

Differences in Leaf Morphology among Three Native Dandelion Species in the Central Part of Honshu, Japan

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本州中部における在来タンポポ3種の葉の形態的差異

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要旨: フィールド調査において、タンポポ在来種を見分ける花以外の基準を得ることを目的として葉の形態を調査した。2010 から 2014 年春季に、本州中央部において、シナノタンポポ (*Taraxacum platycarpum* ssp. *hondoense*) 8 地点, トウカイタンポポ (*T. platycarpum* ssp. *platycarpum*) 8 地点, セイタカタンポポ (*T. elatum*) 4 地点およびカンサイタンポポ (*T. japonicum*) 4 地点の葉を採集し、画像解析によって葉のサイズを計測して葉形を解析した。各形質の種間差を比較するとともに、要因 (種、地点) の影響を分散分析によって判定した。その結果、葉のサイズ, 葉形 (細長度および複雑度) はいずれも種および地点による影響が有意であった。しかし地点間のばらつきが大きく、種間差は葉のサイズのみで検出され、トウカイタンポポとセイタカタンポポ > シナノタンポポとカンサイタンポポであった。葉形について、トウカイタンポポの変異が大きいため 4 者それぞれのグループを明確に分けることはできず、シナノタンポポとカンサイタンポポのグループが分離していたのみであった。よって、タンポポ在来種を葉の形態のみで分類することは困難で、適応放散のあまり進んでいない種群であることが確かめられた。

キーワード: タンポポ属, 在来種, 葉, 分類

Key words: *Taraxacum*, Native species, Leaf shape, Taxonomy

Introduction

To distinguish between native and exotic dandelion species, the most informative and stable taxonomic character is the morphology of the outer involucre bracts of the capitulum (e.g., Ohwi, 1992; Shimizu, 1997). However, in the Koshin region of central Japan, leaf morphology has proven to be

sufficiently robust and taxonomically informative: the leaf shape of the native species *Taraxacum platycarpum* ssp. *hondoense* generally tends to be more slender and simpler than that of the exotic species, *Taraxacum officinale* (Arase *et al.*, 2013; Arase *et al.*, 2014).

In Japan, many native dandelion species exist in addition to *T. platycarpum* ssp. *hondoense*. Nearly



Figure 1 Geographic location of investigation area and the sampling sites of native *Taraxacum* species.

2,000 agamospecies are in the genus *Taraxacum* around the world, and the Japanese archipelago is one of the five areas where they are distributed with unusually high concentrations (Richards, 1972). The most informative and stable taxonomic character to distinguish native dandelion species is also the morphology of the outer involucre bracts of the capitulum (e.g., Ohwi, 1992; Shimizu, 1997), which requires even more careful observation than needed to distinguish native and exotic species. In addition, native dandelion species have similar life history characteristics (Sawada *et al.*, 1982). Fortunately, the geographic distribution of each native dandelion species is so different that the species can be guessed with a high degree of reliability. However, it is difficult to identify them in an area where two or more distributions of native dandelion species overlap. It is even more difficult during periods when the plants lack inflorescences.

We therefore focused on dandelion leaf morphology, as dandelion leaves can be observed throughout the year and are thus well suited for identification purposes. However, few studies on dandelion leaf morphology have been published to

date, and the influence of environmental factors on leaf shape is unclear (Denawa *et al.*, 1979; Arase *et al.*, 2013). Furthermore, information on the differences in leaf shape between species is limited to descriptions in several illustrated plant guidebooks (e.g., Ohwi, 1992; Shimizu, 1997).

In order to collect data and confirm the validity of leaf morphology as a new taxonomic character for identifying native dandelion species lacking inflorescences, this study examined the size and shape of native dandelion leaves harvested in the field. We expanded the research area from the Koshin district (Arase *et al.*, 2014) to the central part of Honshu, where three native dandelion species are distributed.

Methods

We surveyed areas in the central part of Honshu in Japan (Figure 1), where the native dandelions of *T. platycarpum* (ssp. *hondoense* and ssp. *platycarpum*), *T. elatum* and *T. japonicum* (Figure 2) are common (Ohwi, 1992; Shimizu, 1997). The capitulum morphology of *T. elatum* (Figure 2b) is described as intermediate between those of *T.*

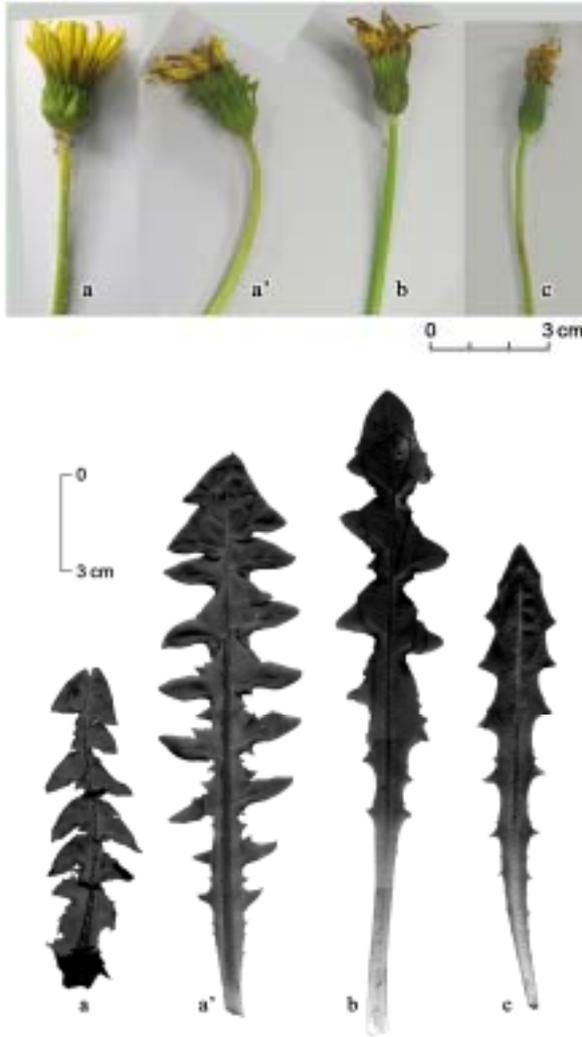


Figure 2 Capitula and leaves of native *Taraxacum* species in the investigation area. a: *T. platycarpum* ssp. *hondoense* (site No. 8), a': *T. platycarpum* ssp. *platycarpum* (site No. 9), b: *T. elatum* (site No. 18) and c: *T. japonicum* (site No. 24).

platycarpum ssp. *hondoense* (Figure 2a) and *T. japonicum* (Figure 2c) (Shimizu, 1997).

In the spring of 2010, 2011, 2012, 2013 and 2014, 24 sites where native dandelion species grew abundantly were surveyed (Figure 1 and Table 1). Two leaves were collected per individual and four individuals were collected per site (24 sites × 4 individuals × 2 leaves = 192 samples in total). Images of individual leaves were captured with a scanner at a resolution of 400 dpi (i.e., 0.0635 mm per dot) and the traits of leaf length (L), leaf width (W), leaf margin circumference (C) and leaf area

Table 1 Sampling sites of native *Taraxacum* species

Site No.	Latitude °	Elevation (m)	Habitat
<i>T. platycarpum</i> ssp. <i>hondoense</i>			
1 Kijima-daira	36 49	10 580	Bank around a shrine
2 Hakuba	36 42	55 710	Levee of paddy fields
3 Komoro	36 19	46 560	Levee of paddy fields
4 Azumino	36 18	33 580	Levee of paddy fields
5 Hokuto	35 49	49 700	Roadside
6 Minami-minowa	35 51	49 760	Forest road
7 Minobu	35 28	54 235	Forest road
8 Shimojo	35 23	18 590	Levee of paddy fields
<i>T. platycarpum</i> ssp. <i>platycarpum</i>			
9 Kawadzu	34 47	16 160	Roadside
10 Fujino-miya	35 11	56 125	Roadside
11 Omaezaki	34 40	44 120	Levee of tea fields
12 Hamamatsu	34 51	32 50	Roadside
13 Shinshiro	34 59	13 140	Orchard
14 Seto	35 13	26 180	Roadside
15 Minami-chita	34 44	38 7	Roadside
16 Higashi-shirakawa	35 40	4 405	Roadside
<i>T. elatum</i>			
17 Echizen	35 59	56 70	Orchard
18 Gujo	35 55	44 450	Roadside
19 Motosu	35 31	22 80	Forest road
20 Takashima	35 30	54 380	Roadside
<i>T. japonicum</i>			
21 Seki	35 27	25 83	Park
22 Kuwana	35 8	24 125	Forest road
23 Ohmi-hachiman	35 9	10 90	Park
24 Iga	34 50	38 230	Unused land

(A) per leaf were measured with Moti Images Plus 2.0S image-processing software (Speed Fair Co. Ltd., Hong Kong, China).

To further characterize leaf shape, we employed indices for slenderness and intricateness (Arase *et al.*, 2013), which can be expressed as follows:

$$\text{Slenderness} = L / W \quad (1)$$

$$\text{Intricateness} = C^2 / A \quad (2)$$

where the minimum value of (2) is 4π if the shape of the leaf approximates a perfect circle.

Results

From the 24 sites in the research area, we collected *T. platycarpum* ssp. *hondoense* at 8 sites (inland area of the Koshin district), *T. platycarpum* ssp. *platycarpum* at 8 sites (mainly the Pacific coast), *T. elatum* at 4 sites (the coast between Biwa Lake and the Japan Sea), and *T. japonicum* at 4 sites (mainly the southern coast of Biwa Lake) (Figure 1 and Table 1).

The leaf sizes of native dandelion species are shown in Figure 3. Analysis of variance showed that species and sites were both significantly

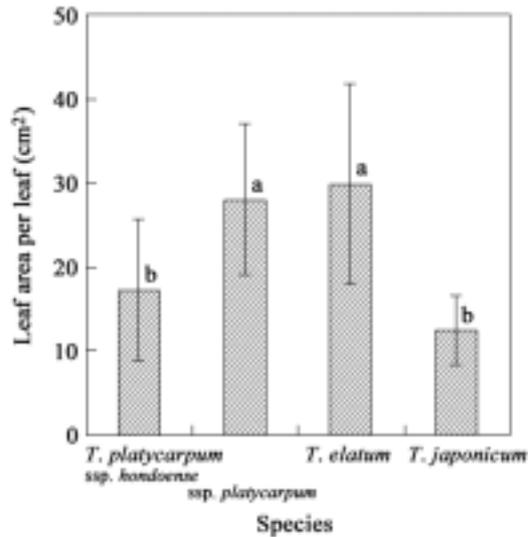


Figure 3 Mean leaf sizes of native *Taraxacum* species. Each bar means \pm standard deviation among sites ($n=8$ in each 2 subspecies of *T. platycarpum*, $n=4$ in *T. elatum* and *T. japonicum*). Different letters in the figure denote significant difference determined by Tukey's HSD ($p < 0.05$).

related (F-test, $p < 0.001$) to all traits (leaf area (A), slenderness (L/W) and intricateness (C^2/A)). Leaf area (A) was significantly larger in *T. platycarpum* ssp. *platycarpum* and *T. elatum* (27.9 and 29.7 cm^2) than in *T. platycarpum* ssp. *hondoense* and *T. japonicum* (17.2 and 12.4 cm^2) (Tukey's honestly significant difference (HSD) test, $p < 0.05$; Figure 3). However, no significant differences among species were detected in slenderness ($L/W = 4.4$ to 5.6) or intricateness ($C^2/A = 114.7$ to 171.1) because of the large deviation among sites.

Figure 4 is a scatter plot of average leaf shape (i.e., slenderness vs. intricateness) for each species at the different sites examined in this study. *T. platycarpum* ssp. *hondoense*, *T. platycarpum* ssp. *platycarpum*, *T. elatum* and *T. japonicum* were fairly intermingled. In particular, *T. platycarpum* ssp. *platycarpum* formed a wide-ranging group, overlapping with other species. Figure 4 demonstrates the distinction between the *T. platycarpum* ssp. *hondoense* and the *T. japonicum* groups: the leaf shape of *T. japonicum* is slenderer and simpler.

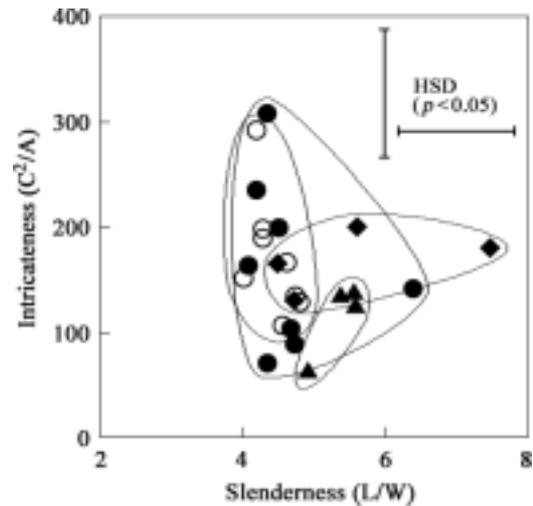


Figure 4 Variations in leaf shape of native *Taraxacum* species among sites. Each symbol of \circ , \bullet , \blacksquare and \blacktriangle denotes *T. platycarpum* ssp. *hondoense*, *T. platycarpum* ssp. *platycarpum*, *T. elatum* and *T. japonicum*, respectively.

Discussion

Our findings showed that leaf size appeared to be species-specific (Figure 3). However, leaf size is a quantitative trait that may change according to growth environment (for example, richness of the soil, intensity of light, temperature). In Figure 3, a significant difference among species was detected between *T. elatum* and *T. japonicum* only; differences between the two subspecies of *T. platycarpum* could be attributable not only to the difference in taxonomic groups, but also to differences in the geographic environment. Thus, leaf size does not seem reliable enough for distinguishing native dandelion species.

The leaf shape parameters of slenderness and intricateness also could not be used to distinguish species (Figure 4). *T. platycarpum* ssp. *hondoense*, *T. platycarpum* ssp. *platycarpum*, *T. elatum* and *T. japonicum* were intermingled. Although the *T. platycarpum* ssp. *hondoense* and *T. japonicum* groups seemed distinguishable, the observed differences were not sufficiently reliable because *T. japonicum* was examined at only 4 sites. The

influence of environmental factors on leaf shape is reportedly unclear (Denawa *et al.*, 1979; Arase *et al.*, 2013; Arase *et al.*, 2014), and our results also do not elucidate the reasons for the variation observed in the leaf shape of dandelion leaves.

The morphology of the capitulum of *T. elatum* is described as intermediate between *T. platycarpum* ssp. *hondoense* and *T. japonicum* (Shimizu, 1997). However, the leaf size of *T. elatum* was significantly larger than both *T. platycarpum* ssp. *hondoense* and *T. japonicum* (Figure 3), and the leaf shape of *T. platycarpum* ssp. *hondoense* and *T. japonicum*, especially its slenderness, did not vary between the two species (Figure 4). Therefore, these aspects of leaf morphology do not seem to adequately characterize or allow distinguishing of native dandelion species, and they do not seem to correlate with capitulum morphology. This supports the notion that the genus *Taraxacum* exhibits relatively little adaptive radiation except for its capitulum morphology (Richards, 1972).

Consequently, additional surveys of leaf shape need to be conducted in order to more accurately evaluate the robustness of leaf shape for its use in taxonomic studies. The potential application of other leaf shape characteristics that have not yet been investigated also needs to be examined.

Conclusions

We examined leaf morphology traits of native species of dandelions from the central part of Honshu, Japan. We surveyed 24 sites where native dandelion species were abundant in the spring of 2010, 2011, 2012, 2013 and 2014. Scans of the leaf samples were analyzed and morphometric differences among the species were compared statistically.

Analysis of variance showed that the effects of species and site were significant for leaf size and leaf shape (slenderness and intricateness). However, a significant difference among species was detected only for leaf size (leaves of *T. platycarpum* ssp. *platycarpum* and *T. elatum* were larger than those of *T. platycarpum* ssp. *hondoense*

and *T. japonicum*), possibly because of the large variance among sites. Thus, leaf shape was not a reliable way of distinguishing among species, and it did not appear to correlate with the capitulum morphology.

These results suggest that leaf morphology may not be sufficient to distinguish between native dandelion species, confirming that the genus *Taraxacum* exhibits relatively little adaptive radiation.

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