

FORM 2

THE PATENTS ACT, 1970
(39 of 1970)
AND
THE PATENTS RULES, 2003

**COMPLETE
SPECIFICATION**

(See Section 10; rule 13)

TITLE OF THE INVENTION

**“BIOSEPARATION COMPOSITIONS AND METHODS FOR MAKING
AND USING SAME”**

APPLICANT

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The following specification particularly describes
the invention and the manner in which
it is to be performed

WHAT IS CLAIMED IS:

1. A composition for use in bioseparation, the composition comprising:
a plurality of hollow particles, the hollow particles comprising a siliceous surface;
and
5 a surface-modifying agent bonded to the hollow particles, wherein the surface-modifying agent comprises a binding segment and a reactive segment, wherein the binding segment comprises a silyl group, and wherein the reactive segment comprises a reactive nitrogen group;
wherein the plurality of hollow particles have an average density between about
10 0.05 and 0.4 g/ml.
2. The composition according to claim 1, wherein the hollow particles comprise glass bubbles.
3. The composition according to any one the preceding claims, wherein a size distribution of the plurality of hollow particles has a span of less than 1.0.
- 15 4. The composition according to any one the preceding claims, wherein a mean particle size of the plurality of hollow particles ranges from about 10 micrometers to about 100 micrometers.
5. The composition according to any one the preceding claims, wherein the binding segment has a formula:
20
$$-\text{Si}(\text{R}^1)_{3-x}(\text{R}^2)_x;$$

wherein $x = 0, 1, \text{ or } 2$;
wherein each group R^1 comprises independently OH or a hydrolyzable group from among halo, alkoxy, aryloxy, aralkoxy, and acyloxy; and
wherein each group R^2 comprises independently a non-hydrolyzable group from
25 among alkyl, aryl, and aralkyl.
6. The composition according to any one the preceding claims, wherein the surface-modifying agent further comprises a linking segment, the linking segment comprising alkylene, arylene, or both, and optionally further comprising $-\text{NH}-$ or alkyleneoxy, or both.
- 30 7. The composition according to any one the preceding claims, wherein the reactive segment has a formula:
$$-\text{N}(\text{R}^3)_2, \text{ or salts thereof};$$

wherein each group R^3 comprises independently hydrogen, alkyl, aryl, or aralkyl.
8. A method for making a bioseparation composition, the method comprising:

size fractionating a first plurality of hollow particles having a first particle size distribution with a first span to form a second plurality of hollow particles having a second particle size distribution with a second span that is less than the first span; and

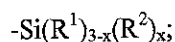
5 surface-modifying either or both of the first and second plurality of hollow particles with a surface modifying agent to form surface-modified hollow particles, wherein the surface-modifying agent comprises a binding segment and a reactive segment, wherein the binding segment comprises a silyl group, and wherein the reactive segment comprises a reactive nitrogen group.

10 9. The method according to claim 8, wherein the first and second plurality of hollow particles comprise glass bubbles.

10. The method according to any one of claims 8-9, wherein the second span is less than 1.0.

11. The method according to any one of claims 8-10, wherein a mean particle size of the second plurality of hollow particles ranges from about 10 micrometers to about 100 micrometers.

12. The method according to any one of claims 8-11, wherein the binding segment has a formula:



20 wherein $x = 0, 1, \text{ or } 2$;

wherein each group R^1 comprises independently OH^- or a hydrolyzable group from among halo, alkoxy, aryloxy, aralkyloxy, and acyloxy; and

wherein each group R^2 comprises independently a non-hydrolyzable group from among alkyl, aryl, and aralkyl.

25 13. The method according to any one of claims 8-12, wherein the surface-modifying agent further comprises a linking segment, the linking segment comprising alkylene, arylene, or both, and optionally further comprising $-\text{NH}-$ or alkyleneoxy, or both.

14. The method according to any one of claims 8-13, wherein the reactive segment has a formula:

30 $-\text{N}(\text{R}^3)_2$, or salts thereof;

wherein each group R^3 comprises independently hydrogen, alkyl, aryl, or aralkyl.

15. A method for capturing an analyte the method comprising:

providing a bioseparation composition comprising:

a plurality of hollow particles; and

35 a surface-modifying agent bonded to the hollow particles, wherein the surface-modifying agent comprises a binding segment and a reactive segment,

wherein the binding segment comprises a silyl group, and wherein the reactive segment comprises a reactive nitrogen group;
wherein the plurality of hollow particles have an average density between about 0.05 and 0.4 g/ml; and
5 contacting the bioseparation composition with a solution comprising an analyte, wherein the surface-modifying agent is capable of coupling to the analyte.

16. The method according to claim 15, further comprising separating the bioseparation composition from the solution.
17. The method according to claim 16, wherein separating the bioseparation composition
10 from the solution comprises allowing the bioseparation composition to float to an upper surface of the solution.
18. The method according to any one of claims 15-17, wherein the hollow particles comprise glass bubbles.
19. The method according to any one of claims 15-18, wherein a size distribution of the
15 plurality of hollow particles has a span of less than 1.0.

Dated this 16 day of June 2014

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