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$^{99m}$Tc-H-D RBC as the spleen scanning agent is prepared by using reducing agent, $^{99m}$TeO$_4^-$ and heating (49.5°C, 15 minutes). The object of the present study is to evaluate the osmotic fragility of the membrane of $^{99m}$Tc-H-D RBC to evaluate the mechanism of the spleen sequestration of $^{99m}$Tc-H-D RBC. For this purpose the C.P.C. (Coil Planet Centrifuge) method was used. This new method provided us with more sensitive and quantitative information than the ordinary Parpart method on the osmotic fragility of the membrane of RBC.

The results were as follows: 1) The osmotic fragility of the membrane of $^{99m}$Tc-H-D RBC is remarkably elevated by the heating (15 or 30 minutes), whereas the difference between 15 and 30 minutes is not clearly seen. Clinically the agent heated 15 minutes is most suitable for spleen specific scan but the one heated 30 minutes isn’t suitable for the imaging of liver and spleen. 2) The addition of reducing agent and/or $^{99m}$TeO$_4^-$ also showed slight elevation of the osmotic fragility of the membrane. It is concluded that osmotic fragility is not the single factor of splenic sequestration of $^{99m}$Tc-H-D RBC.

Application of $^{99m}$Tc-Labelled Heat-Denaturaed Erythrocytes to Splenic Clearance Study

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Splenic clearance rate of denaturated erythrocytes has been measured usually with $^{51}$Cr-labelled cells to investigate splenic function. In this report, we examined if or not $^{99m}$Tc-labelled erythrocytes can be used for this purpose in association with spleen scintigraphy.

Erythrocytes were at first treated with stannous pyrophosphate in saline solution for 5 minutes at the room temperature and then incubated with pertechnetate. The labelled erythrocyte suspended in the saline were incubated at 49.0±0.5°C for 15 minutes. At the end of each step, erythrocytes were washed sufficiently with the saline.

On observation with scanning electron microscope, SEM, erythrocytes appeared just slightly spherocytic after the tinning procedure. Their osmotic resistance was slightly reduced as examined by coil planet centrifuge method.

After the heat-denaturation process, $^{99m}$Tc-labelled erythrocytes were more uniformly spherocytic than $^{51}$Cr-labelled ones which beared numerous protrusions and might be called echinocantho-spherocyte. Significant difference was not observed in the 50% hemolysing saline concentration between $^{99m}$Tc-heat-denatureted cells and $^{51}$Cr-cells which had been incubated at the same temperature for 45 minutes, by “Parpart’s method”, while the range of hemolysis was significantly more narrow in the former than in the latter. The disappearance curve of the former in the circulation was prone to simulate single exponential fashion. When the individual variation of the damage degree was corrected using osmotic fragility, average difference in clearance rate between these cells was only 4.3% of an average of normal clearance rate in 32 measurement in 30 cases.

From these results, it was concluded that tinning process brought about a slight damage on the cell membrane, which enhanced the labelling efficiency and made the denaturation effect more