Certified Specialist Programme in Cell Culture Optimization

Automation and High-Throughput Screening

Automated Liquid Handling – a system that precisely transfers liquids using programmable robots. Related terms: pipetting robot, dispensing module. Enables reproducible reagent addition in 96-, 384-, or 1536-well plates. Example: a Hamilton STAR performs 10 µL transfers for a kinase assay. Practical application: large-scale drug-screening campaigns where manual pipetting would be error-prone. Challenges include cross-contamination, tip-clogging, and the need for regular calibration to maintain accuracy.

Automated Microscopy – integration of motorized stages, autofocus, and image-capture software to acquire high-content data without human intervention. Related terms: high-content imaging, image analysis pipeline. Used to monitor cell morphology after siRNA knock-down. Practical application: time-lapse monitoring of cell proliferation across dozens of plates. Challenges involve maintaining focus across uneven plates, data storage demands, and ensuring consistent illumination.

Automated Plate Reader – an instrument that measures absorbance, fluorescence, or luminescence in multi-well plates via robotic loading and detection. Related terms: multimode reader, spectrophotometer. Example: a BioTek Synergy H1 reads 384-well plates for a cell-viability assay. Practical application: rapid readout of enzymatic reactions in high-throughput screens. Challenges include well-to-well variability, edge effects, and the need for regular wavelength calibration.

Automation Workflow – the sequence of programmed steps that link liquid handling, incubation, detection, and data capture. Related terms: protocol scripting, process integration. A typical workflow may include cell seeding, compound addition, incubation, and plate reading. Practical application: end-to-end drug-screen pipelines that run unattended overnight. Challenges are error handling, synchronization of disparate devices, and ensuring software compatibility across vendors.

Batch Processing – grouping of samples or plates for simultaneous handling to maximize throughput. Related terms: plate pooling, run queue. Example: processing four 384-well plates as a single batch in a liquid-handling robot. Practical application: reduces set-up time and improves consistency. Challenges include limited flexibility for plate-specific conditions and the risk of propagating a single error across many samples.

Cell Counter – an automated device that enumerates cells using image analysis or flow cytometry. Related terms: hemocytometer, image-based cytometer. Example: a Countess II uses bright-field imaging to count suspension cells. Practical application: rapid determination of seeding density before plating. Challenges involve distinguishing debris from cells, handling clumped cells, and calibrating for different cell sizes.

Cell Dispensing – precise delivery of a defined number of cells into wells using automated equipment.

Related terms: droplet printer, acoustic dispenser. Example: a Thermo Fisher Multidrop Combi dispenses 2 000 cells per well into a 384-well plate. Practical application: uniform seeding for high-content screens. Challenges include maintaining cell viability during dispensing, preventing nozzle clogging, and achieving low coefficient of variation (CV) across wells.

Cell Line Authentication – verification that a cultured cell line matches its expected genetic profile, often using STR profiling. Related terms: mycoplasma testing, identity verification. Essential before large-scale screens to avoid misidentified cells. Practical application: routine authentication after 10 passages. Challenges are added time, cost, and integrating authentication data into laboratory information management systems (LIMS).

Cell Line Optimization – systematic adjustment of culture conditions (media, supplements, seeding density) to improve cell health and assay performance. Related terms: media formulation, growth curve. Example: testing five serum concentrations to maximize ATP signal in a viability assay. Practical application: increasing assay window for high-throughput screens. Challenges include combinatorial explosion of variables and the need for robust statistical analysis.

Cell Viability Assay – a test that measures the proportion of living cells, typically via metabolic activity (e.g., MTT, resazurin) or ATP levels. Related terms: cytotoxicity assay, live/dead staining. Example: CellTiter-Glo luminescence readout after 72 h drug exposure. Practical application: primary readout in compound screening. Challenges include assay interference from colored compounds, edge effects, and signal drift over long runs.

Cell Seeding Density – the number of cells plated per well, a critical parameter for assay reproducibility. Related terms: confluence, plating efficiency. Example: seeding 5 000 HeLa cells per well in a 96-well plate for a 48-h proliferation assay. Practical application: optimizing density to avoid over-confluence that masks drug effects. Challenges involve precise dispensing, accounting for cell growth rates, and adjusting for different cell lines.

Cell Tracking – computational analysis of cell movement over time using time-lapse images. Related terms: trajectory analysis, motility assay. Example: using ImageJ TrackMate to follow individual fibroblasts after treatment. Practical application: assessing migration inhibitors in a high-content screen. Challenges include image quality, cell overlap, and processing large datasets.

CellTiter-Glo – a luminescent assay that quantifies ATP as a proxy for viable cells. Related terms: ATP assay, luminescence readout. Used in 384-well format for rapid viability measurement. Practical application: primary screen of 10 000 compounds for cytotoxicity. Challenges include luminescence quenching by certain chemicals and the need for rapid plate reading to avoid signal decay.

Compound Library – a curated collection of chemical entities for screening. Related terms: chemical diversity, hit library. Example: a 50 000-compound library spanning drug-like space. Practical application: source of test agents in high-throughput screens. Challenges include solubility, stability, and ensuring consistent DMSO concentration across plates.

Control Wells – wells designated for positive, negative, or reference conditions within a plate. Related terms: assay controls, reference standards. Example: using 0.1 % DMSO as a negative control and a known inhibitor as a positive control. Practical application: normalizing data and assessing assay performance. Challenges involve placement to minimize edge effects and selecting appropriate controls for each assay type.

Cross-Contamination Prevention – strategies to avoid unintended transfer of liquids or cells between wells. Related terms: tip washing, aerosol control. Includes using filtered tips and de-contamination cycles. Practical application: maintaining library integrity during automated dispensing. Challenges are increased cycle time and the need for rigorous validation of cleaning protocols.

Data Analysis Pipeline – a series of computational steps that transform raw readouts into actionable results. Related terms: normalization, hit selection. Example: applying plate-based Z-score, then filtering by % inhibition. Practical application: rapid identification of active compounds. Challenges include handling large data volumes, batch effects, and integrating multiple assay readouts.

Data Integration – combining heterogeneous datasets (e.g., imaging, luminescence, genomics) into a unified analysis framework. Related terms: multi-omics, data fusion. Example: linking cell-morphology features with viability outcomes. Practical application: uncovering mechanistic insights from high-content screens. Challenges involve data standardization, storage, and cross-platform compatibility.

Data Normalization – statistical adjustment of raw assay signals to reduce systematic variation. Related terms: Z-score, robust scaling. Example: normalizing each plate to its median of control wells. Practical application: improving comparability across batches. Challenges include selecting appropriate reference points and avoiding over-correction that masks true biological effects.

Edge Effect – systematic variation in signal intensity observed at the outer wells of a plate, often due to temperature gradients or evaporation. Related terms: peripheral bias, plate uniformity. Example: higher luminescence in edge wells of a 384-well plate. Practical application: designing plate layouts that place controls away from edges. Challenges include mitigating with humidified incubators and using plate seals.

Equipment Calibration – routine verification and adjustment of instrument performance parameters. Related terms: validation, routine maintenance. Example: calibrating a liquid-handling robot’s pipette volume using gravimetric measurement. Practical application: ensuring accuracy of reagent dispensing. Challenges are time-consuming procedures and the need for skilled technicians.

Flow Cytometry – a technique that analyzes physical and fluorescent characteristics of cells in suspension. Related terms: fluorescence-activated cell sorting, FACS. Used for high-throughput phenotyping of cultured cells. Practical application: measuring surface marker expression after CRISPR perturbation. Challenges include sample preparation, data complexity, and instrument throughput limitations.

High-Content Screening (HCS) – a form of high-throughput screening that captures detailed cellular images and extracts multiparametric data. Related terms: phenotypic screening, image-based assay. Example: using an Opera Phenix to assess nuclear size after drug treatment. Practical application: discovering compounds that modulate subcellular localization. Challenges involve large data volumes, image analysis algorithm development, and longer assay times.

High-Throughput Screening (HTS) – rapid testing of thousands to millions of compounds against a biological target using automated platforms. Related terms: primary screen, hit identification. Example: a 10 000-compound library screened for kinase inhibition in 384-well format. Practical application: early-stage drug discovery. Challenges include assay miniaturization, false-positive/negative rates, and managing large data sets.

Incubator Integration – linking plate incubators directly to robotic workflows for seamless plate handling. Related terms: environmental control, robotic arm. Example: a Liconic automated incubator moves plates to a liquid-handler after 24 h. Practical application: reducing manual transfer and maintaining sterile conditions. Challenges are communication protocol compatibility and handling different plate formats.

Liquid-Handler Tip Management – strategy for selecting, loading, and disposing of pipette tips. Related terms: disposable tips, tip rack. Using filtered tips reduces aerosol contamination. Practical application: ensuring each dispense uses a fresh tip for critical reagents. Challenges include tip consumption cost, waste generation, and tip-clogging with viscous liquids.

Miniaturization – scaling down assay volumes and well sizes to increase throughput and reduce reagent consumption. Related terms: microplate format, nanoliter dispensing. Example: moving from 100 µL to 5 µL wells in a 1536-well plate. Practical application: cost-effective screening of expensive compounds. Challenges involve maintaining assay sensitivity, handling evaporation, and ensuring accurate dispensing at low volumes.

Multi-Mode Reader – an instrument capable of detecting multiple signal types (absorbance, fluorescence, luminescence) on the same platform. Related terms: plate reader, spectrofluorometer. Example: a PerkinElmer EnVision can read both FRET and luminescence assays. Practical application: consolidating assay types to a single device, saving space and time. Challenges include switching times between modes and calibration for each detection type.

Multiplex Assay – an assay that simultaneously measures multiple analytes or readouts in a single well. Related terms: dual-luminescence, bead-based assay. Example: combining ATP viability and caspase-3 activity in one plate. Practical application: increasing data density per sample. Challenges include assay interference, signal cross-talk, and more complex data analysis.

Normalization Controls – internal standards used to correct for plate-to-plate variation. Related terms: reference wells, calibrators. Example: adding a constant concentration of a fluorescent dye to each well. Practical application: improving comparability across runs. Challenges involve selecting a control that does not affect the biology under study.

Optical Calibration – process of verifying that detection optics (filters, lenses) produce accurate measurements. Related terms: wavelength verification, detector linearity. Using a fluorescence standard to set gain on a plate reader. Practical application: ensuring consistency across instruments in multi-site projects. Challenges include drift over time and the need for periodic re-calibration.

Plate Layout Design – strategic arrangement of samples, controls, and replicates within a plate to minimize systematic errors. Related terms: randomization, block design. Example: placing positive controls in a checkerboard pattern. Practical application: reducing edge effects and improving statistical power. Challenges are balancing randomization with practical constraints of robotic dispensing.

Plate Mapping – the digital representation of well positions and associated sample information. Related terms: well indexing, barcode linking. Mapping a 384-well plate to a CSV file that lists compound IDs. Practical application: tracking samples through the automation workflow. Challenges include preventing mismatches between physical and digital maps.

Plate Sealer – device that applies a breathable or airtight film to plates after dispensing. Related terms: foil seal, gas-permeable membrane. Example: using a heat-seal to prevent evaporation during long incubations. Practical application: maintaining consistent assay conditions. Challenges involve ensuring seal integrity without damaging plates or interfering with downstream detection.

Plate Reader Kinetic Mode – capability to measure signal changes over time directly in the plate reader. Related terms: real-time assay, time-course measurement. Example: monitoring fluorescence increase from an enzymatic reaction every minute for 30 min. Practical application: kinetic enzyme inhibition studies in HTS. Challenges include data volume, temperature control, and avoiding plate movement artifacts.

Plate Uniformity Assessment – evaluation of signal consistency across all wells of a plate. Related terms: Z′-factor, coefficient of variation. Performing a uniformity test with a fluorescent dye to calculate CV. Practical application: qualifying new plates before screening. Challenges include detecting subtle gradients and establishing acceptance criteria.

Positive Control – a well-treated sample known to produce a maximal response, used to validate assay performance. Related terms: reference compound, assay benchmark. Example: staurosporine at 1 µM for a cytotoxicity assay. Practical application: calculating Z′-factor and monitoring assay drift. Challenges are ensuring stability of the control compound and avoiding saturation of the detection system.

Process Automation Software – the suite that programs, monitors, and logs robotic operations. Related terms: workflow engine, scripting interface. Example: using Tecan EVOware to script a 96-well dispensing protocol. Practical application: reproducible execution of complex assay sequences. Challenges include software bugs, version incompatibility, and steep learning curves for non-programmers.

Quality Control (QC) Metrics – statistical parameters that assess assay reliability (Z′-factor, signal-to-noise, %CV). Related terms: assay validation, performance indicators. Example: a Z′-factor >0.5 indicates an excellent assay. Practical application: deciding whether to accept a screening run. Challenges are setting realistic thresholds for diverse assay types and maintaining QC over long campaigns.

Robotic Arm – a mechanical manipulator that transports plates between devices. Related terms: gantry robot, deck mover. Example: a Hamilton robot arm moves plates from a liquid handler to an incubator. Practical application: fully unattended screening cycles. Challenges include collision avoidance, precise positioning, and handling different plate formats.

Sample Tracking – the process of recording the movement and identity of each plate or well throughout a workflow. Related terms: barcode scanning, LIMS integration. Using RFID tags on plates to log each step. Practical application: preventing sample mix-ups in large screens. Challenges include ensuring reliable read-outs and synchronizing data across multiple instruments.

Scheduling Software – tool that queues and prioritizes automated tasks to maximize instrument utilization. Related terms: job scheduler, queue manager. Example: a master scheduler assigns liquid-handling jobs during off-peak hours. Practical application: optimizing throughput while avoiding conflicts. Challenges include handling emergency overrides and integrating diverse device APIs.

Signal-to-Noise Ratio (SNR) – the proportion of true assay signal relative to background variation. Related terms: dynamic range, assay sensitivity. An SNR of >10 is often desired for primary screens. Practical application: guiding assay optimization decisions. Challenges involve reducing background fluorescence and ensuring consistent detector settings.

Software API (Application Programming Interface) – a set of protocols that allow external programs to control instruments. Related terms: SDK, command line interface. Example: using a Python API to trigger a plate reader from a custom script. Practical application: integrating disparate devices into a unified workflow. Challenges include version drift, documentation quality, and security considerations.

Throughput – the number of samples processed per unit time, typically expressed as plates per day. Related terms: capacity, assay speed. A 384-well liquid handler may achieve 5 000 wells/hour. Practical application: planning campaign timelines. Challenges are bottlenecks in downstream detection and data analysis, which can limit overall throughput despite fast dispensing.

Validation Run – a pilot screening that confirms assay robustness before full-scale deployment. Related terms: pilot study, pre-screen. Example: testing 100 compounds in duplicate to assess Z′-factor. Practical application: identifying protocol tweaks early. Challenges include allocating resources for a run that does not generate primary data.

Viscosity Compensation – adjustments made to dispensing algorithms to account for liquid resistance. Related terms: shear-thickening, tip speed. Using slower aspiration rates for high-% DMSO solutions.

Practical application: preventing tip blockage and volume errors. Challenges involve measuring viscosity accurately and updating software parameters for each new reagent.

Workflow Integration – the seamless connection of all steps from cell culture to data output within an automated system. Related terms: end-to-end automation, process orchestration. Example: linking cell seeding, compound addition, incubation, and plate reading without manual intervention. Practical application: enabling 24/7 operation of a screening facility. Challenges include cross-vendor compatibility, error propagation, and maintaining flexibility for assay-specific tweaks.

Yield Optimization – strategies to maximize the number of usable data points per experiment. Related terms: assay window, dynamic range. Adjusting seeding density to increase signal while avoiding saturation. Practical application: improving hit detection rates. Challenges are balancing cell health with assay sensitivity and managing trade-offs between speed and data quality.

Z′-Factor – a statistical parameter that quantifies assay suitability for HTS; values >0.5 denote an excellent assay. Related terms: assay robustness, QC metric. Calculated from the means and standard deviations of positive and negative controls. Practical application: deciding whether a new assay can be deployed at scale. Challenges include variability in control wells, plate-to-plate drift, and the impact of outliers on the calculation.