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## Original article

Morphological and functional diversity of foliar damage on *Quercus mongolica* Fisch. ex Ledeb. (Fagaceae) by herbivorous insects and pathogenic fungiJae-Cheon Sohn<sup>a</sup>, Nang-Hee Kim<sup>b</sup>, Sei-Woong Choi<sup>a,c,\*</sup><sup>a</sup> Institute of Littoral Environment, Mokpo National University, Muan, Jeonnam, Republic of Korea<sup>b</sup> Department of Biology, Mokpo National University, Muan, Jeonnam, Republic of Korea<sup>c</sup> Department of Environmental Education, Mokpo National University, Muan, Jeonnam, Republic of Korea

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## ABSTRACT

Insect-feeding damage on plants provides insight into plant–insect interactions along environmental gradients. However, cataloging such damage has been limited for fossil flora or extant tropical vegetation. We explored the diversity of insect-feeding damage on *Quercus mongolica*; one of the major tree species in the East Asian temperate forests. Eighty-six types of damage were cataloged from five sites on three mountains in Korea. Each damage type identified from our study was compared to previously proposed types of damage. The possible or confirmed origin of each damage type was discussed. The diversity of the damage types was highest from Mt. Seongjusan, followed by Mt. Jirisan and Mt. Hallasan. The frequency of each damage type on the leaves varied among research sites. Damage involving sap-sucking arthropods and pathogenic fungi was most frequent and exhibited greater among-site variation among the eight functional categories.

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## Introduction

Plant-feeding insects, which constitute about 25% of all described species, are a ubiquitous feature of terrestrial life (Schoonhoven et al 1998; Strong et al 1984). Accordingly, understanding the evolution of insect herbivory and its diversity is essential to understanding the evolution of terrestrial ecosystems. Mitter et al (1988) showed that the evolution of phytophagous habits in insects accelerated their diversification. This process involves an arms race between plants and herbivorous insects. The evolution of various defenses in plants facilitates host shifts or exploitation of various strategies to attack plant parts among insects (Winkler and Mitter 2008). Several studies (e.g., Farrell and Sequeira 2004; Kaila et al 2011; Sohn et al 2016) have demonstrated that diverse feeding modes are

one of the major features in species-rich lineages of phytophagous insects.

Insect-feeding modes have been studied through the traces that they leave behind, rather than *in situ* observation of their feeding activity. The feeding damage caused by plant-feeding insects and mites is regarded as a good indicator of their taxonomic and functional diversity (reviewed in Andrew et al 2012). Accordingly, such data have been used to investigate the effects of extant or ancient environmental gradients on insect herbivory (Adams et al 2010; Currano et al 2008, 2010; Wilf and Labandeira 1999) and the evolutionary diversification of the insect groups (Labandeira 2006). These data are also potentially useful for developing effective pest-control (Hartman et al 2015) or conservation (Kathiresan 2003) programs. For such studies, the feeding damage needs to be reliably identified at the species level or categorized by functional types representing feeding guilds. To date, morphotyping of insect-feeding damage has primarily been applied to fossilized leaves (Labandeira et al 2007). In their study of an extant tropical forest, Carvalho et al (2014) showed that only a portion of the insect-feeding damage was preserved in fossils. Therefore, comprehensive monitoring of the damage on extant flora is necessary to enable

\* Corresponding author. Tel.: +82 61 450 2788.

E-mail address: [choisw@mokpo.ac.kr](mailto:choisw@mokpo.ac.kr) (S.-W. Choi).

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a better understanding of the diversity of insect-feeding guilds. In this study, we attempted to catalog the insect-feeding damage on the temperate vegetation for the first time with the Mongolian oaks (*Quercus mongolica*) as an example.

Mongolian oaks are one of the major components of the East Asian cool-temperate deciduous forests (Zeng et al 2015). These trees play a fundamental role in the forests, principally through serving as food plants for numerous insect herbivores that are important food sources for other animals (Kim and Kil 2000; Yang et al 2014). There are many records of insects feeding on *Q. mongolica* that usually consist of simple statements in taxonomic papers or field notes, while synthetic studies of such records are rare. For example, Teramoto (1993) compiled lepidopterans feeding on *Q. mongolica* that belong to 25 families and 254 species from Japan. It is well known that the composition of insects associated with the oak species can differ by biotic and abiotic factors (Hata et al 2011).

This study was conducted to compile all of the damage types caused by the feeding activity of insects and mites on Mongolian oaks on three representative mountains of Korea and to compare these with other morphotyping systems of insect-feeding damage. Moreover, we discussed its possible makers, frequency in sampled leaves, and among-site variation.

## Materials and methods

### Collecting samples

The leaves of *Q. mongolica* were collected from a total of five sites of three mountains: two on Mount (Mt.) Hallasan, Sajebi Hill (SJB: 3322'32.2"N 12629'58.8"E, alt. 1410 m) and lower Yongsil (LYS: 3320'54.8"N 12629'47.6"E, alt. 1210 m); on Mt. Seongjusan, Boryeong (SJS: 361950.5"N 12637'54.3"E, alt. 380 m); and two on Mt. Jirisan, Seongsamjae (SSJ: 3518'20.8"N 12730'44.6"E, alt. 1073 m) and Sangseonam (SSA: 3517'31.5"N 12729'39.4"E, alt. 682 m). Foliar damage tends to be accumulated with the lapse of time. To maximize the accumulation, we collected leaves in autumn, before they changed color (12 September 2015 in Mt. Hallasan, 17 September 2015 in Mt. Seongjusan, and 21 September 2015 in Mt. Jirisan). In each research site, we collected two or three branches with all attached leaves that were randomly selected from two randomly-sampled tree stands. After detachment from the branches, the leaves were placed into layers of newspaper and a plastic envelope labeled with collection data, then stored in a -20°C freezer. A total of 246 leaves were collected from Mt. Jirisan (140 in SSJ; 106 in SSA), 88 from Mt. Seongjusan, and 330 from Mt. Hallasan (199 in SJB; 131 in LYS).

Insect-feeding damage is any trace involving the removal or modification of plant tissue through feeding activity of insects and mites, which is sometimes followed by the healing process by hosts or fungal infection. Labandeira et al (2007) expanded the range of insect-feeding damage to include any damage caused by terrestrial arthropods and pathogenic fungi. We followed this definition when identifying damage on Mongolian oaks. Damage types were identified and compiled from all the collected leaves at each survey site. Only the damage to foliar parts including laminae, veins, and petioles were considered and assigned to types according to their position on the leaves, relation to the veins, and overall shape. We tried to assimilate our morphotyping systems into two previous ones, that were, Labandeira et al (2007) and Carvalho et al (2014), but found some problems. Labandeira et al (2007) typed the damage from fossilized leaves and considered the final outcomes after fossilization. Carvalho et al (2014) used extant leaves in tropical forests and considered the process of each type of damage through their field observation of damage makers. However, these procedures could not be applied to our study; therefore, we decided to devise our own morphotyping system for damage types on *Q. mongolica* and to

compare those with two previous morphotyping systems. We numbered each damage type with a prefix of abbreviation for each functional damage category (MF – marginal feeding; HF – hole feeding; SF – surface feeding; SK – skeletonization; PS – piercing/sucking; MI – mining; GA – galling; FG – fungal).

The representative damage and its variation in each damage type were photographed using a digital camera (NIKON D30, Nikon Corp., Tokyo, Japan) attached to a stereoscope (LEICA L2, Leica Microsystems, Wetzlar, Germany). The leaves examined were tagged with numbered white tape, dried in the standard plant press (BioQuip, CA, USA), and finally stored in plastic envelopes. The leaf collection was deposited at the Department of Environmental Education, Mokpo National University, Muan, Jeonnam Province, South Korea.

### Functional classification

We followed the guide developed by Labandeira et al (2007) for classification of functional feeding groups for terrestrial arthropod damage types on plants. We treated only the damage on plant leaves so that seed predation was excluded from our classification.

The functional feeding groups in the present study include the following.

- (1) MF: damage makers consume entire or only nonvascular foliar tissue including marginal or apical area. Direction can be from the central area to margins or *vice versa*.
- (2) HF: damage makers consume entire foliar tissue except marginal area, leaving various sizes and shapes of windows that can be limited by leaf veins.
- (3) SF: damage makers consume only one layer of the epidermis and entire or partial mesophyll tissue. The damage progresses horizontally, leaving discolored or transparent areas depending on the degree of consumption of mesophyll. The resulting damage can be reticulate if foliar-vascular tissues are not completely consumed.
- (4) SK: Labandeira et al (2007) and Feng et al (2014) defined SK as feeding damage caused by the complete consumption of the interveinal leaf tissue. Certain surface feeding traces resemble skeletonization, but one layer of the epidermis and leaf veins remain in surface feeding.
- (5) PS: this damage comprises various shapes, sizes and arrangements of puncture marks caused by sap-feeding arthropods in Labandeira et al (2007). Observation of such marks is often difficult in intact leaves. Instead, we identified piercing/sucking damage with discoloration or malformation of foliar tissue due to fungal infection.
- (6) MI: damage makers consume partial or entire mesophyll tissue, leaving both epidermal layers intact. Direction and shape of mining path vary in each morphotype. There can be trails or patterns of dropping derived from mine makers.
- (7) GA: damage makers induce abnormal growth of foliar tissue for breeding chambers. The resulting damage can be swelling, curls, blisters, nipples, or erineae on the upper or lower leaf surface.
- (8) FG: damage makers are not arthropods, but pathogenic fungi. These types of damage are included in the present study because of their frequent association with arthropods in the infestation stage. Fungal infection may result in either decay or abnormal growth of foliar tissue.

### Annotation entries

Compatible types: observed foliar damage was compared to morphotypes identified by Labandeira et al (2007) and Carvalho et al

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