The Macaque Monkey as an Experimental Paratenic Host for *Paragonimus westermani* (Kerbert, 1878) Braun, 1899

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Five macaque monkeys, three *Macaca fascicularis*, one *M. fuscata*, and one *M. cyclops* were inoculated orally with 120 metacercariae of *Paragonimus westermani* and they were necropsied 70 to 400 days after infection. None of the infected monkeys became ill or manifested apparent clinical signs of paragonimiasis, although all the animals examined showed marked peripheral blood eosinophilia. The eosinophilia appeared soon after inoculation, reached peak by the 5th or 7th week, remained on a high level for about ten weeks, and thereafter declined gradually. The recovery of the inoculated metacercariae varied from 35.0 to 51.7%, with an average of 44.2%. All worms recovered were on the youngest developmental stage and 93.2% (88.1 to 100%) of them apparently remained in a slightly developed stage in the skeletal muscles, irrespective of the species of the monkey. These flukes of the youngest stage recovered from the muscles of monkeys were infective for cats and 74.3% (60.0 to 90.0%) of the worms given orally developed into mature adults capable of producing normal eggs in the feces of the recipient cats 41 to 66 days after transfer. Thus, it was found that the three species of macaques were the paratenic host of *P. westermani* under laboratory conditions. The gross lesions which might be attributable to migration of immature *P. westermani* and the histopathologic findings in the experimental monkeys are described and illustrated. —*Key words*: Lung fluke, Migration, Monkey.

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In the recent studies on the geographical distribution of *Paragonimus westermani* (Kerbert, 1878) Braun, 1899 in the southern prefectures of the Kinki district, Japan, the authors have found several prevalent areas of metacercariae of this lung fluke in two prefectures, Mie and Nara [9, 10]. The availability of the metacercariae spurred the authors to investigate the susceptibility of the monkey to *P. westermani* infection and biological features of host-parasite relationships since until recently the fluke has been considered to be a rare parasite of macaques [3]. The present study was carried out to study the infection, the growth and development of the parasites, the clinical effects, the location, and the lesions produced. Additionally, the role of macaques as an experimental paratenic host was investigated by exposing noninfected cats to immature *P. westermani* recovered from the monkeys 70 to 150 days after infection.

**MATERIALS AND METHODS**

*Source of metacercariae of P. westermani*: Metacercariae of *P. westermani* were harvested from the visceral organs and body muscles of the crab, *Geothelphusa dehaani*, which were collected from several prevalent areas in Mie and Nara [9, 10] prefectures.

*Monkey hosts*: Five monkeys of the genus *Macaca* were obtained from local zoological gardens in Osaka Prefecture. Three of them belonged to *Macaca fascicularis* (crab-eating macaque), one to *M. fuscata* (Japanese macaque), and the other one to *M. cyclops* (Formosan rock macaque). Upon arrival, they were kept isolated individually and their sto-
ols were examined for eggs of intestinal parasites. They were fed pelleted commercial monkey feed with fruit supplements and water *ad libitum*. When they adjusted themselves to the new environment and became free from eggs of parasites, they were subjected to the experiment.

**Inoculation:** All the monkeys were infected by feeding a small piece of banana in which freshly isolated 120 *P. westermani* metacercariae had been embedded. As all feed was withheld for at least 16 hours, the infective material was consumed within a few minutes.

**Clinical examinations:** All the monkeys were regularly observed for changes in their feed intake, behavior, and general health.

**Hematological examinations:** About 1 ml of blood was withdrawn into a sterile syringe from the cephalic vein before and at weekly intervals after infection. Blood preserved in EDTA-stopped tubes was examined with a “Toa” microcell counter for total leucocyte (WBC) and erythrocyte (RBC) counts. Determinations for packed cell volumes (PCV), and total plasma proteins were done by routine, standardized techniques. Differential leucocyte counts were made on Giemsa-stained smears; 400 leucocytes were identified, and the absolute number of each type per µl of blood was calculated.

**Postmortem examinations:** Routine post-mortem examinations were carried out on each monkey shortly after lethal intravenous injection of barbiturate at the end of the experiment. Gross lesions which might be attributable to parasitic invasion were recorded. For histological studies, selected tissue samples including those with the gross lesions were subjected to routine histological technique and stained with hematoxylin and eosin.

**Recovery of flukes:** The procedure of recovering flukes from the monkeys has been described [2, 7]. After the removal of the gastrointestinal tract, liver, pancreas and spleen from the peritoneal cavity, the lungs and heart were removed from the pleural cavity. These organs were rinsed with warmed saline (0.85% NaCl, 37°C) in an attempt to remove any *P. westermani* which might have been adhered to the surface. The serosal surfaces of the peritoneal and pleural cavities were then flushed repeatedly with warmed saline. These washings were kept in separate sedimentation beakers for later examination for flukes. After evisceration, the rest of the carcass was divided into six portions; (1) the head and neck, (2) the thoracic limbs, (3) the pelvic limbs, (4) the anterior half region of the trunk, (5) the posterior half region of the trunk, and (6) the skin with underlying tissues. To recover immature worms of the fluke from the liver, heart, lungs, diaphragm, skeletal muscles, and skin with underlying tissues, these organs and tissues were cut into 2-to 4-mm thick and separately kept in warmed saline at 37°C for 12 hr. The solution was drawn off into flat, marked petri dishes and examined for worms under a dissecting microscope. The flukes recovered were preserved in saline (0.85% NaCl) at room temperature and counted. The worms found in histological sections from the blocks of various organs and tissues were included in the total of each region.

**Morphological observations of immature *P. westermani***: Part of the immature worms recovered were compressed between two slide glasses before being fixed with 70% alcohol. For morphological observation, they were stained with borax carmine and mounted with balsam.

**Transfer of immature *P. westermani* to cats:** Part of the immature worms recovered from the muscle tissues of monkeys were fed to cats as early as possible: two cats were fed each ten worms from monkey No. 1, two cats, each ten to 20 worms from monkey No. 4, and one cat, 20 worms from monkey No. 2. Prepatent periods were determined by examining the feces of these cats from about 35 days after infection. All cats were killed
and necropsied 90 to 250 days after infection for recovery of adult flukes from the lungs and pleural cavity.

RESULTS

1. Clinical findings

No apparent clinical signs of paragonimiasis were observed in any of the infected monkeys. The animals remained in good clinical condition and showed no evidence of inappetence or coughing throughout the course of the experiment. No excretion of Paragonimus eggs in the stool was indicated during the experimental period.

2. Hematological changes

Four monkeys were examined for changes in the hematological values. The total and differential WBC counts of two monkeys, Nos. 2 and 3, are presented in Figs. 1 and 2, respectively. The total WBC count ranged from 6,200 to 11,000 per μl in these four animals before infection. It increased gradually to a range of 12,400 to 24,300 per μl between the third and 14th week of infection. The absolute number of eosinophils began to increase three or four weeks after infection and continued to increase progressively up to about the seventh week of infection, i.e., from the preinfection value of 100 to 400 per μl to 1,200 to 3,900 per μl (No. 3; Fig. 2). Such increase remained relatively constant for about ten weeks. Subsequently, both the total WBC counts and the absolute eosinophil levels started to decline gradually, but remained on considerably high levels even at the end of the experiment.

In these four infected monkeys, there was no significant alteration in the RBC count, PCV value, total plasma protein concentration, nor in the monocyte and basophil counts during the period of observation. Similarly, no marked change was observed in absolute lymphocyte nor neutrophil count in all monkeys except No. 3, in which they revealed rather high levels throughout the experiment with week-to-week fluctuations.

3. Distribution of immature P. westermani in monkeys

![Graph showing hematological response of monkey No. 2 following inoculation with 120 P. westermani metacercariae.](image-url)

Fig. 1. Hematological response of monkey No. 2 following inoculation with 120 P. westermani metacercariae.
Fig. 2. Hematological response of monkey No. 3 following inoculation with 120 *P. westermani* metacercariae.

Table 1. Number and location of immature *P. westermani* recovered from tissues and organs of monkeys experimentally infected with 120 metacercariae

<table>
<thead>
<tr>
<th>No.</th>
<th>Species</th>
<th>Body weight at necropsy (kg)</th>
<th>Sex</th>
<th>Duration of infection (days)</th>
<th>% recovery</th>
<th>Total</th>
<th>Pleural cavity</th>
<th>Peritoneal cavity</th>
<th>Lungs</th>
<th>Head and neck</th>
<th>Thoracic limbs</th>
<th>Pelvic limbs</th>
<th>Anterior half region of trunk</th>
<th>Posterior half region of trunk</th>
<th>Posterior half region of trunk</th>
<th>Diaphragm</th>
<th>Liver</th>
<th>Heart</th>
<th>Skin with underlying tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>M. fascicularis</em></td>
<td>4.5</td>
<td>M</td>
<td>70</td>
<td>50.8</td>
<td>61</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>12</td>
<td>41</td>
<td>21</td>
<td>1</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td><em>M. fascicularis</em></td>
<td>2.5</td>
<td>F</td>
<td>150</td>
<td>51.7</td>
<td>62</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>8</td>
<td>4</td>
<td>12</td>
<td>34</td>
<td>2</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td><em>M. fascicularis</em></td>
<td>4.4</td>
<td>M</td>
<td>400</td>
<td>35.0</td>
<td>42</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>9</td>
<td>11</td>
<td>4</td>
<td>12</td>
<td>0</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td><em>M. fuscata</em></td>
<td>8.8</td>
<td>M</td>
<td>100</td>
<td>45.8</td>
<td>55</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>18</td>
<td>3</td>
<td>31</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td><em>M. cyclopes</em></td>
<td>7.9</td>
<td>M</td>
<td>100</td>
<td>37.5</td>
<td>45</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>11</td>
<td>7</td>
<td>27</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Overall</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>44.2</td>
<td>265</td>
<td>6</td>
<td>1</td>
<td>2</td>
<td>18</td>
<td>44</td>
<td>38</td>
<td>145</td>
<td>4</td>
<td>5</td>
<td></td>
<td></td>
<td>11</td>
</tr>
</tbody>
</table>

The organs examined and the number and location of the flukes recovered from five infected monkeys are shown in Table 1. The recovery of flukes was relatively high in the monkeys and remained on a comparatively high level until the end of the experiment; the total recovery of flukes was 44.2%, ranging from 35.0 to 51.7%. Of a total of 265 worms, 247 or 93.2% (88.1 to 100%) were recovered from the skeletal muscles. With regard to the distribution of worms in the carcass of the infected monkeys, the largest number of worms was always found in the musculature of the posterior half region of the trunk, irrespective of the species of the monkey and the duration of infection. The next largest number was found in the pelvic limb muscles or the musculature of the anterior
half region of the trunk. The numbers of immature worms recovered from other regions were six in the peritoneal cavity, five in the liver, four in the diaphragm, one in the pleural cavity, one in the heart, and one in the skin with underlying tissues. No worm was detected from the lung parenchyma of the infected monkeys.

4. Dimensions and morphology of immature *P. westermani*

All flukes recovered from various tissues of the infected monkeys were mobile in warmed saline. They were pale brown in color, leaf-like in appearance, and all in the youngest developmental stage as shown in Figs. 3 and 4. The excretory bladder was still a conspicuous structure due to its contents of excretory granules.

The measurements of worms are given in Table 2. The mean sizes of flukes obtained from the muscles of the posterior half region of the trunk in three cases were 732±142 μm×342±74 μm for specimens from monkey No. 1 killed 70 days after infection, 972±110 μm×452±30 μm for those from monkey No. 5 killed 100 days after infection, and 904±123 μm×509±57 μm for those from monkey No. 3 killed 400 days after infection.
Table 2. Measurements (mean±S.D.; range in brackets) in μm of *P. westermani* recovered from various tissues of monkeys

<table>
<thead>
<tr>
<th>Monkey No.</th>
<th>Duration of infection (days)</th>
<th>Location of worms</th>
<th>No. of worms measured</th>
<th>Length</th>
<th>Width</th>
<th>Transverse diameter</th>
<th>Longitudinal diameter</th>
<th>Transverse diameter</th>
<th>Longitudinal diameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>70</td>
<td>Muscle of posterior half region of trunk</td>
<td>5</td>
<td>732±42(520-890)</td>
<td>342±74(230-450)</td>
<td>76±16(55-98)</td>
<td>100±17(80-118)</td>
<td>105±8(93-113)</td>
<td>108±18(80-125)</td>
</tr>
<tr>
<td>5</td>
<td>100</td>
<td>Muscle of posterior half region of trunk</td>
<td>5</td>
<td>972±110(840-1090)</td>
<td>452±30(420-500)</td>
<td>94±6(83-98)</td>
<td>109±5(103-118)</td>
<td>136±39(100-170)</td>
<td>133±15(115-143)</td>
</tr>
<tr>
<td>3</td>
<td>400</td>
<td>Muscle of posterior half region of trunk</td>
<td>9</td>
<td>904±123(770-1100)</td>
<td>509±57(440-620)</td>
<td>128±10(113-143)</td>
<td>147±17(133-175)</td>
<td>154±18(138-185)</td>
<td>170±30(150-203)</td>
</tr>
<tr>
<td>3</td>
<td>400</td>
<td>Liver</td>
<td>2</td>
<td>940(930-950)</td>
<td>520(500-540)</td>
<td>119(108-130)</td>
<td>136(133-138)</td>
<td>147(145-148)</td>
<td>164(163-165)</td>
</tr>
<tr>
<td>3</td>
<td>400</td>
<td>Skin with underlying tissue</td>
<td>1</td>
<td>920</td>
<td>530</td>
<td>128</td>
<td>133</td>
<td>128</td>
<td>183</td>
</tr>
</tbody>
</table>

Fig. 5. Ventral surface of the lungs of monkey No.4 killed 100 days after infection, showing a pleural lesion similar to a worm cyst, about 0.7 cm in diameter, in the right caudal lobe (arrow). Thickening of the pulmonary pleura and adhesions between the lung lobes are also seen.

Infection. No significant difference in length or width was found among worms from these three monkeys. Similarly, no marked difference in diameter of the oral or ventral sucker was detected among flukes from these three monkeys, although dimensions of the former were always smaller than those of the latter. In 22 mounted specimens examined, the primordium of the ovary, 22–124 μm×8–62 μm, was observed.

Fig. 6. Pleural microabscess with a core of necrotic eosinophils in the lung of monkey No. 1 killed 70 days after infection. Fibrous thickening of the pulmonary pleura with connective tissue tags containing numerous eosinophils is also noted. HE stain. ×20.
μm in size, was lying just posterior to the ventral sucker and the testicular primordia, 8–66 μm×4–26 μm in size, were found adjacent to the excretory bladder about midway between the ventral sucker and posterior end of the body. The average length of these flukes showed only 1.1- to 2.3-fold increase from that of excysted metacercariae (av., 472 μm) of this trematode which were obtained from its second intermediate host, G. dehaani.

5. Pathological features in monkeys

On gross and microscopic examinations, neither fluke nor worm cyst formation was seen in the pulmonary parenchyma of the infected monkeys; however, lesions of various degrees were present in the pleurae, heart, liver and flank, which might be attributable to migration of immature P. westermani. These pathological changes are described below.

*Pulmonary pleura:* On gross examinations, thickening of the pulmonary pleura with adhesions of the lungs to the thoracic wall and between the lung lobes, and focal fibrin deposits were present in four monkeys, Nos. 1 to 4. These lesions in two animals, Nos. 2 and 3, were less extensive than those in the other two, Nos. 1 and 4. No macroscopic change was observed in the pleura of monkey No. 5. In addition, a rust-colored nodular lesion, about 0.7 cm in diameter, with overlying yellowish-brown fibrinous exudate was present on the ventral surface of the caudal lobe of the right lung of monkey No. 4 (Fig. 5). A pleural nodular lesion similar to a worm cyst, approximately 0.6 cm in diameter, was also seen on the dorsal surface of the same lung lobe of monkey No. 1. Microscopic examinations revealed that these two nodular pleural lesions were eosinophilic microabscesses consisting of a central area of necrotic eosinophil debris surrounded by granulation tissue with eosinophils (Fig. 6). In the corresponding alveolar parenchyma immediately beneath the pleura, a few minute areas of eosinophilic pneumonitis with multinucleated giant cells and epithelioid cells
were also noted in two monkeys, Nos. 1 and 4. No other abnormality was observed in the pulmonary parenchyma. Immature flukes or migratory tracks were unable to be detected in spite of careful examinations of many sections from the lungs of all the experimental monkeys. In accordance with macroscopic observations, the mesothelial cells were hyperplastic and this finding, together with fibroplasia and inflammatory cell infiltration, resulted in various degrees of thickening of the pulmonary pleura in two monkeys, Nos. 1 and 4. Their pleurae were partly covered with thick projections of granulation tissue containing numerous eosinophils, or fibrin strands.

Diaphragmatic pleura: On gross examinations, pleural foci of fibrous thickening, 0.5 to 5.0 cm in diameter, with fibrin deposits were found on the thoracic surface of the diaphragm of all infected monkeys. In monkeys Nos. 3 and 4, in which such changes were more extensive, there were a small number of rust-colored spots 0.4 to 0.6 cm in diameter, on the diaphragmatic pleura of the boundary between the tendinous and muscular portions of the diaphragm (Fig. 7). Microscopic examinations of these spots showed proliferation of granulation tissue, containing mass of eosinophils and scattered lymphocytes, plasma cells, pigment-laden macrophages and multinucleated giant cells, on the diaphragmatic pleura and between the serosa and tendon fibers or muscle bundles. No migrating lung fluke was found in the sections of diaphragms from all the experimental monkeys.

Heart: Macroscopically, no evidence of cardiac invasion by *P. westermani* was found in any monkey except monkey No. 1, in which one barely discernible grayish-white focus was detected underneath the epicardium of the left ventricle. Microscopically, the lesion consisted of a focal migratory track, containing a cross section of an immature
fluke surrounded primarily by eosinophils and red blood cells with a few macrophages and lymphocytes. At the peripheral zone of the track, an almost complete rim was formed by necrotic cardiac muscle cells, but no proliferation of granulation tissue was seen at the junction between the normal parenchyma and the necrotic tissue (Fig. 8).

Liver: Macroscopically, relatively small numbers of hemorrhagic spots up to 0.5 cm in diameter and irregular lines of various length were visible on the external and cut surface of the livers of four monkeys, Nos. 1 to 4. In addition, a few circular whitish fibrous lesions, 0.2 to 0.4 cm in diameter, were observed in the hepatic capsule of three monkeys, Nos. 1, 2 and 4 (Fig. 9). Such macroscopic changes of the livers were less pronounced in two animals, Nos. 1 and 3, then in the other two, Nos. 2 and 4. No macroscopic lesion was detected in the liver of monkey No. 5. On microscopic examinations, the main pathologic changes were the formation of focal migratory tracks containing necrotic eosinophilic debris, eosinophils, red blood cells, fibrin and even serous fluid. Within the track, a cross section of immature *P. westermani* was detected by histologic examinations of several blocks of the liver from monkey No. 2 (Fig. 10). The walls of the tracks consisted mainly of degenerated and necrotic hepatic cells with or without infiltration of eosinophils. Additional findings were thickening of the portal triads due to hyperplasia of fibroblasts and large aggregates of eosinophils, and focal fibroplasia with eosinophil infiltration in the hepatic capsule.

Flank: Macroscopically, only a few whitish-gray specks with short fibrous tags were found on the peritoneal surface of the flanks of two monkeys, Nos. 1 and 3. The flank of the former was slightly thick and edematous, while the structure of the latter appeared to be normal in its thickness. No special pathologic change was detected in the remaining three animals, Nos. 2, 4 and 5. On microscopic examinations, the most important feature was the formation of focal migrating tracks, containing necrotic eosinophilic debris, eosinophils, red blood cells and some macrophages. These tracks were located mainly in the intramuscular connective tissue, and surrounded by infiltration of eosinophils with edematically dissociated collagen fibers. By histologic examinations of several blocks of the flank from monkey No. 1, cross sections of two immature flukes were detected within the tracks (Fig. 11). Additional findings were thickening of the intramuscular connective tissue due to focal or diffuse hyperplasia of eosinophilic granulation tissue and inflammatory edema, and the formation of scattered foci of acute or chronic eosinophilic myositis.
Table 3. Results of experimental inoculation of cats with immature *P. westermani* recovered from monkeys 70 to 150 days postinfection

<table>
<thead>
<tr>
<th>No.</th>
<th>Body weight at necropsy (kg)</th>
<th>Sex</th>
<th>No. of immature worms transferred</th>
<th>Age of immature worms (days)</th>
<th>Donor monkey No.</th>
<th>Prepatent period (days)</th>
<th>Duration of infection (days)</th>
<th>% recovery</th>
<th>No. of adult worms recovered from</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.10</td>
<td>F</td>
<td>10</td>
<td>70</td>
<td>1</td>
<td>40</td>
<td>90</td>
<td>90.0</td>
<td>9</td>
</tr>
<tr>
<td>2</td>
<td>0.95</td>
<td>F</td>
<td>10</td>
<td>70</td>
<td>1</td>
<td>54</td>
<td>250</td>
<td>90.0</td>
<td>9</td>
</tr>
<tr>
<td>3</td>
<td>1.50</td>
<td>F</td>
<td>10</td>
<td>100</td>
<td>4</td>
<td>55</td>
<td>200</td>
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<td>4</td>
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<td>5</td>
<td>3.20</td>
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<td>2</td>
<td>65</td>
<td>110</td>
<td>75.0</td>
<td>15</td>
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</tbody>
</table>

6. Transfer of immature *P. westermani* from the monkeys to cats

A known number of immature *P. westermani* recovered from three monkeys, Nos. 1, 2 and 4, which were killed 70 to 150 days after infection were fed to five cats in order to examine for the survival, growth and egg-laying capacity of these worms in their suitable definitive hosts. The results showed that the transferred monkey worms matured, formed worm cysts in the feline lungs, and started to discharge eggs into the feces of their new hosts 41 to 66 days after transfer (Table 3). Sixty to 90.0% (av., 74.3%) of the immature flukes given were recovered from the lungs and pleural cavity of the cats as adult parasites 90 to 250 days after infection. Thus, the worm life cycle was completed in the cats, and the macaque monkey was defined as a paratenic host for the lung fluke, *P. westermani*.

**DISCUSSION**

The studies clearly demonstrated that a great majority of *P. westermani* metacercariae ingested by three species of monkeys of the genus *Macaca* migrated to the host muscle and remained there in a slightly developed state for at least 400 days, without showing apparent clinical signs of paragonimiasis in any of the monkeys. Furthermore, these flukes of the youngest stage recovered from the muscles of monkeys were infective to cats and the worms developed into mature adults discharging normal eggs in the feces of the recipient hosts. Thus, it was concluded that the macaque monkey is a paratenic host of *P. westermani* under laboratory conditions.

Miyazaki et al. [7] have shown experimentally that in two Japanese macaques metacercariae of *P. pulmonalis* (Baelz, 1880) Miyazaki, 1978 can survive in a slightly developed state for at least 200 days after infection. The vast majority of parasites in these two animals, with returns of 44.5 to 47.7%, occurred in the host muscles, principally those of the trunk and pelvic limbs, with only a few gaining entry into the pleural cavity and lung. They also demonstrated that these worms of the youngest stage were able to mature when fed to three dogs and one cat. Based upon these observations, they concluded that under laboratory conditions the Japanese macaque served as a paratenic host for *P. pulmonalis* as in the case of many other animals such as wild boars, pigs, rats, mice, hamsters, guinea pigs, rabbits and hens [1, 6]. The present results with *P. westermani* metacercariae and three species of macaque monkeys are very similar to their findings.

Recently, comparative studies on the development and preferred locations of *P. pulmo-
nalis from Kyushu, Japan, and P. westermani filipinus Miyazaki, 1978 from Leyte, the Philippines, were made in four crab-eating macaques [2]. In P. pulmonalis, only two adult flukes were found within a worm cyst formed in the lungs of one of three monkeys and a great majority of the parasites still remained in the skeletal muscles in a slightly developed state even 148 to 150 days after infection. The recovery rate of these youngest flukes from the host muscles was almost the same as that in the monkeys in the present studies with P. westermani metacercariae. In contrast, in P. w. filipinus, flukes were recovered usually from worm cysts in the lungs and about one-half of the flukes reached sexual maturity until the end of the experimental period (187 days postinfection).

From the previous observations [2] and the present results, it became clear that the development and preferred locations of P. w. filipinus in the crab-eating macaque were quite different from those of P. pulmonalis and P. westermani in the same species of the monkey. Nevertheless, the crab-eating macaque may be more important as the final host rather than as the paratenic host of P. westermani in Malaysia, since natural infection with the adult worms in the crab-eating macaque had been recorded in this area [4, 8]. It should be, however, emphasized that experimental infection studies may be essential to elucidate the nature of the host-parasite relationship in that mammal in Malaysia.

The gross lesions which might be attributable to migration of immature P. westermani, and the attendant histopathologic findings in the pleurae, heart, liver and flank of the monkeys were described and illustrated for the first time. The great majority of the metacercariae administered migrated directly from the intestinal lumen through the peritoneal cavity to the body muscles. They remained there in a slightly developed state until the end of the experiment, inciting an intense eosinophilic reaction around them (Fig. 11). Furthermore, it is considered that only a few parasites migrated from the intestinal lumen through the peritoneal and pleural cavities to the lungs. Although P. westermani was not recovered from the lungs of any of the five monkeys, an eosinophilic microabscess was found in the pulmonary pleura of each of two monkeys, Nos. 1 and 4. In the corresponding alveolar parenchyma immediately beneath the pleura, a few minute areas of eosinophilic pneumonitis with multinucleated giant cells and epithelioid cells were also noted in these two animals. Such histopathologic changes may indicate host response to the parasite penetrating to the lung from the pleural cavity. It seemed likely, therefore, that immature P. westermani reached the lungs but was destroyed by the intense tissue reaction.

The most significant hematological findings after experimental P. westermani infection were marked leucocytosis due primarily to an increase in number of eosinophils. It is considered that as in the case of many other tissue-invading helminths [5], migrating immature P. westermani stimulated the production and circulation of eosinophils for a long period of time, and the worms were always surrounded by a dense coat of eosinophils in the various organs and tissues, as was detected by histological examinations (Figs. 8, 10 and 11).

Based on the results of the present experiments, the authors wish to recommend that all macaque monkeys from endemic areas of P. westermani not only in Japan but also in foreign countries be screened for this fluke before use in biomedical research. Investigators who will intend to use the wild macaque monkey should be made aware of the occurrence of immature flukes in this animal, a paratenic host of P. westermani.

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要約

ウェステルマン肺吸虫 Paragonimus westermani (Kerbert, 1878) のパラテニックホスト（待機宿主）に関する実験的研究：サルについての検索成績：杉山 広・園田二朗・奥田 純・富村 保（大阪府立大学農学部家畜病理学教室）——ウェステルマン肺吸虫（両性生産卵型）に対する感受性。体内における虫体発育状況。寄生部位、および本虫感染にもとづくサルの病態を調べ、カニクイザル、ニホンザルおよびタイワザルがウェステルマン肺吸虫（両性生産卵型）のパラテニックホスト（待機宿主）になることを明らかにした。カニクイザル3頭、ニホンザル1頭、タイワザル1頭に本虫のメタサルリアをそれぞれ120日間投与し、70～400日目に剖検した。全例とも臨床症状、肺吸虫卵排出は認められなかったが、持続性の著明な好酸球増多症が観察された。虫体の回収率は35.0～51.7%（平均 41.2%）で、サルの種類に関係なく、回収虫体の大多数（平均93.6%)は、ほとんど未発育の状態で肺内、とくに胸壁および四肢から検出された。胸腔、腹腔、横隔膜、肝臓、心筋、皮膚、皮下組織などからも、数例ながら幼若虫が回収されたが、肺に虫卵形成が認められず、虫体も証明できなかった。これらの幼若虫を終宿主であるネコに経口投与すると、虫体の回収率は60～90%で、肺に虫卵の形成を認め、投与後41～66日間に糞便内排卵が開始した。