Vitamin C Reserves of British Troops in England and Scotland during the Winter and Spring, 1941-2*

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The object of this work was to study the vitamin C status ('reserve') of the Army in relation to its bearing upon the prevalence of (a) gingivitis, and (b) scurvy or antecedent conditions and symptoms.

EXPERIMENTAL

Plan of investigation

The general plan of the investigation was drawn up by Major-General D. T. Richardson, C.B., M.C., K.H.S., Director of Hygiene, as follows:

Comparisons were instituted between recruits with less than I month's service, as indicating the condition of the civilian working-class population, and soldiers of over I year's service. Should a considerable difference be shown to exist, it might be argued that it would be unwise to accept a standard of nutrition in the Army which was demonstrably below that of civilians, especially as the troops were liable to face hardships and severe exercise, which, combined perhaps with a further lowering of nutritional standard, might be expected to lead to the risk of outbreaks of scurvy among those with low reserves.

It was decided that a hundred men should be examined in each Command, in winter and late spring, namely:

Western and Scottish Commands: soldiers who had been stationed in an industrial area for 6 months or over.

Northern and Southern Commands: soldiers who had been stationed in an agricultural area for 6 months or over.

Eastern Command: recruits who had come from civil employment in an industrial area.

South-eastern Command: recruits who had come from civil employment in an agricultural area.

Where possible, fifty soldiers were taken from a field ambulance which had a reception station, where the standard of messing was presumably good. The remaining fifty were to come from an infantry unit.

In the final arrangements the men who volunteered for the tests were as follows:

Northern Command: infantry and field ambulance men stationed in a country district in Norfolk.

* The investigation was undertaken upon instructions from the Director-General, Army Medical Services, Lieutenant-General Sir Alexander Hood, K.C.B., C.B.E., M.D., K.H.P. Permission to publish has been granted.

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Southern Command: infantry and field ambulance men in Hampshire.

Western Command: pioneers in a town, Crewe.

Scottish Command: infantry and R.A.M.C. personnel of a military hospital in Glasgow.

Eastern and South-eastern Commands: recruits, as arranged, in Essex and Aldershot districts, respectively; the latter had been in the Army less than 2 weeks.

Operational procedure for the test

General. The test selected was that described by Harris & Abbasy (1937) in which the vitamin C status is ascertained by giving large daily doses and analysing a sample of urine collected 3-5 or 6 hr. after administration, and noting the number of days necessary for saturation. When the vitamin is excreted in quantity, saturation is said to be reached or approximated to. The method has been used extensively, standardized and critically examined (see, for example, Harris, 1942, 1943; Nutrition Society, 1945).

The men received 0.75 g. synthetic vitamin C, L-assorbic acid, daily till saturated. The urine was collected and analysed; the volume was measured and the total amount excreted in the period calculated, since it is upon this—and not the concentration in the sample—that the test depends.

The work in each Command occupied a week, and the Commands were visited in the same order—South-eastern, Eastern, Southern, Western, Northern, Scottish. Starting on 2 December 1941, the winter tour ended on 17 January 1942.

To see how the troops had fared during late winter and spring, when green foods were scarce and potatoes had lost much of their vitamin C, a second tour was begun 4 months later on 13 April 1942 and ended on 24 May.

The tests were carried out by the author, who did all the chemical analyses. The preliminary work was done in the Royal Army Medical College, Hygiene Laboratory, then at Mytchett, under the late Colonel C. S. Ryles, O.B.E., and Major Stanley Elliott, O.B.E., T.D., F.R.I.C., to whom the author is much indebted for help and advice.

Period of retention. The period of retention, 3 hr., was sometimes found inconveniently long, and when shorter periods had to be taken, a rough correction was introduced on the assumption that the volume was a rectilinear function of the time, which is probably approximately true if the time is not too long.

In the spring series, a period of 2 hr. was rigorously adhered to. Though giving strictly comparable results as between the different Commands, this may have led to a systematic error on the high side, since the peak of excretion is about 4 hr. after dosing. It was considered that this was unlikely to be serious. In the winter, saturation was taken to be approached with an excretion of 50 mg. of the vitamin, but in the spring with 35 mg. for the shorter period.

Measurement of vitamin C. Pure L-ascorbic acid was used to adjust and check the concentration of the 2:6-dichlorophenolindophenol reagent at each station.

In the winter series the volume of each sample of urine was measured and 10.0 ml. were pipetted into a bottle containing 1.0 ml. glacial acetic acid. Thus for each sample a calculation had to be made to convert the concentration into the actual output of vitamin.

Furthermore, concentrated and turbid urines were troublesome to titrate. In the spring series, samples were made up to 0.5 or 1 l., using tap water, and this did not introduce any error. Thus each burette reading corresponded to a definite quantity of vitamin, and much time was saved. Since in all about 5000 samples were titrated, the reduction in labour was considerable, as compared with the standard technique. The end-point of the titrations was also better.

RESULTS AND DISCUSSION

Volume of urine passed

The volume of urine passed after 3 hr. retention varied in the winter series from o to 1090 ml., being for the South-eastern, Eastern, Southern, Western, Northern and Scottish Commands, respectively, 0-1060, 6-460, 80-1090, 50-750, 15-660 and 65-1025 ml. The average volumes for the first ten samples of each Command were 320, 90, 290, 360, 220 and 410 ml. The urines of low volume in the Eastern Command were mostly turbid and difficult to titrate. This was occasioned by the issue of an order, only discovered by the investigator after 2 days, that the men should not drink or have their dinner during the 6 hr. period. The men of the South-eastern and Eastern Commands were recruits and gave quite similar results, so it did not appear that the much smaller volume of urine had introduced any serious difference in the quantity of vitamin excreted. It was not possible to ascertain the range of urine volumes over which the time of retention is related to the quantity of vitamin excreted irrespective of the volume passed. It may have been wide, for on the 1st day soldiers W 59 and W 60 passed, respectively, 520 and 50 ml., yet the ascorbic acid excreted was 6.7 and 7.5 mg. Some men consistently passed little or no urine and had accordingly to be classed in the lowest group. They were advised to drink more.

Normal effect of dosing with vitamin C

Table 1 shows the excretion of vitamin C after each dose, either measured or calculated for a period of retention of 3 hr. These results are typical.

In some instances there was an immediate excretion of over 50 mg. in 3 hr., commonly accepted as denoting saturation. More usually for 2 or 3 days little or no change in output occurred. Thereafter, more or less suddenly, there was a rise. Unfortunately, the four doses allowed for in the winter series were too few in number to determine when saturation occurred in those men whose vitamin level was low, and these 112 men (19% of the total, omitting casualties) were divided up so that those showing an approach to saturation were classed as becoming saturated after a fifth dose, though they never received it. Those who, though still low, had almost doubled their initial excretion were classed as 5A. Several of the graphs suggest that these, taken as saturated after six doses, would in fact have been saturated after five. Those classed as 5B showed little or no response after five doses. For graphic representation, they were taken as saturated after seven doses, though some might have been saturated after six, possibly even after five, and some might have required

over seven. It is important to give doses adequate to classify the most resistant, as these are the ones likely to give trouble. The Scottish tests on personnel of the Royal Navy showed that at least five doses were necessary (McNee & Reid, 1942).

Table 1. Daily excretion of vitamin C by seventeen subjects receiving a daily dose of 0.75 g. of the vitamin

No. of subject	Г Г	2	3	4	Day of saturation
4	61	100		_	I
18	81	149		_	I
7	II	72	<i>—</i>		2
29	I	113		_	2
8	3	13	88	_	3
98	3	2	96	—	3
7x	4	21	230	—	3
10	2	4	34	127	4
97	2	8	22	71	4
94	2	3	9	176	4
79	6	3	4	16	5
61	4	5	7	18	5
21	3	7	14	12	5?
39	0	0	2	4	?
II	0	0	5	0	?
4 <i>x</i>	8	6	7	4	?
32	2	2	5	5	?

In the spring series, up to five doses were given to recruits and to the Southern Command men. Up to six doses were given in the Western, Northern and Scottish Commands; and the hard residue in the last should have had seven or more doses but, owing to vomiting, the doses were discontinued after six. The classifications 6, 6Aand 6B also 7, 7A and 7B were similarly used for indicating residues after five and six doses respectively, in the same way as already explained when four doses were given.

Effect of dosing beyond the accepted value for saturation, and some abnormal cases

Table 2 shows that, when a subject has come to the condition of heavy excretion, further dosing might result in no further increase, or even an appreciable decrease from the maximum, as if a state of tolerance were reached. The maximum excretion was 220-230 mg. in 3 hr.

There were occasional instances of a fall where a rise might have been expected nos. 45, 16 and 65—and of an almost steady pre-saturation level for a couple of days nos. 52, 5 and 79. Harris (1940, fig. 1) records, too, several cases of the rise to saturation followed by a diminishing output for 1 or 2 days before this again reached a maximum, but as his outputs are plotted on a logarithmic scale the phenomenon is less noticeable. Dr Harris has since informed me that this occurrence has been frequently noted (e.g. Harris, 1942, 1943).

Saturation tests in the six Commands

The tables giving the analytical results for each of the 1200 men for each day have been omitted. Their use lay in the comparisons with the conditions of the men's

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No. of	Ex	Day of				
subject	I	2	3	4	saturation	
39x	135	75		—	I	
94x	80	62		_	I	
98 <i>x</i>	184	92		- -	I	
15	82	83			I	
53	163	117	91		I	
41	2	62	224		2	
83	6	60	218	—	2	
60	5	111	78	53 *	2	
45	23	74	54	177	2	
95	35	26	194	<u> </u>	3	
16	31	4	72		3	
89	5	13	149	136	3	
52	4	39	36	129	4	
92	32	37	48	—	4	
5	5	6	43	31	5	
79	4	6	30	36	5	
65	2	7	25	2	5?	

Table 2. Daily excretion of vitamin C by seventeen subjects receiving a daily dose of 0.75 g. of the vitamin when dosing was continued beyond saturation; also some unusual occurrences

* Small volume (20 ml.) of urine excreted.

mouths as regards gingivitis. The results of the 5000 tests have been summarized in Table 3. The men whose vitamin C reserve was measured were also examined for gingivitis at the same time by the late Brigadier H. Stobie, Consulting Dental Surgeon to the Army, whose report showed that there was no connexion between incidence of gingivitis and vitamin C reserve. This is in agreement with the earlier results of Surgeon Rear-Admiral McNee and his co-workers, obtained on naval personnel (McNee & Reid, 1942).

In using the term 'reserves of vitamin C' no suggestion is implied that the amount present may be without function save as a reserve. Again, some prefer to write of status or level, since vitamin C is not stored in special parts of the body to any considerable degree and it is thought that the test indicates rather the 'degree of saturation' of the tissues.

The data of Table 3 may be grasped more readily by inspection of Figs. 1-3, which show, respectively, the results for men from country and industrial districts, and the total for recruits as against total for soldiers. In these figures a clearer picture is given by plotting not, as heretofore customary, the number saturated after each dose, but the total number that had reached saturation at that period (Atkins, 1943). It may be seen that in each of Figs. 1-3 the full lines, denoting recruits, lie above the broken lines, denoting soldiers, for the same season.

For industrial districts, Fig. 2, and the totals, Fig. 3, the recruits, even in spring, are superior to the soldiers both during winter and spring. Such differences amount to one or two daily doses and even to three for those with least reserve. Furthermore, though almost all the recruits became saturated, a percentage, increasing during winter, of the soldiers failed to saturate. This is shown in Fig. 4. The importance of this

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Table 3. Number of men in South-eastern (SE), Eastern (E), Southern (S), Wes	stern
(W), Northern (N) and Scottish (SC) Commands becoming saturated after a	loses
as tabulated, also percentage of total for recruits (civilians, SE and E)	(<i>C</i>),
soldiers (M) and total (T) in winter series (WR) and spring series (SP)	

-	s	E	H	2	s		W	•	N	[so	2	C	2	Ν	1	Г	
Cate- gory #	ŴR	SP	ŴŔ	SP	ŴŔ	SP	WR	SP	ŴR	SP	WR	SP	WR	SP	WR	SP	WR	SP
I	9	0	0	0	2	0	0	0	0	0	0	٥	4.2	o	0.2	0	1.9	0
2	31	10	27	10	20	4	7	2	14	5	2	0	29.0	10.4	11.3	2.9	17.4	5-4
3	32	52	47	39	34	14	36	14	43	9	28	4	40	47 ·1	36.3	10.8	37.4	23.0
4	24	30	15	29	28	38	25	19	24	35	27	30	19	30.6	26.8	32.0	24'3	31.2
5	4	3	6	15	9	17	12	31	9	30	20	28	5	9.3	12.9	27.8	10.5	21.0
5A	ò	ō	3	ō	5	ò	10	ం	6	0	4	o	1.2	0	6.2	0	4·8	0
5 B	I	1	Ĩ	0	3	o	8	0	I	o	10	0	1	0	5.2	0	4.0	0
6	0	0	0	3	õ	11	0	11	0	9	0	12	0	1.2	ο	11.3	0	8
6 <i>A</i>	0	0	o	ō	0	4	0	0	0	I	0	0	0	0	ο	1.3	0	0.0
6 <i>B</i>	0	o	о	1	0	7	0	0	ο	1	0	r	0	I	0	2.3	ο	1.0
7	0	0	0	0	0	ò	0	4	` о	4	0	9	0	0	0	4.4	0	2.9
7A	0	0	о	0	o	0	o	5	o	2	0	3	0	o	0	2.6	0	1.8
7 <i>B</i>	0	0	0	o	0	о	o	6	0	4	0	7	0	0	ο	4.4	0	2.9
Cas ua ltic	es o	5	I	3	I	4	5	4	4	5	9†	I			<u> </u>			
Total	101	101	100	100	102	99	103	96	101	105	100	95	100	99.9	100	99·8	100	99.9

• See text, p. 277.

[†] One man, saturated on 2nd day, had been in the country up to the previous week; he has now been included in the casualties.



Fig. 1. Time taken to reach saturation by recruits and soldiers from and in the country, in different Commands, receiving a daily dose of 0.75 g. synthetic vitamin C. The graphs show the total number of men who had reached saturation up to the given day inclusive. WR, winter series; SP, spring series; _____, South-eastern Command, recruits; ____, Southern Command, soldiers; _____, Northern Command, soldiers.



Fig. 2. Time taken to reach saturation by recruits and soldiers from industrial areas, in different Commands, receiving a daily dose of 0.75 g. synthetic vitamin C. The graphs show the total number of men who had reached saturation up to the given day inclusive. WR, winter series; SP, spring series; ______, Eastern Command, recruits; ----, Western Command, soldiers; -----, Scottish Command, soldiers.



Fig. 3. Time taken to reach saturation by recruits (total examined 200) and by soldiers (total examined 400) from all Commands, receiving a daily dose of 0.75 g. synthetic vitamin C. The graphs show the total number of men who had reached saturation up to the given day inclusive. WR, winter series; SP, spring series; -----, recruits; ----, soldiers.

finding lies in the fact that it is this percentage that indicates the potential victims of scurvy and its premonitory symptoms when more severe conditions are encountered; such a percentage, negligible in civilians, is seen to be large in the Army in spring.

In the Southern and Northern Commands, R.A.M.C. and Infantry were used in the trials, all being in the country. In the Southern Command, the Infantry were rather better than the R.A.M.C., but the Northern R.A.M.C. were close to the Southern



Fig. 4. Number of recruits and soldiers in town and country areas in different Commands remaining unsaturated after five or six doses of 0.75 g. synthetic vitamin C. WR, winter series; SP, spring series; SE, South-eastern Command (recruits, country); E, Eastern Command (recruits, town); S, Southern Command (soldiers, country); W, Western Command (soldiers, town); N, Northern Command (soldiers, country); SC, Scottish Command (soldiers, town); ———, six doses; ----, five doses.

Infantry and tailed off less. The Northern Infantry were decidedly the lowest. They were a normal infantry unit, and probably had more severe training than the Southern, which was a Defence Company. Both R.A.M.C. units were Field Ambulances, and did a considerable amount of marching.

Storage of vitamin C in the body

An attempt was made to get some information on storage within the limits of the saturation-test routine.

In the spring series three men were dosed to saturation and dosed again after omitting the dose for 1 or 2 days. A had no dose on the 3rd and 4th days; one on the 5th saturated him fully. B, with similar omissions, became nearly saturated on the 5th day, and on the 6th day excretion fell to 10 mg. with no dose. D had no dose on the 4th day, but became saturated again when dosed on the 5th. Thus, the excess of vitamin appeared to be quickly eliminated and destroyed, but a saturated man not dosed for 2 days could be restored to saturation by one dose.

It was possible, however, to compare the results for thirty-seven men of an industrial

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area, Western Command, who happened to be dosed in both the winter and spring series, respectively from 29 December 1941 to 1 January 1942 and 5 May to 9 May 1942, and also for two men in Southern Command (country) and one man in Scottish Command (city). No change was shown by 30 % in the day upon which they became saturated; 35 % were 1 day later in spring; the remainder were earlier by 1 day (10 %)



Fig. 5. Time taken to reach saturation in the spring series by recruits and soldiers of Eastern and Western Commands, receiving daily doses of 0.75 g. synthetic vitamin C. The graphs show the total number of men who had reached saturation up to the given day inclusive. - (above), Eastern Command (recruits); -------- (below), Western Command (soldiers); ...-..., Western Command, soldiers saturated 4 months previously; ----, Western Command, soldiers who had received four doses 4 months previously; -----, Western Command, soldiers not dosed before.

or 2 days (5 %) or later by 2 days (12.5 %) or 3 days (7.5 %). There was thus no great difference between these and men not previously saturated, but Fig. 5 shows that the recruits from industrial districts were somewhat superior even to those soldiers who had been dosed to saturation in the winter series 4 months before.

The recruits were markedly superior to the Western Command sample as a whole, and a small but definite difference could be seen to exist between the men previously dosed to saturation and those who had received four doses 4 months previously, though not all of them had become saturated. The lowest in reserves were the Western Command men who had not been dosed in the winter series. Of these, 33 % required over five doses to saturate, as against 28 % for the Western as a whole; 22 % of those dosed before required five doses; but only 4 % of those dosed and saturated 4 months previously required five doses, and only 4 % of the Eastern Command recruits.

Thus, some effect of saturation 4 months previously still remained detectable; alternatively, one might conclude that the metabolism of the men who became saturated easily was in each series detectable by the test, and that they stood out in each series because they used the vitamin more efficiently, or destroyed less of it in the body.

Effect of dosing before and after a meal

In the winter series in Glasgow, fifty men were hospital orderlies, fifty from an infantry regiment. The R.A.M.C. were in sections, A and B, merely for A.R.P. duties, they messed and worked together. The results, however, were very different as between the sections, B reaching saturation sooner than A. This at first appeared to be a severe blow to one's confidence in the method. Inquiries failed to reveal any difference between the sections, such as weight or occupation. The results were reported to the Director of Hygiene at the time without any explanation being possible.



Fig. 6. Time taken to reach saturation by soldiers receiving daily doses of 0.75 g. synthetic vitamin C before (twenty-two men) or after (twenty-one men) breakfast. The graphs show the number of men who had reached saturation up to the given day inclusive. ————, section A men, dosed before breakfast; – – –, section B men, dosed after breakfast.

The same hospital was visited 4 months later, and further inquiries elicited the fact that the section A men had paraded at 7.30 a.m. before breakfast, as they had been busy attending to patients. They were dosed at once. The Infantry then arrived, and section B men had breakfast and were dosed afterwards. This routine was followed on subsequent days in order to have an exact repetition from day to day. Thus the vitamin seemed to suffer less destruction and to be better utilized when taken after food. As, however, it appeared unwise to draw a conclusion from a single experiment although fifty men were used, the tabulated results were submitted to statistical analysis and the results were published (Atkins & Fisher, 1944), since the question was not included in the original object of the work.

Four doses were given. As before, those saturated on the 5th day were shown as such, with 5A denoting those likely to be saturated after six doses, and 5B those so low after four doses that no prediction could be made; the latter are plotted as for seven doses. The results are shown in Fig. 6.

To ascertain the probability of the distribution found being due to chance, of the forty-three cases (there were seven casualties out of the fifty) in the two groups to be compared, a line may be drawn between the nineteen saturated in the first 3 days and

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the twenty-four others taking longer. One finds in the first group four A and fifteen B, in the second group eighteen A and six B. The chance of getting so large a discrepancy, if the numbers in A and B were really proportional, is about 1:1867. Thus there is no doubt of the statistical significance of the difference observed. The only ascertainable difference between the two groups was that men of section A, who took longer to saturate, had their vitamin C on an empty stomach.

Pathological effects of the doses

Under the term pathological effect one may consider those cases in which the dosing caused, or was associated with, stomach troubles.

(a) Winter series. S 21 said he always vomited after taking tablets, and he did so on this occasion. No more were given. N 101 was much surprised at the effect, almost immediate vomiting. The thirty tablets in a cup of tap water at Mytchett gave a slightly acid taste, the reaction being about pH 3.5.

(b) Spring series. E38 refused to take the tablets on the 2nd day, saying that the taste still persisted since he had chewed them on the 1st day. N 56 reported that he often suffered from indigestion and used to take kaolin before meals. After four doses he excreted 25 mg., and no further vitamin was given as he complained of a renewal of his indigestion. He was rated as saturated on the 5th day. N 86 complained of heartburn and indigestion, which he did not remember ever having had before. The dosing was stopped. He excreted 12 mg. after the fourth dose, indicating probable saturation on the 6th day, possibly on the 5th. SC 12 vomited after the tablets on the 5th day, when he excreted 25 mg. He made no complaint, and vomited again on the 6th day, but excreted 42 mg. so must have absorbed a considerable amount of each dose, as he was finally saturated, 35 mg. for a 2 hr. period being taken as the criterion. SC 34 vomited on the 6th day and excreted 7 mg., so was classed 7 B. SC 59 also vomited on the 6th day.

On account of the vomiting no further doses beyond six were given, though patients with scurvy may receive up to ten or twelve. Thus McNee & Reid (1942) found that a case of scurvy took eleven 0.7 g. doses, 7.7 g. in all, for saturation and the condition of the gums remained unchanged after 14 days, though the circumfollicular lesions were fading between the fourth and fifth dose.

The thirty-two who had six doses seemed much puzzled as to why they should be so much behind and said they ate potatoes and other vegetables like the rest.

SUMMARY

1. The saturation test of Harris & Abbasy (1937) was applied to recruits, as indicating the condition of civilians, and to soldiers of 1 year's service and upwards, during the winter and late spring 1941-2.

2. The recruits showed a superior vitamin C status compared with the soldiers, so that even in spring their 'reserves' were greater than those of the soldiers as a whole had been 4 months previously in winter.

3. The recruits from the country were slightly superior to those from industrial

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areas, and soldiers from country districts had definitely greater reserves than those from industrial areas.

4. These differences become striking when one considers those with the lowest reserves, not saturated even after receiving five or six doses of 0.75 g. L-ascorbic acid. Whereas this was 1 % for the recruits, the soldiers of Scottish Command (Glasgow) showed in spring 34 % not saturated after five doses and 21 % not saturated after six doses. Though no symptoms of approaching scurvy were reported and the men all seemed well, yet exposure to hardships, severe exercise and restricted food might have been expected to evoke scurvy sooner in these men than in the recruits.

5. It would appear unwise to accept as satisfactory a reserve of vitamin C considerably lower than that of the young civilian working-class population. Harris (1943) has shown that the large majority of men receiving the 30 mg. of vitamin C, recommended as the 'standard requirement' of the League of Nations, became saturated in 2 or 3 days; so the soldiers serving in Great Britain had received less during the winter, since large numbers of them were unsaturated after even five or six doses.

6. The evidence available pointed to the desirability of increasing the vitamin C supply of the troops in winter, especially as it was found that forty men saturated in January were only slightly above those not previously dosed, when compared 4 months later in May. Those men previously saturated left a very small unsaturated residue after five doses, namely 4% as against 33% for those not dosed before.

7. One test with forty-three men showed that saturation was reached more rapidly in those who had the vitamin dose after breakfast rather than before it.

8. Administration of the vitamin tablets in water resulted in vomiting in two soldiers in winter and two in spring, when also there were two cases of stomach trouble attributed to the tablets.

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