Technical Note:

Baffled or Non-Baffled Shake Flasks, that is the Question.

Introduction

Microbial shake flask experiments are the lifeblood of biotechnological research, providing a controlled environment for microbes to thrive. Microbes, used in various biotechnological applications, rely on oxygen, carbon, nitrogen, phosphate, and trace elements for growth and production. Oxygen, a critical element for microbial growth and production, is a focal point in optimizing these experiments, and ensuring proper oxygen supply is vital.

Several factors influence oxygen transfer from the surrounding environment into the liquid phase, and understanding them is crucial for optimizing microbial cultures. These factors include the surface/volume ratio, filling volume/shake flask size, shaking frequency/shaking diameter, flask wall properties (hydrophobic/hydrophilic), viscosity, and osmolarity/salinity. The oxygen transfer rate (OTR) and oxygen uptake rate (OUR) are crucial considerations.

<u>Oxygen Transfer Rate</u>

The oxygen transfer rate measures the amount of oxygen transferred into a specific volume of liquid per unit time, and is typically expressed in mmol/L/h. It is a critical parameter in aerobic cell-based bioprocesses, such as bioreactors, spinner flasks, and shake flasks, because it governs the oxygen available for microorganisms or cells to metabolize substrates efficiently.

<u>Oxygen Uptake Rate</u>

In environmental and industrial contexts, oxygen uptake rate measures the rate at which microorganisms consume oxygen in aerobic cell-based bioprocesses. It is determined by monitoring dissolved oxygen (DO) over time in a sample. High OUR indicates high biological activity, while low OUR suggests a reduced microbial metabolism.

Shake flasks are available with or without baffles. When should a baffled flask be selected over a non-baffled one?

Baffled Shake Flasks

Baffled flasks are designed with indentations or baffles along the inner surface that are specifically designed to improve critical aspects of the cell culture process.

<u>Advantages of baffled shake flasks</u>

Enhanced Oxygen Transfer. Baffles disrupt laminar flow, creating turbulence that increases the liquid's surface area exposed to air. This significantly boosts the oxygen transfer rate (OTR), often doubling it compared to a standard, non-baffled flask. This is especially important for fast-growing or oxygen-demanding microbes, as oxygen limitation can slow growth and reduce yields.

A 250mL capacity Bellco baffled shake flask with 3 lower side baffles and a break resistant lip.

Part number 2542-00250

Better Mixing and Reduced Stratification. The turbulence from baffles promotes more uniform mixing, preventing oxygen-deprived zones and ensuring all parts of the culture receive adequate oxygen. This is particularly beneficial for filamentous, aggregated, or fungal organisms, which can be challenging to oxygenate evenly.

Improved Growth and Productivity. By maintaining higher oxygen levels, baffled flasks can support faster growth rates and higher biomass yields, which can translate into better product titers in bioproduction.

Consistent Surface-to-Volume Ratio. Baffles increase the effective surface area for gas exchange, optimizing the surface/volume ratio for oxygen uptake.

Support for High-Density Cultures. The improved aeration and mixing make baffled flasks suitable for scaling up small-scale cultures toward larger bioreactor-like conditions, helping maintain viability and productivity.

Reduced Foam Formation (in some designs). Some advanced baffled flasks are engineered to minimize foam while still providing vigorous mixing, thereby reducing mechanical stress on cells.

Disadvantages of baffled shake flasks

Variations. Achieving consistent results across multiple experiments can be challenging due to variations in flask design, baffling patterns, and manufacturing processes from different suppliers.

Turbulence. Increased turbulence within baffled flasks may lead to excessive foam formation, necessitating.
additional measures to control foam and prevent potential cell damage.

Shear Stress. The enhanced mixing in baffled flasks may introduce shear stresses that can affect the growth and normal development of delicate microbial cells, potentially impacting their performance.

Mathematical Modeling. Baffled flasks present difficulties in developing and applying mathematical models due to their complex design, making it challenging to predict and control oxygen transfer accurately. This is compounded if baffled flasks of different designs are used in parallel or consecutive experiments.

Non-Baffled Shake Flasks

In contrast, non-baffled flasks lack the internal indentations, have a smooth internal wall, and a much simpler, cleaner design. This simpler design facilitates greater reproducibility across experiments.

<u>Advantages of standard shake flasks</u>

Lower Turbulence. The absence of baffles reduces turbulence; the smooth walls generate laminar flow, which effectively reduces the OTR. Standard shake flasks work well with microorganisms that require a low OTR.

Foam Formation. Minimizing foam formation reduces the mechanical stress on the cells and eliminates the need for anti-foams, thus simplifying experimental control.

Shear Stress. Non-baffled flasks, by creating a more uniform, less turbulent, and more laminar flow mixing environment, subject microbes to lower shear stresses. This can be critical when working with delicate cells that are sensitive to mechanical forces.

<u>Disadvantages of standard shake flasks</u>

Oxygen Transfer Rate. Under identical operating conditions, cultures in non-baffled flasks will exhibit lower oxygen transfer rates than in baffled flasks. Thus, potentially impacts microbial growth and production.

Conclusions.

The choice between baffled and non-baffled flasks is determined by the specific needs of the microbial strain and the goals of the experiment. The advantages and challenges of each flask format must be considered, given the cell type and its susceptibility and response to factors such as reproducibility, foam formation, shear sensitivity, high or low OTR, and cell density. Understanding the nuances of both baffled and non-baffled flasks is crucial for the successful development of an experimental plan.

References

1. Meier, K., et al. (2016). *Correlation for the maximum oxygen transfer capacity in shake flasks for a wide range of operating conditions and for different culture media. Biochemical Engineering Journal*, **109**, 228–235.
2. Büchs, J. (2004). *Introduction to advantages and problems of shaken cultures. Biochemical Engineering Journal*, **17**(3), 91–98.
3. McDaniel, L. E., et al. (1965). *Effect of Oxygen-Supply Rates on Growth of Escherichia coli: I. Studies in Unbaffled and Baffled Shake Flasks. Applied Microbiology*, **13**(1), 109–114.

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