



CHIRONOMUS

Newsletter on Chironomidae Research

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Chironomini indet. (*Paratendipes?*) from La Selva Biological Station, Costa Rica. Photo by Carlos de la Rosa.

CHIRONOMUS Newsletter on Chironomidae Research

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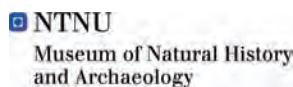
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Would you like to see your picture on the front page? Please send us your favourite midge photograph or drawing (torbjorn.ekrem@ntnu.no).

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Front page layout: Chironomid in title from photograph by Steve Marshall, Graphic design by Kolbjørn Skarpnes, NTNU Information Division.

Front page photo: Chironomini indet., possibly *Paratendipes*?
Photo by Carlos de la Rosa.

Editorial

Inventories — What are they good for?

Do we have the same interpretation of species? Do we use the same literature? Can we trust any chironomid species identification if it is not documented, either through description, reference literature or reference material? - In my opinion the answers are “no”.

We have all experienced that re-identification of a midge sometimes results in a species or species group different from the one the specimen originally was identified as. It could be our own original identification or it could be someone else's. The fact that many Chironomidae species are difficult to identify perhaps makes us experience this more often in our group, I don't know, but it is clearly a problem for static inventories and check-lists.

Moreover, many if not most identifications made in faunistic and ecological studies are unreferenced in publications. Thus, there is no possibility for the reader to know what literature was used, if reference specimens were examined, or even if the material has been deposited in a publicly accessible collection. How can the reader know if the author(s) used the most recent and geographically relevant taxonomic revisions and identification keys? It definitely makes a difference in the interpretation of a species identification (e.g. *Tanytarsus mendax*) if it is clear that the latest clarification of its taxonomy (Spies 1998) is used and not an older revision (Reiss & Fittkau 1971). So apart from giving credit where it belongs, citing the taxonomic literature is scientifically important and should be as mandatory as any other methodological reference.

The increased use of molecular markers in species identification, discovery and delineation (i.e. DNA barcoding) provides an additional set of objective characters for species in Chironomidae. The [Barcode of Life Data Systems](#) database (BOLD) currently holds almost 16 000 sequences of more than 1000 named species and offer a tool to unify the taxonomy and interpretation of chironomid species world-wide. Through our work with DNA barcoding of Chironomidae in Trondheim, we have discovered numerous cryptic species and molecular data has helped us understand species boundaries as well as phylogenetic relationships. There is no doubt that this is part of the future for chironomid taxonomy and thus also disciplines that rely on solid taxonomic foundations for identifications and analysis of species distributions and ecology.

To avoid the creation of useless inventories and erroneous check-lists we should all be careful in how we use and document our identification procedures. It doesn't matter if they are based on morphology or molecules. If the results are not made repeatable by proper description of the methods, it is not science. Therefore, cite your methods and references, and deposit your specimens in scientific collections!

Torbjørn Ekrem

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DR. WILLIAM P. COFFMAN: CELEBRATING 50 YEARS OF RESEARCH ON CHIRONOMIDAE

“Midges are an addiction. Can’t get through the day without my midge fix.” WPC

Bill Coffman’s addiction to midges has been a revelation and an inspiration to his students and colleagues for many years. His knowledge of the family, his uncanny ability to differentiate species by the pupal exuviae, and his enthusiasm over the range of wonders, from tiny structures to biogeographical patterns, is well known and respected. From humble beginnings at Thiel College, Pennsylvania (where he first met his wife of over 40 years), to a long and fruitful career at the University of Pittsburgh, Bill Coffman exemplifies the focus and the dedication that few scientists have for their subjects. He trained with the best, including Dr. John Stall at Thiel, Dr. Ken Cummins at Pitt and followed the steps of giants in the field through post-docs at a government laboratory in Baden-Württemberg, Germany. He also spent time at the Max Plank Institute in Plön, Germany, where names such as Thienemann, Lenz and others adorn not only the halls of the institution but the taxonomic literature as well. He has been honored with his own chironomid genus (*Coffmania*) and a species (*Robackia coffmanii*), immortalizing his name for generations to come.



Dr. William P. Coffman, University of Pittsburgh, Pennsylvania, September 2012.

His work has delved into methodologies and defining characters, through ecology and biogeography, taxonomy and evolution. His insights into the Chironomidae family species richness and diversity, evolutionary patterns, extinction rates, biogeographical puzzles and adaptation have been profound and are likely to shape the directions in this field of research in the future. His collection of world-wide pupal exuviae is unparalleled and extensive, with over 1,000 undescribed species, many from the Neotropics. But perhaps his greatest contribution so far is a family of scientists and researchers that have been touched, bitten perhaps, by the “bug” of knowing and using this family to resolve long-standing questions of evolution, biogeography, taxonomy and conservation.

Bill Coffman’s collection, now housed at the La Selva Biological Station of the Organization for Tropical Studies in Costa Rica, is the foundation for the development of species-level fine tools for bioassessment and ecology, with practical applications to habitat valuation and conservation. His undescribed species, keys and insights, captured in exquisitely prepared specimens (over 100,000 in the collection) and in his detailed notes, sketches and illustrations, will continue to train students in the fine points of chironomid taxonomy and applications. His name will continue to appear in the chironomid literature for many years after he is gone.

“Chironomids are like the bacteria of the aquatic systems” WPC

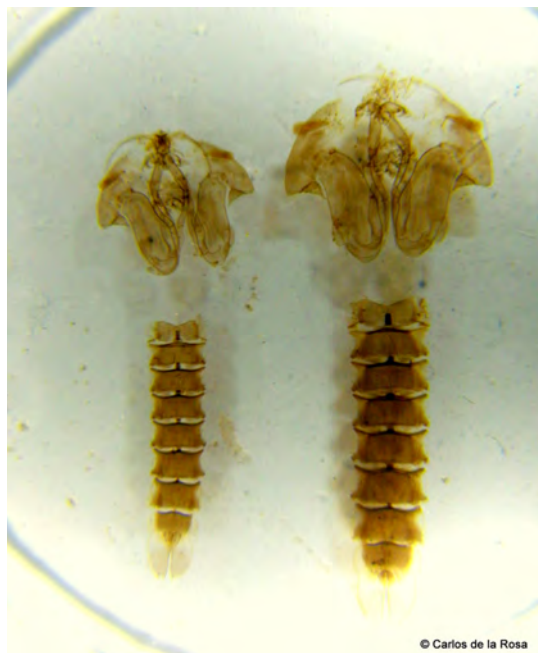
Bill Coffman thought of chironomids as the bacteria of the aquatic systems, the most diverse, species rich and ubiquitous organism in the wide range of aquatic habitats, from the Arctic Circle to the deep tropics. Midges occupy habitats unsuitable to most other organisms, able to thrive from thermal waters to ice-packed lakes; they fulfill every trophic role in aquatic ecosystems, playing the part of predators, herbivores, detritivores, filter feeders, parasites and specialists, feeding on wood, diatoms and other insects. They have an extraordinary range of life habits and an uncanny consistency of life history, which allowed specialists like Bill to learn from them how streams and lakes function. They are a family-level model of entire communities, offering species-level definition that allows us to ask questions rarely possible with other groups. In Bill’s words, they are “*the middle man in aquatic*



William Coffman collecting aquatic insects in the Murg River, northern Black Forest near Rogstag, Germany, 1968



Journal and first collection of chironomids by William Coffman, Winona Lake, Indiana, 1961.



Alotanypus sp. pupal exuviae mounted by William Coffman.

ecosystems,” supporting and maintaining the energy flow of most freshwater ecosystems. And their diversity is barely known in much of the world. Thousands of species remain to be discovered and described, up to 20,000 estimated around the world, including perhaps the most puzzling and difficult fauna of all: terrestrial chironomids. In spite of the challenges, the accumulated knowledge that a handful of specialists like Bill Coffman have bequeathed us will continue to build through the continued work on his and other collections. It is only unfortunate that chironomid taxonomy, as a field of study, has not been pursued or supported by most world universities or institutions. Very few true specialists continue to work on the family and the new generation is not coming up in the needed numbers, at a time when the scientific world needs them the most. The current wave of species extinction, mostly through habitat destruction and compounded by climate change, will make the techniques and knowledge application of this fauna more important than ever.

A new beginning for tropical chironomids

The relocation of Bill Coffman’s collections to Costa Rica, a country well known for its respect and love for biological diversity, will ensure a long life and a continued use of this amazing resource. The new *William P. Coffman Laboratory of Aquatic Entomology* at the La Selva Biological Station will make the collection available to scientists and students, to continue to develop it and add to it, explore it, publish its secrets and train a new generation of local ecologists and taxonomists. His unique collections from the Afrotropical Region, India and other locations around the world will be made available for regional scientists to study and build upon. The most extensive Nearctic collection of chironomid pupal exuviae, an important part of the latest version of the Nearctic Keys to the family, will be housed and curated here. Workshops on the use of chironomid pupal exuviae as tools in biodiversity surveys, bioassessments, pollution studies and habitat conservation, will truly revolutionize the way we approach river and stream conservation in the Neotropics. Printed and electronic keys will make use of the latest visualization technologies, accelerating the learning process for these new chironomid scientists. Other technologies, such as genetic barcoding, will help elucidate and resolve long standing taxonomic issues, all based on the strong foundation provided by many years of experience in anatomy and morphology. Future techniques, such as computer-assisted pattern recognition, will eventually allow students and field biologists recognize species much like we

currently use them to recognize fingerprints and faces. And its applications to conservation, pollution monitoring and control and priority setting for conservation and management, are just some of the many upcoming benefits to society.

“When a door opens, and you are young, you go for it” WPC

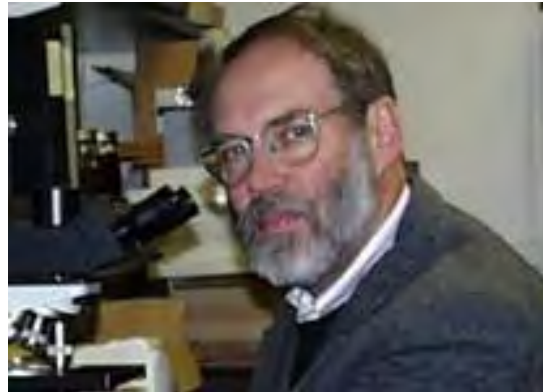
During a celebration of Bill’s career at the Department of Biology of the University of Pittsburgh, he recounted his early experiences in biology, as an undergraduate in Thiel College. His introduction to chironomids, like for most of us that have followed similar paths, was in part fortuitous, and in part absorbing. So many species and so many forms! How could this have evolved? He spent four summers collecting and sorting, trying to overcome the dearth in literature and the resistance from other biologists to study this difficult group. Even when redirected as a student of Ken Cummins in Pittsburgh to develop keys to all the aquatic insects of Linesville Creek, he continued to “*chip away at the resistance to name*” these many species, to figure out how can there be so many of them in a single stream. His doggedness paid off, inspiring a dozen students and challenging established notions. And this legacy will go on, through his papers, his extraordinary collection, his notes and drawings and his insights. His legacy will continue to open doors for those few curious students, filled with energy and frustration and deep and unfulfilled desire to learn and figure out the roots of this diversity and apply it to today’s biodiversity challenges.

A Passionate Scientist, a Scholar and a Teacher

Bill Coffman is, most of all, a well-rounded scientist, dedicated to his field with a quiet but intense passion, the same passion and attention to detail



William Coffman introducing his daughter Kendal to Chironomidae, 1969.



Dr. William P. Coffman at the University of Pittsburgh, Pennsylvania.

he applied to his personal life. For many years he collected stamps with a depth of knowledge and thoroughness only comparable to his knowledge of the midges. He studied history with a critical and discriminating eye, amassing an extraordinary library that he voraciously consumed and internalized. His expertise in World War II, in tropical exploration and history is hardly known by anyone outside his near family circle. He lived a relatively solitary professional life, teaching courses at Pitt (including Introductory Biology) that inspired those that cared to listen with his understanding and craving for knowledge. Largely lost in a young generation, those that did listen were inspired and moved, as the many testimonials from students attest. Bill was not one to go to many professional meetings, nor did he pursue research grants or feel that needed them. His tools were simple: two microscopes, a drawing tube, his notebooks, and few hand tools. His biggest investment was time: time to seek, see, compare, discriminate and resolve. After nearly 50 years of looking at millions of chironomid exuviae with a single-minded and hyper-trained eye, he broke through the extraordinary fog of species and resolved the characters and the keys, fine-tuning the pieces of the puzzle that have stumped researchers before him for over 100 years. Much like deciphering a hieroglyph, the tiny exuviae released their secrets to him, which he captured and preserved in his slides and notes, in his impromptu lectures to a fortunate few, in his field observations and his deep biogeographical and taxonomic insights. This is one of William Coffman’s most important scientific legacies.

However, his most important legacy has been the gift of his friendship, his mentorship, his love for science and his family, his relentless pursuit of knowledge and his deep human qualities. Many people benefitted personally and professionally from their association with Bill Coffman. He developed long lasting relationships with people like

Sam Roback, the Dean of North American chironomid workers, and Ken Cummins and his wife Peggy Wilzbach, who collected and shared many specimens from their trips around the world. Scientists like Jim Sedell, Norman Anderson, Henri Laville, Jan Sykora and former students like Rick Jacobson, Lou Yurasits, Len Ferrington, Dave Wartinbee, Bill Walker, Roger Carrillo and I (among others) benefited and made careers from our close relationship with Bill, providing specimens, insights, writing papers and developing theories that together represent the accumulated knowledge of a productive career. Our gratitude cannot be expressed with simple words, but it is immortalized by our joint accomplishments, the papers we wrote, the courses we taught, the species we discovered and the memories we shared. Thank you, Bill, for all that you have done for all of us.

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DEAR SEPP!

We miss your activity, your scientific leadership and your encouraging laughter.

Much has been said about your numerous merits, in the congratulations to your 75th and 80th birthday in *Chironomus Newsletter*^{1,2}, in the celebration of the award of the Meigen-medal³, and on the occasion of your retirement as director of the ZSM⁴.

Only one honourable act is missing from these accounts: the opening of „Sammlung Fittkau“ 2011 in the Museum of Ethnology in Munich, a large collection of different South American artifacts of mainly Amazonas tribes, showing the outcome of your and your dear wife's friendship with those people. One newspaper declared the collection as „the second part of your scientific life“. I congratulated you at the time for stepping on the road to immortality. Well, you are still immortal in this sense.

I remember an early visit to Plön, where to a special room of family Fittkau the entrance for visitors was strongly forbidden, because of the danger that one would not return alive as a consequence of the many poisoned arrows etc. in the room.

We would not like to repeat all your merits here. But we thought that those who were closely connected with you, should report on their most memorable reminiscences of you, so to speak „The best of Sepp“.

Wolfgang

Wolfgang Wülker:

In 1968, you and I were together at the International Congress of Entomology in Moscow. Both of us were selected to take part in a post-congress excursion to Borok on the river Volga. Our Russian colleagues still acted with some reservation towards the ugly Germans in Moscow, but in Borok they were thawing remarkably and we had nice evenings with various local families. We both were directed to stay in a small house together with a nice young lady as our supervisor (most probably the daughter of a mighty Russian). In the beginning, she wanted to take a bath and was not sure whether or not this was ok in the presence of two wild Germans in the same house. I never forget you saying „we are very shy persons“ – which indeed we were - and so the bathing could go on!

Elisabeth Stur:

Sepp became my 'Doktorvater' in 1993, right after he retired as director of the ZSM. He had been persuaded by Ulrike Nolte to take one 'last' PhD-student to work with chironomids in Brazil. – Well, as it turned out, I was not his last PhD student, – he could not say “no” to supervise the remaining 'last students', my Brazilian friends and colleagues Angela, Sofia and Maria. Sepp and his beloved wife Elise has been part of my life since then, and I am very grateful for all their support.



Elise and Sepp at the train station in Icking, Germany, July 2011. Photo: Angela Sanseverino.



Joachim Illies and Ernst Josef Fittkau on their way to collect at the Wasserkuppe, 1948. Photo: Family archive.

I have many fond memories of Sepp. As supervisor he was very patient in explaining morphological structures and teaching how to draw, and he was always encouraging in the writing process. However, his care for students and colleagues beyond the professional activity was perhaps even more remarkable – no matter what background the person might have.

One memory I have is from a warm sunny February-day in Munich when I could convince him to leave the office for the benefit of some hours in the nearby Biergarten. It was such a fine day and we had a very joyful conversation of bygone days; including stories from his time with Thienemann, Brundin and many other chironomologists whom I only knew from literature. I still remember this day with affection.

Angela Sanseverino:

„The lines are too straight; your drawing needs more movement and life.“ It was a spring morning in 2000, and with those words Sepp introduced me the art of taxonomy. Immediately he took his pen and ink, and showed me how to give to a seta a

simple and elegant movement. Sepp was always filling our lives with his enthusiasm and friendship. And of course, making us laugh with his brilliant stories, which are so well known by those who had the privilege to have been part of his life. I met Sepp for the first time in 1994, during the first Brazilian Chironomidae meeting. At that time I was a biology undergraduate student and after some minutes talking to him, I knew for sure that in the future I would like to do my doctoral dissertation under his supervision. It took five years more until I could finally be ready to start my PhD in Munich. Sepp generously accepted to supervise me, even though it would cost hours of his time, which he was aiming to spend entirely with his wife Elise and family after so many years dedicated to science and research. He was not only a supervisor, but a mentor and a dear friend who had always a kind word and smile and a house with opened doors to his students. Elise and Sepp have always welcomed me with open arms to their home. Like the warm welcome they gave me and my family last year in July, when we visited them in Icking after the Chironomid Symposium in Trondheim. We had a very nice conversation about family, friends, old stories and the future. On May 18, 2012 I came

to Icking to say goodbye to Sepp. It was also a spring morning. We did not exchange words, it was not necessary. Some flies, bees and butterflies were flying around. A light wind blew making flowers and trees move exactly the way Sepp once intended to explain to me.

Sofia Wiedenbrug:

I was lucky to be Sepp's PhD student and there are many stories that I could tell. He impressed me quite a lot right from the beginning! I had brought many samples from Brazil to Germany, but in Munich I realized that this material was insufficient to work on my PhD and I was so desolated about that! So, Sepp told me, he was invited to the "I Encontro Brasileiro sobre Chironomidae" in Rio and that I should attend this meeting, too. He offered himself to go with me to south Brazil in order to do some sampling after the Congress. At that

time, I was not an experienced driver so he himself drove the car from Rio de Janeiro to Porto Alegre which, by the way, is a 1.500 km trip, teaching me all the way down to the south a lot about the fauna and flora of my own country. He was very pleasant company and a gentleman!

He is still present in my mind while I'm working, I can hear his comments about the drawings and the importance of being able to see and interpret difficult structures. I miss him as a teacher and I miss him as a friend. Thank you, Sepp.

Maria C Messias:

I was introduced to Sepp by my adviser Sebastião de Oliveira, in Instituto Oswaldo Cruz, Rio de Janeiro, 1995. Both had met after a period of 20 years, when Sepp and Oliveira celebrated this occasion for the peer-reviewing of my Ph.D. thesis proposal to Instituto Oswaldo Cruz and Zoolog-



Young family in Manaus, 1963. Photo: Family archive.

che Staatssammlung, Germany. Later, several times I heard Sepp encourage me to advance the research work telling me „go on, Maria, the work is going ok, a big master does not fall from heaven“. What I learned from him was not only on Chironomidae, but personal values such as happiness of living, to share and celebrate friendship. A master might not fall from heaven, but a master like Sepp stimulated us to grow with him, to dream and work hard, so that we can achieve our dreams, heaven and perfection.

Martin Spies:

In my early 1980s undergraduate years – long before I succumbed to the siren song of the midges (you never ordered or coaxed me into this group) – on the way to a field course you told me some things about your work on chironomids. Suddenly you burst into a desperate cry (like I never heard from you again): “That [personal name omitted here] is destroying my [taxon name omitted here] systematics!” In the years to follow, you proceeded to collaborate closely with that same person to produce several standard references for everyone studying the taxon in question. You tried hard not to let differences in professional opinion affect personal relations, and in the interests of the latter you were ready to make compromises in the former.

In your words of gratitude upon being awarded the Meigen medal⁵, you wrote: “It has been a particularly important aim for me to coordinate chironomid research internationally and to provide training for junior colleagues.” I feel fortunate to have watched you do both, and to be among those aware of the resulting benefits we keep enjoying. You have succeeded, Sepp, and – in contrast to some elbow societies in other disciplines – our community has been the better for that work of yours.

When someone approached you with an idea for something to study, you often responded with a broad smile and your jovial “Machen Sie mal!” (the closest translation, unfortunately, has become a copyrighted slogan for a global sportswear seller). I cannot thank you enough for all your optimistic encouragement, infinitely patient guidance, and unselfish help.

If there is any place where we may meet again, I’m positive that you’ll receive us there with new energy and enthusiasm, and share with us, immediately after our arrival, what you’ve discovered in this next new world of wonder.

Roland Gerstmeier:

In 1980 Sepp gave me the opportunity to prepare my doctoral thesis at the ZSM. I really appreciated the topic – especially when I was in a small boat on large Starnberger See, hauling an Ekman-Birge sampler with profundal benthos and substrate for up to 120 m from bottom to surface. Much deeper still, however, was my gratitude for the total freedom that Sepp offered me and all his students.

I greatly enjoyed coffee breaks at the museum with him and Frieder Reiss, when the two recounted their adventures in South America – stories about indigenous people in Amazonia, for example Sepp’s affiliation with the Canela tribe, or about struggles with flight attendants when he brought whole spears, blowpipes or canoes into the cabin. One evening in a hotel room shared with Frieder, Sepp intentionally broke a giant traditional, old and precious clay pot. When Frieder cried out why he did this, Sepp’s answer was: “How else should I bring this on the airplane? I can glue the pieces back together in Munich” (which he did). In 1983, at the age of 56, Sepp travelled to Peru with some considerably younger companions. After a 45-minute drive from Lima to the high Andes (4800 m), Sepp jumped out of the car and galloped across the slopes like a mountain goat, while his fellow travelers suffered from bad headache and lethargy.

Sepp has opened my way and my senses to many things – he will always be in my mind and in my heart.

Claus Orendt:

I remember Sepp as a master of „very dry humour“ or stories, which seemed unbelievable to students of today. One example of each:

When we were on our first field campaign for my diploma thesis, Sepp accompanied me on our boat at Lake Chiemsee hauling some corers for sediment samples on a beautiful and warm day of spring. Everything was great: the weather, the sampling site and landscape, and in the background the gorgeous scenery of the nearer mountains of the Alps. We all were really impressed by this paradise-like surrounding, and when we thankfully realized and expressed this with words at the same moment, he concluded: „Really, you should pay an extra charge to be allowed to do this work here.“

The other story happened even earlier, when we were on excursion to study aquatic invertebrates in a student’s course. The weather was cold and rainy, and none of us were very motivated to stay

longer in the nature. We'd rather go home and study the animals collected the day before or alcohol-preserved material in the warm lab. In this moment, when he noticed our severe decline in motivation, Sepp started to tell stories of tough excursion conditions from times when he was a student and a young researcher. During one cold excursion they were happy to find a (cold) spring, in which they were able to warm up their hands... However, Sepp overestimated the pedagogical use of his description. We weren't just impressed, but also shivered, and not convinced to get a grip on ourselves.

In his job, Sepp had only very restricted time or space for personal things. For us as students and a very long time after the examinations, he was „Professor Fittkau“ or simply „Fittkau“. But later, he dearly tried to compensate for this and realized that he had neglected something which should be brought in order. One of the nicest moments for me was - I think it was at his 70th birthday party taking place at the Zoologische Staatssammlung Munich –, when he came to me (and others), at a certain time, clapped friendly on my shoulder, and declared with a bright and very happy smile: „I told the others [i.e. students, and perhaps some staff from the museum], that I am ‚Sepp‘“. That came deeply from his heart and showed his sense of responsibility for a familiar atmosphere. This was greatly appreciated.

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DR. MARTA MARGREITER - KOWNACKA (1939 – 2012)



Dr. Marta Margreiter - Kownacka (1939-2012)

Marta Margreiter–Kownacka was born on 14 September 1939 in Kraków into the family of the renowned Polish archaeologist, Professor Stanislaus Buratyński. After graduating from high school in 1956 Marta was accepted into the Faculty of Biology and Earth Sciences at the Jagiellonian University in Krakow to study biology. In October 1961 Marta was employed by the Department of Water Biology, Polish Academy of Sciences and embarked on a decade of important research. She joined the team that was conducting a comprehensive study of high-mountain streams, lakes and small reservoirs, mainly of the Tatra mountains, and comparing the results with other mountain

ranges in Europe. In 1968, Marta defended her thesis “Bottom animals of the Sucha Woda stream (High Tatars) on an annual basis,” and was awarded her PhD. The results of this period of research she published together with her first husband, Andrew Kownacki. In the same time, together we started research on alpine Chironomidae, describing several new species for science of the genus *Diamesa*.

In 1972 Marta gained a scholarship at the University of Innsbruck. An injury while skiing resulted in her long-term treatment in Austria, which ultimately became her home. From 1973 until 1986 she was a contract researcher at the University of Innsbruck, and in 1986, at the Austrian Academy of Sciences in Vienna. When the contracts expired, Marta established her own Institut für Ökometrie (Institute for Ecometrics) in Vienna, where she carried out research on the impact of hydrodevelopment on alpine streams for both government and commercial organizations. Marta met her second husband, Gerhard Margreiter in Austria. He died on 7 March 2011 in Vienna.

Marta published more than 50 scientific papers and expert opinions and participated actively in many scientific congresses and conferences. She was honoured with the naming of a new species of fly (Chironomidae, Diptera) in her name–*Diamesa marta*. Her biography is published in “The Great Encyclopedia of the Tatra Mts” (Paryska, Paryski, 1995).

Andrzej Kownacki

CURRENT RESEARCH

CHIRONOMIDAE (DIPTERA) IN THE HIMALAYAN LAKES – A STUDY OF SUB-FOSSIL ASSEMBLAGES IN THE SEDIMENTS OF TWO HIGH ALTITUDE LAKES FROM NEPAL

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Abstract

Chironomid head capsules were identified from sediment cores taken from Lake Gokyo (4750 m) and Lake Gosaikunda (4350 m) in the east-central Himalaya, to determine taxonomic composition of chironomid assemblages over time. The sedimentation rate of Lake Gosaikunda was estimated at 0.05 cm/yr and that of Gokyo was estimated at 0.07 cm/yr by ²¹⁰Pb and ¹³⁷Cs dating. *Micropsectra* sp. was dominant in the sediments of both lakes. Lake Gokyo sediments also contained *Pseudodiamesa* sp., *Eukiefferiella/Tvetenia* sp., *Orthocladus/Cricotopus* sp. and *Rheocricotopus* sp. The concentration of headcapsules was nearly an order of magnitude greater in the Lake Gosaikunda core, which contained mostly *Micropsectra* sp. and *Pseudodiamesa* sp. These taxa are typical of cold oligotrophic lakes. Differences in lake depth, dissolved minerals, plus epi- and hypolimnetic temperature and dissolved oxygen suggest that stratification and temperature-induced increases in primary production may affect chironomid assemblages in these pristine lakes. Palaeolimnological studies of Himalayan lakes should include replicate cores within lake, to increase headcapsule sample sizes given potentially high rates of sedimentation from glacial runoff.

Introduction

High-altitude lakes are considered to be very sensitive to climate change (Parry et al. 2007). Climate change is an especially pressing environmental issue for a country like Nepal with its location in the Central Himalaya. The normal seasonal pattern of monsoon rainfall appears to be changing, but there is little data on variation in the form and amount of precipitation across the Himalaya, and consequently much uncertainty about the fate of

Himalayan glaciers in the face of climate warming (Bolch et al. 2012). Shrinkage of many of these glaciers affects the discharge of rivers and lakes in the region. Palaeolimnological techniques present the possibility to infer change in environmental conditions over time, such as lake water temperature (Walker et al. 1991) using sub-fossil chironomid head capsules preserved in lake sediments. However, climate reconstruction modeling requires a calibration dataset based on samples of chironomid head capsules from a large number of lakes spanning a gradient of average temperature. The strong association between lake temperature and lentic chironomid distribution has been well-established by numerous calibration datasets from Western Europe and the Nearctic (Eggermont and Heiri 2011). A calibration dataset for Nepalese lakes has not yet been constructed: this study represents a necessary first step towards establishing the relationship between chironomid assemblages and temperature for Himalayan lakes by sampling two remote very high altitude lakes. There is hardly any information available on chironomid assemblages and temperature for Himalayan lakes and this study is therefore a necessary first step, based on which several other high altitude lakes are planned to be studied in the future.

The chironomid fauna of the high altitude lakes in the Nepalese Himalaya in particular are poorly known. This is due to the physical difficulty in reaching these lakes, as well as government restrictions on access. Fortunately, most of the high altitude lakes in Nepal are protected by law and categorized as National Parks and Nature Reserves and thus have experienced relatively little direct human disturbance. Limnological studies in the Nepal Himalaya have focused on low altitude lakes (Bhandari 1993; Swar 1980) due to the vis-

ible impact of nutrient enrichment and eutrophication. In contrast, the water chemistry of high-elevation lakes is mainly determined by weathering of rocks (Tartari et al. 1998); similar high altitude lakes in Sikkim and Kashmir (Zutshi 1991; Khan and Zutshi 1980; Sharma and Pant 1979) exhibited very low concentrations of dissolved nutrients and minerals and also low phytoplankton abundance.

Previous studies of the chironomid fauna of nine Himalayan lakes, located at altitudes from 4830 m to 5580 m, were conducted by Manca et al. (1998), Reiss (1968), Roback and Coffmann (1987), and Loeffler (1969). The first detailed study on morphology, bathymetry, physico-chemical, and biology of high altitude lakes in Nepal was provided by Lami et al. (1998), and included a palaeolimnological component. The World Wildlife Fund/Nepal (2010) reported the first survey of the Lake Gokyo Series, including bathymetrics, sediment-dating and a description of the chironomid fauna.

The aim of this study was to describe the sub-fossil chironomids assemblages in sediments from the deepest part of two high-elevation Himalayan lakes, in conjunction with temperature and oxygen measurements. These data establish the distribution of lentic chironomid taxa at the low-temperature/high elevation extreme and thus contribute to the construction of a calibration dataset specific to the Himalayan region.

Materials and Methods

Study sites

Two lakes in the main Himalayan range were studied: Gokyo (“Third Lake”) which lies in the eastern zone of Everest National Park, and Lake Gosaikunda, lying in the central zone of Langtang National Park (Figure 1, Table 1). Lake Gokyo is fed by drainage from Ngozumba glacier and springs from Renjo La Pass to the north-west. Although there are four other major lakes in the headwaters of Kosi River system, they have no surface-water connection to Lake Gokyo. Lake Gosaikunda is fed by snowmelt and by a perennial spring source to the north-east, considered a holy place by Hindus and Buddhists. There are many lakes in the series connecting Lake Gosaikunda with the River Trisuli in the Gandaki river system. The geology surrounding the lake consists mostly of exposed bedrock and glacier debris (Bortolami 1998). The lake bottom is composed of silt-clay sediment in the deeper water and sand-silt in the littoral zone. The average minimum temperature in the Everest region of these high-altitude lakes is -7.7°C in January, and the average maximum temperature is 16.2°C in August (Tartari et al. 1998). Vegetation in the catchment is sparse but consists of juniper, rhododendron and herbs.

Bathymetric maps of the lakes were compiled using GIS software from sounding data collected using



Figure 1. Location map and photos of the study area (not in scale). Lake Gokyo (Mt. Everest National Park) is shown to the right and Lake Gosaikunda (Langtang National Park) to the left.

ing a GPS (Garmin Venture SC) in tandem with an Ecosounder model PLASTIMO ECHOTEST II (Sharma et al. 2011). Sediment samples were collected from the deepest part of the lakes (Figure 2) using a Uwitec gravity corer, Ø 60mm diameter (<http://www.uwitec.at>) operated from an inflatable boat. Sediment dating was performed by analysis of ^{210}Pb and ^{137}Cs isotopes at the Tibetan Plateau Research Institute of the Chinese Academy of Sciences in Beijing (WWF/Nepal 2010). Two core samples were collected from Gokyo Lake, and one from Gosaikunda Lake; cores were cut into 5 mm slices and transported in an ice box. Only the core 2 was actually dated from Lake Gokyo.

Chironomid head-capsule analysis

Although the chitin head capsule of chironomid larvae may remain preserved in lake sediments for

Table 1. Parameters of Lake Gokyo, sampled May 2009, and Lake Gosaikunda, sampled October 2010 (Aquatic Ecology Centre, Kathmandu University, personal communication).

Name of lake	Gokyo	Gosaikunda
Coordinates	86°41'N 27°57' E	28°5'N 85°25' E
Elevation	4750 m	4426 m
Surface area	42.9 ha	13.8 ha
Max. depth	42.0 m	24.0 m
Temp (top/bottom)	8.0/4.5 °C	9.2/7.5 °C
DO (top/bottom)	8.3/6.3 mg/L	6.1/3.6 mg/L
Na	0.37 mg/L	1.93 mg/L
K	0.26 mg/L	1.80 mg/L
Fe	0.78 mg/L	0.80 mg/L

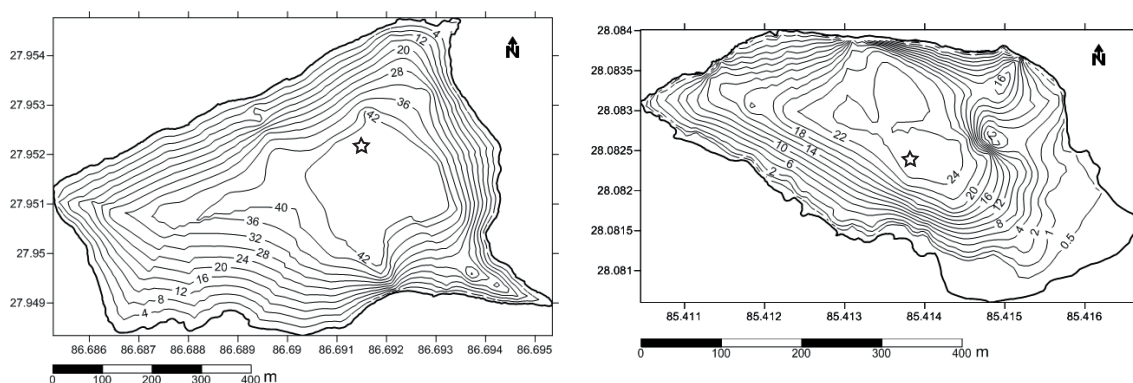


Figure 2. Bathymetric map of Lake Gokyo (left) and Lake Gosaikunda (right). Stars indicate sites where the sediment cores were taken.

thousands of years, they are usually lacking delicate morphological features; yet they can be identified to genus level, or at least grouped into consistent morphotypes. Extraction of head capsules from sediment samples followed the methods of Walker (2001) for air dried sediment samples, but with slight modifications. Samples were deflocculated in 10% KOH overnight (up to 18 hrs) without heating. The sediment was then passed through a 100µm mesh sieve and chironomid head capsules were removed by a fine probe under a stereomicroscope at 40×. After dehydration in 95% ethanol, head capsules were slide-mounted in Euparal. Identifications were made at 400×, based on the keys of Wiederholm (1983), Roback and Coffmann (1987), Walker (2007), and Brooks et al. (2007). Head capsules with a complete mentum or two corresponding halves were counted as one individual. Slides are retained at the Aquatic Ecology Centre of the School of Science, Kathmandu University.

Results

In Lake Gosaikunda, the number of head capsules per sub-sample within the core ranged from 62 to 175, whereas in Lake Gokyo the concentration of head capsules was lower, from 7 to 41 per sub-sample of core 1 and from 8 to 30 for core 2 (Figure 3). In Lake Gokyo, numbers decreased from the sediment surface to 4 to 5 cm depth; density then started to increase towards 6 cm depth, and then decreased at 15 to 10 cm depth in core 2. The natural sedimentation rate in Lake Gokyo was estimated at 0.07 cm, and in Lake Gosaikunda as 0.05 cm per annum on average. Extrapolation of this value to the sediment cores suggests that cores from Lake Gokyo might represent a time span of approximately 140 years, and that of Lake Gosaikunda as 1000 years. However, a single core does not necessarily reflect sediment loading for the lake as a whole (Lami et al. 1998).

The sub-fossil chironomid assemblage in Lake Gokyo was dominated by *Micropsectra* sp. (Tanytarsini) and Orthoclaadiinae, with some *Pseudodia-*

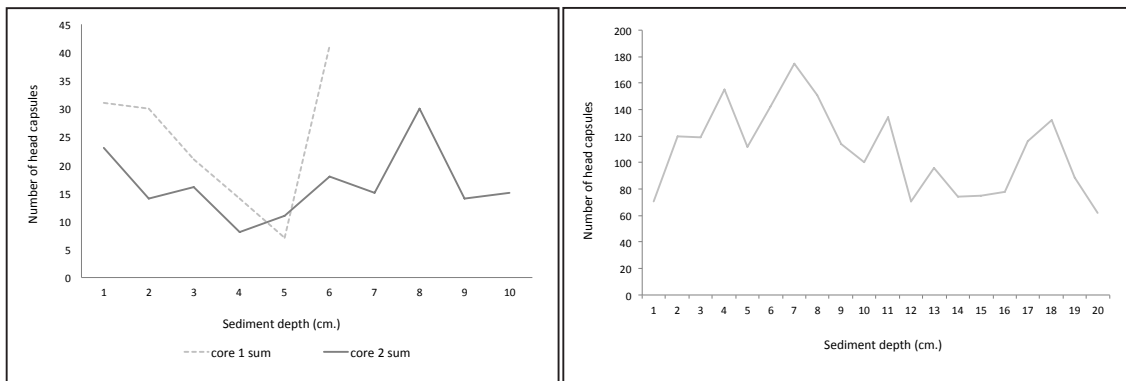


Figure 3. Head capsules count in two different cores from Lake Gokyo (left), and single core in Lake Gosaikunda (right).

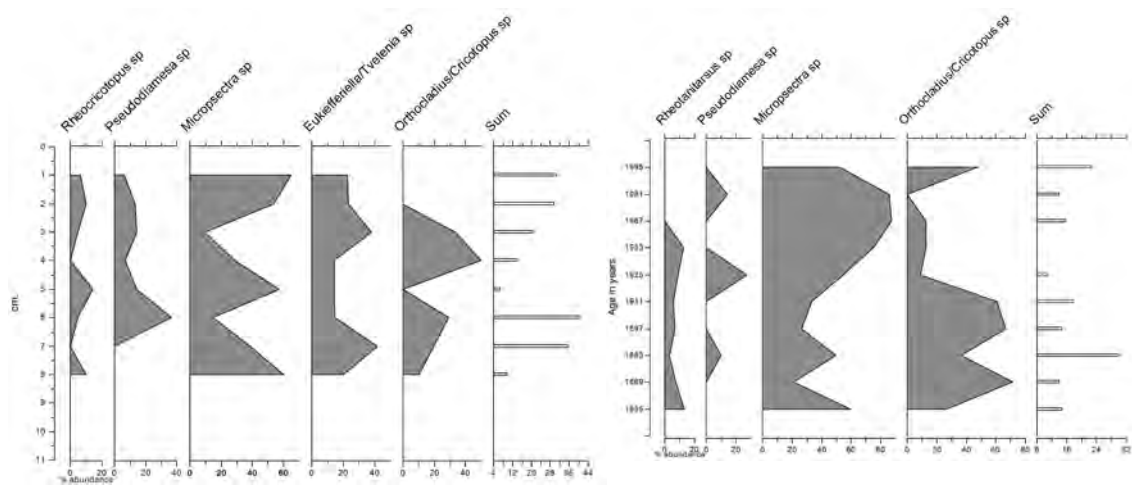


Figure 4. Chironomid stratigraphy of Lake Gokyo in core 1 (left), and core 2 (right). Chironomid taxa are given as % abundance; sum represents the total number of head capsules.

mesa (Diamesinae; Figure 4). The Orthoclaadiinae were represented by an *Orthocladus* / *Cricotopus* morphotype and an *Eukiefferiella* / *Tvetenia* morphotype. The remaining orthoclad, *Rheocricotopus* sp. was distinctive and was found in both of the Gokyo cores. In contrast, Lake Gosaikunda was dominated by *Microsectra* sp. followed by *Pseudodiamesa* sp. (Figure 5) with occasional occurrence of *Orthocladus* sp.

Discussion

The greatest difference in chironomid assemblages was seen in the concentration of head capsules in the sediment cores, which was almost an order of magnitude greater in Lake Gosaikunda (Figures 4, 5). This may be due to greater sedimentation rates in Lake Gokyo, which receives direct glacial meltwater. The trend of lower numbers of head capsules at shallower sediment layers might possibly reflect increased sedimentation from glaciers, which have generally been receding in the central and eastern Himalaya (Bolch et al. 2012). The three cores from these two lakes were more similar in terms of relative abundance of chironomid

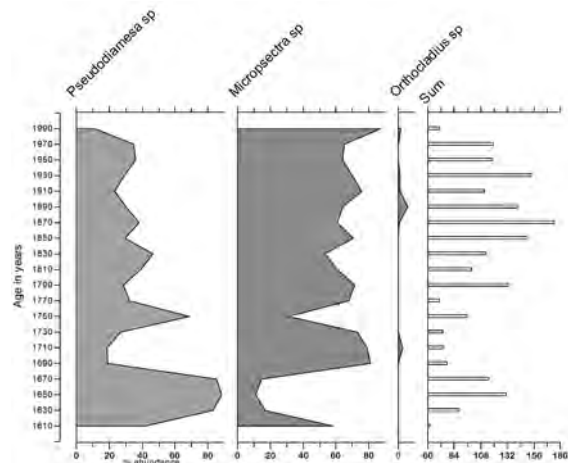


Figure 5. Chironomid stratigraphy of Lake Gosaikunda. Taxa are given as % abundance and sum represents the total number of head capsules.

taxa. The Sørensen distance (based on relative abundance data) between the two assemblages from Lake Gokyo was 34.5%, whereas the distances between the Lake Gosaikunda assemblage and those of Lake Gokyo cores 1 and 2 were slightly

larger at 47.4% and 41.0% respectively. Overall, *Micropsectra* sp. was dominant and composed up to half of the individuals per core in both lakes (Table 2). *Pseudodiamesa* was sub-dominant in Lake Gosaikunda (41%), whereas in Lake Gokyo *Orthocladius/Cricotopus* was subdominant.

In the Nepal Himalaya above 2000 m, Roback and Coffmann (1987) reported *Micropsectra* sp. as the dominant Chironominae. Manca et al. (1998) also found *Micropsectra* larvae in 27 lakes out of 28 studied, located between 4532 m and 5580 m in the Everest National Park. They believed that there

Pseudosmitta were also reported by Löffler (1969) in the region at lower altitudes between 2500-2600 m. *Rheocricotopus* found in both the cores from Lake Gokyo is also reported by Hamerlik et al. (2010) from Sharmar Tso Lake in Tibet.

Given that both lakes are fed by glacial meltwater and/or snowmelt in landscapes dominated by bare rock and glacial debris, it is likely that a combination of high sedimentation and low primary production rates explain the low abundance and taxonomic composition of chironomid head capsules in these lakes. Higher concentrations of head cap-

Table 2. Relative abundance (in %) of chironomid taxa recorded in the lake sediments of both the lakes.

Taxon	Lake Gokyo		Lake Gosaikunda
	core 1 (6 cm)	core 2 (10 cm)	(20 cm)
<i>Eukiefferiella/Tvetenia</i> sp.	25	0	0
<i>Micropsectra</i> sp.	38	53	58
<i>Orthocladius /Cricotopus</i> sp.	18	37	0
<i>Orthocladius</i> sp.	0	0	1
<i>Pseudodiamesa</i> sp.	14	5	41
<i>Rheocricotopus</i> sp.	5	5	0

were at least two forms of *Micropsectra* larvae in the region, the most widespread being similar to other undescribed species reported by Roback and Coffmann (1987). Hamerlik et al. (2010) reported *Micropsectra* sp. as the most abundant taxon in the Tibetan lakes they studied, and considered this to represent a species close to *Micropsectra nepalensis* Sawedal, as described in Roback and Coffmann (1987). Across the Holarctic, *Micropsectra* is typical of cold, unproductive lakes (Eggermont and Heiri 2011).

Pseudodiamesa is also a dependable indicator of cold temperature lakes in the Holarctic (Eggermont and Heiri 2011). *Pseudodiamesa* sp. in this study probably represent *Pseudodiamesa (Pachydiamesa) nepalensis* Reiss as reported by Reiss (1968) above 5000 m a.s.l. in the Nepal Himalaya. Manca et al. (1998) mentioned that in the Himalaya *P. nepalensis* probably replaces both *P. nivosa* (Goetghebuer), which is wide-spread in the palae-arctic region, and *P. branickii*, a Holarctic species found at 850-2500 m elevation, and at higher elevations in Central Asia and Nepal (Pagast, 1947; Sæther, 1968).

Manca et al. (1998) reported Orthoclaadiinae as fairly common and in the Everest region, and composed of three genera, of which the commonest is *Cricotopus*. However, samples of adult chironomids made during this survey contained *Orthocladius* sp. but not *Cricotopus* sp. *Acricotopus* and

sules obtained from Lake Gosaikunda may reflect the morphometry of Lake Gosaikunda which has a smaller surface area and is almost half as deep than Lake Gokyo (Table 1). Though Lake Gosaikunda was sampled in October (vs. May for Lake Gokyo) the surface water temperatures of the two lakes differed by little more than a degree; in contrast the bottom water of Lake Gosaikunda was three degrees warmer and contained only 57% as much dissolved oxygen compared to Lake Gokyo (Table 1). These data, together with higher concentrations of dissolved Sodium and Potassium from weathering, suggest that Lake Gosaikunda supported slightly higher productivity of chironomids, due to warmth and possibly autochthonous production. Lake depth, the degree of stratification and hypolimnetic oxygen deficit may relate to the differences in chironomid assemblages observed (Heiri et al. 2003; Velle et al. 2010).

The low and variable concentration of head capsules, especially in the Lake Gokyo cores, creates uncertainty in interpretation of the shifts in chironomid composition observed. This variation may reflect changes in silt load from glacial melt-water. In general, it is more difficult to infer recent temperature change, because variation in local environmental factors masks our ability to detect small temperature changes of only a couple of degrees (Velle et al. 2010, Velle et al. 2012, Eggermont & Heiri 2012). Lake morphometry influences variation in sediment cores, due to eco-

logical responses of chironomid taxa to depth and development of the littoral zone and stratification (Heiri et al. 2003). Chironomid calibration datasets are commonly based on single cores from the deepest part of the lake. A comparison of within-lake spatial variability in chironomid assemblages and its impact on temperature modeling found significant heterogeneity among cores from the deepest part of five lakes, as well bias due to change in assemblage structure in cores along littoral to profundal transects (Heiri et al. 2003). Change in lake trophic status may or may not be related to average temperature change, thus the cause of shifts in chironomid assemblage composition must be interpreted judiciously in temperature reconstruction (Velle et al. 2010).

Given the spatial variability of precipitation within the Himalaya (Bolch et al. 2012) as well as global warming, Lakes Gokyo and Gosaikunda provide valuable baseline data on subfossil chironomid assemblages of high-altitude, unproductive lakes. Given the large amount of variability in lake-specific factors such as morphometry, productivity and exposure of the catchment to persistent snowpacks, future sampling effort may best be directed towards increasing the number of lakes sampled and measuring additional temperature proxies from the cores, e.g. diatoms or pollen (Chase et al. 2008; Velle et al. 2010; Eggermont and Heiri 2012). Thus, across lakes, a clearer signal of regional temperature change may be inferred. Sampling design for construction of a Himalayan calibration dataset must also address covariation of temperature and trophic state: while the coldest, highest altitude lakes are least impacted by human activity, lakes at lower elevations are not only warmer, but are more likely to be impacted by anthropogenic nutrient enrichment. The remote location of Lakes Gokyo and Gosaikunda is a reminder that it can be difficult to quantify average annual air temperature appropriately; however the measurement of epi- and hypolimnion temperature and dissolved oxygen, as reported here, is valuable in modeling the relation of chironomid assemblages to lake temperature (Eggermont and Heiri 2012). Replicate within-lake cores will be vital for future palaeolimnological studies of those Himalayan lakes where sedimentation rates may be high and chironomid productivity is low. In the Himalaya, continued deposition of debris by receding glaciers is forming many new lakes (Bolch et al. 2012), which will provide an opportunity to observe both ontological changes on lake productivity as well as responses to climate change.

Acknowledgements

The GOKYO Project team deserves special thanks including staff at Aquatic Ecology Centre, Kathmandu University. We are thankful in particular to Dr. Chhatra Mani Sharma who helped in sampling and Ms. Sikha Karki who prepared the slides that helped in identification. Authors are also thankful to the Editor, Chironomus Newsletter, and the two anonymous reviewers, who reviewed the manuscript seriously, and that has substantially improved the quality of this paper. The research was supported in part by WWF/Nepal, Fulbright Commission/USA and OeAD/ Austria.

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NON-DESTRUCTIVE DNA EXTRACTION FROM CHIRONOMIDAE, INCLUDING OF FRAGILE PUPAL EXUVIAE, EXTENDS ANALYSABLE COLLECTIONS AND ENHANCES VOUCHERING

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Introduction

It is well established that the traditional taxonomy and nomenclature of Chironomidae relies on adult males whose usually characteristic genitalia provide evidence of species distinction. In the early days some names were based on female adults of variable distinctiveness – but females are difficult to identify (Ekrem *et al.* 2010) and many of these names remain dubious. In Russia especially, a system based on larval morphology grew in parallel to the conventional adult-based system. The systems became reconciled with the studies that underlay the production of the Holarctic generic keys to Chironomidae, commencing notably with the larval volume (Wiederholm, 1983). Ever since Thienemann's pioneering studies, it has been evident that the pupa, notably the cast skins (exuviae) provide a wealth of features that can aid in identification (e.g. Wiederholm, 1986). Furthermore, the pupae can be readily associated with name-bearing adults when a pharate ('cloaked') adult stage is visible within the pupa. Association of larvae with the name-bearing later stages has been much more difficult, time-consuming and fraught with risk of failure. Yet it is identification of the larval stage that is needed by most applied researchers due to the value of the immature stages of the family in aquatic monitoring for water quality, although the pupal stage also has advocates (reviewed by Sinclair & Gresens, 2008). Few use the adult stage for such purposes as their provenance and association with the water body can be verified only by emergence trapping, and sampling of adults lies outside regular aquatic monitoring protocols.

An answer to this mismatch in demand and taxonomic expertise may lie with the actual and potential use of molecular data. In the past two decades researchers have used data derived from DNA to estimate phylogenies for their inherent interest (e.g. Guryev *et al.* 2001), in supplementing morphological characters (Cranston *et al.* 2012), in estimation of support for biogeographic hypotheses (e.g. Krosch *et al.* 2009; Cranston *et al.* 2010) and

increasingly to propose identification of any or all stages (Carew *et al.* 2007; Ekrem *et al.* 2007). In the realm of understanding midge diversity and community structure, cryptic diversity has been detected (Carew *et al.* 2005) including by use specifically of a 'DNA barcoding' approach (Pfenninger 2007; Sinclair & Gresens 2008; Ekrem *et al.* 2010). Phylogeographic molecular genetic studies have been undertaken to reveal the origin and maintenance of chironomid populations (e.g. Martin *et al.* 2002; Krosch *et al.* 2009; Kaiser *et al.* 2010) and fine-scale patterns of gene flow among adjacent streams have been assessed (Krosch *et al.* 2011a). Early on, adult stages were used in such studies with immature stages incorporated in some. Hence, many techniques for DNA extraction from single chironomid specimens were developed specifically for adult bodies and, thus, relatively larger amounts of tissue (e.g. Willassen 1999; Wang & Wang 2012). Although molecular techniques have been used to associate the sexes of adults (Willassen 2005), explicit attempts made to associate previously unknown immature stages with adult stages have been few. Species associations were either 'known' from cultivation (e.g. *Chironomus*, Guryev *et al.* 2001) or by rearing in the field (e.g. *Archaeochlus* / *Austrochlus*, Cranston *et al.* 2002) or in the laboratory (Sinclair & Gresens 2008). Targetted 'taxonomic' associations of life histories solely via DNA have been often incidental and remain rare in chironomid studies.

In seeking to maximize numbers and/or diversity of taxa for large-scale molecular phylogenetic and population genetics studies, we sought the aquatic immature stages, using conventional techniques with aquatic nets and kicking substrates or interception of drifting individuals (Cranston *et al.* 2010, 2012; Krosch *et al.* 2011a, b). In over 2 decades of working intermittently with molecular aspects of Chironomidae, between us we believe we have encountered most problems, solved many, and have unpublished insights into what can go wrong, what works well, and what can be im-

proved in our protocols. Largely in response to requests from interested colleagues, we describe here the simple technique that makes possible retrieval of PCR-quality DNA from chironomids including even from single pupal exuviae. Previous attempts to extract DNA from exuviae using a modified salting out protocol (Miller *et al.* 1988) failed, and we assumed such specimens lacked sufficient remaining tissue post-adult emergence for DNA extraction. However with careful handling of specimens and more sensitive modern extraction techniques described below, good DNA can be recovered, thereby allowing access to greater numbers of immature stages, and avoiding the need for laborious and often unrewarding rearing, making molecular associations between life stages simpler.

Methods

Field

We use all traditional and some less usual sampling when collecting material in the field, according to the purpose. For population genetics studies we sample intensively in a narrowly defined area containing the appropriate microhabitat(s), seeking known target taxa. For phylogenetic studies in which we seek also to maximise diversity, we sample all visible microhabitats. At all lotic sites we place a drift net downstream of other collecting activity for the duration of the visit, or longer, including overnight if feasible, for pupal including exuvial sampling. In contrast to most studies, we sort specimens from the substrate at or close to the sampling location using dissector microscopes. Live sorting is preferable with specimens being picked with fine forceps and placed immediately into isopropanol or molecular grade ethanol, and never into formalin, methanol or ethanol that has in any way been 'denatured'. Only if a sample cannot be sorted 'live' in timely manner do we preserve in alcohol, after removal of larger particulate matter and sieving for medium-sized organic removal. Isopropanol, although more expensive, is widely available for use in molecular labs, preserves DNA very well, is less volatile and less flammable (important if flights are involved in field work) and is not hygroscopic – your 100% alcohol remains 'absolute'. Most failures to extract DNA from specimens are associated with the age and storage of older material (which should be kept cold and dark) or unwitting use of certain denatured ethanols as supplied to many 'wet labs'. We suspect that some total DNA destruction is caused by customs irradiation of international mailed material but cannot verify this.

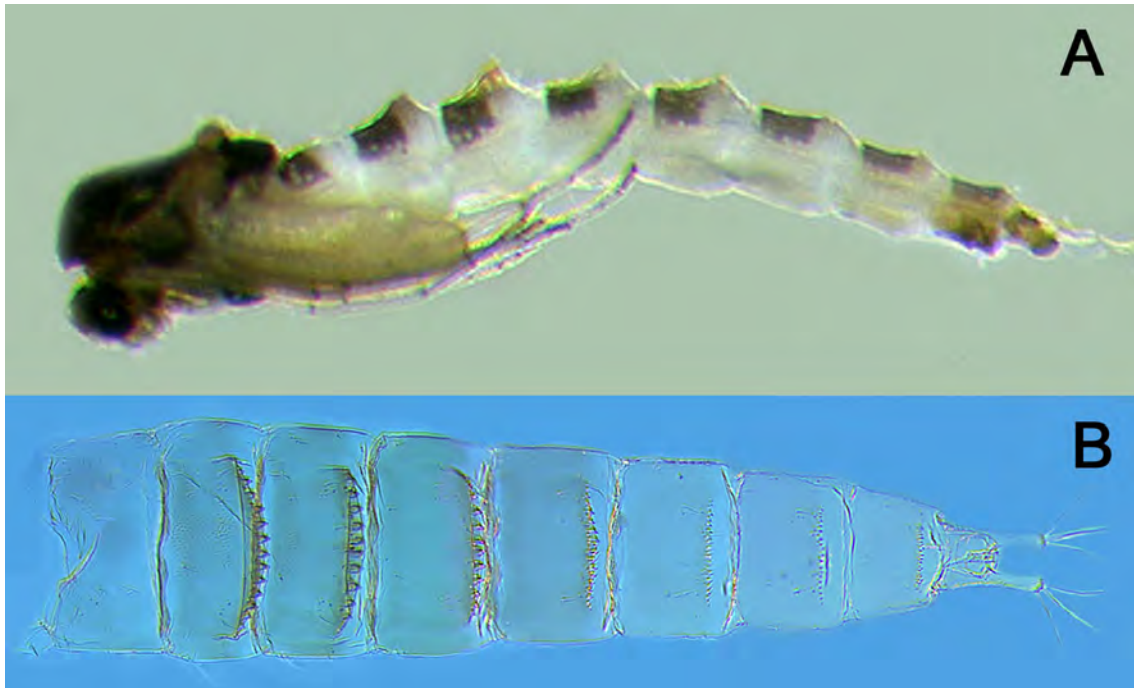
For some taxa, life history stage and location it

is possible to know the specimen identity prior to laboratory processing for molecular work. For instance, reared monoculture such as species of *Chironomus* may be assumed to be that species, and those familiar with the fauna of a particular area can identify much without slide preparation. However as we know, accurate identification using morphology requires slide preparation, which is *prima facie* incompatible with DNA extraction. For larvae we amputate the head and prepare it as a temporary slide mount (in Hoyers mountant) for rapid identification prior to decision on DNA extraction: the same applies to male genitalia. If sequenced, these vouchers are remounted in Euparal for permanence. For pupae, identification often can be made without prior slide preparation.

Laboratory

We extract total genomic DNA from specimens using a Qiagen DNeasy® Blood and Tissue Kit (Qiagen, Hilden, Germany) following manufacturer's protocol. Modifications to this protocol were as follows: (1) tissue was digested with proteinase K overnight at 37°C; (2) after digestion, cuticles were removed carefully using fine-tipped forceps and transferred to 90% isopropanol before vouchering as slides in Euparal; (3) the final elution volume was 100 µL to increase the final DNA concentration of the eluate; and (4) the incubation time for elution was increased to 5 minutes. Furthermore, digestions were never vortexed while cuticles remained in solution to avoid damage to diagnostic morphological characters. At all stages, care with handling is essential and we found using the solution's surface tension, rather than direct pressure from forceps, to be sufficient for transferring specimens between micro-centrifuge tubes. We anticipate that extraction kits from other manufacturers may produce similar results under the same modification, but this remains untested currently.

We report extraction success from pupal exuviae only here and do so simply in terms of reporting successful amplification and sequencing of the barcode region of the mitochondrial *COI* gene and the slower evolving nuclear 28S rRNA region. We acknowledge that using more stringent quality control measures (e.g. NanoDrop or BioAnalyzer technologies) may provide more information about actual DNA concentration and quality. However, we envisage that the majority of end-users of extracted DNA from pupal exuviae will be interested in utilising DNA for traditional PCR and Sanger sequencing and thus we believe our qualitative assessment valid in this case. Reactions were carried out in a total volume of 25 µL, containing 2-5 µL



A. Pharate adult pupa of *Stictocladius lacuniferus* Freeman, from New Zealand. Note the leg sheaths are all extended and none recurved under wing sheath. B. Pupal exuvia: abdomen of *Stictocladius uniserialis* Freeman, from Australia. The genus *Stictocladius*, following morphological review by Cranston & Sæther (2010) and Sæther & Cranston (2012) is now the subject of molecular study by Matt Krosch and Peter Cranston. The pupal stage not only provides good morphological features, but has provided valuable DNA even from exuvia, as outlined in the article.

of template DNA, 0.6 μL of each primer (10 pmol/ μL – manufactured by Geneworks, Adelaide, Australia), 1.75–3.5 μL of 5X MyTaq Red Reaction Buffer (Bioline, London, UK) and 0.2 μL of MyTaq DNA Polymerase (Bioline). Primer sequences and PCR protocols for both genes can be found in Krosch *et al.* (2011b). Amplified products were purified using an ISOLATE PCR and Gel Kit (Bioline) according to manufacturer’s protocols, but using only 13 μL of elution buffer and with a longer incubation time following addition of elution buffer to increase final concentration of amplified fragments. As with DNA extraction, choice of manufacturer of kits for the purification of amplified products is likely not to influence success rates greatly. Purified fragments were sequenced using ABI BigDye® Terminator v3.1 chemistry, precipitated using a standard ethanol-based protocol and analysed on an ABI3500 sequencing platform at the Molecular Genetics Research Facility (QUT, Brisbane). For more detailed information on methods and success of ‘routine’ DNA extraction from larvae or pupae we refer the reader to our existing publications cited below that use molecular data.

Vouchering specimens

In the early days, the few people working with chironomid DNA had a rather casual approach to vouchering material needed to verify that sequenc-

es obtained actually derived from a named and retained specimen (Jon Martin U. of Melbourne, pers. comm, Peter Cranston, pers. obs.). This arose partially in the belief that DNA extraction from small insects involved grinding up (destroying) at least a complete specimen, sometimes more than one, because of the demands of the salting-out protocol (Miller *et al.* 1988). Maintenance of reliable vouchers followed with: 1. growing awareness that not all published sequences were what they were purported to be; 2. recognition that ‘inessential’ parts of a single specimen could provide enough DNA by newer extraction protocols, and the rest be retained as a voucher; 3. recognition that the cuticle alone, after gentle extraction of DNA, provides a complete voucher; and 4. multiple specimens from natural populations should and need never be co-extracted. However, the latter point does remain viable for multiple specimens from laboratory culture lines or egg masses for use in downstream applications that require large quantities of DNA, such as optimisation of DNA systems or PCR primers.

Procedures are a little time-consuming and need accurate recording – we give each potential molecular specimen a unique ‘site (year date) plus sequential numerical code’ at the outset – whether or not DNA is obtained. This unique identifier is as-

sociated with the DNA throughout processing, and with the cuticle obtained after extraction, or the slide-mounted larval head capsule if it is only the body that is taken through extraction. These slide mounted materials are treated with the same care in preparation, curation and specified repository as type material in a taxonomic study. The unique specifier code and associated data remains linked to the data submitted to Genbank (<http://www.ncbi.nlm.nih.gov/genbank/>).

Results

This technique, which already has worked well across other life stages from many subfamilies of Chironomidae, was trialed on exuvial specimens across a range of Orthoclaadiinae taxa. Of 58 exuvial specimens derived from separate field collections made at various sites across Australia during 2011 and 2012, extracted as described above, 27 have been successfully amplified and sequenced for *COI* and 17 for *28S*. Sequence quality always was acceptable under standard QC metrics (e.g. signal strength) applied by the sequencing software and sequencing success was repeatable via both multiple attempts using the same purified PCR product and multiple initial PCR reactions. Moreover, vouchers made from cuticles post-digestion showed that diagnostic characters remained intact and morphology - and molecular-based identifications converged for all specimens. A number of specimens, in addition to those successfully sequenced for either locus, amplified successfully (as assessed using 1.5% agarose check gels) but did not produce clean or useable sequence. This suggests that further optimisation of protocols may be necessary, especially with regard to the ratio of DNA template to primer in sequencing reactions.

It is not surprising that even this technique can fail to recover usable DNA from some specimens. Extraction of DNA from pupal exuviae relies on the presence of muscle tissue that is left behind on the inner surface of the cuticle by the emerging adult. DNA cannot be extracted from the cuticle itself. Thus, we suspect that the success of this technique is highly correlated with how long exuviae have been exposed to the aquatic environment after adult emergence. The longer the exposure time prior to collection, the greater the likelihood of any remaining muscle tissue being scoured or degraded. The same principle applies to the duration that specimens spend in alcohol collections. Furthermore, we note also problems encountered with degraded ethanol for preservation and quarantine of shipped specimens using X-rays (particularly to the USA). These factors act to shear DNA

strands and greatly reduce efficiency of extraction and amplification, often to the point of complete failure for entire collections. Moreover, given the much higher copy number of mitochondrial genomes within each cell, we anticipated that DNA extracted from exuviae would perform better for amplifying mtDNA loci than nuclear. Although we did not observe a dramatic difference in total numbers sequenced for each locus here, *28S* amplification required more troubleshooting than *COI*. The use of intermediate primers for *COI*, which often produce sequence data from older more degraded specimens (Jon Martin, pers. comm.) may allow even sub-optimal specimens to be used.

Discussion

The addition of PCR-quality DNA sequences from 'wild caught' pupal exuviae described here expands the number of sequenceable specimens for chironomid researchers who collect in aquatic environments. The greatest benefit of this technique will likely be in aiding the development of molecular associations between life stages of species by negating the need for rearing of larvae through to adults. This technique also has potential importance for those investigating population genetic structure, gene flow and dispersal where sample sizes must be maximised from each study site. We encourage other researchers to retain, and attempt to extract DNA from exuviae and not consider these important specimens valueless for molecular purposes.

However the addition of a 'new' life history stage, the pupal exuviae, to the repertoire of those engaged in molecular studies of Chironomidae should not obscure that even this suite of techniques do not provide the perfect answer to all questions. We note that there are sporadic and difficult to diagnose difficulties with extraction for some specimens or collection conditions in addition to the unknown age and history of collected exuviae, or interceptions of larvae, pupae and adults by netting. There are sporadic problems with amplification of DNA from mermithids rather than the midge host, apparently restricted to 18S rRNA and probably due to primer degeneracy.

The procedures and protocols associated with specimen tracking, vouchering and truly geocoded and permanently labelled specimens are particularly important as we recognise increased cryptic diversity. Unique vouchers allow us to revisit actual specimens (and locations) of 'known' molecular identity and seek to validate perhaps obscure or overlooked morphology that will allow non-molecular identification. Furthermore, GenBank

is not prescriptive concerning vouchering and thus the database is error-ridden and requires validation and correction for many anomalous taxa. To date we have no complete and reliable library to test our species identifications (Ekrem *et al.* 2007) and thus morphological vouchers for DNA sequences remain as important as morphological types. Webb *et al.* (2012) illustrate a salutary case of difficulties in tracking down morphological vouchers for molecular databased N. American Ephemeroptera, despite the DNA barcode initiative's long-standing instructions concerning appropriate vouchering. DNA taxonomy does not over-ride the common sense understanding of how to go about systematic science, regarding repeatability and documentation of evidence.

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KIEFFERULUS BARBATITARSIS (KIEFFER, 1911) AND *KIEFFERULUS TAINANUS* (KIEFFER, 1912) ARE DISTINCT SPECIES, NOT SYNONYMS

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Abstract

Morphological and molecular studies indicate that *Kiefferulus tainanus* (Kieffer 1912) is quite distinct from *K. barbatitarsis* (Kieffer 1911), and not a synonym of the latter species, as has previously been suggested by Chaudhuri and Guha (1987). The suggestion of synonymy seems to have been based on a comparison of the figures of *K. tainanus* in Sasa (1979), rather than an examination of the types. While the adults show some similarities, other characters, and mitochondrial COI sequence, clearly indicate that the two species are not identical, and not particularly closely related. Australian material previously considered to be *K. tainanus* is probably incorrectly identified and represents a separate, but closely related species.

Introduction

In his paper on chironomids of Thailand, Cranston (2007) noted that the identity of *Kiefferulus* species of South East Asia was not clear. One group for which uncertainty has existed in the past, is three species described by Kieffer: *K. barbatitarsis* (Kieffer 1911), *K. tainanus* (Kieffer 1912), and *K. biroi* (Kieffer 1918). Kieffer originally placed *K. barbatitarsis* and *K. biroi* in *Chironomus*, but he described *K. tainanus* as a *Tendipes*. *Kiefferulus barbatitarsis*, described from India, remained in *Chironomus*, although Sublette and Sublette (1973) classed it as unknown Chironomini. Chaudhuri and Ghosh (1986) re-examined the types in the Indian Museum, along with rearings, and re-described the species as *Kiefferulus*.

Kiefferulus tainanus was originally described from Tainan, Taiwan. It was variously placed in *Phytochironomus* (Kieffer 1921) and *Glyptotendipes* (Goetghebuer 1937-54), before Sasa (1979) re-described it for all stages from Japanese specimens. He placed it in *Chironomus*, although noting that it did not fit the strict definition. Hashimoto *et al.* (1981) also placed it in *Chironomus*, but noting that it was “rather related to *Kiefferulus* and *Glyptotendipes*”. Chaudhuri and Ghosh (1987) placed it in *Kiefferulus*, when they placed it as a probable synonym of *K. barbatitarsis*, then Cranston and Martin (1989) placed it in *Nilodorum*, before restoring it to *Kiefferulus* in an analysis that included

a syntype in the British Museum (Cranston *et al.* 1990).

K. biroi was originally described from Colombo, Sri Lanka, and later from Australia, India and Japan. Freeman (1961) placed it in the subgenus *Nilodorum* of *Chironomus*, and later as the genus *Nilodorum* (Freeman & Cranston 1980). Saxena *et al.* (1985) also referred Indian specimens to *Nilodorum*. Hashimoto *et al.* (1981) had synonymised it with *C. tainanus*, but the rather obscure publication was generally unknown. The synonymy was restated by Cranston and Martin (1989), and again when *K. tainanus* was returned to the genus *Kiefferulus* (Cranston *et al.* 1990). However, the possible synonymy of these species with *K. barbatitarsis* has been largely ignored. Chaudhuri and Guha (1987) apparently did not examine any type material of *K. tainanus*, but rather relied on the quite detailed re-description of Sasa (1979) since they attribute *K. tainanus* to Sasa in their listing of synonymies. The purpose of this paper is to provide morphological and molecular data to clarify that the two species, *K. barbatitarsis* and *K. tainanus* as re-described by Chaudhuri and Ghosh (1987) and Sasa (1979) respectively, are not in question and that it is clear that they are morphologically distinct.

Material Examined

While the conclusions here are largely based on a comparison of previously published work, some additional specimens were examined.

Kiefferulus barbatitarsis:

1 male Mai Ping N.P., Lamphung Province, Thailand. 6 III.2002, coll: P.S.Cranston. Part of abdomen used for DNA extraction (BOLD CoTW018-08), and photo of hypopygium used in Figure 1.

Kiefferulus tainanus:

1 male believed to be syntype, bearing three labels, respectively ‘tainanus Kieff. det Kieffer’, ‘Formosa Sauter’, ‘Purchd. from Budapest Mus. BM 1922-72’ (British Museum Natural History) - used to establish *K. tainanus* by Cranston *et al.* (1990), and a photograph of the hypopygium used in Figure 1.

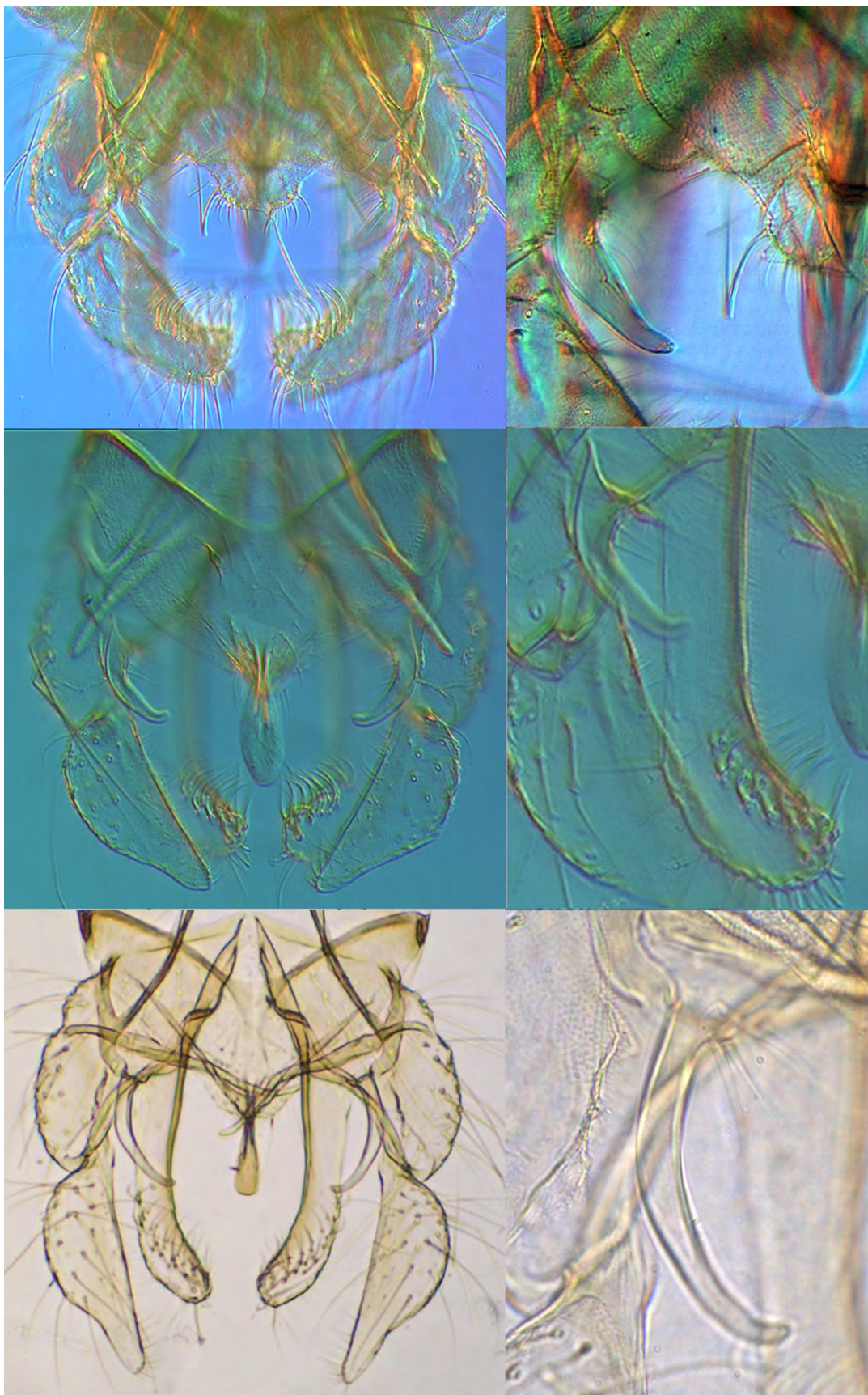


Figure 1. Male hypopygium (left) and superior volsella (right) of *K. barbatitarsis* (top), *K. tainanus* (middle), and Australian *K. "tainanus"* (below). Photos at top and middle courtesy of P.S. Cranston.

1 male Okhla, nr. Delhi, India, OK1, coll. S. Taneja (now Saxena); 1 male pupal exuviae Yamuna River, Okhla, nr. Delhi, India OK2; larva Honshu, Japan, 9.IX.2001 GenBank accession no. DQ648225), 1 male Mai Ping N.P., Lamphung Province, Thailand. 6.III.2002, coll: P.S.Cranston. Part of abdomen used for DNA extraction.

Australian specimens previously considered to be *K. tainanus*:

1 male Goanna Lagoon, Alligator Rivers Region, Northern Territory, 15.IX.1979, Coll. R. Marchant; 1 male Somerset Dam, Queensland AQ.20.10 M2, 26.V.1971, coll. J.Martin; Hutchins Lagoon, Ayr, Queensland, 14.VI.1974, coll: B.V.Timms; 4 larvae with associated chromosome squashes, Somerset Dam, Queensland, AQ.20.4, 23.I.1969, from egg mass #1, coll. J. Martin. Additional specimens were included in the morphological studies of Cranston *et al.* (1990) and the cytological studies of Saxena *et al.* (1985).

Observations

The published re-descriptions and the analysis of additional specimens leaves no doubt that material described as *K. barbatitarsis* is distinct from that described as *K. tainanus*. Both species were collected together at a site in Thailand, and there was no difficulty in separating them, as indicated by the molecular analysis of two such specimens (see below). On the other hand, there is no doubt that the adults of the two species are somewhat similar in gross morphology. Both have an AR around 4, and an LR around 1.25. The male hypopygium is also basically similar, including the presence of setae on the inner margin of the superior volsella (see Fig. 1).

Closer examination reveals that there are definite differences, in all life stages as seen in Table 1.

Possibly the most obvious is the relatively shorter palps of *K. tainanus*, as can be seen in Table 1, which had led to it being placed in the genus

Nilodorum (e.g. Cranston and Martin 1989). Another obvious difference is in the shagreen pattern of the pupa. While both species have an anterior and posterior row of spines on tergite II, spines on other tergites of *K. barbatitarsis* are relatively small (Chaudhuri & Ghosh 1986), while those of *K. tainanus* are larger and more extensive (Fig. 2). The tergal spines of *K. tainanus* were well illustrated by Sasa (1979). It might be noted that material identified as *K. tainanus* in Australia (Saxena *et al.* 1985, as *N. biroi*; Cranston *et al.* 1990, Bugledich *et al.* 1999) is probably a distinct but closely related species. The most obvious difference is that the male superior volsella is longer and narrower

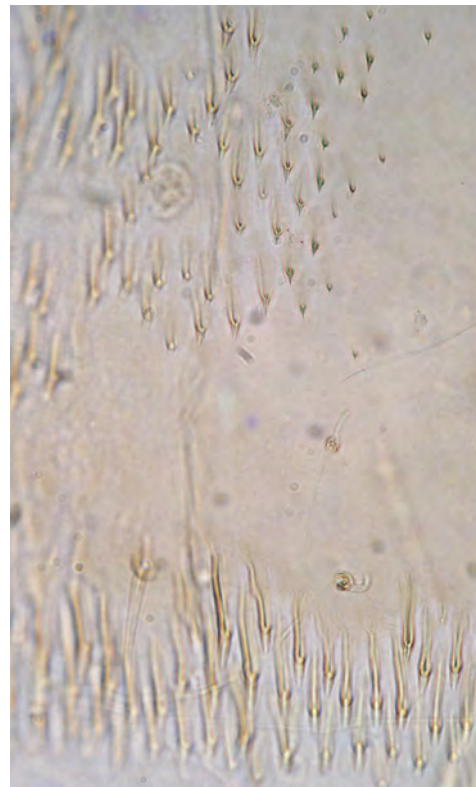


Figure 2. Spinose patches on posterior two thirds of tergite V of pupa of *Kiefferulus tainanus*. Note the shorter median spines and longer posterior spines.

Table 1. Listing of most obvious differences between *Kiefferulus barbatitarsis* and *Kiefferulus tainanus*. Abbreviations here and in text as in Sæther (1980).

Character	<i>Kiefferulus barbatitarsis</i>	<i>Kiefferulus tainanus</i>
Adult male		
Anal tergal band	H-type	Y-type
Palp ratios	4: 3: 7: 11: 15	4: 3: 5: 8: 10
IV (Fig. 1)	more swollen distally	less swollen distally
Pupa		
Spines of tergites III-VI	relatively small	larger & more extensive (Fig. 2)
Larva		
S1 setae	deeply feathered	palmate

(Fig. 1), while further evidence can be drawn from the cytological comparison of Indian and Australian specimens (Saxena *et al.* 1985), where a small number of fixed differences were noted in the banding patterns of the salivary gland chromosomes in the two continents. Further study will be required to clarify the situation.

In the larvae, the main difference is in the S1 setae (Table 1), which differ in the manner previously used as a distinction between the genera *Kiefferulus* and *Nilodorum* (Cranston *et al.* 1990).

Finally, the DNA barcode sequence of the mitochondrial COI gene of *K. tainanus* from Japan has been published (Martin *et al.* 2007), and a further sequence was obtained from an adult from Thailand. These can be compared with the equivalent data for *K. barbatitarsis* (Fig. 3). As previously noted, the *K. barbatitarsis* sequence came from the same specimen as the hypopygium in Figure 1.

This comparison shows that, while there are 14 polymorphic sites between *K. tainanus* from Japan and India (2.3%), there are 42 (6.8%) and 39 (6.3%) respectively between the Japanese and Thai sequences of *K. tainanus* and the sequence of *K. barbatitarsis*. While the difference between the

ered to be in different genera (see Introduction). In a subsequent listing of Indian Chironomidae, Chaudhuri *et al.* (2001) did not mention *K. tainanus* at all, but included *K. biroi* in the genus *Nilodorum*. Since *K. biroi* is accepted as a synonym of *K. tainanus* (see Introduction), this provides further confirmation that this species is quite distinct from *K. barbatitarsis*.

Acknowledgements

I am grateful to Peter Cranston for helpful discussion and for making available material of *K. tainanus* and *K. barbatitarsis* from Thailand for DNA extraction, and for providing the photographs of the hypopygia of both species. Also to Kimio Hirabayashi for providing larvae of *K. tainanus* from Japan for DNA sequencing. Sumitra Saxena provided specimens of *K. tainanus* from India, while material from Australia was collected in the course of fieldwork funded by the Australian Research Grants Committee and the Office of the Supervising Scientist of the Alligator Rivers Region, with further funding supplied by The University of Melbourne.

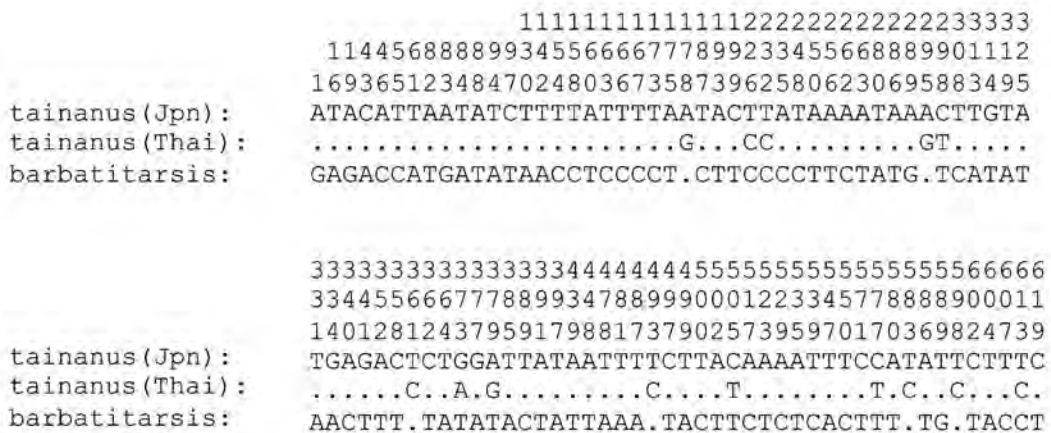


Figure 3. Polymorphic sites in 621 bases of the mitochondrial COI sequences of two populations of *K. tainanus*, and *K. barbatitarsis*

two *K. tainanus* samples is well within the arbitrary five percent limit for intraspecific variation of this sequence, the difference of the *K. barbatitarsis* sequence falls outside that limit. In a Neighbor-joining tree of *Kiefferulus* species (not shown), the two species do not cluster together.

It therefore must be concluded that the gross similarity of some adult characters does not indicate any particularly close relationship. Differences exist for larvae and pupae, as well as for the adults. Indeed the two species were previously consid-

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SHORT COMMUNICATIONS

An easy to make and simple designed rearing apparatus for Chironomidae

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Introduction

Identification of immature Chironomidae can be impossible, as most species are identified based on adult males, for which the associated pupae or larvae are not known or described. To overcome this, Chironomidae researchers rear larvae and pupae in aquaria to obtain the adults. This is usually done on fourth or final instar larvae collected from aquatic habitats, and mass reared (e.g. Biever 1965; Hein and Mahadeva 1993). An advantage of mass rearing is that a large number of specimens can be obtained for identification. However, with mass rearing, matching individual larvae with pupal and adult stages is not possible. To overcome this, larvae can be grown individually, a potentially costly undertaking. Even though most chironomids can withstand the room temperatures, most cannot tolerate low oxygen levels that are created in the vials (Mendes 2002).

In this paper we describe an inexpensive and rapid method of constructing an apparatus so that all three stages of individual specimen can be obtained, and that maintains sufficient oxygen during the rearing process.

Materials and Methods

Rearing vials were placed in a modified base constructed from centrifuge tube stands made of styrofoam, which are readily available in chemistry or biology labs. A standard foam is 20cm long, 16cm wide and 3cm deep, and has 25 holes for individual centrifuge tubes. To make the base we cut a foam stand in half to produce a 10cm by 16 cm stand, and glued this to a second stand to create a stepped structure (Figure 1i).

The rearing chamber was constructed from two centrifuge tubes to create an angled chamber with a screw cap on each end. The bottom end with screw cap was made water tight using plumbers tape and further sealed from the outside of the screw cap using aquarium glue. The top cap was left removable to allow access, and removal of emergent adults.

To modify the tubes, the bottom tube was cut perpendicularly at the 7.5 cm height (Figure 1ii). The bottom section had two drilled holes, one sized to receive an air tube that feeds the centrifuge tube from an air pump, and the second used to feed the larvae. We found that the second hole could be dispensed with, and reared 4th instar larvae successfully in using stream water with no feeding access port. When the second hole was installed, it was outfitted with a small plastic tube, 1.5 cm in length to allow feeding (Figure 1iii). The top section was cut diagonally at the tip at a 45° angle (Figure 1ii). The bottom vial was then inserted into the top at a 135° angle (Figure 1iii) and the joint of these two sealed using aquarium silicon. Finally, both the feeding tube and the air tube were made watertight using aquarium cement.

Using this apparatus, stream water and individual larvae were added to the rearing chamber from the top, and the chamber secured to the base using tape. The bottom fitted the stand holes and was secure.

Because the apparatus is small, any air pressure entering needed to be low to prevent water from splashing in the chamber. We adjusted the air pressure using hose clamps, and found that 1-2 air bubbles every second provided sufficient aeration.

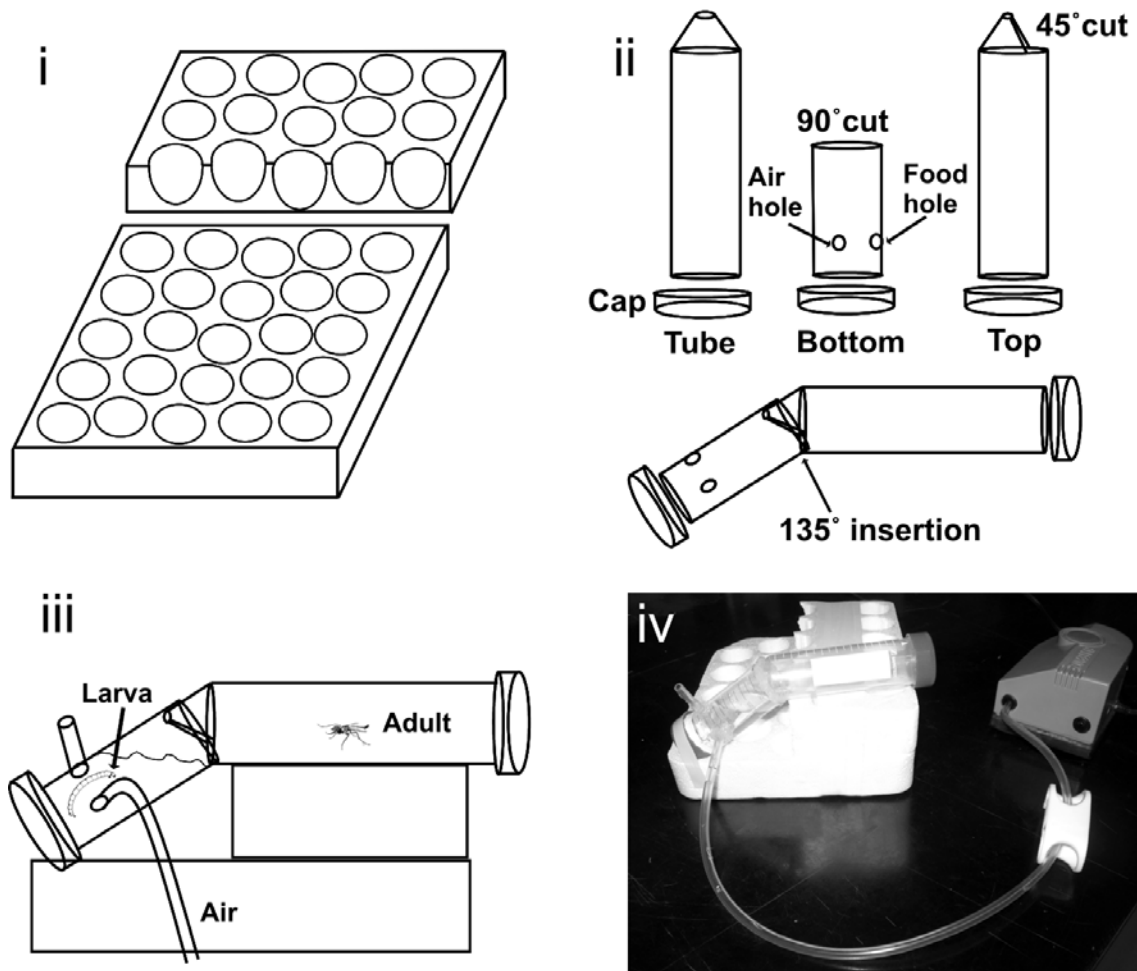


Figure 1: i) Apparatus stand made from centrifuge tubes stands ii) Centrifuge tubes, bottom and top parts, the position of holes and cuts, and assembling the top part to the bottom part iii) Apparatus on stand, position of larvae and adults iv) The apparatus assembled.

The larvae were placed at the base of chamber, and adults emerged into the top section, and remained dry due to the lip. Pupae exuviae and larva head capsule were obtained from the bottom part once the adults were removed (Figure 1iii). Using this apparatus we have been able to acquire all three stages of Chironomidae for identification.

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Some proposed emendations to larval morphology terminology

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Despite the publication of an intended consistent terminology for the morphology of chironomid larvae (Sæther 1980) as applied in Wiederholm *et al* (1983), two areas have remained controversial, with inconsistently applied terminology. The first and perhaps most confusing concerns the dorsal head sclerites, the second, the terminology of the mandibular teeth of Tanypodinae. In preparation for the 2nd edition of the Holarctic larval keys, I have attempted to reconcile differing approaches to naming the parts of these larval cephalic structures.

I started with recognising that we need hypotheses that (a) cover putative homology across insects as a whole, (b) conform across all ‘Nematocera’ with complete head capsules, and (c) can be implemented across all Chironomidae. Of course homologies are hypotheses derived from data (observations) and thus are not ‘right’ or ‘wrong’, only tested / refuted by further data. All names of anatomical (morphological) parts, purportedly ‘neutral’ or not, are hypotheses of ‘sameness’ = homology. Thus we should use names for parts that clearly hypothesise homology – especially when they are available and have meaning outside the subfamily Chironominae. Equally evident is that we cannot and should not have a separate morphological terminology from closely related groups and our use in chironomids should not be at variance with standard entomological works such as Snodgrass (1935) and Torre-Bueno (1989), and modern textbooks (e.g. Gullan & Cranston 2010).

First I sought to understand the dorsal head, which proves variation so useful in taxon-discrimination in, for example, the Chironominae (e.g. *Goeldichironomus* in Reiss, 1974, *Endochironomus* group in Grodhaus, 1987, and *Kiefferulus* in Cranston, 2007) (Fig. 1). The terminology applied previously to the various parts has been a mixture of homology and ‘neutral’ terms, creating some confusion and inconsistency in their application. Starting from basic insect head morphology, it should be noted that there is no 1:1 relationship between sclerotised parts of heads and primitive segmentation. Although there is thus a certain arbitrariness in defining head parts, never-the-less regions of the head across all insects can be recognised by sutures – these parts are the frons, clypeus, labrum in posterior-anterior order. There are no other recognised regions in any group that are available for insertion in this sequence.

At least across Nematocera, but seemingly much more widely, there are defining cephalic setae and sutures (differentiated borders) associated with each part. Thus S1 and S2, the labral setae, belong on the labrum, S3, the clypeal setae, on the clypeus, and S4 and S5, the frontal setae, on the frons. There is no need for the term frontal apotome – ‘apotome’ is either redundant or misleading. In Torre-Bueno (1989) its use is ascribed only to the Chironomidae. The frontal apotome is the frons.

Starting posteriorly, cephalic setae S4 and S5 lie on the frons, a uniform sclerite that fills the area completely, with the exception of a thinned area, the fenestra, in certain Chironomini (*fen* in Fig. 1). The frons lacks membranous parts that otherwise would allow flexibility and therefore it is rigid. It is bounded posteriorly and laterally by the ecdysial lines along which the cuticle opens at ecdysis (emergence of the next stage). Anteriorly often is a boundary of less sclerotised cuticle, anterior to which lies the head region called the clypeus (*clyp*, delimited in Figs 1A, B, E by a red boundary line). This region includes membranous and denser cuticle – the thicker median area, termed a sclerite, is of variable shape and size but rarely fills the entire clypeal area. The S3 (clypeal setae) can (rarely) lie on the clypeal sclerite, but are located at the postero-lateral corner of the clypeus, usually on membranous cuticle that allows increased flexion. The anterior margin of the clypeus also is membranous, abutting the labral sclerite (*lab*) which varies in degree of subdivision. The S1 labral setae often, but not always, lie on the labral sclerite. Laterally the labrum bears the S2 labral setae. Here there may be up to 3 pairs of lateral sclerites which may be complete, partially or totally fragmented, or not discernible from the background. These visible lateral sclerites may be distinctive and numbered 1-3 as in Fig. 2, but more usually the structures are partially or completely fused with indistinct subdivision (see variation in this development in Fig. 1, especially in unlabelled G, H, I). Anteriorly the labrum bears the SI - SIV setae, labral chaetae etc and terminates antero-laterally at the tormal bar.

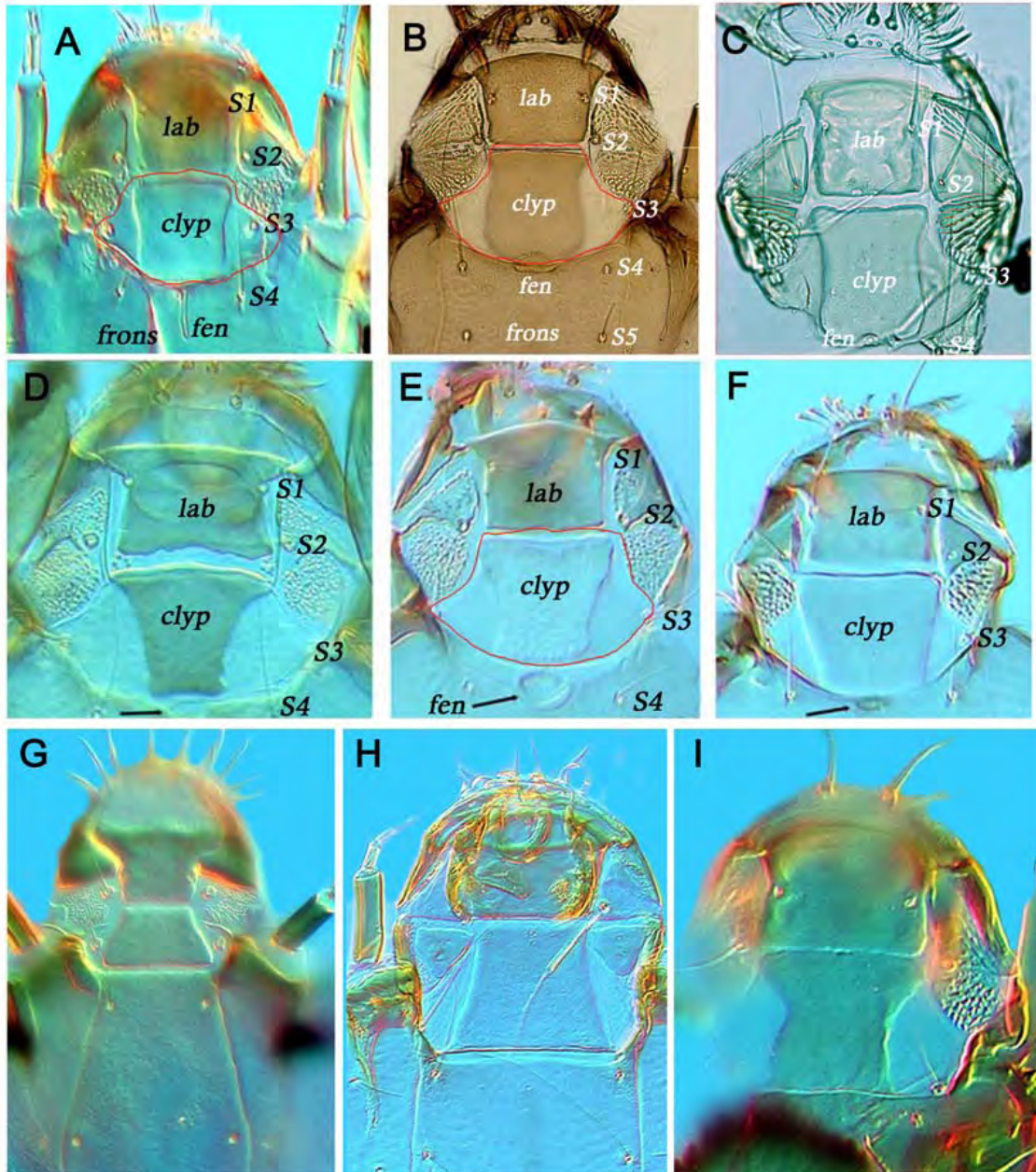


Figure 1. Dorsal larval heads of Chironomini. A. *Dicrotendipes septemmaculatus* (Becker); B. *Glyptotendipes paripes* Edwards); C. *Glyptotendipes* 'sp. E' Epler 2001; D. *Kiefferulus longilobus* (Kieffer); E. *Kiefferulus calligaster* Kieffer; F. *Kiefferulus barbatitarsis* Kieffer; G. *Endotribelos* sp.; H. *Goeldichironomus carus* (Townes); I. *Goeldichironomus* sp.

This unit comprising the frons plus clypeus plus labrum is 'split off' as a single structure at ecdysis – it is completely bounded by weak ecdysial lines. The genae are not part of this, and there are no genal elements on the dorsal head. This is the basic pattern against which variants can be defined. Thus fusion of the clypeus with frons, with S3-5 on a single unit – is a frontoclypeus. Fusion of frontoclypeus with labrum is frontoclypeolabrum, bearing all S setae (1-5). Rarely (uniquely) there is a fusion of clypeus and labrum to form a clypeolabrum (*Kribiodorum*). The anterior margin of the frons or frontoclypeus may be straight, concave or convex. The frons, whether separate from the clypeus or fused, can have a fenestra (interiorly thinned cuticle). We do not know if 'pits' are homologous to oval fenestra, but certainly 'pits' do not co-occur with fenestra and are not 'pits' (depressions in the external cuticle).

The test of this hypothesis is its applicability in all Chironominae, and hopefully beyond. An issue is that in taxonomic studies dorsal heads are not always illustrated. In many cases this seems to be because of apparent uniformity, for example in Tanypodinae, the *Harnischia* cpx and many Tanytarsiini. However, all

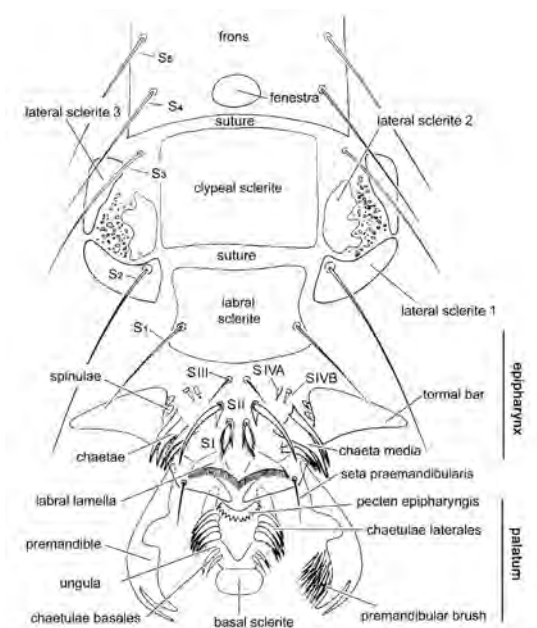


Figure 2. Labelled schematic for dorsal larval head, Chironominae.

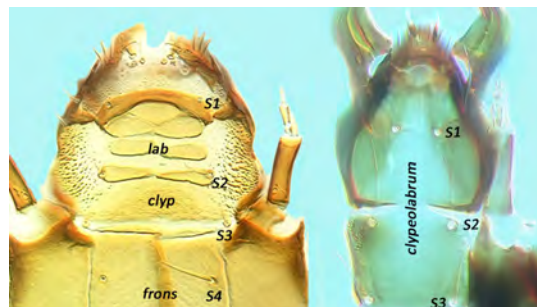


Figure 3. Dorsal larval heads of Orthoclaadiinae. A. *Brillia*, B. *Elpiscladius*

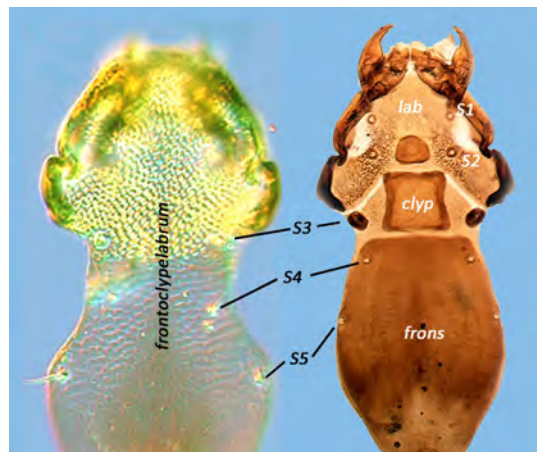


Figure 4. Dorsal larval heads of Telmatogetoninae. A. *Telmatogeton*, B. *Thalassomyia*

images in the Lucid on-line key (<http://keys.lucidcentral.org/keys/v3/Chironomidae/>) have been viewed. The morphology of *Glyptotendipes*, which include the greatest variation and demonstrate most complexity, was discussed with Michael Heyn.

Since the scheme as labelled in Figures 1 and 2 ‘worked’ even for problematic taxa, I verified (and sometimes emended) the generic diagnoses for the revision of the Holarctic Chironomidae for which the feature is applicable. Difficulties were encountered only in the strange arrangement in *Kribiodorum* but I could locate no description or figure that could be reconciled with the ‘divided labral sclerite 1 and 2’ of Sæther’s glossary (1980: fig. 62). However the situation occurs only in *Xenochironomus* – but I don’t believe that this anomalous dorsal head structure damages the proposed scheme. The caricature dorsal head, as labelled in Figure 2, thus appears applicable across all Chironominae

The dorsal heads of Orthoclaadiinae generally follow Hirvenoja’s (1973) figures, including cephalic setal terminology and locations - except in *Brillia* group in which there are interesting variations (Fig. 3). All can be incorporated into the new naming scheme, although the situation in South African *Elpiscladius* (Fig. 3B) is unusual. In contrast to the original interpretation (Cranston 2008), now I interpret *Elpiscladius* as showing a fused clypeo-labrum in which there is secondary near division into anterior section with S1 seta, and posterior, section with S2 and S3 (the separate frons, bearing S4 and S5, is omitted from the image).

The distinctive variation in dorsal head morphology in the Telmatogetoninae was recognised by text and drawings in Cranston (1983). Terminology can be emended easily to follow the new system, as labelled in Figure 4.

The most misleading interpretation concerned the dorsal head of the Tanypodinae, but fortunately the feature has been little used in identification. The problem is misinterpretation of the cephalic S setae, including by Müh (1985) who mistakenly termed and labelled the frontal seta S5 as S4 (no S5 is labelled or mentioned in Müh). This, or a similar error, may explain the strange statement “clypeus covering almost entire head” (Fittkau and Roback, 1983: p. 33). Actually it is the frons that extends nearly entirely to cover the dorsal head, with S1, 2 and 3 on poorly-differentiated, far-anterior, clypeo-labral microsclerites.

The proposed scheme thus seems validated across the family. In implementation it is important to recognise Gail Grodhaus who interpreted the S3 / clypeus issue (Grodhaus. 1987) as a problem of logic. Of course, Gail worked also with mosquito larvae, so his outgroup was appropriate.

Turning to the tanypod mandible: Fittkau & Roback (1983) used a different terminology for describing the various associated teeth. However, the terms 'apical tooth' and 'inner teeth' can and should be used as in other subfamilies. If we recognise the widespread term 'mola' for the inner mandibular margin basal to the insertion of the seta subdentalis, then distal projections of the mola should not be treated as 'inner' teeth, as so termed in all other subfamilies, but molar projections (see Fig. 5). The inner teeth of the tanypod mandible, when present, should be restricted to those generally small teeth that lie distally between the mola / site of seta subdentalis insertion and the base of the strong apical tooth.

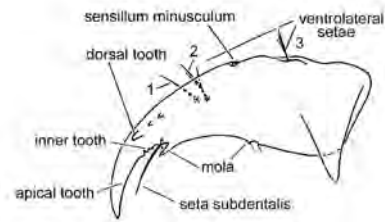


Figure 5. Labeled schematic for Tanypodinae larval mandible.

Acknowledgements

I thank particularly John Epler for engaging discussions on chironomid larval morphology, and Mike Heyn for correspondence concerning the dorsal heads of *Glyptotendipes*.

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Chironomids in Quaternary permafrost deposits in the Siberian Arctic

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Fossil chironomids from lacustrine sediments are frequently used as bioindicators for the reconstruction of Late Quaternary environments (Smol et al. 2005; Self et al. 2011), but there are very few records concerning chironomids from periglacial permafrost deposits (Ilyashuk et al. 2006).

Permafrost records, accessible at outcrops along the coast of Oyogos Yar at the NE-Siberian Dmitry Laptev Strait provided unique insights into the environmental history of Beringia during the last interglacial (Kienast et al. 2011). A paleontological study was carried out on the last interglacial (Kazantsevo, Eemian, Sangamoian, MIS 5) terrestrial and freshwater organism assemblages preserved in frozen deposits of a shallow paleo-lake outcropped at a permafrost cliff at Oyogos Yar (72.68°N; 143.53°E), the mainland coast of the NE-Siberian Dmitry Laptev Strait, Republic Yakutia, in the Russian Federation (Fig. 1). The Dmitry Laptev

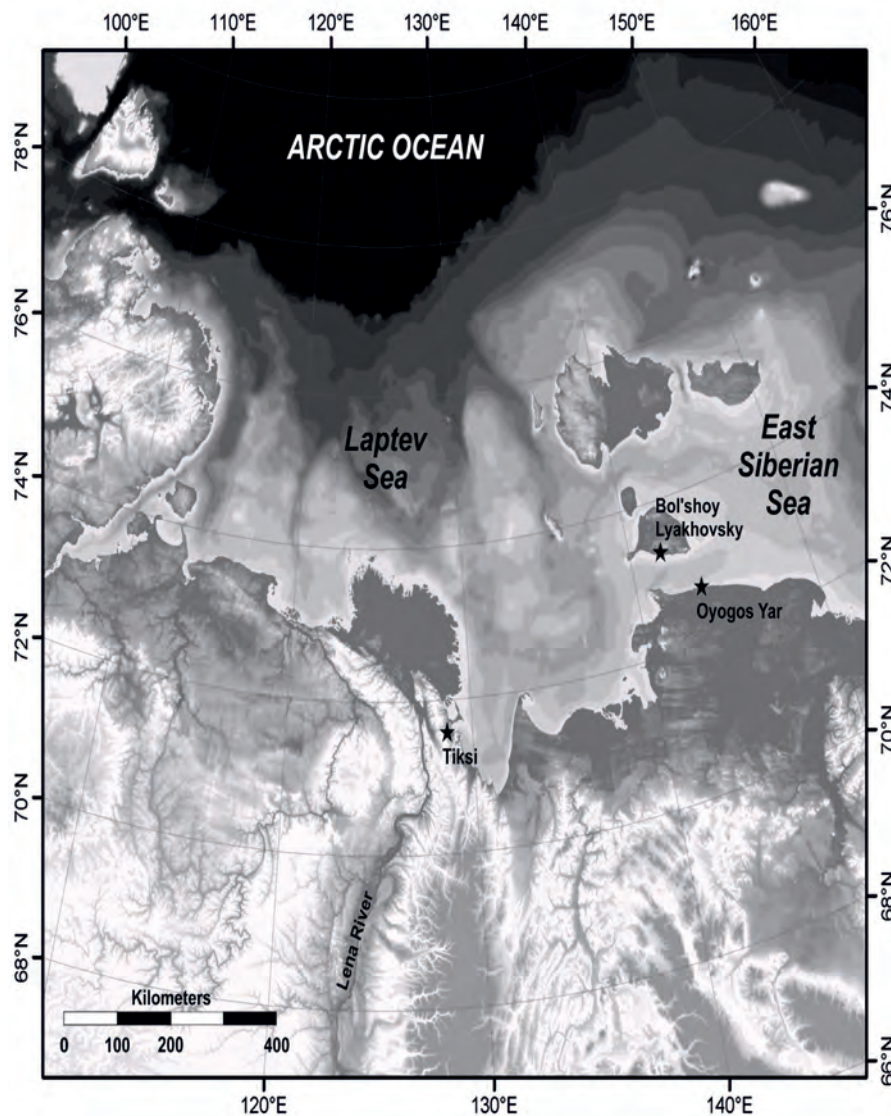


Figure 1. Location of the Bol'shoy Ljakhobskiy Island and coastal sector of Oyogos Yar in north-eastern Siberia.

Strait connects the Laptev and East Siberian seas, which are the widest and shallowest shelves worldwide. The mean temperature of the warmest month at the coast is about 4°C, the mean temperature of the coldest month is about -30°C (Station Cape Svyatoy Nos, 72° 53' N, 140°, 45' E in Rivas-Martínez, 1996-2009).

From a total of 50 chironomid head capsules, recovered from Eemian deposits, 16 taxa were identified (Table 1). Eight of these taxa belong to the subfamily Orthocladiinae and eight to the subfamily Chironominae (five Chironomini, three Tanytarsini). The dominant taxa were *Limnophyes*, *Smittia foliacea*-type and the *Psectrocladius sordidellus*-type. The majority of the taxa are indicators of temperate shallow lakes or littoral conditions that are associated with macrophytes. *Limnophyes* and the subdominant taxa, *Metriocnemus eurynotus*-type and *Parametriocnemus/ Paraphaenocladus* are also frequently associated with macrophytes (Cranston et al., 1983; Brodin, 1986). Furthermore, these taxa are typically indicative of lake level fluctuations (Massaferro & Brooks, 2002). *Smittia foliacea*-type could be indicative of erosional processes or unstable lake level conditions (Cranston et al., 1983). The subdominant taxa *Chironomus anthracinus*-type and *Cricotopus laricomalis*-type are among the most frequently occurring taxa in present-day Yakutian lakes and can be found in a broad range of ecological conditions (Nazarova et al., 2005, 2008, 2011). *Tanytarsus pallidicornis*-type 1 is characteristic of relatively warm and productive lakes (Brodin, 1986). In contrast, *Tanytarsus lugens*-type and *Parakiefferiella triquetra*-type are cold stenotherms, occurring in oligotrophic cold subarctic lakes (Walker & Mathewes, 1989; Brodin, 1986). *Brillia* and *Endochironomus albipennis*-type often are associated with submerged wood and dead leaves (Cranston et al., 1983). Some species of the group *E. albipennis*-type are leaf or stem miners (Pinder & Reiss, 1983).

The chironomid record indicates the presence of submerged coarse plant debris in the paleo-lake, most probably the remains of trees and shrubs. Most of the identified chironomids indicate shallow water conditions and a pronounced macrophyte zone (Cranston et al., 1983; Brodin, 1986).

The Eemian was an interglacial period which began about 130,000 years ago and ended about 114,000 years ago. It was the second-to-latest interglacial period of the current Ice Age, the most recent being the Holocene which extends to the present day. The prevailing Eemian climate is believed to have been similar to that of the Holocene. Comparison of the Eemian fauna and modern chironomid assemblages from the 35 small permafrost lakes of Bolshoj Ljakhobskiy Island and Oyogos Yar mainland coastal area has shown certain faunistic differences (Table 1) with a higher proportion of cold tolerant taxa in modern chironomid assemblages. Remains of terrestrial and freshwater organisms, including chironomids, preserved in frozen deposits of a shallow Eemian paleo-lake indicate a boreal climate at today's Arctic during that time. The use of transfer functions on the base of chironomid assemblages (Nazarova et al. 2011) brought a mean July air temperature reconstruction of 12.9±0.9°C and a water depth of 1.00±0.34 m for the time of deposition.

Table 1. Eemian chironomid fauna from Oyogos Yar permafrost deposits and most frequent chironomid taxa from Bolshoj Ljakhobskiy Island and Oyogos Yar mainland coastal area (Northern Yakutia, Russia. Taxa that are found in modern and in Eemian sediments are given in bold.

Modern lake deposits		Eemian Permafrost deposits (130,000 to 114,000 years ago)	
16 most frequent taxa	%	Taxa	%
<i>Paratanytarsus penicillatus</i> -type	22.4	<i>Limnophyes</i>*	22.0
<i>Chironomus anthracinus</i>-type	17.0	<i>Smittia foliacea</i> -type	18.0
<i>Psectrocladius sordidellus</i>-type	13.9	<i>Psectrocladius sordidellus</i>-type	14.0
<i>Orthocladus</i> type I	10.1	<i>Chironomus anthracinus</i>-type	6.8
<i>Derotanyptus</i>	5.4	<i>Cricotopus (I.) laricomalis</i>	6.0
Chironomini larvula	2.9	<i>Metriocnemus eurynotus</i>	6.0
<i>Limnophyes</i>	2.7	<i>Parametriocnemus</i>	6.0
<i>Corynoneura arctica</i> -type	2.7	<i>Brillia</i>	4.0
<i>Paratanytarsus austriacus</i> -type	2.2	<i>Microtendipes pedellus</i> -type	4.0
<i>Cladotanytarsus mancus</i> type 1	1.9	<i>Endochironomus albipennis</i> -type	2.0
<i>Orthocladus oliveri</i> -type	1.3	<i>Parachironomus varus</i> -type	2.0
<i>Metriocnemus eurynotus</i>-type	1.2	<i>Parakiefferiella triquetra</i> -type	2.0
<i>Orthocladus</i> type S	1.2	<i>Polypedilum nubeculosum</i> -type	2.0
<i>Trissocladius</i>	1.2	<i>Zavrelia</i>	2.0
<i>Acricotopus</i>	1.2	<i>Tanytarsus lugens</i>-type	2.0
<i>Tanytarsus lugens</i>-type	1.1	<i>Tanytarsus pallidicornis</i>-type 1	2.0

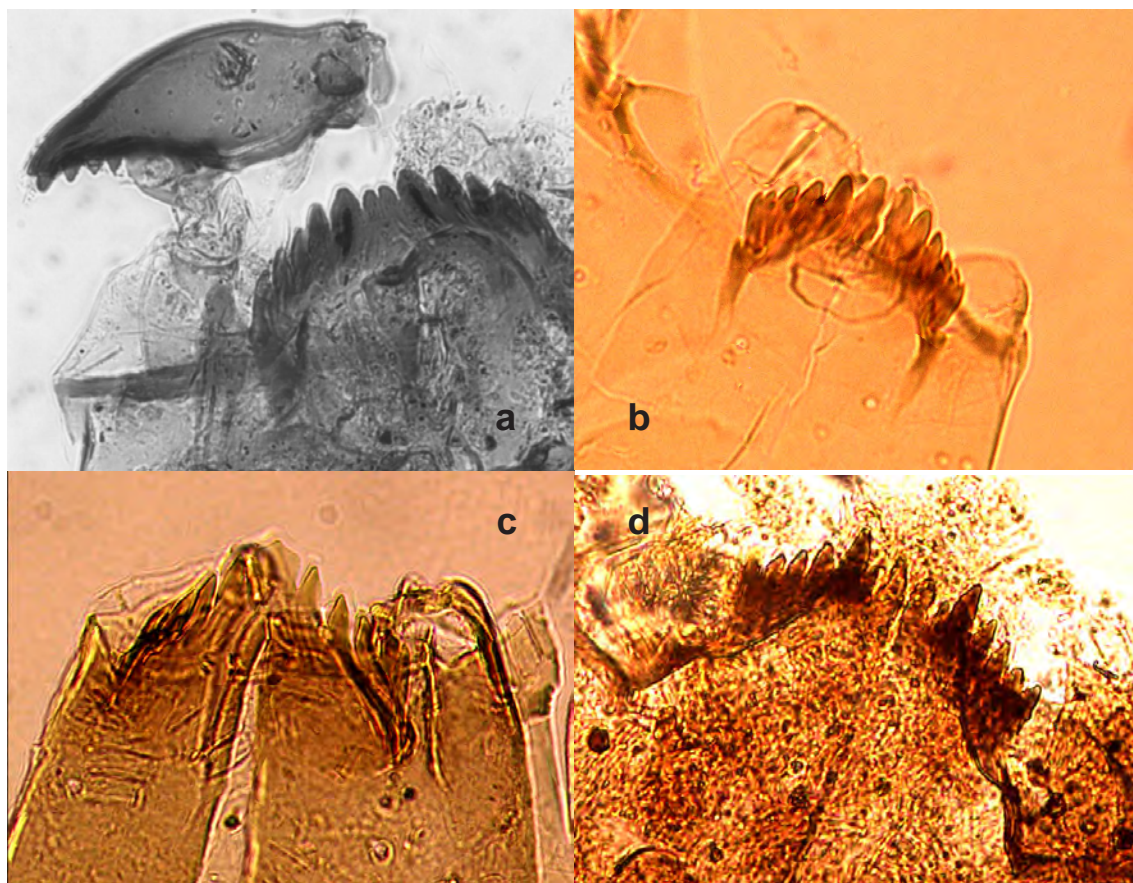


Figure 2. Photos of chironomids from Eemian permafrost deposits: (a) *Metriocnemus eurynotus*-type; (b) *Limnophyes*; (c) *Smittia foliacea*-type; (d) *Microtendipes pedellus*-type.

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New keys to European Chironomidae larvae

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Chironomini (Diptera: Chironomidae). Keys to Central European larvae using mainly macroscopic characters.

2nd, revised edition.

By Orendt, C. & Spies, M. (2012).

64 p., more than 450 illustrations.

ISBN 978-3-00-038842-2

The second edition of these richly illustrated keys separates larvae of Central European non-biting midges by subfamilies, and especially members of the tribe Chironomini by genera or some smaller taxa. The work is also directed at workers with little previous experience in chironomid larvae. The keys' emphasis is on morphological features that are visible macroscopically without elaborate preparations.

Chironomus (Meigen). Key to the larvae of importance to biological water analysis in Germany and adjacent areas.

By Orendt, C. & Spies, M. (2012).

Bilingual edition (English/German).

24 p., 47 illustrations.

ISBN 978-3-00-038789-0

This key identifies 11 species and species groups of significance to water quality evaluation whose ecological requirements are known sufficiently, at present, and whose larvae are reasonably diagnosable in terms of taxonomy. One of the aims of this work is to reduce existing uncertainties and misinterpretations, particularly in biological water analysis.

Chironomidae larvae in brackish waters of Germany and adjacent areas.

By Orendt, C., Dettinger-Klemm, A. & Spies, M. (2012).

Editor: Federal Environment Agency, Berlin.

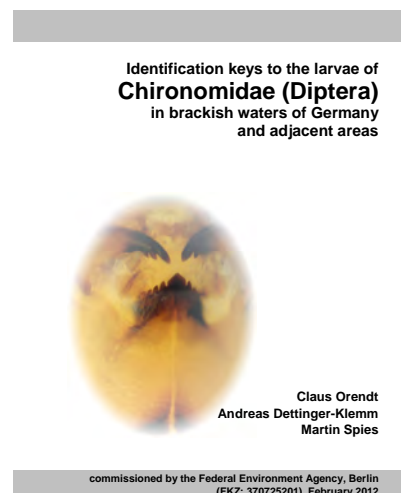
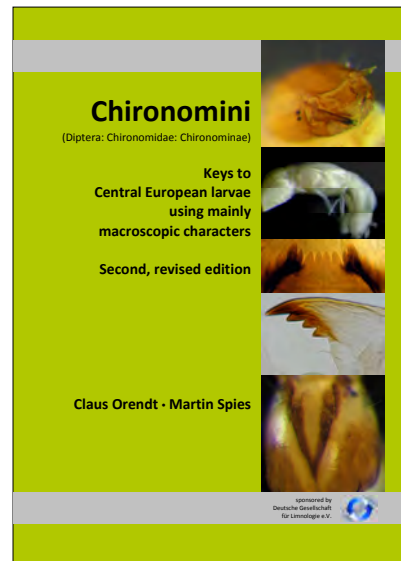
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214 p., 1154 illustrations.

ISSN 2194-7902

The key includes about 90 taxa (genera, species groups, species) that can be identified in the larval stage and have been recorded from brackish waters in Germany and adjacent North Sea and Baltic Sea areas. With its broad geographical scope and practical approach, this monograph is unique and of equal interest for scientists and technical offices in countries along the North and Baltic Seas.

Previews, informations and orders: www.hydro-bio.de/chironomidae.htm



Proceedings of the 18th International Symposium on Chironomidae

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We are happy to announce that the Proceedings of the 18th Chironomidae symposium held in Trondheim last year now is published and available open access through the Fauna norvegica website: http://www.ntnu.no/ojs/index.php/fauna_norvegica.

For people that wish a paper copy:

The editor of *Fauna norvegica* has informed us that there are extra copies of the proceedings available. The costs are NOK 300,- (Norwegian kroner), and includes postage.

To order please contact:
Torkild Bakken, Editor in chief
NTNU, Museum of Natural History and Archaeology
E-mail: torkild.bakken@ntnu.no

We wish to thank all authors for their contributions, and hope you all enjoy reading the papers!

Fauna norvegica Vol. 31, 2012



An online bibliography of the Chironomidae

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We are very happy to announce that the Online Bibliography of Chironomidae is available online at <http://literature.vm.ntnu.no/Chironomidae/>

As many of you know, there has been a number of people involved with the printed bibliography over the years and it is therefore nice to be able to make their work available in electronic form.

The database currently holds 27780 entries (nov. 20, 2012), but we hope that you all will contribute with missing data and add references to your own published work. To do so, just follow the link in the top right corner of the introductory page.

Thank you!

Author	Year	Title	Journal/Book/Publisher
"L'In Société de Naturalistes et d'Agriculteurs"	1816-1819	Nouveau dictionnaire d'histoire naturelle, appliquée aux arts, à l'agriculture, à l'économie rurale et domestique, à la médecine, etc. Tome 10	Deterville, Paris
Abbott, I.	1996	Phidromus of the occurrence and distribution of insect species in the forested part of south-west Western Australia	CALMScience 1, 365-483
Abbott, I. M., Sheehan, D. P. and Harrison, S.	2009	Bit activity affected by sewage effluent in Irish rivers	Biol. Conserv. 142, 2004-2014
Abulafia, A. M., Ezzion, D. R.	2003	Environmental factors controlling the distributions of benthic invertebrates on rocky shores of Lake Malawi, Africa	J. Or Lakes Res. 29, 202-216
Abulafia, E.	1980	Distribution of the bream Abramis brama in lakes of the Khaszmi Oblast, Uzbek SSR, USSR	Uzb. Biol. Zh. 1980, 4, 43-45

A World Catalogue of Chironomidae (Diptera). Part 2. Orthocladiinae

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Part 2 of A World Catalogue of Chironomidae (Diptera) is now available and was published on the 1st October 2012. In Part 2 there are a total of 174 valid genera, 36 subgenera and 2,275 valid species plus 19 subspecies. In addition, there are a further 113 genus-group names and 1,470 species-group names which are either synonyms, nomina dubia or unavailable. The total number of taxa (genus-group + species-group names) in Part 2 is 4,087. The full citation is as follows:

Ashe, P. & O'Connor, J. P. 2012. *A World Catalogue of Chironomidae (Diptera). Part 2. Orthocladiinae*. Irish Biogeographical Society and National Museum of Ireland, Dublin. xvi+968pp.

Due to the large size it is printed in two sections, Section A (i-xvi, 1-468 pp) & Section B (469-968 pp). The two sections each weigh about 1.5 kg and to save on postage costs each section is posted separately. The total cost of a copy of Part 2 (including both sections) plus postage (Airmail only) and packing can be determined from the table given below.

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Catalogue Part 2 is published by The Irish Biogeographical Society in association with The National Museum of Ireland. Details about this publication will be included on the Irish Biogeographical Society website: <http://www.irishbiogeographicalsociety.com/>

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Copies of Part 1 are still available:

Ashe, P. & O'Connor, J. P. (2009) *A World Catalogue of Chironomidae (Diptera). Part 1. Buchonomyiinae, Chilenomyiinae, Podonominae, Aphroteniinae, Tanypodinae, Usambaromyiinae, Diamesinae, Prodiamesinae and Telmatogetoninae*. Irish Biogeographical Society and National Museum of Ireland, Dublin. 445pp.

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TOTAL	Euro 50.00	Euro 55.00	Euro 55.00	Euro 60.00

The 12th Workshop on Subfossil Chironomidae

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Dates: 10-13th June 2013

Venue: Beaulieu Hotel, New Forest, Hampshire, Southern UK (the beautiful English countryside!)

Costs (all incl VAT): Total price pp for single occupancy would be £309.60 pp. Double occupancy would be £269.00 pp. Triple occupancy would be £255.00 pp. Day delegate rate £37.80 pp (incl lunch). Dinner as an extra add on for any non-residents would be £25.00 pp. If staying 10-12th (ie leave before 13th) cost would be £228.60.

We have some bursaries for Postgraduates from a QRA (Quaternary Research Association) award, and also bursaries from PAGES support for those coming from 'low GDP' and/or former Eastern Block countries - they will need to write a 300 word application outlining their expertise and why they need the money, i.e. make a case for support. If awarded support this will lower the costs from these groups.

In terms of themes, and given it is a workshop, we are keen for delegates to consider their data/thoughts in relation to the following topics:

Limnological functioning:

What can chironomids tell us about (palaeo) lake processes Eutrophication, stratification, changes in food-web structure (e.g. recent Quinlan paper in JoPL)?

Impacts on lakes:

Can chironomids help distinguish when a lake passes a critical transition? Do they show early warning signals (e.g. Wang et al Nature paper)?

If a lake is losing resilience, how do chironomids respond?

Transfer functions:

How do the current debates on use of transfer functions affect our training set data/research?

Are there aspects about chironomid data that help/hinder this debate (see Brooks et al. reply to Velle et al. in The Holocene).

Climate and other environmental reconstructions:

Following on from the above - how good are our chironomid inferred climate reconstructions?

Can we get better at estimating the real magnitude of change, and real errors?

A website for registration of participants will soon be set up at the University of Southampton. Please contact Peter G. Langdon (P.G.Langdon@soton.ac.uk) if you have any questions.

Finnish Chironomidae

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The current list of Chironomidae from Finland comprises 781 species altogether (Table 1). This includes 41 undescribed species, 12 ‘problematic’ species (cf. or ?), 1 exuvial and 4 larval types. Compared to the previous version from March 20th 2009, 1015 additions and 23 removals in the provincial records have been made.

Regional and national check-lists are often based on records from a large number of scientists with variable knowledge of both species distributions and taxonomy. For the list of species from Finland, over 90 % of the records are now my own identifications. These are mainly of own sweep-net samples (74%) from the period 1967 - 2012 and the massive whole-season Malaise-trap sampling by esp. Jukka Salmela during the last ten years (71% of all Malaise-trap samples). Some additional records come from collections of exuviae (mainly by Janne Raunio) and larvae (mainly by me). An overview of the sampling efforts divided by bioprovinces (Figure 1) and method is given in table 2.

The material is deposited mainly in my own collection consisting of thousands of slides and alcohol vials. The hypopygia of the undescribed spp. have been photographed and drawn. DNA barcoding using partial cytochrome c oxidase subunit 1 (COI) sequences has been done on 226 specimens of 77 species from southwest Finland, including two types in *Procladius choreus*-agg. and three types in *Chironomus pseudo-thummi*- agg. Papers giving summaries of the local fauna in the main habitat types are now in preparation.

Table 2. Number of species by sampling efforts in bioprovinces. 14 light-trap nights are included in the sweep-net samples. Most Malaise-traps were operated through the whole season. Only sampling lines/stations are regarded as larval samples.

Bioprov.	Species	Sweep-net	Malaise-traps	Emerg. traps	Exuvial samples	Larval sites
Al (A)	192	22	19			
Ab (V)	510	794	78	37	15	310
N (U)	442	134	42	12	14	187
Ka (EK)	378	163	3		30	40
St (St)	467	149	25	10	3	114
Ta (EH)	523	365	44	20	52	388
Sa (ES)	414	137	2		10	160
Kl (LK)	261	39			5	9
Oa (EP)	426	302	34	13	18	136
Tb (PH)	481	241	20		23	139
Sb (PS)	436	189	24			108
Kb (PK)	452	147	33	3		139
Om (KP)	415	159	5	4	16	136
Ok (Kn)	354	33	12			101
Oba (OP)	384	41	32			121
Obb (PeP)	411	33	32		5	87
Ks (Ks)	459	42	65			319
Lkoc (KiL)	466	186	45			82
Lkor (SoL)	444	52	26	18		78
Le (EnL)	494	137	55		1	52
Li (InL)	553	471	36	4	1	96
Total	781	3836	632	121	193	2802



Figure 1. The bioprovinces in Finland.

Table 1. Records of Chironomidae from Finland divided by bioprovinces from south to north (Al - Li). X = ident. by L. Paasivirta, x = ident. by others, M = collection of Museum of Zoology Helsinki (confirmed by Lauri Paasivirta), E = exuviae collected and identified by J. Raunio (mostly confirmed by Lauri Paasivirta).

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
Total number of species		192	510	442	378	467	523	414	261	426	481	436	452	415	354	384	411	459	466	444	494	553
Identified by L. Paasivirta (%)		95	99	92	92	98	95	96	90	91	99	97	97	92	99	98	97	98	98	92	96	97
Podonominae																						
<i>Boreochlus thienemanni</i> Edwards, 1938							X				X			X		X	X	X	X	X	X	X
<i>Lastodiamesa armata</i> Brundin, 1966							X				X	X		X		X	X	X				X
<i>L. gracilis</i> (Kieffer, 1924)			X	M			X		X		X	X	X						X	X	X	X
<i>L. sphagnicola</i> (Kieffer, 1925)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Parochilus kiefferi</i> (Garrett, 1925)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Trichotanypus mariae</i> Wirth & Sublette, 1970																			X			X
<i>T. positivalis</i> (Lundbeck, 1898)																	X	X	X	X	X	X
Tanypodinae																						
<i>Ablabesmyia longistyla</i> Fittkau, 1962		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>A. monilis</i> (Linnaeus, 1758)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>A. phatta</i> (Egger, 1863)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Anatopynia plumipes</i> (Fries, 1823)		X	X	x		x	X	X	x		X	X	X	x					X			X
<i>Apsectrotanypus trifascipennis</i> (Zetterstedt, 1838)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Arctopelopia barbaris</i> (Zetterstedt, 1850)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>A. griseipennis</i> (v.d. Wulp, 1858)		X	X	X	X		X	X													X	X
<i>A. melanosoma</i> (Goetghebuer, 1933)																					X	X
<i>Clinotanypus nervosus</i> (Meigen, 1818)		x	X	X	X	X	X	X	X	x	X	X	X	x		X		X				
<i>Conchapelopia aagaardi</i> Murray, 1987																				X	X	X
<i>C. hittmairorum</i> Michiels & Spies, 2002		X	X	X	X	X	X	X	E	E	X	X	E	E	X	E	E	X	X	X	X	X
<i>C. intermedia</i> Fittkau, 1962		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. melanops</i> (Meigen, 1818)		x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. pallidula</i> (Meigen, 1818)		X									X			E	X							
<i>Guttipelopia guttipennis</i> (v.d. Wulp, 1861)		X	X	X	X	X	X	X	X	X	X	X	X	X								X
<i>Hayesomyia tripunctata</i> (Goetghebuer, 1922)				X	X	X	X	X	X													
<i>Krenopelopia binotata</i> (Wiedemann, 1817)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>K. nigropunctata</i> (Staeger, 1839)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Labrundinia longipalpis</i> (Goetghebuer, 1921)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Larsia atrocincta</i> (Goetghebuer, 1942)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Macropelopia adlaucta</i> Kieffer, 1916		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. nebulosa</i> (Meigen, 1804)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. notata</i> (Meigen, 1818)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
<i>T. laeta</i> (Meigen, 1818)				X							X											
<i>T. lentiginosa</i> (Fries, 1823)		X	X	X	X	X	X		X	X		X		X	X	X	X	X	X	X	X	X
<i>T. pseudocarnica</i> Murray, 1976		X	X	X	X	X	X	X						X				X	X	X	X	X
<i>T. vitellina</i> (Kieffer, 1916)									X	X				X								
<i>Trissopelopia longimana</i> (Staeger, 1839)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Xenopelopia falcigera</i> (Kieffer, 1911)							E															
<i>X. nigricans</i> (Goetghebuer, 1927)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Zavrelimyia barbatipes</i> (Kieffer, 1911)	X	M	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Z. hirtimana</i> (Kieffer, 1918)		X	X	X	X	X	x															
<i>Z. melanura</i> (Meigen, 1804)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Diamesinae																						
<i>Diamesa aberrata</i> Lundbeck, 1889				X	X	X			X									M	x	X	X	X
<i>Diamesa arcica</i> (Boheman, 1865)																			X			X
<i>D. bertrami</i> Edwards, 1935																					X	X
<i>D. bohemani</i> Goetghebuer, 1932											X	X	X	X	X	X	X	X	X	X	X	X
<i>D. hyperborea</i> Holmgren, 1869																					X	X
<i>D. incallida</i> (Walker, 1856)				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>D. insignipes</i> Kieffer, 1908													X	X	X	X	X	X	X	X	X	X
<i>D. latitarsis</i> (Goetghebuer, 1921)																						X
<i>D. permacra</i> (Walker, 1856)	X			X												X	X					
<i>D. serratosioi</i> Willassen, 1985																						X
<i>D. tonsa</i> (Walker, 1856)																						X
<i>Pothisia gaedii</i> (Meigen, 1838)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. longimanus</i> (Kieffer, 1922)	X	X	X	X	X	X	X	E	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. pastoris</i> (Edwards, 1933)	X	X	X	X	X	X		M			X	X	X	X	X	X	X	X	X	X	X	X
<i>Protanypus caudatus</i> , Edwards, 1924	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x	x	X
<i>P. morio</i> (Zetterstedt, 1838)	X	x	E	X	X	X	X	X	X	X	X	X	X	x	X	X	X	x	X	X	X	X
<i>Pseudodiamesa branickii</i> (Nowicki, 1873)	X	X	M	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. nivosa</i> (Goetghebuer, 1928)							X									X	X		X	X	X	X
<i>Pseudokiefferiella parva</i> (Edwards, 1932)														X		X	X	X	X	X	X	X
<i>Symptothastia fulva</i> (Johannsen, 1921)																			X	X	X	X
<i>S. huldeni</i> Tuiskunen, 1986. 1																						X
<i>Syndiamesa</i> sp., indet. larv., 1a	X			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x
Telmatogoninae																						
<i>Telmatogeton japonicus</i> Tokunaga, 1935																						
Prodiamesinae																						
		E	E																			

Biogeographical province																					
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
	X	x		X	X	X	X	x	X	X	X	x	X	X	X	X	X	X	x	X	
<i>Monodiamesa bathyphila</i> (Kieffer, 1918)				X	X	X	X		X	X	X	x	X	X	X	X	X	X	x	X	
<i>M. ekmami</i> (Brundin, 1947)				X	X	X	X		X			X	X	X	X	X	X	X	X	X	
<i>Odontomesa fulva</i> (Kieffer, 1919)	X	X		X	x			X					X			X					
<i>Prodiamesa olivacea</i> (Meigen, 1818)	X	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	
Orthocladinae																					
<i>Aagaardia protensa</i> Saether, 2000	X	X	x	X	X	X	X		X	X	X	X		X			X	X		X	
<i>A. sivertseni</i> (Aagaard, 1979)	X		x		X				X	X	X	X	X	X	X	X	X	X	X	X	
pr. <i>Aagaardia</i> sp.1									X	X		X							X	X	
<i>Abiskomyia paravirgo</i> Goetghebuer, 1940									X	X	X		X	X	X	X	X	X	X	X	
<i>A. virgo</i> Edwards, 1937									X	X	X		X	X	X	X	X	X	x	x	
<i>Acamptocladius reissi</i> Cranston & Saether, 1982, 2				X	X				X	X	X		X	X			X	X	X	X	
<i>A. submontanus</i> (Edwards, 1932)	x		X	X		M			X	x	x		X	X	X	X	X	X	X	X	
<i>Acricotopus lucens</i> (Zetterstedt, 1850)	X	X	X	X	X	x		X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>Allocladius bothnicus</i> (Tuiskunen, 1984)														X						X	
<i>Boreosmittia inariensis</i> Tuiskunen, 1986	X	X		X	X	X	X	X	X		X		X	X	X	X	X	X	X	X	
<i>B. kareloborealis</i> Tuiskunen, 1986	X	X	x	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>Brillia bifida</i> Kieffer, 1909	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>B. longifurca</i> Kieffer, 1921, 2a	X	X	X	X	X	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	
<i>Bryophaenocladus aestivus</i> (Brundin, 1947)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x	X	
<i>B. dentatus</i> (Karl, 1937)																		X	X	X	
<i>B. flexidens</i> (Brundin, 1947), 3	X	X		X	X				X		X		X	X	X	X	X	X	X	X	
<i>B. ictericus</i> (Meigen, 1830)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>B. ilimbatus</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>B. cf. impectinus</i> Saether, 1976										X										X	
<i>B. inconstans</i> (Brundin, 1947)	X	X	X	X	X	X	X	X	X				X	X	X	X	X	X	X	X	
<i>B. cf. laticaudus</i> Saether, 1973	X								X								X	X	X	X	
<i>B. muscicola</i> Kieffer, 1906	X	X																			
<i>B. nidorum</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>B. nigrus</i> Albu, 1974	X																				
<i>B. nitidicollis</i> (Goetghebuer, 1913)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	M	X	X	
<i>B. pectinatus</i> Albu, 1974								X							X						
<i>B. psilacrus</i> Saether, 1982	X		M	X	X	X	X	X	X	X				X	X	X	X	X	X	X	
<i>B. saanae</i> Tuiskunen, 1986					X												X	X	X	X	
<i>B. scanicus</i> (Brundin, 1947)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>B. cf. sclerus</i> Wang & al., 2004				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>B. subparallelus</i> (Malloch, 1915)				X								X					X	X	X	X	

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
	<i>B. subvernalis</i> (Edwards, 1929)	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X		X	X
	<i>B. tuberculatus</i> (Edwards, 1929)	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X
	<i>B. vernalis</i> (Goetghebuer, 1921)										X											
	<i>B. xanthogyne</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	<i>B. sp. pt. xanthogyne</i>	X	M	X	X	X	X												X		X	X
	<i>B. sp. 1</i> "Syöte"	X	X	X	X	X	X				X			X		X	X	X	X			
	<i>B. sp. 2</i> "Syöte"															X		X				
	<i>B. sp. 3</i> "Malla"																					X
	<i>B. sp. 4</i> "Tsarmi"																	X			X	X
	<i>B. sp. 5</i> "Pallas"						X	X					X						X		X	X
	<i>B. sp. 6</i> "Rommas"															X						
	<i>Camptocladus stercorarius</i> (De Geer, 1776)	x	X	X	X	X	X	x	X	X	x	x	X	X	X	X	X	X	x	X		X
	<i>Cardiocladius capucinus</i> (Zetterstedt, 1850)	X	E	E	X	E,M	E		E	X	X	X		X						X		X
	<i>C. fuscus</i> Kieffer, 1924	X	X	E			E		E	X	X	X				X	X	X	X	X	X	X
	<i>Chaetocladus acuminatus</i> Brundin, 1956										X					X	X	X	X	X	X	X
	<i>C. binotatus</i> (Lundström, 1915)																		X		X	X
	<i>C. britae</i> Säwedal, 1976																				X	X
	<i>C. crassisaetosus</i> Tuiskunen, 1986																					X
	<i>C. dentiforceps</i> (Edwards, 1929), 4									X	X		X					X		X		X
	<i>C. dissipatus</i> (Edwards, 1929)			X		X	X		X	X	X	X	X			X	X	X		X		X
	<i>C. glacialis</i> (Lundström, 1915)												X								X	X
	<i>C. gracilis</i> Brundin, 1956, 4a	X	X	X									X			X	X	X	X	X	X	X
	<i>C. grandilobus</i> Brundin, 1956	X	X	M	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X
	<i>C. laminatus</i> Brundin, 1947	X	X	X	X	X	X	X	X	X	X	X				X	X	X	X	X	X	X
	<i>C. maeaeeri</i> Brundin, 1947																				X	
	<i>C. melaleucus</i> (Meigen, 1818)			X		X	X		X	X	X	X	X			X	X	X	X	X	X	X
	<i>C. sp. pt. melaleucus</i>	X	X	X					X	X											X	X
	<i>C. muliebris</i> Tuiskunen, 1986																				X	X
	<i>C. perennis</i> (Meigen, 1830)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	<i>C. piger</i> (Goetghebuer, 1913)	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X
	<i>C. cf. rusticus</i> (Goetghebuer, 1932)															X	X	X	X	X	X	X
	<i>C. suecicus</i> (Kieffer, 1916), 4b	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	<i>C. tenuistylus</i> Brundin, 1947															X	X	X			X	X
	<i>Clunio balticus</i> Heimbach, 1978	x	x	x	x	x	x															
	<i>Corynoneura arctica</i> Kieffer, 1923	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	<i>C. brundini</i> Hirvenoja & Hirvenoja, 1988	X					M						X	X	X	X	X	X	X	X	x	X

Biogeographical province																					
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
	X	X	X	X	X	M	X	X	X	X	X	X	X	X	X						
<i>C. carriana</i> Edwards, 1924																					
<i>C. celeripes</i> Winnertz, 1852	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. celtica</i> Edwards, 1924	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. coronata</i> Edwards, 1924	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. edwardsi</i> Brundin, 1949	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. fitkaui</i> Schlee, 1968	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. grattias</i> Schlee, 1968	X			X	X	X	X	X	X		X	X				X	X				
<i>C. gynocera</i> Tuiskunen, 1983																					x
<i>C. lacustris</i> Edwards, 1924	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. lobata</i> Edwards, 1924	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. magna</i> Brundin, 1949																					X
<i>C. scutellata</i> Winnertz, 1846	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. sp. 1</i> "Tvärminne"	X	x		X	X		X	X	X	X	X				X		X	X	X	X	X
<i>C. sp. 2</i>																					X
<i>Corynoneurella paludosa</i> Brundin, 1949				X	X	X	X	X	X	X	X				X	X	X	X	X	X	X
<i>Cricotopus (Cricotopus) albiforceps</i> (Kieffer, 1916)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. annulator</i> (Goetigebauer, 1927)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. bicinctus</i> (Meigen, 1918)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. caducus</i> Hirvenoja, 1973	X	X	x	X			X	X	X	X	X										
<i>C. coronatus</i> Hirvenoja, 1973					X	X		X	X		X					X	X	X	X	X	X
<i>C. cumulatus</i> Hirvenoja, 1973																					X
<i>C. cylindraceus</i> (Kieffer, 1908)	X		X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. ephippium</i> (Zetterstedt, 1838)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. festivellus</i> (Kieffer, 1906)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. flavocinctus</i> (Kieffer, 1924)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. fuscus</i> (Kieffer, 1909)	X	X	X	X	X	X	X	X			X	X			X						X
<i>C. magus</i> Hirvenoja, 1973					X																X
<i>C. pallidipes</i> Edwards, 1929																X					X
<i>C. patens</i> Hirvenoja, 1973	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. pilidorsum</i> Hirvenoja, 1973												X									X
<i>C. pilosellus</i> Brundin, 1956	X	x	X	X	X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. pirifer</i> Hirvenoja, 1973						X															x
<i>C. polaris</i> Kieffer, 1926	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. sp. pr. polaris</i>																					X
<i>C. pulchripes</i> Verrall, 1912								X								X					X
<i>C. septentrionalis</i> Hirvenoja, 1973					X			X				X	X	X	X	X	X	X	X	X	X

Biogeographical province																				
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
	X											X	X	x	?E	X	x		X	X
<i>C. similis</i> Goetghebuer, 1921												X	X	X		X	X	X	X	X
<i>C. slossonae</i> Malloch, 1915, 5a	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. tibialis</i> (Meigen, 1804)	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. tremulus</i> (Linnaeus, 1758)	X	X	X	X					X		X	X	X	X	X	X	X	X	X	X
<i>C. tritannulatus</i> (Macquart, 1826)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. trifascia</i> Edwards, 1929	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. tristis</i> Hirvenoja, 1973															E		X	X	X	X
<i>C. vierriensis</i> Goetghebuer, 1935	X	X																		
<i>C. villosus</i> Hirvenoja, 1973																		x	X	X
<i>C. (Isocladius) arcuatus</i> Hirvenoja, 1973	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x	X	X
<i>C. brevipalpis</i> Kieffer, 1909					x															
<i>C. interseclusus</i> (Staeger, 1839)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. laetus</i> Hirvenoja, 1973	X	X	x	X	X		x		X		X	X	X	X	X				X	X
<i>C. laricomalis</i> Edwards, 1932					X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. maurii</i> Spies & Saether, 2004, 5b	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. obnixus</i> (Walker, 1856)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. sp. cf. obnixus</i> Hirvenoja, 1973					X															
<i>C. obtusus</i> Hirvenoja, 1973	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. ornatus</i> (Meigen, 1818)	x	X	x					X												
<i>C. perniger</i> (Zetterstedt, 1850)				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. pilicauda</i> Hirvenoja, 1973																	x			X
<i>C. pilitarsis</i> (Zetterstedt, 1850)	X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. reductus</i> Hirvenoja, 1973																				x
<i>C. reticens</i> Hirvenoja, 1973				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. reversus</i> Hirvenoja, 1973				E	X	X	X	X	X	X	X	X	X	X	X	X	X	x	X	X
<i>C. suspiciosus</i> Hirvenoja, 1973					X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. sylvestris</i> (Fabricius, 1794)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. tricornatus</i> (Meigen, 1818)	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. trifasciatus</i> (Meigen, 1813)	X	X	X	X	x	X	X	X	X	X	X	X	X	X	E	X	X	X	X	X
<i>Cricotopus (Nostocloadius) lygropis</i> Edwards, 1929								X								X	X	X	X	X
<i>pr. Cricotopus sp. I</i> "Tapionaho"								X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Diplocladius cultriger</i> Kieffer, 1908	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Donricotopus dentatus</i> Tuiskunen, 1985				x					X											X
<i>Dramalia potamophylaxi</i> (Fittkau & Lellak, 1971)									X	X	X	X	X	X	X	X	X	X	X	x
<i>Epoiclocladius ephemeræ</i> (Kieffer, 1924)	X	X	x	x	X	X	x	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Eukiefferiella boeuvrensis</i> Brundin, 1956	X																			X

Biogeographical province																					
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. brevicar</i> (Kieffer, 1911)																					
<i>E. claripennis</i> (Lundbeck, 1898)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. clypeata</i> (Kieffer, 1923)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. devonica</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. ditimari</i> Lehmann, 1972	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. graciei</i> (Edwards, 1929)	X		X	X					X					X							
<i>E. sp.pr. graciei</i>														X	X						
<i>E. ilkeyensis</i> (Edwards, 1929)	X	X	X	X	X	X	E	E	E	E	E	E	E	X	X	X	X	X	X	X	X
<i>E. minor</i> (Edwards, 1929)														X	X	X	X	X	X	X	X
<i>Eurycnemus crassipes</i> (Panzer, 1813)														X	X	X	X	X	X	X	X
<i>Georhocladius luteicornis</i> (Goetghebuer, 1941)	X	x	X	X	X	E	X	X	X	X	x			X	X	X	X	X	X	x	
<i>G. platystylus</i> Saether & Sublette, 1983	X																				
<i>Gymnometrioctenus brunaldi</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>G. subnudus</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>G. volitans</i> (Goetghebuer, 1940), 6	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Halocladus variabilis</i> (Staeger, 1839)	X	X	X	X	X		X	X													
<i>Heleniella omaticollis</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>H.serratostioi</i> Ringe, 1976	X	X	X	X	X		X	X	X	X				X	X	X					
<i>Heterotanytarsus apicalis</i> (Kieffer, 1921)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>H. brun dini</i> Fittkau, 1956																	X			X	X
<i>H. sp. 1</i>																		X		X	X
<i>Heterorissocladius brun dini</i> Saether & Schnell, 1988																					x
<i>H. grinshtavi</i> (Edwards, 1929)	X	X	E	X	X	X	X	X	X	X	X	E	X	X	X	X	X	X	X	X	X
<i>H. maeaeeri</i> Brundin, 1949	X	X	X	X	X	X	X	X	X	X	X	x		X	X	X	X	X	X	X	X
<i>H. marcidus</i> (Walker, 1856)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>H. subpilosus</i> (Kieffer, 1911)	x								X	x	X	x				X	X	X	X	X	X
<i>H. sp.1</i> "Kolmperät"	X	M	M	X	M	X	X	X	X	X	X			X	X						
<i>Hydrobaenus conformis</i> (Holmgren, 1869)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>H. fuscistylus</i> (Goetghebuer, 1933)																X	X	X	X	X	X
<i>H. lapponicus</i> (Brundin, 1956)																X	X	X	X	X	X
<i>H. martini</i> Saether, 1976																					
<i>H. pilipes</i> (Malloch, 1915)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>H. spinnae</i> Saether, 1976		M				M										X	X	X	X	x	X
pt. <i>Hydrobaenus</i> sp.1																	X			X	X
<i>Hydrosmittia oxoniana</i> (Edwards, 1922)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>H. rutneri</i> Strenzke & Thienemann, 1942	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
<i>Krenosmittia boreoalpina</i> (Goetghebuer, 1944)		X	X			X	X				X							X	X	X		X	
<i>K. campophleps</i> (Edwards, 1929)																						X	
<i>K. halvorseni</i> (Cranston & Saether, 1986)		X				X	X					X				X	X	X	X	X	X	X	
<i>Lappokiefferiella platytarsus</i> Tuiskunen, 1986							X				X										X		
<i>Limmophyes aagaardi</i> Saether, 1990, 6b		X				X	X								X	X	X	X	X	X	X	X	
<i>L. angelicae</i> Saether, 1990		X					X	X				X		X		X		X					
<i>L. asquamatus</i> Andersen, 1937		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>L. bidumus</i> Saether, 1990																X	X	X	X	X	X	X	
<i>L. sp. pr. bidumus</i>																						X	
<i>L. brachytonus</i> (Kieffer, 1922)		X	X			X	X			X	X			X	X	X	X	X	X	X	X	X	
<i>L. difficilis</i> Brundin, 1947		X	X			X	X				X		X	X	X	X	X	X	X	X	X	X	
<i>L. edwardsi</i> Saether, 1990		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. er</i> Saether, 1985																						X	
<i>L. habilis</i> (Walker, 1856)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. margaretae</i> Saether, 1975																						X	
<i>L. minimus</i> (Meigen, 1818)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. natalensis</i> (Kieffer, 1914)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. ninae</i> Saether, 1990		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. pentaplastus</i> (Kieffer, 1921)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. pumilio</i> (Holmgren, 1869)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. sp. pr. pumilio</i>							X	X			X	X			X	X		X	X	X	X	X	
<i>L. schnelli</i> Saether, 1990																						X	
<i>L. spinigus</i> Saether, 1990		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>L. torulus</i> Saether, 1990																X	X	X	X	X	X	X	
<i>Mesocricotopus thiemanni</i> (Goetghebuer, 1940)		X	X			X	X	X			X	X	X	X	X	X	X	X	X	X	X	X	
<i>Mesosmittia flexuella</i> (Edwards, 1929)		X	X			X	X				X							X	X	X		X	
<i>Mertiocnemus acutus</i> Saether, 1995													X		X	X	X	X	X	X	X	X	
<i>M. albolineatus</i> (Meigen, 1818)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. atriclava</i> Kieffer, 1921		X	X	X	X	X	M				X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. beringensis</i> (Cranston & Oliver, 1988)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. caudigus</i> Saether, 1995		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. corticalis</i> Strenzke, 1950 s. Saether 1989						X					X	X		X								X	
<i>M. eurynotus</i> (Holmgren, 1883)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. exitiacis</i> Saether, 1995		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. fuscipes</i> (Meigen, 1818)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
<i>M. intergerivus</i> Saether, 1995							X															X	

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li		
<i>O. smolandicus</i> Brundin, 1947		X	X	X	X	X	X	X	X	X	X	X				X	X	X	X	X	X	X	X	
<i>Parachaetocladus abnobaicus</i> (Wulker, 1959)		X	X	X	X	X	X	X	X	X	X	X	X			X	X	X	X	X	X	X	X	
<i>Paracladius alpicola</i> (Zetterstedt, 1850)											X	X	X							X		X	X	
<i>P. conversus</i> (Walker, 1856)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X			X	X	X
<i>P. quadrimodosus</i> Hirvenoja, 1973								X			X						X	X	X			X	X	
<i>Paraticotopus niger</i> (Kieffer, 1912)		X	X	X	X	E	X	X	X	X	X			X	X							X	X	X
<i>P. uliginosus</i> (Brundin, 1947)	X	M				X	M		X	X	X	X	X	X	X		X	X	X	X	X	X	X	X
<i>Parakiefferiella bathophila</i> (Kieffer, 1912)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. bilobata</i> Tuiskunen, 1986						X								X										
<i>P. coronata</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. fennica</i> Tuiskunen, 1986			X	X	X	X	X	X	X		X	X	X	E			X					X	X	X
<i>P. finmarkica</i> Tuiskunen, 1986	X						X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. gynocera</i> (Edwards, 1937)						E	X	X	X		X	X	X			X	X	X	X	X	X	X	X	X
<i>P. minuta</i> Tuiskunen, 1986		X	X	X	X				X	X	X	X	X					X	X	X				
<i>P. nigra</i> Brundin, 1949						X	X	X	E				X	E			X					X	X	X
<i>P. scandica</i> Brundin, 1956						X	X	X	X		X	X	X						X	X		X	X	X
<i>P. smolandica</i> (Brundin, 1947)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. subaterrima</i> (Malloch, 1915)				X					X						X	X	X					X		
<i>Paralimnophyes longiseta</i> (Thienemann, 1911)	X	X	X	X	X	X	X	X	X	X	X	X	X	M		X	X	X	X	X	X	X	X	X
<i>Parametrioconemus borealpinus</i> Gouin, 1942															X			X				X	X	X
<i>P. stylatus</i> (Kieffer, 1924)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. sp. 1</i>																								X
<i>Paraphaenocladus exagitans</i> (Johannsen, 1905)		X	X	X	X	X	X	M		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. sp. pt. exagitans, 7a</i>																		X				X	X	X
<i>P. impensus</i> (Walker, 1856)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. intercedens</i> Brundin, 1947		X					X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. sp. pt. intercedens</i>																						X		X
<i>P. irritus</i> (Walker, 1856)		X	X	X	X	X	X	M		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. pseudirritus</i> Strenzke, 1950		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. triangulus</i> Saether & Wang, 1995																X								
<i>Parasmittia carinata</i> Strenzke, 1950	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. sp. 1 "Luukki"</i>		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Pararichocladus rufiventris</i> (Meigen, 1830)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. skirwithensis</i> (Edwards, 1929)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Paratrissocladus excerptus</i> (Walker, 1856)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Biogeographical province																				
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
	X		x	x	X	X		X	X	X										
<i>Propilocerus jacuticus</i> (Zvereva, 1950)			x	x	X	X		X	X	X										
<i>P. fl. komensis</i> (Zvereva, 1950)			x		X				X	X										
<i>P. saetheri</i> Wang, Liu & Paasivirta, 2007	X					X		X			X		X	X					X	X
<i>Prosmittia jemilandica</i> (Brundin, 1947)	X							X	X			M		M		X	M	X		X
<i>P. rectangularis</i> Tuiskunen, 1985																				x
<i>Psectrocladius</i> (<i>Allop.</i>) <i>conjungens</i> (Brundin, 1947), 8	X		X	X	X	X		X	X	X	X				X	X	X	X	X	X
<i>P. obivus</i> (Walker, 1856)	x	X	x	X	X	X		X	X	X	X	x		X	X	X	X	X	X	X
<i>P. platypus</i> (Edwards, 1929)	X			X	X	X		X	X	X	X			X	X	X	X	X	X	X
<i>P. (Mesop.) barbatus</i> Kieffer, 1923			E	X	X	X		X	X	X	X			X	X	X	X	X	X	X
<i>P. (Monop.) calcaratus</i> (Edwards, 1929)	X		x	X	X	X		X	X	X	X	X		X	X	X	X	X	X	X
<i>P. (Psectrocl.) barbimanus</i> (Edwards, 1929), 8a	X			x	X			X	X		X						X	X	X	X
<i>P. bisetus</i> Goetghebuer, 1942	X		M	X	X	X		X	X	X	X	X			X		X	X	X	X
<i>P. fennicus</i> Stora, 1939	X			X	X	X		X	X	X	X			X	X	X	X	X	X	X
<i>P. limbatellus</i> (Holmgren, 1869)	X		X	X	X	X		X	X	X	X	X		X	X	X	X	X	X	X
<i>P. octomaculatus</i> Wulker, 1956	X		X	X	X	X		X	X		X			X		X	X	X	X	X
<i>P. oligosetus</i> Wulker, 1956			M	X	X	X		X	X	X	X	X				X	X	X	X	X
<i>P. oxypura</i> Langton, 1985	X		E	X	X	X	E	X	X	X	X	X			X	X	X	X	X	X
<i>P. psilopterus</i> (Kieffer, 1906)	X		X	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X
<i>P. schliezni</i> Wulker, 1956	X		E	X	X	X		X	X	X	X	X		X	X	X	X	X	X	X
<i>P. sordidellus</i> (Zetterstedt, 1838)			X	X	X	E		X	X		X	X		X			M	X	X	X
<i>P. ventricosus</i> Kieffer, 1925	M	X	X	X	X									X	X					
<i>P. zetterstedti</i> Brundin, 1949	X			X	X				X	X	X							X		X
<i>Pseudorthocladius curtisylus</i> (Goetghebuer, 1921)	X		X	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X
<i>P. filiformis</i> (Kieffer, 1908)	X		X	X	X			X	X	X	X			X	X	X	X	X	X	X
<i>P. pilosipennis</i> Brundin, 1956	X		X	X	X	X		X	X	X	X	X		X	X	X	X	X	X	X
<i>Pseudosmittia albipennis</i> (Goetghebuer, 1921)	X			X	X															
<i>P. angusta</i> (Edwards, 1929)	X		X	X	X			X	X		M	X								X
<i>P. danconai</i> (Marzucci, 1947)	X		X		X			X					X	X				X		
<i>P. forcipata</i> (Goetghebuer, 1921)	X		X	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X
<i>P. sp. pr. forcipata</i>																				X
<i>P. gracilis</i> (Goetghebuer, 1913)			X	X				X	X	X	X	X		X	X	X	X	X	X	X
<i>P. mathildae</i> Albu, 1968						X			X			M		X	X	X	x			X
<i>P. obiusa</i> Strenzke, 1960	X	X	X	X	X	M		X	X	X	X	X		X	X	X	X	X	X	X
<i>P. trilobata</i> (Edwards, 1929)	X	X	M	X	X			X		X		X		X						
<i>Psilometriocnemus europaeus</i> Tuiskunen, 1985	X	X	X	X	X	x		X	X	X	X	X		X	X	X	X	X	X	X
<i>P. sp. I</i> "Tervola"	X														X					X

Biogeographical province																							
	Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li		
<i>Rheocricotopus atripes</i> (Kieffer, 1913)	X	X	X	X	X	X	X	X	X	X	X	X			X		X	X	X	X			
<i>R. chalybeatus</i> (Edwards, 1929)	X	X	X	X	X	X	X	X	X		X		X	X	X	X	X	X	X	X	X	X	
<i>R. chapmani</i> (Edwards, 1935)													X				X	X		X	X	X	
<i>R. effusus</i> (Walker, 1856)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>R. fuscipes</i> (Kieffer, 1909)	X	X	X	X	M					X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>R. glabricollis</i> (Meigen, 1830)												X			X	X	X	X	X	X	X	X	X
<i>R. redundus</i> Saether & Schnell, 1988										X													
<i>R. sp. pr. unidentatus</i> Saether & Schnell, 1988	X	X				X						X											
<i>Rheosmittia languida</i> (Brundin, 1956)															M								M
<i>R. spinicornis</i> (Brundin, 1956)	E										X		X	X	X	X	X	X	X	X	X	X	X
<i>Smittia alpilonga</i> Rossaro & Lencioni, 2002																							X
<i>S. cf. amoena</i> Caspers, 1988										X													
<i>S. aterrima</i> (Meigen, 1818)	X	X	X	X	X	X	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	X	X
<i>S. sp. pr. aterrita</i>																							X
<i>S. betuletorum</i> Edwards, 1941													X		X	X	X	X	X	X	X	X	X
<i>S. contingens</i> (Walker, 1856)											X												
<i>S. edwardsi</i> Goetghebuer, 1932	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>S. foliacea</i> (Kieffer, 1921)	X	X			X																		
<i>S. leucopogon</i> (Meigen, 1804)	X	X	X	X	X	X	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	X	X
<i>S. nudipennis</i> (Goetghebuer, 1913)	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>S. paranudipennis</i> Brundin, 1947	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>S. pratorum</i> (Goetghebuer, 1927)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>S. scutellosetosa</i> Caspers, 1988	X					X	M																
<i>S. stercoraria</i> Rossaro, 2000	X	X	X	X	X	X			X	X			X		X	X	X	X					X
<i>S. sp.1</i> s. Tuiskunen	X	X	X			X	M			X	X	X	X	X	X	X	X	X	X	X	X	X	x
<i>S. sp. pr. sp. 1</i>										X													
<i>S. sp. 2</i> "Jehkats"																							X
<i>Stabelbergina praeclara</i> Shilova & Zelentsov, 1978												X					X						
<i>Stiloladius intermedius</i> Wang, 1998																		X					X
<i>Synortholadius semivirens</i> (Kieffer, 1909)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Tavastia australis</i> Tuiskunen, 1985						X				X	X				X								X
<i>T. yggdrasilia</i> Brodin, Lundström & Paasivirta, 2008	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Thienemannia gracilis</i> Kieffer, 1909	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Thienemannia acuticornis</i> (Kieffer, 1912)	X	X	M	M	X	E		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>T. clavicornis</i> (Kieffer, 1911)																							X

Biogeographical province																				
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
	X	X	X	X	X	X					X	X	X	X	X	X	X	X	X	X
X	X			X	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	M		X	x		X	X	X	X	X	E	X	X	X	X	X	X	X	X
X				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	E	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	E		X	X	X		E	X	X	X	X	X	X	X	X	X	X	X	X
																X				X
											X	X	X	X	X			X	X	X
									X	X	X	X	X	X	X	X	X	X	X	X
									X	X	X	X	X	X	M		X		X	X
									X	X	X	X	X	X	X	M			X	X
									X											X
X	X	x	x		X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	x	x		X	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X
									X											
x	X	X	X	X	X	X	x	X	X	X	X	X	X	X	X	x	X	X	X	M
X	X	X	X	X	M		X	X	X	X	X	X	X							
X	X	X	X	X	X	X	X	X	X	X	x	X	X	X	X	X				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	x	x	X											
X	X	X	X	X	X	X	x	x												
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
<i>C. anthracinus</i> Zetterstedt, 1860		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. aprilius</i> Meigen, 1830		X	X	X	X	X		X		X													
<i>C. beljaninae</i> Wulker, 1991								x						x			X						
<i>C. brevidentatus</i> Hirvenoja & Michailova, 1998											x							X	X	X	X	X	X
<i>C. borokensis</i> Kerkis & al., 1988																							
<i>C. cingulatus</i> Meigen, 1830		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. clarus</i> Hirvenoja, 1962		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. coactaneus</i> Hirvenoja, 1998																							
<i>C. dorsalis</i> s. Strenzke, 1959				X	X	X				X		X	X					x	X	X	X	X	X
<i>C. entis</i> Shobanov, 1989		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x	X	X
<i>C. esai</i> Wulker, 1997							X	X		X	X			x									
<i>C. fraternus</i> Wulker, 1991							x			x				x				X					
<i>C. heteroplicicornis</i> Wulker, 1996			X				X			x	X			X				X					
<i>C. islandicus</i> (Kieffer, 1913) coll.									X														
<i>C. inermifrons</i> Goetghebuer, 1921						X	X	X	X	X	X	X	X						X	X	X	X	X
<i>C. jonnartini</i> Lindeberg, 1979, coll.	X	X	x			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. longistylus</i> Goetghebuer, 1921			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. lugubris</i> Zetterstedt, 1850													x										x
<i>C. luridus</i> Strenzke, 1959		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. melanescens</i> Keyl, 1962						X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. melanotus</i> Keyl, 1961		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. muratensis</i> Ryser & al., 1983		x					x				x												
<i>C. neocorax</i> Wulker & Butler, 1983		X	x	x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. piger</i> Strenzke, 1959		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. pilicornis</i> (Fabricius, 1787)							X			x	X		X	X	X	X	X	X	X	X	X	X	X
<i>C. plumosus</i> (Linnaeus, 1758)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. pseudothummi</i> Strenzke, 1959, coll.		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. riihimakiensis</i> Wulker, 1973		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. riparius</i> Meigen, 1804		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. salinarius</i> Kieffer, 1915		X																					
<i>C. saxatilis</i> Wulker & al., 1981				X				X															
<i>C. saxatilis</i> agg.		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. sollicitus</i> Hirvenoja, 1962		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. sororius</i> Wulker, 1973										x													
<i>C.?staegeri</i> Lundbeck, 1898													X					x					x
<i>C. tenuistylus</i> Brundin, 1949		X			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Biogeographical province												Li								
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X			
<i>C. (Lobochironomus) dorsalis</i> Meigen, 1818, 9																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x		
<i>C. improvidus</i> Hirvenoja, 1998																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. mendax</i> Storå, 1937																				
	X	X	X	X	X	X	X	x	X	X	X	X	X	X	X	x	X	X	X	X
<i>C. storai</i> Goetghebuer, 1937																				
	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X	x	X	X	X	X
<i>Cladopelma bicarinatum</i> (Brundin, 1947)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. edwardsi</i> (Kruseman, 1933)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. goetghebueri</i> Spies & Saether, 2004, 10																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. virescens</i> (Meigen, 1818)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. viridulum</i> (Linnaeus, 1767)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Cryptochironomus albofasciatus</i> (Staeger, 1839)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. denticulatus</i> Goetghebuer, 1921																				
	X	X	X	X	X	X	E	E	X	X	E	X	X	X	X	X	X	X	X	X
<i>C. obreptans</i> (Walker, 1856)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. psittacinus</i> (Meigen, 1830)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. redekei</i> (Kruseman, 1933)																				
	X	X	X	X	X	X	X	X	X	X	M	X	X	X	X	X	X	X	X	X
<i>C. rostratus</i> Kieffer, 1921																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>C. supplicans</i> (Meigen, 1830)																				
	X	X	X	X	X	X	X	X	X	X	x	X	X	X	X	X	X	X	X	X
<i>C. ussouriensis</i> (Goetghebuer, 1933)																				
<i>Cryptotendipes darbyi</i> (Sublette, 1960)																				
<i>C. pflugfelderi</i> Reiss, 1964																				
		M			E															
<i>C. pseudotener</i> (Goetghebuer, 1922)																				
		E																		
<i>C. usmaensis</i> (Pagast, 1931), 11																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Demejerea rufipes</i> (Linnaeus, 1761)																				
x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	x	X	X	X
<i>Demicryptochironomus</i> (<i>D.</i>) <i>vulneratus</i> (Zetterstedt, 1838)																				
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>D. sp. Pe 1 s. Langton</i> , 1991																				
		E			E		E	E				E	E	E	E					
<i>D. (Irmakia) neglectus</i> Reiss, 1988																				
		E			E															
<i>Dicrotendipes lobiger</i> (Kieffer, 1921)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	M	X	X	X	X	X	X
<i>D. nervosus</i> (Staeger, 1839)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>D. notatus</i> (Meigen, 1818)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>D. pulsus</i> (Walker, 1856)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>D. tritonus</i> (Kieffer, 1916)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Einfeldia pagana</i> (Meigen, 1838)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. pectoralis</i> (Kieffer, 1924), s. Hirvenoja 1998																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Endochironomus albipennis</i> (Meigen, 1830)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. stackelbergi</i> Goetghebuer, 1935																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>E. tendens</i> (Fabricius, 1775)																				
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Glyptotendipes aequalis</i> (Kieffer, 1922)																				

Biogeographical province																					
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
			X	x																	
	X		X	X	X	X	E	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	X	x	x	x	X	X	X	x	X	X	X	x	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	x	X	X	X	E				X	X			X							
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
	X	M	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
x	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	M	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
X	X	M	E	X	M	E															

Biogeographical province																				
Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li
	X				X	X			X	X	X	X		X	X	X	X	X	x	X
<i>C. vanderwulpi</i> (Edwards, 1929)	X	X	X	X	X	X	E	X		X	X	X	X		x	X	X		X	X
<i>Constempellina brevicosta</i> (Edwards, 1937)	X	E	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Corynocera ambigua</i> Zetterstedt, 1838	X	x		X	X	X	X	x	X	x	X		X	X	X	X	X	X	x	x
<i>C. oliveri</i> Lindeberg, 1970					X	X			X										x	x
<i>Microspectra appendica</i> Stur & Ekrem, 2006	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. apposita</i> (Walker, 1856)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. atrofasciata</i> (Kieffer, 1911)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. attenuata</i> Reiss, 1969	X	X		X	X	X		X	X	X	X									
<i>M. californis</i> Stur & Ekrem, 2006					X	X											X	X	X	X
<i>M. chionophila</i> (Edwards, 1933)	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. insignilobus</i> Kieffer, 1924	X		E		X	X	X		X	X	X	x				X		X	X	X
<i>M. junci</i> (Meigen, 1818)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. klinki</i> Stur & Ekrem, 2006	X	X	X	X	X	X		X	X	X	X		X	X	X	X	X	X	X	X
<i>M. lacustris</i> Säwedal, 1975					X	X			X	X	X	X	X	X	X	X	X	X	X	X
<i>M. lindebergi</i> Säwedal, 1976	X				X	X			X	X	X	X	X	X	X	X	X	X	X	X
<i>M. logani</i> (Johannsen, 1928)			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. malla</i> Gilka & Paasivirta, 2008					X	X								X						
<i>M. nana</i> (Meigen, 1818)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. notescens</i> (Walker, 1856)	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. pallidula</i> (Meigen, 1830)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. radialis</i> Goetghebuer, 1939		X			X	X			X	X	X					X	X	X	X	X
<i>M. recurvata</i> Goetghebuer, 1928	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. ritensis</i> Gilka, 2001	X	X	X	X	X	X		X	X	X	X					X				
<i>M. roseiventris</i> (Kieffer, 1909)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. schrankelae</i> Stur & Ekrem, 2006																	X			
<i>M. sofiae</i> Stur & Ekrem, 2006	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X
<i>M. snyiata</i> Reiss, 1969	X																X			
<i>Neozavrelia cuneipennis</i> (Edwards, 1929), 17				E		M														
<i>Paratanytarsus abiskoensis</i> Reiss & Säwedal, 1981																X		X		X
<i>P. austriacus</i> (Kieffer, 1924)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. bituberculatus</i> (Edwards, 1929)				X																
<i>P. dimorphis</i> Reiss, 1965															X	X				
<i>P. dissimilis</i> Johannsen, 1905	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>P. grimmii</i> (Schneider, 1885)		x	X				E													
<i>P. hyperboreus</i> Brundin, 1949								X								X	X	X	X	X

Biogeographical province		Al	Ab	N	Ka	St	Ta	Sa	Kl	Oa	Tb	Sb	Kb	Om	Ok	Oba	Obb	Ks	Lkoc	Lkor	Le	Li	
<i>T. trux</i> Gilka & Paasivirta, 2007																							X
<i>T. usmaensis</i> Pagast, 1931		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>T. verralli</i> Goetghebuer, 1928		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>T. volgensis</i> Missetko, 1967		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
<i>Virgatantarsus arduennensis</i> (Goetghebuer, 1922)		X	E	X	X	E	E	E	E	E	X	X	E	E				X	X	X	X	X	X
<i>Zavrelia pentatoma</i> Kieffer, 1913		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Notes: **1:** ?syn. with *takatenis* (Tokunaga, 1936) (Makarchenko 1994); **1a:** mostly hardly separable from *Pseudokiefferiella*; **2:** ?syn. with *submontanus*; **2a:** ?syn. with *flavifrons* (Johannsen, 1905). The differences in the colour and AR of the species (Sublette 1967) may be due to the water temperature (different generations); **3:** might be earlier confused with *subvernalis*; **4:** obviously belongs to *Zalutschia*; **4a:** ?syn. with *dissipates*; **4b:** includes just splitted *longivirgatus* Stur & Spies, 2011 (Stur & Spies 2011); **5a:** syn. *claripes* Hirvenoja, 1973; **5b:** syn. *polychaetus* Hirvenoja, 1989 (Spies & Sæther 2004); **6:** ?syn. *brumalis*; **6b:** partly masked by *minimus*; **7:** repl. *bicolor* (Zeiterstedt, 1838) (Spies & Sæther 2004), includes colour form *anderseni* Sæther, 1977; **7a:** see Cranston, Oliver & Sæther (1989); Fig. 9.58 sp. D and F; **8:** ?syn. with *platypus*; **8a:** in the provinces Lkoc-Li hypopygium resembles *fennicus*; **8b:** syn. *Chironomus (Loboch.) dissidens* (Walker, 1856); **9:** syn. *longipes* Staeger, 1839; **10:** syn. *lateralis* (Goetghebuer, 1934); **11:** ?syn. *nigronitens* (Edwards, 1929); **12:** syn. *gripekoveni* (Kieffer, 1913); **13:** syn. *mancunianus* (Edwards, 1929); **14:** hypopygium separable from *imbecilis* s. Langton & Pinder 2007; **14a:** syn. *arcuatus* (Goetghebuer, 1919); **15:** syn. *nudisquama* (Edwards, 1929) and *transcaucasicus* Chernovski, 1949; **16:** syn. *uncinatum* (Goetghebuer, 1921) (Dettinger-Klemm 2002); **16a:** syn. *wexionensis* Brundin, 1947 (Gilka 2011); **17:** syn. *longappendiculata* Albu, 1980 (Ekrem, 2006); **18:** syn. *Stempellinella distinctissimus* Brundin, 1947; **19:** replaces *minor* (Edwards, 1929) (Spies & Sæther 2004); **20:** syn. *deceptus* Lindeberg, 1967 and *palmeni* Lindeberg, 1967 and *socialis* Lindeberg, 1967 (Ekrem 2004); **21:** syn. *simulans* Lindeberg, 1967 (Ekrem 2004).

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