Note

Relation between Blood Clearance and Hepatic Uptake of ^{99m}Tc -Phytate in Rats with Hepatic Injury[†]

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The relation among the blood clearance of ^{99m}Tc -phytate (^{99m}Tc -P), the hepatic uptake of ^{99m}Tc -P and the severity of hepatic injury was investigated by using the rats with carbon tetrachloride (CCl_4), D-galactosamine (Gal N), α -naphthylisothiocyanate (ANIT) or DL-ethionine (EthN) induced hepatic injury.

After the administration of CCl_4 , GalN or ANIT, serum GPT activity increased significantly with the increase of dose level, and the degree of this increase was in the order: GalN > CCl_4 > ANIT. However, the mild increase in serum GPT activity was observed after EthN administration. The blood clearance rate of ^{99m}Tc -P and the hepatic uptake ratio of ^{99m}Tc -P decreased with the increase of dose level after CCl_4 , GalN or ANIT administration, but significant changes were not found after EthN administration. The degree of decrease in the blood clearance rate of ^{99m}Tc -P was in the order: GalN $\approx$ CCl_4 > ANIT, and the degree of decrease in the hepatic uptake ratio of ^{99m}Tc -P was in the order: GalN $\approx$ CCl_4 > ANIT.

These results suggest that the disorder in the hepatocytes may be one of causes for inducing the decrease in the hepatic uptake of ^{99m}Tc -P, and the consequence of this decrease may induce the decrease in the blood clearance of ^{99m}Tc -P.

Key Words: technetium-99m-phytate, blood clearance, hepatic uptake, hepatic injury, serum glutamic pyruvic transaminase, carbon tetrachloride, D-galactosamine, α -naphthylisothiocyanate, DL-ethionine

1. Introduction

^{99m}Tc -phytate (^{99m}Tc -P) is a radioactive substance which has been widely used in a field of nuclear medicine¹⁾⁻³⁾ since it was developed in 1973⁴⁾. Intravenous administration of ^{99m}Tc -P causes immediate chelation with blood calcium, and this makes microcolloidal substance which is taken up by the reticulo-endothelial system (RES).⁴⁾ In addition, since ^{99m}Tc has an appropriately short physical half-life of about 6 hours and emits 89% of

γ rays at 141 keV but no β ray, this substance can be treated with safety and it is easy to handle animals treated with this substance. Then, we have attempted to apply ^{99m}Tc -P for the evaluation of hepatic dysfunction in laboratory animals, and reported that the blood clearance test of ^{99m}Tc -P showed a sensitive reaction for the acute hepatic dysfunction in rats, equally to serum transaminase test, serum bilirubin test or histological test⁵⁾⁻⁷⁾. Furthermore, we reported that a major part of ^{99m}Tc -P intravenously administered was taken up by the liver (approximately 92% of total activity in body), while the remainder of small amount by the spleen, kidneys, and so on in the normal rats⁵⁾.

[†] 肝障害ラットにおける ^{99m}Tc -フィチン酸の血液クリアランスと肝摂取の関係。柴田 浩, 久野友幸, 河野正宏: 山口大学農学部家畜薬理学教室, 753 山口市吉田1677-1。

However, the relation among the blood clearance of ^{99m}Tc -P, the hepatic uptake of ^{99m}Tc -P and the severity of hepatic injury has not been clarified. Then, by using the rats with carbon tetrachloride (CCl_4), D-galactosamine (GalN), α -naphthylisothiocyanate (ANIT) or DL-ethionine (EthN) induced hepatic injury, the blood clearance rate of ^{99m}Tc -P, the hepatic uptake ratio of ^{99m}Tc -P and serum GPT activity were examined.

2. Materials and Methods

Male Wistar strain rats weighing 220–260 g were used at the age of 7–8 weeks, and were allocated to each group consisting of 12 heads.

CCl_4 at dose of 0.05, 0.1, 0.25, 0.5 or 1.0 ml/kg, GalN at dose of 50, 100, 250, 500 or 1000 mg/kg, ANIT at dose of 10, 25, 50, 100 or 200 mg/kg, or EthN at dose of 100, 250, 500, 1000 or 2000 mg/kg were intraperitoneally administered to the rats⁵⁾. The blood clearance rate of ^{99m}Tc -P, the hepatic uptake ratio of ^{99m}Tc -P and serum GPT activity were examined 48 hours after the administration of each dose, respectively.

The blood clearance rate of ^{99m}Tc -P was represented by the *K*-value. Methods to prepare ^{99m}Tc -P, to measure the radioactivity of ^{99m}Tc -P in blood sample and to calculate the *K*-value were described in the previous papers^{5)–7)}.

Since the uptake ratio of ^{99m}Tc -P by the liver reached the maximum 15 minutes after the intravenous administration⁵⁾, the value of uptake ratio at 15 minutes was employed as an index in the examination of hepatic uptake of ^{99m}Tc -P.

To assess the degree of hepatic injury, serum GPT activity was determined by the pyruvate oxidase method (Wako Pure Chemical Industries, Ltd.).

3. Results

After the administration of CCl_4 , GalN or ANIT, serum GPT activity increased significantly with the increase of dose level, and the degree of this increase was in the order: GalN > CCl_4 > ANIT (Tables 1, 2, 3). However, the mild increase in serum GPT activity was

Table 1 Dose-dependent changes of blood clearance rate of ^{99m}Tc -P, hepatic uptake ratio of ^{99m}Tc -P and serum GPT activity after CCl_4 administration

CCl_4 (ml/kg)	<i>K</i> -value	UR(%)	GPT(KU)
Control	0.133 ± 0.012	93.2 ± 5.4	28.2 ± 5.2
0.05	0.089 ± 0.008	86.1 ± 3.9	33.2 ± 10.8
0.1	0.085 ± 0.007	78.1 ± 0.8	50.2 ± 18.1
0.25	0.065 ± 0.006	72.9 ± 3.1	103 ± 35
0.5	0.063 ± 0.005	69.0 ± 3.3	398 ± 76
1.0	0.062 ± 0.005	67.2 ± 6.3	954 ± 260

Values show mean $\pm$ S.D. of 12 rats.

Table 2 Dose-dependent changes of blood clearance rate of ^{99m}Tc -P, hepatic uptake ratio of ^{99m}Tc -P and serum GPT activity after GalN administration.

GalN (mg/kg)	<i>K</i> -value	UR(%)	GPT(KU)
Control	0.133 ± 0.012	93.2 ± 5.4	28.2 ± 5.2
50	0.116 ± 0.010	91.5 ± 4.9	32.2 ± 5.3
100	0.108 ± 0.010	81.9 ± 1.7	43.4 ± 7.3
250	0.094 ± 0.008	75.7 ± 1.5	620 ± 100
500	0.068 ± 0.007	72.0 ± 1.0	1300 ± 412
1000	0.051 ± 0.005	66.7 ± 1.9	1900 ± 490

Values show mean $\pm$ S.D. of 12 rats.

Table 3 Dose-dependent changes of blood clearance rate of ^{99m}Tc -P, hepatic uptake ratio of ^{99m}Tc -P and serum GPT activity after ANIT administration

ANIT (mg/kg)	<i>K</i> -value	UR(%)	GPT(KU)
Control	0.133 ± 0.012	93.2 ± 5.4	28.2 ± 5.2
10	0.122 ± 0.010	90.7 ± 5.7	36.3 ± 9.9
25	0.105 ± 0.010	90.1 ± 1.6	42.6 ± 10.3
50	0.094 ± 0.008	86.0 ± 1.8	111 ± 20
100	0.075 ± 0.006	83.9 ± 2.7	444 ± 80
200	0.060 ± 0.005	81.3 ± 2.5	460 ± 98

Values show mean $\pm$ S.D. of 12 rats.

observed after EthN administration (Table 4).

The blood clearance rate of ^{99m}Tc -P (*K*-value) and the hepatic uptake ratio of ^{99m}Tc -P (UR) decreased with the increase of dose level after CCl_4 , GalN or ANIT administration, but significant changes were not found except for the tendency of decrease in them after EthN administration (Tables 1, 2, 3, 4).

The degree of decrease in the blood clearance rate of ^{99m}Tc -P was in the order: GalN

Table 4 Dose-dependent changes of blood clearance rate of $^{99m}\text{Tc-P}$, hepatic uptake ratio of $^{99m}\text{Tc-P}$ and serum GPT activity after EthN administration

EthN (mg/kg)	K-value	UR(%)	GPT(KU)
Control	0.133±0.012	93.2±5.4	28.2± 5.2
100	0.133±0.012	92.7±1.2	28.4± 6.6
250	0.128±0.011	88.7±1.8	29.3± 6.8
500	0.128±0.010	88.9±1.6	36.8± 9.2
1 000	0.124±0.010	86.8±2.0	45.1± 9.6
2 000	0.116±0.010	85.3±2.1	50.8±10.7

Values show mean±S.D. of 12 rats.

≡ $\text{CCl}_4 > \text{ANIT}$, and the degree of decrease in the hepatic uptake ratio of $^{99m}\text{Tc-P}$ was in the order: $\text{GalN} \equiv \text{CCl}_4 > \text{ANIT}$ (Tables 1, 2, 3).

4. Discussion

To induce hepatic injury, four types of hepatotoxins, CCl_4 which causes degeneration and necrosis in hepatocytes⁹⁾, GalN which causes hepatocellular necrosis and fatty liver that closely resemble human viral hepatitis^{10),11)}, ANIT which causes not only marked hyperbilirubinemia and cholestasis but also hepatocellular disorder^{12),13)}, or EthN which induce inhibition of hepatic protein synthesis and fatty liver¹⁴⁾, were administered to rats in this study.

The present study has made it clear that the blood clearance rate of $^{99m}\text{Tc-P}$ and the hepatic uptake ratio of $^{99m}\text{Tc-P}$ decreased with the increase in serum GPT activity. Since GPT, which is normally found in both the cytosol and mitochondria of hepatocytes, is a serum enzyme marker for hepatic injury, the degree of increase in serum GPT level indicate the degree of the disorder in the hepatocytes. However, the relation between the hepatocellular disorder and liver RES function has not been clarified. A clear correlation between the alteration of RES activity and the degree of hepatic injury in both CCl_4 and EthN treated mice was not noted^{15),16)}, while altered RES function contributed to the development of GalN -induced hepatocellular necrosis in rats^{17),18)}. In addition, the blood clearance rate of $^{99m}\text{Tc-P}$ may be affected not only by the RES function but by intravascular

coagulation and fibrinolysis, platelet aggregation and disaggregation and blood flow through the RES^{19),20)}. Therefore, the blood clearance of $^{99m}\text{Tc-P}$ may reflect the hepatic function in a broad sense, including the RES function.

Finally, the results in our study suggest that the disorder in the hepatocytes may be one of causes for inducing the decrease in the hepatic uptake of $^{99m}\text{Tc-P}$, and the consequence of this decrease may induce the decrease in the blood clearance of $^{99m}\text{Tc-P}$.

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