

EVALUATION OF NOVEL CORONAVIRUS SARS-COV-2 NUCLEOCAPSID PROTEIN INTERACTION WITH SPECIFIC ANTIBODIES BY TOTAL INTERNAL REFLECTION ELLIPSOMETRY

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SARS-CoV-2 outbreak started at the end of 2019 and is still a major global public health concern. With 219 million cases and 4.5 million deaths (as of October, 2021) various methods of quick analyses are needed, for either antibody or antigen detection, especially in populations where herd immunity is not yet achieved. SARS-CoV-2 is a virus consisting of 4 structural proteins: envelope, membrane, spike and nucleocapsid. This work focuses on the nucleocapsid protein and its' interaction with specific antibodies [1].

During the infection process the nucleocapsid protein enters the host cell with viral RNA to begin virus particle replication. Nucleocapsid protein is highly immunogenic and can be detected in high concentration during an infection with SARS-CoV-2. Thus, while infected the host produces high amount of specific antibodies targeting spike and nucleocapsid proteins to combat the infection [2].

For the development of SARS-CoV-2 tests for either nucleocapsid antigen or antibody detection, information based on antigen/antibody binding kinetics is needed. To obtain such results various methods are utilised, however, non-destructive, label-free tools are of great importance [3]. Thus, recent attention is directed on the application of optical methods. One of such methods is spectroscopic ellipsometry, in total internal reflection mode (TIRE). Using TIRE, spectroscopic ellipsometry is combined with surface plasmon resonance effect to achieve high sensitivity. Due to, spectroscopic ellipsometry registering two parameters Ψ (light intensity amplitude) and Δ (light phase shift after reflection) simultaneously it is reported to be more accurate in detecting mass changes in the solid/liquid interface than commercial SPR, where only light intensity is registered. Thus, TIRE can be successfully applied for antigen/antibody affinity interaction evaluation [4].

In the presented work, we have evaluated the SARS-CoV-2 nucleocapsid protein interaction with specific antibodies isolated from immunized mice using TIRE method. Calculations of steric factor concluded that the complex formation requires strict orientation parameters. A mathematical model was applied to calculate rate of association, dissociation and affinity constants. Additionally, thermodynamic parameters of antigen/antibody complex formation were evaluated.

References

- [1] R. Lu *et al.*, "Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding," *Lancet*, vol. 395, no. 10224, pp. 565-574, Feb. 2020, doi: 10.1016/S0140-6736(20)30251-8.
- [2] Y. Cong *et al.*, "Nucleocapsid Protein Recruitment to Replication-Transcription Complexes Plays a Crucial Role in Coronaviral Life Cycle," *J. Virol.*, vol. 94, no. 4, pp. 1-21, 2019, doi: 10.1128/jvi.01925-19.
- [3] I. Plikusienė *et al.*, "Total internal reflection ellipsometry for kinetics-based assessment of bovine serum albumin immobilization on ZnO nanowires," *J. Mater. Chem. C*, vol. 9, no. 4, pp. 1345-1352, 2021, doi: 10.1039/d0tc05193d.
- [4] I. Plikusienė *et al.*, "Evaluation of kinetics and thermodynamics of interaction between immobilized SARS-CoV-2 nucleoprotein and specific antibodies by total internal reflection ellipsometry," *J. Colloid Interface Sci.*, vol. 594, pp. 195-203, 2021, doi: 10.1016/j.jcis.2021.02.100.