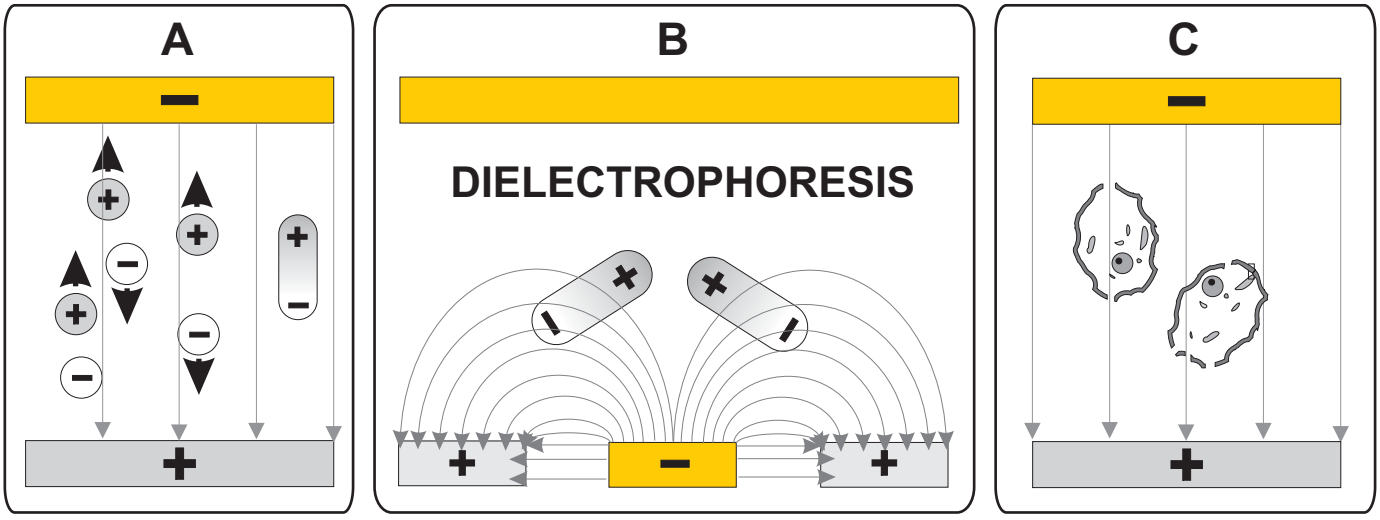
TIRF Labs, Inc.

Application Note

TIRF-EC - the Utility of ElectroChemical and Electric Field Control for TIRF Detection

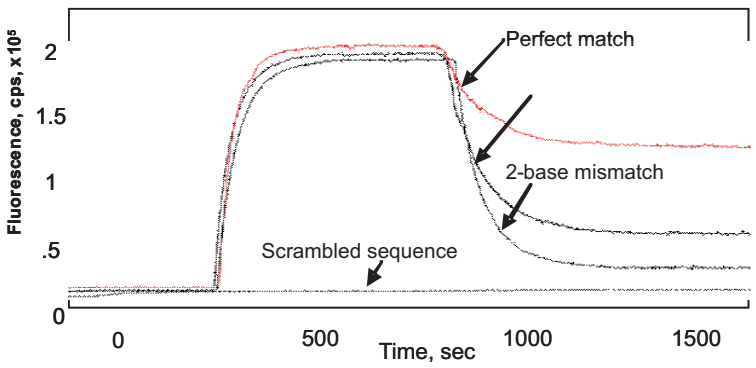


A- Electrostatic and electrochemical effects. Charged particles migrate between electrodes due to electrostatic and electrokinetic effects, permanent dipoles orient along the electric field. Strong electric field ~ 10^7 V/m in the double electric layer causes gradients of ions concentration, changes of surface chemistry and wettability, thus affects surface behavior of biomolecules, membranes and cells.

B- Dielectrophoretic (DEP) effects. Inhomogeneous AC electric field induces dipoles that migrate in gradients of electric field along the gradient (positive DEP), or against the gradient (negative DEP). Combination of DEP with electrostatic and electrochemical effects can be used to perform lysis or electroporation of cells and electrofocusing of low abundance DNA.

C - Effects of Pulsed Polarization. Short pulses of electrochemical polarization perform lysis - disintergation of cells, spores and tissues or electroporation - reversible disintegration of cell membranes to extract cell content or stimulate intake of exogenous compounds.

EC-Stimulated Dissociation for Discriminating SNP in DNA Targets

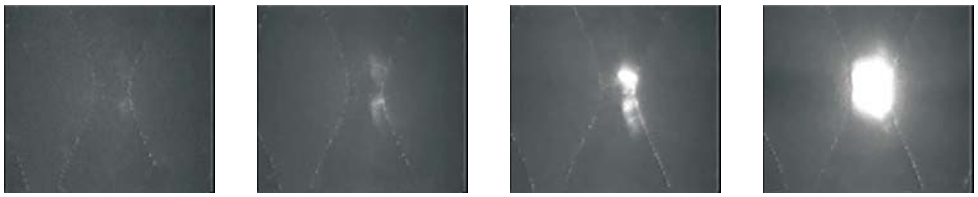


Discrimination of mismatched sequences of DNA target oligomers using EC stimulated dissociation. Molecular beacon assay is immobilized at the surface of TIRF-EC slide. The association (hybridization) stage exhibits close kinetics for single and double mismatched DNA sequences. Binding of DNA targets to molecular beacons is kinetically irreversible at room temperature. EC polarization stimulates accelerated dissociation, which kinetics is different for perfectly matched and mismatched DNA sequences.

Utility of Electric Field Control for TIRF Detection

TIRF-EC - the Utility of ElectroChemical and Electric Field Control for TIRF Detection

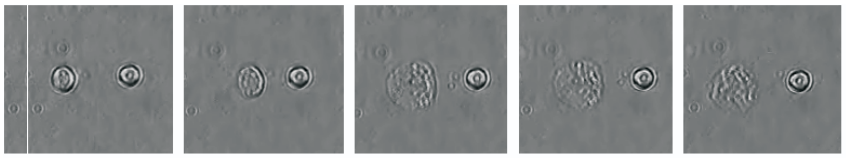
EC Focusing (Concentrating) of DNA in TIRF-EC Cell



1s 2s 4s 6s

Combination of electrostatic, electrophoretic, and dielectrophoretic forces stimulates focusing (concentrating) of low abundance DNA between patterned ITO electrodes at the surface of TIRF-EC slide. Then, the plug of concentrated DNA solution can be moved in microchannel along TIRF-EC surface for sensitive detection by an array of immobilized molecular beacon assays.

Dielectrophoretic Lysis of an Individual Cell



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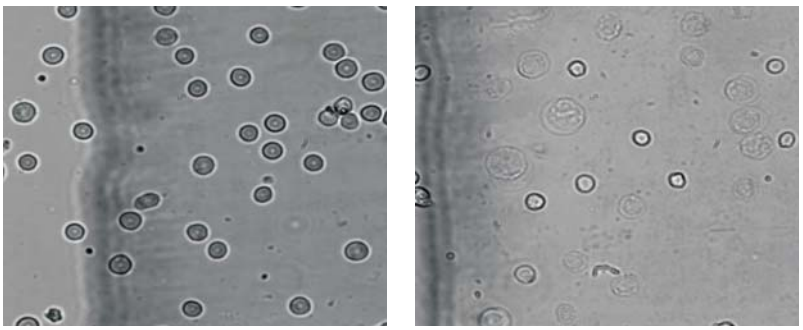
Dielectrophoretic Lysis of a Group of Cells



0 s 10 s