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Research review paper

Effects of shear stress on microalgae – A review

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ABSTRACT

Cultivation of microalgae requires consideration of shear stress, which is generated by operations such as mixing, circulation, aeration and pumping that are designed to facilitate mass and heat transfer as well as light distribution in cultures. Excessive shear stress can cause increased cell mortality, decreased growth rate and cell viability, or even cell lysis. This review examines the sources of shear stress in different cultivation systems, shear stress tolerance of different microalgal species and the physiological factors and environmental conditions that may affect shear sensitivity, and potential approaches to mitigate the detrimental effects of shear stress. In general, green algae have the greatest tolerance to shear stress, followed by cyanobacteria, haptophytes, red algae, and diatoms, with dinoflagellates comprising the most shear-sensitive species. The shear-sensitivity of microalgae is determined primarily by cell wall strength, cell morphology and the presence of flagella. Turbulence, eddy size, and viscosity are the most prominent parameters affecting shear stress to microalgal cells during cultivation.

1. Introduction

Microalgae encompass a large group of unicellular and simple multicellular aquatic organisms with vast genetic diversity. They are simple to cultivate, grow rapidly under heterotrophic or photoautotrophic conditions (Chisti, 2007), and have been employed in numerous applications including human food supplements, feed for aquaculture (Harun et al., 2010), and the production of pigments, antioxidants, and other novel algal products (Pulz and Gross, 2004). There is a particular interest in large-scale cultivation of microalgae for biomitigation of CO_2 (Wang et al., 2008) and the sustainable production of biofuels (Li et al., 2008).

Shear stress, a type of hydrodynamic stress generated by mixing and/or aeration, is a critical factor requiring careful consideration in microalgal cultivation at different scales. On one hand, shear stress is associated with aeration and mixing, both of which are essential for microalgal cultures and are particularly important for large-scale microalgal farming. On the other hand, excessive shear stress can cause reduced cell growth and productivity, severe cell damage and even cell lysis (Rodriguez et al., 2009). A balance must be found where mixing and aeration are sufficient for optimal cell growth without compromising cell integrity, which could be particularly challenging for species that are sensitive or ultrasensitive to shear stress.

This review aims to provide a comprehensive survey on the sources of shear stress in different microalgal cultivation systems, the tolerance of different microalgal species to shear stress, and environmental conditions that could help mitigate shear stress-induced damage to microalgal cells.

2. Source of shear stress

Microalgal cultivation is a complex process with multiple operations where shear stress can be generated. These operations can be classified into the following categories: 1) mechanical mixing, 2) aeration, and 3) pumping. Shear stress imposed to microalgal cells could be estimated using the following equation,

$$\tau = \gamma \cdot \mu \quad (1)$$

where τ is shear stress (Pa), γ is shear rate (s^{-1}), and μ is apparent viscosity (Pa s).

Sufficient mixing is crucial for both laboratory cultivation and industrial farming of microalgae, where microalgae are grown at cell-densities much higher than that in natural niches (Suh and Lee, 2003). Mixing circulates fluid, which helps maintain a homogenous culture suspension and ensure that each cell has access to CO_2, other nutrients, and light (Contreras et al., 1998; Hodaifa et al., 2010). It also prevents cell sedimentation (Leupold et al., 2013), facilitates the removal of O_2 as a by-product of photosynthesis, and prevents cell growth on the reactor walls (Ramírez-Duque et al., 2012). Shear stress is generated in mixing by micro-eddies of similar or smaller size than microalgal cells.

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Aeration is required in most microalgal cultures, particularly those of large scale, for the following functions: 1) supplying CO_2; 2) serving as the sweeping gas for the removal of O_2 produced in photosynthesis; and 3) in some cases, providing the power for mixing and circulation. While fluid circulation created by aeration may contribute to certain extent, shear stress in aeration is mostly generated by the bursting of bubbles – which also results in other types of detrimental forces such as the tension force (Chalmers, 2015) – at the culture surface where gas-liquid separation occurs.

Pumping of cell suspensions is needed in some large-scale cultivation systems for purposes such as transportation, circulation, deoxygenation and mixing. When centrifugal pumps are used, shear stress is generated by the high-speed rotation of pump impellers. This usually creates much larger shear stress than is observed in mechanically agitated cultures (e.g., stirred-tank photobioreactor, alias PBR). Therefore, air-lift pumps are becoming a popular alternative.

2.1. Mechanically agitated cultures

Mechanical agitation is commonly used in open ponds and stirred tank PBRs for mixing.

2.1.1. Open pond

Agitation is achieved by a rotating arm in circular ponds or by paddlewheels in raceway ponds. As agitation levels are low, shear stress from mixing is usually not a significant concern. Aeration with CO_2-enriched air is usually necessary for open ponds. Since these ponds are typically shallow, and air bubbles tend to be much larger than microalgal cells, shear stress associated with aeration is also negligible in most cases.

Open ponds are simple to construct, inexpensive to operate, and of relatively low shear stress. However, the limited control over operating conditions greatly reduces the potential for microalgal biomass production and biosynthesis (Wang et al., 2012). Mixing in an open pond is also typically insufficient for proper mass transfer of CO_2, leaving a heterogenous distribution of cells and nutrients (Koller, 2015). Recently, greater efforts have been made in developing and implementing high-efficiency closed systems, which allow for greater productivity and reduced risk of contamination although at much higher capital and operational costs (Brennan and Owende, 2010; Wang et al., 2012). Nevertheless, these closed system are in general expensive to build and operate, and suffers from the complexity in association with problems such as biofouling. For instance, when shear tolerant green alga *Tetraselmis suecica* was cultivated in a 40 L tubular PBR with a centrifugal pump for recirculation, biofouling started to occur in the tubes 1 day at the maximum pumping rate, i.e., 3.6 $m^3 h^{-1}$, which represented a hydraulic residence time < 1 min (Michels et al., 2016).

2.1.2. Stirred tank PBR

Stirred tank PBRs contain stirring devices to provide agitation (Singh and Sharma, 2012). While impellers are commonly used for mechanical agitation in stirred tank reactors (Gumery et al., 2009), other devices such as agitation bars (Fadlallah et al., 2016) or magnetic stirrers (Massart et al., 2014) are typically used for small vessels. Alternatively, cultures can be shaken (Camacho et al., 2007; Ojo et al., 2014) or wave-mixed (Hillig et al., 2014).

Aeration is required in stirred tank PBRs to provide CO_2 as a carbon source and remove O_2 (Suh and Lee, 2003). The shear stress generated by aeration is generally inconsequential relative to that by agitation (Perez-Garcia and Bashan, 2015). The majority of shear stress is associated with the circulation of fluid, interactions between cells and the stirring apparatus and reactor walls (Brindley Alías et al., 2004; Bronnenmeier and Märkl, 1982), and most importantly, the micro-eddies necessary for mixing.

Agitation is commonly expressed by revolutions per minute (rpm) (Hodaifa et al., 2010) or tip speed (m s^{-1}) (Leupold et al., 2013). Tip speed is determined by:

$$u_{tip} = r\,\omega \tag{2}$$

where r is the radius of the stirring apparatus or orbit in metres (m)and ω is the angular velocity (Leupold et al., 2013). In this review, when applicable, rpm rates have been converted to m s^{-1} and Eq. (1) is converted to the following:

$$u_{tip} = (2\pi\, r\, n)/60 \tag{3}$$

where n is the rotation rate in rpm and 60 is used to convert rpm to rps.

In a stirred-tank PBR, assuming laminar flow, shear rate is given by the following equation (Newton's Law):

$$\gamma = \frac{du}{dy} \tag{4}$$

Eq. (1) is therefore given as follows,

$$\tau = \mu\frac{du}{dy} \tag{5}$$

However, most industrial photobioreactors and algal ponds work in turbulent regime and laminar flow exists only in boundary layers and is of secondary importance. In this situation, the relative velocity that determines the stress acting on microalgal cells is the velocity of the turbulent fluctuations. The turbulence of flow is created by eddies of different sizes and the energy content of eddies decreases with their size. Shear stress imposed to cells is determined mainly by eddies of a size comparable to that of the cells because larger eddies are followed by the cells as a convective movement, whereas smaller eddies have too low energy intensity to be important. For estimation of the Reynold shear stress in the velocity field of a turbulent flow, please refer to the book of Henzler (2000).

Stirred tank PBRs are widely used in laboratory settings and are valuable apparatuses for determining the shear sensitivity of different microalgal species. Scale-up of stirred tank reactors for heterotrophic cultures is relatively simple (Perez-Garcia and Bashan, 2015). However, they have a low surface area to volume ratio, which limits the overall light exposure for cells in autotrophic cultures, especially upon scale-up (Singh and Sharma, 2012). It is also generally believed that stirred tank PBRs are not suitable for the cultivation of fragile microalgae due to significant shear stress, especially at the tip ends of impellers (Koller, 2015). Nevertheless, a scale-up study for a shear-sensitive dinoflagellate microalga using stirred tank PBR with low impeller speed showed positive results (Camacho et al., 2011).

2.2. Air-agitated cultures

Aeration is usually required in microalgal cultures for gas exchange, i.e., supplementation of CO_2 as a carbon source and removal of O_2 (Ugwu et al., 2008). In some PBRs, aeration is also used as a means of delivering power for mixing and circulation. In such air-agitated cultures, shear stress is generated by fluid circulation, micro-eddies, and the rupturing of air bubbles at the culture surface (Leupold et al., 2013). Cells can also be damaged by air bubbles at the sparger (Barbosa et al., 2003), with small bubbles being more damaging than large bubbles (Barbosa et al., 2003; Sobczuk et al., 2006). Shear stress imposed by aeration is typically less than that by mechanical agitation (Perez-Garcia and Bashan, 2015).

The rate of aeration can be expressed as specific air flow rate Q (volume air per volume culture per minute, vvm) (Khoo et al., 2016) or superficial gas velocity U_g (m s^{-1}) (Barbosa et al., 2003) for a given volumetric gas flow rate of F_g (m^3 s^{-1}) as follows:

$$Q = F_g/V \tag{5}$$

$$U_g = 4F_g/(\pi D^2) \tag{6}$$

here, V (m^3) is the working volume of the PBR, and D (m) is the