the hydrogen bonding between the carboxylate group of histidinate (His) and the polar side group of asparaginate (Asn), serinate (Ser), threoninate (Thr), *etc.* (referred to as AA) in the ternary systems M(AA)(His) was confirmed by evidence from synthetic and CD and NMR spectral studies [3]. The infrared spectra of the isolated complexes indicated the existence of geometric isomerism in the diastereomers of M(A)(B) and M(AA)(His). On the basis of the steric requirements for the intramolecular ligand–ligand interactions, the geometry of M(L-A)(L-B) and M(D-AA)(L-His) has been inferred to be *trans* and that of M(D-A)(L-B) and M(L-AA)(L-His) *cis* with respect to the amine nitrogens. This is further supported by our recent infrared spectral study of the crystalline ^{63}Cu- and ^{65}Cu(AA)(L-His) complexes (AA = L-Asn, D-Asn, L-Thr, L-Ser, or D-Ser), which shows that only Cu(L-AA)(L-His) exhibits a metal isotope shift (*ca.* 1 cm^{-1}) of the Cu–NH_2 symmetric stretching band. The finding is reasonably explained by the *cis* structure for Cu(L-AA)(L-His) as established for AA = Asn andThr by X-ray analysis and the *trans* structure for Cu(D-AA)(L-His) where the isotope shift is not observed because the symmetric stretching mode does not involve the displacement of Cu.

Possibilities of ligand–ligand interactions in mixed ligand heteronuclear complexes will also be discussed.

1 T. Sakurai, O. Yamauchi and A. Nakahara, *Bull. Chem. Soc. Jpn., 49,* 169 (1976).
2 O. Yamauchi and A. Odani, *J. Am. Chem. Soc., 103,* 391 (1981).
3 O. Yamauchi, T. Sakurai and A. Nakahara, *J. Am. Chem. Soc., 101,* 4164 (1979);
A. Odani and O. Yamauchi, *Bull. Chem. Soc. Jpn., 54,* 3773 (1981).

B18

Chemistry and Molecular Biology of Anticancer Platinum Drugs

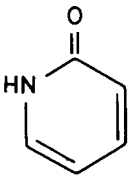
STEPHEN J. LIPPARD

Department of Chemistry, Massachusetts Institute of Technology, Cambridge, Mass. 02139, U.S.A.

The anticancer platinum drug *cis*-diamminedichloroplatinum(II) (*cis*-DDP) binds avidly to DNA both *in vitro* and *in vivo*. The mechanism by which *cis*-DDP kills tumor cells is thought to involve DNA–Pt interactions [1], but further experimental work to establish this point would be valuable. Accordingly, in joint studies with M. Poirier and her group at the National Cancer Institute, we have developed an antibody to *cis*-DDP-DNA adducts formed *in vitro* which reacts specifically with *cis*-DDP–DNA adducts extracted from leukemia cells of mice treated with the platinum drug [2]. Though these antibody studies and other work using nucleases, especially restriction enzymes and exonuclease III, to probe the nature of the *cis*-DDP–DNA binding stereochemistry we find substantial evidence to support the hypothesis that the *cis*-$(NH_3)_2Pt^{2+}$ unit forms an intrastrand crosslink between two adjacent or nearby guanine bases on DNA [3]. The stereochemistry of this adduct has been deduced by NMR studies of a model complex formed between the self-complementary hexanucleoside pentaphosphate d(ApGpGpCpCpT) and *cis*-DDP. In this complex the two adjacent guanine bases are coordinated to the *cis*-diammineplatinum(II) moiety through their N-7 nitrogen atoms [4].

When *cis*-DPP is administered in chemotherapy it is invariably used in combination with other anticancer drugs. How do these drugs affect the chemistry of *cis*-DDP within the cell? Moreover, agents within the cell such as glutathione are known to form complexes with *cis*-DDP. Will these intracellular reactions affect the DNA-*cis*-DDP interaction? With these questions in mind we have studied the binding of *cis*-DDP to DNA plasmids and restriction fragments. Our results demonstrate that the minor groove intercalating drug ethidium bromide switches the nuclease sensitive binding sites of *cis*-DDP [5] and that glutathione also alters the DNA binding properties of the antitumor drug.

Finally, an investigation has been conducted using ^{195}Pt NMR spectroscopy which demonstrates the kinetic mobility of diammineplatinum(II) adducts of the nucleobase analog α-pyridone [6]. In particular, the head-to-head isomer of $[(en)Pt(C_5H_4NO)]_2^{2+}$



α-pyridone

rearranges thermally to the head-to-tail isomer by a process that is reversible, intramolecular, and dissociatively activated. The possible relevance of this isomerization reaction to *cis*-DPP–DNA adducts will be discussed.

Acknowledgements. This work was supported by PHS grant number CA 34992 awarded by the National Cancer Institute, DHHS. We thank Englehard Industries for a generous loan of K_2PtCl_4 from which all platinum compounds were prepared.

1 J. J. Roberts and A. J. Thomson, *Progr. Nucleic Acid Res. Molec. Biol., 22,* 71 (1979).
2 M. C. Poirier, S. J. Lippard, L. A. Zwelling, H. M. Ushay, D. Kerrigan, C. C. Thill, R. M. Santella, D. Grunberger and S. H. Yuspa, *Proc. Nat. Acad. Sci. U.S.A., 79,* 6433 (1982).
3 S. J. Lippard, *Science, 218,* 1075 (1982).
4 J. P. Caradonna, S. J. Lippard, M. J. Gait and M. Singh, *J. Am. Chem. Soc., 104,* 5793 (1982).
5 T. D. Tullius and S. J. Lippard, *Proc. Natl. Acad. Sci. U.S.A., 79,* 3489 (1982).
6 T. V. O'Halloran and S. J. Lippard, submitted for publication.

B19

A Novel System Involving Exchange-Coupling between a Heme and an Iron–Sulfur Cluster

ECKARD MÜNCK* and JODIE A. CHRISTNER

Gray Freshwater Biological Institute, University of Minnesota, Navarre, Minn. 55392, U.S.A.

PETER A. JANICK and LEWIS M. SIEGEL

Department of Biochemistry, Duke University Medical Center, Durham, N.C. 27710, U.S.A.

The β-subunits of *E. coli* sulfite reductase (SiR) contain an Fe-isobacteriochlorin, termed siroheme, and a [4Fe-4S] cluster. Mössbauer and EPR studies [1] of oxidized SiR have demonstrated that the siroheme and the iron–sulfur cluster are exchange-coupled. Such a coupling implies a covalent link between the two chromophores; it is reasonable to assume that the cluster is attached to the heme iron by an as yet unspecified bridging ligand. In oxidized SiR the iron-sulfur cluster is in the 2+ oxidation state, a state in which [4Fe-4S] clusters are normally diamagnetic. Through exchange interactions with the siroheme the cluster acquires paramagnetism; the experimental observations have been explained qualitatively in a model which involves isotropic exchange between the heme iron and one iron site of the cluster [2]. Recent studies [3] of one-electron and two-electron reduced SiR, a nitrite 'turnover' (ferroheme-NO) complex, and studies of SiR in chaotropic agents show that the coupling is maintained in many oxidation-, spin-, and complexation-states of the enzyme. We have also studied SiR complexed to cyanide (in three oxidation states) and carbon monoxide. Exchange-coupling is indicated in the oxidized cyano complex; in the reduced CN^- and CO complexes the heme is low-spin ferrous and thus in a state unfavorable for the development of interatomic exchange.

1 J. A. Christner, E. Münck, P. A. Janick and L. M. Siegel, *J. Biol. Chem., 256,* 2098 (1981).
2 E. Münck, in 'Iron–Sulfur Proteins', T. G. Spiro, ed., Wiley–Interscience, New York, 1982, p. 147.
3 J. A. Christner, E. Münck, P. A. Janick and L. M. Siegel, submitted.

B20

Multifunctional Behavior as an Aid in Deducing Metalloenzyme and Model Reaction Mechanisms

DANIEL L. LEUSSING

Chemistry Department, The Ohio State University, Columbus, Ohio 43210, U.S.A.

The ability of a substance to participate in competing reactions presents difficulties but if the problems can be resolved a clearer picture of the behavior in all reaction modes may be forthcoming. Oxalacetate is involved in a number of different biological reactions which are catalyzed by metalloenzymes. Many of these reactions are amenable to independent investigation in model systems employing metal ions.

Decarboxylation is one reaction mode of $oxac^{2-}$ which has attracted interest for many years owing to strong resemblances between the rate dependencies of the enzymic and model systems on metal ion concentration. The Steinberger–Westheimer mechanism had early been accepted as the means by which metal ions catalyzed $oxac^{2-}$ decarboxylation:

$$M^{2+} + oxac^{2-} \rightleftharpoons M(oxac) \xrightarrow{-CO_2}$$

$$M(enolatepyruvate) \xrightarrow{H^+} M(pyruvate)$$

Prevailing evidence indicates that the activation barrier is lowered by the complexation of the high energy enolate of pyruvate.

Enolization and hydration reactions of $oxac^{2-}$ procede within the same time frame and are closely entwined. Resolved rate data show that the reactions are subject to acid and base catalysis. Proton catalysis appears to be equally effective for both, but large differences are evident in base catalysis. Hydration rates are sensitive to bases possessing an oxygen donor atom. OH^- is a very efficient catalyst and even H_2O catalyzed rates are appreciable. Tertiary amines are found to be weak catalysts. Enolization appears to be more susceptible to softer bases. The rate constant for the OH^- catalyzed path is 1/6 as large as that determined for hydration, and H_2O catalysis is negligible; however, tertiary amines are potent catalysts and the more basic ones exceed OH^- in activity. Different sites are involved in catalysis. Enolization involves the removal of a $-CH_2-$ proton from