# A Comparative Study of Single and Dual Tray Incubator Shakers for HEK293 Cell Cultures

Iris Bodenmann iris.bodenmann@newbiologix.com
Mélissa Vona melissa.vona@newbiologix.com
Alexandre Felix alexandre.felix@newbiologix.com

NEW BIOLOGIX

***In cell culture*** **research, the choice of incubator shaker configuration can significantly impact experimental outcomes and laboratory efficiency. While single trays have long been a staple in cell culture laboratories, the advent of dual tray technology introduces a new paradigm, offering several distinct advantages over traditional setups:**

## Enhanced Capacity

Compared to single trays, which are limited by their singular workspace, dual trays effectively double the available surface area for culturing, enabling higher throughput and scalability.

## Improved Flexibility and Control

With dual trays, researchers can segregate cultures onto separate trays, minimizing the risk of cross-contamination and ensuring the reliability of experimental results.

## Streamlined Workflow

Dual trays optimize laboratory workflow by providing a more organized and efficient workspace. By compartmentalizing experiments onto separate trays, researchers can better manage samples, reduce the potential for errors, and enhance overall productivity.

## Space and Cost Efficiency

While single trays are limited to a single workspace, dual trays consolidate two incubator shakers into a single unit, maximizing laboratory space utilization. By eliminating the need for multiple standalone units, dual trays offer a cost-effective solution without compromising performance.

This application note presents a comparative study between single and dual tray incubator shakers for cultivating HEK293 and CHO cell cultures under various experimental conditions. The study evaluates the effects of tray configuration, cell culture volume, and position within the incubator on cell growth parameters, including density, doubling time, and water loss in the tubes.

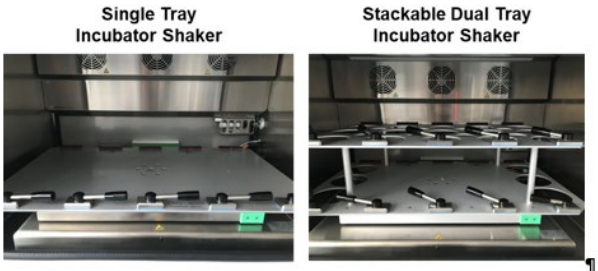


**Figure 1. Comparison of single tray and dual tray incubator shaker configurations**

Visual comparison between the single tray and dual tray configurations of the incubator shaker system used in the study.

## Experimental Design

A first experiment was designed with CHO cells in different volumes (10 mL and 30 mL). Cell cultures were shaken at 320 rpm using the standard Single Tray, which is our standard condition. Cell cultures were shaken at 300 rpm using the Dual Tray with a shaking diameter of 25 mm, as it was the maximum allowed speed with the Dual Tray.

A second experiment with HEK293 cell cultures was performed, with cultures prepared at different volumes (10 mL and 30 mL) with a 50 mm shaking diameter, at a speed of 225 rpm for both types of trays.

For both experiments, the cultures were placed at various positions across the rack levels to assess any potential differences in growth parameters. Output measurements included cell density, doubling time, and water loss in the tubes.

**Table 1 Summarized conditions for Experiment 1 and Experiment 2**

| | *Experiment 1* | *Experiment 2* |
|---|---|---|
| ***Cell Line*** | **CHO** | **HEK293** |
| ***Volumes*** *(mL)* | **10, 30** | **10, 30** |
| ***Rotor shaking diameter*** *(mm)* | **25** | **50** |
| ***Speed*** *(rpm)* | **300/320** | **225** |

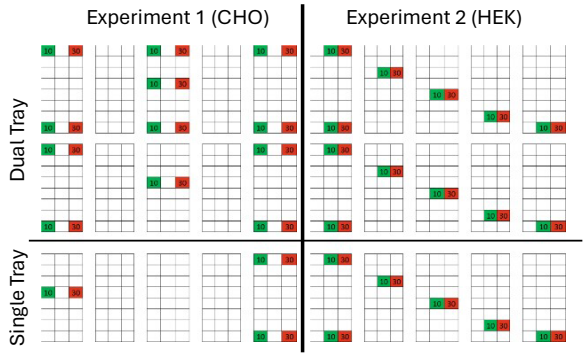


**Figure 2 Schematic Representation of Tube Positions in Single and Dual Tray Configurations and their volume.**

Schematic illustration detailing the arrangement of tubes within both Single and Dual Tray configurations of the incubator shaker system. Additionally, the figure delineates the different volumes of CHO or HEK293 cell cultures tested during the study.

# A Comparative Study of Single and Dual Tray Incubator Shakers for HEK293 Cell Cultures

## Results

Cell Density:

The results indicate that both Single and Dual Tray configurations supported homogeneous cell growth of HEK293 cells (Fig. 3) and CHO cells (Fig. 4) across the tested volumes (10 mL and 30 mL) and tray types. There was also no significant variation between the upper and lower levels within the Dual Tray for HEK293 cells. A slight increase in VCD was observed for CHO cells in both 10 mL and 30 mL volumes with the upper level within the Dual Tray. However, this increase was not significant and was not observed in the other experiment using HEK293 cells.

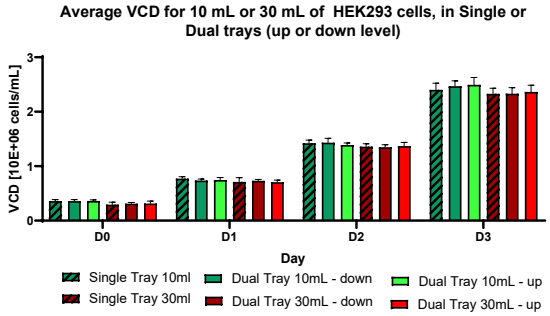


**Figure 3. Viable Cell Density (VCD) of HEK293 cells Across Different Volumes and Positions in Single and Dual Tray Configurations**

Viable cell density (VCD) observed across volumes (10 or 30mL) and positions in both Single and Dual Tray configurations of the incubator shaker system. The figure provides insight into how cell density varies under different experimental conditions over days of cultivation.

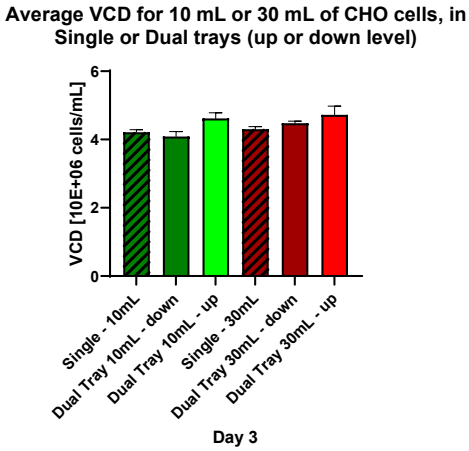


**Figure 4. Viable Cell Density (VCD) of CHO cells Across Different Volumes and Positions in Single and Dual Tray Configurations**

Viable cell density (VCD) observed across volumes (10 or 30mL) and positions in both Single and Dual Tray configurations of the incubator shaker system. Measurement at day 3 post seeding.

Viability :

The viability was also monitored, but did not show any differences across volume, tray configuration or localisation within the incubator. The average viability at day 3 for the HEK293 was of 94.7%, with a standard deviation of 1.4, and of 98.7% with a standard deviation of 0.4 for CHO cells.

Doubling Time:

Analysis of doubling time revealed consistent growth kinetics across all experimental conditions. There was no significant difference in doubling time between single and dual tray configurations (Fig. 5) or across rack levels.

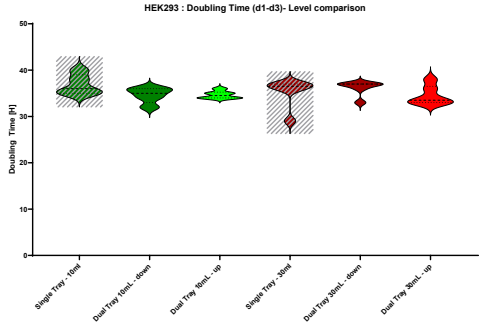


**Figure 5. Doubling Time of HEK293 Across Different Volumes and Positions in Single and Dual Tray Configurations**

Doubling time observed across various volumes and positions within both single and dual tray configurations of the incubator shaker system.

Water Loss:

Evaluation of water loss in the tubes showed minimal variation across different tray configurations and positions within the incubator. The residual mass of water exhibited small fluctuations (0.001-0.1g) compared to the control incubator, indicating efficient humidity control regardless of tray type or position.

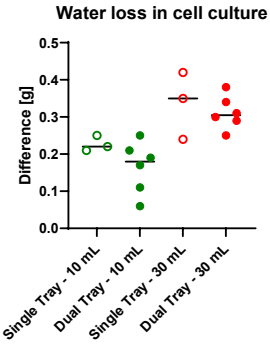


**Figure 6: Water Loss in Tubes Across Different Positions and Tube Volumes**

Water loss observed in the tubes positioned at various locations within the incubator shaker system, as well as the influence of different tube volumes on this phenomenon. The graph provides valuable insights into the dynamics of water evaporation under different experimental conditions.

# A Comparative Study of Single and Dual Tray Incubator Shakers for HEK293 Cell Cultures

## Results

Visual observations :

The movement of the wave generated within the culture by the shaking incubator can be evaluated by observing the ring of dead cells. Interestingly, this movement differs between the Dual Tray and the Single Tray (Fig. 7). However, as shown by the results presented above, there was no impact on cell growth and viability, regardless of the tray type.

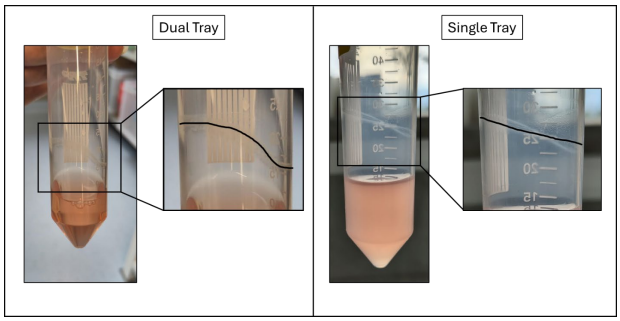


**Figure 7: Picture of cultures placed in the two types of incubator shakers.**

Dead cells form a rings deposited at the top of the wave movement created by the incubator shaker. The wave created by the Dual Tray upper level (left) or Single Tray (right) differ.

## Conclusions

The comparative study between Single and Dual Tray incubator shakers for HEK293 and CHO cell cultures demonstrates comparable performance in supporting cell growth under various experimental conditions. These findings highlight the suitability of both Single and Dual Tray incubator shakers for cell culture applications, offering researchers flexibility and efficiency in experimental design and execution.

In conclusion, Dual Trays represent a significant advancement in incubator shaker technology, offering unparalleled benefits over traditional Single Tray configurations. With their increased capacity, flexibility, experimental control, workflow optimization, and cost efficiency, Dual Trays empower researchers to conduct more robust and productive experiments in cell culture research.