

QX-302 User Manual

QuantomiX Ltd. 2005. All rights reserved.

This publication is the copyright of QuantomiX ltd. and contains information that may not be used or reproduced unless agreed in writing. QuantomiX ltd. reserves the right to alter without notice the specifications, design or supply of any product or service.

The information provided in this User Manual is believed to be accurate. It is the user's responsibility to confirm the technical aspects and the suitability of the technology for any particular application.

QuantomiX Ltd., 12 Hamada St., Tamar Science Park, Rehovot

POB 4037, Nes-Ziona 70400, Israel

Tel: +972-8-946-2244, Fax: +972-8-946-5874

http://www.quantomix.com

Table of Contents

1	Safety		7
2	Technica	al Data	9
	2-1	QX-302 Capsules	9
	2-2	Capsule Plates (MP-12)	12
	2-3	Imaging Buffer (IB-74)	13
	2-4	Spacers	13
3	Introduc	tion	15
	3-1	Manual Scope and Contents	15
	3-2	References	
	3-3	Technology and Components	17
4	Using th	e QX-302 Capsules	23
	4-1	Getting Started	23
	4-2	Specimen Preparation	24
	4-3	Inserting Specimens into the QX-302 Cap	sule .27
	4-4	Storing Specimens in the QX-302 Capsule	ə30
5	Imaging		31
	5-1	Optimal Imaging Conditions	
	5-2	Recommendations for SEM Imaging with	
		QX- 302 Capsules	34
6	Appendi	ces	35
	6-1	Appendix A: Protocols for Specific Applica	ations 35
	6-2	Appendix B: Glossary	
	6-3	Appendix C: Troubleshooting	
	6-4	Appendix D: Ordering Information	
	6-5	Appendix E: Legal Notices	
		-	

List of Figures

Figure 1. Sealed Capsule - Dimensions	10
Figure 2. The Capsule Plate	12
Figure 3. QX-302 Capsule Parts	18
Figure 4. QX-302 Capsule, Positioned in the Capsule Plate	
Figure 5. Opening the QX-302 Capsule	28
Figure 6. Placing a Sample in the Specimen Dish	29

List of Tables

TABLE 1: Material Compatibility Table	11
TABLE 2: Chapters and Appendices in the User Manual	16
TABLE 3: Features Enhanced by Different Staining Agents	26
TABLE 4: Recommendations for SEM Imaging with QX-302	
Capsules	34
TABLE 5: Troubleshooting	37

Cited registered trademarks: Whatman[®] Tween[®] Auto/Zyme[®] VibratomeTM Triton[®]

All trademarks are the properties of their respective owners.

Chapter 1: Safety

Cautions

Please observe the following cautions while using the QuantomiX QX-302 capsules:

- Correct sealing of the capsule is essential for its proper functioning. To ensure that correct sealing is achieved, please note that the wings of the middle section of the capsule should be aligned with the wings of the upper and lower sections.
- The QX-302 sealing stub includes an O-ring, a circular rubber ring found in the lower section of the sealing stub. If the O-ring accidentally detaches from the sealing stub, it should be re-positioned inside the sealing stub (see Figure 3. on page 18).
- Use powder-free gloves to maintain cleanness and sterility of the QX-302 capsules. Powdered gloves should be avoided.
- 4. Do not place the liquid dish or the capsule with the capsule membrane facing down, except in the capsule plate. To prevent rupture, avoid touching the capsule membrane at all stages.

Chapter 2: Technical Data

2.1 QX-302 Capsules

Storage	The capsules should be stored in a dry, dark environment at room temperature.
Shelf life	12 months from specified production date (printed on the box).
Application	The capsules are intended for single use and are not reusable. They are intended for research pur- poses only. The QX-302 capsule is suitable for imaging thick, non-adherent samples such as tissue biopsies, plants, and material specimens, in a wet environ- ment. The capsule is suitable for various sample sizes, with a maximum diameter of 3 mm. Sample thickness can range between 300-1000 microns.
Dimensions	Specimen dish - diameter 3 mm, Sealed capsule - see Figure 1 on page 10. In cases where the QX-302 capsule does not fit your SEM stage, inquire for available adaptors.
Sterility	Gamma-sterilized. When sterility is required, opening of the packaging should be done in a sterile environment (laminar flow). Opened packages should be stored under sterile conditions.
Operation Temperature	4° to 40° C

Material
CompatibilityFor compatibility of reagents with the
QX-302 capsule materials, see Table 1
on page 11. For other materials or spe-
cific concentrations, please contact
tech@quantomix.com. For materials not
compatible with the QX-302 capsule,
inquire about other capsule types.
Reagents containing DMSO should not
be used during SEM imaging (DMSO
can be present in the sample prepara-
tion process).

Figure 1. Sealed Capsule - Dimensions



Dimensions are given in mm.

Material	Compatible	Not Compatible
Acetone		x
DMSO		x
Ethanol	х	
Ethyl Acetate		x
Formalin	x	
2% Glutaraldehyde	x	
Isopropanol	x	
Methanol	x	
4% Paraformaldehyde	x	
1% Tannic Acid	x	
0.5% Triton [®] X-100	x	
0.5% Tween [®] 20	x	
Toluene		x
Xylene		x

2.2 Capsule Plates (MP-12)

Storage	Should be stored in a dry, dark environment at room temperature.	
Shelf Life	Three years from specified production date.	
Application	The plates should be used for all stages in which cap- sules are being handled.	
Dimensions	85 x 128 x 33 mm	
Sterility	ETO sterilized	
Operation Temperature	4° to 40°C	

Figure 2. The Capsule Plate



Capsule plate, as seen from above



2.3 Imaging Buffer (IB-74)

Storage	The lyophilized product is stored at room temperature. After reconstitution, store in the dark at 4°C. The reconstituted product is stable for 1 month at 4°C.
Shelf life	12 months. See expiry date printed on the bottle.
Application	Imaging buffer should be added to the specimen dish prior to SEM imaging in order to protect specimen from beam damage.

2.4 Spacers

Paper type	Whatman [®] 3 mm CHR	
Diameter	3 mm	
Thickness	0.30 mm	
Packaging	Spacers are supplied inside the Imaging Buffer con- tainer.	
Application	Spacers should be used to enhance the attachment of the specimen to the QX-302 capsule's membrane.	

Chapter 3: Introduction

Imaging fully hydrated samples has been a long-needed capability in electron microscopy. The WETSEMTM technology developed by QuantomiX combines the resolution that characterizes Scanning Electron Microscopes (SEM) with the easy sample preparation that characterizes light microscopy.

The technology is based on a thin, electron-transparent membrane that completely isolates the sample from the vacuum in the microscope chamber.

Fully hydrated specimens are placed in the capsule. The capsule is sealed, and imaging is done through the electron-transparent membrane.

3.1 Manual Scope and Contents

This manual provides a detailed description of the components required for using the QX-302 capsules. It also provides guidelines for inserting samples into the capsules and imaging.

References for specific protocols for preparing the samples are provided in Appendix A of this manual.

This User Manual consists of the following chapters:

Chapter	Heading	Provides
1	Safety	Cautions for safe use of the QX-302 cap- sules
2	Technical Data	Technical data about the QX-302 capsules and accessories
3	Introduction	User Manual descrip- tion, References, and Components descrip- tions
4	Using the QX-302 Capsules	Instructions for prepar- ing samples for use with the QX-302 cap- sule, inserting sam- ples into the capsule, and storing samples in the capsules
5	Imaging	Instructions for imag- ing with the QX-302 capsules
6	Appendices	Protocols for Specific Applications, Glos- sary, Troubleshooting, Ordering Information, and Legal Notices

3.2 References

3.2.1 Sites

http://www.quantomix.com

3.2.2 Technical Support

For technical support please contact your local distributor or tech@quantomix.com.

3.3 Technology and Components

The WETSEM[™] technology is a proprietary technology developed by QuantomiX Ltd., which enables direct imaging of fully hydrated samples in scanning electron microscopes. The patented technology is based on a thin, electron-transparent membrane, which seals the sample from the vacuum in the microscope chamber. No coating or embedding of the sample are required, enabling electron microscopy imaging with easy sample preparation comparable with light microscopy.

The QX-302 capsules can be used for imaging various materials and biological samples. The samples can be visualized either directly or by following the appropriate contrast enhancement staining or labeling procedures, for which some typical protocols are provided on the QuantomiX website at http://www.quantomix.com.

The QuantomiX components required for using the technology are listed below.

- The QX-302 capsule
- Capsule plate
- Spacers
- Imaging Buffer

3.3.1 The QX-302 Capsule

The QX-302 capsule shown in Figure 3 on page 18 is a sterile single use specimen enclosure consisting of a specimen dish and a sealing stub.

The base of the capsule is the specimen dish, which is designed as a small dish in which specimens can be placed. The top of the capsule (the sealing

stub) is designed to seal the specimen chamber and to ensure the attachment of the specimen to the capsule's membrane.

- 1. Prior to sealing the capsule, ensure the O-ring is properly positioned in the sealing stub (as seen in Figure 3 on page 18).
- 2. The specimen dish should be positioned in the capsule plate (as seen in Figure 4 on page 19).
- 3. Place the sealing stub on top of the specimen dish.
- 4. Hold the sealing stub from the middle and turn clockwise until full closure is obtained. Check that the wings of the sealing stub are aligned with the wings of the specimen dish.



Figure 3. QX-302 Capsule Parts

Figure 4. QX-302 Capsule, Positioned in the Capsule Plate



Note

Prior to closing the capsule, ensure that the O-ring is positioned appropriately in the sealing stub.

Caution

a. Correct sealing of the capsule is essential for its proper functioning.

b. Capsule sealing is achieved when the wings of the specimen dish and the sealing stub are aligned.

c. Do not use sharp objects, such as sharp-ended forceps, to hold the rubber seal, in order to prevent damage to the sealing.

3.3.2 Capsule Plate (MP-12)

The QX capsule plate is a sterile disposable plastic holder for the capsules. It is designed to enable parallel handling of a number of individual QX-302 capsules. It serves for holding the capsules during specimen preparation, and for storage.

The QX capsule plates are supplied sterile and intended for single use. They are not compatible with autoclave sterilization.

Note

Placing the QX-302 capsule in the QX capsule plate protects the membrane from rupture.

3.3.3 Imaging Buffer (IB-74)

QX-302 Imaging Buffer is a solution optimized for imaging samples in an SEM with QX-302 capsules and is formulated to minimize damage to the samples by the electron beam. Imaging of samples should be done in QX Imaging Buffer whenever possible, especially for biological samples.

QX Imaging Buffer should be used unless a specific experimental requirement dictates a different medium.

QX-302 Imaging Buffer is applied on samples after sample preparation steps and prior to imaging, as described in Section 4.3 on page 27.

3.3.4 Spacers

The spacer is a piece of absorbent paper that should be placed in the specimen dish above the sample. The spacer is used to protect the sample from damage that could be incurred by the plunger of the sealing stub. In addition, in case of a thin sample, multiple spacers are placed above the sample, enhancing the attachment of the sample to the membrane. Sample attachment to the membrane is critical for SEM imaging.

The spacers are included in the Imaging Buffer vial (Cat. No. IB-74).

Note

For additional product details, please refer to instructions accompanying the product.

3.3.5 Calibration Capsule (RT-58)

The Calibration Capsule (Cat. No. RT-58) is a QX-302 capsule designed for finding the optimal imaging conditions for wet samples with WETSEM[™] technology.

It is strongly recommended that new users initially use the Calibration Capsule to find the optimal working conditions in the SEM.

Imaging of samples with a QX-302 capsule in an SEM differs from standard SEM imaging in some aspects. The factors that affect imaging vary among applications and differ from one SEM model to another. The Calibration Capsule contains nanoparticles (40 and 500 nm in size) stably attached to the capsule membrane. The particles are easily imaged in an SEM and provide a convenient means to calibrate the parameters for optimal wet imaging conditions.



Note

Always use the Calibration Capsule with QX Imaging Buffer.

For details of use and calibration, please refer to the instruction page accompanying the product.

Chapter 4: Using the QX-302 Capsules

The following procedures are general and appropriate for many different QX-302 applications. For specific applications protocols, please see the QuantomiX website, at http://www.quantomix.com.

4.1 Getting Started

The QX-302 capsules and the QX multi-capsule plates (MP-12) are supplied sterile in a sealed container. If maintaining sterility is desired, the opening and further handling of capsules should be performed in a sterile environment.



Note

It is recommended to open the capsules only after specimen preparation (section 4.2 on page 24) has been com-



Caution

To prevent rupture of the membrane, avoid touching the membrane at all stages. Keep the specimen dish properly positioned in the capsule plate at all times.

- Unwrap the QX-302 capsule container by peeling back the paper cover.
- Position the individual capsules in the QX capsule plate.
- Open the positioned capsules by turning the middle part of the sealing stub counter-clockwise (see Figure 5 on page 28). Do not open the top of the sealing stub.

- Put aside the sealing stub until all preparations are completed and the sample is ready for imaging. It is recommended to use the original QX-302 capsule package for storing the sealing stubs.
- Cover the QX-302 capsule plate with the lid to keep the specimen dish undamaged.

To prepare a specimen for SEM imaging, follow the steps described in the following section.

4.2 Specimen Preparation

Any fresh or freshly-fixed soft specimens can be used and imaged in the QX-302 capsule. Experience shows that older biological specimens, even when fixed, tend to disintegrate and loose their internal structure. For best results, these samples should be avoided.

Note

Hard or sharp specimens can damage the capsule membrane and tear it. It is not recommended to use the QX-302 capsule for imaging hard or sharp specimens.

4.2.1 Fixation

Most biological specimens require fixation prior to further treatments. The purposes of the fixation are:

- To preserve the biological structures as close to the living state as possible.
- To protect the specimen from morphological alteration and damage during subsequent treatments.

The most common fixation reagent is formalin. However, other fixatives can be applied to the specimens. Since no fixative preserves all biological struc-

tures, the choice of a fixation reagent and conditions is dependent on the specimen itself and the features of interest.

For specific applications please consult your local distributor or tech@quantomix.com.

4.2.2 Slicing and Cutting

Using the QX-capsules, the electron beam can penetrate a specimen to a depth of up to 3 microns. Thickness of specimens beyond 3 microns does not have an effect on the quality of the imaging.

Specimens prepared for use with the QX-302 capsule should be sliced to a thickness of 300-1000 microns. Samples can be sliced manually using a scalpel knife or with slicing equipment such as Vibratome[™] (www.vibratome.com).

Manually slicing is a simple and fast process. Its disadvantages are the lack of accuracy of final sample thickness and the roughness of the sliced surface.

New, sharp blades must be used. The quality of the final slices will depend on the skill and experience of the operator.

The use of slicing equipment will provide slices of a defined, uniform thickness. The process will usually require additional steps in order to stabilize the specimen in agarose prior to slicing. For best results, please consult the user manual of the instrument to be used.

Once a specimen is sliced to the required thickness, use a knife to reduce its size. The final section should be small enough to fit into the QX-capsule specimen dish, which is 3mm in diameter.

Note

The final specimen should be less than 1000 microns thick and smaller than 3 mm in diameter.

4.2.3 Contrast Enhancement

The imaging contrast in QX-302 capsules is created from variations in the average atomic number of the sample constituents. Thus, heavy metal stains, such as Uranium and Osmium compounds, are best suited for improving the general contrast of biological samples. Table 3 on page 26 describes some common staining reagents and the features highlighted by them.

Reagent	Features Enhanced
Uranyl Acetate	General contrast agent. Binds to nucleic acids, proteins and membranous structures
Osmium Tetroxide	Lipids, membranous structures, vesicles
PTA (Phosphotungstic acid)	Positively charged structures such as basic pro- teins associated with nuclear DNA and nucleoli.

TABLE 3: Features Enhanced by Different Staining Agents

General staining protocols are provided on the QuantomiX website at http://www.quantomix.com. These should be used as guidelines and be modified to suit the particular specimen.

4.2.4 Immunogold Labeling

Immunogold labeling techniques are common in electron microscopy and are suitable for imaging with the QX-302 capsule. These techniques allow the visualization of a single gold colloid attached to a single molecule, thus enabling the localization and quantification of specific receptors.

General guidelines for immunogold labeling are provided on the QuantomiX website at http://www.quantomix.com. Since there is no standard labeling protocol which is suitable for all labeling reactions, optimal labeling conditions should be established on the basis of prior experience with the particular antibody and antigen or on preliminary experiments.

4.3 Inserting Specimens into the QX-302 Capsule

4.3.1 Preparing the capsule

Place a new QX-302 capsule in the capsule plate (see Figure 4 on page 19).

Turn the middle part of the capsule (the sealing stub) counter-clockwise (see Figure 5 on page 28) and place it inside the capsule plate lid. The specimen dish should be resting in the capsule plate during the entire procedure.

1	\Box	

Note

The specimen dish contains the membrane that enables viewing of the samples with an SEM. The membrane is very delicate and must not be touched at any time. Special care should be taken when using sharp instruments.

The sealing stub contains a plunger that, once the capsule is closed, pushes the sample towards the membrane and mechanically attaches it to the membrane. This action ensures that the sample is within the electron beam penetration range and enables SEM imaging.

Figure 5. Opening the QX-302 Capsule



4.3.2 Placing the sample

Carefully place your sample into the specimen dish. The sample should have a diameter of 3 mm or less in order to fit into the dish. Use a spacer as an aid in this procedure, as described in Section 4.3.3 on page 29.



4.3.3 Using spacers

Reconstitute the QX-302 Imaging Buffer with 1 ml of double-distilled water. The spacers will absorb the Imaging Buffer. Remove a spacer from the vial and use it as described below:

- 1. Place a spacer on the working surface and place the sample gently on the spacer. Holding the spacer with tweezers, turn it over and place it in the specimen dish. (See Figure 6. on page 29.) Remember that the sample itself must be touching the membrane, and the spacer is to be placed above the sample, further from the membrane.
- 2. If your sample is thinner than 300 microns, place an additional spacer, in order to make sure that the sample is held in close contact with the capsule membrane.



Figure 6. Placing a Sample in the Specimen Dish

4.3.4 Adding Imaging Buffer

Carefully add 2 to 5 microliters of Imaging Buffer into the specimen dish.

4.3.5 Sealing the capsule

Now that the sample is placed in the specimen dish, the capsule may be sealed. While keeping the specimen dish in the capsule plate, gently place the sealing stub on it. Turn the sealing stub clockwise until the "wings" are aligned. At this point, the capsule is tightly closed and ready for imaging in the SEM (see Chapter 5 on page 31). To ensure it remains intact, leave the capsule in the capsule plate until it is placed in the SEM. For further protection, the capsule plate itself should be covered.

4.4 Storing Specimens in the QX-302 Capsule

Samples can be stored in sealed capsules for a limited period. Sample quality will deteriorate over time. The rate of deterioration is a function of the nature of the sample itself and the quality of the storage conditions. Over a period of approximately one week samples could gradually dry out and therefore lose their original features. It is recommended to store samples at 4°C.



Note

The sealed capsules are conveniently stored in the capsule plate.

Chapter 5: Imaging

The factors that affect imaging vary with different applications and different SEMs. This chapter provides guidelines for achieving the best imaging results for wet samples with WETSEM[™] technology.

The imaging conditions are best optimized using the Calibration Capsule. The Calibration Capsule contains nanoparticles, which are easily visualized in the SEM and provide a convenient means to calibrate the parameters for optimal imaging conditions. The Calibration Capsule is provided with an image to evaluate the optimization process. Please read the instructions accompanying the product prior to use.

5.1 Optimal Imaging Conditions

Follow the guidelines below to find the optimal imaging conditions.



Note

Recommendations for SEM parameters are summarized in Table 4 on page 34. Optimize the conditions first with the Calibration Capsule, and then move on to imaging your sample.



Caution

The QX-302 capsule is taller than the average specimen. Bring the stage to a low position before placing the capsule in the SEM.

- 1. Place the closed QX-302 capsule on the microscope stage as a conventional 'stub' with the capsule membrane facing up. In case the QX-302 capsule does not fit your SEM stage, inquire for available adaptors.
- 2. The QX-302 capsules are suitable for use with high vacuum or low vacuum modes.
- 3. Ensure sufficient working distance. Note that the QX-302 capsules may be taller than conventional SEM stubs. Adjust the height of the stage as necessary.
- 4. Perform the imaging with a Back Scattered Electron Detector (BSED). The capsule also enables X-ray microanalysis using suitable detectors. Secondary Electron (SE) imaging can also be performed, but with a lower signal. For systems without a BSED, consult QuantomiX representatives regarding the use of secondary electron detectors in conjunction with the QX-302 capsule.
- 5. For best imaging, adjust the working distance (WD) to maximize the sample BSE signal. See Table 4 on page 34 for recommended WD range.
- 6. To obtain best imaging conditions, start with an acceleration voltage of 30 kV and a mid-range spot size.
- 7. Increase the contrast until the desired signal from the sample is obtained (usually maximum contrast). It is recommended to start imaging with a low scan speed (a few seconds per frame). Note that since the signal from the sample is generally weaker and of lower contrast than the signal from the support grid, the optimization of contrast and brightness should be carried out with respect to the sample and not to the grid.
- Focus on the sample. If you have difficulties in focusing on your sample at this stage, focus first on the supporting grid of the capsule using SE (secondary electron) detector, then switch back to BSE detector.
- 9. The range of recommended acceleration voltage is 10 kV to 30 kV. Different acceleration voltages correspond to different penetration depths.

- 10. Probe current/spot size is determined empirically by the optimal signal obtained. A higher probe current generates a larger signal. A higher probe current may also damage the sample. The optimal probe current/ spot size is determined by optimizing the signal and minimizing damage to the sample. For maximal allowed probe current see Table 4 on page 34.
- 11. Scan speed should be adjusted according to the signal from the sample. For low contrast samples, it is recommended to work with lower scan speeds. For samples sensitive to higher beam damage, it is recommended to integrate several frames instead of scanning a single frame at a lower scan speed.

5.2 Recommendations for SEM Imaging with QX-302 Capsules

TABLE 4: Recommendations for SEM Imaging with QX-302Capsules

Parameter	Recommended range	Comments					
Acceleration voltage	15-30 kV	Not lower than 10 kV					
Probe current (based on source type)							
Tungsten filament	0.4-1.0 nA	Not higher than 1.0 nA					
FEG	0.1-0.5 nA	Not higher than 0.5 nA					
Working distance (based on detector type)							
Semiconductor (BSE)	6-10 mm	Acceptable: 5-15 mm					
Robinson (BSE)	10-20 mm	Better efficiency at higher kV					
Scintillator (BSE)	6-10 mm	Acceptable: 6-10 mm					
Everhart- Thornley (SE)	8-12 mm	Acceptable: 6-15 mm					
In-lens / Through the lens (all detectors)	2-4 mm	Manufacturer dependent					

Chapter 6: Appendices

6.1 Appendix A: Protocols for Specific Applications

For the latest applications protocols, please see the list of published protocols at www.quantomix.com, on the Technology tab.

Each of the protocols provided on our web-site includes a list of the required reagents, a step-by-step procedure and Notes highlighting issues important for the successful completion of the protocol.

It should be noted that the protocols are based on work conducted at QuantomiX laboratories. They are given as a starting point which will facilitate the user's first steps in acquiring the desired imaging results. It is the user's responsibility to determine the suitability of any protocol published by QuantomiX to their applications. Users may find it necessary to modify protocols in order to obtain the information required for their study.

Note

All fixing, staining and labeling protocols of samples for QX-302 capsules can be done in small test-tubes. During incubation steps, samples should be gently agitated in the appropriate reagent to ensure a homogeneous reaction.

6.2 Appendix B: Glossary

BSED	Back-scattered electrons detector
BSE	Back-scattered electrons
Calibration Capsule	QX-capsule with reference sample used for optimiza- tion of imaging conditions
ЕТО	Ethylene Oxide Sterilization
MP-12	Capsule plate; a sterile, transparent holder for parallel handling of up to 12 individual QX-302 capsules, serving to hold and store the capsules
QX-302	Capsule used for SEM-imaging of various wet, thick non-adherent samples.
QX Imaging Buffer	Buffer optimized for imaging samples in an SEM with QX-302 capsules
SE	Secondary electrons
SED	Secondary electron detector
SEM	Scanning electron microscope
Sealing Stub	Part of the QX-302 capsule used for sealing the cap- sule and for holding the capsule in the SEM
Spacer	A round piece of absorbent paper designed to enhance specimen attachment to the QX-302 capsule membrane.
Specimen Dish	QX-302 capsule base designed as a dish for placing specimens

6.3 Appendix C: Troubleshooting

TABLE 5: Troubleshooting

Phase	Problem	Possible Cause	Solution
Immediately after sample insertion	There is a drop- let on the multi- capsule plate, underneath the membrane.	Membrane was rup- tured during sample insertion.	A new sample must be prepared. A cap- sule with a torn membrane should not be placed in the SEM.
Imaging	No signal is observed.	The sample is not in con- tact with the membrane.	Another spacer should be added above the sample.
	The image is not clear.	There is insufficient contrast between the constituents of the sam- ple.	The sample may require contrast enhancement, such as heavy metal staining. See Sec- tion 6.1 on page 35.
	High signal arti- facts appear during imaging.	No Imaging Buffer was added.	Add 2 to 5 microli- ters of Imaging Buffer prior to imag- ing.
	High signal imaging (charg- ing effect)	Sample is old and has dried out.	Prepare a new sample.
		Sample has not been kept at 4°C and has dried out.	Prepare a new sample.

6.4 Appendix D: Ordering Information

Please see www.quantomix.com for your local distributor or QuantomiX Sales Representative or contact sales@quantomix.com.

6.5 Appendix E: Legal Notices

6.5.1 Product Warranty, Liability and License for Use

- QuantomiX guarantees the performance of all Products in the manner described in our product literature. The Purchaser must determine the suitability of the product for its particular use or application. Should any product fail to perform satisfactorily, within a period of twelve months following the date of delivery, due to any reason other than misuse or unsuitable application, QuantomiX will replace it free of charge or refund the purchase price. Please contact tech@QuantomiX.com or your local distributor or sales representative.
- QuantomiX QX capsules have been validated for single-use in Scanning Electron Microscopes of the major manufacturers. Reuse of capsules is prohibited, is deemed misuse of the Products and releases QuantomiX from all warranty obligations.
- 3. QuantomiX products are covered by patents owned by QuantomiX Ltd. as well as patents owned by Yeda Research and Development Co. Ltd and licensed to QuantomiX. Upon delivery of the Products, QuantomiX shall be deemed to have granted the Purchaser a non-transferable, nonexclusive license to use the Products for the sole purpose of performing research and development applications. The Purchaser will not have the right to market, sub-license, or otherwise grant any right in, or to, the Products or make any commercial use or any disposition whatsoever in the Products.
- 4. The limited warranties set forth in these terms and conditions are given to the Purchaser only, are not enforceable by any other entity or person, and are the sole and exclusive warranties given by QuantomiX with respect to the Products. QuantomiX expressly disclaims any and all implied warranties, including but not limited to, implied warranties of merchantability and fitness for a particular purpose, title and noninfringement.
- 5. In no event shall QuantomiX bear any liability, obligation or responsibility for any indirect, incidental or consequential damages in connection with the Products, regardless of the form of action, including but not limited

to, loss of revenue or anticipated profits arising in any way in connection with the use of the Products. In no event shall QuantomiX be liable for any amount greater than the amount paid to it in respect to the specific product giving rise to the liability.