BASICS PAPER

WHAT IS SHAKING

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Safety recommendations with regard to the weight distribution:

- Please ensure that the shaker's platform is loaded symmetrically
- Avoid placing the heavier vessels on the edge of the shaker or, in the case of a multi-tier structure, on the upper level

WHAT IS SHAKING

By definition, shaking is a back and forth motion of objects. In a laboratory context, the term Shaking is used especially with respect to mixing different solutions or to keeping suspensions in motion. The first automated shaker was patented and launched in 1959 as a vortexer. Since then, shakers in different versions have become indispensable in laboratories. Besides the traditional vortexers, laboratory staff can nowadays also benefit from different platform shakers. In the following sections, you will learn more about the different types of motion of platform shakers. Particular attention will be paid to the orbital shaking movement theory as this is the most commonly used type of motion in many microbiology and cell culture laboratories.

What is a platform shaker?

A platform shaker is laboratory equipment with a platform that oscillates in two or three directions. It is used to move and mix the mostly liquid contents of different vessels. The platform of a shaker is moved by appropriate motors and mechanic. There are devices with an orbital (circular), circular vibrating (vortexing), reciprocating (back and forth), wave or rocking movement pattern. In most cases, different attachments and clamps can be fixed on the platform to enable the use of different vessels. The following sections highlight the different movement patterns of platform shakers focusing on the orbital movement.

Focus on different types of motion

ORBITAL

The orbital shaking movement is a basic technique in laboratories used to mix solutions, cultivate cells and to perform various experiments. The understanding of the theory behind the orbital movement and how orbital shakers work can help you to optimize their use for specific applications.

Theory behind the orbital movement

The orbital movement of the orbital shakers is generally based on the principles of the circular motion and the centripetal force, both of them being central concepts of classical mechanics.

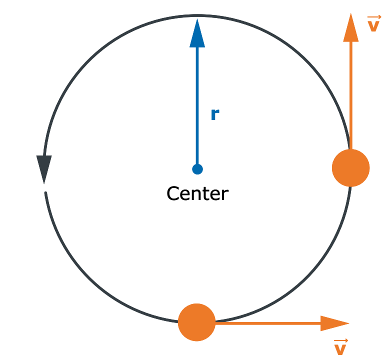

Fig. 1: Schematic representation of the circular motion

Circular motion

An object moving with constant speed on a circular orbit undergoes a continuous change of direction. This means that it constantly accelerates although its speed remains constant.

Centripetal force

By Newton's second law, each acceleration is caused by a force. In the case of the circular motion, the centripetal acceleration is caused by a centripetal force. This force points in the direction of the circular orbit's center and keeps the object on this orbit. Mathematically, the centripetal force (Fc) is given by the formula,

$$F_c = \frac{mv^2}{r}$$

where m is the object's mass, v its tangential velocity and r the radius of the circular orbit.

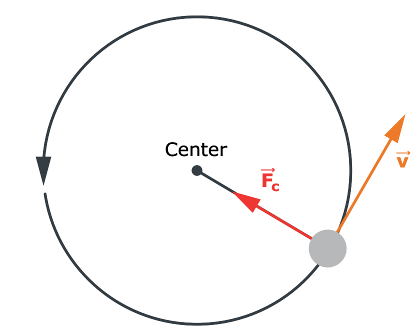

Fig. 2: Schematic representation of the centripetal force

The orbital shaking movement includes a circular motion where a platform moves on a fixed circular orbit. This kind of motion is achieved by mounting the platform on a central pivot or an axis rotating around a fixed point. The most important parameters defining this movement are the orbital diameter (radius of the circular orbit)[1] and the shaking speed (revolutions per minute/rpm).

Radius of the circular orbit and rotation speed (rpm)

The radius of the circular orbit defines the level of the mixing effect, where larger diameters cause a stronger shaking and a more thorough mixing movement while maintaining the same speed. The rotation speed (rpm) is the speed which moves the platform on its circular orbit. Higher speeds increase the transferable power to the medium. This power can enhance the mixing movement but can also lead to splashes and damages to sensitive samples.

[1] Common alternative terms are e.g. orbit size, rotation diameter, stroke length.

RECIPROCATING

The reciprocating movement is a violent horizontal shaking movement and is therefore suitable for different extraction applications, e.g. babcock analysis or the determination of pesticide residues. Particularly where an increased number of samples has to be handled during a liquid-liquid extraction, used with suitable holding clamps for separatory funnels, the workflow can be facilitated.

WAVE AND ROCKING

The particularly gentle movement of 2D rocking and 3D wave shakers is used, among other things, for staining membranes after blotting processes. In addition, both types of motion are often used to prevent coagulation in blood samples or when cell cultures have to be gently flushed by medium.

Why is motion so important for cell growth?

The next sections cover important parameters which have a decisive influence on cell growth and can be positively influenced using platform shakers.

Improved oxygen supply

The motion of cell suspensions improves the volumetric mass transfer coefficient (kLa)[1] as well as the oxygen transfer rate (OTR)[2]. Both parameters are a measure for the efficiency of oxygen transfer from the gaseous phase into the liquid phase in a culture system. By shaking the culture medium, shakers increase the contact surface (exposed to air) to the atmospheric oxygen and thus enhance an efficient gas exchange. The continuous motion helps to distribute oxygen in the whole medium and prevents oxygen gradients being able to lead to local fatigue. This constant mixture also ensures that nutrients and oxygen are evenly distributed, optimizing cell growth and cell activity. Moreover, the improved oxygen transfer increases the total yield and productivity of the culture process. Shakers contribute to an homogeneous culture environment by preventing sedimentation and by keeping cells in suspension. This mechanical movement not only supports a better oxygen supply but also helps to maintain a constant pH value and a constant temperature in the whole culture vessel. Hence the use of shakers is crucial to obtain cultures with high cell density and to maximize the efficiency of biological processes. In conclusion, laboratory shakers significantly increase the oxygen transfer rate in cell and bacterial cultures supporting healthier and more productive cultures.

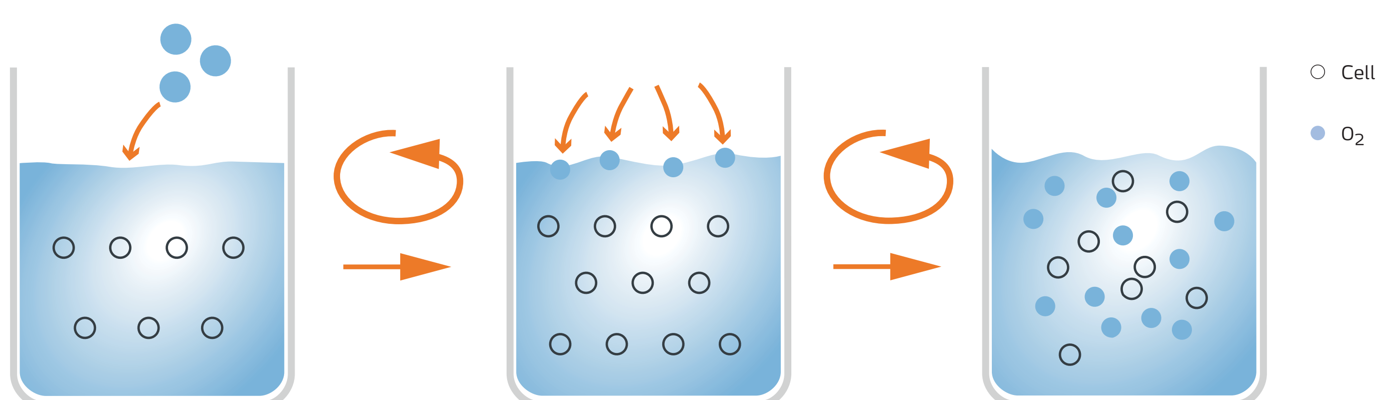

Fig. 3: Simplified representation of oxygen distribution in an agitated vessel

1 Volumetric mass transfer coefficient (kLa) = parameter that transfers the oxygen rate or the rate of other gases into the culture per volume unit per hour
2 Oxygen transfer rate OTR = parameter describing the speed at which gases are delivered into the liquid medium (mmol O2/(Lxh))

Uniform distribution of nutrients

The orbital shaking movement ensures a uniform distribution of nutrients in the culture medium by mixing the solution continuously. Hence, all nutrients contained in the nutrient medium are distributed homogeneously. Prokaryotic as well as eukaryotic cells thus have a better access to essential medium components being necessary for growth and metabolism. An increased cell viability, improved rates of growth and more homogeneous cell populations are only some of the advantages of this uniform distribution of nutrients. The cells are not excluded from critical nutrients or exposed to local fatigue zones. This optimal environment supports robust cell cultures and a higher reproducibility of experiments.

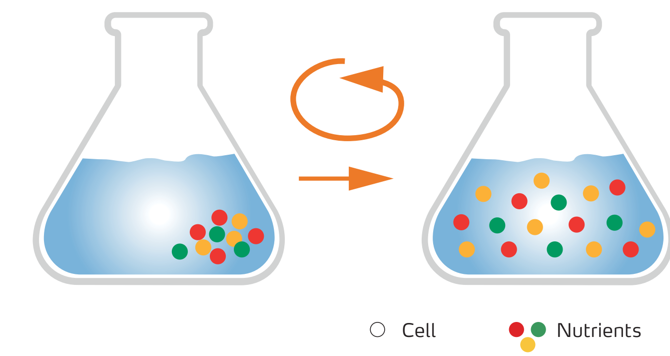

Fig. 4: Simplified representation of uniform distribution of nutrients in an Erlenmeyer flask

Reduced formation of cell clumps

As the culture medium moves continuously, the orbital movement in shakers reduces the formation of cell clumps. Hence, the cells are kept in suspension and are evenly distributed. In the case of bacterial cells, this motion avoids the formation of aggregates hindering the access to nutrients and oxygen, supports a more uniform growth and reduces the risk of local hypoxic or low-nutrient zones. In the case of mammalian cell cultures, the reduced formation of cell clumps ensures a better cell-cell interaction and minimizes the stress resulting from physical collisions within dense clusters. The improved distribution promotes a more consistent culture environment which leads to improved growth rates, viability and general cell health.

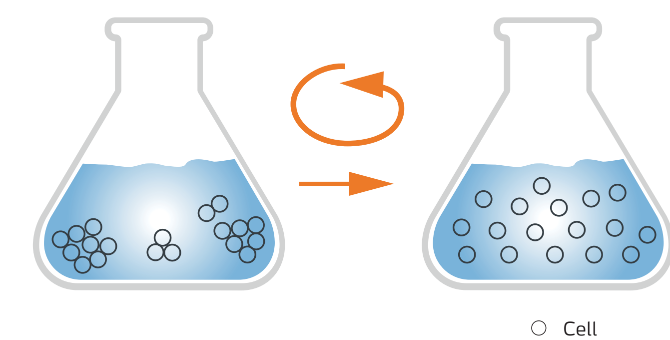

Fig. 5: Schematic representation of the dissolution of cell clumps induced by motion

Summary

The use of orbital shakers, 2D rocking shakers or even 3D wave shakers is a crucial technology in laboratory environments improving the efficiency of different applications. Of particular note is the continuous circular motion that improves the oxygen supply (kLa and OTR) and, at the same time, provides an homogeneous distribution of nutrients and reduces the formation of cell clumps. These factors are decisive for a healthy cell growth. For bacterial cultures and mammal cells, this means a better oxygen transfer and even access to nutrients, improved cell-cell interactions and minimized stress. Generally, the optimized environment created by an even movement supports a robust cell growth, higher viability and more reproducible experimental results. This comprehensive understanding emphasizes the use of different platform shakers in laboratories.

Sources: **1)** Madigan, M. T., Martinko, J. M., Stahl, D. A., Clark, D. P., Biology of Microorganisms, Brock, 2012, 13. Auflage; **2)** S. Schmitz, Der Experimentator: Zellkultur, 2011, Spektrum Akademischer Verlag, Heidelberg, 3. Auflage; **3)** https://doi.org/10.1007/978-3-8274-2573-7; **4)** L.M/H.L/S.Gä, Lexikon der Biologie: Zellkultur; www.spektrum.de/lexikon/biologie/zellkultur/71584; **5)** https://www.leifiphysik.de/mechanik/kreisbewegung; **6)** https://www.leifiphysik.de/mechanik/kreisbewegung/grundwissen/zentripetalkraft

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