Abstract

Haemophilus influenzae is one of the main initiators of meningitis and pneumonia in children. Implementation of fast, cheap and instrumentally accessible method for detection of this pathogen would enable an early and targeted treatment of patients. The development of amplification methods in the last decades enables, apart from commonly used PCR method, application of alternative approaches, such as the LAMP. The focus of this Bachelor thesis was the study (research) of the alternative method LAMP as a tool for detection of *Haemophilus influenzae.*

The LAMP method was successfully implemented for *Haemophilus influenzae,* however, it has contended with the false positive results of negative control in case of longer incubation times. Therefore, the optimized LAMP method was designed in presence of deoxyuridine triphosphate and uracil-DNA glycosylase. Its aim was to change the structure of LAMP products via the incorporation of uracils to amplified regions of DNA and subsequent removal of uracils with influence of uracil-DNA glycosylase, and therefore prevent their replication during potential contamination of reaction mixtures and consequently reduce the risk of false positive results of negative controls to minimum. The concentration of deoxyuridine triphosphate in reaction mixtures was optimized for the purposes mentioned above. The fundamental contribution of the present thesis of optimized LAMP method for detection of *Haemophilus influenzae* was the finding that the uracil-DNA glycosylase effects the structure of the amplified DNA by the LAMP method in presence of deoxyuridine triphosphate. The approximate diagnostic window of the LAMP method without deoxyuridine triphosphate and of the optimized LAMP in presence of deoxyuridine triphosphate was determined.