Prevalence of *M. canis* in cats in a Dutch animal shelter at the moment of admission

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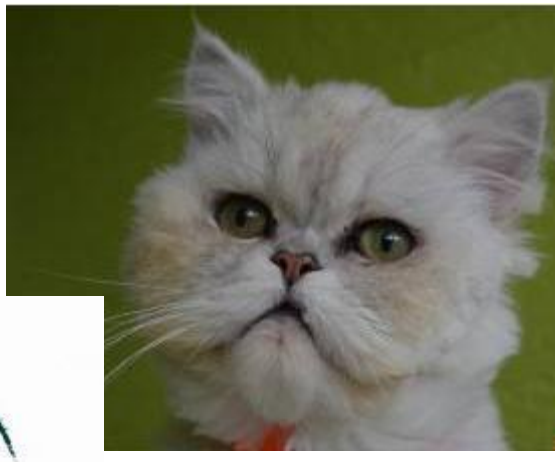

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Abstract

The risk of introduction of *M. canis* into Dutch animal shelters is evaluated in this study. *M. canis* is responsible for the infection with dermatophytosis in more than 90 percent of the cases in cats. The prevalence in a Dutch animal shelter is evaluated over a period of two months by testing the newly admitted cats (n=60) for fungal infections by using the MacKenzie method and culturing on Sabouraud and Selective Agar for three weeks. Fungal colonies were microscopically determined and reported as positive or negative for *M. canis*. The hypothesis : *M. canis* in Dutch shelter cats is a prevalent infection (>7,6%) at the moment of infection. No cats tested positive for *M.Canis* so the prevalence found in this research is 0%. Concluding: based on this outcome the hypothesis is rejected.

Introduction

There are about 100 animal shelters in the Netherlands. Cats and other companion animals like dogs and rabbits who are surrended by owners are brought to these shelters. The aim of the animal shelter is to find a new owner for these animals. Dutch animal shelters rehome approximately 35.000 cats a year. Dermatophytosis, also called 'ringworm' is one of the top-three diseases in animal shelters and is known and feared for its outbreaks [1] and is of particular importance in these facilities because of its zoonotic and easily transmittable character [2].

There is little information about the situation concerning *M. canis* in the Netherlands . Because dermatophytosis is a problem in animal shelters, the aim of this study is to survey the prevalence of *M. canis* at intake in a Dutch animal shelter of new cats presented to the shelter to identify the extent of this problem exposed to this facility so statements can be made about the situation in The Netherlands. The reason the cats are examined at the intake, instead of examining the current population of cats in the shelter, is to study the prevalence of *M.canis* in the cat population presented to Dutch animal shelters instead of the incidence of *M.canis* in a typical Dutch shelter. The incidence is the total number of infected cats in a particular time period and depends on sources of infection, contamination of the environment and management of the facility. Previous studies, some of them listed in appendix I examined the incidence of *M.canis* rather than the prevalence. This study however examines the prevalence. The prevalence of *M.canis* is an exposure risk for the animal shelter, the incidence is something they can influence by management interventions.

The hypothesis of this study is based on one of the few prevalence studies performed by *Newbury et al.*. Information about this study is discussed in the paragraph '*previous studies'*.

Hypothesis: *M. canis* in Dutch shelter cats is a prevalent infection (>7,6%) at the moment of admission.

Dermatophytosis

Dermatophytosis is a superficial fungal infection of the skin. In cats, *M. canis* is responsible for the infection in more than 90 percent of the cases. Other rarely isolated species include *M. gypseum, M. persicolor and T. mentagrophytes* [3]. An important epidemiological factors of *M. canis* is the occurrence of (asymptomatic) carrier cats. Cats are considered the major natural host of *M. canis* [4, 5]. The disease is transmitted through direct contact with (sub)clinically infected animals and mechanical carriers of spores, mainly cats. Infection occurs also via indirect contact: brushes, blankets, furniture, clothes, so called fomites, and through contaminated surroundings[4].

As dermatophytes cannot penetrate healthy skin cats can become passive carriers of arthrospores (the infectious product of dermatophytes). Cats who are mechanically carrying spores on their coat are so called 'dust mop cats'. This group of cats is contamined but not infected, however they are a source of infection to other cats. In many cases dust mop cats will remove the spores themselves by grooming [6]. Becoming clinically infected depends on endogenous en exogenous factors such as: age, immune status, skin trauma, hygiene. Dermatophyte spores require (micro) trauma to the skin to access the skin and cause infection, (micro) trauma can be caused by example through fighting, external parasites and even high humidity [6].

Arthospores are highly resistant which makes them difficult to eradicate from the surroundings. In a dry environment they can survive up to twelve months or longer[4].
The incubation period of dermatophytosis caused by *M. canis* is one to three weeks. The infection is mainly at the level of the stratum corneum and the hair shafts, causing hair loss and an inflammatory skin response resulting in alopecia and scaling around the eyes, ears and nose. [4].
Clinical signs are often mild and self limiting with hair loss and scaling. In some cases the infection deteriorates and can become chronic. Dermatophytosis is known for its numerous presentations including the following clinical signs: hair loss, erythema, easily broken hairs, excessive shedding, pruritus, follicular plugging, hyperpigmentation of the skin, hair loss, otitis externa, inflammation of the ear margin, pododermatitis, papules, pustules, "feline symmetrical alopecia", scaling and crusting[6].
There are different diagnostic techniques to diagnose a dermatophytosis. The gold standard for the detection of dermatophytes is a culture on agar with material obtained by using the Mackenzie method (1 minute brushing with a sterile toothbrush). Mackenzie discovered this method in 1960 at a girls' school with fungal infection problems when culturing the hairbrushes of the schoolgirls [7]. Other methods are: Wood's lamp examination, direct microscopic examination of fluorescing hairs and in-house dermatophyte test media [4]. Using the colony forming units (CFU) scoring system/pathogen score system described by Moriello and Newbury, distinction can be made between fomite carriers and truly infected cats. This system uses colony forming units to come to three pathogen scores, Newbury also used this system in her study in 2007[2]. This information along with presence or absence of lesions at the time of culture can help identify culture positive cats and differentiate 'dust mop cats' from truly infected cats [6].
The treatment of dermatophytosis is time consuming and expensive, especially in animal shelters. It consists of treatment of the cats and disinfection of the environment [4]. Although the disease is self limiting, it can have devastating consequences for a shelter that houses groups of animals because of *M.canis* highly transmittable character. A shelter needs to close its facility to the public, cannot let animals be adopted and the zoonotic infection poses a potential risk to the staff. The animal(s) who is/are the source of infection even face the risk of being euthanized if the infection cannot be controlled[6].

Previous studies

Previous other studies are not clear in their description of the materials and methods about the moment of specimen collection and type of housing. One can imagine that the time spent in a facility or in a group of cats contaminated with *M.canis* increases the risk of finding positive cultures and higher prevalences.However, these results are truly incidence numbers and not prevalence numbers. Few real prevalence studies are done. The current study investigates prevalence by examining the cats within 48 hours after intake in the shelter, cats are not moved prior to specimen collection and through swabs taken from the cat quarantine housing, the environmental contamination is known.

Appendix I shows a table with data of previous studies from 1961 to 2011 in which the incidence or prevalence of *M.canis* is investigated. It is not possible to report if the results of the studies are incidences of prevalences as is mentioned in the previous paragraph so it is mentioned as prevalence/incidence here. An overview is given of studies performed with asymptomatic, dermatophytosis suspected and shelter cats. The location, sample size, percentage of *M. canis* positive cats, type of study, type of cat, sampling procedure and culturing procedure are listed to compare between previous studies and the current study. A remarkable difference in incidences/prevalences is seen in this table. The difference between symptomatic and asymptomatic cats is clear because studies with suspected cats take place in a selected group of cats with a greater chance of having actual dermatophytosis. However between similar studies the incidence/prevalence also differs. For example the incidence/prevalence in asymptomatic cats ranges from 0% [8-13] to 88.46% [14]. Different explanations can be brought up to declare this variety: climate, background of the sampled cats, differences in sampling procedures[15], differences in culturing procedures, epidemiological differences between continents and countries, perhaps all plausible explanations and some of these are evidence based. However this variety in outcomes makes it difficult to estimate the expected prevalence in The Netherlands so a selection of European and shelter data is made out of the data from appendix I to predict reasonable prevalence for the Dutch situation.

Because there is no data concerning the situation of feline dermatophytosis in the Netherlands the situation in Europe is surveyed in figure 1.Only in a few countries studies are done as is seen in Figure 1. The studies use asymptomatic and symptomatic cats. That is why there is an extensive variety in incidences/prevalences. Studies in surrounding countries with asymptomatic cats have different outcomes. In Belgium Mignon *et al.* found a prevalence of 15.7% in impounded cats, these cats however lived together in a group and had close contact which is different from the situation in this study. Connole did one of the first studies and found no infections. The group she tested was rather small and statistics were not used in this study. Romano tested street cats in Italy, a country were incidences/prevalences are remarkably higher than in other countries and came to a incidence/prevalence of 47.4%. In 2009 Alpun *et al.* studied homeless shelter cats in Istanbul (n=50) and found a prevalence of 8%.This group is similar to the situation in this study, however the sample size is smaller than other similar studies performed in the United States that is why this study did not use this data in its hypothesis[8, 16-18].

Table 1 lists the studies of (asymptomatic) shelter cats. These numbers were used to come to our hypothesis : *M. canis* in shelter cats is a prevalent infection (>7,6%) at the moment of admission. Newbury, Moriello, Verbrugge and Thomas studied a new treatment protocol for cats infected with *Microsporum canis*. For this study they screened 4019 shelter cats at intake in a shelter in Wisconsin, 304 cats were found positive resulting in a prevalence of 7.6%. The population is comparable to the population present in a Dutch animal shelter and the study is with a large group of animals that is why the hypothesis is based on this study[2]. Woodgyer, Moriello and Boyanowski's results were respectively 6,5%, 4% en 5%, also similar studies to this study only smaller sample sizes. Their outcomes concerning incidence or prevalence (moment of sample collection not clear described in matrials and methods) are lower so it is not unthinkable that the prevalence mentioned in the hypothesis is found too high[19-21].

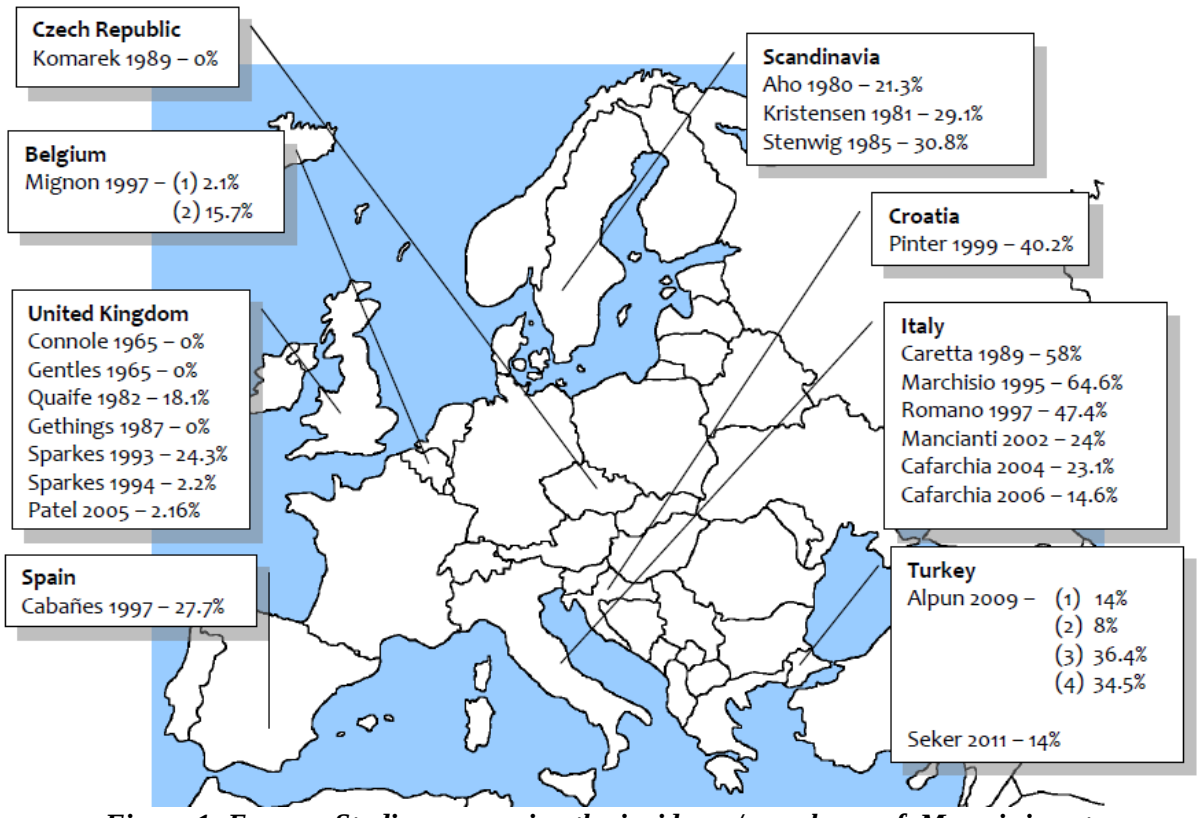


Figure 1: Europe: Studies concerning the incidence/prevalence of M.canis in cats

Table 1: Studies concerning the incidence/prevalence of M.canis in shelter and/or stray cats

Author	Year	Sample size (n)	Location	% *M.canis* positive	Comment
Connole[8]	1965	18	United Kingdom	0%	*Small sample size*
Woodgyer[21]	1977	199	New Zealand	6,5%	*Similar group as this study*
Moriello *et al.*[20]	1994	200	United States	4%	*Warm and cold region, positives in warm region*
Khosravi[22]	1996	100	Iran	26%	*Stray (street) cats*
Mignon *et al.* [18]	1997	134	Belgium	15.7%	*Cats were housed in a group, close contact*
Romano *et al.*[17]	1997	173	Italy	47.4%	*Stray (street) cats*
Boyanowski *et al.* [19]	2000	200	United States	5%	*Similar group as this study*
Newbury *et al.* [2]	2007	4019	United States	7.6%	*Similar group as this study*
Alpun *et al.* [16]	2009	50	Turkey	8%	*Similar group as this study, small sample size*

Materials and Methods

Cats

The research group consisted of 60 cats admitted to "Dierenopvangcentrum (DOC) De Doornakker" in Eindhoven, Noord-Brabant, The Netherlands and examination took place from December 2012 to February 2013. One cat was admitted twice at two different times and is also examined twice. In table 2 is information about the age, sex and breed distribution of the cats in the research group. Appendix II and III show the cat - and laboratory journal with detailed information about the cats and their fungal cultures. Cats included in this study were all considered non-feral (see also 'Character' in Appendix II), feral cats were excluded from the study because of the safety of the cats and investigators. Aggressive cats were examined as good as possible.

Table 2: Age, Sex and Breed distribution research group

Age			Sex		Breed			
Kitten	*Juvenile*	*Adult*	*Male*	*Female*	*European Shorthair*	*Norwegian Forrestcat*	*Maine Coon*	*Persian*
18	9	33	33	27	57	1	1	1

Shelter

The shelter is situated in a suburb in the western part of the city Eindhoven and is one of the biggest shelters in the Netherlands. It rehomes stray animals and animals given up for adoption from the region Brabant South-East which has fourteen municipalities. Cats, dogs, rabbits and rodents live in the shelter. The shelter staff consists of seven fulltime employees and nearly 300 volunteers.

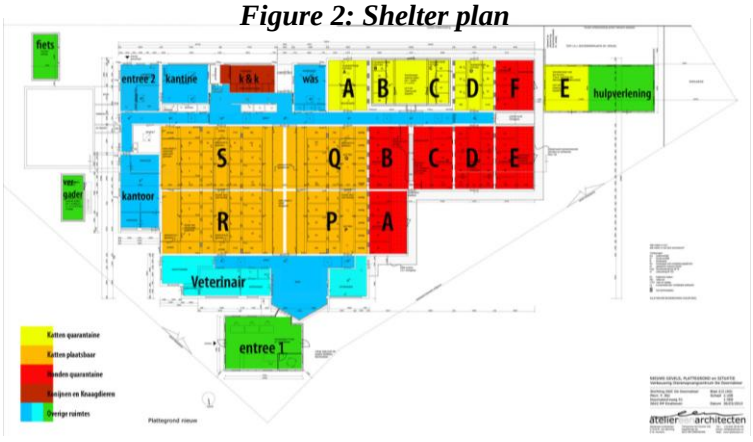
Table 3 shows the number of admitted and clinical (suspected) dermatophytosis cats during last years who are treated for the infection in this shelter. Infection was only based on clinical signs and in-house dermatophyte tests. The data are from the software program of the shelter. It is not known if this were outbreaks or individual cases. The percentage of dermatophytes on the total number of admitted cats is remarkable low concerning the data of previous shelter studies. The shelter staff experiences dermatophytosis not as an extensive problem.

Table 3: Number of admitted cats and dermatophytosis cases

Year	Admitted cats (n)	Clinical dermatophytosis	Percentage%
2012	1335	8	0.60%
2011	1513	5	0.33%
2010	1549	5	0.32%

In 2012 the shelter is renovated to focus on the care for cats, more cat facilities are build at the expense of the dog kennels. Figure 2 is a sketch of the new situation in the shelter. The standard procedure is to keep new admitted cats in quarantine (yellow: B,C,D) for at least two weeks with the exception of the animals surrendered by their owners (yellow: A) as they are completely vaccinated at the time of arrival they are housed in individual open cages together in area A (yellow). Young kittens are homed in foster families. The examination room is situated in the light blue area 'veterinair'.

Figure 2: Shelter plan



Research period
Sampling of the new admitted cats took place from December 2012 to January 2013. These months are in the winter season of the Netherlands (Europe).

General procedures at intake
The cats were subjected to a full general and dermatologic examination after the specimen collection to determine their health status and to check the animal for skin lesions and alopecia suggesting dermatophytosis. All data was recorded in a file (appendix II).
The animals were provided with a shelter number and a name at admission, during the examination the cats all got a microchip to make sure they had a unique identification number for the research and when the animal was adopted the shelter registered it together with the new owner.

Specimen collection
Cats included in this research were at the shelter for a maximum of 48 hours. In these 48 hours they stayed in an individual cage and were moved and touched as minimal as possible in the shelter.

Specimens were collected following a standard procedure:

1) The surfaces in the examination room were cleaned and disinfected using a bleach solution. The solution was made in a small bucket with warm water and a sprout of bleach so the exact solution is unknown. The bleach used contained <5% sodium hypochlorite.
2) The transportation cage was cleaned using a bleach solution.
3) The examiner put on the protective clothing, all disposables:
    a. Overall
    b. Shoe covers
    c. Mob cab
4) The examiner cleaned her hands, first with water and soap and afterwards with antiseptic gel on her dry hands.
5) The cat was transported from its cage to the examination room in the transportation cage.
6) Specimens were collected by using the Mackenzie method[23]. Cats were brushed over the entire body for one minute with a sterile toothbrush (Lactona). After the brushing the toothbrush was placed in a paper envelope and the envelope was closed. Written on the envelope was the name, shelter – and microchipnumber of the cat.
7) The cat was examined and microchipped after specimen collection by performing a broad general and dermatologic exam ("examination and identification", all the data is recorded in appendix II.
8) At the end the cat was examined with the Wood's lamp. The Wood's lamp radiates ultraviolet light with a wavelength of 366 nm which fluoresces hairs containing *M. canis* in 30 to 60% of the cases[24]. Cats were brought to a darkened room and examined from head to tail. The reason this exam took place at the end was because this needed to take place in another (dark) room, the researchers wanted to move the cat as little as possible before specimen collection.
9) The cat was brought back to its own cage. The complete route is seen in figure 3.

Figure 3: Route cat – Specimen collection, Examination, Identification, Wood's lamp

Environmental contamination

To rule out environmental contamination cats were examined within 48 hours of the moment of arrival at the shelter. Cats were kept in strict quarantine for at least two weeks and only kittens from the same litter were housed together in the quarantine. After the quarantine period cats were moved to another area where they were also housed individually (not in social groups). The cats were only moved from their cage to the veterinary area and back for vaccination, castration, examination etc.
Environmental samples were taken with a Cutisoft ® non-woven compress from a visible clean surface. The veterinary examination room and cages of the cats were sampled every two weeks and cultured on Sabouraud B agar and Selective Agar (Biotrading) fungal plates.

Culturing procedure

The culturing procedure took place at the Veterinary Microbiologic Diagnostic Centre (VMDC) of the Clinical Infectiology Division of the Faculty of Veterinary Medicine, Utrecht University.Toothbrush specimens were inoculated on to Sabouraud B agar and Selective Agar (Biotrading) fungal plates. The technical data sheet with the typical formulas is included in appendix IV. The tootbrush was pushed softly into the agar ten times and individual hairs were obtained from the brush by sterile forceps and pressed onto the surface. Fungal colonies resembling dermatophytes were in an early stage subcultured on Sabouraud B agar. In doing so a pure culture of possible dermatophyte colonies were made.
This procedure took place in a ventilated cabinet that was cleaned before, in between and afterward with alcohol. The forceps were sterilized by dipping it into methylated spirit and flaming afterwards.
The plates were incubated at 25˚C for 21 days and examined three times a week, the laboratory journal can be found in Appendix III.

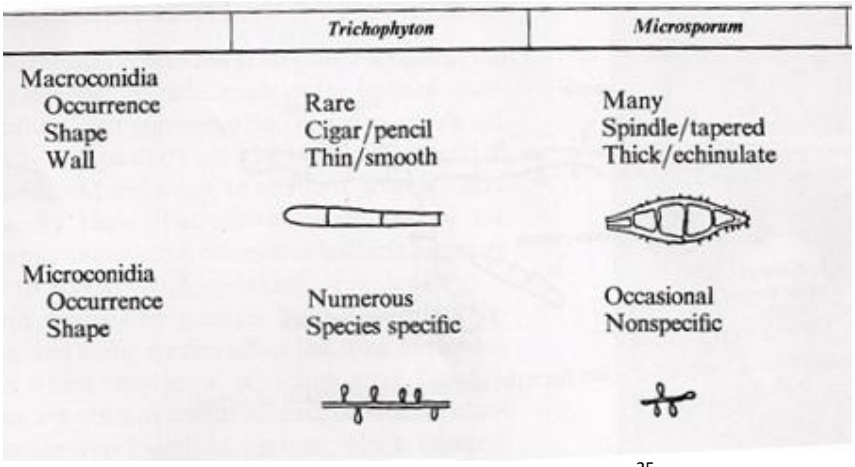


Figure 4: Sporulation characteristics[25]

Suspected colonies were identified microscopically in adhesive tape preparations stained with lactophenol cotton blue. Students were trained to recognize *Microsporum, Trichophyton* and *Aspergillus* macroscopically and microscopically (macro- and microconidia, figure 4)[25]. The staff of the VMDC supported the students with difficult cases and in coming to a final result. At the end cultures were regarded positive or negative for *M.canis*.

Statistical analysis

With the veterinary epidemiology software Win Episcope 2.0 the sample size with a 95% confidence interval was determined for detection of disease with the prevalence and the population of shelter cats in The Netherlands. So statements can be made about the outcome and the hypothesis. In the Netherlands in 2010 and 2011 about 35.000 cats are taken in by Dutch animal shelters of the Dutch Association for the Protection of Animals[26].

Results

Cats

All specimens were cultured in the laboratory of the VMDC for 21 days or until the agar plates were overgrown. The full laboratrium journal is found in appendix III. The health status of the cats is listed in table 4 and in table 5 are the results of the fungal cultures and the number of overgrown cultures.

Table 4: Cats – Health Status

Health Status	Number of Cats (Total: n=60)
Healthy	24
Upper Respiratory Disease	10
Underweight (BCS < 4)	18
Obese (BCS > 6)	5
Heart murmur	2
Fleas (suspected)	2
Pregnant	1
Urotlithiasis	1

Prevalence M.Canis

None of the cats had a *M.canis* positive culture making the prevalence in this group of animals 0%. In table 5 an overview of the results is listed. The full laboratory journal is found in Apppendix III.

In this population of 35.000 shelter cats a sample size of n=60 is sufficient to find a prevalence of 4.9% and higher with a confidence interval of 95% (Win Episcope 2.0). However, the prevalence of 0% cannot be supported by statistics. If the prevalence is lower than 4.9% in this population of animals a bigger sample size is needed to have a significant value, to detect a prevalence of 1% in this population with a confidence interval of 95% the required sample size is 297 animals (Win Episcope 2.0). The sample size used in this is sufficient to make the following statements:

- The prevalence of M.canis in cats admitted to a Dutch shelter is lower than 4.9% within a confidence interval of 95%.
- The hypothesis, '*M. canis* in Dutch shelter cats is a prevalent infection (>7,6%) at the moment of infection', is rejected with the results of this study.

Table 5: Results Fungal Cultures

M.canis (Result)		Overgrown (both fungal plates)		
Positive	**Negative**	**< Day 7**	**< Day 14**	**< Day 21**
0	60	0	12	15

Wood's lamp examination

As is seen in table 6, there were a few cats positive at the Wood's lamp examination. These cats were suspected because of fluorescing spots that were not removable by grooming. However, no positive fungal cultures were found so these results are considered to be false positive and will be discussed later.

Table 6: Results Wood's lamp examination

Wood's lamp examination	Number of Cats
Positive	7
Negative	53

Environmental contamination

Fungi that had grown on the agars of environmental specimens taken every two weeks were not found to be pathogenic. There was fungal growth on all the environmental fungal plates. This technique was used to investigate the environment for fungal contamination and can also be used to evaluate cleaning procedures of the environment after an infection with dermatophytosis.

Research period

December 2012 and January 2013 were cold winter months as is seen in figures 3 and 4, temperatures were often below 0˚C and it was snowing for some days in both months.

Figure 3: Temperature curve December 2012
[27]

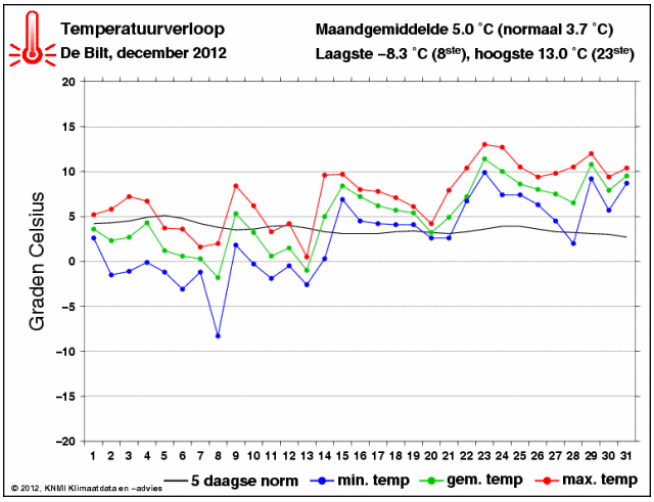
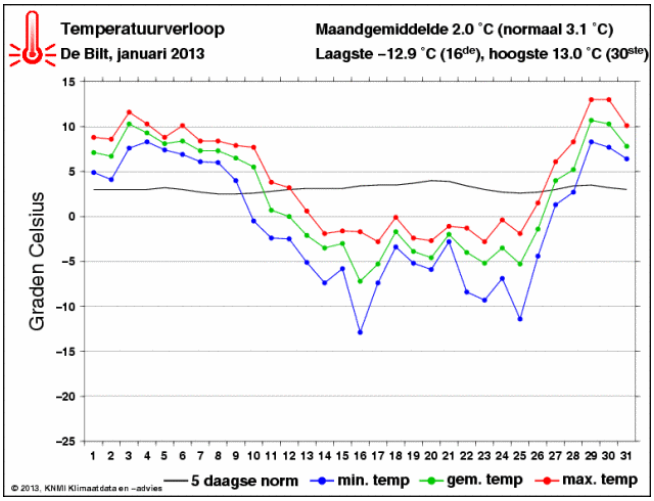


Figure 4: Temperature curve January 2013
[27]



Discussion

Sample size

The sample size of 60 cats is statistically large enough when expecting a prevalence of 7.6% according to previous studies[2]. However regarding the cases of clinical dermatophytosis in the shelter in the last three years as seen in table 3, the hypothesis is perhaps overestimated for this particular situation. This is not surprising because no studies are ever done in The Netherlands so it is difficult to estimate a prevalence without knowledge about the situation. It could have been better to use the information of the shelter although this was not scientific and diagnosis were made based on clinical presentation and in-home fungal cultures. Although based on these facts the estimated prevalence in the hypothesis would be lower, estimated around 1% to include fomite carriers and truly infected cats. As is said before a sample size of 297 cats is needed to test this hypothesis with a confidence level of 95%.

With the sample size of 60 cats prevalences of 4.9% and higher could be detected with a confidence level of 95%. However, if the prevalence is lower in the population of admitted shelter cats in The Netherlands, the sample size was not sufficient to detect it. Further research with a larger sample size over a longer period of time is needed to make statements about the situation concerning the prevalence of *M.canis* in shelter cats in The Netherlands.

Results

There are other studies that found a prevalence of 0% of *M.canis* in cats. However most of these studies are done with similar or smaller sample sizes, complete information is seen in table 1 and Appendix I. [8-10, 12, 13, 28]. There is only one study of Moriello and DeBoer that is done with a larger sample size of 172 healthy pet cats belonging to students, staff and faculty at the University of Wisconsin School of Veterinary Medicine. The fungal flora of the coat of these pet cats was examined to determine the prevalence of *Microsporum canis* carrier status in the population. Moriello and DeBoer found no positive cats for *M.canis*, while they did found one positive sample of *M. gypseum* and one positive sample of *M. vanbreuseghemii*. Their explanations for finding no positive culture for *M. canis* were a difference in climate and environment (Great Lakes region of the United States) with other studies and difference in the population surveyed. The cats in this study belonged to students and employees at a veterinary school and perhaps these cats received different care than other populations of cats in catteries, shelters, living on the street and belonging to the general public. Also showcats who are frequently shampooed and groomed prior to shows may have a changed fungal culture making them more hospitable to *M.canis*[12].
This study is done with admitted shelter cats so a higher prevalence is expected than in healthy pet cats. However the investigators noticed that the cats at intake at the DOC in Eindhoven seemed previously owned by judging their behavior towards people and their appearance. Other studies with shelter cats also examined '"wild" cats and real street cats[17, 22] and found higher prevalences. In this study feral cats were excluded because examination of these animals was not possible without sedation. This could be an explanation for finding no positive culture of *M.canis*. Another explanation for the low prevalence could be the moment of specimen collection in this study. Cats included in this study were not longer than 48 hours in the shelter to rule out environmental contamination. Other studies are not very clear about the moment of specimen collection and type of housing. One can imagine that the time spent in a facility or in a group of cats contaminated with *M.canis* increases the chance of finding a positive culture and higher prevalences.

Sampling period

As is written earlier sampling of the new admitted cats took place from December 2012 to January 2013. December and January were cold winter months as is seen in figures 3 and 4, temperatures were often below 0˚C and it was snowing for some days in both months. According to Kaplan *et al.*, Cafarchia *et al.* and Kristensen *et al. M.canis* is found more frequent in fall and winter[24, 29, 30].
However Kaplan *et al.* also stated that snow covers the fungus and its spores so the snow could be a reason for finding no fomite carriers or infected cats in the shelter in this period[30].

Type of cats

The cats included in this study were considered to be all previous owned by judging their behavior and appearance. However some of the cats were admitted with health problems (mainly upper respiratory disease, condition problems or age problems like urolithiasis), emaciated or had not been groomed in the past by their owner or could not groom themselves due to obesity (table 4, Appendix II). These problems caused the cats to have dirty coats and also stress prevented them from grooming[31].

Wood's lamp examination

Nasal- and ocular discharge fluoresces under the Wood's lamp causing some false "positive" examinations which were not reported as positive in the cat journal (appendix II). Some cats showed fluorescing spots under the Wood's lamp, this may occur due to the application of soap, petroleum and other medicaments [32] or perhaps discharge that was hard to remove and can be considered as false positive because there were no positive cultures. The cats were not washed prior to the examination. The history of the cats before admission to the shelter however is not known which makes it hard to explain the presence of the fluorescing spots. Seven of the sixty cats fluoresced with Wood's lamp examination and all were found negative for *M.canis* in the laboratorium. Wood's lamp examination is perhaps not very usefull in shelters because these cats often carry dirt and discharge on their coats making the Wood's lamp examination hard to evaluate[31].

Overgrowth fungal plates

By using the Mackenzie method for specimen collection a lot of hairs and dirt was collected during the one minute brushing of the dirty cats.This caused a substantial number of culture plates to be overgrown with saprophytic fungi before they reached day 21. The main cause for overgrowth were *Aspergillus* spp[12]. Other research of Moriello and DeBoer studies the fungal flora of the coat of pet cats and found only 79% positive for fungi (thirteen genera of saprophytes and two genera of pathogens (*Microsporum (M.gypseum* and *M. vanbreuseghemii)* and *Trichophyton spp.))*. The most frequent fungal isolates were *Apergillus*, *Cladosporium*, *Penicillium* and *Alternaria* spp. In this study only dermatophytes (*Microsporum* and *Trichophyton*) and *Aspergillus spp*. were microscopically specified. Non-dermatophytes were only determined macroscopically so no statements can be done about the different fungal isolates because they are not microscopically determined. It is interesting to notice that nearly all (116 out of 120 fungal plates, Appendix III) the plates had saprophytic fungal growth and a variety of fungi. Although this study is done with shelter cats and the earlier mentioned study included only healthy pet cat. In the future a similar research as the one from Moriello and DeBoer can be done with Dutch shelter cats, one can expect different outcomes based on the current findings.

Further Research

This research could be the start of new studies investigating the situation of *M.canis* in (shelter)cats in the Netherlands. This research shows that new research, including more cats creating a larger sample size and sampling for a longer period of time (a full year minimal, longer is preferred) is necessary to make statements about the situation in The Netherlands. The population of shelter cats is relatively small and studies including more cats or more shelters are in theory not difficult to perform when time is available to sample for a longer period. It could also be very interesting to research the situation in pet cats and carriers among them in relation to the zoonotic risk of dermatophytosis. A much larger sample size is needed because the population of pet cats in The Netherlands is approximately 2.9 million (2011)[33]. When the testing period is longer than one full year the chance that significant differences in different risk factors like season and age will be observed is greater.

Another interesting research is examining the fungal flora of the coat of (shelter) cats, as said before. The variation and quantity of fungi found is this study is different from other studies and interesting to investigate further. However a mycologist is needed to identify the different fungal isolates making it an expensive and time consuming study.

Conclusion

The aim of this study was to evaluate the risk of introduction of *M.canis* into Dutch animal shelters by testing cats during two months at the time of admission to a shelter in The Netherlands. The results led to a prevalence of *M.canis* of Dutch shelter cats at the moment of admission. Because no fungal cultures were found positive for dermatophytes and in particular *M.canis* the outcome of this study is a prevalence of 0%. The sample size of 60 cats is statistically sufficient to find prevalences of 4.9% and higher with an confidence interval of 95%. We can conclude that the found prevalence is not statistically supported and can be as high as 4.9%. Further research over a longer period of time and with larger sample sizes is needed to come to more accurate results on the situation concerning the prevalence of *M.canis* in shelter cats in The Netherlands.

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Appendix

Appendix I Studies concerning the incidence/prevalence of M.canis from 1961 to 2011

Author	**Year**	**Location**	**Sample size (n)**	**% *M.canis* positive**	**Type of study**	**Type of cat***	**Sampling procedure**	**Culturing procedure**
Kaplan *et al.* [30]	1961	USA	1232	29.5% (363)	Submission to diagnostic lab	C	Hair and skin scrapings	Selective medium containing cycloheximide (Acti-dione), penicillin and streptomycin, 25˚C, 1 month
Keep [34]	1963	Sydney, Australia	1059	5.9% (63)	Patients Syndney University Clinic	A, C	Plucked hairs **only** of Wood's lamp positive cats	Sabouraud's medium
Connole [8]	1965	Glasgow, Scotland, Europe	18	0%	Submitted to veterinarian at receiving home	A, S	Hairbrush sampling technique	4% malt extract agar incorporating penicillin (20µg/ml), streptomycin (40µg/ml) and with or without cycloheximide ("Actidione", 0.5mg/ml), 28˚C, 21 days, Subculture: malt extract agar

Gentles *et al.* [10]	1965	Glasgow, Scotland, Europe	30	0%	Submitted to veterinary clinic	A, C	Hairbrush sampling technique	4% malt extract agar supplemented with cyclohedimide (0.5g/L) penicillin (20units/ml) and streptomycin (40units/ml), 24˚C
Al-Doory *et al.*[35]	1968	Texas, USA	6	50% (3)	Submitted to veterinary clinic	C	Skin scrapings	Mycosel agar, 25˚C, 21 days
Baxter [36]	1973	Palmerston North, New Zealand	200	36% (72)	Visitors veterinary clinic randomly selected	A	Brushing with a plastic brush	Sabouraud's dextrose agar containing 0.05% chloramphenicol and 0.5% cycloheximide, 25˚C, 14 days
Woodgyer [21]	1977	Wellington, New Zealand	199	6,5% (13)	Cats SPCA at veterinary clinic	A, S	Ive's modification of the Mackenzie method	Sabouraud's dextrose agar with 0.5% cycloheximide (Actidione) and 0.05% chloramphenicol, 27˚C, 14 days
Aho** [37]	1980	Scandinavia, Europe	61	21.3% (13)	Unknown	C	Unknown	Unknown
Kristensen *et al.*[24]	1981	Denmark, Europe	227	29.1% (66)	Submission to diagnostic lab	C	Hairs and skinscrapings	Sabouraud's and Selective agar (Merck), 28˚C, 21-25 days
Quaife *et al.*[38]	1982	UK, Europe	216	18.1% (39)	Show cats at four cat shows	A	1) Fur plucked	Ink blue antibiotics, 26˚C,

							from the dorsum 2) Brushing with nylon nail brush	21 days, Subculture: Sabouraud's agar containing cycloheximide (actidione)
Stenwig [39]	1985	Norway, Europe	279	30.8% (86)	Submission to diagnostic lab	C	Skinscrapings	Sabouraud dextrose agar and Mycobiotic agar (Difco) both containing 5µg chloramphenicol per ml, 30˚C, 14 days
Zaror *et al.*** [14]	1986	Brazil	104	88.46% (92)	Healthy housecats	A	Unknown	Unknown
Gethings *et al.*[9]	1987	South-west England, UK, Europe	51	0%	Farm cats on 22 sheep farms	A	Sterile 20cm plastic scalp brush, rigorously combed	Standard Sabouraud, 27˚C, 10 days
Komarek *et al.* *** [11]	1989	Prague, Czech Republic, Europe	32	0%	Unknown	A	Mariat and Tapia method	Unknown
Thomas *et al.*** [28]	1989	North Carolina, USA	1) 50 2) Unknown	1) 0% 2) 29%	1) Healthy pet cats 2) Cattery	1) A 2) C (history of dermatophytosis)	Toothbrush	Unknown
Caretta *et al.*[40]	1989	Tuscany, Italy, Europe	93	58% (54)	Submitted to the Department of Animal Pathology, Veterinary Medicine, Pisa University, suspected fungal disease	C	Sterile fine metal comb	Sabourauds dextrose agar containing 20 units of penicillin, 40 of streptomycin, 0.05mg

								chloramphenicol and 0.5mg cyclohexidine/ml, 28˚C, 4 months, Subculture: Sabouraud's, cornmeal agar with 1% dextrose and Emerson's agar medium
Moriello *et al.*[12]	1991	Winsconsin, USA	172	0%	Healthy pet cats belonging to students, staff and faculty at the University of Wisconsin School of Veterinary Medicine	A	Sterile toothbrush, 3 minutes	Sabouraud dextrose agar and Dermatophyte Test Medium (DTM), 23-26˚C, 30 days, Subculture: ricegrain medium, 21 days
Lewis *et al.*[41]	1991	Louisiana, USA	408	13.7% (56)	Submission to diagnostic lab	C	"in-house" submissions	"in-house" submissions
Moriello *et al.*[13]	1991	Midwestern USA	1) 118, cats from catteries with a history of dermatophytosis 2) 58, cats from catteries without a history of dermatophytosis	1) 100% when resampled 2) 0%	Catteries with a long standing problem of dermatophytosis and catteries with no history of dermatophytosis	A, C	Sterile toothbrush, 3 minutes	Sabouraud glucose agar and Dermatophyte Test Medium (DTM), 23-26˚C, 30 days. Subculture: rice grain medium, 21 days

Sparkes *et al.* [32]	1993	ÙK, Europe	3407	24.3% (827)	Submitted to diagnostic lab	C	Samples of hair submitted to lab	Sabouraud's dextrose agar containing cycloheximide (0.04%) and chloramphenicol (0.005%), 30˚C, 14 days. Subculture: Takashios' agar
DeBoer *et al.*[42]	1993	Midwestern USA	99	16.2% (16)	Seven show catteries and control group (lab colony, healthy cats)	A, C (all positive cats from endemic catteries 3 cats with skin lesions, other cats history of dermatophytosis in previous year)	Sterile toothbrush, 3 minutes	Dermatophyte Test Medium plates, 25˚C, three weeks
Gambale *et al.* ** [43]	1993	Brazil	100	8% (8)	Unknown	A	Unknown	Unknown
Moriello *et al.*[20]	1994	Pennsylvania, New York, Wisconsin, Florida, USA	200 (100 cold dry climate, 100 warm humid region)	4% (8), all from warm humid region	Shelter cats, apparently healthy, from animal shelters in two different geographic regions	A, S	Sterile toothbrush	Sabouraud glucose agar and Dermatophyte Test Medium (DTM), 23-26˚C, 30 days. Subculture: rice grain medium, 21 days
Sparkes *et al.*[44]	1994	Bristol, UK, Europe	181	2.2% (4)	Healthy pet cats attending a veterinary clinic	A	Modified Mackenzie hair brush technique	Sabouraud's dextrose agar containing cycloheximide (0.04%) and

								chloramphenicol (0.005%), 30˚C, 21 days. Subculture: Takashios' agar
Marchisio *et al.*[45]	1995	Turin, Italy, Europe	105	64.6% (53)	Suspected pet cats attending the Medical Clinic of the Veterinary Faculty, University of Turin	C	Samples from lesions (hairs, skin scrapings, contents from pustules or vesicles)	Dermasel (Oxoid) agar supplemented with 0.4 g l^{-1} cycloheximide, 24˚C, 15 days followed by 15 days at room temperature. Subculture: Sabouraud glucose agar with yeast extract (5 g l^{-1}) or Borelli Lactrimel agar
Simpanya *et al.*[46]	1996	Palmerston North, New Zealand	178	18.5% (33)	SPCA premises, Palmerston North	A, C	Bruhing with plactic brush	Sabouraud dextrose agar containing antibiotics, 25˚C, 21 days
Khosravi [22]	1996	Isfahan, Iran	100	26% (26)	Stray cats from different districts of the city	A (n=96), C (n=4, all positive), (S)	Sterile toothbrush, 3 minutes	Sabouraud glucose agar and Dermatophyte Test Medium (DTM), 26˚C, 30 days. Subculture: rice grain medium, 21 days
Mignon *et al.*[18]	1997	Belgium, Europe	1) 467 2) 134 3) 27	1) 2.1% 2) 15.7% 3) 100%,	1) Healthy pet cats belonging to veterinary students	A, C, S	Sterile toothbrush	Sabouraud glucose agar with cycloheximide

			4) 4	4) 100% In group 3 and 4 one infected cat was responsible for all the positive cultures of the other cats	2) Impounded cats 3) Stray cats in a private refuge 4) Longhaired pet cats living together			(0.2%) and chloramphenicol (0.05%), 28˚C, 15 days. Subculture: Takashio's agar
Cabañes *et al.*[47]	1997	Barcelona, Spain, Europe	56	27.7% (18)	Submitted to diagnostic lab	C	Plucked hairs and scraped scales	Mycosel agar, 28˚C, 1 month
Romano *et al.*[17]	1997	Siena, Italy, Europe	173	47.4% (82)	Asymptomatic stray cats	A, (S)	Mackenzie method	Sabouraud glucose agar with chloramphenicol and cycloheximide, 25˚C, 21 days. Subculture: Sabouraud medium or DTM.
Pinter *et al.*** [48]	1999	Croatia, Europe	1838	40.2% (738)	Cats with suspected skin disease	C	Unknown	Unknown
Boyanowski *et al.* [19]	2000	West coast, USA	200	5% (10)	Shelter cats from four different geographical regions	A, S	Modified Mackenzie method	Potato flake agar with 0.4 mg mLl^{-1} cycloheximide and chloramphenicol 0.05 mg mL^{1}, 25˚C, 30 days. Subculture: potato flake agar without

								added antibiotics
Mancianti *et al.*[49]	2002	Tuscany, Italy, Europe	7650	24% (1836)	Submitted to diagnostic lab	C	Mackenzie's brush technique	Mycobiotic agar (Difco, USA), 25˚C, 15 days
Khosravi *et al.*[50]	2003	Iran	186	47.8% (89)	Submitted to diagnostic lab	C	Different hair and skin specimens	Sabouraud dextrose agar incorporating cyclomeximide (0.04%) and chloramphenicol (0.005%), 30 and 37˚C, 1 month
Brilhante *et al.*[51]	2003	Northeast Brazil	38	36.8% (14)	Submitted to diagnostic lab	C	Skin scrapings, nail fragments, plucked fur, scales from lesions	Sabouraud dextrose agar with or without choloramphenicol and Mycosel agar, 28˚C, 1 month. Subculture: rice medium, Borelli's lactrimel agar
Cafarchia *et al.* [29]	2004	South-Italy, Europe	156	23.1% (36)	Samples from practitioners and the Faculty of Veterinary Medicine of the University of Bari	C	Hair and skin scrapings	Mycobios Selective agar (Biolife®), 24˚C, 15 days. Subculture: potato glucose agar (Biolife®).
Patel *et al.*[52]	2005	Southeast England, UK, Europe	169	2.16% (3)	One closed colony (n=30) and cats brought into a first-opinion clinic for routine vaccination or elective surgery (n=139)	A, (S)	Brushing using a sterilized human scalp brush	Sabouraud's dextrose agar containing Actidione and chloramphenicol, 32˚C, 21 days
Cafarchia *et*	2006	South-Italy,	1) 56	1) 53.6% (30)	*1)* Cats belonging to	A	Brush	Sabouraud agar

al.[53]		Europe	2) 192	2) 14.6% (28)	individuals affected by tinea corporis caused by *M.canis* 2) Cats belonging to individuals without tinea corporis caused by M.canis		technique, 8cm diameter plastic brush for at least 3 minutes	with chloramephenicol (0.5%) and actidione (0.4%), 25˚C, 15 days.
Newbury *et al.*[2]	2007	Wisconsin, USA	4019	7.6% (304)	Population of cats surrendered to the Dane County Humane Society (Shelter)	A, C, S	Tootbrush	Mycosel agar modified with phenol red as colour indicator, 25-27˚C, 21 days
Alpun *et al.*[16]	2009	Istanbul, Turkey, Europe	1) 50 2) 50 3) 33 4) 29	1) 14% (7) 2) 8% (4) 3) 36.4% (12) 4) 34.5% (10)	Specimens from veterinary clinics and shelters located in Istanbul. 1) clinically healthy/owned, 2) clinically healthy/ownerless, 3) suspected dermatophytosis owned, 4) suspected dermatophytosis/ownerless	A, C, S	Scrapings from lesions and brushing with sterile brush	Sabouraud dextrose agar and Dermatophyte Test Medium (DTM), 25˚C, 21 days, DTM 14 days
Seker *et al.*[54]	2011	Western Turkey, Europe	164	14% (23)	Samples sent in by veterinary practitioners from veterinary clinics and shelters, submitted to diagnostic lab	C, S	Hair and skin scrapings from lesions	Sabouraud dextrose agar medium containing 0.5mg/ml chloramphenicol, 25˚C, 21 days.

* ***A*** *= Asymptomatic,* ***C****= Clinical signs,* ***S*** *= Shelter/Stray cats*

*** Information acquired from the abstract of the article and review articles, the full text of these articles was not available via the University Library of the University Utrecht*

**** Article in Chezch, information from English abstract*

Appendix II Cat journal

Cats - Identification and Description

Number	Name	Admission	Examination	Hours in Shelter	Shelter number	Origin	Breed	Sex	Age	Hairlength
1	Fyta	10-12-2012	11-12-2012	17.5	19731	Stray	ES	F	A	S
2	Emile	13-12-2012	14-12-2012	25.5	19736	Stray	ES	M	K	S
3	Bono	15-12-2012	17-12-2012	36.1	19748	Stray	ES	M	J	S
4	Eastwood	18-12-2012	19-12-2012	17.5	19753	Given up for adoption	ES	M	A	S
5	Forrest	18-12-2012	19-12-2012	18	19752	Given up for adoption	ES	M	A	S
6	Mees	20-12-2012	21-12-2012	20	19759	Stray	ES	M	K	S
7	Mars	20-12-2012	21-12-2012	20	19760	Stray	ES	M	K	S
8	Nonna	21-12-2012	21-12-2012	2	19761	Stray	ES	F	K	S
9	Mylo	22-12-2012	23-12-2012	18.5	19765	Stray	ES	M	K	S
10	Athos	28-12-2012	29-12-2012	22	19776	Given up for adoption	ES	M	K	S
11	Coco	28-12-2012	29-12-2012	22	19777	Given up for adoption	ES	M	K	S
12	Bjorn	28-12-2012	29-12-2012	5	19780	Stray	ES	M	J	S
13	Isis	28-12-2012	29-12-2012	22	19778	Given up for adoption	ES	F	K	S
14	Mia	28-12-2012	29-12-2012	22	19779	Given up for adoption	ES	F	K	S
15	Vandor	29-12-2012	30-12-2012	23	19781	Stray	ES	M	A	S
16	Poekel	30-12-2012	30-12-2012	7	19782	Stray	ES	M	A	S
17	Frits	30-12-2012	30-12-2012	2	19785	Stray	ES	M	J	S
18	Phil	4-1-2013	5-1-2013	19.5	19796	Stray	ES	M	K	S
19	Bibi	6-1-2013	7-1-2013	19	19801	Stray	ES	F	K	S
20	Stuart	6-1-2013	7-1-2013	17.5	19800	Stray	ES	M	A	S
21	Quinty	7-1-2013	7-1-2013	0	19764	Stray	ES	F	K	S
22	Timmy	9-1-2013	10-1-2013	21	19807	Stray	ES	M	K	S
23	Ayla	9-1-2013	10-1-2013	21	19808	Stray	ES	F	K	S
24	Leyla	9-1-2013	10-1-2013	21	19809	Stray	ES	F	K	S
25	Dewi	9-1-2013	10-1-2013	21	19810	Stray	ES	F	K	S
26	Saar	9-1-2013	10-1-2013	21	19811	Stray	ES	F	K	S
27	Bruce	9-1-2013	10-1-2013	24	19806	Stray	ES	M	K	S
28	Ditch	9-1-2013	10-1-2013	22	19812	Stray	ES	M	A	S

29	Simon	10-1-2013	11-1-2013	18	19818	Stray	ES	M	A	S
30	Polar	10-1-2013	11-1-2013	21	19816	Given up for adoption	Norwegian FC	F	A	L
31	Elmo	11-1-2013	11-1-2013	1	19819	Stray	ES	M	J	S
32	Sultan	12-1-2013	13-1-2013	20	19826	Stray	ES	M	A	S
33	Sjors	13-1-2013	14-1-2013	25	19830	Stray	ES	M	A	S
34	Faya	14-1-2013	15-1-2013	17	19839	Stray	ES	F	A	S
35	Diddle	17-1-2013	19-1-2013	41.5	19844	Stray	Maine Coon x	M	A	M
36	Sylvester	17-1-2013	19-1-2013	42	19847	Stray	ES	M	A	S
37	Puck	18-1-2013	19-1-2013	21	19849	Given up for adoption	ES	F	A	S
38	Amy	18-1-2013	19-1-2013	19	19850	Stray	ES	F	A	S
39	Jackson	19-1-2013	20-1-2013	22	19852	Stray	ES	M	J	S
40	Emily	24-1-2013	25-1-2013	24	19861	Stray	ES	F	A	S
41	Frans	24-1-2013	25-1-2013	21	19862	Stray	ES	M	A	S
42	Enzia	25-1-2013	25-1-2013	5	19863	Given up for adoption	ES	F	A	S
43	Garfield	26-1-2013	27-1-2013	21	19867	Stray	ES	M	A	S
44	Bjorn (2)	26-1-2013	27-1-2013	21	19780	Stray	ES	M	A	S
45	Shari	26-1-2013	27-1-2013	22	12880	Given up for adoption	ES	F	J	S
46	Teske	26-1-2013	27-1-2013	24	18944	Given up for adoption	ES	F	A	S
47	Tora	26-1-2013	27-1-2013	24	18945	Given up for adoption	ES	F	J	S
48	Lassie	29-1-2013	30-1-2013	18	3796	Given up for adoption	ES	F	J	S
49	Byeol	29-1-2013	30-1-2013	20	19871	Given up for adoption	ES	F	A	S
50	Jang-Soo	29-1-2013	30-1-2013	20	19872	Given up for adoption	ES	M	A	S
51	Ventoux	29-1-2013	30-1-2013	18	19874	Given up for adoption	ES	F	A	S
52	Melon	29-1-2013	30-1-2013	18	19875	Given up for adoption	ES	F	A	S
53	Emiel	30-1-2013	1-2-2013	37	19880	Stray	Persian	M	A	L
54	Zyra	2-2-2013	4-2-2013	43	19885	Given up for adoption	ES	F	A	S
55	Tybo	2-2-2013	4-2-2013	43	19886	Given up for adoption	ES	M	A	S
56	Fee	3-2-2013	4-2-2013	24	19889	Stray	ES	F	A	S
57	Aurora	2-2-2013	4-2-2013	46	19882	Stray	ES	F	A	S
58	Mr. X	4-2-2013	5-2-2013	21	19891	Stray	ES	M	J	S
59	Misty	8-2-2013	8-2-2013	2.5	19905	Given up for adoption	ES	F	A	S
60	Donald	8-2-2013	8-2-2013	2	19906	Stray	ES	M	A	S

ES = European Shorthair, Age: K=Kitten (0-6 months), J=Juvenile (6-12 months), A=Adult (> 1 year), Hairlength: S=Short, M=Medium, L=Long

Cats – Examination

Number	Weight (kg)	BCS (1-9)	Breathing	Circulation	Temperature	Other	Dermatology	Wood's lamp	Skin Lesions	Character
1	3.09	3	28, CA	140, KRES	38.5	Bad teeth	Tattered coat, flee poop, loose hair	Positive spot on the right ear	No	Nice, but painful on palpation head area
2	1.25	3	24, CA	156, KRES	38.6	Full abdomen, Nose & eyes are dirty	Some scaling	Negative	Alopecia on left ear	Very nice
3	1.70	3	30, CA	240, KRES, murmur 4/6	not examined	Conjunctiva red OS and protrusion MN	Smooth, shiny, ears: earmite	Negative	No	Very nice
4	4.8	7	20, CA	240, KRES	not examined	Urolithiasis (bladder)	Scaling, loose hairs (severe)	Negative	No	Nice, anxious
5	4.30	5	20, CA	200, KRES	not examined	Shortening of the left ear (scarring)	Scaling, loose hairs (mild)	Negative	No	Nice, anxious
6	1.68	4	64, CA	180, KRES	not examined	Brother of Mars, together in cage	Smooth, shiny coat, no scaling	Negative	No	Nice
7	1.65	4	32, CA	210, KRES	not examined	Brother of Mees, together in cage	Smooth, shiny coat, no scaling	Negative	No	Nice
8	2.5	4	20, CA	240, KRES	38.2	-	Smooth, shiny coat, no scaling	Negative	No	Nice
9	1	3	30, CA	240, KRES	38.5	Upper respiratory disease, Fleas	Dull, dirty and loose hairs (moderate), fleas	Negative	No	Nice
10	2.2	5	22, CA	200, KRES	not examined	Brother of Coco, Isis and Mia	Smooth, shiny coat, no scaling	Negative	No	Nice
11	2.5	5	50, CA	240, KRES	not examined	Bother of Athos, Mia and Isis	Smooth, shiny coat, no scaling	Negative	No	Nice
12	3.33	4	50, CA	180, KRES	not examined	Fleas, itching ear	Scaling, loose hairs (severe)	Negative	Alopecia on back, left	Angry
13	1.65	4	50, CA	240, KRES	not examined	Sister of Athos, Mia and Coco. Tense abdomen.	Smooth, shiny coat, no scaling	Negative	No	Nice
14	1.95	4	60, CA	204, KRES	not examined	Sister of Athos, Isis and Coco	Smooth, shiny coat, no scaling	Negative	No	Nice

15	2.5	2	36, CA	205, KRES	not examined	Pupil deformation, missing piece in both eyes OS, crests on nose	Loose hairs, crest on planum nasale,	Negative	No	Nice, timid
16	3.0	3	60, CA	266, KRES	not examined	Dirty ears, wet paws, probably blind	Tattered coat, crests on nose, loose hairs (severe)	Negative	No	Nice, anxious
17	2.76	4	36, CA	156 on auscultation	not examined	-	Smooth, shiny coat, no scaling	Negative	No	Very nice
18	1.9	3	Purring	204 on auscultation	not examined	Tearflow OD, nasal discharge (serous), dirty ears	Dull, smooth coat. Alopecia of the right ear, left lateral hindpaw, right lateral hock	Positive hairpoints on top of the head, cannot be erased by grooming	Alopecia of the right ear, left lateral hindpaw, right lateral hock	Nice, playful
19	2.02	4	54, CA	240 on auscultation	not examined	Lnn: left mandibular bigger than the right mandibular	Smooth, shiny coat, no scaling	Negative	No	Nice
20	3.75	4	28, CA	150, KRES	not examined	Dirty ears, foetor ex ore	Tangled hairs around the neck region and caudodorsal, a lot of tangled hairs, scaling (mild), Dull coat, alopecia caudal to the whiskers (probably fighting)	Negative	Alopecia caudal to the whiskers (probably fighting)	Very nice
21	1.25	4	60, CA	not examined	not examined	-	Alopecia: right frontleg, right hindleg (3th toe), head cranial to the right ear. Dirty hairs on the left frontleg	Negative	Alopecia: right frontleg, right hindleg (3th toe), head cranial to the right ear	Very nice, active
22	1.91	3	60	240, KRES	not examined	Group of kittens 9-1-2013	Smooth, shiny, no alopecia	Negative	No	Nice, playful
23	1.75	3	60	240, KRES	39,4	Group of kittens 9-1-2013	Smooth, shiny, no alopecia	Negative	No	Nice
24	1.85	3	60	240, KRES	not examined	Group of kittens 9-1-2013	Smooth, shiny, no alopecia	Negative	No	Nice, quiet

25	2	4	60	216, KRES	not examined	Group of kittens 9-1-2013	Smooth, shiny, no alopecia	Negative	No	Nice, quiet
26	2.23	4	60, superficial	240, KRES	not examined	Group of kittens 9-1-2013	Loose hairs, smooth, shiny	Negative	No	Nice, quiet
27	2	4	purring	144, KRES	not examined		Some scales, smooth, shiny, itch of the ear, fleas	Negative	No	Nice, active
28	-	4	not examined	not examined	not examined	Found in the water: Hypothermic and in shock, received an injection of Dexamethason	Not examined	Not examined	Not examined	Very anxious and stressed
29	4.42	5	24, CA	192, KRES	not examined	-	Smooth, shiny, no alopecia	Negative	No	Nice, playfull and active
30	3.66	4	60, CA	108, KRES	not examined	-	Fluffy, a lot of tangled hairs, some scaling, smooth, a little bit dull	Negative	No	Very nice, anxious and stressed
31	2.7	4	40, CA	108, KRES	not examined	Was found poured with gasoline and burning marks on the paws	Smooth, shiny, no alopecia and scaling	Negative	No	Nice, timid
32	3.99	5	50, CA	198 KRES	not examined	-	Loose hairs, scaling (mild)	Negative	No	Nice
33	5.8	7	66, CA	200, purring	not examined	Might have been hit by car. Was walking poorly when admitted to shelter; now he's doing fine	Lots of loose hairs, scaling (severe), some little (fight) wounds. Lot of tangled hairs	Negative	No	Littlebit scared, stressed
34	2.3	2	50, superficial	200, KRES	not examined	Dirty eyes and nose, suspect of calici or other upper respiratory virus. Treatment with doxy has been started yesterday	Fluffy, not shiny, long nails	Green on head, ears and back. Might be saliva or from eyes/nose. Partly on hair shafts too.	No	Nice, scared. Examined in cage.
35	3.1	2	24, CA	140	not examined	Dirty eyes,	Fluffy, tangled hairs	Positive point	Alopecia right	Stressed, not well

						sticking out the tongue		on the tip op the ear and the right ear	ear	examinable
36	3.93	5	60	120, KRES	not examined	Suspicion of fleas	Smooth, shiny	Negative	No	Very nice and playfull
37	-	6	36	not examined	not examined	-	Dull, dirty, a lot of loose hairs	Not examined	No	Very scared, agressive
38	2.94	5	24	180, KRES	not examined	-	Smooth, shiny, no alopecia	Negative	No	Nice, curious
39	3.0	3	36, sniffing	160, KRES	not examined	Upper respiratory problems, sniffing and sneezing	Dull, fluffy, no alopcia	Negative	No	Very nice
40	3,6	6	60	220	not examined	Pregnant	Smooth, shiny, no alopecia	Negative	No	Very scared, agressive
41	2.24	1 to 2	60	not examined	not examined	Very severe upper respiratory disease complex (treatment with doxycycline)	Dull, fluffy, dirty - the cat was just washed with animal shampoo	Negative (head fluroisced green because of the discharge and crusts)	Alopecia, little wound on the right frontpaw	Nice
42	2.05	3	40	160	not examined	-	Smooth, shiny, no alopecia	Not examined	No	Extremely stressed, anxious, not examinable, earlier that day seen by shelter vet who did a general exam)
43	6.7	8 to 9	54	198, on auscultation	not examined	Dirty eyes, dirty ears and earmargins	Very severe felt fomation on the back probably due to no grooming, almost like there is a second get on the back of this cat, dirty smelly, greasy coat	Negative	No	Obese, not active, stressed
44	see 12	see 12	see 12	see 12	see 12	see 12	see 12	see 12	see 12	see 12
45	4.2	7	28, CA	180, on auscultation	not examined		Smooth, shiny, no alopecia	Negative	No	Stressed
46	2.4	3	30, CA	150, KRES	not examined	Sister of Tora	Smooth, shiny, no alopecia	Negative	No	Nice
47	2.65	4	48, CA	180, on auscultation	not examined	Sister of Teske	Smooth, shiny, no alopecia, some loose hairs	Negative	No	Nice

48	4.12	5	40, CA	180, KRES	not examined	Heartmurmur 3/6	Smooth, shiny, some loose hairs	Negative	No	Nice, stressed
49	2.79	4	28	126, KRES	not examined	-	A lot of loose hairs, moderate shiny and smooth	Negative	No	Stressed
50	5.05	8	44	198 on auscultation	not examined	-	Smooth, shiny, a lot of loose hairs	Negative	No	Nice, stressed
51	3.62	5	80, superficial	240	not examined	-	Smooth, shiny, moderate loose hairs	Negative	No	Scared, agressive when forced
52	3.52	4	32, CA	240, KRES	not examined	-	Smooth, shiny, a lot of loose hairs	Negative	No	Nice
53	4.5	5	32, CA	180, KRES	not examined	Severe tangled hairs, shaved incorrect, dirty ears, wound on the right ear	Very dirty coat, a lot of tangled hears, needs to be shaved	Doubtfull (because of all the dirt in the coat a lot of colors under Wood's lamp examination)	Wound on the right ear	Very nice
54	3.2	4	30	200, KRES	not examined	-	Smooth, shiny, a lot of loose hairs	Negative	No	Nice
55	4.2	5	40, CA	240, KRES	not examined	-	Smooth, shiny, a lot of loose hairs	Negative	No	Nice, stressed
56	2.9	2	24, CA	250, KRES	not examined	Fleas	Not shiny and smooth, dull, a lot of loose hairs	Spot on the ear (dirty)	No	Nice
57	2.5	3	40, CA	170, KRES	not examined	Severe upper respiratory disease complex	Dull and dirty	Negative	No	Nice
58	3.65	5	50, CA	150, KRES	not examined	-	Smooth and shine, moderate loose hairs	Green spot above the right eye	No	Nice
59	3.6	5	72, superficial	240, KRES	not examined	-	Smooth and shiny	Negative	No	Nice, stressed
60	4.92	6	40,CA	192, KRES	not examined	Scarring on the ears, crusts on his head, probably from fighting	A lot of loose hairs and moderate scaling, crusts on the head, probably from fighting	Negative	Scarring on the ears	Nice

Appendix III Laboratorium journal

No.	Result	Final day - Overgrown day	Culture Date	Inocu-lation Date	Check 1	Check 2	Check 3	Check 4	Check 5	Check 6	Check 7	Check 8	Check 9
1	Negative	Day 21: no pathogenic fungal growth	12-11-2012	12-11-2012	Day 2: both negative	Day 6: Sabouraud agar white colony (+/- 2 cm), not suspect, microscopy: aspergillus	Day 8: Sabouraud and Selective agars colonies of Aspergillus (microscopy)	Day 11: Sabouraud - filled with white colonies, no suspicion of dermatofytes . Determintation: Aspergillus. Selective agar - 6 colonies, 4 green, 2 white --> Determination: unsure.	Day 13: see day 8. No pathogen funghi found. No clear determination.	Day 16: See day 13	Day 19: Selective Agar - 5x greenbrown 3mm-1cm, 2x white powdery, 2 cm. Sabouraud - white/yellow, completely full, looks like stucwork.	Day 21: idem day 19	
2	Negative	Sabouraud: day 16 - overgrown Aspergillus, Sabouraud B plate: day 19 overgrown Aspergillus, Selective agar: Day 21 no pathogenic fungal growth	12-14-2012	12-14-2012	Day 3: both negative	Day 5: Sabouraud agar 2 small colonies, suspected, not determinable.	Day 8: Sabouraud - 4 white colonies. Determination: 1x aspergillus, 1x unsure. Sel. Agar - 1 small white colony	Day 10: selective agar - aspergillus. Sabouraud - Snowball next to red aspergillus -> re-cultured on a new Sabouraud.	Day 13: selective agar - same as day 10. Sabouraud - overgrowth Aspergillus. B-plate: 3 colonies, probably no dermatophyt.	Day 16: Sabouraud - overgrowth Aspergillus (garbage!) Selective agar - 1x white, rounded 2.5cm, 2x white, 1cm, 1x carrotorange 1 cm. B-plate: Overgrowth Aspergillus? 2x white, 1 cm	Day 19: Sel. Agar - 3x green 1-3cm, 1x orange 1cm, 1x white 2cm, Sab. B plate - green overgrowth	Day 21: Sel. Agar: green islet, 1x white, 1x orange	
3	Negative	Selective agar and Sabouraud: Day 19 overgrown black fungus, no pathogen, Sabouraud B plate: no pathogenic fungal growth, aspergillus pollution	12-17-2012	12-17-2012	Day 2: both negative	Day 5: Sabouraud - multiple yellow-white mini colonies. Sel.agar- 1 green colony	Day 7: same as day 5. Determination: bacteria's and doubt. Re-culture on a new Sabouraud.	Day 10: Sabouraud and Selective agar: yellow mini colonies stay flat, but grow. B-plate is suspect. Determination shows maybe Chrysosporum or Trychophyton	Day 13: Selective agar - yellow white mini colonies, >20. 1x green, brown with white 6cm. Sabouraud: 4x white, 3-4 cm. 1x green powdery, yellow white flat colonies >20, 3-5mm. B-plate: 4x white colonies 1cm. Powdery aspect with snowflake-like boundries.	Day 16: Sel. Agar - whiteblack flag 7cm and multiple minicolonies, Sab. - 1x black 4cm, 1x white 3cm, 5x white 1cm, multiple mini colonies, Sab. B plate - idem Day 13	Day 19: Sel. Agar - idem day 16, Sab. 1x black 5 cm, 1x white 4 cm, Sab. B plate - 6x white, powdery. Determination: no M. Canis or Trychopyton ?	Day 22: Sabouraud B plate: idem day 19 plus 1x green	

									Determination: No M. canis or trychophyton probably chrysosporium				
4	Negative	No pathogenic fungal growth	12-19-2012	12-19-2012	Day 4: both negative	Day 5: both negative	Day 8: both negative	Day 11: Selective agar 1x white, 4mm	Day 14: Sab. - 1x white 1-2mm, Sel. Agar - 3x white 1-7mm	Day 17: Sel. Agar - 1x white 1cm, 1x white yellow 3mm multiple bulbs, 1x yellow 1-2mm, 1x white around hair bulging, Sab.- 2x whiteyellow bulging 2mm	Day 20: Sab. Idem day 17, Sel. Agar - 1x white growth aroud a hair has become a white island	Day 22: idem	
5	Negative	Sabouraud and Selective agar: day 17 Aspergillus, Sabouraud B plate day 20: Aspergillus	12-19-2012	12-19-2012	Day 4: both negative	Day 5: Sabouraud: 4x mini colony, white. 1x colonoy 3-4 mm, white green	Day 8: Selective agar: big yellowgreen colony and few small colonies. Sabouraud: big green colonies. Not suspect.	Day 11: Sabouraud: green colonies, 2x white bulging. Selective agar: 1 suspect colony, snowflake growth --> new Sabouraud B-plate.	Day 14: Sab. - >20 darkgreen 1 - 40mm, Sel. Agar - idem, Sabouraud Bplate: 5x white snowflake, Determination: No M. Canis or Trychophyton, probably chrysosporium	Day 17: Sab. - >20 darkgreen to black donutshaped 1 mm - 3 cm, 2x white, Sel. 7x white and green overgrowth, Sab. B plate: 5x white yellow snowflakelike colonies - Determination: No M. canis or trychophyton probably chrysosporium, 2x white bulging	Day 20: Sabouraud B plate: 5x whitbrown starshaped, 2x green 2-3cm		
6	Negative	Sabouraud: day 11 - overgrown Aspergillus, Selective agar: day 20 no pathogenic fungal growth	12-21-2012	12-22-2012	Day 2: both negative	Day 5: Sabouraud: 2x big green, 3cm.	Day 8: Sabouraud: 2x big green, 6cm	Day 11: Sel. Agar - 1x green mini colony, Sab.- overgrowth green	Day 14: Sel. Agar - 1x darkgreen 2mm	Day 17: Sel. Agar - 1x darkgreen 3mm	Day 20: Sel. Agar - 11x darkgreen white border		
7	Negative	Sabouraud: day 8 - overgrown Aspergillus, Selective agar: day 11 overgrown Aspergillus	12-21-2012	12-22-2012	Day 2: both negative	Day 5: Selective agar: 2x 1.5 cm white/green. Sabouraud: 2x green 2cm, 1x mini white	Day 8: Sabouraud: carbahe, overgrowth. Selective agar: 2x green, 3cm.	Day 11: Sel. Agar - green overgrowth					
8	Negative	Sabouraud: day 14 -	12-21-2012	12-22-2012	Day 2: both negative	Day 5: Selective agar: 1x mini	Day 8: Selective agar: 3x white, 1-	Day 11: Sel. agar - 3x white 1mm, 2x	Day 14: Sab. - 1x green 10cm, 10x	Day 17: Sel. Agar - whitegreen			

		overgrown Aspergillus, Selective agar: day 17 - Aspergillus				white	2mm, 1 greenwhite 4mm. Sabouraud: 1x greenwhite, 3cm. 2x white 1-3mm, 1 white/yellow 2-4 mm	green bulging 5-10mm, 2x yellow red 1-2mm, Sab. - green islet 6 cm, 1x white bulging 1cm, 1x yellow 1cm	white bulging, 1x red, Sel. Agar - 7x whitegreen 1-15mm	bulging 2mm - 2cm		
9	Negative	Sabouraud: day 17 - suspect Aspergillus, Selective agar: day 17 no pathogenic fungal growth	12-23-2012	12-24-2012	Day 3: Selective agar: 1x white colony	Day 6: Sabouraud: 2x white small colonies, 2-3mm, Selective agar: 12 small white colonies, 1-4mm, 1x green 5mm, 1 orangegreen, 1cm	Day 8: Sab. > 10 small white 1-10mm, 1x whitebrown 1cm, Sel. Agar - 1x darkgreen 1cm, 1x orange 1.5-2cm, multiple white (>10) 1-9mm	Day 11: Sel. Agar - 30 white and yellow bulging 2-10mm, 2x darkgreenblue (vulcano) 3-15mm, 1x orange red green blue 2cm starpattern, Sab.- 17x whiteyellow bulging 1-15mm, 1x white suspect colony -> New Sabouraud B-plate	Day 14: Sabouraud and Selective agar idem Day 11 (Sab: variation white, whiteyellow, whitebrown all bugling)	Day 17: Sab - >10 white bulging, 1x green powdery (thrown away), Sel. Agar - idem day 14 (thrown away), Sab. B plate - negative	Day 21: B plate - 9x small white colonies, yellow center. 1x greenish fluffy colony, suspect Aspergillus	Day 24: B plate - overgrowth with Aspergillus. Thrown away.
10	Negative	Sabouraud: day 16 - overgrown orange colonies, Day 22 selective agar - no pathogenic fungus	12-29-2012	12-30-2012	Day 4: both negative	Day 7: Sab.- 4x whitepink bulging 4mm, Sel. Agar - negative	Day 10: Sel. Agar - negative, Sab. - 4x whitepink bulging 1cm	Day 13: Sab. - 4x salmonpink 2 cm, white in the middle, Sel. Agar - 1x white bulging 2mm	Day 16: Sel A - 1x 3 mm white, bulging. Sab: 4x orange, 2cm with a yellow or orange center. Grows like a vulcano. (thrown away)	Day 19: Sel. Agar - 1x white bulging colony, 3 mm, the same as on day 16.	Day 22: Sel. Agar - >10 whitedarkgreen	
11	Negative	Selective agar: day 7 - overgrown Aspergillus, Sabouraud: day 10 - no pathogenic fungus overgrowth, Day 12: Sab. B plate - overgrown Aspergillus	12-29-2012	12-30-2012	Day 4: Sel.agar - 2x white 2mm, Sab.- negative	Day 7: Sel. Agar - 2x yellowpink bulging in the middle 7mm, 1x green 2mm, 1x white powderish suspect -> New Sabouraud plate	Day 10: Sab. - darkgreen island, Sab. B plate - 5x white 2 of which are bulging 1cm	Day 12: Sab. B - 2x greenwhite Aspergillus 2-3 cm				
12	Negative	Selective agar: day 10 - suspect Aspergillus. Sabouraud:	12-29-2012	12-30-2012	Day 4: Sel. agar - 1x green, 1x white, Sab. - negative	Day 7: Sab. - 2x whitepink 1-2mm, 1x white bulging 2mm, Sel. agar - 1x	Day 10: Sel. Agar - 1x black 4cm, 1x whitegreen 4cm, 1x yellow 1cm, Sab. - 5x	Day 13: Sab. 1x whiteyellow bulging 1cm, 3x white bulging 0.5cm, 2x	Day 16: Sab: idem, thrown away.			

		day 16 - overgrown Aspergillus				darkgreen 2cm, 1x whitegreen 1.5cm, 1x white-yellow-pink 8mm	whiteyellow bulging 1-8mm, 4x whitegrey bulging 1-4mm	darkgreen 1-4mm, 1x white bulging 1cm, 1x whitegreen bulging 1cm				
13	Negative	Selective agar: day 10 - overgrown Aspergillus. Sabouraud: day 16 - overgrown with orange colonies	12-29-2012	12-30-2012	Day 4: both negative	Day 7: Sab.- 1x white bulging 4mm, 2x yellow-pink-white 6mm, Sel. agar - 3x white bulging 2-5mm, 1x darkgreen bulging 2mm, 1x yellow 5mm	Day 10: Sel. Agar: 1x yellow 1cm, 1x greenwhite 2cm, 1x black 5mm, 2x white 5mm --> New Sabouraud B plate, Sab. 2x orangeyellow 1cm, 1x white vulcano 1cm	Day 13: Sab. - 2x salmonpink 2cm, white in the middle, 1x white 1cm vulcano, 1x white 2mm, Sab. B plate - 7x white bulging pluche (thrown away)	Day 16: Sab - the same as day 14, only bigger colonies (thrown away)			
14	Negative	Sabouraud: day 19 - overgrown other fungi. Selective agar day 21: no pathogenic fungi	12-29-2012	12-30-2012	Day 4: both negative	Day 7: Sab. - 3x yellowpink 1-4mm, Sel. agar - 1x yellow 5mm	Day 10: Sel. Agar - 1x yellowred 1cm, Sab. - 3x whiteyellow bulging 8mm	Day 13: Sab. 2x salmonpink 2cm, 1x greygreen 1cm, Sel. Agar - 1x yelloworange 2cm, 1x white 2mm	Day 16: Sel A - 1x yellow orange colony, 2.5 cm grows like a vulcano. 2x grey colonies, 3mm. Sab - 2x salmon pink, 3 cm. 1x grey, 2cm vulcano.	Day 19: Sel. Agar - the same as day 16, plus 2x white bulging colony 1 mm. Sab. - the same as day 16 but bigger and plus 3x white bulging 1 mm. (thrown away)	Day 21: Sel. Agar - 2x white bulging 4mm, 1x whiteyellow 5cm
15	Negative	Selective agar: day 7 - overgrown Aspergillus, Sabouraud: day 10 - Aspergillus determination	12-30-2012	12-30-2012	Day 4: Sab. - 4x white 1-6mm, Sel. agar - negative	Day 7: Sab. - 2x white bulging 3mm, 1x white reliëf/hilly, Sel. agar: 2x white 1-2mm, 3x white bulging 2-3cm, 1x green islet	Day 10: Sab. - 1x white 5cm, Determination: Aspergillus, 2x whiteyellowgreen 1cm				
16		Selective agar and Sabouraud: Day 7 suspect Aspergillus	12-30-2012	12-30-2012	Day 4: Sab. - 3x white bulging 1mm-1cm, Sel. agar - 2x white 1mm	Day 7: Sab.: green white (bulging), 2x suspected white -> New Sabouraud plate, Sel. agar - growth around all hairs, 2x black 2cm, 2x white bulging, over 20 white mini colonies	Day 10: Sab. B plate: colonies on graft spots, bulging, white	Day 12: Sab. B plate - 1x greenwhite 3-4 cm bulging, 1x greenwhite 1cm, 2x white flat 5-10mm --> New Sabouraud B2 plate	Day 13: Sab. B2 plate: negative	Day 19: B2 plate - 5x white poudery, 1 cm. Determination: lots of round microsporidiae around hyphae. Trychophyton?	Day 24: Sab. B plate suspect trychopyton, Microscopic determination is not consistent with macroscopic findings, UREASE test performed (takes 7 days)
17	Negative	Sabouraud: day 7 -	12-30-2012	12-30-2012	Day 4: Sab.- 1x white	Day 7: Selective agar: negative	Day 10: Sel. Agar - 2x white bulging				

		overgrown Aspergillus. Sel A: day 10 - overgrown Aspergillus			colony 1cm, Selective agar - negative		fluffy 3mm							
18	Negative	Sabouraud day 6 - overgrown Aspergillus, Selective agar: no (known) pathogen fungal growth	5-1-2013	5-1-2013	Day 4: Sel. Agar - negative, Sab. 1x white bulging 1 cm	Day 6: Sab. - 1x greenwhite Aspergillus 3cm, Sel. Agar - 1x whiteyellow 1-2mm	Day 7: same as day 6	Day 10: Sel A - same as day 6	Day 13 - same as day 6, 5mm now	Day 16: Sel. Agar - 2x white 1-8mm	Day 18: Sel. Agar - idem day 16	Day 20: Sel. Agar - idem day 18	Day 23: idem (end)
19	Negative	Both Day 10 - overgrown Aspergillus	7-1-2013	8-1-2013	Day 3: both negative	Day 4: Sab. - 1x spiderweb white 3mm, 1x white snowflake 3mm, Sel. Agar - negative	Day 7: Sab - 1x spiderweb 2cm, bulging. 1x grey/green bulging 2cm, 1x whitegreen 2mm. Sel A - 1x green 2cm	Day 10: both overgrown Aspergillus					
20	Negative	Both day 13 - overgrown all sorts of fungus no pathogens	7-1-2013	8-1-2013	Day 3: both negative	Day 4: Sel. Agar - 1x white 2mm, Sab. - 1x white 2mm	Day 7: Sab - 1x white vulcano 1 cm, 3x whiteyellow 3mm bulging, 1x whitegreen 2mm, 1x greenyellow 4mm. Sel A - 5x yellowwhite 2-5mm, 1x white vulcano 2cm, 1x white 2mm	Day 10: Sab - 3x yellowwhite 1 cm bulging, 1x grey 2mm, 1x white poudery 4 cm, 1x greenred 1.5 cm and 1x white bulging 0.5 cm. Sel A - 4x yellowwhite 1 cm, 1x yellow island 4cm, 1x yellowwhite 3mm	Day 13: idem day 10				
21	Negative	No pathogenic fungal growth	7-1-2013	8-1-2013	Day 3: Sab. - negative, Sel. Agar - 2x whiteyellow 1mm	Day 4: Sab. - negative, Sel. Agar - 4x whiteyellow 1mm	Day 7: Sab - negative. Sel A - 9x white yellow 1-4 mm	Day 10: Sel A - 6x yellow flat 4 mm, 4x white bulging 5mm. Sab - 1x white bulging 3mm	Day 13: Sab. - 1x white bulging 5mm, Sel. Agar - 6x yellow 5-10mm, 1x orangewhite 1cm, 3 white 5-10mm, 1x whitegreen 1cm	Day 15: Sab. - 1x white bulging 1cm, 1x pinkred bulging 1mm, Sel. Agar - 4x yellow 4mm, 2x white 6mm, 1x pink vulcano 1cm, 1x green 2cm, 1x brown 2cm	Day 17: Sel. Agar - idem + 4x white 2mm, Sab. - idem day 15	Day 20: Sab. Idem day 17, Sel. Agar - multiple colours and shapes	Day 22: Sab,-2x white bulging 3-20mm, 1x mini white --> new Sab. B plate, Sel. Agar - diverse (end)
22	Negative	Selective agar day 15: overgrown darkgreen	10-1-2013	10-1-2013	Day 2: Sel. Agar - negative, Sab. - 1x yellow	Day 5: Sel A - negative. Sab: 1x white 2mm	Day 8 : Sab - 1x white flat 1 cm. Determination: lots of loose	Day 11: Sab. - 1x white flat 2cm, Sel. Agar - >10 darkgreenwhite	Day 13: Sel. Agar - idem day 11, Sab. - 1x white flat 2cm, 5x white	Day 15: Sab. - 1x white 4cm, 1x white 1cm, Sel. Agar - >50%	Day 18: Sab. - 2x white 3-5cm, Sab. B plate: 5x white little bit bulging 2-	Day 20: Sab. - 1x white > 50%, 2x white 5mm-4cm, 1x penicillium-like	

		white fungus, Sabouraud day 20: overgrown white fungus not pathogenic			1mm		microsporidiae. Chrysosporum? Sel A - negative	cluster	5mm flat - Microscopic determination: no (known) pathogen	darkgreenwhite	10mm	6mm, 1x green 1cm, Sab. B plate - 4x white little bit bulging, Determination: suspect chrysosporium	
23	Negative	Sabouraud day 11: white propellor like overgrowth, Selective agar: no pathogenic fungal growth	10-1-2013	10-1-2013	Day 2: both negative	Day 5: Sel A - negative. Sab: 1x white 1cm	Day 8: Sab - 1x white 3 cm poudery, 1x greenwhite 3mm. Sel A - 2x white bulging 1-3mm	Day 11: Sab. 1x white 6cm, 1x greenwhite 5mm, Sel. Agar - 2x whitebulging 6-8mm, 1x whiteyellow 3mm	Day 13: Sel. Agar - 2x white bulging 1cm, 1x white 2mm	Day 15: Sel. Agar - idem	Day 18: Sel. Agar - 1x white bulging 1,5cm, 1x whiteyellow 3mm, 1x whiteorangegreen bulging 1cm	Day 20: Sel. Agar - 1x white bulging 2cm, 1x whitegreen 1,5cm, 1x whiteyellow 5mm	Day 23: idem day 20
24	Negative	Sel. Agar day 15: overgrown white with yellow powder fungus, Sabouraud: no pathogenic fungal growth	10-1-2013	10-1-2013	Day 2: both negative	Day 5: Sel A - 1 white 1 mm, Sab - 1x white 1 mm	Day 8: Sab - 1x greenwhite 1 cm fluffy, 1x whiteyellow 2mm. Sel A - 1x white 2cm circulair	Day 11: Sab. - 2x white fluffy bulging 4mm, Sel. Agar - 1x whiteyellow 4cm, 1x white 2mm	Day 13: Sel. Agar - 1x white 4cm, 1x white green center 4mm, Sab. - 2x white bulging 1-2cm	Day 15: Sab. - 1x white bulging 3cm, 2x greenyellowhite 2-15mm, Sel. Agar - idem day 13, now bigger and white with yellow powder	Day 18: Sab. - 1x white bulging 4cm, 1x mossgreen powdery donutshaped 1,5cm	Day 20: Sab. - 1x white bulging 4cm, 1x greengranular donut 1,5cm	Day 23: idem now bigger
25	Negative	No pathogenic fungal growth	10-1-2013	10-1-2013	Day 2: both negative	Day 5: both negative	Day 8: Sel A - 1x 3 mm white, around the hairs a white demarcation zone. Sab - negative	Day 11: Sel. Agar - 1x white around the loose hairs, 2x greenwhite 5cm, Sab. - negative	Day 13: Sab. - negative, Sel. Agar - 1x white around the hairs 3cm, 2x green 5mm	Day 15: Sab. - negative, Sel. Agar - 1x greenwhitebrown 5cm	Day 18: idem day 15	Day 20: Sab. - negative, Sel. Agar - idem now 70% of the plate (end)	Day 23: idem
26	Negative	Sabouraud day 13: overgrown white non pathogenic fungus also greenwhite colonies, Selective agar: no pathogenic fungal growth	10-1-2013	10-1-2013	Day 2: both negative	Day 5: Sel A - negative. Sab - 1x white 1 cm, 1x white 2mm bulging	Day 8: Sab - 1x white poudery 3cm vulcano, 2x greenwhite 3mm, 1x brown 3mm, 1x green 2 mm (Ruth!) Sel A - 1x yellow 2 mm	Day 11: Sab. - 1x white 6cm, 3x greenwhite 8-15mm, Sel. Agar - 1x yellow 5mm, 1x greenorange 4mm	Day 13: Sel. Agar - idem day 11, Sab. - 1x white 50% of the plate, 3x greenwhite 6mm, 1x green 5mm	Day 15: Sel. Agar - 1x redbrown 5mm, 1x yellow 5mm	Day 18: idem day 15	Day 20: idem day 18 + 1x whitegreen 3mm	Day 23: idem day 20
27	Negative	No pathogenic fungal growth	10-1-2013	10-1-2013	Day 2: both negative	Day 5: both negative	Day 8: both negative	Day 11: Sel. Agar - negative, Sab. 1x white bulging 5mm	Day 13: Sab. 1x white bulging 8mm, Sel. Agar - 1x orangewhite 1mm	Day 15: Sel. Agar - 1x redwhite 3mm, Sab. - 1x whitegreen 2cm, 1x white 3mm	Day 18: Sab. - 1x greenwhite 1,5cm, 1x white bulging 7mm, 1x yellow 1mm, Sel. Agar - 1x redwhite 3mm	Day 20: idem day 18	Day 23: idem day 20
28	Negative	No pathogenic fungal growth	10-1-2013	10-1-2013	Day 2: both negative	Day 5: both negative	Day 8: Sab - 1x brown 1mm, 1x	Day 11: Sel. Agar - 9x miniwhite, Sab.	Day 13: Sab. - 1x brown 6mm, 1x	Day 15: Sab. - idem day 13 + 1x	Day 18: Sab. - 1x darkgreen 3cm, 1x	Day 20: idem day 18	Day 23: idem day 20

							white 1 mm	- 1x brown 4mm, 1x whiteyellow 4mm, 1x white 2-3mm	grey fluffy 5mm, 2x white 1mm, 1x whityellow 3mm, 1x greenwhite 2mm, Sel. Agar - 11x whiteyellow 1-3mm	white flat 8mm, Sel. Agar - idem day 13 now 2-6mm	greengrey 1cm, 1x black 1,5cm, 10 white bulging 1-10mm, Sel. Agar - 11x whiteyellow bulging 3-7mm	
29	Negative	Sabouraud day 7: overgrown white powdery bulging, Selective agar day 10: overgrown white fluffy fungus	11-1-2013	11-1-2013	Day 4: Sab - 17x white 2mm-8mm, poudery. Sel A - 14x the same white poudry colonies. Suspect? Determination next time!	Day 7: Sel A - 8x powdery white bulging Sab - overgrown with white bulging colonies.	Day 10: Sel. Agar - overgrown white fluffy fungus						
30	Negative	Sabouraud day 10: greenwhite overgrowth, Day 22: Selective agar - no pathogenic fungal growth	11-1-2013	11-1-2013	Day 4: both negative	Day 7: Sel A - 9x white 1mm, 1x yellow 1 mm. SAB - 1x white 1.5 cm, 2x whitebrown 3mm (Ruth!) Determination: 'kralenketting' 2x greenyellow 2mm	Day 10: Sab. - 1x greenwhite 7cm, Sel. Agar - 1x white 2-10mm, 1x yellow 4mm, 1x darkgreen 1cm	Day 12: Sel. Agar - 11x white 1-10mm, 1x yellow 4mm, 1x brown 2cm, 1x black 1mm	Day 14: Sel. Agar - 12x white 4-15mm, 1x yellow 9mm, 3x black 2mm, 1x salmonpink 1mm, 1x blackgreen 4cm	Day 17: Sel. Agar - 1x black 5cm, 12x white 1cm, 1x yellow 1cm	Day 19: Sel. Agar - 1x black 5cm, 12 white 1cm, 1x yellow 1cm	Day 22: idem day 19 + 1x green, 1x orange, 1x black
31	Negative	No pathogenic fungal growth	11-1-2013	11-1-2013	Day 4 : both negative	Day 7: Sel A - 1 mm white	Day 10: Sab. - negative, Sel. Agar - 1x greenwhite 1cm	Day 12: Sab. - negative, Sel. Agar - 1x greenwhite 2-3 cm	Day 14: Sab. - negative, Sel. Agar - 1x greenwhite 3-4cm, 2x whitegreen 3mm	Day 17: Sel. Agar 1x darkgreen white powdery 6cm, >10 cluster darkgreenwhite 2-7mm, Sab. - 1x darkgreenwhite 1cm	Day 19: Sel. Agar - darkgreenwhite overgrowth, 1 mini supsect, new Sab. B plate, Sab. - 1x darkgreenwhite 2cm	Day 22: Sab. B plate 1x whitegreen bulging 7mm, Sab. Idem
32	Negative	Sabouraud day 7: yellowwhite granular overgrowth, Day 18: Selective agar - overgrown yellowwhite with black and brown powder	13-1-2013	14-1-2013	Day 4: Sab - 4x white bulging 4mm Sel A - yellowwhite around hairs	Day 7: Sel. Agar - yellowwhite around the hairs 80% of the plate, 1x greenwhite 1cm, Sab. - 3 white 3cm, 6x white 2mm	Day 9: Sab. 90% of the plate yellow white granular, Sel. Idem day 7	Day 11: Sel. Agar - idem day 9, greenwhite 2cm macroscopicly supsect Aspergillus	Day 14: Sel. Agar - idem, greenwhite is now brownwhite powdery 2cm	Day 16: Sel. Agar - white sludge and idem day 14	Day 18: Sel. Agar - overgrown whiteyellow whith black and brown powder	

33	Negative	Sabouraud and Selective agar: Day 7 green growth	14-1-2013	14-1-2013	Day 4: Sab - 3x white 2mm	Day 7: Sel.Agar - 1x green 3cm, 3x white 1-10mm 2 bulging 1 flat --> New Sab. B plate, Sab. - 1x greenwhite, 1 yellowwhite, 1x orangewhite, 4x whitegreen 3-5mm all bulging	Day 9: Sab. B plate: negative	Day 11: Sab. B plate - 1x white bulging 3mm	Day 14: Sab. B plate - 5x white bulging 1-15mm	Day 16: Sab. B plate - 4x white 4-10mm, 1x grey 1mm, Determiniation: a lot of hyphae, few micro's, no macro's	Day 19: Sab. B plate: 4x white 1-2cm, 1x black 3mm, Not supsect		
34	Negative	Day 11: Sabouraud and Selective agar overgrown darkgreen black fungus	15-1-2013	17-1-2013	Day 4: both negative	Day 6: Sab. - 2x darkgreen flat 1cm, Sel. Agar - 2x darkgreen 4-10mm, 1x white flat fluffy 3mm	Day 8: Sab. - 1x darkgreenblack 5cm, 1x white 8mm, 1x darkgrey 2mm, Sel. Agar - 1x darkgreenblack 4cm, 1x white powdery 8mm	Day 11: Sab. And Sel. Agar - overgrown darkgreen black > 50% of the plate						
35	Negative	Day 8: Sabouraud overgrown green brown black grey fungus, Day 10: Selective agar - overgrown blackgreen fungus and yeasts	19-1-2013	20-1-2013	Day 3: Sab - negative, Sel. A. - white sludge around the loose hairs	Day 5: Sab. - 1x darkgreenwhite 3cm around the loose hairs, >10 small white around the hairs like yoghurt, Sel. Agar - 1x darkgreen 1cm, >10 white smalle like yoghurt around the loose hairs	Day 8: Sab. - overgrown green brown black grey fungus, Sel. Agar - 3x darkgreen 2cm, 1x greenwhite 1cm, >10 white 2-4mm	Day 10: Sel. Agar - blackgreen and 'yeasts', end						
36	Negative	No pathogenic fungal growth	19-1-2013	20-1-2013	Day 3: both negative	Day 5: both negative	Day 8: Sel. Agar - 4x white 1-3mm, suspect too small now, 1x greenwhite 5mm, Sab. - 1x greenwhite 5mm, 1x white 1mm	Day 10: Sab. - 1x greenwhite 1cm, 1x yellowwhite3mm, Sel. Agar - 5x greenwhite 3-10mm, 2x white 1-4mm, 1x black 1mm	Day 13: Sel. Agar - 4x greenwhite 1,5-3cm, 2x white bulging 2-5mm, 1x whiteyellow 1mm, Sab.- 6x whitegreen 1-2mm, 1x green 1cm	Day 16: Sel.- 3x darkgreenwhite granular 2-3cm, 1x yellow 1cm, 2x white bulging 2-10mm, Sab. - 6x darkgreen(white) 3mm-1,5cm	Day 18: Sab.- 4x darkgreen 1-2cm, Sel.- idem day 16 + 6x whiteyellow 1-8mm	Day 20: Sab. 3x d.green 1-3cm, 1x whitegreen 2mm, Sel. - 3x dgreen 3-4cm, 1x yellow 1,5cm, 2x whiteyellow 2-10mm, 5x whitegreen 2mm	Day 23:idem day 20
37	Negative	Day 8: Sabouraud : overgrown brown white green fungus, Day 10:	19-1-2013	20-1-2013	Day 3: both negative	Day 5: Sab.- 1x greenwhite 1cm around the loose hairs, 1x white powdery 2mm, sel. Agar - 1x	Day 8: Sab. - overgrown brownwhitegreen fungus, Sel. Agar - overgrown, 3 minicolonies	Day 10: Sel. Agar - idem day 8, end. Sab. B plate - 5x white star	Day 13: Sab. B plate - 6x white little bit bulging, not supsect	Day 16: idem Day 13	Day 18: idem day 16	Day 20: idem day 18		

		Selective agar: overgrown, Sab. B: no pathogenic fungal growth				greenwhite 5mm around the loose hairs, 2x white powdery 1-3mm	suscpect --> New Sab. B plate						
38	Negative	No pathogenic fungal growth	19-1-2013	20-1-2013	Day 3: both negative	Day 5: Sab. - negative, Sel. Agar - 5x white bulging 1-3mm	Day 8: Sab. - 3x white yellow 1-3mm, 1x greenwhite 3mm, Sel. Agar - 6x white 3-10mm, 1x darkgreen 2mm	Day 10: Sel. Agar - 1x white vulcano 1cm, 2x white bulging 4-6mm, 1x whitegreen3mm, 3x yellowwhite 'yeast', 1x whiteywllow 4mm, Sab.- 1x green 3mm, 2x greygreen 2-4mm, 6x yellowwhite 1-2mm	Day 13: Sel. Agar - 1x whitegreen 1,5cm, 2x white 1-1,5cm, 6x whiteyellow 2-6mm, 1x yellowgreen 3mm, Sab. - 1x green spiral 8mm, 2x whitegreen 4mm, 2x yellow 2mm, 4x white 1mm	Day 16: Sel. - 2x darkgreenwhite 5mm-2,5cm, 3x white bulging 2mm-2cm, 4x whiteyellow 2mm-1cm, 1x blackbrown 6mm, Sab. - 7x piles yellow, brown, green and white, 4x white 1mm	Day 18: idem day 16	Day 20: Idem day 18 bigger	
39	Negative	No pathogenic fungal growth	20-1-2013	22-1-2013	Day 3: Sab. - 2x white 1mm, Sel. Agar - 2x white 1mm	Day 6: Sab. - 9x white 3-20mm, Sel. Agar - 17x white 3-6mm	Day 8: Sel. Agar - 1x white 4mm, >20 white' ýeast', Sab. - 8x whiteyellow 'yeast', 1x white bulging 5mm	Day 11: Sel. Agar - +/- 50% white yellow 'yeast', 2x white bulging 5mm, Sab. - 9x whiteyellow 'yeast' 3mm-2cm, 1x white bulging 1cm	Day 14: Sel. And Sab- idem day 11	Day 16: Sel. And Sab. Idem day 14	Day 18: Sel.- yeast 50%, 2x white bulging 7-10mm, Sab. - idem day 16	Day 21: Sab. - 8x yeast 3mm-4cm, 1x white 2cm, Sel.- 50% yeast, 2x whitebulging 5-15mm	
40	Negative	Day 16 Sabouraud overgrown all sorts of fungus	25-1-2013	27-1-2013	Day 3: Sel. Agar - negative, Sab. - 1x yellow mini	Day 7: Sab. - 6x white mini, Sel. Agar - white yellow around all the hairs, 1x white bulging 7mm	Day 9: Sab. - 15 darkgreenwhite 1-7mm, >20 whiteyellow mini, Sel. - 1x white island 5cm, 5x whiteyellow 1-2mm	Day 11: Sab. And Sel. Idem day 9	Day 13: Sel. - 1x white 70% fluffy, 10x whiteyellow 1-2mm, yeast, Sab. Idem	Day 16: Sab. - 12x greenwhite 1-2cm, 30 yellow 2-8mm, 6x white diverse 3-6mm (END), Sel. -idem day 13	Day 18: Sel - idem	Day 20: Sel - idem	Day 23: idem, END
41	Negative	No pathogenic fungal growth	25-1-2013	27-1-2013	Day 3: negative	Day 7: Sab. - negative, Sel. Agar - 2x white mini	Day 9: Sel.- 8x white 1-2mm, Sab. - 1x white bulging 2mm	Day 11: Sab. And Sel. Idem day 9	Day 13: Sel.- 9x whiteyellow 1-3mm, Sab. - 6x white mini-4mm	Day 16: Sab. >10 white bulging 1-6mm, Sel. - 8x white 2-4mm, little web, 4x yellow 2-4 mm	Day 18: Sab - > 20 white, bulging, 1-6 mm. Sel - idem day 16	Day 20: Sab - idem but bigger, 1-10mm. Sel - idem	
42	Negative	No pathogenic fungal growth	25-1-2013	27-1-2013	Day 3: negative	Day 7: Sel. Agar - 3x white 1-3mm, Sab. - 4x white mini	Day 9: Sel. - 2x whitegreen 1cm, 1x white 3mm, Sab.- 5x white bulging 8mm	Day 11: Sel. - 2x whitegrey 1cm, 2x white 1-2mm, Sab. - 6x white(grey) 1cm, 3x white 1mm	Day 13: Sab. - 9x whiteyellowgrey 1-10mm, Sel.- idem bigger	Day 16: Sel. - 2x whitegrey 3-4cm, 3x white 1-6mm, Sab. - 6x whitegrey 2cm, 2x whiteyellow 2-4mm	Day 18: Sab - idem, Sel - idem	Day 20: Sel - 2x white black and white colonies, 3-4cm. 3x white 1-7mm. Sab - 1x grey 8 cm, yellow and white colonies inside, 1x white	

												2mm.
43	Negative	Day 9: Sabouraud overgrown green white fungus, Day 13: Selective Agar overgrown blackgrey fungus	27-1-2013	27-1-2013	Day 3: negative	Day 7: Sel. Agar - 4x green 1cm, 13x white 1-5mm, Sab.- 2x green 2-3cm, 2x white 2mm	Day 9: Sel. - 1x green 7 cm, 7x white fluffy 2-10mm, Sab.- > 95% green white fluffy (END)	Day 11: Sel. - >90% blackgrey with 5x white 1cm	Day 13: Sel. Idem (END)			
44	Negative	No pathogenic fungal growth	27-1-2013	27-1-2013	Day 3: negative	Day 7: negative	Day 9: Sab. - 4x white 1-3mm, Sel. 1x white 2mm	Day 11: Sab. - 3x white 1-4mm, Sel. - idem day 9	Day 13: Sel. - 1x white 3mm, 1x black 1mm, Sab. - 5x white 1-10mm, 4x black 1-3mm	Day 16: Sel - 2x white 3mm-6mm, 1x black 2mm, 1x whitegreen 8 mm. Sab - 6x whiteyellow 2-12mm, 7x black 1-2mm.	Day 18: Sab - idem, Sel - idem	Day 20: Sab - 5x whiteyellow 4-12 mm, 1x brown 9 mm, 8x black 1-2mm. Sel - 1x darkgreen, 13mm, 2x white island 5cm, 3x black 1-3mm
45	Negative	No pathogenic fungal growth	27-1-2013	27-1-2013	Day 3: negative	Day 7: Sab. - 7x white bulging 1-3mm, Sel. Agar - white sludge around the hairs, 1x yellow shiny 2mm	Day 9: Sel. - yeasts around the hairs, on top of the hais 3x white bulging 3mm, Sab. - 8x green white 5-10mm, 1x white bulging 2mm	Day 11: Sab. - idem day 9, Sel. Idem day 9, on top of the hairs now 8x greenwhiteyellow 2-4mm	Day 13: Sab. And Sel. - idem day 11	Day 16: Sel. - 6x greenwhite 3-10mm, 1x white 4mm, yeast around the hairs, Sab. - 1x greenwhite 10cm, >10 white 1-10mm	Day 18: Sab - white became whitegreen, further idem. Sel - idem	Day 20: Sel - idem. Sab - 8-% of the plate is greenwhite
46	Negative	No pathogenic fungal growth	27-1-2013	27-1-2013	Day 3: negative	Day 7: Sab. - 17x white some of them bulging 1-2mm, 3x whitegreen 2mm, Sel. Agar - >10 white around the hairs 1-2mm	Day 9: Sel. - >10 green 2-5mm, 11x white bulging 2-4mm, 1x yellowgreen 2mm, Sab.- 12xgreen(white) 3mm-2cm, 5x white bulging 2-10mm, >10 white mini suspect --> New Sab. B plate	Day 11: Sab B.- 4x white mini, 1x greenwhite bulging 2mm, Sab. - >15 green 5-10mm, 3x beige 5-10mm, 3x whiteyellow 2-5mm, 1x white bulging 4mm, Sel. - >10 greenwhite 5mm, 1x white 5mm	Day 13: Sab.- idem + 13x miniwhite, Sel. 3x white 5mm, 2x orangbrown 3mm, green idem, Sab.B- 5x whitegreen 4mm	Day 16: Sel. >10 green 5cm, 1x brown 3mm, 3x white vulcano 4mm-1cm, Sab.- idem bigger, Sab. B. - 5x greenwhite 1cm, 2x white 1mm	Day 18: Sel - idem. Sab - idem. Mostly green. B plate - 6x green, 3x white fluffy	Day 20 - Sel - idem. Sab - lots of green colonies, 3x brown, 2x white fluffy. B plate - 9x green white border
47	Negative	Day 16: Sabouraud overgrown white granular and green fungus. Sel - no pathogenic fungal growth	27-1-2013	27-1-2013	Day 3: negative	Day 7: Sel. Agar - white yellow sludge around the hairs, Sab. - 7x whitegreen 2-3mm	Day 9: Sab. - 1x white 3cm, 7x green 2-8mm, 1x white bulging 2mm, Sel. - yeast yellow around the hairs, 8x greenwhite 2-	Day 11: Sab. And Sel. Agar - idem day 9 but bigger	Day 13: Sel.- yeast, >10 greenwhite 2-5mm, 1x white brown 1.5cm, 4x white 1-4mm, Sab. - 1x white 60%, 7x white	Day 16: Sel. Yeast >40%, 1x green 4cm, 1x brownwhite 2,5cm, 3x whitebulging 4mm, 1x white 2mm --> New Sab.	Day 18: Sel - idem. B plate - negative	Day 20 - Sel - brownwhite 3cm, lots of yeasts, 4x whiteyellow 8 mm and green cluster of 3cm. B plate - 5x white 8 mm, fluffy.

							4mm, 2x yellowwhite 2-5mm		bulging 1-5mm, 1x green 4cm	B plate, Sab. - 80% granular whiteyellow, >10 green 1mm-2cm (END)				
48	Negative	No pathogenic fungal growth	30-1-2013	1-2-2013	Day 4: negative	Day 6: Sel. - little web around the hairs, Sab. - 2x white 2mm	Day 8: Sel. - idem day 6 + 1x brownwhite 1mm, Sab. - 2x greenwhite 4mm, 3x brownwhite 2mm	Day 11: Sel.- yeastweb _ 1x brown 3mm, Sab.- 4x d.green 2-10mm, 1x greenwhite 1cm	Day 13: Sel - >70% yeasts around the hairs, 1x darkbrown 4mm. Sab - 4x green 3mm-15mm, 1x whitegreen vulcano 1.5cm	Say 15: Sab - 4x geen 5-15mm, 19x whitegreen 1-2mm. Sel - idem	Day 18: Sab - 2x darkgreen, 1x whitegreen 2cm, >15 whiteyellow 1-2mm. Sel - idem	Day 20: Sel - yeast, 1x black 1cm, Sab - >20 d.green, 2x whitegreen 5mm-3cm	Day 22: both idem, END
49	Negative	No pathogenic fungal growth	30-1-2013	1-2-2013	Day 4: negative	Day 6: Sab.- 1x white 1mm, Sel.- negative	Day 8: Sab.- 1x white 8mm, Sel. - negative	Day 11: Sab. - 1x yellow 4mm, Sel.- negative	Day 13: Sab - 1x yellow 1 cm. Sel - negative	Day 15: Sab - 1x yellow yeast 1 cm. Sel - 1x white fluffy 3mm	Day 18: Sab - 1x yellow yeast. Sel - 1x greenwhite 2 cm	Day 20: Sel - 1x greenwhite 4cm, Sab - 1x yellowwhite yeast 2cm	Day 22: both idem, END
50	Negative	Day 15: Selective agar overgrowth (black)	30-1-2013	1-2-2013	Day 4: negative	Day 6: Sab.- 2x white 1-3mm, Sel. 2x white 1-3mm	Day 8: Sel. - 1x whiteblack 1cm, 1x green 2mm, 2x white 2-5mm, Sab. - 2x white 3-6mm, 1x grey 2mm	Day 11: Sab. - 1x d.green 6mm, 1x d.greenwhite 1cm, 1x greywhite 1cm, Sel.- 2x white bulging 2-10mm, 1x black powdery 2,5cm, 1x d.green 2m	Day 13: Sab - 1x neongreen 15mm, 1x brownwhite 15mm, 1x brown 8 mm. Sel - 1x white fluffy 15mm, w whiteyellow 3mm, 3x black 1-3mm.	Day 15: Sab - 1x neongreenyellow 15mm, 1x brownwhite 15mm, 3xdarkgreen 1mm-1cm. Sel - 1x black 4 cm, 2x black smaller, 2x white 3-15 mm (END)	Day 18: Sab - 2x darkgreen 1mm and 1 cm, 1x whitegreen 2mm. Sel - 1x whitegreenneon 2 cm, 1x whitebrown 1.5 cm	Day 20: Sab - 1x greenyellowwhite 3cm, 1x whitebrown 2cm, 2x d.green 8-15mm	Day 22: END
51	Negative	No pathogenic fungal growth	30-1-2013	1-2-2013	Day 4: negative	Day 6: Sel. - 2x white 1mm, Sab.- negative	Day 8: Sab.-1x white 2mm, Sel.- 2x white bulging 2-4mm	Day 13: Sab - idem but 1 cm. Sel - 1x white fluffy bulging 1cm, 1x yeast 3mm.	Day 15: Sab - idem, 12 mm. Sel - idem	Day 18: Sel - 1x white 12 mm, 1x whiteyellow 3 mm. Sab - idem 1.5 cm	Day 20: Sab - 1x whitebrown 3cm, Sel - 3x whiteyellow 3mm-2cm		
52	Negative	Day 13: Selective agar overgrowth (black)	30-1-2013	1-2-2013	Day 4: negative	Day 6: Sab. - 1x greenwhite 3mm, 1x yellow 2mm, Sel.- 1x green 2cm	Day 8: Sel.-1x green 5cm, 2x white 3-6mm, Sab.-2x greenwhite4mm, 3x black 1-3mm, 2x white 1-2mm	Day 11: Sab. - 4x greenwhite 1-2cm, 5x d.green 1-4mm, Sel.-1x darkgreen 7cm, 1x greenwhite 1cm	Day 13: Sab - 1x greenwhite 3cm, 7x darkgreen 1-9mm, 1x whitegeen 1cm, 1x green 1cm. Sel - 80% blackgreen, 1x greenwhite 2cm (END)	Day 15: sab - idem	Day 18: sab - greenwhite 3cm, whitegreen 1cm, lightgreen 1cm, darkgreen 1mm-1cm, cluster whitegreen >15	Day 20: Sab darkgreen/white cluster, <15. 50% plate	Day 22: Sab -idem. END
53	Negative	No pathogenic fungal growth	1-2-2013	1-2-2013	Day 4: Sab.- 1x white 1mm, Sel.- 5x white	Day6: Sel.- >20 white 1-3mm, 2x yellow 3-4mm,	Day 8: Sab: 20x white bulging with red discoloration,	Day 11: Sab. 3x d.green 1-3cm, >20 white bulging	Day 13: Sel - idem, sludge. Sab - idem	Day 15: Sel - idem. Sab - idem, 80% of plate	Day 18: both idem	Day 20: both idem	Day 22: idem. END

					1-3mm	1x greenwhite 2mm, sludge around the hairs, Sab.- >20 white transparent, 2x greenwhite bulging 3-7mm	3x greenwhite 2cm, 2x brown 2mm, Sel.->20 white bulging 1-8mm with red discoloration, 2x greenwhite 3-5mm, yeast around the hairs, 1x white fluffy 1cm	1cm, Sel.-3x darkgreen, >15 white bulging 1cm					
54	Negative	No pathogenic fungal growth	4-2-2013	5-2-2013	Day 2: negative	Day 4: negative	Day 7: negative	Day 9: Sel - negative. Sab - 1x whitegreen 4mm.	Day 11: Sel - negative. Sab - 1x whitegreen 15 mm, 1x white 3mm	Day 14: Sel - negative. Sab - 2x greenwhite 1 and 1 cm	Day 16: Sab - d.green 4cm, Sel - negative	Day 18: Sel - negative. Sab - 2x darkgreen 3.5 cm	Day 21: Sel - negative. Sab - idem
55	Negative	No pathogenic fungal growth	4-2-2013	5-2-2013	Day 2: negative	Day 4: negative	Day 7: negative	Day 9: negative	Day 11: negative	Day 14: Sel - 1x white 1 mm bulging. Sab - negative	Day 16: Sel - 2x white 1-2mm, Sel negative	Day 18: Sel - 2x white bulging 1-2mm. Sab - negative	Day 21: Sab - negative. Sel - idem
56	Negative	No pathogenic fungal growth	4-2-2013	5-2-2013	Day 2: Sab.- 1x mini yellow, Sel. - negative	Day 4: Sab.-1x miniyellow, 1x miniwhite, Sel.- negative	Day 7: Sel.- 4x white 103mm, Sab.-2x white 8mm, 3x yellow 1mm, 1x darkgreen 2mm	Day 9: Sab - 1x white 1cm, 3x green 1-3mm, 1x yeast, 1x white 1mm. Sel - 4x white 1-3mm, 1x whitegreen 5 cm, 1x yeast 4mm.	Day 11: Sab - 4x white 2-10mm, 3x green 3mm, 1x white 1mm, 1x green 1mm. Sel - 1x yeast 5mm, 1x whiteyellow 1-2mm, white 3mm, greenwhite 5mm	Day 14: Sel - 2x yeast, 2x white 3-7mm, 1x whitebrown 1cm, 2x yellow 1mm and 5mm, 2x black 1mm. Sab - 2x white 1.5cm, 2x darkgreen 3-4mm, 1x yellow 4mm	Day 16: Sel - 3x whitegrey 5-15mm, 3x yellow 2-10mm, Sab - 1x greenwhite 2,5cm, 2x d.green4mm, 1x white 2cm, 1x yellow 8mm	Day 18: Sel and sab - idem	Day 21: Sel - 5x white 1-10mm, 2x browngrey 1-2cm, 3x black 1-3mm. Sab - 3x white 2mm-3cm.1x brownwhite 4cm, 1x yellow 1cm, 1x black 5mm
57	Negative	Day 18: Sabouraud overgrowth (whiteyellow sandy)	4-2-2013	5-2-2013	Day 2: negative	Day 4: Sel. - negative, Sab.- 1x white 8mm	Day 7: Sab. - 17x white 1-20mm, 2x greenwhite 5mm, >20 darkgreen1mm, 1x red 1mm, Sel.- 15 white 1-10mm, 7x d.green1-3mm	Day 9: Sab - divers, 1x white star on new plate. Sel - divers.	Day 11: Sab - divers. Sel - divers. B plate - 4x white fluffy bulging 3-10mm	Day 14: Divers. B plate - white bulging 4x merging. 5cm	Day 16: Sab B - 95% white, Sab and Sel - diverse	Day 18: Sab - 95% overgrowth whiteyellow (END) Sel - divers. B plate: 100% white	Day 21: idem
58	Negative	Day 20: Sabouraud overgrowth (80% black), Selective agar overgrowth	5-2-2013	5-2-2013	Day 2: negative	Day 4: negative	Day 7: Sab - 1x darkgreen 2cm, 4x white bulging 1-8mm, 1x greenwhite 4mm, Sel.- 1x white 2cm,	Day 9: Sab - 1x brown 4cm, 2x white 1 cm, 1x green 1cm, 1x whitebrown 8 mm. Sel - 1x white 4cm,	Day 11: Sab - darkgreen 4-5cm, 2x white 1cm, 1x greenwhite 1cm. Sel - idem, 5cm.	Day 14: Sab - 1x darkgreen 5-6 cm, 3x whitegreen 1cm. Sel - whiteyellow poudery 6cm	Day 16: Sel - 80% yellow powdery, Sab - 50% blackbrowngreen, 4x greenwhite 1-2cm	Day 18: Sel - 95% yellow poudery. Sab - idem >60% plate	Day 20: Both overgrown

		(yellow poudery)					1x yellow 1mm	1x yellow yeast 3mm.					
59	Negative	No pathogenic fungal growth	8-2-2013	11-2-2013	Day 2: negative	Day 4: negative	Day 7: Sel - negative. Sab - 1x white 2mm	Day 9: Sel - negative, Sab - 1x greenwhite 1,5cm, 1x white 1cm	Day 11: Sab - 1x black 2cm, 1x white 1 cm. Sel - negative	Day 14: Sab - 1x blackwhite 5 cm, 1x white bulging 2cm. Sel - negative	Day 16: Sel - 1x black 1mm. Sab - 1x black 5cm, 1x grey 2cm.	Day 18: both idem	Day 21: idem. END
60	Negative	No pathogenic fungal growth	8-2-2013	11-2-2013	Day 2: negative	Day 4: Sab - 2x white star. Sel - sludge	Day 7: Sab - 2x white 1cm, 1x yellow yeast. Sel - sludge, yeast around hairs, 1x white 1cm	Day 9: Sel- sludge, 3x white 1-15mm, Sab - 5x white 1mm - 1,5cm, 1x yellowgreen 2cm	Day 11: Sel - sludge and white bulging. Sab - 6x white 1-20mm, 1x whitegreen 2cm, 2x darkgreen 1-2mm.	Day 14: Sab - 9x white 1-30mm, 1x greenwhite 3cm, 2x black 1-2mm. Sel - sludge, 1x white bulging 3mm, 1x whitebrown 2.5cm	Day 16: Sab - 7x white 2mm-2.5cm, 1x darkgreen 2mm, 1x greenwhite 3cm. Sel - sludge and 4x white 2mm-2cm	Day 18: Sab - idem. Sel - 5x white 1-2.5cm, 1x yellow 8 mm, 1x darkgreen white 1cm, 1x greenwhiteyellow 3.5cm	Day 21: idem. END

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Technical Data Sheet

Medium	***SABOURAUD B***
Recipe number	***567***

Intended use	Refer to appropriate references.
Typical formula (g/l)*	Peptic digest of animal tissue 5.0; Pancreatic digest of casein 5.0; Dextrose 40.0; Agar 15.0; Inositol 10.0; Cycloheximide 0.2; Vitamin B 1.0; Depomycin 3.0
Interpretation	Refer to appropriate references.

Quality Control

Sterility test	7 days at 30°C
Final pH at 20-25°C	5.6 ± 0.2
Appearance	Buff, opalescent gel

Growth performance

Control strains	Test method	Criteria
C. albicans ATCC 10231	semi-quantitative	recovery >80%

Remarks	n.a.

** Adjusted and/or supplemented as required to meet performance criteria*

Storage instructions: petridishes 2-15°C; petridishes P090MF-R055QC/tubes/bottles/jars/bags 2-25°C; minimize exposure to light.
Version 2, 01-01-2009

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