ELECTROPHORETIC STUDIES OF SERUM PROTEIN PATTERNS IN NEWBORN INDIAN INFANTS

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The concentration of gamma globulin in the serum of apparently healthy adults from tropical areas is considerably higher than that of residents in temperate zones (Holmes, Stanier, Semambo and Jones, 1951; Satoskar and Lewis, 1954; Deegan, Gilles and McGregor, 1956; Kulkarni, 1957b; Edozien, 1957). In studies of plasma protein patterns in 100 Indian medical students (Satoskar and Lewis, 1954) gamma globulin and albumin levels were found to be twice as high as those among non-tropical residents. Furthermore there was no obvious correlation between past illness and the plasma proteins. Although some correlation was noticed between the dietary protein source and the level of gamma globulin, a definite cause for this rise could not be found. It would therefore be interesting to study the serum protein patterns of umbilical cord blood to see whether the newborn Indian infant also has a high level of globulin, particularly the gamma fraction, as compared with infants born in temperate zones.

Studies in the U.S.A. have shown the serum gamma globulin of cord blood to be as high as, or even higher than, that of maternal blood (Longsworth, Curtis and Pembroke, 1945; Orlandini, Sass-Kortsak and Ebbs, 1955; Saito, Gittleman, Pincus and Sobel, 1956; Jencks, Smith and Durrum, 1956). But similar studies on African infants showed the serum gamma globulin of cord blood to be lower than that of the maternal blood at delivery (Stanier and Thompson, 1954). Our previous studies on plasma proteins in normal pregnant women at term showed a lower serum albumin level than that reported by foreign workers; however the serum gamma globulin in these women was much higher (Coryell, Beach, Robinson, Macy and Mack, 1950; Mack, Robinson, Wiseman, Schoeb and Macy, 1951; Brown, 1954; Purandare, Satoskar and Lewis, 1954). The average birthweight of Indian newborn infants is considered to be lower

than the Western birthweight standard. Since the diet of the average Indian is qualitatively and quantitatively low in protein content, it would be of interest to find out if this is in any way related to birthweight and levels of the different protein fractions of the newborn.

In the present study serum protein fractions of the newborn infants and their correlations with birthweights and with the protein pattern of maternal blood at delivery have been investigated. The effect of pre-eclampsia in the mother on the protein pattern of infants has also been studied.

Materials and Method

The subjects of this study were divided into two main groups according to their economic status. Group A consisted of mothers admitted to Nowrosjee Wadia Maternity Hospital and belonged to a class with a low income, representing a sample from the majority of the population of Bombay. More than 80% were Hindus, who ate meat or meat products only occasionally. Group B included mothers from a private maternity hospital and belonged to a class with a much higher economic status. Moreover, these women were Indian Christians who had had meat and fish in their daily diet. They, however, represented a sample from the minority of the population.

Blood from the umbilical cord was collected in clean dry test tubes at the time of delivery from infants born normally. Clear, non-haemolysed serum was separated by centrifugation. The weight of the infant was recorded immediately after birth. Blood samples from the mothers were collected at the time of delivery and treated in the same way. The total protein content was determined by the copper sulphate specific gravity method (Phillips, Van Slyke, Dole, Emerson, Hamilton and Archibald, 1943). Paper electrophoretic separation, staining of electrophoretograms and the quantitative estima-

TABLE 1
ANALYSIS OF BIRTH WEIGHTS OF INDIAN NEWBORN INFANTS

Weight Group (lb.)	Low Income Group A			High Income Group B		
	Males (No.)	Females (No.)	Total (No.)	Males (No.)	Females (No.)	Total (No.)
3.00-3.99	61	44	105	1	1	2
4.00-4.99	162	163	325	2	3	5
5.00-5.99	445	461	906	13	18	31
6.00-6.99	489	474	963	45	57	102
7.00-7.99	171	117	288	47	35	82
8.00-8.99	36	22	58	19	10	29
9.00-9.99	4	4	8	3	1	4
10.00-10.99	0	0	0	1	0	1
Total	1,368	1,285	2,653	131	125	256
Mean weight for all groups (lb.)	5.90	5.83	5.87	6.98	6.63	6.81
S.D.	1.05	1.00	1.03	1.05	0.95	1.01
S.E.	0.030	0.028	0.020	0.092	0.085	0.063
No. of babies weighing below 4.84 lb.	205 (15%)	192 (15%)	397 (15%)	3 (2.3%)	4 (3.2%)	7 (2.7%)
No. of premature babies according to W.H.O. Standard	410 (30.0%)	422 (32.8%)	832 (31.3%)	10 (7.6%)	13 (10.4%)	23 (9.1%)

tion of proteins were carried out as described previously (Kulkarni, 1957a).

Results

Infant's Birth Weight and Mother's Socio-economic Status. In order to establish an average normal Indian birth weight standard the weights of 2,653 infants (51.6% males and 48.4% females) born of mothers belonging to group A were analysed. The results are presented in Table 1. The average birth weight of males was 5.90 ± 1.06 (S.D.) lb. and that of females 5.83 ± 1.00 (S.D.) lb., the average for the whole group being 5.87 ± 1.03 (S.D.) lb. Though the average weight of males was slightly more than that of females, this difference was not statistically significant ($p > 0.05$). A histogram shows the frequency distribution of the infants in various weight groups (Fig. 1). The birth weights

FIG. 1.—Frequency distribution of Indian infants, born of mothers with low economic status, according to their birth weights.

of the majority of normal Indian infants from the general population varied between 4.84 and 6.90 lb. and this range is taken as normal. According to this standard, 15% of the infants studied were below normal average weight. However, according to World Health Organization Standards, 31.3% of the infants were premature (below 5.5 lb.).

Table 1 includes an analysis of the birth weights of infants from the private maternity hospital (group B). Of the 256 infants studied, 51.1% were males and 48.9% were females. The average birth weight of males (6.98 ± 1.05 lb.) in this group was higher than that of females (6.63 ± 0.95 lb.), the difference being statistically significant ($p < 0.01$). The mean birth weight for the whole group was 6.81 ± 1.01 (S.D.) lb. The difference between the average birth weights of infants in this group and those in group A is highly significant ($p < 0.001$). Furthermore only 2.7% of the infants in group B were below the normal Indian birth weight standard mentioned above and only 9.1% of them were below the standard laid down by W.H.O., as compared to the high percentage of prematurity observed in group A.

Serum Protein Pattern and Infant's Birth Weight.

Table 2 shows the mean figures of different protein fractions of (1) group A infants with higher weights and their mothers, (2) group A infants with lower weights and their mothers, and (3) group B infants and their mothers.

The total protein content of infants with higher weights from group A was more than that of infants with lower weights ($p < 0.01$), and this difference was mainly at the expense of a significant rise in

TABLE 2

MEAN SERUM PROTEIN FRACTIONS (G./100 ML.) OF CORD AND MATERNAL BLOOD WITH STANDARD DEVIATION AND STANDARD ERROR

Group	No.	Birthweight (lb.)	Total Proteins	Albumin	Globulin			
					Alpha 1	Alpha 2	Beta	Gamma
Infants group A, weights above 4.84 lb...	20	6.13	5.64	3.67	0.14	0.30	0.39	1.14
		0.61	0.58	0.38	0.03	0.06	0.09	0.23
		0.14	0.13	0.09	0.01	0.01	0.02	0.05
Mothers of above	20	—	5.94	2.69	0.27	0.66	0.92	1.40
		—	0.56	0.42	0.06	0.11	0.16	0.29
		—	0.13	0.09	0.01	0.02	0.04	0.06
Infants group A, weights below 4.84 lb...	21	3.87	5.09	3.15	0.18	0.33	0.44	0.99
		0.86	0.67	0.44	0.04	0.06	0.10	0.31
		0.19	0.15	0.10	0.01	0.01	0.02	0.07
Mothers of above	21	—	5.76	2.50	0.28	0.65	0.92	1.41
		—	0.45	0.28	0.05	0.13	0.11	0.32
		—	0.10	0.06	0.01	0.03	0.03	0.07
Infants group B	10	7.09	6.21	3.93	0.17	0.35	0.50	1.26
		0.82	0.78	0.47	0.06	0.14	0.17	0.26
		0.26	0.25	0.15	0.02	0.04	0.06	0.08
Mothers of above	10	—	6.77	3.46	0.29	0.68	0.93	1.41
		—	0.45	0.43	0.07	0.20	0.17	0.32
		—	0.14	0.14	0.02	0.06	0.05	0.10
Infants of pre-eclamptic mothers group A	11	4.69	5.13	3.09	0.16	0.32	0.47	1.09
		1.74	0.44	0.45	0.06	0.06	0.24	0.15
		0.53	0.13	0.14	0.02	0.02	0.07	0.05
Mothers of above	11	—	5.09	1.92	0.30	0.67	0.84	1.36
		—	0.64	0.46	0.08	0.16	0.22	0.37
		—	0.19	0.14	0.03	0.05	0.07	0.11

serum albumin ($p < 0.01$). However, on comparing the serum protein patterns of mothers from both the higher and lower weight groups, no significant difference was noticed.

The infants born of mothers with higher economic status had higher serum total protein content than those of the low income group ($p < 0.05$). This was due to a small rise in all the serum protein fractions. The mothers with high economic status had 0.8 g. of total proteins more than that observed in mothers with low economic status. It should be emphasized that this rise was due solely to a 30% increase in the albumin fraction. As there was no significant change in the globulin fraction in either group, the A:G ratios were 0.82 ± 0.06 (S.E.) and 1.04 ± 0.12 (S.E.) in groups A and B respectively, the difference being highly significant ($p < 0.001$).

The correlation of the birth weight with the total proteins, albumin, total globulins and gamma globulin values is shown in Fig. 2. The serum total proteins and albumin showed a considerable rise with the increase in birthweight. The regression analysis confirmed that this rise in total proteins and albumin with increasing birthweight is highly significant ($p < 0.001$). Gamma globulin also revealed a similar rise but to a lesser extent ($p < 0.005$). However, the total globulins did not show any significant change.

Correlation of Mother's Serum Protein Pattern with that of the Infant. Fig. 3 shows the correlation of the total proteins, albumin and total globulins of cord blood with those of maternal blood. Serum total proteins of cord blood showed a progressive rise with the increase in the total protein content of maternal blood ($p < 0.01$). A similar trend was also observed in total globulin values ($p < 0.002$), but the albumin fraction did not show a significant change ($p > 0.1$).

Pre-eclampsia in Mother and Infant's Serum Protein Fractions. The average values of different protein fractions of pre-eclamptic mothers at delivery and their newborn infants are shown in Table 2. The pre-eclamptic mothers had notably low serum total proteins. This drop was mainly due to 30% reduction in serum albumin; the respective globulin fractions were not significantly different. The protein pattern of infants in this group was very similar to that of infants in the low weight group.

Serum Protein Fractions in Twins. Table 3 shows the values of different protein fractions of the cord blood obtained from twin infants born of mothers from group A. The average birth weight and the mean serum protein pattern of twins were very

TABLE 3
SERUM PROTEIN FRACTIONS (G./100 ML.) OF CORD BLOOD FROM TWINS

Pair No.	FIRST BABY							SECOND BABY						
	Birth weight (lb.)	Total Proteins	Albu-min	Globulins				Birth weight (lb.)	Total Proteins	Albu-min	Globulins			
				Alpha 1	Alpha 2	Beta	Gamma				Alpha 1	Alpha 2	Beta	Gamma
1	3.13	4.7	2.82	0.15	0.32	0.42	0.99	2.12	4.6	2.82	0.15	0.29	0.37	0.97
2	4.31	3.6	2.20	0.12	0.24	0.36	0.68	3.75	4.7	3.01	0.16	0.34	0.42	0.77
3	2.25	4.3	2.51	0.17	0.43	0.53	0.66	2.50	4.0	2.60	0.17	0.26	0.33	0.64
4	4.00	5.0	2.66	0.13	0.34	0.53	1.34	2.00	4.7	2.40	0.24	0.49	0.60	0.97
5	3.00	4.0	2.64	0.17	0.27	0.25	0.67	2.87	3.5	2.06	0.17	0.25	0.40	0.62
6	3.19	5.1	3.19	0.18	0.27	0.30	1.16	3.94	5.3	3.15	0.22	0.36	0.38	1.19
7	4.25	5.0	3.03	0.14	0.29	0.47	1.07	3.87	4.7	3.16	0.13	0.30	0.33	0.78
8	3.19	4.9	2.47	0.25	0.52	0.76	0.90	2.75	5.3	2.84	0.30	0.44	0.61	1.11
9	6.00	4.9	2.81	0.23	0.37	0.42	1.07	5.12	5.3	2.89	0.23	0.38	0.59	1.21
10	4.50	5.5	3.29	0.25	0.31	0.43	1.22	3.62	5.3	2.85	0.23	0.35	0.53	1.34
Mean	3.78	4.70	2.76	0.18	0.34	0.45	0.97	3.25	4.74	2.78	0.20	0.35	0.45	0.96
S.D.	1.07	0.54	0.32	0.05	0.09	0.14	0.24	0.97	0.61	0.34	0.05	0.08	0.11	0.25
S.E.	0.34	0.17	0.10	0.01	0.03	0.05	0.08	0.31	0.19	0.11	0.02	0.02	0.04	0.08

FIG. 2.—Relationships of serum total proteins, albumin, total globulins and gamma globulin with the birth weights of Indian infants.

FIG. 3.—Relationship of serum total proteins, albumin and total globulin content of maternal blood with those of cord blood.

similar to those of infants with low birthweights. Statistical analysis showed that the differences in the protein fractions between each pair were not significant.

Discussion

That the definition of prematurity based on weight alone is unsatisfactory is demonstrated in the National Survey on 'Maternity in Great Britain' where 29% of infants weighing 5.5 lb. or less at birth were not considered premature by length of gestation (Douglas, 1950). Our results show that

the average birth weight of the newborn Indian infant is much lower than the reported Western standard of 7.3 lb. (Bowes, 1950; Humphreys, 1954). Furthermore about 32% of Indian infants will have to be considered premature according to the W.H.O. standard.

The effect of the mother's socio-economic and nutritional status on the infant's birth weight has been noted previously (Drillien, 1957). The nutritional and socio-economic status of the average Indian is lower than that in Western countries, and this is reflected in lower levels of serum total protein and albumin observed during pregnancy in Indian women (Purandare *et al.*, 1954; Kulkarni, 1957b). Judged by the Registrar General's 'Classification of Occupations', 1950 (quoted by Drillien, 1957), our group consisting of mothers with low incomes corresponds to classes IV and V, whereas mothers in group B belonged to classes I and II. The average birth weight of an Indian infant born of a mother belonging to a low income group was about 5.9 lb., and this figure is in good agreement with that reported from other parts of India (Tampan, 1955; Sen, 1955; Bhonsale, 1955). The low levels of serum protein and albumin in the cord blood from infants born of mothers in the low income group are probably explained by the low level of the dietary protein which is prevalent in India. This is supported by the fact that the mothers with high economic status as well as their infants had serum protein and albumin values very similar to those reported by Western workers (Table 4). Also the average birth weight of these infants was similar to the reported Western standard; while the prematurity rate in this group was only 9.1%. As the serum total protein and albumin were correlated with the birth weights, the low weights observed in infants

born of mothers with low economic status indicate the low nutritional status of their mothers.

The average concentrations of different protein fractions in infants with weights below 4.84 lb. were lower than those in infants weighing above 4.84 lb. from the same group. This finding is in general agreement with the observations of Saito *et al.* (1956).

The increase in gamma globulin level of the apparently normal Indian adult (Satoskar and Lewis, 1954; Kulkarni, 1957b) suggested the possibility that this represented a continuation into adult life of an infantile trait. This study indicates that Indian infants are born with higher gamma globulin levels than those reported in temperate zones (Table 4). Furthermore, this gamma globulin level shows very little further rise up to adult age, so that no significant difference has been observed between the gamma globulin level of an infant and an adult of 30 years of age (Kulkarni, unpublished). Another difference is that the Indian infant is born with a lower gamma globulin level than its mother, whereas the Western infant is born with a gamma globulin level higher than its mother. Similar findings in African infants have been reported by Stanier and Thompson (1954).

In contrast to the differences in serum total protein and albumin values, the gamma globulin levels observed in infants of groups A and B were very similar, irrespective of their mothers' nutritional and socio-economic status. The gamma globulin levels of mothers in these two groups were also very similar and definitely higher than that reported by foreign workers. It is concluded therefore that the increased gamma globulin level observed in Indian infants is due to a high gamma globulin level in the maternal blood. The direct transmission of homo-

TABLE 4
SERUM PROTEIN FRACTIONS (G./100 ML.) OF CORD AND MATERNAL BLOOD, REPORTED FROM VARIOUS COUNTRIES

Subjects	No.	Country	Method	Total Proteins	Albumin	Globulins				Author
						Alpha 1	Alpha 2	Beta	Gamma	
Newborn infants ..	11	U.S.A.	E	6.18	3.82	0.29	0.49	0.60	0.97	Longworth <i>et al.</i> (1945)
Mothers at delivery ..	11			7.17	3.56	0.47	0.79	1.62	0.72	
Newborn infants ..	14	Africa	E	5.31	3.26	0.73 0.98		0.26	1.03	Stanier <i>et al.</i> (1954)
Mothers at delivery ..	9			5.61	2.45			0.75	1.42	
Newborn infants ..	28	Canada	C	6.24	3.63					Orlandini <i>et al.</i> (1955)
Mothers at delivery ..	28			6.68	3.50					
Newborn infants ..	14	U.S.A.	PE	5.44	3.31	0.28	0.50	0.51	0.84	Saito <i>et al.</i> (1956)
Newborn infants ..	41	U.S.A.	PE	5.70	3.94	0.14	0.41	0.44	0.77	Jencks <i>et al.</i> (1956)
Mothers at delivery ..	325			6.60	3.80	0.33	0.73	0.90	0.84	
Newborn infants ..	20	India	PE	5.64	3.67	0.14	0.30	0.39	1.14	This paper
Mothers at delivery ..	20			5.94	2.69	0.27	0.66	0.92	1.40	

E = Electrophoresis.

PE = Paper electrophoresis.

C = Chemical.

logous maternal albumin and gamma globulin to the foetal circulation has recently been demonstrated in rhesus monkeys; furthermore, it was found that the gamma globulin was transmitted some 15 to 20 times more easily than albumin, whereas the alpha and beta globulins did not reach the foetal circulation (Bangham, Hobbs and Terry, 1958).

The serum proteins of infants born of pre-eclamptic mothers showed a pattern similar to that seen in infants with weights below 4.84 lb. However, the serum total proteins and albumin in these mothers were considerably lower than those in normal mothers from the same group. Similar protein patterns have been reported previously in pre-eclamptic women at term (Purandare *et al.*, 1954). These changes in the protein pattern of mothers are attributed to probable liver damage. It seems however that this liver damage, if any, in the mother, does not considerably affect the serum protein pattern of her infant. Slight lowering of the total proteins and albumin observed in infants of pre-eclamptic mothers, as compared with those of infants with higher weights, might be due to their premature births.

Summary

The birth weights and serum protein patterns of Indian newborn infants and their mothers have been studied. Infants of mothers of low income group had lower birth weights and serum total protein and albumin than those born of mothers with high economic status. The serum total globulins of these infants showed a definite relation to that of their mothers. The gamma globulin levels in these infants were higher than the corresponding figures reported from temperate zones. This rise could not be correlated with the nutritional status of the mothers.

The serum protein patterns revealed no significant difference in each pair studied.

The serum protein pattern of infants born of pre-eclamptic mothers resembled that of infants in the low weight group, although the pre-eclamptic mothers had considerably lower serum total protein and albumin content.

The implications of these findings are discussed.

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