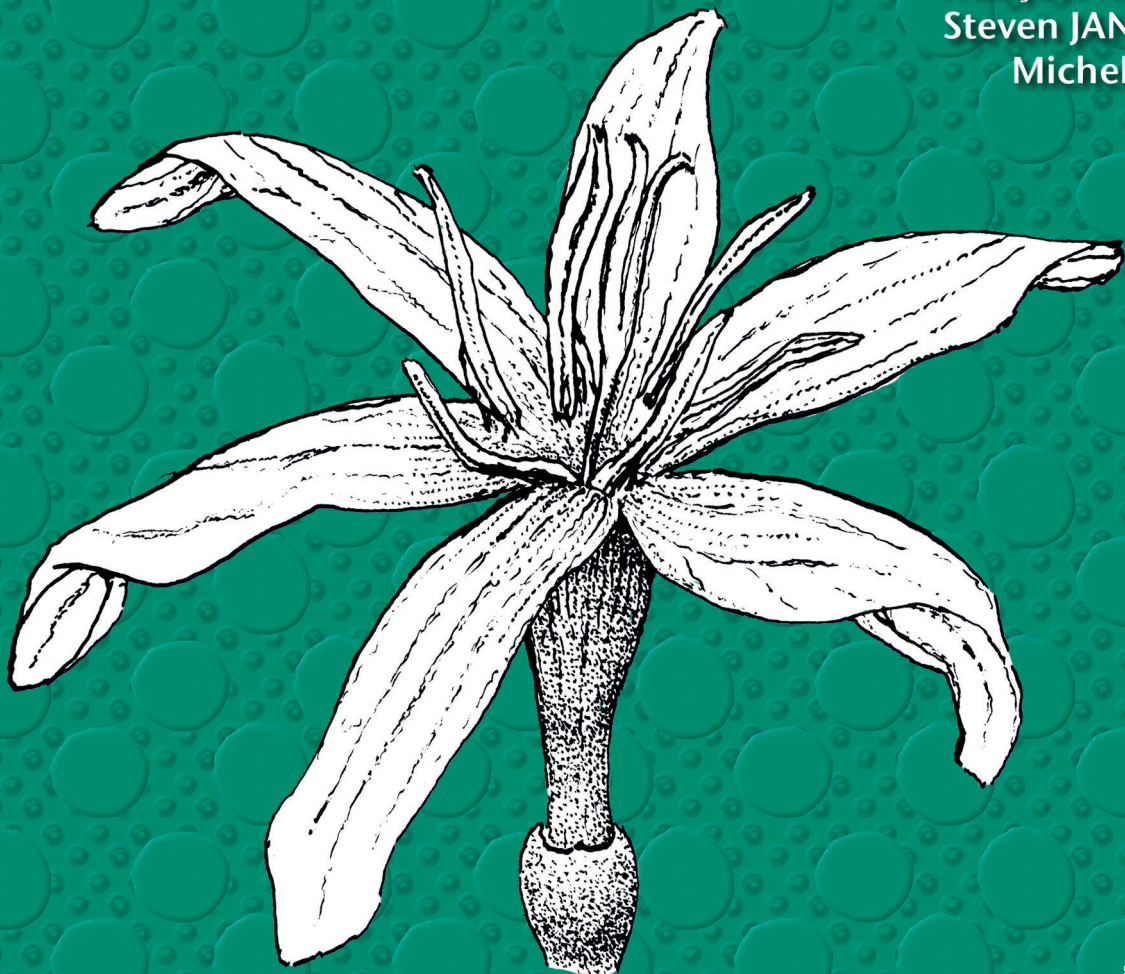


# adansonia

2021 • 43 • 26

A new coffee species from South-West Cameroon,  
the principal hotspot of diversity for *Coffea* L.  
(Coffeae, Ixoroideae, Rubiaceae) in Africa

Piet STOFFELEN,  
François ANTHONY,  
Steven JANSSENS &  
Michel NOIROT



DIRECTEUR DE LA PUBLICATION / PUBLICATION DIRECTOR: Bruno David  
Président du Muséum national d'Histoire naturelle

RÉDACTEUR EN CHEF / EDITOR-IN-CHIEF: Thierry Deroin

RÉDACTEURS / EDITORS: Porter P. Lowry II; Zachary S. Rogers

ASSISTANT DE RÉDACTION / ASSISTANT EDITOR: Emmanuel Côté (adanson@mnhn.fr)

MISE EN PAGE / PAGE LAYOUT: Emmanuel Côté

COMITÉ SCIENTIFIQUE / SCIENTIFIC BOARD:

P. Baas (Nationaal Herbarium Nederland, Wageningen)  
F. Blasco (CNRS, Toulouse)  
M. W. Callmänder (Conservatoire et Jardin botaniques de la Ville de Genève)  
J. A. Doyle (University of California, Davis)  
P. K. Endress (Institute of Systematic Botany, Zürich)  
P. Feldmann (Cirad, Montpellier)  
L. Gautier (Conservatoire et Jardins botaniques de la Ville de Genève)  
F. Ghahremaninejad (Kharazmi University, Téhéran)  
K. Iwatsuki (Museum of Nature and Human Activities, Hyogo)  
A. A. Khapugin (Tyumen State University, Russia)  
K. Kubitzki (Institut für Allgemeine Botanik, Hamburg)  
J.-Y. Lesouef (Conservatoire botanique de Brest)  
P. Morat (Muséum national d'Histoire naturelle, Paris)  
J. Munzinger (Institut de Recherche pour le Développement, Montpellier)  
S. E. Rakotoarisoa (Millenium Seed Bank, Royal Botanic Gardens Kew, Madagascar Conservation Centre, Antananarivo)  
É. A. Rakotobe (Centre d'Applications des Recherches pharmaceutiques, Antananarivo)  
P. H. Raven (Missouri Botanical Garden, St. Louis)  
G. Tohmé (Conseil national de la Recherche scientifique Liban, Beyrouth)  
J. G. West (Australian National Herbarium, Canberra)  
J. R. Wood (Oxford)

COUVERTURE / COVER:

Réalisée à partir des Figures de l'article/*Made from the Figures of the article.*

*Adansonia* est indexé dans / *Adansonia is indexed in:*

- Science Citation Index Expanded (SciSearch®)
- ISI Alerting Services®
- Current Contents® / Agriculture, Biology, and Environmental Sciences®
- Scopus®

*Adansonia* est distribué en version électronique par / *Adansonia is distributed electronically by:*

- BioOne® (<http://www.bioone.org>)

*Adansonia* est une revue en flux continu publiée par les Publications scientifiques du Muséum, Paris  
*Adansonia is a fast track journal published by the Museum Science Press, Paris*

Les Publications scientifiques du Muséum publient aussi / *The Museum Science Press also publish: Geodiversitas, Zoosystema, Anthropozoologica, European Journal of Taxonomy, Naturae, Cryptogamie* sous-sections *Algologie, Bryologie, Mycologie, Comptes Rendus Palevol*

Diffusion – Publications scientifiques Muséum national d'Histoire naturelle  
CP 41 – 57 rue Cuvier F-75231 Paris cedex 05 (France)  
Tél.: 33 (0)1 40 79 48 05 / Fax: 33 (0)1 40 79 38 40  
[diff.pub@mnhn.fr](mailto:diff.pub@mnhn.fr) / <http://sciencepress.mnhn.fr>

© Publications scientifiques du Muséum national d'Histoire naturelle, Paris, 2021  
ISSN (imprimé / *print*): 1280-8571/ ISSN (électronique / *electronic*): 1639-4798

# A new coffee species from South-West Cameroon, the principal hotspot of diversity for *Coffea* L. (Coffeae, Ixoroideae, Rubiaceae) in Africa

**Piet STOFFELEN**

Meise Botanic Garden  
Nieuwelaan 38, 1860 Meise (Belgium)  
piet.stoffelen@plantentuinmeise.be

**François ANTHONY**

Unité Mixte de Recherche IPME, IRD  
911 avenue d'Agropolis, F-34394 Montpellier cedex 5 (France)  
francois.anthony@ird.fr (corresponding author)

**Steven JANSSENS**

Meise Botanic Garden  
Nieuwelaan 38, 1860 Meise (Belgium)  
steven.janssens@plantentuinmeise.be

**Michel NOIROT**

Unité Mixte de Recherche IPME, IRD  
911 avenue d'Agropolis, F-34394 Montpellier cedex 5 (France)  
michel.noirot@ird.fr

Submitted on 16 November 2020 | accepted on 9 May 2021 | published on 21 December 2021

Stoffelen P., Anthony F., Janssens S. & Noirot M. 2021. — A new coffee species from South-West Cameroon, the principal hotspot of diversity for *Coffea* L. (Coffeae, Ixoroideae, Rubiaceae) in Africa. *Adansonia*, sér. 3, 43 (26): 277-285. <https://doi.org/10.5252/adansonia2021v43a26>. <http://adansonia.com/43/26>

## ABSTRACT

*Coffea rizetiana* Stoff. & M.Noirot, sp. nov., a diploid species only known from a single wild population in South-West Cameroon, is formally described here. It is probably extinct in the wild, but is present in the germplasm collection in Bassin Martin (Réunion, France) and Meise Botanic Garden (Belgium). Phylogenetic analyses reveal a close relationship with *C. montekupensis* Stoff., a species endemic to the same region but occurring at higher elevations, and with *C. liberica* Bull. ex Hiern, a species with a wide Central and West African distribution. The new species can be distinguished from other *Coffea* species from Central Africa by its large black fleshy fruits with thick mesocarp. It can be differentiated from its close relative *C. montekupensis* by its larger less obovate leaves, longer corolla tube and larger fruits; and from *C. liberica* by its shorter petiole, acuminate leaf tip, cuneate leaf base, thinner leaf blade, shorter corolla tube, longer corolla lobes and black fruits with a thick fleshy pericarp. The evolutionary importance of the fruit colour is discussed, as well as the role of Mount Kupe, the Bakossi Mountains and the broader South-West Cameroon region as the principal *Coffea* diversity hotspot on the African continent.

## KEY WORDS

Rubiaceae,  
*Coffea*,  
Cameroon,  
Bakossi Mountains,  
Mount Kupe,  
new species.

## RÉSUMÉ

*Une nouvelle espèce de caféier du sud-ouest du Cameroun, le principal point chaud de diversité de Coffea L. (Coffeae, Ixoroideae, Rubiaceae) en Afrique.*

*Coffea rizetiana* Stoff. & M.Noiro, sp. nov., une espèce diploïde connue uniquement d'une seule population sauvage dans le sud-ouest du Cameroun, est formellement décrite. La nouvelle espèce est probablement éteinte à l'état sauvage et est présente dans la collection de matériel génétique à Bassin Martin (La Réunion, France) et au Jardin botanique de Meise (Belgique). Des analyses phylogénétiques révèlent d'étroites relations avec *C. montekupensis* Stoff., une espèce qui est endémique de la même région mais observée à des altitudes plus élevées que l'espèce nouvellement décrite, et avec *C. liberica* Bull. ex Hiern, une espèce avec une vaste distribution en Afrique centrale et occidentale et aussi présente dans cette région. La nouvelle espèce peut être distinguée de n'importe quelle autre espèce d'Afrique centrale par ses gros fruits pulpeux de couleur noire avec un mésocarpe épais. Elle se différencie de l'espèce proche *C. montekupensis* par ses feuilles moins grandes obovales, le tube de la corolle plus long et des fruits plus gros, et de *C. liberica* par son pétiole plus court, sa feuille à apex acuminé et base cunéiforme et le limbe plus fin, le tube de la corolle plus court et des lobes plus longs, et des fruits noirs avec un péricarpe pulpeux plus épais. L'importance évolutive de la couleur des fruits est discutée et le rôle du Mont Kupe et des Montagnes de Bakossi ainsi que du sud-ouest du Cameroun dans un sens plus large est mis en évidence, car c'est le principal point chaud de diversité des *Coffea* sur le continent africain.

## MOTS CLÉS

Rubiaceae  
*Coffea*,  
Cameroun,  
Montagnes de Bakossi,  
Mont Kupe,  
espèce nouvelle.

## INTRODUCTION

During the 1980's, the genetic resources of the genus *Coffea* L. conserved in Côte d'Ivoire were enhanced by IRD (ex-ORSTOM) during extensive prospecting of the rainforests of Cameroon (Anthony *et al.* 1985) and Congo-Brazzaville (Namur *et al.* 1987). Several of these new accessions were considered to be new species (Anthony 1992), however they were never formally described. Stoffelen (1998) made a taxonomic revision of the Central and West African *Coffea* species where several of these informal taxa were defined as unplaced. It was not possible to describe these informal taxa due to the lack of specimens with fruits or flowers.

The availability of additional fruiting and flowering specimens led to the description of *C. charrieriana* Stoff. & F.Anthony from South-West Cameroon (Stoffelen *et al.* 2008) and *C. anthonyi* Stoff. & F.Anthony (Stoffelen *et al.* 2009). In 2013, fruits and flowers were collected from another potentially new species, *Coffea* sp. 'Koto', from the *Coffea* germplasm collection in Bassin Martin (Réunion, France), described below.

## MATERIAL AND METHODS

### TAXON DELIMITATION AND BOTANICAL DESCRIPTION

Taxon delimitation and botanical description are based on preserved collections: herbarium specimens and liquid preserved fruit and flowers. Herbarium vouchers and liquid preserved spirit collections were made from material cultivated in Meise Botanic Garden (Belgium) and the Bassin Martin coffee collection (Réunion, France). Specimens were deposited at BR and duplicates were sent to P, K and WAG following herbarium abbreviations (Holmgren *et al.* 1981).

The specimens were compared against herbarium specimens of other *Coffea* species native to Central and West Africa housed at BR, K, WAG and P. These herbaria were also screened for the new species, but no additional specimens were found. Observations and description were according to the method described for tropical woody Rubiaceae by Robbrecht (1988). The morphological characters that are summarized in the results were based on the observation of three herbarium specimens.

### PHYLOGENETIC ANALYSES

The taxon sampling for this study was based on previous molecular phylogenetic studies of *Coffea* (Lashermes *et al.* 1997; Cros *et al.* 1998; Maurin *et al.* 2007; Anthony *et al.* 2010; Hamon *et al.* 2017), with particular focus on the phylogenetic position of *C. rizetiana* Stoff. & M.Noiro, sp. nov. within the genus. Based on the overall morphology of the species and the composition of the different clades within the genus, a reduced dataset including two accessions of the new *Coffea* species, *C. brevipes* Hiern (1 accession), *C. canephora* Pierre ex A.Froehner (2), *C. heterocalyx* Stoff. (2), *C. humilis* A.Chev. (2), *C. kapakata* (A.Chev.) Bridson (2), *C. liberica* Bull. ex Hiern var. *dewevrei* (De Wild. & T.Durand) Lebrun (1), *C. liberica* Bull. ex Hiern var. *liberica* (1) *C. magnistipula* Stoff. & Robbr. (1) and *C. montekupensis* Stoff. (1), was used to determine the relationship of the new species to closely related species. Two specimens of *C. rhamnifolia* (Chiov.) Bridson were used as outgroup. Specimen vouchers and GenBank accession numbers for the phylogenetic study are summarized in Table 1.

Total genomic DNA was isolated from silica-dried leaf material using a modified CTAB protocol, which was optimized for *Impatiens* (Janssens *et al.* 2006, 2008). Nuclear ribosomal intergenic transcribed spacer (ITS) and the

TABLE 1. — List of material studied in the phylogenetic analyses: vouchers and references in GenBank.

Species	Voucher	trnLF	accD	ITS
<i>Coffea brevipes</i> Hiern	GenBank	DQ153827	DQ153460	DQ153591
<i>Coffea canephora</i> Pierre ex A.Froehner (1)	GenBank	DQ153829	DQ153462	DQ153593
<i>Coffea canephora</i> (2)	GenBank	—	—	AY780425
<i>Coffea congensis</i> A.Froehner (1)	GenBank	DQ153868	DQ153501	DQ153632
<i>Coffea congensis</i> (2)	GenBank	DQ153834	DQ153467	DQ153598
<i>Coffea humilis</i> A.Chev. (1)	GenBank	DQ153847	DQ153480	DQ153611
<i>Coffea humilis</i> (2)	GenBank	DQ153854	DQ153487	DQ153618
<i>Coffea kapakata</i> (A.Chev.) Bridson (1)	GenBank	DQ153832	DQ153465	DQ153596
<i>Coffea kapakata</i> (2)	GenBank	DQ153850	DQ153483	DQ153614
<i>Coffea liberica</i> Bull. ex Hiern var. <i>dewevrei</i> (De Wild. & T.Durand) Lebrun	GenBank	DQ153839	DQ153472	DQ153603
<i>Coffea liberica</i> Bull. ex Hiern var. <i>liberica</i>	GenBank	DQ153846	DQ153479	DQ153610
<i>Coffea magnistipula</i> Stoff. & Robbr.	GenBank	DQ153876	DQ153509	DQ153640
<i>Coffea mayombensis</i> A.Chev.	GenBank	DQ153828	DQ153461	DQ153592
<i>Coffea montekupensis</i> Stoff. (1)	GenBank	DQ153826	DQ153459	DQ153590
<i>Coffea montekupensis</i> (2)	SD2717	MZ028620	MZ028622	—
<i>Coffea montekupensis</i> (3)	BSO5385	MZ028619	MZ028621	—
<i>Coffea rhamnifolia</i> (Chiov.) Bridson (1)	GenBank	DQ153825	DQ153458	DQ153589
<i>Coffea rhamnifolia</i> (2)	GenBank	FR832850	FR828681	FR832860
<i>Coffea rizetiana</i> Stoff. & M.Noïrot, sp. nov. (1)	20110297-63	MZ028617	MZ028623	MZ028616
<i>Coffea rizetiana</i> Stoff. & M.Noïrot, sp. nov. (2)	20110301-67	MZ028618	MZ028624	MZ028615

plastid *accD-psa1* and *trnL-F* gene markers were amplified following White *et al.* (1990) and Maurin *et al.* (2007). PCR reactions for all three gene markers investigated in this study consisted of 2 min initial denaturation at 94°C and 30 cycles of 30 s denaturation at 94°C, 30 s primer annealing at primer specific temperature and 1 min extension at 72°C. Primer annealing for ITS, *accD-psa1* and *trnL-F* were at 49°C, 51°C and 50°C, respectively. Amplification reactions were carried out on a GeneAmp PCR system 9700 (Applied Biosystems). Purified amplification products were sent to Macrogen, Inc. (Seoul, South Korea) for sequencing. Sequences generated in this study were submitted to GenBank.

Contiguous sequences were assembled using Geneious v7.0.6 (Biomatters, New Zealand). Automatic alignments were carried out with MAFFT (Katoh *et al.* 2002) under an E-INS-i algorithm, a 100PAM/k=2 scoring matrix, a gap open penalty of 1.3 and an offset value of 0.123. Subsequent manual fine tuning of the aligned dataset was done in Geneious v7.0.6. Congruency between the nuclear and chloroplast datasets was inferred by a partition homogeneity test as implemented in PAUP\*4.0b10a (Swofford 2002). In addition, possible conflicts between nuclear and chloroplast matrices were visually inspected, searching for conflicted relationships within each topology that are strongly supported (Johnson & Soltis 1998). The best-fit nucleotide substitution model for each plastid and nuclear dataset was determined using jModelTest 2.1.4 (Posada 2008) under the Akaike information criterion (AIC). The TVM+G model was found as best fit for ITS, whereas the GTR model was calculated as best substitution model for *accD-psa1* and *trnL-F*. A mixed-model approach was used in which the combined dataset is partitioned in order to apply a different model of evolution on each DNA region (Ronquist & Huelsenbeck 2003). Bayesian inference (BI) analyses were conducted with MrBayes v3.1 (Huelsenbeck & Ronquist

2001) on three individual data partitions and a combined data matrix. Each analysis was run twice for 10 million generations. Trees were sampled every 2500 generations. Inspection of chain convergence and ESS parameters was done with TRACER v1.4 (Rambaut & Drummond 2007). Bayesian inference BPP (Bayesian Posterior Probabilities) values between 0.50 and 0.95 using the 50% majority-rule consensus tree were considered to be weak, whereas BPP values above or equal 0.95 were taken into consideration (Suzuki *et al.* 2002). Maximum likelihood (ML) analyses were carried out on the CIPRES web portal using RAxML v7.2.8 (Stamatakis *et al.* 2008) under the GTRGAMMA model. Non-parametric ML bootstrapping analysis was calculated with 1000 bootstrap replicates.

## RESULTS

### MORPHOLOGICAL DELIMITATION OF

#### *COFFEA RIZETIANA* STOFF. & M.NOÏROT, SP. NOV.

In 1983, François Anthony, Emmanuel Couturon and Christian de Namur sampled a population of *Coffea* in Cameroon that appeared to contain a new species (Anthony *et al.* 1985). In 1992, Anthony identified it informally as a new variety of *C. liberica*. On the basis of his description, «feuilles allongées et étroites terminées par un acumen spatulé, et de très gros fruits noirs, contenant une pulpe épaisse et fibreuse rappelant celle de la variété *liberica*», Stoffelen (1998) suggested that the *Coffea* 'Koto', could be conspecific with *C. leonimontana* Stoff. A morphological study comparing herbarium and liquid preserved specimens showed that *C. leonimontana* was distinct by having smaller leaves 20-24 cm vs 24-34 cm, shorter acumen *c.* 1 cm vs 1.3 cm, thick vs thin leaves, elliptic vs obovate leaves, 2 vs 1 bracteal cup, longer corolla tube 4-7 mm vs 2-3 mm long, shorter and broader corolla lobes 15-17 × 5-7 mm vs 20 × 4-5 mm, round black fruit vs elongated red

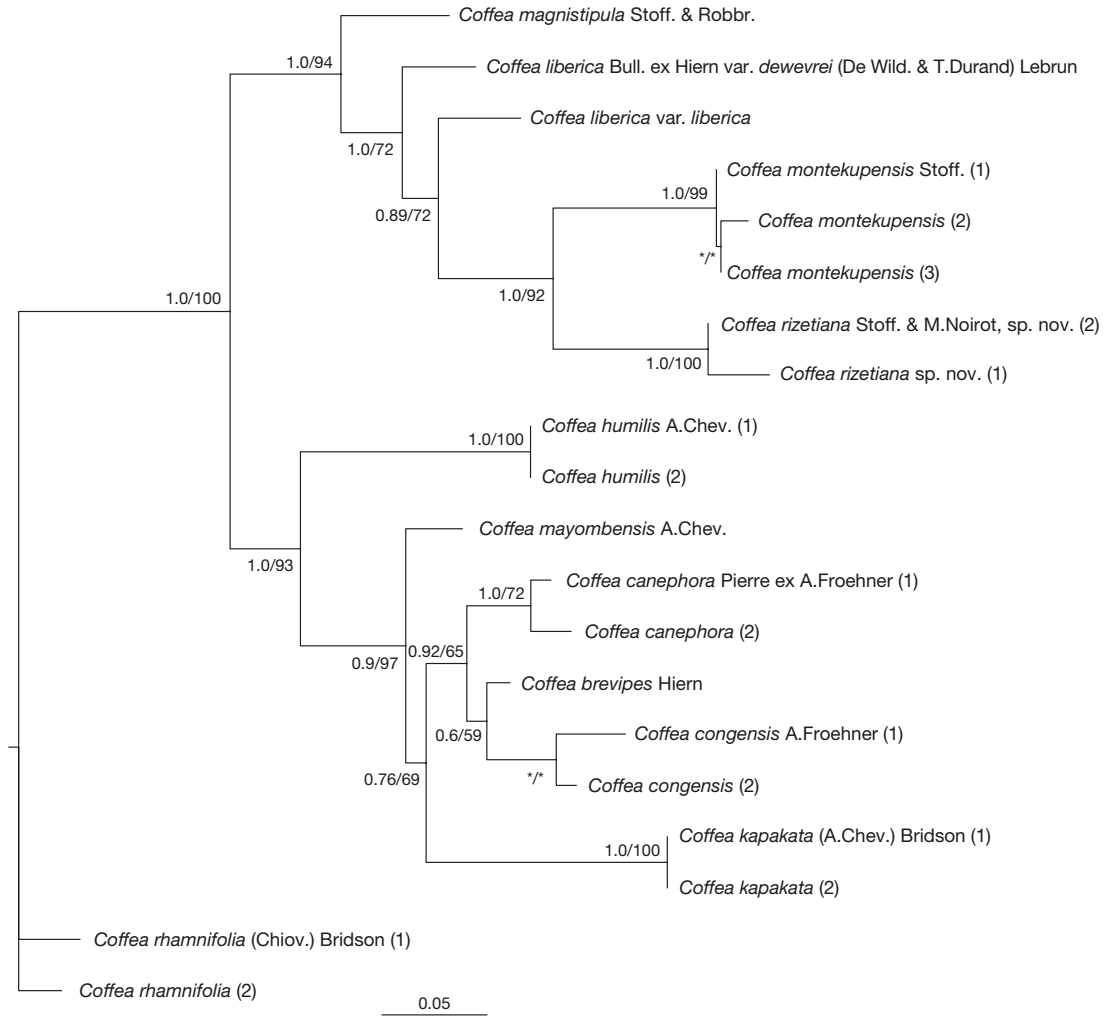


FIG. 1. — Phylogram based on combined *trnLF*, *accD-psa1* and ITS data. Numbers on branches represent Bayesian Posterior Probabilities and Maximum Likelihood Bootstrap Support, respectively. An asterisk indicates a lack of support.

TABLE 2. — Sequence statistics for the aligned data matrix.

	ITS	<i>accD-psa1</i>	<i>trnL-F</i>	combined
No. taxa	20	19	19	20
Aligned matrix length	801	1033	855	2689
No. variable characters	82	23	12	117

fruit, and larger fruit, more than 2 cm vs less than 2 cm. Also, the new species appeared to be morphologically and genetically related to *C. montekupensis* and *C. liberica sensu lato*. (Anthony 1992). However, it differed from *C. montekupensis* and *C. liberica* through vegetative and generative characters. These characters are mentioned in the diagnosis.

PHYLOGENETIC RELATIONSHIPS

Sequence characteristics of chloroplast and nuclear data matrices are summarized in Table 2. Bayesian inference (BI) and maximum likelihood (ML) analyses of both chloroplast and nuclear gene markers resulted in topologies with low to moderate Bayesian posterior probabilities (BPP) and ML

bootstrap support values (ML-BS; data not shown), with the exception for the relationship of the new *Coffea* species. Therefore, in case of incongruence between the different datasets, the incongruent lineages were not supported. In addition, the partition homogeneity was not significant ( $P > 0.05$ ) indicating that the chloroplast and nuclear partitions were not in conflict. The combined dataset resulted in a better resolved phylogeny with nodes that have a higher support compared to the separate plastid and nuclear dataset.

Fig. 1 shows the phylogenetic trees where four main results have been emphasized: 1) sister group relationships within the selected *Coffea* clade correspond to previous studies (Lashermes et al. 1997; Cros et al. 1998; Maurin et al. 2007; Anthony et al. 2010; Hamon et al. 2017), but generally better resolved; 2) the new species *C. rizetiana* Stoff. & M.Noïrot, sp. nov. belongs to the *C. liberica* group but not related to the *C. canephora* group; 3) both accessions of the new taxon *C. rizetiana* Stoff. & M.Noïrot, sp. nov. appear as most closely related lineages (BPP: 1.0, ML-BS: 100); and 4) the sister group relationship of the new species with *C. montekupensis* is also highly supported (BPP: 1.0, ML-BS: 92).

## SYSTEMATICS

Family RUBIACEAE Juss.

Genus *Coffea* L.*Coffea rizetiana* Stoff. & M.Noiro, sp. nov.  
(Figs 1-3)

The new species *C. rizetiana* Stoff. & M.Noiro, sp. nov. is related to *C. montekupensis* Stoff. and *C. liberica* Bull. ex Hiern, but it differs from *C. montekupensis* by having: elliptic vs obovate leaves; larger leaves (14)20-24(26) cm long and (5)6-9(10) cm wide vs 7-18(19) cm long and (2)2.5-5(6.5) cm wide; fewer secondary veins (8-10 pairs vs 12-13 pairs); longer corolla tube (4-7 mm vs 2.5-4 mm) and larger black fruits vs smaller red fruits and larger seeds (14-20 mm long × 8-13 mm vs 11-15 mm × 8-9 mm). It differs from *C. liberica* by having: shorter petioles (less than 1 cm vs 1-1.5 cm); acuminate leaf tips (vs apiculate or obtuse in *C. liberica*); a cuneate leaf base (vs rounded or rarely cuneate); a thinner leaf blade; shorter corolla tube; longer corolla lobes; and, black fruit (vs red or rarely yellowish or blackish; Fig. 3) with a thicker fleshy pericarp.

INFORMAL NAMES USED IN LITERATURE. — *Coffea* sp.: ‘Koto’ in Hamon *et al.* (2009, 2015), Razafinarivo *et al.* (2012), Noiro *et al.* (2016), Hamon *et al.* (2017).

*Coffea liberica* ‘Koto’: in Noiro *et al.* (1993), Cros *et al.* (1994), Anthony *et al.* (1997), Dussert *et al.* (1997a, 1997b), Lashermes *et al.* (1996, 1997), Stoffelen (1998: 102, 103, 110, 125), Stoffelen *et al.* (1999), Fernandez & Lashermes (2002), Noiro *et al.* (2003), Campa *et al.* (2004, 2005a, 2005b, 2008), Dubreuil-Tranchant *et al.* (2011).

TYPE SPECIMEN. — *Stoffelen 2045* (holo-, BR[BR0000026083674V]; iso-, P, YA), 15.IV.2016 collected in the greenhouses of Meise Botanic Garden, Belgium; grown from material collected in 1983 in Cameroon, south of the Mouyouka-Kompina forest, 4°22'N, 9°34'E, a forest road, to the left of the Penda-Mboko asphalt road, after 2.6 km leads to the population.

OTHER LIQUID PRESERVED SPECIMENS STUDIED. — *Noiro EC66a*, 3.XII.2012, flowering (BR, WAG); *Noiro EC66b*, 31.III.2013, fruiting (BR); *Noiro EC54*, 31.I.2013, flowering (BR, WAG); *Noiro EC52* (BR), sterile; *Noiro EC61*, sterile (BR); *Noiro EC63*, sterile (BR, WAG); *Noiro EC66*, 30.X.2012. All these specimens were collected in the germplasm collection in the Bassin Martin coffee collection.

DISTRIBUTION. — Cameroon, only known from one population at the south of the Mouyouka-Kompina forest, N 4°22', E 9°34'. A forest road, on the left of the Penda-Mboko asphalt road, leads to the population after 2.6 km.

HABITAT. — Understory of secondary rain forest, forest at 60 m above sea level, sympatric with *C. canephora*.

ETYMOLOGY. — This species is dedicated to the Professor Georges Rizet, Geneticist at the Orsay University Paris XI. Scientific adviser to the ORSTOM Director, he was central to the survey and genetic studies of the wild coffee genetic resources collected by IRD (ex-ORSTOM), Cirad and MNHN from 1966 to 1984.

## DESCRIPTION

Small tree up to 6 m tall, young branchlets glabrous. Leaf-blades elliptic, (14)20-24(26) cm long, (5)6-9(10) cm wide, tip acuminate, acumen *c.* 1 cm, with rounded tip; small pit domatia at the junction of the midrib and the secondary veins, glabrous; 8-10 secondary veins at either

side of the midrib. Petiole 0.5-1(1.3) cm long. Stipules 0.2-0.4 cm long, truncated to slightly obtuse, in young stage covered with exudate.

1-2(3) inflorescence per axil, each with 1(2) flowers and supported by two truncate bracteal cups, lower cup with 2 small reduced leaves-like appendages, 1-1.5 mm long; inflorescence stalk and peduncle in total *c.* 4 mm long.

Flowers 6 (7)-merous. Calyx reduced to a small rim, truncate. Corolla white; tube 4-7 mm long; lobes 15-17 mm long, 5-7 mm broad; anthers 7-12 mm long, filament 2-5 mm long; ovary glabrous, not surrounded by the bracteal cups; disc *c.* 1 mm high.

Infructescence 1-2 per axil. Fruit 1 per infructescence. Fruit black to dark maroon-brown, fleshy pericarp (Fig. 3). Short peduncle *c.* 5 mm. Fruit almost spherical, somewhat truncate at top, (1.5) 2-3 cm long, 1.6-2.3 wide along the septum and 2-2.6 cm perpendicular to the septum. Disk and calyx reduced, small and not accrescent. Seeds small elliptic to elliptic, 14-20 mm long, 8-13 mm wide and *c.* 5-7 mm thick. Habitus, fruits and flowers are illustrated in Fig. 2.

## DISCUSSION

The genus *Coffea* L. *sensu stricto* comprises 103 species (Davis *et al.* 2006) and occurs naturally in tropical Africa, Madagascar, the Comoros and the Mascarenes (Réunion and Mauritius). *Coffea* is a member of the tribe Coffeae (comprising 11 genera) and is closely allied to the genus *Psilanthus* Hook.f., with which it shares the typical “coffee bean” fruit morphology (Davis *et al.* 2007; Maurin *et al.* 2007). Davis (2011) subsumed *Psilanthus* into the genus *Coffea*. This merge of the genera increased the number of species to 124, extended the distribution area of the genus *Coffea* to tropical Asia and Australasia and broadened the morphological characterisation of the genus.

*C. rizetiana* Stoff. & M.Noiro, sp. nov. has the typical “coffee bean” seed morphology, a character which is restricted to the genus *Coffea s.s.* and the genus *Psilanthus* (Stoffelen 1998; Davis *et al.* 2006). The exerted style and anthers place it in the genus *Coffea s.s.* (Stoffelen 1998) or *Coffea* subgenus *Coffea sensu* Davis *et al.* (2005). It is diploid (Cros *et al.* 1994; Noiro *et al.* 2003) like most coffee species.

The new species has only been reported from the original collected locality in 1983 by Anthony, Couturon and de Namur. After screening the different herbaria, we concluded that *C. rizetiana* Stoff. & M.Noiro, sp. nov. has not been collected elsewhere. Moreover, the site where it was collected in 1983 has been cleared for agriculture. The species is therefore presumed to be extinct in the wild. The species is however still present in the living collections at Réunion and the Meise Botanic Garden.

*C. rizetiana* Stoff. & M.Noiro, sp. nov. has particular fruit characteristics, namely its colour (black instead of red) and a very thick fruit mesocarp, these are both exceptional for *Coffea* species. On the African continent, the black fruits

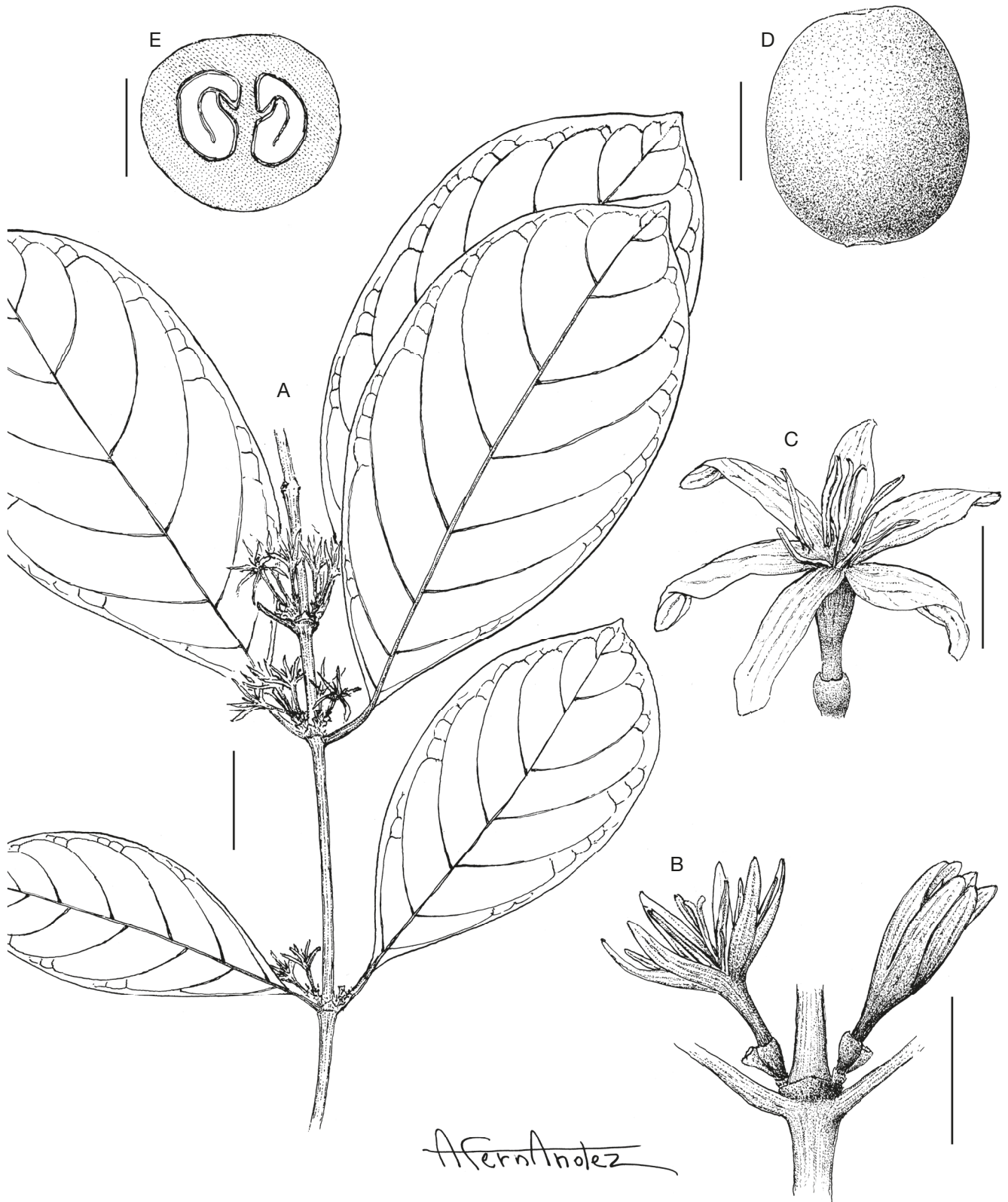


FIG. 2. — *C. rizetiana* Stoff. & M.Noïrot, sp. nov.: **A**, habit; **B**, node with flowers and petioles; **C**, detail of flower; **D**, fruit; **E**, transection of the fruit with two seeds. Vouchers deposited in BR: **A-C**, Stoffelen 2045; **C, D**, Noïrot EC66, 30.X.2012 (liquid preserved collection). Scale bars: A, 3 cm; B, 2 cm; C-E, 1 cm.

have only been reported for *C. stenophylla* G.Don and *C. togoensis* A.Chev., and the fleshy fruits of *C. rizetiana* Stoff. & M.Noïrot, sp. nov. are the largest on the African

continent. Colour and size of ripe fruits are important characters as they are related to the concept of co-evolution (Ehrlich & Raven 1964; Janzen 1966). Fruit traits have a





FIG. 3. — *C. rizetiana* Stoff. & M.Noïrot, sp. nov., fruit showing its basis (right) and top (left) from the *Coffea* L. collection at Bassin Martin (Réunion, France).

direct impact on attracting vertebrates that disperse fruits and seeds (Gauthier-Hion *et al.* 1985). According to these authors, two elements are important in the relationship between fruit traits and vertebrate dispersal agents: (1) fruit size versus dispersal agents, from birds to large mammals including large rodents and elephants; and (2) red to green fruits where dispersion is mainly carried out by monkeys and squirrels respectively. The role of monkeys and bats in the seed dispersion of *C. canephora* was studied in the Kibale forest of Uganda (Oryem-Origa 1999). If this is taken into account, *C. rizetiana* Stoff. & M.Noïrot, sp. nov. and its sister *C. montekupensis*, where both size and colour of fruits differ, should be dispersed by different animals. The shift in fruit colour and size coupled to the change of dispersers are relatively recent events as they emerged in a final clade. This recent divergence in both species would be driven by a co-evolution process affecting a limited number of genes.

*C. rizetiana* Stoff. & M.Noïrot, sp. nov. and *C. montekupensis* are two sister species that not only probably have different dispersers, but also occupy different ecological niches: lowland rain forest near to Mount Kupe (c. 90 m elevation) vs (sub)montane rain forest on the slopes of Mount Kupe, respectively. The driving force behind this speciation event is unknown. Based on the current data, we consider Mount Kupe an interesting region to investigate speciation patterns within the genus *Coffea*. At least six different *Coffea* and *Psilanthus* species occur on Mount Kupe and its immediate surroundings, namely *C. rizetiana* Stoff. & M.Noïrot, sp. nov., *Coffea liberica*, *C. bakossii* Cheek & Bridson, *C. brevipes*, *C. montekupensis* and *P. mannii*. Each species occupies a specific ecological niche with the exception of *C. liberica* and *P. mannii* which seem to be less specific in their niche preference. *C. rizetiana* Stoff. & M.Noïrot, sp. nov. is a lowland species, only occurring in lowland rain forest. While, *C. liberica* is a species with a wide Guineo-congolian distribution from low land to submontane forest. On Mount Kupe, *C. liberica* does not occupy a particular niche and has a broad altitudinal range. *C. liberica* is sympatric (or at least parapatric) with *C. montekupensis* and *C. bakossii*, and probably also with *P. mannii*. *C. brevipes* is also widely

spread with a lower Guinean and Congolian distribution, including altitude. On Mount Kupe it has only been found on the drier north face. *C. montekupensis* in contrast occurs on the wet western and southern slopes of Mount Kupe at an elevation of 800 m to 1500 m. It is quite abundant within this altitudinal range on Mount Kupe and the adjoining Bakossi Mountains (Stoffelen *et al.* 1997). Finally, *C. bakossii* is a submontane species, occurring at elevations between 700 and 900 m (Cheek *et al.* 2002). It is rare and limited in the number of sites. Population studies can give some insight in the genetic isolation and genetic variability within the species. Despite *C. canephora* (robusta coffee) being cultivated for more than 50 years in lowland areas, it does not occur in the forests of Mount Kupe and the Bakossi Mountains. *C. canephora* has been reported from the lowland rain forest in the region of Mount Cameroon.

In the broader region of South-West Cameroon, including Mount Cameroon, several other species (*P. aff. ebracteolatus*, *C. charreriana*, *C. heterocalyx*, *C. leonimontana* and *C. mayombensis* A.Chev.) are found. South-West Cameroon comprises 12 accepted species, this is the most important diversity hotspot for the genus *Coffea* on the African continent. This area should be designated as a priority for *in situ* and *ex situ* conservation of the wild relatives of *Coffea* and many other plant species.

#### Acknowledgements

We are grateful to all the curators and technicians of the different herbaria (P, WAG, KEW, LISC, COI, G, MO, MA, YA, YBI) who sent specimens on loan to BR or who allowed Piet Stoffelen to visit and study their *Coffea* and *Psilanthus* collections. Special thanks to Antonio Fernandez for the preparation of the drawing and to the curators and gardeners who were in charge of the *ex situ* collections where the living *Coffea* specimens were growing. We also thank Henry Engledow for English revision and André Charrier and an anonymous referee, who both helped us in revising the manuscript.

REFERENCES

- ANTHONY F. 1992. — *Les ressources génétiques des caféiers: collecte, gestion d'un conservatoire et évaluation de la diversité génétique*. ORSTOM, Paris, 320 p. (Travaux et Documents micro-édités; 81).
- ANTHONY F., COUTURON E. & DE NAMUR C. 1985. — Les caféiers sauvages du Cameroun: résultats d'une mission de prospection effectuée par l'ORSTOM en 1983. *Proceeding of the 11<sup>th</sup> International Congress of ASIC*: 495-505. <https://www.asic-cafe.org/conference>
- ANTHONY F., BERTRAND B., LASHERMES P. & CHARRIER A. 1997. — La biologie moléculaire en appui à l'amélioration génétique du caféier Arabica. *Plantations, Recherche, Développement* 4: 369-374. <https://agritrop.cirad.fr/389637/>
- ANTHONY F., DINIZ L. E. C., COMBES M. C. & LASHERMES P. 2010. — Adaptive radiation in *Coffea* subgenus *Coffea* L. (Rubiaceae) in Africa and Madagascar. *Plant Systematics and Evolution* 285: 51-64. <https://doi.org/10.1007/s00606-009-0255-8>
- CAMPA C., BALLESTER J. F., DOULBEAU S., DUSSERT S., HAMON S. & NOIROT M. 2004. — Trigonelline and sucrose diversity in wild coffee species. *Food Chemistry* 88: 39-43. <https://doi.org/10.1016/j.foodchem.2004.01.020>
- CAMPA C., DOULBEAU S., DUSSERT S., HAMON S. & NOIROT M. 2005a. — Diversity in bean caffeine content among wild *Coffea* species. Evidence of a discontinuous distribution. *Food Chemistry* 91: 633-637. <https://doi.org/10.1016/j.foodchem.2004.06.032>
- CAMPA C., DOULBEAU S., DUSSERT S., HAMON S. & NOIROT M. 2005b. — Quantitative relationship between caffeine and chlorogenic acid contents among wild *Coffea* species. *Food Chemistry* 93: 135-139. <https://doi.org/10.1016/j.foodchem.2004.10.015>
- CAMPA C., RAKOTOMALALA J. J., DE KOCHKO A. & HAMON S. 2008. — Chlorogenic acids: diversity in green beans of wild coffee species, in HEMANTARANJAN A. (ed.), *Advances in Plant Physiology*. Vol. 10. Scientific Publishers, Jodhpur, India: 421-437.
- CHEEK M., CSIBA L. & BRIDSON D. 2002. — A new species of *Coffea* (Rubiaceae) from Western Cameroon. *Kew Bulletin* 57: 675-680. <https://doi.org/10.2307/4110998>
- CROS J., GAVALDA M. C., CHABRILLANGE N., RÉCALT C., DUPERRAY C. & HAMON S. 1994. — Variations in the total nuclear DNA content in African *Coffea* species (Rubiaceae). *Café Cacao Thé* 38: 3-10.
- CROS J., COMBES M. C., TROUSLOT P., ANTHONY F., HAMON S., CHARRIER A. & LASHERMES P. 1998. — Phylogenetic analysis of chloroplast DNA variation in *Coffea* L. *Molecular phylogenetics and evolution* 9: 109-117. <https://doi.org/10.1006/mpev.1997.0453>
- DAVIS A. P. 2011. — *Psilanthus manni*, the type species of *Psilanthus*, transferred to *Coffea*. *Nordic Journal of Botany* 29: 471-472. <https://doi.org/10.1111/j.1756-1051.2011.01113.x>
- DAVIS A. P., BRIDSON D. & RAKOTONASOLO F. 2005. — A reexamination of *Coffea* subgenus *Baracoffea* and comments on the morphology and classification of *Coffea* and *Psilanthus* (Rubiaceae-Coffeae), in KEATING R. C., HOLLOWELL V. & CROAT T. B. (eds.), *A Festschrift for William G. D'Arcy: the Legacy of a Taxonomist*. Missouri Botanical Garden, St. Louis: 399-420. <https://www.biodiversitylibrary.org/page/59962457>
- DAVIS A. P., GOVAERTS R., BRIDSON D. M. & STOFFELEN P. 2006. — An annotated taxonomic conspectus of the genus *Coffea* (Rubiaceae). *Botanical Journal of the Linnean Society* 152: 465-512. <https://doi.org/10.1111/j.1095-8339.2006.00584.x>
- DAVIS A. P., CHESTER M., MAURIN O. & FAY M. F. 2007. — Searching for the relatives of *Coffea* (Rubiaceae, Ixoroideae): the circumscription and phylogeny of Coffeae based on plastid sequence data and morphology. *American Journal of Botany* 94: 313-329. <https://doi.org/10.3732/ajb.94.3.313>
- DE NAMUR C., COUTURON E., SITA P. & ANTHONY F. 1987. — Résultats d'une mission de prospection des caféiers sauvages du Congo. *Proceeding of the 12<sup>th</sup> International Congress of ASIC*: 397-404. <https://www.asic-cafe.org/conference>
- DUBREUIL-TRANCHANT C., GUYOT R., GUELLIN A., DURET C. DE, MARE M. DE LA, RAZAFINARIVO N., PONCET V., HAMON S., HAMON P. & KOCHKO A. DE 2011. — Site-specific insertion polymorphism of the MITE *Alex-1* in the genus *Coffea* suggests interspecific gene flow. *International Journal of Evolutionary Biology*: 9 p. <https://doi.org/10.4061/2011/358412>
- DUSSERT S., CHABRILLANGE N., ANTHONY F., ENGELMANN F., RECALT C. & HAMON S. 1997a. — Variability in storage response within a coffee (*Coffea* spp.) core collection under slow growth collection. *Plant Cell Reports* 16: 344-348. <https://doi.org/10.1007/BF01088294>
- DUSSERT S., ENGELMANN F., CHABRILLANGE N., ANTHONY F., NOIROT M. & HAMON S. 1997b. — *In vitro* conservation of coffee (*Coffea* spp.) germplasm, in RAZDAN M. K. & COCKING E. C. (eds), *Conservation of Genetic Resources in vitro*. Vol. 1. Science Publishers, New York: 287-305.
- EHRlich P. R. & RAVEN P. H. 1964. — Butterflies and plants: a study in coevolution. *Evolution* 18: 586-608. <https://doi.org/10.1111/j.1558-5646.1964.tb01674.x>
- FERNANDEZ D. & LASHERMES P. 2002. — Molecular tools for improving coffee (*Coffea arabica* L.) resistance to parasites, in MOHAN JAIN S., BRAR D. S. & AHLWALIA B. S. (eds), *Molecular Techniques in Crop Improvement*. Springer Netherlands: 327-345.
- GAUTHIER-HION A., DUPLANTIER J. M., QURIS R., FEER F., SOURD C., DECoux J.-P., DUBOST D., EMMONS L., HECKETSWEILER P., MOUNGAZI A., ROUSSILHON C. & THIOLLAY J.-M. 1985. — Fruit characters as a basis of fruit choice and seed dispersal in a tropical forest vertebrate community. *Oecologia* 65: 324-337. <https://doi.org/10.1007/BF00378906>
- HAMON P., SILJAK-YAKOVLEV S., SRISUWAN S., ROBIN O., PONCET V., HAMON S. & KOCHKO A. DE 2009. — Physical mapping of rDNA and heterochromatin in chromosomes of 16 *Coffea* species: a revised view of species differentiation. *Chromosome Research* 17: 291-304. <https://doi.org/10.1007/s10577-009-9033-2>
- HAMON P., RAKOTOMALALA J. J., AKAFFOU S., RAZAFINARIVO N. J., COUTURON E., GUYOT R., CROUZILLAT D., HAMON S. & KOCHKO A. DE 2015. — Caffeine-free species in the genus *Coffea*, in PREEDY V. R. (ed.), *Coffee in Health and Disease Prevention*. Elsevier, London: 39-44. <https://doi.org/10.1016/B978-0-12-409517-5.00005-X>
- HAMON P., GROVER C. E., DAVIS A. P., RAKOTOMALALA J. J., RAHARIMALALA N. E., ALBERT V. A., SREENATH H. L., STOFFELEN P., MITCHELL S. E., COUTURON E., HAMON S., KOCHKO A. DE, CROUZILLAT D., RIGOREAU M., SUMIRAT U., AKAFFOU S. & GUYOT R. 2017. — Genotyping-by-sequencing provides the first well-resolved phylogeny for coffee (*Coffea*) and insights into the evolution of caffeine content in its species: GBS coffee phylogeny and the evolution of caffeine content. *Molecular Phylogenetics and Evolution* 109: 351-361. <https://doi.org/10.1016/j.ympev.2017.02.009>
- HOLMGREN P. K., KEUKEN W. & SCHOFIELD E. K. 1981. — *Index Herbarium*. Dr. W. Junk B. V., The Hague, 452 p.
- HUELSENBECK J. P. & RONQUIST F. 2001. — MRBAYES, Bayesian inference of phylogenetic trees. *Bioinformatics* 17: 754-755. <https://doi.org/10.1093/bioinformatics/17.8.754>
- JANSENS S., GEUTEN K., YUAN Y.-M., SONG Y., KÜPPER P. & SMETS E. 2006. — Phylogenetics of Impatiens and Hydrocera (Balsaminaceae) using chloroplast atpB-rbcL spacer sequences. *Systematic Botany* 31: 171-180. <https://doi.org/10.1600/036364406775971796>
- JANSENS S. B., VIAENE T., HUYSMANS S., SMETS E. F. & GEUTEN K. P. 2008. — Selection on length mutations after frameshift can explain the origin and retention of AP3/DEF like paralogues in Impatiens. *Journal of Molecular Evolution* 66: 424-435. <https://doi.org/10.1007/s00239-008-9085-5>
- JANZEN D. H. 1966. — Coevolution of mutualism between ants and acacias in Central America. *Evolution* 20: 249-275. <https://doi.org/10.1111/j.1558-5646.1966.tb03364.x>
- JOHNSON L. A. & SOLTIS D. E. 1998. — Assessing congruence:

- Empirical examples from molecular data, in SOLTIS P. S., SOLTIS D. E. & DOYLE J. J. (eds), *Molecular Systematics of Plants II: DNA Sequencing*. Kluwer, Boston: 297-348.
- KATOH K., MISAWA K., KUMA K. & MIYATA T. 2002. — MAFFT: a novel method for rapid multiple sequence alignment based on Fourier transform. *Nucleic Acids Research* 30: 3059-3066. <https://doi.org/10.1093/nar/gkf436>
- LASHERMES P., COMBES M. C., TROUSLOT P., ANTHONY F., HAMON S. & CHARRIER A. 1996. — Inheritance and restriction fragment length polymorphism of chloroplast DNA in the genus *Coffea* L. *Theoretical Applied Genetics* 93: 626-632. <https://doi.org/10.1007/BF00417958>
- LASHERMES P., COMBES M. C., TROUSLOT P. & CHARRIER A. 1997. — Phylogenetic relationships of coffee-tree species (*Coffea* L.) as inferred from ITS sequences of nuclear ribosomal DNA. *Theoretical and Applied Genetics* 95: 947-955. <https://doi.org/10.1007/S001220050500>
- MAURIN O., DAVIS A. P., CHESTER M., MVUNGI E. F., JAUFEEALLY-FAKIM Y. & FAY M. F. 2007. — Towards a phylogeny for *Coffea* (Rubiaceae): identifying well-supported lineages based on nuclear and plastid DNA sequences. *Annals of Botany* 100: 1565-1583. <https://doi.org/10.1093/aob/mcm257>
- NOIROT M., HAMON S. & ANTHONY F. 1993. — L'obtention d'une core collection de caféiers. Définition des groupes d'échantillonnage et méthodologie. *Proceeding of the 15<sup>th</sup> International Congress of ASIC*: 183-190. <https://www.asic-cafe.org/conference/15th-international-scientific-colloquium-coffee>
- NOIROT M., PONCET V., BARRE P., HAMON S. & DE KOCHKO A. 2003. — Genome size variations in diploid African *Coffea* species. *Annals of Botany* 92: 709-714. <https://doi.org/10.1093/aob/mcg183>
- NOIROT M., CHARRIER A., STOFFELEN P. & ANTHONY F. 2016. — Reproductive isolation, gene flow and speciation in the former *Coffea* sgenus: a review. *Trees* 30: 597-608. <https://doi.org/10.1007/s00468-015-1335-8>
- ORYEM-ORIGA H. 1999. — Fruit and seed ecology of wild robusta coffee (*Coffea canephora* Froehner) in Kibale National Park, Uganda. *African Journal of Ecology* 37: 439-448. <https://doi.org/10.1046/j.1365-2028.1999.00200.x>
- POSADA D. 2008. — jModelTest: phylogenetic model averaging. *Molecular Biology and Evolution* 25: 1253-1256. <https://doi.org/10.1093/molbev/msn083>
- RAMBAUT A. & DRUMMOND A. J. 2007. — *Tracer v1.4*. Available from <http://beast.bio.ed.ac.uk/Tracer> (accessed: 30 March 2016).
- RAZAFINARIVO N. J., RAKOTOMALALA J. J., BROWN S. C., BOURGE M., HAMON S. ET AL. 2012. — Geographical gradients in the genome size variation of wild coffee trees (*Coffea*) native to Africa and Indian Ocean islands. *Tree Genetics & Genomes* 8: 1345-1358. <https://doi.org/10.1007/s11295-012-0520-9>
- ROBBRECHT E. 1988. — *Tropical Woody Rubiaceae*. Opera Botanica Belgica 1, 272 p.
- RONQUIST F. & HUELSENBECK J. P. 2003. — Mr. Bayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics* 19: 1572-1574. <https://doi.org/10.1093/bioinformatics/btg180>
- STAMATAKIS A., HOOVER P. & ROUGEMENT J. 2008. — A rapid bootstrapping algorithm for the RAxML web servers. *Systematic Biology* 57: 758-771. <https://doi.org/10.1080/10635150802429642>
- STOFFELEN P. 1998. — *Coffea and Psilanthus in tropical Africa: a systematic and palynological study, including a revision of the West and Central African species*. Dissertation, Katholieke Universiteit, Leuven, 270 p.
- STOFFELEN P., CHEEK M., BRIDSON D. & ROBBRECHT E. 1997. — A new species of *Coffea* (Rubiaceae) and notes on Mount Kupe (Cameroon). *Kew bulletin* 52: 989-994.
- STOFFELEN P., ROBBRECHT E. & SMETS E. 1999. — A new species of *Coffea* (Rubiaceae) from central Africa with notes on tentative other taxa. *Systematics and Geography of Plants* 69: 119-124.
- STOFFELEN P., NOIROT M., COUTURON E. & ANTHONY F. 2008. — A new caffeine-free coffee from Cameroon. *Botanical Journal of the Linnean Society* 158: 67-72. <https://doi.org/10.1111/j.1095-8339.2008.00845.x>
- STOFFELEN P., NOIROT M., COUTURON E., BONTEMS S., DE BLOCK P. & ANTHONY F. 2009. — *Coffea anthonyi*, a self-compatible Central African coffee species, closely related to an ancestor of *C. arabica* L. *Taxon* 58: 133-140. <https://doi.org/10.1002/tax.581014>
- SUZUKI Y., GLAZKO G. V. & NEI M. 2002. — Over credibility of molecular phylogenies obtained by Bayesian phylogenetics. *Proceedings of the National Academy of Science USA* 99: 16138-16143. <https://doi.org/10.1073/pnas.212646199>
- SWOFFORD D. L. 2002. — *PAUP\*: Phylogenetic Analysis Using Parsimony (\*and other methods), version 4.0b10*. Sinauer Associates, Sunderland, USA.
- WHITE T. J., BRUNS T. D., LEE S. B. & TAYLOR J. W. 1990. — Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics, in INNIS M. A., GELFAND D. H., SNINSKY J. I. & WHITE T. J. (eds.), *PCR Protocols: A Guide to Methods and Applications*. Academic Press, San Diego, California: 315-322.

Submitted on 16 November 2020;  
accepted on 9 May 2021;  
published on 21 December 2021.