Naphthalene in Formosan Subterranean Termite Carton Nests

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Naphthalene was detected in nest cartons of Formosan subterranean termites, *Coptotermes formosanus* Shiraki. The concentration of naphthalene ranges from 50.56 to 214.6 μg/kg. This is the first report of naphthalene being associated with termites. Some possible functions of naphthalene in a termite society are discussed.

Keywords: *Naphthalene; Coptotermes formosanus; termite nest*

INTRODUCTION

Coptotermes formosanus Shiraki, a native of mainland China, has become an important international structural pest (Su and Tamashiro, 1987). After its initial discovery in the continental United States in Houston, TX, in 1965 (King and Spink, 1969), its distribution has included Alabama, California, Florida, Georgia, Louisiana, Mississippi, South Carolina, and Tennessee (Su and Tamashiro, 1987; Haagsma et al., 1995). *C. formosanus* may in some cases displace native *Reticulitermes* spp. (La Fage, 1987).

Foraging areas of *C. formosanus* are continually expanded by enlarging or building accessory nests (Tang and Li, 1959; Lin, 1987). Soil, masticated wood, and excrement, cemented by saliva and excrement (called "carton"), are used to make nests (Tang and Li, 1959; King and Spink, 1969). Galleries and shelter tubes connect primary nests to accessory nests and feeding sites (Tang and Li, 1959; King and Spink, 1969).

In this study, naphthalene was discovered and quantified in termite carton nests by using gas chromatography−mass spectrometry (GC−MS). The discovery of naphthalene in nest carton poses interesting questions related to colony function.

MATERIALS AND METHODS

Nest Carton Materials. Termites with their nests were collected from colonies infesting houses and trees in New Orleans and Lake Charles, Louisiana. Fresh carton material was used in this experiment.

Sample Extraction. For quantification purpose, colonies from New Orleans and Lake Charles, LA, were collected. Twenty-five grams of carton material from each colony were ground into a powder by using a ceramic mortar and pestle and then placed in a 125 mL flask. Before extraction, a 20:1 azulene (99+%, Sigma Chemical Co., MS) hexane solution at a concentration of 0.178 mg/mL was added to each sample as an internal standard. Each sample was extracted twice with 25 mL of dichloromethane and concentrated to 0.1 mL by using a rotary evaporator and nitrogen flow before analysis by GC−MS. As a control, 50 mL of dichloromethane alone was concentrated to 0.1 mL and analyzed.

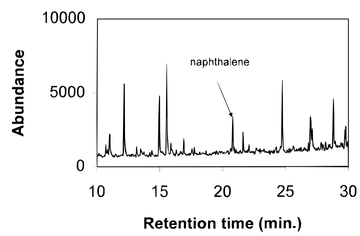

Figure 1. Total ion chromatogram of Formosan subterranean termite nest carton extract.

Instrumental Methods. A Hewlett-Packard 5890 gas chromatograph with a 5970 GC−MS (Palo Alto, CA) equipped with a 50 m J&W (Folsom, CA) DB5-MS capillary column (0.2 mm i.d. and 0.33 m stationary phase) was used. The injection temperature was 250 °C. Initial GC temperature was 50 °C for 1 min; the temperature was then increased to 300 °C at 5 °C/min and held for 9 min. The instrument was operated in linear scanning mode from *m*/*z* 35 to 450.

Identification of Naphthalene. Identification of naphthelene was made by comparison of mass spectra and GC retention times of peaks in the sample with those of a naphthalene standard (99+%, Sigma Chemical Co., MS).

RESULTS

Naphthalene in Termite Nest Carton. A typical total ion chromatogram of termite nest carton extract is shown in Figure 1. The retention time of naphthalene was 20.86 min under this GC−MS system. The retention time and mass spectrum of the peak at 20.86 min (Figures 1 and 2) are matched with those of the naphthalene standard shown in Figures 3 and 4. The

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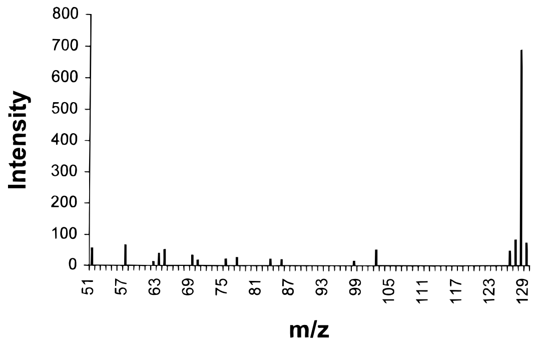

Figure 2. Typical mass spectrum of the peak at GC retention time 20.86 min of the nest carton extracts.

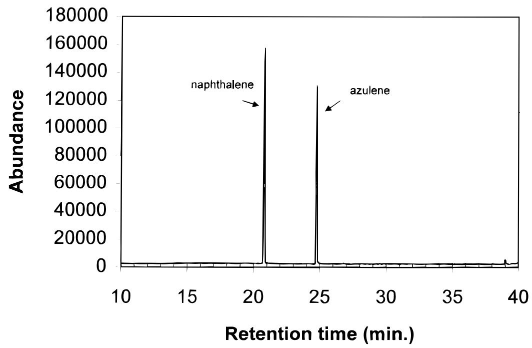

Figure 3. Total ion chromatogram of naphthalene standard and internal standard azulene.

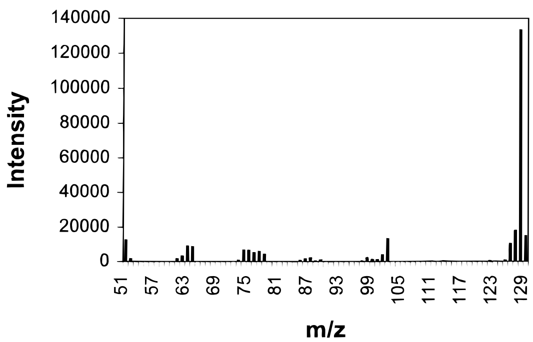

Figure 4. Typical mass spectrum of the naphthalene standard.

Table 1. Naphthalene Concentrations in Nest Cartons of Formosan Subterranean Termites

colony	mean, μg/kg	standard error
1	214.6	45.9
2	50.56	8.0
3	183.5	103.8
4	84.23	15.69

concentration of naphthalene ranged from 50.56 to 214.6 μg/kg (Table 1). Naphthalene was not detected in the solvent system.

DISCUSSION

This is the first report of naphthalene found in association with termites. A termite nest is a closed system which protects termites from air movement and provides a microclimate which is controlled and different from that of the surrounding environment (Noirot, 1970). Naphthalene at 50.56−214.6 μg/kg in termite carton nests may constitute a unique chemical defense strategy of Formosan subterranean termites in that the closed system of termite nest may allow for fumigation as a defense strategy. Naphthalene is a common arthropod fumigant and also is used as a repellent of bats, pigeons, sparrows, squirrels, starlings, and rabbits (Office of Pesticides and Toxic Substance, 1981). Naphthalene also is an antimicrobial and anthelmintic agent (Bolton and Eaton, 1968). Such nest fumigation may prevent or repel other invertebrates, such as ants, and other animals from entering the carton nests and inhibit any pathogenic microorganisms from proliferating. It is probable that termites are biochemically and physiologically adapted to their nest environment, which is harsh to other organisms. As a soil-dwelling creature, Formosan subterranean termites confront many adversaries such as ants, fungus, bacteria, and nematodes. Antimicrobial ability is believed to be one of the most important attributes of ants and one which makes them so successful in the soil habitat (Wilson, 1971). This may be true for Formosan subterranean termites as well. Fumigating the nest with naphthalene and other volatile compounds may play an important role in inhibiting microorganism and invertebrate invaders in the nest.

The origin of the naphthalene in the nest carton is unknown. Naturally occurring naphthalene is known to originate from coal and petroleum and incomplete combustion of organic materials such as forest fires (GDCH-Advisory Committee on Existing Chemicals of Environmental Relevance, 1989). Since termites use soil, masticated wood, and excrement to make their nests (Tang and Li, 1959; King and Spink, 1969; Wilson, 1971), one possible source of naphthalene is the processed food of termites or soil. The finding of Azuma et al. (1995) that naphthalene is in *Magnolia* flower suggests the possibility of a food source origin. Plants contain a large amount of organic molecules which also could serve as precursors of naphthalene. Naphthalene also was recently found in the forehead region of male white-tailed deer, *Odocoileus virginianus* (Gassett et al. 1997). This suggests that biosynthesis of naphthalene in animals is also possible. Another possible origin of naphthalene is microbial biosynthesis in the termite nest, in the gut, or on the food. However, no direct evidence of naphthalene synthesis by any microorganisms, invertebrates, or vertebrates has been found.

With the development of the petroleum industry, pollution of petroleum hydrocarbons including naphthalene in the marine and estuaries environments is a worldwide phenomenon (Connell and Miller, 1980). Naphthalene has been found in some aquatic animals such as oysters, mussels, and fish as a result of environmental contamination (GDCH-Advisory Committee on Existing Chemicals of Environmental Relevance, 1989). Although soil pollution with naphthalene from the petroleum industry is possible, it was very unlikely that the widely dispersed termite nests examined in the experiments reported herein were all contaminated by extraneous sources of naphthalene.

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