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[Continued on next page]

(54) Title: EXTRACTION OF LIPID FROM CELLS AND PRODUCTS THEREFROM

(57) Abstract: Processes for obtaining a lipid from a cell by lysing the cell, contacting the cell with a base and/or salt, and separating the lipid, as well as lipids prepared by said process are disclosed. Also disclosed are microbial lipids having a particular anisidine value, peroxide value, and/or phosphorus content.

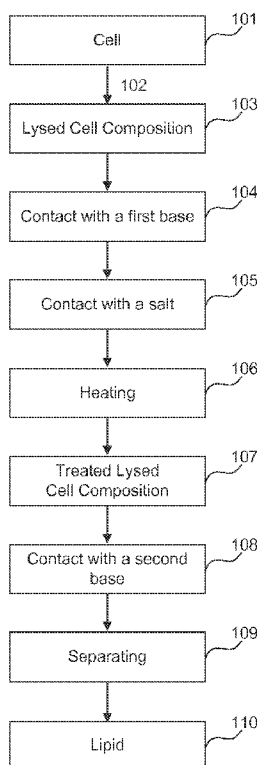


FIG. 1



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ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

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## INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 11/38768

## A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - C12P 7/64; C11B 1/00 (2011.01)

USPC - 435/134; 554/8

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)

IPC(8): C12P 7/64; C11B 1/00 (2011.01)

USPC: 435/134; 554/8

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 practicable, search terms used)

WEST (PGPB,USPT,EPAB,JPAB), Google Scholar, esp@cenet ; Search Terms: cell, lipid, pH, salt, less, solvent, absence, absent, impeller, tip, speed, demulsify, microbial, docosahexaenoic, docosapentaenoic, ratio, triglyceride, anisidine, peroxide, phosphorus, n-6, ppm, 3:1, 5:1, 6:1

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category*   | Citation of document, with indication, where appropriate, of the relevant passages   | Relevant to claim No.               |
|-------------|--|-------------------------------------|
| X<br>—<br>Y | US 2006/0099693 A1 (KOBZEFF et al.) 11 May 2006 (11.05.2006), abstract; para [0008], [0011], [0014], [0028], [0029], [0030].   | 1, 2<br>-----<br>3-5, 10-13, 37, 38 |
| X           | US 2008/0107791 A1 (FICHTALI et al.) 8 May 2008 (08.05.2008), para [0038], [0044], [0045], [0046], [0071], [0074], [0076], [0082], [0085], [0092].   | 51                                  |
| X<br>—<br>Y | US 2005/0115897 A1 (DUEPPEN et al.) 2 June 2005 (02.06.2005), abstract; para [0010], [0011], [0035], [0056], [0064], [0067], Table 2, Table 5.   | 52, 54, 55<br>-----<br>53           |
| Y           | WO 2008/088489 A2 (BIRSCHBACH et al.) 24 July 2008 (24.07.2008), abstract; pg 1, ln 3-6; pg 13, ln 23-31; pg 21, ln 30-pg 22, ln 3.  | 3-5, 10-13, 37, 38                  |
| Y           | ZHU et al. Extraction of lipid from sea urchin ( <i>Strongylocentrotus nudus</i> ) gonad by enzyme-assisted aqueous and supercritical carbon dioxide methods. Eur Food Research Technol. 20 January 2010, Vol 230, Pages 737-743, especially page 737, abstract; page 740, right col, second para. | 4, 5, 10                            |

 Further documents are listed in the continuation of Box C.

\* Special categories of cited documents:

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

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&amp;" document member of the same patent family

Date of the actual completion of the international search

11 November 2011 (11.11.2011)

Date of mailing of the international search report

14 DEC 2011

Name and mailing address of the ISA/US

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## INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 11/38768

## C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

| Category* | Citation of document, with indication, where appropriate, of the relevant passages              | Relevant to claim No. |
|-----------|---|-----------------------|
| Y         | US 2007/0138094 A1 (BOMBERGER et al.) 21 June 2007 (21.06.2007), abstract; para [0085], [0087]. | 38                    |
| Y         | EP 1419780 A1 (KYLE) 19 May 2004 (19.05.2004), abstract; para [0045], [0082].                   | 38                    |
| Y         | US 2008/0148433 A1 (METZ et al.) 19 June 2008 (19.06.2008), abstract, para [0220].              | 53                    |

## Continuation of Box (III): Lack of Unity

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

The special technical feature of the inventions listed as Groups I and II is an extracted microbial lipid, wherein the lipid contains less than 5% by weight of an organic solvent. This special technical feature fails to provide a contribution over the prior art, as evidenced by US 2008/0038800 A1 to Ruecker et al. (published February 14, 2008). Ruecker discloses a method for extracting lipids from microbes (para [0023] - "method for obtaining lipids from microorganisms"), wherein the lipid contains less than 5% by weight of an organic solvent (para [0023] - "using a substantially organic solvent free extraction process, i.e., a "solventless" extraction process . . . The term "solventless extraction process" refers to an extraction process which when an aqueous solvent is used, the aqueous solvent comprises less than about 5% of an organic solvent, preferably less than about 4%, more preferably less than about 2%, and most preferably less than 1%."). In the absence of a contribution over the prior art, the shared technical feature is not a shared special technical feature. Furthermore, while the process of Group I can be used to extract the microbial lipid composition of Group II, it is readily apparent that the microbial lipid composition of Group II may be extracted by a variety of methods not limited by any special feature to the process of Group I.

Unity of invention exists only when the same or corresponding technical feature is shared by the claimed inventions. Without a shared special technical feature, the inventions of Groups I and II lack unity of invention.

## INTERNATIONAL SEARCH REPORT

International Application No.

PCT/US 11/38768

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

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

1.  Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:
  
2.  Claims Nos.:  
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
  
3.  Claims Nos.: 6-9, 14-36, 39-50, 56-59  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

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

This International Searching Authority found multiple inventions in this international application, as follows:  
This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees must be paid.

Group I: Claims 1-5, 10-13, and 37-38, drawn to a process for obtaining a lipid from a cell.

Group II: Claims 51-55, drawn to a microbial lipid composition.

----- Please see extra sheet for continuation -----

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

**Remark on Protest**

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