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SERUM GLYCOPROTEINS IN DIABETES MELLITUS AND SCHIZOPHRENIA.

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Abstract

Chemical determination of serum glycoproteins as proteins-bound hexose and sialic acid has been studied in 54 healthy male donors, 52 diabetic patients and 45 schizophrenic pateints. The average values of protein-bound hexose and sialic acid were found to be 125±25 mg %, 47.8±12.5 % in normal persons, 133±45 mg %, 41.36±10.2 mg% in diabetic patients 231±36 mg %, 49.9±9.6 mg % in schizophrenic patients respectively. The discrepancies among the group were discussed.

Introduction :

Glycoprotein consists of a polypeptide chain covalently bound with carbohydrate as oligosaccharide chains usually through serine, threonine and asparagine. The sugars in the oligosaccharides one amino sugars, N-acetyl glucosamine and N-acetyl-galactosamine; neutral sugars, mannose, galactose and fucose; and derivatives of neuraminic acid or sialic acids (1). Human serum contains a very large number of glycoproteins such as orosomucoid, fetuin, ceruloplasmin, alpha 2 - glycoproteins, haptoglobins, alpha 2 - macroglobulins, 7 - 5 gammaglobulins, 19-5 gammaglobulins and transferrin. Many of these specifically play a transport role in blood (2). Some are antibodies and hormones. Other functions of these serum glycoproteins are still being investigated.

It has been accumulated in literatures regarding the elevation of the protein-bound carbohydrate of serum in a great number of diseases, such as cancer, tuberculosis, rheumatiod arthritis, pneumonia and myocardial infarction (3). Decreased levels of some glycoproteins of plasma have been discussed. This might be due to a failure of normal hepatic synthesis

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or to a loss of glycoprotein in the urine.

Goodman et al. (4) reported that orosomucoid was elevated in schizophrenic patients who had no other pathological conditions. It might be of interest to investigate serum glycoproteins in Thai Schizophrenic persons and diabetes mellitus patients. It is the purpose of this article to report the level of protein-bound hexoses in serum of normal persons, schizophrenic persons, and diabetes millitus in northern Thailand. A discrepancy among the groups will be discussed.

Materials and methods

Normal blood serum was obtained from 45 male donors, aged 20 - 35 years. 52 blood serum of diabetes mellitus and 45 of schizophrenics from Suanprung Hospital were brought for analyses of bound hexoses and sialic acids.

Determination of protein-bound hexoses

Protein-bound hexoses is defined as those total neutral sugars covalently linked with protein precipitated by 95 % ethanol. It was determined by the method of winzler (5) 0.2 ml of serum was well-mixed with 5 ml of 5 % ethanol in a graduated centrifuges tube. The tube was spun for 15 minutes. The supernatant was discarded and the precipitate was dissolved with 0.2 ml of 0.1 N NaOH. 0.1 ml. of the mixture in duplicates was pipetted into a test tube (15×150 mm.) and diluted upto 1.0 ml with distilled water. A blank was made with 1 ml distilled water 0.25, 0.5 and 1 ml of galactose and mannose mixture (0.2 mg/ml) was used as the standards. The final volume of all samples and standards was mede up to 1.0 ml 8.5 ml of orcinol-H_2SO_4 reagent * were added to all the tubes and well-mixed by conversion. The tubes were covered with marbles to minimize the evaporation and heated in a water bath at 80°C for 15 minutes. Optical densities were read at 540 mu by Bausch & Lomb Spectronic 20.

Determination of total Sialic Acid

Total sialic acid was analysed by thiobarbiturate method (6). To 0.3 ml serum in a 12×100 mm test tube, 3.2 ml of 5 % TCA was added, and mixed. The tube was heated in water bath at 100°C for 15 minutes, cooled in tap water, and centrifuged for 20 minutes, 0.5 ml filtrate was drawn mixed with 0.1 ml of 0.04 M periodic acid and then cooled at 0°C for 20 minutes, 1.25 ml resorcinol-HCl ** was added and cooling at 0°C was centinued

* Orcinol-sulfuric acid reagent = a mixture of 60 % H_2SO_4 and 1.6 % orcinol in 30% H_2SO_4 in a ratio 7.5 : 1.

** Resorcinol-HCl reagent = 0.6 gm Resorcirol in 60 ml of 28 % HCl plus 25 u mole $CuSO_4$ and 40 ml distilled water.

for another 5 minutes. The final mixture was placed in boiled water at 100°C for 15 minutes and consequently in a water bath at 37°C for 3 minutes. The optical density was read at 630 mu by Spectronic 20.

Results

Each of Tables 1, 2, 3 shows the comparison of serum protein-bound hexose and sialic acid in 54 normal subjects, 52 diabetes mellitus patients and 44 schizophrenic patients respectively.

The average values with standard deviation of serum. protein-bound hexose and sialic acid also demonstrated.

Discussion

The average values of protein-bound hexoses and sialic acid in normal Thai serum determined in the present study were 125±25 mg% and 47.8±12.5 mg% respectively (Table 1). The hexose content is close to that reported by Winzter as about 121±2.1 mg% (5). By diphenylamine reaction the level of sialic acid in normal American serum reported by the same author was 60.0±3.1 mg% (5). In our studies, sialic acid was determined by resorcinol method, therefore this might give the lower values. Besides different technics of the methods, the discrepancy might be due to physiological variations, races and ages of the subjects. Haralambie (7) found that physiological exercises caused an increase of serum glycoproteins.

In this report there seemed to be no concentation change of glycoproteins in serum obtained from 52 diabetic patients. High concentation of glucose in their blood does not interfere the protein-bound hexoses in the serum. The free sugar was separated and washed out during protein precipitation with 95% alcohol. Total sialic acid concentation was also normal. There was a fluctuation in analyses of serum bound hexoses and sialic acid (Table 2). Shetlar (8) showed no significant change of serum glycoprotein in diabetes mellitus, but Srinivasa et al. (9) reported an increase of galactose-containing glycoproteins in renal cortical tissue of the diabetic patients. The latter report also showed the relationship between serum glycoprotein concentration and the severity of the disease. It is still a matter of conjecture.

It has been previously reported by Goodman et al (4) that serum glycoproteins were greatly increased in schizophrenic patients. Our work firmly supported the investigation, since protein-bound hexoses in serum of Thai schizophrenic patients was two times higher than the normal Thais (table 3), some rised upto three times more. All achizophrenic patients we observed had no history of pathological diseases or any physical sickness. the

TABLE 1

Quantitative Analysis of Protein-bound Hexoses and Sialic Acid in Normal Serum

No.	mg.% protein-bound hexose	mg. % sialic acid	No.	mg.% protein-bound hexose	mg.% sialic acid
1	101	41.0	28	117	79.1
2	90	54.6	29	100	45.5
3	104	43.2	30	208	78.0
4	93	56.0	31	124	52.7
5	154	–	32	141	64.1
6	125	38.5	33	150	64.1
7	149	49.0	34	130	62.2
8	107	–	35	182	48.0
9	237	43.2	36	109	46.0
10	105	35.0	37	155	46.0
11	97	38.5	38	116	42.1
12	129	35.5	39	143	–
13	90	36 2	40	172	43.3
14	139	55.5	41	127	47.0
15	102	43.2	42	149	–
16	112	36 2	43	116	50.0
17	110	35.0	44	127	42.1
18	95	32.7	45	190	–
19	120	45.5	46	97	52.5
20	100	35.0	47	148	50.0
21	190	52.7	48	108	38.5
22	118	43.3	49	153	–
23	123	40.8	50	137	45.5
24	111	62.0	51	121	37.4
25	128	57.4	52	155	–
26	73	78.0	53	110	–
27	109	62.0	54	95	–

Protein-bound hexose mean = 125 $\pm$ 25 mg. %

Sialic acid mean = 47.8 $\pm$ 12.5 mg. %

TABLE 2 Quantitative Analysis of Protein-bound Hexoses and Sialic in Serum Obtained from Diabetic Patients

No.	mg% protein-bound hexose	mg% sialic acid	No.	mg% protein-bound hexose	mg% sialic acid
1	111	–	28	170	48.0
2	198	42.0	29	180	56.0
3	177	39.4	30	201	–
4	109	44.2	31	100	39.8
5	109	49.0	32	178	58.4
6	100	38.5	33	203	52.5
7	84	50.5	34	158	51.5
8	115	–	35	115	26.8
9	150	55.0	36	81	35.1
10	93	49.0	37	213	42.0
11	101	44.2	38	111	24.5
12	116	–	39	210	42.0
13	145	–	40	183	28.0
14	117	37.0	41	144	42.0
15	135	49.0	42	131	37.4
16	93	45.5	43	103	33.8
17	125	55.3	44	195	37.0
18	88	–	45	124	33.8
19	85	35.0	46	212	–
20	101	53.7	47	203	44.5
21	75	–	48	189	–
22	81	36.2	49	185	52.5
23	80	31.5	50	102	29.1
24	142	37.4	51	191	74.5
25	69	31.5	52	102	43.2
26	152	44.5			
27	109	37.4			

Protein-bound hexose mean = 133 ± 45 mg %

Sialic acid mean = 41.36 ± 10.2 mg %

TABLE 3 Quantitative Analysis of Protein-bound Hexoses and Sialic Acid in Serum Obtained from Schizophrenic Patients

No.	mg% protein-bound hexose	mg% sialic acid	No.	mg% protein-bound hexose	mg% sialic acid
1	200	40.7	23	222	39.4
2	151	40.7	24	215	39.4
3	206	55.0	25	215	53.9
4	198	36.0	26	238	45.6
5	167	36.0	27	192	50.1
6	177	46.0	28	237	54.0
7	163	48.0	29	204	66.5
8	198	33.6	30	350	54.0
9	234	50.1	31	263	46.5
10	195	45.2	32	267	73.0
11	220	57.0	33	211	46.5
12	220	58.4	34	206	57.0
13	206	54.0	35	225	51.8
14	202	64.0	36	247	67.0
15	160	31.0	37	200	38.0
16	187	46.0	38	291	54.0
17	246	50.1	39	229	50.1
18	297	55.0	40	206	57.0
19	208	46.0	41	285	73.0
20	257	68.3	42	267	55.0
21	191	40.7	43	172	46.0
22	215	35.0	44	340	55.0
Protein-bound hexose mean = 231 $\pm$ 36 mg%					
Sialic acid mean = 49.9 $\pm$ 9.6 mg%					

marked increase of serum glycoproteins might be due to the mental stress and internal metabolic changes in the patients rather than due to tissue destruction or proliferation.

Instead of increasing as did the bound hexoses, total sialic acid did not change in the schizophrenic serum. The change of sialic acid should be seen along with that of bound hexoses since sialic acid is contained as terminal units of serum glycoproteins (10, 11). There is a tendency to state that those increased glycoproteins contain less or no sialic acid. Are these glycoproteins abnormal. To answer the question intensive investigations on the glycoproteins in schizopzophrenia are needed.

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