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K-kit instructions for use

K-kit has been primarily developed for TEM imaging of liquid samples in their native state. The **Liquid or Wet Mode** of operation is especially useful for studying chemical reactions in liquids, such as the gold reduction process in AuCl4 solution by electron beam energy.

It is also possible to use K-kit for controlled drying of liquids. By using this **Thin Layer Dried Mode** with K-kit, one can get images of nanoparticles distributed uniformly without aggregation and are similar to those in liquid. This is especially useful for particles that tend to aggregate when drying on copper TEM grids using more conventional methods.

K-kit prevents particles from aggregating and maintains them in-situ as they would be when suspended in liquid, allowing nanoobjects, aggregates, and agglomerates (NOAAs) to be characterized. Studies have shown that when using Thin Layer Dried Mode a thin liquid layer will be formed on the walls of K-kit trapping particles in-situ until they have dried. Results are comparable to those observed in Wet Mode.

If you are familiar with the handling and use of standard TEM grids, then you should find that K-kit is relatively easy and straightforward to load and prepare liquid specimens for TEM observation.

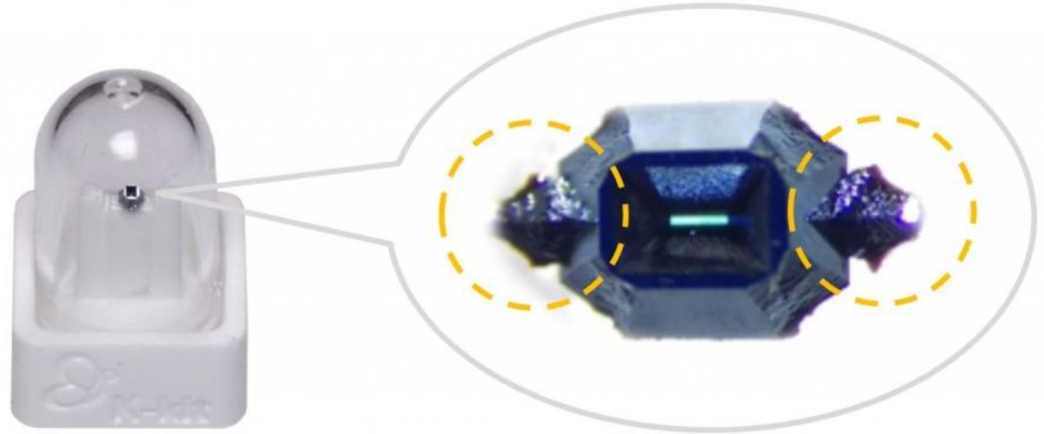
Guidelines for using K-kit

1. Unpack the K-kit carrier

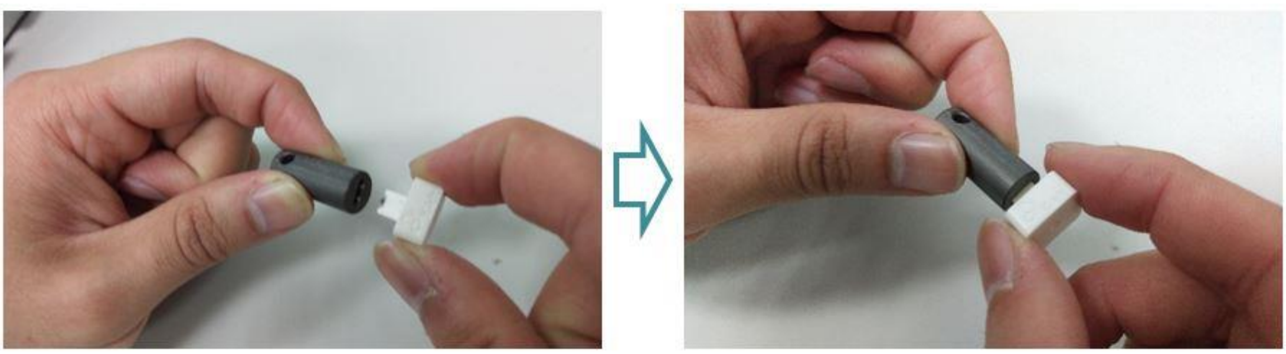
Remove a complete K-kit and carrier from the shipping box. Keeping the clear cap in place to protect the K-kit, push a paper clip or similar point through the holes at the corners to push out the bottom compartment, which contains the copper hole grid. You will require this at a later stage in the process.

2. Remove the channel tips to expose the microchannel

With the K-kit mounted on its carrier use the channel opener tool to open the microchannel by breaking off the channel tips. The microchannel of each K-kit is hermetically sealed and protected with channel tips at each end to ensure the membrane surfaces remain pristine until ready for use.



Remove the channel tips by inserting the K-kit carrier top into the opening of the tool and gently pushing to the end. The channel opener tool has a self-guiding slot and a mechanism to safely break off the tips before the carrier top plate reaches the end stop.

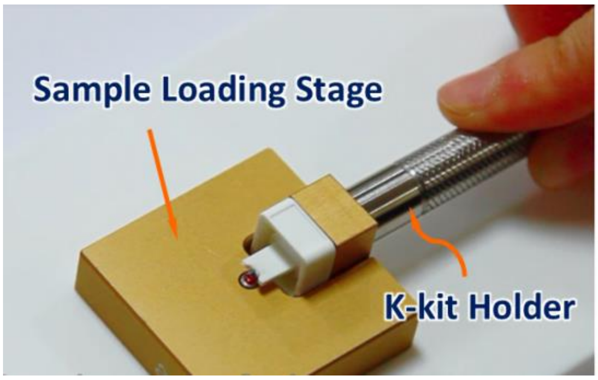


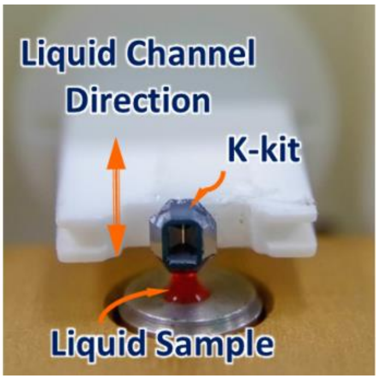
3. Liquid Loading

The loading stage and K-kit holder enable your liquid sample to be loaded into a K-kit in a controlled manner. Place about 2 microlitres of liquid at the centre of the sample loading stage and then place the white K-kit carrier holding the K-kit onto the end of the K-kit holder.

Place the notch of the K-kit holder onto the horizontal rod of the loading stage. This enables the K-kit and carrier to pivot on the loading stage allowing the K-kit to be moved towards the liquid.

By gently lifting the back of the K-kit holder lower the K-kit so that it makes contact with the liquid, allowing it to fill the microchannel by capillary action. You can observe the liquid surface being “pulled up” by the K-kit. Keep the K-kit steady for approximately one minute to allow filling to complete.





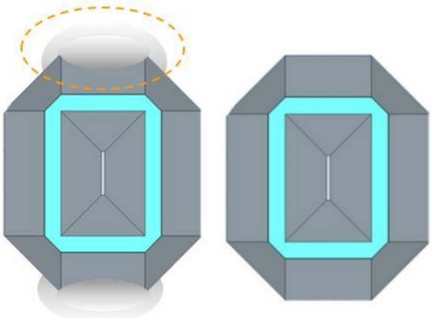
4. Vacuum seal

Prepare the sealing and mounting glues on a glass slide. Each glue is made up of two components which need to be mixed. Take a small drop of the two parts of the Torr Seal epoxy sealing glue and mix on the slide by using a stirrer (supplied) or cocktail stick. Similarly, mix the two components of the mounting glue on separate part of the glass slide. The sealing glue is vacuum compatible and has very low outgassing during the curing process, which also ensures that no volatised gases from it are able to flow inward to react with the liquid sample inside.

Place the K-kit carrier on the gluing stand. Use the needle pen to pick and apply the Torr Seal epoxy on to the channel openings. Cover the channel openings at both ends with an adequate amount of glue.
NB. To ensure that the liquid is retained in the K-kit, it is recommended that the channel-seal gluing is completed within one minute of liquid loading.

If you plan to use the Thin Layer Dried Mode of K-kit this step is not required as both ends of the microchannel are left open to atmosphere.

In Wet Mode the microchannel is sealed at both ends (a) and in Thin Layer Dried Mode the ends are left open (b).



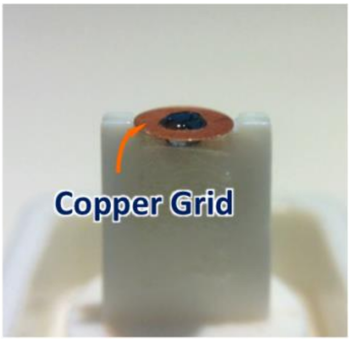
a b

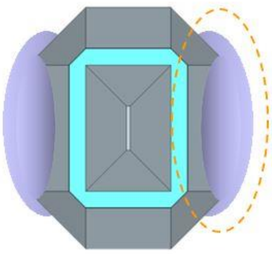
5. Copper grid mounting

A copper hole grid (supplied) is required to load and seat the K-kit correctly in a TEM grid holder. This needs to be attached to the prepared K-kit using the epoxy mounting glue for both Wet Mode and Thin Layer Dried Mode.

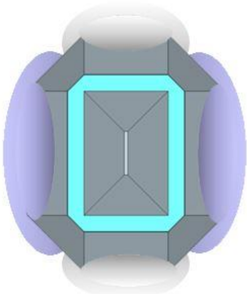
Keeping the K-kit carrier on the gluing stand use the needle pen to pick and apply the mounting glue from the mixed components on the glass slide onto the long side edges of the K-kit.
Place the copper hole grid onto the top of the K-kit. Steps on the carrier top plate facilitate centring and levelling of the copper grid.

NB. Take care not to get glue on the channel ends of K-kit when using the Thin Layer Dried Mode of observation as this will inhibit drying of the microchannel.





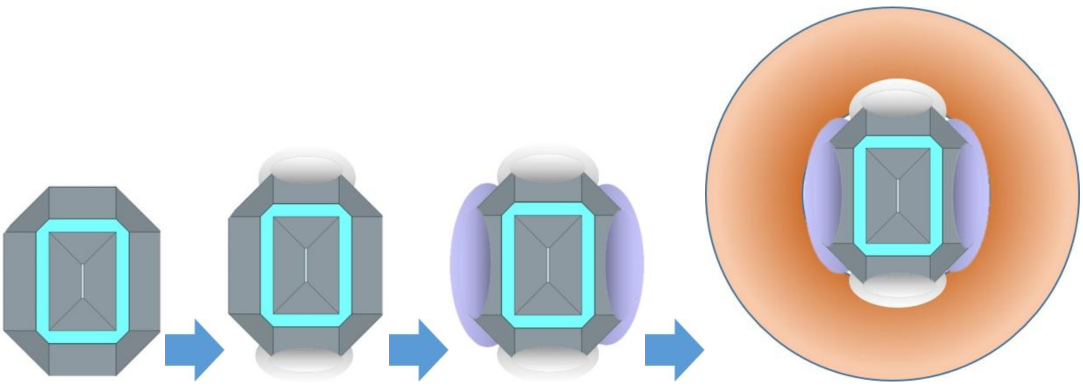
Thin Layer Mode (dried)



Wet Mode

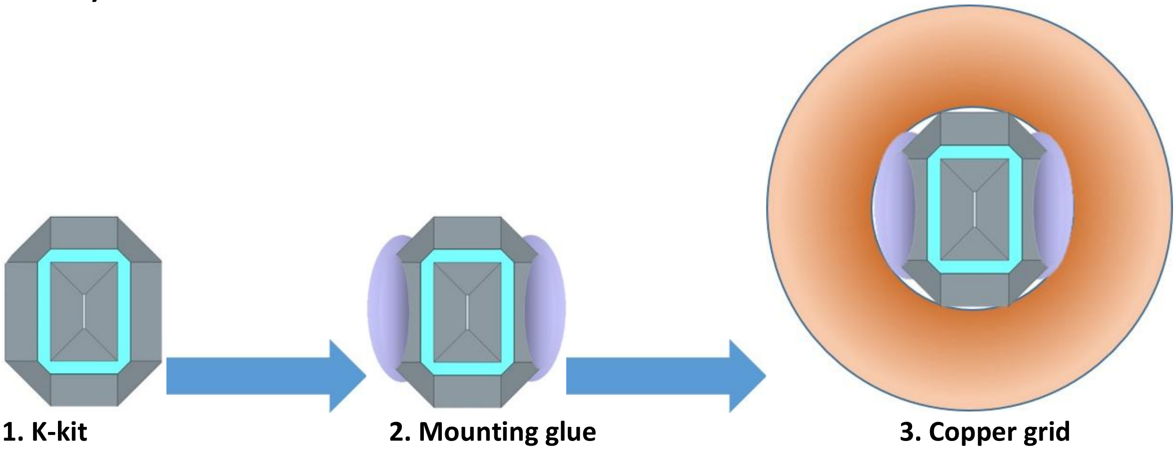
K-kit gluing procedures

Wet Mode



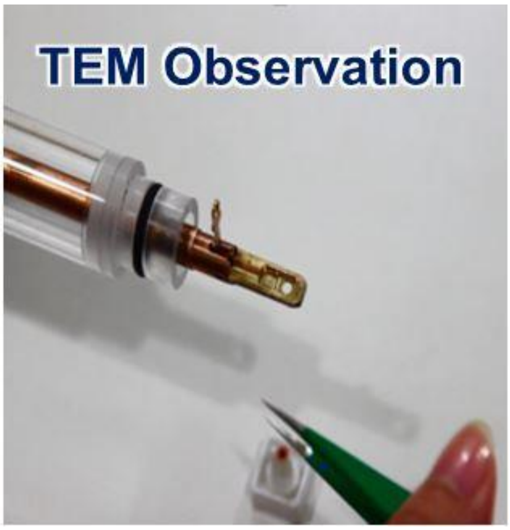
1. K-kit **2. Vacuum glue** **3. Mounting glue** **4. Copper grid**

Thin Layer Dried Mode



6. Vacuum pumping

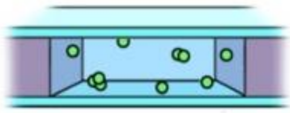
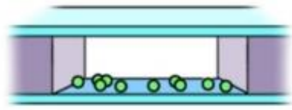
Place the glued K-kit (on its carrier without the clear cap) into a small vacuum device. Vacuum pump the K-Kit for 30 minutes. The K-kit is then prepared and ready for TEM observation. Remove and load into the TEM grid holder.

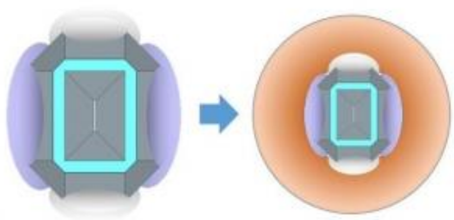


K-kit Wet and Dried modes

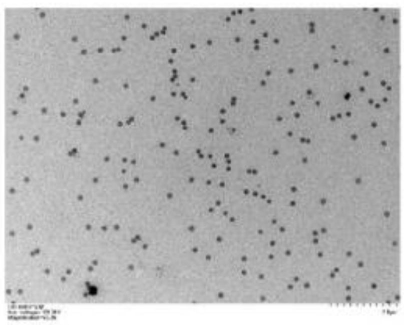
K-kit can be used in two ways depending on the application and the required purpose for observation. These are: i) Dried Mode (Thin Layer Mode) and ii) Wet Mode

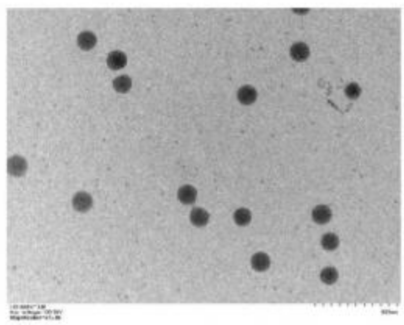
For particle characterisation where the prime objective is to image the shapes, sizes, or aggregation and agglomeration (NOAAs) of particles in liquid, Dried Mode of K-kit is recommended. This will usually give very good image quality and resolution in a dried state.
If the objective is to study chemical reactions in liquid, such as the gold reduction process in $AuCl_4$ solution by TEM electron beam energy, then Wet Mode of K-kit is recommended.

Sample Preparation	Wet Mode	Thin Layer Mode
Inner Status of K-kit	With Liquid 	Dried 
Imaging Resolution	Good	Excellent
Gap Size (Considered)	300～500nm	2000～3000nm
Particle Size (Loadable)	10nm～300nm	3nm～2000nm
Particle Shape	Keeping original	Potentially, could be deformed.
Chemical Reduction or Potential Damage by Electron Energy	High	Low



Wet Mode





The loaded liquid sample is sealed and imaged using TEM in the native liquid environment.

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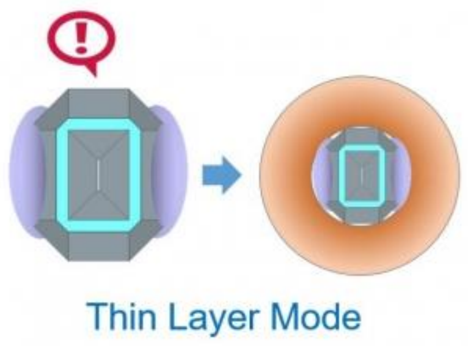
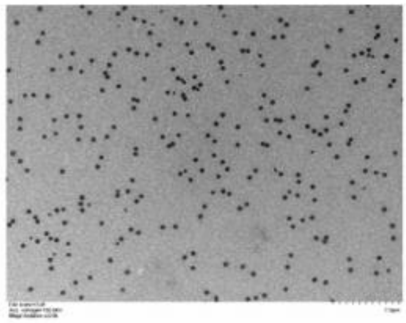
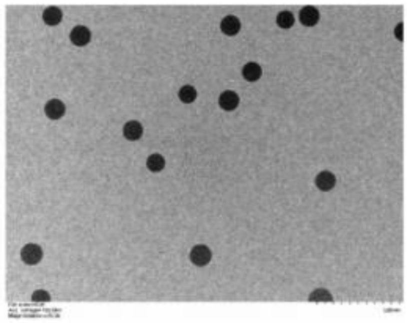
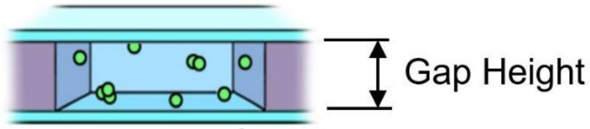






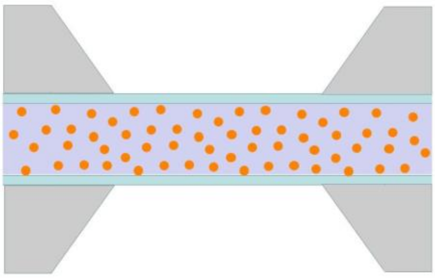
A patented liquid drying protocol preserves the original morphology and physical state of nanomaterials with improved imaging resolution.

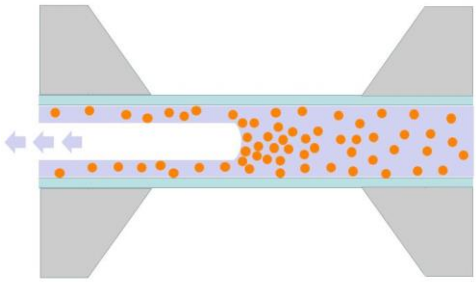
The channel gaps of K-kit can also make a big difference. Basically, we recommend using a 2µm gap K-kit for Thin Layer Dried Mode and a 0.2µm gap for Wet Mode. This is because liquid loaded by a K-kit with a larger gap height is normally much easier to evaporate or pump out at room temperature than by in one with as smaller gap height. To ensure that the sample liquid is well dried it is essential to keep both ends of the channel open to atmosphere. The gluing step is omitted.



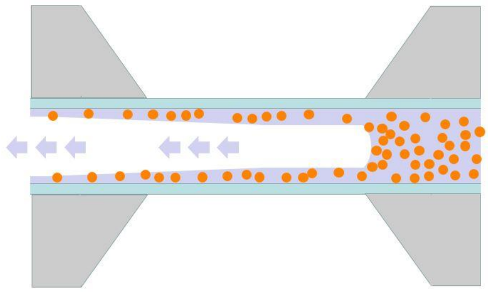
Gap Height (um)	0.1	0.2	0.5	1.0	2
Wet Mode	●	●	◐		
Thin Layer Mode	◐	●	●	●	●

Mechanism of Thin Layer Dried Mode

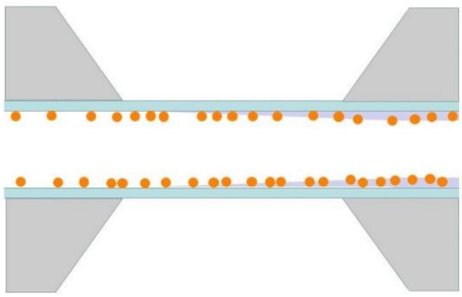

K-kit filled with liquid sample



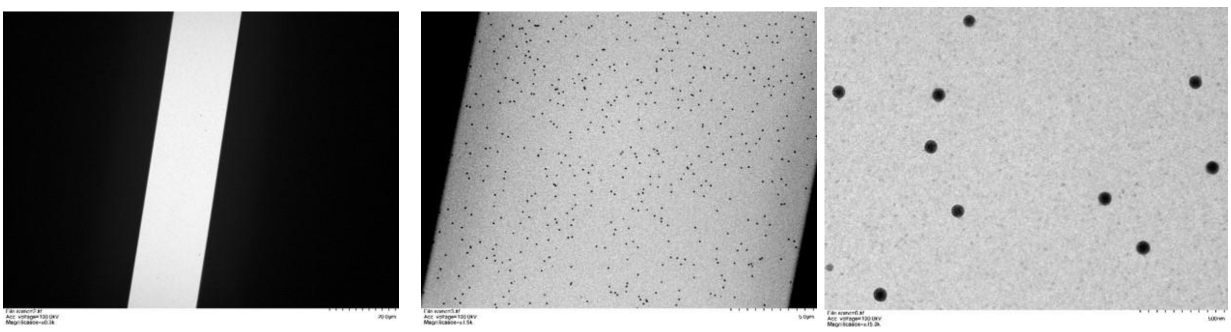
Drying by vacuum pumping



A thin liquid layer formed on the inner walls with in-situ trapping the particles during drying



The native state of particles is maintained after drying onto the chamber walls



An example of Thin Layer Dried Mode K-kit using a gap of 0.2µm. A polystyrene bead solution

Using Thin Layer Dried Mode can allow the formation of a stable liquid layer of a certain thickness on the inner wall of K-kit. Nanoparticles suspended in the liquid will be trapped in-situ and kept in their native state until the liquid has dried.

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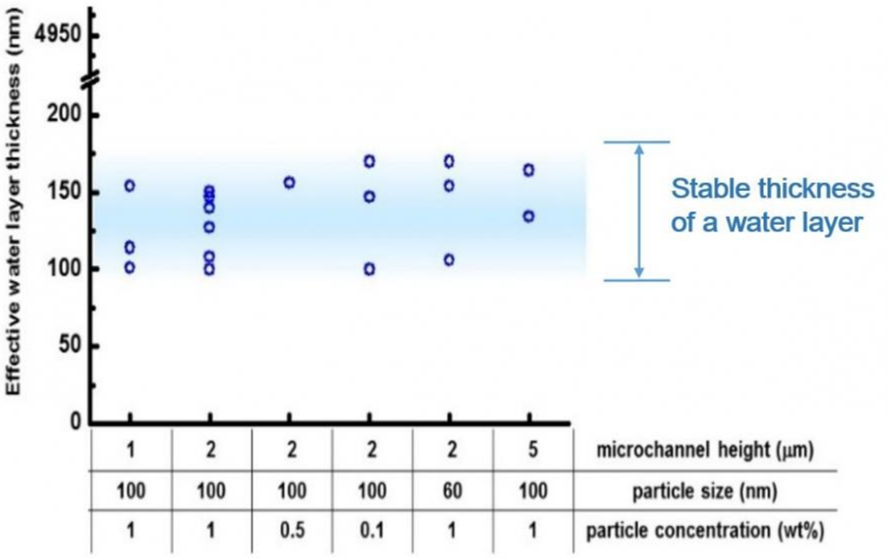
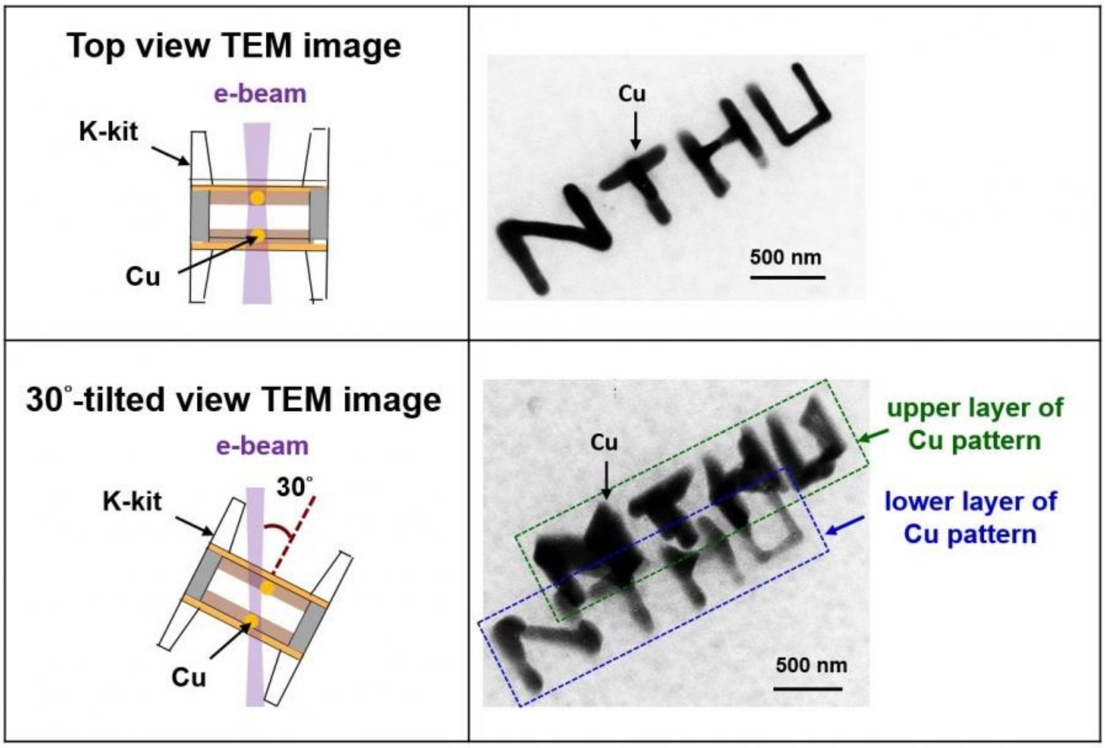
Liquid layer observations in K-Kit Thin Layer Dried Mode

Quantitative counting and EELS data showing a water of a consistent thickness layer in Thin Layer Dried mode



Channel Gap Height	1um	2um	5um
Water Layer Thickness	100 – 155 nm	100 – 170 nm	130 – 160 nm



TEM images of Cu pattern, formed by e-beam writing directly on a K-kit loaded with $AuCl_4$ in Thin Layer dried mode.

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References

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3. "Direct-Writing of Cu Nano-Patterns with an Electron Beam,"Microsc. Microanal. 21, 1639–1643, 2015.
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