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ORIGINAL ARTICLE

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Shoot morphology of *Aucuba japonica* incurred by anisophylly: ecological implications

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Abstract Anisophylly, having leaves different in size and/or shape, was quantified in adult *Aucuba japonica* and simulations were carried out to evaluate the effects of anisophylly on the extent of self-shading at the single-shoot level as well as at the whole-canopy level. Clear anisophylly was observed in the individual after switching from the single-stemmed juvenile stage to the multi-stemmed adult stage. In such plants, leaf area in the canopy abruptly increased. The effective display of adult foliage involved a variety of morphological changes in addition to anisophylly, most prominently reduction in leaf size compared to juveniles. The simulation results indicate that diversity of leaf size and shape is an effective means of minimizing self-shading as well as allowing the efficient exploitation of a larger canopy volume in adult plants. Anisophylly also increased the biomass use efficiency of individual plants at maturity. Taken together, having diverse leaf forms is superior to having a single leaf form for maximizing area acquisition and for efficiently filling the acquired area. We therefore conclude that the anisophylly expressed in *A. japonica* is adaptive.

Key words *Aucuba japonica* · Anisophylly · Dorsiventrality · Self-shading · Leaf diversity

Introduction

Plants show morphological adjustments at both the shoot and the whole-crown levels to intercept light efficiently (Fisher 1978; Chazdon 1985; Niklas 1992; Kikuzawa et al. 1996; Yamada and Suzuki 1996). The adjustment of shoot morphology is critically important for avoiding self-shading (Osada et al. 2002; Takenaka 1994) because inefficient light capture lowers photosynthetic production (Takenaka 1994; Yamada et al. 2000) and survivorship (Yamada et al. 2000). Therefore, the ability to adjust shoot morphology to reduce self-shading should be under selection pressure (Takenaka 1994). A variety of features like leaf size and shape, internodal length, petiole length (Niklas 1988), leaf lamina narrowness (Takenaka 1994), dynamic adjustment of leaf deployment, pattern of leaf growth (Yamada et al. 2000) and leaf orientation (Niklas 1988; Yamada et al. 2000), all contribute to efficient display of photosynthetic surfaces to avoid self-shading (Valladares et al. 2000). Because of the number of interacting traits that can contribute to minimizing self-shading, various shoot morphologies with very different phyllotactic patterns and crown architecture are capable of efficient light capturing (Valladares et al. 2002), compensating even for phylogenetically determined phyllotaxis that may be somewhat sub-optimal in a given environment (Niklas 1988).

The leaf is the principal photosynthetic organ, and as such, its effective function in light capture and photosynthesis is critical to plant survival (Kessler et al. 2001). In the light-deficient understorey, a low degree of self-shading could have a large impact on net photosynthesis near the compensation-point light level (Givnish 1984). Minimum leaf overlap in the horizontal plane increases the mean light capture per unit leaf area in shaded habitats (Pearcy and Yang 1998; Pearcy and Valladares 1999); and in small gaps where stems are present in extremely high density (Brokaw 1985), light coming from angles nearer to the horizon is mostly intercepted and plants depend on light mostly coming from angles nearer to the vertical (Yamada et al. 2000). Thus, effective acquisition and efficient use of light can be critical in the forest understorey where light is very variable and also strongly limiting to plant life (Muraoka et al. 2003). This is particularly important for seedlings in the understorey where light scarcity causes growth suppression (Canham 1985). Because of their smaller size and limited tolerance, seedlings are more vulnerable to shade stress.

Leaves are the key components of the shoot system (Tsukaya 1998) upon which the functional shoot

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morphology largely depends. For effective tailoring of leaf area to maximize light capture, having a single leaf form (shape and size) or having only smaller leaves (Osada et al. 2002) within a crown might not offer sufficient flexibility to compensate for phyllotactic constraints. In these instances, anisophylly, the formation of leaves differing in shape and/or size (Ali and Kikuzawa 2005), may be advantageous. Orientation of anisophyllous leaves would have significant impact on light interception (Goebel 1900) as well as on capture of canopy space by individual plants. Anisophylly is associated with a range of phyllotactic patterns (Dengler 1999) and is viewed as an expression of adjustment of leaf area to light interception (Morley 1973). Different species expressed anisophylly to varying degrees in all or part of the shoot system (Sánchez-Burgos and Dengler 1988). Thus, from the view of optimal adjustment of leaf area to maximize light interception, different degrees of anisophylly are acceptable (Morley 1973), depending on, for example, plant structure and shoot orientation.

Anisophylly occurs exclusively upon plagiotropic shoots (Goebel 1900) and from the earliest documentation of anisophylly, it has been thought to be an adaptation to dimly lit understory environments (Goebel 1900; Givnish 1984), yet there is a dearth of ecological studies dealing with anisophylly (Dengler 1999). In this paper, we use *Aucuba japonica* (Cornaceae), an understory shrub, to explore the ecological implications of anisophylly in the forest understory. Over time, *A. japonica* could suffer from severe self-shading due to sudden increase in leaf deployment in the tree canopy (Takenaka et al. 1998), for example, when a canopy gap closes in which the seedling *A. japonica* is established. Mature *A. japonica* shrubs express moderate anisophylly (Ali and Kikuzawa 2005) that could be a rescue mechanism for reducing self-shading (Goebel 1900; Dengler 1999; Morley 1973).

We address the following questions with regard to *Aucuba japonica*: (1) Does anisophylly reduce self-shading? (2) Does anisophylly enhance area acquisition and economic filling of acquired area? To answer these questions, we estimated self-shading and area occupation by individual canopy through simulations at a single-shoot level (considering different shapes of leaf) as well as at the whole-canopy level (considering anisophylly and isophylly).

Material and methods

To evaluate the ecological implications of anisophylly, we selected juvenile (yet to flower) and adult (flowering) *Aucuba japonica* Thunb. plants growing in the Botanical Garden, Faculty of Science, Kyoto University, situated at 35°02′N and 135°47′E and 60 m above sea level. The site is on the Kitashirakawa alluvial fan with sandy soil. Annual mean temperature was 16.1°C, ranging from a maximum of 36.0°C in July to a minimum of −5.0°C in January. Mean annual precipitation was 1,539 mm.

The genus *A. japonica* is distributed from the Himalayan region through south China and Taiwan to the Japanese archipelago; *Aucuba japonica* is distributed at the easternmost edge of the range (Kanai 1963 and Hara 1966 as cited in Ohi et al. 2003). *A. japonica* is an evergreen, dioecious, woody understory shrub species widely distributed from warm temperate to cool temperate forests in Japan. The minimum age of flowering is 4 years but reproductive maturity can be delayed for as many as 15 years (Kume and Ino 2000). *A. japonica* produces two types of terminal buds, vegetative and reproductive, that commonly have two pairs of bud scales. Phyllotaxis is almost always decussate from vegetative appendages, including bud scales, to the reproductive ones (Hara 1980). Juvenile plants extend a single new shoot every year (Kume and Ino 2000) from the terminal vegetative bud until reproductive maturity is attained. Once a plant is mature (flowering), successive new shoots generally keep flowering each season, and multiple shoots extend from the shoot apex. Adult plants produce terminal reproductive and/or terminal vegetative buds. Axillary buds in the axils of bud scales of the terminal reproductive bud develop into new shoots and a terminal reproductive bud has the potential to yield up to four shoots (Hara 1980). In adult plants, shoots are produced through syllepsis (Tomlinson 1978). *A. japonica* expresses moderate anisophylly on every other node of the adult shoot system. The degree of anisophylly varies depending on nodal position on the shoot system, and on crown light regime (Ali and Kikuzawa 2005). Depending on light conditions, shoot orientation ranged from nearly horizontal to almost orthotropic.

Geometric measurements

We selected naturally growing juvenile plants ($n = 14$) of 25–70 cm in height and adult plants (flowered for the first time) ($n = 40$) of 38–122 cm, all within the age range 5–7 years to measure the following parameters: (1) laminar length and maximum width of all leaves, (2) petiole length of all leaves, (3) distance of maximum width from petiole base (the attaching point to the stem) of all leaves, (4) internode (IN) lengths and stem diameters, (5) distance between daughter shoot tips, and (6) crown diameter in two directions perpendicular to each other for all the individual plants. Further, along with current-year growth, we measured annual growth characteristics in the previous years (identified by bud scale scars), IN length, and the number of leaves (by leaf scars) produced in a previous year.

Measurement of area of mature leaves

We estimated individual leaf area by the equation:

$Y = 0.682 \times X$ ($r = 0.999$),

where X equals lamina length × width. This relationship was established between leaf area (measured by scanning) and lamina dimensions (lamina length × width) of fresh sample leaves ($n = 81$). Lamina length was measured as the distance from the attaching point of the petiole to the tip of the lamina. Lamina width was measured as the distance between the lamina margins at the widest point.

Measurement of the degree of anisophylly

To express the degree of anisophylly, we used leaf area ratio, the ratio of the leaf areas of the two leaves opposite to each other on the same node. For the even-numbered nodes (e.g., N_2, N_4) on an adult shoot, we used the ratio of outer to inner leaf area. For odd-numbered nodes (e.g., N_1, N_3 where leaves grown tangential to the inflorescence) and shoots emerging from a terminal vegetative bud on adult plants and juveniles, we selected the numerator and the denominator of the ratio for each nodal position randomly because of difficulty in distinguishing the inner leaf versus outer leaf.

For adult plants, the position in between the sides of inner leaves of the shoots was designated as the inner canopy and that between the sides of the outer leaves of the shoots as outer canopy (Fig. 1a); this distinction was impossible for juvenile plants. The smaller leaf of an anisophyllous pair is always oriented to the inner canopy position and that of the larger leaf to the outer canopy.

Leaf-lamina narrowness

We defined leaf lamina narrowness as the ratio of laminar length to the maximum width for all leaves; a larger value indicates a narrower lamina.

Leaf-shape index

We defined leaf-shape index (LSI) as the ratio of the distance from the petiole base on the stem to the point on the midrib where the lamina width was the maximum to the total distance from the petiole base to the lamina tip (Fig. 1b). For a given size of a leaf, if the maximum width occured at distances of 0, 20, 40, 60, 80 and 100% away from the shoot stem, then LSI value would be 0, 0.2, 0.4, 0.6, 0.8 and 1.0, respectively. The leaves with larger LSI values would reduce crowding of leaf area around the stem and thus, reduce mutual shading among the neighbouring

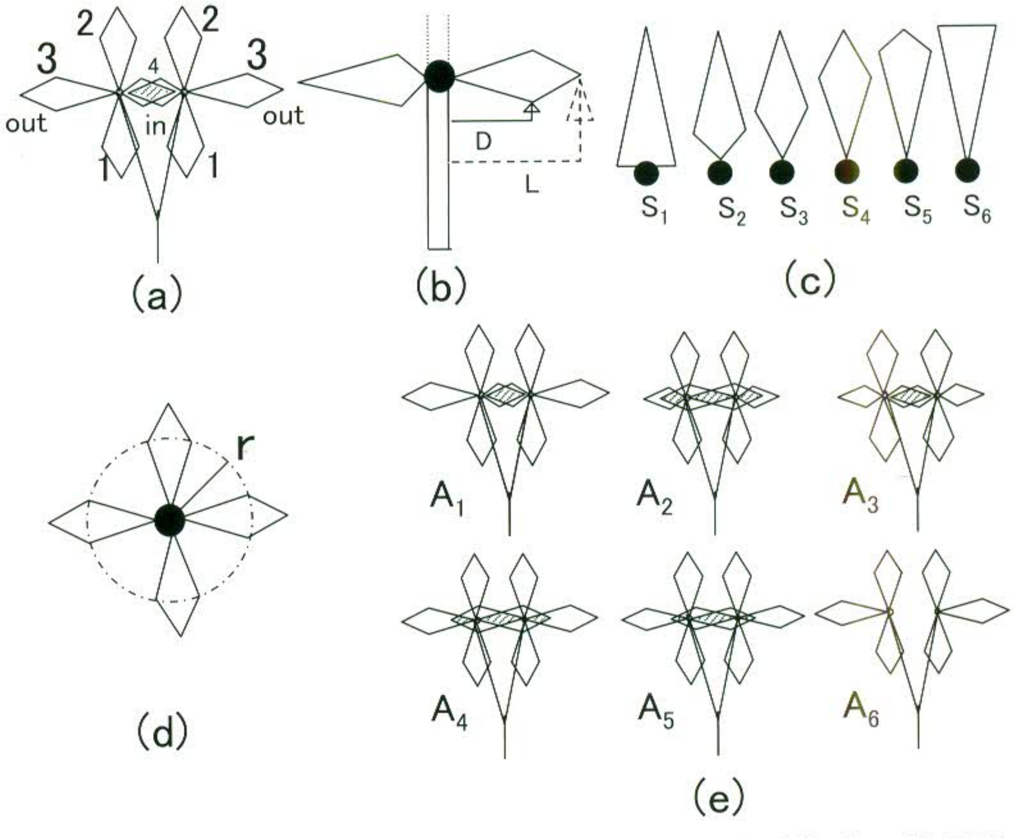

Fig. 1a–d. Canopy stature, leaf shape index and alternative canopy forms. **a** Canopy structure of adult *Aucuba japonica* plant assuming two sister shoots (each with two pairs of leaves) grown from the terminal reproductive bud at maturity. Shoot orientation almost orthotropic with nearly horizontal leaf display. Integers *1* and *2* constitute the basal pair (isophyllous) and *3* and *4* constitute the second pair (anisophyllous) while *font size* indicates intra-pair leaf size difference. *In* and *out* indicate inner and outer canopy, respectively. **b** Determination of LSI. *D* denotes distance between the petiole base at the stem and the point on the midrib where the line for maximum width of the lamina crosses. *L* denotes total distance from the petiole base to the lamina tip. The *black circle* indicates stem cross-section wherever shown. Two different leaf shapes shown on a node. **c** Varying shapes to simulate self-shading at shoot level (*1–6* in regard to LSI values of 0, 0.2, 0.4, 0.6, 0.8 and 1.0). The juvenile plant canopy was comprised of a single leaf shape very similar to S_4 while the adult canopy possessed two different shapes similar to S_3 and S_4. **d** Determination of DPSS of a shoot having two pairs of leaves (with S_4). Top view of a shoot. **e** Alternative canopy forms depending mainly on size and change in orientation of anisophyllous leaf pair. Extreme anisophylly gives rise to hollow canopy (A_6). A_1 is the prototype of crown form in field condition. In these simulations, only two pairs of current leaves (one isophyllous and one anisophyllous pair) have been considered although number of pairs could vary from one to four. Also, the year-old leaves beneath the current leaves were ignored

leaves. For each value of LSI, we modelled leaf shapes, S_1, S_2, S_3, S_4, S_5 and S_6 (Fig. 1c), for simulating the self-shading.

Simulating effects of the leaf shape

To quantify the effect of the diversity of leaf shapes, we estimated the degree of potential self-shading (DPSS) at current shoot level for both isophyllous juvenile and anisophyllous adult shoots. Occurrence of the maximum width along petiole–leaf lamina length could be an important factor for self-shading on a shoot (here we considered a single shoot and ignored the effect of neighbouring shoots). Assuming leaves on a shoot are arranged decussately on the circumference of a circle that encircles the shoot stem on a horizontal plane, we defined DPSS as the ratio of sum of maximum width of all the leaves residing on the circumference to the circumference of the circle. Circumference of a circle equals $2\pi r$, where r denotes radius of the circle. The centre of the circle was the centre of shoot stem cross-section. From the centre, we measured the distances to the maximum width for all the leaves on the shoot. Then we determined the mean of the measured distances to get the radius of the circle. $\text{DPSS} > 1$ indicates overlapping among the neighbouring leaves on the circumference, $\text{DPSS} = 1$ indicates no overlap with leaves just touching each other, and $\text{DPSS} < 1$ indicates distant arrangement of leaves. For a given leaf size, we simulated DPSS by changing the leaf shape (S_1, S_2, S_3, S_4, S_5 and S_6) but keeping the maximum width constant. In the case of S_1, the circumference of the circle was the circumference of the stem cross-section. For each leaf shape, we determined the radius and circumference of the circle, and then DPSS.

Measurement of canopy area

We measured crown diameter (a, b) in two directions perpendicular to each other and then averaged. Canopy area we estimated as:

$$C_{\text{area}} = \pi r^2,$$

where r denotes the radius estimated as $(a + b)/4$.

Leaf-area index at canopy level

For both juvenile and adult plants, we estimated leaf-area index (LAI) (expressed as $\text{cm}^2\ \text{cm}^{-2}$) as the amount of leaf area displayed by the crown per unit ground area occupied by the canopy.

Simulating percent leaf-area overlap at canopy level

We estimated percent leaf-area overlap (PLAO) in the crown as:

$$\text{PLAO} = \left(\frac{\text{TLA} - \text{PLA}}{\text{TLA}}\right) \times 100,$$

where TLA denotes the total leaf area (cm^2) per crown and PLA denotes projected leaf area (cm^2). To simulate crown leaf area, we measured 40 adult plants. Then, we calculated mean dimension of leaf lamina, stem diameter and distance between the tips of sister shoots. Depending on leaf laminar dimensions, we determined TLA as the sum of the area of all the simulated leaves of the canopy; we determined the area of each leaf using Adobe Photoshop software. We also determined PLA using the same software. In these simulations, for a single canopy, we used two different leaf shapes, S_3 (similar to the smaller leaf of the anisophyllous pair) and S_4 (similar to the larger leaf of the anisophyllous pair), at a time.

To compare PLAO and canopy size of adult plants, we used six alternative scenarios (A_1–A_6) representing changes in orientation and size of anisophyllous leaves in the N_2 position. The orientation of the isophyllous (N_1) pair was unchanged for all the alternatives. A_1 is the prototype in which the smaller anisophyllous leaf in the N_2 position is oriented to the inner canopy and the larger leaf is oriented to the periphery of the canopy as is observed in real plants. A_2, the opposite of A_1, has the larger leaf oriented to the inside and the smaller leaf to the periphery of the canopy. This could happen if the initial primordial size were to dominate the leaf developmental course, since at inception, the inner primordium is much larger than the outer one (Ali and Kikuzawa 2005). For A_3, we considered leaves similar to the anisophyllous smaller leaf on both sides of the shoot. This could also happen if growth of both the primordia were suppressed equally. For A_4, we considered leaves similar to the larger leaf of the anisophyllous pair on both sides of the shoot. This could occur in the absence of an inflorescence as in juvenile plants. For A_5, we used average leaf size of two anisophyllous leaves on both sides of the shoot. It can be thought that the outer leaf became larger at the cost of the inner leaf by taking advantage of the unfavourable growth conditions of the inner leaf. If there were equal opportunity, then isophyllous leaf size would be about the mean size of existing leaves. A_6 was the case of extreme anisophylly. This could happen if growth of the inner smaller leaf was extremely suppressed or turn into a bud scale.

In these simulations, we considered two sister shoots (each with two pairs of leaves) extended from a single supporting axis of a juvenile plant at maturity. Under natural conditions, the number of leaf pairs varied from two to four and there were year-old leaves beneath the current-year leaves. However, we neglected these for simplicity. We assumed that the model leaves were oriented decussately on a horizontal plane.

Determination of aboveground biomass

For determination of plant aboveground biomass (dry weight of stem, petiole and leaves), we collected five juvenile and five adult plants among the previously measured sample plants. Immediately after harvesting, we separated each plant into stem and leaf with petiole. Samples were oven-dried at 80°C to constant weight.

To estimate aboveground biomass for the other sample plants measured, we used the following equations:

(1) Stem weight (g) is expressed as

$$Y_S = 0.1369(SL \times SD) - 0.0276\ (r = 0.9;\ P < 0.001),$$

where SL and SD are stem length (cm) and stem diameter (cm), respectively. We assumed similar wood density for juvenile and adult plants because of the relatively small range of plant ages.

(2) Petiole weight (g) is expressed as

$$Y_P = 0.0069(PL \times PD) - 0.0013\ (r = 0.91;\ P < 0.001),$$

where PL and PD are petiole length (cm) and petiole diameter (cm), respectively.

(3) Leaf weight (g) is expressed as

$$Y_L = 0.0066LA - 0.0004\ (r = 0.95;\ P < 0.001),$$

where LA denotes individual leaf area (cm^2). We assumed similar leaf thickness for all the individuals (juveniles and adults).

Determination of C:F ratio

We determined C:F ratio as the ratio of total dry mass of leaf support tissues (stem plus petiole) to total foliage weight at individual plant level.

Biomass use index

We defined biomass use index as the ratio of canopy area to aboveground biomass of the individual plant. The unit of the index is $cm^2\ g^{-1}$. We used this as an indicator of biomass use efficiency in terms of the occupation of canopy space.

Statistical analyses

For comparing leaf size and degree of anisophylly, we used the Mann-Whitney *U*-test in SPSS software, one-way ANOVA for leaf lamina narrowness and LSI, and Excel software for graphs.

Results

Single-stemmed juvenile plants turned into multi-stemmed adult form upon achieving reproductive maturity. Reproductive maturity was also accompanied by anisophylly. In juveniles, two leaves on any node grew almost equally, and thus the index of anisophylly was close to unity (Fig. 2a). Conversely, adult shoots showed well-marked anisophylly on alternate nodal positions (Fig. 2b). When leaves of a pair (e.g., N_1, N_3) were grown tangentially to the inflorescence axis, the ratio was close to unity, but when leafing direction

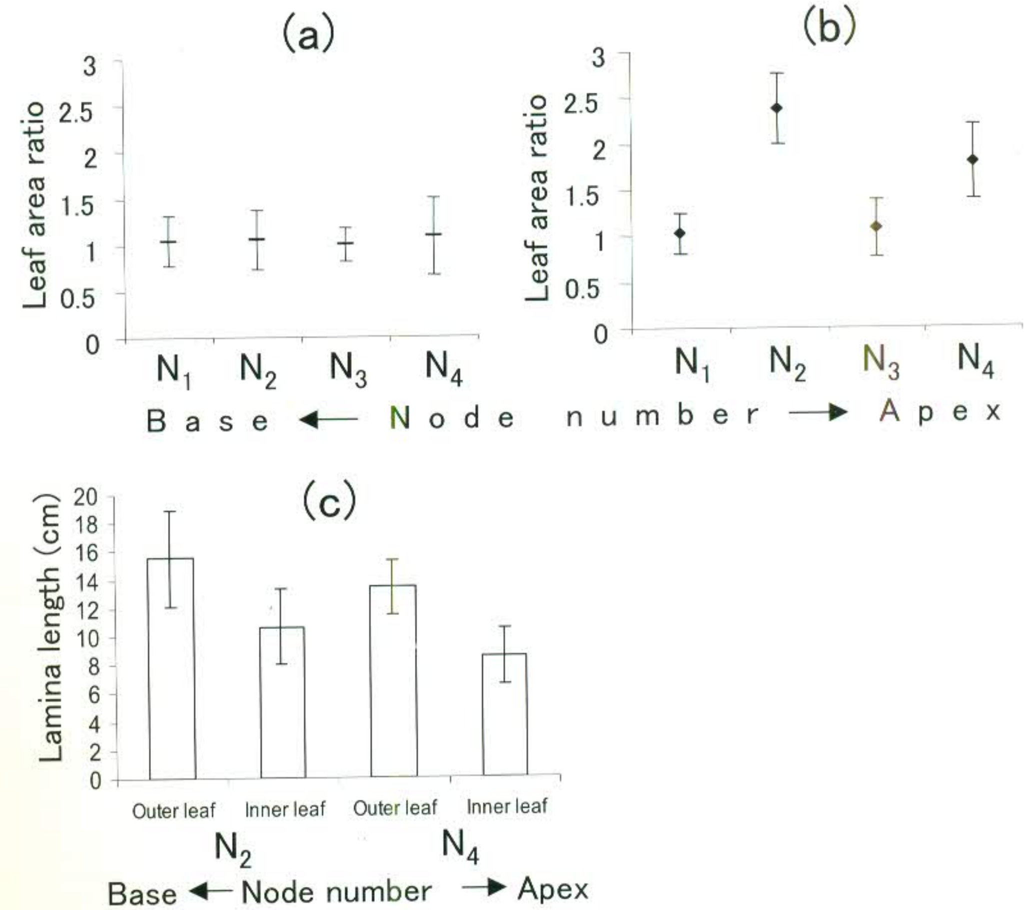

Fig. 2a–c. Leaf area ratio along shoot length. **a** Juvenile plant (*dash*), **b** adult plant (*filled diamond*) and **c** comparison of leaf laminar lengths on anisophyllous nodes. *Vertical bar* indicates SD. Leaf area ratio is used to express the degree of anisophylly, i.e., the magnitude of difference between two leaves of a pair

changed by 90° (e.g., N_2, N_4), the mean ratio (outer to inner) was significantly greater than unity ($P < 0.001$, Mann-Whitney *U*-test; more than 2 for N_2 and nearly 2 for N_4; Fig. 2b). The mean ratio for N_2 was significantly larger than N_4 ($P < 0.01$, Mann-Whitney *U*-test), i.e., the degree of anisophylly reduced acropetally (Fig. 2b). Anisophylly caused significantly larger leaf area display in the outer compared to the inner canopy (2.38 times the inner leaf in the N_2 and 1.79 times in the N_4 position; Fig. 2b). For anisophyllous pairs, mean laminar lengths of outer and inner leaves were 15.5 ± 3.41 (mean ± SD) and 10.65 ± 2.68 for N_2 and 13.45 ± 1.97 and 8.49 ± 2.0 for N_4 positions, respectively (Fig. 2c).

Juvenile plants were shorter than adult plants (Table 1). Before maturity, shoot structure of juvenile plants, e.g., length of comparable INs and number of INs between two consecutive years did not change remarkably (Fig. 3a–c).

Even after entering the adult stage, shoot structure did not change significantly (Fig. 3d) except in the total number of shoots. Until maturity, leaf production (number of leaves per year per individual) between 2 consecutive years did not change significantly, but the transition from juvenile to adult form caused a sudden significant increase in leaf production (Fig. 3e) without significant increase in canopy area (Table 1). As a result, mean LAI was significantly larger for adult plants than juvenile plants (Table 1). The distance between tips of two sister shoots (extended from a single supporting axis at maturity) ranged from 5–20 cm, and the mean distance was 12.3 ± 3.92 cm (Fig. 3f). This distance is shorter than the leaf lamina length especially on anisophyllous nodes where inner leaves on neighbouring shoots face each other (Fig. 2c) and thus, could shade each other easily.

Adult plants produced a larger number of leaves and mean leaf size was reduced significantly at the switch from the juvenile to adult stage (Table 1). Both stages showed acropetal reduction in leaf size (Fig. 4a). Anisophylly caused additional, position-dependent reduction in leaf size

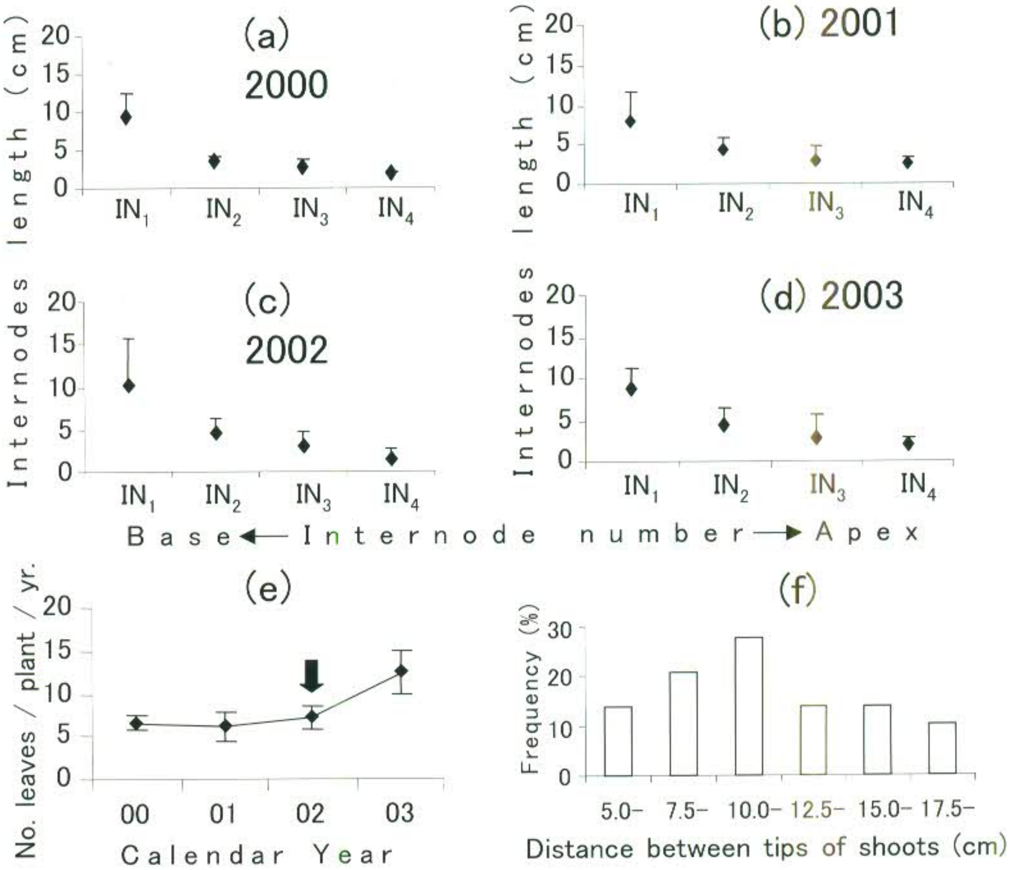

Fig. 3a–f. Shoot structure and leaf production in the canopy. **a–c** Shoot structure during juvenile phase (in years 2000–2002), **d** shoot structure during adult phase (in year 2003), **e** leaf production in different years, and **f** frequency distribution of distance (cm) between two daughter shoots extended from a single mother axis. *Arrow* indicates transition from juvenile to adult form. *IN* denotes internode

Table 1. Size parameters of juvenile and adult *Aucuba japonica* plants

Parameters	Juvenile plant (mean ± SD)	Adult plant (mean ± SD)	Level of significance
Plant height (cm)	50.5 ± 13.36	72.97 ± 21.54	$P < 0.05$ (one-way ANOVA)
Canopy area (cm^2)	1,291.74 ± 396.98	1,378.91 ± 606.3	$P > 0.05$ (Mann-Whitney *U*-test)
Leaf size (cm^2)	91.41 ± 40.99	62.35 ± 32.19	$P < 0.001$ (one-way ANOVA)
LAI ($cm^2\ cm^{-2}$)	0.67 ± 0.14	1.05 ± 0.42	$P < 0.01$ (Mann-Whitney *U*-test)

To estimate mean leaf size, only current-year leaves were taken into account

(Fig. 4a). For an anisophyllous pair, the mean size of the outer leaf, i.e., leaf no. 3 (88.82 ± 36.61), was not significantly different in size from any other position. Following leaf size, petiole length showed a similar descending pattern towards the tip of the shoot (Fig. 4b). Leaf lamina narrowness increased acropetally except for the inner leaves of the anisophyllous pairs (Fig. 4c).

For the anisophyllous pairs, the inner leaf, i.e., leaf no. 4 of N_2, was significantly wider (proportionally) than the outer leaf (leaf no. 3, $P < 0.001$, one-way ANOVA), while those in the N_4 position did not differ significantly. For juvenile plants, LSI was almost similar to any leaf in any nodal position (0.61 ± 0.06) where maximum width of lamina occurred after the midpoint (Fig. 4d). For adult plants, LSI for N_1 (0.61 ± 0.09) was similar to that of juvenile plants, but the indices for N_2 and N_4 were significantly smaller ($P < 0.05$, one-way ANOVA). The LSI between the outer (0.57 ± 0.07) and the inner leaf (0.47 ± 0.07) in the N_2 position was significantly different ($P < 0.001$, one-way ANOVA) but the difference was not significant in the N_4 position. A smaller LSI indicates closer occurrence of maximum width to the stem, which was conspicuous for the inner leaves in the N_2 and N_4 positions (Fig. 4d).

At the shoot level, a consideration of DPSS using the different simulated leaf shapes indicated that the closer the occurrence of the maximum width to the stem the higher the self-shading and vice versa (Table 2). There was a sharp decline in DPSS between S_1 and S_2 while DPSS reduction was rather gradual from S_2 (Table 2). For a given leaf shape, DPSS was significantly smaller for adult shoots (Table 2).

Although occurrence of the maximum width farther from the stem reduced self-shading, proportionally higher biomass investment into the petiole (Fig. 5a) was needed for leaf display.

The mean area of the simulated crown was significantly larger for A_1 (328.25 ± 24.86), the prototype of naturally grown seedlings, than for any other alternative canopy forms except A_4 and A_6 ($P < 0.05$, Mann-Whitney U-test; Fig. 5a). A_4 acquired an area similar to A_1, but PLAO was significantly larger (Fig. 5b). PLAO was significantly smaller ($P < 0.05$, Mann-Whitney U-test) for A_6 (0) followed by A_1 relative to other alternatives. The very high crowding of leaf area in the inner canopy caused maximum PLAO (13.55 ± 0.96) for A_2, while the inner canopy of A_6 was vacant due to extreme anisophylly.

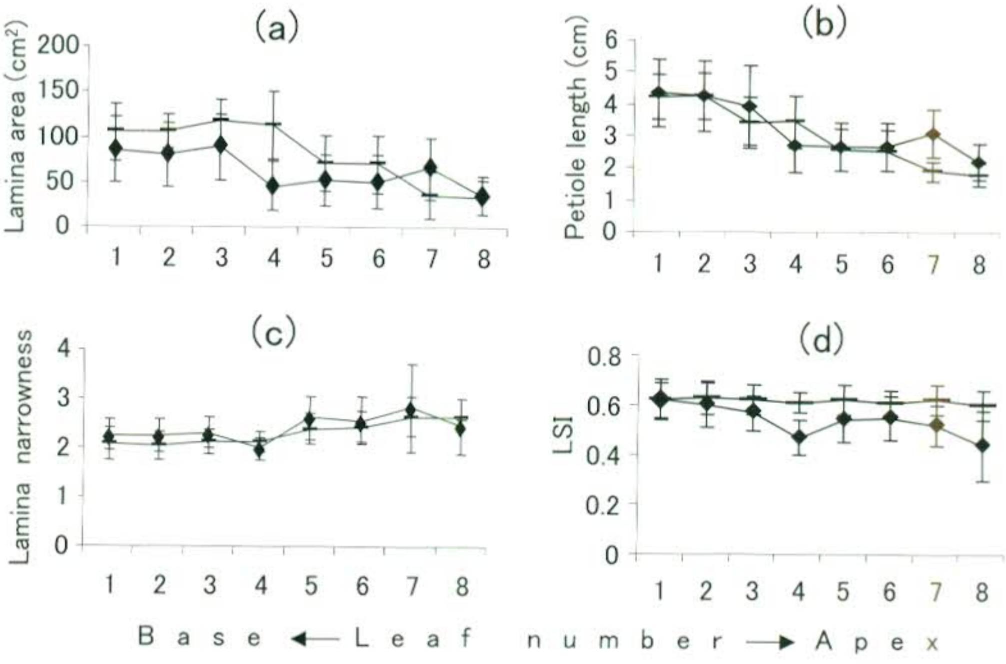

Fig. 4a–d. Comparison of foliage leaf morphology between juvenile and adult plants. **a** Individual leaf size (acropetally), **b** petiole length, **c** leaf lamina narrowness and **d** leaf shape index (LSI). *Vertical bar* indicates SD. *Dash* Juvenile, *filled diamond* adult plant. Two consecutive leaves made a pair

Table 2. Results of simulation of varying leaf shapes on the degree of potential self-shading

Leaf shape	Juvenile plant (mean ± SD)	Adult plant (mean ± SD)	Level of significance
S_1	26.25 ± 4.36	22.73 ± 8	$P < 0.05$ (Mann-Whitney U-test)
S_2	2.06 ± 0.39	1.82 ± 0.29	$P < 0.01$ (one-way ANOVA)
S_3	1.03 ± 0.19	0.91 ± 0.15	$P < 0.01$ (one-way ANOVA)
S_4	0.69 ± 0.13	0.61 ± 0.09	$P < 0.01$ (one-way ANOVA)
S_5	0.52 ± 0.09	0.45 ± 0.07	$P < 0.01$ (one-way ANOVA)
S_6	0.41 ± 0.09	0.36 ± 0.06	$P < 0.01$ (one-way ANOVA)

In this simulation, variations in the occurrence of maximum width were made for a single leaf shape for juveniles and for two different leaf shapes for adult shoots, because on adult shoots, the shape of anisophyllous leaves was different. The difference in DPSS between juvenile and adult plants was mainly due to anisophylly

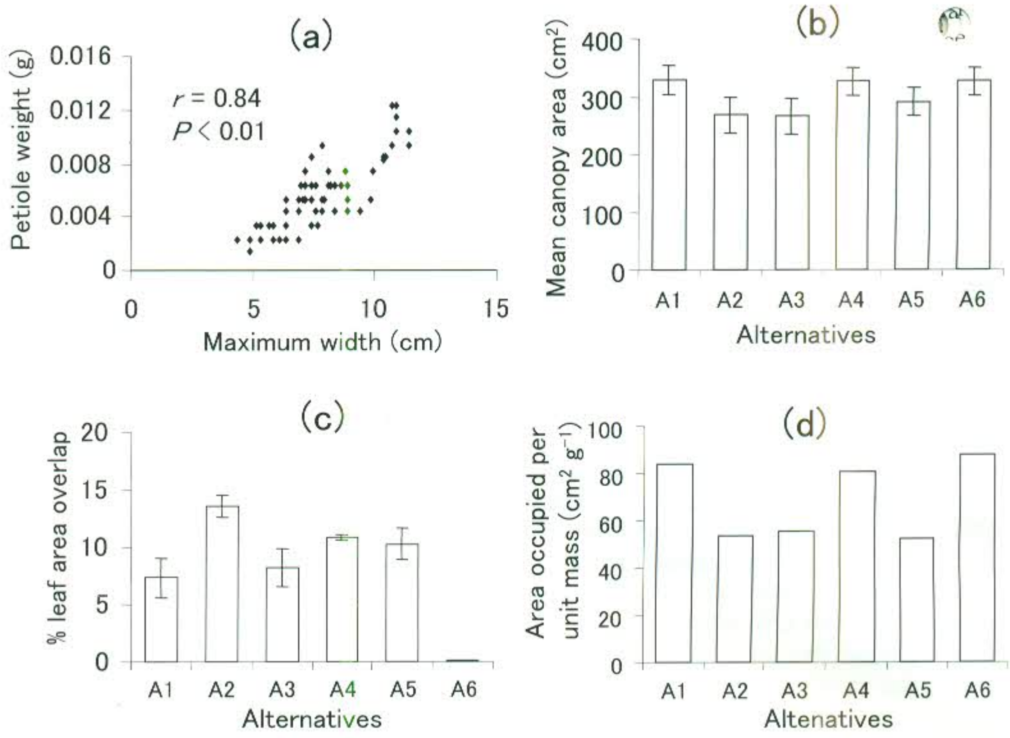

Fig. 5a–d. Leaf display cost, relative size of alternative crown forms and cost of area acquisition. **a** Relationship between the occurrence of maximum width of leaf lamina and the biomass of petiole of the leaf. **b** Mean canopy size of simulated crown. **c** Percentage of leaf area overlap of simulated crown. **d** Mean ground area occupation per unit biomass under field conditions compared to other alternatives

Table 3. Biomass use efficiency of *Aucuba japonica* plants

Parameters	Juvenile plant (mean ± SD)	Adult plant (mean ± SD)	Level of significance
C:F ratio	0.76 ± 0.12	1.09 ± 0.23	$P < 0.001$(Mann-Whitney U-test)
Biomass use index (cm^2 g^{-1})	135.72 ± 33.24	84.13 ± 31.9	$P < 0.001$ (one-way ANOVA)

The C:F ratio for adult plants was significantly larger than the ratio for juvenile plants. Because of greater investment of biomass into foliage, the biomass use index of juvenile plants was significantly larger than adult plants (Table 3). In contrast, despite proportionally reduced investment into foliage, anisophylly increased biomass use efficiency of adult *Aucuba japonica* plants. In the context of field conditions, A_1 had significantly greater biomass use efficiency than A_2, A_3, and A_5. The canopy space occupied per unit aboveground biomass was due to less self-shading as a consequence of anisophylly. A_1 showed similar efficiency to A_4 but less than A_6 (Fig. 5d).

Discussion

Juvenile plants maintained nearly a constant leaf area in the crown until maturity (Fig. 3e), most probably due to structural limitation (single-stemmed structure) for leaf deployment. Isophyllous leaves could find accommodation in rotation around the single stem without significant overlap (Morley 1973). Only at maturity were *Aucuba japonica* plants able to overcome this structural limitation by producing multiple shoots from a single supporting axis. This led to a sudden increase in leaf production (Fig. 3e) without a simultaneous increase in canopy area (Table 1), which necessitated morphological readjustment of leaf size and shape and also petiole length to minimize potential self-shading. Reduction in leaf size was the dominant trait in these morphological readjustments. As plants matured, mean leaf size was reduced in general (Table 1). Adult plants produced a large number of smaller leaves instead of making a smaller number of larger leaves; making smaller leaves acts to reduce mutual shading of adjacent shoots (Osada et al. 2002) and thus, enhances light penetration in the crown (Horn 1971). This may be particularly important for evergreen *A. japonica* because the older leaves, especially the year-old leaves (immediately below the current leaves), which are still photosynthetically active, would otherwise suffer from light starvation. As part of the correlated morphological readjustments, anisophylly contributed to further place-dependent reductions in leaf size. Making variously sized leaves is more important for efficiently tailoring leaf area on the shoot (Goebel 1900; Givnish 1984) than making only smaller leaves because efficient tailoring is essential for avoiding mutual shading at the shoot level as well as at the whole-crown level (Osada et al. 2002; Takenaka 1994; Valladares et al. 2002).

Diversity of leaf shape is clearly an important factor for the efficient display of leaf area. Differentiation in leaf shape caused greater reduction in leaf overlap (Osada et al.

2002) by positional adjustment of leaves around the stem (Takenaka 1994). A larger LSI (outer leaf of anisophyllous pair) placed leaves more in the crown periphery of the shoot while a smaller LSI (inner leaf of anisophyllous pair) with proportionally wider leaf lamina (Fig. 4c, d) filled the nearby space of the supporting stem on one hand and reduced potential shading of neighbouring sister shoots on the other hand. Both basipetal increase in petiole length and acropetal increase in leaf lamina narrowness (Ali and Kikuzawa 2005; Takenaka 1994) reduced crowding of leaf area in close vicinity of the stem and consequently, reduced leaf overlap (Takenaka 1994). The occurrence of maximum leaf width closer to the stem, e.g., S_1 (Fig. 1c), although economically and mechanically safer, casts the highest DPSS. To reduce self-shading, leaf size must be reduced. In contrast, placing maximum leaf width distally, e.g., S_6 (Fig. 1c), reduces the probability of self-shading, but such a leaf shape needs proportionally greater biomass investment in non-photosynthetic petioles (Yamada et al. 2000). Given all these considerations, leaf shape S_4, which is similar to actual leaf shape, does indeed appear to be most suitable for minimizing the risk of self-shading. Combinations of different leaf shapes, e.g., S_3 (as inner leaf) and S_4 (for all other leaves), appeared to be the most efficient leaf area arrangement in terms of space filling and reducing self-shading, in contrast to the single-shaped leaves typical of juvenile plants (Fig. 4d), which inefficiently fill the space near the stem. Self-shading was significantly lower on adult shoots due to the combination of diverse leaf shapes and double (general and place-dependent) reduction in leaf size. Reduction in leaf size caused reduction in width, while variation in shape (Fig. 4d) was due to anisophylly, as shape was independent of leaf size (Dale 1986). Therefore, anisophylly, accompanied by a combination of modified characters such as leaf size and shape, petiole length, and lamina narrowness (Fig. 4), and thus, serving as a synthetic character, played a significant role in displaying the components of photosynthetic surfaces to maximize light capture (Niklas 1988; Takenaka 1994; Valladares et al. 2002) at the shoot level as well as at the whole-crown level of *Aucuba japonica*.

Anisophylly not only caused effective filling of space but also resulted in acquired greater total area per individual (Fig. 5b) as the larger leaf in the pair was oriented with the longer petiole in the outer canopy. This developmentally correlated leaf orientation increased both canopy projection area (Yamada et al. 2000) and biomass use efficiency, and thus should increase competitive ability of *Aucuba japonica* seedlings. Greater space acquisition by anisophylly helped *A. japonica* to provide space for suddenly increased leaf area at maturity and thus, diluted the crowding of leaf area. This was particularly conspicuous for anisophyllous pairs where plants displayed significantly greater leaf area towards the outer canopy. Because of this dilution effect (Goebel 1900), leaf overlap was minimal (Fig. 5c) and light capture per unit leaf area increased (Pearcy and Yang 1998; Pearcy and Valladares 1999). From the perspective of photosynthesis, this architecture should enhance plant performance (Niklas 1988). Anisophylly increased area acquisition, economic filling of acquired area and minimization of leaf area overlap, and thus can be viewed as a rescue mechanism ensuring an effective transition in light-capture systems from juvenile to adult *A. japonica* plants.

The ontogenetic disruption of light-capture systems due to disproportionate increases in annual leaf area production versus woody shoot structure (Fig. 3a–c, e) forces *Aucuba japonica* plants to readjust morphologically to minimize self-shading. Responding to this ontogenetic imperative, *A. japonica* leaves show evidence of adaptive plasticity (Dale 1986). Anisophyllous leaves exhibited plastic responses in both shape and size. In the adult shoot system, occurrence of anisophylly was not random but rather occurred at specific position(s) where possibility of self-shading was greater. This was perhaps due to selection pressure to reduce self-shading. Isophyllous development of adult shoots due to desuppression by excision of inflorescence (Ali and Kikuzawa 2005) indicated that the anisophyllous leaf did not permanently lose its potential to grow larger, but rather that growth was just suppressed (Basile and Basile 1993). This clearly indicated that anisophylly is an adaptation lending the advantage of less self-shading, increased biomass use efficiency, and greater area occupancy to *Aucuba*. Although extreme anisophylly eliminated the possibility of self-shading, it caused a hollow crown and inefficient use of acquired space. In overall consideration, *A. japonica* expressed anisophylly to the necessary extent and is adaptive to it.

While analyzing biomass cost of area occupancy, it appeared that, compared to adult plants, greater investment of aboveground biomass into foliage (Table 1) relative to the support structure might enable juvenile plants to occupy significantly greater areas per unit biomass. Since juvenile plants are smaller in size and thus less vulnerable to stresses, such as wind, maximizing area occupation through investment in foliage might be cheaper and more efficient for them. Larger investment into foliage increases competitive ability of juvenile plants in two ways, by increased area occupancy and by increased light capture through horizontal spreading of foliage, a key factor for plant growth and survival in the understory (Canham 1985, 1988). In contrast, greater proportion of biomass investment into leaf support structure of adult plants could not increase area occupancy or decrease self-shading without anisophylly.

Conclusion

The form and the extent of anisophylly expressed by *Aucuba japonica* are adaptive. The problem of potential self-shading posed by unparallel production of leaf area and supporting shoot structure was resolved in an integrated way by making smaller leaves in general and by making anisophyllous leaves on the same node. Anisophylly caused greater area acquisition to accommodate excess leaf area produced in the crown with minimal self-shading. For efficient tailoring of leaf area, diverse leaf forms accompanied by anisophylly are superior to monotonic single leaf forms.

The plants increased biomass use efficiency and economic filling of the acquired area.

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References

Ali MS, Kikuzawa K (2005) Anisophylly in *Aucuba japonica* (Cornaceae): an outcome of spatial crowding in bud. Can J Bot 83:143–154

Basile DV, Basile MR (1993) The role of place-dependent suppression of cell division in plant morphogenesis and phylogeny. Mem Torrey Bot Club 25:63–84

Brokaw NVL (1985) Gap-phase regeneration in a tropical forest. Ecology 66:682–687

Canham CD (1985) Suppression and release during canopy recruitment in *Acer saccharum*. Bull Torrey Bot Club 112:134–145

Canham CD (1988) Growth and canopy architecture of shade-tolerant trees: response to canopy gaps. Ecology 69:786–795

Chazdon RL (1985) Leaf display, canopy structure and light interception of two understory palm species. Am J Bot 72:1493–1502

Dale JE (1986) Plastic responses of leaves. In: Jennings DH, Trewavas AJ (eds) Plasticity in plants. Symposium of the Society of Experimental Biology. Company of Biologists, Cambridge, pp 287–305

Dengler NG (1999) Anisophylly and dorsiventral shoot symmetry. Int J Plant Sci 160(6 Suppl.):S67–S80

Fisher JB (1978) A quantitative study on terminalia branching. In: Tomlinson PB, Zimmerman MH (eds) Tropical trees as living system. Cambridge University Press, Cambridge, UK, pp 285–320

Givnish TJ (1984) Leaf and canopy adaptations in tropical forests. In: Medina E, Mooney HA, Vazquez-Yanes C (eds) Physiological ecology of plants of the wet tropics. Junk, The Hague, pp 51–84

Goebel K (1900) Organography of plants, English edn. Oxford University Press, Oxford

Hara N (1980) Shoot development of *Aucuba japonica* I. Morphological study. Bot Mag Tokyo 93:101–116

Horn HS (1971) The adaptive geometry of trees. Princeton University Press, Princeton, p 144

Kessler S, Kim M, Pham T, Weber N, Sinha N (2001) Mutations altering leaf morphology in tomato. Int J Plant Sci 162:475–492

Kikuzawa K, Umeki K, Lechowicz MJ (1996) Some evidence for an adaptive linkage between leaf phenology and shoot architecture in sapling trees. Funct Ecol 10:252–257

Kume A, Ino Y (2000) Differences in shoot size and allometry between two evergreen broad-leaved shrubs, *Aucuba japonica* varieties in two contrasting snowfall habitats. J Plant Res 113:353–363

Morley B (1973) Ecological factors of importance to *Columnea* taxonomy. In: Heywood VH (ed) Taxonomy and ecology. Academic Press, London, pp 265–281

Muraoka H, Koizumi H, Pearcy RW (2003) Leaf display and photosynthesis of tree seedlings in a cool-temperate deciduous broadleaf forest understorey. Oecologia 135:500–509

Niklas KJ (1988) The role of phyllotactic pattern as a "developmental constraint" on the interception of light by leaf surface. Evolution 42:1–16

Niklas KJ (1992) Plant biomechanics: an engineering approach to plant form and function. University of Chicago Press, Chicago

Ohi T, Kajita T, Murata J (2003) Distinct geographic structure as evidenced by chloroplast DNA haplotypes and ploidy level in Japanese *Aucuba* (Aucubaceae). Am J Bot 90:1645–1652

Osada N, Takeda H, Furukawa A, Awang M (2002) Changes in shoot allometry with increasing tree height in a tropical canopy species, *Elateriospermum tapos*. Tree Physiol 22:625–632

Pearcy RW, Valladares F (1999) Resource acquisition by plants: the role of crown architecture. In: Press M, Scholes JD, Barker MG (eds) Physiological plant ecology. Blackwell Science, London, UK, pp 45–66

Pearcy RW, Yang W (1998) The functional morphology of light capture and carbon gain in the redwood-forest understorey plant, *Adenocaulon bicolor* Hook. Funct Ecol 12:543–552

Sánchez-Burgos AA, Dengler NG (1988) Leaf development in isophyllous and facultatively anisophyllous species of *Pentadenia* (Gesneriaceae). Am J Bot 75:1472–1484

Takenaka A (1994) Effects of leaf-blade narrowness and petiole length on the light capture efficiency of a shoot. Ecol Res 9:109–114

Takenaka A, Inui Y, Osawa A (1998) Measurement of three-dimensional structure of plants with a simple device and estimation of light capture of individual leaves. Funct Ecol 12:159–165

Tomlinson PB (1978) Branching and axis differentiation in tropical trees. In: Tomlinson PB, Zimmermann MH (eds) Tropical trees as living systems. Cambridge University Press, Cambridge, pp 187–207

Tsukaya H (1998) Genetic evidence for polarities that regulate leaf morphogenesis. J Plant Res 111:113–119

Valladares F, Wright SJ, Lasso E, Kitajima K, Pearcy RW (2000) Plastic phenotypic response to light of 16 congeneric shrubs from Panamanian rainforest. Ecology 81:1925–1936

Valladares F, Skillman JB, Pearcy RW (2002) Convergence in light capture efficiencies among tropical forest understorey plants with contrasting crown architectures: a case study of morphological compensation. Am J Bot 89:1275–1284

Yamada T, Suzuki E (1996) Ontogenetic change in leaf shape and crown form of a tropical tree species, *Scaphium macropodum* (Sterculiaceae) in Borneo. J Plant Res 109:211–217

Yamada T, Okuda T, Abdullah M, Awang M, Furukawa A (2000) The leaf development process and its significance for reducing self-shading of a tropical pioneer tree species. Oecologia 125:476–482