Available online at www.sciencedirect.com
SCIENCE DIRECT®
Palaeogeography, Palaeoclimatology, Palaeoecology 237 (2006) 19 – 31
PALAEO
www.elsevier.com/locate/palaeo

Environmental information from guano palynology of insectivorous bats of the central part of the United States of America

Louis J. Maher Jr. *

University of Wisconsin, Department of Geology and Geophysics, 1215 W. Dayton Street, Madison, WI 53706-1692, USA

Received 13 February 2003; accepted 8 November 2005

Abstract

Bat droppings accumulate in caves, and the resultant guano contains a stratigraphic record of the environment analogous to the record from lake sediment and peat. The bats forage at night for insects; they return to the cave during the day to sleep and care for their young. They attach themselves to suitable perches in the cave ceiling, and their excrement accumulates on the floor below. Flying requires a lot of energy, and bats of temperate regions consume large numbers of night-flying insects. In some situations the guano can reach a depth of meters in hundreds to thousands of years, and it has a valuable chronostratigraphy. The bat scats occur as small pellets that represent the non-digestible portion of the animal's diet in the preceding few hours; hence the diet provides information about the time of the year the feeding occurred. Bat guano contains, among other things, insect fragments, hair, pollen, and mineral matter. Night-flying insects do not normally visit flowers for the pollen; many species do not eat during the flying phase of their life cycle, and those that do generally are nectar feeders. Although the insects are not after the pollen, they do fly through a pollen-laden environment, and the pollen and dust adheres to their bodies. The insects essentially act as living traps for airborne debris. The bats also are furry pollen traps; during grooming they ingest pollen and dust enmeshed in their fur, and this also is excreted. The pollen in an individual scat contains a record of the atmospheric pollen during a single day in the past. This kind of detail is rarely available from lake sediment. Chemical analysis of individual bat scats in a time series can chart the changing environment caused by agriculture, industry, volcanic dust, and a host of other details that depend only on the cleverness of the researcher. Careful ^{14}C analysis can isolate the times when bats did not use the cave, and that may be useful in interpreting past conditions. If the insect types in the guano change over time, that may provide evidence of changing climate. Pollen was analyzed from guano samples taken from Tumbling Creek Cave near Protem, Missouri, USA. The cave contains a maternal colony of the Grey Bat (*Myotis grisescens*) that occupies the cave for a short time each year. Scats collected from the base of a 70 cm thick cone of guano yielded an AMS ^{14}C date of 2810±40 yr BP. The fecal material has a crumbly structure below the surface; it was of mahogany color (7.5 YR 2/1 to 3/2) and had no noticeable odor. Guano can be processed like normal sediment, but simple washing in a weak detergent solution followed by acetolysis appears adequate.

Keywords: Bat guano; Central USA; Tumbling Creek Cave; Palynology; *Myotis grisescens*; Insects

1. Introduction

In 1990 the author was asked to conduct a palynological study of bat guano in Tumbling Creek Cave. The cave is located near the small town of Protem in extreme southern Missouri, USA. It is owned by Thomas J. and

* Fax: +1 608 262 0693.
E-mail address: maher@geology.wisc.edu.

0031-0182/$ - see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.palaeo.2005.11.026

Catherine L. Aley and operated as the Ozark Underground Laboratory. It has designated walkways but no electric lighting. The cave has been named a National Historic Landmark; it is said to contain the most diverse fauna of any cave west of the Mississippi River.

It also hosts a large maternal colony of the Grey Bat (*Myotis grisescens*), an endangered species. Various sheets and mounds of guano occur under places where the bats roost. The author was informed that another researcher's effort to examine fungal elements in the guano was thwarted because it contained an overwhelming abundance of pollen grains. Therefore T.J. Aley suggested the study of the guano's pollen.

The idea was intriguing: insects pick up pollen by visiting flowers; many temperate-region bats are insectivorous; pollen grains are difficult to digest and should accumulate stratigraphically in the mounds of guano under the bat roosts. The author's initial assumption was that the piles of guano would yield a record of insect-pollinated plants that might complement the wind-pollinated types in lake-sediment cores. This hypothesis can be easily falsified, as demonstrated in this paper.

The cave was visited on two separate occasions. Preliminary samples were collected in late May, 1990. Then in mid-August, 1991, photographs were taken, and additional guano samples were gathered.

Fenton (1983) provides an excellent account of bat behavior, diets, navigation, reproduction, and lineage. As a group bats are known to eat insects, other arthropods, fish, mice, bats, and other vertebrates as well as fruit, nectar, and pollen. The bats of the temperate region of the New World are generally insectivorous. The aforementioned Grey Bat (*M. grisescens*) is one of the Plain-Nosed Bats (Vespertilionidae). This family has the largest number of species, and it is found almost everywhere that bats are found.

Bats are often maligned, and whole colonies are ruthlessly killed when they come into contact with humans (Clark et al., 1988; Clawson, 1992). Some illnesses such as rabies and histoplasmosis may be associated with bats (Sorley et al., 1979; McMurray and Russell, 1982), but in the main they are very beneficial little animals. McCracken and Gustin (1987) estimate that a colony of Mexican free-tailed bats living in a cave in Texas, nightly consume about half of their own body weight in insects. Given an average body weight of 14 g per bat, the twenty million bats in the colony may consume 140 metric tons of insects each night.

There appear to be few published references to the palynology of guano (Pons and Quézel, 1958; see Leroy and Simms, this volume), but a number of palynologists have told the author that they have looked for pollen in guano. Owen Davis (personal communication, 1991) mentioned he had processed a sample of guano but found no pollen, and this led to a fascinating discussion of whether certain bat species may have evolved a way to digest pollen exine to gain more energy from their food. There is a body of palynological studies of caves, but these usually deal with the mineral sediments. Coles et al. (1989) review the literature and discuss the many ways that pollen can get into caves (air, streams, seepage, insects, and animals); they do not, however, mention the important role played by bats with their guano.

This paper provides information about Tumbling Creek Cave and the surface vegetation that surrounds it. A portion of the cave plan is provided, and areas and types of guano deposits are noted. The reason the guano deposits are separate and of different shapes is discussed, and it is shown that they can be explained by a simple mathematical model based on the bats' expected roosting behavior. Procedures are discussed for sampling the guano and for extracting its pollen. Observations are also made about the guano organic and mineral content. It will be demonstrated that the pollen found in the guano of insectivorous night-flying bats is a good representation of the pollen in the atmosphere through which the insects and the bats fly. Finally, suggestions are made for devices that can extract guano samples without deforming the material. A bulk sample of the bodies of night-flying insects can be correlated with the pollen in lake sediments in the central United States.

2. Methods

2.1. Tumbling Creek Cave

The cave (36°38′18″ N, 92°48′23″ W) is in Taney County Missouri, and it can be found on the Protem 7.5 minute Quadrangle (1968, scale 1/24,000, contour interval 20 ft) in SW 1/4, Sec 26, T22N, R17W (Fig. 1). The cave system has developed on the Ozark Plateau in the Cotter Dolomite of Ordovician age. The entrance elevation is 244 m (800 ft) above sea level.

Lying far south of the southern limit of Pleistocene glaciation, the area's soils are poor, and the surface often has a thick residuum of chert. The surface is deeply-dissected with a relief of about 200 m. The regional arboreal vegetation is characterized by Küchler (1964) as *Quercus* (oak), *Carya* (hickory), *Juniperus* (cedar), and *Pinus* (pine).

The local vegetation is primarily *Quercus* (oak) and *Carya* (hickory) forest. Secondary species consist of *Juglans nigra* (black walnut), *Celtis occidentalis* (hackberry), *Juniperus virginiana* (eastern red cedar), *Nyssa sylvatica* (black gum), *Gleditsia triacanthus* (honey locust), and *Platanus occidentalis* (American sycamore). Herbaceous plants include *Andropogon* (big and little bluestem), *Sorghastrum* (Indian grass), and other prairie components.

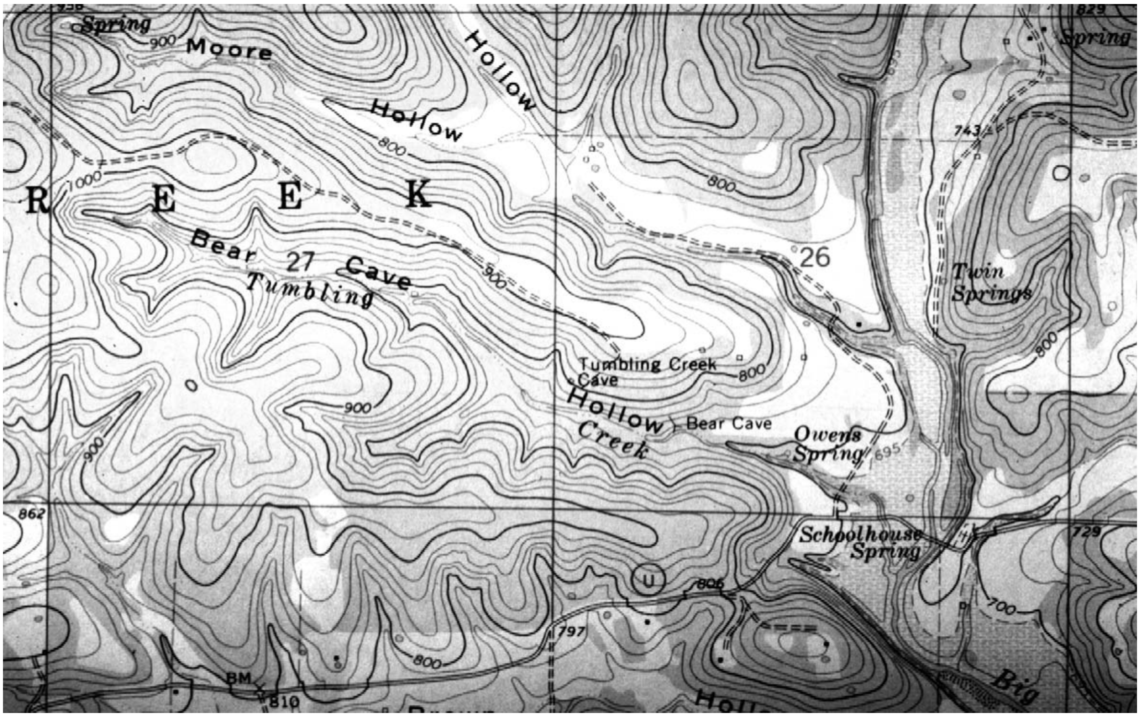

Fig. 1. Portion of the Protem, MO 7.5 minute quadrangle (1968, scale 1/24,000, contour interval 20 ft). Tumbling Creek Cave is in the SW 1/4 of Section 26. Sections are 1 mi (1.6 km) on each side. The label "TCC" on Fig. 10 marks the position of Tumbling Creek Cave in the central part of the United States.

The Grey Bat that occupies Tumbling Creek Cave travels up to 40 km in a night's feeding. It gets its food mainly along riparian corridors, but it also feeds over the top of the forest canopy and in clearings (T. Aley, personal communication, 2003).

Fig. 2 is a map of a portion of the cave. People now enter through a vertical shaft with stairs; the bats use natural openings. The passages were formed by subterranean streams, but some channels are now abandoned and dry. The main guano deposit is in one of the dry channels. The bats use "Hibernation Hall" as a nursery.

Initial guano samples were taken from a 70 cm high conical mound in the East Passage, about 500 m from the cave entrance. During the walk to this mound, a few bats were seen clinging to the ceiling in places where the surface had sufficient roughness for their claws to get a purchase. They avoid sites directly over the water and ceiling areas that are too smooth. At one place a thin carpet-like layer of guano stretched along the path for several meters (Fig. 3); it was a meter wide, several centimeters thick in the middle, feathering to a thin edge at each margin. The sheet of guano contained a number of light-colored patches of fungus. Guano is about the only energy source for the small creatures living in the cave, and the molds convert it to a form that some life forms find edible.

The isolated guano cone appears to have formed at a place the bats congregate. A number of samples from the margins of the mound were taken in glass vials; these would be used to experiment with processing techniques. A standard Livingstone piston sampler, designed to take lake-sediment cores, was used to obtain a core from the mound. After anchoring the piston cable, the corer was carefully pushed into the guano mound. The whole core

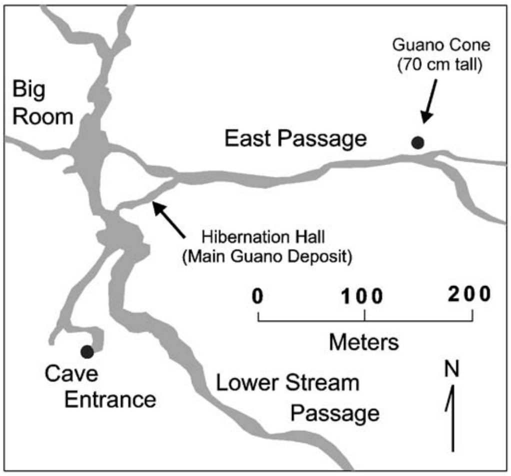

Fig. 2. Underground map of a portion of Tumbling Creek Cave.

barrel was wrapped in plastic foil when it was removed in order to avoid handling the adhering feces. The core was extruded later after return to the surface.

Although the corer had penetrated the entire 70 cm of the guano mound, even chipping a small fragment of dolomite from the cave floor, the extruded core was compressed to 35 cm, i.e. half its original length. The author resolved to use another type of sampler for future work; this will be discussed later. The core of guano was wrapped in plastic and aluminum foils for transfer to the pollen laboratory at the University of Wisconsin-Madison.

The various shapes of the cave's guano deposits were intriguing. On the assumption that the first bat to roost will choose a comfortable place on the cave ceiling, and late-comers will group around the first for warmth and company, the choice position establishes a mean position for the guano deposit, and the grouping of the surrounding bats contributes to a normal sort of distribution around the mean. The author later wrote a short computer program to model how this behavior would influence the shape of a pile of guano. The program accepts a value for the mean and standard deviation and draws random samples from that distribution. The assumption is made that the draw represents the position of the excreting bat and that the scat will drop vertically to the floor below. The program also keeps track of the growing heap, allowing "gravity" to cause any chance scat pinnacles to collapse down hill, and plots the results on the computer screen. Fig. 4 shows an average run in which 26,000 scats were allowed to accumulate. The program switches the falling guano between black and white after every 2000 scats have fallen. It is interesting that this simple assumption so well matched the guano heaps in the cave. The first 2000 scats closely resemble the sheet of guano in Fig. 3, and the entire run looks very much like the 70 cm tall mound. The simulation makes it obvious that the thickest part of the guano pile should be sampled in order to obtain the best time resolution.

Fig. 4. Computer-generated mound of guano based on assumption that roosting bats will be distributed normally around a favored place on the ceiling. Given a frame width of 440 pixels, the initial roosting bat was assigned a position of 220 pixels with the density of the surrounding members decreasing with a standard deviation of 65 pixels. The computer drew 26,000 random scats, changing the color at 2000-scat intervals. See text for discussion.

2.2. Laboratory procedures

The 35 cm core of guano was opened in the lab, split longitudinally, and placed near the simple apparatus (Fig. 5) the author uses for recording core descriptions. The color of the fresh guano surface is 7.5 YR ranging from 2/1 to 3/2 (mahogany). It had no noticeable odor. Samples were taken at 1 cm intervals using cut-off lengths of plastic drinking straws as little core tubes. The portion with the feces was put in a labeled shell vial to

Fig. 3. Photograph of a thin carpet-like layer of guano under a favored roost. The deposit extends for several meters left to right, a meter from front to back, and it is several centimeters deep near its central axis. Light-colored patches are fungus.

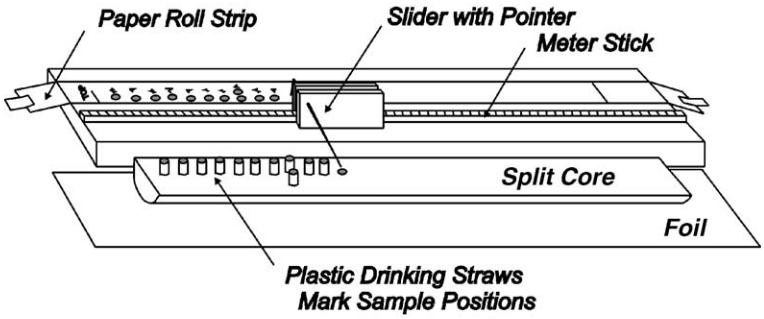

Fig. 5. A simple device for sampling and recording the characteristics of the guano core.

be processed, and another short piece of the plastic straw was pushed into the guano to record where the sample had been taken (Fig. 6). Eight additional samples were taken at 5-cm intervals for loss-on-ignition study. The core was re-wrapped and stored in a laboratory freezer.

2.3. Pollen

The plastic drinking straw in which the feces was collected is removed and discarded during the initial wash. Guano can be processed as if it were lake sediment using standard palynology procedures (Fægri and Iversen, 1964). However the 10% HCl, and l0% NaOH treatments are not needed; the guano is too acidic to contain much carbonate. Good recovery can be obtained by using an initial wash in warm water containing a trace of dish-washing detergent, followed by 5 to 10 min of acetolysis solution in a boiling-water bath, staining, and mounting in glycerine. The guano is quickly broken down in the detergent solution. Avoid concentrations that result in excessive suds, and if foam does occur, disperse it with a few drops of alcohol before centrifuging. The residue was mounted in glycerine in case an individual sample should require additional treatment, such as needing to employ 48% HF in the rare cases when siliceous residue caused a problem making slides. The glycerine can be washed away and replaced with silicone fluid (2000 cs) should it be desired.

2.4. Radiocarbon date

A fragment of guano from the base of the compressed core was submitted to the Center for Accelerator Mass Spectrometry at Lawrence Livermore National Laboratory. This yielded a radiocarbon age of 2810±40 yr B.P. (CAMS85667), corresponding to 2890 calibrated yr B.P. (Stuiver et al., 1998).

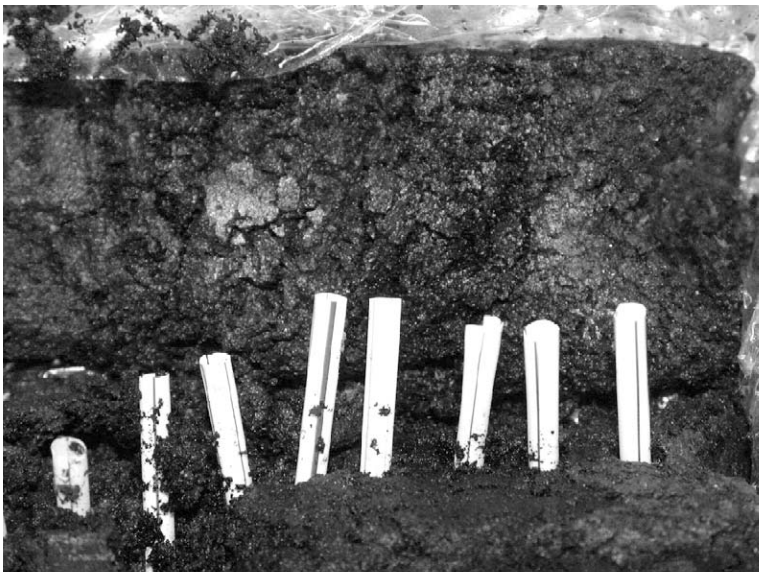

Fig. 6. Photograph of the upper 10 cm of the guano core. The drinking straws mark the position of the individual samples.

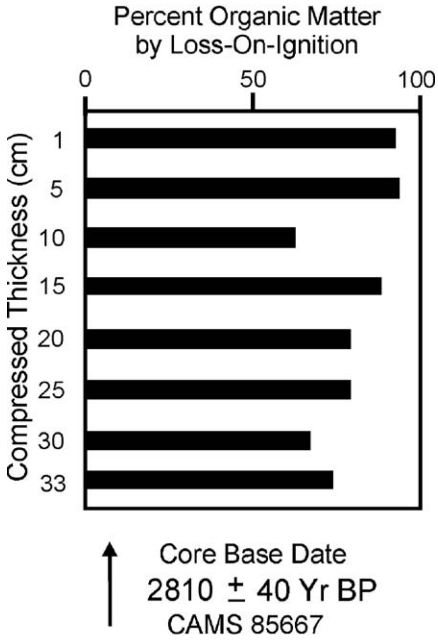

Fig. 7. Loss-on-ignition measurements on the compressed core. The AMS ^{14}C sample came from organic fragments at the base.

2.5. Organic matter

The eight samples taken for loss-on-ignition (LOI) study (Dean, 1974) were put in weighed ceramic crucibles, dried over night at 105 °C, and then fired at 550 °C for 2 h in a muffle oven. A sample's loss of weight on firing is considered to be its organic content; half of that weight usually is allotted to organic carbon. Fig. 7 illustrates a general loss of organic material in the deeper layers, probably as the result of microbial and other chemical action through time.

2.6. X-ray analysis

X-ray diffraction analysis allows minerals to be identified that are too small to identify under the microscope. The analysis of the mineral fraction of the guano was done in the Geology and Geophysics Department's S. W. Bailey X-ray Diffraction Laboratory using standard mineralogical techniques.

3. Results and discussion

3.1. Source of pollen in the guano of insectivorous bats

Fig. 8a shows a view of a guano sample after an initial wash in a detergent solution. Hair makes up a major component of this sample; it was probably ingested as the bat was grooming. A single pollen grain of *Carya* (hickory) is found near the top center. Hair is removed with further processing (Fig. 8b). A *Carya* grain is at the right, and a member of the Chenopodiaceae is at the left. Other grains in the same sample included *Pinus* (pine) (Fig. 8c) and *Quercus* (oak) (Fig. 8d). In fact, the great majority of the pollen grains in the guano were the same kind of wind-blown pollen that are found in cores from lake sediment.

The guano contains insect fragments, hair, pollen, and some mineral matter. Night-flying insects do not normally visit flowers for the pollen; many species do not eat during the flying phase of their life cycle, and those that do generally are nectar feeders. Although the insects are not after the pollen, they do fly through air that contains a lot of it, and the pollen and dust adheres to their bodies. The insects essentially act as living traps for airborne debris.

To verify this conclusion the author trapped moths and other insects that were attracted to a yard light at his home in Wisconsin. The insect bodies were processed as if they were regular palynological samples. In two samples collected during the month of June, pollen was found of *Acer* (maple), *Betula* (birch), *Juglans* (walnut), *Pinus* (pine), *Quercus* (oak), Poaceae (grass family), and *Ambrosia*-type (ragweed). The night-flying insects were not visiting any of these plants; they simply were flying through a pollen-laden environment.

The bats also are furry pollen traps. During grooming they ingest pollen and dust enmeshed in their fur, and this also is excreted. The mineral portion left over from one of the guano loss-on-ignition samples was determined by X-ray diffraction to be composed of the mineral quartz with some feldspar. Bats usually do not encounter these minerals at their ceiling roosts in limestone caverns. These minerals do compose the wind-blown siliceous dust in the atmosphere which coats both the bat and its prey.

An unusual event occurred many years ago that verifies that pollen is trapped in bat fur. Open petri dishes of glycerine are placed on high shelves in some pollen laboratories to check for pollen contamination from incoming air. Phenol crystals are added to prevent mold growth. The author once found a bat lying on one of the dishes; presumably it had been trapped in the laboratory, ingested some of the glycerine, and died. As a temporary measure the bat's body was placed in a sealed plastic bag in a freezer; and then it was forgotten. While working with the guano samples the author remembered the frozen bat, retrieved it and clipped off some of its body fur (Fig. 8e). The hairs (Fig. 8f) were very fine, and the ends were tapered to points rather than

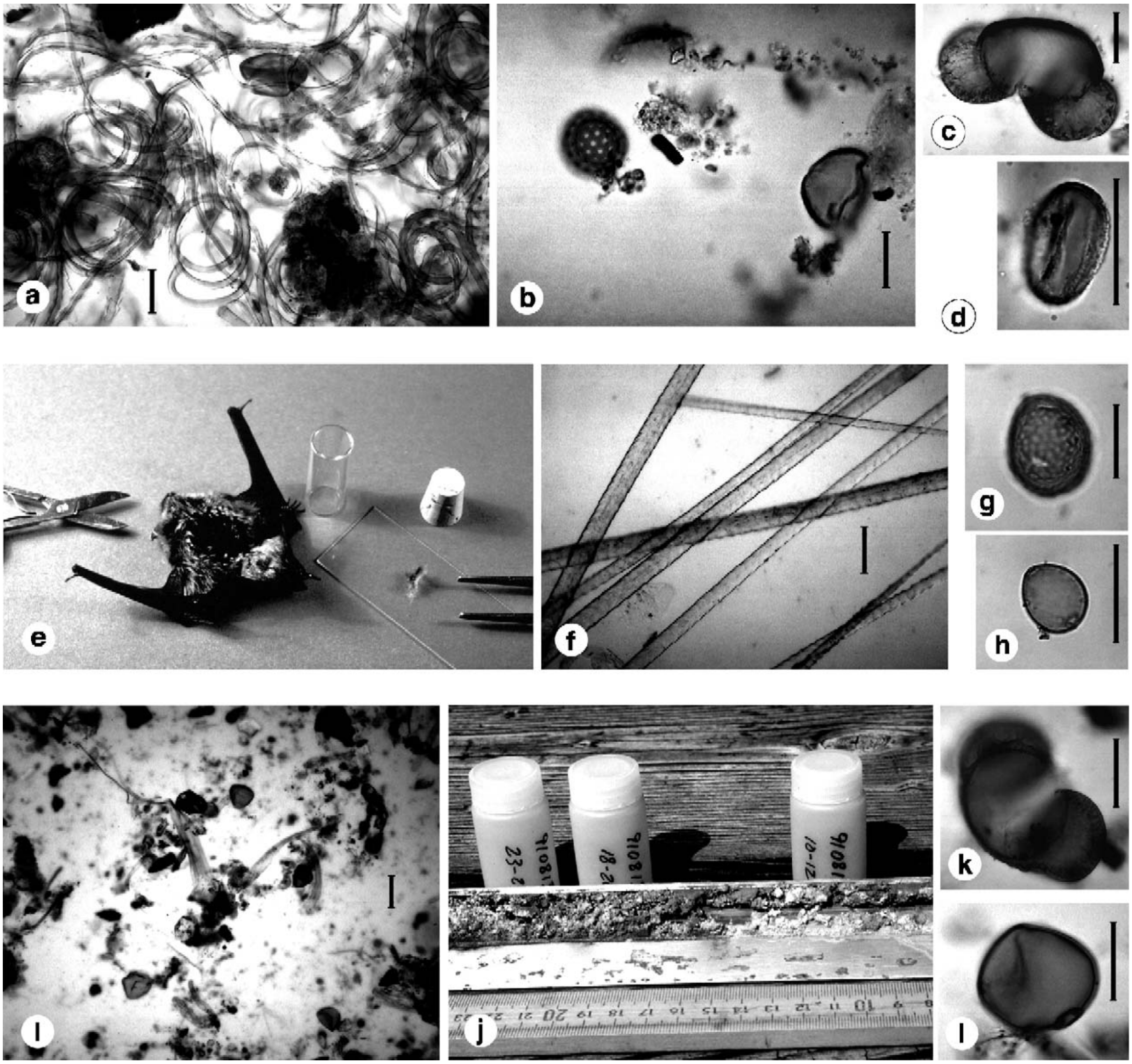

Fig. 8. All scale bars represent 25 μm. (a) Photomicrograph of detergent-washed guano containing a quantity of hair with two pollen grains: *Carya* (top center) and *Pinus* (left). (b) Processed sample with *Carya* (right) and Chenopodiaceae (left) pollen. (c) *Pinus* and (d) *Quercus* grains from the same sample as (b). (e) Photograph of a dead bat that provided a sample of hair. (f) Photomicrograph of the bat's hair. (g) *Ulmus* and (h) *Morus* pollen recovered from the bat's hair. (i) Low-power photomicrograph of insect debris recovered from the housing of an outdoor mercury-vapor lamp; a number of pollen grains are in the view. (j) Samples from old degraded guano; see text. (k) *Pinus* and (l) Poaceae pollen recovered from material shown in (j).

being broken off, suggesting the bat was very young. A clump of its fur was chemically processed, and during a scan of the slide *Ulmus* (elm) (Fig. 8g) and several grains of *Morus* (mulberry) (Fig. 8h) were found. These represent a sample of the atmospheric pollen in Wisconsin when the bat was alive.

Another chance opportunity to study the pollen carried by night-flying insects occurred when a mercury-vapor lamp burned out at the Geology Department in Madison, Wisconsin. The lamp is mounted above the freight dock, and it had been burning nightly for at least several years. The unit is protected from the elements by a glass cover. A rubber gasket separates the glass lens from its rectangular metal base, and there is a small gap at the lower part of the gasket to allow for changes of pressure. Thousands of flying insects had approached the light through that bright gasket tunnel, immolating themselves in the process. More than 100 ml of their dry bodies were collected when the bulb was replaced. Some of the debris is shown in Fig. 8i; a myriad of pollen grains are visible among the insect fragments. When a sample of this material was processed and counted, it compared very well with the upper sediment of Devils Lake in southern Wisconsin (Fig. 9). This suggests that a region's airborne pollen will be very similar, whether collected on a flying animal's body or deposited on a lake's water surface.

The American Pollen Database has a collection of surface samples of modern pollen from the United States and Canada: ftp://ftp.ncdc.noaa.gov/pub/data/paleo/pollen/asciifiles/modern/napd/ MODPOL.EXE (Maher, 2000) was used to calculate squared-chord dissimilarity coefficients (DC) between the mercury-vapor lamp sample and all the modern pollen sites in eastern North America. The 21 sites with DC values ≤0.150 are plotted on Fig. 10. Overpeck et al. (1985) consider sites with that degree of similarity to represent analog sites. The pollen on the insect bodies in the mercury-vapor lamp has the same general composition as the atmosphere through which they flew, and they have the same composition as the terrestrial pollen accumulating in the region's lakes and bogs.

It is difficult to over-emphasize the importance of this observation. Night-hunting insectivorous bats are eating these same insects. The bat guano contains a record of the pollen content of the atmosphere while they were feeding.

If the pollen in guano is mainly of atmospheric pollen that coats the bat and the food it eats, then the accumulation of guano may be considered analogous to lake sediment. Whereas lake sediments accumulate in a water-filled depression, insectivorous bats gather night-flying insects that are covered with atmospheric pollen, and the pollen accumulates stratigraphically with the guano. Pollen reaches lake sediment by many and varied routes either by wind or water. Much pollen is scavenged from the air by rain drops (McDonald, 1962). Wax-coated pollen is too buoyant to sink. While floating at the surface it serves as a nutrient source to many aquatic organisms whose excrement accumulates

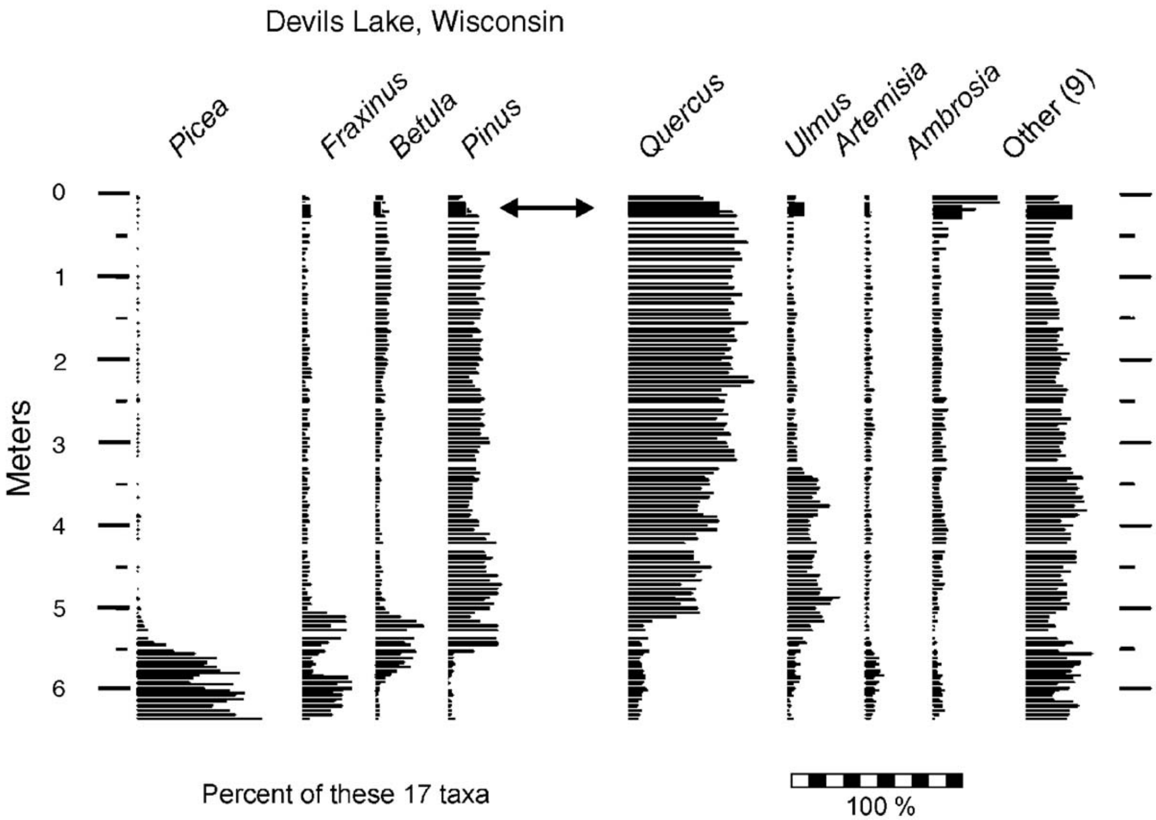

Fig. 9. The black bars (see arrows) show that the pollen spectrum recovered from the residue of the insects trapped in the mercury-vapor lamp closely match the upper sediment levels of Devils Lake, Sauk Co., Wisconsin (Maher, 1982).

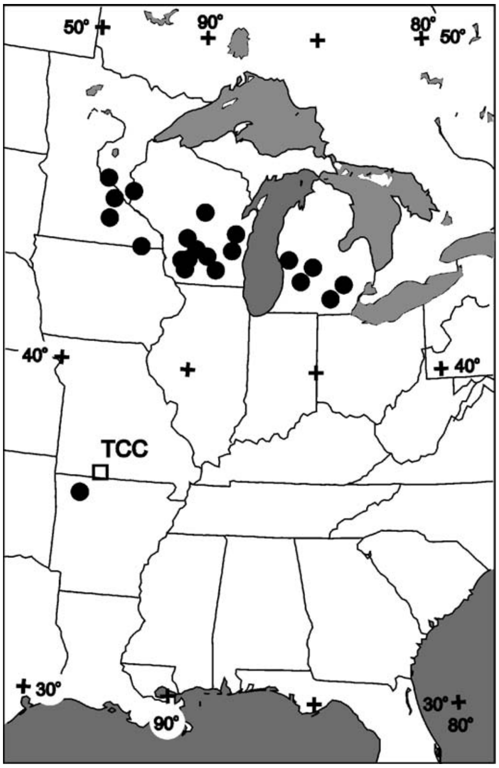

Fig. 10. The map shows the 21 sites in the American Pollen Database's modern pollen set whose squared-chord dissimilarity with the insect sample from the mercury-vapor lamp differ by ≤0.150. The square labeled "TCC" is Tumbling Creek Cave.

stratigraphically on the bottom. The pollen in both guano and water-laid sediments involve intermediary organisms in the depositional process. Both such deposits may yield paleoclimate information.

3.2. Guano palynology of Tumbling Creek Cave

Fig. 11 is a simple pollen diagram that allows the comparison of data from a number of different materials. The taxa and their display order are those used by Huber (1990). The upper ten samples are from four of Huber's sites in southeastern Missouri; they are included to show pollen spectra from moss and wetlands in southern Missouri. The first five samples from Buttonbush Bog are from moss polsters; the last is from surface mineral sediment. The two samples from Ozark Sink Pond are both from moss, whereas Gooseneck and Round Spring are from surface sediment. Huber indicates he counted about 400 grains per sample. He does not describe the pollen types included in his category "Other Pollen."

The TCC Moss sample was obtained in a wooded valley just outside the main entry to Tumbling Creek Cave. Its pollen composition should reflect the local vegetation in the forested valley and the farmed uplands; 407 grains were counted.

The guano sample was taken from several pollen samples from the top 2 to 3 cm of the core shown in

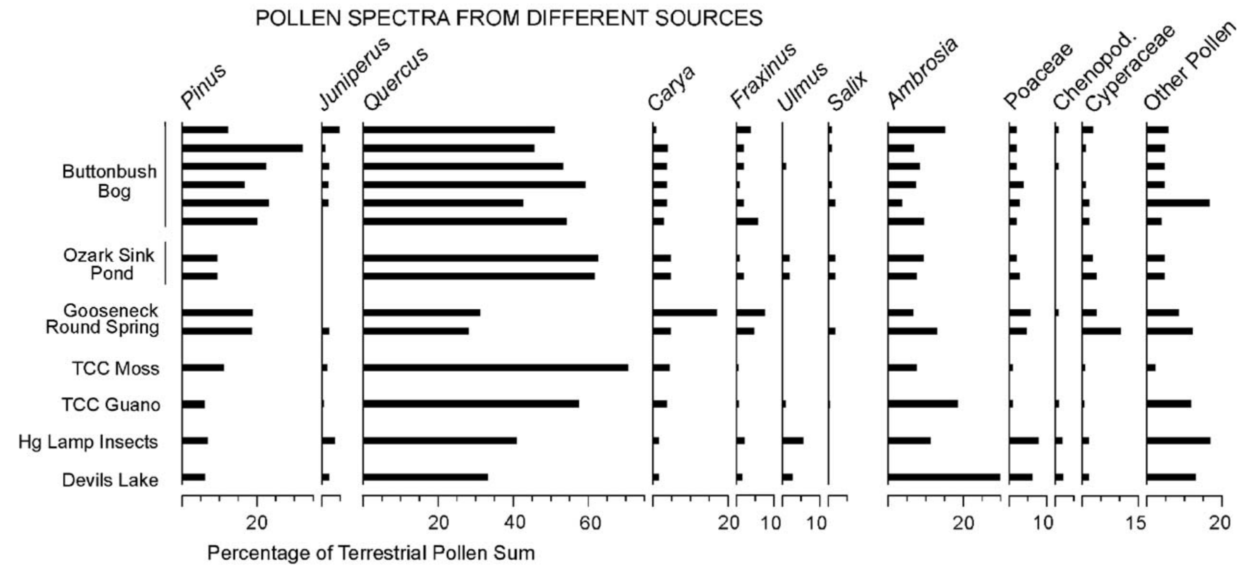

Fig. 11. The upper ten pollen spectra are from surface sample sites of Huber (1990) in southeast Missouri. The label "TTC Moss" indicates the pollen spectrum from moss by the entry to Tumbling Creek Cave. "TCC Guano" is the spectrum from the upper 2–3 cm of the compressed guano core. "Hg Lamp Insects" represents the pollen spectrum from the sample of dead insects trapped in the mercury-vapor lamp in Madison, Wisconsin. "Devils Lake" refers to the pollen spectrum from the combination of its upper three sediment levels; see also Fig. 9.

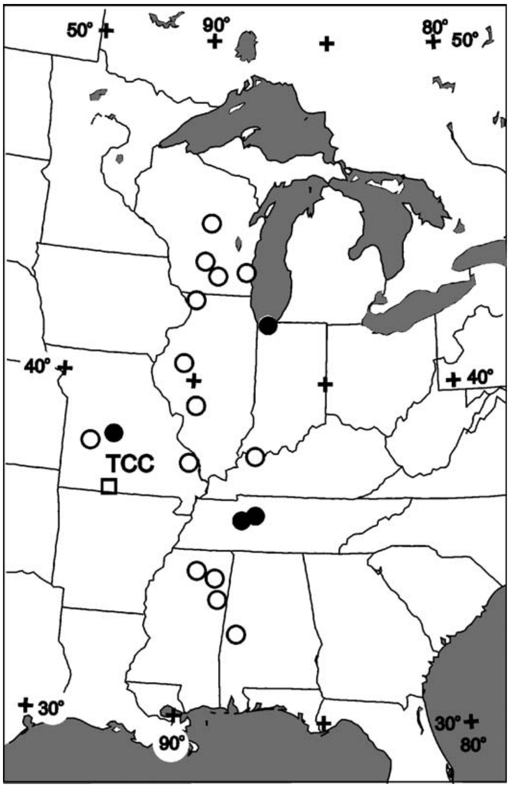

Fig. 12. The solid circles show the four sites in the National Climatic Data Center's M70 modern pollen data set whose squared-chord dissimilarity with the top of the guano core differ by ≤0.150. The site in Indiana is from Cole and Taylor (1995), the two sites in Tennessee are from Delcourt (1978); that in Missouri is from Peterson (1978). The open circles are 14 sites whose DC range between 0.150 and ≤0.200. The square labeled "TCC" is Tumbling Creek Cave.

Fig. 6. Its composition should represent the vegetation in the feeding range of the bats, thought to extend 40 km from the cave. Its proportions are based on a combined pollen sum of 815 grains. In this case the category "Other Pollen" comprises 12% of the guano's pollen. It includes in descending order of abundance: Other Echinate Compositae (35 grains), *Celtis* (29 grains), and *Acer* (7 grains). Pollen types with five or fewer grains were: *Plantago*, *Platanus*, *Liquidambar*, Apiaceae, Urticaceae, Brassicaceae, *Morus*, Polygonaceae, and Saxifragaceae. All of these types are commonly found in lake sediments. Seven grains could not be identified.

About 500 grains were counted in the sample of insects taken from the mercury-vapor lamp in Wisconsin. The Devils Lake percentages are calculated on 1900 grains from its upper three levels.

It would be impossible to pick any one of these 14 samples and state whether it came from outside surface samples, from guano, or from the dead night-flying insects from a mercury-vapor lamp. The figure illustrates the remarkable similarity of pollen deposition in the oak-hickory-pine region of the central U.S.

MODPOL.EXE was used to compare the pollen in the Tumbling Creek Cave guano with all the modern pollen samples in eastern North America. Fig. 12 shows the position of four analog sites that yielded squared-chord DC ≤0.150. There were 14 other sites whose DC range between >0.150 and ≤0.200. All 18 of these localities are found in the longitudinal belt between 87° and 95° W. The pollen in the guano represents the regional vegetation much like lake and bog sediments that are usually used for palynology.

The original core (Fig. 6) had been compressed when it had been extruded from the piston sampler. To avoid guano compaction, the author decided to use a side-wall cutter that is generally known as a "Russian peat sampler." The device is pushed to the desired depth in the deposit and then rotated, cutting a half-cylinder of material from the sediment and protecting it while the

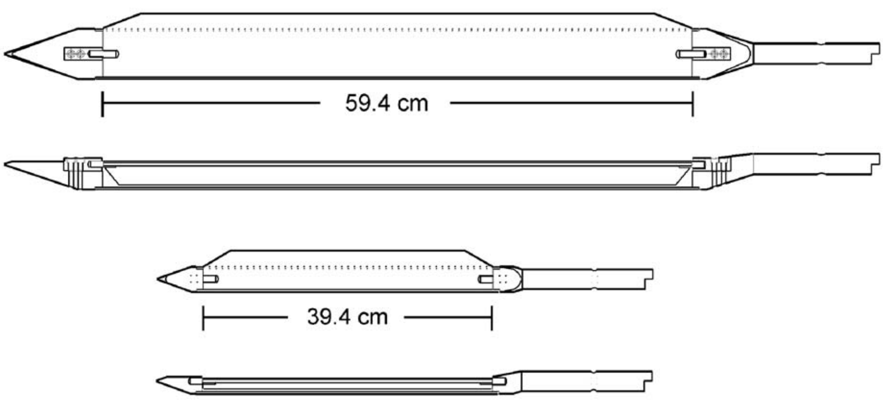

Fig. 13. Standard and miniature Russian peat samplers that were used for sampling bat guano. The small version is useful for penetrating into crevices.

Fig. 14. Russian sampler in the 70 cm high guano cone shown in Fig. 2.

sampler is withdrawn (Jowsey, 1966). Fig. 13 shows two such implements that were made in the Department of Geology's machine shop—a full-sized one, and a much smaller model that could penetrate into confined spaces. Fig. 14 shows the upper part of the sampler projecting from the 70 cm high guano cone from which the original core was obtained. It did succeed in obtaining material that retained its original low-density structure.

A larger deposit was identified from "Hibernation Hall" (Fig. 15). This passageway is used from time to time as a nursery by the maternal Grey Bat colony. To get some estimate of the thickness of the deposit, the smaller sampler was pushed into the deposit near its margin, penetrating about 2 m into the cave floor. The material recovered from that depth was a white crumbly mineral substance that represents a highly degraded guano (Fig. 8j). Pollen from this material contained well-preserved pine and grass grains (Fig. 8k–l).

X-ray diffraction analyses of this material recorded several different mineral substances that could be classified as hydrous phosphates (S. Welch, personal communication, 2002). The primary one was phase number 40-0041, $K_4H_5Al_3(PO_4)_6 \cdot 11H_20$; a reasonably good fit also occurred for the mineral Tananakite, phase number 29-0981, $H_6K_3Al_5(PO_4)_8 \cdot 18H_2O$. Several other mineral phases in the database matched at least a couple of the diffraction lines: phase 28-0082, potassium ammonium hydrogen phosphate $(NH_4, K)_3HP_2O_7 \cdot H_2O$; phase 35-

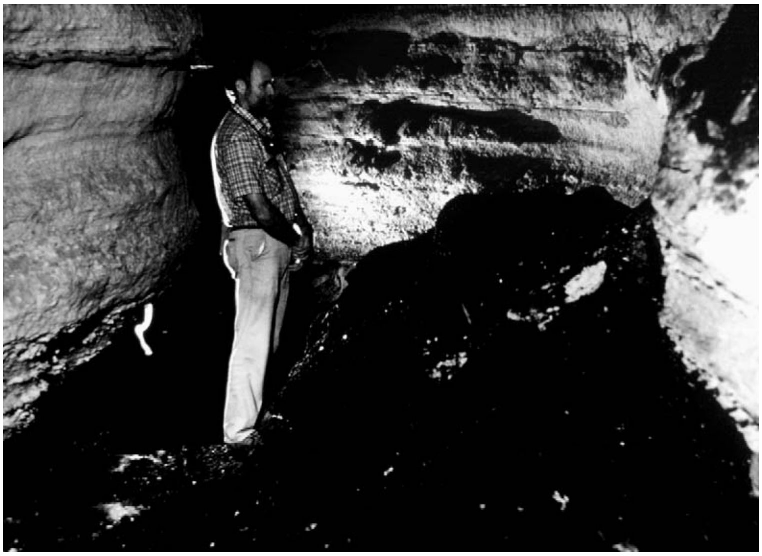

Fig. 15. The large deposit of guano in Hibernation Hall. See Fig. 2 for location.

0179, ammonium potassium hydrogen phosphate $(NH_4, K)_3HP_2O_7$; and phase 40-0404, aluminum ammonium hydroxide phosphate $Al_2NH_4OH(PO_4)_2 \cdot 2H_2O$. All the combinations suggest reaction between the constituents of the guano and the underlying dolomite with the latter being replaced by the acid decomposition products from the organic guano. Quattropani et al. (1999) also reported the growth of phosphate minerals in a reaction zone between guano and the cave wall.

The guano of insectivorous bats can contain pollen that matches rather well the atmospheric pollen that accumulates in the surface sediment of lakes and peat. Perhaps in areas without suitable lakes and bogs a researcher might reasonably turn to guano in attempting to reconstruct an area's vegetation and climate history. However the analyst should consider a number of problems before launching a project using bat guano.

Although the accumulation of guano may have analogies to accumulating lake sediment, there are distinct differences. While potential unconformities can be recognized in a lake sediment profile, it is more difficult to recognize hiatuses in piles of guano without numerous AMS ^{14}C measurements. Palynologists often assume—perhaps naively—that a few dates in a lake core will establish the intervening sediment accumulation rates. Lake sediment probably accumulates in deep basins as long as a lake exists, but is the deposition of guano continuous once it begins?

The same general hydrological processes of transportation and deposition might be assumed to exist whenever a lake contains water, but would that be true of guano if different species of bat occupied the cave at different times? If a hiatus were demonstrated in a guano deposit, it might be supposed that the bats were absent because of a change in climate or that disease or some other catastrophe had destroyed the colony. But could the lack of bats simply mean that their entry to the cave had been temporarily blocked, as often occurs, by a landslide?

An unusual phenomenon has puzzled the author about the compressed core from the 3000 year-old mound. A sample from near the core's base contained far less pollen than levels either above or below. Its pollen spectrum contained only a few different types rather than several dozen, and the two main grains (*Pinus* and *Carya*)—easy to identify even when preservation is poor—seemed small in size. The maximum breadth of 20 *Pinus* grains near the surface of the core were compared to the same measurement from 20 grains in the interval with bad preservation: mean and standard deviation was 75.7 ± 10.3 versus 36.7 ± 9.1 μm, respectively. The diameter of the *Carya* grains in the same two levels was also compared, although the author was able only to measure 12 from the lower one: 40.4 ± 5.4 versus 21.3 ± 4.1 μm. The linear dimensions of these two grain types were half as large in the level of poor presentation, and this means their volumes were only one-fourth as large. Pollen taxa processed in the same manner generally do not vary in size by a factor of two. It seems unlikely that flying insects attract only small pollen, nor is the author aware of pine or hickory species with pollen that small. Was this size change the result of microorganisms that process guano? Or were the bats occupying the cave at the earlier time able to absorb nutrients from the pollen exine? The author does not know the answer to this question nor has he found it discussed in the literature. But it does not seem merely to be a function of the deposit's age; the size of the much older pollen from the degraded guano under the Hibernation Hall (Fig. 8j–l) was well-preserved and of normal size.

Perhaps some of the questions of depositional continuity could be answered if vertical sections were excavated through a guano deposit. However, it would be difficult to maintain a stable cut surface because of guano's crumbly nature. The author has wondered if it might be feasible to collect guano by freezing it on to a metal probe like the "frozen finger" sampler that has been used to collect soft lake sediment (Wright, 1980). Fig. 16 is a sketch of a hypothetical wedge-shaped metal container, filled with

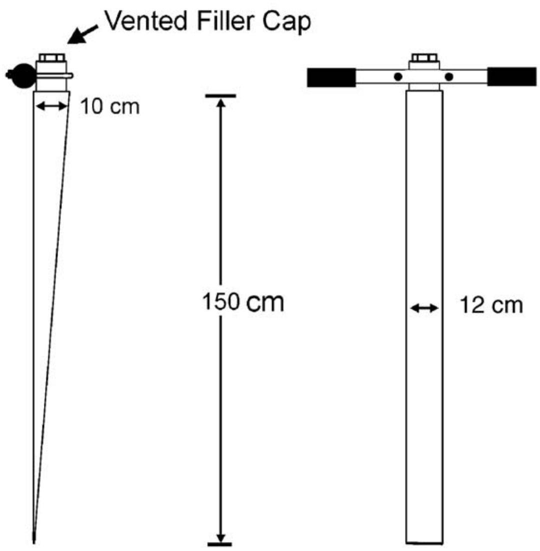

Fig. 16. A theoretical sampler for freezing guano to a flat metal surface. The device is cooled by a mixture of dry ice and normal butyl alcohol, and works on the same principle as the "Frozen Finger" sampler of Wright (1980). With the dimensions shown, the device would hold ~9.5 l of the working fluid. The wide-mouth filler tube needs a vented cap to allow the exit of CO_2 from the sublimating dry ice. For the sake of the bats, no spillage should be allowed.

dry ice and isopropyl alcohol, and pushed into the guano by its insulated handles. It would be left until the guano froze to the flat surface and then removed. (If water were poured along the flat surface before the sampler was pushed into the guano, a thin sheet of ice would separate the guano from the metal, and this would help later to release the guano sample.) The frigid mixture of dry ice and alcohol could then be drained into a suitable container. The sampler would be refilled with warm water to melt enough of the ice frozen to the metal to release the still-frozen guano on to an awaiting tray. The guano crust should then be kept frozen until it reached the laboratory. If several of these frozen samples were taken across a deposit of guano, it might be possible to recognize unconformities in the sediment; this would help choose where ^{14}C samples were needed.

There are always risks in life, but working with bat guano adds extra ones. Protective clothing, gloves, and respirators should be used by anyone disturbing guano. Caves may have confining passages and slippery slopes. But in spite of the problems, the information that can be extracted from guano about bats—their nutrition, parasites, habits, and the environment in which they lived—seems limited only by the imagination of the researcher.

Acknowledgements

I thank Thomas and Catherine Aley for allowing me to visit and take samples from Tumbling Creek Cave. James Miller and Jane Maher assisted with the sampling and photography in the cave. Susan Welch generously did the X-ray diffraction measurements and interpreted the results. A grant from the University of Wisconsin College of Letters and Science provided the funds for constructing the two Russian samplers that were used in this project. David Novotny fashioned the two ingenious samplers and the connecting rods needed for their use. I benefited from the help and advice of department artist, Mary Diman. The Lewis G. Weeks Bequest to my department supplied the funds for the radiocarbon date.

References

Clark Jr., D.R., Bagley, F.M., Johnson, W.W., 1988. Northern Alabama colonies of the endangered Gray Bat *Myotis grisescens*: organochlorine contamination and mortality. Biological Conservation 43, 213–226.

Clawson, R.L., 1992. Pesticide contamination of endangered gray bats and their prey in Boone, Franklin, and Camden Counties, Missouri. Transactions of the Missouri Academy of Science 25, 13–19.

Cole, K.L., Taylor, R.S., 1995. Past and current trends of change in a dune prairie/oak savanna reconstructed through a multiple-scale history. Journal of Vegetation Science 6, 399–410.

Coles, G.M., Gilbertson, D.D., Hunt, C.O., Jenkinson, R.D.S., 1989. Taphonomy and the palynology of cave deposits. Transactions of the British Cave Research Association, Cave Science 16, 83–89.

Dean Jr., W.E., 1974. Determination of carbonate and organic matter in calcareous sediments and sedimentary rocks by loss on ignition: comparison with other methods. Journal of Sedimentary Petrology 44, 242–248.

Delcourt, H.R., 1978. Late-Quaternary vegetation history of the eastern highland rim and adjacent Cumberland plateau of Tennessee. Ph.D. thesis, University of Minnesota, Minneapolis, Minnesota, USA.

Fægri, K., Iversen, J., 1964. Textbook of Pollen Analysis. Hafner Company Inc, New York.

Fenton, B.M., 1983. Just Bats. University of Toronto Press, Toronto, Canada.

Huber, J.K., 1990. Modern pollen rain in the southeast Missouri Ozarks. American Midland Naturalist 124, 263–268.

Jowsey, P.C., 1966. An improved peat sampler. New Phytologist 65, 245–248.

Küchler, A.W., 1964. Potential natural vegetation of the conterminous United States. New York American Geographical Society Special Publication no. 36. (Map with 116 p. manual).

Maher Jr., L.J., 1982. The palynology of Devils Lake, Sauk County, Wisconsin. In: Knox, J.C., Clayton, L., and Mickelson, D.M. (Eds.), Quaternary History of the Driftless Area. Wisconsin Geological and Natural History Survey Field Trip Guide Book Number 5, University of Wisconsin-Extension, Geological and Natural History Survey, Madison, WI, pp. 119–135.

Maher Jr., L.J., 2000. MODPOL.EXE: a tool for searching for modern analogs of Pleistocene pollen data. INQUA Sub-Commission on Data-Handling Methods. Newsletter, vol. 20. Internet Source: http://www.kv.geo.uu.se/inqua/news20/n20-ljm.htm.

McCracken, G.F., Gustin, M.K., 1987. Bat mom's nightmare. Natural History 96 (10), 66–73.

McDonald, J.E., 1962. Collection and washout of airborne pollens and spores by raindrops. Science 135, 435–437.

McMurray, D.N., Russell, L.H., 1982. Contribution of bats to the maintenance of *Histoplasma capsulatum* in a cave microfocus. American Journal of Tropical Medicine and Hygiene 31, 527–531.

Overpeck, J.T., Webb III, T., Prentice, I.C., 1985. Quantitative interpretation of fossil pollen spectra: dissimilarity coefficients and the method of modern analogs. Quaternary Research 23, 87–108.

Peterson, G.M., 1978. Pollen spectra from surface sediments of lakes and ponds in Kentucky, Illinois, and Missouri. American Midland Naturalist 100, 333–340.

Pons, A., Quézel, P., 1958. PremiΠres remarques sur l'étude palynologique d'un guano fossile du Hoggar. Compte Rendus Hebdomadaires des Séances de l'Académie des Sciences 246, 2290–2293.

Quattropani, L., Charlet, L., de Lumley, H., Menu, M., 1999. Early Palaeolithic bone diagenesis in the Arago Cave at Tautavel, France. Mineralogical Magazine 63, 801–812.

Sorley, D.L., Levin, M.L., Warren, J.W., Flynn, J.P.G., Gerstenblith, J., 1979. Bat-associated histoplasmosis in Maryland (USA) bridge workers. American Journal of Medicine 67, 623–626.

Stuiver, M., Reimer, P.J., Bard, E., Beck, J.W., Burr, G.S., Hughen, K.A., Kromer, B., McCormac, F.G., v.d. Plicht, J., Spurk, M., 1998. INTCAL98 Radiocarbon Age Calibration, 24,000–0 cal BP. Radiocarbon 40, 1041–1083.

Wright Jr., H.E., 1980. Cores of soft lake sediment. Boreas 9, 107–114.