

ANNEX A: AUXILIARY REQUEST 1

1A. A process for obtaining a lipid from a microbial cell, said process comprising:

- a) lysing a cell to form a lysed cell composition;
- b) adding a base to the lysed cell composition, raising the pH of the lysed cell composition to 8 or above to demulsify the cell composition;
- c) one or more of c1, c2, c3 and c4;
 - c1) adding a salt to the lysed cell composition to demulsify the cell composition
 - c2) heating the lysed cell composition to demulsify the cell composition
 - c3) agitating the lysed cell composition to demulsify the cell composition
 - c4) adding a second base to the lysed cell composition to demulsify the cell composition;

and

- d) separating a lipid from the demulsified cell composition;

wherein the lipid contains less than 5% by weight of an organic solvent and wherein the lysing comprises enzymatic treatment.

2A. The process according to claim 1A, wherein the process comprises adding a salt to the lysed cell composition to demulsify the cell composition.

3A. The process of claim 2A, wherein the salt is added in an amount of 0.1% to 20% by weight of the lysed cell composition.

4A. The process of claim 2A or 3A, wherein the salt is selected from the group consisting of: alkali metal salts, alkali earth metal salts, sulfate salts, and combinations thereof.

5A. The process according to any preceding claim, wherein the process comprises heating the lysed cell composition to demulsify the cell composition.

6A. The process according to claim 5A, wherein the heating is performed after the adding a salt.

7A. The process according to any preceding claim, wherein the process comprises agitating the lysed cell composition to demulsify the cell composition.

~~8. The process of any preceding claim, wherein the raising of the pH of the lysed cell composition is performed after the adding a salt or the heating.~~

~~98A.~~ The process of any preceding claim, wherein the process comprises agitating the lysed cell composition by stirring, mixing, blending, shaking, vibrating, or a combination thereof.

~~10. The process of any preceding claim, wherein the lysing comprises mechanical treatment, physical treatment, chemical treatment, enzymatic treatment, or a combination thereof.~~

~~149A.~~ The process of any preceding claim, wherein the separating comprises centrifuging.

~~1210A.~~ The process of any preceding claim, wherein the process provides a lipid having an anisidine value of 26 or less.

~~1311A.~~ The process of any preceding claim, further comprising refining the lipid.

~~1412A.~~ The process of claim ~~1311A~~, wherein the refining is selected from the group consisting of: refining, degumming, acid treatment, alkali treatment, cooling, heating, bleaching, deodorizing, deacidification, and combinations thereof.

~~1513A.~~ The process of any preceding claim, wherein the microbial cell is an algae, bacteria, fungus, protist, or combination thereof.

~~1614A.~~ The process of claim ~~1513A~~, wherein the microbial cell is a member of the order Thraustochytriales.

~~1715A.~~ The process of claim ~~1614A~~, wherein the microbial cell is of the genus *Thraustochytrium*, *Schizochytrium*, or mixtures thereof.

~~1816A.~~ The process of any preceding claim, wherein the lipid comprises one or more polyunsaturated fatty acids.