CLAIMS

1. A method for obtaining a stabilised suspension of red blood cells (RBCs) encapsulating an active ingredient, from resealed RBCs incorporating the active ingredient, the method comprising the incubation of the resealed RBCs in an incubation solution, at an osmolality of no less than 280 mOsmol/kg, for a time of 30 minutes or more, the incubation solution being a solution that does not contain a bridging or cross-linking agent which is denaturing for the RBC membrane, the liquid medium is then removed from the incubated suspension and the RBCs obtained are placed in suspension in a solution allowing the injection of the suspension in a patient.

2. The method of claim 1, comprising, before incubation, encapsulating by lysis-resealing the active ingredient into RBCs and obtaining resealed RBCs comprising the active ingredient.

3. The method according to claim 1 or 2, wherein the incubation is performed at an osmolality of between about 280 and about 380 mOsmol/kg.

4. The method according to claim 1 or 2, wherein the incubation is performed at an osmolality of between about 290 and about 330 mOsmol/kg.

5. The method of any one of claims 1 to 4, wherein the incubation has a duration that is 1 h or longer.

6. The method of any one of claims 1 to 5, further comprising washing the resealed RBCs before the incubation.

7. The method of claim 6, wherein washing comprises at least one washing cycle comprising dilution of or placing in suspension the resealed RBCs in a solution, at an osmolality of no less than 280 mOsmol/kg, then separation.

8. The method of claim 7, wherein washing comprises 2 or 3 washing cycles.

9. The method according to claim 1, comprising the following steps:

(a) encapsulating an active ingredient inside RBCs, comprising the contacting with a hypotonic medium, contacting with the active ingredient, resealing of the RBCs;

(b) obtaining a suspension or residue comprising RBCs incorporating the active ingredient and a solution at an osmolality of 280 mOsmol/kg or higher;

(c) incubating the residue or suspension of step (b) as such or after addition of an incubation solution at an osmolality of 280 mOsmol/kg or higher, for a time of 30 minutes or longer;

(d) eliminating the liquid medium from the suspension incubated at step (c),

(e) placing in suspension the RBCs obtained at (d) in a solution allowing injection of the suspension in a patient.

10. A method as claimed in claim 9 wherein the solution of step (e) is a preservation solution.

11. The method according to claim 9 or 10, wherein step (b) comprises at least one 1 washing cycle, by dilution of the suspension or residue obtained at (a) in a solution, at an osmolality of 280 mOsmol/kg or higher, then obtaining a residue or suspension of RBCs.

12. The method according to any one of claims 9 to 11 wherein at step (d) either a separation is performed, or a dilution before separation.

13. The method according to any one of claims 1 to 12, wherein the incubation step is conducted at a temperature of between about 2 and about 39 °C.

14. The method according to any one of claims 1 to 13, wherein:

- the incubation step is conducted at low temperature between about 2 and about 10 °C, and lasts about 1 h to about 72 h; or
- the incubation step is conducted at a temperature between about 20 and about 39 °C, and lasts about 30 min to about 1 h.

15. The method according to any one of claims 1 to 14, comprising the recovering of a stabilised RBC suspension having an extracellular haemoglobin level maintained at 0.5 or lower, or 0.3 or lower, or 0.2 or lower, or 0.15 or lower, or 0.1 g/dl or lower and/or a haemolysis rate maintained at 2 or less, or 1.5 or less, or 1 % or less at 72 h after placing in suspension in the preservation solution and at a temperature of between 2 and 8 °C.

16. The method according to any one of claims 1 to 15, comprising the recovering of a stabilised RBC suspension having an extracellular haemoglobin level maintained at 0.5 g/dl or lower, or 0.3 g/dl or lower, or 0.2 g/dl or lower, or 0.15 g/dl or lower, or 0.1 g/dl or lower and/or a haemolysis rate maintained at 2 % or less, or 1.5 % or less, or 1 % or less for a time of between 24 h and 20 days, or between 24 and 72 h after placing in suspension in the preservation solution and at a temperature of between 2 and 8 °C.

17. The method according to any one of claims 1 to 16, wherein the active ingredient is chosen from among: L-asparaginase, IHP, ADI, Factor VIII, Factor IX, alglucosidase, beta-glucosidase, bisphosphonates, 2^{nd} and 3^{rd} generation bisphosphonates, uricase, thymidine phosphorylase, adenosine deaminase.

18. A stabilised suspension of RBCs encapsulating an active ingredient, obtained by implementing the method according to any one of claims 1 to 17.

19. The suspension according to claim 18, wherein an extracellular haemoglobin level is maintained at 0.5 g/dl or lower, or 0.3 g/dl or lower, or 0.2 g/dl or

lower, or 0.15 g/dl or lower, or 0.1 g/dl or lower and/or a haemolysis rate is maintained at 2 % or less, or 1.5 % or less, or 1 % or less at 72 h after placing in suspension in the preservation solution and at a temperature of between 2 and 8 °C.

20. The suspension according to claim 18, wherein an extracellular haemoglobin level maintained at 0.5 g/dl or lower, or 0.3 g/dl or lower, or 0.2 g/dl or lower, or 0.15 g/dl or lower, or 0.1 g/dl or lower and/or a haemolysis rate is maintained at 2 % or less, or 1.5 % or less, or 1 % or less for a time of between 24 h and 20 days, or between 24 and 72 h after placing in suspension in the preservation solution and at a temperature of between 2 and 8 °C.

21. The suspension according to any one of claims 18 to 20, wherein the active ingredient is chosen from among: L-asparaginase, IHP, ADI, Factor VIII, Factor IX, alglucosidase, beta-glucosidase, bisphosphonates, 2^{nd} and 3^{rd} generation bisphosphonates, uricase, thymidine phosphorylase, adenosine deaminase.