

Species Diversity of Adult Caddisflies (Order Trichoptera)
in Northern Lao People's Democratic Republic
ความหลากหลายชนิดของตัวเต็มวัยแมลงหนอนปลอกน้ำ (อันดับ Trichoptera)
ในภาคเหนือของสาธารณรัฐประชาธิปไตยประชาชนลาว

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ABSTRACT

Diversity of Trichoptera adults was investigated from 2 rivers, Nam Song and Nam Khan, Lao PDR. in November 2017 by using an ultraviolet light trap. A total of 1,952 male individuals in 6 families, 15 genera and 30 species of Trichoptera were found. The most abundant species was *Psychomyia intorachit* (0.49%) and followed by *Cheumatopsyche charites* and *Pseudoleptonema quinquefasciatum* (each 0.12%). Species richness at Nam Song (23 species) was greater than that of Nam Khan (10 species) but Shannon-Wiener species diversity index in Nam Khan (1.95) were higher than that in Nam Song (1.82). Sørensen's similarity coefficient of Trichoptera assemblages between the two rivers was 0.22. *Cheumatopsyche charites*, *Psychomyia arthit* and *P. intorachit* were found in both rivers. Fifteen species of Trichoptera were new reported, which increased species richness recorded of Lao PDR. up to 254 species.

บทคัดย่อ

การศึกษาคความหลากหลายชนิดของตัวเต็มวัยแมลงหนอนปลอกน้ำในน้ำของและน้ำขุ่น ประเทศสาธารณรัฐประชาธิปไตยประชาชนลาว ดำเนินการในเดือนพฤศจิกายน พ.ศ. 2560 โดยเก็บตัวอย่างตัวเต็มวัยด้วยกับดักแสงไฟอัลตราไวโอเล็ต พบตัวเต็มวัยเพศผู้จำนวนรวม 1,952 ตัว ใน 6 วงศ์ 15 สกุล 30 ชนิด ชนิดที่มีความชุกชุมมากที่สุดคือ *Psychomyia intorachit* (0.49%) รองลงมา คือ *Cheumatopsyche charites* และ *Pseudoleptonema quinquefasciatum* (ชนิดละ 0.12%) น้ำของพบแมลงหนอนปลอกน้ำจำนวน 23 ชนิดมากกว่าที่น้ำขุ่นซึ่งพบจำนวน 10 ชนิด แต่ดัชนีความหลากหลาย (Shannon-Wiener Diversity Index) ที่น้ำขุ่น (1.95) มีค่าสูงกว่าน้ำของ (1.82) สัมประสิทธิ์ความคล้ายคลึงกัน (Sørensen's similarity coefficient) ของชุมชนแมลงหนอนปลอกน้ำตัวเต็มวัยในแม่น้ำทั้งสองแห่งนี้มีค่าเท่ากับ 0.22 แมลงหนอนปลอกน้ำ *Cheumatopsyche charites*, *Psychomyia arthit* และ *P. intorachit* เป็นชนิดที่พบในแม่น้ำทั้งสองแห่ง ผลการศึกษานี้พบแมลงหนอนปลอกน้ำที่เป็นการพบครั้งแรกในประเทศจำนวน 15 ชนิด ทำให้จำนวนชนิดแมลงหนอนปลอกน้ำที่มีรายงานการพบในประเทศสาธารณรัฐประชาธิปไตยประชาชนลาวเพิ่มขึ้นเป็น 254 ชนิด

Keywords: New records, Trichoptera adult, Lao PDR.

คำสำคัญ: รายงานครั้งแรก ตัวเต็มวัยแมลงหนอนปลอกน้ำ สาธารณรัฐประชาธิปไตยประชาชนลาว

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Introduction

Trichoptera (T) or caddisflies is one of the largest groups of aquatic insects, which are holometabolous. The Trichoptera are high abundance and they inhabit a variety of freshwaters such as springs, streams, rivers, ponds, wetlands and lake banks (Wiggins, Mackay, 1978). Trichoptera larvae are an important component of freshwater benthic community because they contributed to transfer energy and nutrients through all trophic levels, which had a substantial role in food web (Resh, Rosenberg, 1984; Wiggins, 1996). Various species of Trichoptera larvae had the very different susceptibilities to pollutants and other types of environmental disturbances (Rosenberg, Resh, 1993). Therefore, they are popular to use in the biological assessment and monitoring of water quality, which combined with the mayfly larvae (Ephemeroptera, E) and the stonefly larvae (Plecoptera, P) especially used in temperate zone because knowledge in taxonomy and ecology of EPT have been well studied (Chamorro et al., 2007). In Lao People's Democratic Republic (Lao PDR.), studies of aquatic insects were scarcely reported. The present study aims to investigate species diversity of Trichoptera adults at Nam Song and Nam Khan, where is located in northern Lao PDR.

Methodology

Collecting samples and identification

The study sites are located at the Nam Khan, Xieng Ngeun District, Luang Prabang Province and Nam Song, Vang Vieng District, Vientiane Province in Lao PDR. (Figure 1 and Figure 2). Details of the study sites were shown in Table 1. The field sampling was carried out in November 2017. Three replicates of surface water samples were collected from each site in order to measure some physical and chemical parameters of water quality. Air temperature ($^{\circ}\text{C}$) was measured with thermometer, water channel width (m) and water depth (cm) were measured with tape measure, water velocity (m/s) was measured with flow velocity indicator model D625F, turbidity (NTU) was measured with turbidimeter Hach model 2100N. pH, electrical conductivity (EC, $\mu\text{S}/\text{cm}$) and total dissolved solid (TDS, mg/l) were measured by using pH/EC/TDS meter Hanna model HI 98129. Nitrate nitrogen (NO_3^- -N, mg/l) and orthophosphate (PO_4^{4-} , mg/l) were analyzed using titration method. Suspended solid (SS, mg/l) was measured with photometric method. Chlorophyll a (mg/l) content was measured using extracted methanol method. Biochemical oxygen demand (BOD, mg/l) and dissolved oxygen (DO, mg/l) were measured by using dissolved oxygen meter YSI model 550A. Adult of caddisflies were collected using an ultraviolet light trap (8-W fluorescent powered by a 18-volt DC battery) placed near the stream margin to operate simultaneously from 5.00 pm to 7.00 am. The specimens were preserved in 95% ethanol for later identification. The characters of the spur formula, the presence or absence of ocelli and numbers of segments of male maxillary palps based on that of Holzonthal (2007) and Malicky (2010) were used for identification to the species level. An apex of abdomen of each male adult was dissected under the stereo-microscope. Genitalia preparation was made by clearing the apex of abdomen in warm 10% potassium hydroxide (KOH)

solution for 30 minute-1 hour after rinsing the KOH with water and 95% ethyl alcohol, the apex of abdomen was transferred to glycerin for further examination, which was modified from Malicky (2010).

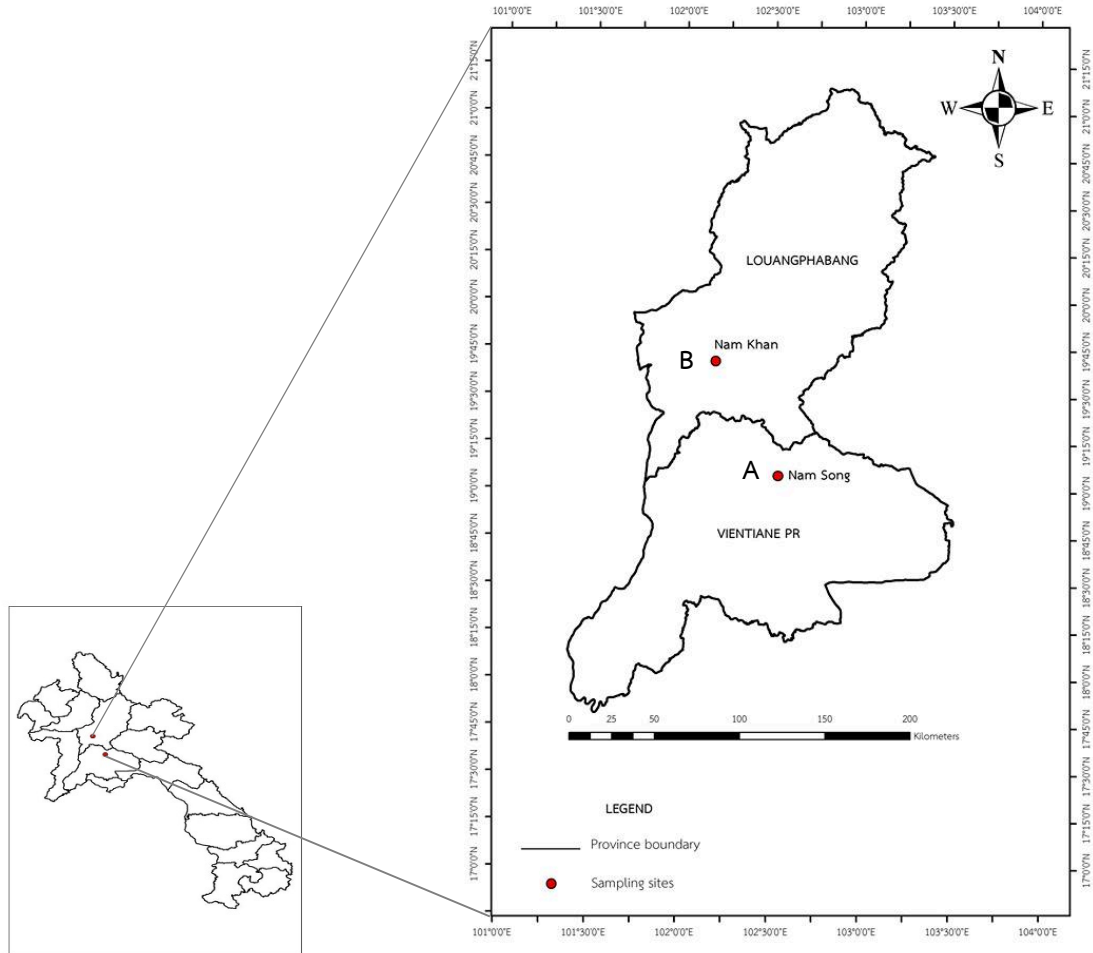


Figure 1 Showing study site in Lao PDR. A: Nam Song, B: Nam Khan

Table 1 List of study sites with geographic data (a.s.l.=above sea level)

Study site	Geographic coordinates	Altitude (a.s.l.) (m)	Substrate	Land use
Nam Song	N 19° 06' 13.07" E 102° 30' 03.16"	315	boulder, cobble, gravel and sand	forest areas
Nam Khan	N 19° 43' 55.58" E 102° 09' 24.31"	332	coarse gravel, sand and substrates were covered by silt	agricultural, rural areas and heavy erosion

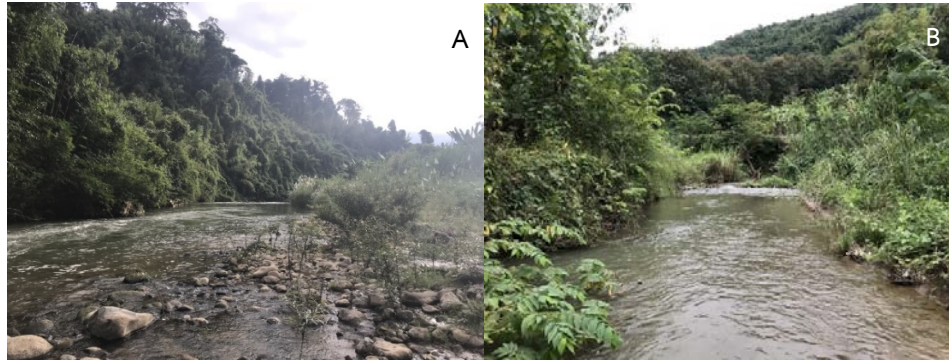


Figure 2 Showing habitats of each study site from Lao PDR. A: Nam Song, B: Nam Khan

Statistical analysis

The difference of physico-chemical parameters of water were analyzed between the two study sites by Independent t-test in SPSS program version 19.0 (SPSS Inc., Chicago, IL, USA). Richness, Shannon-Wiener species diversity index (H'), evenness index (E') and Sørensen's similarity coefficient (S) (Krebs, 1999) were applied to analyze the species diversity of Trichoptera adults as following:

$$H' = - \sum_{i=1}^s (p_i)(\ln p_i)$$

$$p_i = n_i/N$$

$$E' = H'/\ln s$$

$$S = 2C/(A+B)$$

Where	H'	=	Shannon-Wiener species diversity index
	s	=	Number of species
	p_i	=	n_i/N (where n_i =number of individuals in each species, N =Total number of all species)
	E'	=	Evenness index
	S	=	Sørensen's similarity coefficient
	C	=	Number of species in community A and B
	A	=	Number of species in community A but not in community B
	B	=	Number of species in community B but not in community A

Results

Physico-chemical parameters

Mean \pm SD of physico-chemical parameters of water quality from both study sites are shown in Table 2. Independent t-test revealed that average values of water channel width, dissolved oxygen, electrical conductivity, total dissolved solid, suspended solids, biochemical oxygen demand and chlorophyll a were significant higher in Nam Song than those of Nam Khan ($p < 0.05$). Depth, water velocity, nitrate nitrogen, orthophosphate and turbidity were non-significant difference ($p > 0.05$) between both rivers.

Diversity of adult Trichoptera

A total of 22,935 Trichoptera adults were collected from the combined results of both study sites (Table 3). At the Nam Song, 21,781 individuals (male=1,931, female=20,850) representing 8 families were collected. The highest to the lowest numbers in each family was followed: Psychomyiidae (male=, 1,225, female=17,428), Hydropsychidae (male=603, female=2,047) and Leptoceridae (male=100, female=835). The families with only males were Odontoceridae (n=2) and Stenopsychidae (n=1). The families with only females were Hydroptilidae (n=9), Philopotamidae (n=528) and Xipocentronidae (n=3). At the Nam Khan, 154 individuals (male=21, female=133) representing 3 families were collected.

In the present study, all male specimens were identified to the species level. Female specimens were not able to identify to species because they lack of necessary characters for the species level identification. Such specimens were not included in any analyses. As shown in Table 4, male adult Trichoptera representing 6 families and 30 species were identified. Hydropsychidae (12 species) was the richest family and followed by Leptoceridae (9 species), Psychomyiidae (6 species), Odontoceridae, Philopotamidae and Stenopsychidae (1 species), respectively. *Psychomyia intorachit* Malicky & Chantaramongkol, 1993 was the most abundant species (0.49%) followed by *Cheumatopsyche charites* Malicky & Chantaramongkol, 1997 (0.12%) and *Pseudoleptonema quinquefasciatum* Martynov, 1935 (0.12%), respectively (Figure 3).



Figure 3 Showing A: *Psychomyia intorachit*, B: *Cheumatopsyche charites*, C: *Pseudoleptonema quinquefasciatum* the abundant species in two study sites of Lao PDR. (Scale bar=1 mm)

Table 2 Mean±SD of physico-chemical parameters of water quality at the Nam Song and Nam Khan and results of Independent samples test (*=significantly difference at $p < 0.05$, ns=non-significantly difference at $p > 0.05$)

Parameter	Nam Song	Nam Khan	p-Value	Significantly difference
Air temperature (°C)	24.00	19.50	-	-
Water temperature (°C)	25.00±0.00	21.10±0.00	-	-
Water channel width (m)	20.00±0.00	4.80±0.62	0.001	*
Water depth (cm)	21.33±8.14	25.67±5.13	0.479	ns
Water velocity (m/s)	0.68±0.06	0.53±0.13	0.137	ns

Table 2 Mean±SD of physico-chemical parameters of water quality at the Nam Song and Nam Khan and results of Independent samples test (*=significantly difference at $p < 0.05$, ns=non-significantly difference at $p > 0.05$) (Cont.)

Parameter	Nam Song	Nam Khan	p-Value	Significantly difference
Dissolved oxygen (mg/l)	5.65±0.19	6.60±0.15	0.002	*
pH	8.14±0.00	8.03±0.00	-	-
Electrical conductivity (µS /cm)	251.00±1.73	413.33±0.58	0.043	*
Total dissolved solids (mg/l)	132.30±0.58	219.00±0.00	0.034	*
Nitrate nitrogen (mg/l)	0.23±0.06	0.33±0.06	0.099	ns
Orthophosphate (mg/l)	0.36±0.17	0.23±0.05	0.259	ns
Suspended solids (mg/l)	9.00±3.46	20.33±4.62	0.043	*
Turbidity (NTU)	1.63±0.28	1.36±0.17	0.234	ns
Biochemical oxygen demand (mg/l)	0.38±0.23	2.52±0.42	0.002	*
Chlorophyll a (mg/l)	0.63±0.21	2.15±0.66	0.046	*

Table 3 Numbers of male and female Trichoptera adults of each family from the Nam Song and Nam Khan

Family	Nam Song River		Nam Khan River	
	male	female	male	female
Hydroptilidae	0	9	0	0
Hydropsychidae	603	2,047	7	125
Leptoceridae	100	835	4	8
Odontoceridae	2	0	0	0
Philopotamidae	100	528	0	0
Psychomyiidae	1,125	17,428	10	0
Stenopsychidae	1	0	0	0
Xipocentronidae	0	3	0	0
Total	1,931	20,850	21	133
Total numbers in each study sites	22,781		154	
Overall	22,935			

Family Hydropsychidae consisted of *Amphipsyche gratiosa* Navas (1922) *Cheumatopsyche charites* Malicky & Chantaramongkol (1997), *C. dubitans* Mosely (1942), *Hydropsyche boreas* Malicky & Chantaramongkol (2000), *Macrostemum floridum* Navas (1929), *Oestropsyche vitrina* Hagen (1859), *Potamorphanus muluensis* Barnaed (1980), *Potamyia chinensis* Ulmer (1915), *P. elektra* Malicky & Chantaramongkol (2001), *P. periboia* Malicky & Chantaramongkol (1997), *P. phaidra* Malicky & Chantaramongkol (1997) and *Pseudoleptonema quinquefasciatum* Martynov (1935). The species found in both study sites was *C. charites*. The species were only found in the Nam Song as follows: *A. gratiosa*, *C. dubitans*, *H. boreas*, *O. vitrina*, *P. muluensis*, *P. chinensis*, *P. elektra*, *P. phaidra* and *P. quinquefasciatum*. The species were only found in the Nam Khan as follows: *M. floridum* and *P. periboia*.

Family Leptoceridae consisted of *Ceraclea harpalyke* Malicky & Chantaramongkol (2002), *C. trismegistos* Malicky (2008), *Setodes argentiguttatus* Gordon & Schmid (1987), *S. brevicaudatus* Yang & Morse (1989), *S. endymion* Malicky & Chantaramongkol (2000), *S. klakahana* Ulmer (1951), *S. meriones* Malicky & Chantaramongkol (2006), *S. sarapis* Malicky & Chantaramongkol (2006) and *S. sp.1*. The species were only found in the Nam Song as follows: *C. Harpalyke*, *C. trismegistos*, *S. brevicaudatus*, *S. klakahana*, *S. kleio*, *S. meriones*, *S. sarapis* and *S. sp.1*. The species were only found in the Nam Khan as follows: *S. argentiguttatus* and *S. endymion*.

Family Odontoceridae consisted of *Marilia aerope* Malicky & Chantaramongkol (1996), which was found in only the Nam Song.

Family Philopotamidae consisted of *Chimarra okuihorum* Mey (1998), which was only found in the Nam Song.

Family Psychomyiidae consisted of *Psychomyia arthit* Malicky & Chantaramongkol (1993), *P. intorachit* Malicky & Chantaramongkol (1993), *P. isaschar* Malicky (2009), *P. lak* Malicky & Chantaramongkol (1993), *P. samanaka* Malicky & Chantaramongkol (1993), *Paduniella semarangensis* Ulmer (1913). The species found in both study sites were *P. arthit* and *P. intorachit*. The species were found only in the Nam Song as follows: *P. lak* and *P. semarangensis*. The species were only found in the Nam Khan as follows: *P. isaschar* and *P. samanaka*.

Family Stenopsychidae consisted of *Stenopsyche haimavatika* Schmid 1969, which was only found in the Nam Song.

Shannon-Wiener species diversity index (H'), evenness index (E') and Sørensen's similarity coefficient (S) of male adult Trichoptera summarized in Table 4, total numbers of individuals and taxa richness in the Nam Song were higher than those in the Nam Khan, whereas Shannon-Wiener species diversity index and the evenness index in the Nam Khan were higher than those in Nam Song. Sørensen's similarity coefficient of adult Trichoptera between both study sites was 0.22.

Table 4 Numbers of male adult Trichoptera of each family at the Nam Song and Nam Khan, Lao PDR. in November 2017

Male adult Trichoptera	Nam Song	Nam Khan
1. Family Hydropsychidae (12 species)		
<i>Amphipsyche gratiosa</i> Navas, 1922	10	0
<i>Cheumatopsyche charites</i> Malicky & Chantaramongkol, 1997	232	5
<i>C. dubitans</i> Mosely, 1942	16	0
<i>Hydropsyche boreas</i> Malicky & Chantaramongkol, 2000	1	0
<i>Macrostemum floridum</i> Navas, 1929	0	1
<i>Oestropsyche vitrina</i> Hagen, 1859	81	0
<i>Polymorphanisus muluensis</i> Barnaed, 1980	1	0
<i>Potamyia chinensis</i> Ulmer, 1915	13	0
<i>P. elektra</i> Malicky & Chantaramongkol, 2001	4	0
<i>P. periboia</i> Malicky & Chantaramongkol, 1997	0	1
<i>P. phaidra</i> Malicky & Chantaramongkol, 1997	14	0
<i>Pseudoleptonema quinquefasciatum</i> Martynov, 1935	231	0
2. Family Leptoceridae (9 species)		
<i>Ceraclea harpalyke</i> Malicky & Changthong, 2002	4	0
<i>C. trismegistos</i> Malicky, 2008	1	0
<i>Setodes argentiguttatus</i> Gordon & Schmid, 1987	0	1
<i>S. brevicaudatus</i> Yang & Morse, 1989	5	0
<i>S. endymion</i> Malicky & Chantaramongkol, 2000	0	1
<i>S. klakahana</i> Ulmer, 1951	38	0
<i>S. meriones</i> Malicky & Chantaramongkol, 2006	47	0
<i>S. sarapis</i> Malicky & Chantaramongkol, 2006	5	0
<i>S. sp.1</i>	0	2
3. Family Odontoceridae (1 species)		
<i>Marilia aerepe</i> Malicky & Chantaramongkol, 1996	2	0
4. Family Philopotamidae (1 species)		
<i>Chimarra okuihorum</i> Mey, 1998	100	0
5. Family Psychomyiidae (6 species)		
<i>Psychomyia arthit</i> Malicky & Chantaramongkol, 1993	89	1
<i>P. intorachit</i> Malicky & Chantaramongkol, 1993	964	1
<i>P. isaschar</i> Malicky, 2009	0	1

Table 4 Numbers of male adult Trichoptera of each family at the Nam Song and Nam Khan, Lao PDR. in November 2017 (Cont.)

Male adult Trichoptera	Nam Song	Nam Khan
<i>P. lak</i> Malicky & Chantaramongkol, 1993	39	0
<i>P. samanaka</i> Malicky & Chantaramongkol, 1993	0	7
<i>Paduniella semarangensis</i> Ulmer, 1913	33	0
6. Family Stenopsychidae (1 species)		
<i>Stenopsyche haimavatika</i> Schmid, 1969	1	0
Total numbers of individuals	1,931	21
Total taxa richness	23	10
Shannon-Wiener species diversity index (H')	1.82	1.95
Evenness index (E')	0.58	0.85
Sørensen's similarity coefficient (S)		0.22

Discussion and Conclusions

Statistical analysis of 15 parameters of water quality revealed that the water channel width, dissolved oxygen, electrical conductivity, total dissolved solid, suspended solids, biochemical oxygen demand and chlorophyll a were significantly higher ($p < 0.05$) in Nam Khan than those of Nam Song. Depth, water velocity, nitrate nitrogen, orthophosphate and turbidity were non-significant difference ($p > 0.05$). The results showed that water quality in Nam Song was better than that of Nam Khan. The Nam Song is located at the forest; substrates consisted of boulder, cobble, gravel and sand and was less disturbance from human activity. Whereas, the Nam Khan is located close to the rural areas and agricultural land; river bed consisted of coarse gravel, sand and substrates were covered by silt. The Nam Song had heterogeneity habitats, which led to an increase in species diversity (Cramer, Willig, 2005) comparing to homogeneity habitats with erosion of fine sediments in Nam Khan. Such conditions in Nam Khan prevented retreat building of net spinning caddisflies (Albertson, Daniels, 2016). A total of 21,781 individuals (male=1,931, female=20,850) in Nam Song was higher than that of 154 individuals (male=21, female=133) in Nam Khan. Sex ratio was different between both sites, however females were greater than males. The results of the present study were agreed with the finding of Nowinszky (2014) who reported that *Hydropsyche* has a generally high proportion of females (proterogyny). He also stated than an uneven sex ratio because of the higher mortality rate for males in the larval or pupal stages. Species richness was higher in Nam Song (23 species) than that of Nam Khan (10 species); Shannon-Wiener species diversity index and evenness index were higher in Nam Khan than those of Nam Song. Due to some species in Nam Song were most individuals (dominant species) including *Psychomyia intorachit* (964 individuals), *Cheumatopsyche charites* (232 individuals) and *Pseudoleptonema quinquefasciatum* (231 individuals). The first three diverse families were Hydropsychidae, Leptoceridae

and Psychomyiidae, which were supported by many researchers included Laudee, Prommi (2011) and Prommi, Thani (2014). In this study, adults of *Amphipsyche*, *Cheumatopsyche*, *Hydropsyche* and *Potamyia* (Family Hydropsychidae) were found, which were agreed with the occurrence as more diverse and abundance of Trichoptera in streams at Vang Vieng area, Lao PDR. (Jung et al., 2012). Recently, Laudee, Malicky (2017) reported 20 species in 6 families of Trichoptera from Li Phi Falls, the Mekong River, southern Lao PDR. and Hydropsychidae was the most diverse taxa. *Cheumatopsyche charites*, *Psychomyia arthit* and *P. intorachit* occurred in both rivers. *Cheumatopsyche charites* had a wide distribution, which was found in streams and rivers in both high gradient and low gradient beds (Laudee, Prommi, 2011; Prommi, Thani, 2014). Two hundred thirty-nine species of Trichoptera were recorded in Lao PDR. (Malicky, 2010; Laudee, Malicky, 2017; Malicky, Laudee, 2017). Among 30 species of Trichoptera presented in the present study, 15 species are the first reported. They consisted of *Potamyia chinensis* Ulmer, 1915, *P. elektra* Malicky & Chantaramongkol, 2001, *Ceraclea trismegistos* Malicky, 2008, *Setodes argentiguttatus* Gordon & Schmid, 1987, *S. endymion* Malicky & Chantaramongkol, 2000, *S. klakahana* Ulmer, 1951, *S. sarapis* Malicky & Chantaramongkol, 2006, *Marilia aérope* Malicky & Chantaramongkol, 1996, *Chimarra okuihorum* Mey, 1998, *Psychomyia intorachit* Malicky & Chantaramongkol, 1993, *P. isaschar* Malicky, 2009, *P. lak* Malicky & Chantaramongkol, 1993, *P. samanaka* Malicky & Chantaramongkol, 1993, *Paduniella semarangensis* Ulmer, 1913 and *Stenopsyche haimavatika* Schmid, 1969. From the present study, it increased species richness recorded of Lao PDR. up to 254 species.

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