



Improved liquid foam-bed photobioreactor design for microalgae cultivation

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ABSTRACT

The liquid foam-bed photobioreactor is a recently developed photobioreactor concept that allows for cost reduction in microalgae cultivation. Long term operation was not yet achieved, due to degradation of the surfactant. In this study, Pluronic F68 was applied for foam stabilization. In order to compensate for the low algae partitioning to Pluronic F68 stabilized foam, liquid recirculation was introduced into the reactor design. The microalgal suspension was continuously pumped from the bottom of the reactor to the top of the foam column where it was allowed to drain down again through the foam. This new design allowed increased mixing and, thereby, a homogenous algae distribution within the reactor. The volumetric mass transfer coefficient for CO₂ in the foam-bed was 0.14 s⁻¹, revealing that the gas transfer rate is an order of magnitude higher compared to bubble column reactors. This characteristic, together with a very high gas residence time, allows for a dramatic reduction in gas flow rate and a high carbon dioxide utilization efficiency. Long-term cultivation (> 500 h) of *Chlorella* sp. was achieved in a stable foam-bed. The areal productivity of the foam-bed photobioreactor was 57 g m⁻² d⁻¹, which is slightly lower than maximally achieved in flat panels under similar conditions. This is possibly related to substantial light scattering taking place in the foam leading to a steeper light gradient and increased reflection. The reactor dilution rate and the liquid recirculation rate were not optimized during our experiments and there is room for further improvement. During continuous reactor operation, biomass densities of > 20 g L⁻¹ could be maintained. This biomass density is a factor of 10 higher compared to traditional, liquid phase photobioreactors, thereby, contributing to reduced energy requirements for microalgae harvesting.

1. Introduction

Current photobioreactors are not yet economically feasible for mass production of microalgae [1–3]. In order to reduce production costs, an alternative photobioreactor design, utilizing foam instead of liquid for microalgae production was developed [4]. This liquid foam-bed photobioreactor is a promising microalgae cultivation concept, since it will allow for the reduction of harvesting costs due to the high biomass densities achieved. In addition, reduction in energy requirements are expected because of reduced gassing rates, enhanced mass transfer, and lower pressure drop in the foam-bed photobioreactor. The reduced gassing rates are achieved as a result of the enhanced mass transfer and increased gas residence time.

In the first explorative experiments on the foam-bed photobioreactor, the protein Bovine Serum Albumin was applied as surfactant. However, due to the loss of the foaming properties of this molecule, only short-term growth experiments could be carried out with a

duration of 8 h [4]. A detailed comparison of surfactants showed that Pluronic F68 is a promising surfactant for the foam-bed photobioreactor [5]. Pluronics are amphiphilic, non-ionic block copolymers, consisting of poly(ethylene oxide) and poly(propylene oxide) blocks [6]. Pluronics are widely used in cosmetics [7] or biomedical applications [8], and Pluronic F68 specifically is applied for shear protection in animal cell cultures [9]. When the liquid foam-bed photobioreactor is operated with Pluronic F68 as a surfactant instead of proteins, a longer operation time is expected since it showed no biodegradability in two week tests. In contrast to its good foaming properties and lack of toxicity, the drawback of this surfactant is the low microalgae partitioning to the foam phase. Therefore, the microalgal cells are not equally distributed over the foam and the underlying liquid phase; upon foaming the microalgal concentration is enhanced in the underlying liquid and reduced in the foam phase. Thus, a large fraction of the algal culture is not illuminated, resulting in reduced growth rates.

In order to eliminate the problem of low algal partitioning to the

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foam phase when Pluronic F68 is applied, a new reactor design was established that circumvents algal gradients in the reactor. The high density microalgal suspension underneath the foam-bed is pumped to the top of the reactor and allowed to drain back down through the foam-bed. This liquid recirculation ensures equal algae distribution in the reactor, and also provides better mixing in the foam phase. The significant mixing expected in the foam column underneath the liquid addition point is related to convective motion and internal circulation in the froth [10,11]. By distributing the microalgal broth on the top of the foam column, the additional liquid will drain down in the foam structure, thereby, enhancing the liquid fraction of the foams [12]. At higher liquid fractions, both coarsening and coalescence is reduced, therefore, also more stable foams are produced [13,14]. In foams without liquid addition, most of the liquid resides in the nodes and Plateau borders, while the liquid films are relatively thin (130–160 μm [15]). When considering mass transfer in foams, the thin films are prone to saturation [16,17]. With increasing liquid fraction the film thickness increases, which results in increased concentration gradient in the film and also film saturation is eliminated [18]. Together with better liquid advection [16], the additional liquid supply leads to increased mass transfer rates. Besides influencing mass transfer, the enhanced liquid fraction and algae concentration also alters the light profile in foams [19].

The formed foam has to be broken in order to refresh the gas phase in the entrapped bubbles, and ensure sufficient carbon dioxide (CO_2) for microalgal growth. The liquid jets originating from the liquid recirculation can serve as a foam breaking method [15,20]. More specifically, foam breakage originates from the impact of the liquid droplets falling on the foam [21]. Spraying cell culture on foam was already reported to efficiently break foam [20]. Since the necessity of liquid recirculation also offers the possibility to break the foam, the need for external foam breakers is avoided.

This study aims to reveal the potential of a novel liquid foam-bed photobioreactor design with internal circulation of the microalgae suspension over the foam-bed. Although liquid recirculation might be beneficial for a range of non-ionic surfactants, this study is focused on the application of Pluronic F68 due to its good foaming characteristics and its low toxicity and biodegradability [5]. Batch and continuous operation were carried out in order to test the system and determine reactor productivity and photosynthetic efficiency. Additionally, the mass transfer coefficient, mixing characteristics, and the light profile were characterized in this new photobioreactor design.

2. Materials and methods

2.1. The setup

The foam-bed photobioreactor consists of a glass column surrounded by circular lights. The column is 55 cm high, has a diameter of 10 cm from height 0 to 6 cm and 49 to 55 cm, and a diameter of 10.4 cm in between. The gas distributor plate is installed at 3 cm height from the bottom of the glass column. Between 44 and 50 cm height, the reactor has a narrowing; at 47 cm height the column reaches the narrowest point with a diameter of 4 cm. A water jacket used for temperature control surrounds the glass column. Foam was formed by a mixture of N_2 and CO_2 gas, supplied via mass flow controllers (Brooks instruments, The Netherlands). The gas entered the reactor via a distributor plate containing conical holes of 100 μm diameter at the bottom and 30 μm at the top, all holes were placed 3 mm from each other. The N_2 flow was humidified through a water bottle at 2 °C. After mixing the N_2 and CO_2 streams the gas dew point was analysed (E + E Elektronik dew point sensor). Finally, the gas was led over a filter (Whatman® PolyVENT 500, 0.2 μm) to sterilize the gas before entering to the reactor.

The algae suspension was recirculated from the bottom of the reactor to the top. The point of intake of the algae suspension was 2 mm above the gas distributor plate. It was pumped to the top of the foam

column by two peristaltic pumps. Both streams were further divided into two more streams. The total liquid volume in the tubes of the recirculation system was 20 mL. The resulting four streams were dispensed at the top of the reactor narrowing (5.5 cm below the lid) by four metal pipes with an inner diameter of 2 mm. The falling liquid contributed to foam breaking. However, when complete foam deconstruction did not take place at a sufficient rate, the foam level was still maintained at 5.4 cm below the top by a level sensor, controlling the inflow of the gas. More specifically, when the foam was in contact with the level sensor the gas inflow was automatically and temporarily stopped until the foam-bed dropped again below the level sensor. The gas was forced to leave the reactor through a condenser at 2 °C after which the gas dew point was measured and gas composition analysed (infrared analyser for CO_2 and a paramagnetic analyser for O_2 , Servomex, UK).

The temperature of the foam-bed was continuously monitored. A temperature probe was fitted inside a hollow metal tube filled with water, hanging in the foam bed 10 cm underneath the lid. Temperature regulation took place by an external cooling/heating unit recirculating water through the water jacket. The water temperature of this unit was adjusted manually to reach the desired temperature in the foam-bed. A pH probe was connected to a T-junction placed within the liquid recirculation line. In addition, a liquid sampling port was placed in the recirculation line. The foam-bed inside the cylindrical reactor was illuminated from all sides resulting in a homogenous light level over the reactor surface. More specifically, a circular light source was constructed, consisting of 8 vertical light panels placed around the column as an octagon. Each panel was composed of 24 warm-white LEDs (Avago ASMT-MY22-NMP00) equipped with a plastic lens with FWHM of 25.5° (Part no. 10393, Carclo-optics, UK).

The growth experiments were conducted in batch and chemostat modes. During chemostat mode media was pumped in and out by two peristaltic pumps. The inflow media bottle was continuously mixed with a magnetic stirrer to keep the surfactant dispersed and prevent its accumulation at the liquid surface. This media was sterilized by pumping through a 0.20 μm filter (Sartolab P-20 or Sartobran 300, Sartorius, Germany). Reactor weight, weight of the media bottle (inflow), and weight of the harvest bottle (outflow) were measured by analytic balances. These balances were linked to a data acquisition system interfaced via a computer by means of a LabView (National Instruments, USA) virtual instrument allowing for continuous data logging and process control. The reactor weight and, as such, liquid volume were maintained constant by continuously adjusting the inflow pump rate, while the outflow pump was running at a fixed speed. A picture and a schematic overview of the setup are presented in Fig. 1. A short video of the operation of the reactor is provided as Supplementary material (see Appendix F).

2.2. Reactor operation

To monitor microalgal growth in the reactor, two samples were taken daily and analysed for optical density, cell count and biovolume (cell volume per liquid volume), PSII quantum yield, and microscopic analysis. Microalgal dry weight (DW) concentration was also measured daily from the outflow bottle during continuous operation with *C. vulgaris*. After the measurements, the outflow bottle was emptied, and the measured DW represents an average over 24 h. For *C. sorokiniana*, the biovolume was translated to a dry weight value with a conversion factor of 0.5 g dry weight per millilitre cell volume [22]. For *C. vulgaris*, following the direct DW measurements from the outflow bottle, a calibration curve was made and a correlation factor of 0.516 g per millilitre was found, which was applied to convert biovolume to DW in the reactor samples.

The reactor was operated aseptically and was autoclaved only between operation with different strains. The volume at the bottom of the reactor directly above the sparger (3 cm) was covered with aluminium

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