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(54) IMPROVED MICROALGAL FLOUR

VERBESSERTES MIKROALGENMEHL

AMÉLIORATIONS APPORTÉES À DE LA FARINE DE MICROALGUES

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(73) Proprietor: **Corbion Biotech, Inc.**
South San Francisco, CA 94080 (US)

(72) Inventors:
- **NORRIS, Leslie**
  South San Francisco, CA 94080 (US)
- **PIECHOCKI, John**
  South San Francisco, CA 94080 (US)
- **BALIU, Enrique**
  South San Francisco, CA 94080 (US)
- **DESAI, Ruchir**
  South San Francisco, CA 94080 (US)
- **RUYET, Marie, Le**
  F-62400 Bethune (FR)
- **PATINIER, Samuel**
  F-59000 Lille (FR)
- **PASSÉ, Damien**
  F-59500 Douai (FR)
- **DRUON, Amandine**
  F-59800 Lille (FR)

(74) Representative: **Carpmaels & Ransford LLP**
One Southampton Row
London WC1B 5HA (GB)

(56) References cited:
WO-A1-2010/120923
WO-A2-2010/063031
WO-A2-2011/130578
WO-A2-2011/130578
US-A1- 2010 297 296
US-A1- 2010 297 296
US-A1- 2010 303 989

- **MÓNICA FRADIQUE ET AL: "Incorporation of Chlorella vulgaris and Spirulina maxima biomass in pasta products. Part 1: Preparation and evaluation", JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE, vol. 90, no. 10, 15 August 2010 (2010-08-15), pages 1656-1664, XP055027243, ISSN: 0022-5142, DOI: 10.1002/jsfa.3999**

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(Cont. next page)

- **KIRCHHOFF ET AL.: 'Quantitation of Odor-Active Compounds in Rye Flour and Rye Sourdough Using Stable Isotope Dilution Assays' JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY vol. 50, 15 August 2002, pages 5378 - 5385, XP055278633**
- **FRADIQUE ET AL.: 'Incorporation of Chlorella vulgaris and Spirulinamaxima biomass in pasta products. Part 1: Preparation and evaluation' JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE vol. 90, no. ISS. 1, 13 May 2010, pages 1656 - 1664, XP055027243**
- **SZABO ET AL.: 'Safety evaluation of a high lipid Whole Algalin Flour (WAF) from Chlorella protothecoides' REGULATORY TOXICOLOGY AND PHARMACOLOGY vol. 63, 28 March 2012, pages 155 - 165, XP055278652**

Description

Technical Field

[0001] The present invention relates to microalgal flour with improved flavor and methods of producing the flour.

Background

[0002] As the human population continues to increase, there is a growing need for additional food sources, particularly food sources that are inexpensive to produce but nutritious. Moreover, the current reliance on meat as the staple of many diets, at least in the most developed countries, contributes significantly to the release of greenhouse gases. There is a need for new foodstuffs that are less harmful to the environment to produce.

[0003] Requiring only "water and sunlight" to grow, algae have long been looked to as a potential source of food. While certain types of algae, primarily seaweed, do indeed provide important foodstuffs for human consumption, the promise of algae as a foodstuff has not been fully realized. Algal powders made with algae grown photosynthetically in outdoor ponds or photobioreactors are commercially available but have a deep green color (from the chlorophyll) and a strong, unpleasant taste. When formulated into food products or as nutritional supplements, these algal powders impart a visually unappealing green color to the food product or nutritional supplement and have unpleasant fish, seaweed or other flavors.

[0004] There are several species of algae that are used in foodstuffs today, most being macroalgae such as kelp, purple laver *(Porphyra*, used in nori), dulse (*Palmaria palmate)* and sea lettuce (*Ulva lactuca).* Microalgae, such as Spirulina (*Arthrospira platensis)* are grown commercially in open ponds (photosynthetically) for use as a nutritional supplement or incorporated in small amounts in smoothies or juice drinks (usually less than 0.5% w/w). Other microalgae, including some species of *Chlorella* are popular in Asian countries as a nutritional supplement.

[0005] Poor flavor is a major factor that has impeded the widespread adoption of microalgae in food. WO2010/12093 discloses methods of making and using microalgal biomass as a food. That reference discloses the growth of microalgae in the dark, to produce a microalgal biomass.

[0006] However, further improvements in flavor of microalgal biomass should promote further adoption.

Summary

[0007] The present invention relates to microalgal flour with acceptable sensory characteristics and a method of producing the flour. The flour can be produced by cultivating microalgal cells of a strain of *Chlorella protothecoides* under conditions of acceptable pH and dissolved oxygen to produce a desired amount of lipid. The microalgal cells can be lysed, heat-treated, washed and dried to produce a microalgal flour that can be incorporated into a variety of products.

[0008] The present invention provides a microalgal flour suitable for use in food, the flour comprising microalgal cells of *Chlorophyta* and characterized as set out in claim 1. Preferred features of the microalgal flour of the present invention are set out in dependent claims 2 to 13. The present invention also provides a method of preparing a microalgal flour as set out in claim 14.

Brief Description of the Drawings

[0009] The features of the invention will be more readily understood by reference to the following detailed description, taken with reference to the accompanying drawing, in which:

Fig. 1 shows a flow diagram depicting a method of producing a flour in accordance with the present invention; and

Fig. 2 shows a PCA clustering analysis with points representing microalgal flour samples with acceptable and inferior flavor.

Detailed Description of the Invention

Definitions

[0010] In connection with a culture medium, "dissolved oxygen", abbreviated as "DO", means the relative oxygenation of the culture medium as compared to the oxygenation of a culture medium that is in oxygen equilibrium with the atmosphere.

[0011] A "microalgal flour" is a dry, particulate composition, fit for human consumption, comprising cells of microalgae.

[0012] As used herein, an "off-flavor" means a flavor that a consumer would not expect and/or is undesired in a food,

for example a baked food, such as a cake. Examples of off-flavors include flavors of cabbages or fish. Although specific flavors may be measured by modern analytical techniques such as Gas Chromatography-Mass Spectrometry (abbreviated as GC-MS), often the most convenient and effective tool for measuring off-flavors is a tasting panel comprised of humans. In connection with human perception of off-flavors, these may be determined by a sensory panel of, for example, 10 people, where absence of a flavor or odor is established when 2 or fewer of the 10 people can detect the flavor, or by performing enough tests to establish statistical significance.

Overview

[0013] The present invention is rooted in the discovery that certain strains of microalgae can produce an appetizing biomass in terms of flavor, odor and color, when cultivated and processed under particular conditions. The improved flavor is believed to result not just from the absence of off-flavors but from the presence of desirable flavor compounds produced during cultivation and/or processing. In the Examples below, the microalgae is a strain of *Chlorella protothecoides* cultivated heterotrophically, in the dark, but could be another species of *Chlorella* or other species of Chlorophyta, provided that a non-green color can be produced via heterotrophic cultivation and careful processing such as by using the methods given below. By use of these techniques, the product may fall within the newly identified acceptability criterion disclosed here.

[0014] Human sensory panel data on multiple batches of microalgal flour was correlated with data from an extensive analysis of flavor and odor compounds of varying solubility in water to identify a clustering in flavor/odor space as represented by a principal component analysis. Thus, a microalgal flour that falls within the identified cluster has a high probability of being acceptable for human consumption.

[0015] Fig. 1 is a flow diagram of a process for producing microalgal flour having low amounts of off-flavors, in accordance with the invention. The resulting flour may be incorporated into a variety of foods and beverages.

[0016] Fig. 2 is a plot showing a representative PCA clustering analysis with points representing microalgal flour samples with acceptable and inferior flavor.

Process for the production of improved microalgal flour

[0017] *Chlorella protothecoides* microalgae are cultured (step 105) in the dark. It has been found that culturing the microalgae in the dark creates microalgal biomass having lower levels of off-flavors such as mushroom and cabbage or fish flavors; e.g. when microalgal flour dispersed in deionized water at 10% (w/v) and evaluated by a human sensory panel. Thus, the microalgae are cultured heterotrophically, in the dark on glucose as a fixed (i.e. non-CO_2) carbon source. While glucose is used in the method of claim 14 and in the examples below, other fixed carbon sources such as fructose, sucrose/fructose mixtures, or acetic acid/acetate may produce comparable results and can be used to produce the flour of claim 1. The sugar concentration can be controlled by continuous feeding. Favorable results have been achieved with a glucose concentration of between 3 and 10 g/l.

[0018] To produce the flour of claim 1, suitable genera of microalgae include *Chlorella* and *Prototheca*. For example, *Chlorella protothecoides, Prototheca moriformis* or *Prototheca zopfii* may be used. Other species of *Chlorella* used for human nutrition, such as *Chlorella protothecoides,* can also be grown and processed as disclosed here. Combinations of microalgal species or strains may also be used. Optionally, the microalgal cells are mutated and a strain selected to be substantially reduced in pigment that may change the color of a food product into which the biomass is incorporated. In the examples below, it was found that suitable flavor and no observable green color could be obtained from cells of *Chlorella protothecoides.* For example, the flour may comprise less than 200, 20 or 2 ppm of chlorophyll. In the examples below, the color was found to be yellow/gold, but could also be, for example, pale-yellow, off-white or white, depending on the strain and cultivation/processing conditions used.

[0019] In the process of claim 14, the microalgae are cultured to a desired density and lipid concentration. The lipid concentration may be increased by culturing under nutrient-limiting and especially nitrogen-limiting conditions. Culturing may be performed under conditions of limiting nitrogen so that the microalgae reach 10-20%, 20-30% 40-50%, 40-60%, 30-70%, 35-75%, 50-60%, 60-70% or 70-85% lipid, as measured by dry cell weight. In the exemplified embodiments, the microalgae comprise about 50% lipid. Elevated levels of lipid are especially useful in producing food products with improved fat and cholesterol profiles or improving the mouthfeel of such products. When a high lipid microalga is used to produce the flour, the stickiness of the lipid can be an impediment to forming a flour that is measurable and/or flowable. Alternatively, cultivation under nitrogen-replete conditions can give a high-protein microalgal flour. Such a flour can have, for example 5-20% or 10-18% lipid by dry cell weight. As described below, drying methods have been identified that give a flowable powder while retaining the desirable taste, odor and color characteristics.

[0020] The microalgae may be cultured in an opaque culture vessel. The microalgae may be cultured under aerobic conditions. Surprisingly, it has been found that increasing the oxygen level to 30% DO or more during heterotrophic culture of *Chlorella protothecoides* can result in a microalgal biomass having improved flavor. Variation of ±30% in DO

(i.e. 30±9%DO) is contemplated. In addition, elevated oxygen (e.g. >40% DO, >50% DO, >60% DO or 60-70% DO) during fermentation can result in a microalgal biomass having a white or off-white color with low amounts of off-flavors. Whiteness may be measured with a Hunter colorimeter. In an embodiment, the whiteness is greater than the whiteness of a control sample of microalgal biomass grown at about 30-40% DO. In a specific embodiment, the oxygen is elevated to about 60-70% dissolved oxygen. Increased oxygenation can be achieved, for example, by the introduction of purified oxygen.

[0021] The flavor may be improved by culturing the microalgae at a desired pH. In the process of claim 14, the starting pH is 6.8. For example, the pH could be from 4 to 9 or from 5 to 8. The pH may be controlled using buffering and/or pH monitoring with titration. If an acidic pH is used, the pH can be neutralized by adjusting to a pH of 6 to 8, 6.5 to 7.8 or about 7; e.g. prior to drying to avoid astringent flavor. The final flour may be characterized by a pH of 5.5-8.5, 6.0-8.0 or 6.5-7.5 for a 1% w/v solution of flour in water.

[0022] After culturing, the microalgae are inactivated (step 110). Inactivation conditions are chosen to be sufficient to inactivate enzymes that produce off-flavors. These conditions may also kill the microalgae or stop growth of the microalgae and contaminating species, if any. It has been found that rigorous pasteurization (i.e. at high temperature and/or long times) can lead to undesirable flavor/odor, while treatment that is not rigorous enough also can lead to unacceptable flavor/odor. Thus, when pasteurization is used, a delicate balance must be struck. Experiments have shown that, according to the process of claim 14, a high-temperature-short-time pasteurization ("HTST") treatment regime can be used to produce an acceptable microalgal biomass product. For example, the temperature of the treatment may be from 70°C to 95°C or 72°C to 90°C for from 10 to 180, 30 to 120 or 45 to 90 seconds. In the process of claim 14, microalgae are treated at 75°C for 1 minute, optionally by flowing the cultured microalgal broth through a heat exchanger into a collection vessel. Cooling of the HTST output is preferred to avoid prolonged heating. Similar results should be obtainable by adjustment of both time and temperature. Delay prior to inactivation should be minimized so as to prevent the development of off-flavors, which are believed to be created by enzyme activity. Thus, in an embodiment of the present invention, the step of inactivating enzymes is performed without delay of a time sufficient to allow production in the microalgae of enzymatically developed off -flavors. Culture at an acidic pH may also allow for an even more gentle pasteurization to be used. For example, the microalgal cells can be cultured at a pH of from 5 to 6.5, followed by pasteurization at from about 60 to about 70°C for 1 minute and neutralization prior to drying.

[0023] To further improve flavor, the microalgal cells may be washed (step 115). Without wanting to be bound by theory, the washing may remove off-flavors. In addition, using an inactivation step prior to washing may permeabilize the cells or otherwise promote the removal of unwanted flavors or odors from the microalgal biomass. Washing may be performed by centrifugation, filtration, dialysis or other method known in the art. Optionally, the washing is performed with a volume of wash liquid (e.g. water or buffer) that is as great or greater than the volume of the microalgal cells (e.g. as measured by centrifugation). The volume of wash liquid may be twice the volume of the cells or, preferably, at least 3 times the volume of the cells. In the method of claim 14, centrifugation in 6.4 times the cell volume gave a microalgal biomass with favorable flavor. To produse a flour according to claim 1, the cells may be washed with between 3 and 12 volumes of water. For these purposes, measurement of the cell volume is accomplished by dewatering the cells (i.e. removing them from the liquid growth medium). For example, the cells may be dewatered by centrifugation or filtration. Optionally, the washing step may be repeated one or more times.

[0024] Optionally, after washing, a preservative may be added (step 120). For example, sodium benzoate and/or potassium sorbate may be added as a bacteriostatic and fungistatic agent. Since sodium benzoate is more active under acidic conditions, the pH may be lowered as necessary. In that case, the pH can be raised later in the process to avoid an unwanted acidic flavor.

[0025] In the process of claim 14, the microalgal cells are then lysed (step 125) by milling. The lysis may be partial, or complete. For example, from 5% to 95% or a majority (>50%) of the cells may be lysed. Lysis may be especially desirable to release lipids in a high-lipid microalgae, where release of the lipids improves the quality or nutritional value of a food product into which the microalgal biomass is incorporated. Lysis may be accomplished with a bead mill. Optionally, a majority of the cells can be lysed. In one embodiment, about 30-75% of the microalgal cells are lysed. In another embodiment, about 30-75% of the microalgal cells are lysed and the microalgal cells have about 30-75% lipid by dry cell weight. In yet another embodiment, the microalgal cells are 60-90% lysed. This combination of parameters is believed to lead to a microalgal biomass that improves the mouthfeel, air-holding capacity or other functional parameters of a food into which it is integrated, while avoiding difficulties in drying or other processing steps that may be associated with highly lysed cells. In Example 3 below, the cells were lysed to about 80%.

[0026] Optionally, the biomass may be homogenized (step 130). For example, the suspension containing the cells and/or lysed cells may be forced through a narrow channel or orifice at elevated pressure (i.e. use of a high-pressure homogenizer). Other types of homogenizers such as blade or ultrasonic homogenizers may also be employed.

[0027] In the process of claim 14, an antioxidant is added to enhance the shelf life of the biomass (step 135). For example, tocopherols, BHA, BHT, rosemary extract or other suitable food-grade antioxidants can be used. In addition to enhancement of shelf life, addition of antioxidant at the stage may prevent unwanted oxidation flavors from forming

in the drying step. At this stage, addition of a base to raise the pH may prevent astringent flavors associated with a low pH if low pH conditions were used in upstream processes.

[0028] Prior to drying (e.g. after homogenization and before or after the optional addition of antioxidant), the microalgae can be held at elevated temperature for a period of time (140). Without wanting to be bound by theory, it is believed that this step promotes stability of the flavor, ensures inactivation of enzymes and may promote the formation of positive flavors. For example, a suspension of lysed microalgae can be held at 70-85° for 1-6 minutes. In the Example 3 below for which acceptable sensory properties were obtained in the flour produced, this heating step was performed at 77°C for 3 minutes. Comparable results may be obtained, for example, by heating at about 87°C for about 90 seconds or about 67°C for about 6 minutes.

[0029] In the process of claim 14, the biomass is then spray dried (step 145). The spray drying may use, for example, a box-dryer, a tall-form spray-dryer, a fluidized bed dryer or a moving fluidized bed dryer (e.g. a FilterMat® spray dryer, GEA Process Engineering, Inc.). Example 3 describes conditions used for drying with a FilterMat drier.

[0030] The resulting flour may be measureable or flowable, even if high in lipid (e.g. 30-70 or 40-60% lipid by dry cell weight). In a specific embodiment, the flour has an aerated density of 0.30 to 0.50, a bulk density of 0.50 to 0.65, an oversize of 15-35% by weight at 2000 μm (i.e. % too large to pass through a 2000 μm sieve), 40-70% at 1400 μm and 1-20% at 800 μm, a wettability of 1-25 mm and a surface area of 0.1 to 0.7 m^2/g.

[0031] To test wettability:

- introduce 500 ml of deionized water at 20°C into a 600 ml squat-form beaker (Fisherbrand FB 33114);
- place 25 g of the microalgal flour powder uniformly at the surface of the water, without mixing;
- observe the behavior of the powder after 3 h of contact; and
- measure the height of the product that has penetrated the surface of the water and settled at the bottom of the beaker.

[0032] The aerated bulk density is determined using a conventional method of measuring aerated bulk density, i.e. by measuring the mass of an empty container (g) of known volume and by measuring the mass of the same container filled with the product to be tested.

- The difference between the mass of the filled container and the mass of the empty container, divided by the volume (ml), then gives the value of the aerated bulk density.
- For this test, the 100 ml container, the scoop used for filing and the scraper used are supplied with the apparatus sold by the company Hosokawa under the trademark Powder Tester type PTE.
- To perform the measurement, the product is screened through a sieve with apertures of 2000 μm (sold by SAULAS). The density is measured on the product that is not retained on that screen.

[0033] The specific surface area is determined over the whole of the particle size distribution of the microalgal flour granules, e.g. by means of a Quantachrome specific surface area analyzer based on a test for absorption of nitrogen onto the surface of the product subjected to the analysis, carried out on a SA3100 apparatus from Beckmann Coulter, according to the technique described in the article BET Surface Area by Nitrogen Absorption by S. BRUNAUER et al. (Journal of American Chemical Society, 60, 309, 1938).

[0034] The microalgal flour is tested for acceptable flavor, color, odor and/or mouthfeel (step 150). For example, a human sensory panel may be employed and/or analytical technology such as headspace GC-MS, SPME or SBSE. Optionally, the flavor may be evaluated to determine if it is grouped with or falls within boundaries associated with acceptable flavor determined by a prior sensory panel and/or analytical testing. The groupings/boundaries may be determined with the use of principal component analysis (PCA) (see Examples below). An acceptable lot may then be selected for packaging and future use.

[0035] After drying and optional testing, the biomass may undergo any further processing or packaging (step 155) needed to make a microalgal flour or a food product that incorporates the biomass. For example, to make microalgal flour, the biomass may be agitated or passed through a screen. The microalgal flour may also be mixed with other ingredients to make a soup, sauce, dough, cake, cookie, dry baked-good mix etc. Testing can also be performed according to Examples 4, 5 and 8 below.

[0036] In accordance with embodiments of the invention, any two or more of the above-mentioned techniques can be combined to reach a heretofore unprecedented flavor in a microalgal flour. For example, HTST treatment followed by washing with liquid as described above can produce microalgal flour having low off-flavor. Oxygenation during cultivation and other steps as described above may further improve the flavor.

[0037] By selecting an appropriate microalgal strain and using the methods disclosed herein, a microalgal flour made from the biomass having acceptable sensory characteristics may result. The microalgal flour may be non-green and have undetectable levels of fish, mushroom or cabbage flavors or odors when diluted in water at a ratio (by volume) of 1:2, 1:5, 1:10, 1:20, 1:30 or 1:40. In an embodiment, off-flavors of fish and cabbage are undetectable when diluted 1:20

by volume in water, as detected by a tasting panel.

[0038] The following flavor/odor compounds were determined by the methods of Examples 4 or 5 and are believed to correlate with acceptable sensory testing: undecalactone (400-1800ppb), 3-methyl butanal (0-11,000ppb), pentanal (160-10,700ppb), 2-methyl butanal (0-2500ppb), 2-pentanone (39-10,600ppb), 3-pentene-2-one (0-1500ppb).

[0039] Acceptable samples also had less than threshold amounts of pyrrole, pyrazine or pyridinecontaining compounds, while these compounds were found in the sample of *Chlorella vulgaris* obtained from www.nuts.com, which was green and unacceptable in flavor and odor.

[0040] In an embodiment, the microalgal flour produced by the methods described above retain the low amounts of off-flavors mentioned for at least 2 weeks, 1 month, 3 months or 6 months when stored in the dark at room temperature in moisture- and oxygen- impermeable packaging (e.g. a Mylar® food storage bag).

[0041] Optionally, larger particles, granules or pellets can be made from the dried microalgal material. For example, the flour can be agglomerated, granulated, extruded or pelletized using a variety of methods known in the art.

Example 1. Production of microalgal flour at low pH and using a low-pigment strain.

[0042] Multiple fermentations of *Chlorella protothecoides* were performed at scales ranging from 7 L to 1000 L. Two strains of *Chlorella protothecoides* were used: strain A and strain B, a low-pigment mutant. Fermentation was performed in the dark on glucose as a fixed carbon source at a pH of about 5 to 6. After fermentation, the fermentation broth containing the microalgae was heat treated to inactivate the microalgae, immediately diluted with excess water and centrifuged to wash and concentrate the microalgae. The cells were lysed by milling, then spray-dried to make a microalgal flour. The microalgal flour made from Strain A was light yellow in color and the microalgal flour made from strain B was tan in color. A fermentation of strain B was also performed at about neutral pH.

Example 2. Low-color flour using high oxygen conditions.

[0043] Strain B was cultivated in at high (about 60%-70%) and low (about 30-40%) levels of dissolved oxygen and treated as in Example 1 to form microalgal flour. For the high oxygen experiment, reduced yellow color was noted in the broth, centrifuged biomass and in the final flour as compared to the microalgae produced at lower oxygen.

Example 3: Production of improved microalgal flour.

[0044] A seed culture of *Chlorella protothecoides* was added to a defined medium broth to give 9,000 L of culture. Heat-sterilized glucose (55% w/w) was used as a carbon source. Dissolved oxygen was held to a minimum of 30% by controlling aeration, backpressure and agitation in the fermentor. The cultivation temperature was 28°C. The pH of the broth was 6.8 at the start of cultivation and dropped to about 6 over the course of cultivation. Glucose was fed to a concentration of 3-10 g/L concentration. Growth was continued over 4-5 days to the mid-log-phase as measured by OD750. The resulting product had a dry cell weight (DCW) of 18.5% w/v. The nitrogen level in the growth medium was limiting to force the microalgae to accumulate approximately 50% lipid as a result of extended sugar feeding.

[0045] The broth was then heat-treated by online HTST at 75°C for 1 min, cooled to 6.2°C and then stored at 7°C. The HTST-treated broth was then washed by 6.4-fold dilution in decarbonated water and centrifuged using an Alfa Laval FEUX 510 centrifuge.

[0046] The pH was lowered to pH to 4.1 with 75% phosphoric acid and 500 ppm sodium benzoate /1000ppm potassium sorbate (on dry basis) were added as a preservative.

The material was then stored under agitation below 10°C.

[0047] Lysis was accomplished by milling in a NETZSCH LME500 bead mill using 0.5 mm zirconium silicate beads to give 88% cell disruption. The outlet was cooled to 6°C.

[0048] Ascorbic acid (150 ppm on a dry basis) and mixed tocopherols (500 ppm on a dry basis) were added to the material to prevent oxidation. Potassium hydroxide was added to neutralize the pH.

[0049] The material was then heated to 77°C for 3 minutes.

[0050] Drying was accomplished on a Filtermat FMD125 drier with a cyclone. The nozzle pressure was 160-170bar.

Example 4: SPME (Solid Phase Micro Extraction)

[0051] Samples (500 mg) plus 3 mL distilled water plus 1gm NaCl plus 5 μL 0.022 μg/uL 2-undecanone in ethanol internal standard were incubated at 50°C for 10 min and then extracted by SPME at 50°C for 20 min while stirring with the orbital shaker of the Gerstel MPS2. The SPME fiber used was DVB/CAR/PDMS (Divinylbenzene/Carboxen/Poly-

dimethylsiloxane), df 50/30 μm The fiber was desorbed at 260°C in the Agilent split/splitless injector for 3 min. Volatiles were desorbed into a Leco Pegasus GC-TOFMS and separated on a DB5-MS column (30m, 0.25 mm, 0.25 um) with helium carrier gas flow at 1.0 mL/min. The initial column temperature was 40°C (for 3 min) and then increased to 270°C at 15°C/min and held at 270°C for 5 min. Mass detection was performed in the electron impact mode (EI). All injections were splitless. Peak identification is based on comparison of EI mass spectra in samples to EI mass spectra of the NIST Library. Data is reported as relative concentration compared to the internal standard expressed in ppb.

Example 5: SBSE (Stir Bar Sorptive Extraction)

[0052] Samples (500 mg) plus 10 mL distilled water plus 5 μL 0.022 μg/μL 2-undecanone internal standard in ethanol were extracted for 1hr while stirring at 1000 rpm using a 2cm Gerstel PDMS Twister. One gram of NaCl was then added to the sample and extraction was continued for another hour. The technique is known as sequential SBSE. The Twister is then removed from the sample, rinsed with distilled water, patted dry with a lintless cloth and thermally desorbed in a Gerstel TDU used in the splitless mode. With the TDU, desorbed volatiles were initially trapped at -100°C; the volatiles trapped on the Twister were then desorbed at 280°C for 3 min. Volatiles were desorbed into an Agilent GC-MSD and separated on a DB5-MS column (30m, 0.25 mm, 0.25 μm) with helium carrier gas flow at 1.0 mL/min. The initial column temperature was 40°C (for 3 min) and then increased to 270°C at 10°C/min and held at 270°C for 5 min. Mass detection was performed in the electron impact mode (EI). All injections were splitless. Peak identification is based on comparison of EI mass spectra in samples to EI mass spectra of the NIST Library. Data is reported as relative concentration compared to the internal standard expressed in ppb.

Example 6: Flavor/Odor Data for Acceptable Sample of Example 3

[0053] The sample produced in Example 3 was tested by sensory panel and analyzed by SPME and SBSE as in Examples 4 and 5. The results are reported in the table below in units of parts per billion, determined relative to the 2-undecanone internal standard. In the tables below, a is used to represent alpha, d for delta, g for gamma. CAS numbers for the compounds are listed in Example 7.

Chemical	Mean relative concentration
Dimethyl.sulfide	0
2.3.Butanedione	248
Butanal	9.5
Propanal..2.methyl.	75
Furan..3.methyl.	67.5
Ethyl.Acetate	1671.5
2.Butenal...E..	47.5
Butanal..3.methyl.	0
1.Butanol	26
Butanal..2.methyl.	0
Thiophene	0
1.Penten.3.ol	0
1.Penten.3.one	7
2.Pentanone	38.5
2.3.Pentanedione	688.5
Pentanal	2876
Furan..2.ethyl.	2
Thiazole	0
3.Penten.2.one	7.5
Disulfide..dimethyl	42
2.Pentenal...E..	89.5
Pyrrole	0
Oxazole..4.5.dimethyl.	0
2.Penten.1.ol...Z..	0
Thiophene..3.methyl.	68.5
Hexanal	16198

(continued)

Chemical	Mean relative concentration
4.Methylthiazole	0
Pyrazine..methyl.	0
Furfural	0
Oxazole..trimethyl.	0
Butanoic.acid..3.methyl.	0
Butanoic.acid..2.methyl.	0
2.Hexenal	0
1.Hexanol	0
4.Heptanone	415
Pyridine..2.6.dimethyl.	0
Thiazole..2.4.dimethyl.	0
3.Heptanone	174
2.Heptanone	104
3.Heptanol	2426.5
Heptanal	700.5
Methional	0
Pyrazine..2.5.dimethyl.	0
Pyrazine..2.6.dimethyl.	0
Pyrazine..ethyl.	0
Pyrazine..2.3.dimethyl.	0
Pyrazine..ethenyl.	0
Thiazole..4.5.dimethyl.	0
2.Heptanone..6.methyl.	0
Hexanal..2.ethyl.	75
2.Heptenal...Z..	493
5.Nonen.2.one	0
2.Furancarboxaldehyde..5.methyl.	0
Benzaldehyde	231
hexanoic.acid	38.5
1.Octen.3.ol	173
Dimethyl.trisulfide	0
2.5.Octanedione	87.5
5.Hepten.2.one..6.methyl.	107.5
Furan..2.pentyl.	1.5
2.4.Heptadienal...E.E..	0
Pyrazine..2.ethyl.6.methyl.	0
Octanal	1067
Pyrazine..trimethyl.	0
Pyrazine..2.ethyl.3.methyl.	0
2.4.Heptadienal...E.E...1	13.5
Pyrazine..2.ethenyl.6.methyl.	0
1.Hexanol..2.ethyl.	11445.5
3.Octen.2.one...E..	0
2H. Pyran .2.one..5.6.dihydro.	1472
Benzeneacetaldehyde	0
3.5.Octadien.2.one...E.E..	0
Acetophenone	74
1.Decen.3.one	0
Pyrazine..3.ethyl.2.5.dimethyl.	0
Pyrazine..tetramethyl.	0

(continued)

Chemical	Mean relative concentration
5.Methyl.2.thiophenecarboxaldehyde	0
g.Heptalactone	0
Linalool	0
Nonanal	1436.5
Thymol	0
Phenylethyl.Alcohol	0
2.3.5.Trimethyl.6.ethylpyrazine.	0
Acetic.acid..phenylmethyl.ester	179.5
Safranal	0
2.Decenal...E..	150
g.octalacone	0
o.Amino.acetophenone	0
2.4.Decadienal	0
g.Nonlactone	0
Ionone	0
Geranyl.acetone	0
Ionene	0
g.Nonlactone.1	0
2.4.Nonadienal...E.E..	0
2.4.Decadienal.1	17.980041
g.Heptalactone.1	0
Ionone.1	0
Geranyl.acetone.1	0
a.Ionone	0
Peach.lactone.g.undecalactone	46.4516735
d.Decalactone	186.835836
cis.Geranylacetone	0
d.dodecalactone..δ.Nonyl.δ.valeralactone.	1582.590707
d.Undecalactone	11295.4731

Example 7: PCA Analysis

[0054] Multiple production lots of *Chlorella protothecoides* microalgal flour were produced according to methods given above. In addition, a commercial sample of Chlorella powder was obtained from nuts.com; the product information as of the date of filing. http://www.nuts.com/ assigns the flour to a Korean source, with heterotrophic production. A total of 12 samples, measured in duplicate by SBSE and SPME as in Examples 4 and 5, were used. In addition, sensory testing was done using a panel of volunteers. Scaled principal component analysis (using a correlation matrix) was performed with R software version 2.15.1 (The R project for Statistical Computing, www.r-project.org) using the prcomp function. Three principal components were found that well characterize the variation in flavor/odor compounds. Vectors defining the three principal components are listed in the table below as PC1, PC2, and PC3 along with the method used for determining each compound. A cluster of samples was found in this reduced-dimensional space that correlated with the samples having acceptable sensory characteristics.

Chemical Name	CAS	PC1	PC2	PC3	GC Method
Dimethyl sulfide	75-18-3	0.0076	-0.154649	0.1379564	SPME
2,3-Butanedione	431-03-8	-0.05341	0.116238	0.1384577	SPME
Butanal	123-72-8	-0.0612	0.021748	-0.1541993	SPME
Propanal, 2-methyl-	78-84-2	-0.0248	-0.203551	0.1420793	SPME
Furan, 3-methyl-	930-27-8	-0.13905	0.053489	-0.0400092	SPME
Ethyl Acetate	141-78-6	0.02303	0.078633	0.1490604	SPME

(continued)

Chemical Name	CAS	PC1	PC2	PC3	GC Method
2-Butenal, (E)-	123-73-9	0.0346	-0.007869	-0.2288552	SPME
Butanal, 3-methyl-	590-86-3	-0.01585	-0.209996	0.152554	SPME
1-Butanol	71-36-3	0.01482	0.147081	0.1203239	SPME
Butanal, 2-methyl-	96-17-3	-0.06977	-0.186611	0.1433748	SPME
Thiophene	110-02-1	-0.14535	0.003674	-0.0107213	SPME
1-Penten-3-ol	616-25-1	0.10591	0.05907	-0.0208901	SPME
1-Penten-3-one	1629-58-9	0.02932	-0.055926	-0.1865801	SPME
2-Pentanone	107-87-9	0.01895	-0.168215	-0.1843823	SPME
2,3-Pentanedione	600-14-6	0.03772	-0.074626	-0.0103901	SPME
Pentanal	110-62-3	-0.05954	-0.059048	-0.1301291	SPME
Furan, 2-ethyl-	3208-16-0	-0.00841	-0.0761	-0.0141672	SPME
Thiazole	288-47-1	-0.14288	-0.031332	0.0205445	SPME
3-Penten-2-one	625-33-2	0.03658	-0.118624	0.1932202	SPME
Disulfide, dimethyl	624-92-0	0.00766	0.07675	-0.030508	SPME
2-Pentenal, (E)-	1576-87-0	0.02904	0.005659	-0.0633539	SPME
Pyrrole	109-97-7	-0.14542	0.001009	-0.0083546	SPME
Oxazole, 4,5-dimethyl-	20662-83-3	-0.14535	0.003674	-0.0107213	SPME
2-Penten-1-ol, (Z)-	1576-95-0	-0.14181	-0.022408	-0.0072056	SPME
Thiophene, 3-methyl-	616-44-4	0.00669	0.144512	0.1163417	SPME
Hexanal	66-25-1	0.02329	0.064197	-0.1621187	SPME
4-Methylthiazole	693-95-8	-0.14535	0.003674	-0.0107213	SPME
Pyrazine, methyl-	109-08-0	-0.13884	-0.055436	0.0337262	SPME
Furfural	98-01-1	-0.14535	0.003674	-0.0107213	SPME
Oxazole, trimethyl-	20662-84-4	-0.14535	0.003674	-0.0107213	SPME
Butanoic acid, 3-methyl-	503-74-2	-0.14535	0.003674	-0.0107213	SPME
Butanoic acid, 2-methyl-	116-53-0	-0.14535	0.003674	-0.0107213	SPME
2-Hexenal	505-57-7	0.02747	-0.052249	-0.2361552	SPME
1-Hexanol	111-27-3	0.03121	0.198559	0.0119837	SPME
4-Heptanone	123-19-3	-0.00358	-0.135096	0.0100197	SPME
Pyridine, 2,6-dimethyl-	108-48-5	-0.14535	0.003674	-0.0107213	SPME
Thiazole, 2,4-dimethyl-	541-58-2	-0.14535	0.003674	-0.0107213	SPME
3-Heptanone	106-35-4	0.02161	-0.184446	-0.1716557	SPME
2-Heptanone	110-43-0	-0.09702	-0.058868	0.0154171	SPME
3-Heptanol	589-82-2	0.02303	-0.205456	-0.1113283	SPME
Heptanal	111-71-7	-0.11331	0.141566	-0.0259176	SPME
Methional	3268-49-3	-0.11001	-0.130401	0.0939776	SPME
Pyrazine, 2,5-dimethyl-	123-32-0	0.02063	-0.11695	-0.0042558	SPME
Pyrazine, 2,6-dimethyl-	108-50-9	-0.14539	-0.007146	-0.0010984	SPME
Pyrazine, ethyl-	13925-00-3	-0.14544	-4.79E-05	-0.0074156	SPME
Pyrazine, 2,3-dimethyl-	5910-89-4	-0.14541	0.001518	-0.0088075	SPME
Pyrazine, ethenyl-	4177-16-6	-0.14535	0.003674	-0.0107213	SPME
Thiazole, 4,5-dimethyl-	3581-91-7	-0.14535	0.003674	-0.0107213	SPME
2-Heptanone, 6-methyl-	928-68-7	0.14535	0.003674	-0.0107213	SPME
Hexanal, 2-ethyl-	123-05-7	0.01846	-0.027007	-0.1799374	SPME
2-Heptenal, (Z)-	57266-86-1	0.02161	-0.093801	-0.1905916	SPME
5-Nonen-2-one	27039-84-5	-0.14535	0.003674	-0.0107213	SPME
2-Furancarboxaldehyde, 5-methyl-	620-02-0	0.01921	-0.109621	0.1754483	SPME
Benzaldehyde	100-52-7	-0.14243	0.046336	0.0247769	SPME
hexanoic acid	109-52-4	-0.00113	0.064879	-0.0160903	SPME

(continued)

Chemical Name	CAS	PC1	PC2	PC3	GC Method
1-Octen-3-ol	3391-86-4	-0.09067	-0.045064	-0.1354748	SPME
Dimethyl trisulfide	3658-80-8	0.0289	-0.064852	-0.1508671	SPME
2,5-Octanedione	3214-41-3	0.02899	-0.075905	-0.0937522	SPME
5-Hepten-2-one, 6-methyl-	110-93-0	-0.14527	0.00547	-0.0141759	SPME
Furan, 2-pentyl-	3777-69-3	-0.07838	0.16758	-0.0356101	SPME
2,4-Heptadienal, (E,E)-	4313-03-5	0.024	-0.071588	-0.1450388	SPME
Pyrazine, 2-ethyl-6-methyl-	13925-03-6	-0.14535	0.003674	-0.0107213	SPME
Octanal	124-13-0	0.06342	0.197764	-0.0144755	SPME
Pyrazine, trimethyl-	14667-55-1	-0.14463	-0.018889	0.0093576	SPME
Pyrazine, 2-ethyl-3-methyl-	15707-23-0	-0.14535	0.003674	-0.0107213	SPME
2,4-Heptadienal, (E,E)-	4313-03-5	0.03375	-0.100784	-0.1998281	SPME
Pyrazine, 2-ethenyl-6-methyl-	13925-09-2	-0.14535	0.003674	-0.0107213	SPME
1-Hexanol, 2-ethyl-	104-76-7	0.01545	-0.147033	-0.1738968	SPME
3-Octen-2-one, (E)-	18402-82-9	0.02243	-0.027669	-0.1418	SPME
2H-Pyran-2-one, 5,6-dihydro-	3393-45-1	0.04024	0.008083	-0.0019753	SPME
Benzeneacetaldehyde	122-78-1	0.01141	-0.200551	0.1476711	SPME
3,5-Octadien-2-one, (E,E)-	30086-02-3	0.02431	0.191552	-0.0405352	SPME
Acetophenone	98-86-2	0.03482	0.112029	0.0678319	SPME
1-Decen-3-one	56606-79-2	0.01487	-0.007144	0.0679731	SPME
Pyrazine, 3-ethyl-2,5-dimethyl-	13360-65-1	-0.14539	0.002524	-0.0097007	SPME
Pyrazine, tetramethyl-	1124-11-4	-0.14544	-0.003912	-0.0054264	SPME
5-Methyl-2-thiophenecarboxaldehyde	13679-70-4	-0.14535	0.003674	-0.0107213	SPME
g-Heptalactone	105-21-5	0.01298	0.140814	0.1183756	SPME
Linalool	78-70-6	-0.14535	0.003674	-0.0107213	SPME
Nonanal	124-19-6	0.05356	0.198786	-0.1092893	SPME
Thymol	89-83-8	-0.14535	0.003674	-0.0107213	SPME
Phenylethyl Alcohol	60-12-8	-0.14506	-0.014282	0.003239	SPME
2,3,5-Trimethyl-6-ethylpyrazine	17398-16-2	-0.14538	0.002837	-0.0099785	SPME
Acetic acid, phenylmethyl ester	140-11-4	0.04544	0.114759	0.1539536	SPME
Safranal	116-26-7	-0.14535	0.003674	-0.0107213	SPME
2-Decenal, (E)-	3913-81-3	0.03435	-0.01297	-0.2149363	SPME
g-Octalactone	104-50-7	0.01639	0.142953	0.0964521	SPME
o-Amino acetophenone	551-93-9	0.02232	0.204042	0.0183701	SPME
2,4-Decadienal	2363-88-4	0.01791	0.169004	-0.0389474	SBSE
g-Nonlactone	104-61-0	0.01493	0.18923	0.0333768	SPME
a-Ionone	127-41-3	-0.14535	0.003674	-0.0107213	SPME
Geranyl acetone	3796-70-1	-0.14542	-0.002004	-0.0085515	SPME
a-Ionene	14901-07-6	-0.14535	0.003674	-0.0107213	SBSE
g-Nonalactone	104-61-0	0.01637	-0.075372	-0.0496326	SBSE
2,4-Nonadienal	6750-03-4	0.03136	-0.023742	-0.1745061	SBSE
2,4-Decadienal	2363-88-4	0.02952	0.094377	-0.1710607	SBSE
g-Heptalactone	105-21-5	0.01775	0.158721	-0.0198467	SBSE
a-Ionone	127-41-3	-0.14535	0.003674	-0.0107213	SBSE
Geranyl acetone	3796-70-1	-0.14535	0.003674	-0.0107213	SBSE
a-Ionone	127-41-3	-0.14535	0.003674	-0.0107213	SBSE
g-Undecalactone	104-67-6	0.09703	-0.071462	0.0844344	SBSE
d-Decalactone	705-86-2	0.03467	-0.188054	0.0770618	SBSE
cis-Geranylacetone	3879-26-3	0.01193	0.016184	-0.0633938	SBSE
d-Dodecalactone..	713-95-1	0.13073	-0.059213	0.0333184	SBSE

(continued)

Chemical Name	CAS	PC1	PC2	PC3	GC Method
d-Undecalactone	710-04-3	0.05183	-0.042457	-0.1311766	SBSE

[0055] Fig. 2 shows the PCA analysis clustering. Each plotted point represents a microalgal powder sample plotted in a space defined by the principal components PC1, PC2, and PC3 (dim1, dim2 and dim3 respectively). The solid circles represent *Chlorella protothecoides* flour samples that has acceptable flavor. The open circles represent *Chlorella protothecoides* flour samples with inferior flavor. The open square represent the *Chlorella vulgaris* obtained from Nuts.com.

Example 8: Determination of bounds for acceptable flavor

[0056] Based on the PCA analysis of Example 7, the FactomineR package v. 1.2.1 (Husson, et al.) was used to statistically define the cluster of samples that correlated with the acceptable sensory testing. The result of the FactomineR analysis was 3 ellipsoids in the three dimensions of PC1, PC2 and PC3; the ellipsoids characterize 1, 2, and 3 standard deviations from center point of the cluster associated with the positive human sensory analysis (solid circles from the graph of Example 7). Each 3-dimensional ellipsoid is defined by 3 orthogonal 2-dimensional ellipses defined by the equation $Ax^2+Bxy+Cy^2+Dx+Ey+F = 0$ using the data in the table below for the values of A, B, C, D, E, and F. Thus, samples falling within the smallest ellipsoid will be expected to have a positive sensory analysis by a human panel about 99.7% of the time, samples falling within only the mid-sized ellipsoid will be expected to have a positive sensory analysis by a human panel about 95% of the time and samples falling only within the largest ellipsoid will be expected to have a positive sensory analysis by a human panel about 68% of the time.

Equation for confidence intervals: Equation: $Ax^2+Bxy+Cy^2+Dx+Ey+F = 0$

[0057]

Standard Deviations	X Dimension	Y Dimension	A	B	C	D	E	F
3	PC1	PC2	0.00348 1467	0.00036 6174	3.79437 E-05	0.00062 8924	4.27301 E-05	1.51548 E-05
3	PC1	PC3	0.00173 4328	0.00028 6969	1.89401 E-05	0.00031 8201	2.8099E -05	1.12003 E-05
3	PC2	PC3	0.35621 8856	0.28921 9807	0.35693 6631	0.08519 1149	0.04023 7159	0.13812 915
2	PC1	PC2	0.00047 7458	5.02181 E-05	5.2037E -06	8.62524 E-05	5.86012 E-06	3.01302 E-06
2	PC1	PC3	0.00023 785	3.93556 E-05	2.5975E -06	4.3639E -05	3.85357 E-06	1.76892 E-06
2	PC2	PC3	0.04885 2827	0.03966 4394	0.04895 1264	0.01168 3347	0.00551 8234	0.00911 8978
1	PC 1	PC2	2.78319 E-05	2.9273E -06	3.03333 E-07	5.0278E -06	3.41597 E-07	2.11154 E-07
1	PC 1	PC 3	1.38647 E-05	2.29411 E-06	1.51413 E-07	2.54379 E-06	2.24631 E-07	1.11963 E-07
1	PC2	PC 3	0.00066 5829	0.00046 6136	0.00015 2694	0.00038 0618	0.00013 6456	4.14371 E-05

Example 9: QC Analysis using results of PCA analysis

[0058] The ellipsoids of Example 8 can be used to determine if a sample falls within the cluster associated with positive flavor. For example, a quality-control experiment can be performed on a batch of microalgal flour produced according to the methods given above. The flour is analyzed by SPME and SBSE as in Examples 4 and 5 and then one determines if the data falls within one or more of the ellipsoids of Example 8.

[0059] To do this, one can use the following procedure (though others may be applicable). Start with relative concentration for 105 compounds. From each concentration, subtract its center factor and divide by its scale factor (given in

the table below). This centers and scales the data. Take the dot product of the scaled and centered data and the principal component (PC) loadings. This will yield one value for each PC. Divide each value by its associated plotting factor. This will allow the data point to be plotted in three dimensional algal-chemical space. If the point falls within the space bounded by the confidence ellipsoid, it is not statistically different ($p < 0.05$). For example, if the point falls within the space bounded by the 95% confidence ellipsoid, it is not statistically different ($p < 0.05$).

Chemical	Center	Scale	PC1	PC2	PC3
Dimethyl.sulfide	15.04166667	52.10586179	0.007602386	0.154648539	0.13795639
2.3.Butanedione	573.4583333	687.3035077	0.053406645	0.116238372	0.138457708
Butanal	165.0833333	291.8766733	0.061200873	0.021748265	0.154199309
Propanal..2.methyl.	294.25	321.9922006	-0.02479716	0.203551061	0.142079295
Furan..3.methyl.	254.0833333	364.0905752	0.139050167	0.053488926	0.040009249
Ethyl.Acetate	1534.958333	721.2414001	0.023033335	0.078632968	0.149060426
2.Butenal...E..	56.95833333	67.74264748	0.034598984	0.007869304	0.228855217
Butanal..3.methyl.	2368.958333	3305.894731	0.015854973	0.209996041	0.152553963
1.Butanol	236.75	723.0508438	0.01482126	0.147080874	0.120323863
Butanal..2.methyl.	858.0416667	1132.843254	0.069765232	0.186610612	0.143374765
Thiophene	0.708333333	2.453738644	0.145349572	0.003673658	0.010721336
1.Penten.3.ol	111.2916667	123.2715883	0.105910877	0.059069801	0.020890092
1.Penten.3.one	10.625	18.86570361	0.029319785	0.055925743	0.186580083
2.Pentanone	429.875	520.4705967	0.018948769	0.168215403	0.184382338
2.3.Pentanedione	392.625	359.8726495	0.037715762	0.074625863	0.010390137
Pentanal	5315.166667	4258.727501	-0.05954475	-0.05904769	0.130129097
Furan..2.ethyl.	32.75	24.43590875	0.008414663	0.076099651	0.014167153
Thiazole	70.16666667	199.0549642	0.142882049	0.031332244	0.020544457
3.Penten.2.one	442.125	470.5612763	0.036579138	0.118623927	0.193220234
Disulfide..dimethyl	77.45833333	105.2821875	0.007660621	0.076749927	0.030508003
2.Pentenal...E..	116.7083333	200.60312	0.029036734	0.005658787	0.063353931
Pyrrole	12.29166667	41.79846579	0.145424967	0.001008736	0.008354639
Oxazole..4.5.dimethyl.	15.83333333	54.84827557	0.145349572	0.003673658	0.010721336
2.Penten.1.ol...Z..	45.25	118.0232065	0.141807908	0.022407562	0.007205637
Thiophene..3.methyl.	108.5416667	279.7959856	0.006693629	0.144512146	0.116341706
Hexanal	26189.95833	17886.61913	0.023290612	0.064196972	0.162118696
4.Methylthiazole	1.958333333	6.783865663	0.145349572	0.003673658	0.010721336
Pyrazine..methyl.	135.2083333	326.6405766	0.138842567	0.055435505	0.03372617
Furfural	34.5	119.5115057	0.145349572	0.003673658	0.010721336
Oxazole..trimethyl.	64	221.7025034	0.145349572	0.003673658	0.010721336
Butanoic.acid..3.methyl.	58.58333333	202.9386196	0.145349572	0.003673658	0.010721336
Butanoic.acid..2.methyl.	3.833333333	13.27905619	0.145349572	0.003673658	0.010721336
2.Hexenal	25.58333333	50.09710268	0.027469429	0.052249399	-0.23615517
1.Hexanol	106.1666667	155.9474465	0.031207096	0.198558566	0.011983686

(continued)

Chemical	Center	Scale	PC1	PC2	PC3
4.Heptanone	360.5833333	577.8576749	0.003575779	0.135096305	0.010019679
Pyridine..2.6.dimethyl.	2.958333333	10.24796728	0.145349572	0.003673658	0.010721336
Thiazole..2.4.dimethyl.	15.58333333	53.98225017	0.145349572	0.003673658	0.010721336
3.Heptanone	111.625	94.41016052	0.021607662	-0.18444557	0.171655667
2.Heptanone	380.875	288.460973	0.097016748	0.058868123	0.015417076
3.Heptanol	1193.041667	1008.348074	0.023029974	0.205456135	0.111328282
Heptanal	1396.791667	920.0702903	0.113307135	0.141565621	0.025917554
Methional	79.625	148.3023823	0.110012922	0.130400953	0.093977633
Pyrazine..2.5.dimethyl.	3.333333333	7.857634774	0.020631611	0.116950274	0.004255769
Pyrazine..2.6.dimethyl.	178.2083333	574.8013672	0.145388496	0.007146465	0.001098366
Pyrazine..ethyl.	15.95833333	53.8796885	0.145442956	-0.0000479	0.007415618
Pyrazine..2.3.dimethyl.	439.2083333	1498.775644	0.145413873	0.001518449	0.008807482
Pyrazine..ethenyl.	1.416666667	4.907477288	0.145349572	0.003673658	0.010721336
Thiazole..4.5.dimethyl.	3.583333333	12.41303079	0.145349572	0.003673658	0.010721336
2.Heptanone..6.methyl.	53.75	186.1954618	0.145349572	0.003673658	0.010721336
Hexanal..2.ethyl.	78.41666667	124.9672381	0.018460956	0.027007294	0.179937424
2.Heptenal...Z..	645.25	937.3877266	0.021607084	0.093800543	0.190591625
5.Nonen.2.one	13.33333333	46.18802154	0.145349572	0.003673658	0.010721336
2.Furancarboxaldehyde..5.methyl.	21.25	40.57288615	0.019206035	0.109620677	0.175448337
Benzaldehyde	872.875	1358.161493	0.142431906	0.046335544	0.024776943
hexanoic.acid	176.25	216.4210438	0.001128927	0.064879481	0.016090326
1.Octen.3.ol	369.6666667	350.9919277	0.090672545	0.045064295	0.135474824
Dimethyl.trisulfide	14.33333333	21.56315601	0.028899179	0.064852089	0.150867075
2.5.Octanedione	23.95833333	44.27674248	0.028988465	-0.07590479	0.093752193
5.Hepten.2.one..6.methyl.	1503.833333	4827.634134	0.145266246	0.005470194	0.014175912
Furan..2.pentyl.	633	967.4016276	0.078384616	0.167579691	0.035610073
2.4.Heptadienal...E.E..	20.83333333	43.16371231	0.024003523	0.071588186	0.145038829
Pyrazine..2.ethyl.6.methyl.	21	72.74613392	0.145349572	0.003673658	0.010721336
Octanal	1243.041667	897.5365644	0.063418428	0.197764097	-0.01447548
Pyrazine..trimethyl.	348.6666667	1051.439497	0.144625394	0.018888681	0.009357594
Pyrazine..2.ethyl.3.methyl.	87.33333333	302.5315411	0.145349572	0.003673658	0.010721336
2.4.Heptadienal...E.E...1	26.33333333	40.42070427	0.033749609	0.100784032	0.199828071
Pyrazine..2.ethenyl.6.methyl.	5.541666667	19.19689645	0.145349572	0.003673658	0.010721336

(continued)

Chemical	Scale	Center	PC1	PC2	PC3
1.Hexanol..2.ethyl.	5078.453328	5684.541667	0.015454406	0.147033095	0.173896762
3.Octen.2.one...E..	462.4334412	196.375	0.022433793	0.027668713	0.141800019
X2H.Pyran.2.one..5.6.dihydro.	845.025291	683.3333333	0.040235145	0.008083104	0.001975331
Benzeneacetaldehyde	60.74811383	31.83333333	0.01141478	0.200551415	0.147671091
3.5.Octadien.2.one...E.E..	426.6112306	455.125	0.024307307	0.191552198	0.040535191
Acetophenone	56.41088104	42.375	0.034819826	0.112028714	0.067831917
1.Decen.3.one	9.100761706	3.125	0.014871492	0.007143686	0.067973089
Pyrazine..3.ethyl.2.5.dimethyl.	174.3908228	50.75	0.145387371	0.002524067	0.009700663
Pyrazine..tetramethyl.	3113.918129	951.4583333	0.145437121	-0.00391206	0.005426362
5.Methyl.2.thiophenecarboxaldehyde	198.7528302	57.375	0.145349572	0.003673658	0.010721336
g.Heptalactone	6.92820323	2	0.012980337	0.140814237	0.118375646
Linalool	34.06366588	9.833333333	0.145349572	0.003673658	0.010721336
Nonanal	1335.036088	1528.416667	0.053558189	0.198785653	0.109289305
Thymol	556.2769844	160.5833333	0.145349572	0.003673658	0.010721336
Phenylethyl.Alcohol	416.085189	135.9583333	0.145061726	-0.01428243	0.003239013
2.3.5.Trimethyl.6.ethylpyrazine.	718.7459552	208.7083333	0.145377878	0.002836895	-0.00997845
Acetic.acid..phenylmethyl.ester	205.6043337	213.875	0.045438482	0.114758954	0.153953593
Safranal	163.8231389	47.29166667	0.145349572	0.003673658	0.010721336
2.Decenal...E..	78.60616976	55.04166667	0.034351801	0.012969523	-0.21493625
g.octalacone	28.57933535	10.625	0.016392036	0.14295305	0.096452129
o.Amino.acetophenone	32.17070943	15.5	0.022315438	0.204041622	0.018370134
2.4.Decadienal	24.16781606	9.416666667	0.0179089	0.169004115	0.038947428
g.Nonlactone	40.20345982	13.5	0.01493418	0.189230257	0.033376822
Ionone	351.0289637	101.3333333	0.145349572	0.003673658	0.010721336
Geranyl.acetone	2137.396627	652.75	0.145423518	0.002004031	0.008551463
Ionene	553.5345706	159.7916667	0.145349572	0.003673658	0.010721336
g.Nonlactone.1	22.81994259	6.58755	0.016371012	0.075372449	0.049632645
2.4.Nonadienal...E.E..	30.64101284	18.07305674	0.031363408	0.023742328	0.174506137
2.4.Decadienal.1	85.11825112	50.4716275	0.029518821	0.094376773	0.171060695
g.Heptalactone.1	42.07909242	17.25928968	0.017750131	0.158720982	0.019846703
Ionone.1	689.4126429	199.0162875	0.145349572	0.003673658	0.010721336
Geranyl.acetone.1	3049.421811	880.2922516	0.145349572	0.003673658	0.010721336
a.Ionone	1160.638915	335.0475951	0.145349572	0.003673658	0.010721336

(continued)

Chemical	Center	Scale	PC1	PC2	PC3
Peach.lactone.g.undecalactone	72.77877498	34.06000193	0.097029409	0.071461906	0.084434422
d.Decalactone	85.57314465	106.5309321	0.034674859	-0.18805394	0.077061807
cis.Geranylacetone	5.9584	20.64050306	0.011926134	0.016184168	0.063393798
d.dodecalactone..δ.Nonyl.δ.valeralactone.	. 1400.955104	491.4817796	0.130734715	0.059212775	0.033318423
d.Undecalactone	6472.792302	6394.323609	0.051826724	0.042456918	0.131176612

Plotting Factor:

PC Standard Deviation *

Square Root of number of samples from the model

PC1	PC2	PC3
23.79781	12.25408	11.48665

Claims

1. A microalgal flour suitable for use in food, the flour comprising microalgal cells of *Chlorophyta* and **characterized by** a flavor descriptor falling within an ellipsoid in a flavor-description space having dimensions of PC1, PC2 and PC3, the flavor descriptor produced by using SPME and/or SBSE analysis to determine concentrations of the following compounds:

Dimethyl.sulfide
2.3.Butanedione
Butanal
Propanal..2.methyl.
Furan..3.methyl.
Ethyl.Acetate
2.Butenal...E..
Butanal..3.methyl.
1.Butanol
Butanal..2.methyl.
Thiophene
1.Penten.3.ol
1.Penten.3.one
2.Pentanone
2.3.Pentanedione
Pentanal
Furan..2.ethyl.
Thiazole
3.Penten.2.one
Disulfide..dimethyl
2.Pentenal...E..
Pyrrole
Oxazole..4.5.dimethyl.
2.Penten.1.ol...Z..
Thiophene..3.methyl.
Hexanal
4.Methylthiazole
Pyrazine..methyl.
Furfural
Oxazole..trimethyl.
Butanoic.acid..3.methyl.
Butanoic.acid..2.methyl.
2.Hexenal
1.Hexanol
4.Heptanone
Pyridine..2.6.dimethyl.
Thiazole..2.4.dimethyl.
3.Heptanone
2.Heptanone
3.Heptanol
Heptanal
Methional
Pyrazine..2.5.dimethyl.
Pyrazine..2.6.dimethyl.
Pyrazine..ethyl.
Pyrazine..2.3.dimethyl.
Pyrazine..ethenyl.
Thiazole..4.5.dimethyl.
2.Heptanone..6.methyl.
Hexanal..2.ethyl.
2.Heptenal...Z..

5.Nonen.2.one
2.Furancarboxaldehyde..5.methyl
Benzaldehyde
hexanoic.acid
1.Octen.3.ol
Dimethyl.trisulfide
2.5.Octanedione
5.Hepten.2.one..6.methyl.
Furan..2.pentyl.
2.4.Heptadienal...E.E..
Pyrazine..2.ethyl.6.methyl.
Octanal
Pyrazine..trimethyl.
Pyrazine..2.ethyl.3.methyl.
2.4.Heptadienal...E.E...1
Pyrazine..2.ethenyl.6.methyl.
1.Hexanol..2.ethyl.
3.Octen.2.one...E..
2H.Pyran.2.one..5.6.dihydro.
Benzeneacetaldehyde
3.5.Octadien.2.one...E.E..
Acetophenone
1.Decen.3.one
Pyrazine..3.ethyl.2.5.dimethyl.
Pyrazine..tetramethyl.
5.Methyl.2.thiophenecarboxaldehyde
g.Heptalactone
Linalool
Nonanal
Thymol
Phenylethyl.Alcohol
2.3.5.Trimethyl.6.ethylpyrazine.
Acetic.acid..phenylmethyl.ester
Safranal
2.Decenal...E..
g.octalacone
o.Amino.acetophenone
2.4.Decadienal
g.Nonlactone
Ionone
Geranyl.acetone
Ionene
g.Nonlactone.1
2.4.Nonadienal...E.E..
2.4.Decadienal.1
g.Heptalactone.1
Ionone.1
Geranyl.acetone.1
a.Ionone
Peach.lactone.g.undecalactone
d.Decalactone
cis.Geranylacetone
d.dodecalactone..δ.Nonyl.δ.valeralactone.
d.Undecalactone

relative to an internal standard,
the ellipsoid being defined by the equation $Ax^2+Bxy+Cy^2+Dx+Ey+F = 0$ and parameterized according to the following table:

X Dimension	Y Dimension	A	B	C	D	E	F
PC1	PC2	0.003481467	-0.000366174	3.79437E-05	-0.000628924	4.27301E-05	1.51548E-05
PC1	PC3	0.001734328	0.000286969	1.89401E-05	-0.000318201	-2.8099E-05	1.12003E-05
PC2	PC3	0.356218856	0.289219807	0.356936631	0.085191149	-0.040237159	-0.13812915

wherein, the falling within the ellipsoid is determined by the procedure of:

for each compound, determining relative concentrations;
for each compound, subtracting center factors according to the table below;
for each compound, dividing by the scale factors according to the table below;
taking the dot product of the scaled and centered data to yield values for PC1, PC2 and PC3; and
determining if the flavor descriptor defined by PC1, PC2 and PC3 falls within the ellipsoid:

Chemical	**Center**	**Scale**	PC1	PC2	PC3
Dimethyl.sulfide	15.04166667	52.10586179	0.007602386	-0.154648539	0.13795639
2.3.Butanedione	573.4583333	687.3035077	-0.053406645	0.116238372	0.138457708
Butanal	165.0833333	291.8766733	-0.061200873	0.021748265	-0.154199309
Propanal..2.methyl.	294.25	321.9922006	-0.02479716	-0.203551061	0.142079295
Furan..3.methyl.	254.0833333	364.0905752	-0.139050167	0.053488926	-0.040009249
Ethyl.Acetate	1534.958333	721.2414001	0.023033335	0.078632968	0.149060426
2.Butenal...E..	56.95833333	67.74264748	0.034598984	-0.007869304	-0.228855217
Butanal..3.methyl.	2368.958333	3305.894731	-0.015854973	-0.209996041	0.152553963
1.Butanol	236.75	723.0508438	0.01482126	0.147080874	0.120323863
Butanal..2.methyl.	858.0416667	1132.843254	-0.069765232	-0.186610612	0.143374765
Thiophene	0.708333333	2.453738644	-0.145349572	0.003673658	-0.010721336
1.Penten.3.ol	111.2916667	123.2715883	-0.105910877	0.059069801	-0.020890092
1.Penten.3.one	10.625	18.86570361	0.029319785	-0.055925743	-0.186580083
2.Pentanone	429.875	520.4705967	0.018948769	-0.168215403	0.184382338
2.3.Pentanedione	392.625	359.8726495	0.037715762	-0.074625863	-0.010390137
Pentanal	5315.166667	4258.727501	-0.05954475	-0.05904769	-0.130129097
Furan..2.ethyl.	32.75	24.43590875	-0.008414663	-0.076099651	-0.014167153
Thiazole	70.16666667	199.0549642	-0.142882049	-0.031332244	0.020544457
3.Penten.2.one	442.125	470.5612763	0.036579138	-0.118623927	0.193220234
Disulfide..dimethyl	77.45833333	105.2821875	0.007660621	0.076749927	-0.030508003
2.Pentenal...E..	116.7083333	200.60312	0.029036734	0.005658787	-0.063353931
Pyrrole	12.29166667	41.79846579	-0.145424967	0.001008736	-0.008354639
Oxazole..4.5.di methyl.	15.83333333	54.84827557	-0.145349572	0.003673658	-0.010721336
2.Penten.1.ol...Z..	45.25	118.0232065	-0.141807908	-0.022407562	-0.007205637
Thiophene..3.methyl.	108.5416667	279.7959856	0.006693629	0.144512146	0.116341706
Hexanal	26189.95833	17886.61913	0.023290612	0.064196972	-0.162118696
4.Methylthiazole	1.958333333	6.783865663	-0.145349572	0.003673658	-0.010721336
Pyrazine..methyl.	135.2083333	326.6405766	-0.138842567	-0.055435505	0.03372617
Furfural	34.5	119.5115057	-0.145349572	0.003673658	-0.010721336
Oxazole..trimethyl.	64	221.7025034	-0.145349572	0.003673658	-0.010721336
Butanoic.acid..3.methyl.	58.58333333	202.9386196	-0.145349572	0.003673658	-0.010721336
Butanoic.acid..2.methyl.	3.833333333	13.27905619	-0.145349572	0.003673658	-0.010721336
2.Hexenal	25.58333333	50.09710268	0.027469429	-0.052249399	-0.23615517
1.Hexanol	106.1666667	155.9474465	0.031207096	0.198558566	0.011983686
4.Heptanone	360.5833333	577.8576749	-0.003575779	-0.135096305	0.010019679
Pyridine..2.6.dimethyl.	2.958333333	10.24796728	-0.145349572	0.003673658	-0.010721336
Thiazole..2.4.dimethyl.	15.58333333	53.98225017	-0.145349572	0.003673658	-0.010721336
3.Heptanone	111.625	94.41016052	0.021607662	-0.18444557	-0.171655667
2.Heptanone	380.875	288.460973	-0.097016748	-0.058868123	0.015417076
3.Heptanol	1193.041667	1008.348074	0.023029974	-0.205456135	-0.111328282
Heptanal	1396.791667	920.0702903	-0.113307135	0.141565621	-0.025917554
Methional	79.625	148.3023823	-0.110012922	-0.130400953	0.093977633

(continued)

Chemical	Center	Scale	PC1	PC2	PC3
Pyrazine..2.5.dimethyl.	3.333333333	7.857634774	0.020631611	-0.116950274	-0.004255769
Pyrazine..2.6.dimethyl.	178.2083333	574.8013672	-0.145388496	-0.007146465	-0.001098366
Pyrazine..ethyl.	15.95833333	53.8796885	-0.145442956	-0.0000479	-0.007415618
Pyrazine..2.3.dimethyl.	439.2083333	1498.775644	-0.145413873	0.001518449	-0.008807482
Pyrazine..ethenyl.	1.416666667	4.907477288	-0.145349572	0.003673658	-0.010721336
Thiazole..4.5.dimethyl.	3.583333333	12.41303079	-0.145349572	0.003673658	-0.010721336
2.Heptanone..6.methyl.	53.75	186.1954618	-0.145349572	0.003673658	-0.010721336
Hexanal..2.ethyl.	78.41666667	124.9672381	0.018460956	-0.027007294	-0.179937424
2.Heptenal...Z..	645.25	937.3877266	0.021607084	-0.093800543	-0.190591625
5.Nonen.2.one	13.33333333	46.18802154	-0.145349572	0.003673658	-0.010721336
2.Furancarboxal dehyde.. 5.methyl	21.25	40.57288615	0.019206035	-0.109620677	0.175448337
Benzaldehyde	872.875	1358.161493	-0.142431906	0.046335544	0.024776943
hexanoic.acid	176.25	216.4210438	-0.001128927	0.064879481	-0.016090326
1.Octen.3.ol	369.6666667	350.9919277	-0.090672545	-0.045064295	-0.135474824
Dimethyl.trisulfide	14.33333333	21.56315601	0.028899179	-0.064852089	-0.150867075
2.5.Octanedione	23.95833333	44.27674248	0.028988465	-0.07590479	-0.093752193
5.Hepten.2.one.. 6.methyl.	1503.833333	4827.634134	-0.145266246	0.005470194	-0.014175912
Furan..2.pentyl.	633	967.4016276	-0.078384616	0.167579691	-0.035610073
2.4.Heptadienal...E.E..	20.83333333	43.16371231	0.024003523	-0.071588186	-0.145038829
Pyrazine..2.ethyl.6.methyl.	21	72.74613392	-0.145349572	0.003673658	-0.010721336
Octanal	1243.041667	897.5365644	0.063418428	0.197764097	-0.01447548
Pyrazine..trimethyl.	348.6666667	1051.439497	-0.144625394	-0.018888681	0.009357594
Pyrazine..2.ethyl.3.methyl.	87.33333333	302.5315411	-0.145349572	0.003673658	-0.010721336
2.4.Heptadienal...E.E...1	26.33333333	40.42070427	0.033749609	-0.100784032	-0.199828071
Pyrazine..2.ethenyl.6.methyl.	5.541666667	19.19689645	-0.145349572	0.003673658	-0.010721336
1.Hexanol..2.ethyl.	5684.541667	5078.453328	0.015454406	-0.147033095	-0.173896762
3.Octen.2.one...E..	196.375	462.4334412	0.022433793	-0.027668713	-0.141800019
X2H.Pyran.2.one.. 5.6.dihydro.	683.3333333	845.025291	0.040235145	0.008083104	-0.001975331
Benzeneacetaldehyde	31.83333333	60.74811383	0.01141478	-0.200551415	0.147671091
3.5.Octadien.2.one...E.E..	455.125	426.6112306	0.024307307	0.191552198	-0.040535191
Acetophenone	42.375	56.41088104	0.034819826	0.112028714	0.067831917
1.Decen.3.one	3.125	9.100761706	0.014871492	-0.007143686	0.067973089
Pyrazine..3.ethyl. 2.5.dimethyl.	50.75	174.3908228	-0.145387371	0.002524067	-0.009700663
Pyrazine..tetramethyl.	951.4583333	3113.918129	-0.145437121	-0.00391206	-0.005426362
5.Methyl.2.thiop henecarboxaldehyde	57.375	198.7528302	-0.145349572	0.003673658	-0.010721336
g.Heptalactone	2	6.92820323	0.012980337	0.140814237	0.118375646
Linalool	9.833333333	34.06366588	-0.145349572	0.003673658	-0.010721336
Nonanal	1528.416667	1335.036088	0.053558189	0.198785653	-0.109289305
Thymol	160.5833333	556.2769844	-0.145349572	0.003673658	-0.010721336
Phenylethyl.Alcohol	135.9583333	416.085189	-0.145061726	-0.01428243	0.003239013
2.3.5.Trimethyl. 6.ethylpyrazine.	208.7083333	718.7459552	-0.145377878	0.002836895	-0.00997845
Acetic.acid..phe nylmethyl.ester	213.875	205.6043337	0.045438482	0.114758954	0.153953593
Safranal	47.29166667	163.8231389	-0.145349572	0.003673658	-0.010721336
2.Decenal...E..	55.04166667	78.60616976	0.034351801	-0.012969523	-0.21493625

(continued)

Chemical	Center	Scale	PC1	PC2	PC3
g.octalacone	10.625	28.57933535	0.016392036	0.14295305	0.096452129
o.Amino.acetophenone	15.5	32.17070943	0.022315438	0.204041622	0.018370134
2.4.Decadienal	9.416666667	24.16781606	0.0179089	0.169004115	-0.038947428
g.Nonlactone	13.5	40.20345982	0.01493418	0.189230257	0.033376822
Ionone	101.3333333	351.0289637	-0.145349572	0.003673658	-0.010721336
Geranyl.acetone	652.75	2137.396627	-0.145423518	-0.002004031	-0.008551463
Ionene	159.7916667	553.5345706	-0.145349572	0.003673658	-0.010721336
g.Nonlactone.1	6.58755	22.81994259	0.016371012	-0.075372449	-0.049632645
2.4.Nonadienal...E.E..	18.07305674	30.64101284	0.031363408	-0.023742328	-0.174506137
2.4.Decadienal.1	50.4716275	85.11825112	0.029518821	0.094376773	-0.171060695
g.Heptalactone.1	17.25928968	42.07909242	0.017750131	0.158720982	-0.019846703
Ionone.1	199.0162875	689.4126429	-0.145349572	0.003673658	-0.010721336
Geranyl.acetone.1	880.2922516	3049.421811	-0.145349572	0.003673658	-0.010721336
a.Ionone	335.0475951	1160.638915	-0.145349572	0.003673658	-0.010721336
Peach.lactone.g. undecalactone	72.77877498	34.06000193	0.097029409	-0.071461906	0.084434422
d.Decalactone	85.57314465	106.5309321	0.034674859	-0.18805394	0.077061807
cis.Geranylacetone	5.9584	20.64050306	0.011926134	0.016184168	-0.063393798
d.dodecalactone..δ.Nonyl.δ. valer alactone.	1400.955104	491.4817796	0.130734715	-0.059212775	0.033318423
d.Undecalactone	6472.792302	6394.323609	0.051826724	-0.042456918	-0.131176612.

2. The microalgal flour of claim 1, wherein the flavor descriptor falls within a narrower ellipse parameterized by the table below:

PC1	PC2	0.000477458	-5.02181E-05	5.2037E-06	-8.62524E-05	5.86012E-06	3.01302E-06
PC1	PC3	0.00023785	3.93556E-05	2.5975E-06	-4.3639E-05	-3.85357E-06	1.76892E-06
PC2	PC3	0.048852827	0.039664394	0.048951264	0.011683347	-0.005518234	-0.009118978

3. The microalgal flour of claim 1 or claim 2, wherein the flavor descriptor falls within a yet narrower ellipse parameterized by the table below:

PC 1	PC 2	2.78319E-05	-2.9273E-06	3.03333E-07	-5.0278E-06	3.41597E-07	2.11154E-07
PC 1	PC 3	1.38647E-05	2.29411E-06	1.51413E-07	-2.54379E-06	-2.24631E-07	1.11963E-07
PC 2	PC 3	-0.000665829	0.000466136	- 0.000152694	0.000380618	-0.000136456	-4.14371E-05

4. A microalgal flour of any of claims 1 to 3, obtainable by the process of:

cultivating a broth of cells of *Chlorella protothecoides* in the dark in the presence of glucose as a fixed carbon source with a starting pH of 6.8, while maintaining the dissolved oxygen level above 30%, subjecting the broth to a high-temperature-short-time process of 75°C for 1 minute, harvesting the cells by centrifugation with a dilution of 6.4 fold in water, adding an antioxidant, lysis of the cells by milling, and spray drying.

5. A microalgal flour of any of claims 1 to 4, comprising undecalactone (400-1800ppb), 3-methyl butanal (0-11,000ppb), pentanal (160-10,700ppb), 2-methyl butanal (0-2500ppb), 2-pentanone (39-10,600ppb), and/ or 3-pentene-2-one (0-1500ppb) as determined by SPME or SBSE.

6. A microalgal flour of any of claims 1 to 5, having an undetectable fish or cabbage flavor when the flour is dispersed in deionized water at 10% (w/v), as detected by a tasting panel.

7. A microalgal flour of any of claims 1 to 6, having a flowability **characterized by** an oversize of 15-35% by weight at 2000 μm.

8. A microalgal flour of any of claims 1 to 7, wherein the flour is white, pale yellow or yellow in color.

9. A microalgal flour of any of claims 1 to 8, wherein the flour comprises

(i) 5-20% lipid, or
(ii) 30-70% lipid, or
(iii) 40-60% lipid.

10. A microalgal flour of any of claims 1 to 9, wherein the pH of the flour when dissolved in water at 1% (w/v) is between:

(i) 5.5 and 8.5, or
(ii) 6.0 and 8.0, or
(iii) 6.5 and 7.5.

11. A microalgal flour of any of claims 1 to 10, having less than 200 ppm of chlorophyll, optionally less than 2 ppm chlorophyll.

12. A microalgal flour of any of claims 1 to 11, further comprising an added antioxidant.

13. A microalgal flour of any of claims 1 to 12, wherein the majority of the cells in the flour are lysed and optionally between 50 and 90% of the cells are lysed.

14. A method of preparing a microalgal flour, the method comprising the steps of:
cultivating a broth of cells of *Chlorella protothecoides* in the dark in the presence of glucose as a fixed carbon source with a starting pH of 6.8, while maintaining the dissolved oxygen level above 30%, subjecting the broth to a high-temperature-short-time process of 75°C for 1 minute, harvesting the cells by centrifugation with a dilution of 6.4 fold in water, adding an antioxidant, lysis of the cells by milling, and spray drying.

Patentansprüche

1. Mikroalgenmehl, das sich zur Verwendung in Lebensmitteln eignet, wobei das Mehl Mikroalgenzellen von *Chlorophyta* umfasst und durch ein Aromamerkmal gekennzeichnet ist, das in einem Ellipsoiden liegt, welcher sich innerhalb eines Aromabeschreibungsraums mit den Abmessungen PC1, PC2 und PC3 befindet, wobei das Aromamerkmal unter Verwendung einer SPME- und/oder SBSE-Untersuchung erhalten wurde, um die Konzentrationen der folgenden Verbindungen zu bestimmen:

Dimethylsulfid
2,3-Butandion
Butanal
2-Methylpropanal
3-Methylfuran
Ethylacetat
2-(E)-Butenal
3-Methylbutanal
1-Butanol
2-MethylBbutanal
Thiophen
1-Penten-3-ol
1-Penten-3-on
2-Pentanon
2,3-Pentandion

Pentanal
2-Ethylfuran
Thiazol
3-Penten-2-on
Dimethyldisulfid
2-(E)-Pentenal
Pyrrol
4,5-Dimethyloxazol
2-(Z)-Penten-1-ol
3-Methylthiophen
Hexanal
4-Methylthiazol
Methylpyrazin
Furfural
Trimethyloxazol
3-Methylbutansäure
2-Methylbutansäure
2-Hexenal
1-Hexanol
4-Heptanon
2,6-Dimethylpyridin
2,4-Dimethylthiazol
3-Heptanon
2-Heptanon
3-Heptanol
Heptanal
Methional
2,5-Dimethylpyrazin
2,6-Dimethylpyrazin
Ethylpyrazin
2,3-Dimethylpyrazin
Ethenylpyrazin
4,5-Dimethylthiazol
6-Methyl-2-Heptanon
2-Ethylhexanal
2-(Z)-Heptenal
5-Nonen-2-on
5-Methyl-2-Furancarboxaldehyd
Benzaldehyd
Hexansäure
1-Octen-3-ol
Dimethyltrisulfid
2,5-Octandion
6-Methyl-5-hepten-2-on
2-Pentylfuran
2,4-(E,E)-Heptadienal
2-Ethyl-6-methylpyrazin
Octanal
Trimethylpyrazin
2-Ethyl-3-methylpyrazin
2,4-(E,E)-Heptadien-1-al
2-Ethenyl-6-Methylpyrazin
2-Ethyl-1-hexanol
3-(E)-Octen-2-on
5,6-Dihydro-2H-pyran-2-on
Benzolacetaldehyd
3,5-(E,E)-Octadien-2-on
Acetophenon

1-Decen-3-on
3-Ethyl-2,5-dimethylpyrazin
Tetramethylpyrazin
5-Methyl-2-thiophencarboxaldehyd
γ-Heptalacton
Linalool
Nonanal
Thymol
Phenylethylalkohol
2,3,5-Trimethyl-6-ethylpyrazin
Essigsäurephenylmethylester
Safranal
2-(E)-Decenal
γ-Octalacon
o-Aminoacetophenon
2,4-Decadienal
γ-Nonalacton
Ionon
Geranylaceton
Ionen
γ-1-Nonalacton
2,4-(E,E)-Nonadienal
2,4-Decadien-1-al
γ-1-Heptalacton
Ion-1-on
1-Geranylaceton
α-Ionon
Pfirsichlacton, γ-Undecalacton
δ-Decalacton
cis-Geranylaceton
δ-Dodecalacton, δ-Nonyl-δ-valeralacton
δ-Undecalacton

unter Bezugnahme auf einen internen Standard,
wobei der Ellipsoid durch die Gleichung $Ax^2+Bxy+Cy^2+Dx+Ey+F = 0$ abgegrenzt ist und seine Kennwerte der folgenden Tabelle entsprechen:

X-Abmessung	Y-Abmessung	A	B	C	D	E	F
PC1	PC2	0,003481467	-0,000366174	3,79437E-05	-0,000628924	4,27301E-05	1,51548E-05
PC1	PC3	0,001734328	0,000286969	1,89401E-05	-0,000318201	-2,8099E-05	1,12003E-05
PC2	PC3	0,356218856	0,289219807	0,356936631	0,085191149	-0,040237159	-0,13812915

wobei anhand der folgenden Vorgehensweise bestimmt wird, ob es in dem Ellipsoiden liegt:

Bestimmen der relativen Konzentrationen für jede der Verbindungen;
Abziehen, für jede der Verbindungen, der Mittenfaktoren gemäß der unterstehenden Tabelle;
Teilen, für jede der Verbindungen, durch die Maßstabsfaktoren gemäß der untenstehenden Tabelle;
Erhalten von Ergebniswerten für PC1, PC2 und PC3 ausgehend von dem Punktprodukt der maßstabs- und mittenbezogen verarbeiteten Daten; und
Bestimmen, ob das Aromamerkmal, welches anhand von PC1, PC2 und PC3 bestimmt wurde, in dem Ellipsoiden liegt:

Chemischer Stoff	**Mitte**	**Maßstab**	PC1	PC2	PC3
Dimethylsulfid	15,04166667	52,10586179	0,007602386	-0,154648539	0,13795639
2,3-Butandion	573,4583333	687,3035077	-0,053406645	0,116238372	0,138457708
Butanal	165,0833333	291,8766733	-0,061200873	0,021748265	-0,154199309
2-Methylpropanal	294,25	321,9922006	-0,02479716	-0,203551061	0,142079295
3-Methylfuran	254,0833333	364,0905752	-0,139050167	0,053488926	-0,040009249
Ethylacetat	1534,958333	721,2414001	0,023033335	0,078632968	0,149060426
2-(E)-Butenal	56,95833333	67,74264748	0,034598984	-0,007869304	-0,228855217
3-Methylbutanal	2368,958333	3305,894731	-0,015854973	-0,209996041	0,152553963
1-Butanol	236,75	723,0508438	0,01482126	0,147080874	0,120323863
2-Methylbutanal	858,0416667	1132,843254	-0,069765232	-0,186610612	0,143374765
Thiophen	0,708333333	2,453738644	-0,145349572	0,003673658	-0,010721336
1-Penten-3-ol	111,2916667	123,2715883	-0,105910877	0,059069801	-0,020890092
1-Penten-3-on	10,625	18,86570361	0,029319785	-0,055925743	-0,186580083
2-Pentanon	429,875	520,4705967	0,018948769	-0,168215403	0,184382338
2,3-Pentandion	392,625	359,8726495	0,037715762	-0,074625863	-0,010390137
Pentanal	5315,166667	4258,727501	-0,05954475	-0,05904769	-0,130129097
2-Ethylfuran	32,75	24,43590875	-0,008414663	-0,076099651	-0,014167153
Thiazol	70,16666667	199,0549642	-0,142882049	-0,031332244	0,020544457
3-Penten-2-on	442,125	470,5612763	0,036579138	-0,118623927	0,193220234
Dimethyldisulfid	77,45833333	105,2821875	0,007660621	0,076749927	-0,030508003
2-(E)-Pentenal	116,7083333	200,60312	0,029036734	0,005658787	-0,063353931
Pyrrol	12,29166667	41,79846579	-0,145424967	0,001008736	-0,008354639
4,5-Dimethyloxazol	15,83333333	54,84827557	-0,145349572	0,003673658	-0,010721336
2-(Z)-Penten-1-ol	45,25	118,0232065	-0,141807908	-0,022407562	-0,007205637
3-Methylthiophen	108,5416667	279,7959856	0,006693629	0,144512146	0,116341706
Hexanal	26189,95833	17886,61913	0,023290612	0,064196972	-0,162118696
4-Methylthiazol	1,958333333	6,783865663	-0,145349572	0,003673658	-0,010721336
Methylpyrazin	135,2083333	326,6405766	-0,138842567	-0,055435505	0,03372617
Furfural	34,5	119,5115057	-0,145349572	0,003673658	-0,010721336
Trimethyloxazol	64	221,7025034	-0,145349572	0,003673658	-0,010721336

(fortgesetzt)

Chemischer Stoff	Mitte	Maßstab	PC1	PC2	PC3
3-Methylbutansäure	58,58333333	202,9386196	-0,145349572	0,003673658	-0,010721336
2-Methylbutansäure	3,833333333	13,27905619	-0,145349572	0,003673658	-0,010721336
2-Hexenal	25,58333333	50,09710268	0,027469429	-0,052249399	-0,23615517
1-Hexanol	106,1666667	155,9474465	0,031207096	0,198558566	0,011983686
4-Heptanon	360,5833333	577,8576749	-0,003575779	-0,135096305	0,010019679
2,6-Dimethylpyridin	2,958333333	10,24796728	-0,145349572	0,003673658	-0,010721336
2,4-Dimethylthiazol	15,58333333	53,98225017	-0,145349572	0,003673658	-0,010721336
3-Heptanon	111,625	94,41016052	0,021607662	-0,18444557	-0,171655667
2-Heptanon	380,875	288,460973	-0,097016748	-0,058868123	0,015417076
3-Heptanol	1193,041667	1008,348074	0,023029974	-0,205456135	-0,111328282
Heptanal	1396,791667	920,0702903	-0,113307135	0,141565621	-0,025917554
Methional	79,625	148,3023823	-0,110012922	-0,130400953	0,093977633
2,5-Dimethylpyrazin	3,333333333	7,857634774	0,020631611	-0,116950274	-0,004255769
2,6-Dimethylpyrazin	178,2083333	574,8013672	-0,145388496	-0,007146465	-0,001098366
Ethylpyrazin	15,95833333	53,8796885	-0,145442956	-0,0000479	-0,007415618
2,3-Dimethylpyrazin	439,2083333	1498,775644	-0,145413873	0,001518449	-0,008807482
Ethenylpyrazin	1,416666667	4,907477288	-0,145349572	0,003673658	-0,010721336
4,5-Dimethylthiazol	3,583333333	12,41303079	-0,145349572	0,003673658	-0,010721336
6-Methyl-2-Heptanon	53,75	186,1954618	-0,145349572	0,003673658	-0,010721336
2-Ethylhexanal	78,41666667	124,9672381	0,018460956	-0,027007294	-0,179937424
2-(Z)-Heptenal	645,25	937,3877266	0,021607084	-0,093800543	-0,190591625
5-Nonen-2-on	13,33333333	46,18802154	-0,145349572	0,003673658	-0,010721336
5-Methyl-2-Furancarboxaldehyd	21,25	40,57288615	0,019206035	-0,109620677	0,175448337
Benzaldehyd	872,875	1358,161493	-0,142431906	0,046335544	0,024776943
Hexansäure	176,25	216,4210438	-0,001128927	0,064879481	-0,016090326
1-Octen-3-ol	369,6666667	350,9919277	-0,090672545	-0,045064295	-0,135474824
Dimethyltrisulfid	14,33333333	21,56315601	0,028899179	-0,064852089	-0,150867075
2,5-Octandion	23,95833333	44,27674248	0,028988465	-0,07590479	-0,093752193
6-Methyl-5-hepten-2-on	1503,833333	4827,634134	-0,145266246	0,005470194	-0,014175912
2-Pentylfuran	633	967,4016276	-0,078384616	0,167579691	-0,035610073
2,4-(E,E)-Heptadienal	20,83333333	43,16371231	0,024003523	-0,071588186	-0,145038829
2-Ethyl-6-methylpyrazin	21	72,74613392	-0,145349572	0,003673658	-0,010721336
Octanal	1243,041667	897,5365644	0,063418428	0,197764097	-0,01447548
Trimethylpyrazin	348,6666667	1051,439497	-0,144625394	-0,018888681	0,009357594
2-Ethyl-3-methylpyrazin	87,33333333	302,5315411	-0,145349572	0,003673658	-0,010721336
2,4-(E,E)-Heptadien-1-al	26,33333333	40,42070427	0,033749609	-0,100784032	-0,199828071
2-Ethenyl-6-methylpyrazin	5,541666667	19,19689645	-0,145349572	0,003673658	-0,010721336

(fortgesetzt)

Chemischer Stoff	Mitte	Maßstab	PC1	PC2	PC3
2-Ethyl-1-hexanol	5684,541667	5078,453328	0,015454406	-0,147033095	-0,173896762
3-(E)-Octen-2-on	196,375	462,4334412	0,022433793	-0,027668713	-0,141800019
5,6-Dihydro-2H-pyran-2-on	683,3333333	845,025291	0,040235145	0,008083104	-0,001975331
Benzolacetaldehyd	31,83333333	60,74811383	0,01141478	-0,200551415	0,147671091
3,5-(E,E)-Octadien-2-on	455,125	426,6112306	0,024307307	0,191552198	-0,040535191
Acetophenon	42,375	56,41088104	0,034819826	0,112028714	0,067831917
1-Decen-3-on	3,125	9,100761706	0,014871492	-0,007143686	0,067973089
3-Ethyl-2,5-dimethylpyrazin	50,75	174,3908228	-0,145387371	0,002524067	-0,009700663
Tetramethylpyrazin	951,4583333	3113,918129	-0,145437121	-0,00391206	-0,005426362
5-Methyl-2-thiophencarboxalde hyd	57,375	198,7528302	-0,145349572	0,003673658	-0,010721336
γ-Heptalacton	2	6,92820323	0,012980337	0,140814237	0,118375646
Linalool	9,833333333	34,06366588	-0,145349572	0,003673658	-0,010721336
Nonanal	1528,416667	1335,036088	0,053558189	0,198785653	-0,109289305
Thymol	160,5833333	556,2769844	-0,145349572	0,003673658	-0,010721336
Phenylethylalkohol	135,9583333	416,085189	-0,145061726	-0,01428243	0,003239013
2,3,5-Trimethyl-6-ethylpyrazin	208,7083333	718,7459552	-0,145377878	0,002836895	-0,00997845
Essigsäurephenylmethylester	213,875	205,6043337	0,045438482	0,114758954	0,153953593
Safranal	47,29166667	163,8231389	-0,145349572	0,003673658	-0,010721336
2-(E)-Decenal	55,04166667	78,60616976	0,034351801	-0,012969523	-0,21493625
γ-Octalacon	10,625	28,57933535	0,016392036	0,14295305	0,096452129
o-Aminoacetophenon	15,5	32,17070943	0,022315438	0,204041622	0,018370134
2,4-Decadienal	9,416666667	24,16781606	0,0179089	0,169004115	-0,038947428
γ-Nonalacton	13,5	40,20345982	0,01493418	0,189230257	0,033376822
Ionon	101,3333333	351,0289637	-0,145349572	0,003673658	-0,010721336
Geranylaceton	652,75	2137,396627	-0,145423518	-0,002004031	-0,008551463
Ionen	159,7916667	553,5345706	-0,145349572	0,003673658	-0,010721336
γ-1-Nonalacton	6,58755	22,81994259	0,016371012	-0,075372449	-0,049632645
2,4-(E,E)-Nonadienal	18,07305674	30,64101284	0,031363408	-0,023742328	-0,174506137
2,4-Decadien-1-al	50,4716275	85,11825112	0,029518821	0,094376773	-0,171060695
γ-1-Heptalacton	17,25928968	42,07909242	0,017750131	0,158720982	-0,019846703
Ion-1-on	199,0162875	689,4126429	-0,145349572	0,003673658	-0,010721336
1-Geranylaceton	880,2922516	3049,421811	-0,145349572	0,003673658	-0,010721336
α-Ionon	335,0475951	1160,638915	-0,145349572	0,003673658	-0,010721336
Pfirsichlacton, γ-Undecalacton	72,77877498	34,06000193	0,097029409	-0,071461906	0,084434422
δ-Decalacton	85,57314465	106,5309321	0,034674859	-0,18805394	0,077061807

(fortgesetzt)

Chemischer Stoff	Mitte	Maßstab	PC1	PC2	PC3
cis-Geranylaceton	5,9584	20,64050306	0,011926134	0,016184168	-0,063393798
δ-Dodecalacton, δ-Nonyl-δ-valeralacton	1400,955104	491,4817796	0,130734715	-0,059212775	0,033318423
δ-Undecalacton	6472,792302	6394,323609	0,051826724	-0,042456918	-0,131176612

2. Mikroalgenmehl nach Anspruch 1, wobei das Aromamerkmal in einer enger gefassten Ellipse liegt, deren Kennwerte der nachstehenden Tabelle entsprechen:

PC1	PC2	0,000477458	-5,02181E-05	5,2037E-06	-8,62524E-05	5,86012E-06	3,01302E-06
PC1	PC3	0,00023785	3,93556E-05	2,5975E-06	-4,3639E-OS	-3,85357E-06	1,76892E-06
PC2	PC3	0,048852827	0,039664394	0,048951264	0,011683347	-0,005518234	-0,009118978

3. Mikroalgenmehl nach Anspruch 1 oder Anspruch 2, wobei das Aromamerkmal in einer noch enger gefassten Ellipse liegt, deren Kennwerte der nachstehenden Tabelle entsprechen:

PC1	PC2	2,78319E-05	-2,9273E-06	3,03333E-07	-5,0278E-06	3,41597E-07	2,11154E-07
PC1	PC3	1,38647E-05	2,29411E-06	1,51413E-07	-2,54379E-06	-2,24631E-07	1,11963E-07
PC2	PC3	-0,000665829	0,000466136	-0,000152694	0,000380618	-0,000136456	-4,14371E-05

4. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 3, wobei es mittels des folgenden Verfahrens erhalten werden kann:
Anzüchten einer Brühe von *Chlorella-protothecoides*-Zellen in der Dunkelheit in Gegenwart von Glucose als festgelegter Kohlenstoffquelle mit einem Ausgangs-pH-Wert von 6,8, wobei das Niveau an gelöstem Sauerstoff bei mehr als 30 % gehalten wird, Einwirkenlassen eines Hochtemperatur-Kurzzeit-Verfahrens von 75 °C über 1 Minute auf die Brühe, Gewinnen der Zellen durch Zentrifugation, wobei diese um den Faktor 6,4 in Wasser verdünnt werden, Zusetzen eines Antioxidans, Lyse der Zellen durch Zerkleinern, sowie Sprühtrocknen.

5. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 4, wobei es Undecalacton (400 bis 1.800 ppb), 3-Methylbutanal (0 bis 11.000 ppb), Pentanal (160 bis 10.700 ppb), 2-Methylbutanal (0 bis 2.500 ppb), 2-Pentanon (39 bis 10.600 ppb) und/oder 3-Penten-2-on (0 bis 1.500 ppb) umfasst, gemäß einer Bestimmung mittels SPME oder SBSE.

6. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 5, bei welchem keinerlei Fisch- oder Kohlaroma nachweisbar ist, wenn dieses Mehl zu 10 % (w/v) in deionisiertem Wasser aufgelöst wird, wobei der Nachweis durch ein Verkostungspanel erfolgt.

7. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 6, mit einer Fließfähigkeit, die durch eine Übergröße von 15 bis 35 Gewichts-% bei 2.000 μm gekennzeichnet ist.

8. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 7, wobei das Mehl von weißer, blassgelber oder gelber Farbe ist.

9. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 8, wobei das Mehl Folgendes umfasst

(i) 5 bis 20 % an Fettstoffen, oder
(ii) 30 bis 70 % an Fettstoffen, oder
(iii) 40 bis 60 % an Fettstoffen.

10. Mikrolagenmehl nach beliebigen der Ansprüche 1 bis 9, wobei der pH-Wert des Mehl, wenn es zu 1 % (w/v) in Wasser aufgelöst wird, im folgenden Bereich liegt:

(i) 5,5 bis 8,5, oder
(ii) 6,0 bis 8,0, oder
(iii) 6,5 bis 7,5.

11. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 10, wobei es weniger als 200 ppm an Chlorophyll, möglicherweise weniger als 2 ppm Chlorophyll aufweist.

12. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 11, wobei es weiterhin ein zugesetztes Antioxidans umfasst.

13. Mikroalgenmehl nach beliebigen der Ansprüche 1 bis 12, wobei die überwiegende Anzahl an Zellen in dem Mehl eine Lyse erfahren haben und möglicherweise zwischen 50 und 90 % der Zellen eine Lyse erfahren haben.

14. Verfahren zur Herstellung eines Mikrolalgenmehls, wobei das Verfahren die folgenden Schritte umfasst: Anzüchten einer Brühe von *Chlorella-protothecoides*-Zellen in der Dunkelheit in Gegenwart von Glucose als festgelegter Kohlenstoffquelle mit einem Ausgangs-pH-Wert von 6,8, wobei das Niveau an gelöstem Sauerstoff bei mehr als 30 % gehalten wird, Einwirkenlassen eines Hochtemperatur-Kurzzeit-Verfahrens von 75 °C über 1 Minute auf die Brühe, Gewinnen der Zellen durch Zentrifugation, wobei diese um den Faktor 6,4 in Wasser verdünnt werden, Zusetzen eines Antioxidans, Lyse der Zellen durch Zerkleinern, sowie Sprühtrocknen.

Revendications

1. Farine de microalgue appropriée pour une utilisation dans un produit alimentaire, la farine comprenant des cellules de microalgue de *Chlorophyta* et étant **caractérisée par** un descripteur d'arôme se situant dans un ellipsoïde dans un espace de description d'arôme possédant des dimensions de PC1, PC2 et PC3, le descripteur d'arôme étant produit en utilisant une analyse SPME et/ou SBSE pour déterminer des concentrations des composés suivants :

sulfure de diméthyle
2,3-butanedione
butanal
2-méthylpropanal
3-méthylfuranne
acétate d'éthyle
2-(E)-buténal
3-méthylbutanal
1-butanol
2-méthylbutanal
thiophène
1-pentén-3-ol
1-pentén-3-one
2-pentanone
2,3-pentanedione
pentanal
2-éthylfuranne
thiazole
3-pentén-2-one
disulfure de diméthyle
2-(E)-penténal
pyrrole
4,5-diméthyloxazol
2-(Z)-pentén-1-ol
3-méthylthiophène
hexanal
4-méthylthiazole
méthylpyrazine
furfural
triméthyloxazol
acide 3-méthylbutanoïque

acide 2-méthylbutanoïque
2-hexénal
1-hexanol
4-heptanone
2,6-diméthylpyridine
2,4-diméthylthiazole
3-heptanone
2-heptanone
3-heptanol
heptanal
méthional
2,5-diméthylpyrazine
2,6-diméthylpyrazine
éthylpyrazine
2,3-diméthylpyrazine
éthénylpyrazine
4,5-diméthylthiazole
6-méthyl-2-heptanone
2-éthylhexanal
2-(Z)-hepténal
5-nonén-2-one
5-méthyl-2-furannecarboxaldéhyde
benzaldéhyde
acide hexanoïque
1-octén-3-ol
trisulfure de diméthyle
2,5-octanedione
6-méthyl-5-heptén-2-one
2-pentylfuranne
2,4-(E,E)-heptadiénal
2-éthyl-6-méthylpyrazine
octanal
triméthylpyrazine
2-éthyl-3-méthylpyrazine
2,4-(E,E)-heptadiénal
2-éthényl-6-méthylpyrazine
2-éthyl-1-hexanol
3-(E)-octén-2-one
5,6-dihydro-2H-pyran-2-one
benzèneacétaldéhyde
3,5-(E,E)-octadién-2-one
acétophénone
1-décén-3-one
3-éthyl-2,5-diméthylpyrazine
tétraméthylpyrazine
5-méthyl-2-thiophènecarboxaldéhyde
γ-heptalactone
linalool
nonanal
thymol
alcool phényléthylique
2,3,5-triméthyl-6-éthylpyrazine
ester de phénylméthyle d'acide acétique
safranal
2-(E)-décénal
γ-octalactone
o-aminoacétophénone
2,4-décadiénal

γ-nonalactone
ionone
géranylacétone
ionène
γ-1-nonalactone
2,4-(E,E)-nonadiénal
2,4-décadién-1-al
γ-1-heptalactone
ion-1-one
1-géranylacétone
α-ionone
lactone de pêche, γ-undécalactone
δ-décalactone
cis-géranylacétone
δ-dodécalactone, δ-nonyl-δ-valéralactone
δ-undécalactone

par rapport à une référence interne,
l'ellipsoïde étant défini par l'équation $Ax^2 + Bxy + Cy^2 + Dx + Ey + F = 0$ et paramétrisé selon le tableau suivant :

Dimension X	Dimension Y	A	B	C	D	E	F
PC1	PC2	0,003481467	-0,000366174	3,79437E-05	-0,000628924	4,27301E-05	1,51548E-05
PC1	PC3	0,001734328	0,000286969	1,89401E-05	-0,000318201	-2,8099E-05	1,12003E-05
PC2	PC3	0,356218856	0,289219807	0,356936631	0,085191149	-0,040237159	-0,13812915

le fait qu'il se situe dans l'ellipsoïde étant déterminé par la procédure suivante :

pour chaque composé, détermination de concentrations relatives ;
pour chaque composé, soustraction de facteurs centraux selon le tableau ci-dessous ;
pour chaque composé, division par les facteurs d'échelle selon le tableau ci-dessous ;
prise du produit scalaire des données mises à l'échelle et centrées pour donner des valeurs pour PC1, PC2 et PC3 ; et
détermination si le descripteur d'arôme défini par PC1, PC2 et PC3 se situe dans l'ellipsoïde :

Composé chimique	**Centre**	**Échelle**	PC1	PC2	PC3
sulfure de diméthyle	15,04166667	52,10586179	0,007602386	-0,154648539	0,13795639
2,3-butanedione	573,4583333	687,3035077	-0,053406645	0,116238372	0,138457708
butanal	165,0833333	291,8766733	-0,061200873	0,021748265	-0,154199309
2-méthylpropanal	294,25	321,9922006	-0,02479716	-0,203551061	0,142079295
3-méthylfuranne	254,0833333	364,0905752	-0,139050167	0,053488926	-0,040009249
acétate d'éthyle	1534,958333	721,2414001	0,023033335	0,078632968	0,149060426
2-(E)-buténal	56,95833333	67,74264748	0,034598984	-0,007869304	-0,228855217
3-méthylbutanal	2368,958333	3305,894731	-0,015854973	-0,209996041	0,152553963
1-butanol	236,75	723,0508438	0,01482126	0,147080874	0,120323863
2-méthylbutanal	858,0416667	1132,843254	-0,069765232	-0,186610612	0,143374765
thiophène	0,708333333	2,453738644	-0,145349572	0,003673658	-0,010721336
1-pentén-3-ol	111,2916667	123,2715883	-0,105910877	0,059069801	-0,020890092
1-pentén-3-one	10,625	18,86570361	0,029319785	-0,055925743	-0,186580083
2-pentanone	429,875	520,4705967	0,018948769	-0,168215403	0,184382338
2,3-pentanedione	392,625	359,8726495	0,037715762	-0,074625863	-0,010390137
pentanal	5315,166667	4258,727501	-0,05954475	-0,05904769	-0,130129097
2-éthylfuranne	32,75	24,43590875	-0,008414663	-0,076099651	-0,014167153
thiazole	70,16666667	199,0549642	-0,142882049	-0,031332244	0,020544457
3-pentén-2-one	442,125	470,5612763	0,036579138	-0,118623927	0,193220234
disulfure de diméthyle	77,45833333	105,2821875	0,007660621	0,076749927	-0,030508003
2-(E)-penténal	116,7083333	200,60312	0,029036734	0,005658787	-0,063353931
pyrrole	12,29166667	41,79846579	-0,145424967	0,001008736	-0,008354639
4,5-diméthyloxazol	15,83333333	54,84827557	-0,145349572	0,003673658	-0,010721336
2-(Z)-pentén-1-ol	45,25	118,0232065	-0,141807908	-0,022407562	-0,007205637
3-méthylthiophène	108,5416667	279,7959856	0,006693629	0,144512146	0,116341706
hexanal	26189,95833	17886,61913	0,023290612	0,064196972	-0,162118696
4-méthylthiazole	1,958333333	6,783865663	-0,145349572	0,003673658	-0,010721336
méthylpyrazine	135,2083333	326,6405766	-0,138842567	-0,055435505	0,03372617
furfural	34,5	119,5115057	-0,145349572	0,003673658	-0,010721336
triméthyloxazol	64	221,7025034	-0,145349572	0,003673658	-0,010721336
acide 3-méthylbutanoïque	58,58333333	202,9386196	-0,145349572	0,003673658	-0,010721336
acide 2-méthylbutanoïque	3,833333333	13,27905619	-0,145349572	0,003673658	-0,010721336

(suite)

Composé chimique	Centre	Échelle	PC1	PC2	PC3
2-hexénal	25,58333333	50,09710268	0,027469429	-0,052249399	-0,23615517
1-hexanol	106,1666667	155,9474465	0,031207096	0,198558566	0,011983686
4-heptanone	360,5833333	577,8576749	-0,003575779	-0,135096305	0,010019679
2,6-diméthylpyridine	2,958333333	10,24796728	-0,145349572	0,003673658	-0,010721336
2,4-diméthylthiazole	15,58333333	53,98225017	-0,145349572	0,003673658	-0,010721336
3-heptanone	111,625	94,41016052	0,021607662	-0,18444557	-0,171655667
2-heptanone	380,875	288,460973	-0,097016748	-0,058868123	0,015417076
3-heptanol	1193,041667	1008,348074	0,023029974	-0,205456135	-0,111328282
heptanal	1396,791667	920,0702903	-0,113307135	0,141565621	-0,025917554
méthional	79,625	148,3023823	-0,110012922	-0,130400953	0,093977633
2,5-diméthylpyrazine	3,333333333	7,857634774	0,020631611	-0,116950274	-0,004255769
2,6-diméthylpyrazine	178,2083333	574,8013672	-0,145388496	-0,007146465	-0,001098366
éthylpyrazine	15,95833333	53,8796885	-0,145442956	-0,0000479	-0,007415618
2,3-diméthylpyrazine	439,2083333	1498,775644	-0,145413873	0,001518449	-0,008807482
éthénylpyrazine	1,416666667	4,907477288	-0,145349572	0,003673658	-0,010721336
4,5-diméthylthiazole	3,583333333	12,41303079	-0,145349572	0,003673658	-0,010721336
6-méthyl-2-heptanone	53,75	186,1954618	-0,145349572	0,003673658	-0,010721336
2-éthylhexanal	78,41666667	124,9672381	0,018460956	-0,027007294	-0,179937424
2-(Z)-hepténal	645,25	937,3877266	0,021607084	-0,093800543	-0,190591625
5-nonén-2-one	13,33333333	46,18802154	-0,145349572	0,003673658	-0,010721336
5-méthyl-2-furannecarboxaldéh yde	21,25	40,57288615	0,019206035	-0,109620677	0,175448337
benzaldéhyde	872,875	1358,161493	-0,142431906	0,046335544	0,024776943
acide hexanoïque	176,25	216,4210438	-0,001128927	0,064879481	-0,016090326
1-octén-3-ol	369,6666667	350,9919277	-0,090672545	-0,045064295	-0,135474824
trisulfure de diméthyle	14,33333333	21,56315601	0,028899179	-0,064852089	-0,150867075
2,5-octandione	23,95833333	44,27674248	0,028988465	-0,07590479	-0,093752193
6-méthyl-5-heptén-2-one	1503,833333	4827,634134	-0,145266246	0,005470194	-0,014175912
2-pentylfuranne	633	967,4016276	-0,078384616	0,167579691	-0,035610073
2,4-(E,E)-heptadiénal	20,83333333	43,16371231	0,024003523	-0,071588186	-0,145038829
2-éthyl-6-méthylpyrazine	21	72,74613392	-0,145349572	0,003673658	-0,010721336
octanal	1243,041667	897,5365644	0,063418428	0,197764097	-0,01447548
triméthylpyrazine	348,6666667	1051,439497	-0,144625394	-0,018888681	0,009357594
2-éthyl-3-méthylpyrazine	87,33333333	302,5315411	-0,145349572	0,003673658	-0,010721336
2,4-(E,E)-heptadiénal	26,33333333	40,42070427	0,033749609	-0,100784032	-0,199828071
2-éthényl-6-méthylpyrazine	5,541666667	19,19689645	-0,145349572	0,003673658	-0,010721336
2-éthyl-1-hexanol	5684,541667	5078,453328	0,015454406	-0,147033095	-0,173896762
3-(E)-octén-2-one	196,375	462,4334412	0,022433793	-0,027668713	-0,141800019

(suite)

Composé chimique	Centre	Échelle	PC1	PC2	PC3
5,6-dihydro-2H-pyran-2-one	683,3333333	845,025291	0,040235145	0,008083104	-0,001975331
benzèneacétaldéhyd e	31,83333333	60,74811383	0,01141478	-0,200551415	0,147671091
3,5-(E,E)-octadién-2-one	455,125	426,6112306	0,024307307	0,191552198	-0,040535191
acétophénone	42,375	56,41088104	0,034819826	0,112028714	0,067831917
1-décén-3-one	3,125	9,100761706	0,014871492	-0,007143686	0,067973089
3-éthyl-2,5-diméthylpyrazine	50,75	174,3908228	-0,145387371	0,002524067	-0,009700663
tétraméthylpyrazin e	951,4583333	3113,918129	-0,145437121	-0,00391206	-0,005426362
5-méthyl-2-thiophènecarboxald éhyde	57,375	198,7528302	-0,145349572	0,003673658	-0,010721336
γ-heptalactone	2	6,92820323	0,012980337	0,140814237	0,118375646
linalool	9,833333333	34,06366588	-0,145349572	0,003673658	-0,010721336
nonanal	1528,416667	1335,036088	0,053558189	0,198785653	-0,109289305
thymol	160,5833333	556,2769844	-0,145349572	0,003673658	-0,010721336
alcool phényléthylique	135,9583333	416,085189	-0,145061726	-0,01428243	0,003239013
2,3,5-triméthyl-6-éthylpyrazine	208,7083333	718,7459552	-0,145377878	0,002836895	-0,00997845
ester de phénylméthyle d'acide acétique	213,875	205,6043337	0,045438482	0,114758954	0,153953593
safranal	47,29166667	163,8231389	-0,145349572	0,003673658	-0,010721336
2-(E)-décénal	55,04166667	78,60616976	0,034351801	-0,012969523	-0,21493625
γ-octalacone	10,625	28,57933535	0,016392036	0,14295305	0,096452129
o-aminoacétophénone	15,5	32,17070943	0,022315438	0,204041622	0,018370134
2,4-décadiénal	9,416666667	24,16781606	0,0179089	0,169004115	-0,038947428
γ-nonalactone	13,5	40,20345982	0,01493418	0,189230257	0,033376822
ionone	101,3333333	351,0289637	-0,145349572	0,003673658	-0,010721336
géranylacétone	652,75	2137,396627	-0,145423518	-0,002004031	-0,008551463
ionène	159,7916667	553,5345706	-0,145349572	0,003673658	-0,010721336
γ-1-nonalactone	6,58755	22,81994259	0,016371012	-0,075372449	-0,049632645
2,4-(E,E)-nonadiénal	18,07305674	30,64101284	0,031363408	-0,023742328	-0,174506137
2,4-décadién-1-al	50,4716275	85,11825112	0,029518821	0,094376773	-0,171060695
γ-1-heptalactone	17,25928968	42,07909242	0,017750131	0,158720982	-0,019846703
ion-1-one	199,0162875	689,4126429	-0,145349572	0,003673658	-0,010721336
1-géranylacétone	880,2922516	3049,421811	-0,145349572	0,003673658	-0,010721336
α-ionone	335,0475951	1160,638915	-0,145349572	0,003673658	-0,010721336
lactone de pêche, γ-undécalactone	72,77877498	34,06000193	0,097029409	-0,071461906	0,084434422
δ-décalactone	85,57314465	106,5309321	0,034674859	-0,18805394	0,077061807
cis-géranylacétone	5,9584	20,64050306	0,011926134	0,016184168	-0,063393798

(suite)

Composé chimique	Centre	Échelle	PC1	PC2	PC3
δ-dodécalactone, δ-nonyl-δ-valéralactone	1400,955104	491,4817796	0,130734715	-0,059212775	0,033318423
δ-undécalactone	6472,792302	6394,323609	0,051826724	-0,042456918	-0,131176612

2. Farine de microalgue selon la revendication 1, le descripteur d'arôme se situant dans une ellipse plus étroite paramétrisée par le tableau ci-dessous :

PC1	PC2	0,000477458	-5,02181E-05	5,2037E-06	-8,62524E-05	5,86012E-06	3,01302E-06
PC1	PC3	0,00023785	3,93556E-05	2,5975E-06	-4,3639E-05	-3,85357E-06	1,76892E-06
PC2	PC3	0,048852827	0,039664394	0,048951264	0,011683347	-0,005518234	-0,009118978

3. Farine de microalgue selon la revendication 1 ou la revendication 2, le descripteur d'arôme se situant dans une ellipse encore plus étroite paramétrisée par le tableau ci-dessous :

PC1	PC2	2,78319E-05	-2,9273E-06	3,03333E-07	-5,0278E-06	3,41597E-07	2,11154E-07
PC1	PC3	1,38647E-05	2,29411E-06	1,51413E-07	-2,54379E-06	-2,24631E-07	1,11963E-07
PC2	PC3	-0,000665829	0,000466136	-0,000152694	0,000380618	-0,000136456	-4,14371E-05

4. Farine de microalgue selon l'une quelconque des revendications 1 à 3, pouvant être obtenue par le procédé de : culture d'un bouillon de cellules de *Chlorella protothecoides* dans l'obscurité en la présence de glucose comme source de carbone fixe avec un pH de départ de 6,8, tout en maintenant le taux d'oxygène dissous au-dessus de 30 %, soumission du bouillon à un procédé de haute température-courte durée de 75 °C pendant 1 minute, récolte des cellules par centrifugation avec une dilution de 6,4 fois dans l'eau, ajout d'un antioxydant, lyse des cellules par broyage, et séchage par pulvérisation.

5. Farine de microalgue selon l'une quelconque des revendications 1 à 4, comprenant de l'undécalactone (400 à 1 800 ppb), du 3-méthylbutanal (0 à 11 000 ppb), du pentanal (160 à 10 700 ppb), du 2-méthylbutanal (0 à 2 500 ppb), de la 2-pentanone (39 à 10 600 ppb) et/ou de la 3-pentén-2-one (0 à 1 500 ppb), comme déterminé par SPME ou SBSE.

6. Farine de microalgue selon l'une quelconque des revendications 1 à 5, possédant un arôme de poisson ou de chou indétectable lorsque la farine est dispersée dans de l'eau désionisée à raison de 10 pour 100 (p/v), comme détecté par un panel de dégustation.

7. Farine de microalgue selon l'une quelconque des revendications 1 à 6, possédant une fluidité **caractérisée par** un surdimensionnement de 15 à 35 % en poids à 2 000 μm.

8. Farine de microalgue selon l'une quelconque des revendications 1 à 7, la farine étant d'une couleur blanche, jaune pâle ou jaune.

9. Farine de microalgue selon l'une quelconque des revendications 1 à 8, la farine comprenant

(i) 5 à 20 % de lipides, ou
(ii) 30 à 70 % de lipides, ou
(iii) 40 à 60 % de lipides.

10. Farine de microalgue selon l'une quelconque des revendications 1 à 9, le pH de la farine lorsqu'elle est dissoute dans de l'eau à raison de 1 % (p/v) étant compris entre :

(i) 5,5 et 8,5, ou
(ii) 6,0 et 8,0, ou
(iii) 6,5 et 7,5.

11. Farine de microalgue selon l'une quelconque des revendications 1 à 10, possédant moins de 200 ppm de chlorophylle, éventuellement moins de 2 ppm de chlorophylle.

12. Farine de microalgue selon l'une quelconque des revendications 1 à 11, comprenant en outre un antioxydant ajouté.

13. Farine de microalgue selon l'une quelconque des revendications 1 à 12, la majorité des cellules dans la farine étant lysées et éventuellement entre 50 et 90 % des cellules étant lysées.

14. Procédé de préparation d'une farine de microalgue, le procédé comprenant les étapes de :
culture d'un bouillon de cellules de *Chlorella protothecoides* dans l'obscurité en la présence de glucose comme source de carbone fixe avec un pH de départ de 6,8, tout en maintenant le taux d'oxygène dissous au-dessus de 30 %, soumission du bouillon à un procédé de haute température-courte durée de 75 °C pendant 1 minute, récolte des cellules par centrifugation avec une dilution de 6,4 fois dans l'eau, ajout d'un antioxydant, lyse des cellules par broyage, et séchage par pulvérisation.

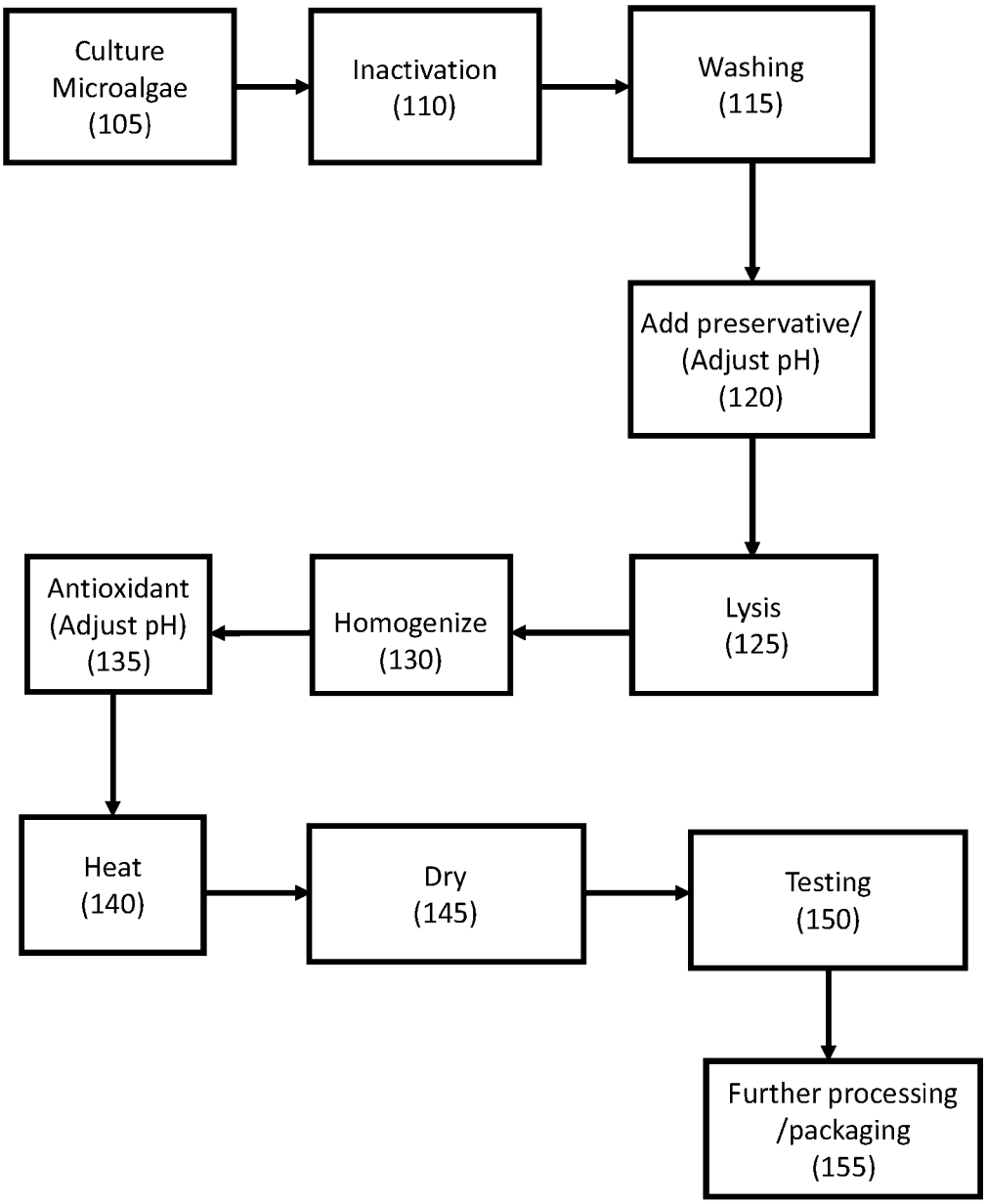
Culture Microalgae (105)
Inactivation (110)
Washing (115)
Add preservative/ (Adjust pH) (120)
Lysis (125)
Homogenize (130)
Antioxidant (Adjust pH) (135)
Heat (140)
Dry (145)
Testing (150)
Further processing /packaging (155)

Fig. 1

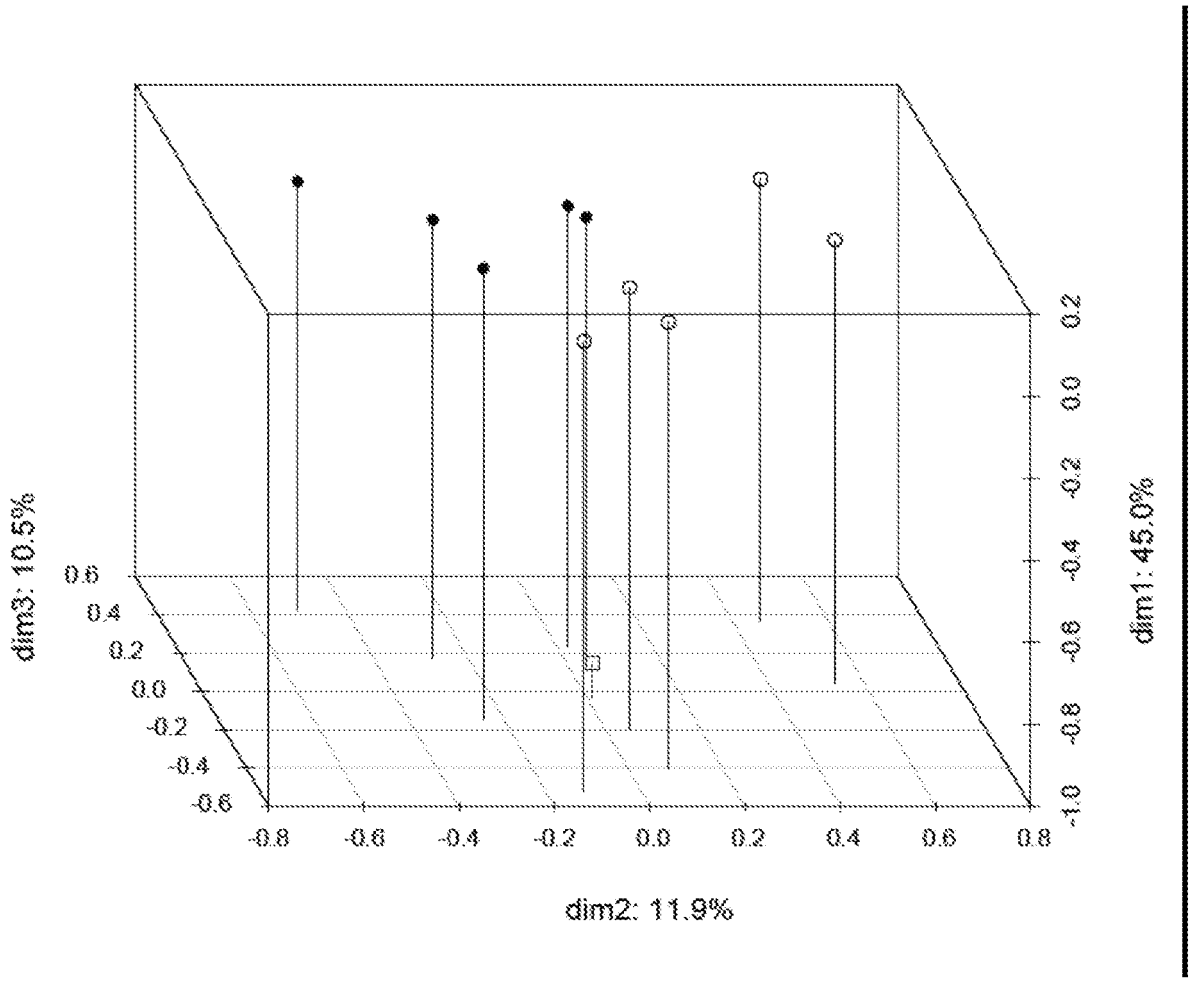
dim3: 10.5%
0.6
0.4
0.2
0.0
-0.2
-0.4
-0.6
-0.8
-0.6
-0.4
-0.2
0.0
0.2
0.4
0.6
0.8
dim2: 11.9%
0.2
0.0
-0.2
-0.4
-0.6
-0.8
-1.0
dim1: 45.0%

Fig. 2

REFERENCES CITED IN THE DESCRIPTION

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Patent documents cited in the description

- WO 201012093 A **[0005]**

Non-patent literature cited in the description

- **S. BRUNAUER et al.** BET Surface Area by Nitrogen Absorption. *Journal of American Chemical Society,* 1938, vol. 60, 309 **[0033]**