

What is claimed is:

1. A flow cell, comprising:
a patterned substrate including depressions separated by interstitial regions;
5 sequencing surface chemistry attached to each of the depressions, the sequencing surface chemistry including:
a functionalized coating layer; and
a primer grafted to the functional coating layer; and
a hydrogel on at least the sequencing surface chemistry.
10
2. The flow cell as defined in claim 1, wherein the hydrogel is also on at least some of the interstitial regions.
3. The flow cell as defined in any of claims 1 or 2, wherein the functionalized coating
15 layer is poly(N-(5-azidoacetamidylpentyl)acrylamide-co-acrylamide).
4. The flow cell as defined in any of claims 1 through 3, wherein the hydrogel is selected from the group consisting of a poly(N-(5-azidoacetamidylpentyl) acrylamide-co-acrylamide), crosslinked polyacrylamide, an agarose gel, and crosslinked polyethylene glycol.
20
5. The flow cell as defined in any of claims 1 through 4, wherein the hydrogel is non-grafted.
6. The flow cell as defined in any of claims 1 through 5, wherein:
25 the patterned substrate includes at least one flow channel;
the depressions are defined in the at least one flow channel; and
the flow cell further comprises a spacer layer attached to other interstitial regions of the patterned substrate such that the spacer layer defines a perimeter of the at least one flow channel.
7. The flow cell as defined in claim 6, further comprising a lid attached to the spacer
30 layer.

8. A method, comprising:

applying a functionalized coating layer in depressions of a patterned flow cell substrate,
wherein the depressions are separated by interstitial regions;

5 grafting a primer to the functionalized coating layer to form a grafted functionalized
coating layer in the depressions; and

applying a hydrogel on at least the grafted functionalized coating layer.

9. The method as defined in claim 8, wherein the hydrogel is selected from the group
10 consisting of a poly(N-(5-azidoacetamidylpentyl) acrylamide-co-acrylamide), crosslinked
polyacrylamide, an agarose gel, and crosslinked polyethylene glycol.

10. The method as defined in claim 8 or 9, wherein the hydrogel is disposed on the
grafted functionalized coating layer and on some of the interstitial regions.

15

11. The method as defined in any of claims 8 through 10, wherein prior to applying the
functionalized coating layer, the method further comprises treating a surface of the patterned
flow cell substrate to attach a functional group to the surface to form treated depressions and
treated interstitial regions.

20

12. The method as defined in claim 11, wherein applying the functionalized coating layer
in the depressions includes:

applying the functionalized coating layer in the treated depressions and on the treated
interstitial regions; and

25 polishing the functionalized coating layer from the treated interstitial regions.

13. The method as defined in any of claims 8 through 12, wherein applying the hydrogel
involves applying an aqueous mixture including from about 0.001% up to about 0.1% (mass to
volume) of a hydrogel material.

30

14. The method as defined in any of claims 8 through 13, wherein a perimeter of the patterned flow cell substrate has a spacer layer bonded thereto, and wherein after the hydrogel is applied, the method further comprises bonding a lid to the spacer layer.

5 15. The method as defined in any of claims 8 through 14, wherein after the functionalized coating layer is applied and before the primer is grafted, the method further comprises bonding a lid to at least some of the interstitial regions.

10 16. The method as defined in any of claims 8, 9, and 11 through 15, wherein applying the hydrogel includes selectively depositing the hydrogel on the grafted functionalized coating layer.

17. A method, comprising:
attaching a silane or a silane derivative to a surface of a patterned substrate including a
15 flow channel having depressions defined therein, wherein the depressions are separated by interstitial regions, thereby forming silanized depressions and silanized interstitial regions;
applying a functionalized coating layer in the silanized depressions and on the silanized interstitial regions;
polishing the functionalized coating layer from the silanized interstitial regions;
20 grafting a primer to the functionalized coating layer in the silanized depressions to form a grafted functionalized coating layer in the depressions; and
applying a hydrogel on the grafted functionalized coating layer in the depressions.

18. The method as defined in claim 17, wherein applying the hydrogel involves applying
25 an aqueous mixture including from about 0.001% up to about 0.1% (mass to volume) of a hydrogel material.

19. The method as defined in claim 17 or 18, wherein the hydrogel material is selected from the group consisting of a poly(N-(5-azidoacetamidylpentyl) acrylamide-co-acrylamide),
30 crosslinked polyacrylamide, an agarose gel, and crosslinked polyethylene glycol.

20. The method as defined in any of claims 17 through 19, wherein:
a spacer layer is bonded to the patterned substrate and defines a perimeter of the flow
channel; and
after the hydrogel is applied, the method further comprises bonding a lid to the spacer
5 layer.

21. The method as defined in any of claims 17 through 20, wherein after the
functionalized coating layer is polished and before the primer is grafted, the method further
comprises bonding a lid to at least some of the interstitial regions.
10

22. The method as defined in any of claims 17 through 21, wherein applying the
hydrogel includes selectively depositing the hydrogel on the grafted functionalized coating layer
in the depressions.