of multispecies biofilm communities were described in BV [5]. Interestingly, G. vaginalis was present in most cases and accounted for the majority of the biofilm biomass. Further studies demonstrated that biofilm-forming G. vaginalis presented higher tolerance to external stresses [6].

Taking these data into consideration, we hypothesized that strains of G. vaginalis that were able to form biofilms could be the causative agent of BV. To test our hypothesis, we isolated more than 30 bacterial species from BV patients and also several strains of G. vaginalis from healthy women, and tested biofilm forming ability, initial adhesion to human vaginal cells, cytotoxicity activity, antimicrobial resistance and gene expression of known virulent genes.

Our results revealed that G. vaginalis outcompeted all the other bacterial species in the initial adhesion to the epithelial cells. Furthermore, when comparing BV-associated G. vaginalis strains to strains isolated from healthy women, we found that all 7 strains from BV were more virulent than the 7 strains colonizing healthy women, as measured by the higher cytotoxicity resistance profiles. Interestingly, no significant differences in expression of known virulence genes were detected, suggesting that the higher virulence of the BV-associated G. vaginalis was due to a yet unknown virulence determinant. We then tested virulent G. vaginalis against other known BV-associated anaerobe pathogens, namely Mobiluncus mulieris, Atopobium vaginae, Prevotella bivia and Fusobacteria nucleatum in mixed biofilm formation quantification. Interestingly, while the other tested anaerobes did not reveal a higher initial adhesion, they did enhance biofilm formation by G. vaginalis.

Overall, our data suggests that virulent variants of G. vaginalis have the potential to be the etiological agent of BV, while acknowledging that other anaerobes do enhance G. vaginalis virulence.

Keywords: Bacterial vaginosis; multi-species biofilms; virulent variants; gene expression; citotoxicity assays

References

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