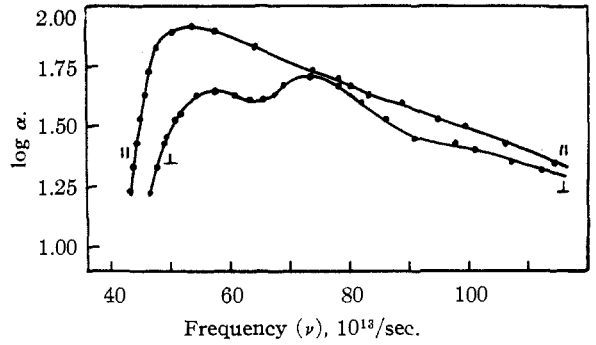

Fig. 1.—Absorption spectra of bis-(dimethyloxime)-copper.

no metal–metal interaction. Similar measurement (Fig. 2) revealed that the corresponding nickel compound exhibits a remarkable dichroism for the corresponding absorption band, maximum absorption being observed with electric vector along the *c*-axis.[9] The following data were

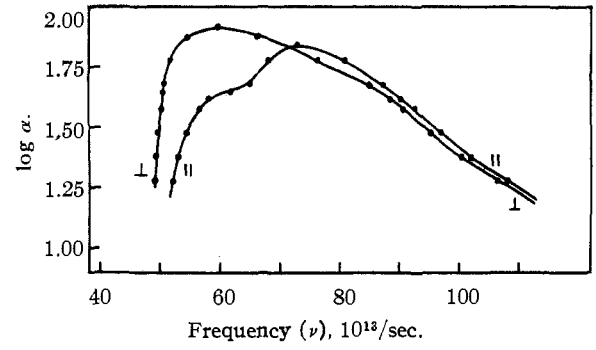

Fig. 2.—Absorption spectra of bis-(dimethylglyoxime)-nickel.

obtained: for ∥ absorption, $\nu = 60 \times 10^{13}$/sec. and log $\alpha = 1.62$; for ⊥ absorption, $\nu = 59.2 \times 10^{13}$/sec. and log $\alpha = 1.91$. These data indicate that the relation on the dichroism with this crystal is reverse to that in the ordinary case. A direct interaction between nickel atoms may be considered as responsible for the reversal of the effect. Similar dichroism measurement was made with chocolate-colored acicular crystals of the corresponding platinum complex. Although the crystal structure has not been determined, judging from the data of analogous compounds, planar complexes are supposed to be arranged parallel to each other to a greater or lesser extent, with their planes nearly perpendicular to the needle-axis. On the basis of the assumed structure, it is established that ⊥ absorption is bathochromic and hyperchromic to ∥ absorption. This relation is reverse to that for planar complexes of an ordinary type.[5b] The metal–metal interaction, as in the crystal of the corresponding nickel compound, may be expected in the platinum compound.

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(9) Godycki, *et al.*, reported (ref. 4) that maximum absorption was observed with the electric vector perpendicular to the *c*-axis.

BIOLOGICAL ACTIVITY OF A METABOLITE OF *p*-AMINOBENZOIC ACID (PABA) IN A HYDROXYLATING SYSTEM

Sir:

Sloane, Crane and Mayer[1] reported that resting cells of *Mycobacterium smegmatis* (101) hydroxylate aniline to *p*-aminophenol. Further investigations[2,3] indicated that the hydroxylation is an energy-coupled reaction. It was determined that chlortetracycline (Aureomycin[4]) and oxytetracycline (Terramycin[5]) compounds which uncouple oxidative phosphorylation[6,7] inhibit the hydroxylation reaction without affecting the oxygen uptake of mycobacterial cells[8] at concentrations ranging from 1.3 to 5.4 $\times 10^{-6}$ *M*.[9] Penicillin, chloramphenicol, dihydrostreptomycin, viomycin and neomycin do not inhibit the hydroxylation reaction by the mycobacteria at these concentrations.[9]

Isochlortetracycline, the biologically inactive rearrangement product of chlortetracycline[10] does not inhibit the hydroxylation, thus the antibiotic activity of chlortetracycline parallels its ability to inhibit the hydroxylation reaction.

It is the purpose of this communication to describe some biological and chemical properties of a metabolite of PABA, which non-competitively reverses the activity of chlortetracycline and oxytetracycline in this system. The data are shown in Table I. The metabolite appears to function as a

TABLE I

THE CHLORTETRACYCLINE–OXYTETRACYCLINE REVERSING ACTIVITY OF PABA-METABOLITE IN THE HYDROXYLATING SYSTEM

	M. Tuberculosis (#607 ≃ 600 mg. dried cells per 25 ml. of buffer–citrate–metals solution per 250-ml. flask (vigorous aeration (1))	μM. *p*-aminophenol, 16 hr.
1	Cells + buffer–citrate–metals + aniline (107 μM.)	4.62
2	1 + 0.136 μM. chlortetracycline	0
3	2 + 9.12 μM. PABA—metabolite[a]	1.54
4	2 + 4.56 μM. PABA—metabolite[a]	1.54
5	2 + 2.28 μM. PABA—metabolite[a]	0.93
6	2 + 1.14 μM. PABA—metabolite[a]	0.49
7	2 + 0.57 μM. PABA—metabolite[a]	0.13
8	4 without aniline	0
9	4 without aniline and chlortetracycline	0
10	Buffer–citrate–metals + 4.56 μM. PABA metabolite + 107 μM. aniline	0

[a] Maximum solubility of metabolite is 4.5 μM. per 25 ml.

cofactor or cosubstrate in the hydroxylation reaction. Amino acids, purines, pyrimidines and vita-

(1) N. H. Sloane, C. Crane and R. L. Mayer, *J. Biol. Chem.*, **193**, 453 (1951).

(2) N. H. Sloane, M. Samuels, C. Ritter, C. Crane and R. L. Mayer, *Federation Proc.*, **11**, 288 (1952).

(3) N. H. Sloane, M. Samuels and R. L. Mayer, *J. Biol. Chem.*, in process of publication.

(4) Aureomycin is the registered trade name of Lederle Laboratories Division, American Cyanamid Co.

(5) Terramycin is the registered trade name of Chas. Pfizer and Co.

(6) W. F. Loomis, *Science*, **111**, 474 (1950).

(7) Y. Miura, Y. Nakamura, H. Matsudaira and T. Komeiji, *Antibiotics and Chemotherapy*, **2**, 152 (1952).

(8) Non-pathogenic mycobacteria perform this hydroxylation.

(9) N. H. Sloane, unpublished data.

(10) C. W. Waller, B. L. Hutchings, C. F. Wolf, A. A. Goldman, R. W. Broschard and J. H. Williams, THIS JOURNAL, **74**, 4981 (1952).

mins did not reverse the activity of chlortetracycline at varying concentrations. The metabolite does not affect the antibiotic activity of these compounds, as determined by the standard antibiotic assay technique.

It is interesting to note that while 2,4-dinitrophenol (5×10^{-4} *M*) also inhibits the reaction[2,3] the action of this uncoupling agent[11] is not reversed by the PABA metabolite. These data suggest that different inhibitors of oxidative phosphorylation do not necessarily effect the same locus in the enzyme complex. Witter, Newcomb and Stotz[12] have previously discussed this concept.

The compound designated PABA-metabolite was obtained as a crystalline free base from culture filtrates of *M. smegmatis* which was grown in the presence of 0.1% PABA. The compound was purified by partition between water and ethyl acetate (*p*H 10.0) after removal of the silver precipitable material from the culture filtrate at *p*H 7. The metabolite (ethyl acetate soluble at *p*H 10) was crystallized from water (*p*H 6.8) and then from ethyl acetate, chloroform–petroleum ether mixtures or hot methanol to constant analyses. The yield of recrystallized metabolite is in the order of 500 μg. per liter. The compound has the empirical formula $C_{14}H_{14}N_2 \cdot 1/2H_2O$[13] and melts[14] at 198–199° uncor., with darkening.

Anal. Calcd. for $C_{14}H_{14}N_2 \cdot 1/2H_2O$: C, 76.69; H, 6.90; N, 12.78; mol. wt., 219. Found:[15] C, 76.96; H, 7.23; N, 12.81; mol. wt., 248 (Signer, 10^{-3} *M* in acetone).

The ultraviolet absorption spectrum of the metabolite (free base) in ethanol shows maxima at 258 mμ (ε 23,400) and at 295 mμ (ε 3900). The ultraviolet absorption spectrum of the hydrochloride in ethanol shows a marked change, the absorption peaks at 258 and 295 mμ are replaced by an inflection at 255 mμ (ε 3600). The metabolite shows one aromatic amine group by Bratton–Marshall test[16] per mole (slow development of the color).

The major infrared bands are at 2.9, 6.14, 6.54, 7.07, 7.57, 7.96, 8.45, 8.91, 9.27, 10.03 (broad), 10.67, 11.45, 12.25 and 13.25 microns. Work is now in progress for the elucidation of the structure of this compound.

(11) W. Loomis and F. Lipmann, *J. Biol. Chem.*, **173**, 827 (1948).

(12) R. F. Witter, E. H. Newcomb and E. Stotz, *ibid.*, **202**, 291 (1953).

(13) Microanalyses were performed by Mr. Louis Brancone and coworkers.

(14) The compound decomposes if heated slowly; a sharp melting point is obtained if compound is placed in preheated bath in vacuum capillary.

(15) Analyses of five preparations agreed.

(16) A. C. Bratton and E. K. Marshall, *J. Biol. Chem.*, **128**, 537 (1939).

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BOOK REVIEWS

Physical Chemistry of Metals. By LAWRENCE S. DARKEN, Ph.D., Research Laboratory, United States Steel Corporation; and ROBERT W. GURRY, Ph.D., Research Laboratory, United States Steel Corporation; with a collection of problems by MICHAEL B. BEVER, Sc.D., Department of Metallurgy, Massachusetts Institute of Technology. McGraw-Hill Book Company, Inc., 330 West 42nd Street, New York 36, N. Y. 1953. ix + 535 pp. 16.5 × 23.5 cm. Price, $8.50.

This book will appeal to both the chemist and the metallurgist although it was written expressly for the latter. As a text for students who have completed the usual one-year introductory course in physical chemistry, it should find its place in many metallurgy curricula. It will be a valuable guide to the research worker in this field of applied physical chemistry.

After a brief introductory chapter the next four, approximately one-fourth of the book, are devoted to gases, solids and liquids. The discussion of atomic structure is excessively brief but contains a complete table of arrangement of orbital electrons. Bonding and resonance phenomena are treated briefly but competently. In addition the chapter on solids discusses plastic deformation, Hume-Rothery's classification, atomic radii and crystallography of the elements and imperfections in crystals. It contains a rather full exposition of Pauling's theory of valence and atomic radius in metals.

The chapter on solid solutions and intermetallic compounds includes quantitative discussions of the effect of size factor and electronegativity on extent of solid solubility. Long and short range order and intermediate phases are discussed.

Chapter 5 contains an excellent summary of the structure of liquids as deduced from X-ray diffraction data. The authors make a strong case for ordering in certain liquid metallic solutions. A discussion of the "hole" theory of liquid structure contains the only mention of viscosity.

Chapters 6–10 present the classical approach to thermodynamics with applications to metallic solutions and other systems of especial metallurgical interest. Statistical mechanics is considered beyond the scope of the book but its conclusions are used freely, especially in connection with the third law. This procedure is likely to prove baffling to the student who has not been told about the relation between entropy and randomness.

The treatment of solutions will be especially helpful to research metallurgists. The authors employ the function $\alpha_1 = \ln \gamma_1/(1 - N_1)^2$ which is useful in interpolating activity data and in graphical integration of the Gibbs–Duhem equation. This chapter and a later one on free energy–composition diagrams are especially recommended to the physicist or metallurgist who is unfamiliar with the elegant methods developed by chemists for the thermodynamic treatment of solutions.

Two chapters on the phase rule and heterogeneous equilibria contain the basic principles applicable to one- and two-component systems. Systems of three or more components are not discussed.

Chapter 14 contains summaries of many useful metallurgical data on the free energy of formation of oxides, sulfides, carbides, nitrides and chlorides. In it also is found a brief summary of the authors' own fine work on the system iron–oxygen. The two following chapters are treatises on the two important systems Fe–N and Fe–C. They include