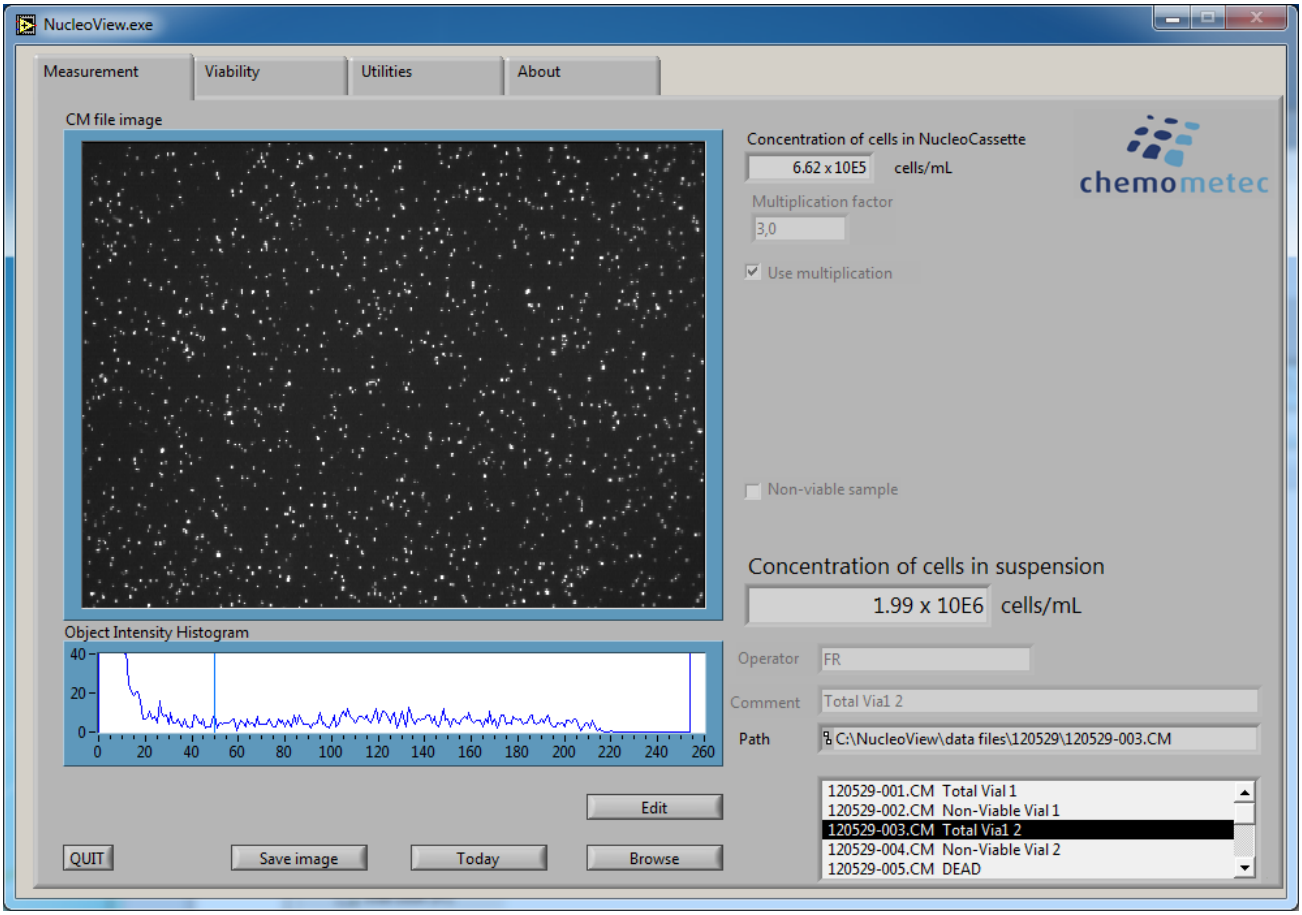
NucleoView™

User's Guide

P/N 991-0006

Version 2.5



Technology that counts

chemometec

NucleoView™

A part of the NucleoCounter®™ NC-100 and
NucleoCounter® YC-100 ™system

Manual No. 991-0006 (English)

Version 2.5

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Customer support

If you have questions about installing or using your NucleoView™ software check this user's guide first - you will find answers to most of your questions here. If you need further assistance, please contact customer support at ChemoMetec A/S.

Contacting support

Technical information including product literature, answers to questions regarding the operation of the NucleoCounter™ not covered in this document and information on software upgrade is available through the following:

- For Email support, send questions to NucleoCounter Technical Support on the address support@chemometec.com
- Check out the FAQ section under support at www.chemometec.com
- To speak with a Technical Support Specialist, call (+45) 48 13 10 20.

Please note the NucleoCounter™ serial number and have it available when contacting ChemoMetec A/S for support. The NucleoCounter™ serial number is found on a label affixed to the bottom of the instrument and in the display upon start-up. The version number of the NucleoCounter firmware shall also be noted. Refer to the User's Guide of you NucleoCounter instrument in order to retrieve that.

Sales and ordering information

For pre-sales assistance with NucleoCounter™ or the NucleoView™ software, to place an order for a NucleoCounter™ or consumables, call (+45) 48 13 10 20, or mail to Sales@chemometec.com

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Intended use

The NucleoView™ software is developed as a dedicated accessory to the NucleoCounter™ NC-100 and the NucleoCounter™ YC-100 instruments; it is intuitive and easy to use. NucleoView™ offers the user several features when connected to the NucleoCounter™.

With NucleoView™, correction for dilution of the samples and calculation of viability becomes easy. When documentation of results is required, NucleoCounter™ results can be printed or saved for later use using the NucleoView application. NucleoView™ offers several additional features, which are described in detail in this manual.

Though the NucleoView software can be operated in a safe way the program is not CFR Part 11 compliant.

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Table of contents

1 Installation

When the NucleoCounter is connected to a computer running NucleoView it is possible to register the estimated cell concentration and the collected image of the NucleoCassette chamber. Connecting a NucleoCounter to a computer does not affect the operation of the NucleoCounter in any way and the NucleoView software performs no additional image analysis.

The installation[1] procedures below describe the installation of the NucleoView software and the USB driver on computers running Windows 7. It is possible to install NucleoView and the USB driver on computers running other operating systems than Windows 7. For a list of supported operating systems please refer to chapter 8 Technical specifications.

The communication between the NucleoCounter and NucleoView is based on the USB 1.1 protocol, thus the computer must be equipped with an USB 1.1 compatible port.

NOTICE - IMPORTANT!

Before installation of NucleoView Package ver. 2.4, check if an older version of NucleoView is installed on the computer.

Any other version of NucleoView is most likely to be located in the folder C:\Nucleoview.

If present it must be uninstalled by executing the uninst.exe file placed in the folder C:\Nucleoview.

If no older version of NucleoView is installed on the computer follow the directions in chapter 1.1 Installation of the NucleoView Package ver. 2.4 - Windows 7.

If an older version of NucleoView is installed on the computer, it has to be uninstalled before NucleoView Package ver. 2.4 can be installed. Follow the 3 steps below.

1. Uninstall the older version of NucleoView (see chapter 1.2.1 Removal of NucleoViewTM - Windows 7)

2. Install NucleoView Package ver. 2.4 by following the directions in chapter 1.1 Installation of the NucleoView Package ver. 2.4 - Windows 7

The NucleoCounter will then be ready for use with the NucleoView software ver. 2.2[1].

[1] This installation is called ”NucleoView Package ver. 2.4”, since it contains a new driver for 64-bit OS also. However the actual NucleoView program is still ver. 2.2.

1.1 Installation of the NucleoView Package ver. 2.4 - Windows 7[2]

When the computer has been turned on and is finished with the start-up process, insert the USB Flash disk supported with the NucleoCounter to the computer. Let the computer install the USB Flash disk and the open the DISK.

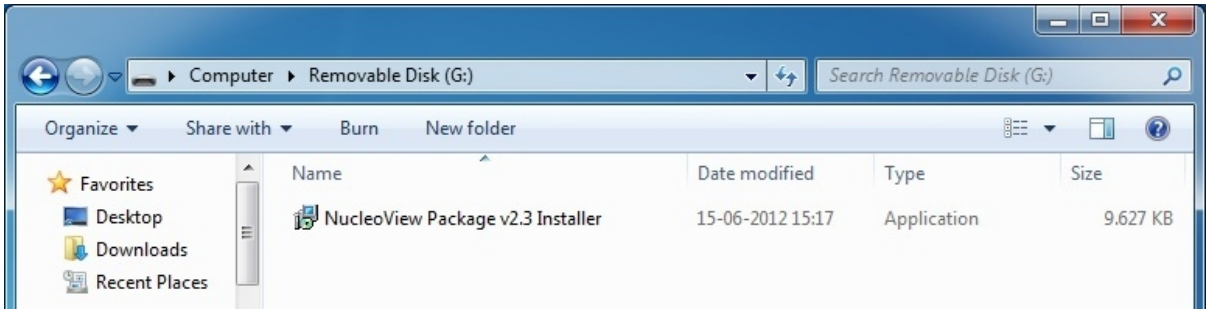


Figure 1 NucleoView Package v2.4 Installer located on the USB Flash disk

Activate the application "NucleoView Package v2.4 Installer" and allow (hit Yes) the Installer program to make changes to the computer (see Figure 2 User Account Control)

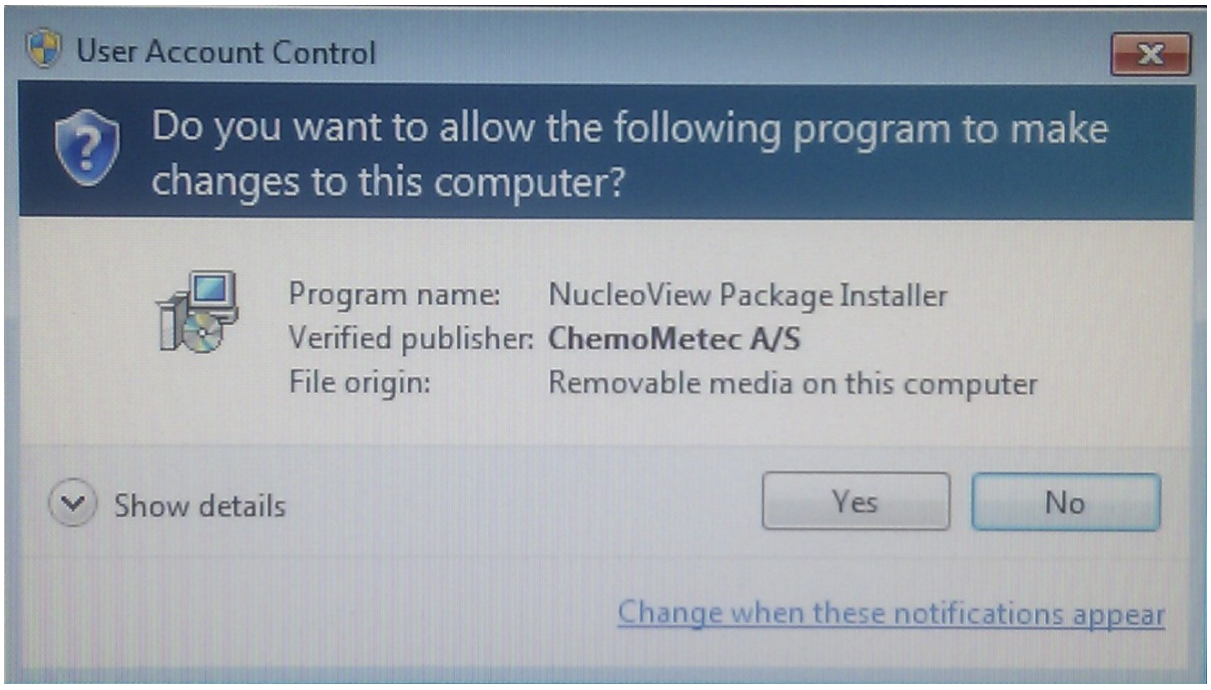


Figure 2 User Account Control

The Installer will welcome you and you can now continue the installation in a very controlled manor.

[2] This description applies to Windows 7 only.

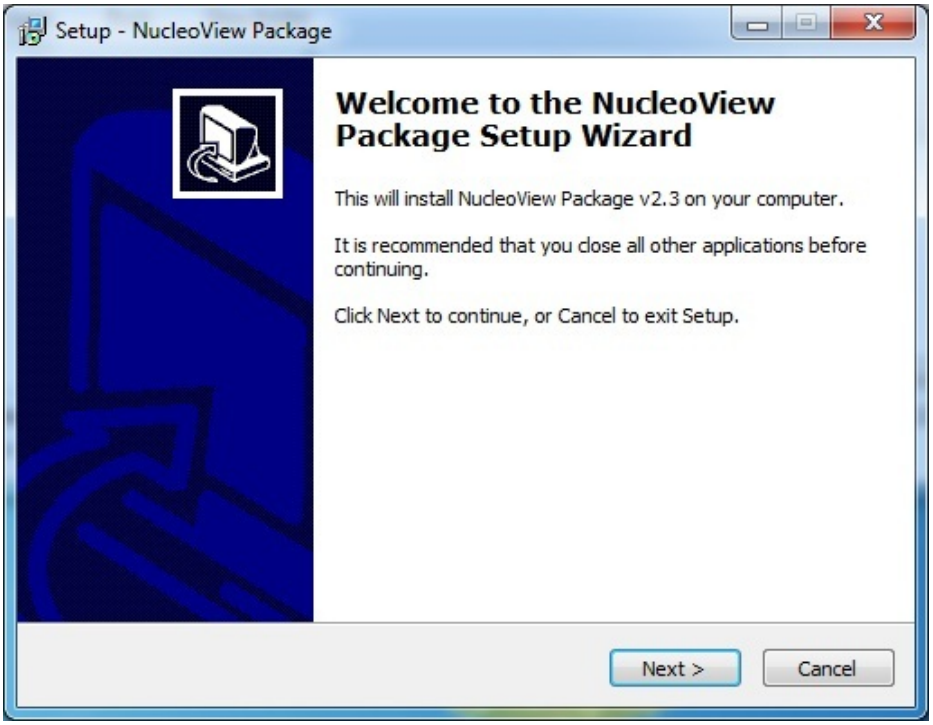


Figure 3 Installer Setup - Welcome

Hit “Next”

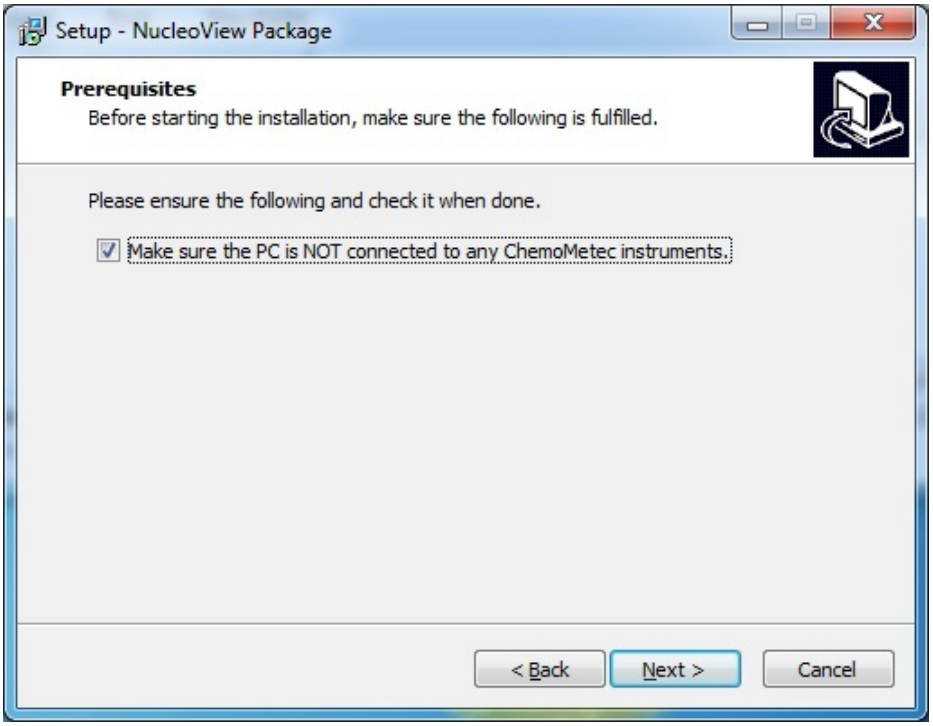


Figure 4 Installer Setup - Prerequisites

Check the check box in Figure 4 Installer Setup - Prerequisites and hit “Next”.

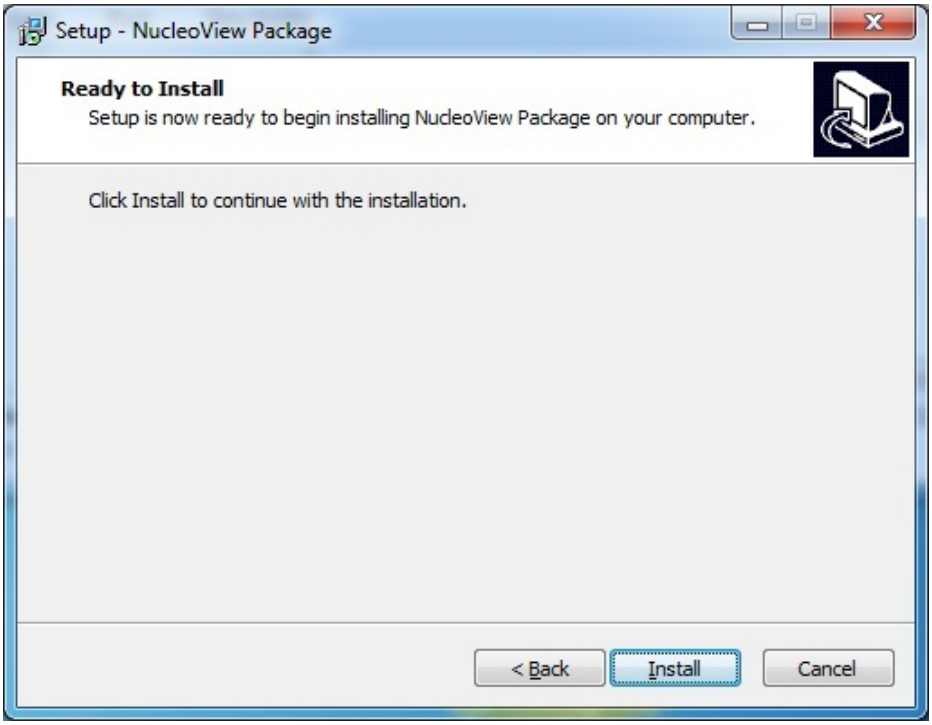


Figure 5 Installer Setup – Ready to Install

Hit “Install”.

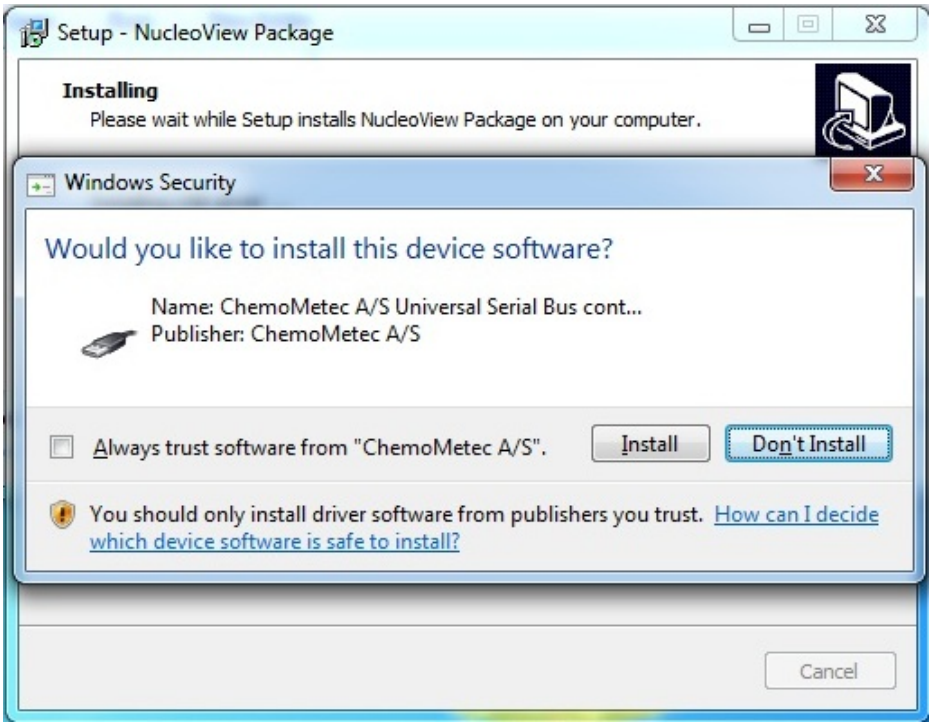


Figure 6 Installer Setup – Installing and Windows Security

After the actual Installation is started you will be prompted to accept the installation of the device software for the USB driver. Hit “Install”

In the background the installation continues and when this is over the following dialogue appears. Then hit “Finish”.

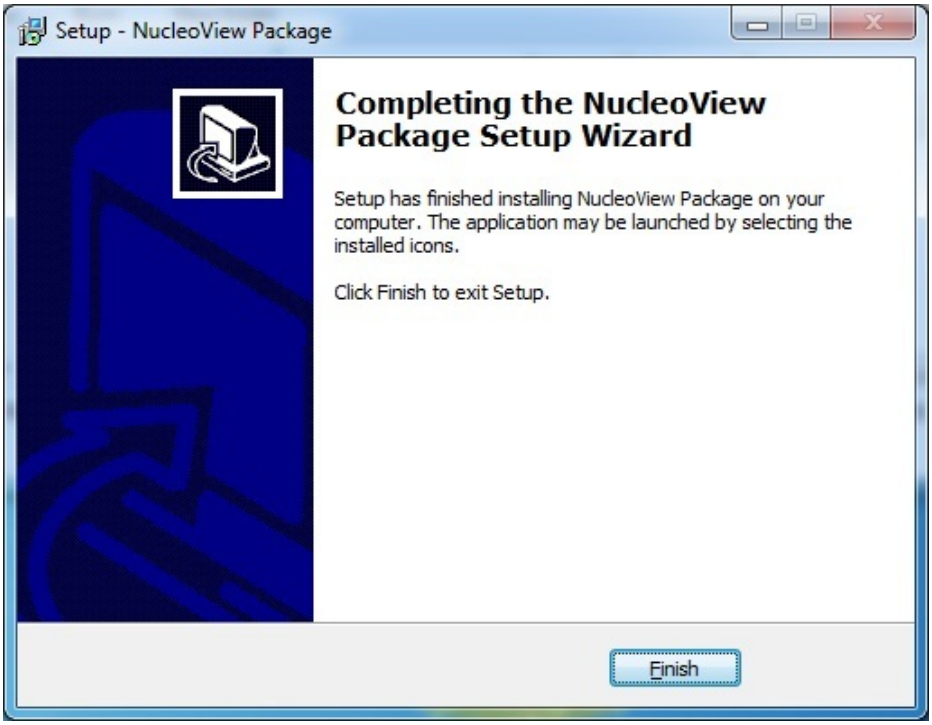
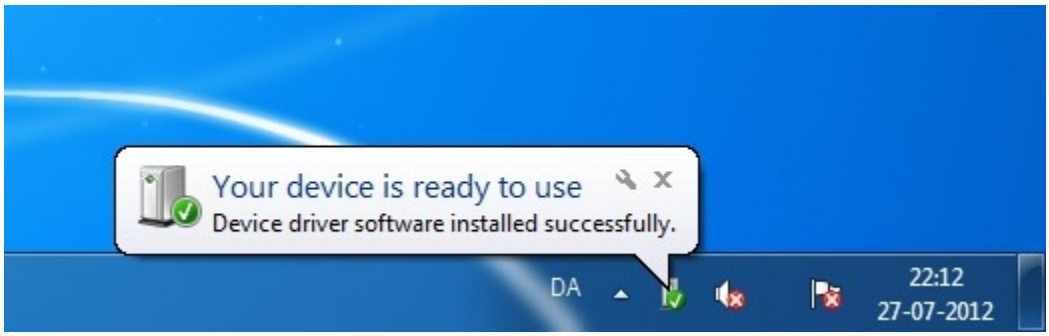


Figure 7 Installer Setup – Completed

The first time a NucleoCounter is turned on while connected to the computer, the computer will identify the NucleoCounter as new hardware and install the drivers for it. So the screen will show the following message.



1.2 Removal and upgrade of the NucleoView software

For removal[3] of the NucleoView software follow the directions below. If you upgrade from NucleoView 2.0 or 2.1 these version **must** be removed first.

[3] For removal of NucleoView 2.0 or 2.1 refer to the original manual.

1.2.1 Removal of NucleoView™ - Windows 7[4]

To remove the NucleoView software[5] open the NucleoView folder (see Figure 8 Contents of the NucleoView folder (before uninst000.exe is activated).). The folder should be located in the root directory of the local hard disk (C:\Nucleoview) if the location was not changed during the installation.

In the NucleoView folder double-click on "uninst000.exe[6]" and confirm that the NucleoView software is to be uninstalled. The NucleoView software will then be removed from your computer. Any saved data will remain on the computer.

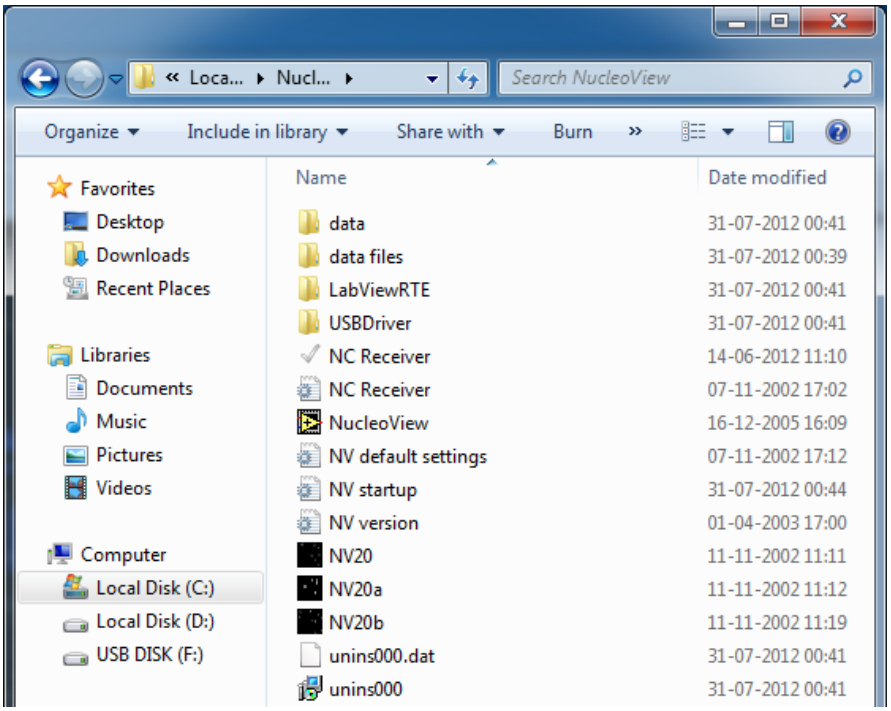


Figure 8 Contents of the NucleoView folder (before uninst000.exe is activated)[7].

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[4] This description applies to Windows 7 only.

[5] In order to remove the program correct, first quit the NC Receiver.exe program if it is running.

[6] "uninst000.exe" or "uninst000" depending on your folder settings in your OS.

[7] If you at any time created your own Default settings you will also have a file called "NV default settings.ini".

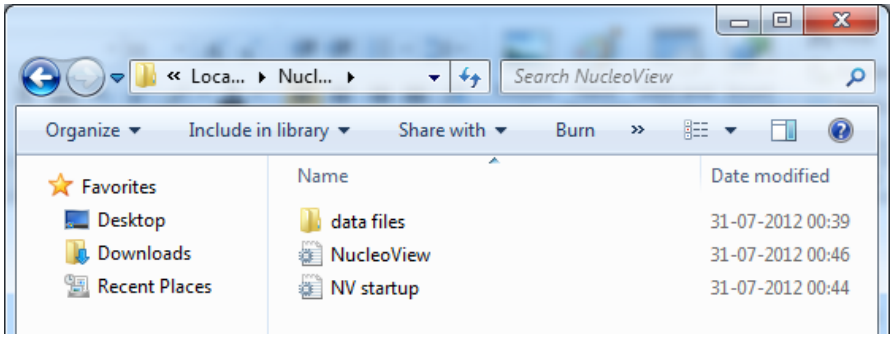

Figure 9 Contents of the NucleoView folder (after uninst000.exe is activated)[8].

If the program has to be removed completely you can just delete the files and folders now. Remember you will loose ALL data, if you delete the contents of the "data files" folder from Figure 9.

In case you are upgrading the NucleoView program, DO NOT delete any files or folders after "uninst000.exe" was activated.

1.2.2 Removal of all installed components[4]

In case all installed components must be removed the following procedure can be used.

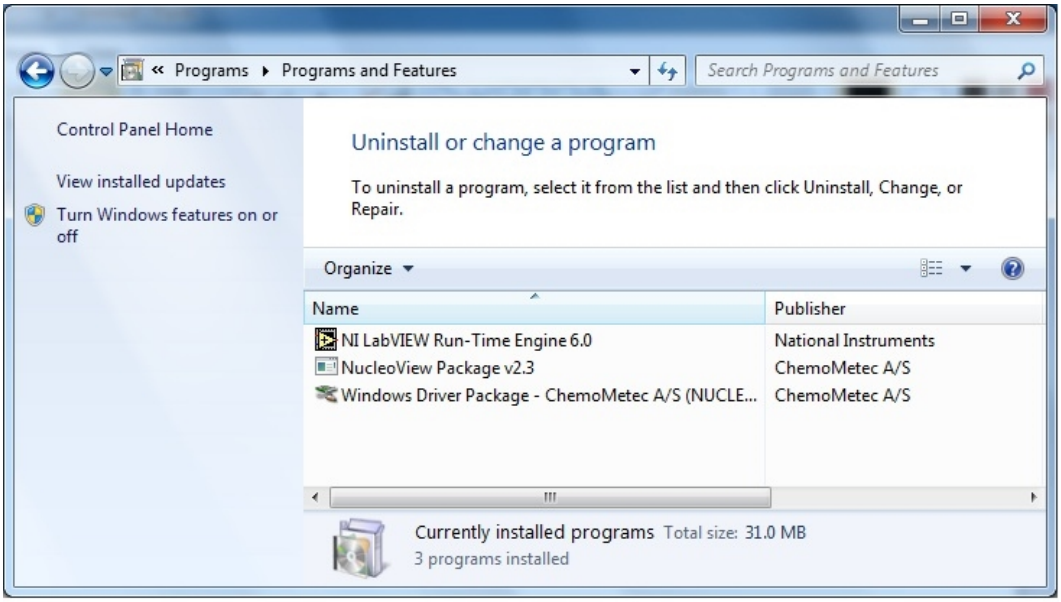

Figure 10 Remove all components.

Via the start button select programs – Control Panel – Programs and Features. From the list you can locate the three components listed above and by a right click on the mouse you are able to uninstall the components individually.

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[8] If you at any time created your own Default settings you will also have a file called "NV default settings.ini".

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2 Getting started with NucleoView™

NucleoView allows the user to register the results of cell counts with the NucleoCounter on a computer. This is useful when documentation of results and data is needed.

2.1 Launch NucleoView™

NucleoView can be activated through the Windows “Start” menu. First select “NucleoView 2.2” from the “Programs” submenu, and then select “NucleoView”.

When NucleoView is launched, both NucleoView and the NC Receiver appear on the task bar. The NC Receiver is an application, which must be active when receiving data from the NucleoCounter. NC Receiver is launched automatically when NucleoView is launched.

Data from NucleoCounter can only be transferred if the NC Receiver is active, therefore it must not be closed.

2.2 Selection of program mode

When NucleoView is launched for the first time, the window shown in figure 11 appears.

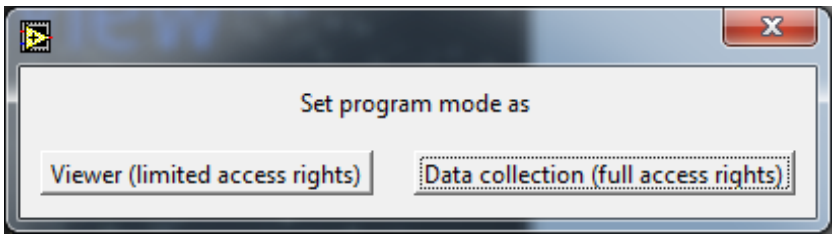

Figure 11 This start-up window allows for selection of the appropriate program mode.

You shall now select “Data collection” mode[9] for your NucleoView installation in order to get full access rights. Once this program mode has been selected (click it using the mouse) NucleoView is ready for use.

[9] Data collection is the normal mode when an instrument is connected.

2.2.1 Viewer (limited access rights) mode

The Viewer mode is recommended for installations retrieving and viewing data over a data network – not for collection of data.

In this mode it is possible to retrieve and view all data from the CM files received from a NucleoCounter. It is possible to change all settings in the program, but it will not be saved to the files.

2.2.2 Data collection (full access rights) mode

The Data collection program mode is recommended for the computer directly connected to the NucleoCounter via the USB interface.

In this mode it is possible to modify and save settings and information in the CM file for later use. This mode is required if data has to be saved with the CM file.

The information, which can be saved, is:

- Multiplication factor (from the dilution of the sample)
- Operator (16 characters)
- Comments (45 characters)
- Data concerning the dilution
    - Exact amounts of Lysis buffer
    - Exact amounts of Stabilizing buffer
    - Exact amounts of sample
    - External dilution factor

2.2.3 Change program mode

In case you want to change the program mode follow the directions below.

1. Close NucleoView using the “QUIT” button on the Measurement tab.
2. Locate the folder “c:\nucleoview”.

3. Locate and delete the file “NV startup.ini” in the folder “c:\nucleoview”.

4. Launch NucleoView again, and you will have the possibility of making the choice of modes again.

DO NOT use this procedure to perform regular changes between Viewer and Data collection mode, since it can affect the ability to change parameters in your older files. Only use this procedure if you during initial startup accidentally chose the wrong program mode.

2.2.4 Configure text editor

First time the Viability- or Utilities tab is entered NucleoView will prompt to have a text editor configured. In the Viability tab click “OK” and continue the calculations (see chapter 5.3 Configure text editor) or proceed to the Utilities tab to configure a text editor (see chapter 6.7 Configure text editor).

2.3 Quit NucleoView™

To quit NucleoView click the “QUIT” button in the Measurement tab.

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3 The structure of NucleoView™

NucleoView is operated through a number of tabs. The different tabs are selected by clicking on the corresponding tab. The tabs are shown in figure 12.

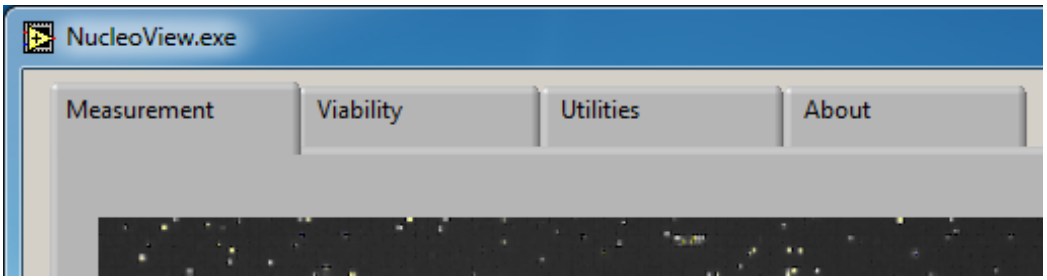

Figure 12 The four tabs of NucleoView.

The default startup screen is the Measurement tab. For detailed descriptions of the different tabs see the following chapters. An overview of the four different tabs is given below.

3.1 Measurement tab

This is where data from the CM file received from the NucleoCounter is presented. The presented data is: Concentration of cells in the NucleoCassette, Multiplication factor, Concentration of cells in the suspension, Operator, Comment. Parameters such as the multiplication factor, operator and comment can be changed and saved to the CM file (provided the Data collection program mode is chosen). Please refer to chapter 4 for a detailed description of each feature.

3.2 Viability tab

On the Viability tab it is possible to calculate the viability on basis of two files or measurements. Choose the files and NucleoView calculates the viability (%) and the concentration of viable cells in the original cell suspension. The result of the calculation can then be saved for printing or exporting purposes.

3.3 Utilities tab

Here it is possible to print and export data from a chosen session or from the saved viability calculations. The header of the prints and exports includes various information and may be customized through the Utilities tab. Furthermore it is possible to change the default settings for the measurements (multiplication factor etc.).

3.4 About

This tab displays version information of the NucleoView.

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4 Measurement tab

Measurement is the default start-up tab of NucleoView. In this tab, data and results transferred from the NucleoCounter are automatically displayed. An example of the Measurement tab is shown in figure 13.

Once NucleoView has been launched the Measurement tab is automatically displayed. Usually there are no files in the active session upon start-up. The object intensity histogram is then replaced with a small text box saying, “Please select a valid folder”. Start the measurements and the object intensity histogram will appear as in figure 13 or use the Browse button to navigate to another session (see chapter 4.5 Browse and Today).

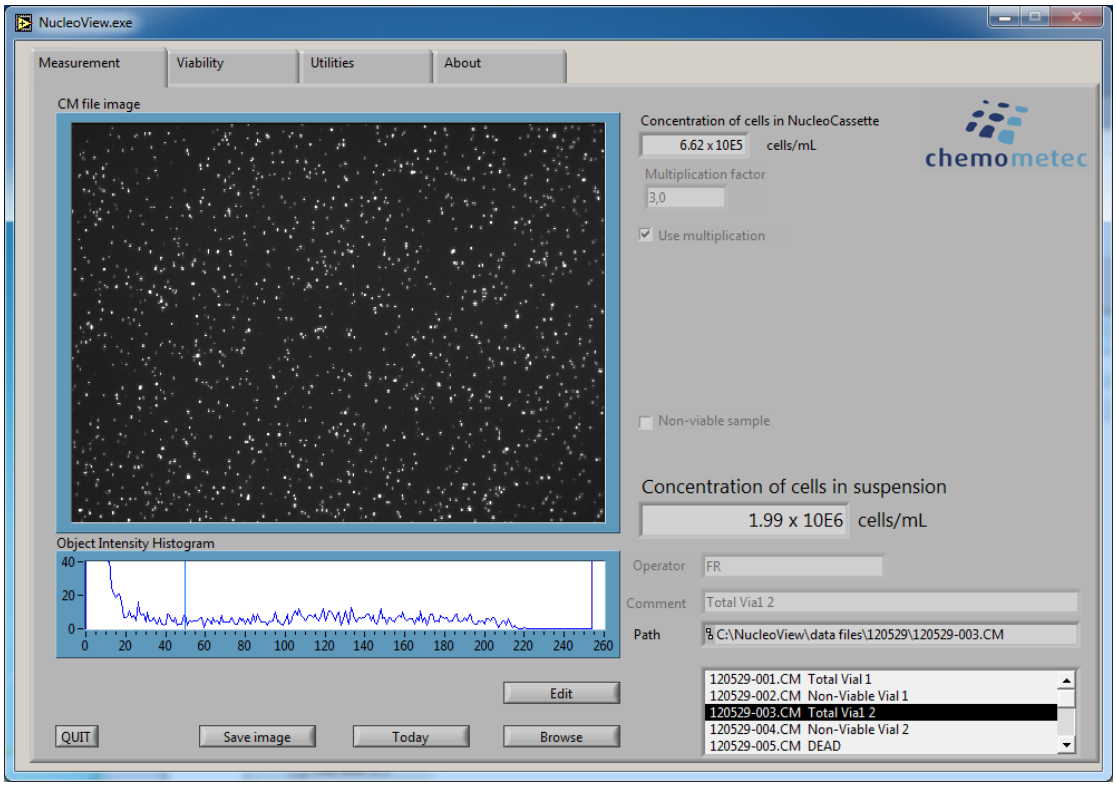

Figure 13 The NucleoView Measurement tab.

When results from cell analyses are received from the NucleoCounter, the cell concentration, image of the NucleoCassette chamber etc. are displayed. Each analysis is saved on the computers hard disk in a separate data file.

4.1 Concentration of cells in the NucleoCassette™

The estimated cell concentration of the cell lyzate in the NucleoCassette is displayed at the top of the window in a field labeled “Concentration of cells in NucleoCassette”. The cell concentration is given as cells per mL.

Most often the cell suspension has been diluted with the Lysis and Stabilizing buffers before the analysis. To obtain the concentration of cells in the original cell suspension the result has to be corrected using the multiplication factor (see chapter 4.2 Concentration of cells in suspension and Multiplication factor below).

4.2 Concentration of cells in suspension and Multiplication factor

The result of the cell counts adjusted for the appropriate dilutions (using the multiplication factor) is displayed near the bottom of the window using large types. The multiplication factor is used to correlate the observed cell concentration in the NucleoCassette to the concentration of cells in the original cell suspension. Thus, the concentration of cells in the NucleoCassette times the multiplication factor equals the concentration of cells in the original cell suspension.

When there is a checkmark in the field next to “Use multiplication factor” (as shown in figure 13) the multiplication factor can be changed manually. The multiplication factor can be changed after the CM file is received, either by entering a new value in the field below “Multiplication factor”. The factory set default value of the multiplication factor is 3 and changes to this is described in section 6.6 Default settings for NucleoViewTM.

When the checkmark next to “Use multiplication factor” is removed (see figure 14) it becomes possible to enter information about the various volumes used for the analyses. NucleoView then calculates the multiplication factor according to the formula shown below.

$$MF = MF_{EX} \cdot \frac{V_{LB} + V_{SB} + V_S}{V_S}$$

MF The multiplication factor used for correction of the concentration of cells in the NucleoCassette.

MF_{EX} The external multiplication factor e.g. from a pre-dilution.

V_{LB} The volume of the Lysis buffer.

V_{SB} The volume of the Stabilizing buffer.

V_S The volume of the sample.

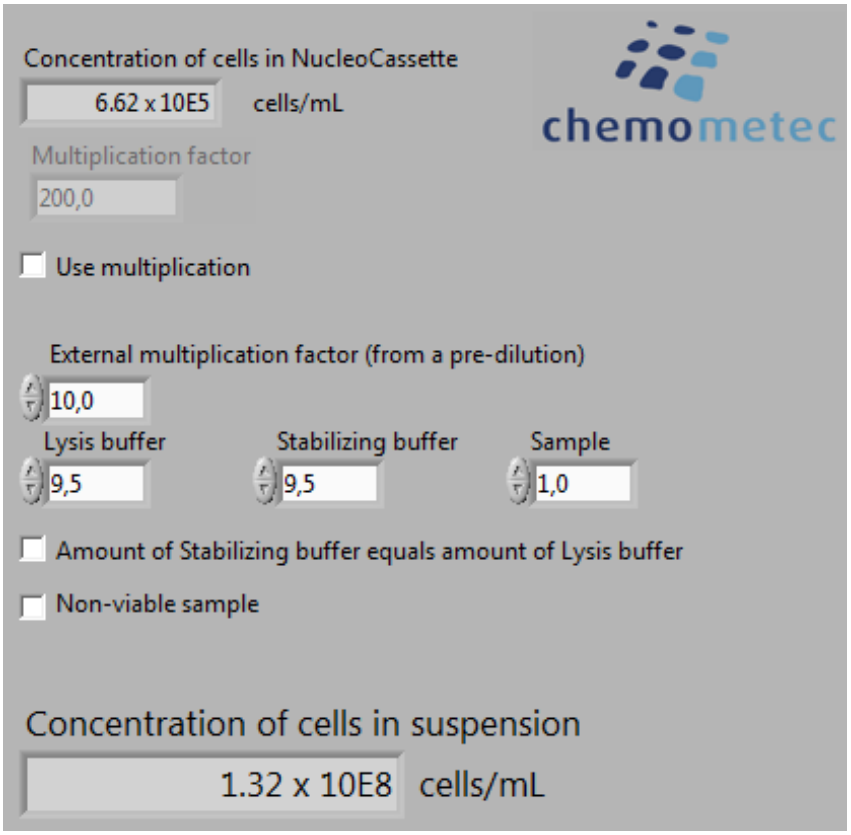

Figure 14 Data relating to the dilution of the sample.

All data regarding the multiplication factor is stored in the individual data file, thus making it possible to keep track of the individual settings for each analysis. Once NucleoView has received a new file from the NucleoCounter it automatically saves the default settings for the multiplication factor to the file. The factory set of default settings are:

- Multiplication factor = 3.0 (Valid range: 1.0-50000.0).
- Use multiplication factor[10], this feature is enabled by the default settings.
- External multiplication factor = 1.0.
- Volume of Lysis buffer = 1.0.
- Volume of Stabilizing buffer = 1.0.
- Volume of Sample = 1.0.

[10] This is enabled when the (✓) symbol, is present in the selection box.

- Amount of Stabilizing buffer equals amount of Lysis buffer[11], this feature is enabled by the default settings.

These default settings can be changed in the Utilities tab, see chapter 6.6 Default settings for details.

For further description of the multiplication factor and representation of the result refer to the NucleoCounter User's Guide.

4.3 Non-viable sample

The check button “Non-viable sample” is used for quick setting of the multiplication factor when analyzing non-viable samples. The initial default value for the multiplication factor for a non-viable sample is 1.0. The multiplication factor is changed to 1.0 by setting a checkmark using the mouse in the box to the left of the text Non-viable sample.

The multiplication factor for non-viable samples can be changed manually. Once this multiplication factor is changed, the new value is stored and becomes the default multiplication factor for non-viable samples as long as NucleoView is *running*. This is useful when e.g. Yeast samples are analysed, since pre-dilution of e.g. 50-500 is common.

4.4 Operator and Comment

To increase the manageability of the files, it is possible to attach information to each file in the operator and comment fields (both fields are empty at start-up). To do this, activate the operator or the comment field by clicking it using the mouse. The operator or comment can then be entered. The operator and comment fields are limited to 16 and 45 characters respectively. If a comment longer than 45 characters is entered in the comment field text will be truncated. The entered comment and operator will automatically be saved in the CM-file.

Once a comment or operator has been entered and saved to a file, they will be transferred to the next files when new data is transmitted from the NucleoCounter. The entered operator will be transferred as it is, while the entered comment will be transferred with three question marks, “???”, appended as a reminder that this comment has not yet been modified.

If typing the comments or operator settings is in progress during the transfer of new data from the NucleoCounter, the new file will be presented once the typing has ended.

[11] This is enabled when the (✓) symbol is present in the selection box.

4.5 Browse and Today

Each day NucleoView is launched in data collection mode, a new session (folder) is created. The folder's name is the date of the session (yymmdd). When NucleoView is launched in the Viewer mode, no new folder is created. It is possible to browse sessions created at an earlier date in both program modes.

When a measurement is performed and data is transferred to the computer, a file containing the measurement data is created. The file is shown in the file list, and the file name consists of the date of analysis and a running number as "yymmdd – nnn", see figure 15.

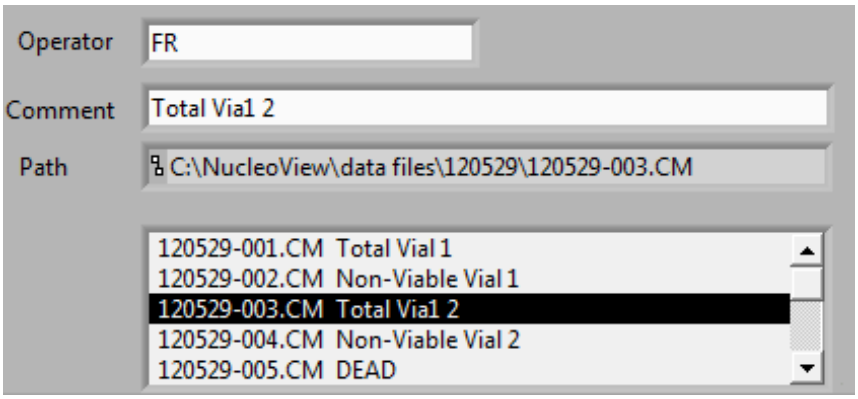

Figure 15 The file list (bottom), Path -, Comment - and Operator field.

It is possible to browse through files or results from the active session. To select and view a file from the list use the mouse. The selected file will be displayed including all manually entered data such as operator, comment and multiplication factor.

To browse files from other sessions, follow the directions below.

1. Click the "Browse…" button. This activates a dialogue where another session, which is to become the active session, can be located.

2. Use the mouse to double click on the session or highlight it and click "Open".

3. Click "Select Cur Dir" and the session will be displayed in NucleoView. The files will appear in the file list and it is possible to browse through them using the mouse.

The selection of a session from a previous date does not affect the transmission of data from the NucleoCounter. If the active session is not the session of the day when data is

received from the NucleoCounter then NucleoView detects this and changes the active session to the session of the day, displaying the most recent results.

The "Today" button will also change the active session to the current days session.

If comments have been added to a file this will be shown next to the name of the CM file eg. File 040831-054.CM from figure Figure 15 has the comment “Total Vial 2” and is shown next to the file name. This makes it easy to navigate and locate the files.

4.6 Storage of data

Using NucleoView it is possible to save information about the operator, data relating to the dilution of the sample and general comments. In figure 14 the data relating to the dilution of the sample is presented.

The settings described in chapter 4.2 are automatically stored to the CM-file when another file is transferred, another tab is activated or when the program is shut down using the Quit button. Do not shut Windows down without shutting NucleoView 2.2 down first or you may loose date entered in the very last file shown.

4.7 Changing data in CM-files

The data transferred from the NucleoCounter to the computer is stored on the computers hard disk as a file with type identifier (file extension) “.CM” (e.g. 020511-007.CM). To preserve the integrity of the data NucleoView upholds strict rules for changing information in the file and NucleoView will detect if the contents of the file is changed using other applications. NucleoView also only allows the modification of a file while it is in its original position on the hard disk.

Information can be changed and saved only in CM-files, which are still placed on the original file path.

In other words; information can only be changed and saved on the computer that collected the files in the first place, and if the files have not been moved from their original folder.

Also modifying information is only possible if NucleoView is installed in the Data collection mode. When browsing through files from an earlier date an “Edit”-button appears to the left of the file list. Click the “Edit”-button to change and save data in the file highlighted in the file list.

When NucleoView is unable to change and save the information in the CM-file (e.g. on a computer where NucleoView is installed in the Viewer program mode), a small icon appears left to the path field as shown in Figure 16. Information can be changed but the changes are not saved to the file.

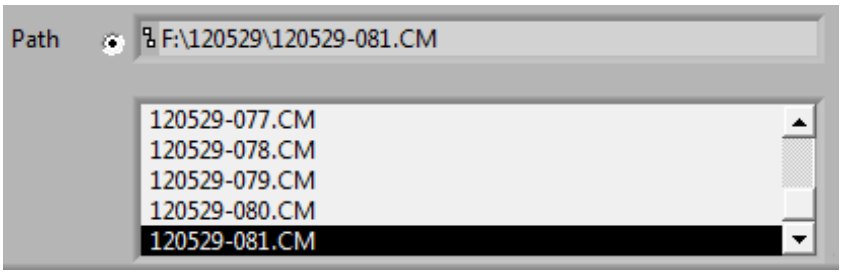

Figure 16 Data cannot be saved to the CM-file when the radio button is shown.

It is possible to have NucleoView installed on an office computer and then access CM-files placed on a laboratory PC used for data collection. However operated in this way it is not possible to save changes to any information in the CM-file. Figure 17 shows a possible configuration of computers on network when working with NucleoCounter.

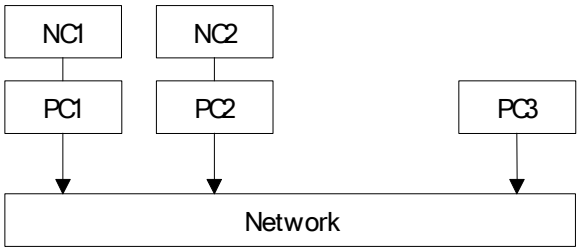

Figure 17 NucleoCounters and computers in a network.

NucleoCounter (NC1) is connected to PC1. NucleoView installation on PC1 is in data collection mode. NucleoCounter (NC2) is connected to PC2. NucleoView installation on PC2 is also in data collection mode. Both PC1 and PC2 are connected to the network. From PC1 CM-files on PC2 can be accessed but not changed and visa-versa. Data placed on PC1 can only be changed from PC1 and likewise for PC2. PC3 can access data on the other PCs but not save any changes to the CM-files.

When a CM-file is transferred to a computer and then presented in NucleoView, the default settings will be saved to the file. In case the default settings do not reflect the conditions for the analysis the operator *must* adjust these settings to reflect the appropriate values. When this file is opened the next time these modified settings will be presented. As discussed previously there are restrictions to how the settings can be changed. However it is always possible to open any file and adjust the multiplication factor to see the impact it has on the result.

When CM-files are not from today's session the "Edit" button is shown. This button must be activated before changes of the settings can be made, however these changes will only be saved to the CM-file if accessed using the PC that collected the files, and if the files still remain in their original folder. The files cannot be changed from other PCs or if they have been moved from the original folder.

It is not possible to save changes to CM-files generated with versions of NucleoView prior to version 2.0.

4.8 Collected image and Object intensity histogram

When estimating cell concentration with the NucleoCounter, an image of the stained nuclei in the NucleoCassette chamber is collected. This image is transferred to the computer and NucleoView presents it. The image bears resemblance to a starry sky, where each of the stars is a stained nucleus. Figure 18 shows an example of an image displayed by NucleoView.

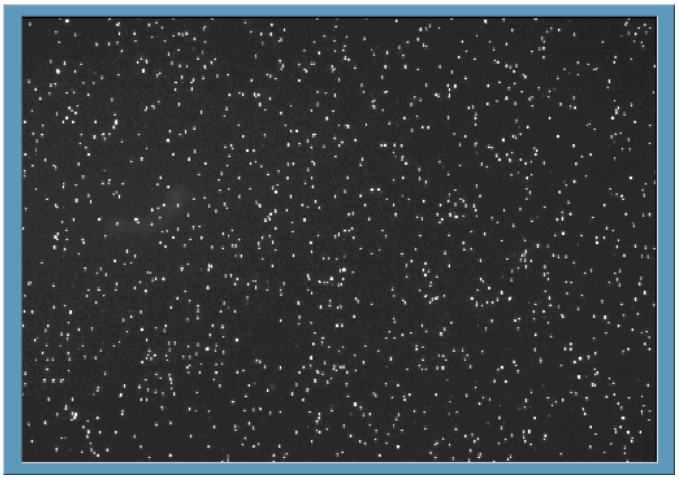

Figure 18 NucleoView presents the images from the NucleoCounter like a starry sky. Each "star" represents a stained nucleus.

The NucleoCounter records the image of the NucleoCassette chamber when counting cells. The image is then processed in the build-in computer of the NucleoCounter to give an estimate of the cell concentration, counting every star or white dot as a nucleus.

The object intensity histogram is shown below the image of the nuclei, see figure 19 The object intensity histogram. It shows the distribution of the number of fluorescence signals (particles) as a function of the intensity of the objects. In other words, the unit of the x-

axis is signal intensity on a scale from 0 to 255. The unit of the y-axis is the number of stained nuclei or objects.

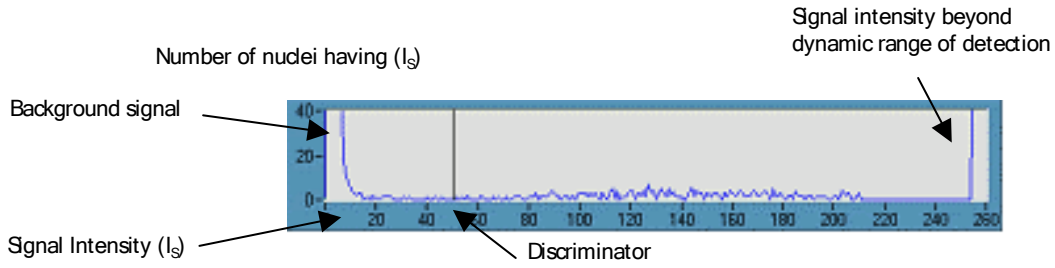

Figure 19 The object intensity histogram.

Variation of the background level of the image gives rise to a number of signals of low intensity. These signals are found to the far left of the object intensity histogram, generally the intensities of the background signals are below 20.

The stained nuclei give rise to higher signal intensities, which is illustrated in figure 19, as the rise in the number of signals to the right of the background signals. There is often an increase in the number of signals at the maximum intensity (255). These signals originate from stained nuclei having fluorescence intensities, which exceed the dynamic range of the detection system. These nuclei are also included in the counts.

Since there can be fluorescent objects with intensities which are only slightly higher in intensity than features in the background it is necessary to determine a limit, which discriminates between signals from a nuclei and signals from objects in the background. At the signal intensity of 50 the discriminator is placed. The discriminator defines that any object with signal intensity above 50 is included in the cell count, while signals of intensities below 50 are regarded as background signals.

Due to the high signal to noise ratio of the NucleoCounter, illustrated by the low frequencies of signals of intensities between 20 and 60, the position of this discriminator is not critical. When measuring samples of similar origin (mammalian cells) the fluorescence signals from all nuclei will have similar intensities since the DNA content of nuclei is similar. Thus the discriminator does not have to be adjusted individually for each sample.

4.9 Save image

The image of the NucleoCassette chamber can be saved for documentation purposes. How to save the image is explained below.

To obtain a copy of the image, use the "Save image" button. This will activate a standard save dialogue where the location and name of the resulting image is defined. The image is saved as a .BMP image, a graphical format, which can be used with variety of common applications, such as word processing software. The size of the BMP image is 140 KB and it looks as shown in figure 20.

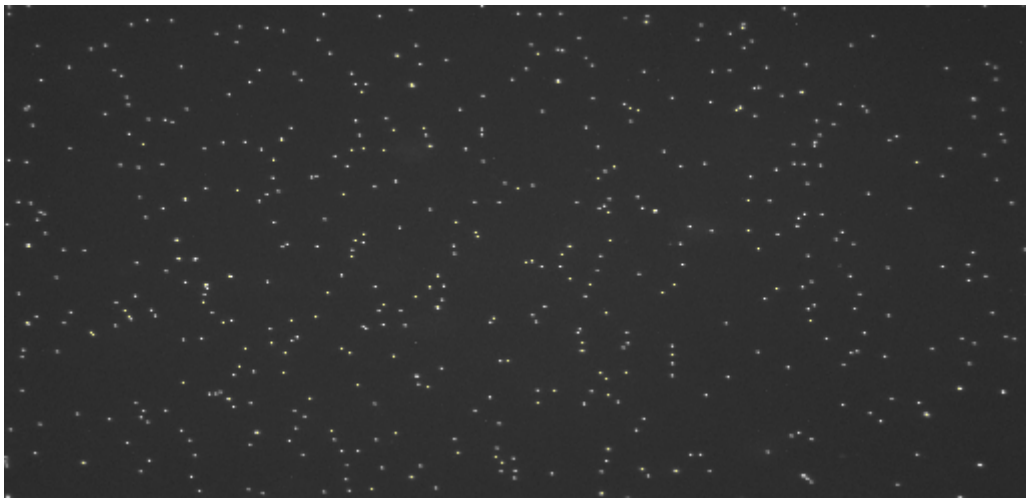

Figure 20 Image created with “Save image” button.

The BMP image, which is saved, contains the actual data originally measured by the NucleoCounter. The actual image consists of 510 x 246 pixels representing an area of 4.8 by 3.6 mm. This area has the ratio 4:3 (width: height). However the ratio of the pixels width and height is 2.07:1 and therefore the image created by the button “Save image” will look like figure 20 when viewed as a BMP image. To restore the appropriate aspect ratio of the image it is therefore necessary to scale the image. The following is an example of the scaling of an image using Microsoft Word.

1. Click on the image using the right mouse button. This activates a pop-up menu.
2. Click on “Format Picture…” and the dialogue shown in figure 21 appears.
3. Click the “Size” tab in the dialogue.
4. Remove the “Lock aspect ratio” checkmark by clicking it.
5. Adjust the Height to 155 and the Width to 100. Usually only the Height needs to be adjusted.
6. Click OK, and the image will look as shown in figure 22.

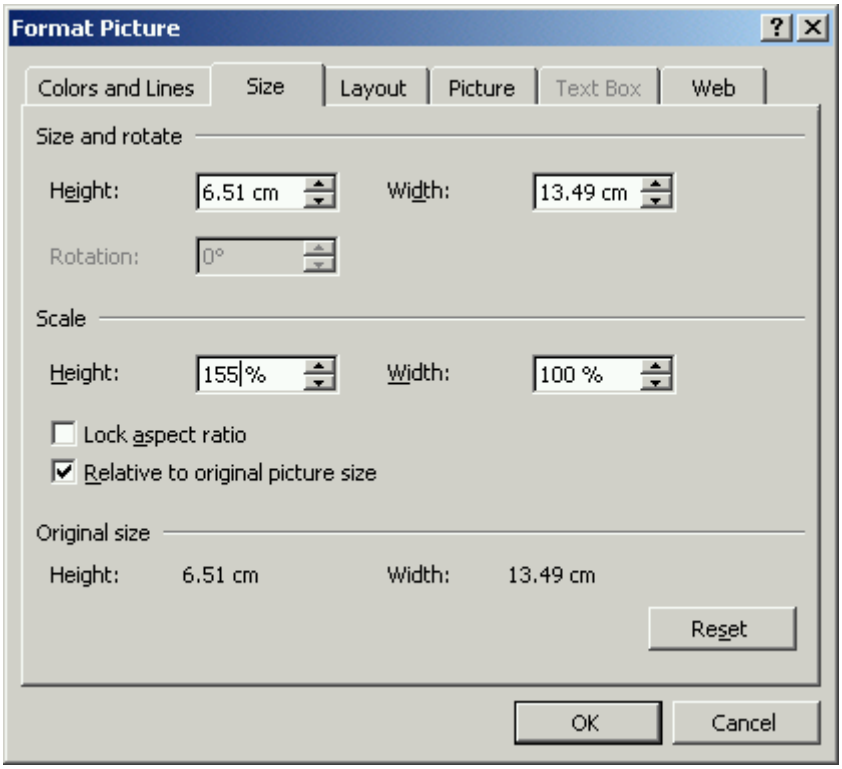

Figure 21 Resizing the image created using the "Save image" button.

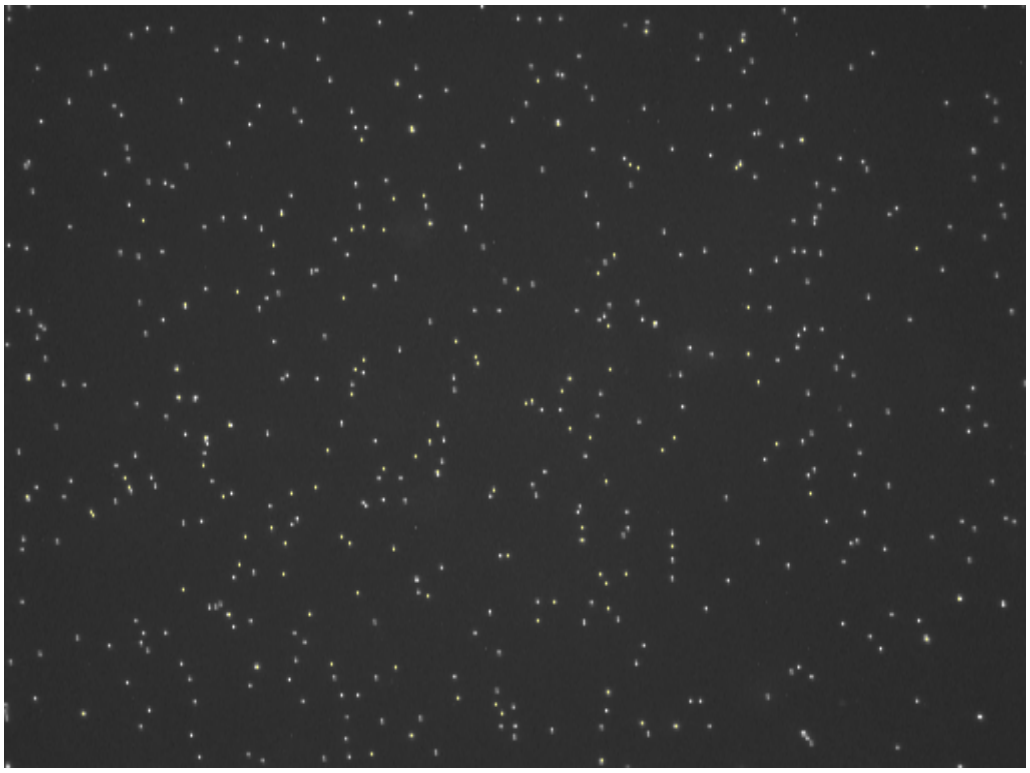

Figure 22 Resized image created using the "Save image" button.

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5 Viability tab

NucleoView offers the possibility of automatic calculation of the percentage and concentration of viable cells. The results can be saved and printed or exported later (see chapter 6 Utilities). The Viability tab looks as shown in figure 23. The upper and middle parts of the Viability tab consists of data relating to the total cell count and the non-viable cell count, respectively. The results of the viability calculations are found in the bottom of the tab.

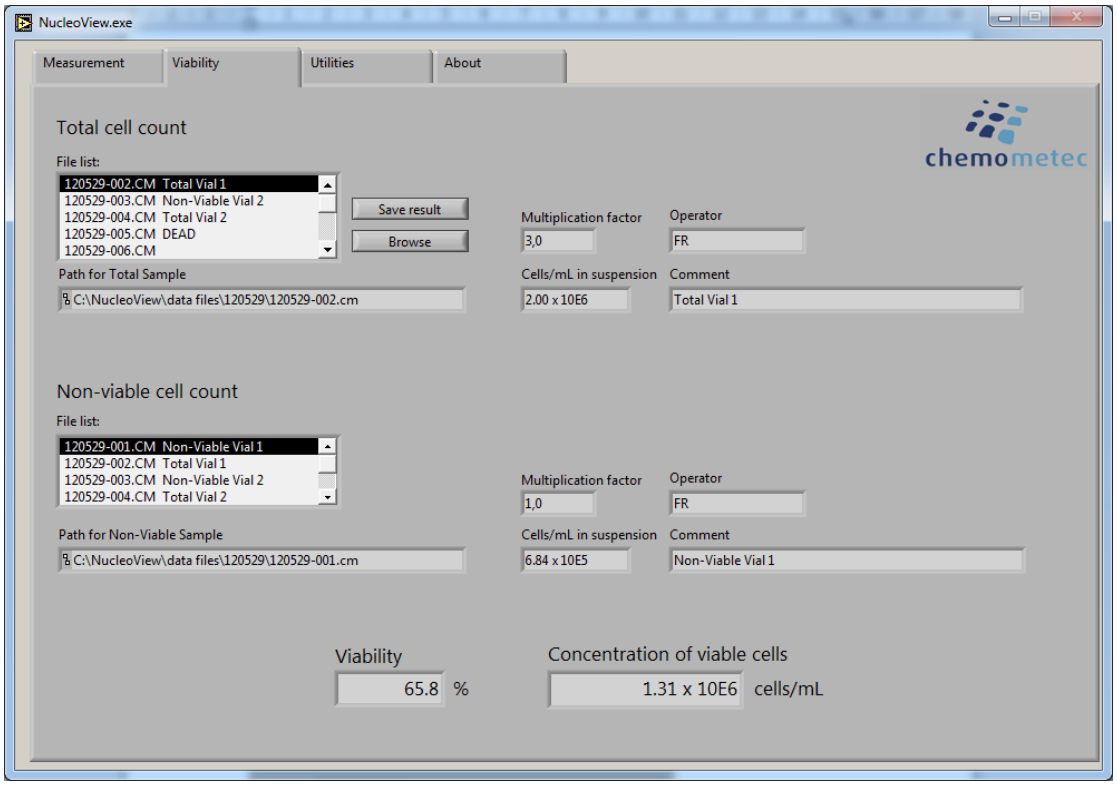

Figure 23 The Viability tab.

5.1 Choose files for the calculation and Save result

To the far left of the Viability tab are the file lists and the paths for the total and non-viable samples. The paths show the location of the files, which are being browsed.

In the file lists it is possible to browse through the files of the current session to choose which file is to be used in the viability calculation as the total cell count. Once the file for Total cell count is selected NucleoView suggest that the sample analyzed *prior* to this to be the non-viable cell count. The result of the viability calculation is shown immediately. However, it is possible to choose another file for the non-viable cell count by selecting a different file from the non-viable file list. Again, the result of the viability calculation is

shown immediately. If comments have been added to a file this will be shown next to the name of the CM file.

If the first file is selected as the total cell count, no file is automatically selected as the non-viable cell count and only the file list is shown (the information about the file and the results disappear). Select the file that is the non-viable cell count manually and the information about the file and results will be shown.

If the same file is chosen for the total cell count and the non-viable cell count an error message will appear, see figure 24. Click OK and NucleoView changes the non-viable file to be the file prior to the file chosen for the total cell count.

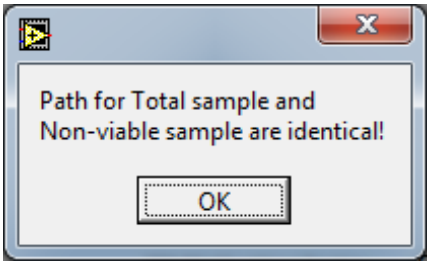

Figure 24 Error message when the same files are chosen for the total and non-viable cell count.

To choose results from another session click the “Browse”-button. Follow the directions described in chapter 4.5 Browse and Today.

To save the result of a viability calculation click the “Save result”-button. The result will be saved instantly and no dialogue will pop up. After a result has been saved it is not possible to delete it again.

The saved results can be printed or exported to a tabulator-separated file, which can be imported into several applications such as word processors or spreadsheets. See chapter 6.3 Viability report and chapter 6.4 Import data into Excel.

5.2 Viability (%) and Concentration of viable cells (cells/mL)

Once the two files have been selected for the viability analysis the results are present at the bottom of the Viability tab. The results are the Viability in % and the Concentration of viable cells in cells/mL. The representation of the results of the calculations follows the result mode for the total cell count. The result mode is selected on the NucleoCounter. Please refer to the NucleoCounter User's Guide for further information on result mode and calculation of viability.

The representation of the "Concentration of viable cells" follows the result mode used for the total cell count. However the calculations of Viability and Concentration of viable cells are based on the full precision results from the analyses and not the truncated values, which are present when result mode 2 or 4 are active.

Therefore, when the NucleoCounter is set to result mode 2 or 4 the displayed results of the viability calculations may differ from manually calculated results.

If the viability cannot be calculated the result fields will show "**N/A**".

5.3 Configure text editor

When the Viability tab is opened for the first time, the following message box appears.

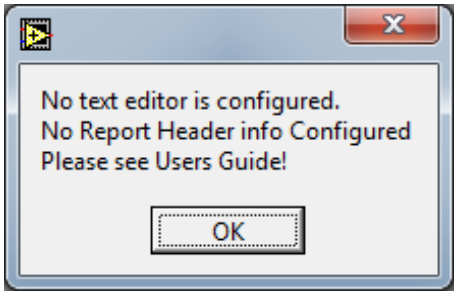

Figure 25 No Text editor configured.

Click OK and continue your analysis or go to Utilities page for setting up a text editor (see chapter 6.7 Configure text editor).

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6 Utilities tab

In the Utilities tab (figure 26) it is possible to print and export results and viability calculations. Furthermore it is possible to change the information on the header of the prints and exports, and the default settings, which are used when data from the NucleoCounter is received in the Measurement tab.

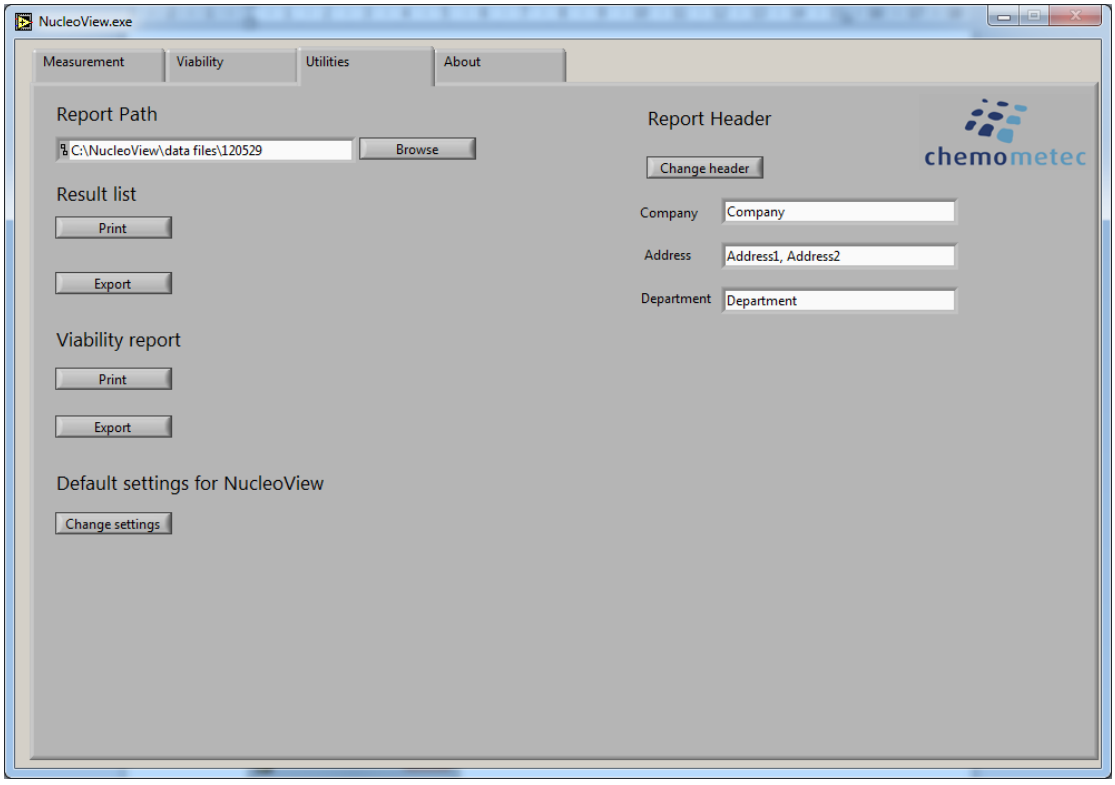


Figure 26 The Utilities tab.

6.1 Report path

The report path is found in the top left corner of the Utilities tab. It shows which session forms the basis for the result list and viability report. The active session can be changed by clicking the “Browse”-button, see chapter 4.5 Browse and Today for instructions.

6.2 Result list – Print and export

The result list contains information about the files in the active session. The information in the result list is:

- File name
- Concentration of cells pr. mL in the NucleoCassette (Cells/ml (cass.))
- Multiplication factor (MF)
- Concentration of cells pr. mL in the original cell suspension (Cells/ml (susp.))
- Comment
- Operator

On the top of the result list a header is printed. The header consists of the location, and name of the exported file is printed path is printed, company information, address, department, NucleoCounter serial no., print date and print time are also part of the header. It is possible to change the information about company, address and department, see chapter 6.5 Report header.

The results from the active session can be printed (using the default printer) and/or exported by clicking the “Print”- or the “Export”-button, respectively. Follow the directions below.

1. When the “Print” or the “Export” button is activated a file list and three further buttons appear, see **Error! Reference source not found.**

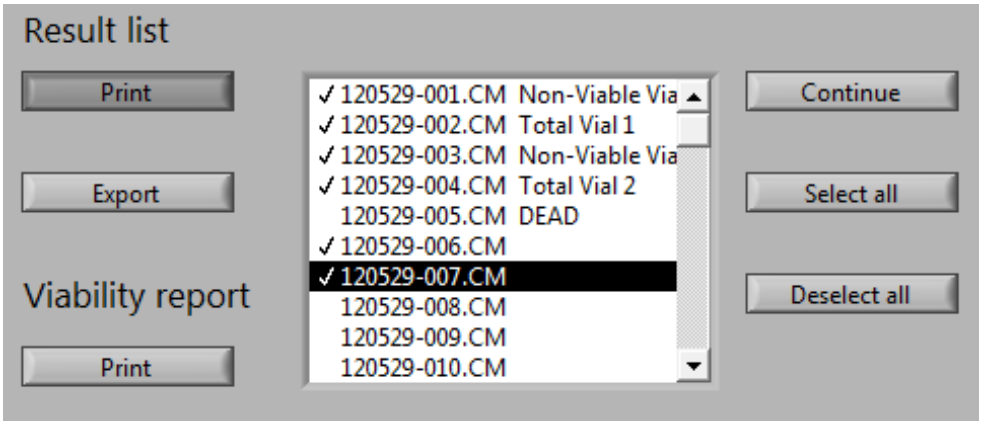


Figure 27 When the “Print”- or “Export”-button is activated a file list and three further buttons (“Continue”, “Select all” and “Deselect all”) appear.

2. Select the files to be included using the “Select all”- or “Deselect all”-button or use the mouse to select and deselect files individually (using a single click). Once a file is selected, a small check mark (✓) will appear next to the file. If only one file is present in the list, you must use the “Select all”- or “Deselect all”-buttons.

3. When the appropriate files has been selected, click “Continue” and the files will be printed or exported. If many files are printed or exported it can take a few minutes. For an example of a printed result list, see Appendix 1: Printed result list.

When files are exported a message box appears, see figure 28. The message box shows the location and name of the exported files.

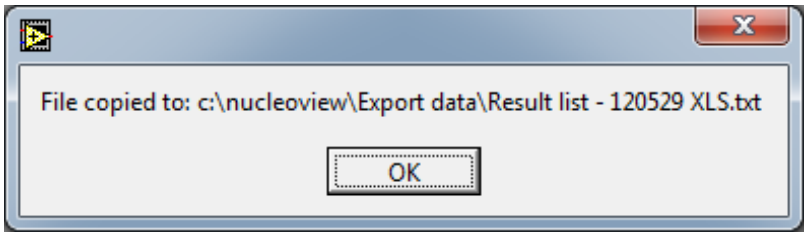


Figure 28 The message box displayed when files are exported. It shows the location and name of the exported file.

When exporting a result list the data from each file is arranged in a single line, separated by a tabulator character. Thus, the data is placed in columns for easy processing using a spreadsheet solution.

The files can also be printed and exported at the same time. To do this, activate both the “Print” and “Export” button before selecting the files. When “Continue” is clicked the files will be printed and exported simultaneously.

6.2.1 Location and name of exported files

When files are exported one file containing the exported data is generated. All files generated for export are present in the folder:

C:\Nucleoview\Export data

All the generated reports are present in this folder. In order not to have more than one version of the same data a report file is simply deleted if a new is generated from the same session. Both the exported viability report and the report list are placed here.

The filenames of the exported files are composed of a prefix, a middle and a last part. The prefix is Result list -, the middle part is the date of the session from which the data is exported, YYMMDD, and the last part of the filenames, XLS, indicates that the file can be imported into e.g. MS Excel (see chapter 6.4 Import data into Excel). The files are tabulator-separated txt-files. Because of the individual filenames all exported files can be stored in the same directory.

An example of a filename for an exported file is:

Result list – 021030 XLS.txt

The message box, which appears when files are exported, shows the location and name of the exported file, see figure 28.

6.3 Viability report – Print and export

The viability report contains information about the saved results of viability calculations for the active session. The information in the viability reports, both the print and the export, is:

- Filename for the total and non-viable cell count, TC and NV respectively.
- Concentration of cells pr. mL in the NucleoCassette for the total and non-viable cell count (Cells/mL (cass.)).
- Multiplication factor (MF) for the total and non-viable cell count.
- Concentration of cells pr. mL in the original cell suspension for the total and non-viable cell count (Cells/mL (susp.)).
- Comment and operator for the total and non-viable cell count.
- Concentration of viable cells/ml.
- Percentage of viable cells.

As for the result list a header is printed at the top of the viability report. The header contains the same information with one exception. Instead of the print date and time, the creation date and time is included in the header. The creation date and time refers to the date and time the first viability calculation for that particular session was saved. The information about company, address and department can be changed, see 6.5 Report header. Please note that changes made to the header will affect both viability reports and result lists.

The viability report can be printed (using the default printer) or exported by clicking the "Print" or the "Export" button. All of the saved results are printed or exported when the buttons are activated, it is not, as for the result list, possible to choose which results are to be printed or exported.

Once a viability report is exported a message box appears. The message box shows the location and name of the exported report, see figure 29. If Excel is processing the file from the session to export data from, the export operation cannot be completed.

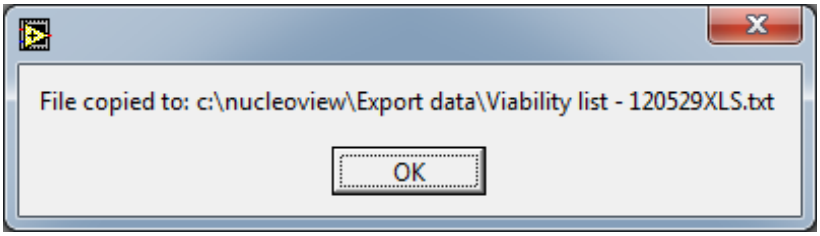


Figure 29 The message box displayed when a viability report is exported. It shows the location and name of the exported file.

When exporting a viability report the data from each analysis is arranged in a single line, separated by a tabulator character. Thus, the data is placed in columns for easy processing using a spreadsheet solution.

For further details on the location and name of the exported file see the chapter below.

If no viability data is saved in the selected session a message box will appear, see figure 30. Check if the active session is correct and then try again. If the active session is correct there is no viability data saved. Go to the Viability tab and save the needed viability calculations.

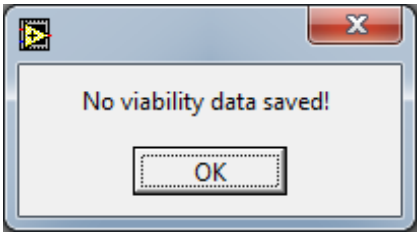


Figure 30 No viability data present for the selected session.

6.3.1 Location and name of exported files

The exported viability reports are saved in the same folder as the result lists:

C:\Nucloeview\Export data

The file names of the viability reports are constructed in the same manner as the result lists. The first part is a prefix, Viability list -, the middle part is the date of the session from which the viability report is derived, YYMMDD. The last part is “XLS” to indicate that the file can be imported into e.g. MS Excel. The exported viability reports are tabulator-separated txt files.

An example of a filename of an exported viability report is:

Viability list – 021030 XLS.txt

The message box, which shows the location and filename of the viability reports, is shown above in figure 29.

6.4 Import data into Excel

To import the exported files into Excel follow the directions below. The directions are identical for result lists and viability reports.

Before importing the first files make sure that the decimal symbol is set to “.” (period). If the decimal symbol is “,” (comma) then Excel cannot use the imported data for calculations. See chapter 6.4.1 Set decimal symbol for directions of how to change the decimal symbol.

1. To import a report first open Excel and activate the “Open…” function e.g. through the file menu. The dialogue shown in figure 31 appears.

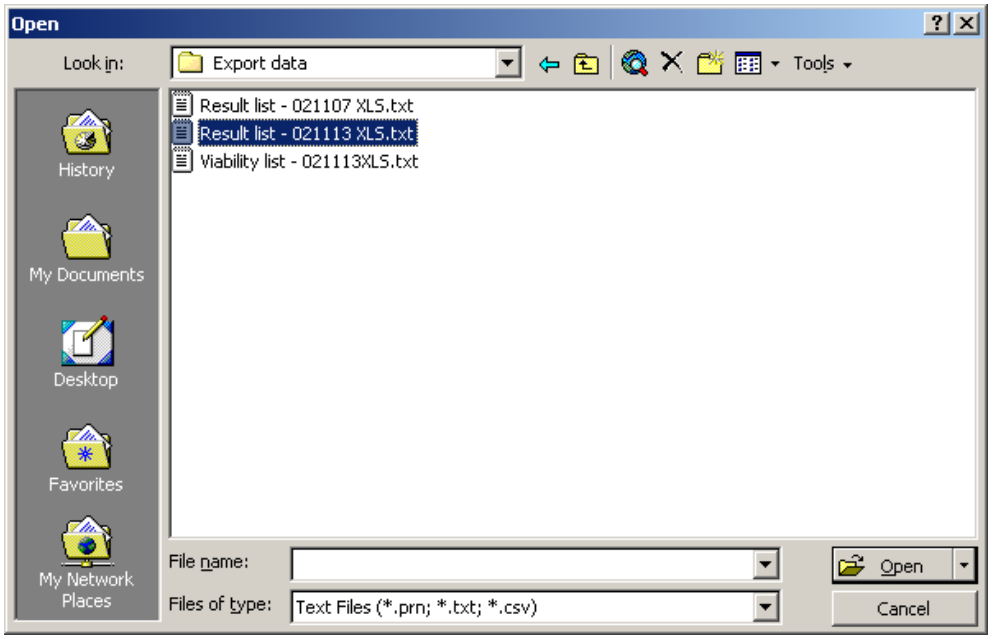


Figure 31 Open Excel

2. Locate the file to import in the directory: "C:\Nucleoview\Export data" and open it (doubleclick it or highlight it and click the “Open” button). If you cannot find the file in the directory ensure that you have selected “Text Files” in the "Files of type" field at the bottom of the dialogue. Once the txt-file has been opened the Text Import Wizard appears, see figure 32.

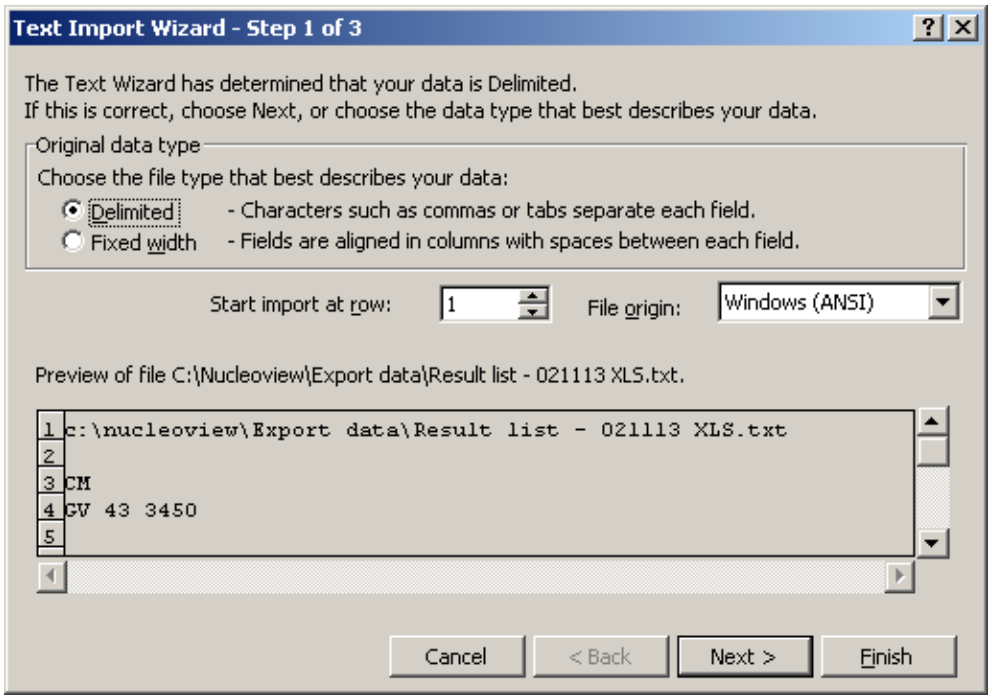


Figure 32 Import to Excel step 1.

3. Check that the radio button under “Original data type” is set to “Delimited”. If this is the case, click “Finish” to import the file.

   If the radio button is set to “Fixed width” change it to “Delimited” (using the mouse). Click “Next” and a new dialogue will appear, see figure 33.

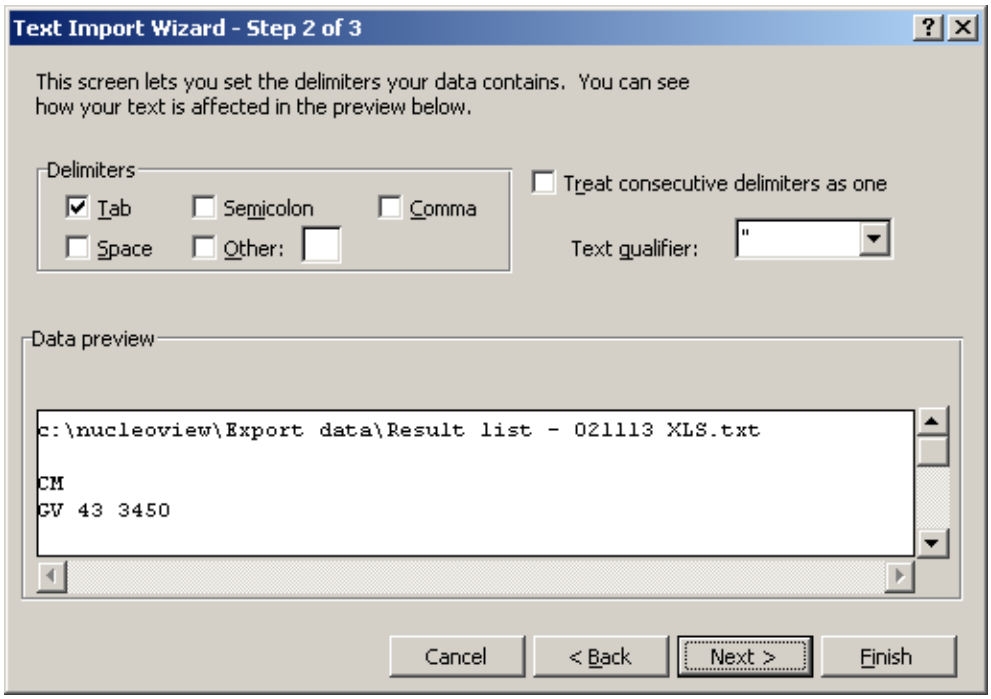
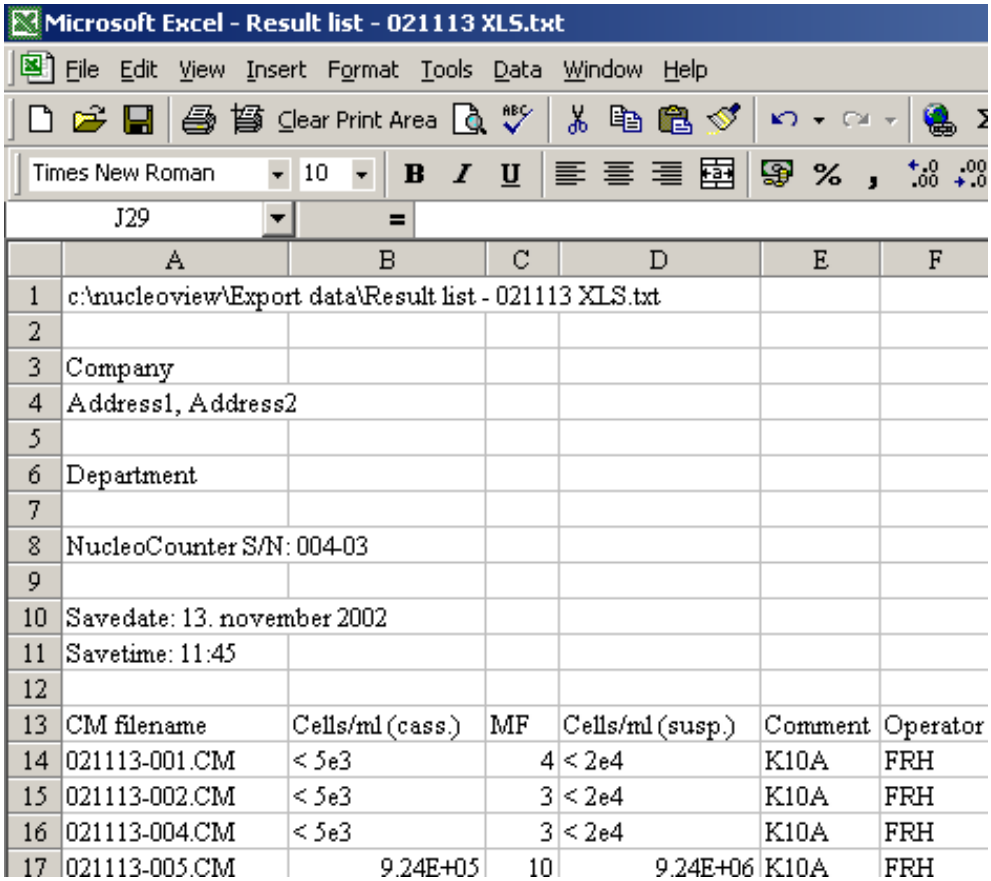


Figure 33 Import to Excel step 2.

4. The delimiter must be set to “Tab” as shown in figure 33. This is normally the default setting for Excel. Click “Finish” to import the data into Excel.

5. Following this procedure above will omit the third step in the "Text Import Wizard" for Excel.

The file is now imported into Excel and ready for any further processing, see figure 34.



Microsoft Excel - Result list - 021113 XLS.txt

File Edit View Insert Format Tools Data Window Help

Clear Print Area

Times New Roman 10

J29 =

	A	B	C	D	E	F
1	c:\nucleoview\Export data\Result list - 021113 XLS.txt					
2						
3	Company					
4	Address1, Address2					
5						
6	Department					
7						
8	NucleoCounter S/N: 004-03					
9						
10	Savedate: 13. november 2002					
11	Savetime: 11:45					
12						
13	CM filename	Cells/ml (cass.)	MF	Cells/ml (susp.)	Comment	Operator
14	021113-001.CM	< 5e3	4	< 2e4	K10A	FRH
15	021113-002.CM	< 5e3	3	< 2e4	K10A	FRH
16	021113-004.CM	< 5e3	3	< 2e4	K10A	FRH
17	021113-005.CM	9.24E+05	10	9.24E+06	K10A	FRH

Figure 34 Data imported to Excel

6.4.1 Set decimal symbol

To import reports to Excel ensure that the decimal symbol used by Windows is set to "." (period) and not "," (comma). To check that the decimal point is correct or to change it, follow the directions below.

1. Open “Control Panel” through the Windows Start menu.
2. Select the "Region and Language" options.
3. When the Region and Language Options dialogue appears, click the “Formats” and the “Numbers” tab and the dialogue will look as shown in figure 35 (only part of the dialogue is shown).

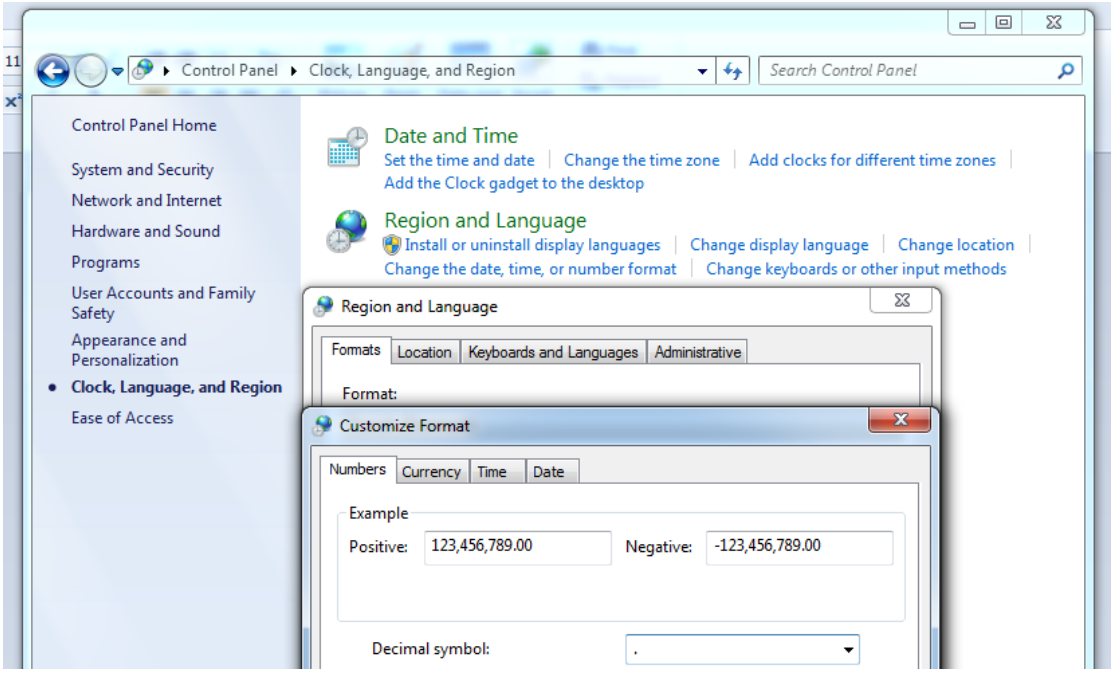


Figure 35 Setting decimal symbol using the Region and Language options in Windows Control panel. Only part of the dialogue is shown.

4. Check if the Decimal symbol is “.” (period), see figure 35. If the Decimal symbol is “,” (comma) change it to “.” (period).

The decimal symbol has now been set and import of files into e.g. Excel can start. Please note that changing the Region and Language options affects other programs.

6.5 Report header

The report header for the result lists and viability reports can be changed to fit individual companies and departments.

To change the report header follow the directions below.

1. Click “Change header”.
2. Change the fields Company, Address and Department.
3. Click “Save header” to save the changes.

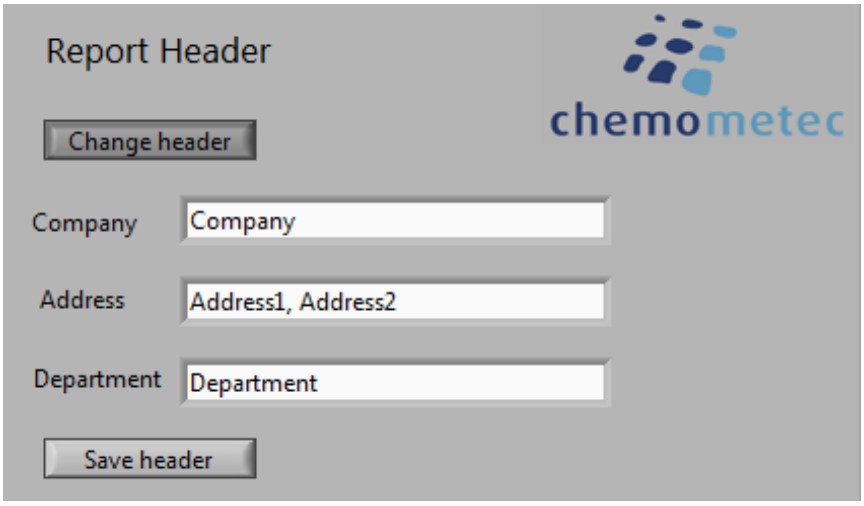


Figure 36 Report header information.

Changes made to the report header will affect the result lists, exported data and the viability reports. A viability report will only show the changed report header if it was changed before the first viability calculation of that particular report was saved.

6.6 Default settings for NucleoView™

NucleoView offers the possibility to change the default settings used when data is transferred from a NucleoCounter. See figure 37 for the settings it is possible to change.

1. To change the default settings click the "Change settings" button. This will activate a dialogue as shown in figure 37.
2. Customize the settings.
3. Click "Save settings". The new settings will be used for the next transfer of data from a NucleoCounter.

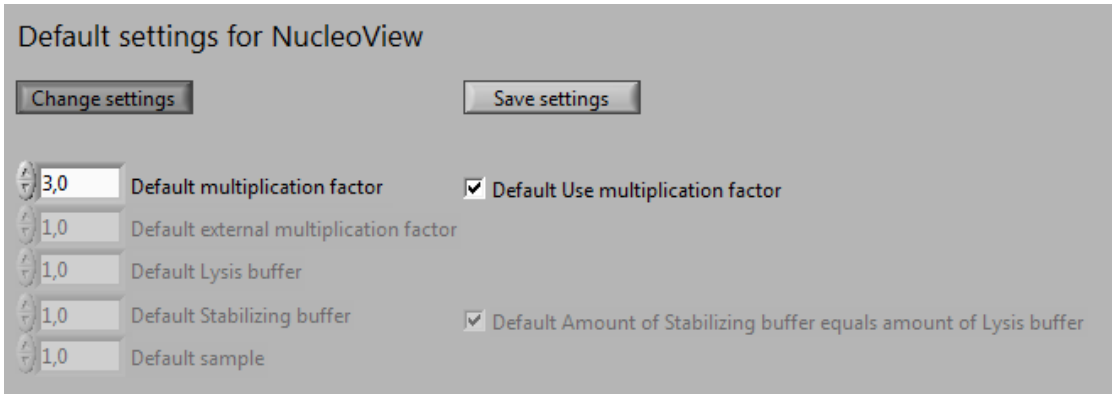


Figure 37 Customized default settings for NucleoView.

Next time the "Change settings" button is activated, the new default settings appear again. The old default settings will appear only if NucleoView has been closed down and launched again, this can be used as an easy method to change the settings back to the factory set default values (by clicking "Save settings").

If the "Change settings" button has been activated but no changes are to be made, click "Change settings" again. The possibility to change settings will disappear and no changes are made.

When NucleoView is restarted, the settings shown in Figure 37 will revert to the factory set default values.

For users with a NucleoCounter YC-100 for analysis of samples with very high cell counts in e.g. Beer or Wine Yeast fermentation samples using only Reagent Y100, please refer to Appendix 3: Default settings for Yeast samples.

6.7 Configure text editor

It is necessary to configure a text editor to be able to print result lists and viability reports from the Utilities tab. Either WordPad or Notepad can be configured as text editor. Follow the directions below to configure a text editor.

1. When the Utilities tab is activated for the first time, the message shown in figure 38 appears. Click OK.

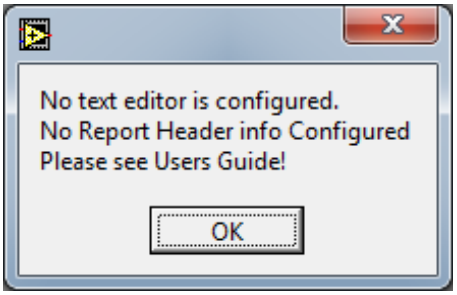


Figure 38 This message box is displayed when no text editor is configured and the Utilities tab is activated.

2. Activate the "Configure"-button below "Text Editor" in the Utilities tab.

3. Click OK when the message box shown in figure 39 appears.

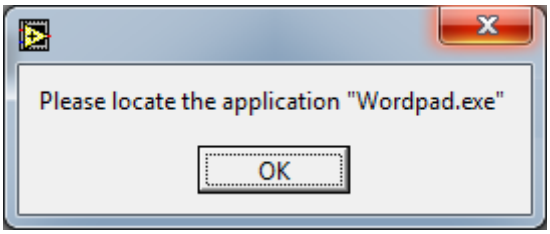


Figure 39 When this message box is displayed click OK and locate the WordPad application.

4. The dialogue shown in figure 40 appears. Locate “wordpad.exe”, highlight it (using the mouse) and click Open.

   Either WordPad or Notepad[12] can be configured as text editors. Therefore, if it is not possible to locate “wordpad.exe”, try to locate “notepad.exe” and open it instead of “wordpad.exe”. If neither “wordpad.exe” nor “notepad.exe” can be located use the search function of Windows Explorer to locate one of them or contact your administrator for assistance.

[12] Before Notepad can be selected as text editor the field “Files of type:” must be changed to “All files (*.*)”.

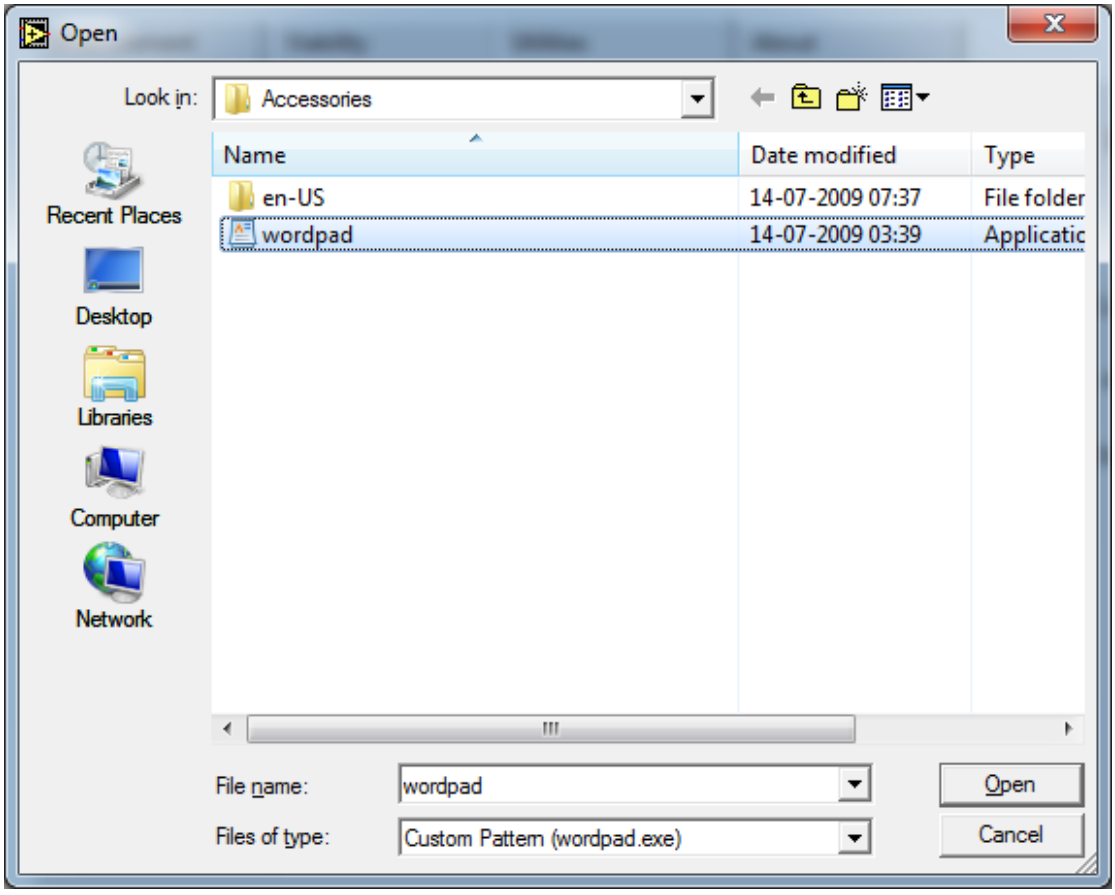


Figure 40 This dialogue appears when WordPad or Notepad is configured as text editor.

5. The message box shown in figure 41 appears when WordPad or Notepad has successfully been configured as text editor. Click OK and proceed to define the font for the text editor.

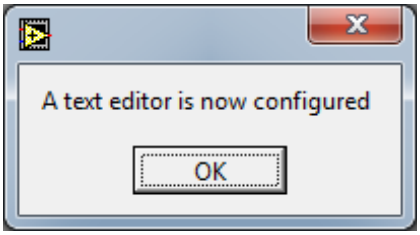


Figure 41 This message box indicates that a text editor has been successfully configured.

6.7.1 Define font for WordPad

To create reports with results in columns the font for the text editor must be defined. This is done differently for WordPad and Notepad. Follow the directions below to define a font for WordPad.

1. Open WordPad and select “Save as” from the File menu. The dialogue shown in figure 42 appears.

2. Choose the location to save the file to be the Nucleoview folder (usually C:\Nucleoview).

3. In the bottom of the dialogue change the “Save as type:” to “Text Document”. Make sure there is a checkmark in the check box to the left of “Save in this format by default”. If there is no checkmark present set it using the mouse.

4. Change the file name to “Document.txt”.

5. Click “Save”.

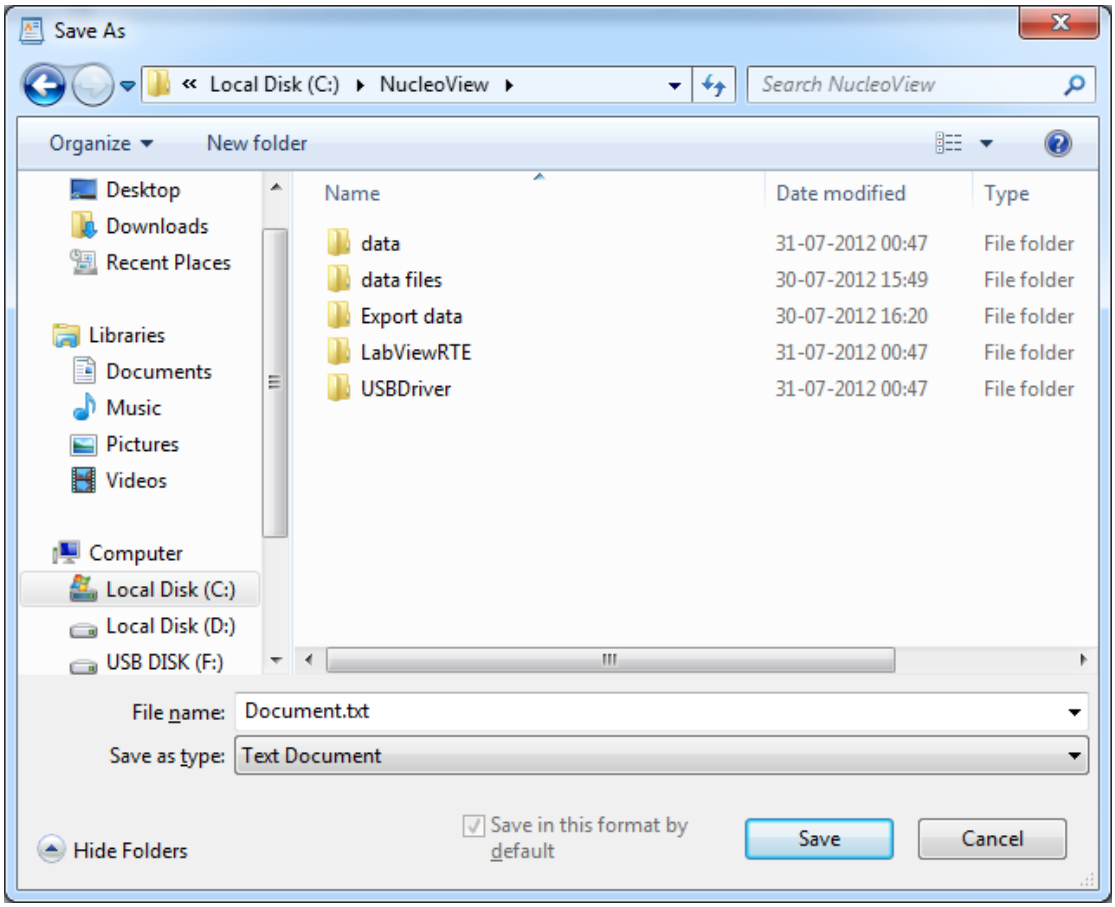


Figure 42 The dialogue shown when setting up the font for WordPad.

6. After clicking “Save” the dialogue shown in figure 43 appears. Click “Yes” and the font for WordPad is defined. Now WordPad will by default use the font Courier, size 10 regular.

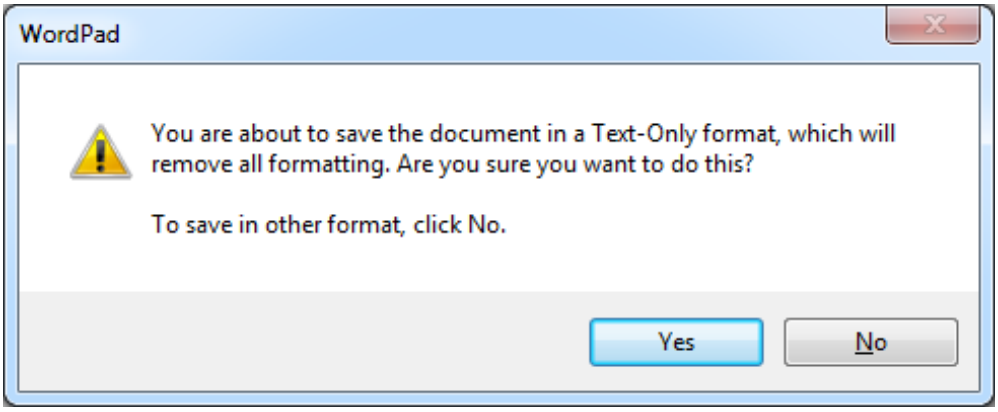


Figure 43 Confirm the setup for WordPad by clicking “Yes”.

7. Exit WordPad.

6.7.2 Define font for Notepad

Similarly Notepad can be set up for NucleoView, follow the directions below.

1. Open Notepad.
2. Choose “Font…” through the Format menu. The dialogue is shown in figure 44.

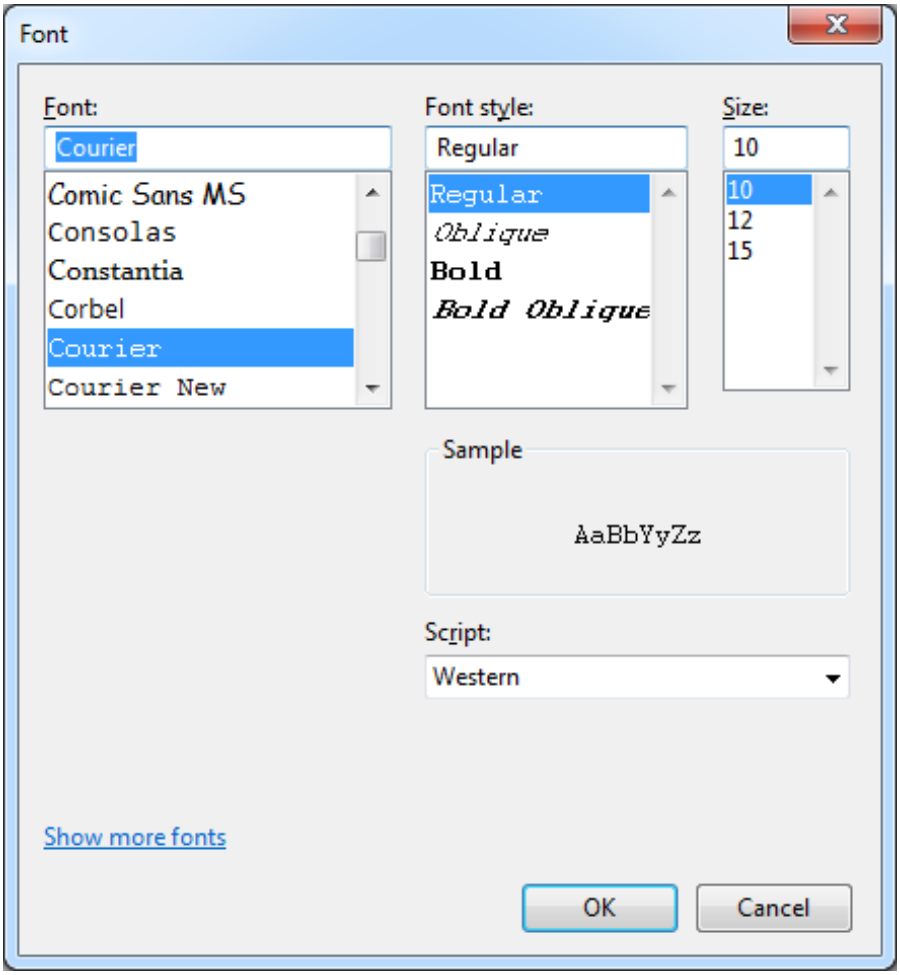


Figure 44 Defining the font for Notepad.

3. Choose the font Courier, Font style Regular and Size 10 as in figure 44. Now Click OK and exit Notepad. Notepad will use the font Courier, size 10 regular.

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7 Troubleshooting

Occasionally difficulties with the NucleoView software may occur. In the following a short description and suggested action is given.

7.1 Quality of the sample

Through the CM file image it is possible visually to assess the quality of the sample preparation and thereby the reliability of the measurement by looking at the “starry sky”. The measurement is reliable if the stars or white dots are distributed separately and evenly over the image. Naturally, this rule only applies when a representative sample has been drawn from the original cell suspension.

Be aware of large objects in the image. These objects can be external material, such as dust or a piece of hair. These objects are ignored in the image analysis, so the obtained result correctly reflects the number of individual cells in the sample. On the other hand, it is important to be aware of these objects, since if it is a cell aggregate or a clump this might indicate that the sample preparation has been sub optimal and the quality of the estimate of cell concentration may be compromised.

7.2 Data not received

If NucleoView does not receive data from the NucleoCounter, please verify that the USB cable is connected correctly to both the NucleoCounter and the computer. Then look at the task bar on the computer to see if the NC Receiver is started.

Figure 45 NC Receiver icon is Green on the Windows task bar when connected and Grey when disconnected

The NC Receiver must be started to receive data from the NucleoCounter. If the NC Receiver is not connected, quit and restart NucleoView. NC Receiver should then appear on the task bar.

Generally the NC Receiver has two states in the Instrument Status field when it is started:

- Not connected
- Connected

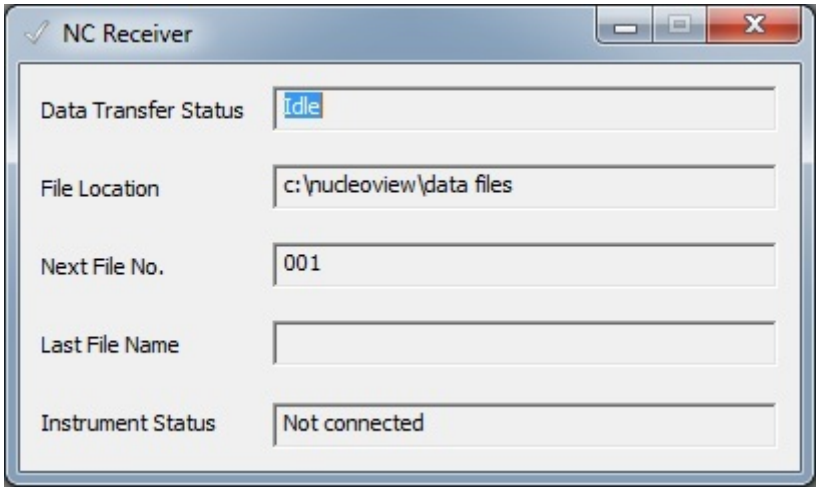


Figure 46 Not connected

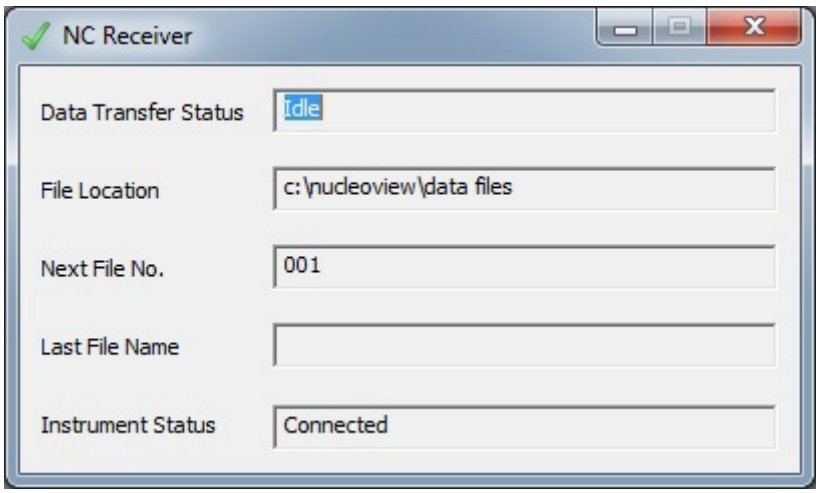


Figure 47 Connected

If "Not connected" is the status then check the USB cable. When the NucleoCounter is turned off this status will also be the case.

If data still is not received quit NucleoView and close the NC Receiver. Then open NucleoView and check that the NC Receiver is also started. If necessary, restart the computer before opening NucleoView again.

The status "Connected" is present when and only when:

- The NucleoCounter is turned On, and
- The USB Cable is properly connected, and
- The NC Receiver is working properly

In case you are certain that all cables etc. are as required you can try to use the F51 Testimage to PC function in the NucleoCounter (only valid from firmware version 4.00 and

higher). This F51 will simply transfer a predefined testimage to the PC and NucleoView will recognize the image if it is transferred.

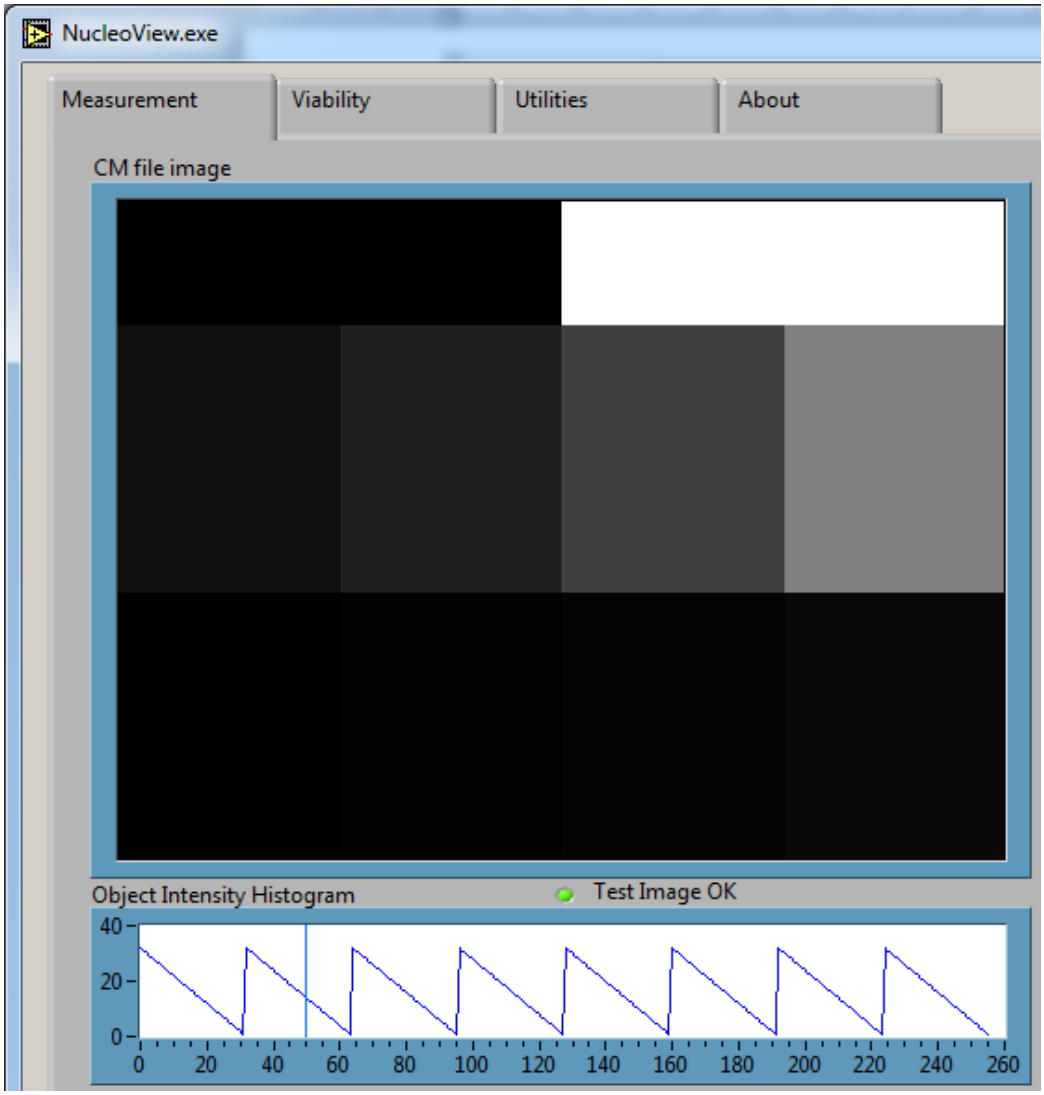
The following procedure shows this.

7.2.1 Test Image transfer from the NucleoCounter.

- Press <F> <5> <1> on the keyboard of the NucleoCounter.
- Press enter <↵>.
- The display of the NucleoCounter shows for 1-2 seconds

```
1  Testimage <-> PC
2
3
4
```

- When the NucleoCounter responds with “Ready” again in the display, the NucleoView display will appear like



- The Test Image is present, a “saw-tooth” pattern is shown in the Object Intensity Histogram and if the “Test Image OK” indicator is visible(and green) the image data transfer from the NucleoCounter to NucleoView is working.

7.3 NucleoView “freezes”

If NucleoView ”freezes” or does not respond, quit the program using the “QUIT”-button in the Measurement tab. Close down the NC Receiver. Then restart NucleoView and the program should be ready for use again. Any transmission of data from the NucleoCounter while the NucleoView is disabled is received and stored on the computer, as long NC Receiver is active. Such results can be viewed the next time NucleoView is activated. If the “QUIT”-button does not respond (after only few seconds), you can close NucleoView down using the Windows Task Manager. For instructions on the use of Windows Task Manager please refer to the documentation supplied with Windows.

If any of the fields starts to blink or behave "strange" try to select a new file from the result list or a new folder using the Browse button.

Low resources on the PC running NucleoView can cause the result field to show the following message "Not SP-100". You can check your resources on the PC using the Windows Task Manager. Please show **caution** when performing the following or contact your local PC administrator. The Windows Task is activated in the following way[13]:

Press the 3 keys "Ctrl" + "Alt" + "Delete" and the same time, then press the Task Manager button, hit Performance and the following appears (shown partly):

[13] This applies only to Windows 2000.

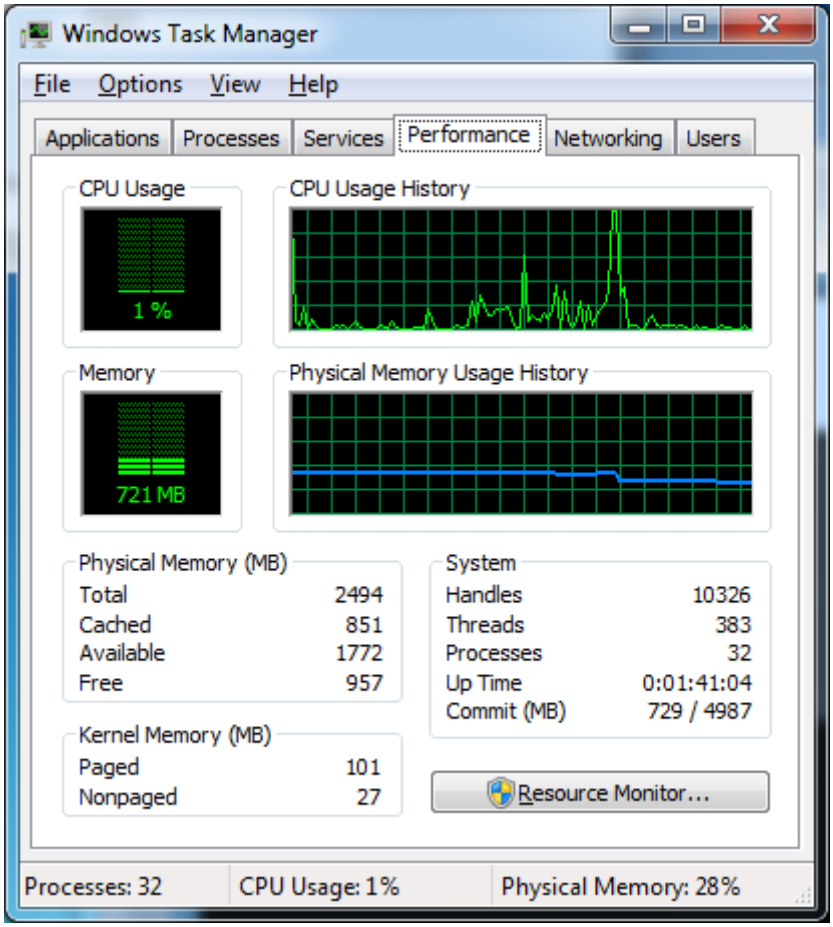


Figure 48 Windows Task Manager – Performance when running NucleoView

Figure 48 shows the Windows Task Manager. During normal operation of NucleoView the CPU usage will typically be less than 20% however it can peak to 90%, when a new file is received or when another file from the file list is selected.

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8 Technical specifications

8.1 NucleoView™ software

System requirements

For operating systems see table 1 below.

USB 1.1 port[14]. Display Screen Area: Minimum 1024 x 768[15].

Table 1 Functionality of NucleoView and the USB driver on computers running different operating systems.

Operating system	Functionality of application	Installation tested	Installation documented
Win XP (32/64 bit)	+	+	-
Win 7 (32/64 bit)	+	+	+[16]
Mac OS[17]	+	+	-

When using other operating systems than Windows 7 the installation procedures can differ slightly from the procedures described in chapter 1 Installation.

[14] The NucleoCounter system does not support all types of USB hubs. Please note that some computers have internal hubs, the NucleoCounter system may not function on such computers.

[15] For Display Screen Area settings refer to the Display option in the Windows Control Panel.

[16] Only 64 bit version.

[17] The NucleoView program can be installed on a vitual macihine on Mac OS, which then have a Windows OS installed. Refer to your IT administrator regarding this.

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9 Appendix 1: Printed result list

Below is an example of what the result list looks like when it is printed. On top of the page is a header and below are the chosen results from the active session including comment and operator.

c:\nucleoview\data files\021218\Result list - 021218 PRN.txt

Company
Address1, Address2

Department

NucleoCounter S/N: 002-09

Printdate: 18. december 2002
Printtime: 10:45

File name	Cells/ml (cass.)	MF	Cells/ml (susp.)
021218-001.CM	6.5 x 10E4	1.0	6.5 x 10E4
Comment:	CHO Non-viable		
Operator:	LB		
021218-002.CM	5.80 x 10E5	3.0	1.74 x 10E6
Comment:	CHO Total		
Operator:	LB		
021218-003.CM	5.8 x 10E4	1.0	5.8 x 10E4
Comment:	CHO Non-viable		
Operator:	LB		
021218-004.CM	5.88 x 10E5	6.0	3.53 x 10E6
Comment:	CHO Total		
Operator:	LB		
021218-005.CM	6.4 x 10E4	1.0	6.4 x 10E4
Comment:	CHO Non-viable		
Operator:	LB		
021218-006.CM	6.16 x 10E5	9.0	5.54 x 10E6
Comment:	CHO Total		
Operator:	LB		

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10 Appendix 2: Printed viability report

Below is an example of a printed viability report. On top of the page is a header and below that are the files, results of the analyses and the calculated viability in cells pr. mL and %.

C:\Nucleoview\data files\021218\Viability list - 021218PRN.txt

Company
Address1, Address2

Department

NucleoCounter S/N: 002-09

Creation date: 18. december 2002
Creation time: 08:45

Total cell count

File name	Cells/ml (cass.)	MF	Cells/ml (susp.)
021218-002.CM	5.80 x 10E5	3.0	1.74 x 10E6

Comment: CHO Total
Operator: LB

Non-viable cell count

File name	Cells/ml (cass.)	MF	Cells/ml (susp.)
021218-001.CM	6.5 x 10E4	1.0	6.5 x 10E4

Comment: CHO Non-viable
Operator: LB

Concentration of Viable cells/ml 1.67 x 10E6
Concentration of Viable cells(%) 96.3

Total cell count

File name	Cells/ml (cass.)	MF	Cells/ml (susp.)
021218-004.CM	5.88 x 10E5	6.0	3.53 x 10E6

Comment: CHO Total
Operator: LB

Non-viable cell count

File name	Cells/ml (cass.)	MF	Cells/ml (susp.)
021218-003.CM	5.8 x 10E4	1.0	5.8 x 10E4

Comment: CHO Non-viable
Operator: LB

Concentration of Viable cells/ml 3.47 x 10E6
Concentration of Viable cells(%) 98.4

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11 Appendix 3: Default settings for Yeast samples

The settings below are *suggested* default settings for users with very high cell count. This can be the case when Yeast fermentation samples are being analyzed using the NucleoCounter YC-100 instrument.

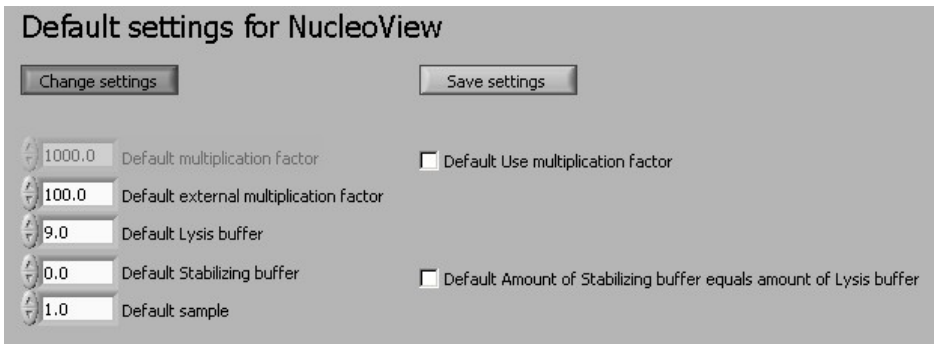

Figure 49 Suggested settings for very high cell counts in e.g. Beer or Wine Yeast fermentation samples using the NucleoCounter YC-100.

If the multiplication factor for Non-viable samples is changed, the new value is stored and becomes the default multiplication factor for non-viable samples as long as NucleoView is **running**. This is useful when e.g. Yeast samples are analysed, since pre-dilution of e.g. 50-500 is common.

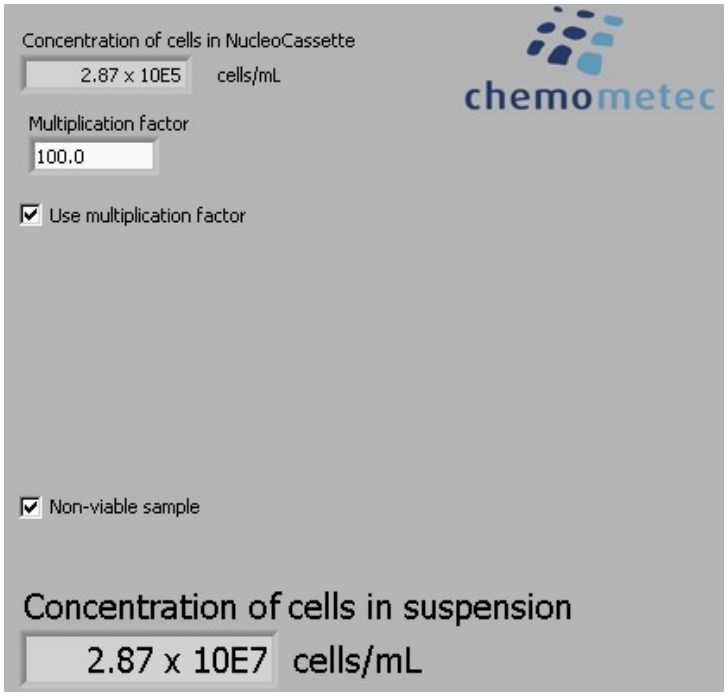

Figure 50 Setting of the Multiplication factor to other than 1 for Non-viable samples.

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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 the NucleoCounter™ and other cell counting methods may occur. As such, nothing contained or stated herein including results obtained from use of the NucleoCounter™ or NucleoCassette™ shall be construed to imply any warranty or guarantee. ChemoMetec A/S and affiliated companies shall not be held liable for damages and customers shall indemnify ChemoMetec A/S and affiliated companies against liability flowing from use of potentially inaccurate data generated by the NucleoCounter™. It is recommended that all results obtained with the NucleoCounter™ are validated against appropriate reference methods and/or traditional laboratory methods at regular intervals.