Appendix I

VitroPrep™ Sample Processing Procedure

1- Vortex PreservPlus™ Solution containing specimen* for about 10 seconds.

2- Pour the specimen into a 15 mL conical centrifuge tube.

3- Centrifuge the tube for 10 min @ 1000 ×g.

4- Gently and quickly decant the supernatant liquid from the tube.

5- Depending on the cellular pellet size, add CytoBase™ Solution in a ratio of 3 parts CytoBase™ Solution to 1 part of cellular pellet, typically 250-300 µL, into the tube and vortex the tube about 20 seconds.

6- Withdraw 45 µL of the mixture (step 5) and spread gently on a standard glass slide.

7- Allow drying of the slide at room temperature for 3-4 hours, and follow standard Papanicolau staining protocol.

*For specimens containing blood, allow PreservPlus™ Solution containing specimen to sit for 24 hours for effective hemolysis.

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Appendix I

*Gynecological Papanicolau Staining Procedure

Stain Station	Reagent	Suggested Time
1	95% Ethanol	3 min
2	70% Ethanol	1 min
3	50% Ethanol	1 min
4	Distilled Water	1 min
5	Hematoxylin I	7 min
6	Distilled Water	10 sec
7	Clarifier I	1 min
8	Distilled Water	1 min
9	Bluing Solution	30 sec
10	Distilled Water	30 sec
11	50% Ethanol	30 sec
12	95% Ethanol	30 sec
13	OG-6 Solution	2 min
14	95% Ethanol	15 sec
15	95% Ethanol	15 sec
16	EA-50 Solution	4 min
17	95% Ethanol	1 min
18	95% Ethanol	1 min
19	100% Ethanol	30 sec
20	100% Ethanol	30 sec
21	100% Ethanol	30 sec
22	Xylene	1 min
23	Xylene	3 min
24	Xylene	1 min

*Please note: The Staining Procedures in Appendix I & II are only suggested staining procedures.

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Appendix II

*Non-Gynecological Papanicolau Staining Procedure

Stain Station	Reagent	Suggested Time
1	95% Ethanol	3 min
2	95% Ethanol	3 min
3	Distilled Water	30 sec
4	Hematoxylin 7211	40 sec
5	Distilled Water	90 sec
6	Clarifier I	30 sec
7	Distilled Water	1 min
8	Bluing Solution	1 min
9	Distilled Water	1 min
10	95% Ethanol	30 sec
11	OG-6 Solution	2 min
12	95% Ethanol	15 sec
13	95% Ethanol	15 sec
14	EA-50 Solution	3 min
15	95% Ethanol	15 sec
16	95% Ethanol	15 sec
17	100% Ethanol	1 min
18	100% Ethanol	1 min
19	100% Ethanol	1 min
20	Xylene	1 min
21	Xylene	1 min
22	Xylene	1 min

*Please note: The Staining Procedures in Appendix I & II are only suggested staining procedures.

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