

**Titolo: Sviluppo di un modello cellulare complesso di epitelio vaginale e ottimizzazione della colonizzazione da parte di batteri probiotici/patogeni**

Title: Set up of a complex cellular model of human vaginal epithelium and optimization of probiotic/pathogen colonization

**Breve descrizione dell'attività di ricerca**

Le infezioni e le condizioni infiammatorie del tratto genitale femminile rappresentano una delle cause più comuni di accesso alle cure mediche. I lattobacilli vaginali esercitano un'azione antagonista nei confronti della crescita eccessiva dei patogeni genitali, ma il loro contributo alla risposta immune locale e alla funzione di barriera dell'epitelio è ancora poco conosciuta, anche a causa della mancanza di modelli cellulari appropriati per lo studio di questi eventi.

L'obiettivo del presente progetto è lo studio di dell'interazione tra l'epitelio vaginale, i batteri benefici membri del microbiota e i patogeni, nel contesto della salute femminile. Questo obiettivo verrà raggiunto mediante lo sviluppo di un modello cellulare di epitelio vaginale umano e l'ottimizzazione di protocolli di infezioni/colonizzazione con patogeni e lattobacilli vaginali. La messa a punto del modello di interazione è funzionale alla successiva analisi del sistema mediante tecnologie -omiche.

Infections and inflammatory conditions of the female genital tract represent one of the most common reasons for women to seek medical care. Vaginal *Lactobacillus* spp. are known to exert antagonistic activity towards genital pathogen overgrowth, whereas their contribution to local immune response and epithelium barrier function is still very fragmented in the vaginal environment, also because of the lack of appropriate models to study these events in detail. The aim of the present project is to dissect the crosstalk between the vaginal epithelium, the beneficial members of the resident microbiota and pathogens, in the major context of vaginal health. Such aim will be achieved by the development of a reliable model of vaginal epithelium, that will be used for the optimization of infection protocols with vaginal lactobacilli and pathogens. Upon the establishment of the interaction model, cell cultures and their supernatants will be subjected to 'Omics' analysis.

**Progetto di Ricerca**

Infections and inflammatory conditions of the female genital tract represent one of the most common reasons for women to seek medical care. Aerobic vaginitis is characterized by inflammation of the vaginal epithelium and abnormal vaginal microflora containing aerobic and enteric bacteria, mainly *Escherichia coli*, Group B streptococci, and *Enterobacteriaceae*. These bacteria generally behave as vaginal commensals but, in certain conditions, can become pathogens, negatively impacting women's quality of life and reproductive health, besides causing high social costs. *Lactobacillus* spp. are known to exert antagonistic activity towards genital pathogen overgrowth, whereas their contribution to local immune response and epithelium barrier function is still very fragmented in the vaginal environment, also because of the lack of appropriate models to study these events in detail.

The aim of the present project is to dissect the crosstalk between the vaginal epithelium, the beneficial members of the resident microbiota and pathogens, in the major context of vaginal health. Such aim will be achieved by the development of a reliable model of vaginal epithelium, based on human vaginal cell lines. Different approaches for the realization of a complex, 3D model

of vaginal epithelium will be employed and the resulting cellular model will be used for the optimization of infection protocols. Both vaginal beneficial microbes and pathogens will be used to colonize and interact with the vaginal epithelium.

### **Piano di Attività**

The researcher will be involved in:

-Set up and optimization of a cellular model of vaginal epithelium: a multilayer vaginal cell models will be developed by culturing VK2/E6E7 or V19I (ATCC reference PCS-480-010) cells in trans-well insert systems or microcarrier beads. [Months 1-8]

-Optimization of probiotic/pathogen colonization of the vaginal model: different amounts of probiotic strains of *Lactobacillus crispatus* and of selected pathogen strains (*Entorococcus faecalis*, *E. coli*, *Streptococcus agalactiae*) will be employed to colonise/infect vaginal cells and the proper value of MOI (multiplicity of infection) will be determined. [Months 9-12]