

# Variation in the Leaf Number of *Arisaema iyoanum* subsp. *nakaianum* and *A. ovale* var. *ovale* (Araceae)

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## ABSTRACT

*Arisaema iyoanum* Makino subsp. *nakaianum* (Ohba) H. Ohashi et J. Murata and *A. ovale* Nakai var. *ovale* are known to have one-leaved phenotype in both males and females; however, we discovered two-leaved individuals of these species. To elucidate the relationship between growth stage and leaf number of *A. iyoanum* subsp. *nakaianum* and *A. ovale* var. *ovale*, we conducted a morphological analysis of these plants. Our analysis suggested that the two-leaved individuals of *A. iyoanum* subsp. *nakaianum* and *A. ovale* var. *ovale* appeared only at the female phase. This suggested that one-leaved *A. iyoanum* subsp. *nakaianum* and *A. ovale* var. *ovale* individuals could not store the resources and hence changed to two-leaved individuals. This transformation could be explained by the fact that these species occur at high altitudes in mountain areas or regions covered in snow of the Japan Sea side, and their flowering time is also late compared with that in other sympatric *Arisaema* species.

**Keywords:** *Arisaema iyoanum* subsp. *nakaianum*; *A. ovale* var. *ovale*; Leaf Number

## 1. Introduction

Araceae is a large family of mostly herbaceous plants, with a great variety in vegetative habit. This family mainly includes herbaceous plants with aerial stems or underground tubers or rhizomes, as well as a few woody members. *Arisaema* is a genus of the Araceae family that includes about 150 species that are found in eastern and central Africa, Asia, and eastern North America [1]. *Arisaema* includes perennial plants that are typically male when young and female or hermaphroditic when fully grown. The sex of a plant changes depending on resources conditions, and a single plant can change its sex several times during its life-span (e.g., [1-3]). In general, most species in the section *Pedatisecta* in *Arisaema* have constant numbers of leaves either one or two. The section *Pedatisecta* is the spring ephemeral, which flower in early spring and store the resources for a limited period [3]. Among them, however, *Arisaema tosaense* Makino has characteristic leaf numbers, with both one- and two-leaved plants. Muramatsu *et al.* [4] reported that one-leaved individuals of *A. tosaense* appeared at the beginning of both male and female phases, suggesting that they contribute to the earlier

appearance of the fertile phase from sterile phase. Considering the result of the leaf number of *A. tosaense*, *Arisaema* species described as one-leaved may have the potential to become two-leaved during its life-span.

*Arisaema iyoanum* Makino subsp. *nakaianum* (Ohba) H. Ohashi et J. Murata and *A. ovale* Nakai var. *ovale* are mainly one-leaved [1]. The former is distributed in the high altitudes of the mountain areas of Shikoku, and the latter occurs in the Japan Sea side of the limited mountain areas of Honshu, Japan [3,5,6], where it remains covered with snow during winter to spring. Interestingly, we found two-leaved individuals of *A. iyoanum* subsp. *Nakaianum* and *A. ovale* var. *ovale* (**Figure 1**) in different areas of Japan. Hence, we attempted to elucidate whether the number of leaves varies from one to two leaves during the male to female transition of these plants. Nonetheless, determining the growth stage of *Arisaema* species is difficult. Bierzychudek [2] indicated that the leaf area of *Arisaema* species was correlated to their growth stage. In addition, Takasu [7] suggested high correlations of the leaf area with the leaflet number, biomass, and pseudostem diameter in *Arisaema* species. Therefore, we thought that the pseudostem diameter in field could reflect the growth stage of *Arisaema* species. The relationship between leaf number and growth stage

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**Figure 1.** A two-leaved individual of *Arisaema ovale* var. *ovale*. Arrows indicate leaves.

(sexual stage) of *A. iyoanum* subsp. *nakaianum* and *A. ovale* var. *ovale* was determined by conducting a morphological analysis of these species during their growth stage (pseudostem diameter measurement).

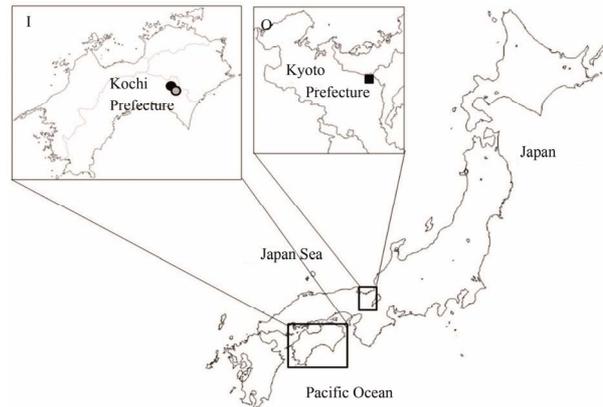
## 2. Materials and Methods

In this study, *A. iyoanum* subsp. *nakaianum* were analysed between April and June 2012 and *A. ovale* var. *ovale* were analysed in May 2011. In all, 120 sterile plants (54 *A. iyoanum* subsp. *nakaianum*; 66 *A. ovale* var. *ovale*), 108 males (54 *A. iyoanum* subsp. *nakaianum*; 54 *A. ovale* var. *ovale*), and 63 females (31 *A. iyoanum* subsp. *nakaianum*; 32 *A. ovale* var. *ovale*) plants were marked using numbered and staked wire flags in the Kyoto Prefecture (*A. ovale* var. *ovale*) and Kochi Prefecture (*A. iyoanum* subsp. *nakaianum*) (Table 1, Figure 2). The pseudostem diameter of the plants was measured using a digital calliper (Mitutoyo), and the sexual state of each plant was recorded during the growing season.

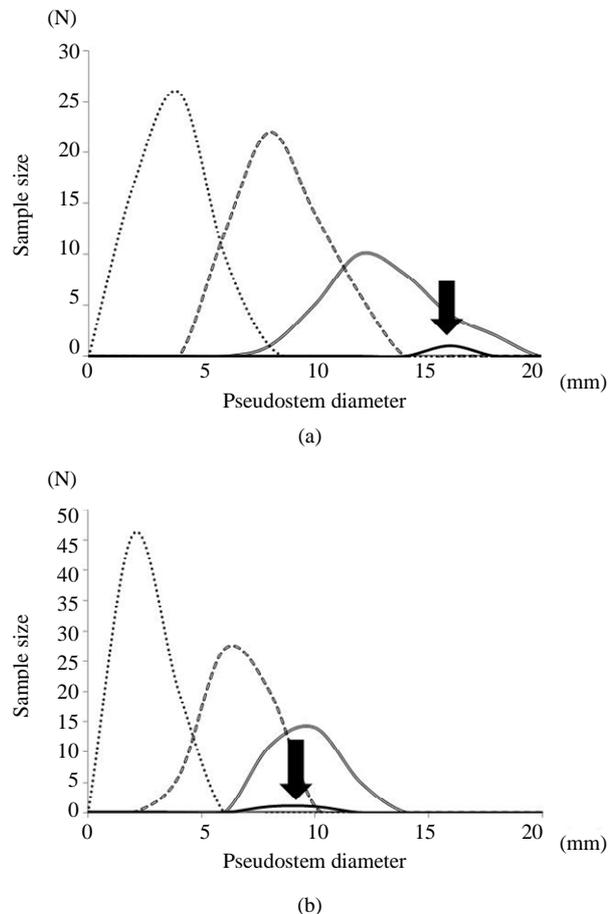
## 3. Results and Discussion

Our results indicated that the pseudostem diameters of sterile, male, and female individuals of *A. iyoanum* subsp. *nakaianum* ranged from 0.64 to 6.88, 4.2 to 11.71, and 6.09 to 17.63 mm (Figure 3(a), Table 2), and those of *A. ovale* var. *ovale* ranged from 0.29 to 3.86, 3.66 to 8.37, and 6.1 to 11.56 mm (Figure 3(b)), respectively. The pseudostem diameter of *A. iyoanum* subsp. *nakaianum* with 2 leaves was 14.48 mm and those of *A. ovale* var. *ovale* were 7.88 and 8.24 mm during the female phase.

These results suggested that the generally one-leaved individuals of *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* became two-leaved at the female phase; however, the rate of appearance of two-leaved individuals was very low (*A. ovale* var. *ovale*: 1.32%; *A. iyoanum* subsp. *nakaianum*: 0.72%). The reason for the appear-



**Figure 2.** Sampling localities of this study. I: *Arisaema iyoanum* subsp. *nakaianum*; O: *A. ovale* var. *ovale*. Black circle indicates Kochi Pref., Kami city, Monobe-cho Kubokage; Gray circle indicates Kochi Pref., Kami city, Monobe-cho befu; Black square indicates Kyoto Pref., Nantan city, Miyama-cho, Ashiu.



**Figure 3.** Relationship between growth stage and pseudostem diameter. Dotted line indicates the sterile phase, white short dashed line is the male phase with one-leaf, gray line is the female phase with one-leaf, and black line is the female phase with 2 leaves. Arrows indicate the female phase with 2 leaves. (a) *Arisaema iyoanum* subsp. *Nakaianum*; (b) *A. ovale* var. *ovale*.

**Table 1. Sampling localities.**

Species	Locality	Latitude	Longitude	Sample Size		
				Sterile	Male	Female
<i>Arisaema iyoanum</i> subsp. <i>nakaianum</i>	Kochi					
	Prefecture, Kami City	33°48'	133°57'	38	18	5
	Monobe-Cho					
	Kubokage					
<i>A. ovale</i> var. <i>ovale</i>	Kochi					
	Prefecture, Kami City	33°47'	134°01'	16	36	26
	Monobe-Cho					
	Befu					
<i>A. ovale</i> var. <i>ovale</i>	Kyoto					
	Prefecture, Nantan City	35°19'	135°44'	66	54	32
	Miyama-Cho					
	Ashiu					

**Table 2. Pseudostem diameter (mm) of sterile, male, female in *Arisaema iyoanum* subsp. *nakaianum* and *A. ovale* var. *ovale*.**

Species	Sterile	Male	Female
	Mean ± S.D.	Mean ± S.D.	Mean ± S.D.
<i>Arisaema iyoanum</i> subsp. <i>nakaianum</i>	2.88 ± 1.38	7.56 ± 1.87	12.13 ± 2.59
<i>A. ovale</i> var. <i>ovale</i>	1.76 ± 0.72	5.63 ± 1.15	8.55 ± 1.45

ance of two leaves in *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* during the female phase was not clear. One of the reasons could be that the resources to produce an additional leaf could not be stored, although the single leaf of *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* is very large and can store many resources. *A. ovale* var. *ovale* is distributed in the Japan Sea side of Honshu, and the climate of this region differs from that of the Pacific Oceanic side because Japan has a lofty backbone range that extends along the axis of Honshu and divides the island into the Japan Sea side and the Pacific Oceanic side. The Japan Sea side receives considerable snow and has late spring. On the other hand, *A. iyoanum* subsp. *nakaianum* occurs in the high altitude of the mountain areas of Shikoku, and its flowering time

is late compared with that of other sympatric *Arisaema* species [3,8]. From similar environmental conditions of flowering time in different areas, *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* could not be easily to store in the resources of appearing two leaves. The resources of *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* could not be stored efficiently in the male phase, and there was no need to produce an additional leaf; however, during the shift from male to female phase, the stored resources in the female phase were used to not only produce seeds and fruits but also form an additional leaf. Thus, most *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* species are one-leaved during the female phase, and appearance of two-leaved individuals at this phase is very rare. In addition, our results indicated that the rate of appearance of two-leaved individuals of *A. ovale* var. *ovale* and *A. iyoanum* subsp. *nakaianum* was lower than that of *A. tosaense* ([4]: 58.45%), which has both one- and two-leaved individuals. *A. tosaense* plants occur at lower or the same altitude as that of *A. iyoanum* subsp. *nakaianum*, but the former occur at lower altitude than the latter; hence, *A. tosaense* can easily store resources compared to *A. iyoanum* subsp. *nakaianum*. In a short spring season with environmental conditions such as high altitude and considerable snow, one-leaved *Arisaema* species could have better fitness than the two-leaved individuals.

Recent studies have reported interspecific hybrids among *Arisaema* species on the basis of morphological and molecular analyses. For example, Maki and Murata [9] indicated that *A. ehimense* J. Murata et Ohno is a hybrid origin between *A. serratum* (Thunb.) Schott and *A. tosaense*. Moreover, Hayakawa *et al.* [10-12] reported the formation of hybrids and introgressions between *A. sikokianum* and *A. tosaense* and between *A. sikokianum* and *A. japonicum* Blume (recognized as *A. serratum* in [12]). These results suggested that interspecific hybrids could be easily generated using *Arisaema* species. It would be interesting to determine the leaf number of interspecific hybrids between *A. ovale* var. *ovale* or *A. iyoanum* subsp. *nakaianum* and two-leaved *Arisaema* species.

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