Seasonal Periodical Appearance of *Hepatozoon canis* Gamont in the Peripheral Blood

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(Received 22 December 1992/Accepted 7 June 1993)

**ABSTRACT.** We investigated the appearance of gamonts in peripheral leukocytes of 13 *Hepatozoon canis* infected dogs from June 1990 to December 1992. In almost all the dogs, the numbers of gamonts increased from spring to autumn, and decreased from autumn to winter. Particularly in the dogs used for hare hunting, a large number of gamonts was detected from spring to autumn. This finding reveals that the gamont increases in number in the dog which ingested the infected ticks during the hunting season. ---KEY WORDS: canine, *Hepatozoon canis*, seasonal periodicity.

*Hepatozoon canis* infection is one of the worldwide protozoan parasitic diseases of dogs [5]. *H. canis* infection occurs through India, South Africa, Nigeria, Israel, the Philippines, Malaysia, North America and Japan [2-4, 6, 8, 9, 11, 12]. In our observations, infected dogs had many gamonts in the peripheral leukocytes and the number of gamonts gradually decreased toward winter [9], so, we think there is possible seasonal periodical appearance of gamont in the peripheral blood. The seasonal occurrence of hepatocoenosis was also elucidated in rodents by field survey [7, 13].

We surveyed 11 of 58 apparently healthy infected dogs on the appearance of *H. canis* gamonts in the blood from June 1990 to December 1992 in Fukuoka and Yamaguchi prefectures. At the same time, 2 infected dogs were observed under the tick free condition.

Dog No. 1 was used for hare hunting in winter from 1990 to 1991, but not in the next winter from 1991 to 1992. Dogs No. 2, 3, 4, 5 and 6 were hunting dogs and used for hunting for 2 years. Dogs No. 8 and 9 were young and had not been used for hunting. Dogs No. 7 and 10 were hunting dogs but not used for hunting. Dog No. 11 was a house dog and not chained all day long during the investigation period.

Two naturally infected dogs (No. 12 and 13) with *H. canis* were kept strictly free from tick infestation in our laboratory. Dog No. 12 was observed from his birth to about 21 months of age. At 16 months of age, the dog was subcutaneously injected with prednisolone in a dose of 5 mg/kg B.W./day for 1 month, splenectomized at 20 months of age, and euthanatized with pentobarbital sodium for histopathological examination at 22 months of age. Dog No. 13 received a bone marrow biopsy under general anesthesia at 8 months of age.

White blood cells were counted by an automatic cell counter. The number of *H. canis* gamonts was estimated by examination of 2,000 leukocytes on peripheral blood smears stained with Giemsa at intervals of 3 to 13 weeks.

Large numbers of *H. canis* gamonts were observed in April in all the dogs except those kept free from tick infestation. Gamonts reached a maximal number (mean: 2,689.3/μl) from May to August and decreased in September to become a minimum number (mean: 14.2/μl) during winter from December to February. Dog No. 1 was parasitized with large numbers of gamonts in July 1990 but was not so in the summer of 1992 (Fig. 1a).

Dog No. 7, 10 and 11 and No. 8 and 9 (newborn puppies) had small numbers (mean: 226.1/μl) of gamonts from April to September 1990 (Fig. 1b).

Dog No. 2 showed a peak in the number of gamonts in November 1990 but had no gamont in November 1991. Injection of prednisolone and splenectomy had no influence on parasite burden of this dog. No meront was observed in the main visceral organs at autopsy. Dog No. 13 showed the appearance of a small number of gamonts from autumn of 1991 to summer of 1992 but no gamont was detected in September 1992. A mature meront was detected from this dog in the bone marrow. The number of gamonts did not differ between before and after biopsy (Fig. 1c).

White blood cell counts were almost constant in all the dogs throughout all seasons.

From the dogs No. 1, 2, 3 and 5, five species of ticks were collected, namely, *Haemaphysalis campanulata*, *H. longicornis*, *H. flava*, *Ixodes ovatus*, and *I. nipponensis*.

In the infected dogs used for hare hunting, the number of gamont apparently differed between summer and winter. On the other hand, the dogs not used for hare hunting and the house dogs showed slight increases in the number in the next summer. The appearance of gamonts in the blood differed between the dogs used and those not used for hunting because of the possible contact with infected ticks in the hunting season.

Nordgren and Craig [10] detected *H. canis* gamonts in dogs 3 weeks after the experimental exposure to tick infestation and one oocyst in only one of approximately 120 ticks. Our data showed that the number of gamonts in the blood of infected dogs seasonally differed between summer and winter, and that gamonts hardly appear in the blood in the hunting season. These findings suggest that there may be little chance for tick to be infected with *H. canis* by biting infected dogs in the hunting season, and also that dogs were infected with the parasite by ingesting infected ticks in the hunting season. It is also suspected that the appearance of gamonts in the blood synchronizes with the seasonal activity of ticks, and that the infected
Table 1. Dogs infected with *Hepatozoon canis*

<table>
<thead>
<tr>
<th>No.</th>
<th>Sex</th>
<th>Age</th>
<th>Breed</th>
<th>Hunting</th>
<th>Babesia</th>
<th>Remark</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>m</td>
<td>1y</td>
<td>b</td>
<td>y</td>
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<td>VDF</td>
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<tr>
<td>2</td>
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<td>3</td>
<td>m</td>
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<td>y</td>
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</tr>
<tr>
<td>4</td>
<td>f</td>
<td>1y</td>
<td>m</td>
<td>y</td>
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</tr>
<tr>
<td>5</td>
<td>f</td>
<td>6m</td>
<td>b</td>
<td>y</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>f</td>
<td>10y</td>
<td>b</td>
<td>y</td>
<td></td>
<td>+</td>
</tr>
<tr>
<td>7</td>
<td>f</td>
<td>6y</td>
<td>b</td>
<td>n</td>
<td></td>
<td>D(3)</td>
</tr>
<tr>
<td>8</td>
<td>m</td>
<td>2d</td>
<td>b</td>
<td>n</td>
<td></td>
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</tr>
<tr>
<td>9</td>
<td>f</td>
<td>31d</td>
<td>b</td>
<td>n</td>
<td></td>
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</tr>
<tr>
<td>10</td>
<td>f</td>
<td>6y</td>
<td>m</td>
<td>n</td>
<td></td>
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</tr>
<tr>
<td>11</td>
<td>m</td>
<td>13y</td>
<td>m</td>
<td>h</td>
<td></td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>m</td>
<td>26d</td>
<td>m</td>
<td>i</td>
<td>B, P, S&amp;A</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>f</td>
<td>6m</td>
<td>m</td>
<td>i</td>
<td>BMB</td>
<td></td>
</tr>
</tbody>
</table>

a) m: male, f: female.
b) Age at the first detection of the parasite; y: year, m: month, d: day.
c) b: beagle, m: mongrel.
d) Experience of hare hunting during the investigation; y: yes, n: no, b: house dog, i: isolated from other dogs and kept free from ticks.
e) with *Babesia gibsoni* infection before the investigation.
f) Ventral disk fenestration for intervertebral disk protrusion.
g) Delivery (times) during the investigation.
i) BMB: bone marrow biopsy.

The route of transmission could not be clarified in this survey, but elucidation of this is a very important problem in the future.

The seasonal periodical appearance of gamont might be caused by depression of host immunity, but dogs No. 12 and 13 strictly free from tick infestation also showed a slight parasitemia with gamonts through the year. Of these dogs, No. 12 showed a single peak of parasitemia in November 1990 possibly caused by bacterial colitis and from then on a continuous slight parasitemia appeared through his life. These two dogs (No. 12 and 13) were parasitized by no ticks and developed on clinical signs caused by reinfection after ingestion of infected ticks during the investigation period. On the other hand, the other infected dogs may develop strong immunity against *H. canis*, so the animals would show no severe clinical signs after the peak and seasonal appearances of gamonts in the peripheral blood and the reinfection with *H. canis*. The seasonal periodical appearance of *H. canis* gamont suggests that reinfection plays an important role in the appearance in the blood throughout the seasons. These problems remain to be experimentally proved.

In this investigation, *H. longicornis*, a vector tick of *Babesia gibsoni*, was collected from the dogs infected with *H. canis*. This shows that these species of tick will transmit both *H. canis* and *B. gibsoni* because many mixed infections with both *Hepatozoon* and *Babesia* have been reported [8, 11].

Canine hepatozoonosis has been described to produce severe clinical symptoms in 1985 [1] and was one of the
most important diseases of dogs. Our result suggests that the period from spring to autumn is suitable for practitioners to detect *Hepatozoon* infection in dogs.

**ACKNOWLEDGEMENTS.** We gratefully thank Dr. Kozo Fujisaki, National Institute of Animal Health, Ministry of Agriculture, Forestry and Fisheries, Japan, for identification of the ticks, and Dr. Magoshiro Maeda, Mine Animal Clinic, Mine-city, Yamaguchi, for collecting the materials, and Dr. Yukio Hara for discussion of hepatozoonosis and babesiosis.

**REFERENCES**