

# The identity of *Cintractia carpophila* var. *kenaica*: reclassification of a North American smut on *Carex micropoda* as a distinct species of *Anthracoidea*

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**Abstract:** *Cintractia carpophila* var. *kenaica*, a neglected taxon described from Alaska more than half a century ago, is re-described and illustrated. Its nomenclature and taxonomic status are discussed. This smut species is characterised by small spores with a very finely verruculose surface rarely enclosed by a thin, hyaline, mucilaginous sheath, a wall with 2–5 distinct internal swellings, and parasitism on *Carex micropoda* (*Carex* sect. *Dornera*). It is reallocated to the genus *Anthracoidea* as a distinct species, *Anthracoidea kenaica* comb. nov., and assigned to *Anthracoidea* section *Leiosporae* which includes species having smooth or very finely verruculose spores. Morphological and biological characteristics of the five most similar *Anthracoidea* species are contrasted and discussed.

**Key words:**

*Anthracoidea*  
*Carex*  
*Cintractia*  
Historical collections  
North America  
Smut fungi  
*Ustilaginales*

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## INTRODUCTION

*Anthracoidea* is the most species-rich genus of smut fungi on *Cyperaceae*. Currently, 106 species are accepted in this genus (Denchev & Denchev 2011a, 2011b, 2012, Vánky & Abbasi 2011, Vánky 2012, Savchenko *et al.* 2013), but this is certainly not a final number. The magnitude of host plants reported in different publications for some putative species complexes suggests that more species are likely to exist, some of which may be well-delimited morphological species, while others are probably cryptic species that could be uncovered by molecular methods. Some species of *Anthracoidea* were recently included in molecular systematic studies (Hendrichs *et al.* 2005, Begerow *et al.* 2007, Bauer *et al.* 2007, Lutz *et al.* 2012, Savchenko *et al.* 2013, Vánky *et al.* 2013), but sequence data are not available for the vast majority. Distinct species could still be hidden under different generic names, especially under historical names that have not been reassessed in recent years (Piątek 2012). Such historical names should be critically re-examined in addition to any comprehensive molecular studies directed to the description of novel *Anthracoidea* species.

*Cintractia carpophila* var. *kenaica* is such a neglected taxon name and a likely candidate to be a distinct member of *Anthracoidea*. This smut was described from a specimen of *Carex pyrenaica* subsp. *micropoda* collected in the Kenai Peninsula of Alaska. That sedge is now accepted as a distinct species, *Carex micropoda*, belonging to *Carex* sect. *Dornera* (syn. sect. *Callistachys*) (Murray 2002a). Saville (1952) provided the following description of *Cintractia carpophila*

var. *kenaica*: “Teliosporae 16.0–23.5 × 11.5–19.5 μm, compressae, ellipsoideae, nunquam angulater. Episporium 0.6–1.3 μm, castaneum, leve; saepius interne gibberibus 2–5 munitum.” Zambettakis (1978) included it in *Anthracoidea*, as “*Anthracoidea heterospora* Kukkonen var. *kenaica* (Saville) nov. comb.”, but without any indication of the basionym or a reference to the place of its valid publication, rendering the combination invalid (Art. 41.5). Likewise, Kukkonen (1963) and Piepenbring (2000) considered this fungus to be a member of *Anthracoidea*, but again without further treatment and any formal nomenclatural and taxonomic decisions. Vánky (2012) included this smut in two places in his monograph: first as a synonym of *Anthracoidea heterospora* and later under excluded or invalidly published taxa, in both cases without detailed observations.

The aim of the present work is to clarify the nomenclatural and taxonomic status of *Cintractia carpophila* var. *kenaica*, and to provide a detailed characterization of this smut fungus as it lacks a detailed description and any iconography.

## MATERIALS AND METHODS

Sori and spore characteristics were studied using dried herbarium material deposited in DAOM, S, and WRSL. Specimens were examined either by light microscopy (LM) and scanning electron microscopy (SEM) or only by light microscopy (LM).

For light microscopy (LM), hand-cut sections of sori or small pieces of sori were mounted in lactic acid, heated

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**Table 1.** Spore size range, and mean spore sizes with standard deviation of *Anthracoidea kenaica* specimens examined in this study.

Spore size range (µm)	Average spore size with standard deviation (µm)	Specimen
(14.5–)15.0–20.5(–21.5) × 12.0–17.5(–18.5)	18.1 ± 1.6 × 15.2 ± 1.7	USA, Alaska, Kenai Peninsula, Head of Palmer Creek Valley, 26 July 1951, J.A. Calder 6229 (DAOM 28108 – holotype)
17.0–20.5(–22.0) × 12.0–18.0(–20.5)	19.2 ± 1.3 × 16.1 ± 1.8	Same locality, date and collector (S F-36682 – isotype)
(14.0–)15.0–20.5(–21.0) × (11.5–)12.0–17.5(–18.5)	18.2 ± 1.7 × 15.2 ± 1.8	USA, Alaska, St. Paul, Pribilof Island, 22 Aug. 1914, J.M. Macoun (DAOM 66925)
(14.5–)17.0–20.5(–22.0) × 13.5–18.5(–19.0)	18.5 ± 1.2 × 15.9 ± 1.4	Canada, British Columbia, Bella Coola, Mt. Fougner, 23 Aug. 1956, J.A. Calder, J.A. Parmelee & R.L. Taylor (DAOM 70101)

to boiling point and cooled, then examined under a Nikon Eclipse 80i light microscope. LM micrographs were taken with a Nikon DS-Fi1 camera. Spores were measured using NIS-Elements BR v. 3.0 imaging software. Spore size range, mean spore size, and standard deviation of 50 measured spores of each investigated specimen were calculated (Table 1). The species description includes combined values from all measured specimens. The spores were measured in plane view and measurements were adjusted to the nearest 0.5 µm. Spore size ranges were assigned to one of the three groups distinguished by Savile (1952): (1) small-sized spores, 13–21(–23) × 9–17(–20) µm; (2) medium-sized spores, 15–25(–27) × 10–21 µm; and (3) large-sized spores, 18–33 × 13–28 µm.

For scanning electron microscopy (SEM), spores taken directly from dried specimens were dusted onto carbon tabs and fixed to an aluminium stub with double-sided transparent tape. The stubs were sputter-coated with carbon using a Cressington sputter-coater and viewed under a Hitachi S-4700 scanning electron microscope, with a working distance of ca. 11 mm. SEM micrographs were taken in the Laboratory of Field Emission Scanning Electron Microscopy and Microanalysis at the Institute of Geological Sciences of Jagiellonian University (Kraków).

## RESULTS

Detailed morphological characteristics of the holotype, isotype, and two non-type specimens of *Cintractia carpophila* var. *kenaica* are embraced in the species description and illustrated (Figs 1–2). The internal soral structure in the holotype was typical of species of *Anthracoidea* in that the spores were produced directly on the outer surface of the achene, and not within U-shaped cavities embedded in sterile stroma, a characteristic of *Cintractia* (Kukkonen 1963, Kukkonen & Vaissalo 1964, Piepenbring 2000). This indicated this smut fungus was better placed in *Anthracoidea*, as was suggested in other studies (Kukkonen 1963, Zambettakis 1978, Piepenbring 2000). The spores were uniform in shape and size ranges between collections (Table 1). My examination of specimens of *Cintractia carpophila* var. *kenaica* matched well the short description given by Savile (1952), although the spore surface was not smooth as stated in the protologue, but smooth or very finely punctate in LM, and very finely verruculose in

SEM. The very fine ornamentation of spores was probably outside the limits of resolution of Savile's light microscope. In general, the present examination confirms the decision of Savile (1952) to consider this smut as distinct. However, a specific status seems to be appropriate for this taxon. This is in line with the conclusion of Kukkonen (1963), who, however, did not formally make the transfer. Accordingly, a new combination is necessary.

## TAXONOMY

***Anthracoidea kenaica*** (Savile) M. Piątek, **comb. nov.**  
MycoBank: MB804512

(Figs 1–2)

*Basionym:* *Cintractia carpophila* var. *kenaica* Savile, *Can. J. Bot.* **30**: 419 (1952).

*Synonym:* *Anthracoidea heterospora* var. *kenaica* (Savile) Zambett., *Bull. trimest. Soc. mycol. Fr.* **94**: 177 (1978), nom. inval. (Art. 41.5).

*Type:* **USA:** Alaska: Kenai Peninsula, Head of Palmer Creek Valley, 60°49'N, 149°33'W, on *Carex micropoda* (syn. *Carex pyrenaica* subsp. *micropoda*), 26 July 1951, J.A. Calder 6229 (DAOM 28108 – holotype, S F-36682 – isotype).

*Description:* Sori in all or single ovaries of the inflorescence, black, globose or ovoid, about 1–1.5 mm diam, at first covered by a silvery membrane and perigynium that later ruptures revealing agglutinated spores, powdery on the surface, the sori are partly hidden by the perigynium and scales. Sori develop around reduced achenes that are consecutively surrounded by a thin dark layer of the remnants of achene epidermis, a hyaline layer of sporogeneous hyphae with young spores, a layer of gradually maturing dark spores, and a thin membrane of host origin. Spores usually more or less flattened, chestnut brown, reddish brown to dark brown, quite regular in shape and size, globose, subglobose or broadly ellipsoidal, small, (14.0–)15.0–20.5(–22.0) × (11.5–)12.0–18.5(–20.5) µm [av. ± SD, 18.5 ± 1.5 × 15.6 ± 1.7 µm, *n* = 200/4], rarely enclosed by a thin, hyaline, mucilaginous sheath; wall even, 1.0–1.5 µm thick, somewhat darker than the rest of the spore, without protuberances and light-refractive spots, but with 2–5 distinct internal swellings; surface smooth or very finely punctate in LM, spore profile smooth, surface very finely verruculose in SEM.

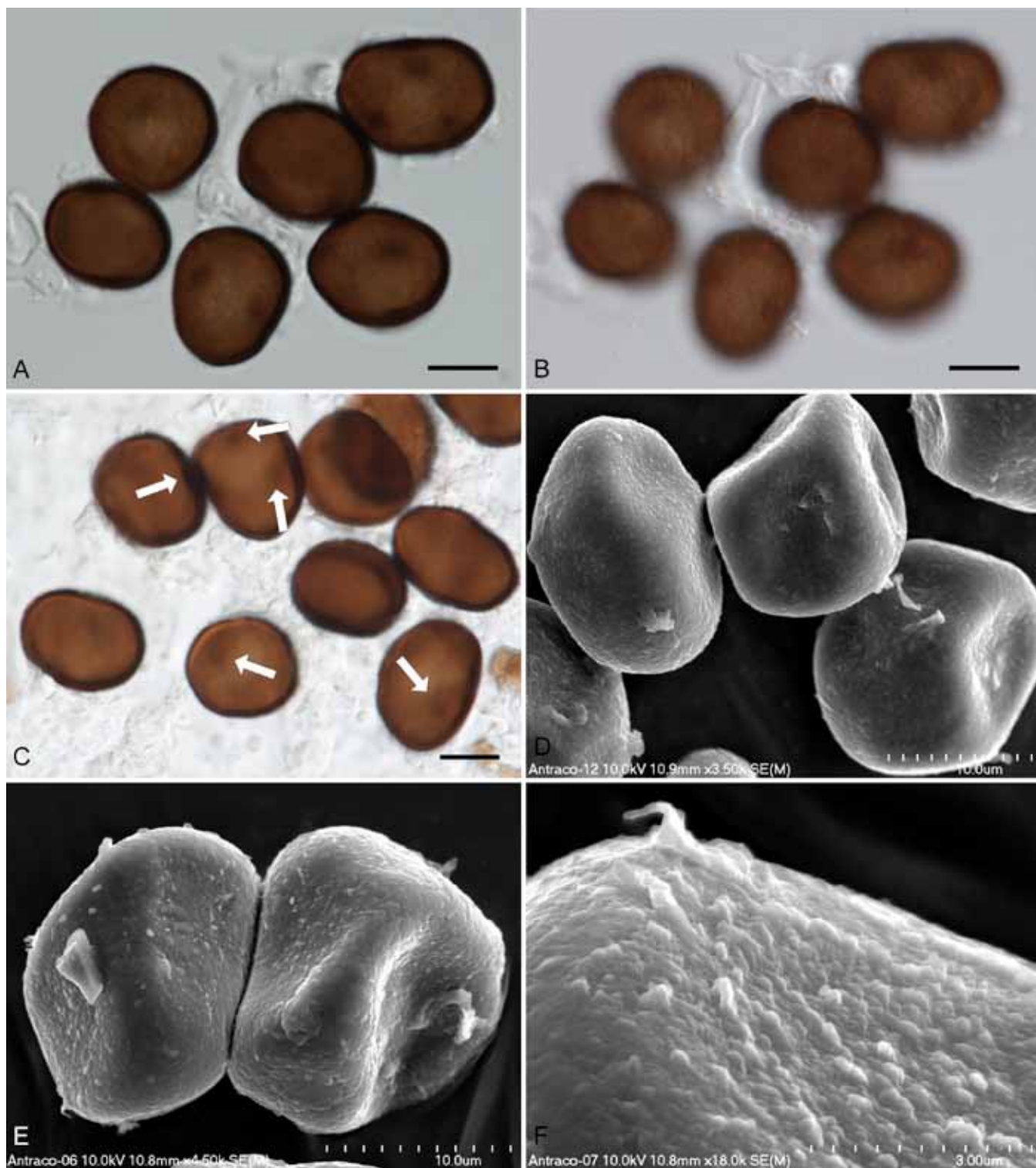


**Fig. 1.** *Anthracoida kenaica* on *Carex micropoda*. **A.** Sori in the ovaries (S F-36682). **B.** Transverse section through the sorus showing reduced achene surrounded by spores (DAOM 28108). **C–D.** Enlarged area close to the achene surface (DAOM 28108). **E.** The spore formation (DAOM 28108). **F.** Cells of the soral membrane, indicated by arrows (DAOM 28108). Abbreviations: n – achene, e – dark layer of the remnants of the achene epidermis, s – layer of young hyaline spores, m – layer of gradually maturing dark spores. Bars: A = 1 mm, B = 500  $\mu$ m, C–D = 20  $\mu$ m, E–F = 10  $\mu$ m.

**Additional specimens examined:** **Canada:** British Columbia: Bella Coola, Mt. Fougner, on *Carex micropoda*, 23 Aug. 1956, J.A. Calder, J.A. Parmelee & R.L. Taylor (DAOM 70101). – **USA:** Alaska: St. Paul, Pribilof Island, on *Carex micropoda*, 22 Aug. 1914, J.M. Macoun (DAOM 66925).

**Host and distribution:** On *Carex micropoda* (*Carex* sect. *Dornera*); Canada (British Columbia) and USA (Alaska).

**Notes:** The nomenclature of *Cintractia carpophila* var. *kenaica* needs some clarification. The name *Uredo carpophila*



**Fig. 2.** *Anthracoidea kenaica* on *Carex micropoda* (S F-36682). **A–C.** Spores seen by LM, median and superficial views, note internal swellings indicated by arrows. **D–E.** Spores seen by SEM. **F.** Spore wall seen by SEM. Bars: A–E = 10  $\mu$ m, F = 3  $\mu$ m.

Schum. (Schumacher 1903) was introduced as superfluous replacement of *Uredo caricis* Pers. (Nannfeldt & Lindeberg 1965) and is therefore illegitimate and to be rejected (Art. 52.1). Consequently, the name *Cintractia carpophila* (Schum.) Liro (Liro 1938), based on *Uredo carpophila*, is also illegitimate. Also, Liro's treatment cannot constitute a valid description of a new species to be attributed to him alone due to the absence of a Latin diagnosis (Nannfeldt & Lindeberg

1965), required in the period 1 January 1935 to 31 December 2011 (Art. 39.1). Vánky (2012) considered *Uredo carpophila* as an illegitimate name, which is correct, but also as a *nomen nudum*, which is not correct, since Schumacher (1903) provided a short description of this species: "U. carpophila, pulvere nigro capsulas subnude ambiente. Ured. Caricis Pers. Synops. pag. 225. In capsulis Caricis caespitosae. Julio". Furthermore, Vánky (2012) considered the name *Cintractia*

*carpophila* var. *kenaiica* also as illegitimate, but the varietal name *Cintractia carpophila* var. *kenaiica* is legitimate since an infraspecific name may be legitimate even if its final epithet was originally placed under an illegitimate species name. Similarly, *Cintractia carpophila* var. *verrucosa* Savile (Savile 1952), was accepted as a legitimate varietal name that was elevated to the species rank as *Anthracoidea verrucosa* (Savile) Nannf. (Nannfeldt 1977, Vánky 2012). Therefore, the name *Cintractia carpophila* var. *kenaiica* is used here to elevate this taxon to species rank.

The type host of *A. kенаica* is *Carex micropoda*, but in addition Savile (1952) assigned a single smut collection on *Carex deweyana* (in *Carex* sect. *Deweyanae*; Naczi 2002) to his concept of *Cintractia carpophila* var. *kenaiica*. Zambettakis (1978) included two SEM pictures of spores from a specimen on *C. deweyana* and they are distinctly verrucose, unlike spores of specimens on *C. micropoda*. Indeed, the *Anthracoidea* on *Carex deweyana* represents a distinct species – *Anthracoidea deweyanae* (Denchev & Denchev 2012). *Anthracoidea kенаica* was previously reported from the type locality on the Kenai Peninsula and two other collections from the same region (Savile 1952). Here the smut is newly reported from Pribilof Island (AK) and from Mt Fougner in British Columbia; this last collection represents the first record of this species in Canada.

## DISCUSSION

*Cintractia carpophila* var. *kenaiica* cannot be treated as a synonym or variety of *Anthracoidea heterospora* since this species is different in having spores with better developed ornamentation, thicker walls (1–2.5 µm), and occurs on host plants belonging to *Carex* sect. *Phacocystis* (Vánky 2012). It can be assigned to *Anthracoidea* sect. *Angulosporae* (Kukkonen 1963). In contrast, the morphology of *Anthracoidea kенаica* is characteristic of members of *Anthracoidea* sect. *Leiosporae*, which includes species with smooth or very finely verruculose spores (Kukkonen 1963). Within this section, *Anthracoidea kенаica* may be comparable only to five *Anthracoidea* species having spores similar in size and with a smooth or very finely verruculose (not papillate) surface: *A. elyanae*, *A. externa*, *A. macranthae*, *A. nardinae*, and *A. scirpi*. The main morphological differences between these species include differences in wall thickness, the presence and the number of internal swellings, and the presence of a hyaline mucilaginous sheath enclosing the spores. Furthermore, all of them are restricted to host species belonging to different sections of *Carex*, or to different genera (*Kobresia*, *Trichophorum*), which could be used as supportive taxonomic characters. Characters used to discriminate these five species of *Anthracoidea* from *A. kенаica* (Table 2) are contrasted and discussed below.

*Anthracoidea elyanae* is distinguished from *A. kенаica* by the mostly smooth spores with a thicker wall, fewer internal swellings, a more or less evident mucilaginous sheath, and occurrence on *Kobresia*. The internal swellings in *A. elyanae* are weakly visible in LM (Savile 1952, Kukkonen 1961, Vánky 1994, 2012), and recent TEM studies of spores from a Romanian collection did not report internal swellings (Parvu

*et al.* 2009). In fact, the spore presented in figure 3 of the latter study has a shallow thickening on the lower flattened side, which may be interpreted as a weak internal swelling. By contrast, internal swellings of *Anthracoidea kенаica* are prominent and clearly visible in LM. *Anthracoidea externa* is morphologically distinct in having absolutely smooth spores surrounded by a thick mucilaginous sheath, a thicker spore wall without internal swellings, and occurrence on species of *Carex* sect. *Filifoliae* (Mastrogioseppe 2002). *Anthracoidea macranthae* differs from *A. kенаica* as it has completely smooth spores with prominent and common hyaline caps (a mucilaginous sheath) on the flattened sides, an absence of internal swellings, a somewhat thinner spore wall, and in occurring on *Kobresia* (Guo & Wang 2005). Although not discussed in the protologue, the occurrence of a mucilaginous sheath in the form of hyaline caps is the most valuable diagnostic character of *Anthracoidea macranthae*. The combination of characters seen places this species close to *A. externa*.

*Anthracoidea nardinae* appears to be most similar to *A. kенаica*. It is distinguished by nearly smooth spores, a thicker spore wall, fewer internal swellings, and occurrence on *Carex* sect. *Nardinae* (Murray 2002b). The type host of *A. nardinae* is *C. nardina*, but Kukkonen (1963) assigned this smut to two collections on *C. elynoides*, which belongs to a different *Carex* section (sect. *Filifoliae*). The examination of one collection of *Anthracoidea* on *C. elynoides* [“Plants of Southern Colorado, *Carex elynoides* Holm n. sp., near Pagosa Peak, Aug. 1899, leg. C.F. Baker”, WRSL s.n., extracted from the isotype of *C. elynoides* in WRSL (phanerogamic herbarium)], had globose, subglobose to broadly ellipsoidal spores, (14.5–)15.0–19.5 × (11.0–) 12.0–17.5 µm, av. ± SD, 17.2 ± 1.2 × 14.8 ± 1.7 µm, *n* = 50, wall even, 1.0–1.5 µm, without internal swellings, surface smooth without mucilaginous sheath], though different from those studied by Kukkonen, revealed a complete absence of internal swellings typical of *A. nardinae*. It could be yet another distinct species or a form of *A. externa* without a hyaline sheath.

*Anthracoidea scirpi* is distinguished from *A. kенаica* by the minutely punctate spores, which are usually surrounded by hyaline, mucilaginous sheaths on the flattened sides, a thicker spore wall, the absence of internal swellings, and occurrence on *Trichophorum* species (Vánky 1994, 2012).

Differences between smut specimens in the ovaries of *Carex micropoda* and all aforementioned *Anthracoidea* species from sect. *Leiosporae* support *A. kенаica* as a distinct species specialised to a host in *Carex* sect. *Dornera*. Except for the host of *A. scirpi*, host plants of these *Anthracoidea* species are placed in one of the four/five major clades of the tribe *Cariceae*, the so called “Core Unispicate Clade”, which includes *Carex* subgen. *Psyllophora* p.p., *Kobresia*, and *Uncinia* (Starr & Ford 2009). Whether this may indicate a close evolutionary relation between members of *Anthracoidea* sect. *Leiosporae* is uncertain, and the problem remains open for future studies using molecular methods. In a recent molecular phylogenetic study, by Hendrichs *et al.* (2005), the only accessioned smooth-spored species, *A. elyanae*, was recovered as sister to the verruculose-spored species *A. curvulae* on *Carex curvula*, which is also a member of the “Core Unispicate Clade”.

Table 2. Host plants and morphological characters of *Anthracoidea kenaica* and the five most similar species of the section *Leiosporae*.

Species	Host plant(s)	Spores ( $\mu\text{m}$ )	Wall ( $\mu\text{m}$ )	Internal swellings	Spore ornamentation	Mucilaginous sheath	References
<i>Anthracoidea elymae</i>	<sup>1</sup> <i>Kobresia humilis</i> , <i>K. laxa</i> , <i>K. macrolepis</i> , <i>K. myosuroides</i> , <i>K. schoenoides</i> , <i>K. sibirica</i> , <i>K. smirnovii</i>	(14–)16–22(–25) × (9–)10–20(–22)	1–2.5(–3)	frequent 1–2(–3) weak internal swellings	smooth, seldom dotted	usually covered by a gelatinous sheath	Kukkonen (1963)
<i>Anthracoidea externa</i>	<i>Carex filifolia</i> ( <i>Carex</i> sect. <i>Filifoliae</i> )	17–22(–25) × (14–)15–18.5	1–2.5(–3)	often with 1–3 weak internal swellings	smooth to finely punctate on the flat sides	more or less evident hyaline sheath	Ványkó (1994, 2012)
<i>Anthracoidea kenaica</i>	<i>Carex filifolia</i> ( <i>Carex</i> sect. <i>Filifoliae</i> )	15–22(–23) × 11–20	0.7–2.5	absent	absolutely smooth	always covered by a gelatinous sheath	Kukkonen (1963)
<i>Anthracoidea kenaica</i>	<i>Carex micropoda</i> ( <i>Carex</i> sect. <i>Dornera</i> )	17–21 × 15–20 (14–)15–20(–22) × (11.5–)12–18.5(–20.5)	0.8–1.5 1–1.5	absent 2–5 distinct internal swellings	apparently smooth smooth to very finely punctate in LM, verruculose in SEM	present in most of the spores rarely enclosed by a thin mucilaginous sheath	Ványkó (2012) this study
<i>Anthracoidea macranthae</i>	<i>Kobresia macrantha</i>	15–18(–19.5) × 13–17.5	0.5–1	absent	smooth	hyaline caps common on the flattened side	Guo & Wang (2005)
<i>Anthracoidea nardinae</i>	<i>Carex nardina</i> ( <i>Carex</i> sect. <i>Nardinae</i> ), ? <i>Carex elynoides</i> ( <i>Carex</i> sect. <i>Filifoliae</i> )	15–18.5(–20.5) × 13.5–16	1	absent	smooth	present on the flattened sides and often around the entire spore	Ványkó (2012)
<i>Anthracoidea nardinae</i>	<i>Carex nardina</i> ( <i>Carex</i> sect. <i>Nardinae</i> ), ? <i>Carex elynoides</i> ( <i>Carex</i> sect. <i>Filifoliae</i> )	(15–)16–22(–23) × (10–)11–20(–21)	1–3	always 1–3 more or less clear internal swellings	smooth, sometimes obscurely dotted	absent, or at most, rare	Kukkonen (1963)
<i>Anthracoidea scirpi</i>	<i>Trichophorum cespitosum</i> , <i>T. pumilum</i>	(15–)16–23 × 13–20(–21)	ca. 2	1–3 clearly seen	almost smooth, only dotted by hardly discernible dots	no data	Nannfeldt (1979)
<i>Anthracoidea scirpi</i>	<i>Trichophorum cespitosum</i> , <i>T. pumilum</i>	(15–)16–22(–23) × 13–20(–21)	1.5–2	1–3 more or less clear internal swellings	almost smooth or sometimes obscurely punctate	no data	Ványkó (1994, 2012)
<i>Anthracoidea scirpi</i>	<i>Trichophorum cespitosum</i> , <i>T. pumilum</i>	(16–)17–24(–25) × 12–20(–23)	1–2.5(–3)	absent	smooth or rarely very slightly verruculose	often covered by a gelatinous sheath	Kukkonen (1963)
<i>Anthracoidea scirpi</i>	<i>Trichophorum cespitosum</i> , <i>T. pumilum</i>	17–23 × 14–21	1.5–2	absent	smooth or very minutely punctate	often covered by gelatinous sheaths on the flattened sides	Ványkó (1994, 2012)

<sup>1</sup> Host plants taken from Ványkó (2012), but at least some of them may harbour different *Anthracoidea* species.

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