

Photobioreactors

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Abstract

Photobioreactors are a new class of bioreactors in which microorganisms or vegetal cells use solar or artificial light for converting different substrates into the desired compounds. These equipments are of flat or tubular types and could be analyzed comparatively by means of specific criteria, photosynthesis efficiency and the yield of illumination being the most suggestive ones. In this paper we present the main characteristic of the most used photobioreactors.

Keywords: photobioreactors, microalgae, photobiosynthesis, kinetic model, light intensity.

Introduction

Photobioreactors are bioreactors in which photosynthesing microbial or vegetal cells (especially microalgae) are cultivated, in septic or aseptic conditions. These bioreactors are used for biomass or for bioactive compounds production (Table 1) [1].

Table 1. Compounds obtained by photobiotechnology

Products	Producer	Current or potential utilizations
Amphidinols and amphidins	<i>Amphidinium sp.</i>	Agents against tumors
Astaxanthin	<i>Haematococcus pluvialis</i> , <i>Chlorella sp.</i>	Pigment
β -Carotenes	<i>Dunaliella</i>	Dyes, additives for food
Docosahexenoic acid	<i>Isochrysis galbana</i>	Essential fatty acid
γ -Linolenic acid	<i>Spirulina sp.</i>	Essential fatty acid
Biomass, proteins	<i>Laminaria</i> (over 350,000 tonnes d.w./year), <i>Undaria</i> (over 13,000 tonnes d.w./year), <i>Porphyra</i> (over 340,000 tonnes d.w./year), <i>Eucheuma</i> (over 20,000 tonnes d.w./year)	Human or animal nutrition
Polyunsaturated fatty acids	<i>Phaeodactylum tricornutum</i> , <i>Isochrysis galbana</i>	Medicine, additives for food
Biohydrogen	<i>Rhodobacter sp.</i> , <i>Rhodopseudomonas sp.</i> , <i>Rhodospirillum sp.</i> , <i>Rhodovulum sp.</i>	Fuel
Fucoxantin	<i>Phaeodactylum tricornutum</i>	Antioxidant agent
Goniodomins	<i>Alexandrium hiraoui</i>	Antifungal agents
Oscillopeptins	<i>Oscillatoria agardhii</i>	Elastase inhibitor
Ficobiliproteins	Red algae, cyanobacteria	Dyes
Ficocyanine	<i>Spirulina platensis</i>	Dye

The design of photobioreactors has to be adapted to the requirements for biological functions of the cells and to the physical characteristics of the particular systems, as follows [2-5]:

- adequate space area exposed to light
- efficient feeding with carbon dioxide, as principal substrate for photosynthesis
- degassing system to remove the oxygen produced, because its accumulation induces the phenomena of product inhibition
- efficient temperature, pH and concentration control
- efficient harvesting systems.

The influence of the above mentioned factors are included in some particular mathematical models describing the microbial growth and biosynthesis, these models being useful for designing, optimizing and scaling-up the photobioreactors.

In this review, we present the most important kinetic models applied in photobiosynthesis and the constructive and operational characteristics of the most representative photobioreactors.

Kinetic Models

As stated by Lee and Low (1992) [6,7], Putz and Sheinbenbogen respectively (1998) [8], the productivity of the photobiosynthesing cultures is controlled by the illumination intensity. Generally, by amplifying the illumination the specific rate of biomass growth, μ , increases, reaches a maximum value, μ_{\max} , decreasing then owing to the photoinhibition phenomena [10,11].

Many kinetic models proposed for microbial growing in photobioreactors are derived from the ideal model proposed by Monod [1,12-18]:

- Tamiya et al. (1953)
$$\mu = \frac{\alpha \cdot \mu_{\max} \cdot I}{\mu_{\max} + \alpha \cdot I} \quad (1)$$

- Van Oorschot (1955)
$$\mu = \mu_{\max} \cdot \left(1 - e^{-\frac{I}{I_{\max}}} \right) \quad (2)$$

- Steele (1977)
$$\mu = \frac{\mu_{\max} \cdot I}{\mu_{\max}} \cdot e^{-\frac{I}{I_{\max}}} \quad (3)$$

- Molina et al. (1994)
$$\mu = \frac{\mu_{\max} \cdot I^{\beta}}{A^{\beta} + I^{\beta}} \quad (4)$$

- Molina et al. (2001)
$$\mu = \frac{\mu_{\max} \cdot I_m^{\beta}}{I_k^{\beta} + I_m^{\beta}} \quad (5)$$

where: α , β - constants specific to the system

A - cells affinity for light, $\mu\text{E}/\text{m}^2\text{s}$

I - light intensity on the culture surface, $\mu\text{E}/\text{m}^2\text{s}$

I_{\max} - saturation value of light intensity, $\mu\text{E}/\text{m}^2\text{s}$

I_m - average light intensity inside the culture, $\mu\text{E}/\text{m}^2\text{s}$

I_k - constant depending on the cultivated microorganisms and photobiosynthesis conditions.

Photoinhibition, however, exhibits an important negative influence on the photobiosynthesis processes, and for outdoor photobioreactors it appears in the period of the day with maximum solar light intensity. As it can be seen from Figure 1, the photoinhibition induces the reduction of photosynthetic activity of the microorganisms for about four hours after the moment with maximum illumination intensity (in the noon) [19].

Photosynthetic activity

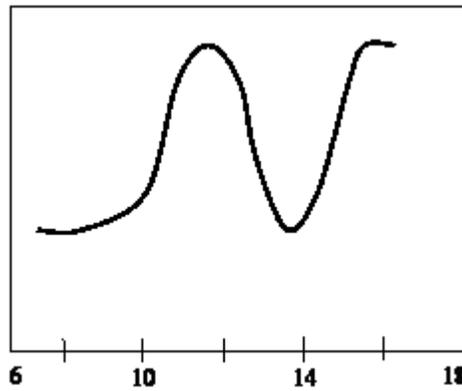


Figure 1. Variation of the microalgal photosynthetic activity during one summer day.

The effect of light intensity has been tested for the photobiosynthesis of some pigments by *Haematococcus pluvialis* in a tubular photobioreactor of air-lift type. The experiments carried out by Garcia-Malea et al. (2006) indicated that the effect of the illumination must be correlated with the dilution rate, because the increasing of the dilution rate could diminish or avoid the phenomena of photoinhibition (Figure 2) [11].

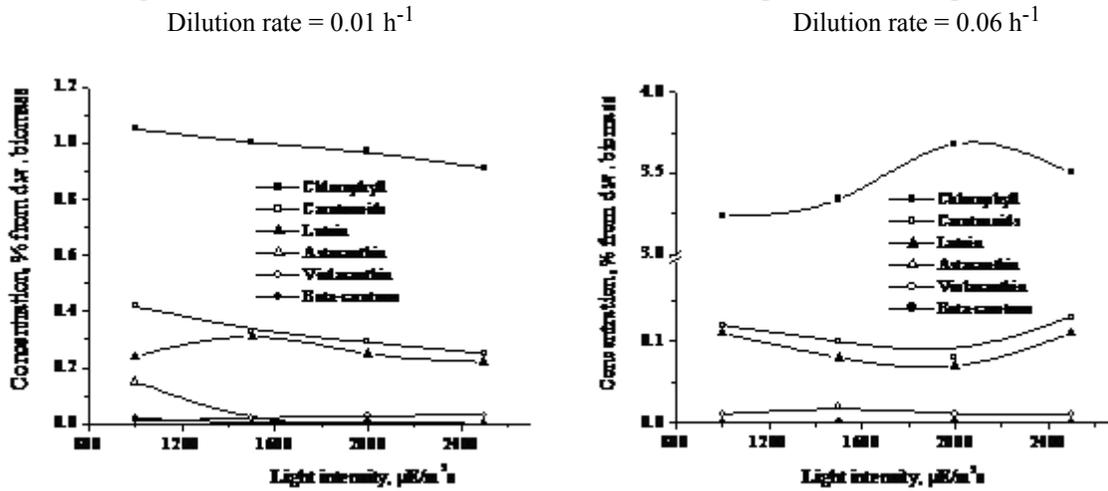


Figure 2. Influence of the illumination intensity on some pigments photobiosynthesis.

Because the correlations (1) - (5) did not take into account the photoinhibition which inherently appears at higher light intensity, the above equations have been modified for respecting the real photobiosynthesis processes. The expressions of the new models either are similar with the classical kinetic models for the biosynthesis with inhibition, or are particular ones [9,19-26]:

- Bannister (1979)

$$\mu = \frac{\mu_{max} \cdot I}{\left(K_i^\gamma + I^\gamma\right)^\gamma} \tag{6}$$

- Aiba (1982)

$$\mu = \frac{\mu_{max} \cdot I}{K_S + I + \frac{I^2}{K_i}} \tag{7}$$

- Ancien et al. (1998)

$$\mu = \frac{\mu_{max} \cdot I_m^{\alpha + \frac{\beta}{I_0}}}{\left\{A \cdot \left[1 + \left(\frac{I}{K_i}\right)^\gamma\right]\right\}^{\alpha + \frac{\beta}{I_0}} + I_m} \tag{8}$$

where: γ - constant specific to the system

K_i - photoinhibition constant, $\mu E/m^2s$

K_S - saturation constant, $\mu\text{E}/\text{m}^2\text{s}$

I_0 - light intensity perpendicular to the culture surface, $\mu\text{E}/\text{m}^2\text{s}$.

The equation (8) has been verified for the cultures of *Phaeodactylum tricornutum*, the obtained values of the kinetic parameters being as follows: $\mu_{\max} = 0.063 \text{ h}^{-1}$, $A = 94.3 \mu\text{E}/\text{m}^2\text{s}$, $K_i = 3426 \mu\text{E}/\text{m}^2\text{s}$, $\alpha = 3.04$, $\beta = 1.209$, $\gamma = 514.6$ [19].

Photobioreactors

Although photobioreactors configurations are various, these equipments can be included in two main classes [1]:

- a. flat photobioreactors
- b. tubular bioreactors

each of these categories being subdivided in function of the configuration of the constructive elements, broth circulation, light supply, position etc.

Some of the commercial photobioreactors for microalgae cultures are given in Table 2 [1,18].

Table 2. Commercial photobioreactors for microalgae cultures

Photobioreactor	Algae	Volume , m ³	Country
Reservoir	Various	10	Different countries
Pond	<i>Dunaliella salina</i>	10 ⁶	Australia
Pond with rotative arm	<i>Chlorella sp.</i>	15	Taiwan, Japan
With channels	<i>Chlorella sp.</i> , <i>Spirulina sp.</i> , <i>Dunaliella salina</i>		Japan, Taiwan, USA, Thailand, China, India, Israel, Vietnam, Chile, Peru, Russia
Cascade with baffles	<i>Chlorella sp.</i>	30	Czechia, Bulgaria
Classical bioreactor	<i>Chlorella sp.</i> , <i>Cryptocodinium cohnii</i>	> 1	Japan, Indonesia, USA
Combined: closed bioreactor and pond with mixing	<i>Haematococcus pluvialis</i>	20	USA

The open flat bioreactors, with free or forced media circulation, are the most used, but their characteristics allow only few microorganisms to be cultivated, especially due to the difficulty to perform aseptic fermentations (the control of the sterility is mainly made by using high alkaline or saline media).

The **pond photobioreactors** (Figure 2) gained considerable popularity, due to their cheap construction, simple maintenance and operation [1]. But, they have several drawbacks, namely as the biomass deposition, low yields, unstable microalgal populations, difficulty to reach the uniform distribution of nutrients.

The main sizes of these bioreactors are: length 16 - 20 m, width 8 - 10 m. Some of the ponds from Australia have the surface over 250 hectares and are placed inside of the natural lakes.

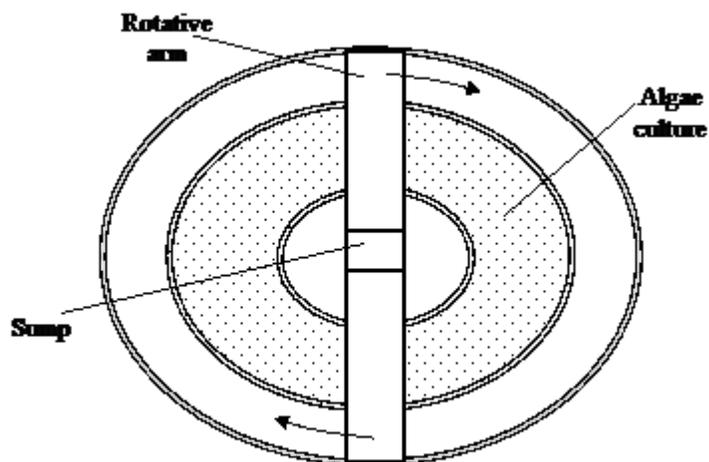


Figure 3. Pond photobioreactor with mixing.

This photobioreactor is used for *Dunaliella salina* or diatoms cultures in enriched marine water.

In the **deep channeled photobioreactors** the culture is slowly circulated through channels configured as a recirculating loop (Figure 4) [1,27]. They can be used for aseptic cultures of bacteria or microalgae (*Spirulina sp.*, *Chlorella sp.*), as well as for wastewater treatment.

These bioreactors are more productive than the pond ones, but also more expensive. In function of the used technology, the costs for 1 kg of d.w. biomass vary between 50 and 300 - 600 \$, comparatively with 9 - 25\$ in the case of pond photobioreactors.

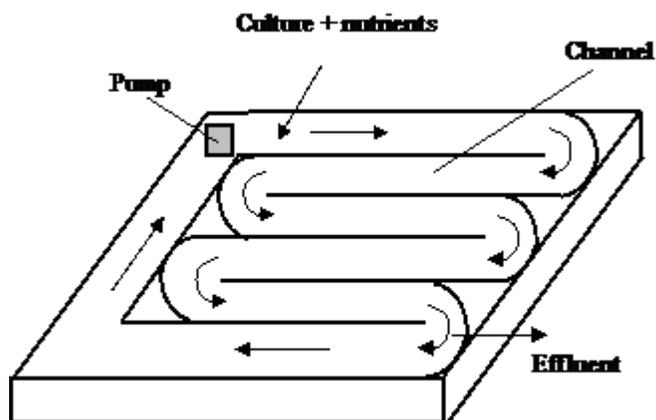


Figure 4. Deep channeled photobioreactor.

Photobioreactors with shallow circulation are provided with an intense mixing [1,28]. In these bioreactors, the algal cultures and the air are completely enclosed in a transparent tube of polyethylene and circulated with a pump (Figures 5 and 6). The tube diameter is of 5 - 10 cm, the culture depth of 1 cm and the broth velocity of 7.5 cm/s, thus avoiding the biomass deposition.

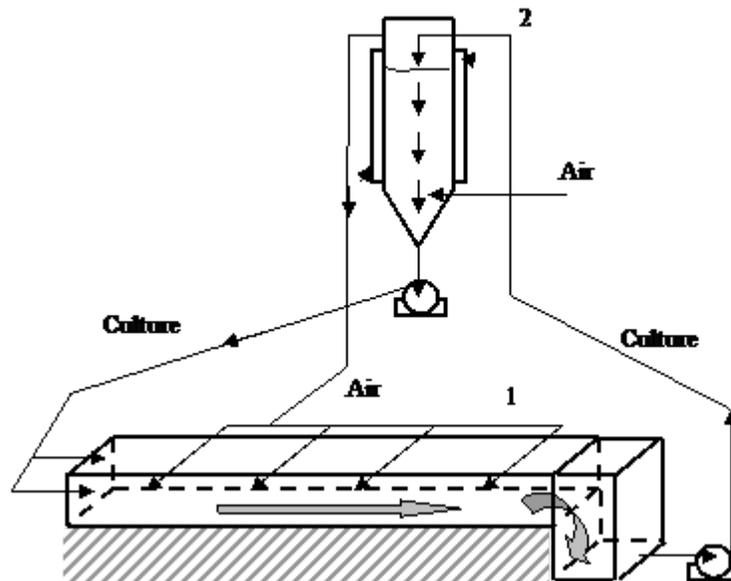


Figure 5. Photobioreactor with shallow-cascade circulation
(1 - closed tubular module, 2 - tower for aeration and heat exchange).

In the cascade photobioreactor, high growth rate has been obtained for *Chlorella sp.*, the culture depth being lower than 1 cm. In Australia, the productivity of this system reached 25 g d.w. biomass/m² day, the overall circulation surface of the bioreactor being of 0.5 hectares [1,29].

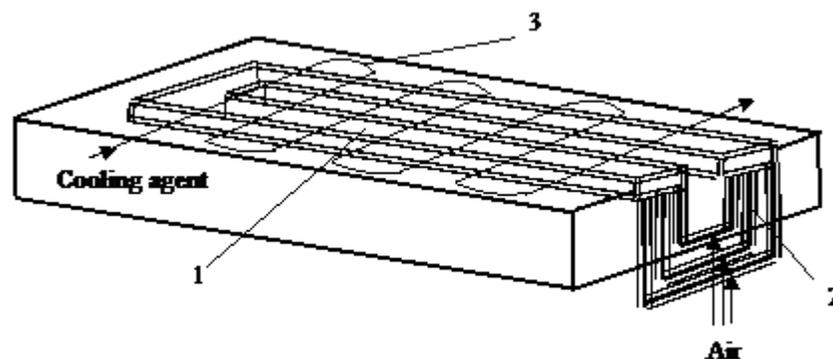


Figure 6. Photobioreactor with shallow circulation
(1 - channel, 2 - recirculation system, 3 - heat exchanger).

All the above presented photobioreactors are placed outdoor, especially due to the solar light requirements. Although the outdoor photobiosynthesing systems have been applied for many years and in various cultivating methods, an economically feasible production of microalgae has not been established yet. One of the reasons is the demand of microalgal cultures in light energy and, consequently, the requirement of large surface areas. On the other hand, large surface areas are expensive and cannot be effectively controlled from the viewpoint of all the environmental parameters affecting the microalgal growth. These requirements combined with the seasonal change in the climatic conditions limit the productivity of the outdoor photobioreactors.

Another limitation of the outdoor systems is the low stability of the medium consistency and of the final product quality, due to the high rates of water evaporation and to the inclusion of foreign materials (dust, birds, insects etc.).

Therefore, various types of photobioreactors have been designed for indoor cultivations of microorganisms, in closed systems. The process can be more efficiently controlled, but the lower space available for photosynthesis constitutes an important limitation of these bioreactors.

The **helicoidal photobioreactor**, given in Figure 7, has been designed for maximum control of the growth conditions and ability to scaling-up [1,30,31]. The helicoidal part is made from 200 m of polyethylene or glass tubes, in four sections, each having 50 m length. The circulation velocity of the broth is maintained at a level which allows low stationary times into the dark regions of the tubes, of maximum 5 minutes. In this bioreactor can be cultivated microorganisms resistant to the shear forces and which do not adhere to the tubes wall.

The productivity reached in the helicoidal photobioreactor is for several times greater than that corresponding to the outdoor photobioreactors.

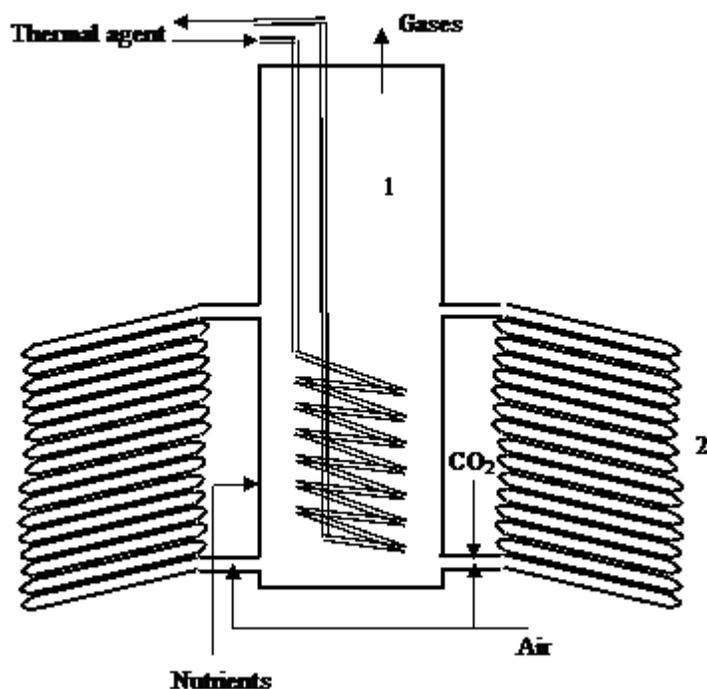


Figure 7. Helicoidal photobioreactor (1 - column, 2 - helicoidal loop).

The **semi-spherical photobioreactors**, designed in 2006 in Japan, work as the gas-lift bioreactors, the mixing being achieved by supplying air or air and carbon dioxide through six orifices, each placed at 60° (Figure 8) [32]. For supplementary mixing, the bioreactors are provided with a blade attached to the sparger, its movement being induced by the gas jet.

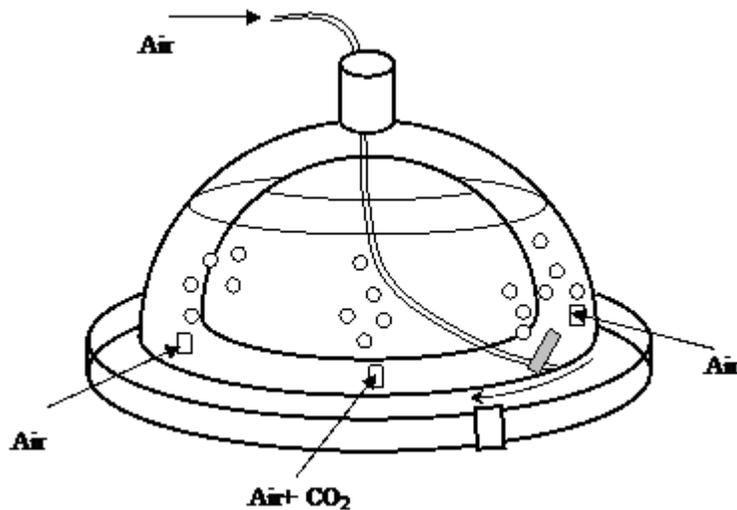


Figure 8. Semi-spherical photobioreactor.

The ellipsoidal shape avoids the biomass deposition on the wall, the biomass adhesion leading to the reducing of the depth of light penetration.

Similar photobioreactors are those with cylindrical and parabolic shapes, used in Japan for the microalgae *Chlorococum littorale* and *Chaetoceros calcitrans* cultivation in all seasons. Among them, the most efficient is the cylindrical photobioreactor, its productivity for 1 m² being for about 1.8 times greater than that of the others similar photobioreactors (Table 3) [32-34].

Table 3. Growth rates of microalga *Chlorococum littorale* in different closed flat photobioreactors

Photobioreactor	Growth rate		
	<i>g d.w./day</i>	<i>g d.w./l day</i>	<i>g d.w./m² day</i>
Parabolic	6.05	0.086	14.94
Cylindrical	10.25	0.146	20.5
Semi-spherical	12.38	0.095	10.95

The **multitubular photobioreactor**, also called “*medusa photobioreactor*”, due to its configuration, is designed to intersperse the tubular elements with the light sources (Figure 9) [35]. The broth circulation is promoted by the air and carbon dioxide supplying through one of the tubular element.

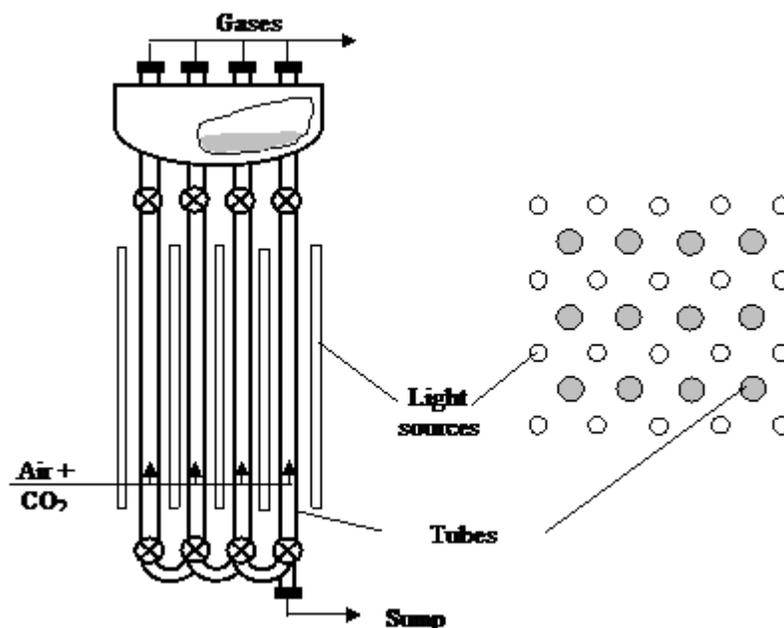


Figure 9. “Medusa” photobioreactor.

The medusa photobioreactor allow to using small spaces with high volumes (100 l occupies 1 m² ground).

Walter et al. (2003) succeeded to scaling-up this photobioreactor at pilot and industrial level by means of the light intensity criteria, for microalga *Chlorella salina* and bacteria *Lyngbya taylorii* [35]. For this purpose, the volumetric coefficient of irradiation, I_V , has been used:

$$I_V = I_0 \frac{A}{\varepsilon \cdot d_t \cdot C_X}, \mu\text{E}/\text{m}^3\text{s} \quad (9)$$

where: A - specific illuminated surface, m²/m³

ε - light extinction coefficient of the broth, m²/kg

d_t - tubes diameter, m

C_X - biomass concentration, kg d.w./m³.

Therefore, in Table 4 are given the results obtained for three operation levels by maintaining constant the value of I_V [35,36].

Table 4. Comparative data obtained for different operational scales of “Medusa” photobioreactor (cultures of *Chlorella salina*, $I_V = 3 \times 10^4 \mu\text{E}/\text{m}^3\text{s}$)

Volume, l	Specific rate of growth, h ⁻¹	Maximum biomass concentration, g d.w./l	Productivity, mg d.w./l.h
0.5	0.063	3.9	17.9
10	0.056	4.3	21.3
100	0.052	4.5	33.8

These data indicated the similitude between the analyzed parameters, only the productivity being significantly higher for 100 l working volume.

The comparative analysis of the performances of different types of photobioreactors is made taking into account the efficiency of light supply, the facility of fermentation control, the possibility to cultivate only a single microorganism in aseptic conditions etc. The qualitative comparison between the photobioreactors is exemplified in Table 5 [1,37-46].

Table 5. Qualitative comparison of photobioreactors performances

Photobioreactor	Mixing	Efficiency of light utilization	Efficiency of gas-liquid mass transfer	Effect of shear forces	Sterility
Pond	Very low	Low	Low	Very low	Septic cultures
Reservoir	Low	Very low	Low	Very low	Septic cultures
Pond with rotative arm	Good	Good	Good	Low	Septic cultures
Pond with channels and mixing	Good	Good	Low	Low	Septic cultures
Stirred bioreactor with illumination	Non-uniform	Good	Rather good	High	Aseptic cultures
Air-lift	Uniform	Good	Very good	Low	Aseptic cultures
Tubular	Uniform	Excellent	Good	Rather high	Possible aseptic cultures

Helicoidal	Uniform	Excellent	Good	Rather high	Possible aseptic cultures
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Moreover, according with Akkerman et al. (2002), the comparative analysis can be made also by means of two specific parameters: the photosynthesis efficiency, FE, defined as the proportion of the absorbed light intensity which is used for biomass growing or product biosynthesing from the overall light intensity absorbed into the media, respectively the yield of illumination, $Y_{B/E}$ (amount of biomass or product/unit of absorbed light energy) (Table 6) [47-49].

Table 6. Comparative analysis of some closed photobioreactors used for biohydrogen production

Photobioreactor	Microorganism	FE (%), $Y_{B/E}$
Column and Air-lift	<i>Phaeodactylum tricornutum</i>	$Y_{B/E} = 0.82 - 0.84$
Flat: - vertical - inclined	<i>Spirulina platensis</i>	FE = 16%, $Y_{B/E} = 1.48$ FE = 10 - 20
Tubular: - diameter 2.5 cm - diameter 5.3 cm	<i>Phaeodactylum tricornutum</i> , <i>Spirulina platensis</i>	$Y_{B/E} = 0.48 - 0.63$ $Y_{B/E} = 0.68 - 0.95$

The values given in Table 6 indicate that the most efficient photobioreactors are the flat ones, even if the biomass concentration is high and, consequently, the penetration depth of the light is reduced. The tubular photobioreactors should theoretically offer higher photobiosynthesis yields, owing to the shorter duration of the light-dark cycles inside the broths, but the above results do not support this assumption (probably due to the most efficient illumination and oxygen removal in the case of the flat photobioreactors).

Conclusions

The progress of the biotechnology implicitly leads to the design of new bioreactors which use the solar or artificial light, namely the photobioreactors. These equipments are of special construction of flat or tubular types. The comparison between the performances of these bioreactors can be made using different specific criterias, among them the photosynthesis efficiency and the yield of illumination being the most suggestive.

Therefore, using the photobioreactors and the renewable sources, the „solar biotechnology” is going to give a cleaner environment and improved economic balance for the production of food, fuel and chemicals [1,50,51].

References

1. CAȘCAVAL D., ONISCU C., GALACTION A.I., *Inginerie biochimica si biotehnologie. 2.Bioreactoare*, InterGlobal, Iasi, 2002.
2. ASADA Y., MIYAKE J., *J. Biosci. Bioeng.*, **88**, 16 (1999).
3. LEE C.G., *Biotechnol. Bioprocess Eng.*, **4**, 78 (1999).
4. RICHMOND A., CHENG-WU Z., *J. Biotechnol.*, **85**, 259 (2001).
5. YEH M.S., WEI Y.H., CHANG J.S., *Biotechnol. Progr.*, **21**, 1329 (2005).
6. LEE H.S., SEO M.W., KIM Z.H., LEE C.G., *Enz. Microb. Technol.*, **39**, 447 (2006).
7. LEE K.S., LIN P.J., CHANG J.S., *Int. J. Hydrogen Energy*, **31**, 465 (2006).
8. RUBIO F., ACIEN F.G., SANCHEZ J.A., CAMACHO F., MOLINA E., *Biotechnol. Bioeng.*, **62**, 71 (1999).
9. ACIEN F.G., FERNANDEZ J.M., SANCHEZ J.A., MOLINA E., CHISTI Y., *Chem. Eng. Sci.*, **56**, 2721 (2001).
10. AKKERMAN I., JANSSEN M., ROCHA J., WIJFFELS R.H., *Int. J. Hydrogen Energy*, **27**, 1195 (2002).
11. GARCIA-MALEA M.C., ACIEN F.G., FERNANDEZ J.M., CERON M.C., MOLINA E., *Enz. Microb. Technol.*, **38**, 981 (2006).

12. MOLINA E., CAMACHO F., ACIEN F.G., GOMEZ A., *J. Chem. Technol. Biotechnol.*, **61**, 167 (1994).
13. MOLINA E., ACIEN F.G., CAMACHO F., CHISTI Y., *J. Biotechnol.*, **70**, 231 (1999).
14. DE PHILIPPIS R., SILI C., PAPERI R., VINCENZINI M., *J. Appl. Phycol.*, **13**, 293 (2001).
15. BARBOSA M.J., ROCHA J., TRAMPER J., WIJFFELS R.H., *J. Biotechnol.*, **85**, 25 (2001).
16. BABCOCK R.W., MALDA J., RADWAY J.C., *J. Appl. Phycol.*, **14**, 169 (2002).
17. MOLINA E., ACIEN F.G., CAMACHO F., CHISTI Y., *J. Appl. Phycol.*, **12**, 355 (2001).
18. MOLINA E., FERNANDEZ J., ACIEN F.G., CHISTI Y., *J. Biotechnol.*, **92**, 113 (2001).
19. DEL RIO E., ACIEN F.G., GARCIA-MALEA M.C., RIVAS J., MOLINA E., *Biotechnol. Bioeng.*, **91**, 808 (2005).
20. MIRON A.S., CAMACHO F.G., GOMEZ A.C., GRIMA E.M., CHISTI Y., *A.I.Ch.E.J.*, **46**, 1872 (2000).
21. PARK K.H., LEE C.G., *Biotechnol. Bioprocess Eng.*, **6**, 189 (2001).
22. PULZ. O., *J. Appl. Microbiol. Biotechnol.*, **57**, 287 (2001).
23. CARLOZZI P., *Biotechnol. Bioeng.*, **81**, 305 (2003).
24. GUERIN M., HUNTLEY M.E., OLAIZOLA M., *Trends Biotechnol.*, **21**, 210 (2003).
25. JEON Y.C., CHO C.W., YUN Y.S., *Biochem. Eng. J.* **27**, 127 (2005).
26. KIM Z.H., KIM S.H., LEE H.S., LEE C.G., *Enz. Microb. Technol.*, **39**, 414 (2006).
27. HOEKEMA S., BIJMANS M., JANSSEN M., TRAMPER J., WIJFFELS R.H., *Int. J. Hydrogen Energy*, **27**, 1331 (2002).
28. LORENZ R.T., CYSEWSCHI G.R., *Trends Biotechnol.*, **18**, 160 (2000).
29. DEGEN J., UEBELE A., RETZE A., SCHMID-STAIGER U., TROSCHE W., *J. Biotechnol.*, **92**, 89 (2001).
30. FABREGAS J., OTERO A., MASEDA A., DOMINGUEZ A., *J. Biotechnol.*, **89**, 65 (2001).
31. FABREGAS J., DOMINGUEZ A., MASEDA A., OTERO A., *Appl. Microbiol. Biotechnol.*, **61**, 545 (2003).
32. SATO T., USUI S., TSUCHIYA Y., KONDO Y., *Energy Convers. Manag.*, **47**, 791 (2006).
33. SUH I.S., JOO H.N., LEE C.G., *J. Biotechnol.*, **125**, 540 (2006).
34. HE D., BULTEL Y., MAGNIN J.P., WILLISON J.C., *Enz. Microb. Technol.*, **38**, 253 (2006).
35. WALTER C., STEINAU T., GERBSCH N., BUCHHOLZ R., *Biomolec. Eng.*, **20**, 261 (2003).
36. WY S.Y., HUNG C.H., LIN C.N., CHEN H.W., LEE A.S., CHANG J.S., *Biotechnol. Bioeng.*, **93**, 934 (2006).
37. ZHANG D.H., LEE Y.K., *Appl. Microbiol. Biotechnol.*, **55**, 537 (2001).
38. WU X., MERCHUK J.C., *Chem. Eng. Sci.*, **56**, 3527 (2001).
39. HALLENBECK P.C., BENEMANN J.R., *Int. J. Hydrogen Energy*, **27**, 1185 (2002).
40. ZOU N., ZHOU B., LI B., SUN D., ZENG C., *Biomolec. Eng.*, **20**, 281 (2003).
41. CHOI L.S., SUH I.S., LEE C.G., *Enz. Microb. Technol.*, **33**, 403 (2003).
42. HALL D.O., FERNANDEZ F.G.A., GUERRERO E.C., RAO K.K., MOLINA E., *Biotechnol. Bioeng.*, **82**, 62 (2003).
43. LIN C.Y., LAY C.H., *Int. J. Hydrogen Energy*, **29**, 275 (2004).
44. LABABPOUR A., HADA K., SHIMAHARA K., KATSUDA T., KATOH S., *J. Biosci. Bioeng.*, **98**, 452 (2004).
45. OROSA M., FRANQUEIRA D., CID A., ABALDE J., *Biores. Technol.*, **96**, 373 (2005).
46. CHEN C.Y., CHANG J.S., *Process Biochem.*, **41**, 2041 (2006).
47. HARRIGAN C.C., GOETZ G., *J. Appl. Phycol.*, **14**, 103 (2002).
48. KOBAYASHI M., *Biotechnol. Bioprocess Eng.*, **8**, 322 (2003).
49. OLAIZOLA M., *Biomolec. Eng.*, **20**, 459 (2003).
50. ZOU N., RICHMOND A., *J. Biotechnol.*, **70**, 351 (1999).
51. GALACTION A.I., CAȘCAVAL D., *Metaboliti secundari cu aplicatii farmaceutice, cosmetice si alimentare*, Venus, Iasi, 2006.