



LigandTracer® for interaction analysis

TECHNOLOGY NOTE

LigandTracer® monitors molecular interactions on cells in real-time. The instrument is particularly suited for real-time monitoring of e.g. on-off rates, affinity or retention of ligands to cell-surface receptors. This note explains the basics of the LigandTracer technology.

LigandTracer technology

LigandTracer technology generates kinetic data based on repeated measurements of bound ligand concentration in a cell-covered area. The technology behind the LigandTracer is schematically outlined in Figure 1. Target cells are seeded in a local part of a circular cell dish and are allowed to attach firmly to the dish surface. The dish is then put on an inclined slowly rotating support and liquid containing labelled ligand is added to the dish. A detector is mounted over the elevated part of the dish, collimated to read an essentially liquid-free area. As the dish rotates the target cells will pass by the detector and a response from bound ligand will be recorded (See Figure 2). As the ligand interacts with the cells over time, a growing or declining response is generated, representing the kinetic behaviour of the interaction, as shown in Figure 3. Since LigandTracer measures interactions in real-time without the use of washes, a binding trace is obtained both during and after incubation. The differential signal (cell area minus reference area) becomes a background corrected measure of the amount of ligand attached to the cell-surface receptors.

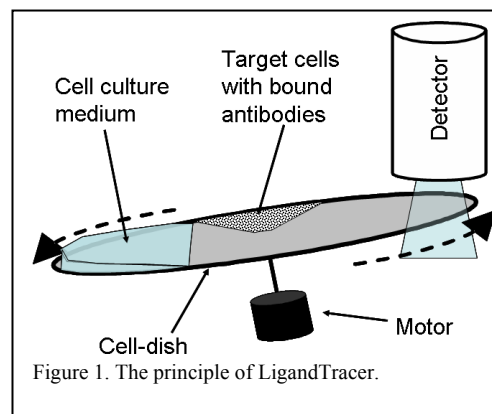


Figure 1. The principle of LigandTracer.

Results

LigandTracer has been used to characterize interactions between several different cells (including A431, SKOV-3, SKBR-3, SCC9, and HPAC) and ligands (including natural ligands, antibodies, and affibody molecules). The results obtained with LigandTracer agree with historical data obtained using manual protocols, and require less reagents. The unique real-time monitoring with LigandTracer has provided unperceived resolution of the binding data for interactions with rapid kinetics and on-off rates have been determined.

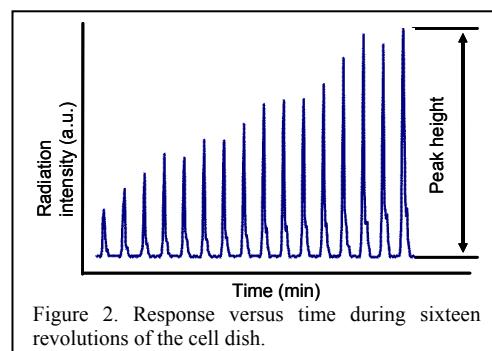


Figure 2. Response versus time during sixteen revolutions of the cell dish.

Conclusions

LigandTracer offers a simple and accurate method for real-time measurement of how ligands bind to cells. Real-time binding data is obtained during minutes, hours or days. Some applications are e.g. measurement of on-off rates, affinity or retention of ligands to cell-surface receptors. Measurements can be performed both during and after incubation and the required amount of reagents is reduced (compared to manual protocols). LigandTracer is available for radiolabelled ligands with γ -emission over 10 keV or β -emission over 100 keV.

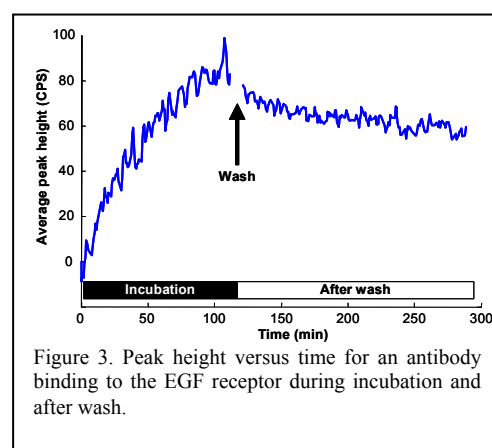


Figure 3. Peak height versus time for an antibody binding to the EGF receptor during incubation and after wash.

References

Application notes and references are available at www.ridgeviewinstruments.com