BRIEF NOTE

Transplantation of Intracranial Young Adult Angiostrongylus cantonensis into the Subdural Spaces of the Brains of Rats and Guinea Pigs

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Angiostrongylus cantonensis develops to sexual maturity in the pulmonary artery of rats. In several other mammals including man, however, the parasite develops only up to the young adult stage in the brain and shortly dies there [1]. In an attempt to better understand the migration and development of A. cantonensis in the normal and abnormal hosts, a simple method was developed for the surgical transfer of the intracranial young adult worms into the subdural spaces of the brains of small laboratory animals. The present study was also undertaken to determine whether the young adult worms introduced by this technique into the rats and guinea pigs migrate to the pulmonary artery and sexually mature.

The intracranial worms were collected from the brain surface of experimentally infected donor rats and mice, and washed in sterile minimum essential medium (Nissui Seiyaku, Tokyo) supplemented with 100 units/ml penicillin. Recipient animals were anaesthetized with sodium pentobarbital. The head of each animal was shaved and a longitudinal cut of approximately 3 cm was made in the skin covering the skull. A hole measuring 8×8 mm in size was made in the parietal portion of the skull with a motor-driven Model B-313 drill (Takahashi Shoten, Tokyo) and a longitudinal incision of approximately 7 mm was then made in the dura mater. Ten worms (5 males and females each) were placed carefully on the brain surface under the dura mater with a dissecting needle. Thenceforth, the dura mater was covered with a piece of Spongel® (Yamanouchi Seiyaku, Tokyo) and then, with a glass-ionomer cement Type I (G-C Dental Industrial Corp., Tokyo). The outer skin was finally closed with 4 to 5 separate stitches and the animal was injected intramuscularly with Chloromycetin (Sankyo Seiyaku, Tokyo), 100 mg per animal. The whole operation took about 25 min per animal.

The worm recovery and stool examination of the recipient animals as well as the measurements of the recovered worms were made as described previously [5]. The morphological observations of the worms were performed by the method of Ohmori and...
Table 1. Infection rates and worm recoveries of rats and guinea pigs which were transplanted into the subdural spaces of the brains with the intracranial worms from homologous and heterologous donors.

<table>
<thead>
<tr>
<th>Group</th>
<th>Donor</th>
<th>Recipient</th>
<th>Age (days) of worm at transfer</th>
<th>Day of necropsy after transfer</th>
<th>Infection rate</th>
<th>Worm recovery*</th>
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<tbody>
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<td></td>
<td></td>
<td></td>
<td>Operated</td>
<td>Examed</td>
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<tr>
<td>A</td>
<td>Rat</td>
<td>Rat</td>
<td>16</td>
<td>42-43</td>
<td>4**</td>
<td>3</td>
</tr>
<tr>
<td>B</td>
<td>''</td>
<td>''</td>
<td>23</td>
<td>35-36</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>C</td>
<td>Guinea pig</td>
<td>16</td>
<td>43</td>
<td>3†</td>
<td>1</td>
<td>0</td>
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<td>D</td>
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<td>23</td>
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<td>E</td>
<td>Mouse</td>
<td>Rat</td>
<td>14</td>
<td>44-45</td>
<td>5</td>
<td>5</td>
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<td>F</td>
<td>''</td>
<td>''</td>
<td>21</td>
<td>37-38</td>
<td>4¶</td>
<td>3</td>
</tr>
</tbody>
</table>

Remarks.
SD: Standard deviation.
*: Worm recovery was studied on the animals which survived throughout the course of the experiment.
**: One individual died 40 days post-transplant.
***: Figures outside parentheses mean the total number of worms and those in parentheses denote the male/female worms.
†: Two of them died 2 days post-transplant.
§: One individual died 25 days post-transplant.
¶: One individual died 31 days post-transplant.

Suzuki [3].

The intracranial worms obtained from the donors either 14 to 16 days or 21 to 23 days postinfection were transferred to both Wistar strain male rats weighing 320-445 g and Hartley strain female guinea pigs weighing 325-440 g. The mortality (50% = 3/6) of transplanted guinea pigs during the course of the observations was higher than that (13% = 2/15) of the rats (Table 1). Stool examinations revealed that all individuals of six groups were negative for A. cantonensis first stage larvae throughout the course of the study. At necropsy, however, it was found that two rats which had been transplanted with either 16-day old rat worms (Group A) or 14-day old mouse worms (Group E) harbored each a single adult worm in their pulmonary arteries (Table 1). A single female worm recovered from Group A could not be measured because it was injured when collected. This worm, however, was found to be sexually matured due to the presence of ova in the uterus (Fig. 1). Likewise, a single male worm from Group E, measuring 20.9 mm in length, was also considered mature since its seminal vesicle was filled with sperms (Fig. 2).

The present study clearly shows that if the young adult worms are introduced into the subdural spaces of the brains of normal rats, migration and adult establishment will ensue though the infection rate and worm recovery rate were extremely low. This study was also indicative that if the mouse worms are transferred to the rat, they are capable of migrating to the pulmonary artery, although such a migration does not occur in the mouse [2]. Additionally, it is of interest to note that the rats transplanted with 21 to 23-day old worms were negative for A. cantonensis infections (Table 1). The reason for this is still unknown. However, the younger worms may have an advantage in being transplanted more easily than the large ones. Another explanation is that the introduc-
tion of about 15-day old worms into the brain surface of recipient animal might constitute a prerequisite for completion of migration of the worms from the brain to the lung.

Our preliminary study indicated that in guinea pigs, the transferred worms failed to migrate to the pulmonary artery even when the young adult worms, normally developed in the rat brain, were transplanted (Table 1). Although further experimentation is of course required to confirm this, the above finding is quite interesting when one considers our another recent observation that *A. cantonensis* could not develop to sexual maturity in guinea pigs even when the young adult worms from the rat brain were directly transferred into the pulmonary arteries of the animals [6]. Thus, the results of these transplantation studies might provide experimental evidences to support partially the previous observations that in guinea pigs infected with *A. cantonensis*, the development of the parasite is analogous to that in rats until about 15 to 20 days postinfection, but the distinct delay of development occurs at later stages and the worms die shortly without migrating to the lung [2–4].

References


Explanation of Figures

Fig. 1. Female worm recovered from a rat transplanted with 16-day old rat worms, at Day 43 post-transplant. Note some ova in the uterus. ×650.

Fig. 2. Male worm recovered from a rat transplanted with 14-day old mouse worms, at Day 44 post-transplant. Note sperms in the seminal vesicle. ×650.