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May 1970

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Far-Infrared Magnetic Resonance in Fe(III) and Mn(III) Porphyrins, Myoglobin, Hemoglobin, Ferrichrome A, and Fe(III) Dithiocarbamates

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ABSTRACT

Far infrared spectroscopic techniques are used to study the magnetic resonance of Fe(III) and Mn(III) ions in molecular sites with large axial and rhombic fields. Measurements of transmission spectra over the range 3-100 cm^{-1} are discussed for a group of polycrystalline compounds, including several biological complexes, at temperatures between 1.3 and 50°K and in applied magnetic fields up to 50 kOe. The spectra show magnetic resonance absorptions which are consistent with a number of cases of the spin Hamiltonian formulation, and the spin Hamiltonian parameters ($D,E \gtrsim 1\ cm^{-1}$) are directly obtained from the spectra. The observation of resonances due to high spin Mn(III) and to the ferromagnetic resonance of $((C_2H_5)_2NCS_2)_2$ Fe(III)Cl is reported. These measurements show that present far-infrared techniques offer a direct method for the investigation of the effects of large ligand fields on paramagnetic ions in molecules.

I. INTRODUCTION

A number of powerful techniques have been applied to the investigation of the electronic properties of paramagnetic ions in molecules. Considerable information has been obtained from spectroscopic measurements in the microwave, near infrared, visible, and ultraviolet regions of the electromagnetic spectrum, and from Mössbauer resonance at higher frequencies. Although the detailed structure of the electronic spectrum is complex, it has frequently been found that the magnetic properties of the ground term could be adequately described by a simple Hamiltonian first proposed by Abragam and Pryce[1] in 1950. The simplicity of the description results from the interaction of the ion with the surrounding ligands. The ligand field splits the multiply-degenerate free-ion ground state, and the new ground term, which is often an orbital singlet and spin multiplet, is further split by second-order spin-orbit coupling. The ground term may then be described by the spin Hamiltonian

$$\mathcal{H} = \mu_B \, \underset{\sim}{H} \cdot \underset{\approx}{g} \cdot \underset{\sim}{S} + D[S_z^2 - S(S+1)/3] + E[S_x^2 - S_y^2] \qquad (1)$$

where $\underset{\sim}{H}$ is the applied magnetic field, $\underset{\approx}{g}$ is the g-tensor, $\underset{\sim}{S}$ the electronic spin, and D and E are parameters which describe the effects of axial and rhombic ligand fields, respectively.

Conventional electron paramagnetic resonance techniques have been widely used to investigate ions whose ground terms are approximately described by Eq. (1). Such measurements, on compounds for which the values of the ligand field parameters correspond to microwave frequencies, have demonstrated that this description is often adequate, and have obtained accurate values for D and E. However, there is a large class of interesting compounds where the values of the spin Hamiltonian parameters are considerably larger ($D, E \gtrsim 1\ cm^{-1}$). This class of compounds contains many extensively-studied biological molecules, the most well-known of which are various derivatives of the hemoproteins, such as myoglobin and hemoglobin. In addition, it also includes a number of metal-organic complexes, such as the transition-metal porphyrins. Microwave measurements on these compounds have been interpreted in terms of Eq. (1), but although the observed resonances can be used to obtain estimates of the spin Hamiltonian parameters, they only depend upon D in second order, and are relatively insensitive to the effects of small modifications of the spin Hamiltonian. However, higher frequency magnetic resonance measurements, using far-infrared spectroscopic techniques, can obtain detailed information on the spin Hamiltonian for substances in this class.

We have measured the far-infrared transmission spectra of a group of such compounds containing Fe(III) and Mn(III). The measurements were made over the frequency interval 3-100 cm^{-1}. The samples were polycrystalline powders or frozen solutions at temperatures between 1.3 and 50°K, and in magnetic fields up to 52 kOe. The spectra show magnetic field dependent absorptions due to magnetic dipole transitions between states of the paramagnetic ground multiplet. Our experiments may be thought of as high frequency, high field elec-

tron paramagnetic resonance, with the exception that a continuous range of both frequency and field is available. The measurements have enabled us to investigate the validity of the spin Hamiltonian approximation, and to obtain directly values for the parameter D (and occasionally E).

In this paper, we shall present results for ferrichrome A, tris (pyrrolidyl dithiocarbamato) Fe(III), and certain complexes of ferrimyoglobin, ferrihemoglobin, Fe(III) porphyrins, Mn(III) porphyrins, and bis Fe(III) dithiocarbamates. This group of compounds is particularly well suited for such a general study because it contains illustrative examples for a variety of cases of Eq. (1). In terms of the spin Hamiltonian parameters, the cases investigated include: $S=3/2$, 2, and 5/2; $D > 0$ and $D < 0$; $E/D \ll 1$, and $E/D=0.25$; and $g\mu_B H/D \ll 1$ to $g\mu_B H/D \gg 1$. In addition, one of the compounds, $((C_2H_5)_2NCS_2)_2Fe(III)Cl$, is ferromagnetic, and several show spectra with strong magnetic field-independent absorptions which may be due to low-frequency molecular vibration modes. Our experiments on the manganese porphyrins have obtained the first magnetic resonance data for these complexes which can definitely be attributed to Mn(III). Finally, the results for myoglobin, hemoglobin, and ferrichrome A indicate that detailed information on the effects of the ligand field can be obtained from

investigation of intact biological molecules in the far-infrared region.

The organization of this paper is as follows: Section II includes a discussion of the relevant cases of the spin Hamiltonian of Eq. (1) and brief comments on the calculation methods used to analyze the data; Section III contains an outline of the experimental techniques employed; Section IV presents our experimental results for each compound together with some discussion.

II. THEORY

A. The Spin Hamiltonian

Excellent discussions of crystal or ligand field theory[2,3] and of more general forms of the spin Hamiltonian[1,4] are available, and a complete derivation of the spin Hamiltonian for ferrihemoglobin has been given by Weissbluth.[5] In this section, we shall confine our discussion to the cases of the spin Hamiltonian of Eq. (1) which are applicable to our study. Although the general theory is widely understood, the details of the predictions of Eq. (1) for specific cases may be unfamiliar.

We shall consider a simpler form of Eq. (1) which is sufficient to analyze our polycrystalline spectra:

$$\mathcal{H} = g\mu_B \underset{\sim}{H} \cdot \underset{\sim}{S} + D[S_z^2 - S(S+1)/3] + E[S_x^2 - S_y^2]. \qquad (2)$$

In Eq. (2), the $\underset{\approx}{g}$ tensor has been replaced by an isotropic g-factor with the free spin value g=2.00. Our results also do not require the small quartic terms in the spin operators which are occasionally included[6] in Eq. (1).

B. Eigenvalues and Eigenfunctions

The energy-level spectrum of Eq. (2) is distinguished by two general characteristics:

(i) In zero magnetic field, the eigenvalues of Eq. (2) are separated by "zero-field splittings", which are functions of the parameters D and E. For S=3/2 and S=5/2, the states are Kramers doublets. For S=2, the states are all singlets except in the case E=0 where there are two doublets and a singlet.

(ii) In a magnetic field, the eigenvalues are further split by the Zeeman interaction, and they are strongly dependent on both the magnitude and direction of the applied field.

The behavior of the spin Hamiltonian spectrum for each spin value can be most easily investigated in terms of the dimensionless parameters $\lambda=E/D$ and $H'=g\mu_B H/D$. Departures from axial symmetry are indicated by the value of λ. The range $0 \leqslant \lambda \leqslant 1/3$ describes all of the distinct physical possiblities; for $\lambda > 1/3$, a new set of coordinate axes may be chosen[7] in which the spin Hamiltonian has new parameters D' and E' and a value $\lambda' < 1/3$.

We have calculated the variation of the eigenvalues and eigenfunctions as a function of H' and λ for S=3/2, 2, and 5/2. Although the detailed behavior is complex, a few simple comments can be made which are very useful in the interpretation of our experimental spectra. In the following discussion, we shall use the eigenstates of S_z as a basis set.

The variation of the zero-field eigenvalues for each S as a

function of λ is given in Fig. 1. For $\lambda=0$ (E=0), the eigenfunctions of Eq. (2) are eigenstates of S_z with Kramers doublets characterized by the value of $|m_s|$. The zero field splittings are integer multiples of the axial crystal field parameter, D. For $D > 0$, the ground state is given by the minimum value of $|m_s|$; for $D < 0$, the level system is inverted. If λ is non-zero, the term $E[S_x^2 - S_y^2]$ couples states differing by $\Delta m_s = 2$. The eigenfunctions are no longer pure eigenstates of S_z and this admixture is reflected in a shift of the Kramers doublets for $S = 3/2$ and $S = 5/2$, and a splitting for $S = 2$ (Kramers' theorem does not hold for ions with an even number of electrons.) The zero-field splittings are therefore a function of λ, and this dependence can be used to obtain a value of λ as discussed below.

The eigenvalue spectrum as a function of H' for Eq. (2) with $\lambda=0$ is given for each S in Fig. 2. The eigenvalues depend upon the magnitude and orientation of the external field $\underset{\sim}{H}$ with respect to the coordinate system in which the spin Hamiltonian is written. For $\lambda=0$, only the polar angle θ_H of the field with respect to the z-axis is required to determine the spectrum, and we have given curves for $\theta_H = 0$ and $\pi/2$. (For $\lambda \neq 0$, the spectrum also depends upon the azimuthal angle ϕ_H, and can differ markedly from the curves shown, although the qualitative characteristics remain the same[8]).

For $\underset{\sim}{H} \parallel \hat{z}$ ($\theta_H = 0$), the eigenfunctions of Eq. (2) are eigenstates of S_z, and the Zeeman splitting is linear in H'. For $\underset{\sim}{H} \perp \hat{z}$ ($\theta_H = \pi/2$), the Zeeman term $g\mu_B \underset{\sim}{H} \cdot \underset{\sim}{S}$ mixes states differing by $\Delta m_s = \pm 1$. For $H' \ll 1$, this admixture produces a linear splitting of the zero field $m_s = \pm 1/2$ doublet for $S = 3/2$ and $5/2$, with an effective g-value which

depends upon S (for S=3/2, $g_\perp=4$; for S=5/2, $g_\perp=6$). The remaining doublets, for each S, split quadratically. For $H' \sim 1$, the eigenfunctions are mixtures of all eigenstates of S_z, and the variation of the eigenvalues with H' is complex. However, for large H', the eigenfunctions are nearly pure eigenstates of S_H, the projection of $\underset{\sim}{S}$ onto the magnetic field $\underset{\sim}{H}$, and the variation of all eigenvalues with H' then becomes linear. This behavior is completely analogous to the Zeeman effect in atomic multiplets split by the spin-orbit interaction, and the high field limit corresponds to the Paschen-Back effect. The limiting values of m_s and m_H for $\underset{\sim}{H} \parallel \hat{z}$ and $\underset{\sim}{H} \perp \hat{z}$ respectively are indicated on the figures for the case D > 0; for D < 0, the signs of m_s and m_H should be reversed.

The most important feature of the spectrum for powder spectroscopy is the large variation of the eigenvalues and eigenfunctions with the orientation of the applied magnetic field. In practice, this results in broad, complex absorption lineshapes.

C. Polycrystalline Absorption

The transmission spectra of samples containing paramagnetic ions whose ground multiplet is described by Eq. (2) show absorptions due to magnetic dipole transitions between the states discussed in the previous section. The observed spectrum for a single crystallite can be described by an absorption coefficient $\alpha(\nu)$, which is, in general, a function of D, E, H, θ_H, and ϕ_H, as well as of the direction of propagation and polarization of the incident radiation. For a given transition n between initial state $|i_n\rangle$ and final state $|f_n\rangle$, the absorption coefficient may be written[9]

$$\alpha_n(\nu) = \frac{4\pi^2 e^2 N_o}{\hbar m^2 c^3} \nu \, \rho(\nu-\nu_n) |\langle f_n | \hat{k} \times \hat{\pi} \cdot \underset{\sim}{S} | i_n \rangle|^2 \qquad (3)$$

where N_o is the concentration of paramagnetic ions, ρ is a lineshape function, $\hat{k}$ and $\hat{\pi}$ are unit vectors in the direction of propagation and electric field polarization respectively of the incident radiation, $\underset{\sim}{S}$ is the spin operator, and ν_n is the frequency corresponding to the difference of the eigenvalues of the initial and final states. In addition, at a given temperature T, the absorption coefficient must be multiplied by $P_n(T)$, the difference in the thermal population of the two states. The total absorption coefficient for a single crystallite is then

$$\alpha(\nu) = \sum_n \alpha_n(\nu) \, P_n(T) \qquad (4)$$

where the sum is over all transitions within the ground multiplet. The spectrum thus described will show a series of absorption lines, one for each transition.

The transmission spectrum of a polycrystalline specimen in a fixed magnetic field may be obtained by averaging Eq. (4) over all crystallite orientations. In our experiments, the incident radiation was unpolarized and was strongly scattered within the sample, which was placed within a low-Q transmission cavity. Under these conditions, the average over crystallite orientations reduces to three independent averages over the directions of polarization $\hat{\pi}$, propagation $\hat{k}$, and magnetic field $\underset{\sim}{H}$. In general, the first two averages may be carried out analytically, but the last must be performed numerically. (Later, we shall briefly discuss the important case $H' \gg 1$, in which the absorption coefficient may be

obtained in closed form.) After the averages over polarization and the direction of propagation have been calculated, the expression for the average of Eq. (4) may be written[9]

$$\alpha(\nu) = \frac{\pi N_o e^2 \nu}{\hbar m^2 c^3} \sum_n \int [\ \rho(\nu-\nu_n)\ P_n(T)\ W_n] d\Omega_H \tag{5}$$

where

$$d\Omega_H = \sin\theta_H d\theta_H d\phi_H$$

$$W_n \equiv 1/3\ \{|\langle S_x \rangle_n|^2 + |\langle S_y \rangle_n|^2 + |\langle S_z \rangle_n|^2\}$$

and

$$|\langle S_x \rangle_n|^2 \equiv |\langle f_n | S_x | i_n \rangle|^2.$$

The integral over the orientation of $\underset{\sim}{H}$ in Eq. (5) is most easily calculated by numerical methods because W_n and ν_n are in general not simple functions of the parameters of the spin Hamiltonian of Eq. (2).

The results of a calculation of $\bar{\alpha}$ depend upon a few simple properties of the integrand in Eq. (5). The factor of ν, which appears because α is defined as the power absorbed per unit length, enhances high frequency transitions. The term $P_n(T)$ strongly suppresses transitions from states elevated more than ~ kT above the ground state. Furthermore, at high temperatures, when $D/kT \ll 1$, $P_n(T)$ is very small for all transitions. The magnetic dipole transition probability W_n only allows transitions with $\Delta m_s = \pm 1, 0$. Finally, the solid angle $d\Omega_H$ strongly

enhances transitions corresponding to $H \perp z$.

As an example of the effects of $P_n(T)$ and W_n, we consider the absorption coefficient for $S = 5/2$ in zero applied field. For low temperatures, only transitions from the ground state will contribute appreciably. If $\lambda = 0$, only the transition from the ground state to the first excited state is allowed by the selection rules of W_n, and $\bar{\alpha}$ will show only one peak. However, if $\lambda \neq 0$, the admixture of states described previously will allow a transition to the second excited state. The strength of this second transition increases rapidly with λ, and for $\lambda \gtrsim 0.1$, two strong peaks will appear. The frequencies of the two corresponding experimental absorptions are sufficient to measure both D and λ.

If the temperature is sufficiently high to populate the first excited state, a third peak due to the transition between the first and second excited states will appear in $\bar{\alpha}$ for any value of λ; in particular, for small λ, the positions of the first and third peaks can be used to obtain D and λ. Therefore, the zero-field spectrum is sufficient in principle to determine the parameters of the spin Hamiltonian. We shall later discuss specific examples of these two cases. Similar observations may be made for the zero-field spectrum in the case $S = 2$, with the exception that the splitting of the doublets with λ provides additional information. However, for $S = 3/2$, measurement of the zero-field absorption coefficient is not sufficient to obtain values for D and λ, and it is necessary to measure the absorption spectrum in an applied field.

In order to compare our experimental spectra for $S = 3/2$ with the predictions of the spin Hamiltonian, we have written a program[9] to calculate $\alpha(\nu)$ for a specified D, E, H, T, and linewidth. A Gaussian

lineshape function, due to site inhomogeneities, was assumed, and the linewidth was chosen to fit the zero field spectrum. An example of the results of this calculation for $D < 0, |D|/kT = 0.8$, and a small value of λ are shown for several values of $H' = g\mu_B H/D$ in Fig. 3. The upper graph shows the contributions of the various transitions to the total lineshape, and the lower diagram is a composite plot of the variation of $\bar{\alpha}$ with H'. These curves illustrate the complex nature of the powder lineshape. We shall compare these calculations with experiment in a later section.

For $H' \gg 1$, the bare polycrystalline absorption coefficient (calculated assuming $\rho(\nu-\nu_n) = \delta(\nu-\nu_n)$) can be obtained in closed form,[9] because the quantities ν_n and W_n can be expressed as simple functions of the spin Hamiltonian parameters and the angles specifying the orientation of $\underset{\sim}{H}$. This can be seen by writing Eq. (2) in a coordinate system $\{x'y'z'\}$ where $H \parallel z'$, and choosing the eigenstates of $S_{z'}$ as a basis set. In this system, off-diagonal terms may be neglected, since the eigenfunctions are very nearly eigenstates of $S_H \equiv S_{z'}$. The magnetic dipole transition probability W_n is large only for transitions between adjacent levels, and the transition frequencies cluster about $\nu = g\mu_B H$ with a spread that is a different linear function of D and λ for each transition. The lineshape for each transition can be calculated by expressing the solid angle $d\Omega_H$, which is a function of two angles, in terms of the transition frequency and one angle using a Jacobian determinant. The double integral in Eq. (5) then reduces to a single integral which can be expressed in terms of the complete elliptic integral of the first kind. The resulting individual lineshapes can then be multiplied by the appropriate factors $P_n(T)$ and W_n, and

added to give the total bare absorption coefficient. We shall compare the results of this calculation for S = 5/2 with our experimental results for ferrichrome A in a later section of the paper.

III. EXPERIMENTAL TECHNIQUES

Our spectra were obtained using the techniques of far-infrared Fourier transform spectroscopy.[10] The Michelson interferometer, sample dewar, and detection system employed have been recently described elsewhere.[11] Although the results obtained are analogous to those of electron paramagnetic resonance, there are a number of significant differences. Our spectra were measured for samples in fixed magnetic fields over a continuous range of frequencies roughly determined by the spectral bandpass of the sample, and the experimental methods used more nearly resemble those of near infrared spectroscopy than conventional microwave techniques. In addition, the lack of intense broad-band far-infrared sources places severe restrictions upon the application of far-infrared spectroscopy to problems of the type discussed in this paper. Since these restrictions do not generally apply to other magnetic resonance methods, we shall briefly discuss them.

The most widely-used far infrared source is the Rayleigh-Jeans region of the black body spectrum emitted by a high pressure mercury arc lamp. The intensity of the radiation emitted by the lamp varies approximately as ν^2, and the total power radiated by the lamp used in our experiments into the f:1.5 optics of the Michelson interferometer in the region 0-100 cm^{-1} is $\sim 2 \times 10^{-5}$ W. Our experiments were typically made over the smaller spectral range 0-30 cm^{-1}, where the power falling on the detector, with no sample in place, is of the order of 10^{-8} W.

This very small power requires the use of high sensitivity, low temperature detectors; the Ge bolometer[12] used in our experiments has a noise equivalent power of $\sim 10^{-12} W/\sqrt{Hz}$, and it is the limiting source of noise in our detection system. The methods of Fourier transform spectroscopy are especially suited[10] to this situation. However, in practice the small available source intensity means that:

(i) Spectral features in the sample which absorb less than 5 per cent of the incident radiation cannot be accurately measured.

(ii) Spectral regions in which the sample transmission is less than 1% are virtually inaccessible to our present techniques.

These two practical limitations have several implications for the present study. For example, accurate measurements of magnetic dipole lineshapes in the far infrared require samples which contain in excess of $\sim 10^{19}$ spins: for compounds such as myoglobin where the paramagnetic ion concentration is very dilute, a typical sample consists of 1 gram of material. Many large biological molecules are difficult to obtain in such quantities. In addition, the sample temperature must be low enough so that there is an appreciable Boltzmann population difference $P_n(T)$. For values of D on the order of a few cm^{-1}, liquid helium temperatures are necessary. Low temperatures are also required because the thermal population of higher frequency excitations in the sample, such as vibrational modes, may substantially reduce the low frequency transmission, and because magnetic resonance linewidths generally increase rapidly with temperature.

We have also observed strong broad-band absorption at high frequencies in all of the compounds investigated. This absorption, which may be

due to low-lying vibrational modes, has an onset which varies roughly inversely with molecular weight, and it essentially creates a low frequency spectral window which limits the frequency range in which our measurements may be performed. For example, the available frequency range for our measurements of myoglobin and hemoglobin was approximately 3.5-16 cm^{-1}. The low frequency limit is approximately the same for all samples, and is due to the small source intensity at low frequencies.

Within the restrictions outlined above, however, far infrared magnetic resonance has several advantages over other techniques. The major advantage is that both frequency and field information can be obtained over relatively wide ranges; in particular, large zero field splittings for polycrystalline samples may be directly measured. The observed spectra clearly show the variation of the transition frequencies with field, and the individual transitions are easily identified. Paramagnetic resonances which are too broad to be accurately measured by microwave techniques can be observed more easily at higher magnetic fields in the far infrared, and the high frequency information which can be obtained is particularly sensitive to the approximations of the spin Hamiltonian. Finally, far infrared results can be used to determine the parameters of the spin Hamiltonian even when the zero field splittings lie in the microwave region (we shall illustrate this point in the discussion of our results for ferrichrome A).

Our far infrared transmission spectra show effects due to both magnetic resonance transitions and the background transmission spectrum of the sample. In order to remove the background, spectra were customarily

obtained at several values of applied magnetic field, and ratios of spectra for different fields were computed. An example of this method is illustrated in Fig. 4. The upper graph shows an observed spectrum for zero applied field compared with the spectrum obtained with no sample in place. The shape of the latter curve is due to the ν^2 rise of the source intensity, the frequency variation of the efficiency of the dielectric film beam splitter, and the attenuation of low-pass filters which are used to eliminate unwanted high frequencies. The sample spectrum shows a weak magnetic resonance absorption at $\nu \cong 33\ cm^{-1}$, and a strong broad band attenuation at frequencies above $\sim 20\ cm^{-1}$. The broad band attenuation is due both to the absorption in large molecules previously mentioned and to the onset of large scattering within the polycrystalline sample at wavelengths approximately equal to crystallite dimensions.[13] The lower graph is a plot of the ratio of a spectrum for high applied field to the spectrum for zero field. The ratio shows clearly the zero field absorption and the resonance due to the Zeeman splitting of the ground state, as well as noise on either end of the frequency range which is enhanced by computing the ratio of small numbers. This technique works well whenever the shift of the magnetic resonance spectrum with field is large compared to the linewidths, but for spectra such as those described in Fig. 3, the analysis is difficult.

Several of our spectra show effects due to alignment of the sample crystallites in a magnetic field. The alignment is due to the torque produced by the interaction of the low temperature ionic moment with the

applied field. This torque is widely used [14] to measure the low temperature magnetic susceptibility of paramagnetic ions whose ground term is described by Eq. (2). The torque acts to align one of the coordinate axes of the spin Hamiltonian with the applied field, and therefore strongly affects the observed magnetic resonance spectrum. In addition, the crystallite alignment can significantly increase or decrease the high frequency background attenuation due to scattering if the crystallites are plate-like. We have attempted to avoid these effects in our measurements by dispersing the crystallites in transparent glasses, such as mineral oil, and fine magnesium oxide powders, or by packing samples which are too valuable to disperse.

The samples were typically placed within the far-infrared light pipe in cylindrical polyethylene containers with a sheet of thin Mylar covering the top. The container diameter was ~ 1.1 cm, and the sample length varied from 0.1 to 2.5 cm. Metal cones were placed above and below the sample container to form a low-Q transmission cavity. For temperatures $T \leqslant 4.2°K$, the sample mount was immersed in liquid helium. For higher temperatures, the sample mount was placed in an evacuated tube and heated by means of a heating coil. The temperature was monitored by measuring the resistance of a GaAs diode in thermal contact with the sample mount.

IV. EXPERIMENTAL RESULTS

A. Ferrichrome A

Ferrichrome A[15] is a metabolic product of the smut fungus Ustillago sphaerogena; its precise biological function is unknown. The crystal and molecular structure of ferrichrome A tetrahydrate have been recently obtained by X-ray crystallography.[16] The molecule contains one Fe(III) ion in the configuration shown in Fig. 5. Although the iron coordination is roughly octahedral, the local symmetry is that of a left-handed propeller, which suggests that there should be a large rhombic component in the ligand field at the iron site.

Electron paramagnetic resonance measurements[7] at 9 GHz for polycrystalline ferrichrome A have been reported by Wickman, Klein, and Shirley. These measurements showed a broad asymmetric resonance at 1550 Oe for temperatures between 1.0 and 300°K and additional smaller structure was observed at both higher and lower fields in the spectra for temperatures less than 4.2°K. The spin Hamiltonian Eq. (2) was used to analyze

the spectra, under the assumption that the Zeeman term was small compared to the ligand field terms, and reasonable agreement was obtained for $S=5/2$, $D > 0$, and $\lambda=0.25 \pm 0.04$. The assignment $D > 0$ was based upon a fit of the low field effective g-values for the two possible ground doublets to the additional low temperature structure, and the range of λ was obtained from an approximate fit to all of the observed resonances. The temperature variation of the spectra was used to give a rough estimate of 3.5 cm^{-1} for the zero field splitting between the two lowest Kramers' doublets, and the temperature dependence of subsequent Mössbauer effect measurements[17] was used to obtain an improved estimate of 2.4 - 3.5 cm^{-1} for this splitting.

Our far infrared spectra for approximately 70 mg of polycrystalline ferrichrome A, kindly supplied by Dr. M. P. Klein, showed no absorptions in zero field above our experimental lower frequency limit of 3.0 cm^{-1}; spectra were obtained at T = 1.3 and 4.2 °K. In high applied field, a broad asymmetric absorption which peaked at a frequency slightly less than $\nu = 2\mu_B H$ was observed. The average of three experimental absorption coefficient measurements for H=52.2 kOe and T=4.2°K is plotted in Fig. 6. The positions of the absorption maximum and the shoulders on either side were quite reproducible. No evidence of crystallite orientation due to magnetic torques was observed.

The absence of zero field absorptions above 3.0 cm^{-1} suggests that the observed high field lineshape corresponds to the limit $H' \gg 1$. The bare polycrystalline absorption coefficient in this limit was calculated according to the method outlined in Section II-C for various values of D and λ, and the results for two sets of values are also plotted in Fig. 6.

The theoretical absorption coefficient at T = 4.2°K is largely given by the sum of the lineshapes for the two transitions from the ground and first excited states. The small delta-function at $\nu=2\mu_B H$ on the curve for D < 0 is due to the transition from the second excited state, and the contribution of the remaining transitions is negligible. The major features of the calculated $\bar{\alpha}$ are the absorption edges and the position of the maxima; the frequencies at which they occur can be easily expressed in terms of the spin Hamiltonian parameters. The absorption edges occur at frequencies ν_i given by

$$\nu_{1a} = g\mu_B H + 2D(1+3|\lambda|)$$
$$\nu_{1b} = g\mu_B H - 4D$$

$$\nu_{2a} = g\mu_B H + D(1+3|\lambda|$$
$$\nu_{2b} = g\mu_B H - 2D$$

where i = 1,2 indicate the transitions from the ground and first excited states respectively, and the maxima occur at frequencies ν_i' given by

$$\nu_1' = g\mu_B H + 2D(1-3|\lambda|)$$

$$\nu_2' = g\mu_B H + D(1-3|\lambda|).$$

These frequencies are indicated in Fig. 6 for the solid curve. Although D and λ have the same sign,[7,8] the calculated absorption coefficient for a given field depends only upon the sign and magnitude of D and the magnitude $|\lambda|$. Therefore, apart from the effects of the Boltzmann term $P_n(T)$, the lineshape for D < 0 may be obtained from the lineshape for D > 0 by

reflecting through the line $\nu = g\mu_B H$. For $|\lambda| = 1/3$, the absorption coefficient depends only upon the magnitude $|D|$, and the sign of D can be regarded as arbitrary. This result is due to the symmetry properties of the spin Hamiltonian.[7,8] As $|\lambda|$ approaches 1/3, the distinction between positive and negative D becomes smaller, and consequently the sign of D becomes more difficult to obtain from paramagnetic resonance spectra.

The theoretical absorption coefficient for $g = 2.00$, $D = -0.27\ cm^{-1}$, and $|\lambda| = 0.25$, which is plotted in Fig. 6, represents our best fit to the observed lineshape; reasonable agreement places limits of $\pm\ 0.01\ cm^{-1}$ on D and $\pm\ 0.02$ on $|\lambda|$. Considerably poorer fits are obtained for positive values of D, since the calculated maxima lie at frequencies higher than $\nu = 2\mu_B H$ for $|\lambda| < 1/3$. The best fit to all of the observed features for $D > 0$ requires $|\lambda| = 1/3$ and $D = +0.27\ cm^{-1}$; however, as indicated above, the same curve would be obtained for a negative D. The curve for these values is also plotted in Fig. 6. Both sets of values derived from our data yield zero field splittings between adjacent Kramer's doublets in the range 0.8 to $1.0\ cm^{-1}$. These values are considerably smaller than the estimates previously obtained.

The analysis of the paramagnetic resonance spectra of ferrichrome A, given by Wickman, Klein, and Shirley, rests upon the assumption that the Zeeman terms of Eq. (2) are much smaller than the ligand field terms, which is approximately equivalent to the statement that the energy of the microwave quantum is much less than the zero-field splittings. Our measurements indicate that this assumption is not justified at the experimental microwave frequency of $0.3\ cm^{-1}$. It has recently been pointed out[8] that the effective g-factors in finite fields (such that $g_{eff}\ \mu_B H \sim D,E$) may vary considerably from those calculated in the limit

of infinitesimal fields, and that resonances due to transitions between states originating in adjacent zero field doublets may also be observed. Under these conditions, the analysis of the low temperature paramagnetic resonances for ferrichrome A in the small field approximation is inconclusive with respect to the sign of D. Our data are fit more accurately by negative values of D, as indicated in Fig. 6. Preliminary calculations of the X-band electron paramagnetic resonance spectrum[18], using the methods outlined in Ref. 8, indicate that the values $D = -0.27\ cm^{-1}$ and $|\lambda| = 0.25$ obtained from our data can account for the published resonances. More accurate values of D and λ could possibly be obtained by further analysis of X-band spectra, or by directly obtaining the zero field splittings at K-band.

B. Tris (pyrrolidyl dithiocarbamato) Fe(III)

A series of interesting tris Fe(III) dithiocarbamates, Fe(III) $(S_2CNRR')_3$ where R,R' are alkyl groups, have recently been synthesized.[19] Although X-ray crystallographic data are unavailable, the molecular structure of these compounds is presumed on steric grounds[20] to be that shown in Fig. 7. A comparison of Fig. 5 and Fig. 7 illustrates the similarity of the iron environment in these complexes to that in ferrichrome A.

Most of these substances show room temperature magnetic moments which lie between those of the limiting "low spin" (S=1/2) and "high spin" (S=5/2) states of the Fe(III) ion. Visible and infrared spectra and the temperature and pressure dependence of the magnetic susceptibility for several compounds have been reported,[20] as well as Mössbauer effect[21] and nuclear magnetic resonance[22] studies. Good agreement with the qualitative effects observed in these measurements has been obtained by assuming that the octahedral $^2T_2$ and $^6A_1$ terms of

Fe(III) are separated by only a few hundred cm^{-1} in these complexes. However, quantitative agreement with the experimental results has been frequently hampered by the lack of detailed information on the effects of the ligand field at the iron site. For example, the similarity of the iron site in the tris Fe(III) dithiocarbamates to that in ferrichrome A suggests that the ligand field should have a strong rhombic component in these complexes. Such information can in principle be obtained by electron paramagnetic resonance techniques, and a study at microwave frequencies is now in progress.[18]

One of the compounds, tris (pyrrolidyl dithiocarbamato) Fe(III), shows only high spin behavior. We have measured the far infrared spectra of a polycrystalline sample of this substance at T=4.2°K. The sample was supplied by Dr. A. M. Trozzolo. Data obtained from our spectra are plotted in Fig. 8. The zero field spectrum shows a strong absorption at 8.4 cm^{-1} and a weaker one at 13.2 cm^{-1}. The first absorption corresponds to a transition between the ground and first excited Kramers doublets, and the second corresponds to a transition between the ground and second excited doublets. As discussed in Section II-C, the presence of the second absorption at low temperature implies that $\lambda \neq 0$. Our zero field data are fit by the values $D = -2.14 \pm 0.05$ cm^{-1} and $\lambda = -0.10 \pm 0.01$. The sign of D was established by observing that the first absorption occurs at a frequency more than half of the total zero field splitting, 13.2 cm^{-1}; if $D > 0$, the reverse is true. In addition, no absorption due to the Zeeman splitting of the ground doublet was observed, which is further evidence for a negative value of D.

Our measurements indicate that the ligand field in this compound does in fact have an appreciable rhombic component, as expected. It is

interesting to note that low temperature microwave magnetic resonance measurements[18] for frozen solutions of this compound in N,N dimethyl formamid indicate that $|\lambda| \sim 1/3$ and that $|D|$ is somewhat smaller. Previous experiments[19] have also shown differences between the room temperature magnetic moments of solid and dissolved compounds of this series.

C. Fe(III) Porphyrins

Metalloporphyrins[23] have been investigated by a variety of techniques because of their occurrence as the prosthetic group of paramagnetic ions in many biological molecules. In particular, studies on iron prophyrins (hemes)[24] have been stimulated by interest in related work in hemoproteins.[25] The approximate structure of such heme compounds and the local coordination of the iron ion in the porphyrin molecule are indicated in Fig. 9. The iron is coordinated to the four pyrrole nitrogens of the porphyrin, and two other coordination positions, labeled 5 and 6, are available in positions approximately perpendicular to the plane defined by the four nitrogen atoms. In the compounds we shall discuss, one of these positions is unoccupied, and the other is occupied by one of a number of ligands. Under these conditions, X-ray crystallographic measurements[26] have indicated that the iron lies slightly above the nitrogen plane in the direction of the fifth ligand. Hemes are also identified by the presence of various groups attached to the periphery of the porphyrin skeleton. Protoheme (iron protoporphyrin), the prosthetic group of hemoglobins, myoglobins, and several other hemoproteins, is the best-known.

The far-infrared spectra of several halogeno deutero- and protoporphyrin Fe(III) complexes have recently been reported.[27] The spectra for these compounds were successfully analyzed in terms of the spin Hamiltonian in Eq. (2) for $S = 5/2$, $D > 0$, and $\lambda \cong 0$. The observed zero field splittings for the compounds investigated were quite large, ranging from 11.1 cm^{-1} to ~ 33 cm^{-1}. Preliminary results for several other complexes were also reported.

We have measured the far-infrared spectra of four polycrystalline Fe(III) porphyrins. The sample preparation and characterization techniques were similar to methods recently described elsewhere.[28] These measurements have yielded more accurate values of the ligand field parameters for two compounds included in the previous work, iodo- and azido deuteroporphyrin IX dimethyl ester Fe(III). In addition, we have also obtained data for two additional compounds, fluoro- and azido protoporphyrin IX dimethyl ester Fe(III). The last two substances are of particular interest because of the existence of the corresponding myoglobin and hemoglobin complexes.

The spectra for all of the compounds investigated showed absorptions characteristic of high spin Fe(III) and positive values of D. A typical transmission spectrum has been previously shown in Fig. 4. The frequencies of the experimental absorption peaks at $T = 4.2°K$ are plotted as a function of field for fluoro protoporphyrin IX dimethyl ester Fe(III) in Fig. 10. The observation of absorptions corresponding to the Zeeman splitting of the ground doublet establishes the sign of D as positive. The frequency of the zero-field absorption corresponds to the zero-field splitting Δ_1 between the ground and first excited Kramers doublets.

If $\lambda = 0$, as we would expect for this complex,[29] $\Delta_1 = 2D$. The calculated frequencies of strong transitions for the spin Hamiltonian in Eq. (2) for $D = +5.0\ cm^{-1}$, $\lambda = 0$, $T = 4.2°K$, and $\underset{\sim}{H} \perp \hat{z}$ are also plotted in Fig. 10 for comparison. The values of the zero field splittings and derived values of D obtained from our data for the two halogeno complexes investigated and previously reported values for related complexes are listed in Table I.

The azide complex of ferrihemoglobin has a static magnetic susceptibility[30] characteristic of the low spin S = 1/2 state of Fe(III). Electron paramagnetic resonance measurements[31] show a strongly asymmetric g-tensor which has been successfully analyzed[32,33] in terms of a rhombic ligand field. Similar results have been obtained for the azide complex of ferrimyoglobin. The origin of the rhombic field in these complexes is in dispute. It has been ascribed to the attachment of the distal histidine[31] (which is coordinated to the iron in the fifth position in these compounds), to the displacement of the iron atom out of the nitrogen plane,[33] and to a non-axial attachment of the azide ion.[32] An X-ray crystallographic study of ferrimyoglobin azide,[34] which showed that the azide ion is inclined at 21° to the porphyrin plane, has given support to the latter explanation.

Our measurements for the corresponding heme compound, azido protoporphyrin IX dimethyl ester Fe(III) and the similar deuteroheme complex show spectra characteristic of high spin Fe(III). However, an orientation of the azide ligand in these substances similar to that observed in ferrimyoglobin should produce a substantial rhombic component in the ligand field. In order to determine the value of λ for these

complexes, zero-field spectra were obtained at T = 4.2 and 50°K. At the higher temperature, an additional absorption was observed in the spectra at a frequency corresponding to the second zero-field splitting Δ_2 between the first and second excited Kramers doublets. As discussed in Section II-C, the values of Δ_1 and Δ_2 obtained in this fashion can be used to obtain values for D and λ. The observed splittings and the derived values of D and λ for the two compounds are also listed in Table I. Both complexes show non-zero values of λ, indicating that the ligand field does in fact have a rhombic component. The value of λ for the protoheme complex is particularly large compared to the limiting value $\lambda = 1/3$ for a completely rhombic field. (A typical value of λ for an axial complex, obtained from high-temperature data for chloro protoporphyrin IX Fe(III),[27] is $\lambda = 0.01 \pm 0.01$.) Although the iron atom in these compounds may be displaced out of the nitrogen plane, an axial displacement cannot account for a rhombic component to the ligand field.[5] The most likely source of such a field is an inclination of the azide ion to the porphyrin plane such as that observed in ferrimyoglobin azide. If this is the case, our measurements on these complexes indicate that the orientation of the azide ligand can significantly contribute to the rhombic character of the ligand field in ferrimyoglobin and ferrihemoglobin.

D. Mn(III) Porphyrins

Studies of manganese porphyrins, like those of iron porphyrins, have been stimulated by interest in their properties in relation to biological systems. For example, several studies have investigated the possible role of these compounds in the oxidation-reduction systems of photosynthesis.[35,36] In addition, synthetic enzymes made by replacing

heme prosthetic groups with Mn porphyrins have been shown to have partial activity.[37] Elemental analysis and magnetic susceptibility measurements on manganese hematoporphyrins[35] have shown that the stable oxidation state of the manganese in these complexes is Mn(III), with a spin S=2. However, no electron paramagnetic resonances that can be directly attributed to Mn(III) have been observed.

We have measured the far infrared spectra of four polycrystalline Mn(III) deuteroporphyrins. The spectra for three of the compounds investigated show magnetic resonance absorptions which are consistent with the predictions of the spin Hamiltonian in Eq. (2) for S=2, $D < 0$, and $\lambda \cong 0$. Data for a sample of azido deuteroporphyrin IX dimethyl ester Mn(III) at T=4.2°K are shown in Fig. 11. The zero-field spectrum for this complex showed only one absorption, indicating that $\lambda \sim 0$. For small λ, the zero-field spectrum should show two absorptions separated by $6E$, as indicated in Fig. 1. The width of the observed zero-field lineshape was used to obtain the upper limit $E \leqslant 0.1\ cm^{-1}$. Since transitions corresponding to the Zeeman splitting of the pair of zero field states with $m_s \cong \pm 2$ are forbidden to first order, the absence of such absorptions in the observed spectra does not imply that D is positive. In order to establish the sign of D, we have obtained spectra for this complex in zero field at temperatures up to 40°K. No additional absorptions appeared above $3.5\ cm^{-1}$, which shows that D is negative and that therefore the observed zero-field splitting is $\Delta_1 = 3|D|$. These observations yield $D = -3.08 \pm 0.10\ cm^{-1}$ and $\lambda \leqslant 0.04$. The calculated transition frequencies for Eq. (2) with $D = -3.08\ cm^{-1}$, $\lambda = 0$, and $\underset{\sim}{H} \perp \hat{z}$ are also plotted in Fig. 11 for comparison. Similar observations were used to derive values of D

and λ for chloro deuteroporphyrin IX dimethyl ester Mn(III). However, no absorption in zero field was observed for the third complex, bromo deuteroporphyrin IX dimethyl ester Mn(III). The spectra in an applied field showed substantial changes in the background attenuation at high frequencies due to crystallite orientation. In addition, a broad magnetic resonance absorption with a peak at $\nu = 2\mu_B H + 3.4\ cm^{-1}$ was observed at high field. These observations are only consistent with a small negative value of D, and analysis of the observed high field lineshape in the manner described for ferrichrome A yields the value $D = -1.1 \pm 0.1\ cm^{-1}$. The derived values of the spin Hamiltonian parameters for these three complexes are listed in Table II. The fourth compound studied, iodido deuteroporphyrin IX dimethyl ester Mn(III), showed spectra similar to those observed for the bromo derivative, indicating a small absolute value of D. However, the extremely broad absorptions observed prevented an accurate measurement of either the sign or magnitude of D for this complex.

Our measurements have therefore shown magnetic resonance absorption which is definitely due to high-spin (S=2) Mn(III). The observed large values of D, coupled with the small transition probabilities for transitions between Zeeman-split "doublets" for S=2, may account for the lack of microwave paramagnetic resonance signals for these complexes. In addition, we can make some interesting comparisons of the spin Hamiltonian parameters obtained for corresponding Mn(III) and Fe(III) porphyrins. For example, the values of D obtained for halogeno proto- and deuteroheme complexes increase in the order F < Cl < Br < I, as indicated in Table I. The relation of this effect to various chemical series (nephelauxetic,

electronegativity, and metal-halide bond strength) and to π-bonding in the porphyrin system has been recently discussed.[38] The algebraic values of D obtained for halogeno manganese porphyrins also show this behavior. It is interesting to note that the small negative value of D for the bromo derivative implies that D for the iodido complex may be positive. In addition, the algebraic value of D for the azido derivatives of both Fe(III) and Mn(III) deuteroporphyrins is less than that for the chloro derivatives. Finally, the limits of λ for azido deuteroporphyrin IX dimethyl ester Mn(III) include the measured value for the corresponding deuteroheme compound, indicating that the effects of the azide ligand are similar in the two complexes.

E. Ferrihemoglobin and Ferrimyoglobin

Hemoglobins and myoglobins are found in all vertebrates: hemoglobin in the red blood cells and myoglobin in the tissues. A large number of chemical, biological, and physical techniques have been applied to the study of these compounds because of their central importance to the process of respiration.

Both hemoglobin and myoglobin contain iron atoms coordinated to a protoporphyrin prosthetic group and to a nitrogen atom of a histidine residue of the globin. Hemoglobin contains four such iron atoms, and myoglobin contains one. In the respiratory process, the iron atom is in the Fe(II) state, and the sixth coordination position is available for the reversible bonding of molecular oxygen. Studies of hemoglobin and myoglobin where the iron is in this state are therefore of greater value to the understanding of the biological function of these

compounds. However, a large amount of useful information has been obtained from measurements on complexes containing Fe(III). Perhaps the most striking examples of such measurements are the determination of the orientation of the heme planes in hemoglobin[39] and myoglobin[40] by electron paramagnetic resonance measurements on Fe(III), in the high spin (S=5/2) state. In particular, such paramagnetic resonance measurements have been restricted to complexes containing Fe(III) since the low spin (S=0) Fe(II) derivatives have no paramagnetism and since no electron paramagnetic resonance has been observed for high spin (S=2) Fe(II) compounds.

Hemoglobin and myoglobin derivatives in which the sixth coordination position is occupied by a water molecule (we shall refer to these as met-hemoglobin and met-myoglobin) or a fluoride ion have been shown to contain Fe(III) in the high spin state. Static magnetic susceptibility measurements for these complexes[41] have found room-temperature magnetic moments slightly less than the value of 5.92 Bohr magnetons expected for S=5/2. Microwave electron paramagnetic measurements[42] for these complexes have found $g_{\parallel} = 2.0$ and an isotropic $g_{\perp} = 6.0$, which is characteristic of the Zeeman splitting of the ground doublet for the spin Hamiltonian in Eq. (2) for S=5/2, $D > 0$, and a magnitude of D much larger than the microwave quantum. A number of indirect methods have been used to obtain values of D for these compounds: for example, microwave paramagnetic resonance,[43] temperature dependence of the magnetic susceptibility,[44] torque magnetometer measurements of magnetic anisotropy,[14] and Mössbauer resonance.[45] As the results discussed for heme compounds indicate, far-infrared spectroscopy

can, in principle, directly measure the zero-field splitting and thus the value of D for such compounds.

We have measured the far-infrared spectra of bovine hemoglobin and sperm whale myoglobin with both water and fluoride ligands. The spectra for each compound show absorptions corresponding to the Zeeman splitting of the ground doublet in applied fields up to 52.2 kOe, and to the zero-field splitting for the fluoro derivatives. The polycrystalline sample of met-myoglobin was supplied by Dr. G. Feher. The remaining samples were pastes made by mixing approximately 1 gm. of lyophilized material (obtained from Mann Research Laboratories and Sigma Chemical Company) into 1 ml of distilled water buffered to pH 7.0 with a drop of 1 M mixed phosphate buffer. The fluoro derivatives were obtained by using an aqueous solution containing approximately four molar equivalents of fluoride. The composition of the paste samples was verified by measuring the optical spectra of suitably diluted aliquots.

Data obtained from the far-infrared spectra of the two fluoro complexes at 4.2°K are shown in Fig. 12. Since $\lambda \sim 0$ for these complexes, the observed absorption in zero field corresponds to $\Delta_1 = 2D$. Curves for the transitions calculated from Eq. (2), using values of D derived from the zero field splitting, are also plotted for $\lambda = 0$ and $\underset{\sim}{H} \perp \hat{z}$. The frequency region in which accurate data can be obtained from the far infrared spectra for these compounds is indicated by the plotted range. For comparison, the typical transmission range for a heme compound sample with approximately the same number of Fe(III) ions is indicated in Fig. 4. In addition, the high-frequency attenuation in hemoglobin and myoglobin increases very rapidly with frequency. For example, a reduction of the sample

path length by a factor of two only increases the available frequency range by ~ 1 cm^{-1}.

Absorption corresponding to the Zeeman splitting of the ground doublet was observed in the spectra for met-hemoglobin and met-myoglobin. However, no absorptions were observed in zero field below the maximum frequency limit for these compounds of ~ 16 cm^{-1}. This observation implies $D \gtrsim 8$ cm^{-1} for these complexes. A more accurate estimate of D for met-myoglobin can be obtained from our data for the Zeeman splitting of the ground doublet, shown in Fig. 13. The points plotted are the average values of the measured frequencies of peak absorption obtained from eight experimental runs at 4.2°K. The curves are the transition frequencies calculated from Eq. (2) with $\lambda = 0$, $\underset{\sim}{H} \perp \hat{z}$, and the indicated values of D. A comparison of this figure with the data obtained for the fluoro derivative in Fig. 12 indicates that $D \cong 9$ cm^{-1}, and a conservative estimate places D in the range[46] $D = 9.5 \pm 1.5$ cm^{-1}. Preliminary observations for met-hemoglobin indicate that D is slightly larger (~ 10.5 cm^{-1}) in this compound.

The values of D derived from the far-infrared data are listed in Table III. We have also included several values of D obtained from indirect measurements, and have reproduced the value for fluoro protoporphyrin IX dimethyl ester Fe(III), for comparison. It is interesting to note that the value of D for the fluoro derivatives of myoglobin, hemoglobin, and protoheme are very similar, indicating that the influence of the protein on the ligand field at the iron site is relatively small. In addition, no evidence for inequivalent sites due to the two different protein chains in hemoglobin was found in the far-infrared spectra for

the fluoro derivative. If two inequivalent sites are assumed, the observed zero-field lineshape places an upper limit of ~ 0.4 cm^{-1} on the difference of the value of D between the two sites.

F. Bis Fe(III) Dithiocarbamates

Further studies of the tris Fe(III) dithiocarbamates, discussed in Section IV-B, have resulted in the synthesis of a series of novel bis Fe(III) dithiocarbamate compounds, the bis (N,N dialkyl dithiocarbamato) Fe(III) halides: $(R_2NCS_2)_2$ FeX where R is an alkyl group and X is a halogeno ligand. X-ray crystallographic measurements[47] on one of the complexes in this series have obtained the molecular structure shown in Fig. 14. The iron atom in these compounds is pentacoordinate, and lies approximately at the centroid of a rectangular pyramid formed by the four sulfur atoms of the two dithiocarbamate lagands and the halide atom. The local symmetry of the iron site is thus nearly square pyramidal, but the total symmetry is much lower (C_{2v}).

Measurements of the static magnetic susceptibility,[47,48] electron paramagnetic resonance,[49] and Mössbauer resonance[50] have shown that the Fe(III) ground multiplet is described by the spin Hamiltonian in Eq. (2) with the unusual "intermediate spin" value S=3/2, $\lambda \cong 0$, and large values of D. Although the energy of the $^4T_{1g}$ octahedral state never lies lowest,[51] the low C_{2v} symmetry of the iron site in these complexes completely removes the degeneracy of the d^1 levels. A simple argument[9] shows that under these conditions it is possible to stabilize a $^4A_2$ ground state derived from the $(t_{2g}^4 e_g)$ $^4T_{1g}$ octahedral state.

In order to investigate the validity of the spin Hamiltonian approximation and to obtain values for the spin Hamiltonian parameters,

we have measured the far-infrared spectra of seven polycrystalline compounds in this series. A brief account of this work has been recently published elsewhere.[52] Data obtained from the far-infrared spectra of $((i\text{-}C_3H_7)_2NCS_2)$ Fe(III) Cl at 4.2°K are shown in Fig. 15. As discussed in Section II-C, the zero field spectrum for S=3/2 shows only one absorption, and is therefore not sufficient to determine D and λ. However, electron paramagnetic resonance measurements on this compound[48] have obtained the value $\lambda=0.036\pm0.003$. Using this value, the measured zero-field splitting $\Delta = |D|[1+3\lambda^2]^{1/2}$ gives $|D| = 2.35 \pm .03\ cm^{-1}$. A comparison of the polycrystalline absorption coefficient, calculated using the program discussed in Section II-C, with the observed spectra showed that $D < 0$. The frequencies of the calculated absorption maxima for $D= -2.35\ cm^{-1}$, $\lambda = 0.036$, and $T = 4.2°K$ are also plotted in Fig. 15. The calculated polycrystalline absorption coefficients for these parameters have been shown in Fig. 3. The fit between the observed absorptions and the calculated maxima is excellent. The zero field splittings and derived values of the spin Hamiltonian parameters obtained in this manner from the far-infrared spectra for the compounds investigated are listed in Table IV. Since the polycrystalline spectra are relatively insensitive to values of $\lambda < 0.1$, we have only listed the values of λ obtained for two compounds. The listed values of D for the remaining compounds were obtained from the zero field splitting, assuming $\lambda = 0$. In addition, the calculated polycrystalline absorption coefficient is relatively insensitive to small changes in the spin Hamiltonian, such as the assumption of a slightly anisotropic g-factor.

One of the compounds investigated, $((C_2H_5)_2NCS_2)$ Fe(III) Br, can be obtained in relatively large crystals (0.5 × 3 × 3 mm). The far-infrared spectra of a polycrystalline sample of this compound at 4.2°K showed sharp absorptions that were approximately consistent with the predictions of the spin Hamiltonian in Eq. (2) for $D > 0$ and θ_H near $\pi/2$, indicating a substantial alignment of the crystallites by the low-temperature magnetic torque discussed in Section III. (A calculation of the torque for this compound at T = 4.2°K and H = 52.2 kOe yields a maximum torque per unit volume of 8.65×10^5 dyne/cm^2 which tends to rotate the crystallites toward $\underset{\sim}{H} \perp \hat{z}$.) In order to obtain a more accurate comparison of the predictions of the spin Hamiltonian with the observed spectra, an ordered sample of this complex was constructed with the crystallites approximately oriented with $\underset{\sim}{H} \perp \hat{z}$. The data obtained from the far-infrared spectra of this sample is plotted in Fig. 16. A "best fit" to the observed spectra was calculated from Eq. (2) by varying θ_H and using values of D and λ chosen to fit the zero-field splitting and the Zeeman splitting of the ground doublet ($g_\perp \sim 4$). The calculated transition frequencies for the values obtained, $\theta_H \sim 3\pi/8$, $D = +7.50$ cm^{-1}, and $\lambda = 0.067$, are also plotted in Fig. 16. The value of θ_H obtained was consistent with both the construction of the sample and the effects of the magnetic torque. Although the fit to the observed absorptions is quite good, the deviation of the high-frequency experimental absorption maxima from the calculated curves is more than the experimental error (0.1 cm^{-1}). This small discrepancy cannot be explained by imperfect alignment of the crystallites. It is more likely due to the assumption of an isotropic g-factor made in Eq. (2).

Another compound in this series, $((C_2H_5)_2\ NCS_2)_2$ Fe(III) Cl, has been shown[50] to be a ferromagnet with a low transition temperature $T_c = 2.43°K$. Mössbauer resonance data for this complex in the paramagnetic state have indicated that $D < 0$. Our zero-field spectra for this compound at 4.2°K showed only a very weak, broad absorption in the range 2.5 to 5 cm^{-1}, in contrast to the sharp (half width ~ 0.4 cm^{-1}) absorptions observed for other complexes in this series. The large width of the zero-field resonance is probably due to the effects of exchange broadening, since the Heisenberg exchange coupling parameter J is of the same order as D. For T = 1.3°K, the spectra showed a sharp ferromagnetic resonance absorption at 3.85 cm^{-1} in zero field. The zero-field resonance frequency is related to the anisotropy of the iron environment which is reflected in the strong axial ligand field. A simple classical calculation,[9] suggested by Dr. A. M. Portis,[53] can be used to derive the relationship between the zero field resonance frequency at T = 0, ν_o, and D. This calculation obtains the effective anisotropy field $\underset{\sim}{H}_A$ at T=0 due to the axial term in the spin Hamiltonian by expanding it in a power series in the polar angle. The result, for $D < 0$, is $\nu_o = 2|D|S$. This expression, which is exact in the limit of large spin, must be corrected by the factor[54] $\eta = (1-1/(2S))$. Thus, for spin $S = 3/2$, $\nu_o = 2|D|$. In this case, the zero-field resonance should broaden, and the absorption maximum may shift,[9] as T approaches T_c. Our measurements, obtained for temperatures between 1.3 and 2.3°K, showed a progressive broadening and a decrease of only ~ 0.05 cm^{-1}. We have therefore taken $2|D| = 3.85\ cm^{-1}$. Data obtained from the spectra for a polycrystalline sample of this compound at T = 1.3°K are shown in Fig. 17. The observed absorption maxima lie

close to the line $\nu = 2\mu_B H + 3.85\ cm^{-1}$, which corresponds to an orientation of $\underset{\sim}{H} \parallel \hat{z}$ (or $\underset{\sim}{H} \parallel \underset{\sim}{H}_A$). These observations are consistent with the simple theory of ferromagnetic resonance[55] and with the effects of the expected large magnetic torque.

The ferromagnetic coupling in this complex presumably arises from a super-exchange interaction via the sulfur atoms of neighboring molecules, which lie at normal Van de Waals distances.[46] This observation is interesting in the light of recent measurements on the iron-sulfur protein spinach ferridoxin[56] which indicate that the two iron atoms in this compound are antiferromagnetically coupled at low temperatures.

The similarity of the chemical and macroscopic properties of the bis dithiocarbamates indicates that they form an isostructural series obtained by substitution of either the halide ligand or the alkyl groups. As an example of the effects of such a substitution, we have listed in Table IV the change in the zero field splitting on substitution of a chloro ligand for a bromo ligand for a fixed alkyl group. This quantity, $\Delta_{Br}-\Delta_{Cl}$, is nearly independent of the dithiocarbamate ligand for the first two pairs of complexes, which implies that the effect of alkyl group substitution upon Δ is either small or independent of the halide ligand. However, $\Delta_{Br}-\Delta_{Cl}$ for the pyrrolidynyl derivatives is very different, indicating a substantially different bonding in these complexes. This effect may be due to a greater degree of π-bonding for the pyrrolidynyl derivatives because of the existence of a pseudo-ring structure including the nitrogen atom. A similar effect may explain the temperature-independent high spin magnetic moment of tris (pyrrolidyl dithiocarbamato) Fe(III), discussed in Section IV-B, compared with the temperature-dependent

magnetic moments of the other tris dithiocarbamates.

All of the compounds investigated also showed an increase in the algebraic value of D upon substitution of bromo for chloro ligands for a fixed dithiocarbamate ligand. As discussed in Section IV-D, similar effects are observed in the Fe(III) and Mn(III) porphyrins. Furthermore, we may compare the values of D obtained for the bis Fe(III) dithiocarbamates with the quadrupole splitting ΔE_Q, obtained from Mössbauer effect measurements, which is also listed in Table IV. Since ΔE_Q and D both depend upon the strength and asymmetry of the ligand field, we might expect a correlation between them. This comparison has been made for the Fe(III) porphyrins,[38] where an approximately linear variation of ΔE_Q with D, passing through the origin, is observed. Our data clearly eliminates such a correlation for the bis dithiocarbamates, but does not discriminate against other possible zero-intercept functions. For example, the data can be fit to a saturating function of D with either even or odd parity. Mössbauer resonance and far-infrared measurements on the fluoride and iodide complexes would help to clarify this point.

G. Additional Spectral Features

A number of the compounds investigated in this study showed additional sharp structure which was not magnetic field-dependent. These absorptions were typically at higher frequencies than the observed paramagnetic resonances, and were stronger by factors ranging from approximately two to ten. In general, more sharp lines were observed in the spectra of compounds with relatively small molecular weight, such as the bis Fe(III) dithiocarbamates, than in the spectra for the large biological molecules. However,

all of the compounds showed the broad high-frequency attenuation discussed in Section III.

In most cases, the samples used for the magnetic resonance measurements did not transmit sufficient far-infrared radiation to observe such absorptions at frequencies above approximately 30 cm^{-1}. In order to investigate these absorptions in $((C_2H_5)_2NCS_2)_2$ Fe(III) Br, we measured the far-infrared spectrum of a thin polycrystalline sample at T = 4.2°K. The spectrum obtained in zero applied field is shown in Fig. 18. Although the linewidths and strengths of the observed absorptions are typical, the number of observed lines is not. For example, similar measurements on another bis Fe(III) dithiocarbamate, $((i\text{-}C_3H_7)_2NCS_2)_2$ Fe(III) Cl, showed only one such absorption in the same frequency interval. The frequencies of the absorption maxima for several compounds, obtained from the far-infrared spectra at 4.2°K, are listed in Table V. It is interesting to note that a sharp absorption (width ~ 0.75 cm^{-1}) was observed at 8.9 cm^{-1} in the spectra for the fluoro derivative of ferrimyoglobin. The strength of this absorption was approximately twice that of the zero-field magnetic resonance. In addition, we have included data from preliminary measurements on phenoxo deuteroporphyrin IX dimethyl ester Mn(III). High-temperature spectra for this compound show an increase in the strength of the field-independent absorptions for temperatures up to 77°K.

Although we have not investigated the source of the additional structure in the far-infrared spectra of these compounds, our observations suggest that they are due to transitions between excited molecular vibration states. The linewidths are much too small for absorptions due to lattice vibrations. The temperature dependence of the structure

in the manganese deuteroporphyrin compound indicates that the transitions observed are between excited states. In addition, the structure depends strongly upon changes in ligands within a series of similar compounds. Finally, the observed strengths indicate that the absorptions are due to electric quadrupole transitions, rather than electric dipole transitions.

V. CONCLUSIONS

Our measurements have shown that far-infrared spectroscopic techniques offer a direct method for the investigation of the effects of large axial and rhombic ligand fields upon paramagnetic ions in molecules. The polycrystalline magnetic resonance absorptions observed are consistent with the predictions of a wide variety of cases of the simple spin Hamiltonian of Eq. (2). We have directly obtained values of the axial ligand field parameter D from the spectra for twenty-one compounds, and have measured $\lambda \equiv E/D$ for several substances with a large rhombic ligand field. In addition, our investigation has shown that such detailed information can also be obtained for paramagnetic ions in intact biological molecules.

ACKNOWLEDGEMENTS

It is a pleasure to acknowledge the assistance of Dr. G. Feher, Dr. M. P. Klein, and Dr. A. M. Trozzolo, who kindly supplied samples for this investigation. In addition, we have had a number of very informative discussions concerning the interesting properties of these compounds with Dr. G. Feher, Dr. M. P. Klein, Dr. W. E. Blumberg, Dr. A. J. Bearden, Dr. K. Tsushima, Dr. G. Harris Loew. Finally we wish to acknowledge the assistance of Dr. A. M. Portis in analyzing the ferromagnetic resonance of $((C_2H_5)_2NCS_2)_2Fe(III)Cl$. This work was performed under the auspices of the U. S. Atomic Energy Commission.

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Table I. Experimental values of the zero field splittings Δ_1 and Δ_2 and derived values of the spin Hamiltonian parameters for several heme compounds obtained from the far-infrared spectra at H=0 for T=4.2 and 50°K.

Compound	Ligand	Δ (cm^{-1})	D (cm^{-1})	λ
Protoporphyrin IX Dimethyl Ester Fe(III)	Fluoro	Δ_1=10.0±.20	5.0±.10	~0
	Chloro[a]	Δ_1=13.9±.28	6.95±.14	~0
	Azido	Δ_1=19.5±.30 Δ_2=36.0±.75	9.10±.15	.085±.025
Deuteroporphyrin IX Dimethyl Ester Fe(III)	Fluoro[a]	Δ_1=11.1±.22	5.55±.11	~0
	Chloro[a]	Δ_1=17.9±.36	8.95±.18	~0
	Bromo[a]	Δ_1=23.6±.46	11.8±.23	~0
	Iodo	Δ_1=32.8±.30	16.4±.15	~0
	Azido	Δ_1=14.8±.10 Δ_2=29.2±.15	7.32±.05	.036±.015

[a] Data from Ref. 28.

Table II. Experimental values of the zero field splitting Δ_1 and derived values of the spin Hamiltonian parameters for several Mn(III) deuteroporphyrins obtained from the far infrared spectra at T = 4.2°K.

Compound	Ligand	Δ_1 (cm^{-1})	D (cm^{-1})	λ
Deuteroporphyrin IX Dimethyl Ester Mn(III)	Chloro	7.6 ± .05	-2.53 ± .02	⩽ .005
	Bromo	⩽ 3.5	-1.10 ± .10	~ 0
	Azido	9.25 ± .15	-3.08 ± .10	⩽ .04

Table III. Observed zero field splittings and derived values of D for ferrihemoglobin and ferrimyoglobin obtained from the far-infrared spectra at 4.2°K. Several indirect values of D and the far-infrared results for fluoro protoheme are included for comparison.

Compound	Ligand	Δ_1 (cm^{-1})	D (cm^{-1})	Indirect value D (cm^{-1})
Ferrimyoglobin	Aquo	> 16	9.5 ± 1.5	~ 12[a] 4.38 ± .6[b]
	Fluoro	11.88 ± .16	5.94 ± .08	~ 7[c]
Ferrihemoglobin	Fluoro	12.60 ± .24	6.30 ± .12	~ 7[d]
Protoporphyrin IX Dimethyl Ester Fe(III)	Fluoro	10.0 ± .20	5.0 ± .10	

[a]Ref. 14
[b]Ref. 43
[c]Ref. 44
[d]Ref. 45

Table IV. Zero-field splittings Δ and derived values of the spin Hamiltonian parameters for bis (N,N dialkyl dithiocarbamato) Fe(III) halides [$(R_2NCS_2)_2$ Fe(III) X], obtained from the far-infrared spectra. The difference in the zero field splitting for compounds with the same dithiocarbamate ligand and different halogeno ligands, $\Delta_{Br} - \Delta_{Cl}$, and the quadrupole splitting obtained from Mössbauer data[a] are also included.

Ligands		Δ (cm^{-1})	$\Delta_{Br}-\Delta_{Cl}$ (cm^{-1})	D (cm^{-1})	λ	ΔE_Q (cm/sec)
R = CH_3	X=Br	14.60 ± .20	18.80	+ 7.30 ± .10		.290
	X=Cl	4.20 ± .04		-2.10 ± .02		.266
R = C_2H_5	X=Br	15.10 ± .20	18.95	+ 7.50 ± .10	.067 ± .005	.288
	X=Cl	3.85 ± .02		-1.93 ± .01		.268
R = (i-C_3H_7)	X=Cl	4.70 ± .06		-2.35 ± .03	.036 ± .003[b]	.268
NR_2=Pyrro-lidyl	X=Br	16.33 ± .20	11.13	+8.17 ± .10		.277
	X=Cl	5.20 ± .09		+2.60 ± .05		.268

[a] Ref. 50

[b] Ref. 49

Table V. Frequencies of absorption maxima for the magnetic field-independent absorptions observed in the far-infrared spectra of several compounds at 4.2°K.

Compound	Absorption Maxima (cm^{-1})
Fluoro ferrimyoglobin	8.9
Bromo deuteroporphyrin IX dimethyl ester Mn(III)	25.2, 34.0, 39.3, 46.0
Phenoxo deuteroporphyrin IX dimethyl ester Mn(III)	13.3, 31.0
Bromo bis (N,N diethyl dithiocarbamato) Fe(III)	27.3, 31.5, 34.5, 42.9, 44.7, 46.5, 52.8
Chloro bis (di-isopropyl dithiocarbamato) Fe(III)	26.5
Chloro bis (N,N diethyl dithiocarbamato) Fe(III)	33.0, 38.8, 43.6
Bromo bis (pyrrolidyl dithiocarbamato) Fe(III)	27.0, 29.8

FIGURE CAPTIONS

Fig. 1. Zero field eigenvalues in units of D as a function of $\lambda = E/D$ for the spin Hamiltonian Eq. (2) with S = 3/2, 2, and 5/2. The m_s values of the states for $\lambda = 0$ are indicated at the left, and doubly-degenerate eigenvalues are identified at the right.

Fig. 2. Eigenvalues in units of D as a function of $H' = g\mu_B H/D$ for the spin Hamiltonian Eq. (2) with $\lambda = 0$ and S = 3/2, 2, and 5/2. The dotted curves are for $H \parallel z$, the solid curves for $H \perp z$. The m_s and m_H values of the states for large H' and $D > 0$ are indicated at the right.

Fig. 3. The polycrystalline absorption coefficient $\bar{\alpha}$ calculated from Eq. (5) for S = 3/2, $D < 0$, $D/kT = 0.8$, $\lambda = 0.036$, and various values of H'. The contribution of each transition to the total lineshape is shown in the upper curve; the transition index n is identified in the inset diagram. The lower composite plot indicates the variation of the total lineshape with H'. The scale of $\bar{\alpha}$ for H' = 0 has been reduced for clarity.

Fig. 4. Raw data and ratios as obtained from the Fourier spectrometer for ~ 200 mg of polycrystalline iodo deuteroporphyrin IX dimethyl ester Fe(III).

Fig. 5. Absolute configuration of the ligands surrounding the Fe(III) ion in Ferrichrome A, after Zalkin, Forrester, and Templeton, Ref. 16.

Fig. 6. Theoretical and experimental absorption coefficients for polycrystalline ferrichrome A at H = 52.2 KOe and T = 4.2°K. The points are the average of three experimental lineshapes, and the bars indicate plus or minus one standard deviation. The experi-

mental points have been corrected for small linear variations of the background, due to instrumental effects, obtained from the value of the absorption coefficient at frequencies below and above the plotted range. The solid curve is the theoretical bare absorption coefficient for g=2.0, D= -0.27 cm^{-1} and $|\lambda| = 0.25$; the dotted curve was calculated for D= +0.27 cm^{-1} and $\lambda = 0.333$. The frequencies of the absorption edges and maxima, defined in the text, are indicated for the solid curve.

Fig. 7. Probable molecular structure for Fe(III) $(S_2CNR_2)_3$, after Ewald, Martin, Ross, and White, Ref. 20. R is an alkyl group.

Fig. 8. Data for polycrystalline tris (pyrrolidyl dithiocarbamato) Fe(III) at T=4.2°K, obtained from the far-infrared spectra. The points are the frequencies of the observed absorption maxima, and the bars show the approximate width of the absorptions. Smooth curves have been drawn through the points.

Fig. 9. Approximate structure of the porphyrin skeleton and local coordination of the iron atom in heme compounds.

Fig. 10. Data for a sample of ~ 150 mg of polycrystalline fluoro protoporphyrin IX dimethyl ester Fe(III) at T = 4.2°K. The points indicate the position of the experimental maxima, and the bars the approximate width of the observed absorptions. The curves are the transition frequencies predicted by the spin Hamiltonian in Eq. (2) for D = +5.0 cm^{-1}, $\lambda = 0$, and $\underset{\sim}{H} \perp \hat{z}$; the dotted portions indicate weaker absorption due to the effects of the Boltzmann term P_n(T=4.2°K). The approximate spectral bandpass of the sample is indicated by the range of frequencies plotted.

Fig. 11. Data for a sample of ~ 100 mg of polycrystalline azido deuteroporphyrin IX dimethyl ester Mn(III) at T = 4.2°K. The format is the same as that used in previous figures. The curves are the predicted transition frequencies calculated from Eq. (2) for D= -3.08 cm^{-1}, $\lambda = 0$, and $\underset{\sim}{H} \perp \hat{z}$.

Fig. 12. Data obtained from the far-infrared spectra of paste samples containing ~ 1 gm of the fluoro derivatives of ferrihemoglobin (HbF) and ferrimyoglobin (MbF) at 4.2°K. The points are the frequencies of the measured absorption maxima, and the bars indicate the approximate width of the observed absorptions. The curves are the frequencies of strong transitions calculated from Eq. (2) for $\lambda = 0$, $\underset{\sim}{H} \perp \hat{z}$, and D = +6.30 cm^{-1} (HbF), and D = +5.94 cm^{-1}(MbF); the dotted portions indicate regions of weak absorption. The range of frequencies plotted indicates the approximate spectral bandpass of the samples.

Fig. 13. Data from the far-infrared spectra of ~ 1 gm of polycrystalline met-myoglobin at 4.2°K. The points are the average values of the frequencies of the measured absorption maxima for eight experimental runs. The curves are the calculated transition frequencies using Eq. (2) with $\lambda = 0$, $\underset{\sim}{H} \perp \hat{z}$, and the values of D indicated at the right.

Fig. 14. Molecular structure of $(R_2NCS_2)_2$ Fe(III) X, after Hoskins, Martin, and White, Ref. 47. R is an alkyl group, and X is a halogeno ligand.

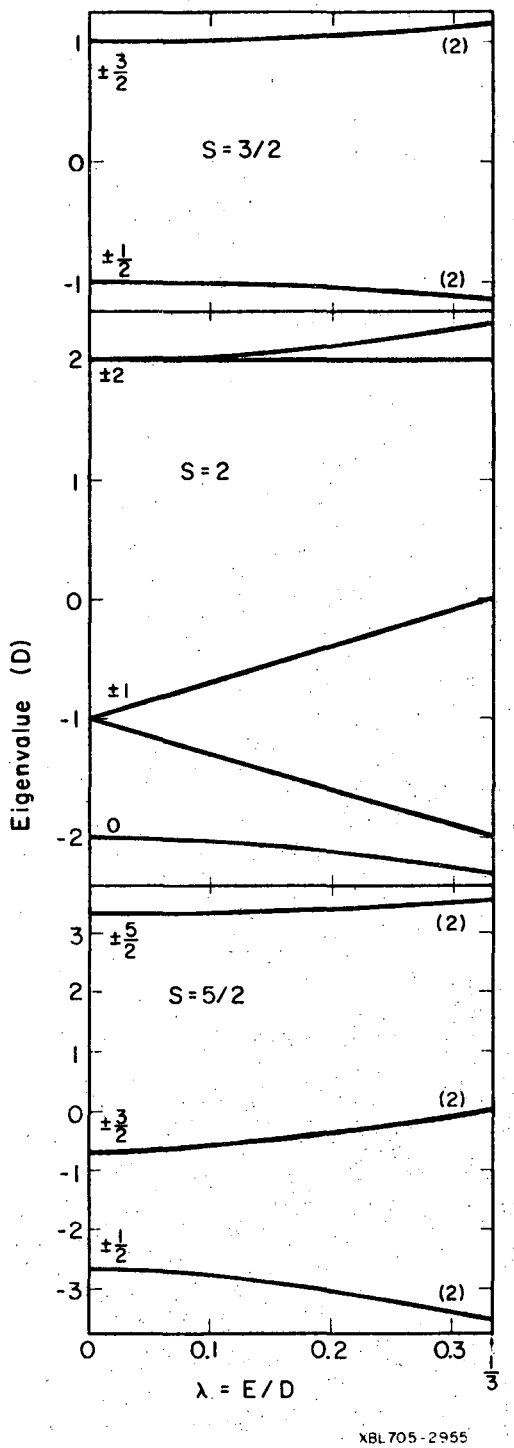
Fig. 15. Data obtained from the far-infrared spectra of ~ 100 mg of polycrystalline bis (di-isopropyl dithiocarbamato) Fe(III) chloride. The points are the frequencies of the observed absorption maxima and

the bars indicate the approximate width. The curves plot the principle maxima of the polycrystalline absorption coefficient, calculated using the program described in Section II-C for $D = -2.35\ cm^{-1}$, $\lambda = 0.036$, and $T = 4.2°K$. The dotted portions indicate regions of weak absorption.

Fig. 16. Data for the ordered sample of bis (N,N diethyl dithiocarbamato) Fe(III) bromide, described in the text at 4.2°K. The points are the frequencies of the observed maxima and the bars indicate the approximate width. The curves are the transition frequencies calculated from Eq. (2) for $\theta_H \sim 3\pi/8$, $D = +7.50\ cm^{-1}$, and $\lambda = 0.067$.

Fig. 17. Data obtained from the far-infrared spectra of a polycrystalline sample of bis (N,N diethyl dithiocarbamato) Fe(III) chloride in the ordered state at T = 1.3°K. The points are the frequencies of the single sharp absorption maxima observed, and the bars indicate the experimental width. The line is $\nu = 2\mu_B H + 3.85\ cm^{-1}$.

Fig. 18. Far-infrared transmission spectrum of ~ 50 mg of polycrystalline $((C_2H_5)_2NCS_2)_2$ Fe(III) Br at T=4.2°K and H=0. The absorption due to the zero-field splitting is indicated at $\Delta = 15.1\ cm^{-1}$, and the transmission with no sample in place is given by the dotted curve.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S = 3/2
±3/2
±1/2
(2)
S = 2
±2
±1
0
S = 5/2
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±3/2
±1/2
λ = E/D
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0.2
0.3
1/3
XBL705-2955

Fig. 1

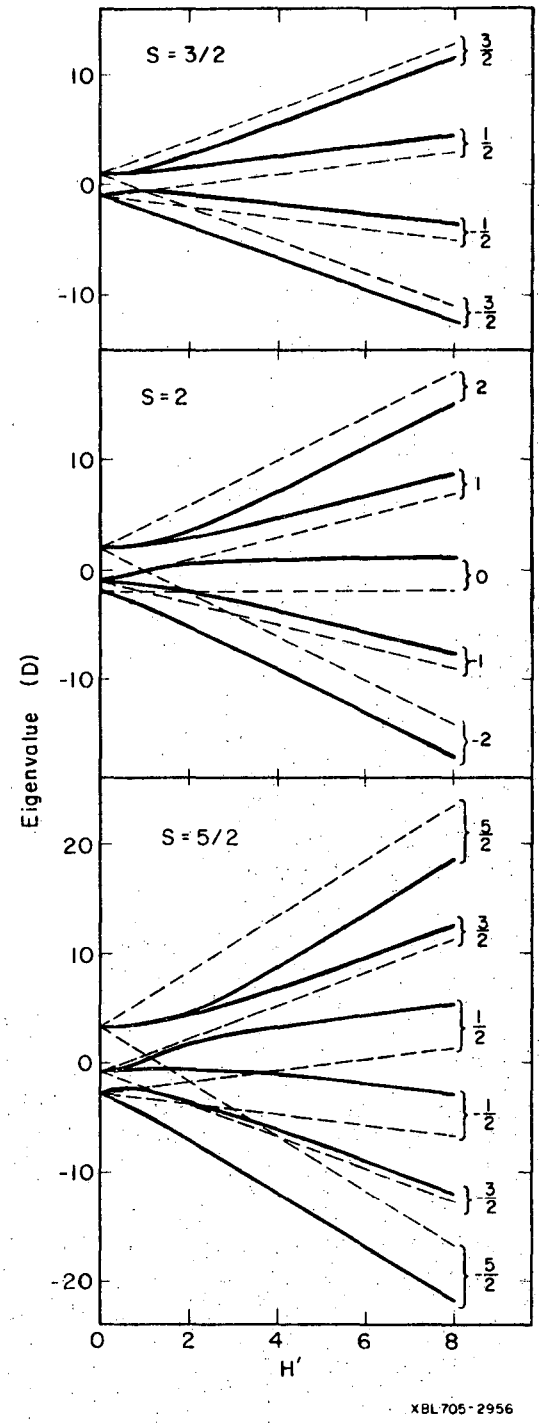
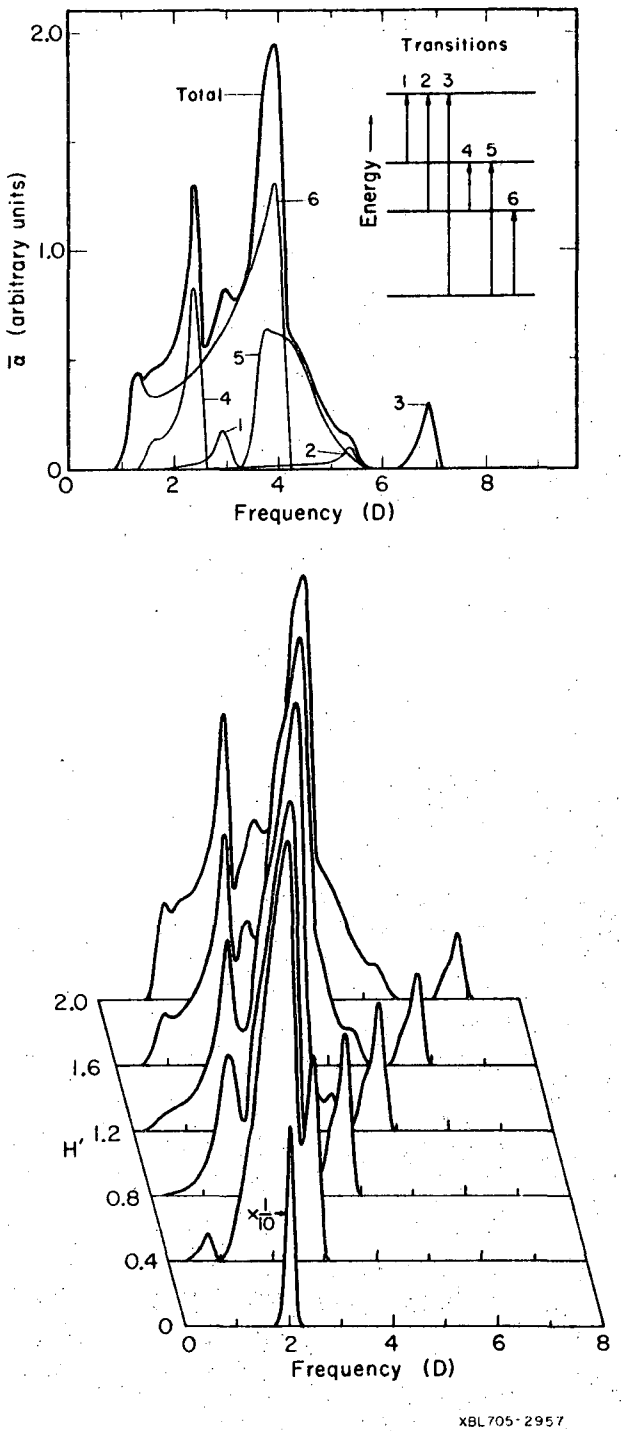


Fig. 2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4 5
6
Energy
Total
6
5
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1
2
3
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1.0
0
$\bar{\alpha}$ (arbitrary units)
0 2 4 6 8
Frequency (D)
2.0
1.6
H' 1.2
0.8
0.4
0
$\times\frac{1}{10}$
0 2 4 6 8
Frequency (D)
XBL705-2957

Fig. 3

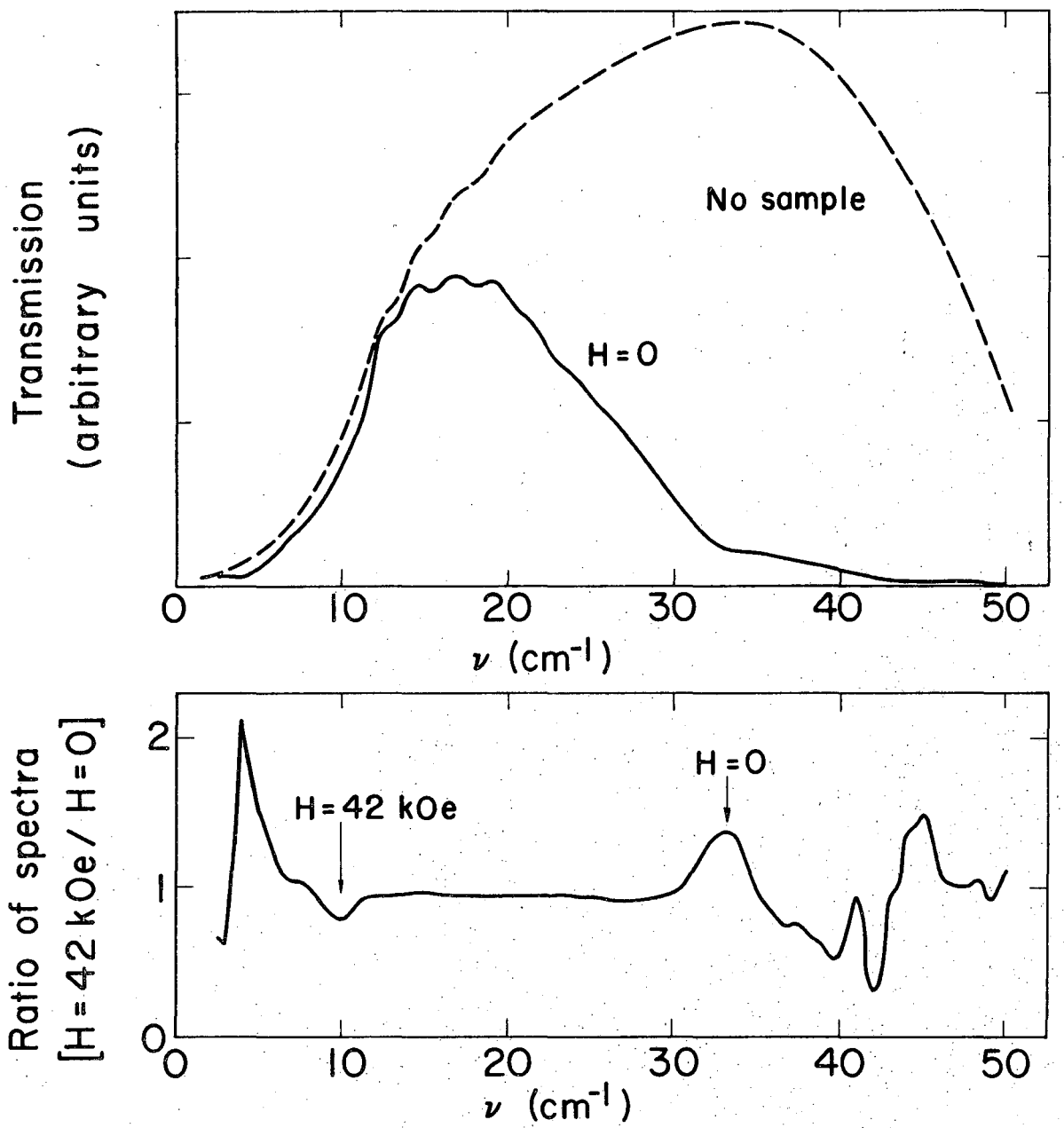

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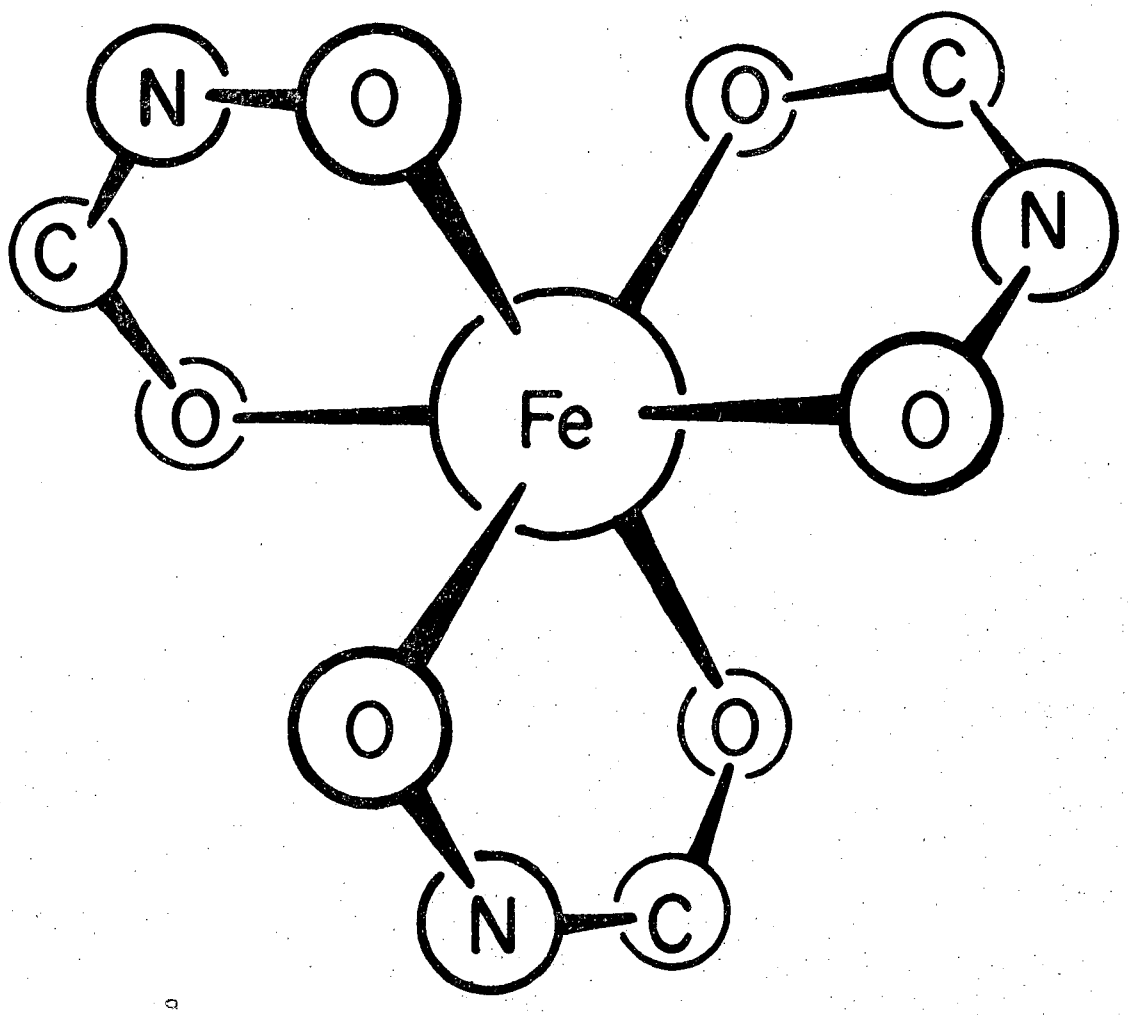
Fig. 4



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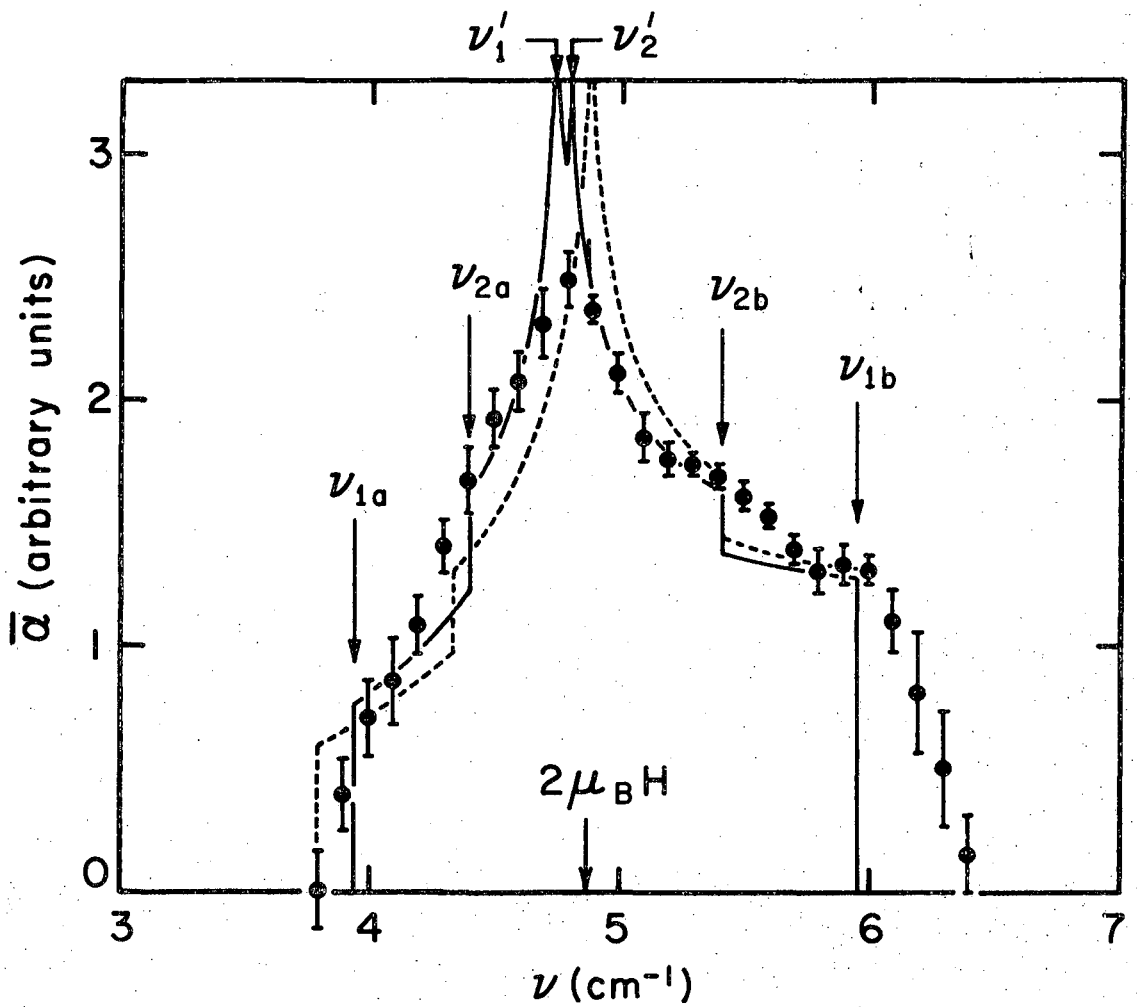

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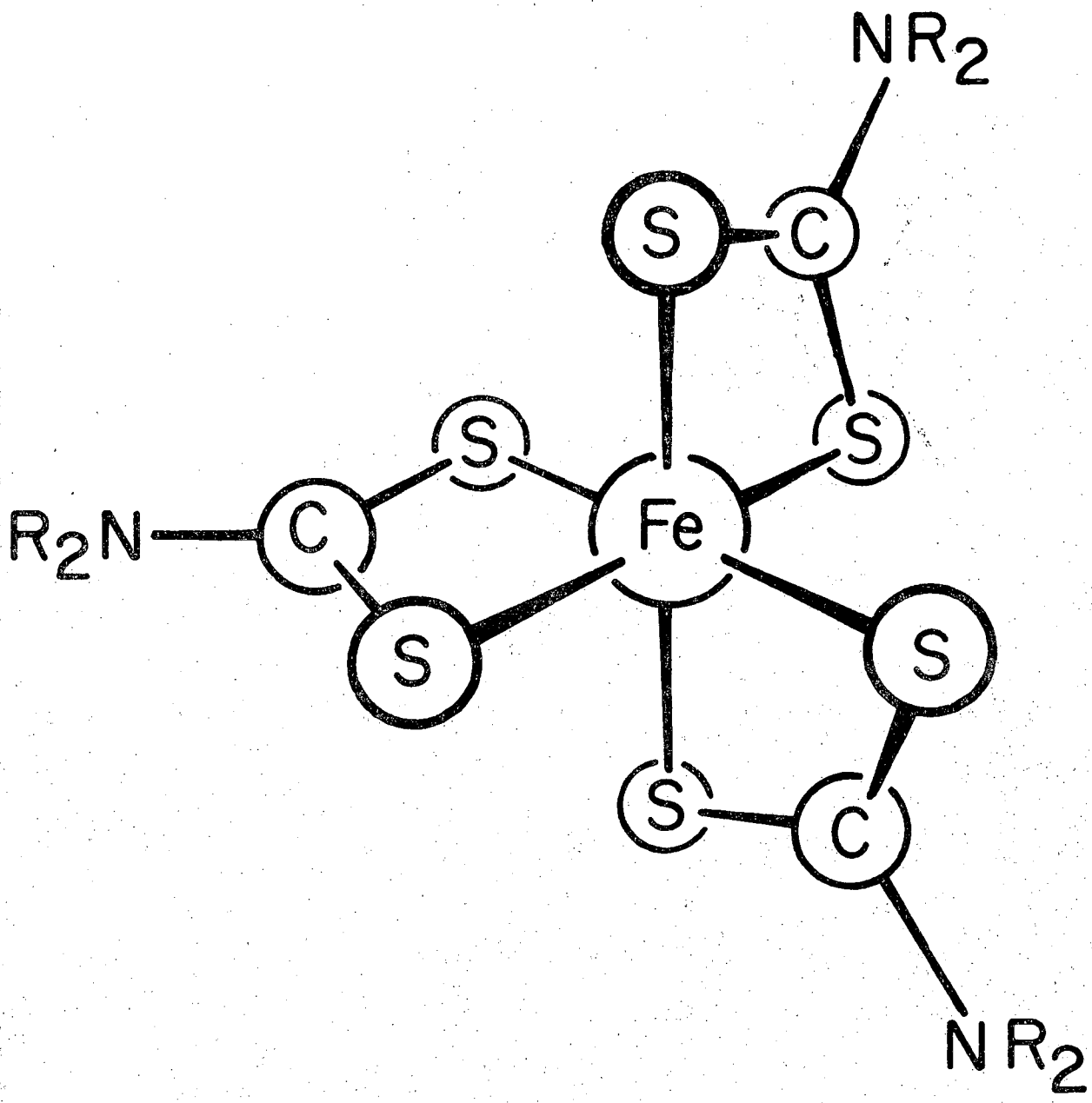

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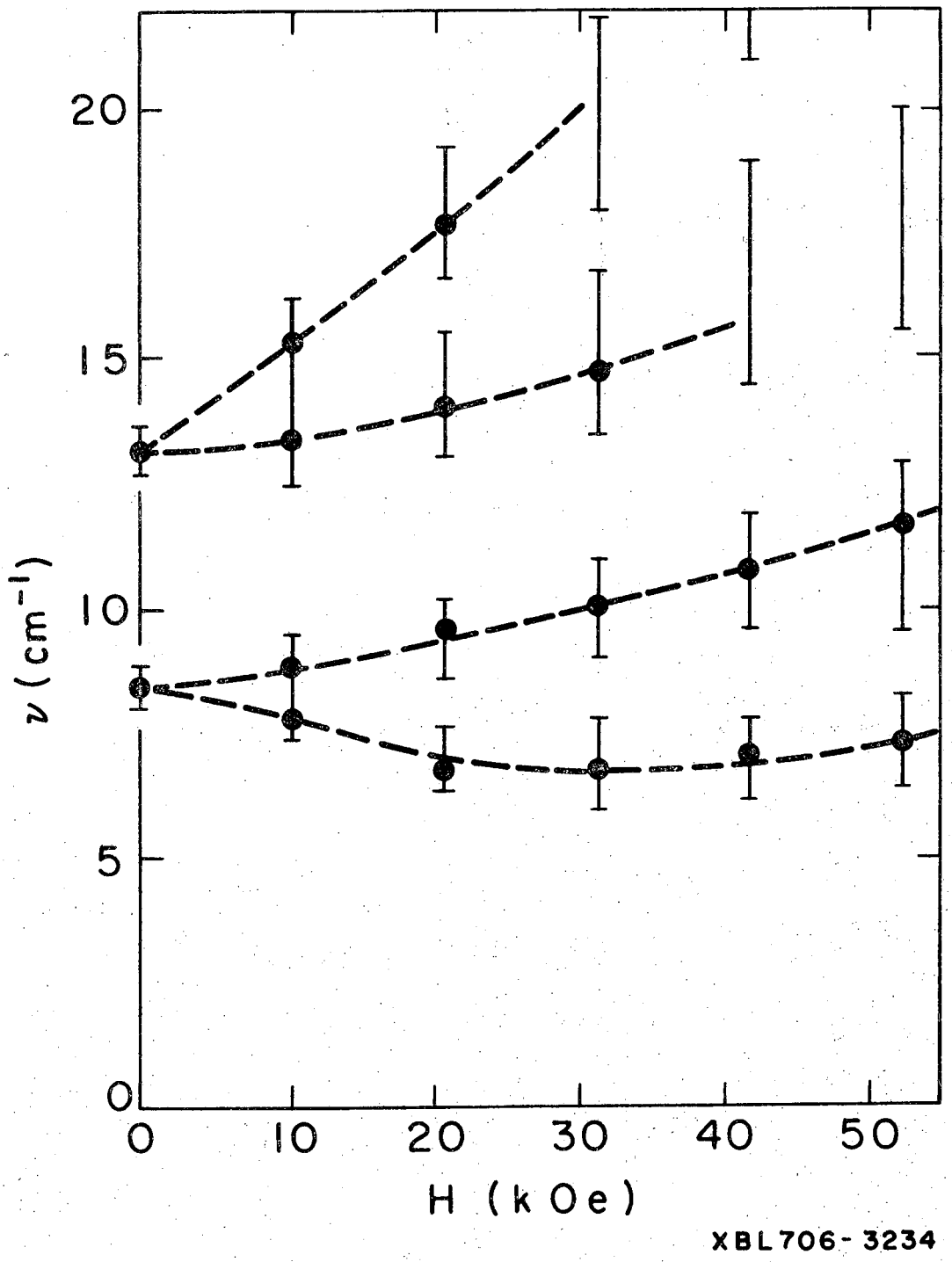

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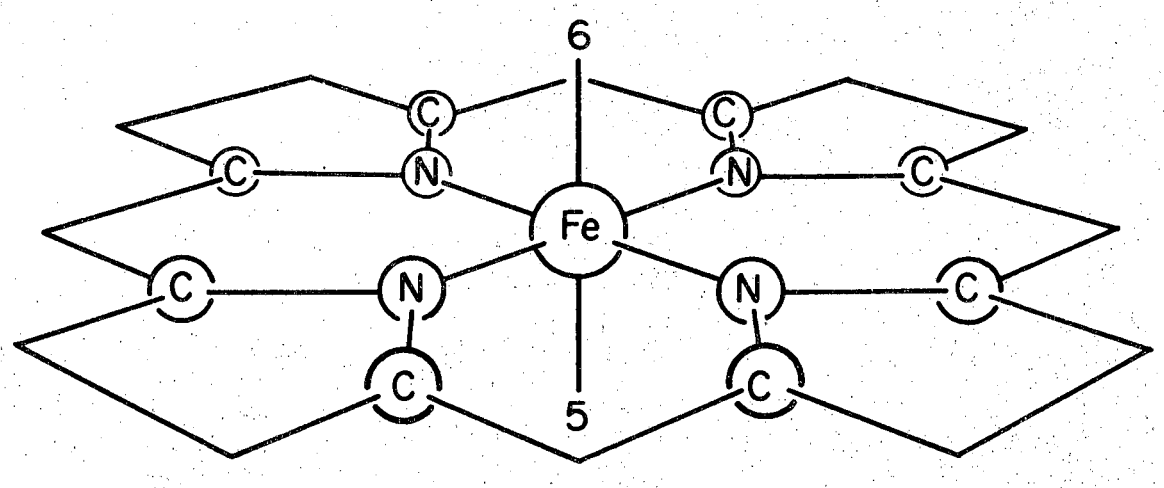

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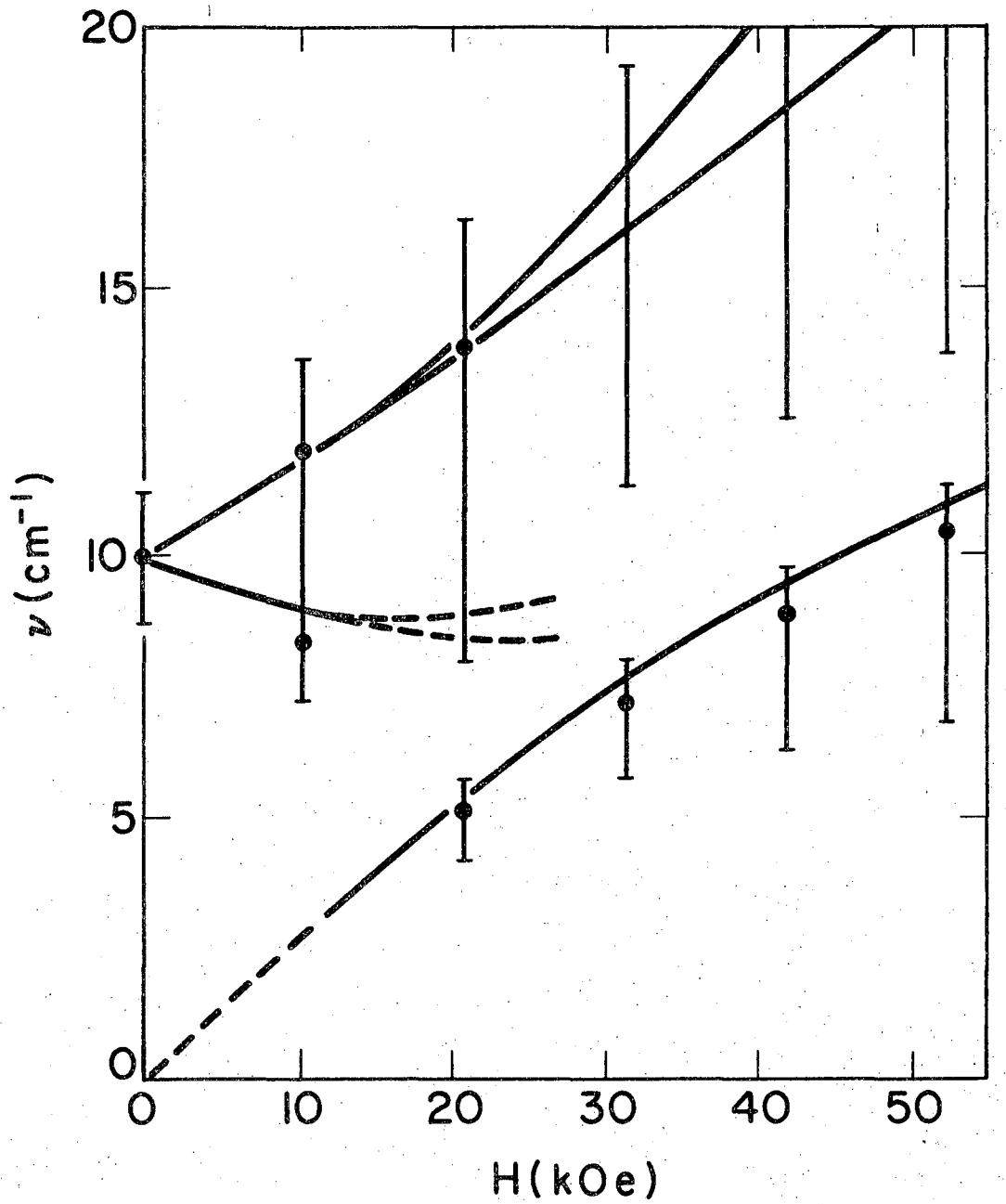
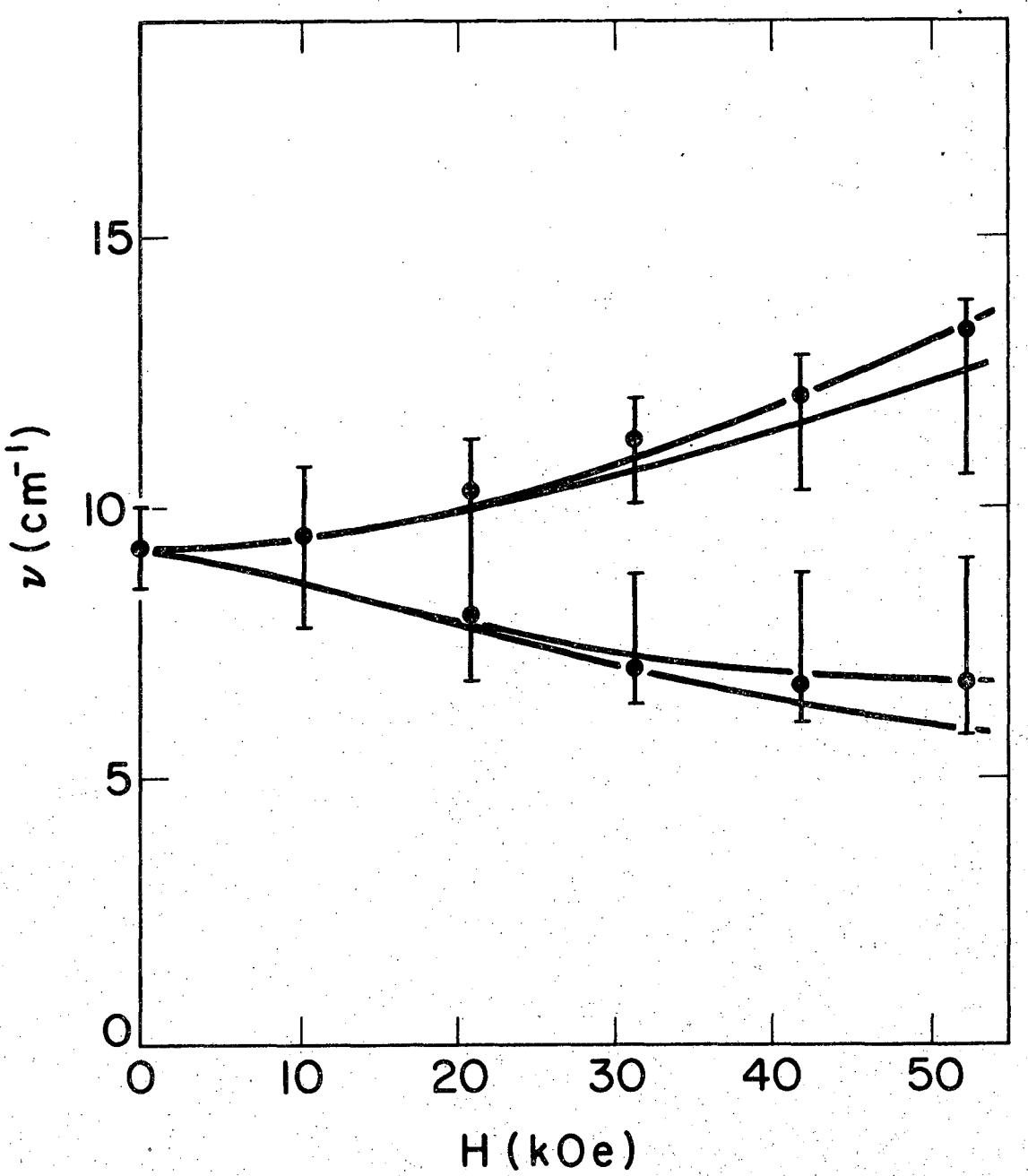
Fig. 9



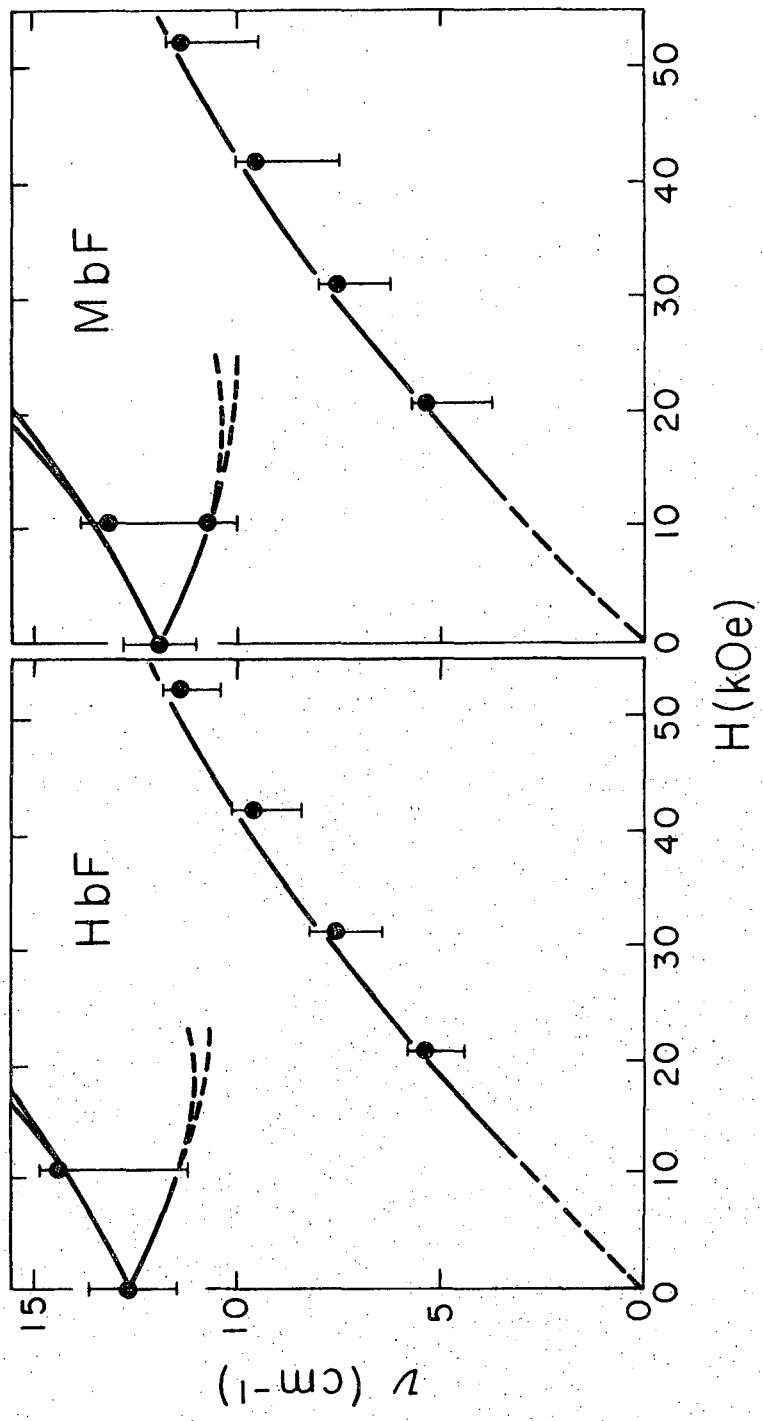
XBL706-3181

Fig. 10



XBL706-3182

Fig. 11



XBL706-3236

Fig. 12

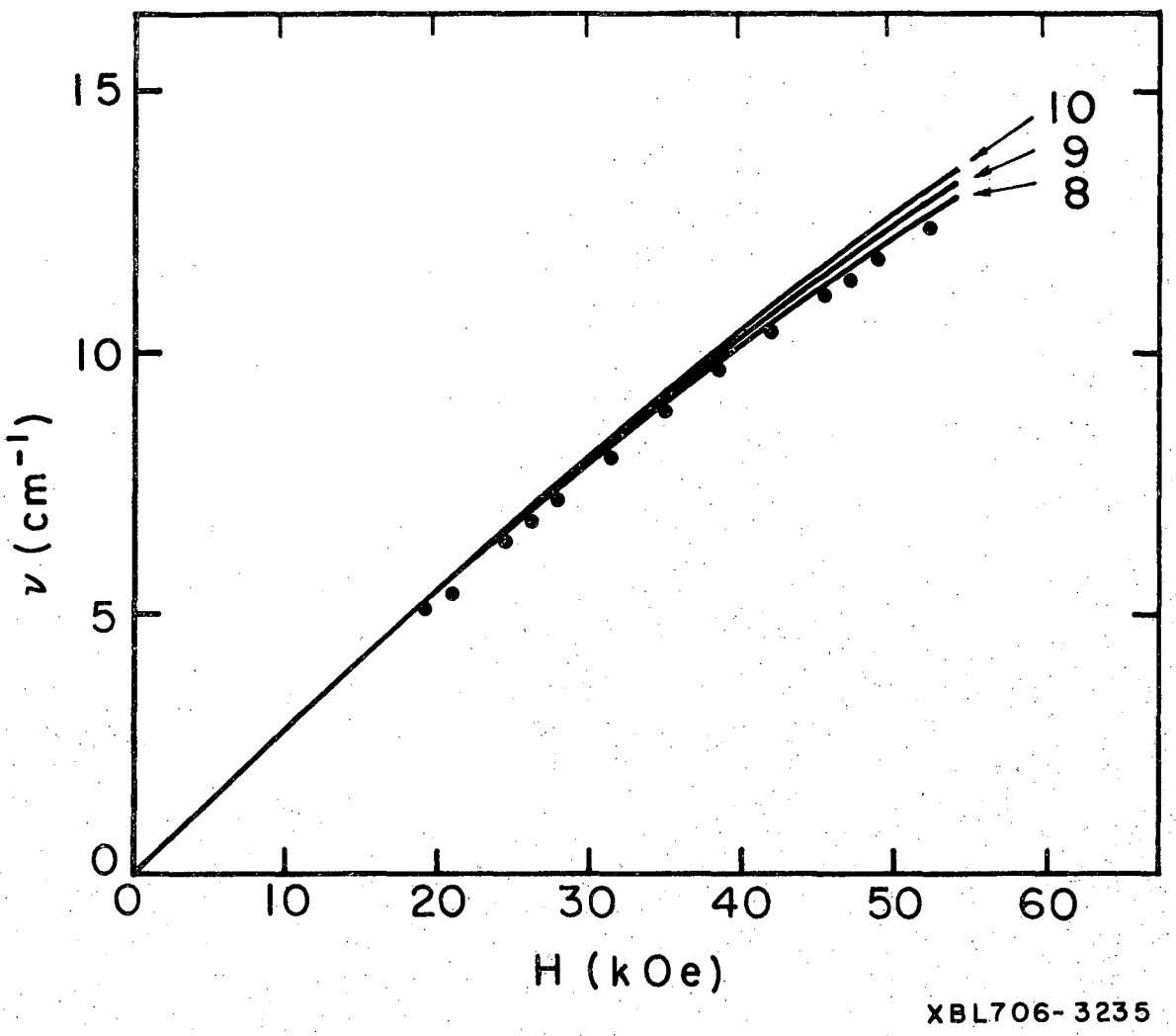
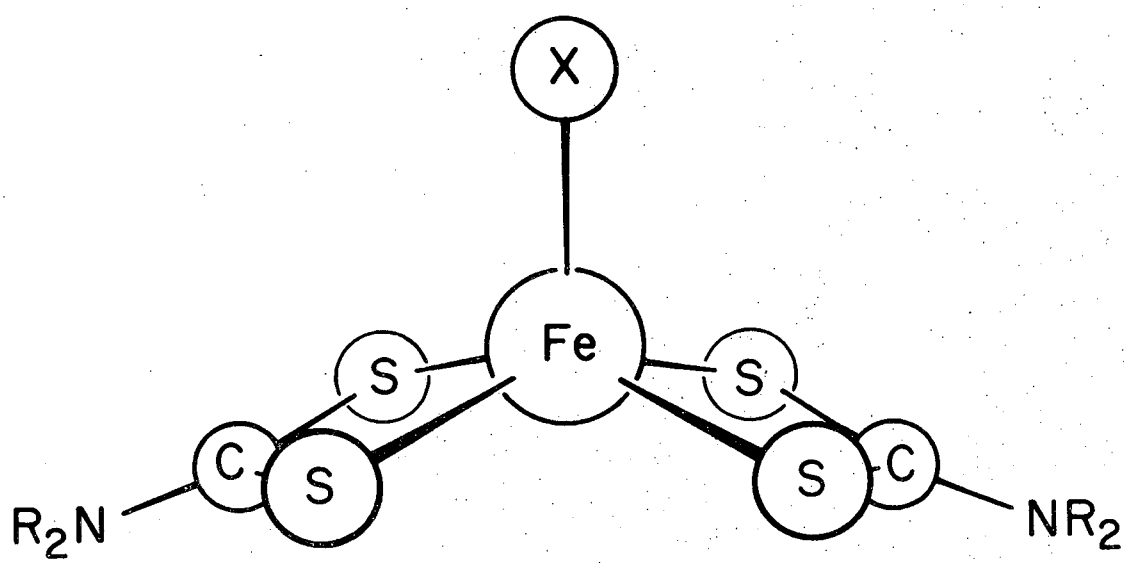
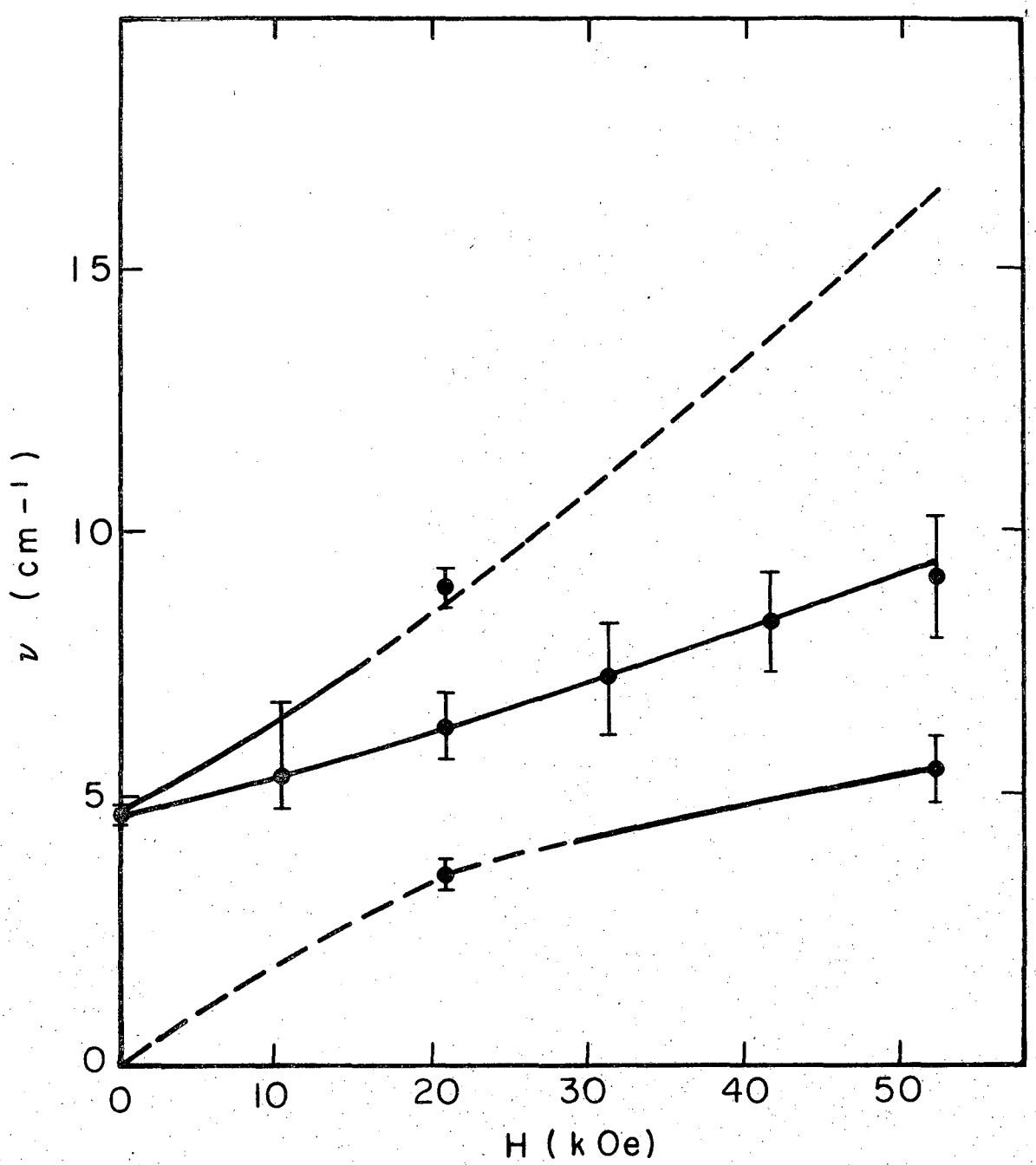


Fig. 13



XBL705-2959

Fig. 14



XBL6912-6385-A

Fig. 15

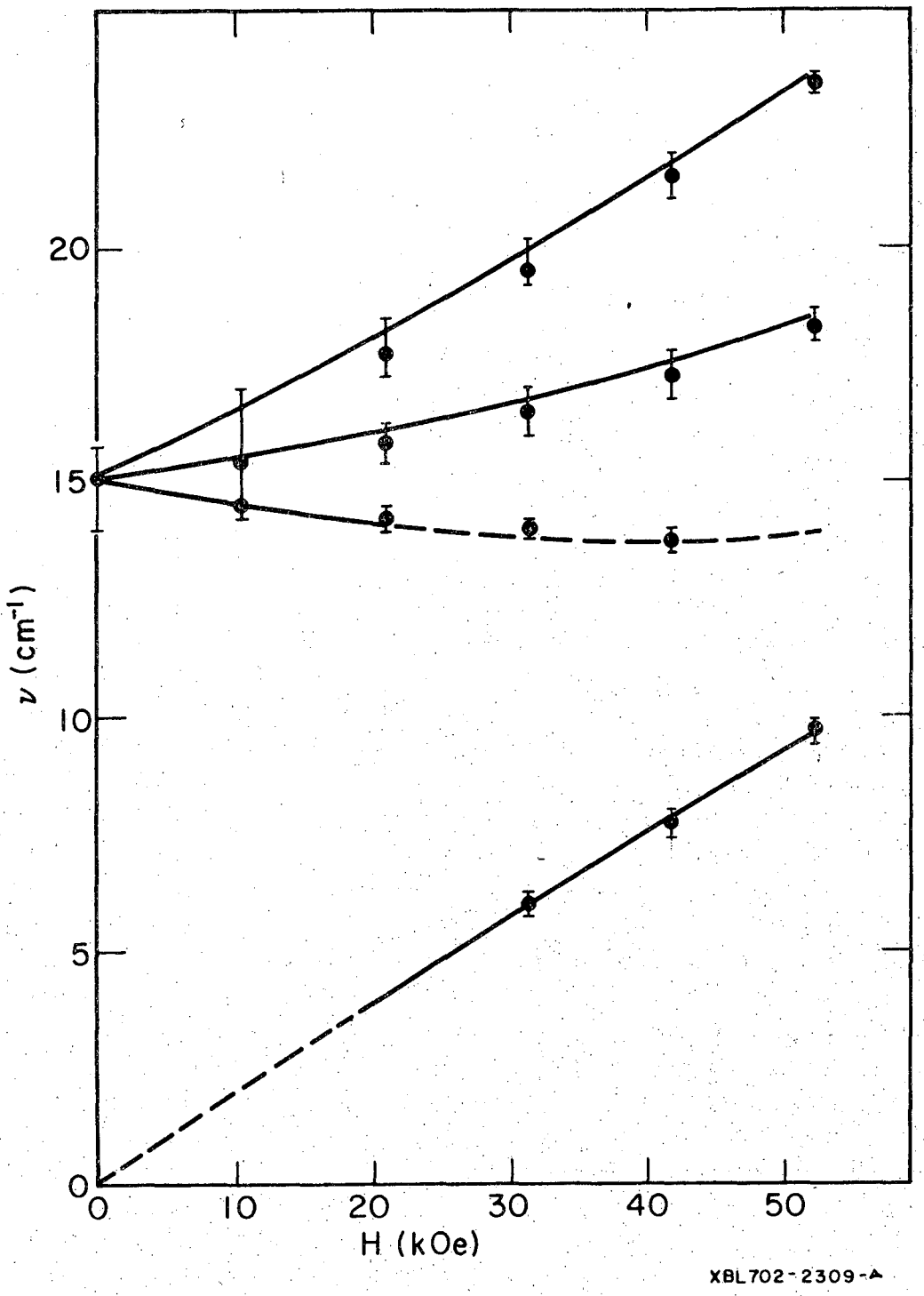
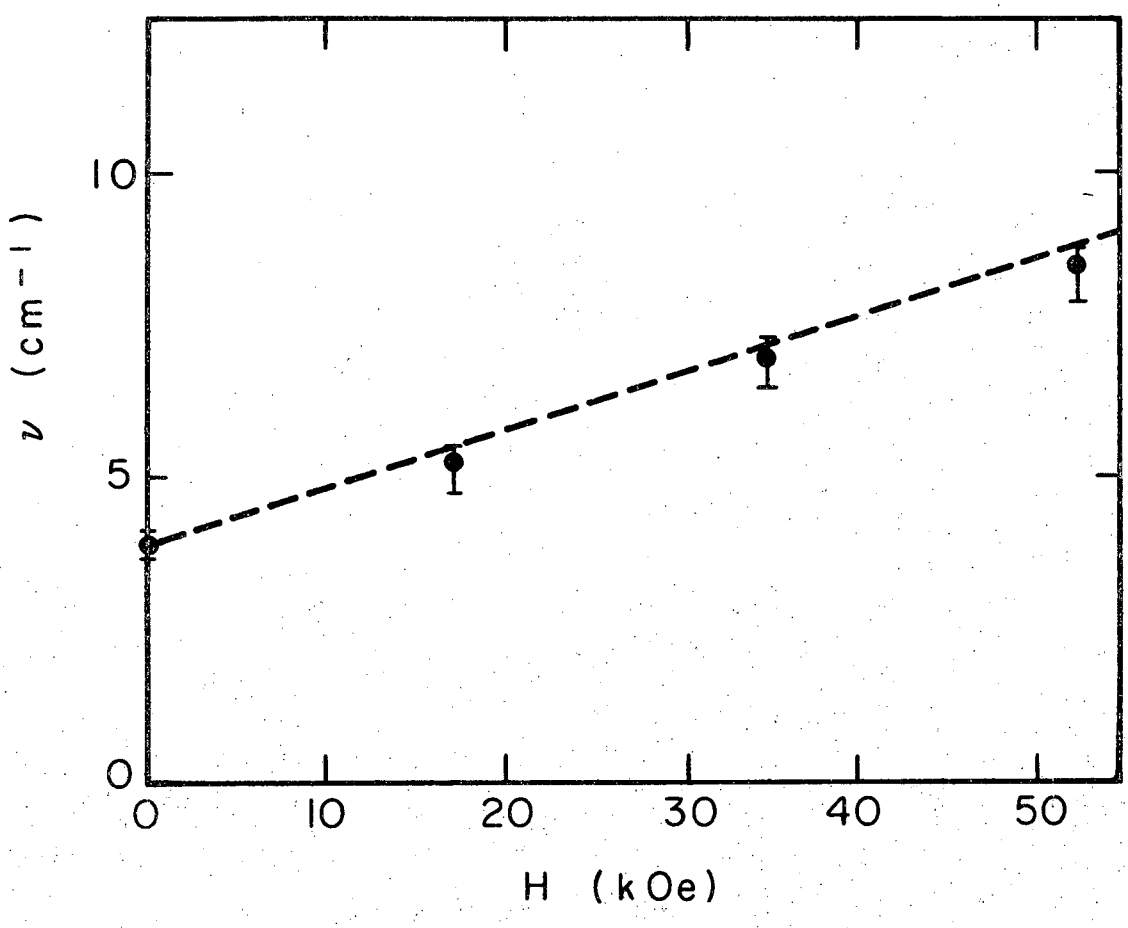


Fig. 16



XBL6912-6386-A

Fig. 17

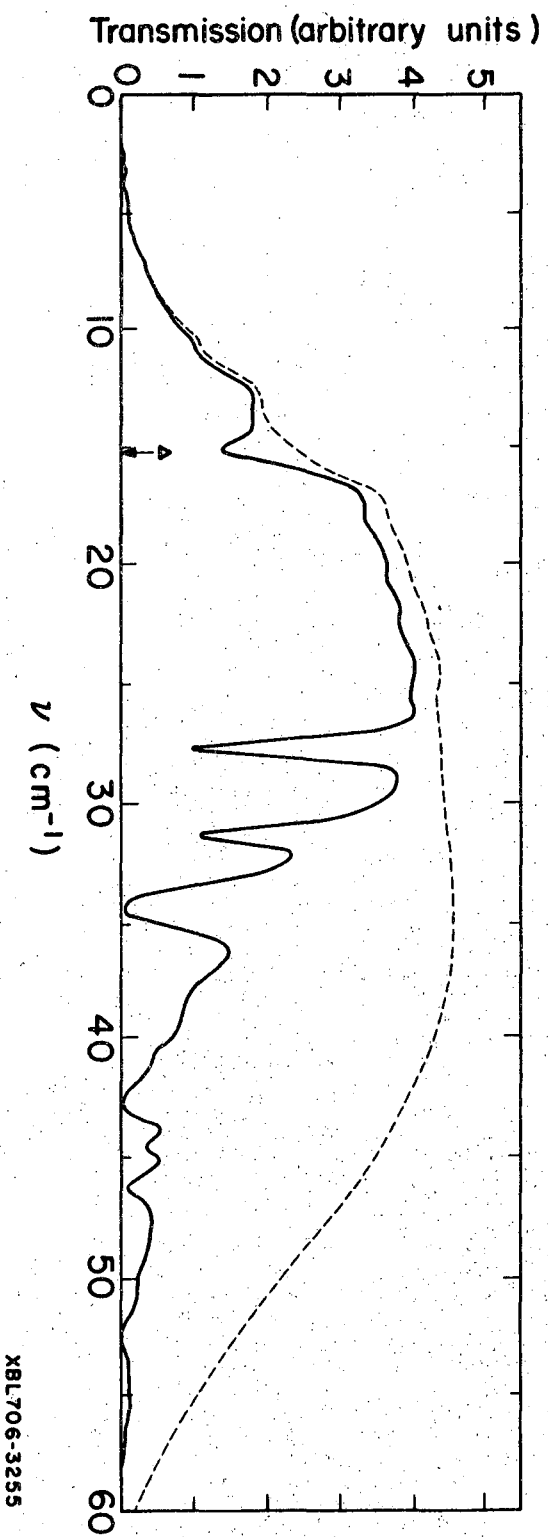

Transmission (arbitrary units)
0
1
2
3
4
5
0
10
20
30
40
50
60
ν (cm⁻¹)
Δ
XBL706-3255

Fig. 18

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