ThinPrep® NGYN Applications introducing Cellient™ Automated Cell Block System

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Applications Specialist
ThinPrep® Technology

1 Dispersion:  

2 Cell Collection:  

3 Cell Transfer:
• Semi-automated (Approx 25 per hour)

• Processes Gyn, Non-Gyn & UroCyte samples (full range of application specific filters)
ThinPrep® 5000 Benchtop Processor

Automated small batch processor

• Walk-away automation
• Up to 20 vials per run
• GYN /NGYN or Urocyte
• Chain of custody
• Minimal maintenance
ThinPrep® 5000 benchtop components

- Filter Waste Bin
- Touch Screen interface
- Fixative Bath & Staining Rack
- Waste Bottle
- Carousel
ThinPrep® 5000 BT

Specifications

<table>
<thead>
<tr>
<th>Specification</th>
<th>Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>Instrument Dimensions</td>
<td>Width: 86 cm/34”, Height: 56 cm/22”, Depth: 66 cm/26”</td>
</tr>
<tr>
<td>Weight</td>
<td>80 kg/175 lb (approx.)</td>
</tr>
<tr>
<td>Waste container</td>
<td>Width: 15 cm/6”, Height: 43 cm/17”, Depth: 15 cm/6”</td>
</tr>
<tr>
<td>Operating Temperature</td>
<td>16 – 32°C / 60 - 90°F</td>
</tr>
<tr>
<td>Operating Humidity</td>
<td>20% – 80% RH non-condensing</td>
</tr>
<tr>
<td>Electrical Voltage</td>
<td>100/130 VAC at 2 amps 220/240 VAC at 1 amp</td>
</tr>
<tr>
<td>Frequency</td>
<td>47 – 63 Hz</td>
</tr>
<tr>
<td>Power</td>
<td>Maximum 240 watts</td>
</tr>
<tr>
<td>Clearances</td>
<td>Width: 94 cm/37”, Height: 109 cm/43”, Depth: 74 cm/29”</td>
</tr>
</tbody>
</table>
T5000 Autoloader

- Bulk slide loading door pivots upward
- Fixed center window creates continuous viewing area
- Sheetmetal waste access door
- Common vial tray door used four places
- Sheetmetal filter access door
- Cosmetic groove detail on plastic panel
- Sheetmetal side panel is integral with frame
**T5000 Autoloader**

- Large Batch processor
  - Up to 160 samples/batch
- Multiple sample types
  - NGYN, Urocyte, GYN
- Flexible
- Walk away time
- Full chain of custody
Why use ThinPrep for General Cytology?

- **Optimal preservation and Standardised**
  - less operator dependency
  - Improved Consistency
  - Improved Quality
  - Screening simplified
    - Cost saving

- **Ease of use**

- **Ancillary testing**
Applications in Non-Gyn Cytology

- Body fluids
  - Pleural effusion
  - Peritoneal effusion
  - Pericardial effusion

- Respiratory tract
  - Sputum
  - Bronchial brushing, washing
  - Bronchoalveolar lavage

- Urines
  - Voided urines
  - Bladder washing
  - Ileal conduit

- Digestive tract
  - Esophageal, colonic brushing
  - Anal cytology

- FNAs
  - Thyroid
  - Breast
  - Pancreas.... Lymph Nodes

- Cytology
  - Cells Block
  - DNA ploidy
  - Immunocytochemistry
  - ISH
  - FISH
  - Cytogenetic
  - Special stains
Required Materials List

- ThinPrep® Microscope Slides
- Non-Gyn TransCyt™ Filters (Blue)
- Multi-Mix™ Racked Vortexor
- CytoLyt® and PreservCyt® Solutions
- 50 ml capacity centrifuge
- 50 ml centrifuge tubes
- Slide staining system and reagents
- 1 ml plastic transfer pipettes, graduated
- Standard laboratory fixative
- Coverslips and mounting media
- Glacial acetic acid, DTT
BODY FLUIDS
(Serous Effusions, Urinary and Cerebrospinal Fluids)

1) COLLECTION
Collect body fluids fresh.
Fluids collected in Cytolyt® Solution require a Cytolyt® Solution wash prior to processing.
For extremely bloody fluids, start with only 10ml of fresh fluid.

2) CONCENTRATE BY CENTRIFUGATION
(600g for 10 minutes)

3) POUR OFF SUPERNATANT AND RESUSPEND CELL PELLET
Resuspension can be done on a vortexor or may be achieved by syringing the pellet back and forth with a plastic pipette.

4) CYTOLYT® SOLUTION WASH
- Add 30ml Cytolyt® solution
- Centrifuge
- Pour off supernatant
- Resuspend cell pellet

5) EVALUATE CELL PELLET APPEARANCE
If cell pellet is not free of blood, repeat step 4.

6) ADD SPECIMEN TO PRESERVICYT® SOLUTION VIAL
Allow to stand in PreservCyt® Solution for 15 minutes.

7) RUN ON THINPREP® PROCESSOR USING SEQUENCE 2 (FLU/FNA)
(Fix, Stain and Evaluate)

MUCOID SPECIMENS
(Respiratory and Gastrointestinal specimens)

1) COLLECTION
Sputum: Collect sample directly into Cytolyt® Solution
Brushings: Deposit the collection brush directly into a pre-filled Cytolyt® Solution tube
Washings/Lavage: Collect sample with a balanced electrolyte solution (BES). In the laboratory, the BES is replaced with 30ml of Cytolyt® Solution.
The use of Distilled Water (DW) has shown to reduce the amount of mucoid present in respiratory samples. 2.
To use DW with the ThinPrep® system, prepare a stock solution by adding 2.5g DTT to 100ml of Cytolyt® Solution. This solution is suitable for use for 1 week when stored at room temperature (15-30°C). Add 1ml of stock solution to the sample.

2) MECHANICAL AGITATION
Place 50ml specimen tubes on the Multi-Mix™ Racked Vortexor for 10 minutes or blend the Cytolyt®/Sample mixture for a few seconds.

3) CONCENTRATE BY CENTRIFUGATION
(600g for 10 Minutes)

4) POUR OFF SUPERNATANT AND RESUSPEND CELL PELLET
Resuspension can be done on a vortexor or may be achieved by syringing the pellet back and forth with a plastic pipette.

5) EVALUATE CELL PELLET APPEARANCE
Confirm the cell pellet is in liquid form. If not, add 30ml of Cytolyt® Solution and repeat step 2, 3 and 4.

6) ADD SPECIMEN TO PRESERVICYT® SOLUTION VIAL
Allow to stand in PreservCyt® Solution for 15 minutes.

7) RUN ON THINPREP® PROCESSOR USING SEQUENCE 3 (MUCOID)
(Fix, Stain and Evaluate)
FINE NEEDLE ASPIRATES (FNA)
(Serous Effusions, Urinary and Cerebrospinal Fluids)

1) COLLECTION
Collect sample directly into 30ml of CytoLyt® Solution
CytoLyt® Solution is an alcohol based solution. If a fresh,
non-alcoholic fixed specimen is clinically indicated, collect
the specimen into a balanced electrolyte solution (BES). In
the laboratory, the BES is replaced with 30ml of CytoLyt® Solution.

2) CONCENTRATE BY CENTRIFUGATION
(600g for 10 Minutes)

3) POUR OFF SUPERNATANT AND RESUSPEND CELL PELLET
Resuspension can be done on a vortexor or may be achieved
by syringing the pellet back and forth with a plastic pipette.

4) EVALUATE CELL PELLET APPEARANCE
If cell pellet is not free of blood, add 30ml of CytoLyt® Solution
and repeat step 2.

5) ADD SPECIMEN TO PRESERVCYT® SOLUTION VIAL
Allow to stand in PreservCyt® Solution for 15 minutes.

6) RUN ON THINPREP® PROCESSOR USING SEQUENCE 2 (FLU/FNA)
(Fix, Stain and Evaluate)

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ThinPrep® UroCyte™
(For use with Urine Cytology Processing and/or Slide-based Molecular Testing)

1) Collection
Collect urine directly into the ThinPrep™ UroCyte™ Urine Collection Kit,
OR process urine fresh. (Note: Fresh urine can be mixed with a 2:1 urine
to PreservCyt® ratio and stored for up to 48 hours before processing.)

2) Concentrate by Centrifugation
(600g for 10 Minutes)

3) Pour Off Supernatant and Resuspend Cell Pellet
Resuspension can be done on a vortexor or may be achieved by syringing
the pellet back and forth with a plastic pipette.

4) Add CytoLyt® Solution Wash
• Add 30ml of CytoLyt® Solution
• Centrifuge
• Pour Off Supernatant
• Resuspend Cell Pellet

5) Evaluate Cell Pellet Appearance
If cell pellet is not free of blood, add 30ml of CytoLyt® and repeat from step 2.

6) Add Specimen to PreservCyt® Solution Vial
Allow to stand in PreservCyt® Solution for 15 minutes.

7) Run on ThinPrep® Processor using Sequence 5 (UroCyte™)
(Fix, stain, and evaluate cytology or perform molecular diagnostic testing
according to the manufacturer's instructions for use.)

Refer to the ThinPrep® Processor Operator's Manual for detailed instrument and
specimen preparation instructions. See section D and E for details.
Achieving optimal results in Non-Gyn ThinPrep® Processing
TPNG Shortcuts: Where do you end up?
TPNG Shortcuts: Where do you end up?

- The outcome if you take shortcuts on TPNG prep
- The reasons for following ThinPrep NGYN protocols
- Review of a case study
Problem- Customer complained T2000 processed NGYN Serous Fluid Preparations were better than T5000 NGYN preparations.

T2000

T5000

Please note no difference if adhere to correct Hologic NGYN protocols!
A. Processed following protocol correctly

B. Received in Cytolyt and did not carry out a wash step
Microscopic Result

A. Correct cell distribution

B. Scanty uneven cell distribution
Macro comparison of sample following decanting of supernatant

A. Correct decant

B. Incorrect decant
Microscopic Result following decanting of supernatant

A. Correct cell distribution

B. Scanty and uneven cellular distribution
Cytolyt Wash
(especially applicable for bloodstained samples)

A. Cytolyt wash performed as suggested
B. No cytolyt wash done
Microscopic Result

A. Correct Cell distribution

B. Scanty and uneven cell distribution
Correct Vol vs Too much specimen added to Vial

A. Correct amount

B. Incorrect amount
Microscopic Result

A. Correct volume added

B. Too much specimen added to vial
ThinPrep™ Sample
NGYN BW
Adenoca lung
Non-Keratinising Sq Cell Ca - BW

13/03/2014
Nigel Boucher
CAS/AE
Hologic
Small cell anaplastic Ca-BW
PL. FL Small Cell Ca
RS cells Hodgkin's Lymphoma PF
Carcinosarcoma of Lung

Sarcomatous element
Adenoca element
Grade 1 TCC (x60)

- Apoptosis
- Mitosis
Grade 2 TCC (x20)
TCC high Grade
Adenoca Bladder 1°
Urine SCC (x40)

Note intercellular bridges
Renal Casts

Note blood in cast indicating tubular damage
Strongyloides stercoralis
cellient™ AUTOMATED CELL BLOCK SYSTEM
Importance & role of cell blocks

Is it cancer?
Where did it originate?
What type is it?

Cytology

Cell Blocks

Histology
• **Improved capture**
  – Vacuum assisted
  – Based on proven ThinPrep® technology

• **Improved presentation**
  – produces an even distribution of cells in block

• **Improved consistency**
  – Fully automated
  – Rapid Processing (30-45mins)
  – Less cross contamination
After embedding
Example of Traditional Method Versus Cellient™ Automated Cell Block System

Traditional Cell Block

Cellient™ System

Pleural Fluid Positive For Carcinoma
Reactive changes in pleural effusion

AGAR

Cellient
Cellient, HE

Sq Cell Ca- Lung
Pleural effusion

Giemsa

Cellient, HE

Metastatic Adenoca- Breast
2 case studies
Case 1- Ascitic Fluid

- Female
- 52
- Abdominal pain, Large Ascites
Ascites, Giemsa / PAP
PAS

PAS-dias
BerEp4  CK 7
WT-1
Conclusion

• Clear cell adenocarcinoma of the ovary

• By using additional immunocytochemistry, a more specific diagnosis can be given:

• This improves service delivery.
Peritoneal fluid – Case 2

- Female

- 56 years

- Suspicious of a Malignant Ovarian Tumor
Serous Papillary Ca Ovary
CK7 positive – Ovarian Ca
WT1 Positive- Ovarian Ca
Results

• Morphology: adenocarcinoma

• Immunocytochemistry: CK 7, WT1 positive

• CK 20, TTF-1 negative

• Conclusion: adenocarcinoma of the ovary.
Morphology conclusion

• Cellient gives a more clear staining pattern

• improved nucleus/cell detail

• More cells to diagnose
Cellient™ Tips
Vial to Block

Following protocol and good practice is essential to achieve optimum cell blocks from the Cellient processor.

*ThinPrep® 2000/5000 Processor*

*Operator’s Manual*
The Cellient™ Processor operators manual

Table 1.1: Sample Quantity

<table>
<thead>
<tr>
<th>Centrifuge Tube Quantity</th>
<th>PreservCyt® Solution Vial</th>
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</table>

Quantity Process

- Centrifuge tube empty Vial with residual sample Run vial on the Cellient System.

- Tube with cell pellet Vial empty Put a maximum of 10 drops (approximately ¼ ml) of cell pellet into a vial containing 20 ml of PreservCyt Solution.

- Tube with cell pellet Vial with residual sample Put a maximum of 10 drops (approximately ¼ ml) of cell pellet into a vial containing 20 ml of PreservCyt Solution.

- Top off vial with PreservCyt Solution if needed.

- Tube empty Vial empty Sample insufficient for cell block.
Clinical Study

Abstract

*Diagn Cytopathol.* 2013 Feb 27. [Epub ahead of print]

**Effective application of the methanol-based PreservCyt(™) fixative and the cellient(™) automated cell block processor to diagnostic cytopathology, immunocytochemistry, and molecular biology.**

*van Hemel BM, Suurmeijer AJ.*

Source

*Department of Pathology and Medical Biology, Cytology Laboratory, University Medical Center Groningen, University of Groningen, The Netherlands.*
Hologic® offers The Complete Solution for Cervical Health Screening

1. Sample Collection
2. ThinPrep® Cytology Processing
3. ThinPrep® Staining
4. ThinPrep® Imaging with Dual Screening
5. Molecular Testing

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www.cytologystuff.com
www.cellentsystem.com
www.cervistahpv.com
Thank you