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(54) Title: METHOD FOR GROWING MICROALGAE, AND DEVICE FOR IMPLEMENTING SAID METHOD

(57) Abstract: This invention relates to a method and to a device to implement said method, to cultivate microalgae and to obtain the simultaneous separation of oleic and protein parts, reducing the required space and drawing mainly from renewable energy sources. The method is characterized by the fact to include the following phases: • said watery mixture, containing said inoculum, follows a path (B) from an inlet point (C) to an outlet point (D), along which it is irradiated by a radiation spectrum suitable to the development and the growth of said microalgae; • along said path (B) they are added NPK salts (containing nitrogen, phosphorus and potassium) and CO₂, these additions, together with the diffusion of said radiation spectrum, causing an intense growth of said algae; • said mixture, strongly enriched of microalgae, is flooded by the ultrasounds that destroy the grown-up algae, splitting them in oleic and in protein components, said action causing the formation of a new watery mixture in which a oleic fraction and a protein fraction are present; • said new watery mixture undergoes a spontaneous gravimetric separation in such a way that: • a oleic fraction, lighter, migrate in the upper part of said new mixture; • a protein fraction, heavier, migrate in the lower part of said new mixture; • a neutral fraction composed almost exclusively of water remains in the intermediate part of said new mixture; • said three fractions are individually taken. The device (A) is characterized by the fact to include: • a basin (1) fitted to contain said watery mixture; • one or more baffles (3, 4, 5) fitted to delimit a path (B) from a point (C) to point (D), said one or more baffles (3, 4, 5) being homogeneous diffuser panels of radiation spectrum suited to the cultivation phase; • means fitted to provide, to said fluid mixture, NPK salts nitrogen, phosphorus and potassium salts) and CO₂, said means being arranged along said path (B); • means (9) fitted to produce ultrasounds, positioned at the final point (D) of said path (B), said ultrasounds being of sufficient power to destroy the grown-up algae splitting them in oleic and protein components, giving rise to a new fluid mixture in which there are present a oleic phase, a protein phase and a neutral phase; • means fitted to diffuse said new fluid mixture, in order to carry out a gravimetric separation of said oleic, protein and neutral phases; • means fitted to separately collect said oleic, protein and neutral phases.

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METHOD FOR GROWING MICROALGAE, AND DEVICE FOR IMPLEMENTING SAID METHOD

DESCRIPTION

This invention relates to a method and to a device to implement said
5 method, to cultivate microalgae and to obtain the simultaneous separation of
oleic and protein parts, reducing the required space and drawing mainly from
renewable energy sources.

It is strongly felt the need to replace fossil fuels with other renewable
and more compatible with the environment. The spinneret of algal crops is
10 producing different solutions ("open ponds", tubular, bioreactors in
greenhouses, etc.). The aim is to obtain concentrations of dry substance
such as to justify the high costs of extraction. Another limitation suffered by
the actual plants, derives from the choice to move the algal mass (process
characterized by a high energy consumption), with actions necessary to
15 keep it in suspension as well as to move it, to exchange its positioning in
order to bring it to be conditioned by the light (exhausting its effectiveness
after the first 20/30 cm of algal mass depth, or even less if thicker and when it
would need more light for its exponential growth). In particular, it is not
possible to bring a specific radiation spectrum, in a pervasive and deep way,
20 with a drastic cost reduction for the mechanical movement of the culture
medium. A limitation derives also from the possibility of biological and
chemical contamination from the environment, because the algal mass is in a
large contact with the environment itself (see the "open ponds" situations)
and it is heavily exposed to the prevalent thermal cycles (often not suitable to
25 the processes of growth) inside it. Some problems are often encountered
even in the phase of collection and selection of the algal mass to be
forwarded to the following processes, that proceeds through the massive
processing of large volumes (by filtration and concentration) that, due to
previous limitations (contamination and uncertain conditions of growth),
30 remain at low concentrations. This invention overcomes many of these
limitations, and reaches high rates of productivity.

The purpose of this invention is to propose a method and a device for implementing said method, respectively conform to claims 1 and 4, for the cultivation and the growth of microalgae present in small quantities (inoculum) in an watery mixture, obtaining the simultaneous separation of oleic and protein parts with relevant indices of growth and in continuous cycle, in tight spaces and compatible with the urban and suburban logistics.

The method is characterized in that it includes the following steps:

- said watery mixture containing said inoculum, follows a path from a point of entry to a point of exit, along which it is irradiated by a radiation spectrum suitable to the development and growth of the said microalgae;
- along said path they are added NPK salts (containing nitrogen, phosphorus and potassium) and CO₂, these additions, together with the diffusion of said radiation spectrum, causing an intense growth of said algae;
- said mixture, strongly enriched of microalgae, is flooded by the ultrasound that destroy the algae splitting them in oleic and in protein components, said action causing the formation of a new watery mixture in which a oleic fraction and a protein fraction are present;
- said new watery mixture undergoes a spontaneous gravimetric separation in such a way that:
 - the oleic fraction, lighter, migrate in the upper part of said new mixture;
 - the protein fraction, heavier, migrate in the lower part of said new mixture;
 - a neutral fraction composed almost exclusively of water remains in the intermediate part of said new mixture;
- said three fractions are individually taken.

According to a preferred embodiment, the method in accordance to the invention further provides the step of measuring out the power of said ultrasound in such a way as to preserve a small amount of microalgae, said

microalgae being then recovered together with said neutral fraction that constitutes the inoculum of said watery mixture of departure. In this way, a continuous cycle for the production of said oleic and protein fractions is obtained.

5 The device is characterized in that it includes:

- a basin fitted to contain said watery mixture;
- one or more baffles fitted to delimit a path from an initial point to a final point, said one or more baffles being diffuser panels of homogeneous radiation spectrum suited to the culture phase;
- 10 • means fitted to provide said fluid mixture, NPK salts (nitrogen, phosphorus and potassium salts) and CO₂, said means being arranged along said path (B);
- means fitted to produce ultrasound, positioned at the final point of said path, said ultrasound being of sufficient power to destroy the algae
- 15 splitting in oleic and protein components, giving rise to a new fluid mixture in which there are present a oleic phase, a protein phase and a neutral phase;
- means fitted to diffuse the said new fluid mixture, in order to carry out a gravimetric separation of said oleic, protein and neutral phases;
- 20 • means fitted to separately collect said oleic, protein and neutral phase.

Other characteristics, such as for example the ability to organize the linear development, in a configuration as a spiral (with a baffle separator to determine the second compartment, which acts as before), with the homogeneous optical diffusers that simultaneously feeds one stretch and the

25 following lap, may be more appropriate to insert the device in existing structures, will be the subject of the dependent claims.

Other characteristics, such as for example the possibility to equip the second compartment with a device fitted to extract more effectively the oleic and the protein components, resorting to gravimetric principles in a state of

30 rest, will be the subject of the dependent claims.

Other characteristics, such as for example the presence of

circumscribed mixers, will allow an adequate support to the processes of cultivation in the vicinity of the collection and support higher productivity, will be the subject of the dependent claims.

The use of a device according to the invention allows to produce components ready for specific following industrial uses, for example for the chemical and pharmaceutical industry.

It can also be used as a retrofit of FER plants (wind and photovoltaic) in agronomic compartments, absorbing redundancies and storing in raw materials, favouring the reclamation of the agronomic waste.

The invention will now be described for illustrative and not limitative purposes, according to a preferred embodiment and with reference to the drawings enclosed, wherein:

- Figure 1 shows the device according to the invention;
- Figure 2 is a plan view of the device according to the invention, illustrating the path of the fluid mixture in treatment.

With reference to figures 1 and 2, with (A) it is indicated a device according to the invention, fitted to cultivate microalgae and to separate the oleic and protein components. Said device (A) comprises a basin (1), predisposed for the phases of culture and extraction in the condition of communicating vessels, inside which it is positioned a first baffle (2) which divides said basin (1) in a first part (1a) and in a second part (1b). Inside said first part (1a) of the basin (1) it is present a plurality of alternate baffles (3, 4, 5) fitted to delimit a sinuous path (B) from a point (C) to a point (D), said alternate baffles (3, 4, 5) being homogeneous diffuser panels of a radiation spectrum suitable to the cultivation phase, for example of the type described in the Italian patent application no. MI2014A002105, in the name of the same proprietors.

In order to be able to properly irradiate the mixture, the thickness of said flow is preferably not higher than 20 ÷ 30 cm. Furthermore, along the path (B) are present means (not shown) to provide the fluid mixture with NPK salts (nitrogen, phosphorus and potassium) and CO₂.

The tank (1) is preferably insulated outside and heated from below, for example by radiant systems to the floor (not shown) for supporting a slight excitation through convective motions so as to create the ideal conditions for the culture.

5 The flow of the mixture, watery based, containing the inoculum, including a small amount of microalgae to be cultured, indicated with the arrow (I), is introduced in the first part (1a) of the basin (1) through a first inlet pipe (6), vertically positioned in coincidence of the point (C) of the beginning of the path (B). The mixture comes out the tube (6) through a plurality of
10 holes (6a) aligned along a generatrix of said first inlet tube (6).

The watery mixture, after the path (B), reaches the point (D) and runs into a second tube (7), through a plurality of holes (7a) aligned along a generatrix of said second tube (7).

In order to obtain a more regular flow, the inlet of the watery mixture
15 takes place in the bottom part of the first inlet tube (6), while the output of said mixture takes place at the top of the second tube (7), according to a scheme of reverse return.

From the second pipe (7), the watery mixture passes into a third tube (8) and, from this, passes in said second part (1b) of the tub (1). The
20 passage from the third tube (8) to said second part (1b) of the tub (1) takes place through a plurality of holes (8a) aligned along a generatrix of said third tube (8). The part of the third tube (8) in which it is present said plurality of holes (8a) is arranged horizontally and is positioned at about half the height of the basin (1).

25 In the top vertical position of said third tube (8), preferably in coincidence of the zone in which the second pipe (7) enters in said third tube (8), is positioned a sonotrode probe (9), the function of which will be described hereinafter.

The mixture leaving the third tube (8) passes through the second part
30 (1b) of the basin (1) and reaches, on the opposite side, three outlet tubes, intermediate (10), upper (11) and lower (12) parallelly arranged to said third

tube (8).

Said three outlet tubes (10), (11) and (12) are provided with holes (10a), (11a) and (12a) respectively, aligned along a generatrix. In the enlarged detail of the upper tube (11) the holes (11a) are shown; said enlarged detail is representative of the holes made on all tubes inside the basin (1).

In the path from the third tube (8) to said three outlet tubes (11), (12) and (13), the mixture is subjected to the separation of the fractions oleic, that goes upwards, and protein, that goes downwards, leaving at half height a fraction composed almost exclusively of water. Proceeding towards the outlet tubes, the watery fraction is directed mainly towards the central tube (10), from which it comes out, as indicated by the arrow (O1). Similarly the oleic fraction, lighter, goes upwards and comes out from the upper tube (11), forming a flow (O2), while the protein fraction, heavier, goes downwards and comes out from the lower tube (12), forming a flow (O3).

The intermediate flow (O1) containing the inoculum, enriched by water, is recycled through said first inlet tube (6) for a new treatment.

The operation of the device (A), that is also the method of treatment according to the invention, is the following:

- the watery mixture containing the inoculum, substantially constituted by a small amount of microalgae, is introduced in the first part (1a) of the basin (1) through the first inlet tube (6);
- said mixture follows said path (B), from the inlet point (C) to the outlet point (D) along which it is irradiated by a radiation spectrum suitable to the development and the growth of the microalgae;
- along said path (B) they are added the NPK salts (containing nitrogen, phosphorus and potassium) in appropriate titre, and CO₂, these additions, together with the diffusion of an appropriate radiation spectrum, causes an intense growth of algae, said growth being able to reach a hourly rate of growth between 10% and 20%;
- arrived in the fourth tube (8), the mixture is flooded by the ultrasounds emitted by the sonotrode probe (9) that destroys the algae splitting

them in oleic and protein components, said division being suitably measured out through the adjustment of the power of the sonotrode, to preserve a small amount of algae;

- the resulting mixture, composed i.e. by a oleic fraction, a protein fraction and a small amount of not damaged algae, runs into the second part (1b) of the basin (1), where it undergoes a gravimetric separation;
- the oleic fraction, lighter, collects in the upper outlet tube (11) and form the output flow (O2), while the protein fraction, heavier, collects in the lower outlet tube (12) and form the output flow (O3);
- the neutral fraction, largely composed of water containing a small amount of not damaged algae, is recycled to the inlet tube (6) for a new cycle of culture.

The process (depending on the unicellular strain and of its chemical-biological structure) can be supported by flocculating or chemical agents to facilitate the separation and its collection (functional as well as to any subsequent treatments). The oleic (high) and the protein (low) components are extracted according to rates of flow correlated to the concentration of the relevant "solute" (detected by suitable densitometers), and the volumes of water must be properly replenished.

According to a preferred embodiment (not shown), said path (B) can be spiral-shaped, said path being delimited by panels, also in the shape of spiral, provided with optical diffusers for the radiative irradiation, that simultaneously feeds one stretch and the following lap.

In addition, along said path (B) they may be positioned localized mixers, for example of the type described in the Italian patent application no. MI2014A002103, in the name of the same proprietors, said mixers allowing an adequate support to the processes of cultivation in the closeness of the picking, obtaining greater productivity.

As it is clear from the foregoing description, in the described device, it is possible to obtain large amounts of oleic and of protein material from small amounts of algae. For a full ecological application of the device, the energy

sources, to be used to favour the growth of the algae and their treatment, will be of renewable type.

The invention has been described for illustrative and not limitative purposes, according to some preferred embodiments. The person skilled in
5 the art could devise several other embodiments, all included within the scope of protection of the enclosed claims.

CLAIMS

1. Method for the cultivation and the growth of microalgae present in small quantities (inoculum) in an watery fluid mixture, obtaining the simultaneous separation of the oleic and of the protein parts characterized by the fact to include the following phases:
- said watery mixture, containing said inoculum, follows a path (B) from an inlet point (C) to an outlet point (D), along which it is irradiated by a radiation spectrum suitable to the development and the growth of said microalgae;
 - along said path (B) they are added NPK salts (containing nitrogen, phosphorus and potassium) and CO₂, these additions, together with the diffusion of said radiation spectrum, causing an intense growth of said algae;
 - said mixture, strongly enriched of microalgae, is flooded by the ultrasounds that destroy the grown-up algae, splitting them in oleic and in protein components, said action causing the formation of a new watery mixture in which a oleic fraction and a protein fraction are present;
 - said new watery mixture undergoes a spontaneous gravimetric separation in such a way that:
 - an oleic fraction, lighter, migrate in the upper part of said new mixture;
 - a protein fraction, heavier, migrate in the lower part of said new mixture;
 - a neutral fraction composed almost exclusively of water remains in the intermediate part of said new mixture;
 - said three fractions are individually taken.
2. Method for the cultivation and the growth of microalgae, according to the claim 1, characterized in that the power of said ultrasounds is regulated in such a way as to preserve a small amount of microalgae, said microalgae being then recovered together with said neutral fraction that constitutes the

inoculum of said watery mixture of departure.

3. Method for the cultivation and the growth of microalgae, according to claim 1, characterized in that said spontaneous gravimetric separation is supported by flocculating or chemical agents.

5 4. Device (A) for the cultivation and the growth of microalgae present in small quantities (inoculum) in a watery fluid mixture, obtaining the simultaneous separation of oleic and proteic parts, characterized by the fact to include:

- a basin (1) fitted to contain said watery mixture;
- one or more baffles (3, 4, 5) fitted to delimit a path (B) from a point (C) to point (D), said one or more baffles (3, 4, 5) being homogeneous
10 diffuser panels of radiation spectrum suited to the cultivation phase;
- means fitted to provide, to said fluid mixture, NPK salts (nitrogen, phosphorus and potassium salts) and CO₂, said means being arranged along said path (B);
- 15 • means (9) fitted to produce ultrasounds, positioned at the final point (D) of said path (B), said ultrasounds being of sufficient power to destroy the grown-up algae splitting them in oleic and protein components, giving rise to a new fluid mixture in which there are present a oleic phase, a protein phase and a neutral phase;
- 20 • means fitted to diffuse said new fluid mixture, in order to carry out a gravimetric separation of said oleic, protein and neutral phases;
- means fitted to separately collect said oleic, protein and neutral phases.

25 5. Device (A) for the cultivation and the growth of microalgae, according to the claim 4, characterized by the fact to include a separator baffle (2) suitable to divide said basin (1) in a first part (1a), in which the culture and the growth of microalgae takes place, and a second part (1b), in which said gravimetric separation of said oleic, protein and neutral phases takes place.

30 6. Device (A) for the cultivation and the growth of microalgae, according to claim 4, characterized by the fact to include:

- a first inlet duct (6), vertically positioned in coincidence with said point

(C) of the beginning of the path (B), the mixture coming out from said first pipe (6) through a plurality of holes (6a) aligned along a generatrix of said first inlet pipe (6);

- a second pipe (7), vertically positioned in coincidence with said point (D) at the end of the path (B), the mixture running into said second pipe (7) through a plurality of holes (7a) aligned along a generatrix of said second outlet pipe (7);

the inlet of the watery mixture occurring from the lower part of said first inlet pipe (6), and the outlet of said mixture occurring from the upper part of said second outlet pipe (7), according to a scheme of reverse return.

7. Device (A) for the cultivation and the growth of microalgae, according to the claim 4, characterized by the fact that said one or more separatory baffles (3, 4, 5) are arranged in alternating sequence and define a winding path (B).

8. Device (A) for the cultivation and the growth of microalgae, according to the claim 4, characterized by the fact that said one or more separatory baffles (3, 4, 5) are arranged in such a way as to define a spiral path.

9. Device (A) for the cultivation and the growth of microalgae, according to the claims 4 and 5, characterized by the fact that said means (9), fitted to produce ultrasounds, are positioned in coincidence with a pouring pipe (8) fitted to transfer the fluid mixture from said first part (1a) to said second part (1b) in which it is divided the basin (1).

10. Device (A) for the cultivation and the growth of microalgae, according to claims 4 and 9, characterized by the fact that said means to diffuse said new fluid mixture, so as to carry out a gravimetric separation of said oleic, protein and neutral phases, include a part of said pouring pipe (8), arranged horizontally at a first end of said second part (1b) of the basin (1), said fluid mixture passing through a plurality of holes (8a) aligned along a generatrix of said pouring pipe (8).

11. Device (A) for the cultivation and the growth of microalgae, according to the claims from 4 to 10, characterized by the fact that said means fitted to separately collect said oleic, protein and neutral phases include an

intermediate outlet pipe (10), an upper outlet pipe (11) and a lower outlet pipe (12), arranged horizontally at a second end of said second part (1b) of the basin (1), said three outlet pipes (10), (11) and (12) being provided with holes (10a), (11a) and (12a) respectively, aligned along a generatrix.

5 12. Device (A) for the cultivation and the growth of microalgae, according to at least one of the claims from 4 to 11, characterized by the fact to provide some localized mixers positioned along said path (B).

13. Device (A) for the cultivation and the growth of microalgae, according to at least one of the claims from 4 to 12, characterized by the fact to include
10 means, positioned in coincidence with said second part (1b) of the basin (1), fitted to spread flocculating and/or chemical agents fitted to favour the gravimetric separation of the fractions that constitute said new fluid mixture.

14. Device (A) for the cultivation and the growth of microalgae, according to at least one of the claims from 4 to 13, characterized by the fact to include
15 means designed to insulate said basin (1) and to warm said basin (1) from the bottom so as to support a slight excitation through convective motions, to create the ideal conditions for the cultivation.

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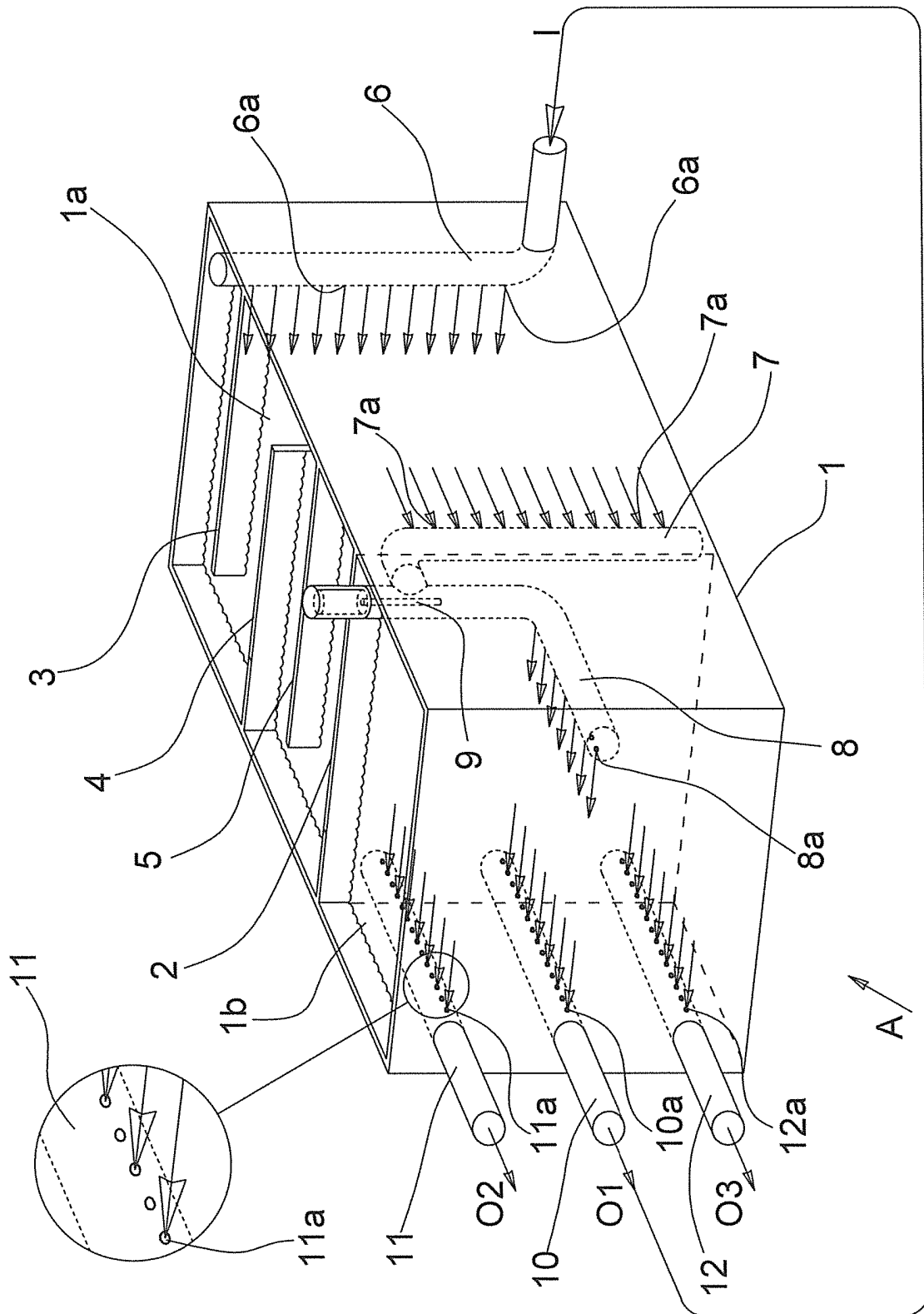


Fig. 1

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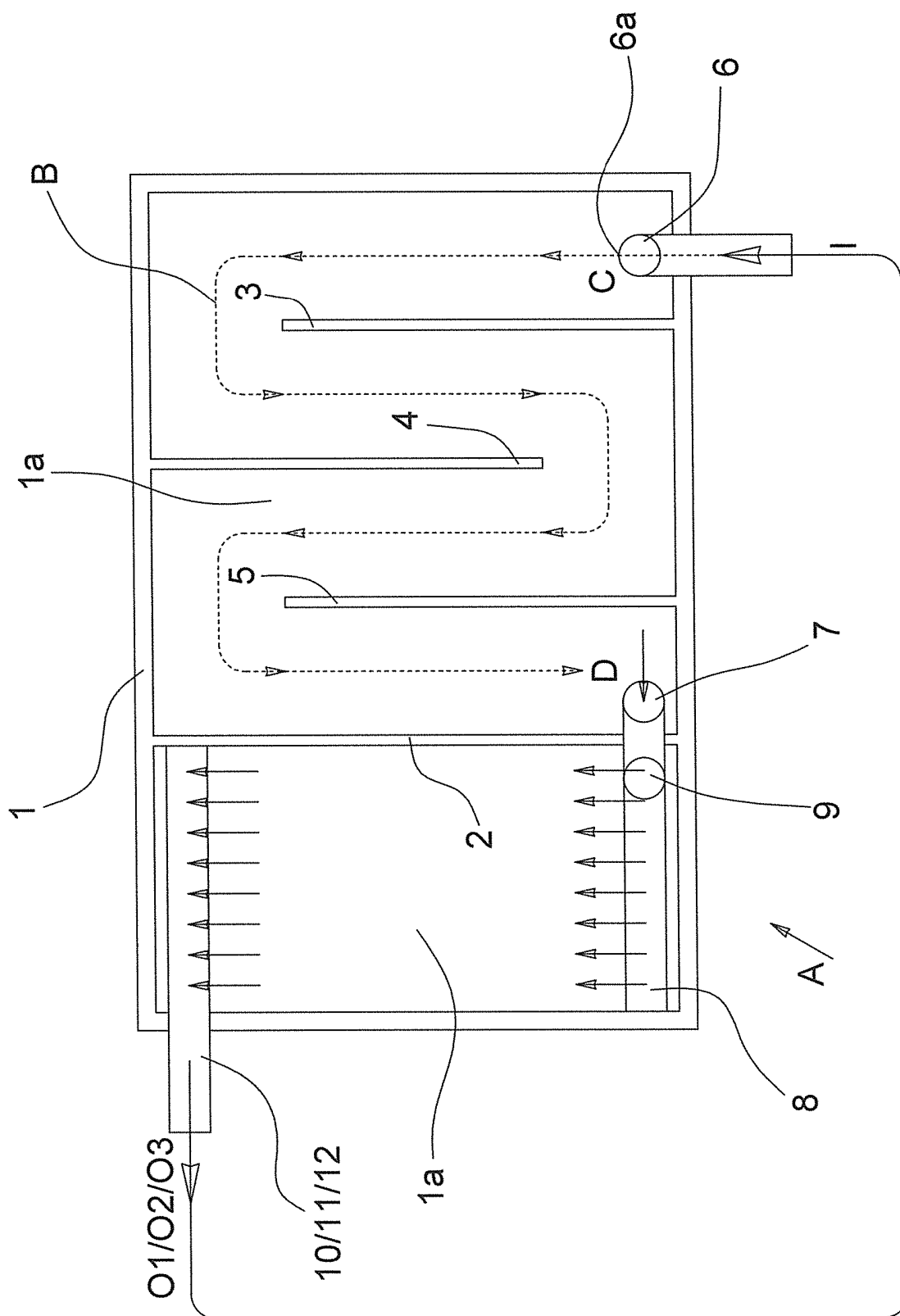


Fig. 2

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(54) Title: PHOTOSYNTHETIC OIL PRODUCTION WITH HIGH CARBON DIOXIDE UTILIZATION

(57) Abstract: A system for processing oil from algae is disclosed. Specifically, the system recycles byproducts of the process for use as nutrients during algae growth and oil production. The system includes a conduit for growing algae and an algae separator that removes the algae from the conduit. Also, the system includes a device for lysing the algae and an oil separator to remove the oil from the lysed matter. Further, the system includes a biofuel reactor that receives oil from the oil separator and synthesizes biofuel and glycerin. Moreover, the algae separator, oil separator and biofuel reactor all recycle byproducts back to the conduit to support further algae growth.

PHOTOSYNTHETIC OIL PRODUCTION WITH HIGH CARBON DIOXIDE UTILIZATION

FIELD OF THE INVENTION

The present invention pertains generally to processes for harvesting oil from algae. More particularly, the present invention pertains to a cost efficient supply of nutrients to support the growth of algae cells having a high oil
5 content. The present invention is particularly, but not exclusively, useful as a system and method for recycling byproducts of an algae oil harvesting process for use in supporting algae cell growth and oil production.

BACKGROUND OF THE INVENTION

As worldwide petroleum deposits decrease, there is rising concern over
10 shortages and the costs that are associated with the production of hydrocarbon products. As a result, alternatives to products that are currently processed from petroleum are being investigated. In this effort, biofuels such as biodiesel have been identified as a possible alternative to petroleum-based transportation fuels. In general, biodiesel is a fuel comprised of mono-alkyl
15 esters of long chain fatty acids derived from plant oils or animal fats. In industrial practice, biodiesel is created when plant oils or animal fats are reacted with an alcohol, such as methanol.

For plant-derived biofuel, solar energy is first transformed into chemical energy through photosynthesis. The chemical energy is then refined into a
20 usable fuel. Currently, the process involved in creating biofuel from plant oils is expensive relative to the process of extracting and refining petroleum. It is possible, however, that the cost of processing a plant-derived biofuel could be reduced by maximizing the rate of growth of the plant source. Because algae is known to be one of the most efficient plants for converting solar energy into
25 cell growth, it is of particular interest as a biofuel source. However, current algae processing methods have failed to result in a cost effective algae-derived biofuel.

In overview, the biochemical process of photosynthesis provides algae with the ability to convert solar energy into chemical energy. During cell growth, this chemical energy is used to drive synthetic reactions, such as the formation of sugars or the fixation of nitrogen into amino acids for protein synthesis. Excess chemical energy is stored in the form of fats and oils as triglycerides. Thus, the creation of oil in algae only requires sunlight, carbon dioxide and the nutrients necessary for formation of triglycerides. Nevertheless, with the volume requirements for a fuel source, the costs associated with the inputs are high.

In light of the above, it is an object of the present invention to provide a system and method for processing oil from algae which reduces input costs. For this purpose, a number of systems have been developed, such as those disclosed in co-pending U.S. Patent Application No. 11/549,532 for an invention entitled "Photosynthetic Oil Production in a Two-Stage Reactor," co-pending U.S. Patent Application No. 11/549,541 for an invention entitled "Photosynthetic Carbon Dioxide Sequestration and Pollution Abatement" and co-pending U.S. Patent Application No. 11/549,552 for an invention entitled "High Photoefficiency Microalgae Bioreactors," which are filed concurrently herewith and assigned to the same assignee as the present invention, and are hereby incorporated by reference. Another object of the present invention is to provide a recycling system for feeding oil harvesting byproducts back to the conduit where high oil content algae is grown. Still another object of the present invention is to provide a system for supplying nutrients to algae cells in the form of processed algae cell matter. Another object of the present invention is to provide a system for recycling the glycerin byproduct from the creation of biofuel as a source of carbon to foster further oil production in algae cells. Another object of the present invention is to provide a system for processing oil from algae that defines a flow path for continuous movement of the algae and its processed derivatives. Yet another object of the present invention is to provide a system and method for processing algae with high oil content that is simple to implement, easy to use, and comparatively cost effective.

SUMMARY OF THE INVENTION

In accordance with the present invention, a system is provided for efficiently processing oil from algae. For this purpose, the system recycles byproducts of the process for use as nutrients to support algae cell growth and the cellular production of oil. Structurally, the system includes a chemostat that defines a conduit for growing algae cells. The chemostat's conduit includes input ports for feeding material into the conduit as well as an output port. Further, the system includes a plug flow reactor that defines a conduit for fostering oil production within the algae cells. For the present invention, the plug flow reactor has an input port that is positioned to receive material from the output port of the chemostat.

In addition to the chemostat and plug flow reactor, the system includes an algae separator. Specifically, the algae separator is positioned in fluid communication with the plug flow reactor to remove the algae cells from the plug flow reactor's conduit. Structurally, the algae separator includes an outlet for the remaining effluence which is in fluid communication with the input port of the chemostat. Further, the system includes a device for lysing algae cells to unbind oil from the algae cells. For purposes of the present invention, the lysing device is positioned to receive algae cells from the algae separator.

Downstream of the lysing device, the system includes an oil separator that receives the lysed cells and withdraws the oil from remaining cell matter. For purposes of the present invention, the oil separator has an outlet for the remaining cell matter which is in fluid communication with the input port of the chemostat. Further, the system may include a hydrolyzing device interconnected between the oil separator and the chemostat. In addition to the cell matter outlet, the oil separator includes an outlet for the oil. For the present invention, the system includes a biofuel reactor that is in fluid communication with the outlet for oil. In a known process, the biofuel reactor reacts an alcohol with the oil to synthesize biofuel and, as a byproduct,

glycerin. Structurally, the biofuel reactor includes an exit for the glycerin that is in fluid communication with the input port of the plug flow reactor.

In operation, algae cells are grown in the chemostat and are continuously transferred to the plug flow reactor. In the plug flow reactor, the
5 algae cells increase the rate of intracellular oil production. Thereafter, the algae separator removes the algae cells from the remaining effluence in the plug flow reactor. The remaining effluence is diverted back to the chemostat to serve as a source of nutrition for the algae cells growing therein while the algae cells are delivered to the cell lysis device. At the cell lysis device, the
10 cells are lysed to unbind the oil from the remaining cell matter. This unbound cell material is received by the oil separator from the cell lysis device. Next, the oil separator withdraws the oil from the remaining cell matter and effectively forms two streams of material. The stream of remaining cell matter is transferred to the hydrolysis device where the cell matter is broken into
15 small units which are more easily absorbed by algae cells during cell growth. Thereafter, the hydrolyzed cell matter is delivered to the chemostat to serve as a source of nutrition for the algae cells growing therein. At the same time, the stream of oil is transmitted from the oil separator to the biofuel reactor. In the biofuel reactor, the oil is reacted with an alcohol to form biofuel and a
20 glycerin byproduct. The glycerin byproduct is fed back into the plug flow reactor to serve as a source of carbon for the algae cells therein during the production of intracellular oil.

BRIEF DESCRIPTION OF THE DRAWINGS

The novel features of this invention, as well as the invention itself, both
25 as to its structure and its operation, will be best understood from the accompanying drawing, taken in conjunction with the accompanying description, in which the Figure is a schematic view of the system for processing oil from algae in accordance with the present invention.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

Referring to the Figure, a system for processing oil from algae in accordance with the present invention is shown and generally designated 10. Specifically, in the system 10 byproducts of the processing method are recycled to foster growth of algae cells having high oil content. As shown, the system 10 includes a conduit 12 for growing algae cells with high oil content (exemplary cells depicted at 14). As further shown, the conduit 12 includes an upstream conduit section 16 that is defined by a continuously stirred first stage reactor or chemostat 18. Also, the conduit 12 includes a downstream conduit section 20 that is defined by a plug flow second stage reactor 22. As shown, each conduit section 16, 20 includes input ports 24a-e. Further, the upstream conduit section 16 includes an output port 26. As shown, the output port 26 of the upstream conduit section 16 and the input port 24c of the plug flow reactor 22 are in fluid communication. In this manner, the conduit 12 passes through the chemostat 18 and the plug flow reactor 22.

As further shown in the Figure, the system 10 includes an algae separator 28 that is in fluid communication with the downstream conduit section 20 in the plug flow reactor 22. For purposes of the present invention, the algae separator 28 removes algae cells 14 from the downstream conduit section 20. As shown, the algae separator includes outlets 29a and 29b. Also, the system 10 includes a cell lysis device 30 that receives algae cells 14 from the outlet 29a of the algae separator 28 via pipe 32. As shown, the cell lysis device 30 is in fluid communication with an oil separator 34. Specifically, a pipe 36 interconnects the cell lysis device 30 and the oil separator 34. For purposes of the present invention, the oil separator 34 is provided with two outlets 38a-b. As shown, the outlet 38a is connected to a hydrolysis device 40 by a pipe 42. Further, the hydrolysis device 40 is connected to the input port 24b in the upstream conduit section 16 of the chemostat 18 by a pipe 44.

Referring back to the oil separator 34, it can be seen that the outlet 38b is connected to a biofuel reactor 46 by a pipe 48. It is further shown that the biofuel reactor 46 includes two exits 50a-b. For purposes of the present invention, the exit 50a is connected to the input port 24e in the downstream
5 conduit section 20 of the plug flow reactor 22 by a pipe 52. Additionally or alternatively, the exit 50a may be connected to the input port 24b in the upstream conduit section 16 of the chemostat 18 by a pipe 54 (shown in phantom). As further shown, the exit 50b is connected to a pipe 56 which may connect to a tank or reservoir (not shown) for purposes of the present
10 invention.

Referring now to the algae separator 28, it can be seen that the outlet 29b is in fluid communication with the input port 24a of the chemostat 18. Further, a blowdown 57 is shown to be interconnected between the algae separator 28 and the input port 24a. Specifically, a pipe 59 connects the
15 outlet 29b and the blowdown 57, and a pipe 61 connects the blowdown 57 and the input port 24a.

In operation of the present invention, algae cells 14 are initially grown in the upstream conduit section 16 in the chemostat 18. Specifically, a medium with a nutrient mix is continuously fed through input port 24a into the
20 upstream conduit section 16 at a selected rate. Further, the conditions in the upstream conduit section 16 are maintained for maximum algal growth. For instance, in order to maintain the desired conditions, the medium and the algae cells 14 are moved around the upstream conduit section 16 at a fluid flow velocity in the range of approximately ten to two hundred centimeters per
25 second, and preferably at fifty centimeters per second. Further, the amount of algae cells 14 in the upstream conduit section 16 is kept substantially constant. Specifically, the medium with nutrient mix is continuously fed into the input port 24a and an effluence 58 containing algae cells 14 is continuously removed through the output port 26 of the upstream conduit
30 section 16 as overflow. Under preferred conditions, approximately ten grams of algae per liter of fluid circulate in the upstream conduit section 16.

Preferably, the residence time for algae cells 14 in the upstream conduit section 16 is about one to ten days.

After entering the input port 24c, the effluence 58 containing algae cells 14 moves through the downstream conduit section 20 in the direction of arrows 60 in a plug flow regime. Preferably, the effluence 58 moves through the downstream conduit section 20 of the plug flow reactor 22 at a rate of between ten and two hundred centimeters per second. Further, as the effluence 58 moves downstream, a modified nutrient mix may be added to the downstream conduit section 20 through the input port 24d. This modified nutrient mix may contain a limited amount of a selected constituent, such as nitrogen or phosphorous. The absence or small amount of the selected constituent causes the algae cells 14 to focus on energy storage rather than growth. As a result, the algae cells 14 form triglycerides.

At the end of the downstream conduit section 20, the algae separator 28 removes the algae cells 14 from the effluence 58. To facilitate this process, the depth of the downstream conduit section 20 may be increased near the algae separator 28. The corresponding increase in the fluid flow cross-sectional area, and decrease in fluid flow rate, allows the algae cells 14 to settle to the bottom or float to the top of the conduit section 20, depending on the oil content of the algae cells 14. In certain embodiments, the modified nutrient mix may include a limited amount of a predetermined constituent to trigger flocculation of the algae cells 14 in the downstream conduit section 20. The predetermined constituent may be the same as the selected constituent such that a shortage of nitrogen, for example, causes both the production of triglycerides and the flocculation of the algae cells 14.

After the algae cells 14 are removed from the conduit 12 by the algae separator 28, the remaining effluence (indicated by arrow 63) is discharged from the algae separator 28 through the outlet 29b. As shown, the remaining effluence 63 passes through the blowdown 57 where impurities, such as salt, are removed. Then, additional nutrients (indicated by arrow 65) may be added to the remaining effluence 63 for replenishment to support further cell

growth in the chemostat 18. After being replenished, the remaining effluence 63 is fed back into the chemostat 18 through the input port 24a.

While the remaining effluence 63 is discharged through outlet 29b, the algae cells 14 removed by the algae separator 28 are delivered to the cell lysis device 30. Specifically, the algae cells 14 pass through the outlet 29a and the pipe 32 to the cell lysis device 30 as indicated by arrow 60. For purposes of the present invention, the cell lysis device 30 lyses the algae cells 14 to unbind the oil therein from the remaining cell matter. After the lysing process occurs, the unbound oil and remaining cell matter, collectively identified by arrow 62, are passed through pipe 36 to the oil separator 34. Thereafter, the oil separator 34 withdraws the oil from the remaining cell matter as is known in the art. After this separation is performed, the oil separator 34 discharges the remaining cell matter (identified by arrow 64) out of the outlet 38a and through the pipe 42 to the input port 24b of the chemostat 18.

In the chemostat 18, the remaining cell matter 64 is utilized as a source of nutrients and energy for the growth of algae cells 14. Because small units of the remaining cell matter 64 are more easily absorbed or otherwise processed by the growing algae cells 14, the remaining cell matter 64 may first be broken down before being fed into the input port 24b of the chemostat 18. To this end, the hydrolysis device 40 is interconnected between the oil separator 34 and the chemostat 18. Accordingly, the hydrolysis device 40 receives the remaining cell matter 64 from the oil separator 34, hydrolyzes the received cell matter 64, and then passes hydrolyzed cell matter (identified by arrow 66) to the chemostat 18 through pipe 44.

Referring back to the oil separator 34, it is recalled that the remaining cell matter 64 was discharged through the outlet 38a. At the same time, the oil withdrawn by the oil separator 34 is discharged through the outlet 38b. Specifically, the oil (identified by arrow 68) is delivered to the biofuel reactor 46 through the pipe 48. In the biofuel reactor 46, the oil 68 is reacted with alcohol, such as methanol, to create mono-alkyl esters, i.e., biofuel fuel. This biofuel fuel (identified by arrow 70) is released from the exit 50b of the biofuel

reactor 46 through the pipe 56 to a tank, reservoir, or pipeline (not shown) for use as fuel. In addition to the biofuel fuel 70, the reaction between the oil 68 and the alcohol produces glycerin as a byproduct. For purposes of the present invention, the glycerin (identified by arrow 72) is pumped out of the exit 50a of the biofuel reactor 46 through the pipe 52 to the input port 24e of the plug flow reactor 22.

In the plug flow reactor 22, the glycerin 72 is utilized as a source of carbon by the algae cells 14. Importantly, the glycerin 72 does not provide any nutrients that may be limited to induce oil production by the algae cells 14 or to trigger flocculation. The glycerin 72 may be added to the plug flow reactor 22 at night to aid in night-time oil production. Further, because glycerin 72 would otherwise provide bacteria and/or other non-photosynthetic organisms with an energy source, limiting the addition of glycerin 72 to the plug flow reactor 22 only at night allows the algae cells 14 to utilize the glycerin 72 without facilitating the growth of foreign organisms. As shown in the Figure, the exit 50a of the biofuel reactor 46 may also be in fluid communication with the input port 24b of the chemostat 18 via the pipe 54 (shown in phantom). This arrangement allows the glycerin 72 to be provided to the chemostat 18 as a carbon source.

While the particular Photosynthetic Oil Production with High Carbon Dioxide Utilization as herein shown and disclosed in detail is fully capable of obtaining the objects and providing the advantages herein before stated, it is to be understood that it is merely illustrative of the presently preferred embodiments of the invention and that no limitations are intended to the details of construction or design herein shown other than as described in the appended claims.

What is claimed is:

1. A system for processing oil from algae which comprises:
 - a conduit for growing algae cells with high oil content, said conduit having an input port;
 - 5 an algae separator in fluid communication with the conduit for removing the algae cells from remaining effluence, with the remaining effluence being a byproduct;
 - a device for lysing the algae cells removed from the conduit to unbind oil within the algae cells;
 - 10 an oil separator for withdrawing the oil from remaining cell matter, with the remaining cell matter being a byproduct;
 - a reactor for receiving the oil from the oil separator and for synthesizing biofuel and glycerin from said oil, with said glycerin being a byproduct; and
 - 15 a means for recycling at least one byproduct through the input port to the conduit to support growth of algae cells with high oil content.

2. A system as recited in claim 1 wherein said algae separator has an outlet in fluid communication with the input port of the conduit for recycling the remaining effluence to the conduit to support growth of high oil content
20 algae cells.

3. A system as recited in claim 1 wherein said reactor has an exit in fluid communication with the input port of the conduit for recycling the glycerin to the conduit to support growth of high oil content algae cells.

4. A system as recited in claim 1 wherein said oil separator has an
25 outlet in fluid communication with the input port of the conduit for recycling the remaining cell matter to the conduit to support growth of high oil content algae cells.

5. A system as recited in claim 4 wherein the remaining cell matter includes biopolymers, and the system further comprises a means for hydrolyzing the remaining cell matter to reduce the biopolymers therein to smaller subunits, with said hydrolyzing means being interconnected between
5 the outlet of the separator and the input port of the conduit.

6. A system as recited in claim 1 wherein the conduit includes a first conduit section formed in a chemostat for growing algae cells therein, with the first conduit section including a first input port, and further wherein said oil separator has an outlet in fluid communication with the first input port
10 for recycling the remaining cell matter to the first conduit section to support growth of algae cells therein.

7. A system as recited in claim 1 wherein the conduit includes a first conduit section formed in a chemostat for growing algae cells therein, with the first conduit section including a first input port, and further wherein
15 said reactor has an exit in fluid communication with the first input port for recycling the glycerin to the first conduit section to support growth of algae cells therein.

8. A system as recited in claim 1 wherein the conduit includes a second conduit section formed in a plug flow reactor for increasing the oil
20 content of the algae cells therein, with the second conduit section including a second input port, and further wherein said reactor has an exit in fluid communication with the second input port for recycling the glycerin to the second conduit section to support oil production within the algae cells therein.

9. A system for processing oil from algae which comprises:
a conduit for growing algae cells with high oil content, said
conduit having an input port;
an algae separator in fluid communication with the conduit for
5 removing the algae cells from remaining effluence, with the remaining
effluence being a byproduct;
a device for lysing the algae cells removed from the conduit to
unbind oil from the algae cells; and
an oil separator for withdrawing the oil from remaining cell
10 matter, said oil separator having an outlet in fluid communication with
the input port of the conduit for recycling the remaining cell matter to
the conduit to support growth of high oil content algae cells.
10. A system as recited in claim 9 further comprising a means for
hydrolyzing the remaining cell matter to reduce the remaining cell matter to
15 smaller subunits, with said hydrolyzing means being interconnected between
the outlet of the separator and the input port of the conduit.
11. A system as recited in claim 9 further comprising:
a reactor for receiving the oil from the oil separator and for
synthesizing biofuel and glycerin from said oil, said reactor having an
20 exit in fluid communication with the input port of the conduit for
recycling the glycerin to the conduit to support growth of high oil
content algae cells.

12. A system as recited in claim 11 wherein the conduit includes a first conduit section formed in a chemostat for growing algae cells therein, with said first conduit section including the input port, and further wherein said outlet of said oil separator is in fluid communication with the input port of the first conduit section for recycling the remaining cell matter to the first conduit section to support growth of algae cells therein.

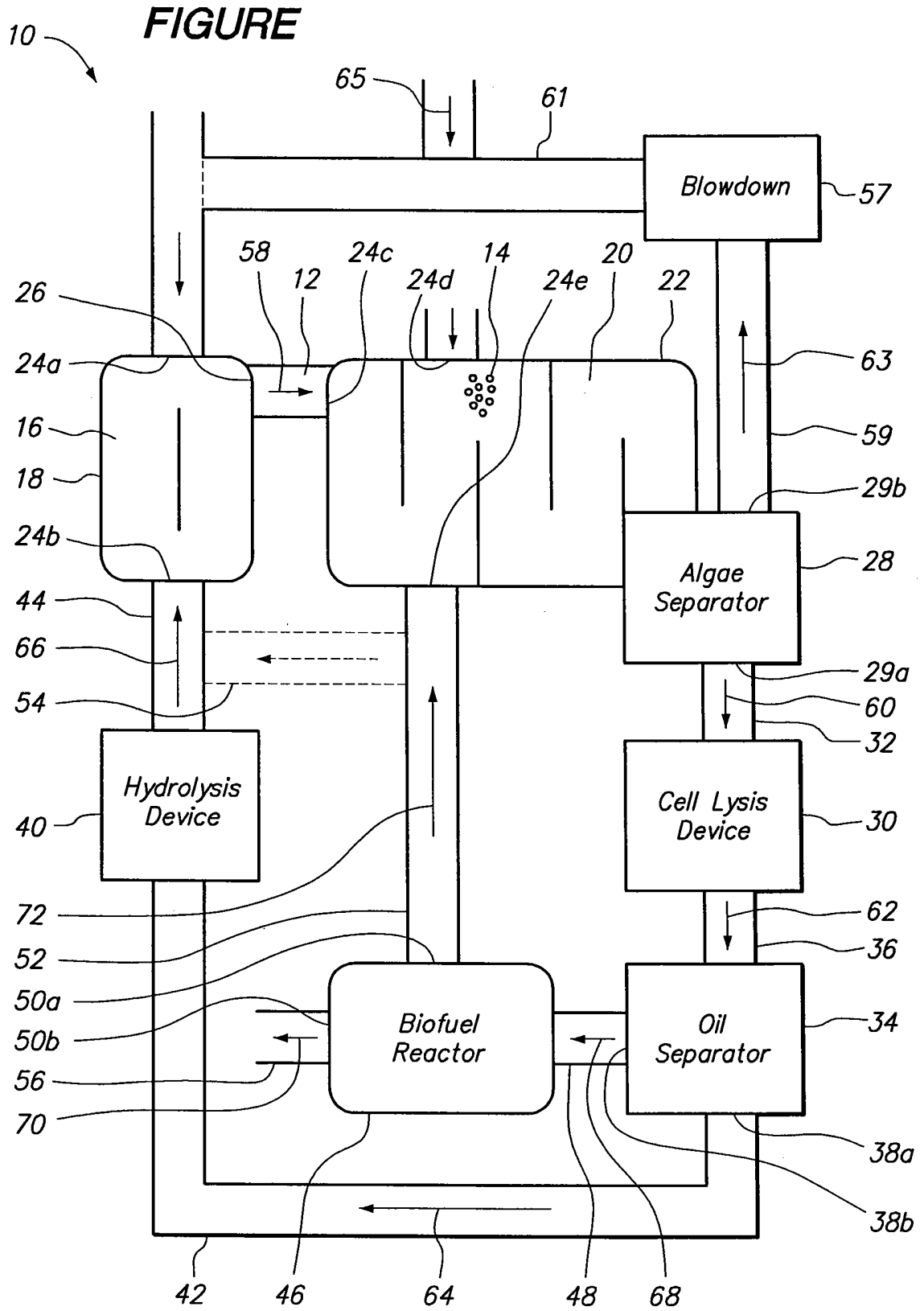
13. A system as recited in claim 12 wherein said exit of said reactor is in fluid communication with the input port of the first conduit section for recycling the glycerin to the first conduit section to support growth of algae cells therein.

14. A system as recited in claim 12 wherein the conduit includes a second conduit section formed in a plug flow reactor for increasing the oil content of the algae cells therein, with said second conduit section including an input port, and further wherein said exit of said reactor is in fluid communication with the input port in the second conduit section for recycling the glycerin to the second conduit section to support oil production within the algae cells therein.

15. A method of processing oil from algae which comprises the steps of:
- growing algae cells with high oil content in a conduit;
 - removing the algae cells from the conduit, with the remaining effluence being a byproduct of the removing step;
 - lysing the algae cells removed from the conduit to unbind oil within the algae cells;
 - withdrawing the oil from remaining cell matter, with the remaining cell matter being a byproduct of the lysing step;
 - synthesizing biofuel and glycerin from the withdrawn oil, with said glycerin being a byproduct of the synthesizing step; and
 - recycling at least one byproduct to the conduit to support growth of algae cells with high oil content.
16. A method as recited in claim 15 wherein the glycerin is recycled to the conduit to support growth of high oil content algae cells.
17. A method as recited in claim 15 wherein the remaining effluence is recycle to the conduit to support growth of high oil content algae cells.
18. A method as recited in claim 15 wherein the remaining cell matter is recycled to the conduit to support growth of high oil content algae cells.

19. A method as recited in claim 18 further comprising the step of hydrolyzing the remaining cell matter to reduce the remaining cell matter to smaller subunits before the remaining cell matter is recycled to the conduit to support growth of high oil content algae cells.

5 20. A method as recited in claim 15 wherein the conduit includes a first conduit section formed in a chemostat and a second conduit section formed in a plug flow reactor, and wherein the growing step includes developing algae cells in the first conduit section and facilitating oil production in the algae cells in the second conduit section, and further wherein the
10 recycling step includes delivering the remaining cell matter to the first conduit section and delivering the glycerin to the second conduit section.



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(57) Abstract: The presently described invention relates to the method of extraction of oils from photosynthetic cultures using hydrodynamic cavitation technology for the production of biofuels or other products. This method is referred to herein as Hydrodynamic extraction.



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HYDRODYNAMIC EXTRACTION OF OILS FROM PHOTOSYNTHETIC CULTURES

Related Applications

[0001] This application claims the benefit of priority of U.S. Provisional Application No. 61/105,190, which is hereby incorporated by reference in its entirety.

Technical Field

[0002] The disclosed invention relates to the method of extraction of oils from photosynthetic cultures using hydrodynamic cavitation technology for the production of biofuels or other products.

Background Art

[0003] Microalgae and other photosynthetic cultures produce and store lipids, fatty acids, monoglycerides, and diglycerides that can make up a significant percentage of their total ash free dry weight. The hydrocarbons produced by microalgae and other photosynthetic cultures often form oils. Microalgae contain a wide variety of oil lipids, which include membrane-bound polar lipids and non-polar lipids that also encompass free fatty acids and fatty acids. Lipid fractions as high as 70-85% have been reported in some microalgae.

[0004] Microalgae oil plays an essential nutritional role in the marine animal world. A 60 ton-blue-whale may have 2 tons of microalgae plankton in its gut for nutrition. The oil contents of whale, fish, and shark-liver oil are the condensates of oil droplets originally stored in the microalgae cells. For marine culture of zooplankters, larval shrimp, and juvenile oysters the aquaculture industry has long used microalgae as a food source not only because of their characteristically high lipid and fatty acid content, but also because of their abundance of certain polyunsaturated fatty acids (PUFAs) essential to the marine animal diet.

[0005] In addition to cultivating microalgae as an oil-rich nutritional source for aquaculture, oils derived from cultivated microalgae are used for pharmaceutical, nutraceutical, and cosmetic purposes. Products made from microalgae oils command a very large per acre revenue (over \$600,000 per acre per year). With such high profit margins production cost efficiencies and new technologies are not aggressively pursued.

[0006] Another potential application of oils derived from cultivated microalgae is for use in the production of biofuels, which are fuels suitable for burning in standard internal combustion engines that are derived from biological sources. In fact most of the fossil fuels extracted from the seas (both former and current) are derived from the oils synthesized and stored by the microalgae of the past ages. With the increasing demand and shrinking supply of fossil fuels, there is a present need for alternative fuels and the promise of biofuels derived from microalgae oils have recently been a source of major interest and investments.

[0007] In comparison to the pharmaceutical, nutraceutical, or cosmetic products made from microalgae oils, biofuels from microalgae have a much lower per acre revenue (under \$30,000 per acre per year) and thereby highly sensitive to operating cost efficiencies. Current technologies used in the core processes of microalgae oil production are inefficient and have high operating costs. New technologies must be developed to deliver cost-efficient production processes before microalgae biofuels become commercially economic.

[0008] An important process step in harvesting microalgae oil is extraction. Extraction is the process of removing the oils from the microalgae cells. Microalgae oils are extracted through a wide variety of methods. Current extraction technologies are costly and do not lend themselves to an efficient and cost effective production systems. Estimates of the current costs to extract oil from microalgae vary, but are likely to be around \$1.80/kg or \$2.91/liter (\$11.00 gallon)

[0009] Mechanical pressing is the simplest method of extraction. Because different strains of microalgae vary widely in their physical attributes, various press configurations (screw, expeller, piston, etc) work better for specific microalgae types. Using typical methods, microalgae are harvested, dried, and then can be "pressed" out with an oil press. A press can extract between 70-75% of the oils out of microalgae. Often, mechanical pressing is used in conjunction with chemical solvents. While simple in design, this is a highly energy intensive and extraction efficiency is low.

[0010] Chemical solvent extraction, used alone or in combination with other methods, is another common methodology for extracting microalgae oils. Oils from the algae are extracted through repeated washing, or percolation, with an organic solvent under reflux in special glassware. Benzene and ether have been used, but a more popular chemical for solvent extraction is hexane, which is widely used and is less expensive. A downside to using solvents for oil extraction is the inherent dangers involved in working with the chemicals. Care must be taken to avoid exposure to vapors and direct contact with the skin, either of

which can cause serious damage. For example, benzene is classified as a carcinogen. Additionally, chemical solvents also present the problem of being an explosion hazard.

[0011] Hexane solvent extraction can be used in isolation or it can be used along with the mechanical press method. After the oil has been extracted using a mechanical press, the remaining pulp can be mixed with cyclohexane to extract the remaining oil content. The oil dissolves in the cyclohexane, and the pulp is filtered out from the solution. The oil and cyclohexane are separated by means of distillation. These two stages (cold press & hexane solvent) together will be able to derive more than 95% of the total oil present in the microalgae.

[0012] Another extraction method is enzymatic extraction which uses enzymes to degrade the cell walls with water acting as the solvent, making fractionation of the oil much easier. The costs of this extraction process are estimated to be much greater than hexane solvent extraction. The enzymatic extraction can be supported by ultrasonication. The combination "sonoenzymatic treatment" causes faster extraction and higher oil yields.

[0013] Ultrasonic-assisted extraction, a branch of sonochemistry, can greatly accelerate extraction processes. Using an ultrasonic reactor, ultrasonic waves are used to create cavitation bubbles in a solvent material, when these bubbles collapse near the cell walls, it creates shock waves and liquid jets that cause those cells walls to break and release their contents into the solvent. A variant of ultrasonic extraction is electrostatic shockwave extraction where cavitation bubbles are created by an ultra-high electric impulse rather than by an acoustic impulse. Sonochemistry can be done wet or dry. If done wet the water will need to be extracted from the mash before extraction of oils with a solvent.

[0014] Osmotic shock is yet another method used for extraction. Osmotic shock is a sudden reduction in osmotic pressure that can cause cells in a solution to rupture. Osmotic shock extraction can be performed by taking a high saline growth medium, harvesting to a sludge, and then dumping the sludge into distilled water which will burst nearly all the cells and then the oil can be skimmed off the surface.

[0015] Another method of extraction is supercritical fluid extraction. In supercritical fluid/CO₂ extraction, CO₂ is liquefied under pressure and heated to the point that it has the properties of both a liquid and a gas, this liquefied fluid then acts as the solvent in extracting the oil. This method requires special equipment for containment and pressure in the supercritical fluid/CO₂ extraction. Supercritical fluid extraction does not need to be

absolutely dry as by varying the pressure and temperature one can fractionate the sample being extracted.

[0016] Electroporation is yet another extraction method. With electroporation, ultra high electric impulses are directed toward the microalgae perforating the cell walls to release the oil contents.

[0017] All these extraction methods are too costly and complex for commercial use on a large scale. In addition these extraction methods are static batch processes and do not lend themselves to a cost efficient system. These extraction methods are also dependant on the effectiveness of the preceding post-cultivation processes of harvesting and de-watering which are required for removing the microalgae from its growth medium and increasing the microalgae cell density by removing most of the water content in order to prepare the microalgae for current oil extraction methods; current technologies used for harvesting and de-watering processes have very high operating costs. In summary, current extraction methods are not suitable for the low-cost production of biofuels from microalgae oils on a commercial scale. To enable low-cost microalgae biofuel production an extraction process needs to have a low capital cost, ultra low operating costs, and lend itself to an integrated, economical, and continuous production system on a commercial scale.

Summary of the Invention

[0018] The presently described invention relates a method for the continuous extraction of hydrocarbons from photosynthetic organism, and the apparatus for performing the method. In a preferred embodiment the method comprises the step of applying hydrodynamic cavitation to a continuous flow of microalgae in its growth medium to rupture the cell walls and extract the microalgae oil.

[0019] Another preferred embodiment of the invention comprises the step of applying hydrodynamic cavitation to a continuous flow of microalgae in a fluid medium to rupture the cell walls and extract the microalgae oil.

[0020] As part of the hydrodynamic cavitation process the growth medium or fluid medium used is sterilized for reuse.

[0021] The presently invention relates to a method of rupturing a microalgae cell wall, comprising providing a continuous flow of a fluid medium comprising one or more microalgae to a hydrodynamic cavitation device; applying hydrodynamic cavitation in sufficient quantity to rupture one or more microalgae cells, whereby microalgae oil is released

from the microalgae into the medium; and extract the microalgae oil from the medium. In one embodiment, the method further comprises the step of dewatering the medium. In one embodiment, the photosynthetic organism is a diatom, such as a *Chaetoceros* species. In another embodiment of the invention, the hydrodynamic cavitation is applied using a multi-stage hydrodynamic cavitation reactor, and in another, the hydrodynamic cavitation is applied using a magnetic impulse cavitation reactor. In another aspect of the invention, the processed medium after hydrodynamic cavitation is separated into components comprising microalgae oil, microalgae cell walls, and the processed fluid medium. The separated components can be further processed into a biofuel, such as biodiesel, and such further processing can comprise one or more additional rounds of hydrodynamic cavitation to produce transesterification. The processed medium can also be recycled for use as microalgae cultivation medium, and the processed medium is subjected to one or more additional rounds of hydrodynamic cavitation. Another embodiment of the invention relates to a biofuel produced by the disclosed methods, where a particular biofuel that can be prepared is a biodiesel.

Detailed Description of the Invention

[0022] The presently described invention relates to continuous production and extraction methods for harvesting hydrocarbons from microalgae and other photosynthetic cultures. Extraction methods that enable a continuous production process are preferred over static batch processes because continuous production methods significantly reduce the cost of producing finished biofuels or other products. Hydrodynamic cavitation is a preferred method of extracting hydrocarbons of interest from microalgae and other photosynthetic cultures.

[0023] The choice of extraction technologies will depend largely on the nature of the photosynthetic organism in culture. Organic-wall microalgae are very suitable to hexane solvent and enzymatic extraction. Living silica-wall microalgae (diatoms) however render their own cell walls extremely insoluble. In addition silica creates a physically strong and chemically inert protective covering since the cell walls cannot be attacked enzymatically. Silicon uptake and deposition by diatoms involves less metabolic energy expenditure than formation of equivalent organic walls resulting in faster growth rates than their organic-wall counterparts which makes diatoms attractive for high yield cultivation. Current extraction technologies however greatly inhibit diatom cultivation for oil production and favor organic wall microalgae cultivation. The silica cell structure of diatoms requires the use of cell disruption technologies that liberates the oils from the cultured organisms and allows for the

isolation of the high-quality silica (diatomite). A preferred cell disruption technology is hydrodynamic cavitation which can be applied effectively to both organic- and silica-wall photosynthetic organisms. Extraction results for different microalgae species at various volume densities indicate that hydrodynamic cavitation achieves near theoretical maximum extraction volumes (see Table 1).

Table 1
Extracted Volume vs. Theoretical Maximums

<i>Microalgae Species</i>	Density Volume		
	1.0%	3.0%	5.0%
<i>Chaetoceros</i>	96%	98%	97%
<i>Chlorella</i>	93%	95%	96%
<i>Scenedesmus obliquus</i>	92%	93%	94%
<i>Botryococcus braunii</i>	92%	94%	95%

Hydrodynamic Cavitation

[0024] Cavitation is the formation of partial vacuums in a liquid by a swiftly moving solid body such as a propeller or by high-intensity sound waves. The partial vacuums are used to rupture the photosynthetic organisms. A variety of examples of hydrodynamic cavitation devices are known in the art. Examples of suitable devices include U.S. Patent Application No. 2009/0192159, as well as U.S. Patent Nos. 6,279,611, 6,365,555, 6,846,365, 6,935,770, 7,086,777, 7,207,712, and 7,338,551, all of which are hereby incorporated by reference in its entirety.

[0025] In a preferred embodiment, a device for creating hydrodynamic cavitation in a fluid is utilized. Typically, the device includes a flow-through chamber having various portions and a plurality of baffles within one of the downstream portions of the chamber. One or more of the baffles is configured to be movable into an upstream portion of the chamber to generate a hydrodynamic cavitation field downstream from each baffle moved into the upstream portion of the chamber.

[0026] In another preferred embodiment, a device for creating hydrodynamic cavitation is utilized in which case the device creates hydrodynamic cavitation of the fluid stream by applying magnetic impulse. Magnetic impulse hydrodynamic cavitation offers a more uniform cavitation distribution over flow through chambers hydrodynamic cavitation for processing of liquids in a turbulent flow.

[0027] Cavitation (the formation, growth, and implosive collapse of gas or vapor-filled bubbles in liquids) can have substantial chemical and physical effects. While the chemical effects of acoustic cavitation (i.e., sonochemistry and sonoluminescence) have been extensively investigated during recent years, little is known about the chemical consequences of hydrodynamic cavitation created during turbulent flow of liquids.

[0028] Hydrodynamic cavitation is the formation of cavitation bubbles and cavities within a liquid stream or at the boundary of the streamlined body resulting from a localized pressure drop in the liquid flow. If, during the process of movement of the liquid, the pressure at some point decreases to a magnitude under which the liquid reaches a boiling point for this pressure (“cold boiling”), then a great number of vapor-filled cavities and bubbles are formed. These vapor-filled cavities and bubbles are called cavitation cavities and cavitation bubbles. Insofar as the vapor-filled bubbles and cavities move together with the flow, they then move into the elevated pressure zone. Then, almost instantaneously, vapor condensation takes place in the cavities and bubbles, and they collapse, creating very large pressure impulses. The magnitude of the pressure impulses within the collapsing cavitation bubbles may reach 150,000 psi. The result of these high-pressure implosions is the formation of shock waves that emanate from the point of each collapsed cavitation bubble. Such high-impact loads result in the breakup of any medium found near the collapsing cavitation bubbles. Collapse of a cavitation bubble near the boundary of phase separation of a liquid–solid particle in suspension results in the breakup of the suspension particles: A dispersion process takes place. Collapse of a cavitation bubble near the boundary of phase separation of a liquid–liquid type results in the breakup of drops of the disperse phase: Cavitation process takes place. Thus, the use of kinetic energy from collapsing cavitation bubbles and cavities is used in the described cavitation process to extract the oils from microalgae and to sterilize the growth medium for reuse.

[0029] The following is a description of one embodiment of a suitable cavitation device. As described in the art, a suitable cavitation device or apparatus is capable of producing appropriate bubbles that produce the cavitation effect. All components inside the apparatus are influenced by pressure impulses and advanced hydrodynamic cavitation. Suitable devices stimulate cavitation in hydrodynamic liquids to the point where the end result of processed fluid meets intended emulsification or dispersion criteria.

[0030] A particularly preferred embodiment comprises a nano-cavitation generator that utilizes flow-through nano-cavitation technology for producing biodiesel fuel. The nano-

cavitation generator will typically include a casing or housing that encloses a flow-through region. The flow-through region will typically comprise an inlet, a flowmeter passage, an intermediate coupling, a reaction chamber having an inlet and an outlet, a reaction chamber cover, and an outlet fitting.

[0031] The inlet is a fitting that passes through a portion of the housing. The inlet includes a coupling, whereby an external fluid line is connected to supply a fluid medium or other reaction components to the generator. The inlet is secured to the housing by a retaining ring which holds the inlet in place and provides sealing against leaks. The inlet fitting is connected to a flowmeter passage which includes a flowmeter to measure the flowrate of process fluids. The flowmeter passage is connected to an inlet of the reaction chamber by an intermediate coupling. The connection between the intermediate coupling and the inlet is sealed by an o-ring or other similar structure. The reaction chamber includes a reaction chamber passageway that connects the inlet to the outlet. The reaction chamber cover is connected to the reaction chamber and partially defines the reaction chamber passageway. The outlet fitting of the generator is integral with the reaction chamber cover.

[0032] The reaction chamber passageway defines a series of compartments having varying diameters and surface features. In a first preferred embodiment, the series of compartments in sequence from the inlet to the outlet are as follows: inlet compartment, constriction compartment, first reaction compartment, second reaction compartment, final reaction compartment and outlet compartment. A plasmator is positioned in the passageway through the constriction compartment and the first reaction compartment. The configuration and operation of the plasmator will be described below.

[0033] A number of the fittings and couplings in the generator are sealed using retaining rings, o-rings or similar structures. The outlet fitting includes an o-ring which forms a water-tight seal in the junction between the outlet fitting or reaction chamber cover and the reaction chamber. Another o-ring forms a water-tight seal in the connection between the reaction chamber and the intermediate coupling. The connection between the intermediate coupling and the flowmeter passage should also be sealed by an o-ring or similar structure, as well as the connection between the inlet fitting and the flowmeter passage. The inlet fitting is retained and sealed against the housing by a retaining ring as described above.

[0034] A pressure gauge is positioned in the housing adjacent the reaction chamber. A sensor from the pressure gauge enters the reaction chamber through an access passage. The pressure gauge and sensor are designed to measure the overall pressure in the reaction

chamber. As discussed elsewhere, the overall pressure of the reaction chamber should remain at about atmospheric pressure for the generator to operate as intended.

[0035] The nano-cavitation generator is static, i.e., contains no moving parts, and is configured for operation at a set fluid velocity and pressure of fluid medium. As described below, the changing of cavity diameters and surface features within the generator causes the generation of cavitation fluid features, i.e., bubbles and localized elevations of temperature and pressure. These localized elevations of temperature and pressure come in the form of eddies of internal temperature and pressure increases. The subsequent collapse of the cavitation bubbles and eddies is such that the outlet liquid stream is homogenized into a stable, ultra-thin emulsion or dispersion.

[0036] The inventive device creates nano-cavitation in fluids in a flow-through region between the fluid inlet fitting and the fluid outlet fitting. The flow-through nano-cavitation reactor is a multi-stage process whereby reaction components are manipulated through localized high temperature and pressure impulses and advanced nano-cavitation principles.

[0037] Fluid medium enters the generator at the inlet fitting as indicated by flow arrow. As described briefly above, the reaction chamber passageway comprises various compartments of varying diameter and internal surface features such that the cross-sectional area of each changes in relation to the previous compartment, the plasmator can be positioned in the junction between the constriction compartment and the first reaction compartment.

[0038] The plasmator can also comprise a constrictor plate having a stem topped by a conical cap. A series of orifices are positioned in the constrictor plate around the stem. The plasmator can be oriented such that the conical cap is centered in the constriction compartment to force the fluid medium to an outer circumferential flow path, i.e., the gap between the wall of the constriction compartment and the edge of the conical cap. The circumferential flow path provides a greatly reduced flow area compared to the open flow area of the inlet compartment. This greatly reduced flow area is thought to lead to the nano-cavitation process described above. The orifices in the constrictor plate 46 provide another point at which the available flow area is greatly reduced and the nano-cavitation process is increased. Finally, sequential compartments in the reaction chamber passageway vary the available flow area and then match the flow area of the inlet fitting.

[0039] Processed fluid medium exits the generator at the outlet fitting as indicated by flow arrow. The nano-cavitation process takes place in the reaction chamber, specifically the reaction chamber passageway. The design of the nano-cavitation generator and the theory

behind the fluid process taking place is based solely on the static mechanical and physical construction of the device, i.e., the changing diameters, flow areas and cross-sectional areas.

[0040] All reactions that take place in the nano-cavitation generator occur at ambient temperature. No agitation or mixing time is required. The nano-cavitation process is run at pressures between 100 psi and 1000 psi, ideally at around 500 psi. The nano-cavitation generator produces an instant reaction process, due to the bonding at the molecular level of free fatty acids (FFA) in the oil or fat with the reaction catalysts. The transesterification process is completed in seconds and finished product is produced immediately. Complete separation of finished biodiesel and glycerin can be achieved within 8-15 minutes via gravitational processes and instantly via centrifugal processes.

[0041] While processing vegetable oils, yellow grease, tallow and other animal fats (below 5% percent FFA content) with necessary components in a flow-through nano-cavitation reactor the molecules of FFA are broken apart in micro-explosions. Such micro-explosions result in instant glycerol separation, increased yield, decreased viscosity, increased cetane number, as well as, improvement of power parameters of produced fuel. The inventive generator also increases the effectiveness of any catalysts used in the reaction, as well as, the rate and efficiency of the esterification reaction. Thus, the inventive apparatus not only increases the quality and quantity of pure biodiesel fuel output but also its production rate.

[0042] Flow-through nano-cavitation is produced by pressure variations, which are obtained using the geometry of the passageways in the reactor creating variations in velocity and pressure. For example, based upon the geometry of the first preferred embodiment, an interchange of pressure and kinetic energy can be achieved resulting in the generation of cavities as in the case of the orifices in the constrictor plate. The cavitating conditions are generated just after the orifices in the reaction chamber passageway and hence the intensity of the cavitating conditions strongly depends on the number and geometry of the orifices.

[0043] When the reaction liquid passes through the orifices, the flow velocities increase due to the sudden reduction in the area offered for the flow, resulting in a decrease in the pressure. In the inventive device, the velocities are increased such that the localized pressure drops below the vapor pressure of the liquid medium under operating conditions and cavities are formed. Such cavities are formed at multiple locations in the reaction chamber. The location of formation strongly depends upon the number of compartments and the configuration of the same in the reaction chamber passageway. However, downstream of the orifices, due to an increase in the flow area, the velocities decrease giving rise to increasing

pressures and greater pressure fluctuations. The change in pressure and resultant pressure fluctuations control the different stages of cavitation, namely formation, growth and collapse.

[0044] The various devices known in the art makes it possible to accelerate the cavitation reaction causing bubbles to collapse and unite on a molecular level and allow for the production of biodiesel fuel without the addition of large amounts of energy and avoids high-pressure operation. The devices can produce biodiesel fuel using oils or fats. Soaps formed during base catalyzed transesterification are not present after the cavitation transesterification process has been completed, when provided with appropriate conditions. This simplifies the separation of the product phases and prevents the formation of emulsions if a water wash procedure is used for the finished fuel. The amount of water and FFA in the biolipids (oil or fat) are important parameters for the process and should be set using methodologies known to those of ordinary skill in the art to avoid unwanted side products.

Principle of operation of cavitation mixer-homogenizer reactor

[0045] In its simplest form, basic cavitation consists of the flow-through chamber, with cavitation generator located at the entry. The shape of the cavitation generator significantly affects the character of the cavitation flow and, correspondingly, the quality of dispersing. The optimal cavitation generator design is chosen in a multi-stage cavitator. In general, the cavitation generator works in the following manner. The stream of components to be processed under pressure P1 is charged with the aid of an auxiliary pump at the entry of the flow through chamber. Further, the stream flows around cavitation generator, after which, as a result of the localized pressure constriction, a cavitation cavity is formed. This cavity with its tail part comprises numerous bubbles. The cavitation bubbles flow with the stream to the exit of the flow through chamber into the elevated pressure zone P2. In this zone, the cavitation bubbles collapse, resulting in the dynamic influence on the emulsion drops, particles, or aggregate particles in suspension.

[0046] However, in the currently described process a precisely calculated engineered design is used in order to maximize the physical principle of a multi-stage hydrodynamic cavitation operation.

Advantage of multi-stage cavitation

[0047] Independent of the physical principle of its operation, the particle size achieved is dependent on one primary parameter in the process of dispersion—the level of energy

dissipation in the cavitation reactor and cavitation pump. The higher the level of energy dissipation in the cavitator chamber of the reactor, the smaller the particle size that can be achieved with any given medium.

[0048] The preferred multi-stage hydrodynamic cavitation reactor can achieve the smallest particle sizes. The level of energy dissipation in a cavitation reactor is mainly dependent on three vital parameters in the cavitation bubble field: the sizes of the cavitation bubbles, their concentration volume in the disperse medium, and the pressure in the collapsing zone. Given these parameters, it is possible to control the cavitation regime in the reactor and achieve the required quality of dispersion. These parameters are proprietary information.

[0049] In the above examples, the volume concentration of cavitation bubbles was on the order of 10%, which is at the low end of the concentration levels normally achieved in a cavitation reactor. By changing the type of cavitation in the reactor, it is possible to change the volume concentration of bubbles in the field from 10 to 60%, and their sizes from 10 to 1000 μm . The very high levels of energy dissipation produced during the collapse of a large number of cavitation bubbles allows the cavitation mixing pump and multi-stage hydrodynamic reactor to produce a very small particle size and very uniform particle size distribution. The results are produced at 500 psi operating pressures, which makes the equipment safe for a daily processing operation.

[0050] Magnetic impulse hydrodynamic cavitation creates cavitation bubbles in a turbulent flow of liquid by applying magnetic impulses which create the cavitation bubbles. Pressures created through magnetic impulse hydrodynamic cavitation are similar to those obtained through those created in flow-through baffle hydrodynamic cavitation devices, but the distribution of the cavitation is more uniform and predictable.

[0051] For the purposes of this patent, hydrodynamic cavitation is used to refer to hydrodynamic cavitation created in a continuous flow of liquid – whether created by a flow-through baffle device, a magnetic impulse device, or other similar device capable of creating hydrodynamic cavitation in a turbulent continuous flow of liquid without any moving parts.

Hydrodynamic Extraction

[0052] Preferably, the hydrodynamic cavitation technology described here is used to extract the oils produced by the cultivated photosynthetic organism. An advantage of this technology is that it eliminates the need for dewatering steps required in other extraction

processes. In one embodiment, after harvest, the harvested medium is directly subjected to hydrodynamic cavitation which disrupts the microalgae cell structure and extracts the oils from the microalgae cells. The resulting medium consisting of microalgae oil, microalgae cell biomass, and the harvested medium is flowed through to a separation process for separation. After separation the harvested medium can then be reused.

[0053] Another advantage of this technology is that it eliminates the need for harvesting steps. In another embodiment, a significant portion of the growth medium is directly subjected to hydrodynamic cavitation which disrupts the microalgae cell structure and extracts the oils from the microalgae cells. The resulting medium consisting of microalgae oil, microalgae cell biomass, and the harvested medium is flowed through to a separation process for separation. After separation the harvested medium can then be reused.

[0054] The oils and biomass produced from a first round of hydrodynamic cavitation can be subjected to subsequent rounds of hydrodynamic cavitation.

[0055] Hydrodynamic extraction enables the production of low-cost biofuels from microalgae oils because it is easily integrated into an economic and continuous process. The cost of hydrodynamic extraction using a 10 gallon/minute reactor is approximately \$0.002 per gallon of fluid processed which is several orders of magnitude smaller than the alternative combined costs of harvesting, de-watering, and existing extraction technologies. New higher flow-rate reactor designs will significantly bring down the costs. Furthermore hydrodynamic extraction does not require the addition and subsequent removal of costly additives or chemicals. Hydrodynamic extraction also enhances the adoption of diatoms for microalgae oil production.

Biofuel Products

[0056] Following extraction and processing, the oil, fats, fatty acids, triglycerides, etc. harvested from the microalgae and other photosynthetic organisms can be proceed to a variety of different useful products. For example, biodiesel can be produced from the products extracted from the cultivated organisms using standard techniques well know to those of ordinary skill in the art. For example, the production of biodiesel (fatty acid methyl esters) is well understood in the art. A discussion of such methods is provided in U.S. Patent Application No. 20090071064, which is hereby incorporated by reference.

[0057] According to some embodiments of the present invention, microalgae lipids are harvested and converted to biodiesel using transesterification. The hydrodynamic cavitation

devices described herein are effective to perform this conversion. In a particular embodiment, harvested lipids, etc., are subjected to further rounds of hydrodynamic cavitation to achieve the desired result. Further, after the biodiesel has been produced, it can be readily, energy-efficiently, and economically separated from the other chemicals in the reactor effluent using equipment common in the chemical industry.

Microalgae and Other Photosynthetic Organisms

[0058] The phrase "microalgae and other photosynthetic organism," as used herein, includes all algae capable of photosynthetic growth as well as photosynthetic bacteria. Eukaryotic algal strains are preferred for use with the disclosed methodology. Examples include *Botryococcene sp.*, *Chlorella sp.*, *Gracilaria sp.*, *Sargassum sp.*, *Spirolina sp.*, *Dunaliella sp.* (e.g., *Dunaliella tertiolecta*), *Porphyridum sp.*, and *Plurochrysis sp.* (e.g., *Plurochrysis carterae*). Diatoms, such as *Chaetoceros sp.* are particularly preferred algal strains for use with the presently described invention. These terms may also include organisms modified artificially or by gene manipulation.

[0059] *Chaetoceros* is particularly well suited for use with the presently described invention. There are over 400 species and subspecies known throughout the world. The growth rate of this organism is rapid, with 4 doublings per day, which permits cultures to be grown quickly. These organisms are known to have broad tolerances to temperature and salinity. *Chaetoceros* is also known to have a favorable lipid content and thus do not require manipulation to produce high quantities of oil.

Cultivation

[0060] The organisms selected for culture can be grown in open or closed systems. Open systems are preferred because they require less energy for maintenance and are typically more stable than closed systems. A preferred culture method for maintaining a dominant strain in culture using an open system is described in U.S. Patent No. 6,673,592, which is hereby incorporated by reference.

[0061] Briefly summarized, the cultivation system comprises a container for holding a culture medium. The culture medium includes an initial aqueous solution and a seed stock of photosynthetic organism. The initial aqueous solution is prepared such that optimal conditions for culturing photosynthetic organism of interest are established. Once the optimal conditions are established, the aqueous solution is inoculated with a seed stock of

photosynthetic organism. The resulting culture medium is pH controlled in a set range. A light source, preferably the sun, delivers light and heat to the culture medium, facilitating the growth of the photosynthetic organism culture. Periodically, a percentage of the photosynthetic organism culture medium is harvested. The harvested medium is replaced with a non-sterile medium, such as seawater. Alternatively the harvested medium can be replaced by culture medium from which the photosynthetic organisms have been harvested using the hydrodynamic methods disclosed herein. The method is continually repeated, thereby providing for uninterrupted harvests.

[0062] Optimal conditions for culturing a selected photosynthetic organism are typically established in the aqueous medium. Optimal conditions are those that allow a seed stock of photosynthetic organism to grow and outcompete predators, contaminants and other potential scavengers. Creating such a medium allows for the mass production of photosynthetic organism outdoors and under non-sterile conditions. Preferably, optimal conditions are attained in the aqueous medium by initially adjusting the concentrations of some or all of the following constituents: nitrogen, phosphorous, vitamin B₁₂, iron chloride, copper sulfate, silicate and Na₂EDTA. The pH of the culture medium is monitored, with adjustments, such as carbon dioxide treatments, performed to maintain the pH at a desired level.

[0063] In a preferred embodiment, the present system is used for culturing *Chaetoceros sp.* as the photosynthetic organism. The container holds an aqueous medium having the following starting characteristics: a carbon dioxide controlled pH of about 8.2, a starting nitrogen concentration of at least 3.0 mg N/liter, a starting phosphorous concentration of at least 2.75 mg P/liter, a starting vitamin B₁₂ concentration of at least 5 micrograms/liter, a starting iron chloride concentration of at least 0.3 mg/liter, a starting copper sulfate concentration of at least 0.01 mg/liter, a starting silicate concentration of at least 10 mg SiO₂/liter, and a Na₂EDTA concentration of 5 mg/liter. The medium is inoculated with a seed stock of *Chaetoceros sp.* photosynthetic organism and exposed to direct sunlight. The photosynthetic organism grows in the open environment and is periodically and continuously harvested. The harvested volume is replaced with a new seed stock of *Chaetoceros sp.* photosynthetic organism and culturing is repeated.

[0064] While any light source may be used in the present system, culturing the photosynthetic organism under full strength sunlight is the most economical option.

[0065] A percentage of the culture is periodically harvested. Preferably, about 60, 70, 80, 90, 95, or 99% of the culture volume is harvested at the conclusion of each period. In

preferred embodiments of the present system and method, the culture is harvested once a day, or approximately once every twenty-four hours. As sterile conditions are not required, the harvested volume is readily replaced with non-sterile seed stock of photosynthetic organism, such as seawater. Alternatively, the harvested volume can be replaced by medium subjected to hydrodynamic cavitation. Replaced of the harvested volume with treated medium is advantageous, particularly when some portion of the organisms present in the treated medium remain viable. The volume is preferably manually harvested or harvested using any acceptable harvesting machine or apparatus.

[0066] The container, which may have any acceptable dimensions and be constructed of any acceptable material, and preferably has an open top. Preferably large tanks are used as the containers. The tanks may be positioned above ground to permit sunlight to be passed through the sides of the containers. Alternatively, the tanks may be positioned within the ground. A transparent, light-passing cover may be positioned over the open top. In one embodiment, the cover is removably positioned over the open top.

[0067] By culturing photosynthetic organism in the optimal conditions, the production of large quantities of photosynthetic organism is possible in a cost effective manner. A single container is situated in an outdoor environment such that the contents of the container are directly exposed to natural light. No artificial light sources or additional transfer tanks are needed. Contaminants and predators are not a problem, as the established media conditions allow the photosynthetic organism to outcompete and overcome unwanted or detrimental species.

[0068] By establishing the optimal culture conditions for the photosynthetic organism, the present system provides for an environment where the photosynthetic organism out competes other species of photosynthetic organism from the culture. That enables the photosynthetic organism to be cultured continuously in large, outdoor containers using natural light. The need for labor intensive and costly systems designed to exclude other species from the culture is eliminated. The use of natural light greatly decreases the costs and problems associated with artificial lights.

[0069] The following examples are offered to illustrate but not to limit the invention.

Example 1

Hydrodynamic Extraction of oil from Harvested *Chaetoceros* Microalgae

[0070] *Chaetoceros sp.* is a microalgae diatom species that is particularly suited for fuel production because of desirable growth rates, growing conditions, and oil profile (e.g., lipid composition, lipid concentration as a percentage of mass).

[0071] Each day 90% of the culture volume was removed and stored in a harvesting tank. The culture in the harvesting tank was circulated through a foam fractionator column from evening until morning. Air was bubbled upward through the column from the bottom creating foam at the surface of the water that contained concentrated photosynthetic organisms. This foam was collected from the surface of the water. This foam upon condensing into a liquid contained approximately 3% dry matter content.

[0072] This harvested medium consisting of 10% of the culture volume with a 3% dry matter content was then directly flowed into a hydrodynamic cavitation reactor that processed 10 gallons per minute at 500 psi operating pressure for Hydrodynamic Extraction. Three hundred and Twenty (320) liters were processed in under 9 minutes. Total processing cost was \$0.17.

[0073] The hydrodynamic extraction extracted over 98% of the *Chaetoceros sp.* estimated ash free dry weight oil content and produced over 2.9 liters of microalgae oil at a cost of \$0.06 per liter of oil (\$0.22 per gallon of oil). This compares to \$2.91/liter for extraction using current technologies, not including the cost of the required de-watering steps.

[0074] The medium that then flowed out of the hydrodynamic extraction was directly flowed to a separation unit where the microalgae oil, the silica cell walls (diatomite), and the remaining fluid medium were separated. The separated microalgae oil can then be processed for use as biofuel or other product. The separated diatomite can be sold commercially. The separated fluid medium is then directly re-circulated back for cultivation.

Claims

1. A method of rupturing a microalgae cell wall, comprising:
providing a continuous flow of a fluid medium comprising one or more microalgae to a hydrodynamic cavitation device;
applying hydrodynamic cavitation in sufficient quantity to rupture one or more microalgae cells, whereby microalgae oil is released from the microalgae into the medium; and
extract the microalgae oil from the medium.
2. The method of claim 1, further comprising dewatering the medium.
3. The method of claims 1 or 2, wherein the photosynthetic organism is a diatom.
4. The method of claim 3, wherein the diatom is a *Chaetoceros* species.
5. The method of any one of the preceding claims, wherein hydrodynamic cavitation is applied using a multi-stage hydrodynamic cavitation reactor.
6. The method of any one of the preceding claims, wherein hydrodynamic cavitation is applied using a magnetic impulse cavitation reactor.
7. The method of any one of the preceding claims, wherein the processed medium after hydrodynamic cavitation is separated into components comprising microalgae oil, microalgae cell walls, and the processed fluid medium.
8. The method of any one of the preceding claims, wherein the microalgae oil is used for biofuel production.
9. The method of any one of the preceding claims, wherein the processed fluid medium is recycled for microalgae cultivation.
10. The method of any one of the preceding claims, wherein the processed medium is subjected to one or more additional rounds of hydrodynamic cavitation.

11. The method of any one of the preceding claims, wherein any of the separated microalgae oil is subjected to one or more additional rounds of hydrodynamic cavitation to produce transesterification.

12. The method of claim 7, wherein any of the separated components are subjected to one or more additional rounds of hydrodynamic cavitation.

13. A biofuel produced by the method of claim 11.

14. The biofuel of claim 13, wherein the biofuel is biodiesel.

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2009/060722

A. CLASSIFICATION OF SUBJECT MATTER
INV. C11B1/06 C11C3/04 C10L1/02

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
C11B C11C C10L

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, BIOSIS, COMPENDEX, FSTA

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X,P	WO 2009/063296 A2 (TANTILLUS SYNERGY LTD [GB]; VAN ALSTYNE DAVID C [GB]; VAN ALSTYNE LAYL) 22 May 2009 (2009-05-22) paragraph [0136]; claims 1,6	1,3,7-9, 13-14
X	WO 2008/089321 A2 (MCCALL JOE [US]) 24 July 2008 (2008-07-24) paragraphs [0002], [0008], [0014], [0049], [0052], [0053]; claims 5,6,7,9,17; figures 4,5	1-14
X	WO 01/46115 A1 (COMMW SCIENT IND RES ORG [AU]; MARCUCCIO SEBASTIAN MARIO [AU]; WALTER) 28 June 2001 (2001-06-28) example 1	1-2,7-8, 13-14
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Further documents are listed in the continuation of Box C.

See patent family annex.

* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
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