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### Geographic Distribution and Habitat Differentiation in Diploid and Triploid *Lilium lancifolium* of South Korea

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Habitats and geographic distribution were investigated for diploid and triploid *L. lancifolium* grown in islands and mainland areas covering a whole country of South Korea. Of 367 populations investigated, 185 (50.4%) and 182 (49.6%), respectively, were diploid and triploid populations. Diploid was intensively distributed only along western to southeastern coastal areas and islands, whereas triploid spread dominantly in inland areas of the Korean Peninsula. Diploid was specific to such seaside habitats as coastal cliffs or beaches but never to inland habitats. Triploid tended to prefer often–disturbed inland habitats such as roadsides, arable lands, hilly fields and riversides, though 23% of triploid individuals were growing in the coastal cliffs. The facts indicate that adaptability for disturbed habitats are crucial for the distribution range of each cytotype. Nonetheless, triploids were rarely found in coastal habitats within the diploid distribution range. Autotriploid formation from diploid ancestors under natural conditions may be suppressed by rareness of higher unreduced gamete productivity within the diploid species and/or by minority cytotype exclusion.

#### INTRODUCTION

Tiger lily (*Lilium lancifolium*) had been considered to be cytologically conspicuous species because it was the only triploid in the genus (Takenaka and Nagamatsu, 1930; Noda, 1966). Natural populations of diploid *L. lancifolium* were firstly found in the Tsushima Islands, Japanese western islands near the southern coast of the Korean Peninsula, by Noda (1978). Thereafter, it has been well confirmed that *L. lancifolium* is a polyploid complex involving both diploid and triploid forms (Noda, 1978, 1986, 1991).

Several reports (Wilson, 1925; Lightly, 1968; Noda, 1986; Noda and Lee, 1980; Song, 1997) mentioned the distribution of *L. lancifolium* in Korea, but little has been known about exact geographic distribution of each ploidy form.

Noda (1986) reported that the diploid form is confined to the southern part of the Korean Peninsula including Jejudo Island of South Korea and the Tsushima Islands of Japan, whereas the triploid form is widely distributed in East Asia. However, his description of the cytotype distribution in Korea stood on the investigation only on 11 sites of South Korea. Recently, Kim *et al.* (2005) found that 13 islands of the Bay of Kyunggi, the western region of South Korea, were inhabited by diploid alone in a high population density with up to 2000 reproductive individuals. Such dense and wide

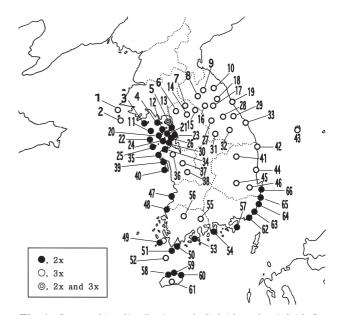
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diploid distribution has never been reported in the other places except the Tsushima Islands. The facts suggest the wider distribution of diploid *L. lancifolium* in the Korean Peninsula.

We analyzed the cytotypes of a large number of *L*. *lancifolium* individuals collected from insular and the mainland populations covering an entire country in South Korea to understand the origin of triploid forms in nature.

#### MATERIALS AND METHODS

Three hundred and eighty seven individuals of *L. lancifolium* were collected from 367 populations of 66 different localities including 23 islands throughout the



**Fig. 1.** Geographic distribution of diploid and triploid *L. lancifolium* in South Korea.

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Site No.	Locality	Habitat	No. of populations found	No. of plants observed	Ploid form
1	Incheon City, Backryungdo	Island, coastal cliff	10	10	Зx
2	Incheon City, Sochungdo	Island, coastal cliff	11	21	Зx
3	Incheon City, Daiyeonpyungdo	Island, coastal cliff	11 5	11 5	2x 2x
4	Incheon City, Suckmodo	Island, coastal cliff			
5	Incheon City, Kanghwado	Island, coastal cliff	8	8	2x
		Island, disturbed habitat	6	6	Зx
6	Kyunggido, Jucksung	Mainland, disturbed habitat	3	3	Зx
7	Kyunggido, Yeonchun	Mainland, disturbed habitat	4	4	Зx
8	Gangwondo, Chulwon	Mainland, disturbed habitat	14	14	Зx
9	Gangwondo, Sangseo	Mainland, riverside	11	11 7	Зх Зх
10	Gangwondo, Yanggu	Mainland, disturbed habitat	7		
11	Incheon City, Achado	Island, coastal cliff	19	19	2x
12	Incheon City, Yungjongdo	Island, coastal cliff	7	7	2x
13	Incheon City, Songdo	Mainland, coastal cliff	2	2	2x
14	Kyunggido, Pochun	Mainland, disturbed habitat	7	7	Зx
15	Kyunggido, Gapyung	Mainland, disturbed habitat	3	3	Зx
16	Gangwondo, Hanam	Mainland, riverside	12	12	Зx
17	Gangwondo, Chuncheon,	Mainland, riverside	9	9	Зx
	Seomyun				
		Mainland, disturbed habitat	3	3	Зx
18	Gangwondo, Chuncheon, Dongmyun	Mainland, riverside	7	7	3x
		Mainland, disturbed habitat	4	4	Зx
19	Gangwondo, Yangyang	Mainland, riverside	4	4	Зx
20	Incheon City, Eulwangdo	Island, coastal cliff	4	4	2x
21	Incheon City, Mooeuido	Island, coastal cliff + beach	3	3	2x
	• /	Island, beach	3	3	2x
22	Incheon City, Deokjuckdo	Island, coastal cliff	13	13	2x
23	Incheon City, Jawoldo	Island, coastal cliff	8	8	2x
		Island, beach	3	3	2x
24	Incheon City, Youngheungdo	Island, beach	4	4	2x
25	Incheon City, Backado	Island, coastal cliff	13	13	2x
26	Incheon City, Jangbongdo	Island, coastal cliff	13	13	2x
27	Gangwondo, Heongsung	Mainland, disturbed habitat	3	3	Зx
28	Gangwondo, Hongchun	Mainland, disturbed habitat	5	5	Зx
29	Gangwondo, Pyungchang	Mainland, disturbed habitat	7	7	Зx
30	Choongchungnamdo, Dainanjido	Island, coastal cliff	3	3	2x
31	Gangwondo, Yungwol	Mainland, riverside	2	2	Зx
32	Gangwondo, Jungseon	Mainland, riverside	2	2	Зx
33	Gangwondo, Samchuck	Mainland, coastal cliff	3	3	Зx
34	Choongchungnamdo, Taeahn, Iwon	Mainland, coastal cliff	3	3	2x
35	Choongchungnamdo, Taeahn, Wonbook	Mainland, coastal cliff	4	4	2x
36	Choongchungnamdo, Seosan, Sungyeonmyun	Mainland, disturbed habitat	4	4	Зx
37	Choongchungnamdo, Yesan, Yesanub	Mainland, disturbed habitat	2	2	3x
38	Choongchungnamdo, Gongjoo	Mainland, disturbed habitat	5	5	3x
39	Choongchungnamdo, Taeahn, Nammyun	Mainland, coastal cliff	4	4	2x
40	Choongchungnamdo, Taeahn, Anmyun	Island, coastal cliff	3	13	2x
41	Kyungsangbookdo, Andong	Mainland, riverside	8	8	3x
42	Kyungsangbookdo, Uljin	Mainland, coastal cliff	6	6	Зx
43	Kyungsangbookdo, Urleungdo	Island, coastal cliff	1	1	Зx
44	Kyungsangbookdo, Yungduck	Mainland, coastal cliff	7	7	Зx
45	Kyungsangbookdo, Yungcheon	Mainland, disturbed habitat	2	2	Зx
46	Kyungsangbookdo, Kyungjoo	Mainland, riverside	3	3	Зx
47	Jeolabookdo, Booahn	Mainland, coastal cliff	4	4	2x
48	Jeolanamdo, Youngkwang	Mainland, coastal cliff	1	1	2x
49	Jeolanamdo, Jindo	Island, coastal cliff	4	4	2x
50	Jeolanamdo, Wando	Island, coastal cliff	5	5	2x
51	Jeolanamdo, Bogildo	Island, coastal cliff	1	1	2x

Site No.	Locality	Habitat	No. of populations found	No. of plants observed	Ploidy form
52	Jeolanamdo, Choojado	Island, coastal cliff	3	5	Зx
53	Jeolanamdo, Goheung			4	2x
54	Jeolanamdo, Namhae	Mainland, coastal cliff	3	3	2x
55	Jeolanamdo, Sooncheon	Mainland, disturbed habitat	2	2	Зx
56	Jeolanamdo, Goorye	Mainland, disturbed habitat	1	1	Зx
57	Kyungsangnamdo, Geojedo	Island, coastal cliff	3	3	2x
58	Jejudo, Aewol <sup>z</sup>	Island, coastal cliff	3	3	2x
59	Jejudo, Goojwa	Island, coastal cliff	6	6	2x
60	Jejudo, Hwasoon	Island, coastal cliff	2	2	2x
61	Jejudo, RDA center	Mainland, disturbed habitat	1	1	Зx
62	Pusan, Gijanggun	Mainland, coastal cliff	3	3	2x
63	Pusan, Gijanggun	Mainland, coastal cliff	1	1	2x
64	Ulsan, Uljoogun	Mainland, coastal cliff	6	6	2x
65	Ulsan, Bookgu	Mainland, coastal cliff	4	4	2x
66	Pohang, Gampo	Mainland, coastal cliff	2	2	2x
	Total		367	387	

<sup>z</sup> Triploid was found by Song (1997), but not in this experiment.

whole country of South Korea (Fig. 1, Table 1). Number of individuals collected from each population was 1 to 21. A bulbil or bulb was sampled from each reproductive plant in summer and autumn seasons from 1998 to 2005. The populations geographically isolated at least more than 100m distance from other populations were judged as independent populations. Habitats of the populations were grouped into four categories; that is, 1) coastal cliffs, 2) coastal beaches, 3) riversides, and 4) disturbed habitats often affected by human activities including roadsides, arable fields, graveyards and grassy hills.

The ploidy level was determined by counting chromosome number of root tip cells and/or measuring relative DNA content of leaf cells with a flow cytometer (Partec PA Ploidy Analyzer, Germany). Procedures of chromosome observation and flow cytometric analysis were previously reported (Kim *et al.*, 2005).

#### RESULTS

## Geographic distribution of diploid and triploid L. lancifolium

We found 367 populations of *L. lancifolium* throughout the country of South Korea. The localities and habitat description of the populations, and the ploidy forms detected for each population are shown in Table 1. Population frequencies were almost the same between the cytotypes with 185 (50.4%) diploid and 182 (49.8%) triploid populations (Table 2).

Diploid individuals were concentrated in the western (site Nos. 3–5, 11–12, 13, 20, 21–26, 30, 40) and southern (site Nos. 49–51, 59–60) islands and along western to southeastern coasts of the mainland (site Nos. 34–35, 39, 47–48, 53–54, 57, 62–66), but never found in inland areas (Fig. 1). Of 185 diploid populations, 138 (74.6%) were found in islands and 47

(25.4%) in the mainland (Table 2).

By contrast, triploid individuals were the representative cytotype in inland areas of the mainland of South Korea (Fig. 1). Inland individuals were more easily found in northern provinces than in southern provinces. Exceptionally, only in three sites (site Nos. 33, 42, 44) of the mainland, triploid was growing in coastal populations (Table 1).

Insular populations of the triploid were rather minority; 32 (17.6%) of the 182 triploid populations were found only in six of 23 islands (Table 2). There were no diploid, but only triploid individuals in Backryungdo (site No. 1) and Sochungdo (site No. 2), the northernmost islands in the west coast, Urlengdo (site No. 43), an island in the east coast, and Choojado (site No. 52), a southern island (Table 1). Eight diploid and six triploid populations occupied Kanghwado (site No. 5), one of the nearest islands to the west coast, with close distance. One triploid population was found in the southern island, Jejudo, where diploid populations were rather dominant. Triploid plants in these islands set no capsules.

# Habitat differentiation between diploid and triploid *L. lancifolium*

Diploid individuals persisted in the coastal habitats, whereas triploid individuals tended to prefer inland habitats often affected by human activities or on the riverside (Table 1). Even in the sites where the both cytotypes were located in a relatively close distance (Site No. 5), they never grew sympatrically in the same population; i.e., diploid individuals were growing on coastal cliffs, but triploid in the roadside.

Among 185 diploid populations, 175 (94.6%) were observed on coastal cliffs (Table 2). These populations were growing in the narrow zones from 2–3m above sea level to the margin of the forest and rarely found in the

	No. of populations observed in the habitats classified below					
Ploidy forms and distribution area	Coastal cliff	Beach	Riverside	Disturbed habitat <sup>z</sup>	Total	
2x in the mainland	47	0	0	0	47	
2x in islands	128	10	0	0	138	
2x total	175	10	0	0	185	
3x in the mainland	16	0	58	76	150	
3x in islands	25	0	0	7	32	
3x total	41	0	58	83	182	

 Table 2.
 Observation frequency of diploid and triploid L. lancifolium populations for four habitat types in South Korea

<sup>z</sup> Habitats often affected by human activities including roadsides, arable fields, graveyards and grassy hills.

forest. In islands, 10 (5.4%) were growing on the beach.

Among 182 triploid populations, 83 (45.6%) were growing in such ruderal habitats as arable lands, roadsides, graveyards and hilly zones. Fifty-eight (31.9%) populations were found on the riverside in inland areas. The remaining 41 (22.5%) were growing in the habitats similar to those inhabited by diploid forms, namely, coastal cliffs.

#### DISCUSSION

Before the present study, the most detailed information on cytotype distribution of L. lancifolium has been recorded in Japan, where diploid forms were found only in the Tsushima Islands and Iki Island, the islands between Kyushu Island in Japan and Pusan in South Korea, but never in the other areas (Noda, 1978, 1986; Hiramatsu, unpublished data). Jugging from the previous and the present study in which no diploid was found in the whole inland areas, the east coastal areas, and the northernmost islands in the west coast, it is highly probable that geographic distribution of diploid L. lancifolium is restricted in western and southern coastal areas of the Korean Peninsula. The description of geographic distribution for diploid L. lancifolium in the present study is more precise than that previously reported (Noda, 1986; Kim et al., 2005).

Opposite to the limited distribution of diploid *L. lancifolium*, its triploid seems to maintain considerable wide distribution in Eastern Asia including the Japan Islands, the Korean Peninsula, the north–east district of China as shown in the present and previous studies by Noda (1986). A more widespread distribution of the polyploid than that of the ancestral diploids is a well–known, typical output phenomenon of success in plant polyploidy (Stebbins, 1950). Cytotype distribution data shown in the present study imply that triploid is adaptive to inland habitats often disturbed by human activities or floods, but diploid is not. Such ability specific to triploid could explain its widespread distribution.

Since no tetraploid *Lilium* species has been found under natural conditions so far, only diploid that can produce functional unreduced gametes could be the resource of triploid, and therefore, triploid individuals of early generations are expected to grow within or near their diploid progenitors. Only in three areas (Kanghwado, Choojado and Jejudo Island), however, triploid forms were parapatrically growing within the diploid distribution range.

Though it is difficult to explain why coexistence of the both cytotypes has not been detected for the moment, we can only propose two possible reasons. First, 'minority cytotype exclusion', a model of frequency-dependent selection against the newly formed rarer cytotype when two populations with different ploidy levels grow in sympatry (Levin, 1975), may work critically in populations densely dominated by diploid individuals. This hypothesis is consistent with the fact that triploids are growing dominantly on coastal cliffs where diploids are absent, as we confirmed in the several sites. Second, since variations in rate to develop functional unreduced gametes have been observed in many nonhybrid crop species (Bretagnolle and Thompson, 1995) and in interspecific hybrids of *Lilium* (Lim et al., 2001), frequency of autotriploid emergence may be significantly different among different diploid lineages of L. *lancifolium*. In order to answer which factor is attributable to the emergence of autotriploid in natural conditions, more precise investigations if both cytotypes grow in more close distance, if there is a close genetic relationship between such sympatric and parapatric cytotypes, and if production frequency of functional unreduced gametes is genotype-dependent are required.

Noda (1986) hypothesized from his cytological studies that the triploid *L. lancifolium* may be the allotriploid produced by 'interspecific' hybridization between a 'diploid' *L. lancifolium* and the taxonomically close 'diploid' species, *L. maximowiczii*. However, the possibility for allotriploids originated from the interspecific hybridization between these diploid species may be hardly accepted yet, because *L. maximowiczii* has never been found sympatrically and parapatrically with the 'diploid' *L. lancifolium* in previous (Noda, 1986) and our investigations (unpublished data).

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