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# Mitigating Spatial Bias in Cell Culture: Reducing Edge-Related Variability with Expansify™ Plates

**Introduction:**

Edge effect in cell culture is a common phenomenon where cells at the outer wells of multi-well plates grow differently from those in the interior. Factors such as increased evaporation, temperature gradients, and uneven nutrient distribution can cause slower or inconsistent proliferation at the plate edges. This variability can compromise experimental reproducibility, affect quantitative assays, and introduce bias in downstream applications such as high-throughput screening, imaging analyses, and cell-based assays. Data presented in this document demonstrate that Expansify™ 24-well plates effectively reduce or eliminate edge effect, promoting uniform cell growth across the wells and enabling more reliable outcomes for various downstream experimental applications.

**Procedure (Figure 1)**

1. Seed 5x10$^4$ cells per well in both standard polystyrene and Expansify™ 24-well plates.
2. Perform complete media change for Expansify™ plates only on Days 4, 5 and 6.
3. Quantify cell growth in polystyrene plates on Day 3.
4. Quantify cell growth in Expansify™ plates on Day 7.

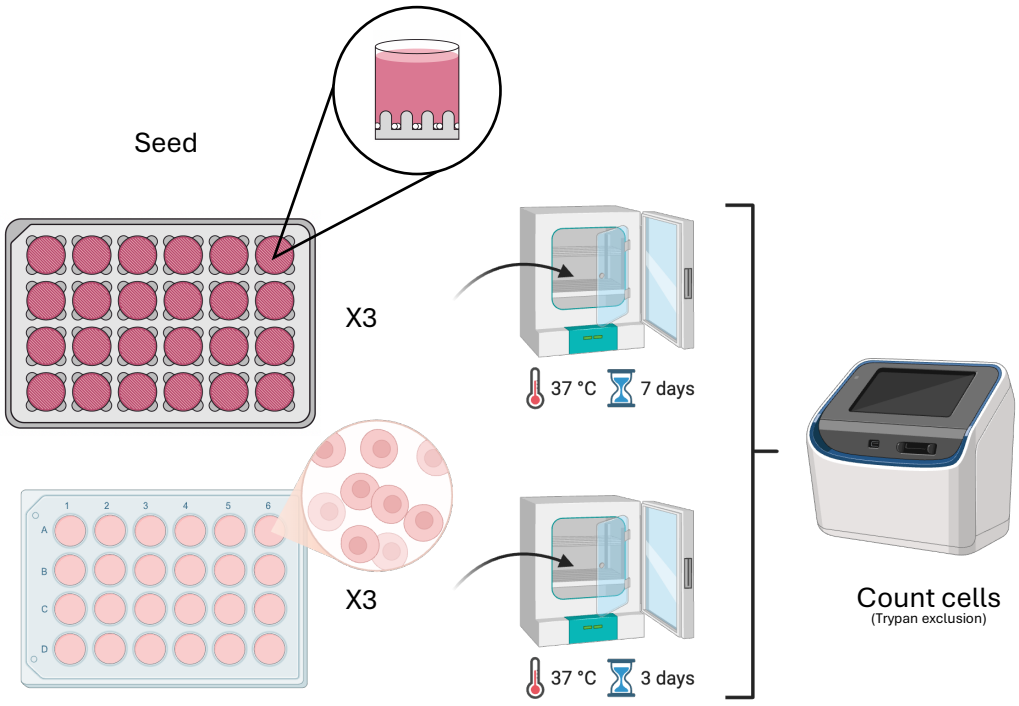


Figure 1. Overview of the experimental timeline comparing cell growth in standard polystyrene and Expansify™ 24-well plates

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## Expansify™ Plate Design Eliminates Positional Bias, Delivering Uniform Cell Growth Across the Entire Culture Surface (Figures 2, 3)

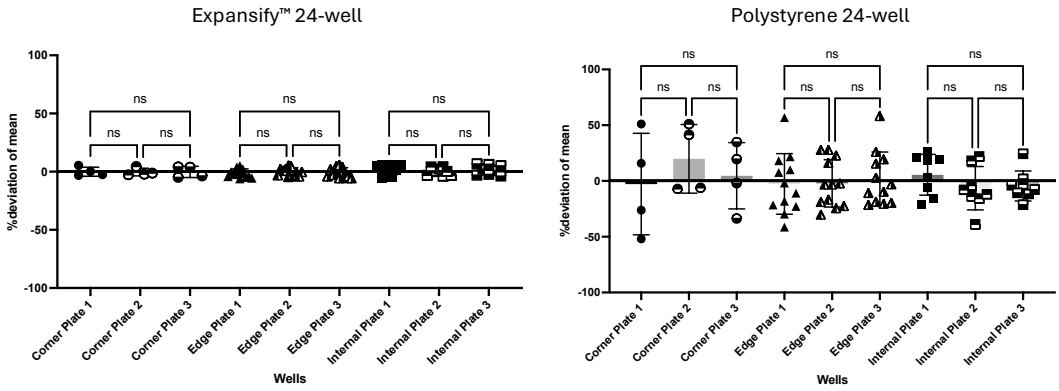


Figure 2. Two-way ANOVA with Tukey's test showed no significant positional differences (all adjusted *P* > 0.63, 95% CI overlapping zero). The Expansify™ plates has a low coefficient of variation (CV = 3.86%), confirming highly uniform and reproducible cell growth. In contrast, polystyrene plates show a much higher CV = 25.0%, indicating greater positional variability and reduced consistency across wells.

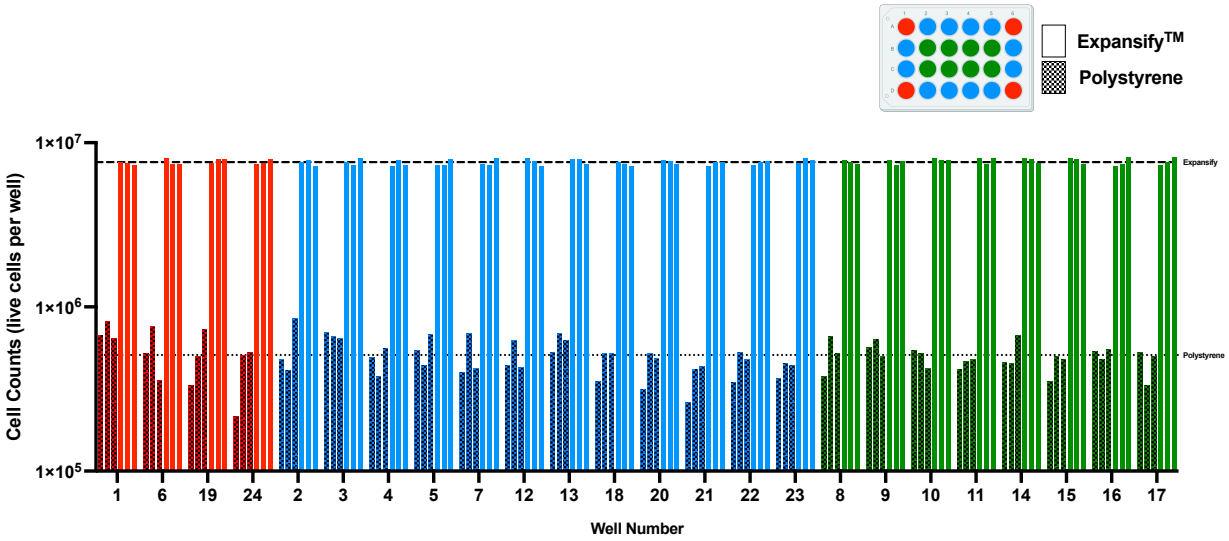


Figure 3. Bar graph showing raw cell count distribution across corner (red), edge (blue), and internal (green) wells. The Expansify™ plates show tightly clustered, consistent bar heights across all well positions, indicating uniform cell growth and absence of positional bias. In contrast, the polystyrene plates display broader variability, particularly at corner and edge wells, suggesting nonuniform growth and the presence of mild edge effects.

### Summary and Further Considerations:

Expansify™ plates exhibit uniform cell growth across all well positions, confirming consistent performance and minimal edge effects. In contrast, polystyrene plates show greater variability, indicating positional influence on cell growth. As the well area and media volume decrease in higher-density formats (48-, 96-, and 384-well plates), edge effects in polystyrene plates are expected to become more pronounced. These results highlight the superior reproducibility and spatial consistency of Expansify™ plates, making them well-suited for precision cell-based assays.