



(51) International Patent Classification:  
C12C 1/15 (2006.01)

(21) International Application Number:  
PCT/US2010/036178

(22) International Filing Date:  
26 May 2010 (26.05.2010)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:  
61/181,790 28 May 2009 (28.05.2009) US  
61/233,549 13 August 2009 (13.08.2009) US

(71) Applicant (for all designated States except US):  
COASTAL BIOMARINE, LLC [US/US]; P.O. Box 6,  
Bridgewater, CT 06752 (US).

(72) Inventor; and

(75) Inventor/Applicant (for US only): WILKINSON, Loy  
[US/US]; 250 Northrup Street, Bridgewater, CT  
06752-1610 (US).

(74) Agent: GREELEY, Paul, D.; Ohlandt, Greeley, Rug-  
giero & Perle, L.L.P., One Landmark Square, 10th Floor,  
Stamford, CT 06901-2682 (US).

(81) Designated States (unless otherwise indicated, for every  
kind of national protection available): AE, AG, AL, AM,  
AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ,  
CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO,  
DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT,  
HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP,  
KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD,  
ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI,  
NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD,  
SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR,  
TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

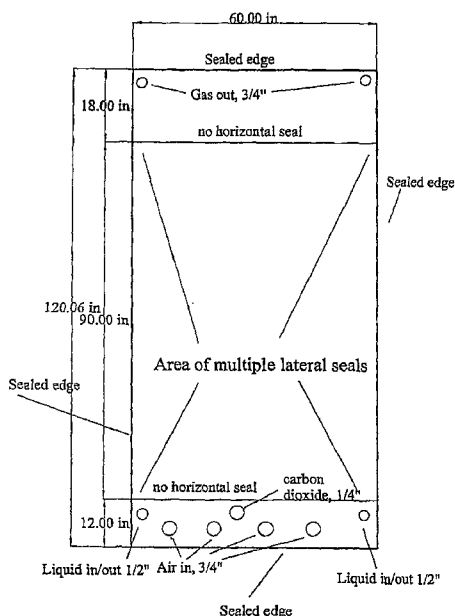
(84) Designated States (unless otherwise indicated, for every  
kind of regional protection available): ARIPO (BW, GH,  
GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG,  
ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ,  
TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK,  
EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU,  
LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK,  
SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ,  
GW, ML, MR, NE, SN, TD, TG).

[Continued on next page]

(54) Title: PHOTOBIOREACTOR AND METHOD FOR CULTURING AND HARVESTING MICROORGANISMS

FIG. 1

Front Layer - Culture Chamber



(57) Abstract: This disclosure relates to a photobioreactor and methods for culturing and harvesting a microorganism culture and biomass. The photobioreactor comprises a container having at least a front wall, a rear wall and an interior volume. At least a portion of the front wall and the rear wall is transparent to light. The container has at least a top portion, a bottom portion, and a plurality of parallel passageways extending from the bottom portion to the top portion for the throughflow of a liquid medium. The container has one or more liquid medium inlet openings at the bottom portion for introducing at least liquid medium into the container; one or more product outlet openings at the bottom portion for removing at least product from the container; one or more gas inlet openings at the bottom portion for introducing at least carbon dioxide gas and optionally other gases into the container; and one or more gas outlet openings at the top portion for removing at least one of excess gas and waste gases from the container. A heat exchanger system can be used for controlling the temperature of the photobioreactor. The gas supply source can be sequestered carbon dioxide. The microorganism culture can be used in biofuel production, aquaculture food production, mammalian food production, and recombinant protein synthesis.

WO 2010/138571 A1



---

**Declarations under Rule 4.17:**

— *of inventorship (Rule 4.17(iv))*

**Published:**

— *with international search report (Art. 21(3))*

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments (Rule 48.2(h))*

PHOTOBIOREACTOR AND METHOD FOR CULTURING  
AND HARVESTING MICROORGANISMS

Field of the Disclosure

**[0001]** This disclosure relates to a photobioreactor for the cultivation and harvesting of photosynthetic organisms and biomass. The photobioreactor of this disclosure allows for cost-effective cultivation of microorganisms, e.g., microalgae, on a large scale.

Background of the Disclosure

**[0002]** Cultivation of microorganisms, particularly algae, has been utilized for creating nutritional supplements, fertilizers, food additives, energy products such as biofuels, and other applications. Methods of such cultivation include, for example, open pond systems and closed system bioreactors.

**[0003]** The open pond systems can provide a low cost growing environment but suffer from a number of drawbacks. Such drawbacks include, for example, evaporation, lack of temperature control, contamination by undesired organisms, slow or no response to control measures, and the like. These limitations can reduce productivity in open pond systems making them not cost effective.

**[0004]** Closed system photobioreactors typically can be used for commercial cultivation of microorganisms. Closed system photobioreactors have several advantages over the open pond systems, for example, better control of the microorganism culture, better regulation of gas transfer, better control of light intensity, reduction of evaporation of the liquid medium, more uniform temperature control, and better protection from contamination. However, while closed system photobioreactors overcome many of the problems associated with open pond systems, the closed system photobioreactors are commercially not cost effective and have high capital costs.

**[0005]** For example, photobioreactors used to cultivate microorganisms on an industrial scale include glass and stainless steel photobioreactors having

expensive materials of construction. These industrial photobioreactors also have expensive and complicated parts and technologies. Operating costs associated with industrial glass and stainless steel photobioreactors can be high due to low productivity yields and required special operating techniques such as sterilization. Microorganism cultures grown in these commercial photobioreactors, and products derived therefrom, are expensive, and competing commercially in the marketplace is difficult.

**[0006]** There is a need for industrial scale photobioreactors that can be constructed and operated economically, e.g., low capital investment, and at high productivity. In addition, there is a need for commercial cultivation of microorganisms, e.g., microalgae, that is economically practical and cost effective for competing in the marketplace, and that can be conducted at high productivity. It would therefore be desirable in the art to provide such a photobioreactor and cultivation method that possesses some, or preferably all, of the above characteristics.

#### Summary of the Disclosure

**[0007]** This disclosure relates in part to a photobioreactor comprising:

- a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;
- said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;
- one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;
- one or more product outlet openings at said bottom portion for removing at least product from said container;
- one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container.

**[0008]** This disclosure also relates in part to a photobioreactor for culturing and harvesting at least one of a microorganism culture, said photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more microorganism culture outlet openings at said bottom portion for removing microorganism culture from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container.

**[0009]** This disclosure further relates in part to a method for culturing and harvesting microorganisms, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more microorganism culture outlet openings at said bottom portion for removing microorganism culture from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings; and

h) harvesting a microorganism culture from said container via said one or more microorganism culture outlet openings.

**[00010]** This disclosure yet further relates in part to a method of producing biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the production of said biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings; and

h) removing said biomass from said container via said one or more biomass outlet openings.

**[00011]** This disclosure also relates in part to a method for producing a biofuel from biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;

h) removing said biomass from said container via said one or more biomass outlet openings; and

i) converting said biomass to said biofuel.

**[00012]** This disclosure further relates in part to a method for producing and recovering a recombinant protein from biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;



one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;

h) removing said biomass from said container via said one or more biomass outlet openings; and

i) producing and recovering said recombinant protein from said biomass.

**[00013]** This disclosure yet further relates in part to a method for producing aquaculture food from biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;

h) removing said biomass from said container via said one or more biomass outlet openings; and

i) converting said biomass to said aquaculture food.

**[00014]** This disclosure also relates in part to a method for producing mammalian food from biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;

h) removing said biomass from said container via said one or more biomass outlet openings; and

i) converting said biomass to said mammalian food.

**[00015]** The photobioreactor and methods of this disclosure allow for cost-effective cultivation of microalgae on a large scale. The photobioreactor and methods have several features, for example, the use of inexpensive materials of construction, low capital investment, a form which maximizes utilization of incident sunlight, modularity that affords efficient swapping of units, and high biomass productivity. The photobioreactor is designed to provide favorable use of energy inputs (for example, light) for growing microalgae and provide good control over operating parameters including uniform internal turbulence. The closed system photobioreactor of this disclosure can also limit problems associated with open pond systems such as evaporation of water, allow for recycling of used water and medium, and reduce or eliminate chances of introducing foreign organisms or contamination.

**[00016]** Other goals and advantages of this disclosure will be further appreciated and understood when considered in conjunction with the following description and accompanying drawings. While the following description may contain specific details describing particular embodiments of the disclosure, this should not be construed as limitations to the scope of the disclosure but rather as an exemplification of preferable embodiments. For each aspect of this disclosure, many variations are possible as suggested herein. A variety of changes and modification can be made within the scope of the disclosure without departing from the spirit thereof.

#### Brief Description of the Drawings

**[00017]** Fig. 1 is a schematic front view representation of a culture chamber portion of an illustrative photobioreactor of this disclosure showing illustrative dimensions.

**[00018]** Fig. 2 is a schematic back view representation of a heat exchanger portion of an illustrative photobioreactor of this disclosure showing an enlarged portion of parallel passageways and also showing illustrative dimensions.

**[00019]** Fig. 3 is a schematic front view representation of a culture chamber portion of an illustrative photobioreactor of this disclosure showing an enlarged

portion of a gas sparger (segment) positioned in the parallel passageways and also showing illustrative dimensions.

**[00020]** Fig. 4 is a schematic front view representation of a culture chamber portion of an illustrative photobioreactor of this disclosure showing illustrative internal flow patterns.

**[00021]** Fig. 5 is a graphical representation showing that a riser with an inflated diameter of 0.75 inch requires a gas flow in the range of .015 cubic feet per minute (cfm) to 0.25 cubic feet per minute to achieve a range of turbulent flows in the liquid phase equivalent to a Reynolds number in the range of 1800 to 3000.

**[00022]** Fig. 6 depicts an illustrative sparger assembly useful in a photobioreactor of this disclosure.

#### Detailed Description of the Disclosure

**[00023]** The photobioreactor of this disclosure possesses features that are essential for commercial viability, particularly for applications that involve the production of commodities such as fuel and food. Those features include among others a low construction cost and a high productivity of biomass for solar radiation that impinges on the surfaces of the photobioreactor. The low cost photobioreactor of this disclosure is achieved by the use of components fabricated from low cost materials (for example, plastics) and produced in mass production operations such as extrusions. The designed components can also be assembled into the photobioreactor by simple techniques. High productivity is achieved by employing turbulent flow in the parallel passageways.

**[00024]** As indicated above, this disclosure relates in part to a photobioreactor. As also indicated above, this disclosure relates in part to a photobioreactor, a photobioreactor for culturing and harvesting at least one of a microorganism culture, and a method for culturing and harvesting microorganisms.

**[00025]** The photobioreactor can be made of inexpensive materials and is preferably made of plastic. Illustrative plastic materials include polyethylene,

polypropylene, polycarbonate, polyvinylchloride, polyester and the like. Different types of plastic may be used for their desirable properties on the different photobioreactor components or be laminated to each other or other suitable materials (for example, polyamides) to combine their properties. For example, rigid polyvinylchloride may be used for components requiring stiffness and exceptional strength while plasticized polyvinylchloride may be used for components requiring flexibility and light transmittance, or polyethylene and nylon may be laminated to join polyethylene's vapor barrier with nylon's strength. The plastic may be treated for resistance to ultraviolet radiation to prevent degradation of the plastic and to decrease photoinhibition of the algal growth. The photobioreactor may be made of materials that support the cost effective production of low value products (such as fuel) as well as high value products (such as cosmetics, nutritional supplements, therapeutics or enzymes). The photobioreactor or components of the photobioreactor can be made of recycled materials (for example, recycled plastics).

**[00026]** In an embodiment, the container can be formed by fusion bonding two or more suitable sheets of plastic material along predetermined seams. The plastic sheets can be cut in an approximately elongated rectangular shape and superposed one over the other. The plastic sheets can then be fusion bonded together by conventional methods to form seams along the peripheries and also seams making up the parallel passageways. Prior to fusion bonding the plastic sheets together, rigid plastic face fittings, e.g., bosses, can be fusion bonded at locations corresponding to the one or more liquid medium inlet openings, the one or more microorganism culture outlet openings, the one or more gas inlet openings, the one or more air inlet openings, and the one or more gas outlet openings. These face fittings can provide suitable mechanical attachment points for each of the corresponding input and output lines.

**[00027]** The container can be formed by fusion bonding two sheets of plastic material along predetermined seams to provide at least an interior volume. The interior volume has at least a top portion, a bottom portion, and a plurality of

parallel passageways extending from the bottom portion to the top portion for the throughflow of the liquid medium.

**[00028]** The container can be formed by fusion bonding three sheets of plastic material along predetermined seams to provide at least an interior volume and a second interior volume. The interior volume has at least a top portion, a bottom portion, and a plurality of parallel passageways extending from the bottom portion to the top portion for the throughflow of the liquid medium. The second interior volume has at least a top portion, a bottom portion, and a plurality of parallel passageways extending from the bottom portion to the top portion for the throughflow of a heat transferring fluid. The second interior volume can be a heat exchanger for circulating heat transferring fluid therethrough. The interior volume and the second interior volume have opposed top portions, bottom portions and parallel passageways. The opposing parallel passageways in horizontal cross section comprise a series of parallel crests intercalated with troughs along the width of the container, wherein pairs of opposed troughs are joined at an inner wall, thereby defining a series of adjacent parallel passageways in the interior volume and the second interior volume. An embodiment of this disclosure comprises altering the volume of the parallel passageways of the interior volume using pressure resulting from heat transferring fluid in opposed parallel passageways of the second interior volume.

**[00029]** The container can be formed by fusion bonding four sheets of plastic material along predetermined seams to provide at least the interior volume, a second interior volume and a third interior volume. The second interior volume is positioned between the interior volume and the third interior volume. The interior volume and the third interior volume have at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to the top portion for the throughflow of the liquid medium. The second interior volume has at least a top portion, a bottom portion, and a plurality of parallel passageways extending from the bottom portion to the top portion for the throughflow of a heat transferring fluid. The second interior volume is a heat exchanger for circulating heat transferring fluid therethrough. The interior

volume, the second interior volume and the third interior volume have opposed top portions, bottom portions and parallel passageways. The opposing parallel passageways in horizontal cross section comprise a series of parallel crests intercalated with troughs along the width of said container, wherein pairs of opposed troughs are joined at an inner wall, thereby defining a series of adjacent parallel passageways in the interior volume, the second interior volume and the third interior volume. An embodiment of this disclosure comprises altering the volume of the parallel passageways of the interior volume and the third interior volume using pressure resulting from heat transferring fluid in opposed parallel passageways of the second interior volume.

**[00030]** The photobioreactor of this disclosure comprises a non-rigid, semi-rigid or rigid container, preferably a non-rigid container, having at least a front wall, a rear wall and an interior volume. The photobioreactor is preferably a flat panel photobioreactor. The flat panel photobioreactor has a corrugated appearance resulting from the parallel passageways. At least a portion of the front wall and rear wall is transparent and/or translucent. The front wall and rear wall allow sufficient light to enter the liquid medium for the photocultivation of microalgae. The container can be substantially rectangular in structure having a length of from about 100 centimeters to about 600 centimeters and a width of from about 45 centimeters to about 300 centimeters, preferably a length of from about 150 centimeters to about 400 centimeters and a width of from about 120 centimeters to about 250 centimeters.

**[00031]** The bottom portion of the photobioreactor can contain one or more gas spargers for introducing gas into the parallel passageways. One photobioreactor typically can have 4 or more gas spargers (segments) in the bottom portion. The gas spargers comprise at least one gas inlet line having a plurality of secondary gas inlet lines protruding therefrom. The gas inlet line extends from the gas inlet opening and the secondary gas inlet lines extend into inlet openings in at least a portion of the parallel passageways (risers as described herein) for introducing gas into the risers. Typically, a gas sparger segment has from about 4 to about 10, preferably from about 4 to about 8, secondary gas inlet



lines extending into the same number of risers. The bottom portion also contains diffusers for introducing carbon dioxide into the parallel passageways.

**[00032]** Creation of controllable turbulent flow within the photobioreactor is important to a high productivity of algae biomass and provides the means to thwart the effects of over radiation including phototoxicity. Turbulent flow is achieved by injecting individual streams of gas into each riser. These gas streams should be uniform from riser to riser. Control of the gas stream should be such that when the gas flow is changed, the gas flow in each riser is uniformly changed.

**[00033]** The sparger useful in the photobioreactor achieves the essential uniform flows among the risers by static means. No mechanical controls from riser to riser need be employed. The sparger useful in the photobioreactor is self clearing, after it is flooded by an influx of water when the gas flow is interrupted and the gas pressure drops. The sparger useful in the photobioreactor accounts for the contraction of the horizontal dimension of the photobioreactor when gas is applied and the device inflates.

**[00034]** Uniform flows are achieved by direct injection of equal volumes of gas into each riser through an inserted individual tube. Diameters for the headers and for the individual tubes are chosen such that the pressure drop during periods of gas flow is sufficient to prevent the back flow of water. The diameters of the headers from which the individual tubes project are chosen such that the pressure drop during periods of gas flow is sufficient to prevent the back flow of water. Further diameters of the individual tubes and the headers are chosen such that water that may inflow during an interruption of the gas flow is expelled by gas pressure. The diameters of the individual tubes and headers are chosen on the basis of the pressure drop they impose when the required amount of gas flows through it.

**[00035]** The required amount of gas is the volume that impels the liquid up the riser to achieve a range of turbulent flows in the liquid phase equivalent to a Reynolds number in the range of 1800 to 3000. For example, a riser with an inflated diameter of 0.75 inch requires an gas flow in the range of .015 cubic feet

per minute (cfm) to 0.25 cubic feet per minute to achieve the above range of Reynolds numbers. See, for example, the graph depicted in Fig. 5. These ranges of flows will expel accumulated water and will provide equal distribution of gas flow to all individual tubes when the velocity of the gas imposes an overall pressure drop of 1.0 psi or more, when measured from the inlet to the header to the outlet of the individual tubes.

**[00036]** Spargers can be customized and produced by various molding techniques. Spargers can also be assembled from common pipe, tubing and fittings in diameters to match the minimum pressure drop requirement of 1.0 psi.

**[00037]** The data in Table 1 of the Examples below indicate that a sparger that provides good gas flow distribution and is capable of expelling liquid accumulations, imposes a large pressure drop at high turbulence. High turbulence is required in situations of high radiation where damage by phototoxicity is a threat. These situations may be prolonged in tropical or desert locales.

**[00038]** In an embodiment of this disclosure, two or more parallel spargers to the same bank of risers is a solution to a high pressure drop while delivering the required gas volume. The data in Table 1 of the Examples indicate that a Reynolds number of 3000 can be achieved with 2 spargers operating with a pressure drop of less than 2.5 psi. In the 2 sparger embodiment, each sparger will produce 0.14 cfm per riser and experience a pressure drop of less than 2.5 psi. The justification of 2 or more spargers is an economic tradeoff between the cost of energy for gas compression for one sparger and the cost of an additional sparger for the 2 or more sparger embodiment and its controls.

**[00039]** When the gas pressure is applied to a photobioreactor fabricated from non-rigid material, the horizontal dimension contracts as the photobioreactor is inflated. The sparger does not contract because it is not subject to inflationary forces and is made of a stiffer material.

**[00040]** Contraction is accommodated for by segmenting the header portion of the sparger to a length that matches the contracted width of the inflated photobioreactor. A header can accommodate 20 individual tubes or more, but a header length that will accommodate 4 to 10 individual tubes is preferable. The

lesser length of the header is such that the individual tubes projecting from the header align with the inlet to the risers after the photobioreactor is inflated.

**[00041]** For photobioreactors employing numerous risers, multiple spargers can be utilized. For example, a photobioreactor that employs 30 risers could utilize up to 5 or 6 spargers or more.

**[00042]** In the photobioreactor, at least a portion of the parallel passageways are configured for the flow of liquid medium from the bottom portion to the top portion. These are known as risers. Also, at least a portion of the parallel passageways are configured for the flow of liquid medium from the top portion to the bottom portion. These are known as down comers. For each photobioreactor, the number of risers and down comers can vary over a wide range to optimize flow through of product and thereby optimize productivity. Typically, there can be approximately 4 risers for every 1 down comer, but other combinations may also be desirable. The internal diameter of the down comers can be wider than the internal diameter of the risers in order to balance the downward flows with the upward flows. The number of parallel passageways in a photobioreactor is not narrowly critical and can vary over a wide range. Typically, there can be from about 4 to about 6 groupings of risers/down comers (for example, one grouping comprises about 4 risers for every 1 down comer) in a photobioreactor.

**[00043]** As indicated above, a heat exchanger system can be used for controlling the temperature of the photobioreactor. A fluid can travel through or be contained in an interior volume of the photobioreactor that is adjacent to the interior volume where cellular cultivation occurs. A warm or cool fluid, depending on the desired temperature within the photobioreactor, can be used to heat or cool the surfaces of the photobioreactor, which in turn controls the temperature within the reactor. The temperature of the fluid is transferred to the circulating biomass inside. A fluid used to control the temperature can be water. Alternatively, for photobioreactors without an interior volume heat exchanger system, the photobioreactor can be sprayed with warm or cool water or operated in a controlled environment (for example, a greenhouse).

**[00044]** Operating conditions of the photobioreactor can vary over a wide range depending on the particular microorganisms, microorganism cultures and biomass used. Operating conditions such as temperature, pressure and residence (mixing) time may vary greatly and any suitable combination of such conditions may be employed herein. The temperature may range from about 18°C to about 35°C, preferably from about 24°C to about 30°C. The pressure may range from about 1 psig to about 9 psig, preferably from about 2 psig to about 5 psig. The pressure will be limited by the strength of the materials of construction (for example, plastic film) of the photobioreactor. The starting materials (for example, liquid medium and carbon dioxide) can be added to the photobioreactor in any order. The mixing time employed can range from about 1 to about 20 days, preferably from about 2 to 8 days. Additionally, the operating pH is dependent on the particular microorganisms, microorganism cultures and biomass used. The pH can range from about 5 to about 9, preferably from about 7 to about 8.5.

**[00045]** The photobioreactor of this disclosure can be operated in any suitable environment (for example, arid or tropical environments). The heat exchanger described herein can be used for controlling the temperature of the photobioreactor in a particular environment. The photobioreactors of this disclosure can also be located and operated underground or indoors using artificial light sources as described herein.

**[00046]** The photobioreactor can further comprise a liquid medium feed line connected to the liquid medium inlet opening. The liquid medium feed line can extend from the liquid medium inlet opening exteriorly from the bottom portion for delivery of liquid medium into the interior volume. The liquid medium feed line can optionally contain one or more liquid medium flow control valves therein for control of flow of the liquid medium therethrough. The liquid medium feed line can be connected to a liquid medium supply source.

**[00047]** The photobioreactor can further comprise a microorganism culture discharge line connected to the microorganism culture outlet opening. The microorganism culture discharge line can extend from the microorganism culture outlet opening exteriorly from the bottom portion for removal of microorganism

culture from the interior volume. The microorganism culture discharge line can optionally contain one or more microorganism culture discharge control valves therein for control of flow of the microorganism culture therethrough. The microorganism culture discharge line can be connected to a collector.

**[00048]** The photobioreactor can further comprise a gas feed line connected to the gas inlet opening. The gas feed line can extend from the gas inlet opening exteriorly from the bottom portion for delivery of gas into the interior volume. The gas feed line can optionally contain one or more gas flow control valves therein for control of flow of the gas therethrough. The gas feed line can be connected to a gas supply source. The gas supply source can be sequestered carbon dioxide. The carbon dioxide can also be extracted from air or a waste stream (for example, treated stack gas, chemical plant off gas, a non-toxic waste gas, and the like).

**[00049]** The photobioreactor can further comprise one or more air inlet openings at the bottom portion of the container for introducing at least air into the container. The bottom portion of the photobioreactor can contain one or more gas spargers for introducing air into the parallel passageways. One photobioreactor typically can have 4 or more gas spargers (segments) in the bottom portion. The gas spargers comprise at least one air inlet line having a plurality of secondary air inlet lines protruding therefrom. The air inlet line extends from the air inlet opening and the secondary air inlet lines extend into inlet openings in at least a portion of the parallel passageways (risers as described herein) for introducing air into the risers. Typically, a gas sparger segment has from about 4 to about 10, preferably from about 4 to about 8, secondary air inlet lines extending into the same number of risers.

**[00050]** The photobioreactor can further comprise one or more air feed lines connected to the one or more air inlet openings. The one or more air spargers in the bottom portion extend from the one or more air inlet openings. The one or more air feed lines can extend from the one or more air inlet openings exteriorly from the bottom portion for delivery of air into the interior volume. The one or more air feed lines can optionally contain one or more air flow control

valves therein for control of flow of the air therethrough. The one or more air feed lines can be connected to one or more air supply sources.

**[00051]** The photobioreactor can further comprise a gas discharge line connected to the gas outlet opening. The gas discharge line can extend from the gas outlet opening exteriorly from the top portion for removal of gas from the interior volume. The gas discharge line can optionally contain one or more gas discharge control valves therein for control of flow of the gas therethrough. The gas discharge line can be connected to a collector or a gas treatment system.

**[00052]** The photobioreactor can be oriented, angled or tilted toward the sun or other light source. This orientation of the photobioreactor can be determined based on facing the photobioreactor in an improved manner for receiving light absorption by orienting the light absorbing surfaces normal to the sun at any time during the day or season of the year. The photobioreactor can be oriented according to the position of the sun throughout the day in an attempt to minimize the angle of incidence of light energy to the surface of the photobioreactor. For artificial light sources, the artificial light source can be oriented to face the light absorbing surfaces of the photobioreactor.

**[00053]** Although the photobioreactor preferably utilizes natural sunlight, an artificial light source providing light at a wavelength able to drive photosynthesis may be utilized instead of or in supplement to natural sunlight. Examples of artificial light sources include, for example, LEDs, light bulbs, halogen lamps, and the like. Another artificial light source can be light transmitted by fiber optic. Reflective or refractive light may also be a suitable light source. A photobioreactor utilizing both sunlight and an artificial light source may be configured to utilize sunlight during the daylight hours and artificial light in the night hours, so as to increase the total amount of time during the day in which the photobioreactor can convert carbon dioxide to biomass through photosynthesis. Artificial light sources are useful for under cover photobioreactors (for example, photobioreactors that are located and operated underground). The artificial light source can also be configured to face light absorbing surfaces of the photobioreactor. Additionally, a support component

capable of supporting said photobioreactor can be configured to tilt the photobioreactor based on the position of the sun or other light source as described herein.

**[00054]** The photobioreactor surfaces of light absorption may be physically or chemically modified to improve light absorption. This can include modification to reflect undesired light (for example, ultraviolet light) while absorbing desired light (for example, photosynthetically active radiation between 400 and 700 nm or light in the visible spectrum) for any angle of incidence.

**[00055]** In some instances, a wavelength selective surface comprises a wavelength selective coating. In some instances, a wavelength selective surface comprises a wavelength selective plastic. A wavelength selective surface can involve adding chemicals to the plastic (for example, doping the plastic), layering different kinds of plastic, coating plastic with other chemicals.

**[00056]** Examples of types of wavelength selective coatings for a photobioreactor include, for example, coatings that block ultraviolet light, coatings that prevent light-induced degradation of the plastic, coatings that block infrared light, coatings that minimize reflectance at certain wavelengths, coatings that maximize reflectance at certain wavelengths, coatings that minimize/maximize transmission of certain wavelengths, and coatings that minimize/maximize absorption of certain wavelengths.

**[00057]** A plurality of photobioreactors can be arranged to form a system for the growth and production of a photosynthetic biomass. A photobioreactor system can comprise one of a plurality of identical or similar photobioreactors interconnected in parallel, in series, or in a combination of parallel and series configurations. Configuring a system of photobioreactor into series or parallel arrangements will provide the opportunity for close control of the reaction conditions module to module and further optimize the output of the overall photobioreactor system. All such configurations and arrangements of the photobioreactors are included within the scope of this disclosure.

**[00058]** In some instances, each photobioreactor in a system of photobioreactors can operate independently. The photobioreactor can be modular

and easily swapped if desired. For example, if one photobioreactor becomes contaminated with another species of algae or other organism, it can be swapped for a different photobioreactor. For photobioreactors in series or in parallel in which a photobioreactor has become contaminated, this disclosure allows for the containment and/or isolation of the contaminants.

**[00059]** Although a system of photobioreactors can be intended to be modular and self-contained, harvest processes, medium recycling, water storage, power generation and other processes may be centralized and distributed to individual photobioreactors. Independent photobioreactors can be connected in a network so that dispersal of medium and collection of biomass products can be centrally coordinated.

**[00060]** A control system and methodology can optionally be utilized in the operation of a photobioreactor which is configured to enable automatic, real-time optimization and/or adjustment of operating parameters to achieve desired or optimal photomodulation and/or growth rates for a particular environmental operating conditions. Also, methods and systems can be provided for preselecting, adapting, and conditioning one or more species of photosynthetic organisms to specific environmental and/or operating conditions to which the photosynthetic organisms will subsequently be exposed during utilization in a photobioreactor.

**[00061]** A computer implemented system can optionally be used to control light exposure, media flow rates, gas exchange rates, internal turbulence, orientation of the photobioreactor in respect to the sun or other light source, heating and cooling of the photobioreactor, mixing, and harvesting of the biomass. The computer control system can have the ability to adjust different parameters to optimize growth of the biomass in the photobioreactor. The system can be implemented to adjust parameters automatically. For example, a computer implemented system can calculate light exposure intervals to determine the duration of exposure of the biomass, on average, to light intensities both above and below an optimum intensity required to drive photosynthesis in a log-based



manner. In another example, the system can determine the frequency of exposure of the algae to light and dark periods of the biomass.

**[00062]** Control of the photobioreactor can be achieved using conventional hardware or software-implemented computer and/or electronic control systems together with a variety of electronic sensors. Using the control systems, components for nutrient level maintenance, pH control, and other factors can be added automatically directly into the liquid phase within the photobioreactor, if desired. The control system can also be configured to control the temperature in the photobioreactor by either or both of controlling a heat exchanger system or heat control system within or connected with the photobioreactor.

**[00063]** The photobioreactor can further comprise sensors for measuring a number of parameters that are representative of the growth of said microorganism culture. A control unit can be connected to the sensors and at least one of the liquid medium inlet openings, gas inlet openings and air inlet openings for introducing liquid medium, gas and air in order to stimulate growth of the microorganism culture in accordance with the measured parameter values.

**[00064]** The computer implemented system can optionally be part of or coupled with a photobioreactor. The system can be configured or programmed to control and adjust operational parameters of the photobioreactors as well as analyze and calculate values. The computer implemented system can send and receive control signals to set and control operating parameters of the photobioreactor and, optionally, other related apparatuses. The computer implemented system can be remotely located with respect to the photobioreactor. It can also be configured to receive data from one or more remote photobioreactors via indirect or direct means, such as through an ethernet connection or wireless connection. The control system can be operated remotely, such as through the Internet.

**[00065]** Part or all of the control of a system or photobioreactor can be accomplished without a computer (for example, using a thermostat to control temperature). Other types of control may be accomplished with physical controls. In an instance, a control system can be a manual system operated by a user. In

another example, a user may provide input to a control system as described. A suitable pressure gauge may be used to monitor air pressure (for example, air and carbon dioxide) in the container. The air pressure gauge can have a suitable shut-off valve that may be preset to shut off the supply of air and/or carbon dioxide to the container if the pressure therein exceeds a predetermined value. Such a system is useful in case of a blockage of the outflow of waste gases, which could otherwise lead to a buildup of pressure inside the container, eventually bursting the container.

**[00066]** The photobioreactor is designed to provide favorable use of energy inputs (for example, light) for growing microalgae. Photosynthetic organisms such as algae use sunlight as an energy source to grow, divide, and/or make products, where the organisms and/or products can be used to create an eventual product output of the photobioreactor. In some instances, too much sunlight can be detrimental to photosynthetic or algal growth, resulting in what is described as phototoxicity. Phototoxicity is caused by excessive light penetrating or impinging upon a cell and can lead to reduction in biosynthesis efficiency and, in some cases, can lead to cell death. Because of phototoxicity, light intensity should be regulated along with monitoring specific wavelengths of light for improvement of biosynthesis efficiency. The exposure of individual cells to light intensity can be adjusted by control of internal turbulence. By creating adjustable levels of turbulence in the parallel passageways, this disclosure allows for optimal exposure of the microorganisms and biomass to sunlight, thereby optimizing growth.

**[00067]** The photobioreactor may be used in conjunction with a support component capable of supporting said photobioreactor. The support component should be mechanically stable and capable of supporting the photobioreactor. For example, the support component can comprise a frame. The frame can be rectangular comprising substantially parallel and horizontal upper and lower load carrying members. The support component can be configured to tilt the photobioreactor based on the position of the sun or other light source.

**[00068]** Referring to Figs. 1-6, operation of the photobioreactor involves introducing liquid medium into the parallel passageways via a liquid medium inlet opening. The liquid medium can be supplied to the photobioreactor from a liquid medium supply source through a liquid medium feed line connected to the liquid medium inlet opening. Carbon dioxide can be introduced into the liquid medium via a gas inlet opening. The carbon dioxide can be supplied from a gas supply source through a gas feed line connected to the gas inlet opening. The gas supply source can include, for example, air, treated stack gas, chemical plant off gas, any non-toxic waste gas, and the like. The gas supply source provides carbon dioxide for the microorganism culture. The gas supply source can provide an impelling gas that creates circulation and turbulence in the photobioreactor. Air can also be introduced into the liquid medium via an air inlet opening. Air can be supplied from an air supply source through an air feed line connected to the air inlet opening. The air supply source can include, for example, air, treated stack gas, chemical plant off gas, any non-toxic waste gas, and the like. The air supply source provides an impelling gas that may or may not contain carbon dioxide, and that creates circulation and turbulence in the photobioreactor.

**[00069]** As used herein, the terms “gas”, “gases” and “air” are not intended to be limiting in any manner and can be used interchangeably in instances herein. For example, in instances herein, the terms “gas” and “gases” refer to air, treated stack gas, chemical plant off gas, any non-toxic waste gas, and other gases, that may or may not contain carbon dioxide. Also, for example, in instances herein, the term “air” refers to air, treated stack gas, chemical plant off gas, any non-toxic waste gas, and other gases, that may or may not contain carbon dioxide. Both “gas” and “air” can be used as a source of carbon dioxide for the microorganism culture. Both “gas” and “air” can be used as an impelling gas for creating circulation and turbulence in the photobioreactor. Such terms should be construed in conjunction with the particular description and context in which they are used, and should not be construed as limitations to the scope of this disclosure.

**[00070]** The photobioreactor can be oriented to receive adequate light or, in the case of artificial light, the artificial light can be oriented to face light absorbing

surfaces of the photobioreactor. A support component capable of supporting the photobioreactor can be configured to tilt the photobioreactor based on the position of the sun or other light source as described herein. Growth occurs by exposing the liquid medium to carbon dioxide and light. After the biomass reaches a predetermined density, the operation can become continuous by providing a continuous flow of liquid medium to the photobioreactor via the liquid medium inlet opening and allowing liquid and biomass material to exit the photobioreactor via the microorganism culture outlet opening. The microorganism culture outlet opening is connected to a microorganism culture discharge line for removing microorganism culture from the photobioreactor. Gas formed in the photobioreactor can be removed via a gas discharge line connected to a gas outlet opening.

**[00071]** Once entering the photobioreactor, the liquid medium is circulated in a reflux fashion. The liquid medium is impelled by the flow of gas up to the top portion of the photobioreactor via the risers. By the force of gravity, the liquid medium flows back to the bottom portion of the photobioreactor via the down comers where the medium is mixed with incoming medium. For each photobioreactor, the number of risers and down comers can vary over a wide range to optimize flow through of product and thereby optimize productivity. Typically, there can be approximately 4 risers for every 1 down comer, but other combinations may also be desirable. The internal diameter of the down comers can be wider than the internal diameter of the risers in order to balance the downward flows with the upward flows.

**[00072]** The liquid medium used herein contains one or more microorganisms and nutrients for the growth of the microorganisms and for the production of biomass by photosynthesis. In particular, the liquid medium contains at least one photosynthetic microorganism, water, nutrients for photosynthetic growth, and optionally other liquids suitable for growing photosynthetic microorganisms. The preferred photosynthetic microorganism is microalgae. The liquid medium may also contain bacteria and/or archaea (i.e., an

ancient form of bacteria). The microalgae can have a coexistence with the bacteria and/or archaea in the photobioreactor as described herein.

**[00073]** Microorganisms and the microorganism cultures and biomass produced in accordance with this disclosure include all organisms capable of photosynthetic growth, such as plant cells and microorganisms in unicellular and multicellular form and products produced by the photosynthetic organism. The microorganisms are capable of growth in the liquid medium. Illustrative microorganisms that can be cultivated and harvested include those modified by natural selection, induced mutation, directed evolution, synthetic assembly, or genetic manipulation. While the cultivation of microalgae is a preferred cultivation in accordance with this disclosure, other photosynthetic microorganisms may be utilized in place of or in addition to microalgae.

**[00074]** Illustrative microorganisms and microorganism cultures and biomass that can be grown in the photobioreactor of this disclosure include various types of microalgae, for example, cyanobacteria species (for example, *microcystis*), green algae (for example, *Chlorella*, *Botryococcus*, *Ankistrodesmus*, *Chlamydomonas*, *Dunaliella*), and diatoms (for example, *Thalassiosira*, *Navicula*). The photobioreactor can also include heterotrophic growth as a component. In some instances, the algae can be cultivated in a suitable medium that supports either autotrophic or both autotrophic and heterotrophic growth. The photobioreactor may also be used for the cultivation of multicellular aquatic plants, including macroalgae such as seaweeds and red algae. Various types of bacteria and/or archaea can be grown in the photobioreactor of this disclosure. Mixtures of bacteria and/or archaea, including mixtures of bacteria and/or archaea with other microorganisms, can also be grown in the photobioreactor of this disclosure.

**[00075]** The microalgae can comprise marine or fresh water microalgae. Suitable marine microalgae can be selected from *Skeletonema*, *Chaetoceros*, *Thalassiosira*, *Tetraselmis*, *Isochrysis*, and *Nanochloropsis*. Suitable fresh water microalgae can be selected from *Haematococcus*, *Chlamydomonas*, and *Spirulina*.

**[00076]** Other microorganisms and microorganism cultures and biomass that can be grown in the photobioreactor of this disclosure include, for example, genetically modified microalgae, sterile microalgae, sterile genetically modified microalgae, mixtures of microalgae, mixtures of genetically modified microalgae, sterile mixtures of microalgae, sterile mixtures of genetically modified microalgae, mixtures of microalgae with other microorganisms, mixtures of genetically modified microalgae with other microorganisms, sterile mixtures of microalgae with other microorganisms, and sterile mixtures of genetically modified microalgae with other microorganisms.

**[00077]** In an embodiment, the microalgae can have a coexistence with bacteria and/or archaea in the photobioreactor. The bacteria and/or archaea can include one or more species of bacteria and/or archaea, preferably a population of different species of bacteria and/or archaea. In such a coexistence, the bacteria and/or archaea graze on microalgae waste. The bacteria and/or archaea remove waste that would otherwise inhibit growth and division rate of the microalgae culture. The commercial benefit is that higher cell densities can be obtained when the inhibitors are removed. For example, one of the waste products is a growth inhibitor that limits microalgae cell densities to about 5 grams per liter. By eliminating the growth inhibitor, microalgae cell densities can be increased to about 40 grams per liter or more. The cost of cell recovery is less for high density microalgae cultures than for low density microalgae cultures. Suitable bacteria and/or archaea include those species that can have a coexistence with the microalgae and consume microalgae waste. Preferably, the bacteria and/or archaea are present in an amount sufficient to reduce or eliminate microalgae waste in the photobioreactor.

**[00078]** Selected bacteria and/or archaea can be added to sterile microalgae cultures such that the sterile microalgae can have a coexistence with the selected bacteria and/or archaea in the photobioreactor. Suitable selected bacteria and/or archaea include those species that can have a coexistence with the sterile microalgae and consume sterile microalgae waste without contaminating or otherwise interfering with sterility of the microalgae culture. Preferably, the

selected bacteria and/or archaea are present in an amount sufficient to reduce or eliminate sterile microalgae waste in the photobioreactor without any contamination or adverse effect on sterility of the microalgae culture.

**[00079]** The photobioreactor of this disclosure preferably utilizes conditions that enable efficient levels of algal autotrophic growth (for example, using only inorganic carbon as a carbon source). The photobioreactor can also utilize conditions using autotrophic and heterotrophic growth in combination (for example, using carbon dioxide and other organic molecules, such as sugar, acetate or starch, as the carbon source). This is also known as mixotrophic growth.

**[00080]** In an embodiment, the microorganisms and microorganism cultures and biomass can be grown under sterile conditions. Also, heterotrophic and/or mixotrophic microorganisms can be grown under autotrophic conditions sufficient for said heterotrophic and/or mixotrophic microorganisms to adopt autotrophic characteristics. Autotrophic microorganisms can be grown under mixotrophic conditions or autotrophic conditions.

**[00081]** Algal cells can be kept at a density within the photobioreactor whereby cells divide at a logarithmic rate. To maintain a desired cell density in the culture with growing and dividing cells, fresh medium (for example, water, water with sugar, and nutrients) may be directed into the photobioreactor to dilute the culture, and algal and medium may be directed out of the photobioreactor to be harvested. In order to be harvested, medium containing algal biomass can be pumped out of the photobioreactor, or can flow out as a consequence of fresh medium being added. The density of the microorganisms and microorganism culture and biomass in the photobioreactor can range from about 0.2 grams per liter (dry basis) to about 40 grams per liter (dry basis), preferably from about 2 grams per liter (dry basis) to about 10 grams per liter (dry basis).

**[00082]** The medium that can be directed into the photobioreactor can comprise water or a saline solution (for example, sea water or brackish water) containing sufficient nutrients to facilitate viability and growth of algae and/or other photosynthetic microorganisms. A medium can be utilized that comprises brackish water, sea water (filtered or unfiltered) and other non-potable water. The

medium can be obtained from a locality in which the photobioreactor will be operated. Particular medium compositions containing water plus nutrients required or suitable for use in maintaining a growing culture of algae or other photosynthetic organism are well known in the art. The medium can include a source of nitrogen, phosphate, and micronutrients such as essential metals. The medium may also contain a protein or sugar source.

**[00083]** Carbon dioxide gas can be pumped into the photobioreactor in order to grow biomass. The carbon dioxide can be filtered air or unfiltered air that may or may not be supplemented with certain gases to improve growth and algal productivity. The gas entering the photobioreactor can also be used for heating or cooling as well as mixing the liquid growth medium and biomass. The flow rate of carbon dioxide and optionally other gases (for example, nitrogen source gases) in the photobioreactor can range from about 0.01 percent volume of inlet gas flow to about 5 percent volume of inlet gas flow, preferably from about 0.2 percent volume of inlet gas flow to about 1.5 percent volume of inlet gas flow.

**[00084]** Gas can be pumped into the photobioreactor in order to grow biomass. The gas can be filtered air or unfiltered air that may or may not be supplemented with certain gases (for example, carbon dioxide) to improve growth and algal productivity. The gas entering the photobioreactor can also be used for heating or cooling as well as mixing the liquid growth medium and biomass. Introducing gas into the parallel passageways creates a turbulence in the liquid medium sufficient to prevent sedimentation of microorganisms, limit exposure of individual cells to light intensity, and prevent adherence of microorganisms to walls of the parallel passageways. The turbulence in the liquid medium can be controlled by airflow in the parallel passageways. The flow rate of gas in the photobioreactor is preferably of sufficient volume to create laminar or turbulent flow of the liquid medium in the parallel passageways. Reynolds numbers of the flowing media can range from about 500 to about 4,000, preferably from about 2100 to about 3,000.

**[00085]** The biomass and liquid medium inside the photobioreactor may be mixed. In some instances, mixing can allow for dispersal of nutrients and carbon



dioxide, efficient removal of waste products, maintenance of optimal light conditions for each individual algal cell, and reduction of algal colony adherence on the walls of the photobioreactor. Mixing can be achieved through the pumping of water and/or medium into the photobioreactor.

**[00086]** A liquid medium can be self-shading where the cells more interior are shaded by the cells more exterior. In a dense medium, cells more than a few centimeters below the surface may receive insufficient light. If the medium is mixed rapidly, this can produce a flashing light effect. The algae can grow as efficiently with intermittent exposure to light as under constant light at the same intensity. To maximize the growth, the biomass material should move freely throughout the parallel passageways without any stagnated movement. Gas can be introduced into the parallel passageways to cause turbulence in the liquid medium, thereby disrupting any stagnant biomass material and creating a continuous and even flow of material through the parallel passageways. For photobioreactors configured in series or in parallel, different levels of turbulence can be produced in the individual photobioreactors as desired.

**[00087]** As indicated above, turbulence in the liquid medium can be controlled by airflow in the parallel passageways. The introduction of gas into the parallel passageways creates turbulence in the liquid medium sufficient to prevent sedimentation of microorganisms, limit exposure of individual cells to light intensity, and prevent adherence of microorganisms to walls of the parallel passageways. Preferably, in operating the photobioreactor of this disclosure, uniform internal turbulence can be created in the parallel passageways by controlling gas flow in each of the parallel passageways. Internal turbulence in the parallel passageways of a single photobioreactor or a series of photobioreactors can be adjusted by controlling the gas flow in the parallel passageways. In some instances in which sunlight is used as the light source, turbulence may be adjusted to higher levels to prevent phototoxicity (for example, at midday) and to lower levels during other times of the day (for example, at night).

**[00088]** As indicated above, one or more gas spargers are used for introducing gas into the parallel passageways. Turbulent flow is created by injecting individual streams of gas through the one or more spargers into each riser. The gas streams are preferably uniform from riser to riser. The gas streams are controlled such that when the gas flow is changed, the gas flow in each riser is uniformly changed.

**[00089]** In accordance with this disclosure, energy (or light) from the sun is used to drive photosynthesis. The photobioreactor is preferably a non-rigid container that has at least a front wall and a rear wall, and at least a portion of the front wall and rear wall is transparent to light of a wavelength capable of driving photosynthesis, for example, light of a wavelength between about 400-700 nm. The photobioreactor can also have a source of light capable of driving photosynthesis associated therewith. Since different types of biomass can require different light exposure conditions for optimal growth and proliferation, light modification apparatus or devices can be utilized in the construction of a photobioreactor. The light can be natural or artificial.

Control of the concentration of biomass within the photobioreactor can be important both from the standpoint of maintaining a desirable level of algal growth and proliferation. Biomass can be harvested periodically or continuously to maintain the desired concentration range during operation. The harvesting can take place in a continuous or semi-continuous fashion, meaning that biomass is constantly removed, or only a portion of the biomass is removed from the photobioreactor at a given time. Harvesting can be accomplished by pumping out medium, or by pumping in medium to displace biomass out of the photobioreactor in a controlled way. Harvested algae and medium can be separated, and medium can be tested and adjusted for reuse (for example, for nutrient concentration and pH). The medium may be filtered before use to ensure it is free of contaminants when entering the photobioreactor.

**[00090]** After discharge from the photobioreactor, it may be desirable to separate the microorganism culture medium into a liquid phase and a solid phase that contains the microorganisms (biomass). Such a separation can be achieved

by various processes. The biomass can be separated from the liquid phase by centrifugation, flocculation or with a filter unit.

**[00091]** During the photosynthetic process, carbon dioxide is absorbed and molecular oxygen is generated. The molecular oxygen can be released from the photobioreactor through the gas outlet openings. The molecular oxygen may also be recovered and used in another process. The absorption of carbon dioxide creates a favorable environmental effect.

**[00092]** Water-rich gas coming out of the photobioreactor may be put through a water condensing or water sequestering apparatus to decrease water loss from the system.

**[00093]** The method of this disclosure can be conducted in batch, semi-batch or continuous mode. The microorganism culture produced by the method of this disclosure can be used, for example, in biofuel production (for example, biodiesel), aquaculture food production, mammalian food production, and recombinant protein synthesis (for example, pharmaceutical and industrial enzymes). In addition, carbon dioxide can be utilized from sequestered sources.

**[00094]** In an embodiment, this disclosure relates in part to a method of producing biomass. The biomass produced by the method of this disclosure can be used, for example, in biofuel production (for example, biodiesel), aquaculture food production, mammalian food production, and recombinant protein synthesis (for example, pharmaceutical and industrial enzymes). Conversion of biomass can be carried out by conventional procedures known in the art.

**[00095]** In an embodiment, this disclosure relates in part to a method for producing biomass and a biofuel from the biomass. Biofuels can be produced from biomass by conventional procedures known in the art. For example, biodiesel can be produced through the transesterification of organically derived oils or fats from biomass. Biodiesel can be used as a replacement for or as a component of diesel fuel, jet fuel or heating oil. Illustrative biofuels include, for example, biodiesel, bioethanol, and the like.

**[00096]** In an embodiment, this disclosure relates in part to a method for producing and recovering a recombinant protein from biomass. Recombinant

proteins can be produced from biomass by conventional procedures known in the art. For example, recombinant DNA can be inserted into the chromosomal DNA of microalgae. The microalgae will produce recombinant protein based on the recombinant DNA. Illustrative recombinant proteins include, for example, antibodies, antigens, hormones, enzymes, and the like.

**[00097]** In an embodiment, this disclosure relates in part to a method for producing aquaculture food from biomass. Aquaculture food can be produced from biomass by conventional procedures known in the art. For example, aquaculture food can be produced by drying and pelletizing the biomass. Aquaculture food can be used, for example, in fish farming (e.g., salmon, talapia, catfish, cod and shellfish). Illustrative aquaculture food includes, for example, shrimp food, shrimp food supplements, and the like.

**[00098]** In an embodiment, this disclosure relates in part to a method for producing mammalian food from biomass. Mammalian food can be produced from biomass by conventional procedures known in the art. For example, mammalian food can be produced by drying and pelletizing the biomass. Mammalian food can be used, for example, in swine and cattle feed. Illustrative mammalian food includes, for example, extract oils, and the like.

**[00099]** Large-scale commercialization of algae applications, e.g., biofuels, is dependent on the development of a highly productive photobioreactor system that has a low cost. Flat panel photobioreactors, such as those of this disclosure, are significantly more productive than competing designs and are expected to be the type that will be used commercially. Productivity of photobioreactors is expressed in terms of grams of algae biomass per square meter per day. Flat panel designs achieve productivities of 50 to 70 grams per meter squared per day. Productivity of tubular designs is in the range of 20 to 30 grams per meter squared per day. For ponds and race ways, productivity is in the range of 10 to 15 grams per meter squared per day.

**[000100]** The conventional approach for achieving low cost processing systems in general is to build facilities with larger capacities and take advantage of the economies of scale that result from use of bigger processing equipment.

Economies of scale apply to commonly used processing equipment such as tanks, refining columns, pumps, compressors and their interconnecting piping. The economy of scale does not apply to photobioreactors because they are basically two dimensional devices. The only way to increase the capacity of a high productivity photobioreactor system is to add more photobioreactors, or in other words increase the area. The economies of scale do not apply for systems where capacity is increased simply by increasing the area.

**[000101]** However, the low cost requirement is met by a photobioreactor that can be mass produced and assembled in the field by simple operations. A mass produced photobioreactor is essential to successful commercialization of algae applications, e.g., biofuels. Economic analysis of algae biofuels production indicate that commercial feasibility can be achieved if the installed cost of the photobioreactor is \$4 per square foot or less and if the productivity is 50 grams per liter per square meter per day or more. Only a mass produced flat panel photobioreactor can meet these requirements.

**[000102]** The photobioreactor of this disclosure uniquely lends itself to mass production at low cost. The surface components, the fittings and the internals can be produced by various automated extrusion techniques. Assembly of these into a photobioreactor is a series of simple operations that can be performed by robotic assembly stations.

**[000103]** The photobioreactor of this disclosure also lends itself to high productivity. The photobioreactor of this disclosure can be used for commercial cultivation of microorganisms on an industrial scale.

**[000104]** As used herein, the terms “microorganism(s)”, “microorganism culture(s)” and “biomass” are not intended to be limiting in any manner and can be used interchangeably in instances herein. For example, in instances herein, the term “microorganism(s)” refers to those microorganisms contained in the liquid medium that is fed to the photobioreactor; the term “microorganism culture(s)” refers to a product cultured and harvested by the method of this disclosure; and the term “biomass” refers to a product manufactured by the method of this disclosure. Such terms should be construed in conjunction with the particular

description and context in which they are used, and should not be construed as limitations to the scope of this disclosure.

**[000105]** Various modifications and variations of this disclosure will be obvious to a worker skilled in the art and it is to be understood that such modifications and variations are to be included within the purview of this application and the spirit and scope of the claims.

#### Examples

**[000106]** A sparger system was configured to provide air to eight risers with an inflated diameter of 0.75 inch as depicted in Fig. 6. The sparger system included a header assemble from ¼ ID inch tubing with appropriate fittings, 1/8 inch diameter individual tubes, and at the end of each tube was a 1/8 inch to 1/16 inch reducer acting as a nozzle. Air flow to the sparger and resulting pressure drop data and calculated Reynolds numbers for this configuration are set forth in Table 1 below.

Table 1

<u>Flow to riser, cfm</u>	<u>Pressure drop, psi</u>	<u>Reynolds number</u>
0.083	0.9	< 1000
0.167	2.5	1900
0.25	5.2	2800
0.28	6.8	3000

**[000107]** The above data indicate that a sparger that provides good air flow distribution and is capable of expelling liquid accumulations, imposes a large pressure drop at high turbulence. High turbulence is required in situations of high radiation where damage by phototoxicity is a threat. These situations may be prolonged in tropical or desert locales.

**[000108]** Two parallel spargers to the same bank of risers in accordance with this disclosure is a solution to a high pressure drop while delivering the required

air volume. The data in Table 1 above indicates a Reynolds number of 3000 can be achieved with two spargers operating with a pressure drop of less than 2.5 psi. In the two sparger embodiment of this disclosure, each sparger will produce 0.14 cfm and experience a pressure drop of less than 2.5 psi.

Claims

1. A photobioreactor for culturing and harvesting at least one of a microorganism culture, said photobioreactor comprising:
  - a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;
  - said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;
  - one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;
  - one or more microorganism culture outlet openings at said bottom portion for removing microorganism culture from said container;
  - one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and
  - one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container.
2. The photobioreactor of claim 1 wherein said container is made from a plastic material.
3. The photobioreactor of claim 1 wherein said container is non-rigid, semi-rigid or rigid.
4. The photobioreactor of claim 1 further comprising one or more air inlet openings at said bottom portion for introducing at least air into said container.
5. The photobioreactor of claim 2 wherein said plastic material is selected from polyethylene, polypropylene, polycarbonate, polyvinylchloride, polyester and laminates thereof.



6. The photobioreactor of claim 2 wherein said plastic material comprises a laminate of polyethylene and nylon.
7. The photobioreactor of claim 2 wherein said plastic material has a wavelength selective surface.
8. The photobioreactor of claim 1 comprising a flat panel photobioreactor.
9. The photobioreactor of claim 1 wherein said bottom portion contains one or more gas spargers for introducing gas into said parallel passageways.
10. The photobioreactor of claim 1 wherein said bottom portion contains two or more parallel gas spargers for introducing gas into said parallel passageways.
11. The photobioreactor of claim 9 wherein said one or more gas spargers comprise at least one gas inlet line having a plurality of secondary gas inlet lines protruding therefrom, said secondary gas inlet lines extending into inlet openings in at least a portion of said parallel passageways for introducing gas into said parallel passageways.
12. The photobioreactor of claim 1 wherein said bottom portion contains diffusers for introducing carbon dioxide into said parallel passageways.
13. The photobioreactor of claim 1 wherein at least a portion of said parallel passageways (risers) are configured for the flow of said liquid medium from said bottom portion to said top portion to achieve recycle of medium.
14. The photobioreactor of claim 1 wherein at least a portion of said parallel passageways (down comers) are configured for the flow of said liquid medium from said top portion to said bottom portion.

15. The photobioreactor of claim 1 having one or more groupings of risers and down comers in which one grouping comprises at least about 4 risers and 1 down comer.

16. The photobioreactor of claim 15 having from about 4 to about 6 groupings of risers and down comers.

17. The photobioreactor of claim 15 wherein the internal diameter of said down comers is greater than the internal diameter of said risers.

18. The photobioreactor of claim 2 wherein said container is formed by fusion bonding two sheets of said plastic material along predetermined seams to provide at least said interior volume, said interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of said liquid medium.

19. The photobioreactor of claim 2 wherein said container is formed by fusion bonding three sheets of said plastic material along predetermined seams to provide at least said interior volume and a second interior volume, said interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of said liquid medium, and said second interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a heat transferring fluid.

20. The photobioreactor of claim 19 wherein said second interior volume is a heat exchanger for circulating heat transferring fluid therethrough.

21. The photobioreactor of claim 19 wherein said interior volume and said second interior volume have opposed top portions, bottom portions and parallel passageways.
22. The photobioreactor of claim 21 in which the opposing parallel passageways in horizontal cross section comprise a series of parallel crests intercalated with troughs along the width of said container, wherein pairs of opposed troughs are joined at an inner wall, thereby defining a series of adjacent parallel passageways in said interior volume and said second interior volume.
23. The photobioreactor of claim 19 further comprising altering volume of the parallel passageways of said interior volume using pressure resulting from heat transferring fluid in opposed parallel passageways of said second interior volume.
24. The photobioreactor of claim 2 wherein said container is formed by fusion bonding four sheets of said plastic material along predetermined seams to provide at least said interior volume, a second interior volume and a third interior volume, said second interior volume positioned between said interior volume and said third interior volume, said interior volume and said third interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of said liquid medium, and said second interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a heat transferring fluid.
25. The photobioreactor of claim 24 wherein said second interior volume is a heat exchanger for circulating heat transferring fluid therethrough.
26. The photobioreactor of claim 24 wherein said interior volume, said second interior volume and said third interior volume have opposed top portions, bottom portions and parallel passageways.

27. The photobioreactor of claim 26 in which the opposing parallel passageways in horizontal cross section comprise a series of parallel crests intercalated with troughs along the width of said container, wherein pairs of opposed troughs are joined at an inner wall, thereby defining a series of adjacent parallel passageways in said interior volume, said second interior volume and said third interior volume.

28. The photobioreactor of claim 24 further comprising altering volume of the parallel passageways of said interior volume and said third interior volume using pressure resulting from heat transferring fluid in opposed parallel passageways of said second interior volume.

29. The photobioreactor of claim 1 further comprising a liquid medium feed line connected to said liquid medium inlet opening, said liquid medium feed line extending from the liquid medium inlet opening exteriorly from said bottom portion for delivery of liquid medium into said interior volume, the liquid medium feed line optionally containing one or more liquid medium flow control valves therein for control of flow of the liquid medium therethrough.

30. The photobioreactor of claim 29 wherein said liquid medium feed line is connected to a liquid medium supply source.

31. The photobioreactor of claim 1 further comprising a microorganism culture discharge line connected to said microorganism culture outlet opening, said microorganism culture discharge line extending from the microorganism culture outlet opening exteriorly from said bottom portion for removal of microorganism culture from said interior volume, the microorganism culture discharge line optionally containing one or more microorganism culture discharge control valves therein for control of flow of the microorganism culture therethrough.

32. The photobioreactor of claim 31 wherein said microorganism culture discharge line is connected to a collector.
33. The photobioreactor of claim 1 further comprising a gas feed line connected to said gas inlet opening, said gas feed line extending from the gas inlet opening exteriorly from said bottom portion for delivery of gas into said interior volume, the gas feed line optionally containing one or more gas flow control valves therein for control of flow of the gas therethrough.
34. The photobioreactor of claim 33 wherein said gas feed line is connected to a gas supply source.
35. The photobioreactor of claim 34 wherein said gas supply source is sequestered carbon dioxide, treated stack gas, chemical plant off gas, or a non-toxic waste gas.
36. The photobioreactor of claim 4 further comprising an air feed line connected to said air inlet opening, said air feed line extending from the air inlet opening exteriorly from said bottom portion for delivery of air into said interior volume, the air feed line optionally containing one or more air flow control valves therein for control of flow of the air therethrough.
37. The photobioreactor of claim 36 wherein said air feed line is connected to an air supply source.
38. The photobioreactor of claim 37 wherein said air supply source is air, treated stack gas, chemical plant off gas, or a non-toxic waste gas.
39. The photobioreactor of claim 1 further comprising a gas discharge line connected to said gas outlet opening, said gas discharge line extending from the gas outlet opening exteriorly from said top portion for removal of gas from said

interior volume, the gas discharge line optionally containing one or more gas discharge control valves therein for control of flow of the gas therethrough.

40. The photobioreactor of claim 39 wherein said gas discharge line is connected to a collector.

41. The photobioreactor of claim 1 further comprising a collector for collecting microorganism culture discharged from said interior volume.

42. The photobioreactor of claim 1 further comprising sensors for measuring a number of parameters that are representative of the growth of said microorganism culture, and a control unit connected to the sensors and at least one of the liquid medium inlet openings, gas inlet openings and air inlet openings for introducing liquid medium, gas and air in order to stimulate growth of the microorganism culture in accordance with the measured parameter values.

43. The photobioreactor of claim 1 further comprising a computer implemented system for controlling at least one of light exposure, media flow rates, gas exchange rates, internal turbulence, orientation of said photobioreactor in respect to the sun or other light source, heating and cooling of said photobioreactor, mixing, and harvesting of biomass.

44. The photobioreactor of claim 11 wherein introducing gas into said parallel passageways creates a turbulence in said liquid medium sufficient to prevent sedimentation of microorganisms, limit exposure of individual cells to light intensity, and prevent adherence of microorganisms to walls of said parallel passageways.

45. The photobioreactor of claim 11 wherein introducing gas into said parallel passageways is conducted at a flow rate of sufficient volume to create laminar or turbulent flow of said liquid medium in said parallel passageways.

46. The photobioreactor of claim 11 wherein introducing gas into said parallel passageways creates a turbulence in said liquid medium having a Reynolds number of from about 500 to about 4000.

47. The photobioreactor of claim 44 wherein turbulence in said liquid medium is controlled by airflow in said parallel passageways.

48. The photobioreactor of claim 44 wherein exposure of individual microorganism cells to light intensity is controlled by controlling internal turbulence in said parallel passageways.

49. The photobioreactor of claim 1 wherein said front wall and said rear wall allow sufficient light to enter said liquid medium for the photocultivation of microalgae.

50. The photobioreactor of claim 49 wherein said light can be natural or artificial light.

51. The photobioreactor of claim 49 wherein said light can be reflective or refractive light.

52. The photobioreactor of claim 1 further comprising a support component capable of supporting said photobioreactor.

53. The photobioreactor of claim 52 in which the support component is configured to tilt the photobioreactor based on the position of the sun or other light source.

54. The photobioreactor of claim 50 in which artificial light can be oriented to face light absorbing surfaces of the photobioreactor.

55. The photobioreactor of claim 1 wherein said liquid medium comprises microorganisms and nutrients for the development of said microorganisms and for the production of biomass by photosynthesis.

56. The photobioreactor of claim 55 wherein said microorganisms comprise microalgae.

57. The photobioreactor of claim 55 wherein said microorganisms comprise genetically modified microalgae.

58. The photobioreactor of claim 55 wherein said microorganisms comprise sterile microalgae.

59. The photobioreactor of claim 55 wherein said microorganisms comprise sterile genetically modified microalgae.

60. The photobioreactor of claim 55 wherein said microorganisms comprise mixtures of microalgae.

61. The photobioreactor of claim 55 wherein said microorganisms comprise mixtures of genetically modified microalgae.

62. The photobioreactor of claim 55 wherein said microorganisms comprise sterile mixtures of microalgae.

63. The photobioreactor of claim 55 wherein said microorganisms comprise sterile mixtures of genetically modified microalgae.

64. The photobioreactor of claim 55 wherein said microorganisms comprise mixtures of microalgae with other microorganisms.



65. The photobioreactor of claim 55 wherein said microorganisms comprise mixtures of microalgae with bacteria and/or archaea.
66. The photobioreactor of claim 65 wherein said microalgae have a coexistence with said bacteria and/or archaea.
67. The photobioreactor of claim 65 wherein said bacteria and/or archaea are present in an amount sufficient to reduce or eliminate microalgae waste in the photobioreactor.
68. The photobioreactor of claim 55 wherein said microorganisms comprise mixtures of genetically modified microalgae with other microorganisms.
69. The photobioreactor of claim 55 wherein said microorganisms comprise mixtures of genetically modified microalgae with bacteria and/or archaea.
70. The photobioreactor of claim 69 wherein said genetically modified microalgae have a coexistence with said bacteria and/or archaea.
71. The photobioreactor of claim 69 wherein said bacteria and/or archaea are present in an amount sufficient to reduce or eliminate genetically modified microalgae waste in the photobioreactor.
72. The photobioreactor of claim 55 wherein said microorganisms comprise sterile mixtures of microalgae with other microorganisms.
73. The photobioreactor of claim 55 wherein said microorganisms comprise sterile mixtures of microalgae with bacteria and/or archaea, wherein said bacteria and/or archaea are selected and added to said sterile mixtures in an amount sufficient to reduce or eliminate microalgae waste in said photobioreactor.

74. The photobioreactor of claim 73 wherein said microalgae have a coexistence with said bacteria and/or archaea.
75. The photobioreactor of claim 73 wherein microalgae cell density is at least about 40 grams per liter.
76. The photobioreactor of claim 55 wherein said microorganisms comprise sterile mixtures of genetically modified microalgae with other microorganisms.
77. The photobioreactor of claim 55 wherein said microorganisms comprise sterile mixtures of genetically modified microalgae with bacteria and/or archaea, wherein said bacteria and/or archaea are selected and added to said sterile mixtures in an amount sufficient to reduce or eliminate genetically modified microalgae waste in said photobioreactor.
78. The photobioreactor of claim 77 wherein said genetically modified microalgae have a coexistence with said bacteria and/or archaea.
79. The photobioreactor of claim 77 wherein genetically modified microalgae cell density is at least about 20 grams per liter.
80. The photobioreactor of claim 56 wherein said microalgae comprises marine or fresh water microalgae or genetic modifications thereof.
81. The photobioreactor of claim 80 wherein said marine microalgae is selected from *Skeletonema*, *Chaetoceros*, *Thalassiosira*, *Tetraselmis*, *Isochrysis*, and *Nanochloropsis*.
82. The photobioreactor of claim 80 wherein said fresh water microalgae is selected from *Haematococcus*, *Chlamydomonas*, and *Spirulina*.

83. The photobioreactor of claim 56 wherein said microalgae comprises cyanobacteria, green algae, or diatoms.

84. The photobioreactor of claim 56 wherein said microalgae comprises cyanobacteria species selected from *microcystis*, green algae selected from *Chlorella*, *Botryococcus*, *Ankistrodesmus*, *Chlamydomonas*, and *Dunaliella*, and diatoms selected from *Thalassiosira* and *Navicula*.

85. The photobioreactor of claim 1 wherein said liquid medium comprises autotrophic or mixotrophic microorganisms.

86. The photobioreactor of claim 1 which is connected in series or in parallel to one or more other photobioreactors.

87. A photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more product outlet openings at said bottom portion for removing at least product from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container.

88. The photobioreactor of claim 87 further comprising one or more air inlet openings at said bottom portion for introducing at least air into said container.

89. The photobioreactor of claim 87 wherein said bottom portion contains two or more parallel gas spargers for introducing gas into said parallel passageways.

90. A method for culturing and harvesting microorganisms, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more microorganism culture outlet openings at said bottom portion for removing microorganism culture from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases, through said parallel passageways;

- f) allowing said microorganisms to grow to a desired yield;
- g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings; and
- h) harvesting a microorganism culture from said container via said one or more microorganism culture outlet openings.

91. The method of claim 90 wherein said container of said photobioreactor is made from a plastic material.

92. The method of claim 90 wherein said container of said photobioreactor is non-rigid, semi-rigid or rigid.

93. The method of claim 90 wherein said container of said photobioreactor further comprises one or more air inlet openings at said bottom portion for introducing at least air into said container.

94. The method of claim 93 further comprising providing said air into said container via said one or more air inlet openings.

95. The method of claim 91 wherein said plastic material is selected from polyethylene, polypropylene, polycarbonate, polyvinylchloride, polyester and laminates thereof.

96. The method of claim 91 wherein said plastic material comprises a laminate of polyethylene and nylon.

97. The method of claim 90 wherein said photobioreactor comprises a flat panel photobioreactor.

98. The method of claim 90 wherein said bottom portion of said photobioreactor contains one or more gas spargers for introducing gas into said parallel passageways.

99. The method of claim 90 wherein said bottom portion of said photobioreactor contains two or more parallel gas spargers for introducing gas into said parallel passageways.

100. The method of claim 98 wherein said one or more gas spargers comprise at least one gas inlet line having a plurality of secondary gas inlet lines protruding therefrom, said secondary gas inlet lines extending into inlet openings in at least a portion of said parallel passageways of said photobioreactor for introducing gas into said parallel passageways.

101. The method of claim 90 wherein said bottom portion of said photobioreactor contains diffusers for introducing carbon dioxide into said parallel passageways.

102. The method of claim 90 wherein at least a portion of said parallel passageways of said photobioreactor (risers) are configured for the flow of said liquid medium from said bottom portion to said top portion to achieve recycle of medium.

103. The method of claim 90 wherein at least a portion of said parallel passageways of said photobioreactor (down comers) are configured for the flow of said liquid medium from said top portion to said bottom portion.

104. The method of claim 90 wherein said photobioreactor has one or more groupings of risers and down comers in which one grouping comprises at least about 4 risers and 1 down comer.

105. The method of claim 104 wherein said photobioreactor has from about 4 to about 6 groupings of risers and down comers.

106. The method of claim 104 wherein the internal diameter of said down comers is greater than the internal diameter of said risers.

107. The method of claim 91 wherein said container of said photobioreactor is formed by fusion bonding two sheets of said plastic material along predetermined seams to provide at least said interior volume, said interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of said liquid medium.

108. The method of claim 91 wherein said container of said photobioreactor is formed by fusion bonding three sheets of said plastic material along predetermined seams to provide at least said interior volume and a second interior volume, said interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of said liquid medium, and said second interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a heat transferring fluid.

109. The method of claim 108 wherein said second interior volume of said photobioreactor is a heat exchanger for circulating heat transferring fluid therethrough.

110. The method of claim 108 wherein said interior volume and said second interior volume of said photobioreactor have opposed top portions, bottom portions and parallel passageways.

111. The method of claim 110 in which the opposing parallel passageways in horizontal cross section comprise a series of parallel crests intercalated with troughs along the width of said container, wherein pairs of opposed troughs are joined at an inner wall, thereby defining a series of adjacent parallel passageways in said interior volume and said second interior volume of said photobioreactor.

112. The method of claim 108 further comprising altering volume of the parallel passageways of said interior volume using pressure resulting from heat transferring fluid in opposed parallel passageways of said second interior volume.

113. The method of claim 91 wherein said container of said photobioreactor is formed by fusion bonding four sheets of said plastic material along predetermined seams to provide at least said interior volume, a second interior volume and a third interior volume, said second interior volume positioned between said interior volume and said third interior volume, said interior volume and said third interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of said liquid medium, and said second interior volume having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a heat transferring fluid.

114. The method of claim 113 wherein said second interior volume of said photobioreactor is a heat exchanger for circulating heat transferring fluid therethrough.

115. The method of claim 113 wherein said interior volume, said second interior volume and said third interior volume of said photobioreactor have opposed top portions, bottom portions and parallel passageways.



116. The method of claim 115 in which the opposing parallel passageways in horizontal cross section comprise a series of parallel crests intercalated with troughs along the width of said container, wherein pairs of opposed troughs are joined at an inner wall, thereby defining a series of adjacent parallel passageways in said interior volume, said second interior volume and said third interior volume of said photobioreactor.

117. The method of claim 113 further comprising altering volume of the parallel passageways of said interior volume and said third interior volume using pressure resulting from heat transferring fluid in opposed parallel passageways of said second interior volume.

118. The method of claim 90 further comprising a liquid medium feed line connected to said liquid medium inlet opening of said photobioreactor, said liquid medium feed line extending from the liquid medium inlet opening exteriorly from said bottom portion for delivery of liquid medium into said interior volume, the liquid medium feed line optionally containing one or more liquid medium flow control valves therein for control of flow of the liquid medium therethrough.

119. The method of claim 118 wherein said liquid medium feed line is connected to a liquid medium supply source.

120. The method of claim 90 further comprising a microorganism culture discharge line connected to said microorganism culture outlet opening of said photobioreactor, said microorganism culture discharge line extending from the microorganism culture outlet opening exteriorly from said bottom portion for removal of microorganism culture from said interior volume, the microorganism culture discharge line optionally containing one or more microorganism culture discharge control valves therein for control of flow of the microorganism culture therethrough.

121. The method of claim 120 wherein said microorganism culture discharge line is connected to a collector.

122. The method of claim 90 further comprising a gas feed line connected to said gas inlet opening of said photobioreactor, said gas feed line extending from the gas inlet opening exteriorly from said bottom portion for delivery of gas into said interior volume, the gas feed line optionally containing one or more gas flow control valves therein for control of flow of the gas therethrough.

123. The method of claim 122 wherein said gas feed line is connected to a gas supply source.

124. The method of claim 123 wherein said gas supply source is sequestered carbon dioxide, treated stack gas, chemical plant off gas, or a non-toxic waste gas.

125. The method of claim 90 further comprising an air feed line connected to said air inlet opening of said photobioreactor, said air feed line extending from the air inlet opening exteriorly from said bottom portion for delivery of air into said interior volume, the air feed line optionally containing one or more air flow control valves therein for control of flow of the air therethrough.

126. The method of claim 125 wherein said air feed line is connected to an air supply source.

127. The method of claim 126 wherein said air supply source is air, treated stack gas, chemical plant off gas, or a non-toxic waste gas.

128. The method of claim 90 further comprising a gas discharge line connected to said gas outlet opening of said photobioreactor, said gas discharge line extending from the gas outlet opening exteriorly from said top portion for removal of gas from said interior volume, the gas discharge line optionally containing one

or more gas discharge control valves therein for control of flow of the gas therethrough.

129. The method of claim 128 wherein said gas discharge line is connected to a collector.

130. The method of claim 128 wherein molecular oxygen is recovered.

131. The method of claim 90 in which said photobioreactor further comprises a collector for collecting microorganism culture discharged from said interior volume.

132. The method of claim 90 in which said photobioreactor further comprises sensors for measuring a number of parameters that are representative of the growth of said microorganism culture, and a control unit connected to the sensors and at least one of the liquid medium inlet openings, gas inlet openings and air inlet openings for introducing liquid medium, gas and air in order to stimulate growth of the microorganism culture in accordance with the measured parameter values.

133. The method of claim 90 in which said photobioreactor further comprises a computer implemented system for controlling at least one of light exposure, media flow rates, gas exchange rates, internal turbulence, orientation of said photobioreactor in respect to the sun or other light source, heating and cooling of said photobioreactor, mixing, and harvesting of biomass.

134. The method of claim 100 wherein introducing gas into said parallel passageways creates a turbulence in said liquid medium sufficient to prevent sedimentation of microorganisms, limit exposure of individual cells to light intensity, and prevent adherence of microorganisms to walls of said parallel passageways.

135. The method of claim 100 wherein introducing gas into said parallel passageways is conducted at a flow rate of sufficient volume to create laminar or turbulent flow of said liquid medium in said parallel passageways.

136. The method of claim 100 wherein introducing gas into said parallel passageways creates a turbulence in said liquid medium having a Reynolds number of from about 500 to about 4000.

137. The method of claim 134 wherein turbulence in said liquid medium is controlled by airflow in said parallel passageways of said photobioreactor.

138. The method of claim 134 wherein exposure of individual microorganism cells to light intensity is controlled by controlling internal turbulence in said parallel passageways.

139. The method of claim 90 wherein said front wall and said rear wall of said photobioreactor allow sufficient light to enter said liquid medium for the photocultivation of microalgae.

140. The method of claim 139 wherein said light can be natural or artificial light.

141. The method of claim 139 wherein said light can be reflective or refractive light.

142. The method of claim 90 in which said photobioreactor further comprises a support component capable of supporting said photobioreactor.

143. The method of claim 142 in which the support component is configured to tilt the photobioreactor based on the position of the sun or other light source.

144. The method of claim 140 in which artificial light can be oriented to face light absorbing surfaces of the photobioreactor.
145. The method of claim 90 wherein said liquid medium comprises microorganisms and nutrients for the development of said microorganisms and for the production of biomass by photosynthesis.
146. The method of claim 145 wherein said microorganisms comprise microalgae.
147. The method of claim 145 wherein said microorganisms comprise genetically modified microalgae.
148. The method of claim 145 wherein said microorganisms comprise sterile microalgae.
149. The method of claim 145 wherein said microorganisms comprise sterile genetically modified microalgae.
150. The method of claim 145 wherein said microorganisms comprise mixtures of microalgae.
151. The method of claim 145 wherein said microorganisms comprise mixtures of genetically modified microalgae.
152. The method of claim 145 wherein said microorganisms comprise sterile mixtures of microalgae.
153. The method of claim 145 wherein said microorganisms comprise sterile mixtures of genetically modified microalgae.

154. The method of claim 145 wherein said microorganisms comprise mixtures of microalgae with other microorganisms.
155. The method of claim 145 wherein said microorganisms comprise mixtures of microalgae with bacteria and/or archaea.
156. The method of claim 155 wherein said microalgae have a coexistence with said bacteria and/or archaea.
157. The method of claim 155 wherein said bacteria and/or archaea are present in an amount sufficient to reduce or eliminate microalgae waste in the photobioreactor.
158. The method of claim 145 wherein said microorganisms comprise mixtures of genetically modified microalgae with other microorganisms.
159. The method of claim 145 wherein said microorganisms comprise mixtures of genetically modified microalgae with bacteria and/or archaea.
160. The method of claim 159 wherein said genetically modified microalgae have a coexistence with said bacteria and/or archaea.
161. The method of claim 159 wherein said bacteria and/or archaea are present in an amount sufficient to reduce or eliminate genetically modified microalgae waste in the photobioreactor.
162. The method of claim 145 wherein said microorganisms comprise sterile mixtures of microalgae with other microorganisms.
163. The method of claim 145 wherein said microorganisms comprise sterile mixtures of microalgae with bacteria and/or archaea, wherein said bacteria and/or

archaea are selected and added to said sterile mixtures in an amount sufficient to reduce or eliminate microalgae waste in said photobioreactor.

164. The method of claim 163 wherein said microalgae have a coexistence with said bacteria and/or archaea.

165. The method of claim 163 wherein microalgae cell density is at least about 20 grams per liter.

166. The method of claim 145 wherein said microorganisms comprise sterile mixtures of genetically modified microalgae with other microorganisms.

167. The method of claim 145 wherein said microorganisms comprise sterile mixtures of genetically modified microalgae with bacteria and/or archaea, wherein said bacteria and/or archaea are selected and added to said sterile mixtures in an amount sufficient to reduce or eliminate genetically modified microalgae waste in said photobioreactor.

168. The method of claim 167 wherein said genetically modified microalgae have a coexistence with said bacteria and/or archaea.

169. The method of claim 167 wherein genetically modified microalgae cell density is at least about 20 grams per liter.

170. The method of claim 146 wherein said microalgae comprises marine or fresh water microalgae or genetic modifications thereof.

171. The method of claim 170 wherein said marine microalgae is selected from *Skeletonema*, *Chaetoceros*, *Thalassiosira*, *Tetraselmis*, *Isochrysis*, and *Nanochloropsis*.

172. The method of claim 170 wherein said fresh water microalgae is selected from *Haematococcus*, *Chlamydomonas*, and *Spirulina*.

173. The method of claim 146 wherein said microalgae comprises cyanobacteria, green algae, or diatoms.

174. The method of claim 146 wherein said microalgae comprises cyanobacteria species selected from *microcystis*, green algae selected from *Chlorella*, *Botryococcus*, *Ankistrodesmus*, *Chlamydomonas*, and *Dunaliella*, and diatoms selected from *Thalassiosira* and *Navicula*.

175. The method of claim 90 wherein said liquid medium comprises autotrophic or mixotrophic microorganisms.

176. The method of claim 90 wherein said photobioreactor is connected in series or in parallel to one or more other photobioreactors.

177. The method of claim 90 wherein the density of said microorganism culture in said photobioreactor can range from about 0.2 grams per liter (dry basis) to about 40 grams per liter (dry basis).

178. The method of claim 90 wherein the flow rate of said at least carbon dioxide gas and optionally other gases in said photobioreactor can range from about 0.1 percent volume of inlet gas flow to about 5 percent volume of inlet gas flow.

179. The method of claim 90 wherein the flow rate of said gas in said photobioreactor flow rate is of sufficient volume to create turbulent flow of said liquid medium in said parallel passageways.



180. The method of claim 90 which is conducted in batch, semi-batch or continuous mode.
181. The method of claim 90 further comprising allowing said microorganisms to grow under sterile conditions.
182. The method of claim 90 comprising growing heterotrophic and/or mixotrophic microorganisms under autotrophic conditions sufficient for said heterotrophic and/or mixotrophic microorganisms to adopt autotrophic characteristics.
183. The method of claim 90 comprising growing autotrophic microorganisms under mixotrophic conditions.
184. The method of claim 90 comprising growing autotrophic microorganisms under autotrophic conditions.
185. The method of claim 90 further comprising using said microorganism culture in biofuel production, aquaculture food production, mammalian food production, and/or recombinant protein synthesis.
186. The method of claim 185 wherein biofuel production comprises biodiesel, jet fuel or heating oil production.
187. The method of claim 185 wherein recombinant protein synthesis comprises pharmaceutical and industrial enzyme synthesis.
188. The method of claim 90 wherein said microorganism culture comprises microalgae.

189. The method of claim 90 wherein said microorganism culture comprises genetically modified microalgae.
190. The method of claim 90 wherein said microorganism culture comprises sterile microalgae.
191. The method of claim 90 wherein said microorganism culture comprises sterile genetically modified microalgae.
192. The method of claim 90 wherein said microorganism culture comprises mixtures of microalgae.
193. The method of claim 90 wherein said microorganism culture comprises mixtures of genetically modified microalgae.
194. The method of claim 90 wherein said microorganism culture comprises sterile mixtures of microalgae.
195. The method of claim 90 wherein said microorganism culture comprises sterile mixtures of genetically modified microalgae.
196. The method of claim 90 wherein said microorganism culture comprises mixtures of microalgae with other microorganisms.
197. The method of claim 90 wherein said microorganism culture comprises mixtures of microalgae with bacteria and/or archaea.
198. The method of claim 197 wherein said microalgae have a coexistence with said bacteria and/or archaea.

199. The method of claim 197 wherein said bacteria and/or archaea are present in an amount sufficient to reduce or eliminate microalgae waste in the photobioreactor.

200. The method of claim 90 wherein said microorganism culture comprises mixtures of genetically modified microalgae with other microorganisms.

201. The method of claim 90 wherein said microorganism culture comprises mixtures of genetically modified microalgae with bacteria and/or archaea.

202. The method of claim 201 wherein said genetically modified microalgae have a coexistence with said bacteria and/or archaea.

203. The method of claim 201 wherein said bacteria and/or archaea are present in an amount sufficient to reduce or eliminate genetically modified microalgae waste in the photobioreactor.

204. The method of claim 90 wherein said microorganism culture comprises sterile mixtures of microalgae with other microorganisms.

205. The method of claim 90 wherein said microorganism culture comprises sterile mixtures of microalgae with bacteria and/or archaea, wherein said bacteria and/or archaea are selected and added to said sterile mixtures in an amount sufficient to reduce or eliminate microalgae waste in said photobioreactor.

206. The method of claim 205 wherein said microalgae have a coexistence with said bacteria and/or archaea.

207. The method of claim 205 wherein microalgae cell density is at least about 20 grams per liter.

208. The method of claim 90 wherein said microorganism culture comprises sterile mixtures of genetically modified microalgae with other microorganisms.
209. The method of claim 90 wherein said microorganism culture comprises sterile mixtures of genetically modified microalgae with bacteria and/or archaea, wherein said bacteria and/or archaea are selected and added to said sterile mixtures in an amount sufficient to reduce or eliminate genetically modified microalgae waste in said photobioreactor.
210. The method of claim 209 wherein said genetically modified microalgae have a coexistence with said bacteria and/or archaea.
211. The method of claim 209 wherein genetically modified microalgae cell density is at least about 40 grams per liter.
212. The method of claim 188 wherein said microalgae comprises marine or fresh water microalgae or genetic modifications thereof.
213. The method of claim 212 wherein said marine microalgae is selected from *Skeletonema*, *Chaetoceros*, *Thalassiosira*, *Tetraselmis*, *Isochrysis*, and *Nanochloropsis*.
214. The method of claim 212 wherein said fresh water microalgae is selected from *Haematococcus*, *Chlamydomonas*, and *Spirulina*.
215. The method of claim 90 wherein said liquid medium comprises autotrophic or mixotrophic microorganisms.
216. A method of producing biomass, said method comprising:  
a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the production of said biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings; and

h) removing said biomass from said container via said one or more biomass outlet openings.

217. The method of claim 216 further comprising using said biomass in biofuel production, aquaculture food production, mammalian food production, and/or recombinant protein synthesis.

218. A method for producing a biofuel from biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

- g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;
- h) removing said biomass from said container via said one or more biomass outlet openings; and
- i) converting said biomass to said biofuel.

219. The method of claim 218 wherein said biofuel comprises biodiesel, jet fuel or heating oil.

220. A method for producing and recovering a recombinant protein from biomass, said method comprising:

- a) providing a photobioreactor comprising:
  - a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;
  - said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;
  - one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;
  - one or more biomass outlet openings at said bottom portion for removing biomass from said container;
  - one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and
  - one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;
- b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

- c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;
- d) exposing said photobioreactor to a light source;
- e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;
- f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;
- g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;
- h) removing said biomass from said container via said one or more biomass outlet openings; and
- i) producing and recovering said recombinant protein from said biomass.

221. The method of claim 220 wherein said recombinant protein comprises an antibody, antigen, hormone, or enzyme.

222. A method for producing aquaculture food from biomass, said method comprising:

- a) providing a photobioreactor comprising:
  - a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;
  - said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;
  - one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;
  - one or more biomass outlet openings at said bottom portion for removing biomass from said container;



one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;

h) removing said biomass from said container via said one or more biomass outlet openings; and

i) converting said biomass to said aquaculture food.

223. A method for producing mammalian food from biomass, said method comprising:

a) providing a photobioreactor comprising:

a container having at least a front wall, a rear wall and an interior volume, wherein at least a portion of said front wall and said rear wall is transparent to light;

said container having at least a top portion, a bottom portion, and a plurality of parallel passageways extending from said bottom portion to said top portion for the throughflow of a liquid medium;

one or more liquid medium inlet openings at said bottom portion for introducing at least liquid medium into said container;

one or more biomass outlet openings at said bottom portion for removing biomass from said container;

one or more gas inlet openings at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container; and

one or more gas outlet openings at said top portion for removing at least one of excess gas and waste gases from said container;

b) providing said liquid medium into said container via said one or more liquid medium inlet openings, wherein said liquid medium contains microorganisms and nutrients for the growth of said microorganisms and for the production of biomass;

c) providing said at least carbon dioxide gas and optionally other gases into said container via said one or more gas inlet openings;

d) exposing said photobioreactor to a light source;

e) circulating said liquid medium and said at least carbon dioxide gas and optionally other gases through said parallel passageways;

f) allowing said microorganisms to grow to a desired yield and said biomass to be produced to a desired amount;

g) allowing said at least one of excess gas and/or waste gases to leave said container continuously via said one or more gas outlet openings;

h) removing said biomass from said container via said one or more biomass outlet openings; and

i) converting said biomass to said mammalian food.

FIG. 1

Front Layer - Culture Chamber

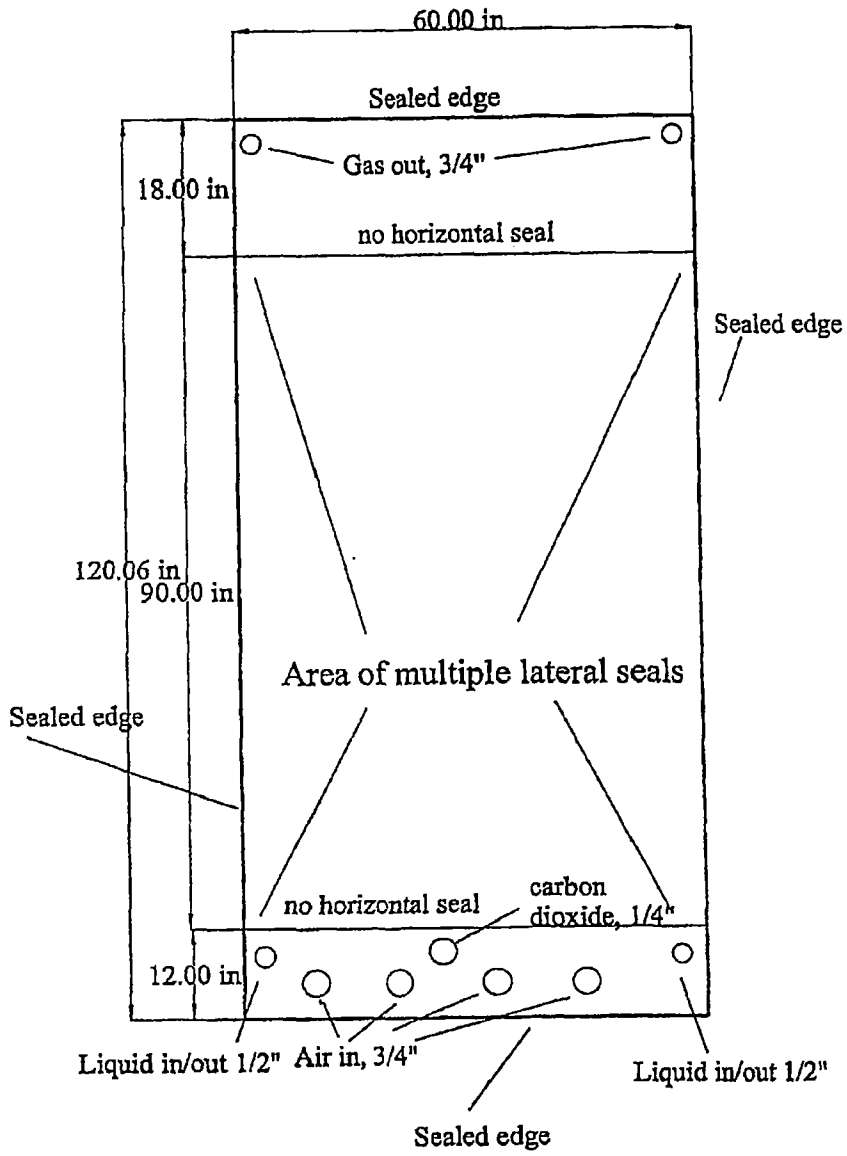


FIG. 2

Back Layer - Heat Exchanger

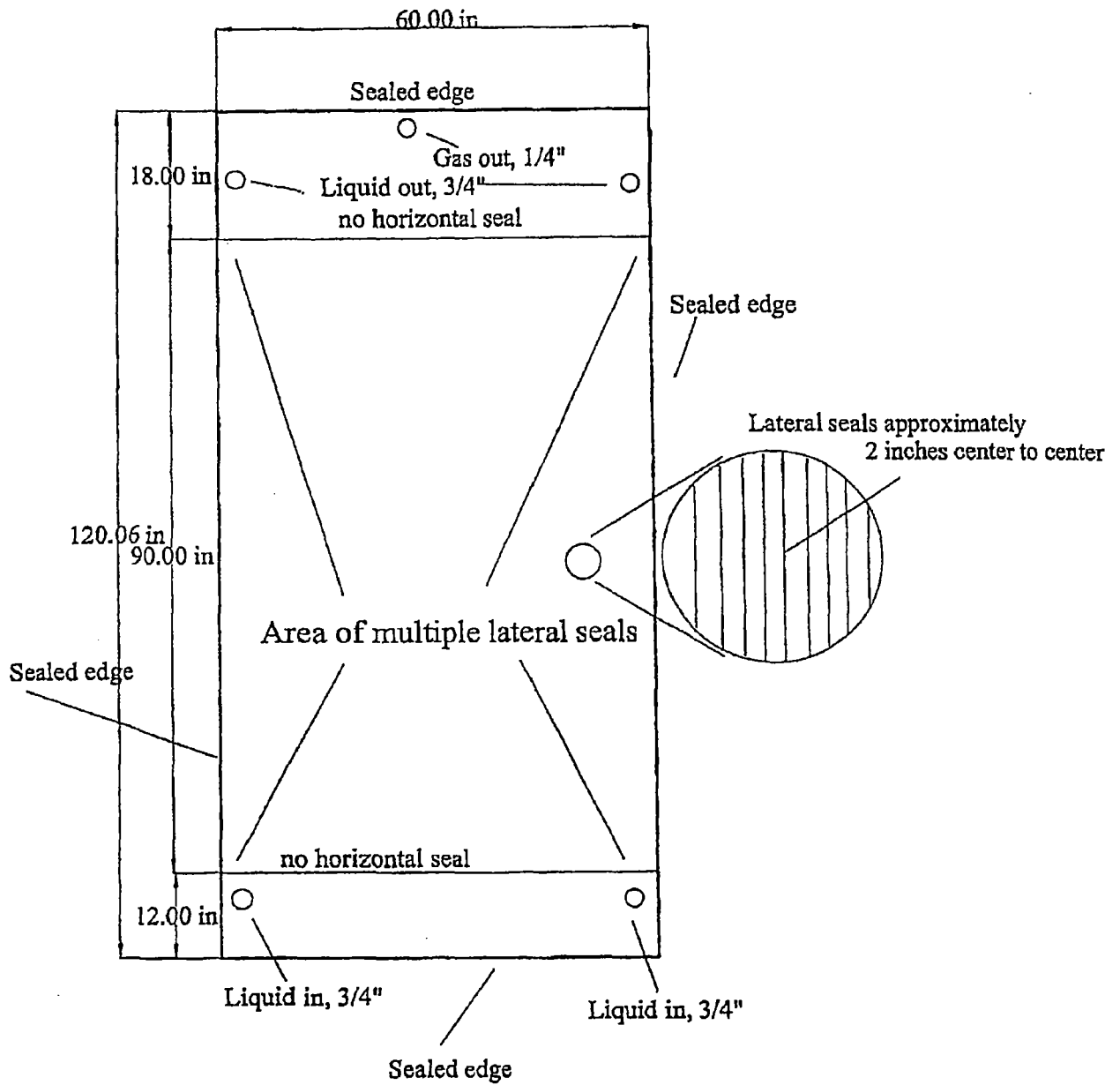


FIG. 3

Front Layer -Culture Chamber  
with internal view of sparger  
and verticle channels

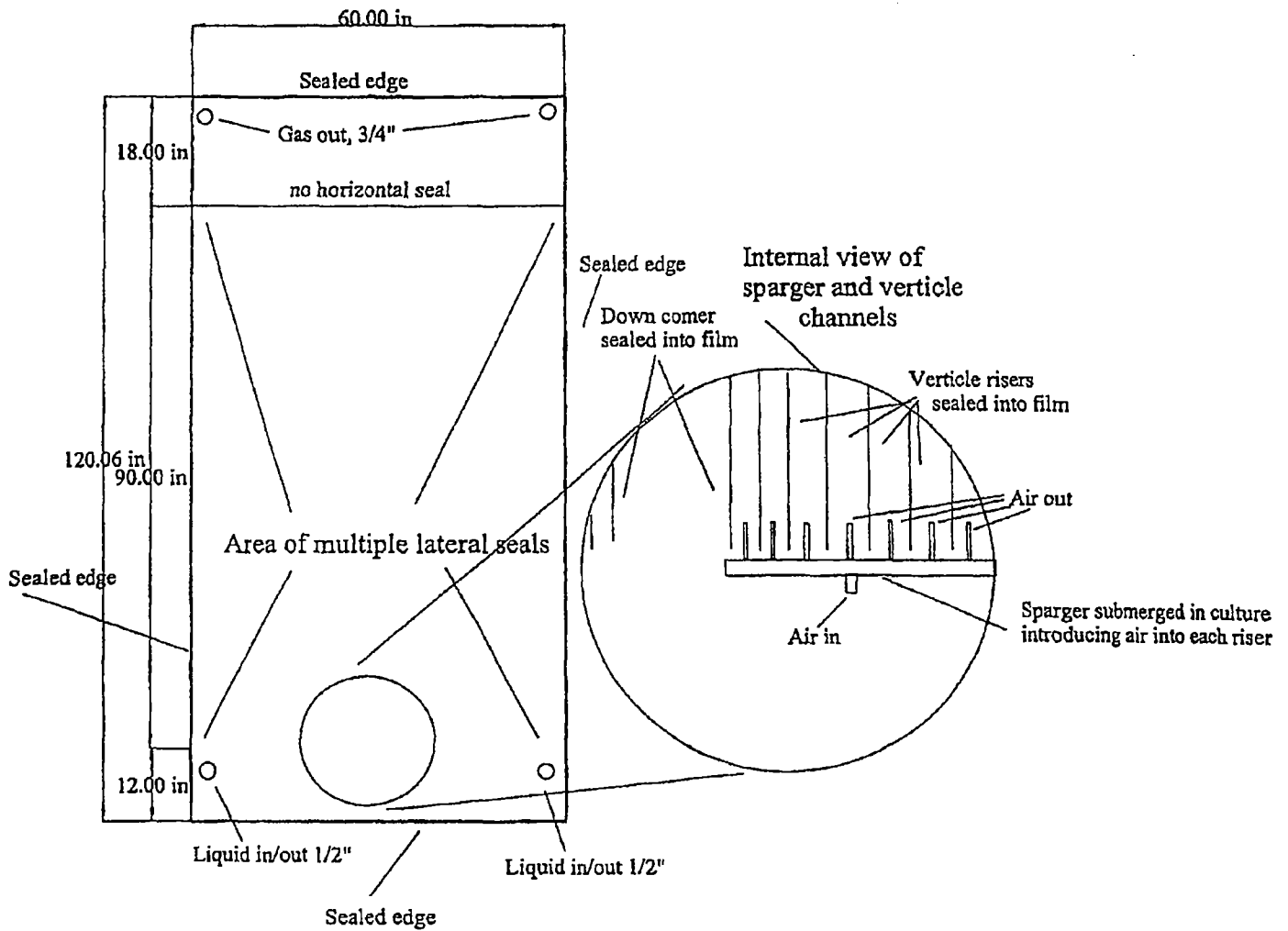


FIG. 4

Internal Flows of Photobioreactor

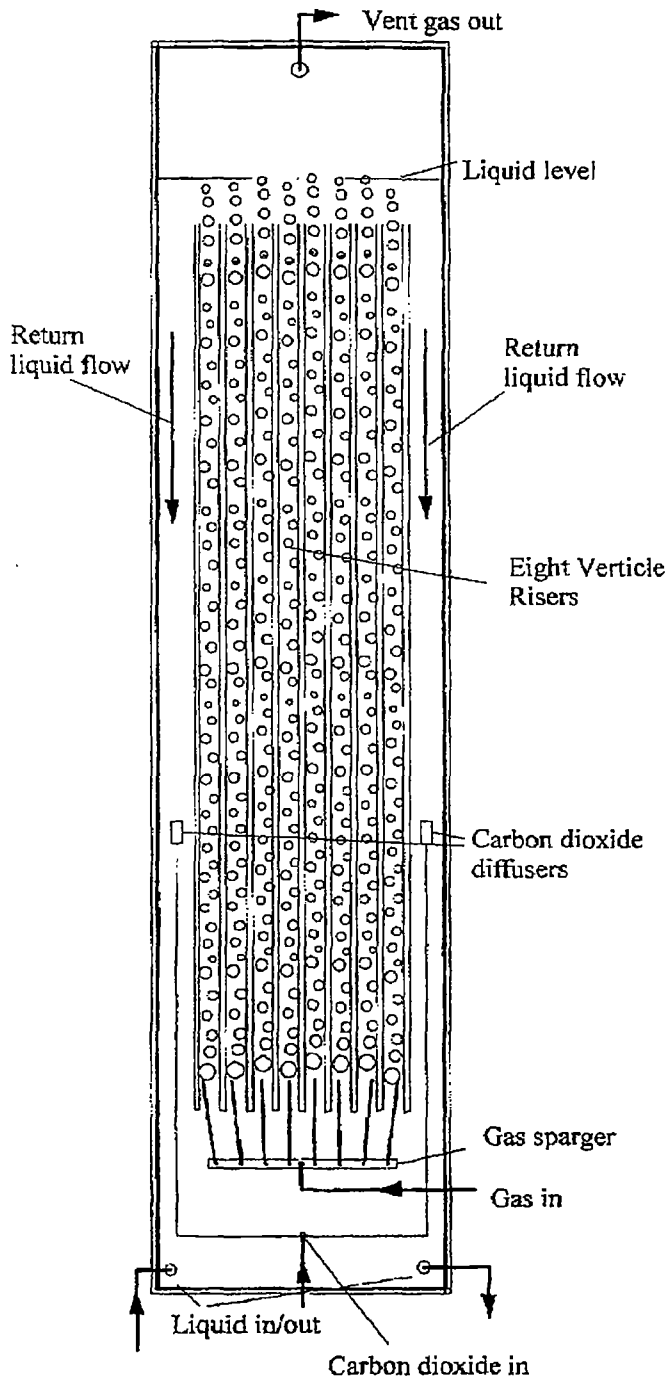


FIG. 5

Effect of Gas Flow on Reynolds Number

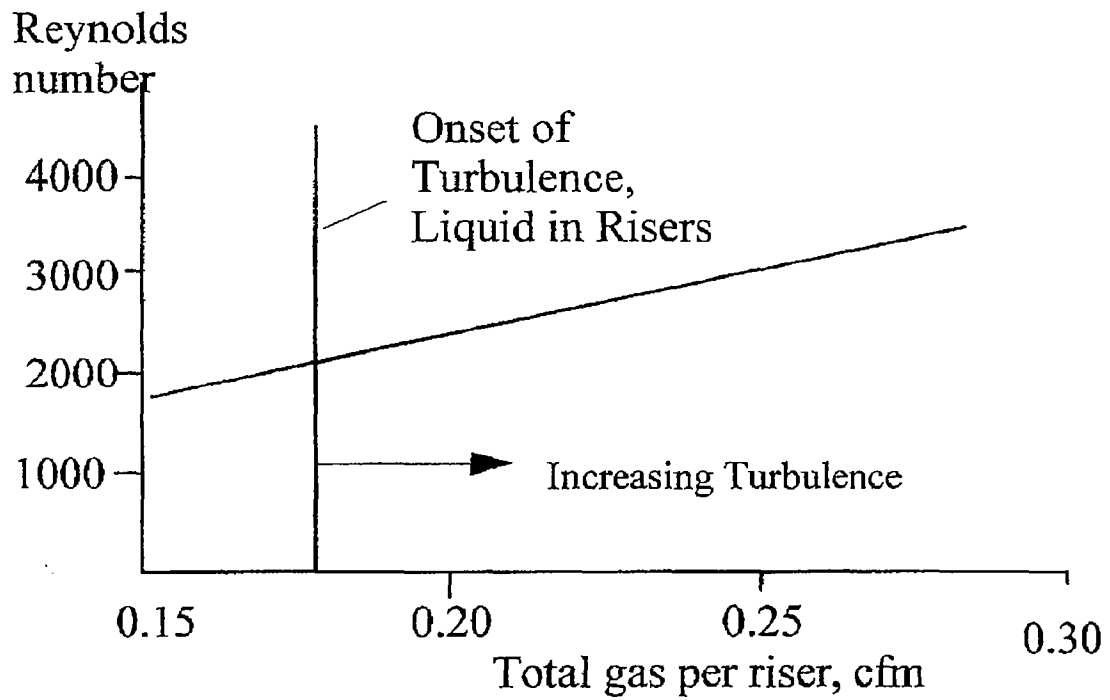
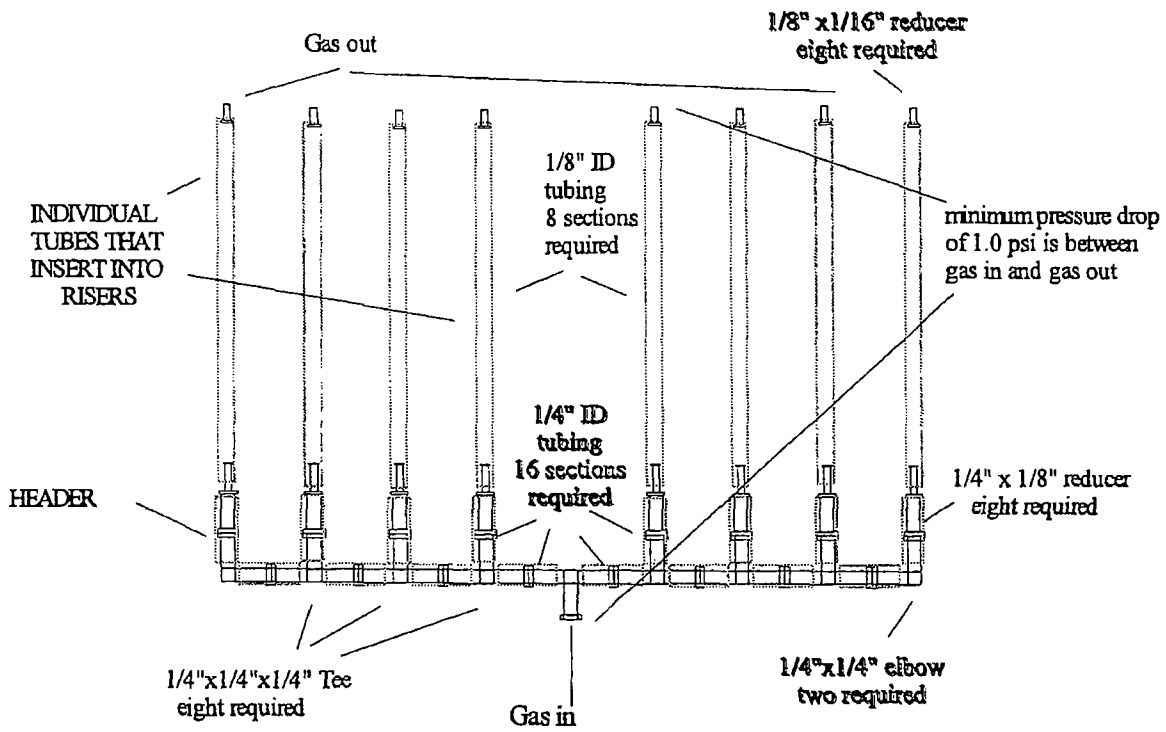


FIG. 6

Sparger Assembly





## INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 10/36178

<b>A. CLASSIFICATION OF SUBJECT MATTER</b> IPC(8) - C12C 1/15 (2010.01) USPC - 435/292.1 According to International Patent Classification (IPC) or to both national classification and IPC		
<b>B. FIELDS SEARCHED</b> Minimum documentation searched (classification system followed by classification symbols) IPC(8) - C12C 1/15 (2010.01) USPC - 435/292.1 Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched USPC - 435/292.1; 428/36.6; 435/289.1; 435/257.1 (Text Search) Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) PubWEST (PGPB, USPT, USOC, EPAB, JPAB); Google Scholar. Search Terms: photobioreactor, photo-bioreactor, bioreactor, gas, CO <sub>2</sub> , carbon dioxide, inlet, outlet, riser, down comer, air, polyethylene, nylon, laminat\$, wavelength, select, select\$, surface, filter, flat, panel, recycle, recirculat\$, throughflow, through flow,		
<b>C. DOCUMENTS CONSIDERED TO BE RELEVANT</b>		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 2008/0286851 A1 (WHITTON) 20 November 2008 (20.11.2008) fig 1A, 5B, 10, 12; para [0005], [0010]-[0014], [0016]-[0020], [0022], [0040], [0041], [0045], [0064], [0070], [0071], [0078], [0079], [0080], [0087], [0088], [0092].	1-89
Y	US 2008/0160591 A1 (WILLSON et al.) 03 July 2008 (03.07.2008) fig 1; para [0009], [0017], [0039], [0045], [0046], [0057], [0058], [0064], [0074], [0075], [0079], [0081]-[0083], [0089], [0096], [0120], [0140], [0141], [0143]-[0145].	1-89
Y	US 2003/0059932 A1 (CRAIGIE et al.) 27 March 2003 (27.03.2003) fig 1; claim 1; para [0015], [0025]-[0027].	4, 29-40 and 88
Y	US 2008/0017558 A1 (POLLOCK et al.) 24 January 2008 (24.01.2008) para [0122], [0123], [0142].	13-17
Y	US 2005/0260553 A1 (BERZIN) 24 November 2005 (24.11.2005) para [0016], [0059].	44 and 47-48
Y	US 5,137,828 A (ROBINSON et al.) 11 August 1992 (11.08.1992) col 1, ln 14-23; col 2, ln 5-54.	51
Y	US 2009/0011492 A1 (BERZIN) 08 January 2009 (08.01.2009) para [0100], [0116].	52-54
Y	US 2007/0048848 A1 (SEARS) 01 March 2007 (01.03.2007) para [0007]-[0010], [0061].	64-79
Y	US 2003/0228684 A1 (BURBIDGE et al.) 11 December 2003 (11.12.2003) para [0034].	86
<input type="checkbox"/> Further documents are listed in the continuation of Box C. <input type="checkbox"/>		
* Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family		
Date of the actual completion of the international search 30 September 2010 (30.09.2010)		Date of mailing of the international search report <b>06 OCT 2010</b>
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-3201		Authorized officer: Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.  
PCT/US 10/36178

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

- 1.  Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:
  
- 2.  Claims Nos.:  
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
  
- 3.  Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:  
This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1.

- Group I: Claims 1-89, directed to a photobioreactor.
- Group II: Claims 90-215, directed to a method for culturing and harvesting microorganisms.
- Group III: Claims 216-217, directed to a method of producing a biomass.
- Group IV: Claims 218-219, directed to a method of producing a biofuel.

.....Continued in Supplemental  
Box.....

- 1.  As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
- 2.  As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.
- 3.  As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
  
- 4.  No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:  
1-89

- Remark on Protest
- The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
  - The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
  - No protest accompanied the payment of additional search fees.

Continuation of:

Box No. III - Observations where unity of invention is lacking:

Group V: Claims 220-221, directed to a method for producing and recovering a recombinant protein.

Group VI: Claim 222, directed to a method for producing aquaculture food.

Group VII: Claim 223, directed to a method for producing mammalian food.

The groups of inventions listed above do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

Groups II - VII represent separate methods of use for the photobioreactor in Group I. In accordance with ISPE Guidelines sections 10.12 - 10.16, unity of invention under PCT Rule 13 permits the inclusion of any one combination of an independent claim for a given product, an independent claim for a process specially adapted for the manufacture of the said product, and an independent claim for a use of the said product. In this case, multiple combinations are claimed through the recitation of multiple methods of use.

Further, a single general inventive concept must link the claims in the various categories. However, the claims of Groups I - VII are not linked by a single general inventive concept.

The technical features shared by the groups are those of the photobioreactor of claim 1. These shared technical features fail to make a contribution over the prior art, as exemplified by US 2008/0286851 A1 (Whitton).

Regarding claim 1, Whitton discloses a photobioreactor for culturing and harvesting at least one of a microorganism culture (para [0010]-[0013]), said photobioreactor comprising:

—a container having at least a front wall, a rear wall and an interior volume (fig 12, 1A, para [0070], [0092]), wherein at least a portion of said front wall is transparent to light (para [0022], [0070], [0080]);

—said container having at least a top portion, a bottom portion and a plurality of parallel passageway extending from said bottom portion to said top portion for flow of a liquid medium (fig 10a, 12, part 89a; para [0010], [0071], [0079], [0087], [0092]);

—one or more liquid medium inlet opening at said bottom portion for introducing at least liquid medium into said container (fig 12, part 130; para [0010], [0017], [0018], [0022], [0092]-"liquid fill line");

—one or more microorganism culture outlet openings at said bottom portion for removing microorganism culture from said container (fig 12, part 130; para [0010], [0017], [0018], [0022], [0092]-"liquid drain line");

—one or more gas inlet opening at said bottom portion for introducing at least carbon dioxide gas and optionally other gases into said container (fig 12, part 126, part; para [0022], [0040], [0092]); and

one or more gas outlet opening at said top portion for removing at least one of excess gas and waste gas from said container (fig 12, part 122; para [0016], [0022], [0092]).

Whitton does not expressly disclose that at least a portion of said rear wall is transparent to light, however, this would have been obvious to one of ordinary skill in the art in view of Whitton that discloses that at least one of the front, back and side walls of the photobioreactor is transparent (fig 5B, part 42gg; para [0022], [0070], [0080]), because one of ordinary skill in the art would have been able to choose without undue experimentation the proper wall(s) that should be transparent.

Thus, the groups of claims listed as Groups I - VII lack unity of invention under PCT Rule 13 because they do not share a same or corresponding special technical feature providing a contribution over the prior art.