Residual Pathologic Changes and $^{99m}$Technetium Pyrophosphate Uptake Following Coxsackievirus B3 Perimyocarditis in Mice

Akira Matsumori, Kazunori Kadota, Hirofumi Kambara, Chuichi Kawai

The diagnosis of viral myocarditis is often difficult because the clinical presentation of viral myocarditis shows wide variations ranging from total absence of clinical manifestations to sudden unexpected death. Recently, we have found a high incidence of

Fig.1. Ninety days after inoculation with coxsackievirus B3. Marked fibrosis with calcification was noted in the right ventricle. A high $^{99m}$Tc-PYP uptake ratio (0.49) was shown in the heart. Hematoxylin and eosin stain. $\times$120.

Key Words:
$^{99m}$Technetium pyrophosphate
Perimyocarditis
Coxsackievirus B3

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severe perimyocarditis in the right ventricle in weanling BALB/c mice inoculated with coxsackievirus B3 (Nancy strain). Using this experimental model, we studied myocardial uptake of technetium-99m stannous pyrophosphate (99mTc-PYP) and found that 99mTc-PYP myocardial scintigraphy may be useful when making a clinical diagnosis of viral perimyocarditis. This report describes studies of 99mTc-PYP uptake in experimental viral perimyocarditis on the 3rd–90th day after inoculation with the virus.

METHODS

Induction of Experimental Viral Perimyocarditis

Methods are the same as used for the study of experimental coxsackievirus B3 perimyocarditis. The animals were sacrificed 3, 5, 7, 14, 28, and 90 days after the inoculation with coxsackievirus B3.

Three to four week old mice serving as the controls were inoculated intraperitoneally with 0.1 ml of virus-free HeLa cell culture fluid and sacrificed seven days after the inoculation.

Distribution Studies of 99mTc-PYP

Seventeen mice treated with virus-free control fluid and 154 mice inoculated with coxsackievirus B3 were sacrificed and organs were excised one hour after the injection of 10 μCi of 99mTc-PYP through a vein in the tail. Tissue uptake of the tracer in the skull, heart, lung, liver, kidney, spleen and blood was counted in a well type gamma scintillation counter as count/min per gram of tissue. The concentration of the tracer was calculated by the ratio of cpm/gm for the tissues to cpm/gm for the skull.

RESULTS

Tissue Distribution Studies

99mTc-PYP uptake ratio of 17 control mice was 0.074 ± 0.023 (mean ± SD) in the blood, 0.036 ± 0.021 in the heart, 0.067 ± 0.044 in the lung, 0.044 ± 0.011 in the liver, 0.024 ± 0.006 in the spleen, 0.023 ± 0.016 in the pancreas and 0.264 ± 0.091 in the kidney.

99mTc-PYP Uptake Ratio of the Heart

The uptake ratio of 99mTc-PYP of hearts inoculated with coxsackievirus B3 did not increase before the third day (Table I). 99mTc-PYP ratio of hearts with positive lesions appeared to increase five days after virus inoculation, reached a maximum on the seventh day and began to decrease on the 14th day. However, on the 90th day, 99mTc-PYP ratio remained high in mice with severe perimyocardial lesions with calcification.

TABLE I 99mTc-PYP UPTAKE RATIO OF THE HEART IN CONTROL AND INOCULATED MICE

<table>
<thead>
<tr>
<th>Days after inoculation with coxsackievirus B3</th>
<th>Controls</th>
<th>5</th>
<th>14</th>
<th>28</th>
<th>90</th>
</tr>
</thead>
<tbody>
<tr>
<td>mean ± SD</td>
<td>0.333 ± 0.194</td>
<td>0.352 ± 0.386</td>
<td>0.327 ± 0.576</td>
<td>0.383 ± 2.310</td>
<td>0.139 ± 0.350</td>
</tr>
<tr>
<td>range</td>
<td>0.111–0.670</td>
<td>0.130–2.130</td>
<td>0.030–0.550</td>
<td>0.004–0.215</td>
<td>0.004–0.771</td>
</tr>
<tr>
<td>No. of animals</td>
<td>n = 16</td>
<td>n = 25</td>
<td>n = 20</td>
<td>n = 20</td>
<td>n = 20</td>
</tr>
</tbody>
</table>

| Hearts with positive pathologic lesions | mean ± SD | 0.035 ± 0.009 | 0.036 ± 0.013 | 0.041 ± 0.018 | 0.041 ± 0.028 | 0.042 ± 0.055 |
| range | 0.021–0.064 | 0.017–0.068 | 0.007–0.075 | 0.005–0.093 | 0.005–0.092 |
| No. of animals | n = 19 | n = 17 | n = 14 | n = 15 | n = 14 |

| Hearts with negative pathologic lesions | mean ± SD | 0.024 ± 0.007 | 0.025 ± 0.010 | 0.033 ± 0.016 | 0.034 ± 0.022 | 0.036 ± 0.050 |
| range | 0.018–0.055 | 0.015–0.055 | 0.015–0.055 | 0.009–0.055 | 0.009–0.055 |
| No. of animals | n = 11 | n = 10 | n = 9 | n = 10 | n = 10 |

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Fig. 2. Ninety days after the inoculation (The same section of Fig. 1). Von Kossa positive calcification was shown. Von Kossa stain. × 120.

(0.225 ± 0.169, range 0.06 - 0.49). Figures 1 and 2 show the heart of a mouse with maximum $^{99m}$Tc-PYP ratio of 0.49.

**DISCUSSION**

Previous studies in experimental animals and man have indicated that $^{99m}$Tc-PYP is an extremely sensitive technique for detecting acute myocardial infarction. Our previous studies demonstrated that measurement of $^{99m}$Tc-PYP uptake of the heart is an extremely sensitive technique for detecting acute viral perimyocarditis in the experimental animal. $^{99m}$Tc-PYP uptake ratio of the heart began to increase five days after virus inoculation when myocardial necrosis was evident. On the seventh day, $^{99m}$Tc-PYP ratio reached a maximum and histologically, fine dystrophic calcification was seen in the necrotic fibers. After the 14th day, $^{99m}$Tc-PYP ratio began to decrease. However, on the 90th day, $^{99m}$Tc-PYP ratio remained high in mice with severe pathologic lesions.

Olson and co-workers reported a positive $^{99m}$Tc-PYP scintigram in patients with acute pericarditis, and Oka and co-workers reported a case of coxsackie B4 myocarditis with a positive $^{99m}$Tc-diphosphonate scintigram. A positive scintigram has also been reported in a case of cholesterol pericarditis and in a case of pericarditis following acute myocardial infarction. Other workers have described normal $^{99m}$Tc-PYP images in patients with acute pericarditis. However, it cannot be ruled out that the disease was inactive or in a healing phase at the time of study.

The marked sensitivity of $^{99m}$Tc-PYP uptake of viral perimyocarditis, at least in the experimental model, offers great promise as a reliable noninvasive method for detecting this process. The present findings provide further basis upon which $^{99m}$Tc-PYP imaging may be applied to perimyocarditis in humans.

**SUMMARY**

The myocardial uptake of technetium-99m pyrophosphate ($^{99m}$Tc-PYP) in perimyocarditis induced by coxsackievirus B3 in BALB/c mice was studied on 3rd-90th day after the inoculation. $^{99m}$Tc-PYP uptake ratio, measured by the ratio of cpm/gm for the heart to cpm/gm for the skull, began to increase five days after virus inoculation and reached a maximum on the seventh day. After the 14th day, $^{99m}$Tc-PYP ratio began to decrease, however, on the 90th day, a high $^{99m}$Tc-PYP uptake was shown in mice with severe perimyocardial fibrosis and
calcification.

The present findings may provide a further basis upon which $^{99m}$Tc-PYP imaging may be applied to viral perimyocarditis in humans.

REFERENCES


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