THE INTERPRETATION OF HUMAN SERUM PROTEIN VALUES IN CENTRAL AMERICA AND PANAMA

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THE INTERPRETATION OF HUMAN SERUM PROTEIN VALUES IN CENTRAL AMERICA AND PANAMA[1, 2]

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Serum protein levels are commonly considered to reflect protein nutritional status and dietary protein intake. The determination of serum total protein has become a routine hospital procedure and recommendations for diet therapy are often based on it. Serum or plasma proteins are almost universally determined when nutrition surveys employ laboratory techniques. The determination of plasma protein by specific gravity method is the only chemical or physical blood test in the list of minimal laboratory observations suggested for public health nutrition studies in the *National Research Council Bulletin* on nutrition surveys (1). Section 3 of this bulletin states that even a slight reduction of the concentration of total serum protein *is significant* in persons who are ambulatory and without signs of complicating disease.

These suggestions of the value of serum or plasma protein determinations are not without impressive support. Many observations on animals confirm the development of serum protein deficiencies when they are kept on a diet quantitatively and qualitatively poor in protein (2). In man, many studies in famine areas during both World War I and World War II demonstrated low serum total protein (3, 4, 5, 6, 7). Nor are the observations confined to famine. Hegsted and co-workers (8) found total proteins decreased in adults fed vegetable diets at a low level of protein intake. Kark and his co-workers (9) in a study of troops in tropical areas find a correlation between the average intake of protein and its concentration in the serum. They report Indian troops with lower protein intakes to have lower serum protein values than corresponding British and American troops.

On the other hand, Anderson et al (10) for Mexico, the Netherlands Red Cross Feedingteam for the East Indies (11), Clements for Australia (12), the report on malnutrition and starvation in Western Netherlands (13), and Scrimshaw et al for Panamá (14), have already described high serum protein values in poorly nourished groups in these areas. Other workers in Europe after World War II failed to find a lowering of serum protein in groups in which they had

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[2] The following nutritionists have rendered invaluable assistance: Marina Flores and Emma Reh, Guatemala; Lucila Sogandares, El Salvador; Martha Thomason, Panamá; Margaret Moen, Margarite Barrett, and Virginia Stevenson, Rochester; and Audrey Johnson, calculations of El Salvador and Guatemala dietary data. The dietary histories in Rochester were made possible by a grant from the Nutrition Foundation.

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expected it, and those serum protein levels did not correlate well with the incidence of famine edema (2, 3, 11, 13). Even in the United States and Canada, many surveys have singularly failed to demonstrate hypoproteinemia even in low income groups (15, 16, 17). In recent unpublished studies of nutrition in pregnancy, neither W. J. Darby and his group (18) working in Nashville, Tennessee, nor ours (19) working in Rochester, New York, could find a positive correlation between dietary intake and serum protein levels. Whenever a study fails to reveal a positive correlation between protein intake and serum protein levels, it is customary to conclude that the deficiency in dietary protein is not sufficiently severe to influence the blood level of the protein. Sachar et al (20) find that a reduction in plasma albumin concentration of 0.5 grams per 100 cc involves a tissue loss of about 510 grams. This tends to support the concept that the deficiency of protein must be sufficiently advanced to produce a clearly demonstrable effect on blood levels.

However, the problem of interpretation which is posed by the serum protein data is not only one of failure of low protein intake to be reflected in serum protein levels, but also the occurrence of a distinct elevation of serum protein above normal levels under certain conditions in relatively undernourished individuals in the Central American area. This elevation above normal levels, even in undernourished groups, is suggested in the 7.39 to 7.61 grams per cent reported for different age and sex groups of Otomi Indians in Mexico (10) and by 7.75 for normal subjects in Mexico City (21). Recently, serum protein values for pregnant women in low income groups in Panama were reported to be higher than those for high income ones, and highest of all in the pregnant women of two very poor rural villages (22). The senior author has also observed personally normal or high serum protein values in patients in hospitals of Mexico City, Central America, Panamá, and Lima, Perú. These values were common in persons believed to be well hydrated but distinctly undernourished. It is also easy to find patients in all of these hospitals with very severe, often terminal, nutritional deficiency in which the serum proteins are greatly lowered. There is no doubt that *severe* deficiency produces marked lowering of blood protein. It seems equally clear from the data which follow, that under certain conditions, common in tropical or subtropical areas, a factor or factors other than protein intake can cause an *increase* in serum protein levels even in the presence of mild to moderate protein deficiency indicated by dietary history or clinical examination.

Although it has been suggested (23, 24) that protein deficiency may be detected at an earlier stage if the volume of the circulating plasma is determined simultaneously with the protein concentration in the blood, the serum protein values observed are often too high to be accounted for by changes in blood volume alone. Further, the patients themselves give no obvious reasons for suspecting that dehydration with its accompanying hemoconcentration plays an important role.

METHODS

All of the serum protein determinations presented have been done by the density gradient method of Lowry and Hunter (25) with the exception of the protein fractionation studies. Protein gradient tubes were set up in each region and determinations made on refrigerated specimens a few hours after they had been drawn. The separation of protein fractions was carried out by the technique of Kibrick and Blonstein (26) using 27.2% sodium sulphate and incubation for one hour. The final concentrations were determined by the biuret method (27).

The dietary intakes in Panamá have been estimated by a short-form interview method soon to be described. The dietary data for Salvador and Guatemala were obtained by daily visits to each family to obtain seven-day diet records. These data were calculated on the basis of analysis for regional foods carried out at M. I. T. (28, 29, 30) and by the food analysis section of the Institute of Nutrition of Central America and Panamá. The dietary intakes reported for Rochester were obtained by a single detailed interview patterned after that of Burke (31).

TABLE 1

Comparison of the total protein intake and serum protein levels of pregnant women, Gorgas Hospital, Panama Canal Zone

	NO. OF WOMEN	PROTEIN INTAKE GMS.	NO. OF WOMEN	SERUM PROTEINS GMS. %*	
U. S. White	51	62.8	48	6.42	High Income
Panamanian	52	66.1	18	6.53	
Panamanian	102	51.1	58	6.62	Low Income
W. I. Negro	57	46.8	60	7.03	

* Serum protein tends to decrease with gestation. For convenience the overall average for all of the gestation periods is taken, since the results are not altered thereby and serum protein changes with pregnancy are not a concern of this paper.

RESULTS

Pregnant Women, Panamá

In the Panamá Canal Zone, Gorgas Hospital patients are sharply divided into two groups on the basis of their payroll status. There is no overlapping between these groups. On the contrary, the economic status and security is very favorable in the high income group and generally unfavorable in those confined to the low income scale. Under these circumstances it is only to be expected that the high income groups would show a significantly higher intake of total protein. In Table 1 it is apparent that this is the case although even the protein intake in the high income groups does not meet National Research Council standards. It was expected that the somewhat superior status, economically and nutritionally, of the high income groups would be reflected in higher serum protein values. This was *not* the case. In table 1 it will be seen that Panamanians on a low income and poorer dietary protein intake had a slightly higher serum protein level than the Panamanians in the high income group. The protein intake of the West Indian Negroes was still lower and the serum protein still higher. Differences in protein intake between the high and low income groups have high statistical significance (Table 2) while the differences in protein intake between the two racial groups in each category are not significant. When blood levels are examined, high and low income Panamanians, while differing in dietary protein, show no significant difference in serum protein. On the other hand, the West Indian Negroes whose diet though grossly lower in protein is not significantly different statistically from that of the Panamanians in the same economic category, have a highly significant increase in serum total protein. It can be concluded from this table that in pregnant women in Gorgas Hospital, the

TABLE 2

Significance of differences in dietary and serum protein among rural and economic groups, Gorgas Hospital patients, Panama Canal Zone

	DIETARY PROTEIN			SERUM PROTEIN		
	t	d.f.	P	t	d.f.	P
High Income, American vs. Panamanian	1.08	101	0.27	1.18	74	0.23
High Income vs. Low Income Panamanian	4.41	102	*.0001*	1.33	70	0.19
Low Income, Panamanian vs. West Ind. Negro	1.22	107	0.22	2.70	116	*.007*

TABLE 3

Dietary protein in comparison with serum protein in pregnant white North Americans

	PROTEIN INTAKE	SERUM PROTEIN		
		0–16 weeks	17–28 weeks	29+ weeks
Panama				
Mean	62.8	6.47	6.41	6.45
Number	51	21	34	10
Rochester				
Mean	81.4	6.50	6.29	6.04
Number	204	237	222	117
t =	8.6	0.38	1.58	0.43
Probability	< .0001	0.70	0.11	0.65

groups with better nutrition as well as better general social and economic status, have lower serum protein.

When white North American pregnant women in Panamá are compared with those in Rochester, New York, in table 3, it will be noted that the superior protein intake estimated for the women in Rochester is not reflected by higher serum protein values. On the contrary, serum protein values in Panamá, even among the North Americans, are the same or higher than those for Rochester. The difference in protein intake is highly significant statistically. The differences in mean serum protein levels are not. It has been mentioned previously (14) that the values for pregnant women in Panamá in two very poor interior villages with diets certainly inferior to any of those shown in this table have a mean serum protein level higher than that of any of the Canal Zone groups.

Adults, Panamá and Guatemala

It will be seen in table 4 that the staff members of the Institute of Nutrition in Guatemala and their associates have a mean serum protein level that is not significantly different from adults in several rural villages where nutritional status is inferior. The rural Panamá group, with the highest mean serum protein average of the three, is the group believed to have the lowest protein intake. The difference in serum protein between adults in rural Panamá and rural Guatemala is highly significant statistically and cannot be explained by differences in protein intake. The Guatemalan groups are living at 5000–6000 feet and the Panamanians near sea level.

TABLE 4

Serum protein values in Panama and Guatemala adults

	NO.	MEAN	σ
Laboratory Staff			
Guatemala	71	7.29	.37
Poor rural villages			
Guatemala	78	7.26	.36
Panamá	22	7.94	.68
Poor rural villages Guatemala vs. Panamá	t = 4.53	P = <.0001	

TABLE 5

Serum protein values in rural school children

PLACE	NO.	MEAN	σ
Sacatepequez, Guatemala	164	7.26	.50
El Naranjo, Guatemala	24	7.62	.56
t = 3.00		P = .003	

School Children in Central America

In Table 5 a group of rural children in El Naranjo near Chicacao at a lower altitude in Guatemala than those in the Sacatepequez villages, have a significantly higher mean serum protein level. Since Hurtado (32) has reported a moderate but definite *increase* in serum proteins at very high altitudes, moderate differences in altitude would tend, if they had any effect at all, to result in lower values for El Naranjo on this table and rural Panamá on table 4. Thus, an altitude effect would presumably minimize the differences rather than explain them.

Urban school children in a moderately expensive private school (Table 6) showed no differences in serum protein levels from those for the orphanage in the same town or for the nearby rural villages. The values reported for all of these schools and for a public school in Honduras, are higher than the mean for Detroit (33) or New York (34).

In compiling table 7, children in El Salvador and Guatemala schools with serum total protein values above and below one standard deviation from the mean were placed in separate high and low protein groups. A few in each group

TABLE 6

Serum protein values in urban school children

PLACE	NO.	MEAN	σ
Tegucigalpa, Honduras	43	7.19	.44
Private School, Guatemala	25	7.31	.39
Orphanage, Guatemala	52	7.33	.37
Detroit U. S.*	545	7.00	.29–.58

* Beach et al, 1948.

TABLE 7

Dietary protein intake of school children with very high and low serum protein values Guatemala and El Salvador

	TOTAL PROT.		ANIMAL PROT.	
Serum Protein Group	<6.46	>7.64	<6.46	>7.64
Number	11	14	11	14
Daily Protein Intake	*40.5*	*32.6*	*5.1*	*3.9*
Standard Deviation	5.7	7.2	3.4	4.4
Student t	2.83		0.76	
Probability	0.01		0.45	

TABLE 8

Rural school children, Guatemala

TOTAL PROT.	NO.	MEAN	ALB.	% ALB.	GLOB.	% GLOB.	A/G RATIO
<7.64	27	7.93	4.33	54.7	3.60	45.3	1.22
6.46–7.64	81	7.16	4.02	56.2	3.15	43.8	1.30
>6.46	7	6.37	3.58	56.2	2.79	43.8	1.31
Variance ratio F* =			1.43		1.14		1.06

* A value of 3.08 is necessary for a probability of .05.

so separated had already been included in dietary surveys. For these few, the daily total protein and animal protein intake could be calculated. It is remarkable to note that the low serum protein group actually had the higher average dietary protein intake, according to these records. In the case of animal protein intake, the same trend will be noted in the raw figures. However, the standard deviation is so large in comparison with the mean that the differences in animal protein have no statistical significance. Table 8 demonstrates an actual *inverse* relationship between dietary protein intake and serum total protein levels in these groups.

Protein Fractionation Studies

It became important to know what protein fraction might be responsible for these abnormally high serum protein values. The most obvious hypothesis would be that the rise was due to an increase in globulin secondary to parasitic infection or chronic disease of some sort. Kagan (35), for example, has demonstrated that an increase in total protein generally consists of an elevation of globulin. Unfortunately, an apparatus for electrophoresis was not available, but table 8 shows the results of chemical albumin and globulin determinations. The children were again divided into three groups based on those with serum proteins above and below one standard deviation from the mean and those in the intermediate range. There is no difference in percentage of albumin and globulin observed between the low and intermediate groups. In the case of the high group, as compared with the intermediate and low, the albumin percentage is very slightly lower and that of the globulin, very slightly higher, but neither these differences

TABLE 9

Serum protein levels in rural school children, Guatemala, before and after school feeding

	SURVEY			RESURVEY			t	P
	No.	Mean	σ	No.	Mean	σ		
Milk and snack (16 weeks).........	35	7.38	.52	35	7.19	.37	1.81	.07
Control............................	33	7.31	.32	28	7.27	.30	.50	.53

nor those in albumin globulin ratio are of statistical significance. The correlation coefficient between albumin and globulin for the overall group is − 0.15.

It can be concluded from these data that in the children studied, the increase is in the albumin fraction as well as in the globulin fraction, although the proportionate increase is not equal.

Effect of School Feeding and Parasite Treatment

In table 9 protein values are tabulated for two schools in Guatemala. In Santa María Cauqué, the children are receiving a mid-morning supplementary feeding consisting of milk and various fruits and vegetables. After sixteen weeks of this "snack," the serum total protein value had dropped slightly and the variation within the group had decreased. That is, some of the extreme values had become more normal. Neither the drop in serum protein nor that in standard deviation is quite statistically significant. In the case of a control village, no change in mean or standard deviation occurred. All of these children had at least *Ascaris lumbricoides* and many had other intestinal parasites. They were not treated for these during the period reported here. Urban school children on a milk supplement alone in El Salvador also showed a drop but here also the variability was so great that the difference was not statistically significant. In two other urban schools in El Salvador parasite treatment alone apparently brought about a significant reduction in serum total protein and a combination of a complete lunch with milk and therapy for parasites appeared to achieve similar results.

Pregnant Women, Rochester, New York

The results of serum protein analysis in a recent pregnancy nutrition study in Rochester, New York, suggest that this anomalous finding has a wider distribution, although it seems more difficult to detect in temperate regions. In this project, 500 consecutive women presenting themselves for medical care in the clinics of Strong Memorial Hospital, and a similar number treated by staff physicians in their private offices were compared biochemically. Almost half of these also had dietary histories taken. In table 10 the dietary history results are as expected. The private patients had a higher intake of both total protein and animal protein. These were obtained using a detailed interview method, essentially that of Burke. Probabilities of .008 and less than .001 are highly significant.

The serum protein results however are *not* as expected. The difference in dietary protein appears to be related in the same inverse way to the serum protein

TABLE 10

Protein intake and serum proteins, in pregnant women, Rochester, New York

	DIETARY PROTEIN		SERUM PROTEIN			
	Total	Animal	0–16 weeks	17–28 wks.	29–36 wks.	37 + wks.
Number*	414	414	381	406	294	123
Private Patients	81 gms.	59 gms.	6.50	6.29	6.09	5.93
Clinic Patients	75 gms.	47 gms.	6.67	6.42	6.20	6.15
$t =$	2.66	5.84	1.84	2.06	1.23	1.67
Probability	.008	.0001	.06	.04	.22	.09

* The total includes 210 consecutive private patients and 204 consecutive clinic patients.

level as previously described for Panamá, Central America and the East Indies. In every gestation category the clinic patients had *higher* serum protein levels than the private patients. These differences were not statistically significant however, except in the 17 to 28 week gestation range.

Here in a situation where intestinal parasites are rare we encounter these same anomalously higher serum total proteins in the lower economic group on a diet lower in protein. It is interesting to note in retrospect that Pett and Hanley in their 1947 nutrition survey in Canada reported a range of serum protein values from 3.1 to 10.9 with the statement "no explanation of the few high values is available at present."

DISCUSSION

It is much easier to present these data than to explain them. Kagan considered values greater than 7.50 grams per cent as abnormal. On this basis, 20% of the individuals in the groups described from Central America are abnormal. But what is abnormal about them except their elevated serum protein? Diseases known to increase serum protein, such as certain malignancies, are not present. Dehydration does not seem to be the factor. These variations occur within racial groups as well as between them.

Intestinal parasites may be a contributing factor in Central America but are certainly not in Rochester, New York. The high incidence of iodine deficiency and endemic goiter may be a contributory factor since proteins tend to be high in hypothyroidism. All of the groups with higher serum protein have proportionately more vegetable protein in the diet. Whether there is something in a predominantly vegetable diet that disturbs the electrolyte balance or in some other way brings about an increase in the serum protein should be investigated. Current unpublished studies of the Institute of Nutrition of Central America and Panamá demonstrate a high incidence of positive cephalin flocculation tests in school children in Guatemala, but liver disease such as might be indicated by these results produces a decrease in albumin, not an increase.

We do not have the answer to this problem but are studying all of these possibilities. In the meantime we feel it important that this phenomenon of high serum protein values be generally recognized and critically studied, especially since the nutritional interpretation of serum protein values in tropical and subtropical areas is so seriously affected.

SUMMARY

1. Serum protein levels cannot be used for the detection of mild to moderate degrees of protein deficiency in humans.

2. Factors other than protein intake *per se* can bring about an increase in serum protein levels above normal averages even in the presence of mild to moderate protein deficiency as determined by diet interview or clinical examination.

3. Neither racial differences, altitude differences, nor simple dehydration explain this phenomenon.

4. The influence of endemic goiter and the high proportion of vegetable protein in the diet should be investigated in relation to it.

5. The increase in serum proteins appears to be an increase in both albumin and globulin fractions.

6. These phenomena of high serum protein values in the presence of mild to moderate protein deficiency are of special significance for tropical and subtropical regions.

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