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# Stem cambial variants of selected Cucurbitaceae plants in Taiwan

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ABSTRACT: The Cucurbitaceae family is characterized by bicollateral vascular bundles with both outer and inner phloem. Studies on the secondary growth of bicollateral vascular bundles are scarce. The secondary growth of cambia were observed in approximately 12 Cucurbitaceae climbers from Taiwan. The experimental results showed that stems cross-section of cambial variant types presented irregular conformation, axial vascular elements in segments, furrows xylem of the phloem wedge, successive cambia, external secondary vascular cylinder, and a combination type of two or three cambial variants. Most species have ten bicollateral vascular bundles and arranged in the outer and inner cylinders, as well as few species have 14 and 20 bundles. *Neoalsomitra clavigera* (Wall.) Hutch. has14 bicollateral vascular bundle, which divided by two broad rays and arranged in two cylinder equally. *Momordica cochinchinensis* (Lour.) Spreng. exhibits 20 vascular bundles, which divided by four broad rays, five in inner cylinder and 15 in outer cylinder. *Gynostemma pentaphyllum* (Thunb.) Makino developed three secondary vascular bundles in the ray dilatation region. *Momordica charantia* L. developed external secondary vascular cylinder from the outer cylinder of the cortical bicollateral vascular bundle. In the young stem, *Sinobaijiania taiwaniana* (Hayata) C. Jeffrey & W.J. de Wilde has 12 bicollateral vascular bundles arranged unequally, five in outer and seven in inner cylinder. Different species will be validated based on the features of secondary growth in cambia. This report provides fundamental information about cambial variants of selected Cucurbitaceae climbers as common taxonomic characteristics.

KEY WORDS: Bicollateral vascular bundles, centrifugal vascular bundles, phloem wedge, ray dilatation, successive cambia.

## INTRODUCTION

Climbers can generally be classified into liana and herbaceous vines based on the degree of stem lignification. Lianas have fibrous, thick, and truly lignified stems, whereas herbaceous vines have slender and flexible stems (Gentry, 1992). Many lianas have stiff searching branches or a self-supporting shrub structure at the beginning of their growth. Moreover, the inner secondary xylem of the self-supporting phase is characterized by a few narrow vessels and thick fibers. However, the xylem of the following climbing phase is characterized by wide vessels, low density, and intermixed soft and stiff tissues. One of the structures that cause irregular stem structures is the presence of cortical vascular bundles. Additionally, it is common for tropical forests to have lianas or herbaceous vines that exhibit these unusual patterns (Beck, 2011).

Globally, the Cucurbitaceae family consists of approximately 95 genera and about 1000 species of climbers, primarily found in the tropics and subtropics, but are very rare in temperate regions (Wu *et al.*, 1994–2004, Schaefer and Acevedo-Rodríguez, 2021). Of these, 20 genera and 32 species are found in Taiwan (Liu, 1993). Concerning herbaceous vines and perennial climbers (lianas), this family uses twining tendrils as a climbing strategy (Yang and Chen, 2021). Cucurbitaceae differ from other families in having a bicollateral vascular bundle, in which the phloem is situated on the opposite side of the xylem. In the transverse section of the *Cucurbita* stem, the external and inner phloems occur at both ends of the vascular bundle (Esau, 2006). The bicollateral vascular bundles of Cucurbitaceae are generally arranged in two rings: the inner ring and the outer ring (Beck, 2011).

The stem cross-sections of *Cucurbita* genus consist of an inner cylinder of a larger bicollateral vascular bundle adjacent to the pith, and an outer cylinder of smaller cortical bicollateral vascular bundle adjacent to the cortex (Beck, 2011). The vascular bundles are divided into two parts: the centripetal and the external parts (Schweingruber *et al.*, 2011). The centripetal part of vascular bundle consists of a centripetal phloem and centrifugal xylem, and cambium does not exsist between xylem and phloem. In contrast, the external part of a vascular bundle have a centripetal xylem, a cambium, and a centrifugal phloem. The interfascicular cambium of Cucurbitaceae is located between the vascular bundles and contributes to radial growth (Schweingruber *et al.*, 2011). As the primary rays widen, the bicollateral vascular bundles are divided, secondary rays (also called new rays) appear in the xylem segments, and new rays are produced with a larger diameter.

Lianescent vascular syndrome in lianas is associated with secondary growth, and numerous studies have been conducted on different families (Metcalfe and Chalk, 1985; Carlquist, 1992a, 2001, 2007, 2013; Caballé, 1993; Jansen *et al*., 2002; Isnard and Silk, 2009; Angyalossy *et al*., 2012, 2015; Yang and Chen, 2016, 2017; Pace *et al.*, 2018; Dias-Leme *et al*., 2021; Yang *et al*., 2021, 2022). Cucurbitaceae has been reported to have single cambial variants or combinations of two or three cambial variants. These include axial vascular elements in segments (also known as xylem in plates), successive cambia, interxylary phloem, furrowed xylem of phloem wedges, and irregular conformation (Angyalossy *et al*., 2012, 2015; Yang *et al*., 2022).

Taiwan is located in the subtropical monsoon region, and owing to the favorable climate, approximately 550 climbers naturally occur in this region (Yang *et al.,* 2022). In this study, we investigated cambial variants of Taiwan's Cucurbitaceae lianas and herbaceous vines. The main aim of the present study was to elucidate anatomical variations and to compare variations in the secondary growth of some Cucurbitaceae species growing in Taiwan. Our results contribute to the classification of climbers, and may ultimately provide accurate morphologies of climbers' secondary growth.

## MATERIALS AND METHODS

All the members of the Cucurbitaceae family do not have bicollateral bundles, in this study we select and observe the species with this feature and submitted the herbarium number. Twelve species, including nine genera of Cucurbitaceae, were collected from open ground or mountainous areas at medium or low altitudes in Taiwan, namely: 1. *Coccinia grandis* (L.) Voigt (PPI 62956), 2. *Gynostemma pentaphyllum* (Thunb.) Makino (PPI 41672), 3. *Melothria pendula* L. (PPI 67148), 4. *Momordica charantia* L. (PPI 80580), 5. *Momordica cochinchinensis* (Lour.) Spreng. (PPI 68515), 6. *Neoalsomitra clavigera* (Wall.) Hutch. (PPI 77772), 7. *Luffa aegyptiaca* Miller, 8. *Mukia maderaspatana* (L.) M. Roem. (PPI 80635), 9. *Sinobaijiania taiwaniana* (Hayata) C. Jeffrey & W. J. de Wilde (PPI 77130), 10. *Trichosanthes homophylla* Hayata (PPI 79957), 11. *Trichosanthes ovigera* Blume (PPI 80455), and 12. *Zehneria mucronata* (Blume) Miq. (PPI 80387). Among these, seven species are lianas and six are herbaceous vines. Among them, the morphologies of *M. charantia* were diverse, we collected two individuals for observing their differences.

We collected plant stems of various sizes from different habitats in Taiwan to observe vascular bundle development. The samples of 13 climbers were collected at breast height (1.3 m from the ground). The fresh materials were divided into pieces of approximately 5 cm in length, and a flat cross-section of each stem was cut using a blade cutter. Images of the stem cross-sections were taken using a Nikon D80 SLR digital camera (Lens AF Micro Nikon 60 mm 1:2.8D; Nikon Corporation, Tokyo, Japan), and qualitative and quantitative anatomical traits were determined using ImageJ software (Ferreira and Rasband, 2012). All photographs were obtained through macroscopic observations. The specimens were dried in an oven (60 °C) for 4–5 d and then stored at -20 °C for 3–4 d. All collected specimens were deposited in the herbarium of Provincial Pingtung Institute (PPI) at the National Pingtung University of Science and Technology, Pingtung, Taiwan, for subsequent identification. The nomenclature follows the Flora of Taiwan Volume III (Liu, 1993) and the Editorial Committee of the Red List of Taiwan Plants (2017).

The following stem features of each species were investigated: habit (liana; herbaceous vine), stem diameter, stem shape, cambial variants, irregular conformation, axial vascular elements in segments, furrowed xylem of phloem wedges, successive cambia, bicollateral vascular bundle number, number of external and inner bicollateral vascular bundles, rays and average ray width, secondary rays, vessels and average vessel diameter, successive cambia layers, secondary vascular bundles, and ray wedges. The features of the cork and cambial variants of the stem were investigated based on the definitions provided by Angyalossy *et al.* (2016), Yang *et al*. (2021, 2022).

## TAXONOMIC TREATMENT

The diagnostic characteristics of the Cucurbitaceae species in Taiwan are presented as follows (Table 1).

**Lianas**

a. ***Coccinia grandis***: the stem cross-section is pentagonal, bark is gray-brown and membranous with dotted lenticels. There are 10 bicollateral vascular bundles. Each bundle consists of two wider, v-shaped rays (Fig. 1A) that measure 0.9–2.3 mm in width, with an average of 1.3 mm, and five to six secondary rays are present in each xylem segment. Ray width generally varied from the stem thickness. The cambial variants types are axial vascular elements divided in radial segments by broad rays of ground tissue combined with irregular conformation, and a furrowed xylem of the phloem wedge. The vessels are diffuse-porous, and the vessel pores are 79–244 μm in diameter, with an average diameter of 169 μm; pith small cavity.

b. ***Gynostemma pentaphyllum***: the cross-section of the stem is slightly pentagonal when the stem diameter is approximately 3 mm and the cork thickness is 0.7 mm (Fig. 1B). The bark is brown and vertically ridged. There are 10 bicollateral vascular bundles; three secondary vascular bundles are exhibited in the ray dilatation region, when the stem diameter is approximately 8 mm. The width of the rays range from 0.06–0.17 mm, with an average of 0.11 mm. The broad rays are wedge-shaped and black (Fig. 1C). The cambial variant types are axial

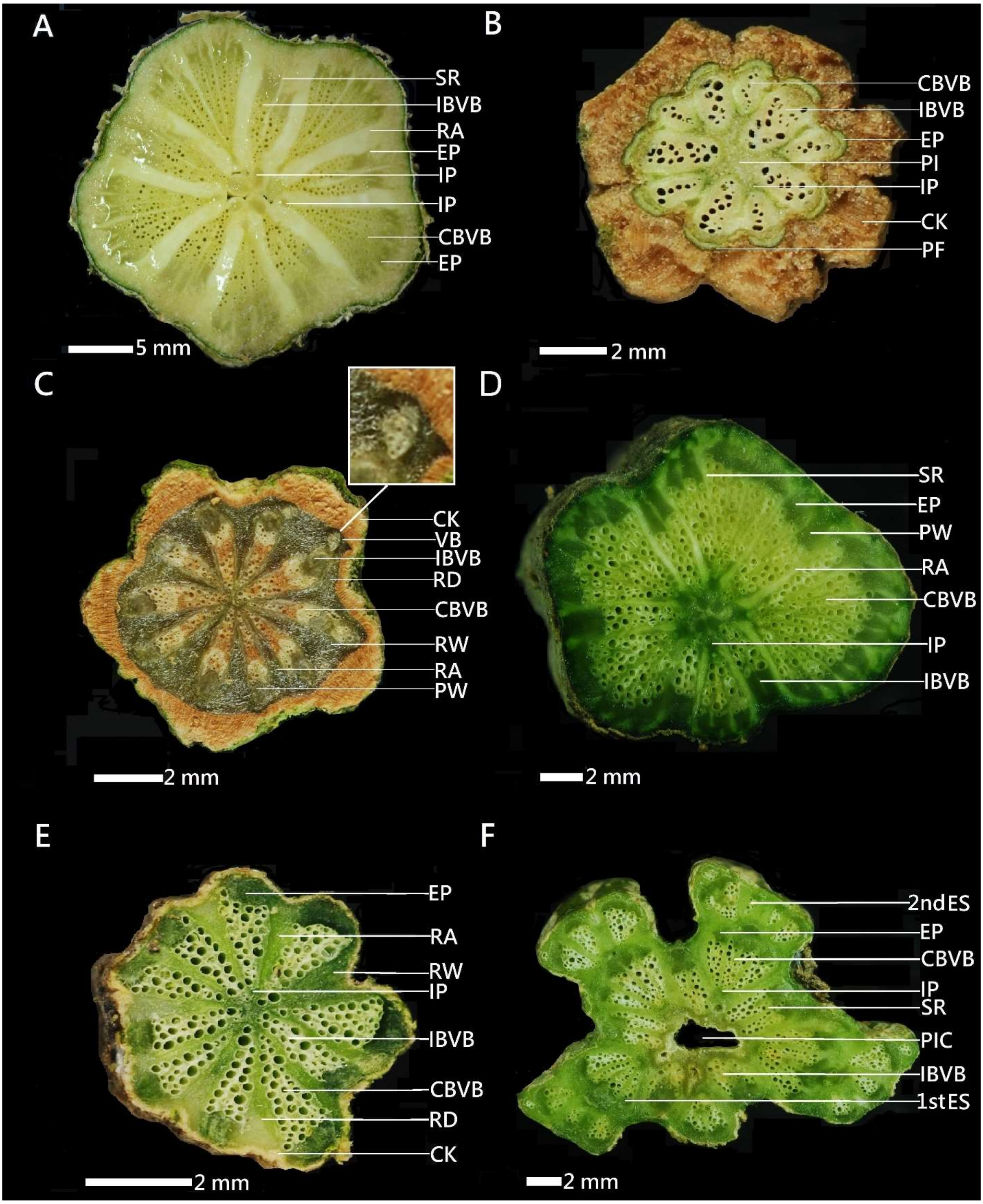


**Fig. 1.** The cambial variants of some Cucurbitaceae plants. **A**. ***Coccinia grandis***, numerous secondary rays, and two wide rays which split the external cylinder of the cortical bicollateral vascular bundle. **B**. ***Gynostemma pentaphllum***, stem with a thick cork. **C**. ***G. pentaphllum***, two to three secondary vascular bundles developed, furrowed xylem of phloem wedges, ray wedges evident. **D**. ***Luffa aegyptiaca***, secondary rays numerous. **E**. ***Melothria pendula***, with wedge-like rays and ray dilatation. **F**. ***Momordica charantia***, stem penta-lobed. **Abbreviation**: CK: cork, PW: phloem wedge, RA: ray, RD: ray dilatation, RW: ray wedge, PI: pith, PIC: pith cavity, ES: external secondary vascular cylinder,1st ES: the first layer of external secondary vascular cylinder, 2nd ES: the second layer of external secondary vascular cylinder, VB: vascular bundle, EP: external phloem, IP: internal phloem, IBVB: inner bicollateral vascular bundle, CBVB: cortical bicollateral vascular bundle, SR: secondary ray, PF: phloem fibers.

**Table 1.** Stem characteristics of Taiwan Cucurbitaceae species.

| Scientific name | Habit (L/V) | SD (mm) | SS | CV | Cork (mm) | BVB (no.) | CBVB (no.) | IBVB (no.) | RAY (mm) | ARW (mm) | SR (+/−) | VD (μm) | AVD (μm) | SC (layers) | SV (+/−) | RW (+/−) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *Coccinia grandis* | L | 24 | PEN | VS,IC,PW | - | 10 | 5 | 5 | 0.90–2.30 | 1.30 | + | 79–244 | 169 | - | - | - |
| *Gynostemma pentaphllum* | L | 3, 8 | PEN | VS,IC,PW | 0.7 | 10 | 5 | 5 | 0.06–0.17 | 0.11 | - | 24–107 | 61 | - | + | + |
| *Melothria pendula* | L | 5 | PEN | VS,IC | 0.1 | 10 | 5 | 5 | 0.18–0.83 | 0.40 | - | 33–200 | 116 | - | - | + |
| *Momordica charantia* (1) | L | 12 | PEL | VS,IC,ES | - | 10 | 5 | 5 | 0.35–0.91 | 0.62 | + | 46–274 | 160 | - | - | - |
| *Momordica charantia* (2) | L | 12 | PEN | VS,IC,ES | 0.1 | 10 | 5 | 5 | 0.12–0.23 | 0.17 | + | 44–180 | 92 | - | - | - |
| *Momordica cochinchinensis* | L | 18, 44 | CYL | VS,PW,SC | - | 20 | 15 | 5 | 0.25–0.66 | 0.48 | - | 150–468 | 274 | 2 | - | - |
| *Neoalsomitra clavigera* | L | 6, 9 | CYL | VS,PW | - | 14 | 7 | 7 | 0.11–0.74 | 0.45 | - | 63–315 | 190 | - | - | - |
| *Luffa aegyptiaca* | V | 15 | PEN | VS,PW | - | 10 | 5 | 5 | 0.20–0.57 | 0.38 | + | 148–462 | 270 | - | - | - |
| *Mukia maderaspatana* | V | 5 | CYL | VS,PW | + | 10 | 5 | 5 | 0.13–0.53 | 0.31 | - | 33–327 | 152 | - | - | - |
| *Sinobaijiania taiwaniana* | V | 5 | PEN | IC | - | 12 | 5 | 7 | 0.01–0.49 | 0.24 | - | 92–340 | 183 | - | - | - |
| *Trichosanthes homophylla* | V | 5 | CYL | VS | 0.3 | 10 | 5 | 5 | 0.19–0.34 | 0.27 | - | 10–259 | 111 | - | - | - |
| *Trichosanthes ovigera* | V | 6 | CYL | VS | + | 10 | 5 | 5 | 0.26–0.57 | 0.41 | - | 34–403 | 219 | - | - | - |
| *Zehneria mucronata* | V | 8 | CYL | VS | - | 10 | 5 | 5 | 0.65–1.09 | 0.82 | - | 116–334 | 217 | - | - | - |

**Note**: L: liana; V: herbaceous vine; SD (mm): stem diameter; SS: stem shape; PEN: pentagonal, PEL: penta-lobed, CYL: cylindrical or tri-lobed; CV: cambial variants; IC: stem cambium normal in production but irregular conformation; VS: axial vascular elements in segments; PW: furrowed xylem of phloem wedge; ES: external secondary vascular cylinder; SC: successive cambia; BVB no.: bicollateral vascular bundle number; CBVB no.: cortical bicollateral vascular bundle number; IBVB no.: inner bicollateral vascular bundle number; RAY (mm): ray min-max width (mm); ARW (mm): average rays width (n = 10) (mm); SR (+/−): secondary rays; VD (mm): vessel min-max diameter (mm); AVD (μm): average vessels diameter (n = 25) (μm); SC (layers): number of successive cambia; SV (+/−): secondary vascular bundle; RW (+/−): ray wedge; (+/−): present/absent.

vascular elements divided in radial segments by broad rays of ground tissue combined with irregular conformation and furrowed xylem of the phloem wedges. The diffuse-porous vessels have a diameter of 24–107 μm, and an average diameter of 61 μm, with a compressed pith.

c. ***Melothria pendula***: the cross-section of the stem is pentagonal in shape, wavy; cork thickness is approximately 0.1 mm, and the bark is white-green with vertical ridges. There are 10 bicollateral vascular bundles. The xylem segments are 0.6–1.0 mm wide, with an average of 0.75 mm; rays are dilated and widened centrifugally with a width of 0.18–0.83 mm, and an average width of 0.4 mm, forming wedge-shaped rays (Fig. 1E). The cambial variants have axial vascular elements divided in radial segments by broad rays of ground tissue combined with an irregular conformation. The vessels are diffusely porous, with solitary and a few multiple pores, a diameter of 33–200 μm, with an average of 116 μm, a vasicentric parenchyma, and a compressed pith.

d-1. ***Momordica charantia***: the stem cross-section is pentalobe-shaped, and the bark is white-green with prominent vertical flakes. There are 10 bicollateral vascular bundles. The rays are approximately 0.35–0.91 mm wide, with an average of 0.62 mm; each xylem segment contains secondary rays. Two layers of external secondary vascular cylinder are formed from the cortical bicollateral vascular bundle. The cambial variant types are axial vascular elements divided in radial segments by broad rays of ground tissue combined with irregular conformation and external secondary vascular cylinder (Fig. 1F). The vessels were diffuse-porous with solitary and a few multiple pores, measuring 46–274 μm in diameter, with an average of 160 μm, and with a pith cavity.

d-2. ***Momordica charantia***: the stem cross-section is pentagonal, the cork thickness is approximately 0.1 mm, and the bark is white-green with prominent vertical flakes. There are 10 bicollateral vascular bundles. The rays are approximately 0.12–0.23 mm wide, with an average of 0.17 mm. There are a few secondary rays present in each xylem segment. Two or three layers of external secondary vascular cylinder are formed from the cortical bicollateral vascular bundle (Fig. 2A). The cambial variant types have axial vascular elements divided in radial segments by broad rays of ground tissue combined with irregular conformations and external secondary vascular cylinder. The vessels are diffusely porous, containing solitary and a few multiple pores, measuring 44–180 μm in diameter, with an average of 92 μm. In addition, they have a vasicentric parenchyma and a compressed pith.

e. ***Momordica cochinchinensis***: the stem cross-section is cylindrical when the diameter reaches approximately 18 mm (Fig. 2B), and the bark is light brown with shallow depressesions arranged vertically like waves. The bicollateral vascular bundles are 20 and

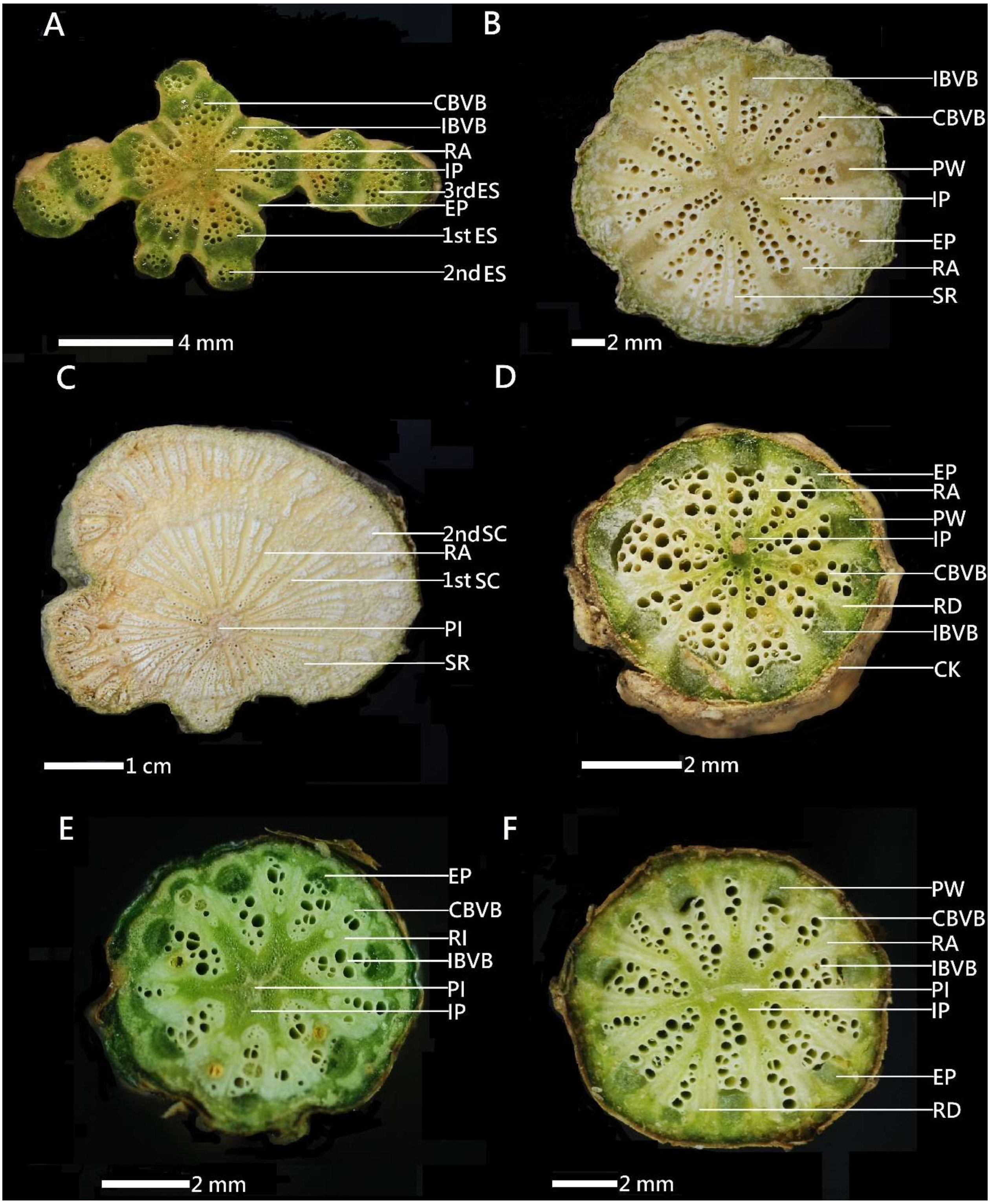


**Fig. 2.** The cambial variants of some Cucurbitaceae plants. **A**. ***Momordica charantia*** external secondary vascular cylinder. **B**. ***M. cochinchinensis***, four wider rays split the external cylinder of cortical bicollateral vascular bundles. **C**. ***M. cochinchinensis***, successive cambia. **D**. ***Mukia maderaspatana***, furrowed xylem of phloem wedges. **E**. ***Neoalsomitra integrifolia***, bicollateral vascular bundle 14. **F**. ***N. integrifolia***, ray dilatation. **Abbreviation**: PW: phloem wedge, EP: external phloem, RA: ray, RD: ray dilatation, IP: internal phloem, PI: pith, SC: successive cambia, 1st SC: the first layer successive cambia, 2nd SC: the second layer successive cambia, ES: external secondary vascular cylinder,1st ES: the first layer of external secondary vascular cylinder, 2nd ES: the second layer of external secondary vascular cylinder, 3rd ES: the third layer of external secondary vascular cylinder, IBVB: inner bicollateral vascular bundle, CBVB: cortical bicollateral vascular bundle, RI: ray initials, CK: cork, SR: secondary rays.

arranged in two cylinders. The inner cylinder has five bundles, the outer cylinder is divided in radial segments by four broad rays of ground tissue into 15 bundles. A two-layered successive cambium can be observed when the stem diameter is approximately 4.4 cm (Fig. 2C). The rays widen centrifugally and measure between 0.25 and 0.66 mm in width, on average measuring 0.48 mm. Cambial variant types are axial vascular elements divided in radial segments by broad rays of ground tissue combined with the furrowed xylem of phloem wedges and successive cambia. The vessels are diffuse-porous, with solitary and a few multiple pores, they range in diameter from 150 to 468 µm, with an average of 274 µm. The parenchyma is vasicentric, and the pith is pentagonal.

f. ***Neoalsomitra clavigera***: the stem cross-sectionh is cylindrical, and the bark is light-brown, vertically and shallowly depressed with dotted lenticels. There are 14 bicollateral vascular bundles in two rings (Fig. 2E-F). The ray dilatation is evident and the ray width is approximately 0.11–0.74 mm, with an average value of 0.45 mm. The cambial variant types are axial vascular elements divided in radial segments by broad rays of ground tissue combined with the furrowed xylem of phloem wedges. The vessels are diffuse-porous, with solitary and a few multiple pores ranging from 63 to 315 µm in diameter, with an average diameter of 190 µm. The parenchyma is vasicentric, and the pith is slightly heptagonal in shape.

**Herbaceous vines**

In this study, herbaceous vines included *Luffa aegyptiaca* (Fig. 1D), *Mukia maderaspatana* (Fig. 2D), *Sinobaijiania taiwaniana* (Fig. 3A), *Trichosanthes homophylla* (Fig. 3B), *Trichosanthes ovigera* (Fig. 3C), and *Zehneria mucronata* (Fig. 3D).

a. ***Luffa aegyptiaca***: the stem cross-section is pentagonal when the stem diameter is approximately 1.5 cm (Fig. 1D), and the bark is green-yellow with prominent vertical flakes. There are 10 bicollateral vascular bundles, numerous secondary rays are found in each xylem segment, and the secondary phloem is triangular and black in color. The cambial variant types are axial vascular elements divided in radial segments by broad rays of ground tissue combined with furrowed xylem of phloem wedges. Diffuse-porous vessels have single and a few multiple pores, measuring 148–462 µm in diameter, with an average diameter of 270 µm. Its parenchyma is vasicentric, and its pith is rounded.

b. ***Mukia maderaspatana***: the stem cross-section is cylindrical and slightly corked when the stem diameter is approximately 5 mm (Fig. 2D). The bark is white-yellow with dotted lenticels. Rays have a centrifugal and dilatant expansion, ranging from 0.13–0.53 mm in width, and the average width is 0.31 mm. The cambial variant types are axial vascular elements divided in radial segments by broad rays of ground tissue combined with the furrowed xylem of phloem wedges. Vessels are diffuse-porous, with solitary and a few multiple pores, measuring 33–327 µm in diameter, with an average of 152 µm. The parenchyma is vasicentric, and the pith is rounded.

c. ***Sinobaijiania taiwaniana***: the stem cross-section is pentagonal when the diameter of the stem is approximately 3.6–6.3 mm, and the bark is pink-green. Twelve bicollateral vascular bundles are embedded in the parenchyma tissue. Five cortical bicollateral vascular bundles are located on the angular side, and seven inner bicollateral vascular bundles are located near the pith (Fig. 3A). The external primary phloem, perivascular fibers, and collenchyma of vascular elements are distributed throughout the stem cross-section. The width of ray initials is 0.01–0.49 mm, with an average of 0.24 mm. The cambial variant has an irregular conformation. The vessels are diffusely porous with solitary and a few multiple pores. Their diameter ranges from 92–340 µm, with the average diameter is 183 µm; the pith is rounded and green in color.

d. ***Trichosanthes homophylla***: the stem cross-section is cylindrical, the gray-white bark is membranous with thin cork and dotted with black lenticels (Fig. 3B). There are 10 bicollateral vascular bundles, and rays are dilatated. Rays are approximately 0.19–0.34 mm wide, with an average width of 0.27 mm. The cambial variant is axial vascular elements divided in radial segments by broad rays of ground tissue. These vessels are diffuse-porous, have solitary and a few multiple pores, measuring 10–259 µm in diameter, with an average diameter of 111 µm, and are compressed at the pith.

e. ***Trichosanthes ovigera***: the stem cross-section is cylindrical, the bark is brown, membranous, with thin cork. There are 10 bicollateral vascular bundles, and ray dilatation is present. Ray widths range between 0.26 and 0.57 mm, with an average width of 0.41 mm. The cambial variant is axial vascular elements divided in radial segments by broad rays of ground tissue (Fig. 3C). The vessels are diffuse-porous, with solitary pores and a few multiple pores, a diameter ranging from 34 to 403 µm, with an average diameter of 219 µm, and a compressed pith.

f. ***Zehneria mucronata***: the stem cross-section is cylindrical, the bark is gray-white, membranous, with a few dotted lenticels. There are 10 bicollateral vascular bundles (Fig. 3 D), and the rays are wide with evident dilatation; rays are 0.65–1.09 mm in width with an average width of 0.82 mm. The cambial variant is axial vascular elements divided in radial segments by broad rays of ground tissue. The vessels are diffusely porous, with solitary and a few multiple pores. Their diameters range from 116 to 334 µm, with an average of 217 µm, and their piths are compressed.

Stem cambial variants of the Taiwan Cucurbitaceae climbers were identified using the following the bracket keys.

1. Bicollateral vascular bundle numbers 10–20 …..………..……….. 2
1. Bicollateral vascular bundle numbers 10 ……….………………..… 4

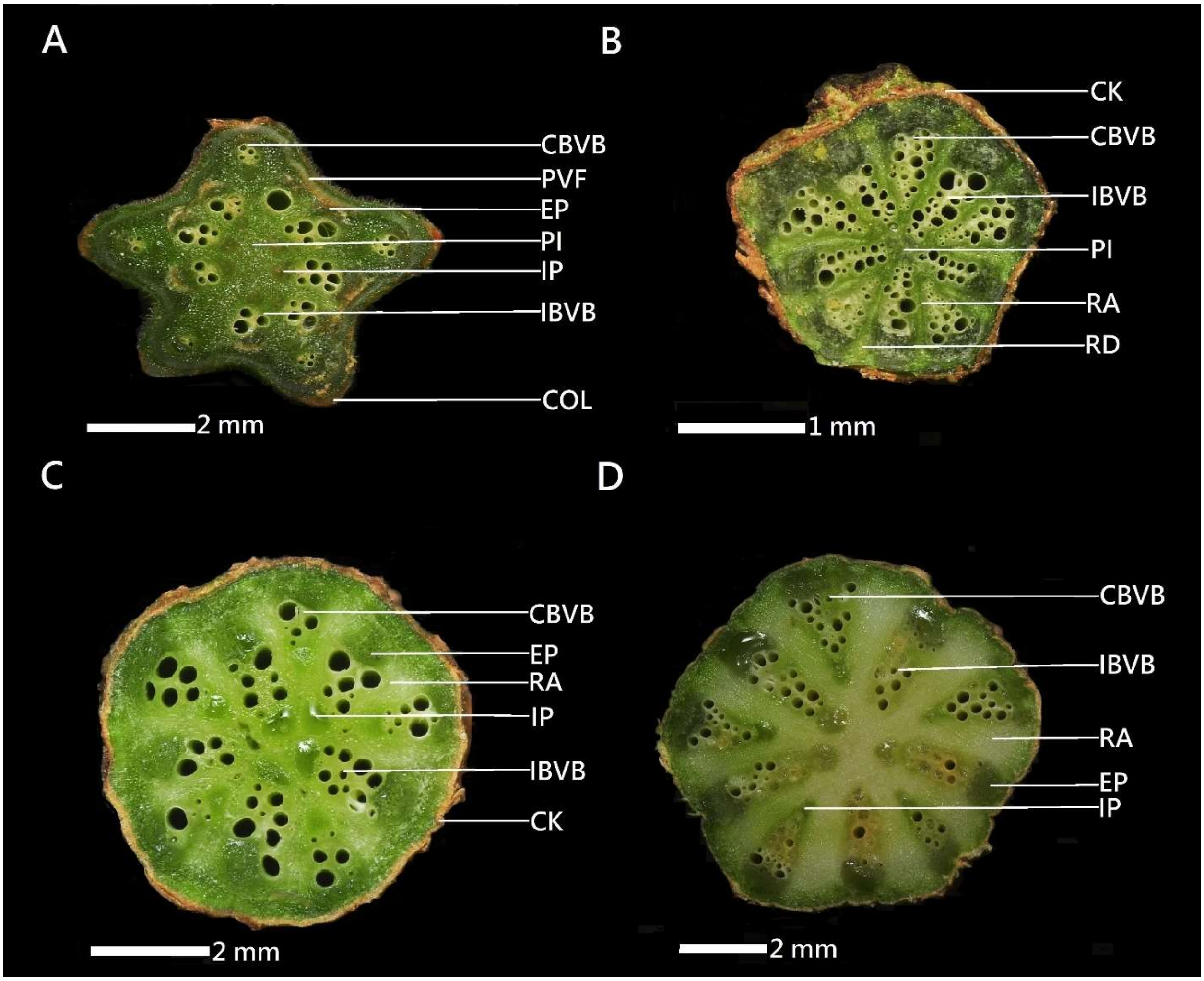


**Fig. 3.** The cambial variants of some Cucurbitaceae plants. **A**. ***Sinobaijiania taiwaniana***, external cylinder of cortical bicollateral vascular bundle five, inner bicollateral vascular bundle seven. **B**. ***Trichosanthes homophylla***, the external cylinder of the cortical bicollateral vascular bundle is split into two rays. **C**. ***T. ovigera***, the external cylinder of cortical bicollateral vascular bundle is divided into two wide rays. **D**. ***Zehneria mucronata***, ray dilatation evident. **Abbreviation**: EP: external phloem, RA: ray, RD: ray dilatation, IP: internal phloem, PI: pith, IBVB: inner bicollateral vascular bundle, CBVB: cortical bicollateral vascular bundle, PVF: perivascular fibers, COL: collenchyma.

2. Bicollateral vascular bundle numbers 20 …………………………… ………………………………………… ***Momordica cochinchinensis***
2. Bicollateral vascular bundle numbers 12 or 14 …………………… 3
3. Bicollateral vascular bundle numbers 14 …... ***Neoalsomitra clavigera***
3. Bicollateral vascular bundle numbers 12 ... ***Sinobaijiania taiwaniana***
4. With axial vascular elements in segments only ………………… 5
4. With two or three cambial variants types ………………………… 7
5. Without cork …………………………………… ***Zehneria mucronata***
5. With cork ……………………………………………………… 6
6. Bark gray-white with black dotted lenticels ………………………… ………………………………………… ***Trichosanthes homophylla***
6. Bark brown with few brown dotted lenticels ………………………… ……………………………………………… ***Trichosanthes ovigera***
7. With two cambial variants types ……………………………………… 8
7. With three cambial variants types ……………………………………… 9
8. With ray wedges ……………………………… ***Melothria pendula***
8. Without ray wedges ………………………… ***Mukia maderaspatana***
9. Stem with external secondary vascular cylinder ……………………… ……………………………………………… ***Momordica charantia***
9. Stem with furrowed xylem of phloem wedges ………………………. 10
10. Secondary rays none; with cork …....… ***Gynostemma pentaphllum***
10. Secondary rays numerous; without cork ……………………… 11
11. Bark gray-brown and membranous with dotted lenticels ………… ………………………………………………… ***Coccinia grandis***
11. Bark green-yellow with prominent vertical flakes ………………… …………………………………………………… ***Luffa aegyptiaca***

## DISCUSSION

The number of bicollateral vascular bundles showed three patterns (Table 1): 1. Ten vascular bundles type: in most species, both the outer and inner cylinders were composed of five bundles each, and two cylinders were split by two broad rays, for example, *C. grandis* (Fig. 1A). 2. Fourteen vascular bundles type: both the outer and inner cylinders are composed of seven bundles each. Each cylinder is split by two broad rays, such as in *Neoalsomitra clavigera* (Fig. 2E, 2F). 3. Twenty vascular bundles type: the outer and inner cylinders consist of 15 and five bundles, respectively. The cylinders are divided by four wide rays, such as in *M. cochinchinensis* (Fig. 2B, 2C). *Sinobaijiania taiwaniana* (Fig. 3A) had twelve bicollateral vascular bundles in the younger stem, of which five are outer cylinders, and seven are inner cylinders. The number of outer and inner cylinders changed in older stems must be observed and determined continuously.

The species *Coccinia indica* developed new secondary rays in the fascicular region of the cambium. The secondary xylem was arranged in plates (Patil *et al*., 2011), also called as axial vascular elements in segments (Angyalossy *et al*., 2016). In addition, the interfascicular region of the cambium continued to expand centrifugally, resulting in discontinuities cambium. Furthermore, the secondary xylem was arranged in plates in *C. grandis* (Carlquist, 1992b). In the present study, the secondary rays of *C. grandis* were found in each xylem segment and the rays widened approximately 1.3 mm. These features were consistent with those of Patil *et al*. (2011). *G. pentaphyllum* exhibited three secondary vascular bundles near the ray dilatation region, which originated from parenchymal proliferation. How much secondary vascular bundles will develop and form concentric rings or external secondary vascular cylinders are needed to study.

The outer cylindr of the cortical bicollateral vascular bundles of *M. charantia* had cambium, forming external secondary vascular cylinders (Fig. 2A). In addition, *M. cochinchinensis* showed continuous successive cambia (Fig. 2C) in the larger stem, approximately 4.4 cm in diameter. The external secondary vascular cylinders and successive cambia types exhibited by these three *Momordica* species were diagnostic features to distinguish the other Cucurbitaceae species. The distribution of these unique cambial variant types should be determined by examining the mature stems of additional *Momordica* species. *N. clavigera* had axial vascular elements in segments combined with the furrowed xylem of phloem wedges and pronounced ray dilatation (Fig. 2D).

The early stage of *Clematis grata* developed phloem fibers in dense bundles beneath the angular extension of the stem (Yang *et al*. 2021) when the stem diameter was approximately 3.5 mm. *S. taiwaniana* developed primary phloem, perivascular fibers, and collenchyma, similar to the developmental structures of *C. grata* in the earlier stage (Fig. 3A). Therefore, the later stage of *S. taiwaniana* has ten or more bicollateral vascular bundles warrants further study.

The rays of some Cucurbitaceae species dilated in a centrifugal direction, resulting in a compressed pith. The outer phloem, located in the inner cylinder of the bicollateral vascular bundle, usually developed the furrowed xylem of phloem wedges, such as in *M. cochinchinensis* and *N. clavigera*. In this study, the genus *Momordica*, which included three species, exhibited external secondary vascular cylinders and successive cambia, these results were consistent with Beck (2011) suggestion that cortical vascular bundles are one of the structures that result in irregular stem structures, such as successive cambia or furrows xylem of phloem wedges.

In summary, most species of Cucurbitaceae in Taiwan can be identified by the number of bicollateral vascular bundles, number of broas rays within the bicollateral vascular bundle, number of secondary rays, furrowed xylem of phloem wedges, ray dilatation, and ray wedges. The developmental processes of stem vascular elements are beneficial for understanding the changes in the vascular bundles of lianas. Sampling different stem sizes and fresh stems to investigate the development of each lianas are more important. The cambial variant types were more prevalent in rosids and asterids (Stevens, 2001), and the furrowed xylem type was an effective cambial variant for identifyingof climbers. We added the cambial variants of selected Cucurbitaceae species here will be available to assess the relationships among the order and families of climbers for academic study. The presence of multiple cambial variant types may also be indicative of taxonomic values.

## CONCLUSION

The bicollateral vascular bundles of Cucurbitaceae are diagnostic characteristic, and the secondary growth of cambia are diverse. Approximately five cambial variants types found in twelve Cucurbitaceae species were irregular conformation, axial vascular elements in segments, furrowed xylem of phloem wedge, external secondary vascular cylinder, and successive cambia. Most lianas exhibited a combination of two or three cambial variants. Based on the division of two or four broad rays and numbers of ten, fourteen, and twenty bicollateral vascular bundles, can be classified. These cambial variants and the three types of vascular bundles provide further evidence for the taxonomy of the Cucurbitaceae family. The presence of secondary rays, different developmental stages, and phloem wedges or ray wedges are also significant available for identifiying the Cucurbitaceae family. Cambial variants of Taiwan's Cucurbitaceae climbing plants will provide information on establishing cambial variants as common taxon-specific charactersistics.

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