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Understanding the vaginal microbiota: *Gardnerella vaginalis* in women with different clinical conditions

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Background: *Gardnerella vaginalis* (GV) is a facultative anaerobic, pleomorphic, Gram-negative to Gram-variable bacterium. GV is the most common species found in bacterial vaginosis (BV), a condition characterized by a depletion of vaginal lactobacilli and an overgrowth of anaerobes. Despite being the most prevalent and virulent species found in BV, GV can also be a part of the vaginal microbiota in a significant proportion of healthy women. Besides this aspect, less is known about the role of GV in other clinical conditions affecting the genital tract of women, such as vaginal candidiasis and *Chlamydia trachomatis* (CT) infections. The aim of this study was therefore to evaluate and compare GV load in the vaginal tract in 4 different conditions: healthy, BV, vaginal candidiasis and CT infection.

Material/methods: From July 2016, all the pre-menopausal women attending the STI Outpatients Clinic of Sant'Orsola-Malpighi Hospital in Bologna (Italy) and meeting one of the following criteria were enrolled: presence of vulvo-vaginal symptoms (as vaginal discharge, abnormal bleeding, itching) or presence of risk factors for CT infection (age < 25 years, multiple sex partners, unsafe intercourse). Exclusion criteria comprised the use of any antibiotics or vaginal medications in the past month, the use of estrogen-progestin products and the presence of chronic diseases. For all the patients, after a

clinical visit, a vaginal swab for molecular CT detection (Versant CT/GC DNA 1.0 Assay; Siemens) was collected. Moreover, Amsel criteria were used for BV assessment and a microscopic examination of secretions were performed for vaginal candidiasis. For each patient, starting from the remaining eluate of vaginal swab of Versant PCR plate, a quantitative real-time PCR targeting 16s rRNA gene of GV was performed. Results were expressed as GV DNA copies/reaction. Statistical comparisons of mean GV DNA load between groups were performed with one-way ANOVA. The study was approved by the Hospital Ethical Committee.

Results: Among all the women enrolled for the study, 24 were considered healthy (any symptoms and no microbiological positivity), 16 received a diagnosis of candidiasis, 18 of BV and 23 were positive for CT. As shown in the figure, the mean GV DNA load was significantly different between the groups ($P = 0.0186$). Healthy and CT positive women showed similar and lower mean loads (195274 and 162627 copies, respectively) compared to BV and candidiasis groups (562807 and 3.4×10^6 copies).

Conclusions: Vaginal GV load varies significantly considering different clinical conditions: CT positive women have low level of GV DNA comparable to healthy subjects, while women with candidiasis show even higher level of GV DNA than women with BV. Further studies are needed to better understand the role of GV and if the variations in GV levels precede or come after the onset of these conditions.

