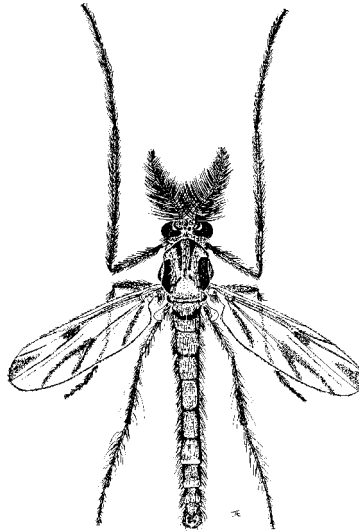


IDENTIFICATION MANUAL
FOR THE
LARVAL CHIRONOMIDAE (DIPTERA)
OF
NORTH AND SOUTH CAROLINA



John H. Epler, Ph.D.

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INTRODUCTION

and key to subfamilies

1



HIRONOMIDAE - a word that brings either a smile, a groan or a look of terror, or perhaps all! Chironomidae - a fantastically diverse large group of small flies whose larvae inhabit just about every niche possible in most freshwater aquatic ecosystems - not to mention marine and terrestrial forms.

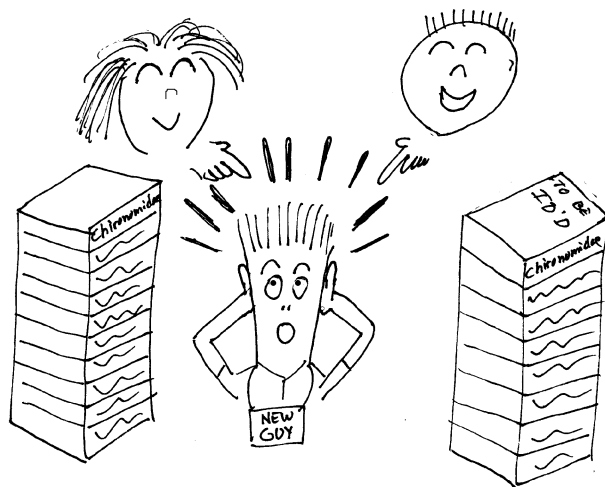
As a benthologist, at one time or another you will have some sort of interaction with Chironomidae. Whether it is a pleasant or unpleasant encounter may depend on how well you feel about and what you know about the organisms with which you are working. Yes, identifying chironomids can be a daunting task, but it is possible - sometimes even easy! - and the amount of information one can gather can be prodigious. Too many studies list Chironomidae only at the family level; identification at the generic level introduces many more data, while species-level identifications provide the most data, especially when biodiversity is an issue.

The Chironomidae typically have been shunned by many benthologists because of perceived difficulties in specimen preparation, identification, taxonomy, morphology and literature.

The Family Chironomidae

The Chironomidae are a relatively primitive (phylogenetically speaking) group of flies (Diptera) in the suborder Nematocera. Commonly called non-biting midges, or “blind mosquitoes”, as adults and “bloodworms” as larvae, chironomids are closely related to mosquitoes (Culicidae) and biting midges (Ceratopogonidae). Unlike their nasty relatives, female chironomids do not bite!

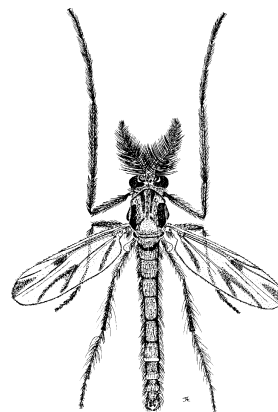
The Chironomidae are usually the most abundant macroinvertebrate group, in numbers of species and individuals, encountered in the majority of freshwater aquatic habitats. In addition, chironomids



Hey - we've got a new guy in the lab! Let's make him do the chironomid identifications!!!

Depending on the taxa, identification can be easy or perhaps difficult; I've examined collections of Chironomidae that were basically 100% correctly identified, but I've also seen some collections with as many as 65% of the specimens misidentified!

Hopefully, this manual will make your life with Chironomidae easier. It is possible that, **with experience**, you will eventually be able to identify most chironomids to genus, or even species. Note that some identifications can be done while the larvae are still in fluid preservative (BUT only with much experience!!!!)



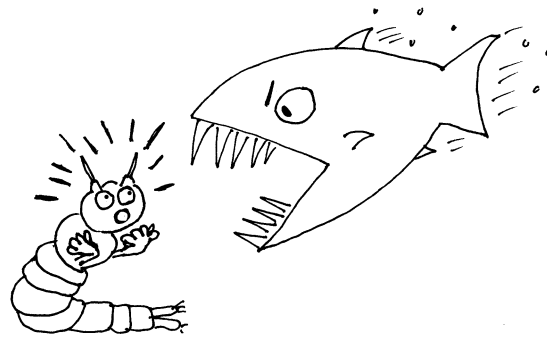
An adult midge, Dicotendipes thanatogratus

have invaded the sea, being found along coastlines world wide and occurring at least 30 m down in the ocean, and the land, where they may be encoun-

tered in a corn field or in dry hardwood forest litter. They occur on all continents - chironomids are the only free-living holometabolous (meaning with complete metamorphosis; a four stage life cycle) insects to do so - and are found living from heights of 5600 m on glaciers in Nepal down to depths of over 1000 m in Lake Baikal. Chironomid larvae, pupae and adults form an integral part of the food web, serving as food for larger invertebrates, fish, amphibians and birds. Many larvae possess giant chromosomes and have been used extensively in genetic research. Chironomid adults are considered nuisances when large emergences occur in close proximity to human habitations. They have also been implicated in allergenic reactions in humans (see Ali (1991) and Armitage et al. (1995) for an overview of pestiferous Chironomidae). Chironomids are recorded as pests in rice fields, where the larvae mine the leaves and eat the seeds and seedlings. In somewhat of a turnabout, relatives of these pest species (mostly members of the genus *Cricotopus*) may find a use in biological control of nuisance aquatic plants in the southern US.

However, to benthologists, the Chironomidae have long been known as potential indicators of water quality. Some groups of genera and/or species are known to inhabit water of high quality; others are well known dwellers in water of poor quality. Unfortunately, many of the larvae have been (and some still are) very difficult to identify, and much of the literature is burdened with studies done with Chironomidae that were misidentified.

Some of the confusion is due to the complexity of the taxonomy of the family. The Chironomidae have suffered a "double whammy" of sorts: a) their names were confusing due to changes necessitated by the Code of Zoological Nomenclature at the time, and b) there were two systems of classification, one based on adults, the other on immature stages. Ashe (1983) gives an excellent review of the taxonomic problems the Chironomidae have gone through. Today these differences have been largely reconciled because knowledgeable workers utilize characters from all life stages to separate species and delimit



Chironomid larvae, pupae and adults form an integral part of the food web, serving as food for larger invertebrates, fish, amphibians and birds.

genera. A comprehensive update of our knowledge of the biology and ecology of the Chironomidae has recently been published (Armitage et al. 1995). Identifications of chironomid larvae became much more realistic in 1983 when the first volume of the Entomologica Scandinavica "Holarctic Keys", dealing with larvae, was published (Wiederholm 1983). A pupal volume was produced three years later (Wiederholm 1986), followed by the adult volume (Wiederholm 1989). The volumes combine keys, excellent illustrations and, most importantly, diagnoses for each genus. (A diagnosis is a short description of the characters of a taxon that will separate it from other similar taxa.) However, many new genera and previously unknown larvae have been described and some previously described genera have been reorganized since the publication of the larval volume. These books remain a necessity for chironomid workers but must be supplemented with more recent literature.

The family is divided into 11 subfamilies, seven of which occur in North America. Two of the subfamilies, Telmatogetoninae and Podonominae, are relatively restricted in habitat; two other subfamilies, the Diamesinae and Prodiamesinae, are, depending on your locality, relatively uncommon. The majority of Chironomidae you will encounter will probably be members of the subfamilies Tanypodinae, Orthoclaadiinae and Chironominae.

How to use this manual

Area covered: This manual was written for use in the states of North and South Carolina, and will identify all genera known to me from these states. In actuality, this manual should identify larvae of most genera and species encountered on what is commonly called the Southeastern Coastal Plain. States covered include Alabama, Florida, Georgia, eastern Kentucky, Mississippi, North and South Carolina, eastern Tennessee and most of Delaware, Pennsylvania and Virginia. The manual should be useful for most of the eastern United States, with the caveat that the further that one is from the Southeast, the less effective the manual will be.

Illustrations and abbreviations: The majority of the illustrations in this manual were produced by the author from southeastern US specimens, most of which were reared or otherwise associated. Some are somewhat schematic in that all parts of a structure are not drawn. For example, in the Chironominae, often only one ventromental plate is drawn, and only a portion of the ventromental striae are shown; premandibular brushes are often not drawn unless they are an important character, and the pecten mandibularis is not fully drawn on most mandibles. When specimens were unavailable or not suitable for illustration, figures were borrowed from other sources. Thus, some illustrations differ in the amount of shading, structures included, etc. If the illustrations were from publications other than my own, the source of each figure is cited at least once within the manual.

Abbreviations used are explained in the Glossary that begins on page 1.10.

Taxonomy: In general, I have not used the author's name(s) for genera and species within the text and keys; complete names are listed in chapter 10. For arrangement of tribes and subgenera, see Caldwell et al. (1997) or Oliver et al. (1990).

Many larvae are undescribed or unassociated with the adult stages. Species definitions in the Chironomidae are, for the most part, based on the adult male. Several undescribed larval "types" are known on the genus and species level. These have

been given letter or number designators, such as "*Tanytarsus* sp. A" or "Chironomini genus III". These may represent taxa with described adults, or species new to science. When reared or otherwise associated with an identifiable life stage, the names can be updated. Many new, undescribed species are included in this manual. However, a manual such as this is not the proper place to publish new names and descriptions. Thus, as is noted in the text, these new species will be described in papers currently in progress. However, the following changes are proposed in this manual:

Corynoneura taris Roback is considered a junior synonym of *C. lobata* Edwards

Einfeldia austini (Beck & Beck) is moved to *Chironomus* and is now *Ch. (Lobochironomus) austini* (Beck & Beck).

The Layout: This manual is divided into ten chapters. This introduction is the first chapter, followed by seven subfamily chapters, which are then followed by a Bibliography and a checklist of the Chironomidae of North and South Carolina. The subfamily chapters are arranged phylogenetically; chapters are paginated separately. Each subfamily chapter has a key to genera which is followed by "generic units" in alphabetical order. Undescribed genera are at the end of each chapter. Each genus unit consists of several parts:

A **Diagnosis**, or short descriptive summary of the genus' morphological characters which will separate it from similar taxa. Although this manual is intended for stand-alone use, it will be most effective when used in conjunction with the more detailed diagnoses in Wiederholm (1983). **Note that the diagnoses in this manual pertain to character states of southeastern taxa!** Some genera include species that are different from those in the Southeast. For instance, *Dicrotendipes lobiger*, a species not known to occur in the Southeast, has a frontoclypeal apotome - all southeastern *Dicrotendipes* have a frontal apotome.

A **Notes** section which contains additional information concerning the taxonomy and biology of the genus.

An **Additional References** section lists additional literature that may give more information. *Wiederholm (1983, 1986, 1989) is always considered to be an additional reference.*

Illustrations of important body structures are included for each genus; a **Key** to species and a **Notes on species** section are included when possible.

The Keys: *The keys are written for fourth instar larvae!* Measurements are only valid for fourth instar larvae, but ratios may still be useful for other instars. Illustrations are usually arranged from left to right and/or top to bottom with regards to the order of statements in the couplet(s). If you are new to chironomids, you'll have to start with the key for subfamilies that starts on page 1.41 at the end of this chapter. Key your larva to subfamily, then go to the subfamily chapter. There you may key your larva to genus. If it fits, then key your specimen to species, if a species key is available for that genus. When (if!) you get to the species, check to see if there are additional notes ("Notes on species") concerning that species.

How to use a dichotomous key: The identification keys used in this manual are dichotomous keys. Never used a dichotomous key before? Read on!

A dichotomous key is basically a series of either/or statements ("dichotomous" means "dividing into two parts"). One runs through the statements making choices and eventually should end up with an answer or "identification". For example, let's say you have 4 objects – a red triangle named Phil, a white square called Bobby, a white circle named Mickey and a red circle named Jerry. Here's how a key would work to identify them. The first couplet (group of choices) would read:

1	Object red in color	2
1'	Object white in color	3

Here you have a choice – your object is either red or white. If it's red, go to couplet "2"; if it's white go to couplet "3". When we go to "2" we see:

2(1)	Object triangular	Phil
2'	Object circular	Jerry

What's that "1" doing in parentheses after the "2", you may ask? That's the number of the couplet that led you to where you are now! In a long key, you can use these numbers to trace your steps back. Sometimes you may have to venture forward in a key when you're not sure which couplet fits your taxon, i.e., try going both ways from a couplet. With the parenthetical numbers, it's easier to retrace your steps. You can even start at an endpoint in the key (your "identification") and run through the key backwards!

The key ends with:

3(1)	Object square	Bobby
3'	Object circular	Mickey

Now you may think you've identified your objects, but now comes a very important part – you must check your "identification" against a diagnosis (a brief synopsis of the characters that distinguish your taxon from others that may resemble it) or a description (a full blown listing of the characters of the taxon). Pictures may also help, but pictures can also be a trap into which you can easily fall. Why? Because the picture may be of a structure that is similar in several organisms and may not illustrate a definitive character for your organism. When Wiederholm (1983) first appeared, featuring figures of many Palaearctic species, there was an increase in "records" of those species from the US. Hmmm ...

Remember that if you start to key something, that something will end up somewhere in the key – but that doesn't mean you've identified it correctly. If you go to key a larva to species in the wrong genus key, sure, you'll end up with an identification – but it'll be wrong (trust me – I've seen this a lot!!). Another thing to remember about using a key – don't insert information that is not specifically put forth in the key at the point in the key at which you are working; i.e., answer only the question(s) in that specific couplet.

And, most importantly, you must have your identifications verified by a qualified expert! Be sure to read the sections on "Identifying Chironomidae" (page 1.14) and "Quality Assurance" (page 1.28)

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(Clemson University), Peggy Morgan (FDEP, Tampa), John Morse (Clemson University), Steve Moulton, (USGS, Denver, CO), Chuck Parker (Great Smoky Mountains National Park), Bill and Jan Peters (Florida A&M University), Bob Rutter (FDEP, Punta Gorda), Wolfgang Schacht (Zoologische Staatssammlung, Munich, Germany), Michelle Scharer (University of Puerto Rico), Marty Sneen (EA Engineering, Science, & Technology, Inc., Deerfield, IL), Martin Spies (Zoologische Staatssammlung, Munich, Germany), Doug Strom (FDEP, Port St. Lucie), David Wahl (American Entomological Institute, Gainesville, FL), Harold Walther (Canadian National Collection), Ford Walton, Charles Watson, Cliff Webber (Auburn University), Tom Wilda and Doug Wymer (Tennessee Tech University) for the loan, gift or opportunity to examine some of the specimens utilized in this study.

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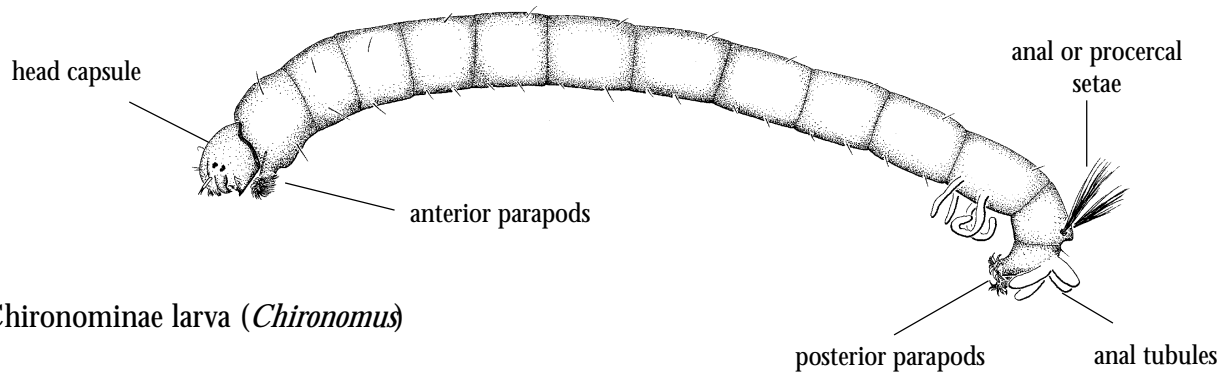
Judy and Barry Merrill (Merrill Consultants, Dallas, TX) supplied much of the funding for my laboratory and computer equipment- thanks again, guys! Their generosity has made it far easier to accomplish the gargantuan task of producing this manual.

My wife Linda helped secure funding for this project, has handled the administrative tasks, proofed text, prepared the table of contents and has put up with the isolation that results from my being almost totally involved with the production of this manual for the last four years. I am forever grateful for her help, love and understanding.

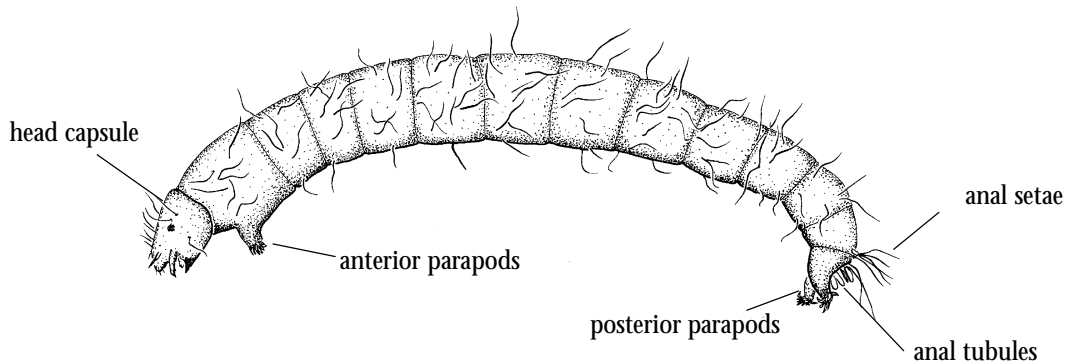
Morphology

Another component of the complexity of the Chironomidae is their morphology. Just as many species have gone under several different synonyms, many anatomical structures have often gone under several names. For example, the mentum has been called the hypochilum, hypostoma, hypostomium, hypostomial plate, labium, or labial plate. Many of these changes in structure names were evolutionary, due to our increased knowledge of chironomid morphology. Sæther (1980a) produced a glossary of chironomid morphology terminology that is largely followed today.

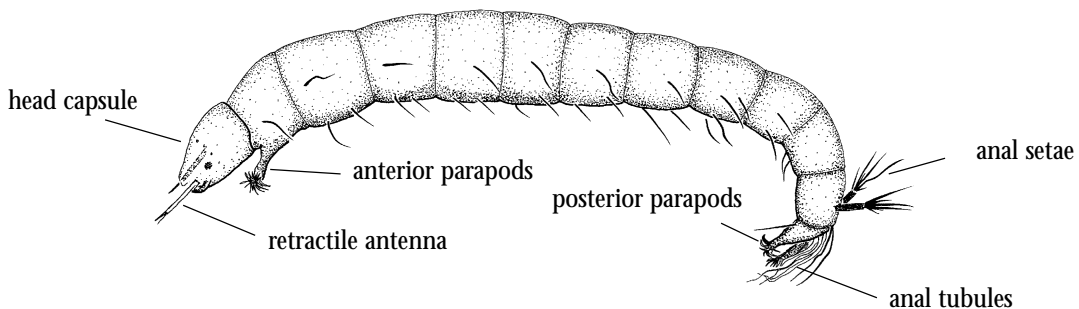
Chironomid larvae bear a sclerotized, non-retractile head capsule, with opposing mandibles, on a narrow, cylindrical body. There usually is one pair of unjointed anterior parapods ("prolegs") on the first body segment, one pair of unjointed posterior parapods on the last body segment, which also bears a pair of setae-bearing procerci, and one to three (usually two) pairs of anal tubules. Some terrestrial chironomids and others living in specialized environments have lost one or both pairs of parapods. There are usually no spiracles, except in some members of the subfamily Podonominae. There are four larval instars (the larvae sheds its skin four times before pupating).



Chironominae larva (*Chironomus*)



Orthoclaadiinae larva (*Tvetenia*)

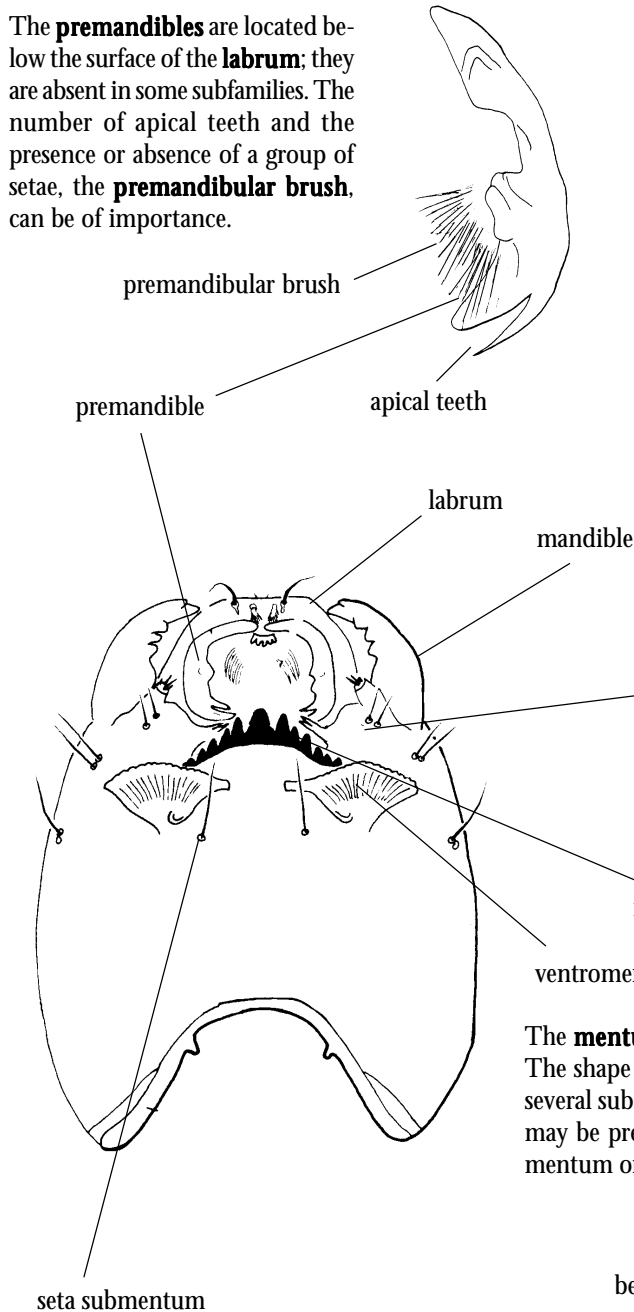


Tanypodinae larva (*Pentaneura*)

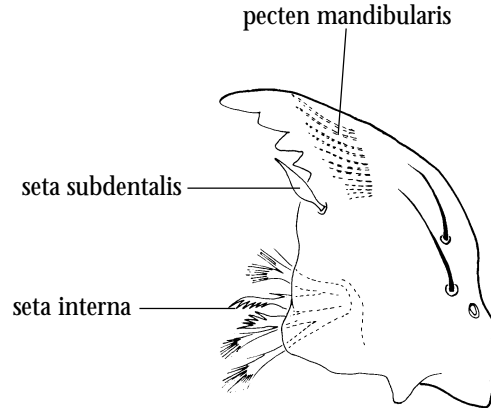
The Head Capsule

The majority of the characters used for larval identification are found on the sclerotized head capsule, with most of the more easily found characters located on the ventral side of the head. Below is a ventral view of the head capsule of *Dicrotendipes*, a typical member of the subfamily Chironominae; most other Nearctic subfamilies are generally similar, except for the Tanypodinae (see page 6).

The **premandibles** are located below the surface of the **labrum**; they are absent in some subfamilies. The number of apical teeth and the presence or absence of a group of setae, the **premandibular brush**, can be of importance.

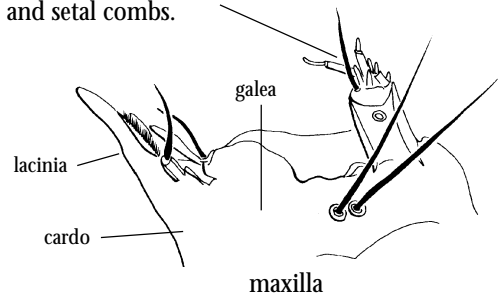


The placement and/or shape of a pair of setae posterior to the mentum, the **setae submenti**, are of importance in some taxa.

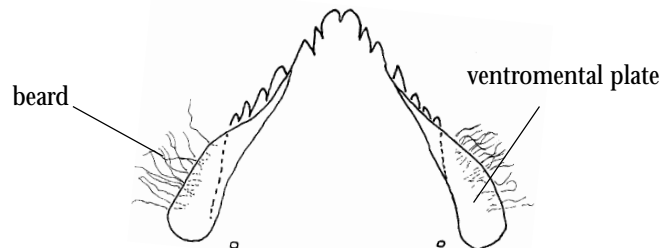


Many characters useful in identification are located on the **mandible**. These include the number and shape of the inner, apical and dorsal teeth, the presence or absence of a **seta interna**, the morphology of the **seta subdentalis** and the **pecten mandibularis**.

The **maxilla** bears structures useful in identification, such as the **maxillary palp** and various setae and setal combs.

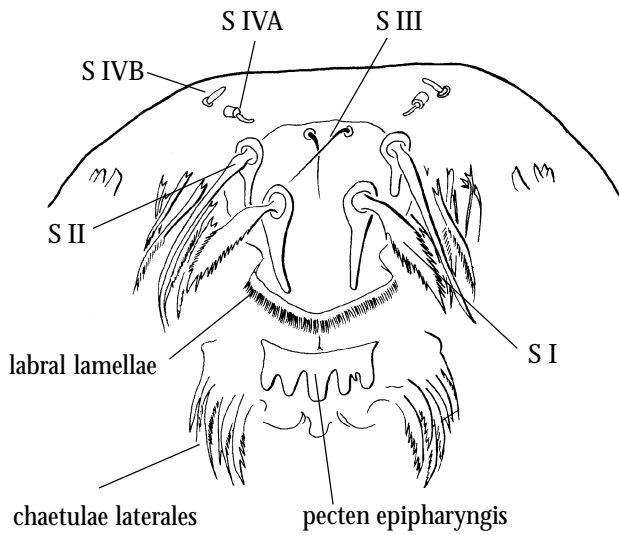


The **mentum** is often one of the most noticeable structures of the head capsule. The shape and number of teeth can be of great importance in identification. In several subfamilies, ventromental plates are present. A group of setae, the **beard**, may be present under or near the ventromental plates and/or the margin of the mentum or the maxilla.



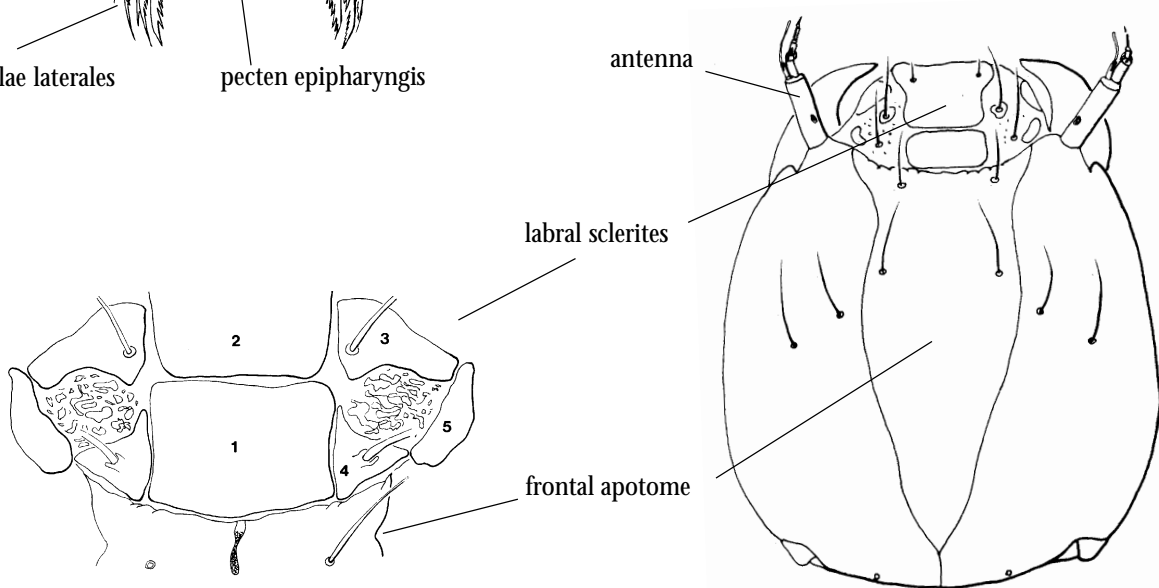
Mentum of an Orthoclaudiinae larva, *Rheocricotopus robacki*, showing beard setae beneath ventromental plates.

Dorsal view of the labrum



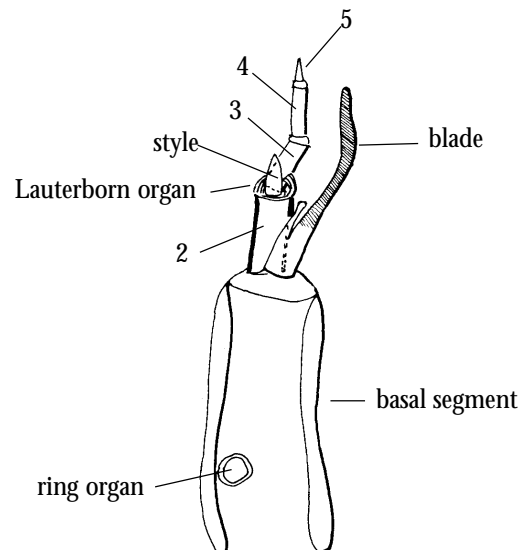
Although located on the dorsum of the head capsule, the labrum is usually folded under in slide mounts and is most often viewed "dorsally" in a ventral aspect. Several very important setae and other structures are located on or near the labrum - the **S setae**, the **labral lamellae** and the **pecten epipharyngis**.

Dorsal view of the head capsule



Posterior to the labrum are the **labral sclerites**. All of the labral sclerites figured above are not always present, dependent upon various degrees of fusion of the sclerites. Labral sclerite 1 is often fused with the apotome, forming a frontoclypeal apotome.

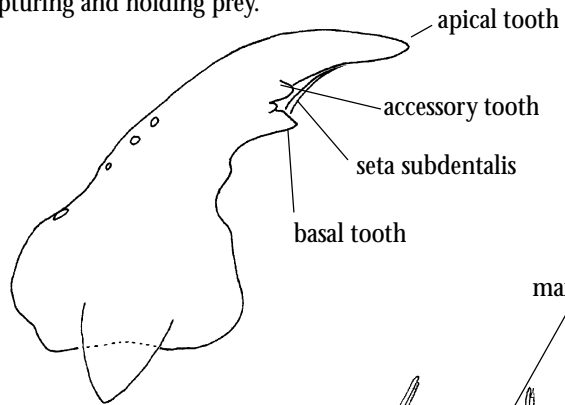
In all subfamilies, the **antennae** provide important characters. A commonly used character is the antennal ratio, **AR**. This is the length of the basal antennal segment divided by the combined lengths of the remaining segments (the remaining segments are collectively termed the **flagellum**). The apical segments are sometimes difficult to discern, especially in those genera with 6-, 7- or 8-segmented antennae. The placement and shape of the **Lauterborn organs**, sensory structures usually located on the second antennal segment or at its apex, are important, as is the location of the **ring organ**. Phase-contrast optics aid greatly in observing these hyaline (translucent) structures of the antennae and the dorsum of the labrum.



The tanypodine head capsule

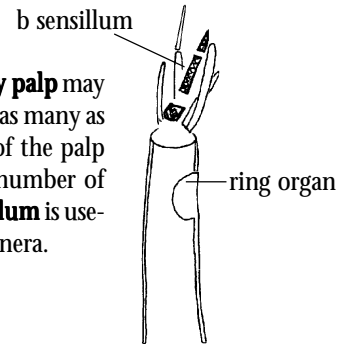
Members of the subfamily Tanypodinae differ from all other subfamilies in having retractile antennae and numerous other uniquely modified structures. Many of the specialized structures, such as the **ligula**, **paraligula** and the **M appendage**, are modifications of the premento-hypopharyngeal complex and associated structures, located dorsally of the mentum.

All Tanypodinae are predacious; the long apical tooth of the **mandible** is well suited for capturing and holding prey.

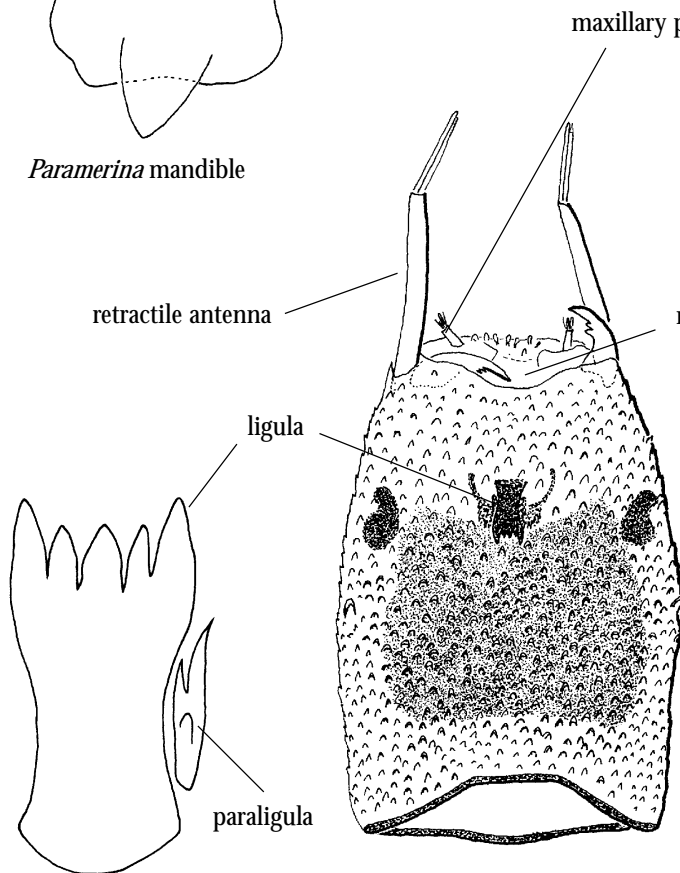


Paramerina mandible

In tanypods, the **maxillary palp** may be simple or divided into as many as 6 sections. At the apex of the palp are several sensillae; the number of "segments" of the **b sensillum** is useful in separating some genera.

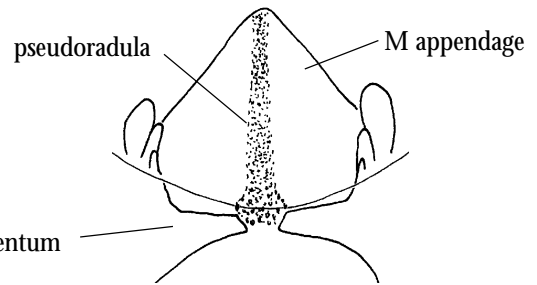


Conchapelopia maxillary palp, with three segmented b sensillum

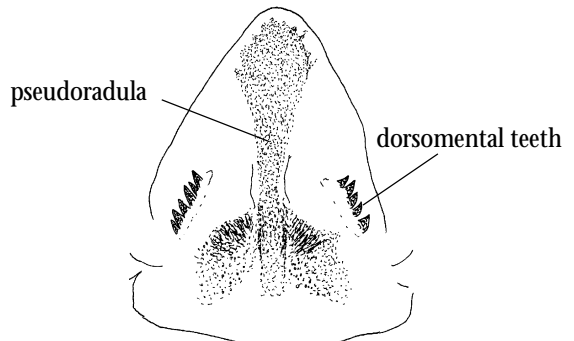


Labrundinia sp. A ligula and paraligula

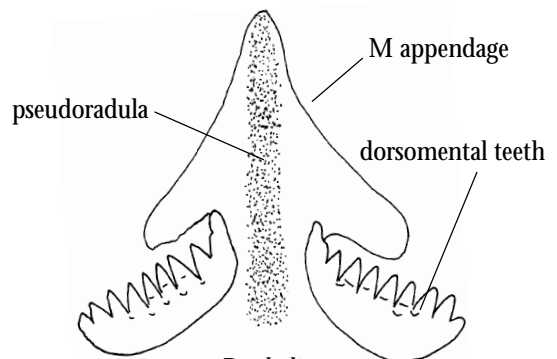
Labrundinia sp. A head capsule



Labrundinia



Coelotanypus



Procladius

The **ligula** is often the most noticeable feature of tanypod head capsules. While the **mentum** is quite distinct in many chironomids, it is weakly developed in many tanypods, with large teeth present only in phylogenetically "primitive" taxa. The **M appendage** of the mentum usually bears a strip of chitinized points, the **pseudoradula**.

Glossary and Abbreviations

Outdated terms are in italics; plurals are in parentheses.

Most structures are illustrated in figures on the previous pages.

- accessory blade** - smaller elongate structure adjacent to antennal blade, usually partially fused with antennal blade at base .
- accessory tooth** - in Tanypodinae, small tooth between basal tooth and apical tooth of mandible; see also **dorsal accessory tooth**.
- anal seta (setae)** - seta(e) located on apex of procercus; also termed procercal setae.
- antennal blade** - elongate structure adjacent to antennal flagellum, arising from apex of first segment.
- apotomal fenestra** - circular to oval to quadrate area, usually anteromedial, on apotome that is lighter in color, a different thickness or of a different “texture” than the remainder of apotome.
- apotome** - see frontal apotome.
- ANSP** - Academy of Natural Sciences of Philadelphia.
- AR** - antennal ratio. In larvae, the ratio of the length of the basal antennal segment divided by the length of the combined apical segments (the **flagellum**). When I measure the flagellum, I measure from the bottom of segment 2 to the apex of the last segment; intersegmental membranes are included.
- b sensillum** - small, cylindrical, one to three sectioned (“segmented”) sensillum on apex of maxillary palp; useful in delimiting genera in the *Thienemannimyia* group of tanypod larvae.
- basal tooth** - large tooth near base of seta subdentalis of tanypod mandible.
- beard** - in chironomid larvae, a group of setae present beneath or adjacent to the lateral margin of the mentum and ventromental plates. A cardinal beard, the type most often found in orthoclads, is one which originates from the cardo of the maxilla; it often appears as setae beneath the ventromental plates. A ventromental beard originates from the dorsal, inner surface of the ventromentum; it is found in prodiamesines and the orthoclad *Diplocladius*.
- bifid** - divided into two parts.
- cardo (cardines)** - the inner basal portion of the maxilla.
- chaetulae laterales** - simple or pectinate blades on each side of the pecten epipharyngis.
- clypeus** - dorsal sclerite of the head immediately anterior to the frontal apotome that bears the S 3 setae.
- conjunctiva (conjunctivae)** - intersegmental membrane(s).
- corona** - in Tanypodinae pupae, the clear area near apex of thoracic horn.
- crenulate** - incised in a regular manner, so that a margin appears to have a series of small rounded or truncated teeth, as in the margin of a scallop shell; adjective: **crenulated**.
- digitiform** - finger-like.
- distal** - towards the apex.
- dorsal** - referring to the upper surface or “top” side.
- dorsal accessory tooth** - dorsal tooth or teeth of mandible in addition to the “normal”, more apical and larger dorsal tooth; present in several species of *Tanytarsus*.
- dorsum** - the upper surface; the “top” side.
- exuviae** - shed skin. “Exuviae” is the singular and plural form of this word; the use of the word “exuvium” is incorrect.
- FAMU** - Florida A & M University, Tallahassee, FL.
- FDEP** - Florida Department of Environmental Protection.
- flagellum** - collective term for the apical segments of the antenna.
- frontal apotome** - elongate plate at center of dorsum of head formed by sutures that, in most taxa, will split and allow the pupa to emerge. If the clypeus is fused to the apotome, it is termed the **frontoclypeal apotome**.
- frontal pit** - small to medium, internal pit found near middle of anterior margin of the frontal apotome of some larvae (*Dicrotendipes*) or directly anterior to apotome (*Glyptotendipes*). This is not the same struc-

- ture as the apotomal fenestra (q.v.).
- frontoclypeal setae** - the S 3 setae, borne on the fused clypeus and frontal apotome.
- in part** - in the keys, this means that the taxon appears in the key more than once.
- labial plate** - mentum.
- labral lamella (lamellae)** - scale-like to plumose structures near median anterior margin of labrum.
- labral sclerite(s)** - central sclerite(s) directly anterior to clypeus and frontal apotome on dorsum of head.
- labrum** - the anterior dorsal portion of the head capsule, essentially the upper lip.
- lateral** - towards the side (also **laterad**)
- Lauterborn organs** - sensory organs on antennae, usually located on apex of second segment, but may arise elsewhere. Usually digitiform but may be on pedicels and collectively may appear leaf-like (in Tanytarsini).
- ligula** - in Tanypodinae, a sclerotized, toothed, tongue-like internal plate near center of head.
- M appendage** - membranous, triangular to arrow-head shaped appendage (in Tanypodinae) near anterior center of prementum; usually bears the pseudoradula (q.v.).
- maxilla (maxillae)** - mouthpart located near base of mandible; bears the maxillary palp. Composed of cardo, galea, lacinia, stipes (these structures essentially fused in chironomid larvae) and maxillary palp.
- maxillary plate** - basal ventral side of maxilla that lies above striae of ventromental plate; the striae of the maxillary plate join with the striae of the ventromental plate to form tubes through which silk may be expressed.
- medial** - referring to the middle or towards the middle.
- mentum (menta)** - (usually) toothed plate on anterior ventral margin of head capsule, composed of a fused ventromentum and dorsomentum.
- mola** - inner portion of mandible below teeth.
- nomen dubium (nomina dubia)** - a scientific name that is considered doubtful or unknown in its application; it usually refers to a name that can not be reliably connected to a taxon because there is no extant material of the taxon and/or the taxon can not be identified from its description.
- NCDENR** - North Carolina Department of Environment and Natural Resources.
- palmate** - like a hand, with finger-like processes.
- paralabial plate** - ventromental plate.
- paraligula** - small sclerotized structure adjacent to ligula.
- parapod(s)** - "legs" of larva; most larvae have a pair of anterior and a pair of posterior parapods (posterior pair often absent in terrestrial larvae).
- pecten epipharyngis** - structure located beneath the anterior central margin of the labrum, often composed of three scales, lamellae or spines (most Orthocladiinae), or may be a pectinate comb (many Chironominae).
- pecten galearis** - small, usually comb-like structure on the dorsal surface of the galea of the maxilla.
- pecten hypopharyngis** - in Tanypodinae, the comb-like structures on either side of the base of the ligula.
- pecten mandibularis** - group of setae near ventral apex of mandible.
- pedestal** - in tanytarsine larvae, the tubercle on the dorsum of the head capsule from which the antenna arises.
- pectinate** - comb-like.
- pedicel** - stalk or stem.
- pharate** - stage within the cuticle of the preceding stage, such as the pharate pupa developed within the larval skin, or a pharate adult developed within the pupal skin.
- plastron** - in Tanypodinae, the apical porous plate on the pupal thoracic horn.
- plumose** - featherlike, extremely finely divided.
- postmentum** - ventral area of head capsule posterior to the mentum.
- premandible** - one of a pair of elongate movable structures beneath the labrum, lacking or vestigial in some subfamilies (Podonominae, Tanypodinae).
- premandibular brush** - group of setae near premandible.

- prementum** - internal, soft, ventral lobe of the premento-hypopharyngeal complex, located dorsal of the mentum, that carries the ligula, paraligula, labial palp and M appendage.
- procercus (procerci)** - tubercle (may be elongate, especially in Tanypodinae) located above the anus; bears the anal setae apically. Absent or vestigial in some taxa.
- proximal** - towards the base.
- pseudoradula** - longitudinal band of fine to coarse points present on middle of M appendage.
- ring organ** - a circular structure (campaniform sensillum) found on the basal segment of the maxillary palp and the antenna.
- rugose** - wrinkled or corrugated.
- S I, S II, S III, SIVA, S IVB** - major setae of the anterodorsal surface of the labrum.
- S 1 – S 12** - setae of the head capsule (not including the setae of the labrum listed above). They are numbered from the anterior end to the posterior end of the head and may have specific names for the structure they arise from or to which they are closest. Those used in this manual are the S 1 and S 2 setae, also termed the labral setae; the S 3 setae, termed the clypeal or frontoclypeal setae; and the S 9 and S 10 setae of the tanypod genus *Larsia*.
- seta interna** - seta located near base of dorsal side of mandible; it is usually apically branched.
- seta subdentalis** - seta on mandible proximal to inner teeth.
- setae submenti** (singular: **seta submenti**) - pair of setae immediately posterior to mentum; in some taxa displaced farther posteriorly.
- sternite** - ventral portion of a segment (in pupae refers to an abdominal segment).
- stria (striae)** - fine, impressed line; usually refers to lines on the ventromental plates of Chironominae ("**strial ridges**"). Striate or striated refers to a structure having striae.
- style** - small (usually) cylindrical sensory organ usually located at tip of second antennal segment; occasionally located near middle of segment.
- supraanal setae** - setae ventral to procerci and dorsal to anal tubules.
- taenia (taeniae)** - flattened, ribbon-like setae; adjective form: **taeniate**.
- taxon (taxa)** - a taxonomic unit, such as a species, genus, family, etc.
- tergite** - dorsal portion of a segment (in pupae refers to an abdominal segment).
- teneral** - recently molted. Teneral individuals usually do not have the "normal" coloration and portions of the body are not yet completely sclerotized ("hardened").
- thoracic horn** - structure near the "shoulder" of pupa; may be a simple bag-like structure, tubular, ellipsoid, branched, plumose or absent.
- triangulum occipitale** - the roughly triangular area between the posterior margin of the head capsule and the first suture anterior to it (the secondary postoccipital margin).
- tribe** - a taxonomic unit between the subfamily and genus; e.g., similar genera within a subfamily are grouped into tribes. The only tribes used in this manual are those of the subfamily Chironominae: Chironomini, Pseudochironomini and Tanytarsini.
- tubules** - tubular gill-like structures originating from body segments X-XI (ventral and/or caudolateral tubules) or from near anus (anal tubules).
- USGS** - United States Geological Survey.
- VP** - ventral pore, a sensory structure on the venter of the head capsule.
- venter** - the lower or "bottom" side.
- ventral** - referring to the lower or "bottom" side.
- ventromental plate(s)** - plate-like or shelf-like ventral outgrowth of the head capsule adjacent to each side of the mentum.
- vortex (vortices)** - circular group of spines located on posterolateral portion of some pupal abdominal sternites, formerly termed "pedes spurii A".

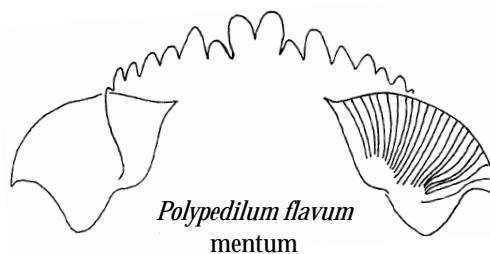
About the Names ...

As noted previously, one of the confusing aspects of working with chironomids has been the apparently rampant changing of names. There are good reasons for some of this “name changing”. First is the Principle of Priority, one of the main principles of the International Code of Zoological Nomenclature (International Commission on Zoological Nomenclature 1999). Simply put, the first name given an organism is the one that has priority over other names applied to the same organism at a later date. As with everything, there are exceptions! The “rules” have been suspended in some cases. An example would be when a name that has been in use for a long time is discovered to be a junior synonym of a name that has not been used since it was published; the older name may be suppressed by a special ruling of the International Commission on Zoological Nomenclature. This has happened with several names in the Chironomidae, including the family name itself! See Ashe (1983) for a more detailed explanation of some of the family’s name changes.

Other name changes may occur when a species is transferred from one genus to another. In Latin, there are three genders: masculine, feminine and neuter. The gender of the species name must agree in gender with the genus name. Thus, when a species is moved from one genus to another with a different gender, the spelling of the species name may change. For example, Johannsen described the species *Chironomus flavus*. *Chironomus*, ending in *-us*, is masculine, so *flavus* is masculine to agree with it.

When the species was moved to *Polypedilum* (which ending in *-um*, is neuter), *flavus* was changed to *flavum* to agree in gender with the neuter *Polypedilum*. It seems pretty simple, doesn’t it? HA! There are also many other things to consider when coining names, such as the derivation of the name, its case, its tense, etc. And, just to confuse things, in plants, many genera ending in *-us* are feminine! For an insight on the formation and meaning of scientific names, see Brown (1956) and Ride et al. (1985).

We also have problems with mistaken identities. The example I just used, *Polypedilum flavum*, is the correct name for the species that has been called *Polypedilum convictum* in this country for years! This happened because Townes (1945) synonymized the Nearctic *Chironomus flavus* with the Palaearctic *Chironomus convictus*, because the adults were apparently inseparable (the species were originally described in *Chironomus*; later taxonomic work showed that they belonged in the genus *Polypedilum*). However the immature stages are quite different. The necessity for a “name change” was postulated in Epler (1992, 1995 - and note that I failed to correct the specific name for gender!) and finally made “official” in Oyewo & Sæther (1998).



Collecting and Preserving Chironomidae

Larval Chironomidae can be collected with any of the standard benthic collection devices. Larvae are best preserved in 70-80% ethanol. Formalin preserved larvae (and other life stages) can be difficult to clear if left in formalin for more than a few days or weeks.

Many workers add Rose Bengal stain to samples to facilitate “picking” - if you don’t absolutely need it,

please don’t use it. I strongly recommend that this stain **NOT** be used! Rose Bengal often excessively stains many head capsules, making them too dark for proper light transmission. This obscures many of the tiny structures present on chironomids, rendering specimens very difficult to identify. Mountants, such as CMCP-9AF, that stain specimens should also not be used.

Identifying Chironomidae

Believe it or not, identifying chironomid larvae is easier than identifying the larvae of some other aquatic groups! Chironomidae have fascinated many aquatic scientists (it seems there is always a plentiful supply of “nuts”), and a large number of taxa have been reared and associated with their adult forms. The adults are important because historically, most Chironomidae species have been described based on characters of the adult male. Compared to some other families of aquatic flies, larval Chironomidae offer numerous morphological characters for identification. Problems arise when all these easily seen characters are similar in closely related (or sometimes not so closely related) taxa. Some characters are minute and hyaline, and are visible best under a high power oil immersion lens, using phase contrast or Nomarski optics/lighting.

Because of the similarity of some of these easily observed characters, it often is not possible to identify a chironomid by simply matching structures of your larva with a picture of a structure. This apparently has been the case in many misidentified North American specimens I've seen; many of these specimens were apparently matched with pictures of structures illustrated in Wiederholm (1983), for they bear names of species known only from the Palaearctic. Match-the-picture technology may work with identifying birds, but not, in general, with chironomids! Of course, there are many exceptions; they wouldn't be chironomids if there were no exceptions!! There are many species with Holarctic distribution patterns - and we will continue to find more such taxa as our knowledge of the Nearctic fauna grows - but one must exercise caution when applying names to taxa which are apparently extralimital.

Identifying an organism does not mean just running it through a key and coming up with an “identification” - after all, if you start to key something out, it will key somewhere! That does not mean you have arrived at the correct destination. Once a specimen has been keyed, you must double check your findings by consulting a diagnosis or description of the taxon you believe you have, or check it

against specimens in a reference collection, if such a collection is available *and* if the collection has been verified by an expert. You should also look for information on distribution and hopefully some illustrations of morphological structures unique to that taxon; a major failing of many identification guides is their lack of diagnoses or descriptions, and insufficient illustrations. While well written keys will work for some groups for some geographical areas, this is not always the case with our wonderful chironomids! A lot of misinformation has been “provided” by poorly written or researched keys. You should also keep in mind that the specimen you're keying out may be new to science and/or was not seen by the person who wrote the key(s) you are using.

There is no single document that will allow one to identify all North American chironomid larvae to the species level; Wiederholm (1983, 1986, 1989) and Coffman & Ferrington (1996) go only to genus. There are two ways you can try to identify larvae to species:

- 1) Go to the literature and search for revisions of the taxa that you have already identified to genus. If the genus has been revised, you may be able to find keys or descriptions that will enable you to identify your specimen.

- 2) Look for regional guides to larval identification. Unfortunately, for North America there are very few such guides, and of those, two (Beck 1976, 1979; Webb & Brigham 1982) should not be used because of the inaccurate information they contain. Regional guides such as those by Epler (1995) and Simpson & Bode (1980) are reasonably accurate, although they are both out of date taxonomically (what's new?). The 1995 Epler guide was updated via the World Wide Web, but this current manual supercedes it and will be the one updated via my web site from now on (see Sources).

The credibility of good identifications with a regional guide may be high if the guide is extensively researched. Take for example the guide to Florida

larval Chironomidae (Epler 1995). This guide was possible because of the large amount of reared material that was available to the author. Previous workers in Florida, such as Bill and Beth Beck, Annelle Soptonis and myself, had reared a considerable number of the species in Florida, and additional *reared* material and information had been provided by other workers such Broughton Caldwell, Bohdan Bilyj, Martin Spies, Mike Bolton, Jim Sublette, Bob Rutter, Mike Heyn, Charles Watson and many others. A larval guide to species level identification was possible because many of the larvae had been associated with the adult stage (although, of course, not enough!) in a relatively small area (Florida). This current guide to the Carolinas was possible for the same reasons - and most of the same people (see Acknowledgements). I also studied a large number of type specimens and in essence did mini-revisions of many of the genera in this manual.

In order to really get a handle on the larval Chironomidae of North America, more revisionary work utilizing all life stages is necessary. And more regional guides to larvae and pupae are needed!

Regional guides can sometimes be used outside of the region they cover, but one must exercise caution! Regional guides often omit taxa that may be present in your area, and include taxa that probably will not be present in your area. In general, the chironomid fauna of any area is elucidated by the collection of adults (remember, most of the names of species are based on characters of adult males) and the rearing of larvae to adulthood, thus (hopefully) making it possible to identify the larvae. The Southeast US is one of the better known areas of the Nearctic, but we still have dozens of undescribed species. We must use letter or number designators (sp. A, sp. 1, etc.) to "identify" such species until the immature stages are associated with described adults, or the complete metamorphosis of the new species is described.

Do not identify specimens to a level beyond your capability or the capability of current taxonomy. Given the incomplete status of our knowledge of some chironomid taxa, many times an identification

to a species group or complex will be all that is currently possible. Also, remember that most keys are based on fourth instar larvae. The fourth instar is the last larval instar; reared associations usually consist of the fourth instar larval exuviae, the pupal exuviae and the adult. Thus the fourth instar may be the only larval instar known with any morphological "precision". Some larval characters may change from one instar to the next; this is especially true for relative/comparative lengths of some body parts; earlier instars may not, or in some cases, *will not*, key correctly. There is no shame in listing a specimen as a "*Dicrotendipes* sp." when you can not be positive of your determination at the species level (without fourth instar specimens or associated material this may be necessary for many larvae). I have seen numerous studies and species lists based solely on larval identifications that would be impossible to achieve without associated specimens - just how were those specimens identified? By comparing pictures? Wishful thinking? Intuition? Did somebody pin a list on a wall and throw darts at it? (I've seen some collections that apparently were identified in a manner similar to this!) If you can't realistically and accurately put a species name on a specimen, drop back and punt at the generic level!

If you are uncertain of a generic or species identification but are relatively sure that you're close to being correct with a name, you can use the modifier "cf.". This is an abbreviation for a Latin term that means "compare to". If you're uncertain about the genus, use: "cf. *Meropelopia*" (or whatever genus); if uncertain of the species use: "*Goeldichironomus* cf. *natans*" (or whatever). Question marks can also be used, but many workers place them incorrectly, which leads to confusion! This confusion is best avoided by not using question marks in names. Also, do not use the modifier "nr." (an abbreviation for "near"; an example would be "*Polypedilum* nr. *illinoense*"). This implies a close phylogenetic relationship between your specimen and another species. Many keys are artificial constructs used to identify organisms; they do not necessarily imply phylogenetic relationships. Thus if your specimen keys to a couplet but doesn't quite fit, it does not automatically follow that your specimen is "near" the other taxon in that couplet.

Materials and Equipment Required for Larval Chironomid Identification

Microscopes: You will need a dissecting (stereo) microscope for sorting larvae and mounting them on microscope slides. A compound microscope is necessary for identification; one with phase-contrast optics or better is recommended. The compound microscope should have several objective lenses: a low power scanning lens (4X, which gives “40 power” with a 10X eyepiece), which makes it easier to locate your specimens on your slide, a 40X (“400 power”) lens for most work and a 100X (“1000 power”) oil-immersion lens; 10X and 20X objectives may be desirable, but are not necessary. Phase-contrast optics and a high power oil-immersion lens may be expensive, but are necessary for observing minute hyaline structures such as the S I, labral lamellae and the apical sensilla of the maxillary palpus. Another necessity is a measuring reticle (a glass disc etched with a grid or ruler line, which fits into one of the microscope’s eyepieces); this accessory is needed to provide accurate length measurements (often the only way to separate some species) and to calculate ratios. Be sure to calibrate your reticle with a stage micrometer (usually, a precisely etched glass slide is used) at all magnifications you will be using.

Microscope slides, cover slips (glasses) and boxes: Whatever size you find convenient. Use glass cover slips. **DO NOT USE PLASTIC COVER SLIPS!!!** It is often necessary to press down on cover slips to reposition or flatten larvae; plastic cover slips will scratch and become impossible to see through. Round or square cover slips from 12 to 22 mm work well for most larvae. I favor the round ones because they allow more rotation and better positioning of your specimens. Although I have no empirical data, it also seems that round coverslips are less prone to air fingers than square or rectangular ones. Note that small

round coverslips, from 6 to 10 mm, are useful for mounting associated larval and pupal exuviae with the emergent adult. Good slide boxes for maintaining reference collections are a necessity; don’t scrimp on quality!

Mounting medium. There are two major kinds of mounting media:

- 1} media in which chironomid larvae can be mounted directly from water or alcohol. CMC-10 is the most widely used of these; Hoyer’s (or Berlese’s) is also sometimes used. These media almost always must be ringed to achieve any degree of longevity; slides made with them may be considered at best to be “semi-permanent”.
- 2} media which require “clearing” and/or dehydration of the material to be mounted. This includes mountants such as Canada balsam and Euparal. In general, these media can produce museum quality slides that can be considered “permanent”.

Both types have their advantages and disadvantages - of course! Nothing is ever simple!! We’ll discuss CMC first, since it apparently is the medium of choice for most benthologists involved in large studies where hundreds or thousands of slides must be made.

CMC The negligible or reduced specimen preparation time no doubt makes CMC a favorite time saver. CMC medium comes in several varieties - CMC-10 (and to a much lesser extent, CMC-9) appears to be the most widely used for invertebrate slide making. CMC is a water-based medium; material can be mounted in it from alcohol or water, and the medium does impart a clearing action (clearing refers to the maceration or “digestion” of inner muscle tissue, thus allowing light rays to pass through the body. Remember that a compound microscope

essentially shines a light through the viewed subject; if there is too much muscle tissue obstructing the light beams, one can not readily observe the structures necessary for identification.) Note that CMC may not sufficiently clear larvae with thick, dark head capsules (some diamesines, orthocladids and chironomines) or specimens that have been heavily stained with Rose Bengal; in such cases, the larvae may have to be cleared in KOH before mounting (see under Canada balsam technique).

advantages. quick mounting; clearing action; possible to mount large numbers of larvae in short time; can be thinned with water or alcohol; water-base makes it easy to soak off old cover slips and remount material.

disadvantages. medium may develop “air fingers” unless (or even if!) coverslip is ringed; medium can crystallize; some larvae will not clear sufficiently for identification without prior maceration in KOH; some larvae may effervesce and produce gas bubbles after the coverslip is put on; medium is temporary or semi-permanent at best; bad odor and may be hazardous.

Hoyer's medium apparently can no longer be purchased in the US because it contains chloral hydrate, a federally controlled substance. However, if you wish to go through the agony of obtaining a federal permit (or if you have a friend in the chemistry department), Hoyer's may be prepared from the following ingredients:

- 30 g gum arabic (ground crystals or powder, *not* flakes)
- 200 g chloral hydrate
- 20 cc glycerin
- 50 cc water

After mixing (it will take some time for all materials to go into solution - do not heat the mixture to speed the process!), filter the mixture through glass wool before usage and/or storage.

advantages. same as those for CMC.

disadvantages. same as those for CMC; not available commercially.

Canada balsam and Euparal. These media require xylene, cellosolve (ethylene glycol monoethyl ether), or for Euparal, “Euparal essence”, for thinning, and specimens must be cleared and dehydrated before mounting.

advantages. produces superior slides for permanent storage.

disadvantages. specimens must be cleared and dehydrated before mounting; long drying time.

Storage and dispensing of mounting media. In general, one should store media in the containers in which they were shipped; media may be affected by exposure to air or light. These containers are usually too large to work as a dispensing unit. There are containers made specifically to hold mountant, such as Wheaton balsam bottles (figured below). These bottles usually come with a rod or wand for dispensing the mountant. To help prevent evaporation or desiccation of the mountant, I put a small amount of petroleum jelly on the bottom lip of the bottle top. CMC, Canada balsam and Euparal can be stored in and dispensed from Wheaton bottles, but I also use a small plastic squeeze bottle (I use the little bottles from pH testing kits made for pools) for dispensing CMC.



Wheaton balsam bottle

Also see page 1.25 for a checklist of other materials that you may need.

Sorting Chironomid Larvae

Mounting every chironomid from a sample is often unrealistic when huge numbers are collected. I always sort chironomids before slide mounting them. If you are mounting several specimens on one slide, you (hopefully) should at least have similar taxa under the cover slip and will be in the same part of the book when trying to identify them!

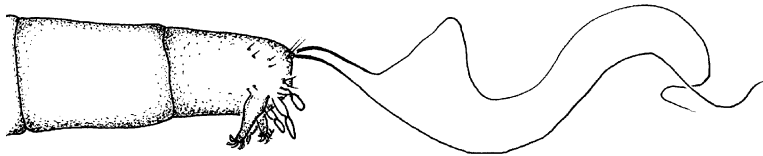
Many Chironomidae can be sorted to genus, even species, while still in fluid preservative, and representatives from each sorted group can be mounted.

NOTE, however, that you should have considerable experience before identifying unmounted larvae from fluid, and you should frequently mount specimens from groups you already “know” in order to be sure you’re still looking at the same taxa. It is always best to remain skeptical of one’s abilities!

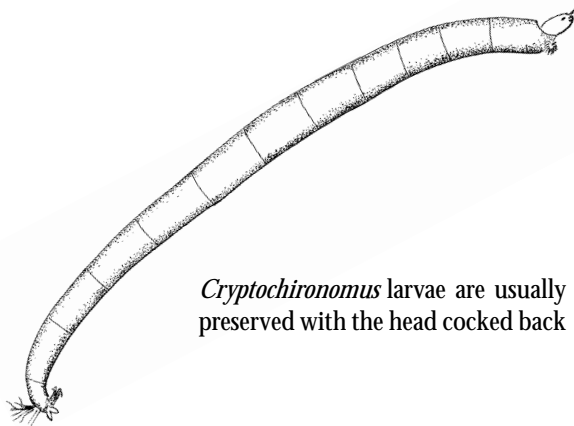
Chironomid larvae possess an abundance of characters that can be observed while in alcohol. Some of these are:

1. The general appearance of the body

Is the body setose (“hairy”)? Are the setae scattered, arranged in lines along the side of the body or are they grouped as tufts near the posterior corners of a segment? Are there setae at the end of the abdomen? If so, are they long or short? Is the body curved or the head distinctly bent? Are posterior parapods present? Are there any darkened claws on the posterior parapods? What is the condition of the anal tubules?

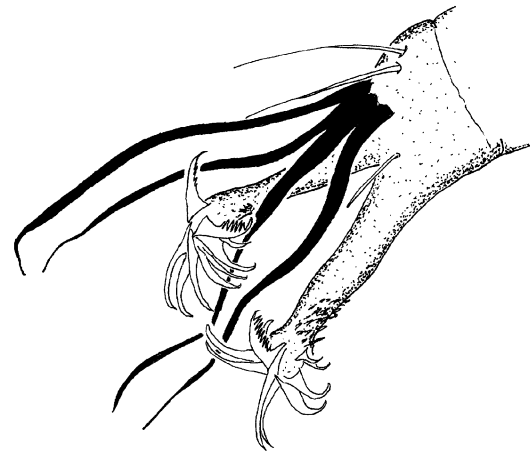
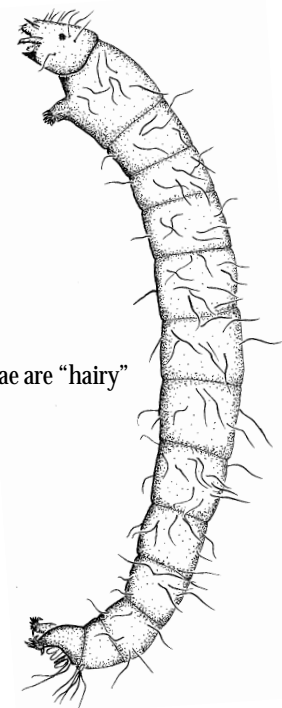


Parachaetocladius has extremely long anal seta
(Note: some other taxa also have long anal setae.)



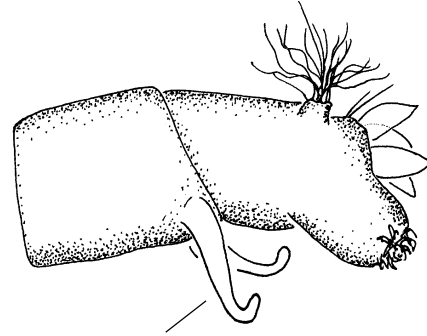
Cryptochironomus larvae are usually preserved with the head cocked back

Tvetenia larvae are “hairy”

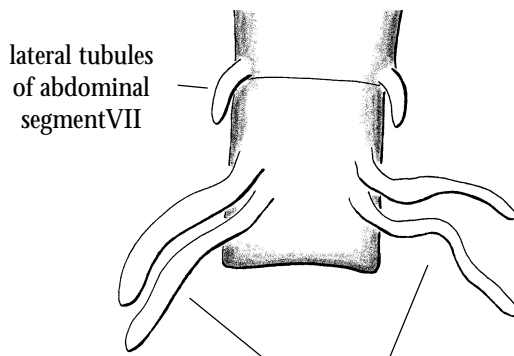


The anal tubules of *Nilotanypus* and *Pentaneura* are longer than their posterior parapods

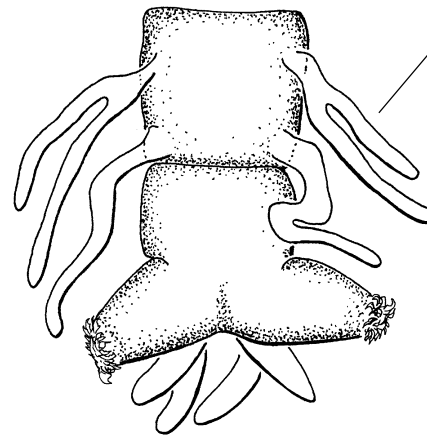
Are ventral or lateral tubules present near the end of the abdomen? If ventral tubules are present, how many pairs are there and how are they shaped? Note the shape and length of the anal tubules.



ventral tubules on abdominal segment VIII of *Glyptotendipes barbipes*

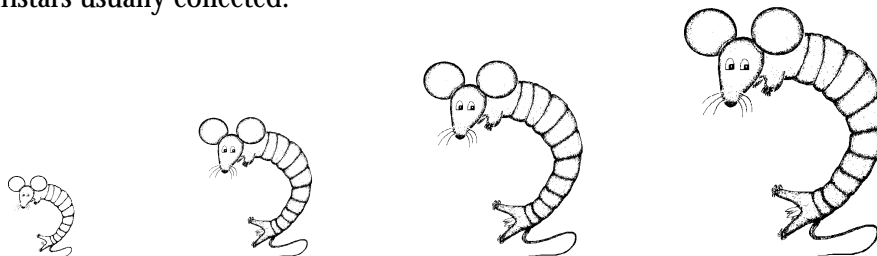


ventral tubules on abdominal segment VIII of a *Chironomus* species



ventral tubules on abdominal segment VIII of *Goeldichironomus holoprasinus* - note the bifid first pair

Size can also be considered; some larvae are huge (*Chironomus*, *Glyptotendipes*); others are tiny (*Corynoneura*, *Thienemanniella*). But, remember that although different taxa may be different sizes, different instars of one species will be of different dimensions. First instar larvae are tiny and may be generally planktonic; depending on the mesh size of your net or sieves, you may collect second instar larvae as well as the third and fourth instars usually collected.



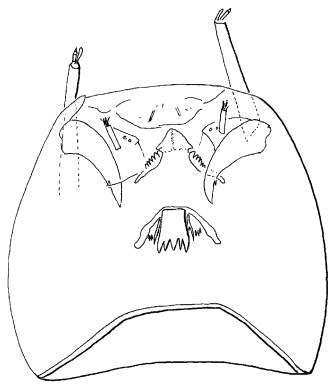
different instars of the same species will be of different sizes

2. Color of the body

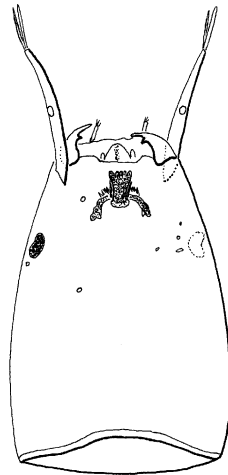
Best seen with live or fresh specimens (most alcohol preserved specimens will bleach). Some larvae may be white, cream, red, green or even purple! Some larvae may have color bands on the body. The larva of *Cricotopus lebetis* has its second and third thoracic segments colored a bright blue!

3. Shape and structures of the head capsule

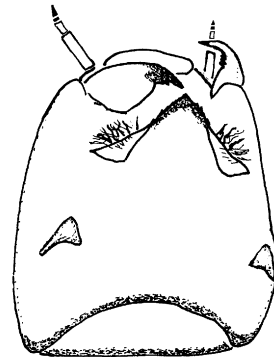
Some larvae have rounded head capsules, others are apically pointed; some are flattened. There are larvae with bumps, knobs or projections on the head capsule. Note the length of the antennae (see 5). The mentum and associated structures are easily visible on larger larvae and can get you right to genus with some taxa. In a lateral or ventral view, the triangulum occipitale provides a good character to separate larvae of *Kiefferulus* and *Goeldichironomus* (where it is large) from those of *Dicrotendipes*, *Einfeldia* and *Chironomus* (where it is small) while still in fluid preservative.



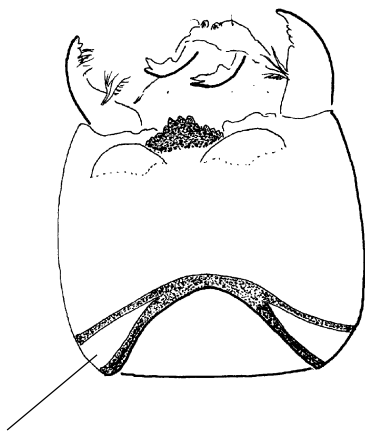
the head capsule of *Brundinella* is rounded



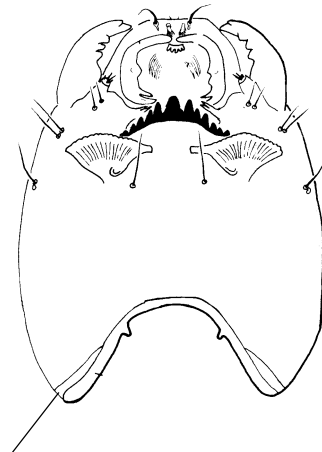
Denopelopia has a narrow head capsule



Rheocricotopus tuberculatus has a pair of small tubercles ventrally



the triangulum occipitale of *Goeldichironomus* is large



the triangulum occipitale of *Dicrotendipes* is small

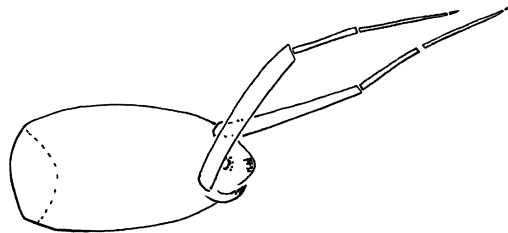
4. Color or markings of the head capsule.

As with body colors, head capsules come in a variety of colors, from colorless to black. The dorsum and/or venter of the head may bear stripes, spots or bars. The postmentum (the area of the head capsule posterior to the mentum, also called the gular region) or the posterior margin of the head capsule may be darkened. Note how many eyespots are present and how they are arranged.

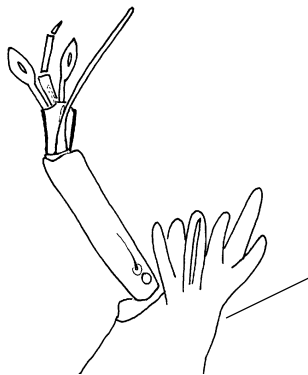
5. Antennae.

The shape and length of the antennae are diagnostic for many taxa. Larvae of the subfamily Tanypodinae have retractile antennae (they can be pulled into the head capsule) and are thus easily identified to subfamily while alive or in alcohol.

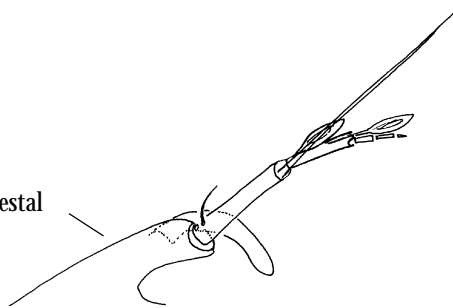
Note the long antennae of *Corynoneura*.



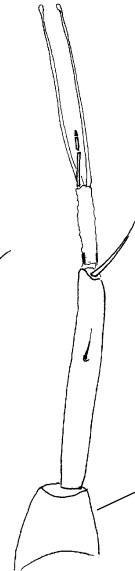
Note that the members of the tribe Tanytarsini (subfamily Chironominae) have their antennae mounted on elongated pedestals which are sometimes adorned with spines and other projections.



Stempellina - note the ornate pedestal



Stempellinella - this particular species has a long curved projection from the pedestal



the unadorned pedestal of a *Tanytarsus* species



this *Micropsectra* has a spine on its pedestal

6. Cases

Many benthic larvae build tubes of detritus, feces, and other available materials that are cemented with silk (*Chironomus*, *Glyptotendipes*, *Tanytarsus*); some larvae build transportable cases and they may often be collected while still in their cases. Note the distinctive cases of *Zavreliella* (like a hydroptilid caddisfly case) or the cases of *Rheotanytarsus*. Although it is not possible to identify most *Rheotanytarsus* larvae to species, they can be separated into two major groups by the type of larval/pupal case - the *Rh. pellucidus* group has a case with a long attachment stem; the *Rh. exiguus* group's case is attached along its side.

Slide Preparation

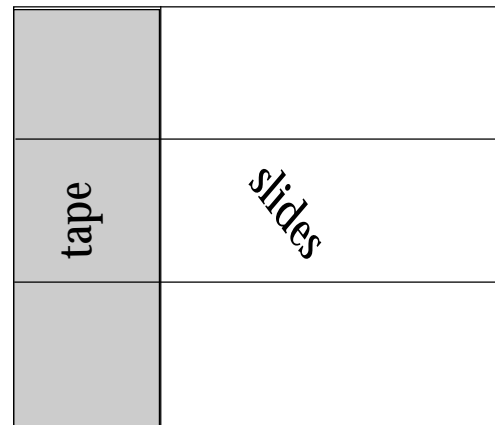
What follows below are the “long-winded”, detailed directions for slide-making, replete with all kinds of good tips. A simple, basic version is provided on page 29, which may be easier to refer to while making slides. **HOWEVER**, be sure to read *ALL* of the material below before you make slides for the first time!! - or if you think there is a chance you may learn something new here ...

- 1 Always start with clean slides and cover slips. Note that although the box of cover slips may read “pre-cleaned”, apparently a different definition of the term is being used than that accepted by most biologists; slides and especially cover slips can always benefit from a quick wipe before use (as long as a clean tissue is used).
- 2 Label the slide! Basically, slides without good labels are useless!! Few things are as irritating to a taxonomist than finding an interesting specimen with no collection data! Using codes or sample collection numbers is fine while the slide is in the lab, etc., but be sure to label the slide with complete information before sending it off to an expert for identification or verification, and before putting it into your reference collection. Years from now there is a good chance that nobody in the lab will remember to what the codes referred.



Always include complete collection data on any slides in your reference or voucher specimen collections!!!

There are several ways to put a label on a slide. Peel off and stick on labels are satisfactory, as are frosted slides, providing the “frosting” is fine. Avoid coarsely frosted slides - they do not allow fine writing. Satisfactory labels can also be made with transparent (regular “Scotch”) tape. The tape method can be quite useful when making large numbers of slides; one can line up several slides and run a length of tape across all of them, and then use a knife or razor edge to separate the now labeled slides. Tape can be written on with pen or pencil; if using a pen, use at least waterproof ink. Alcohol-proof India ink is recommended - sooner or later, someone will spill something like alcohol over the slide and ...



tape can be applied to several slides at once and then cut with a knife, etc.

Remember that a compound microscope inverts the image. If you want to be able to look at your specimen and read the label on the slide, turn the slide around. This way you can read the slide label while the slide is on the microscope stage, and the head capsule will be oriented correctly.

Now you're ready to mount your specimens. What you do next depends on the type of mounting medium you are using. We'll discuss the most popular method using CMC first.

CMC method

Most specimens need no preparation before mounting in CMC; larvae may be mounted from alcohol or water. Place 2-5 drops of CMC on the slide (the amount will vary with how many specimens you are mounting, their size, etc.). It is generally to your advantage to use a small excess of mountant; this will reduce the possibility of "air fingers" from forming near the edge of the coverslip. However, if you are mounting many (more than seven or so) larvae under one coverslip, too much mountant may ooze out from under the slip and may carry a few larvae with it. If this happens, you can pick up the coverslip and try again, or you can sometimes push the larva(e) back under the slip.

Place the specimen(s) in the mountant, laying the larva(e) ventral side up (and head pointed up). You will note that allowing the larvae to sit in the mountant for a short period will soften them a bit and make them easier to manipulate. Tease out some of the larger bubbles that may form (don't worry about getting them all; most will disperse on their own). When mounting from alcohol or water, if too much liquid is carried with the larvae, the mountant may spread out too much - it sometimes is necessary to wick the liquid off the larvae by gently touching them to a paper towel or similar absorbent surface.

The number of specimens and coverslips per slide is your choice. Certainly, "special" larvae deserve their own separate slides, but for efficiency it is possible to mount numerous larvae on a slide. Note that while it is possible to mount 10 or more larvae under a single large coverslip (22 mm), and to put two such coverslips on a slide, such a technique frequently results in numerous larvae being oriented in a less than satisfactory position.

Some workers remove the

head and mount it under a separate coverslip. This is not necessary for most larvae, but certain recalcitrant larvae do require decapitation in order to position the head capsule correctly. Be certain to mount the body on the same slide! Both head capsule and detached body can often be mounted under the same coverslip.

Using forceps (I use an old pair with relatively blunt points), take a clean coverslip and gently lower it over the mountant at an angle. Try not to drop the slip onto the mountant - this may trap air bubbles. If your mountant is a bit thick or has set up prematurely, dip the coverslip in alcohol, wipe it off, and then dip it back in the alcohol again to pick up a tiny drop of alcohol, then place it over the mountant; the drop of alcohol helps the coverslip settle over the medium more smoothly. Once the mountant has filled in under the coverslip, you can finish arranging your larva(e) under the slip. By pushing the coverslip from one side, etc., you can roll the larva(e) to the position desired. Then, gently press down on the coverslip over the head capsule with blunt forceps, pencil eraser, etc. to spread the mouthparts, and over the anal end to spread the claws of the hind parapods (especially important with tany-pod larvae). It is important to spread the mandibles so that their inner teeth can be viewed.

Lay the slide on a flat surface and allow it to cure at room temperature for 2-3 hours. As it dries, the medium may shrink and produce air fingers at the

TIP: When mounting several larvae under one coverslip, it may be easier to lay them all on one side (all facing the same direction) and roll them over on their backs with the coverslip (after it is put on). Also, allowing the mountant to set up a bit after placing the larvae in it but before placing the coverslip on may help them stay in place when you finally do put the coverslip on.

edge of the cover slip; if any air fingers are present, fill them in with more CMC (place a small amount next to the affected area; capillary action will usually draw it in under the slip) and allow to cure for another 1-2 hours. For best results and more permanent slides, use clear fingernail polish or more CMC to ring the coverslips; this creates a seal that prevents more air fingers from forming. Note: if you are making large numbers of slides, it may be

time saving to examine the specimens before ringing them, and only ringing the slides with unusual, important or new specimens. After this the slides can be placed in a drying oven (do not exceed 55° C) for a day or two (usually longer at room temperature) and are then ready for detailed examination. Slides may be examined earlier, but do not use the oil immersion lens on slides until the medium has dried; otherwise, the coverslip may lift off or shift sideways and specimens may be damaged, or you may coat your objective lens with mountant!

“Bad” slides can be remounted easily by soaking the coverslip off in water; old slides may require soak-

ing for several days. If the coverslip has already been ringed with fingernail polish, the sealant can be removed with ethyl acetate.

If CMC slides are examined within 3-4 weeks (and perhaps after even longer periods of time), it is usually possible to reorient larvae under a coverslip by applying pressure with blunt forceps over the area that needs to be moved. I do this at 40X power - the low power scanning objective of my compound microscope. You may be amazed how much you can move or squash specimens. Use caution about too much pressure or the coverslip will break!

Canada Balsam/Euparal Method

Important or potentially important material should be mounted in a good permanent medium. The two media most often used are Canada balsam and Euparal. Although both of these media can impart a slight clearing action, it generally is not sufficient for observation of many characters (although small larvae will often be sufficiently cleared). Thus we have several more steps in this slide making procedure involved in preparing the specimens before mounting. These steps include clearing the larvae followed by baths in various liquids. You will need several small containers, one for each bath - two of which will need lids to prevent evaporation of the liquids. I use very small, nesting watch glasses. Bear in mind that there are several techniques for treating specimens before mounting. What follows are the best methods I have used. Note that if you are mounting larval or pupal exuviae (shed skins), you need only soak the exuviae in 95% propanol before mounting in either balsam or Euparal.

1 - Begin with placing the larvae in a 10% solution of potassium hydroxide (KOH). KOH will digest the inner muscle tissue and leave the sclerotized portions of the larval exoskeleton, including the body. Larvae can be left in the solution over night at room temperature and by the next morning they should be sufficiently clear to begin the next step, dehydration. Or, to speed things up, the KOH solution with the larvae can be gently heated to just

below its boiling point. You may wish to put the larvae and KOH in test tubes and then immerse the tubes in a beaker with boiling water, essentially a water bath. Or the KOH solution can be warmed directly on a hot plate using small ceramic pots containing the solution. Of course one must be very cautious not to over clear the specimens.

2 - Whether using heated or room temperature KOH, the next step for the specimens after clearing is a bath in distilled water. Note that some clearing action may continue in the water - larvae that have been over cleared may seem to “disappear” at this stage! Don’t worry too much - the specimens are still there. If the specimens are not easily visible, try lighting from below, or tilt the container; often bodies will line up along the meniscus. Transfer the specimens using a small dropper, or drape them over a needle, or lift them between the tips of forceps - be gentle! Specimens should sit in the water bath approximately 3 to 10 minutes, depending on the number of specimens and their size.

3 - Following the water bath, specimens are placed in glacial acetic acid (this container should remain covered) for 3 -10 minutes.

4 - Traditionally, the last bath may differ whether Euparal or Canada balsam is used. If using balsam, specimens go into a bath of 95% propanol layered

over cedar wood oil. Note that it is probably not necessary to use cedar wood oil, but be sure to test this before you mount important material without the cedar wood oil bath. I have had success using a final bath of just 95% propanol. If you are using Euparal, place in a bath of just 95% propanol. As with previous baths, allow 3 -10 minutes. With either fluid, the container should be covered to slow evaporation or absorption of atmospheric water.

From this point, directions are similar to the CMC method. Apply several drops of mountant on the slide. Using fine forceps or a needle, transfer the larva(e) to the mountant. The liquid transferred with the larva(e) will thin the mountant a bit. Arrange your larva(e) and put on a cover slip. Note that because the head capsule has been cleared, it will not take as much pressure to spread the mandibles, etc. Often, the weight of the coverslip will suffice. It is not necessary to ring slides made with Canada balsam or Euparal.

Once the slide is made, it should be placed in a drying oven. Slides made with balsam or Euparal take a much longer time to dry than those made with CMC. It may take two to three weeks before a Euparal slide can be looked at under an oil immersion lens.

If the mountant hardens too quickly, it may be thinned. With Canada balsam, use a bit of xylene; 95% propanol will also work, but xylene usually does a much better job. Euparal can be thinned on the slide with 95% propanol; Euparal essence should be used to thin Euparal that has become too thick or has dried out in its dispensing or storage containers. If slides need to be remade or cover slips need to be replaced, use xylene for balsam slides and Euparal essence or 95% propanol for Euparal mounts.

Cellosolve (ethylene glycol monoethyl ether) may be used as a thinner for Canada balsam (if all the xylene, normally used as a thinner for balsam, has been evaporated from the balsam). Material that doesn't require extensive clearing (lightly sclerotized body parts, etc.) can be placed in a bath of cellosolve

and then directly in balsam. However, cellosolve is very prone to contamination with atmospheric water. This contamination can produce a cloudy mountant - and many times you won't know this until several weeks after the slides have been made. I no longer recommend using cellosolve.

When large numbers of larvae from several sites must be processed, an efficient assembly line procedure can be set up with both the CMC and Canada balsam methods.

Checklist of necessary lab equipment and materials:

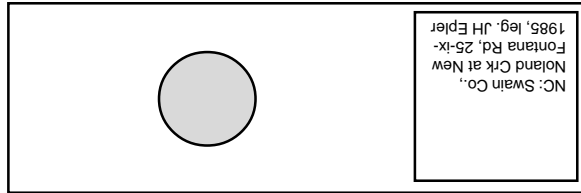
- slides
- coverslips
- slide labels
- mounting medium
- slide storage boxes
- compound microscope with (at least) phase contrast optics
- stereo (dissecting) microscope
- forceps ("tweezers")
- labware (dishes, vials, watch glasses, etc.)
- 70-80% ethanol
- ethyl acetate
- clear fingernail polish
- * 95% propanol (used because it evaporates much more slowly than ethanol)
- * glacial acetic acid
- * cedar wood oil
- * xylene
- * hot plate
- * Euparal essence (only if using Euparal)
- * = if using Canada balsam or Euparal

A note on forceps. I use forceps with the finest points available, Dumont #5's. It is important to keep your forceps as sharp as possible. I've been in labs where technicians were using old, blunt forceps that felt like two huge trees under the scope. Obtain a hard honing stone for maintaining sharp points; you'll be amazed at the comfort difference using sharp forceps makes!

Slide making with CMC

(see text for more details)

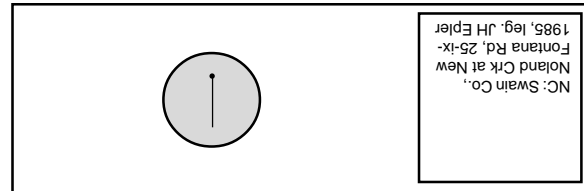
Start with a clean slide. The first thing you need to do is to label the slide! A short code, etc., is OK, but be sure to add complete label data if the slide will be a voucher or reference specimen, or if you send it to an expert for identification or verification.



Then turn the slide around. Remember that a compound microscope inverts the image. This way you can read the slide label while the slide is on the microscope stage, and the head capsule will be oriented correctly.

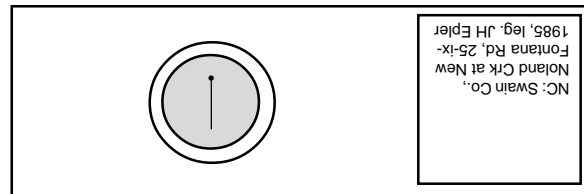
Place 2-5 drops of CMC on the slide (the amount will vary with how many specimens you are mounting, their size, etc.)

Place the specimen(s) in the mountant, laying the larva(e) ventral side up (and head pointed up). Tease out some of the larger bubbles that may form (don't worry about getting them all; most will disperse on their own).



Take a *clean* cover slip and gently lower it over the mountant at an angle. Try not to drop the slip onto the mountant - this will trap air bubbles.

Once the mountant has filled in under the cover slip, you can finish arranging your larva(e) under the slip. By pushing the cover from one side, etc., you can roll the larva(e) to the position desired. Then, *gently* press down on the cover slip over the head capsule with your forceps, pencil eraser, etc. to spread the mouthparts, and over the anal end to spread the claws of the hind parapods.



Lay the slide on a flat surface and allow it to cure at room temperature for 2-3 hours. Check for air fingers; if any are present fill them in with more CMC and allow to cure for 1-2 hours. Then use clear fingernail polish or more CMC to ring the coverslips. Slides can be placed in a drying oven (do not exceed 55° C) for a day or two (perhaps longer at room temperature) and are then ready for examination.

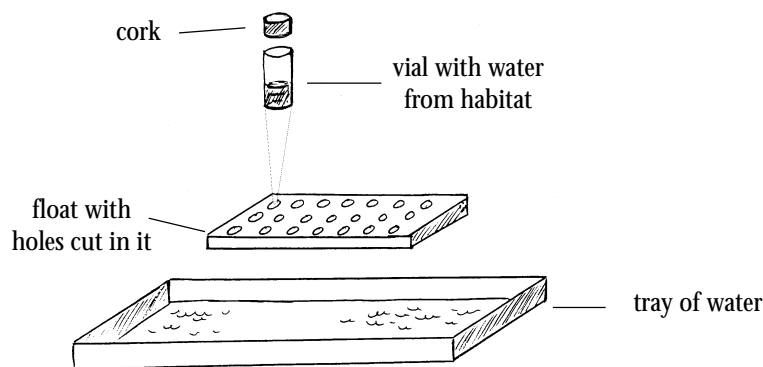
Rearing Larvae

Identification of some larvae is greatly aided by, or sometimes requires, associated pupal or adult material. Sometimes we get lucky and collect late fourth instar larvae in which developing pupal structures, such as the thoracic horn or various setal structures, can be observed; these can be particularly useful for separating genera of the *Thienemannimyia* complex or *Cricotopus* larvae from those of *Orthocladius*. Or one may collect pupae with larval exuviae still attached; sometimes pupae are found with developed adult genitalia within while the larval exuviae is still attached. Most often, though, one must collect living larvae and rear them to adulthood to make an association that may enable you to put a species name on a larva or with larval complexes that have not yet been deciphered.

Rearing refers to the process of allowing isolated larvae to go through the remaining stages of metamorphosis in order to associate the adult stage with the larval stage (and, of course, the pupal stage as well). Rearing most chironomid larvae is simple: collect live larvae in the field. Whether using a net or any other sampling device, place the material collected in a white (or any light color) pan, then sort through the detritus for larvae. Use an eye dropper for collecting and transferring larvae - do not pick up living larvae with forceps - they damage very easily! Place each larva in a separate 2 or 4 dram vial with a small amount of water from the habitat, and place a cork or other permeable stopper in it (don't use cotton unless you have nothing else; adults may lose legs and antennae in it). The vials can then be placed

in racks or floats; I use closed cell foam with holes bored through it that are just a bit smaller than the diameter of the vials. Maintain an even temperature. This may require using a cooler; larvae are difficult to overcool for short periods, but succumb easily to high heat.

If field time is limited, take a container (cooler) with samplers with live insects back to lab, and sort larvae there. In the lab, place the vials in rafts floating in water, or if air conditioned, they can probably be left at room temperature. I've had good results at temperatures ranging from 15°-20+° C. This will vary depending on what and where you have collected, time of year, etc. Lentic taxa tend to be easier to rear than lotic taxa; in the Southeast US, psammophilic taxa (sand dwellers) seem to be the most difficult to rear. Check vials daily. If you have collected fourth instar larvae, they may pupate, and eventually an adult may emerge. Allow the adult to harden for several hours or a day, and then knock it into the water with a squirt of alcohol. There you have it: an adult with its shed pupal and larval skins! Be sure to add preservative in an adequate strength (70-80% ethanol). Incomplete rearings (larva died in transition to pupa, or pupa died before adult emerged) can also be extremely valuable. Reared larvae may be sent to experts for identification. You could be the person who makes an important association which allows better identifications for everyone. And, rearing and observing live larvae, pupae and adults can be quite interesting, educational and even fun!



a simple rearing setup

Quality Assurance

An often neglected factor in macroinvertebrate studies is the quality assurance of identifications. Although stringent guidelines may be in place for sample collection methods, chemical analyses or statistical tests, etc., too often too little attention is paid to monitoring or improving the abilities of lab personnel performing identifications.

We're all human - we all make mistakes - and we're working with biological units that are subject to variation. It's easy to mislabel a slide or write the wrong thing down if one is momentarily distracted - perhaps the person next to you in the lab had a big meal of beans the night before, etc. I view these errors as "mechanical" mistakes that are basically inevitable; regular QA/QC procedures should handle or catch many of these types of errors.

Identifying something incorrectly is another story. Novices and masters - all of us - need the feedback concerning the accuracy of our identifications. Then why don't more workers have their work checked?

I can postulate a number of reasons. The number one reason related to me by most biologists is that, although they'd like to have their work verified, they don't have the funds to have their material checked by outside experts. This is *not* a good reason. After all, the beginning data points for any study involving organisms are the organisms, and everything should be done to insure the accuracy of those data. And, sorry to say, passing the specimens around the lab for various opinions as to their identity is in many instances not a satisfactory solution. Funds for verification by experts should be incorporated into every study. In the past, it was often possible to get "free" identification help from taxonomists at museums, universities, etc. In some cases, this is still possible, but for many groups it no longer is. Museums have cut back on personnel, retiring systematists at universities are replaced by ecologists or molecular biologists with insufficient training in taxonomy, and many of the "old guard" have passed on. There are not that many students studying systematics today - why should they when the prospects for a job in systematics are bleak? Talk about

"biodiversity" is cheap and hypocritical when systematics is not supported. Administrators must be made aware of the importance of correct identifications and the necessity (sometimes) to use an outside expert.

Reason number two is that many naive biologists actually believe they know what they're doing when it comes to identifying organisms. However, identifying an organism does not mean just running it through a key and coming up with an "identification" (see the section on Identifying Chironomidae above). If you don't have your work checked by someone who knows what he/she is doing, there is a good chance you have misidentified some taxa.

Reason number three is related to number two - some biologists believe that their work is so "good" that there is no reason to have it checked - it is a matter of personal ego. But this is supposed to be science we're doing, and a mainstay of science is the process of peer review.

And just in case there are non-believers out there who think that their work doesn't need to be checked, peruse the table below. It is a small sampling of the larval chironomid collections that I've examined recently. They come from a variety of sources: county, state and federal labs, universities and private consulting firms.

Collection	Number of specimens	Number misidentified	% misidentified
G	70	4	6
J	57	4	7
H	185	22	12
A	14	2	14
C	73	13	18
D	32	9	28
I	85	26	31
E	61	26	43
B	50	24	48
F	86	52	60

Can you imagine a collection with 60% of the specimens misidentified? Can you imagine writing a report utilizing such data? Imagine no longer, because it's been done!

Voucher specimens. All studies should have a collection of voucher specimens, i.e., specimens that are representatives of the organisms identified from that study. Ideally, such a collection would be verified by an expert.

Experts. Just who qualifies as an expert? How do you know whether you have a diamond or a cubic zirconium? A few things to consider:

The first thing one needs to realize is that mere possession of a Ph.D. does not mean that the bearer is an expert in taxonomy. The worst collections I've examined were "identified" by people with Ph.Ds. However, if that person earned his/her Ph.D. by doing a generic revision or similar systematic work, he/she could be considered expert at least with the taxon or taxa studied. Workers who have earned a Ph.D. may have been exposed to more serious taxonomic experience than those persons who have not spent as much time in a laboratory, but note that several North American experts do not hold a Ph.D.! Experience gained through years of working may be more instructive than taking courses, provided that the experience has been tempered with ample verification of identifications. Nothing beats the opportunity to examine material that has been correctly identified; some taxa bear nuances (or a "gestalt") that illustrations or descriptions don't quite bring out. In some cases it may even be necessary to study type material (but please note that such a circumstance would be relatively rare!).

Has your "expert" published on the taxonomy of the group in question? What we're talking about here is not papers dealing with new records for distribution or life history studies, but genuine taxonomic work, such as describing new taxa, redescribing taxa, generic revisions, reviews of museum specimens, etc. In lieu of published taxonomic experience, a conscientious worker with many years of

experience, an up to date library and an extensive collection of *verified* reference specimens might be considered as an expert. Quite often, benthologists have seen more material of some taxa than the museum taxonomist!

Is your "expert" able to confirm or deduce larval identities based on associated pupal or adult stages? Without such an ability, it would seem hard to deem such a worker as an expert, although, as outlined above, there are many workers who have a great deal of expertise with only one life stage.

In general, we chironomid taxonomists have our areas of taxonomic and geographic specialization; if we have a specimen whose identity is unclear, we tend to send it to the worker who is the specialist with that group (usually the last person that did any taxonomic work on it - barring those who have already passed on) or region. A major problem is that there are not enough people doing genuine systematic research (and publishing it!) with Chironomidae in North America - the active publishing experts can be counted on two hands, and several fingers will be left over. Don't forget that these experts are often fooled (just read the literature!) and biological entities seem sometimes to get a bit cranky if you try to put a name on them. For instance, although I've worked with the genus *Dicrotendipes* for over 25 years, I still can not consistently separate some specimens of the common species *D. modestus*, *D. tritonus* and *D. neomodestus*, in either the larval, pupal or adult stages. Whether this is due to "natural" variation, hybridization or the presence of unrecognized, cryptic species is not known. It still is not possible to separate at the genus level many species of *Orthocladius* larvae from those of *Cricotopus* and/or *Paratrichocladius*, although with dedication, larval/pupal/adult associations and lots of experience, the larvae of many species of these three genera can be identified at the species level.

If you learn one thing from this manual, it should be:

You must have your identifications verified by a qualified expert!!

Literature

One of the most important things a good taxonomist has, in addition to lots of experience and a collection of correctly identified reference specimens, is an extensive, up to date library. Keeping up with the literature can be a daunting task, but today that task is aided by such helpful things as the annual NABS bibliographies and other literature available from Internet sites such as the Chironomid Home Page (see Sources).

Literature that should be in every lab is listed below. Literature that is not to be trusted is Beck (1976, 1979) and Webb & Brigham (1982); these publications are fraught with mistakes and misinformation.

If you're going to seriously work with Chironomidae, you should at a minimum obtain the following literature:

Armitage, et al. 1995
 Coffman & Ferrington 1996
 Epler 1995
 Oliver et al. 1990
 Oliver & Dillon 1994b
 Sæther 1980a
 Wiederholm 1983, 1986, 1989

Regional Guides

Epler 1995
 Oliver et al. 1978
 Simpson & Bode 1980

Other handy literature, including bibliographies, checklists, works on eggs, pupae and adults; some of these publications deal with the fauna of other areas, but they may include some taxa that occur in the Nearctic or may eventually be found here.

Ashe 1983
 Caldwell et al. 1997
 Fittkau 1962
 Fittkau et al. 1976
 Hoffrichter & Reiss 1981
 Hudson et al. 1990
 Langton 1991
 Nolte 1993
 Oliver & Roussel 1983b
 Pinder 1978
 Roback 1957, 1971
 Schmid 1993
 Simpson 1982
 Simpson & Bode 1980
 Spies & Reiss 1996
 Townes 1945

Sources

Listed below are sources for laboratory materials, literature and additional information. Note that these are my recommendations and that the mention of a company, product or service does not indicate endorsement by any government agency!

Lab Materials:

CMC mounting medium:
Masters Company, Inc.
890 Lively Blvd.
Wood Dale, IL 60191
(630) 238-9292
Fax: (630) 238-9297

General lab products:
VWR Scientific Products
<http://www.vwrsp.com>

Entomology equipment:

BioQuip Products
17803 LaSalle Avenue
Gardena, CA 90248-3602
(310)-324-0620
e-mail: bioquip@aol.com

BioQuip is the best source for almost all entomological equipment and many books.

Livesay's, Inc.
456 West Columbus Drive
Tampa, FL 33602
(813)229-2715

Source for extra fine point Dumont number 5 forceps (Swiss made; expensive, but the best).

Literature:

Unfortunately, the larval volume (Wiederholm 1983) of the Holarctic Keys is out of print. The other Entomologica Scandinavica Holarctic Keys and other Ent. scand. chironomid related papers can be purchased from the following:

Scandinavian Entomology Ltd.
P.O. Box 24
S-240 17 S. Sandby,
Sweden
e-mail: Lennart.Cederholm@zool.lu.se

Entomologica Scandinavica on the web:

<http://darwin.biol.lu.se/systzool/zoomus/ZooDoc/Publ> will take you to an index; click on the ESS numbers for a listing of Entomologica Scandinavica Supplements and their prices.

The Holarctic Keys are also available from:

Apollo Books
Kirkeby Sand 19
DK-5771 Stenstrup
Denmark
email: apollobooks@vip.cybercity.dk
web: <http://www.apollobooks.com>

Information sources on the World Wide Web:

Chironomidae and Water Beetles of Florida:
<http://www.concentric.net/~jhepler/index.html>
Features checklists of the Chironomidae of Florida, North Carolina, South Carolina and updates, additions and corrections to Epler's manuals, including this current manual.

The Chironomid Home Page

<http://www.ouc.bc.ca/fwsc/iwalker/intpanis/>

Central source of general information on Chironomidae and chironomid workers; includes a world-wide directory of chironomid workers and access to an extensive bibliography dealing with Chironomidae.

Chironomidae-L listserv

A listserv is like a distribution house for communications on certain subjects. Subscribers send in e-mail messages to a central address, and the computer there sends that message out to all subscribers. To subscribe, simply send an e-mail message to [<majordomo@cf.ac.uk>](mailto:majordomo@cf.ac.uk) with the text: "subscribe Chironomidae-L".

A Tour of the Subfamilies

As noted before, seven of the described subfamilies of Chironomidae occur in North America. We'll take a short morphologically-based tour of these subfamilies in presumed phylogenetic order, beginning with the most "primitive". Note that characters referred to below pertain to North American members of the subfamilies; in other parts of the world some subfamilies have members which may differ. This tour is followed by a key to the subfamilies of the southeastern United States.

Subfamily Telmatogetoninae

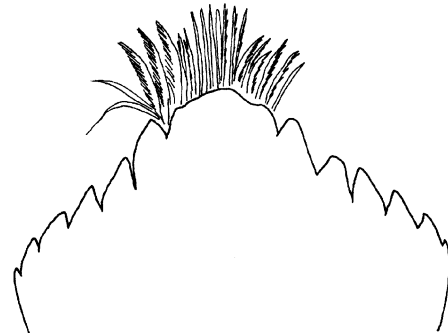
Recognized by the short 4 segmented antennae (less than 1/5 the length of the mandible), well developed mentum, the dense median brush of the prementum, and the lack of procerci and anal tubules. Two genera occur in North America; *Telmatogeton* is found on both coasts north to and including Canada; *Thalassomya* is apparently restricted to south Florida. All Nearctic members of this subfamily are marine coastal organisms, where they usually occur on algae on rocks, but note that some Hawaiian *Telmatogeton* species have invaded freshwater.



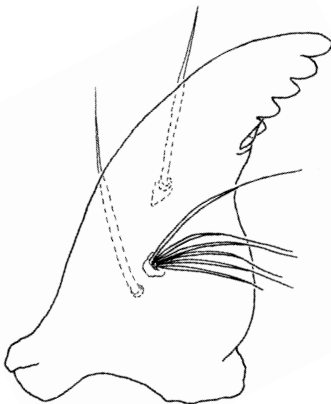
4 segmented antenna of *Telmatogeton*



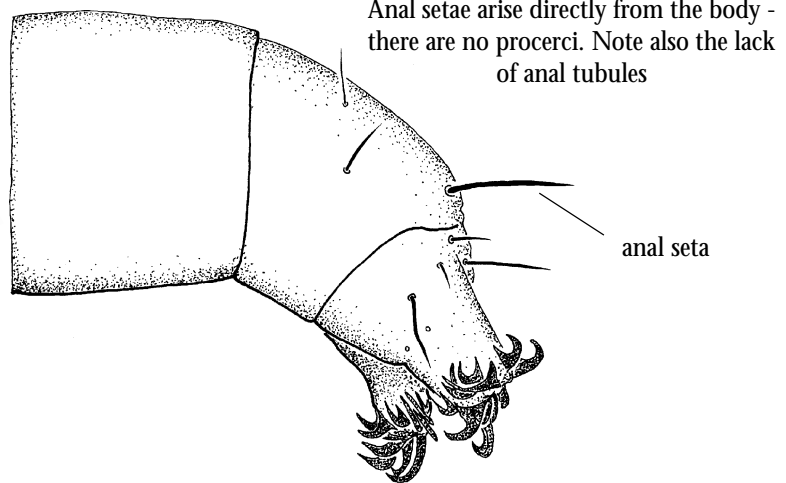
Telmatogeton prementum with dense median brush



Thalassomya mentum with brush of prementum visible dorsal to it



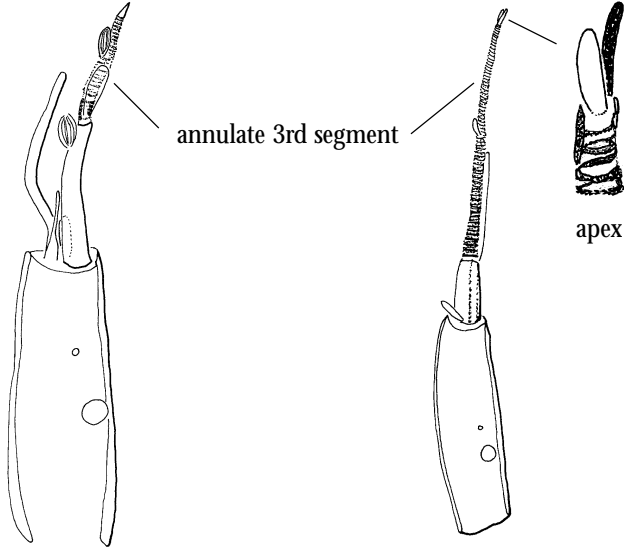
Telmatogeton mandible



posterior segments of *Telmatogeton*

Subfamily Podonominae

Recognized by annulate 3rd antennal segment, lack of premandibles, a well developed mentum and long, well developed procerci. Five genera are found in North America. Podonomines are usually uncommon and restricted to cold springs, brooks and streams where the larvae are often associated with mosses.

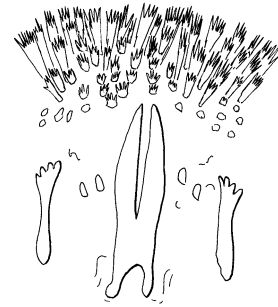


annulate 3rd segment

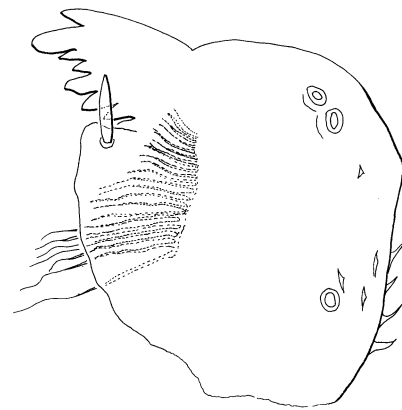
apex

Paraboreochlus antenna

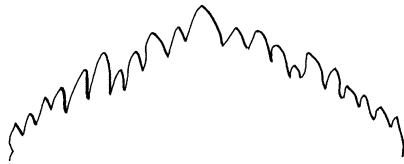
Boreochlus antenna



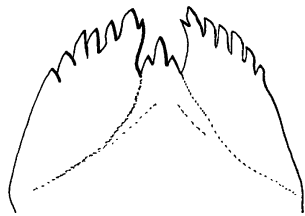
Boreochlus premento-hypopharyngeal area



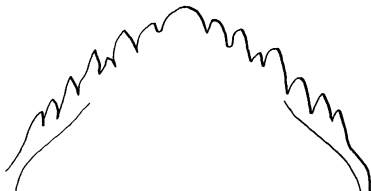
Paraboreochlus mandible



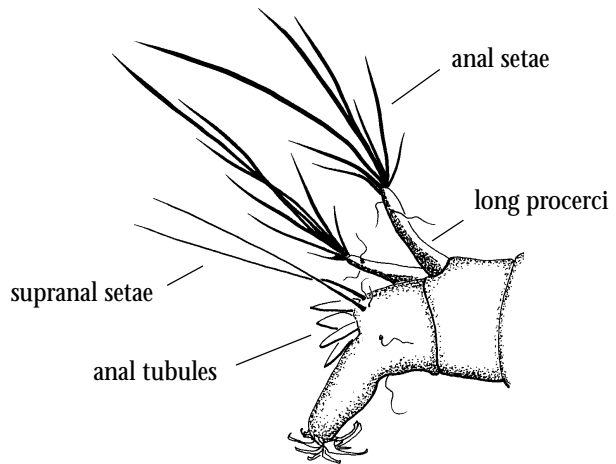
Lasiodiamesa mentum



Trichotanypus mentum



Paraboreochlus mentum



anal setae

long procerci

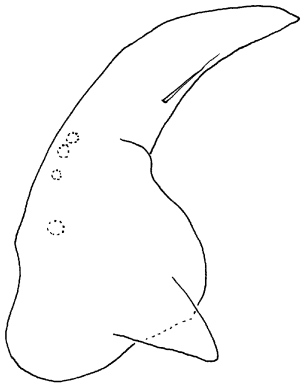
supranal setae

anal tubules

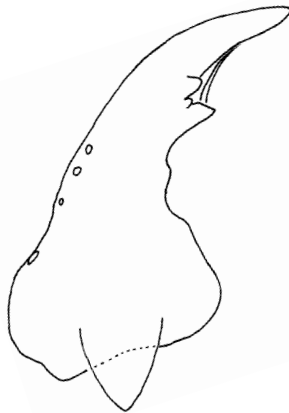
posterior segments of *Paraboreochlus*

Subfamily Tanypodinae

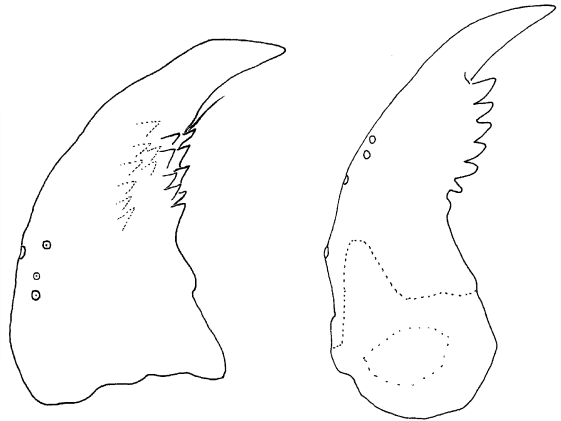
Members of this subfamily are easily recognized by the retractile antennae, lack of premandibles, a well developed ligula and well developed procerci. The setae and sensory pits of the head capsule are also very useful in identifying genera and some species. These setae were reviewed in detail by Kowalyk (1985). At least 39 genera occur in the Nearctic. All tanypods are predacious; larvae are found in all types of water bodies, including brackish water. Some genera possess hemoglobin and can live in low oxygen environments.



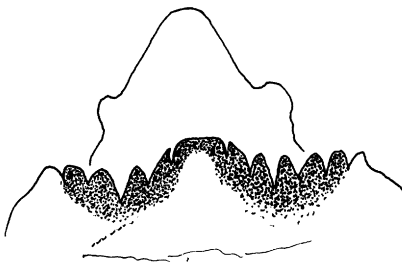
Tanypod mandibles can run from the extremely simple one of *Rheopelopia*



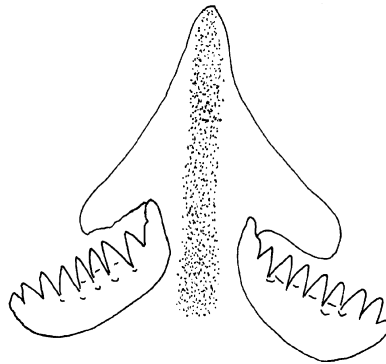
to one with a few teeth (*Paramerina*)



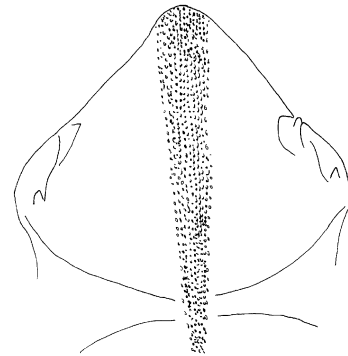
to one with many teeth (*Fittkauimyia*, *Psectrotanypus*)



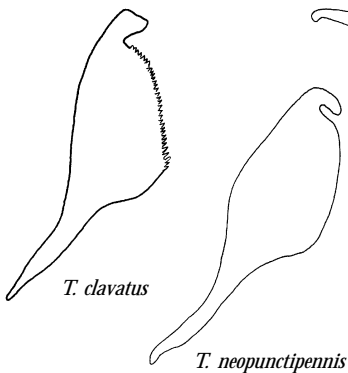
The mentum may be complete (*Tanypus*)



consist of two divided plates (*Procladius*)

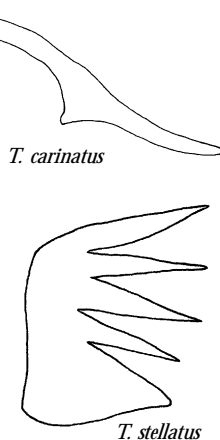


or have only a few widely separated blunt "teeth" (*Ablabesmyia*)



T. clavatus

T. neopunctipennis



T. carinatus

T. stellatus



Ablabesmyia

Clinotanypus

Fittkauimyia

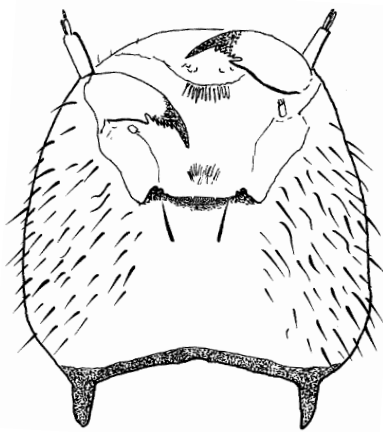
Labrundinia

Claws on the posterior parapods can be of importance; above are 4 examples from the genus *Tanypus*

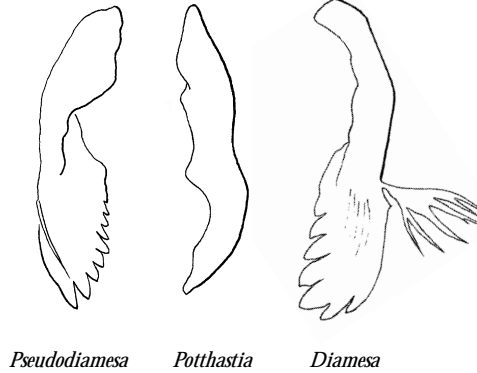
The ligula is also very important in tanypod taxonomy

Subfamily Diamesinae

Most Nearctic genera have an annulate 3rd antennal segment, a well developed mentum and premandibles and 3 well developed brushes of setae on the prementum. *Protanypus*, the sole Nearctic diamesine without an annulate 3rd antennal segment, can be recognized by the numerous setae on the head capsule, which also bears two long ventrolateral posteriorly directed processes. Larvae of the *Potthastia longimana* group lack teeth on the mentum. Ventromental plates may be vestigial to well developed; in some genera they obscure the teeth of the mentum. There are no beard setae associated with the ventromentum. Procerci may be present or absent. Eleven genera occur in the Nearctic, with at least one additional "genus" from the SE US known only as a larva. Diamesines are most often found in cold or cool flowing water, but also are found in springs and lakes.

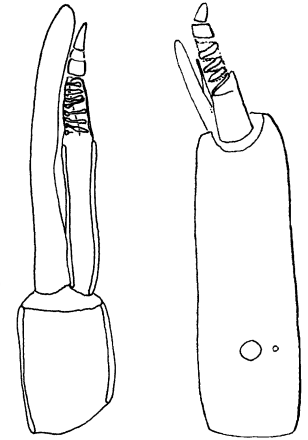


Protanypus head capsule



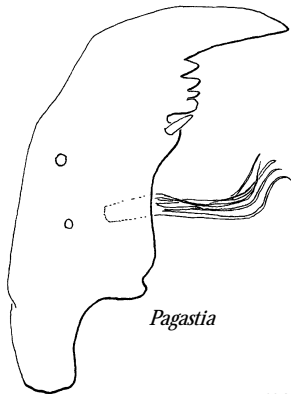
Pseudodiamesa *Potthastia* *Diamesa*

premandibles



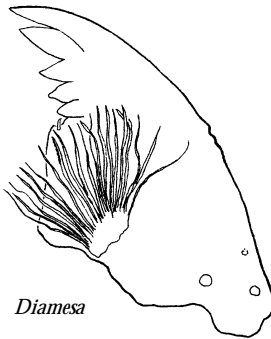
Potthastia longimana grp. *Diamesa*

antennae

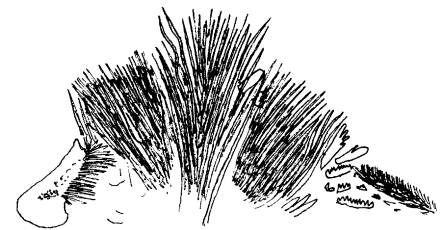


Pagastia

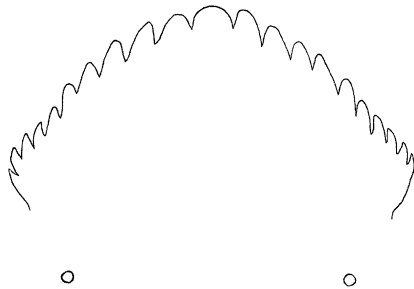
mandibles



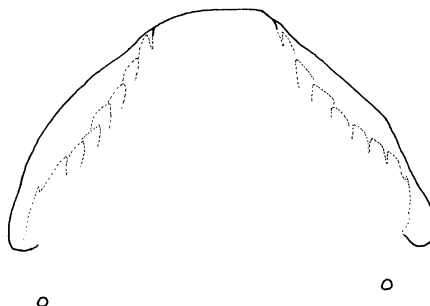
Diamesa



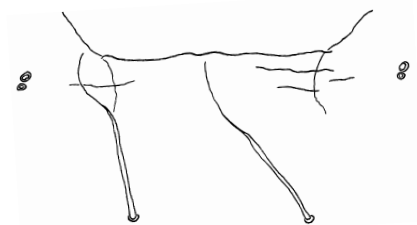
Diamesa prementum
with 3 large brushes



Diamesa mentum



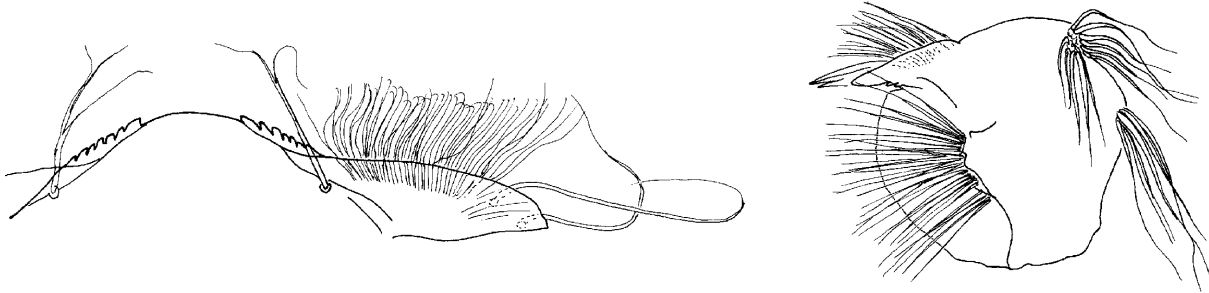
Potthastia gaedii grp. mentum



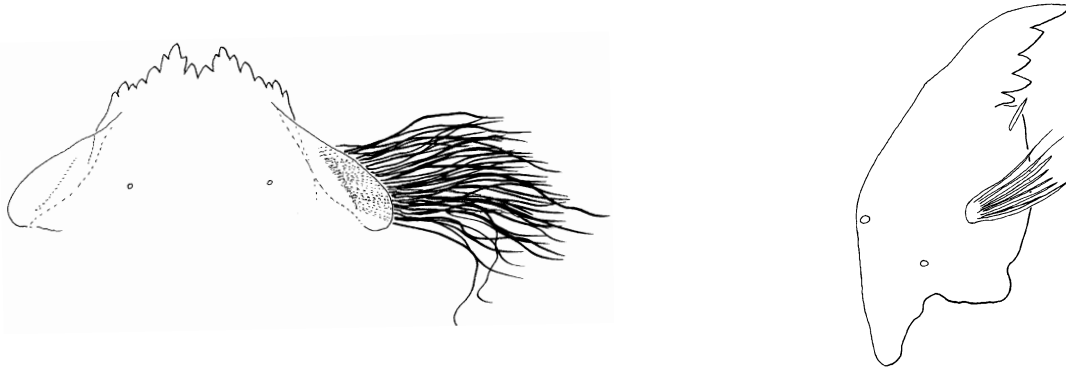
Potthastia longimana grp. mentum

Subfamily Prodiamesinae

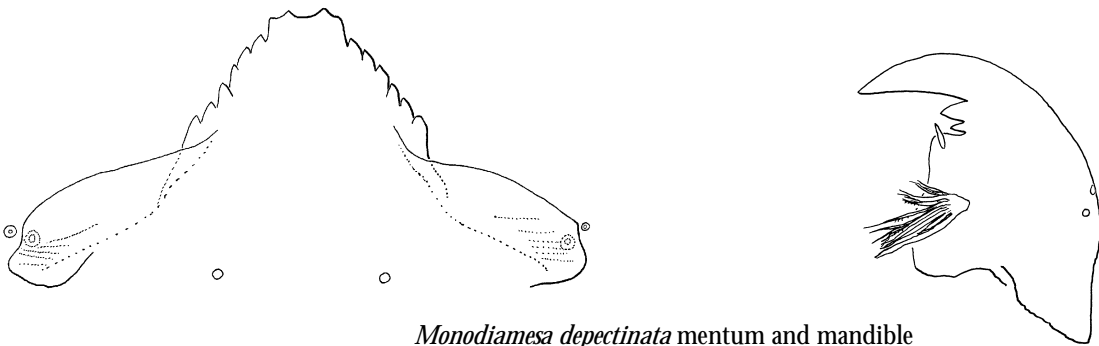
Recognized by the 4 segmented antennae, with segments 3 and 4 very small; well developed premandibles, prementum without a brush or brushes of setae and the well developed mentum with large unstriated ventromental plates, these plates with setae (often very long) beneath or adjacent to them. Four genera are known from the Nearctic, where larvae are found in springs, streams/streams, ponds and the littoral zone of lakes.



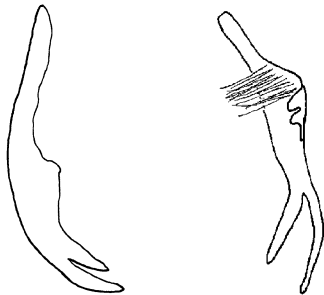
Odontomesa fulva mentum and mandible



Prodiamesa olivacea mentum and mandible



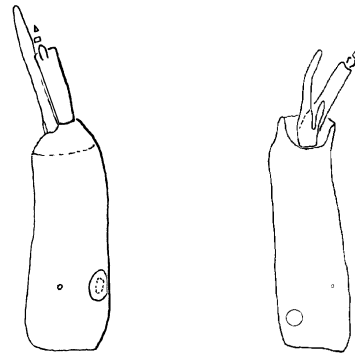
Monodiamesa depectinata mentum and mandible



Prodiamesa olivacea

Odontomesa fulva

premandibles



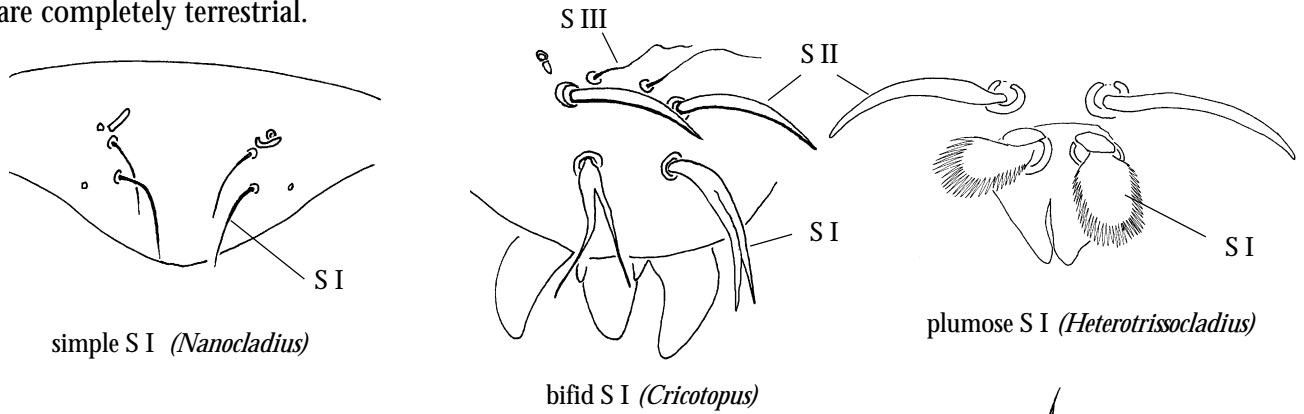
Monodiamesa

Odontomesa fulva

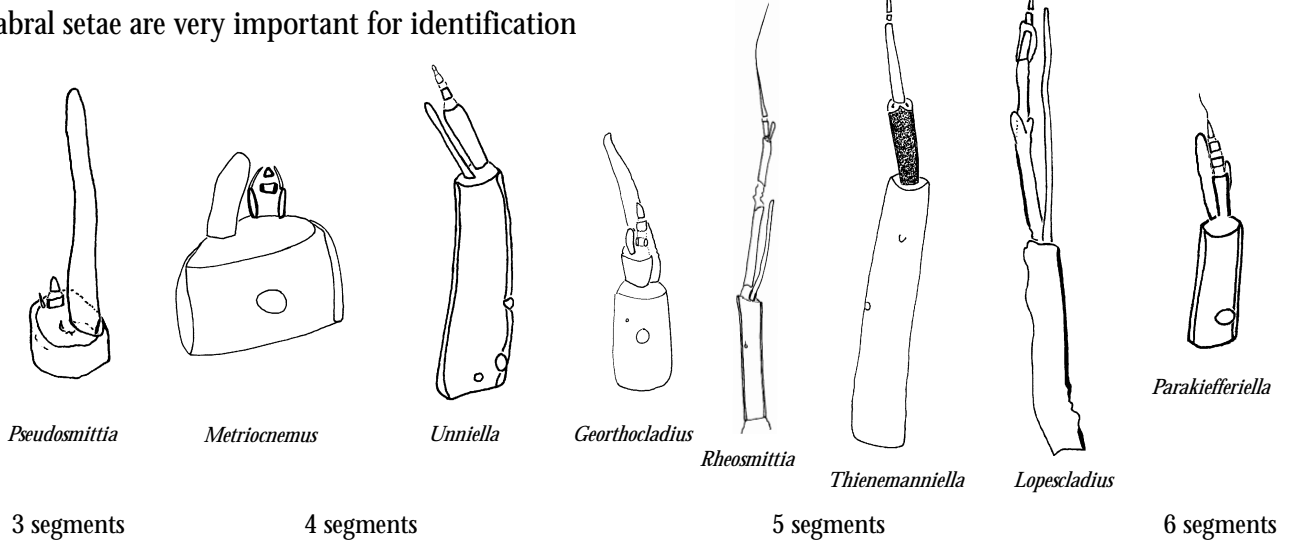
antennae

Subfamily Orthoclaadiinae

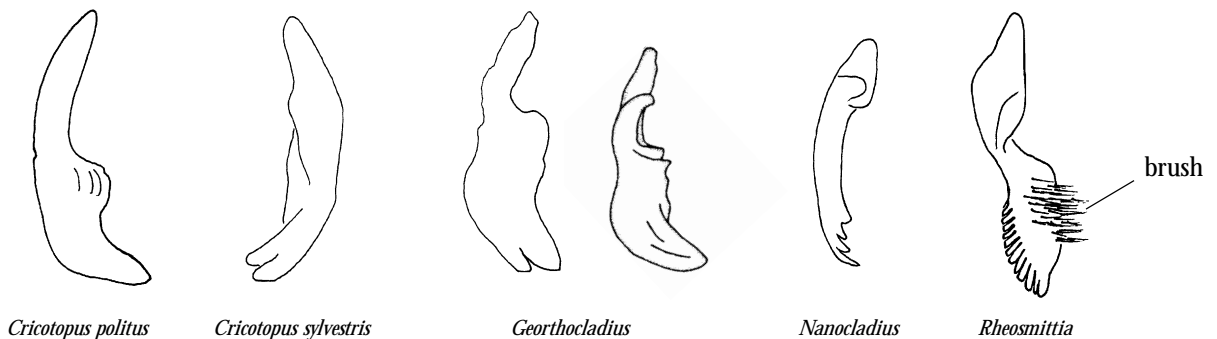
A morphologically and ecologically diverse subfamily, usually with well developed antennae (although sometimes strongly reduced) with 3-7 segments, a prementum without a dense brush or brushes of setae; well developed premandibles, and a well developed mentum, with or without unstriated ventromental plates (although weak ridges may be present in some taxa), with or without setae adjacent to or beneath them; some terrestrial/semi-aquatic larvae lack procerci, anal tubules and/or anterior and posterior parapods. At least 81 described genera occur in North America, with several additional taxa that probably represent new genera. Larvae are found in all aquatic habitats, including coastal marine areas; some taxa are completely terrestrial.



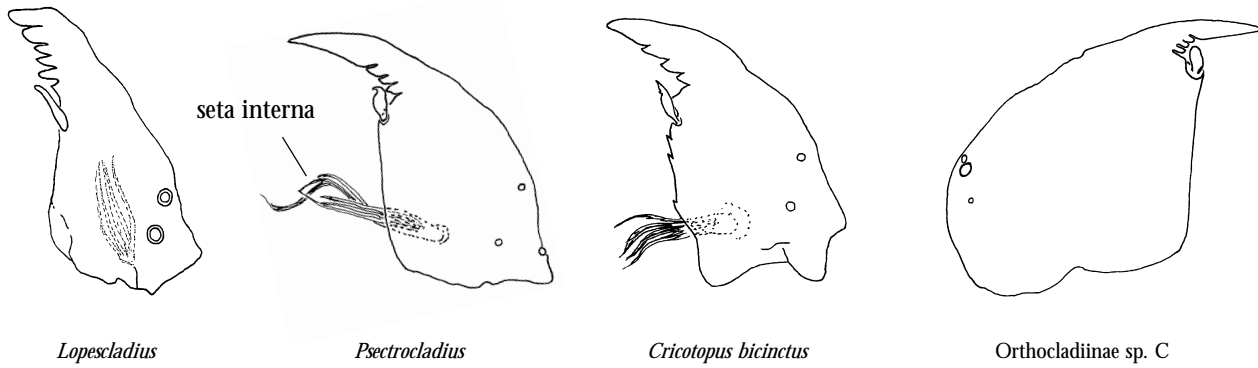
Labral setae are very important for identification



Orthoclad antennae exhibit a wide range of segmentation and design



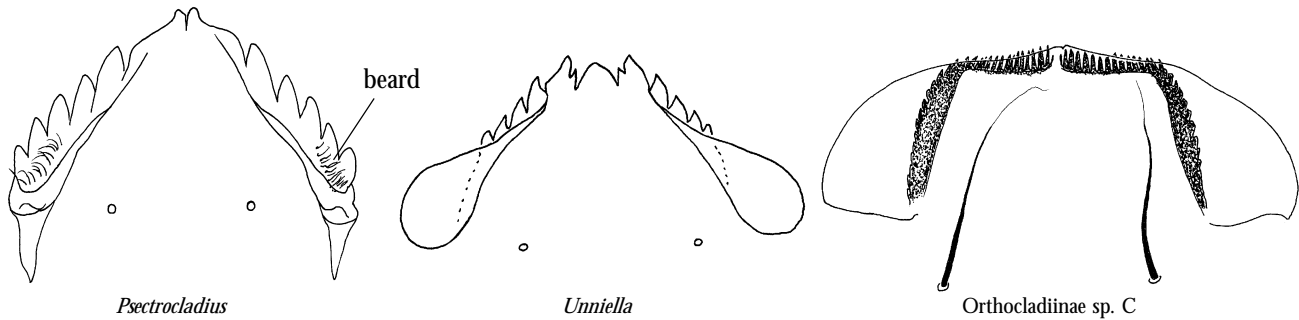
Premandibles may be simple or multi-toothed, with or without a brush of setae



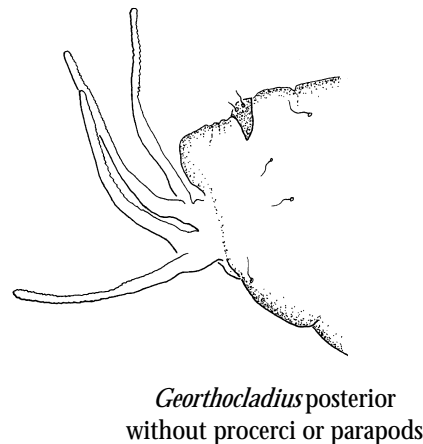
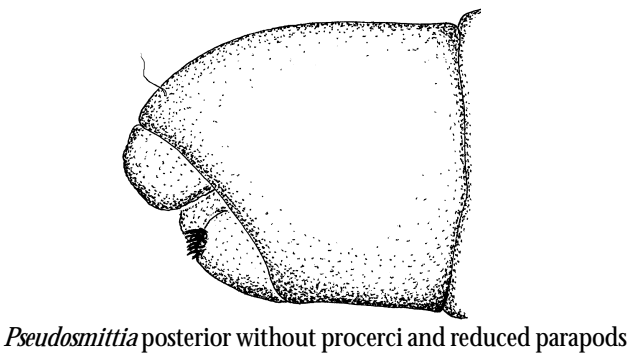
Orthoclad mandibles offer many characters for identification - their shape, number of teeth, presence or absence of a seta interna, and numerous other features



The mentum may lack ventromental plates, or ...



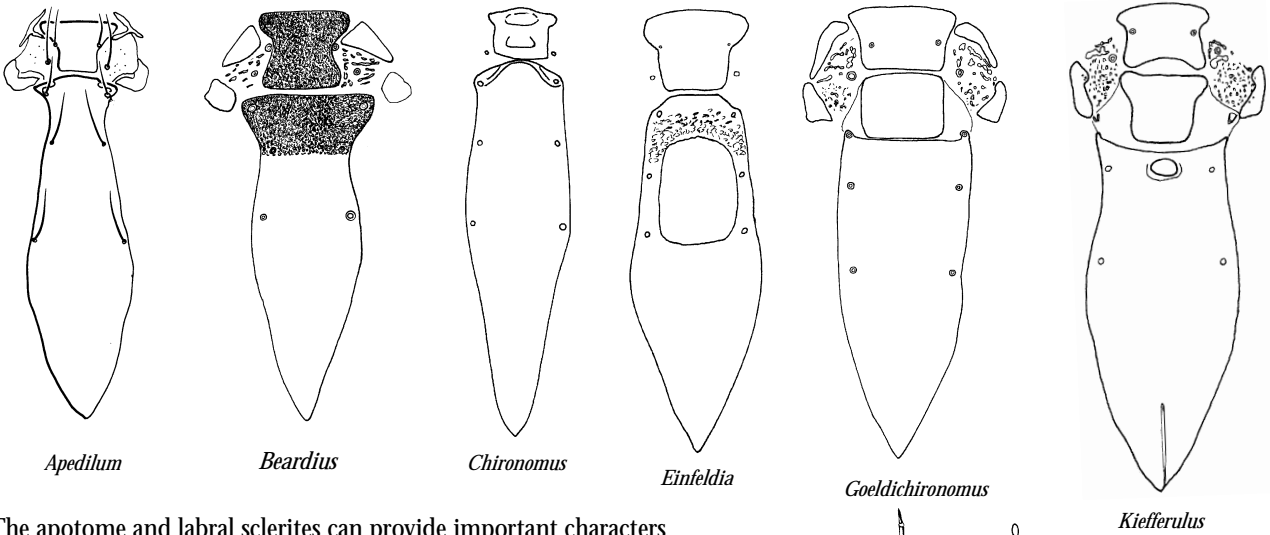
ventromental plates may be small to large and may have a beard beneath or adjacent to them



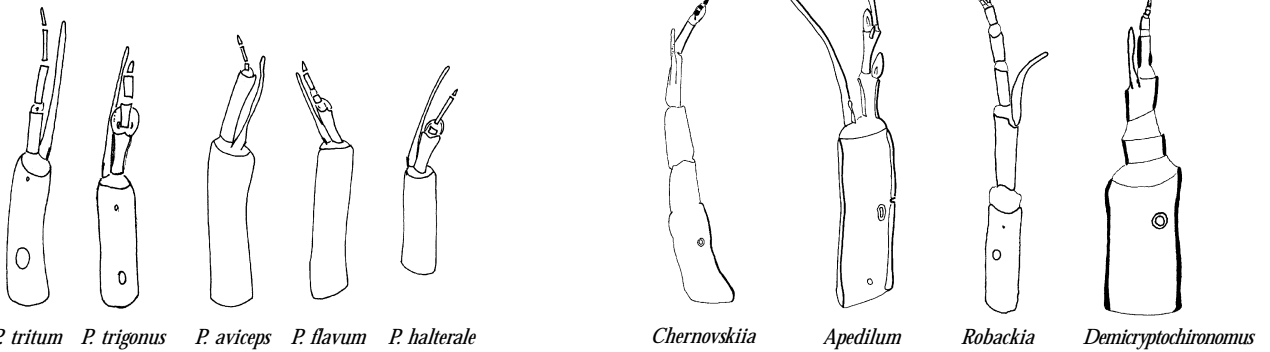
Some species modified for terrestrial or semi-terrestrial life have reduced or vestigial parapods, proceri and/or anal tubules

Subfamily Chironominae

Nearctic genera possess antennae with 5-8 segments; premandibles are present; the prementum does not bear large brushes. Most Nearctic chironomines possess a well developed mentum with striated ventromental plates (these plates are reduced and unstriated in the leaf-mining/wood-boring genera *Stenochironomus* and *Xestochironomus*); a beard is not present in members of this subfamily. Two pairs of anal tubules are usually present; some genera may bear additional lateral and/or ventrolateral tubules; procerci and parapods are usually well developed. At least 71 described genera are found in North America, with several additional undescribed taxa that probably represent new genera. Larvae are found in all aquatic habitats, including coastal marine areas; some taxa can withstand extended periods of desiccation.

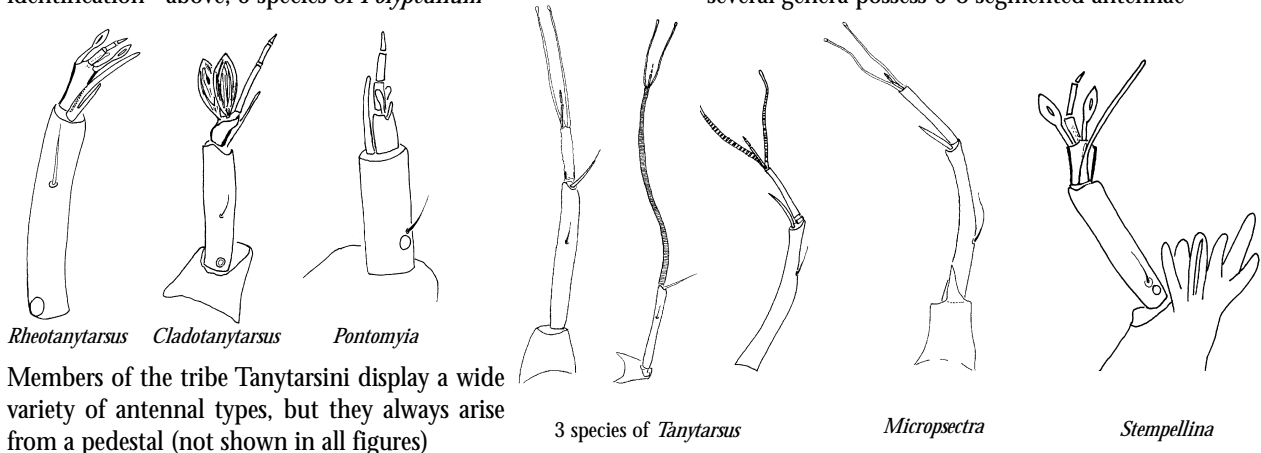


The apotome and labral sclerites can provide important characters

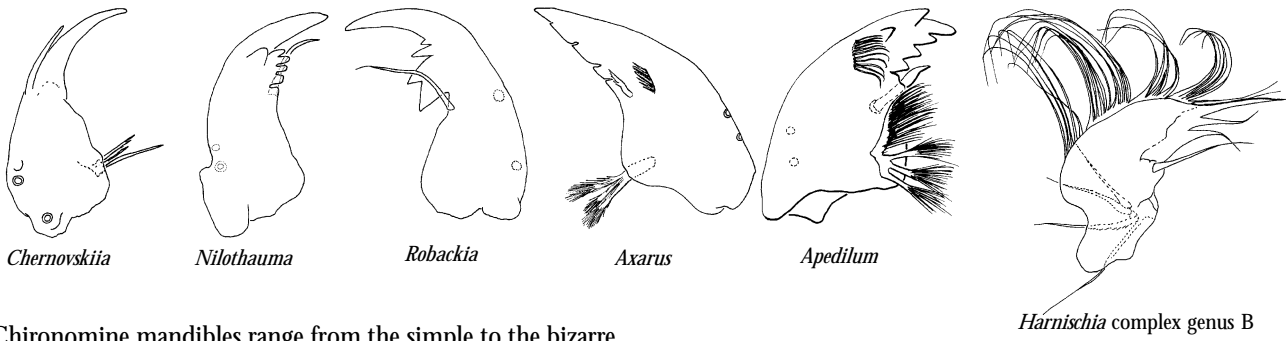


Antennae may provide important characters for species identification - above, 5 species of *Polypedilum*

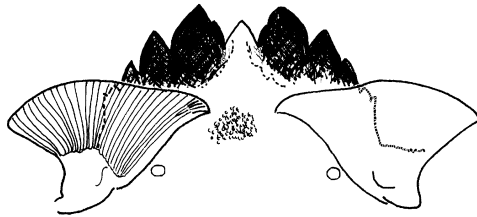
Although most Chironominae have 5 segmented antennae, several genera possess 6-8 segmented antennae



Members of the tribe Tanytarsini display a wide variety of antennal types, but they always arise from a pedestal (not shown in all figures)



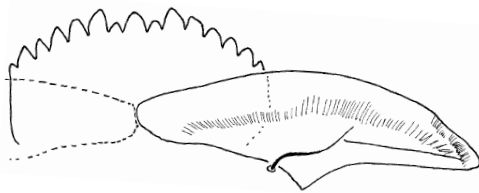
Chironomine mandibles range from the simple to the bizarre



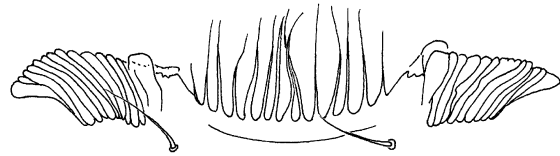
Beardius



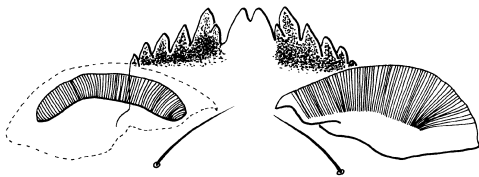
Stenochironomus



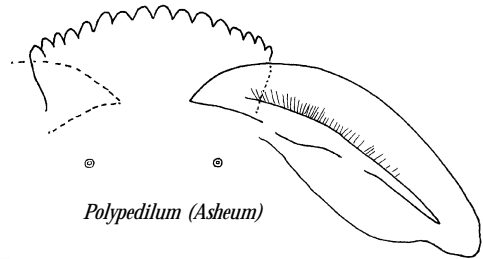
Axarus



Harnischia complex genus B



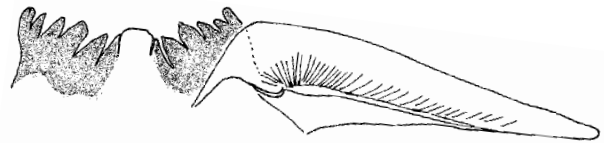
Apedilum



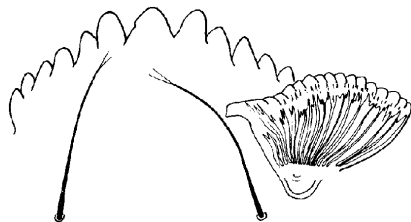
Polypedilum (*Asheum*)



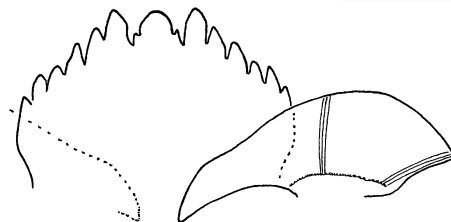
Chernovskia



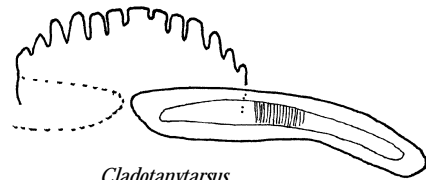
Cryptochironomus



Dicrotendipes



Goeldichironomus



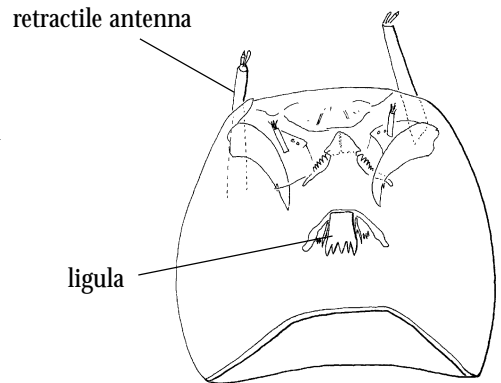
Cladotanytarsus

For identification purposes, the mentum and ventromental plates provide some of the most important characters, such as the number and arrangement of teeth on the mentum, its shape and the shape of the ventromental plates and the number of striae on them

Key to the subfamilies of Chironomidae of the southeastern United States

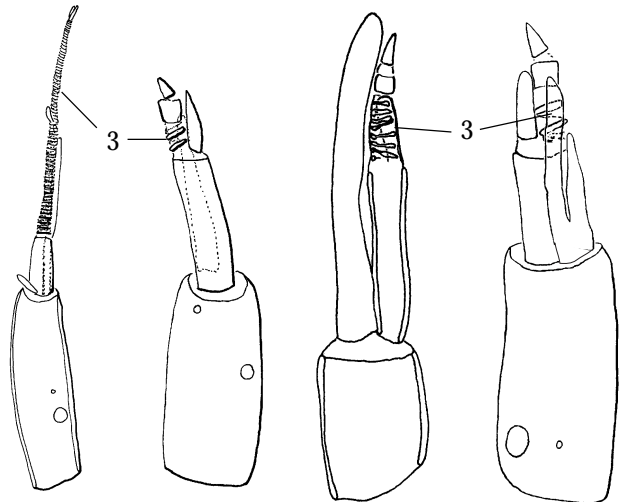
Use this key in conjunction with the figures found in the "Tour of the Subfamilies" beginning on page 1.32

1 Antennae retractile within head capsule; a large well sclerotized ligula present ... **Tanypodinae** (Chapter 4)



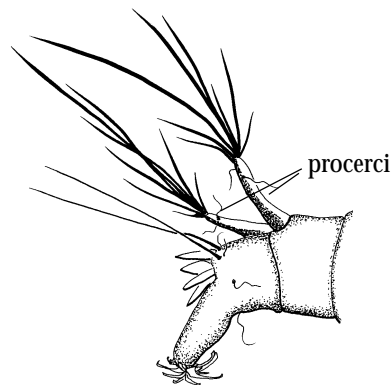
1' Antennae not retractile within head capsule; a well sclerotized ligula not present 2

2(1') Third antennal segment annulated .. 3

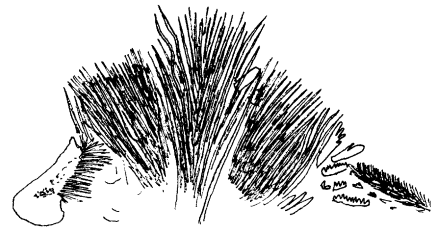


2' Third antennal segment never annulated (second antennal segment or Lauterborn organ pedicels may be annulated) 4

3(2) Premandibles absent; prementum without 3 large brushes of setae; procerci well developed, very long **Podonominae** (Chapter 3)

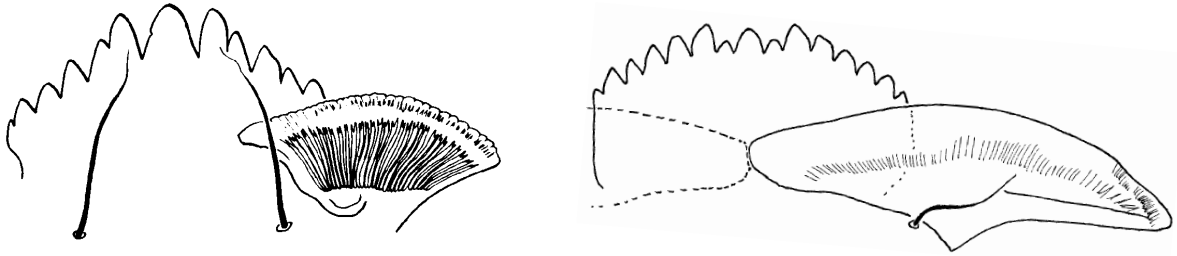


3' Premandibles present; prementum with 3 dense brushes of setae; procerci vestigial or small, never long **Diamesinae** (Chapter 5)



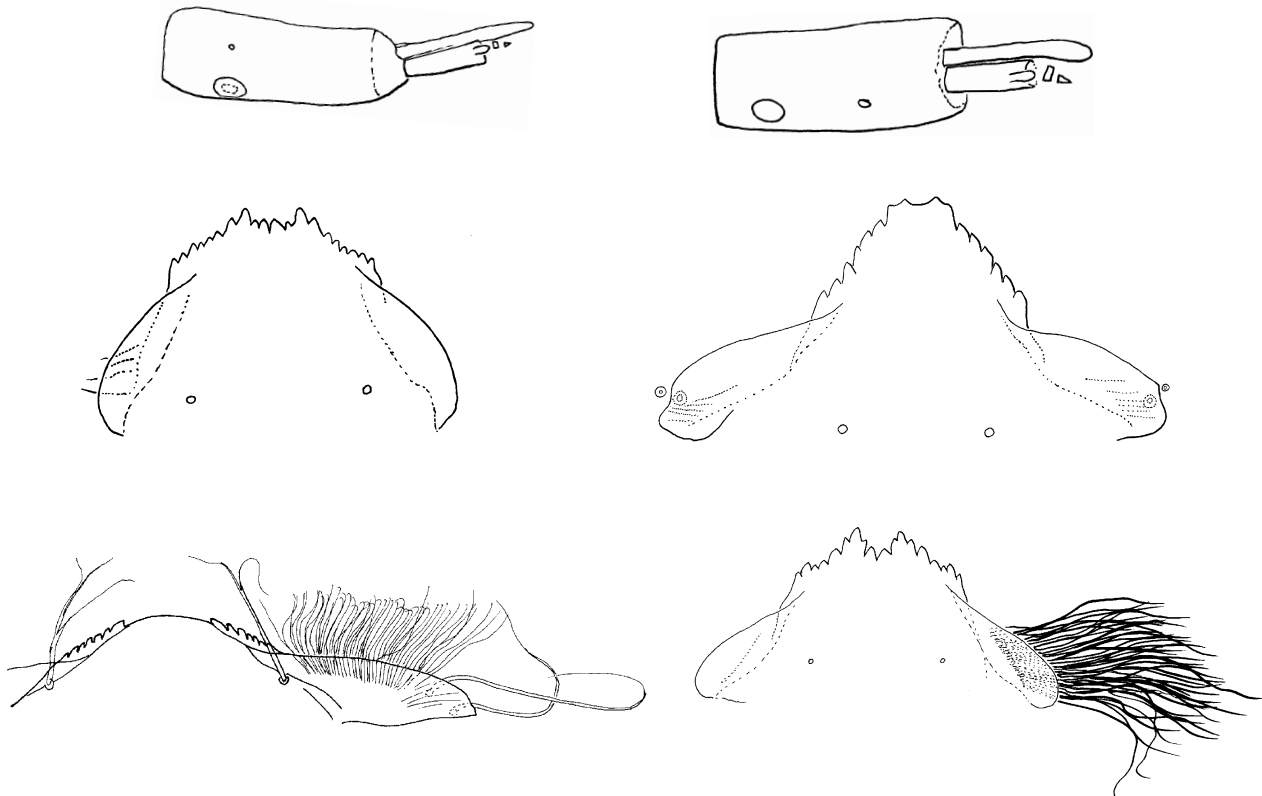
prementum

4(2') Striated ventromental plates present; no beard (setae) present beneath ventromental plates ..
..... **Chironominae** (in part) (Chapter 8)



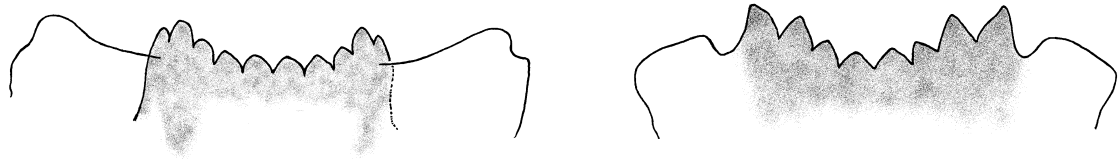
4' Ventromental plates, if present, without striae (some *Nanocladius*, p. 7.89, may have a few markings on ventromental plates) 5

5(4') Ventromental plates strongly expanded, with beard (setae) beneath (beard very well developed in 2 genera); antennae 4 segmented, with segments 3 and 4 very small **Prodiamesinae** (Chapter 6)



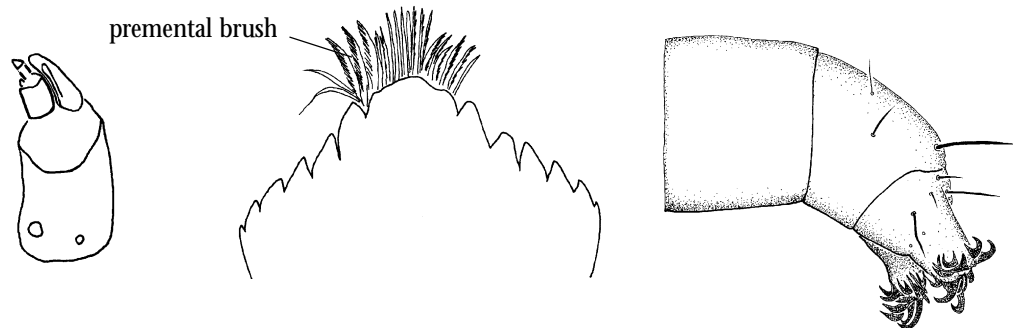
5' If ventromental plates strongly expanded, then beard either absent or antennae not 4 segmented ..
..... 6

6(5') Ventromental plates fused to maxillae so not apparent; mentum concave; head flattened dorsoventrally, chisel-shaped; larvae mining in dead submerged wood and/or submerged leaves; may be common in Hester-Dendy samplers constructed of plant materials
..... **Chironominae** (in part) (Chapter 8)



6' Mentum, ventromental plates and head not as above; in a variety of habitats that may include submerged dead wood or leaves 7

7(6') Antennae short, 4 segmented; prementum with well developed median brush; beard, procerci and anal tubules absent; exclusively marine in Southeast **Telmatogetoninae** (Chapter 2)



7' Antennae 3-7 segmented; prementum without well developed median brush; beard present or absent; procerci and anal tubules present or absent (usually present, but often absent in terrestrial genera); larvae in a variety of habitats, but if marine, then antennae with 5 segments
..... **Orthoclaudiinae** (Chapter 7)

