Neither was cholesterol with albumen in the patients in the group of obstructive jaundice as is present in the cases with hepatic diseases.

DR. JOHN G. MATEER (Detroit, Mich.) (closing the discussion): We agree fully with Dr. Mann's emphasis upon the *dissociation* of different liver function tests. Because of limited time, we were unable to show our lantern slide tables of the results of the four newer liver function tests in *individual* cases. These tables of individual cases show in a striking manner the *dissociation* of the results of the different tests. A survey of these tables reveals the need, therefore, of performing at least *several* tests in each case, rather than conducting only the single test which yields the highest per cent of positive results in a group of cases.

Dr. White's inquiry about the different statistical results obtained by different workers with the cephalin test is a very reasonable question. In this study we have made no attempt to use this test to differentiate obstructive and hepatogenous jaundice. We have used the test simply as an index of *impairment* of hepatic function, regardless of whether the patient was jaundiced or not. (As a matter of fact, no *obstructive* jaundice cases were included in this clinical material). Hanger, in his second paper, and Pohle and Stewart in their recent communication attempted to evaluate the cephalin test as a method to differentiate the two common types of jaundice. The difference in their experience, we believe, was due, at least in part, to the fact that Hanger's jaundiced patients, as a group, were studied at a somewhat *earlier* stage in the course of the jaundice. So much depends upon the *duration* of the jaundice when the test is performed, that no *liver function* test should be expected to serve as a *final* differential criterion between hepatogenous and obstructive jaundice. The earlier any liver function test can be conducted in the course of jaundice, the more reliable will be its differential aid.

We agree with Dr. Rosenberg's *explanation* of the apparent differences between his experience and ours with the cephalin test. Evidently he has been using *ripened* cephalin. We have used *unripened* cephalin, following Hanger's published technique. Hanger's more recent suggestion to use ripened cephalin represents an improvement in the method. The need is thus eliminated for *excluding* the 25 per cent of *faintly* positive one + results from the total positive results obtained with *unripened* cephalin. According to Hanger, *false* faintly positive reactions upon *normal* subjects do not occur with *ripened* cephalin.

As to the optimum dose of bromsulphthalein for evaluation of liver function, Macdonald's recent experiments would suggest that the 5 mg. dose per kilo provides a more sensitive test than the 2 mg. dose. However, regardless of which dose may be selected, the employment of the *serial* method will *increase* the sensitivity of the test.

A New Galactose Test for Differentiation of Obstructive from Parenchymatous Jaundice*

By

A. M. BASSETT, M.D., T. L. ALTHAUSEN, M.D.

and

G. C. COLTRIN, M.D.

SAN FRANCISCO, CALIFORNIA

SINCE Claude Bernard first demonstrated the participation of the liver in carbohydrate metabolism, investigators of hepatic physiology have stressed the importance of the liver in an increasing number of physiologic processes. Metabolism of carbohydrates, fats and proteins, erythropoiesis, detoxification, production of prothrombin, and water balance are but a few of the many vital processes with which the liver is concerned. Pathologic lesions rarely impair all functions of the liver equally; one or more functions are disturbed in various degrees while others may to all appearances be spared. Therefore the expression, "impaired liver function" is ambiguous unless the function is specifically defined. Precise evaluation of hepatic disease requires the application of various tests each of which is designed to test a specific function. In some instances the status of a particular function may be extremely valuable in distinuishing between two different hepatic disorders which stimulate each other. Only in this limited sense can one speak of a "best" liver function test. We offer the intravenous galactose clearance test as a relatively accurate measure of the glycogenic function of the liver and as a test which in our experience, in addition to being generally useful, has proved superior to other liver function tests in the differentiation of obstructive from parenchymatous jaundice.

Bauer (1) in 1906 suggested the use of galactose as an agent for testing the glycogenic function of the liver. Subsequent investigation has confirmed the wisdom of this choice. It has been proved in several species of mammals, including man, that only the liver can utilize galactose in significant amounts (2, 3, 4, 5), that this utilization is independent of insulin (6), and that there is no renal threshold for galactose (7). The quantitative determination of galactose in the blood is simple and accurate (8). Galactose thus fulfills the requirements for a testing agent of the glycogenic function of the liver.

The original galactose test, which involves oral administration of the sugar and measurement of its urinary excretion, has proved inadequate. Most workers consider it unreliable in the differentiation between obstructive and parenchymatous jaundice; in chronic liver disease, notably cirrhosis, it is of even less value. However, certain features of this test suggest that the technic of application rather than the choice of testing agent is responsible for its

*Aided by a grant from the Christine Breon Fund for Medical Research.

From the Department of Medicine, University of California Medical School, San Francisco, Calif.

Read at the Annual Meeting of the American Gastro-Enterological Association at Atlantic City, N. J., May 5, 1941.

limited usefulness. Particularly intestinal absorption and renal excretion of galactose may vary sufficiently to mask the functional capacity of the liver, and quantitative determinations of urinary galactose are inaccurate in the presence of bile. These deficiencies of the conventional galactose tolerance test were recognized by several earlier workers. In 1933 Roe and Schwartzman (9) described a modification of the test in which 1 gm. of galactose per kilogram of body weight was given orally and the function of the liver was measured by the resulting blood galactose curve. Although this method was an improvement, it failed to differentiate between variations in intestinal absorption of sugars, which are by no means rare (10), and utilization of galactose by the liver. In 1937 Jankelson, Segal and Aisner (11) proposed a galactose liver function test in which a standard dose of 25 gm. of galactose was injected intravenously and determinations of galactose in the blood were made at intervals. This test also was better than the original oral test, as shown by the fact that in 64 per cent of the patients with cirrhosis of the liver so tested the outcome was positive. On the other hand, the amount of galactose was below the optimum and the use of a standard dose failed to take into account the essential difference between oral and intravenous administration of galactose. In the former case the maximal absorptive capacity of the intestine automatically controls the amount of galactose which enters the blood stream according to the size of the patient. In the latter case the task imposed on the liver is identical in all patients regardless of size. Recently Maclagan (12) reported work with a test similar to that of Roe and Schwartzman except that he used a standard oral dose of galactose (40 gm.).

After publication of our preliminary paper (13) on the intravenous galactose test, King and Aitken (14), who consider that there is no advantage in graded doses of galactose, reported their results with the technic of Jankelson, Segal and Aisner. Their conclusions regarding the differential value of the intravenous galactose test in obstructive and parenchymatous jaundice agree with ours although their absolute values for galactose levels differ, probably because they used different brands of galactose and yeast and different chemical methods. The objections to the use of a standard dose of galactose have already been stated. The favorable results obtained by these workers may have been due to an accidental lack of variation in body weight in a small series of cases (10 patients with obstructive jaundice and 15 patients with acute hepatitis).

In devising our intravenous galactose clearance test, we attempted to eliminate all variables except the glycogenic function of the liver. We found that the rate of clearance of galactose injected intravenously depended upon the size of the dose in terms of body weight as well as upon the functional state of the liver. Therefore the dose was graded according to the patient's ideal weight. An amount was sought which would be large enough to detect slight impairment of function and yet small enough to permit rapid intravenous injection with a luer syringe of available size. One-half gram of galactose per kilogram of body weight best fulfilled these requirements. Originally the blood galactose level was determined every 15 minutes for two hours following the injection. Subsequently the critical period was established at 75 minutes and the 60-minute specimen was used as a check.

TECHNIC OF THE TEST

After an oxalated blood sample has been obtained, a dose consisting of 1 cc. of a 50 per cent solution of galactose per kilogram of body weight, is injected intravenously over a period of four to five minutes. Oxalated blood samples are again secured 60 to 75 minutes after the injection. Glucose is removed from the blood samples by fermentation with yeast according to Raymond and Blanco's (8) modification of Somogyi's method. The filtrates are analyzed for the nonfermentable reducing substance by the Hagedorn-Jensen method. In order to obtain the galactose content of the blood, the figure for reducing substances in the fasting blood is subtracted from the corresponding figure in the 60 and 75 minute specimens. A correction of 24 per cent must be added if conversion tables for glucose are used. The details of the procedure and its adaptation to the Folin-Wu method have been described elsewhere (15).

The test is usually performed on the fasting patient, but in our experience such food as toast and coffee has produced no rise in the galactose level of the blood. The injection is made with a 100 cc. syringe with eccentric tip, fitted with a short 19-gauge needle. Contamination of the samples of blood by galactose may be avoided by using opposite arms or separate veins of the same arm for injection and collection. The galactose solution used in our studies was prepared by dissolving chemically pure galactose (Pfanstiehl) in triple distilled water. The solution should be prepared fresh each morning because on cooling small crystals of galactose form which may escape notice on reheating. Over 200 tests have been performed in this manner without the occurrence of systemic reactions. Occasionally small amounts of the solution have been injected extravenously, but aside from a transitory burning pain no local reactions have resulted.

RESULTS

Normal State. Fifteen adults who had no evidence clinically or by other function tests of impaired liver function, served as normal controls. In all these the blood had been cleared of galactose 75 minutes after the injection. No galactose remained in the blood of most of the young adults (under 30 years of age) 60 minutes after the injection. Further study may show that complete clearance of galactose in 60 minutes is normal for this group. For the present, however, the more conservative 75-minute limit will be used in all cases. We have not determined the normal value for children. The 15 controls are represented on Chart 1. All values given in this paper refer to milligram per cent of galactose in the 75-minute specimen of blood.

Acute Jaundice—(Hepatitis and Extra-Hepatic Obstruction). The intravenous galactose clearance test was performed on 71 patients with acute jaundice (Table I). In the 31 patients with parenchymatous jaundice the mean galactose blood level was 48 mg. per cent ($\sigma = 2.85$). In the 33 patients with obstructive jaundice of less than six months' duration, the mean galactose blood level was 13.5 mg. per cent ($\sigma = 1.71$); however, in the seven patients with obstructive jaundice of longer than six months' duration

the corresponding mean galactose level was 22 mg. per cent. The difference between the means of the two types of jaundice is statistically highly significant. Even more important for the differential diagnostic value is the limited scatter of individual cases from the mean. As will be seen from Chart 1, there is little overlap between the galactose values for the two types of jaundice. If 20 mg. per cent of galactose is arbitrarily chosen as the critical blood level (represented in the chart by the broken horizontal line), then 82 per cent of the patients with obstructive jaundice of less than six months' duration had less than this amount while 81 per cent of those with "idiopathic" parenchymatous jaundice had more than this amount at the end of the test.* In four of the seven patients with whom the obstructive jaundice had persisted longer than six months, appreciable impairment of galactose clearance occurred. However, in the group in which the obstructive jaundice was of shorter duration, the degree of damage was not proportional to the duration of the jaundice. In fact, four of the six patients with obstructive jaundice who had been jaundiced for less than one month had galactose values

TABLE I.
COMPARISON OF RESULTS OF GALACTOSE CLEARANCE TEST IN HEPATITIS AND OBST. JAUNDICE.

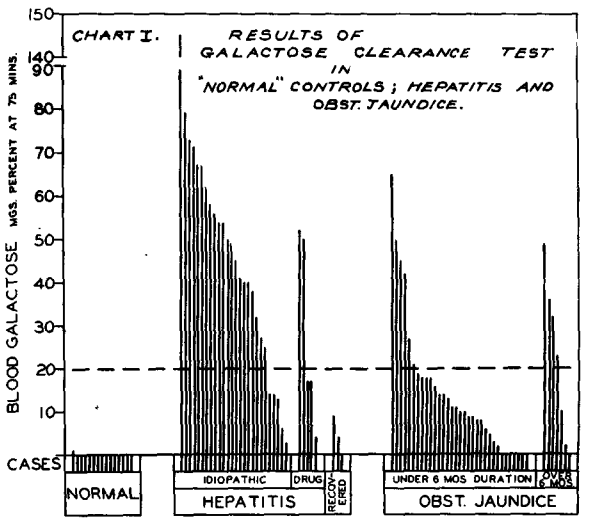
	ETIOLOGY	CASES	BLOOD GALACTOSE MGS. PERCENT AT 75 MIN.		
			MORE THAN 20	LESS THAN 20	MEAN OF GROUP
HEPATITIS	DRUG TOXICITY	5	2=40%	3=60%	
	SULFANILAMIDE	2	1	1	
	CINCHOPHEN	2	1	1	
	ARSPHENAMINE	1	0	1	
	IDIOPATHIC	26	21=81%	5=19%	48 MGS.%
	ACUTE YELLOW ATROPHY	1	1	0	
	NONSPECIFIC	25	20	5	
	RECOVERED	3	0	3	
OBST. JAUNDICE	DURATION				
	LESS THAN 6 MO.	33	6=18%	27=82%	13.5 MGS.%
	1 MONTH	16	4	12	
	1-3 MONTHS	11	2	9	
	3-6 MONTHS	6	0	6	
	MORE THAN 6 MO.	7	4=57%	3=43%	22 MGS.%

above 20 mg. per cent, and in all of the six patients in whom the jaundice had been present from three to six months, the galactose values were below the 20 mg. per cent limit. These observations suggest that the high values obtained in obstructive jaundice of less than six months' duration may represent preexisting liver damage rather than damage secondary to extrahepatic obstruction. We had the opportunity to perform the test within a few days after onset of jaundice in several patients with acute hepatitis. The resulting uniformly high values suggested that the functional impairment is often maximal at the onset. In patients who recovered, the figures for galactose declined from the original high level. In fatal cases they rose.

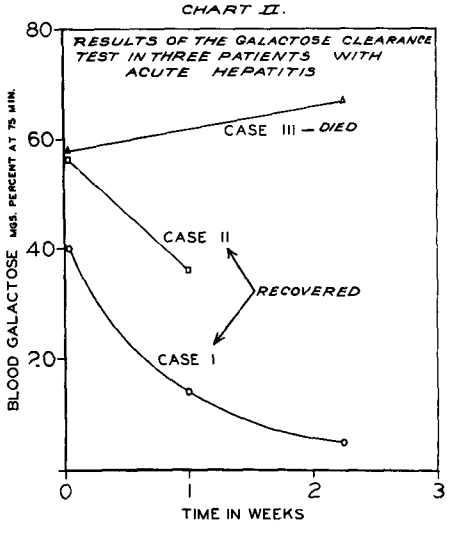
In 31 of the 40 patients with obstructive jaundice, the diagnosis was confirmed at operation or at autopsy. In the remaining nine patients the clinical and laboratory data were conclusive but associated circumstances contraindicated surgical intervention. Five of the 31 patients with acute parenchymatous

*Of 5 patients with obstructive jaundice who had appreciably more than 20 mg. per cent of galactose 2 were found at operation to have also cirrhosis of the liver. Among 5 patients with parenchymatous jaundice who had less than 20 mg. per cent of galactose 1 was rapidly recovering at the time of the test and 2 had only slight jaundice which cleared in 7 and 10 days respectively.

damage of the liver had drug poisoning caused by arsphenamine, cinchophen or sulfanilamide. Six of the remaining 26 patients died and autopsy was secured in four. In three of these acute nonspecific hepatitis was found. In the fourth patient, in whom the galactose clearance test demonstrated the greatest liver damage we have ever seen (145 mg. per cent of



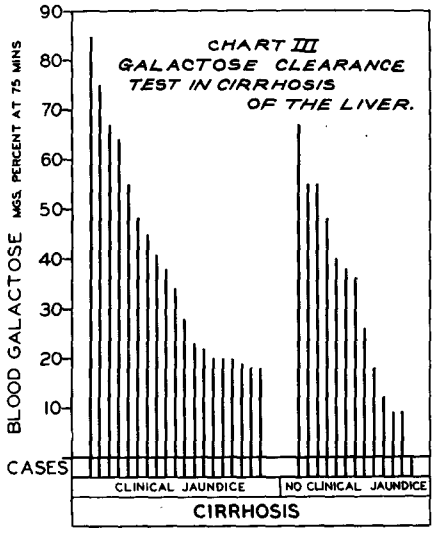
galactose), the clinical diagnosis of acute yellow atrophy of the liver was confirmed. The final diagnosis of the 21 patients who recovered was established by the clinical course and relevant laboratory data. Further classification of cases in this group does not seem warranted because of the uncertain state of nomenclature and etiologic concepts. Suffice it to say that all the patients manifested evidence of an acute diffuse involvement of the liver associated with jaundice of sudden onset and that the majority made a complete recovery. The results of the test in three



patients after recovery from acute parenchymatous jaundice are recorded on Chart I.

The intravenous galactose test is of value in prognosis as well as in diagnosis. Serial tests indicate whether liver function is improving, remains unchanged, or is decreasing. This use of the test is demonstrated in Chart II. Each point represents a

separate test and the slope of the connecting line indicates whether liver function is improving (downward slope) or decreasing (upward slope). Curve 1 represents the recovery phase of a case of acute hepatitis. Decreasing icterus index, increasing excretion of hippuric acid and clinical improvement paralleled the improvement of liver function as demonstrated by the intravenous galactose test. In Case 2 the improvement of hepatic function indicated by the galactose clearance test was the first sign of eventual recovery



from hepatitis, since the clinical course and hippuric acid excretion had remained unchanged during the period spanned by the two tests. Case 3 at first closely resembled Case 2 both clinically and in degree of impairment of liver function as measured by the intravenous galactose test. Two weeks later, however, increasing hepatic damage was recorded by the test which eventually resulted in the death of the patient. Obviously, two determinations are often not sufficient to establish the eventual prognosis. Chart II demonstrates that valuable information regarding the trend of the illness may be obtained from the test.

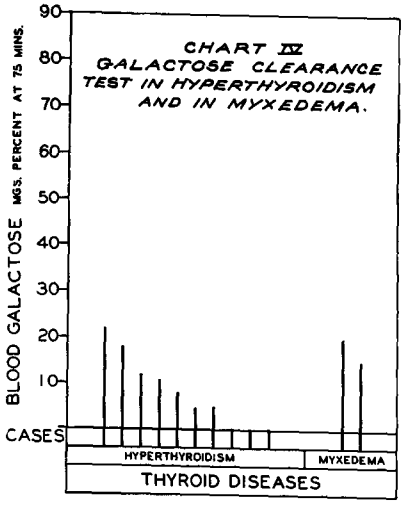
Cirrhosis of the Liver. While the oral galactose test has been of no value in the diagnosis of cirrhosis, the intravenous galactose clearance test performed by our method demonstrated impairment of hepatic function (average, 36 mg. per cent of galactose) in 31 of 32 patients (97 per cent). The results obtained in these 32 patients with cirrhosis of the liver, divided into those with and those without clinical jaundice (icterus index exceeding 15 units), are recorded in Chart III. The mean galactose values were somewhat higher in the patients with jaundice than in those without jaundice. In the former group, however, the degree of jaundice was unrelated to the severity of the functional impairment as measured by the intravenous galactose test.

Neoplasms of the Liver. Six patients with metastatic malignancy and one patient with primary carcinoma (hepatoma) of the liver were tested. Two of the six patients with metastatic malignancy had normal galactose clearance rates; the other four showed slightly impaired clearance (2 to 10 mg. per cent of galactose). The galactose retention in the patient with primary carcinoma of the liver was 15 mg. per cent. The frequent background of cirrhosis of the liver in these cases would lead one to anticipate an even more marked impairment of galactose clearance.

Acute Infections. The intravenous galactose clearance test was performed on a few patients with severe infection accompanied by generalized systemic reactions (ulcerative colitis, pneumonia, infected hydronephrosis, osteomyelitis and brain abscess). In about 75 per cent appreciable impairment of galactose clearance was demonstrated (20 to 43 mg. per cent of galactose). It would seem that this test is capable of detecting secondary toxic damage to the liver.

Endocrine Disorders. DIABETES: Reference has already been made to the independence of the utilization of galactose from the insulin mechanism. We have found that intestinal absorption and hepatic utilization of galactose are normal in the average patient with diabetes. Even such complications as moderate infection or marked lipemia do not affect the galactose clearance. Occasionally, however, a patient with diabetes suddenly becomes "insulin resistant," that is, insulin in doses up to 1000 or 1500 units daily fails to correct the hyperglycemia and glycosuria. We have tested four such patients and in all have demonstrated impaired clearance of galactose (18, 40, 58 and 61 mg. per cent of galactose respectively). Further study is needed to determine whether this impairment of liver function is responsible for the "insulin resistance" or whether it is merely an associated finding.

Thyroid Disease. In 1937, Althausen and Wever (16) reported that the ingestion of 40 gm. galactose produced abnormally high levels of galactose in the blood of hyperthyroid patients. Later, animal experiments (17), in which intestinal absorption was studied directly, demonstrated markedly increased absorption of sugars, including galactose, in the hyperthyroid state. Recently Maclagan and Rundle (18) confirmed the presence of high post-absorptive galactose blood levels in clinical hyperthyroidism but ascribed it to impaired liver function which is known to occur not



infrequently in patients with hyperthyroidism (19). In order to solve this problem, the rate of clearance of galactose from the blood should be determined in patients who have high post-absorptive levels of galactose in the blood. These studies had been made previously (15); but since the technic and standards of the intravenous galactose clearance test are now better understood, they have been repeated. In each of the ten cases of hyperthyroidism represented on Chart IV the post-absorptive levels of galactose in the

blood were markedly elevated. Nevertheless the utilization of galactose as measured by the intravenous galactose clearance test was normal in three, slightly impaired in five and significantly abnormal in only two patients. Equally conclusive are the observations in two cases of myxedema in which despite significantly impaired utilization of galactose by the liver, the post-absorptive values of galactose in the blood were well below normal. Thus, in clinical as well as in experimental hyperthyroidism, the increased intestinal absorption of galactose is sufficient to account for the high blood values although the presence of impaired liver function accentuates this effect.

COMPARISON OF THE GALACTOSE CLEARANCE TEST WITH OTHER LIVER FUNCTION TESTS

The main purpose of this report is to present data which demonstrate the value of the intravenous galactose clearance test in measuring the glycogenic function of the liver and to point out its merits in differentiating obstructive from parenchymatous jaundice. A systematic study of hepatic function tests is not attempted, but for purposes of orientation the intravenous galactose clearance test is compared with some of the other tests commonly employed in determining the functional state of the liver. Drs. Lucia and Aggeler (20) have investigated the relationship between prothrombin concentration and hippuric acid excretion in some of the patients included in this study and have kindly allowed us to use their results. In each of the main classifications of hepatic disease previously listed, icterus index, rose bengal test, oral hippuric acid test and blood prothrombin values were compared with the galactose clearance test in the following manner. The incidence of impaired function detected by the galactose clearance test was compared with the incidence of impaired function detected by each of the other tests. Furthermore, a calculation was made of the coefficient correlation* between the degree of impairment demonstrated by the galactose clearance test and that demonstrated by the other tests (Table II).

TABLE II.
COMPARISON OF THE GALACTOSE CLEARANCE TEST WITH OTHER LIVER FUNCTION TESTS.

LIVER FUNCTION TEST	TYPE OF LIVER DISEASE					
	OBST. JAUNDICE		HEPATITIS		CIRRHOSIS	
	PERCENT ABNORMAL	CORRELATION TO GAL. CLEARANCE	PERCENT ABNORMAL	CORRELATION TO GAL. CLEARANCE	PERCENT ABNORMAL	CORRELATION TO GAL. CLEARANCE
IV GALACTOSE CLEARANCE	18 (<20 MG%)		81 (>20 MG%)		97	
ICTERUS INDEX	100	NONE	100	NONE	59	NONE
ROSE BENGAL	50	NONE	76	NONE	95	SLIGHT
HIPPURIC ACID	100	NONE	100	HIGH	92	NONE
PROTHROMBIN	36	NONE	38	GOOD	54	NONE

Obstructive Jaundice. The galactose clearance test demonstrated seriously impaired function of the liver (more than 20 mg. per cent galactose in the blood at the end of the test) in but 18 per cent of the patients with jaundice of less than six months' duration. The icterus index was elevated in 100 per cent but showed no correlation with the galactose clearance values. As would be expected, the rose bengal retention paralleled the degree of jaundice and also showed no correlation with the galactose clearance test. Excretion of hippuric acid was impaired in all of the 13 patients tested. However, the coefficient of correlation between the degree of excretion and the degree of galactose clearance was not significant. Prothrombin determinations were performed in 14 patients, in five of whom (36 per cent) the level was less than 70 per cent of normal. All low prothrombin values returned to normal when Vitamin K was administered. No correlation existed between the prothrombin level and the rate of galactose clearance.

Acute Hepatitis. The galactose clearance test indicated serious impairment of hepatic function in 81 per cent of the patients with nonspecific hepatitis. All of these had an elevated icterus index but the degree of jaundice was not correlated with the degree of galactose clearance. The retention of rose bengal dye was roughly proportional to the degree of jaundice. Again no strict correlation existed between the degree of dye retention and the rate of galactose clearance. The excretion of hippuric acid was diminished in all ten patients in whom it had been tested. A highly significant coefficient of correlation existed between the amount of hippuric acid excreted and the degree of galactose clearance. The prothrombin level was less than 70 per cent of normal in five (38 per cent) of the 13 patients in whom it was tested. In these patients the deficiency of prothrombin was not corrected by administration of Vitamin K. Here too a significant coefficient of correlation existed between the prothrombin level and the galactose clearance.

Cirrhosis of the Liver. As stated previously, galactose clearance was impaired in 97 per cent of the patients with cirrhosis of the liver. The icterus index was elevated to the level of clinical jaundice (15 units or higher) in 59 per cent. Although a significant difference in the degree of galactose clearance was noted between patients with jaundice and those without jaundice, no correlation could be made between the height of the icterus index and the degree of galactose clearance. The rose bengal test was abnormal in 19 of the 20 patients tested. A barely significant degree of correlation existed between the values of the rose bengal and of the galactose clearance tests. The hippuric acid test was performed in 13 patients with cirrhosis of the liver, 12 of whom (92 per cent) showed impaired excretion. The coefficient of correlation between the hippuric acid excretion and the galactose clearance was not significant. In seven of 13 patients (54 per cent) the prothrombin level was less than 70 per cent of normal. In none of these could the deficiency of prothrombin be corrected by administration of Vitamin K. Here too the calculated coefficient of correlation between the prothrombin level and the rate of galactose clearance was not significant.

*The significance of the correlation was determined by the formula for the correlation of rank (21): $P = 1 - \frac{6\Sigma D^2}{N(N^2-1)}$ where D = difference in rank.

SUMMARY AND CONCLUSIONS

1. An intravenous galactose clearance test is offered as a relatively accurate measure of the glycogenic function of the liver.

2. This test proved in our experience to be of great diagnostic value in distinguishing between extrahepatic obstructive jaundice and intrahepatic parenchymatous jaundice. Repeated tests were of prognostic value especially in acute hepatitis.

3. The galactose clearance test consistently demonstrated impaired liver function in hepatic cirrhosis, but showed little or no functional impairment in carcinoma of the liver.

4. Galactose clearance was normal in most cases of uncomplicated diabetes but was markedly impaired in cases characterized by "insulin resistance."

5. The galactose clearance was normal, or nearly normal, in most cases of hyperthyroidism even when the post-absorptive galactose blood values were very high.

6. A comparison of the intravenous galactose clearance test with other liver function tests (the icterus index, the rose bengal test, the oral hippuric acid test, and the prothrombin level) was made in obstructive jaundice, in acute hepatitis and in cirrhosis of the liver.

REFERENCES

1. Bauer, R.: Weitere Untersuchungen über alimentäre Galactosurie. *Wien. med. Wchnschr.*, 56:2537, 1906.
2. Draudt, L.: Über die Verwertung von Laktose und Galaktose nach partieller Leberausschaltung (Ecksche Fistel). *Arch. f. exper. Path. u. Pharm.*, 72:457, 1913.
3. Bollman, J. L., Powers, M. H. and Mann, F. C.: Relation of Liver to Metabolism of Galactose. Proc. Staff Meet. Mayo Clinic, 6:724, 1931.
4. Mann, F. C. and McGath, T. B.: Studies on the Physiology of the Liver. *Arch. Int. Med.*, 30:73-171, 1922.
5. Foster, G. L.: Studies on CHO Metabolism. *J. Biol. Chem.*, 55: 291-303, 1923.
6. Roe, J. H. and Schwartzman, A. S.: Galactose Tolerance of Normal and Diabetic Subjects, the Effect of Insulin Upon Galactose Metabolism. *J. Biol. Chem.*, 96:717, 1932.
7. Harding, V. J. and Van Nostrand, F. H.: Variation in Blood and Urinary Sugar After Ingestion of Galactose. *J. Biol. Chem.*, 85:765, 1930.
8. Raymond, A. L. and Blanco, J. G.: Blood Sugar Determinations and Separation of Sugars with Live Yeast. *J. Biol. Chem.*, 79:649, 1928.
9. Roe, J. H. and Schwartzman, A. S.: Galactose Tolerance as a Measure of Liver Function. *Am. J. Med. Sci.*, 186:425, 1933.
10. Althausen, T. L.: A Test for Intestinal Absorption. *Am. J. Dig. Dis.*, 6:544, 1939.
11. Jankelson, I. R., Segal, M. and Aisner, M.: Intravenous Galactose Liver Function Test. *Am. J. Dig. Dis. and Nutrit.*, 3:889, 1937.
12. Maclagan, N. F.: Galactose Tolerance as a Test of Liver Function. *The Quart. J. Med.*, 9:151, 1940.
13. Bassett, A. M., Althausen, T. L. and Coltrin, G.: A New Intravenous Galactose Clearance Test for Differentiation of Obstructive from Parenchymatous Jaundice. *Proc. Soc. Exp. Biol. and Med.*, 45:405, 1940.
14. King, E. J. and Aitken, R. S.: An Intravenous Galactose Tolerance Test. *Lancet*, 2:543, 1940.
15. Althausen, T. L., Lockhart, J. C. and Soley, M.: A New Diagnostic Test (Galactose) for Thyroid Disease. *Am. J. Med. Sci.*, 199:342, 1940.
16. Althausen, T. L. and Wever, G. K.: Galactose Tolerance in Hyperthyroidism. *J. Clin. Invest.*, 16:257, 1937.
17. Althausen, T. L. and Stockholm, M.: Influence of the Thyroid Gland on Absorption in the Digestive Tract. *Am. J. Physiol.*, 123:577, 1938.
18. Maclagan, N. F. and Rundle, F. F.: Liver Function in Thyrotoxicosis. *The Quart. J. Med.*, 9:215, 1940.
19. Wever, G. K., Althausen, T. L. and Biskind, G. R.: Liver Function in Hepatic and Extrahepatic Diseases. *Am. J. Dig. Dis. and Nutrit.*, 2:93, 1935.
20. Lucia, S. P. and Aggeler, P. M.: The Influence of Liver Damage on the Plasma Prothrombin Concentration and the Response to Vitamin K. *Am. J. Med. Sci.*, 201:326, 1941.
21. Fisher, A. F.: Statistical Methods for Research Workers. London, Oliver and Boyd, 1936.

DISCUSSION

DR. T. L. ALTHAUSEN (San Francisco, Calif.): Mr. President and Members of the Association: I want to answer several of the comments made by the discussants; first, one by Dr. Jankelson about the amount of galactose to be used. We tried various amounts and found that half a cc. of a 50 per cent solution per kilogram of body weight, represents the optimum dose. The smaller the dose, the less reliable the results of the test become; the greater the dose, the more difficult it is to administer, technically, because we have to use a three-way stopcock instead of the single needle and syringe.

Now, so far as the comparison between the oral and the intravenous galactose tests is concerned, as mentioned by Dr. Shay, the oral test has the disadvantage, that two barriers are interposed between the test and the results. One is the rate of intestinal absorption for we know that differences in absorption up to a hundred per cent occur not infrequently.

The second is that the excretion of galactose is estimated in the urine. Here I want to emphasize another very practical point which will answer Dr. Schiff's question as to whether we performed parallel studies between oral and intravenous galactose tests. We tried to perform such studies and also tried to get an idea as to how much galactose was lost through the urine in the intravenous test. Since we were chiefly interested in jaundiced patients, we soon found out that titration of galactose in the urine of a markedly jaundiced person is almost an impossible task.

Another point of interest has been raised in his paper by Dr. Rosenberg about hyperfunction of a diseased liver. Anyone who has done experimental work with various doses of hepatic poisons probably has observed hyperfunction of the liver in early stages of intoxication, especially when small doses are given. We worked with phosphorus, chloroform, manganese chloride, and with various other toxins, and almost regardless of what liver function test was used there was early evidence of a better than normal function, on the part of the liver. We explained this on the basis of excessive irritability of the hepatic cells.

I also want to say how much I was impressed by Dr. Mateer's approach to the problem of liver function tests in his paper. I think that many other clinical function tests should be compared on such a strictly objective basis —also we shouldn't limit ourselves merely to gauging the sensitivity of the various tests but should also compare them on the basis of clinical usefulness.

Toxicity Studies on Stilbestrol*

By

A. H. AARON, M.D., FRANK MEYERS, M.D., MORTON H. LIPSITZ, M.D.
and
ROGER S. HUBBARD, Ph.D.
BUFFALO, NEW YORK

A STUDY was undertaken to determine the toxic properties of stilbestrol. For the purpose of this investigation a group of patients with chronic arthritis was chosen inasmuch as it has been reported that natural estrogens are of distinct benefit in this condition (1, 2, 3, 4, 5), and such material therefore seemed especially suitable for both a clinical and toxicological study of this hormone. Moreover these

*The effect of stilbestrol in chronic arthritis will be reported elsewhere. Read at the Annual Meeting of the American Gastro-Enterological Association at Atlantic City, N. J., May 5, 1941.