DL-phenylalanines by a method analogous to that described in this communication.[5] The use of acetyl-DL-phenylalanine in preference to other acyl-DL-phenylalanines is based upon previous studies on the role of the acyl group in determining the stereochemical course of the reaction under discussion[5,6] and the fact that acetyl-DL-phenylalanine is sufficiently soluble in water to permit the use of relatively concentrated solutions. *p*-Toluidine was chosen as the base because it is a solid, is available in a relatively pure state, and is relatively stable in contact with air.

The authors wish to acknowledge the assistance of W. H. Schuller who checked the procedure given below.

Experimental[7]

Reagents.—Acetyl-DL-phenylalanine, lustrous plates, m.p. 152–154°, was prepared in yields of 93–96% from DL-phenylalanine (Dow) by acetylation of 0.8 mole of the amino acid with 2.4 moles of acetic anhydride and 1.6 moles of sodium hydroxide at a temperature below 10°. Thirty grams of finely ground papain (Wallerstein) was stirred with 150 ml. of water for 3 hours at 5°, the suspension centrifuged for 15 minutes at 2000 r.p.m. and the supernatant solution reserved for use. *p*-Toluidine (Merck and Co., Inc.) was used without further purification.

Acetyl-L-phenylalanine-*p*-toludide (I).—To 2 l. of a 0.5 *M* acetic acid–0.5 *M* sodium acetate buffer containing 9 g. of L-cysteine hydrochloride was added 155.5 g. (0.75 mole) of acetyl-DL-phenylalanine, the suspension warmed to 50° to effect complete solution, 80 g. (0.75 mole) of *p*-toluidine and the above enzyme solution added, the total volume brought to 3 liters with the acetate buffer, and the clear solution, *p*H 4.6, incubated at 40° for 7 days. The reaction mixture was maintained at 5° for 2 hours prior to the collection of the precipitate which was washed with 1 l. of water and air-dried to give 102–106 g. (92–95%) of I, m.p. 215–217°. This product was dissolved in 1.25 l. of hot 96% ethanol, the hot solution filtered, the filtrate held at 5° overnight, the precipitate collected, washed with 500 ml. of cold 96% ethanol, and air-dried to give 86–89 g. of I, m.p. 219°; $[\alpha]^{25}D$ +35 ± 1° (*c*, 4 in pyridine). *Anal.* Calcd. for $C_{18}H_{20}O_2N_2$ (296): C, 73.0; H, 6.8; N, 9.5. Found: C, 72.8; H, 6.7; N, 9.4. An additional 9–10 g. of I, m.p. 219°, was recovered from the mother liquor to give a total yield of recrystallized I of 86–89%.

L-Phenylalanine (II).—A suspension of 86 g. of recrystallized I in 1100 ml. of 20% hydrochloric acid was heated under reflux for 16 hours, the clear solution evaporated to dryness *in vacuo*, the residue dissolved in 300 ml. of water, again evaporated to dryness, the residue dissolved in 250 ml. of water and 450 ml. of 28% aqueous ammonia cautiously added to this solution. After the reaction mixture was held at 5° for 2 hours the precipitated *p*-toluidine was collected, washed with cold water, the filtrate and washings combined, the solution extracted with two 300-ml. portions of chloroform, the volume of the aqueous phase reduced to *ca.* 500 ml. by boiling, the solution cooled to 5° (2 hours), the precipitate collected, washed successively with 100 ml. of water and 40 ml. of 96% ethanol and air-dried to give 28 g. of II, lustrous flat plates; $[\alpha]^{25}D$ −34 ± 1° (*c*, 2 in water). Concentration of the mother liquor gave two additional crops of II of 6 and 4 g., respectively, or a total yield of 38 g. (82%).

D-Phenylalanine (III).—The filtrate remaining after the collection of I was evaporated *in vacuo* below 50° to one-half of its original volume, acidified with 120 ml. of concentrated hydrochloric acid, stored at 5° overnight, the crystalline precipitate collected, washed with 200 ml. of cold water and recrystallized from 20% aqueous methanol to give 50–65 g. (77–84%) of acetyl-D-phenylalanine (IV), m.p. 162–164°; $[\alpha]^{25}D$ −32 ± 1° (*c*, 2 in methanol). A suspension of 50 g. of IV in 300 ml. of 20% hydrochloric acid was heated under reflux for 5 hours, the clear solution evaporated to dryness, the residue dissolved in 300 ml. of water, 100 ml. of 28% aqueous ammonia added, the solution boiled to remove excess ammonia, decolorized with 5 g. of Norite, the clear colorless filtrate stored at 5° overnight, the crystalline precipitate collected, washed successively with 100 ml. of water and 50 ml. of 96% ethanol, and dried to give 27 g. of III; $[\alpha]^{25}D$ +34 ± 1° (*c*, 1 in water). A second crop of 3 g. of III was obtained from the mother liquor to give a total yield of 30 g. (77%).

The average over-all yields of D and L-phenylalanine from DL-phenylalanine were 59 and 68%, respectively.

CALIFORNIA INSTITUTE OF TECHNOLOGY
PASADENA 4, CALIFORNIA
RECEIVED AUGUST 7, 1950

---

[5] E. L. Bennett and C. Niemann, THIS JOURNAL, **72**, 1800 (1950).

[6] E. L. Bennett and C. Niemann, *ibid.*, **72**, 1798 (1950).

[7] All melting points reported are corrected.

On the Interaction between Hexacyanatoferrate(III) Ions and (a) Hexacyanatoferrate(II) or (b) Iron(III) Ions[1a,2]

BY JAMES A. IBERS AND NORMAN DAVIDSON[1b]

In view of the occurrence of non-additive light absorption ("interaction absorption") in hydrochloric acid solutions containing iron(II) and (III),[3] it was of interest to determine whether the hexacyanatoferrate(II) and (III) ions exhibited this same effect. The present note reports experiments showing that the answer to this question is in the negative. At the same time we have examined the well-known brown coloration of solutions containing iron(III) ions and hexacyanatoferrate(III) ions and have evaluated the equilibrium constant for the formation of the uncharged complex species, $FeFe(CN)_6$, responsible for this color.

Figure 1 shows the absorption spectra of 0.1 *F* $Fe(CN)_6^{-3}$ and 0.1 *F* $Fe(CN)_6^{-4}$ solutions buffered at a *p*H of 6.8 (where the ratio $[HFe(CN)_6^{-3}]/[Fe(CN)_6^{-4}]$ is of the order[4] of 0.003). The data agree fairly well with those given by Kortüm.[5] A 1:1 mixture of these two solutions had optical densities in the wave length range 270–350 mμ which agreed to within 1% with those predicted by Beer's law. No interaction absorption was observed when the ionic strength of the solution was raised from 1.9 to 4.4 by making it 2.5 *F* in potassium chloride. It was thought that such a high ionic strength might diminish the electrostatic repulsion between the $Fe(CN)_6^{-4}$ and $Fe(CN)_6^{-3}$ ions.

It is noteworthy that, by virtue of the short light paths used, the absorption of a moderately concentrated solution of $Fe(CN)_6^{-4}$ and $Fe(CN)_6^{-3}$ has been measured in the wave length region close to the strong ultraviolet "electron-transfer" absorption bands of the components; this is the region where interaction absorption, when it occurs, is most marked.[6,7] One may conclude that there is little or no optical interaction absorption between $Fe(CN)_6^{-4}$ and $Fe(CN)_6^{-3}$ in aqueous solution. It has already been suggested[3] that interaction absorption

---

[1a] Research supported by the O. N. R. under Contract N6onr-244.

[1b] To whom inquiries concerning this article should be addressed.

[2] For a table of the extinction coefficients of solutions of $K_3Fe(CN)_6$, $K_4Fe(CN)_6$, $Fe(ClO_4)_3$ and $FeFe(CN)_6$ order Document 2944 from American Documentation Institute, 1719 N Street, N. W., Washington 6, D. C., remitting $0.50 for microfilm (images 1 inch high on standard 35 mm. motion picture film) or $0.50 for photocopies (6 × 8 inches) readable without optical aid.

[3] H. McConnell and N. Davidson, THIS JOURNAL, **72**, 5557 (1950).

[4] I. M. Kolthoff and W. J. Tomsicek, *J. Phys. Chem.*, **39**, 955 (1935).

[5] G. Kortüm, *Z. physik. Chem.*, **B33**, 254 (1939).

[6] J. Whitney and N. Davidson, THIS JOURNAL, **71**, 3809 (1949).

[7] H. McConnell and N. Davidson, *ibid.*, **72**, 3168 (1950).

between iron(II) and (III) requires a bonding "bridge" between atoms of the two oxidation states; such a bridge is not possible for the case at hand.

Measurements of the variation of the optical density with concentration of Fe^{+3} and $Fe(CN)_6^{-3}$ were made at the wave lengths 475, 500 and 525 mμ for solutions at a total ionic strength of 1.00 ± 0.02 (moles/liter) and at $[H^+] = 0.102$ *M*. The data have been interpreted assuming the approximate validity of the mass action law at the constant ionic strength used. Let *a* and *b* be the concentrations of free Fe^{+3} and $Fe(CN)_6^{-3}$, and a_0 and b_0 the formal (total) concentrations of these components. Let c_{ij} be the concentrations of the complexes $[Fe_i^{+3}(Fe(CN)_6)_j^{-3}]^{3(i-j)}$, ϵ_{ij} their extinction coefficients, and K_{ij} their formation constants, so that $c_{ij} = K_{ij}a^ib^j$.

One set of results was obtained for a series of solutions in which $a > b$ so that $a \cong a_0$, and so that only the complexes $[Fe_i^{+3}(Fe(CN)_6)_1^{-3}]^{3i-3}$ are likely to have been present. Let D_c be the optical density for a 1.0 cm. light path of a solution due to the complexes present, *i.e.*, corrected for the light absorption by Fe^{+3} and $Fe(CN)_6^{-3}$. Then

$$D_c = \sum_i \epsilon_{i1}c_{i1} = (\Sigma\epsilon_{i1}K_{i1}\, a^ib_0)/(1 + \Sigma K_{i1}\, a^i)$$

and

$$(ab_0/D_c) = (1 + \Sigma K_{i1}\, a^i)/(\Sigma\epsilon_{i1}K_{i1}\, a^{i-1}) \qquad (1)$$

Assuming that $\epsilon_{11}K_{11} > \epsilon_{i1}K_{i1}a_0^{i-1}$ $(i > 1)$ (*i.e.*, that the contribution to D_c from the 1:1 complex is greater than that from higher complexes), one expands the denominator of the right side of the above expression

$$(\epsilon_{11}K_{11})(ab_0/D_c) = 1 + a\left(K_{11} - \frac{\epsilon_{21}K_{21}}{\epsilon_{11}K_{11}}\right) + a^2 \times \left(K_{21} - \frac{\epsilon_{21}K_{21}}{\epsilon_{11}} + \frac{\epsilon_{21}^2K_{21}^2}{\epsilon_{11}^2K_{11}^2} - \frac{\epsilon_{31}K_{31}}{\epsilon_{11}K_{11}}\right) + \cdots \qquad (2)$$

Figure 2 exhibits plots of (ab_0/D_c) *vs.* *a* for a series of solutions at 26 ± 2° containing $Fe(ClO_4)_3$, $K_3Fe(CN)_6$, $NaClO_4$ and $HClO_4$. The non-dependence of ab_0/D_c on b_0 justifies the assumption that only complexes containing one $Fe(CN)_6^{-3}$ are important for these solutions. The linear nature of the plots demonstrates that a complex containing one Fe^{+3} ion is formed (the points at low *a* are especially convincing in this respect). The intercepts of the curves give values of $1/\epsilon_{11}K_{11}$. However, as may be seen in equation (2), the apparent linear nature of the plots for larger *a* does not prove that there is no significant formation of 2:1 complexes; it is possible that the positive and negative terms in the coefficient of a^2 approximately cancel each other. The ratio of slope to intercept for a curve is $K_{11} - (\epsilon_{21}K_{21}/\epsilon_{11}K_{11})$. There is no systematic dependence of this ratio on wave length (Table I); this *suggests* that the $\epsilon_{21}K_{21}/\epsilon_{11}K_{11}$ term is small and that the entries in the second column of the table are essentially K_{11}. Values of ϵ_{11} calculated on this basis are also given.

TABLE I

COMPLEXING OF $Fe(CN)_6^{-3}$ BY EXCESS Fe^{+3}

λ, mμ.	$\epsilon_{11}K_{11} \times 10^{-3}$	$K_{11} - (\epsilon_{21}K_{21}/\epsilon_{11}K_{11})$, liter/moles	
475	17.4	21.5	809
500	11.9	19.4	611
525	8.5	21.4	397

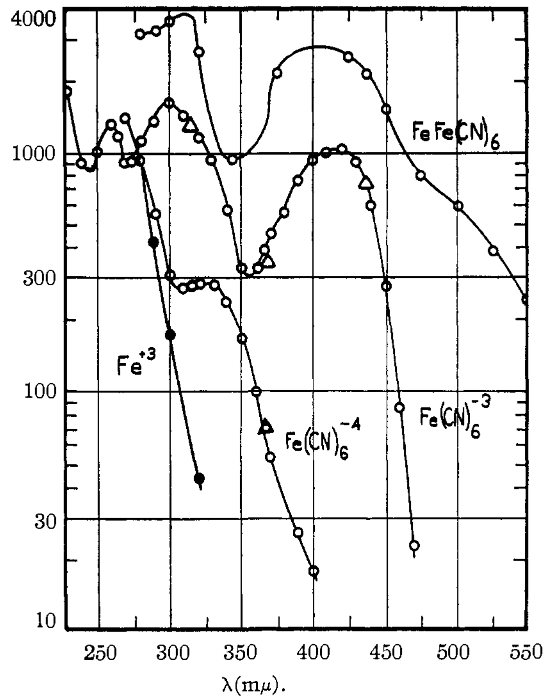

Fig. 1.—Extinction coefficients ($\epsilon cl = \log_{10}(I_0/I)$) of: 0.1006 *F* $K_3Fe(CN)_6$, *p*H 6.8 (phosphate buffer; the absorption spectrum was the same in water and at $(H^+) = 0.50$ *M*); 0.1043 *F* $K_4Fe(CN)_6$, *p*H 6.8 (Δ, date of Kortüm for the above two substances); 1.01×10^{-3} *M* $FeFe(CN)_6$ (see text); 0.0141 *F* $Fe(ClO_4)_3$ in 0.5 *M* $HClO_4$. Data obtained with 1.00, 0.10 and 0.010 cm. cells.

It is important to note that due to some decomposition the optical densities of the solutions used

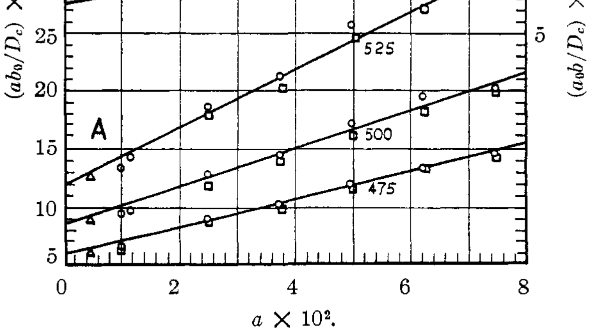

Fig. 2.—A, plot of (ab_0/D_c) *vs.* *a* for $a_0 > b_0$ in a perchlorate medium: ○, $b_0 = 1.01 \times 10^{-3}$ *F*; □, $b_0 = 5.05 \times 10^{-4}$ *F*; Δ, $b_0 = 3.36 \times 10^{-4}$ *F*. The points at $a = 4.13 \times 10^{-3}$ *M* and $a = 1.12 \times 10^{-2}$ *M* were obtained with 10 cm. cells. B, Plot of (a_0b/D_c) *vs.* *b* for $b_0 > a_0$ in a nitrate medium: ○, $a_0 = 9.6 \times 10^{-4}$ *F*; □, $a_0 = 4.8 \times 10^{-4}$ *F*.

for the above measurements changed with time; freshly prepared solutions were measured within 5 minutes of mixing and corrected to zero time using the following observed increases in D per 5-minute interval: 475 mμ, 1%; 500 and 525 mμ, 2.5%. Other small corrections were: (1) using 21 for K_{11}, a rather than its first approximation, a_0, has been used in applying equation (2); (2) the values of a and of the contribution of Fe^{III} to the optical density have been corrected for the hydrolysis[8] of Fe^{+3} to $FeOH^{++}$.

The extinction coefficients of the complex $FeFe(CN)_6$, for 280–550 mμ, given in Fig. 1 were calculated from the absorption spectrum of a solution that was 0.00705 F in $Fe(ClO_4)_3$, 0.0056 F in $K_3Fe(CN)_6$ and 0.50 M in $HClO_4$. The concentration of $FeFe(CN)_6$ of 1.01 (±0.02) × 10^{-3} M in this solution, calculated from the optical densities at 475, 500 and 525 mμ and the extinction coefficients of Table I, corresponds to a K_{11} of 36 in 0.50 M $HClO_4$.

A second set of data was obtained for $b_0 > a_0$ where one should consider the complexes $Fe(Fe(CN)_6^{-3})_j^{3\,-\,3j}$. These solutions at 26 ± 2° and at an ionic strength of 1.00 contained $Fe(ClO_4)_3$, $K_3Fe(CN)_6$, $NaNO_3$ and 0.102 M HNO_3, since potassium perchlorate precipitated from solutions in which sodium perchlorate and perchloric acid were used to maintain the acidity and ionic strength.

Figure 2 shows that (a_0b/D_c) is a linear function of b; the parameters of the straight lines are given in Table II. The apparent values of $\epsilon_{11}K_{11}$ and K_{11} are smaller than those of Table I; an attempt has been made to resolve this discrepancy by considering the formation of $FeNO_3^{++}$, with formation constant N, so that $(FeNO_3^{++}) = Na(NO_3^-)$. Because the ionic strength was 1.0, $(NO_3^-) + 6b_0 = 1$. The equation that results is

TABLE II

COMPLEXING OF Fe^{+3} BY EXCESS $Fe(CN)_6^{-3}$ IN NITRATE SOLUTIONS

λ, mμ	(Intercept)$^{-1}$ × 10^{-3}	Slope/ intercept	N	K_{11}
475	12.9	11.8	0.35	17
500	9.4	10.2	.26	15
525	6.25	8.2	.36	12.5

$$(\epsilon_{11}K_{11})(a_0b/D_c) = 1 + N + b\left(K_{11} - 6N - \frac{\epsilon_{12}K_{12}}{\epsilon_{11}K_{11}}\right) + b^2$$

Accepting equation (3) and the values of $\epsilon_{11}K_{11}$ of Table I, the intercepts recorded in Table II have been used to calculate N. From the ratio of slope to intercept one can then calculate K_{11} (neglecting the $\epsilon_{12}K_{12}/\epsilon_{11}K_{11}$ term). The results for N agree as well as can be expected. The average deviation of the K_{11} values is greater than that of Table I, and the average (15) is 25% less than that of Table I. These discrepancies and the trend in the calculated values of K_{11} with wave length may be due to contributions of the $(\epsilon_{12}K_{12}/\epsilon_{11}K_{11})$ term. We feel that all of the results are sufficiently concordant to indicate the essential validity of the interpretations given here.

Experimental.—$Fe(ClO_4)_3 \cdot 10H_2O$ was prepared by evaporation of a mixture of $FeCl_3 \cdot 6H_2O$ with excess 60% $HClO_4$ and recrystallization. It was free of Cl^- and ClO_3^-. Iron(III) was determined as recommended by Swift.[9] Ferro- and ferricyanide solutions were prepared from recrystallized $K_4Fe(CN)_6 \cdot 3H_2O$ and $K_3Fe(CN)_6$ and were determined as recommended by Swift.[10]

---

[8] Using 10^{-3} for the hydrolysis constant, E. Rabinowitch and W. H. Stockmayer, THIS JOURNAL, **64**, 335 (1942).

[9] E. H. Swift, THIS JOURNAL, **51**, 2682 (1929).

[10] E. H. Swift, "A System of Chemical Analysis," Prentice-Hall, Inc., New York, N. Y., 1946, pp. 438–441.

CONTRIBUTION NO. 1476 FROM
THE GATES AND CRELLIN LABORATORIES OF CHEMISTRY
PASADENA, CALIF. RECEIVED SEPTEMBER 30, 1950

Shifted Position of Infrared Absorption Bands Associated with the C=O Linkages in Polycyclic Compounds

BY MARIE LOUISE JOSIEN AND NELSON FUSON

Although a great deal of research has been made on the Raman and infrared band assigned to the C=O stretching vibration in long chain and single ring compounds, spectroscopic study of the C=O linkage in polycyclic compounds is still relatively unexplored. Among the studies which have been made in this latter field may be cited those of Dulou,[1] Kohlrausch,[2] Biquard,[3] Lecomte,[4] Flett,[5] R. N. Jones,[6] and Josien and Fuson.[7] In the course of an infrared spectroscopic study of some polycyclic hydrocarbons and their oxidation products the following values for the carbonyl group absorption bands have been obtained which we believe have not yet been reported in the literature:

Compound[8]	Wave length (in microns) of band assigned to the C=O stretching vibration
Pyrenequinone-3,8	6.10
Pyrenequinone-3,10	6.10
Acetylpyrene	6.02
Chrysenequinone-1,2	6.03
Acenaphthenequinone	5.63 and 5.79
9-Fluorenone-1-carboxylic acid	5.73 and 6.00

These spectral wave length values were obtained for crystalline samples prepared in paraffin oil paste form. A comparison of these results with other results given in the literature cited suggests the following comments: (a) We have assigned the two bands, 5.73 μ and 6.00 μ, of 9-fluorenone-1-carboxylic acid to the carboxylic acid C=O and the cyclic ketone C=O, respectively. The first assignment is in agreement with the usual band location for carboxylic acids[9]; the second is without doubt located at such a position because of the effect of conjugation. (b) From the appearance of two bands, 5.63 μ and 5.79 μ, in acenaphthene-

---

[1] Dulou, Thèse, 1933, Bordeaux, France.

[2] Kohlrausch, *Z. Elektrochem.*, **43**, 282 (1937).

[3] Biquard, *Bull. soc. Chim. France*, **7**, 895 (1940); **8**, 55, 725 (1941).

[4] J. Lecomte, *J. Phys.*, **6**, 259 (1945).

[5] Flett, *J. Chem. Soc.*, 1441 (1948).

[6] R. N. Jones, *et al.*, THIS JOURNAL, **70**, 2024 (1948); **71**, 241 (1949); **72**, 86 and 956 (1950).

[7] M. L. Josien and N. Fuson, *Compt. rend.*, in press.

[8] These compounds were prepared by Dr. St. Elmo Brady, Professor of Chemistry, Fisk University, Nashville, Tennessee.

[9] H. M. Randall, R. G. Fowler, N. Fuson and R. Dangl, "Infrared Determination of Organic Groups," D. Van Nostrand Co., New York, N. Y., 1949, p. 20.