

Pollinators and dispersing insects of seeds in *Fritillaria koidzumiana* (Liliaceae)

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Naohiro Naruhashi¹, Yuko Takata¹ and Hisashi Negoro²: Pollinators and dispersing insects of seeds in *Fritillaria koidzumiana* (Liliaceae)

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The genus *Fritillaria* L. (Liliaceae) containing 100 species, occur in West Europe to East Asia and North America (Mabberley 1997). In Japan, nine species have been recorded (Naito 2005), of which six species are listed as endangered plants in The Red Data Plants (Yahara 2003).

Fritillaria japonica group (Noda 1975 ; Noda and Naruhashi 1988 ; Naruhashi et al. 1997) containing *F. koidzumiana* Ohwi are unique in that they exhibit achiasmate meiosis in pollen mother cells (Noda 1968, 1975), which is not observed in other plants in the world. Several years ago we began to examine *F. koidzumiana* with the aim to reveal the life history of this remarkable plant from its reproductive strategy because it is morphologically distinct from other *Fritillaria* species. Furthermore it is a member of spring ephemerals, shows the unique meiosis in PMCs and its taxonomically related species are endangered.

Though Bakhshi Khaniki and Persson (1997) reported pollinators of *Fritillaria*, no pollinators of Japanese *Fritillaria* species except for *F. camtschatcensis* are known until now (Yashima et al. 1997). On the other hand, *F. koidzumiana* was reported as cross pollination type in breeding system by Kawano et al. (2004) and that is confirmed (Naruhashi and Takata, unpublished). Therefore, it is highly meaningful study to reveal pollinators of the plant.

Fritillaria has a capsule which is upright and dry in matured time, and flat seeds with a wing are dispersed by wind. However, plants of Japanese *F. japonica* group are not upright and not dry up in matured capsules and have somewhat globose seeds without a wing. Supposing that

morphological characters, we can speculate no wind dispersal of the plant.

Erythronium japonicum belonging to the Liliaceae, is one of the famous spring ephemerals and growing with *F. koidzumiana* and blooming the same time, i.e., the early to the middle April in low elevation and the late April to early May in rather higher elevation in mountains in Toyama Prefecture. Both species are perennial herbs of the temperate deciduous forests of Japan. The former, *E. japonicum* has seeds accompanied by small elaiosomes and is reported to be myrmecochorous (Kawano et al. 1982). Kawano et al. (2004) suggested that the seeds of *F. koidzumiana* have elaiosomes and the possibility of dispersal seeds by ants. Doubtless an appendage of the seed of *Fritillaria* seems to be an elaiosome. Therefore, we expected to observe the seeds of *F. koidzumiana* to be moved around by ants.

The purpose of this paper is to report on the pollinators and dispersal insects of seeds in *F. koidzumiana*.

Materials and methods

Pollinators were observed in the peak of flowering at four sites, see Table 1. Visiting insects were captured by net after confirmation of insects climbing into flower. Pollen on the insect body are examined at laboratory after capture.

As noted previously, the seeds of *Fritillaria* are accompanied by small elaiosomes, and thus are assumed to be susceptible to myrmecochory. In order to know the dispersibility of *Fritillaria* seeds by ants, seeds were presented to ants in the right neighboring place of the habitats of the



Fig. 1. Pollinators and ants dispersing seeds of *Fritillaria koidzumiana*. A and B: *Andrena benefica* visiting flower. C: Seeds of *Fritillaria koidzumiana*, with scale of 1 mm. D: *Lasius japonicus* carrying a seed. E: *Aphaenogaster famelica* carrying a seed. F: *Formica japonica* carrying a seed.

Table 1. Sites, dates, time, weather and temperature for investigation of pollination of *Fritillaria*

Sites, altitude	Date Time	Weather Temperature
Toyama Pref., Toyama-shi, Yatsuo-machi, Yuzunoki, 130 m	Apr. 15, 2006 10:00–11:00	Cloudy 13.4°C
Toyama Pref., Toyama-shi, Yatsuo-machi, Naka, 120 m	Apr. 18, 2006 10:30–11:00	Fine to slightly cloudy 22°C
Toyama Pref., Toyama-shi, Yatsuo-machi, Naka, 120 m	Apr. 24, 2006 9:30–11:30	Cloudy 16.4°C
Toyama Pref., Nanto-shi, Fukumits-machi, Tori-dam, 360 m	May 4, 2006 13:30–14:30	Fine 23°C

Table 2. Investigation sites, date and dispersal ants of *Fritillaria* seeds

No. Sites and altitude	Date	Weather and time	Ants
1 Naka in Yatsuo-machi, Toyama-shi, 120 m	May 30, 2006	Fine, 14:20–14:50	<i>Formica hayashi</i> , <i>Aphaenogaster famelica</i> , <i>Lasius japonicus</i>
2 Naka in Yatsuo-machi, Toyama-shi, 120 m	May 30, 2006	Fine, 14:50–15:10	<i>Lasius japonicus</i>
3 Naka in Yatsuo-machi, Toyama-shi, 120 m	May 30, 2006	Fine, 15:10–15:30	<i>Formica japonica</i>
4 Campus of Univ. of Toyama, Toyama-shi, 10 m	May 30, 2006	Fine, 16:10–16:20	<i>Lasius japonicus</i>
5 Kurehayama, Toyama-shi, 15 m	Jun. 1, 2006	Fine, 13:00–15:00	<i>Formica japonica</i>
6 Naka in Yatsuo-machi, Toyama-shi, 120 m	Jun. 3, 2006	Fine, 13:30–16:00	<i>Lasius japonicus</i>
7 Sitanomyo in Yatsuo-machi, Toyama-shi, 220 m	Jun. 5, 2006	Fine, 17:00–17:30	<i>Formica japonica</i>

1, 2, 3 and 7, right neighboring place of population ; 4 and 5, place out of population : 6, in population.

plant (see, 1, 2, 3 and 7 in Table 2). After carrying the seeds into their nests, the ants were captured for identification. Captured insects in the present study are held in trust in the sampling room of the Toyama Science Museum.

Results and discussion

Bees flying about, striding into bell-shaped *Fritillaria* flower and sucking nectar and collecting pollen, were observed (Fig. 1 A and B). These bees are shown in Table 3. In the table a figure are a number of captured insects. For example on 24th of April we observed ten bees which in-

truded into the flower, five bees, however, were captured. Even four times and in all places *Andrena benefica* Hirashima (Hymenoptera, Andrenidae) was pollinator and moreover two bees, *An. hebes* Pérez in Yuzunoki and *An. minutula* (Kirby) in Tori-dam were observed as pollinator. These three *Andrena* are common species in Japan (Tadauchi and Hirashima 1986).

Behavior of female *Andrena* bees when they visited the flowers of *Fritillaria* was observed to be as follows. They landed on the outside of tepal, intruded into the inside of tepal, there sucked nectar and afterward collected pollen

with thorax, postical abdomen, and hind-legs clinging or moving on a style. They sometimes stopped on the style and sucked nectar of tepal. After moving to apex portion of style or to apical part of tepal, they ultimately departed from the flower. Pollen was adhered on antical thorax of *An. benefica* and *An. hebes* when they made a stop inside of the flower, but was not adhered in small *An. minutula*. Pollen grains were clung on the stigma when bees moved on style and departed from the flower. We observed the *Fritillaria* pollen grains on the body (thorax, abdomen and hind-legs).

However, *Lasius japonicus* might be not a pollinator though the ant agitated similarly and sucked nectar in the flower inside. Because of a larger flower size and a larger amount of nectar we expected bumble-bees (*Bombus*) to act as pollinator of *Fritillaria*. However we could not observe their visit of the flower.

On the basis of this result, the pollinator of *Fritillaria* in Toyama Prefecture is believed to be *Andrena* species.

Andrena is the largest genus of bees in England and active in March and April (Proctor and Yeo 1973). Tadauchi (1992) reported *Andrena* in Japan visited flower from spring to early summer and oligotropic. Spring ephemeral *Fritillaria koidzumiana* employing *Andrena* species as pollinator is thought to do so as strategy for survival.

Erythronium japonicum of the same family which grows with the present plant and blooms at the same time, has many kinds of pollinators, such as Lepidoptera, Coleoptera, Hemiptera, Diptera and Hymenoptera (Kawano and Nagai 1982). The difference of pollinator from *Fritillaria* may be explained as that of the flower morphology, flower scent and nectar component.

The pollinator of *F. camschatcensis* in Mt. Hakusan located in neighboring prefecture was reported as blowfly, *Aldrichina grahami* (Diptera, Cyclorrhapha) by Yashima et al. (1997). *Fritillaria camschatcensis* and *F. koidzumiana* are growing in the same geographical area, Hokuriku District in Japan Sea side, but ecologically both plants are different, because the former occurs in alpine region, the latter occurs in lower mountains, lower than deciduous forest

zone. Because of severe environmental conditions, there are only a few insects acting as pollinators in the alpine zone. On the other hand since *F. camschatcensis* is pollinated by blowfly, it must show a myophilous syndrome (somewhat evil-smelling to attract blowfly, style apex deeply parted and greatly recurved, etc.).

Different shapes of nectar glands and different amounts and composition of nectar in the genus *Fritillaria* is reported well by Rix and Rast (1975) and Bakhshi Khaniki and Persson (1997). Pollinators of many species are bees or bumblebees, those of *F. crassifolia* and *F. graeca* group are wasps and that of *F. imperialis* is bird (Bakhshi Khaniki and Persson 1997).

In the case of *F. koidzumiana*, flower is broadly campanulate, nodding and nectar glands occur in grooves running along the perianth segments (Naruhashi et al. 1997; Rix 2005). No nectar sugars, i.e., proportion of fructose and glucose, were reported of the present flower. Generally speaking, pollination of *F. koidzumiana* is a type of bees or bumblebees.

The seeds of *Fritillaria* are nearly ellipsoidal and have an elaiosome (Fig. 1 C). The results of the experiment of removing seeds by ants are shown in Table 2. We observed ants hold the part of elaiosome on a seed and carried the seed into their nests (Fig. 1 D, E and F). The ants observed in natural population of *Fritillaria* were *Lasius japonicus* Santschi (Formicidae, Lasiini). Similarly, the ants observed as dispersal ant at right neighboring places of populations were *Formica hayashi* Terayama and Hashimoto (Formicidae, Formicini), *For. japonica* Motschulsky (Formicidae, Formicini) and *Aphaenogaster famelica* (Fr. Smith) (Formicidae, Plagiopelidini).

At Kurehayama out of the habitat of *Fritillaria*, an experiment, how do ants carry seeds was carried out. A sample was 10 seeds collected at Naka in Yatsuo-machi, and was left near the nest of *Lasius japonicus*. The time of seeds removal by ants was eleven minutes.

A list of ants of myrmecochores in warm-temperate zone of Japan by Nakanishi (1988) contains these four ants (described *For. japonica* as *For. fusca* and *L. japonicus* as *L. niger* in his paper). We consider that these four ants are famous for seed dispersal. In the above mentioned

Table 3. Pollinators in each site of *Fritillaria* (see, Table 1)

Sites (date)	Pollinators	Others
Yuzunoki (4/15)	<i>Andrena hebes</i> 4 ♀ <i>An. benefica</i> 1 ♀	
Naka (4/18)	<i>An. benefica</i> 4 ♀	
Naka (4/24)	<i>An. benefica</i> 5 ♀	<i>Lasius japonicus</i>
Tori-dam (5/4)	<i>An. minutula</i> 4 ♀ <i>An. benefica</i> 1 ♀	<i>Lasius japonicus</i>

paper no seeds of Japanese *Fritillaria* species are shown, because of the plants occurring in cool-temperate zone. Furthermore, Nakanishi (1988) reported on the weight of disseminule (seed) for myrmecochory plant, the lightest 0.178 mg in *Corydalis racemosa*, the heaviest 47.537 mg in *Akebia trifoliata* and 0.4–2 mg in the most species per single seed weight. Since a seed weight of the present *Fritillaria* is about 5.3 mg, it seems to be heavy for carrying by ant. Certainly the seed of *Fritillaria* is considered to be large compared with the body size of ant, especially that of *L. japonicus*.

These four ants are common and they are distributed in lowland to mountainous region from Hokkaido to Kyushu and are mainly in the edge of forests in their habitat. Moreover their nests are made in soil (Sonobe and Onoyama 1991; Terayama and Yamauchi 1991; Watanabe and Yamane 1992). The seeds of *Fritillaria* are not dispersed but are prevented from being feeded by other animals and from dessication after removal into their nests by ants.

That *Fritillaria* is myrmecochorous, is not reported in Japan (Nakanishi 1999) and even in the world (Beattie 1983). This is the first record of the genus on myrmecochorous plant and may be understood as a result of a quite different habitat, because many species of *Fritillaria* in the world occur in meadows and the present species grows in woodland. Furthermore, many species in *Fritillaria* in meadows are dispersed by the wind because of upright dry capsules and flat seeds with a wing (cf. Turrill and Sealy 1980). On the contrary, *F. koidzumiana* occur-

ring in woodland is myrmecochorous because of nodding wet capsules and wingless globose seeds with a prominent elaiosome.

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- (Received July, 27, 2006; accepted September 30, 2006)
- 鳴橋直弘¹・高田由子¹・根来 尚²: ユリ科コシノコバイモの送粉昆虫と種子散布昆虫
- 世界の植物の中で、花粉作成の減数分裂時に、相同染色体がキアズマを作らない唯一の植物群であるコバイモ (*Fritillaria japonica* group) は、8種からなり、その内コシノコバイモ (*Fritillaria koidzumiana*) 以外は絶滅危惧植物に指定されている。それ故早急にこの群の生活史の解明が急がれている。コシノコバイモは産地もまた個体数も多く実験に適した植物であり、筆者らは数年前よりこの植物を生殖戦略という面から分析している。この植物が他殖であることを観察 (河野他 2004; 鳴橋・高田 未発表) したが、送粉昆虫は不明だったので、2006年4月15日から5月4日に4回、富山県の3カ所の生育地において観察・調査した。その結果、4回とも共通して膜翅目ヒメハナバチ科のウヅキヒメハナバチ (*Andrena benefica*) が訪花して、花粉を媒介していたことを観察した。それ以外に同属のヤヨイヒメハナバチ (*An. hebes*) とマメヒメハナバチ (*An. minutula*) も送粉昆虫であることを確認した。
- コシノコバイモの種子にエライオソームが観察されアリ散布が期待されたので、コシノコバイモの個体群内1カ所での観察と生育地ではあるが個体群外4カ所、および生育地外2カ所で、種子をアリが運ぶかどうかの実験を行った。その結果、ハヤシクロヤマアリ (*Formica hayashi*)、クロヤマアリ (*F. japonica*)、アシナガアリ (*Aphaenogaster famelica*)、トビロケアリ (*Lasius japonicus*) が種子を口にくわえて運ぶ姿を観察した。本種が属するバイモ属は、欧州、アジア、北米と北半球に広く分布し、世界に約100種存在するが、種子のアリ

散布はこの属での最初の報告である。本属の多くの種は草原性で、それらの種子は扁平で翼を持ち、風散布と考えられるのに対して、コバイモは森林性で、種子は楕円体に近く翼がなく、エライオソームを持ち、アリ散布である。本実験において捕獲した昆虫

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