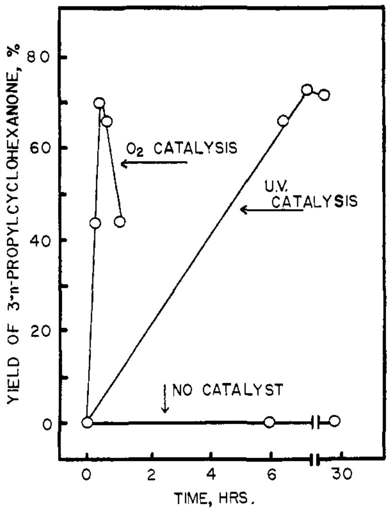

Figure 1. Reaction of tri-*n*-propylaluminum (0.25 *M*) with 2-cyclohexenone (0.25 *M*) in heptane at −78°. The effect of free-radical initiators.

Table I. Photochemically Induced Reaction of Tri-*n*-propylaluminum with Representative α,β-Unsaturated Ketones^a

Ketone	Product^b	Bp, °C^c	Yield, %^{d,e}	Time^f
2-Cyclohexenone	3-*n*-Propylcyclohexanone	211	75 (70)^g	7 hr
3-Penten-2-one	4-Methyl-2-heptanone	156	60	1 hr
Methyl vinyl ketone	2-Heptanone	152	30^h	1 min

^{a} Reaction conditions: 5 mmol of organoaluminum added to 5 mmol of ketone dissolved in heptane and maintained at −78°. Ultraviolet light shone on the mixture for an indicated period of time. ^{b} All products were either compared with authentic samples or exhibited analytical and spectral data in accordance with the assigned structures. ^{c} 745 mm. ^{d} Yields calculated using starting ketone as limiting reagent. ^{e} By glpc analysis. ^{f} Time of reaction at which maximum yield was obtained, in all cases starting materials still present. ^{g} The value in parentheses refers to an experiment in which oxygen was utilized as the radical initiator. ^{h} Polymerization occurred.

hexanone (after hydrolysis) is obtained in 7 hr under otherwise identical conditions.[15]

Essentially identical yields are obtained when traces of oxygen[16] are employed as the free-radical initiator except that the maximum yield is attained more quickly. The results of the photochemical and oxygen-induced reactions are shown graphically in Figure 1.

Galvinoxyl, an efficient free-radical scavenger, has been used successfully to inhibit extremely fast free-radical chain reactions of organometallic compounds.[1,17] We have found that the addition of 5 mol % of galvinoxyl effectively stops the otherwise facile photochemically induced reaction of tri-*n*-propylaluminum with 2-cyclohexenone. The results are shown graphically in Figure 2.

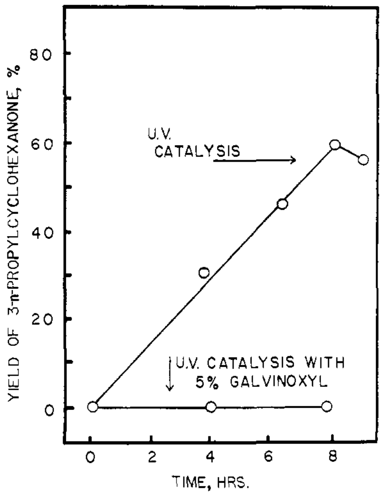

Figure 2. Reaction of tri-*n*-propylaluminum (0.25 *M*) with 2-cyclohexenone (0.25 *M*) in ethyl ether. The effect of added galvinoxyl (in mole %).

The reaction appears to be a general one. Thus, 1,4-addition products are obtained in the reactions of tri-*n*-propylaluminum with a variety of α,β-unsaturated ketones. In all cases, no 1,2-addition products were observed. Furthermore, we have made no effort to maximize yields as of this date. Our results are summarized in Table I. The experimental details are essentially those described earlier.[15]

The discovery that tri-*n*-propylaluminum can be made to participate in such a facile free-radical addition process opens up a major new area for exploration. Such a study is in progress.

Acknowledgment. The authors wish to thank the Research Corporation for support of this work.

George W. Kabalka,* Richard F. Daley

Department of Chemistry, The University of Tennessee
Knoxville, Tennessee 37916

Received February 2, 1973

---

[15] H. C. Brown and G. W. Kabalka, *J. Amer. Chem. Soc.*, **92**, 712 (1970).

[16] Traces of oxygen are most conveniently added to the reaction mixture by simply adding air at a slow rate (~0.1 ml/min) [H. C. Brown and G. W. Kabalka, *J. Amer. Chem. Soc.*, **92**, 714 (1970)].

[17] P. D. Bartlett and T. Funahashi, *J. Amer. Chem. Soc.*, **84**, 2596 (1962).

Carbon-13 Nuclear Magnetic Resonance and Infrared Spectroscopic Studies of ^{13}CO Binding to Rabbit Hemoglobin[1]

Sir:

Recently it has been reported[2] that two ^{13}C resonances of equal intensity, chemical shift separation 2 ppm, can be distinguished for ^{13}CO bound to Fe(II) of the heme units of the α and β chains of rabbit hemoglobin. The chemical shift separation reported for the two ^{13}CO resonances of rabbit hemoglobin is quite large considering that (1) although two ^{13}CO resonances can be distinguished in the spectra of carbonyl hemoglobins of other species (*e.g.*, mouse, dog, and human), not only are there smaller chemical shifts (~0.4 ppm) between the α and β chains but also between the ^{13}CO resonances of carbonyl hemoglobins from different species,[2,3] and (2) the ^{13}CO resonance of

---

[1] This work was performed under the auspices of the U. S. Atomic Energy Commission and the U. S. Public Health Service (Grant No. HL-15980).

[2] R. B. Moon and J. H. Richards, *J. Amer. Chem. Soc.*, **94**, 5093 (1972).

[3] N. A. Matwiyoff, P. J. Vergamini, T. S. Needham, R. T. Eakin, C. Barlow, C. T. Gregg, and W. C. Caughey, manuscript in preparation.

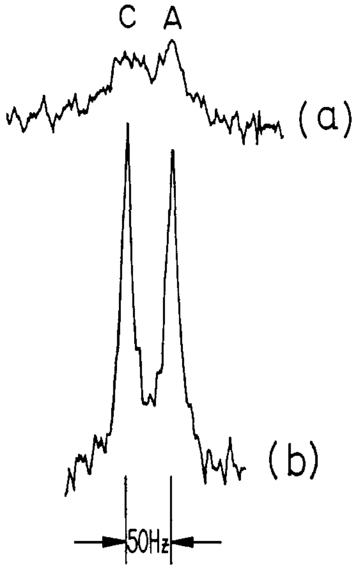

Figure 1. Carbon-13 nmr spectra in the carbonyl region of rabbit blood (Harvey I) carbonylated with CO (80% ^{13}C), 65,000 pulses: (a) whole blood; (b) supernate from red blood cell hemolysate. Samples were treated with CO for 60 min.

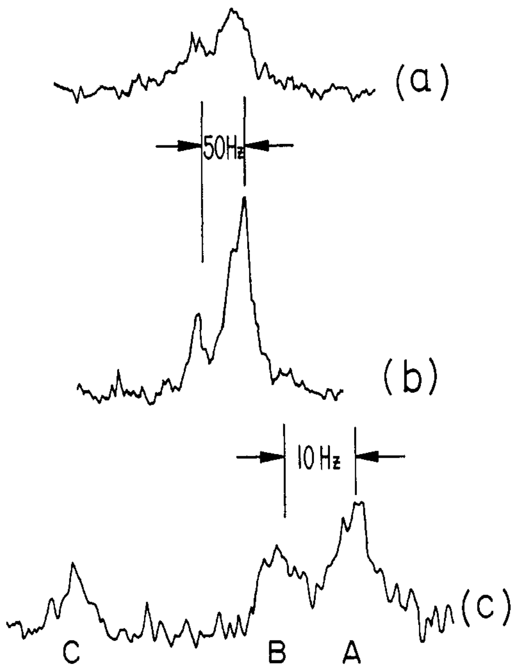

Figure 2. Carbon-13 nmr spectra in the carbonyl region of rabbit blood (Harvey II) carbonylated with CO (80% ^{13}C), 65,000 pulses: (a) whole blood; (b) supernate from red blood cell hemolysate; and (c) spectrum b at higher resolution. Samples were treated with CO for 60 min.

the CO adduct of the *N*-methylimidazole complex of protoheme dimethyl ester differs in chemical shift by only 1 ppm from the center of gravity of the ^{13}CO resonances of mouse, dog, and human hemoglobins.

We report here results of ^{13}C nmr studies which show that *three* distinct Fe(II)–^{13}CO coordination sites can exist in rabbit blood. The sites are thus not those of the α and β subunits of a single type of hemoglobin molecule. Rather, the likely explanation is hemoglobin heterogeneity which varies among individual rabbits and involves a functionally different hemoglobin subunit whose abnormality is reflected in (1) a large downfield shift of the ^{13}C resonance and a low CO stretching frequency for CO bound to the heme, (2) preferential oxidation of the Fe(II) of that subunit by limiting amounts of oxidizing agents, (3) facile displacement of CO in the subunit by O_2, and (4) susceptibility of the subunit to precipitation in citrate buffers at pH <5.

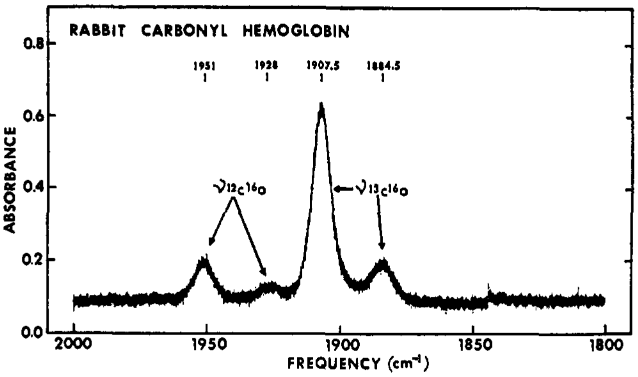

Figure 3. Infrared difference spectrum of rabbit (Harvey I) carbonyl homoglobin. Oxyhemoglobin was treated with CO (80% ^{13}C) for 120 min.

The carbonyl region of the ^{13}C nmr spectrum of a whole blood from a Dutch lop rabbit (Harvey I) carbonylated with CO (80% ^{13}C) is shown in Figure 1a. The set of broad low field signals (ratio *ca.* 1:1) shown is absent from the spectrum of the same blood treated with $^{12}C^{16}O$ and is therefore assigned to $^{13}C^{16}O$ bound to heme Fe(II). The chemical shifts, −77.5 and −79.5 ppm with respect to external benzene, are in agreement with those reported by Moon and Richards[2] and are similar to the ^{13}C nmr chemical shifts of the $^{13}C^{16}O$ complexes of protein-free protoporphyrin IX dimethyl ester–iron(II).[3] After hemolysis, the rabbit blood also exhibited two ^{13}C nmr bands in the ^{13}CO region (Figure 1b). The ^{13}C nmr spectrum of the hemolysate containing the hemeprotein in a homogeneous solution shows the ^{13}C nmr resonances to be substantially narrower than those for whole blood containing intracellular heme protein. The narrowness presumably reflects an enhanced mobility at the CO binding site of the heme protein in a homogeneous solution.[4,5] However, the chemical shifts are the same within experimental uncertainty, ±0.2 ppm, for the two systems.

The spectra of carbonylated blood from another rabbit (Harvey II) exhibited *three* resonances in the carbonyl region of the spectrum (Figure 2) having approximate relative intensities C(1):B(2):A(3). The chemical shifts of resonances A and B are nearly identical with those for ^{13}CO bound to the α and β chains of human, dog, and mouse hemoglobin.[2,3] The infrared spectrum, Figure 3, of this rabbit hemoglobin exposed to a mixture of $^{12}C^{16}O$ and $^{13}C^{16}O$ in about a 1:4 ratio reveals two ν_{CO} bands in about 5:1 ratio of intensities for each CO: 1951 and 1928 cm^{-1} for $^{12}C^{16}O$ and 1907.5 and 1884.5 cm^{-1} for $^{13}C^{16}O$. Band widths ($\Delta\nu_{1/2}$) were 8 and 10 cm^{-1} for the major and minor bands, respectively. Frequency, isotopic shift, and

---

(4) N. A. Matwiyoff and T. E. Needham, *Biochem. Biophys. Res. Commun.*, **49**, 1158 (1972).

(5) R. T. Eakin, L. O. Morgan, C. T. Gregg, and N. A. Matwiyoff, *FEBS* (*Fed. Eur. Biochem. Soc.*) *Lett.*, **28**, 259 (1972).

band widths were similar to those found for normal human HbA.[6] In samples of $^{12}C^{16}O$ with ^{13}C present in natural abundance, only the 1951- and 1928-cm^{-1} bands were observed. The infrared spectrum of the carbonylated blood of Harvey I exhibited two ν_{CO} bands in a 1 : 1 intensity ratio at 1951 and 1928 cm^{-1} for $^{12}C^{16}O$. The infrared band at 1928 cm^{-1} thus corresponds to ^{13}CO resonance C whereas the band at 1951 cm^{-1} corresponds to resonances A and B. That all CO binding sites were saturated under the conditions of Figures 1–3 was confirmed by visible spectroscopy and by the fact that the relative intensities of the bands were not affected by longer exposure to CO, even in the presence of dithionite.

The infrared[7] and nmr parameters[2,3] for resonance C suggest an unusual chemical environment for CO in that rabbit hemoglobin component, and this is reflected in some chemical reactions we have monitored with ^{13}C nmr spectroscopy. Addition of citrate buffer (0.5 *M*, pH 4) to the rabbit hemolysate results in precipation of hemoglobin and a pronounced reduction in the intensity of resonance C relative to those of resonances A and B. The selective precipation of component C is complete at a hemolysate pH 4.8. Treatment of the hemolysate stepwise with fractional equivalents of oxidants like $K_3Fe(CN)_6$ results first in the exclusive oxidation of the heme unit corresponding to resonance C. When heme unit C has been consumed, oxidation of heme units A and B occur. Finally, partial carbonylation of rabbit oxyhemoglobin results first in the appearance of ^{13}CO resonances A and B; similarly, partial oxygenation of carbonyl hemoglobin results in the preferential displacement of ^{13}CO from heme unit C.

An analysis of the specific structural changes (*e.g.*, amino acid substitutions) and the attendant genetic aspects that cause these differences in rabbit hemoglobin properties must await examination of a larger population of rabbits. We could point out here, however, that the amino acid substitutions in these hemoglobins are electrophoretically silent in that the electrophoretic patterns of the hemoglobins from Harvey I and II are indistinguishable.[8] In addition the ^{13}C nmr spectra of carbonylhemoglobin from Harvey I and II were independent of rabbit age (6–11 months) and the frequency of bleeding.

Pulse Fourier transform ^{13}C nmr spectra were obtained at 25.2 MHz using a Varian XL-100-15 spectrometer interfaced to a Supernova computer. The Fourier transform was accomplished using 8K data points in the time domain. Samples were contained in 12-mm (o.d.) tubes and the magnetic field was stabilized using the 15.4-MHz deuterium lock signal of external D_2O. The infrared techniques were similar to those reported.[6,9] Both infrared (Perkin-Elmer Model 180 spectrometer) and visible spectra (Cary 14 spectrometer) were recorded in the same cells of 0.027-mm path length with CaF_2 windows. In infrared measurements a resolution of 3 cm^{-1} was selected to give a high signal to noise ratio while maintaining nearly true absorption intensities[10] at a scan speed of 1 cm^{-1} per 200 sec.

---

(6) J. O. Alben and W. S. Caughey, *Biochemistry*, 7, 175 (1968).

(7) W. S. Caughey, J. O. Alben, S. McCoy, S. Charache, P. Hattsway, and S. Boyer, *Biochemistry*, 8, 59 (1969); W. S. Caughey, M. C. O'Toole, and J. A. Volpe, *Fed. Proc., Fed. Amer. Soc. Exp. Biol.*, 31, 483 (1972).

(8) T. Bradley, R. C. Wohl, C. T. Gregg, and N. A. Matwiyoff, work in progress.

(9) S. McCoy and W. S. Caughey, *Biochemistry*, 9, 2387 (1970).

(10) D. Z. Robinson, *Anal. Chem.*, 23, 273 (1951).

N. A. Matwiyoff,* P. J. Vergamini, T. E. Needham, C. T. Gregg
Los Alamos Scientific Laboratory, University of California
Los Alamos, New Mexico 87544

J. A. Volpe, W. S. Caughey
Department of Biochemistry, Colorado State University
Fort Collins, Colorado 80521

Received April 27, 1973

Structure of a Compound with a Molybdenum-to-Molybdenum Bond of Order Three and One-Half

Sir:

It is now well established that the element molybdenum in its lower valence states shows an extraordinary tendency to form multiple bonds. Bonds of order 4 are very common; they occur in the acetate, $Mo_2(O_2CCH_3)_4$,[1] and a number of other carboxylates,[2,3] in the trifluoroacetate[4] and the pyridine adduct of the trifluoroacetate,[5] in the $Mo_2Cl_8^{4-}$ ion,[6–8] in the $Mo_2(SO_4)_4^{4-}$ ion,[9,10] in $Mo_2(S_2COC_2H_5)_4$,[11] and presumably in various other compounds such as $Mo_2^{4+}(aq)$,[10] $[Mo_2en_4]Cl_4$,[10] $[Mo_2(CH_3CO_2C_2H_5)_4](CF_3SO_3)_4$,[12] and the $Mo_2Cl_4(PR_3)_4$ complexes.[13] There is also the quadruply bonded organometallic compound, $Mo_2(C_3H_5)_4$.[14] The existence of a triple bond in $Mo_2[CH_2Si(CH_3)_3]_6$ has been demonstrated.[15]

We wish to report the first compound containing an Mo-to-Mo bond of order 3.5. According to the molecular orbital description of a quadruple bond,[16–18] the order of the orbitals is σ, π, δ, ($\sigma_n^{(1)}$, $\sigma_n^{(2)}$, δ^*) π^*, σ^*, with eight electrons occupying the lower three orbitals, *viz.*, $\sigma^2\pi^4\delta^2$. Clearly then a species with a bond order of 3.5 should have an unpaired electron in the δ orbital and, in principle, there is an opportunity to gain detailed information about the δ bond through epr spectroscopy. Moreover, the role of the δ com-

---

(1) D. Lawton and R. Mason, *J. Amer. Chem. Soc.*, 87, 921 (1965); T. A. Stephenson, E. Bannister, and G. Wilkinson, *J. Chem. Soc.*, 2538 (1964).

(2) L. Dubicki and R. L. Martin, *Aust. J. Chem.*, 22, 1571 (1969).

(3) A. B. Brignole and F. A. Cotton, *Inorg. Syn.*, 13, 81 (1972).

(4) F. A. Cotton and J. G. Norman, Jr., *J. Coord. Chem.*, 1, 161 (1972).

(5) F. A. Cotton and J. G. Norman, Jr., *J. Amer. Chem. Soc.*, 94, 5697 (1972).

(6) J. V. Brencic and F. A. Cotton, *Inorg. Chem.*, 8, 7 (1969).

(7) J. V. Brencic and F. A. Cotton, *Inorg. Chem.*, 8, 2698 (1969).

(8) J. V. Brencic and F. A. Cotton, *Inorg. Chem.*, 9, 346 (1970).

(9) C. L. Angell, F. A. Cotton, B. A. Frenz, and T. R. Webb, *J. Chem. Soc., Chem. Commun.*, in press.

(10) A. R. Bowen and H. Taube, *J. Amer. Chem. Soc.*, 93, 3287 (1971).

(11) L. Ricard, P. Karagiannidis, and R. Weiss, *Inorg. Chem.*, in press.

(12) T. Backstrom, F. Schoenewolf, Jr., and E. H. Abbott, *J. Coord. Chem.*, in press; see also Abstracts of Papers, 165th National Meeting of the American Chemical Society, Dallas, Texas, April 8–13, 1973.

(13) J. San Filippo, Jr., *Inorg. Chem.*, 11, 3140 (1972).

(14) F. A. Cotton and J. R. Pipal, *J. Amer. Chem. Soc.*, 93, 5441 (1971).

(15) F. Huq, W. Mowat, A. Shortland, A. C. Skapski, and G. Wilkinson, *Chem. Commun.*, 1079 (1971); W. Mowat, A. Shortland, G. Yagupsky, N. J. Hill, M. Yagupsky, and G. Wilkinson, *J. Chem. Soc., Dalton Trans.*, 533 (1972).

(16) F. A. Cotton, *Inorg. Chem.*, 4, 334 (1965).

(17) F. A. Cotton and C. B. Harris, *Inorg. Chem.*, 6, 924 (1967).

(18) M. J. Bennett, K. G. Caulton, and F. A. Cotton, *Inorg. Chem.*, 8, 1 (1969).