Sir,

Tinea caused by *Trichophyton equinum* is currently a very rare infection in Europe. During the period 1951 to 1987 only one out of 4571 cases of tinea diagnosed in Santiago de Compostela, Spain, was attributed to *T. equinum* (tinea corporis) (1). In England a case of tinea capitis was reported in 1994 in a woman on systemic therapy with corticosteroids who went horse-riding (2). Another case was reported from Germany in 1998; a girl with tinea of the neck who had been infected by her pony (3). A racing horse was the source of infection with *T. equinum* in a case of onychomycosis reported from Finland in 1998 (4). Tinea of the eyebrows was acquired by another child through contact with a pony in Spain in 2001 (5). We report here on a child infected by *T. equinum* and show for the first time that sequencing can be used successfully in a clinical setting as a diagnostic tool to differentiate *T. equinum* from *T. tonsurans*.

**CASE REPORT**

A 5-year-old boy presented with a slowly enlarging hairless plaque on the back of his head that had been progressing for 2 weeks. In addition, some small erythematous scaling lesions had occurred on his body. His general health was good. The boy lived on a farm that bred horses. Some of the horses had previously had tinea, which had been treated successfully by a veterinary surgeon, but an identification of the causal dermatophyte had not been attempted. The boy had also had contact with dogs, cats and cattle. His siblings and playmates had no skin lesions.

On clinical examination an area of slightly erythematous and scaling skin with a diameter of approximately 5 cm was seen on the back of the head. The lesion was sharply demarcated and almost free of hair (Fig. 1). On the trunk and extremities several small distinct erythematous and scaling plaques were seen. The nuchal lymph nodes were slightly enlarged.

Hair plucked from the margin of the occipital lesion and scales collected from this site did not reveal fungi by direct microscopy (KOH). DNA was extracted from the scales and used for PCR; a positive result occurred with a pair of primers designed to detect the species of the *T. tonsurans* complex, but not with primers specific for the *T. rubrum* complex. In a first culture on Sabouraud agar a *Trichophyton* species was cultivated that was subsequently analysed further by morphological, physiological and genetic methods. Conventional methods as well as the subsequent sequence analysis of the ribosomal internal transcribed spacer (ITS) region finally allowed us to identify the strain unambiguously as *T. equinum*. Systemic therapy with griseofulvin (10 mg/kg body weight) plus topical application of ciclopirox resulted in complete healing and re-growth of hair after 3 months.

**DISCUSSION**

Identification of *T. equinum* is difficult, but epidemiologically and clinically important. Morphological criteria have to be complemented by physiological tests (3, 6). On Sabouraud agar our strain rapidly developed a flat colony with a granular surface, a radial border and a slightly yellow reverse side. Microscopically clavate and globose microconidia and sparse small thin- and smooth-walled macroconidia with few chambers were seen. As a characteristic feature of *T. equinum* (3, 6) no growth occurred on Trichophyton agars 1–4, 6 and 7, but supplementation with nicotinic acid (Trichophyton agar 5) yielded good growth. Tests for urease and hair perforation were positive. These results allowed identification of *T. equinum* (3, 6), but in consideration of the arguable synonymization of this species with *T. tonsurans* (7, 8) verification by genetic analysis was considered appropriate.

For this purpose DNA was extracted from culture material and used in analysis of the ITS region of the isolate. A single C/T substitution out of more than 600 base pairs (bp) sequenced within the first 30 bp of ITS-1 was detected that discriminates *T. equinum* from *T. tonsurans* (Tables I and II). This is a rather subtle dif-
Table I. Differentiation of *Trichophyton equinum* and *T. tonsurans* by use of the internal transcribed spacer (ITS)-sequence (655 bp) shown in the validated IDNS™ (Integrated Database Network System) database supplied by SmartGene (Zug, Switzerland). The clinical isolate shows 100% identity with the *T. equinum* reference strain (AF170458) in the database. In the column “Mismatches” the 1 bp difference from the *T. tonsurans* reference strain (AF170479) is documented. Similar sequences are found in the IDNS™ database.

<table>
<thead>
<tr>
<th>Dataset</th>
<th>Accession</th>
<th>Organism</th>
<th>Sequence length (bp)</th>
<th>Identities (bp)</th>
<th>Mismatches (bp)</th>
<th>Match length (bp)</th>
<th>Score</th>
</tr>
</thead>
<tbody>
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<td>Dermatophytes ITS</td>
<td>AF170458</td>
<td><em>Trichophyton equinum</em></td>
<td>845</td>
<td>655 (100%)</td>
<td>0</td>
<td>655</td>
<td>1298</td>
</tr>
<tr>
<td>Dermatophytes ITS</td>
<td>AF170479</td>
<td><em>Trichophyton tonsurans</em></td>
<td>845</td>
<td>654 (99.8%)</td>
<td>1</td>
<td>655</td>
<td>1291</td>
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<tr>
<td>Dermatophytes ITS</td>
<td>Z98014</td>
<td><em>Arthroderma vanbreuseghemii</em></td>
<td>717</td>
<td>646 (98.6%)</td>
<td>9</td>
<td>655</td>
<td>1215</td>
</tr>
</tbody>
</table>

Table II. Internal transcribed spacer (ITS)-sequence alignment of the first 60 bp of the ITS-1 of the clinical isolate (upper row) and the *Trichophyton tonsurans* reference strain (lower row) by use of the freely available and validated database for identification of dermatophytes (www.cbs.knaw.nl) supplied by the Centraalbureau voor Schimmelcultures (CBS, Utrecht, The Netherlands).

gatattagcccagagccgagagctcggccceccagatatgggcaaacgtcgctca
x
atattagccagagccgagagctcggccceccagatatgggcaaacgtcgctca

The C/T substitution that discriminates both species is marked by an “x” (26th base pair) between the rows.