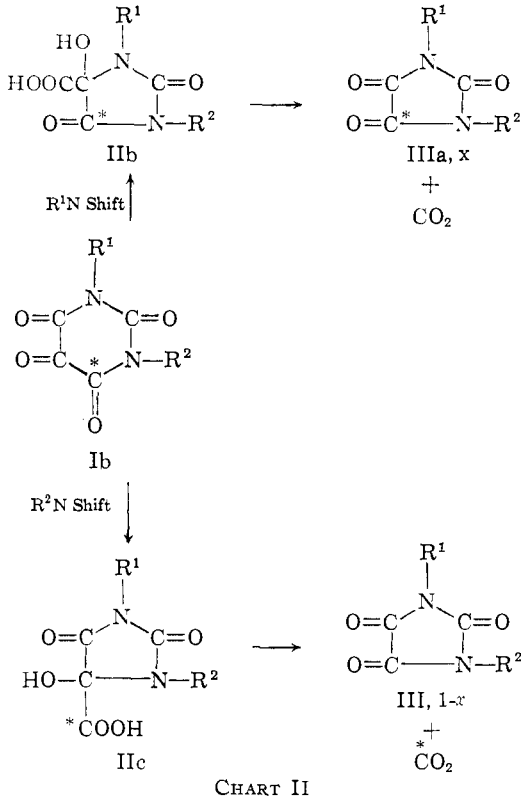


CHART II

next were prepared; such derivatives, designated Ib (Chart II), were subjected to most of the same conditions of rearrangement as was the isotope position isomer Ia. The values ($x$) and ($1 - x$) are the mole fractions of product formed through shift of $R^1N$- or $R^2N$, respectively. If no isotope effect is exhibited, it would be expected, when $R^1 = R^2$, that $x = 0.500$, whereas if $R^1 \neq R^2$, $x$ could have values of zero to unity. The results of experiments with Ib are given in Table I.

Although the data of Table I deviate somewhat from the theoretical values of $x = 0.500$ and $x = 1.00$ (excluding for the moment lines 8 and 9 concerning N-methylalloxan-4-$C^{14}$), the deviations are no greater than those to be expected from the operation of normal intramolecular or intermolecular isotope effects. We therefore conclude that: (a) each rearrangement takes place with exclusive shift of nitrogen, rather than of carbon, and (b) in the rearrangements of N-phenylalloxan-4-$C^{14}$ the shift of N–Ph takes place to the exclusion of unsubstituted nitrogen.[6] Turning now to the data for the rearrangement of N-methylalloxan-4-$C^{14}$ (lines 8 and 9, Table I), auxiliary experiments with N-methylalloxan-5-$C^{14}$ [Ia, $R^1 = CH_3$, $R^2 = H$] indicate that the parabanic acid [IIIa, $R^1 =$ $CH_3$, $R^2 = H$] obtained upon rearrangement at $p$H < 1 contained 93.7% and at $p$H 7–8 contained 96.7% of the original radioactivity of reactant N-methylalloxan-5-$C^{14}$. From lines 8 and 9 (Table I) it can thus be calculated that the migration ratios, or "migratory aptitudes," of $CH_3$-N *versus* H–N under the two reaction conditions studied are 3:1 and 4:1, respectively.

alkali to produce the mono-substituted iminobarbituric acid [W. Traube, *Ber.*, **33**, 3039 (1900)]. Hydrolysis of the imine to the barbituric acid with 6 *N* HCl and then chromic acid oxidation[4] produced the N-methyl- or N-phenylalloxan-4-$C^{14}$. Alloxan-4-$C^{14}$ and N,N-dimethylalloxan-4-$C^{14}$ were prepared according to the procedure of ref. 4, using carboxyl-labeled malonic ester and urea or dimethylurea as reactants.

(6) This result is to be contrasted with the observations of W. E. Doering, T. I. Taylor and E. F. Schoenewaldt, *J. Am. Chem. Soc.*, **70**, 455 (1948), and of O. K. Neville, *ibid.*, **70**, 3499 (1948), that in the rearrangement of phenylglyoxal to mandelic acid it is only the hydrogen which undergoes migration.

TABLE I

RADIOCHEMICAL RESULTS OF THE REARRANGEMENT OF ALLOXAN-4-$C^{14}$ AND DERIVATIVES [Ib]

| $p$H *ca.* | Substituent in I, II and III $R^1$ | $R^2$ | $x^a$ | $1 - x^b$ |
|---|---|---|---|---|
| <1 | H | H | 0.533 | 0.467 |
| 7.5 | H | H | .542 | .458 |
| 9.4 | H | H | .503 | .497 |
| 7–10 | H | H | .498 | .502 |
| >13 | H | H | .550 | .450 |
| <1 | $CH_3$[c] | $CH_3$[c] | .528 | .472 |
| 7–8 | $CH_3$ | $CH_3$ | .529 | .471 |
| <1 | $CH_3$[d] | H[d] | .754 | .246 |
| 7–8 | $CH_3$ | H | .803 | .197 |
| <1 | Ph[e] | H[e] | .973 | .027 |
| 7–8 | Ph | H | .960 | .040 |
| >13 | Ph | H | .990 | .010 |

[a] Based on radioactivity assay of the appropriate parabanic acid [III]. [b] By difference. [c] I and III, $R^1 = R^2 = CH_3$: E. Fischer, *Ber.*, 14, 1912 (1881); A. Strecker, *Ann.*, **118**, 174 (1861). [d] I, II and III, $R^1 = CH_3$, $R^2 = H$: E. Fischer, *Ber.*, **15**, 455 (1892); H. B. Hill, *ibid.*, 9, 1092, 1093 (1876); I and III, $R^1 = Ph$, $R^2 = H$: N. M. Winslow, *J. Am. Chem. Soc.*, 61, 2089 (1939); H. Kammerer, *Ber.*, 40, 3741 (1907).

(7) This paper is based upon work performed at Oak Ridge National Laboratory, which is operated by Union Carbide Corporation for the Atomic Energy Commission.

(8) Predoctoral Fellow of the Oak Ridge Institute of Nuclear Studies.

DEPARTMENT OF CHEMISTRY[7]
UNIVERSITY OF DELAWARE
NEWARK, DELAWARE
HAROLD KWART
RICHARD W. SPAYD[8]

CHEMISTRY DIVISION
OAK RIDGE NATIONAL LABORATORY
OAK RIDGE, TENN.
CLAIR J. COLLINS

RECEIVED APRIL 24, 1961

## AMIDO NITROGEN MIGRATION IN A BENZILIC ACID REARRANGEMENT AS PART OF THE RING CONTRACTION OF ISOQUINOLINEDIONES TO PHTHALIMIDES.

*Sir:*

Kwart and Sarasohn in a paper just appeared[1] have raised the question of whether amido-nitrogen migration may occur in the benzilic acid rearrangement of alloxan to alloxanic acid; the reaction previously has been interpreted both as occurring by migration of carbon[2] and by migration of nitrogen.[3] We wish to report a resolution of this problem for a system containing a somewhat similar heterocyclic ring, phthalonimide(III). This compound and its N-methyl derivative, like alloxan, dissolve readily in aqueous alkali, presumably because of formation of the anion of a *gem*-diol derived from the ketonic carbonyl.

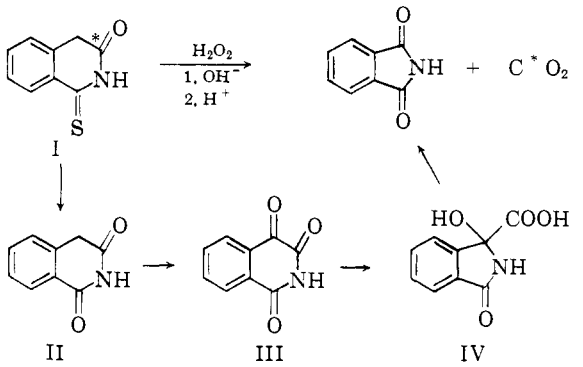
Our investigations have been concerned with a

(1) H. Kwart and I. M. Sarasohn, *J. Am. Chem. Soc.*, **83**, 909 (1961).

(2) S. Selman and J. F. Eastham, *Quart. Revs.*, **14**, 221 (1960).

(3) H. Biltz, M. Heyn and M. Bergius, *Ann.*, **413**, 68 (1916).

novel, one-step, ring-shrinking process, in which the benzilic acid rearrangement appears to constitute a phase, which has the effect of converting 1,3(2,4)-isoquinolinediones (II) (homophthalimides) or their monothio analogs (I) to phthalimides. The change is accomplished by treatment with excess hydrogen peroxide and aqueous alkali at room temperature or slightly above for periods of less than three hours, then acidification and brief warming. We have effected it with seven examples (2a-thiohomophthalimide, 4-methyl-, 4-chloro-, 5-methyl-, 5-methoxy-, and 5,6-benzo-thiohomophthalimides, and N-methylhomophthalimide) in yields of 46 to 81%.




The reaction with monothiohomophthalimides, with which most of our experiments were carried out, proceeds by initial oxidative desulfurization to homophthalimides, which can be isolated in good yield if the amount of peroxide is limited to four molar equivalents. Oxidation to phthalonimides (III) through epoxidation of the enol double bond presumably follows, but such compounds cannot be isolated, for they are too rapidly altered in alkaline solution[4] (oxidation in acid solution gives isolable phthalonimide, however[4]). If the peroxide is destroyed before acidification, a substance is formed which agrees with the described[4] alteration product of phthalonimide (largely 3-hydroxy-phthalimidine-3-carboxylic acid (IV), "phthalonamic acid"[4]), but not phthalimide. If the reaction mixtures (containing excess peroxide) are kept cold during acidification, phthalimide is not formed and carbon dioxide is not evolved until the mixture subsequently is warmed gently. Phthalonimide prepared independently gives the same products as homophthalimide under similar conditions.

Although the over-all reaction in effect deletes a methylene group, it is the carbonyl group of I that is eliminated. When the reaction was carried out with thiohomophthalimide prepared[5] from phenylacetic acid 1-$C^{14}$, nearly all the radioactivity appeared in the evolved carbon dioxide. The carbon dioxide obtained by the Hofmann rearrangement of the resulting phthalimide had activity only just detectably above normal background. The ring-shrinking step is thus a rearrangement of the benzilic acid type, in which the bond-breaking and bond-forming steps occur at a nitrogen atom. Instances of benzilic acid rearrangement in a nitrogenous heterocyclic system have been reported before,[3] but we believe this is the first case where migration of nitrogen has been established.

In view of the possibility that the ring-shrinking step occurs through a phthalonimide anion and an intermediate three-membered ring, analogous to the hetero Favorskii rearrangement proposed by Sarel and Greenberger,[6] we carried out the reaction with 2-methyl-1,3(2,4)-isoquinolinedione (N-methylhomophthalimide), also labelled at the 3-position (the "aliphatic" carbonyl). The same over-all conversion occurred readily, and N-methylphthalimide was obtained in good yield. However, the evolved carbon dioxide had only 85% of the radioactivity to be expected if it had been derived solely from the 3-carbonyl, and a significant amount of isotope was detected in the carbon dioxide derived from the resulting N-methylphthalimide carbonyls by conversion to phthalimide followed by Hofmann rearrangement. Thus even when phthalonimide anion cannot be formed, migration by nitrogen predominates, although aryl migration becomes a minor competing process. Phthalamic acids are not intermediates, for they are not converted to phthalimide under the reaction conditions; hydrolytic ring opening to a phthalonamic acid followed by reclosure at the ketonic carbonyl is not ruled out (nor is the analogous scheme for the rearrangement of alloxan), although there are reasons to doubt it. A mechanism analogous to one proposed for the rearrangement of alloxan at high *p*H by Kwart and Sarasohn[1] conforms best to the facts, and our results provide strong support for their proposal.

Because of its simplicity, mildness, and satisfactory yields, we believe this reaction has useful application in synthesis as well as structure proof. It already has enabled us to identify as 2a-thiohomo-1,2-naphthalimide (5,6-benzo-1-thio-1,3(2,4)-isoquinolinedione) the compound earlier believed[5] to be its isomer, 8a-thio-1-homo-1,8-naphthalimide; both give by simple hydrolysis decarboxylic acids of the same melting point, but the imides of one less carbon are strongly differentiated. Furthermore, by a combination of the present reaction with the recently reported synthesis of thiohomophthalimides,[5] some phthalimides can be synthesized that were previously available only by circuitous routes.

(4) S. Gabriel and J. Colman, *Ber.*, **33**, 996 (1900).
(5) P. A. S. Smith and R. O. Kan, *J. Am. Chem. Soc.*, **82**, 4753 (1960).
(6) S. Sarel and A. Greenberger, *J. Org. Chem.*, **23**, 330 (1958).

DEPARTMENT OF CHEMISTRY PETER A. S. SMITH
UNIVERSITY OF MICHIGAN
ANN ARBOR, MICH. ROBERT O. KAN

RECEIVED MARCH 18, 1961

## EVIDENCE FOR MALIC SYNTHETASE IN ANIMAL TISSUES

*Sir:*

Malic synthetase, one of the key enzymes of the glyoxylate cycle,[1] was demonstrated both in microorganisms[2,3] and in plants.[4,5] Recent report by Madsen[6] indicated that the key enzymes of the

(1) H. L. Kornberg and H. A. Krebs, *Nature*, **179**, 988 (1957).
(2) D. T. O. Wong and S. J. Ajl, *J. Am. Chem. Soc.*, **78**, 3230 (1956).
(3) H. L. Kornberg, *Biochem. J.*, **68**, 549 (1958).
(4) H. L. Kornberg and H. Beevers, *Biochim. Biophys. Acta*, **26**, 531 (1957).
(5) C. Bradbeer and P. K. Stumpf, *J. Biol. Chem.*, **234**, 498 (1959).
(6) N. B. Madsen, *Biochim. Biophys. Acta*, **27**, 199 (1958).