Liver Function Study using ^{131}I-BSP*

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Introduction

Liver function tests are indispensable for diagnosis of liver diseases. It is desirable to conduct several kinds of test for over-all evaluation of liver function, considering the various aspects of metabolism performed in the liver. Diagnosis of the existence and severity of liver diseases, especially in cases without jaundice and judgment of prognosis and effects of treatment must be made by various liver function tests.

So far, the BSP loading test has been widely used as the most sensitive index of liver function, since BSP greatly contributes to the diagnosis of liver disease without jaundice and evaluation of prognosis. However, the clinical use of the BSP test seems to be decreasing, because side effects such as pain and anaphylaxis-like shock, occasionally causing death, have been reported (1). Indocyanine-Green (ICG) appears to be replacing the BSP test as a safer dye for the dye excretion test (2, 3), but it has not yet been widely used for clinical purposes because of problems of product stability and measurement procedures.

The authors previously used ^{35}S-BSP to analyze the mechanism of liver uptake of the dye and determined maximum hepatic uptake capacity of BSP in normal controls and for various liver diseases (4). At the same time it was noted that, in the presence of function disturbance, the liver cannot adequately handle even a tracer dose of BSP, and blood clearance of a tracer dose of BSP is not exclusively dependent on hepatic blood flow, but is also dependent on the hepatocellular function of dye uptake (5).

BSP and ICG have been also used as a bilirubin analogue for analysis of clearance after a single intravenous injection (6) and to clarify the mechanism of hepatic uptake and biliary excretion of the dye under continuous infusion in normal subjects as well as in various liver diseases (7).

A clinical test must be safe and simple for routine use. The measurement of ^{35}S-BSP is rather complicated while ^{131}I-BSP, the basic properties of which were intensively studied(8), and which was used clinically as an agent for sequential liver scanning(9), is simple to measure. Therefore ^{131}I-BSP is an ideal agent for a simple liver function test based on a single injection and a single blood sampling.

In this paper it is confirmed that BSP tests have hardly been used for liver function studies

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at Tokyo University Hospital. As a simple and safe liver function examination which might replace the conventional BSP test, a 30-minute retention test with ^{131}I-BSP was introduced and compared with the conventional BSP 45-minute retention test.

Subjects and Methods

1. Number of BSP tests performed at Tokyo University Hospital.

The frequency of BSP tests performed was examined and a histogram of frequency distribution was made for 14 years from June 1955 to June 1969. The number of transaminase (GOT) measurements done, starting with the latter half of 1962, was also examined for comparison.

2. ^{131}I-BSP retention test.

Ninety ^{131}I-BSP retention tests were performed in 83 subjects, including: 10 normal controls, 8 cases of schistosomiasis japonica showing normal liver function, 12 cases of schistosomiasis japonica with abnormal liver function tests, 24 hepatitis, 21 liver cirrhosis, 4 liver malignancies, 4 obstructive jaundice, and 4 cases with miscellaneous diseases including cholelithiasis.

Twenty-six patients in this group underwent regular BSP 45-minute retention tests 2 to 5 days later for comparison. In 7 cases, urine was collected for 24 hours to study the correlation between urinary excretion and blood retention rate.

3. Method and Calculation

Irrespective of body weight, 0.5mg of ^{131}I-BSP in a volume of 0.5ml, equivalent to 50~100uCi, was injected intravenously. Thirty minutes later, 2~3ml of blood was drawn from the opposite antecubital vein. An aliquot of 0.1 ml was then measured using a well-type scintillation counter. The same volume of ^{131}I-BSP solution as injected was diluted to 1000 ml, and 1.0 ml was accurately withdrawn to serve as a standard. The calculation was carried out as shown in the formula (Fig. 2). The denominator indicates the total rdaioactivity injected in c.p.m. while the numberator shows that of BSP remaining in the total blood at 30 minutes after injection. The figure

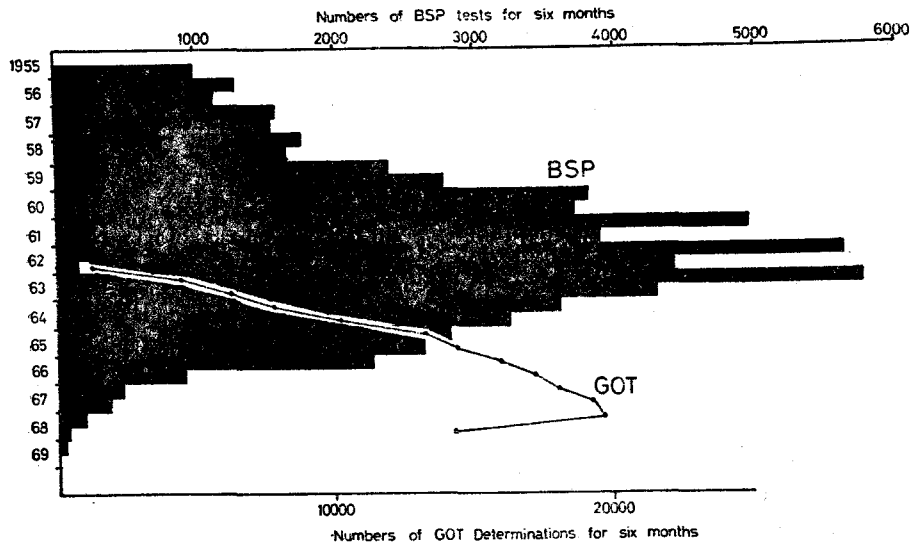

Fig. 1. Changes in the semiannual cumulative numbers of BSP tests. Transaminase determinations performed at the Central Laboratory of Tokyo University Hospital.

^{131}I-BSP RETENTION TEST

1) Dose injected : 0.5ml (0.5mg, 50~100μCi)

2) Blood sampling : 30min. after injection

3) Calculation of retention rate :

$$= \frac{\text{cpm/ml (blood)} \times \text{B.W. (gr)} \times 0.072}{\text{cpm/ml (standard)} \times 1000\,(\text{ml})} \times 100\,(\%)$$

$$= 7.2 \times \text{B.W. (kg)} \times \frac{\text{cpm/ml (blood)}}{\text{cpm/ml (standard)}}\ \%$$

Fig. 2. Method and calculation of ^{131}I-BSP 30-minute retention test.

obtained by multiplying body weight by 0.072 indicates the circulating blood volume as the BSP dilution space.

Results

Figure 1. shows the change in numbers of BSP tests performed at the Central Laboratory of Tokyo University Hospital. The tests gradually increased in number after 1955, when the Central Laboratory started, and showed marked increments after 1959. They reached a peak in the first of 1963 and then drastically decreased. Now the BSP test is practically not performed in this hospital.

Results of ^{131}I-BSP retention tests performed in various liver diseases are shown in Figure 3. Normal controls averaged 2.46±0.66% retention with an upper limit of 4%. In all patients with schistosomiasis japonica with normal liver function ^{131}I-BSP retention was within normal limits, while patients showing abnormal gamma globulin concentration showed abnormal values without exception. A high retention rate was observed in some cases with hepatitis or liver cirrhosis. As indicated by arrows, normalization of the ^{131}I-BSP retention rate was seen in the course of recovery from hepatitis. Abnormal retention rates were observed in three out of four patients with liver malignancy who showed otherwise normal liver function. All cases with obstructive jaundice showed abnormal retention values.

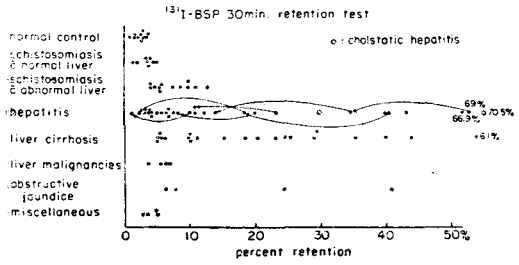

Fig. 3. Results in a series of ^{131}I-BSP retention tests.

Figure 4 shows the relationship between ^{131}I-BSP retention tests and conventional 45-minute BSP retention tests. A relatively high correlation was found to exist between these tests as indicated by a correlation coefficient of 0.782. In some cases, considerable discrepancies were observed between the tests. In most of such cases, the BSP retention rate was higher than the value with ^{131}I-BSP.

Urinary excretion rates expressed as percent of the dose administered were plotted against percent blood retention, as shown in Figure 5. Approximately 5% of the activity was excreted into the urine during 24 hours when BSP retention rate was abnormal but less than 10%. However, no more than 7% was excreted even when blood retention was as high as 30%.

Discussion

In 1959, BSP injection was reported to cause anaphylaxis-like shock very much like an abnormal reaction to penicillin, occasionally leading to death(1). In Japan, a report of death probably caused by such a reaction was also reported in the same year (10). The number of BSP examinations increased for 3 years after that year, but drastically decreased after the deter-

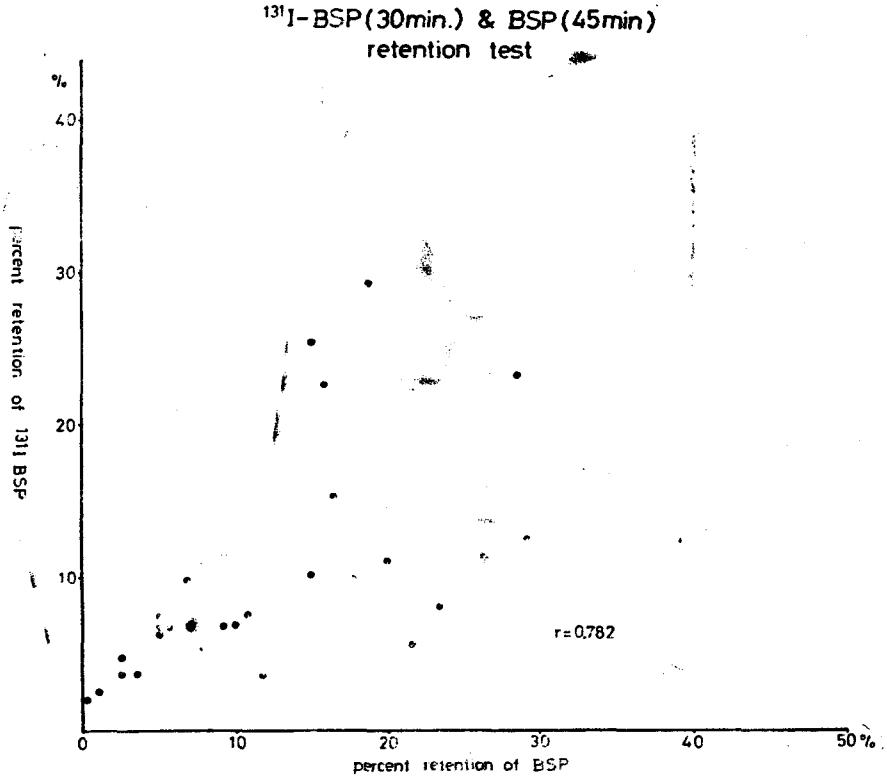

Fig. 4. Correlation between ^{131}I-BSP test and the conventional BSP 45-minute test.

mination of serum transaminase was started at the Central Laboratory. Serum transaminase values are highly significant for the detection of hepatic dysfunction in non-jaundice hepatitis as well as for evaluation of prognosis.

The increased use of serum transaminase determinations and the reports of abnormal reactions caused by BSP (11) are the chief reasons for the decreased use of the BSP test.

The most common BSP test procedure is based on intravenous injection of a dose of BSP of 5 mg per kilogram of body weight and blood sampling 30 or 45 minutes after injection. There is the possibility of overestimating the retention rate when body weight is abnormally large, such as in obese persons or patients with ascites(12). It is assumed in this method that the dye is injected into a plasma volume of 50 ml/kg body weight, and that with instantaneous mixing concentration of 10 mg/100 ml of plasma is achieved. This value is considered to represent 100 percent retention.(13)

In the ^{131}I-BSP test reported here, the dose injected is not varied with body weight. The amount of ^{131}I-BSP retained in the blood is estimated from the radioactivity in a 1-ml blood sample multiplied by the circulating blood volume. Body weight is used as an index in estimating circulating blood volume as in the conventional BSP test. As a result, there is the same possibility of overestimating circulating blood volume in obese subjects or patients with ascites. It would be ideal to measure total blood volume in each individual case, but that is practically impossible. As reported previously(9), there is a method for obtaining initial concentr-

ation by extrapolation from sequential blood sampling. However, frequent sampling is not likely to be acceptable for routine examinations. Another method is to take a blood sample 2 or 5 minutes after injection, followed by another sampling at a certain time later. The first sample is supposed to indicate 100% retention. However, it should be noted that, in the case of a single intravenous injection, mixing in the blood space and liver uptake take place simultaneously. In other words, 2 or 5 minutes after injection, a large percentage, possibly more than half, of the ^{131}I-BSP has been already taken up by the liver in normal subjects. If this value is supposed to represent complete mixing and to set the base for 100 percent concentration, this standard must be different for normal subjects and for patients with liver disease. Therefore there may not be a good separation between normal and abnormal values. Considering the difficulties in determining dilution space or the base value for 100% retention from the single-injection method, sequential sampling appears to be the best method for studying blood disappearance rate or liver uptake rate. The procedure, however, is inconvenient for an every-day screening test.

One can set a sampling time rather freely within a certain range in which separations between normal and abnormal values is most conspicuous. The reasons for using 30 minutes for blood sampling are as follows:

(a) The trace dose clearance of ^{131}I-BSP is faster than the BSP clearance of a regular dose and is comparable with the clearance using a BSP dose of 2.5 mg/kg body weight.

(b) The normal clearance curve for ^{131}I-BSP shows abend about 30 minutes after injection and enters slower phase.

(c) Previous investigations comparing sampling times showed that 30 minutes is preferable to 45 minutes.(9)

As seen in the results with the ^{131}I-BSP retention test, abnormal results were obtained in obstructive jaundice where liver blood flow is said to remain within normal limits. In case of Rotor syndrome, the ^{131}I-BSP retention rate was considerably high in spite of a normal value for ^{198}Au colloid clearance which indicated normal hepatic blood flow.(4) In a study of the clearance of a tracer dose of ^{35}S-BSP it was recognized that a liver with disturbed function cannot handle adequately even a tracer dose of the dye. This phenomenon is also observed with regard to the clearance of ^{131}I-Rose Bengal in tracer dose(13) and the clearance of ICG where the dose is 0.25—0.5 mg/kg body weight, considerably smaller than with conventional BSP(3). Therefore it was concluded that the clearance of a trace dose of dyes represents the uptake function of liver cells in case of liver disease. On the contrary, as far as the clearance of single injection is concerned, even 5 mg/kg body weight dose cannot escape the influence of liver blood flow. Therefore, the real liver uptake capacity must be measured by a double-loading method using different doses or a continuous infusion method, despite the problems involved.

However, as BSP 45 minute retention served the purpose of a screening test for the detection of liver disease, the ^{131}I-BSP retention test is also considered a useful method because of its capability of separating liver disease from normal states.

The metabolism of ^{131}I-BSP is considerably different from that of BSP or ^{35}S-BSP.(14) In order to establish the true value of the ^{131}I-BSP test further investigation is required, including a study of the discrepancies observed between the ^{131}I-BSP and BSP tests.

Conclusion

It is felt strongly that a safe and simple dye excretion test is required, now that the BSP retention test has fallen into disuse. ^{131}I-BSP is probably one of the most ideal dyes for a retention test, because its metabolism is rather clearly defined and its concentration is easy to measure. It has been shown that a simple retention test is possible using a tracer dose of ^{131}I-BSP. The average normal value by the procedure described is 2.46 ± 0.66. The data available so far define the normal range as less than 4 per cent. Correlation with the conventional BSP 45 minute test is good ($r=0.782$).

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