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Miscellanea

Distribution of *Toxoplasma* Cysts in the Central Nervous System of Slow Loris, *Nycticebus coucang*

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Faculty of Medicine, University of Singapore

Introduction

It is generally believed that in many animals *Toxoplasma* tend to encyst more frequently in the central nervous system than in other organs of the body. However, there is little information about the precise distribution of the cysts in various parts of the central nervous system itself. A recent work in which an assessment was made on these lines is by Verma & Bowles (1967) using mice as an experimental animal. We have, on the other hand, studied the distribution of the cysts in a primate and believe that these observations would give a better indication of the possible distribution of cysts in man.

The experimental animal used in the present investigation is the slow loris, a small primate confined to the South East Asian Region. In the adult animal the brain is 3.0–3.4 cm in length (Krishnamurti, 1966) and it is, therefore, possible to obtain serial sections of the whole organ without much difficulty. In a recent survey some serologically positive animals were detected and a strain of *Toxoplasma* was isolated from a slow loris (Zaman & Goh, 1968). This prompted us to study the whole of the central nervous system from an infected animal so as to map out the distribution of the cysts.

TABLE 1

*Distribution of toxoplasma cysts in the central nervous system of a slow loris*

Total number of cysts encountered 211 (7 in white matter, 204 in grey matter)

<table>
<thead>
<tr>
<th>Central Nervous System</th>
<th>Number of Cysts</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spinal cord</td>
<td>nil</td>
<td>nil</td>
</tr>
<tr>
<td>Hind brain</td>
<td></td>
<td></td>
</tr>
<tr>
<td>medulla</td>
<td>nil</td>
<td>nil</td>
</tr>
<tr>
<td>pons</td>
<td>nil</td>
<td>nil</td>
</tr>
<tr>
<td>cerebellum</td>
<td>1</td>
<td>0.47</td>
</tr>
<tr>
<td>Mid brain</td>
<td>9</td>
<td>4.27</td>
</tr>
<tr>
<td>Fore brain</td>
<td></td>
<td></td>
</tr>
<tr>
<td>diencephalon</td>
<td>74</td>
<td>35.07</td>
</tr>
<tr>
<td>sub cortical</td>
<td>34</td>
<td>16.11</td>
</tr>
<tr>
<td>nuclear masses</td>
<td></td>
<td>95.26</td>
</tr>
<tr>
<td>cerebral cortex</td>
<td>93</td>
<td>44.08</td>
</tr>
</tbody>
</table>

* Department of Parasitology.
** Department of Anatomy.
TABLE 2

Distribution of toxoplasma cysts in slow loris

<table>
<thead>
<tr>
<th>Cerebral Cortex</th>
<th>Number of Cysts</th>
<th>% of Total in Cerebral Cortex</th>
</tr>
</thead>
<tbody>
<tr>
<td>Olfactory lobe</td>
<td>7</td>
<td>7.53</td>
</tr>
<tr>
<td>Frontal lobe</td>
<td>33</td>
<td>35.48</td>
</tr>
<tr>
<td>Parietal lobe</td>
<td>28</td>
<td>30.11</td>
</tr>
<tr>
<td>Temporal lobe</td>
<td>9</td>
<td>9.68</td>
</tr>
<tr>
<td>Occipital lobe</td>
<td>16</td>
<td>17.20</td>
</tr>
</tbody>
</table>

Material and Methods

The infected slow loris was anaesthetized intraperitoneally with sodium pentobarbital (28 mg/kg of body weight) and was perfused with 0.9% saline followed by formol saline. The whole brain and the spinal cord were removed and embedded in celloidin, serially sectioned at 25 μ thickness in the coronal plan and stained with cresyl violet. The serial sections were studied microscopically in order to find out the distribution of Toxoplasma cysts in the entire central nervous system of this animal. The cyst size was measured with a Reichert Visopan.

Observations

The cysts appeared as dense bodies with sharply demarcated outline. The contents were granular representing the nuclei of the parasite. The size varied from 25 μ to 50 μ. The cysts in the grey matter were generally spherical (figs. 1–3) while those in the white matter were oval (fig. 4). The cysts took a deep violet colour and stood out clearly from the lighter background of the host tissue.

The total number of cysts found in the central nervous system of this animal was 211, of these 204 were in grey matter and 7 in white matter. Moreover, the distribution was mainly confined to the forebrain which accounted for 95.26% of the total number. Tables I and II show the total number of cysts encountered, their detailed distribution and their relative percentage in various parts of the central nervous system.

In addition to the presence of cysts which were clearly observed by this method of staining, there was a marked cellular infiltration of the meninges and the brain substance (fig. 5). The cellular infiltration in the brain was generally of a focal nature. There was also perivascular infiltration of the blood vessels throughout the brain.
Discussion

The remarkable feature of the distribution of the Toxoplasma cysts is that the cell bodies of the neurons rather than their processes appear to be the site of preference for encystation as revealed by the fact that out of 211 cysts encountered 204 were found in the grey matter while only 7 were located in the white matter. Moreover, the concentration of cysts is the maximum in the forebrain (95.26%), which is in accordance with the findings of Verma & Bowles (1967), who had found 80.97% of the cysts in the forebrain of the laboratory infected mice. In the forebrain itself the total number of cysts was the highest in the cerebral cortex. However, when the distribution was compared in relation to the size of the various areas of the forebrain, thalamus and hypothalamus exhibited the highest concentration. An additional feature of interest found in this study is a progressive decrease in the concentration of the cysts from forebrain (95.26%) to midbrain (4.27%) to hind brain (0.47%) to spinal cord (nil %). These findings, therefore, suggest that the parasite is more likely to interfere with the higher cortical as well as thalamic and hypothalamic functions rather than with the basic functions of the hind brain and the spinal cord.

Furthermore, the cerebral cortex, subcortical nuclei and the nuclear masses of the thalamic complex, which show heavy concentration of the cysts, are all supplied by the carotid system of the circle of Willis (Krishnamurti, 1968) while the cerebellum, midbrain, pons, medulla and spinal cord, which show sparse or no distribution of the cysts, are all supplied by the vertebral system of the circle of Willis. It is, therefore, likely that the infection has spread via the internal carotid arteries explaining the heavy concentration of the cysts in the forebrain.

The areas of focal cellular infiltration, seen very frequently in the brain substances, were never found around the cysts. This could very well mean that the parasite, so long as it is encysted, does not seem to elicit any tissue reaction from the host.

References


Fig. 1. Coronal section of the brain of slow loris showing *Toxoplasma* cyst in the Diencephalon. Cresyl violet. × 66. Inset × 200.

Fig. 2. Coronal section of the brain of slow loris showing *Toxoplasma* cyst in the subcortical nuclear masses. Cresyl violet. × 66. Inset × 200.
Fig. 3. Coronal section of the brain of slow loris showing *Toxoplasma* cyst in the cerebral cortex. Cresyl violet. $\times 66$. Inset $\times 200$.

Fig. 4. Coronal section of the brain of slow loris showing *Toxoplasma* cyst in the white matter. Cresyl violet. $\times 65$. Inset $\times 200$. 
Fig. 5. Coronal section of the brain of slow loris showing cellular infiltration. Cresyl violet. × 5. Inset × 34.