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Dermatomycoses Due to *Nannizzia praecox* (Formerly *Microsporum praecox*) in Germany: Case Reports and Review of the Literature

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Abstract *Nannizzia praecox*, formerly known as *Microsporum praecox*, is a geophilic dermatophyte. Up to now 31 cases of human tinea have been reported in the literature, most of them with an inflammatory course. Three recent cases diagnosed in Germany within 1 year suggest that the fungus might be a more common cause of human dermatophytosis than reported so far. This might be based on the fact that *N. praecox* is often found in an equine environment and that horse riding is becoming more popular recently.

Keywords *Nannizzia praecox* · *Microsporum praecox* · New taxonomy · Geophilic dermatophyte · Horse · Inflammatory tinea

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Introduction

Most recently, based on a compilation of molecular data the taxonomy of the dermatophytes has been revised. Nearly all anthropophilic dermatophytes are now classified in the genera *Trichophyton* and *Epidermophyton*, along with some zoophilic species that regularly infect humans. *Microsporum* is now restricted to some species around *M. canis*, while the geophilic species and zoophilic species that are more remote from the human–environment are divided over *Arthroderma*, *Lophophyton* and *Nannizzia* [1]. But, also in these genera, there are some dermatophytes of importance in clinical medicine. *Nannizzia (N.) praecox* (Padhye, Ajello and McGinnis) Graeser and de Hoog, *comb. nov.*, formerly known as *Microsporum praecox*, is a geophilic dermatophyte present in soil and equine environments (saddles, straw, stables, etc.). It is rarely reported as a cause of human tinea, particularly after contact with horses. *N. praecox* can be isolated from horse hair in the absence of clinical lesions [2–17].

The following article summarizes three recent cases of inflammatory *N. praecox* infections in Germany and gives a review of the literature.

Case Reports

1. A 15-year-old female horse rider suffered from scaling, itching and small blistering at her right

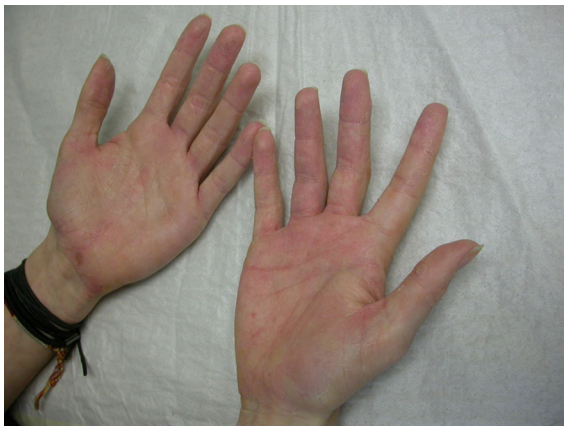


Fig. 1 15-year-old girl (patient 1) with erythema, desquamation and tiny blisters on both hands. Tinea manus due to *Nannizzia praecox*

hand with subsequent spreading to the left one. Initially, under the preliminary diagnosis of allergic contact dermatitis a topical and oral therapy with glucocorticosteroids was started. At that time, an erythema with desquamation and tiny blisters on both hands was complicated by a severe itch (Fig. 1). Direct microscopy was positive, and *N. praecox* was found in culture. The macro- and micromorphological identification was confirmed by sequencing of internal transcribed spacer region (ITS). Brush cultures of 10 horses in the riding stable remained negative. The lesions cleared completely after a systemic antifungal treatment with terbinafine 250 mg/d for 21 days.

2. A 10-year-old girl suffered from tinea corporis with erythematosquamous and centrifugal growing, sparse itching lesions of her right lower arm. Source of infection was horses at a horse-riding centre where the girl regularly was riding. Fluorescence optical Blancophor® preparation from skin scrapings revealed fungal hyphae. On Sabouraud's dextrose agar, the fast growing dermatophyte formed flat, peripheral radiating and convolved colonies with white, slightly yellowish to beige-brown-stained granular and powdery surface (Fig. 2a, b). The reverse side of the colonies was smooth with luminous yellow colour (Fig. 2c). In the subculture, according to Ito and Refai the strain developed flat colonies with white–yellowish and granular surface with a conspicuous annular morphology (Fig. 2d). Microscopically, a multitude of thin-walled spindle-shaped and echinulate (with

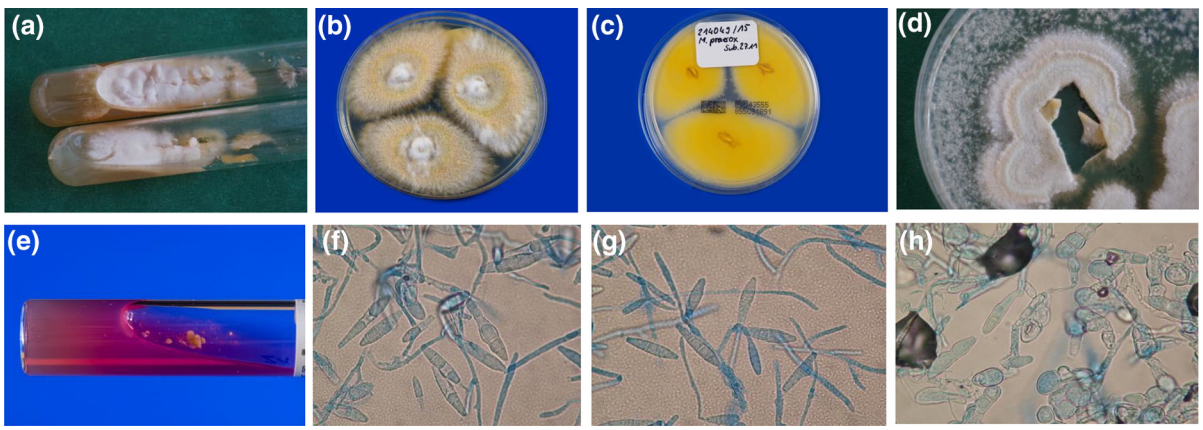


Fig. 2 **a** Primary culture of *Nannizzia praecox* (strain 2) on Sabouraud's dextrose agar. On tubes with tilted agar from skin scrapings developed flat colonies with white powdery and granular surface. **b** Three-week-old subculture of strain 2 on Sabouraud's dextrose agar without cycloheximide. Slightly yellowish colonies showing with brownish pigmentation of the granular surface. **c** The reverse side of the colonies showed brightening yellow pigmentation. **d** Subculture according to Ito and Refai: *Nannizzia praecox* develops flat colonies with white-yellowish and granular surface with conspicuous annular morphology. **e** Urease activity was positive at Christensen-Agar (Becton Dickinson, Heidelberg, Germany. **f** Microscopically, a multitude of row-walled, spiny, echinulate (with small spins) and lanceolate macroconidia were observed. Spearhead-like in the centre raised macroconidia are typical for this dermatophyte. Lactophenol cotton blue preparation. **g** The spindle-like macroconidia form three to six or seven cross septae. Lactophenol cotton blue preparation. **h** Round and irregular formed chlamydospores—occurring single or in chains—were found in a hugh scale. Lactophenol cotton blue preparation. (Color figure online)

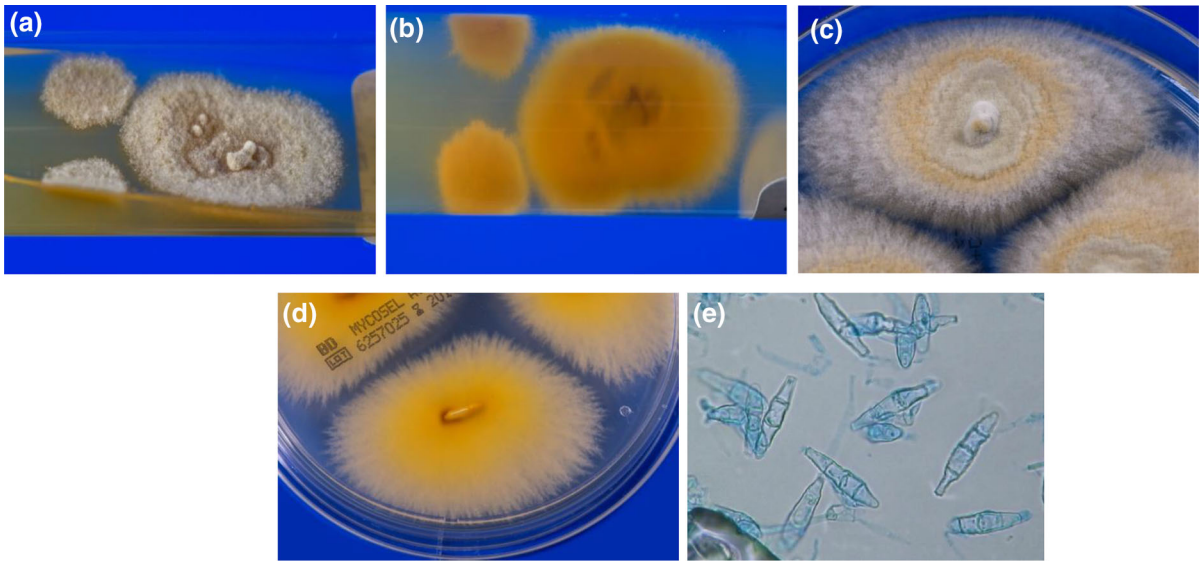


Fig. 3 **a** Primary culture of *Nannizzia praecox* (strain 3) on Sabouraud's dextrose agar. On tubes with tilted agar developed flat colonies with white to beige powdery and granular surface. **b** Yellow-stained reverse side of the primary culture. **c** Three-week-old subculture of strain 3 on Sabouraud's dextrose agar without cycloheximide. White granular colonies with annular brownish pigmentation. **d** The reverse side of the colonies showed brightening yellow pigmentation. **e** Microscopically, a multitude of row-walled, spiny and lanceolate macroconidia appeared. Lactophenol cotton blue preparation. (Color figure online)

small spins) and lanceolate macroconidia appeared (Fig. 2e–g). Urease activity was positive. First, sequence analysis of the ribosomal ITS region (18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA) and of the translation elongation factor 1-alpha (TEF 1-α) gene revealed the dermatophyte species *M. praecox*. Topical treatment was done using ciclopirox olamine cream.

3. A 17-year-old girl presented with a scaly plaque on the right lower leg existing since 2 weeks. There was no objective evidence for an animal source of infection. From skin scrapings, *M. praecox* was isolated (Fig. 3a–e). Suspicion diagnosis of *M. praecox* was based on results of matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) analysis of the strain. Species identification has been confirmed by sequencing the ITS region of the rDNA. Topical treatment by fusidic acid ointment was ineffective. Clotrimazole 1% plus triamcinolone in basic ointment lead to improvement in the skin lesion.

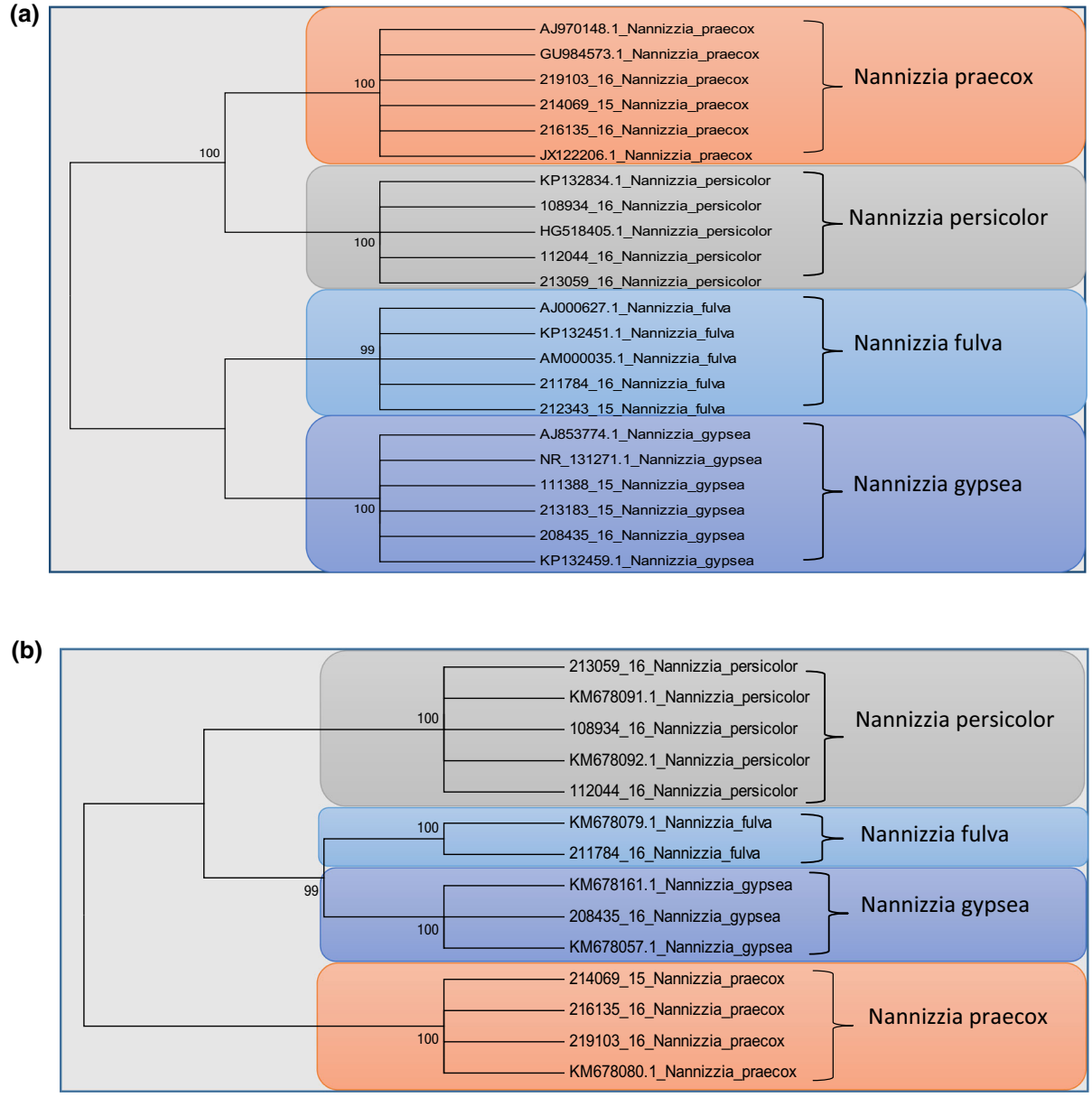
Table 1 summarizes the clinical and mycological data of all three *N. praecox* infections.

Identification of *N. praecox* by Morphologic and Physiologic Parameters

On Sabouraud's dextrose agar, the fast growing dermatophyte formed flat, peripheral radiating and convolved colonies with white, slightly yellowish to

Table 1 Clinical and mycological data of all three cases, caused by *N. praecox*

Case No.	Patients	Clinical presentation	Direct microscopy	Clinical course and outcome
1	15-year-old girl	Tinea manuum dyshidrotic	Positive	Terbinafine 250 mg/d 21 d; complete healing
2	10-year-old girl	Lesions right forearm	Positive	Ciclopirox olamine cream
3	17-year-old girl	Single lesion lower leg	n.d.	1% clotrimazole with triamcinolone in basis ointment



beige-brown-stained granular and powdery surface (Figs. 2a, b, d, 3a, c). The reverse side of the colonies is smooth with luminous yellow colour (Figs. 2c, 3b, d). Microscopically, a multitude of thin-walled spindle-shaped and echinulate (with small spins) and lanceolate macroconidia appeared [18, 19]. The small based macroconidia are raised in the middle and end part, however, pointy at the end ("spear-head") (Figs. 2f-h, 3e). They are 6–9 celled and up to 65 × 9 µm. The small piriform microconidia, when present, have an orthotropic arrangement. Chlamydospores are also formed. Urease activity is positive (Fig. 2e).

◀ **Fig. 4** *Nannizzia praecox*: Phylogenetic tree (dendrogram) based on sequencing of the ITS and TEF 1-α gene region, Mega5, statistical method: neighbour-joining, 1000 bootstrap replicates, bootstrap branch supports above 70%. NCBI—National Center for Biotechnology Information (NCBI), Bethesda, Maryland; CBS—Centraalbureau voor Schimmelcultures, Utrecht, The Netherlands; DSM—German Collection of Microorganisms and Cell Cultures (Deutsche Sammlung von Mikroorganismen und Zellkulturen) Braunschweig, Germany. **a** Sequencing of the ITS region, used dermatophyte strains. *Nannizzia praecox:* AJ970148.1—NCBI database, CBS 468.74; GU984573.1—NCBI database, 009.10 (Austria); 219101_16—University Clinics Gießen and Marburg, Prof. P. Mayser, Gießen, DSM strain number 109798; 214069_15—Laboratory for Medical Microbiology, Mölbis; DSM strain number 103445; 216135_16—Laboratory Dr. Stein + Colleagues, Mönchengladbach, DSM strain number 104497; JX122206.1—NCBI database, bM 137 (Switzerland). *Nannizzia persicolor:* KP132834.1—NCBI database, ISHAM-ITS: MITS2651 (accession number); 108934_16—Laboratory for Medical Microbiology, Mölbis; HG518405.1—NCBI database, CCF<CZE>: 4541; 112044_16—Laboratory for Medical Microbiology, Mölbis; 213059_16—Laboratory for Medical Microbiology, Mölbis. *Nannizzia fulva:* AJ00627.1—NCBI, CBS 287.55 (accession number); KP132451.1—NCBI ISHAM-ITS: MITS2051 (accession number); AM000035.1—NCBI, CBS 529.71 (accession number); 211784_16—Laboratory for Medical Microbiology, Mölbis; 212343_15—Laboratory for Medical Microbiology, Mölbis. *Nannizzia gypsea:* AJ853774.1—NCBI database, ISHAM-ITS: ID MITS2061 (accession number); NR_13127.1—NCBI database, CBS database strain number 258.61; 111388_15—Laboratory for Medical Microbiology, Mölbis; 213183_15—Laboratory for Medical Microbiology, Mölbis; 208435_16—Laboratory for Medical Microbiology, Mölbis; KP132459.1—NCBI database, ISHAM-ITS database MITS2060 (accession number). **b** Sequencing of the TEF 1-α gene region. *Nannizzia persicolor:* 108934_16—Laboratory for Medical Microbiology, Mölbis; 213059_16—Laboratory For Medical Microbiology, Mölbis; KM678092.1—NCBI database, CBS strain number 422.74; 112044_16—Laboratory for Medical Microbiology, Mölbis; KM678091.1—NCBI database, CBS strain number 421.74. *Nannizzia fulva:* KM678079.1—NCBI database, CBS strain number 287.55; 211784_16—Laboratory for Medical Microbiology, Mölbis. *Nannizzia gypsea:* 208435_16—Laboratory for Medical Microbiology, Mölbis; KM678161.1—NCBI database, CBS strain number 130820; KM678057.1—NCBI database, IFO 8228 (accession number). *Nannizzia praecox:* 214069_15—Laboratory for Medical Microbiology, Mölbis; DSM strain number 103445; 216135_16—Laboratory Dr. Stein + Colleagues, Mönchengladbach; DSM strain number 104497; 219103_16—University Clinics Gießen and Marburg, Prof. P. Mayser, Gießen; DSM strain number 109798; KM678080.1—NCBI database, CBS strain number 288.55

Molecular Identification

Molecular testing for dermatophyte DNA of *Microsporum canis* with a previously described PCR–ELISA using a specific primer target gene topoisomerase II was negative [20, 21].

Identification of *M. praecox* by DNA Sequence Analysis

The sequence analysis of the ITS of the ribosomal DNA and of the TEF 1-α gene was performed for the three *N. praecox* strains [22]. The respective DNA fragment was amplified with the universal fungal primer pair V9D and LSU266. Sequences of the amplification products were determined by Sanger sequencing using these oligonucleotide primers. For species determination, sequences were compared to the National Center for Biotechnology (NCBI) database, Bethesda, Maryland, using standard nucleotide blast.

Sequence analysis of the ribosomal ITS region (18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA) and of the TEF 1-α gene revealed the dermatophyte species *N. praecox* (formerly *M. praecox*). Comparison was made with the known sequences of in the NCBI-deposited *N. praecox* strains (*M. praecox* AJ970148.1 [3] and *M. praecox/N. praecox* U984573.1 [4]). Moreover, DNA sequences of the here described three wild strains of *N. praecox* matched 99% with the *M. praecox/N. praecox* isolate JX122206.1 [6]. Sequence analysis using the TEF 1-α gene revealed a 100% concordance of all three wild strains with at the NCBI-deposited sequences of *M. praecox/N. praecox* [10].

The phylogenetic tree or the dendrogram of *N. praecox* and further *Nannizzia* species is shown in Fig. 4. *N. praecox*—in the upper part of the dendrogram—can be clearly distinguished from *N. persicolor*, *N. fulva* and *N. gypsea* (bootstrap values were over 90%) [23]. Sequencing of the ITS region (Fig. 4a) and of TEF 1-α gene (Fig. 4b) of the DNA allowed clear discrimination of *N. praecox* from any other *Nannizzia* species.

Table 2 Characterization of the three *N. praecox* isolates: deposition of culture material at culture collection and gene sequences at DNA databases

No	Date of isolation laboratory number	Fungal culture (DSMZ)	ITS gene (NCBI) nucleotide sequence accession number	TEF 1-α gene (NCBI) nucleotide sequence accession number	ITS (ISHAM) nucleotide sequence accession number	References
1	1/2011 219103/16	DSM 104498	KY769914	KY769913	MITS3917	[16]
2	11/2015 214069/15	DSM 103445	KX866686	KY769910	MITS3918	[17]
3	7/2016 216135/16	DSM 104497	KY769912	KY769911	MITS3919	–

DSMZ German Collection of Microorganisms and Cell Cultures (Deutsche Sammlung von Mikroorganismen und Zellkulturen), *NCBI* National Center for Biotechnology Information, *ISHAM* International Society for Human and Animal Mycology

All three isolates (culture material) were deposited at the German Collection of Microorganisms and Cell Cultures (Deutsche Sammlung von Mikroorganismen und Zellkulturen, DSMZ, https://www.dsmz.de/) at Braunschweig, Germany (Table 2).

The DNA sequences of the three strains have been deposited at the NCBI database (ITS and TEF 1-α gene) and additional at the database of the International Society for Human and Animal Mycology (ISHAM) (Table 2).

Discussion

Nannizzia (N.) praecox (Padhye, Ajello and McGinnis) Graeser and de Hoog, *comb. nov.*, formerly known as *M. praecox*, a geophilic dermatophyte, is present in soil and equine environments. It was described first in 1954 by Rivalier as *Sabouraudites praecox* [24]. In 1978, the nomenclature changed to *M. praecox* [25]. However, in the new classification published in 2017 this geophilic species is now classified in the genus *Nannizzia*, as it is more remote from the human–environment [1].

Since 1954, less than 40 cases (Table 3) of human dermatophytosis due to *N. praecox* have been described in the literature, mainly from France [3, 4, 7–14, 22], Belgium [6] and the USA [5, 12]. The culture morphology of *N. praecox*, however, is not very specific. The white, slightly yellowish to beige-brown-stained granular and powdery surface might resemble *T. mentagrophytes* at the first look. By microscopic features, e.g. by the shape of macroconidia, the geophilic species *N. gypsea* (formerly *M. gypseum*) and *N. fulva* (formerly *M. fulvum*) should be excluded. The hair perforation test may be a useful tool, as it is negative in case of *N. praecox*, but positive with *N. gypsea*. In contrast, urease activity is not very valid as it is positive for most of the *Microsporum* and *Nannizzia* species. Therefore, molecular methods are currently considered as reference for species identification.

For these problems of identification in particular, with conventional methods, *N. praecox* might be underestimated in clinical medicine underlined by the fact that we were able to detect three cases in Germany within a period of 5 years.

In humans, *N. praecox* can cause tinea corporis as well as tinea capitis, the majority of cases being related to contact with horses (Table 3). In our first case, however, we were not successful with the isolation of *N. praecox* direct from horses which could be asymptomatic carriers. The geophilic *N. praecox* therefore might be more associated with equine environments (saddles, straw, stables, etc.), but data are sparse (2).

In circumscribed lesions, topical antifungal therapy is often successful. As the infections by geophilic fungi are often very inflammatory and pruritic, topical combination therapy with topical steroids might be of advantage in the first days. Extended and/or high inflammatory lesions as well as tinea capitis may require systemic antifungal therapy with terbinafine, griseofulvin or one of the azoles.

However, as eczema is the main differential diagnosis single use of corticosteroids as in our first case should be avoided. To confirm or to rule out a diagnosis of tinea direct microscopy of skin scrapings

Table 3 Clinical features of 34 cases of *N. praecox* dermatophytosis reported in the literature. Adapted from [2]

Case No. (ref.)	Year	Geographic origin	Sex/age (years)	Localization	Contact with horses
1 (22)	1954	France	m/42	Wrist	NA
2 (3)	1968	France	f/40	Arm	NA
3 (3)	1972	France	f/23	Hand, wrist, forearm	NA
4 (3)	1977	France	f/15	Hand	NA
5 (5)	1977	USA	f/NA	NA	+
6 (4)	1979	France	f/23	Instep	+
7 (6)	NA	Belgium	f/13	Forearm	+
8 (6)	NA	Belgium	f/12	Instep	+
9 (6)	NA	Belgium	f/14	Forearm	+
10 (6)	NA	Belgium	f/35	Thigh	+
11 (6)	NA	Belgium	f/30	Elbow	+
12 (6)	NA	Belgium	f/18	Leg	+
13 (7)	NA	France	f/25	Achille's tendon	+
14 (8)	1985	France	f/50	Wrist	NA
15 (9)	1986	France	f/19	Buttock	+
16 (9)	1987	France	f/12	Ankle	+
17 (9)	1987	France	f/19	Instep	+
18 (11)	1976	France	f/12	Forearm	NA
19 (11)	1986	France	m/13	Hand	+
20 (11)	1988	France	f/18	Foot	+
21 (12)	1981	France	f/13	External malleolus	NA
22 (12)	1983	France	m/12	Instep	+
23 (12)	1987	France	f/10	Achille's tendon	+
24 (12)	1989	USA	f/30	Scalp	NA
25 (13)	1990	France	m/10	Instep	NA
26 (13)	1990	France	f/20	Instep	+
27 (14)	1992	France	f/18	Internal malleolus	+
28 (14)	1993	France	m/19	Wrist	+
29 (14)	1993	France	f/12	hand	+
30 (2)	2007	France	f/28	External malleolus	+
31 (15)		Austria			
32 (16)	2016	Germany	f/15	Hands	+
33 (17)	2017	Germany	f/10	Forearm	
34 PR	2017	Germany	f/17	Lower leg	

PR present report, *NA* not available

from the lesions is therefore an essential tool. For the identification of the aetiologic agent, phenotypic and genomic methods should be combined. If a direct or indirect contact to horses is reported in a case of dermatophytosis, *T. verrucosum*, *T. mentagrophytes*, *T. equinum*, *T. bullosum* and, in particular, *N. praecox* should be taken into consideration.

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Compliance with Ethical Standards

Conflict of interest The authors including co-authors declare that they have no conflict of interest.

Research Involving Human Participants and/or Animals All applicable international, national and/or institutional guidelines for the care and use of animals were followed. This article does not contain any studies with human participants performed by any of the authors. This article does not contain any studies with animals performed by any of the authors.

Informed Consent Informed consent was obtained from all individual participants included in the study.

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