



# Guide to Operations

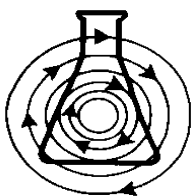
## **NucleoCounter<sup>®</sup> YC-100<sup>™</sup>** **Automatic Yeast Cell Counter** (software version 3.00 or higher)

MANUAL No. M1293-0055

Revision A

March 28, 2005

For a printed copy of this manual, please contact NBS.



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***CAUTION!***

This equipment *must* be operated as described in this manual. If operational guidelines are not followed, equipment damage and personal injury *can* occur. Please read the entire User's Guide before attempting to use this unit.

## Disclaimer Notices

New Brunswick Scientific Co., Inc. reserves the right to change information in this document without notice. Updates to information in this document reflect our commitment to continuing product development and improvement.

The information contained herein is to the best of our knowledge accurate and complete. However, cell species and cell environments may vary in property, therefore systematic and/or random deviation between estimates obtained by *NucleoCounter*<sup>®</sup> and other cell counting methods may occur. As such, nothing contained or stated herein, including results obtained from the use of *NucleoCounter*<sup>®</sup> and/or *NucleoCassette*<sup>™</sup>, shall be construed to imply any warranty or guarantee. New Brunswick Scientific (NBS) and affiliated companies shall not be held liable for damages, and customers shall indemnify NBS and affiliated companies against liability from use of potentially inaccurate data generated by *NucleoCounter*<sup>®</sup>. We recommend that all results obtained with *NucleoCounter*<sup>®</sup> be validated against appropriate reference methods and/or traditional laboratory methods at regular intervals.

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*NucleoCounter*<sup>®</sup>, *YC-100*<sup>™</sup>, *Reagent Y100*<sup>™</sup>, *NucleoCassette*<sup>™</sup> and *NucleoView*<sup>™</sup> are trademarks of and are manufactured by ChemoMetec A/S, Gydevang 43, DK-3450 Allerød, Denmark.

## Manual Conventions

### Bold

Text in bold face type emphasizes key words or phrases.

### NOTE:

Notes contain essential information that deserves special attention.



### CAUTION!

*Caution* messages appear before procedures which, if caution is not observed, could result in personal injury or damage to the equipment.



### ELECTRICAL HAZARD!


*Electrical Hazard* messages alert you to specific procedures or practices which, if not followed correctly, could result in serious personal injury from electric shock.



## WARRANTY

Every New Brunswick Scientific instrument is warranted to be free from defects in material and workmanship. This apparatus with the exception of glassware, lamps and electrodes (where supplied), is warranted for 1 year against faulty components and assembly and our obligation under this warranty is limited to repairing or replacing the instrument or part thereof, which shall, within one year after date of shipment, prove to be defective after our examination. This warranty does not extend to any NBS products that have been subjected to misuse, neglect, accident or improper installation or application; nor shall it extend to products that have been repaired or altered outside the NBS factory without prior authorization from the New Brunswick Scientific Co., Inc.

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# 1 OVERVIEW

*NucleoCounter<sup>®</sup> is based on a patented technology for the counting of yeast cells in suspension. Compared to a variety of other methods, the most important difference is the ability to stain DNA and count the individual cells containing DNA without having to handle hazardous dyes. The sample pre-treatment not only allows effective staining of cellular DNA, but it will also help to dissolve cell aggregates.*

## 1.1 **Symbol**

Wherever the  symbol appears on the *NucleoCounter* instrument, it is an indication that the manual needs to be consulted for precautions and/or warnings.

## 1.2 **Intended Use**

The *NucleoCounter<sup>®</sup> YC-100<sup>™</sup>* system is comprised of the *NucleoCounter<sup>®</sup>* automatic cell counter, the *NucleoCassette<sup>™</sup>*, a cell preparation lysing buffer (Reagent Y100<sup>™</sup>), and *NucleoView<sup>™</sup>* software for use with an optional computer interface.

The *NucleoCounter* system enables the user to determine yeast cell concentration in samples containing dispersed yeast cells in suspension. The cell counts rest on the ability of the fluorescent dye (propidium iodide) to bind with the yeast cells' DNA.

The *NucleoCounter* system provides fast, efficient and reliable cell counts. It is intended for research only, not for human or veterinary diagnostic use.

## 1.3 **Measurement Volume**

One key benefit of the *NucleoCounter* system is the use of a disposable sample device called the *NucleoCassette<sup>™</sup>*. This small plastic apparatus is preloaded with a DNA-staining dye.

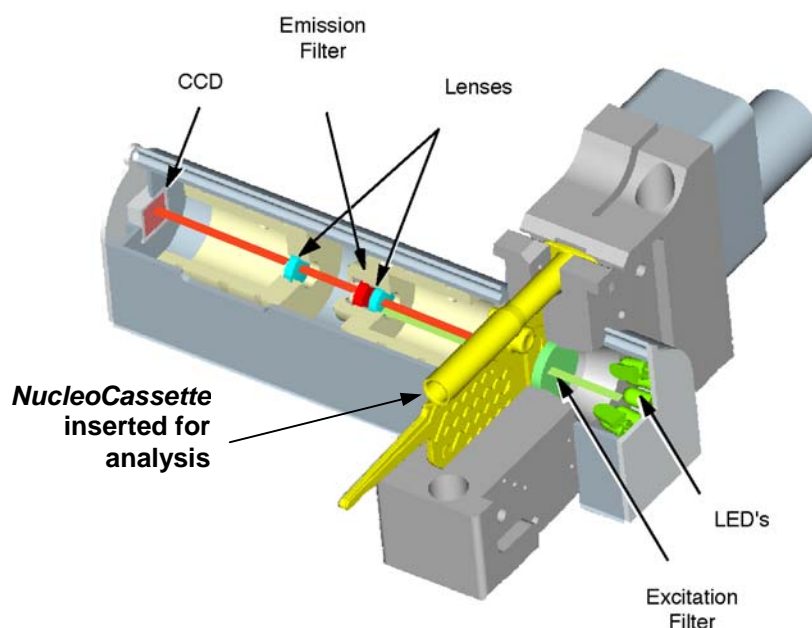
The measurement volume, which is the sample volume analyzed for yeast cells, is approximately 1 µl. This is 10-20 times the volume usually sampled by conventional hemacytometers.

## 1.4 Fluorescence Microscope

The compact fluorescence microscope (see Figure 1) integrated into *NucleoCounter* comprises LEDs (light-emitting diodes) as the excitation light source, optics (lenses), excitation and emission filters, and a CCD (charged coupled device) camera. *NucleoCounter* also contains advanced software for image processing.

While it is true that this principle is used in several other methods of cell counting, in *NucleoCounter* it is used in combination with novel technologies and techniques that offer substantial advantages.

**Figure 1: Fluorescence Microscope**



This is how the fluorescence microscope system works:

1. When the sample is brought into the *NucleoCassette*'s measurement chamber, it is illuminated by green excitation light emitted by the LEDs.
2. This green light is passed through the excitation filter before it reaches the chamber; the filter allows only green light of pre-determined wavelengths to pass, in order to excite the propidium iodide (PI) dye.
3. The green light reaches the measurement chamber, excites the dye that is bound to the cell DNA.
4. In return, the PI dye emits red fluorescent light.
5. Not all of the green light is absorbed by the dye; after passing through the chamber, the remainder of the green light, plus the red light, is focused through an optical lens and passed through the emission filter.
6. The emission filter allows only the red light emitted by the PI to pass; the green light is absorbed.

7. The red light is then focused through another optical lens before the CCD camera detects it.
8. When the red light reaches the CCD camera, the photons are transformed into electrical signals.
9. An image of the measurement chamber is formed from these electrical signals (see Figure 2). Each dot in the image represents DNA stained with PI. If a computer is connected and running *NucleoView*, this is the image shown by *NucleoView* each time an analysis is run.

**Figure 2: Image of Cell DNA**



#### **1.4.1 Rapid Analysis**

One obvious advantage of the *NucleoCounter* is the relatively large volume of sample that is analyzed for particles at one time. In addition, despite the size of the volume, the analysis is performed rapidly and accurately, allowing the entire analysis of the sample to be completed in less than one minute.

#### **1.4.2 Cell Identification**

Because the cell's DNA is stained with a dye that is recognized by the fluorescence microscope, the system does not rely on any particular morphological feature of cells in order to identify them for counting.

### **1.5 Data Logging**

*NucleoCounter* can operate both as a stand-alone cell counter and connected to a PC through a USB interface.

Working as a stand-alone, the instrument shows results in the instrument display.

When connected to a computer that is running *NucleoView*<sup>™</sup> application software, the instrument transfers the collected image as well as the calculated result to the PC for data entry, where the information will be available for later use. *See NucleoView user's guide for details.*

---

## 2 INSPECTION & UNPACKING OF EQUIPMENT

### 2.1 *Inspection of Boxes*

After you have received your order from New Brunswick Scientific, inspect the box(es) carefully for any damage that may have occurred during shipping. Report any damage to the carrier and to your local NBS Sales Order Department immediately.

### 2.2 *Packing List Verification*

Unpack your order, saving the packing materials for possible future use. Also be sure to save the User's Guide, for instruction and reference.

Verify against your NBS packing list that you have received the correct materials, and that nothing is missing.



#### **NOTE:**

**The CD-ROM packed with your instrument has the software needed to install the *NucleoCounter* utilities if you plan to use the optional *NucleoView* software in connection with a PC.**

If any part of your order was damaged during shipping or is missing, or fails to operate, please fill out Customer Satisfaction Form 6300 (located in the envelope that contains the Warranty Card) and return it by fax or mail. You can also call the New Brunswick Scientific Service Department.

## 3 INSTALLATION

### 3.1 *Physical Location*

*NucleoCounter* is a small, lightweight instrument that can be used on any benchtop, desk or counter. The unit is for indoor use only.

**Figure 3: Dimensions**



### 3.2 *Installing the NucleoCounter*

Remove the protective plastic wrap and save it with the packing materials.

### 3.3 *Power Supply*

*NucleoCounter* is powered via a detachable 12VDC power supply cord, provided with the instrument, that connects the cell counter to the electrical supply. The instrument automatically detects the input voltage; no manual voltage selection is required.

## 3.3.1

## Electrical Warnings &amp; Cautions

**ELECTRICAL HAZARD!**

Make sure that your power supply outlet is grounded. Verify that the power supply cord shipped with your *NucleoCounter* is appropriate to your power outlet and supply voltage.



*NucleoCounter* may **only** be used with one of the following external power supplies:

Power Supply Class	I	I	II
Manufacturer	Proton Electronic Industrial Co., Ltd.	Powerbox Europe AB	Celetron USA Inc.
Model	SPN-260-12	EBH 03 131	ZVC40LT12E
Input Rating	100-240 VAC 50-60 Hz, 1.6A	100-240 VAC 47-63 Hz, 0.8A	100-240 VDC 50-60 Hz, 1.2A
Output Rating	12 VDC, 4.3A	11-13 VDC, 2.7A	12 VDC, 3.3A



The detachable power supply cord, provided with the instrument, and the appliance inlet of the external power supply are considered to be the disconnecting device.



The power supply cord and its plugs comply with any applicable national regulations.



If the user attempts to utilize the *NucleoCounter* and/or the external power supply in a manner not specified by the manufacturer, the protection provided by the *NucleoCounter* and the external power supply may be impaired.

## 3.3.2

## Making Power Connection

Once you have verified that the power supply cord's bladed plug is appropriate for your electrical outlet, connect the "phone jack" end of the 12 VDC power supply cord to the DC power socket on the rear panel of the instrument (*see Figure 4 on the following page*).

**Figure 4: Power Supply Cord Connection (Rear Panel)**



When you are ready to use *NucleoCounter*, plug the bladed end of the power supply cord into your electrical outlet.

### **3.4 Electromagnetic Interference**

#### **NOTE:**

**This equipment has been tested and found to comply with the limits for a Class B digital device, pursuant to Part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference in a residential installation.**

**This equipment generates, uses and can radiate radio frequency energy and, *if not installed and used in accordance with the instructions contained in this manual*, may cause interference harmful to radio communications.**

Even with proper installation and operation, there is no guarantee that interference will not occur at a particular location. If this equipment does cause harmful interference to radio or television reception, which can be determined by turning the equipment on and off and monitoring the results, we encourage you to try to correct the interference by implementing one or more of the following measures:

- Reorient or relocate the receiving antenna of the radio and/or television.
- Increase the distance that separates *NucleoCounter* from the receiver.
- Connect *NucleoCounter* to an outlet that is on a different circuit than the outlet to which the receiver is connected.
- Consult the dealer or an experienced radio/TV technician for help.

**NOTE:**

Changes or modifications not expressly approved by the party responsible for compliance could void the user's authority to operate the equipment.

### 3.5 USB Cable Cautions

On the left side of your *NucleoCounter*'s rear panel is a USB connector. This is only to be used to communicate with a PC via the shielded USB cable supplied with the *NucleoCounter* to ensure that you maintain the appropriate EMI classification for the intended environment.



The *NucleoCounter* USB interface connector must be connected **only** to SELV circuits. External computing devices connected to the *NucleoCounter* USB interface connector must comply with UL 1950 and IEC/EN 60950 standards.



The following table lists essential information for the *NucleoCounter* USB interface connector:

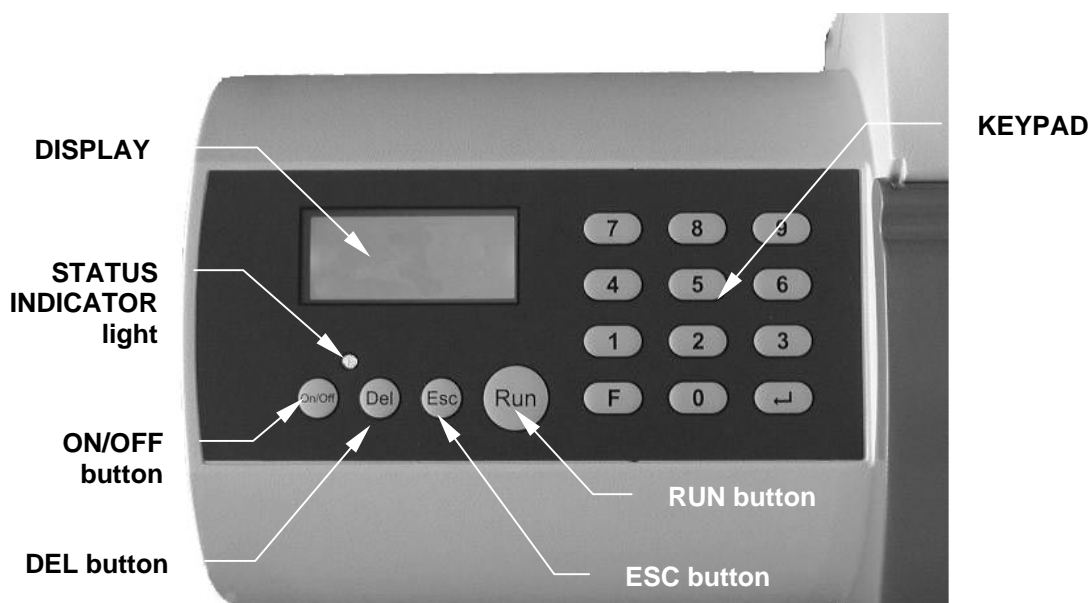
Pin Number	Designation	Maximum Voltage Level*
1	+5V	+5 VDC
2	D-	+3.5 VDC
3	D+	+3.5 VDC
4	DGND	0 VDC
Metal enclosure	Shield (connected to DGND)	0 VDC

\* in normal operation (refers to Pin number 4).

Consult the *NucleoView* software user's guide for further information regarding the use of a PC in communication with *NucleoCounter*.

## 4 OPERATING CONTROLS & DISPLAYS

Figure 5: Keyboard Panel



### 4.1 Interactive Controls

#### 4.1.1 Control Buttons

There are four control buttons on the front of the unit (*see Figure 5*). Only two, **ON/OFF** and **RUN**, will be needed for everyday operation:

<b>On/Off</b>	Use this button to turn the instrument on or off.
<b>Run</b>	Use this button to start measuring.
<b>Esc</b>	<i>Reserved for special use.</i>
<b>Del</b>	<i>Reserved for special use.</i>

The other two are reserved for special use. Do not press either of them unless so instructed by this manual.

#### 4.1.2 Keypad

There is a keypad (*see Figure 5*) comprised of 12 keys: **0 – 9**, **F** and **↵** (“Enter”). You will rarely have occasion to use the keypad.

## **4.2      *Interface Displays***

A small Status Indicator light above the **ON/OFF** button (*see Figure 5*) illuminates when the unit is on. It is green when the instrument is ready to work, red while it is measuring, and green again when the results are displayed.

The Display screen communicates information to the user. It indicates, at various times: unit identification (software version and unit serial number), start-up status, operating functions, cell counts, and shutdown status.

## 5 GETTING STARTED



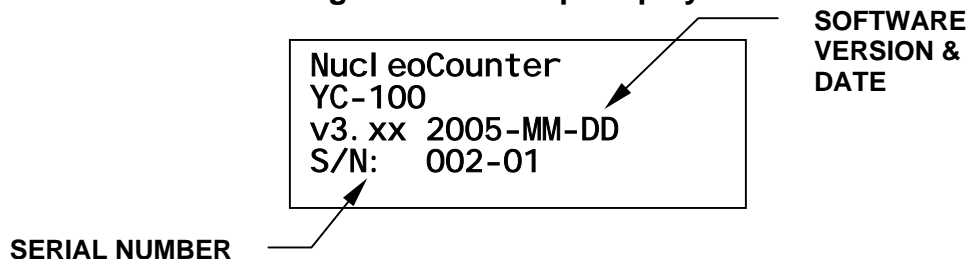
### **CAUTION!**

During start-up, *NucleoCounter* performs a self-check. Never insert a cassette prior to, or during, start-up.

### 5.1 Power On

1. Turn the instrument on by pressing the **ON/OFF** button.
2. During start-up, you will hear a brief noise as the motor turns on. This is perfectly normal.
3. While the instrument is starting up, the display should look like this:

**Figure 6: Start-Up Display**



### **NOTE:**

The start-up display shows the version number (“v3.xx” in *Figure 6*) and date (“2005-MM-DD” in *Figure 6*) of the integral software and the serial number of your instrument (“002-01” in *Figure 6*).

**Record this information for future reference. You will need to provide the serial number whenever you contact New Brunswick Scientific for assistance.**

4. When the unit has started successfully, the display should read: **NucleoCounter Ready** and the green status indicator light should be illuminated.
5. To shut down the instrument, press the **ON/OFF** button. While *NucleoCounter* shuts down, the display will momentarily read: **Bye...**

6. If the instrument has performed as described above, it is ready for use. If the instrument displays an error message at start-up, *see Section 8*.

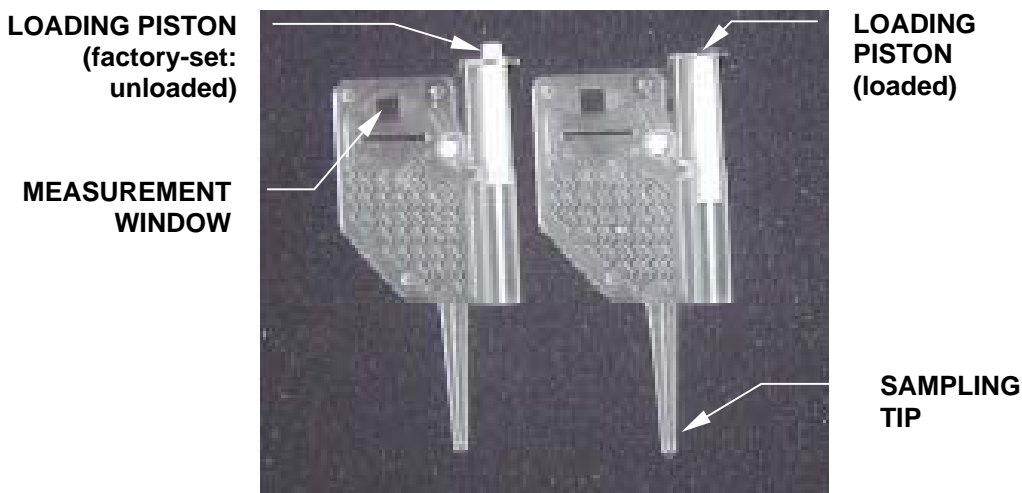
## 5.2 **Installation of NucleoView™**

If you plan to use the optional *NucleoView* management software, install it at this time. Please refer to your *NucleoView* User's Guide for installation instructions.

## 5.3 **About NucleoCassette™**

The *NucleoCassette*™ is the easy-to-use component that stains the cell DNA with propidium iodide and collects the fluorescent image. The cassette is a small, custom-made, disposable plastic device that was designed for optimal sample handling characteristics and easy, safe disposal.

**Figure 7: NucleoCassette**



The tapered tip of the cassette is used to easily remove a sample from a small sample vial. You can load a volume of approximately 50 µl into the cassette by pressing the white loading piston while you immerse the tip into the sample (*see Section 5.5, Loading a Sample*).

### 5.3.1 **Fluorescent Dye**

The cassette is preloaded with a dried form of fluorescent DNA-staining dye. This propidium iodide is immobilized in the first portion of the flow system, ready to dissolve immediately into and mix with the sample as the cell suspension is loaded into the cassette.

**CAUTION!**

**Propidium iodide, the staining dye used in the *NucleoCassette* may be mutagenic and/or carcinogenic. Never attempt to reuse the cassette or to empty it once you have loaded it.**

**If a cassette becomes damaged or begins to leak, any spill should be handled as potentially hazardous waste.**

Due to the fluorescent nature of the dye, it absorbs green light, then emits red light, which *NucleoCounter* uses to detect the stained cells. In fact, the ability to fluoresce is significantly enhanced when the dye binds to the cell DNA, substantially enhancing selectivity.

During the analysis, the stained mixture is transported through the flow system, from the area where it was mixed with the PI toward the chamber where the actual cell counting is performed. The precise volume analyzed is determined individually for each cassette during the analysis. This volume is approximately 1 $\mu$ , which is 10-20 times that of a conventional hemacytometer.

### 5.3.2 Loading Piston

In production, the cassette-loading piston is adjusted to a position that provides optimal loading of the cassette when you press the piston down slowly and gently with your thumb, until its top is flush to the surface (*see Figure 7*).

As the sample is aspirated into the cassette (*see Section 5.5 for instructions*), the sample dissolves the dry staining dye within a few seconds. The time it takes to install the cassette in the instrument is usually more than adequate to dissolve and mix with the dye.

### 5.3.3 Sample Volume

As it is manufactured, each cassette is marked with a dot code that specifies the precise depth or thickness of the measurement chamber of that cassette. *NucleoCounter* reads and deciphers the dot code during analysis. The volume analyzed is determined by multiplying the imaged area by the depth (thickness) of the cassette's measurement chamber.

### 5.3.4 Cassette Handling

The *NucleoCounter*'s built-in image analysis method offers considerable stability. It eliminates non-cellular objects from the image, including any scratches or smears on the *NuceloCassette* windows. In this way, you are assured that the results are valid even under extreme conditions. Nevertheless, the quality of the cell count can be optimized by keeping the windows of the cassette as clean as possible.

#### NOTE:

**To maximize the quality of the cell count, do not touch the cassette windows. Should you need to wipe the surface of the cassette for any reason, be sure to use a clean, soft cloth to avoid scratching the plastic windows.**

After analysis, remove and dispose of the *NucleoCassette*, which still contains the entire sample, as biological waste.

### 5.4 Sample Preparation: Lysing

Reagent Y100, the lysis buffer for yeast cells available for use with your *NucleoCounter*, is suitable for the sample preparation of most yeast cell suspensions. It is highly effective and generally achieves instantaneous lysing of yeast cells in suspension. There are, however, certain species of yeast cells that might require a different preparation procedure. If you encounter problems relating to insufficient lysing of your yeast samples, please contact NBS for assistance.

1. Using a pipette, collect a precise volume of thoroughly mixed sample. We recommend that you collect at least 100  $\mu\text{l}$  of the cell suspension, but throughout this manual, a sample size of **200  $\mu\text{l}$**  is used. It is very important that the sample be representative of the cell suspension.
2. Place the sample in a convenient sample vial for treatment (*see Figure 8*)



#### **CAUTION!**

- **Never pipette by mouth.**
- **Avoid specimen contact with skin or mucous membranes.**
- **Avoid cross-contamination of the samples during preparation, as this can compromise the quality of the measurement results.**
- **Handle any biological specimen as though it were capable of transmitting infection, and dispose of it with proper precautions according to your federal, state and local regulations.**

**Figure 8: Pipetting Sample**

3. Using the same pipette (but with a new tip) to avoid dilution, add 200  $\mu$ l of lysing buffer to the cell sample. Alternatively, dispense the buffer using the available reagent-dispensing pump.

 **NOTE:**

If you are not using reagent-dispensing pumps, place an appropriate amount of buffer in a temporary container from which you will draw the needed volume. In this way you will protect the source reagent from contamination.

 **NOTE:**

Be sure to validate, at regular intervals, the precision and accuracy of any dispenser and/or pipette used to dispense the buffer.

4. Mix the sample and buffer thoroughly for a few seconds, by turning the carefully closed vial upside down 5-10 times, or using the pipette, or mixing the closed vial on a vortex mixer.

**CAUTION!**

If you mix the sample in a vial fitted with a lid, open the lid very carefully: the surfactant in the buffer may cause a liquid film to form across the vial opening, just under the lid. Opening the lid could cause the film to burst, spattering droplets into the air.

Take care also to gently break the film if it remains, to avoid contaminating your pipette or dispenser as you introduce it into the vial.

 **NOTE:**

The reagent/cell mixture may be stored at room temperature or refrigerated for some time before analysis, depending on the yeast type.

**NOTE:**

Remember to mix the sample well (as explained in Step 4) before aspirating any volume or loading the *NucleoCassette*.

5. Load the mixture, called the lyzate, into the *NucleoCassette* (see Section 5.5 below).

## 5.5 Loading a Sample

**CAUTION!**

- Never pipette by mouth.
- Avoid specimen contact with skin or mucous membranes.
- Avoid cross-contamination of the samples during preparation, as this can compromise the quality of the measurement results.
- Handle any biological specimen as though it were capable of transmitting infection, and dispose of it with proper precautions according to your federal, state and local regulations.

To load the *NucleoCassette*:

1. Take care to fully immerse the loading tip into the sample, to avoid air bubbles in the flow system.
2. Press the loading piston gently with your thumb, until your thumb and the top of the piston are flush to the surface of the cassette.

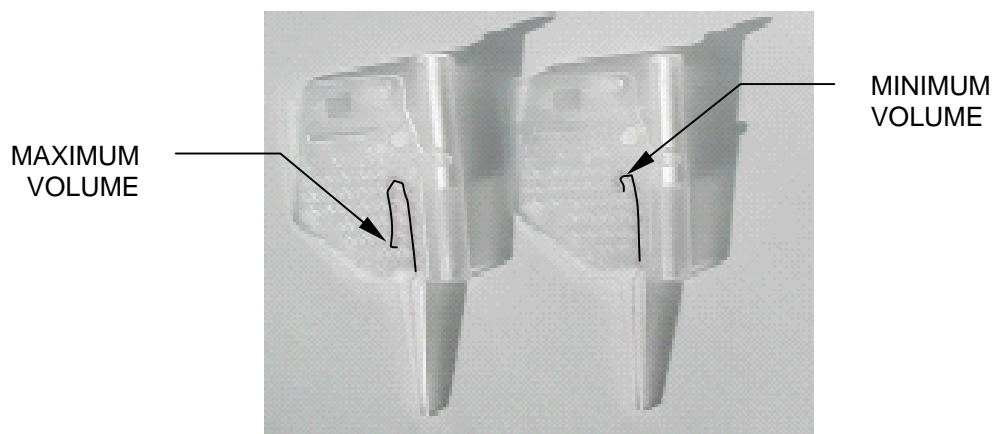
**Figure 9: Loading the Cassette**



3. After loading, release the piston (it will remain in place). Make sure no droplets are attached to the tip.
4. Inspect the cassette to make sure it contains no large air bubbles.

Although the final concentration of the dye is not critical to a successful operation, it is important to load an appropriate volume of sample into the cassette. Inspect the cassette after loading to assure that the volume is adequate. Figure 10 shows a minimum and a maximum load, with the sample colored for the sake of illustration.

**Figure 10: Cassette Min/Max Volume**



## 6 OPERATION

### 6.1 *Ready Mode*

After you turn the *NucleoCounter* on by pressing the **ON/OFF** button, the display will indicate that the instrument is ready for cell counting when it reads **NucleoCounter Ready**. The status indicator light will also be green.

The instrument is ready to receive the sample and begin counting.



#### **NOTE:**

**The same message (“NucleoCounter Ready”) will also be displayed if the ESC button is activated at any time in a run other than measuring or shutdown. In association with the ESC button, however, the status indicator light will be red, not green.**

### 6.2 *Inserting the Cassette*

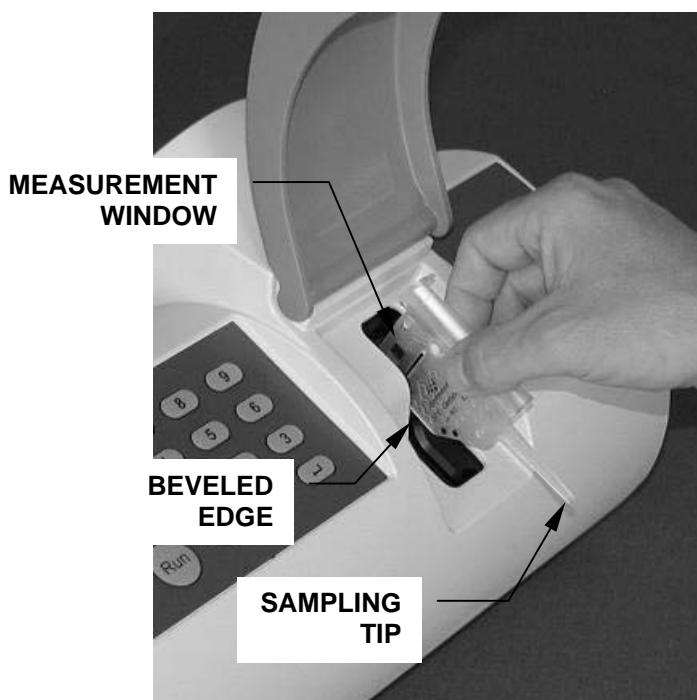
When the cassette has been properly loaded with the lyzate mixture, as explained in the previous chapter, you are ready to load it into the *NucleoCounter* instrument.

The sample compartment lid (*see Figure 11*) covers the sample insertion area. This lid is designed to protect the area from dust and other potential contaminants. It also keeps external light from interfering with the fluorescent image recording and the measurement analysis. The lid is fitted with magnetic hinges, so it can be easily removed for cleaning purposes.

Unless you are inserting or removing a cassette or cleaning the insertion area, keep the lid closed at all times.

To insert the cassette:

1. Flip the lid up, the way you open a cell phone.
2. Grip the cassette on either side of the piston barrel, but taking care not to touch the measurement windows.
3. Insert the cassette, beveled edge down, into the insertion slit (*see Figure 12*). Be sure to keep the sampling tip parallel to your hand, pointing toward your wrist.

**Figure 11: Sample Compartment Lid****Figure 12: Cassette Insertion**

4. Press the cassette down until you hear a click. The tip will lay parallel to the opening, along the body of the instrument.
5. Close the lid gently.

### **6.3 Measuring**

Once the cassette has been loaded with lyzate and inserted into the *NucleoCounter*, activate measurement by pressing the **Run** button.

When you press **Run**, the actuator motor moves the piston rod to release the sample/dye mixture and send it flowing toward the cassette measurement chamber. The form and dimensions of the flow channels facilitate effective mixing of sample and DNA-staining dye. Sensors monitor sample flow; when the sample has been loaded into the image chamber, the piston is stopped.

The sample is allowed to settle for a few seconds, then the fluorescent image is recorded. Once recorded, it is analyzed by the *NucleoCounter*'s integral software. Upon completion of the analysis, the result is shown on the display (see *Figure 13*, which presents the lyzate cell density in Mode 3—see Section 6.4 for an explanation of the available modes). The entire process takes approximately 30 seconds.

**Figure 13: Sample Analysis Result Display**

Cell s/ml :

1.234 x 10E6

If an external computer is connected, the results and the recorded fluorescent image can be viewed there using *NuceloView* software.

When the process is finished, the cassette can be safely removed (see Section 6.5). Sometimes, however, the complete process described above may be interrupted. Please familiarize yourself with *Troubleshooting Error Messages*, Section 8.

## 6.4 Choosing Display Parameters

The measured results can be indicated in three different ways on the *NucleoCounter* display. These are called *Modes*. Modes 0 and 1 are not available; Modes 2, 3 and 4 are explained below:

Display Mode	Description	Sample Display Result
2	The result is limited to 1 decimal place	1.2x10E6
3	For maximum precision, the result is shown with 1, 2 or 3 decimal places, corresponding to a full representation of the number of cells per $\mu$ l.	1.2x10E4 1.23x10E5 1.234x10E6
4	The result is rounded off according to a statistical table. In this mode, the result is represented with values that reflect the precision of the <i>NucleoCounter</i> system. The even number to which the result is rounded off is generally within a half standard deviation of repeatability. The rounding off does not affect any significant digit in the result.	See Section 11.2 for a detailed look at Mode 4 results.

E = exponent

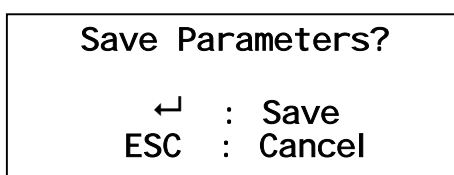
The default setting is Mode 3. If you wish to change the display mode, follow these instructions, using the keypad on the front panel:

1. Sequentially press the **F**, **2** and **1** keys.
2. Press the **↵** (“Enter”) key.
3. Press the number that corresponds to the mode you wish to select, e.g. **4**.
4. Press the **↵** key.

The newly chosen display parameters will be effective with the next analysis.

When you turn *NucleoCounter* off, prior to shutdown you will see the following message in the display:

**Figure 14: Mode Change Shutdown Display**



As indicated in the display:

5. To save the new mode you have programmed, press the **↵** (“Enter”) key.
6. To cancel the change and return to the previous mode setting, press the **ESC** button on the front panel.

The next time you perform an analysis, the results will be displayed in the mode you have selected.

## **6.5 Removing the Cassette**

To remove the cassette after analysis:

1. Lift the lid.
2. Grab the cassette as before (wearing protective gloves as always).
3. Remove the cassette.
4. Dispose of the cassette properly, according to federal, state and local regulations regarding the nature of the mixture it contains.

## **6.6 Other Possible Settings**

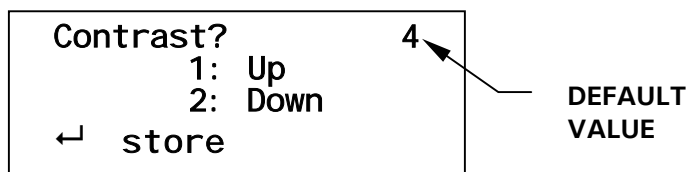
### **6.6.1 Reset Display Contrast**

You can adjust the contrast on the LCD display to make it brighter or darker. The factory-set default value is 4; you can choose values from 0 to 9.

To adjust the LCD contrast using the front panel keypad:

1. Turn the instrument on.
2. Sequentially press these keys: **F, 2, 2, 0**.
3. Press the **↵** (“Enter”) key.
4. You will see the following prompt in the display:

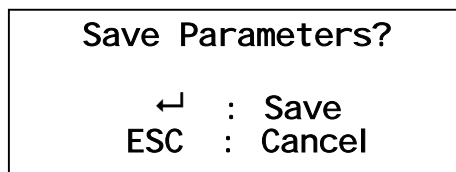
**Figure 15: Contrast Adjustment Display**



5. As indicated in the display, press the **1** key or the **2** key to adjust the contrast in the appropriate direction. **Do not press any other number key:** press the **1** (increase) or **2** (decrease) key as many times as necessary to arrive at the desired value (e.g., to change the value from 4 to 6, press the **1** key twice).
6. Press the **↵** key to save the new contrast value or the **ESC** key to cancel your entry and return to the previous setting.

If you saved a new contrast value, it will be applied throughout the current session. When you turn *NucleoCounter* off, prior to shutdown you will see the following message in the display:

**Figure 14: Mode Change Shutdown Display**



As indicated in the display:

7. To save the new mode you have programmed, press the **↵** (“Enter”) key.
8. To cancel the change and return to the previous mode setting, press the **ESC** button on the front panel.

## 6.6.2 Set Time & Date

If you are not using a PC or a printer with your *NucleoCounter*, skip this section.

If you are using a PC or a printer with your *NucleoCounter*, you can set the time of day and the date using the keypad on the front panel.

To set the time of day, using a 24-hour clock:

1. Turn the instrument on.
2. Sequentially press these keys: **F, 2, 0, 1**.
3. Press the **↵** (“Enter”) key.
4. The display will prompt you, showing the current time and, below it, hh:mm (hours:minutes). Press the numbers that correspond to the time you wish to program, e.g., for 2:05 pm (14:05), press **1, 4, 0**, and **5**. Be sure to use zero as the first digit for any hour or minute numbered from 1 to 9 (e.g., press **0, 9, 3**, and **8** for 9:38 am).
5. Press the **↵** key to save the time or the **ESC** key to cancel your entry if you have made an error.

To set the date, using a year-month-day (yy-mm-dd) format:

1. Turn the instrument on.
2. Sequentially press these keys: **F, 2, 0, 0**.
3. Press the **↵** (“Enter”) key.
4. Press the numbers that correspond to the date you wish to select: the last two digits of the year, then the two digits that represent the month, then the two digits that represent the day. Be sure to use zero as the first digit for any month or day numbered from 1 to 9 (e.g., **0, 5, 0, 4, 2, 8** will be displayed 05-04-28, meaning April 28, 2005).
5. Press the **↵** key to save the time or the **ESC** key to cancel your entry if you have made an error.

### 6.6.3 Reset the Counter

**If you are not using a printer** with your *NucleoCounter*, skip this section.

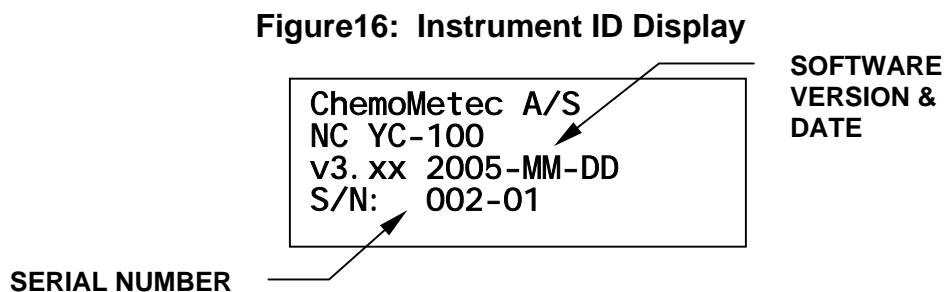
If you are using a printer with your *NucleoCounter*, NucleoCounter automatically counts each analysis performed by the instrument, ascribing a number from 1 to 9999 (after which it reverts to 1). You may have occasion to reset the counter prior to reaching 9999. To reset the counter to 1, follow these instructions using the keypad on the front panel:

1. Turn the instrument on.
2. Sequentially press these keys: **F, 3, 0**.
3. Press the **↵** (“Enter”) key.
4. Press the **↵** key again to reset the counter or the **ESC** key to cancel the resetting and revert to the most recent analysis number. If you press the **↵** key, the display will read **Resetting...** for a brief time.

#### 6.6.4 Instrument ID

If you need a reminder of your *NucleoCounter*'s version number and date and/or the instrument's serial number, you can use the front panel keypad to access the instrument information display:

1. Sequentially press these keys: **F, 1, 0, 0**.
2. Press the ↵ ("Enter") key.
3. The instrument identification display will appear:



4. Press the ↵ ("Enter") key again to exit this display.

## 7 MAINTENANCE

Preventive maintenance keeps your equipment in proper working condition. When performed routinely, maintenance results in longer life for your equipment. It also reduces time lost due to equipment failure.

***ELECTRICAL HAZARD!***

Always disconnect the power supply from the *NucleoCounter* rear panel before performing maintenance.

***CAUTION!***

Never immerse *NucleoCounter* in liquid of any kind.

### 7.1 Cleaning

#### 7.1.1 Exterior

Clean the *NucleoCounter* body exterior with a soft moist cloth, gently wiping the surface. If there is any contamination that does not come off immediately, rub it gently with a soft cloth moistened with mild detergent.

***CAUTION!***

Never use organic solvents or aggressive detergents to clean the exterior of *NucleoCounter* as they can damage the surface.

When you clean outside the insertion area, take great care to avoid the introduction of any liquid or dust particles inside *NucleoCounter*.

#### 7.1.2 Interior

Any material (dust, dirt, cell contamination) on the surface of an optical component inside *NucleoCounter*'s cassette chamber can influence the collected image (visible and recordable with an optional computer connection and the *NucleoView*<sup>TM</sup> software).

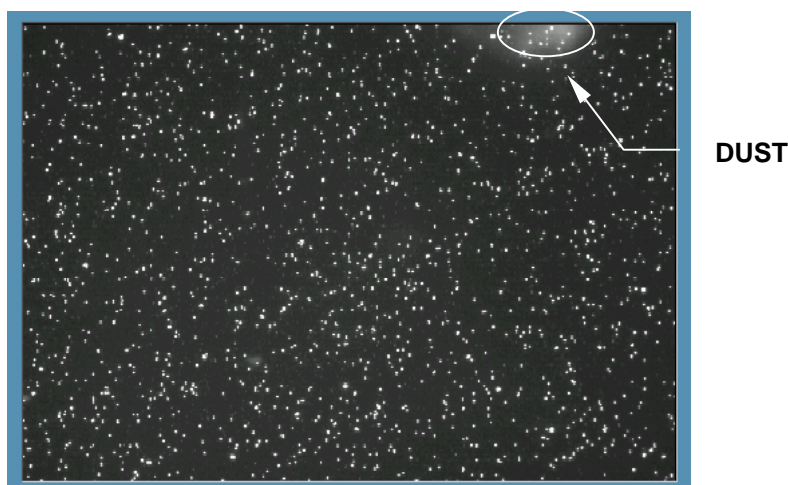


### CAUTION!

- **Never wipe the cassette insertion area with a moist or a wet cloth, as drops of liquid may inadvertently enter the insertion area.**
- **Never attempt to clean inside the cassette insertion area other than with dry compressed air; you may damage the optical system. Call NBS for maintenance by a technician.**

A dust particle will normally be visible as a faint object in the displayed image. Since it is also possible that such contamination is on the surface of the *NucleoCassette*, not the optical components, check for any objects that appear in all images, in the same location. Such phenomena are most easily perceived in samples with only a few cells. An example of a relatively large contamination of the optical system is shown in Figure 16 below.

**Figure 16: Optical System Contamination**



The presence of a dust particle normally will not influence the cell count. *NucleoCounter*'s built-in image analyzer will distinguish between cells and dust particles, since the cell image is generally much smaller.

Remove dust from the insertion region using dry compressed air (*see Figure 17 on the following page*).

If liquid is spilled on the instrument, it may contaminate the components of the optical system. While this sort of contamination can show up in the image in a variety of ways (and have diverse causes), a common feature is that it cannot be removed with compressed air. **Do not attempt to clean it yourself; call New Brunswick Scientific to request a technician's assistance.**

**Figure 17: Removing Dust from Cassette Compartment**



## 7.2 Fuse Replacement

To replace the fuse, follow these steps:



1. **Unplug the power supply cord from the outlet**, and disconnect the power supply cord from *NucleoCounter*'s rear panel.
2. Remove the fuse holder, located above the power supply cord connector (*see Figure 18 below*) using a turn CCW screwdriver.

**Figure 18: Replacing a Fuse**



3. Remove the fuse from its holder and inspect it. If the fuse is not broken, skip to Step #7.

4. If the fuse needs to be replaced, use a UL-listed 600mA/250VAC fuse (UL 248).
5. Insert the new fuse into the holder.
6. Re-insert the holder at its location above the power supply cord connector.
7. If *NucleoCounter* does not turn on after replacing the fuse, or if the fuse was not broken, contact New Brunswick Scientific for technical assistance.

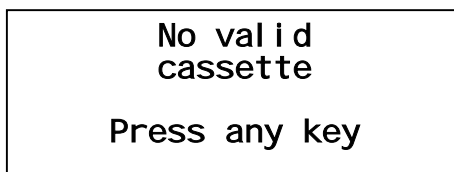
## 8 TROUBLESHOOTING

*Certain conditions will cause NucleoCounter to display error messages during operation. Corrective actions are suggested in this chapter, for responding to error messages and other operational problems. If these actions do not correct the problem, however, be sure to contact New Brunswick Scientific for technical assistance. Do not attempt to repair the instrument yourself.*

### 8.1 No Valid Cassette

During analysis, *NucleoCounter* may determine that the *NucleoCassette* inserted is not valid. This will cause the instrument to display this error message:

**Figure 19: Invalid Cassette**



There can be several reasons for *NucleoCounter* to determine that the cassette is invalid:

- This *NucleoCassette* has been analyzed previously.
- The cassette was not inserted properly into *NucleoCounter*.
- There is no cassette inside *NucleoCounter*.

To remedy the problem:

1. Remove the *NucleoCassette* (if there is one), and examine it. If it has been analyzed before, replace it with a new cassette and begin the process again.
2. If the cassette is new and ready for analysis, re-insert it, taking care to do so properly (make sure it clicks in place).
3. If no cassette was found in the insertion area, insert a loaded cassette.
4. The *NucleoCounter* system is now ready to perform an analysis.

## 8.2 Analysis Aborted

*NucleoCounter* displays the error message shown in Figure 20 when the analysis has been automatically aborted because the stained mixture inside the *NucleoCassette* has not flowed into the measurement chamber within a prescribed, factory-set time.

**Figure 20: Aborted Analysis**



Analysis aborted  
Flow sensor error  
Refer to manual  
Press any key

The most common cause is that too small a sample has been loaded into the cassette (see Section 5.5 and Figure 10 for information about minimum and maximum loading):

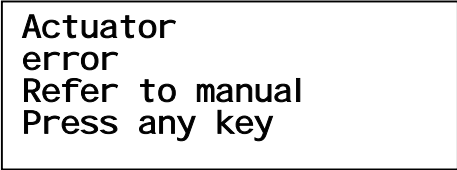
1. Open the lid, remove the cassette.
2. **If it was improperly inserted**, skip to the following paragraph. **If not**, dispose of the cassette (following appropriate regulations).
3. Close the lid.
4. Press any key on the keypad to reset *NucleoCounter*.
5. Load a new cassette, taking care to press the loading piston gently, then repeat the analysis.

Another frequent cause is improper cassette insertion. If the cassette is not fully inserted, or not correctly seated, *NucleoCounter* cannot push the loading piston down to send the sample into the measurement chamber. If the cassette was improperly inserted:

1. Open the lid, remove the cassette.
2. Press any key on the keypad to reset *NucleoCounter*.
3. Re-insert the cassette, making sure it clicks into place.
4. Close the lid and press the **Run** button.

## 8.3 Actuator Errors

The actuator is the mechanical component that presses down the piston in the *NucleoCassette*, to send the sample into the measurement chamber. If the actuator misfunctions, an error message will appear in the display (see Figure 21):

**Figure 21: Actuator Error Message**A rectangular box with a black border containing the text: Actuator error, Refer to manual, Press any key.

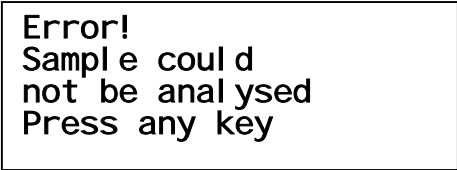
Actuator  
error  
Refer to manual  
Press any key

If you receive this error message, follow these steps:

1. Open the lid.
2. Examine whether the *NucleoCassette* has been properly inserted. If the cassette is properly inserted, do not remove it and skip to Step #6.
3. If the cassette is improperly seated, remove it and re-insert it correctly, until it clicks.
4. Press any key on the keypad to reset *NucleoCounter*.
5. Close the lid and press the **Run** button.
6. If the cassette was properly inserted, close the lid, press any key on the keypad to reset the instrument, then turn *NucleoCounter* off and on a few times using the **On/Off** button.
7. If you hear the actuator motor during start-up, it is most likely the error has been corrected.
8. If you still have a problem when you run the analysis, call NBS for technical assistance.

#### **8.4 Analysis Impossible**

When *NucleoCounter* cannot determine the cell concentration during analysis, the following error message is displayed:

**Figure 22: No Analysis**A rectangular box with a black border containing the text: Error!, Sample could not be analysed, Press any key.

Error!  
Sample could  
not be analysed  
Press any key

This error message appears if the concentration of stained DNA inside the cassette measurement chamber is far too high because the sample is not sufficiently diluted.

This message may also appear if the lid was not closed properly, in which case ambient light may have interfered with the analysis.

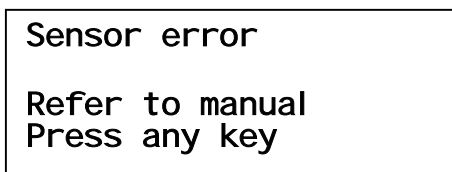
Follow these steps:

1. If the lid was closed properly, skip to step #2. If the lid was not properly closed, remove and reinsert the cassette, close the lid and run the analysis again.
2. Open the lid, remove the *NucleoCassette* and dispose of it (following appropriate regulations).
3. Close the lid and press any key to reset *NucleoCounter*.
4. Dilute the cell suspension from which the sample was taken, according to the instructions in Section 5.4.
5. Load a sample of the diluted lyzate into a new *NucleoCassette*.
6. Open the lid, properly insert the new cassette, close the lid and press the **Run** button to run a new analysis.

## 8.5 **Sensor Error**

Sensors inside *NucleoCounter* read the black dot(s) printed onto the *NucleoCassette* each time you run an analysis. The dots code thickness of the measurement chamber in the cassette, which affects the volume of sample it contains; this is an important parameter for each analysis. If one or more of these sensors breaks, the following error message will be displayed:

**Figure 23: Sensor Error**



If this error message is displayed, analyses can still be made (after you press any key on the keypad to reset the instrument). The accuracy of the results, however, cannot be guaranteed as the broken sensor(s) may cause *NucleoCounter* to read the cell volume incorrectly.

Call New Brunswick Scientific for technical assistance.

## 8.6 **Power-On Failure**

If you find that you cannot turn *NucleoCounter* on, follow these steps to troubleshoot the problem:

1. Verify that the power supply cord is connected, both to *NucleoCounter*'s rear panel and to your electrical outlet.

2. Make sure the power outlet is functioning.
3. After Steps #1 & #2, if *NucleoCounter* still does not turn on, inspect the fuse and replace it if necessary, as outlined in Section 7.2. The fuse holder is located above the power plug connector on the rear panel.

## 9 SPECIFICATIONS

### 9.1 *NucleoCounter*

<b>Specificity</b>	Counts yeast cells stained with the DNA-specific fluorescent dye, propidium iodide.		
<b>Sample Type</b>	Suspension of dispersed yeast cells		
<b>Sample Consumption</b>	The sample is mixed with a lysis buffer prior to analysis. We recommend that at least 100 $\mu$ l of the sample be used for this dilution in order to reduce the probability for errors in sample handling.		
<b>Reagent Consumption</b>	We recommend using equal amounts of lysis buffer and sample.		
<b>Lyzate Mixture Consumption</b>	We recommend using at least 100 $\mu$ l of lyzate mixture (sample + reagent) for cassette loading. Approximately 50 $\mu$ l of lyzate will load into the cassette.		
<b>Analysis Volume</b>	Approximately 1 $\mu$ l of lyzate mixture is analyzed in the <i>NucleoCounter</i> YC-100.		
<b>Measurement Range (cell density in lyzate)</b>	The total range is $5 \times 10^3$ to $2 \times 10^6$ cells/ml. The recommended range is $5 \times 10^4$ to $2 \times 10^6$ cells/ml.		
<b>Presentation of Result</b>	YC-100 presents an accurate estimate of cell density in the lyzate contained in the cassette being analyzed. To obtain cell concentration in a cell suspension, the analysis result must be multiplied by the appropriate multiplication factor to compensate for dilution of the sample.		
<b>Analysis Time</b>	Within 30 seconds of pressing the <b>Run</b> button.		
<b>Operation</b>	Menu-controlled by means of keyboard and LCD display. For indoor use only.		
<b>Dimensions</b>	<b>Height</b>	<b>Width</b>	<b>Depth</b>
	10.2 in./26 cm	15 in./38 cm	8.7 in./22 cm
<b>Weight</b>	6.6 lbs / 3 kg		
<b>Power Input*</b>	12 VDC (11-13 VDC)		
<b>Power Supply*</b>	100-240 VAC from electrical outlet, transformed by power supply cord (provided)		
<b>Power Supply Cord*</b>	See Section 9.1.1 below.		
<b>Fusing (UL-Listed)</b>	600mA/250 VAC (UL 248)		
<b>Power Consumption</b>	<b>Peak</b>	<b>Ready Mode</b>	<b>Standby Mode</b>
	25 W	2.5 W	2 mW
<b>Ambient Operating Conditions</b>	Maximum relative humidity 80% for temperatures up to 31° C decreasing linearly to 65% relative humidity at maximum 35°C. Minimum temperature 15°C.		
<b>USB Connection*</b>	USB version 1.1; does not support USB Hubs.		

\* Be sure to read Section 3.3, *Power Supply* and Section 3.5, *USB Cable Cautions*.

### 9.1.1 Power Supply Cord

Following are the specifications for the detachable cord set for Class I and Class II power supply, and for U.S. and European use:

<b>Class I-Specified Power Supplies*</b>	
<b>U.S. 125 VAC</b>	UL-listed, type SVT, rated min. 60C, 18 AWG, 3 conductors. Provided with molded-on grounding-type (NEMA 5-15P) attachment plug, rated min. 125 VAC, min. 2.5A. Opposite end terminates in molded-on IEC320-style connector, rated min. 125 VAC, min. 2.5A.
<b>Europe 250 VAC</b>	Cord type min. H05RR-F or min. H03VV-F or min. H03VVH2-F, rated min. 60C, 3x0.75 mm <sup>2</sup> . Provided with molded-on grounding-type attachment plug, rated min. 250 VAC, min. 2.5A. Opposite end terminates in molded-on IEC320-style connector, rated min. 250 VAC, min. 2.5A.
<b>Class II-Specified Power Supplies*</b>	
<b>U.S. 125 VAC</b>	UL-listed, type SPT-2 or SVT, rated min. 60C, 18 AWG, 3 conductors. Provided with molded-on ungrounding-type (NEMA 5-15P) attachment plug, rated min. 125 VAC, min. 2.5A. Opposite end terminates in molded-on IEC320-style connector, rated min. 125 VAC, min. 2.5A.
<b>Europe 250 VAC</b>	Cord type min. H05RR-F or min. H03VV-F or min. H03VVH2-F, rated min. 60C, 2x0.75 mm <sup>2</sup> . Provided with molded-on ungrounding-type attachment plug, rated min. 250 VAC, min. 2.5A. Opposite end terminates in molded-on IEC320-style connector, rated min. 250 VAC, min. 2.5A.

\* Be sure to read Section 3.3, *Power Supply*.

## 9.2 Safety Standards

*NucleoCounter* complies with EMC and EMI safety standards as follows:

<b>EN61326: 1977 + A1: 1998 (Class B) + A2: 2000</b>	Electrical equipment for measurement, control and laboratory use – EMC requirements (emission and immunity). Annex B and C from A1: 1998 is used.
<b>Part 15 of the FCC Rules</b>	Operation is subject to the following two conditions: 1. This device may not cause harmful interference. 2. This device must accept any interference received, including interference that may cause undesired operation.

# 10 PARTS & ACCESSORIES

## 10.1 Spare Parts

Item Description	NBS Part Number
Fuse, Slo-Blo® 600mA/250 VAC (UL 248)	P0380-3930
External USB Cable	P0720-9890
Power Supply Cord	P0720-9880

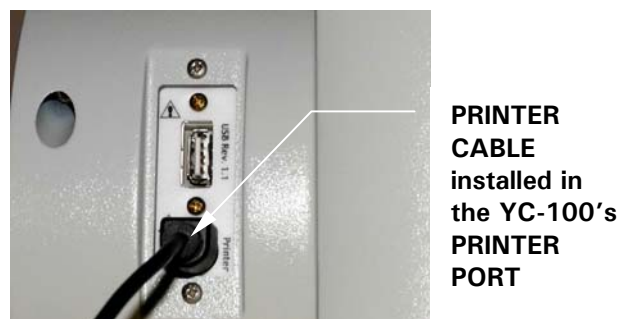
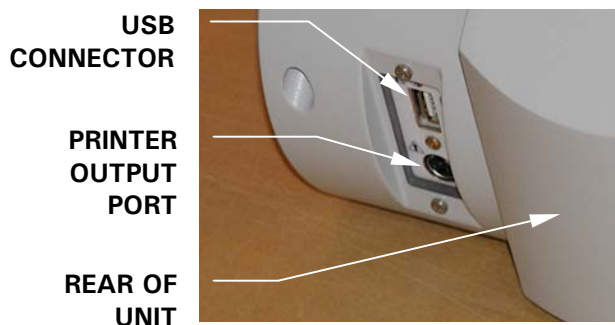
## 10.2 Accessories

Item Description	NBS Part Number
100 <i>NucleoCassettes</i> : one box of 10 foil bags, 10 cassettes per bag	M1293-0100
Lysing buffer, Reagent Y100	P0820-5185
Buffer Dispenser Pump (for Y100)	P0820-5179
<i>NucleoView</i> ™ software CD	P0540-5440
Reagent holder	P0860-5178
Printer, impact type, starter kit	P0620-6440

### 10.2.1 Optional Printer

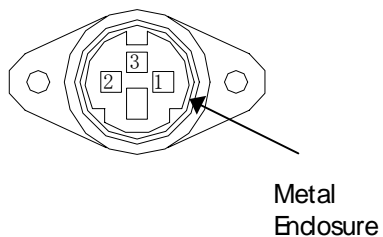
NucleoCounter YC-100 is equipped with a printer output port for use with an external printer. The port is located on the rear panel, just below the USB connector (see *Figures 24a and 24b*):

**Figure 24a: Printer Port Location**      **Figure 24b: Installing Printer Cable**



The printer output connector has a metal enclosure and 3 pins that mate to the printer cable (see *Figure 25*):

**Figure 25: Printer Port**



Pin Number	Pin Designation	Maximum Voltage Level*
1	unassigned	no signal
2	Rx	$\pm 10$ VDC
3	Tx	$\pm 10$ VDC
metal enclosure	DGND	0 VDC

\* in normal operation

# 11 APPENDICES

## 11.1 *Appendix A: List of Illustrations*

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## 11.2 *Appendix B: Mode 3 vs. Mode 4 Results*

To help guide your choice of analysis display mode, the table below provides an overview of the results displayed in Mode 3 (maximum precision), compared to Mode 4 (statistical table):

Lower limit (mode 3)	Upper limit (mode 3)	Mode 4
N/A	N/A	$< 5 \times 10^3$
N/A	$7 \times 10^3$	$5.0 \times 10^3$
$8 \times 10^3$	$1.3 \times 10^4$	$1.0 \times 10^4$
$1.4 \times 10^4$	$2.5 \times 10^4$	$2.0 \times 10^4$
$2.6 \times 10^4$	$3.5 \times 10^4$	$3.0 \times 10^4$
$3.6 \times 10^4$	$4.5 \times 10^4$	$4.0 \times 10^4$
$4.6 \times 10^4$	$5.5 \times 10^4$	$5.0 \times 10^4$
$5.6 \times 10^4$	$6.5 \times 10^4$	$6.0 \times 10^4$
$6.6 \times 10^4$	$7.5 \times 10^4$	$7.0 \times 10^4$
$7.6 \times 10^4$	$8.5 \times 10^4$	$8.0 \times 10^4$
$8.6 \times 10^4$	$9.5 \times 10^4$	$9.0 \times 10^4$
$9.6 \times 10^4$	$1.05 \times 10^5$	$1.0 \times 10^5$
$1.06 \times 10^5$	$1.15 \times 10^5$	$1.1 \times 10^5$
$1.16 \times 10^5$	$1.25 \times 10^5$	$1.2 \times 10^5$
$1.26 \times 10^5$	$1.35 \times 10^5$	$1.3 \times 10^5$
$1.36 \times 10^5$	$1.45 \times 10^5$	$1.4 \times 10^5$
$1.46 \times 10^5$	$1.55 \times 10^5$	$1.5 \times 10^5$
$1.56 \times 10^5$	$1.65 \times 10^5$	$1.6 \times 10^5$
$1.66 \times 10^5$	$1.75 \times 10^5$	$1.7 \times 10^5$
$1.76 \times 10^5$	$1.85 \times 10^5$	$1.8 \times 10^5$
$1.86 \times 10^5$	$1.95 \times 10^5$	$1.9 \times 10^5$
$1.96 \times 10^5$	$2.10 \times 10^5$	$2.0 \times 10^5$
$2.11 \times 10^5$	$2.30 \times 10^5$	$2.2 \times 10^5$
$2.31 \times 10^5$	$2.50 \times 10^5$	$2.4 \times 10^5$
$2.51 \times 10^5$	$2.70 \times 10^5$	$2.6 \times 10^5$
$2.71 \times 10^5$	$2.90 \times 10^5$	$2.8 \times 10^5$
$2.91 \times 10^5$	$3.10 \times 10^5$	$3.0 \times 10^5$
$3.11 \times 10^5$	$3.30 \times 10^5$	$3.2 \times 10^5$
$3.31 \times 10^5$	$3.50 \times 10^5$	$3.4 \times 10^5$
$3.51 \times 10^5$	$3.70 \times 10^5$	$3.6 \times 10^5$
$3.71 \times 10^5$	$3.90 \times 10^5$	$3.8 \times 10^5$
$3.91 \times 10^5$	$4.10 \times 10^5$	$4.0 \times 10^5$
$4.11 \times 10^5$	$4.30 \times 10^5$	$4.2 \times 10^5$
$4.31 \times 10^5$	$4.50 \times 10^5$	$4.4 \times 10^5$
$4.51 \times 10^5$	$4.70 \times 10^5$	$4.6 \times 10^5$
$4.71 \times 10^5$	$4.90 \times 10^5$	$4.8 \times 10^5$
$4.91 \times 10^5$	$5.12 \times 10^5$	$5.00 \times 10^5$
$5.13 \times 10^5$	$5.37 \times 10^5$	$5.25 \times 10^5$
$5.38 \times 10^5$	$5.62 \times 10^5$	$5.50 \times 10^5$
$5.63 \times 10^5$	$5.87 \times 10^5$	$5.75 \times 10^5$
$5.88 \times 10^5$	$6.12 \times 10^5$	$6.00 \times 10^5$
$6.13 \times 10^5$	$6.37 \times 10^5$	$6.25 \times 10^5$

...continued...

Lower limit (mode 3)	Upper limit (mode 3)	Mode 4
6.38 x 10E5	6.62 x 10E5	6.50 x 10E5
6.63 x 10E5	6.87 x 10E5	6.75 x 10E5
6.88 x 10E5	7.12 x 10E5	7.00 x 10E5
7.13 x 10E5	7.37 x 10E5	7.25 x 10E5
7.38 x 10E5	7.62 x 10E5	7.50 x 10E5
7.63 x 10E5	7.87 x 10E5	7.75 x 10E5
7.88 x 10E5	8.12 x 10E5	8.00 x 10E5
8.13 x 10E5	8.37 x 10E5	8.25 x 10E5
8.38 x 10E5	8.62 x 10E5	8.50 x 10E5
8.63 x 10E5	8.87 x 10E5	8.75 x 10E5
8.88 x 10E5	9.12 x 10E5	9.00 x 10E5
9.13 x 10E5	9.37 x 10E5	9.25 x 10E5
9.38 x 10E5	9.62 x 10E5	9.50 x 10E5
9.63 x 10E5	9.87 x 10E5	9.75 x 10E5
9.88 x 10E5	1.024 x 10E6	1.00 x 10E6
1.025 x 10E6	1.074 x 10E6	1.05 x 10E6
1.075 x 10E6	1.125 x 10E6	1.10 x 10E6
1.126 x 10E6	1.175 x 10E6	1.15 x 10E6
1.176 x 10E6	1.225 x 10E6	1.20 x 10E6
1.226 x 10E6	1.275 x 10E6	1.25 x 10E6
1.276 x 10E6	1.325 x 10E6	1.30 x 10E6
1.326 x 10E6	1.375 x 10E6	1.35 x 10E6
1.376 x 10E6	1.425 x 10E6	1.40 x 10E6
1.426 x 10E6	1.475 x 10E6	1.45 x 10E6
1.476 x 10E6	1.525 x 10E6	1.50 x 10E6
1.526 x 10E6	1.575 x 10E6	1.55 x 10E6
1.576 x 10E6	1.625 x 10E6	1.60 x 10E6
1.626 x 10E6	1.675 x 10E6	1.65 x 10E6
1.676 x 10E6	1.725 x 10E6	1.70 x 10E6
1.726 x 10E6	1.775 x 10E6	1.75 x 10E6
1.776 x 10E6	1.825 x 10E6	1.80 x 10E6
1.826 x 10E6	1.875 x 10E6	1.85 x 10E6
1.876 x 10E6	1.925 x 10E6	1.90 x 10E6
1.926 x 10E6	1.975 x 10E6	1.95 x 10E6
1.976 x 10E6	N/A	2.00 x 10E6
N/A	N/A	> 2 x 10E6

N/A: Not Applicable

E: Exponent

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