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Taxonomic revision of the genus *Calendula* (Asteraceae) in the Canary Islands

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Abstract

A taxonomic revision of the genus *Calendula* (Asteraceae – Calenduleae) for the Canary Islands is presented. An extensive morphometric analysis was conducted on specimens held in several herbaria and collected by the team, as well as chromosome counts and genome size estimates. Four species of *Calendula* were recognized in the islands: *C. arvensis*, *C. officinalis*, *C. ricardoi*, and *C. sventenii*, the latter two being new taxa, described for the first time in this work. Descriptions, an identification key, distribution areas, ecology and conservation assessments for each taxon are presented, as well as discussions of the possible origins of the taxa in the territory.

Keywords: Calenduleae, taxonomy, DNA nuclear content, chromosome number

Introduction

Calendula L., a small genus belonging to the tribe Calenduleae (Asteraceae) and primarily distributed in the Mediterranean region, has been, for many years, considered taxonomically and evolutionary very complex. The 15 species recognized in the most recent partial revision works (Gonçalves 2018, Gonçalves *et al*. 2023) are distributed across Europe (especially in the south), Morocco, Algeria (including the southernmost populations of the genus in the Ahaggar Mountains in the Sahara), Tunisia, Libya, and Egypt. Morocco is considered as the center of diversification of the genus, where the taxa with the lower chromosome numbers occur (*Calendula stellata*, *2n*=14; *Calendula maroccana*, *2n*=18). The easternmost populations of the genus occur in the Arabian Peninsula and Iran (also found in the Yemen Mountains) and the westernmost ones are found throughout the Macaronesian region, in Madeira, the Azores and the Canary Islands (Norlindh 1946, 1977, Heyn *et al.* 1974, Nordenstam 2007, Nordenstam & Källersjö 2009, Gonçalves *et al.* 2018).

Most of the authors who studied *Calendula* (Lanza 1919, Heyn *et al*. 1974, Ohle 1974, 1975a, 1975b, Meikle, 1976), did not agree on the number of taxa (between 10 to 25) or on which characters are of taxonomic importance to distinguish among species, and provided different taxonomic treatments. These divergences are rooted in the great morphological and cytological variability of *Calendula*, which has hindered the establishment of strong taxonomic characters that undoubtedly separate taxa. The hybridization of species with different chromosome numbers and polyploidization, which have been considered the most probable hypothesis for speciation in the genus (Heyn & Joel 1983), also contributed to the complexity of these taxonomic studies.

Achene morphology has been one of the main morphological characters used to distinguish among species in *Calendula*, together with life form, habit, leaf shape, indumentum, and diameter of the capitula. However, more recently, information regarding chromosome numbers and nuclear DNA content has also been used to aid this process

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(Heyn *et al*. 1974, Gonçalves 2018). In complex genera such as *Calendula*, genetic information can be a powerful ally and often indicates taxonomic heterogeneity and influences taxa delimitation (Greilhuber & Speta 1985).

With seven islands and a group of small islets of volcanic origin that form a west-southwest to east-north-east archipelago, the Canaries possess over 680 endemic taxa, comprising at least 50% of the native described species (Santos-Guerra 2001). Despite the known high percentage of endemic flora of the Canary Islands, a taxonomic work concerning *Calendula* is still missing, apart from the occasional mention in regional flora publications (Buch 1819, 1825, 1833, Webb & Berthelot 1846, Pitard & Proust 1908, Lindinger 1926, Bramwell 1971, Hansen & Sunding 1985, Acebes-Genovés *et al*. 2001, 2004, 2010).

Considering this framework, the aim of this study is to improve the taxonomical knowledge about the genus *Calendula* in the Canary Islands, establishing a formal and up-to-date taxonomical treatment that includes morphological and ecological data, but also karyological information such as chromosome numbers and genome size for all the taxa.

Material and Methods

Plant Material

The morphological study was based on plant material from the Canary Islands, gathered during field collections by the team (c. 130, held at AVE and ORT) and herbarium specimens (c. 300) deposited in BCN, BM, BR, C, COI, K, LPA, MA, MACB, MAF, O, ORT, RAB, RNG, S, SALA, SEV and TFC. Additionally, fruits collected during the field collections were germinated and chromosome counts were obtained from the cultivated plants in the Instituto Superior de Agronomia (ISA) of the University of Lisbon, while genome size estimations was carried out on samples gathered during the field collections.

Morphometric analysis

Morphological measurements were performed on dry material for all taxa described in this paper using a ruler and a digital caliper. For the morphometric analysis of each taxon, 27 quantitative characters (Table 1) were measured in at least 10 individuals, when possible (Appendix I), and boxplots were elaborated with percentile and median values, representing species variability.

Statistical analysis

Analysis of variance was performed on morphological data for 21 quantitative characters from Table 1, and on genome size data to determine significant differences between the taxa. When the data presented a normal distribution, a t-test was used (between two groups) or One-way ANOVA (between three or more groups). When normality was not met, Mann-Whitney test (between two groups), and Kruskal-Wallis test were used (between three or more groups). The Dunn and Tukey's post hoc tests were then used to identify which groups were significantly different from the others for each of the characters considered. For each quantitative character, statistical parameters such as mean, percentile 25%, percentile 75%, minimum value, maximum value, standard deviation (SD) and standard error (SE) were calculated and represented in boxplots. For genome size data the mean, standard deviation (SD), coefficient of variation (CV), minimum values and maximum values of the holoploid (2C, pg) were also calculated.

Distribution maps and conservation assessments

Distribution maps for each taxon were elaborated based on latitude and longitude coordinates from specimens gathered during field collections as well as the herbaria specimens that could be georeferenced in DIVA-GIS, v. 7.5 (http://www.diva-gis.org).

Regional or global (when applicable) conservation status of three native species was assessed following the IUCN Red List Categories and Criteria (http://www.iucnredlist.org/). Area of occupancy (AOO) and extent of occurrence (EOO) were assessed using the Geospatial Conservation Assessment Tool (GeoCAT) (http://geocat.kew.org/).

TABLE 1. List of quantitative characters used in this study.

Characters	Abb.	Unit/Scale
Length of the longest branch	LB	cm
Basal leaf length	LL	mm
Basal leaf width	LW	mm
Basal leaf distance from base to point of maximum width	LD	mm
Ratio basal leaf length/point of maximum width	R1	ratio
Ratio basal leaf length/width	R2	ratio
Basal leaf thickness	LT	mm
Head diameter	HD	mm
Involucre length	IL	mm
Ligule length	LG	mm
Ratio ligule/involucre	R3	ratio
Rostrate achene length	RL	mm
Rostrate achene width	RW	mm
Rostrate achene dorsal spine	RS	mm
Sub-rostrate achene length	SRL	mm
Sub-rostrate achene width	SRW	mm
Sub-rostrate achene dorsal spine	SRS	mm
Bialate achene length	BL	mm
Bialate achene width	BW	mm
Trialate achene length	TL	mm
Trialate achene width	TW	mm
Cymbiform achene length	CL	mm
Cymbiform achene width	CW	mm
Vermiform-alate achene length	VAL	mm
Vermiform-alate achene width	VAW	mm
Vermiform-exalate achene length	VEL	mm
Vermiform-exalate achene width	VEW	mm

Chromosome number

Chromosomes were observed and counted in mitotic cells isolated from apical root meristems of cultivated adult plants or seedlings of *Calendula* (see Table 5). Achenes were germinated in Petri dishes with filter paper after the removal of the tegument. When seedlings were developed, they were transplanted into Jiffy-7 pots (www.jiffy.com) and kept in a growth chamber (max. temperature 20ºC, min. temperature 10 ºC, 12 hours of light) until the plants developed and produced roots. Roots were pre-treated in ice-cold water for 15 h, and then fixed with Carnoy fixative solution (absolute ethanol: glacial acetic acid in proportion 3:1) for 7 h at room temperature. The fixed material was stored in Carnoy fixative solution until used at -20ºC. For the analysis, the fixed material was digested in an enzyme mixture containing cellulase Sigma-Aldrich (from *Trichoderma* sp.), pectolyase Sigma-Aldrich (from *Aspergillus japonicus*), and cytohelicase Sigma-Aldrich (from *Helix pomatia*) (Esmaeili *et al*. 2020) at 37ºC for 3 h. Slides were prepared according to the squash technique and screened with a phase contrast microscope to find metaphase plates. VECTASHIELD Mounting Medium with DAPI (Vector Laboratories, Burlingame, California, USA) was added to the permanent chromosome slides. The Axio Imager.Z1 epifluorescence microscope (Carl Zeiss) using the Zeiss filter sets 49 for DAPI (excitation 365 nm, beam splitter 395 nm, and emission 445/50 nm) was used to acquire chromosome images.

Genome size assessments

For the genome size estimations, fresh leaves from 100 specimens (see Table 3) were collected and maintained in refrigerated conditions until being analyzed by flow cytometry following the methodology described in Guilengue *et*

al. (2020). For that, 50 mg of leaf tissue and 50 mg of standard leaves *Pisum sativum* (2C= 9.09 pg; Doležel *et al*. 1992) were chopped in 1 ml of woody plant (WPB) buffer (200 mM Tris.HCl, 4mM $MgCl_2.6H_2O$, 2 mM EDTA $Na_2.2H_2O$, 86 mM NaCl, 10 mM sodium metabisulfite, 1% PVP-10, 1% (v/v) Triton X-100, pH 7.5; Loureiro *et al*. 2006) in a Petri dish using a razor blade. After chopping, the solution was filtered through a nylon mesh with 30 μm, and then added 100 $\mu g.mL^{-1}$ of propidium iodide.

Samples were then analyzed in a CyFlow Space flow cytometer (Sysmex, Norderstedt, Germany) equipped with a 30 mW green solid-state laser emitting at 532 nm for optimal PI excitation. For each sample, five replicas were made. Nuclear FlowMax software v2.4d (Sysmex) was used to measure nuclear DNA content and fluorescence pulse integral in linear scale (FL) graphs were generated from data measurements. The nuclear DNA content of the samples was calculated based on the mean values of G1 peaks, following Doležel & Bartoš (2005).

DNA content estimates were only considered when the coefficient of variation of G0/G1 peaks (CV peak) was below 5%. Statistical analysis was performed with Mann-Whitney U test (between two groups) for non-normal data.

Results

Variability of *Calendula* species in the Canary Islands

Twenty-one quantitative characters from Table 1 were selected considering their importance in the distinction between taxa and submitted to analysis of variance to test for significant differences. The results of these tests are summarized in Table 4 and showed significant differences between groups ($P<0.0001$) in the 21 characters analyzed, except for the length of spines in rostrate and sub-rostrate achenes and the length and width of sub-rostrate achenes. Boxplots representing the variability of 27 quantitative characters for the *Calendula* species are presented in Figures 1, 2, and 3. Appendix I contains a table with the results of the morphometric analysis, used for the elaboration of the boxplots. Some of the characters are only present in one or two groups, such as the trialate achenes length and width (Figure 3).

The morphometric analysis allowed to distinguish four groups of specimens in the Canary Islands. Two of these groups corresponded to already established species, *C. arvensis* and *C. officinalis.* However, the other two corresponded to new taxa, since their morphology was not consistent with any other species known in the genus, for the Canary Islands or any other territory of the distribution of *Calendula,* especially concerning the presence or absence of certain morphological characters of the achenes and floral morphology. The specimens from these groups were named *C. sventenii* (Figures 9, 10c, and 11b) and *C. ricardoi* (Figures 8, 10d, and 11c). These differences were further confirmed with the multiple comparison tests done on the morphometric analysis which showed significant differences between the two taxa in several characters, such as HD, LL, IL, R1, RL, RW, TL, and TW. The main morphological differences between these two taxa and the most similar species, *C. tripterocarpa* and *C. arvensis*, are presented in Table 2.

Chromosome number and genome size

This work includes, for the first time, genome size estimates for the *Calendula* taxa of the Canary Islands, which varied from 3.1 pg (3014.2 Mbp) in *C. officinalis* to 5.9 pg (5925 Mbp) in *C. arvensis.* Significant differences were found between species ($H=285.163$; $p < 0.0001$). The mean CV for most of the analyzed samples was lower than 5% (global mean of 4.2%). Chromosome numbers varied from $2n= 32$ in *C. officinalis* (Figure 4B), to $2n=44$ for *C. arvensis* (Figure 4A), and $2n=54$ for *C. sventenii* and *C. ricardoi* (Figure 4C and D, respectively). Different taxonomic treatments, chromosome number counts, and genome size estimates made in this work are summarized in Table 5.

Conservation assessments

An evaluation of the conservation status of *C. arvensis, C. sventenii*, and *C. ricardoi* for the Canary Islands was made following the IUCN Red List Categories and Criteria (http://www.iucnredlist.org/). *Calendula officinalis* was excluded from this assessment, being cultivated and sometimes escaped from cultivation in the territory. *Calendula arvensis* was assessed as Least Concern (LC), being a widely distributed species across the seven islands, without major threats to its populations. Although not as cosmopolitan as the previous, *C. sventenii* was also assessed as LC, considering its stable population trends and distribution range. *Calendula ricardoi* was assessed as vulnerable (VU D1+2), since only

one population of less than 1000 mature individuals is known, with a small distribution range (EOO = 0.5 km^2, AOO = 12 km^2). Even though it is a taxon restricted to only one of the islands (Fuerteventura), its main distribution area is included in a natural protected area where collection is prohibited without permission, and we have no knowledge of an observed or estimated continuing decline of this population.

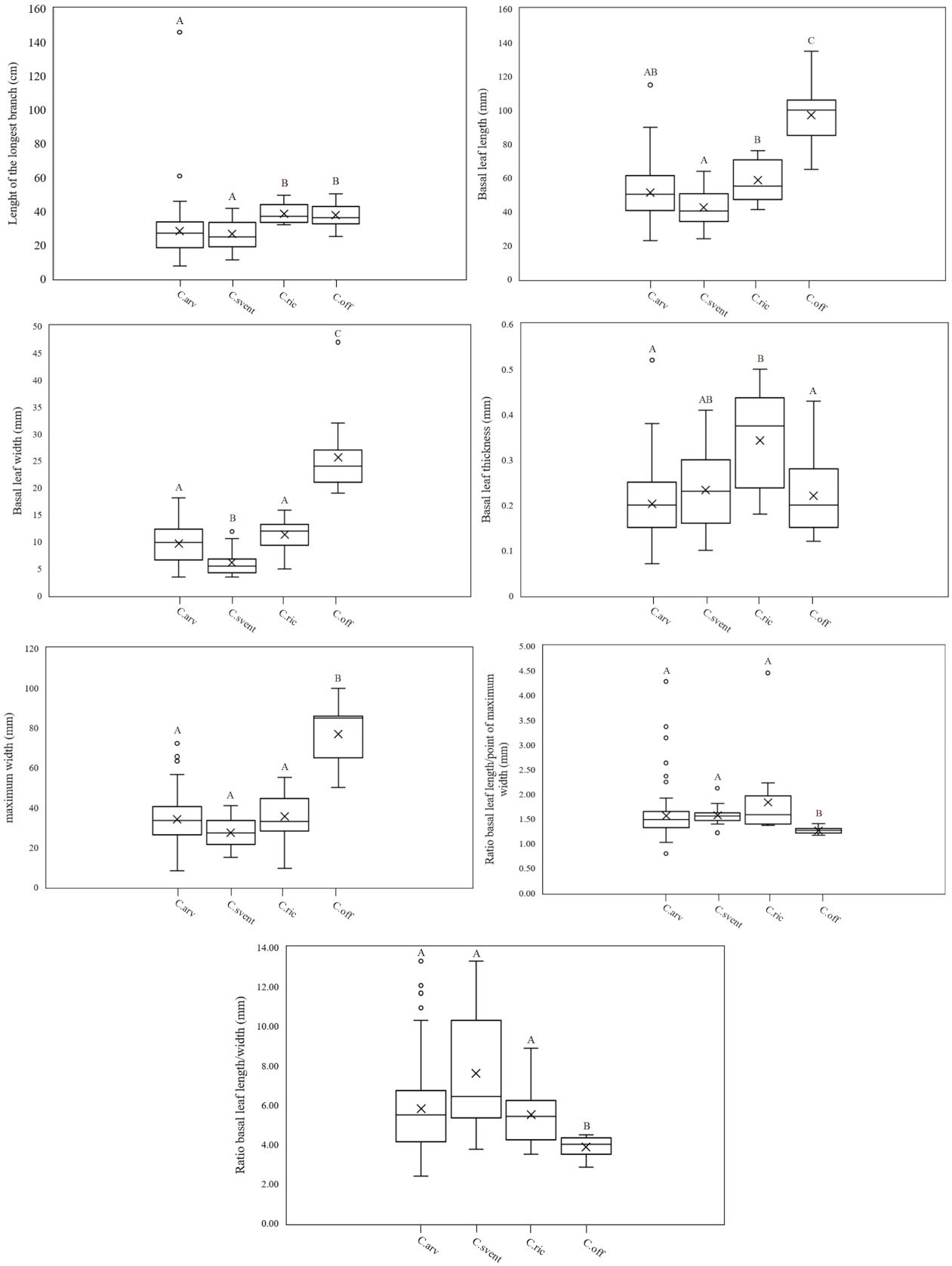

FIGURE 1. Boxplots representing the variability of foliar quantitative characters in *Calendula* species. Different letters reveal statistical differences at $P < 0.05$.

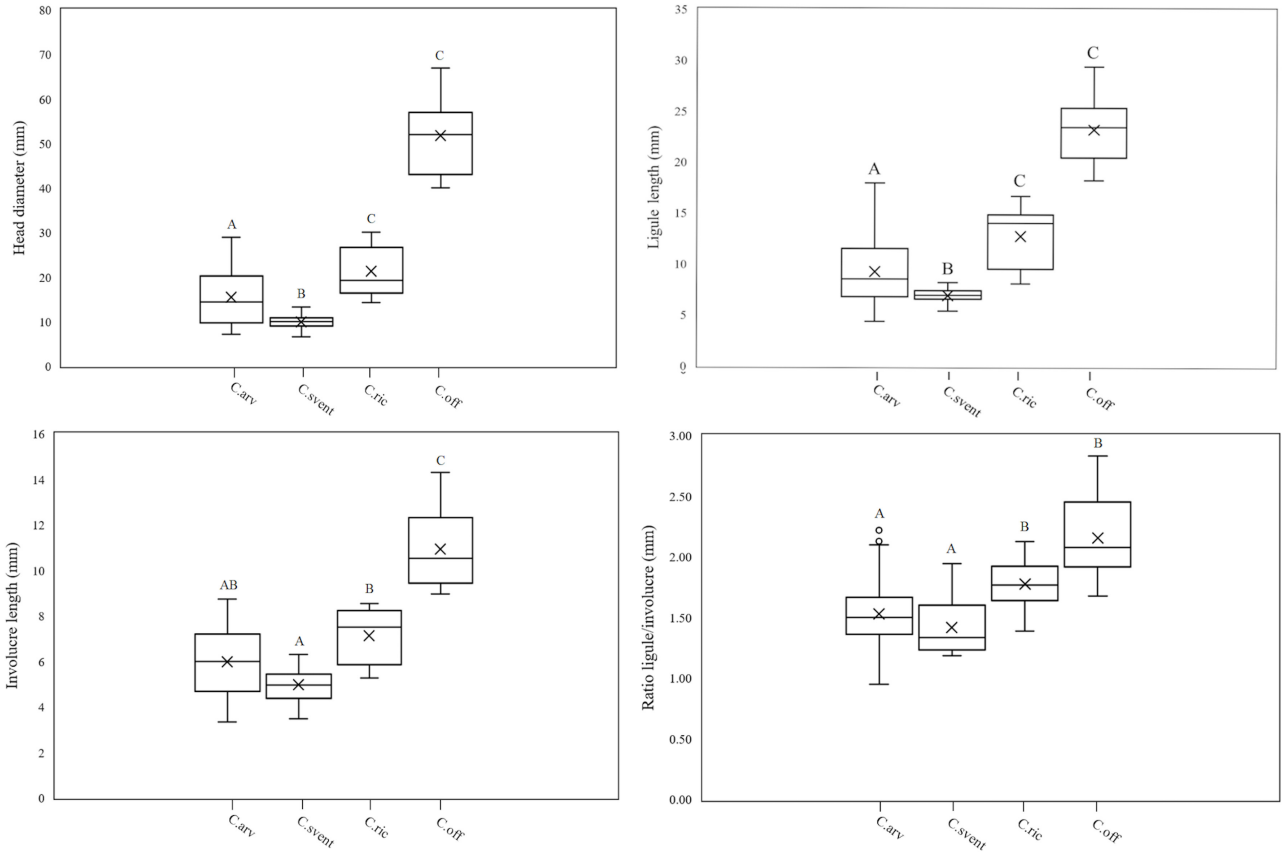

FIGURE 2. Boxplots representing the variability of floral quantitative characters in *Calendula* species. Different letters reveal statistical differences at $P < 0.05$.

TABLE 2. Main diagnostic characters between the new taxa reported for the Canary Islands (*C. sventenii* and *C. ricardoi*) and the most similar taxa *C. arvensis*, also from the Canary islands, and *C. tripterocarpa*, from Morocco (Gonçalves *et al.* 2023).

Characters	***C. arvensis***	***C. tripterocarpa***	***C. sventenii***	***C. ricardoi***
Height	(7.5) 18.5–33.4 (146) cm long, erect	(2) 5.6–15.8 (30) cm long, decumbent to diffuse	(11) 20–32.4 (41.5) cm long, ascending to erect	(32) 33–42 (50) cm long, occasionally longer, erect
Leaf shape	Oblanceolate, higher leaves usually oblanceolate to lanceolate	Basal leaves linear-oblong, upper leaves oblanceolate to lanceolate	Strictly linear-oblong	Ovate to oblong, less frequently oblanceolate
Capitula diameter (cm)	(0.7) 0.9–2 (2.9)	(1.1) 1.2–1.4 (1.6)	(0.7) 0.9–1.1 (1.3)	(1.4) 1.7–2.7 (3)
Ligules (mm)	(4.4) 6.8–11.5 (18) × (0.64) 1.23– 1.98 (2.58) Without purple/reddish spots under the ligules	(5.8) 6–7.7 (8.7) × 0.6–1.2 Without purple/reddish spots under the ligules	(5.4) 6.6–7.4 (8.2) × (0.7) 0.9–1.1 (1.6) Without purple/reddish spots under the ligules	(8.1) 9.6–14.9 (16.7) × (1) 1.9–2.3 (2.5) Often with purple/ reddish spots under the ligules
Disc florets	18–20 Yellow-orange or brown-purplish	(4) 6–20 Yellow-orange	13–23 Yellow-orange	39–50 Yellow-orange
Involucre length	(3.3) 4.7–7.1 (8.7) mm	No data	(3.5) 4.5–5.4 (6.3) mm	(5.3) 6.1–8.1 (8.5) mm

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TABLE 2. (Continued)

Characters	*C. arvensis*	*C. tripterocarpa*	*C. sventenii*	*C. ricardoi*
Rostratre achenes (mm)	(4.7) 7.4–11 (20.3) × (0.9) 1.7–2.4 (3.7) Without an enlarged embryo, and with a relatively short, bent, rostrum, which do not end in a hook-like tooth	Absent	(4.9) 6.2–10.6 (13.5) × (1.3) 1.8–3.4 (3.9) With an enlarged embryo and, frequently, a long strait rostrum, ending in a hook-like tooth	(10.4) 13.7–22 (25) × (1.8) 2.6–3.6 (5.1) With an enlarged embryo and, frequently, a long strait rostrum, ending in a hook-like tooth
Trialate achenes, length (mm)	Absent*	(5.7) 6.3–7.9 (9.5)	(5.7) 7–8.6 (9.1)	(8) 8.6–10.5 (11.5)
Bialate achenes (mm)	(4.1) 6.3-7.7 (10.4) × (1.5) 6.1-7.9 (9.3) Absent, or present (in the macroptera form)	Absent	Absent	Absent
Genome size	5.4 pg ± 0.2	3.44 ± 0.06 pg	3.7 pg ± 0.16	5.1 pg ± 0.7
Chromosome number	*2n* = 44	*2n* = 30	*2n* = 54	*2n* = 54

*Some bialate achenes could be confused with trialate achenes, but they have an incomplete ventral wing, deep incised lateral wings and dorsal spines. The trialate achenes display complete ventral wings, lateral wings entire or almost entire, and no dorsal spines or small spines.

TABLE 3. *Calendula* specimens used in the flow cytometry analysis.

Taxa	Locality	Coordinates	Voucher No.
C. arvensis	LANZAROTE: surroundings of Playa Blanca	28°52’24” N, 13°49’54” W	3384.1
	LANZAROTE: surroundings of Playa Blanca	28°52’42” N, 13°49’57” W	3385.1
	LANZAROTE: surroundings of Playa Blanca	28°52’42” N, 13°49’57” W	3385.3
	LANZAROTE: surroundings of Playa Blanca	28°52’42” N, 13°49’57” W	3385.4
	LANZAROTE: surroundings of Playa Blanca	28°52’42” N, 13°49’57” W	3385.5
	LANZAROTE: surroundings of Playa Blanca	28°52’41” N, 13°49’58” W	3386
	LANZAROTE: Teguise	29°03’50” N, 13°32’59” W	3389.1
	LANZAROTE: Teguise	29°03’50” N, 13°32’59” W	3389.2
	LANZAROTE: Manguía	29°04’14” N, 13°31’31” W	3390.1
	LANZAROTE: Manguía	29°04’14” N, 13°31’31” W	3390.2
	LANZAROTE: Mirador de Guinate	29°11’06” N, 13°30’04” W	3392.1
	LANZAROTE: Mirador de Guinate	29°11’06” N, 13°30’04” W	3392.2
	LANZAROTE: El Jable	29°03’55” N, 13°35’05” W	3393.1
	LANZAROTE: El Jable	29°03’55” N, 13°35’05” W	3393.2
	LANZAROTE: El Jable	29°03’55” N, 13°35’05” W	3394
	LANZAROTE: El Jable	29°03’55” N, 13°35’05” W	3395.1
	LANZAROTE: El Jable	29°03’55” N, 13°35’05” W	3395.2
	LANZAROTE: El Jable	29°03’55” N, 13°35’05” W	3395.3
	LANZAROTE: Punta Mujeres	29°08’19” N, 13°27’18” W	3124a
	LANZAROTE: Punta Mujeres	29°08’19” N, 13°27’18” W	3124b
	LANZAROTE: Punta Mujeres	29°08’19” N, 13°27’18” W	3125
	LANZAROTE: Orzola	29°08’19” N, 13°27’18” W	3126
	FUERTEVENTURA: along FV-102 towards La Caldereta	28°35’05” N, 13°51’49” W	3370
	FUERTEVENTURA: along FV-102 towards La Caldereta	28°35’05” N, 13°51’49” W	3371
	FUERTEVENTURA: agricultural fields near Montaña Del Frontón	28°35’59” N, 13°55’06” W	3373
	FUERTEVENTURA: agricultural fields near Montaña Del Frontón	28°35’59” N, 13°55’06” W	3374

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TABLE 3. (Continued)

Taxa	Locality	Coordinates	Voucher No.
	FUERTEVENTURA: agricultural fields near Montaña Del Frontón	28°35’59” N, 13°55’06” W	3375
	FUERTEVENTURA: Betancuria	28°25’42” N, 14°03’29” W	3327.i2
	FUERTEVENTURA: Betancuria	28°25’42” N, 14°03’29” W	3327.i3
	GRAN CANARIA: Ingenio. El Roque	27°56’52” N, 15°28’49” W	3420.1
	GRAN CANARIA: Ingenio. El Roque	27°56’52” N, 15°28’49” W	3420.2
	GRAN CANARIA: Ingenio. El Roque	27°56’52” N, 15°28’49” W	3420.3
	GRAN CANARIA: Ingenio. El Roque	27°56’52” N, 15°28’49” W	3420.4
	GRAN CANARIA: Ingenio. El Roque	27°56’52” N, 15°28’49” W	3420.5
	GRAN CANARIA: Valsequillo. Borde de la Caldera de los Marteles	27°57’31” N, 15°31’58” W	3421
	GRAN CANARIA: Cruz de Tejeda	28°00’20” N, 15°36’00” W	3422.1
	GRAN CANARIA: Cruz de Tejeda	28°00’20” N, 15°36’00” W	3422.2
	GRAN CANARIA: Barranco de Agaete	28°06’06” N, 15°42’17” W	3425
	GRAN CANARIA: Mogán. Cortadores	27°51’41” N, 15°40’44” W	3428.1
	GRAN CANARIA: Mogán. Cortadores	27°51’41” N, 15°40’44” W	3428.2
	GRAN CANARIA: Mogán. Cortadores	27°51’41” N, 15°40’44” W	3428.3
	GRAN CANARIA: Mogán. Cortadores	27°51’41” N, 15°40’44” W	3428.4
	GRAN CANARIA: Mogán. Cortadores	27°51’41” N, 15°40’44” W	3428.5
	TENERIFE: C. del Calvario between Tacoronte and Lomo Colorado	28°29’00” N, 16°24’22” W	3397.1
	TENERIFE: C. del Calvario between Tacoronte and Lomo Colorado	28°29’00” N, 16°24’22” W	3397.2
	TENERIFE: C. del Calvario between Tacoronte and Lomo Colorado	28°29’00” N, 16°24’22” W	3397.3
	TENERIFE: C. del Calvario between Tacoronte and Lomo Colorado	28°29’00” N, 16°24’22” W	3397.4
	TENERIFE: La Laguna	28°30’04” N, 16°18’46” W	3398.1
	TENERIFE: La Laguna	28°30’04” N, 16°18’46” W	3398.2
	TENERIFE: Las Canteras	28°30’24” N, 16°10’41” W	3399
	TENERIFE: Bejía	28°32’53” N, 16°18’04” W	3400
	TENERIFE: Bejía	28°32’53” N, 16°18’04” W	3401
	TENERIFE: Bejía	28°32’53” N, 16°18’04” W	3402
	TENERIFE: Carr. Tahodio	28°29’20” N, 16°14’56” W	3403
	TENERIFE: street near Buenavista	28°23’00” N, 16°50’18” W	3405
	TENERIFE: Buenavista del Nte.	28°22’21” N, 16°50’58” W	3406
	TENERIFE: Buenavista del Nte.	28°22’21” N, 16°50’58” W	3407
	TENERIFE: Buenavista del Nte.	28°22’21” N, 16°50’58” W	3408
	TENERIFE: TF-375 near Las Manchas	28°16’55” N, 16°47’51” W	3410
	TENERIFE: TF-375 near Las Manchas	28°16’55” N, 16°47’51” W	3411.1
	TENERIFE: TF-375 near Las Manchas	28°16’55” N, 16°47’51” W	3411.2
	TENERIFE: TF-82 near Barranco de Ramallo	28°12’14” N, 16°46’10” W	3412.1
	TENERIFE: TF-82 near Barranco de Ramallo	28°12’14” N, 16°46’10” W	3412.2
	TENERIFE: TF-82 near Barranco de Ramallo	28°12’14” N, 16°46’10” W	3413.1
	TENERIFE: TF-82 near Barranco de Ramallo	28°12’14” N, 16°46’10” W	3413.2
	TENERIFE: Tejina de Isora	28°11’01” N, 16°45’55” W	3414
	TENERIFE: fields along Av. el Palm-Mar	28°01’35” N, 16°41’18” W	3415.1
	TENERIFE: fields along Av. el Palm-Mar	28°01’35” N, 16°41’18” W	3416.1
	TENERIFE: fields along Av. el Palm-Mar	28°01’35” N, 16°41’18” W	3416.2
	TENERIFE: fields along Av. el Palm-Mar	28°01’35” N, 16°41’18” W	3417
	TENERIFE: Teno Alto. Bajada a las Cuevas	28°20’58” N, 16°53’39” W	3427

.....continued on the next page

TABLE 3. (Continued)

Taxa	Locality	Coordinates	Voucher No.
C. officinalis	LANZAROTE: Femés	28°54'45" N, 13°46'47" W	3388
	TENERIFE: Near Puerto de Erjos	28°19'41" N, 16°48'12" W	3409.1
	TENERIFE: Near Puerto de Erjos	28°19'41" N, 16°48'12" W	3409.2
	GRAN CANARIA: Valleseco. Sobre Mesas de Galaz	28°00'54" N, 15°35'25" W	3423
C. sventenii	LANZAROTE: Punta Pechiguera	28°51'22" N, 13°52'18" W	3383.1
	LANZAROTE: Punta Pechiguera	28°51'22" N, 13°52'18" W	3383.2
	LANZAROTE: Punta Pechiguera	28°51'22" N, 13°52'18" W	3383.3
	LANZAROTE: Punta Pechiguera	28°51'22" N, 13°52'18" W	3383.4
	LANZAROTE: Punta Pechiguera	28°51'22" N, 13°52'18" W	3383.5
	LANZAROTE: Punta Pechiguera	28°51'22" N, 13°52'18" W	3383.6
	LANZAROTE: surroundings of Playa Blanca	28°52'24" N, 13°49'54" W	3384.2
	LANZAROTE: surroundings of Playa Blanca	28°52'42" N, 13°49'57" W	3385.2
	LANZAROTE: surroundings of Playa Blanca	28°52'41" N, 13°49'58" W	3387.1
	LANZAROTE: surroundings of Playa Blanca	28°52'41" N, 13°49'58" W	3387.2
	LANZAROTE: surroundings of Playa Blanca	28°52'41" N, 13°49'58" W	3387.3
	LANZAROTE: mirador de Guinate	29°11'06" N, 13°30'04" W	3391.1
	LANZAROTE: mirador de Guinate	29°11'06" N, 13°30'04" W	3391.2
	FUERTEVENTURA: along FV-102 towards La Caldereta	28°35'05" N, 13°51'49" W	3372
	FUERTEVENTURA: Gran Tarajal	28°15'23" N, 14°01'04" W	3381
	FUERTEVENTURA: Corralejo	28°44' N, 13°52' W	3123a
	FUERTEVENTURA: Corralejo	28°44' N, 13°52' W	3123b
	TENERIFE: fields along Av. el Palm-Mar	28°01'35" N, 16°41'18" W	3415.2
	TENERIFE: fields along Av. el Palm-Mar	28°01'35" N, 16°41'18" W	3515.3
	TENERIFE: fields along Av. el Palm-Mar	28°01'35" N, 16°41'18" W	3415.4
	TENERIFE: fields along Av. el Palm-Mar	28°01'35" N, 16°41'18" W	3415.5
C. ricardoi	FUERTEVENTURA: Betancuria	28°25'42" N, 14°03'29" W	3327.i1
	FUERTEVENTURA: Betancuria	28°25'42" N, 14°03'29" W	3327.i4
	FUERTEVENTURA: Betancuria	28°21'25" N, 14°04'51" W	3327.l.1/2

Taxonomic implications for the species of the Canary Islands

Based on our study of the herbarium specimens and living plants in the field and under cultivation, and on the morphometric analysis and statistical tests, we were able to establish four groups of specimens for the Canary Islands. Since the statistical tests performed on these characters showed significant differences between groups, a selection of these characters was considered of taxonomic importance and used in the elaboration of the identification key for the species of *Calendula* in the Canary Islands. The presence or absence of certain morphological characters of the achenes was also a criterion considered in the elaboration of the identification key (see Table 2). Furthermore, the karyological tools applied in the study, such as genome size assessments and, in particular, chromosome counts, help corroborate the distinction of these groups as separate species.

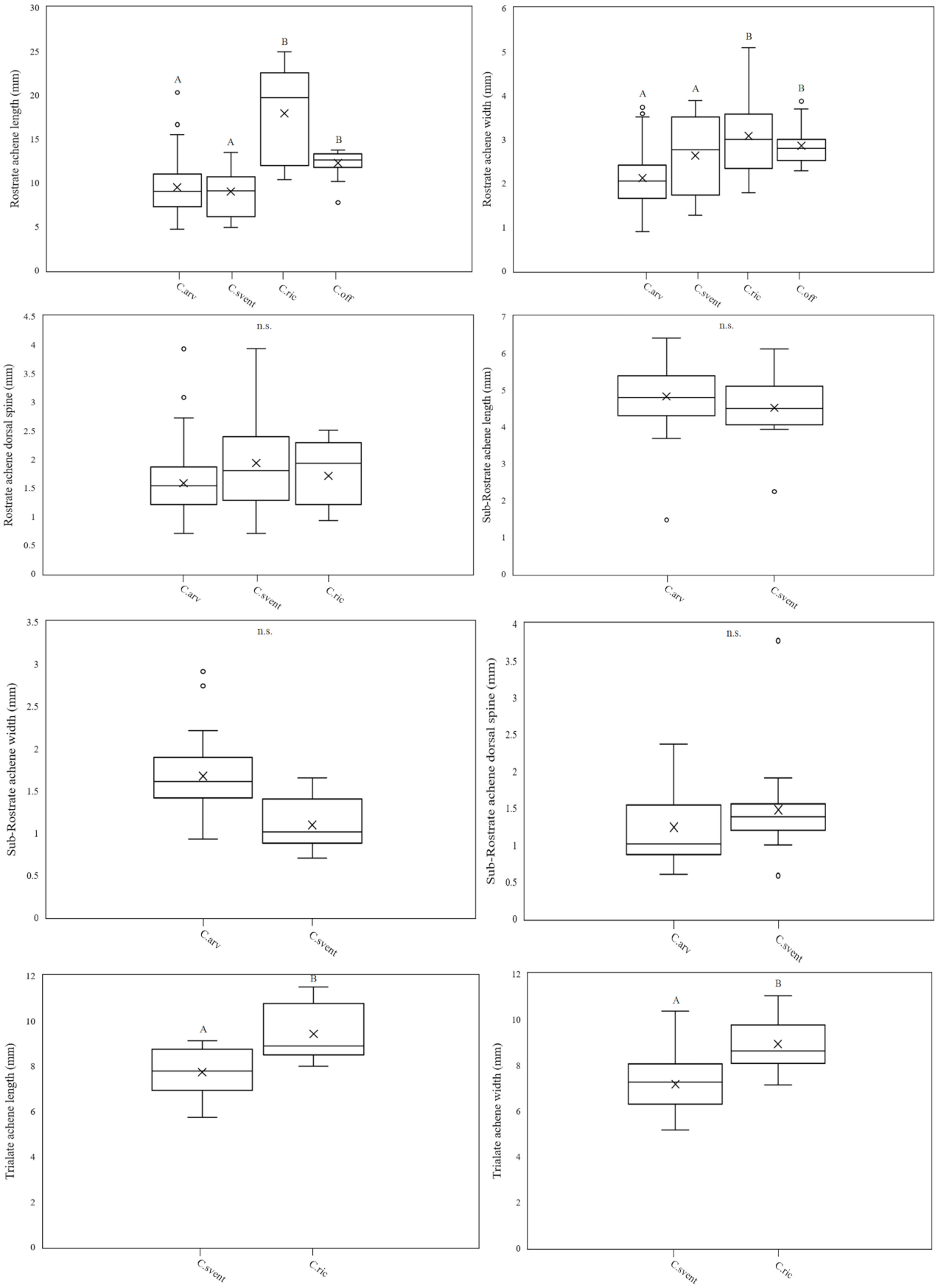

FIGURE 3. Boxplots representing the variability of fruit quantitative characters in *Calendula* species. Different letters reveal statistical differences at $P < 0.05$; n.s. means that no statistical differences were found.

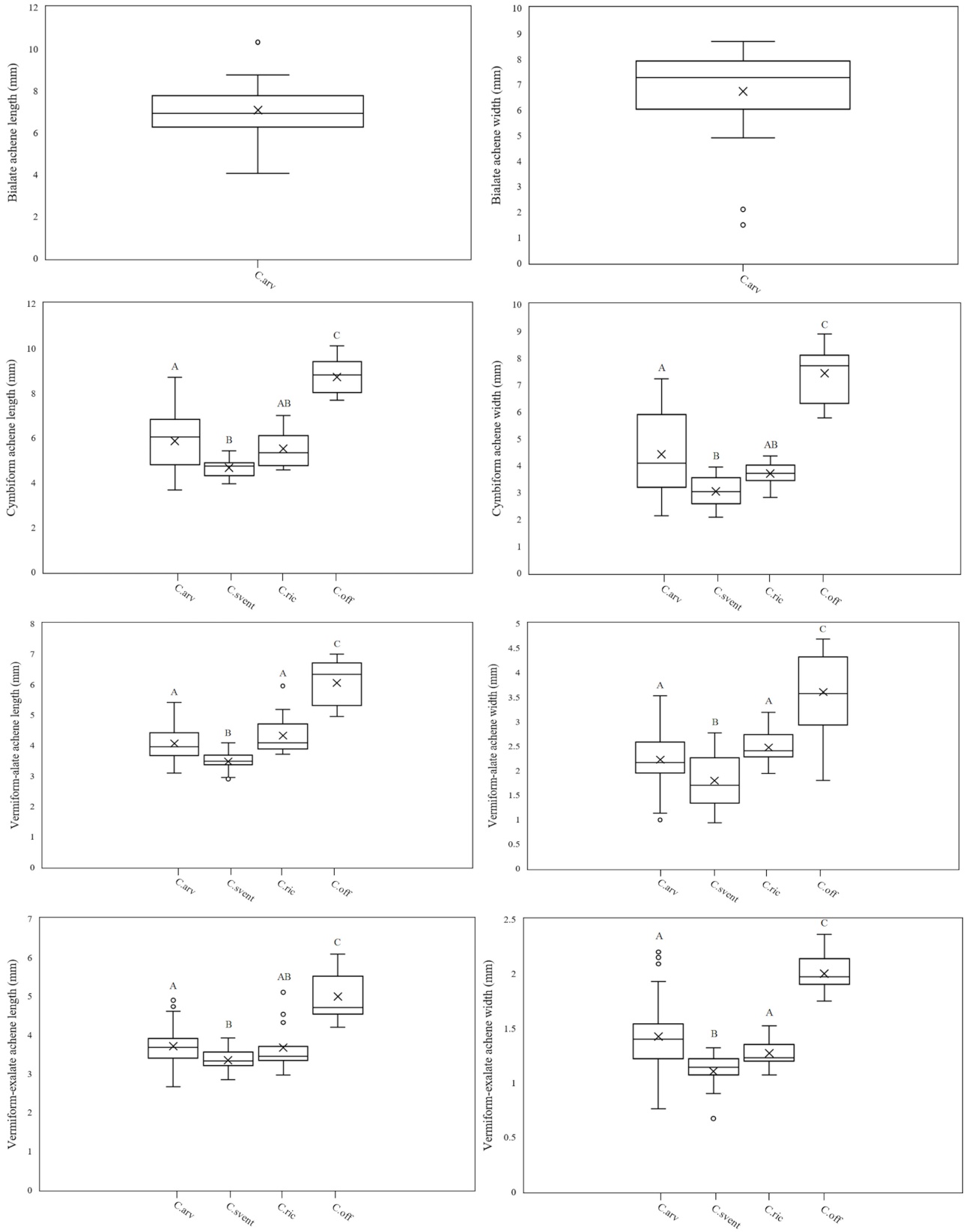

FIGURE 3. (Continued)

TABLE 4. Univariate statistical analysis of *Calendula* species from the Canary Islands.

Morphological trait	d.f.	Statistical test	P
Length of the longest branch	3	H=24.455	<0.0001
Basal leaf length	3	H=44.884	<0.0001
Basal leaf width	3	H=55.325	<0.0001
Basal leaf distance from base to point of maximum width	2	F=78.938	<0.0001
Basal leaf thickness	3	H=21.123	<0.0001
Head diameter	3	H=60.162	<0.0001
Involucre length	3	H=54.992	<0.0001
Ligule length	3	H=60.643	<0.0001
Ratio ligule/involucre	3	H=45.053	<0.0001
Rostrate achene length	3	H=38.113	<0.0001
Rostrate achene width	3	H=24.484	<0.0001
Sub-rostrate achene length		U=357	0.286
Sub-rostrate achene width	53	t=4.639	<0.0001
Trialate achene length		U=42	<0.0001
Trialate achene width	32	H=47.789	<0.0001
Cymbiform achene length	3	H=50.250	<0.0001
Cymbiform achene width	3	H=47.789	<0.0001
Vermiform-alate achene length	3	H=56.749	<0.0001
Vermiform-alate achene width	2	F=38.057	<0.0001
Vermiform-exalate achene length	3	H=42.801	<0.0001
Vermiform-exalate achene width	2	F=133.869	<0.0001

One-way ANOVA and t-test: F and t, respectively, for characters with normal distributions. Kruskal-Wallis and Mann-Whitney test: H and U, respectively, for characters with non-normal distributions. d.f. – degrees of freedom.

TABLE 5. Chromosome numbers and genome size in *Calendula* taxa from the Canary Islands.

Dalgaard (1986)	Baltisberger & Widmer (2006)	This study	*ncells*	*2n*	Genome size (2C/pg)				
					Mean	Min.	Max.	CV	n
	C. arvensis	*C. arvensis*	15	44	5.4±0.2[a]	4.9	5.9	3.9	71
		*C. ricardoi***	27	54	5.1±0.7[b]	3.7	5.3	3.6	4
*C. tripterocarpa**		*C. sventenii***	23	54	3.7± 0.16[ab]	3.5	3.9	4.4	21
		C. officinalis	7	32	3.2±0.06[c]	3.1	3.2	4.7	4

The table represents the medium 2C-value in pg for each of the taxon analyzed in this study, with respective standard deviation, minimal and maximum value, number of populations studied and individuals (n), analyzed chromosome number in the present study (2n), number of cells counted for each taxon (ncells) and coefficient of variation of the G0/G1 peak in % (CV); * nomenclatural change; **newly described taxa

Taxonomic treatment

Calendula Linnaeus (1753: 921). Lectotype (designated by Green in Hitchcock & Green 1929: 183): *Calendula officinalis* L.

Annual or perennial herbs covered with glandular and non-glandular hairs in different proportions. Stems (7.5) 20.7–36.15 (146) cm prostrate, ascending, diffuse or erect, branched, frequently glandular and aromatic. Leaves (22.9) 41–65.4 (135) × (3.4) 6.5–12 (47) mm, (0.1) 0.16–0.26 (0.5) mm thick, alternate, simple, uninervate, varying from spatulate to sub-spatulate, obovate, oblanceolate, oblong or linear, with margins entire, repand, undulate to dentate; base attenuate, truncate or auriculate; apex acute to obtuse; upper leaves usually similar in shape to the basal ones but

progressively smaller towards the apex, many times sessile and auriculate. Capitula (0.65) 1–2.3 (6.7) cm, solitary, heterogamous and radiate. Involucre (3.3) 4.9–7.7 (14.3) mm, with 1–2 rows of linear or acuminate bracts, apex acute, and a narrow scarious margin. Ray florets (8) 13–19 (45), (4.4) 7–13.2 (29.4) mm, ligulate, female and fertile, with a yellow or orange corolla, hairy at the base and 3-lobed at the apex. Disc florets 13–50, over 50 in *C. officinalis*, (2.4) 2.75–3.7 (4.7) mm, functionally male, with a yellow to orange, sometimes brown or violet-purple tubular corolla, 5-lobed. Anthers (1) 1.15–1.97 (2.37) mm, sagittate-caudate, with free filaments. Styles (1.5) 1.9–3.2 (4.3) mm. Receptacle flat, without palea. Outer achenes rostrate (4.7) 7.7–12.8 (24.9) × (0.9) 1.9–2.98 (5.1) mm, straight to deeply curved, often with developed dorsal spines, sometimes bialate (4.1) 6.3–7.7 (10.4) × (1.5) 6.1–7.9 (9.3) mm, trialate (5.7) 7.7–9 (11.5) × (5.2) 7.1–8.6 (11.1) mm or sub-rostrate (1.48) 4.2–5.3 (6.4) × (0.7) 1.2–1.7 (2.9) mm; middle achenes cymbiform (3.7) 4.8–6.9 (10.1) × (2.1) 3.2–5.9 (8.9) mm, less usually bialate or trialate, sometimes with dorsal wings or spines; inner achenes vermiculate exalate (2.7) 3.4–4.1 (6.1) × (0.67) 1.2–1.6 (2.36) mm or with 2 lateral wings (2.9) 3.7–4.6 (7) × (0.93) 1.94–2.7 (4.7) mm, many times both, circular, hook-shaped or falcate. Pappus absent.

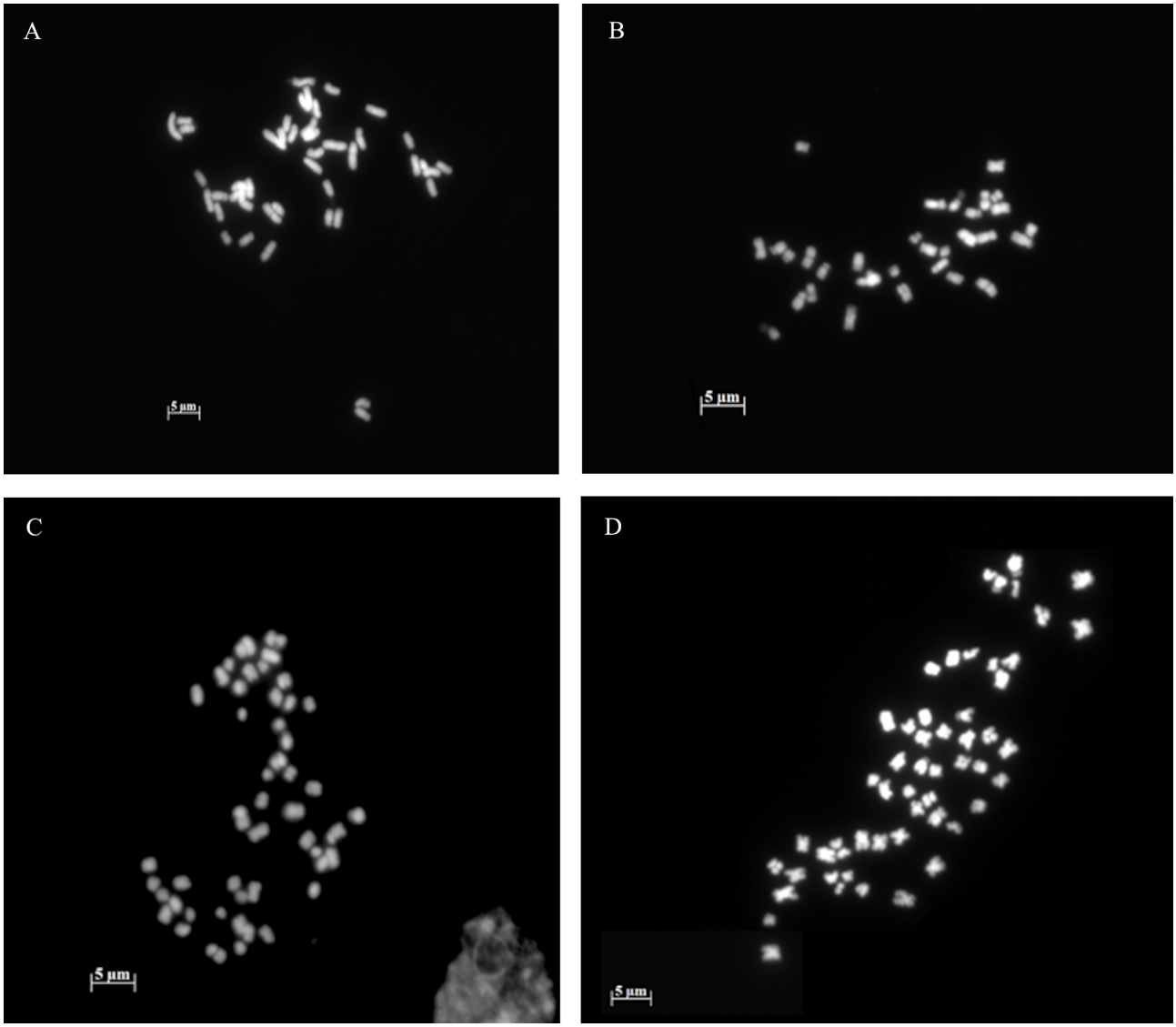

FIGURE 4. Metaphase somatic cells of (A) *Calendula arvensis* (*2n*=44; 3327.e); (B) *C. officinalis* (*2n*=32; 3409); (C) *C. sventenii* (*2n*=54; 3327.a); (D) *C. ricardoi* (*2n*=54; 3327g).

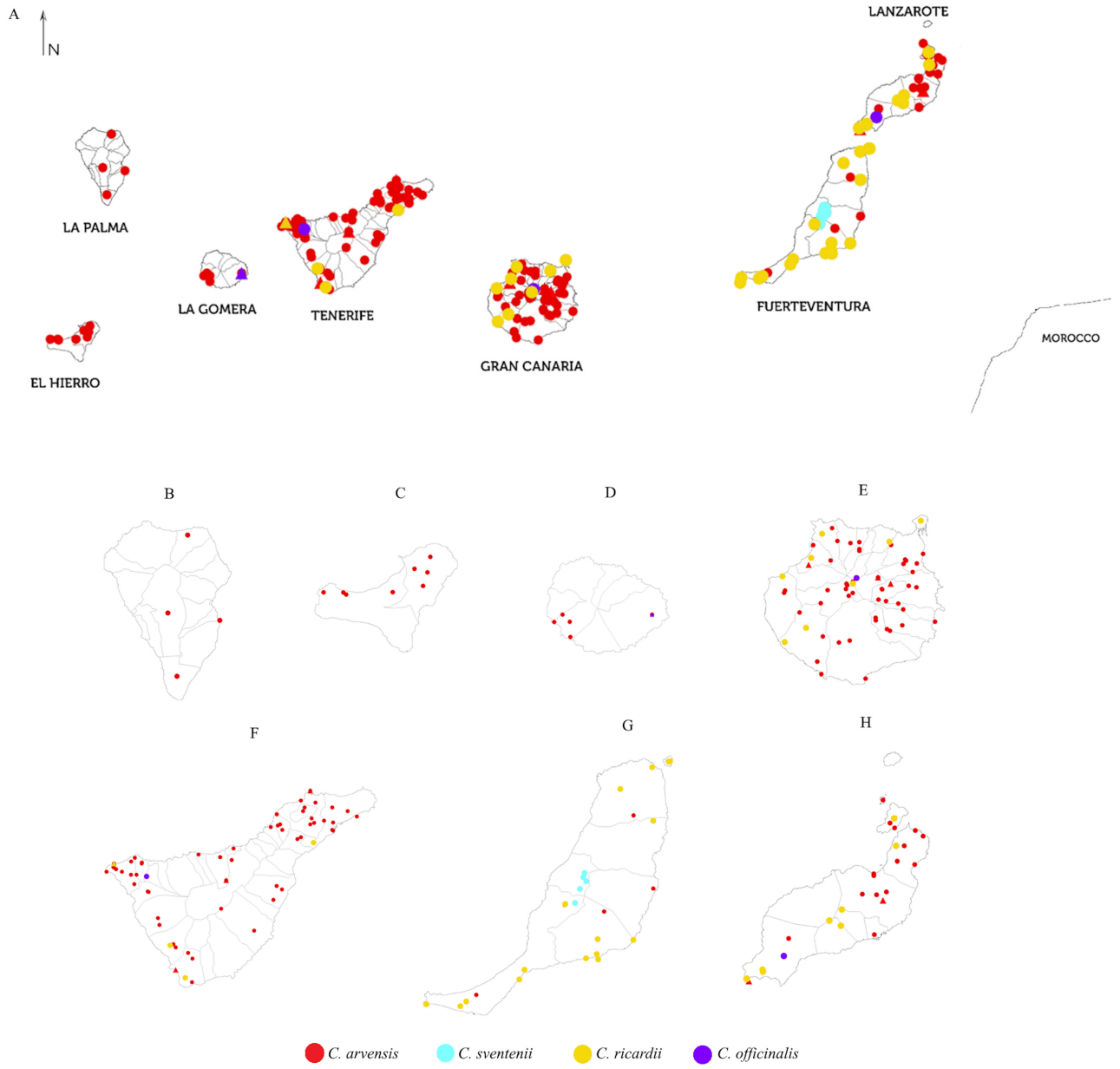

FIGURE 5. Geographic distribution of *Calendula* in the Canary Islands from herbarium specimens and from collected material in the 2014 and 2022 field expeditions. A. general view of the Canary Islands; B. La Palma; C. El Hierro; D. La Gomera; E. Tenerife; F. Gran Canaria; G. Fuerteventura; H. Lanzarote. The triangles represent herbarium specimens with a database entry, but whose identifications were not confirmed; the colour code is the same for triangles and circles; all forms of *C. arvensis* are under the same colour.

Key to the species of *Calendula* in the Canary Islands

1. Cymbiform achenes with a more or less developed ventral wing; disc florets at least 60 ..... 2. *C. officinalis*
– Cymbiform achenes without a ventral wing; disc florets up to 50, more frequently between 15–25 2
2. Outer achenes rostrate or bialate with incise-dentate lateral wings, never trialate ..... 1. *C. arvensis*
– Outer achenes rostrate and trialate with entire to sub-entire lateral wings 3
3. Capitula diameter (0.7) 0.9–1.1 (1.3) cm; rostrate achenes (4.9) 6.2–10.6 (13.5) mm long; leaves strictly linear-oblong ..... 4. *C. sventenii*
– Capitula diameter (1.4) 1.7–2.7 (3) cm; rostrate achenes (10.4) 13.7–22 (25) mm long; leaves ovate to oblong, less frequently oblanceolate ..... 3. *C. ricardoi*

1. *Calendula arvensis* Linnaeus (1763: 1304); Buch (1825: 151, 171; 1833: 497); Webb & Berthelot (1846: 341); Pitard & Proust (1908: 239); Lanza (1919: 112); Lindinger (1926: 170); Hansen & Sunding (1985: 20); Acebes-Genovés *et al.* (2001: 104; 2004: 131; 2010: 158); Beierkuhnlein *et al.* (2021: 21). Lectotype (designated by Heyn *et al.* (1974: 182)):—EUROPE. 'In Europae arvis', *Löfling s.n* (LINN! 1035.1).

Calendula aegyptiaca Persoon (1807: 492); Hansen & Sunding (1985: 20). Type:—EGYPT. Bords des champs, Matarych près de Caire, *L. Kralik s.n.* (holotype FI! 000477).

Calendula bicolor Rafinesque (1810: 82); Hansen & Sunding (1985: 20); Acebes-Genovés *et al*. (2001: 104; 2004: 131; 2010: 158). *Calendula arvensis* var. *bicolor* (Raf.) Candolle (1837: 452); Pitard & Proust (1908: 239). Type:—not found.

Calendula parviflora Rafinesque (1810: 83), *nom. illeg.*, non Thunberg (1800: 163). *Calendula arvensis* var. *parviflora* (Raf.) Battandier & Trabut (1888: 478); Pitard & Proust (1908: 239). Type:—not found.

Description:—Annual herbs. Stems (7.5) 18.5–33.4 (146) cm long, erect, branched at the base, covered in glandular and non-glandular hairs. Basal leaves usually (2.3) 4.1–6 (11.5) × (0.3) 0.7–1.2 (1.8) cm, (0.1) 0.2–0.3 (0.5) mm thick, oblanceolate, with a ± long petiole, acute to abruptly acute, margins sub-entire to repand, sometimes dentate, higher leaves usually smaller, oblanceolate to lanceolate, sessile and auriculate, possessing both glandular and non-glandular hairs. Capitula concolorous or discolorous, with (0.7) 0.9–2 (2.9) cm diameter, solitary. Involucre (3.3) 4.7–7.1 (8.7) mm, composed of one to two rows of bracts, lanceolate and acute, green with a ± defined purple apex, glandular pubescent. Ray florets 11–26, (4.4) 6.8–11.5 (18) × (0.64) 1.23–1.98 (2.58) mm, arranged in one row, yellow or orange and tri-lobed. Disc florets 18–20, (3) 3.1–3.7 (4) × (0.6) 0.8–1 (1.7) mm, five-lobed, yellow, brown, sometimes reddish, or even purple. Anthers 1.1–1.8 mm long, styles 1.95–2.4 mm, bifid. Outer achenes rostrate (4.7) 7.4–11 (20.3) × (0.9) 1.7–2.4 (3.7) mm, most of the times curved in a ± 90º angle and with a long rostrum, dorsal spines more or less developed, and one to two ventral teeth, one at the apex and one at the base, many times both, sometimes bialate, (4.1) 6.3–7.7 (10.4) × (1.5) 6.1–7.9 (9.3) mm, with dentate to incise lateral wings; sub-rostrate achenes (1.5) 4.3–5.3 (6.4) × (0.9) 1.4–1.9 (2.9) mm, with dorsal spines and two ventral teeth; middle achenes cymbiform (3.7) 4.8–6.8 (8.7) × (2.1) 3.2–5.8 (7.2) mm, slightly curved without ventral or dorsal wings but with ventral teeth and sometimes dorsal spines; inner achenes vermiform-alate (3.1) 3.7–4.4 (5.4) × (1) 1.9–2.5 (3.5) mm, circular to hook-shaped, with two lateral wings and ventral teeth; vermiform-exalate (2.7) 3.4–3.9 (4.9) × (0.8) 1.2–1.5 (2.2) mm.

Observations:—In the Canary Islands, at least four variations of *C. arvensis* can be found: the typical *C. arvensis*, with stems 15–40 cm long, external rostrate achenes, exalate, possessing, more or less, long beaks and capitula diameter 1–1.5 (2) cm; the *aegyptiaca* form (*C. aegyptiaca* Pers.), similar to the previous, but plants not exceeding 15–20 cm, with shorter and usually straighter rostrate achenes, and smaller capitula, resembling *C. tripterocarpa*, 0.5–1.5 cm in diameter; the *macroptera* form (*C. arvensis* var. *macroptera* (Rouy) O.Bolòs & Vigo), which possesses external bialate achenes with dentate lateral wings, and the *bicolor* form (*C. arvensis* var. *bicolor* Raf.), with discolourous capitula.

Habitat and Distribution:—All forms are densely dispersed throughout the islands, growing in a variety of habitats. Found in the margins of roads, at ruderal sites, agricultural fields, grasslands, rocky cliffs, near the beach in sandy soils, and in semi-desert to desert regions. Present in *Launaeo nudicaulis-Resedetum lancerotae*, *Raphanus raphanistrum-Scandix pecten-veneris, Convolvulus arvensis* community and *Raphanus raphanistrum-Scandix pecten-veneris* communities. Present in all islands.

Elevation:—From sea level to 1600 m.

Illustrations:—Figures 6, 10B and 11A.

Conservation status:—Least Concern (LC).

Chromosome number:—2n=44.

Genome size:—5.4 pg ± 0.2.

FIGURE 6. *Calendula arvensis* (Silveira, P., Simão, I. and Reyes-Bet. 3403, 3411). A, B. habit (3411 and observation on site, respectively); C, D, E. diversity of flower capitula (observation on site); F, G. diversity of fruiting capitula (3403 and observation on site, respectively).

2. *Calendula officinalis* Linnaeus (1753: 921); Hansen & Sunding (1985: 20); Acebes-Genovés *et al*. (2001: 104; 2004: 131; 2010: 158); Beierkuhnlein *et al*. (2021: 21). Lectotype (designated by Alavi (1983: 195)):—EUROPE. 'In Europae arvis', *Löfling s.n.* (LINN! 1035.4).

Description:—Annual or perennial herbs, sometimes woody at the base. Stems (25) 33–41 (50) cm long, occasionally longer, erect, branched at the base, covered with glandular and non-glandular hairs. Basal leaves usually (6.5) 8.8–10.6 (13.5) × (1.9) 2.3–2.7 (4.7) cm, (0.1) 0.2–0.3 (0.4) mm thick, oblanceolate to spatulate, with a ± long petiole, acute or obtuse, margins sub-entire to repand, higher leaves usually smaller, oblanceolate to lanceolate, sessile and auriculate, possessing both glandular and non-glandular hairs.

Capitula concolorous or discolorous, with (4) 4.6–5.7 (6.7) cm diameter, solitary. Involucre (9) 9.45–12.2 (14.3) mm, composed of one to two rows of bracts, linear-lanceolate, acute, green with a hyaline margin, glandular pubescent. Ray florets 30–45, (18.2) 20.8–25.2 (29.4) × (4.6) 5.5–6.4 (7.2) mm, arranged in one row, yellow or orange and tri-

lobed. Disc florets over 50, (3.8) 4–4.4 (4.7) × (1.4) 1.5–1.9 (2) mm, five-lobed, yellow, orange, or purple. Anthers 1.9–2.4 mm long, styles 3.2–4.3 mm, bifid.

Outer achenes rostrate (7.7) 11.8–13.2 (13.7) × (2.3) 2.6–3 (3.9) mm, most of the times only slightly curved with a long rostrum, sometimes with an 90º angle, small dorsal spines, and one to two ventral teeth, one at the apex and one at the base, many times both; middle achenes cymbiform (7.7) 8.1–9.3 (10) × (5.8) 6.7–8.1 (8.9) mm, slightly curved with a developed ventral wing, one or two ventral teeth; inner achenes vermiform-alate (4.9) 5.4–6.7 (7) × (1.8) 3.1–4.3 (4.7) mm, circular to hook-shaped, sometimes falcate, with two lateral wings and ventral teeth; vermiform-exalate (4.2) 4.6–5.4 (6.1) × (1.8) 1.9–2.1 (2.4) mm.

Habitat and Distribution:—Cultivated as an ornamental plant, sometimes escaping into ruderal grounds. Present in all major islands.

Elevation:—350–1500 m.

Illustrations:—Figures 7, 10E and 11D.

Chromosome number:—*2n*=32.

Genome size:—3.2 pg ± 0.06.

FIGURE 7. *Calendula officinalis* (Silveira, P., Simão, I. and Reyes-Bet. 3388, 3409). A, B. diversity of flowering capitula capitula (3388 and 3409, respectively); C. fruiting capitulum (3409).

3. *Calendula ricardoi* I.Simão, Reyes-Bet. & P.Silveira, ***sp. nov.*** Holotype:—CANARY ISLANDS. Fuerteventura: Road from Betancuria to Pájara, 30 March 1975, *P. L. Perez & J. R. Acebes 4802* (TFC! 4802).

Diagnosis:—This species is distinguishable from *C. tripterocarpa* by the well-developed rostrate achenes, usually with a long rostrum, and the larger size of the plants and leaves, and also by the flowering capitula with purple/reddish spots under the ligules. It is distinguishable from *C. arvensis* by the trialate achenes with entire to sub-entire lateral wings.

Description:—Annual herbs. Stems (32) 33–42 (50) cm long, occasionally longer, erect, branched at the base, with dispersed glandular and non-glandular hairs. Basal leaves usually (4.1) 4.9–6.9 (7.6) × (0.5) 0.95–1.3 (1.6) cm, (0.2) 0.3–0.4 (0.5) mm thick, ovate to oblong, less frequently oblanceolate, acute, margins entire to sub-entire, sometimes dentate, higher leaves similar to the basal ones but smaller, sessile, auriculate, possessing both glandular and non-glandular hairs. Capitula concolorous, (1.4) 1.7–2.7 (3) cm in diameter, solitary. Involucre (5.3) 6.1–8.1 (8.5) mm long, bracts in 1–2 rows, lanceolate, acute, green with a defined purple apex, glandular pubescent. Ray florets 13–21, (8.1) 9.6–14.9 (16.7) × (1) 1.9–2.3 (2.5) mm, arranged in one row, yellow or orange, sometimes with purple spots, tri-lobed. Disc florets 39–50, (2.5) 2.7–2.9 (3.7) × (0.6) 0.8–1 (1.2) mm, five-lobed, yellow to orange. Anthers 1.4–1.5 mm long, styles 1.9–2.3 mm, bifid. Achenes rostrate (10.4) 13.7–22 (25) × (1.8) 2.6–3.6 (5.1) mm, most of the times forming a ± 90º angle, sometimes only slightly curved, with a long rostrum and poorly developed dorsal spines, and one to two ventral teeth, one at the base or one at the apex, many times both; achenes trialate, (8) 8.6–10.5 (11.5) × (7.1) 8.1–9.5 (11.1) mm, lateral and ventral wings sub-entire, rarely dentate, flat in the back; achenes cymbiform (4.6) 5.1–6.1 (7) × (2.8) 3.5–4 (4.4) mm, strongly curved without ventral or dorsal wings but with ventral teeth, sometimes muricate in the back; achenes vermiform-alate (3.7) 3.9–4.5 (6) × (1.9) 2.3–2.7 (3.2) mm, circular to hook-shaped, with two lateral wings and ventral teeth; vermiform-exalate (3) 3.4–3.7 (5.1) × (1.1) 1.2–1.3 (1.5) mm.

Habitat and Distribution:—Limited to the central-western part of Fuerteventura, between Betancuria and Pájara, growing along the margins being part of ruderal nitrophilous and subnitrophilous weed communities. These annual communities are found in a degraded landscape characterized by shrubby Chenopodiaceae species or by better-preserved shrub communities dominated by succulent *Euphorbia* species.

Elevation:—250–400 m.

Illustrations:—Figures 8, 10D and 11C.

Conservation status:—Vulnerable (VU).

Chromosome number:—*2n*=54.

Genome size:—5.1 pg ± 0.7.

Etymology:—Named after Spanish botanist Ricardo Mesa Coello (b. 1954), who worked on the monitoring of the endangered flora of the Canary Islands, contributing to its conservation and better knowledge.

4. *Calendula sventenii* I.Simão, Reyes-Bet. & P.Silveira, ***sp. nov.*** Holotype:—CANARY ISLANDS. Lanzarote: Punta de Pechiguera, 13 February 2010, *A. Reyes-Betancort & R. Glez Glez 41487* (ORT! 41487).

Calendula aegyptiaca var. *platycarpa auct. non* Cosson (1857: 703), *nom. nud.*; Pitard & Proust (1908: 239).

Calendula tripterocarpa auct. non Ruprecht (1856: 231); Acebes-Genovés *et al.* (2010: 158); Beierkuhnlein *et al*. (2021: 21).

Calendula aegyptiaca subsp. *tripterocarpa auct. non* (Rupr.) Lanza (1919: 110); Acebes-Genovés *et al.* (2001: 104, 2004: 131).

Diagnosis:—This species resembles *C. tripterocarpa*, but differs from the latter by the well-developed rostrate achenes, usually with a long rostrum. It is distinguishable from *C. arvensis* by the trialate achenes with entire to sub-entire lateral wings, and a smaller size of the plant and capitula.

Description:—Annual herbs. Stems (11) 20–32.4 (41.5) cm long, ascending to erect, branched at the base, with dispersed glandular and non-glandular hairs. Basal leaves usually (2.4) 3.5–4.9 (6.4) × (0.3) 0.4–0.6 (1) cm, (0.1) 0.2–0.3 (0.4) mm thick, linear-oblong, sometimes with a small petiole, acute, margins sub-entire to repand, sometimes dentate, higher leaves usually smaller, lanceolate, amplexicaul, possessing both glandular and non-glandular hairs. Capitula concolorous, (0.7) 0.9–1.1 (1.3) cm in diameter, solitary. Involucre (3.5) 4.5–5.4 (6.3) mm long, bracts in one to two rows, lanceolate, apex acuminate, green, sometimes purplish when dry, glandular pubescent. Ray florets 8–20, (5.4) 6.6–7.4 (8.2) × (0.7) 0.9–1.1 (1.6) mm, arranged in one row, yellow-orange and tri-lobed. Disc florets 13–23, (2.4) 2.5–2.8 (3.5) × (0.7) 0.9–1 (1.1) mm, five-lobed, yellow to orange. Anthers 1–1.3 mm long, styles 1.5–2.15 mm, bifid. Achenes rostrate (4.9) 6.2–10.6 (13.5) × (1.3) 1.8–3.4 (3.9) mm, very variable, some strongly curved, others forming a ± 90º angle, with a long rostrum and poorly developed dorsal spines, and one to two ventral teeth at the base or at the apex; achenes trialate (5.7) 7–8.6 (9.1) × (5.2) 6.4–8 (10.4) mm, with lateral and ventral wings sub-entire to entire, flat

in the back, sometimes with dorsal spines; achenes cymbiform (3.9) 4.4–4.9 (5.4) × (2.1) 2.6–3.5 (3.9) mm, strongly curved without ventral or dorsal wings but with ventral teeth and dorsal spines, sometimes only muricate in the back; achenes vermiform-alate (2.9) 3.4–3.7 (4.1) × (0.9) 1.4–2.2 (2.8) mm, circular to hook-shaped, less frequently falcate, with two lateral wings and ventral teeth; achenes vermiform-exalate (2.8) 3.2–3.6 (3.9) × (0.7) 1.1–1.2 (1.3) mm.

Habitat and Distribution:—Growing along the margins of roads, at ruderal sites in semi-desert to desert regions, and in agricultural fields. The natural habitat of this species are non-nitrophilous herb communities (*Tuberarietea guttatae*) associated with the *Euphorbia canariensis* and *Euphorbia balsamifera* shrubblands, reaching the lower areas of the *Juniperus canariensis* woodlands. Also, it can grow in sandy habitats as part of the *Resedo lanceolatae-Moricandion* communities associated with those of the class *Polycarpaeo-Traganetea moquinii*.

FIGURE 8. *Calendula ricardoi* (Silveira, P., Simão, I. and Reyes-Bet. 3327.j, 3327.l). A. habit (observation on site); B. flowering and fruiting capitula (3327.j); C. fruiting capitulum (3327.l).

Only confirmed for La Graciosa, Lanzarote, Fuerteventura, Gran Canaria and Tenerife.

Elevation:—10–450 m.

Illustrations:—Figures 9, 10C and 11B.

Conservation status:—Least Concern (LC).

Chromosome number:—2n=54.

Genome size:—3.7 pg ± 0.16.

Etymology:—Named after Eric Sventenius (1910–1973), Swedish botanist, who dedicated his work to the Macaronesian Flora, first working in the Jardín de Aclimatación de La Orotava and then founding the Jardín Botánico Canario Viera y Clavijo.

FIGURE 9. *Calendula sventenii* (Silveira, P., Simão, I. and Reyes-Bet. 3383, 3384.2, 3391). A. habit (3384.2); B, C. flowering capitula (3391); D. fruiting capitulum (3383).

FIGURE 10. Variability of achene morphology of *Calendula*. A. *C. arvensis* form *arvensis* (Silveira, P. and Gonçalves, A.C.R.S. 3327.e); B. *C. arvensis* form *macroptera* (Silveira, P. and Gonçalves, A.C.R.S. 3327.c); C. *C. sventenii* (Silveira, P. and Gonçalves, A.C.R.S. 3327. a); D. *C. ricardoi* (Silveira, P. and Gonçalves, A.C.R.S. 3327.g); E. *C. officinalis* (Silveira, P. 2986).

Discussion

Chromosome number and genome size

Basic chromosome number reports for the genus have varied from x=7, 8, 9, 11, and 15, with x=9 being considered the main basic number (Norlindh 1977, Heyn & Joel 1983). Chromosome numbers are also diverse, with *2n*= 14, 18, 30, 32, 44 to 88 reported for different species of *Calendula* (Nora *et al*. 2013). Prior to this work, *Calendula* chromosome reports for the Canary Islands were very limited (Dalgaard 1986, Baltisberger & Widmer 2006), and included only two species. Chromosome data from this study (Figure 4) is consistent with these reports for *C. arvensis 2n*=44 and for the taxon with *2n*=54, although the latter does not correspond to *C. tripterocarpa* as originally stated but to a new species described in this work, *C. sventenii*.

Several authors discussed the positive correlation between genome size and chromosome number (Srivastava & Lavania 1991; Soliman 2003), and the same trend was observed in several *Calendula* taxa from other territories (Nora *et al.* 2013; Gonçalves 2018).

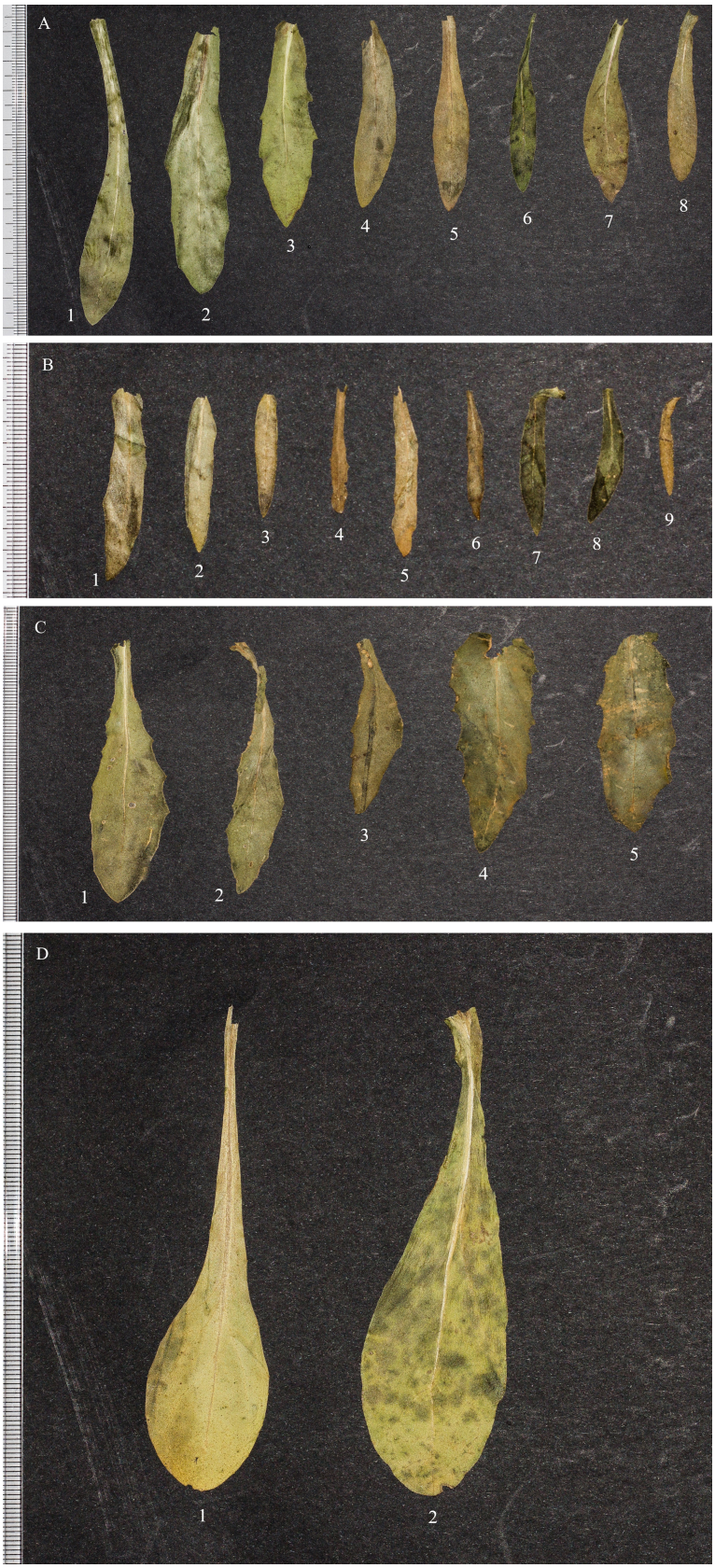

FIGURE 11. Variability of leaf morphology of *Calendula*. A. *C. arvensis* (1. Reyes-Bet. *in* P. Silveira 3426.3; 2. Reyes-Bet. *in* P. Silveira 3426.5; 3. Reyes-Bet. *in* P. Silveira 3119.1; 4. Silveira, Simão and Reyes-Bet. 3413.2; 5. Silveira, Simão and Reyes-Bet. 3413.3; 6. Silveira, Simão and Reyes-Bet. 3412.2; 7, 8. Silveira, Simão and Reyes-Bet. 3397.7); B. *C. sventenii* (1. Silveira, Simão and Reyes-Bet. 3383.2; 2, 3. Silveira, Simão and Reyes-Bet. 3380; 4–6. Silveira, P. and Gonçalves, A.C.R.S. 3327.a; 7–9. Silveira, P. and Gonçalves, A.C.R.S. 3327. b); C. *C. ricardoi* (1, 2. Silveira, P. and Gonçalves, A.C.R.S. 3377; 3. Silveira, P. and Gonçalves, A.C.R.S. 3327.i.4; 4, 5. Silveira, P. and Gonçalves, A.C.R.S. 3327.l); D. *C. officinalis* (1. Silveira, Simão and Reyes-Bet. 3421.1; 2. Silveira, Simão and Reyes-Bet. 3409).

However, in the Canary Islands, the species with the highest chromosome numbers, *C. sventenii* and *C. ricardoi* (both with *2n*=54) possess a lower genome size than most *C. arvensis* populations (*2n*=44). This may indicate that the Canary Islands taxa represent a distinct evolutionary line from *C. arvensis* and other *Calendula* species, which might be plausible considering these representing endemic insular species, while *C. arvensis* is known to be a taxon with a large distribution range.

An intraspecific genome size variability was observed in *C. ricardoi*, with 2C-values fluctuating from 3.7—5.3 pg, regardless of no significant morphologic distinction. The scarcity of analyzed specimens, namely in the rural park of Betancuria, a natural protected area in Fuerteventura, hindered the effort of obtaining an accurate genome size for *C. ricardoi*. Nevertheless, all individuals collected from the population share *2n*=54.

This is not the first report of genome size variability for the genus. Statistically significant differences in 2C-value were reported for *C. suffruticosa*, a taxon from the Iberian Peninsula with several subspecies, reflecting intraspecific variation (Nora *et al*. 2013, Silveira *et al*. 2013). Nora (2009) also reported a DNA content of 6.5 pg for a population of *C. arvensis* from Pujaire, Andalucía. The presence of supernumerary B chromosomes, which were reported in the genus (Oberprieler & Vogt 1993, Vogt & Oberprieler 2008) and are common in the Asteraceae (Camacho 2005), has been considered a contributing factor for intraspecific genome size variation (Nora *et al*. 2013), although, in this work, they were never observed in the analyzed specimens. The presence of secondary metabolites may also cause artefactual variation in genome size. Yet, the samples with CV% ≤ 5% discard this option as the main reason for the 2C-value fluctuation (Loureiro *et al*. 2006).

An intrapopulation genome size variability has also been reported for taxa of other genera of the Asteraceae family, such as *Picris, Hieracium* and *Centaurea* (Suda *et al*. 2007, Slovák *et al*. 2009; Dydak *et al*. 2009). It seems there are specific groups that are more likely candidates for genome size variation, such as long-term isolated populations, which may be the case for *C. ricardoi* (Slovák *et al*. 2009).

Distribution maps, species richness and ecology

Distribution maps for the *Calendula* species are presented in Figure 5. The list of specimens used can be consulted in Appendix III.

Tenerife and Gran Canaria were the islands with the highest amount of information regarding *Calendula* specimens. In most of the territory, *C. arvensis* appears as dominant taxa, growing in a variety of habitats, on roadsides, in ruderal sites, on agricultural fields, grasslands, rocky cliffs, near the seashore on sandy soils, and in semi-desert to desert regions, from sea level to 1600 m. *Calendula sventenii* emerges predominantly in the oldest islands, Fuerteventura, and Lanzarote, up to 450 m, at ruderal sites in semi-desert to desert regions, and in agricultural fields. *Calendula officinalis* is cultivated as an ornamental plant, sometimes escaping to ruderal grounds.

Unlike the other insular taxa, *C. ricardoi* was only found in the central-western part of Fuerteventura, between Betancuria and Pájara, at elevations of 250–400 m. The higher regions of Fuerteventura, such as the Betancuria massif (but also the Jandía peninsula) have been associated with the higher average rainfall values on the island (Herrera & Custodia 2014, Bechtel 2016). Considering the limited distribution of this taxon, with most of its individuals occurring at elevations higher than 400 m, the annual precipitation may be a defining factor in the establishment of new populations.

Other species previously recorded from the islands (Beierkuhnlein *et al*. 2021), such as *C. tripterocarpa*, *C. stellata* and *C. suffruticosa*, were not observed either in the expeditions or in herbarium material, and as such, were not shown in the maps. Regarding these taxa, herbarium specimens and literature were consulted in order to elaborate a list of sites to visit and check for the taxa that were given for the territory. The few herbaria specimens which we had access to and which were identified as *C. stellata* (ORT 30212) and *C. suffruticosa* (TFC 48.837), were misidentified and in fact belong to *C. arvensis*. *Calendula suffruticosa*, specifically, was considered to exist in only two sites in the Canary Islands, in Tacoronte and in a cliff area in Punta de Teno, Tenerife. However, upon visit, only specimens of *C. arvensis* were found. It is worth noting that several specimens of *C. arvensis* from Tenerife were bigger than the typical form, almost resembling the perennial taxa because of their size, with ligules more than two times the length of the involucre, one of the characteristics associated with *C. suffruticosa*. The nuclear DNA content of these specimens (AVE, ORT 3397) was analyzed and did not correspond to the reported 2C-value for the *C. suffruticosa* group (3.2 pg ± 0.11-3.4 pg ± 0.13) (Nora *et al*. 2013), but corresponded to *C. arvensis* (5.5 pg ± 0.04).

Although we cannot state for sure that *C. suffruticosa* never existed in the area, we can suspect that perhaps these bigger specimens of *C. arvensis* might have been misidentified in the past as *C. suffruticosa* due to their morphologic variability. Another possibility to explain the records of *C. suffruticosa* in the island is that the species may have

existed, at some point, at least in Punta de Teno, but because of its restricted habitat combined with the heavy tourism pressure (Bramwell 1971, Caujapé-Castells *et al*. 2010) may have gone locally extinct with time. *Calendula stellata* also was not spotted during the field expedition and may have been misidentified in the floristic checklists as its specimens in herbarium collections appeared to belong to *C. arvensis* and *C. arvensis* forma *macroptera* specimens with bigger discolorous capitula, which are not big enough to be considered belonging to *C. stellata*.

Possible origins and mechanisms of speciation

The region of SW Morocco corresponds to the closest continental mass to the Canary Islands, with 96.5 km separating the African coast from Fuerteventura (Arco-Aguilar & Rodríguez-Delgado 2018). The biogeographical and floristic similarities between the Canary Islands and the coastal and subcoastal areas of SW Morocco have led some authors to consider these regions as part of a "Moroccan-Macaronesian sector" (Peltier 1982, Kim *et al*. 1996), although, at the moment, the SW Moroccan region is not considered part of the Macaronesia (Médail & Quézel 1999, Msanda *et al*. 2021). Regardless of the biogeographical classification, this proximity has had evident consequences regarding plant colonization: the Moroccan area played a fundamental role as a plant-source territory for the Canary Islands, especially to those closest to the mainland, such as Fuerteventura and Lanzarote. Molecular studies on the origin of several Macaronesian plants have shown this relation (Msanda *et al*. 2021). Although the scope of this work is not the origin of the *Calendula* in the Canary Islands, for which a molecular phylogenetic study would be needed, we established two possibilities for the origin of the ancestors of the Canary Islands *Calendula*.

The first option would be the colonization of the easternmost islands from SW Morocco by long-distance dispersal. This phenomenon has been reported for other taxa; for example, pollen transport has been reported from Morocco to Tenerife through trade winds (Izquierdo *et al*. 2011). Furthermore, the Canaries often suffer from a meteorological event called "Calima" (dust invasions from the Saharan desert), which has a big influence on the climate of the islands closest to the African coast (Morales *et al*. 2009), but could also have functioned, at some point, as a transport vector for *Calendula*. In this scenario, the *Calendula* heterocarpy might have been fundamental for the establishment of populations in the islands since the presence of several dispersal syndromes confers colonization advantage to species (Vargas *et al*. 2015).

The second option for the origin of the genus in the islands would be the introduction of the ancestor taxa by men, starting in the 15th century with the European conquest and settlements of colonists in the Canaries (Arco-Aguilar & Rodríguez-Delgado 2018) or maybe even before this period, by the aboriginal peoples (Guanches) who came to the Canaries from Europe and Africa (Arnaiz-Villena *et al*. 2015). As already mentioned, the first records of *Calendula* in the Islands date back to the beginning of the 19th century when *C. arvensis* was considered an introduction (Buch 1819, 1833). Since more than 50% of the present vascular flora of the Canary Islands is introduced (Arco-Aguilar & Rodríguez-Delgado 2018), *Calendula* might have been part of the plants brought by the Spanish trade routes to the islands, either intentionally or by accident. It is worth noting that this second hypothesis is based on the flora records we have access to for the genus in the islands, which means these introductions may have taken place earlier than the 15th century. The similarities found between the taxa from the Canaries that occur in the oldest islands, and *C. pinnatiloba* and *C. lanzae* from Morocco, particularly the morphology of the achenes, also seem to possibly point out to an earlier period of colonization.

Once established, the spatial isolation and reduced gene flow between the continent and the islands favored the beginning of the gradual speciation process that took place and resulted in chromosomal rearrangements, morphological differentiation, and, at some point in time, two new taxa: *C. sventenii* and *C. ricardoi*. This process was certainly promoted by several attributes of *Calendula*: small populations, self-pollination capacity, and a short generation time (Crawford & Archibald 2016).

Conclusions

With this work we present the first taxonomic revision of *Calendula* for the Canary Islands focused on morphologic and genetic information, including four taxa, two of them newly described species.

Calendula ricardoi and *C. sventenii* are considered two separate taxa based on their morphologic differences but also their ecological preferences and distribution patterns. The chromosome counts for *C. sventenii* (*2n*=54) helped to corroborate its distinction as a species separate from *C. tripterocarpa* (*2n*=30), confirming the reported *2n* for *C. sventenii* plants cited in the literature.

Genome size estimates for the Canary Islands *Calendula* are also reported for the first time. Intraspecific variation in genome size was detected in the population of *C. ricardoi*, without morphological differences being detected. Further research focused on the genome size of *C. ricardoi* is needed to confirm the 2C-values reported for the species in this work.

For genera with the level of complexity of *Calendula*, there is an increasing interest in the use of genetic information such as nuclear DNA content and chromosome numbers in the establishment of meaningful taxonomic characters that distinguish taxa. However, although they are useful tools, genetic information cannot be used isolated from the morphological and ecological analysis of the specimens, which continues to be the best approach for characterization of species within the genus.

A future molecular phylogenetic study will be fundamental to better unravel the relationships between taxa and to understand how *Calendula* evolved in the Canary Islands.

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Disclosure statement

The authors report there are no competing interests to declare.

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APPENDIX I

Results of the morphometric analysis of *Calendula* from the Canary Islands.

	C. arvensis						*C. sventenii*						*C. ricardoi*						*C. officinalis*					
Characters	**N**	**Min.**	**25%**	**Mean**	**75%**	**Max.**	**N**	**Min.**	**25%**	**Mean**	**75%**	**Max.**	**N**	**Min.**	**25%**	**Mean**	**75%**	**Max.**	**N**	**Min.**	**25%**	**Mean**	**75%**	**Max.**
LB	88	7.5	18.5	26.9	33.4	146.0	19	11.0	19.7	24.8	32.4	41.5	9	32.0	33.6	36.9	42.0	49.5	15	25.0	32.8	36.0	41.1	50.3
LL	88	22.9	40.8	50.2	60.4	115.0	18	23.9	35.5	40.3	49.0	63.8	15	41.2	48.6	55.1	69.4	135.0	15	65.0	88.0	100.0	105.5	135.0
LW	87	3.4	6.6	9.9	12.2	18.1	18	3.4	4.4	5.5	6.4	12.0	15	4.9	9.5	12.0	13.1	15.8	15	19.0	21.0	24.0	27.0	47.0
LD	87	8.3	26.5	33.5	40.5	72.2	18	14.9	22.5	27.2	32.5	40.8	15	9.3	29.2	33.0	43.6	55.2	15	50.0	65.0	85.0	85.5	100.0
R1	87	0.8	1.3	1.5	1.6	4.3	18	1.2	1.5	1.6	1.6	2.1	15	1.4	1.5	1.6	1.9	4.5	15	1.2	1.2	1.3	1.3	1.4
R2	87	2.4	4.2	5.5	6.7	13.3	18	3.8	5.6	6.4	9.7	13.3	15	3.5	4.3	5.4	6.1	8.9	15	2.8	3.5	4.0	4.3	4.5
LT	87	0.1	0.2	0.2	0.3	0.5	19	0.1	0.2	0.2	0.3	0.4	16	0.2	0.3	0.4	0.4	0.5	15	0.1	0.2	0.2	0.3	0.4
HD	87	7.1	9.8	14.4	20.1	29.0	19	6.5	9.2	10.0	10.7	13.2	15	14.3	16.7	19.3	26.6	30.0	15	40.0	45.5	52.0	56.5	67.0
IL	87	3.3	4.7	6.0	7.2	8.7	19	3.5	4.5	5.0	5.4	6.3	15	5.3	6.1	7.5	8.1	8.5	15	9.0	9.5	10.5	12.2	14.3
LG	87	4.4	6.8	8.6	11.5	18.0	19	5.4	6.7	7.0	7.4	8.2	15	8.1	9.6	14.0	14.9	16.7	15	18.2	20.8	23.5	25.2	29.4
R3	87	1.0	1.4	1.5	1.7	2.2	19	1.2	1.2	1.3	1.6	2.0	15	1.4	1.7	1.8	1.9	2.1	15	1.7	1.9	2.1	2.4	2.8
RL	64	4.7	7.4	9.0	11.0	20.3	18	4.9	6.2	9.1	10.6	13.5	15	10.4	12.4	19.7	22.0	24.9	15	7.7	11.8	12.6	13.2	13.7
RW	64	0.9	1.7	2.1	2.4	3.7	18	1.3	1.8	2.8	3.4	3.9	15	1.8	2.6	3.0	3.6	5.1	15	2.3	2.6	2.8	3.0	3.9
RS	64	0.7	1.3	1.6	1.9	3.9	18	0.7	1.3	1.8	2.3	3.9	15	0.9	1.2	1.9	2.2	2.5	-	-	-	-	-	-
SRL	40	1.5	4.3	4.8	5.3	6.4	15	2.2	4.1	4.5	5.0	6.1	-	-	-	-	-	-	-	-	-	-	-	-
SRW	40	0.9	1.4	1.6	1.9	2.9	15	0.7	0.9	1.0	1.4	1.7	-	-	-	-	-	-	-	-	-	-	-	-
SRS	40	0.6	0.9	1.2	1.6	2.5	15	0.6	1.2	1.4	1.6	3.8	-	-	-	-	-	-	-	-	-	-	-	-
BL	32	4.1	6.3	6.9	7.7	10.4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
BW	32	1.5	6.1	7.1	7.9	9.3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
TL	-	-	-	-	-	-	19	5.7	7.0	7.8	8.6	9.1	15	8.0	8.6	8.9	10.5	11.5	-	-	-	-	-	-
TW	-	-	-	-	-	-	19	5.2	6.4	7.3	8.1	10.4	15	7.1	8.1	8.6	9.5	11.1	-	-	-	-	-	-
CL	71	3.7	4.8	6.0	6.8	8.7	19	3.9	4.4	4.7	4.9	5.4	19	4.6	4.8	5.3	6.1	7.0	15	7.7	8.1	8.8	9.3	10.1
CW	71	2.1	3.2	4.1	5.8	7.2	19	2.1	2.6	3.0	3.5	3.9	19	2.8	3.5	3.7	4.0	4.4	15	5.8	6.7	7.7	8.1	8.9
VAL	82	3.1	3.7	4.0	4.4	5.4	19	2.9	3.4	3.5	3.7	4.1	16	3.7	3.9	4.1	4.5	6.0	15	4.9	5.4	6.3	6.7	7.0
VAW	82	1.0	2.0	2.2	2.5	3.5	19	0.9	1.4	1.7	2.2	2.8	16	1.9	2.3	2.4	2.7	3.2	15	1.8	3.1	3.6	4.3	4.7
VEL	83	2.7	3.4	3.7	3.9	4.9	15	2.8	3.2	3.3	3.6	3.9	15	3.0	3.4	3.5	3.7	5.1	15	4.2	4.6	4.7	5.4	6.1
VEW	83	0.8	1.2	1.4	1.5	2.2	15	0.7	1.1	1.1	1.2	1.3	15	1.1	1.2	1.2	1.3	1.5	15	1.8	1.9	2.0	2.1	2.4

APPENDIX II
Herbarium specimens used in the statistical analysis.

***Calendula arvensis* L.**
SPAIN. **Canary Islands**
Fuerteventura: along FV-102 towards La Caldereta (28°35'05" N, 13°51'49" W), 133m, 26-3-2022, *Silveira, P., Simão, I.* 3371 (AVE, ORT); Betancuria (28°25'42" N, 14°03'29" W), 446m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* .i.2/.3 (AVE); FC-30 towards Pájara, near Sra. de La Peña (28°23'46" N, 14°04'02" W), 308m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.k (AVE); agricultural fields near Montaña del Frontón (28°35'59" N, 13°55'06" W), 241m, 26-3-2022, *Silveira, P. Simão, I.* 3373/4/5 (AVE).
Gran Canaria: Botanical Canarian Garden Viera y Clavijo (28°03'50" N, 15°27'45" W), 330m, 27-3-2022, *Silveira, P., Simão, I.* 3382.1/.2/.3/.4/.5 (AVE, ORT); Ingenio. El Roque (27°56'52" N, 15°28'49" W), 855m, 10-4-2022, *Reyes-Betancort, J. A. 3.3/.4/.5 In Silveira, P.* 3420.3/.4/.5 (AVE, ORT); Valsequillo. Edge of the boiler of "los Marteles" (27°57'31" N, 15°31'58" W), 1546m, 10-4-2022, *Reyes-Betancort, J. A. 4 In Silveira, P.* 3421 (AVE); Las Mesas Altas (28°06'08" N, 15°29'57" W), 9-4-2022, *Reyes-Betancort, J. A. 1.1/.2/.3 In Silveira, P.* 3418.1/.2/.3 (AVE, ORT); Mogán. Cortadores (27°51'41" N, 15°40'44" W), 726m, 30-4-2022, *Reyes-Betancort, J. A. 11.2/.3/.4/.5 In Silveira, P.* 3428.2/.3/.4/.5 (AVE, ORT).
Lanzarote: Punta Mujeres (29°08'19" N, 13°27'18" W), in a ditch by the side of the road, 29-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.c/d/e (AVE); Órzola (29°12'32" N, 13°26'02" W), in the sands of the roadsides and paths, 29-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.f (AVE); surroundings of Playa Blanca (28°52'24" N, 13°49'54" W), 42m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3384.1 (ORT); Teguise (29°03'50" N, 13°32'59" W), 321m, 30-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3389.1 (AVE); Guinate viewpoint (29°18'46" N, 13°50'07" W), 354m, 30-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3392.1 (AVE); El Jable (29°03'55" N, 13°35'05" W), 145m, 30-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3394 (AVE).
Tenerife: C. del Calvario between Tacoronte and Lomo Colorado (28°29'00" N, 16°24'22" W), 479m, 31-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3397.2/.4/.5/.6/.7 (AVE, ORT); La Laguna (28°30'04" N, 16°18'46" W), 532m, 31-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3398.1/.2 (AVE, ORT); Bejía (28°32'53" N, 16°18'04" W), 575m, 31-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3400/3401 (AVE, ORT); street near Buenavista (28°23'00" N, 16°50'18" W), 47m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3405 (AVE, ORT); Buenavista del Nte. (28°22'21" N, 16°50'58" W), 99m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3407/3408 (AVE, ORT); TF-375 near Las Manchas (28°16'55" N, 16°47'51" W), 1054m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3410 (AVE); TF-82 near Barranco de Ramallo (28°12'14" N, 16°46'10" W), 626m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3412.1/.2/3413.1/.2 (AVE, ORT); Tejina de Isora (28°11'01" N, 16°45'55" W), 553m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3414 (AVE, ORT); fields along Av. el Palm-Mar (28°01'35" N, 16°41'18" W), 81m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3415.1/3416.1/.2 (AVE, ORT); ascent to Teno Alto through the old hermitage of San Mateo in Teno Bajo (28°21'19" N, 16°54'04" W), 292m, 22-4-2022, *Reyes-Betancort, J. A. 9.1 In Silveira, P.* 3426.1 (AVE, ORT); (28°21'21" N, 16°54'05" W), 247m, 22-4-2022, *Reyes-Betancort, J. A. 9.3/.4 In Silveira, P.* 3426.3/.4 (AVE, ORT); (28°21'22" N, 16°54'07" W), 207m, 22-4-2022, *Reyes-Betancort, J. A. 9.5 In Silveira, P.* 3426.5 (AVE, ORT); Teno Alto. Descent to las Cuevas (28°20'58" N, 16°53'39" W), 561m, 22- 4-2022, *Reyes-Betancort, J. A. 10 In Silveira, P.* 3427 (AVE, ORT).

***Calendula officinalis* L.**
SPAIN. **Canary Islands**
Gran Canaria: Valleseco. Sobre Mesas de Galaz (28°00'54" N, 15°35'25" W), 1587m, 10-4-2022, *Reyes-Betancort, J. A. 6.1 In Silveira, P.* 3423.1 (AVE, ORT).
Tenerife: Near Puerto de Erjos (28°19'41" N, 16°48'12" W), 1007m, 1-4-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3409 (AVE, ORT).

***Calendula ricardoi* I.Simão, Reyes-Bet. & P.Silveira**
SPAIN. **Canary Islands**
Fuerteventura: Betancuria (28°26'24" N, 14°03'15" W), 29-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.g (AVE); (28°25'42" N, 14°03'29" W), 446m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.i.1/.4 (AVE); FC-30 towards Pájara, near Sra. De La Peña (28°23'46" N, 14°04'02" W), 308m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.j

(AVE); Pájara (28°21’25” N, 14°04’51” W), 262m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.l.1 (AVE).

Calendula sventenii **I.Simão, Reyes-Bet. & P.Silveira**
SPAIN. **Canary Islands**:
Fuerteventura: Corralejo (28°44’ N, 13°52’ W), 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.a, b (AVE); Gran Tarajal (28°15’23” N, 14°01’04” W), 91m, 26-3-2022, *Silveira, P., Simão, I.* 3380, 3381 (AVE, ORT).
Lanzarote: Punta Pechiguera (28°51’22” N, 13°52’18” W), 18m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3383.1/.2/.3/.4/.5/.6 (AVE, ORT); surroundings of Playa Blanca (28°52’42” N, 13°49’57” W), 49m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3385.2 (AVE); (28°52’41” N, 13°49’58” W), 49m, 29-3-2022, *Silveira, P.. Simão, I. & Reyes-Betancort, J. A.* 3387.1/.2 (AVE, ORT).
Tenerife: fields along Av. el Palm-Mar (28°01’35” N, 16°41’18” W), 81m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3415.5 (AVE, ORT).

Other selected specimens studied.
Calendula pinnatiloba **(Cosson ex Maire) A.C. Gonç. & P. Silveira**
MOROCCO.
Agadir: (30°36’ N, 9°46’ W), 27m, 10-4-2011, *Silveira P.* 3130 (AVE); (30°37’34” N, 9°51’27” W), 12m, 10-4-2011, *Silveira P.* 3134 (AVE); (29°36’50” N, 10°01’25” W), 256m, 11-4-2011, *Silveira P.* 3138 (AVE).

Calendula lanzae **Maire**
MOROCCO.
Taroudant: (30°28’35” N, 8°48’03” W), 257m, terrace on the left side of Sous River 26-3-2013, *Silveira, P., Gonçalves, A.C.R.S., Ouhammou, A.* 3292 (AVE); Tiout (30°23’48” N, 8°42’17” W), 424m, 26-3-2013, *Silveira, P., Gonçalves, A.C.R.S., Ouhammou, A.* 3293a (AVE).

Calendula stellata **Cav.**
MOROCCO.
Khemisset: Oued Beht (33°52’47” N, 5°55’49” W), 132m, 8-5-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3329 (AVE).
Zaio: Zaio (34°59’52” N, 2°49’47” W), 213m, 12-5-2014, *Silveira, P. Gonçalves, A.C.R.S.* 3337 (AVE).

Calendula tripterocarpa **Rupr.**
MOROCCO.
Marrakech: Jebilet (31°52’09” N, 7°57’08” W), 624m, 25-3-2013, *Silveira, P., Gonçalves, A.C.R.S., Ouhammou, A.* 3282 (AVE); Menara (31°35’00” N, 8°00’44” W), 25-3-2013, *Silveira, P., Gonçalves, A.C.R.S., Ouhammou, A.* 3283 (AVE).
Taroudant: (30°28’32” N, 9°02’05” W), 176m, 26-3-2013, *Silveira, P., Gonçalves, A.C.R.S., Ouhammou, A.* 3289 (AVE); (30°06’41” N, 8°27’50” W), 1656m, 26-3-2013, *Silveira, P., Gonçalves, A.C.R.S., Ouhammou, A.* 3296 (AVE).

APPENDIX III

List of specimens consulted for the elaboration of the distribution maps[1].

***Calendula arvensis* L.**

SPAIN. **Canary Islands**

Fuerteventura: Pozo Negro (28°20' N, 14°00' W), 31-7-1858, Lowe, R.T. 154 (BM); Gran Tarajal (28°12' N, 14°01' W), 0m, waste ground, 3-4-1936, Brooke, W.M.A. 323 (BM); (28°12'53.30"N, 14° 1'13.04"W)*, 15-3-1946, Sventenius, E. R. (ORT); Castillo (28°23'48.48"N, 13°51'49.76"W)*, 29-3-1956, *Sventenius, E. R.* 21583 (ORT); Lobos (28°44'59.55"N, 13°49'22.27"W)*, 24-3-1956, *Sventenius, E. R.* 24004 (ORT); Corralejo (28°44' N, 13°52' W), 23-3-1986, *Hansen, A.* 208 A (C); Pájara, Jandía, Pico de la Zarza (28° 6'6.99"N, 14°21'20.19"W)*, 26-2-1986, *Navarro, B. Montelongo, V.M.* 14150 (LPA); Pájara, La Lajita (28°21'3.17"N, 14° 6'35.08"W)*, 0-50m, 30-3-1989, *Montelongo, V.M.* 017596 (LPA); along FV-102 towards La Caldereta (28°35'05" N, 13°51'49" W), 133m, 26-3-2022, *Silveira, P., Simão, I.* 3370/3371 (AVE, ORT); Betancuria (28°25'42" N, 14°03'29" W), 446m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.i.2/.3/.5/.6 (AVE, ORT); FC-30 towards Pájara, near Sra. de La Peña (28°23'46" N, 14°04'02" W), 308m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.k (AVE); agricultural fields near Montaña del Frontón (28°35'59" N, 13°55'06" W), 241m, 26-3-2022, *Silveira, P. Simão, I.* 3373/4/5 (AVE);

Gran Canaria: Lomitos de Correa (28°00' N, 15°30' W), 16-6-1889, *Buysman, M.* 341 (COI); San Mateo (28°01' N, 15°32' W), 1927, *Bannerman, D.A.* s.n. (BM); 18-4-1935, *Tejida, J.* 1196 (MAF); 1000m, 18-3-1970, *Borgen, L.* 30 (O); Arucas (28°00' N, 15°36' W), 21-3-1921, *Rahn, K.* 743 (C); ?-12-1964, *Pedersen, A.* s.n. (C); 30-12-1964, *Hansen, A.* s.n. (C); 6-1-1965, *Kaae, N.* 1 (C); 18-12-1972, *Kaae, N.* 1 (C); 21-12-1972, *Kaae, N.* 2 (C); 20-12-1972, *Kaae, N.* 3 (C); 13-4-1973, *Hansen, A.* s.n. (C); edge of ditch on the hillside, 15-1-1974, *Pajari, A.* s.n. (C); 6-4-1997, *Gelert, O.* s.n. (C); Valle de los Nueve (27°59'26.95"N, 15°25'56.77"W)*, 150m, 15-3-1947, *Sventenius, E. R.* 7336 (ORT); Roque Nublo (27°58'5.96"N, 15°36'39.00"W)*, 30-3-1947, *Sventenius, E. R.* 7338 (ORT); Ayagaure (27°51'0.73"N, 15°36'30.43"W)*, 22-4-1947, *Sventenius, E. R.* 7335 (ORT); 22- 4-1947, *Sventenius, E. R.* 7334 (ORT); Entre Veneguera y S. Nicolás (27°55'28.20"N, 15°44'26.21"W)*, ?-2-1955, *Sventenius, E. R.* 24233 (ORT); Lomo Pedro Alfonso (27°50'43.21"N, 15°38'12.82"W)*, 400m, 7-1-1966, *Kunkel, G.* 3971 (LPA); Monte Doramas (28° 5'12.62"N, 15°34'59.20"W)*, 800m, 20-1-1967, *Kunkel, G.* 4875 (LPA); S. Nicolás village (27°58'34.85"N, 15°46'53.56"W)*, 11-3-1950, *Sventenius, E. R.* 7333 (ORT); Bco. Las Goteras (28° 1'51.29"N, 15°26'32.73"W)*, 430m, 30-1-1967, *Kunkel, G.* 4968 (LPA); 14-3-1973, *Sventenius, E.R* 13493 (LPA); San Cristobal (28° 4'43.69"N, 15°24'54.49"W)*, 50m, 16-1-1967, *Kunkel, G.* 4818 (LPA); Tafira Alta (28° 3'10.33"N, 15°27'58.65"W)*, 300m, 21-11-1965, *Kunkel, G.* 3742 (LPA); 5-10-1972, *Sventenius, E.R* 13495 (LPA); Tafira, Carr. A. Marzagan (28° 3'12.28"N, 15°25'53.20"W)*, 240m, 24-1-1965, *Kunkel, G.* 3553 (LPA); Barranco de la Angostura (27°52'50.28 "N,15°30'37.37"W)*, 600m, 5-1-1966, *Kunkel, G.* 3936 (LPA); Monte Tafira (28° 3'10.84"N, 15°27'58.04"W)*, 330m, 19-2-1967, *Kunkel, G.* 5124 (LPA); Bco. de Tenteniguada (27°59'11.18"N, 15°31'25.23"W)*, 20-1-1971, *Sventenius, E.R* 13508 (LPA); Bco. Arguineguin (27°45'43.18"N, 15°40'59.92"W)*, 8-3-1972, *Sventenius, E.R* 13500 (LPA); Los Tiles (28° 5'30.28"N, 15°34'58.59"W)*, 28-2-1972, *Sventenius, E.R* 13512 (LPA); Santa Lucia (27°54'36.50"N, 15°32'24.49"W)*, 21-11-1972, *Sventenius, E.R* 13503 (LPA); Roca Frente a Corralillos (27°53'26.68"N, 15°28'11.90"W)*, 8-3-1972, *Sventenius, E.R* 13506 (LPA); Teror, Barranco de Madrelagua, La Culata (27°58'33.12"N, 15°36'0.21"W)*, 10-2-1972, *Kunkel, G.* 15204 (LPA); 890m, 13-2-2011, *Marrero, Á.* 27149 (LPA); Tafira (28° 3'39.49"N, 15°27'35.14"W)*, 6-6-1972, *Sventenius, E.R* 13501 (LPA); Barranco de Guayadeque (27°54' N, 15°23' W), 19-12-1979, *Diez, J., Gallego, M.J., Silvestre, S.* s.n. (SEV); Maspalomas (27°45' N, 15°34' W), 12-3-1979, *Duvigneaud, J.* 21 Can (MA); Moya (28° 6'34.67"N, 15°34'58.15"W)*, ?-5-1980, *Rodríguez, C. S. Pérez, P. L.* 49782 (TFC); Cazadores: Telde (27°57'16.39"N, 15°30'45.49"W)*, 3-4-1982, *Rodrigo, J.* 016028 (LPA); Santa María de Guía (28° 6'26.81"N, 15°36'21.84"W)*, 200-50m, 17-3-1982, *Rodrigo, J.* 8757 (LPA); Barranco de Tenteniguada (27°59'7.20"N, 15°31'20.84"W)*, 1150m, 30-4-1986, *Montelongo, V.M. Roca, A.* 13543 (LPA); El Risco (28°03' N, 15°43' W), 12-1-1996, *Evrard, C.* 12139 (BR); Telde. Mountain of Las Palmas (27°59'40.56"N, 15°27'11.63"W)*, 1-2-2000, *Navarro, B.* 019903 (LPA); Gáldar. Los Covideros (28° 8'49.17"N, 15°39'16.88"W)*, 20-1-2000, *Navarro, B.* 019902/019905 (LPA); Barranco de Anzofé (28° 6'46.36"N, 15°38'26.00"W)*, 20-1-2000, *Navarro, B.* 019904 (LPA); Agaete. El Sao (28° 3'42.22"N, 15°39'30.93"W)*, 31-3-2000, *Roca, A.* 019901 (LPA); Ingenio/Aguimes, Barranco de Guayadeque (27°55'57.44"N, 15°28'2.02"W)*, 600-750m, 26-4-2000, *Roca, A.* 37896 (LPA); Las Lagunas (27°54'13.16"N, 15°32'22.98"W)*, 610-15m, 6-3-2010, *Marrero, Á.* 30750 (LPA); 610-15m, 6-3-2010, *Marrero, Á.* 30751 (LPA); Santa Lucía de Tirajana, La Sorrueda (27°54'37.39"N, 15°32'25.02"W)*, 400-

1 The coordinates marked with * were deduced from the gazetteer.

20m, 6-3-2010, *Marrero, Á.* 30797 (LPA); Reserva Integral de Inagua, Santa Juana (27°56'56.35"N, 15°41'4.14"W)*, 829m, 16-3-2010, *Pérez, M. M.* 029642 (LPA); Barranco de Madrelagua. La Culata (27°58'32.77"N, 15°36'1.20"W)*, 890m, 13-2-2011, *Marrero, Á.* 27147/27148 (LPA); Botanical Canarian Garden Viera y Clavijo (28° 3'49.95"N, 15°27'44.94"W)* 320 mm, 20-1-2010, *Marrero, Á.* 32421/32422 (LPA); 320m, 3-12-2020, *Marrero, Á.* 38628/38629 (LPA); 320m, 9-2-2021, *Marrero, Á.* 38997/38998 (LPA); (28°03'50" N, 15°27'45" W), 330m, 27-3-2022, *Silveira, P., Simão, I.* 3382.1/.2/.3/.4/.5 (AVE, ORT); Ingenio. El Roque (27°56'52" N, 15°28'49" W), 855m, 10-4-2022, *Reyes-Betancort, J. A. 3.3/.4/.5 In Silveira, P.* 3420.3/.4/.5 (AVE, ORT); Valsequillo. Edge of the boiler of "los Marteles" (27°57'31" N, 15°31'58" W), 1546m, 10-4-2022, *Reyes-Betancort, J. A. 4 In Silveira, P.* 3421 (AVE); Las Mesas Altas (28°06'08" N, 15°29'57" W), 9-4-2022, *Reyes-Betancort, J. A. 1.1/.2/.3 In Silveira, P.* 3418.1/.2/.3 (AVE, ORT); Barranco de la Angostura (27°52'34" N, 15°30'07" W), 295m, 10-4-2022, *Reyes-Betancort, J. A. 2.1/.2 In Silveira, P.* 3419.1/.2 (AVE, ORT); Ingenio. El Roque (27°56'52" N, 15°28'49" W), 855m, 10-4-2022, *Reyes-Betancort, J. A. 3.1/.2/.3/.4/.5 In Silveira, P.* 3420.1/.2/.3/.4/.5 (AVE, ORT); Valsequillo. Borde de la Caldera de los Marteles (27°57'31" N, 15°31'58" W), 1546m, 10-4-2022, *Reyes-Betancort, J. A. 4 In Silveira, P.* 3421 (AVE, ORT); Cruz de Tejeda (28°00'20" N, 15°36'00" W), 1509m, 10-4-2022, *Reyes-Betancort, J. A. 5.1/.2 In Silveira, P.* 3422.1/.2 (AVE, ORT); 850m, dry ground, 20-4-1936, *Brooke, W.M.A.* 150 (BM, BM 813634); Bco. de Tejeda (27°59'17.91"N, 15°37'6.28"W)*, 19-4-1972, *Sventenius, E.R* 13504 (LPA); Tejeda, Reserva Integral de Inagua (27°59'46.36"N, 15°36'54.71"W)*, 420m, 3-3-2010, *Soto, M. E.* 029615 (LPA); Barranco de Agaete (28°06'06" N, 15°42'17" W), 21m, 10-4-2022, *Reyes-Betancort, J. A. 7.1/.2/.3/.4/8 In Silveira, P.* 3424.1/.2/.3/.4/3425 (AVE, ORT); ascent to Teno Alto through the old hermitage of San Mateo in Ten (28°21'19" N, 16°54'04" W), 292m, 22-4-2022, *Reyes-Betancort, J. A. 9.1/.2/.3/.4/.5 In Silveira, P.* 3426.1/.2/.3/.4/.5 (AVE, ORT); Teno Alto. Bajada a las Cuevas (28°20'58" N, 16°53'39" W), 561m, 22-4-2022, *Reyes-Betancort, J. A. 10 In Silveira, P.* 3427 (AVE, ORT); Tabaibal de Mogán (27°53'2.77"N, 15°43'22.43"W)*, 21-2-1973, *Sventenius, E.R* 13502 (LPA); Mogán. (27°53' N, 15°43' W), 350m, 14-2-1993, *Vašák, V.* s.n. (BR); Cortadores, Charco del Burro (27°47'42.03"N, 15°41'37.99"W)*, 12-3-2011, *Marrero, Á.* 027257/027258/027259 (LPA); Mogán. Cortadores (27°51'41" N, 15°40'44" W), 700m, 12-3-2011, *Marrero, Á.* 027256/027260 (LPA); 726m, 30-4-2022, *Reyes-Betancort, J. A. 11.1/.2/.3/.4/.5 In Silveira, P.* 3428.1/.2/.3/.4/.5 (AVE, ORT).

Lanzarote: Masdache (29°00' N, 13°40' W), 15-7-1858, *Lowe, R.T.* 14 (BM); La Corona (29°11'5.02"N, 13°28'51.01"W)*, 450m, 15-4-1948, *Sventenius, E. R.* 22381 (ORT); Yaiza (28°57'21.37"N, 13°46'3.77"W)*, 200m, 5-5-1948, *Sventenius, E. R.* 22382 (ORT); 300m, 8-5-1948, *Sventenius, E. R.* 22383 (ORT); Alegranza (29°17'52.90"N, 13°32'0.87"W)*, 16-5-1953, *Sventenius, E. R.* 23418 (ORT); Graciosa, el Portillo (29°13'45.55"N, 13°30'17.36"W)*, 14-4-1957, *Sventenius, E. R.* 1188 (ORT); Montaña Clara, Caldera (29°17'52.90"N, 13°32'0.87"W)*, 16-4-1957, *Sventenius, E. R.* 23555 (ORT); 17-4-1957, *Sventenius, E. R.* 1375 (ORT); Graciosa (29°14'28.98"N, 13°30'58.16"W)*, 14-4-1957, *Sventenius, E. R.* 1187 (ORT); Graciosa, Central Part (29°15'14.78"N, 13°30'14.31"W)*, 14-4-1957, *Sventenius, E. R.* 1189 (ORT); Haría (29° 8'47.44"N, 13°29'53.97"W)*, 14-4-1968, *Sventenius, E. R.* 223?9 (ORT); Haría, mountain "La Higuera" (29° 8'48.57"N, 13°29'53.33"W)*, 14-4-1968, *Sventenius, E. R.* 22380 (ORT); Punta Pechiguera (28°51' N, 13°52' W), 24-12-1973, *Duvigneaud, J.* can1504 bis (BR); Teseguite (29°03' N, 13°32' W), 4-1-1974, Duvigneaud, J. can61 (BR); Riscos de Famara (29° 6'55.98"N, 13°33'22.90"W)*, 24-3-1984, *Montelongo, V.M.* 13887 (LPA); 24-3-1984, *Montelongo, V.M.* 14139 (LPA); Arrecife (28°57'54.59"N, 13°33'18.23"W)*, 4-4-1993, *Reyes-Betancort, J. A.* 37408 (TFC); Punta Mujeres (29°08'19" N, 13°27'18" W), in a ditch by the side of the road, 29-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.c/d/e (AVE); Orzola (29°13'21.67"N, 13°27'12.05"W)*, 17-2-1964, *Von Stranes, Dr. R.* 22384 (ORT); (29°12'32" N, 13°26'02" W), in the sands of the roadsides and paths, 29-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.f (AVE); surroundings of Playa Blanca (28°52'24" N, 13°49'54" W), 42m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3384.1 (AVE, ORT); (28°52'42" N, 13°49'57" W), 49m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3385.1/.3/.4/.5 (AVE, ORT); (28°52'41" N, 13°49'58" W), 49m, 29-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3386 (AVE, ORT); Teguise (29°03'50" N, 13°32'59" W), 321m, 30-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3389.2 (AVE, ORT); Manguía (29°04'14" N, 13°31'31" W), 273m, 30-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3390.1/.2 (AVE); Guinate viewpoint (29°18'46" N, 13°50'07" W), 354m, 30-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3392.2 (ORT); El Jable (29°03'55" N, 13°35'05" W), 145m, 30-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3393.1/.2/3394/3395 .1/.2/.3 (AVE, ORT).

Tenerife: Puerto de la Cruz (28°19' N, 16°34' W), 1845, *Bourgeau, E.* 839 (BM, K); 20-1-1921, Rahn, K. 463 (C); 21-2-1921, Rahn, K. 517 (C); 24-2-1921, Rahn, K. 555 (C); 25-10-1965, Kaae, N. 1 (C); 25-3-1967, Kaae, N. s.n. (C); 20-10-1969, Kaae, N. 2 (C); 120m, 26-2-1969, Bramwell, D. 807 (C); 600m, 27-12-1973, Lewalle, J. 7343 (RAB); 390m, 24-3-1975, Dalgaard, L. Dalgaard, V. 6011 (C); 2-4-1972, Hansen, A. s.n. (C); 1-11-1974, Hansen, A. s.n. (C);

14-3-1979, Olgaards, P. s.n. (C); Las Palmas (28°20’ N, 16°50’ W), 100m, dry semmy ground, 25-2-1936, *Brooke, W.M.A.* 1 (BM); Realejo Alto (28°23’7.70”N, 16°35’3.10”W)*, 28-1-1944, *Sventenius, E. R.* 13057 (ORT); Pto. Cruz (28°24’41.08”N, 16°32’41.89”W)*, 23-1-1944, Sventenius, E. R. 13107 (ORT); La Perdoma (28°22’38.43”N, 16°33’0.74”W)*, 6-2-1944, *Sventenius, E. R.* 13110 (ORT); Realejo Alto (28°23’7.70”N, 16°35’3.10”W), 28-1-1944, *Sventenius, E. R.* 13057 (ORT); Roque del Fraile (28° 0’46.91”N, 16°40’7.21”W)*, 23-5-1944, *Sventenius, E. R.* 13105 (ORT); El Chorrillo (27°59’ N, 15°40’ W), 22-3-1959, Bellot, F. Casaseca, B. s.n. (MACB); Los Silos (28°21’58.69”N, 16°49’4.35”W)*, 17-1-1946, *Sventenius, E. R.* 13109 (ORT); 26-4-1996, *Sauquillo, V. L. Arco, M. J.* 40554 (TFC); Valverde (27°47’35.42”N, 17°56’29.84”W)*, 400m, 7-5-1949, *Sventenius, E. R.* 18164 (ORT); Punta de Teno (28°20’31.51”N, 16°55’22.60”W)*, 4-3-1950, *Sventenius, E. R.* 13106 (ORT); 6-3-1950, *Sventenius, E. R.* 1694 (ORT); 25-3-1969, *Sventenius, E. R.* 13059 (ORT); Tacoronte (28°28’ N, 16°24’ W), 12-8-1954, *Morales, A.* s.n. (MACB); waste ground at roadside, 1-4-1975, *Cannon, J.F.M. Cannon, M.J., Cannon, P.F.* 4410 (BM); Carretera entre La Esperanza e Izaña (28°26’42.02”N, 16°20’40.95”W)*, 1700 m, 19-2-1961, *Sventenius, E. R.* 13058 (ORT); Adeje, Bco. del Infierno (28° 7’34.68”N, 16°43’25.32”W)*, 23-3-1962, *Sventenius, E. R.* 13103 (ORT); Anaga, Bco. Ijuana (28°31’59.47”N, 16°15’4.06”W)*, 21-2-1963, *Sventenius, E. R.* 13108 (ORT); Anaga (28°29’39.01”N, 16°16’32.71”W)*, 21-2-1967, *Sventenius, E. R.* 13056 (ORT); Bco. S. Andrés (28°31’22.26”N, 16°12’20.93”W)*, 450 m, 17-3-1968, *Sventenius, E. R.* 30212 (ORT); Bajamar (28°33’14.98”N, 16°20’40.35”W)*, 26-2-1969, *Bramwell, D.* 13511 (LPA); Montañas de Teno (28°20’32.24”N, 16°52’36.22”W)*, 100m, 19-2-1969, *Bramwell, D.* 13494 (LPA); San José above La Rambla (28°23’36.10”N, 16°38’57.92”W)*, 200m, 14-1-1969, *Bramwell, D.* 13507/13510 (LPA); Teno (28°20’ N, 16°51’ W), 100m, at side of road at Teno Bajo, 19-2-1969, *Bramwell, D.* 752 (SEV, C); San Jose (28°38’ N, 17°45’ W), fields and roadsides, 14-1-1969, *Bramwell, D.* 529 (SEV); Los Cristianos (28°03’ N, 16°43’ W), 22-12-1970, Duvigneaud, J. can924 (BR); Masca (28°18’19.27”N, 16°50’23.69”W)*, 19-4-1971, *Sventenius, E. R.* 13104 (ORT); Adeje (28°07’ N, 16°43’ W), 9-1-1972, Kaae, N. 2 (C); Arico (28°10’ N, 16°29’ W), 660m, 27-1-1973, Aldridge, A.E. 519 (BM); Bajamar (28°32’ N, 16°20’ W), 100m, 25-1-1973, Aldridge, A.E. 440 (BM); Punta del Hidalgo (28°35’ N, 16°19’ W), 200m, in rocks above the water canal, 24-1-1973, Aldridge, A.E. 414 (BM);Guimar (28°18’ N, 16°25’ W), 8-9-1975, Cabezudo, B., Talavera, S. s.n. (SEV); San Andres (27°46’ N, 17°57’ W), 200m, rough grassland, 4-4-1977, Jarvis Murphy 68 (BM, RNG); Arona (28°05’ N, 16°40’ W), uncultivated land, ruderalized, anthropized, 10-3-1979, Pérez de Paz, P.L. 20790 (MA); Arona (28° 6’0.30”N, 16°40’47.22”W)*, 10-3-1979, Pérez de Paz, P. L., Arco Aguilar, M. J.20790 (TFC); 10-3-1979, Pérez de Paz, P. L., Arco Aguilar, M. 20790dup. (TFC); La Laguna (28°29’ N, 16°19’ W), ?-3-1979, Wildpret, W., del Arco, M. s.n. (MAF, TFC, SALA); Santa María del Mar (28°25’43.97”N, 16°18’25.53”W)*, 1-3-1979, *Wildpret, W., M.*. 24399 (TFC); El Escobonal (28°15’29.79”N, 16°25’35.31”W)*, 6-3-1981, Rodríguez, O. Cabrera, P. G. 12669 (TFC); La Laguna (28°29’14.64”N, 16°18’57.26”W)*, 20-11-1981, *García Gallo, A.* 22049 (TFC); Pto. de la Cruz (28°24’41.09”N, 16°32’41.86”W)*, 11-4-1981, *González, C.* 29501 (ORT); Las Cañadas (28°13’55.37”N, 16°34’54.77”W)*, 13-4-1984, *Santos, A.* 28661 (ORT); Lomo Marrera (28°17’20.85”N, 16°24’3.38”W)*, 25-3-1985, Rodríguez Delgado, O. 27684 (TFC); Las Magarzas, Montaña Birmagen (28°26’21.70”N, 16°21’17.59”W)*, 700-800m, 1-3-1989, *Montelongo, V.M.* 017615 (LPA); Buenavista (28°22’ N, 16°54’ W), 1-4-1990, Bouharmont, J. 21117 (BR); Tegueste (28°31’22.22”N, 16°20’16.56”W)*, 13-2-1994, Felipe, T. J. Felipe, T. J. 36604 (TFC); El Sauzal (28°28’29.92”N, 16°26’2.94”W)*, 30-1-1994, Rodríguez, F. R. 37624 (TFC); Erjos (28°19’44.10”N, 16°48’16.12”W)*, 30-5-1996, Sauquillo, V. L. Arco Aguilar, M. J. 39996 (TFC); Montaña Aregume (28°22’9.86”N, 16°49’3.42”W)*, 23-2-1996, Saquillo, V. C. Acosta, C. 40100 (TFC); Tacoronte (28°28’47.36”N, 16°24’47.31”W)*, 26-12-1993, *Rodríguez, F. R.* 37639 (TFC); 28-2-2007, *Navarro, M. L. R.* 48770 (TFC);6-11-2008, *Navarro, M. L.* 48837 (TFC); buena vista del norte (28°22’ N, 16°54’ W), 300m, 2-5-1997, Panero, J.L., Ortega JF 6976 (MA); Punta del Hidalgo (28°35’ N, 16°19’ W), 200m, 12-2-2003, Bouharmont, J. 32103 (BR); Jb La Concepción (28°27’52.54”N, 16°14’56.73”W)*, 8-4-2010, *Sánchez, P. S.* 028215 (LPA); San Tiago del Teide (28°17’ N, 16°48’ W)*, 900m, *Dürbye/Kirchhoff 1116 in BGBD 718 in Silveira, P.* 3082 (AVE); (28°30’47” N, 16°24’59” W), 136m, 31-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3396.1/.2 (AVE, ORT); Universidad La Laguna (28°28’54”N, 16°19’01”W)*, 13-4-2021, *Montelongo, C. G.* 50501 (TFC); C. del Calvario between Tacoronte and Lomo Colorado (28°29’00” N, 16°24’22” W), 479m, 31-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3397.1/.2/.3/.4/.5/.6/.7 (AVE, ORT); La Laguna (28°30’04” N, 16°18’46” W), 532m, 31-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3398.1/.2 (AVE, ORT); Las Canteras (28°30’24” N, 16°10’41” W), 540m, 31-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3399 (AVE, ORT); Bejía (28°32’53” N, 16°18’04” W), 575m, 31-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3400/3401/3402 (AVE, ORT); carr. Tahodio (28°29’20” N, 16°14’56” W), 39m, 31-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3403/3404 (AVE, ORT); street near Buenavista (28°23’00” N, 16°50’18” W), 47m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3405 (AVE, ORT); Buenavista del Nte. (28°22’21” N, 16°50’58” W), 99m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3406/3407/3408 (AVE,

ORT); TF-375 near Las Manchas (28°16'55" N, 16°47'51" W), 1054m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3410/3411.1/.2 (AVE, ORT); TF-82 near Barranco de Ramallo (28°12'14" N, 16°46'10" W), 626m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3412.1/.2/3413.1/.2 (AVE, ORT); Tejina de Isora (28°11'01" N, 16°45'55" W), 553m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3414 (AVE, ORT); fields along Av. el Palm-Mar (28°01'35" N, 16°41'18" W), 81m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3415.1 (AVE, ORT); 81m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3416.1/.2/3417 (AVE, ORT); ascent to Teno Alto through the old hermitage of S (28°21'19" N, 16°54'04" W), 292m, 22-4-2022, *Reyes-Betancort, J. A. 9.1 In Silveira, P.* 3426.1 (AVE, ORT); (28°21'21" N, 16°54'05" W), 247m, 22-4-2022, *Reyes-Betancort, J. A. 9.3/.4 In Silveira, P.* 3426.3/.4 (AVE, ORT); (28°21'22" N, 16°54'07" W), 207m, 22-4-2022, *Reyes-Betancort, J. A. 9.5 In Silveira, P.* 3426.5 (AVE, ORT); Teno Alto. Descent to las Cuevas (28°20'58" N, 16°53'39" W), 561m, 22- 4-2022, *Reyes-Betancort, J. A. 10 In Silveira, P.* 3427 (AVE, ORT).

La Palma: Between Fuencaliente and chapel of Sta. Cecilia (28°30'25.81"N, 17°50'47.47"W)*, 25-4-1963, *Sventenius, E. R.* 3444 (ORT); Barlovento (28°49'31.99"N, 17°49'20.67"W)*, 12-8-1964, *Sventenius, E. R.* 3443 (ORT); El Paso (28°39' N, 17°52' W), 1300m, 27-3-1980, Machado, A. s.n. (MA).

La Gomera: Bco. Iguala (28° 5'7.39"N, 17°18'6.59"W)*, 22-3-1959, *Sventenius, E. R.* (ORT); Valle Gran Rey (28°06' N, 17°19' W), 600m, roadside, 7-3-1973, *Aldridge, A.E.* 1062 (BM); 13-4-1981, *Pérez, B. M. Acebes, J. R.* 13623 (TFC); El Molinito (28°06' N, 17°08' W), 7-3-1990, *Hansen, A.* 80 (C); 12-5-1992, *Forfung, A.S.* s.n. (C); Laderas sobre La Dama (28° 3'15.89"N, 17°18'2.36"W)*, 30-1-2002, *Pérez de Paz, P. L.. Arco, M. J.* 46434 (TFC).

El Hierro: Sabinosa (27°45' N, 18°06' W), 50m, cliffs, ?-8-1971, *Bramwell, D. Humphries, C.J.* 3316 (BM); La Frontera (27°45'15.68"N, 18° 0'33.30"W)*, 350m, 5-5-1959, *Sventenius, E. R.* 18162 (ORT); El Pozo de la Salud (27°45'13.80"N, 18° 6'20.24"W)*, 8-4-1971, *Sventenius, E. R.* 18163 (ORT); Lomo Negro (27°45'14.98"N, 18° 8'39.38"W)*, 10-4-1979, *Wildpret, W., Arco, M.J., Pérez de Paz, P. L., Hdez. Padrón, C.* 8086 (TFC); Tenecedra (27°49'24.03"N, 17°56'8.80"W)*, 13-7-1981, *Pérez de Paz, P. L., Hdez. Padrón, C.* 34380 (TFC); Jarales (27°48' N, 17°58' W), 23-2-1991, *Hansen, A.* 45 (C); Frontera (27°45'15.58"N, 18° 0'33.46"W)*, 23-3-1998, *Pérez de Paz P. L., Cruz Trujillo, G.* 42972 (TFC).

***Calendula officinalis* L.**

SPAIN. **Canary Islands**

Fuerteventura: (28°20' N, 14°00' W), 10m, 25-2-2011, *Peterson, T.* s.n. (S); 27-2-2011, *Peterson, T.* s.n. (S); 28-2-2011, *Peterson, T.* s.n. (S); 1-3-2011, *Peterson, T.* s.n. (S).

Gran Canaria: Valleseco. Sobre Mesas de Galaz (28°00'54" N, 15°35'25" W), 1587m, 10-4-2022, *Reyes-Betancort, J. A. 6.1/.2 In Silveira, P.* 3423.1/.2 (AVE, ORT).

Lanzarote: Femés (28°54'45" N, 13°46'47" W), 371m, 29-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3388 (AVE, ORT).

Tenerife: Near Puerto de Erjos (28°19'41" N, 16°48'12" W), 1007m, 1-4-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3409 (AVE, ORT).

La Gomera: (28°06' N, 17°08' W), ?-5-47, *Marcos* 1519 (BCN).

***Calendula ricardoi* I.Simão, Reyes-Bet. & P.Silveira**

SPAIN. **Canary Islands**

Fuerteventura: Betancuria (28°25' N, 14°03' W), 15-3-1974, *Leskinen, M.* s.n. (C); (28°26'24" N, 14°03'15" W), 29-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.g (AVE); (28°25'42" N, 14°03'29" W), 446m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.i.1/.4 (AVE); FC-30 towards Pájara, near Sra. de La Peña (28°23'46" N, 14°04'02" W), 308m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.j (AVE); Pájara (28°21'25" N, 14°04'51" W), 262m, 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.l.1/.2 (AVE).

***Calendula sventenii* I.Simão, Reyes-Bet. & P.Silveira**

SPAIN. **Canary Islands**:

Fuerteventura: Lobos (28°44'56.15"N,13°49'13.16"W)*, 5-4-1955, *Sventenius, E. R.* 1276 (ORT); 20-3-1988, *Hansen, A.* 452 (C); Lobos (28°44'56.15"N,13°49'13.16"W), 5-4-1955, *Sventenius, E. R.* 1276 (ORT); Pájara, La Lajita (28°21'3.35"N,14° 6'29.11"W)*, 0-50m, 30-3-1989, *Montelongo, V.M.* 17586 (LPA); 125m, 16-12-2018, *Scholz, S.* 36470/36471 (LPA); Jandía, Playa Esmeralda (28° 8'40.85"N, 14°14'8.42"W)*, 1-5-1981, *Méndez Pérez, B., Acebes Genovés, J. R.* 13547/13652 (TFC); Jable de Lajares (28°40'22.49"N,13°57'17.76"W)*, 18-2-1984, *Montelongo, V.M.*

13825 (LPA); Morro del Recogedero (28°10’17.62”N,14°13’16.43”W)*, 350m, 24-2-1986, *Navarro, B. Montelongo, V.M.* 13886 (LPA); Cofete (28°05’ N, 14°23’ W), 14-3-1989, *Hansen, A.* 828 (C); Peninsula de Jandía (28°03’ N, 14°30’ W), 12-3-1989, *Hansen, A.* 731 (C); (28° 4’33.45”N,14°29’38.74”W)*, 9-2-2005, *Scholz, S.* 46116 (TFC); 7-3-2007, *Scholz, S.* 47950 (TFC); Playa de Gran Valle (28°15’16.02”N, 13°55’7.42”W)*, 12-4-2005, *Cruz Trujillo, G.. Pérez de Paz, P. L. , González, R.* 48535 (TFC); Jorós (28° 4’14.07”N, 14°23’59.32”W)*, 14-1-2005, *Pérez de Paz, P. L., González, R.* 48441 (TFC); Tuineje, Playa de Agando (28°12’12.28”N,14° 3’0.66”W)*, 10m, 9-12-2018, *Scholz, S.* 36451 (LPA); along FV-102 towards La Caldereta (28°35’05” N, 13°51’49” W), 133m, 26-3-2022, *Silveira, P. Simão, I.* 3372 (AVE); Corralejo (28°44’ N, 13°52’ W), 3-4-1985, *Hansen, A.* 76 (C); 23-3-1986, *Hansen, A.* 212 (C); 28-3-2014, *Silveira, P., Gonçalves, A.C.R.S.* 3327.a/.b (AVE); Gran Tarajal (28°12’53.56”N,14° 1’13.24”W)*, 24-2-1986, *Marrero, Á.* 13885 (LPA); (28°15’23” N, 14°01’04” W), 91m, 26-3-2022, *Silveira, P., Simão, I.* 3380, 3381 (AVE, ORT).

Gran Canaria: Isleta (28° 9’59.43”N,15°25’14.74”W)*, 40m, 22-1-1966, *Marrero, Á.* 4062 (LPA); (28°00’ N, 15°36’ W), 75m, 26-3-1976, *Dalgaard, L. Dalgaard, V.* 7026 (C); Andén Verde (28° 1’9.97”N, 15°47’7.61”W)*, 28-3-1988, *Wildpret, W. Rodríguez Delgado, O., Arco Aguilar, M.* 36896 (TFC); Gáldar, Amagro (28° 7’53.69”N,15°40’52.08”W)*, 18-1-2000, *Olangua, M., Suárez, C. & Navarro, J.* 019900 (LPA); Mogán, Rampa de Tabaibales (27°50’46.80”N,15°46’48.43”W)*, 200-250m, 14-2-2006, *Marrero, Á.* 30300/30301 (LPA); Montaña Amagro (28° 6’40.03”N,15°30’14.32”W)*, 22-2-2014, *Garcia, R.* 38024 (LPA); 1-5-2016, *Garcia, R.* 38232 (LPA); Mogán (27°53’2.20”N,15°43’23.75”W)*, 450m, 12-3-2011, *Marrero, Á.* 30429 (LPA); Bco. de La Palma (28° 4’6.93”N,15°42’36.03”W)*, 14-2-1973, *Sventenius, E.R* 13498 (LPA).

Lanzarote: Graciosa (29°15’8.75”N,13°30’22.12”W)*, 14-4-1957, *Sventenius, E. R.* 23802 (ORT); (29°00’ N, 13°40’ W), 9-3-1978, *Hansen, A.* 172 (C); Montaña Blanca (28°59’13.53”N,13°38’13.18”W)*, 12-1-1992, *Pérez de Paz, P. L., Gómez, M. M., Alemó, E. C.* 35310 (TFC); Teguise (29° 1’37.87”N,13°38’7.87”W)*, 16-3-1997, *Reyes-Betancort, J. A.* 40443 (TFC); Punta Pechiguera (28°51’20.08”N, 13°52’21.00”W)*, 13-2-2010, *Reyes-Betancort, J. A., Glez Glez, R.* 41487 (ORT); (28°51’22” N, 13°52’18” W), 18m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3383.1/.2/.3/.4/.5/.6 (AVE, ORT); surroundings of Playa Blanca (28°52’24” N, 13°49’54” W), 42m, 29-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3384.2 (AVE); (28°52’42” N, 13°49’57” W), 49m, 29-3-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3385.2 (AVE, ORT); (28°52’41” N, 13°49’58” W), 49m, 29-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3387.1/.2/.3 (AVE, ORT); mirador de Guinate (29°11’06” N, 13°30’04” W), 354m, 30-3-2022, *Silveira, P. Simão, I. & Reyes-Betancort, J. A.* 3391.1/.2 (AVE, ORT).

Tenerife: Santa María del Mar (28°25’44.44” N, 16°18’23.26”W)*, 1-3-1979, *Wildpret, W. Arco, M.* 24399 (TFC); Adeje (28° 7’21.55”N,16°43’58.80”W)*, 20-2-1984, *Beltrán E., León, M. C., González, L.* 7613 (TFC); fields along Av. el Palm-Mar (28°01’35” N, 16°41’18” W), 81m, 1-4-2022, *Silveira, P., Simão, I. & Reyes-Betancort, J. A.* 3415.2/.3/.4/.5 (AVE, ORT).