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SCALAR COUPLING BETWEEN ^{31}P AND SPIN-1/2 METAL NUCLIDES IN ^{31}P NMR OF COMPLEXES WITH A LIGAND CONTAINING PHOSPHONATE DONOR GROUPS*

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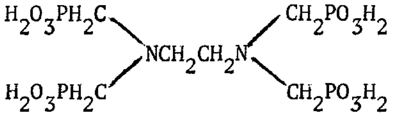
SUMMARY: Scalar couplings between ^{31}P and spin-1/2 isotopes of cadmium, mercury, lead, and tin are reported for the respective metal complexes with the chelating agent (ethylenedinitrilo) - tetramethylenephosphonic acid.

Many biochemically interesting compounds which contain phosphate groups, such as the nucleoside di- and triphosphates, exhibit affinities for divalent cations with formation constants of $>>10^3$ M^{-1}. However, the rate of dissociation of the complexes appears to be sufficiently rapid to bring about "fast exchange" between ligands and metal ions with a consequent loss of any scalar spin-spin coupling between magnetically active metal nuclides and ^{31}P in the ^{31}P NMR spectra. Since the rate of dissociation of metal ion-"phosphate" complexes may be retarded upon association of the complex with an enzyme, there is interest in the magnitude of the scalar coupling between spin-1/2 metal nuclides and ^{31}P in complexes with M-O-P coordinate bonds.

The multidentate chelating agent ENTMP[1]



forms highly stable complexes with multivalent metal ions(1). With ratios

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[1] Abbreviation used: ENTMP(ethylenedinitrilo)-tetramethylenephosphonic acid. The name of this compound in Chemical Abstracts is 1,2-ethanediylbis [nitrilobis(methylene)] tetrakis (phosphonic acid).

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of ENTMP to metal ion near unity, the rates of symmetrical ligand exchange reactions as well as rates of dissociation of the complexes are slow with respect to the magnitude of the scalar coupling between ^{31}P and some of the common spin-1/2 metal nuclides. This communication reports the scalar coupling observed in the ^{31}P NMR spectra for complexes of ENTMP with Cd(II), Pb(II), Hg(II) and Sn(II).

Materials and Methods

ENTMP was synthesized by the method of Moedritzer and Irani(2). Samples for NMR studies were prepared from the octacid of ENTMP and the following metal salts: $Pb(NO_3)_2$, $Cd(NO_3)_2$, HgO, and $SnSO_4$. The pH of the samples was adjusted with concentrated KOH, and the solutions were diluted 1:1 with ^2H_2O to provide an internal ^2H lock signal. Chemical shifts were measured from 85% phosphoric acid contained in a concentric capillary. ^{31}P NMR spectra were obtained at 24.39 MHz with a Varian NV-14 spectrometer modified for pulsed Fourier transform spectroscopy. Spectra for a sample of 96% enriched ^{113}Cd ENTMP were obtained at 40.48 MHz with a Jeol PFT-100 spectrometer. Spectra were obtained with and without broad-band proton decoupling.

Results and Discussion

The ^{31}P NMR spectrum of the free ligand consists of a 1:2:1 triplet which collapses to a singlet upon irradiation of the methylene protons. The ENTMP complexes with the metal ions gave ^{31}P spectra with a central signal for the non-magnetic isotopes flanked symmetrically by satellite signals from the complexes with the less abundant spin-1/2 isotopes of the metal ions[2]. Chemical shifts and ^{31}P-metal nuclide coupling constants are summarized in Table I. The single set of ^{31}P signals observed for each of the complexes indicates either a rapid intramolecular exchange of phosphonate donor groups between positions in-plane and out-of-plane with respect to the nitrogen donor groups or a negligible magnetic non-equivalence for ^{31}P in these two positions(3).

The ^{113}Cd enriched sample was also investigated at 40.480017 MHz in an experiment in which a simultaneous irradiation of protons (broad-band) and

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[2] The positions of the satellite signals were independent of the frequency of sample spinning.

Table I
Chemical Shifts and Metal-Phosphorus Coupling Constants Obtained from ^{31}P NMR Spectra for ENTMP Complexes

Complex[a]	Isotope	δ (ppm)[b]	J_{P-M} (Hz)	pH
SnENTMP[c]	$^{117}Sn, ^{119}Sn$	16.9 ± .1	31.4 ± .5	10.2
CdENTMP[c]	$^{111}Cd, ^{113}Cd$	15.4 ± .1	45.9 ± .7	11.2
PbENTMP	^{207}Pb	20.3 ± .1	54.6 ± .5	11.2
HgENTMP	^{199}Hg	15.4 ± .1	127 ± 2	11.2

[a] The concentration of complex was 0.1M in each case. Spectra were recorded at ambient probe temperature (25-30°C).

[b] Chemical shifts relative to external sample of 85% H_3PO_4. (A positive shift is downfield.)

[c] Only a single set of satellite resonances is observed for two isotopes with very similar magnetic moments.

the ^{113}Cd (narrow-band) was effected. The ^{31}P doublet signal at 15.4 ppm was collapsed to a singlet when the ^{113}Cd was irradiated at a frequency of 22.181140 MHz. Based on the data of Maciel and Borzo(5), this frequency corresponds to an upfield shift for ^{113}Cd ENTMP of 113 ppm with respect to a 0.1M aqueous solution of $Cd(ClO_4)_2$.

The ^{31}P-metal nuclide coupling observed for the ENTMP complexes may be mediated through the M-N-C-P or M-O-P linkages. Since the latter route involves only two bonds, this linkage likely accounts for most of the observed scalar coupling. However, this conjecture cannot be proven at present. Subject to the above ambiguity and to intrinsic differences between phosphates and phosphonates, the experiments with complexes of ENTMP indicate that the magnitudes of M-O-P coupling constants are large enough to permit NMR observation of the coupling in complexes with M-O-P coordination whose lifetimes are in the msec region (i.e. $\tau > 1/2\pi J$).

A potential example of a long-lived metal-phosphate complex on an enzyme is given by E. coli alkaline phosphatase, where direct metal-phosphate coordination has been inferred from ^{31}P NMR experiments with the cobalt(II) derivative(4). Experiments with the ^{113}Cd-enriched cadmium phosphoenzyme have already been performed(6,7), and while they have ruled out a coupling as large as the 46 Hz seen with ENTMP, there is an indication that a coupling of approximately 10 Hz may be present(6).

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