

CLAIMS

1. A heterotrophic method of culturing algae comprising:
 - (a) Growing algae in a medium comprising a carbon source and a nitrogen source, wherein the oxygen saturation of the medium is above 75%.
- 5 2. A method according to claim 1, wherein the carbon source comprises a sugar, carbohydrate or polyol, or any combination thereof.
3. A method according to claim 2, wherein the sugar is selected from a monosaccharide, disaccharide, oligosaccharide, polysaccharide or any combination thereof; preferably the sugar is selected from: glucose, mannose, ribose, xylose, fructose, galactose, sucrose,
 10 lactose, isomaltose, trehalose, and maltose, maltitriose, raffinose, stachyose, verbascose, maltodextrose, cellodextrose, or any combination thereof.
4. A method according to claim 2 wherein the polyol is selected from glycerol, erythritol, inositol, lactitol, mannitol, xorbitol, xylitol, or any combination thereof, and wherein the carbohydrate is selected from: isomalt, cellulose, hemicellulose, pectin, starch, glycogen,
 15 chitin, chitosan, guar gum, beta-glucan, alginate, acacia gum, beta mannan, inulin, tara gum, xanthan gum, carrageenan gum, polydextrose, glucomannan, or any combination thereof.
5. A method according to claim 1, wherein the carbon source comprises glucose, fructose, sucrose, or glycerol, or any combination thereof.
- 20 6. A heterotrophic method of culturing algae comprising:
 - (b) Growing algae in a medium comprising a carbon source and a nitrogen source, wherein a majority of the carbon source is glycerol and wherein the oxygen saturation of the medium is above 75%.
7. A method according to any of claims 1-6, wherein the medium is acidic.
- 25 8. A method according to any preceding claim, wherein the nitrogen source is a sole nitrogen source, preferably wherein the sole nitrogen source is a base.
9. A heterotrophic method of culturing algae comprising:
 - (a) growing algae in an acidic medium comprising a carbon source and a sole nitrogen source, wherein the sole nitrogen source is a base which is added into the acidic medium during culture and which modifies the pH of the acidic medium.
- 30 10. A method according to claim 9, wherein the carbon source is as defined in any of claims 2-5.
11. A heterotrophic method of culturing algae comprising:
 - (a) growing algae in an acidic medium comprising a carbon source and a sole nitrogen source, wherein a majority of the carbon source is glycerol; and wherein the sole nitrogen source is a base which is added into the acidic medium during culture and which modifies the pH of the acidic medium.
- 35 12. A method according any of claims 9-11 wherein the oxygen saturation of the medium is above 75%.
- 40 13. A method according to any of claims 1-12, wherein the oxygen saturation of the medium is above 80%, above 85%, above 90%, above 95%.

14. A method according to any of claims 1-13- wherein at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% of the carbon source is glycerol, preferably the carbon source consists of glycerol.
- 5 15. A method according to any of claims 6-8 or 11-14, wherein the glycerol is least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, least 95%, at least 99% pure.
16. A method according to any of claims 8-15, wherein the nitrogen source comprises a source of ammonia, preferably wherein the sole nitrogen source is selected from ammonia, ammonium hydroxide, ammonium sulphate, and ammonium phosphate.
- 10 17. A method according to any of claims 8-16, wherein the nitrogen source is ammonia gas.
18. A method according to any of claims 1-17, wherein addition of the nitrogen source into the acidic medium is automated, preferably by a pH control system.
19. A method according to any of claims 7-18, wherein the acidic medium is about pH 2.
- 15 20. A method according to any preceding claim wherein the algae is a red algae of the class *Cyanidiophyceae*, preferably of the order *Cyanidiales*, preferably of the family *Cyanidiaceae* or *Galdieriaceae*, preferably of the genus *Cyanidioschyzon*, *Cyanidium* or *Galdieria*, preferably of the species *Cyanidioschyzon merolae* 10D, *Cyanidioschyzon merolae* DBV201, *Cyanidium caldarium*, *Cyanidium daedalum*, *Cyanidium maximum*, 20 *Cyanidium partitum*, *Cyanidium rumpens*, *Galdieria daedala*, *Galdieria maxima*, *Galdieria partita* or *Galdieria sulphuraria*.
21. A method according to claim 20, wherein the algae is *Galdieria sulphuraria*, preferably selected from the following strains: ACUF141, SAG 108.79, 074G, 074G-G1, 074G-G2, CCME 5587.1, SAG 108.71, and UTEX#2919, preferably the algae is *Galdieria sulphuraria* 25 strain ACUF141.
22. A method according to any preceding claim wherein the method is continuous.
23. A method according to any preceding claim wherein the temperature of the medium is between 35 to 55°C, preferably the temperature of the medium is about 42°C.
24. A method according to any preceding claim, wherein the method further comprises a step 30 of collecting algal biomass from the culture medium.
25. A method according to any preceding claim, wherein the method produces between 35 and 45 g.L⁻¹.day⁻¹ of algal biomass.
26. A method of producing phycocyanin comprising culturing algae according to the method of any of claims 1-25.
- 35 27. A method according to any preceding claim, wherein the algal biomass has a mean intracellular concentration of phycocyanin of between 25-50 mg.g⁻¹ dry cell weight.
28. A method according to claims 26 or 27, wherein the method produces between 1-2.5 g.L⁻¹.day⁻¹ of phycocyanin.
29. Algal biomass produced by the method of any of claims 1-28.

30. Algal biomass having a mean intracellular concentration of phycocyanin of at least 25 mg.g⁻¹ dry cell weight and a mean intracellular concentration of allophycocyanin of less than 1.0 mg.g⁻¹ dry cell weight.
31. A method of preparing a valuable chemical from the algal biomass of claims 29 or 30.
- 5 32. A method according to claim 31 comprising the steps of (a) lysing the algal cells and (ii) purifying the valuable chemical from the lysate.
33. A method according to claims 31 or 32, wherein the valuable chemical comprises phycocyanin.
34. A reactor for culturing algae comprising a container for housing a culture medium, 10 wherein the container has a total volume and a working volume, wherein the working volume is a part of the total volume which is operable to contain the culture medium, and wherein the ratio of the total volume: working volume of the container is at least 2:1.
35. The reactor according to claim 34, further comprising an automated pH control system.
36. The reactor according to claims 34 or 35 wherein the reactor further comprises a pair of 15 baffles located within the container.
37. The reactor according to claim 36, wherein the baffles comprise a triangular profile.
38. The reactor according to any of claims 34-37, wherein the ratio of the total volume:working volume of the container is at least 3:1, preferably at least 4:1.