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KERATINOPHILIC FUNGI FROM THE FEATHERS OF FREE-LIVING BIRDS

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Abstract. Mycological examinations of feather samples taken from 1 120 birds belonging to 78 species revealed keratinophilic fungi in 170 specimens (15.2 %). Species of the genus *Anixiopsis*, *Arthroderma*, *Chrysosporium*, *Thielavia* and *Trichophyton* were isolated. Dermatophytes known as causative agents of superficial mycoses, namely *Trichophyton mentagrophytes* (*Fulica atra*, *Sylvia curruca* and *Emberiza schoeniclus*) and *Nannizzia persicolor* (*Acrocephalus scirpaceus* and *A. schoenobaenus*) were found in the feathers of five species of birds. The presence of keratinophilic fungi in the feathers of birds was influenced by bird bionomy (contact with soil, breeding biology, synanthropisation etc.). The sporadic occurrence of agents of superficial mycoses found in the feathers confirms the hypothesis that birds play only a limited role in the transport and dispersal of these agents.

The feathers of birds, like the hair of small mammals, represent a suitable substrate for the colonization by keratinophilic fungi. Studies focused on the knowledge of mycoflora of this substrate have gradually revealed that the findings of keratinophilic micromycetes in the feathers are not accidental either from the qualitative or quantitative aspect, but reflect specific relationships between the fungi and their hosts. The English authors (Pugh and Evans 1970, Pugh 1972) pointed out the differences in the frequency of the occurrence of keratinophilic micromycetes in the feathers of different bird species on the basis of long-term studies. They looked for an explanation in the different bionomy of the bird hosts as well as in the differences of microbiotopes represented by the feathered integument of these vertebrates. New studies of the Soviet authors (Kuzmina and Sharapov 1975, Sharapov and Kuzmina 1976) have also directed attention to the importance of specific microclimatic and biotic condition of feather colonization by keratinophilic fungi. The present paper has been inspired by the results obtained in the studies on interactions of micromycetes and birds in Czechoslovakia (Otčenášek et al. 1967, Hubálek 1972, 1974a, b, 1976, Hubálek et al. 1973, Hubálek and Balát 1974) and should be additional contribution to the present knowledge on the influence of ecological factors upon the distribution of microscopic fungi which metabolically use keratin.

MATERIAL AND METHODS

The feather samples were taken from birds captured by current methods (collection from nests, catches in nets etc.). The birds were captured in the period from April to August 1977 in 10 localities in East Bohemia (Bohdaneč, Bukovka, Jílovka, Rohovládová Bělá, the Skříň fishpond, the Strážov fishpond, Sopřeč, the Tichá fishpond, Trhoňka and Vyšehněvice). In the mentioned localities predominant were biotopes typical of the forest and field fishponds with different intensity of reed growths, but also meadow biotopes, forest edges and gardens were represented. The birds investigated numbered 1 120 specimens belonging to 78 species. Each specimen yielded one feather sample originating from three different topographic localizations. Tufts of feather collected from sites on the head, neck and under the wings were taken by pincette disinfected in 1 % Ajatin solution and stored in sterile test tubes. The samples were treated by the so-called hair bait method: the feathers, mixed with sterile soil and particles of children's hair in petri dishes, were moistened by sterile distilled water and incubated in the darkness at 27 °C for 4 weeks. The fungi growing from positive samples

were isolated on Sabouraud dextrose agar (SDA) with chloramphenicole and subsequently identified. So far as the isolated strains corresponded with the descriptions of species in which perfect stages are known but cleistothecia had not been formed in the hair cultures, additional media were used to stimulate their formation: oat flake agar (Hejtmánková and Hejtmánek 1965) and modified de Vroey's medium (Young 1968).

RESULTS

Keratinophilic fungi were found in the feathers of 170 (15.2 %) out of 1 120 birds investigated. Positive findings came from 42 species of these vertebrates. The following survey arranged according to host orders contains information about the particular findings (micromycetes isolated and number of isolations are given in brackets).

Anseriformes: *Anas platyrhynchos* (*Anixiopsis stercoraria*, *Arthroderma quadrifidum*, *Chrysosporium* sp., *Ch. keratinophilum*, *Ch. tropicum*, *Trichophyton terrestre* 12), *Anas strepera* (*Chrysosporium* sp., *Thielavia sepedonium*, *Trichophyton terrestre*), *Anas clypeata* (*Ctenomyces serratus*), *Aythya fuligula* (*Arthroderma insingulare* 2, *Chrysosporium keratinophilum*, *Trichophyton terrestre*), *Aythya nyroca* (*Arthroderma quadrifidum*, *Trichophyton terrestre* 4).

Falconiformes: *Falco tinnunculus* (*Arthroderma quadrifidum*).

Galliformes: *Phasianus colchicus* (*Trichophyton terrestre*).

Ralliformes: *Rallus aquaticus* (*Arthroderma multifidum*), *Fulica atra* (*Arthroderma quadrifidum*, *Chrysosporium tropicum*, *Trichophyton mentagrophytes*).

Charadriiformes: *Actitis hypoleucos* (*Chrysosporium pannorum*).

Columbiformes: *Columba livia* f. *domestica* (*Chrysosporium tropicum*).

Apodiformes: *Apus apus* (*Arthroderma insingulare*, *A. lenticulare*, *A. tuberculatum*, *Chrysosporium* sp.).

Passeriformes: *Sylvia borin* (*Arthroderma multifidum*), *Sylvia communis* (*Chrysosporium* sp.), *Sylvia curruca* (*Arthroderma lenticulare*, *Chrysosporium keratinophilum*, *Trichophyton mentagrophytes*), *Phylloscopus collybita* (*Anixiopsis stercoraria*, *Arthroderma curreyi*, *A. insingulare*, *A. lenticulare*, *A. quadrifidum* 2, *A. tuberculatum* 2, *Chrysosporium keratinophilum*, *Ch. tropicum*), *Phylloscopus trochilus* (*Chrysosporium keratinophilum*), *Locustella luscinioides* (*Arthroderma insingulare*), *Acrocephalus arundinaceus* (*Arthroderma insingulare*, *A. lenticulare*, *Chrysosporium* sp.), *Acrocephalus palustris* (*Arthroderma curreyi*, *Chrysosporium* sp.), *Acrocephalus scirpaceus* (*Arthroderma ciferrii*, *A. curreyi* 3, *A. lenticulare*, *A. multifidum* 2, *A. tuberculatum* 2, *Chrysosporium* sp., *Ch. asperatum*, *Ch. keratinophilum*, *Nannizzia persicolor*, *Trichophyton terrestre*), *Acrocephalus schoenobaenus* (*Arthroderma curreyi* 3, *A. insingulare* 2, *A. lenticulare* 2, *A. multifidum* 6, *A. tuberculatum* 5, *A. quadrifidum*, *Chrysosporium* sp., *Nannizzia persicolor*, *Thielavia sepedonium*), *Turdus merula* (*Arthroderma ciferrii*, *A. curreyi*, *A. quadrifidum* 3, *Chrysosporium evolceanui*, *Trichophyton terrestre*), *Turdus philomelos* (*Chrysosporium keratinophilum*), *Erithacus rubecula* (*Trichophyton terrestre*), *Lanius collurio* (*Arthroderma lenticulare*, *Chrysosporium tropicum*, *Trichophyton terrestre*), *Parus caeruleus* (*Arthroderma curreyi*), *Parus major* (*Arthroderma quadrifidum*), *Remiz pendulinus* (*Arthroderma tuberculatum*, *Chrysosporium tropicum*), *Panurus biarmicus* (*Anixiopsis stercoraria*, *A. curreyi*), *Hirundo rustica* (*Arthroderma insingulare*, *A. tuberculatum* 2), *Riparia riparia* (*Arthroderma insingulare* 2, *Trichophyton terrestre*) *Carduelis carduelis* (*Arthroderma quadrifidum*, *Chrysosporium keratinophilum*), *Carduelis cannabina* (*Arthroderma quadrifidum*, *Chrysosporium asperatum*, *Trichophyton georgiae*), *Carduelis chloris* (*Arthroderma quadrifidum*, *Chrysosporium* sp., *Trichophyton terrestre*), *Serinus serinus* (*Arthroderma insingu-*

Table 1. Frequency of particular keratinophilic fungi species isolated in bird orders examined

Bird order / Fungus species	Podicipediformes n = 3	Anseriformes n = 76	Falconiformes n = 5	Galliformes 1	Ralliformes n = 38	Charadriiformes n = 22	Columbiformes n = 10	Strigiformes n = 5	Cuculiformes 1	Apodiformes n = 33	Coraciiformes 1	Piciformes n = 5	Passeriformes n = 920	Total 1120	% of particular species
Anixiopsis stercoraria		1											2	3	1.7
Arthroderma ciferrii													3	3	1.7
A. curreyi													17	17	9.4
A. insingulare		1								1			16	18	9.9
A. lenticulare										1			17	18	9.9
A. multifidum					1								9	10	5.5
A. quadrifidum		2	1		1								24	28	15.5
A. tuberculatum										1			15	16	8.8
Arthroderma sp.													1	1	0.6
Chrysosporium sp.		2								1			7	10	5.5
Ch. asperatum													5	5	2.8
Ch. evolceanui													1	1	0.6
Ch. keratinophilum			1										7	8	4.4
Ch. pannorum						1							1	2	1.2
Ch. tropicum		1			1		1						5	8	4.4
Ctenomyces serratus		1												1	0.6
Nannizzia persicolor													2	2	1.1
Thielavia sepedonium		1											2	3	1.7
Trichophyton georgii													1	1	0.6
T. mentagrophytes					1								2	3	1.7
T. terrestre		10		1									12	23	12.6
Number of negative birds	3	57	3	—	34	21	9	5	1	29	1	5	772	950	—
Number of keratinophilic fungi isolated	—	19	2	1	4	1	1	—	—	4	—	—	149	181	100.0

lare, *Trichophyton terrestre*), *Emberiza citrinella* (*Arthroderma lenticulare*, *A. tuberculatum*), *Emberiza schoeniclus* (*Arthroderma curreyi*, *Trichophyton mentagrophytes*), *Passer domesticus* (*Arthroderma* sp., *A. curreyi* 5, *A. insingulare* 7, *A. lenticulare* 7, *A. quadrifidum* 12, *A. tuberculatum*, *Chrysosporium asperatum* 3, *Ch. keratinophilum*, *Ch. pannorum*, *Ch. tropicum*, *Thielavia sepedonium*, *Trichophyton terrestre* 5), *Passer montanus* (*Arthroderma ciferrii*, *A. curreyi*, *A. lenticulare*, *A. quadrifidum* 2), *Motacilla flava* (*Arthroderma multifidum*), *Sturnus vulgaris* (*Arthroderma quadrifidum*).

The frequency of isolates of particular keratinophilic fungi species is given in Table 1, where the host range contains 13 groups corresponding with relevant bird orders. *Arthroderma quadrifidum* isolated from feathers of 16.4 % of positive birds was absolutely dominant. A high frequency was also revealed in the two remaining members of the conidial species *Trichophyton terrestre*, *Arthroderma insingulare* and *A. lenticulare* (both found in 10.6 % of birds). The findings of all three components of the *T. terrestre* complex are moreover included in the data that in 13.5 % of positive birds this geophilic dermatophyte was found without being differentiated according to sexual stages (cleistothecia failed to be induced). *Arthroderma curreyi* and *A. tuberculatum* were also among the more abundant keratinophilic fungi species.

Of the dermatophytic fungi, known to be agents of superficial mycoses, *Nannizzia persicolor* (2) and *Trichophyton mentagrophytes* (3) were isolated. However, in no bird investigated any lesions indicating a skin disease were ascertained.

Table 2. Frequency of keratinophilic fungi occurrence in orders and families of birds examined

Order of birds examined	Family	Total examined		% of birds with contaminated feathers	
Podicipediformes			3		—
Anseriformes		76		20.3	
Falconiformes			5		1*
Galliformes			1		1*
Ralliformes		38		10.5	
Charadriiformes		22		4.5	
Columbiformes			10		—
Strigiformes			5		—
Cuculiformes			1		—
Apodiformes		33		12.1	
Coraciiformes			1		—
Piciformes			5		—
Passeriformes		920		15.4	
	Muscicapidae		6		—
	Sylviidae	387		15.1	
	Turdidae	62		12.9	
	Laniidae	17		17.6	
	Paridae	65		6.2	
	Paradoxornithidae		9		2*
	Hirundinidae	54		9.3	
	Sittidae		2		—
	Fringillidae	210		6.7	
	Ploceidae	80		61.3	
	Motacillidae		3		1*
	Sturnidae		12		1*
	Oriolidae		1		—

Digits indicated by * are given in absolute value

Table 3. Keratinophilic fungi occurrence in feathers depending on some aspects of bird ecology

	Migration			Sociability		Contact with soil		Synanthropisation		
	migratory	roving birds	non-migratory	gregarious	non-gregarious birds	+	—	eusynanthropic	hemisynanthropic birds	exoanthropic
Number of birds examined	811	218	91	213	907	1019	101	187	147	786
% of birds with contaminated feathers	24.0	9.6	56.0	28.2	13.2	19.6	14.8	31.0	15.3	13.3

Table 4. Keratinophilic fungi occurrence in feathers depending on feeding habits of birds

	Feeding habit			Feeding place				
	Phytophagous	Zoophagous birds	Polyphagous	Ground (dry soil)	Ground (humid soil)*	Trees and shrubs	Water	Air (on the wing)
Number of birds examined	440	597	83	278	192	515	83	52
% of birds with contaminated feathers	29.1	13.7	7.6	24.1	13.7	14.3	14.5	10.4

* rotten plant remains on stream banks etc.

Table 2 shows a total relative frequency of keratinophilic fungi occurrence without species differentiation. The host range is given again on the level of orders, the order Passeriformes, with the highest number of birds represented being subdivided into families. Among the birds whose feathers are most frequently colonized by keratinophilic micromycetes are primarily Anseriformes (20.3 %), then Passeriformes (15.4 %), Apodiformes (12.1 %) and Ralliformes (10.5 %). In the order Passeriformes the keratinophilic fungi are of high frequency of occurrence, primarily in Ploceidae.

Table 3 suggests the dependence of the keratinophilic fungi occurrence on factors relating to the bionomy of birds. Positive findings predominated in non-migratory birds; the fungi were found twice as often in gregarious birds e.g. in their resting places, in contrast to non-gregarious birds. The frequency of occurrence of keratinophilic fungi was positively influenced by the contact of birds with the soil and by the degree of synanthropisation: positive findings were most frequent in eusynanthropic birds, e.g. in members of the genus *Passer*.

As far as the dependence on feeding habit and on the type of feeding habitat of birds is concerned, the keratinophilic micromycetes were found primarily in the feathers of phytophagous birds and in the feathers of birds seeking food on the ground (Table 4).

Table 5. Keratinophilic fungi occurrence depending on the type of inhabited biotope and nesting place of birds

	Biotope				Nesting places			
	I	II	III	IV	V	VI	VII	VIII
Number of birds examined	408	217	235	260	59	393	500	168
% of birds with contaminated feathers	17.4	16.1	9.6	19.9	20.3	18.8	14.2	29.2

I = towns and villages
II = fields and meadows
III = forest
IV = water and reeds
V = cavities
VI = nests on the ground
VII = nests in trees
VIII = nests in buildings and rocks

Table 6. Keratinophilic fungi occurrence depending on sex of birds and degree of maturity

	Adult		Immature	
	birds			
	♂	♀	fledgelings	ready for flight
Number of birds examined	196	209	217	498
% of birds with contaminated feathers	23.5	11.6	10.1	20.5

Conversely, the least contaminated feathers were detected in bird species feeding on the wing. Marked was also the frequency of findings influenced by the type of biotopes in which the birds lived (Table 5). Colonized by fungi were primarily feathers of aquatic birds and feathers of cavity nesting birds, living in rocks, and in human dwellings. Conversely, the least number of findings came from birds inhabiting forest biotopes. Sex was another factor influencing the percentage of feathers contaminated by keratinophilic micromycetes: it was twice as high in females as in males (Table 6). The table also shows that in immature birds keratinophilic fungi were more frequently found in the feathers of fledglings ready for flight.

DISCUSSION

Keratinophilic mycoflora in the feathers is considerably influenced by the occurrence of these fungi in the soil to be found in the territory inhabited by particular bird species. Though we cannot rely on the close ecologic association with soil substrates such as in the case of small terrestrial mammals (e.g. rodents and insectivores), to a certain extent the colonization of bird feathers by keratinophilic fungi is a reflection both of the qualitative and quantitative representation of micromycetes occurring in the soil of the biotope inhabited by birds. This dependence is also evident from the results of our investigations, which demonstrate a more frequent occurrence of keratinophilic fungi in birds active on the ground (search for food, collection of materials necessary for nest building etc.). The influence of active contact with soil and the influence of cavity nesting upon the higher frequency of keratinophilic fungi was also proved by Hubálek (1974b). The results of this author and our findings show that colonization of feathers by fungi is also promoted by suitable microclimatic conditions during the bird's stay in the nest; primarily nesting in cavities and in the nests built in human dwellings or in the immediate vicinity (stables, barns) promote the frequency of keratinophilic fungi occurrence in feathers. The influence of synathropisation phenomenon was disclosed in previous papers (Hubálek 1972). Our results show that this phenomenon influences not only the quantitative representation of particular fungus species, but also the scope of the species spectrum of these organisms.

The differences in the frequency of keratinophilic fungi in the feathers of various bird orders may be also determined by factors unrelated to environment in which the birds live, but concerning the birds themselves. After some authors e.g. the fat in feathers is an important factor which may stimulate or inhibit the growth of keratinophilic micromycetes (Pugh and Evans 1970, Pugh 1972). A stimulating influence might be anticipated in the high frequency of keratinophilic fungi found in the feathers of aquatic birds (Anseriformes).

The rare occurrence of pathogenic agents of dermatophytoses in the feathers of free-living birds, as confirmed by our results, is in harmony with the published data on the sporadic occurrence of skin mycoses in these vertebrates (Soper and Hosking 1961, Pepin and Austwick 1968). The high temperature on the skin surface of birds, 40 to 41 °C (Pugh 1972), is obviously unfavourable to the cutaneous propagation of the agent, which mostly belongs to typical mesophilic organisms. In the feathers of birds examined even typical ornitophilic species of dermatophytes — *Trichophyton gallinae* and *T. simii* — failed to be demonstrated, despite their higher occurrence in birds than in other natural substrates (Dvořák and Otčenášek 1969). Although the frequency of occurrence of agents causing dermatophytoses in the birds' feathers is very low, important findings of *Microsporum persicolor* and *Trichophyton mentagrophytes* in migratory birds testified that wild birds may transport pathogenic dermato-

phytes over great distances. However, in contrast to small terrestrial mammals, birds evidently play a limited role as a source of infection for man and food-producing animals.

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КЕРАТИНОФИЛЬНЫЕ ГРИБЫ ИЗ ПЕРЬЕВ СВОБОДНОЖИВУЩИХ ПТИЦ

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Резюме. Путем микологического обследования проб перьев, взятых от 1120 птиц, относящихся к 78 видам, обнаружены кератинофильные грибы у 170 особей (15,2 %). Выделены виды родов *Anixiopsis*, *Arthroderma*, *Chrysosporium*, *Thielavia* и *Trichophyton*. В перьях пяти видов птиц обнаружены дерматофиты известные как возбудители суперфициальных микозов: *Trichophyton mentagrophytes* (*Fulica atra*, *Sylvia curruca* и *Emberiza schoeniclus*) и *Nannizzia persicolor* (*Acrocephalus scirpaceus* и *A. schoenobaenus*). На наличие кератинофильных грибов в перьях птиц оказала влияние биономия птиц (контакт с почвой, способ гнездования, синантропизация и т. д.). Единичные находки возбудителей поверхностных микозов в перьях птиц подтверждают предположение, что птицы играют ограниченную роль в транспорте и дисперсии этих возбудителей.

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