

with small vacuoles, Ψ_m is small relative to Ψ_p and Ψ_s . If we disregard Ψ_m , Eq. 5 becomes:

$$\Psi_{\text{cell}} = \Psi_s + \Psi_p \quad (6)$$

It will be recalled that DPD was defined by Meyer (2) as:

$$\text{DPD} = \text{OP} - \text{TP} \quad (7)$$

To compare the terms of Eq. 6 and Eq. 7, we express the potentials in Eq. 6 as their algebraically equivalent pressures: $\Psi_s = -\text{OP}$, $\Psi_p = \text{TP}$, and $\Psi_{\text{cell}} = -(\text{OP} - \text{TP})$. Thus, it can be seen that Ψ_{cell} or Ψ_w and DPD are numerically equal but are opposite in sign, that is,

$$\Psi_w = -\text{DPD} \quad (8)$$

The change from DPD to Ψ_w is relatively simple except for the fact that Ψ_w is a negative number and must be thought of as becoming lower with increasing water stress. Thus, water potential (Ψ_w) increases in magnitude but decreases in absolute value (becomes more negative) as water stress increases. The same situation occurs in reading temperatures below zero on a thermometer. For example, -5°C is a higher temperature than -10°C and -5 bars is a higher potential than -10 bars.

The potential terminology can be applied to soil water. Total potential of soil water includes: osmotic potential (Ψ_s), gravitational potential (Ψ_g), matric or capillary potential (Ψ_m), and potential caused by external gas pressure (Ψ_p).

$$\Psi_{\text{soil}} = \Psi_s + \Psi_g + \Psi_m + \Psi_p \quad (9)$$

However, the *total soil-moisture stress* of certain investigators (5) refers only to the matric and osmotic forces, hence:

$$\Psi_{\text{soil}} = \Psi_m + \Psi_s = -\text{TSMS} \quad (10)$$

The total free energy level or water potential of plant tissue or soil can be determined by measuring its vapor pressure (6). Measurements made on soil with pressure plates or pressure membranes indicate only the matric or capillary potential (Ψ_m) of the soil water.

In summary, terms such as DPD and Ψ_w are merely names applied for convenience to the basic property of water known as its chemical potential, which is an expression of its free energy status. Ψ_w can be regarded as a measure of the driving force which

causes water to move into any system (7), such as plant tissue or soil, or from one part of the system to another.

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31 May 1966

Infrared Study of the Hydroxyl Bands in Clinoamphiboles

Abstract. Sharp single peaks in the fundamental and first-overtone bands of the O-H stretching vibration in pure Mg^{2+} and Fe^{2+} amphiboles split into a maximum of four sharp peaks, corresponding to hydroxyl groups linked to 3 Fe, 2 Fe + Mg, 2 Mg + Fe, and 3 Mg, in mixed Fe^{2+} - Mg^{2+} amphiboles. Within any one solid-solution series, the frequencies of these peaks can be correlated with the electronegativity of ions in the M_1 and M_3 positions, and differences between series can be correlated with the size of ions in the M_4 position. The O-H vector lies approximately normal to z in the (010) plane. The distribution of Fe^{2+} and Mg^{2+} ions between the (M_1, M_3) and (M_2, M_4) positions in the cummingtonite-grunerite series, and between the (M_1, M_3) and M_2 positions in the tremolite-ferroactinolite series, has also been estimated.

The observation (1) of possible overtones of the O-H stretching vibration as strongly polarized bands in the near-infrared spectra of certain amphiboles led us to investigate both the fundamental and first-overtone regions of this vibration. Minerals of the tremolite-ferroactinolite [$\text{Ca}_2(\text{Mg,Fe})_5\text{Si}_8\text{O}_{22}(\text{OH})_2$] and cummingtonite-grunerite [$(\text{Mg,Fe})_7\text{Si}_8\text{O}_{22}(\text{OH})_2$] series were chosen initially because of their simple chemistry.

The fundamental band (3600 to 3700 cm^{-1}) was recorded on a Perkin-Elmer model 337 spectrophotometer modified for ordinate and abscissa expansions, the KBr disc method being used. Spectra of the polarized first-overtone bands at 7050 to 7200 cm^{-1} were recorded on a Cary model 14 spectrophotometer with orientated thin (30 μ) sections by means of a polarizing-microscope technique (2). This technique was developed further to enable the indicatrix orientation to be determined over the whole wavelength range from 4000 to 22,000 Å. Measurements were made at room temperature on five members of the cummingtonite-grunerite series with 35 to 98 percent Fe^{2+} , and on four members of the tremolite-ferroactinolite series with 0 to 48 percent Fe^{2+} (all percentages mentioned in this report are 100 $\text{Fe}^{2+}/(\text{Fe}^{2+} + \text{Mg}^{2+})$).

The fundamental band (Fig. 1)

shows a single sharp peak (A) in iron-free tremolite. Additional peaks (B, C, D) appear at lower frequencies as the ferroactinolite content increases, and peak A simultaneously decreases in intensity. There is no significant wavelength shift of individual peaks with Fe^{2+} concentration analogous to that observed in the electronic spectra (1). The fundamental band in grunerite with 98 percent Fe^{2+} consists of a single sharp peak (D') with a small pip (C') on the high-frequency side. With increasing Mg content, C' first grows, and then B' and A' appear and continue to grow, with D' and later C' declining in importance (Fig. 1). The variation of intensities with composition is similar to that seen in the tremolite-ferroactinolite series, and there is again no significant shift of individual peaks as the ratio of Fe to Mg varies.

The overtone band shows similar features (1). Single, sharp peaks occur at 7180 and 7068 cm^{-1} in tremolite and grunerite, respectively, with additional peaks in mixed Fe-Mg amphiboles (Table 1). In the actinolite with 15 percent Fe^{2+} , the intensity ratios for light vibrating along α , β , and γ are $A_\alpha = 22$; $A_\beta = 0$; $A_\gamma = 5$; and the extinction angle γ/z is $19^\circ \pm 2^\circ$ in the obtuse angle beta. In both the fundamental and the overtone bands, the four peaks in the tremolite-ferroactino-

lite series occur at wave numbers higher (4 to 13 cm^{-1}) than the corresponding peaks in the cummingtonite-grunerite series (Table 1).

The fundamental band represents the transition from the ground state to the first excited state ($\nu=0 \rightarrow \nu=1$) which is allowed by the selection rules for a harmonic oscillator-type system (3). The overtone at 7050 to 7200 cm^{-1} represents the forbidden transition $\nu=0 \rightarrow \nu=2$, which is intensified by anharmonicity (departure of the potential energy curve from a parabola). The hydroxyl group in the amphibole structure lies in a large cylindrical cavity formed by the linking of two Si_6O_{18} rings, and closed at each end by $(\text{Fe}, \text{Mg})_3\text{OH}$ groups. This arrangement is very similar to that in many sheet silicates. The O-H bond projects toward the central void away from the metal ions. The proton, therefore, lies in a highly noncentrosymmetric force field, which accounts for the anharmonicity. The observed intensity ratio of fundamental and overtone bands is approximately 40/1, and the anharmonicity factor $2\omega x$ is about 160 cm^{-1} . The narrow bandwidth of 11 cm^{-1} at half-height reflects the relative isolation of the OH group from perturbing influences.

Each hydroxyl group in the amphibole structure is coordinated to three cations, two of which are in crystallographically equivalent M_1 positions, with the third in the M_3 position (4, 5). In the tremolite-ferroactinolite and cummingtonite-grunerite series, Mg and Fe^{2+} ions occupy these positions. There are eight distinguishable ways of distributing Fe and Mg over three inequivalent positions, reduced to six by the equivalence of the two M_1 positions, and further reduced to four by the pseudo-trigonal symmetry of the $(M_1M_1M_3)\text{OH}$ groups. These four distinguishable distributions may be designated $(\text{MgMgMg})^1$, $(\text{MgMgFe})^3$, $(\text{FeFeMg})^3$, and $(\text{FeFeFe})^1$, where the superscripts refer to the numbers of indistinguishable possibilities belonging to each distribution. The manner in which the intensities of peaks A, B, C, and D vary with the ratio of Fe to Mg in the two series demonstrates that $A = (\text{MgMgMg})^1$, $B = (\text{MgMgFe})^3$, $C = (\text{FeFeMg})^3$, and $D = (\text{FeFeFe})^1$. The shift to lower frequencies along the series A, B, C, D is related to the different electronegativities of Fe^{2+} (1.8) and Mg (1.2). Metal-oxygen bonds are strengthened, and hence the hydrogen-oxygen bond is weakened, when Fe^{2+}

replaces Mg in the M_1 and M_3 positions. The higher frequency of peaks in the tremolite-ferroactinolite series, relative to those of analogous peaks in the cummingtonite-grunerite series, probably results from compression of the Si_6O_{18} ring when (Mg,Fe) in the M_4 position is replaced by the larger Ca^{2+} ion. The

analogy between the hydroxyl environment in clinoamphiboles and that in certain sheet silicates is strengthened by the ready replacement of OH^- by F^- in both amphibole and mica structures. Splitting of the hydroxyl stretch fundamental by Fe^{2+} -Mg substitution has been observed in talc (6), and the

Table 1. Peak positions of the fundamental and first-overtone O-H stretching bands in amphiboles.

Tremolite-ferroactinolite series				Cummingtonite-grunerite series			
<i>Fundamental peak maxima (cm^{-1})</i>							
(A) 3673	(B) 3660	(C) 3648	(D) 3625	(A') 3665	(B') 3650	(C') 3635	(D') 3615
<i>First-overtone peak maxima (cm^{-1})</i>							
7180	7147	7120	7078	7172	7143	7112	7068
<i>Anharmonicity, $2\omega x$ (cm^{-1})</i>							
166	173	176	172	158	157	158	162
<i>Coordinating group to OH ($M_1M_1M_3$)</i>							
MgMgMg	MgMgFe MgFeMg FeMgMg	FeFeMg FeMgFe MgFeFe	FeFeFe	MgMgMg	MgMgFe MgFeMg FeMgMg	FeFeMg FeMgFe MgFeFe	FeFeFe

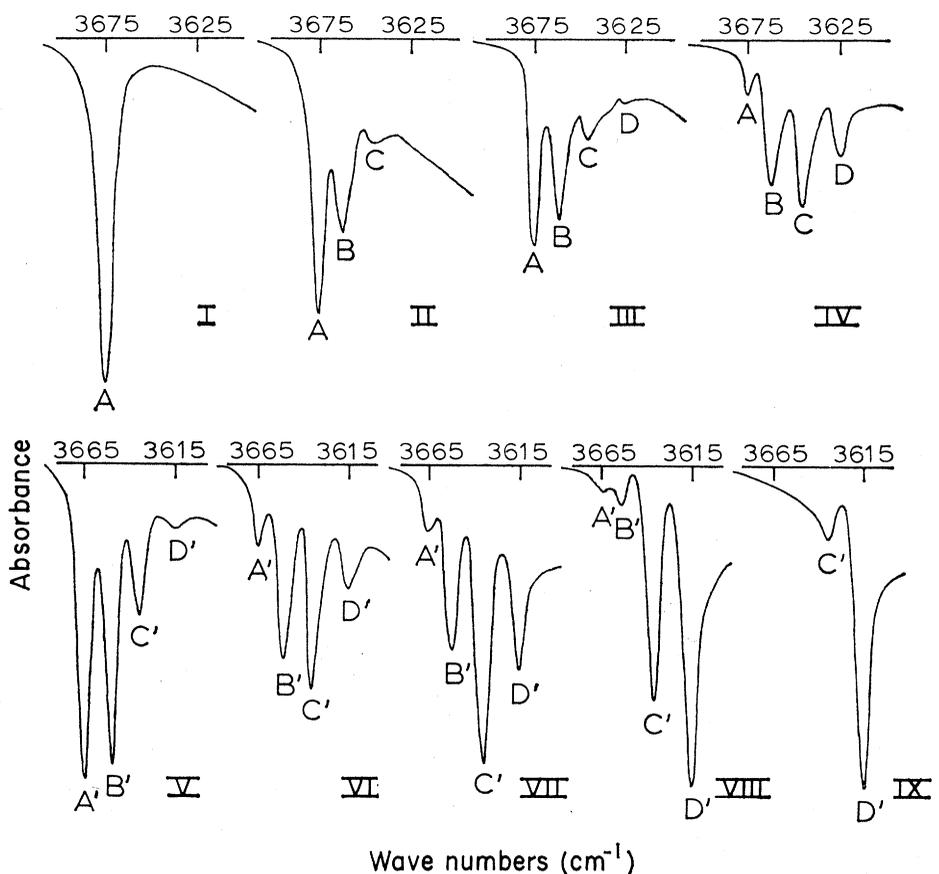


Fig. 1. The fundamental O-H stretching band in amphiboles. I to IV, The tremolite-ferroactinolite series; V to IX, The cummingtonite-grunerite series. I, Tremolite, 0 percent Fe^{2+} (analyst: Strens; by optics); II, actinolite, 15.2 percent Fe^{2+} (analyst: Burns; by electron microprobe; Berkeley Collection No. 14785); III, actinolite, 30.0 percent Fe^{2+} (analyst: Burns; by electron microprobe; U.S. National Museum No. 80714); IV, actinolite, 47.9 percent Fe^{2+} [analyst: Mueller (9); No. 12BA]; V, cummingtonite, 36.0 percent Fe^{2+} (analyst: Burns; by electron microprobe; U.S. National Museum No. 118125); VI, cummingtonite, 53.4 percent Fe^{2+} [analyst: Mason (10)]; VII, cummingtonite, 65.1 percent Fe^{2+} [analyst: Klein (6); No. 7]; VIII, grunerite, 88.8 percent Fe^{2+} [analyst: Klein (8); No. 1]; IX, grunerite, 98.0 percent Fe^{2+} [analyst: Mueller (9); No. 1B]. Percentages are 100 $\text{Fe}^{2+}/(\text{Fe}^{2+} + \text{Mg}^{2+})$.

Table 2. Distribution of Fe²⁺ in clinoamphiboles.

Specimen	Fe ²⁺ (%)	Total Fe (per formula unit) (%)	Position of Fe			
			In <i>M</i> ₁ , <i>M</i> ₃	Per <i>M</i> ₁ , <i>M</i> ₃	In <i>M</i> ₂ or <i>M</i> ₄	Per <i>M</i> ₂ or <i>M</i> ₄
<i>Tremolite-ferroactinolite series</i>						
Berkeley 14785	15.2	0.76	0.32	0.11	0.44	0.22
USNM 80714	30.0	1.50	.62	.21	.88	.44
Mueller, 12BA	47.9	2.40	1.56	.52	.84	.42
<i>Cummingtonite-grunerite series</i>						
USNM 118125	36.0	2.52	0.72	0.24	1.80	0.45
Mason	53.4	3.74	1.50	.50	2.24	.56
Klein, 7	65.1	4.56	1.96	.65	2.60	.65
Klein, 1	88.8	6.22	2.56	.85	3.66	.92
Mueller, 1B	98.0	6.86	2.95	.98	3.91	.98

polarization-dependence of the hydroxyl peak in phlogopite (6) is similar to that in the amphiboles.

The absence of overtone contributions in the polarized β spectra indicates that the O-H bond lies in the (010) plane. Its direction in this plane may be calculated from the observed ratio of absorbances for light vibrating $\parallel\alpha$ and $\parallel\gamma$ by applying the relation

$$\sin^2 \theta / \cos^2 \theta = A_\alpha / A_\gamma$$

where θ is the angle between α and the O-H bond. From the ratio $A_\alpha / A_\gamma = 22/5$, the value of θ is $24^\circ \pm 6^\circ$. The angle γ/z at the overtone frequency is $19^\circ \pm 2^\circ$ in the obtuse angle beta ($\beta = 105^\circ$), so that the O-H bond makes an angle of $[90 - (24 \pm 6 - 19 \pm 2)] = 85^\circ \pm 8^\circ$, with z in the (010) plane. Inspection of the structure model suggests that the bond is, in fact, normal to z . If this is so, sections of tremolite (which is obtainable in large single crystals) normal to z would be very efficient polarizers over narrow bands centered on 3673 and 7180 cm^{-1} .

The relative intensities of peaks *A*, *B*, *C*, and *D* may be used to estimate the distribution of Fe²⁺ between the *M*₂ and (*M*₁,*M*₃) positions in the tremolite-ferroactinolite series, and between the (*M*₂,*M*₄) and (*M*₁,*M*₃) positions in the cummingtonite-grunerite series, provided the total iron content is known.

$$\Sigma \text{Fe}^{2+} (M_1M_3) = 0 \cdot A + 1 \cdot B + 2 \cdot C + 3 \cdot D$$

$$\Sigma \text{Mg}^{2+} (M_1M_3) = 3 \cdot A + 2 \cdot B + 1 \cdot C + 0 \cdot D$$

The proportion of Fe²⁺ ions in (*M*₁,*M*₃) positions is $3\Sigma \text{Fe} / (\Sigma \text{Fe} + \text{Mg})$, and the number in *M*₂ or (*M*₂,*M*₄) positions is obtained by difference (Table 2). The results indicate that the Fe/(Fe + Mg) ratio in the *M*₂ position is approximately twice that in the (*M*₁,*M*₃) positions in actinolites with up to 30 percent Fe²⁺, and that the (*M*₂,*M*₄) posi-

tions are similarly favored in the cummingtonite-grunerite series. Similar conclusions were reached by less direct x-ray methods (4, 7).

The spectrum of a manganiferous cummingtonite (8) showed no evidence of the presence of Mn²⁺ in the (*M*₁,*M*₃) positions, but all peaks were shifted to higher frequencies by 2 to 5 cm^{-1} , suggesting that the large Mn²⁺ ion, like Ca²⁺, enters the *M*₄ position. In chemically more complex amphiboles, increasing numbers of peaks of ever-decreasing intensity appear, and in common hornblende the O-H fundamental is a single, broad, jagged band.

Determination of the orientation of the O-H bond from overtone bands (when present) rather than the funda-

mental has the advantage that conventional petrographical thin sections can be used, enabling minerals with good cleavage to be studied in any orientation. Previous single-crystal work with the fundamental band has been largely confined to minerals of which suitable cleavage flakes could be obtained, and orientations were restricted to zero or low-tilt angles (6). The microscope technique should prove applicable to a wider variety of minerals, although limited to those with overtone bands.

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2 May 1966

Selection of Sucrose-Dependent *Escherichia coli* To Obtain Envelope Mutants and Fragile Cultures

Abstract. *Mutants isolated as sucrose-dependent include many with apparent envelope defects, which fact often leads to filamentous growth or lysis in the absence of sucrose. One mutant can grow exponentially in 4 percent sucrose, but is very fragile: it releases all of its RNA and soluble protein when treated with 0.5 percent sodium deoxycholate. These characteristics permit the study of unstable structures and rapid processes in actively growing cells.*

Since the basis of bacterial rigidity is the cell wall and since 10 to 20 percent sucrose can stabilize otherwise unstable spheroplasts (1), it seemed probable that there is a class of mutants, with defects of the cell wall, which is dependent on high concentrations of sucrose for growth, and that some of these mutants would be fragile in its absence. We isolated such mutants.

Escherichia coli, strains K12 3000 B₁⁻ and AB 301 *met*⁻, or a derivative of AB 301 constitutive for alkaline phosphatase, were grown exponentially at 37°C in trypticase soy broth to 2×10^8

cells per milliliter. Cultures were then treated with 30 $\mu\text{g}/\text{ml}$ of the mutagens ethylmethane sulfonate or *N*-methyl-*N'*-nitro-*N*-nitroso-guanidine for 30 minutes, and samples were diluted and plated on nutrient agar containing 20 percent sucrose. After replica plating on media with and without sucrose, colonies that grew only in the presence of sucrose were purified and tested in liquid medium.

Of 30 independent *sud* (sucrose-dependent) mutants that showed an absolute requirement for sucrose on plates, 22 required sucrose for growth