

Impedance-based cellular analysis – Z-factor

Assay validation and Z-factor calculation of GPCR activity with the **Bionas Discovery™ adcon** reader

INTRODUCTION

The quality and suitability of an assay is of major importance when testing the effectiveness and sensitivity of drugs during drug target identification. The Z-factor calculation is a useful mathematical tool to assess the quality and the effect size of an assay and is therefore used for assay validation and optimization. Here we performed a study with the **Bionas Discovery™ adcon** reader and determined a dynamic Z-factor calculation.

For that purpose, HeLa and U2OS cells were used to investigate the activation of S1P receptors. S1P receptors are G-protein coupled receptors (GPCRs) and are activated by sphingosine-1-phosphate (S1P). S1P is a bioactive sphingolipid metabolite formed by the phosphorylation of sphingosine which is ubiquitously expressed and implicated in the regulation of a variety of physiological cellular processes. Thus, S1P is an eligible measure and positive control for assay performance and Z-factor calculation.

RESULTS

For assay quality and stability assessment, dynamic Z-factor calculation was performed for HeLa cells (Figure 1) and U2OS cells (Figure 2) with the adcon software. The application of different concentrations of S1P resulted in a dose-dependent increase of the impedance in both cell lines. The dynamic Z-factor calculation provided essential information about the (time-dependent)

quality of the assay. In general, Z-factor values between 0.5 and 1 define an excellent assay¹. Within a few minutes, the values reached a stable level above 0.5 which was kept until the end of the experiment.

Cell line	Z-factor	Time-point
HeLa	0.76	45 min
U2OS	0.77	8 min

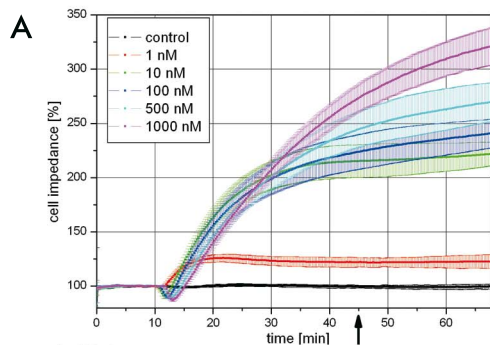


Fig. 1: HeLa cell assay evaluation.

A: Monitoring of the concentration-dependent S1P impedance. The arrow points to the time of best Z-factor determination. B: Dynamic Z-factor calculation for assay quality determination. The red bar indicates the timepoint with the highest Z-factor of 0.76, which was 45 min after adding S1P.

