Effect of corticosteroid therapy on bromsulphthalein excretion in active chronic hepatitis

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The aetiology of active chronic hepatitis or lupoid hepatitis (Saint, King, Joske, and Finckh, 1953; Bearn, Kunkel, and Slater, 1956; Mackay, Taft, and Cowling, 1956; Bartholomew, Cain, Baggenstoss, and Hagedorn, 1960; Read, Sherlock, and Harrison, 1963; MacLachlan, Rodnan, Cooper, and Fennell, 1965) is not completely understood. It is a stage in a continuous process beginning with an acute hepatic insult, which may sometimes be virus hepatitis, and ending in post-necrotic cirrhosis (Bartholomew, Hagedorn, Cain, and Baggenstoss, 1958; Page and Good, 1960; Jones and Castleman, 1962). The place of corticosteroids in both short- and long-term treatment is undecided. The present report concerns bromsulphthalein (BSP) excretion and metabolism in patients in a controlled trial of this treatment which is currently in progress at the Royal Free Hospital. Additional studies in other patients with active chronic hepatitis who were not in the trial, and in patients with cholestatic virus hepatitis and extrahepatic obstructive jaundice are also reported.

PATIENTS

In all patients the diagnosis was confirmed by serum biochemistry and hepatic histology obtained by aspiration biopsy specimens (Read *et al.*, 1963).

Twenty-two patients with active chronic hepatitis who had not previously received corticosteroids or ACTH had bromsulphthalein (BSP) disappearance curves determined. They were in the controlled trial, and were divided into two random groups by a blind technique using sealed envelopes. One group received 15 mg prednisolone daily indefinitely; the second was not treated with corticosteroids. Other treatment, *ie*, diuretics, and supportive measures, was similar in each group. Tables I and II give the sex, age, and initial BSP and biochemical indices in the 22 patients in the two groups. The mean time between inclusion to the trial and the last BSP estimation was 32 and 28 weeks in the two groups respectively.

In an attempt to elucidate the mechanism of the alter-

ation in BSP retention produced in the patients in the controlled trial by corticosteroids, BSP storage (S), and transport maximum (Tm) estimations were done in two further groups of patients with active chronic hepatitis. Twenty-four patients with active chronic hepatitis had a single BSP storage (S) and transport maximum (Tm) determination; they included patients 1, 2, 4, and 12 in the controlled trial (Table III). Fourteen of them were receiving 10 to 30 mg prednisolone daily; 10 were not having corticosteroids. In 29 other patients with active chronic hepatitis who were not all in the therapeutic trial, and were not therefore strictly randomly selected, serial BSP storage (S) and transport maximum (Tm) estimations were made; they included patients 7, 8, 9, 10, 15, 17, 19, 20, 21, and 22, who were studied in the controlled trial. Sixteen had received corticosteroids or ACTH in the past, but not for at least 12 months. In 16, estimations were performed before and after starting 15 mg prednisolone daily (Table IV); the other 13 acted as controls (Table V).

Storage (S) and transport maximum (Tm) for BSP were estimated in a further four patients—three with cholestatic virus hepatitis and one with extrahepatic obstructive jaundice—in order to obtain additional information about the mechanism of the alteration in BSP excretion during corticosteroid treatment (Table VI). Three of them were given 30 mg prednisolone daily.

In 21 of the patients with active chronic hepatitis, nine of whom were in the controlled trial (nos. 7, 8, 9, 10, 17, 19, 20, 21, and 22), plasma samples during BSP disappearance curves were subjected to chromatographic analysis of BSP. Twelve had estimations before and after starting 15 mg prednisolone daily; nine acted as controls (Tables VII and VIII).

METHODS

Bromsulphthalein (BSP) disappearance curves were performed after an overnight fast following the intravenous injection of 5.0 mg BSP per kg body weight. Venous blood was taken from the opposite arm at three, eight, 12, 20, 30, and 45 minutes after injection of BSP. Plasma BSP was estimated on the same day by a method based on that of Seligson, Marino, and Dodson (1957). To 3.5 ml alkaline buffer (pH 10.6) (12.2 g Na₂HPO₄·7H₂O, 1.77 g Na₃PO₄·H₂O, and 3.20 g of sodium *p*-toluene sulphonate, made up to 500 ml with distilled water), 0.5 ml plasma was added and the colour estimated with a Unicam spectrophotometer at 580 mμ. After the

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addition of 0.1 ml sodium dihydrogen phosphate (69 g $\text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$ to 250 ml with distilled water), a second reading was obtained and subtracted from the first. Variation in turbidity, depth of jaundice, and mild haemolysis were thus eliminated by this method, but obviously haemolysed samples were discarded. Standard BSP solutions were treated as the test plasma. By extrapolation to zero on semi-log paper using three, eight, and 12-minute readings, the initial venous BSP level was obtained. Percentage retention at 30 minutes, disappearance half time ($t_{1/2}$), and plasma disappearance rate (PDR) were determined, the latter from the formula:

$$\text{PDR} = \frac{0.693}{t_{1/2}} \times 100$$

Relative hepatic storage (S) and transport maximum (Tm) for BSP were estimated by the method of Wheeler, Meltzer, and Bradley (1960). By this indirect technique, observations of biliary and hepatic venous BSP levels are unnecessary. Tests were performed after an overnight fast. Infusion of BSP was with a constant infusion pump (Bowman Process and Instruments, 15 Stone Avenue, Brooklyn, New York). Intravenous Braunule size 1 (Armour) needles were used for infusion. Two infusions of constant rate were given (Adams, Gordon, and Combes, 1966) (Tables IV to VI). These were continued for at least 30 minutes before venous sampling was started; this consisted of four or five samples taken from the opposite arm over the next 20-30 minutes. When serial estimations were performed in the same patient, the rates of infusion were as far as possible identical on the two occasions. The rates of infusion were calculated to maintain an approximately constant level of plasma BSP. From the rates of change of plasma BSP and an estimated plasma volume, hepatic storage (S) (mg BSP stored/mg liver tissue/100 ml plasma BSP) and transport maximum (Tm) (mg BSP excreted/minute) were calculated from the simultaneous equations:

$$\begin{aligned} I_1 &= \text{Tm} + \Delta P_1 (S + \text{PV}) \\ I_2 &= \text{Tm} + \Delta P_2 (S + \text{PV}) \end{aligned}$$

where I_1 and I_2 are the infusion rates of BSP, ΔP_1 and ΔP_2 the rates of change of plasma BSP during the two infusions, and PV the plasma volume, calculated from the formula (Wheeler *et al.*, 1960):

$$\text{PV (ml)} = 41.2 \times \text{body weight in kg}$$

Chromatographic studies of plasma BSP were made on samples taken at three, eight, 12, 20, 30, and 45 minutes after the intravenous injection of BSP by a method based on that of Javitt, Wheeler, Baker, Ramos, and Bradley (1960). Two ml plasma samples were deproteinized by mixing with 8.0 ml acetone and centrifugation. The supernatant fluid was separated and slowly evaporated to dryness. The residue after the addition of 0.1 ml distilled water was subjected to ascending chromatography on Whatman no. 4 paper in a solvent system of *n*-butanol: glacial acetic acid: distilled water at a ratio 4:1:2. Chromatograms were rapidly dried, sprayed with 20% potassium carbonate, and photographed. A fast moving component was repeatedly shown to be identical with commercially produced BSP. Several slower moving components were conjugates of BSP with glutathione and to a lesser extent glycine, glutamic acid, and cysteine

(Grodsky, Carbone, and Fanska, 1959; Javitt, Wheeler, Baker, and Ramos, 1959). The spots were separately eluted with the alkaline buffer used for the plasma BSP estimation and the relative concentrations of conjugated and unconjugated BSP were obtained. Absolute values for conjugated and unconjugated BSP were obtained by reference to total plasma BSP.

Total and conjugated bilirubin were estimated by a modification of the method of Michaëlsson (1961), serum aspartate transaminase by that of Reitman and Frankel (1957), and alkaline phosphatase by that of Bell and Collier (1964). Serum proteins (albumin and globulin) were measured by the method of Wolfson, Cohn, Calvary, and Ichiba (1948). The biochemical tests were done on the same day as one or more of the BSP estimations.

Hepatic histology was assessed in all patients from aspiration biopsy specimens. 'Activity', which was based on the severity of necrosis and inflammatory cell infiltration, was divided into four grades, namely, + + +, + +, + and 0.

RESULTS

BROMSULPHTHALEIN DISAPPEARANCE CURVES IN PATIENTS WITH ACTIVE CHRONIC HEPATITIS IN THE CONTROLLED TRIAL Tables I and II give results of BSP disappearance following intravenous injection in the patients in the controlled trial. In the corticosteroid group, liver histology from four patients was graded + + + and from six + +; eight had well developed cirrhosis. In the control group six were graded + + + and six + +; 10 had cirrhosis. Expressed as BSP retention at 30 minutes, nine patients in the corticosteroid group (mean 38.4%) and all of those in the control group (mean 42.9%) had abnormal results before inclusion in the trial (Fig. 1). Expressed as plasma disappearance rate (PDR) only two patients, in the corticosteroid group, had normal results (Schoenfield, McGill, and Foulk, 1964); the mean values were 5.4 (2.8-12.6) %/min and 4.3 (2.5-8.7) %/min respectively. In the treated group, all except one patient had a decrease in BSP retention after starting corticosteroids. Six control patients showed a decrease and six an increase in retention. The mean reduction in the 30 minutes' BSP retention when the first and last measurements were compared was significantly greater in the corticosteroid than in the control group ($P < 0.02$). There was not a significant correlation between change in BSP retention and the degree of histological activity or presence of cirrhosis. The difference between the mean increase in PDR was not significant ($P < 0.20$). Plasma BSP at zero time was estimated by extrapolation of the disappearance curve on semi-log paper (Tables I and II). Mean values were 8.3 mg/100 ml in the corticosteroid and 7.7 mg/100 ml in the control group. At the last BSP determination this value was higher

TABLE I

BROMSULPHTHALEIN (BSP) DISAPPEARANCE AFTER AN INTRAVENOUS INJECTION OF 5.0 mg/kg BODY WEIGHT IN THE 'CORTICOSTEROID' GROUP (OF THE CONTROLLED TRIAL) AND BIOCHEMICAL INDICES

Case No.	Sex	Age (yr)	Time after Inclusion in Trial ¹ (weeks)	BSP Retention at 30 Minutes (% of initial dose)	Plasma Disappearance Rate (PDR) (%/min)	Plasma BSP Level at Zero Time (mg/100 ml)	Serum Bilirubin (mg/100 ml)	Serum Aspartate Transaminase (AST) (iu/litre) ²	Alkaline Phosphatase (KA units/100 ml)	Serum Albumin (g/100 ml)	Serum Total Globulin (g/100 ml)
1	F	46	0	48.3	2.9	11.7	4.2	92	15	2.3	4.7
			2	43.1	4.1	10.6					
			4	30.6	5.3	10.7					
			6	31.5	6.3	8.8					
			36	22.6	6.3	8.0					
2	M	15	76	18.2	6.9	9.5	0.5	23	12	3.5	4.2
			0	46.1	3.2	6.4	3.3	172	30	3.6	7.4
			1	40.1	4.1	7.8					
			5	40.8	5.3	7.6					
			13	20.5	8.7	8.7	1.6	35	19	3.8	5.7
3	F	8	0	5.0	12.6	9.0	1.2	64	22	4.1	4.4
			51	3.9	13.9	8.4	0.5	6	19	5.2	3.4
4	F	51	0	52.0	2.8	8.8	5.5	45	31	2.9	4.2
			3	48.0	4.3	7.4					
			7	52.3	3.2	6.5					
			11	50.7	3.7	7.3					
			35	50.0	3.2	8.4					
5	F	50	67	59.2	2.2	8.3	22.0	70	38	3.3	3.9
			0	53.5	2.8	10.2	2.5	130	32	2.2	5.6
			6	43.7	3.5	10.4					
			10	34.4	5.3	10.8					
			26	16.6	8.1	11.0	0.5	25	11	3.6	2.5
6	F	56	0	51.4	3.1	7.2	1.8	52	20	2.5	6.5
			4	28.9	6.3	9.0					
			12	34.2	4.8	8.8					
			54	30.1	7.3	12.0	0.8	15	14	4.0	4.9
7	M	51	0	37.8	5.8	7.9	2.0	51	44	2.7	5.9
			0	41.7	4.9	8.1					
			4	22.0	7.7	8.3	1.4	27	40	3.6	6.0
8	F	23	0	38.0	5.5	7.5	2.2	160	47	4.0	4.6
			3	20.8	9.2	7.3					
			11	16.3	8.7	7.0	0.5	40	24	3.3	4.6
9	F	15	0	30.1	5.3	7.5	1.7	500	6	2.9	7.7
			16	19.7	6.9	9.5	0.7	25	8	3.5	5.3
10	F	51	0	22.3	9.9	6.6	1.5	35	70	2.2	7.4
			2	22.6	9.9	7.0					
			6	20.8	10.7	8.8	1.0	28	75	2.5	5.0

¹For each patient, the first estimation was done immediately before starting corticosteroids.

²Normal range <17 i.u./litre.

than the first in six patients in the corticosteroid and 10 in the control group; the difference is not significant. Serum bilirubin, aspartate transaminase, alkaline phosphatase, and albumin and globulin values at the initial and last BSP estimation are shown in Tables I and II. The correlation coefficient (*r*) between change in BSP retention and change in serum bilirubin was + 0.456 ($P < 0.05$); that between BSP retention and serum aspartate transaminase + 0.514 ($P < 0.02$); and that between BSP retention and serum total globulin + 0.597 ($P < 0.01$). Changes in BSP retention in the patients in both

groups therefore occurred *pari passu* with changes in the biochemical indices of 'activity' of the hepatic lesion.

STORAGE (S) AND TRANSPORT MAXIMUM (T_m) IN PATIENTS WITH ACTIVE CHRONIC HEPATITIS Table III summarizes the results of single S and T_m estimations in 24 patients with active chronic hepatitis. Fourteen were receiving corticosteroids and 10 were not being treated. The degree of histological activity in the patients receiving prednisolone was +++ in four, ++ in six, and + in four; all except one had

TABLE II

BROMSULPHTHALEIN (BSP) DISAPPEARANCE AFTER AN INTRAVENOUS INJECTION OF 5.0 mg/kg BODY WEIGHT IN THE 'CONTROL' GROUP (OF THE CONTROLLED TRIAL) AND BIOCHEMICAL INDICES

Case No.	Sex	Age (yr)	Time after Inclusion in Controlled Trial (weeks)	BSP Retention at 30 Minutes (% of initial dose)	Plasma Disappearance Rate (PDR) (%/min)	Plasma BSP Level at Zero Time (mg/100 ml)	Serum Bilirubin (mg/100 ml)	Serum Transaminase (AST) (iu/litre)	Alkaline Phosphatase (KA) (units/100 ml)	Serum Albumin (g/100 ml)	Serum Total Globulin (g/100 ml)
11	M	24	0	11.9	8.7	9.6	2.0	116	16	4.2	3.8
			10	7.8	10.7	10.0					
			23	5.0	17.3	10.0	0.3	15	32	4.4	3.1
12	F	47	0	25.5	6.3	7.7	0.7	34	11	3.1	5.4
			2	20.2	6.9	8.9					
			14	13.9	7.7	9.0					
			45	11.9	9.2	9.1	0.5	19	14	4.9	2.8
13	F	15	0	51.2	3.5	8.1	5.1	195	27	2.2	9.2
			4	55.8	2.5	8.5	3.8	230	28	2.6	9.4
14	F	61	0	43.6	3.5	8.6	7.0	170	34	1.3	6.6
			6	49.5	3.1	9.1	8.0	290	23	1.4	7.2
15	M	31	0	37.9	5.8	6.6	5.5	115	120	2.5	6.2
			11	45.3	4.2	6.8					
			29	45.1	4.0	6.2					
			49	42.0	4.8	6.1					
			78	46.2	4.6	7.9	4.4	94	70	2.5	7.0
16	F	27	0	45.3	3.4	7.8	1.0	44	29	2.5	4.3
			43	43.7	4.2	8.7	2.0	26	30	2.6	4.5
17	F	11	0	55.1	3.1	6.3	4.5	95	44	3.0	6.7
			56	41.9	4.3	7.4	2.6	65	40	2.4	6.1
18	M	44	0	49.4	3.1	8.5	2.3	110	33	2.1	4.2
			9	55.3	2.6	7.5	2.3	135	23	2.2	7.3
19	F	50	0	51.7	3.1	8.5	11.0	310	45	3.3	5.1
			4	52.8	2.9	9.0					
			10	48.3	3.3	10.2					
			24	29.4	5.1	12.6	0.6	19	12	4.3	3.7
20	F	48	0	51.4	3.1	7.0	2.1	21	36	3.8	3.9
			5	48.9	3.5	8.4					
			16	51.3	3.1	7.2					
			20	47.4	3.4	6.6					
			29	52.8	2.7	7.1	2.3	26	46	2.2	3.7
21	F	57	0	33.5	5.8	8.3	2.6	49	46	3.0	5.6
			8	34.6	5.1	7.8	2.2	45	54	3.4	4.5
22	M	39	0	58.9	2.5	5.5	3.9	61	17	3.0	5.7
			8	56.5	3.1	7.9	5.2	151	17	2.2	5.6

TABLE III

SUMMARY OF THE MEAN STORAGE (S) AND TRANSPORT MAXIMUM (Tm) FOR BSP AND BIOCHEMICAL INDICES IN PATIENTS WITH ACTIVE CHRONIC HEPATITIS WITH ONLY ONE ESTIMATION

Group	No. Studied	Sex M	Sex F	Age (yr) (Mean and Range)	Storage (S) (mg/mg/100 ml)	Transport Maximum (Tm) (mg/min)	S/Tm	Serum Bilirubin (mg/100 ml)	Serum Aspartate Transaminase (iu/litre)	Serum Alkaline Phosphatase (KA) (units/100 ml)	Serum Albumin (g/100 ml)	Serum Total Globulin (g/100 ml)
Receiving corticosteroids at time of test	14	5	9	31 (10-60)	30.6 (9.0-67.8)	4.3 (2.2-7.3)	7.8 (2.6-14.1)	3.2 (0.5-22.5)	45 (10-108)	22.7 (5-67)	3.8 (2.9-4.6)	3.9 (2.6-5.4)
No corticosteroids	10	2	8	49 (11-74)	40.2 (16.7-69.1)	3.9 (2.6-6.2)	10.7 (5.8-21.7)	1.3 (0.5-2.3)	30 (10-52)	26.3 (8-79)	3.2 (1.7-4.3)	4.2 (2.0-6.2)

FIG. 1. Initial bromsulphthalein (BSP) retention at 30 minutes and plasma disappearance rate (PDR) after an intravenous injection of 5.0 mg per kg body weight in two randomly selected groups of patients with active chronic hepatitis before inclusion in the controlled therapeutic trial. The hatched areas indicate normal ranges.

cirrhosis. In the untreated group, one patient was graded +++, four ++, and five +; all except one had cirrhosis. The differences between the mean storage (S), transport maximum (Tm) and S/Tm ratio between the two groups is not significant. There is a significant correlation between storage (S) and serum aspartate transaminase ($r = -0.545$; $P < 0.01$) in the individuals of both groups. The correlation between storage (S) and serum bilirubin ($r = -0.224$), and storage and serum total globulin ($r = -0.116$) is not significant. The correlation between transport maximum (Tm) and serum alkaline phosphatase ($r = -0.558$) is significant ($P < 0.01$).

Tables IV and V summarize results of serial estimations of BSP storage and transport maximum. Results for initial S and Tm values are compared in Figure 2. Mean levels for S were 26.5 (5.4-50.0) mg/mg/100 ml in the group subsequently treated with prednisolone, and 35.0 (21.8-58.0) mg/mg/

100 ml in the control group. Those for Tm were 3.9 (2.2-7.0) mg/min and 4.8 (1.4-11.2) mg/min respectively. Mean S/Tm ratios were 7.8 (1.3-18.5) and 9.3 (3.3-18.3). The change in storage (S) between the first and last determinations in the two groups is shown in Fig. 3; the mean increase in S is significantly greater in the group treated with prednisolone than in the control group ($P < 0.02$). The change in transport maximum (Tm) between

FIG. 2. Initial values for storage (S) and transport maximum (Tm) for BSP in 'prednisolone' and 'control' groups of patients with active chronic hepatitis. Normal values (Williams, 1965) are indicated by the hatched areas.

the first and last estimations is, however, not significantly different between the two groups ($P > 0.10$) (Fig. 4). The percentage change in S/Tm is also not significantly different between the groups ($P < 0.10$). Figure 5 summarizes the results of BSP storage (S) and Tm changes in a patient with active chronic hepatitis who was treated with prednisolone (patient no. K). Values for other biochemical indices were also estimated at the same time as S and Tm determinations (Tables IV and V). A significant correlation exists between percentage change in storage (S) and change in serum bilirubin ($r = -0.461$; $P < 0.05$), and between change in storage (S) and change in serum total globulin

FIG. 3.

FIG. 4.

FIG. 3. Percentage change in BSP storage (S) in 'prednisolone' and 'control' groups of patients with active chronic hepatitis. The difference between the means is significant ($P < 0.02$).

FIG. 4. Percentage change in transport maximum (Tm) for BSP in 'prednisolone' and 'control' groups of patients with active chronic hepatitis. The difference in the means is not significant ($P > 0.10$).

FIG. 5. Serum bilirubin, aspartate transaminase, BSP retention at 30 minutes, storage (S) and transport maximum (Tm) in a patient (no. K) with active chronic hepatitis treated with prednisolone.

FIG. 5.

TABLE IV

SERIAL ESTIMATIONS OF STORAGE (S) AND TRANSPORT MAXIMUM (Tm) FOR BSP IN PATIENTS WITH ACTIVE CHRONIC HEPATITIS TREATED WITH CORTICOSTEROIDS (15 mg PREDNISOLONE DAILY) AND BIOCHEMICAL INDICES

Case No.	Sex	Age (yr)	Activity of Liver Histology	Interval between First and Final Estimation ¹ (weeks)	Body Weight (kg)	Infusion Rates (mg BSP/min)		Storage (S) (mg/mg/100 ml)	Transport Maximum (Tm) (mg/min)	S/Tm	Serum Bilirubin (mg/100 ml)	Serum Aspartate Transaminase (iu/litre)	Serum Alkaline Phosphatase (KA units/100 ml)	Serum Albumin (g/100 ml)	Serum Total Globulin (g/100 ml)	
						High	Low									
a	M	35	++	C	28	52.3 55.5	11.27 11.04	1.98 1.75	27.9 33.3	4.6 3.8	6.1 8.8	2.2 1.4	24 16	19 20	2.7 2.9	4.2 3.7
b	F	61	++	C	21	45.7 55.0	14.70 13.21	2.55 2.19	9.7 15.6	4.2 4.4	2.3 3.5	5.0 1.0	240 39	16 8	3.4 4.6	4.4 3.7
c	F	14	++	C	22	58.6 65.0	16.50 16.46	2.73 2.72	33.5 35.7	4.1 5.8	8.2 6.2	1.0 1.3	24 44	18 21	2.4 3.1	5.4 4.6
d	M	51	++	C	4	71.0 72.5	13.00 14.58	2.10 2.49	27.4 34.6	4.1 3.7	6.7 9.3	2.0 1.4	51 27	44 40	2.7 3.6	5.9 6.0
e	F	23	+++	C	13	37.3 41.0 44.6	10.60 10.39 12.36	1.66 1.81 1.96	30.4 44.5 62.5	2.5 2.7 3.3	12.2 16.5 18.9	0.5 — 0.5	142 — 40	60 — 24	2.7 3.6 2.7	8.3 — 4.6
f	F	15	+++	C	16	45.9 50.0	8.63 9.68	1.35 1.69	30.9 42.3	2.2 3.5	13.7 12.0	1.7 0.7	500 25	6 8	2.9 3.5	7.7 5.3
g	F	51	++	C	2	53.1 54.5	11.78 12.06	2.03 2.11	50.0 50.2	2.7 2.5	18.5 20.1	1.5 1.0	35 28	70 75	2.2 2.5	7.4 5.0
h	F	57	+	C	6	51.8 51.8	13.65 13.05	2.16 2.34	12.1 23.8	7.0 3.7	1.7 6.4	0.5 2.1	35 39	13 11	4.5 5.3	3.0 3.0
i	F	71	+	C	3	60.0 61.4	13.75 13.81	2.37 2.35	19.7 33.5	4.9 4.5	4.0 7.5	0.8 1.4	20 25	15 11	2.5 3.2	4.1 3.6
j	F	28	++	C	3	59.1 57.7	15.60 16.70	2.50 2.65	14.0 23.0	4.9 3.5	2.9 6.6	— 2.4	— 45	— 23	3.7 4.3	3.6 2.8
k	F	49	++	—	7	47.7 47.6 47.5 45.4	12.76 9.81 12.49 12.54	2.15 1.76 1.97 2.04	28.8 33.6 39.8 54.3	4.8 4.8 6.8 5.4	5.9 6.9 5.8 10.0	0.5 0.5 — 0.5	50 20 — 11	16 — — 8	3.9 — — 3.9	3.1 — — 3.4
l	M	17	++	C	4	48.1 48.1	8.62 9.75	1.53 1.71	23.7 25.2	2.3 2.4	10.3 10.5	1.0 0.9	112 35	10 7	4.5 4.3	2.3 2.6
m	M	21	+	—	3	54.1 54.1 54.2 54.4	12.41 12.38 12.96 12.50	2.12 2.07 2.26 2.20	48.7 48.8 52.6 74.4	3.5 4.0 4.4 5.0	13.9 12.2 11.9 14.9	1.5 — — 1.2	110 — — 13	17 — — 8	3.8 — — 4.4	3.9 — — 3.9
n	F	16	++	—	4	50.5 50.5 50.9 52.0	10.46 10.71 10.54 10.21	1.78 1.70 1.86 1.76	28.6 35.9 16.1 25.0	3.4 1.6 3.4 3.0	8.5 22.2 4.7 8.3	8.4 — — 4.7	93 — — 70	26 — — 27	— 4.7 3.8 3.8	— 5.3 7.5 4.8
o	M	42	+++	C	20	69.1 85.5	14.10 16.00	2.41 2.78	33.1 33.2	2.9 4.8	8.8 6.9	3.7 0.5	85 16	27 19	3.4 3.7	5.0 3.3
p	F	59	++	C	4	70.7 69.9 66.4	12.89 14.10 13.13	2.30 2.56 2.25	5.4 15.3 18.7	4.2 4.5 4.7	1.3 3.4 4.0	4.0 — 2.0	66 — 20	21 — 27	3.8 — 3.7	3.6 — 3.9

¹For each patient, the first estimation was done immediately before starting corticosteroids.

C = cirrhosis.

($r = -0.501$; $P < 0.01$) in individual patients in the two groups. The correlation between percentage change in storage (S) and change in serum aspartate transaminase is not significant ($r = -0.267$; $P < 0.10$). There is not a significant correlation between percentage change in transport maximum (Tm) and change in serum alkaline phosphatase in the two groups ($r = +0.110$).

Urinary BSP during the S and Tm determinations was estimated in most patients studied; this was approximately 5% of the total BSP infused, and in no case greater than 10%. Urinary BSP excretion was not influenced by corticosteroids.

STORAGE (S) AND TRANSPORT MAXIMUM (Tm) IN CHOLESTATIC VIRUS HEPATITIS AND OBSTRUCTIVE JAUNDICE Table VI summarizes the results. Figure 6 shows the changes in serum bilirubin and BSP S and Tm in a patient with cholestatic virus hepatitis (patient P.B.). After four weeks of jaundice, prednisolone (30 mg daily) was started. Serum bilirubin and aspartate transaminase fell rapidly; storage (S) increased from 16.0 to 33.9 mg/mg/100 ml; Tm fell from 4.4 to 2.6 mg/min; and the S/Tm ratio increased from 3.6 to 13.0. The two patients treated with prednisolone had an increase in BSP storage (S) of 112% and 99% respectively during the first

TABLE V

SERIAL ESTIMATIONS OF STORAGE (S) AND TRANSPORT MAXIMUM (Tm) FOR BSP IN A GROUP OF PATIENTS WITH ACTIVE CHRONIC HEPATITIS NOT RECEIVING CORTICOSTEROIDS AND BIOCHEMICAL INDICES

Case No.	Sex	Age (yr)	Activity of Liver Histology	Interval between First and Final Estimation (weeks)	Body Weight (kg)	Infusion Rates (mg BSP/min)		Storage (S) (mg/100 ml)	Transport S/Tm (mg/min)		Serum Bilirubin (mg/100 ml)	Serum Aspartate Transaminase (iu/ litre)	Serum Alkaline Phosphatase (KA units/ 100 ml)	Serum Albumin (g/100 ml)	Serum Total Globulin (g/100 ml)	
						High	Low									
q	M	31	++	C	22	52.7	14.82	2.54	28.3	5.0	5.7	6.2	150	100	2.6	5.9
r	F	11	+++	C	37	55.9	12.75	2.21	35.0	3.6	9.7	3.2	52	79	—	—
						30.0	6.92	1.14	23.1	1.6	14.4	1.4	92	44	4.0	4.2
s	F	50	++	C	19	34.6	6.54	1.11	25.4	1.5	16.9	4.5	95	44	3.0	6.7
						58.2	14.25	2.50	22.5	4.9	4.6	—	—	2.2	5.5	
t	F	48	++	C	14	66.8	14.10	2.41	18.4	5.5	3.3	0.6	19	12	4.3	3.7
						69.1	11.22	1.92	51.1	2.8	18.3	3.1	31	31	3.1	3.5
u	F	57	++	C	9	68.7	9.70	1.69	41.9	2.3	18.2	—	—	3.1	3.5	
						68.6	10.11	1.79	32.7	3.1	10.5	2.3	25	38	2.0	4.4
v	F	57	++	C	9	46.8	10.56	1.84	24.1	2.1	11.5	1.6	19	24	3.0	5.1
						48.0	10.21	1.82	40.2	1.9	20.0	2.2	45	54	3.4	4.5
w	M	39	++	C	9	85.4	16.82	2.79	41.1	4.5	9.1	3.6	71	24	3.0	5.7
						90.5	15.85	3.03	33.9	4.4	7.7	5.2	151	17	2.2	5.6
x	M	30	+++	C	2	62.5	12.98	2.21	37.1	4.5	8.2	3.5	40	23	3.0	3.8
						62.2	12.49	2.07	33.0	3.7	8.9	3.5	40	23	—	—
y	F	66	+	—	5	80.0	13.66	2.28	21.8	6.7	3.3	0.5	15	11	3.5	3.6
						80.0	12.97	2.10	26.7	6.3	4.2	0.5	15	11	—	—
z	M	46	+	—	3	82.7	16.62	2.85	45.2	8.4	5.4	0.5	18	8	4.4	2.9
						84.5	16.53	2.70	40.4	7.2	5.7	0.7	35	6	4.0	2.6
ai	M	22	+	C	10	61.8	11.91	2.06	39.9	4.8	8.3	0.5	17	6	4.7	2.6
						61.8	14.12	2.52	41.4	4.4	9.4	0.9	12	—	4.0	2.8
bi	F	66	+	C	13	73.2	16.49	2.64	58.0	11.2	5.2	0.5	15	13	3.7	3.6
						75.9	12.50	2.17	30.3	5.8	5.2	0.5	15	13	—	—
ci	F	17	++	C	34	63.6	9.62	1.56	24.8	1.4	17.7	5.8	77	63	3.3	3.5
						65.5	11.60	1.91	22.9	3.5	6.5	6.0	19	78	4.0	2.9
ci	M	12	+	—	25	41.8	12.84	2.06	39.1	4.0	9.8	0.5	45	54	4.0	3.0
						41.8	9.45	1.57	70.6	4.4	16.0	0.5	29	23	3.8	2.4

C=cirrhosis

72 hours; the third patient, who was not given a corticosteroid, had an increase of 14% during 72 hours and 2% during the following seven days. In the treated patients, the transport maximum (Tm) decreased by 41% and 14% respectively, and in the control it increased by 4% in 72 hours, and 13% in the next seven days. Serum bilirubin fell from 24.0 to 13.0 mg/100 ml and 2.2 to 1.1 mg/100 ml in the treated patients; it was relatively constant in the control. Aspartate transaminase fell from 285 to 115 and 148 to 35 iu/litre in the treated patients, and from 39 to 28 iu/litre in the control. Alkaline phosphatase values showed little change.

Figure 7 shows the changes in serum bilirubin and S and Tm in a patient with carcinoma of the head of the pancreas (patient W.H.). Bromsulphthalein storage (S) was 16.1 mg/mg/100 ml before corticosteroids and 15.5 mg/mg/100 ml five days later; the fall was 4% of the initial level. Transport maximum at the corresponding times was 2.7 and 2.6 mg/min; an increase of 4%. Storage/transport maximum was 6.0 before and after corticosteroids. Biochemical indices remained essentially constant.

CHROMATOGRAPHIC STUDIES OF BSP IN PATIENTS WITH ACTIVE CHRONIC HEPATITIS Tables VII and VIII summarize the results. The mean interval between the two BSP estimations was five (2-16) and seven (2-14) weeks in the group treated with prednisolone and the control group respectively. High conjugated BSP levels were common in the early samples (Plaa, Sparks, and Hunter, 1962), and were not influenced by corticosteroids. Attention was confined to 30-minute specimens (Tables VII and VIII). The level of conjugated BSP at the first estimation was 29.1 (3.4 to 82.6)% in the treated, and 39.4 (7.8 to 94.9)% in the control group. The mean difference between the percentage of BSP conjugated at the first and second estimation was +22.4 (−55.0 to +79.5)% in the treated, and −7.8 (−68.2 to +57.6)% in the control group; the difference between the means is significant ($P < 0.02$) (Fig. 8). All of these patients also had BSP storage (S) and Tm estimations. The correlation between percentage change in storage (S) and change in the level of conjugated BSP in the peripheral blood is not significant ($r = +0.340$; $P < 0.10$). The correlation between percentage change in transport

FIG. 6.

W.H. M56. Ca. pancreas.

FIG. 7.

FIG. 8.

FIG. 6. Serum bilirubin and BSP storage (S) and transport maximum (Tm) in a patient (P.B.) with cholestatic virus hepatitis before and 72 hours after starting prednisolone.

FIG. 7. Serum bilirubin and BSP storage (S) and transport maximum (Tm) in a patient (W.H.) with extrahepatic obstructive jaundice due to pancreatic carcinoma before and five days after starting prednisolone.

FIG. 8. Change in the percentage of BSP conjugated at 30 minutes after an intravenous injection of 5.0 mg per kg body weight in 'prednisolone' and 'control' groups of patients with active chronic hepatitis between two estimations. The difference between the two means is significant ($P < 0.02$).

TABLE VI

STORAGE (S) AND TRANSPORT MAXIMUM (T_m) FOR BSP IN PATIENTS WITH CHOLESTATIC VIRUS HEPATITIS AND OBSTRUCTIVE JAUNDICE AND BIOCHEMICAL INDICES

Case	Sex	Age (yr)	Date	Body Weight (kg)	Cortico- steroids ¹	Infusion Rate (mg BSP/min)		Storage (mg/mg/ 100 ml)	Transport Maximum (T _m) (mg/min)	S/T _m	Serum Bilirubin (mg/100 ml)	Serum Aspartate Trans- aminase (iu/litre)	Serum Alkaline Phos- phatase (KA units /100 ml)
						High	Low						
Virus Hepatitis													
P.B.	M	18	21.9.64	65.4	O	12.02	1.96	16.0	4.4	3.6	24.0	285	24
			22.9.64		S						23.5	—	—
			23.9.64		S						22.0	270	32
			24.9.64		S						18.5	135	—
			25.9.64	66.8	S	12.77	2.17	33.9	2.6	13.0	13.0	115	30
			26.9.64		S						10.0	—	—
			28.9.64		S						9.1	67	34
			29.9.64		S						8.9	—	—
			30.9.64	69.1	S						7.5	78	20
Virus Hepatitis													
J.D.	M	24	18.2.65	66.8	O						2.1	200	26
			22.2.65		O						2.2	148	25
			26.2.65	66.8	O	12.86	2.35	22.3	4.8	4.6	—	—	—
			1.3.65	67.7	S	12.29	1.93	44.5	4.1	10.8	1.1	35	18
			4.3.65		S						1.2	25	15
			8.3.65		S						1.3	19	12
Virus Hepatitis													
G.B.	M	31	5.11.64		O						2.3	33	27
			10.11.64	73.2	O	14.08	2.54	47.4	4.4	10.8	1.1	39	30
			13.11.64		O	15.30	2.70	53.9	4.6	11.7	1.1	—	—
			16.11.64	72.7	O						1.0	28	24
			19.11.64		O						0.9	17	20
			20.11.64		O	14.58	2.61	55.1	5.2	10.6	0.8	19	24
Extrahepatic Obstruction (Carcinoma of Pancreas)													
W.H.	M	56	17.12.64	63.2	O						6.4	84	52
			22.12.64		O						6.2	84	56
			29.12.64		O						12.0	73	66
			30.12.64	63.2	O	13.30	2.27	16.1	2.7	6.0	—	—	—
			31.12.64		S						10.4	73	—
			1. 1.65		S						13.2	74	—
			2. 1.65		S						14.6	73	69
			4. 1.65	61.8	S	11.90	2.05	15.5	2.6	6.0	12.4	68	—
			5. 1.65		S						12.6	88	—
			6. 1.65		S						11.8	70	—
			7. 7.65		S						13.2	66	37
			8. 1.65	60.4	S						14.4	54	—

^aS=oral corticosteroids (Prednisolone 30 mg daily)

TABLE VII

CONJUGATION OF BROMSULPHTHALEIN (BSP) IN VENOUS BLOOD AT 30 MINUTES AFTER INTRAVENOUS INJECTION OF 5.0 mg BSP PER kg BODY WEIGHT IN ACTIVE CHRONIC HEPATITIS TREATED WITH CORTICOSTEROIDS

Case No.	Interval between Two Estimations (weeks)	Before Prednisolone				After Oral Prednisolone (15 mg daily)			
		% BSP Retention at 30 Minutes	Total BSP at 30 Minutes (mg/100 ml)	Conjugated BSP at 30 Minutes (mg/100 ml)	% BSP Conjugated at 30 Minutes	% BSP Retention at 30 Minutes	Total BSP at 30 Minutes (mg/100 ml)	Conjugated BSP at 30 Minutes (mg/100 ml)	% BSP Conjugated at 30 Minutes
		Minutes	(mg/100 ml)	(mg/100 ml)	Minutes	Minutes	(mg/100 ml)	(mg/100 ml)	Minutes
i	7	44.4	3.30	0.19	5.7	46.8	3.40	1.09	32.0
ii	4	41.7	3.40	0.16	5.2	22.0	1.80	0.58	32.2
iii	8	20.8	1.46	0.40	27.4	16.3	1.14	0.83	72.8
iv	16	30.1	2.26	0.29	13.2	19.7	1.40	1.25	89.2
v	2	22.3	1.45	1.00	68.9	22.6	1.58	0.22	13.9
vi	3	37.4	3.50	0.12	3.4	42.7	3.60	0.46	12.8
vii	6	25.4	2.80	0.11	3.9	34.7	3.30	0.30	10.9
viii	2	48.7	5.08	3.30	64.9	43.1	4.57	3.70	80.9
ix	2	17.6	1.27	0.07	5.5	10.0	0.80	0.68	85.0
x	3	36.1	3.01	1.80	60.0	19.3	2.00	1.80	90.0
xi	2	22.0	2.13	1.76	82.6	10.9	1.14	1.02	89.5
xii	3	45.0	4.10	0.36	8.8	43.0	3.90	0.35	8.9

TABLE VIII

CONJUGATION OF BROMSULPHTHALEIN (BSP) IN VENOUS BLOOD AT 30 MINUTES AFTER INTRAVENOUS INJECTION OF 5.0 mg BSP PER kg BODY WEIGHT IN ACTIVE CHRONIC HEPATITIS NOT TREATED WITH CORTICOSTEROIDS

Case No.	Interval between Two Estimations (weeks)	Initial Estimation			Second Estimation				
		% BSP Retention at 30 Minutes	Total BSP at 30 Minutes (mg/100 ml)	Conjugated BSP at 30 Minutes (mg/100 ml)	% BSP Conjugated at 30 Minutes	% BSP Retention at 30 Minutes	Total BSP at 30 Minutes (mg/100 ml)	Conjugated BSP at 30 Minutes (mg/100 ml)	% BSP Conjugated at 30 Minutes
xiii	12	55.1	3.43	0.62	18.2	42.7	2.98	2.26	75.8
xiv	14	48.3	4.93	0.80	16.2	29.4	3.70	0.22	5.9
xv	9	47.4	3.13	1.15	36.7	52.8	3.70	1.40	37.8
xvi	8	33.5	2.78	2.64	94.9	34.6	2.70	0.72	26.7
xvii	8	58.9	3.24	2.20	67.9	56.5	4.46	0.65	14.6
xviii	2	42.0	3.15	0.58	18.4	44.9	3.46	0.07	2.0
xix	5	16.2	1.91	0.15	7.8	18.1	2.25	0.18	8.0
xx	3	5.2	0.63	0.10	15.9	5.4	0.63	0.10	15.9
xxi	7	10.4	0.70	0.55	78.6	10.6	0.89	0.78	87.6

maximum (T_m) and change in the level of conjugated BSP is also not significant ($r = +0.302$; $P < 0.10$). Plasma from six healthy volunteers and six patients with active chronic hepatitis were incubated with BSP *in vitro* and treated in an identical way to the samples in the study. In no case was an appreciable amount of BSP conjugated. In most patients, chromatography of urinary BSP was performed, and in all most was conjugated.

DISCUSSION

Intravenously injected bromsulphthalein is excreted mainly by the liver (Rosenthal and White, 1925) although a part may be played by extrahepatic uptake (Cohn, Levine, and Kolinsky, 1948; Rosenau, Carbone, and Grodsky, 1959), and by an entero-hepatic circulation (Lorber, Oppenheimer, Shay, Lynch, and Siplet, 1953). With moderate BSP loads as used in the present study, however, insignificant exchange occurs outside the liver (Leevy, 1961; Norberg, Senning, and William-Olsson, 1962) and the studies reported here give precise information on hepatic cellular function.

Increased BSP retention has been shown in the patients with active chronic hepatitis; this agrees with other observers (MacKay *et al.*, 1959; Page and Good, 1960; Reynolds, Edmondson, Peters, and Redeker, 1964). In patients in the controlled trial a significant decrease in BSP retention has been shown during the first few weeks after starting corticosteroids, and this confirms previous uncontrolled observations (Page and Good, 1960; Kern, Vinnik, Struthers, and Hill, 1963). Plasma disappearance rate (PDR) or fractional clearance (MacDonald, 1939; Ingelfinger, Bradley, Mendeloff, and Kramer, 1948; Richards, Tindall, and Young, 1959; Leevy, 1961), which may be abnormal in the

presence of a normal 30-minute retention (MacDonald, 1939; Richards *et al.*, 1959), also improved more often in patients treated with corticosteroids; this difference is not, however, significant. The calculated plasma BSP level at zero time and therefore the volume of distribution of BSP relative to body weight varied considerably in the individual patients in this study. Although corticosteroids expand plasma volume and cause fluid retention (Bongiovanni and Eisenmenger, 1951), they have not significantly influenced the volume of distribution of BSP in this study.

The correlation between change in BSP retention and change in serum bilirubin, aspartate transaminase, and total globulin in both groups was significant. Decreased retention seems therefore to reflect a general improvement in hepato-cellular function. This preliminary study of patients in the controlled trial has therefore shown a significant improvement in liver cell function soon after starting corticosteroids.

In order to investigate further the mechanism of the decrease in BSP retention following corticosteroids, BSP transport maximum (T_m) and storage (S) were estimated in a further group of patients with active chronic hepatitis. Impairment of storage (S) and transport maximum (T_m) in patients with untreated active chronic hepatitis (Preisig, Williams, Sweeting, and Bradley, 1963) has been confirmed. Although there was no significant difference between these indices in a group of patients receiving corticosteroids compared with a control group, the following study showed a significant increase in S in patients with active chronic hepatitis after starting corticosteroids. Improvement in S has therefore been shown to account for at least part of the decrease in BSP retention after corticosteroids. This improvement correlates significantly with a decrease in serum

bilirubin and total globulin. Storage (S) is a more labile factor than Tm, for in extrahepatic portal obstruction (Thompson, Williams, and Sherlock, 1964) and old age (Thompson and Williams, 1965) it shows a disproportionate reduction. In old age, diminution in storage is paralleled by a diminution in liver size (Thompson and Williams, 1965). It is therefore of interest that corticosteroids can increase liver size (Gyorgy and Bluemle, 1951; Chapman, Kark, Keeton, Calloway, Consolazio, Weigend, Dyniewicz, and Kyle, 1952; Clark, 1953), and this might account for at least part of the increase in S. In animals some therapeutic agents increase BSP uptake by the liver (Fujimoto, Eich, and Nichols, 1965); their effects on storage and transport maximum have not, however, been investigated. In patients with active chronic hepatitis the depressed transport maximum (Tm) improved much less than S. The Tm for BSP is probably constant, and analogous to renal tubular transport (Bradley, Ingelfinger, Bradley, and Curry, 1945; Mason, Hawley, and Smith, 1948; Lewis, 1950; Verschure, 1952; Taleisnik, 1955; Wheeler *et al.*, 1960; Schoenfield *et al.*, 1964). Whether or not there are separate Tm rates for free and conjugated BSP has not yet been determined (Schoenfield, 1965). There was not a significant correlation in the present study between change in Tm and change in serum alkaline phosphatase.

In two patients with cholestatic virus hepatitis, corticosteroids resulted in a very rapid increase in BSP storage (S). This paralleled closely the fall in serum bilirubin and aspartate transaminase and probably reflects an acceleration of the improvement in hepatic function which occurs during natural recovery. One of the patients studied showed a striking increase in BSP storage (S) although the initial serum bilirubin was only 2.2 mg/100 ml. The change is not therefore a result of the fall in the serum bilirubin level. Improvement in S during recovery occurs before that in Tm (Schoenfield *et al.*, 1964; Preisig *et al.*, 1966).

Suggested mechanisms for the fall in bilirubin after corticosteroids are increased cholestasis (Patterson, Dingman, Shwachman, and Thorn, 1954), increased urinary excretion, and decreased red cell destruction. All of these, however, seem unlikely (Williams and Billing, 1961). A direct effect of corticosteroids on the liver (Katz, Ducci, and Alessandri, 1957; Summerskill, Clowdus, Bollman, and Fleisher, 1961), or an altered or additional metabolic pathway (Chalmers, Gill, Jernigan, Svec, Jordan, Waldstein, and Knowlton, 1956; Williams and Billing, 1961) have also been suggested as an explanation. In the present study, the patient with complete extrahepatic biliary obstruction showed

very little change in BSP storage (S) or the serum bilirubin level. Although hepatic storage for bilirubin has not been demonstrated, this must presumably exist. The present study suggests that in cholestatic virus hepatitis corticosteroids increase bilirubin storage in parenchymal cells; this would account for the rapid fall in serum bilirubin during the first 48-72 hours of treatment (Williams and Billing, 1961). In all cases in the present study, Tm showed much less variation than S; excretion of bilirubin has also been shown to be by a rate-limited process with a fixed Tm (Weinbren and Billing, 1956; Arias, Johnson, and Wolfson, 1961; Brown, Grodsky, and Carbone, 1964). Conjugation of bilirubin is also probably increased by corticosteroids (O'Donnell, Mistilis, Schiff, and Cobb, 1963). The cause of the sudden fall in serum aspartate transaminase after corticosteroids is not adequately explained (O'Brien, Goble, and MacKay, 1958; Redeker, Reynolds, and Kuzma, 1961); it is certainly not due to an abrupt cessation of hepatocellular destruction as this continues microscopically throughout the course of the disease. This may also be due, in part at least, to a general increase in hepatic storage.

Conjugation of BSP is largely with glutathione and takes place mainly in the liver (Brauer, Pessotti, and Krebs, 1955; Combes, 1959; Grodsky *et al.*, 1959; Meltzer, Wheeler, and Cranston, 1959; Javitt *et al.*, 1959). In the present study, the 30-minute level of conjugated BSP increased significantly more in the patients who received corticosteroids than in the controls. In infective hepatitis and cirrhosis, low levels of conjugate are present (Carbone, Grodsky, and Hjelte, 1959; Plaa, *et al.*, 1962; Schoenfield *et al.*, 1964); but the level rises in infective hepatitis during recovery (Plaa *et al.*, 1962). The increase in conjugated BSP after corticosteroids is probably also due to parenchymal cell recovery, and it may reflect an adaptive improvement in enzyme action (Burns, Conney, and Koster, 1963) analogous to that following therapeutic agents (Fujimoto *et al.*, 1965). Conjugation is probably important in the biliary excretion of BSP (Philp, Grodsky, and Carbone, 1961) but the present study suggests that it is not essential; there was not a significant correlation between change in storage (S) and conjugation, or between change in Tm and conjugation. As expected, most urinary BSP in the present study was conjugated (Carbone *et al.*, 1959).

SUMMARY

Serial intravenous bromsulphthalein (BSP) disappearance curves have been performed in two randomly selected groups of patients with active chronic hepatitis who had not previously received

corticosteroids. Bromsulphthalein retention decreased significantly more in the group treated with corticosteroids than in the control group.

Decrease in BSP retention has been shown to correlate well with improvement in other biochemical indices of hepatocellular function, and was not dependent on initial serum bilirubin levels.

Relative hepatic storage (S) for BSP increased more in a group of patients with active chronic hepatitis after starting treatment with corticosteroids than in a control group who were not so treated. In both groups, the transport maximum (Tm) for BSP remained relatively constant.

Similar results for S and Tm were seen in two patients with cholestatic virus hepatitis who were treated with corticosteroids. A patient with complete biliary obstruction due to carcinoma of the pancreas showed no significant change in S or Tm after corticosteroids.

Patients with active chronic hepatitis given intravenous BSP showed a significant increase in plasma conjugated BSP after corticosteroids.

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