[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF THE UNIVERSITY OF CALIFORNIA.]

THE RATES OF CONVERSION OF THE STEREOISOMERIC CINCHONA ALKALOIDS INTO THEIR TOXINES.[1]

(ON CATALYSES WITH WEAK ACIDS. IX.)

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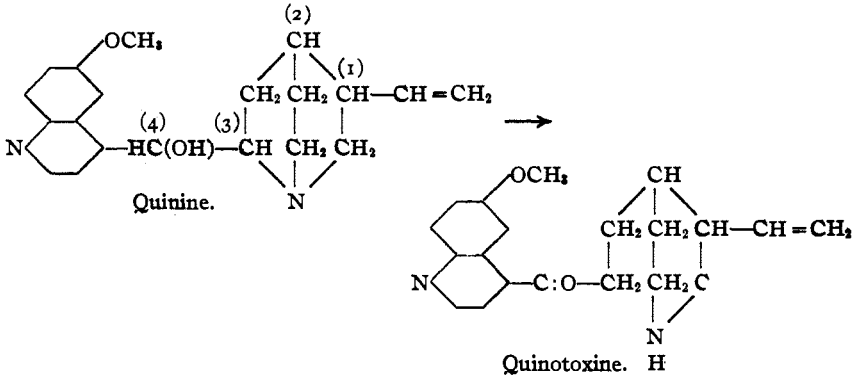
Received February 1, 1916.

1. Introduction.

The stereoisomeric relation between cinchonine and cinchonidine renders of interest the comparative rates of conversion of these two alkaloids into cinchotoxine. Like interest centers about the rates of conversion of quinine and quinidine into quinotoxine. The relation between each pair of alkaloids and its corresponding toxine may be graphically represented as follows:



Quinidine, it may be noted, is stereoisomeric with quinine. Cinchonine and cinchonidine are also stereoisomeric and these compounds and cinchotoxine differ from quinine, quinidine and quinotoxine in constitution only in that they have hydrogen in place of the methoxy group.

Of importance in the consideration of these alkaloids is the question as to the exact differences in stereoisomerism. In the formulas of the two groups of alkaloids given there are present in each case, as is seen, four asymmetric carbon atoms. As has been shown from the numerous investigations of Skraup, König, and Rabe[2] both in the case of quinine and quinidine and in that of cinchonine and cinchonidine; two (1 and 2) of the three asymmetric carbon atoms within the bridged ring[3] do not differ in configuration. The third asymmetric carbon atom (3) within

[1] THIS JOURNAL, 37, 2088 (1915) and preceding articles.

[2] *Ann.*, 373, 91 (1910). Through an oversight, Rabe was previously (THIS JOURNAL, 37, 2083 (1915)) *incorrectly* represented as attributing the stereoisomerism of cinchonine and cinchonidine *solely* to the spatial arrangement about the third asymmetric carbon atom (3) within the ring.

[3] According to a suggestion of König's a bridged ring of this type may be designated as a quinuclidine ring. *Ber.*, 37, 3244 (1904); 38, 3049 (1905).

the ring, however, presents the possibility of an antipodal spatial arrangement and, as Rabe concludes from the relation of desoxycinchonine to desoxycinchonidine and of desoxyquinine to desoxyquinidine, such antipodal configuration actually exists about this carbon atom in the case of each of the two pairs of isomers. But apparently conflicting evidence arises from the fact that both cinchonine and cinchonidine are oxidized to the same cinchoninone and quinine and quinidine under like conditions yield the same quinoninone. This finds reasonable explanation, however, as Rabe clearly shows in the keto-enol tautomerism presented by the oxidation product. This tautomerism points to a dynamic equilibrium existing between four forms (two keto, two enol). The explanation is upheld by the phenomena of mutarotation shown by the salts of cinchoninone and those of quinoninone.

As to the remaining asymmetric carbon atom (4), Rabe calls attention to the fact that experimental evidence is insufficient to draw a definite conclusion here regarding the configuration presented in the case of either pair of isomers. More recently Kaufmann and Huber[1] have succeeded in reducing hydrocinchoninone to a mixture of hydrocinchonine (over 50%) and of hydrocinchonidine (about 10%). While not advancing this as any evidence regarding the configuration about the fourth asymmetric carbon atom, these investigators conclude that hydrocinchoninone itself cannot be a mixture of two optical isomers in equivalent quantities. Definite evidence regarding the configuration about this asymmetric carbon atom must, consequently, await further investigation.

In the study of the conversion of quinine and quinidine into quinotoxine which has been carried out in this laboratory,[2] it has been shown that the same laws obtain as in the corresponding conversion of cinchonine and cinchonidine into cinchotoxine. The rates of conversion as in the latter case are directly proportional to the concentration of the univalent alkaloid ion and furthermore these rates are a linear function of the concentration of the undissociated organic acid, that is to say, the increments in speed are directly proportional to the increasing concentration of the undissociated organic acid.[3] In comparing the conversion rates of quinine and quinidine, however, it was noted that the speeds in cases under like conditions approach a common value. This raised the question as to whether the slightly different rates previously observed under like conditions in the case of cinchonine and cinchonidine[4] were real or only

[1] *Ber.*, 46, 2913 (1913).

[2] Unpublished thesis of H. N. Massey.

[3] In view of the extraordinary nature of the catalysis of the cinchona alkaloids and its dependence upon the univalent ion, the study is being extended to the alkyl cinchona salts in which, in one case, the alkyl radical is attached to the quinoline nitrogen and, in the other, to the quinuclidine nitrogen.

[4] Biddle and Butzbach, THIS JOURNAL, 37, 2082 (1915).

apparent. A reconsideration of these rates has shown that in the case of the two alkaloids, the speed of change to a mean rotational value under like conditions approaches identity within the limits of experimental error, and that furthermore this mean rotational value probably represents the true rotation of the completely converted, *pure*, *nonracemized* cinchotoxine.

2. The Optical Rotation of Cinchotoxine.

As can readily be shown from a consideration of the equations employed in the calculation of reaction rates, slight variations in the rotatory values assigned to cinchotoxine would suffice to account for slight, but appreciable errors. In comparing reaction rates of stereoisomeric alkaloids it is, consequently, of importance to fix the rotatory value of the cinchotoxine with some degree of definiteness. The rotation (Ventzke degrees) observed by Biddle and Rosenstein[1] in a decimeter length of a 0.1 molal solution of cinchotoxine in 1.7 molal acetic acid at 18°, was 2.08° and in 3.2 molal acetic, was 2.70°. The value later noted by Biddle and Brauer[2] under like conditions in concentrations of acetic acid varying from 0.4 to 1.0 molal was 3.00°. This last value was employed by them in all their calculations.

The rotatory power of cinchotoxine salts has been studied by several investigators whose results are not in full agreement. Some preliminary measurements were made by Howard[3] in 1872. Thereafter, Hesse[4] secured definite measurements of the oxalate which were subsequently confirmed by Roques.[5] The results obtained by the last two investigators are given in Table I, the specific rotations being reduced to Ventzke degrees and calculated for 0.1 molal concentrations of the alkaloid.

TABLE I.

No.	Investigator.	Substance.	Concentration of alkaloid used.	Rotation referred to alkaloid as 0.1 molal.
1	Hesse	Cinc. oxalate, $(C_{19}H_{22}N_2O)_2C_2H_2O_4 + 3H_2O$ (from cinchonine)	0.0546	2.40°
2	Hesse	Cinc. oxalate (from cinchonidine)	0.0546	2.38°
3	Hesse	Cinc. oxalate + 2 mols. H_2SO_4	0.0546	2.72°
4	Roques	Cinc. + 2 mols. HCl	0.0340	2.434°

The salts employed by Hesse were prepared from amorphous cinchotoxine; those of Roques, from the crystallized base. Later, Roques more carefully prepared his material and from salts of the more highly purified base obtained a rotation higher than that previously observed and which is in close agreement with the value obtained by Biddle and

[1] THIS JOURNAL, **35**, 421 (1915).
[2] *Ibid.*, **37**, 2065 (1915).
[3] *J. Chem. Soc.*, **25**, 102 (1872).
[4] *Ann.*, **166**, 277 (1873); **178**, 262 (1875).
[5] *Compt. rend.*, **120**, 1170 (1895); *Bull. soc. chim.*, [3] **13**, 1005 (1895).

Brauer. In Table II are presented for comparison the values for equivalent quantities of the alkaloid, reduced in all cases to Ventzke degrees. Measurements given in both tables were in general made at temperatures ranging from 15° to 18°.

TABLE II.

No.	Investigator.	Substance.	Concentration of alkaloid used.	Rotation referred to alkaloid as 0.1 molal.
1	Roques	Cinc. + 2 mols. HCl (from cinchonine)	0.034	3.20°
2	Roques	Cinc. + 2 mols. HCl (from cinchonidine)	0.034	3.26°
3	Roques	Cinc. + 4 mols. HCl (from cinchonine)	0.034	3.02°
4	Roques	Cinc. + 4 mols. HCl (from cinchonidine)	0.034	2.95°
5	Roques	Cinc. oxalate, $(C_{19}H_{22}N_2O)_2C_2H_2O_4 + 4H_2O$ (from cinchonine)	0.0296	3.16°
6	Roques	Cinc. oxalate (from cinchonidine)	0.0296	2.98°
7	Roques	Cinc. nitrate, $C_{19}H_{22}N_2O.HNO_3$	0.028	3.04°
8	Biddle and Brauer	Cinc. + 4 mols. to 10 mols. acetic acid	0.10	3.00°

It is of interest to note in this connection that the rotations of equivalent solutions of cinchotoxine are practically identical irrespective of whether the base is derived from cinchonine or cinchonidine. Both Howard and Hesse earlier observed the same fact when dealing with impure cinchotoxine. As Roques points out, this leads to the conclusion that the impurity arising in cinchotoxine during the process of formation is identical irrespective of the source from which the base is derived.

The final results obtained, as Roques indicates, are of further interest in that they point to the apparent correctness of an hypothesis first postulated by Howard, but which from his data he was unable to establish, *viz.*, that the rotatory power of cinchotoxine should be the arithmetical mean of those of cinchonine and cinchonidine.

Furthermore, as is seen, the solutions given in Table II, even though presenting considerable variation in concentration of H^+ ion, yet differ little in rotation. It seems reasonable, consequently, to assume, as Biddle and Brauer have done, that rotational variations due to this cause are in this case negligible in calculating the reaction rates.

3. The Conversion Rates of Cinchonine and Cinchonidine to Cinchotoxine.

From the studies of Hesse and Roques it seems evident then that in the process of the conversion of cinchonine or cinchonidine into cinchotoxine some other reaction takes place to a slight extent with the result that the unpurified product gives a rotation lower than that of pure cinchotoxine. Such a side reaction would to some extent affect the quantitative data employed in studying the relative speed of conversion of the two alkaloids into cinchotoxine. The error, though naturally slight, would lead to an apparent increase in the speed of conversion of the cinchonine and an apparent decrease in that of the cinchonidine.

In order that a direct comparison might be made with regard to the simultaneous conversion of the two alkaloids under the same conditions, experiments were carried on with equivalent mixtures of cinchonine and cinchonidine dissolved in an excess of acetic acid. Assuming that the two alkaloids do not mutually interfere with each other, opportunity is thus afforded to observe the variations in rotation during the course of the reaction. In view of the fact that solutions of the cinchona alkaloids become discolored much less rapidly in the absence of the oxidizing influence of air, the heating was carried out in sealed tubes which had been exhausted until the pressure was reduced to 20 mm. The removal of the air was of additional advantage in that changes in rotation during equal time intervals were found to be to a considerable extent decreased thereby. Both these facts point to the conclusion that the air is one of the factors at least in part responsible for deviations in these reactions.

The results of an experiment are given in Table III.

TABLE III.

Cinchonine and cinchonidine, each 0.05 molal. Acetic acid, 1.7 molal. Temp., 99.4° (±0.2°).

Time.....	0 hr.	4 hrs.	6 hrs.	23 hrs.	25 hrs.	52 hrs.	111 hrs.	335 hrs.
Rotation[1].....	3.21°	3.11°	3.00°	2.55°	2.55°	2.25°	1.75°	1.00°

A consideration of this table is instructive. As is seen, there is a gradual but continuous fall in rotation to the close of the period, even though the conversion to cinchotoxine may, for all practical purposes, be regarded as complete before the close of the first fifty hours.

If now the rotation of the cinchotoxine were assumed to be that of the sixth reading, 2.25°, the gradual fall in rotation observed cannot be accounted for in case the rate of conversion of the cinchonine either exceeds or equals that of cinchonidine. The observed readings might be obtained, however, as can be shown by calculation, if the conversion rate of the cinchonidine slightly exceeded that of its isomer. But the assumption of a rotational value of 2.25° in the above experiment, fails entirely to explain the continued fall in rotation after the conversion to cinchotoxine is practically complete.

If, on the other hand, the rotatory power of cinchotoxine is the arithmetical mean of those of the other two alkaloids, then, as will be evident, the observed rotations should remain constant throughout the conversion, unless the rate of conversion of one alkaloid exceeds that of the other, or unless such constancy is disturbed by side reactions. The direction of deviations in rotation, if dependent upon difference in rate, would naturally be determined by which of the two alkaloids presented the more rapid rate of conversion. The decrease in rotation observed might at first glance seem to favor the conclusion that the rate of conversion

[1] Readings made in this and the following experiments at 18°.

of cinchonine exceeds that of cinchonidine as previously reported. The fall in rotation would in such case, however, not be continuous, but after attaining a maximum value would return to the original reading. Thus, for example, the final reading in Table III should, under these conditions, practically equal the initial reading of 3.21°.

If, then, we assume that the specific rotation of cinchotoxine is the arithmetical mean of those of the other two alkaloids, the conclusion must be reached that the continuous fall in rotation must be due to a secondary reaction, or reactions, the nature of which will be considered later in this paper.

It will be noted from Table III that the fall in rotation amounts only to about 0.2° in six hours. If, consequently, the rates of conversion of cinchonine and cinchonidine into cinchotoxine were studied during the early part of the reaction under the given conditions, the error introduced by this deviation may be reduced to a minimum. With this in view, the conversion rates of the two alkaloids were remeasured simultaneously for one concentration of acetic acid under precisely the same experimental conditions. The mode of procedure had the additional advantage of eliminating other slight errors which may have arisen in previous determinations from differences in manipulation due to the personal equation, since the former measurements for cinchonine were made by one experimenter and those for cinchonidine by another. The solutions were heated, as in the previous experiment, in sealed tubes evacuated to a pressure of 20 mm. The arithmetical mean of the initial readings of the two alkaloids is chosen as the rotation of the corresponding cinchotoxine solution. The results are recorded in Tables IV and V.

TABLE IV.

Cinchonine, 0.1 molal; Acetic acid, 1.7 molal; L = 1.0 dm.; T = 99.4 (±0.2°).

t.	α.	K_1.	K_2.
0 hr.	20.9	....	....
0.5 hr.	19.5	0.0716	0.0716
1.5 hrs.	17.1	0.0701	0.0694
2.5 hrs.	14.9	0.0722	0.0752
3.5 hrs.	12.9	0.0749	0.0818
4.5 hrs.	11.8	0.0699	0.0525
5.5 hrs.	10.2	0.0736	0.0900
α hrs.	3.25	....	....
	Mean,	0.0721	0.0734

TABLE V.

Cinchonidine, 0.1 molal; Acetic acid, 1.7 molal; L = 1.0 dm.; T = 99.4 (±0.2°).

t.	α.	K_1.	K_2.
0 hr.	—14.4	....	....
0.5 hr.	—13.0	0.0716	0.0716
1.5 hrs.	—10.6	0.0701	0.0694
2.5 hrs.	— 8.6	0.0692	0.0678
3.5 hrs.	— 6.7	0.0712	0.0759
4.5 hrs.	— 5.3	0.0699	0.0658
5.5 hrs.	— 4.0	0.0703	0.0717
α hrs.	3.25	....	....
Mean,		0.0704	0.0704

In the light of the facts already discussed, a consideration of the results given in Tables IV and V leads to the unavoidable conclusion that the rates of conversion of cinchonine and cinchonidine to cinchotoxine are identical within the limits of experimental error, providing we assume

that the specific rotation of cinchotoxine is the arithmetical mean of the corresponding rotations of the other two alkaloids. A reaction rate *apparently* slightly greater in the case of the cinchonine was to be expected. Small differences in rate, if at all existent, can be detected only by more refined experimental methods. The same principle obtains apparently as well with respect to the rates of conversion of quinine and quinidine into quinotoxine.

In order that confirmatory evidence might be obtained regarding the comparative reaction rates of the two alkaloids, recourse was had to a former method of measurement. Unfortunately, the procedure of Biddle and Rosenstein based upon the separation of the alkaloid from cinchotoxine by means of ammonium hydroxide in the presence of an ammonium salt was not readily applicable, since cinchonidine is considerably more soluble under these conditions than is cinchonine. A modification of the earlier ether extraction method, however, proved of service. The cinchotoxine was weighed in each case as such, being freed from traces of unchanged alkaloid by re-solution in *absolute* ether in which both cinchonine and cinchonidine are difficultly soluble. While the method naturally does not present the accuracy[1] of that dependent upon optical rotation, the results obtained fully agreed with those already given as to the identity of the two rates of conversion.

We must conclude then, that *in the conversion of stereoisomeric cinchona alkaloids into their toxines as represented on the one hand by cinchonine and cinchonidine to cinchotoxine and on the other by quinine and quinidine to quinotoxine, the rates of conversion of stereoisomers, as indicated by the rates of approach to a position of mean optical rotation, are identical within the limits of experimental error.*

4. Cause of the Diminishing Rotation.

Roques accounted for the low specific rotations first observed in the study of cinchotoxine by assuming the presence of a common impurity irrespective as to whether the toxine was derived from cinchonine or from cinchonidine. What the nature of this impurity was, however, he did not venture to suggest.

In considering the data presented in Table III, one is struck with the fact that the fall in rotation is *continuous* throughout the entire period of the reaction, regardless as to whether the solution at any moment contained three optically active substances or the cinchotoxine alone. While under ordinary conditions the oxidizing influence of the air may have some influence upon this change, it is obvious that under the ex-

[1] It is to be regretted that apart from the data presented by optical rotation a quantitative method is lacking which would enable us to measure with precision small differences of reaction rate. Some such method is to be desired in order that the comparative conversion rates of stereoisomers may be subjected to more rigid examination.

perimental conditions employed such influence is totally inadequate to account for the results obtained. Indeed, in view of the facts presented only one reasonable explanation can be offered and that is one dependent upon the slow racemization of the optically active substances present. In the protracted heating of the cinchona alkaloids under the conditions of the experiment, partial racemization might naturally be expected. In the case of cinchonine and cinchonidine this racemizing action might extend to any one or all of the four asymmetric carbon atoms (see graphic formulae at beginning of this paper); in the case of cinchotoxine it would naturally be confined to carbon atoms one and two.

A slow racemizing action of this kind fully accords with the facts observed. Thus, it accounts for the supposed impurity which Roques thought was derived indifferently either from cinchonine or cinchonidine, since, as he noted, both these alkaloids were converted under similar conditions to a cinchotoxine of like low rotation. It explains further the divergent specific rotations of cinchotoxine given in the literature and indicates why extraordinary care was found necessary to obtain a cinchotoxine of high rotation.

As a result of this racemizing influence the reaction rates obtained by Biddle and Brauer for cinchonine are a little high and those given by Biddle and Butzbach for cinchonidine are a little low. These factors, however, materially affect only the comparative rates of conversion of the two alkaloids. The influence upon the data establishing the two principles, *viz.*, the action of univalent alkaloidal ion and that of the concentration of the undissociated organic acid upon the reaction, is consequently negligible.

Summary.

1. The specific rotatory power of cinchotoxine closely approaches, if it does not equal, the arithmetical mean of the corresponding rotatory powers of the isomeric cinchonine and cinchonidine.

2. In the conversion of the two alkaloids into the common toxine under the influence of an organic acid the rotational values are subject to a slight but continuous error, due to a partial racemization of the alkaloids concerned. The effect of this error upon the reaction rates calculated is that those obtained for cinchonine will invariably be a little high and those for cinchonidine will be a little low.

3. In the conversion of stereoisomeric cinchona alkaloids into their toxines as represented on the one hand by cinchonine and cinchonidine to cinchotoxine and on the other by quinine and quinidine to quinotoxine, the reaction rates, or the rates of approach to a position of mean optical rotation, are identical within the limits of experimental error.

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