Sir,

The epidemiology of dermatophyte infections is changing due to immigration, travel and socioeconomic development. At the beginning of the 20th century *Trichophyton violaceum* and *T. tonsurans* caused approximately 45% of the dermatophyte infections in Denmark (1). These species apparently disappeared in the 1930s, but are once more causing infections in Denmark (1). In the other Nordic countries a similar re-emergence of endemic anthropophilic species has been observed, especially of *T. violaceum*, *Microsporum audouinii*, *T. soudanense* and *T. tonsurans* among immigrants with tinea capitis from endemic areas, e.g. Africa and the Middle East (2, 3).

The present laboratory-based study focuses on the species distribution of dermatophyte infections in Denmark within a 10-year-period in order to present an updated review on local epidemiology and clarify possible trends in aetiology.

**MATERIALS AND METHODS**

The study is a retrospective review of laboratory data of 24,752 skin, hair and nail specimens obtained from 21,273 patients and examined in 2003 at the mycology laboratory, Statens Serum Institut, Copenhagen, Denmark. These data are compared with similar data from 1993 performed at the same laboratory and extract of a national report (4). Statens Serum Institut receives patients’ skin specimens from general practitioners, dermatologists and hospitals from the whole country. The majority of specimens were submitted from general practitioners (49.6%) and dermatologists in private practice (49.1%), whereas only 1% was submitted from hospitals in 2003. The catchment area was unchanged over the study period. Only samples with information about body sites were included in the calculation of total positive rates. *T. terrestre* was isolated from 36 specimens. It is not regarded as a pathogen and was therefore included among the negative samples in the calculations and tables.

Mycological examinations were performed according to national and international standards and included direct microscopy and culture. Direct microscopy was performed in 30% potassium hydroxide using a light microscope (phase contrast). Cultures were performed using Sabouraud-glucose-agar (SGA) supplemented with cycloheximide and chloramphenicol (SSI, Diagnostika, Hillerød, Denmark) and incubation at 25°C for up to 4 weeks. Identification of species was carried out according to the micro- and macro-characteristics of the cultures (5). No differentiation between *T. mentagrophytes* and *T. interdigitale* was performed; they were thus recorded as *T. mentagrophytes*. Dermatophytes, which were not identified to species level, were not included in the report in 1993 (4). In order to avoid bias when comparing the data, 72 dermatophyte isolates not further identified were similarly excluded from the calculation of species distribution in 2003.

The data from 1993 were available only in tabular form, containing all cultured specimens, and this defines the format of the data presentation in this paper. Out of a total of 24,752 cultured specimens from 2003, 6251 (25%) came from patients who contributed two or more specimens. This implies a lack of stochastic independence that makes it impossible to make a statistically valid comparison between the relative frequencies from 1993 and 2003. The results are therefore presented without (obviously biased) statistical *p*-values.

**RESULTS**

The total number of specimens examined by culture increased by 19.2% in the study period (20,772 in 1993 vs. 24,752 in 2003), mainly due to a four-fold increase in the number of nail specimens, whereas the number of skin samples decreased remarkably. The overall positive rate remained relatively constant, at 21.6–23.2%, although a decrease was observed for hair specimens and a slight increase was observed for nail and skin specimens. Concomitant presence in samples of two dermatophyte species was detected in 49 specimens in 2003 (nail (*n* = 22), skin from feet (*n* = 21), skin unspecified (*n* = 2) and unknown body site (*n* = 4)).

Overall, *T. rubrum* was the predominant species (Table I). The proportion of isolates of *T. mentagrophytes*, *M. canis* or *Epidermophyton floccosum* decreased from 35.8% to 19.8%, whereas that of endemic anthropophilic infections, such as *T. violaceum*, *T. tonsurans* and *M. audouinii*, increased from 1.0% to 2.8% of all the isolates (Table I). The most common clinical presentation was tinea capitis (46% of cases) followed by skin infections (20%), tinea unguium (18%) and tinea pedis (5%). Information on the origin of the specimen was missing in 10% of cases.

The proportion of samples (out of the total number of samples) which derived from hair was unchanged during the 10-year-period, but the absolute number of hair specimens increased by 16% and the positive rate declined by 7.5% (Table II). *M. canis* was the predominant species, though the frequency decreased. On the other hand, infections caused by *T. violaceum* tripled, and thus the proportion of infections caused by dermatophytes other than *Microsporum* species increased from 16.0% to 39.9% (Table II). *T. violaceum* and *M. audouinii* were almost exclusively isolated from hair infections, accounting for 37.8% of these in 2003 (Table II).

The number of nail specimens received for analysis increased four-fold, with a slightly higher positive

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rate in 2003 (Table II). *T. rubrum* was by far the most frequently isolated pathogen and was increasing over the 10-year-period. *T. mentagrophytes* was the second most frequently cultured dermatophyte and surprisingly, 50% of the *T. tonsurans* specimens were isolated from nails. The less terbinafine-susceptible *Microsporum* species was rarely cultured from nail specimens.

The total number of skin samples from the feet received for analysis declined by 34%. As in nails, *T. rubrum* was the predominant and increasing pathogen followed by *T. mentagrophytes*. Although declining, *T. mentagrophytes* was more than twice as frequent in skin from feet as in nails (Table II).

The total number of samples analyzed from skin locations other than feet, and from skin without body site information declined by 46.7% over the 10-year period (Table II).

### DISCUSSION

The main findings of this 10-year study were: (i) a decrease in the proportion of *M. canis*, but an increase in anthropophilic dermatophytes (mainly *T. violaceum*) in hair samples; (ii) a remarkable increase in the number of nail samples and positive nails with *T. rubrum*, which currently is the cause of almost 90% of nail infections; and (iii) a 50% reduction in the number of skin samples received for mycological examination. Even though *M. canis* is still the major pathogen of tinea capitis in Denmark, anthropophilic infections are emerging, as seen in other European countries (6). It is therefore increasingly important to examine all close contacts in order to detect asymptomatic carriers and to avoid further transmission (7). The four-fold increase in number of nail specimens received for analysis was not associated with a decrease in positive rate. This suggests that the prevalence of tinea unguium, and not just sampling frequency, has increased. This could reflect an increase in number of elderly people, who have an increased risk of tinea unguium or people active in sport, who have a higher risk of exposure to dermatophytes. On the other hand, people may, in general, have become more aware of, and less tolerant to, possible nail infections, in part due to an increased exposure to commercials for antifungal treatment. The number of skin samples received for analysis has declined by 34% (feet) and 47% (skin unspecified body sites) during the 10-year-period. This is a contradiction in terms, as: (i) diagnosed cases of tinea unguium, which are believed to be preceded by dermatophyte infection of the skin (8) have not decreased; (ii) topical terbinafine was introduced to the Danish market in 1991 and, since 1994 to 1995, has been available over the counter, leading...
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...to a four-fold increase in use based upon registered defined daily dose (DDD) sold (1994: 711,450 DDD vs. 2003: 3,045,411 DDD. Source: Danish Medicine Agency, www.dkma.dk). It is most likely, that this has led to increased self-medication of skin infections without prior sampling and mycological diagnosis. Such self-medication could also contribute to the apparent dramatic decrease in skin infections, and especially *M. canis* skin infections, as these are typically inflammatory and easily recognized, and to the relatively high proportion of *T. tonsurans* isolates deriving from nails, despite *T. tonsurans* being considered mainly a skin and hair pathogen.

As in other studies (9, 10), the major limitation of this laboratory-based study of dermatophyte epidemiology is that it has not been possible to exclude repeated specimens from the same individual. The results are also influenced by possible confounders, such as changes in threshold for seeking a doctor or arbitrary decisions by the doctor on when to submit specimens for laboratory diagnostics. Nevertheless, we find that this study strongly suggests that the pattern of dermatophyte infections is changing. Up-to-date knowledge of epidemiology, in terms of clinical presentation, available treatment options and risk of disease spreading, is relevant, but also from the perspective of the mycology laboratory, as increased awareness of emerging species and the development of rapid molecular tests for the most prevalent agents, thus allowing a more rapid diagnosis (11).

**REFERENCES**