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Bird feather fungi from Svalbard Arctic

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Abstract Despite feather fungi being an important component of the Arctic fungal flora, their ecological role and diversity are not fully known. In the current study, fungal cultures were isolated from feathers (barnacle goose, common eider, and glaucous gull) collected in the Ny-Ålesund region, Svalbard. Isolates were identified by ITS region sequences, which include the ITS1, ITS2, and 5.8S rRNA. The result showed culturable yeast and filamentous fungi belonging to three classes: Ascomycota (*Pyrenochaetopsis pratorum, Cladosporium herbarum, Thelebolus microsporus*, *Aspergillus versicolor, Penicillium commune,* and *Venturia* sp.), Basidiomycota (*Mrakia blollopis* and *Rhodotorula mucilaginosa*), and Zygomycota (*Mucor flavus*). Most of the fungal isolates appeared to be cold-tolerant, and about 60 % of the isolates showed keratinase activity. The reasonably low fungal diversity colonizing feathers indicates that the birds of Svalbard are casual carriers of fungi which may result in a negligible impact on their health. To the best of our knowledge, this is the first record of fungal communities present on the feathers of birds in the high Arctic.

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Introduction

Fungi have enormous ecological importance due to wide morphological diversity, life strategies, and also the ability to interact with various biotic and abiotic components of the environment (Hawksworth 1991 ; Peay et al. 2008). Polar fungi have the ability to grow in oligotrophic cold environments (Connell et al. 2008) and to produce cold-active enzymes (Buzzini et al. 2012), but their ecological role is poorly understood (Buzzini et al. 2012; Dynowska et al. 2013).

Mycological exploration in Svalbard began with the study of Karsten (1872). Elvebakk et al. (1996) taxonomically listed 389 species of fungi belonging to Myxomycota, Oomycota, Chytridiomycota, Zygomycota, Ascomycota, Deuteromycota, and Basidiomycota. From moss colonies of Svalbard, Hoshino et al. (1999) isolated *Pythium ultimum* Throw var. *ultimum*. From Norway and Svalbard, Aarnæs (2002) prepared a catalogue on "macro- and micromycetes". Kurek et al. (2007) characterized soil filamentous fungi from Bellsund region of Spitsbergen. Pang et al. (2008, 2009) had delineated two novel fungal species from marine habitat of Svalbard. Singh et al. (2012) reported 19 species under 14 genera (*Acremonium*, *Arthrinium*, *Aspergillus*, *Cladosporium*, *Corynespora*, *Emericella*, *Geomyces*, *Mortierella*, *Mucor*, *Myrothecium*, *Penicillium*, *Phialophora*, *Preussia*, and *Xylaria*) from soils of Ny-Ålesund. Recently, pathogenic yeasts have also been reported from the throat and cloaca of the little auk (*Alle alle*), an Arctic colonial seabird (Dynowska et al. 2013). However, the composition of fungal flora in Arctic bird's feathers has not been investigated.

In cold environments, fungi have been isolated from various substrates and habitats such as permafrost (Ivarson 1965; Ozerskaya et al. 2009); glacial ice (Reeve et al. 2002; Butinar et al. 2007); cryoconite holes (Säwström et al. 2002); soil (Callaghan et al. 2004); puddles (Pathan et al. 2009); and Arctic coastal environments (Butinar et al. 2011). Fungi colonize either as normal harmless biota on animals and humans (Barnett et al. 2000) or as pathogens (Ghannuaum and Abu-Elteen 1990; Dynowska and Kisicka 2005a). However, a few fungal species (*Aspergillus fumigates*, *A. flavus*, *Candida albicans*, *C. tropicalis*, *C. glabrata,* etc.) are harmless to some animal species but may be pathogenic to others (Dynowska and Kisicka 2005a, b; Tsiodras et al. 2008). Arctic birds have been considered an excellent host and/or vector of fungi (Dynowska et al. 2013) and bacteria (Sjolund et al. 2008). In addition to the Arctic, birds from various other areas have also been reported as fungal carriers (Dupont et al. 1994; Tsiodras et al. 2008). Knowledge of fungi growing on birds from the Arctic (Wojczulanis-Jakubas et al. 2011; Dynowska et al. 2013) and Antarctic (Del Frate and Caretta 1990; Singh et al. 2014) region is very limited. The presence of fungi on the feathers of common, clinically healthy birds from tropical, subtropical, and other than polar regions (Buck 1983; Buck and Chabasse 1998; Mancianti et al. 2002; Czeczuga et al. 2004; Dynowska and Kisicka 2005a, b; Cafarchia et al. 2006; Kutty and Philip 2008; Mandeel et al. 2011; Miljković et al. 2011; Gungnani et al. 2012) has also been documented. The migration and its epidemiological significance of pathogenic fungi among different habitats with respect to wild birds have been reported (Tsiodras et al. 2008).

Due to the impact of climate change, the Arctic has experienced a twofold higher increase in temperature compared to most parts of world (IPCC 2007). As a result, there is the possibility of an emergence of different fungi and an increase in the prevalence of potentially pathogenic fungi colonizing birds (Dynowska and Kisicka 2005a). Moreover, due to increasing temperatures, the dormant mesophilic fungal spores (including bird's pathogens) which are carried by birds, air, and anthropogenic activities may emerge and colonize areas of Arctic. Therefore, there is a need to examine more birds from the Arctic regions.

Some birds such as barnacle goose (*Branta leucopsis*), common eider (*Somateria mollissima*), and glaucous gull (*Larus hyperboreus*) were sampled in Svalbard during the Arctic summer. The barnacle goose builds their nests on small islands and mountain cliffs, while the common eider builds its nest close to the coasts. Both the barnacle goose and the common eider choose to feed in the wetland area. The glaucous gull builds nests on the ground or on cliffs and feeds in Kongsfjorden coasts and on the wetlands. These birds breed in Svalbard during the Arctic summer and are considered to be an important component of the Svalbard terrestrial ecosystem.

Fungi have various nutritional modes and ecological behaviours (saprotrophy, necrotrophy, and biotrophy); however, numerous fungi do not restrict themselves to a single mode but exhibit varying degrees of flexibility in response to changes in their environment through mechanism of heterokaryosis and differential gene expression (Cooke and Whipps 1993). Keratinophilic fungi are considered to be natural colonizer of partially degraded keratin substrate which are used as a source of carbon, nitrogen, and sulphur (Griffin 1960; De Vries 1962). However, keratinolytic fungi are only those that are truly capable of attacking and demolishing keratin (Majchrowicz and Dominik 1969; Dominik et al. 1973; Filipello Marchisio 2000). The specialized fungal organs (hyphae) and enzymes (keratinases) together are accountable for keratinolytic activity (Kunert 1972; Böckle et al. 1995; Filipello Marchisio 2000). It has been reported that, like many other fungal biochemical activities, keratinolysis does not have a species-specific character (Filipello Marchisio 1986; Filipello Marchisio et al. 1991, 1994). An individual species within the same environmental conditions has both active and non-active strains, and each strain/isolate showed variations in the manner and intensity in which they attack the keratin substrate and differentiate specialized structures, therefore termed as a "potentially keratinolytic species" (Filipello Marchisio 2000).

The current study was planned to evaluate the species composition and prevalence of filamentous fungi and yeasts on feathers of birds in Svalbard. Furthermore, we have investigated keratinophilic/keratinolytic activity of feather fungi. Moreover, we have also examined the biosafety level of the fungal species.

Materials and methods

Study site and sampling methods

Spitsbergen is the largest island of the Svalbard archipelago. The mean air temperature is −14 °C in the coldest month (February) and +5 °C in the warmest month (July) (Nygaard 2009). In the present study, bird's feathers were sampled from their breeding places near the Kongsfjorden coast as well as the coast of an island inside the fjord and mountain cliffs around Ny-Ålesund (78°55′N, 11°56′E) in Svalbard (Fig. 1). Without harming birds, three feather samples were collected from five adult individuals of each bird species (barnacle goose, common eider, and glaucous gull) using sterile gloves, forceps, and a sample collector (HiMedia Laboratories) from 6 to 7 July 2012. The samples were stored at −20 °C for one day and transported, with ice packs, within 24 h to the laboratory. The bird's feathers vane was analysed for culture-based studies.

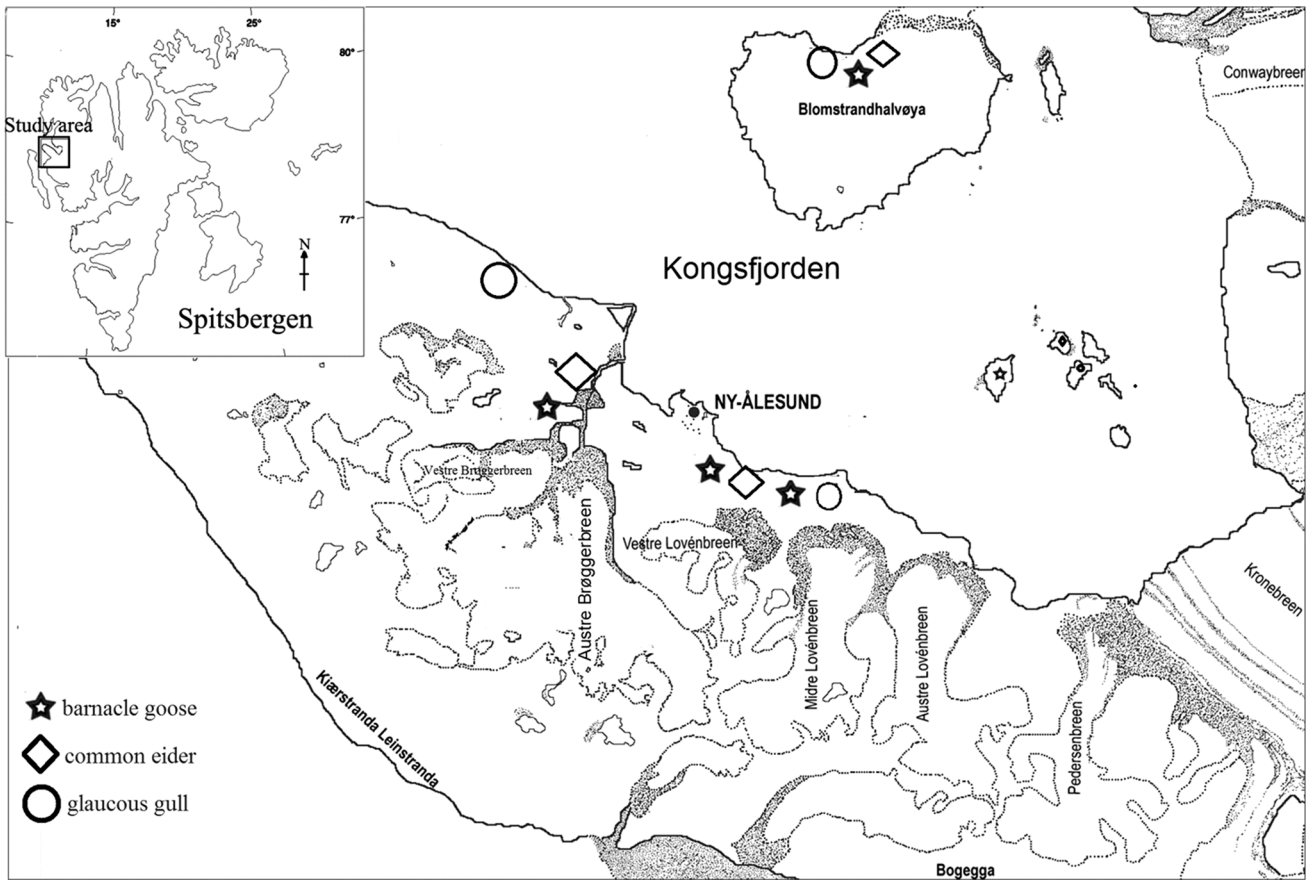

Fig. 1 Map of Svalbard showing sample locations of feathers of glaucous gull (*Larus hyperboreus*), common eider (*Somateria mollissima*), and barnacle goose (*Branta leucopsis*)

Isolation of fungi and their characteristics

The collected feathers were inoculated onto a Sabouraud dextrose agar (SDA) artificial mycological medium. This medium was composed of a mixture of peptic digest of animal tissue and pancreatic digest of casein (1:1) 10 g/l, dextrose 40 g/l, and agar 15 g/l with pH, after sterilization, of 5.6 ± 0.2. To get psychrophilic fungal isolates for screening of keratinase activity and to avoid the growth of mesophilic fungi, the plates were incubated in 4 and 15 °C for 3 weeks. Cultures plates were monitored regularly on the basis of shape, colour, and different morphological features (hyphae, conidiophore, and conidial structure). The distinct colony was picked up, subcultured, and observed for purity of cultures under a microscope. Pure cultures were further incubated at 22, 25, and 30 °C to see whether it had the ability to grow at higher temperatures.

The purified fungal colonies were transferred onto the SDA slants medium solidified in a test tube at about a 35° slant to provide more surface area for fungal growth for detailed study. For morphotaxonomical studies, the fungal mounts were prepared on slides using lactophenol-cotton blue as a mounting medium and observed under Olympus BX-51 and IX-71 model microscopes. Fungal cultures were initially identified on the basis of morphotaxonomy with the help of standard literature (Rapper and Fennell 1965; Ellis 1971, 1976; Barron 1977; Pitt 1979; Schneider 1979; Carmichael et al. 1980; Domsch et al. 1980; Samson and Frisvad 2004; De Hoog et al. 2005; Kirk et al. 2008; de Gruyter et al. 2010; Kurtzman et al. 2011). The isolates with similar morphological characteristics were grouped together, and the representative isolates were subjected to DNA sequence analysis of ITS region. All identified pure cultures were maintained on SDA slants and deposited at the National Fungal Culture Collection of India (NFCCI-WDCM 932) in Pune, India.

Molecular characterization: polymerase chain reaction (PCR), sequencing, and phylogenetic analysis

Total DNA was extracted from cultures (grown on SDA for 3 weeks at 4 °C) using the ISOPLANT II kit (Wako Pure Chemical Industries Ltd., Japan). Extracted DNA was

amplified by PCR method using KOD-plus DNA polymerase (Toyobo Co. Ltd., Japan). The ITS region was amplified using following primers: ITS1F (5′-GTA ACA AGG TTT CCG T) and ITS4 (5́-TCC TCC GCT TAT TGA TAT GC). The amplified DNA was purified using a Wizard® SV Gel and PCR Clean-Up System (Promega KK, Tokyo, Japan). And finally, the purified DNA was sequenced at Macrogen Japan (Tokyo, Japan).

The sequences of isolates (DDBJ AB916505 to AB916517) were deposited in the DNA data bank and were subjected to a NCBI BLAST search. Sequence alignment of ITS region isolates, together with the homologous sequences (retrieved from GenBank) of closely related species, was performed using Clustal W option of MEGA software version 6.05 (Tamura et al. 2013).

To calculate the sequence divergence, the matrix was analysed with the neighbour-joining method (Saitou and Nei 1987) using the Tamura–Nei model (Tamura and Nei 1993) and maximum parsimony method (Tamura et al. 2011). To represent the evolutionary history of the taxa, the bootstrap consensus tree was inferred from 1000 replicates (Felsenstein 1985). The pairwise alignment was performed using EMBOSS Matcher—Pairwise Sequence Alignment tool (http://www.ebi.ac.uk/Tools/psa/emboss_matcher/nucleotide.html).

Screening for keratinase activity

Screening tests on agar plates, using keratin substrate (HiMedia), were performed using the method described by Wawrzkiewicz et al. (1991). The activity of fungus was detected as a clear zone around the colony after incubation of 10 days at a temperature of 4 and 15 °C. The diameter of the clear zone was measured to quantify the enzyme activity.

Biosafety classification

Classification of isolated feather fungi based on biosafety level was done by comparing the species with the database present on www.cbs.knaw.nl and using standard literature (Hoog 1996).

Results

Out of 15 feathers of barnacle goose, 31 isolates were purified from 10 feathers, while the remaining 5 showed no fungal growth on the SDA plates. Likewise, 7 out of 15 feathers of common eider yielded 15 isolates, while the remaining 8 did not give rise to fungal colonies on the plate. Similarly, out of 15 glaucous gull feathers, only 4 of the feathers gave rise to 5 isolates on the SDA plates, while the remaining 11 feathers appeared to be sterile. The total 51 isolates obtained were then classified into 9 groups representing six species of filamentous fungi and three species of yeasts (Table 1). On the basis of ITS region sequence data, the isolates of yeasts and filamentous fungi belonged to six genera (*Pyrenochaetopsis*, *Cladosporium*, *Thelebolus*, *Aspergillus*, *Penicillium*, and *Venturia*) in class Ascomycota, two genera (*Mrakia*, *Rhodotorula)* in Basidiomycota, and one (*Mucor*) in Zygomycota. The seven species of feather fungi (*Pyrenochaetopsis pratorum, Thelebolus microsporus*, *Aspergillus versicolor, Venturia* sp., *Mrakia blollopis, Rhodotorula mucilaginosa*, and *Mucor flavus*) were isolated from barnacle goose, two species (*Cladosporium herbarum* and *Penicillium commune*) were isolated from common eider, and one species *Mucor flavus* was isolated from glaucous gulls (Table 1).

The total sequence lengths after alignment, % sequence similarities, number of positions with base changes, and the NCBI sequence deposition numbers are given in Table 1. The sequence analysis of the 18S rRNA domain of isolate *Pyrenochaetopsis* sp. PG293 (AB916515) indicated their closest relationship with species of *Pyrenochaetopsis pratorum* CBS 445.81 (JF740263), *Cladosporium* sp. PG246 (AB916505) resembled with *Cladosporium herbarum* CBS 399.80 (AJ244227). *Thelebolus* sp. PG278 (AB916508) resembled *Thelebolus microsporus* BI 15-1-1 (GU004196). The results for *Aspergillus* sp. PG277 (AB916513) indicated that its closest relationship is with *Aspergillus versicolor* RF6 (GU232767). The sequence analysis results of isolate *Penicillium* sp. PG291 (AB916511) resembled those of *Penicillium commune* H09-122 (KC009831), and *Venturia* sp. PG255 (AB916509) indicated their closest relationship with species of *Venturia polygoni-vivipari* CBS 114207 (EU035466) by 97.6 % gene similarity and is a novel species yet to be established. *Mrakia* sp. PG265 (AB916506) resembled *Mrakia blollopis* CBS8909 (AY038828). *Rhodotorula* sp. PG294 (AB916512) results indicated their closest relationship with *Rhodotorula mucilaginosa* UOA/HCPF 10538 (HQ702343). *Mucor* sp. PG272 (AB916507) resembled *Mucor flavus* CBS992.68 (JN206067). Phylogenetic trees of fungi of the present study belonging to Ascomycota, Basidiomycota, and Zygomycota are shown in Fig. 2a, b, c.

The feathers, incubated on the SDA medium at 4 and 15 °C for 3 weeks, were shown to be similar types of isolates at both temperatures, but the size of the colony is twofold smaller at 4 °C than at 15 °C. Most of isolates were able to grow up to 25 °C, but only 3 isolates of *Penicillium* and 2 *Aspergillus* isolates were able to grow at 30 °C. These results indicate that some bird feather fungi are psychrotolerant in nature.

Results of keratinase screening revealed that out of 51 isolates, thirty were keratinase positive. The isolates of

Table 1 Identification of fungal isolates (strains) by ITS region sequences, covering ITS1, ITS2, and 5.8S rRNA sequences similarity (%), and keratinase activity

Number of isolates	Identification	Sequence deposition no.	Total sequence length	No. of base changes	Bootstrap support %	18S rRNA gene sequences similarity (%)	Host of the isolates	Keratinase activity
6	*Pyrenochaetopsis* sp. PG293	AB916515	532	0	100	*Pyrenochaetopsis pratorum CBS 445.81 (JF740263)* by 100 %	Barnacle goose	S++
7	*Cladosporium* sp. PG246	AB916505	557	0	100	*Cladosporium herbarum* CBS 399.80 (AJ244227) by 100 %	Common eider	M+
5	*Thelebolus* sp. PG278*	AB916508	566	4	100	*Thelebolus microsporus* CBS 109799 (AY957552) by 99.2 %	Barnacle goose	S++
						Thelebolus microsporus BI 15-1-1 (GU004196) by 100 %		
7	*Aspergillus* sp. PG277	AB916513	571	0	97	*Aspergillus versicolor* RF6 (GU232767) by 100 %	Barnacle goose	–
8	*Penicillium* sp. PG291	AB916511	583	0	88	*Penicillium commune* H09-122 (KC009831) by 100 %	Common eider	M+
	Penicillium sp. PG290	AB916514	592	0	70	*Penicillium commune* H09-122 (KC009831) by 100 %	Common eider	–
4	*Venturia* sp. PG255	AB916509	553	13	60	*Venturia polygoni-vivipari* CBS 114207 (EU035466) by 97.6 %	Barnacle goose	–
5	*Mrakia* sp. PG256*	AB916516	613	2	53	*Mrakia blollopis* CBS8921^{T} (AY038826) by 99.7 %	Barnacle goose	W
	Mrakia sp. PG274*	AB916517	613	4	29	*Mrakia blollopis* CBS8910 (AY038827) by 99.4 %	Barnacle goose	W
	Mrakia sp. PG265*	AB916506	620	0	100	*Mrakia blollopis* CBS8909 (AY038828) by 100 %	Barnacle goose	W
4	*Rhodotorula* sp. PG294*	AB916512	623	1	85	*Rhodotorula mucilagin* CBS 316^{T} (AF444541) by 99.7 %,	Barnacle goose	–
						Rhodotorula mucilagin UOA/HCPF 10538 (HQ702343) by 99.8 %,		
5	*Mucor* sp. PG272	AB916507	616	5	84	*Mucor flavus* CBS 992.68 (JN206067) by 99.2 %	Glaucous gull	–
	Mucor sp. PG268	AB916510	608	7	100	*Mucor flavus* CBS 992.68 (JN206067) by 98.8 %	Barnacle goose	–

* Represents 'yeast', keratinase activity was measured by observing the size of halozones: S++ represents strong positive, M+ represents moderate positive, W represents week positive, and – represents no activity/negative. [Halozone size (1 mm–1.5 cm) = +, (1.6–2 cm & above) = ++, (lesser than 1 mm) = w]

Pyrenochaetopsis sp. PG293, *Cladosporium* sp. PG246, *Thelebolus* sp. PG278, *Penicillium* sp. PG291, and *Mrakia* sp. PG256 proved to have keratinase activity in general (Table 1). However, the strains of genus *Pyrenochaetopsis* and *Thelebolus* had showed a strong keratinase activity, while strains of genus *Mrakia* had shown a very week keratinase activity. Among the 8 isolates of genus *Penicillium*, 5 have shown moderate activity, while 3 have showed no activity.

All the feather fungal isolates were found to be biosafety level BS-1.

Discussion

Due to oligotrophic condition, the colonization and diversity of fungi are very poor in Arctic environment. There is variation in pattern of distribution of fungal strains in the three birds studied. Occurrence of yeast and filamentous fungi on feathers is less frequent and is observed only on a few samples of the studied birds (barnacle goose, common eider, and glaucous gull) in the area of Kongsfjorden. Likewise, low-frequency occurrence of the fungi has also been observed in throat and cloaca of the little auk in

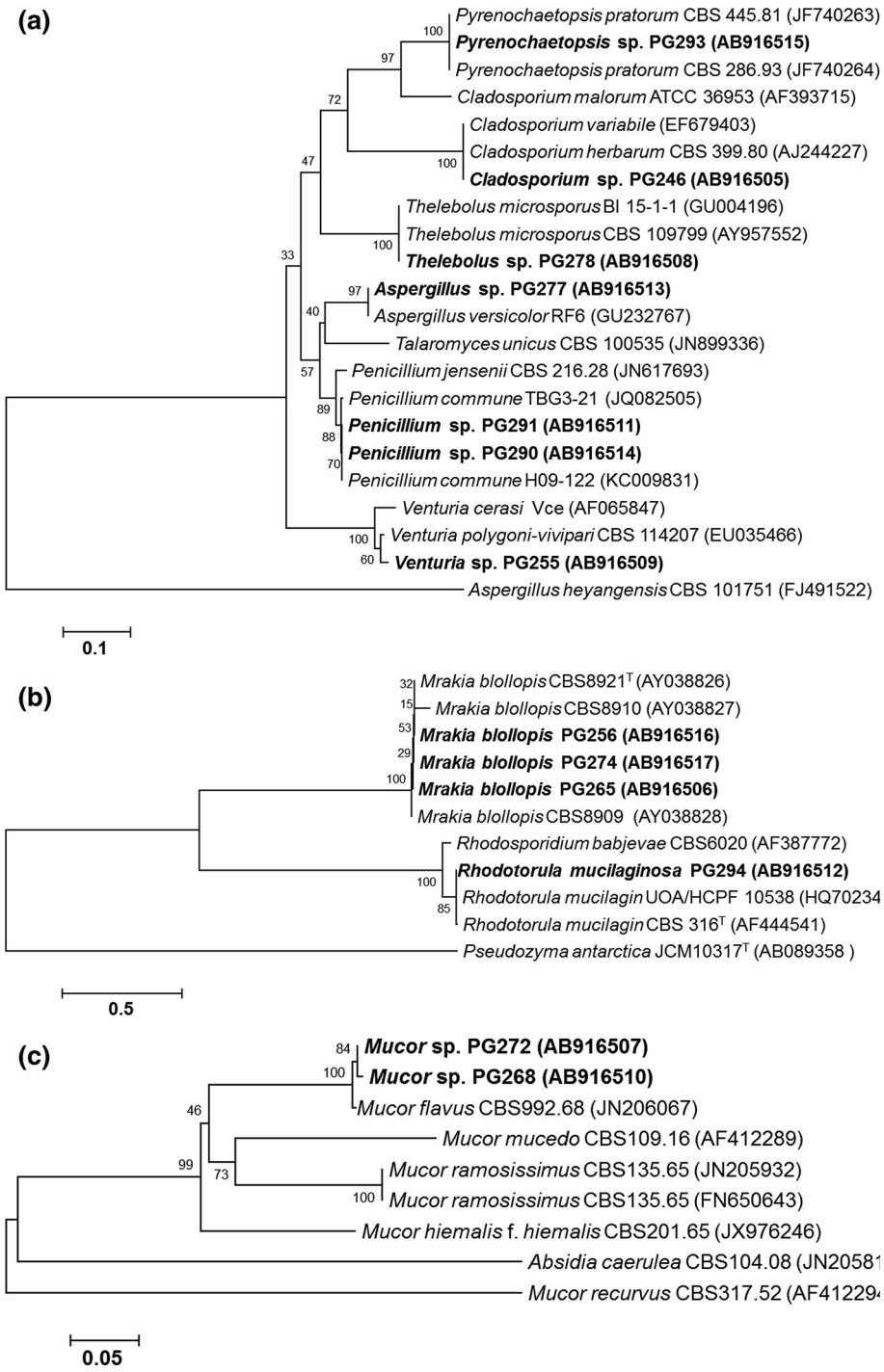

Fig. 2 **a** Phylogenetic tree of Ascomycetous fungal strains (isolates) with closely related species based on ITS region sequences analyses. The accession numbers of strains are shown in parentheses. The tree was constructed with neighbour-joined method. The significance of each branch is indicated by a bootstrap value. The *scale bar* is estimated substitutions per nucleotide position. **b** Phylogenetic tree of Basidiomycetous fungi from bird feathers. The accession numbers of strains are shown in parentheses. Tree was constructed with neighbour-joined method. **c** Phylogenetic tree of Zygomycetous fungi from bird feathers. The accession numbers of strains are shown in parentheses. Tree was constructed with neighbour-joined method

region of Svalbard. Furthermore, the prevalence of the yeast in little auk was reported to be very low (Dynowska et al. 2013). Incidence of yeast has also been reported in other seabirds (Kutty and Philip 2008). *Thelebolus* sp. has also been reported in natural association with feathers and sculls of skuas, petrels, and other birds (Singh et al. 2014; Tsuji et al. 2013a). De Hoog et al. (2005) suggested a distribution of this fungal strain through bird vectors, in various habitats of Antarctica.

Fungi *Cladosporium, Penicillium*, and *Aspergillus* were found during the current study on the feathers of Kongsfjorden, and this has also been reported to be present in the Arctic soils (Holding 1981; Bergero et al. 1999; Kurek et al. 2007; Singh et al. 2012). Most of the feather fungi

isolated were capable of growing between 4 and 25 °C, and a few up to 30 °C, indicating that some of the bird feather fungi are psychrotolerant in nature. Since the birds are migratory in nature, feather colonizing fungi may have survived in a range of temperatures at different geographical locations inhabited by the birds (breeding place to wintering places). This psychrotolerant nature of fungi has also been reported in Arctic soil by Bergero et al. (1999), Kytöviita (2005), and Singh et al. (2012). The physical adaptations that help the fungi overcome low temperature and water stress include formation of chlamydospores and mycelial thickening (Robinson 2001). Similar adaptive features have also been observed in the present study in Svalbard.

The fungal species recorded in current study is different from the species reported present in internal organs (throat and cloaca of little auk). Colonization of fungi in birds depends on many factors such as feeding habit, nature of habitat (feather, throat, and cloaca) and temperature difference on the surface (feathers), and the internal organs (throat and cloaca). The current study in healthy birds shows a low incidence of fungi on feathers when compared to other non-polar birds. Moreover, low frequency of occurrence of yeasts on Arctic bird with minor impact on health has also been reported (Dynowska et al. 2013) and Dynowska and Kisicka (2005a, b) taking into consideration that the birds are random carriers of the fungi. The species composition of fungi examined during the current study belongs to nine genera (Table 1). From the current study, nine species of fungi would be a good indicator of fungal load on feathers. Likewise, Dynowska et al. (2013) also reported 12 species of yeast belonging to eight genera in auk birds. It has been assumed that the reported yeasts from birds are likely to be present in the Arctic habitat (Dynowska et al. 2013).

The occurrence of fungi in feathers of birds could arise due to the possibility of change in nutritional mode allowing them to hydrolysed keratin for their nutrition. Cooke and Whipps (1993) have reported on the nutritional alternative imposed on a fungus by its environment and the narrowing of specialism within a single mode. Keratins are insoluble fibrous proteins, and in nature, there exist as α-keratins and β-keratins (Lehninger 1984). The α-keratins is an insoluble fibrous protein, due to tight packing of their polypeptide chains in α-helix structure and their linkages by disulphide bridges, and hence rendering them poorly biodegradable (Filipello Marchisio 2000). During the current study, the isolates of *Cladosporium, Penicillium*, and *Aspergillus* from feathers were also examined for keratinase activity. This activity is moderately spread among feather fungi. Thus, it can be concluded that keratinase activity is widespread among bird feather fungi and it does not follow that all isolates of the same species will represent similar activity. Keratinolytic fungi such as *Cladosporium, Penicillium*, and *Aspergillus* have also been reported from areas other than polar region (Friedrich et al. 1999). Fungi are able to degrade the keratinic substrates in nature mainly by a biochemical process (keratinases) and morphological structures (hyphae) (Takiuchi et al. 1984; Yu et al. 1969; Malviya et al. 1992; Filipello Marchisio 2000). Therefore, in natural environments, keratinolytic fungi are involved in recycling carbon, nitrogen, and sulphur (Filipello Marchisio 2000). The ecological role of fungi in decomposing complex keratin polymer is important for bioconversion and the nutrient cycle (Friedrich et al. 1999; Peay et al. 2008).

The role of migratory birds entering the Svalbard region for breeding purposes acts as a vector for colonization of fungi in new habitats (glacier) in the Arctic region. *Mrakia* sp. and *Rhodotorula mucilaginosa* have been recorded from Svalbard glaciers (Singh and Singh 2012; Singh et al. 2013), and the occurrence of these two species of yeast on glaciers suggests a possible transmittance of fungi by birds. *Mrakia blollopis* (Thomas-Hall et al. 2010) is a bipolar species and has the unique ability of fermentating various sugars at a low-temperature condition (Tsuji et al. 2013b). Dynowska and Kisicka (2005a, b) opined that the fungi proliferating in marine and terrestrial habitats may passively colonize the avian host through various ways (during nesting incubation, chick rearing, and resting or foraging in water), but the birds need to spend a considerable amount of time in the environment to allow fungal colonization and acclimatization (Dynowska et al. 2013). Over the period of time, these fungi may acclimatize and become harmful to the birds.

To understand the potential hazard faced by scientists (e.g. ornithologists), laboratory personnel, and the environment, the fungal species were classified in accordance with their biosafety levels (Hoog 1996; www.cbs.knaw.nl). Bird feather fungi recorded in the current study belong to biosafety level BS-1, which is suitable for handling and has minimal potential hazard to laboratory personnel and the environment. Dynowska et al. (2013) reported potentially pathogenic yeasts belonging to BSL-1 and BSL-2 category which were isolated from throat and cloaca of Svalbard birds (little auk).

The prevalence and distribution of keratinolytic fungi largely depend on the amount of keratinic material present in the environment (Filipello Marchisio 1986; Ulfig and Ulfig 1990; Filipello Marchisio et al. 1991). The ecological conditions such as pH, temperature, or altitude reported to be less important because these fungi show a wide tolerance towards them (Böhme and Ziegler 1969; Piontelli and Caretta 1974; McAleer 1980; Ogbonna and Pugh 1987). Pugh and Evans (1970) suggested that feather fats play a large part in determining the occurrence of keratinophilic

fungi in birds. The colonization of fungus on feathers in the Arctic may possibly be due to their preference for superficial feather fats. To arrive at such a conclusion, the composition of feather fatty acids needs to be examined. Furthermore, detailed studies on feather fungi in different areas of Arctic may provide an opportunity to learn the details of feather fungal flora and health condition of the birds. Similar such studies will help to establish precautionary measures when scientists handle birds for research purposes.

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