Chapter

Mandibular Structure, Gut Contents Analysis and Feeding Group of Orthopteran Species Collected from Different Habitats of Satoyama Area within Kanazawa City, Japan

Wael M. ElSayed, Shahenda Abu ElEla and Koji Nakamura

Abstract

A study was conducted on assemblies of various orthopteran species from distinct habitats in the Satoyama region, Kanazawa City, Ishikawa Prefecture, Japan, and a total of 50 distinct orthopteran species were registered. These species were represented by 10 families and were belonged to 17 subfamilies and 27 tribes. Results based on stereo-microscopic examination of the mandibular morphology and the analysis of gut contents suggested seven proposed feeding groups for these collected orthopteran species. Among the examined subfamilies, family Tettigoniidae proved to be the most diverse in mandibular structure and four feeding groups were assigned. This was followed by family Acrididae, which showed three feeding groups. Other families contained only single feeding group. It was noted that only five species, from family Acrididae, were graminivorous with their mandibles characterized by comparatively very short incisors and relatively wide molar regions. The analysis of gut contents of these five species proved to contain more than 80% monocotyledonous plant species. Predation and scavenging as feeding habits were also recorded in some orthopteran species.

Keywords: orthoptera, herbivorous, mandibular structure, gut contents analysis, feeding guild

1. Introduction

The strong relationship with diet makes mouthpart morphology an important trait for insect evolutionary biologists [1, 2] and systematists [3]. Isley was one of the first to study the structure and morphology of mouthparts in details and correlate morphological characteristics with various feeding habits [4].

In general, Isely defined three groups of mandibles according to the overall structure and distinctive diet: (i) graminivorous (grass feeding type) with grinding molar and incisors typically merged into a scythe like edge, (ii) forbivorous (forb or broadleaf plant feeding type) having a molar region composed of a depression encircled by elevated teeth and a strong interlocking incisor, (iii) herbivorous (mixed feeding type) that have features of both of the previously mentioned groups. Isely's initial results on mandible group [4] have since been shown to be prevalent in grasshoppers and other entomological taxa. Many authors have performed further thorough research in different locations, important among them.

Isley's initial results on mandible groups [4] have since been shown to be prevalent in grasshoppers and other insect taxa. Further itemized investigations have been directed by numerous authors in various regions, significant among them were Snodgrass [1], Gangwere [5, 6], Gangwere et al. [7] and Patterson in North America [8]; Liebermann; Gangwere and Ronderos in South America [9, 10]; Williams; Kaufmann; Gangwere and Morales [11–13] in Europe; Gangwere and Spiller; Gangwere et al. in the Mediterranean islands [14, 15]; Feroz and Chaudhry; Gapud; Kang et al. and Le Gall et al. in Asia [16–19]; Chapman in Africa [20].

A general scheme for explicating the diet of a given insect species could be started with a prudent inspections of their mandibular morphology [3, 8]. Specifically, the morphological characters of mandibles, incisors and molar surfaces are helpful tools for identifying particular species as either grass feeder or forb feeder [18, 20, 21]. Although most species with forb feeding mandibles feed on a mixture of grasses and forbs; determining an insect's diet should be followed by analyzing the gut contents for further confirmations [22, 23].

Although orthopteran species have often been regarded as polyphagous herbivores; most of these species, particularly grasshoppers, are regarded selective in their diet to some degree of selectivity, demonstrating particular food preferences [3, 24]. Occasionally, grasshoppers with forb-feeding mandibles may regularly feed on grasses or vice versa [20].

Nevertheless, there are some values in evaluating the structure of mouthparts relative to anticipating both the diet selectivity and preference to specific habitat, particularly for many rare or non-economic species that are unlikely to be studied in details. Information pertaining the feeding habits and mouthparts of different orthopteran species co-occurring in diverse habitats located in Satoyama area, Kanazawa City, Ishikawa Prefecture, Japan are fragmentary [25, 26] and there is a shortage of knowledge concerning the mandibular morphology of many orthopteran species inhabiting Satoyama. Thus, there is an urge to study the morphological characteristics and structural adaptations of the mandibles of orthopteran species co-occurring in Satoyama.

2. Materials and methods

2.1 Study area

Survey of orthopteran assemblies was achieved among four sampling sites located in Satoyama area in Kanazawa City, Ishikawa Prefecture, Japan. Satoyama is a region occupying ~74 ha, at an altitude of 150 m and positioned 5 km southwest from the center of Kanazawa City. It includes various habitats ranging from secondary forests occupied predominantly by *Quercus serrata* (Japanese vernacular name is known as Konara), *Q. variabilis* (Abemaki), *Phyllostachys pubescens* (Moso-chiku) and *Cryptomeria japonica* (Sugi) to *Cynodon dactylon* (Gyougi—shiba) and various artificial ecosystems, such as ponds, paddy lands and ordinary and reclaimed farmlands.

2.2 Protocols of sampling and collection

A standard entomological sweep net sampling technique was used for sampling and collecting different orthopteran species from the diverse habitats within

Satoyama area during different seasons of the year. The time for sampling was adjusted between 1000 and 1400 h of the day. Collected specimens were promptly killed in the field and a one liter-capacity containers including a cotton piece soaked with 70% ethanol was used for preserving the collected specimens. These containers were tightly secured with a rubber plug. The collected species were later identified, counted, sorted and kept in individually marked clean glass vials in the laboratory. These vials could be stored in freezer for a year with no apparent damage or impairment to the preserved specimens [22, 24, 27–29].

The collected Orthoptera were identified to species level by following the taxonomic key of Ichikawa et al. [25]. Furthermore, the collected species were also compared with identified museum specimens in Kanazawa University repository for further affirmation.

2.3 Mandibular structure

Mandibles were deliberately and precisely removed from the specimens by lifting the labrum and hauling out each mandible independently with the guide of fine forceps.

For easier manipulation for examination and photographing; the air-dried mandibles were pasted to the head of a #3 or #2 insect pin (depending on mandibular size). The mandibular morphological characters and apparent structure (for both ventral and dorsal sides) of 50 species of orthoptera from 10 families (Acrididae, Eneopteridae, Gryllidae, Mecopodidae, Phaneropteridae, Pyrgomorphidae, Mantidae, Tetrigidae, Tettigoniidae and Trigonididae) were examined under a stereo-fluorescence microscope (Nikon® SMZ800 series) equipped with digital camera for taking photos and TFT LCD Nikon® monitor for easier inspections.

Digital photographs were taken by Nikon® digital camera and these photos were montaged by using the Auto-Montage Syncroscopy scheme facilitated by the Laboratory of Biodiversity at Kanazawa University. For simplifying the categorization of the orthopteran species into proper feeding groups; we adopted the descriptions of mandible types declared by Isley [4]. The examined species were categorized into seven major groups: Forbivorous (referred subsequently as F), Herbivorous or Mixed-feeders (H), Graminivorous (G), Scavengers (S), Herbivorous with observed scavenging behavior (H_S), Forbivorous with scavenging behavior (F_s) and Predators (P). Detailed explanations of these groups are given hereafter in the methodology.

2.4 Field cages experiment

Live different orthopteran species were held and reared under natural environmental conditions in proper wooden cages and were supplied with almost all accessible plant species collected from the sampling field to minimize the hunger effect. Continuous inspections on feeding behavior were performed for continuous 3 h in three replicate field cages in each season of the year. Obtained results from field cages experiments were compared to those acquired from mandibular examination.

2.5 Gut contents analysis and feeding groups

In an attempt to glean the feeding category of each orthopteran species; gut contents analysis was carried out in concordance with mandibular structure examination.

The proportions of four main categories (monocotyledonous plant species, dicotyledonous species, orthopteran or animal parts, and scavenging observations in caged species) were calculated and considered to classify each of the orthopteran species into one of the suggested seven feeding categories:

1. Herbivorous (H): in which the number of fragments of dicotyledonous plant is almost equal to the number of fragments of monocotyledonous species.
2. Herbivorous with scavenging behavior (H_s): the same as herbivorous group with some scavenging actions were recorded in laboratory caged species.
3. Graminivorous (G): the number of fragments of monocotyledonous species is more than 75% of the gut contents.
4. Forbivorous (F): the number of fragments of dicotyledonous plant species is more than 75% of the gut contents.
5. Forbivorous with scavenging behavior (F_s): the same as forbivorous group with some scavenging actions were recorded in laboratory caged species.
6. Scavengers (S): plants species (especially roots or tubers) and dead orthopteran and/or oligochaeta parts were encountered in almost equal proportions.
7. Predators (P): all contents of the gut were insect or other orthopteran body parts with no occurrence of plant fragments.

Collected orthopteran specimens were deposited in a catalogued repository in Kanazawa University in special boxes containing small naphthalene coated sachets for further specimen protection against destructive pests.

3. Results and discussion

A total of 50 orthopteran species, belonging to 10 families representing 17 subfamilies and 27 tribes, were collected from different habitats of Satoyama. These species were sampled from different habitats including open grasslands, forest margins, ponds and paddy fields. The stereo-microscopic examination of these 50 orthopteran species revealed that these species belonged to seven major feeding groups (**Table 1**). Among the examined subfamilies, family Tettigoniidae proved to be the most diverse in mandibular type with four feeding groups could be observed. This was proceeded by family Acrididae which possessed three feeding groups. Other families were observed to contain only sole feeding group (**Table 2**).

Species from the family Acrididae, short-horned grasshoppers, and family Tettigoniidae, long-horned grasshoppers, can occur in a diverse of habitats, usually in dense vegetation like open grasslands or around paddy fields or even pond localities. Species belong to these two families were found to be quite active in both walking and flying. It is interesting to note that species with graminivorous type mandibles, were characterized by extremely slender and elongated bodies and were encountered on the edges of ponds. This was in accordance with the findings of other authors [4, 30, 31]. These species typically grasp the stems of emergent grass or grass-like vegetation such as sedges or cattails, blending in almost perfectly. On the other hand, collected species from family Oedipodinae were split into three mandibular types: graminivorous, forbivorous and herbivorous [32]. This signifies a more grass-dominated diet in their feeding behavior.

DOI: http://dx.doi.org/10.5772/intechopen.88705

Family	Subfamily	Tribe	Acridid species	Feeding group*
Acrididae	Acridinae	Acridini	*Acrida cinerea*	G
		Parapleurini	*Stethophyma magister*	G
	Melanoplinae	Podismini	*Parapodisma Mikado*	F
	Oedipodinae	Aiolopini	*Aiolopus thalassinus tumulus*	H
		Oedopodini	*Sphingonotus japonicus*	F
		Locustini	*Oedaleus infernalis*	G
		Trilophidiini	*Trilophidia annulata*	G
	Oxyinae	Oxyini	*Oxya yezoensis*	G
Eneopteridae	Oecanthinae	Oecanthini	*Oecanthus simulator ichikawa*	F
Gryllidae	Gryllinae	Gryllini	*Acheta domesticus*	S
			Loxoblemmus equestris	S
			Loxoblemmus sylvestris	S
			Loxoblemmus tsushimensis ichikawa	S
			Stethophyma magister	S
			Teleogryllus occipitalis	S
			Teleogryllus emma	S
			Velarifictorus asperses	S
			Velarifictorus Mikado	S
			Velarifictorus ornatus	S
		Modicogryllini	*Modicogryllus siamensis*	S
	Sclerogryllinae	Sclerogryllini	*Sclerogryllus punctatus*	S
Mecopodidae	Mecopodinae	Mecopodini	*Mecopoda niponensis*	F_s
Phaneropteridae	Phaneropterinae	Ducetini	*Ducetia japonica*	F_s
		Phaneropterini	*Phaneroptera falcate*	F_s
			Phaneroptera nigroantennata	F_s
Pyrgomorphidae	Pyrgomorphinae	Atractomorphini	*Atractomorpha lata*	F
Mantidae	Mantinae	Mantini	*Tenodera angustipennis*	P
			Tenodera aridifolia	P
Tetrigidae	Scelimeninae	Criotettigini	*Criotettix japonicas*	F
	Tetrigidae	Tetrigini	*Euparatettix tricarinatus*	F
			Tetrix japonica	F
			Tetrix macilenta	F
			Tetrix minor ichikawa	F
			Tetrix nikkoensis	F
			Tetrix silvicultrix ichikawa	F

Family	Subfamily	Tribe	Acridid species	Feeding group*
Tettigoniidae	Conocephalinae	Conocephalini	*Conocephalus japonica*	F_S
			Conocephalus melaenus	F
		Copiphorini	*Euconocephalus varius*	F
			Ruspolia dubia	F_S
	Tettigoniinae	Decticini	*Chizuella bonneti*	F_S
			Eobiana gradiella ishikawa	H
			Eobiana engelhardti subtropica	F_S
		Gampsocleidini	*Gampsocleis Mikado*	H_S
		Hexacentrinae	*Hexacentrus japonicas*	H_S
	Tettigoniinae	Tettigoniini	*Tettigonia orientalis*	F
			Tettigonia sp. *6***	F
			Tettigonia sp. *8*	F
Trigonididae	Nemobiinae	Pteronemobiini	*Dianemobius furumagiensis*	S
			Pteronemobius fascipes	S
	Trigonidinae	Trigonidini	*Trigonidium pallipes*	S

**F, Forbivorous (Forb-feeder); H, Herbivorous (Mixed-feeder); G, Graminivorous (Grass-feeder); S, Scavengers; H_S, Herbivorous with observed scavenging behavior; F_s, Forbivorous with observed scavenging behavior; P, Predator;*
***Species 6 and 8 according to Ichikawa et al. [25].*

Table 1.
Check-list of orthopteran species inhabiting different habitats of Satoyama area with their family, subfamily, tribe and feeding group.

However, these species are much more divergent in their feeding category in which some may be completely graminivorous or forbivorous. Most of the species were found on the ground in open areas or on bare soil, rarely on plant species or grasses. As a general finding, the greatest diversity in mandibular structure was observed in subfamily Oedipodinae in comparison with other orthopteran subfamilies [32]. Equitably even distribution of the three mouthpart types in this group were also recorded [4, 6, 18].

Mantidae, on the other hand, where observed to be represented by only two *Tenodera* species (*Tenodera angustipennis* and *T. aridifolia*). Concerning their feeding habit, these two species were completely predacious. Their mandibles were characterized by sharp incisor points used to capture and pierce the captured prey, and relatively long terebral ridge used to kill and slice prey into small pieces. Results from the analysis of gut contents of these mantid species revealed fragments of chitinous arthropod exoskeleton and other body parts including parts from antennae, wings or even legs affirming their zoophagous feeding behavior.

It was interesting to observe that tenth of the collected orthopteran species, five species out of the 50 species, were graminivorous, all were from the family Acrididae. These five species were characterized by very short incisors and relatively wide molar region in their mandibles. The molar area of some individuals of *Oxya yezoensis* (as one of these five species) showed a severe erosion in the molar region. It has to be mentioned that, feeding on grasses could be one avenue by which some orthopteran species may avoid toxic chemicals [22, 23, 33].

Family	Number			
	Subfamilies	**Tribes**	**Observed species**	**Feeding group**
Acrididae	4	8	8	3
Eneopteridae	1	1	1	1
Gryllidae	2	3	12	1
Mecopodidae	1	1	1	1
Phaneropteridae	1	2	3	1
Pyrgomorphidae	1	1	1	1
Mantidae	1	1	2	1
Tetrigidae	2	2	7	1
Tettigoniidae	2	6	12	4
Trigonididae	2	2	3	1
Total	17	27	50	7

Table 2.
Number of families, subfamilies, tribes, species and feeding group of orthopteran species co-occurring in different habitats of Satoyama area.

In this process, little or no energy, or other resources, would need to be spent on the detoxification process [34]. Contents of the gut of these five graminivorous species contained silica particles in relatively minor amounts. It is assumed that these silica particles could be ingested accidentally during feeding regime and thus accelerating the erosion of the molar area especially in old individuals due to severe and continuous friction [22, 23].

The analysis of gut contents of these five species revealed that the contents contained more than 80% monocotyledonous plant species. Controversially, *Acrida cinerea* as a graminivorous species, less than 12% of dicotyledons plant species were also encountered in their guts. Some authors in their field and laboratory works on a related acridid, *Acrida pellucida*, observed that this species may select non-graminous plants (dicotyledons) for enhancing the reproductive potential since these dicotyledon species showed a pronounced effects on both fecundity and developmental rates in laboratory rearing and food-choice tests [19, 22, 23, 35–37]. It could be assumed that the acridid, *Acrida cinerea*, may exploit some dicotyledonous plant species for augmenting specified biological and physiological processes. In this study, the acridid species belonging to the subfamily Acridinae are typically considered to be grass-feeders, displaying the classic graminivorous type mandibles [4, 20, 22].

Family Gryllidae was typically represented by 12 gryllidae species. These species showed mandibles with sharp incisors and comparatively long knife-shape terebral ridge. These mandibular modifications could delineate a predacious feeding habit. However, the gut content analysis revealed that parts from plant roots, tubers or even debris (38%) and subterranean arthropod species including amphipod and isopod species (62%) were collected from their guts. Consequently, the feeding group of these 12 gryllidae species could be confined to the scavenging habit.

Examination of Tetrigidae, represented by seven species, revealed that these species were mainly forbivorous (F_m). Their mandibles were characterized by pointed and sharp incisor points while their molar region was relatively small. The contents of the gut of these species contained dicotyledonous plan species without any presence of monocotyledonous ones.

Due to merely one representative species from the three subfamilies Eneopteridae, Mecopodidae and Pyrgomorphidae; determination of the mandibular structure of these families was relatively limited (**Table 1**). However, the supposed major mandible type and in turn the feeding group was mostly confined to the forbivorous type (F) where more dicotyledonous plants with nearly 79% dominance were consumed in much greater amount than monocotyledonous species (21%) as emphasized by gut contents analysis.

The determination of the mandibular structure of three subfamilies, Eneopteridae, Mecopodidae and Pyrgomorphidae, was comparatively restricted due to the fact that only one representative species from each of the three subfamilies could be collected (**Table 1**). However, the main mandibular type and in turn the feeding category was mostly restricted to the forbivorous group where more dicotyledonous species (79%) were devoured in relatively greater quantities than monocotyledonous ones (21%) as verified by the assessment of the analysis of gut contents.

At family level, it is noted that family Tettigoniidae with 12 species, was the most diverse family in both mandibular type and feeding group. It was obvious that four different feeding groups could be detected in Tettigoniidae. This was proceeded by family Acrididae in term of feeding group. Acrididae which harbored eight species had displayed three distinctive feeding groups as indicated in **Table 2**. Diversely, other families possessed only a single feeding group irrespective to the number of species (**Table 2**). Moreover, results perceived from **Table 2** showed that both family Gryllidae and family Tetrigidae (12 and 7 species, respectively) retained only one type of mandible and a single feeding group.

In all cases, a range of food of plant and/or animal origin was used in their diet, even though some were used infrequently. Thus these orthopteran species inhabiting different habitats in Satoyama area could be considered polyphagous species.

Cates [38] depicted the degree of diet specialization into the following three criteria: (1) monophagy: where one or more species within a genus; (2) oligophagy: two or more tightly associated genera; and (3) polyphagy: two or more plant families. In fact, none of the orthopteran species regarded in this research can be considered as either monophagic or oligophagic species. A variety of plant and/or animal foods were consumed in their diet in all instances, although some were rarely devoured. Thus, these species of orthopteran co-occurring in the diverse habitats of Satoyama area could be regarded as polyphagic species.

4. Conclusion

In conclusion, it is far from accuracy to roughly connect between mouthparts morphology and diet. Some authors like Mulkern was persuaded that only the grossest associations between mandibular structure and their diet regime (i.e., graminivorous, forbivorous, and herbivorous) could be made [3]. Some orthopteran species, especially grasshoppers with forb-feeding adapted mandibles, occasionally feed on grasses on a regular basis or vice versa [20, 22]. Nevertheless, the evaluation of mouthpart structure and morphology as a predictive avenue in determining diet and habitat preference in orthopteran species has some importance, particularly for the rare or non-economic species that are unlikely to be studied in details. Thus, the analysis of gut contents in parallel with laboratory examinations and precise observations on feeding behavior could be used as confirmation cues for the discovery of the mandibular structural adaptations. This would solve some hidden aspects that could not be deduced from the morphological characters of the mandibles if they were adopted alone.

Acknowledgements

The authors wish to acknowledge the staff at the Laboratory of Ecology, Graduate School of Natural Science and Technology, Kanazawa University for the laboratory facilities provided. Also, sincere gratitude is extended to Prof. Dr. Nakamura Koji (Kanazawa University, Japan) for his keen hospitality and encourages for completing the research and writing the manuscript.

Conflict of interest

The authors declare that there are no conflicts of interest associated with this article.

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