Real-Time Monitoring of Pore-Forming Toxin Activity in Live Cells Using High-Throughput Impedance Assays

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Pore-forming toxins (PFTs) are major virulence factors in many pathogens, disrupting cell membrane integrity and causing cell death. Beyond their pathogenic role, they can be harnessed for single-molecule sensing, where they are known as biological nanopores. Their capacity to kill cells by pore formation - sometimes involving specific receptors - also offers promising therapeutic potential, particularly in cancer treatment [1, 2]. However, for many PFTs, the detailed dynamics of action are often not completely understood.
In this study, we applied a high-throughput, label-free impedance assay to investigate the kinetics and cellular effects of two PFTs: SaroL-1 from *Salpingoeca rosetta* [3] and Aerolysin from *Aeromonas hydrophila* [4, 5]. Using cancer cell monolayers, we monitored real-time changes in electrical impedance to assess membrane disruption, cell adhesion, barrier function, and overall cellular health in both H1299 and HT-29 carcinoma cells. The method enabled us to differentiate between fast and delayed toxin activities, providing kinetic profiles of toxin action. Additionally, we examined the receptor specificity of SaroL-1 and Aerolysin towards the glycosphingolipid Gb3 by using Gb3-depleted cells and a specific inhibitor. The AtlaZ impedance platform enabled automated, scalable experiments, demonstrating its utility for screening neutralizing agents and elucidating dose-dependent PFT mechanisms. Our findings highlight impedance sensing as a powerful tool for real-time analysis of PFT-induced cytotoxicity and receptor dependency.

References:

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