KinExA® vs. SPR

A summary comparison of KinExA advantages vs. SPR

Technologies considered:
KinExA – Kinetic Exclusion Assay
SPR – Surface Plasmon Resonance

KinExA is a technique for measuring unmodified biomolecules with cell surface receptors or other biomolecules in solution. The following document describes advantages KinExA has over SPR techniques.

Cell Measurements:

Over half of all existing drugs are soluble molecules that target a cell membrane protein[1]. Membrane protein structure is partially dependent on the cell membrane so anytime the protein is isolated from the cell membrane and purified there is a risk that the binding epitope is modified, hidden, or eliminated. This is why there is such interest in measuring binding to the cells, rather than to purified soluble proteins.

KinExA – Measures binding of a drug (or candidate) to intact cells in any liquid matrix (media, buffer, serum, etc). KinExA has been demonstrated on both engineered over expressing cell lines[2,3] and on native cells expressing endogenous protein (unpublished). Measuring directly to the native cells is about as good as it can get in terms of biological relevance. KinExA has the sensitivity to measure sub pM K_d 's.

SPR – To our knowledge, no one has used a commercial SPR instrument with cells but there is literature on special apparatus. The first detects morphological changes in response to a binding event using cells grown on a gold substrate[4]. No binding constant is reported. The second case used SPR microscopy to measure binding of wheat-germ agglutinin (a lectin) to a single cell and reports a K_d of 0.32µM[5]. They also showed a noisy sensorgram for an antibody binding to a membrane protein, no K_d is given.

Solution Measurements:

KinExA – KinExA measures binding of the unmodified drug to the unmodified target in solution[6]. The solute can be anything in any buffer (serum, cell lysate, buffer, etc.). KinExA has been demonstrated accurate for many single digit pM K_d measurements and has measured K_d's as low as 12 fM[7].

SPR– These surface techniques require immobilization of one binding partner on the surface. The immobilization can cause conformational changes in the molecule leading to a change in K_d[8,9] or the surface itself can cause artifactual errors in the measured K_d[8]. Examples using SPR to measure single digit pM K_d's can be found in the literature but most users don't trust it for K_d's below 100 pM, with many not trusting it below 1 nM.

References:

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