

Diphtheria, caused by toxigenic strains of *Corynebacterium diphtheriae*, has been almost eliminated in developed countries thanks to introduction of obligatory antiphtheria vaccinations. However, during the last years in countries with high immunization rates, including Poland, infections caused by nontoxigenic *C. diphtheriae* strains, earlier regarded as apathogenic, have been recorded. Particularly disturbing is the fact that most of these infections are invasive with very high mortality rates. This suggests that the strains have acquired new virulence factors which enable them to disseminate and cause infections in vaccinated populations. In the planned studies, whole genome sequencing of ten *C. diphtheriae* strains will be conducted. The strains selected for investigation were isolated from patients with sepsis, endocarditis, wound infections and diphtheria in Poland, Mainland France and New Caledonia. Obtained sequences will be analyzed among other things in terms of presence of pathogenicity islands, prophages, potential virulence genes, such as genes related to adhesive and invasive properties or iron acquisition genes. The obtained results will enable us to verify the hypothesis concerning acquisition of new virulence genes by *C. diphtheriae* and will give information about bacterial factors influencing development of serious invasive infections.

The whole genome sequencing will be provided using two methods: Illumina DNA sequencing system using MiSeq platform and fourth-generation DNA sequencing system using MinION Nanopore Technologies. The MinION system is in a test stage currently, but will be available on the international market soon. Additionally, evaluation of the new fourth-generation sequencing system developed by Nanopore Technologies will be conducted in this project.

Currently little is known about virulence factors of nontoxigenic *C. diphtheriae*. The results of planned studies will give information not only about evolutionary changes which occurred in this bacterial species but also about virulence factors that are related to invasive and local infections. The results will contribute to understanding pathogenesis of infections caused by nontoxigenic *C. diphtheriae* strains, what can influence on recommendation for prevention and therapy of such infections. In the future, the knowledge about virulence factors of invasive *C. diphtheriae* strains will contribute to development of a new vaccine which will protect not only against diphtheria but also against infections due to nontoxigenic *C. diphtheriae*.