Cypricola 23: 1–12. 8-Sep-2023

Earliest evidence of *Cyprinia gracilis* (Boiss.) Browicz in Cyprus: archaeobotanical data from Khirokitia

Maria Rousou

Abstract. – *Cyprinia gracilis* (Boiss.) Browicz (*Apocynaceae*) is a near-endemic species occurring in Cyprus and Southern Turkey. It is a climber growing through pine forests, as well as maquis formations, mainly in humid locations. This work presents the earliest evidence of *C. gracilis* in Cyprus, through the study and identification of wood charcoal (anthracological) remains recovered at the site of Khirokoitia (Choirokoitia) Vounoi in Cyprus. The results contribute to the study of the biogeography of this species, showing its presence in Cyprus since the early Holocene. It also contributes to the study of the palaeoenvironment during the human occupation of the site, and the exploitation of plant resources.

Address. – Maria Rousou, UMR 7209, MNHN-CNRS: Archaeozoology, Archaeobotany: Societies, Practices and Environments (AASPE), 55 rue Buffon, CP 56, 75005 Paris, France & Department of History and Archaeology, Archaeological Research Unit, University of Cyprus, P.O. Box 20537, 1678 Nicosia, Cyprus; mariarousou30@gmail.com

Introduction

Cyprinia (Boiss.) Browicz is a monotypic genus of plants in the family *Apocynaceae* (formerly attributed to the *Asclepiadaceae* family, now considered as a synonym of the *Apocynaceae*) consisting of a single species, *C. gracilis* (Boiss.) Browicz (Browicz 1966a, b, Holmboe 1914, Meikle 1985). The generic name *Cyprinia* derives from the island of Cyprus, the main region of distribution of *C. gracilis* (Browicz 1966b).

The taxonomy of *C. gracilis* is still disputed and some authors consider it as a synonym to *Periploca gracilis*.

In his 'Flora orientalis', Boissier (1879) described a new species in the genus *Periploca*, named *P. gracilis* Boiss. (*Asclepiadeae*), characterised by smaller flowers, grabrous corolla lobes, and bifid corona lobes. These characteristics distinguished it from *P. graeca*. Schumann (1895) accepted this classification (*P. gracilis*, *Periplocoideae-Periploceae*) and divided the genus *Periploca* into two sections: *Euperiploca* and *Campelepis*. *P. gracilis* was attributed together with *P. reussii* to the section *Euperiploca*.

Browicz (1966a, b) transferred *P. gracilis* to a new genus, named *Cyprinia* Browicz, containing only one species – *C. gracilis,* considering its linear bifid corona lobes as a strong, distinct morphological character. Other important morphological characteristics of *C. gracilis* are the white, thin, glabrous, not retuse, ribbonlike, translucent corolla lobes with two distinct veins. This classification is also accepted in Meikle's (1985) 'Flora of Cyprus'.

According to Venter's (1997) revision of the genus *Periploca*, *C. gracilis* should be considered a synonym of *P. gracilis* (section *Periploca*), due to the trisegmented con-

dition of the corona lobes which is unique to *Periploca*. However, this still needs to be confirmed by molecular analyses.

C. gracilis is a slender woody scrambler or climber, with thin, glabrous shoots, green in the first year, dark brown in older growths, containing milky latex, reaching more than 8 m in height. The leaves are persistent, opposite, simple, entire, coriaceous, glabrous, ovate-elliptic, elliptic to lanceolate, 1–5 × 0.3–2 cm, dark green, shortly stalked. The inflorescence is a (1–)3(–5)-flowered cyme, usually as long or a little longer than the subtending leaves. Its flowering period is from May to August (–September). The corolla is white, greenish, or greenish-yellow, deeply divided into obtuse or subacute, strap-shaped, or narrowly lanceolate lobes 6–7 × 1.5–2 mm. The fruit is a glabrous, cylindrical, straight or slightly arched, pale brown follicle, 5–7 × 0.3–0.5 cm; the seeds are narrowly oblong, flattened, 5–7 × 1.5–2 mm, longitudinally costate, one side with a rounded protrusion, the other with a distinct longitudinal strip across the middle, with an apical tuft of hairs. It ripens from September to November (Meikle 1985, Tsintides & al. 2002). Pollen grains are arranged in planar tetrads (Halbritter & al. 2018, Verhoeven & Venter 1994).

C. gracilis is a near-endemic species, indigenous only to Cyprus and Southern Turkey (Mersin, Cilicia) (Browicz 1966a, b). In Cyprus, it occurs in various localities within the botanical divisions 1–4 and 7 (for the botanical divisions, see Meikle 1977), from 50 to 1225 m a.s.l. (Barbero & Quézel 1979, Hand 2004, Holmboe 1914, Meikle 1985). Recent mapping of the species documented findings in the following localities: Machairas forest, Mantra tou Kampiou, Ezousa river (Kannaviou-Agia Panagia), Akamas, Kavo Gkreko, Episkopi, Kouka, Mosfiloti, Kantara, Kato Amiantos and many places around Troodos (C. S. Christodoulou, G. Hadjikyriakou, pers. comm.).

It grows on igneous or calcareous mountainsides, on marly limestones, marls, white limestones with small flint banks, in arid scrubland and wasteland, together with *Rhus coriaria*, *Rubus sanctus*, *Pistacia* sp., *Rosa* sp., *Lonicera etrusca*, *Berberis cretica*, *Smilax aspera*, *Arbutus andrachne*, *Quercus alnifolia* and *Pinus nigra* subsp. *pallasiana* on the Troodos mountains (Alziar 1999, Barbero & Quézel 1979, Browicz 1966a, b, Meikle 1985), but also in more humid locations through the Oleo-Ceratonion maquis together with *Ceratonia siliqua*, *Olea europaea*, *Pistacia* sp., *Rhamnus* sp., *Prasium majus*, *Calycotome villosa*, *Genista fasselata*, *Anagyris foetida* and *Myrtus communis* (Barbero & Quézel 1979, Delipetrou & Christodoulou 2010, Hand 2004, Holmboe 1914).

Little is known about the evolutionary history of *C. gracilis*, and there are not any palaeontological attestations of this species in the available palaeobotanical record (Hadjikyriakou 2017, Holmboe 1914, Kassapis 2011). Its presence in Cyprus is known so far only from modern specimens observed in the field and herbaria collections (Browicz 1966a, b, Holmboe 1914, Meikle 1985, Tsintides & al. 2002). This study aims to present information on the earliest evidence of *C. gracilis* in Cyprus, identified by charcoal remains recovered at the archaeological site Khirokitia Vounoi, in Larnaka district. This contributes to the discussion concerning the botanical history of this species in Cyprus and the reconstruction of the palaeoenvironment during the human occupation of Khirokitia.

The archaeological site and archaeological context of the charcoal remains

The archaeological site of Khirokitia (alternatively Choirokoitia) Vounoi is situated on the southern coast of Cyprus, about 6 km from the present coastline (Fig. 1a–e). The archaeological site was discovered and investigated between 1936 and 1946 by Porphyrios Dikaios (Department of Antiquities, Republic of Cyprus) (Dikaios 1953) and from 1976 to 2009 by Alain Le Brun (French Archaeological Mission, MAE-CNRS) (Le Brun 1984, 1989, 1994, 2021).

The archaeological investigations have revealed the presence of a large settlement (the surface of the built-up area at the time of its maximum extent may be estimated to 3 ha), located over the sides of a hill (Fig. 1b–e). The village is composed of circular buildings and surrounded by an enclosure wall whose outline has been modified several times (Dikaios 1953, Le Brun 1984, 1989, 1994, 2021). The site is dated to the Late Aceramic Neolithic period, also known as 'Khirokitian culture' and the occupation of the site is dated from around 6800 to 5500 cal BC. The exceptionally well-preserved charred plant macroremains (wood charcoal, seeds, fruits) consist an important source of information on the environment surrounding the site during its occupation, the exploitation and uses of plant resources by the inhabitants of the neolithic village, as well as the palaeoenvironment of Cyprus and its evolution during the Late Aceramic Neolithic (Hansen 1989, 1994, Miller 1984, Parés 2013, 2015, Rousou 2022, 2023a, b, Rousou & al. 2021, Thiébault 2003).

This study discusses ten wood charcoal remains that were recovered in the anthracological assemblages of Khirokitia. They were retrieved from two samples (13990, 14006) from two exterior areas (e.1226 and e.1267, respectively) dated to level F4 (Fig. 1c), that is to the early occupation of the village.

Material and Methods

The study and taxonomic identification of wood charcoal (alternatively anthracological) remains, were carried out at the Plateau Archéobotanique of the UMR 7209: Archaeozoology, Archaeobotany: Societies, Practices and Environments (AASPE) of the Muséum national d'Histoire Naturelle in Paris, using an Olympus BX51M reflecting microscope (brightfield-darkfield reflected microscopy). The anthracological approach lies in the observation of the wood anatomical characteristics of the charcoal remains retrieved from the archaeological site, following the three anatomical sections of the wood: transversal, longitudinal tangential, and longitudinal radial. The taxonomic identification of anthracological remains lies on the comparative anatomy method (Chabal 1997), that is the comparison to well-identified modern specimens of reference collections and descriptions available in published wood anatomy atlases (Crivellaro & Schweingruber 2013, Schweingruber 1990). Specimens available in the reference collection created by the author through an extensive sampling of modern botanical material in Cyprus (Rousou 2022, Rousou & Kouka in review) were also used for this purpose. *C. gracilis* modern wood specimens were collected from one locality near Kakopetria (34°58'29"N, 32°54'46"E, 779 m a.s.l.) on the igneous rocks of the Troodos mountains. Photos of the charcoal specimens were taken using a Scanning Electron Microscope Nikon JCM-5000 JEOL NeoScope.

Results

A detailed comparison between the wood anatomy of *C. gracilis* and the archaeological charcoal specimens is available in Tab. 1.

Tab. 1: Comparison of *Cyprinia gracilis* wood anatomy and archaeological charcoals.

Anatomical characteristic	***Cyprinia gracilis* wood anatomy description**	**Description of archaeological charcoals wood anatomy**
Growth ring boundaries	Growth ring boundaries distinct by the difference in vessel between latewood and earlywood and by radially flatted latewood fibers.	Growth ring boundaries distinct.
Ring porosity	Wood ring-porous.	Wood ring-porous.
Vessels	Earlywood vessels predominantly solitary, latewood vessels in short radial multiples common.	Earlywood vessels predominantly solitary, rarely in multiples of 2 vessels, very broad and rounded in transversal section.
	Simple perforation plates.	Simple perforation plates.
	Intervessel pits alternate, small.	Intervessel pits alternate.
	Helical thickenings absent.	Helical thickenings absent.
Axial parenchyma	Axial parenchyma scanty paratracheal and unilateral paratracheal forming semi-circular hoods or caps only on one side of the vessels and which can extend tangentially or obliquely in an aliform or confluent or banded pattern.	Axial parenchyma scanty paratracheal and parenchyma strands of 2-3-4-5 cells.
Rays	Rays exclusively uniseriate.	Rays exclusively uniseriate, short and sometimes more than 10 cellules high.
	Rays with procumbent, square and upright cells throughout the ray.	Rays heterocellylar with square and procumbent inner cells, also often with 1 or 2 rows with marginal upright cells.
Vessel-ray pits	Vessel-ray pits with distinct borders, similar to intervessel pits in size and shape throughout the ray cell.	Not visible.
Fibers	Fibers with distinctly bordered pits (fiber tracheids).	Fibers with distinctly bordered pits (fiber tracheids).

According to the descriptions available in wood anatomy atlases (Crivellaro & Schweingruber 2013, Schweingruber 1990), as well as the description of *C. gracilis* specimens of the reference collection, the species forms a ring-porous wood, with distinct growth ring boundaries. The earlywood vessels are predominantly solitary, while short radial vessel multiples are common in latewood. It is observed that ring porosity is a common trait in climbers and lower wood density is also a common trait in climbers than self-supporting woody plants (Baas & Schweingruber 1987). As observed by Crivellaro & al. (2012) in a study on the anatomy of woody climbers from Cyprus – comprising among others *C. gracilis* specimens – climbers are characterised by greater vessel areas, vessel lumen diameters and higher incidence of ring porosity, related to the elevated need for hydraulic conductance (long-distance water transport) in comparison to self-support species. Other characteristics of the *C. gracilis* wood anatomy are the simple perforation plates, and small, alternate inter-vessel pits. Axial parenchyma is scanty paratracheal and rays are exclusively uniseriate, with procumbent, square and upright cells. Fiber tracheids are present.

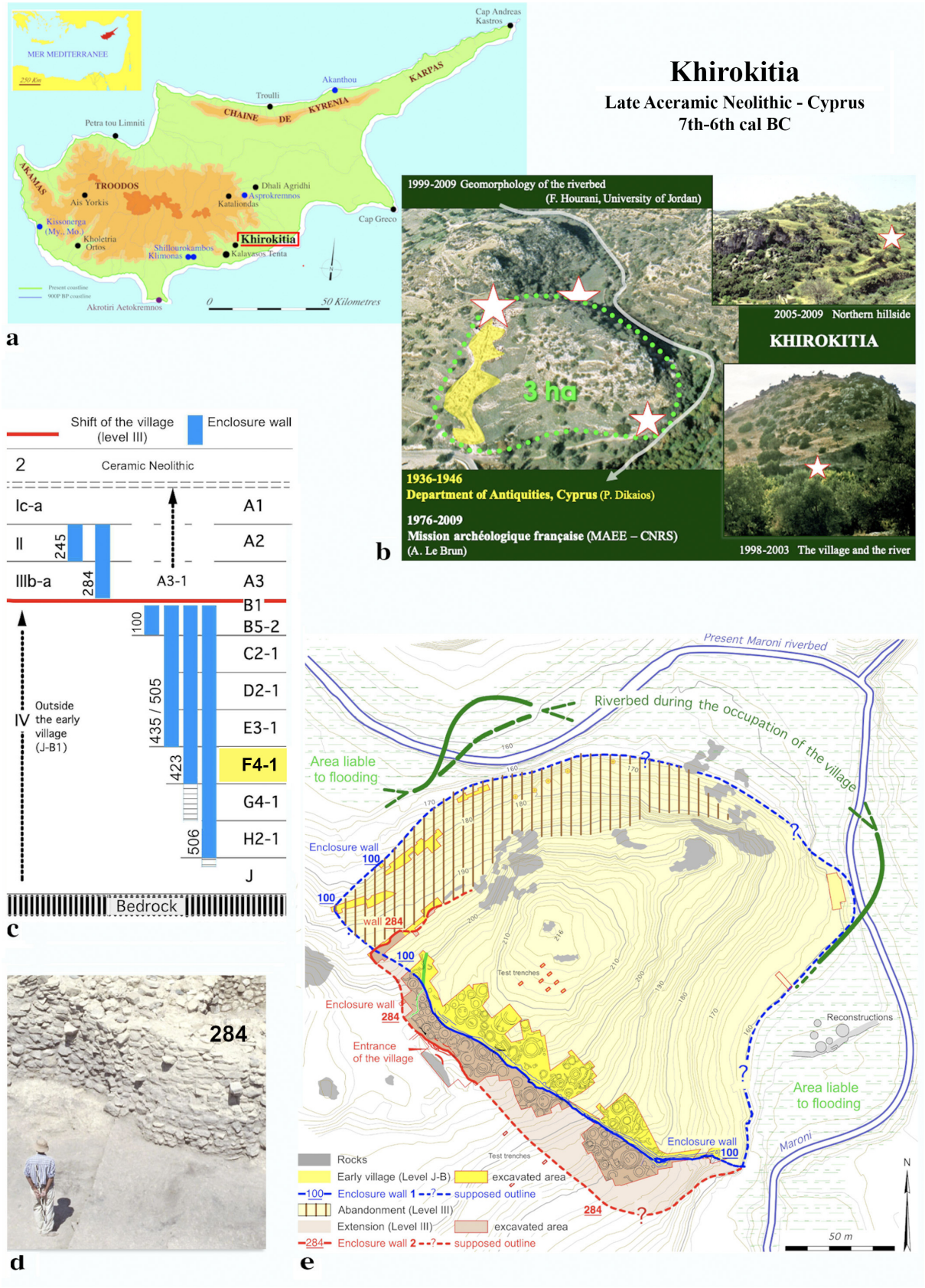

Fig. 1: (a) Map of Cyprus: the Neolithic sites; (b) General view of the site with the location of the excavations; (c) Reference sequence of the settlement; (d) Level III enclosure wall; The level F where the *Cyprinia gracilis* charcoal fragments were recovered is highlighted in yellow; (e) General plan of the settlement showing the shift and the redistribution of the village space, happening in the course of its occupation (Level III) (French Archaeological Mission, MAE-CNRS).

The wood charcoal-remains discussed here (Fig. 2–4) are characterised by ring-porosity, distinct growth ring boundaries, and very broad, solitary earlywood vessels. Perforation plates are simple, while axial parenchyma is scanty paratracheal and organised in parenchyma strands of 2-3-4-5 cells. Rays are exclusively uniseriate, short, and are composed of square and procumbent inner cells and 1 or 2 rows of marginal upright cells. Fiber tracheids are present. Due to these wood anatomical characteristics, these archaeological charcoal specimens are attributed to *C. gracilis*. The other exclusively uniseriate, ring-porous species present in the indigenous flora of Cyprus (*Satureja thymbra, Rubia tenuifolia*, *Pterocephalus multiflorus*, *Hypericum hircinum*) are eliminated by the structure of rays, the presence of helical thickenings in vessels, diffuse axial parenchyma, and septate fibers (Crivellaro & Schweingruber 2013). Homocellular rays are present in *S. thymbra* and *R. tenuifolia*; helical thickenings in vessels are present in *P. multiflorus* and *R. tenuifolia*; diffuse axial parenchyma and septate fibers are present in *H. hircinum* (Crivellaro & Schweingruber 2013).

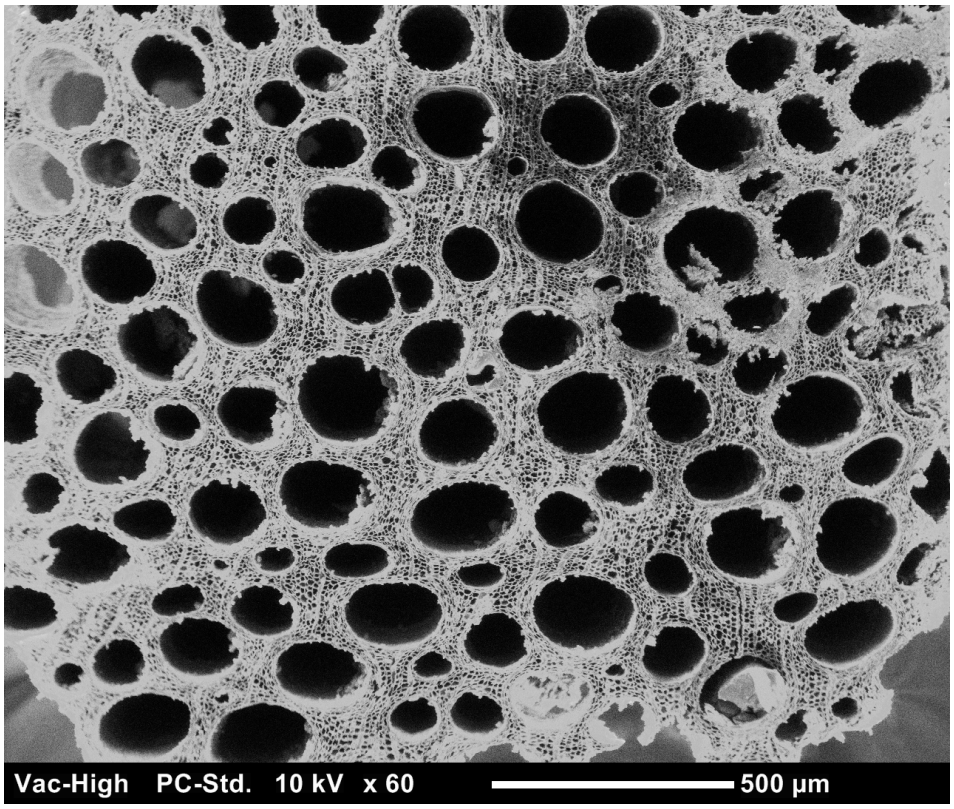

Fig. 2: Transversal section of *Cyprinia gracilis* archaeological charcoal (catalog number 13990). – Maria Rousou.

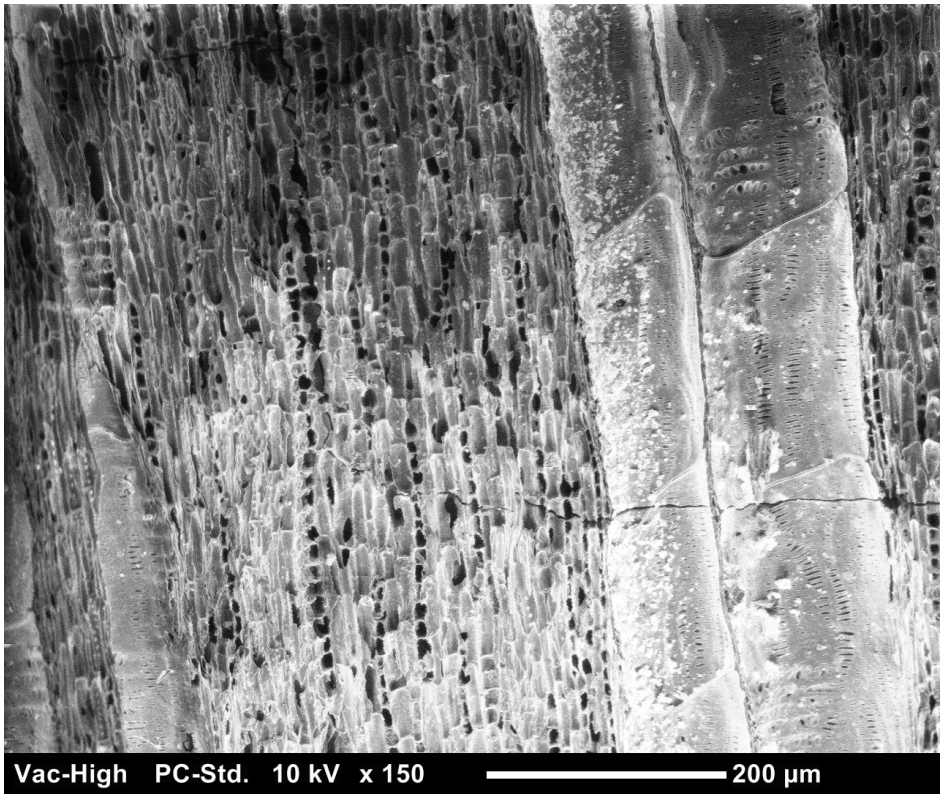

Fig. 3: Longitudinal tangential section of *Cyprinia gracilis* archaeological charcoal (catalog number 13990). – Maria Rousou.

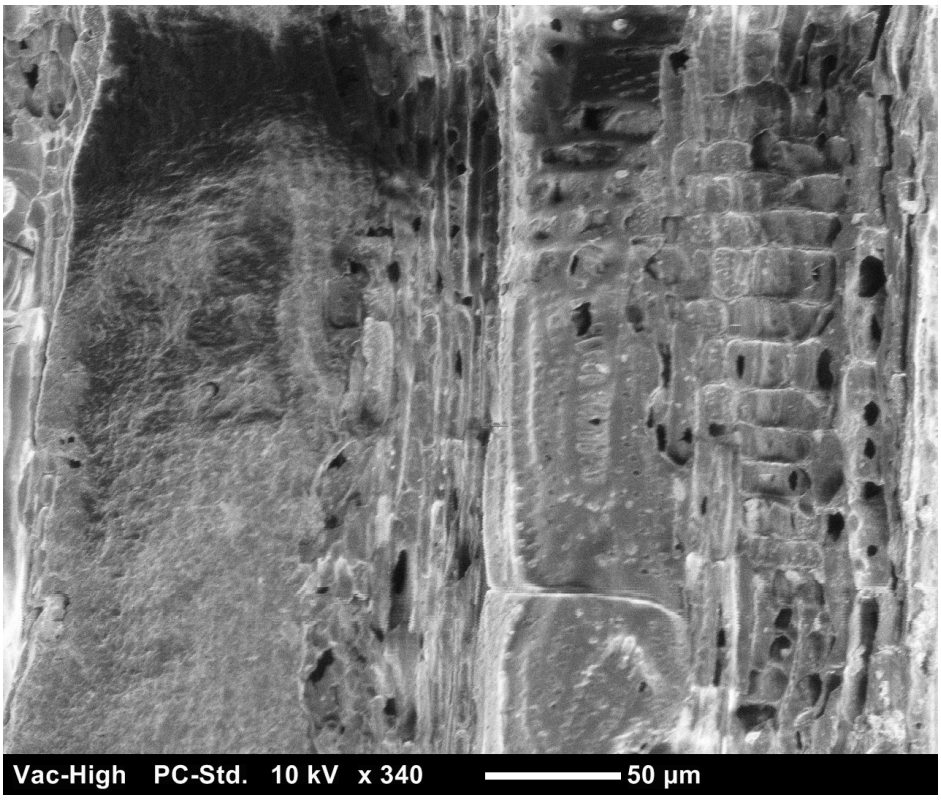

Fig. 4: Longitudinal radial section of *Cyprinia gracilis* archaeological charcoal (catalog number 13990). – Maria Rousou.

Discussion

The good preservation conditions and systematic sampling of soil at the Neolithic site of Khirokitia enabled the discovery of a great number of anthracological remains, exceptional for this early period of Cypriot prehistory, thus allowing the study of the flora of Cyprus and reconstruction of the palaeoenvironment during the occupation of the site (Rousou 2023b). About thirty woody plant taxa are attested by the anthracological approach, including small shrubs (e. g., *Cistaceae*, *Lamiaceae*). The charcoal remains discussed in this study consist the earliest evidence of *C. gracilis* in Cyprus, indicating its presence on the island since the 7th millennium BC. This species is identified for the first time in archaeobotanical context in Cyprus. This study also provides information about the vegetation history and biogeography of this near-endemic species. It also highlights the contribution of the anthracological approach in the study of the biogeography and history of plant species.

The study of anthracological remains of Khirokitia indicates the exploitation of various plant formations, such as maquis (e. g., *Olea europaea*, *Arbutus* sp., *Cistaceae*, *Fabaceae*, *Juniperus* sp., *Lamiaceae*, *Maloideae*, evergreen *Quercus*, *Pistacia* sp.), deciduous oak forests (probably *Quercus infectoria* subsp. *veneris*) and riparian vegetation (e. g., *Styrax officinalis*, *Fraxinus* sp., *Platanus orientalis*, *Tamarix* sp., *Populus/Salix*) (Rousou 2022, 2023b, Thiébault 2003). Considering that the majority of plant taxa attested in the anthracological assemblages of Khirokitia reinforce the hypothesis of the exploitation of Mediterranean maquis formations, the presence of *C. gracilis* could be interpreted in this context. According to the phytosociological observations on the modern flora of Cyprus, this species can be found in more humid locations of the *Oleo-Ceratonion* formations developed on calcareous rocks (Barbero & Quézel 1979, Delipetrou & Christodoulou 2010, Holmboe 1914, Meikle 1985). It is, therefore, possible that this species was also present in more humid locations in the maquis formations – such as near the Maroni river at the foot of the hill of the site –, on the calcareous rocks, that is the principal type of soil attested around Khirokitia. Another plant species that is found in humid locations among the maquis formations on calcareous rocks is *Myrtus communis* (Barbero & Quézel 1979, Delipetrou & Christodoulou 2010, Holmboe 1914), also attested in the assemblages of Khirokitia. The *C. gracilis* charcoals discussed in this study were found together with *Arundo/Phragmites*, *Ficus carica*, *Olea europaea*, *Pinus brutia/halepensis*, *Pistacia* sp., *Platanus orientalis*, deciduous *Quercus*, and *Vitex agnus-castus*, while other taxa identified in level F are *Fabaceae*, *Fraxinus* sp., *Maloideae*, *Myrtus communis*, *Populus/Salix*, *Prunus* sp., evergreen *Quercus*, and *Tamarix* sp. (Rousou 2022).

A second hypothesis for the explanation of the presence of *C. gracilis* at Khirokitia could be the exploitation of plant resources in long-distance localities, that is on the Troodos mountains; however, this is not supported by any clear evidence, e. g. plants restricted on the igneous rocks of the Troodos massif. On the contrary, all available data coming from the study of archaeological and bioarchaeological (archaeobotanical, archaeozoological) material indicates the exploitation of mineral, botanical, and animal resources available in the vicinity of the site (Le Brun 2021, Le Brun & al. 1987,

Parés 2015, Rousou 2022). For these reasons, we consider this second hypothesis less plausible.

C. gracilis presents only a very small number of charcoal remains of the anthracological assemblages of Khirokitia both in terms of frequency (10 out of 14228 studied charcoal fragments, that is 0.07 % of the material) and occurrence (present in only 2 samples). In addition, according to the available ethnobotanical data, there are not any known traditional uses of this species, such as for fruit consumption, hay, medicinal uses, etc (Gennadios 1914). Taking all this into consideration, as well as the fact that *C. gracilis* is a climber, it is highly possible that this species arrived on the site accidentally, together with other woody taxa (for example, *Pistacia* sp., *Quercus* sp., *Olea europaea*) that were collected and used for fuel. According to the spatial analysis of the anthracological remains, the exterior spaces, such as those where *C. gracilis* charcoals were retrieved, were used as discarding areas (Rousou 2022). A similar observation is also made by the analysis of other bioarchaeological and archaeological material (Astruc 2002, Legrand 2007, Parés 2015).

Conclusions and future work

The detailed study of anthracological remains of Khirokitia enabled the discovery of the earliest evidence of *C. gracilis* in the flora of Cyprus, showing that this species, which is nowadays considered a near-endemic, was present in the Cypriot flora since the early Holocene. In addition, this study highlights the importance of the anthracological approach in the study of the biogeography and vegetation history of plant species. These results contribute to the discussion on the palaeoenvironmental reconstruction and the exploitation of plant resources during the Aceramic Neolithic in Cyprus and during the occupation of the site in particular.

The development of anthracological studies on other archaeological sites in Cyprus and in Cilicia could contribute to a better understanding of the biogeography of *C. gracilis* in Cyprus and in the eastern Mediterranean, while a more systematic and in-depth study of the fossil record of palaeobotanical remains in Cyprus could contribute to the study of the evolutionary history of this species. Future work could also include fieldwork for the study, documentation, and geolocation of *C. gracilis* specimens in various localities in Cyprus. Finally, this information could be considered in preservation projects of this near-endemic species.

Acknowledgements

This scientific paper was supported by the Onassis Foundation - Scholarship ID: F ZO 066-1/2018-2019 and within the context of the 7th Scholarship Programme of the Sylvia Ioannou Foundation.

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