Genera of Microgastrinae (Hymenoptera: Braconidae) collected from Khyber Pakhtunkhwa, Pakistan

Fakhruddin and Mian Inayatullah

Abstract
Microgastrines were collected from nine districts of Khyber Pakhtunkhwa, Pakistan. More than 1200 specimens were collected and examined for proper identification. Results show that a total of nine genera occurred in Khyber Pakhtunkhwa viz; Apanteles Foerster, Cotesia Cameron, Dolichogenidea Viereck, Diolcogaster Ashmead, Glyptapanteles Ashmead, Snellenius Westwood, Microplitis Foerster, Microgaster Latreille, and Paroplitis Wasmann. The genus Paroplitis Wasmann had been recorded for the first time as a new record from Pakistan. The most speciose and diverse genus was Cotesia Cameron, having eight species and was recorded from different argro-ecological zones of Khyber Pakhtunkhwa. The other genera Microplitis Foerster and Apanteles Foerster having five species each and Dolichogenidea Viereck, Diolcogaster Ashmead, Glyptapanteles Ashmead, Microgaster Latreille, Snellenius Westwood represented by one species each respectively. Distributional notes and images of key morphological features are provided for each genus and a new key to the microgastrine genera of Khyber Pakhtunkhwa is presented.

Keywords: Taxonomy, wasps, parasitoids, dichotomous key.

1. Introduction
Microgastrinae belongs to Hymenoptera, the leading insect order among all entomophagous insects. More than two thirds of successful biological control cases involving pest species have been achieved by hymenopteran parasitoids (Irshad 2008) [7]. The subfamily Microgastrinae is universally recognized as a monophyletic taxon, easily identified by the characteristic wing venation and the number of flagellomeres invariably fixed at 16. Despite the latter character being a strong apomorphic feature, it was nevertheless recently reconsidered with the description of the unusual genus Kiwigaster (Fernández-Triana et al. 2011), which has males and females with different numbers of antennal flagellomeres (females 17, males 18) [8]. Beyond this consensus, the phylogenetic subdivision of Microgastrinae is still debated, though several attempts were proposed to stabilize it (cf. Whitfield et al. 2002 for a review) [14, 5]. Microgastrines are the most commonly encountered braconid parasitoids of Lepidoptera. Biologically, they form a homogenous group: they are all lepidopteran koinobiont endoparasitoids, parasitizing nearly all families of Lepidoptera. Except for a few species that pupate within the cocoon of the prepupal host, they exit the dead host to spin their cocoon outside. While most species of microgastrine are solitary, some are gregarious with species that parasitize macrolepidoptera (such as Sphingidae and Saturniidae) resulting in hundreds of larvae emerging from a single host. So far, more than one hundred microgastrine species have been used in biocontrol programs against agricultural pests (Whitfield 1997) [1]. As a large part of the hymenoptera world fauna is still poorly known, the actual richness of Microgastrinae is currently expected to range between 5000 and 10000 species (Mason 1981) [9]. Recent collecting shows that this number requires updating. There is no recent or comprehensive checklist to the genera of Microgastrinae in Pakistan, however some scattered work has been done in the Punjab and Khyber Pakhtunkhwa Province of Pakistan. In Khyber Pakhtunkhwa Province this will be an important step to recording the genera and species of Microgastrinae. Such a revision is not only of taxonomic interest but also useful for ecological and biological studies.
2. Materials and methods
The examined specimens have collection localities from all over Khyber Pakhtunkhwa covering 9 districts (Peshawar, Swabi, Mardan, Haripur, Swat, Dir, Chitral, DIK and Nowshehra) distributed from the northern to the southern regions of Khyber Pakhtunkhwa, Pakistan. Identification and illustration were done with the help of a stereozoom Microscope with magnification up to 300x. Alcohol preserved specimens were processed using hexamethyldisilazane and later point mounted. All specimens examined during this study are deposited in the Entomology Museum at The University of Agriculture Peshawar.

3. Results and Discussion
The present research regarding the microgastrine fauna of the Pakhtunkhwa province of Pakistan has yielded 23 species in 9 genera.

Key to the Genera of Microgastrinae in Khyber Pakhtunkhwa Pakistan

<table>
<thead>
<tr>
<th>1. Fore wing areolet absent (Fig. 4)</th>
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<tr>
<td>- Fore wing areolet present (Fig. 22)</td>
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<tr>
<td>2. Hypopygium membranous; ovipositor sheath completely hairy (Fig. 2, 10)</td>
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<tr>
<td>- Hypopygium sclerotized; ovipositor sheath with hairs concentrated apically (Fig.)</td>
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<td>3. Vannal lobe of hind wing concave (Fig.1a)</td>
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<tr>
<td>- Vannal lobe of hind wing convex (Fig.1c)</td>
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<td>4. T1 broadening posteriorly (except glomeratus sp.) (Fig. 6)</td>
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<td>- T1 narrowing posteriorly or parallel (Fig. 16)</td>
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<td>5. Hind coxa shorter than T1: fore wing areolet triangular or quadrangular (Fig. 22)</td>
<td>10</td>
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<tr>
<td>- Hind coxa longer than T1: forewing areolet small and slit-like (Fig. 12)</td>
<td>11</td>
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<tr>
<td>6. Ovipositor sheaths about half as long as hind tibia and setose over most of length, flagellomeres with only a single row of placod</td>
<td>12</td>
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<tr>
<td>- Ovipositor sheaths short as hind tibia and with setae concentrated distally flagellomeres with two ranks of placodes in both sexes</td>
<td>13</td>
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<td>7. T2 usually with raised median area; when this is well-defined, the median area is much longer than wide (Fig. 13)</td>
<td>14</td>
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<td>- T2 usually sculptured without any raised area (Fig. 18)</td>
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<tr>
<td>8. Mesopleuron with distinct (some time complete) epicnemial carina; propodeum double-faced (Fig. 25, 26)</td>
<td>16</td>
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<tr>
<td>- Mesopleuron without distinct epicnemial carina; propodeum single-faced (Fig.20)</td>
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![Fig 1: Vennal lobe of hind wing indicated by an arrow; (a–b) hypothetic Apanteles i.e. Concave or slightly straight, sparsely fringed; (c) Dolichogenidea i.e. Convex and evenly fringed (redrawn from Whitfield 1997).](image)

**Genera Diagnoses and Comments**

**Apanteles Foerster**

Apanteles can be recognized among the world microgastrinae by the following combination of features; fore wing with second r-m vein absent, so that the small areolet is open distally; hind wing with vennal lobe distally flattened and with reduced fringe of hairs; punctuation of posterior part of mesonotum breaking down into more confluent longitudinal sculpturing, especially submedially; propodeum with oval pentagonal, hexagonal or anteriorly open medial areola; first metasomal tergite usually with medial sub apical depression and second metasomal tergite strongly transverse, often with convex or simuate posterior margin; ovipositor and sheath long, exerted, and functional through a desclerotized hypopygium. Apanteles are mostly solitary parasitoids of microlepidoptera, with a few known exceptions. Ankita (2013) [6]. The genus is easily confused with the related, but less diverse, genus Dolichogenidea, which can be separated on the basis of distinct punctures posteriorly on mesonotum, and a convex and evenly fringed hind wing vennal lobe. Some authors (Yu et al. 2012) consider Dolichogenidea to be a junior synonym of Apanteles. The species of the genus Apanteles Foerster are the most frequently encountered amongst all the microgastrines with an estimate of roughly 2,000 species reported across the world (Mason 1981 [8] and Whitfield et al. 2009) [14]. From Indo-Pak approximately 155 species of the genus Apanteles are reported which accounts for only 7% of the estimated number of this highly specious genus.

Mason (1981) [8] correctly pointed out that the paucity of host records reflects the taxonomically uncharacterized composition of the genus. In this survey most of the collecting was done through ground netting and Malaise traps with the exception of some reared species, i.e., Apanteles and Cotesia. Therefore, most specimens lack host information. Wasps of this genus were collected from different districts of Khyber Pakhtunkhwa, Pakistan, both hilly and plain areas. These wasps are in flight April to November.

**Terminology**

Cotesia Cameron

Among microgastrine genera, Cotesia is one of the largest genus, composed of more than one hundred species. Members of this genus primarily attack larvae of larger moth and butterfly families, i.e., Noctuidae, Geometridae and Saturniidae. Cotesia is recognizable among microgastrines by following the features: fore wing with second r-m vein absent, so that the small areolet is open distally (Fig. 8); propodeum coarsely sculptured with medial carina instead of areola (Fig.); 1st and 2nd metasomal tergites usually quadrate in form and highly sculptured (Fig. 7), short ovipositor and sheaths which are barely exerted (Fig. 6). The genus is easily confused with Parapanteles, but the latter contains propodeal areola.

In Khyber Pakhtunkhwa Cotesia is the most diverse genus. Every collection effort yielded specimens of the genus Cotesia. It was collected from all the selected localities throughout the collection period.

Fig 2: Apanteles Foerster from Khyber Pakhtunkhwa, showing declerotized hypopygium with expandable pleats (arrow), ovipositor and sheath longer and exerted. Fig 3: Propodeum; Showing anteriorly open areola, T1; with medially subapical depression, T2; strongly transverse with convex posterior margin. Fig 4: Fore wing with open areolet.

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Dolichogenidea Viereck

Dolichogenidea can be recognized among the microgastrine genera by the following combination of features: fore wing with areolet open (r–m absent); hind wing with vannal lobe margin uniformly convex and hairy, rarely slightly flattened (Fig. 1c); mesoscutum typically shining with coarse, distinctly separated punctures, or sometimes punctuation nearly absent; propodeal areola highly variable, i.e., oval, hexagonal, pentagonal or sometimes poorly defined; first metasomal tergite longer than wide, almost always bearing a median apical longitudinal depression, usually parallel-sided or barrel-shaped, but sometimes slightly widening or narrowing apically; second metasomal tergite wider than long, usually shorter than tergum 3, often with a convex or sinuate posterior margin (Fig. 11); hypopygium moderate to large, sharply folded medially and usually bearing a series of median longitudinal striae; ovipositor sheath usually long and hairy throughout; ovipositor long and exerted (Fig. 10). The hosts of Dolichogenidea are most often microlepidoptera, with few exceptions. This is a very large and widespread genus, easily confused with the closely related but more diverse Apanteles, which differs in having flattened and sparsely fringed hind wing vannal lobe (Fig. 1c) and longitudinal sculpturing on the posterior portion of the mesonotum.

Genus Diolcogaster Ashmead

This genus is highly variable in both form and color pattern, but is generally recognizable by the following combination of characters: fore wing with 2nd submarginal (r–m present), in different forms, i.e., Small, triangular and quadrangular to slit-like (Fig. 14); mesoscutum without notauli, smooth to puncto-reticulate; scutellum mostly smooth with weak punctations; propodeum smooth to punctate with complete median longitudinal carina, always with or without costulae, but lateral carina never present; hind wing with vannal lobe mostly weakly convex to straight, rarely concave, its margin with row of long thick hairs, or hairs sparse or absent; hind coxa large, up to twice as long as first metasomal tergite; hind tibial spurs unequal in length, with inner spur always longer than outer spur, its length varying from half to almost as long as hind basitarsus; tergum 1 broad posteriorly to parallel-sided: medial groove of tergum 1 always present in anterior half; usually for almost entire length of tergite; Tergum 2 variable, but mostly rectangular and wider than long, usually as long as or shorter than tergum 3, tergum 2 usually with well-defined median field; tergum 3 sometimes with median field. Hypopygium short, not more than half the length of hind tibia, evenly sclerotized; ovipositor short, mostly hidden within hypopygium, exposed part much less than half the length of hind tibia; ovipositor sheaths with hairs present on entire length or just a few hairs at apex. This is a medium-sized genus with about 70 known species, most diversity occurring in the tropics and subtropics. They are mostly gregarious parasitoids of macrolepidoptera; this genus is easily distinguished from other microgastrine genera by its lack of a unique derived character and is probably polyphyletic. In this survey, only two specimens have been collected from the Junki (Kohistan) areas of Khyber Pakhtunkhwa. The high elevation of 1000-2500m in Khyber Pakhtunkhwa may be a possible reason for the scarcity of this genus. However, I expect more species to be recovered as collection continues.
Glyptapanteles Ashmead

Glyptapanteles can be distinguished from other Microgastrine genera by the following combination of features: fore wing with areolet open (r–m absent) (Fig. 15); hind wing with distal margin of vannal lobe convex, with or without a fringe of hairs; propodeum usually mostly smooth but often distinctly sculptured on all or part of its surface, rarely with a mid-longitudinal carina and never with even a trace of areola (sometimes with faint striations diverging anteriorly from metasomal foramen); first metasomal tergite narrowing toward apex or sub-parallel for the basal 0.8 then strongly rounded. Second metasomal tergite broadening posteriorly and often nearly triangular (Fig.16). Hypopygium evenly sclerotized, without a mid-longitudinal crease; ovipositor and sheath short, barely exerted and rarely longer than half the length of the hind tibia, mostly concealed by hypopygium. Glyptapanteles is a large cosmopolitan genus of several hundred species, with most diversity occurring in tropics. Species of this genus have been reared from a large variety of macrolepidotera. This genus is not so confusing, although it could be confused with some Distatrix spp. This genus is widely distributed in the Oriental region; about 25 specimens of Glyptapanteles have been recorded from different areas of Khyber Pakhtunkhwa.

Microplitis Foerster

Foerster established the genus Microplitis in 1862 with the type species Microgasters ordipes Nees von Esenbeck. Microplitis is distinguished from other microgastrines by the following features: fore wing with second (r-m) vein present and areolet large (Fig. 19); propodeum roughly sculptured, with distinct or faint medial carina; first metasomal tergite shape variable, often sculptured with antero-medial depression; suture between T2 and T3 faintly defined or absent, if defined than T2 usually shorter than T3, some indistinct median field on T2 (Fig. 20); hypopygium usually well sclerotized and sometimes elongate medially; ovipositor usually very short, often slightly down-curved throughout lengths, sheaths rarely protruding much beyond apex of hypopygium, nearly always with long hairs on exposed part, often with a pointed tuft of fused setae apically. In the plain and hilly areas of Khyber Paakhtunkhwa this is one of the most diverse microgastrine genera other than Cotesia. We have collected 102 specimens of this genus. There are 8 morphospecies examined so far. The Indo-Pak region contains different ecological zones with extremely variable climates and habitats. No attention has given to this region, although some scattered work has been done in this region, although some 95% areas still unexplored.
Microgaster Latreille

This genus can be recognizable by the following combination of features: fore wing second r-m vein present and forming a large quadrangular or sub triangular areolet; propodeum usually strongly rugose all over and a more or less distinct median carina; first metasomal tergite short, broad, a little broader apically and covering almost the entire dorsal surface of tergum 1, its surface mostly coarsely rugose; second metasomal tergum rectangular and without any delimited central area, surface rugose (Fig. 18); ovipositor sheath hairy throughout and ovipositor gradually tapered and manipulatable via a large and medially folded hypopygium (Fig. 17).

The species of Microgaster are mostly Holarctic with a few found in the Indo-Australian Region (Mason 1981) ⁸. However, here we record only two specimens belonging to genus Microgaster, which were collected from the Mardan and Peshawar districts of Khyber Pakhtunkhwa, Pakistan.

Fig 18: Microgaster Latreille from Khyber Pakhtunkhwa showing Propodeum; Strongly rugose with a distinct longitudinal carina, T1; Short and little broader apically and covering entire dorsal surface of tergum 1, T2; Rectangular and Rugolose with no delimited central area, Fig 17. Showing ovipositor sheath; hairy throughout and ovipositor gradually tapered.

Fig 19: Dorsal habitus of Microplitis Foerster from Khyber Pakhtunkhwa, Showing forewing with large areolet. Fig 20: Propodeum; roughly rugose and reticulate with distinct longitudinal carina, T1; sculptured, longer than wide with anterio-medial depression, T2; with sub rectangular small raised area.

Paroplitis Mason

This genus can be recognized by the following combination of features: fore wing second r-m vein present and formed a large quadrangular or triangular areolet; propodeum mostly smooth to mostly rugulose, with a median carina that often becomes faint posteriorly with indications of a transverse carina more or less strongly; First metasomal tergite about twice as long as wide, parallel sided, or a little broadening apically, weakly sculptured or mostly polished; and second metasomal tergum often with a pair of weakly divergent grooves delimiting a smooth sub rectangular area, but grooves absent in some species (Fig. 22); ovipositor sheath hairy throughout and about as long as hind tibia (Fig. 21); ovipositor gradually tapered and weakly decurved; hypopygium is small, sclerotized and sharply folded medially.

Paroplitis is not widely distributed. In this survey only a single large specimen is recorded as new to Pakistan from Baragali, which is 25 kilometers from Abbottabad at an altitude of 7,700 ft (2,350 m) above sea level. It requires a detailed discussion for identification on species level.
Snellenius Westwood
The genus was redescribed by Mason (1981) \cite{9} with additional useful notes provided by Nixon (1965) \cite{10}. It is distinguished from other microgastrines by the following features: fore wing with second r-m vein present, areolet large (Fig. 24); propodeum coarsely reticulate and comprised of two faces that meet transversely at a sharp angle, notaaulae deep and highly crenulate (Fig. 26); first metasomal tergite shape variable, often sculptured with anteromedial depression (Fig. 26); suture between T2 and T3 faintly defined or absent, if defined than T2 usually shorter than T3, some indistinct median field on T2; hypopygium usually well sclerotized and sometimes elongate medially; ovipositor usually very short, often slightly downcurved throughout length, sheaths rarely protruding much beyond apex of hypopygium, nearly always with long hairs on exposed part, often with pointed tuft of fused setae apically. All characters of Snellenius resemble those of the genus Microplitis except for the notaauli and propodeum, which are more coarsely sculptured (Nixon 1965) \cite{10} and have a well-developed prepectal carina (Mason 1981, \cite{8} Austin et al. 1992). The problem in the separation of Microplitis and Snellenius lies with the presence or absence of the prepectal carina (Fig. 25). This problem is more pronounced in Microplitis specimens that have an obscured or faint prepectal carina and notaauli coarsely sculptured. In Snellenius specimens if the mesopleuron is densely pilose then it becomes almost impossible to locate the traces of the prepectal carina (Ankita Gupta 2013) \cite{6}. It appears the limits to and relationships between Snellenius and Microplitis will only be resolved after a detailed study on worldwide basis.

The present study only provides five specimens included in this genus, collected from Swabi and Mardan districts of KP province of Pakistan. These specimens are very small compared to specimens of Microplitis that were collected.

Fig 21: Dorsal habitus of Paroplitis Mason from Khyber Pakhtunkhwa, showing Fig 22: Fore wing; with large triangular areolet, Fig 23: Propodeum: with medial longitudinal and transverse carina T1; longer than wide, broader apically and coarsely rugose, T2; rectangular in shape and rugose entirely.

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4. Acknowledgements
The first author is very thankful to HEC (Higher Education Commission of Pakistan) for providing necessary fund to complete this research work. We are highly thankful to Dr. Michael Sharkey, the University of Kentucky, USA to give us facilities and holotypes for identification and confirmation of microgatrine specimens. We greatly thank Miss Erika Tucker, The University of Kentucky, USA for valuable suggestions and improving the manuscript. We also gratefully acknowledge the staff members of Sharkey lab, college of Agriculture, University of Kentucky, USA and Insect museum, The University of Agriculture Peshawar for their technical support.

5. References