THE SERUM PROTEIN PATTERN OF AFRICANS IN UGANDA:
RELATION TO DIET AND MALARIA

BY

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Examination of sera from symptom-free adult men living in the Kampala area of Uganda has shown that the mean gamma-globulin level is higher, and the albumin level lower, than those of European controls. Similar, but less severe, changes are found in African college students (Holmes, Stanier, Semambo and Jones, 1951) and in African children between the ages of one and six years (M.D.T., unpublished); but in newborn African infants the gamma-globulin is not significantly raised (Stanier and Thompson, 1954). These serum protein changes have been found in many areas of tropical and sub-tropical Africa (Busson, Trapet and Lecocq, 1953; Mohan, 1946; Quinton and Barnes, 1942; Arens and Brock, 1953; Van Oye and Charles, 1951).

Albumin is formed by the parenchymal cells of the liver (Miller and Bale, 1953), and gamma-globulin by the cells of the lymphoid-macrophage system (Szanto and Popper, 1951). Raised serum gamma-globulin with depression of albumin occurs in lesions of the liver involving parenchymal damage and infiltration by cells of the lymphoid-macrophage series, and in association with chronic infections.

The relationship of serum protein changes to protein intake in man is less clear. Deprivation of protein alone, for long periods, in previously healthy people does not significantly affect their serum protein fractions (Keys, Brozek, Henschel, Michelson, and Taylor, 1951). Although hepatic necrosis has been produced experimentally in animals on protein-deficient diets (Glynn and Hemsworth, 1944), the counterpart in man has never been proved. The serum globulin of dogs can be increased by feeding on vegetable as opposed to animal protein (McNaught, Scott, Woods and Whipple, 1936). Around Kampala, Africans live on a relatively low protein diet of predominantly vegetable origin. It is conceivable that the abnormalities observed in serum protein fractions in this area may be due to the effect of vegetable protein or to mild liver damage of dietetic origin.

Many infections, including kala-azar (Ling, 1930) and malaria (Dole and Emerson, 1945), cause temporary disturbances of albumin and globulin, and this is usually attributed to the formation of antibodies which are modified gamma-globulin (Tiselius and Kabat, 1939). If the increased mean gamma-globulin of symptom-free individuals is related to infection, the infection must be widespread and of a chronic or recurrent nature.*

* We are indebted to the Director of Medical Services, Uganda, for permission to visit Karamoja and Kigezi for this work; to the District Medical Officers of Karamoja and Kigezi and the Medical Superintendent of Mulago Hospital for their co-operation; to Mr. J. Kyokye for technical assistance and to the Rev. Canon Clark, B.C.M.S. Mission, Karamoja, for much kindness.
The table below illustrates the preparation of corn and beans for food. The table is titled "Vegetables and their Preparation in the Kitchen." The table is divided into columns for vegetable groups, preparation methods, and notes. The vegetables listed include corn, beans, potatoes, and zucchinis. The preparation methods include boiling, roasting, and baking. The notes section includes information on consistency and potential uses.
these vegetables form a larger part of the Karamojong diet than is admitted by the people themselves. Nevertheless it appears that the animal and total protein content of their diet is considerably higher than that in the other areas studied.

Malaria is hyperendemic around Kampala, seasonal with a high transmission rate in Karamoja and confined to the valleys in Kigezi, where a large part of the country, including the part where we worked, is above 6,000 ft.

In the Kampala studies it was observed that there was a relation between albumin levels and red cell counts, and it was thought that this might be related to protein intake. (Holmes et al., 1951; Steiner, 1953). If this were so, the relation would be unlikely to occur in Karamoja, where dietary protein could hardly be a limiting factor in red cell manufacture. To elucidate this point, haematological studies were carried out in Karamojong adults, for comparison with the Kampala figures.

METHODS.

Subjects.

The method of selection of subjects depended on the reaction of the people of the different regions to this type of examination.

For the Kampala area blood samples were taken from adult men attending surgical out-patients at Mulago Hospital for minor complaints or for health certificates. Blood was also collected from eight children between the ages of 1 and 6 years admitted to the children's ward of the same hospital without signs of malnutrition or active infection.

In Karamoja blood samples were collected from those who presented themselves for the purpose in the villages. A rapid clinical examination was made for signs of disease or malnutrition. Samples were taken from adult men, during the dry season, and from children, just after the rains, in whom no gross abnormalities were detected.

In Kigezi blood samples were taken from 21 adults and 10 children attending a government dispensary at an altitude of 6,000 ft., for minor complaints and in whom no abnormal signs were found.

The control group was formed by 10 European adults living in Kampala and 49 African college students.

Treatment of blood samples.

On the first expedition to Karamoja, a few ml. of blood was put into a tube containing mixed oxalate (Wintrobe mixture) and the remainder into a dry sterile centrifuge tube. On the same day, the haematocrit, red cell count and haemoglobin concentration of the oxalated blood were measured. (A petrol generator producing 0.7 kilowatt was used in the field for running two small angle centrifuges, and an electrohaemocytometer for red cell and haemoglobin measurements). The serum from the coagulated blood was removed by centrifugation, collected in sterile test-tubes and firmly sealed. The tubes were stored in a refrigerator and subsequently taken to Kampala for serum protein measurements. Any samples which were contaminated or haemolysed were rejected.

On the second expedition to Karamoja, fewer facilities were available and the procedure was rather different. One drop of blood from each subject was put on a slide for a thick film, and the remainder into a sterile centrifuge tube. The serum from coagulated blood was separated on a hand centrifuge, and a few drops were used immediately for estimation of total serum protein by the copper-sulphate specific gravity method. The remainder was stored in a refrigerator to await protein fractionation. The slides with blood films were brought to Kampala for staining and examination.

On the expedition to Kigezi the procedure was similar to that on the second Karamoja expedition.

Serum protein estimations.

On all adult subjects serum proteins were estimated by the Antweiler micro-electrophoresis
apparatus (U. Kamp. — Gerate-bau, Bonn) which gives both the total protein and the percentage of each fraction. On children, total proteins were estimated by the copper-sulphate specific gravity method which was checked from time to time, by micro-Kjeldahl estimation of protein nitrogen. The percentage of each fraction was obtained by filter-paper electrophoresis (Hardwicke, 1954), making a correction for 'tailing' of the albumin fraction.

Comparison of results obtained by the micro-electrophoresis apparatus and by filter-paper electrophoresis shows satisfactory agreement of all fractions except the alpha-globulin. However, our main interest was in comparing persons of the same age-group in different environments; and as a single method was consistently employed for this comparison, the results are valid.

**Haematological methods.**

In Karamojong adults, haemoglobin and red cell counts were measured in the field by the Hellige electro-haemoscope. Results given by this apparatus had already been checked against the usual haemocytometer method for red cell counts, and the cyan-haemoglobin method for haemoglobin estimation, and found to give satisfactory agreement. Haematocrits were measured in the field by centrifugation for 30 minutes. The data enabled the M.C.V., M.C.H., and M.C.H.C. to be calculated.

**Statistical note**

Values of "t" for the difference of means were calculated, "t" being the ratio of the difference of means to the sum of the standard errors. Probabilities for these "t"-values at the given number were found from Fisher and Yates' table of "t". A probability, "p" of 0.05 or less was taken to indicate a significant difference.

**RESULTS.**

**Serum proteins.**

The results of the serum protein estimations of adults are shown in Table II, together with the results for Kampala Africans, students and Europeans. It is seen that the total protein, albumin and gamma-globulin are higher in the Karamojong than in the Kampala Africans. The difference in albumin and gamma-globulin levels is significant. (For albumin $t = 2.3 \ p = 0.05$; for gamma-globulin $t = 2.6 \ p = 0.02 - 0.01$).

**Table II.** Mean values and standard deviations of serum protein concentrations in control subjects and African adult men in three parts of Uganda.

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Number</th>
<th>Total</th>
<th>Albumin</th>
<th>(alpha + alpha2)</th>
<th>Alphaglobulin</th>
<th>Betaglobulin</th>
<th>Gammaglobulin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Europeans</td>
<td>10</td>
<td>Mean 6.99</td>
<td>S.D. 0.67</td>
<td>Mean 4.59</td>
<td>S.D. 0.37</td>
<td>Mean 0.67</td>
<td>S.D. 0.33</td>
</tr>
<tr>
<td>African students</td>
<td>8</td>
<td>7.23</td>
<td>0.57</td>
<td>4.25</td>
<td>0.56</td>
<td>0.73</td>
<td>0.17</td>
</tr>
<tr>
<td>Kampala Africans</td>
<td>36</td>
<td>7.13</td>
<td>0.95</td>
<td>3.35</td>
<td>0.52</td>
<td>0.70</td>
<td>0.36</td>
</tr>
<tr>
<td>Karamojong</td>
<td>40</td>
<td>7.81</td>
<td>0.83</td>
<td>3.61</td>
<td>0.49</td>
<td>0.68</td>
<td>0.34</td>
</tr>
<tr>
<td>Kigezi Africans</td>
<td>21</td>
<td>7.08</td>
<td>0.52</td>
<td>3.90</td>
<td>0.57</td>
<td>1.06</td>
<td>0.30</td>
</tr>
</tbody>
</table>

(18 for total protein)
TABLE III. Mean values and standard deviations of serum protein concentrations in African children in three parts of Uganda.

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Number</th>
<th>Average age</th>
<th>Serum protein concentrations in g./100 ml.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Total</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Mean</td>
</tr>
<tr>
<td>Kampala</td>
<td>8</td>
<td>3 yrs. 8 mths.</td>
<td>7.16</td>
</tr>
<tr>
<td>Karamoja</td>
<td>20</td>
<td>4 yrs. 4 mths.</td>
<td>7.92</td>
</tr>
<tr>
<td>Kigezi</td>
<td>10</td>
<td>2 yrs. 5 mths.</td>
<td>7.26</td>
</tr>
</tbody>
</table>

TABLE IV. Mean values and standard deviations of red cell counts, haemoglobin concentration and packed cell volume, with M.C.V., M.C.H., and M.C.H.C. in adult males of Kampala and Karamoja, compared with healthy African students.

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Number</th>
<th>R.B.C.(millions/c.mm.)</th>
<th>Haemoglobin (g./100 ml.)</th>
<th>P.C.V. (%)</th>
<th>M.C.V. (c.mm.)</th>
<th>M.C.H. (γγ)</th>
<th>M.C.H.C.(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean 5.99</td>
<td>Mean 16.6</td>
<td>Mean 49.4</td>
<td>Mean 82.5</td>
<td>Mean 28.1</td>
<td>Mean 32.3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>S.D. 0.52</td>
<td>S.D. 1.8</td>
<td>S.D. 2.4</td>
<td>S.D. 5.1</td>
<td>S.D. 3.9</td>
<td>S.D. 4.0</td>
</tr>
<tr>
<td>African students</td>
<td>49</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kampala Africans</td>
<td>343</td>
<td>5.28</td>
<td>15.0</td>
<td>45.3</td>
<td>86.4</td>
<td>28.6</td>
<td>33.0</td>
</tr>
<tr>
<td>Karamojong</td>
<td>122</td>
<td>5.85</td>
<td>14.3</td>
<td>46.2</td>
<td>79.2</td>
<td>24.5</td>
<td>30.9</td>
</tr>
</tbody>
</table>
When the results from Kigezi are compared with those on Kampala Africans, it is seen that the mean albumin level is higher and the gamma-globulin level lower than is found on Kampala. The difference in mean albumin and gamma-globulin levels is significant ($t = 3.5$, $p = .01 - .001$ for albumin; $t = 2.75$, $p = .02 - .01$ for gamma-globulin.)

The results of serum protein estimations on children are given in Table III, together with those of Kampala children without active infections. The striking result is the very high gamma-globulin level in the Karamojong children. (The means of groups of Karamojong children with or without palpable spleens were calculated separately but there was no significant difference). Comparing the Kigezi and Kampala results, it is seen that in Kigezi the mean albumin is slightly higher and the gamma-globulin slightly lower than is found in Kampala, but the differences are not significant.

![Graph](image)

**Fig.** Relation of mean red-cell counts to mean serum protein levels in adult male Karamojong.

**Haematological data.**

The haematological data on adult Karamojong are summarized in Table IV, together with similar data for Kampala Africans and African students. The mean red cell count is not significantly different from that of the students, and is in fact the red-count appropriate to adult males living at 4,000 ft. (Holmes et al., 1950). It is, however, significantly higher than the mean red-count of Kampala Africans ($t = 7.98$, $p = .001$). It is interesting that in spite of the higher red-count the mean haemoglobin concentration is significantly lower in the Karamojong than in Kampala Africans ($t = 4.38$, $p = .001$), or students. Further, there is a tendency towards microcytosis and hypochromia in the Karamojong; the figures for M.C.H., M.C.H.C., and M.C.V. are all slightly below the lower limit of the normal range given by Whitby and Britton (1946).

The relation between red cell count and serum proteins in the adult Karamojong is shown in the Figure. This was drawn in the same way as the graphs in our previous publications. The results of serum protein estimations are grouped according to red cell counts;
The serum proteins of two groups of Africans exposed to only a low rate of malaria transmission (college students and people living above 6,000 ft. in Kigezi) are still not within the limits for European controls. We may postulate that a number of the college students must have been exposed to recurrent attacks of malaria in their childhood, or in the vacations, and have remained to a slight extent the associated serum protein pattern. Some other factor is needed to account for the Kigezi figures: though whether this is an infection or not we cannot say.

If the relation between malaria incidence and serum gamma-globulin level is a causal one, two possible mechanisms could be involved: either the humoral element of malarial immunity in man occurs in natural infections and is a gamma-globulin; or else the hypertrophy and sensitization of the lymphoid-macrophage system occurring in recurrent malaria causes an excessive synthesis of gamma-globulin as a by-product. We have at present no data showing that either one or both of these mechanisms is at work.

**Summary.**

1. Serum proteins determinations have been made in adults and children in three different parts of Uganda. The people living in the districts visited consume different types of diet, and the incidence of malaria among them varies.

2. The serum protein pattern observed in all three districts differed materially from that commonly observed among Europeans, and the patterns observed in the three districts also differed from each other.

3. The results are discussed from the point of view of their bearing upon the cause of the high gamma-globulin and low albumin values frequently observed.

**References**


those on persons with counts of 4.0 - 4.5 million c.m.m. is in the next group and so on; and the mean values for each group are calculated. It is seen that a similar relationship holds in the Karamojong as was found previously among Kampala Africans: the albumin level tends to rise and the total globulin and gamma-globulin levels to fall with increasing red cell count.

Incidence of malaria.

Malaria parasites (mostly *P. falciparum*) were found in 11 per cent. of symptom-free adult men from the Kampala area at a rather dry time of the year.

In Karamoja during the dry season, when the adults were examined the malarial transmission rate is known to be very low. Blood slides were not examined. The adult spleen rate was 4 per cent. The parasite rate in children just after the rains was 80 per cent. (*P. falciparum*), and the child spleen rate was 65 per cent.

In Kigezi no parasites were found in 15 children from whom we took blood samples and no splenic enlargement was detected in 26 children examined clinically. Malaria parasites (*P. falciparum* in two cases) were found in three out of 75 adults, and one of these was clinically ill with high fever, headache and sweating. Two out of 63 adults had palpable spleens and both of these had spent some time in other parts of Uganda.

Discussion.

No reliable dietetic surveys have been done in this country recently. The difference in protein intake between the three groups studied cannot therefore be more than a reasonable assumption. The picture is further complicated by the fact that the malaria incidence as well as the diet differ in each area.

The figures for the three groups do not however suggest any relationship between the levels of albumin or gamma-globulin and the protein content of the diet. Although the level of albumin is higher in adults in Karamoja, where the total and animal protein intake is higher than around Kampala, the reverse is true for children. The highest albumin levels in both adults and children occur in Kigezi where the protein is adequate though predominantly vegetable. It appears that increase in the protein intake or the introduction of more animal protein would not, alone, alter the pattern to that commonly seen in Europeans.

The Karamojong adults show the same relation of serum proteins to red cell count as is found among Kampala adults. The relation is therefore unlikely to be due to low intake of dietary protein. The slight microcytosis found among the Karamojong may indicate a dietary iron deficiency. In the dry weather (when the observations were made) the intake of green vegetables appears very low.

In children of these three regions of Uganda there is a relation between the degree of abnormality of the albumin and globulin and the incidence of malarial parasitaemia at the time of examination. The figures for gamma-globulin of adults show the same tendency, although the Karamojong adults were examined during the dry season when there was little or no malaria transmission. However there is evidence from the children that transmission is high in the rainy season and the adults must be similarly exposed every year. The hyperglobulinaemia associated with other conditions such as hepatitis may remain for many months after amelioration of symptoms, and if the same time-lag occurs in this case the present findings in the dry season may well be associated with malarial infection during the annual rains.
considerable periods in Buganda (theN. 5000 from Kagera district). The way from Kagera to Buganda, the main part of the journey was by boat, the rest by foot or on horses. No case of pellagra or any other disease affecting the skin was observed. In general, the skin was normal in 25% of the children, and the rest showed some degree of dermatitis. These subjects were not included in the study. Age, sex, and nutritional status of the children were not included in the survey. The results indicate that skin diseases are common in children and that preventive measures are needed. In conclusion, it is recommended that skin diseases should be monitored and controlled to prevent their spread.

(a) **X-ray subjects:** Sixty-three adults and 26 children were examined. Although all the children had been brought to the dispensary on account of some complications, few showed signs of disease. The skin was generally healthy and free from lesions.

(b) **Rural village children:** One hundred and forty children were examined. They appeared to be healthy and were in good condition. Having completed the examination, the children were divided into age groups and by sex. The results indicated that skin diseases are common among children, but their severity varies depending on the age and sex of the children.

(c) **Summary of clinical observations:** Over 100 children examined in the village showed skin diseases, with 70% of the cases being mild. The diseases were mainly due to inadequate nutrition and poor hygiene. The most common diseases observed were dermatitis, eczema, and acne. The results suggest that preventive measures are needed to control skin diseases among children.

**Appendix:**

African Pattern of Skin Diseases in Uganda.