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Research Article

Morphological Characterization of Citrus Species in Sri Lanka

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Abstract

Citrus fruits belong to the Family Rutaceae and are known for their high nutritional and medicinal value due to the presence of various bioactive compounds such as alkaloids, tannins, and phenols. In Sri Lanka, the available *Citrus* germplasm has not been thoroughly characterized, and proper taxonomical classification has yet to be conducted. Therefore, the primary objective of this study is to perform a morphological characterization of 25 samples of *Citrus* species found in Sri Lanka. The morphological characterization involved the preparation of herbarium sheets and the comparison and authentication of these Seventy-four (74) morphological traits of *Citrus* species were examined and documented using ten replicates per each species, including twelve (12) tree morphological traits, fifteen (15) flower morphological traits, fifteen (15) leaf morphological traits, twenty-four (24) fruit morphological traits, and eight (8) seed morphological traits according to the International Plant Genetic Resource Institute descriptors for *Citrus* (IPGRI,1999). Quantitative data were standardized using z-score normalization, while qualitative data were encoded with numerical values. The distances between species were calculated using Gower distance in the R programming environment. A dendrogram was constructed using the Ward's method of hierarchical clustering in IBM SPSS analytical software. The resulting dendrogram revealed five main clusters based on morphological characteristics. Cluster I: Calamansi, Yak dehi (ARS), Yak dehi (Provincial name: Ambul naran), Kumquat, Tahiti lime, Heen dehi (key lime), Nas naran (ARS), Nas naran (Home garden) and Kaffir lime. Cluster II: Lemon dehi (round lemon), Pochchi dehi (long lemon), Peni kudalu orange, Rough lemon, and Trifoliate orange. Cluster III: Heen naran, Ambul dodam (sour orange), Pani dodam (sweet orange), Jama naran, and Philippine red lime. Cluster IV: Citron, Buddha's hand, and Citron hybrid, which are part of a smaller cluster that merges with the cluster formed by Grapefruit and Sidaran. Cluster V: Pomelo, which is distinctly different from the other clusters.

Morphological traits provide valuable insights into the diversity and taxonomy of *Citrus* species. A systematic characterization and documentation of morphological traits could establish a foundation for accurate species identification, classification, and germplasm conservation and supports breeding programs aimed at developing improved *Citrus* varieties.

Keywords: Citrus Germplasm; Dendrogram; Morphological Traits; Rutaceae; Sri Lanka

Abbreviations

ARS: Agricultural Research Station; TH: Tree Height; FA: Flower Arrangement; SIN: Single; FLR: Inflorescence; CR: Circumference; FP: Flower Position; AXI: Axillary; TER: Terminal; PT: Plant Type; TR: Tree; SH: Shrub; FT: Flower type; PRF: Perfect; IMP: Imperfect; PS: Plant shape; EL: Ellipsoid plant; SP: Spheroid Plant; OB: Obloid Plant; STC: Stigma Color; GH: Growth Habit; ER: Erect; SR: Spreading; DR: Drooping; SYC: Style Color; BA: Branch Angle; N: Narrow; M: Medium; W: Wide; FS: Fruit Shape; SPD: Spheroid; ELD: Ellipsoid; PYF: Pyriform; OBL: Obloid; OVD: Ovoid; BD: Branch Density; D: Dense; FBS: Shape of Fruit Base; NK: Necked; CNX: Convex; TRC: Tunicate; CNC: Concave; CC: Concave Collared; CN: Concave Collared Neck; S: Spines; FAS: Shape of Fruit Apex; MMF: Mammiform; ACU: Acute; ROD: Rounded; TRC: Tunicate; DPS: Depressed;

SD: Spine Density; ECI: Epicarp Color (immature); SL: Spine length; ECM: Epicarp Color (mature); DYW: Dark Yellow; STC: Shoot Tip Color; FL: Fruit Length; STS: Shoot Tip Surface; GB: Glabrous; IN: Intermediate; PB: Pubescent; FD: Fruit Diameter; LL: Leaf Length; ET: Thickness of Epicarp; LW: Leaf Width; MT: Thickness of Mesocarp; PO: Petiole; P: Present; AB: Absent; AD: Diameter of Fruit Axis; POS: Petiole shape; OBC: Obcordate; OBD: Obdeltate; OBV: Obovate; FAS: Shape of Fruit Axis; ROU: Rounded; IRG: Irregular; POL: Petiole Length; NFA: Nature of Fruit Axis; HLW: Hollow; SLD: Solid; POW: Petiole Width; NSE: Number of Segment; LD: Leaf Division; SI: Simple; BI: Bifoliate; TRI: Trifoliate; NAS: Nature of Segments; UNF: Uniform; NUF: Not Uniform; LV: Leaf Variegation; P: Present; AB: Absent; LVE: Length of Vesicles; LLA: Leaf Laminar Attachment; SSL: Sessile; BRP: Brevipetiolate; LOP: Longi petiolate; WVE: Width of Vesicles; AOB: Angle of Base; ROU: Rounded; CUN: Cuneate; OBT: Obtuse; EP: Ease of Peeling; E: Easy; HD: Hard; AA: Angle of Leaf Apex; CUN: Cuneate; OBT: Obtuse; ROU: Rounded; ACM: Acuminate; ABS: Adherence Between Segments; LS: Laminar Shape; ELP: Elliptic; OVT: Ovate; OBO: Obovate; LAC: Lanceolate; ORB: Orbicular; FPC: Fruit Pulp Color; LM: Laminar Margin; CRE: Crenate; DEN: Dentate; ENT: Entire; SNT: Sinuate; MC: Mesocarp Color; PNK: Pink; RED: Red; JPL: Junction Between Petiole and Laminar; FU: Fused; ARC: Articulate; PF: Pulp Firmness; L: Low; M: Medium; H: High; LC: Leaf Color; LGR: Light Green; DGR: Dark green; NFS: Nature of Fruit Surface; SMT: Smooth; RUG: Rough; PTD: Pitted; PPL: Papillate; BUP: Bumpy; PL: Petal Length; JC: Juice Color; WHT: White; GR: Green; YW: Yellow; ORG: Orange; PW: Petal Width; PCU: Pulp Color Uniformity; AL: Anther Length; SEN: Number of Seeds; PDL: Pedicel Length; SEL: Length of Seed; CL: Calyx Length; SEW: Width of Seed; FC: Color of Open Flower; PP: Purple; SES: Seed Shape; FUF: Fusiform; CLV: Clavate; CUF: Cuneiform; OVD: Ovoid; SDE: Semi Deltoid; SPD: Spheroid; SSD: Semi- Spheroid; AC: Color of Anther; LYW: Pale Yellow; SEU: Seed Surface; SMT: Smooth; WRK: Wrinkled; HRY: Hairy; NS: Number of Stamen; SEC: Seed Color; CRM: Cream; BW: Brown; NP: Number of Petals; SCC: Cotyledon Color; AS: Anther Length Respect to Stigma; SHT: Shorter; L: Length; SEE: Seed Embryony; MONO: Mono Embryonic; POLY: Poly Embryonic

Introduction

Citrus is one of the most economically and medicinally significant plant genera in the world. It belongs to the subfamily Aurantioideae (Citroideae) within the family Rutaceae. It is believed that *Citrus* originated in Southeast Asia [1] and was cultivated in China as early as 2500 BC [2]. *Citrus* fruits are primarily enjoyed as fresh produce or juices [3]. *Citrus* contains many essential phytochemicals, such as ferulic acid, hydrocinnamic acid, cyanidin glycoside, hesperidin, naringin, polymethoxylated flavones, vitamin C, and carotenoids [4-6]. These compounds exhibit a range of health benefits, including antioxidant, anti-inflammatory, anti-cancer, anti-tumor, anti-fungal, anti-bacterial, and blood clot inhibition activities [7,8]. They act as free radical scavengers, modulating enzymatic activities and providing preventive and protective effects against various diseases.

Specifically, *Citrus* has been associated with the prevention of scurvy [9], the formation of kidney stones [10], and hypertension by interfering with calcium channel blockers [11]. Additionally, these compounds may help reduce the risk of cancer [12,13], coronary disease, chronic asthma [14], diabetes [15], hyperlipidemia [16], liver dysfunction [17], atherosclerosis [18], and osteoporosis [19]. Furthermore, *Citrus* can improve collagen formation, iron absorption, and immune system functions [20,21].

Citrus fruits are also integral to culinary traditions worldwide. They are used to enhance the flavour and aroma of a wide range of dishes and beverages [22]. *Citrus* essential oils, extracted from peels, are used in aromatherapy, cleaning products, and perfumes [23]. Additionally in many cultures, *Citrus* fruits are used in traditional medicine to treat ailments such as colds and digestive issues [24].

Moreover, *Citrus* by-products have industrial applications, including the production of pectin from peels, which is used as a gelling agent in food processing [25]. Bioflavonoids extracted from *Citrus* are used in dietary supplements and pharmaceuticals for their health-promoting properties [26].

Traditionally, the classification of *Citrus* species has relied on morphological characteristics and geographical data. The classification system proposed by [27] recognized 16 *Citrus* species, while [28] identified 162 species. Both systems have gained widespread acceptance. The genetic diversity of *Citrus* species is notably high, leading to complications and controversies in their taxonomy and phylogeny [2]. This complexity arises from factors such as somatic bud mutations, polyploidy [2], and the high sexual compatibility

among various species and related genera like Fortunella, Poncitrus, and Atalantia [29-31]. Recent research has indicated that there are only three true species of Citrus: Citron (*Citrus medica*), Mandarin (*Citrus reticulata*), and Pummelo (*Citrus maxima*). All other *Citrus* species can be categorized as inter-species and intra-species hybrids [32].

Sri Lanka is very famous for superior quality *Citrus* species from ancient times and is being used as fresh fruits, fresh juices, and in Citrus-based industries and Ayurveda and traditional system of medicines for the treatment of an array of aliments. There are about 15 to 20 *Citrus* species in Sri Lanka which belong to four main groups as Oranges, Mandarins, Acid members, and Giant members. Table 1 shows the available *Citrus* species in Sri Lanka.

Citrus group	Botanical name	Common name
Oranges	*Citrus aurantium*	Ambul dodam (sour orange)
Oranges	*Citrus sinensis*	Pani dodam (sweet orange)
Oranges	*Citrus trifoliata*	Trifoliate orange
Oranges	*Citrus* spp.	Peni kudalu orange
Mandarins	*Citrus crenatifolia*	Heen naran
Mandarins	*Citrus reticulata*	Jama naran
Mandarins	*Citrus microcarpa*	Calamansi
Mandarins	*Citrus japonica*	Kumquat
Mandarins	*Citrus japonica*	Nas naran/Odu dehi
Acid members	*Citrus aurantiifolia*	Heen dehi (key lime)
Acid members	*Citrus latifolia*	Tahiti lime
Acid members	*Citrus hystrix*	Kaffir lime
Acid members	*Citrus limonia*	Philippine red lime
Acid members	*Citrus medica*	Citron
		Sidaran/Maha rata dehi
		Buddha's hand
Acid members	*Citrus limon*	Pochchi dehi (long lemon)
		Lemon dehi (round lemon)
Acid members	*Citrus jambhiri*	Rough lemon
Acid members	*Citrus* spp.	Yak dehi/Ambul naran
Giant members	*Citrus maxima*	Pomelo
Giant members	*Citrus paradisi*	Grapefruit

Table 1: *Citrus* species in Sri Lanka.

Sweet oranges are primarily cultivated in the dry and intermediate zones of Sri Lanka. They are widely consumed as fresh fruits and used in juice production. The variety 'Bibila Sweet Orange' is well-known for its sweetness and juicy flesh [33]. Mandarins are grown in the up-country and mid-country regions, benefiting from cooler climates. They are popular as fresh fruits and often used in traditional sweets and beverages. Mandarin varieties like Indu, Madu and Rahangala juicy are popular for their flavour, ease of peel and less seed content. Lemons are extensively grown in home gardens and commercial plantations across the island. Lemons used in culinary applications, beverages, and traditional medicine. The 'Sri Lankan Lemon' variety is renowned for its high juice content and strong aroma [34]. It has two types: long lemon type and round lemon type. Key limes are commonly grown in the coastal and low-country areas. Key limes are essential in local cuisine for flavouring dishes, making pickles, and in beverages [35]. Sour oranges are grown in both home gardens and small-scale plantations. Used in making marmalade, essential oils, and traditional remedies. The variety 'Seville Orange' is often used in culinary and medicinal preparations [36].

Apart from those well-known species, many other *Citrus* species are also found in Sri Lanka. *Citrus medica* (Citron) is cultivated in home gardens and some commercial plantations, *Citrus japonica* (Kumquat) is cultivated in home garden some time as ornamental plants, *Citrus maxima* (Pomelo) is cultivated specially in the wet zone, *Citrus hystrix* (Kaffir lime) which is cultivated in dry zone is another *Citrus* species found in Sri Lanka.

Recent studies conducted using Sri Lankan *Citrus* germplasm revealed the presence of bio-active molecules [37], antioxidant properties [38,39], anti-bacterial, anti-fungal properties [40,41], and anti-inflammatory properties [42], in *Citrus* species in Sri Lanka. Another study which was conducted using 11 *Citrus* species revealed that Sri Lankan *Citrus* have industrially desirable morphological and biochemical traits according to their relevant industries [40]. An additional study based on sweet oranges (*Citrus sinensis*) varieties identified some new superior varieties like arogya, bibile sweet, and sisila with high juice volume, high sweetness, lesser peel thickness which enhance the consumer preference [43].

Citrus species in Sri Lanka face challenges from pests such as *Citrus psyllid* and diseases such as *Citrus* greening [44]. The changes in climate pattern also affecting *Citrus* production, with increased incidences of droughts and irregular rainfall impacting yield and quality. However, Breeding programs aimed at developing disease-resistant and climate-resilient *Citrus* varieties are underway [35].

Citrus species play a significant role in the agricultural landscape of Sri Lanka, with diverse varieties adapted to various climatic conditions across the island. Despite challenges such as pest management and climate change, opportunities for genetic improvement and export potential hold promise for the future. Continued research and sustainable cultivation practices are essential to enhance *Citrus* production and ensure its contribution to the economy and food security.

Morphological traits such as fruit size, shape, color, leaf characteristics, and tree structure provide valuable insights into the diversity and taxonomy of *Citrus* species. By systematically characterizing and documenting these traits, it is possible to establish a foundation for accurate species identification, classification, and germplasm conservation. This process also supports breeding programs aimed at developing improved *Citrus* varieties. Traits such as fruit size, shape, color, and taste are essential targets for breeding efforts to create new *Citrus* varieties with enhanced agronomic characteristics and consumer appeal. Through morphological characterization, breeders can identify promising parental lines and select desirable traits. This approach contributes to the sustainability, productivity, and profitability of *Citrus* cultivation in Sri Lanka while meeting the evolving needs of both local and international market.

Materials and Methods

Sample collection

Samples were collected from the Agricultural Research Station (ARS) in Maduruketiya, Monaragala (46V7+4W5), plant nurseries and home gardens in the Kurunegala (M8CH+PW), Gampaha (53WV+FV5), and Colombo Districts (VVQR+77X) in Sri Lanka. For the preparation of herbarium sheets, parts of the plants were collected, ensuring they included well-bloomed flowers and fruits [45]. Care was taken to avoid diseased or infected plants, as well as deformed or distorted twigs. After collection, the specimens were placed in plastic bags and labeled with specimen tags that included a collection number for all specimens, and tags with the botanical name and author for those that were confirmed. A field record was maintained for each specimen, noting the date of collection, collection number, location (GPS coordinates), microhabitat, a description based on visual observation, the specimen's name (if possible), and details on floral parts that might change upon drying. Additionally, close-up images of the trees, flowers, fruits, and leaves were taken for further identification and confirmation.

Herbarium sheet preparation

Herbarium sheets were prepared following the guidelines provided by [45], with some modifications. After collecting, specimens were treated with 10% formalin by enclosing them in polythene bags and applying formalin spray, followed by sealing the bags tightly. Thereafter, samples were kept within flimsies (newspapers). Lager specimens were mounted in V or N or M shape and leaves were mounted in the dorsoventral position. Extra leaves were removed to avoid overlapping of leaves, flowers, and other parts. Specimens kept inside flimsies were covered on either side by blotter papers and put in herbarium pressure to apply pressure by tightening the straps. During the drying period, blotters were replaced firstly within 1 to 2 days period, then within 2 to 3 days

periods, about a week. Re-pressing was done during that time. After drying, specimens were taken to the National Herbarium, Royal Botanical Gardens, Peradeniya. Dried specimens were mounted on standard-size herbarium papers (29 x 43cm). Each specimen was labeled with a rectangular-shaped main label (10 x 15 cm) in the right bottom corner of the herbarium sheet. Authentication processes of *Citrus* samples were done at the National Herbarium of the Royal Botanical Garden, Peradeniya.

Seventy-four (74) morphological characters of *Citrus* species were examined and documented using ten replicates per each species, including twelve (12) tree morphological traits, fifteen (15) flower morphological traits, sixteen (15) leaf morphological traits, twenty-four (24) fruit morphological traits, and eight (8) seed morphological traits according to the International Plant Genetic Resource Institute descriptors for *Citrus* [46]. Table 2 shows the Morphological characteristics of the *Citrus* species observed [46].

Morphological data were organized into matrix format, with rows representing *Citrus* sample and columns representing morphological traits. The data were submitted to analysis of variance (ANOVA) followed by post-hoc separation of means was done using Fischer's Protected Least Significant Difference (LSD) test at 5% probability [47], using Statistical Package for the Social Sciences (SPSS) software (SPSS Inc., Chicago).

Quantitative data were scandalized using z-score normalization and qualitative data were encoded with numerical value. Distances between species were calculated using Gower distance in the R program for mixed data. Dendrogram was constructed using Ward's method of hierarchical clustering in IBM-SPSS analytical software [48].

Results and Discussion

Morphological characterization was done for twenty-five (25) *Citrus* samples using twenty-two (22) quantitative and fifty-two (52) qualitative data. Table 3 shows the twelve (12) tree morphological traits. Table 4 shows the fifteen (15) leaf morphological traits. Table 5 shows the fifteen (15) floral morphological traits. Table 6 shows the eight (8) quantitative fruit morphological traits and Table 7 shows the sixteen (16) qualitative fruit morphological traits. Table 8 shows the eight (08) seed morphological traits.

Morphological trait	Observations/Measurements	Morphological trait	Observations/Measurements
Tree height (TH)	Measurements in cm	Flower arrangement (FA)	Single (SIN)/Inflorescence (FLR)
Circumference (CR)	Measurements in cm	Flower position (FP)	Axillary (AXI)/Terminal (TER)
Plant type (PT)	Tree (TR)/Shrub (SH)	Flower type (FT)	Perfect (PRF)/Imperfect (IMP)
Plant shape (PS)	Ellipsoid (EL)/Spheroid (SP)/Obloid (OB)	Stigma color (STC)	Yellow (YW)/Green (GR)
Growth habit (GH)	Erect (ER)/Spreading (SR)/Drooping (DR)	Style color (SYC)	Yellow (YW)/White (WHT)/Green (GR)/Purple (PP)
Branch angle (BA)	Narrow (N)/Medium (M)/Wide (W)	Fruit shape (FS)	Spheroid (SPD)/Ellipsoid (ELD)/Pyriform (PYF)/Obloid (OBL)/Ovoid (OVD)
Branch density (BD)	Less (L)/Medium (M)/Dense (D)	Shape of fruit base (FBS)	Necked (NK)/Convex (CNX)/Tunicate (TRC)/Concave (CNC)/Concave collared (CC)/Concave collared neck (CN)
Spines (S)	Present (P)/Absent (AB)	Shape of fruit apex (FAS)	Mammiform (MMF)/Acute (ACU)/ Rounded (ROD)/Tunicate (TRC)/Depressed (DPS)
Spine density (SD)	Absent (AB)/Low (L)/Medium (M)/ High (H)	Epicarp color (immature) (ECI)	Dark green (DGR)/Green (GR)/Light green (LGR)
Spine length (SL)	Measurements in cm	Epicarp color (mature) (ECM)	Green (GR)/Yellow (YW)/Dark yellow (DYW)//Red (RED) orange (ORG)
Shoot tip color (STC)	Green (GR)/Purple (PP)	Fruit length (FL)	Measurements in cm
Shoot tip surface (STS)	Glabrous (GB)/Intermediate (IN)/Pubescent (PB)	Fruit diameter (FD)	Measurements in cm
Leaf length (LL)	Measurements in cm	Thickness of epicarp (ET)	Measurements in cm

Leaf width (LW)	Measurements in cm	Thickness of mesocarp (MT)	Measurements in cm
Petiole (PO)	Present (P)/Absent (AB)	Diameter of fruit axis (AD)	Measurements in cm
Petiole shape (POS)	Obcordate (OBC)/Obdeltate (OBD)/ Obovate (OBV)	Shape of fruit axis (FAS)	Rounded (ROU)/Irregular (IRG)
Petiole length (POL)	Measurements in cm	Nature of fruit axis (NFA)	Hollow (HLW)/Solid (SLD)
Petiole width (POW)	Measurements in cm	Number of segment (NSE)	Count
Leaf division (LD)	Simple (SI)/Bifoliate (BI)/Trifoliate (TRI)	Nature of segments (NAS)	Uniform (UNF)/Not uniform (NUF)
Leaf variegation (LV)	Present (P)/Absent (AB)	Length of vesicles (LVE)	Measurements in cm
Leaf laminar attachment (LLA)	Sessile (SSL)/Brevipetiolate (BRP)/ Longipetiolate (LOP)	Width of vesicles (WVE)	Measurements in cm
Angle of base (AOB)	Rounded (ROU)/Cuneate (CUN)/ Obtuse (OBT)	Ease of peeling (EP)	Easy (E)/Medium (M)/Hard (HD)
Angle of leaf apex (AA)	Cuneate (CUN)/Obtuse (OBT)/ Rounded (ROU)/Acuminate (ACM)	Adherence between segments (ABS)	Low (L)/Medium (M)/High (H)
Laminar shape (LS)	Elliptic (ELP)/Ovate (OVT)/Obovate (OBO)/Lanceolate (LAC)/Orbicular (ORB)	Fruit pulp color (FPC)	White (WHT)/Yellow (YW)/Green (GR)/Orange (ORG)/Red (RED)
Laminar margin (LM)	Crenate (CRE)/Dentate (DEN)/Entire (ENT)/Sinuate (SNT)	Mesocarp color (MC)	White (WHT)/Greenish (GR)/Pink (PNK)/Red (RED)/Orange (ORG)
Junction between petiole and laminar (JPL)	Fused (FU)/Articulate (ARC)	Pulp firmness (PF)	Low (L)/Medium (M)/High (H)
Leaf color (LC)	Light green (LGR)/Green (GR)/Dark green (DGR)/Yellow (YW)	Nature of fruit surface (NFS)	Smooth (SMT)/Rough (RUG)/Pitted (PTD)/Papillate (PPL)/Bumpy (BUP)
Petal length (PL)	Measurements in cm	Juice color (JC)	White (WHT)/Green (GR)/Yellow (YW)/Orange (ORG)/Pink (PNK)
Petal width (PW)	Measurements in cm	Pulp color uniformity (PCU)	Uniform/Not uniform
Anther length (AL)	Measurements in cm	Number of seeds (SEN)	Count
Pedicel length (PDL)	Measurements in cm	Length of seed (SEL)	Measurements in cm
Calyx length (CL)	Measurements in cm	Width of seed (SEW)	Measurements in cm
Color of open flower (FC)	White (WHT)/Yellow (YW)/Purple (PP)	Seed shape (SES)	Fusiform (FUF)/Clavate (CLV)/Cuneiform (CUF)/Ovoid (OVD)/Semi deltoid (SDE)/Spheroid (SPD)/Semi-spheroid (SSD)
Color of anther (AC)	Yellow (YW)/Pale yellow (LYW)/ White (WHT)	Seed surface (SEU)	Smooth (SMT)/Wrinkled (WRK)/ Hairy (HRY)
Number of stamen (NS)	Count	Seed color (SEC)	White (WHT)/Cream (CRM)/Yellow (YW)/Brown (BW)/Green (GR)
Number of petals (NP)	Count	Cotyledon color (SCC)	White (WHT)/Cream (CRM)/Yellow (YW)/Brown (BW)/Green (GR)
Anther length respect to stigma (AS)	Shorter (SHT)/Medium (M)/Length (L)	Seed embryony (SEE)	Mono embryonic (MONO)/Poly embryonic (POLY)

Table 2: Morphological traits of *Citrus* species (IPGRI, 1999) and Observations/Measurements.

Citrus Sample	PH (cm)	CR (cm)	S	LS (cm)	PT	PS	GH	BD	BA	SD	STC	STS
C_01	766.4	32.4	P	0.64	TR	EL	ER	L	N	M	GR	GB
C_02	328.3	16.7	P	0.45	SH	OB	DR	M	M	H	GR	GB
C_03	694.6	38.5	P	0.25	TR	EL	ER	M	N	M	GR	GB
C_04	281.4	19.8	P	0.46	SH	OB	DR	H	W	H	GR	GB
C_05	253.7	42.3	P	0.28	TR	EL	ER	H	N	L	GR	GB
C_06	220.3	14.6	P	0.56	SH	OB	DR	L	W	L	PP	IN
C_07	667.7	44.8	P	0.48	TR	SP	SR	H	M	H	GR	GB
C_08	638.8	59.6	P	0.18	TR	SP	SR	M	M	L	GR	GB
C_09	806.3	62.7	P	0.16	TR	SP	SR	M	N	L	GR	PB
C_10	258.3	32.6	P	0.62	TR	OB	DR	M	M	M	GR	GB
C_11	174.6	16.2	P	0.49	TR	EL	DR	H	N	H	GR	GB
C_12	114.3	14.7	P	0.58	SH	OB	DR	L	W	L	PP	IN
C_13	289.6	53.3	P	0.45	TR	EL	ER	H	N	M	GR	GB
C_14	223.6	20.2	P	0.55	SH	OB	SR	H	M	L	GR	GB
C_15	622.2	43.2	P	0.43	TR	SP	ER	L	M	L	GR	GB
C_16	192.3	13.3	P	1.28	SH	OB	SR	H	W	L	GR	GB
C_17	410.3	18.3	P	3.54	TR	EL	ER	M	M	H	PP	GB
C_18	301.3	15.3	P	1.74	TR	SP	SR	L	W	H	PP	GB
C_19	568.6	49.8	P	0.82	TR	SP	ER	M	M	M	GR	GB
C_20	234.6	17.9	AB	0.00	SH	OB	DR	M	W	L	GR	GB
C_21	407.8	34.5	P	3.28	TR	EL	ER	L	M	H	GR	GB
C_22	263	16.6	P	1.63	TR	EL	SR	L	M	H	GR	GB
C_23	786	56.5	AB	0	TR	EL	ER	M	W	H	GR	GB
C_24	397	22.5	P	0.56	TR	EL	DR	L	W	L	PP	GB
C_25	205.3	15.8	P	0.42	TR	EL	ER	H	N	M	GR	GB

Table 3: Tree morphological data of 25 samples of *Citrus* species.

Citrus sample	LL (cm)	LW (cm)	LD	LV	LS	LM	AOB	AA	LC	LLA	JPL	PO	POL (cm)	POW (cm)	POS
C_01	9.2	4.5	SI	AB	ELP	CRE	OBT	ACM	DGR	BRP	ART	P	1.7	0.5	OBD
C_02	4.1	2.4	SI	AB	ELP	ENT	OBT	ACM	LGR	BRP	ART	P	0.3	0.1	OBD
C_03	3.4	2.2	SI	AB	ELP	ENT	OBT	ACM	DGR	BRP	ART	P	0.4	0.2	OBV
C_04	4.1	2.5	SI	AB	OVT	ENT	CUN	ACM	GR	BRP	ART	P	0.5	0.3	OBD
C_05	9.4	4.2	SI	AB	ORB	DEN	CUN	ROU	DGR	LOP	ART	P	4.6	4.1	OBV
C_06	5.9	3.6	SI	AB	OBV	CRE	CUN	OBT	LGR	SSL	FUS	AB	0	0	-
C_07	8.4	4.7	SI	AB	ELP	ENT	CUN	ACM	DGR	BRP	ART	P	1.5	0.4	OBD
C_08	9.3	4.8	SI	AB	ELP	CRE	CUN	ACM	GR	BRP	ART	P	1.8	0.6	OBC
C_09	9.8	5.3	SI	AB	ORB	SNT	OBT	ACM	DGR	BRP	ART	P	1.7	0.6	OBC
C_10	7.8	3.6	SI	AB	ORB	ENT	CUN	ATT	GR	BRP	ART	P	1.6	0.5	OBV
C_11	3.8	1.9	SI	AB	LAC	ENT	OBT	ACM	GR	BRP	ART	P	0.4	0.2	OBV
C_12	7.8	3.7	SI	AB	OBV	CRE	CUN	OBT	LGR	SSL	FUS	AB	0	0	-
C_13	3.2	1.7	SI	AB	ELP	ENT	OBT	ACM	LGR	BRP	ART	P	0.4	0.2	OBV
C_14	10.9	4.8	SI	AB	OVT	CRE	ROU	OBT	LGR	BRP	ART	AB	0	0	OBD

C_15	12.2	7.8	SI	AB	ORB	DEN	OBT	ACM	DGR	BRP	ART	P	2.9	0.4	OBD
C_16	5.5	2.8	SI	P	OVT	CRE	ROU	OBT	YW	BRP	ART	P	0.8	0.15	OBD
C_17	16.7	5.6	SI	AB	OBV	CRE	CUN	OBT	LGR	SSL	FUS	P	1.8	0.25	-
C_18	15.6	6.3	SI	AB	OBV	DEN	CUN	OBT	LGR	SSL	FUS	AB	0	0	-
C_19	7.8	3.2	SI	AB	ELP	CRE	OBT	ACM	GR	BRP	ART	P	0.7	0.1	OBD
C_20	7.5	3.6	SI	AB	OVT	SNT	OBT	ACM	GR	BRP	ART	P	0.6	0.1	OBV
C_21	10.4	4.8	SI	AB	OVT	SNT	OBT	ACM	GR	BRP	ART	P	1.2	0.2	OBV
C_22	7.3	4.2	TRI	AB	OVT	ENT	CUN	ATT	DGR	BRP	ART	P	1.4	0.2	OBV
C_23	6.4	2.8	SI	AB	OVT	ENT	ROU	ACM	LGR	BRP	ART	P	0.6	0.1	OBV
C_24	14.6	8.3	SI	AB	OBV	CRE	CUN	ATT	LGR	SSL	FUS	AB	0	0	-
C_25	4.6	2.8	SI	AB	ELP	ENT	ROU	ACM	DGR	BRP	ART	P	1.2	0.1	OBD

Table 4: Leaf morphological data of 25 samples of *Citrus* species.

Citrus sample	FC	NP	FA	FP	FT	PL (cm)	PW (cm)	AL (cm)	PDL (cm)	CL (cm)	NS	AS	STC	SYC	AC
C_01	WHT	5	FLR	AXI	PRF	1.5	0.39	0.7	0.36	0.4	4	LNG	YW	GR	YW
C_02	WHT	5	FLR	AXI	PRF	0.8	0.27	0.45	0.38	0.3	4	SRT	YW	WHT	YW
C_03	WHT	5	FLR	AXI	PRF	0.8	0.28	0.48	0.42	0.3	4	LNG	YW	WHT	YW
C_04	WHT	5	SIN	AXI	PRF	0.6	0.28	0.3	0.64	0.5	4	SRT	YW	PP	YW
C_05	WHT	5	SIN	AXI	PRF	0.6	0.26	0.3	0.62	0.5	5	SM	YW	YW	YW
C_06	PP	5	SIN	AXI	PRF	1.2	0.32	0.4	0.47	0.3	5	SM	YW	GR	YW
C_07	WHT	5	FLR	AXI	PRF	1.4	0.32	0.41	0.38	0.4	6	SRT	WHT	GR	YW
C_08	WHT	5	FLR	AXI	PRF	1.4	0.33	0.45	0.55	0.4	8	SM	YW	PP	YW
C_09	WHT	5	FLR	AXI	PRF	1.8	0.42	0.65	0.55	0.4	8	SRT	WHT	YW	YW
C_10	WHT	5	FLR	AXI	PRF	1.6	0.35	0.64	0.56	0.4	4	SM	YW	YW	YW
C_11	WHT	5	FLR	AXI	PRF	0.6	0.21	0.2	0.54	0.4	6	SM	YW	GR	YW
C_12	PP	5	SIN	AXI	PRF	1.2	0.29	0.52	0.48	0.3	4	SM	YW	GR	YW
C_13	WHT	5	FLR	AXI	PRF	0.6	0.22	0.21	0.66	0.4	2	SM	YW	WHT	YW
C_14	WHT	5	SIN	AXI	PRF	1.7	0.42	0.71	0.35	0.5	8	SM	YW	GR	YW
C_15	WHT	5	FLR	AXI	PRF	1.6	0.62	0.4	1.35	0.3	2	SRT	GR	WHT	YW
C_16	WHT	5	SIN	AXI	IMP	0.8	0.33	0.24	0.67	0.2	6	SM	WHT	YW	WHT
C_17	YW	5	FLR	AXI	PRF	1.8	0.74	0.35	1.73	0.6	6	SRT	YW	GR	YW
C_18	YW	5	FLR	AXI	PRF	2.2	0.32	0.4	1.74	0.6	4	SRT	YW	GR	YW
C_19	WHT	5	FLR	AXI	PRF	0.9	0.45	0.2	0.56	0.4	4	LNG	YW	WHT	YW
C_20	WHT	5	SIN	AXI	PRF	1.2	0.53	0.2	0.65	0.4	4	SRT	GR	YW	WHT
C_21	WHT	5	SIN	AXI	PRF	1.7	0.55	0.3	0.85	0.4	6	SM	YW	GR	YW
C_22	WHT	5	FLR	AXI	IMP	1.5	0.64	0.4	0.85	0.5	4	SM	YW	WHT	YW
C_23	WHT	5	FLR	AXI	PRF	1.6	0.34	0.4	0.76	0.5	4	SM	YW	WHT	YW
C_24	PP	5	FLR	AXI	IMP	1.9	0.46	0.78	0.48	0.3	5	SRT	YW	WHT	YW
C_25	WHT	5	FLR	AXI	PRF	0.7	0.24	0.5	0.51	0.3	4	SM	WHT	YW	YW

Table 5: Floral morphological data of 25 samples of *Citrus* species.

Citrus sample	FW (g)	FL (cm)	FD (cm)	ET (cm)	MT (cm)	AD (cm)	LVE (cm)	WVE (cm)
C_01	345	06.53	6.67	0.22	0.32	0.34	1.67	0.2
C_02	70	03.15	3.56	0.21	0.18	0.32	0.82	0.1
C_03	80	03.42	4.68	0.22	0.16	0.28	0.64	0.1
C_04	75	03.65	3.42	0.12	0.14	0.52	1.28	0.1
C_05	117	05.56	4.56	0.24	0.65	0.65	0.54	0.2
C_06	443	07.14	6.65	0.22	0.35	1.86	1.69	0.2
C_07	328	05.42	5.35	0.21	0.23	0.54	1.66	0.2
C_08	564	10.52	12.78	0.24	0.56	1.65	1.89	0.2
C_09	1756	16.22	14.98	0.24	2.52	2.42	2.68	0.3
C_10	423	06.24	6.14	0.21	0.24	0.38	1.55	0.1
C_11	45	03.21	3.25	0.12	0.12	0.22	1.24	0.2
C_12	568	13.34	7.34	0.23	0.84	1.88	1.84	0.2
C_13	53	03.33	3.45	0.12	0.15	0.18	0.87	0.1
C_14	45	03.43	1.83	0.12	0.34	0.34	0.94	0.2
C_15	986	14.82	15.1	0.28	0.32	0.67	2.13	0.3
C_16	48	03.56	4.21	0.20	0.23	0.24	0.96	0.2
C_17	827	18.23	10.43	0.25	3.89	2.34	1.98	0.2
C_18	791	17.34	10.21	0.22	3.45	2.21	1.76	0.2
C_19	87	03.59	4.29	0.21	0.19	0.24	0.68	0.1
C_20	77	03.62	3.32	0.12	0.12	0.55	1.21	0.1
C_21	420	07.23	6.56	0.22	0.32	1.82	1.72	0.2
C_22	329	06.28	6.69	0.21	0.32	0.32	1.65	0.2
C_23	212	05.54	5.35	0.22	0.62	0.54	0.52	0.2
C_24	786	17.56	10.28	0.25	1.45	2.98	1.95	0.2
C_25	39	03.48	3.87	0.21	0.19	0.31	0.83	0.1

Table 6: Fruit morphological quantitative data of 25 samples of *Citrus* species.

Citrus sample	FS	FBS	FAS	ECI	ECM	SFA	NFA	NSE	NAS	EP	ABS	FPC	MC	PF	NFC	JC
C_01	SPD	TRC	TRC	DGR	ORG	ROU	SLD	11	NUF	M	M	PGR	WHT	M	SMT	YW
C_02	SPD	CNX	TRC	DGR	DYW	ROD	SLD	12	NUF	E	E	PGR	WHT	L	SMT	YW
C_03	OBL	TRC	DPS	DGR	ORG	ROU	SLD	12	NUF	E	E	YW	WHT	M	SMT	YW
C_04	ELD	TRC	TRC	GR	YW	ROU	SLD	08	UNF	HD	HD	YW	WHT	M	SMT	YW
C_05	PYF	NK	DPS	DGR	YW	ROU	SLD	07	UNF	HD	HD	YW	WHT	M	RUG	YW
C_06	ELD	TRC	MMF	LGR	YW	IRG	HLW	11	UNF	HD	HD	YW	WHT	L	SMT	YW
C_07	SPD	CNX	ROD	GR	ORG	ROU	SLD	12	UNF	E	E	ORG	WHT	L	SMT	YW
C_08	OBL	TRC	DPS	GR	YW	IRG	HLW	12	NUF	M	M	YW	WHT	L	SMT	ORG
C_09	PYF	NK	DPS	GR	LGR	IRG	SLD	16	NUF	M	E	RED	PNK	H	RUG	PNK
C_10	SPD	CNX	TRC	DGR	ORG	ROU	SLD	12	NUF	M	E	YW	WHT	L	RUG	YW
C_11	SPD	TRC	TRC	DGR	YW	ROU	SLD	11	NUF	M	M	YW	WHT	M	SMT	YW
C_12	ELD	TRC	MMF	GR	YW	IRG	HLW	12	UNF	M	M	YW	WHT	M	SMT	YW
C_13	OBL	TRC	TRC	DGR	ORG	ROU	SLD	10	UNF	E	E	YW	WHT	M	SMT	YW

C_14	ELD	CNX	DPS	GR	ORG	ROU	SLD	05	UNF	HD	HD	ORG	WHT	L	SMT	ORG
C_15	SPD	CNX	TRC	GR	ORG	ROU	SLD	13	UNF	M	M	RED	PNK	M	SMT	PNK
C_16	ELD	CNX	TRC	GR	ORG	ROU	SLD	04	UNF	E	E	ORG	WHT	L	SMT	ORG
C_17	OVD	TRC	MMF	LGR	YW	IRG	HLW	11	NUF	HD	HD	YW	WHT	H	RUG	YW
C_18	OVD	TRC	MMF	LGR	YW	IRG	HLW	12	NUF	HD	HD	YW	WHT	H	RUG	YW
C_19	SPD	CNX	ROD	GR	YW	ROU	SLD	12	UNF	E	E	YW	WHT	L	SMT	YW
C_20	ELD	TRC	MMF	GR	YW	ROU	SLD	09	NUF	HD	HD	YW	WHT	M	SMT	YW
C_21	OVD	TRC	MMF	DGR	YW	ROU	SLD	10	NUF	M	M	YW	WHT	M	RUG	YW
C_22	SPD	CNX	TRC	GR	ORG	ROU	SLD	08	NUF	M	M	ORG	WHT	M	SMT	ORG
C_23	SPD	TRC	ROD	DGR	ORG	ROU	SLD	06	UNF	HD	HD	ORG	WHT	M	SMT	ORG
C_24	OVD	TRC	MMF	GR	YW	IRG	HLW	12	UNF	HD	HD	YW	WHT	M	RUG	YW
C_25	SPD	CNX	TRC	DGR	ORG	ROU	SLD	08	UNF	HD	HD	YW	WHT	M	SMT	YW

Table 7: Fruit morphological qualitative data of 25 samples of *Citrus* species.

Citrus sample	SEN	SEL (cm)	SEW (cm)	SES	SEU	SEC	SCC	SEE
C_01	16	1.34	0.45	CLV	WRK	CRM	GR	MONO
C_02	08	0.89	0.67	OVD	SMT	CRM	GR	MONO
C_03	18	1.16	0.34	CLV	SMT	CRM	CRM	BOTH
C_04	12	1.25	0.64	OVD	SMT	CRM	GR	MONO
C_05	08	1.36	0.45	FUF	WRK	WHT	YW	MONO
C_06	00	-	-	-	-	-	-	-
C_07	18	1.44	0.45	OVD	SMT	WHT	GR	BOTH
C_08	12	1.75	0.67	CLV	WRK	WHT	CRM	MONO
C_09	26	1.89	1.14	SDE	WRK	CRM	BW	MONO
C_10	12	1.25	0.43	OVD	SMT	CRM	GR	MONO
C_11	07	0.85	0.28	OVD	SMT	WHT	GR	MONO
C_12	00	-	-	-	-	-	-	-
C_13	04	0.94	0.26	SPD	WRK	CRM	WHT	MONO
C_14	05	0.87	0.34	OVD	SMT	GR	GR	MONO
C_15	28	1.56	0.94	OVD	SMT	WHT	WHT	MONO
C_16	04	0.85	0.36	OVD	SMT	GR	GR	MONO
C_17	30	0.86	0.43	OVD	SMT	BW	WHT	MONO
C_18	24	0.81	0.41	OVD	SMT	BW	WHT	MONO
C_19	09	1.26	0.32	CLV	SMT	CRM	CRM	POLY
C_20	04	1.24	0.64	OVD	SMT	CRM	GR	MONO
C_21	10	1.52	1.28	OVD	SMT	WHT	WHT	MONO
C_22	12	1.35	0.42	SPD	WRK	CRM	GR	POLY
C_23	07	1.29	0.48	CLV	SMT	CRM	GR	MONO
C_24	52	1.66	1.36	OVD	WRK	CRM	CRM	BOTH
C_25	08	0.86	0.65	OVD	SMT	CRM	WHT	MONO

Table 8: Seed morphological data of 25 samples of *Citrus* species.

A Dendrogram was constructed using ward's method for 25 samples of *Citrus* species combining morphological traits that were represented in Table 3, Table 4, Table 5, Table 6, Table 7, and Table 8 based on their similarity in morphological characteristics [49]. Illustrations of *Citrus* species' morphological features like plant shape, leaf shape, leaf margin shape, and fruit shape were obtained from the descriptor guidelines published in [46].

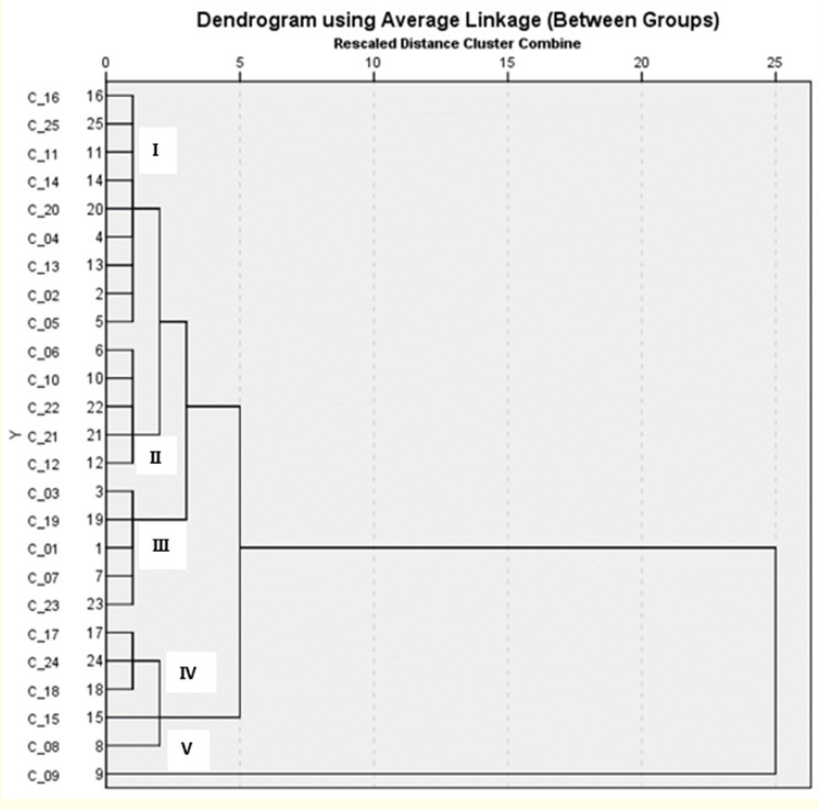

Figure 1: Dendrogram constructed using 74 morphological traits of 25 samples of *Citrus* species.

According to the above data there are five different clusters formed (Figure 1). Calamansi (C16), Yak dehi (ARS) (C 25), Yak dehi (Provincial name: Ambul naran) (C 11), Kumquat (C 14), Tahiti lime (C 20), Heen dehi (key lime) (C 04), Nas naran (ARS)(C 13), Nas naran (Home garden)(C 02), and Kaffir lime (C 05) belong to one cluster (Cluster I) which have common morphological features like tree height about 150 cm to 300 cm, presence of spines, having high branch density, simple leaf arrangement, presence of petiole, oval shape leaf with entire leaf margins, having small white flower with five petals, yellow stigma, yellow anther, having small size fruit with smooth yellow color epicarp, white color mesocarp, ten (10) to twelve (12) fruit segments with solid axis, having six (6) to twelve (12) smooth surfaced seeds. Lemon dehi (round Lemon) (C 06), Pochchi dehi (long lemon) (C 12), Peni kudalu orange (C 10), Rough lemon (C 21), and Trifoliate orange (C 22) belong to another separate cluster (Cluster II) which have common morphological features like tree height 100 cm to 400 cm, presence of spines, having low branch density, simple or trifoliate leaf arrangement, presence of petiole, oval or elliptical shape leaf with crenated leaf margins, having intermediate size white or purple color flower with five petals, yellow stigma, yellow anthers, having intermediate size fruit with green color rough epicarp, white color mesocarp, ten (10) to twelve (12) fruit segments with hollow axis, having zero (0) to twelve (12) rough seeds. Heen naran (C 03), Ambul dodam (sour orange) (C 01), Pani dodam (sweet orange) (C 07), Jama naran (C 19), and Philippine red lime (C 23) belong to a separate cluster (Cluster III) which have common morphological traits like tree height 300 cm to 500 cm, presence of spines, intermediate branch density, having simple elliptical leaves with entire margin, having small white flowers with yellow stigma and yellow anther, having

small or intermediate size fruits with dark green color rough epicarp, white color mesocarp, eight (8) to twelve (12) fruit segments with solid axis, having four (4) to twelve (12) smooth seeds. Citron (C 17), Buddha's hand (C 18), Citron hybrid (C 24) belong to smaller cluster which have common morphological features like tree height 100 cm to 200 cm, absence of spines, low branch density, having simple larger oval shape leaves with crenated leaf margins, having larger purple color flowers with green color stigma and yellow color anthers, having larger size fruits with smooth yellow or light green color epicarp, yellow mesocarp, ten (10) to fourteen (14) fruit segments with hollow axis, having rough seeds more than fifty (50). These three *Citrus* species join into cluster formed by joining Grapefruit (C 15) and Sidaran (C 08) (Cluster IV) which have slight difference from mentioned three *Citrus* species by having white color flowers, orange and yellow color epicarp respectively. Pomelo (C 09) is distinctively different from other clusters (Cluster V) which have tree height of 400 cm to 600 cm, absence of spines, high branch density, having larger leaves with entire margins, having large white color flowers with yellow stigma and yellow color anthers, having larger size fruits with smooth light green color epicarp, pink color mesocarp, having twelve (12) to sixteen (16) fruit segments with solid axis, having twenty (20) to fifty (50) rough seeds.

Conclusion

The morphological analyses of 25 samples of *Citrus* species revealed that Sri Lankan *Citrus* germplasm can be clustered into five main clusters based on 74 morphological traits.

Cluster I - Calamansi, Yak dehi (ARS), Yak dehi (Provincial name: Ambul naran), Kumquat, Tahiti lime, Heen dehi (key lime), Nas naran (ARS), Nas naran (Home gargen), and Kaffir lime. Cluster II – Lemon dehi (round lemon), Pochchi dehi (long dehi), Peni kudalu orange, Rough lemon, and Trifoliate orange. Cluster III - Heen naran, Ambul dodam (sour orange), Pani dodam (sweet orange), Jama naran and Philippine red lime. Cluster IV - Citron, Buddha's hand, Citron hybrid, Grapefruit and Sidaran, Cluster V - Pomelo.

Morphological traits provide valuable insights into the diversity and taxonomy of *Citrus* species. By systematically characterizing and documenting these traits, it is possible to establish a foundation for accurate species identification, classification, and germplasm conservation and supports breeding programs aimed at developing improved *Citrus* varieties.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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