JCI The Journal of Clinical Investigation

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J Clin Invest. 1925;1(5):451-472. https://doi.org/10.1172/JCI100024.

THE PLASMA PROTEINS IN RELATION TO BLOOD HYDRATION

II. In Diabetes Mellitus

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(Received for publication, February 14, 1925)

Rowe (17), in 1917, reported observations of the serum proteins of 10 patients with diabetes. The only remarkable thing was the extreme variability of the findings. Both high and low values were obtained. In the majority of instances albumin and globulin preserved their normal proportions; but in some instances globulin appeared relatively high. Subsequent observers have confirmed Rowe and the general opinion appears to be that there is nothing characteristic about the serum protein level in diabetes.

In table 1 are shown the results of 52 observations of the plasma proteins of 31 patients with diabetes mellitus of varying degrees of severity, studied at different stages of the disease.

EXPERIMENTAL PROCEDURE

Whenever there is no note to the contrary, the blood was taken in the morning, before the patient had breakfast and the morning dose of insulin. The blood was withdrawn in a syringe and coagulation prevented by the addition of enough neutral potassium oxalate to make an 0.2 per cent oxalate solution in the blood. The treatment of the blood is indicated in column 6 of the table. Those specimens marked *cont.* were obtained without stasis and immediately placed over mercury in a blood sampling tube of the type described by Austin, Cullen, Hastings, McLean, Peters and Van Slyke (1). Usually venous blood (*ven.*) was employed, but occasionally arterial (*art.*) was used instead. In specimens indicated as *cap.* the blood was withdrawn without precautions against air contact and brought into equilibrium with 40 mm. of CO_2 in the air at 38°C. by the method previously described (16) before it was placed in the sampling tube over mercury. From the mercury sampling tube part of the blood was transferred to a centrifuge tube

TABLE 1

Case number	Blood sugar	Plasma CO_2	Oxygen capacity	Cell volume	Plasma proteins	Character of blood sample	Remarks
				Section 1			
	mgm. per 100 cc.	*vols. per cent*	*vols. per cent*	*vols. per cent*	*per cent*		
19043	293	52.6	21.4	43.8	6.19	Ven. cap.	Male, aged 62, with mild diabetes admitted for cataract operation. Blood taken the day after admission
5105	231	53.3	21.3	45.6	6.91*	Ven. cap.	Male, aged 38, with mild diabetes. Blood taken day after admission
26903	215	53.0	23.2		7.21	Ven. cap.	Male, aged 56, with mild diabetes, arteriosclerosis and hypertension. Blood taken 6 days after admission
12562	139	48.9	20.0	43.0	7.41	Ven. cap.	Female, aged 45, with mild diabetes and an enlarged liver. Blood taken the morning after admission
77914	141	59.3	20.4	41.5	6.81*	Ven. cap.	Male, aged 44, an outpatient with alimentary glycosuria or mild diabetes. Blood taken before breakfast
10919	132	55.5	22.1	44.3	7.31*	Ven. cap.	Male, aged 40, brother of the preceding patient, with mild diabetes and central nervous system syphilis. Blood taken the morning after admission
10801	411	45.1	19.5	41.7	6.60	Ven. cap.	Female, aged 66, with mild diabetes, arteriosclerosis, hypertension and paralysis agitans. Blood taken 3 days after admission
16727	147	57.5	20.5	44.6	7.36*	Ven. cap.	Male, aged 55, with mild diabetes and hypertrophic spondylitis
15733	319	63.8	17.8	38.5	5.98	Ven. cap.	Female, aged 64, with mild diabetes, arteriosclerosis and hypertension. Had suffered a small cerebral hemorrhage the day before admission, and had also vomited a few times. Blood taken the morning after admission
26143	110	56.0	22.0	45.5	6.94	Ven. cap.	Male, aged 50, with mild diabetes and chronic frontal sinusitis

20387		57.2	18.1	38.2	5.58	Ven. cap.	Female, aged 43, with mild diabetes. On the day of examination she showed a transitory glycosuria and began a rather extraordinary diuresis that lasted 3 days. Urine shows a moderate amount of glucose but no acetone
10629	291	47.8	18.7	41.5	7.48*	Ven. cap.	Female, aged 62, with apparently mild diabetes aggravated by a fracture of her skull. Urine contains large amounts of sugar and acetone
				Section 2			
29127	167	57.4	18.4	42.6	6.72	Ven. cap.	Female, aged 50, with mild diabetes aggravated by presence of lobar pneumonia. Blood taken the morning after admission when urine contained large amounts of sugar but no acetone
15924	112	43.5	14.3	32.3	6.25	Ven. cap.	Female, aged 65, with diabetic gangrene of foot, aggravating mild diabetes, after amputation
10888	311	74.2	17.8	38.3	6.47	Ven. cap.	Female, aged, 53, with chronic and acute arthritis. Temperature at time of examination, 99.6°, arthritis symptoms relieved. In spite of high blood sugar urine is sugar free
15175	244	53.4	20.9	41.9	7.25	Ven. cap.	Female, aged 60, with mild diabetes complicated by a fractured patella and lacerated wounds of the forehead. At time of examination, the second day after admission, urine showed large amounts of sugar but no acetone
34618	309	59.9	17.7	34.2	6.40*	Ven. cont.	Female, aged 33, with moderately severe diabetes, the morning after admission to the hospital. Urine contains considerable sugar, acetone and diacetic acid. Patient is thin, but not extremely emaciated and not dehydrated
1944	423	34.2	19.7	38.3	7.35*	Ven. cap.	Male, aged 17, with severe diabetes and diabetic acidosis, at time of admission to hospital. Marked emaciation, polydypsia and polyuria. Urine shows moderate amount of sugar and considerable acetone

TABLE 1—*Continued*

Case number	Blood sugar	Plasma CO_2	Oxygen capacity	Cell volume	Plasma proteins	Character of blood sample	Remarks
				Section 2—Continued			
	mgm. per 100 cc.	*vols. per cent*	*vols. per cent*	*vols. per cent*	*per cent*		
15267	811	52.8	19.2	37.4	6.28*	Ven. cap.	Male, aged 21, with severe diabetes. Blood taken at time of admission. Urine showed large amount of sugar and moderate acetone. Patient emaciated and dry. Blood pressure: systolic 90, diastolic 60
10572	290	63.3	17.2	36.2	5.75*	Ven. cap.	Male, aged 46, with moderately severe diabetes. At the time of the blood examination, November, 1922, on inadequate diet, urine free from sugar and ketones. Slight edema of ankles
	271	45.1	17.5	41.4	7.68*	Ven. cap.	The same patient a year later, October, 1923, the morning after his admission to the hospital for an acute upper respiratory infection. Urine contains large amounts of sugar and acetone
18067	1,000	16.5	23.3	44.7	5.45	Ven. cap.	Female, aged 37, with acute diabetes. 2 days before admission she was seized with severe abdominal pain and vomiting, which persisted till admission. Examination was made at time of admission when patient was in diabetic coma. She appeared extremely dehydrated. Blood pressure: systolic 80, diastolic 45. Urine contained large amounts of sugar and acetone
				Section 3			
26461	263	56.9	20.4	44.3	6.37	Ven. cont.	Male, aged 33, with moderately severe diabetes, examined in the morning after admission. At this time looked desiccated and had polyuria and polydypsia. Urine contained large amounts of sugar and acetone

	199	64.0	19.2	42.5	6.20	Ven. cont.	Same patient 2 days later, after insulin treatment. Has gained 5 pounds on maintenance diet only. Urine showed only trace of sugar and moderate amount of acetone
29754	234	43.9	20.1	44.0	7.93*	Ven. cont.	Male, aged 37, with severe diabetes, complicated by otitis media, examined the morning after admission. Appears emaciated and dried up. Urine contains large amounts of sugar and acetone
	113	85.0	17.7	40.7	6.66*	Ven. cont.	Same patient 2 days later, after insulin treatment. Patient has gained 5 pounds on maintenance diet only. Urine shows only trace of sugar and acetone
18467	332	59.2	15.3		6.72*	Art. cont.	Male, aged 50, with moderately severe diabetes complicated by an acute respiratory infection, examined the morning after admission. Only slight evidences of desiccation. Urine contains large amount of sugar but no acetone
	133	55.4	16.6		6.26*	Art. cont.	The same patient later in the day, after he had received 40 units of iletin
15120	449	50.6	20.1	46.2	7.14	Ven. cap.	Female, aged 57, with mild diabetes, complicated by lobar pneumonia, examined at time of admission. Semicomatose, dyspneic, with labored breathing and cyanosis. Consolidation of right lower lobe. Urine contains large amount of sugar and considerable acetone
	274	52.1	22.3	46.8	7.43	Ven. cap.	The same patient next day, after iletin treatment. Urine free from sugar and acetone. General condition seemed improved, but patient died later in the day of cardiac failure
29061	308	16.8	22.5	45.8	8.37	Ven. cont.	Female, aged 56, with mild diabetes mellitus, 48 hours before admission patient had a slight cerebral hemorrhage. The next day she became stuporous and her respirations increased. She vomited a few times. Examination of blood was made at time of admission, the night of January 28, when patient was comatose, and urine contained large amounts of sugar and acetone

TABLE 1—*Continued*

Case number	Blood sugar	Plasma CO_2	Oxygen capacity	Cell volume	Plasma proteins	Character of blood sample	Remarks
				Section 3—Continued			
	mgm. per 100 cc.	*vols. per cent*	*vols. per cent*	*vols. per cent*	*per cent*		
	142	41.6	21.0	42.4	8.07*	Ven. cont.	The same patient the next morning, January 29, after insulin and forcing fluids with sugar. Respirations quiet. Urine contains considerable sugar, but little acetone
	229	49.3	19.0	40.6	6.20*	Ven. cont.	The same patient one day later, January 30, improvement continues. Urine shows some sugar, but practically no acetone. Patient has retained considerable fluid
	241	60.8	18.4	38.2	5.85*	Ven. cont.	The same patient one week later, February 6. Urine free from sugar and acetone on an adequate diet with insulin. Consciousness has returned, but she has a definite aphasia. She also has an irregular pulse and the electrocardiograph reveals evidences of coronary disease
15670	441	30.9	13.8	27.8	5.36*	Ven. cap.	Male, aged 26, with severe diabetes, emaciated and dehydrated. Urine contains large amount of sugar and acetone. Examination at time of admission, February 12, 1923
	313	62.1	15.2	30.9	5.03*	Ven. cap.	The same patient 10 days later, February 22, much improved. He has, however, gained 37 pounds and has developed general anasarca. His urine contains a trace of sugar, but no acetone
	273	53.8	14.2			Ven. cap.	The same patient after 10 more days, March 2, has lost 27 pounds and his weight is diminishing steadily although his diet has increased. No signs of edema remain. Urine contains a trace of sugar but no acetone

15096	742	51.1	22.4	47.2	7.29	Ven. cap.	Female, aged 47, with mild diabetes and renal tuberculosis who had recently developed an influenzal pneumonia. She had been vomiting almost continuously for 4 days and had received large quantities of sodium bicarbonate. Examination made at time of admission, the night of December 11, 1922, when patient was stuporous, but breathing quietly. She complained of thirst and appeared dehydrated, but presented a slight edema of ankles. Urine contained large amounts of glucose and acetone
	216	76.5	21.3	45.1	5.77	Ven. cap.	The same patient two days later, December 13, after insulin treatment, without alkali. General condition improved, but vomiting and hiccough continue. Urine contains no sugar nor acetone
	297	54.1	18.4	40.1	6.30	Ven. cap.	The same patient one month later, January 11, 1923, shortly before discharge from hospital. Urine contains no sugar nor acetone and patient is receiving an adequate diet without insulin
22350	616	13.5	20.1		8.02	Ven. cont.	Female, aged 50, with severe diabetes. 4 days before admission patient was seized with severe abdominal pain and vomiting which persisted until admission to hospital. Examination made at time of admission the night of November 19, 1923, when patient was stuporous. She appeared extremely dehydrated. Urine contained large amounts of sugar and acetone
	344	33.7	20.1		7.45*	Ven. cont.	The same patient the next morning, November 20, after insulin treatment, saline hypodermoclysis and sugar solution by mouth. Respirations quieter, mental condition clear. Urine shows considerable sugar and moderate acetone

TABLE 1—*Concluded*

Case number	Blood sugar	Plasma CO_2	Oxygen capacity	Cell volume	Plasma proteins	Capacity of blood sample	Remarks
				Section 3—Concluded			
	mgm. per 100 cc.	*vols. per cent*	*vols. per cent*	*vols. per cent*	*per cent*		
	349	47.8	17.0		5.94*	Ven. cont.	The next morning, November 21. Improvement has continued under treatment with insulin and carbohydrate fluids. Urine contains sugar, but no acetone
	249	73.8	16.9		6.24*	Ven. cont.	6 days later, November 27. Patient receiving an adequate diet and, with insulin her urine remains free from sugar and acetone
	242	67.4	16.7		6.73	Ven. cont.	3 weeks later, December 17. Patient continues well on adequate diet with insulin and urine remains free from sugar and acetone
34611	315	41.4			6.13*	Ven. cont.	Male, aged 19, just recovering from severe diabetic toxemia precipitated by a gastric upset, with vomiting. Urine contains large amounts of sugar, acetone and diacetic acid. Patient appears dehydrated
	341	65.6			5.45*	Ven. cont.	Same patient, 5 days later, on a comparatively low diet, receiving insulin. Urine contains no sugar, acetone nor diacetic acid

and the plasma separated. The latter was then removed to a second sampling tube over mercury and from this samples were taken for analysis. The remainder of the blood was utilized for the determination of cell volume and for other purposes of no immediate importance in this connection.

For the estimation of cell volume an hematocrit of the Daland type manufactured by the International Instrument Company for their centrifuges was employed. Carbon dioxide was determined by the method of Van Slyke and Stadie in a water-jacketed machine calibrated in 0.01 cc. For the oxygen capacity a technique devised by Lundsgaard and Neill[1] which permits the saturation of the blood in the Van Slyke apparatus, was used. In order to reduce the amount of blood required the oxygen capacity was frequently determined not on the whole blood, but on the cell residue. The centrifuge tubes in which the plasma was separated were especially prepared with contracted necks. After the cells had been whirled down the upper level of the plasma was carefully marked. After the plasma had been removed the tube was filled to the mark with 0.9 per cent NaCl solution, the cells were well mixed with the saline, and the resulting emulsion was employed for the determination of oxygen capacity. Values thus obtained agree with those obtained directly from whole blood. (If water is used for the dilution instead of saline the resulting solution becomes so viscous that it is unmanageable.) For the total nitrogen of the plasma an ordinary macro-Kjeldahl technique was employed. Non-protein nitrogen was determined on the whole blood by the Folin-Wu method (7), except that distillation and titration with 0.02 N alkali was substituted for direct Nesslerization. In some instances, indicated by an asterisk in the table, non-protein nitrogen was not determined. In these cases 0.19 per cent (corresponding to 30 mg. of nitrogen per 100 cc.) was subtracted from the values of the total nitrogen in terms of protein. This was only done when there was a reasonable certainty that the blood non-protein nitrogen would prove normal. In no case can this omission introduce any significant error. It is, of course, not strictly proper to use blood non-protein nitrogen values for plasma. Presumably the latter would have proved somewhat less. The necessity for economy of blood made direct determinations on plasma impracticable.

In some of the earlier experiments no special effort to avoid venous stasis was practised in the collection of *cap.* specimens. Partly this was due to a failure to appreciate the importance of such precautions; partly because the blood was intended more particularly for other purposes and the studies of plasma proteins were only incidental. This invalidates none of the important experiments. In the earliest observations the plasma used for protein determinations was made up from all the remnants left from other work on the blood specimens and portions of it were even derived from blood that had been exposed to the air. This probably introduced no serious errors but must be taken into consideration in at least one case, no. 15096.

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[1] Personal communication.

Blood sugar was determined by the method of Folin and Wu (8) except in a few of the earlier studies when Benedict's modification of Lewis and Benedict's method (2) was used.

OBSERVATIONS

The 13 observations in the first section of table 1 are from mild and for the most part uncomplicated cases of diabetes. The majority were made before breakfast the morning after the patients were admitted to the hospital, and, therefore, while glycosuria still persisted and before dietary treatment had been well started. None of these patients showed evidences of serious dehydration or any considerable loss of weight. The plasma proteins are, in all but two of the twelve observations quite normal. The two exceptions, nos. 15733 and 20387 both had rather low proteins. The reduction in 15733 is slight, 5.98, and there were so many complicating factors in the case that it is impossible to ascribe the fault to diabetes. It is unfortunate that only one observation was made on no. 20387. The low protein was quite unexpectedly encountered in the course of an experiment in which the blood was being used for control purposes only. In reviewing the case afterwards it was found that on the day of the blood examination the patient had a transitory glycosuria and an extraordinary diuresis that persisted for 3 days. Of course it is impossible to connect these phenomena with the low proteins directly in the absence of other examinations of the same individual. It may be of some significance that the lowest proteins in this series were associated also with the lowest hemoglobin.

The 10 observations of section 2 represented single examinations of patients with diabetes of inherently greater severity or aggravated by some complicating condition. Although no. 10572 was studied on two occasions it is impossible to compare the two observations because an interval of almost a year had elapsed between them. On the first occasion he came to the hospital extremely malnourished, on a low diet. Extreme dietary reduction was necessary to render him aglycosuric and to overcome an initial acidosis. Under these circumstances, as is so often the case, the early part of his recovery was attended with the development of some edema. It was during this edematous period that the low proteins were found. On the

second occasion he had been taking insulin for some time, with excellent results, and was admitted to the hospital only because he had developed a cold and a slight pleurisy that was attended by an exacerbation of diabetic symptoms and the appearance of large amounts of sugar and acetone in the urine. With this he lost weight very rapidly and, at the time of the examination, appeared distinctly dehydrated. It will be noted that the plasma proteins are near the upper limit of normal. The hemoglobin does not, however, appear elevated.

With one other exception the values observed vary only within normal limits and the variations bear no relation to the level of blood sugar, carbon dioxide or hemoglobin. No. 18067 was an unusual case in every respect. She was admitted in diabetic coma within 48 hours after the onset of an acute attack of abdominal pain attended by persistent vomiting. No previous history of diabetes could be obtained. Blood was taken immediately after she entered the hospital when she was in deep coma, with typical respirations of acidosis. She appeared extremely desiccated, her pulse was weak and rapid and her systolic blood pressure only 80 mm. With insulin she was brought out of coma, and the respirations were temporarily quieted; but she sank back into her original state with extreme rapidity. After her admission to the hospital she passed very little urine and the administration of fluids by mouth and by hypodermoclysis resulted only in the development of edema. She died within 72 hours after her admission to the hospital.

The high hemoglobin at once suggests a concentration of the blood from loss of water and this seems reasonable in view of the vomiting and the overventilation. On the other hand concentration can offer no explanation for the low proteins. In fact no mere alteration of blood water can explain the paradoxical findings. There was no evidence of any preexisting condition that could have determined such extremely low proteins. The immediate disease might have caused a specific destruction of the plasma proteins, but the duration of symptoms was so short that this seems unlikely. It is far easier to believe that proteins had passed from the blood. White and Erlanger (19) found that in surgical shock and allied conditions, although the cellular elements of the blood became concentrated by

loss of water to the tissues, the plasma proteins fell. The low blood pressure and the clinical picture in this case is quite suggestive of "shock" and it is not surprising, therefore, to find an inspissation of the blood with low plasma proteins. If this explanation of the findings is correct the protein change can not be referred directly to the diabetes but must be ascribed to the condition that precipitated the acute crisis. No clue to this underlying condition could be found and autopsy was refused.

On each of the 9 cases in section 3 two or more studies were made at comparatively short intervals and opportunity is therefore given to study the mechanism of some of the variations that occur in different stages of the disease. The first 2 patients, nos. 26461 and 29754, had diabetes of moderate severity, in the latter case complicated by an otitis media. Both had typical diabetic symptoms, and on admission appeared dehydrated. They had been losing weight rapidly and showed marked glycosuria and ketonuria. In each case the administration of insulin and adequate amounts of fluids resulted in a striking increase in weight with a simultaneous fall in both hemoglobin and plasma proteins. In table 2 the relative plasma volumes, calculated from hemoglobin and cell volume and from plasma protein, are compared. In both instances the changes calculated by the two methods are in the same direction; in the second case they are also of the same magnitude. One can not escape the feeling that water exchange is the predominant factor behind such alterations and that the preliminary dehydration and subsequent restoration of the normal fluid content of the body are reflected in the protein values. The fourth case, no. 15120, presents a different picture. The patient, an elderly woman with diabetes and a complicating pneumonia was in a desperate state at the time of admission. Dehydration was quite noticeable and the administration of fluids by mouth difficult. Although with iletin it proved possible to overcome the glycosuria and ketonuria, the overventilation, temperature and inadequate fluid intake resulted probably in a further negative water balance which is associated with an increase of plasma proteins, hemoglobin and cell volume. The changes in nos. 29061, 15096 and 22350 seem to be of the same nature and in the first and last of these three cases the high initial values, 8.37 and 8.02 are indicative of a very definite concentration of the blood.

It is hard to avoid the feeling that these rapid alterations of plasma protein that occur in simple cases are merely expressions of changes in the state of hydration of the blood. The fact that the calculations based on hemoglobin and cell volumes do not agree exactly with those based on plasma protein values does not refute such a theory. Perfect agreement could hardly be expected in view of the profound disturbances of circulation that must accompany conditions of diabetic intoxication and may affect the number of cells circulating in the peripheral blood. Regeneration and destruction

TABLE 2

Case number	Relative plasma volume calculated from		Remarks
	O_2-capacity and cell volume	Plasma proteins	
26461	1.00	1.00	At time of admission
	1.10	1.03	2 days later
29754	1.00	1.00	At time of admission
	1.20	1.19	2 days later
15120	1.00	1.00	At time of admission
	0.89	0.96	The next morning
29061	1.00	1.00	At time of admission, January 28
	1.14	1.04	The next morning, January 29
	1.30	1.35	One day later, January 30
	1.39	1.43	One week later, February 6
15670	1.00	1.00	At time of admission
	0.87	1.07	10 days later
15096	1.00	1.00	At time of admission
	1.09	1.26	2 days later
	1.38	1.16	1 month later

of the proteins themselves must go on and may share in the general metabolic disturbance that exists. With the recognition of the importance of hydration as a factor, however, little has been gained. The production of appreciable changes in the water content of the blood in normal individuals is difficult. To effect a plasma dilution amounting to 30 per cent is almost impossible.

Widal, Abrami, Weill and Laudat (20) have noted similar changes in the concentration of the proteins in the serum of diabetic patients with acidosis after the administration of iletin and they also interpret the diminution of protein as an expression of blood dilution.

They could not, however, connect this directly with the water balance nor with the glycemia, the acidosis nor the assimilation of carbohydrates. In those of our cases in which rapid dilution occurred there is a definite retention of fluids and increase of weight as the protein falls. That the administration of fluids alone is not capable of producing such an effect is, however, well illustrated by case no. 15096 who was admitted with an edema. It is also clear from this case that edemas which occur in the course of diabetes are not invariably associated with low plasma proteins and blood dilution.

Gamble, Ross and Tisdall (9) have shown that fasting results in a loss of water from the body and that this dehydration is one of the effects of acidosis and depletion of base. The well known diuretic effects of ammonium chloride and calcium chloride have been shown by Haldane (10) to depend also on the production of an acidosis, It has repeatedly been demonstrated (5) that the edemas which occur in the course of diabetes can be rapidly delivered by the administration of these acid producing salts and that edema is prone to develop after the administration of sodium bicarbonate. Oehme (14) has made the statement that all conditions that tend towards the production of acidosis result in diuresis, and, vice versa, any increase in the alkalinity of the blood leads to a retention of fluid. At first sight it seems as if in these studies, also, dehydration was associated with acidosis and retention of fluid with the restoration of the normal alkalinity of the blood.

No. 18067 of section 2, mentioned above, is apparently an exception to the rule; but this must not be given too much weight because, as we said above, she was exceptional in every respect. When fluid balance is considered the case seems comparatively clear. From the standpoint of blood hydration, however, frequent inconsistencies appear. For instance, no. 15096, at the time of admission, had a normal plasma CO_2 and an edema; at the same time both hemoglobin and plasma proteins were high, indicating a concentration of the blood. Apparently the hydration of tissues and blood may be dissociated in diabetes just as they have been found to be in nephritis and in cardiac disease.

Although tissue dehydration is in most instances associated with acidosis and retention of fluid with the restoration of the normal

alkaline reserve, the state of hydration apparently bears no relation to the absolute level of plasma CO_2. In only one or two cases was the CO_2 of the plasma above the upper normal level and edema occurred frequently in the presence of a normal CO_2. From a clinical standpoint only, the conditions that usually result in edema in diabetes can be fairly well defined. It usually occurs in poorly nourished individuals who are receiving an inadequate diet, but show no reduction in the plasma bicarbonate. It is especially apt to develop during or just after recovery from a severe attack of diabetic toxemia. Before the advent of insulin it was more common than it is at present. The development of acidosis or the administration of acidosis producing drugs results in rapid diuresis. In the absence of such therapy the edema disappears when the diet is increased to meet the demands of the individual. This is well illustrated by case no. 15670. At the time of the second blood examination he had developed general anasarca. At that time he was receiving only 1340 calories without insulin and had moderate glycosuria. After the addition of enough insulin to clear the glycosuria, the edema disappeared as rapidly as it had come. No. 10572 delivered his edema as soon as he was given a maintenance diet and insulin.

This nutritional factor plays a part in the development of edema that appears to be independent of the acid-base equilibrium, or at least of the level of the plasma or blood carbon dioxide. It may be some of the acid-base elements other than bicarbonate that is at fault. Peters (15), Stillman, Van Slyke, Cullen and Fitz (18), Cullen and Jonas (21) and most recently Bock (3) have noted that after recovery from diabetic acidosis the plasma bicarbonate rises rapidly, while the alveolar carbon dioxide tension remains low. If the alveolar carbon dioxide is the same as that of the arterial blood such a discrepancy can only result in an uncompensated alkalosis. This Bock and his coworkers found in some of their cases. We have also noted a tendency on the part of some of these patients to develop a mild grade of uncompensated alkalosis during the period of recovery.

Gamble, Ross and Tisdall (9) suggest that the water balance is more strictly determined by the level of the alkaline metals in the body and that a retention of these elements is attended by a diminished fluid output. To test this hypothesis we have analyzed the

serum of some of the patients for total base by a method devised by Cullen (4) similar to the Fiske (6) method for the determination of total base in urine. In conjunction with this method the chlorides, carbon dioxide, inorganic phosphates and proteins have also been determined. In this manner the total inorganic anions and kations can be balanced against one another. In table 3 are presented the results obtained on one patient during a period of edema and after diuresis. The edema was very slight. There is no evidence of any change in the level of the total base of the serum in this case and an

TABLE 3

Case 34295

	Blood sugar	Oxygen capacity	Cell volume	Plasma proteins	Plasma CO_2 content	Plasma NaCl	Serum inorganic phosphate	Total inorganic acid	Total base	Difference base-acid	Remarks
	mgm. per 100 cc.	*vols. per cent*	*vols. per cent*	*per cent*	*vols. per cent*	*grams per liter*	*grams per liter*	*mM*	*mM*	*mM*	
August 23....	200	18.0	41.5	6.03*	70.3	5.89	0.035	134.0	154.3	20.4	Slight edema of ankles. Weight 147 pounds, increasing
August 30....	152	16.7		6.45*	68.0	5.90	0.037	133.2	154.9	21.7	No edema. Weight 144 pounds, diminishing pH of plasma 7.39

insignificant alteration of the total inorganic acid. If these observations are typical of diabetic edematous conditions it appears that some factor other than the acid-base balance must be active in the production of the edema. It is possible that undernutrition with its attendant loss of body protein may render these subjects more susceptible than normals to the effects of alkali. In this connection one can not but be reminded of the famine edemas that were so prominent in the War (13).

The fact that it is really the acidosis of diabetes and not the ketosis

or the other metabolic disorders associated with diabetic toxemias that determines the tissue dehydration is well illustrated in no. 15096. This patient had received large doses of alkali before admission and had also vomited large amounts of food and fluid, possibly with the loss of considerable chloride. The consequence was that in spite of the presence of an enormously high blood sugar, marked ketonuria and all the symptoms of impending diabetic coma except the hyperpnea, her plasma CO_2 content proved normal. Under these circumstances she developed an edema during the diabetic intoxication.

Although it is possible to explain the *changes* that occur in the plasma proteins over short periods of time on the basis of alterations of blood hydration in the majority of cases; in a few the facts do not seem to permit such an explanation. For example in no. 18467 the oxygen capacity rose in the course of a few hours from 15.3 to 16.6, while the proteins fell from 6.72 to 6.26. Cell volumes were not determined in this case, but it is inconceivable that the cells could have shrunk enough to account for such a discrepancy. Such occasional exceptions do not seem to us to invalidate our explanation. It would be extraordinary indeed if hemoglobin, cells and proteins were always proportionally distributed in all parts of the circulating blood. The matter can be definitely decided only by the simultaneous application of some other method for the direct determination of plasma volume.

Comparisons of proteins and hemoglobin made at intervals of more than two days in most instances show no parallelism. The methods are inapplicable to studies over long periods because of the play of the forces that determine regeneration and destruction of proteins and blood cells. In case 15096, for instance, at the time of the second examination hemoglobin, cell volume and plasma proteins had all fallen together; one month later at a third examination the hemoglobin and cell volume were still lower, but the plasma proteins had risen to the normal level. No. 22350 shows a similar course of events. One gains the impression that the high proteins found in the first examination were due to the extreme dehydration of a blood that really contained less protein than normal. Relieving the acute condition results in the restoration of the normal plasma

volume and reveals the true level of the proteins, which is low. As improvement in the general condition progresses the total amount of protein in the body increases, possibly because of an increased production. In favor of such a theory is the general nature of the con-

TABLE 4

Case 22350

	Blood sugar	Oxygen capacity	Cell volume	Plasma proteins	Plasma CO_2 content	Remarks
	mgm. per 100 cc.	*vols. per cent*	*vols. per cent*	*per cent*	*vols. per cent*	
September 6, 11 p.m.....	462				19.0	At time of admission. Patient vomiting continuously. Large amounts of glucose and acetone in urine
September 7, 9 a.m.....	370			5.06	27.7	Still vomiting continuously, but has received insulin, carbohydrate and fluids. Acetone has diminished
September 8, 9 a.m.....	233			3.22	47.0	Vomiting has ceased. Improvement continues. Fluid intake larger
September 10, 10 a.m.....	257			4.75	28.0	Vomited again this morning, with recurrence of all symptoms
September 16, 8 a.m.....	270			6.70	59.8	Greatly improved. Receiving adequate diet with large doses of insulin
September 24, 8 a.m.....	462	16.5	37.6	6.14	57.7	Improvement continues. Weight has increased steadily, without occurrence of edema. pH of plasma 7.48
October 7, 8:30 a.m.....	130	16.5	40.9	6.98	65.1	Just before discharge from hospital

ditions associated with low plasma proteins in diabetes. In this series, with the exception of no. 20387, no protein value below 6 per cent was found except in the presence of a severe diabetes with marked evidences of wasting. The same patients invariably had normal plasma proteins when the disease was under control and they

were receiving adequate diets. (See, for examples, nos. 10572, 15096 and 22350.)

Case 22350, was again admitted to the hospital recently after another attack of vomiting, in a desperate condition. The findings on the second admission are shown in table 4. The same rapid fall in the protein level during the first few days is noticeable. This time, however, the initial protein level is much lower and the subsequent depression proportionately greater. But again, after an interval and with the establishment of proper metabolic conditions the proteins rapidly rise to the normal level. The low proteins and blood concentration in this instance are quite similar to the findings in case 18067, mentioned above. It may be that both are due to the same cause and that the explanation previously advanced for 18067 is unnecessary.

It is needless to point out that without insulin these studies could have led to no more satisfactory conclusions than those of previous observers. The miraculous rapidity with which the clinical and metabolic features of a case can be altered by the use of this drug accelerates the blood changes to such an extent that blood dilution can be studied with less danger from the introduction of complicating factors.

As to the underlying cause of the low proteins in severe diabetes only speculation is as yet permissible. It does not seem absurd to connect them with the general malnutrition and protein wastage that these patients exhibit. Kerr, Hurwitz and Whipple (11) have, to be sure, shown that short periods of starvation do not affect the plasma proteins. It is, however, axiomatic that starvation does not result in nutritional disorders similar to those produced by the prolonged use of unbalanced diets. The same authors (12) have shown that the regeneration of protein after plasma depletion is retarded by inadequate diets. One is tempted to conclude that the low proteins in the plasma in severe diabetes are due to an insufficient production or regeneration which is augmented when the normal nutrition and metabolism are restored.

This again opens the question of the diabetic edemas. Usually in the presence of edema the proteins are low. That this is not invariably the case has been demonstrated. This is to be expected if

both low proteins and edema are characteristics of severe diabetes and if edema is provoked by alkalosis, which seldom exists when the blood is dehydrated.

SUMMARY AND CONCLUSIONS

In mild diabetes unassociated with ketosis or malnutrition the plasma proteins are usually normal.

In severe diabetes associated with chronic malnutrition the plasma proteins are usually found reduced. When proper nutrition and metabolic conditions are restored the plasma proteins resume the normal level.

In conditions of severe diabetic toxemia with ketosis the blood becomes concentrated by loss of water. Under these circumstances the plasma proteins appear relatively high. When the toxemia is overcome a rapid dilution of the blood and a reduction of the plasma proteins occur.

The tissue dehydration which is so striking a part of the picture of diabetic toxemias appears to be closely related to the acidosis. Restoration of the alkaline reserve is attended by retention of fluid in the body.

True diabetic edema seldom if ever occurs in the presence of acidosis. A definite alkalosis is, however, not essential to the production of edema. A nutritional factor apparently plays a part in determining the excessive accumulation of fluids, as edemas are characteristic of malnourished patients with severe diabetes who are using insufficient food, and can be eliminated by the administration and utilization of adequate diets. The fact that diuresis can be induced by the production of acidosis indicates that the acid-base equilibrium also plays its part in the etiology of the edema. It is suggested that the state of malnutrition may render the subject peculiarly susceptible to the effects of alkali.

The common association of low plasma proteins with diabetic edema is probably not one of cause and effect but is a result of the fact that both phenomena are characteristic of severe diabetes without acidosis.

BIBLIOGRAPHY

1. Austin, J. H., Cullen, G. E., Hastings, A. B., McLean, F. C., Peters, J. P., and Van Slyke, D. D. Jour. Biol. Chem., 1922, liv, 121. Studies of Gas and Electrolyte Equilibria in Blood. I. Technique for Collection and Analysis of Blood, and for Its Saturation with Gas Mixtures of Known Composition.
2. Benedict, S. R. Jour. Biol. Chem., 1918, xxxiv, 203. A Modification of the Lewis-Benedict Method for the Determination of Sugar in the Blood.
3. Bock, A. U., Field, H., Jr., and Adair, G. E. Jour. Metab. Research, 1923, lv, 27. The Acid-base Equilibrium in Diabetic Coma Being a Study of Five Cases Treated with Insulin.
4. Cullen, G. E. Personal communication.
5. Falta, W. Wien. Arch. f. inn. Med., 1923, v, 581. Ein Beitrag zum diabetischen Oedem.

   Rockwood, R., and Barrier, C. W. Arch. Int. Med., 1924, xxxiii, 643. Calcium Treatment of Edema.
6. Fiske, C. H. Jour. Biol. Chem., 1922, li, 55. A Method for the Estimation of Total Base in Urine.
7. Folin, Otto, and Wu, Hsien. Jour. Biol. Chem., 1919, xxxviii, 81. A System of Blood Analysis.
8. Folin, Otto, and Wu, Hsien. Jour. Biol. Chem., 1920, xli, 367. A System of Blood Analysis. Supplement I. A Simplified and Improved Method for the Determination of Sugar.
9. Gamble, J. L., Ross, G. S., and Tisdall, F. F. Jour. Biol. Chem., 1923, lvii, 633. The Metabolism of Fixed Base During Fasting.
10. Haldane, J. B. S., Hill, R., and Luck, J. M. Jour. Physiol., 1923, lvii, 310. Calcium Chloride Acidosis.

    Haldane, J. B. S. Lancet, 1924, i, 537. Experimental and Therapeutic Alterations of Human Tissue Alkalinity.
11. Kerr, W. J., Hurwitz, S. H., and Whipple, G. H. Amer. Jour. Physiol., 1918, xlvii, 356. Regeneration of Blood Serum Proteins. I. Influence of Fasting upon Curve of Protein Regeneration Following Plasma Depletion.
12. Kerr, W. J., Hurwitz, S. H., and Whipple, G. H. Amer. Jour. Physiol., 1918, xlvii, 370. Regeneration of Blood Serum Proteins. II. Influence of Diet upon Curve of Protein Regeneration Following Plasma Depletion.
13. Kohman, E. A. Amer. Jour. Physiol., 1920, li, 378. The Experimental Production of Edema as Related to Protein Deficiency.
14. Oehme, Court. Arch. exp. Path. Pharm. 1924, cii, 40. Der Wasser-Salzbestand des Körpers in Beziehung zum Säure- Basenhaushalt. I. Mitteilung: Beitrag zu den kolloidchemischen Grundlagen der Purindiurese.
15. Peters, J. P. Amer. Jour. Physiol., 1917, xliv, 84. The Response of the Respiratory Mechanism to Rapid Changes in the Reaction of the Blood.

16. Peters, J. P., Bulger, H. A., and Eisenman, A. J. Jour. Biol. Chem., 1923, lv, 687. Studies of the Carbon Dioxide Absorption Curve of Human Blood. I. The Apparant Variations of pK_1 in the Henderson-Hasselbalch Equation.
17. Rowe, A. H. Arch. Int. Med., 1917, xix, 354. Refractometric Studies of Serum Proteins in Nephritis, Cardiac Decompensation, Diabetes, Anemia and Other Chronic Diseases.
18. Stillman, E., Van Slyke, D. D., Cullen, G. E., and Fitz, R. Jour. Biol. Chem., 1917, xxx, 405. Studies of Acidosis. VI. The Blood, Urine and Alveolar Air in Diabetic Acidosis.
19. White, H. L., and Erlanger, Joseph. Amer. Jour. Physiol., 1920, liv, 1. The Effect on the Composition of the Blood of Maintaining and Increased Blood Volume by the Intravenous Injection of a Hypertonic Solution of Gum Acacia and Glucose in Normal, Asphyxiated and Shocked Dogs.
20. Widal, F., Abrami, P., Weill, A., and Laudat. Pressé Med., 1924, xxxii, 565. L'hydràmie au cours du diabète traité par l'insuline variations de l'indice réfractromâtrique du sérum.
21. Cullen G. E., and Jonas, L. Jour. Biol. Chem., 1923, lvii, 541. The Effect of Insulin Treatment on the Hydrogen Ion Concentration and Alkali Reserve of the Blood in Diabetic Acidosis.