not successful. There will be some agent, removed by charcoal, working to recombine with transferrin and iron, and denaturation may occur in the course of acdification and neutralisation with ascorbic acid and buffer.

Radioimmunoassay was more sentitive than immunoassay, and anti-transferrin serum was not needed, if transferrin was labelled to react with anithuman serum.

Ninety eight % pure transferrin supplied by Hoechst Co. was used to obtain anti-sera and I would like to acknowledge Dr. Izumi Nakashima for his assistance in immunoassay.

## LIBC in Liver Diseases Measured by 59Fe Irosorb

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Irosorb-59 was applied for the measurement of LIBC in liver diseases.

Fundamental studies of this method revealed the following characteristics of the method. 1) plasma showed 153% values of the serum. 2) freezed serum could be used for long period however cooled serum (3-5°C) showed the increase in LIBC. 3) iron absorbing capacity of the sponge (25°C, 1 hr incubation) was 95.1 %  $\pm 1.36$  and 1 sponge could abrorb 91  $\gamma$  of iron. 4) Effect of incubation temperature on resin sponge uptake showed calibration between 5°C~30°C room temperature unnecessary. 5) By diluting serum with veronal buffer (PH 7.3) lineality of this method was proved. 6) Reproducibility of this method was confiermed by repeated measurements of the same samples.

Clinical application of this method was per-

formed on 25 cases of control, 15 cases of hepatitis, 11 cases of liver cirrhosis, 18 cases of schistosomiasis japonicum with liver damage, 19 cases of schistosomiasis japonicum without liver damage 2 cases of iron deficient anemia and 2 cases of aplastic anemia.

LIBC values were found to be  $279.3\pm38.5~\mu gr/dl$  in control cases,  $271.1\pm81.8~\mu gr/dl$  in hepatitis,  $147.0\pm41.0~\mu g/dl$  in liver cirrhosis,  $236.9\pm74.9~\mu gr/dl$  in schistosomiasis japonicum with liver damage,  $294.7\pm58.5~\mu gr/dl$  in schistomiasis japonicum without liver damage, 386 and 336 in iron deficient anemia &93.7 & 195 in aplastic anemia. Negative correlation (r=-0.528) was found between LIBC & Kunkel values in liver disease.

LIBC measured by simplified lrosorb-59 method was found to be an useful index to follow up the progress of liver cirrhosis.

## UIBC and TIBC Values in Liver Diseases Measured by Isotopic Method

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Serum iron  $U_1/_1Bl$  and T/BC values in patients with liver disease of diverse etiology were measured. They were 10 cases of constitutional hyperbilirubinemia, 19 cases of obstructive jaundice, 29 cases of infections hepatitis, and 14 cases of liver cirrhosis with 20 cases of normal control.

Serum Iron values were as follows; normal  $96.4\pm18.3~\mu\mathrm{g/dl}$  constitutional hyperbiliubinamia  $89.9\pm28.8$ , obstructive jaundice  $91.1\pm30.4$ , infections hepatitis  $212.2\pm50.2$  and liver cirrhosis  $137.0\pm36.0$ ,  $\mathrm{U_{1/1}Bl}$  values were as follows; normal  $214.2\pm39.8~\mu\mathrm{g/dl}$  constitutional hyperbiliuemia  $192.3\pm61.7$  obstructive jaundice

 $160.3\pm69.7$ . Acute hepatitis  $117.0\pm63.7$  and liver cirrhosis  $117.0\pm53.7$  respectively.

Ferrokinetics in acute hepatitis revealed that % RCU was within normal range and PIT values showed marked increase in spite

of high serum iron level.

Meanwhile ferrokinetics in constitutional perbilirabinemia indicated normal % RCV and normal PIT levels.

## Evaluation of "59Irosorb" a Kit for the Determination of Unsaturated Iron-Binding Capacity of Serum

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A diagnostic kit"59Irosorb" for the determination of unsaturated iron-binding capacity of serum has been already on the market in U.S.A. as a useful product. Investigators in our country, however, have doubted the accuracy of the Irosorb method because of unreasonably high values.

Our experiments clarified the following defects of the original procedure of the Irosorb method. First, conditions for incubation of serum with a resin sponge are not so strictly specified, although the efficiency of the resin sponge for absorbing iron is significantly influenced by temperature and time. Secondly, it is not taken into consideration that the absorption of iron is to some extent disturbed by serum.

Furthermore, we improved the procedure to determine unsaturated ironbinding capacity with sufficient accuracy for clinical use. The experimental data are as follows.

The resin sponge absorbed iron citrate ammonium completely within one hour at 37°C. However, when serum is contained in the iron

solution, the velocity of absorption became slow and the complete absorption was not attained within several hours. The suitable time for practical use was one hour at 37°C.

The grade of disturbance by serum can be determined using sera saturated with iron. The absorption rate of the resin sponge using these sera was 85–90% (if adsorption was not disturbed by serum, the rate would be 100%). The reciprocal of the figure is the correcting factor for absorption. The unsaturated iron-binding capacity of given sera is calculated from the formula using the factor.

The improved Irosorb method was compared with the spectrophotometric titration method (Rath-Finch's method modified by us) on the same specimens of serum. The results by both methods were in close agreement.

Using the improved Irosorb method, the average unsaturated iron-binding capacity of normal 10 males and 10 females was 227 and  $\mu$ g/dl, respectively, and the percentage saturation was 34.0 and 27.8%, respectively.