THE INFLUENCE OF CERTAIN DIETARY SUPPLE-MENTS ON THE NUTRITION OF THE AFRICAN NATIVE. I.

BY FRANCIS CHARLES KELLY AND JOHN MCASKILL HENDERSON.

(Kenya Medical Service and Rowett Research Institute, Aberdeen.)

(With 3 Figures.)

DURING the last few years a great mass of work has been carried out in connection with the necessity of ensuring an adequate supply of accessory factors and mineral salts in the diets of growing subjects, whether it be children, or experimental animals.

In comparison, little work has been done on full-grown animals; and the requirements of adult man have been particularly neglected.

Accordingly when an opportunity presented itself of using full-grown men as experimental subjects for an investigation of this nature, it was thought well to take advantage of it, particularly since, in the circumstances, some dietary deficiency seemed not improbable.

SUBJECTS.

The 42 subjects used were selected from some 200 male natives serving long-term sentences in Nairobi Prison, Kenya Colony. The selection was carried out with the following objects in view: (1) that the men should all belong to the same tribe or to nearly related tribes (Kikuyu); (2) that they should all be full-grown, free from obvious disease and of average physique; (3) that their occupations should be comparable.

The 42 men thus selected were divided into five groups, there being 14 men in the first group and 7 in each of the others. The constitution of each group was as nearly as possible the same with respect to the following points: (1) the average weight in each group was approximately equal, *i.e.* I = 124.9, II = 125.1, III = 123.6, IV = 124.1, V = 123.6 lb.; (2) as regards their occupations, all were either carpenters or tailors and the proportion of the former to the latter in each group was 2 to 5; (3) the groups were evenly balanced as regards the numbers in each who showed positive Wassermann reactions.

HOUSING AND SUPERVISION.

The men were under the usual strict prison supervision and disciplinary control throughout the experiment. Through the courtesy, however, of the prison authorities the men attended at the prison hospital for the issue of

supplements, collection of excreta, etc. Their daily rations were served from a kitchen specially constructed for the purpose.

PRELIMINARY TREATMENT.

The faeces of the 42 prisoners were examined for protozoal and helminthic infections with a view to having the positive cases deparasitised. It was assumed that the subject was free from parasites on the strength of negative results on three successive examinations. The value of this assumption can be gauged from Callanan's (1927) figures.

As already indicated the Wassermann reaction was determined for each subject.

Differential blood counts and blood examinations for malarial parasites were carried out and anti-malarial treatment administered where necessary.

BASAL DIET.

The basal diet was the "long-term" diet in use at the prison. It consisted of the following:

Beans	•••		•••		0.6 oz. daily
Beef	•••	•••	•••	•••	3.5 "
Whole m	aize	•••	•••	•••	24.0 ,,
Potatoes	•••		•••	•••	8·0 "
\mathbf{Salt}	•••	•••	•••	•••	0.25 ,,
Ghee		•••	•••	•••	0.5 "
Lemons	•••	•••	•••	•••	2·0 per week

On analysis it showed the following composition: moisture, 73 per cent.; total protein, 116.5 grm.; total carbohydrate (by difference), 626 grm.; total fat, 53.4 grm.; energy, 3450 calories; fibre, 12 grm.; ash, 20.7 grm.; calcium, 0.30 grm. Ca; phosphorus, 2.36 grm. P (Kelly and Henderson, 1927).

Supplements.

The groups and additions were arranged as follows:

Group	No. of men	Ration		Daily additions
Ι	14	Basal	+	No additions
II	7	Basal	+	125 c.c. soluble mineral mixture + 15 c.c. cod-liver oil
111	7	Basal	+	15 c.c. cod-liver oil
IV	7	Basal	+	125 c.c. soluble mineral mixture
v	7	Basal	+	15 c.c. olive oil $+0.001$ grm. iodine as KI solution

MINERAL MIXTURE.

Our object was to devise a soluble mineral mixture of approximately the same composition as the ash of cows' milk and to feed daily an amount of it equivalent to the ash in 1 pint of milk. We found it no easy matter to devise a mixture which at one and the same time supplies the appropriate minerals in an easily procurable and soluble form and which is reasonably palatable.

The following, however, was found to meet the requirements. The figures represent one daily dose as fed to Groups II and IV. They are compared with the ash of an average sample of cows' milk.

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Equivalent to	Average amount in ash of 1 pint cows' milk
$0.39 \operatorname{Na_2O}$	0.35 Na2O
$\begin{array}{c} 0.44 \text{ Cl} \\ 0.86 \text{ K}_2 \text{O} \\ 1.21 \text{ Pc} \end{array}$	0·59 Cl 1·04 KgO
$1.31 P_2O_5$ 0.11 MgO 0.954 CaO $0.012 Fe_2O_3$	1·12 P ₂ O ₅ 0·11 MgO 0·944 CaO 0·012 Fe ₂ O ₃
	$\begin{array}{c} 0.39 \text{ Na}_2\text{O} \\ 0.44 \text{ Cl} \\ 0.86 \text{ K}_2\text{O} \\ 1.31 \text{ P}_2\text{O}_5 \\ 0.11 \text{ MgO} \\ 0.954 \text{ CaO} \end{array}$

The above amounts of calcium lactate and iron citrate were made up in one solution with water to 100 c.c. The sodium chloride, potassium di-hydrogen phosphate and the magnesium sulphate were together dissolved and made up to 25 c.c., thus making the total dose per man per day 125 c.c.

The olive oil and iodine were added as the outcome of certain work by one of us (Kelly, 1925), in which the effect of small doses of iodine upon the growth of young pigs had been investigated.

EXPERIMENTAL METHOD.

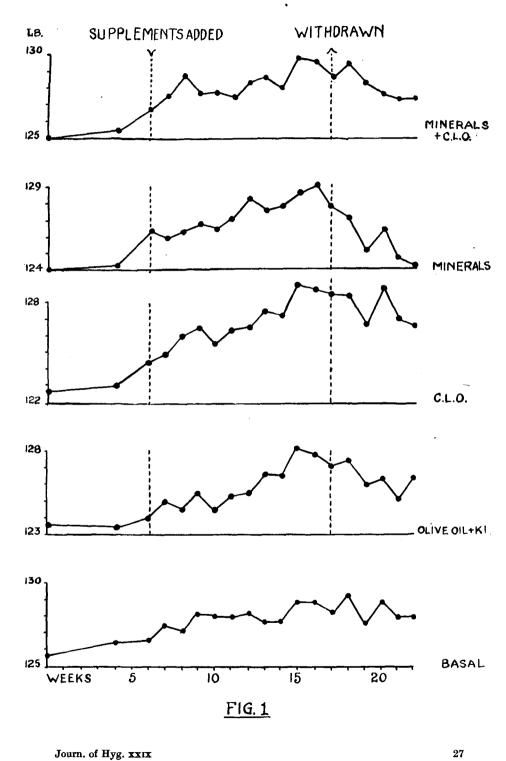
(1) Previous to the addition of supplements to the diet the prisoners were under observation for 6 weeks. The supplements were then given to the groups as indicated above. At the end of a further 11 weeks the supplements were withdrawn and the observations were carried on for a post-period of 5 weeks. These three periods will be referred to respectively as the pre-period, the supplement-period and the post-period.

(2) The prisoners were weighed on three occasions in the pre-period and, thereafter, weekly.

(3) The total blood calcium was determined (Kramer and Tisdall) in the pre-period, towards the end of the supplement-period, and in the last week of the post-period. Objection has lately been made against this method (Pincussen and Schimmelpfeng, 1927) that the time allowed for precipitation is too short. As a matter of routine we have always allowed the precipitation to take place overnight. The error in our duplicate analyses is about ± 3 per cent. We may accordingly regard a rise or fall of 0.5 mg. Ca as significant.

(4) The calcium balances of 5 men (1 from each group) were determined simultaneously. In all, 20 men (4 representative men from each group) were thus examined over the last 4 weeks of the supplement-period. The collection period in each case was a continuous one of 96 hours. During this time the prisoners were under constant observation in the prison hospital, but had their daily exercise as usual. Each man was provided with two covered pails containing a small quantity of toluene to prevent decomposition of excreta. The faeces and urine were thus collected separately. They were analysed at the end of each 48 hours. The calcium was determined by precipitation with oxalate and titration with permanganate.

(5) Throughout the experiment stools were examined for parasites and blood for malaria. These conditions were treated where necessary.





RESULTS.

Weight curves.

It is considered unnecessary to reproduce the individual weight curves. Average curves, however, for each group have been drawn (Fig. 1). In each group the individual curves correspond closely to the composite curve shown. The curves, however, for individuals 1871 (basal group) and 1590 (cod-liver

Table .	I.
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Blood calcium mg. per 100 c.c.			Gain or loss in weight (lb.)			
		Supple-	er 100 c.c.	Supplement-	Post-period minus	
No. of subject	Pre- period	ment- period	Post- period	minus Pre-period	Supplement- period	Intestinal parasites
Basal group	T	1	1	1	T	1
217	9.9	9.8	9.7	- 4.07	+ 0.06	_
1148	9.7	10.8	10.7	+ 1.78	+ 1.89	T - T
2932	8.6	9.8	9.0	+ 4.28	+ 2.24	T and An –
909	8.7	10.2	9.0	+ 2.63	+ 2.27	An –
1866	9·4	9.8	8.9	+ 2.58	+ 3.59	T –
989	9.8	9.2	9.3	+ 0.77	-1.62	
1871	8.4	9.8	9.5	+20.34	+ 0.46	As - T -
950	10.7	10.5	10.3	+ 3.30	-0.97	-
1255	8.4	7.8	9.4	+ 0.87	- 1.50	-
1384	11.0	10.3	10.2	+ 3.14	- 0.34	T - As
3963	10.1	9.9	10.2	+ 0.48	+ 0.72	
2188	10.2	9.2	9.3	+ 3.08	+ 0.75	$-\mathbf{T}$ -
2189	10.0	10.4	10.7	+ 5.22	+ 0.05	- An -
1147	9.0	8.8	10.0	+ 0.91	+ 0.76	-
Minerals and		U 1				_
831	9 •6	10.3	9.9	- 0.41	- 1.49	<u> </u>
1589	9.7	10-1	9-1	+ 4.65	- 0.52	T -
1263	8.0	10.1	9·4	+ 6.15	+ 0.57	$\mathbf{T} - \mathbf{T}$
1844	10.0	10.0	10.2	+ 1.41	- 1.32	Τ –
390	10.2	10.7	10.2	+ 3.99	- 1.24	-
3818	7.6	8.4	8.8	+ 1.73	+ 0.97	-
2740	9 ·6	10.2	10.1	+ 1.12	- 0.45	An –
Cod-liver oil	group					
1251	8.4	9.6	9.2	+ 2.94	+ 5.33	\mathbf{T} –
1533	8.9	10.2	9·4	+ 4.85	- 1.42	T -
1090	8.6	9.5	8.7	+ 4.09	+ 0.86	An –
4298	8.0	10-1	9·4	+ 5.15	- 1.03	T and As –
949	9·4	10.0	9.8	+ 0.72	- 0.27	_
3468	8.9	10.0	9.5	+ 3.51	- 0.98	An and T – T –
1590	9.3	10-1	9.1	+ 8.37	+10.01	Т –
Minerals grou	up					
1166	8.6	9.8	.9.3	+ 0.24	- 0.42	T and An –
947	9.3	9.9	9.7	+ 3.02	-6.72	_
1510	7.6	8.9	9.5	+ 0.42	- 2.04	An – T –
1867	10.1	10.6	9.7	+ 0.34	-5.54	- T -
4021	8.8	9.5	9.5	+ 1.08	+ 0.39	An –
1636	9.7	10.7	10.0	+ 4.48	+ 1.81	An –
2169	8.8	9.7	9.7	+ 8.04	- 1.14	-
Olive oil and	iodine gro	up				
1527	8.7	9.2	9.7	+ 3.53	+ 2.10	Т –
2830	9.2	9.9	9.8	+ 5.14	+ 1.21	-T-
2025	8·4	9.4	10.4	+ 6.94	-2.02	т-
948	$\tilde{9}\cdot\tilde{5}$	8.5	9.4	- 1.16	+ 0.56	_
2739	10-1	10.1	10.1	+ 2.60	+ 0.43	An - T -
1653	7.3	7.9	7.0	+ 1.31	+ 3.76	T –
3929	9.4	8.9	10.0	- 2.50	-5.35	T -

oil group) were omitted when determining the composite curves of these two groups on account of their marked deviation from the group-mean.

Blood calcium.

Table I, above, shows the individual variations in blood calcium and in weight for each period. Data on intestinal parasites are included. A minus sign indicates complete freedom from infection. T, An, As, indicate respectively infection with Taenia, Ankylostoma and Ascaris. Thus in subject 3468, "An and T - T -" indicates that on first examination Ankylostoma and Taenia were both found. On subsequent examination the stool was negative. Later, however, Taenia again appeared, but on the last examination the subject was found free from infection.

The figures for gain or loss in weight per period were arrived at by determining the average weight for each individual throughout each period. In the first weight column the pre-period average was subtracted from that of the supplement-period, a plus sign indicating a rise in weight in the supplement-period and *vice versa*. In the second column the average weight for the supplement-period was subtracted from that of the post-period, a plus sign again indicating a rise in the post-period and a minus sign a fall.

Calcium balances.

The following table gives details of the calcium balances in the 20 cases investigated. The figures are expressed in grammes Ca.

No.	Group and calcium intake per 96 hours	Total calcium urine grm.	Total calcium faeces grm.	Total calcium excreted grm.	Total balance per 96 hours grm.	Average balance of group per 96 hours grm.	Average balance of group per 24 hours grm.
989 2932 909 217	Basal 1·2000 grm.	0·0900 0·0508 0·0930 0·0391	1.6870 1.4000 0.6046 0.6872	1.7770 1.4508 0.6976 0.7263	$ \begin{array}{c} -0.5770 \\ -0.2508 \\ +0.5024 \\ +0.4737 \end{array} $	+0.0371	+0.0093
1263 390 2740 831	Minerals plus cod-liver oil 3·9200 grm.	0·0510 0·2417 0·1506 0·3108	1·2324 3·4526 3·3958 2·9461	1·2834 3·6943 3·5464 3·2569	$\begin{array}{c} +2.6366\\ +0.2257\\ +0.3736\\ +0.6631 \end{array}$	+0.9747	+0.2437
3468 1590 1533 949	Cod-liver oil 1·2000 grm.	0·0540 0·0854 0·2158 0·0541	1·3665 1·1854 1·9318 0·9655	1·4205 1·2709 2·1476 1·0196	$\left.\begin{array}{c} -\ 0.2205 \\ -\ 0.0708 \\ -\ 0.9476 \\ +\ 0.1804 \end{array}\right\}$	-0.2646	-0.0661
4021 1636 947 1867	Minerals 3·9200 grm.	0·1901 0·0503 0·1352 0·2140	5·5786 3·7696 1·5274 3·1601	5·7687 3·8199 1·6626 3·3741	$ \begin{array}{c} -1.8487 \\ +0.1001 \\ +2.2574 \\ +0.5459 \end{array} $	+0.2637	+0.0659
3929 2739 2830 948	Olive oil plus iodine 1·2000 grm.	0·1382 0·0478 0·0789 0·0728	2·0924 1·3010 0·8435 1·4633	2·2306 1·3488 0·9224 1·5361	$\begin{array}{c} -1.0306\\ -0.1488\\ +0.2776\\ -0.3361 \end{array}$	- 0.3095	-0.0774

Table II.

DISCUSSION OF RESULTS.

In considering the weight curves it is necessary to bear in mind that the subjects under the experiment were adult men. Further, they were, during the experiment, and had been for a considerable time previously, on a basal diet which was adequate in energy content (Kelly and Henderson, 1927).

It will be seen from the averaged group curves that while the weight in the basal group is well maintained, and even increases, the weight in the other groups shows a more marked increase. This is further evident from a consideration of the following table, which shows the average increase in weight per man in the respective groups.

T	~1	հไո	γI	Т	т	
T (aı	JIG	; T	T	T	•

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Group	Average gain in weight during supplement-period	Average gain in weight during post-period as compared with weight in supplement-period
Basal	+ 1.92 lb.	+ 0·49 lb.
Minerals and cod-liver oil	+2.66,	-0.49 ,
Cod-liver oil	+2.55 ,,	+0.41
Minerals	+2.51 ,	- 1.95 "
Olive oil and iodine	+2.27 "	+0.10 "

A marked feature of the curves is the drop in weight which follows the withdrawal of the supplements. This is most noticeable in the mineral group. It is less evident in the groups with cod-liver oil, a fact which may be accounted for by the storage of vitamin.

It has suggested itself that it may be possible to interpret certain variations in weight in terms of the minimum temperature curve for the week preceding any particular weighing. This temperature factor is being further investigated; but since it is common to all our groups it cannot vitiate our present results.

Blood calcium.

The blood calcium estimations in the pre-period show that the average (9.2 mg. per 100 c.c. serum) corresponds with the European low normal figure. There are, further, a considerable number definitely sub-normal, such values as 7.3 mg. and 7.6 mg. being encountered.

Towards the end of the supplement-period, while in the basal and in the olive oil and iodine groups only 50 per cent. showed a rise in blood calcium, it is noteworthy that in the other groups all subjects (with one exception, which remained steady) showed a definite rise. This is perhaps most striking in the cod-liver oil group.

It has generally been considered very difficult to raise the blood calcium by oral administration of calcium salts except in such cases as showed initially a definitely sub-normal value (Denis and Minot, 1920). Robinson and Huffman (1926) on the other hand obtained a very strong suggestion that in feeding calves on a calcium-rich ration the blood calcium level was raised. Further, Bauer and Ropes (1926) have recently shown that the blood calcium

level shows a marked though transitory rise (14 per cent. and 28 per cent. above normal) after the ingestion of 5 and 10 grm. doses of calcium lactate. Kahn and Roe (1926) have recorded even more striking rises, *e.g.* as high as 108 per cent. increase after doses of 5 grm. calcium lactate.

It is highly improbable that the rises obtained in our cases were of this latter transient type. Firstly, the rise takes place in the cod-liver oil group where no extra calcium was given. Secondly, as will be seen from Fig. 2, there is a definite positive correlation between rise in blood calcium and increase in weight in the supplement-period. Apart from the general correlation $(0.5)^1$ it is noteworthy that only one subject (No. 831) who showed a rise in blood calcium during the supplement-period showed a fall in weight. This fall only

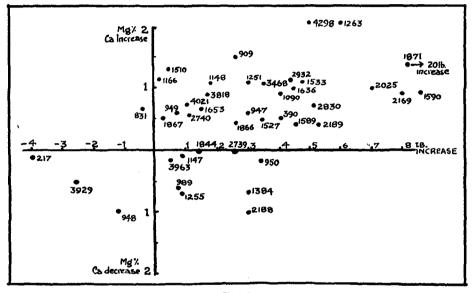


Fig. 2.

amounted to less than half a pound. There is no relationship, however, in the post-period. In this period, while the general blood calcium level is lower than in the supplement-period, it is definitely higher than initially. The fall is probably most marked in the cod-liver oil group.

An attempt was made at the beginning of the experiment to correlate the various blood calcium values with body weight, incidence of syphilis, etc. No relationship was observed. When, on the other hand, freedom from or infection with intestinal parasites (Ankylostoma, Taenia, Ascaris) is considered there is a suggestion that several of the low blood calciums may be in part related to helminthic infection. Whether malaria plays a part is less certain. It will be observed from Fig. 3 that only 3 out of 18 (*i.e.* 17 per cent.) low blood

¹ We are greatly indebted to Mr A. Walter, Statistician to the Conference of East African Governors, who subjected our figures to statistical analysis.

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calcium subjects were free from intestinal parasites (and these three showed sub-tertian malaria parasites in the blood). On the other hand, of the 24 normal blood calcium cases 14 (*i.e.* 58 per cent.) were free from intestinal parasites. Of the 42 only 5 were free from both malarial and intestinal parasites. These five fall in the normal blood calcium group. Work on the possibility of intestinal parasitic infection lowering the blood calcium level is being continued.

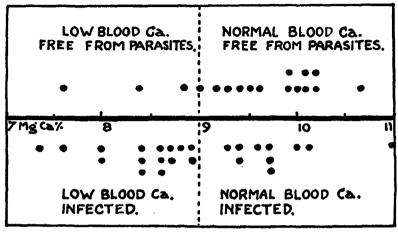


Fig. 3.

Meanwhile the possibility of deparasitisation tending to raise the blood calcium level and the body weight must be borne in mind. Throughout the experiment, however, all groups were alike deparasitised where necessary. This factor, therefore, if it be a factor, probably does not vitiate our general conclusions.

Balances.

It is doubtful if much importance can be attached to the results of the balance experiments. Their duration was undoubtedly too short and only representative subjects were examined. The possibility of fallacy is therefore evident, and the following points are accordingly indicated with the above qualifications in view.

From the table showing the average 96-hour and 24-hour balances for each group, it is obvious that the groups which did not have added calcium are either on a small negative balance or on one just slightly positive. In short, the subjects in the basal, cod-liver oil and olive oil and iodine groups are in calcium equilibrium.

The addition of cod-liver oil to Group III seems to have had little or no effect in promoting an increased retention of calcium. This is possibly related to the low calcium intake which the basal prison diet furnishes, *i.e.* 0.3 grm. Ca per day (Kelly and Henderson, 1927), as compared with a probable optimum requirement of 1.0 grm. per day (Sherman, 1926). Our finding is in agreement with that of Schabad and Sorochowitsch (1911) and Schloss (1914),

who found that the use of cod-liver oil was not always followed by an increased retention of calcium.

In the groups receiving mineral mixture, on the other hand, the subjects examined showed generally a definite positive balance, the higher average balance being in the minerals and cod-liver oil group. This is in agreement with further work by Schloss (1914), and Schabad and Sorochowitsch (1911), who both found that an increased retention of calcium followed the administration of calcium salts with cod-liver oil.

There are individual exceptions, of course, to these broad generalisations, noteworthy being that of subject 4021 in the mineral group, whose balance was very definitely negative. We cannot account for this rather abnormal figure. The highest positive balance is that of subject 1263 in the minerals and cod-liver oil group. It is striking that this particular subject was the one showing the greatest gain in weight in that group.

These balance determinations were carried out largely with a view to ascertaining whether natives could be trained to the routine of accurate excreta-collection technique demanded in work of this type. From this point of view the results were entirely satisfactory and justified the continuance of similar work in subsequent experiments to be described in a future communication.

SUMMARY AND CONCLUSIONS.

1. The addition to the Nairobi prison diet of either a mineral supplement (closely reproducing in composition the ash of cows' milk) or of a small dose of cod-liver oil, or a combination of these two, produced in adult African prisoners a distinct improvement in body weight.

2. Similarly an addition of olive oil and iodine produced a like effect but less in degree. The significance of this will require further investigation.

3. Evidence is adduced which suggests that one of the most important constituents of this soluble mineral mixture is calcium. Thus:

(a) The prison diet would appear on the basis of previous work to be deficient in the absolute amount of calcium present, nor does it seem rich in vitamins A and D. (In this connection it seems apposite to mention that cases of "night blindness" have frequently been reported.)

(b) The blood calcium of the 42 experimental subjects was initially at a low normal level.

(c) In the groups receiving the mineral mixture and/or cod-liver oil (which is a recognised calcium absorption promoting agent) the level of the blood calcium was invariably raised.

(d) As indicated above, the body weights in these groups increased. There appears to be, further, a certain amount of correlation between the increase in body weight and that of the blood calcium level.

(e) Balance experiments show that given favourable conditions (*i.e.* the addition of the mineral mixture alone or along with cod-liver oil) calcium will

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be absorbed in amount considerably in excess of that absorbed from the basal diet alone.

4. In the course of the work some evidence has emerged that certain other factors, *e.g.* intestinal parasites and minimum temperature, may respectively influence the blood calcium level and the body weight increase. Should these prove in future work to be real influences they would not vitiate our present conclusions.

Note. The work referred to in this and the two following papers was done in the course of a joint investigation by the Rowett Research Institute, Aberdeen, and the Kenya Medical Service on deficiency diseases in African natives. Thanks are due to the Director of the Rowett Research Institute and to the Director of the Kenya Medical Service for permission to publish the data.

Thanks are also due to Wazir Chand, Sub-Assistant Surgeon at the Nairobi Prison Hospital, who rendered valuable assistance in connection with the investigation.

It is with very great regret that I have to record the death in Nairobi of Dr J. M. Henderson, which took place after these communications were prepared for publication.

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