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“In the end, we will conserve only what we love; we will love only what we understand and we will understand only what we are taught.”

Baba Dioum

Preface

Aleocharine beetles are among the most poorly known and difficult-to-identify, megadiverse groups of Coleoptera worldwide. Here, we present the first comprehensive synopsis of all aleocharine rove beetle species (Coleoptera, Staphylinidae) recorded from eastern Canada, from Ontario to the Atlantic provinces. This is the first book to treat and illustrate every valid species of this taxonomically difficult and poorly known group of Coleoptera from a large area of the Nearctic region. For many species the illustrations included herein are previously unpublished or are presented here in a more complete way, and include colour habitus and genital diagnostic structures of both sexes. Four hundred and seven valid species (including one newly recorded for Canada) in 96 genera, and 16 tribes are presented. Tribes and subtribes are arranged in phylogenetic order as it is currently recognized. Species are listed alphabetically or in species groups to better reflect their relationships. Species distribution is listed by provinces and territories in Canada and states in the United States, and the geographic origin of each species is categorized as native, Holarctic, adventive or undetermined (either adventive or Holarctic). Every species is presented with a morphological diagnosis including external and genital characters of both sexes. Collection and habitat data are presented for each species, including collecting period, and collecting methods. A list of all eastern Canadian species with their currently known distribution in North America is presented at the end of the book (Appendix A). In concert with this book, we are providing DNA barcode sequences for 104 species in 47 genera treated herein.

NOMENCLATORAL CHANGES. **New synonyms** (first name is valid): *Amarochara duryi* (Casey, 1906) [*Pachycerota*] = *Amarochara formicina* Assing, 2007, and genus *Pachycerota* Casey, 1906 is synonymized with *Amarochara* C.G. Thomson, 1858; *Atheta* (*Alaobia*) *trinotata* (Kraatz, 1856) = *Philhygra pohli* Klimaszewski and Langor, 2011; *Cyphea curtula* (Erichson, 1837) = *Agaricomorpha vincenti* Klimaszewski and Webster, 2016; *Dinaraea aequata* (Erichson, 1837) = *Dinaraea quadricornis* Klimaszewski and Webster, 2013; *Oxypoda sylvia* Casey, 1906 = *Moluciba grandipennis* Casey, 1911. **New lectotype designations:** for *Amarochara duryi* (Casey), and *Oxypoda sylvia* Casey. **New combinations:** *Agaricochara aldersonae* (Klimaszewski and Webster, 2016), described as *Gyrophana*; *Amarochara duryi* (Casey, 1906), described as *Pachycerota* Casey; *Pelioptera americana* (Gusarov, 2002), described as *Tropimenelytron*; *Pelioptera thujae*

(Klimaszewski and Webster, 2016), described as *Atheta*. **New taxa:** *Gatineauleptusa* Klimaszewski, **new subgenus**. **New record:** *Borboropora quadriceps* (LeConte), new genus and species for Canada. **Removed from distribution list in Canada:** *Platyusa sonomae* Casey, 1885 [unlocated voucher specimen/s, erroneous record].

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Introduction

1

The Staphylinidae is the largest family of beetles, consisting of more than 63,495 described species in 3762 genera worldwide, and includes 1774 valid species recorded in Canada (Newton, unpublished database, December 2017). The Aleocharinae is by far the largest subfamily of rove beetles with 16,537 known species in 1310 genera and 62 tribes worldwide (Newton, unpublished database, December 2017). Aleocharinae is generally regarded as one of the most poorly known and most taxonomically challenging groups of Coleoptera worldwide. In Canada, there are 603 known species, and many species remain yet undiscovered including some that are undescribed. Aleocharinae constitutes at least 26% of all species within the family Staphylinidae. Rove beetles, including aleocharines, are useful as ecological indicators of changes in managed forests because of their highly specific microhabitat associations, response to disturbances, high local abundance and species, habitat, and trophic diversity, ease of collection, and availability of identification keys for many groups (Paquin and Dupérré 2002, Pohl et al. 2008, Venier et al. 2017, Klimaszewski et al. 2018). Usually, rove beetles represent a large proportion of the total abundance and species richness of organisms collected in pitfall traps, a sampling method frequently used to sample epigeic arthropods. For example, Klimaszewski et al. (2005, 2007, 2008) reported that rove beetles were five times more abundant and three times more species rich

than ground beetles, another taxon commonly used in the evaluation of impacts of forest management, in yellow-birch-balsam forests in Quebec. In another pitfall study of epigeic fauna in the Cordilleran forests of western Alberta, the number of rove beetle species (including aleocharines) was two times greater and the number of specimens three times greater than that of carabid beetles (Langor, unpublished data). As well, rove beetles are present in high abundance and diversity in dead wood (Langor et al. 2008). Unfortunately, aleocharine assemblages are often neglected in biodiversity studies, including those that consider Staphylinidae, because of the difficulty in identifying many groups due to the dearth of comprehensive modern taxonomic treatments and identification keys and the labor-intensive need to dissect the genitalia of a large proportion of specimens to facilitate identification. However, we hope that this will change (at least in eastern Canada) with this publication that provides the necessary taxonomic keys for identification of 16 tribes, 96 genera and 407 species, complemented by high resolution imagery, including of genitalic structures that are highly diagnostic. This book is the first comprehensive treatment of the known species of Aleocharinae beetles distributed in eastern Canada (Ontario and eastwards). For each species we include a morphological diagnosis, distribution, and natural history data. This work includes all commonly encountered aleocharines in eastern Canada and many uncom-

mon and rarely collected species. We hope that these identification tools and information synthesis will inspire many new taxonomic and ecological studies of Aleocharinae throughout Canada. Additional work will help assess whether there are possible species at risk and will uncover the fascinating biology of this diverse and, often maddeningly, complicated group of beetles.

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Historical Review of Research on Eastern Canadian Taxa

2

No serious attempt was made to inventory the Aleocharinae of Canada until the publication of the first checklist of Canadian Staphylinidae (Campbell and Davies 1991). The aleocharines recorded from Canada at that time clearly showed just how fragmentary and rudimentary the state of knowledge was, mainly because even genus identification was “difficult or impossible without access to type specimens” (Campbell and Davies 1991). It is, therefore, no accident that the earliest genera or subtribes to become the subject of modern taxonomic revisions were readily recognizable by habitus (e.g., *Aleochara*, *Gymnusa*, *Gyrophaena*), and those genera were among the best documented in Campbell and Davies (1991). The first person to provide a significant advancement in our knowledge of the Canadian fauna was Charles Seevers through his revision of North American and European *Gyrophaena* and allied genera (Seevers 1951). This early work is remarkable for illustrating the complicated primary and secondary sexual characters diagnostic of species in this group, and remains useful today. In the 1970s, revisions of the charismatic myrmecophile genus *Xenodusa* (Hoebcke 1976) and the distinctive wetland-dwelling genera *Gymnusa* and *Deinopsis* (Klimaszewski 1979) were completed. In 1978, a revision of the North American genera of Aleocharinae was published, providing a wealth of meticulous illustrations of important morphological characters and a

greatly needed, critical synthesis of generic concepts (Seevers 1978). This work was largely completed by the time of Seevers’ passing in 1965 but was not published until 13 years later with the assistance of Lee Herman and Rupert Wenzel (Seevers 1978). Seevers’ monograph served as an important catalyst for later improvements to generic classification (e.g., Ashe 1984, 1992, Gusarov 2003a), and remains the first reference typically consulted when encountering unfamiliar genera in Canada.

Progress during the 1980s included taxonomic monographs about the wetland-associated genus *Myllaena* (Klimaszewski 1982), the forensically and agriculturally important genus *Aleochara* (Klimaszewski 1984), the genus *Hoplandria* (Génier 1989), and a world revision and subgeneric reclassification of the diverse litter-dwelling genus *Leptusa* (Pace 1989). The earliest significant taxonomic progress on Canadian Athetini, a very diverse and difficult tribe, was accomplished by Lohse et al. (1990), who revised and illustrated genitalia of species from the Canadian Arctic. Many species known only from Alaska in Lohse et al. (1990) were since found to be broadly distributed across northern Canada. Campbell and Davies (1991) reported 113 species of Aleocharinae from eastern Canada, mostly from Ontario (67) and Quebec (74) (Fig. 2.1). Remarkably few species in that checklist were

known from the Canadian Maritime Provinces, with only three species recorded from Prince Edward Island (Fig. 2.1).

There was practically no research on aleocharines in eastern Canada throughout the 1990s. During the early 2000s, taxonomic work on the genera *Placusa* (Klimaszewski et al. 2001), *Earota* (Gusarov 2002a), *Geostiba* (Gusarov 2002b), *Pelioptera* (as *Tropimenelytron*) (Gusarov 2002c), *Seeveriella* (Gusarov 2003b), *Silusa* (Klimaszewski et al. 2003), *Strigota* (Gusarov 2003a) and *Tinotus* (Klimaszewski et al. 2002) resulted in new diagnostic tools for the Canadian fauna. Gusarov (2003a) examined a large number of North American type specimens of Aleocharinae that resulted in some valuable general insights concerning taxonomic work on Nearctic aleocharines. First, it was evident that species described by early workers (e.g., Casey) were often grossly misplaced to genus, causing them to be often overlooked in modern taxonomic treatments. Second, some Aleocharinae, especially Athetini, were found to be much more widespread than previously thought by workers such as Casey, underscoring the importance of checking all North American types as many species were potentially synonyms or wrongly classified. Third, subtle variation in external morphology had led to the proliferation of synonyms (e.g., ten names under *Strigota ambigua*), so to create increased taxonomic stability it was advocated that species concepts should first be based on primary and secondary sexual characters. Gusarov (2003a) also provided tools to reliably identify for the first time several of the most common *Atheta* species in Canada. In 2004, *Neothetalia* was described and added to the eastern Canadian fauna (Klimaszewski and Pelletier 2004), and Canadian species of *Leptusa* were revised (Klimaszewski et al. 2004).

Starting in 2005, regionally focused faunistic projects contributed much to the knowledge of the eastern Canadian fauna, significantly supplementing taxonomic revisions of some genera. Samples from forest survey plots in New

Brunswick generated many new Canadian and provincial records of aleocharines (Klimaszewski et al. 2005a; Fig. 2.1). Klimaszewski et al. (2005b) reviewed the tribe Lomechsuini in the Nearctic region and developed identification tools for the ant-associated genera *Myrmoecia*, *Pella*, *Platyusa*, and *Zyras* in Canada. The genera *Oxypoda*, *Hylota* (Klimaszewski et al. 2006a), and *Tachyusa* (Pašnik 2006) were revised a year later and several new *Atheta* species were discovered in Canada (Klimaszewski et al. 2006b, Majka et al. 2006). A survey of yellow birch dominated forests in southeastern Quebec revealed additional records for the province (Klimaszewski et al. 2007b) and a review of adventive Aleocharinae in Canada (Klimaszewski et al. 2007a) documented the genera *Crataraea*, *Nehemitropia* and *Meotica* for the first time in the fauna, and provided illustrations of their diagnostic characters. In the same year, the poorly known genera *Amarochara* (Assing 2007) and *Euvira* (Klimaszewski and Majka 2007a) were discovered in eastern Canada. In 2008, a combination of reviews of adventive species (Majka and Klimaszewski 2008a, c), studies of coastal littoral fauna (Klimaszewski et al. 2008b, Majka et al. 2008), regional faunistic summaries (Majka et al. 2008), and taxonomic revisions (Assing 2008, Klimaszewski et al. 2008a) resulted in the discovery of two new tribes (Diglottini and Taxicerini) in eastern Canada, the recognition of the genera *Calodera*, *Cypha*, *Diglotta*, *Halobrecta*, *Hydrosmeeta*, *Myrmedonota*, *Myrmecopora*, and *Phloeopora* in the fauna, and many new species records. Nearly all of the aforementioned papers were contained in a special issue of the journal ZooKeys entitled 'Biodiversity, Biosystematics, and Ecology of Canadian Coleoptera' and this collective work resulted in a large spike in knowledge about the composition of the eastern Canadian fauna, especially for Ontario and Nova Scotia (Fig. 2.1). A year later, the publication of the ZooKeys special issue 'Biodiversity, Biosystematics, and Ecology of Canadian

Coleoptera, Part II’ and other papers resulted in the discovery of rarely collected genera *Alisalia* (Klimaszewski et al. 2009d), *Ocyusa* (Webster et al. 2009), *Schistoglossa* (Klimaszewski et al. 2009a, b), and the fungicolous genus *Phymatura* (Webster et al. 2009) in eastern Canada. A review of the Canadian Gyrophaenina by Klimaszewski et al. (2009c) provided a modern update to Seevers (1951). The increase in faunistic knowledge for eastern Canada during 2009 was driven nearly entirely by collections made in New Brunswick, resulting in a significant spike in knowledge for the provincial fauna (Fig. 2.1). The faunistic contribution by Majka and Klimaszewski (2008b, 2010) is noteworthy for providing the most recent significant increase in the known diversity of Aleocharinae in Nova Scotia and Prince Edward Island (Fig. 2.1).

The advances in the taxonomy and faunistics of eastern Canadian Aleocharinae in the last 7 years emerged from three major, region-centered projects. A review of the aleocharine fauna of Newfoundland and Labrador (Klimaszewski et al. 2011) represents the greatest increase to a provincial fauna (130 species) in a single contribution thus far (Fig. 2.1). Many of these were important new eastern records of transboreal species that had been poorly sampled. In the course of this project, a new aleocharine tribe, Boreocyphini (*Boreocypha*), was discovered, and the genera *Brachyusa*, *Gnathusa*, and *Tectusa* (as *Parocyusa*) were newly identified in eastern Canada. Additional new records and species from Newfoundland and Labrador were published five years later (Klimaszewski et al. 2016b) making the fauna of this province one of the best known of any jurisdiction in North America. In 2012, another special issue of ZooKeys with Ontario-focused (Brunke et al. 2012) and New Brunswick-focused (Webster et al. 2012) contributions on aleocharines, together adding 43 species to the fauna of eastern Canada and bringing the total faunal inventory of each province to over 200 species (Fig. 2.1). These contributions also recorded the genera *Agaricomorpha*, *Alevonota*, *Callicerus*,

Dexiogyia, *Phanerota*, *Stethusa*, *Thecturota*, and *Tomoglossa* from Canada for the first time. From 2012 to 2016, the focus of aleocharine research shifted once again toward comprehensive taxonomic reviews, focusing on the subcortical genus *Dinaraea* (Klimaszewski et al. 2013), the northern genera *Gnathusa*, *Mniusa*, and *Ocyusa* (Klimaszewski et al. 2014), and the taxonomically difficult and minute beetles in the genera *Mocyta* (Klimaszewski et al. 2015a), *Clusiota*, and *Atheta* (*Microdota*) (Klimaszewski et al. 2015b). The latest major additions to the knowledge of the aleocharine diversity of eastern Canada was in 2016 when 43 species were recorded as new to the fauna (Fig. 2.1). These most recent publications included a taxonomic review of *Liogluta*, associated with wet litter (Klimaszewski et al. 2016a), and regional studies on the fauna of New Brunswick (Webster et al. 2016) and Newfoundland (Klimaszewski et al. 2016b). Webster et al. (2016) reported the subcortical genus *Anomognathus*, the fungicolous genus *Pleurotobia*, and cool, damp litter-dwelling genus *Blepharhymenus* from eastern Canada for the first time. Most recently, Klimaszewski et al. (2017a, b) reported the riparian genera *Apimela* and *Gyronycha* from Canada for the first time and summarized the state of taxonomic knowledge about Nearctic species.

In the 26 years since the first Canadian checklist (Campbell and Davies 1991), documentation of the fauna of eastern Canada has proceeded at an accelerating rate (Fig. 2.1). The fauna of New Brunswick has gone from nearly unknown (19 species) in 1991 to 299 species in 2017—a 15-fold increase. Other provincial faunas that have seen the most growth in terms of known species composition are Nova Scotia, Newfoundland and Labrador, and Ontario. However, many more species remain to be documented in each province and in eastern Canada as a whole (see Chap. 4). The present work provides the knowledge synthesis and identification tools to hopefully guide and inspire others to continue the documentation of the regional fauna.

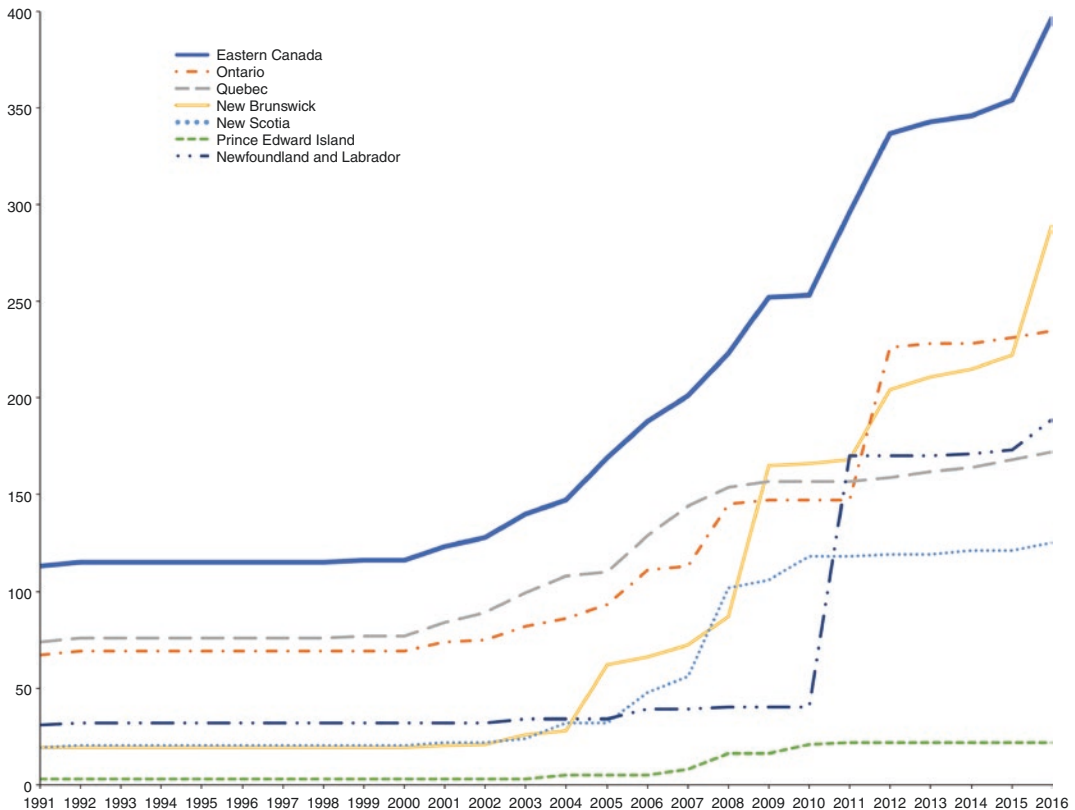


Fig. 2.1 26 years of taxonomic progress on the aleocharine (Coleoptera: Staphylinidae) fauna of eastern Canada: cumulative number of species recorded from six Canadian provinces over time

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Format

The classification of taxa used herein selectively follows concepts expressed by others (Benick and Lohse 1974, Seevers 1978, Klimaszewski 1979, 1984, 1982, Lohse et al. 1990, Ashe 2001, Gusarov 2003, 2011, Elven et al. 2010, 2012, Pašník 2010, Bouchard et al. 2011, Hlaváč et al. 2011, Schülke and Smetana 2015), and several treatments of Canadian genera (Klimaszewski et al. 2001, 2002, 2003, 2004, 2005, 2006, 2008b, 2009a, b, c, 2013, 2015a, b, 2016, Yamamoto and Maruyama 2017). Distributional records and bionomic information (habitat associations, collection dates, collecting methods) are based mostly on published records and on specimens in the collections of Reginald Webster (Charters Settlement, NB), New Brunswick Museum (Saint John, NB), Atlantic Forestry Centre (Fredericton, NB), Laurentian Forestry Centre (Ste-Foy, QC), and the Canadian National Collection of Insects, Arachnids and Nematodes (Ottawa, ON). In the case of unpublished records, we provide specimen data under the respective species. Only records considered to be reliable are listed.

Abbreviations

Jurisdictions in boldface are included in eastern Canada, as defined in this work. We report separately on the jurisdictions of Newfoundland and

Labrador even though together they constitute one province.

AB – Alberta

AK – Alaska

BC – British Columbia

LB – Labrador

MB – Manitoba

NB – New Brunswick

NF – Newfoundland

NS – Nova Scotia

NT – Northwest Territories

NU – Nunavut

ON – Ontario

PE – Prince Edward Island

QC – Quebec

SK – Saskatchewan

YT – Yukon Territory

State abbreviations for the United States of America follow those of the United States Postal Service.

Institution Codens

CNC Canadian National Collection of Insects, Arachnids, and Nematodes, Agriculture and Agri-Food Canada, Ottawa, Ontario, Canada.

FMNH The Field Museum of Natural History, Chicago, Illinois, United States of America.

LFC	Natural Resources Canada, Canadian Forest Service, Laurentian Forestry Centre, R. Martineau Insectarium, Quebec City, Quebec, Canada.
NoFC	Natural Resources Canada, Canadian Forest Service, Northern Forestry Centre, Edmonton, Alberta, Canada.
RWC	Reginald Webster collection, Charters Settlement, New Brunswick, Canada.
TSC	Tim Struyve collection, Mechelen, Belgium.
UAM	University of Alaska Museum, Fairbanks, Alaska, United States of America.
UCC	University of Copenhagen, Copenhagen, Denmark.
USNM	United States National Museum, Washington, DC, United States of America.
UWP	University of Wroclaw, Poland.
ZMH	Zoological Museum, Helsinki, Finland.
VAC	Volker Assing collection, Hannover, Germany.

Diagnostic Features

Aleocharine beetles are highly, diverse taxonomically, morphologically (Fig. 3.1a–l) and ecologically. This subfamily is one of the least known beetle groups in North America and in other parts of the world. Significant challenges with species identification due to the poor state of knowledge of many groups and the dearth of comprehensive diagnostic tools is the main obstacle for understanding species richness, assemblage composition, and ecological roles in terrestrial ecosystems. There are many species, often forming groups of cryptic species, which are externally similar, especially in the large tribes Aleocharini (e.g., *Aleochara*), Athetini (e.g., *Atheta*), Oxypodini (e.g., *Oxypoda*), and Homalotini (e.g., *Gyrophana*). In Canada, these are often represented by pairs of sibling species that can only be distinguished with certainty by the examination of genital structures (e.g., *Aleochara castaneipennis* Mannerheim and *A. castaneimarmotae* Klimaszewski, Webster, and Brunke; *Atheta crenuliventris* Bernhauer and *A. pseudocrenuliv-*

entris Klimaszewski; *Atheta districta* Casey and *A. pseudodistricta* Klimaszewski and Langor; *Atheta modesta* (Melsheimer) and *A. pseudo-modesta* Klimaszewski).

External Body Structures Important for Identification

The terminology used herein follows that used by previous authors (Benick and Lohse 1974, Seevers 1978, Klimaszewski 1979, 1984, Lohse et al. 1990, Ashe 2001, Gusarov 2003).

The body length of aleocharine beetles ranges from 1.0 to 13.0 mm (typically 3.0–5.0 mm) and habitus forms are diverse (Fig. 3.1a–l), reflecting different adaptations to a variety of microhabitats. They range from somewhat flat (e.g., *Anomognathus*, *Homalota*, *Dinaraea*, *Placusa*, and *Xenodusa*) to subcylindrical (e.g., some *Leptusa*), and from robust (e.g., *Aleochara*, *Gymnusa*, and *Oligota*) to slender (e.g., *Atheta*, *Clusiota*, *Myllaena*, *Meotica*, and *Thecturota*).

The term forebody refers to head, pronotum and elytra as a combined structure. The most important body structures used in identification of aleocharines are illustrated (Figs. 3.2–3.4). These relate to the **head** (Fig. 3.2): frontal suture, genae (postocular area, temples), postocular carina, neck; **mouthparts** (Fig. 3.3): labrum, labium, maxillae, labial palps, and ligula; **prothorax** (Figs. 3.2 and 3.4): pubescence pattern, hypomeron, pro-, meso- and metaventrites and their intercoxal processes, isthmus, mesothoracic peritremes (small structures that encompass the spiracles behind the procoxae), and coxae; **elytra** (Figs. 3.2 and 3.4): size and shape, pubescence pattern, elytral suture, basal margin, and lateral emargination; **abdomen** (Fig. 3.5a–e) [10 segmented, position indicated by Roman numerals starting at the base; when describing features of the “first visible tergites” in the keys and diagnoses, we are referring to the basal segments following morphological segments I and II which are reduced and largely hidden by the elytra; segments IX and X are modified]; transverse impressions of basal tergites, structures inside

impressions, and shape of the apical margin of male tergite VIII; **legs** (Fig. 3.2): tarsal formula and length and dimensional proportions of the articles of the hind tarsus.

Genital Structures Important for Identification

The morphological diversification of the genitalia, particularly the shape of the median lobe of the aedeagus and its internal sac structures, including flagellum and internal sclerites (structures), morphology and macro-setal pattern on the apical lobe of parameres, and morphology of the spermatheca, must have played an important role in aleocharine evolution because these structures are variably shaped in different species and are generally the structures most important for species- and genus-level identification. The three-lobed aedeagus of aleocharines, along with complex parameres, is the most distinctive feature of the entire subfamily, and most likely constitutes a synapomorphy for this group.

Aedeagus: The male copulatory organ consists of the median lobe (penis) (Fig. 3.6a, b), and the two parameres (Fig. 3.6c, d).

Median lobe of aedeagus (Figs. 3.6a, b and 3.7a–l): This is a tubular, sclerotized, intromittent organ with an enlarged basal portion called the *bulbus* (Fig. 3.6a, b), and a narrow apical portion, subcylindrical or trough-like, called the *tubus* (Fig. 3.6a, b). The tubus is slender, and narrow towards the apex. The lateral edges of the tubus are extended dorsally in Athetini and form a complete or incomplete arched structure called the *athetine bridge* (Seevers 1978). The shape of the tubus, and particularly its apical portion in lateral view, varies from species to species and is a valuable diagnostic feature. The subspherical bulbus has a small, ventral round opening, the *foramen mediale*, which is an entrance for the ejaculatory duct, the *ductus ejaculatorius*, and one or two anterior projections called the *crista apicalis* or external carina of the bulbus. The crista apicalis provides attachment sites for the muscles connecting the median lobe and the two condylites of the parameres (Fig. 3.6b). The internal space of

the bulbus contains a complex of mainly dorso-ventral muscles attached internally to the base of the bulbus and to the dorsal mobile compressor plate that is oval and weakly sclerotized. Contraction or expansion of the dorso-ventral muscle complex is directly or indirectly responsible for creation of hydrostatic pressure that results in eversion or retraction of the *internal sac* and its associated internal structures (e.g., sclerites, membranous structures, and flagellum). The *flagellum* (also known as the virga or copulatory piece) (Fig. 3.6a, b) is a short or moderately-to-extremely elongate tubular structure (present in some *Aleochara*, *Aleodorus*, and *Oxypoda*) that can be narrow or broad, and it has a swollen basal part connected with the ejaculatory duct. The internal sac membranes frequently bear numerous spinules, denticulate plates, and other structures to aid in retention of the sac in the *vulva* (female copulatory tract). During eversion of the internal sac, the internal sclerites and flagellum are extended (everted) exteriorly filling the internal space of the female vulva and allowing safe transfer of sperm, or packages of sperm contained in spermatophores, with the aid of the flagellum. The internal sac is withdrawn by a set of longitudinal muscles that originate on the proximal surface of the base of the bulbus. The structures and function of the internal sac were previously explained and illustrated for *Aleochara curtula* by Peschke (1978).

The ventral side of the median lobe of the aedeagus is considered to be the side of the bulbus containing the foramen mediale, the entrance of the ductus ejaculatorius, and the adjacent ventral side of the tubus of the median lobe with the internal sac and its structures (this part is referred to as the parameral side in some recent publications (e.g., Gusarov 2003); the opposite side is referred to as the dorsal part.

Parameres (Fig. 3.6c, d): These are the tactile orientation organs, which enable the male to find the tip of the female abdomen just before copulation, using the setal pattern on the female abdomen as a guide (Peschke 1978, Klimaszewski 1984). There are two symmetrical parameres, one on each side of the median lobe of the aedeagus, which are attached to the

crista apicalis (represented by one, two or several carinae) of the bulbus of the median lobe by a complex system of muscles. Each paramere is subdivided into a *paramerite* and a narrowly elongate *condylite*. The paramerite is the main part of the paramere, and consists of a narrow apical lobe bearing a few macrosetae arranged in a particular pattern specific to different genera. Attached to the paramerite is a semimembranous *velar sac* bearing microribbings, and the posterior, slit-shaped structure delimited from the apical portion by a weakly sclerotized strip (Klimaszewski 1984). The morphology of the paramere and the setal pattern of the apical lobe of the paramerite provide diagnostic characters at the generic and higher taxonomic levels.

Spermatheca (*receptaculum seminis*) (Figs. 3.6e and 3.8a–l): This is the female genital organ for storing and perhaps selecting sperm (Peschke 1978, Dybas and Dybas 1981). The spermatheca consists of a bulbous capsule connected to a tubular stem and a thin and membranous, weakly sclerotized seminal canal (*ductus spermathecae*), which connects the spermatheca to the vulva and serves to transport sperm. The seminal canal is not considered to be a part of the spermatheca (Klimaszewski 1984). The capsule is a spherical or tubular structure that may be straight or bent, and bears apical invaginations of various shapes and sizes. The stem may be a simple narrow tube or more complex and subdivided into an elongate chamber and a narrower duct. The shape of the spermatheca varies from species to species and is valuable for species separation. In some genera (e.g., *Acrotona*, *Amischa*, some *Atheta*, *Gnypeta*, *Liogluta*, and *Mocytia*) the shape of the spermatheca may be a better diagnostic character than the shape of the median lobe of the aedeagus.

Terminalia (Fig. 3.5a–e): The terminalia herein refer to the male and female tergite VIII and sternite VIII (Fig. 3.5e) and the *pygidium*, the latter consisting of two to three modified terminal segments closing the end of the abdomen (Fig. 3.5a–d). Tergite VIII and sternite VIII provide useful additional morphological features for species level diagnostics. These structures can be elongate, subquadrate or transverse and can have

varying pubescence patterns. The *antecostal suture* is located subparallel to the base of tergite VIII and sternite VIII. The antecostal suture may be separated by a wide or narrow distance from the base of the disc and may be straight or sinuate in different species. The apical margin of tergite VIII of males is highly modified in many groups of aleocharines, and particularly in athetines, and often bears teeth, denticles, and other projections or emarginations, which provide important taxonomic identification features. The structure of the pygidium is slightly different between males and females (Fig. 3.5a–d). In the male, the pygidium consists of tergite X, accompanied on both sides by tergite IX (completely subdivided or connected by a narrow dorsal strip) which is extended ventrally into two posterior narrow ventral struts (Fig. 3.5a, b). Ventrally located between the subdivided tergite IX of the male is a narrowly oval and flat structure which is regarded as the modified sternite IX. The pygidium of the female is generally similar to that of the male but is lacking ventral struts and an elongate sternite IX (Fig. 3.5c, d). The shape and pubescence pattern of tergite X is often diagnostic for species and genera. Specimens can be sexed by examination of the ventral part of the pygidium for presence (male) or absence (female) of the apical portion of sternite IX which is present in males as a median rounded lobe (Fig. 3.5a, b).

Microdissections and Preparation of Genital Structures for Examination and Diagnostics

Identifying aleocharine species and genera can be challenging due to the small size of most taxa and minute size of many distinctive characters, coupled with a vast number of valid taxa (Ashe 2001, Hanley and Ashe 2003). There are many species, often forming groups of sibling species, which are externally similar, especially in the large tribes Athetini (e.g., *Atheta*), Oxypodini (e.g., *Oxypoda*), and Homalotini (e.g., *Gyrophana*). Thus, properly dissected genital structures (median lobe of aedeagus, parameres,

spermatheca, and tergite VIII and sternite VIII of both sexes) carefully mounted in Canada balsam on plastic microslides for examination, are indispensable for identification of the majority of species and is therefore essential for serious work in aleocharine systematics. The methodology of dissecting aleocharine species mouthparts is sufficiently described by Hanley and Ashe (2003) and is not discussed here as mouthparts are usually not essential for species identification.

Relaxation of Specimens

Specimens stored in 70% ethanol do not need to be relaxed prior to dissection. However, dried mounted specimens need to be relaxed in 5% ammonium hydroxide solution prior to dissections. The duration of relaxation depends on the age of the dried specimens: those from old collections need to be relaxed for several hours but more recently mounted specimens need to be relaxed for only 5–15 min. When the abdomen flexes easily it is usually ready for dissection. Extremely dry and old specimens may be quickly relaxed by immersing them in hot tap water or heating in a boiling water bath for 1–2 min. Some heavily sclerotized structures may be cleared in cold 10% potassium hydroxide for 1–3 min, washed in water, and then dehydrated in absolute ethanol prior to mounting on slides.

Dissection and Dehydration

With the aid of a stereoscopic microscope, specimens are best dissected in water contained in an impression of a glass microscope slide (commercially available slides with impressions used for blood analysis). The abdomen is held in place by forceps while a hooked or straight minuten pin, mounted in a commercially available handle, is inserted between abdominal segments VII and VIII and pulled to dislodge the terminal segments from the rest of the abdomen. The genital structures (aedeagus or spermatheca) are usually inside the dislodged apical part of the abdomen but

sometimes may remain in the basal part of the abdomen and should be removed from there. The dislodged apical part of the abdomen is dissected further by removing tergite VIII and sternite VIII from the pygidium using dissecting needles and then breaking the lateral internal muscles that fix the discs in place and pulling the sclerites apart. Subsequently, the aedeagus or spermatheca are also removed and detached from other structures so that tergite VIII, sternite VIII, aedeagus (or spermatheca), and pygidium are separated from each other. Remnants of muscles attached to structures may be removed mechanically using dissecting needles while structures are immersed in water or alternatively structures may be cleared in cold 10% potassium hydroxide for 1–4 min to digest the soft tissues and then washed in water. The aedeagus needs further dissection by removing the parameres from the median lobe using dissecting needles to break the muscles connecting the condylite of the paramere with the base of the bulbus of the median lobe. All dissected structures need to be washed in clear water, transferred to 75% ethanol for about 1 min and then to 100% ethanol for dehydration for about 1–2 min before mounting. Structures should then be mounted in Canada balsam on a celluloid microslide attached to the pin holding the card-mounted body of the dissected specimen. Celluloid microslides are commercially available in different sizes; the size appropriate for Aleocharinae is 6.6×17.0 mm. One source of celluloid slides is: Gesellschaft zur Kartierung der Wirbellosen, c/o Clemens Brandstetter, Schesasastraße 1, A—6706, Bürs, Austria. For beginners it is best to practice dissection on some unimportant specimens, starting with larger specimens.

The genital structures should be mounted on microslides in a consistent pattern of arrangement to facilitate easy comparison of homologous structures on different microslides. We recommend the following arrangement of structures on the slide: median lobe of aedeagus in lateral view or spermatheca in right upper corner; separated parameres situated below the median lobe on the right side; tergite VIII in left upper corner, sternite VIII below tergite VIII, and pygidium below sternite VIII. For thinning Canada balsam, we use xylene or

non-toxic terpeneol, both commercially available. It is important to note that, at first, only a thin layer of Canada balsam is placed on a microslide to prevent floating of structures into undesirable positions. After the first layer of Canada balsam with the genital structures placed in their optimal position on a microslide is dried, a second layer of Canada balsam can be added to completely, but without excess, cover the structures. We regularly use the above described procedure and we find it facilitates the microscopic examination of genital structures and their photography. The advantage of this system is that the dissected specimen and the genital structures are on the same pin in the collection instead of being stored in two separate collections which can be easily misplaced or lost. This approach is also superior to using genitalia vials which take up more room on the pins, may damage structures when taken out of the vial, and requires more handling time.

Image Production

Images of the entire body and the genital structures were taken using an image processing system (Nikon SMZ 1500 stereoscopic microscope; Nikon Digital Camera DXM 1200F, and Adobe Photoshop software). Images of the dorsal aspects of the head, pronotum, elytra, abdomen, legs and antennae were taken separately in the horizontal plane (except for the head image which was taken while in a natural position) and fused together in Adobe Photoshop. Images of tergite VIII or sternite VIII that were damaged during dissection were 'repaired' by digitally duplicating the undamaged portion using Adobe Photoshop software.

DNA Barcoding

In 2003, "DNA barcoding" was proposed as a way to identify species (Hebert et al. 2003). This method uses a 658 base-pair fragment from the mitochondrial cytochrome c oxidase 1 (CO1) gene as its standard barcode. CO1, as a standard barcoding region is proving to be highly useful for identifying species in many animal groups,

including insects. Previously, biological specimens were identified using mainly morphological features, but if a specimen was damaged, obscuring diagnostic characters, or was of an immature stage of development, even specialists were frequently unable provide identifications. Barcoding helps solve these problems for some specimens, although the specimen age and method of collection/preservation can limit successful amplification of the barcoding region. Thus, DNA barcoding can serve as a valuable tool in the taxonomist's toolbox by providing a set of characters in addition to morphological characters. However, not all species have unique DNA barcodes, so both molecular and morphological characters need to be considered together. One of the most important products assisting molecular characterization of species is a public reference library of molecular data. There are currently two main databases that fill this role: (1) the International Nucleotide Sequence Database Collaboration (available through the GenBank data portal on the National Center for Biotechnology Information website (NCBI: <https://www.ncbi.nlm.nih.gov/>)); and (2) the Barcode of Life Database (BOLD: <http://boldsystems.org/>) that was created and is maintained by the Centre for Biodiversity Genomics in Ontario (Ratnasingham and Hebert 2007). The BOLD offers researchers a way to collect, manage, and analyze DNA barcode data. Specimens are identified by finding the closest matching reference record in the database, and barcoded specimens are grouped together into tentative species using a proxy called the "Barcode Index Number" (BIN). The BIN makes working with unknown species a lot easier and more objective than OTUs (Operational Taxonomic Units) only based on morphology as a BIN is assigned based on a complex algorithm taking into account intraspecific variation by identifying clear discontinuities in variation within clusters (Ratnasingham and Hebert 2013). Barcode sequences in BOLD are associated with vouchered specimens distributed across many museums, and the intention is that voucher specimens will be authoritatively identified by taxonomists to provide a reliable association between the barcode and a particular species

name. In concert with the development of this book we undertook an initiative to obtain barcodes for as many species of eastern Aleocharinae as possible, focusing mainly on authoritatively identified specimens in the LFC. These data are available to the public through a dataset on BOLD (DOI: [x.doi.org/10.5883/DS-ALEOCHAR](https://doi.org/10.5883/DS-ALEOCHAR)).

Barcodes were obtained from 47 genera and 104 species (117 specimens) covered in this book (Fig. 3.9, Table 3.1), about a quarter of the fauna known from eastern Canada. Only records with sequences longer than 300 base-pairs (bp) (full barcodes have 658 bp) were

included to provide accurate reference data for future identifications, compatible with the BIN system. As these barcodes represent only a subset of the eastern Canadian aleocharine species covered herein, no analysis was done to test the level of correspondence of the sequences to the definition of species. A neighbor joining tree (NJT) was created (Fig. 3.9) as a quick reference to the taxa included in the dataset. For further information on these records, please refer to the DOI provided above. As a NJT tree represents similarities of sequences to each other, it is by no means a

Table 3.1 List of barcoded species names covered in this chapter

<i>Acrotona subpygmaea</i>	<i>Dinaraea pacei</i>	<i>Myllaena arcana</i>
<i>Agaricomorpha websteri</i>	<i>Drusilla canaliculata</i>	<i>Myllaena kaskaskia</i>
<i>Aleochara sculptiventris</i>	<i>Euaira micmac</i>	<i>Myllaena procidia</i>
<i>Aleochara verna</i>	<i>Gennadota canadensis</i>	<i>Myllaena vulpina</i>
<i>Alisalia minuta</i>	<i>Geostiba circellaris</i>	<i>Myrmecocephalus gatineauensis</i>
<i>Alisalia testacea</i>	<i>Gnypeta brincki</i>	<i>Myrmedonota aidani</i>
<i>Amarochara duryi</i>	<i>Gnypeta carbonaria</i>	<i>Ocyusa asperula</i>
<i>Amischa analis</i>	<i>Gnypeta nigrella</i>	<i>Oxypoda convergens</i>
<i>Atheta alesi</i>	<i>Gyrophaena affinis</i>	<i>Oxypoda demissa</i>
<i>Atheta atramentaria</i>	<i>Gyrophaena antennalis</i>	<i>Oxypoda frigida</i>
<i>Atheta borealis</i>	<i>Gyrophaena criddlei</i>	<i>Oxypoda opaca</i>
<i>Atheta cryptica</i>	<i>Gyrophaena flavicornis</i>	<i>Oxypoda perexilis</i>
<i>Atheta curtipennis</i>	<i>Gyrophaena fuscicollis</i>	<i>Oxypoda pseudolacustris</i>
<i>Atheta dadopora</i>	<i>Gyrophaena gilvicollis</i>	<i>Oxypoda sylvia</i>
<i>Atheta formicaensis</i>	<i>Gyrophaena gracilis</i>	<i>Parocalea pseudobaicalica</i>
<i>Atheta frosti</i>	<i>Gyrophaena insolens</i>	<i>Pella glooscapii</i>
<i>Atheta klagesi</i>	<i>Gyrophaena involuta</i>	<i>Pella loricata</i>
<i>Atheta modesta</i>	<i>Gyrophaena keeni</i>	<i>Philhygra laevicollis</i>
<i>Atheta newfoundlandica</i>	<i>Gyrophaena laetula</i>	<i>Phloeopora canadensis</i>
<i>Atheta novaescotiae</i>	<i>Gyrophaena lobata</i>	<i>Placusa tacomae</i>
<i>Atheta particula</i>	<i>Gyrophaena meduxnekeagensis</i>	<i>Placusa vaga</i>
<i>Atheta savardae</i>	<i>Gyrophaena modesta</i>	<i>Schistoglossa brunswickensis</i>
<i>Atheta strigosula</i>	<i>Gyrophaena pseudocriddlei</i>	<i>Schistoglossa pseudocampbelli</i>
<i>Atheta terranova</i>	<i>Gyrophaena socia</i>	<i>Schistoglossa sphagnorum</i>
<i>Atheta vestita</i>	<i>Gyrophaena subnitens</i>	<i>Seeveriella globicollis</i>
<i>Boreocypha websteri</i>	<i>Gyrophaena vitrina</i>	<i>Silusa alternans</i>
<i>Boreophilina islandica</i>	<i>Hylota ochracea</i>	<i>Silusa densa</i>
<i>Boreostiba parvipennis</i>	<i>Leptusa elegans</i>	<i>Strigota obscurata</i>
<i>Brachyusa helenae</i>	<i>Liogluta intermedia</i>	<i>Tachyusa smetanai</i>
<i>Callicerus rigidicornis</i>	<i>Liogluta microgranulosa</i>	<i>Tetraleucopora americana</i>
<i>Calodera parviceps</i>	<i>Liogluta pseudocastoris</i>	<i>Thecturota capito</i>
<i>Cordalia obscura</i>	<i>Liogluta castoris</i>	<i>Xenodusa reflexa</i>
<i>Devia prospera</i>	<i>Lypoglossa franclemonti</i>	
<i>Diglotia mersa</i>	<i>Mniusa minutissima</i>	
<i>Dinaraea aequata</i>	<i>Mniusa yukonensis</i>	
<i>Dinaraea borealis</i>	<i>Mocyta sphagnorum</i>	