

Hydrobiologia

Seasonal changes in caddis larvae assemblages in river floodplain habitats along a hydrological connectivity gradient

--Manuscript Draft--

Manuscript Number:	HYDR-D-12-07880R3
Full Title:	Seasonal changes in caddis larvae assemblages in river-floodplain habitats along a hydrological connectivity gradient
Article Type:	Primary research paper
Keywords:	biodiversity; seasonality; Trichoptera; connectivity gradient; wetlands; Rhine
Corresponding Author:	Fred van den Brink, Dr. Province of Limburg Maastricht, Limburg NETHERLANDS
Corresponding Author Secondary Information:	
Corresponding Author's Institution:	Province of Limburg
Corresponding Author's Secondary Institution:	
First Author:	Fred van den Brink, Dr.
First Author Secondary Information:	
Order of Authors:	Fred van den Brink, Dr. Gerard Van der Velde, Prof. Dr. Sander Wijnhoven, Dr.
Order of Authors Secondary Information:	
Abstract:	<p>In order to assess the impact of seasonality versus connectivity on the ecological quality of Lower Rhine river floodplain habitats we studied the seasonal variation in diversity and species assemblages of caddis larvae by monthly sampling the littoral zone of four water bodies over a lateral connectivity gradient. Seasonal variation in diversity showed a general pattern in these floodplain habitats and could be related to caddisfly life history and habitat preferences. Despite this seasonal variation, caddis larvae species assemblages in the water bodies appeared to be rather stable and could be firmly related to the position of their habitats over the connectivity gradient. The main channel (lotic habitat) was clearly separated from the lentic water bodies, and also the sparsely vegetated water bodies from the well vegetated water bodies. It is concluded that the impact of connectivity, and related parameters such as stream velocity and vegetational coverage, outweighed the impact of season on the caddis larvae assemblages.</p>
Response to Reviewers:	<p>Dear Dr. Stendera,</p> <p>We have improved the manuscript according to your suggestions, especially with respect to the introduction, data analysis and discussion sections, including all your editorial remarks.</p> <p>We hope our manuscript is now acceptable for publication in your journal.</p> <p>With kind regards,</p> <p>Fred van den Brink</p>

1 **Seasonal changes in caddis larvae assemblages in river-floodplain habitats along a**
2 **hydrological connectivity gradient**

3

4 Fred W.B. van den Brink¹, Gerard van der Velde^{2,3} & Sander Wijnhoven⁴

5

6 ¹ Province of Limburg, Department of Water and Nature Management, P.O. Box 5700, 6202 MA Maastricht, The
7 Netherlands. e-mail: fwb.van.den.brink@prvlimburg.nl.

8 ² Radboud University Nijmegen, Institute for Water and Wetland Research, Department of Animal Ecology and
9 Ecophysiology, Heyendaalseweg 135, 6525 AJ Nijmegen, The Netherlands. e-mail: g.vandervelde@science.ru.nl.

10 ³ Naturalis Biodiversity Center, P.O. Box 9517, 2300 RA Leiden, The Netherlands.

11 ⁴ Royal Netherlands Institute for Sea Research, NIOZ – Yerseke, Monitor Taskforce, P.O. Box 140, 4400 AC
12 Yerseke, The Netherlands. e-mail: sander.wijnhoven@nioz.nl.

13

14 **Abstract**

15 In order to assess the impact of seasonality versus connectivity on the ecological quality of Lower Rhine
16 river floodplain habitats we studied the seasonal variation in diversity and species assemblages of caddis
17 larvae by monthly sampling the littoral zone of four water bodies over a lateral connectivity gradient.
18 Seasonal variation in diversity showed a general pattern in these floodplain habitats and could be related
19 to caddisfly life history and habitat preferences. Despite this seasonal variation, caddis larvae species
20 assemblages in the water bodies appeared to be rather stable and could be firmly related to the position
21 of their habitats over the connectivity gradient. The main channel (lotic habitat) was clearly separated
22 from the lentic water bodies, and also the sparsely vegetated water bodies from the well vegetated water
23 bodies. It is concluded that the impact of connectivity, and related parameters such as stream velocity
24 and vegetation coverage, outweighed the impact of season on the caddis larvae assemblages.

25

26 **Key-words:** biodiversity, seasonality, Trichoptera, connectivity gradient, wetlands, Lower Rhine

27

28

29 **Introduction**

30 Trichoptera are a major component of the insect fauna of natural rivers and an essential part of the river
31 food-web (Mackay & Wiggins, 1979). Due to their high species richness and their large variety in species
32 specific traits they are useful indicators for ecological monitoring and assessment of river ecosystems
33 (e.g. Chovanec et al., 2005, Waringer et al., 2005). Under natural conditions, river-floodplain ecosystems
34 frequently harbour numerous water bodies which are characterized by high spatial and temporal
35 heterogeneity (Amoros et al., 1987), giving rise to high caddisfly species diversity (Tachet et al., 1994;
36 Graf et al., 2006; Van den Brink et al., 2013). These floodplain water bodies can be arranged according to
37 their hydrological connectivity with the main channel, and connectivity is regarded as a key factor for the
38 functioning of river-floodplain ecosystems (Amoros et al., 1987; Van den Brink et al., 2013).
39 Over the years, caddisfly species assemblages have been examined for several large European rivers,
40 such as the Danube, Rhine, Meuse, Rhône, Neman and Bug. Such recordings on caddisfly species
41 assemblages are generally based on light trap catches of adult specimens (Chantaramongkol, 1983;

42 Waringer, 1989, 1991; Van Urk et al., 1991; Tachet et al., 1994; Czachorowski, 2004; Serafin, 2004; Graf
43 et al., 2006). Surprisingly, studies on the seasonal changes in species composition of caddis larvae in
44 riverine floodplain ecosystems are extremely rare. Several studies have focused on the role of river-
45 floodplain connectivity on the caddisfly assemblages (Tachet et al., 1994; Graf et al., 2006; Van den Brink
46 et al., 2013) or the impact of season on flight periods of caddisfly species (Van der Velde & Bergers,
47 1987; Waringer, 1989, 1991; Sommerhäuser et al., 1997). Information on the seasonal variation of caddis
48 larvae assemblages in different floodplain water bodies over a lateral connectivity gradient is entirely
49 lacking. Such information however may be necessary if we wish to conserve or rehabilitate the caddisfly
50 biodiversity in these ecosystems, since species may be present only during a particular season and may
51 be missed by standard monitoring procedures.

52 As many other large rivers the Lower Rhine has been degraded over the years by water pollution and
53 river regulation (Van den Brink et al., 1996). River regulation reduces the connectivity between the lotic
54 and lentic floodplain water bodies and prevents the creation and rejuvenation of aquatic habitats, with
55 negative consequences for biodiversity. In order to restore the biodiversity of the Lower Rhine river-
56 floodplain ecosystems, restoration works have been carried out recently within the frame of the Rhine
57 Action Plan with variable succes.

58 In order to assess the impact of connectivity on the ecological integrity of aquatic habitats in the Lower
59 Rhine floodplains, we studied the spatio-temporal variation in species diversity and species assemblages
60 of caddis larvae over the whole lateral connectivity gradient. In an earlier study on macroinvertebrate
61 species assemblages in the Lower Rhine river ecosystem we found caddisfly species indicative for
62 hydrologically different types of floodplain water bodies (Van den Brink et al., 1994). In Van den Brink et
63 al. (2013) we extended the connectivity gradient by including the lotic component of the floodplain
64 ecosystem. Here we test the relative importance of seasonality versus variation in connectivity of
65 floodplain water bodies for the occurrence and abundance of caddis larvae. Since water temperature
66 strongly influences growth, activity and flight periods of individual caddisfly species in aquatic ecosystems
67 in the temperate climate zone (Mackay & Wiggins, 1979; Higler, 2005), we expected a major impact of
68 season on the caddis larvae assemblages in floodplain habitats over the connectivity gradient.

69

70 **Methods**

71 Study sites

72 The study area is situated on the left bank of the Lower Rhine in the Netherlands, between river kms 876-
73 894. Highest water levels in the Lower Rhine occur normally outside the growth season, during winter and
74 spring. Over the years, water level fluctuations of 6 m occur in this river stretch. At water levels higher
75 than 12 m above sea level (a.s.l.), all water bodies in the active floodplain are connected to the main
76 channel. The water bodies behind the main dikes, i.e. in the former floodplain, are never flooded but are
77 connected to the main channel via groundwater fluxes only.

78 Seasonal fluctuations in occurrence and relative abundance of caddis larvae have been studied during
79 1991 in four Lower Rhine floodplain water bodies along a lateral hydrological connectivity gradient. The
80 four water bodies were characterized according to the typology of Amoros et al. (1987): eupotamon: main
81 channel; plesiopotamon: disconnected channel at a distance of 10 m from main channel and connected
82 via floods when the water level in the main channel exceeds 8 m a.s.l.; paleopotamon: abandoned
83 meander at a distance of 500 m from the main channel and connected via floods when the water level in
84 the main channel exceeds 11.5 m a.s.l.; and paleopotamon-isolated: abandoned meander in the former
85 floodplain at a distance of 650 m from the main channel, isolated from the active floodplain and
86 connected via seepage only, protected against floods by the main dike with a height of 15 m a.s.l.. A
87 further characterization of the study sites is given in Table 1.

88

89 Sampling methods

90 In the Lower Rhine river channel (eupotamon), artificial substrates (rheolithon) were sampled monthly (De
91 Pauw et al., 1994) using standard procedures of the Dutch Institute for Inland Water Management and
92 Waste Water Treatment (Rijkswaterstaat/RIZA, Lelystad, The Netherlands). In three hydrologically
93 different floodplain water bodies (plesiopotamon, paleopotamon and paleopotamon-isolated), the littoral
94 zone was sampled monthly with a dip-net. In order to analyze the importance of littoral substrate on the
95 occurrence of caddis larvae and in order to get a nearly complete picture of caddis larvae species
96 composition, one site with a mineral-sandy substrate and one site with a organic-clayey substrate was
97 selected in each water body for monthly sampling over one year. The net was quickly pushed through the

98 upper 2 cm of the sediment over a randomly selected surface of approximately 2 m². From April to
99 November, during the growth season, when vegetation was present in the selected habitats, sampling
100 occurred between patches of submerged, nymphaeid and marsh vegetation, avoiding sampling of
101 vegetation, although vegetational fragments and vegetational debris was sampled this way. At high water
102 levels sampling was done with the same net by pushing the net through the sediment, using a rubber
103 boat. Wooden fences in the landscape, close to the sampling sites, were used as orientation marks, so
104 that the same areas could be recognized and sampled even in times of floods. All samples were put in
105 plastic bags which were kept cool and transported to the laboratory. After washing over a 500 µm mesh
106 sieve the caddis larvae were preserved in 70% ethanol, identified (keys: Hickin, 1967; Hiley, 1976;
107 Edington & Hildrew, 1995; Wallace et al., 2003; Higler, 2005) and counted.

108

109 Environmental parameters

110 In order to relate the occurrence of caddis larvae species to environmental parameters the following data
111 were recorded for each water body during sampling of the caddis larvae: current velocity, water level
112 fluctuation, connectivity duration, water temperature, pH, Ca, alkalinity, chlorinity, total-N, DIP, organic
113 matter of sediment, relative abundance of aquatic and marsh vegetation and the chlorophyll-a content of
114 the water column. Hydrological parameters were obtained from Rijkswaterstaat/RIZA. The degree of
115 connectivity, here represented by the connectivity duration between the sites and the main channels, was
116 calculated from floodplain elevation maps and long-term (1901-1990) river water level data. The lowest
117 water level of the river at which the water body becomes connected with the main channel was estimated
118 for each water body. From that the long-term average number of days per year on which the water level
119 in the main channel exceeded the connection level during the year was calculated.

120 The relative abundance of aquatic and marsh vegetation was recorded via an estimation of the
121 percentage coverage of aquatic macrophytes and helophytes within two representative 3 x 3 m plots per
122 water body.

123

124 Data analysis

125 Caddis larvae diversity was expressed in terms of species diversity (H') and species richness (N).
126 Species diversity was calculated according to Shannon, as: $H' = -\sum p_i \ln(p_i)$ with p_i as the proportion of
127 species i in the total number of species per sample per habitat type. Species richness was calculated as
128 the total number of observed species during sampling per habitat type, and presented as the total number
129 of observed species per family during sampling per habitat type.
130 Redundancy analysis (RDA), a direct gradient analysis according to a linear response model (as the
131 gradient length was found to be short as identified with a Detrended Correspondence Analysis; DCA),
132 was used in order to ordinate the caddis larvae species abundances (standardized values to ensure data
133 normality) along habitats and seasons and to analyze the importance of selected environmental
134 parameters (standardized values according to remove effects of measurement units) along which the
135 species and habitats were plotted. The selection of the environmental parameters was based on and
136 classified according to an earlier study (Van den Brink et al., 2013) which showed a high correlation of
137 Ca, alkalinity, chlorinity, organic matter of sediment, total-N and DIP with connectivity and chlorophyll-a.
138 Species which were only incidentally found were excluded from this analysis in order to search for
139 robust patterns. Caddis larvae species abundances were standardized according to the following scale: 0
140 = 0, 1 = 1, 2 = 2-3, 3 = 4-8, 4 = 9-17, 5 = 18-34, 6 = 35-67, 7 = 68-132, 8 = 133-261, 9 = > 261 specimens
141 per m^2 . Hydrological parameters were standardized as follows: connectivity duration (CONN): 0 = 0 $d y^{-1}$;
142 1 = 1–5 $d y^{-1}$; 2 = 6–20 $d y^{-1}$; 3 = 21–120 $d y^{-1}$; 4 = >120 $d y^{-1}$; and current velocity (CURR): 0 = 0 $m s^{-1}$;
143 1 = 0–0.5 $m s^{-1}$; 2 = 0.5–1.0 $m s^{-1}$; 3 = 1.0–1.5 $m s^{-1}$. Water level fluctuation (LEVL) was recorded as the
144 difference between maximum and minimum water level recorded during the sampling year. Temperature
145 (TEMP) and chlorophyll-a (CHLA) were log-transformed. Vegetation abundance of aquatic macrophytes
146 (AQVE) and helophytes (MAVE) were both divided into classes: vegetation abundance: 0 = absent, 1 =
147 coverage < 10%, 2 = coverage between 10 and 50%, 3 = coverage > 50%.

148

149 **Results**

150 The diversity of caddis larvae in the four Lower Rhine floodplain water bodies over a lateral connectivity
151 gradient shows large fluctuations during the year (Fig. 1). However, certain spatial and seasonal trends
152 can be distinguished. Generally, highest peaks in diversity occurred in habitats with a low connectivity

153 (paleopotamon and paleopotamon-isolated) and lowest in habitats with a high degree of connectivity
154 (eupotamon and plesiopotamon). Over the year, highest diversity occurred in the periods March-May and
155 July-September and lowest diversity in June and December-January in all four habitats. The fluctuations
156 in species richness roughly follow this diversity pattern (Fig. 2). Moreover, a clear seasonality for most
157 caddis larvae families can be observed. In the eupotamon, Psychomyiidae, Hydroptilidae and
158 Leptoceridae have been observed during winter and spring only, just as the Limnephilidae in the
159 plesiopotamon, paleopotamon and paleopotamon-isolated habitats. In the latter two habitats,
160 Phryganeidae and Hydroptilidae have been found during summer mainly. In contrast to this, Leptoceridae
161 have been found during the whole year in all three sampled lentic habitats.

162 The relative abundances of caddis larvae over the seasons show more details on the species level,
163 whereas the seasons with larval pupae provide information on flight periods (Table 2). Highest
164 abundances of species of Limnephilidae (*Limnephilus decipiens*, *L. affinis*, *L. lunatus* and *Anabolia*
165 *nervosa*) have been found during late winter and early spring. Species belonging to the Leptoceridae
166 (*Oecetis furva*, *O. lacustris*, *Triaenodes bicolor*, *Athripsodes aterrimus*), Hydroptilidae (*Agraylea*
167 *sexmaculata*, *A. multipunctata*, *Oxyethira flavicornis*) and Polycentropodidae (*Cyrnus crenaticornis*, *C.*
168 *flavidus*) showed their highest abundances in summer, when also larval pupae were found. In contrast to
169 this, *Hydropsyche contubernalis*, *Oecetis ochracea*, *Mystacides longicornis* and *M. nigra*, being the most
170 dominant species in the Lower Rhine habitats, were present in the samples during the whole year.
171 Although present in all the three lentic floodplain water bodies, the maximum abundance of *Mystacides*
172 *longicornis* was found to decrease with increasing hydrological connectivity, and was not found at all in
173 the main channel.

174 The RDA of seasonal data on relative abundance of caddisfly species in the four floodplain water bodies
175 over the hydrological connectivity gradient shows that the eupotamon, plesiopotamon, paleopotamon and
176 paleopotamon-isolated habitats are all well separated, indicating that the impact of hydrological variation
177 is larger than the impact of temporal variation on the occurrence and abundance of caddis larvae species
178 (Fig. 3). A strong species–environment correlation was found for both of the two first redundancy axes
179 together explaining 71.3 % of the total variance in the species data (Table 3). There was a strong positive
180 correlation of the first axis with current velocity, water level fluctuation and connectivity duration and a

181 negative correlation with both aquatic and marsh vegetation coverage, clearly separating the eupotamon
182 from the isolated paleopotamon. The second axis with the strongest (negative) correlation with current
183 velocity but particularly determined by the presence of one species, *Oecetis ochracea*, clearly
184 distinguishes the plesiopotamon from the eupotamon and the isolated paleopotamon. The paleopotamon
185 seemingly does not have a very distinguishing caddis community, with some elements of the isolated
186 paleopotamon and some of the plesiopotamon. The seasons, as the temperature, did not show
187 substantial correlation with one of the two first redundancy axes, and it can be concluded that it therefore
188 only had minor influence on the caddis larvae composition. Characteristic species of the eupotamon are
189 *Hydropsyche contubernalis*, *Ecnomus tenellus*, *Ceraclea dissimilis* and *Psychomyia pusilla*. Species like
190 *Limnephilus decipiens*, *L. lunatus* and *L. affinis* have been found to occur most dominantly in the
191 paleopotamon, whereas *Molanna angustata*, *Mystacides nigra* and *Anabolia nervosa* showed highest
192 relative abundances in the paleopotamon-isolated habitat. The paleopotamon and paleopotamon-isolated
193 habitats differed mainly with respect to their major substrates and nutrient levels (Table 1).
194 With respect to their substrate preference in the floodplain water bodies, species such as *Hydropsyche*
195 *contubernalis*, *H. bulgaromanorum*, *H. exocellata*, *Hydroptila spec.*, *Psychomyia pusilla*, *Tinodes*
196 *waeneri*, *Ceraclea annulicornis*, *C. dissimilis*, *C. senilis*, *Mystacides azurea* and *Oecetis testacea* have
197 been found exclusively on stones in the lotic main channel (rheolithal) (Table 4). In the lentic water
198 bodies, *Oecetis ochracea*, *Mystacides longicornis*, *M. nigra*, *Molanna angustata*, *Limnephilus lunatus* and
199 *Anabolia nervosa* have been found on the sandy, mineral substrates between the vegetational stands
200 (phytopsammal) mainly, whereas species such as *Oecetis furva* and *Triaenodes bicolor* have been found
201 mainly between the vegetational stands on clayey, organic substrates (phytopelal) (Table 4).

202

203 **Discussion**

204 Seasonal variation in caddis larvae diversity showed large fluctuations over the year in a general pattern
205 with peaks in March-May and July-September, clearly related to the occurrence of Limnephilidae and
206 Psychomyiidae in winter/spring and most Leptoceridae, Hydroptilidae, Phryganeidae and
207 Polycentropodidae in summer. A minimum diversity value was found during June, when the species
208 assemblage in the lotic main channel was dominated by *Hydropsyche contubernalis* and in the lentic

209 water bodies by *Mystacides longicornis*. This seasonal variation in diversity was found to be related to the
210 life cycles and flight periods of these semi-aquatic insects. Limnephilid species (*Limnephilus affinis*, *L.*
211 *decipiens*, *L. lunatus*, *L. flavicornis* and *Anabolia nervosa*) appeared some weeks after inundation of the
212 helophyte stands during winter and spring floods (Van den Brink et al., 1994). During winter and spring
213 floods, egg packages of the above-mentioned Limnephilid species came into contact with water, probably
214 giving the trigger for hatching. The extensive helophyte stands in the paleopotamon habitats may be
215 regarded as semi-permanent habitats, since they usually fall dry during the summer months. The
216 observed Limnephilid species are typical inhabitants of such habitats, which survive droughts with an
217 adult diapause in summer (Mackay & Wiggins, 1979; Otto, 1981; Waringer & Graf, 2002; Serafin, 2004;
218 Graf et al., 2008). During spring Limnephilid pupae (*Limnephilus decipiens*, *L. lunatus* and *Anabolia*
219 *nervosa*) were found, so the absence of larvae of these species in summer and autumn can be clearly
220 related to their flight periods (Graf et al., 2008). The occurrence and abundance of Polycentropodidae
221 (*Cyrnus crenaticornis*, *C. flavidus*) and Hydroptilidae (*Agraylea sexmaculata*, *A. multipunctata*, *Oxyethira*
222 *flavicornis*) larvae and pupae in summer was positively correlated with the seasonal development of
223 aquatic macrophytes, mainly of the extensive nymphaeid macrophyte (*Nymphaea alba*, *Nuphar lutea*)
224 stands in the paleopotamon and paleopotamon-isolated habitats. The most abundant species, i.e. *Cyrnus*
225 *crenaticornis* and *C. flavidus*, deposit their eggs underneath the floating leaves of nymphaeids (Van der
226 Velde & Bergers, 1987), whereas *Agraylea sexmaculata*, *A. multipunctata* and *Oxyethira flavicornis* are
227 related to aquatic vegetation and attached algae as a habitat for food and/or material for case building
228 (Higler, 2005; Graf et al., 2008). Although present at the family level during the whole year, Leptoceridae
229 species were found to be most diverse with respect to their occurrence over the seasons. The most
230 abundant species (*Oecetis ochracea*, *Mystacides longicornis*) were present during the whole year with
231 their flight periods in spring and summer. The seasonal differences in abundance of the mentioned most
232 abundant aquatic insect species are generally in agreement with other observations on their life cycles
233 and flight periods (Van der Velde & Bergers, 1987; Waringer, 1991; Sommerhäuser et al., 1997; Higler,
234 2005; Graf et al., 2008). Moreover, a similar pattern in diversity fluctuations during the summer period has
235 been observed in an earlier study (Van der Velde & Bergers, 1987) on caddisfly imagines, caught on
236 adhesive traps on waterlily leaves in the same paleopotamon water body. This pattern was also related to

237 flight periods of most of the same species involved, illustrating the robustness and generality of this
238 seasonal diversity pattern.

239 Despite the large fluctuations in diversity and species richness over the seasons, the impact of spatial
240 variation in the distribution of species over the lateral floodplain connectivity gradient outweighed the
241 impact of season on the caddis larvae species assemblages. In the Lower Rhine, species belonging to
242 the Hydropsychidae and Psychomyiidae have been found exclusively in the eupotamon habitats with
243 *Hydropsyche contubernalis* being the most dominant species there, a situation which was also observed
244 in the natural Neman river (Czachorowski, 2004) and the Danube river (Chantaramongkol, 1983;
245 Waringer, 1991). Hydropsychids such as *Hydropsyche contubernalis*, *H. bulgaromanorum* and *H.*
246 *exocellata* are passive filter-feeders which are dependent on stream velocity and therefore restricted to
247 the lotic eupotamon (Higler, 2005; Graf et al., 2008). Species belonging to Limnephilidae,
248 Polycentropodidae, Phryganeidae and several Hydroptilidae (*Agraylea sexmaculata*, *A. multipunctata*,
249 *Oxyethira flavicornis*) have been found predominantly in the well vegetated lentic paleopotamon and
250 paleopotamon-isolated habitats. These species are typical limnobionts or limnophils, and phytophils
251 (Higler, 2005; Graf et al., 2008), so their absence in the eupotamon and plesiopotamon habitats of the
252 Lower Rhine may be well explained by respectively current velocity and absence of extensive helophyte
253 and nymphaeid macrophyte stands there (Van den Brink et al., 1996). Leptoceridae have been found to
254 occur in all habitats, although most species were highly characteristic. E.g. *Ceraclea dissimilis* occurred
255 on stones in the eupotamon (rheolithal) only, *Oecetis ochracea* was found as a typical inhabitant of the
256 mineral sand flats in the plesiopotamon habitat (Van den Brink et al., 1994), whereas *Oecetis furva* and
257 *Triaenodes bicolor* were mainly found on the organic clayey substrates between the vegetational stands
258 (phytopelal) in the well-vegetated paleopotamon habitats. These different preferences might be related to
259 the material of the cases of these species and their feeding traits. *Ceraclea dissimilis* is a
260 scraper/gatherer which feeds on the diatoms and organic matter on the stones at the Lower Rhine river
261 banks; *Oecetis ochracea* has a mineral case and was found to predate on the chironomid larvae in the
262 sand flats of the plesiopotamon (own observations), whereas *Oecetis furva* and *Triaenodes bicolor* are
263 shredders which feed on aquatic macrophytes and built their case from macrophyte tissue (Higler, 2005;
264 Graf et al., 2008). Although the paleopotamon and paleopotamon-isolated habitats showed comparable

265 caddis larvae family assemblages and species assemblages (Van den Brink et al., 2013), we found
266 differences on the community level, which was related to the higher relative abundances of *Anabolia*
267 *nervosa*, *Mystacides longicornis*, *M. nigra* and *Molanna angustata* in the paleopotamon-isolated habitat.
268 Since all these species have a mineral case their higher abundance might be related to the dominant
269 substrate (psammal) in the latter habitat.

270 In the present study a total number of 34 caddisfly species has been found as larvae in the Lower Rhine
271 floodplain water bodies, which is slightly lower than the 38 species found in an earlier study (Van den
272 Brink et al., 2013). Studies on adult caddisfly species with the use of light traps or adhesive traps along
273 the Lower Rhine did not result in higher species numbers (Van der Velde & Bergers, 1987; Van Urk et al.,
274 1991). This is substantial lower as observed in more natural European rivers (Tachet et al., 1994;
275 Czachorowski, 2004; Serafin, 2004; Graf et al., 2006) or as observed from palaeolimnological data from
276 the Lower Rhine (Klink, 1989), mainly due to the poor habitat variation and eutrophication in the Lower
277 Rhine eupotamon and plesiopotamon habitats (Van den Brink et al., 1996, 2013). Despite this
278 impoverishment, the general stable pattern of caddisfly family distribution over the lateral connectivity
279 gradient along the Lower Rhine shows much resemblance with those in the Danube, the Rhône, and the
280 Neman (Tachet et al., 1994; Czachorowski, 2004; Waringer et al., 2005; Graf et al., 2006), with the same
281 or related species occurring in similar habitats (Van den Brink et al., 2013). In all these rivers
282 Hydropsychidae and Psychomyiidae were found to occur in the eupotamon only. Polycentropodidae,
283 Phryganeidae, Hydroptilidae and Limnephilidae were mainly found in the paleopotamon, whereas
284 Leptoceridae species have been found in all river floodplain habitats. In order to preserve maximum
285 caddisfly diversity it is therefore important that river restoration works should pay attention to the
286 conservation of the whole hydrological range of floodplain habitats over the connectivity gradient,
287 including lotic, lentic, dynamic and low dynamic habitats.

288

289 **Acknowledgements**

290 This study was financially supported by the Dutch Institute for Inland Water Management and Waste
291 Water Treatment, Rijkswaterstaat/RIZA, Lelystad, The Netherlands and the Sandoz Rheinfund, Basel,
292 Switzerland. Rijkswaterstaat/RIZA, Arnhem, provided hydrological data and detailed river maps. Dr.

293 L.W.G. Higler † checked the identification of the Limnephilidae larvae. This is Monitor Taskforce
294 Publication Series 2013-04.

295

296 **References**

297 Amoros, C., A.L. Roux, J.L. Reygrobellet, J.P. Bravard & G. Pautou, 1987. A method for applied
298 ecological studies of fluvial hydrosystems. *Regulated Rivers: Research & Management* 1: 17-36.

299 Chantaramongkol, P., 1983. Light-trapped caddisflies (Trichoptera) as water quality indicators in large
300 rivers: Results from the Danube at Veröce, Hungary. *Aquatic Insects* 5: 33-37.

301 Chovanec, A., J. Waringer, M. Straif, W. Graf, W. Reckendorfer, A. Waringer-Löschenkohl, H.

302 Waidbacher & H. Schultz, 2005. The Floodplain Index – a new approach for assessing the ecological
303 status of river/floodplain systems according to the EU Water Framework Directive. *Large Rivers* 15,
304 *Archiv für Hydrobiologie, Supplement* 155: 169-185.

305 Czachorowski, S., 2004. The last natural river of eastern Europe? Caddisflies (Trichoptera) of the Neman
306 River. *Latvijas Entomologs* 41: 44-51.

307 De Pauw, N., V. Lambert, A. van Kenhove & A. bij de Vaate, 1994. Comparison of two artificial substrate
308 samplers for macroinvertebrates in biological monitoring of large and deep rivers and canals in Belgium
309 and The Netherlands. *Journal of the Environmental Monitoring Association* 30: 25-47.

310 Edington, J.M. & A.G. Hildrew, 1995. A revised key to the caseless caddis larvae of the British Isles, with
311 notes on their ecology. F.B.A. Publication, 43, 91 pp.

312 Graf, W., J. Waringer, P. Wenzl, A. Chovanec & O. Moog, 2006. The river Danube - biodiversity and
313 habitat assessment based on Trichoptera assemblages. *Proceedings of the 36th International Conference*
314 *of the International Association of the Danube. Austrian Communications of the Danube Research,*
315 *Vienna.*

316 Graf, W., J. Murphy, J. Dahl, C. Zamora-Munoz & M.J. Lopez-Rodriguez, 2008. Distribution and
317 ecological preferences of European freshwater organisms. Volume 1. Trichoptera. (Schmidt-Kloiber, A. &
318 D. Hering, eds.). Pensoft Publishers, Sofia-Moscow, 388 pp.

319 Hickin, N.E., 1967. Caddis larvae. Larvae of the British Trichoptera. Hutchinson, London, 476 pp.

320 Higler, B., 2005. De Nederlandse kokerjufferlarven (in Dutch). KNNV Uitgeverij, Utrecht, 159 pp.

321 Hiley, P.D., 1976. The identification of British Limnephilid larvae (Trichoptera). *Systematic Entomology*, 1:
322 147-167.

323 Klink, A., 1989. The Lower Rhine. Palaeoecological analysis. In: *Historical change of large alluvial rivers:*
324 *western Europe:183-201* (G.E. Petts, ed.), John Wiley & Sons Ltd., Chichester.

325 Mackay, R.J. & G.B. Wiggins, 1979. Ecological diversity in Trichoptera. *Annual Review of Entomology* 24:
326 185-208.

327 Otto, C., 1981. Why does duration of flight periods differ in caddisflies? *Oikos* 37: 383-386.

328 Serafin, E., 2004. Species diversity of the caddisflies (Trichoptera) in the left bank river Bug valley. *Teka*
329 *Komunikat Ochrony Kształcenia Srodowisko Przyrody* 1: 195-201.

330 Sommerhäuser, M., B. Robert & H. Schuhmacher, 1997. Flight periods and life history strategies of
331 caddisflies in temporary and permanent woodland brooks in Lower Rhine area (Germany). *Proceedings*
332 *of the 9th International Symposium on Trichoptera*, 425-433. Chiang Mai 1998, Faculty of Science,
333 University of Chiang Mai.

334 Tachet H., P. Usseglio-Polatera & C. Roux, 1994. Theoretical habitat templates, species traits, and
335 species richness: Trichoptera in the Upper Rhône and its floodplain. *Freshwater Biology* 31: 397-416.

336 Van den Brink, F.W.B., M.J. Beljaards, N.C.A. Boots & G. van der Velde, 1994. Macrozoobenthos
337 abundance and community composition in three Lower Rhine floodplain lakes with varying inundation
338 regimes. *Regulated Rivers: Research & Management* 9: 279-293.

339 Van den Brink, F.W.B., G. van der Velde, A.D. Buijse & A.G. Klink, 1996. Biodiversity in the Lower Rhine
340 and Meuse river-floodplains: its significance for ecological management. *Netherlands Journal of Aquatic*
341 *Ecology* 30: 129–149.

342 Van den Brink, F.W.B., G. van der Velde & S. Wijnhoven, 2013. Diversity, occurrence and feeding traits
343 of caddisfly larvae as indicators for ecological integrity of river-floodplain habitats along a connectivity
344 gradient. *Ecological Indicators* 25: 92-98.

345 Van der Velde, G., Bergers, P.J.M., 1987. The temporal and spatial distribution of adult Trichoptera on
346 the upper surfaces of the floating leaves of nymphaeids. A study using adhesive traps. *Proceedings of*
347 *the 5th International Symposium on Trichoptera. Series Entomologica* 39: 319-324. Dr. W. Junk Publ.,
348 Dordrecht.

349 Van Urk, G., F.C.M. Kerkum & A. bij de Vaate, 1991. Caddisflies of the lower Rhine. Proceedings of the
350 6th International Symposium on Trichoptera, 89-94. Adam Mickiewicz University Press, Poznan.

351 Wallace, I.D., B. Wallace & G.N. Philipson, 2003. A key to the case-bearing caddis larvae of Britain and
352 Ireland. FBA Sci. Publ., 61, 269 pp.

353 Waringer, J., 1989. The abundance and temporal distribution of caddisflies (Insecta: Trichoptera) caught
354 by light traps on the Austrian Danube from 1986 to 1987. *Freshwater Biology* 21: 387-399.

355 Waringer, J., 1991. Phenology and the influence of meteorological parameters on the catching success of
356 light-trapping for Trichoptera. *Freshwater Biology* 25: 307-319.

357 Waringer, J. & W. Graf, 2002. Trichoptera communities as a tool for assessing the ecological integrity of
358 Danubian floodplains in Lower Austria. Proceedings of the 10th International Symposium on Trichoptera.
359 Nova Supplementa Entomologica, Keltern 15: 617-623.

360 Waringer, J., A. Chovanec, M. Straif, W. Graf, W. Reckendorfer, A. Waringer-Löschenkohl, H.
361 Waidbacher & H. Schultz, 2005. The Floodplain Index – habitat values and indication weights for
362 molluscs, dragonflies, caddisflies, amphibians and fish from Austrian Danube floodplain waterbodies.
363 *Lauterbornia* 52: 177-187.

364 Table 1. Characterization of four Lower Rhine floodplain habitats over a lateral connectivity gradient.
 365 Annual mean (\pm SD) values of physico-chemical parameters as well as vegetation coverage (last two
 366 rows) in the four habitats are presented (data from Van den Brink et al., 1994).

Parameter	eupotamon	plesiopotamon	paleopotamon	paleopotamon-isolated
Connectivity duration (day.yr ⁻¹)	365	90	1	0
Water level fluctuation (m)	5	3	2	1
Current velocity (m.s ⁻¹)	1.5	0	0	0
pH	7.8 (0.1)	8.1 (0.5)	7.9 (0.2)	8.0 (0.2)
Calcium (mmol.l ⁻¹)	1.7 (0.2)	1.1 (0.3)	1.4 (0.2)	1.2 (0.4)
HCO ₃ ⁻ (mmol.l ⁻¹)	2.2 (0.2)	1.8 (0.3)	2.3 (0.2)	2.8 (0.4)
Cl ⁻ (mmol.l ⁻¹)	4.4 (1.5)	2.6 (0.5)	2.0 (0.4)	1.7 (0.2)
Total-N (μ mol.l ⁻¹)	330 (50)	150 (130)	90 (100)	30 (30)
DIP (μ mol.l ⁻¹)	1.4 (0.4)	0.9 (0.7)	0.3 (0.4)	0.1 (0.01)
Chlorophyll-a (μ g.l ⁻¹)	20 (10)	50 (40)	10 (5)	5 (5)
Organic matter sediment (%)	1	5	20	10
Major microhabitat	rheolithal	psammal	phytopelal	phytopsammal
Coverage aquatic vegetation (%)	0	10-50 ¹	> 50 ²	> 50 ³
Coverage marsh vegetation (%)	0	< 10 ⁴	> 50 ⁵	> 50 ⁶

367 Aquatic vegetation species composition:

368 ¹: *Nymphoides peltata*, *Potamogeton pectinatus*

369 ²: *Nuphar lutea*, *Nymphaea alba*, *Nymphoides peltata*, *Ranunculus circinatus*, *Potamogeton lucens*, *P. pectinatus*
 370 *Fontinalis antipyretica*

371 ³: *Nuphar lutea*, *Nymphaea alba*, *Elodea nuttalli*, *Ranunculus circinatus*, *Potamogeton crispus*, *P. lucens*, *P.*
 372 *pectinatus*, *P. perfoliatus*, *Chara vulgaris*, *Ch. globularis*, *Hippuris vulgaris*, *Fontinalis antipyretica*

373 Marsh vegetation species composition:

374 ⁴: *Carex acuta*, *Phalaris arundinacea*

375 ⁵: *Glyceria maxima*, *Phragmites australis*, *Scirpus lacustris*, *Typha angustifolia*

376 ⁶: *Phragmites australis*, *Typha latifolia*, *Hippuris vulgaris*

377 Table 2. Fluctuations in maximum relative abundances of caddis larvae over the seasons: 1 = 1, 2 = 2-3,
 378 3 = 4-8, 4 = 9-17, 5 = 18-34, 6 = 35-67, 7 = 68-132, 8 = 133-261, 9 = > 261 specimens per m². Shaded
 379 cells indicate season in which pupae were found. Bold lined cells indicate main flight periods according to
 380 Graf et al. (2008). *species codes used in figure 3.

Taxon name	Taxon code*	Winter	Spring	Summer	Autumn
<i>Hydropsyche contubernalis</i> McLachlan	Hyco	8	7	6	9
<i>Hydropsyche bulgaromanorum</i> Malicky		1	1	1	1
<i>Hydropsyche exocellata</i> Dufour			1		1
<i>Ecnomus tenellus</i> (Rambur)	Ecte	1	1	2	1
<i>Hydroptila</i> spec.			1		
<i>Agraylea multipunctata</i> Curtis	Agmu			3	1
<i>Agraylea sexmaculata</i> Curtis	Agse			2	
<i>Oxyethira flavicornis</i> (Pictet)	Oxfl			3	1
<i>Holocentropus picicornis</i> (Stephens)			1	1	
<i>Holocentropus stagnalis</i> (Albarda)	Host		3		
<i>Cyrnus crenaticornis</i> (Kolenati)	Cycr	1	1	4	1
<i>Cyrnus flavidus</i> McLachlan	Cyfl	1	2	5	1
<i>Cyrnus trimaculatus</i> (Curtis)			1	1	
<i>Psychomyia pusilla</i> (Fabricius)	Pspu	1	3		
<i>Tinodes waeneri</i> (L.)		1			
<i>Molanna angustata</i> Curtis	Moan	1	2	5	2
<i>Agrypnia pagetana</i> Curtis				1	
<i>Ceraclea annulicornis</i> (Stephens)			1		
<i>Ceraclea dissimilis</i> (Stephens)	Cedi	1	3		
<i>Ceraclea senilis</i> Burmeister			1		
<i>Athripsodes aterrimus</i> (Stephens)	Atat		2	3	1
<i>Mystacides azurea</i> L.		1			
<i>Mystacides longicornis</i> L.	Mylo	4	9	6	5
<i>Mystacides nigra</i> (L.)	Myni	3	3	4	4
<i>Oecetis furva</i> Rambur	Oefu		1	2	
<i>Oecetis lacustris</i> Pictet	Oela		1	2	
<i>Oecetis ochracea</i> (Curtis)	Oeoc	5	4	2	7
<i>Oecetis testacea</i> (Curtis)			1		
<i>Trienodes bicolor</i> (Curtis)	Trbi		3	6	2
<i>Anabolia nervosa</i> (Curtis)	Anne	3	4	1	
<i>Limnephilus affinis</i> Curtis	Liaf	5	1		
<i>Limnephilus decipiens</i> Kolenati	Lide	3	1		
<i>Limnephilus flavicornis</i> (Fabricius)		1			
<i>Limnephilus lunatus</i> Curtis	Lilu	3	2		

381
 382

383 Table 3. Results of Redundancy Analysis (RDA) between relative abundances of caddisfly species and
 384 seasons and environmental characteristics including the water body typology.

	Axis 1	Axis 2
Summary statistics of first two redundancy axes		
Eigenvalues	0.513	0.200
Species-environment correlations	0.997	0.997
Cumulative percentage variance		
of species data	51.3	71.3
of species-environment relation	53.9	75.0
Correlation of environmental variables with redundancy axes		
Eupotamon (EU)	0.8753	-0.4522
Plesiopotamon (PLE)	0.0421	0.8551
Paleopotamon (PAL)	-0.2607	0.1054
Paleopotamon- isolated (PALi)	-0.6567	-0.5083
Winter	0.0941	0.1460
Spring	-0.0839	-0.0880
Summer	-0.1070	-0.1424
Autumn	0.0967	0.0845
Temperature (TEMP)	-0.1231	-0.1766
Current velocity (CURR)	0.8395	-0.3505
Water level fluctuation (LEVL)	0.7411	0.1694
Connectivity duration (CONN)	0.7221	0.3299
Chlorophyll-a (CHLA)	0.3278	0.2047
Coverage aquatic vegetation (AQVE)	-0.7066	-0.2542
Coverage marsh vegetation (MAVE)	-0.6180	-0.0820

385
 386

387 Table 4. Frequency of occurrence (1 = 1-20 %, 2 = 21-40 %, 3 = 41-60 %, 4 = 61-80%, 5 = 81-100 %) of
 388 caddis larvae per microhabitat.

Taxon name	Rheolithal	Phytopsammal	Phytopelal
<i>Hydropsyche contubernalis</i> McLachlan	5		
<i>Hydropsyche bulgaromanorum</i> Malicky	5		
<i>Hydropsyche exocellata</i> Dufour	5		
<i>Ecnomus tenellus</i> (Rambur)	4	1	
<i>Hydroptila</i> spec.	5		
<i>Agraylea multipunctata</i> Curtis		3	2
<i>Agraylea sexmaculata</i> Curtis		3	2
<i>Oxyethira flavicornis</i> (Pictet)		3	2
<i>Holocentropus picicornis</i> (Stephens)		2	3
<i>Holocentropus stagnalis</i> (Albarda)			5
<i>Cyrnus crenaticornis</i> (Kolenati)		2	3
<i>Cyrnus flavidus</i> McLachlan		3	2
<i>Cyrnus trimaculatus</i> (Curtis)		3	2
<i>Psychomyia pusilla</i> (Fabricius)	5		
<i>Tinodes waeneri</i> (L.)	5		
<i>Molanna angustata</i> Curtis		4	1
<i>Agrypnia pagetana</i> Curtis		1	4
<i>Ceraclea annulicornis</i> (Stephens)	5		
<i>Ceraclea dissimilis</i> (Stephens)	5		
<i>Ceraclea senilis</i> Burmeister	5		
<i>Athripsodes aterrimus</i> (Stephens)		3	2
<i>Mystacides azurea</i> L.	4	1	
<i>Mystacides longicornis</i> L.		4	1
<i>Mystacides nigra</i> (L.)		4	1
<i>Oecetis furva</i> Rambur			5
<i>Oecetis lacustris</i> Pictet		3	2
<i>Oecetis ochracea</i> (Curtis)		5	
<i>Oecetis testacea</i> (Curtis)	5		
<i>Triaenodes bicolor</i> (Curtis)		1	4
<i>Anabolia nervosa</i> (Curtis)		4	1
<i>Limnephilus affinis</i> Curtis		2	3
<i>Limnephilus decipiens</i> Kolenati		2	3
<i>Limnephilus flavicornis</i> (Fabricius)			5
<i>Limnephilus lunatus</i> Curtis		4	1

389
 390

391 **Figure legends**

392

393 Fig. 1. Seasonal variation in caddis larvae diversity in four floodplain habitats (EU= eupotamon, PLE= plesiopotamon,
394 PAL= paleopotamon, PALi= paleopotamon isolated) over a lateral connectivity gradient along the Lower Rhine.

395

396 Fig. 2. Seasonal variation in caddis larvae species numbers (per family) in four floodplain habitats (A: eupotamon, B:
397 plesiopotamon, C: paleopotamon, D: paleopotamon isolated) over a lateral connectivity gradient along the Lower
398 Rhine.

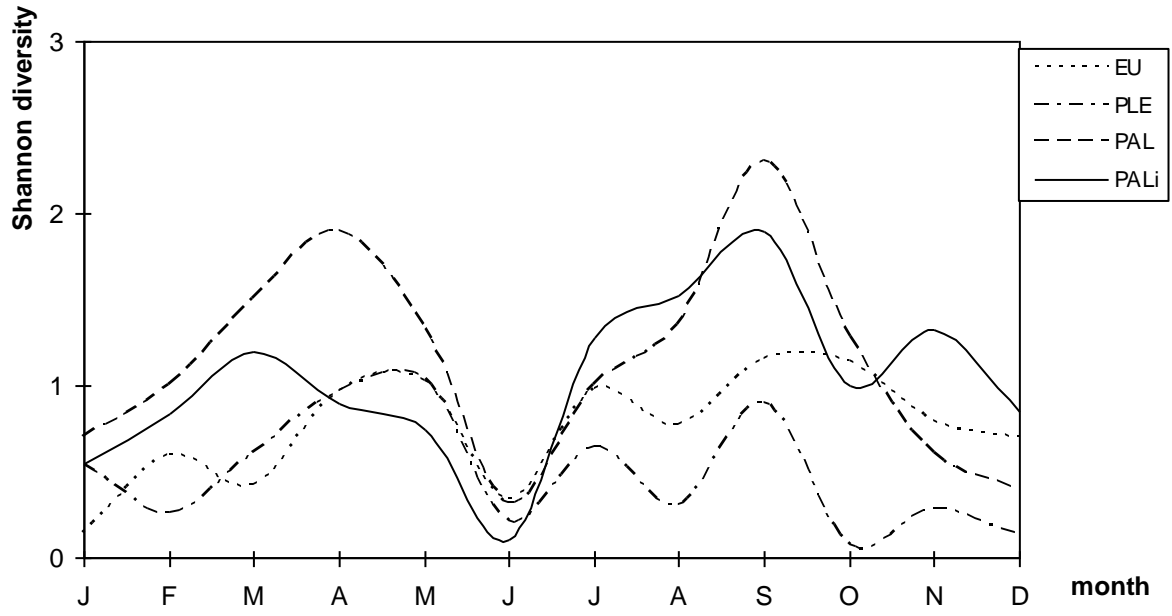
399

400 Fig. 3. Redundancy Analysis (RDA) biplot of caddis larvae (relative abundance data) in four floodplain habitats (EU=
401 eupotamon, PLE= plesiopotamon, PAL= paleopotamon, PALi= paleopotamon isolated) over a lateral connectivity
402 gradient along the Lower Rhine in relation to season and environmental parameters. CONN: connectivity duration,
403 CURR: current velocity, LEVL: water level fluctuation, TEMP: water temperature, CHLA: chlorophyll-a, AQVE:
404 abundance of aquatic macrophytes, MAVE: abundance of marsh vegetation. For explanation of species codes see
405 table 2.

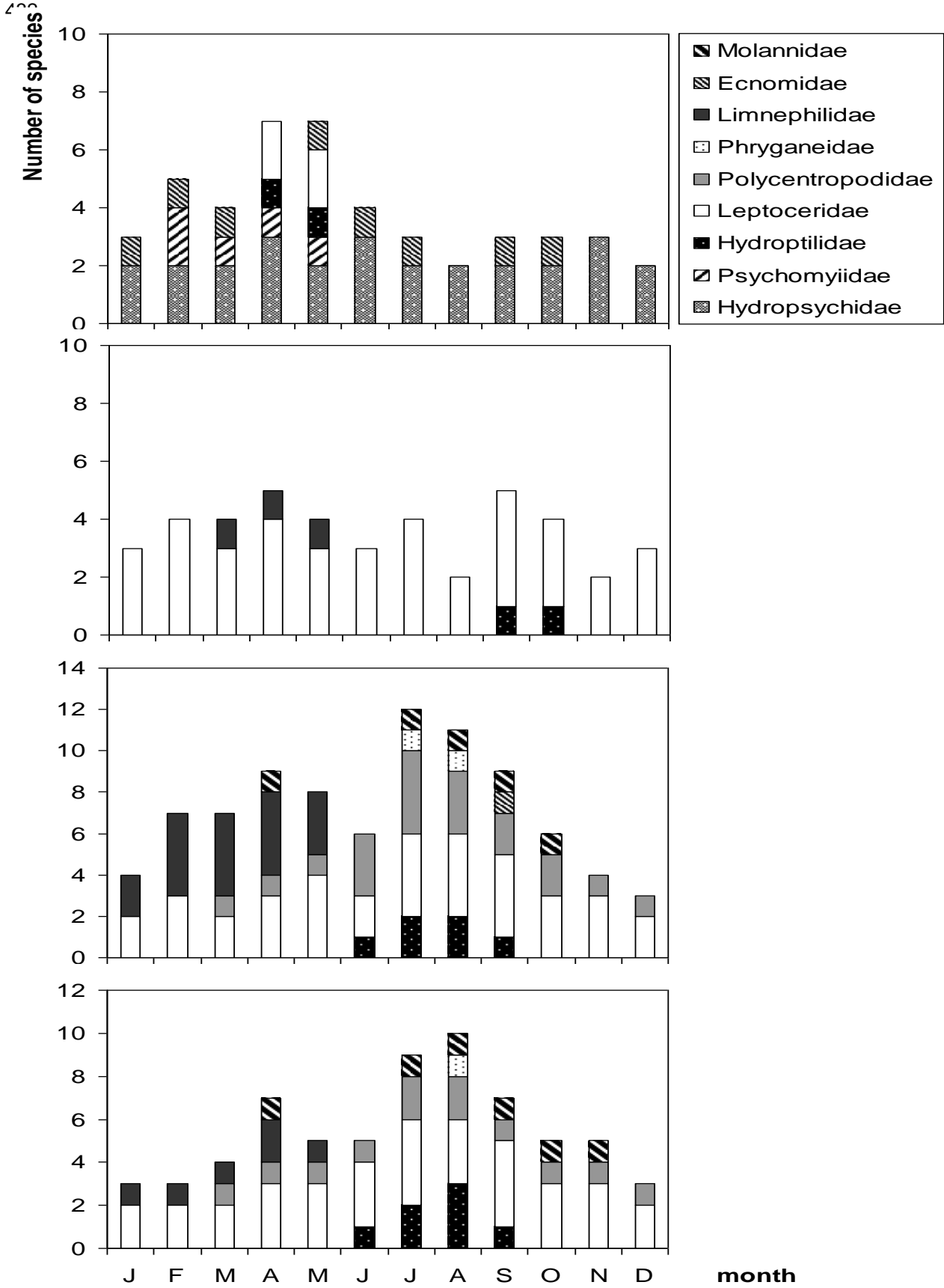
406

407 Fig. 1.

408
409
410
411
412
413
414
415
416
417
418
419
420
421
422
423
424
425
426
427
428
429
430



431 Fig. 2.



433
434

Fig. 3.

