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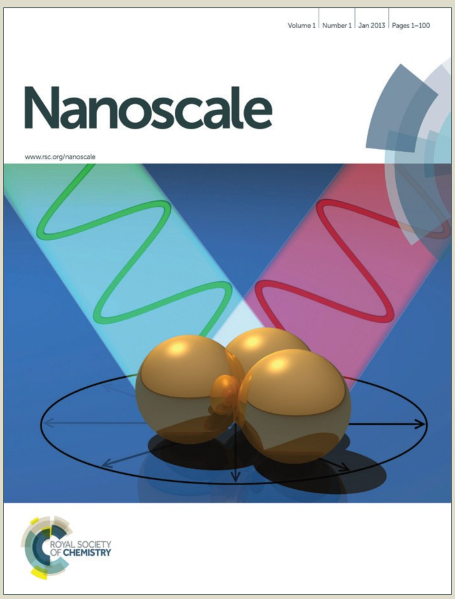

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REVIEW

Recent developments of *in situ* wet cell technology for transmission electron microscopies

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The *in situ* wet cells for transmission electron microscopy (TEM) and scanning transmission electron microscopy (STEM) allow studying structures and processes in liquid environment with high temporal and spatial resolutions, so that have been attracting more and more research interests in many fields. In this review, we highlight the structural and functional developments of the wet cells for TEM and STEM. One of the key features of the wet cells is the sealing technique to isolate the liquid sample from the TEM/STEM vacuum environments, thus the existing *in situ* wet cells are grouped by different sealing methods. The advantages and shortcomings of each type of the *in situ* wet cells are discussed, the functional developments of different wet cells are presented, and the future trends of the wet cell technology are addressed. It is revealed that the *in situ* wet cell TEM/STEM technology will make even more impact in frontier nanoscale research fields in the future.

Introduction

Transmission electron microscopy (TEM) is widely used in modern scientific research due to its high image resolution and the multiple characterization capabilities.[1-4] The vacuum system is one of the key components of TEM, which is extremely important for reducing the electron beam scattering and for protecting the electron source. To prevent damaging the vacuum environment in TEM by the evaporated gases from wet/liquid samples, traditional TEM is mostly used for solid and dry samples. However, there are enormous needs to study structures in liquid, such as in physics, chemistry, nano materials, and biological sciences. Cryogenic technology has been used to freeze water in wet samples in order to analyze them in TEM, however, the frozen samples are no longer in their original form and morphology, and dynamic behaviours in liquids cannot be observed by the cryogenic technology.[5-7] Environmental TEM has created a low pressure vapour space around the sample, while keeps the rest of the instrumental volume in high vacuum, by using differential pumping technology, allowing partially hydrated sample to be analyzed, but still unable to handle most pristine wet samples due to the pressure limitation.[8-10]

The recent developments of *in situ* wet cell (also called liquid cell, etc.) TEM technologies provide unprecedented capabilities to study samples in liquid environment by TEM, and opened new ways to study basic science, nano science, new energy sources, life science etc. in liquid.[11-15] Publications in the field have increased rapidly in the last several years.

So far, wet cell TEM technologies have been used in various fields, including *in situ* chemical sciences and material structural evolutions[16,17], material science,[18] liquid/solid interface,[19, 20] metal nanoparticle growth,[21] and biological science observations,[18, 22] etc. Issues on technical development,[18] design and fabrication of *in situ* fluid cell,[23, 24] coupling *in situ* wet cell with electron and photon beams in TEM,[22] how to reduce electron beam induced artifacts during *in situ* wet cell TEM analysis[25] and factors influencing quantitative analysis using *in situ* wet cell TEMs[26] have also been considered. Besides Niels de Jonge et al.'s review on the brief history of technical development for wet sample characterization with electron microscopies (EMs) (including environmental EMs, and *in situ* wet cells for SEM, TEM and scanning transmission electron microscopy (STEM)),[18] a bunch of other review papers have been published in the *in situ* wet cell EM field. The applications in life sciences and materials sciences are addressed, and the influence of the liquid layer thickness on the image resolution is discussed by de Jonge et al.[18] Zaera reviewed liquid/solid interface characterizations with different technologies, with one section about *in situ* EMs.[20] Tao et al. reviewed *in situ* chemistry and material structure studies in reactive environments using EM and non-EM technologies.[27] Liao et al. reviewed the observation of metal nanoparticle growth process with *in situ* wet cell TEMs.[21] Yu et al. reviewed liquid sample imaging using microfluidic cells with both EM and non-EM technologies.[24] Li et al reviewed design, fabrication, and applications of *in situ* fluid cell TEM developments, in which the technical development is introduced briefly.[23] Evans et al reviewed experimental parameters necessary for biological reaction observations coupling *in situ* wet cell and dynamic TEM (DTEM).[22] Woehl et al. reviewed how to reduce electron beam induced artifacts during *in situ* wet cell TEM analysis.[25] Abellan et al reviewed factors influencing quantitative analysis using *in situ* wet cell TEMs.[26] Wu et al. reviewed the electrochemical investigation of lithium-ion battery in the sealed wet cell.[28] There is also a review book on *in situ* EMs, unfortunately, due to the very broad topic range, there are not much discussions on *in situ* wet cell EMs in it.[29] Overall, these review papers have shed a lot of light on the *in situ* EM research on wet samples, however, due to the rapid development of the technique, there is still a need to systematically address the technological development of the *in situ* wet cells for TEMs. This review is aimed to fill the gaps in the literature to review and address recent technological development of *in situ* wet cells for TEMs.

In situ TEM technologies for liquid / wet samples

The technical breakthrough

Although the origins of *in situ* wet cell EM technologies can be traced back to the beginning of the electron microscope invention, due to the technical challenges, the technique only became readily applicable and developed rapidly over the last decade. The major technical breakthrough was reported by F. M. Ross and her collaborators in 2003, in which microfabricated chips with silicon nitride (SN) windows were used to assemble wet cells for TEM analysis.[30] The SN window membranes are nanometers thin and robust, allowing electron beam penetration while sealing the liquid from the vacuum environment.

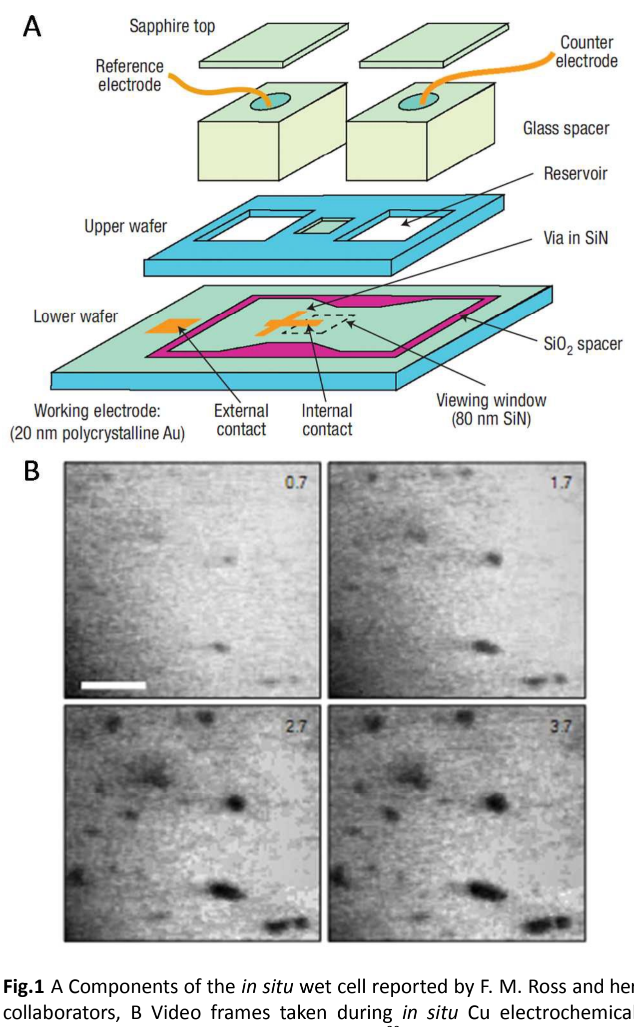

Fig.1 A Components of the *in situ* wet cell reported by F. M. Ross and her collaborators, B Video frames taken during *in situ* Cu electrochemical deposition, in which the scale bar is 500 nm.[30]

A schematic plot of the *in situ* wet cell set up is shown in Fig. 1 A, in which SN thin films were deposited on the upper and lower wafers, and free standing SN viewing windows were made at the center of the wafers by etching away the local wafer substrates using mature lithographic methods. A SiO_2 spacer layer may be patterned on a wafer to control the liquid layer thickness, and epoxy is used to glue and seal the cell parts.

Using this *in situ* cell, real time study of Cu nanoclusters in liquid with 30 images per second time resolution and 5 nm spatial resolution have been achieved.

Besides simple observation of liquid samples, it is further demonstrated that the wet cells can be made multifunctional by incorporating patterned metal electrodes to do electrochemical researches. Fig. 1B shows the video frames taken during *in situ* Cu electrochemical deposition, showing the Cu nanoparticle growth procedure.

Major technical trends of *in situ* wet cell TEMs.

Following the above breakthrough, there are many new technical variations developed for the *in situ* wet cell TEM research. Besides the F. M. Ross approach (noted here as advanced SN window chip approach), the second major trend is using polymer O-rings instead of epoxies to seal the wet cell, which greatly improved the easiness in cell fabrication and assembly. The original design can be seen in Fig. 2A.[31] The design utilizes SN window wafer grids readily available in the market, instead of homemade ones. A custom-built metal holder was used to hold the grids, and several O-rings were used to seal the liquid from the vacuum in the TEM chamber. The *in situ* cell can be assembled in 10-15 min and can be attached to the TEM sample stage with just one clip. A JEOL 2010 LaB_6 TEM system was used to test the design, and different samples such as Al_2O_3 nanoparticles in DI water and multi-walled carbon nanotubes in DI water have been imaged, demonstrating the versatility and reliability of the technique.[31]

The image resolution in the above trends is limited by the thickness of the SN window and liquid layer, mainly due to electron scattering.[32] Another major trend is using low vapor pressure liquids such as ionic liquids (ILs),[33] and study the liquid-solid systems in TEM.[34] The ILs will not evaporate to damage the vacuum system, and thus no sealing cell or differential pumping is needed to perform the research.[12] Fig. 2B is a schematic of the experimental set up. A microdrop of ionic liquid electrolyte (ILE) is put in contact with a bulk $LiCoO_2$ cathode, a single SnO_2 nanowire anode is manipulated and inserted into the ILE. By applying biases to the electrodes, the SnO_2 lithiation processes were observed *in situ* with high-resolution TEM (HRTEM). Besides the observation of an electrode, reaction materials have been dissolved into an ionic liquid to observe nanocrystal formation mechanisms in the solution.[35] Several follow up works are also reported in the literature.[33-37].

A fourth trend is to enclose tiny amount of liquid in atomic thin membrane cells, thus minimizing the window and liquid layer thickness effect. Yuk et al. reported transferring graphene onto gold mesh TEM grids, pipetting liquid solution between two graphene coated grids facing each other to form the *in situ* wet cell.[38] Upon wetting the system, the capillary force deforms the graphene membranes and allows one membrane to detach from its associated grid, forming a graphene enclosed liquid pocket attached to only one TEM grid.[39] The van der Waals force between the graphene sheets is strong enough to seal the liquid from leaking into the vacuum. Atomic resolution images have been obtained during *in situ* Pt nanocrystal growth experiment using an aberration-corrected TEM. Fig. 2C is a schematic illustration of the graphene-sealed liquid cell (GLC), and Fig. 2D shows atomic resolution images demonstrating Pt nanocrystal growth via coalescence and crystal-structure evolution procedure.Besides particles of a single element, the growth of Pt-Pd composite particles in GLC also were reported.[40] The 3D-motions of a single DNA molecules in the liquid have been directly observed in the GLC.[41] Meanwhile, Yuk et al. revealed the anisotropic lithiation on the silicon particle surfaces with GLC,[42] in which the dynamic process of lithium diffusion in silicon was recorded in real time. This makes it possible to prepare "nano-battery" in GLC to observe electrochemical process in higher contrast and resolution. It might become a hot research spot to select appropriate atomic thin sheet materials to replace the SiN window based wet cells in the *in situ* wet cell TEM field.

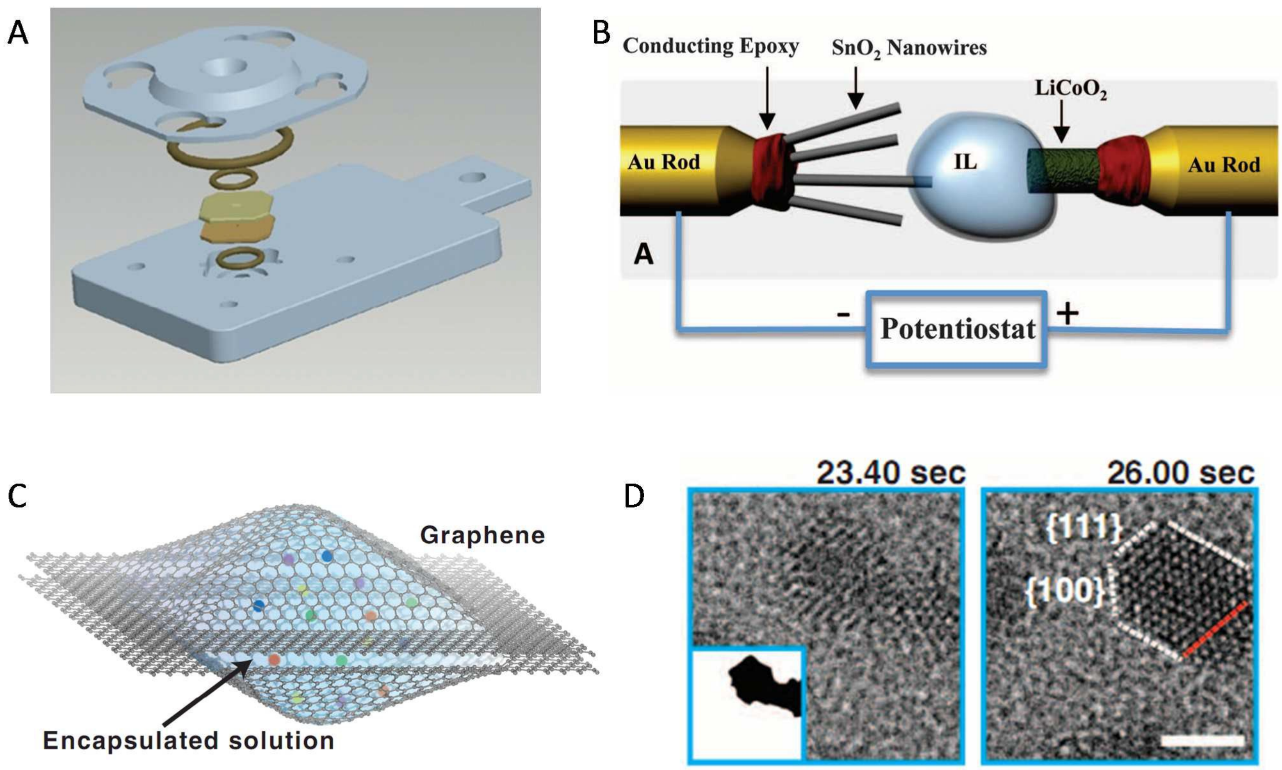

Fig. 2 A O-ring sealed *in situ* wet cell design,[31] B Sealless *in situ* liquid TEM set up utilizing low vapor ionic liquids,[12] C Illustration of *in situ* liquid cell formed by atomic thin graphene membranes, D Atomic resolution images obtained with C showing Pt nanocrystal growth procedure.[38]

Although the latter two trends have been really successful demonstrating high resolution imaging, they both face strong limitations. For instance, the operation of the ILs based set up is relatively challenging, and moreover, the ILs can't be used as solvent or interacting agents for broad materials. Thus, its application scope appears to be relatively narrow.

The GLC approach, on the other hand, needs to avoid electron knock off effect on the carbon atoms of the graphene membranes, thus requires a low operation voltage of 80 kV, which normally limits high resolution imaging for most of the materials and is not applicable for many TEM users. Also, the graphene membranes are highly conductive and atomically thin, there is no simple way to pattern electrodes in such a cell for electrochemical investigation.

Due to these limitations, the most *in situ* wet cell TEM works are currently performed by the first two approach trends, and we will mainly address these two trends in more details in the following sessions.

Technological development of the SN sealed *in situ* wet cells

Advanced SN window chip approach

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REVIEW

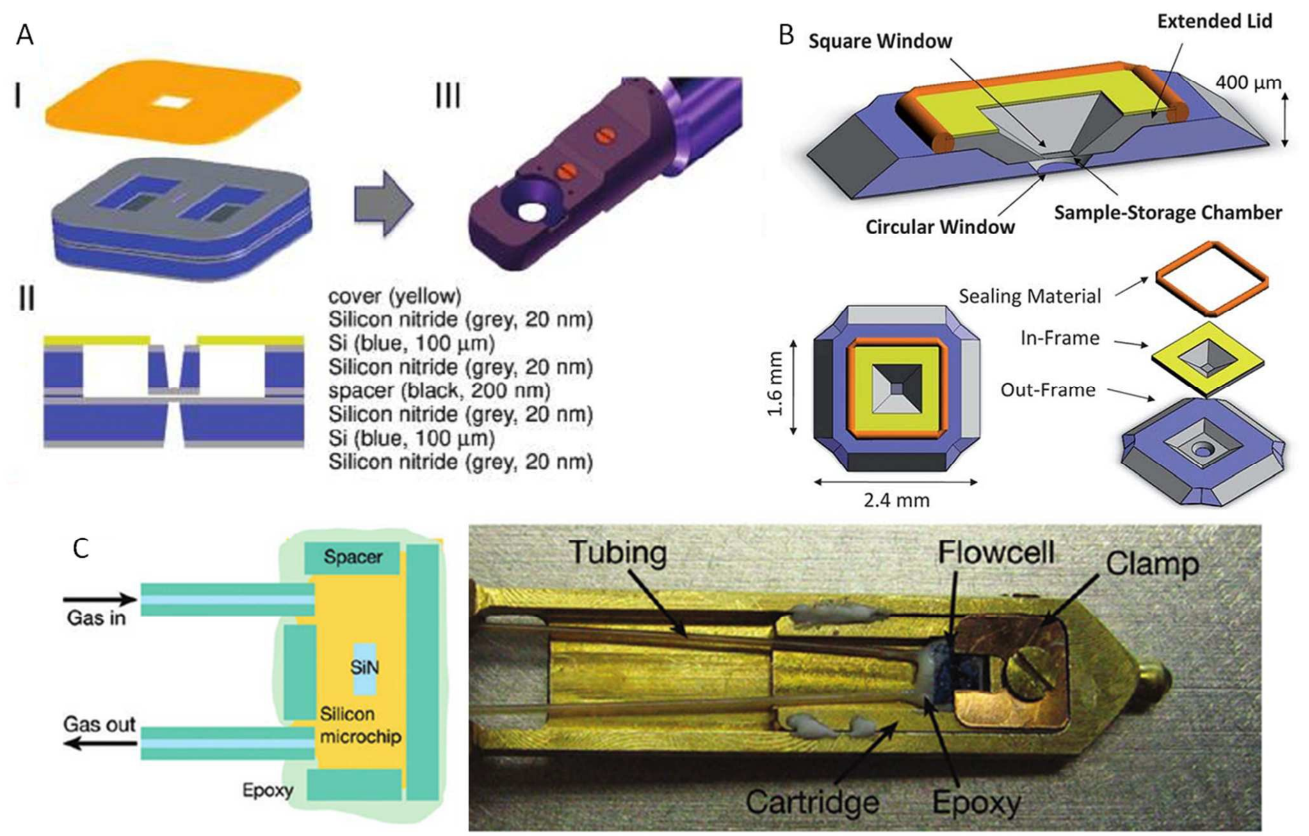

Fig. 3 A The schematic of the mini wet cell that can be mounted on a commercial TEM holder,[47] B Self-aligned wet cell that can be put into regular TEM stages,[49] C The *in situ* fluid cell set up with flow channels.[54]

Following the work of F. M. Ross and her collaborators,[30, 43] the *in situ* wet cell technology based on advanced SN window chip approach has made tremendous progress in the last decade.

Firstly, different *in situ* wet cell structural materials have been tested. Zheng et al utilized indium spacer instead of SiO_2 in the original design.[44-46] Indium, as a soft material, might reduce the tension between the SN window wafers during the cell assembly, and could also improve the sealing level of the *in situ* wet cell.

As micro chips can be scaled down in size easily, mini wet cells have been made to fit into a commercial TEM holder. Fig.3A shows such a design that can be mounted onto an in situ heating holder to perform *in situ* heating experiments. [47] Oscillatory growth behaviour of Bismuth nanoparticles has been observed at 180℃ and bismuth oxide hollow nanoparticle was observed to form through Kirkendall effect at up to 200℃.[47, 48]

Considering the difficulty in aligning the SN window wafers during cell assembly, Huang et al. developed a self-aligned wet cell.[49] As shown in Fig. 3B, one wafer is made into a protruded shape and the other wafer has a socket, realized by microfabrication technology. These two wafers are easily set into position, and after being sealed, the cell can also be of the size of a regular TEM grid, thus can be put into a standard TEM sample holder.

Another development is introducing fluid flow function to the *in situ* wet cells.[32, 50-52] Neils de Jonge et al. made flow channels to the *in situ* wet cell and connected flow tubes to the cell from the TEM stage by glue, allowing not only liquid,[53] but also gas flow into the *in situ* sample cell.[54] Fig. 3C shows the schematic design and the actual set up on a TEM stage.

A lot of work may further be done with the wet cell put into multifunctional TEM stages/systems for advanced *in situ* analysis. Besides the aforementioned *in situ* heating TEM work, J. E. Evans et al also reported *in situ* laser induced growth and electron beam induced growth of lead sulfide nanoparticles using an *in situ* flow cell in a DTEM system.[55]

The wet cell with O-rings

Although the above advanced chip approach have been extremely successful, it still faces some technical difficulties: the epoxy sealing is still relatively technically difficult, non-accuracy of the epoxy amount might cause leaking or contamination to the viewing window region and from the technological view it is relatively challenging to reuse the wet cells. The O-ring sealed wet cell [31, 56] appears to be more advantageous with the above considerations, due to the easiness in assembly, less contamination concern to the viewing region and reusability of the cell body. Thus the o-ring trend is currently gaining strong momentum in the technical development and even commercialization.

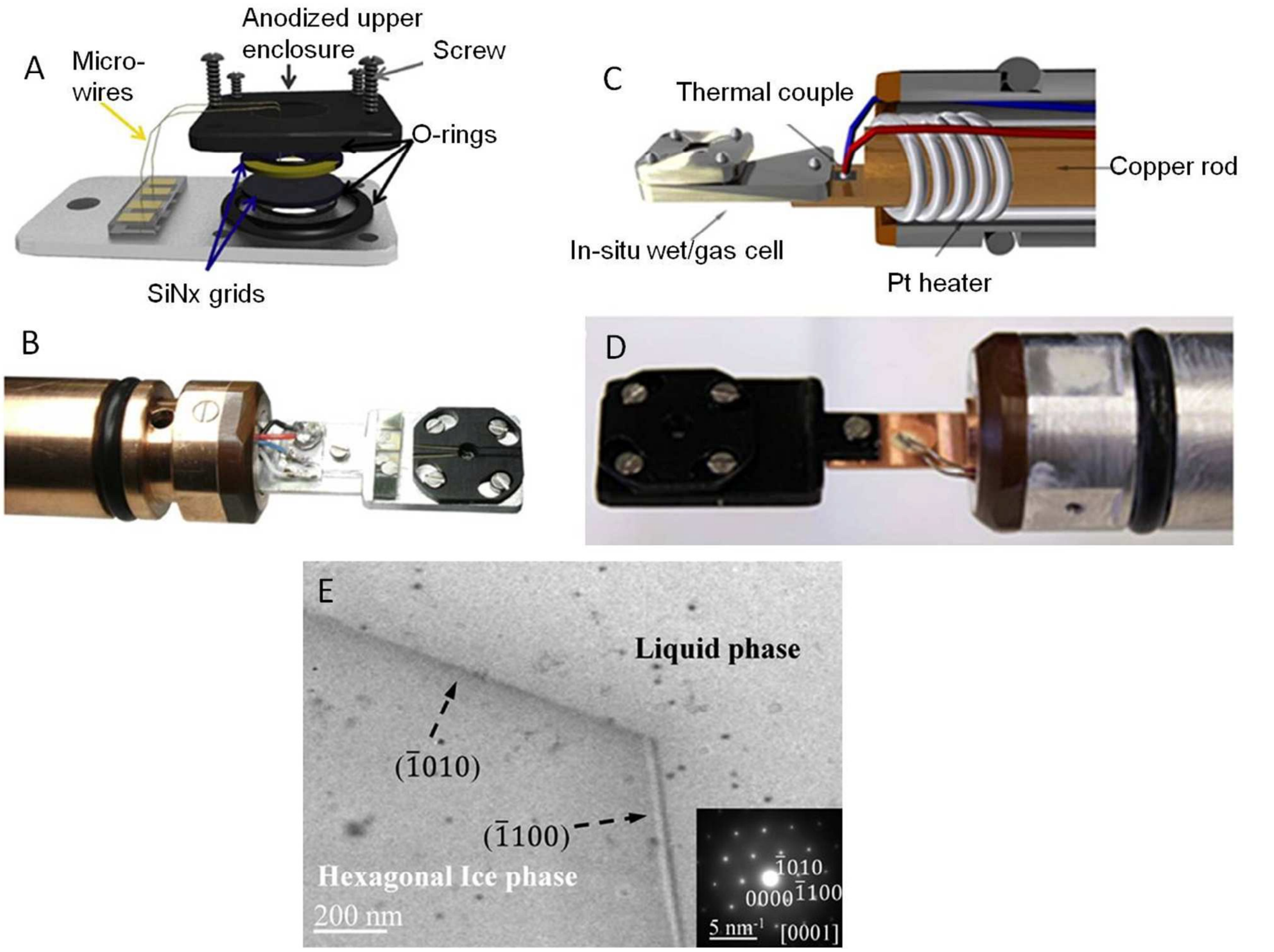

Fig. 4 A schematic illustration of the *in situ* electrochemical wet cell with O-rings, B Photograph of the electrochemical cell mounted on a TEM sample stage,[57] C Illustration of the *in situ* wet cell set up with heating and cooling functions, D Photograph of the wet cell mounted on the heating /cooling TEM holder, E The dynamic interactions of Au nanoparticles at the ice crystallization front observed using the set up in C and D.[59]

X. Chen, et al developed an electrochemical function to the o-ring sealed wet cell.[57, 58] As shown in Fig. 4A, the aluminum wet cell body was anodized to avoid shortage between the electrodes, a commercial SN grid of the wet cell was coated with metal electrodes, micro-wires were connected from the metal electrodes to the outside electrical equipments. Photograph of the electrochemical cell mounted on a TEM sample stage is shown in Fig. 4B.

Heating and cooling functions have also been added to the O-ring based wet cell design by S. J. Dillon's group.[59] As seen in Fig. 4C and D, a special TEM sample stage has been fabricated with a copper rod in the middle, which is thermally isolated from the TEM stage shell. One end of the copper rod is wrapped with Pt metal wires for sample heating, and the other side of the rod is attached to a cold reservoir outside the microscope through a high vacuum feed through (not shown). The temperature of the wet cell mounted on to the copper rod piece can be controlled by adjusting the heating/cooling conditions, and a thermal couple is fixed nearby to monitor the sample temperatures. The sample temperature can be as low as ~150 K using a cold reservoir filled with solid/liquid ethanol, or ~77 K with liquid nitrogen. As an application example, the nanoscale details of ice crystallization procedures have been recorded, and the dynamic interactions of Au nanoparticles at the crystallization front have been studied (Fig. 4E).

M. Gu et al demonstrated a wet cell fabricated with lithium and single Si nanowire electrodes. This set up overcomes the ILs electrolyte limitation by using the sealed cell approach, and can be used to study the lithiation/delithiation behaviour in real lithium ion batteries with commonly used electrolytes.[60] Such electrochemical wet cell has great application potential in new energy field allowing the reaction mechanism to be recorded directly with high spatial resolution.[61, 62]

Further developments include adding flow capability and commercializing the O-ring sealed *in situ* wet cell technology. N. de Jonge et al utilized a fluid STEM/TEM holder with 2 fluid lines from Hummingbird Scientific to observe whole cells in liquid with nanometer resolution.[53] The fluid lines were fed to plastic tubing from Upchurch Scientific. A syringe pump from Harvard Scientific, MA was used to drive the liquid flow through the *in situ* cell. The SN window wafers in the *in situ* cell were custom designed by Protochips Inc, NC, and were fabricated with 50 nm low stress SN thin film on 300 μm thick Si wafers. A polystyrene microsphere layer of 10 μm thickness was solution coated on the 4 corners of the chip to provide spacing for cells and liquid flow.

C. Mueller et al. also exploited a custom made nanofluidic cell set up with active feedback to stabilize the flow conditions and used to take nanometer resolution images for highly scattering metal particles and weakly scattering polymer particles side by side in the fluid.[63] Besides Hummingbird Scientific, *in situ* fluid TEM stages have also been developed by Protochips Inc.[50, 64] Fig. 5 shows the *in situ* liquid flow TEM set up and part of the components. This set up also consists of a liquid flow holder, with SN window wafer wet cell parts encapsulated inside the holder tip, a syringe pump and the plastic tubs.

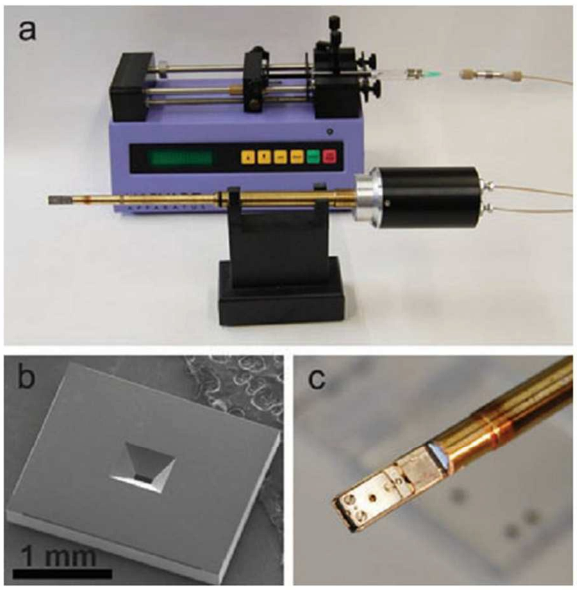

Fig.5 The Protochips Inc. prototype liquid flow TEM holder and its components: A The holder (foreground) and the syringe pump assembly (background) connected together with plastic tubs, B A microchip with a SiN window used in the *in situ* TEM stage, and C The tip of the liquid flow TEM holder with the wet cell microchips assembled and encapsulated within by a screw-secured plate.[50]

The *in situ* wet cell TEM products from Hummingbird and Protochips Inc. are now available in the market, with more detailed product information in the company web pages and advertisements. Protochips Inc. is selling Poseidon™ *in situ* liquid TEM systems with 2 or 3 fluid channels (for flow mixing), and the company produce the E-chips (Environmental chips) used in the *in situ* holder. The Hummingbird *in situ* liquid TEM holder not only can have 2 or 3 flow channels, but also may have electrochemical or heating capabilities. More new products are continually being developed by the companies. More and more people are using commercial *in situ* liquid TEM products and generating important scientific results.[65-69]

Discussions

Although the O-ring based *in situ* TEM cells and the commercial products are gaining momentum, it is by no means the ending of other types of the *in situ* liquid TEM technologies. Although the commercial products are selling more and more in the market, they are currently very expensive; on the other hand, the technology is still very young and new functions and new research topics are still coming out, that still can arise from home made set-ups. Each technology has its own advantages and shall continue to generate new exciting results. We believe the merit of a scientific report should be judged by the scientific content instead of which type of technology is used. Here we try to compare the advantages and disadvantages of each type of technology in Table 1.

It should also be noted that besides the graphene liquid cell approach, atomic resolution images can also be obtained with the other approaches under ideal experimental conditions.[66, 70]

Besides, many times, there are no clear boundaries between each type of technologies. For example, J. M. Gorgan et al. developed a "nanoaquarium", which used sophisticated chip design to couple electrical and liquid flow capability, while using O-rings to seal the *in situ* cell.[71, 72] As the technology advances, graphene windows may also be used in the advanced chip or O-ring based wet cells.

Other than those already mentioned above, here we would like to further address a little about the recent application results in the *in situ* liquid TEM field. Besides the earlier observations on such as the motion and coalescence of nanoparticles[11] and the whole cells and metal labelled whole cells[53], people have further used the technology to observe Ostwald ripening and anti-Ostwald ripening behaviors,[47] the nanoscale Kirkendall effect,[48] the formation of metal core-shell nanostructures,[17] the amorphous to ordered nanostructure transition,[73] the redox reactions with metal particles,[74] and performed advanced researches on lithium ion batteries,[28] etc. The dynamics of soft nanomaterials in water have been observed.[75] Protein complexes have been imaged in their native environment in whole cells,[76] and fine cellular structures and pili motions of E. coli cells have been imaged.[77]

Besides the structural development of the *in situ* wet cell set-ups, there is also the possibility to utilize the electron beam and other built in functions in TEM systems to perform multifunctional *in situ* wet cell TEM research beyond mere imaging. There are already many papers reporting using the electron beam to decompose precursor molecules in the liquid and synthesizing nano materials[25, 46] The electron beam has further been used to control and assemble metal nanoparticles.[45, 46, 78] Selected area diffraction (SAD) is also routinely used to characterize nano material structures in the wet cell, electron energy loss spectroscopy (EELS) and x-ray Energy-Dispersive Spectrometry (EDS) are used to characterize the nano material compositions.[73, 79-81] This topic is out of the scope of this paper. Coupling the multifunctions in the *in situ* wet cell set up design and the multifunctions of the TEM systems may generate more opportunities in the *in situ* liquid TEM research.

Table 1 A comparison of each type of the *in situ* wet cell TEM technology

Technology type	Assembling easiness	*In situ* cell reusableness	Sealing requirement	Sample requirement	Image resolution	TEM system requirement	Multifunctional development capability	Commercialization stage
Advanced chip	Relatively good	Hard to reuse	High	Low	Very good	Low	Good	N/A
O-ring sealed	Good	Reusable	High	Low	Very good	Low	Very good	Commercialized
Low vapor pressure liquid	Technically demanding	N/A	Low	High	Very good	Low	Relatively poor	N/A
Atomic thin membrane cell	Technically demanding	One time use	High	Low	Excellent	High	Relatively poor	N/A

Summary and outlook

There are a couple of ways to set up an *in situ* liquid TEM experiment, including the advanced chip approach, O-ring sealed approach, ion-liquid approach, and grapheme *in situ* cell approach, etc. These technological developments enabled many breakthrough research works in the nanoscale sciences. Moreover, many advanced functions can be coupled in to the experiment, including liquid flow, heating/cooling, electrochemistry, photon irradiation, and the intrinsic functions of the TEM system such as electron beam irradiation, SAD, EELS, EDS, etc. More new functions are being developed and will continue to sprout out in the future.

We envision as the technology is getting mature, the *in situ* liquid TEM product price will soon become affordable to the general TEM users, and the strong and multifunctional characterization powers will be beneficial to a broad range of researches including basic science, nano material fabrication, electrochemistry, new energy material research, and biological sciences.

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Notes and references

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1. C. Cong, K. Li, X. X. Zhang and T. Yu, *Scientific Reports*, 2013, **3**, 1195.
2. D. Golberg, P. M. Costa, M. S. Wang, X. Wei, D. M. Tang, Z. Xu, Y. Huang, U. K. Gautam, B. Liu, H. Zeng, N. Kawamoto, C. Zhi, M. Mitome and Y. Bando, *Advanced Materials*, 2012, **24**, 177-194.
3. A. L. Robinson, *Science*, 1985, **230**, 304-306.
4. G. Van Tendeloo, S. Bals, S. Van Aert, J. Verbeeck and D. Van Dyck, *Advanced Materials*, 2012, **24**, 5655-5675.
5. H. Cui, T. K. Hodgdon, E. W. Kaler, L. Abezgauz, D. Danino, M. Lubovsky, Y. Talmon and D. J. Pochan, *Soft Matter*, 2007, **3**, 945.
6. E. M. Pouget, P. H. H. Bomans, J.A. C. M. Goos, P. M. Frederik, G. de With and N. A. J. M. Sommerdijk, *Science*, 2009, **323**, 1455-1458.
7. C. J. Newcomb, T. J. Moyer, S. S. Lee and S. I. Stupp, *Current Opinion in Colloid & Interface Science*, 2012, **17**, 350-359.
8. P. L. Gai, *Microscopy and Microanalysis*, 2002, **8**, 21.
9. A. Bogner, P. H. Jouneau, G. Thollet, D. Basset and C. Gauthier, *Micron*, 2007, **38**, 390-401.
10. R. Sharma, *Journal of Materials Research*, 2011, **20**, 1695-1707.
11. H. Zheng, R. K. Smith, Y. W. Jun, C. Kisielowski, U. Dahmen and A. P. Alivisatos, *Science*, 2009, **324**, 1309-1312.
12. J. Y. Huang, L. Zhong, C. M. Wang, J. P. Sullivan, W. Xu, L. Q. Zhang, S. X. Mao, N. S. Hudak, X. H. Liu, A. Subramanian, H. Fan, L. Qi, A. Kushima and J. Li, *Science*, 2010, **330**, 1515-1520.
13. E. R. White, M. Mecklenburg, S. B. Singer, S. Aloni and B. C. Regan, *Applied Physics Express*, 2011, **4**, 055201.
14. E. A. Ring, D. B. Peckys, M. J. Dukes, J. P. Baudoin and N. de Jonge, *Journal of Microscopy*, 2011, **243**, 273-283.
15. U. M. Mirsaidov, H. M. Zheng, D. Bhattacharya, Y. Casana and P. Matsudaira, *Proceedings of the National Academy of Sciences of the United States of America*, 2012, **109**, 7187-7190.

16. E. Sutter, K. Jungjohann, S. Bliznakov, A. Courty, E. Maisonhaute, S. Tenney and P. Sutter, *Nature Communications*, 2014, **5**, 4946.
17. K. L. Jungjohann, S. Bliznakov, P. W. Sutter, E. A. Stach and E. A. Sutter, *Nano Letters*, 2013, **13**, 2964-2970.
18. N. de Jonge and F. M. Ross, *Nature nanotechnology*, 2011, **6**, 695-704.
19. S. W. Chee, D. J. Duquette, F. M. Ross and R. Hull, *Microscopy and Microanalysis*, 2014, **20**, 462-468.
20. F. Zaera, *Chemical Review.*, 2012, **112**, 2920-2986.
21. H. G. Liao, K. Niu and H. Zheng, *Chemical communications*, 2013, **49**, 11720-11727.
22. J. E. Evans and N. D. Browning, *Microscopy*, 2013, **62**, 147-156.
23. D. S. Li, M. H. Nielsen and J. J. De Yoreo, in *Methods in Enzymology*, ed. J. D. Y. James, Academic Press2013, Volume 532, pp. 147-164.
24. X.-Y. Yu, B. Liu and L. Yang, *Microfluid Nanofluid*, 2013, **15**, 725–744.
25. T. J. Woehl, K. L. Jungjohann, J. E. Evans, I. Arslan, W. D. Ristenpart and N. D. Browning, *Ultramicroscopy*, 2013, **127**, 53-63.
26. T. J. W. P. Abellan, L. R. Parent, N. D. Browning, J. E. Evansc, I. Arsland *Chemical Communications,*, 2014, **50**, 4873-4880.
27. F. F. Tao and M. Salmeron, *Science*, 2011, **331**, 171-174.
28. F. Wu and N. Yao, *Nano Energy*, 2015, **11**, 196-210.
29. F. M. Ross, *In-situ Electron Microscopy: Applications in Physics, Chemistry and Materials Science,* ed. G. Dehm, J. M. Howe and J. Zweck, Wiley-VCH Verlag GmbH & Co. KGaA, 2012, Volume7, pp.171-188.
30. M. Williamson, R. Tromp, P. Vereecken, R. Hull and F. Ross, *Nature Materials*, 2003, **2**, 532-536.
31. R. Franks, S. Morefield, J. Wen, D. Liao, J. Alvarado, M. Strano and C. Marsh, *Journal of Nanoscience and Nanotechnology*, 2008, **8**, 4404-4407.
32. N. de Jonge, N. Poirier-Demers, H. Demers, D. B. Peckys and D. Drouin, *Ultramicroscopy*, 2010, **110**, 1114-1119.
33. S. Chen, K. Kobayashi, R. Kitaura, Y. Miyata and H. Shinohara, *ACS NANO*, 2011, **5**.
34. C. M. Wang, W. Xu, J. Liu, J. G. Zhang, L. V. Saraf, B. W. Arey, D. Choi, Z. G. Yang, J. Xiao, S. Thevuthasan and D. R. Baer, *Nano Letters*, 2011, **11**, 1874-1880.
35. H. G. Liao, H. B. Shao, C. Wang, Y. Lin, Y. X. Jiang and J. Sun, *Materials Letters*, 2014, **116**, 299-303.
36. D. Chen, S. Indris, M. Schulz, B. Gamer and R. Mönig, *Journal of Power Sources*, 2011, **196**, 6382-6387.
37. W. J. Pemberton, J. E. Droessler, J. M. Kinyanjui, K. R. Czerwinski and D. W. Hatchett, *Electrochimica Acta*, 2013, **93**, 264-271.
38. J. M. Yuk, J. Park, P. Ercius, K. Kim, D. J. Hellebusch, M. F. Crommie, J. Y. Lee, A. Zettl and A. P. Alivisatos, *Science*, 2012, **336**, 61-64.
39. C. Colliex, *Science*, 2012, **336**, 44-45.
40. A. D. Clercq, W. Dachraoui, O. Margeat, K. Pelzer, C. R. Henry and S. Giorgio, *The Journal of Physical Chemistry Letters*, 2014, **5**, 2126-2130.
41. Q. Chen, J. M. Smith, J. Park, K. Kim, D. Ho, H. I. Rasool, A. Zettl and A. P. Alivisatos, *Nano Letters*, 2013, **13**, 4556-4561.
42. J. M. Yuk, H. K. Seo, J.W. Choi and J. Y. Lee, *ACS NANO*, 2014, **8**, 7478-7485.
43. A. Radisic, P. M. Vereecken, P. C. Searson and F. M. Ross, *Surface Science*, 2006, **600**, 1817-1826.
44. J. Park, H. Zheng, G. H. Lee, P. L. Geissler, E. Rabani and A. P. Alivisatos, *American Chemical Society Nano*, 2012, **6**, 2078-2085.
45. H. Zheng, U. M. Mirsaidov, L. W. Wang and P. Matsudaira, *Nano Letters*, 2012, **12**, 5644-5648.
46. H. Zheng, *Nanoscale*, 2013, **5**, 4070-4078.
47. H. L. Xin and H. Zheng, *Nano Letters*, 2012, **12**, 1470-1474.
48. K.-Y. Niu, J. Park, H. M. Zheng and A. Paul Alivisatos, *Nano Letters*, 2013, **13(11)**, 5715–5719.
49. T. W. Huang, S. Y. Liu, Y. J. Chuang, H. Y. Hsieh, C. Y. Tsai, Y. T. Huang, U. Mirsaidov, P. Matsudaira, F. G. Tseng, C. S.

Chang and F. R. Chen, *Lab on a Chip*, 2012, **12**, 340-347.
50. K. L. Klein, I. M. Anderson and N. de Jonge, *Journal of Microscopy*, 2011, **242**, 117-123.
51. J. E. Evans, K. L. Jungjohann, P. C. Wong, P. L. Chiu, G. H. Dutrow, I. Arslan and N. D. Browning, *Micron*, 2012, **43**, 1085-1090.
52. Y. Inayoshi, H. Minoda, Y. Arai and K. Nagayama, *Micron*, 2012, **43**, 1091-1098.53. N. de Jonge, D. B. Peckys, G. J. Kremers and D. W. Piston, *Proceedings of the National Academy of Sciences of the United States of America*, 2009, **106**, 2159-2164.
54. N. de Jonge, W. C. Bigelow and G. M. Veith, *Nano Letters*, 2010, **10**, 1028-1031.
55. J. E. Evans, K. L. Jungjohann, N. D. Browning and I. Arslan, *Nano Letters*, 2011, **11**, 2809-2813.
56. X. C. Y. Liu, K. Noh and S. Dillon, *Nanotechnology*, 2012, **23**, 385302.
57. X. Chen, K. W. Noh, J. G. Wen and S. J. Dillon, *Acta Materialia*, 2012, **60**, 192-198.
58. X. Chen, L.H.Zhou,P. Wang, C. J. Zhao and X. L. Miao, *Chinese Journal of Chemistry*, 2012, **30(12)**, 2839-2843.
59. K. Tai, Y. Liu and S. J. Dillon, *Microscopy and Microanalysis*, 2014, **20**, 330-337.
60. M. Gu, L. R. Parent, B. L. Mehdi, R. R. Unocic, M. T. McDowell, R. L. Sacci, W. Xu, J. G. Connell, P. Xu, P. Abellan, X. Chen, Y. Zhang, D. E. Perea, J. E. Evans, L. J. Lauhon, J. G. Zhang, J. Liu, N. D. Browning, Y. Cui, I. Arslan and C. M. Wang, *Nano Letters*, 2013, **13**, 6106-6112.
61. R. R. Unocic, R. L. Sacci, G. M. Brown, G. M. Veith, N. J. Dudney, K. L. More, F. S. Walden, 2nd, D. S. Gardiner, J. Damiano and D. P. Nackashi, *Microscopy and Microanalysis*, 2014, **20**, 452-461.
62. B. L. Mehdi, M. Gu, L. R. Parent, W. Xu, E. N. Nasybulin, X. Chen, R. R. Unocic, P. Xu, D. A. Welch, P. Abellan, J. G. Zhang, J. Liu, C. M. Wang, I. Arslan, J. Evans and N. D. Browning, *Microscopy and Microanalysis*, 2014, **20**, 484-492.
63. C. Mueller, M. Harb, J. R. Dwyer and R. J. D. Miller, *The Journal of Physical Chemistry Letters*, 2013, **4**, 2339-2347.
64. Protochips Inc, Poseidon510 in-situ electrochemistry flow cell holder for Electron Microscopy.Retrieved Dec.6,2014 from :*http://www.protochips.com/products/poseidon-500.html* .
65. E. R. White, M. Mecklenburg, B. Shevitski, S. B. Singer and B. C. Regan, *Langmuir: the ACS Journal of Surfaces and Colloids*, 2012, **28**, 3695-3698.
66. K. L. Jungjohann, J. E. Evans, J. A. Aguiar, I. Arslan and N. D. Browning, *Microscopy and Microanalysis*, 2012, **18**, 621-627.
67. T. J. Woehl, J. E. Evans, I. Arslan, W. D. Ristenpart and N. D. Browning, *ACS Nano*, 2012, **6**, 8599-8610.
68. D. Li, M. H. Nielsen, J. R. Lee, C. Frandsen, J. F. Banfield and J. J. De Yoreo, *Science*, 2012, **336**, 1014-1018.
69. Y. Liu, X. M. Lin, Y. Sun and T. Rajh, *Journal of the American Chemical Society*, 2013, **135**, 3764-3767.
70. R. Ramachandra, H. Demers and N. de Jonge, *Applied Physics Letters*, 2011, **98**, 93109.
71. J. M. Grogan and H. H. Bau, *Journal of Microelectromechanical Systems*, 2010, **19**, 885-894.
72. J. M. Grogan, L. Rotkina and H. H. Bau, *Physical Review E*, 2011, **83**.
73. M. H. Nielsen, S. Aloni and J. J. De Yoreo, *Science*, 2014, **345**, 1158-1162.
74. E. A. Sutter and P. W. Sutter, *Journal of the American Chemical Society*, 2014, **136**, 16865-16870.
75 M. T. Proetto et al., *Journal of the American Chemical Society,* 2014, **136**, 1162-1165.
76 D. B. Peckys and N. de Jonge, *Microscopy and Microanalysis*, 2014, **20**, 346-365.
77 Y. B. Wang, X. Chen, H. L. Cao, X. D. Cao, C. Deng and P. Wang, *Journal of Analytical Methods in Chemistry*, 2015, in press.
78. V. P. Oleshko and J. M. Howe, in *Advances in Imaging and Electron Physics*, ed. W. H. Peter, Elsevier 2013, Volume 179, pp. 203-262.
79. R. M. Tromp, *Ultramicroscopy*, 2012, **120**, 73-77.
80. Y. Sun, *Materials Today*, 2012, **15**, 140-147.

81. N. J. Zaluzec, M. G. Burke, S. J. Haigh and M. A. Kulzick, *Microscopy and Microanalysis*, 2014, **20**, 323-329.