COMMUNICATIONS TO THE EDITOR

INFRARED ABSORPTION AS A MEASURE OF ENOLIZATION

Sir:

Hilbert, Wulf, Hendricks and Liddell [THIS JOURNAL, **58,** 548 (1936)] discovered the absence of the absorption band at 1.50 μ, which is recognized as being characteristic of the hydroxyl group, in molecules where hydrogen bond formation is possible. Their results have been confirmed in this Laboratory [Buswell, Dietz and Rodebush, *J. Chem. Phys.*, **5,** 84, 501 (1937)], where it was also discovered that the fundamental of the hydroxyl group at 2.75 μ is not missing but is shifted to the neighborhood of 3.0 μ with increased intensity. In one case, however, which was studied by them, dibenzoylmethane, we find no absorption characteristic of the hydroxyl group anywhere in the region 2.75–3.0 μ. We do not believe this to indicate hydrogen bond formation, but rather that the compound is not, as Wulf and his collaborators assumed, largely in the enolic form. A titration with bromine confirms the conclusion that dibenzoylmethane in carbon tetrachloride solution is almost entirely in the ketonic form. The effect of solvent upon enolization has been recognized for a long time but its importance has perhaps not been realized. We have evidence that the percentage of enolization is much greater for example in chloroform than in carbon tetrachloride.

CHEMICAL LABORATORY OF THE UNIVERSITY OF ILLINOIS
URBANA, ILLINOIS

A. M. BUSWELL
W. H. RODEBUSH
M. F. ROY

RECEIVED AUGUST 13, 1937

RELATION OF NICOTINIC ACID AND NICOTINIC ACID AMIDE TO CANINE BLACK TONGUE

Sir:

During the progress of our studies on the isolation of the antipellagra factor two facts have become evident which we feel may have significant application in the treatment of human pellagra.

First, a commercial preparation of nicotinic acid is highly effective in curing canine black tongue. The method of feeding the dogs and testing the concentrates has been described previously [Koehn and Elvehjem, *J. Biol. Chem.*, **118,** 693 (1937)]. Consideration of the solubility relationships and chemical properties of highly active concentrates together with the observation that nicotinic acid had some growth stimulating effect on rats suggested that this compound be tried on dogs. A single dose of 30 mg. of Eastman Kodak Company nicotinic acid gave a phenomenal response in a dog suffering from black tongue. The appetite improved immediately, the animal showed a growth response similar to that obtained on the original liver extract, the diarrhea disappeared, and upon further additions of the nicotinic acid growth continued uninterruptedly. Similar responses have now been obtained in four dogs. One dog on the basal ration has been fed 100 mg. of nicotinic acid per week for three weeks with no indication of recurring symptoms and the dog appears normal in every way. There seems to be no question about the activity of this sample, at least, in the cure of black tongue.

Second, nicotinic acid amide has been isolated from highly active concentrates. Considerable difficulty was encountered in attempts to obtain crystals from liver concentrates (prepared by Eli Lilly and Company). However after distillation of certain concentrates in a molecular still, crystallization of the distillate became very simple.

The distillate proved very active in the cure of black tongue. Upon precipitation with mercuric chloride and subsequent removal of mercury from the precipitate, definite crystals which amounted to about 60% of the total distillate were obtained. These crystals also showed high activity. Recrystallization of the material as the hydrochloride gave a product melting at 227–228° uncorrected. An elemental analysis made by Dr. K. P. Link and Mr. H. Campbell gave the following results: C, 45.8; H, 4.6; N, 17.05. The theoretical results for nicotinic acid amide hydrochloride are C, 45.5; H, 4.4; N, 17.67. The melting point of synthetic nicotinic acid amide hydrochloride was found to be 230° and the mixed melting point was 227–228°. The free base was also prepared from the above hydrochloride and found to melt at 126–127° either alone or when mixed with known nicotinic acid amide.

These and other results demonstrate the close relationship of nicotinic acid and the amide to black tongue. The fact that nicotinic acid amide is an essential ingredient of the diet is not surprising since it is a component of certain coenzymes. However, the observation that a deficiency of this material may be the cause of black tongue is most interesting. Whether these compounds are equally effective in human pellagra can only be answered by clinical trials.

DEPARTMENT OF AGRICULTURAL CHEMISTRY
UNIVERSITY OF WISCONSIN
MADISON, WISCONSIN
C. A. ELVEHJEM
R. J. MADDEN
F. M. STRONG
D. W. WOOLLEY

RECEIVED AUGUST 16, 1937

A WATER-SOLUBLE LIGNIN SULFONIC ACID FROM AN EXTRACTED OAK LIGNIN

Sir:

The authors have succeeded in preparing a water-soluble lignin sulfonic acid from an *extracted* lignin by the following method: solvent-extracted, oak wood meal was acetylated by a modification of the method of Suida and Titsch [*Monatsh.*, **54,** 700 (1929)] and the isolated, acetylated lignin–carbohydrate complex hydrolyzed in cold aqueous acetone solution with sodium hydroxide. Filtration and acidification of the filtrate gave an oak lignin which could be separated into two fractions: (a) a chloroform-soluble (10%), and (b) a chloroform-insoluble fraction (90%). *Anal.* (a) OCH_3, 23.4%; (b) C, 63.9; H, 6.2; OCH_3, 20.6. The fully methylated lignins showed C, 65.2; H, 6.8; OCH_3, 37.6. The oak lignin was readily and completely soluble in sodium bisulfite cooking liquor at 100–125°. Treatment of the lignin sulfonic acid (OCH_3, 23.0) (collaboration of W. L. Hawkins) with alkali yielded a mixture of equal parts of vanillin and syringic aldehyde (total yield 4.6%). Its properties, as well as those of the lignin from which it is derived, are now being investigated thoroughly on account of the importance of such data for use in studies on the mechanism of sulfite pulp manufacture.

DIVISION OF INDUSTRIAL AND CELLULOSE CHEMISTRY
McGILL UNIVERSITY
MONTREAL, CANADA
HAROLD HIBBERT
W. H. STEEVES

RECEIVED AUGUST 9, 1937

THE NITROGEN ISOTOPE (N^{15}) AS A TOOL IN THE STUDY OF THE INTERMEDIARY METABOLISM OF NITROGENOUS COMPOUNDS

Sir:

Urey, Fox, Huffman and Thode [THIS JOURNAL, **59,** 1407 (1937)] have recently succeeded in concentrating the nitrogen isotope of atomic weight 15 (N^{15}). Professor Urey has kindly supplied us with a sample of ammonia enriched in N^{15}. It had a N^{14}/N^{15} ratio of 160 as compared to a normal of 266. All isotope analyses were carried out with a mass spectrometer. Glycine and hippuric acid were synthesized from this ammonia.

For metabolism studies of organic compounds with isotopes it is essential that the labelled atom be stable and not exchange with the same element of other compounds. We have studied the systems glycine–ammonia, hippuric acid–ammonia, hippuric acid–glycine, and tyrosine–glycine, and have found no exchange of the nitrogen.

Two groups of experiments on biological hippuric acid formation were carried out on rats with the new compounds. The urinary hippuric acid, after either feeding or injection of hippuric acid (N^{15}), contained a mixture of about one-third normal acid and two-thirds of the material fed. When glycine and benzoic acid were given together by the same methods, the urinary hippuric acid contained about two-thirds normal hippuric acid and one-third hippuric acid (N^{15}).

The experiments indicate that hippuric acid can be absorbed from the intestinal tract without being hydrolyzed, and furthermore suggest that glycine may be directly utilized for hippuric acid formation. The nitrogen isotope should prove to be as useful in the study of the intermediary metabolism of nitrogenous compounds as deuterium is in the study of fat and sterols.

DEPARTMENT OF BIOLOGICAL CHEMISTRY
COLLEGE OF PHYSICIANS AND SURGEONS
AND DEPARTMENT OF CHEMISTRY
COLUMBIA UNIVERSITY
NEW YORK, N. Y.
RUDOLF SCHOENHEIMER
D. RITTENBERG
M. FOX
ALBERT S. KESTON
SARAH RATNER

RECEIVED AUGUST 2, 1937

CATALYTIC ISOMERIZATION OF *n*-BUTANE AND ISOBUTANE

Sir:

Glasebrook, Phillips and Lovell have recently reported [THIS JOURNAL, **58,** 1944 (1936)] a low temperature, catalytic isomerization of *n*-pentane to isopentane in the presence of aluminum halides.