between five and six hundred copies had been sent out to more than 400 members, 26 state academies of science affiliated with the American Association for the Advancement of Science, 118 other addresses of scientific bodies, libraries, etc., including 28 in thirteen foreign countries. About 20 per cent. of the reserve fund would have to be used in order to balance income and outgo. Respecting the Reelfoot Lake Biological Station he said that the legislature of 1939 had made an appropriation to the academy of $4,000 for the biennium beginning on July 1, 1939, and the State Department of Conservation had added $500 in consideration of researches made there pertaining to the solution of fish and game problems.

Dr. Clinton L. Baker was reelected director of the Reelfoot Lake Biological Station, and Dr. George R. Mayfield, a member of the advisory committee, each for a term of three years. Dr. John T. McGill, in consideration of his services to the academy during the last fourteen years, was elected honorary president for life, with membership on the executive committee without official responsibility.

OFFICERS FOR THE YEAR 1940

President, C. L. Baker, Southwestern, Memphis; *Vice-President,* F. Lynwood Wren, George Peabody College, Nashville; *Secretary-Treasurer,* Kendall E. Born, Division of Geology, Nashville.

Botany Section, *Chairman,* C. R. Freeman, Teachers College, Memphis; *Secretary,* Stanley A. Cain, University of Tennessee, Knoxville.

Geology Section, *Chairman,* Walter F. Pond, Division of Geology, Nashville; *Secretary,* Kendall E. Born, Division of Geology, Nashville.

Physics Section, *Co-Chairmen,* K. L. Hertel, University of Tennessee, Knoxville; Newton Underwood, Vanderbilt University, Nashville.

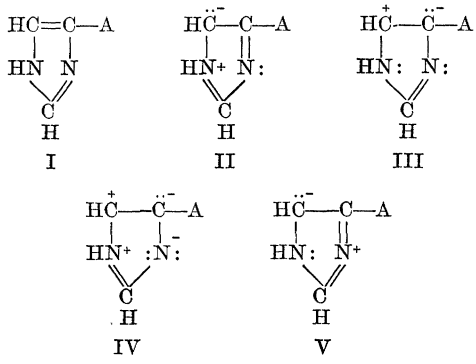
J. T. McGILL

SPECIAL ARTICLES

RESONANCE AND THE CHEMISTRY OF HISTIDINE

CERTAIN of the chemical characteristics of histidine, β-imidazole-α-amino propionic acid, are readily explained by a consideration of the fifteen principal forms of the compound in resonance. These forms can be classified into four groups, each containing structures with similar energies, and the forms of each group may be weighted somewhat arbitrarily (any reasonable assignment of weights leads to the same qualitative results).

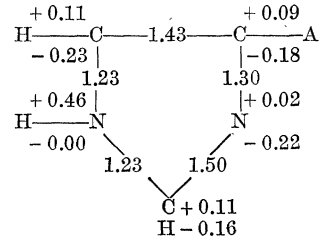
Group 1: The classical structure of histidine (I), where A is the alanine residue. Weighting 30. *Group 2:* Four forms in which there is a single separation of charge and octets about all atoms, *e.g.,* II. Weighting 10. *Group 3:* Five forms containing a single separation of charge, but a sextet about one atom, *e.g.,* III. Weighting 5. *Group 4:* Four structures with a double separation of charge and a sextet about one atom, *e.g.,* IV. In addition, V is placed in this group, due to the two adjacent double bonds. Weighting 2.



I II III IV V

The resonance forms of the other tautomer of histidine are the same, with the functions of the ring nitrogens reversed.

The degree of bondedness between each pair of atoms of the ring and the charges on each atom are summarized in VI as time averages, where one unit of charge (charge on an electron) or bondedness (single bond) is equivalent to a total weighting of 105 (30 + 40 + 25 + 10).



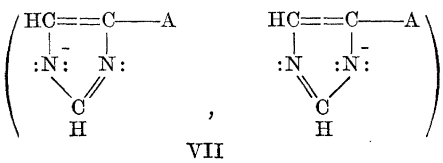
VI

Recalling that a double bond to a nitrogen or a positive charge decreases its basicity and increases its acidity, while a negative charge has the reverse effect, the nature of the three nitrogen atoms of histidine may now be considered. (1) The amino nitrogen of the chain does not participate in the resonance of the ring and therefore is similar both in reactivity and basicity to α-amino groups of other amino acids (*i.e.,* expected pK_b about 5). (2) The secondary amine nitrogen of the ring has a charge of almost $+\frac{1}{2}$ and is not quite $\frac{1}{2}$ double bonded on either side. The similar resonance in pyrrole gives its nitrogen about the same positive charge and degree of bondedness. Thus this nitrogen in histidine, like that of pyrrole, is very weakly basic

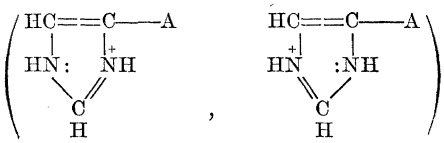
and abnormally acidic. (3) The third nitrogen, resembling in the classical structure the nitrogen of pyridine, possesses a net charge of $-1/5$. It is double bonded 3/10 on one side and 1/2 on the other. On the other hand, the resonance in pyridine gives the nitrogen a relatively small negative charge and makes it slightly less than half double bonded on each side. As a result the histidine nitrogen is much more basic than that of pyridine.

It is doubtful that the negative charge of this "pyridine" nitrogen can entirely compensate for its doublebondedness, and it is therefore probably less basic than the nitrogen in the alanine residue. The formation of salts of histidine indicates two basic nitrogens, $pK_b = 4.85$ and $pK_b = 7.97$, of which the first corresponds to the nitrogen of an amino acid, and the second to a nitrogen much more basic than that of pyridine.

A similar treatment of the nitrogens of histidine could have been accomplished by a consideration of the alternative resonances VII and VIII,[1] in which the ring has a net charge.



VII



VIII

Substitutions that depend on the basicity of the nitrogen occur at the amino group, *e.g.*, $C_3N_2H_3 \cdot CH_2 \cdot CHNH_2 \cdot COOH + C_6H_5 \cdot SO_2Cl \rightarrow C_3N_2H_3 \cdot CH_2 \cdot CHNH\,(C_6H_5SO_2) \cdot COOH + HCl$. Those that depend on removing a hydrogen by a metal, which is afterwards replaced by a group, occur on an acidic nitrogen of the ring, *e.g.*, $AgN_2C_3H_2 \cdot CH_2 \cdot CHNH_2 \cdot COOAg + 2CH_3I \rightarrow CH_3N_2C_3H_2 \cdot CH_2 \cdot CHNH_2 \cdot COOCH_3 + 2AgI$.

TERRELL L. HILL
GERALD E. K. BRANCH

UNIVERSITY OF CALIFORNIA,
BERKELEY

FURTHER EVIDENCE OF SEX VARIATION IN THE UTILIZATION OF IRON BY ANEMIC RATS

PREVIOUS work in this laboratory has indicated that female anemic rats utilize iron more efficiently than male anemic rats. This conclusion was based on the greater gain of the hemoglobin per 100 milliliters of blood shown by female rats when both male and female rats were fed the same iron supplement.[1] The question has been raised as to whether this is really a characteristic of the female organism or is simply due to the difference in the size of the two sexes and therefore different food intakes.[2] In the hope of shedding further light on this question, the total iron content of anemic male and female rats which have been given supplemental iron feedings has been investigated.

Anemia was produced in young rats using the Elvehjem-Kemmerer technique,[3] modified only by the feeding of copper sulfate supplements, as is customary in this laboratory.[4] When the rats were eight weeks old, the hemoglobin level was constant or falling, and ranged from 2.9 to 5.4 gms Hb/100 ml. of blood for the six litters used in this work. At this time representative rats from each litter were killed, while the rest of the rats were placed on $FeCl_3$ supplements varying from 0.05 mg Fe to 0.3 mg Fe daily. At the end of the six weeks' test period and twenty-four hours after the feeding of the last iron supplement, the hemoglobin concentration was determined by the acid-hematin method and the rats killed. After the removal of the digestive tract, the rats were ashed and the hydrochloric acid solution of the ash analyzed for iron, using the method of Farrar.[5]

Results of the iron analyses of the rats given comparable treatment showed remarkable agreement. Six male and seven female rats were killed at the end of the five weeks' iron depletion period on the basal milk diet. Although the hemoglobin concentration ranged from 2.9 to 5.4 gms of Hb/100 ml. blood, the iron content varied only from 0.013 to 0.017 mg Fe/gm of rat with an average of 0.015 mg Fe/gm for both male and female rats. However, as the females weighed a little more than the males, the total iron content of the females (0.8073 mg Fe) was slightly higher than that of the males (0.7065 mg Fe). In another series of experiments the iron depletion period was continued for eleven weeks and although there was a definite decrease in both the hemoglobin concentration and the iron content, again no sex difference in the iron content of rats which had their reserves depleted on a milk diet régime could be detected.

However, Table I shows the results obtained when litter mate rats rendered anemic by the usual procedure were fed varying amounts of iron as $FeCl_3$ solution. It can be seen that the mg Fe/gm of rat was always higher for the females than for the males fed

[1] Jukes and Branch, SCIENCE, 80: 228, 1934.

[1] M. C. Smith and L. Otis, SCIENCE, 85: 125–126, 1937.

[2] H. H. Mitchell and T. S. Hamilton, SCIENCE, 85: 364–366, 1937.

[3] C. A. Elvehjem and A. R. Kemmerer, *Jour. Biol. Chem.*, 93: 189–195, 1931.

[4] M. C. Smith and L. Otis, *Jour. Nutr.*, 14: 365–369, 1937.

[5] G. E. Farrar, Jr., *Jour. Biol. Chem.*, 110: 685–694, 1935.