Structual change of non-crystalline biogenic silica of diatoms by heat treatment

Structural change of non-crystalline biogenic silica of diatoms by heat treatment

Masayuki Okuno[1], Yuka Kitagawa[2], Ryuji Asada[1] and Kazue Tazaki[1]

[1]*Department of Earth Sciences, Faculty of Science, Kanazawa University, Kakuma, Kanazawa, Ishikawa 920-1192 Japan*

[2]*Graduate School of Natural Science and Technology, Kanazawa University, Kakuma, Kanazawa, Ishikawa 920-1192 Japan.*

Abstract

Diatoms are plankton of unicellular whose cell walls (exoskelton, flustule) are made of amorphous silica and organic materials. Therefore, diatom is interested in the fields of material science and biomineralization. In this study, the nano-structure of diatom flustules was investigated by means of X-ray diffraction measurements and micro FTIR spectroscopy. Obtained X-ray diffraction profile of diatoms indicated that diatoms were composed by non-crystalline SiO_2 materials. Its relatively high intensity small angle scattering suggested that the basic structure of this non-crystalline matter is similar to silica gels. The IR spectrum of diatom shows the Si-OH band as well as Si-O band. This is consistent with the result of X-ray measurement. Further, The relative intensity of Si-O-Si band of diatom is lower than the fully polymerized SiO_2 materials such as quartz and vitreous SiO_2. These facts may indicate that network structure of silica base non-crystalline materials of diatom includes some Si-OH bonds and large pore. The structural evolution of this silica base flustule by heat treatment up to 1150 °C was also investigated by means of IR spectroscopy. By the heat treatment up to 400 °C, diatom lose organic matters, OH and Si-OH bond. This may indicate the polymerization of SiO_4 tetrahedra advanced by dehydration of fulustule. Heat treated diatoms show more sharp Si-O band in

IR spectra than non treated ones. This suggests that heat treated diatom has a relatively order SiO_4 tetrahedra in its basic structure.

Key words: Biogenic silica, Pennate diatoms, X-ray diffraction, FTIR, Heat treatment

INTRODUCTION

The diatom is the photosynthesis algae of unicellular which has siliceous exoskeltons (frustules). The frustule is formed in silica deposition vesicle (SDV) which exists in the cell membrane (Vrieling et al. 1999a and 1999b). Some kinds of amino acids with hydroxyl residue in the cell wall protein have been observed (Hecky et al. 1973). Therefore, it is considered that silica of the diatom frustule unites with the hydroxyl residue of the amino acid. Nakashima and Ito (1996) suggested that a silica-protein complex is made on the diatom frustle and silica is important for chemical evolution of the primitive life.

The silica frustules of diatoms deposit and accumulate on the sea floor, after the diatoms die. These made diatomaceous earth and diatom ooze. The diatom earth is used for various industrial materials by their characters such as absorption and heat resistibility.

By the IR spectroscopic study, it was reported that silica flustules of diatoms have some variations among species (Kamatani 1971; Perry 1989). Asada et al. (2002) analyzed the structure of diatom silica frustule by polarized micro IR spectroscopy and reported an anisotropy of their nano-scale structure.

In this study, the structure of diatom flustule is investigated precisely by X-ray diffraction measurement and micro FTIR spectroscopy. Further, the structural evolution of diatom frustules by heat treatment was also investigated by means of IR spectroscopy and we analyzed the detailed characters of nano scale structure.

SPECIMEN AND EXPERIMENTS

Specimen

Pennate diatoms were collected from acidic hot springs at Kamuiwakka Falls, Shiretoko Peninsula, Hokkaido, Japan (Asada et al. 2002). This hot spring water shows

low pH between 1.3 to 1.9 and were well investigated in our previous works (Asada and Tazaki 2001; Asada et al. 2002). Figure 1 is an optical micrograph of the pennate diatom. It shows broad linear valve and the rectangular girdle bands. The diatoms were also observed by a scanning electron microscope equipped by energy dispersive X-ray analyser (SEM-EDX; JEOL JSM-5200LV). SEM observation reveals that long-axis of pennate diatom is about 30 μm in length (Fig. 2). EDX analyses showed that exoskeleton of diatoms are composed of large amount of Si with a little S.

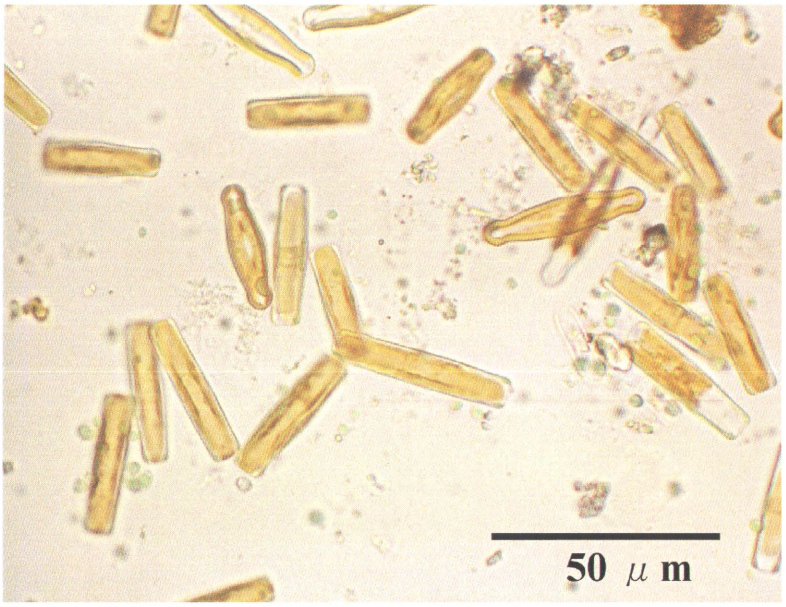

Figure 1 Optical microraph of pennate diatoms collected from an acidic hot spring at Kamuiwakka Falls, Shiretoko Peninsula, Hokkaido, Japan.

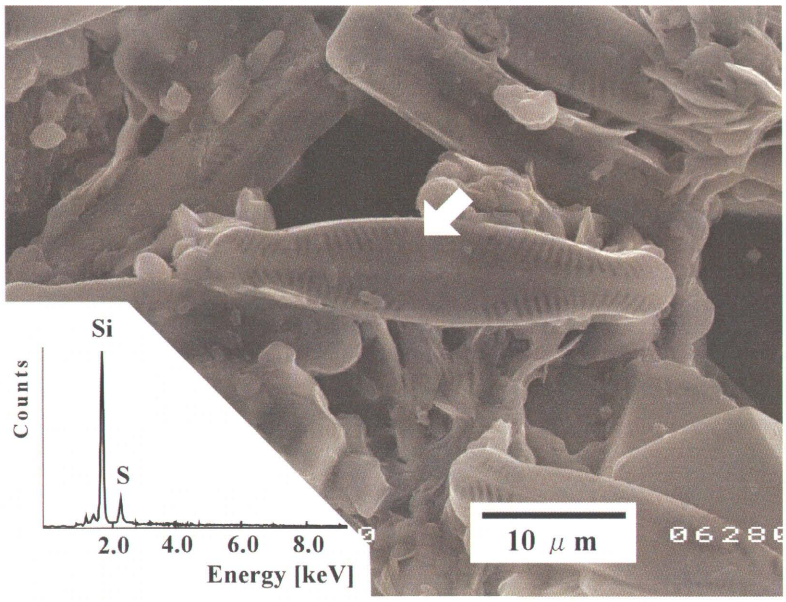

Figure 2 SEM image of pennate diatoms and its EDX analysis (Arrow: analytical point).

Structural analyses of diatom at ambient temperature

X-ray Powder diffraction analysis

The samples were washed by distillated water to remove the contamination derived from hot spring solution. About 10 mg of diatom samples was mounted on a Si plate that show no Bragg diffraction. X-ray diffraction measurements at ambient temperature were carried out on a powder X-ray diffractometer (Rigaku RINT2200). Cu Kα radiation was used for the measurements. A profile of vitreous SiO_2 was also measured as a reference material.

FTIR analyses

The structural analysis of the frustules of diatom at ambient temperature also carried out by means of Fourier Transform Infrared micro spectroscopy (micro-FTIR). Measurements of IR spectra were carried out under a micro-FTIR system consisted of a FTIR spectrometer (JASCO FTIR-610) and an IR microscope (JASCO MICRO-20) with x32 objective by transmission method in the range of $\nu = 650 - 4000\ cm^{-1}$.

A drop of cell suspension of diatom was mounted on a fluorite (CaF_2) disk (0.5 mm in thickness, 10 mm in diameter). After air-drying, several cells were selected under the IR microscopy by a rectangular masking aperture about 30 μm x 40 μm. 100 spectra are accumulated to obtain one final spectrum. In order to compare with the spectrum of diatom, those of SiO_2 gel, low quartz, and vitreous SiO_2 were also measured.

Structure analyses of heat treated diatom

In order to investigate the structural changes of biogenic silica of diatoms by heat treatment, a micro-FTIR spectroscopy was also adopted. A drop of cell suspension of diatoms was put on a platinum disk and held in an electric furnace at 50, 100, 200, 300, 400, 500, 600, 700, 800, 1000, and 1150 °C under the ambient pressure. The duration of heat treatment is about 24 hours for each run. Then, the specimens were rapidly cooled down in air. After the heat treatment, the samples were moved on a CaF_2 disk for micro-FTIR measurement. Measurements were carried out under the same procedure as that for non-treated diatom.

RESULTS

X-ray powder diffraction analyses of the non-treated diatom

Figure 3 shows obtained X-ray powder diffraction profile of non-treated diatom together with that of vitreous SiO_2. The profile of diatom shows no distinct diffraction peaks but a broad peak around $2\theta \cong 20°$. This fact suggests that diatoms are composed by non-crystalline SiO_2 materials.

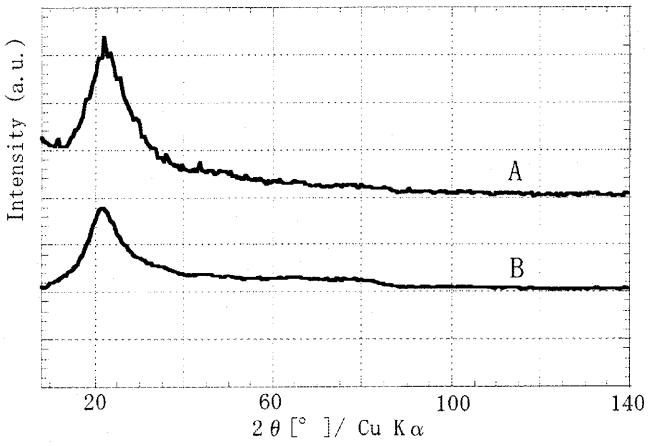

Figure 3 XRD analyses of the diatoms (A) and silica glass (B).

IR spectrum of non-treated diatoms

Figure 4 shows the IR spectrum of several cells of non-treated diatoms. It shows seven distinctive bands. These bands may be assigned to O-H stretching (3100 - 3700 cm^{-1}), C-H stretching (2925 cm^{-1}), C=O stretching and H-O-H bending (1647 cm^{-1}), C-H-N combination (1534 cm^{-1}), Si-O stretching (1000 - 1230 cm^{-1}) and Si-OH stretching (949 cm^{-1}). The Si-OH and Si-O bands mainly results from frustules of diatom. These broads Si-OH and Si-O bands may indicate that the frustles are made of non-crystalline SiO_2

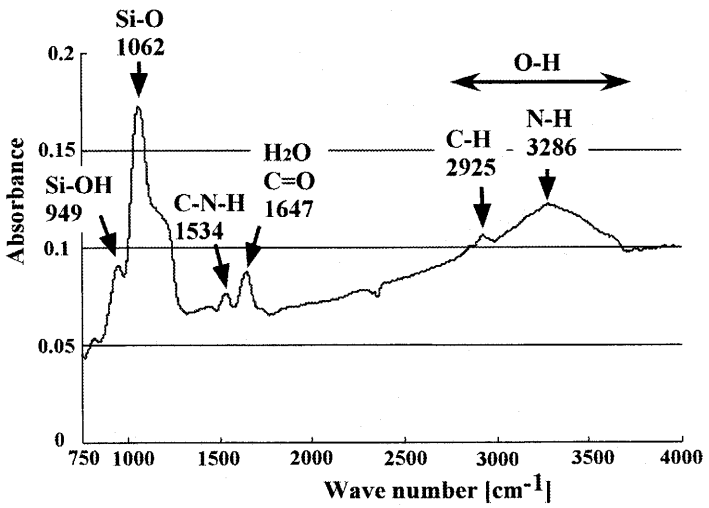

Figure 4 IR absorbance spectrum of diatoms under IR microspectroscopy.

materials with Si-OH bonds. The C-H, C=O, and C-N-H bands mainly results from organic matter such as polypeptides and polysaccharides in the diatoms.

IR spectrum of heat-treated diatoms

Figure 5 shows the infrared spectra of heat-treated diatoms. These spectra show some temperature variations of band shape and intensity. The intensities of O-H, C-H, H_2O, C=O, C-N-H, and Si-OH bands decrease with increasing temperature of treatment, and these bands disappear at 400 °C. The intense broad band of $\nu = 1062$ cm^{-1} has high frequency shoulder. This band separate to two components as increasing temperature, and shows distinct two bands above 600 °C. These two bands gradually shift toward high frequency with temperature. A band at $\nu = 1062$ cm^{-1} (RT) shift to 1075 cm^{-1} (1150 °C). The other band at $\nu \sim 1200$ cm^{-1} (shoulder, RT) shift to 1220 cm^{-1} (1150 °C).

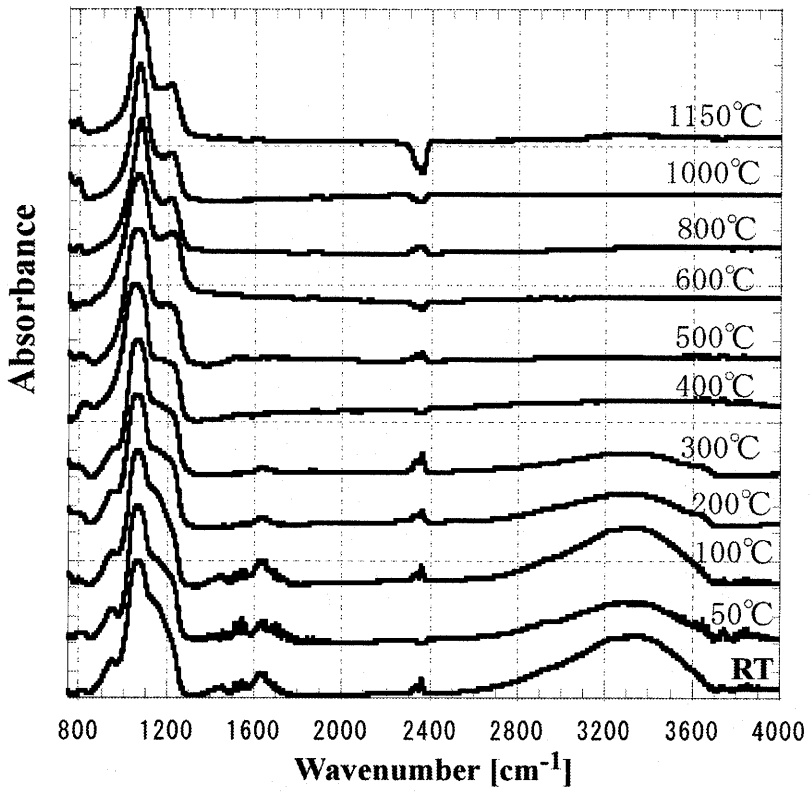

Figure 5 The infrared spectra of the heated diatoms. RT: room temperature.

DISCUSSION

Average structure of non-crystalline frustules of diatom

The X-ray powder diffraction profiles of non-treated diatom shows no sharp peaks like vitreous SiO_2. Therefore, diatoms may be made of non-crystalline SiO_2 materials. Comparing this profile with that of vitreous SiO_2, some small differences were observed (Fig. 3). The intensity in the low 2θ range $2\theta < 10°$ of diatoms is higher than that of the vitreous SiO_2. On the other hand, SiO_2 gel also has similar low angle scattering intensity (Kamatani 1971). SiO_2 gel have primary structures of 10 - 100 nm scale. Therefore, it is possible that micro structure of diatoms have structural units with 10 - 100 nm scale.

Obtained IR spectrum in the region of ν = 700 - 1500 cm^{-1} for non-treated diatom was compared with the spectra of SiO_2 gel, low quartz and vitreous SiO_2 (Fig. 6). All these spectra show strong bands at $\nu \approx 1100$ cm^{-1} and weak band at for $\nu \approx 800$ cm^{-1}. These bands have been assigned to Si-O stretching and Si-O-Si bending modes, respectively (Williams et al. 1993). These facts suggest that basic micro structure of diatoms may be the network of SiO_4 tetrahedra. However, the band intensity at $\nu \approx 800$ cm^{-1} of diatoms is weaker than those of low-quartz and vitreous SiO_2. Further, the spectrum of diatom shows another band at $\nu \approx 950$ cm^{-1}. This band is assigned to Si-OH

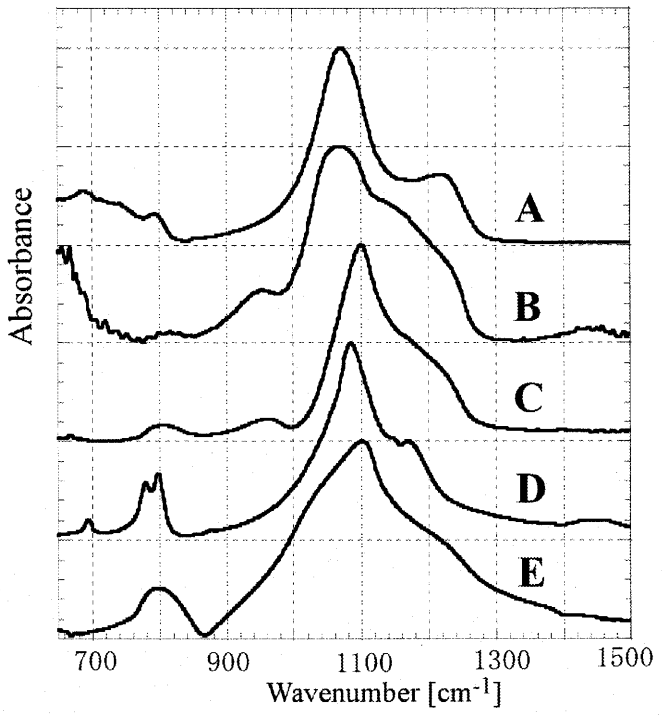

Figure 6 Comparison between IR spectra of heated diatoms: 1000 ℃ (A), untreated diatoms (B), silica gel (C), low-quartz (D) and silica glass (E).

stretching mode. These features of IR spectrum of diatom are also observed on the spectrum of SiO_2 gel. Moreover, the intensity of this band is larger than SiO_2 gel. Therefore, the number of Si-OH bands in diatoms are larger than ones of SiO_2 gel.

These results are consistent with the structural model of diatoms that is a complex of non-crystalline SiO_2 materials and organic matter. However, the shape of the band $\nu \approx 1100\ cm^{-1}$ of diatom is slightly different from that of SiO_2 gel. The band width for the spectrum of diatom is wider than that of SiO_2 gel. Asada et al. (2002) explained this wide band width of Si-O mode in diatoms by the strong anisotropy of SiO_4 networks based on the polarized IR data of diatoms.

Structural evolution of diatom by heat treatments

By the heat treatment, the C=O, C-N-H, O-H, and H_2O bands in IR spectra of diatom disappear at 400 °C (Fig. 5). This fact suggests that organic matters in diatoms decompose by heat treatment. Further, the IR intensity of Si-OH band ($\nu \approx 950\ cm^{-1}$) as well as O-H band decreases by heat treatment and this band also disappear at 400 °C (Fig. 7). On the other hand, the intensity of Si-O-Si band ($\nu \approx 800\ cm^{-1}$) increases by heat treatment (Fig. 5). These variations may indicate that OH- groups decompose forming additional oxygen bridge in the network structure according to

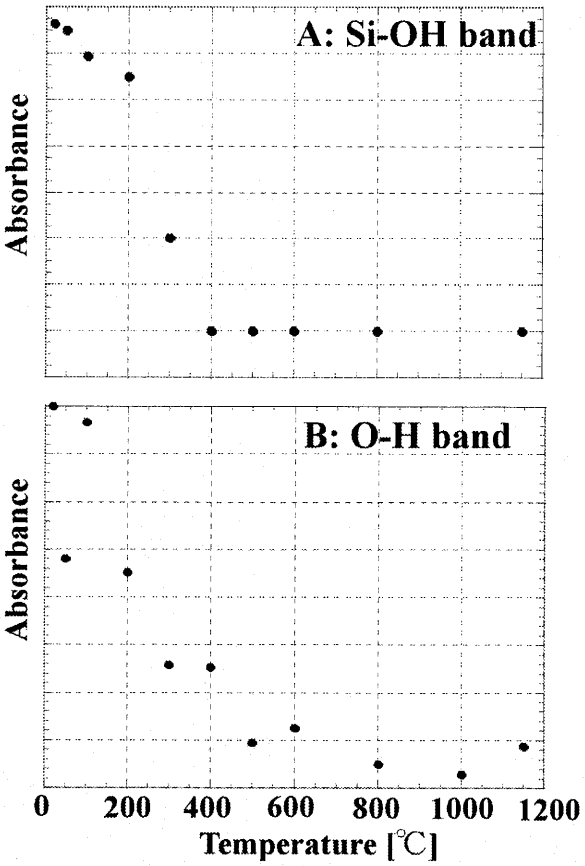

Figure 7 The IR absorbance changes of OH related band in diatoms by heat treatment. A: Si-OH band, B: O-H band.

$$\text{–Si–OH} + \text{OH–Si–} \longrightarrow \text{–Si–O–Si–} + H_2O\uparrow$$

This process included dehydration and additional polymerization of silica. Similar process could be found in a silica

biomineralization of unicellular microbes (Asada and Tazaki 2001) and non-crystalline mullite phase formation by sol-gel method (Okuno et al. 1997).

The position of Si-O band (ν = 1062 cm^{-1} in non-treated diatom) shifts to higher wavenumber (ν = 1075 cm^{-1} at 1050 °C) and a shoulder of this band ($\nu \approx$ 1200 cm^{-1} in non-treated diatom) also shifts to $\nu \approx$ 1220 cm^{-1} at 1050 °C and becomes a sharp clear band. This result may indicate that Si-O bonds in non-crystalline diatom cell become strong and more ordered by heat treatment. Probably this change may relate to dehydration and polymerization of network structure. Asada et al. (2002) reported a similar variation of Si-O band in a pennate diatom cell by polarized IR measurement.

ACKNOWLEDEMENTS

A part of this research was supported by the Japan Society for the Promotion of Science and by the grants from the Japanese Ministry of Education, Science and Culture awarded to Kazue Tazaki.

REFERENCES

Asada, R. and Tazaki, K. (2001) Silica biomineralization of unicellular microbes under strongly acidic conditions. *The Canadian Mineralogist*, **39**, 1-16.

Asada, R., Okuno, M. and Tazaki, K. (2002) Structural anisotropy of biogenic silica in pennate diatoms under Fourier transform polarized infrared microspectroscopy. *Journal of Mineralogical and Petrological Sciences*, **97**, 219-226.

Kamatani, A. (1971) Physical and chemical characteristics of biogenous silica. *Marine Biology*, **8**, 89-95.

Hecky, R. E., Mopper, K., Kilham, P. and Degens, E. T. (1973) The amino acid and sugar composition of diatom cell-walls. *Marine Biology*, **19**, 323-331.

Nakashima, S. and Itoh, Y. (1996) Organic / inorganic interactions and evolution / material cycle of Earth. in Japanese, *Gekkan Chikyu*, **18**, 24-30.

Okuno, M., Shimada, Y., Schmücker, M., Schneider, H., Hoffbauer, W. and Jansen, M. (1997) LAXS and Al MAS NMR studies on the temperature-induced changes of non-crystalline single phase type mullite precursors. *Journal of Non-Crystalline Solids*, **210**, 41-47.

Perry, C. C. (1989) Chemical studies of biogenic silica. In: Mann, S., Webb, J., Williams, R.J.P. (Eds.), Biomineralization: Chemical and Biochemical Perspectives. VCH, Yew York, pp. 223-256.

Vrieling, E. G., Beelen, T. P. M., van Santen, R. A. and Gieskes, W. W. C. (1999a) Diatom silicon biomineralization as an inspirational source of new approaches to silica production. *J. Biotechn.*, **70**, 39-51.

Vrieling, E. G., Gieskes, W. W. C. and Beelen, T. P. M. (1999b) Silicon deposition in diatoms: control by the pH inside the silicon deposition vesicle. *J. Phycol.*, **35**, 548-559.

Williams, Q., Hemly, R. J., Kruger, M. B. and Jeanloz, R. (1993) High Pressure Infrared Spectra of α-Quartz, Coesite, Stishovite and Silica Glass. *J. Geophys Res.*, **98**, B12, 22157-22170.