DISTRIBUTION OF THE LARVAE OF BLACKFLIES (DIPTERA: SIMULIIDAE) AT DOI INTHANON NATIONAL PARK, NORTHERN THAILAND

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Abstract. The larvae of seventeen Simulium species consisting of thirteen known species, three unnamed species and one new species were distributed in various localities with different habitats at 400 - 2,460 meters above sea level in Doi Inthanon National Park, Chiang Mai Province, northern Thailand. Simulium caudisclerum, S. sp G and S. sp H were restricted to the high altitude of 2,460 m at the cool Ang Kha habitat of the summit of Doi Inthanon. On the other hand, S. nakhonense and S. rudnicki occurred only at low altitudes from 400 m - 700 m. Some Simulium species such as S. fenestratum, S. asakoae, S. inthanonense and S. sp J were found in the warmer localities at 1,010 m - 1,280 m altitudes. Moreover, S. feuerborni and S. rufibasis were widely distributed species at different altitudes from 700 m - 2,460 m and 1,010 m - 2,300 m respectively. Therefore, the distribution and abundance of Simulium species seem to correlate with altitudes as well as micro-habitat factors such as water temperature and water velocity.

INTRODUCTION

Blackflies are one of medically important insects belonging to the family Simuliidae of the order Diptera. They are important as pests of humans and animals and also as vectors of filalial nematode of the genus Onchocerca to humans and cattle. The most serious human disease associated with simuliids is onchocerciasis or river blindness, caused by the filalial worm Onchocerca volvulus Leuckart (Peterson, 1984). This disease is common in Tropical Africa. Central America and northern South America. Simuliids are also vector of other Onchocerca species to livestock such as O. gutterosa in cattle (Kettle, 1990). In addition to filalial nematodes, some blackflies can transmit various avian blood protozoans of the genus Trypanosoma and several species of Leucocytozoon among birds including domestic poultry (Peterson, 1984; Kettle, 1990). Moreover, blackflies can also cause a "blackfly fever" which is a syndrome of headache, fever, nausea, dermatitis and allergic asthma.

The family if Simuliidae consists of 24 genera, of these only *Simulium* is the largest and most important genus (Crosskey, 1993). Recently, Takaoka and Davies (1995) reported a total of 37 species of simuliids which were arranged in 3 subgenera of

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the genus Simulium and one species assigned to the genus Sulcicnephia Rubtsov in West Malaysia. Additionally, the geographical distribution of the genus Simulium in the Oriantal and Australasian regions were also reported and mapped (Takaoka, 1996).

In Thailand, knowledge of distribution and abudance of blackflies is very scarce, although the 26 Simulium species were recorded on the basis of external morphological characters (Takaoka and Suzuki, 1984; Takaoka and Saito, 1996; Takaoka and Adler, 1997). It is known that blackflies are a well-known pest to humans and domestic animals at Doi Inthanon National Park, Chiang Mai Province, northern Thailand. They are found in large numbers and cause irritation to the local people, domestic animals and livestock, as well as tourists, Apart from the annoyance, they cause reduced milk production in domestic animals.

In this report, we present the distribution and abundance of the larvae of blackflies from various localities at different altitudes of Doi Inthanon National Park, northern Thailand.

MATERIALS AND METHODS

Study sites

Natural populations of blackfly larvae were collected approximately every three months between April 1996 and February 1998 from twelve locations at different altitudes of Doi Inthanon National

Park, Chiang Mai Province, northern Thailand. These locations are (1) Mae Ya waterfall (520 m), (2) Mae Klang waterfall (400 m), (3) Vang Kwai waterfall (410 m), (4) Vachiratharn waterfall (700 m), (5) Huai Sai Luaeng waterfall (1,010 m), (6) Oab Noi waterfall (1,010 m), (7) Park Headquarters (1,180 m), (8) Royal Project (1,250 m), (9) Siriphum waterfall (1,270 m), (10) Khun Huai Haeng (1,280 m), (11) Kew Mae Pan (2,300 m) and (12) Ang Kha (2,460 m) (Fig 1).

Sample collection

Larvae were removed from stone and vegetable substrates with fine forceps and placed on absorbent tissue to blot off stream water. The larvae were preserved in 80% ethanol for morphological studies.

Taxonomy of larvae

In the laboratory, field samples were sorted carefully. The anatomical parts of sorted larvae were dissected using the method described by Davies (1968) and were then identified following keys of Takaoka (1977, 1979), Takaoka and Suzuki (1984), Takaoka and Davies (1995) and Takaoka and Saito (1996). Some known and unknown larvae were kindly confirmed and identified by Professor Hiroyuki Takaoka, Department of Infectious Disease Control, Faculty of Medicine, Oita Medical University, Japan.

RESULTS

Species distribution and abundance

Doi Inthanon National Park is ruggedly mountainous. The highest peak of Doi Inthanon is at 2,565 m above sea level. The climate is cool all year particularly at higher elevations. The seventeen Simulium species including thirteen known species and four unnamed species were collected from twelve locations which are approximately 400 - 2,460 m altitudes. The four unnamed species have been assigned single letter as species G, H, J and L because of lack of adult and pupal specimens. S. sp G is a new species in subgenus Montisimulium (Takaoka, personal communication). S. sp H, S. sp. J and S. sp. L may represent morphological variants of known species. Further specimens of pupa and adult of these species are needed for final identification. Species distribution, in descending order of dominance, at twelve collection sites, is shown in Fig 1. The detail of species distribution is given in Table 1, showing collection locations, collection dates, season, number of larvae and percentage of species examined morphologically in each sample.

Simulium caudisclerum was the most dominant species at Ang Kha (Table 1). It occurs at low frequencies of 45% and 46% in August 1996 and August 1997 (the mid rainy season) respectively. The frequency of this species increased from 73%

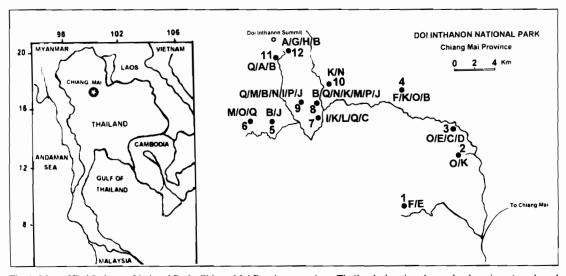


Fig 1-Map of Doi Inthanon National Park, Chiang Mai Province, northern Thailand, showing the twelve locations (numbered 1-12) where the seventeen Simulium species (A-Q) were collected for this study. Location numbers are the same as those listed in Table 1. A = S. caudisclerum; B = S. feuerborni; C = S. nigrogilvum; D = S. siripoomense; E = S. quinquestriatum; F = S. rudnicki; G = S. sp G; H = S. sp H; I = S. tani; J = S. sp J; K = asakoae; L = S. sp L; M = S. fenestratum; N = S. inthanonense; O = S. nakhonense; P = S. chamlongi; Q = S. rufibasis

Percentage of distribution of Simulium species at 12 localities of Doi Inthanon National Park. Chiang Mai Province.

Location (altitude) 1. Mae Ya waterfall (520 m)	Collection date Seaso date Seaso 11 21 Aug 96 wet 30 Oct 96 15 Feb 97 cool 18 May 97 hot 21 Aug 97 wet 21 Aug 97 wet	Season Season Season Cool Cool Hot	S. siripoomense S. nigrogilvum S. feuerborni S. caudisclerum 100 100 100 100 100 100 100 1	S. caudisclerum	S. feuerborni	S. nigrogilvum	1		S. sp G	S. sp J S. tani S. sp H S. sp G S. rudnicki S. quinquestriatum	S. tani	S. sp J	S. asakoae	S. sp L	S. fenestratum	S. inthanonense		S. nakhonense	S. chamlongi S. nakhonense
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5. Huai Sai Luaeng waterfall (1,010 m)	31 Oct 96	wet	334	,	95.8	ı	,		•	1	•	2.1	1	•	1		•		•
6. Oab Noi waterfall (1,010 m)	31 Oct 96	wet	64		1	1	1	,	ı	•	•	1	1	1	76.6	9	1	- 20.3	1
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BLACKFILES LARVAE DISTRIBUTION IN NORTHERN THAILAND

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in the late rainy season (October 1996) to 85% in the cool season (February 1997). A similar situation was also found in October 1997 and February 1998 in which S. caudisclerum occurred at high frequencies of 80% and 88% respectively. S caudisclerum also occurred at Kew Mae Pan with low frequencies ranging from 2 - 10% during the rainy and cool seasons. Most of the larvae from Ang Kha were found attached to fallen leaves in slow flow of small streams 0.5 - 1.0 m wide. In contrast, the larvae from Kew Mae Pan predominated on trailing vegetation in fast flow of medium streams 1.5 - 2.0 m wide.

- S. feuerborni was found to be a widely distributed species at different altitudes from 700 -2,400 m (Fig 1). However, it was the most abundant at the Royal Project (1,250 m) and Huai Sai Luaeng waterfall (1,010 m) (Table 1). In 1996, it was found at high frequencies ranging from 66 -84% at the Royal Project in the cool (November), hot (April) and rainy seasons (August) (Table 1). In 1997 - 1998, the frequency of S. feuerborni decreased from 64% in the hot season (May 1997) to 26% in the mid rainy season (August 1997) to 15% at the end of rainy season (October 1997) and 3% at the end of cool season (February 1998). Only one sample of S. feuerborni was collected from Huai Sai Luaeng waterfall at a high frequency of 96% in the rainy season (October 1996). Larvae of S. feuerborni predominated in shallow and man-made streams 0.2 - 0.5 m wide at the Royal Project and Huai Sai Luaeng waterfall. When the water in the streams dried up, the number of larvae decreased drastically at the end of cool season (February 1998) at the Royal Project and they had been absent since February 1997 at Huai Sai Luaeng waterfall . Pupae and larvae were usually found attached to trailing grasses and fallen leaves. S. feuerborni was also present at Vachiratharn waterfall (700 m), Siriphum waterfall (1,270 m), Kew Mae Pan (2,300 m) and Ang Kha (2,460 m) with different frequencies of 15%, 4%, 0.2% and 0.3% respectively.
- S. nigrogilvum was found at low frequencies ranging from 0.3 -7% at Vang Kwai waterfall during the cool and rainy seasons. It also occurred at a low frequency of 5% at park headquarters in the rainy season (August 1996).
- S. siripoomense was found at a low frequency of 1% at Vang Kwai waterfall in the cool season (February 1997).
- S. quinquestriatum was found at Vang Kwai (20%) and Mae Ya (8%) waterfalls in the cool sea-

son (February 1998).

- S. rudnicki was the most distinctive and abundant species at Mae Ya waterfall year-round. The larvae of this species were collected at a high frequency of 100% from August 1996 to October 1997. It was also found at Vachiratharn waterfall in August 1996 (38%) and February 1998 (92%). Pupae and larvae were firmly attached to rock faces in very fast flowing streams 1.5 -4.0 m wide, exposed to the sun.
- S. tani occurred at park headquarters at low frequencies ranging from 22% in the hot season (April 1996) to 37% in the rainy season (August 1996) while a range of 2 11% was found in the Siriphum waterfall population.
- S. asakoae was a widely distributed species with low abundance levels at different altitudes from 400 1,280 m. A range of 4 34% was found at Mae Klang waterfall (400 m), Vachiratharn waterfall (700 m), park headquarters (1,180 m), the Royal Project (1,250 m) and Khun Huai Haeng (1,280 m).
- S. fenestratum was present at Siriphum waterfall year-round. In 1997 it had high frequencies ranging from 64 70% in the mid rainy season (August 1997) to the end of the rainy season (October 1997), but it was less abundant (27%) in August 1996. It occurred at low frequencies which ranged from 3 13% in early cool season to late cool season (November 1996-February 1997). At the Royal Project, this species was found at low frequencies ranging from 6% in the hot season (May 1997) to 17% in the rainy season (October 1997). However, it was a dominant species at Oab Noi waterfall (77%).
- S. inthanonense occurred at the Royal Project the year round. The highest frequency, 32%, was found in the mid rainy season (August 1997) while a range of 4 15% was found at the end of rainy season (October 1997), cool season (November 1996 and February 1998) and hot season (April 1996 and May 1997). This species was dominant at Khun Huai Haeng in the hot season (50%) and rainy season (66%). However, it was present with low frequencies ranging from 0.6 7.7% in some samples from Siriphum waterfall.
- S. nakhonense was the most abundant species at Vang Kwai waterfall. It was found at high frequencies ranging from 73 98% in all seasons during 1997 and 1998 (Table 1). In addition, it was also found at a high frequency of 93% in the Mae Klang waterfall population. On the other hand, a range of 2 20% was found in the populations from

Vachiratharn and Oab Noi waterfalls. Most of the pupae and larvae were collected from trailing grasses and roots in a small stream (0.3 - 0.5 m) or at the margins of very fast flowing streams 4.0 - 6.0 m wide, rarely on submerged rocks and fallen leaves.

- S. chamlongi was a less abundant species in the populations from the Royal Project and Siriphum waterfall. It was found at low frequencies in the cool seasons (November 1996 and February 1998) at these locations.
- S. rufibasis seemed to be a widely distributed species at different altitudes from 1,010 2,300 m (Fig 1). It was the most dominant species at Kew Mae Pan and Siriphum waterfall the year round. It occurred at high frequencies which ranged from 69 84% in the rainy (August 1997) and cool seasons (February 1998) at Kew Mae Pan. Similarly, it was abundant at Siriphum waterfall during the rainy and early cool months in 1996. It was less abundant at the Royal Project, although the highest frequency of 76% was found in the cool season (February 1998). Pupae and larvae were found on trailing grasses and trailing vegetation in the fast flow of small to medium streams 0.3 2.0 m wide.
- S. sp G occurred only at Ang Kha. It was found at high frequency of 52% in the mid rainy seasons (August 1996 and August 1997). Although the sample in May 1997 showed a high frequency (100%) of S. sp G, this was probable due to a sampling error.
- S. sp H was present only at Ang Kha with low frequencies in all seasons from 1996-1998.
- S. sp J was a rare species which occurred with low frequencies in some locations such as the Royal Project and Siriphum waterfall.
- S. sp L was found only at park headquarters with low frequencies ranging from 20% in the hot season (April 1996) to 23% in the rainy season (August 1996).

Seasonality at Vang Kwai waterfall, Royal Project, Siriphum waterfall and Ang Kha

The timing of weather system at Doi Inthanon National Park is different from that in other places on the mainland. These seasons: hot/dry = March/early May, Wet = late May/October and cool = November/February are recognized in the park. The seasonal variation in frequencies of *Simulium* species collected from some locations during 1996 or 1997 to early 1998 was investigated in the present study.

Vang Kwai waterfall (410 m)

Four Simulium species found in this locality, in descending order of dominance, were S. nakhonense, S. quinquestriatum, S. nigrogilvum and S. siripoomense (Fig 1). There were some unknown species found at this locality.

S. nakhonense was the most abundant species in all seasons from 1997-1998 (Figs 2 and 3). Pupae and larvae of S. quinquestriatum were not present in 1997 but they appeared in one sample collected in February 1998. S. nigrogilvum was found in two samples collected in February and October 1997 but it disappeared in the samples collected in May and August 1997 and February 1998. S. siripoomense seemed to be the least common species at this locality since it occurred only in one sample (February 1997). Pupae and larvae of these species were attached on trailing vegetation at the margin of very fast flowing streams 4 - 6 m wide, rarely on rocks and fallen leaves. Water temperature ranged from 17.0-25.5°C. The pH value was 8.3-8.7.

Royal Project (1,250 m)

In descending order of dominance of the seven Simulium species at the Royal Project were S. feuerborni, S. rufibasis, S. inthanonense, S. asakoae, S. fenestratum, S. chamlongi and S. sp J (Fig 1)

Larvae and pupae of *S. feuerborni* were predominant in all seasons in 1996, but they decreased drastically from the hot season (May 1997) to the late cool season (February 1998) (Figs 4, 5). Larvae of *S. rufibasis* were present at this locality in close association with larvae and pupae of *S. feuerborni* and there was a peak in February 1998. *S. inthanonense* seemed to be associated with *S. feuerborni* and *S. rufibasis* in all seasons except for the rainy season (August 1996). Larvae and pupae of these species were collected from trailing glasses and small stones in slow flow of shallow and man-made stream 0.2 - 0.5 m wide in an open glassy area. Water temperature and pH were 19 - 22°C and 6.9 - 8.0.

Siriphum waterfall (1,270 m)

The number of species and the time of their seasonal appearance are shown in Figs 6, 7. The seven *Simulium* species, in descending order of dominance, were *S. rufibasis*, *S. fenestratum*, *S. feuerborni*, *S. inthanonense*, *S. tani*, *S. chamlongi* and *S.* sp J (Fig 1).

S. rufibasis was the most common species in all seasons from 1996 to early 1998 except for the rainy season 1997 in which S. fenestratum was

■ Unknown

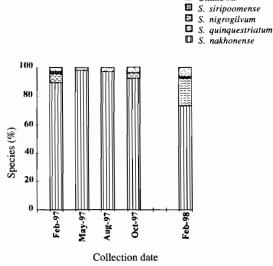


Fig 2-Seasonal succession of Simulium larvae at Vang Kwai waterfall (February 1997-February 1998).

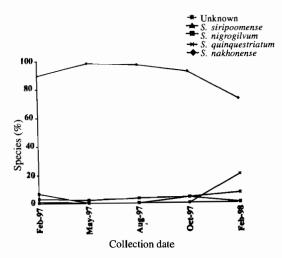
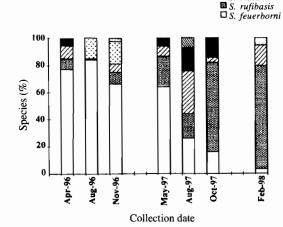


Fig 3-Temporal changes in percentage of distribution of Simulium larvae at Vang Kwai waterfall (February 1977-February 1998).

dominant (Figs 6, 7). S. fenestratum was the second most dominant species. It was found in close association with S. rufibasis in all seasons with a highest peak in August 1997. Larvae and pupae of the other five species, S. feuerborni, S. inthanonense, S. tani, S. chamlongi and S. sp J appeared or disappeared in different seasons from 1996 to early 1998. S. feuerborni occurred at low frequencies in four samples collected in August 1996, February 1997, May 1997 and February 1998. S. inthanonense was found only in the hot season (May 1997) and the late cool season (February 1998). S. tani occurred



S. sp J

■ S.

os.

S. chamlongi

asakoae

S. inthanonense

fenestratum

Fig 4-Seasonal succession of Simulium larvae at Royal Project (April 1996-February 1998).

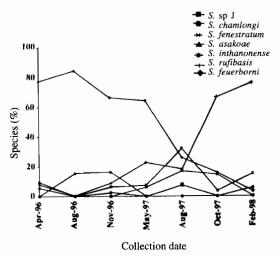


Fig 5-Temporal changes in percentage of distribution of Simulium larvae at Royal Project (April 1996-February 1998).

in all seasons in 1997, but it was not found in 1996 and early 1998. A similar situation was also found with S. sp J which occurred with low frequencies similar to S. tani. Larvae and pupae of these species were found on trailing grasses in a small man-made stream 0.2 - 0.5 m wide. Water temperature and pH were 16.5 - 20.0°C and 7.7 - 7.8 respectively.

Ang Kha (2,460 m)

Ang Kha is located at the summit of Doi Inthanon National Park at the altitude of 2,460 m. The climate is cool all year round because of the

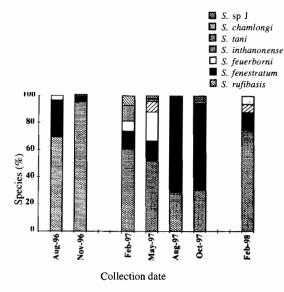


Fig 6-Seasonal succession of Simulium larvae at Siriphum waterfall (August 1996-February 1998).

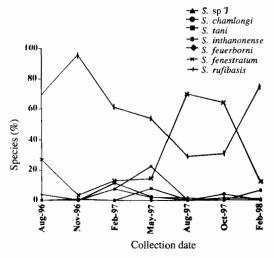
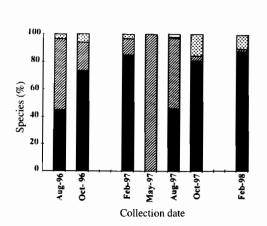


Fig 7-Temporal changes in percentage of distribution of Simulium larvae at Siriphum waterfall (August 1996-February 1998).

high altitude. January is the coldest month with an average temperature of 5.5°C. In the hot season the temperature remains cool and pleasant.

Four Simulium species found in this locality, in descending order of dominance, were S. caudisclerum, S. sp G, S. sp H and S. feuerborni (Fig 1). S. caudisclerum was the most common species (Figs 8, 9). It occurred in all samples collected from August 1996 to February 1998, except for one sample (May 1997) in which only S. sp G was found. S. sp G was the second most dominant species with close



□S. feuerborni

S. caudisclerum

☑ S. sp H

ZS. sp G

Fig 8-Seasonal succession of Simulium larvae at Ang Kha (August 1996-February 1998).

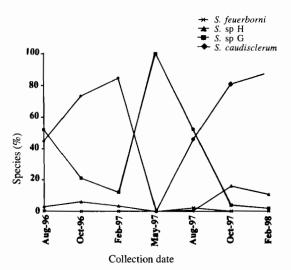


Fig 9-Temporal changes in percentage of distribution of Simulium larvae at Ang Kha (August 1996-February 1998).

association with *S. caudisclerum*. The highest frequency of 100% of *S.* sp G was found on the February 1997 sample. This may be due to sampling error, since all seventeen larvae collected were *S.* sp G. Larvae of *S.* sp H were also found at low frequencies in most samples. Only a few *S. feuerborni* larvae were found in two samples collected in August 1996 and 1997. Larvae of these species were found on fallen leaves and decaying leaves in slow flowing shaded streams 0.5-1.0 m wide. Water temperature and pH were 5.0-15°C and 5.7-6.7 respectively.

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DISCUSSION

The distribution of Simulium species at Doi Inthanon National Park seems to correlate with altitudes because different species were found at difference altitudes with various habitats. S. caudisclerum, S. sp G and S. sp H were restricted to the high altitudes (2,460 m) while S. nakhonense and S. rudnicki were found only at low altitudes (400 - 700 m). However, S. feuerborni was widely distributed at different altitudes (700 - 2,460 m). Some micro-habitat factors such as food (Ulfstrand, 1967); food quality (Carlsson et al, 1977), heterogeneity of flow (Eymann, 1991), substrate types, water velocity and water depth (Lewis and Bennett, 1975) are associated with the distribution of blackfly larvae. The most significant factors affecting the distribution of simuliid larvae were current velocity, substrate types and water depth (Lewis and Bennett, 1975). In addition, nutrient salts may also exert an indirect influence by affecting the amount of microplankton present for feeding by simuliids (Merritt et al, 1978; Chutter, 1968; Ali et al, 1974). In this study, the distribution of larval populations may be influenced by some micro-habitat factors such as substrate types and water velocity, since larvae and pupae of different species from various locations were found on different substrates in stream of different sizes and water velocities. S. caudisclerum, S. sp G and S. sp H were found in slow flowing shaded streams 0.5 - 1.0 m wide, surrounded by upland woods, and with substrates of fallen leaves. In contrast, S. feuerborni prefers to breed in small, shallow and man-made streams in open grassland like at the Royal Project and Huai Sai Luaeng waterfall. These streams were about 0.3 m wide with substrates of trailing grasses and fallen leaves.

A large number of larvae and pupae of S. rudnicki were attached on rocks at Mae Ya waterfall the year-round. Many thousands of S. rudnicki larvae congregated on rocks, forming flattened dark aggregations under the thin layers of water flowing at high velocities. Water velocity affected the micro-distribution of larvae and pupae on rocks of some simuliids (Phillipson, 1957; Maitland and Penney, 1967) although it had no significant effect on the population dynamics of S. argus larvae and pupae; however, it was a significant ecological factor that affected the larval-pupal density of S. canadense (Mohsen and Mulla, 1982). The distribution of S. rudnicki larvae and pupae on rocks strictly followed the direction and intensity of the current as reported concerning S. virgatum (Mohsen and Mulla, 1982). It is believed that water velocity is one of the most important ecological factors influencing the distribution and abundance of blackfly larvae and pupae because water currents bring larval food, supply oxygen to larvae and pupae and help larvae migrate to more suitable positions in streams (Phillipson, 1957; Carlsson, 1962; Maitland and Penney, 1967; Wallace and Merritt, 1980). During 1996 to early 1998, a large number of Simulium larvae and pupae were collected from the fast flowing streams at Siriphum and Vang Kwai waterfalls where they occurred year-round. On the other hand, a smaller number of larvae and pupae were collected from the slow flowing streams at Ang Kha.

Water velocity was not the only ecological variable which influenced the abundance of blackfly larvae and pupae, water conditions are also factors. The water temperature at Ang Kha was generally cool with a range of 5.0-15°C but it is lower than 10°C in the cool season in which the number of larvae decreased drastically. Compared with Ang Kha, the water temperature at Vang Kwai waterfall. the Royal Project and Siriphum waterfall were warm with a range of 17-25.5°C, 19-22°C and 16.5-20°C respectively. These warm habitats were dominated by many multivoltine species which occurred at high frequencies the year round. Therefore, water temperature seems to affect the number and density of the Simulium larvae and pupae at different altitudes of Doi Inthanon National Park. It was found that water temperature strongly influenced the growth and survival rate of S. venustum Say larvae in the laboratory (Mokry, 1976). Additionally, Ross and Merritt (1978) showed that water temperature, by determining hatching time and development rates, not only influenced the geographical and local distribution of five species of simuliids, but also the annual dynamics of species.

In February 1997, all Simulium species larvae and pupae at the Royal Project disappeared because the water in the small, shallow man-made stream dried up due to human activity. Therefore, ecological factors not only influenced the distribution and abundance of species, but some biotic factors, such as humans and predators, also had an effect.

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