

BURECO AG is a GMP and GLP certified bioanalytical service organization for the Life Science Industry worldwide. We offer biochemical, radiometric, immunological and cell based analytical technologies since 1992.

CELL BASED ASSAYS

CELL CULTURE ASSAYS FOR RELEASE AND STABILITY TESTING

Functional cell culture or cell binding assays are quantitative assays with an accuracy and precision approaching that of physico-chemical methods. These assays are used for:

- Biological activity or potency
- Release and stability testing
- Neutralizing antibodies

They are required for release and stability testing of biopharmaceuticals, since the complex molecular structure of biopharmaceuticals cannot be described sufficiently by physico-chemical methods alone. The same assay principals are applied to assess neutralizing antibodies against biopharmaceuticals.

BURECO AG offers all common assay formats for cell based assays such as:

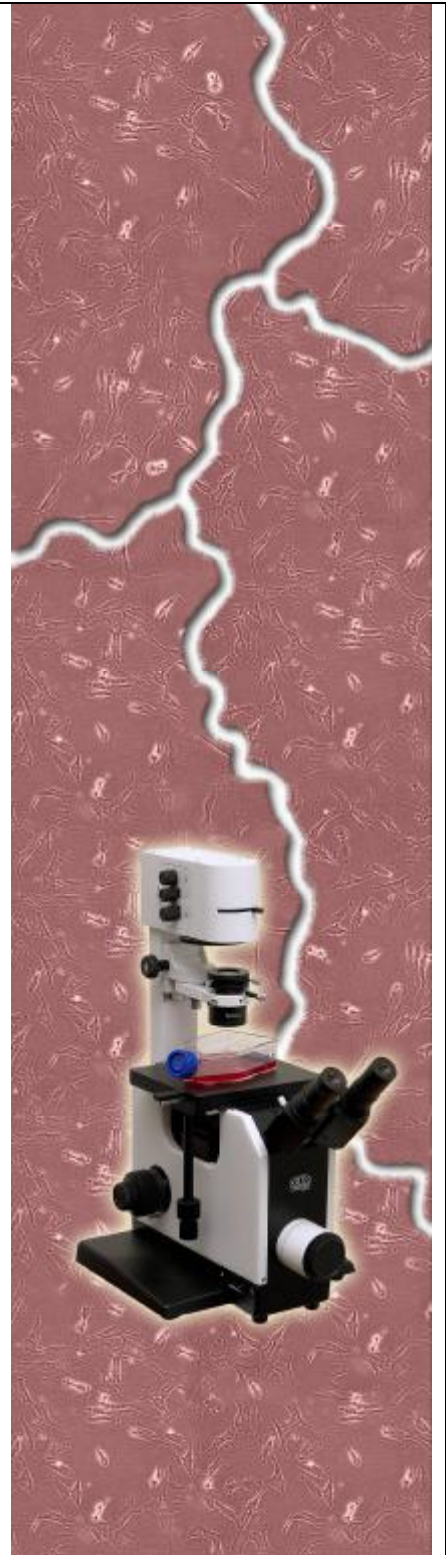
- Proliferation or proliferation inhibition assays
- Reporter gene assays
- Kinase receptor activation (KIRA) assays
- Phospho-specific antibody cell ELISA (PACE)
- Cell binding assays (cell ELISA).

Readout systems include UV/VIS spectrophotometry, chemiluminescence and fluorescence techniques.

Assays may be performed according to protocols compliant with EU and U.S. GMP guidelines ⁽¹⁻⁴⁾ and can be applied for release and stability testing of active pharmaceutical ingredients and final products.

ASSAY TECHNOLOGY FOR RELEASE AND STABILITY TESTING

Functional cell culture assays for release and stability testing are usually conducted in a 96-well format as parallel line assays in which a dose response curve of the unknown sample is compared to that of a reference substance. Typically, a suitable cell line is distributed in the wells of a microtiter plate, any necessary co-factors are added and a separately prepared dilution series of reference substance and test sample is added. Results are read using the appropriate read-out technology after 6 (e.g. reporter gene assays) to 72 (e.g. proliferation assays) hours. Subsequently the data are analysed using parallel line statistics ⁽⁴⁾.



DATA EVALUATION BY PARALLEL LINE STATISTICS

For data evaluation, BURECO AG has established the PLA software tool from Stegmann Systems (PLA 2.0) which is state-of-the-art and fulfils the requirements of EP 5.3 (section: statistical analysis 5.3.3.2). Figure 1 shows snapshots of this tool with real data.

Data evaluation can also be performed in PLA 2.0 using 4- or 5-parameter fit or using ratio of slopes to evaluate parallelism.

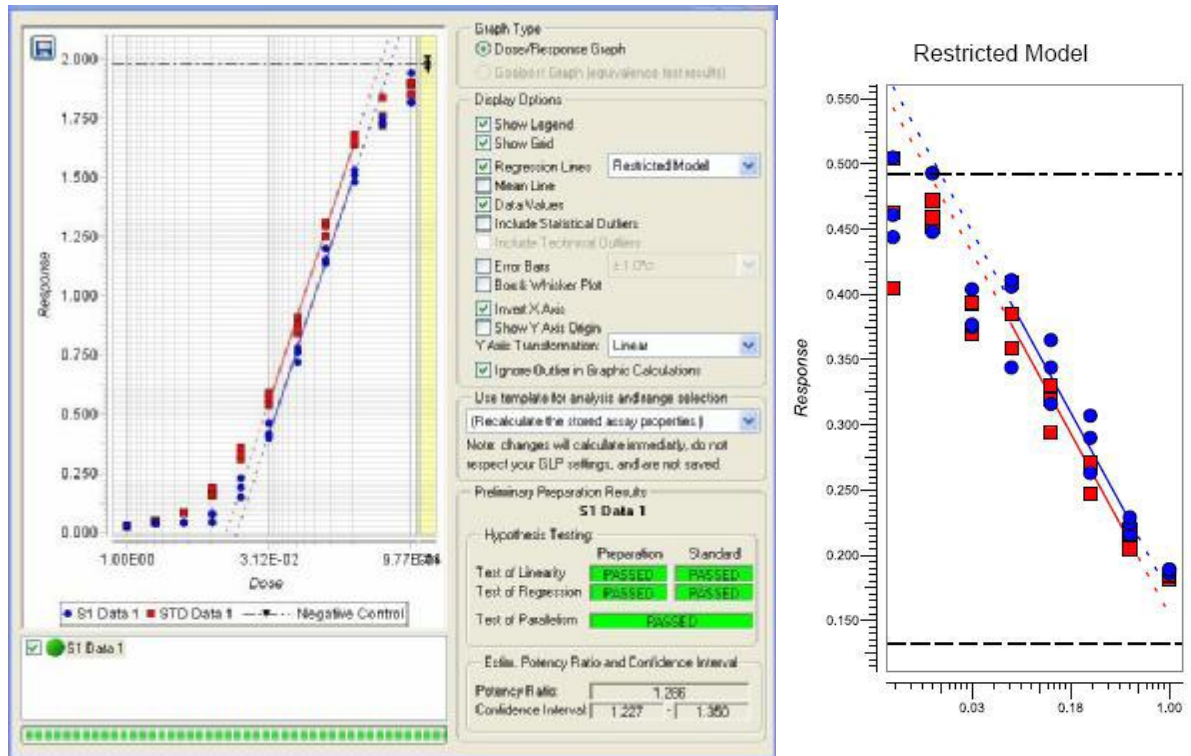


Fig 1: Evaluation of a Parallel Line Assay according to EP 5.3.3 using the software PLA 2.0 (Stegmann Systems)

References

- Ref. 1 EU GMP Guide
- Ref. 2 ICH Q7A, Good Manufacturing Practice for Active Pharmaceutical Ingredients
- Ref. 3 ICH Q5C, Stability Testing of Biological/Biotechnological Products
- Ref. 4 European Pharmacopoeia 5.3. Statistical Analysis, 5.3.3.2. The Parallel Line Model

