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Pet rabbits (*Oryctolagus cuniculus*) and guinea pigs (*Cavia porcellus*) as vehicles of pathogenic and allergenic fungi

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Abstract

Nowadays, rabbits and guinea pigs are frequently adopted as companion animals, representing a vehicle for the dissemination of potentially pathogenic and allergenic fungi to their tutors. This study aimed to characterize the cutaneous mycobiota of these species and evaluate the association between mycological cultures results and several variables related to these animals' husbandry. Hair and scales samples (n=102) were collected from 32 rabbits and 19 guinea pigs: 51 by pulling hairs surrounding lesions and collecting scales (if lesions present) or along the body of the animal (if lesions absent). The other 51 samples were collected using Mackenzie's technique. Samples were inoculated in Sabouraud Chloramphenicol Agar and Dermatophyte Test Media and observed daily during the incubation period. Isolated fungal species were identified based on their macro and microscopic morphology. A questionnaire was provided to the animal tutors to collect information on animal husbandry. The most frequently isolated species corresponded to saprophytic fungi, such as *Aspergillus* spp., *Penicillium* spp., *Scopulariopsis* spp., and yeasts such as *Candida* spp. and *Rhodotorula* spp. were also found. Statistical analysis showed that a positive mycological culture was related to animal's age (p-value 0.00221) and the administration of ongoing medication (p-value 0.01532 for the variable anti-inflammatory drugs and 0.03680 for the variable antibiotics), while the number of isolated fungal species was related to animal's species (p-value 0.00469 for the variable yard) and outdoor access (p-value 0.023260). These fungi have already been reported as responsible for mycotic infections in humans and animals, including dogs and cats, although they usually affect immunocompromised individuals. Therefore, these animals can represent a zoonotic risk, which may be related to animals age, species, ongoing medication, and outdoor access.

Keywords: Fungi, Guinea pigs, Rabbit, Saprophytes, Skin

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Introduction

Fungi are ubiquitous microorganisms. The great majority are saprophytic, which are playing an important role in the environment, contributing to the decaying of organic matter (Freitas, 2010). Similarly, most pathogenic fungi are exogenous, being also present in the environment, water or soil, but can also belong to the animals mycobiota and cause disease (mycosis) when the host presents an organic imbalance (Mitchell, 2013). Finally, fungi can also have a potential allergenic effect (Reedy et al., 1989).

Pets can be potential vehicles for pathogenic and allergenic fungi. The most frequent mycosis in pet rabbits (*Oryctolagus cuniculus*) and guinea pigs (*Cavia porcellus*) is dermatophytosis, with *Trichophyton mentagrophytes* and, less frequently, *Microsporum canis* being the most common associated species in these animals (Donnelly et al., 2000). Some outbreaks of *T. benhamiae* and *T. mentagrophytes* in humans have been reported. These are zoophilic dermatophytic species, frequently transmitted by asymptomatic animals e.g. guinea pigs and rabbits (Contet-Audonneau and Leyer, 2010; Bloch et al., 2016; Maldonado et al., 2021). Typical dermatophytosis lesions are circular, irregular, or diffuse alopecia, localized or generalized, presenting scaling, crusting, and erythema of the borders. Lesions develop most frequently on the head and extremities in both species and also in the thorax of guinea pigs.

These animals can also present asymptomatic or subclinical disease or develop a carrier state (Donnelly et al., 2000; Vangeel et al., 2000; Hoppmann and Barron, 2007; Kraemer et al., 2012, 2013; Turner et al., 2017), representing a potential dissemination vector of zoonotic infections to their tutors, especially to immunocompromised individuals and children (Vangeel et al., 2000; Drouot et al., 2009; Kraemer et al., 2012, 2013).

Pet rabbits and guinea pigs can also present non-dermatophytic mycosis. Despite being rare, cases of cutaneous cryptococcosis in guinea pig (van Herck et al., 1988), cheilitis by *Candida* spp. in guinea pigs (Scott, 2001; Mignon et al., 2003; White et al., 2003), pulmonary and cutaneous aspergillosis in rabbits (Scott, 2001) and infections by *Malassezia* spp. in pet rabbits and guinea pigs were already reported (Radi, 2004; Cabañes et al., 2011; Miller Jr et al., 2013; Quevedo et al., 2014; White et al., 2016).

Some studies have been carried out regarding the characterization of the cutaneous mycobiota of rabbits (Feuerman et al., 1975; Balsari et al., 1981; Vangeel et al., 2000; Kraemer et al., 2012) and guinea pigs (Feuerman et al., 1975; Balsari et al., 1981; Vangeel et al., 2000; Kraemer et al., 2012; d'Ovidio et al., 2014; Dey et al., 2016; Overgaauw et al., 2017), but such studies are not available in Portugal, where there is no information on the mycobiota of these pets, its role on these animals health, and the influence of husbandry on fungal colonization. Such data would contribute to evaluate the role of these pets as potential carriers for zoonotic fungal species.

Therefore, the present study aimed to characterize the skin and hair mycobiota of pet rabbits and guinea pigs presented for consultation at the University Teaching Hospital of Faculty of Veterinary Medicine, University of Lisbon, Portugal, and to determine the association between animals related factors and their colonization by fungi.

Material and Methods

Inclusion criteria

This study included 32 (62.7%) rabbits and 19 (37.3%) guinea pigs. Animals were presented to the medical consultation at the University Teaching Hospital of Faculty of Veterinary Medicine, University of Lisbon, Portugal, between February 5, 2018, and July 6, 2018. No selection was performed based on animal's age, gender, and health status.

Sample collection

From each animal, samples were collected by two methods. The first method consisted of pulling hairs and collecting scales from the periphery of lesions, if present, with sterile Halsted Mosquito hemostatic forceps. In the case of animals without apparent lesions, samples were collected from the areas most frequently associated with dermatophytosis lesions in the rabbit and guinea pig, such as the head and extremities in both species and the thorax in guinea pigs (Donnelly et al., 2000; Turner et al., 2017).

Samples were also collected using the Mackenzie's technique, which consists of passing a sterilized toothbrush through the entire coat of the animal thirty times or until it has hairs and visible skin residues (Moriello, 2001). A total of 102 samples were collected, 51 by the pulling method and 51 by Mackenzie's technique. Samples were collected by trained veterinarians following standard routine procedures with the consent of the owner, and no ethics committee approval was needed.

Sample processing

Samples were inoculated in two different culture media, Sabouraud Chloramphenicol Agar (SCA, VWR, Belgium) and Dermatophyte Test Media (DTM, Merck, USA). All samples were incubated at a temperature of 25-27°C, for 21-days (SCA) or 14-days (DTM) (Moriello, 2001; Moriello et al., 2017). During the incubation period, inoculated media were observed daily. Colonies obtained were identified at the genus level based on their macro and microscopic morphology (Procop, 2016).

Statistical analysis

A questionnaire was provided to the tutors to collect detailed information about the history, husbandry, and characterization of the animals. The statistical analysis was performed in the R software (Anonymous, 2021). Fit models were used to evaluate the relationship between dependent variables (positivity for dermatophytes, positivity in mycological culture, and the number of isolated fungal species) and independent variables (species, gender, age, origin, dental disease, contact with other animals, hygiene, outdoor access, food, and ongoing medication), accessed through anamnesis and the questionnaire.

Two different regression models were used since dependent variables with different structures were observed. First, for the variable's positivity for dermatophytes and positivity in the mycological culture, a logistic regression model was used, which considered the value "1" to represent the presence of dermatophytes or fungal species and the value "0" to represent their absence. Second, a linear regression model was used for the variable number of isolated fungal species.

For each independent variable, the regression coefficient, standard deviation, and p-value were determined after defining a reference category used for comparison. The variable species were divided into two categories, rabbit and guinea pig, with the reference category corresponding to the guinea pig. The variable outdoor access was divided into five categories, street, backyard, public garden, terrace, and none, with the reference category corresponding to none. The variable ongoing medication refers to the medication the animal was taking at the time of sampling and was divided into eight categories, anti-inflammatory, antinematodal drugs, antibiotic, antifungal, analgesic, vitamins, probiotics, and none, with the reference category corresponding to none.

Results

Population characterization

Of the 51 animals sampled, 24 (45.1%) were males and 27 (54.9%) females. Of the 32 rabbits, 18 (56.25%) were male, and 14 (43.75%) were female; in its turn, of the 19 guinea pigs, 6 (31.6%) were male, and 13 (68.4%) were female. Animal's age was between one month and a half and 10 years and 2 months , with an average of 3.70 years and a median of 3.10 years.

Animals were presented for consultation due to several reasons. Approximately half of the animals (n=24; 47.1%) were presented for a preventive medicine consultation, with the remaining being presented for consultation due to problems in other organic systems. The most frequent consultations were of dermatology, neurology, and dentistry, with the same number of cases (9.8% each). Despite that, dermatological signs were found in 14 animals presented for non-related consultations.

In total, 19 animals (37.3%) showed diverse dermatological lesions at the moment of sampling. The most frequent were scaling (n=7, 28.0%), alopecic lesions (n=6, 24.0%), and pododermatitis (n=5, 20.0%), followed by moist dermatitis (n=2, 8.0%) and mites, pruritus, lipoma, otitis externa and thinned skin (n=1 each, 4.0% each). None of the pet owners showed any skin lesions. The following Table 1 summarizes the skin lesions observed, categorized based on by species.

The majority of the animals (n=36; 70.6%) was not taking any medication at the time of sampling, with the following exceptions: 7 animals (13.7%) were taking anti-inflammatories, 8 animals (15.7%) antibiotics, 5 animals (9.8%) antinematodal drugs, 2 animals (3.9%) vitamins, and 1 animal (2.0%) probiotics. Of the animals included in this study, 43.1% (n=22) had contact with individuals at risk, such as children (n=10, 19.6%), elderly (n=10, 19.6%) and pregnant women (n=2, 3.9%). Most were exclusively indoor (n=40, 78.4%), as only 11 animals (n=11, 21.6%) had occasional access to the street, most of which to a backyard (n=7, 13.7%) and the others to the public garden (n=2, 3.9%), terrace (n=1, 2.0%) and street (n=1, 2.0%).

Isolated fungal species

Eleven fungal genera were isolated from the collected samples. Unfortunately, it was not possible to isolate any dermatophytes. Nine genera of saprophytic molds were identified, representing 93.5% of the total number of fungal isolates obtained, and also two genera of yeasts, representing 6.5% of the isolates obtained (Table 2). It was possible to obtain 157 saprophytic moulds isolates, belonging to the following genera: *Aspergillus* (n=45, 26.8%), *Scopulariopsis* (n=26, 15.5%), *Penicillium* (n=25, 14.9%), *Mucor* (n=12, 7.1%), *Rhizopus* (n=12, 7.1%), *Cladosporium* (n=8, 4.8%), *Alternaria* (n=7, 4.2%), *Phoma* (n=2, 1.2%) and *Chaetomium* (n=1, 0.6%). Additionally, 11.3% of the colonies obtained (n=19) were not possible to identify. It was also possible to obtain 11 yeast isolates, identified as *Candida* spp. (n=9, 5.4%) and *Rhodotorula* spp. (n=2, 1.2%).

Statistical analysis

Statistical analysis results are presented in Table 3 and Table 4. The number of isolated fungal genera was significantly related to the animal species since the regression coefficient for the rabbit category was found to be negative. Rabbit samples are more likely to originate cultures with a lower number of fungal species than guinea pig samples (reference category).

Animal age was significantly related to a positive mycological culture. The coefficient for the age category was found to be positive, indicating that older animals are more likely to produce a positive fungal culture. Results obtained from animals adopted from a pet shop were significantly related to a positive mycological culture. However, all these animals were adults at the time of sample collection, a long period after their permanence in the pet shop, which reduces the relevance of this relation for further discussion.

The number of isolated fungal species is related to outdoor access. The coefficients of the categories backyard and public garden were found to be positive, suggesting a higher probability that samples obtained from animals with outdoor access, especially those who have access to a backyard, will originate cultures with a higher number of fungal species than those obtained from animals without outdoor access (reference category). A positive mycological culture was also related to outdoor access, specifically the public garden. However, only one observation contributed to this result, and therefore this result was not considered for further discussion. The same was observed for the number of isolated fungal species and the contact with other animals (dogs).

Finally, a positive mycological culture was found to be related to the animal's ongoing medication. The coefficients of the categories anti-inflammatory, antinematodal drugs, and antibiotics were found to be positive, indicating that animals taking these medications have a higher probability of originating a positive mycological culture than samples obtained from animals that were not taking any medication (reference category).

Discussion

Pet rabbits and guinea pigs can be potential vehicles for zoonotic or allergenic fungi to their tutors. Results revealed that the cutaneous mycobiota of the rabbits and guinea pigs in this study was mainly composed of environmental filamentous saprophytic fungi and yeasts, as described in other studies performed on these animal species (Balsari et al., 1981; Vangeel et al., 2000; Kraemer et al., 2013; d'Ovidio et al., 2014), as well as in others, such as dogs and cats (Balsari et al., 1981; Aho, 1983; Moriello and Deboer, 1991; Khosravi, 1996; Simpanya and Baxter, 1996; Sierra et al., 2000; Paixão et al., 2001; Bernardo et al., 2005; Stojanov et al., 2007; d'Ovidio et al., 2014).

Saprophytic fungi were expected to be found in these animals because of their permanent contact with organic matter present in the hay, food substrate, and the environment. The yeast species identified were

Table 1: Absolute frequency (AF) and relative frequency (RF) of the skin lesions in sampled rabbits and guinea pigs.

Signs	**Rabbit**		**Guinea pigs**		**Total**	
	AF	RF (%)	AF	RF (%)	AF	RF (%)
Alopecia	5.0	31.25	3.0	21.43	8.0	26.67
Mites	0.0	0.0	1	7.14	1.0	3.33
Humid dermatitis	2	12.50	1.0	7.14	3.0	10.0
Crusts	0.0	0.0	1.0	7.14	1.0	3.33
Lipoma	1.0	6.25	0.0	0.00	1.0	3.33
External otitis	1.0	6.25	0.0	0.0	1.0	3.33
Thin skin	0.0	0.00	1.0	7.14	1.0	3.33
Pododermatitis	4.0	25.0	3.0	21.43	7.0	23.33
Itching	1.0	6.25	0.0	0.0	1.0	3.33
Seborrhea	2.0	12.50	4.0	28.57	6.0	20.00
Total	16.0	100.00	14	100.00	30.0	100.00

also expected, as they are commensals of animals and humans microbiota (*Candida* spp.) (Hartigan, 2011) or humid environments (*Rhodotorula* spp.) (Pressler, 2012). As far as we know, this study allowed the isolation of three fungal genera in rabbits and guinea pigs for the first time, namely *Chaetomium* spp., *Phoma* spp., and *Rhodotorula* spp., which were already isolated from dogs and cats (Aho, 1983; Cabañes et al., 1996; Sierra et al., 2000; Paixão et al., 2001; Bernardo et al., 2005).

It was impossible to isolate dermatophytes from the animals under study; however, the other identified fungi were already reported as being responsible for cutaneous lesions in humans, dogs, and cats. In humans, *Aspergillus* spp., *Alternaria* spp., *Candida* spp., *Chaetomium* spp., *Cladosporium* spp., *Mucor* spp., *Penicillium* spp., *Phoma* spp., *Rhizopus* spp., and *Scopulariopsis* spp. have been reported to be responsible for cutaneous infections (Cox and Irving, 1993; Vieira et al., 2001; Hope et al., 2005; Roden et al., 2005; Brusselaers et al., 2011; Ahmed et al., 2016; Avsever et al., 2017).

In dogs and cats, *Alternaria* spp., *Aspergillus* spp., *Candida* spp., *Cladosporium* spp., *Mucor* spp., and *Penicillium* spp. have also been reported as responsible for cutaneous infections (Manning, 1983; Jand and Gupta, 1989; Raposo et al., 1996; Mueller et al., 2002; Avsever et al., 2017). Most of them were not previously associated with infections in pet rabbits and guinea pigs, except for *Aspergillus* spp., which was already related to cutaneous infections in rabbits (Scott, 2001). However, it is important to refer that *Scopulariopsis* spp. was isolated from one animal in the current study, with alopecia and crusting lesions on the extremities. Several factors need to be considered before relating this species to infection, namely the concentration of isolated fungi, the clinical status of the animal, and the possibility of contamination of the sample or culture (Borman and Johnson, 2014).

Although the hypothesis of colonization of a preexisting wound cannot be ruled out, results seem to indicate that this fungus may have been responsible for this lesion, especially because of its association with lesions that are very similar to dermatophytosis in humans has been described (Cox and Irving, 1993). In fact, identified species may have the ability to cause disease in rabbits and guinea pigs, not only in animals with immunosuppression or deficient nutritional status, but also in healthy individuals after abrasive or perforating injury (Avsever et al., 2017). Therefore, veterinarians should consider non-dermatophytic fungal infections as a differential diagnosis for cutaneous lesions.

Many of the variables under study are frequently referred to as predisposing factors for dermatophytosis in guinea pigs and rabbits (Donnelly et al., 2000). Dermatophytes were not isolated, but the association between positive mycological cultures and the number of fungal species isolated with several independent variables was evaluated.

It was possible to observe that samples collected from guinea pigs are likely to originate cultures with a higher number of fungal species than those from rabbits, which seems to indicate that the self-cleaning habits of guinea pigs are less efficient than the ones from rabbits. Also, unlike rabbits, mutual grooming is less frequent between different guinea pigs (Quesenberry et al., 2012). Self-cleaning allows the maintenance of animal hygiene by removing dirt and parasites (Feusner et al., 2009). If not performed, the skin surface presents a higher probability of fungi colonization and dermatological disease development, which may also explain the higher tendency of guinea pigs for dermatophytosis (Rosenthal and Wapnick, 1963; Feuerman et al., 1975; Balsari et al., 1981; Donnelly et al., 2000; Vangeel et al., 2000; Kraemer et al.,

Table 2: Absolute frequency and relative frequency of the skin lesions in sampled rabbits and guinea pigs.

	Species	Absolute frequency	Relative frequency (%)
Filamentous fungi	*Alternaria* spp.	7	4.2
	Aspergillus spp.	45	26.8
	Chaetomium spp.	1	0,6
	Cladosporium spp.	8	4.8
	Mucor spp.	12	7.1
	Penicillium spp.	25	14.9
	Phoma spp.	2	1.2
	Rhizopus spp.	12	7.1
	Scopulariopsis spp.	26	15.5
	Non-identified filamentous fungi	19	11.3
Yeasts	*Candida* spp.	9	5.4
	Rhodotorula spp.	2	1.2
Total	–	168	100.0

Table 3: Results of the analysis of the relationship between the positivity in mycological culture and the independent variables.

Variable of interest (dependent variable)[1]	Independent variable	Categories	Coefficient	Standard deviation	p-value[2]
Positivity in mycological culture	Species	Rabbit	-0.40668	0.24066	0.1050
	Age	–	0.04851	0.02499	0.0647[a]
	Origin	Petshop	0.53630	0.29783	0.0854[a]
		Breeder	0.46008	0.46299	0.35808
		Particular	0.44375	0.40295	0.2821
	Dental disease	Presence	-0.07853	0.12799	0.5462
	Contact with other animals	Dog	0.72085	0.45056	0.1220
		Cat	-0.43221	0.49383	0.3907
		Rabbit	0.36465	0.25930	0.1728
		Guinea pig	0.33537	0.27617	0.2381
		Chinchilla	0.12792	0.66845	0.8489
	Food	Pellets	0.48166	0.47429	0.3115
		Hay	0.39097	0.45443	0.3910
		Fruit	0.47627	0.45909	0.3012
		Vegetables	0.48032	0.44924	0.2867
		Bread	0.38270	0.55330	0.4904
	Outdoor access	Street	0.17066	0.35405	0.6354
		Backyard	0.34288	0.20971	0.1150
		Public garden	0.79459	0.47662	0.0977[a]
		Terrace	-0.20724	0.36657	0.5780
	Hygiene	Yes	0.05344	0.21650	0.8074
	Ongoing medication	Anti-inflammatory	0.44386	0.21230	0.0444[b]
		Antinematodal drug	0.65259	0.32102	0.0460[b]
		Antibiotic	0.50278	0.22179	0.0296[b]
		Antifungal	-0.08474	0.53469	0.8744
		Analgesic	0.10099	0.31883	0.7524
		Vitamins	-0.23892	0.34387	0.4902
		Probiotics	-0.17014	0.53340	0.7504

[1]The table represents the results obtained after regression analysis between the dependent variable positivity in mycological culture and the independent variables.

[1]Symbols presented in front of the p-value indicate that the variable is significant for the most usual levels of statistical significance: a= p<0.1; b= p<0.05.

2012). In fact, a study in cats with dermatophytosis reported that collared cats show more generalized lesions compared to animals without Elizabethan collars that could perform their self-cleaning, suggesting

Table 4: Results of the analysis of the relationship between the number of isolated fungal species and the independent variables.

Variable of interest (dependent variable)[1]	Independent variable	Categories	Coefficient	Standard deviation	p-value[2]
Number of isolated fungal species	Species	Rabbit	-0.944045	0.484200	0.064839a
	Genre	Male	0.002295	0.255143	0.992933
	Age	–	0.048328	0.049690	0.342146
	Origin	Petshop	00.274939	0.591464	0.646941[a]
		Breeder	1.382482	0.918278	0.146825
		Particular	-0.090275	0.798079	0.910995
	Dental disease	Presence	-0.300154	0.258755	0.260123
	Contact with other animals	Dog	1.815347	0.889183	0.053652a
		Cat	0.107732	0.981259	0.913665
		Rabbit	0.460553	0.513964	0.380614
		Guinea pig	0.638197	0.553708	0.263399
		Chinchilla	-1.010326	1.252532	0.423662
	Food	Pellets	1.328621	0.825614	0.109752
		Hay	0.916870	0.790083	0.247789
		Fruit	0.385158	0.799038	0.630519
		Vegetables	0.488230	0.782261	0.533530
		Bread	0.542895	0.948621	0.568140
	Outdoor access	Street	0.380810	0.714589	0.601004
		Backyard	1.012497	0.414648	0.023260b
		Public garden	1.486831	0.847660	0.081777a[a]
		Terrace	0.201894	0.735818	0.786843
	Hygiene	Yes	-0.378714	0.438532	0.397827
	Ongoing medication	Anti-inflammatory	0.461022	0.411607	0.271631
		Antinematodal drug	0.953670	0.596988	0.114909
		Antibiotic	0.683885	0.427860	0.119800
		Antifungal	0.913216	0.970479	0.349053
		Analgesic	0.214098	0.592782	0.719223
		Vitamins	-0.715989	0.646202	0.273205
		Probiotics	-0.049822	0.970504	0.959165

[1] The results of the regression analysis between the dependent variable number of isolated fungal species and the independent variables.

[1] Symbols presented in front of the p-value indicate that the variable is significant for the most usual levels of statistical significance: a= $p<0.1$; b= $p<0.05$.

that the cleaning behavior may limit the development of lesions (Hawkins and Bishop, 2012).

Age was found to be related to positive mycological cultures. Older animals may suffer from diseases that impair cleaning behaviors, such as dental disease associated with oral pain or musculoskeletal disorders that reduce mobility and cause pain (Harcourt-Brown, 2007; Fisher and Carpenter, 2012).

Regarding outdoor access, it was already reported that fungal species present in cats' hair might vary depending on the environment (Cabañes, 2000). Considering that the organic matter present in gardens and backyards constitutes a relevant substrate for saprophytic fungi, it is expected that samples collected from animals with outdoor access originate cultures with a higher number of fungal species.

Prolonged antimicrobial therapy can influence the composition of the commensal microbiota of mucosa and facilitate yeast proliferation (Hartigan, 2011). It is reported that the skin and hair microbiota may vary according to the immune state (Cabañes, 2000). In this study, samples from animals under drug therapy (anti-inflammatory, antinematodal drugs, and antibiotic) presented a higher probability of originating positive cultures. These animals were mainly diagnosed with advanced dental disease or presented neurological signs compatible with *Encephalitozoon cuniculi*. These two situations generally promote a decrease in body condition, poor nutritional status, and an altered degree of activity (Harcourt-Brown, 2007; Fisher and Carpenter, 2012; Hawkins and Bishop, 2012), which may explain this result.

It is important to refer that this study had some limitations. First, dermatophytes were not identified in any sample, which was not expected. However, it is important to refer that few studies are available regarding the true prevalence of dermatophytes in pet rabbits and guinea pigs. It is important to continue performing these studies, including more animals from distinct locations, to understand the impact of dermatophytosis in these animals. Also, in future studies, it will be interesting to evaluate the relationship between a positive mycological culture and the clinical status of the animals, along with an environmental mycological evaluation.

To the best of our knowledge, this is the first study regarding cutaneous mycobiota of pet rabbits and guinea pigs performed in Portugal. Although dermatophytes were isolated, results showed that these animals can carry several filamentous fungi and yeasts in their hair and skin. Three of these genera were isolated for the first time in these animals, namely *Chaetomium*, *Phoma*, and *Rhodotorula*. All the isolated fungi are frequently present in the environment and usually do not cause disease. However, most of the identified fungi infections were already described in animals and humans, especially in immunocompromised individuals. Therefore, these animals can represent a zoonotic risk, which may be related to the animal's age, species, ongoing medication, and outdoor access.

Conclusions

Pet rabbits and guinea pigs are frequently adopted as companion animals in close contact with their owners. In this study, we performed the characterization of animal's skin and hair mycobiota and we evaluated its relationship with the animal's medical records. It was possible to observe that these animals can carry several filamentous fungi and yeasts in their hair and skin. The most frequently isolated species corresponded to saprophytic fungi, such as *Aspergillus* spp., *Penicillium* spp., *Scopulariopsis* spp.; *Candida* spp. and *Rhodotorula* spp. In addition, the three fungal genera *Chaetomium* spp., *Phoma* spp., and *Rhodotorula* spp. were identified for the first time in pet rabbits and guinea pigs. Animal age or the presence of ongoing medication were associated with a positive mycological culture, while the number of isolated fungal species was related to the animal's species and outdoor access. It is worth mentioning that infections related to most of the identified fungi were already described in animals and humans, reinforcing the importance of the mycological assessment in these animals, as they may represent a potential zoonotic risk.

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References

Ahmed, S.A., Khan, Z., Wang, X.w., Moussa, T.A.A., Al-Zahrani, H.S., Almaghrabi, O.A., Sutton, D.A., Ahmad, S., Groenewald, J.Z., Alastruey-Izquierdo, A., van Diepeningen, A., Menken, S.B.J., Najafzadeh, M.J., Crous, P.W., Cornely, O., Hamprecht, A., Vehreschild, M.J.G.T., Kindo, A.J., de Hoog, G.S., 2016. *Chaetomium*-like fungi causing opportunistic infections in humans: A possible role for extremotolerance. Fungal Diversity 76, 11–26. 10.1007/s13225-015-0338-5.

Aho, R., 1983. Saprophytic fungi isolated from the hair of domestic and laboratory animals with suspected dermatophytosis. Mycopathologia 83, 65–73. 10.1007/BF00436886.

Anonymous, 2021. Team-core. a language and environment for statistical computing. https://www.gbif.org/tool/81287/r-a-language-and-environment-for-statistical-computing. access date: 22-12-2021.

Avsever, M.L., Hilmioglu, P.S., Camkerten, I., Aksoy, A., 2017. First isolation of *Alternaria alternata* from a dog in Turkey. Ankara Üniversitesi Veteriner Fakültesi Dergisi 64, 137–139. 10.1501/Vetfak_0000002788.

Balsari, A., Bianchi, C., Cocilovo, A., Dragoni, I., Poli, G., Ponti, W., 1981. Dermatophytes in clinically healthy laboratory animals. Laboratory Animals 15, 75–77. 10.1258/002367781780958649.

Bernardo, F., Lança, A., Guerra, M., Martins, H.M., 2005. Dermatophytes isolated from pet , dogs and cats , in lisbon , Portugal (2000-2004). Rev. Port. Ciências Veterinárias 100, 85–88. URL: https://www.semanticscholar.org/paper/Dermatophytes-isolated-from-pet-%{2C}-dogs-and-cats-%{2C}-%{2C}-Bernardo-Lan%C3%A7a/0606f73251cacb4f944bddf5933f4b0c15c9baf5.

Bloch, M., Cavignaux, R., Debourgogne, A., Dorin, J., Machouart, M., Contet-Audonneau, N., 2016. From guinea pig to man: *Tinea* outbreak due to *Trichophyton mentagrophytes* var. *porcellae* in pet shops in Nancy, France. Journal de Mycologie Medicale 26, 227–232. 10.1016/j.mycmed.2016.05.005.

Borman, A.M., Johnson, E.M., 2014. Interpretation of fungal culture results. Current Fungal Infection Reports 8, 312–321. 10.1007/s12281-014-0204-z.

Brusselaers, N., Blot, S., Vogelaers, D., 2011. Deep-seated *Candida* infections in the intensive care unit. Netherlands Journal of Critical Care URL: https://biblio.ugent.be/publication/1256066.

Cabañes, F., 2000. Dermatofitosis animales. recientes advances. Revista Iberoamericana de Micología 17, 8–12.

Cabañes, F.J., Abarca, M.L., Bragulat, M.R., Castellá, G., 1996. Seasonal study of the fungal biota of the fur of dogs. Mycopathologia 133, 1–7. 10.1007/bf00437092.

Cabañes, F.J., Vega, S., Castellá, G., 2011. *Malassezia cuniculi* spp. A novel yeast species isolated from rabbit skin. Medical Mycology 49, 40–48. 10.3109/13693786.2010.493562.

Contet-Audonneau, N., Leyer, C., 2010. Émergence ddermatophyte transmis par le cochon d et proche de *Trichophyton mentagrophytes* var. *erinacei*: *T. mentagrophytes* var. *porcellae*. Journal de Mycologie Médicale 20, 321–325. 10.1016/j.mycmed.2010.08.001.

Cox, N.H., Irving, B., 1993. Cutaneous 'ringworm' lesions of *Scopulariopsis brevicaulis*. The British Journal of Dermatology 129, 726–728. 10.1111/j.1365-2133.1993.tb03341.x.

Dey, J.C., Rahman, M.K., Rumi, M.A., Dutta, A., Halder, B.C., Mannan, A., Hassan, M.M., Hossain, M.S., Sayeed, M.A., 2016. Prevalence of dermatophytosis in rabbits at SAQTVH, Chittagong, Bangladesh. Journal of Dairy, Veterinary & Animal Research 3. 10.15406/jdvar.2016.03.00100.

Donnelly, T.M., Rush, E.M., Lackner, P.A., 2000. Ringworm in small exotic pets. Seminars in Avian and Exotic Pet Medicine 9, 82–93. 10.1053/AX.2000.5045.

d'Ovidio, D., Grable, S.L., Ferrara, M., Santoro, D., 2014. Prevalence of dermatophytes and other superficial fungal organisms in asymptomatic guinea pigs in Southern Italy. The Journal of Small Animal Practice 55, 355–358. 10.1111/jsap.12216.

Drouot, S., Mignon, B., Fratti, M., Roosje, P., Monod, M., 2009. Pets as the main source of two zoonotic species of the *Trichophyton mentagrophytes* complex in Switzerland, *Arthroderma vanbreuseghemii* and *Arthroderma benhamiae*. Veterinary Dermatology 20, 13–18. 10.1111/j.1365-3164.2008.00691.x.

Feuerman, E., Alteras, I., Hönig, M.D., Lehrer, N., 1975. Saprophytic occurrence of *Trichophyton mentagrophytes* and *Microsporum gypseum* in the coats of healthy laboratory animals (Preliminary report). Mycopathologia 55, 13–15. 10.1007/BF00467084.

Feusner, J.D., Hembacher, E., Phillips, K.A., 2009. Pathologic grooming in animals and humans. CNS Spectrums 14, 503–513. 10.1017/s1092852900023567.

Fisher, P.G., Carpenter, J.W., 2012. Neurologic and musculoskeletal diseases, in: Ferrets, rabbits, and rodents. Elsevier, pp. 245–256. 10.1016/B978-1-4160-6621-7.00019-1.

Freitas, G., 2010. Fungos, in: Sousa, J., de, Ferreira, W., Lima, N. (Eds.), Microbiologia. Edições Técnicas, Lidel, Lisboa, p. 145–165. URL: http://repositorium.sdum.uminho.pt/handle/1822/32687.

Harcourt-Brown, F.M., 2007. The progressive syndrome of

acquired dental disease in rabbits. Journal of Exotic Pet Medicine 16, 146–157. 10.1053/j.jepm.2007.06.003.
Hartigan, P., 2011. Mycology: General features of fungi associated with disease in animals, in: Quinn, P.J., Markey, B.K., Leonard, F.C., Hartigan, P., Fanning, S., Fitzpatrick, E.S. (Eds.), Veterinary Microbiology and Microbial Disease. 2 ed.. Wiley-Blackwell, p. 1102–1117.
Hawkins, M.G., Bishop, C.R., 2012. Disease problems of guinea pigs, in: Ferrets, rabbits, and rodents. Elsevier, pp. 295–310. 10.1016/B978-1-4160-6621-7.00023-3.
van Herck, H., van den Ingh, T.S., van der Hage, M.H., Zwart, P., 1988. Dermal cryptococcosis in a guinea pig. Laboratory Animals 22, 88–91. 10.1258/002367788780746494.
Hope, W.W., Walsh, T.J., Denning, D.W., 2005. The invasive and saprophytic syndromes due to *Aspergillus* spp. Medical Mycology 43 Suppl 1, S207–38. 10.1080/13693780400025179.
Hoppmann, E., Barron, H.W., 2007. Ferret and rabbit dermatology. Journal of Exotic Pet Medicine 16, 225–237. 10.1053/j.jepm.2007.10.002.
Jand, S.K., Gupta, M.P., 1989. Dermatomycosis in dogs. Mycoses 32, 104–105. 10.1111/j.1439-0507.1989.tb02213.x.
Khosravi, A.R., 1996. Fungal flora of the hair coat of stray cats in Iran. Mycoses 39, 241–243. 10.1111/j.1439-0507.1996.tb00133.x.
Kraemer, A., Hein, J., Heusinger, A., Mueller, R.S., 2013. Clinical signs, therapy and zoonotic risk of pet guinea pigs with dermatophytosis. Mycoses 56, 168–172. 10.1111/j.1439-0507.2012.02228.x.
Kraemer, A., Mueller, R.S., Werckenthin, C., Straubinger, R.K., Hein, J., 2012. Dermatophytes in pet guinea pigs and rabbits. Veterinary Microbiology 157, 208–213. 10.1016/j.vetmic.2011.12.005.
Maldonado, I., Elisiri, M.E., Monaco, M., Hevia, A., Larralde, M., Fox, B., Refojo, N., Iachini, R., Santos Muñoz, A., Leitner, R., Luna, P., Meirama, L., Abad, E., Ruiz Diaz, N., Fernández, J., Fernández-Canigia, L., 2021. *Trichophyton benhamiae*: AN emergent zoonotic pathogen in Argentina associated with guinea pigs: Description of 7 cases in Buenos Aires. Revista Argentina de Microbiología 10.1016/j.ram.2021.02.004.
Manning, T.O., 1983. Cutaneous diseases of the paw. Clinics in Dermatology 1, 131–142. 10.1016/0738-081x(83)90048-2.
Mignon, B., Symoens, F., Losson, B., 2003. Selected abstracts from the European Society of Veterinary Dermatology and European College of Veterinary Dermatology 18th Annual Congress Nice, France, 26-28 September 2002. Veterinary Dermatology 14, 237–267. 10.1046/j.1365-3164.2003.00343.x.
Miller Jr, W.H., Griffin, C.E., Campbell, K.L., 2013. Muller and Kirk's Small Animal Dermatology. 7 ed., Saunders.
Mitchell, T., 2013. Medical mycology, in: Brooks, G., Carroll, K., Butel, J., Morse, S., Mietzner, T. (Eds.), Jawetz, Melnick, &Adelberg Medical Microbiology. 26 ed., p. 671–713. URL: https://accessmedicine.mhmedical.com/Content.aspx?bookid=1551&sectionid=94111496.
Moriello, K.A., 2001. Diagnostic techniques for dermatophytosis. Clinical Techniques in Small Animal Practice 16, 219–224. URL: http://dx.doi.org/10.1053/svms.2001.27597, 10.1053/svms.2001.27597.
Moriello, K.A., Coyner, K., Paterson, S., Mignon, B., 2017. Diagnosis and treatment of dermatophytosis in dogs and cats: Clinical Consensus Guidelines of the World Association for Veterinary Dermatology. Veterinary Dermatology 28, 266–e68. 10.1111/vde.12440.
Moriello, K.A., Deboer, D.J., 1991. Fungal flora of the haircoat of cats with and without dermatophytosis. Journal of Medical and Veterinary Mycology 29, 285–292. 10.1080/02681219180000441.
Mueller, R.S., Bettenay, S.V., Shipstone, M., 2002. Cutaneous candidiasis in a dog caused by *Candida guilliermondii*. The Veterinary Record 150, 728–730. 10.1136/vr.150.23.728.
Overgaauw, P.A.M., Avermaete, K.H.A.v., Mertens, C.A.R.M., Meijer, M., Schoemaker, N.J., 2017. Prevalence and zoonotic risks of *Trichophyton mentagrophytes* and *Cheyletiella* spp. in guinea pigs and rabbits in Dutch pet shops. Veterinary Microbiology 205, 106–109. 10.1016/j.vetmic.2017.05.008.
Paixão, G., Sidrim, J., Campos, G., Brilhante, R., Rocha, M., 2001. Dermatophytes and saprobe fungi isolated from dogs and cats in the city of Fortaleza, Brazil. Arquivo Brasileiro de Medicina Veterinária e Zootecnia 53, 568–573. 10.1590/S0102-09352001000500010.
Pressler, B., 2012. Candidiasis and rhodotorulosis, in: Greene, C.E. (Ed.), Infectious Diseases of the Dog and Cat. 4 ed.. Saunders, p. 666–671.
Procop, G.W., 2016. Koneman's Color Atlas and Textbook of Diagnostic Microbiology (Color Atlas & Textbook of Diagnostic Microbiology). 7 ed., Jones & Bartlett Learning, Philadelphia.
Quesenberry, K.E., Donnelly, T.M., Mans, C., 2012. Biology, husbandry, and clinical techniques of guinea pigs and chinchillas, in: Ferrets, rabbits, and rodents. Elsevier, pp. 279–294. 10.1016/B978-1-4160-6621-7.00022-1.
Quevedo, M., Lescano G., J., Fernández A., V., 2014. *Malassezia* spp. associated dermatitis in a pet rabbit (*Oryctolagus cuniculus*). Revista de Investigaciones Veterinarias del Perú 24. 10.15381/rivep.v24i4.2728.
Radi, Z.A., 2004. Outbreak of sarcoptic mange and malasseziasis in rabbits (*Oryctolagus cuniculus*). Comparative Medicine 54, 434–437. URL: https://www.ncbi.nlm.nih.gov/pubmed/15357325.
Raposo, J.B., Nobre, M.D.O., Fernandes, C., Porto, M., 1996. Cutaneous candidiasis in a dog. Rev. da Fac. Zootec. 2, 11–17. URL: http://files.medvetufmt.webnode.com/200000024-cf3d0d0374/candidiase%20cutanea%20cao.pdf.
Reedy, L.M., Miller, W.H., Willemse, T., 1989. Aeroallergens and aerobiology, in: Allergic Skin Diseases of Dogs and Cats. W B Saunders Co, Philadelphia, p. 50–82.
Roden, M.M., Zaoutis, T.E., Buchanan, W.L., Knudsen, T.A., Sarkisova, T.A., Schaufele, R.L., Sein, M., Sein, T., Chiou, C.C., Chu, J.H., Kontoyiannis, D.P., Walsh, T.J., 2005. Epidemiology and outcome of zygomycosis: A review of 929 reported cases. Clinical Infectious Diseases 41, 634–653. 10.1086/432579.
Rosenthal, S.A., Wapnick, H., 1963. The value of Mackenzie's Hair Brush technic in the isolation of *T. Mentagrophytes* from clinically normal guinea-pigs. Journal of Investigative Dermatology 41, 5–6. 10.1038/jid.1963.61.
Scott, D., 2001. Dermatoses of pet rodents, rabbits, and ferrets, in: Muller & kirk's small animal dermatology. Elsevier, pp. 1415–1458. 10.1016/B978-0-7216-7618-0.50025-0.
Sierra, P., Guillot, J., Jacob, H., Bussiéras, S., Chermette, R., 2000. Fungal flora on cutaneous and mucosal surfaces of cats infected with feline immunodeficiency virus or feline leukemia virus. American Journal of Veterinary Research 61, 158–161. 10.2460/ajvr.2000.61.158.
Simpanya, M.F., Baxter, M., 1996. Isolation of fungi from the pelage of cats and dogs using the hairbrush technique. Mycopathologia 134, 129–133. 10.1007/bf00436719.
Stojanov, I., Jaksic, S., Prodanov, J., 2007. Presence and importance of saprophyte fungal organisms on dog skin. Matica Srpska Journal of Natural Sciences , 261–26510.2298/ZMSPN0713261S.
Turner, P.V., Brash, M.L., Smith, D.A., 2017. Guinea pigs, in: Pathology of small mammal pets. John Wiley & Sons, Inc., Hoboken, NJ, USA, pp. 147–191. 10.1002/9781118969601.ch3.
Vangeel, I., Pasmans, F., Vanrobaeys, M., De Herdt, P., Haesebrouck, F., 2000. Prevalence of dermatophytes in asymptomatic guinea pigs and rabbits. The Veterinary Record 146, 440–441. 10.1136/vr.146.15.440.
Vieira, M.R., Milheiro, A., Pacheco, F.A., 2001. Phaeohyphomycosis due to *Cladosporium cladosporioides*. Medical Mycology 39, 135–137. 10.1080/mmy.39.1.135.137.
White, S.D., Bourdeau, P.J., Meredith, A., 2003. Dermatologic problems in guinea pigs. Compendium on Continuing Education for the Practicing Veterinarian URL: https://ucdavis.pure.elsevier.com/en/publications/dermatologic-problems-in-guinea-pigs.
White, S.D., Guzman, D.S.M., Paul-Murphy, J., Hawkins, M.G., 2016. Skin diseases in companion guinea pigs (*Cavia porcellus*): A retrospective study of 293 cases seen at the Veterinary Medical Teaching Hospital, University of California at Davis (1990-2015). Veterinary Dermatology 27, 395–e100. 10.1111/vde.12348.