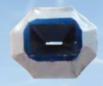
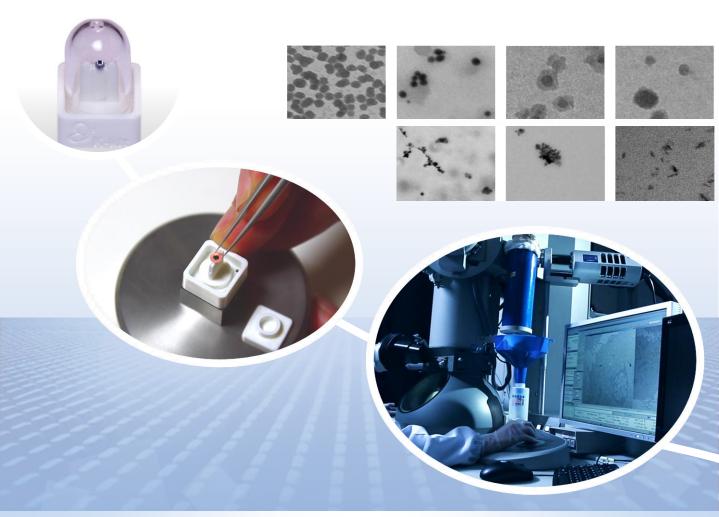
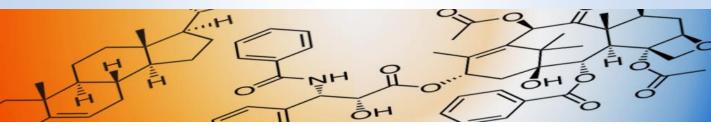
K-kit





A Specimen Holder for Liquid Sample Analysis in TEM

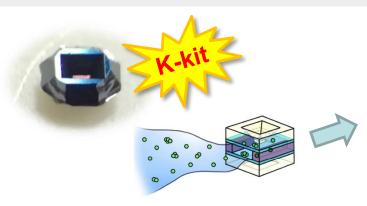


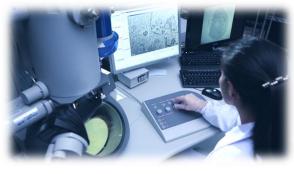




An Innovative Specimen Holder for Liquid Analysis in TEM

- K-kits are sample holders designed to be used for microscopy observation of liquid samples, allowing nanoobjects, aggregates, and agglomerates (NOAAs) in liquid samples to be imaged and characterized in TEM, FIB, and STEM etc. microscopes.
- With vacuum compatible sealing of liquids in electron-transmitting thickness, K-kits are micro reaction chambers for countless experiments in materials, chemical and biological research.



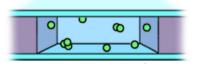


Transmission Electron Microscope (TEM)

Patents being issued and publication

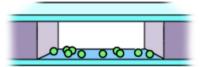
- ✓ US 7807979 B2
- ✓ US 8969827 B2
- ✓ Anal. Chem. 2012, 84: 6312-6316

Wet



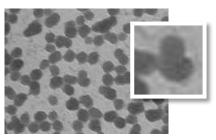
 The loaded liquid sample is sealed and imaged using TEM in the native liquid environment.



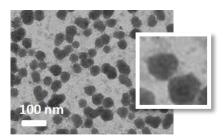


(Loaded liquid was dried out by pumping.)

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



(Acceptable image quality with liquid inside the K-kit.)



(Very good image quality, when liquid was dried out by pumping.)

TEM images shown: Undiluted Chemical-Mechanical Polishing (CMP) slurry directly loaded into K-kit.

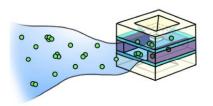


Product Features

Disposable

Free of Cross Contamination

Easy Use

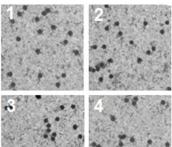


K-Kit

Original physical state

Conventional

Aggregated after drying on Cu grid









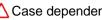


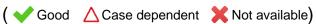


TEM images, as shown: NIST traceable polystyrene beads. Scale bar: 500nm









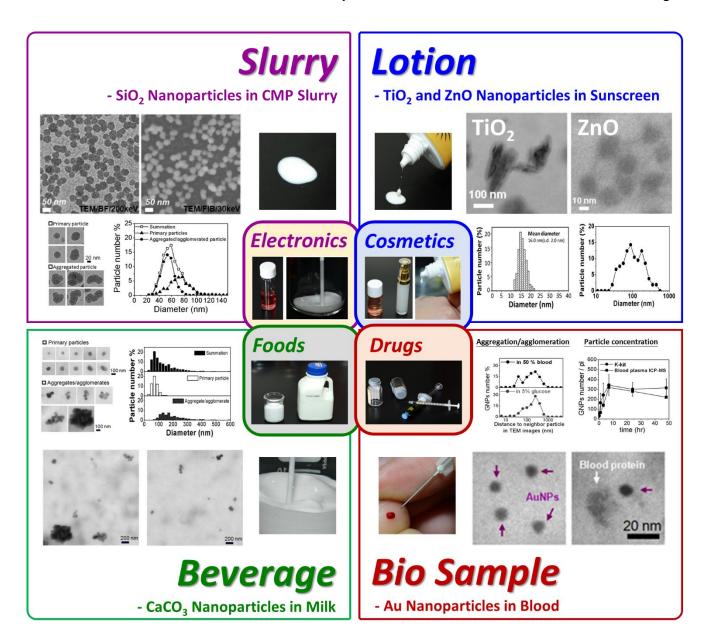
Physicochemical Parameters	K-kit	Cu-Grid
1. Composition	*	*
2. Size	*	*
3. Shape	*	*
4. Size distribution	*	Δ
5. Aggregation and agglomeration in liquid	*	×
6. Particle concentration	*	×
7. Liquid TEM observation	~	×

Compared with other products

Company	Bio MA-Tek	Protochips	Hummingbird Scientific
Liquid Cell	K-kit	E-chip	TEM Holder
Chip Size	1.4 x 1.7 mm (fits in 3 mm grids)	4 x 6 mm	
Special holder	No need	needed	needed
Cost	US. 100-200	US. 100-200 (+Special Holder: US.100K)	US. 150K

Applications

Characterize NOAAs in electronics industry, cosmetics, foods, medical devices, and drugs.



Reference:

- 1. US FDA 2012, Guidance for Industry Safety of Nanomaterials in Cosmetic Products.
- 2. EU/JRC July 2012, Requirements on Measurements for the Implementation of the European Commission Definition of the Term "Nanomaterials".
- 3. ISO/TR13014: 2012, Nanotechnologies -- Guidance on physico-chemical characterization of engineered nanoscale materials for toxicologic assessment.
- 4. ICCR 2012, Characterization of Nanomaterials II Insolubility, Biopersistence and Size Measurement in Complex Media.



K-kit Adaptability









TECNAI F20 Holder

EM31640 STHB Holder

H-7501 SS Holder

Strong Structural Reliability under Vacuum



Torr Seal® Epoxy

- A trusted and widely used glue, suitable for high vacuum systems.



Window

Silicon Nitride

- Material intrinsically tough, durable to withstand drastic pressure changes.

(Torr Seal®, a trade mark owned by Agilent Tech. Inc.)

Sealing glue compatible to many solvents

(MEMS, Micro-Electro-Mechanical Systems)

- The following table shows the test results of Torr Seal Epoxy soaked in chemical solvents for 24 hours and then examined using FTIR (if dissolved) and visual observation (if dispersed).



	Water	PEG400	DMSO	Ethanol	0.1 N HCI	0.1 N KOH
Compatibility (FTIR)	*	*	*	*	~	*
	Hexane	IPA	Methanol	DCM	THF	Acetone
Compatibility (FTIR)	*	~	~	×	×	×

(FTIR, Fourier Transform Infrared Spectroscopy)

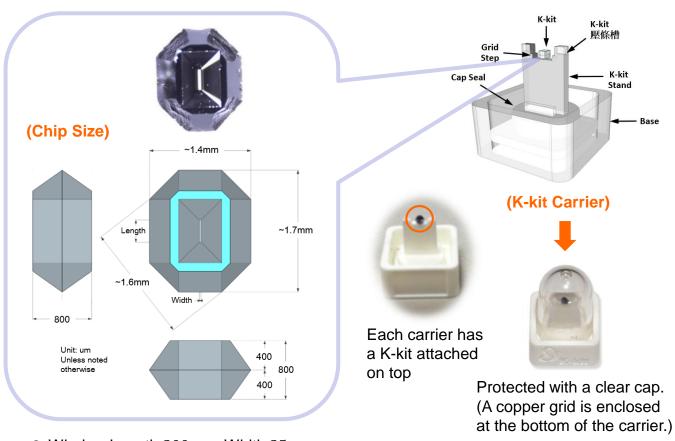
Compatible (FTIR not detected)

Use with care (FTIR detected)

X If you would like to learn more about K-kit or have any question about its applications, do not hesitate to contact us. To enable potential customers to experience the benefits of K-kit, we offer preloaded demo samples for free. Contact us at E-mail: sales@bioma-tek.com

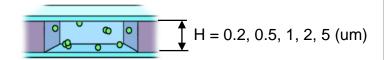


Shipping Packages and Tool Sets



- Window Length 300 μm , Width 25 μm
- Channel Height (H): 0.2 and 2.0 standard

0.5, 1.0 and 5.0 available





4 K-kits

6 K-kits

(Shipping Packages)



Copper Grid

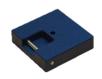


Channel Opener



Starter Box (Glues, Needles, Channel Opener, etc.)

(Tool Sets and Consumables)





K-kit Gluing Stand Sample-loading Stage





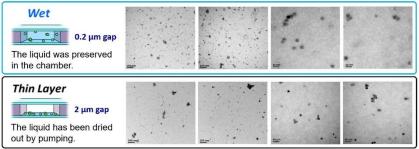




K-kit Meets All Needs for Liquid TEM

1 Native State in Liquid

QDs Particles in Chloroform



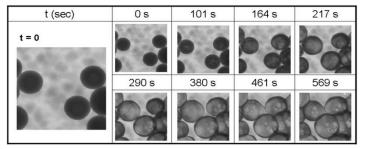


- Available with undiluted solution.
- Preserve the original morphology and physical state in liquid.

In-situ Observation

 Kinetic mechanism of metal growth or physicochemical reaction process in liquid can be in-situ observed with increased reaction time.

Dynamic Observation of Silicate Nanoparticles



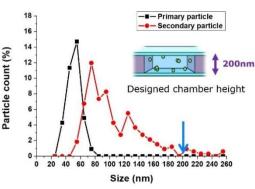
3 Quantitative Analysis

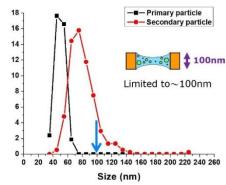
• Software of image recognition for nanoparticle size distribution analysis.

Abrasives in CMP Slurry (K-kit vs. Cyro)

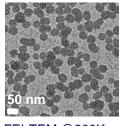
Particle count (%



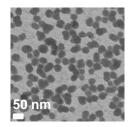




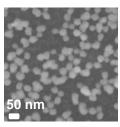
Compatible to Versatile Microscopy Analyses



FEI-TEM @200Kev



Hitachi-TEM @100Kev



FEI-STEM @30Kev

- Applicable to TEM, FIB, and STEM.
- Available for EDX analysis.
- High resistance to most chemicals.
- Working temperature range from -40°C to 120°C.



K-kit Tool Box

• Tool box, we offer a full tool set, including K-kit holder, sample-loading stage, needle pen, K-kit gluing stand, recommended glues, glass slides and some replacement parts.







Accessory Box



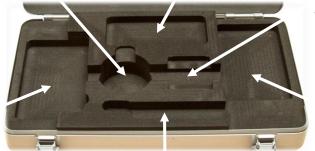
K-kit Gluing Stand



Glass-slide Pack (Six slides inside for free)



Sample-loading Stage





K-kit Holder & Needle Pen

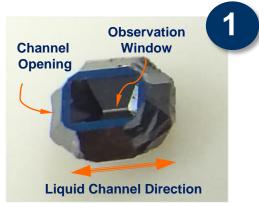


K-kit Shipping Package (Without K-kits)

X K-kit tool box can be in silver or vermillion colors

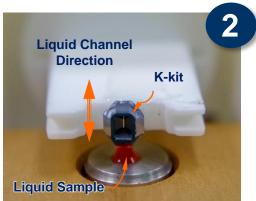


Sample-loading Procedure



1.K-kit:

K-kits are Si-based microchannel devices with silicon nitride windows that allow SEM, FIB, STEM, and TEM observations. The shape is a result of anisotropic wet etching. The liquid channel is parallel to the window, with openings at both sides.



2.Filling:

Liquid fills the channel through capillary force. The liquid surface is "pulled up" by the K-kit. Keep the K-kit steady for approximately 1 min to allow the filling to complete. The aqueous liquid sample should be placed on a glass slide. Both the K-kit and glass surface are hygroscopic. Do not immerse the K-kit in liquid to prevent from the window being contaminated.





3. Torr-seal:

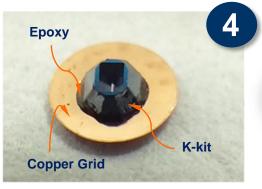
Cover the channel openings at both ends with Torr Seal epoxy after filling the device with liquid. (No need to do this gluing step, if one would like to dry out the liquid and leave the nanoparticles a Thin Layer mode in K-kit.)



Use epoxy to mount the sealed K-kit to a copper grid by fitting it to the precut hole at the center of the grid.



QR code link to demo video





If on-line, please click the link to watch demo video: https://youtu.be/0QDvI0UTBUM



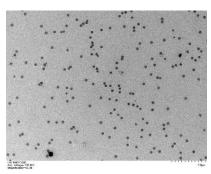
Wet and Thin Layer Mode of K-kit

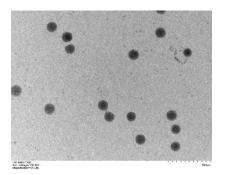
Sample Preparation	Wet Mode	Thin Layer Mode
	With Liquid	Dried
Inner Status of K-kit		080 0800
Imaging Resolution	Good	Excellent
Gap Size (Considered)	300∽500nm	2000〜3000nm
Particle Size (Loadable)	10nm~300nm	3nm∽2000nm
Particle Shape	Keeping original	Potentially, could be deformed.
Chemical Reduction or Potential Damage by Electron Energy	High	Low

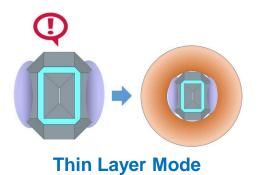
If making a Thin Layer (Dried) mode of K-kit, it's essential to keep both ends of the channel open to atmosphere, no need to do the channel gluing step.

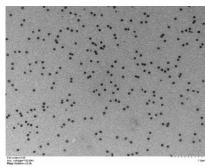
Gap Height (um)	0.1	0.2	0.5	1.0	2
Wet Mode	•	•	•		
Thin Layer Mode	•	•	•	•	•

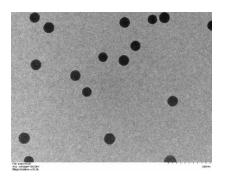






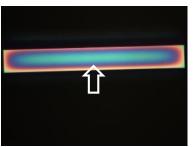




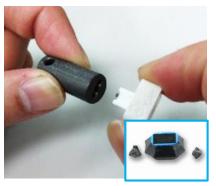


Matters Needing Attention When K-kit in Use



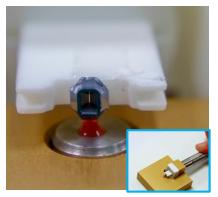


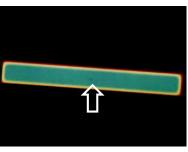
With Newton's rings (Sealed by channel tips)





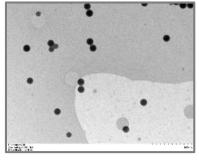
Flat membrane (Open to atmosphere)





With color patterns (After liquid filled)





Liquid well reserved (Soon to glue the openings)



Inspection Before Use

- With Newton's rings on the membrane. (Be sure the channel to keep vacuum sealed)
- Free of any damage on Silicon body of K-kit.



Channel Tips Removal

- Be sure to remove both the channel tips before using K-kit.
- It should be finished the liquid loading within 0.5 hour, after breaking the channel tips.



Liquid Loading

- Keep the K-kit steadily touching on liquid for around 1 minute, to allow the filling to complete.
- Do not immerse the K-kit in liquid.



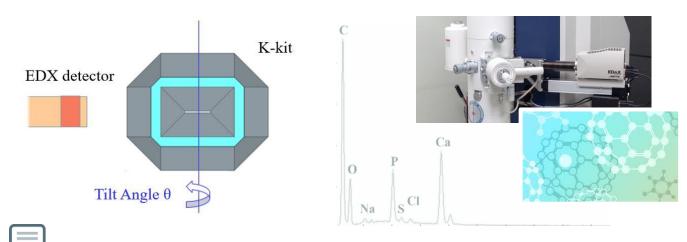
Gluing Process

- Glue both ends of the channel within 1 minute after liquid loaded.
- Be sure not to do the channel gluing step, if making for Thin Layer mode of K-kit.
- Doing the gluing step with care, to avoid the glue flowing into the observation window.

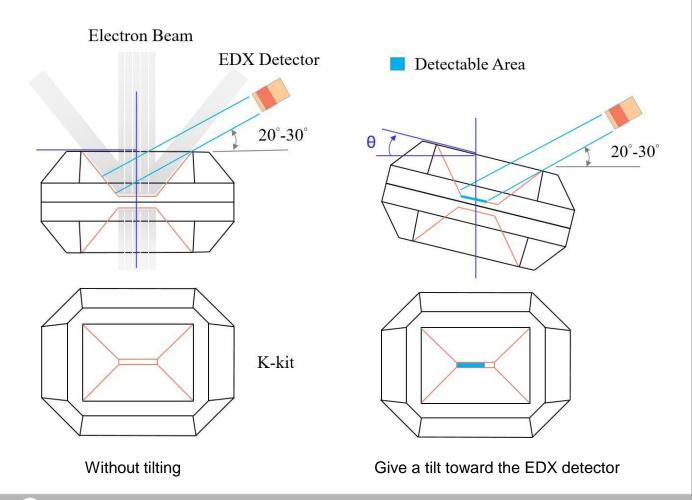


Available for EDX Analysis (1/2)

How to Make EDX Analysis Achievable on a K-kit



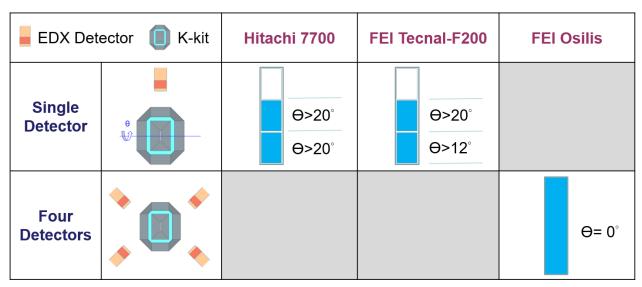
By pointing the window long side to the detector and tilting the holder at some angles, which could make EDX analysis achievable on a k-kit.



Available for EDX Analysis (2/2)

■ EDX available angles for different kinds of TEM equipment

Detectable Area



For some types of TEM installed with multiple EDX detectors, they usually can get a clear X-ray excited signal from K-kit, no need to turn any of body rotation or tilting.

Example:

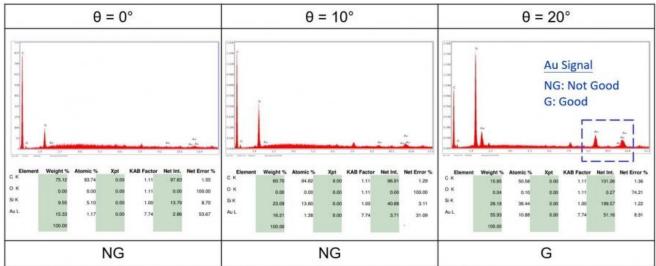
TEM: Hitachi 7700

· EDX: Single Detector

Liquid Sample: AuCl₃

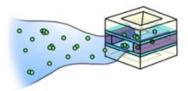
Tilt Angle: 0°, 10°, 20°

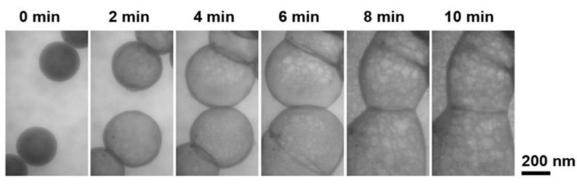




Dynamic observation of NOAAs in liquid

■ Dynamic observation of silicate nanoparticles in water





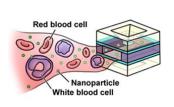
^{*} Exposed electron (100 keV, 4 X 10^4 A/m²)

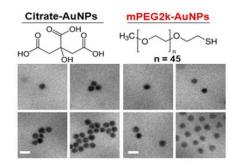
■ Dynamic observation of polystyrene beads

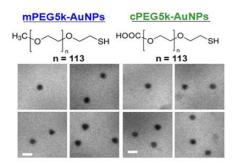
In situ dynamic observation by TEM **Observation Environment** (Hitachi H-7650) 150 s 199 s 0 s Vacuum, 4.0 X 10⁴ A/m² 35 s 0 s 69 s 98 s Air, 2.5 X 10³ A/m² 62 s 199 s 396 s 0 s Water, 2.5 X 103 A/m² 0 s 105 s 208 s 425 s **Buffer/** PBS (Sodium ion), 1.0 X 10⁴ A/m²

NOAAs of Au nanoparticles (NPs) in blood

 K-kit can be used to perform in-vitro and in-vivo physicochemical characterizations of NPs in blood by TEM.

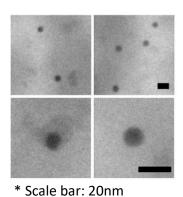


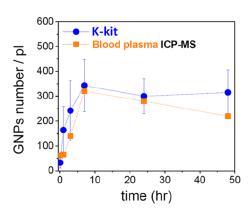




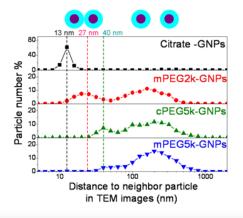
☐ Image-based statistic analysis of particle concentration (K-kit vs. ICP-MS)

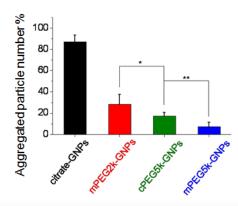






☐ Image-based statistic analysis of aggregation and agglomeration of Au NPs in blood

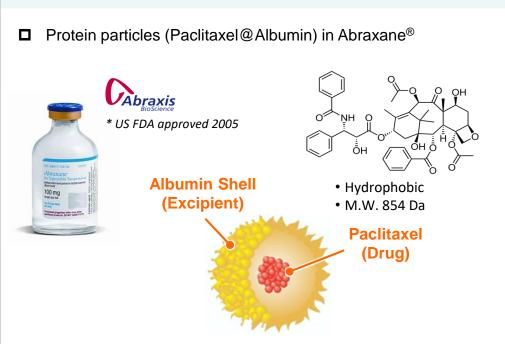


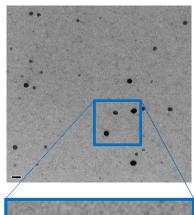


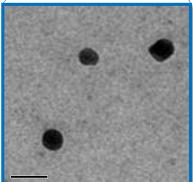


Protein particles in Nanopharmaceuticals

 K-kit can be used for characterizing protein particles in Nanopharmaceuticals by imaging the particle morphology, size and size distribution, to evaluate drug formulation or conduct any bioequivalence study.

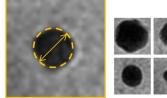


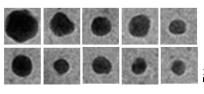




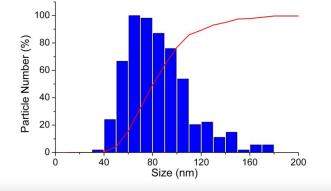
◆ Abraxane in saline _ size & size distribution (D10/ D50/ D90)

* Scale bar: 200 nm









- Total calculated particle #: 319
- Average size: 85.1 nm
- Standard deviation: 27.0 nm

Parameter	Size (nm)	
D10	55.6	
D50	80.1	
D90	122.2	
Span: (D90 - D10) / D50	0.831	

NOAAs of CaCO₃ NPs in milk

 K-kit can be used for characterizing nanoobjects of foods in final product form, to evaluate the safety risks of nanomaterials in food additives and in substances in contact with foods.

☐ Comprehensive physicochemical characterization

P	arameter	Results	Methods	
1	Composition	Calcite CaCO ₃	TEM/EDX, XRD	
2	Size / size distribution	Average Diameter / Standard deviation		
	Crystal particle size	36 / 4 nm	XRD	
	Primary particle size	73 / 26 nm	TEM	
	Powder size	17 / 10 μm	SEM	
3	Shape	Cubic	TEM	
4	Aggregation/Agglomeration			
	in relevant media	Average diameter / Standard deviation	K-Kit / TEM	
	NOAAs	115 / 73 nm	(4wt% in DI water)	
	Nano-Objects	68 / 20 nm (number 58%)		
	Aggregations / Agglomerations	180 / 70 nm (number 42%)		
5	Solubility/Dispersibility	< 0.01% in Ca ²⁺ form	ICP/MS	
		Dispersed in DI water > 4 wt%	K-Kit / TEM	
		(20 ~ 450 nm)		
6	Surface charge	-23.4 ± 1.3 mV (in DI water)	Zeta potential	
7	Surface chemistry	Surface atom:	XPS	
		C (35%), O(48%), Ca(16%)		
8	Specific surface area	18.14 m ² /g	BET	



As Additive

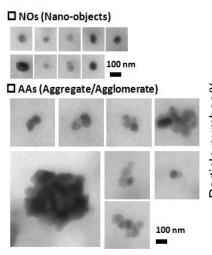


Size and size distribution

■ Size and size distribution of CaCO₃ NOAAs in milk



In Product

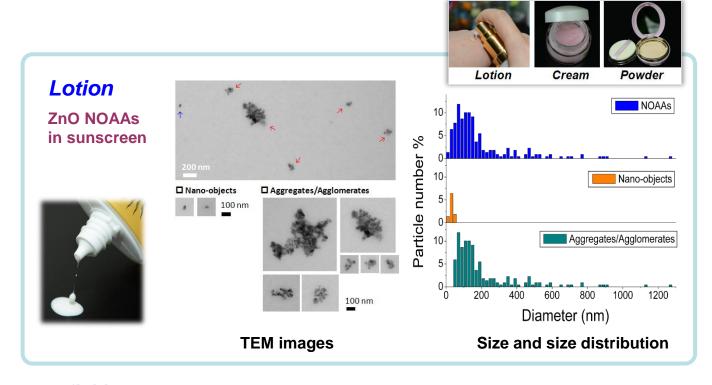


None of the second of the seco

NOAAs of ZnO NPs in sunscreen

- K-kit can be used for characterizing NOAAs of cosmetics in final product forms, including lotion, cream, and powder, to assess the safety risks of nanomaterials in cosmetic ingredients.
- ◆ International Cooperation on Cosmetic Regulation Report (ICCR) 2012 Characterization of Nanomaterials II – Insolubility, Biopersistence and Size Measurement in Complex Media.
- ◆ European Union (EU) Cosmetics Regulatory (EC) No. 1223/2009

 Mandatory labeling of Nanomaterials as ingredients in Cosmetics (effective 2013/07/11)
- United States Food and Drug Administration Guidance (US FDA) 2012
 Guidance of Industry Safety of Nanomaterials in Cosmetic Products



Definitive:

Direct observation in final product form or relevant media, minimizing artifacts.

Quantitative:

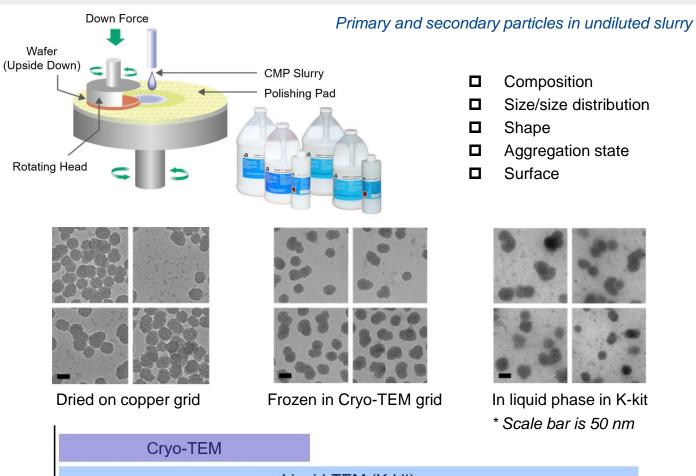
Image-based statistical analysis of aggregation and agglomeration as well as particle concentration.

Comprehensive:

All physicochemical characterization requirements can be addressed.

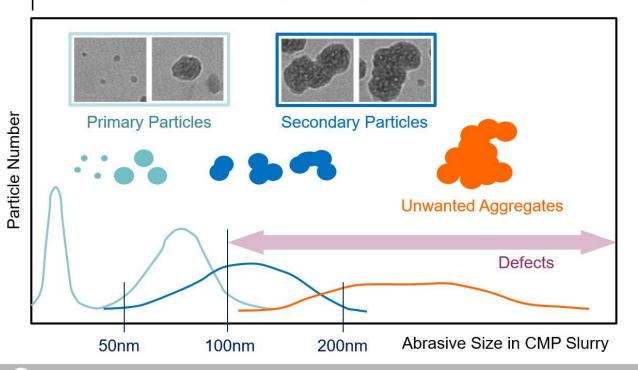


K-kit Application: Abrasives in CMP Slurry



Liquid-TEM (K-kit)

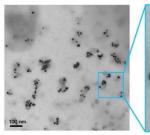
* Abrasive size scales covered by K-kit & cryo-TEM

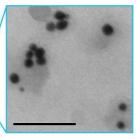




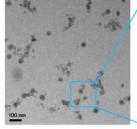
K-kit Application: Liquid-TEM Observation in Nanopharmaceuticals

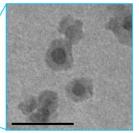
Applicable particle concentration for K-kit: 10¹¹~10¹⁴ particles/ml







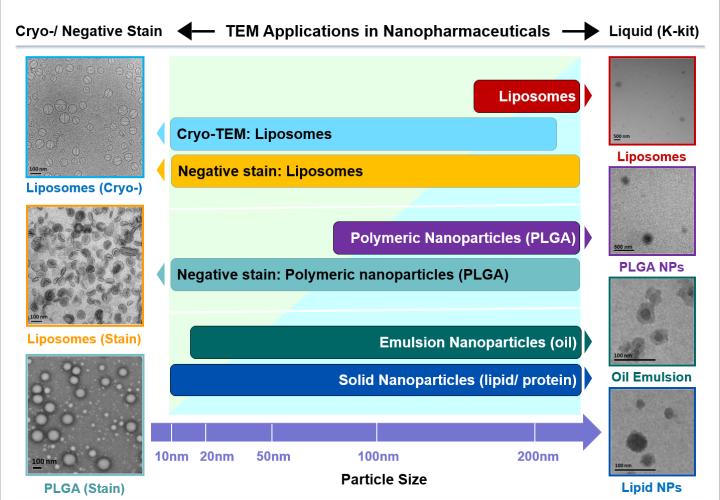




AuroVist® solution was directly loaded and sealed in a K-kit in liquid form.

Oil emulsion in water was loaded and sealed in a K-kit in liquid form.

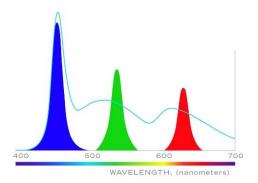
Brand Name of Pharmaceuticals	Doxil ® (1995 approved)	Abraxane ® (2005 approved)	Aurimune ® (Phase II)	Resovist ®	Rexin-G ® (Phase II)
Particle Size	80-100 nm	~ 130 nm	~ 27 nm (AuNPs core), ~ 30-40 nm as hydrated	~ 45-60 nm (Hydradynamic diameter)	~ 100 nm
Particle Concentrations	1.0 x 10 ¹⁴ liposome /ml	4.3 x 10 ¹³ albumin particles /ml	≤ 1.7 x 10 ¹² gold particles /ml	1 x 10 ¹⁴ particles /ml	1-4 x10 ¹¹ cfu





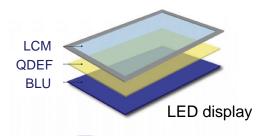
K-kit Application: Quantum Dots in Solution

Quantum dots will enable a market for devices and components worth over \$11bn by 2026.

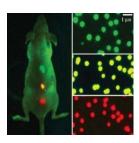




QD inks for printed electronics







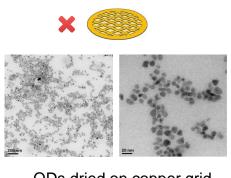
QD imaging diagnosis

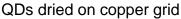


QD: Quantum dots BLU: Backlight Unit

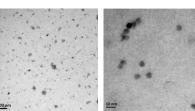
LCM: Liquid Crystal Module

QDEF: Quantum-dot Enhancement Film

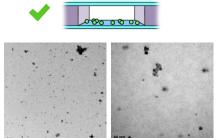






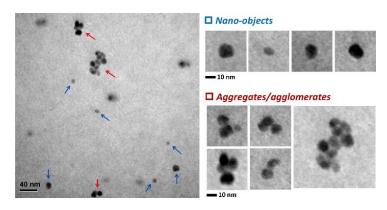


Wet mode of K-kit

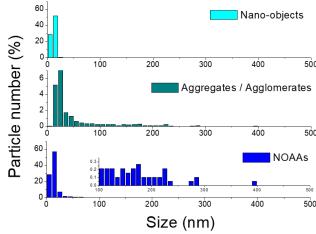


Thin Layer mode of K-kit

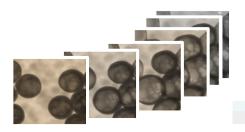
☐ Size and Size Distribution of QDs in Chloroform (Thin Layer Mode of K-kit)



- Sample solution was directly loaded into K-kit
- Nano-objects = Primary particle
- Aggregates/agglomerates = Secondary particle



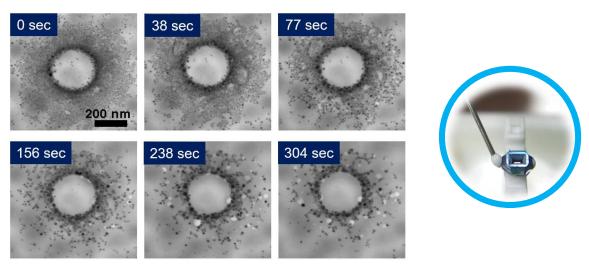
K-kit Application: In-situ Dynamic Observation





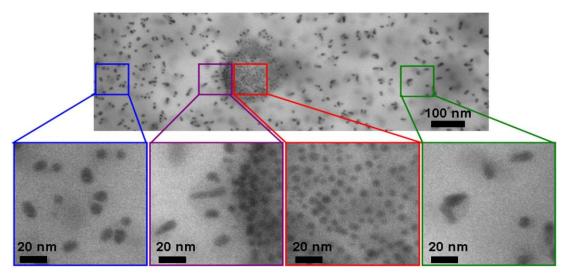
The dynamic changes of reaction processes in liquid can be observed and studied by K-kit.

■ Dynamic observation of polystyrene beads in PBS buffer (sodium ion)



The reduction process of the sodium ions, which induced from the TEM electron energy, in PBS buffer around a polystyrene bead could be observed with prolonged observation time.

☐ Gold metal growth in water with and without polystyrene beads

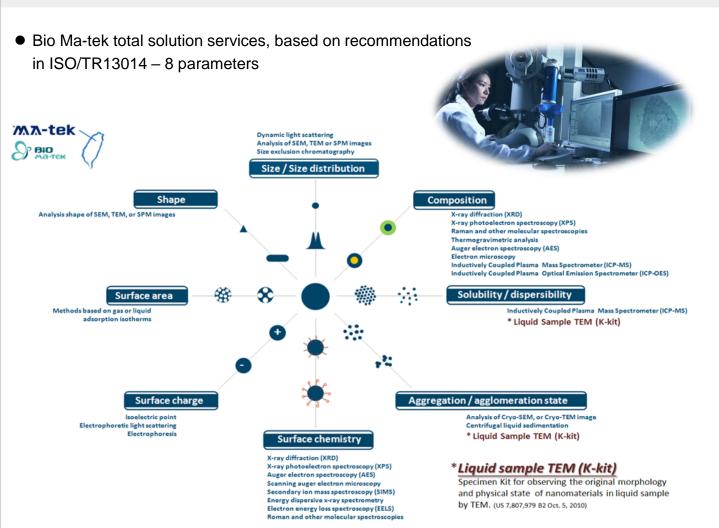


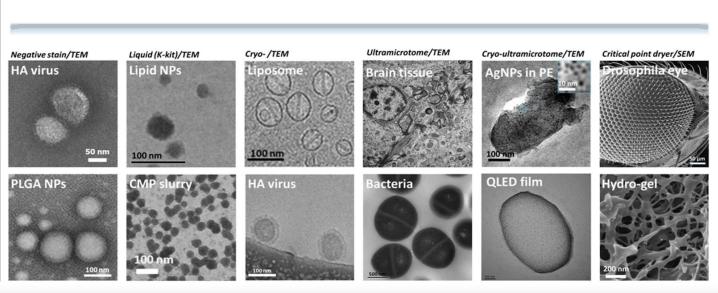
The metal growth of Au ions in water could be observed dynamically at the same time in the areas far away and nearby a polystyrene bead. (As shown in the image, there's a polystyrene bead at the center, with a lot of relatively small Au particles surrounded)



Not Only K-kit

Bio MA-tek provides comprehensive services in bio-EM and physicochemical analysis





Our Services







Bio MA-tek provides analytical services to the following industries:

- Bio-technology
- Pharmaceutical (nano-formulation, nano-drug, etc.)
- · Vaccine (vaccine formulation, adjuvant, etc.)
- Medical devices (dialysis, dressing, etc.)
- · Cosmetics (powder, cream, lotion, mask, etc.)
- Foods (additives, packaging materials, etc.)
- Academic & research organizations
- Electronics industry (Semiconductors, TFT-LCD, LED, PCBs, ...)
- Others

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Service Items

Physico-Chemical Characterization

- Size/Size Distribution: DLS
- Surface Charge: Zeta potential
- Composition and Impurity: TGA, DSC, FT-IR, XRD, UPLC, ICP-MS, LC/MS/MS (QQQ)
- Surface Chemistry: XPS
- Surface Area: BET

Electron Microscopy Analysis

- Negative Stain
- Resin Embedding
- Ultramicrotome
- Cryo-ultramicrotome
- Critical Point Drier (CPD)
- Cryo-transfer System
- Liquid Sample Preparation
- TEM/ EDX
- SEM/ EDX



Bio Materials Analysis Technology Inc.

Established on March 31 2014, Bio Materials Analysis Technology Inc. (Bio MA-tek) and its sole investor, MA-tek, serve as the best R&D partners of high-tech industry. To address the demanding needs for physical and chemical characterization of nanomaterials in bio-medical industry. Bio MA-tek has introduced an array of bio-EM sample preparation and image analysis services as well as a comprehensive list of analytical services per the recommendations of ISO/TR13014.



Vision:

To become a leading brand in bio-medical materials analysis

Business model:

Focus on core technology, leverage external resources, deliver proficient and adequate services

Positioning:

Solution provider of nano-biomaterials characterization and analysis

Service scope:

Characterize and analyze nanobiomaterials in foods, cosmetics, medical materials, drugs, vaccines, biological tissue, etc. Provide proficient and adequate sample preparation, analysis, consultation, and contracted services.



The Best R&D Partner!





Bio MA-Tek, the Best R&D Partner for Your Success!













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