

*Neisseria gonorrhoeae* is a gram-negative bacteria, etiologic agent of gonorrhoea. The only host of *N. gonorrhoeae* is human. This bacteria infects especially male and female genitourinary tract. Male genital tract infections mainly occur with manifestation of symptoms, while female genital tract infections are very often asymptomatic, what makes this infection difficult to eradicate. Beside molecular basis, differences in infection manifestation in men and women are due to different environmental conditions, like presence of hydrogen peroxide and reachability of iron ions in male and female genital tract. Delivery of bacterial virulence factors to environment or directly to host cells is a crucial mechanism for effective pathogenic infection. One of the strategies that is used in this purpose by many pathogenic gram-negative bacteria is production and release of outer membrane vesicles, that beside their function in pathogenesis, play also important role in stress response to environmental conditions. Releasing of outer membrane vesicles from surface of *N. gonorrhoeae* is also crucial for structures called biofilms formation by this bacteria. Bacteria within biofilm are protected from host's immune response and antibiotics. It was demonstrated that environmental factors like hydrogen peroxide and iron ions impact on outer membrane vesicles production as well as on biofilm structure of pathogenic bacteria.

The aim of the project is to investigate impact of environmental factors that are present in male and female reproductive tract, like different reachability of iron ions and hydrogen peroxide on production and cytotoxicity of outer membrane vesicles and structure of biofilm formed by *N. gonorrhoeae*.

In order to cytotoxicity assay of outer membrane vesicles and determination if host's cell death occurs due to induced cell death (apoptosis) or necrosis, commercial kits will be used. Expression of genes encoding proteins involved in apoptosis and antiapoptotic factors will be measured using RT-qPCR technique. This analysis enables determination of expression of many genes during one analysis. For investigation of changes in biofilm structure formed by *N. gonorrhoeae* in different environmental conditions, global proteomic analysis of biofilm extracellular matrix will be conducted using liquid chromatography coupled with mass spectrometry. Concentration of sugars and nucleic acids will be determined using standard colorimetric methods.

In conclusion, conducted analyses will help to understand pathogenesis of *N. gonorrhoeae*, which is a global medical problem, and improve our knowledge in applied and clinical microbiology.