Nat. Hist. Res., Vol. 6 No. 1: 9–14, March 2000

Occurrence of Ammonia Fungi on the Forest Ground After Decomposition of a Dog Carcass

Toshimitsu Fukiharu[1], Koichi Osaku[2], Kiyoshi Iguchi[3] and Masahiko Asada[1]

[1]Natural History Museum and Institute, Chiba
955–2 Aoba-cho, Chuo-ku, Chiba 260–8682, Japan
[2]2–84–2 Higashi-hatsuishi, Nagareyama-shi, Chiba 270–0114, Japan
[3]Hiraoka Environmental Science Laboratory
8–8 Hosoyama, Asao-ku, Kawasaki-shi, Kanagawa 215–0001, Japan

Abstract *Hebeloma vinosophyllum* and *Hebeloma spoliatum* (Basidiomycota, Agaricales), which belong to the ammonia fungi, occurred on the ground in the close vicinity of a decomposed carcass of a dog *Canis familiaris* in a forest dominated by *Quercus serrata,* Musashi-murayama city, Tokyo, central Japan. This is the third record of ammonia fungi occurring at the site of a decomposed dog carcass.

Key words: ammonia fungi, *Canis familiaris,* dog carcass, *Hebeloma spoliatum, Hebeloma vinosophyllum,* Mammalia, *Quercus serrata.*

Sagara (1975, 1992) defined "ammonia fungi" as "a chemoecological group of fungi, which sequentially develop reproductive structures exclusively or relatively luxuriantly, on the soil after a sudden addition of ammonia, some other nitrogenous materials, which react as bases by themselves or on decomposition, or alkalis." Under natural conditions, the ammonia fungi have been reported to appear where the dead body of an animal such as a dog (Sagara, 1976, 1984), cat (Sagara, 1976, 1981), snake (Hilton, 1978), rabbit (Takayama and Sagara, 1981) and kangaroo (Miller and Hilton, 1986) was decomposed on the forest ground. Experimentally, the ammonia fungi appear by the sudden addition of a sufficient amount of urea on the forest ground (Sagara, 1975). More than 50 species of ammonia fungi were listed from Japan (Sagara, 1975, 1992; Fukiharu and Hongo, 1995). In Sept. 1996, basidiocarps were observed in the close vicinity of a decomposed body of an animal in a forest in Tokyo, central Japan. In this paper, we report the results of careful examination of these fungi and the animal.

Materials and Methods

The study site was located in a forest at Noyama-kita-koen, Musashi-murayama city, Tokyo, at 35°46′ N lat., 139°23′ E long., approximately 150 m above sea level, about 1 km distance from Lake Sayama. The mean air temperature is 14.1℃ and annual precipitation is 1,631 mm (at Tachikawa city about 5 km away) and this area belongs to *Camellietea japonicae* zone (Okutomi *et al.*, 1987). The vegetation is dominated by *Quercus serrata* Murray and mixed with deciduous and ever-green hardwoods (*Ilex macropoda* Miq., *Clethra barvinervis* Sieb. et Zucc., *Eurya japonica* Thunb., *Styrax japonica* Sieb. et Zucc.) and conifer (*Chamaecyparis obtusa* (Sieb. et Zucc.) Endl.).

Fungi collected were dried at 60℃, and observed microscopically. Anatomical observations and measurements were made on the preparations mounted in a 5% aqueous solution of KOH. The bones of the animal collected were dried at 60℃, and anatomical observations and measurements were made. Specimens of fungi and animal examined are deposited at the Natural History Museum and Institute, Chiba (CBM).

Results and Discussion

Two fungal species were successively observed from 12 Sept. to 27 Sept. 1996 in close proximity to the decomposed animal carcass (Fig. 1A, B). These fungi were identified as *Hebeloma vinosophyllum* Hongo and *H. spoliatum* (Fr.) Karst. (Basidiomycota, Agaricales). *H. vinosophyllum* was first observed on 12 Sept. On 23 Sept., *H. vinosophyllum* (Figs. 1A, 2) and *H. spoliatum* (Figs. 1B, 3) were both observed. On 27 Sept. only *H. spoliatum* was observed. The animal was identified as *Canis familiaris* Linnaeus. Discussion and notes on morphology, ecology and distribution are provided for these two fungi and the animal.

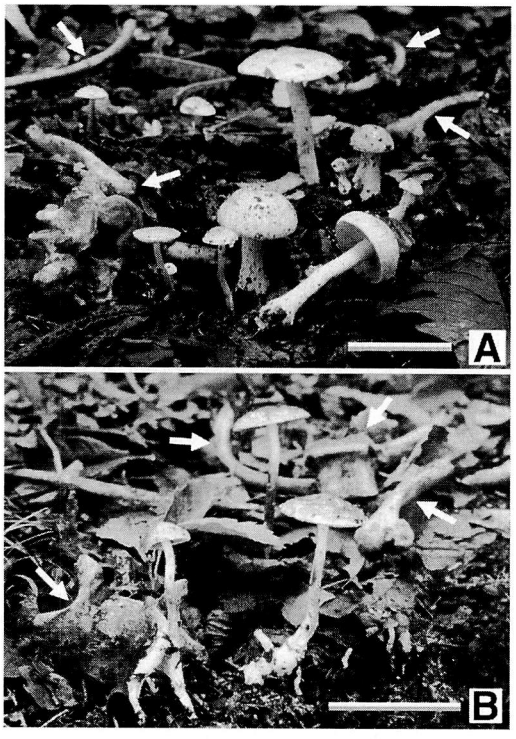

Fig. 1. Ammonia fungi occurring at the site of a decomposed carcass of a dog *Canis familiaris*. After decomposition of the dog carcass in a *Quercus serrata* dominated forest, Noyama-kita-koen, Musashi-murayama city, Tokyo, 23 Sept., 1996. A, *Hebeloma vinosophyllum* (CBM-FB 17141); B, *Hebeloma spoliatum* (CBM-FB 17142). Arrows show the bones of a dog carcass. Scale bars: 2 cm.

1. *Hebeloma vinosophyllum* Hongo
(Figs. 1A, 2)

This fungus is identified as *Hebeloma vinosophyllum* Hongo from the following diagnostic characters of this species (Hongo, 1965): the tricholomatoid basidiocarp (Fig. 2A), cortinate partial veil, medium-sized (9.5–11.0 × 6.0–6.5 μm) densely verrucose amygdaliform basidiospores (Fig. 2C), and purple-brown basidiospore-mass color.

In the early stage of the study of ammonia fungi, this species was confused with *Alnicola lactariolens* Clémençon et Hongo. These two species resemble each other in having the same macroscopic characters of basidiocarp, the same purple-brown basidiospore-mass color and the same microscopic characters of basidiospore. In addition, *A. lactariolens* shows the same behavior as an ammonia fungus (Fukiharu & Hongo, 1995). However, *H. vinosophyllum* (Fig. 2B) had the glutinous upper layer of the pileipellis which distinguishes *Hebeloma* from *Alnicola* (Singer, 1986).

Hebeloma vinosophyllum was described as a new species without any description of behavior that suggested it to be an ammonia fungus (Hongo, 1965). Later, Sagara (1973) reported that the habitat of this species was closely related with the decomposed animal matter such as the carcass or excrement. He named the species a "proteophilous fungus" (Sagara, 1973) and later reclassified it as an ammonia fungus (Sagara, 1975). *H. vinosophyllum* has been reported to occur at the site of a decomposed dead body of a dog (Sagara, 1976), cat (Sagara, 1976, 1995) or rabbit (Sagara, 1981). This is the second report of this fungus at the site of a decomposed dog carcass.

In natural conditions, *H. vinosophyllum* had been reported from Kyoto and Nagaoka-kyo cities, Kyoto Pref. in a *Castanopsis* forest (Sagara, 1976, 1981); from Ootsu city, Shiga Pref. in a mixed forest of *Castanopsis* and *Quercus*, and from *Pinus* forest (Hongo, 1965); from Fujian, China in an ever-green mixed forest of *Castanopsis* and *Quercus* (Hongo *et al.*, 1996). In addition, by application of urea to the soil in the field, this fungus has been reported from Oita, Tottori, Kyoto and Shiga

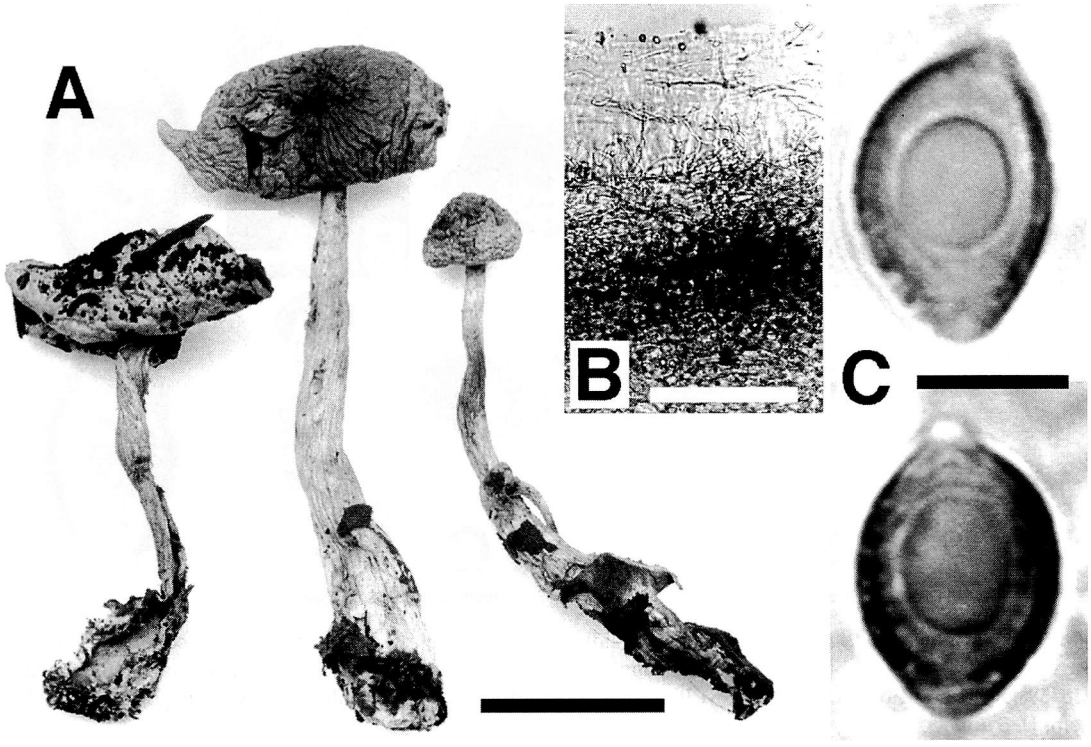

Fig. 2. *Hebeloma vinosophyllum.* A, basidiocarps; B, anticlinal section of the pileal surface; C, basidiospores. (A, dried material; B, C, mounted in 5% KOH solution; A–C, CBM-FB 17060). Scale bars: A, 2 cm; B, 150 μm; C, 5 μm.

Pref. in western Japan in *Castanopsis, Chamaecyparis, Cryptomeria, Pinus, Quercus* and *Pleioblastus* vegetations (Sagara, 1975). By the same method, this fungus has been reported from eastern Japan, from Nishi-nasuno, Tochigi Pref. in a mixed forest of *Pinus* and *Quercus* (Suzuki, 1992); from Mt. Kiyosumi, Chiba Pref. in a mixed forest of *Quercus, Abies, Pinus* and *Tsuga* (Suzuki, 1992); from Abukuma Mountains, Fukushima Pref. in a mixed forest of *Castanopsis, Quercus* and *Abies* (Fukiharu, unpublished data). Biogeographically, the present case fills the blank area of its distribution record in the central part of Japan. *H. vinosophyllum* is known to be a biotrophic (ectomycorrhizal) fungus (Sagara, 1995). The vegetation data from the present case suggests an ectomycorrhizal relationship of *H. vinosophyllum* with the dominant tree species *Quercus serrata.*

Specimens examined. Japan, Honshu, Tokyo, Musashi-murayama city, Noyama-kita-koen, 150 m a.s.l., 12 Sept. 1996 (CBM-FB 17060), 23 Sept. 1996 (CBM-FB 17141).

2. *Hebeloma spoliatum* (Fr.) Karst.
(Figs. 1B, 3)

This species is identified as *Hebeloma spoliatum* (Fr.) Karst. sensu Hongo (Imazeki and Hongo, 1987) from the following diagnostic characters of this species (Imazeki and Hongo, 1987): the tricholomatoid basidiocarp (Fig. 3A), a long rooting stipe (Fig. 1B) sometimes present, brown sticky pileus, an evanescent cortinate partial veil present at the very young stage (Fig. 3B) but lacking later (Fig. 3C), medium-sized (8.5–10.5×5.0–5.5 μm) densely verrucose amygdaliform basidiospores (Fig. 3D) and pale brown spore-mass color.

Hebeloma spoliatum has been reported from Europe (Bresadola, 1927–1933; Lange, 1935–1940; Bruchet, 1970) and from North America (Smith, 1938, 1984), but behavior specific to ammonia fungi has not been known. Sagara (1973) first reported that this species (*H. spoliatum* sensu Hongo) was closely related with decomposed animal matter and clas-

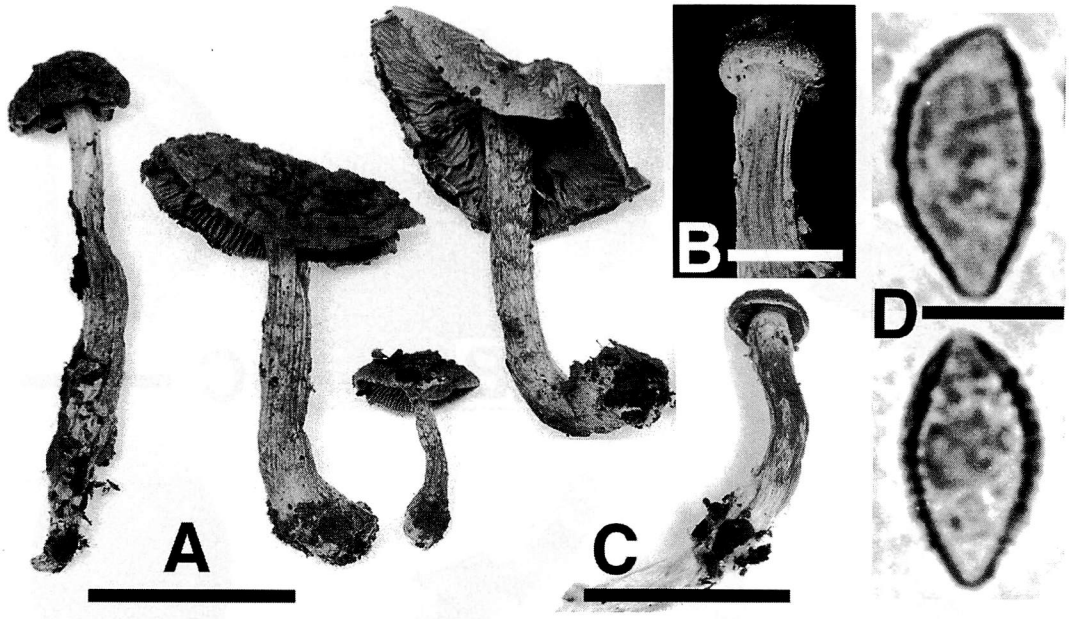

Fig. 3. *Hebeloma spoliatum.* A, matured basidiocarps; B, C, young basidiocarps with cortinate veil at a very early stage (B) but disappearing soon (C); D, basidiospores. (A, B, C, dried material; D, mounted in 5% KOH solution; A, C, D, CBM-FB 17143; B, CBM-FB 17142). Scale bars: A, 2 cm; B, 2 mm; C, 1 cm; D, 5 μm.

sified this species as "an ammonia fungus" (Sagara, 1975). In natural conditions, *H. spoliatum* has been reported to occur at the site of decomposed animal matter, such as feces of cow, pig, fowl and human (Sagara, 1978a, 1995), human excrement night soil (Sagara, 1995), abandoned wasp (*Vespula flaviceps lewisii* Cameron) nest (Sagara, 1979; Sagara *et al.*, 1985), moles (*Mogera imaizumii* Kuroda and *M. wogura wogura* Temminck) midden (Sagara, 1978b, 1980, 1995, 1998), the shrew mole (*Urotrichus talpoides talpoides* Temminck) midden (Sagara *et al.*, 1981) and raccoon dog (*Nyctereutes procyonoides viverrinus* Temminck) midden (Sagara, 1995), from Hiroshima, Hyogo, Kyoto, Aichi and Niigata Pref. in Japan in *Quercus, Pinus, Fagus* dominated forests. Sagara (1977) reported the first case of this fungus from the site of a decomposed dog carcass in Kyoto in a *Castanopsis* dominated forest, and this is the second case. *H. spoliatum* is known to be a biotrophic (ectomycorrhizal) (Sagara, 1995). The vegetation data from the present case suggest an ectomycorrhizal relationship of *H. spoliatum* with dominant tree species *Quercus serrata.*

Specimens examined. Japan, Honshu, Tokyo, Musashi-murayama city, Noyama-kita-koen, 150 m a.s.l., 23 Sept. 1996 (CBM-FB 17142), 27 Sept. 1996 (CBM-FB 17143, 17149).

3. *Canis familiaris* Linnaeus
(Figs. 1, 4)

This animal was identified as a mammal Canidae since the bones had the following diagnostic characters of this family (Evans and Christensen, 1979). The wings of the first cervical vertebrate (atlas) project from the lateral masses (Fig. 4a), the cranial border of scapula is arciform (Fig. 4b) and the cervical tubercle of trochanter major of femur is projected (Fig. 4c, d). Considering from the whole bone size, the shoulder height of this animal is estimated to be ca. 80 cm. The natural mammal Canidae habitants in this area are *Canis familiaris* Linnaeus, *Nyctereustes procyonoides viverrinus* Temminck and *Vulpes vulpes* Linnaeus of which *C. familiaris* is the only animal that has the shoulder height of this animal.

The occurrence of ammonia fungi at the site of a decomposed dog carcass was first reported from Kyoto, Japan with the appearance of *H. vinosophyllum* (Sagara, 1976) and

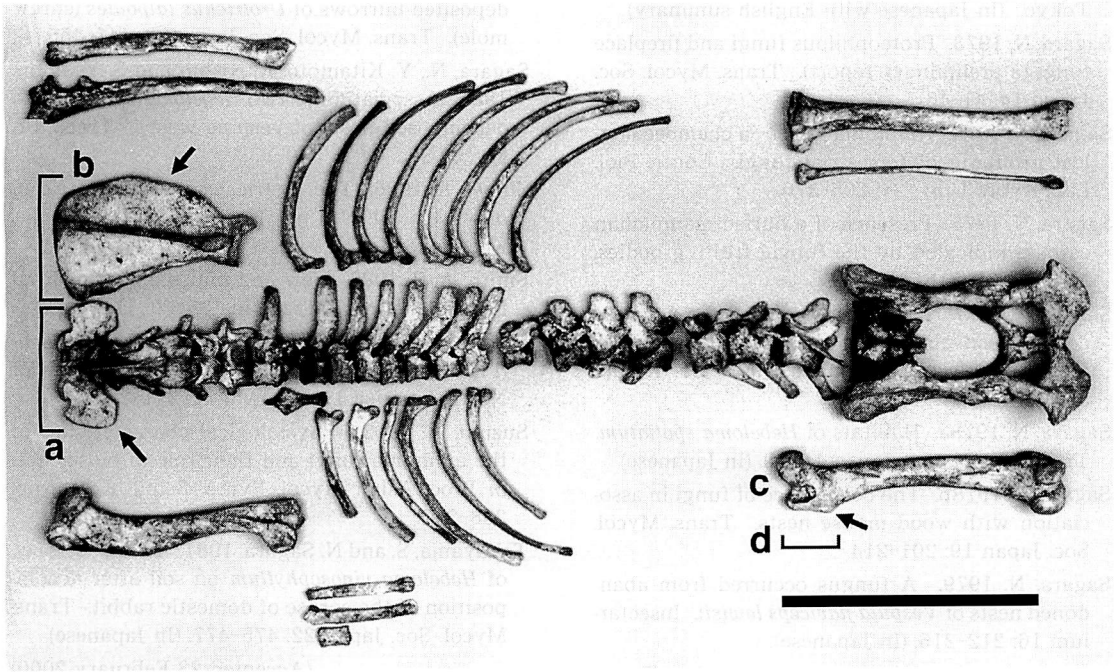

Fig. 4. The bone of a dog *Canis familiaris* (CBM-ZZ 2676). a, The first cervical vertebrate (arrow shows the wing part); b, scapula (arrow shows the cranial border); c, femur; d, trochanter major (arrow shows the cervical tubercule). Scale bar: 20 cm.

H. spoliatum (Sagara, 1977) from the same place. The second case was of *Hebeloma syriense* Karsten found by D. P. Lewis "around well-decayed corpses of dogs" in Texas, North America (Sagara, 1984). Thus, this is the third case to be reported.

Specimen examined. Japan, Honshu, Tokyo, Musashi-murayama city, Noyama-kita-koen, 150 m a.s.l., 23 Sept. 1996 (CBM-ZZ 2676).

Acknowledgments

We express our sincere thanks to Prof. N. Sagara of Kyoto Univ. for critical reading of this manuscript and also to Mr. T. Komiya of Natural History Museum and Institute, Chiba for advice on the mammal bone identification.

References

Bresadola, J. 1927–1933. Iconographia Mycologia. 26 vols. Soci. Bot. Ital. Mus. Civico di Storia Nat. di Trento, Milano.

Bruchet, G. 1970. Contribution a l'étude du genre *Hebeloma* (Fr.) Kummer; partie spéciale. Bull. Soc. Linn. de Lyon 39(6): 1–131.

Evans, H. E. and G. C. Christensen. 1979. Miller's anatomy of the dog, 2nd ed. 1181 pp. W. B. Saunders Company, Philadelphia.

Fukiharu, T. and T. Hongo. 1995. Ammonia fungi of Iriomote Island in the southern Ryukyus, Japan and new ammonia fungus, *Hebeloma luchuense*. Mycoscience 36: 53–58.

Hilton, R. N. 1978. The ghoul fungus, *Hebeloma* sp. ined. Trans. Mycol. Soc. Japan 19: 418.

Hongo, T. 1965. Notes on Japanese larger fungi (17). J. Jpn. Bot. 40 (10): 311–318.

Hongo, T., N. Kinjho, N. Taketomo, N. L. Huang, J. L. Wu and J. T. Lin. 1996. Agaricales in evergreen oak forests in East-Asia. Nippon Kingaku-kai Kaiho 37: 63–64. (In Japanese)

Imazeki, R. and T. Hongo (eds.). 1987. Colored illustrations of mushrooms of Japan. Vol. I. 325 pp. Hoikusha Publishing, Osaka. (In Japanese with Latin names and index)

Lange, J. E. 1935–1940. Flora Agaricina Danica. 5 vols. Soc. Advan. Mycol. Denmark and Danish Bot. Soc., Copenhagen.

Miller, O. K. Jr. and R. N. Hilton. 1986. New and interesting agarics from Western Australia. Sydowia 39: 126–137.

Okutomi, K., S. Okuda, S. Tsuji and Y. Hoshino. 1987. Vegetation of Tokyo. 249 pp. Tokyo-to,

Tokyo. (In Japanese with English summary)

Sagara, N. 1973. Proteophilous fungi and fireplace fungi (a preliminary report). Trans. Mycol. Soc. Japan 14: 41–46.

Sagara, N. 1975. Ammonia fungi—a chemoecological grouping of terrestrial fungi. Contr. Biol. Lab. Kyoto Univ. 24: 205–276.

Sagara, N. 1976. Presence of a buried mammalian carcass indicated by the fungal fruiting bodies. Nature 262: 816.

Sagara, N. 1977. The fungi from the animal matter decomposed place such as mammalian carcass and human feces. Nonezumi 139: 19–21. (In Japanese)

Sagara, N. 1978a. Habitats of *Hebeloma spoliatum*. Trans. Mycol. Soc. Japan 19: 90. (In Japanese)

Sagara, N. 1978b. The occurrence of fungi in association with wood mouse nests. Trans. Mycol. Soc. Japan 19: 201–214.

Sagara, N. 1979. A fungus occurred from abandoned nests of *Vespula flaviceps lewisii*. Insectarium 16: 212–215. (In Japanese)

Sagara, N. 1980. Not mouse but mole. Trans. Mycol. Soc. Japan 21: 519.

Sagara, N. 1981. Occurrence of *Laccaria proxima* in the grave site of a cat. Trans. Mycol. Soc. Japan 22: 271–275.

Sagara, N. 1984. On "Corpse finder". McIlvainea 6(2): 7–9.

Sagara, N. 1992. Experimental disturbances and epigeous fungi. *In* G. C. Carroll and D. T. Wicklow (eds.), The Fungal Community, 2nd ed., pp. 427–454. Marcel Dekker, New York.

Sagara, N. 1995. Association of ectomycorrhizal fungi with decomposed animal wastes in forest habitats: a cleaning symbiosis? Can. J. Bot. 73 (Suppl. 1): 1423–1433.

Sagara, N. 1998. Methods for studying the nesting ecology of moles through observation of mushroom fruiting. Honyurui Kagaku 38(2): 271–292. (In Japanese with English abstract)

Sagara, N., S. Honda, E. Kuroyanagi and S. Takayama. 1981. The occurrence of *Hebeloma spoliatum* and *Hebeloma radicosum* on the dung-deposited burrows of *Urotrichus talpoides* (shrew mole). Trans. Mycol. Soc. Japan 22: 441–455.

Sagara, N., Y. Kitamoto, R. Nishio and S. Yoshimi. 1985. Association of two *Hebeloma* species with decomposed nests of vespine wasps. Trans. Br. Mycol. Soc. 84(2): 349–352.

Singer, R. 1986. The Agaricales in modern taxonomy, 4th ed. 981 pp. Koeltz Scientific Books, Koenigstein.

Smith, A. H. 1938. New and unusual agarics from North America I. Mycologia 38: 20–41.

Smith, A. H. 1984. Studies of species of *Hebeloma* (Fr.) Kummer from the Great Lakes Region of North America 1. Sydowia 37: 271–283.

Suzuki, A. 1992. Physiological characteristics of the ammonia fungi and their role in bioscience. *In* Proc. Asian Mycol. Symp. Seoul, Korea, pp. 238–247.

Takayama, S. and N. Sagara. 1981. The occurrence of *Hebeloma vinosophyllum* on soil after decomposition of the corpse of domestic rabbit. Trans. Mycol. Soc. Japan 22: 475–477. (In Japanese)

(Accepted 23 February 2000)

犬の死体分解跡における アンモニア菌の発生

吹春俊光[1)]・大作晃一[2)]・井口　潔[3)]・浅田正彦[1)]

[1)]千葉県立中央博物館
〒260-8682 千葉市中央区青葉町 955-2
[2)]〒270-0114 千葉県流山市東初石 2-84-2
[3)]財団法人　平岡環境科学研究所
〒215-0001 神奈川県川崎市麻生区細山 8-8

アンモニア菌に属するアカヒダワカフサタケとアシナガヌメリ（担子菌綱，ハラタケ目）が，東京都内のコナラが優占する林内でイヌの死体分解跡に発生した．これまで何種かの動物の死体分解跡からアンモニア菌が発生することは知られており，イヌの死体跡から発生が確認されたのは今回が３例目である．