

## TWO NEW SPECIES OF HOHENBUEHELIA FROM ARGENTINA

ALEJANDRA FAZIO

PRHIDEB-CONICET. Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales, Departamento de Ciencias Biológicas, Ciudad Universitaria, Pab. II, 1428 Buenos Aires, Argentina

EDGARDO ALBERTÓ

Instituto de Investigaciones Biotecnológicas, IIB-INTECH (UNSAM-CONICET). Camino de Circunv. Km 6, 7130 Chascomús, Buenos Aires, Argentina.  
E-mail: ed@intech.gov.ar

**Abstract:** During a revision of the genus *Hohenbuehelia* in Argentina, two new species were found: *H. austrocedri* from the province of Chubut, and *H. minutissima* from the province of Buenos Aires. Both belong to subgenus *Hohenbuehelia* section *Nigricans*.

In the present paper we describe and illustrate the two new species and compare them with *H. singeri*, *H. nigra*, *H. bullulifera* and *H. unguicularis*. In addition, we studied the holotype of *H. pilitomaculoides*, which we reduce to a synonym of *H. nigra*.

**Key Words:** Agaricales, *Hohenbuehelia austrocedri*, *H. minutissima*, *H. sect. Nigricans*, systematics.

## INTRODUCTION

Knowledge of the genus *Hohenbuehelia* in Argentina is scant. Spegazzini described several new taxa as *Agaricus portegnus* Speg., *Pleurotus paraguayensis* Speg., *P. portegnus* var *microspora* Speg. And *P. petaloides* var *victoriensis* Speg. (Spegazzini 1881, 1902, 1921, 1922). Singer & Digilio (1951) published *Hohenbuehelia bullulifera* Singer; while later Raiethelhuber (1984, 1990) described *H. picicola* Raiethelhuber and *H. canubrona* Raiethelhuber. Recently Albertó *et al.*, (1998) studied the *H. nigra* complex and described *H. singeri* (Singer) Albertó & Fazio and *H. nigra* var *pileocystidiata* Fazio & Albertó.

In the present paper we illustrate and describe two new species, one from the Province of Chubut and the other from Province of Buenos Aires, Argentina. We compare them with *H. singeri*, *H. nigra* (Schwein.) Singer, *H. bullulifera* and *H. pilitomaculoides* Libonati-Barnes. After studying the type material of *H. pilitomaculoides*, which also belongs to section *Nigricans*, we consider this species to be a late synonym of *H. nigra*.

## MATERIALS AND METHODS

Dried specimens were studied macro- and microscopically. Color terms and annotations follow Munsell (1954). Abbreviations of author's names are according to Kirk and Ansell (1992). Freehand sections of specimens were mounted in 10 % KOH plus 1 % aqueous solution of phloxine and in Melzer's reagent. All collections are deposited in the mycological Herbarium, Dept. de Ciencias Biológicas, Facultad de Ciencias Exactas y Naturales, University of Buenos Aires (BAFC). Herbaria abbreviations follow Holmgren *et al.* (1990).

## RESULTS

### KEY TO SPECIES OF SECTION NIGRICANS STUDIED

1. Pileocystidia absent ..... 2
- 1 Pileocystidia present ..... 4
  - 2 Leptocystidial pleurocystidia absent or inconspicuous; spores > 6  $\mu\text{m}$  long ..... *H. unguicularis*
  - 2 Leptocystidial pleurocystidia present; spores  $\leq$  6  $\mu\text{m}$  long ..... 3
- 3 Pileus small, up to 2.5 mm diam., pseudostipe present; spores 5.2-6.0 X 3.1-3-6  $\mu\text{m}$  ..... *H. minutissima*
- 3 Pileus larger, up to 5 mm diam., pseudostipe absent; spores 4-5(-5.5) X 3-5  $\mu\text{m}$  ..... *H. singeri*
- 4 Pileocystidia thin-walled ..... 5
- 4 Pileocystidia metuloid ..... 6
- 5 Cheilocystidia leptocystidia, vesiculose-pedicellate ..... *H. bullulifera*
- 5 Cheilocystidia metuloid, ventricose to fusiform ..... *H. subbarbata*
  - 6 Spores 9-10.5  $\mu\text{m}$  long; pleurocystidia hyaline ..... *H. austrocedri*
  - 6 Spores 6.5-8  $\mu\text{m}$  long; pleurocystidia brown ..... 7
- 7 Two types of pileocystidia present; cutis formed by only one type of hyphae ..... *H. nigra* var. *pileocystidiata*
- 7 One type of pileocystidia present; cutis formed by three types of hyphae ..... *H. nigra* var. *nigra*

### *Hohenbuehelia austrocedri* Fazio & Albertó sp. nov.

*Basidiocarpus pileatus, conchatus, nigrus, unguicularis, ad duorum textus, inferus tomentosus brunneo-castaneus, ad margo canescenti. Stipes absens, pileus ad substratum adhaerentibus in locus unicus. Lamellae obscure griseae, subconfertis vel subdistantibus, margo liso, cum lamellulae intercalares. Pulvis sporarum niveis.*

*Sporis 9-10,5 x 4-4,5  $\mu\text{m}$ , cylindricis vel ellipsoideis, hyalinis, laevis, inamyloideis. Basidiis 38-55 x 6-8  $\mu\text{m}$ , longe sterigmaticis. Cheilocystidiis 32-41,5 x 6-12,5  $\mu\text{m}$ , gracilis, extremis globosis, incrustatis, crystallis brunneis amorphibus, in alkali dissolventis. Pleurocystidiis formae magnitudinisque. Pileipellis hyphis curtis, brunneis, crassitunicatis, exornatis. Mycosclereidiis irregularibus, crassitunicatis. Pileocystidiis metuloideis, hyalinis,*

*crassitunicatis, incrustatis; crystallis amorphis in alkali dissolventes. Pilus duorum hyphis formatus: a) brunneis, crassitunicatis, efibulatis; b) hyalinis, crassitunicatis, parce fibulatis chlamydo sporis interdum praesentibus. Subpileus strato gelificatus 150-300  $\mu\text{m}$  et hyphis hyalinis, tenuicatis, fibulatis exornatis.*

*Hab. ad corticium muscosis Austrocedri chilensis*

*HOLOTYPE: ARGENTINA, Chubut, PN. Lago Puelo, Lago Puelo, in Austrocedreto, pr. flumine Los Tineos, intefolia dejecta Austrocedri chilensis, leg. M. Rajchenberg (11.180), 10-V-96 in herbarium BAFC sub n° 34.999 conservatus est.*

Basidiocarp 5-15 X 5-20 mm, pileate, conchate, dark brown to black (5YR 2/2 - 10YR 3/3), with a brown (10YR 4/3) tomentum at the base which covers 2/3 of the pileus surface, then becomes a whitish pruina towards the margin. Stipe absent, the pileus is laterally attached to substratum by part of the underside (Fig. 1A and B). Lamellae very dark gray (7.5YR 3/1), close to subdistant; lamellar margin entire, with small intercalary lamellulae, interlamellar space yellowish brown (10YR). Spore print white.

Spores 9-10.5 X 4-4.5  $\mu\text{m}$ , cylindric to ellipsoid, thin-walled, hyaline, smooth, non amyloid (Fig. 1D and 2I). Basidia 38-55 X 6-8  $\mu\text{m}$ , with sterigmata 7  $\mu\text{m}$  long, clavate, 4-spored, densely guttulated (Fig. 2D). Cheilocystidia 32-41.5 X 6-12.5  $\mu\text{m}$ , thin, apex subglobose, encrusted with small, amorphous brown crystals which are soluble in KOH. Pleurocystidia of identical shape and size (Fig. 2A, C). Pileipellis formed by short, brown, very thick-walled hyphae (Fig. 2G), 5.5-7.5  $\mu\text{m}$  diam. and modified, thick-walled, brown structures similar to mycosclereids (Wright, 1955), 12-22 X 3.5-6  $\mu\text{m}$  very irregular in shape. Pileocystidia 17-26 X 6-12  $\mu\text{m}$ , metuloid, hyaline, very thick-walled, covered with encrusted amorphous crystals which are soluble in 10 % KOH (Fig. 2B). The hairs of the tomentum are formed by two types of erect hyphae: a) brown to ochraceous-brown, thick-walled, tortuous, clampless hyphae, 1.5-3  $\mu\text{m}$  diam., and b) less abundant, hyaline, thick-walled hyphae, with few clamps (Fig. 2F and H). Terminal and intercalary chlamydo spores, 7-20  $\mu\text{m}$  diam. can be observed in some specimens (Fig. 2E). Context formed by a loose trama of hyaline, thin, clamped hyphae with gelatinized walls, 3-8  $\mu\text{m}$  diam., that define a layer 150-300  $\mu\text{m}$  thick.

Habitat: growing among musci on wet bark of *Austrocedrus chilensis*.

Material studied: The holotype: ARGENTINA, Chubut, Pque. Nac. Lago Puelo, Lago Puelo, forest of *Austrocedrus chilensis* next to the stream Los tineos, leg. M. Rajchenberg (11.180) growing on bark, 10-V-96, BAFC 34.999; Chubut, Pque. Nac. Lago Puelo, Lago Puelo, forest of *Austrocedrus chilensis* next to the stream Los tineos, leg. M. Rajchenberg (11.145) growing on bark, 10-V-96 (CIEFAP); Chubut, Pque. Nac. Lago Puelo, Lago Puelo, West branch, International Boundary, in mixed forest of *Austrocedrus chilensis*, *Nothofagus dombeyi* and *Guevinia avellano*, leg. M. Rajchenberg (11.175), 10-V-96 (CIEFAP).

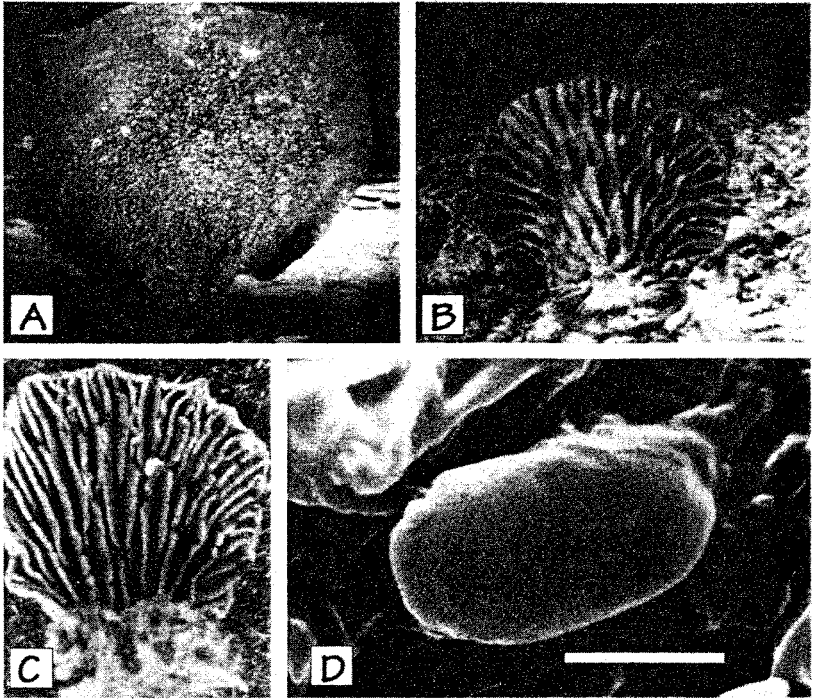


Fig. 1: *Hohenbuehelia austrocedri* and *H. minutissima*. A and B, Basidiocarp of *H. austrocedri*; D, Spore of *H. austrocedri* under SM; C, Basidiocarp of *H. minutissima*. Scale bar Fig. A and B= 10 mm, C = 1 mm, D= 5  $\mu$ m.

***Hohenbuehelia minutissima* Fazio & Albertó sp. nov.**

*Basidiocarpus minutus, petaloideus, spathulatus, substipitatus, flabellatus, pruinosis, obscure cinereus, pruina nivescentibus, pili pseudostipitem abundantibus, concoloribus, hirsutibus, zona ad pilea glabra, cremea vel alba; margo sinuatus.*

*Sporis 5-6 x 3-3,6  $\mu$ m, ellipsoideis, hyalinis, laevis, inamyloideis. Basidiis tetrasporis. Pleurocystidiis claviformibus, 21-27 x 11-16  $\mu$ m, tenuitunicatis, incrustatis, cum yodo metuloideis simulantibus in alkali dissolventis. Pileipellis hyphis hyalinis, tenuitunicatis,*

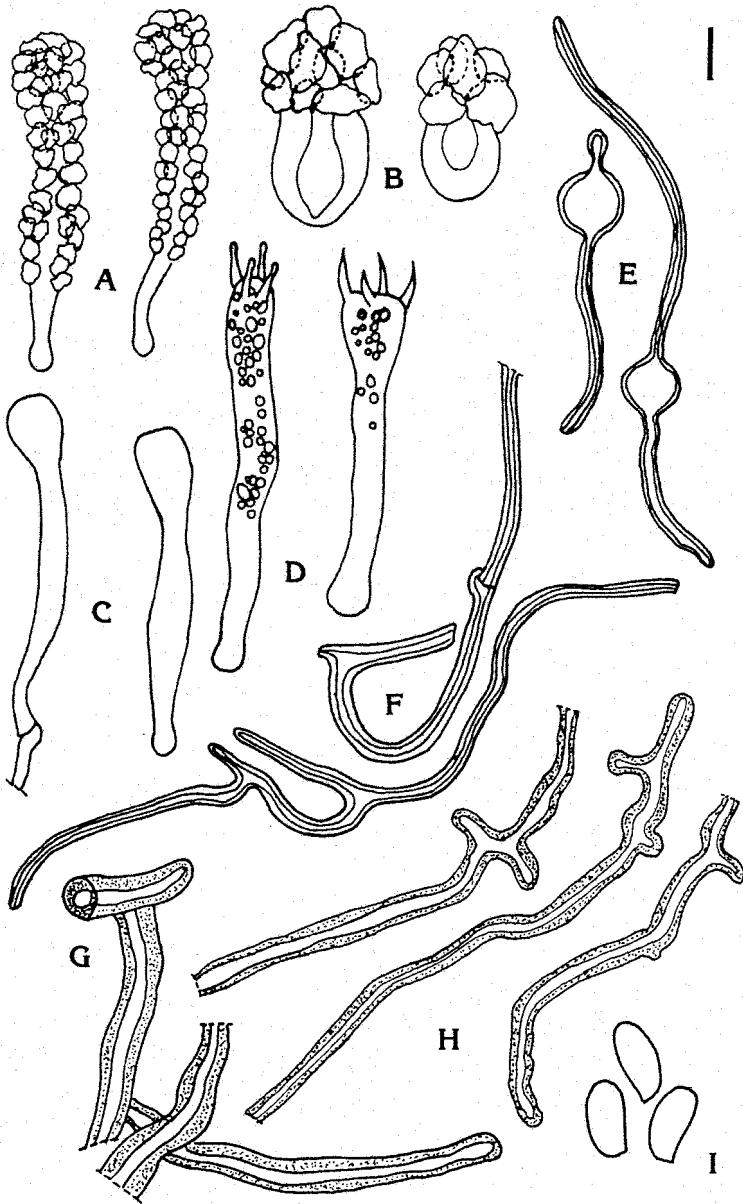


Fig. 2: Microscopic features of *Hohenbuehelia austrocedri*. A, Pleurocystidia; B, Pileocystidia covered with amorphous crystals; C, Pleurocystidia without crystals; D, Basidia; E, Chlamydospores; F and H, Hyphae of the hairs of the tomentum; G, Hyphae of the pileipellis; I, Spores. Scale bar 6  $\mu$ m.

*fibulatis, ramificatis exornatis. Hyphis contextibus idem in matricibus plus minusve gelificatis immersis. trama lamellibus hyphis conductivis pallide brunneis, tenuitunicatis efformantibus.*

*Holotypus:* ARGENTINA, Bonariae, Llavallol, Sta. Catalina, leg. Enrique Fernández, 14.III.94, in herbarium sub. n° BAFC: 34.374 conservatus est.

Basidiocarp 1-3 X 1-2.5 mm, petaloid, spatulate, substipitate, flabelliform, pruinose, very dark gray (10YR 3/1), with whitish pruina; pseudostipe with abundant erect concolorous hairs, then becoming smooth, white to cream colored; margin of pileus slightly festooned (Fig. 1C and 3F). Lamellae gray (10YR 5/1), eccentric, margin entire.

Spores 5.2-6.0 X 3.1-3.6  $\mu\text{m}$ , ellipsoid, hyaline, smooth, non amyloid (Fig. 3E). Basidia 25-27 X 6-7  $\mu\text{m}$ , clavate, 4-spored. Cheilocystidia not observed. Pleurocystidia 22-27 X 11.5-15.5  $\mu\text{m}$ , clavate, thin-walled, encrusted with amorphous crystals, appearing metuloid in Melzer's reagent, crystals soluble in KOH (Fig. 3C and D). Pileipellis formed by hyaline, straight, thin-walled, clumped, branched, interwoven hyphae, 2-3  $\mu\text{m}$  diam., immersed in a more or less gelatinized matrix (Fig. 3A). Trama of the lamellae with pale brown, thin-walled, clumped, conducting hyphae, 2.6-3.1  $\mu\text{m}$  (Fig. 3B).

Material studied: *Holotypus:* ARGENTINA, Buenos Aires, Llavallol, Sta. Catalina, leg. E. Fernández, 14.III.94, BAFC: 34.374

## DISCUSSION

Subgenus *Hohenbuehelia* Section *Nigricans*, includes species with black lamellae and smooth spores. After studying many species of this section it was possible to differentiate clearly two groups: 1) species with metuloid pleurocystidia, thick-walled, with the apex covered with regular polyhedral crystals, which we name "pleurocystidia nigra type" (see Albertó *et. al.* 1998); 2) species with lamprocystidiate pleurocystidia, covered all along the surface by amorphous crystals, which easily dissolve in KOH, appearing metuloid when observed with the crystals but actually thin-walled, which we name "pleurocystidia singeri type" (Fig. 3C and D). *Hohenbuehelia austrocedrii* and *H. minutissima* belong to the latter group.

*Hohenbuehelia austrocedrii* is characterized by a dark pileus covered by a brown tomentum, subdistant lamellae, yellowish brown interlamellar space, thin-walled cystidia, covered by amorphous, opaque crystals that dissolve in KOH, and by long cylindrical spores. We observed that, in some young specimens, lamellae are yellowish-brown, the same color as the interlamellar space. The lamellae become darker during basidiocarp development and, finally, become very dark gray in mature specimens.

*H. minutissima* is characterized by a minute basidiocarp, a pseudostipe with abundant erect hairs, thin-walled cystidia covered by amorphous crystals which are easily soluble in KOH, and by the presence of pale brown conducting hyphae in the trama of the lamellae.

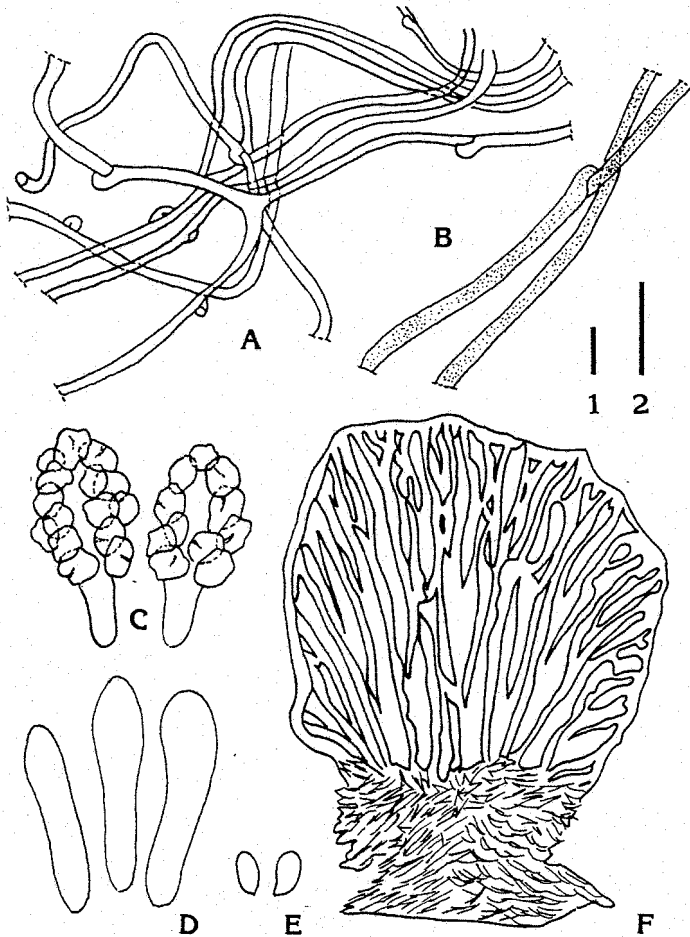


Fig. 3: *Hohenbuehelia minutissima*. A, Hyphae of the pileipellis; B, Conducting hyphae of the trama of the lamellae; C, Pleurocystidia covered with amorphous crystals; D, Pleurocystidia without crystals; E, spores; F, Basidiocarp. Scale bar 1: Figs. A, B, C, D and E= 6.5  $\mu$ m; Scale bar 2: Fig. F= 5 mm.

Both two new species are related with *H. singeri* because of the thin-walled pleurocystidia type, but *H. austrocedri* differs in the distinctly longer spores and the presence of mycosclereids similar to those found in *H. nigra* by Albertó *et al.* (1998).

*Hohenbuehelia minutissima* differs from *H. singeri* because the latter has larger pileus, up to 5 mm diam. with a white pruina abundant in young specimens, spseudostipe absent and spores shorter, 4-5(5.5)  $\mu\text{m}$  long.

Comparing *H. minutissima* with *H. bullulifera* we observed that although both species have a small, dark basidiocarp and spores of similar shape and size, *H. bullulifera* lacks a stipe, has abundant fusoid, thick-walled metuloids pleurocystidia, cheilocystida that are vesiculose-pedicellate, hyaline or incrustated with brown pigment, and has subglobose well-differentiated pileocystidia similar to cheilocystidia.

*H. subbarbata* (Berk. & Curtis) Sing. and *H. bullulifera* are very similar species. Unfortunately we have been unable to study the original material of *H. bullulifera* in which Singer and Digilio (1951) based their description and which was originally deposited in LIL; this material is presumed lost. We based on concept of *H. bullulifera* in the data provide in the protologue.

We also studied the type material of *H. pilitomaculoides*. Libonati-Barnes (1994) reported that *H. pilitomaculoides* was very similar to *H. nigra* (Schwein.) Singer but differed because the former had lacked pileocystidia. Upon studying its holotype (WTU) we could clearly observed the presence of brown, thick-walled, crystal incrustated pileocystidia, very similar to those present in the holotype of *H. nigra* (Albertó *et al.* 1998). We thus conclude that *H. pilitomaculoides* Libonati-Barnes is a synonym of *H. nigra* (Schwein.) Singer.

#### ACKNOWLEDGMENTS

We are very grateful to Dr. Mario Rajchenberg (CIEFAP-ARGENTINA) who kindly sent us his collections of *Hohenbuehelia* from Patagonia, Argentina and the gift of the holotype of *H. austrocedrii* to the BAFC herbarium. We also want to thank the curator of WTU for the loan of specimens in their keeping. We are very grateful to Dr. J. E. Wright who wrote the Latin diagnosis and made invaluable suggestions to improve the paper. Dr. D. Desjardin (SFSU), provided a presubmission review for which we are most grateful. This work was funded by the CONICET; publication n° 131 of PRHIDEB.

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