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Kuehneola warburgiana comb. nov. (*Phragmidiaceae, Pucciniales*), causing witches' brooms on *Rosa bracteata*

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Abstract—Caeomatoid rust infection has been observed on *Rosa bracteata* plants at a single site in Ishigaki Island, Okinawa, Japan, since 1995. The fungus (not previously known in Japan) was identified as *Caeoma warburgiana* by its characteristic systemic infection causing witches' brooms and its spore morphology. Uredinial and telial sori were found on the leaves of the witches' brooms of the infected rose plants at the same site in 2009. The urediniospores were pedicellate and echinulate. The teliospores were composed of two to four linearly arranged, thin-walled cells on a short pedicel. *Caeoma*-type aecia, *Uredo*-type uredinia and pedicellate teliospores with two to four linearly arranged cells are characteristic of the genus *Kuehneola*. Identical telia and teliospores were found in the lectotype of *C. warburgiana*. *Caeoma warburgiana* is recombined as *Kuehneola warburgiana*.

Key words —Asia, life cycle, nomenclature, taxonomy

Introduction

Rosa bracteata is an evergreen perennial shrub, native of southern regions of China, growing in mixed forests, scrub, and sandy hills at low altitudes and seashores (Wu & Raven 2003). The plants also occur at coastal areas of Taiwan and adjacent islands of south Japan (Satake et al. 1989, Liu et al. 2000, Wu & Raven 2003). Although this rose, called "McCartney rose," has been introduced to the United States for breeding new cultivars like "Mermaid" (Beales et al. 1998), it has become a noxious weed in southern States (Texas Invasive Plant and Pest Council 2011).

In the native regions of this rose species, *Caeoma* "*rosae-bracteatae*", *C. warburgiana*, and *Kuehneola japonica* have been reported to occur (Anonymous 1979; Hiratsuka & Chen 1991; Sawada 1943; Zhuang 1983); however, only the last fungus has previously been known in Japan (Hiratsuka et al. 1992, Kobayashi 2007). In December 1995, a caeomatoid rust infection was found on *R. bracteata* plants at a single site at Hirakubo Peninsula, Ishigaki Island, Okinawa. The caeomatoid fungus causing witches' brooms on roses was

previously not known in Japan. Through continued field observations, uredinial and telial sori were found on the leaves of the witches' brooms of the infected rose at the same site in February 2009.

This paper describes the morphology of the whole life cycle of the *R. bracteata* rust fungus and discusses its taxonomic relationships to *C.* "*rosae-bracteatae*."

Materials & methods

Small sorus-bearing pieces were cut out from the herbarium specimens and thin-sectioned with a razor blade under a binocular dissecting microscope. Spores and paraphyses were scraped from sori. Thin-sections, scraped spores, and paraphyses were mounted on microscopic slides and treated by the method described by Ono (2000). The slide preparations were then examined both by bright-field and differential interference contrast microscopy (DIC) with an Olympus BH2 microscope (Olympus, Tokyo, Japan), and measurements were made with an ocular micrometer. Fifty or twenty randomly selected spores and paraphyses were measured for each specimen and five or ten spermogonia were measured. For observation of urediniospore germ pores, spores were mounted in lactic acid on glass slides and heated to boiling point for a few seconds, after which a drop of lactophenol solution with aniline blue was added onto the boiled spores.

For scanning electron microscopy (SEM), spores scraped from herbarium specimens were placed on double-sided adhesive tape on a specimen holder and coated with platinum-palladium at 25 nm thicknesses under a Hitachi E-1030 ion sputter (Hitachi, Tokyo, Japan). Samples were observed with a Hitachi S-4200 SEM operating at 15 kV.

Taxonomy

Kuehneola warburgiana (Henn.) Y. Ono **comb. nov.** Figs. 1–3

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≡ *Caeoma warburgiana* Henn., in Warburg, Monsunia 1: 4, 1899 ["1900"].

Lectotype (**designated here**): on *Rosa* sp., China, Zhejian, Ningbo, 0 + I + III, Dec 1887, O. Warburg (B).

Caeoma "*rosae-bracteatae*" Sawada, Rep Dept Agr Gov Res Inst Formosa 86:131, 1943, nom. inval.

Aecial infection systemic, causing witches' brooms on shoots spreading a large area. Spermogonia on juvenile shoot tips, leaf stipules and petioles (but not on leaf body), subepidermal, of determinate growth, dome-shaped or broadly convex with flat hymenium, 139–206 µm wide, and 58–104 µm high. Aecia strictly on shoots, subepidermal in origin becoming erumpent, of indeterminate growth, neither peridiate nor paraphysate, bright orange-colored, and powdery. Aeciospores formed in basipetal succession, but separated soon after maturation, thus not forming chains; ellipsoid to broadly ellipsoid, often angular, and 20–29 × 12–18 µm in size; wall 1–2 µm thick at sides, apically thickened, colorless, and verrucose with no apparent germ pore.

Figure 1 *Kuehneola warburgiana* (IBAR-7744). A. Witches' brooms exhibited by *Rosa bracteata* shoots with spermogonial/aecial infection. B. Early stage of witches' broom development. Leaf petioles and stipules are densely covered with orange-colored spermogonia. C. Mature caeomatoid aecia on shoots with powdery orange-colored spores. D. Vertical section of a subepidermal spermogonium with a flat hymenium. E. Vertical section of caeomatoid aecium. Aeciospores are formed in basipetal succession. F. Aeciospores. G. Surface structure of an aeciospore. Bars: D = 50 μm; E, F = 10 μm; G = 5μm

Uredinia on abaxial leaf surface, scattered or in loose groups, bright orange-colored and powdery, subepidermal in origin and erumpent, peripherally paraphysate, and often mixed with teliospores; paraphyses irregularly cylindrical, weakly to strongly incurved, apically and dorsally thick-walled, colorless, 25–30 μm high, and 6–9 μm wide. Urediniospores formed singly on a short pedicel, ellipsoid, broadly ellipsoid, obovoid or pyriform, and 18–25 × 11–16 μm in size; wall thin, colorless, and echinulate with no apparent germ pore.

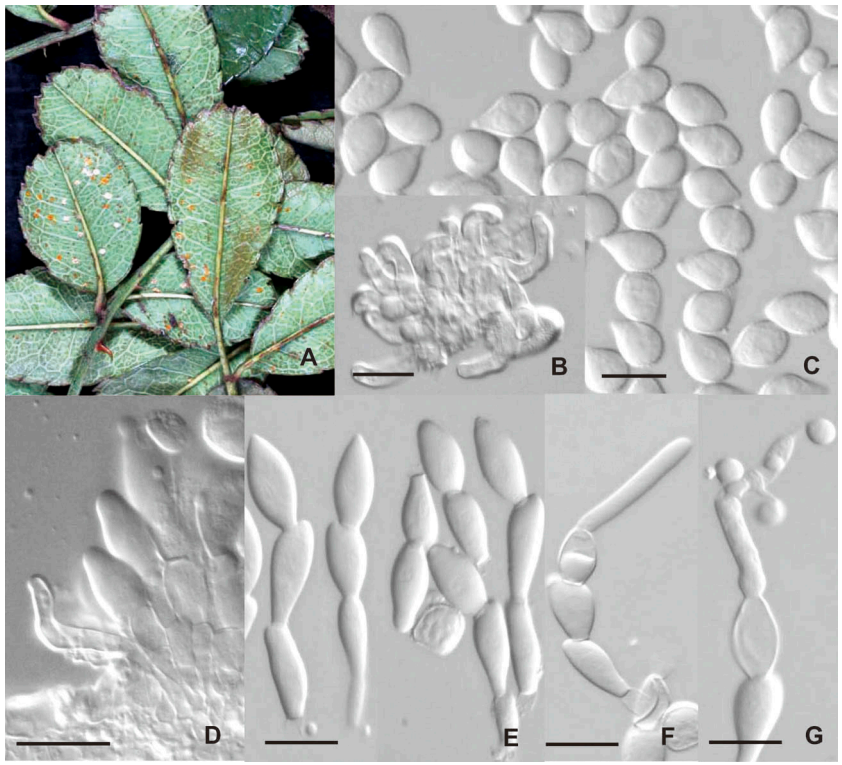

Figure 2 *Kuehneola warburgiana* (IBAR-10130). A. Orange-colored, powdery uredinia and white cottony telia formed on the abaxial leaf surface of *Rosa bracteata*. B. Incurved paraphyses. C. Urediniospores. D. Vertical section of a telium. The sorus is surrounded by incurved paraphyses. The wall is dorsally and apically thickened. E. Two- or three-celled teliospores with a short pedicel (fragment remained at the base). F. Metabasidium development from a distal end cell. G. Basidiospores formed from a four-celled metabasidium (one spore is missing). Bars = 20 µm

Telia on abaxial leaf surface, white, cottony, peripherally paraphysate, and often intermixed with uredinia. Teliospores composed of 2–4 linearly arranged cells formed on a short pedicel; each telial cell ovoid, ellipsoid or broadly ellipsoid, and 21–34 × 10–17 µm in size; wall thin and colorless; germinating in situ to form four basidiospores on a four-celled metabasidium.

Additional specimens examined: On ***Rosa* sp.**: **China**, **Fujian**, Futschau, Yuenfuthal, 0 + I, Dec. 1887, O. Warburg (B).

On ***Rosa bracteata*** J.C. Wendl.: **Japan**, **Okinawa**, Ishigaki, Hirakubo Peninsula, 0 + I, 10 Dec 1995, Y. Ono (IBAR-7744); 0 + I, 7 Nov 1997, Y. Ono (IBAR-7961); 0 + I, 10 Nov 2002, Y. Ono (IBAR-8888); 0 + I + II + III, 17 Feb 2009, Y. Ono (IBAR-10130)

Ecology—The rust infected *R. bracteata* plants spread in the steep slope at the peninsula in Ishigaki. The habitat is heavily utilized for grazing and the

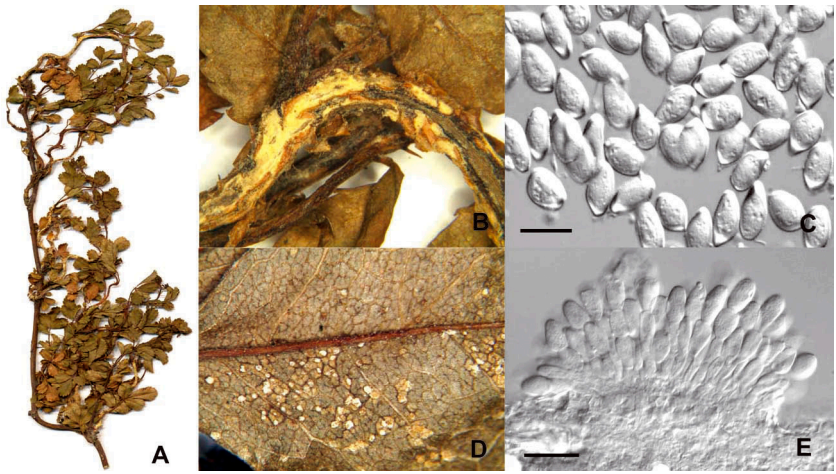

Figure 3 *Caeoma warburgiana* (Lectotype, deposited in B). A. Witches' brooms exhibited by unidentified rose shoots with spermogonial/aecial infection. B. Mature caeomatoid aecia with powdery spores. C. Aeciospores. D. White cottony telia formed on the abaxial leaf surface. E. Vertical section of a telium. Bars = 20 μm

prickly bushes are the property by which the rose is selected for in the habitat, otherwise it is not commonly observed in the island covered with subtropical evergreen forests.

Comments—*Kuehneola japonica* (Dietel) Dietel is the only other *Kuehneola* species to occur on *Rosa* spp. This fungus is microcyclic and forms only telia on 15 species and 5 varieties of *Rosa,* being widely distributed in east and southeast Asia (Anonymous 1979; Arthur & Cummins 1936; Hiratsuka & Chen 1991; Hiratsuka et al. 1992; Kobayashi 2007; Sawada 1918, 1944; Teodoro 1937). This species forms pulvinate bright orange-colored telia, not associated with spermogonia, on both surfaces of leaves, petioles and shoots (Hiratsuka et al. 1992). The sori often become confluent, particularly along the leaf veins and on shoots; however, the telial infection is not systemic. Teliospores are 2–4-celled with a short, persistent pedicel. Each teliospore cell is 22–38 × 15–22 μm in size and thin-walled.

Phylogenetic relationships between *K. warburgiana* and *K. japonica* are not apparent. The non-systemic nature of telial infection of *K. japonica* suggests that the "correlated species" concept (Arthur 1934) and its evolutionary interpretation, Tranzschel's Law (Shattock & Preece 2000; Cummins & Hiratsuka 2003), may not apply to a possible relationship between the two fungi, i.e., a direct evolutionary derivation of microcyclic *K. japonica* from macrocyclic *K. warburgiana*.

In his protologue of *C. warburgiana,* Hennings (1899) cited two collections — "China, Fitschau und Ningpo: auf Zweigen von *Rosa* sp. Dez. 1887. (O. Warburg)." These collections are syntypes, because the protologue did not designate either as the holotype. In B, the collection from Ningpo (Zhejiang) is labeled as "holotypus" and the collection from Fitschau (Fujian) as "isotypus." Yet Hein (1989) listed the Fitschau collection as the "holotypus" and did not mention the other collection. In the Ningpo collection, the telia and teliospores are present (Figs. 3 D,E) and morphologically the same as those found on the Ishigaki collection (IBAR-10130). Therefore, the Ningpo collection is designated here as the lectotype (although it is possible that this collection may already have been designated as lectotype, by a corrective interpretation of the misapplied term "holotypus" on the label of the B herbarium specimen).

Caeoma warburgiana has been reported on *R. bracteata* in Fujian (Zhuang 1983), *R. banksiae* W. T. Aiton var. *banksiae* in Yunnan (Tai 1979), *R. banksiae* var. *normalis* Regel in Yunnan (Tai 1979), *R. hugonis* Hemsl. in Fujian (Zhuang 1983), and *R. longicuspis* Bertol. in Yunnan (Tai 1979).

Caeoma "*rosae-bracteatae*" Sawada was published (without Latin diagnosis) for a fungus on *R. bracteata* var. *bracteata* (as *R. bracteata* "var. *typica*" Lindl.) (Sawada 1943). Four specimens were cited, all from Hsinchu, Taiwan. The sori were described as elongated up to 70 mm and becoming naked with orange-yellow spores; the spores were in chains of 3–4, ellipsoid, globoid or obovoid-ellipsoid, and 22–36 × 11–16 µm in size; the wall was densely verrucose, 2 µm thick and up to 7–8 µm apically. Although none of Sawada's collections were available for the study, all the details described for the fungus support its biological identity with *C. warburgiana*.

Spermogonia of *Kuehneola* species are characterized by their subcuticular origin with a flat hymenium and indeterminate growth (Type 11 of Cummins & Hiratsuka 2003). However, intraepidermally formed spermogonia of indeterminate growth was reported for *K. loeseneriana* (Henn.) H.S. Jacks. & Holw. (Hernández & Hennen 2003). *Kuehneola warburgiana* forms Type 6 spermogonia (subepidermal with a flat hymenium of determinate growth), a type previously known only in *Gymnoconia* (Hiratsuka & Hiratsuka 1980; Cummins & Hiratsuka 2003).

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