CCVI. THE CURATIVE ACTIVITY OF THE ANTINEURITIC VITAMIN OF RICE.

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IN 1926 Jansen and Donath [1926] published a method of isolating a crystalline vitamin from rice polishings capable of protecting bondols from polyneuritis in doses of 0.002 mg./day, and pigeons from loss of weight upon a diet of polished rice together with meat powder in doses of 0.01 mg./day. Since then it has seemed desirable to determine the activity of the crystals in terms of other methods of testing for vitamin B₁.

This paper records tests made in the Department of Biochemistry, Oxford, upon material prepared in Amsterdam.

EXPERIMENTAL.

Rice vitamin. The crystallised specimen of vitamin hydrochloride used for these experiments was prepared from rice polishings and had a melting point of 250°. For the purpose of the experiment it was dissolved in *N*/1000 HCl, to which 15 % ethyl alcohol was added, and kept in cold store while the tests were being carried out.

Tests. These were made by the curative method. The curative pigeon tests were carried out by the technique described by Kinnersley, Peters and Reader [1928] and the curative rat tests by a method to be shortly described by Kinnersley, Peters and Reader. The growth-promoting effects upon *Streptothrix corallinus* were examined by the original technique described by Orr-Ewing and Reader [1928].

It will be seen that the three methods of testing are not absolutely consistent, though they agree that the activity of the crystals lies between 0.005 and 0.009 mg. This is an activity higher than that yet obtained for torulin concentrates. It is to be noted that there is a rather considerable variation in the pigeon tests, all of which have been here included. The variation is larger than that to which we are accustomed with yeast preparations. We are unable at present to account for this; the tables indicate that it is not related

to the time of year; but the average value of 0.009 mg. by mouth and 0.007 mg. by injection is definitely more active than any yeast preparation which we have so far tested. It is also close to the value of 0.01 mg. obtained in the original protective tests upon pigeons by Jansen and Donath. The agreement for *S. corallinus* is so close as to suggest more strongly even than before that vitamin B₁ is the *Streptothrix*-growth-promoting factor.

Table I.

A. Pigeon tests.

Bird	Month of test	Weight g.	Days on diet	Dose mg.	Day cure	Day dose mg.
(a) Doses introduced by the mouth:						
213	July	220	20	0.044	9	0.005
104	"	217	19	"	5	0.008
166	"	240	17	"	3	0.015
123	June	217	26	"	5	0.009
174	July	250	25	"	4	0.011
210	June	300	—	"	11	0.004
153	"	208	13	0.029	9	0.003
130	"	224	20	0.087	8	0.011
149	"	280	20	0.029	2	0.015
						Average 0.009 mg.
(b) Doses injected into breast muscle in 0.5 cc. 0.9 % saline:						
103	Aug.	170	15	0.020	2	0.010
345	"	250	30	0.018	7	0.0025
390	Sept.	294	24	0.024	4	0.006
254	Oct.	202	27	"	2	0.012
411	"	258	27	"	6	0.004
365	"	260	27	"	4	0.006
						Average 0.007 mg.

B. Rat tests.

Rat	Dose mg.	Day cure	Day dose mg.
346	0.029	8	0.004
376	0.029	7	0.004
347	0.020	3	0.007
386	0.020	3.5	0.006
			0.005

C. *Streptothrix* tests.

This gave 0.006 mg. as the amount equivalent to 1 pigeon-day dose.

Summary

Method	Day dose mg.
Pigeon	0.009 (0.007 by injection)
Rat	0.005
<i>Streptothrix</i>	0.006
Average	0.007

Some estimate can be obtained from these figures of the amount required to promote growth in rats. Reader [1930] has shown that a young growing rat needs as a maximum 2.2 curative pigeon doses of vitamin B₁ daily to attain a maximal weight of 250 g. from 100 g.; less vitamin B₁ is needed to reach 100 g. It may be reckoned therefore that the maximum daily dose of

the crystals from rice polishings needed by the rat for growth to maximal weight does not exceed approximately 0.011 mg. daily¹.

Recently, Kinnersley and Peters [1930] stated that the Pauly reaction given by the purest vitamin B₁ preparations from yeast was a much less intense pink than that given by histidine, and that this seemed to constitute a difference between the Dutch preparation and their own. We have accordingly compared the intensity of the Pauly reaction given by one of the purest torulin preparations with an equivalent quantity of the rice vitamin side by side. The pinkness of the torulin preparation was rather greater than that of the rice vitamin; both gave a much less intense reaction than that of histidine. The difficulty therefore in the way of accepting the view that the same curative substance is present in the yeast preparation has disappeared. It must be concluded that the large doses required in the tests by Williams, Waterman and Gurin [1930] of 0.04 mg. *per diem* were a consequence of their method of testing.

SUMMARY.

Curative tests upon the Jansen and Donath rice vitamin crystals gave the following activity per day dose of vitamin: pigeon 0.007 mg. by injection, 0.009 mg. by mouth; rat 0.005 mg. Tests upon the micro-organism *S. corallinus* gave 0.006 mg. *per diem*.

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¹ (Note added at correction of proof.) The vitamin crystals also contain vitamin B₄ (see *Chem Ind.* (1930), **49**, *Proc. Biochem. Soc.* 990).