Raman spectroscopy of synthetic $CaHPO_4 \cdot 2H_2O$– and in comparison with the Cave mineral brushite

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Abstract

The mineral brushite has been synthesised by mixing calcium ions and hydrogen phosphate anions to mimic the reactions in a Cave. The vibrational spectra of the synthesised brushite were compared with that of the natural Cave mineral. Bands attributable to the PO_4^{3-} and HPO_4^{2-} anions are observed. Brushite, both synthetic and natural, is characterised by an intense sharp band at 985 cm^{-1} with a shoulder at 1000 cm^{-1}. Characteristic bending modes are observed in the 300 to 600 cm^{-1} region. The spectra of the synthesised brushite matches very well the spectrum of brushite from the Moorba Cave, Western Australia.

Key words: brushite, monetite, phosphate, Raman spectroscopy, infrared spectroscopy

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Introduction

Brushite is a mineral with the chemical formula $CaHPO_4 \cdot 2H_2O$. The mineral is understood to be a precursor of apatite and is found in guano-rich Caves, formed by the interaction of guano with calcite at a low pH [1,2]. It forms crystals of prismatic shape having a monoclinic crystal structure. The mineral will transform to monetite upon dehydration. Brushite is found in kidney stones [3-7]. However the mineral is often found in Caves and is often regarded as a 'Cave' mineral [8,9]. The mineral has been identified in quite a large number of Australian Caves including the Skipton lava tube Caves (southwest of Ballarat, Victoria, Australia) and at Moorba Cave (Jurien Bay, Western Australia). As such, many minerals formed in these Caves besides brushite include archerite $(K,NH_4)(H_2PO_4)$ [10], mundrabillaite $(NH_4)_2Ca(HPO_4)_2 \cdot H_2O$ [11] and stercorite $Na(NH_4)HPO_4 \cdot 4H_2O$. These minerals occur as stalactites and as crusts on the walls and floors of the Caves. Other minerals found in the Moorba Cave include tanarakite, ardealite, hydroxylapatite, calcite and gypsum. These minerals are formed through the chemical reactions of calcite with bat guano. These Moorba Caves have been in existence for eons of time and have been dated as more than 350 million years old.

The minerals monetite $CaHPO_4$ and brushite $CaHPO_4 \cdot 4H_2O$ have been widely studied [12-15]. In terms of spectroscopy, the minerals are important because of their existence in kidney stones and other biological materials [3,4,16,17]. S.D. Ross in Farmer's treatise (page 393) [18] reported the infrared spectrum of brushite. Data for two mineral samples were provided. However, the results of the two analyses differ. The ν_1 symmetric stretching mode was given as occurring at 1005 cm^{-1}, ν_2 bending mode at 400 and 418 cm^{-1}, ν_3 antisymmetric stretching mode at 1075 and 1135 cm^{-1}, and the ν_2 bending mode at 577 cm^{-1}. Multiple OH stretching vibrations were observed. Other bands associated with the HPO_4 units were found at 750, 875 and 1217 cm^{-1}.

Raman spectroscopy has proven most useful for the study of mineral structure. The detailed comparative Raman spectra of the Cave minerals have not been published. In order to test the brushite from Caves, brushite was synthesised in the laboratory to find how easy the mineral is to form. The idea is to mimic the chemical reactions in the Caves. The objective of this research is to report the Raman spectra of brushite and to relate the spectra to the mineral structure.

Experimental

Synthesis of brushite

The chemicals were purchased as follows: sodium phosphate dibasic from Sigma-Aldrich and calcium chloride from Ajax Chemicals. A calcium chloride solution was prepared by dissolving calcium chloride (25 g) in 100ml water, while a solution of sodium phosphate was made by dissolving sodium phosphate dibasic (32 g) in 600ml of water. To a 1 L beaker, 100ml of deionised water was added (with agitation), along with the simultaneous addition of calcium chloride (20 mL) and sodium phosphate (120 mL) solutions. The pH of the resultant solution was maintained between 4 and 5, using a sodium di-hydrogen phosphate solution to reduce the pH or a sodium carbonate solution to increase the pH. Two appropriate pumps were used to control the flow rates of addition to ensure both $CaCl_2$ and Na_2HPO_4 solutions completed at the same time. The temperature of precipitation was 25 °C. The solids were separated from the liquid by centrifugation, washed with deionised water, and dried overnight at 105 °C in an oven.

Natural Mineral

The mineral brushite was supplied by the Australian Museum (mineral sample number D43843) and originated from Moorba Cave, Jurien Bay, Dandaragan Shire, Western Australia, Australia. Details of the mineral have been published (page 83) [19].

X-ray diffraction

The ‘Cave’ mineral brushite was placed upon an ultra thin film of paraffin wax (Vaseline). In this way a minimal amount of the quite precious ‘Cave’ mineral brushite is used. X-ray diffraction patterns were collected using a Philips X'pert wide angle X-ray diffractometer, operating in step scan mode, with Cu Kα radiation (1.54052 Å). The Philips PANalytical X’pert PRO diffractometer was operating at 40 kV and 40 mA with 0.25° divergence slit, 0.25° anti-scatter between 5 and 15° (2θ) at a step size of 0.0167°. For XRD at low angle section, it was between 1.5 and 8° (2θ) at a step size of 0.0167° with variable divergence slit and 0.125° anti-scatter.

Raman spectroscopy

Crystals of brushite were placed on a polished metal surface on the stage of an Olympus BHSM microscope, which is equipped with 10x, 20x, and 50x objectives. The microscope is part of a Renishaw 1000 Raman microscope system, which also includes a monochromator, a filter system and a CCD detector (1024 pixels). The Raman spectra were excited by a Spectra-Physics model 127 He-Ne laser producing highly polarised light at 633 nm and collected at a nominal resolution of 2 cm^{-1} and a precision of ± 1 cm^{-1} in the range between 100 and 4000 cm^{-1}. Repeated acquisition on the crystals using the highest magnification (50x) was accumulated to improve the signal to noise ratio in the spectra. Spectra were calibrated using the 520.5 cm^{-1} line of a silicon wafer.

Infrared spectroscopy

Infrared spectra were obtained using a Nicolet Nexus 870 FTIR spectrometer with a smart endurance single bounce diamond ATR cell. Spectra over the 4000–525 cm^{-1} range were obtained by the co-addition of 128 scans with a resolution of 4 cm^{-1} and a mirror velocity of 0.6329 cm/s. Spectra were co-added to improve the signal to noise ratio. The infrared spectra are provided in the supplementary information.

Band component analysis was undertaken using the Jandel 'Peakfit' (Erkrath, Germany) software package which enabled the type of fitting function to be selected and allowed specific parameters to be fixed or varied accordingly. Band fitting was done using a Lorentz-Gauss cross-product function with the minimum number of component bands used for the fitting process. The Lorentz-Gauss ratio was maintained at values greater than 0.7 and fitting was undertaken until reproducible results were obtained with squared correlations (r^2) greater than 0.995. Band fitting of the spectra is quite reliable providing there is some band separation or changes in the spectral profile.

Results and discussion

X-ray diffraction

The XRD patterns of the synthesised brushite and the Cave mineral brushite are displayed in the supplementary Information in Figs. S1a and S1b respectively. The XRD pattern of the brushite matches that of the reference pattern exactly. The XRD of the Cave mineral brushite clearly proves that the collected mineral sample was indeed brushite. Traces of calcite and quartz may also be observed but are minor components. The peak at around 45 degrees two theta is due to the presence of paraffin wax.

Raman Spectroscopy

The Raman spectra of synthesised brushite and the Cave mineral in the 650 to 1200 cm^{-1} region are shown in Fig. 1a and in Fig. 1b respectively. The infrared spectrum in the 500 to 1250 cm^{-1} region of the synthesised brushite and the natural brushite are displayed in Figs. S2a and S2b. For the synthetic brushite four Raman bands are observed at 987, 1000, 1063 and 1085 cm^{-1}, While for the Cave mineral, three bands are observed at 985, 1000 and 1055 cm^{-1}. It is noted that there are some differences in the position f the bands between the brushite analogue and the Cave mineral. Perhaps this is not unexpected as the Cave brushite may have some minor partial substitution.

These bands are assigned as follows: 987 cm^{-1} ν_1 PO_4^{3-}, 1000 cm^{-1} ν_1 HPO_4^{2-}, and 1063 and 1085 cm^{-1} ν_3 PO_4^{3-} stretching modes. In the infrared spectrum (Fig. S2a.) of the synthesised brushite, these bands are observed at 987, 1004 and 1061 cm^{-1}. The first two bands display considerable band overlap. In addition, infrared bands are observed at 1118 and 1137 cm^{-1} and are assigned to the ν_3 HPO_4^{2-} antisymmetric stretching modes. For the Cave mineral, infrared bands are observed in similar positions. An intense band is observed at 985 cm^{-1} with a shoulder at 1002 cm^{-1}. Other infrared bands are found at 1056, 1114, 1131 and 1151 cm^{-1}.

An infrared band for synthetic brushite is observed at 1207 cm^{-1}. For the natural brushite, two bands at 1200 and 1219 cm^{-1} are observed. These bands are attributed to the OH in-plain deformation modes of HPO_4^{2-} units. Farmer [18] reported infrared bands at 1200 and 1215 cm^{-1} for brushite and allocated these bands to δ in-plain OH deformations. Indeed the infrared

spectrum of brushite based upon the work of Petrov [20-22] is identical to the infrared spectrum of synthetic brushite reported in this work.

The Raman spectrum of synthetic brushite in the 300 to 650 cm^{-1} region is displayed in Fig. 2a. The Raman spectrum of Cave brushite in the low wavenumber region is displayed in Fig. 2b. For the synthetic brushite two bands are found at 577 and 587 cm^{-1}. A low intensity band at 576 cm^{-1} is found for the Cave brushite. The Raman bands at 577 and 587 cm^{-1} are assigned to the ν_4 PO_4^{3-} bending mode. It should be kept in mind that the natural brushite occurs as a powdered deposit on clacite surfaces. Obviously there will be intensity differences when compared with a highly crystalline synthetic analogue. Intensity differences can arise from the crystallite orientation and polarization. This vibrational mode (ν_4 PO_4^{3-} bending mode) is observed in the infrared spectrum at 579 cm^{-1} (Fig. S2a). The band is found at 577 cm^{-1} for the Cave brushite (Fig. 2b).

The band occurs in different positions for the hydrogen phosphate anion. This ν_4 band for the HPO_4^{2-} anion for the synthetic brushite occurs at 524 and 541 cm^{-1}. The band is observed at 519 cm^{-1} for the Cave brushite. These bands are not observed in the infrared spectrum as the cell absorbs all the radiation below 550 cm^{-1}. Ardealite $Ca_2(HPO_4)(SO_4)\cdot 4H_2O$ is a mineral which also contains the hydrogen phosphate anion. Intense Raman bands are found at 505 and 528 cm^{-1} for ardealite and are assigned to the ν_4 $(HPO_4)^{2-}$ bending modes. It is probable that the bending modes of OPO and HOPO bending modes occur at the same energy, thus two distinct bands are not observed. The mineral newberyite $Mg(PO_3OH)\cdot 3H_2O$ also contains a hydrogen phosphate anion. The Raman spectrum of this mineral has been reported [23]. An intense band was observed for this mineral at 498 cm^{-1}. Rajendran and Keefe [24] reported Raman bands at 521 and 586 cm^{-1} and assigned these bands to (H-O)P=O absorption bands. This assignment is in contrast to the assignment of bands by Ross [18].

For the synthetic brushite, Raman bands are found at 379 and 411 cm^{-1} and are assigned to ν_2 PO_4^{3-} bending modes. For the Cave mineral a Raman band is found at 411 cm^{-1}. Farmer [18] based upon the work of Petrov *et al.* reported these bands at 400 and 418 cm^{-1} which is in agreement with the assignations of this work. It is interesting that these vibrations for the

aqueous HPO_4^{2-} anion occur at much lower wavenumbers (278 and 318 cm^{-1}). In the infrared spectrum, bands are observed at 795 cm^{-1} with shoulder bands at 734 and 860 cm^{-1}. These bands are assigned to the out-of-plane vibrations of POH units. Farmer [18] tabled this band at 750 cm^{-1}.

The Raman spectrum of the synthesised brushite analogue and the natural Cave mineral in the far low wavenumber region are displayed in Figs. 3a and 3b respectively. The synthetic brushite in this spectral region is characterised by an intense band at 205 cm^{-1}. The band is assigned to a librational mode and shows sensitivity to disorder. The band is broad and of low intensity for the natural brushite. This disorder can arise through a partial substitution of calcium ions by other ions for example Mg^{2+} and H^+.

The bands at 879 cm^{-1} for synthetic brushite (Figure 1a) and at 872 cm^{-1} (Figure 2b) for the Cave brushite are assigned to the symmetric stretching vibrations of the POH units. Ross in Farmer [18] reported this band at 875 cm^{-1}. A Raman band is found at 862 cm^{-1} for ardealite. The two infrared bands at 651 and 672 cm^{-1} for synthetic brushite may be due to water librational modes. The bands are observed at 649 and 669 cm^{-1} for Cave brushite.

The position of this band is in harmony with the band position of the aqueous hydrogen phosphate anion at 891 cm^{-1} [25]. This band is assigned to the ν_1 HPO_4^{2-} symmetric stretching mode. The band is due to the presence of hydrogen phosphate units in the ardealite structure. A Raman band was observed in the spectrum of newberyite $Mg(PO_3OH)\cdot 3H_2O$ at 887 cm^{-1} and is attributed to HPO_4^{2-} symmetric stretching mode [23]. Such bands are normally intense in the infrared spectrum but of low intensity in the Raman spectrum. Rajendran and Keefe [24] studied the growth and characterisation of calcium hydrogen phosphate dihydrate and reported the FTIR and Raman spectra. These researchers observed a band at 875 cm^{-1} and assigned this band to the HPO_4^{2-} POP antisymmetric stretching mode. However, this assignment differs from the interpretation reported in this work.

The Raman spectrum of synthetic brushite in the 3000 to 3800 cm^{-1} region and the Raman spectrum of Cave brushite are shown in Figs. 4a and 4b respectively. In the Raman spectrum, four bands are observed at 3166, 3265, 3470 and 3540 cm^{-1}. The bands are assigned to the

symmetric stretching modes of water. Two Raman bands are observed for Cave brushite at 3472 and 3533 cm^{-1}. Other bands may be present but the signal to noise ratio is low and consequently other water bands could not be identified. The infrared spectrum of synthetic brushite in the 2500 to 4000 cm^{-1} region and the infrared spectrum of Cave brushite in the 3000 to 3700 cm^{-1} region are shown in Figs. S3a and S3b respectively. In the infrared spectrum of synthetic brushite, bands are found at 3157, 3275, 3477 and 3539 cm^{-1}. Additional infrared bands for synthetic brushite are observed at 2972, 3157, 3275 and 3384 cm^{-1} and are also assigned to water stretching bands. For the Cave brushite, infrared bands are observed at 3154, 3269, 3475 and 3537 cm^{-1}. The infrared spectra are in harmony with the Raman data which clearly shows the existence of non-equivalent water molecules in the brushite structure.

Jones and Smith [26] reported the OH wavenumbers for the related mineral monetite. Berry and Baddiel [27,28] interpreted the infrared spectrum of brushite in terms of the hydrogen phosphate anion and proposed that the infrared spectrum was based upon the occupation of C_2 sites by HPO_4^{2-} ions. Ross in Farmer [18] stated that the two possible space groups are C^4_s and C^6_{2h}. The first space group is favoured and the centrosymmetric space group requires all eight water molecules in brushite to be equivalent. The infrared spectrum of brushite clearly shows multiple water bands involving different hydrogen bond strengths, thus indicating the water molecules are not all equivalent in the brushite structure. Crystallographic studies show that there are five crystallographic independent hydrogen atoms in the crystal structure of brushite with z=4 in the non-centrosymmetric monoclinic space group Ia [29]. In the structure, two water molecules coordinate differently with different hydrogen bond strengths as indicated by the hydrogen bond distances [29].

Mechanism of formation of brushite

Brushite is formed on calcite surfaces in the Moorba Caves. The source of the phosphate ion is bat guano and is probably present as an ammonium hydrogen phosphate or related chemical. The following reaction is proposed:

227 $(NH_4)_2HPO_4 + Ca^{2+} + 2H_2O \rightarrow CaHPO_4{\cdot}2H_2O$ (brushite) $+ 2NH_4^+$

228 Alternatively the equation may be written as

229 $HPO_4^{2-} + Ca^{2+} + 2H_2O \rightarrow CaHPO_4{\cdot}2H_2O$ (brushite)

230 The calcium ions originate from the dissolution of calcite. Carbonate ions function as an
231 inhibitor of the calcium phosphates. This fact suggests that brushite is formed in solution and
232 is translocated through the Cave system and then precipitates from solution, as is shown in
233 the above equation.

234 For the synthesised brushite the following reaction is proposed:

235 $Na_2HPO_4^{2-} + Ca^{2+} + 2\ Cl^- + 2H_2O \rightarrow CaHPO_4{\cdot}2H_2O$ (brushite) $+ CaCl_2$

236

237 The mineral brushite is formed in renal canals and urinary calculus [6,7,27,30,31]. The proposed
238 chemical reaction above is a mechanism for the formation of brushite in these renal and
239 urinary calculus [32-34]. It is probable that the concentration of calcium ions effects the
240 formation of brushite. High concentrations of Ca^{2+} favours brushite formation, as is found in
241 Caves such as in the Moorba and Jenolan Caves.

242

243 **Conclusions**

244 Brushite was synthesised in order to match the chemical reaction that occurs in Cave systems
245 through the reaction of bat guano with calcite. Brushite is a mineral of significance because
246 of its formation in the urinary tract. No doubt this type of reaction occurs in the urinary tract.
247 The XRD patterns of both the synthesised brushite and the Cave mineral brushite show
248 identical patterns to the standard brushite XRD pattern. The Cave mineral brushite contains
249 minerals other than brushite whereas the synthesised brushite is pure. A mechanism of
250 formation of brushite in Caves is proposed. This mechanism is probably the same type of
251 chemical reaction that occurs in renal canals.

252 Even though the Raman spectrum of the Cave mineral brushite suffers from a lack of signal
253 to noise, the Raman spectrum of the Cave mineral brushite closely resembles that of the
254 synthesised mineral.

Acknowledgments

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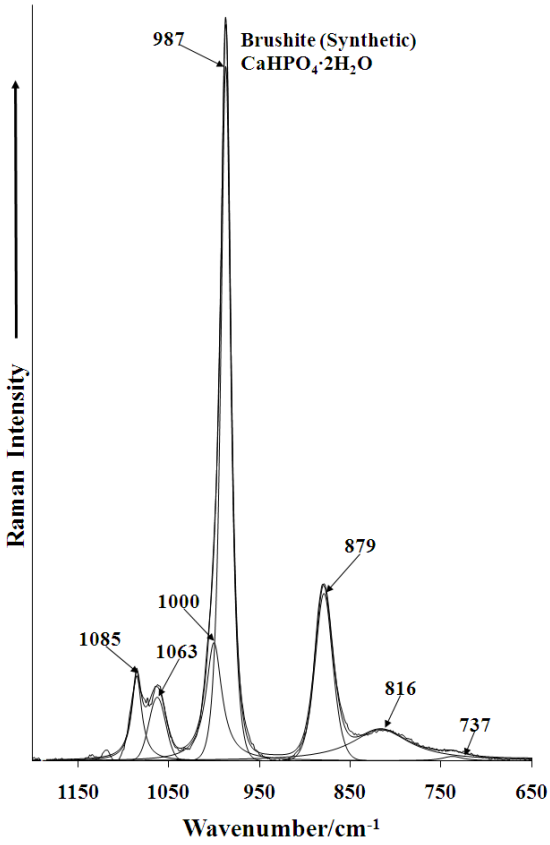
List of Figures

338



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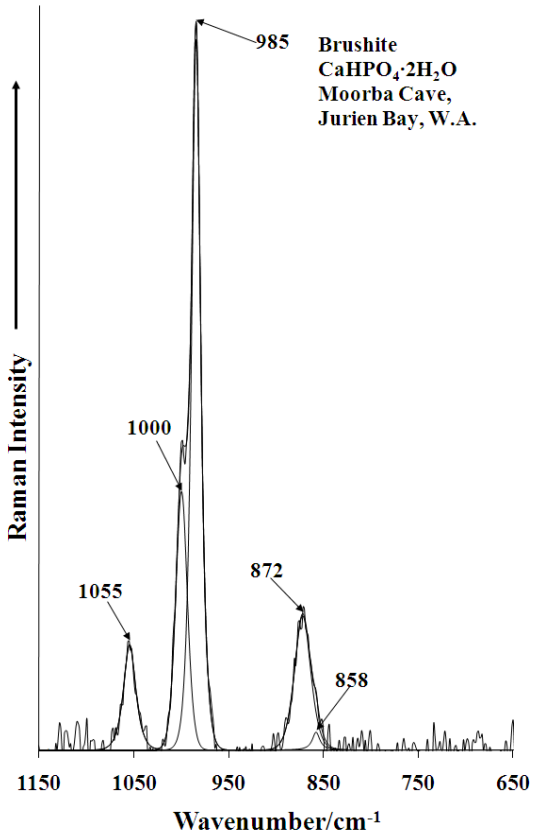

Figure 1b

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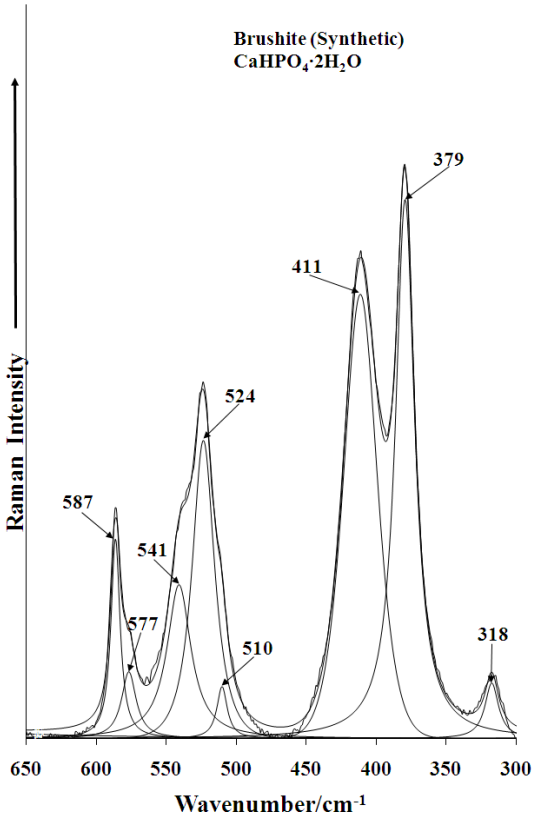

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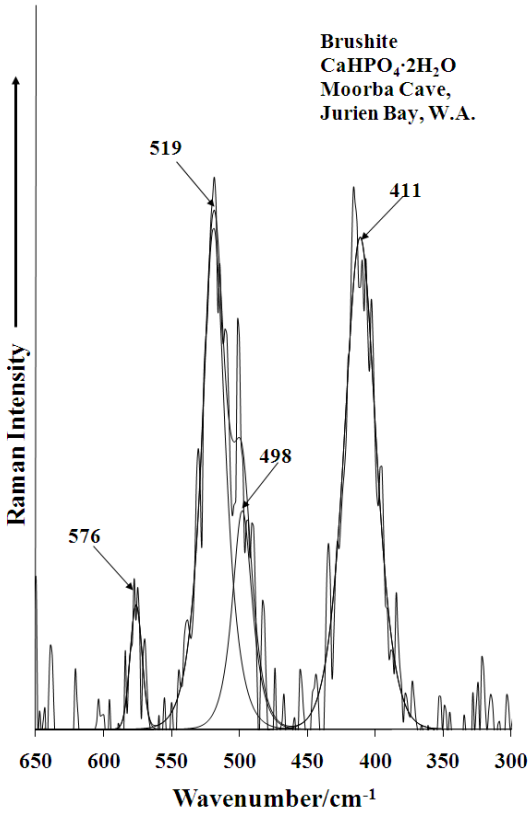

Figure 2b

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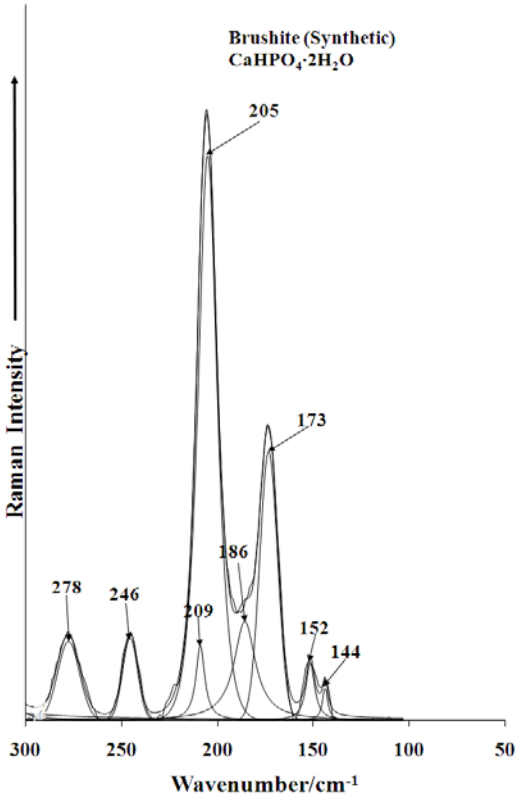

Figure 3a

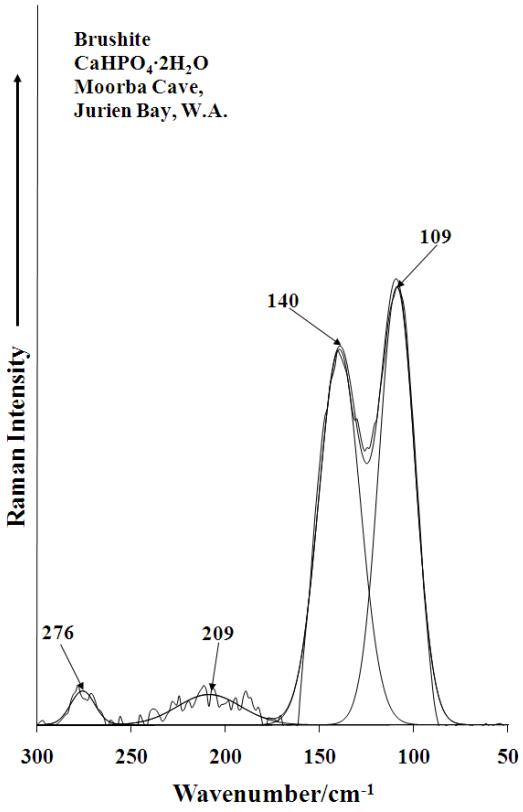

Figure 3b

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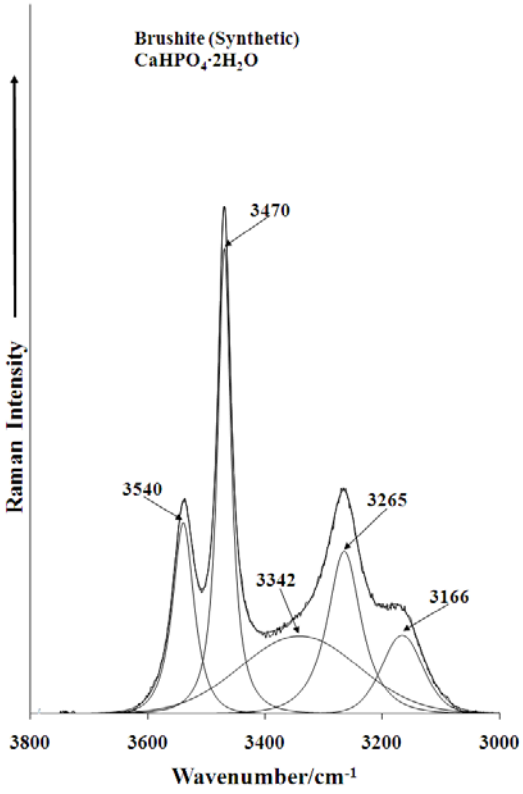

Figure 4a

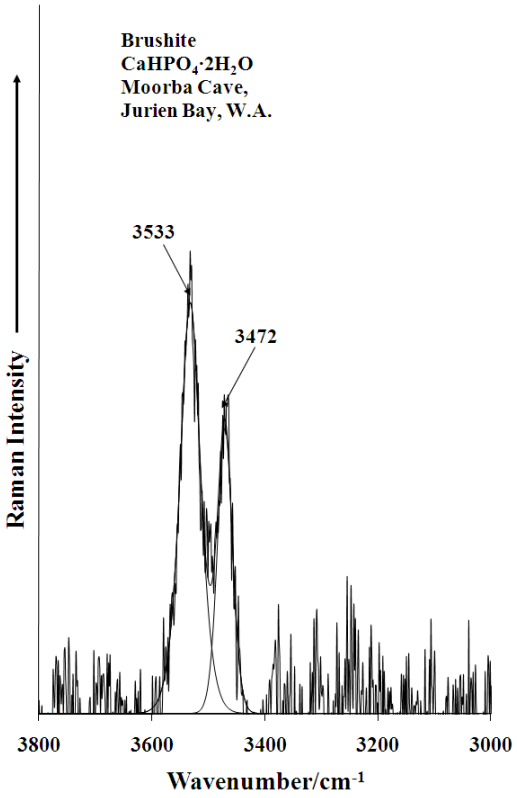

Figure 4b

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