During antibiotic treatment the first observation is a change in L.D.H. activity, followed about one day later by a change in D.N.A. concentration. This may indicate an initial inhibition of disintegration of cellular elements in the sputum as the first effect of the antibiotic, followed later by a reduction in the absolute number of its cellular elements as a result of successful long-term therapy.

The determination of L.D.H. activity has proved to be a valuable method of estimating the efficacy of antibiotic therapy in the treatment of infection in patients with chronic bronchitis. In the case of bacterial resistance developing during antibiotic therapy a rise in L.D.H. activity will occur before clinical deterioration, and thus allow time for a change to other appropriate treatment. The separation of L.D.H. isoenzymes was performed to establish the origin of L.D.H. in bronchial secretions. It seems unlikely that L.D.H. is of bacterial origin, since the electrophoretic migration pattern of its fractions is identical with that of human serum. In uninfected bronchial secretions the L.D.H. isoenzymes resemble those found in normal serum, and it is possible that in this case they originate from that source. However, in infected sputum with a high L.D.H. activity the pattern of L.D.H. isoenzymes corresponds to that of leucocytes and probably mucosal cells.

Summary

Determination of lactic dehydrogenase activity (L.D.H.) in bronchial secretions is a simple method of differentiating between infected and uninfected chronic bronchitics. The values obtained correlate well with the results of the quantitative determination of deoxyribonucleic acid (D.N.A.) in the secretions. The D.N.A. content of sputum can easily be estimated semiquantitatively by fluorescent microscopy, and the results obtained by this method compare well with quantitative D.N.A. estimations.

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Renal Involvement in Human Leptospirosis

VISITH SITPRIJA,* M.D., PH.D.

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Renal involvement is common in leptospirosis. Azotaemia and proteinuria with abnormal urinary sediment are frequently noted. While definite pathological changes of acute tubular necrosis and interstitial nephritis have been described (Gsell, 1952, Areán, 1962), it has been stated that temporary azotaemia and oliguria may occur in the absence of significant renal damage (Edwards, 1959). Clinical improvement after intravenous fluid administration, along with the increase in urinary output, suggests that azotaemia is secondary to deficiency in volume. In one report the decreased renal function was mentioned (Cora, 1956). The glomerular filtration rate was markedly reduced, and the excretion of para-aminohippurate (P.A.H.) was almost negligible. Because of the discrepancy in the clinical data this investigation was undertaken to clarify the clinical patterns of renal function disturbances encountered in leptospirosis.

Materials and Methods

A study was made of 10 patients aged from 24 to 63 years, with leptospirosis proved by haemoculture or serology, whose blood urea nitrogen was over 40 mg./100 ml. Eight were males and two were females. In all cases the causative micro-organisms were *Leptospira bataviae*. They were admitted to hospital on the fifth to the seventh day of illness. Immediately after admission the blood volume was measured by the summation method with both ¹³¹I-labelled albumin and ⁵¹Cr-tagged red blood cells (Moore et al., 1963). Endogenous creatinine and P.A.H. clearances were ascertained by the standard technique (Smith, 1956) before antibiotic therapy. Blood chemistry, including blood urea nitrogen (B.U.N.), serum creatinine, electrolytes, and osmolality, was determined every other day until

the return to normal. Urinalysis was performed at two-day intervals until no abnormal finding was noted. Twenty-four-hour urine was collected on alternate days for determination of urea nitrogen, creatinine, sodium, and osmolality.

Results

Clinical Observations.—Body temperature ranged from 37.3 to 39.2° C., and usually subsided three to four days after penicillin administration. All patients had stable vital signs. Jaundice was present in five patients whose total bilirubin varied from 5 to 24 mg./100 ml. The liver-function test showed only a minimal degree of parenchymal hepatic damage. The patients are classified into two groups according to the serum creatinine. Group 1 comprises five patients with levels above 2 mg./100 ml.; the other five (group 2) had levels below 2 mg./100 ml. The pertinent clinical data are shown in Table I.

Blood Chemistry.—Serum sodium and potassium were within normal limits. Six patients had a CO₂ content below 20 mEq/l., probably indicating metabolic acidosis. Five of these patients were in group 1. The average B.U.N. and creatinine in group 1 were 105.2 and 4.9 mg./100 ml. respectively, in contrast with the values of 54.2 and 1.6 mg. in group 2 (Table I). The association between jaundice and impaired renal function was striking in group 1, and this might reflect the more severe form of infection. The elevated B.U.N. and creatinine returned to normal within 7 to 10 days in group 1 and within 24 to 48 hours in group 2.

Urinary Output and Sediment.—The urine volume on the first hospital day ranged from 180 to 890 ml./24 hours, with an average of 485 ml., in group 1. There was only a slight increase in urine flow after fluid load during the clearance study. In group 2 the urinary output varied from 235 to 700 ml./24 hours, with a mean of 463 ml. (Table I). After

* Senior Lecturer in Medicine, Chulalongkorn Hospital Medical School, Bangkok, Thailand.

TABLE I.—Clinical and Laboratory Data

Case No.	Fever	Jaundice	Plasma						Urine					
			B.U.N.	Creatinine	Na	K	Cl	CO ₂ Content	Volume (ml./24 hr)	Sp. Gr.	Protein	Cast	R.B.C.*	W.B.C.†
			mg./100 ml.		mEq/l.									
Group 1														
1	38.7	+	105	4.4	139	3.6	98	12.2	180	1007	1+	G	4-5	8-9
2	38.2	0	140	4.0	140	3.9	100	18.9	400	1006	2+	G	0	5-6
3	37.3	+	106	2.9	139	3.3	93	17.0	510	1010	0	O	4-6	5-8
4	38.7	+	72	4.2	135	4.1	95	16.8	890	1011	1+	G	1-2	4-6
5	39.0	+	103	9.0	140	3.2	102	9.0	445	1008	1+	O	0	4-5
Average	38.4		105.2	4.9	138.6	3.6	97.6	14.8	485	1008				
Group 2														
6	39.2	0	72	1.4	143	3.4	104	20.0	400	1016	1+	G	2-4	2-3
7	38.2	0	56	1.2	142	3.7	93	18.0	370	1018	0	O	0	0
8	38.5	0	45	1.6	142	3.1	98	21.5	700	1020	2+	G	8-9	6-10
9	38.6	+	50	1.9	139	4.7	101	24.2	610	1018	1+	G	0	3-5
10	39.1	0	48	1.6	140	4.0	96	22.0	235	1022	Trace	G	2-3	5-7
Average	38.7		54.2	1.6	141.2	3.8	98.4	21.2	463	1019				

G = Granular cast. * Red blood cells per high power field. † White blood cells per high power field.

fluid load during the clearance test the urine flow rose to approximately 2 ml./minute. In both groups there was a gradual rise in urinary output during the hospital course; the maximum amount, however, did not exceed 2,500 ml./24 hours. There was no difference in urinary sediment between the two groups of patients. Proteinuria was present in eight cases, and varied from a trace to 2+. The numbers of white blood cells and red blood cells were variable; granular casts were noted in seven cases. Urinalysis became negative four to seven days after admission. The mean urinary specific gravity was 1008 and 1019 in groups 1 and 2 respectively.

Urine Sodium and Osmolality.—The urine sodium varied from 40 to 86 mEq/l., with a mean of 62.2 mEq/l., in group 1; the range in group 2 was between 15 and 60 mEq/l., with an average of 35 mEq/l. The sodium excretion, expressed as a percentage of filtered sodium, averaged 4.63 for group 1 and 0.57 for group 2, indicating decreased sodium reabsorption in group 1 (Table II). Though the urine concentration test was not attempted, the osmotic urine and plasma ratio (Uosm/Posm) above 2 in group 2 indicates good tubular reabsorption of water. In group 1 this ratio ranged from 1.09 to 1.34, suggestive of a defect in concentration (Table II).

TABLE II.—Blood Volume and Clearance Data

Case No.	Blood Volume (ml./kg.)	Clearance (ml./min.)		C _{Cr} / C _{P.A.H.}	Urine Na (mEq/L.)	Na Excretion at Percentage of Filtered Na	Uosm/Posm
		Creatinine	P.A.H.				
Group 1							
1	56.4	8.0	32.3	0.25	86	6.60	1.23
2	69.2	6.2	31.0	0.20	70	6.40	1.09
3	60.4	18.3	31.2	0.59	44	1.53	1.25
4	—	11.2	35.4	0.32	71	2.86	1.30
5	71.2	5.2	30.4	0.17	40	5.77	1.34
Average	64.3	9.8	32.1	0.31	62	4.63	1.24
Group 2							
6	60.1	43.2	212.5	0.20	60	0.96	2.52
7	51.2	54.0	284.3	0.19	15	0.33	2.24
8	52.5	50.0	262.6	0.19	27	0.34	2.11
9	—	32.5	174.7	0.19	19	0.86	2.45
10	—	43.9	232.3	0.19	54	0.87	2.41
Average	54.6	44.7	233.3	0.19	35	0.57	2.35

C_{Cr}/C_{P.A.H.} = Creatinine and P.A.H. clearance ratio.

Blood Volume.—This was determined in seven patients—four in group 1 and three in group 2. The values in the first group varied from 56.4 to 71.2 ml./kg. body weight, with a mean of 64.3 ml./kg. In the second group it was significantly lower, the range being from 51.2 to 60.1 ml./kg., with an average of 54.6 ml./kg., consistent with volume deficit (Table II).

Clearance Study.—There was a decrease in both endogenous creatinine and P.A.H. clearances in both groups. The decrease was more pronounced in group 1, having an average clearance

of 9.8 and 32.1 ml./min. for creatinine and P.A.H. respectively; in group 2 the averages were 44.7 and 233.3 ml./min. The creatinine and P.A.H. clearance ratio averaged 0.31 in group 1 and 0.19 in group 2, indicating disproportional reduction in P.A.H. clearance in group 1 (Table II). The repeat clearances two to three weeks after the first determination showed values that fell within normal limits except in Case 1. This 63-year-old patient had at the time of discharge from hospital creatinine and P.A.H. clearances of 51 and 200 ml./min.

Discussion

The serum creatinine and renal function study shows a distinct difference between the two groups. There was marked reduction of renal clearances in group 1 associated with impaired tubular reabsorption of sodium and water. In group 2 the tubular function pertaining to salt and water reabsorption was retained. The laboratory data in group 2, which showed the decrease in blood volume, Uosm/Posm above 2, and low urine sodium excretion, suggest that volume deficit or dehydration was responsible for azotaemia. Though dehydration may not be detected by clinical methods, the response to fluid load during the clearance study and the increase in urine flow and the rapid fall of B.U.N. support its role in causing oliguria and nitrogen retention. Increased protein catabolism due to fever and decreased caloric intake could also contribute to the development of azotaemia.

The laboratory data in group 1, characterized by Uosm/Posm below 1.35 and the relatively high urine sodium, are compatible with parenchymal renal damage seen in acute tubular necrosis (Eliahou and Bata, 1965; Meroney and Rubini, 1959). Acute tubular necrosis has been described in leptospirosis by Gsell (1952) and Areán (1962). Though the validity of the decreased P.A.H. clearance is questioned in the presence of nitrogen retention and impaired renal function because of decreased P.A.H. extraction (Cargill, 1949; Bradley *et al.*, 1950), the possibility of its decrease from renal tubular damage or reduced renal plasma flow cannot be excluded.

It is interesting that the diuretic phase as seen in acute tubular necrosis was not evident in these patients. The urinary output never exceeded 2,500 ml./24 hours during the period of recovery. It is not clear whether this is characteristic of renal failure in leptospirosis or merely the result of over-hydration during the acute phase. This type of gradual increase in urine flow is often noted in the clinical course of acute glomerulonephritis. The clinical and laboratory data, however, do not support this diagnosis. Leptospirosis has never been cited as a cause of acute glomerulonephritis. In this aspect renal failure in leptospirosis resembles that of acute malaria (Sitprija *et al.*, 1967; Sitprija, 1968), and might have the same pathogenetic mechanisms.

The precise cause of acute renal failure in leptospirosis is not well understood. Bacterial toxin and renal anoxia secondary to haemodynamic changes are among the possibilities thought to account for renal failure (Areán, 1962). Since there was a significant association between renal failure and jaundice in group 1, impaired renal function is evaluated in light of the hepatorenal syndrome. In this syndrome renal failure is attributed to renal haemodynamic changes secondary to liver failure. Though jaundice was intense in this group, liver-function tests showed only slight impairment in function. The striking contrast between the severity of clinical manifestations and the slight deviation of liver function in leptospirosis is well appreciated (Areán and Henry, 1964). In the absence of liver failure the hepatorenal syndrome seems unlikely. Finally, the possibility of the toxic effect of bile on the kidney is raised. It is not known to what extent bilirubin plays a part in renal function. Recently it has been shown that intravenous injection of bilirubin in dogs results in a decrease in tubular reabsorption of sodium (Topuzlu and Stahl, 1966). As some patients in the report of Stiles *et al.* (1946) and one in this report (Case 2) developed renal failure without jaundice, it would seem unlikely that impaired renal function is secondary to jaundice.

Because of a tendency towards bleeding during the acute period of illness renal biopsy was not attempted. In a series of renal biopsies performed late in the course of disease the pathological changes were suggestive of a mild degree of acute tubular necrosis and interstitial nephritis (Penna *et al.*, 1963). The results of renal function study in group 1 support the above pathological findings. The study by electronmicroscopy showed focal thickening of basement membrane, fusion of foot processes, loss of brush border, and mitochondrial depletion of renal tubules (DeBrito *et al.*, 1965).

Since dehydration very often cannot be clinically detected, intravenous fluid therapy may be dangerous when the cause of azotaemia is not known. From this study, for practical purposes, fluid administration in azotaemic patients with leptospirosis should be based on the serum creatinine and Uosm/Posm. When the serum creatinine is above 2 mg./100 ml. and Uosm/Posm is close to 1 azotaemia is most likely due to acute tubular necrosis, and excessive fluid therapy should be avoided. Conversely, when the serum creatinine is below 2 mg./100 ml.

and Uosm/Posm approximates 2 dehydration should be suspected, and fluid load would be the treatment of choice.

Summary

Renal function was studied in 10 azotaemic patients with proved leptospirosis. In five patients azotaemia was ascribed to parenchymal renal failure of acute tubular necrosis. In this group the serum creatinine was higher than 2 mg./100 ml. and the osmotic urine and plasma ratio averaged 1.24, indicating a defect in renal concentration. In another group of five patients the renal function study suggested that volume deficit or dehydration might be the cause of azotaemia. The serum creatinine was lower than 2 mg./100 ml. and the mean osmotic urine and plasma ratio was 2.35. In both groups there was a decrease in endogenous creatinine and para-aminohippurate clearances; the decrease was more pronounced in group 1. Because of the clinical inaccuracy in detecting dehydration the serum creatinine and the osmotic urine and plasma ratio may be used as a clue in ascertaining the cause of azotaemia, and thus provide useful information regarding the fluid therapy of azotaemic patients with leptospirosis.

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Fibrinolytic Response to Moderate Exercise in 50 Healthy Middle-aged Subjects

J. D. CASH,* M.B., CH.B., B.SC., PH.D., M.R.C.P.ED.; D. G. WOODFIELD,† M.B., CH.B., M.R.C.P.ED.

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Previous studies reported from this laboratory on the fibrinolytic response to a short period of moderate exercise in a group of normal young subjects showed a small subgroup whose response to this procedure was poor (Cash, 1966). A similar pattern was also demonstrated when intravenous adrenaline was used as a fibrinolytic stimulant, so that a poor responder to moderate exercise was also a poor responder to intravenous adrenaline (Cash and Allan, 1967). These results led us to conclude that one interpretation of this phenomenon could be a fundamental defect in the mechanisms related to the release of plasminogen activator in an acute stressful situation, and that the subgroup which displayed this anomaly might be at risk to conditions such as atherosclerosis, thrombosis, and

irreversible shock in which defective fibrinolysis has been postulated to be an important aetiological factor (Astrup, 1956; Hardaway, 1966).

The original exercise observations were made on a group of 50 young healthy subjects aged 18-30 (mean 22.3) years. The following communication describes the results from a group of 50 healthy middle-aged volunteers, obtained during the period in which the young subjects were studied, and also a limited 12-month follow-up study on 18 selected middle-aged subjects.

Subjects

The subjects were 25 male and 25 female blood donors aged 42-57 (mean 49.6) years. All agreed to give a detailed medical history and submit themselves to a full clinical examination, which included a resting (supine 15 minutes) blood pressure (by

* Senior Registrar.

† Registrar.

South-east Scotland Regional Blood Transfusion Research Laboratories, Royal Infirmary, Edinburgh 3.