

## On in Vivo Destroy of the Red Blood Cells

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Recently the importance of a spleen scanning increased to differentiate a mass in abdomen and hamatological disturbances. So it is necessary to draw a clear scintigram of  $^{203}\text{Hg}$ -neohydrin has a problem on toxicity on spleen. But former method using  $^{197}\text{Hg}$  or red blood cells and the  $^{51}\text{Cr}$  labeled red blood cell method is sometimes difficult to get a good spleen scintigram, because the destroy of red blood cells is very delicate with temperature.

It was difficult to hold the in cubating temperature accurately in our laboratory. And the grade of destroy of the red blood cells depends on its conditions. For these reason, the grade of aging of red blood cells is uncertain. According to the cell age the  $^{51}\text{Cr}$  deposits mainly on the liver or disappears from the body. By this scintigram an evaluation of a spleen is uncertain. Instead of fever, we discussed about radiation. Red blood cells are irradiated by cobalt-60-rays. We employed

this red blood cells to scan of the spleen. By this method it is easy to destroy red blood cells. And we have good spleen scintigrams. The scintigrams by this method show better contour than fever-method.

For these purpose we investigated the resistance of the red blood cells by  $^{60}\text{Co}$ -ray irradiation. The destroy of the red blood cells begin from 300,000R, and 50% of red blood cells are destroyed at 400,000R. This curve of the grade of destroy shows that the best condition to get a scintigram is between 300,000R and 340,000R.

The grade of deatroy of the irradiated red blood cells shows that 40% one day after irradiation and 80% after two days. The better spleen scintigram is drawn between six hours and twenty four hours after injection.

It is suggested that application of this method to making simple method for determining a red blood cell survivals.

## Studies of Fatty Acid Metabolism with Incorporation of $^{14}\text{C}$ -Acetate into Lipids of Human Whole Blood and Bone Marrow

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As a part of studies of fatty acid metabolism, by method of incorporation of radioactivity, we report some results about incorporation of  $^{14}\text{C}$ -acetate into fatty acids from whole blood and bone marrow cells of patients with hypoplastic anemia, hypochromic (iron deficiency) anemia and leukemias.

Method; as we have already printed, after 4 hours incubation with 5  $\mu\text{Ci}$   $^{14}\text{C}$ -actate under 37°C, lipids extracted due to Folch, and esterified with  $\text{BF}_3$  (Metcalf) and separated by gas liquied gaschromatography. Radio-

activity were determined by liquied scintillation spectrometer. Fractionation into major lipids were performed after Hanahan, Borgstrom and Metcalf with silisic acid column chromatography and so on.

Results as follow:

1) In hypoplastic anemia, radioactivity of total fatty acid decreased in DPM per  $10^6$  white blood cells and bone marrow cells compared with control. Percentage of  $^{14}\text{C}$  in each fatty acid increased in myristic and palmitic acids, and decreased in 20 carbons and more

longer chains fatty acids. But under treatment of ACTH and corticosteroids, percentage of myristic and palmitic acids from whole blood decreased and conversely increased in 20 carbons and more longer chains fatty acids. However, metabolism of bone marrow cells were not affected by treatment of ACTH and corticosteroids.

2) After incubation, material were divided into N(Nucleus), M(Mitochondria) and S (Supernatant) by means of Schneider's gradient centrifugation method. Percentage of DPM of M fraction decreased strikingly in hypoplastic anemia compared with S fraction, on the contrary the one was equal with the other in leukemia and in control.

3) Studying of CPM incorporated into fatty acids from each fractions, radioactivities of myristic and palmitic fatty acids come out of M fraction were increased in hypoplastic anemia compared with in leukemia and in control. These results made it clear that incorporation into M fraction decreased and so mitochondrial elongation system which synthesized 20 carbons and more longer chains fatty acids, were suppressed.

4) Studying of  $^{14}\text{C}$  incorporation into each fatty acid in N,M, and S fractions, we found stearic and oleic acids increased in N frac-

tion, and 20 carbons and more longer chains fatty acids increased in M fractions. The fact shows 20 carbons and more longer chains fatty acids may be synthesized in mitochondrial pathway as Hennes' studies.

5) Percentage of  $^{14}\text{C}$  incorporation into myristic and palmitic acids from whole blood cells as well as bone marrow cells increased in hypochromic anemia, but stearic and oleic acids decreased, especially, oleic acid decreased remarkably.

6) Percentage of  $^{14}\text{C}$  incorporation into myristic and palmitic acids increased in hypochromic anemia as well as hypoplastic anemia, however,  $^{14}\text{C}$  incorporation into unsaturated fatty acids decreased in hypochromic anemia. So, ratio between  $^{14}\text{C}$  incorporation into saturated and unsaturated fatty acids was larger in hypochromic anemia than in hypoplastic anemia.

7) Studying of  $^{14}\text{C}$  incorporation into each fatty acid of major lipids extracted by Hanahan's method with silicic acid column chromatography, in hypoplastic anemia, stearic and oleic acids increased in every group of free fatty acid, glyceride fatty acid, cholesterol fatty acid and phospholipid fatty acid compared with control.

## Plasma Clearance of Radioactive Vitamin B<sub>12</sub> in Various Diseases

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The fate of intravenously injected radioactive vitamin B<sub>12</sub> has been studied in patients with pernicious anemia (PA), chronic myelocytic leukemia (CML), monocytic leukemia, aplastic anemia, hemolytic anemia, liver cirrhosis acute hepatitis, chronic hepatitis, myelitis, diabetes mellitus, nephrosis as well as in patients with no apparent hematologic and renal disorders.

A mixture of 0.8  $\mu\text{g}$ .  $^{57}\text{Co}$  labeled cyanocobalamin and 14.2  $\mu\text{g}$ . of nonradioactive cyanocobalamin was rapidly injected into the antecubital vein and 3 ml. of blood was withdrawn at frequent intervals for 24 hours fol-

lowing the injection. The blood sample and 24-hour urine were counted for  $^{57}\text{Co}$  radioactivity in a well-type scintillator. The total blood volume was estimated at 90 ml. per kg. of body weight in male and 80 ml. in female. In some of the patients, 30 units of hog intrinsic factor was orally given before and during the disappearance test.

The results obtained were as follows:

1) In normal subjects, there was an initial rapid loss of plasma radioactivity during the first 30 minutes, followed by a progressively slower loss of radioactivity in the next 2 hours with a final more horizontal component