A review of the Ephemeroptera of Finnmark – DNA barcodes identify Holarctic relations

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The knowledge of the diversity and distribution of mayflies in Finnmark county, the northernmost part of mainland Norway, is reviewed. Eighty five DNA barcodes representing 23 species in the region are released and used in association of life stages as well as evaluation of morphological characters commonly used in identification of Scandinavian Ephemeroptera. Comparisons of DNA barcodes from North American species indicate close relations between Norwegian and North American populations of *Acentrella lapponica* Bengtsson, 1912, *Heptagenia dalecarlica* Bengtsson, 1912, *Metretopus borealis* (Bengtsson, 1909), *Ephemerella aurvillii* (Bengtsson, 1908) and *Parameletus chelifer* Bengtsson, 1908. The DNA barcode from *Siphlonurus alternatus* (Say, 1824) cluster closely with specimens of the same species from Finland, but are more than 7.8% different from North American populations, indicating that the Fennoscandian specimens might constitute a separate species. Two species new to Finnmark and ten new province records are reported.

Key words: Ephemeroptera, DNA-barcoding, Finnmark, Norway, North America.

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Introduction

The use of the short, standardized gene sequences in the identification and delimitation of species (so-called DNA-barcoding) has ever since Hebert *et al.* (2003) been increasingly popular in most organism groups. Although exceptions exists (e.g. Whitworth *et al.* 2007), most studies confirm the effectiveness of partial cytochrome c oxidase subunit I (COI) sequences in the identification of insect species (e.g. Zhou *et al.* 2009, Ekrem et al. 2010). DNA-barcoding has also been shown to be a valuable and effective tool in identification and association of life stages also in mayflies (Ball *et al.* 2005, Zhou *et al.* 2009, Webb *et al.* 2012). A comprehensive barcode library of mayflies and other freshwater invertebrates will enable future use of environmental barcoding in monitoring of freshwaters (Hajibabaei *et al.* 2011) and facilitate a common understanding of the taxonomy of this interesting group of insects.

The distribution of the Ephemeroptera in Finnmark was relatively poorly known until Reidar Brekke published a list of Norwegian mayflies, which included 18 species from Finnmark (Brekke 1938). Later the works of Huru (1981a,b,c, 1982) and Eie *et al.* (1982), which cover several watercourses in Finnmark, made important contributions. The most intensively investigated river concerning freshwater invertebrates in Finnmark, undoubtedly, is the River Alta. In connection with a major river regulation in the

1980's, sampling of benthic invertebrates was carried out both before (Huru 1984a, Bergersen 1987, 1992) and several times a year in the period 1993–2007, after the regulation. This resulted in a total of 18 recorded Ephemeroptera species from River Alta (Koksvik 1998, Ugedal *et al.* 2002).

In this paper we present results of DNA-barcoding from 23 Ephemeroptera species from Finnmark and discuss their genetic association to North American populations. We also summarize the distribution of all known Ephemeroptera species in Finnmark in accordance with the revised Strand-system (Økland 1981).

Material and methods

Material used in DNA barcoding was collected in connection with a project focusing on selected aquatic insect groups in Finnmark where more than 100 different sites were visited (see Ekrem *et al.* 2012 for details). Immatures, subimagos and imagos were successfully sequenced; a list of barcoded material and all reference numbers are given in the appendix. All data are available through the public project Ephemeroptera of Finnmark [EPHFI] in the Barcode of Life Data Systems 3.0 (BOLD, Ratnasingham & Hebert 2007). Comparative material from North America was obtained from the public BOLD-dataset Ephemeroptera (mayflies) of North America – Phase I (DATASET-EPNA1) (Webb *et al.* 2012). Comparisons of COI-sequences from Finnmark populations with data present in other public databases were done using megablast searches in GenBank (http://blast.ncbi.nlm.nih.gov/) and a selection of tools provided by BOLD, including Neighbor Joining clustering using the Kimura 2-parameter substitution model (Kimura 1980).

Specimens were identified based on Engblom (1996). In addition to the material collected in the project described by Ekrem *et al.* (2012), data on species distributions are based on specimens sampled in 2007 and unpublished material from the University of Tromsø and the University of Oslo. Description of additional localities not given in Ekrem *et al.* (2012) are described below. The catalogue "Limnofauna norvegica" (Brittain *et al.* 1996) and other available literature (Savolainen 1980, Huru 1984b, Studemann *et al.* 1988, Bækken & Aanes 1990, Koksvik & Arnekleiv 1992, Engblom *et al.* 1993, Skjelkvåle *et al.* 2007, 2009, Schartau *et al.* 2011, see introduction for additional references) have been used as well. Additionally, data were obtained from Species Map Service 1.6, an online web-service provided by the Norwegian Biodiversity Information Centre and GBIF-Norway (Artsdatabanken & GBIF-Norge 2007–2012). Species data obtained from the Species Map Service were originally submitted by BioFokus, Norwegian Entomological Society, Norwegian Institute for Nature Research and Tromsø University Museum. Some of the records in Limnofauna norvegica and the Species Map Service are identical since they in part are based on the same sources.

The known distribution of Ephemeroptera species in Finnmark is presented according to the four provinces defined in the revised Strand system (Økland 1981). **FV**= western part of Finnmark, **FI**= inner part of Finnmark, **FN**= northern part of Finnmark, **FØ**= eastern part of Finnmark. Taxonomic and ecological remarks are presented when relevant.

Description of additional localities

FinLoc108 – **FØ**, Sør-Varanger: Pasvik, Skjellbekken, EIS 160, N69.36507° E29.46006°, 75m a.s.l., leg. R. Knudsen (kick net). Samples taken on both slowly and fast flowing areas, bottom with stone and gravel, 4–5m wide, bank with *Betula* and *Pinus*.

FinLoc109 – **FØ**, Sør-Varanger: Pasvik, bog lake, EIS 168, N69.46846° E29.99791°, 24m a.s.l., leg. R. Knudsen (kick net). Small, nutrient poor lake (0.5 hectares) situated in bog area dominated by *Pinus*.

FinLoc110 – **FØ**, Sør-Varanger: Pasvik, Svanvatn, EIS 169, N69.44464° E30.05249°, 21m a.s.l., leg. R. Knudsen (kick net). Slow flowing part of the Pasvik River with high nutrient content; some areas with dense aquatic vegetation, banks with *Salix*, *Betula* and *Pinus*.

FinLoc111 – FI, Kautokeino: Cunovuohppi, EIS 157, N69.07446° E22.85238°, 374m a.s.l., leg. R. Knudsen (kick net). Shallow (< 3m) bay (approximately 32 hectares) rich of aquatic vegetation; part of the large Lake Stuorajavri (2370 hectares), surrounded by *Salix* and *Betula*.

FinLoc112 – FN, Båtsfjord, Sandfjordelva, EIS 185, N70.45247° E30.44145°, 75m a.s.l., leg. G. Kjærstad (kick net). Fast running river with stony bottom, approximately 30m broad; *Salix* along the margins.

FinLoc113 – FN, Båtsfjord: pond, EIS 185, N70.41102° E30.25205°, 115m a.s.l., leg. G. Kjærstad (kick net). Small pond (<50 m^2) with *Carex*, surrounded by scattered groups of *Salix,* occasionally affected by flood from the adjacent Sandfjordelva River.

FinLoc114 – FN, Vardø: Komagelva, EIS 185, N70.29184° E30.1854°, 103m a.s.l., leg. G. Kjærstad (kick net). Samples taken on both slowly and fast flowing areas, stony bottom and with some gravel, approximately 20m broad, *Salix* along the margins.

FinLoc115 – FN, Alta: pond, EIS 172, N70.08968° E22.4898°, 117m a.s.l., leg. J.E. Brittain (kick net). Pond (0,35 hectares) at Tappeluft surrounded by *Betula*.

FinLoc116 – FØ, Sør-Varanger: stream, EIS 168, N69.43198° E29.84098°, 30m a.s.l., leg. J.E. Brittain (kick net). Small stream at Nyheim, Pasvik, banks with *Betula*.

Results and discussion

DNA barcodes were obtained from 85 specimens representing 23 species from Finnmark (see Appendix) and an additional 137 barcodes were added from Canada. Sixteen species were represented by more than one specimen from Finnmark and showed a mean intraspecific Kimura 2-parameter distance of 0.27%. Maximum observed intraspecific distance (1.34%) was considerably lower than the minimum observed interspecific divergence (14.03%), thus the presence of a "barcode-gap" was obvious in our data. With the addition of the Canadian specimens, only *Ephemera danica* Müller, 1764, *Baetis subalpinus* Bengtsson, 1917 and *Caenis horaria* (Linnaeus 1758) were represented by single specimens, and the maximum intraspecific distance increased to 3.2% (between two Canadian specimens of *Metretopus borealis* (Bengtsson, 1909)) if data on *Siphlonorus alternatus* (Say, 1824) was excluded. When including our sequence of S. alternatus the maximum intraspecific distance observed in our combined dataset increased to 9.6%. DNA barcodes of *B. subalpinus* and *C. horaria* match well with sequences from other European populations of these species in BOLD, while our specimen of *E. danica* is the only currently available COI sequence of this species.

Early instars larvae can be difficult to identify to species-level in mayflies. In our material we could associate larvae of *Alainites muticus* (Linnaeus, 1758)*, Baetis rhodani* (Pictet, 1843)*, Leptophlebia marginata* (Linnaeus, 1767)*, Kageronia fuscogrisea* (Retzius, 1738)*, Heptagenia dalecarlica* Bengtsson, 1912*, Ephemerella mucronata* (Bengtsson, 1909) and *Ameletus inopinatus* Eaton, 1887 to adult counterparts and thus secure high taxonomic resolution also for these life stages.

Comparison with a larger data set including specimens from further south in Norway as well as Canada confirmed the presence of an obvious barcode-gap in COI-sequences of Ephemeroptera and revealed some interesting observations (see comments on species below).

A total of 39 of 48 Norwegian Ephemeroptera species are recorded from Finnmark (Table 1). The review of our material resulted in two species new to Finnmark (*Siphlonurus alternatus* (Say, 1824) and *Ephemera vulgata* Linnaeus, 1758) and ten new province records. The inner (FI) and northern (FN) provinces had the highest species number with 32 and 31, respectively. The western part of Finnmark (FV) had 21 species and the eastern part (FØ) 25 species. Of the 39 species occurring in Finnmark, ten have also been reported from North America.

Although the eastern area of Finnmark (FØ) is the smallest among the four provinces, we expect the species number for this region to increase in the future. Due to its geographical location in the county, this area is known to generally

TABLE 1. Distribution of Ephemeroptera in Finnmark based on the revised Strand-system (Økland 1981). Species marked with an asterisk (*) are also known from North America. Capital **X** in bold font represents new province records.

		FV	FI	FN	FØ
	Ameletidae				
1.	*Ameletus inopinatus* Eaton, 1887*	x	x	x	x
	Ametropodidae				
2.	*Metretopus alter* Bengtsson, 1930*	x			
3.	*Metretopus borealis* (Bengtsson, 1909)*	x	x	x	x
	Baetidae				
4.	*Acentrella lapponica* Bengtsson, 1912*	x	x	x	x
5.	*Alainites muticus* (Linnaeus, 1758)	x	x	x	x
6.	*Baetis fuscatus* (Linnaeus, 1761)	x	x	x	
7.	*Baetis macani* Kimmins, 1957		x	x	**X**
8.	*Baetis rhodani* (Pictet, 1843)	x	x	x	x
9.	*Baetis subalpinus* Bengtsson, 1917	x	x	x	x
10.	*Baetis vernus* Curtis, 1834	x	x	x	
11.	*Nigrobaetis digitatus* Bengtsson, 1912				x
12.	*Nigrobaetis niger* (Linnaeus, 1761)		x		**X**
13.	*Centroptilum luteolum* (Müller, 1776)	x	x	x	x
14.	*Cloeon praetextum* Bengtsson, 1914		x	**X**	
15.	*Cloeon simile* Eaton, 1870		x	**X**	
	Caenidae				
16.	*Caenis horaria* (Linnaeus, 1758)	x	x	x	**X**
17.	*Caenis rivulorum* Eaton, 1884				x
	Ephemerellidae				
18.	*Ephemerella aurivillii* (Bengtsson, 1908)*	x	x	x	x
19.	*Ephemerella mucronata* (Bengtsson, 1909)*	x	x	x	x
20.	*Serratella ignita* (Poda, 1761)		x	x	
	Ephemeridae				
21.	*Ephemera danica* Müller, 1764		x	x	**X**
22.	*Ephemera vulgata* Linnaeus, 1758				**X**
	Heptageniidae				
23.	*Arthroplea congener* Bengtsson, 1908		x	x	
24.	*Heptagenia dalecarlica* Bengtsson, 1912*	x	x	x	x
25.	*Heptagenia sulphurea* (Müller, 1776)	x			x
26.	*Kageronia fuscogrisea* (Retzius, 1738)		x	x	x
27.	*Nixe joernensis* (Bengtsson, 1909)*	x	x	x	x
	Leptophlebiidae				
28.	*Habrophlebia lauta* Eaton, 1884			x	
29.	*Leptophlebia marginata* (Linnaeus, 1767)	x	x	x	x
30.	*Leptophlebia vespertina* (Linnaeus, 1758)	x	x	x	**X**

TABLE 1. *continued*

		FV	FI	FN	FØ
	Leptophlebiidae *continued*				
31.	*Paraleptophlebia cincta* (Retzius, 1783)			x	
32.	*Paraleptophlebia strandii* (Eaton, 1901)		x	x	**X**
33.	*Paraleptophlebia submarginata* (Stephens, 1835)		x	x	
34.	*Paraleptophlebia werneri* Ulmer, 1919		x		
	Siphlonuridae				
35.	*Parameletus chelifer* Bengtsson, 1908*	x	x	x	
36.	*Parameletus minor* (Bengtsson, 1909)		x	x	
37.	*Siphlonurus aestivalis* (Eaton, 1903)	x	x	x	x
38.	*Siphlonurus alternatus* (Say, 1824)*		**X**		
39.	*Siphlonurus lacustris* Eaton, 1870	x	x	x	x
Sum		**21**	**32**	**31**	**25**

TABLE 2. Summary of pairwise Kimura 2-Parameter distances distances between Finnmark and North American populations of Holarctic species.

Species	Number of Specimens	Mean Intraspecific Distance (%)	Maximum Intraspecific Distance (%)	Mean Finnmark vs. North America Distance (%)	Range Finnmark vs. North America Distance (%)
Acentrella lapponica	7	1.5	2.8	2.5	2.2–2.8
Ephemerella aurivillii	12	0.48	0.9	0.7	0.6–0.9
Heptagenia dalecarlica + *Heptagenia pulla*	38	0.2	1.2	0.6	0–1.2
Metretopus borealis	10	0.9	3.2	1.3	1.1–1.9
Nixe joernensis	2	0.6	0.6	0.6	0.6
Parameletus chelifer	3	2.4	3.45	3.45	3.45
Siphlonurus alternatus	84	0.6	9.6	8.3	7.8–9.0

accommodate more species compared to other parts of Finnmark. Islands and main land areas close to the coast, except the Varangerhalvøya peninsula are also relatively poorly surveyed. In addition, the majority of the sampling sites are from riffle areas of rivers. To enhance species number, and to achieve a better understanding of species distribution, slow-flowing areas of rivers and smaller freshwater localities like ponds and brooks in little investigated regions should be sampled in forthcoming studies. Based on the distribution of Ephemeroptera of neighbouring areas in Finland (Savolainen 2009a) we expect species such as i.e. *Procloeon bifidum* (Bengtsson, 1912) to be recorded in the future.

Comparison of Holarctic Species. Ten species recorded from Finnmark are also known from North America; *Ameletus inopinatus* (Eaton, 1887), *Metretopus alter* Bengtsson, 1930, *Metretopus borealis* (Bengtsson, 1909), *Acentrella lapponica* Bengtsson, 1912, *Ephemerella aurivillii* (Bengtsson, 1908), *E. mucronata* (Bengtsson, 1909), *Heptagenia dalecarlica* Bengtsson, 1912/*H. pulla* (Clemens, 1913), *Nixe joernensis* (Bengtsson, 1909), *Parameletus chelifer* Bengtsson, 1908 and *Siphlonurus alternatus* (Say, 1824). Barcodes were available from both Finnmark and Canadian populations for

seven of these (Table 2). Three species, *Acentrella lapponica* Bengtsson, 1912, *Metretopus borealis* (Bengtsson, 1909) and *Parameletus chelifer* Bengtsson, 1908, had relatively high divergence between Finnmark and North America populations (1.1-3.1%) and our specimen of *Siphlonurus alternatus* (Say, 1824) clustered with specimens from Finland that were more than 7.8% different from the North American populations present in BOLD. The remaining three species had much less divergence between continents, but only *Heptagenia dalecarlica* Bengtsson, 1912/ *H. pulla* (Clemens, 1913) shared barcodes. The range of divergence values observed suggests that the dispersal of species between the continents did not occur at the same time.

Comments on selected taxa

AMELETIDAE

***Ameletus inopinatus* Eaton, 1887**

Records. FinLoc9, 13 June 2010, 1 nymph; FinLoc10, 18 June 2010, 1 nymph; FinLoc45, 16 June 2010, 1 nymph; FinLoc47, 16 June 2010, 2♂♂ imagos; FinLoc108, 7 June 2009, 1 nymph; FinLoc110, 12 June 2010, 1 nymph; FinLoc112, 21 July 2007, 13 nymphs; FinLoc113, 22 July 2007, 2 nymphs; FinLoc114, 25 July 2007, 7 nymphs.

Remarks. Engblom (1996) separates *A. inopinatus* from *A. alpinus* based on the shape of the subgenital plate (u-shaped in *A. inopinatus* and v-shaped in *A. alpinus*) and the length of the metatarsus relative to the length of the metatibia. In our DNA-barcode analysis, specimens with differently shaped subgenital plates clustered together with *A. inopinatus* larvae, supporting the synonymy of these two species as first suggested by Brekke (1965) and followed by most workers (i.e. Kluge, 2004; Barber-James *et al.*, 2010). *A. inopinatus* seems to be common and widely distributed in Finnmark in both lotic and lentic waters. It is also known from northern North America (Zloty, 1996), but no sequences were available for comparison.

AMETROPODIDAE

***Metretopus borealis* (Bengtsson, 1909)**

Records. FinLoc64, 16 June 2010, 1♂ imago.

Remarks. *M. borealis* is rather common in Finland with several records close to the Norwegian border (Engblom *et al.* 1993). It almost certainly has a wider distribution in Finnmark than what is currently known. This is also the case for *M. alter* Bengtsson, 1930. The single Finnmark specimen of *M. borealis* differed from North American representatives by 1.1–1.8%; when other specimens of *M. borealis* were included from other regions of Norway, the maximum intraspecific divergence increased to 2.5%.

BAETIDAE

***Acentrella lapponica* Bengtsson, 1912**

Records. FinLoc18, 24 July 2010, 1♂ imago; FinLoc49, 27 July 2010, 1♂ imago; FinLoc90, 1 August 2010, 1♂ imago; FinLoc116, 2 July 1974, nymphs**.**

Remarks. This species is widespread in the northern Palearctic and is also known from northern North America (Randolph & McCafferty, 2005). Specimens from Alberta, Canada occurred in a different cluster from Finnmark specimens and differed by 2.2–2.8%, similar to the maximum divergence observed between North American and Finnmark specimens of *Metretopus borealis*.

***Alainites muticus* (Linnaeus, 1758)**

Records. FinLoc18, 12 June 2010, 1 nymph; FinLoc68, 20 June 2010, 1♂1♀ subimagos; FinLoc106, 29 July 2010, 1♀ imago.

***Baetis macani* Kimmins, 1957**

Records. FinLoc116, 2 July 1974, nymphs.

Remarks. Some of the *B. macani* material, particulary the nymphs collected from lentic waters, may belong to the recently described *Baetis jaervii* Savolainen, 2009 from Finland (Savolainen 2009b).

***Baetis rhodani* (Pictet, 1843)**

Records. FinLoc03, 25 July 2010, 1♂ imago;

FinLoc06, 13 June 2010, 1 nymph; FinLoc09, 13 June 2010, 1 nymph; FinLoc08, 23 July 2010, 1♀ subimago; FinLoc10, 18 June 2010, 1 nymph; FinLoc18, 12 June 2010, 1 nymph; FinLoc56, 15 June 2010, 1 nymph; FinLoc106, 18 July 2010, 1♂ imago; FinLoc108, 7 June 2009, 17 nymphs; FinLoc112, 21 July 2007, 10 nymphs; FinLoc114, 25 July 2007, 4 nymphs.

Remarks. *B. rhodani* is the most common Ephemeroptera species in running waters in Norway, and probably the most common also in Finnmark. Previous studies have shown that European *B. rhodani* consist of a complex of cryptic species (Williams *et al.* 2006; Lucentini *et al.* 2011). In our material from Finnmark there is only one cluster of nearly identical DNA-barcodes. These are 99% identical with haplotypes 48, 49, 50 and 52 from Lucentini *et al.* (2011) and fit within their widely distributed group G3 (Lucentini et al. 2011, Figure 3).

***Baetis subalpinus* Bengtsson, 1917**

Records. FinLoc106, 29 July 2010, 1♂ imago.

Remarks. Initially, one barcoded *Baetis* specimen was identified as *A. muticus* based on wing venation (hind wing with three veins, second vein forked). Barcode similarity of the specimen with *B. subalpinus* in BOLD, however, made us reexamine the male specimen and we could confirm its identity as *B. subalpinus* based on genitalia and colour pattern. Most specimens of *B. subalpinus* have an unforked second longitudinal vein, but in other *Baetis* species aberrant specimens having a forked second vein have been reported (Durfee and Kondratieff, 1993). Thus, wing venation should be used with caution as a diagnostic character for identification of Norwegian male Baetidae.

***Nigrobaetis niger* (Linnaeus, 1761)**

Records. FinLoc108, 7 June 2009, 4 nymphs.

Remarks. DNA barcodes of this species from Finnmark match well (>99.5% similarity) with sequences of the same species from Bulgaria in BOLD.

***Centroptilum luteolum* (Müller, 1776)**

Records. FinLoc67, 19 June 2010, 1♂ subimago; FinLoc68, 20 June 2010, 4♂♂1♀ imagos; FinLoc111, 26 August 1998, 1 nymph.

***Cloeon praetextum* Bengtsson, 1914**

Records. FinLoc55, 26 July 2010, 2 nymphs.

Remarks. The two sequences of *C. praetextum* were highly similar (<1%) to those of *Procloeon mendax* (Walsh, 1862) from northern Canada. The generic limits of the 'long-clawed' Baetidae are poorly known, but *P. mendax* shares many characters with the *Cloeon simile* Eaton, 1870 complex, including *C. praetextum*, such as leaf-shaped gills with palmate trachea and only slightly expanded labial palps in the nymphs, and a large somewhat rectangular process between the bases of the forceps in male adults. It is possible that some North American species currently placed in *Procloeon* Bengtsson, 1915 are congeneric with the *C. simile* complex. *C. praetextum* is morphologically very similar to *C. simile* and some authors (e.g. Bauernfeind & Soldán 2012) do not consider the former as a valid species.

***Cloeon simile* Eaton, 1870**

Records. FinLoc115, July 1974, nymphs.

CAENIDAE

***Caenis horaria* (Linnaeus, 1758)**

Records. FinLoc108, 7 June 2009, 1 nymph; FinLoc111, 28 August 1998, 4 nymphs.

Remarks. One specimen was successfully sequenced and the DNA barcode match well (99.2-99.5% similarity) with unpublished specimens from the German Alps deposited in BOLD.

EPHEMERELLIDAE

***Ephemerella aurivillii* (Bengtsson, 1908)**

Records. FinLoc09, 13 June 2010, 1 nymph; FinLoc18, 12 June 2010, 3 nymphs; FinLoc45, 16 june 2010, 1 nymph; FinLoc56, 15 June 2010, 1 nymph; FinLoc108, 7 June 2009, 6 nymphs; FinLoc112, 21 July 2007, 1 nymph.

Remarks. Barcodes from Finnmark specimens were highly similar to those from eastern North America.

***Ephemerella mucronata* (Bengtsson, 1909)**

Records. FinLoc18, 12 June 2010, 5 nymphs, 24 July 2010, 1♀ subimago.

EPHEMERIDAE

***Ephemera danica* Müller, 1764**

Records. FinLoc45, 16 June 2010, 1 nymph; FinLoc108, 7 June 2009, 1 nymph.

***Ephemera vulgata* Linnaeus, 1758**

Records. FinLoc110, 7 June 2008, 4 nymphs.

Remarks. Sequences from our specimens match well (>98.6% similarity) with specimens of the same species from Finland and Germany present in BOLD.

HEPTAGENIIDAE

***Heptagenia dalecarlica* Bengtsson, 1912**

Records. FinLoc05, 11 June 2010, 1 nymph; FinLoc18; 12 June 2010, 4 nymphs; FinLoc64, 27 July 2010, 1♂ imago; FinLoc108, 7 June 2009, 2 nymphs.

Remarks. Finnmark specimens clustered with northern North American specimens identified as *Heptagenia pulla* (Clemens, 1913). The nymphs of *H. dalecarlica* are characterized by reversed asymmetry in the mandibles where the right is angulated and the left is planate, but the cluster of *H. pulla* contains specimens with both normal and reversed asymmetry. Specimens identified as *H. pulla* from eastern North America, however, have the normal mandibular arrangement and form a separate cluster from both *H. dalecarlica* and other *H. pulla* (not shown). Further study of *H. dalecarlica*, *H. pulla* and related species is required to determine the specific limits.

***Kageronia fuscogrisea* (Retzius, 1738)**

Records. FinLoc18, 24 July 2010, 3♂ imagos; FinLoc45, 16 June 2010, 2 nymphs; FinLoc110, 6 June 2009, 30 nymphs; FinLoc111, 28 August 1998, 1 nymph.

Remarks. DNA barcodes of Finnmark populations of this species had no other close matches in GenBank or BOLD.

***Nixe joernensis* (Bengtsson, 1909)**

Records. FinLoc67, 30 july 2010, 1♂ subimago; FinLoc111, 27 August 1998, 1 nymph.

Remarks. This species is sometimes placed in the genus *Afghanurus* Demoulin, 1961. *Nixe* Flowers, 1980 and *Afghanurus* are closely related genera, but whether they should be treated as separate taxa is unclear. Kluge (2004) treats them all as a large 'plesiomorphon' (lacking synapomorphies) under *Afghanurus*/g2. *Nixe* has some synapomorphies, however, that *N. joernesis* shares, including a unique egg sculpturing. The type species of *Afghanurus* lacks these types of eggs and appears to be most closely related to the "*Ecdyonurus*" *simplicioides* -group (which also occurs in the Palearctic). The exact relationship of the *Afghanurus* type and the *Nixe* type is not clear, but based on the shared apomorphic egg type, *N. joernensis* is closely related to the type species of *Nixe*.

LEPTOPHLEBIIDAE

***Habrophlebia lauta* Eaton, 1884**

Remarks. This species is recorded only once in Norway. According to the coordinates given in Huru (1984b) the locality is situated in Porsanger municipality. The record should therefore be placed in the province FN, and not in FI as presented in Limnofauna norvegica (Brittain *et al.* 1996).

***Leptophlebia marginata* (Linnaeus, 1767)**

Records. FinLoc21, 12 June 2010, 1♂ imago; FinLoc45, 16 June 2010 3 nymphs; FinLoc81, 19 June 2010, 1♀ subimago; FinLoc67, 17 June 2010, 1♀ imago; FinLoc110, 6 June 2009, 29 nymphs.

***Leptophlebia vespertina* (Linnaeus, 1758)**

Records. FinLoc109, 11 June 2010, 3 nymphs; FinLoc110, 6 June 2009, 3 nymphs.

Remarks. DNA barcodes match well (99.2% similarity) with specimens from Finland in BOLD.

***Paraleptophlebia strandii* (Eaton, 1901)**

Records. Finloc 47, 27 July 2010, 1♂ imago; FinLoc53, 26 July 2010, 1♂ imago; FinLoc67, 30.

July 2010, 1♂ imago; FinLoc68, 31 July 2010, 1♂ imago; FinLoc92, 30 July 2010, 1♂ imago.

SIPHOLONURIDAE

***Parameletus chelifer* Bengtsson, 1908**

Records. FinLoc113, 22 July 2007, 1 nymph.

Remarks. This species also occurs in North America and comparison of the single sequence available from Finnmark show high similarity (96.95%) with specimens from Churchill, Canada.

***Siphlonurus alternatus* (Say, 1824)**

Records. FinLoc111, 26 August 1998, 1 nymph; 27 August 1998, 4 nymphs + 1♀ subimago.

Remarks. One specimen was successfully sequenced (NO-EPH101) and clusters with two non-public specimens from Finland with 99–100% similarity. The Finnmark specimen was more than 7.8% different from North American specimens, including specimens from the approximate type locality in Manitoba, Canada. It is possible that the Fennoscandian population might constitute a different species but further morphological and molecular analysis is necessary to confirm this.

***Siphlonurus lacustris* Eaton, 1870**

Records. FinLoc3, 25 July 2010, 1♂ imago; FinLoc18, 24 July 2010, 2♂1♀ imagos; FinLoc113, 22 July 2007, 12 nymphs; FinLoc114, 25 July 2007, 3 nymphs.

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APPENDIX. Overview of sequenced specimens and species from Finnmark.

Sample ID	Species	Life Stage	Locality	Collection Date	Collectors	GenBank Accession
NO-EPH21	*Acentrella lapponica*	Male adult	FinLoc59	27-Jul-2010	T. Ekrem & E. Stur	JN299090
NO-EPH22	*Acentrella lapponica*	Male adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299091
NO-EPH23	*Acentrella lapponica*	Male adult	FinLoc90	01-Aug-2010	T. Ekrem & E. Stur	JN299092
NO-EPH43	*Alainites muticus*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299108
NO-EPH45	*Alainites muticus*	Female adult	FinLoc106	29-Jul-2010	T. Ekrem & E. Stur	JN299110
NO-EPH46	*Alainites muticus*	Male subadult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299111
NO-EPH47	*Alainites muticus*	Female subadult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299112
NO-EPH16	*Ameletus inopinatus*	Larva	FinLoc10	18-Jun-2010	T. Ekrem & E. Stur	JN299086
NO-EPH17	*Ameletus inopinatus*	Larva	FinLoc09	13-Jun-2010	T. Ekrem & E. Stur	JN299087
NO-EPH18	*Ameletus inopinatus*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299088
NO-EPH20	*Ameletus inopinatus*	Male adult	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299089
NO-EPH80	*Ameletus inopinatus*	Male adult	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299140
NO-EPH11	*Baetis rhodani*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299141
NO-EPH12	*Baetis rhodani*	Larva	FinLoc06	13-Jun-2010	T. Ekrem & E. Stur	JN299142
NO-EPH13	*Baetis rhodani*	Larva	FinLoc09	13-Jun-2010	T. Ekrem & E. Stur	JN299143
NO-EPH14	*Baetis rhodani*	Female subadult	FinLoc08	23-Jul-2010	T. Ekrem & E. Stur	JN299144
NO-EPH15	*Baetis rhodani*	Male adult	FinLoc106	18-Jul-2010	T. Ekrem & E. Stur	JN299145
NO-EPH93	*Baetis rhodani*	Larva	FinLoc10	18-Jun-2010	T. Ekrem & E. Stur	JN299146
NO-EPH94	*Baetis rhodani*	Larva	FinLoc56	15-Jun-2010	T. Ekrem & E. Stur	JN299147
NO-EPH95	*Baetis rhodani*	Male adult	FinLoc03	25-Jul-2010	T. Ekrem & E. Stur	JN299081
NO-EPH44	*Baetis subalpinus*	Male adult	FinLoc106	29-Jul-2010	T. Ekrem & E. Stur	JN299082
NO-EPH107	*Caenis horaria*	Larva	FinLoc108	07-Jun-2009	R. Knudsen	KC158559
NO-EPH69	*Centroptilum luteolum*	Male subadult	FinLoc67	19-Jun-2010	T. Ekrem & E. Stur	JN299133
NO-EPH70	*Centroptilum luteolum*	Male adult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299134
NO-EPH71	*Centroptilum luteolum*	Male adult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299135
NO-EPH72	*Centroptilum luteolum*	Male adult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299136
NO-EPH73	*Centroptilum luteolum*	Male adult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299137
NO-EPH74	*Centroptilum luteolum*	Female adult	FinLoc68	20-Jun-2010	T. Ekrem & E. Stur	JN299138
NO-EPH89	*Cloeon praetextum*	Larva	FinLoc55	26-Jul-2010	T. Ekrem & E. Stur	JN299149
NO-EPH90	*Cloeon praetextum*	Larva	FinLoc55	26-Jul-2010	T. Ekrem & E. Stur	JN299150
NO-EPH76	*Ephemera danica*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JX571926
NO-EPH113	*Ephemera vulgata*	Larva	FinLoc110	07-Jun-2008	R. Knudsen	KC158560
NO-EPH114	*Ephemera vulgata*	Larva	FinLoc110	07-Jun-2008	R. Knudsen	KC158561
NO-EPH115	*Ephemera vulgata*	Larva	FinLoc110	07-Jun-2008	R. Knudsen	KC158562
NO-EPH36	*Ephemerella aurivillii*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299102
NO-EPH37	*Ephemerella aurivillii*	Larva	FinLoc56	15-Jun-2010	T. Ekrem & E. Stur	JN299103
NO-EPH38	*Ephemerella aurivillii*	Larva	FinLoc09	13-Jun-2011	T. Ekrem & E. Stur	JN299104
NO-EPH39	*Ephemerella aurivillii*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299105
NO-EPH40	*Ephemerella aurivillii*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299106
NO-EPH41	*Ephemerella aurivillii*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299107

APPENDIX. *continued*

Sample ID	Species	Life Stage	Locality	Collection Date	Collectors	GenBank Accession
NO-EPH63	*Ephemerella mucronata*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299127
NO-EPH64	*Ephemerella mucronata*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299128
NO-EPH65	*Ephemerella mucronata*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299129
NO-EPH66	*Ephemerella mucronata*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299130
NO-EPH67	*Ephemerella mucronata*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299131
NO-EPH68	*Ephemerella mucronata*	Female subadult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299132
NO-EPH48	*Heptagenia dalecarlica*	Larva	FinLoc05	11-Jun-2010	T. Ekrem & E. Stur	JN299113
NO-EPH49	*Heptagenia dalecarlica*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299114
NO-EPH50	*Heptagenia dalecarlica*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299115
NO-EPH51	*Heptagenia dalecarlica*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299116
NO-EPH52	*Heptagenia dalecarlica*	Larva	FinLoc18	12-Jun-2010	T. Ekrem & E. Stur	JN299117
NO-EPH53	*Heptagenia dalecarlica*	Male adult	FinLoc64	27-Jul-2010	T. Ekrem & E. Stur	JN299118
NO-EPH57	*Kageronia fuscogrisea*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299122
NO-EPH58	*Kageronia fuscogrisea*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299123
NO-EPH59	*Kageronia fuscogrisea*	Male adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299124
NO-EPH60	*Kageronia fuscogrisea*	Male adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299125
NO-EPH61	*Kageronia fuscogrisea*	Male adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299126
NO-EPH97	*Kageronia fuscogrisea*	Larva	FinLoc110	06-Jun-2009	R. Knudsen	KC158565
NO-EPH98	*Kageronia fuscogrisea*	Larva	FinLoc110	06-Jun-2009	R. Knudsen	KC158564
NO-EPH99	*Kageronia fuscogrisea*	Larva	FinLoc110	06-Jun-2009	R. Knudsen	KC158563
NO-EPH31	*Leptophlebia marginata*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299098
NO-EPH32	*Leptophlebia marginata*	Female subadult	FinLoc81	19-Jun-2010	T. Ekrem & E. Stur	JN299099
NO-EPH33	*Leptophlebia marginata*	Male adult	FinLoc21	12-Jun-2010	T. Ekrem & E. Stur	JN299100
NO-EPH35	*Leptophlebia marginata*	Female adult	FinLoc67	19-Jun-2010	T. Ekrem & E. Stur	JN299101
NO-EPH91	*Leptophlebia marginata*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299151
NO-EPH92	*Leptophlebia marginata*	Larva	FinLoc45	16-Jun-2010	T. Ekrem & E. Stur	JN299152
NO-EPH116	*Leptophlebia vespertina*	Larva	FinLoc109	07-Jun-2008	R. Knudsen	KC158568
NO-EPH117	*Leptophlebia vespertina*	Larva	FinLoc109	07-Jun-2008	R. Knudsen	KC158566
NO-EPH118	*Leptophlebia vespertina*	Larva	FinLoc109	07-Jun-2008	R. Knudsen	KC158567
NO-EPH19	*Metretopus borealis*	Male adult	FinLoc64	16-Jun-2010	T. Ekrem & E. Stur	JX571929
NO-EPH108	*Nigrobaetis niger*	Larva	FinLoc108	07-Jun-2009	R. Knudsen	KC158571
NO-EPH109	*Nigrobaetis niger*	Larva	FinLoc108	07-Jun-2009	R. Knudsen	KC158570
NO-EPH110	*Nigrobaetis niger*	Larva	FinLoc108	07-Jun-2009	R. Knudsen	KC158569
NO-EPH88	*Nixe joernensis*	Male subadult	FinLoc67	30-Jul-2010	T. Ekrem & E. Stur	JN299148
NO-EPH26	*Paraleptophlebia strandii*	Male adult	FinLoc67	30-Jul-2010	T. Ekrem & E. Stur	JN299093
NO-EPH27	*Paraleptophlebia strandii*	Male adult	FinLoc45	27-Jul-2010	T. Ekrem & E. Stur	JN299094
NO-EPH28	*Paraleptophlebia strandii*	Male adult	FinLoc68	31-Jul-2010	T. Ekrem & E. Stur	JN299095
NO-EPH29	*Paraleptophlebia strandii*	Male adult	FinLoc53	26-Jul-2010	T. Ekrem & E. Stur	JN299096
NO-EPH30	*Paraleptophlebia strandii*	Male adult	FinLoc92	30-Jul-2010	T. Ekrem & E. Stur	JN299097
NO-EPH96	*Parameletus chelifer*	Larva	FinLoc113	22-Jul-2007	G. Kjaerstad	KC158572

APPENDIX. *continued*

Sample ID	Species	Life Stage	Locality	Collection Date	Collectors	GenBank Accession
NO-EPH101	*Siphlonurus alternatus*	Larva	FinLoc111	27-Aug-1998	R. Knudsen	KC158573
NO-EPH54	*Siphlonurus lacustris*	Male adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299119
NO-EPH55	*Siphlonurus lacustris*	Female adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JN299120
NO-EPH56	*Siphlonurus lacustris*	Male adult	FinLoc03	25-Jul-2010	T. Ekrem & E. Stur	JN299121
NO-EPH62	*Siphlonurus lacustris*	Male adult	FinLoc18	24-Jul-2010	T. Ekrem & E. Stur	JX571930