

CCLXXXVIII. MAINTENANCE NUTRITION IN THE PIGEON. THE EFFECT OF VITAMIN B₁

By CYRIL WILLIAM CARTER AND JOHN RICHARD O'BRIEN

From the Department of Biochemistry, Oxford

(Received 30 October 1937)

It has been known for many years that vitamin B₁ is essential both for protection from polyneuritis and for maintenance of normal weight in the pigeon. Much of the knowledge of the nutritional influence of vitamin B₁ has been derived from the use of extracts of yeast, rice polishings and cereals which are now recognized to contain other components of the vitamin B complex. In this laboratory the Peters concentrate, an acidic alcoholic eluate of norite charcoal [Kinnersley *et al.* 1933], has been extensively employed as a source of vitamin B₁ in the study of the vitamin B₁ requirements of the pigeon and the rat. The weight-restorative action attributable to the vitamin B₁ content of this concentrate is complicated by the presence of other factors of the vitamin B complex; vitamin B₆ needed by the pigeon and vitamin B₆ required by the rat [Carter *et al.* 1930, 1, 2; O'Brien, 1937]. The availability of crystalline vitamin B₁ has facilitated the investigation of the effect of this vitamin on the growth of the pigeon and allowed a better appreciation of the necessity of other factors for the maintenance of normal nutrition in the pigeon.

Waterman & Ammerman [1935] investigated the influence of a crystalline vitamin B₁ preparation upon the weight of pigeons on a diet of autoclaved whole wheat. Full weight recovery was not observed even when daily doses of 160 γ were administered. Perhaps the most remarkable feature of their work is the fact that the maximum weight response was only obtained with a daily vitamin B₁ intake of 80 γ , though this was only slightly greater than with 40 γ . This contrasts strikingly with the minimum dose required for the cure of acute polyneuritis (2.5 γ). Their use of a basal diet of autoclaved whole wheat raises the possibility of modification of the observed response by the presence of significant amounts of thermostable pigeon factors in the diet which, together with the stores of such factors held tenaciously by the pigeon [Carter & O'Brien, 1935], might exercise a marked effect upon the weight. During the past 18 months we have been engaged in experiments designed to elucidate the nature of these factors and in the course of the work we have gained considerable experience of the response of the pigeon to crystalline vitamin B₁, using a basal diet which we believe to be wholly deficient in components of the vitamin B complex. This paper records results which broadly confirm those of Waterman & Ammerman. Certain additional features of the response to vitamin B₁ are also described.

METHODS

The birds under test were maintained in single cages under conditions previously described. Two types of diet have been employed:

Diet R		Diet S	
Autoclaved polished rice	95%	Caseinogen	18%
McCullum salt mixture	5	Rice starch	68
Caseinogen*		Butter fat	9
		McCullum salt mixture	4
		Agar	1

* 2 g. given daily as separate supplement.

The synthetic diet, S, cooked as biscuits and then broken up into granular form, is readily consumed by the birds. The caseinogen of both diets was rendered as free as possible of flavin. Supplee *et al.* [1936] have pointed out that many commercial and "vitamin-free" caseinogen preparations retain traces of flavin. Different procedures have been adopted to remove such traces, and in agreement with others [Hogan & Richardson, 1932; Halliday & Evans, 1937] we have found that extraction with alcohol frees the caseinogen of most of its flavin. Four extractions are made, first with 97% alcohol, followed by two extractions with 50% alcohol and finally with 97% alcohol. Three samples of vitamin B₁ have been employed. One was prepared from baker's yeast following the method of Kinnersley *et al.* [1933]. The others, kindly placed at our disposal by Prof. Peters, were a natural crystalline product (Merck), and a synthetic preparation (I.G.). No difference in the activity of these crystals has been observed.

RESULTS

Fig. 1 illustrates two typical weight curves of pigeons receiving 40 γ daily of our crystalline vitamin B₁ following a preliminary period of depletion on the basal diet. Curve 1 shows a recovery which proceeds steadily until maximum weight is attained. The curve is not very dissimilar from that of a bird trans-

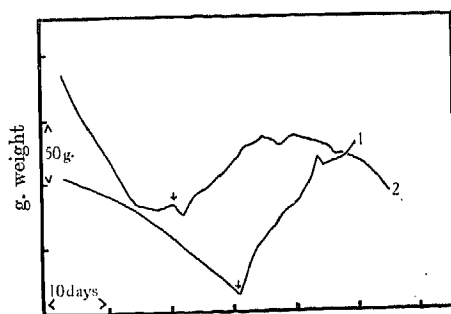


Fig. 1. 40 γ vitamin B₁ administered at arrow. For explanation see text.

ferred after the depletion period to a diet of whole wheat. A feature not clearly shown in these curves, but frequently observed, is the marked gain in weight recorded during the first 2 or 3 days of vitamin B₁ administration. This rise is accompanied by a correspondingly large food intake consequent on the improvement of appetite. Curve 2 shows a rise in weight after administration of vitamin B₁ to a plateau well below maximum weight and followed by a slow decline. The data reported in the present paper represent observations on 75 birds involving 89 individual experiments. Four factors have been studied in regard to their influence on the response to vitamin B₁, namely: (a) duration of the preliminary depletion period, (b) duration of vitamin B₁ administration, (c) magnitude of dose, (d) nature of the basal diet.

Duration of preliminary depletion

In a previous paper [Carter & O'Brien, 1935], in which the source of vitamin B₁ employed was the Peters concentrate, it was reported that in the case of birds whose weight did not fall below 70% of the maximum during the preliminary depletion, full recovery of weight occurred in 70% of cases. On the other hand, where the preliminary depletion weight fell to below 70% of the

maximum only 45% of birds recovered maximum weight on administration of the vitamin B₁ concentrate. Table I summarizes the position when crystalline

Table I. *Effect of vitamin B₁. S diet*

Bird	Max. wt. g.	Depletion wt. and % of max. wt.		Duration of depletion days	Gain in wt. on vitamin B ₁ g.	Duration on vitamin B ₁ admini- stration days
		g.	%			
583	293	244	83	12	36*	11
332	380	292	77	18	71	17
780	412	314	76	23	63	18
568	422	319	75	18	75	16
782	367	277	75	13	78*	13
44	500	371	74	31	59	18
568	425	306	72	12	80	19
782	380	270	71	18	74	8
530	371	264	71	24	75	10
837	500	350	70	29	120	13
28	380	268	70	21	62	11
345	344	242	70	29	120	19
507	396	276	69	26	91	12
605	469	314	67	25	62	9
251	431	292	67	28	94	19
336	430	290	67	26	131	22
446	360	237	66	31	87	15
446	351	238	68	24	71	10
447	334	220	66	29	114	10
410	500	330	66	31	92	23
899	391	268	68	28	62	8
780	428	281	65	34	108	20
410	453	290	64	32	70	18
28	382	247	64	31	89	17
536	432	273	63	30	-16	4
531	491	313	63	27	95	15
447	331	210	63	30	80	11
837	484	308	63	33	131	17
624	412	262	63	23	62	13
583	320	203	63	28	71	13
44	500	310	62	33	68	16
349	362	226	62	25	59	17
641	460	283	61	29	105	15
263	385	237	61	38	93*	15
846	436	266	61	31	87	20
348	400	247	61	29	49	4
881	500	300	60	31	98	17
881	452	272	60	29	87	13
750	444	267	60	37	125	29
750	403	243	60	43	92	17
533	399	237	59	22	53	14
899	412	245	59	35	127	27
700	452	264	58	37	73	17
539	386	227	59	30	96	19
595	364	215	59	16	85	15
263	412	238	57	51	75	12
535	393	226	57	31	62	10
120	386	218	56	31	79	9
264	412	227	55	35	108*	14
813	500	272	54	24	97	12
337	356	180	50	24	79	17
814	500	244	49	24	130*	23

* Subsequently declined in weight while receiving vitamin B₁.

vitamin B₁ in daily doses of 40 γ is employed. In the short depletion group comprising 14 birds 35% regained maximum weight, whereas in the long depletion group (42 birds) only 9% made a full weight recovery. Thus while the present

experiments indicate that a severe period of depletion militates against full weight recovery on administration of vitamin B₁ they also emphasize the relative ineffectiveness of the crystalline material as contrasted with the action of the Peters concentrate. This fact lends indirect support to the view that the latter is a source of an additional pigeon factor or factors which facilitate weight restoration.

Duration of vitamin B₁ administration

Table II records data for a further series in which the duration of vitamin B₁ administration was prolonged for periods up to 140 days. In the case of birds receiving diet S the initial recovery of weight in the majority of cases was checked

Table II. *Effect of vitamin B₁*

Bird	Max. wt. g.	Depletion wt. and %		Max. gain 10γ B ₁ g.	Days	Max. gain 20γ B ₁ g.	Days	Max. gain 30γ B ₁ g.	Days	Increase in wt. max. 40γ B ₁		Final total net gain g.	Total duration on vitamin B ₁ Days
		g.	%							g.	Days		
R diet													
84	436	313	71	—	—	14	42	—	—	47	12	42	94
860	380	271	71	—	—	23	76	—	—	21	13	39	90
77	435	313	71	—	—	35	69	—	—	32	13	53	90
770	465	328	70	—	—	35	75	—	—	39	13	57	93
83	413	282	68	—	—	48	77	—	—	26	3	52	86
857	419	282	67	—	—	21	66	—	—	14	8	21	87
514	456	307	67	13	4	16	13	56	19	20	9	75	56
439	452	302	66	-4	3	27	14	45	20	-1	3	61	41
597	450	292	65	—	—	36	11	32	16	—	—	49	35
523	382	250	65	—	—	2	7	53	22	5	6	56	37
600	418	274	65	-8	3	20	8	91	22	9	6	101	47
561	456	290	63	8	6	26	8	70	20	8	4	83	53
541	444	273	61	—	—	36	8	75	20	23	11	120	50
86	432	267	61	—	—	42	70	—	—	13	9	52	82
858	350	212	60	—	—	22	60	—	—	17	12	30	90
568	410	233	57	-10	4	35	9	27	14	—	—	41	33
S diet													
316	427	350	81	—	—	—	—	—	—	62*	29	29	62
153	500	408	81	—	—	—	—	—	—	64*	33	15	61
291	395	316	80	—	—	—	—	—	—	52*	14	17	39
306	380	284	74	—	—	68	10	—	—	12*	4	30	51
287	405	280	69	—	—	—	—	—	—	95*	20	63	38
85	490	334	68	26	9	26*	15	—	—	-42	53	-27	119
286	390	266	68	—	—	29*	6	—	—	-13	21	16	27
115	475	320	67	0	10	63*	24	—	—	-31	65	-24	135
122	500	338	67	-16	13	41*	13	—	—	—	—	-1	55
75	435	294	67	41	10	27*	34	—	—	—	—	-15	119
986	500	338	67	8	9	39*	29	—	—	-31	55	-20	120
766	430	283	66	6	5	45*	35	—	—	-32	52	-20	116
308	415	260	62	—	—	—	—	—	—	46*	18	27	28
47	500	311	62	9	10	78*	36	—	—	-73	67	-5	140
87	482	300	62	2	5	74*	26	—	—	-60	64	-13	129

* Subsequently declined in weight while receiving vitamin B₁.

and was followed by a period of slow decline. In 8 cases the final weight was actually lower than the weight at the commencement of vitamin B₁ administration. This failure to maintain weight is not due to some failure in absorption since injection of additional amounts of 20-40γ of the vitamin failed to bring about a renewed weight recovery. The phenomenon appears to be due to a slow continued depletion of other essential factors, since full recovery occurs when these are given as additional supplements. The phase of impending exhaustion of reserves of these factors usually begins to manifest itself within 20-30 days after commencement of vitamin B₁ administration, i.e. within 40-60 days from the beginning of the absolute depletion period. In the case of birds on diet R

the final period of decline was either absent altogether or much less conspicuous, even when the test period extended to 94 days. It would appear that diet R may have supplied traces of other factors which prevented decline within the period investigated. It might have been anticipated that a form of dermatitis similar to that described in the chick [Ringrose & Norris, 1931; Kline *et al.* 1934; Lepkovsky & Jukes, 1935] would have developed under the conditions of our experiments. Using young chicks on diet S supplemented by vitamin B₁ we had no difficulty in reproducing a typical dermatitis, but we have observed no analogous condition in the pigeon. It is uncertain whether the pigeon differs from the chick in an immunity to nutritional dermatitis or whether the fact that the pigeons used by us were adult prevented its appearance.

Magnitude of dose

Tables I and II indicate the responses to vitamin B₁ which have been observed with daily doses ranging from 10 to 40 γ . At a level of 10 γ the gains in weight are insignificant and in some cases there is even failure to maintain weight. When the dose is increased to 20 γ there is a partial response. This is always improved on increasing to an intake of 40 γ provided that the store of other factors has not become impaired. In certain cases where a level of 20 γ was maintained for a lengthy period, and where a decline in weight had set in, subsequent increase to 40 γ failed to bring about increase in weight or even to arrest the decline. In our experience a daily intake of 40 γ vitamin B₁ under our conditions represents the optimum level, and in the limited number of cases where the dose has been increased to 60 or even 80 γ the response has been insignificant or nil.

DISCUSSION

The results described, while in general agreement with those reported by Waterman & Ammerman [1935], amplify our knowledge of the response of the pigeon to crystalline vitamin B₁. The extent of this response is conditioned in part by the dose of vitamin B₁ and in part by the availability of other essential factors either stored or present in significant traces in the basal diet (e.g. diet R). Thus with a moderate preliminary depletion a complete restoration to maximum weight is sometimes observed, though in the majority of cases the response falls short of full recovery. If the preliminary depletion is severe, or if the period of vitamin B₁ administration is unduly prolonged, the initial gain in weight is small, and may be succeeded by a slow decline which is not arrested by increasing the dose of the vitamin. It can, however, be reversed by adding other factors. Only when the dose of vitamin B₁ is increased to 40 γ daily is the maximum effect observed.

SUMMARY

1. The response of pigeons, fed on a basal polished rice or synthetic diet, to daily doses of crystalline vitamin B₁ has been determined.
2. The response to vitamin B₁ after preliminary depletion is conditioned by the following factors:
 - (a) Duration of initial depletion period.
 - (b) Duration of vitamin B₁ administration.
 - (c) Magnitude of dose of vitamin B₁.
 - (d) Nature of basal diet.
3. Under the conditions described 40 γ represent the optimum daily dose of vitamin B₁ for maintenance nutrition, and doses rising to 80 γ do not permit greater weight restoration than that observed with the optimum.

4. No nutritional dermatitis comparable to that seen in the chick has been observed in the adult pigeon over periods extending to 140 days.

We desire to express our thanks to Prof. R. A. Peters for his interest and advice, and to Mr H. W. Kinnersley for valuable assistance. One of us (J. R. O'Brien) acknowledges the receipt of a part time grant from the Medical Research Council.

REFERENCES

- Carter, Kinnersley & Peters (1930, 1). *Biochem. J.* **24**, 1832.
— — — — (1930, 2). *Biochem. J.* **24**, 1844.
— & O'Brien (1935). *Biochem. J.* **29**, 2746.
Halliday & Evans (1937). *J. Nutrit.* **13**, 657.
Hogan & Richardson (1932). *Bull. Mo. Univ.* No. 241.
Kinnersley, O'Brien, Peters & Reader (1933). *Biochem. J.* **27**, 225.
Kline, Elvehjem, Keenan & Hart (1934). *J. biol. Chem.* **107**, 10.
Lepkovsky & Jukes (1935). *J. biol. Chem.* **3**, 119.
O'Brien (1937). Unpublished observation.
Ringrose & Norris (1931). *Science*, **71**, 643.
Supplee, Flanigan, Hanford & Ansbacher (1936). *J. biol. Chem.* **113**, 787.
Waterman & Ammerman (1935). *J. Nutrit.* **10**, 161.