Electron
Microscopy
Sciences

INSTRUCTIONAL MANUAL
CAT. FV-102

FlowVIEW Starter Kit - Standard

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Introduction & Features

Creating a liquid environment in SEM

Aquarius is the pioneer of using e-beam to test original fluids and liquid samples. Aquarius can test and monitor coating materials in their original form, especially the liquid state.

The functions of the chip and liquid sample holder are simultaneous versatile and high-resolution inspection (disposable version for slurry testing, inline version for process monitoring, quick assembly version for electrode testing).

The system is capable of displaying the images of sample in its liquid state under original conditions using the anti-vacuum sample holder with nano-membrane and microfluid channels. It can provide analysis results such as particle sizedistribution, dispersity, uniformity, concentration, shape and composition required for R&D and production.

The liquid sample holder in the MFC system is compatible with different preparation tools (such as powder disperser for powder size distribution analysis, centrifuge for bio-sample morphology observation and membrane filter for liquid defect identification) for static inspection in various applications. The sample in the holder can be transferred to different inspection platforms to obtain the correlative information and make versatile and in-depth analysis of the sample possible. Samples can be packaged in 30 seconds without any dilution, drying, or frozen section procedures. Since the nano-membrane is only 20-50 nanometers and provides you a 7-nm resolution, the e-beam can fully penetrate.

Scientific Principles

- **Ultra-Thin Sample Adaptive Chips:** Produced using the semi-conductor manufacturing process together with surface treatment, the membrane can be made hydrophobic, hydrophilic or bifunctional linkers for different applications,allowing samples under test to automatically adhere to the observation window for best image resolution. To ensure image quality, the membrane thickness is designed to be less than 30 nm, yet it is made robust by adjusting the window size and shape to withstand the pressure difference between the vacuum and atmosphere.

- **Liquid Sample Holder:** Combing with microfluidics and high-precision pogo pin with leak-free sealing design, the liquid sample holder is capable of controlling and monitoring the micro-environment. The microfluidics designs were optimized by Computational Fluid Dynamics (CFD) simulations to achieve the best transportation model for the samples. The liquid sample holder has special mechanical design to boost the operation efficiency and can finish the sample loading within a minute. The electrical charging/discharging can also be transmitted to the control system and inside the microfluidics via pogo pin design. Additionally, the heat exchange fluid can be circulated in one of the flow channels, thereby accomplishing the functions of temperature control and fluid properties monitoring, allowing nano-scale in-situ testing of the samples under their original activity.

FlowVIEW Image Analysis Software

This software is included with the FlowVIEW Starter Kit:

- **Fast** - Finish the liquid image analysis within 1 minute.
- **Simple** - The software interface has user-friendly design and it's easy to get started.
- **Multi-Functional** - Functions include Particle Size Distribution, Dispersion, Concentration, Composition and Shape Analysis.
- **Automation** - Full automation analysis with one button.

The FlowVIEW Aquarius "Liquid" SEM Starter Kit contains:

- 24 Microscopic Fluid Chips
- 24 Micro-Channel Substrate
- 24 Tips
- 1 Adapter
- 1 O-Ring
- 1 Gasket
- Tweezer
- Pipette
- Acrylic Case with Holder
- 1 Lite Software

Introduction & Features

Features:

- **Simple** - The liquid sample holder has special mechanical design to boost the operation efficiency and can finish the sample loading within 0.5 minute.
- **High Resolution** - Objects smaller than 10 nm can be easily observed.
- **High Compatibility** - Highly compatible with various models (ex. FEI, JEOL, Hitachi, ZEISS, Phenom, TSCAN, etc.). It can be a shuttle to an Optical Microscope/Fluorescence Microscope for in-situ observation
- **Customized** - It can be placed with the silicon wafer & biochip substrate for in-situ observation

Performance:

Supplementary Packs: After purchasing a starter kit, subsequent materials can be ordered in supplementary packs which include both micro-channel substrates and microscopic fluid chips.

Supplementary Packs include:

- 12 Microscopic Fluid Chips
- 12 Micro-Channel Substrate

Safety

Please observe the following safety guidlines while using the Aquarius Starter Kit to avoid injury or damage:

Do not use HF base acid or hot H_3PO_4.

It is necessary to wear gloves during while using the kit.

Before installing the Microscopic Fluid Chip onto SEM, confirm its integrity. Customers should go to the OM to observe whether the Microscopic Fluid Chip's surface is complete.

Confirm the Micro-Channel Substrate is in the center when placing it. The liquid sample should be loaded into the Micro-Channel Substrate (0.5-1μL maximum). To avoid window damage during sealing, **do not** use too much sample liquid.

Ensure the Micro-Channel Substrate and the fluid Microscopic Fluid Chip are completely sealed.

The membrane (which cannot be disassembled) can be used one time.

Maintain the liquid sample under 15°C-30°C.

Storage: store the kit in a moisture-proof box. The room temperature should be kept below 40°C. The humidity should be kept under 50% RH. The kit should be kept in a dark and dust-free environment.

If liquid leaks due to the membrane breaking (or any other reasons), failure or damage to the scanning electron microscope (SEM) may result. Please operate the product according to the user manual to prevent this. Ensure that the microscopic fluid chip does not get dropped or damaged. Avoid focusing on one specific point continuously and avoid fine scanning repeatedly.

Technical Data

1. **Storage:**
   - Keep the kit in a moisture-proof box.
   - Room temperature should be kept under 40°C.
   - Humidity should be kept under 50% RH.
   - Keep the kit in a dark and dust-free environment.

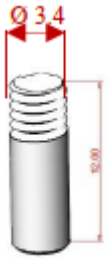
2. **O-Ring Replacement:** Check the o-ring for sealing at least once every 3 months. If any damage is found, the O-ring needs to be replaced.

3. **Disposal:** Make sure to follow the local government rules of disposal for each part of the kit as well as any liquid used for observation. The manufacturers does not handle disposal.

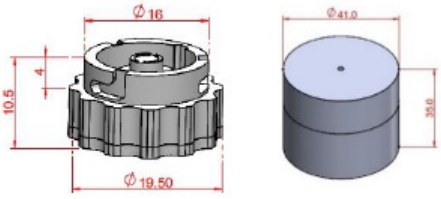
4. **Operation temperature:** Use the kit at an ambient room temperature between 15°C and 30°C.

5. **Material Compatibility:**

   a. **Adapter**
      i. **Function:** Connects stage to SEM.
      ii. **Operation:** Install the adapter under the holder and tightly lock it according to the thread. For fixation, it can be placed in the SEM.
      iii. **Specification:**
         - **Pitch:** M4x0.7
         - **Length:** 12 mm
      iv. **Material:** Stainless.



   b. **Acrylic case with holder**
      i. **Function:**
         - **Holder:** Used to place the Micro-Channel Substrate and sample.
         - **Acrylic case:** Used to combine the holder with the upper cover.
      ii. **Operation:**
         - Place the Micro-Channel Substrate on the stage.
         - Place the sample on top of the substrate.
         - Align the upper cover with the stage joint hole.
         - Use an acrylic case to press down.
         - Rotate to lock it.
      iii. **Specification:**
         - **Diameter:**
            - **Holder:** 16 mm and 19.5 mm
            - **Acrylic Case:** 41 mm
         - **Height:**
            - **Holder:** 10.5 mm
            - **Acrylic Case:** 35 mm
      iv. **Material:**
         - **Acrylic case:** Acrylic
         - **Holder:** Stainless



Technical Data

c. Microscopic Fluid Chip
Standard
High Mag.
Large Window

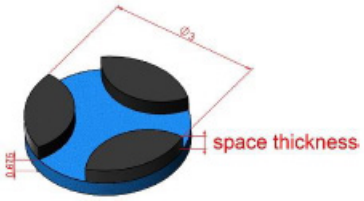
i. **Function:** The Electron transparent window is used to inspect liquids.
ii. **Operation:** Assemble this component together with the stainless holder and the Micro-Channel Substrate in a sandwich structure to inspect liquids.
iii. **Specification:**
- **Diameter:** 19.5 mm
- **Height:** 5 mm
- **Membrane specification:** depends on the product.

iv. **Material:**
- **Mounting:** Conductive plastic
- **Pin:** Stainless
- **Membrane:** Si_3N_4

d. Micro-Channel Substrate:
(a) Space thickness: 2μm
(b) Space thickness: 0.5μm

i. **Function:** Accommodates the sample solution.
ii. **Operation:** Place the sample solution in the internal space of the Micro-Channel Substrate.
iii. **Specification:**
- **Diameter:** 3 mm
- **Height:** 675 μm (depends on whether space thickness 2μm or 0.5 μm is used)

iv. **Material:** Si and SiO_2



e. O-ring

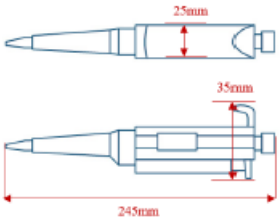
i. **Function:** Sealing.
ii. **Operation:** The o-ring is installed into the stainless holder.
iii. **Specification:** 9 x 1.5 mm
iv. **Material:** Viton

f. Pipette

i. **Function:** Fills with a fixed amount of sample liquid sample from the stage.
ii. **Operation:** Install a tip and fill it with 1μL of the sample liquid from the Micro-Channel Substrate.
- The sampling button has two stages. The first stage is lightly pressed to the sample. The second stage is pressed to the bottom to stake out.
- Select the amount of solution required with the upper rotary knob (0.5~10 μL).
- Tip return button quickly returns the tip.

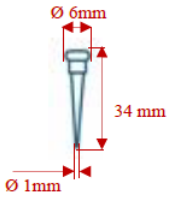
iii. **Specification:**
- **Volume:** 0.5-10μL
- **Size:** 245mm x 25mm x 35mm

iv. **Material:** Fortron



Technical Data

g. Tip



i. **Function:** Consumable pipette tip.
ii. **Operation:** Assemble the tip with the pipette.
iii. **Specification:**
- **Volume:** 10 µL
- **Length:** 34 mm

iv. **Material:** PP

h. Tweezer

i. **Function:** Pick up items with the tweezer.
ii. **Operation:** Use the tweezers to pick up the Micro-Channel Substrate and place it on the holder
iii. **Material:**
- **Body:** ALSI302
- **Head:** PPS

6. Materials Used for the Components:

Components	Materials
Adapter	ɣ-Fe
Acrylic Case	polymethylmethacrylate (PMMA)
Holder	C, Si, Mn, P, S, Ni, Cr, Mo
Microscopic Fluid Chip	Nylon and thermoplastic polyurethane, Si_3N_4
Micro-Channel Substrate	Si, SiO_2
O-ring	Viton
Pipette	Fortron
Tip	PP
Tweezer	C, Si, Mn, P, S, Cr, Ni

7. Prohibited Material of Components:

a. Key:
- **"O":** The content of the restricted substance does not exceed the ppm of reference value.
- **"X":** The prohibited substances are not included with the component.
- **"N/A":** The component known the material but not including prohibited substances.

Prohibited Material	Adapter	Acrylic Case	Holder	Microscopic Fluid Chip	Micro-Channel Substrate	O-Ring	Pippette	Tip	Tweezer
Cadmium and Cadmium Compounds	O <100ppm	X	O <100ppm	O <100ppm	N/A	N/A	N/A	N/A	N/A
Hexavalent Chromium Compounds	N/A	X	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Lead and Lead Compounds	N/A	X	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Lead and Lead Compounds (wire coating)	N/A	X	N/A	N/A	N/A	N/A	N/A	N/A	N/A

Technical Data

7. Prohibited Material of Components:

Prohibited Material	Adapter	Acrylic Case	Holder	Microscopic Fluid Chip	Micro-Channel Substrate	O-Ring	Pippette	Tip	Tweezer
Mercury and its compounds	N/A	X	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Lead, Cadmium, Mercury, Hexavalent Chromium (package and wrapping only)	O <100ppm	X	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Polybrominated Biphenyls (PBBs)	N/A	X	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Polybrominated Diphenyl ethers (PBDEs)	N/A	X	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
DecaBDE	N/A	X	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Bis(2-ethylhexyl) (DEHP)	N/A	X	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Butyl benzyl phthalate (BBP)	N/A	N/A	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Dibutil phthalate (DBP)	N/A	N/A	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Diisobutyl phthalate (DIBP)	N/A	N/A	N/A	O <100ppm	N/A	N/A	N/A	N/A	N/A
Perfluorooctanoic acid (PFOA) and its salts and PFOA-related compounds	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Polychlorinated Biphenyls (PCBs)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Polychlorinated Terphenyls (PCTs)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Short Chain Chlorinated Paraffins	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Tri-substituted Organostannic Compounds	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Asbestos	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Ozone Depleting Substances	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Radioactive Substances	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
PFOS and its Analogous Compounds	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A

Technical Data

7. Prohibited Material of Components:

Prohibited Material	Adapter	Acrylic Case	Holder	Microscopic Fluid Chip	Micro-Channel Substrate	O-Ring	Pippette	Tip	Tweezer
2-(2H-1,2,3-benzotriazol-2-yl)-4,6-di-tert-butylphenol	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexachlorobenzene	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexabromocyclodo-decane (HBCD)	N/A	X	N/A	N/A	N/A	N/A	N/A	N/A	N/A
2,4,6-Tris(tert-butyl) phenol (2,4,6-TTBP)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Phenol,Isopropylated Phosphate(3:1) (PIP 3:1)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Pentachlorothio phenol (PCTP)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hexachlorobutadiene (HCBD)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A

8. Solvent Compatibility:

Chemical Name	Microscopic Fluid Chip	SiN Membrane	Micro-Channel Substrate
Hydroflouric Acid	X	X	X
Hot Phosphoric Acid	X	X	X
Acetic<20%	O	O	O
Acetone	O	O	O
Ammonium Chloride (24%)	O	O	O
Ammonium Hydroxide (28%)	O	O	O
Ammonium Sulfate	O	O	O
Aqua Regia	X	O	O
Benzene	O	O	O
Bleach Solution<20% (Sodium hypochlorite)	O	O	O
Bromine Calcium Chloride (38%)	O	O	O
Chloroform	O	O	O
Citric Acid	O	O	O
Copper Sulfate	X	O	O
Cyclohexane	O	O	O
Diethylether	O	O	O
Diethylamine	O	O	O
Dimethyl Sufloxide 10%	O	O	O
Dimethyl Formamide	O	O	O
Ethanol	O	O	O
Ethylene Diamine	O	O	O
Ethyl Acetate	O	O	O

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8. Solvent Compatibility:

Chemical Name	Microscopic Fluid Chip	SiN Membrane	Micro-Channel Substrate
Ethylene Glycol (Undiluted)	O	O	O
Glycerin (Undiluted)	O	O	O
Gold Chloride	X	O	O
HEPES Buffer	O	O	O
Hexane	O	O	O
Heptane	O	O	O
Hydrogen Peroxide (30%)	O	O	O
Hydrochloric Acid (2%)	X	O	O
Isobutanol	O	O	O
Isopropanol	O	O	O
Ketones	O	O	O
Magenesium Sulfate	O	O	O
Methanol	O	O	O
Methylene Chloride	O	O	O
Mineral Oil	O	O	O
Nitric Acid (10%)	O	O	O
Oleic Acid (40%)	O	O	O
Petroleum Oil Undiluted	O	O	O
Perchloric Acid	X	O	O
Phosphate Buffered Saline	O	O	O
Phosphoric Acid	O	O	O
Potassium Hydroxide	O	O	O
Pyridine	O	O	O
Sodium Bicarbonate (50%)	O	O	O
Sodium Carbonate (20%)	O	O	O
Sodium Chloride (30%)	O	O	O
Sodium Hydroxide (10%)	O	O	O
Sodium Nitrate	O	O	O
Sodium Phosphate (5%)	O	O	O
Sulfuric Acid (<5%)	X	O	O
Tannic Acid (10%)	O	O	O
Tartaric Acid	O	O	O
Tetramethylammonium hydroxide	O	O	O
Toluene	O	O	O
Trichloroacetic Acid	X	O	O
Vegetable Oil	O	O	O
Water	O	O	O
Urea	O	O	O
Xyelene	O	O	O
Zinc Chloride	X	O	O

Technical Data

9. Sample and Chip Window Specifications:

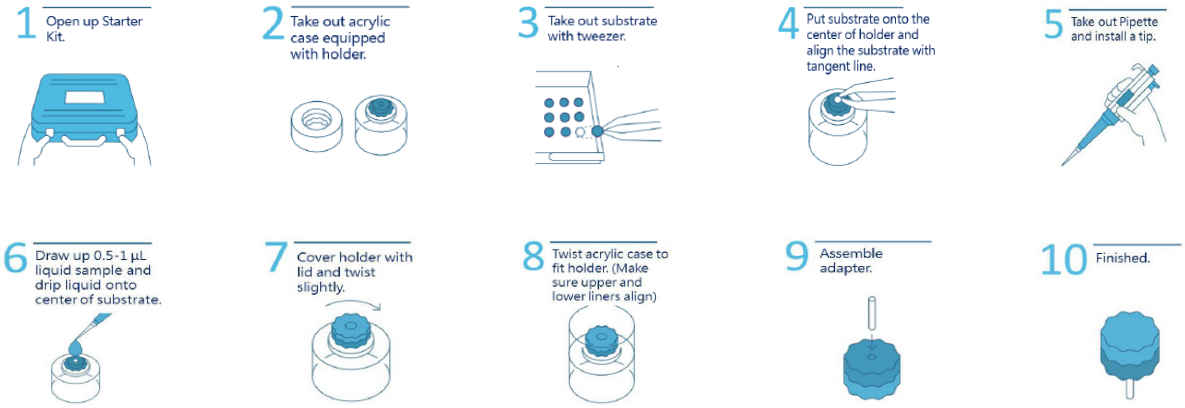
	Standard	High Mag.	Large Window
Applicable Sample	For (metal) samples with high atomic numbers and particle size exceeding 100 nm(ex. silver paste).	For samples with low atomic numbers and particle size not exceeding 100 nm (ex. biological samples, carbon black, SiO_2).	For EDS analysis, low concentration samples, or dry magnetic powders.
Chip Window (Si_3N_4)	**Chip Thickness:** 50nm **Window Size:** 150μm x 150μm	**Chip Thickness:** 20nm **Window Size:** 20μm x 20μm	**Chip Thickness:** 30nm **Window Size:** 250μm x 250μm

10. Micro-Channel Substrate Specifications:

Material Size	0.1μm - 0.5μm	0.5μm-2μm
Space Thickness	0.5μm	2μm
Illustration		

Operation

1. Steps:



Operation

2. Conditions for Operation:

a. Condition for the sample liquid to be observed:
   - There is no particular restriction for the pH value, however, HF base acid and hot H3PO4 cannot be used.
   - There is no particular viscosity restriction for the sample liquid.
   - The sample liquid should be kept at room temperature.

b. The membrane (which cannot be disassembled) should be used only once.

c. Before installing it onto SEM, confirm the integrity of the film. The customer should go to the OM to ensure that the film is a complete square after packaging.

d. Start by focusing on the borderline under the SEM (see Figure 1).

e. Handle the product carefully to avoid damaging the membrane.

3. Instructions:

a. Firmly support the hand holding the pippette with the other hand when using the pipette to avoid poor placement due to hand shaking (see Figure 2).

b. When placing the test sample on the Micro-Channel Substrate, avoid placing the test sample on the Microscopic Fluid Chip because the the Microscopic Fluid Chip is easy to break.

c. When the holder is attached to the upper cover, an acrylic case to fasten it tightly. It is fastened tightly when it is completely in line with the shape (see Figure 3).

d. Please align the tenons before locking when putting the Microscopic Fluid Chip onto the holder (see Figure 4).

e. The O-ring can be removed and fitted to the extraction hole (see the red circle in Figure 5). It is recommended to use the finer tweezers to remove and fit the O-ring. After changing the O-ring, do the sealed vacuum test.

f. Go to the OM to observe the Microscopic Fluid Chip. If it is found to be dusty before use, use a tool to blow off and remove the dust. **Do not** blow air straight at the Microscopic Fluid Chip (see Figure 6).

g. Sealed vacuum test:
   i. Drop 1 μL water in the holder and place it into the vacuum chamber.
   ii. After 60 minutes, open the lid and check the status of the liquid. The base pressure should be around 10-4 Pa.
   iii. The water droplet should still be in its original state and not dried out after 60 mins pumping.

4. Please observe the following guidelines:

a. Wear gloves while using the kit to avoid affecting the sample.

b. The Microscopic Fluid Chip(lid) film on the top cover is very thin and easy to break. The obersvation image will be affected if the film breaks. Be very careful when picking it up. It primarily moves horizontally, and the resistance of moving up and down may damage it. **Do not** touch or use air guns. If there are abnormal streaks in the image, the film on the cover of the Microscopic Fluid Chip(lid) might have been damaged.

Figure 1

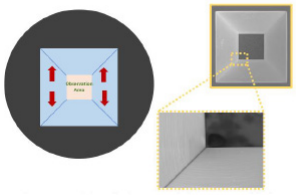


Figure 2

Figure 3

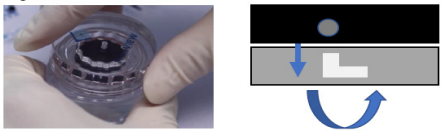


Figure 4

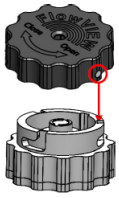
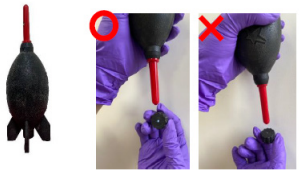


Figure 5

Figure 6



Operation

4. **Please observe the following guidelines:**

   c. **Do not** place the sample on the Microscopic Fluid Chip(lid) film to prevent damage.

   d. Lock the connection between the Adapter and the stage so that placing the SEM does not affect the observation image.

   e. To avoid the risk of contamination or leakage caused by putting in the SEM, the holder and the Microscopic Fluid Chip(lid) must be completely tight.

   f. **Do not** reuse the disposable Microscopic Fluid Chip (lid).

   g. To clean a used holder, wipe it with an alcohol-free and dust-free cloth.

Frequently Asked Questions

1. **Will the FlowVIEW Aquarius Starter Kit fit my SEM?**
   The FlowVIEW Aquarius Starter Kit is compatible with 99% of the SEM models and makes on the market.
2. **Can the Aquarius Starter Kit be used in high vacuum mode?**
   The Aquarius Starter Kit is designed to withstand 1x10-4 Pa.
3. **Why is no drying needed with FlowVIEW technology when it is necessary to dehydrate the sample in conventional SEM imaging?**
   A conventional SEM is a high-vacuum instrument, and the samples would be dried out after putting into the chamber. The FlowVIEW develops a liquid cell whose membrane can protect the sample from being dried out by the pump while allowing the e-beam to fully penetrate and scan the sample.
4. **What is the resolution of the FlowVIEW?**
   The highest resolution possible is 7 nm, varying depending on material properties, solvents used, and SEM used.
5. **How deep below the membrane can imaging be performed?**
   The spacer can restrain the membrane to the thickness of 0.5μm and 2μm. The actual penetration depth would vary depending on material properties, solvents used, and SEM accelerating voltage.
6. **Does the membrane interfere with imaging?**
   After penetrating the membrane the E-beam will slightly scatter.
7. **Could the electron beam damage my sample?**
   Samples usually may be damaged when examined with SEM, and the same may happen with FlowVIEW's liquid cell. If damage occurs, it may not be as severe due to the membrane.
8. **Are there any restrictions to the electron beam current?**
   Restrictions vary according to SEM used. If the current is too high, noise and a low-resolution image may result.
9. **Is there any problem charging with SEM?**
   The charging effect of Aquarius Starter Kit is far less in comparison with conventional SEM. However, charging effects still exist when scanning certain samples. These effects could be largely eliminated by using BSE imaging.
10. **Are the Aquarius Starter Kit compatible with light microscopes?**
    Yes. The membrane that the FlowVIEW creates is not hindered by electron beams or light beam. A light microscope has a relatively low magnification, so it is recommended that the large window version be used.
11. **Can I perform EDS with FlowVIEW technology?**
    Yes. FlowVIEW technology can perform EDS analysis.
12. **What imaging parameters should be used in FlowVIEW technology?**
    Typical acceleration voltages range between 5 and 20kV. It is recommended to use low acceleration voltage for samples with low atom numbers when testing.
13. **Does the liquid sample leak from the Aquarius Starter Kit and contaminate the detector of my SEM?**
    The liquid sample will not leak from the kit under normal operations. Even if there is leakage, the impact on SEM would be negligible due to the small volume of liquid.
14. **Can I image any liquid with the FlowVIEW Aquarius Starter Kit?**
    Samples with concentrations that are too low or sizes that are too small may not be able to be used for observation.

Other Notes

1. **Disclaimer:** The manufactures is not responsible for any damage to Aquarius Starter Kit and SEM equipment and is neither responsible for any claims by government authorities nor claims by any third parties if users do not follow the manual.
2. **EDS Recommended Use – Large Window:** The wafer is an inverted pyramid from the side, with an included angle of 54.7 degrees (all versions of the film are 54.7 degrees, the only difference is in the gray dotted line area, which is the window size we marked: high mag is 20 μm, the standard is 150 μm and large window is 250 μm.

   No matter the take-off angle is 30 degrees or 50 degrees, the angle is fixed without tilting, so the longer the horizontal distance of the window, the higher the height that the x-ray signal can move in the vertical direction, and there are more chances Fly over the Si frame wall and be received by the detector.

   The window size of the High mag membrane is only 20 μm*20μm, and the take-off angle of the x-ray is about 30 degrees, so it may be blocked by the Si frame and cannot reach the detector.