COMPARISON OF HUMAN SERUM PROTEINS OBTAINED BY POLYACRYLAMIDE GEL ELECTROPHORESIS IN DIFFERENT SOCIO-ECONOMIC GROUPS IN URBAN AND RURAL AREAS OF THAILAND (A PRELIMINARY REPORT)

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Serum proteins of 44 persons from a Bangkok university and 53 persons from a rural area in northeast Thailand were examined by electrophoresis in polyacrylamide gel. Serum proteins in the prealbumin and pretransferrin range, including transferrin, were determined quantitatively. The transferrin level and the levels of some alpha-1-globulin proteins were found to be significantly higher in the rural group. The role of nutritional factors in causing these differences is discussed.

Estimation of serum proteins may be a useful parameter for parasite and nutritional field surveys in the tropics (1). We have recently described the determination of serum protein fractions using the molecular sieving effect of 8–6–4.5% polyacrylamide vertical flat-bed gradient gel (2), by modifying the method of Allen and Moore (3). This method was employed to determine serum protein values from two distinct groups of the population, healthy, well-to-do adults in Bangkok and healthy adults in a rural area of northeast Thailand. This paper presents the values of individual proteins and the differences on the serum protein patterns between these two groups.

MATERIALS AND METHODS

Fourty-four individuals, lecturers and staff from Silapakorn University, Bangkok, and 53 persons from the town and neighbourhood of Pol, Muang Pol District, Province of Khon Kaen, about 370 km northeast of Bangkok, participated in the study. Twenty of the Silapakorn University and eighteen of the Muang Pol participants were females. The mean age of the Silapakorn University group was 32 years and of the Muang Pol group 36 years. A detailed history
of each person was taken, and measurements of height and weight as well as a physical examination were done. Fecal examinations for parasites were done for all subjects from the Muang Pol group. Fastening venous blood was drawn and haematocrit and haemoglobin values were measured; the serum was then separated, kept for at least four days at 4°C before being frozen at -70°C. Sera were maintained at this temperature for several weeks and thawed just prior to protein determination.

Whole blood haemoglobin values were determined by the cyanmethaemoglobin method (Aculute reagent, Ortho Diagnostic Corp., U.S.A.). Haematocrit (PCV) values were determined by a microcapillary method and mean corpuscular haemoglobin concentrations (MCHC) were calculated from the results. Electrophoresis was performed in 8–6–4.5% polyacrylamide vertical flat-bed gradient gel, using an Ortec® (Tennessee, U.S.A.) Model 4100/4200 (2). In every gel two reference standards were run simultaneously with the samples. The stained gels were taken to the Ortec Company, Munich, West Germany and protein patterns were read using an Ortec integrating microdensitometer system, Model 4300. (Some gels were damaged during transport, thus we were unable to read every peak in every serum sample.)

RESULTS

All individuals under investigation were considered to be healthy; physical examinations and histories gave no indications of disease. The fecal examinations of the subjects from Muang Pol were negative for parasites. Feces of the university group were not checked; but all persons in this group were under regular medical care. From the analysis of the haematological data no anemia was detected in either group. Weight-height ratios for adults (4) were similar in both groups, ranging from 70% to more than 114%; the mean for the Bangkok group was 98% and for the Muang Pol group 90%.

Values for the proteins under investigation are listed in Table 1. The albumin and transferrin levels of the Bangkok group were close to the values obtained from healthy blood donors in West Berlin, Germany using the polyacrylamide gel technique. Also the range of the first postalbumin peak (Gc-globulin, alpha-1-X-glycoprotein, easily precipitable alpha-1-glycoprotein, alpha-2 HS-glycoprotein) as determined by us, resembled that found in a “healthy” German population using the polyacrylamide technique (5).

As seen in Table 1 values for prealbumin were slightly lower and for alpha-1-acid glycoprotein markedly lower than recorded from German references. But those values were obtained with immunological methods and cannot be strictly compared with the data presented herein.

The first postalbumin peak was often separated into two peaks. The second
Table 1. Serum proteins in the prealbumin and postalbumin range

|                      | Silapakorn University group | Muang Pol District group | Values recorded by other |
|----------------------|-----------------------------|--------------------------|--------------------------|
|                      | N        | X (mg%) | SD     | X±2 SE (mg%) | V% | N        | X (mg%) | SD     | X±2 SE (mg%) | V% | t-test | normal (mg%) | range in (mg%) |
| Prealbumin           | 28       | 13.4    | 3.9    | 11.9–14.8 | 29.1 | 52       | 14.8    | 5.3    | 13.3–16.3 | 35.8 | —     | 25       | 10–40(20)     |
| α₁-acid glycoprotein | 26       | 35.7    | 18.1   | 28.6–42.8 | 50.6 | 46       | 25.9    | 13.3   | 22.0–29.8 | 51.4 | —     | 90       | 55–140(20)    |
| Albumin              | 44       | 4600    | 378    | 4480–4700 | 8.2  | 53       | 4400    | 358.6  | 4300–4490 | 8.2  | —     | 4780     | 4600–4960(5)  |
| Ge-globulin/α₁ X-glycoprotein/easily precipitable α₁-glycoprotein | 10       | 60.7    | 15.9   | 50.6–70.7 | 26.2 | 22       | 140.6   | 28.6   | 128.4–152.8 | 20.3 | p < 0.001 | —       | —            |
| α₂ HS-glycoprotein | 12       | 44.0    | 20.3   | 32.3–55.7 | 46.1 | 22       | 51.1    | 24.6   | 40.6–61.6 | 48.1 | —     | 60       | 40–85(21)    |
| Ge-glob/α₁ X-glyco/a.p. α₁-glycop./α₂ HS-glycop. b | 43       | 123.3   | 32.7   | 113.3–132.9 | 26.5 | 52       | 181.8   | 35.0   | 172.1–191.5 | 19.3 | p < 0.001 | 167      | 93–241(5)    |
| Ceruloplasmin/ Hemopexin | 34       | 44.9    | 15.7   | 39.5–50.2 | 35.0 | 47       | 52.3    | 14.5   | 48.0–56.5 | 27.7 | —     | —       | —            |
| Transferrin          | 38       | 261.5   | 34.9   | 250.2–272.9 | 13.3 | 53       | 306.2   | 51.0   | 292.0–320.4 | 16.7 | p < 0.001 | 222      | 178–266(5)   |

a From a group of subjects from Silapakorn University, Bangkok and a group of people from a rural district of Muang Pol, Province of Khon Kaen, northeast Thailand.

b First post albumin peak.
of these, according to Felgenhauer, should be the alpha-1-HS-glycoprotein peak (6).

Statistically significant differences ($p<0.001$ by student t-test) were found between the two groups in the first postalbumin peak and in the transferrin level. The first postalbumin peak was significantly higher in the Muang Pol group (Table 1). If the peak was separated into two, no significant difference was found between the alpha-1-HS-glycoprotein peaks of the two groups.

The transferrin level was significantly higher in the Pol group than in the Bangkok group. It was found that the transferrin values for females were significantly higher than in males in the Bangkok group. There was no significant difference between transferrin levels in males and females of the Muang Pol group (Table 2).

**Table 2. Transferrin levels in male and females from a group of people in Bangkok with a higher socio economic status and a group in a rural area of northeast Thailand.**

|                      | Silpakorn University group | Muang Pol District group |  |
|----------------------|---------------------------|--------------------------|---|
|                      | N    | X (mg%) | SD  | X+2 SE (mg%) | N    | X (mg%) | SD  | X+2 SE (mg%) | t-test |
| Male                 | 18   | 243.7   | 27.5| 230.7–256.7 | 35   | 302.8   | 52.7| 285.0–320.6 | $p<0.001$ |
| Female               | 20   | 277.6   | 33.5| 262.6–292.6 | 18   | 314.2   | 41.1| 294.8–333.6 | $p<0.005$ |
| t-test               | —    | —       | —   | —            | —    | —       | —   | —            | —      |

**DISCUSSION**

The values of serum proteins may be indicative of nutritional and health status. In our investigation, differences in both groups seemed to indicate a difference in the nutritional status of the Muang Pol group rather than a difference in the normal values of human serum proteins between two groups. High transferrin levels have been associated with iron deficiency anaemia (7). Although the tested individuals showed no clinical signs of anaemia, the iron levels seemed low. There is a high prevalence of iron deficiency anaemia in preschool children in northeast Thailand. The deficiency was not caused by hookworm infection since the prevalence of hookworm is much lower in this area; 90% of the children had iron deficiency but only 10–18% had hookworm infection (8, 9). Although dietary surveys reported by I.C.N.N.D (10) found that the total iron intake in the area was about 10–20 mg/person/day, which seems adequate, further studies in preschool children have indicated that about half of the total dietary iron was derived from rice and vegetables and only a small fraction was of animal origin (11). It was estimated that only one percent of iron in rice and only 3–4% in vegetables could be absorbed and utilized (12).
Therefore it may be assumed that available nutritional iron in the rural population is inadequate.

Recent observations indicated that transferrin may play an important role in the absorption of iron from the gut \(13, 14\). If the lack of iron in the Muang Pol group is due to low availability of iron in the food, it might be presumed that higher transferrin levels are compensatory response to improve iron absorption. The high transferrin levels in females of the Bangkok group is very likely due to the higher physiological iron loss in women.

The rise of the first postalbumin peak in the Muang Pol group also seems to be due to different food intake as compared with the Bangkok group. Another possible explanation would be a higher infection rate in this group, but we failed to obtain any evidence of this. Alpha-1, alpha-2 and beta-glycogobulins are reportedly high in localized and systematic infections \(15\). In that case the proteins belonging to the first postalbumin peak in the polyacrylamide electrophoresis are also elevated, because Gc-globulin migrates between the alpha-1 and alpha-2 glycoproteins, alpha-1-X-glycoprotein and easily precipitable alpha-1-glycoprotein migrates in the alpha-1 range and alpha-2-HS-glycoprotein migrates in the alpha-2 range in paper and cellulose acetate foil electrophoresis \(5, 16\). Alpha-1-acid glycoprotein and haptoglobin are proteins belonging to the alpha-1 and the alpha-2 range respectively and are reportedly high during inflammation \(17, 18\), but in our survey there was no significant difference in alpha-1-acid glycoprotein and haptoglobin between the Bangkok group and the Muang Pol group. We therefore believe that the difference was caused by nutritional factors. Patwardhan et al. \(19\) reported that in acute protein-calorie malnutrition alpha-1 glycoprotein was elevated whereas alpha-2 and gamma-globulins were not altered. In 12 individuals of the Bangkok group and in 22 of the Muang Pol group an alpha-2-glycoprotein (alpha-2-HS-glycoprotein) was observed as a single peak and there was no significant difference between these two groups as seen by the Gc-globulin, alpha-1-X-glycoprotein, easily precipitable alpha-1-glycoprotein peak. As discussed previously \(2\) the peak under discussion represents more proteins, but those which are listed represent the main proportion in this first postalbumin peak. Therefore it is possible that any of these three proteins reacts sensitively in the case of a low content of protein in the diet. As it is assumed that there are some differences in the dietary intake between the population groups examined, further investigations are necessary to prove the above assumptions.

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