Heavy Infestation of the Sticktight Flea 
(*Echidnophaga gallinacea*) in Dog

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A 6-month-old female dog in Kanchanaburi province was heavily infested with the sticktight flea
(*Echidnophaga gallinacea*). The dog showed the signs of weakness, emaciation, as well as pale mucus membranes
and mild dehydration. Hematological analysis revealed moderate anemia, with 28.7% packed cell volume (normal
range 35-55%) and a $4.94 \times 10^6$ erythrocyte count (normal range $5-9 \times 10^6$). However, hemoparasites
and other ectoparasites such as ticks and lice were not found. The finding of this investigation is valuable for
control of the fleas in order to prevent adverse effects on animal health.

Keywords: Ectoparasite, *Echidnophaga gallinacea*, Flea, Dog
Introduction

Fleas (Insecta, Siphonaptera) are small, wingless, laterally compressed, and highly specialized insects. Adult fleas are obligate hematophagous ectoparasites of warm-blooded vertebrates that about 94% of the species infest mammals and the remaining infesting birds (Lewis, 1993).

The sticktight flea (Echidnophaga gallinacea) has been found worldwide and served as an important flea of poultry (Chandler and Read, 1961). It may also infest a wide variety of mammals including cats, dogs, rabbits, rats, insectivores and humans (Wall and Shearer, 1997; Durden and Hinkle, 2009). The flea is the cause of severe nuisance, irritation and allergic reactions in both animals and humans. Heavy flea infestation can cause severe anemia or even death in birds and rodents (Boughton et al., 2006; Cucchi-Stefanoni et al., 2008).

In Thailand, several studies of ectoparasites on dogs have been conducted (Sangvaranond, 1990, Sangvaranond et al., 2000, Nithikathkul et al., 2005; Nuchjangreed and Somprasong, 2007). There is only one reported document on the sticktight flea, E. gallinacea which infested dogs (Changbunjong et al., 2009). However, the studies of clinical symptoms and hematological profiles on dogs have not yet been reported. The present report describes heavy infestation of E. gallinacea in dog, thus provided valuable information for control of the fleas in the animal.
Materials and Methods

Dog and blood samples
A 6-month-old female dog living in the rural area of Kanchanaburi province was examined general health status by physical examination (body temperature, visible mucous membranes and state of dehydration). Blood sample was collected with an EDTA containing tube for hematological and parasitological studies.

Hematological studies
The hematological parameters including red blood cell count (RBCs), hemoglobin concentration (Hb), packed cell volume (PCV) as well as total and differential leucocyte counts were performed at the Veterinary Teaching Hospital, Mahidol University. The blood indices composting of mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated.

Parasitological studies
For ectoparasite examination, all ectoparasites were collected by hand or by using a fine pointed forceps and were preserved in 70% ethanol. Collected fleas were mounted in Hoyer’s medium (Krantz, 1978). They were identified to the species level by using available taxonomic keys of Furman and Catts (1982).

Giemsa stained thin blood smears were performed to examine hemoparasites and microfilariae. Additionally, hemoparasites, which include *Ehrlichia canis*, *Babesia* spp and *Hepatozoon canis* were also confirmed by the multiplex PCR.

Multiplex PCR to detect parasites
DNA extraction and Multiplex PCR amplification of *E. canis* VirB9, Babesia spp 16S rRNA and *H. canis* 16S rRNA were described by Kledmanee et al. (2009). DNA was extracted by proteinase K digestion and phenol/chloroform/ isoamyl (25:24:1). We used the primers *Ehr*1401F (5’CCA TAA GCA TAG CTG ATA ACC CTG TTA CAA 3’), *Ehr*1780R (5’TGG ATA ATA AAA CCG TAC TAT GTA TGC TAG3’), *Ba*103F (5’CCA ATC CTG ACA CAG GGA GGT AGT GAC A3’), *Ba*721R (5’CCC CAG AAC CCA AAG ACT TTG ATT TCT CTC AAG3’), *Hep*001F (5’CCT GGC TAT ACA TGA GCA AAA TCT CAA CTT3’), and *Hep*737R (5’CCA ACT GTC CCT ATC AAT CAT TAA AGC3’) for detecting *E. canis*, *Babesia* spp and *H. canis*, respectively.

Multiplex PCR was carried out in a solution containing 5 μl of extracted DNA and 45 μl of 0.4 pmol of each primer, 300 μM of each dNTP (QIAGEN®, Germany), 4 units of HotStarTaq DNA Polymerase (QIAGEN®, Germany), 1xPCR buffer (20 mM Tris-HCl pH 8.4, 50 mM KCl), 1.5 mM MgC12 and ultrapure sterile water. Amplification was performed in a thermocycler (PTC-200, MJ Research, Water Town, MA) and thermocycling consisting of one step of 15 minutes at 95°C followed by 30 cycles of 45 seconds at 94°C, 45 seconds at 65°C, and 90 seconds at 72°C with a final extension step of 10 minutes at 72°C. The amplicons were separated by electrophoresis in 2.5% agarose gel in 40 mM Tris-acetic acetate pH 8.4, 1 mM EDTA, stained with ethidium bromide (0.5μg/ml) and visualized under UV light.

Results and Discussion
Heavy flea infestation was visually observed on the dog, particularly on the pinna, around the eyes, abdominal and inguinal regions as well as toe pads (Figs. 1, 2). The dog was weak and emaciate. The visible mucous membranes were pale and the animal was mild dehydrated. Hematological profile revealed microcytic normochromic anemia (Table 1). White blood cell (WBC) differential counts showed eosinophilia (Table 2). No hemoparasites were detected in both blood smears and molecular method (Multiplex PCR).
Fleas were the only ectoparasites recovered in this investigation. No attempts were performed to quantify the number of them. Fleas collected from this dog were classified as *E. gallinacea* (Fig. 3). They have no genal and pronotal ctenidia. The head is sharply angled at front. On the head behind the antenna, there are two setae and a well developed occipital lobe usually presents in female. The maxillary laciniae are broad and coarsely serrated (Wall and Shearer, 1997).

The microcytic type of anemia detected in the examined dog would indicate a chronic blood loss with suppressed erythropoiesis (Yulahum et al., 1989). For WBC differential count, it indicated eosinophilia, which probably related to the flea infestation (Stockham and Scott, 2002). In addition, the dogs infested with intestinal parasites such as *Ancylostoma caninum*, *Trichuris vulpis* and *Toxocara canis*, they also showed eosinophilia (Lee-Parritz, 2001). However, intestinal parasites were not examined in this study.

Various studies have been reported about flea infestation dogs in Thailand. *Ctenocephalides felis orientis* and *Ctenocephalides canis* were found to be the common fleas in dogs (Sangvaranond, 1990, Nithikathkul et al., 2005; Nuchjangreed and Somprasong, 2007). A part from those common species, *Echidnophaga gallinacea* is one of the fleas that have been reported to infest dogs and other domestic animals (Wall and Shearer, 1997). The presence of them was in accordance with a report by Changbunjong et al. (2009). They showed the *E. gallinacea* could infest dogs, cats and chickens in Tak province with an infestation rate of 1.1%, 33.3% and 20% respectively.

The sticktight flea infestation seems to cause adverse health problems in dogs, cats, chickens and other animals. The occurrence of this particular flea is non-specific host. Infestations on animals may be persistent if they continually exposed to the sources of infestation. The finding of this investigation is valuable for control of the fleas in order to prevent adverse effects on animal health.

**Figure 1.** Sticktight fleas (*Echidnophaga gallinacea*) on the pinna of the dog.

**Figure 2.** Sticktight fleas (*Echidnophaga gallinacea*) on the inguinal region of the dog.

**Figure 3.** Morphology of an adult female sticktight flea (*Echidnophaga gallinacea*).
Table 1  Hemogram- erythrocytic series.

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<tr>
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<th>Normal range</th>
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<tr>
<td>Red blood cell (x 10^6 μl^-1)</td>
<td>4.94-5.9</td>
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<tr>
<td>Hemoglobin (g/dl)</td>
<td>9.3-10.18</td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>28.70-35.55</td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>58-60.77</td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>18.9-20.25</td>
</tr>
<tr>
<td>MCHC (g/dl)</td>
<td>32.5-32.36</td>
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Table 2  Hemogram- total leukocyte and differential leukocyte count.

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<tr>
<td>White blood cell (μl^-1)</td>
<td>12,300-6,000-17,000</td>
</tr>
<tr>
<td>Monocyte (%)</td>
<td>6-3-10</td>
</tr>
<tr>
<td>Neutrophil (%)</td>
<td>62-60-77</td>
</tr>
<tr>
<td>Lymphocyte (%)</td>
<td>18-12-30</td>
</tr>
<tr>
<td>Eosinophil (%)</td>
<td>14-2-10</td>
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<td>Basophil (%)</td>
<td>0-Rare</td>
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References


