Molds Isolated from Pet Dogs

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Pet dogs have been considered to be involved in the contamination of indoor air by serving as a source of providing molds at houses. Currently, information on the molds originated from pet dogs is rarely available in Korea. The present study was carried out to obtain basic information on the fungi present on pet dogs. For this, fungal isolation was performed to the skin and hairs of 70 pet dogs at different houses and veterinary hospitals. A total of 44 fungal isolates were obtained from skin (27 isolates) and hairs (17 isolates) of the dogs investigated. Based on the observation of microstructures and colony morphology, and the ITS rDNA sequence analysis, the fungal isolates were identified at the level of genus. The identified isolates belong to the genera of *Alternaria*, *Aspergillus*, *Beauveria*, *Chrysosporium*, *Cladosporium*, *Penicillium*, *Scopulariopsis*, and *Trichoderma*. Among these genera, *Aspergillus* (25%), *Cladosporium* (23%) and *Penicillium* (20.5%) were 3 major genera. 63% of the 44 isolates showed color changes on dermatophyte test medium (DTM). When we tested the growth ability of 44 isolates at 37°C, 45% of the isolates were able to grow. These results show that pet dogs could carry fungi having a potentiality of affecting on human health.

KEYWORDS: *Alternaria*, *Aspergillus*, *Cladosporium*, DTM, Indoor molds, Pet dogs

Molds are fungi that commonly found in indoor and outdoor environments. They are important from the point of contamination to foodstuffs, deterioration of diverse natural materials, and deteriorious effect to human health. Regarding human health, the organisms can cause not only infection to human body such as lung and skin but also symptoms such as nasal stuffiness, eye irritation, wheezing, or skin irritation to some peoples (www.cdc.gov/health/mold). Exposed to molds may also cause more severe reactions to those people with serious allergies that from time to time accompany fever and shortness of breath. Recently, with the increase of people's concern on indoor environment, Korean Government initiated making policy and laws that defines the regulation and control of indoor air quality in buildings (Korean Society for Atmosphere Environment, 2004). However, in current law regarding indoor air quality there is no article that regulates mold problems. In addition, information on molds in air, especially, indoor environment is very rare in Korea. Consequently, it is not known yet that how many species of fungi exist in buildings including homes, offices, and public facilities.

A characteristic feature of modern Korean society is that family system becomes a nuclear family and people's hobby has been diversified. This characteristic feature has been partially contributed to the increase of people's demand on pet dogs. Recently, the total number of pet animals in Korea is assumed about three millions and the business relating to pet dogs has been growing rapidly

more than 20% each year since 1998 (www.kca.go.kr). Amid the growth of pet numbers and the increase of chance to pet exposing, very limited knowledge is available about the molds present on pet in Korea. Pet animals such as cats or dogs are one of common sources of mold contamination in houses. To understand the impact of molds on indoor air quality and human health it is prerequisite to obtain information on fungi present on the pet. To gain basic idea on the relation between mold species present on pet and indoor air, in this study we investigated what molds are present on pet dogs cared in houses in Korea.

Fungal samplings were undertaken in July of 2004 against 70 pet dogs at different houses and veterinary hospitals which were located at the area of Cheonan and Asan, Chungcheongnamdo, Korea. Skin debris or hairs of each pet dog were taken using cotton swabs or sterilized forceps, planted on the surface of SDA (Sabouraud Dextrose Agar) supplemented with ampicillin and streptomycin (Sigma Co., USA), and incubated for 5~7 days at 25°C. Fungal mycelia or spores grown out from the hairs and skin debris were transferred new SDA and purely cultured. The purely obtained fungal isolates were grown on diverse media including malt extract agar, corn meal agar, oat meal agar and potato dextrose agar for the observation of mycological features. Dermatophyte test medium (DTM, Asan Pharm. Co., LTD) was used for the detection of fungi causing mycosis on skin. Fungal identification was performed based on the observation of colony morphology and micro-structures, and referring identification books and online tools (De Hoog *et al.*, 2000; Kwon-

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Table 1. Fungi isolated from skin and hairs of 70 pet dogs

Genus	No. of isolates from hairs/ grown at 37°C/ shown DTM positive	No. of isolates from skins/ grown at 37°C/ shown DTM positive	Rate of genus/ grown isolate at 37°C/ shown DTM positive (%)
<i>Alternaria</i> spp	N	1/0/1	2.3/0/3.5
<i>Aspergillus</i> spp.	5/2/3	6/2/5	25/20/29
<i>Beauveria</i> spp.	N	1/1/1	2.3/5/3.5
<i>Chrysosporium</i> spp.	N	2/2/2	4.3/10/7
<i>Cladosporium</i> spp.	5/1/2	5/2/2	23/15/14
<i>Penicillium</i> spp.	3/2/2	6/3/3	20.5/25/18
<i>Scopulariopsis</i> spp.	N	1/1/1	2.3/5/3.5
<i>Trichoderma</i> spp.	1/0/1	N	2.3/0/3.5
Yeast type	3/2/2	5/2/3	18/20/18
Total	17/7/10	27/13/18	100/100/100

N: no isolation.

Chung and Bennett, 1992; Dugan, 2006; Barnett and Hunter, 1998; www.botany.utoronto.ca/ResearchLabs/MallochLab/Malloch/Moulds/Moulds.html) using a phase contrast light microscope (Zeiss, Axioskop 40) and a dissect microscope (Olympus, SZ2-ILST). When the fungi did not show specific morphology, we performed molecular identification by analyzing ITS rDNA sequences (White *et al.*, 1990). ITS rDNA amplification by PCR and sequencing analysis were performed using the method described Kim *et al.* (1999). The identity of the determined ITS rDNA sequences were searched through GenBank DNA database.

From 210 skin or hair samples taken from the 70 pet dogs, 44 fungal isolates were purely obtained. Among these isolates, 27 isolates were from skin and 17 from hairs (Table 1). From hairs, *Aspergillus*, *Cladosporium*, *Penicillium*, *Scopulariopsis*, and *Trichoderma*, and yeast were identified. From skins, *Aspergillus*, *Beauveria*, *Chrysosporium*, *Cladosporium*, *Penicillium*, *Scopulariopsis*, and yeast were identified. Both from skin and hairs, *Aspergillus* was the most commonly found genus and followed by *Cladosporium* and *Penicillium*. The ratio of fungal isolates belonging to these genera was 25, 23, 20.5%, respectively. These results indicate that *Aspergillus*, *Cladosporium*, and *Penicillium* are common groups of molds in pet dogs. This finding agree with that *Aspergillus*, *Cladosporium*, and *Penicillium* are common molds found in indoor air of houses and offices (Lee, 1998). It is interesting to know that house pet dogs carry the same type of fungal species found in inside buildings.

Many species belonging to the genera found at Table 1 are known to affect on human and dogs' health including allergy and mycosis (www.doctorfungus.org; De Hoog *et al.*, 2000). Especially the fungi that can able to grow at 37°C have been considered as potentially more harmful than the fungi that cannot grow at 37°C. Because 37°C is close to human's body temperature. Thus, we grew the 44 isolates at 37°C to investigate if there were fungi that able

to grow at the temperature. 7 of 17 isolates from hairs and 13 of 27 isolates from skin were able to grow at 37°C (Table 1). The 7 isolates of hair origin were found from the groups of *Aspergillus*, *Cladosporium*, *Penicillium* and yeast type. Meanwhile, the 13 isolates of skin origin were found from the groups of *Aspergillus*, *Beauveria*, *Chrysosporium*, *Cladosporium*, *Penicillium*, *Scopulariopsis*, and yeast type. Our data demonstrate that pet dogs carry some fungi that potentially harmful to humans.

Since dermatophytic fungi cause health problems in skin, nails and hair of animals and humans (Kwon-chung and Bennett, 1992), they become important in public health science (Nevoralova, 2006). Thus, we investigated their presence in the 44 isolates from pet dogs using DTM that has been known as one of useful media for detecting dermatophytic fungi at hospitals and clinical laboratories (Taplin *et al.*, 1969). If dermatophytes were cultured on DTM, the color of culture medium would change from yellow (negative) to red (positive) (Fig. 1), DTM test easily discriminate dermatophytic without an extensive knowledge of mycology (Blank and Rebell, 1965). In our DTM test, 10 of 17 isolates from hairs were found to be positive and 18 of 27 isolates from skin were found to be positive (Table 1). DTM positive isolates were found from *Aspergillus*, *Beauveria*, *Chrysosporium*, *Cladosporium*, *Penicillium*, *Scopulariopsis*, and yeast type. However, among the fungi that showed DTM positive results, there is no typical dermatophytic fungal species such as *Ephidermophyton*, *Microsporum*, and *Trichophyton* (Kwon-chung and Bennett, 1992). These fungal groups are special fungi that can infiltrate and exist in the keratinous organization (Miller *et al.*, 1995). Generally sympathizing dermatophytes are *Alternaria* spp., *Penicillium* spp. and *Aspergillus* spp. and so on. These fungi sympathizes with skins of healthy entity, and *Penicillium* spp. and *Aspergillus* spp. become a pathogenic mycosis that happen on various kinds of animal's skin, ear canal, conjunctiva, lung, bronchi, fetus. Considering the facts that DTM positive

Fig. 1. Examples of fungal growth on DTM. When the inoculum is positive on DTM, the color changes from yellow (A) to red (B). Examples of DTM test results with the fungal isolates from pet dogs (C).

fungi can produce alkali metabolites which may serve as irritants of skin and that some of species belonging to these DTM positive genera are able to cause skin irritation, it is suggested that fungi from pet dogs have to be carefully treated as possible sources of skin problems, allergic reactions and infection. DTM positive reaction by saprophytic fungi was also reported by the research of Kim *et al.* (1976).

In conclusion, in this study we first demonstrated in Korea that pet dogs carry diverse molds commonly present in indoor air. Our results indicate that pet dogs can be served as a contamination source of generating fungal bioaerosol into indoor environment. The detection of fungal isolates that can grow at 37°C and react positively on DTM implies that fungi carried by pet dogs have a potential to affect on human health. Further study is required to explore the identity of fungal species and their impact on human and animal health. And with the trends that rais-

ing of pet dogs increase in modern Korean society, our knowledge on fungi as bioaerosol in indoor environment should be extended.

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