

"NanoSuit Solution Type III" - For Cells, Bacteria, etc.

Instruction manual

The NanoSuit method is a technology that enables us to keep many organisms wet in the high vacuum condition of a scanning electron microscope (SEM) by encasing the sample in the NanoSuit membrane. The NanoSuit membrane is a thin, vacuum-proof membrane with sufficient electron conductivity for an SEM observation, and is polymerized from the NanoSuit solution composed of biocompatible polymers by an electron beam or plasma irradiation. The NanoSuit Solution Type III (NST3) is suitable for an SEM observation of small-sized biological samples such as cells, bacteria, viruses, exosomes, and liposomes. The basic procedure is simple as follows; the NST3 is immediately added dropwise to the sample, after removing the solution such as the culture medium which contained those samples. Wipe off the excess NST3 and irradiate it with an electron beam in an electron microscope to start observation.

Follow the instructions below to proceed with your observations and experiments.

1. NanoSuit Solution Type III (NST3) has been developed for observing biological samples such as cells.
2. When handling the NST3, wear glasses or goggles to protect yourself from accidental breakage and r scattering of glass wear. If the NST3 adheres to your skin during work, remove it with running water or detergent.
3. Since the NST3 is polymerized under electron-beam irradiation and forms NanoSuit membrane that protects biological samples, please start electron microscope observation as soon as vacuuming of the sample chamber is completed.
4. Store the NST3 in a freezer. When using, bring it to room temperature and stir well with a pipette. The remaining NST3 can be stored frozen again in a light-shielding bag. When reusing, bring it to room temperature in the same procedure and stir well.

How to use

<For samples such as cells adhering to a glass substrate>

1. If the sample on the substrate is covered with a solution such as a culture medium, remove the solution with a pipette or a filter paper. (In the case of cultured cells, a small cover glass should be placed on the bottom of the culture plate in advance to culture cells on it. Then the cells are easily taken out together with the cover glass.)
2. Before the sample dries up completely, fix the sample (such as substrate glass) on the sample table of the electron microscope using such as a carbon tape. Drop the NST3

on the surface of the sample and immerse. (The shape of the sample may be changed or died due to the difference of osmotic pressure between the original culture solution and the NST3. Please be sure to perform a preliminary experiment before this procedure.)

3. Wipe off the excess solution with filter paper.
4. Set the sample table in the sample chamber of the electron microscope and evacuate.
5. After evacuation, start electron microscope observation immediately. (If the surface image of the sample is vague, the wiping of step 3 described above might be insufficient. After treating the sample with this solution, wipe it off sufficiently. If it does not improve, it might be improved by diluting this solution about two times or more with such as culture medium.)

<For floating samples that have not adhered to the substrate>

1. When the sample is submerged in the bottom of the container, remove the supernatant with a pipette. If the sample is suspended in the culture medium, transfer the medium in a centrifuge tube. After the centrifugation, remove the supernatant gently and reduce the solution around the sample as much as possible.
2. Add the NST3 to the container containing the sample and gently pipette to blend the sample with the solution. (The shape of the sample may be changed or died due to the difference in osmotic pressure between such as the culture solution and the NST3. Be sure to perform a preliminary experiment before this procedure.)
3. Take an appropriate amount of the above-mixed solution containing the sample with a pipette and drop it on the sample table of the electron microscope. Wipe off the excess solution very gently with filter paper.
4. Set the sample table in the sample chamber of the electron microscope and evacuate.
5. After evacuation, start electron microscope observation immediately. (If the surface image of the sample is vague, try diluting this solution 2-fold or more with such as culture medium.)

Note

This NST3 is a product developed for observing ultrafine structures such as cells, but it may be difficult to observe depending on the characteristics and conditions of the sample. Be sure to perform a preliminary experiment before proceeding with this observation and experiment.