

Tuber rufum from Río Negro, Argentina, with notes on Spegazzini's Tuberales

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Knowledge about the Tuberales of Argentina is very poor. Spegazzini (1887) made the first records describing two species belonging to this group in a more general work with five hypogeous fungi. These two species have received no further attention. Although there is not a complete study of the Argentine Tuberales, Crespo and Domínguez de Toledo (1994) have recently reported *Tuber separans* Gilkey, from the province of Córdoba. More recently, Crespo (1996, pers. comm.) observed *T. maculatum* Vitt. and *T. shearii* Harkn. from the same province, and Lorenzo & Calvelo (2000) reported *Tuber maculatum* from Patagonia. Almost simultaneously, another truffle was found in a fruit farm from this area, associated with *Tilia* sp. and *Quercus* sp., both of which are introduced deciduous trees, during autumn. It has been identified as *Tuber rufum* Pico.

This paper describes this new record for Argentina and makes some comments on Spegazzini's type material.

Techniques

Observations and measurements were taken from almost fresh material squash-mounted in 5% KOH and phloxine for optical microscopy. Ascospores were also examined by SEM. Specimens were preserved in BAFC (herbarium abbreviations follow Holmgren *et al.*, 1990). Spegazzini's type material from LPS was also studied.

Taxonomy

I) In the light of modern classification systems, following Trappe (1979):

Order Pezizales

Fam. Tuberales

Tuber rufum Pico: Fr., *Syst. Mycol.* 2: 292, 1823.

For synonymy, see Pegler *et al.* (1993). Figs 1-3, 9

Ascomata hypogeous, subglobose, sometimes lobed, 6-20 mm diam., surface light orange-brown or rufous, glabrous, smooth or commonly minutely warted, solid, firm. *Odour* like humid soil, persistent. *Peridium* 250-400 µm thick, towards the surface composed of subangular, pigmented cells 12-15 µm diam. *Gleba* at first whitish, becoming greyish-yellow to pale orange-brown, marbled with conspicuous, broad, dark brown, branching veins. *Asci* 57-83 x 44-63 µm, subglobose with a short or sometimes elongated stalk, 1 to 5-spored. *Ascospores* 25-32 (-40) x 18-25 (-29) µm excluding ornamentation, broadly ellipsoid to ovate-ellipsoid, yellow-brown, ornamented with pointed, often curved spines 2-3 µm long.

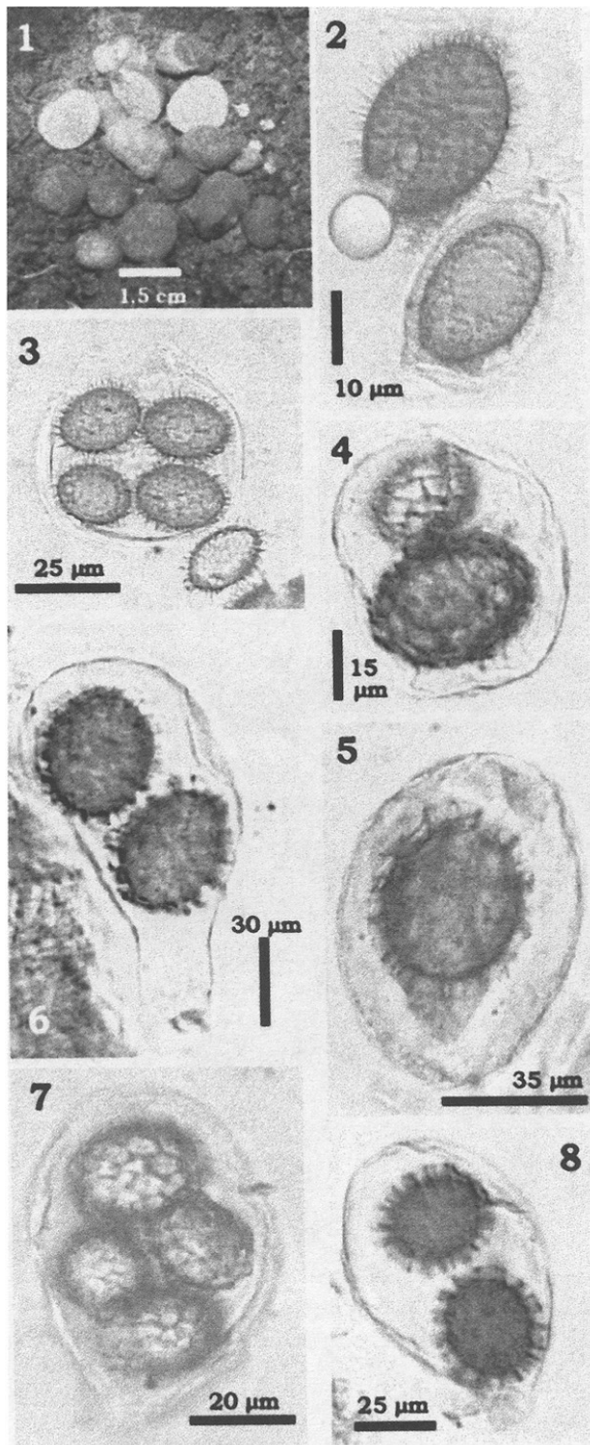
Specimens examined: Argentina: Río Negro, Departamento Gral Roca, Ciudad Gral. Roca, Belgrano and 25 de Mayo, March 1998, leg: M. L. de Hernandez & Blumenfeld, S. Det.: A. I. Romero (BAFC 50.560); *ibid* June-1999 (BAFC 50.561); Feb.-2000, *ibid* (BAFC 50.562).

II) Comments on Spegazzini 's holotypes of *Tuber* species:

Spegazzini (1880) described *Tuber australe* Speg. (Holotype LPS 1304) from Buenos Aires province and later (1887) *T. argentinum* Speg. (Holotype? LPS 1305) from Chaco province. Some years later (1909) he also proposed *T. argentinum* var. *pampearum* Speg. (Holotype LPS 1303) from Buenos Aires.

1) *Tuber australe* Speg., *An. Soc. Cient. Argent.* 10: 149, 1880. Figs 4-5, 11

This seems to resemble *T. maculatum* Vitt. mainly in having similar microscopic features.



Figs 1-3 *Tuber rufum*: Fig 1 Ascomata on the ground; the peridium is usually reddish brown but when cut open shows the whitish gleba, marbled with brown veins corresponding to the spore zones. Fig 2 One monosporous ascus and one free mature ascospore showing the spiny ornament of the wall. Fig 3 One globose ascus bearing four spores. Figs 4-5 *Tuber australe* (Holotype LPS 1304): Fig 4 One globose ascus with two ascospores of different sizes. Fig 5 One subglobose ascus with only one big ascospore. Figs 6-8 *Tuber argentinum* (Holotype LPS 1305) Variations of asci shape and number of ascospores: Fig 6 Stipitate, bisporous ascus. Fig 7 Sessile, subglobose, tetrasporous ascus. Fig 8 Sessile, subglobose, bisporous ascus.

Asci and ascospores are similar in size and shape. The principal differences are those of the ascoma, colour and internal anatomy. The ascomata of *T. australe* Speg., as described in his diagnosis, are white whereas ascomata of *T. maculatum* are at first whitish, becoming pale yellow-brown, with usually brown or reddish brown patches, according to Pegler *et al.* (1993).

Specimen examined: Argentina: Buenos Aires, Capital Federal, Recoleta, 8-VIII-1880, Leg. C. Spegazzini, Holotype LPS 1304.

2) *Tuber argentinum* Speg., *An. Soc. Cient. Argent.* **24**: 123, 1887 nomen dubium Figs 6-8, 10 Apparently the holotype is lost. The LPS 1305 material is not the type. A note inside the envelope reads "probably part of the type", but there are no data of collection. Furthermore, the examination of this specimen has shown that it does not coincide with Spegazzini's original diagnosis based on a collection from Resistencia city, Chaco province in 1886. He described the asci as having 4-12 spores, whereas in this specimen (LPS 1305) the asci have 2, 3 or 4 spores (Figs 6-8), and no 12-spored asci have been observed. Measurements of asci and ascospores given by him are: asci 50-60 x 45-50 µm and ascospores 17-18 µm diam excluding ornamentation (20-22 µm with it). In LPS 1305 the asci are subglobose, 77-95 µm (Figs 7-8) or, they have short stalks 70-90 x 100-120 µm (Fig 6), ascospores are ellipsoid: 35-55 x 40-75 µm (Fig. 10 SEM), some being smaller and globose, 30-40 µm diam.

For these reasons, *T. argentinum* Speg. should be regarded as a *nomen dubium*.

On the other hand, it is hard to identify this material at species level due to the lack of the data of color and size of the ascomata, features of the peridium, etc and due to the condition of the material. All that can be said is that it is a *Tuber* sp.

Specimens examined: *T. argentinum* Speg. LPS 1305, without data, leg. C. Spegazzini.

3) *Tuber argentinum* var. *pampearum* Speg., *An. Mus. Hist. Nac. Buenos Aires ser. 3* 19 (12): 423, 1909.

This variety (LPS 1303) was considered by Gilkey to be a synonym of *Terfezia longii* Gilkey, according to a note made when she examined Spegazzini's material. I agree with her, for although it is not possible to observe the asci, the

typical globose shape of the ascospores is a good character of recognition. Ascospore 20-24 μm diam (Fig 12).

Specimen examined: Argentina: Buenos Aires, Adolfo Alsina, VII-1907, Leg. C. Spegazzini, on roots of *Discaria longispina*, holotype of *Tuber argentinum* var. *pampearum* Speg., LPS 1303. Det.: as *Terfezia longii* by Gilkey.

Conclusion

Tuber rufum is recorded from Argentina for the first time.

As regards the three Spegazzini specimens studied: *Tuber australe* is a good species, *T. argentinum* is a *nomen dubium* and *T. argentinum* var. *pampearum* is a synonym of *Terfezia longii*, the only recorded species of the genus in Argentina.

It seems therefore that five species of Tuberaceae: *T. australe*, *T. separans*, *T. shearii*, *T. rufum* and *T. maculatum* and one from the Terferziaceae have been reported from Argentina, but a further revision of all specimens and more collections are clearly necessary before a full picture can be obtained.

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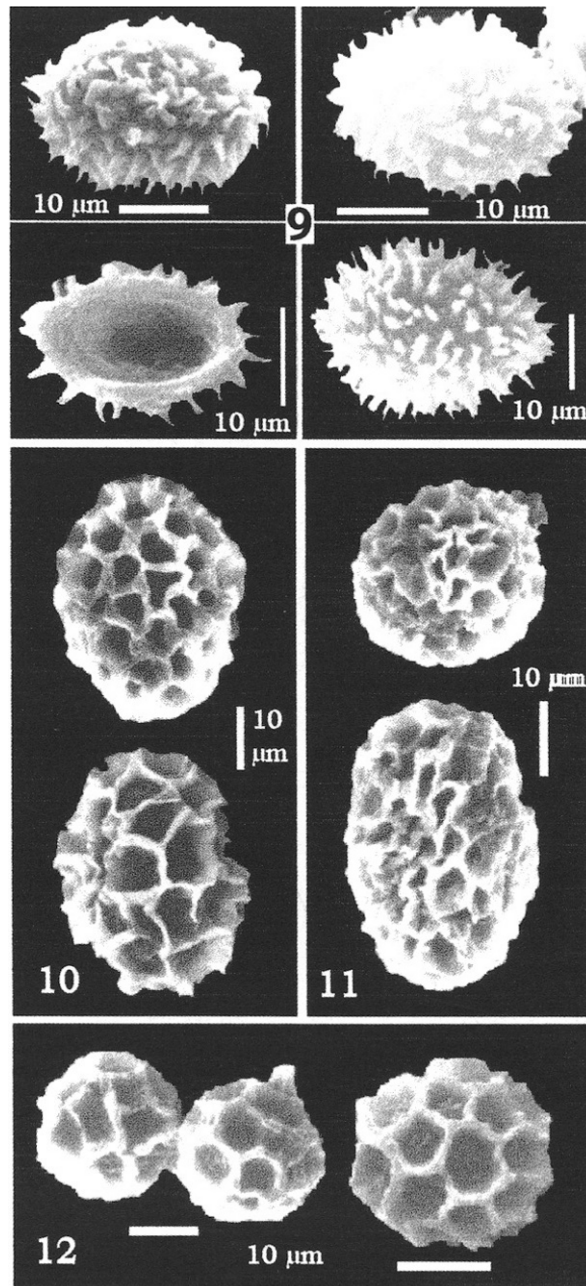


Fig 9 Four ascospores of *Tuber rufum*, in different views showing the spiny wall observed with SEM. Fig 10 Two ascospores of *Tuber argentinum* showing the reticulate wall observed with SEM (Holotype LPS 1305). Fig 11 Two ascospores observed with SEM showing the reticulate wall of *Tuber australe* (Holotype LPS 1304). Fig 12 Spherical ascospores of *Terfezia longii* (Holotype of *Tuber argentinensis* var. *pampearum* LPS 1303) observed with SEM showing the reticulate ornamentation of the wall.

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