

Familial White Piedra Caused by *Trichosporon inkin*

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Abstract

White piedra is a superficial, chronic, asymptomatic mycoses caused by yeast fungi of the genus *Trichosporon*. It affects the hair, especially of the head, less frequently of the pubis, perineum, armpit, beard, mustache, eyebrows and eyelashes, and is characterized by the formation of soft nodules or fungal clusters. It affects all age groups and both sexes, predominantly women. Diagnosis is made by direct examination of the affected hair and culture on Sabouraud dextrose agar. The identification of species occurs through more specific identification procedures, such as mass spectrometry (MALDI-TOF) and PCR). The objective of this work is to report two cases of familial white piedra caused by *T. inkin* identified by PCR.

Keywords

Familial White Piedra, *Trichosporon inkin*, *Trichosporon* sp., Superficial Mycosis, Polymerase Chain Reaction (PCR)

1. Introduction

White piedra is a superficial, chronic, and asymptomatic mycosis, caused by yeast-like fungi belonging to the genus *Trichosporon*. Involvement of the hair shaft occurs, mainly head hairs and less frequently on public hairs, perineum, axilla, beard, moustache, eyebrows, and eyelashes. White piedra is characterized by the presence of nodular, soft, and whitish fungal masses. All age groups and both genders are involved, mainly women. Traditionally diagnosis is by microscopic observation of hair nodules, dermoscopy and culture on Sabouraud dex-

trose agar. Identification of causal fungi is determined by biochemical tests, currently, another more specific identification procedures such as mass spectrometry (MALDI-TOF) and molecular biology (PCR). Therapy mainly consists topical (shampoo) and systemic azolic derivates [1] [2].

We do not know its magnitude (incidence, prevalence), geographic distribution and in members of the same family, only by case reports or systematic reviews, it is not mandatory notification in the health sector, it is frequently confused and managed as head lice [3] [4] [5]. We present 2 cases in a family diagnosed by molecular biology

2. Case Report

Case 1

A 7-year-old female, a resident of Acapulco, Guerrero, she has an 8-month history with whitish structures in her hair, was treated treatment for head lice without improvement.

Examination revealed a normal-looking scalp, abundant, long hair with white nodules were identified throughout the hair, with a predominance in the occipital region (Figure 1(A)), which were better observed with wet hair. To the touch, the concretions are rough, soft consistency.

Case 2

A 52-year-old female, grandmother of the first case, with diabetes mellitus 2, upon examination, numerous concretions of a soft consistency adhered to the hair (**Figure 1(B**)), onset 5 months ago accompanied by itching in the occipital region, without previous treatment.

The time interval between cases was 6 months, history of long hair, habit of tying it up wet and living in the same house.

Clinically, it corresponds to white piedra, and samples of parasitized hair were



Figure 1. Numerous white piedra lesions along the hair shaft in both cases.

taken. A direct examination was performed with 20% potassium hydroxide, and abundant yeasts were observed surrounding the hair (Figure 2(A) $40\times$), and closer up conglomerates of round or slightly polymorphic yeast cells (Figure 2(B) $100\times$).

Cultures were performed on Sabouraud dextrose agar without antibiotics, developing whitish colonies, rough surfaces and soft consistency (**Figure 2(C)**) and microscopic examination with lactophenol blue (Lacto-phenol[®] Delasco, Council Bluffs) revealed septate hyphae and numerous globular cells (**Figure 2(D)**).

Based on history clinic, direct examination of the parasitized hair and phenotypic characteristics of the culture, both isolates were identified as *Trichosporon* sp. To determine the involved species, a polymerase chain reaction test followed by sequencing was performed.

Molecular Identification. DNA extraction. From a monosporic culture aged seven days on SDA, fungal mass was harvested by filtration and macerated in the presence of liquid nitrogen. After the Exgene Plant SV kit (GeneAll Biotechnology, Germany) was used following the manufacturer's instructions. Quality of DNA was verified in a 0.8% agarose gel and quantified by spectrometry (Epoch Bio Tek Instruments). The PCR was targetting the ITS region according to White *et al.* [6] conditions. The oligonucleotides used were: ITS1: 5' tcc gta ggt gaa cct gcg g - 3'; ITS4: 5' tcc tcc gct tat tga tat gc - 3'. Mix reaction in a 50 µL final volumen contained: buffer 1×; 2 mM MgCl₂; 0.25 mM dNTP's (Invitrogen); 2 µM oligonucleotides; 3.5 U polymerase taq recombinant (ThermoScientific); 100 ng DNA. Cycling temperatures were 96°C for 5 min; 30 cycles of de 94°C for 30 s, 58°C for 30 s, 72°C for 30 s; final extension 72°C for 5 min. Isolate from the first case gave an amplicon of about 600 bp and from the second one of 800 bp.

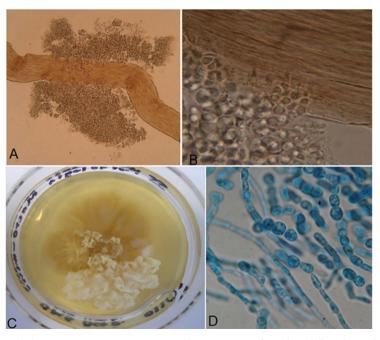


Figure 2. (A) Microscopic examination with KOH 20%, fungal nodules along the hair shaft 40×; (B) Magnification 100×; (C) Culture on Sabouraud dextrose agar, cerebriform colony; (D) Septate hyphae, abundant globular cells lactophenol blue 100×).

Both products were purified using the DNA Clean & Concentrator-5 kit (Zymo Research) (Figure 3). PCR products were sent to sequencing in two senses to Molecular Biology Laboratory from Faculty of Higher Studies, Cuautitlan Campus, UNAM, using the ABI3130/3130×l Genetic Analyzer (Applied biosystems/Hitachi, Foster city, CA, USA. Sequences were compared in the GenBank database, and both showed 100% of identity with *Trichosporon inkin* (isolates CDCF 2096, CDCF2090, CDCF 2102, SCAU167; Sequence IDs: MN809445.1, MN809442.1, MN809448.1, MF135530.1, respectively).

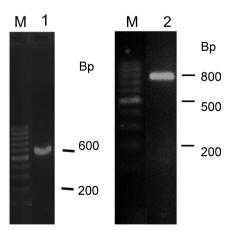
The treatment was to wash the hair daily with 2% ketoconazole shampoo, observing improvement after one month, followed by three months, with no relapses.

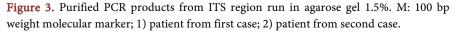
3. Discussion

White piedra was first described as a fungal disease by Beigel in 1865, who isolated the fungus from a wig, but Rabenhorst gave it the name *Pleurotus beigelii* [7]. In 1890 Behrend described another case affecting the beard and the isolated microorganism received the name of *Trichosporon ovoides*. In 1902 Vuillemin named all species so far described as *T. beigelii* [8]. In 1994, Gueho *et al.* rearranged the genus *Trichosporon* in six species: *T. asahii, T. cutaneum, T. asteroides, T. mucoides, T. inkin* and *T. ovoides*. [9]. Currently, based on several phenotypic, biochemical, and molecular studies, there are about 51 *Trichosporon* species, from which 16 have clinical relevance [8].

A systematic literature review was conducted using PubMed, Embase, Academic, SciELO, Google and Virtual Library to 6 June 2022 using the words "white piedra", "white piedra scalp", "white piedra mycoses", "piedra blanca cuero cabelludo", "couro cabeludo de pedra branca".

A total of 9 articles reporting 14 cases of white piedra of the hair from 2004 to date [2] [3] [4] [5] [10] [11] [12] [13] [14], with the following distribution by country: Brazil 8, United States of America 2, India 2, Peru 1 Qatar. From available data in this review 8 were adults (57%) with an age range of 20 to 36 years 7





females and 1 male, in children 6 (43%) range of 3 to 9 years, 5 female and 1 male; the causal agent in 8 was *T. inkin* and 1 by *T. ovoides*.

The patients were treated with topical azoles and/or systemic triazoles. There is no consensus on the treatment of this mycosis, El Attar *et al.* [15] compared three therapy modalities: 1) topical azoles, 2) topical and oral azole; and 3) topical azole and oral terbinafine. The percentages of cure were in 1) 50% in 12 weeks; 2) 75% in four weeks; 3) 60% in eight weeks, the combination of topical and oral azolic drugs showed to be the best therapy.

In 2012 in Brazil Richini *et al.* [5] published the case of a family with white piedra in members of the same family (mother and 2 children) diagnosed by molecular biology, our case in grandmother and granddaughter is the second and the first in Mexico.

The differential diagnosis of white piedra includes black piedra, trichobacteriosis, pediculosis capitis and hair casts [2].

4. Conclusions

Case reports of familial white piedra are few, this is the second by *Trichosporon inkin* diagnosed by molecular biology in Latin America and the first in Mexico.

It is important to sensitize the clinical doctors that when faced with a suspected case of head lice, one of the differential diagnoses is white stone and that molecular biology is the gold standard for identifying the etiological agents.

Ethics Declaration

Written informed consent was obtained from the mother of the minor and the grandmother for the publication of the cases and clinical images.

Authors' Contribution

Marina Romero-Navarrete, Aureliano Castillo-Solana: Design, manuscript writing, clinical study of the patients and sampling of the infected hair.

Francisca Hernandez-Hernandez, Brianda Stephanie Herrera-Ramírez, Erika Córdova-Martínez: Molecular phenotypic study and data analysis.

Roberto Arenas: Revision of technical aspect of the manuscript.

All authors read and approved this final version of the manuscript.

Declaration of the Information Availability

The data that support the findings of this report are available in Pubmed.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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