Rev Argent Microbiol. 2016;48(1):86–90

REVISTA ARGENTINA DE MICROBIOLOGÍA

www.elsevier.es/ram

ORIGINAL ARTICLE

First record of *Talaromyces udagawae* in soil related to decomposing human remains in Argentina

María C. Tranchida[a,*], **Néstor D. Centeno**[b], **Sebastián A. Stenglein**[c], **Marta N. Cabello**[d]

[a] *Instituto de Botánica C. Spegazzini, Facultad de Ciencias Naturales y Museo Universidad Nacional de La Plata, Consejo Nacional de Investigaciones Científicas y Tecnológicas (CONICET-CCT La Plata), La Plata, Buenos Aires, Argentina*
[b] *Laboratorio de Entomología Aplicada y Forense Universidad Nacional de Quilmes, Bernal, Buenos Aires, Argentina*
[c] *Laboratorio de Biología Funcional y Biotecnología (BIOLAB)-CICBA-INBIOTEC-CONICET, Cátedra de Microbiología, Facultad de Agronomía-UNCPBA, Azul, Buenos Aires, Argentina*
[d] *Instituto de Botánica C. Spegazzini, Facultad de Ciencias Naturales y Museo Universidad Nacional de La Plata, Comisión de Investigaciones Científicas de la Provincia de Buenos Aires (CIC), La Plata, Buenos Aires, Argentina*

Received 24 July 2015; accepted 8 October 2015
Available online 5 January 2016

KEYWORDS
Talaromyces udagawae;
Soil fungi;
Cadaver decomposition;
Ascomycota;
Forensic potential

Abstract The morphologic features of *Talaromyces udagawae* Stolk and Samson are here described and illustrated. This teleomorphic Ascomycota fungus was isolated from soil obtained in Buenos Aires province (Argentina) from beneath a human cadaver in an advanced state of decomposition. After washing and serial dilution of the soil along with moist-chamber techniques for fungal cultivation, *T. udagawae* formed very restricted colonies of bright yellow color on different growth media with 8-ascospored asci. The ascospores were ellipsoidal and ornamented. The anamorphic state was not observed. Molecular-genetic techniques identified the species. The present record is the first of the species in Argentina, pointing it as a tool to identify soils where cadaver decomposition occurs.

PALABRAS CLAVE
Talaromyces udagawae;
Hongos de suelo;
Descomposición cadavérica;

Primer registro de *Talaromyces udagawae* relacionado con cuerpos humanos en descomposición, en suelo de la Argentina

Resumen Se describen e ilustran las características morfológicas de *Talaromyces udagawae* Stolk y Samson. Se aisló el estado teleomórfico de este hongo Ascomycota de suelo obtenido en la provincia de Buenos Aires (Argentina), por debajo de un cadáver humano en avanzado

* Corresponding author.
E-mail address: ctranchida@conicet.gov.ar (M.C. Tranchida).

http://dx.doi.org/10.1016/j.ram.2015.10.002

Ascomycota;
Potencial forense

estado de descomposición. Las muestras de suelo fueron analizadas mediante lavado, dilución seriada y cámaras húmedas, técnicas ampliamente usadas para el estudio de hongos de suelo. *T. udagawae* formó colonias muy restringidas de color amarillo brillante en diferentes medios de cultivo, con ascos con 8 ascosporas. Las ascosporas eran elipsoidales y ornamentadas. No fue hallado el estado anamórfico. La especie también fue identificada mediante técnicas moleculares. El presente registro es el primero de la especie en la Argentina y el único que la postula como herramienta para identificar suelos donde ocurre una descomposición cadavérica.

Introduction

The genus *Talaromyces* was established by Chester Ray Benjamin in 1955. The genera mentioned comprise teleomorphic states of *Penicillium*, the species assigned to their corresponding anamorph. *Talaromyces* is characterized by soft, discrete, or confluent globose-to-subglobose surface ascomata of indeterminate growth. Ascomatal coverings vary from scanty to dense and consist of a network of hyphae that may range from very loosely textured to closely knit, but usually surrounded by further generally encrusted growing hyphae that are straight or twisted depending on the species. According to Stolk and Samson[13], ascomatal initials may have various shapes. Asci are short-lived, 4-to-6- or 8-spored, globose to subglobose or slightly ellipsoidal, and borne in chains. Ascospores are likewise globose or ellipsoidal; smooth or showing various ornamentations; and yellow, though rarely becoming reddish[13].

Talaromyces species have been recorded worldwide as being saprophytes utilizing various substrates, and especially organic materials undergoing slow decomposition or rotting wood, in different types of soils. Species such as *Talaromyces stipitatus* (Thom) Bernjamin, *Talaromyces trachyspermus* (Shear) Stolk and Samson, and *Talaromyces flavus* (Klocker) Stolk and Samson are able to degrade cellulose, protein, and keratin; and Eliades *et al.*[8] measured the cellulase, keratinase, and protease activity of *T. stipitatus*, *T. trachyspermus*, and *T. flavus*; *T. stipitatus* also possessed β-glucosidase activity.

The aim of the investigation reported here was to describe and illustrate the presence of *Talaromyces udagawae* in soil taken from beneath a human body in an advanced state of decomposition and to isolate and identify this fungus with a view to its potential forensic use.

This species has been previously described by Stolk and Samson (1972)[13] in relation to decaying wood in soil. The present report, however, constitutes the first documentation of *T. udagawae* in Argentina and the first instance of an association of this species with cadaver decomposition.

Materials and methods

T. udagawae was isolated from a soil sample taken from beneath a human cadaver at the moment it was discovered and removed. The person, whose remains were found by the Buenos Aires province police, had been reported missing 24 days before being discovered. The decaying remains were found in Buenos Aires province (−34°21′41.69″ S; 60°5′22.99″ W), Argentina with the skull already reduced to a skeleton and the arms and legs in a state of advanced decomposition. The site contained temporary puddles from a previous rain. The predominant vegetation was herbaceous, being sorghum the most common plant found in the area. Climatologic data obtained from a government weather station located 60 km away from the site showed that the mean ambient temperature from the time of disappearance to the discovery of the remains had ranged from 15.3 to 24.1 °C, while the average precipitation had ranged between 0 and 41 mm. Soil pH was 5.7 (it corresponds to acid soils of Buenos Aires province)[15].

A sample (composed of random subsamples) of soil in contact with the remains and another sample taken 15 m away (control sample) were collected with a sterile spoon at the time of the discovery and transported to the laboratory in hermetic plastic bags. In the laboratory, the soil specimens were processed in accordance with the following steps as detailed in Tranchida et al.[15] Soil washing was done as described by Parkinson and Williams[10], and serial dilutions were made following the Warcup's method[16]. The moist-chamber incubation was carried out as proposed by Elíades et al.[7]

The fungal morphologic structures were observed by optical microscopy, after lactophenol cotton-blue staining[14]. The ultrastructure of the fungal isolate was examined under a Jeol JSM-6390 scanning electron microscope (JSM-6390LV, Jeol, Akishima, Tokyo, Japan) in the Electronic Microscope Service at the Museum of Natural Sciences of La Plata. The morphology of the colonies was evaluated on malta-extract agar (MEA) (Difco™, USA), potato-dextrose agar (PDA) (Britania S.A., Argentina), and corn-meal–yeast agar (CMYA) (BBL™, USA).

For molecular-genetic identification, the fungal DNA was extracted according to Stenglein and Balatti[12]. The ribosomal DNA internal transcribed spacer (rDNA-ITS) region was amplified by the polymerase-chain reaction (PCR) in an XP thermal cycler (Bioer Technology Co, Hangzhou, China) with primer pairs ITS5 (5′-GGA AGT AAA AGT CGT AAC AAG G-3′) and ITS4 (5′-TCC TCC GCT TAT TGA TAT GC-3′)[18]. The success of the amplification was confirmed by gel electrophoresis using 1.5% (w/v) agarose gels containing GelRed TM (Biotium Inc., CA, USA) at 90 V in 1× Trisborate-EDTA