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Are Callirhytis erythrostoma (Dettmer, 1933) and C. erythrosoma (Dettmer, 1933) synonyms of Callirhytis erythrocephala (Giraud, 1859) or different species? (Hymenoptera: Cynipidae: Cynipini)

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Abstract

A new heteroecious life-cycle of an oak cynipid gallwasp *Callirhytis erythrocephala* (Giraud, 1859) (Hymenoptera, Cynipidae: Cynipini) has been closed experimentally. *Callirhytis hartigi* Förster, 1869 is a **syn. nov.** of *C. erythrocephala*. The sexual galls induced by *C. erythrocephala* are described for the first time. The sexual female and male of *C. erythrocephala* are re-described, and morphological characters of adults for the differentiation of the sexual form from other sexual forms of *Callirhytis* species are also given. It is questionable whether *Callirhytis erythrosoma* is synonymous with *C. erythrocephala* or represents a distinct species.

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KEY WORDS: Cynipidae, *Callirhytis*, *C. erythrocephala*, *C. erythrosoma*, *C. hartigi*, biological cycle, taxonomy.

Resum

Són *Callirhytis erythrostoma* (Dettmer, 1933) i *C. erythrosoma* (Dettmer, 1933) sinònims de *Callirhytis erythrocephala* (Giraud, 1859) o són espècies diferents? (Hymenoptera: Cynipidae: Cynipini).

Un nou cicle heteroècic de cinípid ha estat tancat experimentalment: *Callirhytis erythrocephala* (Giraud, 1859) (Hymenoptera, Cynipidae: Cynipini). *Callirhytis hartigi* Förster, 1869 és una **syn. nov.** de *C. erythrocephala*. Es descriu per primera vegada la gala de la forma sexuada de *C. erythrocephala*. Es redescriuen les femelles sexuades i els mascles de *C. erythrocephala*, i es destaquen les característiques que permeten diferenciar aquesta forma sexuada d'altres ja conegudes en el gènere *Callirhytis*. Es

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discuteix la possibilitat de què *Callirhytis erythrosoma* i *C. erythrocephala* siguin espècies diferents en lloc d'espècies sinònimes.

KEY WORDS: Cynipidae, Callirhytis, C. erythrocephala, C. erythrosoma, C. hartigi, cicle biològic, taxonomia.

Resumen

Son *Callirhytis erythrostoma* (Dettmer, 1933) y *C. erythrosoma* (Dettmer, 1933) sinónimos de *Callirhytis erythrocephala* (Giraud, 1859) o son especies distintas? (Hymenoptera: Cynipidae: Cynipini).

Un nuevo ciclo heteroécico de cinípido ha sido cerrado experimentalmente: *Callirhytis erythrocephala* (Giraud, 1859) (Hymenoptera, Cynipidae: Cynipini). *Callirhytis hartigi* Förster, 1869 es una **syn. nov.** de *C. erythrocephala*. Se describe por primera vez la agalla de la forma sexuada de *C. erythrocephala*. Se redescriben las hembras sexuades y los machos de *C. erythrocephala*, y se destacan las características que permiten diferenciar esta forma sexuada de otras ya conocidas en el género *Callirhytis*. Se discute la posibilidad de que *Callirhytis erythrosoma* y *C. erythrocephala* sean especies diferentes en lugar de especies sinónimas.

KEY WORDS: Cynipidae, *Callirhytis*, *C. erythrocephala*, *C. erythrosoma*, *C. hartigi*, ciclo biológico, taxonomía.

Introduction

It is not easy to close experimentally a cynipid gall-wasp species life-cycle and it very often depends on a correct host plant or plant organ being chosen and given to the gall-wasp (Pujade-Villar *et al.*, 2001). Moreover, even intraspecific preference can strongly influence the result of galling (Stone *et al.*, 2002). For these reasons, pairing possible asexual and sexual generations and

closing a gall-wasp life-cycle is quite remarkable in entomological research. Checking a known cycle is easier because the variables mentioned above are controlled.

In Europe the genus Callirhytis was represented by 5 sexual and 4 asexual forms. Francois Barbotin (1914-1996) was the first researcher to experimentally close the lifecycles of Callirhytis rufescens (Mayr, 1882) and C. glandium (Giraud, 1859). Later, Nieves-Aldrey (1992) published Barbotin's results on his own, and established the alternation of generations for two species: C. rufescens sexual form (with asexual C. glandulosa) and C. glandium asexual form (with sexual C. aestivalis). Sexual forms of these species emerge from galls in the twigs of oaks of Ouercus section Ouercus, while asexual females develop in «stone» galls in acorns of the Cerris section. For this reason, Nieves-Aldrey (1992), after communicating with Barbotin (FB), proposed that the alternate sexual generation of C. erythrocephala, which was known from producing galls in acorns of the Cerris section, could develop in young twigs of the Quercus. The only European Callirhytis species that was known only from the sexual generation and had the tarsal claw simple, without a basal lobe, was Callirhytis hartigi Förster, 1869, and Nieves-Aldrey (1992) proposed C. hartigi as a possible sexual generation of C. ervthrocephala, although the gall of C. hartigi was unknown and he treated C. hartigi in his review of Callirhytis as a separate species. This pairing has been confirmed experimentally by FB and Folliot (RF) independently (see below).

François Barbotin's family donated his collection to Juli Pujade-Villar (JP-V) (Pujade-Villar & Folliot, 2001) and the experimental material from *Callirhytis* species was found and examined. François Barbotin closed the above-mentioned life-cycles in several experiments during 1971-1983 but never published

them, although JP-V has pressed him to since 1984. Barbotin hardly worked on life-cycles of cynipids until 1989-1990. He informed J.L. Nieves-Aldrev and one of the authors of this paper (JP-V) in 1984 about his results on the Iberian species and the preliminary manuscript that he was writing; in 1987, Barbotin informed to JP-V that the paper was close to being finished. Unfortunately, he suffered from Alzheimer's disease from 1991, dying on the 19th of August 1996 (Pujade-Villar & Folliot 2001) and leaving a lot of his research unpublished. Nieves-Aldrey (1992), in his revision of the European Callirhytis species, published Barbotin's life-cycle results; he listed 4 Callirhytis species for Europe but, however, did not mention two other known species from Azerbaijan: C. comantis Belizin & Maisuradze, 1961 and C. reticulatus Belizin & Maisuradze, 1961 (Maisuradze, 1961) (later, Melika (GM) found the second species in material collected around Sochi, Georgia and in the Russian part of the North Caucasus, unpublished data). In the same study, Nieves-Aldrey did not designate the lectotype of C. hartigi after studying the typical series, and he listed Callirhytis villarrubiae Tavares, 1930 as a synonymy of C. rufescens from JP-V's unpublished PhD thesis without his permission.

Thus, on the basis of the data mentioned above, the genus *Callirhytis* is currently represented in the Western Palaearctic by 6 species: four species are quite common in Western and Central Europe (*C. bella* (Dettmer, 1930), *C. erythrocephala*, *C. glandium* and *C. rufescens*) and two species are from Azerbaijan, Georgia and the Krasnodar region of Russia (*C. comantis* Belizin & Maisuradze and *C. reticulatus* Belizin & Maisuradze). *Callirhytis bella* (sexual generation only), *C. comantis* (sexual generation only) and *C. reticulatus* (asexual generation only) are known from a single generation only, so their life-cycles are not closed yet. It is possible that *C. comantis* is the sexual generation of *C. reticulatus*, however, it must be demonstrated experimentally or gene sequences must be obtained.

Callirhytis erythrocephala is native to Central Europe, and a presumed invader in northern Europe where *Q. cerris* has been introduced. As we have mentioned, RF (in 1964-1967) and FB (in 1971-1986) independently closed the life-cycle of this species but their results have never been published until now. Below we discuss these experiments, describe for the first time the gall of the sexual form of *C. erythrocephala*, redescribe the sexual females and males and synonymize *C. hartigi* with *C. erythrocephala*.

Material and methods

Twenty experiments involving sixty-seven agamic females were undertaken by RF in 1964, 1966 and 1967 on *Quercus robur* L., *Q.* pubescens Willd., Q. petraea (Matt.) and Q. cerris L. In each experiment, as is the custom in the study of cynipid life-cycles, an oak twig is isolated for as long as possible from any other gall-making cynipid by a transparent tissue bag. The buds are separated from the bag tissue by a light metallic armature in order to be accessible (see Garbin et al., 2006: figure 3). The insects studied are introduced into the bag after their emergence from galls. The experiments on O. pubescens, Q. petraea and Q. cerris did not give any results. One of the agamic females used on Q. cerris was seen stinging a bud like an Andricus kollari sexual female (= circulans), the head towards the base of the bud but, as mentioned above, nothing was produced. Nine of the experiments were carried out on O. robur. In one of them the result was doubtful but in another experiment reliable results were obtained. This last experiment is described below.

The bag was placed as early as the 14th of September on a twig of O. robur near to la Rochelle (Baillac) in France. On the 26th of March, three black Callirhytis agamic females were introduced. On the 30th of June, the bag was opened. Two living females were collected. The twigs showed three holes (one insect of the sexual form escaped). The «gall» of the sexual form is in fact inside the twigs. One month later it was difficult to find again the sites of the holes, which are closed by a kind of operculum. When dissecting a twig in the place where a hole was previously seen, the «gall» seems to be filled up with plant tissue. Finally, a kind of camouflage makes the gall and its hole almost invisible (this phenomenon was also observed by FB in his own experiments). The experimentally obtained sexual females were included in the FB collection and given to JP-V. This material consists of two sexual females labelled as «Mr. Folliot, exp. 1827, Ed. 30.6.64» and 6 asexual black C. erythrocephala labelled as «Callirhytis 'glandium', Rennes 3eme année». After studying (JP-V) this material, there is no doubt that the asexual forms from RF belong to the black form C. ervthrocephala.

F. Barbotin made his observations and experiments on *Q. robur* from 1971 to 1986. In one experiment he observed, in March, agamic females stinging head down for a long time on oak buds enclosed in a bag, and he saw the thickening of the stinged twigs. According to FB's notes, in late May and at the beginning of June, he obtained many insects of the sexual form, for example 27 males and 95 females in an experiment with three agamic females. The early emergence of these sexual insects is certainly related to the fact that the young oak used was kept indoors. In other experiments the emergence of the sexual form was observed from late June until mid-July. The material

selected by FB, now deposited in JP-V's collection, is kept in this collection as: «Exp 71: 2 females (21.VI.71, Poitiers)», «Exp 74: 7 males (1-15.VII.74, Biard)», «Exp 79A: 12 males & 23 females (21.V.79-7.VI.79, St Malo)», «Exp 80: 7 males (8-10.VI.1980, St Malo)», «Exp 82: 4 females (19.VI.82, St Malo)» and «Exp 83: 1 male & 7 females (12-19.VII.1983, St Malo)».

The current morphological terminology is given after Gibson (1985), Ronquist & Nordlander (1989), and Fergusson (1995). The abbreviations for forewing venation follow Ronquist & Nordlander (1989). The measurements and abbreviations used herein include: F1 - F12, first and subsequent flagellomeres; POL (post-ocellar distance), the distance between the inner margins of the posterior ocelli; OOL (ocellar-ocular distance), the distance from the outer edge of the lateral ocellus to the inner margin of the compound eye; LOL, the distance between the lateral and the frontal ocellus; transfacial distance, the distance between the inner margins of the compound eves measured across the toruli.

The SEM pictures were made at low voltage without coating the specimens in order to preserve the type material. Wing and gall pictures were taken with a digital camera.

Finally, we also studied the type material of *Callirhytis hartigi* Förster, 1869, deposited at the Naturhistorisches Museum Wien (NMW) and we designated the lectotype. All the other material mentioned is deposited at the University of Barcelona (UB) in JP-V's collection or at the Systematic Parasitoid Laboratory (SPL) in GM's collection.

Results

Callirhytis erythrocephala (Giraud, 1859) *Andricus erythrocephalus* Giraud, 1859 (asexual form) *Callirhytis hartigi* Förster, 1869 (sexual form), **syn. nov.**

- Callirhytis erythrocephala (Giraud) Kieffer 1901 (asexual form)
- Callirhytis erythrosoma Dettmer, 1933 (asexual form); synonym in Nieves-Aldrey 1992
- Callirhytis erythrostoma Dettmer, 1933 (asexual form); synonym in Nieves-Aldrey 1992

The adults and galls of the asexual form have been described in Nieves-Aldrey (1992). The sexual form of *C. erythrocephala* (= *C. hartigi* **syn nov.**) is re-described here because no re-description was made in the preceding studies.

Redescription of the sexual form

Type material: Callirhytis hartigi Förster, 1869, 2 males (lectotype and paralectotype are designated here), deposited in NMW. LECTOTYPE with the following labels: «15/ 817» (white label), «Aachen» (white label), «Först» (red label), «male & female» (symbols, white label), «Typ» (white label, hand-written), «Collect. G. Mayr» (white label), «Call. Hartigi Förster type» (white label), «Lectotype of Callirhytis hartigi Förster 1869, desig. J.P-V» (red label), «Callirhytis erythrocephala (Giraud, 1859) sex gen. male det. J.P-V-2006» (white label); PARALECTOTYPE with the following labels: «Typ» (white label, hand-written), «Collect. G. Mayr» (white label), «Call. Hartigi Förster type» (white label), «Paralectotype of Callirhytis hartigi Förster 1869» (red label), «Callirhytis erythrocephala (Giraud, 1859) sex gen. male det. J. P-V-2006» (white label).

Experimental material FROM BARBOTIN (deposited in UB): 2 females (21.VI.71, Poitiers); 7 males (1-15.VII.74, Biard); 12 males & 23 females (21.V.79-7.VI.79, St Malo); Exp 80: 7 males (8-10.VI.1980, St Malo); Exp. 82: 4 females (19.VI.82, St Malo); Exp 83: 1 male & 7 females (12-19.VII.1983, St Malo).

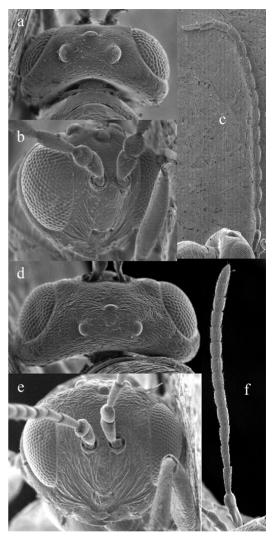


FIGURE 1. Callirhytis erythrocephala (= erythrostoma). (a) head of male, dorsal view, (b) head of male, front view, (c) male antenna, (d) head of female, dorsal view, (e) head of female, front view, (f) female antenna.

FROM FOLLIOT (deposited in UB): 2 females (exp. 1827, Ed. 30-6-64).

Other material examined. «HUNGARY, Dudar, light trap, 1983.VII.4»: 1 female and 1 male (deposited in SPL).

Length: 1.2-2.0 mm. (females), 1.0-1.5 mm. (males)

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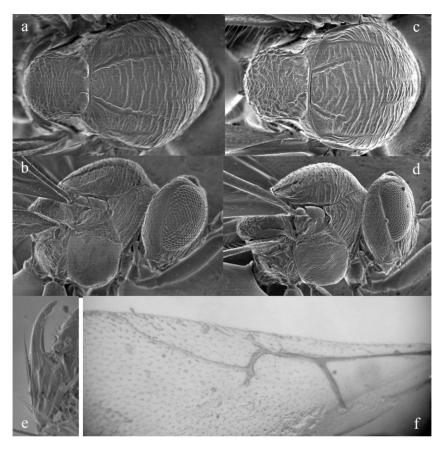


FIGURE 2. *Callirhytis erythrocephala* (= *erythrostoma*). (a) mesosoma of male, dorsal view, (b) head and mesosoma of male, lateral view, (c) mesosoma of female, dorsal view, (d) head and mesosoma of female, lateral view, (e) tarsal claw, (f) radial cell.

Colour: Variable, body yellow-chestnut to chestnut, the head sometimes yellow-ambarine; antenna yellowish to light chestnut, first antennomeres usually lighter; legs yellowish to light chestnut; veins light, yellowish to white.

Female head (Figures 1d, 1e, 4b): Around 2.0 times as broad as long in dorsal view. Gena without punctures, slightly broadened behind compound eye. POL 1.5-1.6 times OOL, OOL around 2.2 times cross diameter of lateral ocellus and around 1.3 times LOL. Frons and vertex coriaceous-granulose, 1.3-

1.4 times as high as broad in frontal view, with short and sparse setae; lower face with striae irradiating from clypeus margin, absent on gena. Transfacial distance around 1.1 times height of compound eye. Diameter of torulus shorter than distance between toruli, and longer than distance between torulus and inner margin of compound eye. Malar space around 0.3 times as long as height of compound eye. Malar sulcus present. Clypeus subquadrangular and shortly bilobed ventrally, with distinct epistomal sulcus and clypeopleurostomal line. Antenna (Figures 1f, 4b): around 0.75 times as long as body length, 14segmented (in some specimens 15 segmented, when there is a distinct suture between F12 and F13); pedicel 1.1-1.3 times as long as broad, F1 1.0-1.2 times as long as F2 and 1.0-1.3 times as long as pedicel; F3 subequal or slightly longer than F4; subsequent flagellomeres very slightly and gradually shorter, last flagellomere 1.5-2.0 times as long as broad; F4-F12 (sometimes F3 as well) broader than F1-F2. Placodeal sensilla present on F3-F12 (sometimes also on F2), but always indistinct on the first flagellomeres.

Female mesosoma (Figures 2c, 2d, 4d): very sparsely pubescent. Pronotum with weak rugae laterally. Scutum with regular and conspicuous transverse rugae, broadly separated; interspaces between rugae transversely sculptured, reticulate; notauli incomplete, distinct in the posterior 1/3 to 1/2 of scutum; median mesoscutal line absent. Scutellum quadrangular, with some rugae, interspaces reticulated; scutellar foveae slightly developed, superficial, smooth or slightly carenated and broadly separated or separated by a distinct carinae. Mesopleuron weakly sculptured, coriaceous-reticulate, with some weak carina in the central area. Lateral propodeal with parallel carinae. Tarsal claws simple, without basal tooth (Figure 2e). Forewings (Figure 2f): hyaline; margin with short cilia. Radial cell around 3.7 times as long as broad; Rs curved and conspicuously projected; R1 not reaching wing margin; 2r curved; areolet and RS+M vein indicated by a weak infuscation.

Female metasoma: Equal in length to head+mesosoma; all tergites smooth, without punctures; metasomal tergite II occupying 0.5 of metasoma length in dorsal view. Ventral spine of hypopygium short, prominent part around 2.0 times as long as broad, with sparse short setae.

Male (Figures 1a-c, 2a-b, 4a, 4c): similar to female, except head around 1.9 times as

broad as high in dorsal view; transfacial distance 0.8 times height of compound eye; malar space around 0.5 times height of compound eye; POL:OOL:LOL around 15:7:3; OOL 0.9-1.2 times as long as diameter of lateral ocellus. Antenna longer, 15-segmented (sometimes 16, as in *C. hartigi* lectotype); F1 excavated and curved, weakly enlarged apically; F2 equal in length to F1 but not excavated and slightly broader. Mesosoma similar to female, but notauli longer; mesopleuron with weaker sculpture, shiny, almost smooth. Metasoma shorter than mesosoma.

Diagnosis: The sexual form of *C. ervthro*cephala is closely related to the sexual form of C. rufescens. We can differentiate the males according to Nieves-Aldrey (1992: 173). In C. ervthrocephala females the notauli are incomplete; the scutellum is quadrangular, the prominent part of the ventral spine of the hypopygium is shorter and the radial cell of the forewing is longer, while in C. rufescens the notauli are complete and reach the pronotum; the scutellum is subovate, the prominent part of the ventral spine of the hypopygium is longer, more than 2.0 times as long as broad; the radial cell is shorter. The adults of C. rufescens are always ambarine in colour but the sexual adults of C. erythrocephala are vellow-chestnut to chestnut on the mesosoma and the metasoma (sometimes also on the head).

Host oaks: In FB's and RF's experiments, the sexual generation galls were induced in *Q. robur* twigs and no galls were obtained with *Q. petraea*, *Q. pubescens* or *Q. cerris*.

Sexual gall (Figure 3c): An extremely cryptic gall induced beneath the bark of branches and shoots, without any visible external deformation of the shoot. Unilocular, often aggregated, small larval chambers are scattered beneath the bark. The galls can easily be located by the emergence holes made by the adults. Sexual galls of C.

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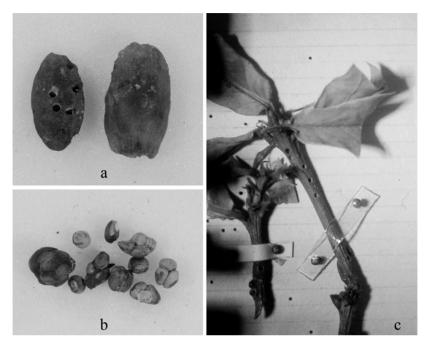


FIGURE 3. Callirhytis erythrocephala. (a) and (b) diversity of asexual galls (see comments in the Discussion): (a) galls of Callirhytis erythrocephala (= erythrostoma), «(a)-type galls»; (b) isolated galls of Callirhytis erythrocephala (= erythrosoma), «(b)-type galls». (c) experimental sexual gall from FB.

erythrocephala are impossible to distinguish from sexual galls of *C. glandium* if these galls are developing in *Q. robur* as well.

Phenology: The sexual generation develops in summer; adults emerge in June-July, rarely by the end of May. The asexual larva completes its development by the time the acorn ripens. The asexual female has a 3-8 year long diapause (according to FB).

Taxonomic note: *Fioriella meunieri* Kieffer, 1902, a species with an uncertain status, might be a possible synonym of *C. erythrocephala* (= *C. hartigi*) (Melika *et al.*, 2001).

Discussion

In acorns of *Q. cerris* two gall models can be observed, according to FB's notes and experiments:

(a) a typical form, named as *C. erythrocephala* (= *erythrostoma*), with an almond-shaped gall, coalescent and hard, often resulting in a complete fusing of single unilocular galls into one large agglomerate in the endocarpium (Figure 3a);

(b) a hemispherical group of gall chambers which can usually be separated into individual gall chambers (Figure 3b), named earlier as *C. erythrosoma* (synonymized to *C. erythrocephala* by Nieves-Aldrey (1992) and was described by Dettmer (1933) without knowing the gall).

The asexual females of both forms are closely related but can be easily distinguished according to FB's notes: females reared from (b)-type galls are red-yellowish while those reared from the (a)-type galls have a black mesosoma. The colouration in *Callirhytis* is a species-specific characteristic, except for the

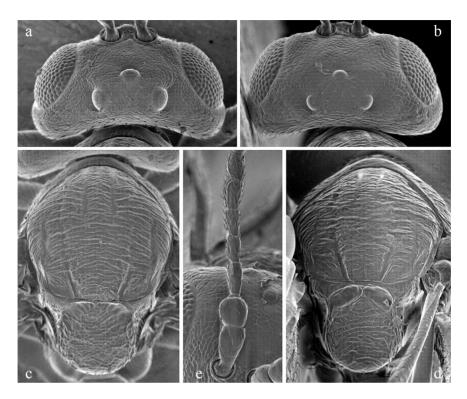


FIGURE 4. *Callirhytis erythrocephala* (= *erythrosoma*), (a) head of male, dorsal view, (b) head of female, dorsal view, (c) mesosoma of male, dorsal view, (d) mesosoma of female, dorsal view,(e) first flagellomeres of female antenna.

chromatic variability in *C. erythrocephala* (Nieves-Aldrey, 1992).

Some morphological differences between the sexual adults reared from the two gall types can also be observed, based on the specimens FB obtained by rearing in his experiments. Figures 1 and 2 correspond to sexual adults reared from the (a)-type galls and Figure 4 corresponds to those reared from the (b)-type galls. Sexual adults from the (b)-type galls have a broader head (Figures 4a-b) and the POL:OOL ratios are larger compared with the ocellus diameter (Figures 4a-b); F2 in females is shorter (Figure 4e); the notauli in males are shorter (Figure 4c); the scutellar contiguous (Figure 4c-d), foveae are separated by a shorter distance. The Callirhytis *hartigi* males from the typical series resemble the males reared from the (a)-type galls in FB's experiments.

It is possible that these two gall forms are induced by different species. However, we need to close the life-cycles again and study the possibility of hybridization between the two sexual forms to ensure it. This is a new objective that could only be attained after a few years of experimentation because of the long diapause of the asexual larvae (3-8 years). Even if we think that we have two different species, in heterogonic Cynipidae it is easier to create a synonymy than to undo one. For this reason, we consider essential a prudent attitude towards it.

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