

NimbleGen MS 200 Microarray Scanner Operator's Manual





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Table of Contents

F10logue	
I. Revision History	i
II. Contact Addresses	i
III. Certificate of Conformity	ii
IV. Warranty	ii
V. Trademarks	ii
VI. Intended Use	ii
VII. License Statements for the Instrument	ii
VIII. Software License Agreement	ii
IX. Preamble	.iii
X. Use of the Instrument Operator's Manual	.iii
XI. Conventions Used in this Manual	. iv
XII. Warnings and Precautions	v
XIII. Disposal of the Instrument and Consumables	vii
A Overview	. 1
1. Introduction	1
2. Specifications	6
2.1 Specifications of the Microarray Scanner	6
2.2 Specifications of the Control Unit	8
2.3 Specifications for Microarray Slides	8
P. Sustem Description	
b System Description	
1. System Packages	11
System Description 1. System Dackages 2. System Description	11 12
System Description 1. System Description 2. System Description 2.1 Description of the Instrument	11 12 12
System Description 1. System Description 2. System Description 2.1 Description of the Instrument 2.2 Slide Magazine.	11 12 12 13
1. System Description 2. System Description 2.1 Description of the Instrument 2.2 Slide Magazine 2.3 Slide Magazine Box	11 12 12 13 14
System Description 1. System Description 2. System Description 2.1 Description of the Instrument 2.2 Slide Magazine 2.3 Slide Magazine Box 3. Installation	11 12 12 13 14 19
1. System Description 2. System Description 2.1 Description of the Instrument 2.2 Slide Magazine 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation	11 12 12 13 14 19 19
1. System Description 2. System Description 2.1 Description of the Instrument 2.2 Slide Magazine 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation 3.2 Unpacking the Instrument	11 12 12 13 14 19 19
 System Description System Description System Description of the Instrument Description of the Instrument Slide Magazine Slide Magazine Box Installation Installation Warning and Recommendation Unpacking the Instrument Space and Power Requirements 	11 12 12 13 14 19 19 19
 System Description 1. System Packages. 2. System Description of the Instrument. 2.1 Description of the Instrument. 2.2 Slide Magazine. 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation 3.2 Unpacking the Instrument 3.3 Space and Power Requirements 3.4 Environmental Requirements 	11 12 12 13 14 19 19 19 19 20
 System Description 1. System Packages. 2. System Description 2.1 Description of the Instrument. 2.2 Slide Magazine. 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation 3.2 Unpacking the Instrument 3.3 Space and Power Requirements 3.4 Environmental Requirements 3.5 Installation of the Control Unit. 	11 12 12 13 14 19 19 19 20 21
b System Description 1. System Description 2. System Description of the Instrument 2.1 Description of the Instrument 2.2 Slide Magazine 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation 3.2 Unpacking the Instrument 3.3 Space and Power Requirements 3.4 Environmental Requirements 3.5 Installation of the Control Unit 3.6 Installation of the Instrument Software	11 12 12 13 14 19 19 19 19 20 21 21
 System Description System Description System Description of the Instrument. Description of the Instrument. Slide Magazine Slide Magazine Box Installation Installation Warning and Recommendation Unpacking the Instrument Space and Power Requirements A Environmental Requirements Installation of the Control Unit. Installation of the Instrument Software Starting the Control Unit, Microarray Scanner, and Instrument Software 	11 12 12 13 14 19 19 19 19 20 21 21 21 25
 System Description System Description System Description of the Instrument. Description of the Instrument. Slide Magazine Slide Magazine Box Installation Installation Warning and Recommendation Unpacking the Instrument Space and Power Requirements Space and Power Requirements Installation of the Control Unit. Installation of the Instrument Software Starting the Control Unit, Microarray Scanner, and Instrument Software Seftware 	11 12 12 13 14 19 19 19 20 21 21 21 25 27
 System Description System Description System Description of the Instrument. Description of the Instrument. Slide Magazine. Slide Magazine Box Installation Installation Warning and Recommendation. Unpacking the Instrument Space and Power Requirements Space and Power Requirements Installation of the Control Unit. Installation of the Instrument Software. Starting the Control Unit, Microarray Scanner, and Instrument Software Setting System Verification: The Initial ILC Test 	11 12 12 13 14 19 19 19 20 21 21 21 25 27 31
 System Description 1. System Packages. 2. System Description of the Instrument. 2.1 Description of the Instrument. 2.2 Slide Magazine. 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation 3.2 Unpacking the Instrument 3.3 Space and Power Requirements 3.4 Environmental Requirements 3.5 Installation of the Control Unit. 3.6 Installation of the Instrument Software 3.7 Starting the Control Unit, Microarray Scanner, and Instrument Software 3.8 Performing System Verification: The Initial ILC Test 	11 12 12 13 14 19 19 19 19 20 21 21 21 25 27 31 31
 System Description 1. System Packages. 2. System Description of the Instrument. 2.1 Description of the Instrument. 2.2 Slide Magazine 2.3 Slide Magazine Box 3. Installation 3.1 Installation Warning and Recommendation 3.2 Unpacking the Instrument 3.3 Space and Power Requirements 3.4 Environmental Requirements 3.5 Installation of the Control Unit. 3.6 Installation of the Instrument Software 3.7 Starting the Control Unit, Microarray Scanner, and Instrument Software 3.8 Performing System Verification: The Initial ILC Test 	11 12 12 13 14 19 19 19 19 20 21 21 21 21 25 27 31 31 31

3.1 Understanding Sessions
3.2 Using the Help Viewer
3.3 Setting Software Options
4. Specifying Parameters
4.1 Working with the Magazine Control
4.2 Specifying Scan Parameters41
5. Specifying Image File Settings46
6. Specifying Scan, Barcode, and Autogain Areas48
7. Starting and Aborting a Scan58
7.1 Before You Start the Scan58
7.2 Starting a Scan58
7.3 Aborting a Scan
7.4 Saving Log Files60
8. Reviewing Images61
8.1 Displaying an Image61
8.2 Zooming in and out61
8.3 Displaying Intensity Data for a Feature62
8.4 Adjusting Image Appearance62
8.5 Displaying a Histogram or Line Profile63
8.6 Choosing the Channel(s) to Display66
D Maintenance and Care67
1. ILC Test
2. Liquid Spill Cleanup68
3. Ozone Filter Replacement
4. Instrument Relocation
5. Instrument Disinfection70
E Appendix73
1. Troubleshooting73
2. Ordering Information79
3. Quick Start
3.1 Starting the Scanner, Control Unit, and Instrument Software80
3.2 Loading Microarray Slides81
3.3 Specifying Parameters81
3.4 Scanning81
3.5 Reviewing Images82
3.6 Instrument Software Workspace82
4. Index

Prologue

I. Revision History

Version	Revision Date
1.0	April 2009

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Questions or comments regarding the contents of this manual can be directed to your Roche NimbleGen Account Manager or Roche Microarray Technical Support. Go to *www.nimblegen.com/arraysupport* for contact information.

Every effort has been made to ensure that all the information contained in the NimbleGen MS 200 Microarray Scanner Operator's Manual is correct at the time of printing. However, Roche NimbleGen, Inc. reserves the right to make any changes necessary without notice as part of ongoing product development.

II. Contact Addresses

Manufacturer	Tecan Austria GmbH Untersbergstrasse 1A A-5082 Grödig/Salzburg AUSTRIA/EUROPE
Distribution	Roche Diagnostics GmbH Sandhofer Straße 116 D-68305 Mannheim GERMANY

III. Certificate of Conformity

Provided with the NimbleGen MS 200 Microarray Scanner.

IV. Warranty

For warranty and service information, contact your Roche NimbleGen Account Manager. Go to *www.nimblegen.com/arraysupport* for contact information.

V. Trademarks

NIMBLEGEN is a trademark of Roche. Other brands or product names are trademarks of their respective holders.

VI. Intended Use

The NimbleGen MS 200 Microarray Scanner is designed to read fluorescently labeled DNA microarrays on 1 x 3 inch standard glass laboratory slides for experiments in research laboratories. This Microarray Scanner is intended for life science research only.

VII. License Statements for the Instrument

Any patents or patent applications are owned by the original equipment manufacturer.

VIII. Software License Agreement

An envelope containing all license and end user agreements is packaged with the NimbleGen MS 200 Microarray Scanner. Please observe all statements made in these documents.

IX. Preamble

Read this entire manual before operating the NimbleGen MS 200 Microarray Scanner. This document is intended as instructions for use for the Microarray Scanner. It provides information about:

- Installation
- Operation
- Maintenance and care
- Troubleshooting

X. Use of the Instrument Operator's Manual

This Operator's Manual describes the operation of the NimbleGen MS 200 Microarray Scanner and contains the following chapters:

Chapter A, *Overview*, provides an introduction to the instrument's capabilities and specifications for the instrument, control unit, and microarray slides.

Chapter B, System Description, contains a description of the instrument's components and consumables and instructions on the installation of the instrument and software.

Chapter C, Operation, describes the operating procedures for the instrument.

Chapter D, Maintenance and Care, describes the maintenance procedures that are required for the instrument.

Chapter E, Appendix, contains troubleshooting and ordering information of the instrument and its accessories and consumables. It also provides a quick start guide to help you get started using the Microarray Scanner quickly.

XI. Conventions Used in this Manual

Text Conventions

To impart information that is consistent and memorable, the following text conventions are used in this Operator's Manual:

Convention	Description
Numbered listing	Steps in a procedure that must be performed in the order listed.
Italic type, blue	Points to a different chapter in this Operator's Manual to consult or to a web site.
Italic type	Identifies the names of controls (checkboxes, option buttons, etc.) in dialog boxes, windows, or message boxes in the instrument software.
Bold type	Identifies buttons and menu names when operating the instrument software.
Underscore and brackets	A placeholder for information such as in the actual name of a directory in a path is enclosed in brackets, e.g. <install path="">. Placeholders (for file names, numbers, dates, etc.) are separated by an underscore (), e.g. <barcode>_<user text="">_<laser wl="">.</laser></user></barcode></install>

Symbols

The following types of notices may be used in this manual to highlight important information or to warn the operator of a potentially dangerous situation:





institutional requirements for disposal of the accessories.

XII. Warnings and Precautions

Handling Requirements

The Microarray Scanner must be used by only trained and skilled personnel. It is essential that the following safety information required for installation and operation of the Microarray Scanner is carefully read and observed. Ensure that this safety information is accessible for every employee working with the Microarray Scanner.

- Always follow basic safety precautions during use to reduce the risk of injury, laser radiation, or electrical shock.
- Read and understand all information in this document. Failure to read, understand, and follow its instructions could result in damage to the instrument, injury to operating personnel, or poor instrument performance.
- Observe all (Warning) and (Caution) statements in this document.

- The Microarray Scanner is rated as a Class 1 (no hazard during normal use) laser product and contains two Class 3b lasers.
- Never force a slide or other accessory into the slide magazine or any component of the instrument.
- Observe proper laboratory safety precautions, such as wearing protective clothing and using approved laboratory safety procedures.

It is assumed that instrument operators, due to their vocational experience, are familiar with the necessary safety precautions for handling chemicals and biohazardous substances. Adhere to the following laws and guidelines:

- National industrial protection law
- Accident prevention regulations
- Safety data sheets of the reagent manufacturers

Safety and Information Labels

Safety and information labels found on the rear of the Microarray Scanner warn of potential hazard or highlight important information. These labels refer to issues that must be taken into account when using this instrument.



Figure 1: Safety and Warning Labels on the Microarray Scanner

XIII. Disposal of the Instrument and Consumables

Disposal of the Instrument

Because components of the Microarray Scanner could be exposed to potentially infectious chemical samples, toxic or corrosive chemicals, or radioactive chemicals, perform appropriate waste management to ensure there is no risk of contamination.



ALWAYS DISINFECT AND DECONTAMINATE THE INSTRUMENT BEFORE DISPOSAL. Depending on the experiments, components of the Microarray Scanner could have been in contact with biohazardous material. Refer to the "Instrument Disinfection" section in Chapter D. Maintenance and Care, for details.

- Make sure to treat this material according to applicable safety standards and regulations.
- Always decontaminate (i.e. clean and disinfect) all components before disposal.
- Follow laboratory procedures for biohazardous waste disposal.



Observe all national, regional, and local regulations for waste disposal and management.



Directive 2002/96/EC on waste electrical and electronic equipment (WEEE): Negative environmental impacts associated with the treatment of electrical and electronic equipment waste:

- Do not treat electrical and electronic equipment as unsorted municipal waste.
- Collect waste from electrical and electronic equipment separately.



Pollution degree: 2



Method of Disposal: Electronic waste, contaminated waste (infectious waste)

Disposal of the Consumables



Biological hazards can be associated with the waste material of the process run on the instrument.



Treat used consumables and all substances in accordance with good laboratory practice guidelines.



Inquire about appropriate collecting points and approved methods of disposal in your country, state, or region.

A Overview

1. Introduction

The Microarray Scanner is a laser-scanning instrument dedicated to acquiring fluorescent images on standard microarray slides (1 x 3 inch). High resolution combined with high sensitivity as well as uniformity, reproducibility, auto calibration, and a dynamic range are key characteristics of the Microarray Scanner.

A slide magazine enables the automatic processing of up to 48 slides, which can be scanned several times using different laser, filter, and photomultiplier tubes (PMTs) gain settings with no further user intervention in a single batch run.

Active air filtering reduces ozone concentration so that the slide processing and storage area is protected from external high ozone concentrations during operation.

The Microarray Scanner produces single- or multi-image Tagged Image File Format files (TIFF, .tif) that can be read by software packages available from Roche NimbleGen and other vendors.

Following is a description of the instrument's key functionality and components:

Autofocus Procedure

Microarray slides can have a variable surface planarity, which can affect the focusing of the scanning laser and may cause problems with measuring fluorescence intensities.

To counter this problem, the Microarray Scanner implements a dynamic autofocus procedure that measures the surface contour (surface modeling) before it scans a slide. During this procedure, the Microarray Scanner performs a multi-line scan using different positions across the entire slide to evaluate the surface. Then the contour of the slide is interpolated to correct for focal distance and tilt while scanning. This unique dynamic autofocus procedure is more reliable than single focusing routines, which concentrate on only one point of the slide. Moreover, this dynamic autofocus procedure is less vulnerable to disturbances such as dust on the slide surface.

Laser Excitation

The Microarray Scanner is designed for microarray imaging using individual solid state lasers that emit light at wavelengths suitable for exciting fluorescent dyes used as labels.

The lasers are able to excite Cy3- and Cy5-labeled samples to measure fluorescence intensities in microarray analysis. Laser emission for excitation of fluorescent dyes can be chosen at these wavelengths:

- Green: 532nm
- Red: 635nm

Other dyes similar to Cy3 and Cy5 can also be used, such as Alexa 555 and Alexa 647 or DY 547 and DY 647.

Excitation intensity can be varied with neutral density filters located in a filter wheel behind the lasers so that each laser can be adjusted separately with its own filter device.

A reference photodiode is used for detecting the laser power.

Emission Filters

The Microarray Scanner is equipped with filters that are optimized for the emission spectrum of the fluorescent cyanine dyes Cy3 and Cy5 and the other dyes specified earlier.

Detection

Fluorescence emission is directed through filters before reaching the photodetector. To detect photons emitted by the tagged molecules, photomultiplier tubes (PMTs) with very high sensitivity and low electronic noise are used to convert the light energy into electric current that can be measured. The resulting current is proportional to the incident emission light at the photodetector.

The amplification of the photon signal can be controlled by the voltage applied to the tube. PMT voltages are adjustable in the PMT gain settings for different channels to equalize intensity values on each slide.

To optimize and increase the dynamics of scanning, adjust the PMT setting using the histogram mode to balance channel intensity. Balance the histogram at all points above background across signal intensity. For more information on histograms, refer to *Chapter C, Operation*.



Histogram mode is not available for $2\mu m$ scans during the run but is available post run.

Repeated use of high power settings for the lasers causes photobleaching of the dye molecules and thus detrimentally affects fluorescence intensity.

Read Speed

The Microarray Scanner can measure two fluorescent dyes simultaneously (parallel mode) at increased overall scan speed. Alternatively, the dyes can be read one at a time (sequential mode). In addition, two scanning velocities are available: 16 lines per second (8Hz) and 24 lines per second (12Hz).

The overall processing time includes loading and unloading of a slide, reading the barcode, performing the autofocus, autogain, and scanning processes, and saving the image file. The file size of an image depends on the resolution and size of the scanned area. Increasing the spatial pixel size is accompanied with an increasing scan speed and decreasing amount of data.

The following table shows approximate file sizes and processing times for the NimbleGen HD2 family of arrays (2.1M, 3x720K, and 12x135K probe formats) using a speed of 24 lines per second, parallel mode, autogain turned off, and scan length of 62mm. File size and processing time will vary if a different scan speed or scan length is specified.

Possibilition (um)	File Siz	e (MB)	Processing Time
	Single Channel	Dual Channel	(min)
2	630	1260	25
5	100	200	12
10	25	50	8
20	6	12	6
40	2	3	5

ILC Module

For periodic testing of the Microarray Scanner's performance, the Microarray Scanner includes an internal Integrated LaserCheck (ILC) module. The ILC slide, which is used by this module, is provided with the Microarray Scanner (refer to page 27 for information on how to load the ILC slide and run the ILC module). The ILC module is designed for durability, contains no degradable parts, and requires no consumables.

The ILC module checks whether the Microarray Scanner is operating according to determined parameters within defined limits. The ILC module should be run on a regular basis. Roche NimbleGen recommends a period of time of 2 weeks.

Once you initiate the ILC run, the ILC module runs fully automated with no user intervention. The instrument software will automatically load and unload the ILC slide as required and perform and evaluate all tests. The following tests are performed, and results are reported (refer to page 29 for information on how to save the results):

Actuators Test. Performs a step-loss check for each actuator (e.g. slide transport, filter wheels).

Barcode Reader Test. Verifies the functionality of the barcode reader.

Oscillator Test. Checks the functionality of the oscillator subsystem and measures the following: (1) the frequency of the oscillator subsystem and the presence of the encoder signal at medium and accelerated frequency, (2) the maximum magnitude of the pixel jitter, and (3) the deviation perpendicular to the scanning direction (stagger).

Electronics Test. Measures the electronic noise and determines the integrity of the detection electronics (analog integration, connections, A/D conversion). Data acquisition is performed with closed detector and gain set to zero.

Detector(s) Test. Evaluates the detector's noise. Data are acquired with closed detector and gain set to defined values listed in the report.

Laser(s) Test. Reviews the integrity of the laser light sources. For each laser, the excitation light is reflected on the ILC slide, and the noise is measured with the detector. Additionally, the laser light intensity is measured with a reference photodiode.

Optical Alignment Test. Verifies the alignment and stability of the optical system. For this test, Z scans on a blank glass area are performed.

Autofocus Test. Checks the reliability of the autofocus and the slide transport subsystem while performing several Z scans using different parameters on the ILC slide.

Optical Resolution Test. Scans defined test structures (i.e. line gratings with high-low reflectivity parallel to the X and Y axes) on the ILC slide to provide information on optical resolution.

Sensitivity Tests. Check the overall performance of components that are relevant for sensitivity: electronics, detectors, lasers, alignment, and filter blocking.

Positioning Test. Verifies and, if necessary, recalibrates the position of the slide transport (X and Y axes) relative to the measurement optics. Defined position marks on the ILC slide are scanned.

Geometric Linearity Test. Checks for X and Y axes and if necessary, recalibrates (Y axis only) the image linearity within scanned images. Defined straight line patterns on the ILC slide are scanned to evaluate the image linearity in the X and Y directions.

2. Specifications

2.1 Specifications of the Microarray Scanner

Specification	Description
Performance	
Sensitivity	<0.04 Fluorescein equivalent/µm ² (Cy5)
Dynamic range	5 orders of magnitude
Pixel resolution	2, 5, 10, 20, or 40µm
Reading speed	16/24 lines per second (8/12Hz)
Optical	
Light sources	Two solid-state lasers (532 and 635nm)
Fluorescence detector(s)	Two low dark current photomultiplier tubes
Gain adjustment	0.01 - 1000%
Optics	High-aperture collection optics (NA 0.7)
Emission filters	One predefined filter per laser
Dichroic beam splitter	Two different beam splitters
Pinholes	Two pinholes of fixed size
Reference detector	Monitors laser power
Autofocus	Automatic, user-selectable slide types
Barcode reader	Capable of reading NimbleGen, Code 39, and Code 128 barcode formats
Mechanical	
Scanner	Linear movement with cone-rod principle
Slide holder	3-axis high-precision module for scanning and automatic alignment of exact focus position
Stacker cover	Opens upon pressing of the insert/eject magazine button

Electrical	
Power supply	Auto-sensing: 100 - 120V / 220 - 240V, 50/60Hz
Power consumption	150VA
Approvals	EN 61010-1:2001, 61010-2-081/A1:2003, 60825-1/A1:2002, UL 61010-1:2004, IEC 61010-2-081/A1:2003, CAN/CSA-C22.2 No. 61010-1:2004, CAN/CSA-C22.2 No. 61010-2-081:2004, CAN/CSA-E60825-1:2003
Processing	
Control	Software-controlled by external PC
PC operating system	Windows Vista Business (64-bit)
Interface	Ethernet RJ45
Data acquisition	High-speed 16-bit A/D conversion
Samples to be measured	Fluorescent features on microscope slides within the specified slide dimensions
Slide magazine	Holds up to 48 slides and allows automatic loading
Environmental	
Ambient temperature	Operation: 18 - 28°C (64 - 82°F) Non-operation: -20 - 60°C (-4 - 140°F)
Relative humidity	Operation: 20 - 80% non-condensing Storage: 10 - 80% non-condensing
Overvoltage category	II
Pollution degree	2
Method of disposal	Electronic waste (infectious waste)
Physical	
Outer dimensions	Width: 60cm (23.6in) Height: 46.6cm (18.5in) Depth: 61.8cm (24.4in)
Weight	Approximately 38kg (84.0lb)

2.2 Specifications of the Control Unit

Component	Specification
Computer	HP dc7800 CMT with Dual Core 2 3GHz (gigahertz)
	Memory: 8GB (gigabytes)
	Disk 1: 160GB for system and applications
	Disk 2: 160GB for use by users
	Disk 3: 1TB (terabyte) Data storage
	CD/DVD: DVD-RW
	Ethernet RJ45: 1 x to connect to the Microarray Scanner (on the computer's PCI card)
	1 x to connect to the lab's network (on the computer's main board)
Monitor	19 inch LC display with sound (1280 x 1024 resolution)
Keyboard	US layout
Mouse	2-button optical scroll mouse
Operating system	Microsoft Vista Business 64-bit

2.3 Specifications for Microarray Slides

Slide Formats

Microarray slides to be processed using the Microarray Scanner should meet the nominal specifications of 25.4 x 76.2mm (1 x 3in) for standard glass laboratory slides or following dimensions:

- Width: 24.6 26.0mm
- Length: 75.0 76.5mm
- Thickness: 0.8 1.2mm

Scan Area, Resolution, and Barcode Specifications

The maximum scan area on a slide is set to 22×75 mm (Figure 2). Pixel resolution is selectable at 2, 5, 10, 20, and 40 μ m.

The scan and barcode dimensions could vary due to experimental and/or manufacturer settings. If a barcode area is designated on the slide, the effective scan area is smaller.



The barcode area cannot exceed 22 x 18mm. Ensure that barcode labels do not interfere with the rails in the slide magazine (Figure 6) or the internal slide transport (Figure 41).

Recommendations for Non-NimbleGen Microarrays

To obtain reliable results, it is strongly recommended that reference spots are placed onto the slide for quality control. These spots should represent every dye used for the experiment and be distributed in a nonsymmetrical pattern across the slide. It is also recommended that "blank" spots be placed on the slide to check for variation of the background in the experiment.



Figure 2: Scan Area Dimensions (in mm) on Standard Slides with Maximal Barcode Area Definition

Notes

B System Description

1. System Packages

The following table lists the components required for the Microarray Scanner and control unit. Check for completeness and inspect packaging prior to installation.



The original shipping boxes must be transferred unopened to the installation site. On delivery, carefully inspect the boxes. Report any suspected damage immediately to Roche NimbleGen and to the shipping agent before accepting the product.



Use only the original packaging for transportation or relocation of the equipment.

Quantity	Component
Microarray Scan	ner (Catalog No. 05394341001)
1	MS 200 Microarray Scanner
1	Power cord (European standard)
1	Power cord (North American standard)
1	Ethernet cable
1	Slide magazine box
1	ILC slide
Control Unit (Ca	talog No. 05394325001)
1	
1	19" LC display with sound
1	2-button optical scroll mouse
1	19" LC display with sound 2-button optical scroll mouse HP PS/2 standard keyboard (US layout)
1 1 1	19" LC display with sound 2-button optical scroll mouse HP PS/2 standard keyboard (US layout) Power cord (European standard)
1 1 1 1	19" LC display with sound 2-button optical scroll mouse HP PS/2 standard keyboard (US layout) Power cord (European standard) Power cord (North American standard)
1 1 1 1 1 Instrument Softw	19" LC display with sound 2-button optical scroll mouse HP PS/2 standard keyboard (US layout) Power cord (European standard) Power cord (North American standard) ware (Catalog No. 05394333001)

2. System Description

2.1 Description of the Instrument

The following illustrations identify the outer attributes of the Microarray Scanner. An overview of the Microarray Scanner's functions is provided in subsequent sections.



Figure 4: Rear





2.2 Slide Magazine

The Microarray Scanner comes with a slide magazine that accommodates up to 48 microarray slides for sequential processing (Figure 6).



Figure 6: Slide Magazine Accommodates up to 48 Microarrays

For loading the slides, the slide magazine contains rails with the slide positions numbered 1 - 48 from the top down.

The latch on top of the slide magazine ensures that the flap remains closed so that all slides are kept safely in position and do not slip out. For instructions on how to unlock and lock the latch, refer to the "Inserting Slides into the Slide Magazine" section on page 14.

NimbleGen MS 200 Microarray Scanner Operator's Manual v1.0

The slot on the side of the slide magazine ensures the correct alignment of the slide magazine when inserting into the Microarray Scanner.

Use the handle for carrying and loading the slide magazine into the Microarray Scanner.

2.3 Slide Magazine Box

The slide magazine box can be used as a storage device for the slide magazine to provide protection against dust (Figure 7). This box is also suitable for placing the slide magazine reclined in the foam slot so that slides can be inserted from above (Figure 9).



Figure 7: Slide Magazine Box

Inserting Slides into the Slide Magazine

Wear powder-free gloves or equivalent when handling slides. Avoid touching the microarray area. If a slide is not coded or labeled, consider a defined orientation to ensure comprehensible image acquisition.

- 1. While lifting the latch's lever with one hand, press the flap's flange with the other hand to open the slide magazine. Release the lever so the latch remains in the unlocked position (Figure 8).
- 2. Hold each slide by its edges with the microarray facing up. Insert the slide into the slide magazine slot using the same rail profile. Insert slides with a barcode with the barcode area first (Figure 8). Numbered slots provide spacing for inserting slides.
- **3.** When finished inserting slides, release the flap's flange, which causes the latch to automatically lock so that slides do not slip out of the slide magazine.



Figure 8: Inserting Slides into the Slide Magazine (Upright)



Figure 9: Inserting Slides into the Slide Magazine (Horizontally in Box)

Inserting or Removing the Slide Magazine

Insertion and removal of the slide magazine is achieved manually and should be done gently and with care (Figure 10). A green light in the upperleft corner of the insert/eject magazine button indicates that the slide magazine is accessible, whereas a locked stacker cover is indicated by a red light.



Figure 10: Inserting the Slide Magazine into the Microarray Scanner

To insert the slide magazine:

- 1. Press the insert/eject magazine button to open the stacker cover.
- 2. Insert the slide magazine, aligning the slot on the slide magazine's side to join with the rail profile inside the Microarray Scanner. The lowering of the slide magazine is interrupted by a mechanical hold point. Apply gentle pressure to complete insertion. When fully lowered, the slide magazine's handle will project slightly above the compartment.
- **3.** Press the insert/eject magazine button to close the stacker cover. The initialization process starts, checking the slide magazine to determine which slots are occupied.
- Do not start the instrument software during the initialization process that the Microarray Scanner performs after slide magazine insertion. Doing so may cause the software to shut down unexpectedly.

To remove the slide magazine:

- 1. Press the insert/eject magazine button to open the stacker cover.
- 2. Wait for the slide magazine to eject from the stacker unit.
- **3.** Remove the slide magazine and press the insert/eject magazine button to close the stacker cover.

Slide Mishandling

Slides are prone to damage by mishandling resulting from them slipping out of or being incorrectly placed into the slide magazine (Figure 11). Avoid stacking or piling slides on each other. Improper slide handling and insertion into the slide magazine could lead to array damage, data loss, and possibly instrument damage.



slides slipping out

tilted or wedged slides

Figure 11: Examples of Slide Mishandling. Use care when unlatching the slide magazine's flap to ensure the slides do not slip out (left image). Avoid tilting or wedging microarrays in the slide magazine (right image).



Never force or wedge a slide into the slide magazine. Be careful when carrying and when inserting the slide magazine into the Microarray Scanner. Do not unlatch the flap, or slides could slip out.

3. Installation

3.1 Installation Warning and Recommendation

Before installing and switching on for the first time or after moving the Microarray Scanner to another site, allow it to stand for at least 3 hours to avoid electrical or optical failure caused by condensation.



🚺 It is recommended that a Roche NimbleGen Account Manager, Field Service Engineer, or Field Application Consultant performs the first installation of the Microarray Scanner.

3.2 Unpacking the Instrument

- Visually inspect the shipping box for damage before opening. Report any damage immediately.
- Place the box in an upright position and open it.
- Refer to the unpacking instructions to unpack the instrument.
- Visually inspect the instrument for loose, bent, or broken parts. Report any damage immediately.

3.3 Space and Power Requirements

Space Requirements

Ensure that the designated site meets the dimension and weight requirements of the Microarray Scanner. Place the instrument on a rigid and level surface that can support 50.0kg (110.0lb). The instrument is 46.6cm (18.5in) high, 60cm (23.6in) wide, and 61.8cm (24.4in) deep (Figure 12).

Leave sufficient distance behind the instrument for ventilation and access to the rear panel. Ensure that the stacker cover and front section can open and microarray loading can occur without hindrance.

Make sure that the main power switch, power cable, and Ethernet cable are accessible at all times and in no way obstructed. Do not place objects onto the Microarray Scanner that might prevent the opening of the stacker cover or stress its casing.

It is recommended to avoid any external vibrations to the Microarray Scanner because these can have a negative impact on its operation.



Figure 12: Dimensions of the Microarray Scanner (Top View and Side View)

Power Requirements

Power supply of the Microarray Scanner is auto-sensing and able to operate without any manual adjustments within the following voltage ranges:

- 100 120V (AC), 50/60Hz, 150VA
- 220 240V (AC), 50/60Hz, 150VA

3.4 Environmental Requirements

The Microarray Scanner is intended for indoor operation under controlled laboratory conditions. For optimal operation and smooth running of the relevant applications, operate the Microarray Scanner only in the specified conditions. Care should be taken to maintain a low-dust, low-vibration environment. Keep liquids and vapors away from the Microarray Scanner.

Temperature

Do not expose or locate the Microarray Scanner near direct sunlight or heat sources to avoid problems with misaligned mechanics and/or optics. To assure optimal performance, it is recommended to operate the Microarray Scanner in a temperature controlled (air conditioned) environment with a constant temperature.

- Temperature for operation: 18°C (64°F) 28°C (82°F)
- Temperature for transport: -20°C (-4°F) 60°C (140°F)

Humidity

The Microarray Scanner is potentially sensitive to condensation under humid conditions. Allow time for thermal equilibrium before opening the shipping box or after moving to a place with changed environmental conditions.

- Humidity for operation: 20 80% non-condensing
- Humidity for storage: 10 80% non-condensing

3.5 Installation of the Control Unit

To install the control unit and connect it to the Microarray Scanner:

- 1. Connect the mouse, keyboard, and monitor to the computer as instructed in the computer's documentation.
- **2.** Connect the Microarray Scanner to the computer's PCI-card Ethernet port using the Ethernet cable provided with the Microarray Scanner.
- **3.** Connect the computer to your local network using the main-board Ethernet port on the back of the computer.
- **4.** Connect the computer, monitor, and Microarray Scanner to a clean uninterruptable power supply.

3.6 Installation of the Instrument Software

Following is information to be aware of before you install the instrument software and details the installation procedures.

3.6.1 Instrument Software Safety

This section covers general introductory safety instructions applicable to the instrument software.

Significance of these Safety Instructions

The instrument software is a pure software product and as such does not contain any hazardous parts. However, the software is used to control hardware devices and options, which may contain parts that can move with great force and at considerable speed.

As a consequence, the safety of users and personnel can only be ensured if the safety instructions in this software documentation as well as the safety instructions of the hardware devices controlled with the software described here are strictly observed and followed. Therefore, all relevant reference materials must always be available to all users working with the instrument software.

What Users Must Know

Users must be qualified and trained to run the instrument and the instrument software. In particular, they must fulfill the following qualifications:

- Have a basic knowledge of the Windows operating system.
- Have a thorough knowledge of the technical system functions.
- Have a thorough knowledge of the application run on the system.
- Be familiar with good laboratory practice guidelines.
- Have read and understood the instructions in this document.

Only users that meet the qualifications prescribed here are eligible to run the instrument software described in this documentation.

3.6.2 Target Computer

The instrument software must be installed on the control unit shipped with the Microarray Scanner. Refer to "Specifications of the Control Unit" on page 8 for detailed computer specifications.

3.6.3 Who Should Install the Software?

A person with administrator rights or a Roche NimbleGen Account Manager, Field Service Engineer, or Field Application Consultant should install the instrument software.

3.6.4 Access Rights

To install the instrument software on a target computer, you need local administrator rights.

3.6.5 General Installation Considerations

Pay attention to the following:

- If a previous version of the instrument software is installed on the target computer, uninstall it before installing the new one.
- Install the new instrument software.
- Create the necessary user accounts.

3.6.6 Saving User-Defined Data

Roche NimbleGen recommends that you save all user-defined data and output files on the dedicated 1TB hard disk drive (not within the default installation path of C:\Program Files (x86)\Roche Diagnostics\NimbleGen MS200). Information in the default installation path will be lost in these situations:

- Installing a new version of the instrument software
- Repairing an existing version of the instrument software
- Removing (uninstalling) the instrument software

3.6.7 Installation Directory

The instrument software will be installed into the C:\Program Files (x86)\Roche Diagnostics\NimbleGen MS 200 directory by default. Roche NimbleGen recommends accepting this directory as the default drive/directory. However, you can install the instrument software into a different drive/directory.

3.6.8 Installing the Instrument Software

If you encounter problems, contact your Roche NimbleGen Account Manager or Roche Microarray Technical Support for technical and installation support to solve any known issues.

- 1. Close all software that is currently running.
- **2.** Log into the Windows operating system as local administrator. Use the following user account and password for the installation procedure:
 - Account: msInstall
 - Password: 1-msInstall

For instructions on using or creating a user account that has administrative permissions, refer to the "To add a new user to the computer" topic in Windows Help or contact your system administrator.

At the first log in, you will be prompted to change the password. The password must meet the following requirements and is valid for 90 days. Store any notation of the password in a secure location.

- Must be at least 8 characters
- Must not contain the user's account name or parts of the user's full name that exceed two consecutive characters
- Must contain characters from three of the following four categories:
 - Uppercase character (A through Z)
 - Lowercase character (a through z)
 - Base 10 digits (0 through 9)
 - Special symbols or non-alphabetic characters (for example: !, \$, #, %, etc.)
- **3.** Insert the installation CD for the instrument software into the computer's CD drive.
- 4. Open Windows File Explorer.
- **5.** Run autorun.exe on the installation CD.
- 6. Click Next to accept the suggested destination folder. To choose a different destination folder, click Browse and click Next when done selecting the folder.
- 7. The Welcome window opens. Click **Install Software** to continue. The instrument software and all necessary components will be installed by an automatic setup program. Wait while the setup process is performed.



Figure 13: Welcome Window

8. After successful installation, carefully read the release notes, if provided, to learn about the latest changes and functionality.

3.7 Starting the Control Unit, Microarray Scanner, and Instrument Software

1. Start the control unit and log into your user account.

The following is a preconfigured account. Your system administrator may set up additional accounts.

- Account: msOperator
- Password: 1-msOperator

2. To start the Microarray Scanner, switch on power using the main power switch on its rear and the on/off button on its left side.

The internal fans switch on. The blue initialize/function light indicates that the Microarray Scanner is ready for operation.

- **3.** To start the instrument software, choose between the following methods:
 - Click the instrument software icon on the computer's desktop.



• Click **Start** in the lower-left corner of the Windows task bar to open the menu. Click **All Programs** and locate the NimbleGen folder. Open the folder and select the program icon to launch the instrument software.

Maintenance	Connect To		
NimbleScan Control Panel			
NimbleGen MS 200 Startup	Default Programs		
1 Back	Help and Support		
Start Search	₽		

- **4.** The instrument software workspace opens (Figure 14). Make sure that the instrument software has completely loaded before continuing.
- 5. Click the Green Laser and Red Laser buttons to switch on the lasers. Allow them to warm for at least 10 minutes.

Refer to *Chapter C, Operation*, for further details on the instrument software workspace and how to use the instrument software. Using the instrument software, you can specify how long the lasers will remain on during periods of non-use before the instrument software automatically turns them off.



Figure 14: Top of Instrument Software Workspace Showing Laser Buttons

3.8 Performing System Verification: The Initial ILC Test

After you install the Microarray Scanner, control unit, and instrument software, perform an initial ILC test.



After the initial test, Roche NimbleGen recommends you perform the ILC test every 2 weeks.



Running the ILC test requires the ILC slide be installed in the Microarray Scanner. You will find the ILC slide in its protective package in the slide magazine box. Wear powder-free gloves or equivalent hand coverings when handling the ILC slide. Avoid touching or damaging the featured slide surface.

If necessary, switch on the Microarray Scanner using the main power 1. switch on its rear and the on/off button on its left side.

You will notice that the internal fans switch on. The blue initialize/function light indicates that the instrument is ready for operation.

- 2. Start the instrument software to connect to the Microarray Scanner. The Microarray Scanner performs an initialization routine.
- **3.** Click the *OC* tab in the instrument software and then click ILC.



Figure 15: QC tab on the Instrument Software Workspace

The Integrated Laser Check window opens (Figure 16) and displays all available test points. The following figure identifies the components of the Integrated Laser Check window.



Figure 16: Integrated Laser Check Window

4. Click the **Start/Stop ILC procedure** button (). When prompted, load the ILC slide into slot 48 of the slide magazine (Figure 17). Insert the slide magazine into the Microarray Scanner (Figure 10).



Figure 17: Inserting the ILC Slide

5. Click **OK** in the instrument software.

The Microarray Scanner removes the ILC slide from the slide magazine and places it in the ILC bay.

After the ILC test is completed, the instrument software produces and displays a comprehensive test report containing all measured parameters, evaluation limits, and results (PASS or FAIL, Figure 18). If you encounter problems, contact your Roche NimbleGen Account Manager or Roche Microarray Technical Support for technical and installation support to resolve any known issues.

For more information on the checks that occur during the ILC test, refer to "ILC Module" on page 4.

- Keep the ILC slide in the Microarray Scanner and remove it only when transporting or moving the instrument to another site. Make sure that slot 48 of the slide magazine is empty before instructing the Microarray Scanner to remove the ILC slide. Click the **Remove ILC slide** button () in the Integrated Laser Check window to move the ILC slide from the ILC bay to the slide magazine.
- 6. To save the results of the ILC test to a file:
 - a. Click the Export Report button () in the upper-left corner of the test results window.
 - **b.** Specify a file name and select a format (.pdf is recommended).



c. Save the file.

Figure 18: Example of a Test Report Integrated LaserCheck

Notes

C Operation

1. Introduction

This chapter describes how to use the instrument software and how to make use of its functionality. The instrument software is a comprehensive application to control the Microarray Scanner. It automates data and image acquisition from the scanning of various microarrays including NimbleGen high-density arrays.



Before installing and running the instrument software, review the "Instrument Software Safety" section on page 22.

2. Using the Instrument Software: A Typical Scanner Experiment

Following are typical steps in a scanner experiment.

- Before starting the software, start the control unit and Microarray Scanner (refer to page 25) and load the slides into the scanner (refer to page 14).
- **1.** Start the instrument software (refer to page 25).
- 2. Start the scanner's lasers to allow for proper warmup (refer to page 25).
- 3. Set parameters:
 - **a.** Specify scan settings (refer to page 41).
 - **b.** Specify image file settings (refer to page 46).
- **4.** Specify/review scan area (refer to page 48).
- **5.** Scan the slides (refer to page 58).
- **6.** Review images (refer to page 61).

3. Starting the Instrument Software

Refer to page 25 for instructions on starting the control unit, instrument software, and lasers.

The instrument software workspace opens and provides menus and functionality for viewing and editing scan specifications and acquiring and viewing images (Figure 19).

The Instrument Software Workspace

The instrument software consists of various input and output options. Its workspace is arranged to help you focus on specifying scan parameters (Figure 19, top figure) and viewing images (Figure 19, bottom figure).



Figure 19: Components of the Instrument Software Workspace

Roche button. Click to display the Application menu, which provides filerelated commands for creating, opening, or saving the session file that contains parameters used for a scanning experiment. The Application menu also enables you to open an image file, access the software's options, and exit the software.

9]() ÷				
	New				
)	Open				
	Save				
<i>i</i>	Open Image				
			Options	🗙 Exit	

Figure 20: Application Menu

Help button. Displays the online help, which provides background information, functionality overviews, and procedural assistance for the Microarray Scanner and the instrument software.

Scan tab. By default, the *Scan* tab is selected. It provides access to capabilities for specifying scan parameters and viewing images.

QC tab. Displays the ILC button to initiate testing of the Microarray Scanner's performance.

Scan Control. Displays the **Start/Stop Scan** button (**)** and a progress bar. During initialization at startup and scanning, the current task that is being performed is identified above the progress bar.

Laser Control. Displays buttons for turning on and off the Microarray Scanner's lasers. Click the laser's button to toggle between on and off modes. The button is bright yellow in color when its respective laser is turned on.

Image Control. Displays options for adjusting the appearance of an image, such as contrast and brightness. Provides control elements for image adjustments.

Magazine Control. Provides settings for each slide loaded into the slide magazine. This control is displayed by default.

Parameter Control. Enables you to adjust scan parameters for the currently selected slide.

File Settings Control. Enables you specify image file settings.

Area Definition Control. Allows you to review and specify the scan, barcode, and autogain areas for a slide. The settings for these areas can be saved in an area file.

Spot View. Displays an enlarged detail around the cursor. Intensity levels and X and Y coordinates are displayed when you position the mouse pointer over a pixel location in the Image View.

Slide View. Displays the defined scan area(s). During scanning, this view shows the currently acquired image. Use the resizable magenta rectangle of each scan grid to display a more detailed view in the Image View.



Do not use the resizable magenta rectangle when scanning at $2\mu m$ resolution.

Image View. Displays the currently scanned or loaded image. Refer to the "Reviewing Images" section on page 61 for more information.

3.1 Understanding Sessions

Session is the term used when the slide magazine is loaded with slides to scan in a user-defined run. Like the slide magazine, a session can handle 48 slide positions. Each position can refer to a slide that is intended to be scanned once or several times with defined parameters.

Within a session, you can customize the tasks to be performed by adjusting parameters (Parameter Control and File Settings Control) and areas (Area Definition Control) for each slide. These parameters can also be saved in several different files for future use.



Figure 21: Relationship between Session, Parameter, and Area Files

Session file (.sessx): Stores parameters for all slide positions. The session file contains all applied parameters and includes the information about the location of the image files.

The session file can be used to reload a complete session into the workspace. It is stored in XML format.

To create, save, and open session files:

Click the **Roche** button (()) to display the Application menu (Figure 20) and select the appropriate command.

Parameter file (.parax): Stores user inputs such as slide type, resolution, speed, channels, gain and laser intensity, as well as image file definition and scan areas can be saved.

A parameter file can be loaded into a session and assigned to one or more slides. For instance, you can create parameter files for specific batch runs, apply and/or save them, and switch between them as you work. The parameter file is stored in XML format.

You will learn more about setting parameters in the "Specifying Scan Parameters" section on page 41.

• Area file (.areax): Stores the size and shape of a single or multiple scan areas. It also stores the coordinates of the autogain and barcode areas.

The area file is stored in XML format.

You will learn more about setting scan areas in the "Specifying Scan, Barcode, and Autogain Areas" section on page 48.



The instrument software does not make any restrictions on overwriting, deleting, or modifying files. Use care when performing file handling tasks.

3.2 Using the Help Viewer

The Help Viewer provides background information, functionality overviews, and procedural assistance for the Microarray Scanner and instrument software. Click the Help button (?) or press F1 to access the Help Viewer.



Figure 22: Help Viewer

At the top of the Help Viewer, you find buttons that enable printing, navigating to the previous or next page, and setting options. In the left pane, the *Contents, Index*, and *Search* tabs provide browse and search functionality.

Contents tab. To browse through the table of contents, click the *Contents* tab. Click the "+" node next to a book icon to reveal sub-books and topic entries. You can navigate through the chapters by expanding/collapsing further nodes. Click a particular entry to display the corresponding topic.

Index tab. Click the *Index* tab to see a list of index entries, and either type a word or scroll through the list. Topics are often indexed under more than one entry. Click an index entry to display the corresponding topic.

Search tab. The *Search* tab allows you to type in the word or phrase for which you want to search for, and then click List Topics. Double-click a search result to display the corresponding topic.

3.3 Setting Software Options

The instrument software provides options for specifying the default storage location for image files, how long the lasers will remain on during periods of non-use before the instrument software automatically turns them off, and the default file format for scanned images.

To specify software options:

Click the Roche button () to display the Application menu and select Options. The Options window opens (Figure 23).

Settings Resources	Roche	ustomize your	version of Roche Nimb	ileGen MS 200	
	Save documents	Ed Date Mil	and a second second		Presso
	Deraun me rocation	ENDatauxin	nbleGen MS 200		Browsen
	Laser settings				
	Laser auto on ume	30	minutes		
	Image settings				_
	Image Format	Roche Fc	ormat		
					Cancel

Figure 23: Options Window

2. Review the default file location to which files are saved. To specify a different directory, click **Browse** and select the new location.

- **3.** Review the default laser auto off time that identifies the length of time (in minutes) the lasers will remain on during periods of non-use before the instrument software automatically turns them off. To specify a different length of time, type the value in the text box.
- **4.** Review the default file format. Currently "Roche Format" is the only available format.
- **5.** Click **OK** to save any changes. Or, click **Cancel** to exit without saving changes.

4. Specifying Parameters

In this section, you learn how to

- Work with the magazine control to identify the slides to scan.
- Set the parameters to use to scan the slides.
- Specify scan, barcode, and autogain areas.

4.1 Working with the Magazine Control

The Magazine Control (Figure 24) provides information for each slide loaded into the slide magazine.



Figure 24: Magazine Control

Slot. Positioning of slides in the slide magazine slots, numbered 1 - 48 from the top down. A maximum of 48 positions can be addressed.

Scan Number. Identifies the number of times to scan the slide. Refer to page 45 for information on how to add cycles.

Slide Present. Each position in the slide magazine is automatically identified as empty or occupied. An occupied position is identified by a green box, an empty position by a light gray box. A red box indicates the instrument software is in the process of checking the status of the slide position.

Select Scan. Select the checkbox to choose the slide for scanning.

Image Exists. After scanning, red and/or green dots corresponding to the laser's color indicate that images in the respective channel were acquired and saved successfully.

Display Images. A blue **Display Images** button () appears after the image has been acquired. Move the mouse pointer over the button to display the directory location and file name of the acquired image. Click the button to display the image in the Slide View and Image View.

Parameters. Scan parameters are assigned to a letter code in alphabetical order. The same letter corresponds to same applied parameter settings. For information on how to change parameters, refer to the "Specifying Scan Parameters" section, below.

Resolution. Displays the selected scan resolution.

Laser. Red and green dots represent selected laser channels.

4.2 Specifying Scan Parameters

Go to the row of the slide and click its Open Parameter Control button

 (③) in the Magazine Control (Figure 24).

If not already open, the Parameter Control opens and provides options and controls for specifying all scan parameters. The following figure identifies the components of the Parameter Control.

Open Parameter Control	Close Parameter Control		
Open Parameter Control Image: Control <th control<<="" td=""><td>Close Parameter Control</td><td> Open/Close Area Definition Control Slide Type Resolution Speed/Sensitivity Channel Gain Laser Intensity Autogain Apply parameter settings to slide(s) </td></th>	<td>Close Parameter Control</td> <td> Open/Close Area Definition Control Slide Type Resolution Speed/Sensitivity Channel Gain Laser Intensity Autogain Apply parameter settings to slide(s) </td>	Close Parameter Control	 Open/Close Area Definition Control Slide Type Resolution Speed/Sensitivity Channel Gain Laser Intensity Autogain Apply parameter settings to slide(s)

Open/Close File Settings Control

Figure 25: Magazine Control (Left) and Parameter Control (Right)

2. Review and as necessary adjust the scan parameters:

Slide Type. The only selection available is "Roche NimbleGen." Additional selections could be available in the future.

Resolution. Select from 2, 5, 10, 20, and 40µm resolutions.

Speed/Sensitivity. Adjusts the scan speed. Your selection has an impact on the overall results based on the speed and sensitivity of the scan. Click the slider in the following locations to select one of these settings:

- Far left = *Fast/Low* (24 lines per second (12Hz) parallel)
- Mid left = *Accelerated/Standard* (24 lines per second (12Hz) sequential)
- Mid right = *Medium/Advance* (16 lines per second (8Hz) parallel)
- Far right = *Slow/High* (16 lines per second (8Hz) sequential)

When scanning in parallel mode, the Microarray Scanner collects data for both channels at the same time. With sequential mode, it performs two individual scans: one for each channel.

Channel. Select the laser channel (532 or 635nm).

Gain. Adjust the gain for the photomultiplier tubes (PMTs) from 0.01 to 1000% in small or large increments.

Laser intensity. Select laser intensity at 0.1, 1, 10, 25, 50, 75, or 100% for each channel.



It is recommended to keep the laser intensity set to 100% and adjust the PMT gain setting if necessary.

Autogain. Select this checkbox to activate the autogain procedure. During the autogain procedure, the instrument software scans the specified autogain area and equalizes the intensity values for the channels.

Apply to slide number x and cycle x. Select this option to apply parameter settings only to the selected slide and corresponding slide cycle.

Apply to slides with parameter set. Select this option to apply parameter settings to all slide positions with the same parameter letter code in the Magazine Control.

- **3.** To save any changes to the settings, click **Apply**. Or, click **Reset** to cancel changes and leave the parameter settings unchanged.
- 4. If desired, click Save to save settings to a parameters file for future use.

4.2.1 Assigning the Same Scan Parameters to Multiple Slides

You can easily assign the same scan parameters to multiple slides.

To assign using the *Apply to slides with parameter set* option button:

Select the *Apply to slides with parameter set* option button in the Parameter Control (Figure 25) to apply parameter settings to all slide positions with the same parameter letter code in the Magazine Control.

To assign using the copy and paste functionality:

- 1. If open, close the Parameter Control by clicking the Close Parameter Control button ((), Figure 25).
- **2.** Go to the row of the slide with the preferred parameters in the Magazine Control, right-click, and select **Copy** from the menu.



- **3.** Highlight the rows for the slides to process using the same parameters, right-click, and select **Paste Parameter Settings** from the menu.

To highlight multiple slide rows, hold down the Ctrl key and click each slide's row. Or, click a row and then hold down the Shift key to highlight a range of slides.

If the slide with the preferred parameters contains more than one cycle, use the copy and paste functionality to specify the complete slide definition.

To assign using a parameters file:

You can apply the settings from an existing parameter file to the selected slide(s).

1. Highlight the slide row in the Magazine Control (Figure 24).

- 2. If necessary, open the Parameter Control (Figure 24) by clicking the slide's Open Parameter Control button (◊) in the Magazine Control.
- **3.** In the Parameter Control, click Load.
- 4. Choose the parameter file and click Open.
- **5.** Click **Apply** to confirm.

4.2.2 Processing the Same Slide Multiple Times

You can scan an individual slide a maximum of 12 times. Each time the slide is processed is referred to as a *cycle*.

To create a cycle:

- 1. If open, close the Parameter Control by clicking the Close Parameter Control button ((), Figure 25).
- **2.** Go to the slide row in the Magazine Control (Figure 24), right-click, and select Add Cycle.

	Scan Select Scan Unselect	
	Add Cycle	
	Delete Cycle	
b	Сору	
	Paste Parameterset	
2	Paste Slide	

3. Specify parameters for the cycle as described in "Specifying Scan Parameters" on page 41.

To delete a cycle:

- 1. If open, close the Parameter Control by clicking the Close Parameter Control button ((), Figure 25).
- **2.** Go to the slide row in the Magazine Control (Figure 24), right-click, and select Delete Cycle.

5. Specifying Image File Settings

The instrument software saves images as a TIFF file (Tagged Image File Format, .tif). In the File Settings Control, you can assign additional information to image file names. You can specify properties, methods, or user-defined text to annotate file names to help identify images. This capability improves consistency and avoids naming conflicts across a batch scan.

- **1.** Highlight the slide row in the Magazine Control (Figure 24).
- 2. If necessary, open the Parameter Control (Figure 25) by clicking the slide's Open Parameter Control button (♥) in the Magazine Control.
- In the Parameter Control, click the Open/Close File Settings button
 (()) to open the File Settings Control (Figure 26).



Figure 26: File Settings Control

- 4. Choose between the following option buttons:
 - *Single-TIFF*: Generates one image file in Tagged Image File Format (TIFF, .tif) per channel.
 - *Multi-TIFF*: Generates one image file in TIFF containing both channels.



For NimbleGen arrays, it is recommended to select the Single-TIFF option button.

- 5. If necessary, change the path to which the images files will be saved. The default path is E:\Data\NimbleGen MS 200. To make changes, click Browse to open a dialog box to specify a location in the directory and click OK to confirm. Otherwise, click Cancel.
- 6. Use the annotation list to add or change annotations to include in file names. Click an annotation in the left list box and then click the right arrow button to add to the right list box. To remove an annotation from the list, select the annotation in the right list box and click the left arrow button.



Make sure to select the "Barcode" annotation (i.e. include it in the right list box) to read the barcode label information on the slide.

7. Once your selection is complete, optionally change the order of the annotations in the file name. Click the annotation in the right list box and click Up or Down to adjust its position.



If you will be using NimbleScan software for data analysis, order the annotations as follows:

<Barcode> <User Text> <Laser WL>.tif

where "WL" means wavelength.

- 8. If you selected to include the date, select a format in the Date Format field.
- 9. If you selected to include user-specified text, type the text in the User Text field.
- **10.** Click Apply to confirm settings.
- 11. To close the File Settings Control, click the Open/Close File Settings button (③). The Filename Preview text box of the Parameter Control displays the entire naming convention of the image file (.tif).

6. Specifying Scan, Barcode, and Autogain Areas

The instrument software includes an Area Definition Control that provides functionality to set scan, barcode, and autogain areas. This functionality helps to automate routine scanning because the settings can be saved for future use.

To display the Area Definition Control:

- 1. Highlight the slide row in the Magazine Control (Figure 24).
- 2. If necessary, open the Parameter Control (Figure 25) by clicking the slide's Open Parameter Control button (◊) in the Magazine Control.
- In the Parameter Control, click the Open/Close Area Definition Control button (2) to open the Area Definition Control (Figure 25). The following figure identifies the components of the Area Definition Control:



Figure 27: Area Definition Control



The barcode and autogain functionalities are not activated using the *Barcode area* and *Autogain area* checkboxes in the Area Definition Control. Selecting these checkboxes allows for the definition of the barcode and autogain areas.

To activate the barcode functionality, specify "Barcode" as part of the image file name using the File Settings Control (Figure 26). To activate the autogain functionality, select the *Autogain* checkbox in the Parameter Control (Figure 25).



Do not adjust the barcode area if using NimbleGen arrays. The default location and size of the barcode area has been designated specifically for these arrays.

6.1 Specifying a Single Scan Area

The scan area determines the region on the slide to scan. It should be large enough to capture the entire feature region on the slide.



For NimbleGen 2.1M, 3x720K, and 12x135K arrays, use the default settings for the scan region.



For NimbleGen 385K and 4x72K arrays, it is recommended that you reduce the scan area to reduce the scan time.

To define and adjust a single scan area on your microarray:

- 1. If necessary, highlight the slide row in the Magazine Control (Figure 24).
- **2.** If necessary, open the Parameter Control (Figure 25).
- If necessary, open the Area Definition Control (Figure 28). 3.
- 4. Select the *Single Area* option button.



Figure 28: Area Definition Control for Specifying a Single Scan Area

- **5.** Capture the entire designated feature area. Move the mouse pointer inside the white scan area rectangle and click to enable the move cursor.
- **6.** Drag the rectangle over the desired feature region. Adjust the size of the rectangle by dragging a corner/side handle.
- 7. Click Apply to confirm your settings.
- 8. If desired, click Save to save settings to an area file for future use.

To use an existing array image as a template for your area layout:

Choose between the following:

To load an image associated with the current session, go to the slide row in the Magazine Control and click the Display Images button () in the slide's Display Images column. The image appears in the Image View.

To use the scan area(s) provided by an existing area file:

If you are confident with the scan area(s) saved to an area file, you can apply the settings in the file.

- 1. Click Load in the Area Definition Control (Figure 28).
- **2.** Select and open the area file.
- 3. Click Apply to confirm your selection.

6.2 Specifying Multiple Scan Areas

If you are using NimbleScan software for data analysis, Roche NimbleGen recommends specifying a single scan area instead of multiple scan areas. For NimbleGen multiplex arrays, you use the software's burst functionality to create individual image files for the arrays.

To identify multiple areas:

- **1.** If necessary, highlight the slide row in the Magazine Control (Figure 24).
- 2. If necessary, open the Parameter Control (Figure 25).
- **3.** If necessary, open the Area Definition Control (Figure 29).
- 4. Select the *Multi Area* option button.

A magenta feature rectangle and a white scan area rectangle are displayed. Both bounding rectangles are merged at their top-left corners to capture and select multiple scan areas.



Figure 29: Area Definition Control for Specifying Multiple Scan Areas

- Position the mouse pointer inside the magenta feature area rectangle. Drag both bounding rectangles to the upper-left corner of the microarray features.
- **6.** Drag the merged rectangles to the top-left segment of the printed area to identify the area to scan. Adjust the size of the magenta rectangle by dragging a corner handle.
- 7. Use the white scan area rectangle to enclose the top (left) feature segment on the array. This is used to set size and shape of all other scan areas. Drag a corner handle or a side handle until that bounding rectangle matches the desired feature size.

- **8.** Specify rows and columns by clicking the arrow buttons according to the array layout. Columns and rows are separated by intersecting lines that give rise to multiple scan area segments wherein additional single scan areas are then displayed.
- **9.** Make sure that the height and width of your initial scan area enables displaying multiple scan areas within the predefined magenta feature area.
- **10.** If necessary, clear the *Link to Geometry* checkbox:

By default, the *Link to Geometry* checkbox is selected. If you resize the initial scan area, the instrument software automatically scales all other areas to the same dimension.

Clear the *Link to Geometry* checkbox to disable this functionality. In each corner and side of a scan area, handles will appear to adjust each scan area individually.



- **11.** Click **Apply** to confirm your settings.
- **12.** If desired, click **Save** to save settings to an area file for future use.

To use an existing array image as template for your area layout:

From the Application menu, select **Open Image** and import the image from the file directory to the Image View. Then click **Show Image** to display the image in the Area Definition View.

Alternatively, click the **Display Images** button () in the Magazine Control in your current session or open a session to retrieve an image. Then click **Show Image** to display the image in the Area Definition View.

To use the scan area(s) provided by an existing area file:

If you are confident with the scan area(s) saved to an area file, you can apply the settings in the file.

- 1. Click Load in the Area Definition Control.
- **2.** Select and open the area file.
- **3.** Click **Apply** to confirm your selection.

6.3 Specifying the Barcode Area

- Do not adjust the barcode area if using NimbleGen arrays. The default location and size of the barcode area has been designated specifically for these arrays.
- **1.** If necessary, highlight the slide row in the Magazine Control (Figure 24).
- 2. If necessary, open the Parameter Control (Figure 25).
- **3.** If necessary, open the Area Definition Control (Figure 29).
- 4. Ensure the *Barcode area* checkbox is selected.
- **5.** Position the mouse pointer inside the respective rectangle and click to enable the move cursor.
- **6.** Drag the barcode rectangle to the desired position on the array. The barcode rectangle is a fixed size of 22 x 7mm.
- 7. Click Apply to confirm your settings.

6.4 Specifying the Autogain Area



The autogain area cannot be less than 3 x 3mm or greater than 22 x 22mm.

- Make sure that the autogain area is located on a feature area when determining PMT gain values automatically. Do not place the autogain area offset between or outside of the feature area to avoid too high or too low PMT gain values.
- **1.** If necessary, highlight the slide row in the Magazine Control (Figure 24).
- **2.** If necessary, open the Parameter Control (Figure 25).
- **3.** If necessary, open the Area Definition Control (Figure 29).
- 4. Ensure the Autogain area checkbox is selected.
- **5.** Position the mouse pointer inside the respective rectangle and click to enable the move cursor.
- **6.** Drag the rectangle to the desired location. As a recommendation, ensure the rectangle is approximately 10mm from the edge of the default scan area.
- **7.** Size the rectangle by dragging the side and corner handles. The following figure provides guidance as to the size of and where to locate the autogain area for NimbleGen arrays.



Figure 30: Examples of Setting the Autogain Area for NimbleGen Arrays

8. Click **Apply** to confirm your settings.

7. Starting and Aborting a Scan

7.1 Before You Start the Scan

- Close any other software that is currently running. The instrument software could stop the scan if the computer's memory becomes overloaded.

Make sure that you are not running out of disk space on your dedicated hard disk drive. Free space on this drive by backing up data or deleting old or unnecessary files.



Ensure that each slide to process is selected in the Magazine Control (Figure 24).

7.2 Starting a Scan

Click the Start/Stop Scan button (\blacktriangleright) in the Scan Control (Figure 19).

Specify the folder and file naming to save the session file. The scan process is then initiated.

If the lasers are not fully warmed up, the Laser Warmup Time Active window opens, asking you to wait. After the necessary warmup time, this window closes, and the scan starts automatically. Click Continue to ignore this advice and immediately proceed with the run. Or, click Cancel to return to the instrument software workspace.



Selecting to continue the run before the lasers have warmed up could affect data quality.

C	Laser WarmUp Information
	Laser Warmup Time Active
	Laser 1 green (532) nm 09 min 53 sec
	Laser 2 red (635) nm 09 min 54 sec
	Laser warmup time is active. Do you want to wait until warm-up time is reached, continue anyway or cancel the measurement?
	Continue

Figure 31: Laser Warmup Time Active Window

Once scanning begins, the Parameter Control and Area Definition Control close, and the Image View, Slide View, and Spot View open. The current task that is being performed is identified above the progress bar.

The Slide View shows all specified scan areas for the currently loaded slide. As the image is acquired, it appears from the bottom upward in the Slide View.



For 5, 10, 20, and $40\mu m$ scans, the Image View mirrors the Slide View, and the Spot View is functional during the run.



For $2\mu m$ scans, the Image View and Spot Views are not functional during the run.

After the scanning experiment is completed, the light in the upper-left corner of the insert/eject magazine button becomes green (Figure 10). Microarray Scanner unlocks the stacker cover, and you can remove the slide magazine.



Figure 32: Run-Time Window

Manually Adjusting the PMT Gain during a Scan

While scanning, *Gain 1* and *Gain 2* spin boxes appear in the Scan Control to enable you to adjust the PMT gain for each channel. To increase or decrease the PMT gain value, click the arrow buttons to incrementally change the value. In the Image Control, select the respective channel to see the immediate effects on the acquired signal.



If you adjust the PMT gain value(s) during the scan, Roche NimbleGen recommends to rescan the slide using the new settings.

For NimbleGen arrays: If using NimbleScan software to extract and analyze data, ensure the fiducial controls are visible and near or above the signal intensity of the surrounding experimental features to help to ensure proper grid alignment. Features with saturated pixels are drawn in magenta on the image. Saturated fiducial controls can cause poor alignment quality.

7.3 Aborting a Scan

At any time, you can stop the scan or batch run. Click the **Start/Stop Scan** button (\triangleright) in the Scan Control (Figure 19). It can take some time to stop the scan, depending upon the stage of the scan process. Any collected data are saved in the default or user-defined file directory for later analysis.

7.4 Saving Log Files

If you encounter errors or exceptions, or if you suspect problems with the instrument or settings that you are using, save the log files that record actions of the scanning operation.



Figure 33: QC Tab Showing the Save Logfiles Button

Click the QC tab and click **Save Logfiles** to save as a .zip file to a userdefined location.
All relevant information, such as error messages and communication and system information (e.g. operating system version, amount of available disk space), are saved and are helpful when troubleshooting any issues with a Roche Field Service Engineer.

8. Reviewing Images

You can explore 5µm and above images during and after the scanning experiment. Two micron images can only be examined after the experiment.

8.1 Displaying an Image

Choose between the following:

- Click the Display Images button () in the Magazine Control (Figure 24) or open another session and click the Display Images button ().
 - The Display Images button links to the directory in which the image was originally saved for the session. If the image's file name is modified or if the file is moved, clicking the Display Images button (
- Click the Roche button (^(C)), select Open Image, and retrieve the image from the file directory.

8.2 Zooming in and out

Select a scan area and zoom out or drag the magenta frame in the Slide View to display an enlarged detail of the captured features in the Image View (Figure 34).



Figure 34: Spot, Slide, and Image Views

8.3 Displaying Intensity Data for a Feature

Position the cursor over a location in the Image View to activate the Spot View (Figure 34). The red crosshair in the Spot View can be placed over a specific feature by moving the cursor within the Image View. This will render intensity data for both channels and the coordinates for that feature.

8.4 Adjusting Image Appearance

You can adjust contrast and brightness using the Image Control (Figure 35) to have a better view of features on the image. Adjusting these settings allows faint features to be more easily seen.



Figure 35: Image Control

To automatically adjust the contrast to enhance the image:

Click Best Fit to automatically adjust contrast values for better discrimination between the brightest signal and the darkest background pixels of the image.

To manually adjust gamma, brightness, and contrast of the image:

Use the Gamma, Brightness, and Contrast sliders to change the settings.

- The Gamma slider adjusts the linearity of the displayed pixels to the scanned intensity.
- The Contrast slider adjusts the difference between dark and bright pixels.
- The Brightness slider adjusts the overall displayed intensities.



Adjusting the gamma, brightness, and contrast settings does not modify the pixel values in the saved image. They only adjust the way in which the image is displayed.

8.5 Displaying a Histogram or Line Profile

Using the Image Control (Figure 35), you can generate histograms or line profiles. Histograms show an intensity frequency profile for a specified region. Line profiles show an intensity profile for a specified vertical or horizontal section.

To display a histogram:

Click Show Histogram to open the Histogram window (Figure 36 and Figure 37). The histogram shows the pixel's intensity distribution of the currently selected area in the Slide View. The displayed color on the histogram corresponds to the selected channel color. You can zoom the boundaries of the histogram range by dragging the slider bars in the histogram window. Linear and Logarithmic Base 10 option buttons allow you to select the scale on the Y axis for the histogram.

For 5μ m and above scans, you can use the PMT gain in conjunction with the histogram to balance or match up the channels during the run. With each adjustment to the PMT gain, the instrument software resets the histogram to reflect the changes in pixel intensity.



Histogram mode is not available for 2μ m scans during the run but is available post run.

The following figure shows a histogram generated during a scanner experiment in which the channels are unbalanced. The Gain 1 and Gain 2 spin boxes enable you to adjust the gain during the scan.

Gain 1 and 2 spin boxes			
(ka)) =			NimbleGen MS 200
Scan Oc Gain 1 Gain 2 2 50 % 400 % 2	Scan	Laser green	Boo Hastogram Overlay Gamma: 4.1 Channel 1 Channel 2
👸 Histogram		-	
Information Start: 0 End: 65536 V-Axis Scale Logarithmic Base 10 Auto Scale	6607 2616 1165 1267 199 34 14 2 1 100 100 100 100 100 100 100 100 100	45445	Zoon Contraction of the second s

Figure 36: Histogram Generated During the Run

Figure 37 shows a histogram generated after the scanner experiment in which the channels are balanced.



Figure 37: Histogram Generated After the Run

To display a line profile:

Use the line profile command to obtain a cross section of intensity values along a line drawn in the image. When clicking the Show Vertical Line Profile or Show Horizontal Line Profile button, the Line Profile window opens (Figure 38). A magenta defining-line is placed within your image in the selected channel mode. Use the mouse to position the defining-line over the pixels to measure. The line can be drawn to any length by dragging either endpoint. You can zoom the boundaries of the line profile plot range by dragging the slider bars in the Line Profile window. *Linear* and *Logarithmic Base 10* option buttons allow you to select the scale on the Y axis for the line profile.



A line profile is not available for $2\mu m$ scans during the run but is available post run.



Figure 38: Line Profile Window

8.6 Choosing the Channel(s) to Display

The *Overlay, Channel 1*, and *Channel 2* option buttons in the Image Control (Figure 35) enable you to view images in single channel or overlay mode.

D Maintenance and Care

The maintenance and care procedures are important to prolong the instrument's life and reduce the need for servicing. This chapter contains information about the following:

- ILC test
- Liquid spill cleanup
- Ozone filter replacement
- Instrument relocation
- Instrument disinfection

1. ILC Test

If this is the first time you are running the ILC test, refer to page 27 for more detailed instruction.

Roche NimbleGen recommends you perform an ILC test every 2 weeks.

- 1. Switch on the Microarray Scanner.
- 2. Start the instrument software.
- **3.** Click the QC tab and then ILC in the instrument software. The Integrated Laser Check window opens (Figure 16).
- **4.** Click the **Start/stop ILC procedure** button ().

After the ILC test is completed, the instrument software produces and displays a comprehensive test report containing all measured parameters, evaluation limits, and results (PASS or FAIL).

For more information on the checks that occur during the ILC test, refer to "ILC Module" on page 4.

2. Liquid Spill Cleanup



Always switch off the instrument before removing any kind of spills. All spills must be treated as potentially infectious. Therefore, always adhere to applicable safety precautions (including the wearing of powder-free gloves, safety glasses, and protective clothing) to avoid potential infectious disease contamination.



All resulting waste from the cleanup must be treated as potentially infectious and the disposal must be performed according to the information given in *Chapter A*, *Overview*. If the spill occurs inside of the instrument, a service technician is required.

- **1.** Switch off the Microarray Scanner.
- Wipe up the spill immediately using absorbent material. 2.
- 3. Dispose of contaminated material appropriately.
- 4. Clean the instrument surfaces with a mild detergent.
- **5.** Disinfect the instrument as described in the "Instrument Disinfection" section on page 70.
- **6.** Wipe dry all cleaned areas.

3. Ozone Filter Replacement

The need for replacing the ozone filter units may vary with the usage of the microarray scanner. To ensure optimal performance, replacement of the ozone filter units will be included in the annual maintenance by your Roche Field Service Engineer or is available upon request.

4. Instrument Relocation

Before you move the instrument to a different location, remember to:

- 1. Remove the ILC slide stored inside the Microarray Scanner:
 - **a.** If necessary, switch on the Microarray Scanner.
 - **b.** If necessary, eject the slide magazine and remove the slide inserted into slot 48.
 - **c.** Start the instrument software.
 - **d.** Click the *QC* tab and then ILC in the instrument software. The Integrated Laser Check window opens (Figure 16).
 - e. Click the Remove ILC slide button (△) in the Integrated Laser Check window.
 - f. Close the instrument software and switch off the Microarray Scanner.
- **2.** Disinfect the instrument as described in the "Instrument Disinfection" section on page 70.



Treat all parts of the instrument that come into contact with biological samples or other hazardous material as potentially infectious areas.



Thoroughly disinfect the instrument before it is removed from the laboratory.

3. Pack the instrument. Use the original packaging to ensure that no damage occurs to the instrument.



Remove the slides from the magazine before you prepare the instrument for relocation. If a slide remains in the instrument, potential jamming or fragmentation of the slide could damage the instrument.



If the original packaging is lost or damaged, contact Roche NimbleGen for replacement packaging.

4. Complete the Certificate of Decontamination (refer to page 71) and affix to the outside of the package.

5. Instrument Disinfection

Authorized trained personnel, wearing disposable gloves, protective glasses, and protective clothing, should perform the disinfection procedure in a well-ventilated room. Be aware that the disinfectant can influence instrument performance if you apply it to the inside of the instrument.



Risk of fire and explosion. Alcohols, such as ethanol or isopropanol, are flammable and when improperly handled can lead to explosions and/or fire. Proper laboratory safety precautions must be observed.



The disinfection procedure should be performed according to national, regional, and local regulations.



Perform the disinfection procedure when cleaning up a spill and when relocating the instrument.

- 1. Disconnect the instrument from the main power supply.
- 2. Disconnect the instrument from any accessories.
- **3.** Carefully wipe all the outside surfaces of the instrument with a clean lint-free cloth that has been soaked in the disinfecting solution, such as:
 - Lysetol (Schülke & Mayr, Norderstedt/Germany, www.schuelke-mayr.com)
 - Aseptisol (Bode Chemie, Hamburg/Germany, *www.bode-chemie.com*)
 - 70% ethanol, if neither of the above are available
- 4. Perform the same disinfection procedure on the slide magazine.
- **5.** Repeat the disinfection procedure on any accessories.
- **6.** Complete the Certificate of Decontamination (refer to page 71) and affix to the outside of the package.

Certificate of Decontamination

I declare that the instrument in this package has been decontaminated or disinfected to
remove or inactivate any biological material, which could be dangerous to personnel, or
that it has never been exposed to any hazardous biological material.
Contact person
Company:
Function:
Phone/Fax:
Email:
Date of decontamination:
Method of decontamination applied:
Date:
Signature:

Notes

E Appendix

1. Troubleshooting

The troubleshooting table below lists possible malfunctions and errors of the Microarray Scanner and provides corrective measures on how to resolve them. The operator can correct some problems or errors without having to contact Roche Microarray Technical Support. For these situations, appropriate actions are listed in the *Corrective Measure(s)* column.

The elimination of more complicated malfunctions or errors is usually performed by Roche NimbleGen according to separate instructions. For these situations, the *Corrective Measure(s)* column directs you to contact Roche NimbleGen.

Problem/Error	Possible Cause(s)	Corrective Measure(s)
Instrument-Level Troub	leshooting	-
Communication error occurred.	Power is not on.	Switch on power to the Microarray Scanner using the main power switch on its rear and using the on/off button on its left side.
	Power/communication was interrupted.	Check cable(s) and plug to ensure they are securely connected. Switch off then switch on power to the Microarray Scanner and computer.
	Windows Vista or local network firewall may prevent communication.	Disable or modify the settings for the firewall. Contact your IT Department if necessary.
	The TCP/IP network properties for the Microarray Scanner may have been altered.	Verify the Microarray Scanner's IP address for the local area connection is set to 169.254.101.202 and its subnet mask is 255.255.0.0. Contact your IT Department if necessary.

Problem/Error	Possible Cause(s)	Corrective Measure(s)
Data loss occurred while scanning causing a scan abort.	Insufficient communication is occurring between the computer and Microarray Scanner.	Verify the provided Ethernet cable is securely connected to the computer and Microarray Scanner. Do not connect the computer and Microarray Scanner to a hub or repeater device. Make sure the computer's network card is configured as a 1GB Ethernet port (RJ45).
Stacker cover does not unlock properly.	Slide cover clashes with the magazine's handle bar. Slide magazine not properly inserted.	Switch off power to the Microarray Scanner. Carefully, push back the stacker flap and lower the slide magazine beyond the mechanical hold point. Switch on power to the Microarray Scanner and wait for initialization to complete. If the problem persists, contact Roche Microarray Technical Support.
Load/unload of slide could not be done successfully.	Mechanical failure occurred.	Follow the instructions in the "Removing a Slide from the Slide Transport" section on page 45 to manually remove the slide. If the problem persists, contact Roche Microarray Technical Support.
No slide was detected.	The slide detection sensor is not able to recognize a slide in that slot.	Unload the slide magazine from the Microarray Scanner. Check slides for damages and correct positioning in slots. Reload and initialize the slide magazine. If the problem persists, contact Roche Microarray Technical Support.

Problem/Error	Possible Cause(s)	Corrective Measure(s)
Shift/misalignment of pixels was apparent in the image.	Instrument needs recalibration.	Switch off then on power to the Microarray Scanner and wait for initialization to complete. Run the ILC test to recalibrate the instrument. If all test points pass successfully, scan a test slide to verify that pixel shift has been corrected. If the problem persists, contact Roche Microarray Technical Support.
Processing-Level Troub	leshooting	
Autofocus procedure failed.	Slide does not match selected slide type.	Verify the slide matches predefined slide type options.
	Slide is not present.	Verify the slide is present in the slide magazine.
	Particles, dust, or finger prints are present on the slide.	Check the slide surface. If possible, remove dust and/or particles from the slide by appropriate methods. (For NimbleGen arrays, refer to the <i>NimbleGen Arrays User's Guide</i> for instructions.) Repeat scan. If the problem persists, run the ILC test. If the problem persists, contact Roche Microarray Technical Support.
The array image appears too bright after applying autogain.	Autogain area is not entirely located with a feature area.	Place the autogain area in a feature area. Refer to page 56 for instructions on how to specify the autogain area. Repeat scan.
	Dust or particles within the autogain area affect the autogain procedure.	Check the slide surface. If possible, remove dust and/or particles from the slide by appropriate methods. (For NimbleGen arrays, refer to the <i>NimbleGen Arrays User's Guide</i> for instructions.) Repeat scan.

Problem/Error	Possible Cause(s)	Corrective Measure(s)
The dark value scan could not be finished successfully.	There is too much noise on the signal.	Perform the ILC test. If the problem persists, contact Roche Microarray Technical Support.
No array image is created.	Slide was inserted with microarray side facing down.	Orient the slide in the slide magazine with microarray side facing upward.
	Improper gain setting(s) were specified.	Verify gain setting(s).
	Improper laser intensity setting(s) were specified.	Verify laser intensity setting(s).
	Poor fluorescence was realized.	Avoid degradation of labeled samples on the array due to exposure to light, ozone, and/or humidity. Correct settings for PMT gain and/or laser intensities. Run the ILC test.
Barcode reading failed.	No barcode is on slide.	Verify the slide has a barcode.
	Barcode is misplaced.	Check the position of the barcode area.
	Slide inserted in the wrong direction.	Check the insertion direction of the slide.
	Barcode type is not supported.	Check if the barcode type matches specifications for Code 39 or Code 128.
	Low contrast or soiled barcode.	Clean or wipe the barcode area. Check the quality of the etched barcode.
	Barcode area specified in the instrument software does not match barcode position on slide.	Check and/or adjust the barcode position in the Area Definition Control.

Problem/Error	Possible Cause(s)	Corrective Measure(s)
Computer-Level Trouble	eshooting	
The TIFF image from incoming scan data could not be generated.	Insufficient computer memory is available.	Check computer performance. Close any unnecessary software that is currently running. Verify the computer hardware meets minimum requirements.
Scan aborts.	Insufficient computer memory is available.	Close any other software that is running. Ensure the autogain area does not exceed 22 x 22mm. Verify the computer hardware meets minimum requirements.
Software-Level Troubles	shooting	-
Missing array images. No Display Images button is available.	Array images were removed from the default file location or renamed.	Check the assigned directory in the File Settings Control.
Instrument software shuts down unexpectedly.	The Microarray Scanner may be performing the initialization process for the slide magazine.	After inserting the slide magazine into the Microarray Scanner, wait for the initialization process to complete before starting the instrument software.

Removing a Slide from the Slide Transport

If you encounter a slide load/unload failure, follow these steps to remove the slide from the instrument:

1. Switch off power to the Microarray Scanner. Unscrew the two stacker locks at the rear of the instrument to release the front cover (Figure 39).



Figure 39: Unlocking the Stacker Locks

2. Open the instrument's front cover by pulling with both hands on both sides of the white part so that the front cover disengages and swings open to the right (Figure 40).



Figure 40: Opening the Front Cover

3. If accessible, manually remove the slide or slide fragments from the slide transport (Figure 41). Use vacuum to remove all slide fragments if necessary.



Figure 41: Removing the Slide from the Slide Transport

4. Close the instrument's front cover (Figure 42). Gently push the front cover into place, making sure to push it all the way into place. Two bolts and a pin secure the cover.



Figure 42: Closing Front Cover

5. Tighten the stacker locks at the rear of the instrument to secure the cover (Figure 43).



Figure 43: Tightening the Stacker Locks

6. Switch on the Microarray Scanner and wait for initialization to complete.

2. Ordering Information

To place an order, contact your Roche NimbleGen Account Manager. Go to *www.nimblegen.com/arraysupport* for contact information.

The following items are included with the purchase of a NimbleGen MS 200 Microarray Scanner system.

Catalog No.	Product
05394341001	MS 200 Microarray Scanner (1 magazine included)
05394325001	MS Control Unit
05394309001	MS 200 Operator Manual
05394333001	MS 200 Software

The following accessory can be purchased:

Catalog No.	Product	
05471478001	Microarray Scanner MS Magazine	

For a complete list of Roche NimbleGen products and services, go to *www.nimblegen.com*.

Technical Support

If you have questions about how to use the Microarray Scanner, contact your Roche NimbleGen Account Manager or Roche Microarray Technical Support. Go to *www.nimblegen.com/arraysupport* for contact information.

3. Quick Start

The quick start provides a general description of a typical workflow:

- Starting the scanner, control unit, and instrument software
- Loading microarray slides
- Specifying parameters
- Scanning
- Analyzing images

It also describes the instrument software workspace. Refer to chapters noted for more details.

3.1 Starting the Scanner, Control Unit, and Instrument Software

- 1. Start the control unit and log into your user account.
- 2. Start the Microarray Scanner (*Chapter B*, *System Description*).
- 3. Start the instrument software (*Chapter B, System Description*).
- **4.** In the instrument software workspace, switch on the laser(s) to use and allow them to warm for at least 10 minutes (*Chapter B*, *System Description*).

3.2 Loading Microarray Slides

Press the insert/eject magazine button on the front of the instrument to open the stacker cover, remove the slide magazine to load/unload slides, and reinsert the slide magazine. Learn more about slide handling and loading/unloading in Chapter B, System Description.



Do not start the instrument software during the initialization process that the Microarray Scanner performs after slide magazine insertion. Doing so may cause the software to shut down unexpectedly.

3.3 Specifying Parameters

The instrument software workspace (refer to page 83) displays a green rectangle in the Magazine Control for an occupied position (Slide Present) in the slide magazine. Choose a slide to scan during the next run by selecting the Select Scan checkbox. Specify scan parameters using the Parameter Control. Adjust the output file format, path for data storage, and the name generation setting using the File Settings Control. Define the scan, barcode, and autogain areas using the Area Definition Control (*Chapter C*, Operation).



For NimbleGen 2.1M, 3x720K, and 12x135K arrays, use the default settings for the scan region.



For NimbleGen 385K and 4x72K arrays, it is recommended that you reduce the scan area to reduce the scan time.

The parameter settings can be saved for future retrieval.

3.4 Scanning

Start scanning all selected slides by clicking Start/Stop Scan in the Scan Control. Store collected data in the default directory (E:\Data\NimbleGen MS 200) or specify another location in the File Definition Control (Chapter C, Operation).

3.5 Reviewing Images

Display the image from a successful scan by clicking the **Display Images** button () in the Magazine Control or load information from a previous scan run by clicking the **Roche** button () and opening the image file. Use the *Gamma, Brightness*, and *Contrast* sliders in the Image Control to enhance the visibility of features. Use the *Zoom function* in the Area Definition Control to change the magnification in the Area Definition View. In addition to the histogram of this (potentially magnified) area, the instrument software offers data assessment with vertical and horizontal line profiles (*Chapter C, Operation*).

3.6 Instrument Software Workspace

The instrument software workspace offers menus and a variety of options for viewing and editing scan specifications. Its controls are arranged to help you focus on setting scan parameters and reviewing scanned images (*Chapter C, Operation*).



4. Index

Index item page
Aborting a scan60
Actuators test4
Alexa dyes2
Application menu
Area Definition Control, components49
Area files
Description of area files
Relationship between session, parameter, and area files
Specifying parameters for area files
Autofocus
Autofocus test
Description of autofocus procedure1
Troubleshooting75
Autogain
Activating the autogain functionality43
Dimensions of the autogain area56
Specifying the autogain area56
Barcode
Acceptable formats
Activating the barcode reader functionality47
Dimensions of barcode area9, 80
Location of the barcode area for NimbleGen arrays55
Specifying the barcode area
Troubleshooting
Barcode reader test4
Blank spots, non-NimbleGen microarrays9
Brightness, adjusting
Certificate of Decontamination
Channels, specifying for a scan
Cleanup, liquid spill
Computer specifications
Contrast, adjusting63
Conventions used in this manualiv

Cy dyes2
Cycle
Decontamination CertificateSee Certificate of Decontamination
Detection, description
Detector(s) test
Dimensions, instrument
Directive 2002/96/ECviii
Disinfection instructions
Disposal
Consumablesviii
Instrumentvii
Distributori
DY dyes2
Electrical specifications, instrument
Electronics test
Emission filters, description
Environmental specifications, instrument
Features, displaying intensity data
File Settings Control, components
Files
Area files
Parameter files
Saving log files
Session files
Setting the default format for image files
Setting the default storage location for image files
Specifying naming conventions for image files
Gain See PMT gain
Gamma,adjusting
Geometry linearity test
Handling requirementsv
Help Viewer, using
Histogram
- Availability of the histogram during a scan
Displaying a histogram
Using the histogram to adjust PMT during scan
Icon, MS 200

ILC module

Actuators test4
Autofocus test
Barcode reader test
Description of the ILC module
Detector(s) test
Electronics test4
Geometry linearity test
Initial ILC test
Laser(s) test
Optical alignment test
Optical resolution test
Oscillator test
Positioning test
Sensitivity test
Setting run parameters
ILC slide
Diagram of the ILC slide
Inserting the ILC slide into the slide magazine
Performing the initial ILC test
Proper handling of the ILC slide
Removing the ILC slide from the instrument
ILC test
Example report
How often to perform the ILC test
Images
Adjusting contrast, brightness, and gamma62
Choosing the channe(s) to display
Displaying intensity data for a feature62
Reviewing Images
Specifying file naming conventions for images
Troubleshooting75
Zooming in and out of images61
Information labels, rear panelvi
Installation, control unit
Installation, instrument
Humidity conditions
Power requirements
Space requirements
Temperature conditions

Unpacking the instrument 19
Warning and recommendation19
Installation, instrument software
Instrument
Front panel, description12
Left side, description
Moving the instrument to a different location
Rear panel, description
Troubleshooting
Instrument software
Access rights
General considerations
Installation directory
Installation information
Installation instructions
Safety instructions
Saving user-defined data
Setting defaults/options
Target computer
Typical scanner experiment
What users must know
Who should install
Workspace description
Intended use, instrumentii
Intensity data, displaying
Keyboard specifications
Lasers
Laser excitation, description
Laser timeout setting
Laser(s) test
Specifying the laser intensity for a scan
Turning on the lasers
Line profile
Availability of the line profile during a scan
Displaying a line profile
Log files, saving
Magazine Control, components
Manual conventions
Manufactureri
Monitor specifications

Mouse specifcations
Mouse specifications
Non-NimbleGen arrays, recommendations9
Operating system
Optical alignment test
Optical resolution test
Optical specifications, instrument
Ordering information79
Oscillator test
Ozone filter, replacement
Parameter Control, components
Parameter files
Assigning scan parameters to a session using an existing file
Description of parameter files
Relationship between session, parameter, and area files
Saving parameter files
Specifying scan parameters
Password requirements
Performance specifications, instrument
Photobleaching
Photodetectors
Photomultiplier tubes
Photomultiplier tubes
Photomultiplier tubes
Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan
Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan
Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan
Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan
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Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan
Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan
Photomultiplier tubes See PMTs PMT gain Adjusting the PMT gain during a scan

Scan area
Specifying a single scan area
Specifying multiple scan areas
Scan parameters
Assigning scan parameters using an existing file
Specifying scan parameters for multiple slides
Specifying scan parameters for one slide 41
Scanned imagesSee Images
Scanner experiment, typical steps
Scanning
Aborting a scan
Adjusting the PMT gain during a scan60
Before you start the scan
Saving log files
Starting a scan
Sensitivity test
Session files
Creating, saving, and opening session files
Description of session files
Relationship between session, parameter, and area files
Slide
Barcode dimensions
Inserting the ILC slide into the slide magazine
Recommendations for non-NimbleGen microarrays
Slide dimensions
Slide mishandling/handling
Specifying a single scan area
Specifying multiple scan areas
Specifying the autogain area
Specifying the barcode area
Specifying the slide type for a scan
Slide magazine
Components of the slide magazine
Inserting or removing the slide magazine from the instrument
Inserting slides into the slide magazine14
Slide magazine box14
Slide transport, removing slide from77
Space requirements, instrument
Specifications, control unit

Specifications, instrument
Electrical7
Environmental7
Mechanical
Optical
Performance
Physical7
Processing
Specifications, microarray slides
Speed/sensitivity, specifying for a scan42
Spills, cleanup
Start menu
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
Starting the control unit, scanner, instrument software
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