Risk factors of feline dermatophytosis in the Netherlands

Contents

Abstract

The purpose of this study was to estimate the prevalence and identify risk factors for feline dermatophytosis in an animal shelter in the Netherlands. In December 2012 and January 2013 all cats admitted to a shelter in Eindhoven were clinically examined and their hair coat was sampled. Sampling was done with a toothbrush using the MacKenzie technique and was followed by a Wood's lamp illumination. The cats admitted to the shelter were stray cats or given up for adoption by their owners and were from different ages, genders and breeds. European shorthairs were over presented. One of the kittens had suspect ringworm lesions on the front legs and head, which did not fluoresced on Wood's illumination.The toothbrush samples were cultured on Sabouraud B and Selective A agar and inoculated for 21 days. All cats were culture negative for dermatophytes, and therefore no risk factors could be identified. A short overview of risk factors described by other authors is included and the possibility that some samples were false negative is discussed. Further research with a larger survey is needed to get a better estimation of the prevalence and to identify possible risk factors.

***Keywords**: cat, feline, dermatophytosis, dermatophytes, ringworm, Microsporum canis, shelter, stray, given up for adoption, prevalence, risk factors, MacKenzie toothbrush technique, Wood's lamp illumination, mycological culture, Sabouraud agar, Selective agar.*

Introduction

Various kinds of skin diseases can be diagnosed on cats. One of them is the pathogenic fungal infection dermatophytosis, which is caused by *Microsporum* spp or *Trichophyton* spp that affects the hair shafts and stratum corneum, the outermost layer of the skin. Fungi of the genera *Microsporum* and *Trichophyton* are the species colonizing animals, and *Microsporum canis* is the most common found dermatophyte in cats (Cafarchia et al. 2006; Nweze 2011; Seker, Dogan 2011).

Affected cats may have localized, multifocal or generalized skin lesions. Alopecia with variable scaling and itching of mild intensity are common seen. The remaining hairs around the skin lesions are often broken (Medleau, Hnilica 2006). Cats can also be carrier of dermatophytes without showing any clinical signs of the disease (Cafarchia et al. 2006). Cats that mechanically carry spores on their coat without having a clinical infection are so-called “dust-mop” cats (Miller, Hurley 2009).

Transmission of dermatophytes occurs via infective spores originating from the hair coats of infected or colonized dogs and cats or even the environment. Infected cats or “dust-mop” cats are a source of contamination in their environment and can provoke airborne presence of viable fungal spores (Mancianti et al. 2003). The human skin can be contaminated with those spores and if skin barrier is insufficient or the body’s immune system fails, these spores can cause a clinical infection in humans. Dermatophytosis is therefore a zoonotic disease (Cafarchia et al. 2006).

A study in 78 dog and cat shelters in the United States of America and showed that dermatophytosis is one of the top-three diseases of concern in shelters (Steneroden, Hill & Salman 2011). Since shelter animals are exposed to high levels of stress, they are more susceptible for infectious diseases like dermatophytosis than other cats. A new environment, a continuously changing social environment and poor body condition score influence the general health of the shelter animals and make them more vulnerable. When cats are under stress, they groom themselves less frequently. Infective spores of fungi, which are normally removed by mechanical grooming, now get a chance to infect. Cats living in groups may fight, increasing the risk of trauma to the skin thus increasing the risk of infection (Carlotti et al. 2010). To warrant the good health of shelter animals, a different approach of small animal veterinary medicine is needed. Instead of focusing on the well-being of the individual animal, a group approach is preferable. Shelter medicine uses this approach to warrant the health of the shelter animals.

The Netherlands has approximately 100 animal shelters, adopting a total of 15.000 dogs and 35.000 cats per year. Little is known about the prevalence of infectious diseases in shelters in the Netherlands which makes shelter medicine difficult to perform. To optimize the results we can achieve with shelter medicine, we need to know more about the prevalence and risk factors of common infectious diseases like dermatophytosis.

Various risk factors for feline dermatophytosis are described. Some authors suggest that the highest incidence of dermatophytosis might be seen in kittens, immunocompromised animals and long-haired animals; Persian cats appear to be predisposed (Medleau, Hnilica 2006; Miller, Hurley 2009). Some researches affirm these risk factors, while other studies have different outcomes. Since factors as humidity, temperature, environment and housing of the cats might also influence the presence of dermatophytosis, the results of different cat populations, in different countries in the world cannot be extrapolated to the Dutch cat population (Boyanowski et al. 2000; Simpanya, Baxter 1996; Moriello, Newbury 2006; Copetti et al. 2006; Iorio et al. 2007; Thomas, Scheidt & Walker 1989). The purpose of this study was to estimate the prevalence and identify risk factors for feline dermatophytosis in an animal shelter in the Netherlands.

Materials & methods

Cats, housing and study area

A total of 60 cats that were admitted to a big shelter in Eindhoven, the Netherlands, between December 2012 and February 2013 were sampled and screened for dermatophytosis. Only cooperative cats older than 1 month of age, which could be sampled within 48 hours after entering the shelter were included in the study. Cats entering the shelter were housed individually, in a cage that was cleaned with bleach (household bleach: unknown concentration) in between cats by the shelter volunteers and employees. Kittens from one litter were not separated, according to the shelter policy, and were housed together in a large cage.

Figure 1 shows a rough map of the shelter, with the rooms for cats noted as: B, C, QB, 1, 2, 3 and 4. Since the shelter was reconstructed during the study, the occupation of rooms changed. At first, stray cats were put in the quarantine rooms marked with C and B and when the rooms were finished, 4 and QB were most frequently used. Room 1, 2 and 3 were non-quarantine rooms which were used for cats given up for adoption by their owner. Room C was used to house the kitten litters.

The standard quarantine period for all stray cats was 21 days, while the cats coming from a household were put up for adoption immediately, provided that their vaccination status was good. Food and water were given multiple times a day; all cats had unrestricted access to water and all the kittens had unrestricted access to food. Cleaned and disinfected toys were available for all cats.

To prevent transmission of diseases, visitors of the shelter could not enter the cat rooms. The shelter staff was trained to clean the cages properly, to take care of the young cats before the older ones and hands were washed and disinfected in between each animal. No protective clothing was worn.

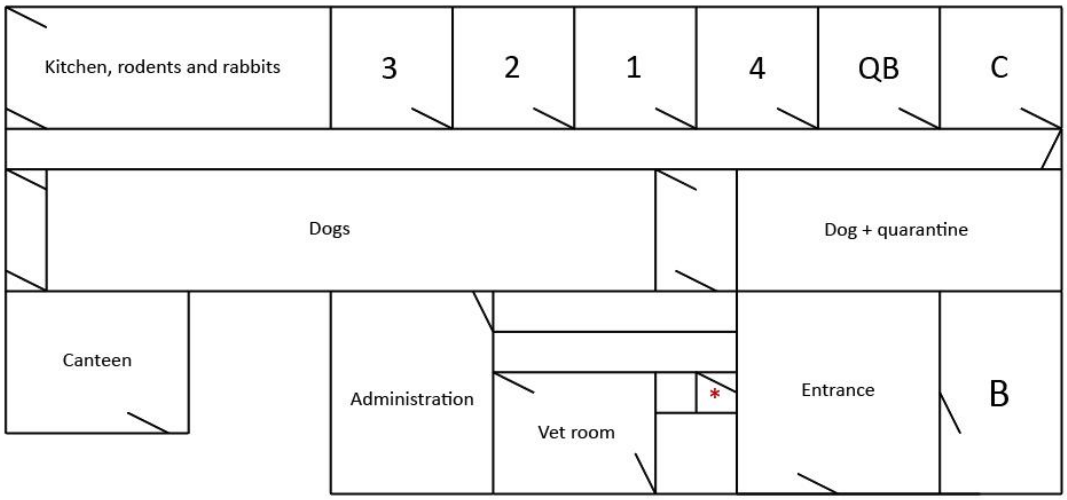

Figure 1: Map of the shelter. Red asterisk indicates the Wood's lamp illumination test room.

Sample collection

All cats were brushed for 60 seconds with a plastic toothbrush (Lactona 18 Nylon Soft Toothbrush, 3 bristle rows, figure 2) over the whole body, with the main focus on the head and fore limbs. Brushing was alternately performed by two researchers, which were wearing disposable overalls, overshoes and hairnets. After brushing the animal, each brush was kept in a separate envelope and transported to the laboratory within 2 days.

Samples were taken in the cat's cage or in the veterinarian room that was cleaned with bleach (approximately 20 ml 5% sodium hypochlorite in 1 L water = 5mg/L) . Transportation occurred in cages cleaned with the same amount of bleach.

Environment samples were taken by swiping a visually clean part of the room with an unfolded sterile gauze (Cutisoft). These samples were taken in the veterinarian room, room C, room

2, room QB and room 3, on day 27, 27, 49, 49 and 61 of the study respectively. After sampling, the gauze was folded and put in a separate envelope.

Figure 2: Toothbrush used for sampling

Data collection

A general examination of the cats was performed after the brush sample was taken. The following data was noted: hours in shelter, reason of admission, breed, sex, weight, hair length, (estimated) age, body condition score (BCS), breathing, pulse, temperature, skin condition, lesions and other abnormalities in the health check. Furthermore, their level of co-operation and character were included. Body temperature was only measured in cases of (suspect) illness, to minimize stress. All cats without a microchip were chipped with a Five Star ID microchip to optimize registration and follow-up. Chipping was done after sampling and the general examination.

Afterwards, each cat was transported to a small dark room where they were examined under Wood's lamp illumination (see figure 1). The Wood's lamp was used after a warming-up period of at least 15 minutes, to reach operating temperature. The examination with the Wood's lamp was performed after the toothbrush samples were taken, to minimalize the risk of environment contamination of the sample.

Laboratory methods

The samples were taken to Utrecht, VMDC (Veterinary Microbiologic Diagnostic Center) for mycological culture.

All toothbrushes were inoculated on Sabouraud B and Selective A agar (Biotrading, Benelux b.v.) by pressing the bristles 10 times into the medium. Hairs were picked off the brush with a sterile forceps and gently pressed into the medium. Cutisoft samples of the environment were unfolded and the sampled surface was gently pressed on the agar.

The Sabouraud B agar contains peptic digest of animal tissue (5.0 g/L), pancreatic digest of casein (5.0 g/L), dextrose (40.0 g/L), inositol (10.0 g/L), cycloheximide (0.2 g/L), vitamin B (1.0 g/L), depomycin (3.0 g/L) and agar (15.0 g/L). The Selective A agar contains peptone from soymeal (10.0 g/L), D(+)glucose (10.0 g/L), cycloheximide (0,4 g/L), chloramphenicol (0.05 g/L) and agar (12.5 g/L). The plates were incubated at 25^{o}C in the dark up to 21 days and examined at least 3 times a week. When suspect colonies were found, microscopic examination was performed to identify the colonies. Microscopic examination was achieved by putting the fungus on a microscope slide with lactophenol cotton blue, by use of a small piece of sticky tape. The slide was then examined under a light microscope. By examination of the hyphae, macroconidia and microconidia a determination of the fungus was performed, to identify dermatophytes.

In case of overgrowth of the plate with a different fungus, suspect colonies were re-cultured on a new plate and observed for another 21 days. The original plates were thrown away to prevent contamination.

In one doubtful case an urease test was performed for further identification.

Statistical analysis

Win-episcope 2.0 was used to evaluate the size of the sample group (Computer-aided Learning In Veterinary Education 2000). A total shelter cat population of 35.000 cats was used for the calculations.

The confidence interval was calculated by the method of Newcombe et al. using the Confidence Interval Calculator (Newcombe 1998; Herbert 2002).

Results

Description of the cats

From the 60 cats included in this study, 27 (45%) were female and 33 (55%) were male. One of the cats was admitted twice to the shelter, during the 2 month period. Since a few weeks passed, he was sampled again to see if he was still negative on dermatophyte culture the second time he entered the shelter.

Twenty (33.3%) of the cats had a history of coming from a multi-cat household or were brought in, in a group. Eleven of them, which were all kittens, were brought in as part of a litter and were also housed together in the shelter. The adult cats were all housed in a single-cat cage, even if they came in together. Table 1 shows the occupation of the different cat rooms.

	Room B	Room C	Room QB	Room 3	Room 4
Single cat cage	2	7	15	15	9
Multi cat cage	0	11	0	0	0

Table 1: number of cats housed in the different cat rooms

Of the 60 cats, 18 (30%) were <6 months, 9 (15%) were 6 months-1 year and 33 (55%) were 1 year or older. Almost all of the cats were European shorthairs (95%); the 3 remaining cats were a Persian, a Norwegian Forest cat and a Maine coon, all with long hair. Nine of the 60 (15%) cats examined were suspect or diagnosed cat flu patient and were started on Doxycycline therapy. Other health problems that were diagnosed include heart murmur (2/60), dental problems (2/60), shock due to drowning (1/60), pregnancy (1/60), urolithiasis (1/60), burn marks on the paws due to gasoline (1/60) and blindness (1/60). Six of the cats had serious flea infestation and two suffered from ear mites.
Of the 60 cats, 40 (66.7%) were stray cats, 16 (26.7%) were given up for adoption and 4 (6.7%) were doubtful. The 4 doubtful cases were part of 1 litter that was given to the ambulance personnel and consisted of healthy, good-looking and properly fed 4-months-old kittens. All the cats from households were older than 6 months. The stray cats were found in a radius of 30 km around Eindhoven (figure 3).

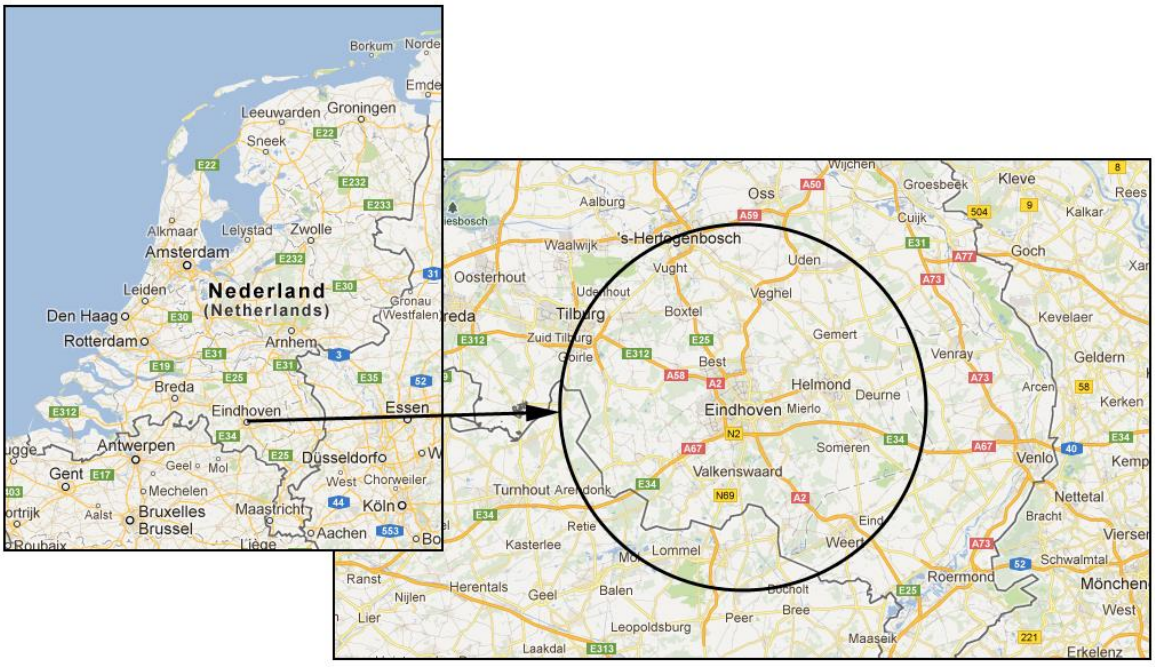

Figure 3: The black circle indicates the area where stray cats were found. Maps obtained from Google Maps.

Sample size

To detect a prevalence of 7.6% in shelter cats, a minimum sample size of 38 cats will be required, with a level of confidence of 95% (Newbury et al. 2007). The 60 cats that were used in this study are an adequate amount to detect a prevalence of 4.87% or higher, with the same level of confidence.

Weather

Figure 4 en 5 show the daily mean temperature and humidity during the sampling period.
The mean temperature and mean relative humidity during these 2 months were 3.6 degrees Celsius and 90% respectively. It snowed or hailed approximately 10 times.

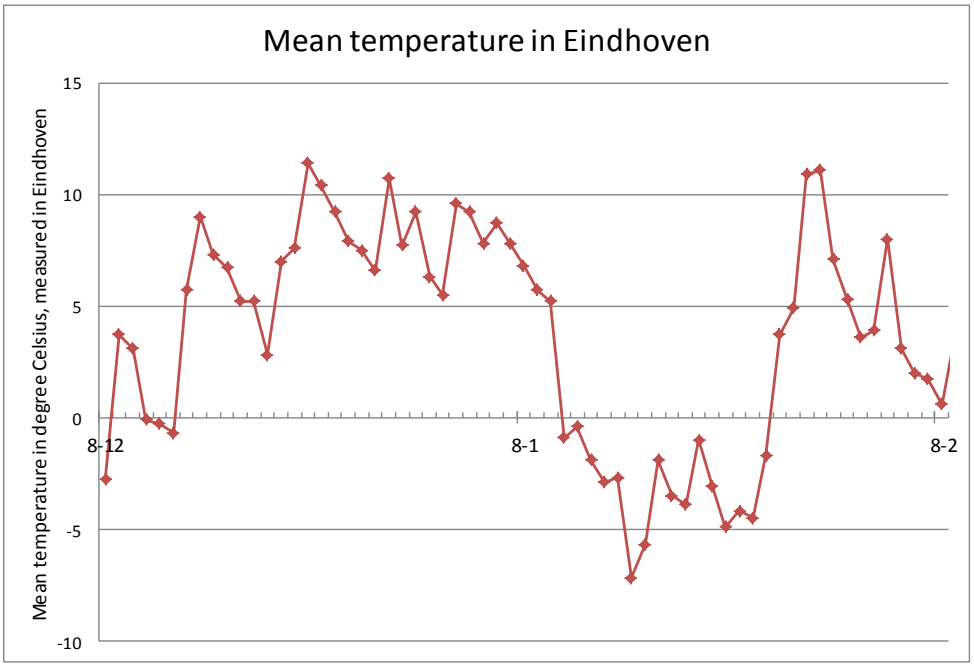

Figure 4: Mean temperature in degree Celsius in Eindhoven, the Netherlands, during the sampling period. Data obtained from KNMI, national data and knowledge center for weather.

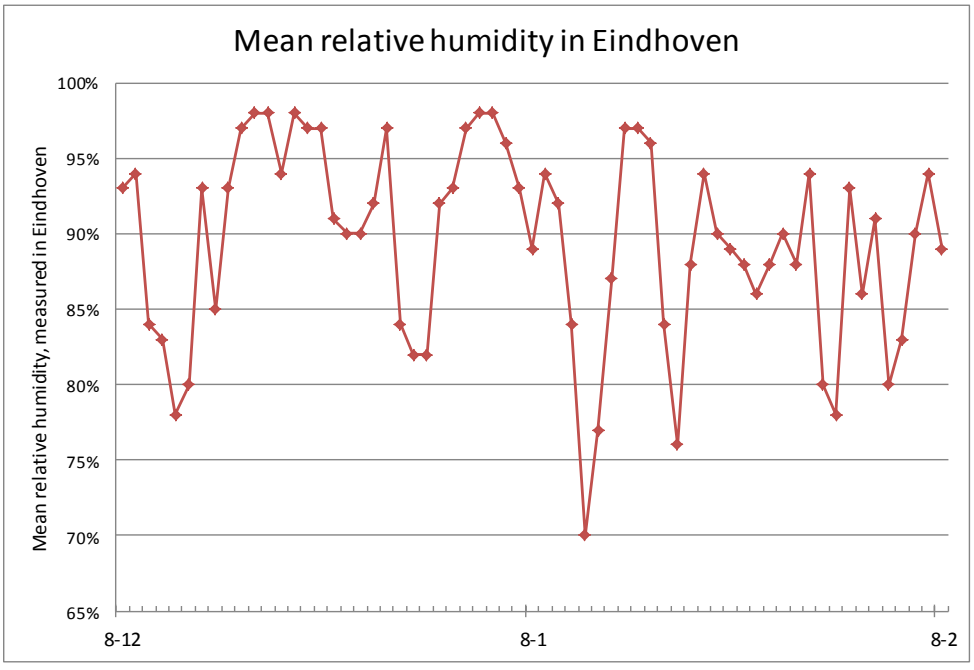

Figure 5: Mean relative humidity in Eindhoven, the Netherlands, during the sampling period. Data obtained from KNMI, national data and knowledge center for weather.

Diagnostic results

The mean time the cats spent in the shelter before sampling was 23 hours, with a range from 0-46 hours.

Of the total of 57 cats examined under Wood's illumination, 49 (86%) were negative and 8 (14%) were doubtful. On dermatological exam, 13 (21.7%) cats had moderate hair loss and 13 (21.7%) had sever hair loss, 6 (10%) cats had local alopecia because of fleas or fighting and 2 (3.3%) had wounds. 6 (10%) cats had lots of tangled hairs; 2 of them needed to be shaved totally. One cat was not dermatological examined, because of his bad temper and shock-status due to almost drowning. Only 1 kitten had suspect ringworm lesions on the front legs and head, which did not fluoresced on Wood's illumination.

A lot of saprophytes grew during the 21 days incubation period of each sample. No determination of these fungi was done. However, some fungi's have been determined and those were mostly *Aspergillus* spp., especially in the first week of the study. Other saprophytic fungi found were *Penicillium* spp and *Alternaria* spp. Only 4 cats were negative on mycological culture on 1 of their plates; 2 Sabouraud B and 2 Selective agar plates remained empty.

Twenty-five Selective A agar plates and 33 Sabouraud B plates were thrown away before the 21 days of inoculation had passed, because of rapid overgrowth (Table 2): 22 cats had both of their plates overgrown before the 21 days had passed. Twelve of these plates contained suspect colonies, which were re-cultured on Sabouraud B agar before throwing away the original agar.

	Inoculation time (in days)																
	6	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**	**20**	**21**	**Total**
Sabouraud B	1	5	3	1	4	3	0	2	1	0	6	2	1	2	2	27	60
Selective A	0	4	0	0	7	2	0	3	0	3	0	3	1	1	1	35	60

Table 2: Inoculaton time of the plates in days

No dermatophytes were found on both Sabouraud and Selective agar. In this study, the prevalence of feline dermatophytosis, and therefore of *Microsporum canis,* is 0% (95% CI: 0-6%).

No dermatophytes were cultured from the environment samples.

Discussion

The 60 shelter cats sampled during this study were all negative for dermatophytosis on mycological culture. Risk factors associated with dermatophytosis could therefore not be estimated. Possible explanations for the absence of dermatophytosis in our survey will be elaborated below.

Sample size

First of all, the sample size used in this study was relatively small. Because the number of cats admitted to the shelter during the winter period is relatively low in comparison with the summer, there were few cats to sample.

With the use of Win Episcope 2.0 a required sample size can be calculated. To do so, an assumption of the prevalence should be made. In 2007 Newbury et al. performed a study in a big shelter in the USA; of the 4017 cats sampled, 304 cats (7.6%) were found to be culture positive for *Microsporum canis* (Newbury et al. 2007). Since the population of cats used in this study corresponds with our population and the sample size is relative large, we assume that a prevalence of 7.6% is also plausible for the Dutch shelter cat population.

Considering a Dutch shelter population of 35.000 cats and a prevalence of 7.6%, 2660 of the cats will be positive for *M. canis*. With a level of confidence of 95%, the required sample size is 38 cats. Assuming abovementioned prevalence, the sample size of 60 cats was high enough.
To determine a prevalence of 0%, a much larger sample size is needed. With a cat population of 35.000 and 60 samples, the prevalence should be at least 4.9% to detect more than 1 positive cat. So in our survey, a prevalence of ≤4.9% is still possible.

False negative:

Furthermore, the option exists that some of the cultures were false negative. Moriello and Newbury think the toothbrush technique is superior when screening cats for dermatophytosis, but false negative fungal culture results are still possible. The most common reason for this are: rapid overgrowth of the plates because of contaminants, a poor culturing technique or poor handling of the toothbrush before inoculation (Moriello, Newbury 2006). Furthermore, the cats could have received an antifungal therapy before culturing. This option cannot be ruled out, since no clinical history is known for most of the cats in our study.

Overgrowth

Contamination of clinical specimens of veterinary origin is a severe limiting factor to cultural procedures in mycological diagnostics (Schmidt 1996). In our study, a lot of plates were thrown away before the 21 days of inoculation had passed, because of rapid overgrowth (Table 2).
Microsporum canis grows within 7-14 days and the growth of *Trichophyton* species may take 21 days (Moriello, Newbury 2006). In asymptomatic cats it may take 21 days before *Microsporum canis* grows (Moriello 1990). Since almost half of the plates were thrown away before the 3 weeks inoculation time had passed, some dermatophytes might be missed. Therefore, some cats might have been cultured negative due to the rapid overgrowth of the plates, instead of the absence of dermatophytes.

It was not determined for all plates which fungi caused the overgrowth, but *Aspergillus spp* was found several times. *Aspergillus* species are aerobic and grow rapidly on standard Sabouraud dextrose agar. Distinct colonies can be seen after incubation for 2 to 3 days, which can be up to 5 cm in diameter after 5 days (Quinn 2007). Spores of *Aspergillus* species are present in dust and air, so contamination of the coat of cats occurs easily. Other authors confirm the finding of *Aspergillus* on the coats of cats without dermatophytosis (Boyanowski et al. 2000; Moriello, DeBoer 1991; Moriello, Kunkle & DeBoer 1994; Moriello, DeBoer 1991; Sierra et al. 2000). To prevent rapid overgrowth with *Aspergillus* spp in a future research, the toothbrush could be disinfected with 90% methanol or 90% ethanol for more than 5 minutes before culturing (Miller, Hurley 2009; Okungbowa, Usifo 2010).

Culturing technique

The samples were taken as described by MacKenzie (Mackenzie, W & R. 1963), by brushing the whole cat with a toothbrush for 60 seconds. The MacKenzie method is noted as the most reliable technique for detecting *Microsporum canis* on the hair coats of both symptomatic and asymptomatic cats (Moriello 1990).

It was sometimes difficult to obtain enough hair from the short-haired kittens for adequate culturing. This might be the result of a too gentle combing technique or might be caused by the toothbrush, which was quite rigid and had few bristles (Figure 2). For further research, a toothbrush with more and softer bristles might be useful.

The Sabouraud B and Selective A agars are routinely used in the VMDC for culturing dermatophytes of different animal species and are found to be very useful for this purpose.

Handling of the toothbrush

All the toothbrushes were separately packed in sealed envelopes after sampling and transported to the laboratory in Utrecht. Between sampling and inoculation on the agars, was a period of up to 72 hours. During this period, the toothbrushes were not exposed to extreme temperatures and the envelopes were only unsealed in the fume cupboard. Poor handling of the toothbrushes before inoculation seems not to be the case.

Other points of attention

The transport cages used in this shelter are in need of improvement. To transport the animals wired cat transport cages are used, which are difficult to clean properly since they contain a lot of bars. Preferably, easier to clean solid cages are used in the future.

Another point of attention is the use of the Wood's lamp. In our study, 8 cases fluoresced green under Wood's lamp illumination, but were culture negative. Hair shafts infected with *Microsporum canis* have an apple green color under Wood's lamp illumination due to tryptophan metabolites. However, false positive fluorescing might occur due to shampoo, ointments, mucus or other topical applications, which was probably the case in our study (Moriello 1990).

Risk factors

Since no cats had positive cultures for dermatophytes, no risk factors could be identified. Other authors identified several risk factors associated with dermatophytosis, which will be described briefly.

Appendix 2 shows the feline data extracted from other studies, with the focus on *Microsporum canis*. A lot of studies have been performed and published in Europe and America, but less is known about the feline dermatophytosis status in Asia and Africa.

Age

Cats of all ages were included in our study. Since none of them was positive for dermatophytosis, no age group can be designated as more susceptible.

Most other studies agree on the fact that young cats are more often culture positive for dermatophytosis than older cats. Menges and Georg were one of the first who made this statement. Of the 109 cats they sampled that were <1 year of age, 67% was positive for *Microsporum canis*. The percentage of positive cats decreased with age, but ringworm was found in cats of all ages (Menges, Georg 1957). Since no statistical analysis is performed or described in the article, the significance of this higher percentage in younger cats remains unknown. However, other articles draw the same conclusion. Several studies examining cats with suspect dermatophytosis lesions, also find a significant higher percentage in cats <1 year (Seker, Dogan 2011; Brilhante et al. 2003; Lewis, Foil & Hosgood 1991; Cafarchia et al. 2004; Torgerson, Abbott 1999; Sparkes et al. 1993; Yahyaraeyat et al. 2009). In studies examining asymptomatic cats, less often a significant lower percentage in cats <1 year of age is found (Cafarchia et al. 2006; Romano, Valenti & Barbara 1997). A study with (mostly) healthy stray cats even describes a higher prevalence in cats >12 months (Natale et al. 2007).

Young cats might be more susceptible to dermatophytosis, due to the fact that young animals are not completely immunocompetent (Cafarchia et al. 2004). However, the significant age differences are mostly found in studies performed on cats with suspect lesions. Younger cats might be over presented in these studies, because they are more likely to develop lesions due to dermatophytes than adult cats, which are more often asymptomatic carrier (Yahyaraeyat et al. 2009).

Gender

Males and female were almost equally represented in our study. Since all of the cats were negative for dermatophytes on mycological culture, no differences can be found between males and females. Most other studies find no statistical difference between male and female cats positive for dermatophytosis.

One study detected a significant higher prevalence in female cats (Natale et al. 2007). This might be due to the disproportion between male and female cats examined. Only 21 males were sampled, compared to 246 females.

Another study found that neutered male cats had a 12 fold higher risk for having dermatophytosis compared to intact male cats (Boyanowski et al. 2000). The wide confidence interval implies a weaker confidence in the increased risk for neutered male cats, but there is no obvious other explanation for this finding.

Season

The samples in our study were taken during the winter months, with a mean temperature of 3.6 degrees Celsius. It hailed or snowed ten times and the ground was covered with snow for most of the time. Other authors suggest that snow decreases soil contact and therefore decreases the amount of from-soil-originating fungi on cat coats (Aho, Padhye & Ajello 1987). The snow may also have influenced our results. First of all it might have covered the dermatophytes in the environment and therefore decreased the exposure to the coats of the stray and free roaming pet cats. Further, it might have influenced the roaming behavior of the cats negatively.

The data on the seasonality of dermatophytosis in the literature is controversial. Some authors suggest that the highest prevalence of dermatophytosis is found in the cold seasons. A 15 year during study in Italy on 7650 suspect pet cats showed a significantly higher recovery rate for *Microsporum canis* in the fall and winter than in summer and spring (Mancianti et al. 2002). Another study on asymptomatic cats also describes a higher prevalence in the winter (Cafarchia et al. 2006). Some other studies also find a higher prevalence, but the significance is unclear (Iorio et al. 2007; Antos, Breuer-Strosberg & Awad-Masalmeh 1996; Ainsworth, Austwick 1955; Kristensen, Krogh 1981; Siesenop, Busse & Böhm 1996).

In the observation on the seasonal variations of ringworm, conducted by Kaplan and Ivans, no clear seasonal incidence of *Microsporum canis* was found. This is in contrast with the clear cut pattern of incidence they found in the dog. The data indicates peak months in Augustus, September, December and January, but the infections are also common during the other months of the year (Kaplan, Ivens 1961). Other authors agree that there is no significant seasonal distribution(Seker, Dogan 2011; Brilhante et al. 2003; Lewis, Foil & Hosgood 1991; Sparkes et al. 1993; Baxter 1973; Khosravi, Mahmoudi 2003; Lopez et al. 2012; Palumbo et al. 2010).

In studies conducted in Italy, a higher prevalence of *Microsporum canis* infection in the summer was found. Cafarchia et al. examined 156 symptomatic pet cats and found a higher percentage of culture positive animals in the summer and autumn (Cafarchia et al. 2004). Another study in Italy showed a peak of infections in late spring, summer and autumn. The authors think it could be related to the seasonal birth of kittens (Marchisio et al. 1995).

Climate

Warm temperatures favor the germination of spores. Moriello and Newburry think that this need for moisture and warmth could explain why dermatophytosis is more common in tropical and semitropical climates and during the warm months in temperate climates (Moriello, Newbury 2006). As said before, the literature is not equivocal about the latter, but the climate might have an influence on the prevalence of dermatophytosis.

In the USA, 2 studies comparing the incidence of dermatophytosis in different regions have been conducted. Moriello et al. compared the incidence of dermatophytosis in shelter cats from the northern cold dry region with those in the southern warm, humid region of the USA. *Microsporum canis* was only isolated from 8% of the cats in the southern region. This was a significantly higher percentage than in the northern region, where 0% of the cats was positive for *Microsporum canis* (Moriello, Kunkle & DeBoer 1994). The other study was performed by Boyanowski et al. in four geographically distinct regions in the Pacific west coast of the USA. It turned out that the basin in Los Angeles was more likely to have cats that cultured positive for dermatophytes, which were all *Microsporum canis*, compared to the basins in San Francisco, Seattle and Sacramento valley. A cautious evaluation of these findings must be made given the low overall prevalence of dermatophytosis in this study. Moreover, the basin in Los Angeles did not routinely euthanize cats, what could have increased the possibility of having a positive cat remain in the population for a longer time, being a source for cross-contamination. Los Angeles had the highest combined temperature and humidity compared to the other regions, which were all warm but had a different level of humidity. Humidity might therefore be more important than temperature (Boyanowski et al. 2000). In Turkey, Seker et al found no statistical difference in incidence of dermatophytosis between Ankara and Izmir (Seker, Dogan 2011).

Appendix 2 shows an overview of dermatophytosis studies and the climates of the areas in which they are conducted. These climate classifications are based on the Köppen-Geiger climate classification system. The percentages *Microsporum canis* positive cats in Europe range from 0%-100%, in North America (USA) from 0%-100%, in South America from 0%-88.5%, in Asia from 0%-47.8% and in Australia from 3.7%-18.5%. The only study in Africa shows a prevalence of 32.5%. However, these prevalence ranges cannot be compared, since the sample size and the design of studies conducted on each continent differ too much.

If the outcomes are sorted on base of climates, no striking differences between the climate classes can be found (Appendix table 3). It is not possible to draw any conclusion from these findings, since the individual studies cannot just be compared mutually.

Breed and hair length

It has been suggested that long-haired cats are more often asymptomatic carriers of dermatophytes. Even if one of the 3 long-haired cats included in our study had been positive for dermatophytosis, no conclusions could be drawn, due to the small number of long-haired cats sampled.

In 1981, Quiafe and Womar examined asymptomatic cats attending cat shows to investigate the prevalence of *Microsporum canis*. At one show, 35% of the long-haired cats were positive. Though some may have had minimal suspect lesions, the hair density causes difficulty in the early detection. In the authors experience, show cats are more likely to carry spores since they live under domestic conditions with a number of other cats. Also, it's possible that the excessive combing and grooming of long-haired show cats produces an electrostatic effect that attracts fungal spores from the environment and other cats (Quaife, Womar 1982). Statistical analysis of the findings in this study is unknown or not performed. Other authors also find a higher percentage of *Microsporum canis* in healthy long-haired or Persian cats, without performing a statistical analysis to check the significance of the results (Brilhante et al. 2003; Antos, Breuer-Strosberg & Awad-Masalmeh 1996).

Sparkes et al. examined samples from cats with suspected dermatophytosis and found a higher prevalence in long-haired cats, including the separate analysis of non-pedigree cats. Definitive conclusions of these findings cannot be drawn owing to the lack of a healthy control population

(Sparkes et al. 1993). Other studies examining symptomatic animals also found a higher percentage in long-haired cats (Mancianti et al. 2002). Lewis et al. found an over representation over Persian cats with dermatophytosis compared to the hospital cases (Lewis, Foil & Hosgood 1991).

All abovementioned studies lack statistical analysis of the results or a healthy control population. Most studies performed in healthy cat populations show no significant differences in *Microsporum canis* positive cultures in short-haired and long-haired cats (Cafarchia et al. 2006; Moriello, Kunkle & DeBoer 1994;; Romano, Valenti & Barbara 1997; Betancourt et al. 2009). A study with only 1 suspect positive case among the healthy positive cats also shows no significant difference in hair coat length (Boyanowski et al. 2000). The same result considering breed is obtained from studies in Barcelona and southern Italy examining cats with suspect dermatophytosis (Cafarchia et al. 2004; Cabanes, Abarca & Bragulat 1997). Moriello et al. performed a study in cats with and without dermatophytosis and found no significant difference in hair length for the isolation of *Microsporum canis*. They think it is possible that *Microsporum canis* is more difficult to eradicate from long-haired cats, causing the impression that it is more common in long-haired breeds (Moriello, DeBoer 1991).

Cat type

The goal of our study was to estimate the prevalence of dermatophytosis and the associated risk factors in cats admitted to a shelter. This is important to know, since cats entering the shelter can cause an outbreak among other cats and can contaminate their environment (Mancianti et al. 2003). This also works the other way around, and to minimalize the chance of contamination from the environment, the cats were sampled as fast as possible after entering the shelter. The mean time spent before sampling was 23 hours, with a 0-46 hours range.

Few studies examined the prevalence of dermatophytosis in shelter cats. Mignon et al. found 15.6% culture positive cats in a group of 134 European cats, collected in a pond over a period of 2 years. However, seventeen of them were clinical and Wood's negative and were noted as asymptomatic carriers; most of them could have had contact with infected cats, since they were all confined in the same room (Mignon, Losson 1997). The real prevalence of *Microsporum canis* in this group of cats could therefore be much lower than 15.6%. Lower percentages are found by Newbury et al. and Boyanowski et al. The study by Newbury et al. is used to estimate the sample size required for our study, since the group of cats used best resembles our study group and the 7.6% positive cultures seems to be a reliable outcome (Newbury et al. 2007). The study of Boyanowski et al. shows a percentage of 5% positive cultures for *Microsporum canis*. The cats used in this study came from 4 different shelters and no information was available regarding the length of their stay in these buildings (Boyanowski et al. 2000). Since almost all positive culture came from 1 shelter, it is possible that only one cat caused the outbreak in the shelter. Another study assessed the shelter admissions of cats to animal welfare shelters in Melbourne. They found a ringworm percentage of 0.7% in the 15.206 cats admitted during the study period. No further information about sampling, culturing or fungi species determination is known (Marston, Bennett 2009). Shelter cats may be more susceptible for dermatophytosis since they are often under more stress than pet cats and therefore groom themselves less frequently. Infective spores are longer present on their coats, getting a bigger chance to infect. Thereby, cats housed in groups may fight increasing the risk of trauma, thus increasing the risk of infection (Carlotti et al. 2010).

The origin of the cats admitted to a shelter also influences the risk of dermatophytosis. Cats coming from multi-cat households are more likely to carry dermatophytes, cat from rural areas might be at higher risk and in a study in central Italy the prevalence of dermatophytosis in stray cats was higher than in pet cats (Cafarchia et al. 2006; Iorio et al. 2007; Thomas, Scheidt & Walker 1989; Sparkes et al. 1993; Sparkes et al. 1994). In our study, 66.7% of the cats were picked up from the street by the animal rescue and they were noted as stray cats. However, based on their appearance and character, most of them had lived with people before. None of the sampled cats was feral. Since pet cats might be less often infected with dermatophytosis than stray cats, this might have had a negative influence on our results. Another 33.3% of the cats sampled were coming in with a group or

were known to be coming from a multi-cat household and were therefore more likely to be positive for dermatophytosis.

Other cats that might be at higher risk for getting dermatophytosis, are the ones suffering from other diseases. A study performed by Mancianti et al. showed a positive correlation between dermatophytosis and FIV infection, but others showed no significant difference between FIV (and FeLV) positive and negative cats (Sierra et al. 2000; Mignon, Losson 1997; Mancianti et al. 1992). In a study of stray cats in Italy, also no correlation between dermatophytosis and other feline pathologies is found (Romano, Valenti & Barbara 1997).

Further, the purebred cats are said to be at higher risk (Sparkes et al. 1993; Keep 1963). Keep et al. showed a higher incidence of dermatophytosis on thoroughbred cats attending the veterinarian. They think that the owners of thoroughbred cats seek veterinary advice more often than regular cat owners. Thereby, ringworm develops and persists more rapidly in groups of cats and thus in the breeding establishments. Menges et al. also suggested a difference in the percentage of infections in purebred cats and mixed breed cats, due to the housing or higher susceptibility of the purebred cats (Menges, Georg 1957).

Further research

To obtain more knowledge about dermatophytosis in shelter cats in the Netherlands, further research is needed.

According to the shelter's administration, in 2012 a total of 1135 cats were admitted to the shelter used in this study. Eight of these cats had dermatophytosis and were treated systemically and isolated from the other shelter animals. It is unknown if all these animal were brought in alone or in groups and whether one animal was the cause of the outbreak in the other cats. These numbers indicate that the prevalence of dermatophytosis in this shelter might be higher than zero.

To obtain the best results in the future, a longer sampling period is indicated. Preferably in the spring and summer, because the largest number of cats are admitted in this period. If feasible, it would be even better to sample cats during a full year or one month each season, to obtain a larger sample size and get a clearer view on the seasonality of dermatophytosis in this region. Ideally, more long-haired cats are included in the study when a longer sampling period is used.

One cat admitted to the shelter was not added in our study, because he was unmanageable and thought to be feral. This might have influenced the results and in future research feral cats should be included.

The maximum amount of time the cats spent in the shelter before sampling was 46 hours. During this period the cats were handled as little as possible, to keep contamination from the environment to a minimum. Since none of the cats was culture positive after this period, this seems to be an appropriate time for a shelter with no suspicion of dermatophytosis. Free roaming of the cats should be kept to a minimum.

The toothbrush method by MacKenzie is a reliable method and should be used in future research. The laboratory methods used in this study are also sufficient. Because only the dermatophytes are determined, no information about other fungi is present. It would be valuable to determine the saprophytic fungi as well, to get an insight in the fungal population on a cat's coat. To achieve this, it is useful to add a mycologist to the research team.

Conclusion

No dermatophytes were isolated from the cats admitted to a shelter in Eindhoven during a 2 month period. The prevalence of dermatophytosis in the shelter cat population is 0% (95% CI: 0-6%). On base of our findings, risk factors as described by other authors, namely gender, age, hair length, breed, origin, and other illnesses, could not be estimated. Also nothing can be said about seasonality and climate. Further research with a larger survey is needed to get a better estimation of the prevalence and to identify possible risk factors.

Appendix 1: clinical data sampled cats

NB: personal information like names and chip numbers are not mentioned.

	H in shelter	Admission	Breed	Sex	Weight	Hair	Age	BCS	Breathing	Heart	Temp	Dermatology	Wood's	Character	Other
1	17.5	Stray	ES	F	3.09	Short	Adult (12 years)	3	28, CA	140, KRES	38.5	Tattered coat, loose hair, fleas	Spot on the right ear	Nice, but painful on palpation head area	Dental problems
2	25.5	Stray	ES	M	1.25	Short	Kitten (12 weeks)	3	24, CA	156, KRES	38.6	Some scales	Negative	Very nice	Worms, cat flu, murmer
3	36.1	Stray	ES	M	1.70	Short	Juvenile (6 months)	3	30, CA	240, KRES	n.a.	Smooth, shiny coat	Negative	Very nice	Cat flu, earmites
4	17.5	Given up for adoption	ES	M	4.8	Short	Adult (6 years)	7	20, CA	240, KRES	n.a.	Scaling, loose hairs (severe)	Negative	Nice, anxious	Urolithiasis
5	18	Given up for adoption	ES	M	3.90	Short	Adult (6 years)	5	20, CA	200, KRES	n.a.	Scaling, loose hairs (mild)	Negative	Nice, anxious	Shortening of the left ear
6	20	Stray	ES	M	1.68	Short	Kitten (3-4 months)	4	64, CA	180, KRES	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	
7	20	Stray	ES	M	1.65	Short	Kitten (3-4 months)	4	32, CA	210, KRES	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	
8	2	Stray	ES	F	2.5	Short	Kitten (4 -5 months)	4	20, CA	240, KRES	38.2	Smooth, shiny coat, no scaling	Negative	Nice	
9	18.5	Stray	ES	M	1.0	Short	Kitten (3 months)	3	30, CA	240, KRES	38.5	Not smooth, dull,dirty, loose hairs, fleas	Negative	Nice	Cat flu, fleas
10	22	Given to ambulance	ES	M	2.2	Short	Kitten (4 months)	5	22, CA	200, KRES	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	
11	22	Given to ambulance	ES	M	2.5	Short	Kitten (4 months)	5	50, CA	240, KRES	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	
12	5	Stray	ES	M	3.33	Short	Juvenile (8 months)	4	50, CA	180, KRES	n.a.	Scaling, loose hairs (severe)	Negative	Angry	Fleas, earmites
13	22	Given to ambulance	ES	F	1.65	Short	Kitten (4 months)	4	50, CA	240, KRES	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	Worms
14	22	Given to ambulance	ES	F	1.95	Short	Kitten (4 months)	4	60, CA	204, KRES	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	
15	23	Stray	ES	M	2.5	Short	Adult (17 years)	2	36, CA	205, KRES	n.a.	Loose hairs, crests on nose	Negative	Nice, timid	Pupil deformation, cat flu
16	7	Stray	ES	M	3.0	Short	Adult (8 years)	3	60, CA	266, KRES	n.a.	Tattered coat, crests nose, loose hairs (severe)	Negative	Nice, anxious	Blind, cat flu
17	2	Stray	ES	M	2.76	Short	Juvenile (8 months)	4	36, CA	156	n.a.	Smooth, shiny coat, no scaling	Negative	Very nice	
18	19.5	Stray	ES	M	1.9	Short	Kitten (3 months)	3	Purring	204	n.a.	Dull, smooth coat.	Positive on	Nice, playful	Cat flu, earmites

											Alopecia of the right ear, left lateral hindpaw, right lateral hock	top of the head		
19	19	Stray	ES	F	2.02	Short	Kitten (4 months)	4	54, CA	240	n.a.	Smooth, shiny coat, no scaling	Negative	Nice	> Lnn. mandibularis
20	17.5	Stray	ES	M	3.75	Short	Adult (8 years)	4	28, CA	150, KRES	n.a.	Lots of tangled hairs, scaling (mild), dull coat, alopecia caudal to the whiskers	Negative	Very nice	Dental problems, earmites?
21	-	Stray	ES	F	1.25	Short	Kitten (2 months)	4	60, CA	n.a.	n.a.	Alopecia: right frontleg, right hindleg (3th toe), head cranial to the right ear. Dirty hairs on the left frontleg	Negative	Very nice, active	
22	21	Stray	ES	M	1.91	Short	Kitten (5 months)	3	60	240, KRES	n.a.	Smooth, shiny, no alopecia	Negative	Nice, playful	
23	21	Stray	ES	F	1.75	Short	Kitten (5 months)	3	60	240, KRES	39.4	Smooth, shiny, no alopecia	Negative	Nice	
24	21	Stray	ES	F	1.85	Short	Kitten (5 months)	3	60	240, KRES	n.a.	Smooth, shiny, no alopecia	Negative	Nice, quiet	
25	21	Stray	ES	F	2	Short	Kitten (5 months)	4	60	216, KRES	n.a.	Smooth, shiny, no alopecia	Negative	Nice, quiet	
26	21	Stray	ES	F	2.23	Short	Kitten (5 months)	4	60, superf	240, KRES	n.a.	Loose hairs, smooth, shiny	Negative	Nice, quiet	
27	24	Stray	ES	M	2	Short	Kitten (5 months)	4	purring	144, KRES	n.a.	Some scales, smooth, shiny, earmites and fleas	Negative	Nice, active	
28	22	Stray	ES	M	n.a.	Short	Adult (5 years)	4	n.a.	n.a.	n.a.	n.a.	n.a.	Very anxious and stressed	Shock, dexamethasone
29	18	Stray	ES	M	4.42	Short	Adult (1 year)	5	24, CA	192, KRES	n.a.	Smooth, shiny, no alopecia	Negative	Nice, playful and active	
30	21	Given up for adoption	NF	F	3.66	Long	Adult (9 year)	4	60, CA	108, KRES	n.a.	Fluffy, a lot of tangled hairs, some scaling, smooth	Negative	Very nice, anxious and stressed	
31	1	Stray	ES	M	2.7	Short	Juvenile (7 months)	4	40, CA	108, KRES	n.a.	Smooth, shiny, no scaling	Negative	Nice, timid	Gasoline; burning marks on the paws
32	20	Stray	ES	M	3.99	Short	Adult (2 years)	5	50, CA	198 KRES	n.a.	Loose hairs, scaling (mild)	Negative	Nice	
33	25	Stray	ES	M	5.8	Short	Adult (8 years)	7	66, CA	200, purr	n.a.	Lots of loose hairs and scaling, some fight wounds, tangled hairs	Negative	Little bit scared, stressed	
34	17	Stray	ES	F	2.3	Short	Adult	2	50, superf	200, KRES	n.a.	Fluffy, not shiny, long nails	Green: head, ears back.	Nice, scared. Examined in cage.	Cat flu, doxycycline started

35	41.5	Stray	Maine Coon	M	3.1	Medium	Adult (2,5 years)	2	24, CA	140	n.a.	Fluffy, tangled hairs	Tip of head, right ear	Stressed, not well examinable	Cat flu
36	42	Stray	ES	M	3.93	Short	Adult (2-3 years)	5	60	120, KRES	n.a.	Smooth, shiny	Negative	Very nice and playful	Fleas
37	21	Given up for adoption	ES	F	n.a.	Short	Adult (8.9 years)	6	36	n.a.	n.a.	Dull, dirty, a lot of loose hairs	n.a.	Very scared, aggressive	
38	19	Stray	ES	F	2.94	Short	Juvenile (7 months)	5	24	180, KRES	n.a.	Smooth, shiny, no alopecia	Negative	Nice, curious	
39	22	Stray	ES	M	3.0	Short	Adult (1 year)	3	36, sniff	160, KRES	n.a.	Dull, fluffy, no alopecia	Negative	Very nice	Cat flu
40	24	Stray	ES	F	3.6	Short	Adult (2 years)	6	60	220	n.a.	Smooth, shiny, no alopecia	Negative	Scared, aggressive	Pregnant
41	21	Stray	ES	M	2.24	Short	Adult (6 years)	1	60	n.a.	n.a.	Dull, fluffy, dirty - just washed with shampoo	Fluoresced	Nice	Cat flu, doxy started
42	5	Given up for adoption	ES	F	2.05	Short	Adult (1 year)	3	40	160	n.a.	Smooth, shiny, no alopecia	n.a.	Extremely stressed, not fully examinable	
43	21	Stray	ES	M	6.7	Short	Adult (8 years)	8	54	198	n.a.	Very severe felt formation on back probably due to no grooming, dirty coat.	Negative	Obese, not active, stressed	Cat flu
44	21	Stray	ES	M	3.33	see 12	Juvenile (8 months)	4	50, CA	128, KRES	n.a.	Smoot, shiny, loose hairs	Negative	Nice	
45	22	Given up for adoption	ES	F	4.2	Short	Adult (5 years)	7	28, CA	180	n.a.	Smooth, shiny, no alopecia	Negative	Stressed	
46	24	Given up for adoption	ES	F	2.4	Short	Juvenile (8 months)	3	30, CA	150, KRES	n.a.	Smooth, shiny, no alopecia	Negative	Nice	
47	24	Given up for adoption	ES	F	2.65	Short	Juvenile (8 months)	4	48, CA	180	n.a.	Smooth, shiny, no alopecia, some loose hairs	Negative	Nice	
48	18	Given up for adoption	ES	F	4.12	Short	Adult (7 years)	5	40, CA	180, KRES	n.a.	Smooth, shiny, some loose hairs	Negative	Nice, stressed	Heartmurmur 3/6
49	20	Given up for adoption	ES	F	2.79	Short	Adult (1 year)	4	28	126, KRES	n.a.	A lot of loose hairs, shiny and smooth	Negative	Stressed	
50	20	Given up for adoption	ES	M	5.05	Short	Adult (1 year)	8	44	198	n.a.	Smooth, shiny, a lot of loose hairs	Negative	Nice, stressed	
51	18	Given up for adoption	ES	F	3.62	Short	Adult (11 years)	5	80, superf	240	n.a.	Smooth, shiny, moderate loose hairs	Negative	Scared, aggressive when forced	
52	18	Given up for adoption	ES	F	3.52	Short	Adult (10 years)	4	32, CA	240, KRES	n.a.	Smooth, shiny, a lot of loose hairs	Negative	Nice	
53	37	Stray	Persian	M	4.5	Long	Adult (6 years)	5	32, CA	180, KRES	n.a.	Very dirty coat, a lot of tangled hears, needs to be shaved, wound ear	Doubtful (because of dirt)	Very nice	
54	43	Given up for	ES	F	3.2	Short	Adult (7 years)	4	30	200, KRES	n.a.	Smooth, shiny, a lot of	Negative	Nice	

		adoption									loose hairs			
55	43	Given up for adoption	ES	M	4.2	Short	Adult (7 years)	5	40, CA	240, KRES	n.a.	Smooth, shiny, a lot of loose hairs	Negative	Nice, stressed	
56	24	Stray	ES	F	2.9	Short	Adult	2	24, CA	250, KRES	n.a.	Not shiny and smooth, dull, a lot of loose hairs	Spot on the ear (dirty)	Nice	Fleas
57	46	Stray	ES	F	2.5	Short	Adult (2 years)	3	40, CA	170, KRES	n.a.	Dull and dirty	Negative	Nice	Cat flu
58	21	Stray	ES	M	3.65	Short	Juvenile (8 months)	5	50, CA	150, KRES	n.a.	Smooth and shine, moderate loose hairs	Green spot above the right eye	Nice	
59	2.5	Given up for adoption	ES	F	3.6	Short	Adult (3 years)	5	72, superf	240, KRES	n.a.	Smooth and shiny	Negative	Nice, stressed	
60	2	Stray	ES	M	4.92	Short	Adult (3 years)	6	40,CA	192, KRES	n.a.	A lot of loose hairs, moderate scaling, crusts on the head, wounds	Negative	Nice	Fightmarks

Appendix table 1: Clinical data from sampled cats.

Abbreviations:

CA = costo-abdominal breathing
ES = European shorthair
H in shelter = Hours in shelter
KRES = powerful, regularly, equal, symmetrical pulse
n.a. = not examined
NF cat = Norwegian Forrest cat

Appendix 2: feline data extracted from other studies

Author & Year	Location	Climate	Group of cats	Clinical status		Technique	Culture plates	% MC positive	Risk factors found				
				Symptomatic	Asymptomatic				Gender	Age	Season	Breed	Cat type
Europe													
(Seker, Dogan 2011)	Turkey	Ankara: B Izmir: Ca	164, pet	Suggestive lesions	-	Hair & skin scrapings	SDA	14%	Not found	< 1 year	Not found	-	-
(Coelho, Alegria & Rodrigues 2008)	Portugal (Vila Real)	Ca	47, pet	Suggestive lesions	-	Hair & skin scrapings	SDA DTM	19.1%	Not found	-	-	-	-
(Iorio et al. 2007)	Italy (central)	Cc	100, pet 100, stray	10 with lesions	190 free of lesions	Lesions: skin scraping. No lesions: toothbrush	Mycobiotic	Pet: 8% Stray: 100%	Not found	Not found	Pet: highest prevalence in cold season, NS	-	Stray cats Pet: living in countryside
(Natale et al. 2007)	Italy (Veneto)	Cb	218, stray	5 of 58 positive cats	54 of 58 positive cats	Toothbrush	Mycobiotic	26.7%	Females	Older cats	-	-	-
(Cafarchia et al. 2006)	Italy (Bari)	Cb	248, single pet	-	Free of lesions, no history	Plastic brush	Sabouraud	23.4%	Not found	< 1 year	Winter	Not found	Cats cohabiting with owners with MC Rural area
(Menelaos 2006)	Greece (Thessaloniki)	Cb	52, pet	Suggestive lesions	-	Hair & skin scrapings	DTM	Unknown 51.9% dermatophytes	Not found	Young cats, US	-	-	-
(Patel, Lloyd & Lamport 2005)	UK	Cc	30, closed colony (CC) 139 pet, feral and stray cats	-	All free of lesions, healthy	Human scalp brush	SDA	CC: none Others: 2.16%	Not found	Not found	-	Long-hair (total dermato phytes)	Not found
(Cafarchia et al. 2004)	Italy (Bari)	Cb	156, pet	Suggestive lesions	-	Hair & skin scrapings	Mycobios	23.1%	Not found	<1 year	Summer, autumn	Not found	-
(Mancianti et al. 2002)	Italy (Tuscany, Pisa)	Ca	7650, pet	Suggestive dermatological signs	-	Toothbrush	Mycobiotic	24%	Not found	< 1 year, US	Fall and winter	Long-hair	-
(Mignon et al. 2002)	Belgium (Liège)	Cc	659, pet	Suspect	-	Hair & skin scrapings	Unknown	22.2%	-	-	-	-	-
(Sierra et al. 2000)	France (Paris)	Cc	85, pet, partly FIV or FeLV positive	1 positive had a ringworm lesions	No lesions	Mariat & Tapia	SDA	7.1%	-	-	-	-	**No** difference in FeLV or FIV positive cats
(Pinter et al. 1999)	Croatia (Zagreb)	Cc	1838, attending vet	Suggestive lesions	No lesions	Hair & skin scrapings	SDA	40.2%	Male, US	<1 year, US	-	Not found, US	Cats with lesions
(Torgerson, Abbott 1999)	Ireland (Dublin)	Cc	36, attending vet	Suspect	-	Hair & skin scrapings	InTray SDA	36.1%	-	Lower age	-	-	

(Mignon, Losson 1997)	Belgium (Liège)	Cc	471 pet, 134 shelter and 27 stray	Some had clinical signs	Healthy pet cats, stray cats and shelter cats	Toothbrush	Sabouraud	3.0% pet cats 15.7% shelter 100% stray	Not found, US	Not found, US	-	Not found, US	**No** association with FIV (small group)
(Romano, Valenti & Barbara 1997)	Italy (Siena)	Cb	173, stray	-	Asymptomatic	Toothbrush	Sabouraud	47.4%	Not found	Young cats	-	Not found	**No** correlation with other pathologies
(Papini, Gazzano & Mancianti 1997)	Italy (Pisa)	Ca	29, laboratory	6, ringworm lesions	23, healthy	Toothbrush	Mycobiotic	65.5%	-	-	-	-	Cats with lesions were positive, US
(Cabanes, Abarca & Bragulat 1997)	Spain (Barcelona)	Ca	12, attending vet 56, unknown	Suspect	-	Hair & skin scrapings	DTM Mycosel	Cultures send: 100% Other: 32.1%	Not found	Not found	Autumn and winter, NS	Not found	-
(Schmidt 1996)	Germany	Cc	78, attending vet	Significant tendency towards	-	Hair & skin scrapings	Sabouraud with growth factors	9.0%	-	-	-	-	-
(Siesenop, Busse & Böhm 1996)	Germany (Hannover)	Cc	3592, attending vet	Suspect	-	Hair & skin scrapings	Kimmig agar	22.1%	-	-	3th and 4th quarter, US Decrease: April-June	-	-
(Antos, Breuer-Strosberg & Awad-Masalmeh 1996)	Austria (Vienna)	Cc	1022, attending vet	-	No lesions	Flea comb	SDA	7.9%	Intact males, US	<3 months, US	February, March, July, US	Long-haired, US	-
(Breuer-Strosberg 1993)	Austria	Cc	384, attending vet	Suspect	-	Unknown	SDA	45.6%	-	-	-	-	-
(Marchisio et al. 1995)	Italy (Turin)	Cc	105, attending vet 1.8% treated	Suspected lesions	-	Hair & skin scrapings	Dermasel Sabouraud Lactrimel	50.4%	Not found, US	<1 year, US	May-June July-August November, US	-	-
(Sparkes et al. 1994)	UK (Bristol)	Cc	181, pet	-	Healthy, no lesions	Scalp massage brush	SDA	2.2%	-, small number of + cats	-, small number of + cats	-	-, small number of + cats	-, small number of + cats
(Sparkes et al. 1993)	UK (Bristol)	Cc	3407, attending vet	Suspect	-	Hair & skin scrapings	SDA	24.3%	Not found	<1 Year	Not found	Longhair Birman Chinchilla Longhair ed	Joint submissions Pedigree
(Mancianti et al. 1992)	Italy (Pisa)	Ca	35 FIV + 55 FIV -	-	No lesions	Toothbrush	Mycobiotic	+: 74.3% -: 25.5%	-	-	-	-	FIV infected
(Wawrzkiewicz et al. 1992)	Poland (Lublin)	Db	85, show and pet	-	No lesions	Toothbrush	Sabouraud	Show: 45.8% Pet: 16.2%	-	-	-	-	Showcats, US
(Caretta, Mancianti & Ajello 1989)	Italy (Tuscany)	Ca	93, attending vet	Suspect	-	Fine metal comb	SDA	58%					

(Wright 1989)	UK (Bristol)	Cc	2925, attending vet or clinical cases	Suspect	-	Unknown	SDA	23.0%	-	-	-	-	-
(Gethings et al. 1987)	UK (Bristol)	Cc	51, farm	Unknown	Unknown	Scalp brush	Sabouraud	0%	-	-	-	-	-
(Aho, Padhye & Ajello 1987)	Finland (southern part)	Db	276, cattery	4 showed skin problems	172 healthy cats	Fine metal comb	Poor base medium Actidione	0%	-	-	-	-	-
(Stenwig 1985)	Norway (SE)	Ce	279, attending vet	Suspect	-	Skin scraping	SDA Mycobiotic	29.7%	-	-	-	-	-
(Aho 1983)	Finland (Helsinki)	Db	75, pet	Suspect	-	Hair & skin scrapings	Poor base medium	20%					
(Quaife, Womar 1982)	UK (Portsmouth)	Cc	241, show	-	Not suspect	Plucked fur and nail brush	Ink blue antibiotics	1: 10% 2: 24% 3: 3.9% 4: 17.8% Total: 12.9%	-	-	-	Up to 35% of long haired cats, US	-
(Kristensen, Krogh 1981)	Denmark (Copenhagen)	Cc	227, attending vet	Suspect	-	Hair & skin scrapings	Sabouraud	29.1%	-	-	Autumn and winter. Peak in September, US	-	-
(Weiss et al. 1979)	Germany (Hannover)	Cc	448, attending vet	Unknown	Unknown	Hair & skin scrapings	Kimmig agar	28.8%	-	-	-	-	-
(Mantovani, Morganti 1977)	Italy (Bologna and Rome)	Cb	283, pet	-	Clinically normal	Hair scrapings	Unknown	26.5%	-	-	-	-	
(Marcelou Kinti et al. 1977)	Greece	Ca	100, unknown	-	No clinical signs	Hair	Sabouraud	17%	-	-	-	-	-
(Connole 1965)	Scotland (Glasgow)	Cc	18, indoor pet	-	No clinical evidence	Nylon hairbrush	Malt extract agar	0%	-	-	-	-	-
(Gentles, Dawson & Connole 1965)	Scotland (Glasgow)	Cc	30, unknown	-	No clinical evidence	Nylon hairbrush	Malt extract agar	0%	-	-	-	-	-
(Ainsworth, Austwick 1955)	UK	Cc	36, unknown	Suspect	-	Hair & skin scrapings	Unknown	47.2%	-	-	Peak of dog and cat ringworm in Nov-Jan	-	-
North America													
(Newbury et al. 2007)	Wisconsin	Db	4019, shelter	Unknown	Unknown	Toothbrush	Mycosel	7.6%	-	-	-	-	-

(Boyanowski et al. 2000)	San Francisco Los Angeles Seattle Sacramento	Ca Ca Cc Ca	50 from each region, shelter	8 had suggestive lesions	192, no lesions	Fluorescing hair with tweezers and all toothbrush	Potato flake agar	5% (2 in SF, 8 in LA)	Neutered male and female > intact male	No found	-	Not found	Cats from LA (highest comb. temperature and humidity)
(Moriello, Kunkle & DeBoer 1994)	Pennsylvania, New York, Wisconsin Gainsvile	Db Db Db Cb	200, stray	9 with skin lesions (none positive)	Except from fleas, no skin lesions	Toothbrush	SDA DTM	North: 0% South: 8% Total: 4%	Not found	Not found	-	Not found	Cats from warm and moist regions
(Moriello, DeBoer 1991)	Midwestern US	Da	176, from 7 catteries	Kittens with lesions	All adults were clinically normal	Toothbrush	Sabouraud DTM	4 catteries: - 2 catteries: 100% 1 cattery: 45.8%	Not found	Not found		Not found for hairlength	
(Moriello, DeBoer 1991)	Wisconsin	Db	172, pet	-	Free of skin disease	Toothbrush	SDA DTM	0%	-	-	-	-	
(Lewis, Foil & Hosgood 1991)	Louisiana	Cb	407, attending vet	Suspect	-	Unknown	Unknown	13.7%	Not found	<1 year	Not found	Persian	-
(Thomas, Scheidt & Walker 1989)	North Carolina	Cc	50 pet, 22 + unknown catteries	Suspect in catteries with history of ringworm	All household pets were clinically normal	Toothbrush	DTM or SDA	Pet: 0% Cattery history: 22.7% asymptomatic, 22.7% symptomatic. Cattery suspect: 100%	-	-	-	-	High-density cat population (small study group!)
(al-Doory, Vice & Olin 1968)	Texas (San Antonio)	Cb	6, pet	Suspect	-	Hair & skin scrapings	Mycosel	50%	-	-	-	-	-
(Kaplan, Ivens 1961)	35 states	-	1232, attending vet	Suspect	-	Hair & skin scrapings	Selective isolation medium	29.5%	-	-	January, December, August, September NS	-	-
(Kaplan, Georg & Ajello 1958)	35 states	-	524, attending vet	Skin lesions	-	Hair & skin scrapings	Selective isolaten medium	36.8%	-	<2 year, US	-	-	-
(Menges, Georg 1957)	32 states	-	281, attending vet	Ringworm lesions	No lesions	Hair & skin scrapings	Cyclo-heximide medium	44.5%	Not found, US	<1 year, US	Fall, too few cases	-	Purebred, US
South America													
(Lopez et al. 2012)	Argentina (Mendoza)	B	45, pet and shelter	With lesion	Without lesions	Lesions: hair & skin scrapings. No: toothbrush	SDA Lactrimel	11.1%	Not found	Not found	-	-	Not found
(Silva et al. 2011)	Brazil	Cb	7, pet	Suspect	-	Hair & skin	SDA	0%	-	-	-	-	-

	(Xanxere)					scrapings							
(Beraldo et al. 2011)	Brazil (Alfenas)	Cb	40, attending vet	2 with suspect esions	38 without lesions	Carpet (Mariat & Tapia)	DTM Sabouraud after color change	47.5%	Not found	Not found	-	Not found, US	-
(Palumbo et al. 2010)	Brazil (Sao Paulo)	Cb	22, pet	Suspect	-	Unknown	Mycosel	Unknown; total including dogs: 79.7%	Not found	Not found	Not found	Sample size too small	-
(Betancourt et al. 2009)	Chile (Temuco)	Cc	50, pet	-	Healthy	Mariat & Tapia Hair extracting tweezers	Sabouraud Lactrimel	60%	Not found	Not found	-	Not found	-
(Copetti et al. 2006)	Brazil (Santa Maria)	Cb	151, pet	Clinical suspicion	-	Hair & skin scraping	Micibiotic Sabouraud	25.2%	-	-	-	-	-
(Brilhante et al. 2003)	Brazil (Fortaleza)	Ac	38, pet	Suspect	-	Hair & skin scrapings	SDA	36.8%	Not found	<1 year	Not found.	50% of Persians. US	-
(Paixão et al. 2001)	Brazil (Fortaleza)	Az	18, pet	Suspect	-	Hair & skin scraping after soap wash	SDA Sabouraud Mycosel	33.3%	Not found, US	<1 year, US	-	Not found, US	-
(Guzman-Chavez et al. 2000)	Mexico	Cc	100, pet and shelter	Presence of lesions	Absence of lesions	Toothbrush	SDA	4%	Not found, all dermatophytes	Not found, all dermatophytes	-	-	>1 cat in same house
(Castañón-Olivares et al. 2001)	Mexico	Cc	20, laboratory	-	No lesions	Mariat & Tapia	Sabouraud	45%	-	-	-	-	-
(Zaror et al. 1988)	Chile (Valdivia)	Cc	56, attending vet	-	Healthy	Mariat & Tapia	DTM	23.2%	-	-	-	-	-
(Zaror et al. 1986)	Brazil (Sao Paulo)	Cb	104, stray	-	Healthy	Mariat & Tapia	DTM	88.5%	-	-	-	-	-
Asia													
(Yahyaraeyat et al. 2009)	Iran (Tehran)	B	124, pet	Dermatological lesions	-	Hair and skin scraping after 70% ethylalcohol	SDA (partly only KOH)	41%	Not found	<1 year	-	-	-
(Tahereh Shokohi, Naseri 2006)	Iran (Sari)	Cb	100, stray	-	Lesion free	Toothbrush	SDA Mycosel DTM	0%	Not found	Not found	-	-	-
(Khosravi, Mahmoudi 2003)	Iran (Tehran)	B	186, vet clinics	Suspect	-	Hair & skin scrapings	SDA	47.8%	Not found, US	<1 year, US	Not found, US	Not found,	-

												US	
(Khosravi 1996)	Iran (Isfahan)	B	100, stray	Clinical signs in 4 kittens	All adults were lesion free	Toothbrush	Sabouraud DTM	26%	Not found, US	Kittens with lesions, US	-	Not found, US	**No** difference between the districts
(Ali-Shtayeh et al. 1988)	West Bank	Ca	8, unknown	-	Healthy hair sampled	Hair	Sabouraud	12.5%	-	-	-	-	-
Africa													
(Nweze 2011)	Nigeria	Ac	77, pet & for sale	Suggestive lesions	-	Hair & skin scrapings	Dermasel	32.5%	-	-	-	-	-
Australia													
(Marston, Bennett 2009)	Autralia (Melbourne)	Cc	15,206 shelter	Unknown	Unknown	Unknown	Unknown	Unknown 0.7% ringworm	-	-	-	-	Kitten only & multiple cat admissions, US
(Simpanya, Baxter 1996)	New Zealand (Palmerston North)	Cc	178, shelter and vet	Unknown	Unknown	Toothbrush	SDA	18.5%	-	-	May-June, correlated with relative humidity	-	-
(McAleer 1980)	Autralia (Perth)	Ca	Unknown, pet, stray	Suspect	-	Toothbrush	SDA	Unknown	-	-	March-May, US	-	
(McLeer 1980)	Australia (Western)	Ca	Unknown, attending vet	Most had clinical signs	Trace human infection source	Hair & skin scrapings	SDA Part Parker Quink stain	Unknown	-	Most kittens, US	-	-	-
(Woodgyer 1977)	New Zealand (Wellington)	Cc	199, attending vet	-	No symptoms	Scalp body massager	SDA	6.5%	-	9/13, <1 year, US	-	-	-
(Baxter 1973)	New Zealand (Palmerston North)	Cc	1480, attending vet 200, pet	Attending vet might have lesions	Most of attending don't have lesions 200: no signs of ringworm	Hair & skin Plastic brush	SDA	3.7% Carriers: 36%	Males, US	Y<1 year, US	Not found	-	-
(Keep 1963)	Australia (Sydney)	Cb	1059, attending vet	Fluorescing on Wood's illumination	Non-fluorescing on Wood's illumination	Fluorescing hairs were plucked	Sabouraud	5.9%	-	<3 months, US	February and April, US	High in Persian, Siamese, US	-

Appendix table 2: Overview feline data extracted from other dermatophytosis/*Microsporum canis* studies.
Abbreviations: MC = Microsporum canis, NS = Not significant, US = Unknown statistics, SDA = Sabouraud dextrose agar, DTM = Dermatophyte Test Medium
Red text = article not written in English
*: Climate based on Köppen-Geiger climate classification system. Group A – Tropical climate: a. Tropical rainforest climate. b. Tropical monsoon climate. c. Tropical savannah climate
Group B – Dry climates. Group C – Mesothermal climates: a. Mediterranean climate. b. Humid subtropical climate. c. Oceanic climate. d. Temperature highland tropical climate with dry winters. e. Subpolar oceanic. Group D – Continental climates: a. Hot summer. b. Warm summer. c. Subarctic. d. Subarctic with severe winters
Group E – Polar climates: a. Tundra. b. Ice cap.

Climate	Prevalences (in %)																													
Ac	32,5	36,8																												
Az	33,3																													
B	11,1	26	41	47,8																										
Ca	8	13	17	19,1	24	50	58	65,5	66,1																					
Cb	0	0	5,9	8	14	23,1	23,4	25,2	26,5	26,7	47,4	47,5	50	88,5																
Cc	0	0	0	1,08	2,2	3,7	4	6,5	7,1	7,9	9	10	16	18,5	22,1	22,2	23	23,2	24,3	28,2	29,1	36,1	36,4	40,2	45,6	45	47,2	50	54	60
Ce	29,7																													
Da	35,1																													
Db	0	0	0	7,6	20	31																								

Appendix table 3: Prevalences sorted by climate. NB: this table shows the mean percentage of different groups of cats in one study. Therefore the 100% prevalence are not included. Studies conducted in regions with different climates that were not mentioning the separate outcomes, are not included (Seker, Dogan 2011, Menges, Georg 1957, Kaplan, Ivens 1961, Kaplan, Georg & Ajello 1958).

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