Preparation of TEM Specimens Having Specific Crystal Orientations Using FIB-SEM and EBSD ～Applications to Allende meteorite～

Misumi Kadoi[1], Masateru Shibata[1] and Ichiro Ohnishi[2]

[1] EP Business Unit, JEOL Ltd. [2] EM Business Unit, JEOL Ltd.

In TEM or STEM observations of micro-crystals or poly-crystals, we need to search for crystalline grains having the primary crystal orientations of the target rather than the random crystal orientations, and, if necessary, we also need to perform specimen tilting and crystal-orientation alignment. These procedures require a significant effort and long hours. To improve this situation, we used a FIB-SEM equipped with an EBSD detector to examine a method to select crystalline grains having specific orientations and then prepare TEM specimens of crystals having the target primary orientations. In our study, we applied the method to Allende meteorite and prepared TEM specimens of olivine grains having the orientations of [100], [010] and [001] from the fine-grained matrix in the meteorite. Our TEM results confirmed that the TEM specimens can be aligned to the target primary orientations with an accuracy of a tilt angle of 5 degrees using the TEM goniometer stage, and they were thin enough to produce good atomic-resolution STEM images. The results suggest that our method is very effective in preparing TEM specimens having specified crystal orientations of the target from not only the matrix of Allende meteorite but also from any specimens composed of various micro-crystals or poly-crystals.

Introduction

Recent development of the aberration correction technology has enabled us to observe atomic resolution TEM (Transmission Electron Microscope) or STEM (Scanning Transmission Electron Microscope) images for crystalline specimens. However, in order to observe the crystal structure at the atomic level, the electron beam has to be irradiated along the target zone axis of the crystal. If this condition is not satisfied, it is necessary to align the orientations by tilting the specimen using the TEM goniometer stage. In general, the powder or poly-crystal specimens such as ceramics, minerals, and so on, contain randomly oriented many crystalline grains and therefore, specimen tilting and orientation alignment are essential procedures, and require many efforts and long hours. Moreover, if the specimen tilt angle is large, the specimen appears thicker, resulting in degrading image resolution by increased chromatic aberration. To avoid these problems, it is useful to obtain the information on crystal orientations of the specimen before or during the preparation of TEM specimen. This orientation information facilitates preparation of TEM specimen having the primary crystal orientations, thus greatly reducing the efforts and hours for observation steps.

The FIB (Focused Ion Beam) technique irradiates and scans a finely-focused gallium ion beam on the surface of bulk sample to make a specially shaped specimen by ion-sputtering. Since FIB can prepare the TEM specimen with specific size and thickness from an arbitrary area, this technique is indispensable for the preparation of TEM specimen. Common FIB instrument configuration includes not only an ion gun but also an electron beam gun, called multi-beam or FIB-SEM system. This system can acquire SEM images such as secondary electron image (SEI) and backscattered electron image (BEI). The instrument can observe the surface morphology and Z contrast of the sample to help us prepare a TEM specimen. With detection of secondary electrons which are generated by the interactions between a gallium ion beam and a sample, the FIB is also able to acquire SIM (Scanning Ion-beam Microscope) images. The SIM image is highly sensitive to the surface structure of sample, compared with the SEM image. In addition, the SIM image shows channeling contrast which depends on the crystal orientation, thus providing the relative difference in the orientations among the crystalline grains. However, such information reflects only qualitative one, and thus the SIM image cannot provide detailed and quantitative information on the crystal orientations.

On the other hand, the EBSD (Electron Back-Scatter Diffraction) method is more useful for obtaining precise crystal orientation information. In the method, the electron back-scatter diffraction pattern (Kikuchi pattern), which is formed by the interaction between electron beam and sample, is acquired using a CCD or CMOS camera. Then, we can conduct quantitative

›› 3-1-2 Musashino, Akishima, Tokyo 196-8558, Japan | E-mail: mkadoi@jeol.co.jp

study of the crystal orientation on the sample surface. By combining EBSD with the scanning function of SEM, a two-dimensional (2D) crystal orientation map of the sample surface can be obtained. Furthermore, the incorporation of an EBSD detector into a FIB-SEM can take a three-dimensional (3D) distribution analysis of the crystal orientations by iterative procedures of milling a sample and then acquiring crystal orientation maps for milling surface at the specified interval.

In our study, we used an FIB-SEM system equipped with an EBSD detector. Then, the crystal orientation information on the sample surface was obtained by the EBSD method, followed by preparing TEM specimens having the target primary crystal orientations by the FIB method. This method was applied to the matrix of Allende meteorite and we prepared TEM specimens of olivine grains having the orientations of [100], [010] and [001] for TEM & STEM observations. From the observation results, we confirmed the effectiveness of our method. In this article, we will report the detailed procedures of this method and the results of observing TEM specimens prepared with the method.

Sample

Allende meteorite was used as a sample for this study (**Fig. 1**). This meteorite belongs to carbonaceous chondrite, which is one of the groups in stony meteorite. Carbonaceous chondrite has chemical composition similar to that of the solar atmosphere. That is, it contains information on the pre-solar or early process of the Solar System, which was lost in many planets and satellites caused by the geological activities after the formation of those bodies. Therefore, it is regarded to be an important sample to study the formation process of the solid materials in the early Solar System. Allende meteorite consists mainly of mm-sized spherical objects, called as chondrules, and irregular shaped Ca, Al-rich inclusions (CAIs) embedded in the matrix composed of fine grained aggregate of olivine, pyroxene and Fe-Ni oxide/sulfide (Fig. 1). Since the grain sizes of constituent minerals of the matrix are very small (<10 µm in size), there have been only a few studies for these grains compared with chondrules and CAIs.

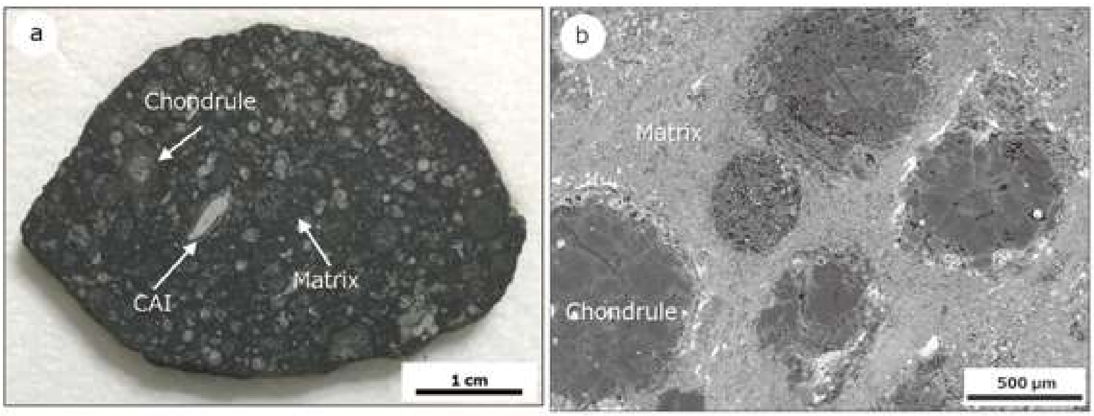
Ohnishi et al. (2018) reported that very thin plate-shaped precipitates (~1 nm in thickness) are generated in the olivine grains from Allende matrix and their long axes are parallel to the (100) plane of the host of olivine. Based on their results, they suggested that the precipitates are Fe, Cr-rich oxides and they were probably formed by the high-temperature oxidation process of olivine [1]. However, it should be noted that the number of olivine grains that they studied is limited and it is still controversial whether the plate-shaped precipitates commonly occur in the matrix olivine. In order to observe the precipitates parallel to the (100) plane, we need to observe them in the specific orientation (for example, [010] or [001]). But the olivine grains are randomly oriented in the matrix and thus, if we study the TEM specimen prepared by using the conventional FIB method or the ion-milling method, many efforts and long hours are required for searching the target grains and the orientation alignment. To overcome this situation, we applied our method to prepare TEM specimens from the matrix of olivine grains in the [001], [010] and [100] orientations and then, we examined whether the precipitates reported in [1] commonly occur or not in olivine grains from the Allende matrix.

For the FIB-SEM observations, we conducted the following two steps. A thick section of Allende meteorite was embedded by conductive resin and the surface of embedded section was mechanically polished. For performing EBSD analysis after these steps, mirror polishing using a colloidal silica was performed to obtain mirror polish of the surface. Furthermore, in order to prevent electrical charging, osmium coating was applied to the mirror polished surface of embedded section.

Instruments

For EBSD analysis and TEM specimen preparation, we used a JEOL Multi-Beam System, JIB-4700F (**Fig. 2**) which is equipped with an EDAX DigiView EBSD detector. The JIB-4700F is a FIB-SEM system equipped with both a gallium ion-beam gun and a Schottky electron gun. This system enables us to perform both FIB milling and high spatial-resolution SEM observation and analyses in a single instrument. The JIB-4700F also includes the Picture Overlay function [2] for performing overlay display of an arbitrary image on the FIB image or SEM image. The use of this unique function allows us to provide overlay-displaying the SEM image on the SIM image, leading

Fig. 1 Optical micrograph of a thick section of Allende meteorite (a) and SEM image obtained from an area of surface of the section (b).



(a) Optical reflection image of Allende meteorite. The meteorite is composed of white-colored CAIs and chondrules (a few mm in size), black-colored matrix which fills the gaps among CAIs and chondrules.
(b) SEM image of Allende meteorite (backscattered electron image). The matrix of aggregated small grains occurs as filling the gaps in chondrules.

to highly precise positional setting of deposition or milling onto the target region. For observing the TEM specimen prepared by FIB milling, a JEOL Atomic Resolution Analytical Electron Microscope, JEM-ARM300F, was used. This microscope is our 300 kV aberration-corrected S/TEM equipped with a cold field emission gun [3-4]. In the JEM-ARM300F, two types of the objective lens pole-pieces; FHP (Full High resolution Pole-piece) for ultra-high resolution imaging and WGP (Wide Gap Pole-piece) for ultrahigh sensitive analysis are selectable. In our study, the microscope equipped with the WGP was used.

Experimental Results and Discussion

The workflow of preparation and observation of TEM specimens using the method in our study is shown in **Fig. 3**. First, the crystal orientation maps were obtained from the matrix in a thick section of Allende meteorite using the EBSD method, and olivine grains were selected for the preparation of TEM specimens. Next, TEM specimens of the selected grains were prepared with FIB function. At that time, using the Picture Overlay function, the acquired crystal orientation map was overlaid on the SEM image for helping us to determine the milling position, and then electron-beam deposition was applied to this position for preparing TEM specimens. Finally, TEM observations were performed to evaluate the prepared TEM specimens. The following descriptions will show each process of the method in detail.

Selecting olivine grains for TEM observations

In **Fig. 4**(a) to (c), an SEM image of the matrix of Allende meteorite and crystal orientation maps of olivine grains, which were obtained from the same field of view using the EBSD method, are shown. From Fig. 4(b) and (c), it is clearly confirmed that olivine grains in the matrix are randomly oriented. From them, the preparation of TEM specimens for observing the grains having the [100], [010] and [001] orientations was carried out with the following procedures. First, as shown in Fig. 4(b), olivine grains having the [001], [100] and [010] orientations, which are aligned within the tilt angle of 20 degrees in the direction vertical to the polished surface of the sample (Z direction), were selected and colored (**Fig. 5**(a)). The crystal structure of olivine belongs to the orthorhombic crystal system, and thus the [100], [010] and [001] orientations are orthogonal to each other (Fig. 4(d)). Therefore, if TEM specimen is prepared for the above-mentioned, selected and colored grains in the direction parallel to the sample surface, it becomes possible to obtain the TEM specimens of olivine grains having the [100], [010] and [001] orientations within at least the tilt angle of 20 degrees. Even if this condition is satisfied, several grains and two kinds of the orientations in the direction parallel to the sample surface become candidates for milling (for example, in the case of grains having the [001] orientation parallel to the Z direction, it is possible to prepare TEM specimens in the two directions of [100] and [010]). From them, we selected grains and directions so that the TEM specimens can be made over the area as large as possible with the FIB milling accuracy. After this selection, we prepared TEM specimen for observing in the [100] direction from the olivine grains having the [001] orientation within the tilt angle of 20 degrees in the Z direction (Fig. 5(b)). In the same way, TEM specimen for observing in the [010] direction was prepared from the olivine grains having the [100] orientation (Fig. 5(c)), and also for observing in the [001] direction from the olivine grains having the [010] orientation (Fig. 5(d)).

Preparation of TEM specimen by FIB

For the preparation of TEM specimens by FIB, we utilized the bulk pickup method, which uses a pickup tool independent of the FIB system, described as follows. First, a thick specimen block is prepared by FIB milling. This block is mounted onto a TEM grid using the pickup tool. After that, FIB milling is performed again for the preparation of a thin specimen.

The detail procedures of preparation of TEM specimen for observation in the [100] direction of olivine, using the bulk pickup method in our study, are shown in **Fig. 6**. First, electron-beam deposition was performed on the target area (grain) to be milled. Next ion-beam deposition was further performed on the electron-beam deposition area. During the electron-beam deposition on the target area, the Picture Overlay function was used to overlay the crystal orientation map on the SEM image,

Fig. 2 Appearance of the JIB-4700F

The JIB-4700F is an FIB-SEM system that is equipped with a gallium ion-beam gun and a Schottky electron gun. The system also includes the 3D observation function. Furthermore, the EDS (Energy Dispersive X-ray Spectroscopy) and the EBSD systems are optionally available, thus utilizing a diversified range of applications.

Fig. 3 Workflow of preparation and observation of TEM specimens using our method.

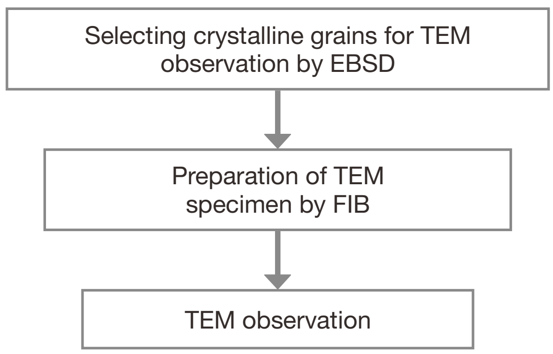
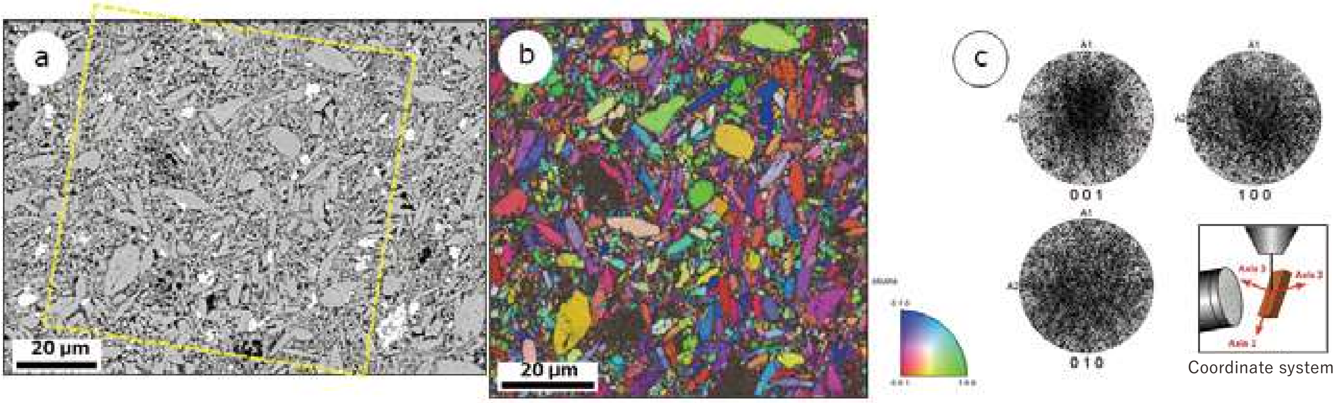
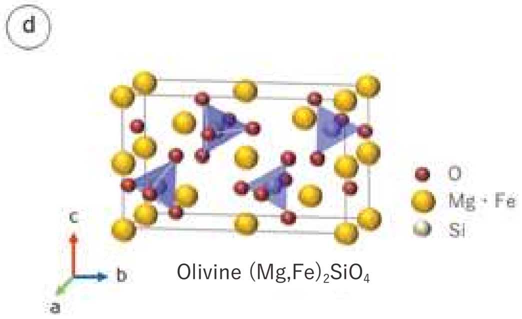


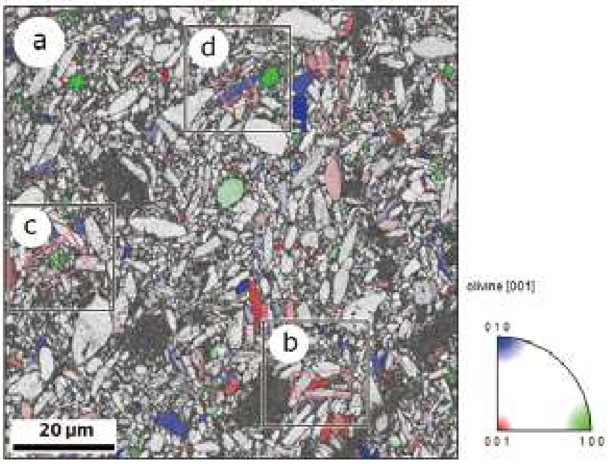
Fig. 4 SEM image of the matrix of Allende meteorite (a), Crystal orientation map of olivine grains (b), Pole Figures (PF) of the (001), (100) and (010) planes (c), and an ideal crystal structure of olivine (d).



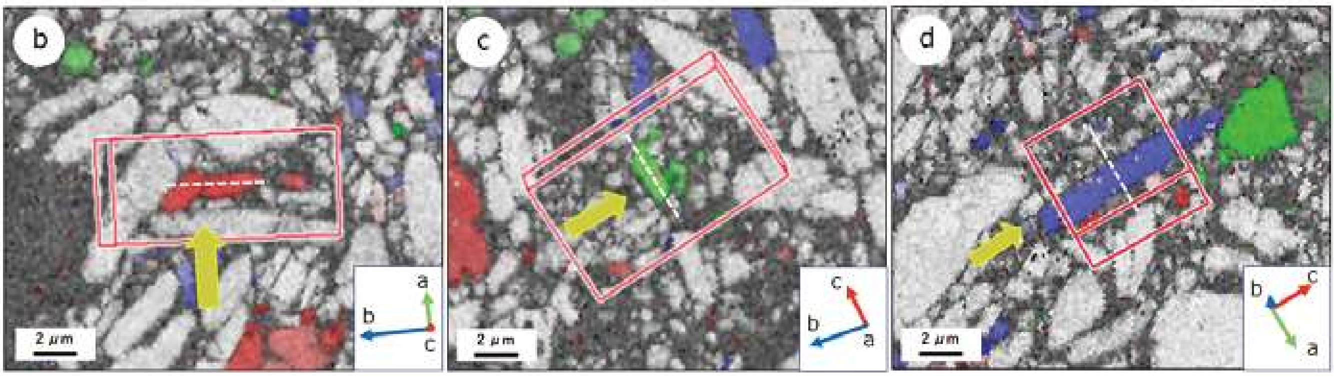


(a) SEM image of the matrix of Allende meteorite (backscattered electron image).
(b) Crystal orientation map (IPF(Z) + Fit) of the olivine grains from the area corresponding to the yellow-colored dotted box in (a). The crystal orientations of the olivine grains in the Z direction (direction vertical to the polished surface of the sample) are exhibited with colors. Different color keys to the lower right of the orientation map indicate the corresponding crystal orientations.
(c) Pole Figures (PF) of the (001), (100) and (010) planes.
(d) Ideal crystal structure of olivine.

Fig. 5 Crystal orientation maps of olivine grains selected for the preparation of TEM specimens.



(a) Crystal orientation map of only the olivine grains having the [001], [100] and [010] orientations within the tilt angle of 20 degrees in the direction vertical to the sample surface (Z direction), which were preferentially selected and colored from the result of Fig. 4(b). (b-d) Enlarged images of the selected and colored olivine grains for the preparation of TEM specimens.
(b) Olivine grains with the orientation close to [001], which were subjected to the preparation of TEM specimen for observing the grain in the [100] direction.
(c) Olivine grains with the orientation close to [100] for observing in the [010] direction.
(d) Olivine grains with the orientation close to [010] for observing in the [001] direction. Red-colored frames in Fig. 5(b-d) show the unit cell models of olivine estimated by EBSD analysis. White-colored dotted lines in Fig. 5(b-d) correspond to the area milled for TEM cross-sectional specimen, and yellow-colored thick arrows in these three figures show the direction of TEM observation.



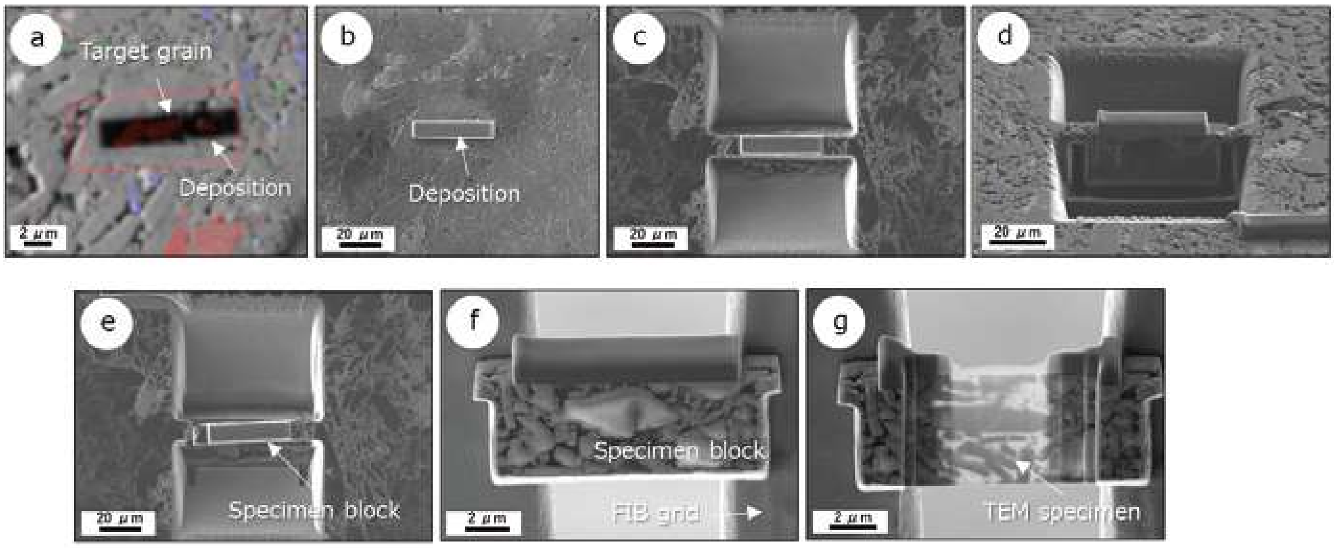
so as to precisely select the grains to be milled and to locate the milling position (Fig. 6(a) and **Fig. 7**(a), (c)). After the deposition, a specimen block, which included the deposition layer and a part of the sample, was cut out at an accelerating voltage of 30 kV (Fig. 6(c-e)). After that, this chunk was moved to the FIB grid using the pickup tool, and it was mounted on the grid by epoxy resin (Fig. 6(f)). Finally, the chunk mounted on the grid was returned to the FIB system and it was thinned using the ion beam (accelerating voltages: 30 kV and 5 kV). With this procedure, the specimen was thinned down to about 80 nm in thickness, which enables us to perform high-resolution TEM/STEM observations (Fig. 6(g)). With the same procedures, TEM specimens were also prepared for observation in the [010] and [001] directions of olivine (Fig. 7).

Evaluation of Prepared Specimens Using TEM

TEM specimens prepared by the above-mentioned method were evaluated using TEM. **Figure 8** shows the upper Selected Area Diffraction (SAD) patterns and the lower atomic-resolution STEM-HAADF (High Angle Annular Dark Field) images in the [100], [010] and [001] direction of olivine, respectively.

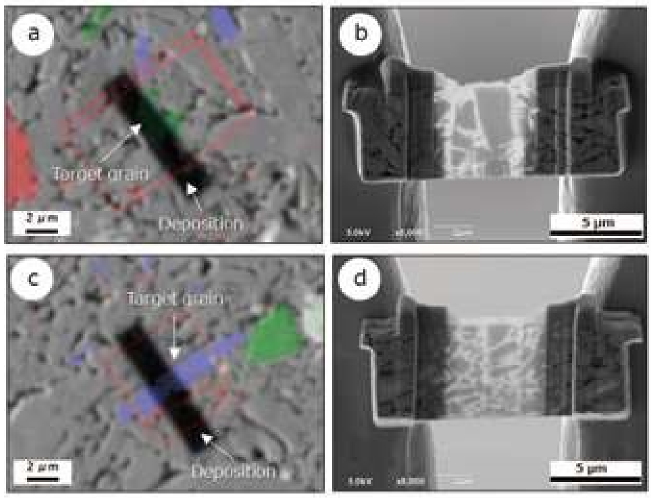
As shown in Fig. 8, atomic-columns were clearly observed in every direction, thus suggesting that all the prepared specimens are thin enough for atomic-resolution observations. From the results of STEM-HAADF observation, we confirmed that plate-shaped precipitates (<1 nm in thickness) occur along the (100) plane of olivine in the [010] and [001] directions of olivine (Fig. 8), while no distinct defects were found in the [100] direction of olivine. These results are consistent with the previous study [1],

Fig. 6 Secondary electron images of TEM specimen prepared for observation of the [100] direction of olivine.



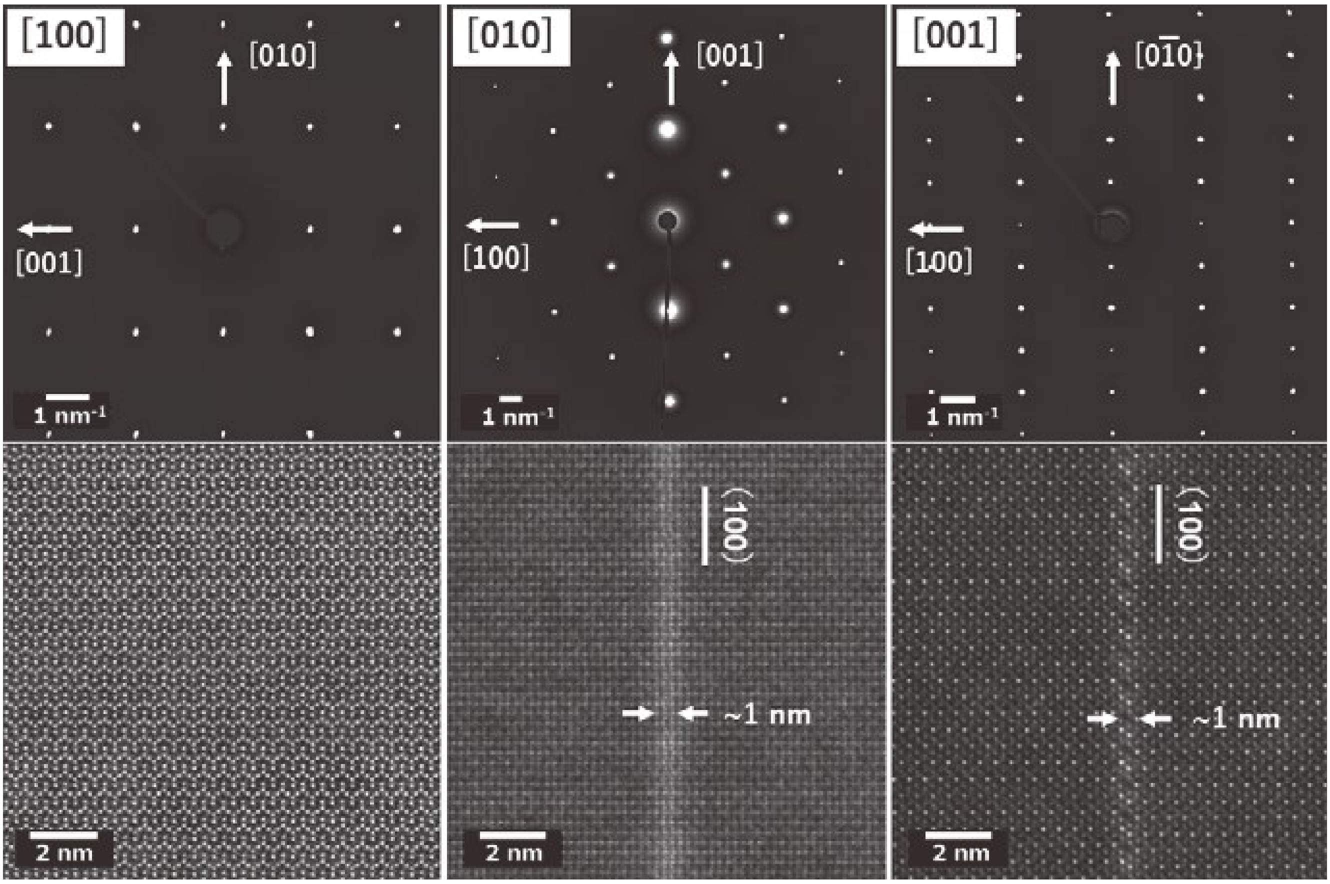
(a) Electron-beam deposition. Using the Picture Overlay function, the crystal orientation map was overlaid on the secondary electron image to perform the deposition precisely on the target milling position. This image was obtained from the same area as that of Fig. 5(b).
(b) Ion-beam deposition directly onto the electron-beam deposition area.
(c) FIB milling was performed for both the front and back side of the deposition area to create a hole.
(d) The sample was tilted so that it can be observed with the FIB-SEM from the oblique direction, and then the bottom portion of the milled specimen was cut.
(e) Both sides of the deposition were cut out to separate a specimen block from the host of the sample.
(f) The cut-out block was mounted on the FIB grid.
(g) Thin specimen was made by milling the specimen block.

Fig. 7 Secondary electron images of area of TEM specimens prepared for observation in the [010] and [001] directions of olivine (a, c), and of the prepared TEM specimens (b, d).



(a, b) olivine grains for observation in the [010] direction.
(c, d) olivine grains for observation in the [001] direction.

Fig. 8 TEM observation results of the prepared specimens of the matrix olivine in Allende meteorite in the [100], [010] and [001] directions.



Upper: Selected electron diffraction patterns in each direction. Lower: Atomic-resolution STEM-HAADF images. Instrument: JEM-ARM300F (WGP). Accelerating voltage: 300 kV.

indicating that plate-shaped precipitates might commonly occur in the matrix olivine of Allende meteorite.

Table 1 shows the tilt angles of the TEM goniometer stage when the SAD patterns were obtained in each orientation. These tilt angles were found to be within 5 degrees for all the prepared specimens. The small tilt angle of the stage can be due to the accuracy of the crystal orientation map obtained by the EBSD method, the FIB milling accuracy for the bulk pickup, the precision for mounting the specimen on the FIB grid and for thin specimen milling, the errors in loading the FIB grid on TEM specimen holder, etc. We successfully achieved the tilt angle of the TEM stage within 5 degrees under any conditions. This result suggests that our method described in this article is very effective for preparation of TEM specimens having the target primary orientations, where EBSD and FIB are combined.

Summary

In our study, we examined a method which combines EBSD and FIB-SEM, for preparing TEM specimens with specific crystal orientations and applied the method to the matrix of Allende meteorite. From the results, we confirmed that our method enables us to prepare TEM specimens having the specific orientations with high accuracy. That is, by using this method, the TEM specimens of olivine grains with [100], [010] and [001] orientations were prepared within the accuracy of tilt angle of 5 degrees using the TEM goniometer stage.

In the preparation of TEM specimens with FIB, the selection of the target crystalline grains by using the EBSD method greatly reduces the efforts and hours required to find the target crystal and adjust the crystal orientation to a specific direction in the TEM observation steps. Thus, it is suitable to apply our method not only to the matrix of Allende meteorite, but also to the powder specimens (ceramics, minerals. etc.) or the poly-crystal specimens composed of randomly oriented grains.

Table 1 Tilt angles of TEM goniometer stage when the selected electron diffraction patterns in Fig. 8 were obtained.

		TEM specimen		
		[100]	[010]	[001]
Tilit	X	-1.7	-3.5	+3.4
	Y	+2.6	+2.7	-0.5

References

[1] I. Ohnishi. et al. (2018) ***81st Annual Meeting of The Meteoritical Society*** 6105.
[2] Y. Ikeda, ***TEM Users Meeting 2014*** abstract (in Japanese).
[3] H. Sawada. et al. (2014) ***JEOL News*** vol.**49** p.51-58.
[4] E. Okunishi. et al. (2015) ***JEOL News*** vol.**50** p.58-63.