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Levels of linear alkylbenzene sulfonates and nonylphenol in wild bat guano samples

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Introduction: Surfactants are substances that are commonly used in the industry, and they pollute the environment and negatively affect living organisms. In this article, we describe, for the first time, the exposure of wild bats to selected surfactants: nonylphenol (NP) and linear alkylbenzene sulfonates (LASs), including LAS C10, LAS C11, LAS C12, and LAS C13 (where "C" with a number indicates the length of the alkyl carbon chain).

Methods: The levels of the abovementioned compounds were evaluated in guano samples collected from various colonies of the greater mouse-eared bat (*Myotis myotis*) using liquid chromatography coupled with mass spectrometry (LC-MS/MS). Results: All surfactants studied were found in the guano samples. Mean levels of LASs (±SD) ranged from 87 ± 28.5 ng/g (median 75.1 ng/g) in the case of LAS C10 to 662 ± 227.2 ng/g (median 560.5 ng/g) in the case of LAS C12. The mean concentration of NP was 65.2 ± 74.9 ng/g (median 23.5 ng/g). Moreover, significant differences in surfactant levels were noted between particular bat colonies.

Discussion: The results showed that wild bats are highly exposed to LASs and NP, and the degree of this exposure shows clear intraregional differences. Moreover, the study demonstrated that guano samples serve as a valuable tool to assess bat exposure to these surfactants.

KEYWORDS

surfactants, wild animals, environmental pollution, LC-MS/MS, biomonitoring

Introduction

Surfactants are a large group of compounds characterized by a specific molecular structure. Their molecules are composed of two parts: a hydrophilic part soluble in a polar medium (with affinity to water) composed of one or several hydrocarbon chains and a hydrophobic part soluble in a nonpolar medium (with affinity to lipids) composed of one or several polar groups (Ying, 2006; Olkowska et al., 2011).

Depending on the charge of the hydrophilic part of the molecule, surfactants are divided into three groups: cationic, anionic, and nonionic substances (Olkowska et al., 2011). This molecular structure determines the properties of surfactants, such as their ability to adsorb on various surfaces and create micelles in solutions (Olkowska et al., 2014). Surfactants are commonly used worldwide in various branches of industry, agriculture, and households. It

is estimated that the annual production of these substances exceeds 13 million metric tons (Olkowska et al., 2014). The widespread use of surfactants leads to their significant presence in both municipal and industrial wastewater, which ultimately enters the natural environment (Villarreal-Reyes et al., 2022). Additionally, surfactants are considered effective bioindicators of the human impact on ecosystems (Hampel et al., 2012). Many surfactants are also toxic to living organisms (Jena et al., 2023). Linear alkylbenzene sulfonates (LASs) and nonylphenol (NP) are surfactants that require special attention due to their widespread use, presence in the natural environment, and toxicity to living organisms (Gejlsbjerg et al., 2001; Ying, 2006).

LASs and NP are found, among others, in laundry and personal care products, paints, pesticides, and plastics (Hampel et al., 2012; Jie et al., 2013). These substances have been observed in both marine and freshwater environments and sediments (Hampel et al., 2012; Lu et al., 2021; Nurulnadia et al., 2023). Moreover, LASs and NP have been found in aquatic organisms, both animals and plants, including fish, invertebrates, and algae (Sáez et al., 2000; Sáez et al., 2001). In contrast to the aquatic environment, knowledge of LASs and NP in the terrestrial environment is relatively scarce; however, it is known that they pollute soil, dust, and plants (Hu et al., 2021; de Bruin et al., 2019).

LASs and NP can adversely impact living organisms. It is known that these substances show developmental toxicity, affect the reproductive and digestive systems, disrupt immune processes, and change hematological parameters (Bakirel et al., 2005; Franco-Belussi et al., 2021; Gouda et al., 2022; Pan et al., 2024). Moreover, NP shows endocrine-disrupting properties and dysregulates the endocrine system, which in turn causes disturbances in the nervous, reproductive, and cardiovascular systems (Hu et al., 2022; Aliakbarzadeh et al., 2023; Ni et al., 2023).

Given their common presence in the environment and their toxic nature, monitoring the exposure of living organisms to LASs and NP is a crucial responsibility of environmental toxicology. Monitoring of the exposure of wild bio-indicative species, such as bats, is crucial (Timofieieva et al., 2023). This is because bats live in various regions on almost all continents, often close to human settlements, and they are very sensitive to all environmental changes, including industrialization, urbanization, and environmental pollution (Sotero et al., 2022).

In the case of protected wild animals, selecting an appropriate matrix is crucial in studies on exposure to harmful substances. One of the best matrices is feces/guano samples (Teampanpong and Duengkae, 2024). Such samples are relatively easy to collect without capturing animals, stressing them, or interfering with their lives. Simultaneously, guano is known to be a suitable matrix for studying the exposure to various organic pollutants (Gonkowski et al., 2023). Moreover, the our previous preliminary study performed on only a few samples (n = 5) showed that bat guano is also an appropriate matrix for studying the LASs and NP levels (Martin et al., 2023). However, to the best of our knowledge, there have been no studies to date on the exposure of wild terrestrial animals to these substances.

The aim of the current study was to assess the levels of selected LASs (LAS C10, LAS C11, LAS C12, and LAS C13, where "C" with a number indicates the length of the alkyl carbon chain) and NP in guano samples collected from colonies of the greater mouse-eared bat (*Myotis myotis*)—the most common species of insectivorous bat in Poland. The results of the present research allow a better understanding of the problem of environmental pollution by LASs and NP and their influence on wild animals.

Materials and methods

Reagents

The commercial LAS mixture comprised C10 (12.3%), C11 (32.1%), C12 (30.8%), and C13 (23.4%) and was obtained from CEPSA (Madrid, Spain). A technical-grade mixture of 4-NP isomers was sourced from Sigma-Aldrich (Milwaukee, WI, United States). Isotopically labeled BPA, specifically IS (BPA-d16) from Cambridge Isotope Laboratories (Tewksbury, MA, United States), was used as an internal standard. Water and methanol, both analytical grade, were obtained from Romil Ltd. (Barcelona, Spain). Octadecyl-functionalized silica (C18) was provided by Sigma-Aldrich (Steinheim, Germany); formic acid and ammonium acetate were supplied by PanReac (Barcelona, Spain). Individual stock standard solutions were prepared at a concentration of 1,000 mg/L in methanol and stored at −18 °C. Working solutions were created by diluting the stock standard solution in methanol.

Sample collection

Samples were collected in August and September 2022. The study involved four summer (nursery) colonies of the greater mouse-eared bat (*Myotis myotis*), which are situated in various parts of Poland, in areas that differ in terms of urbanization and industrialization (Supplementary Table S1). Bat guano sample collection was performed according to the method described previously by Gonkowski et al. (2024), in which samples were collected into flat glass containers placed on the floor in the area where the bat colony was located. The containers were removed after 48 h, and the guano samples were transferred to sterile glass vials and frozen at −20 °C. The samples were stored at this temperature until further analysis.

According to the Act for the Protection of Animals for Scientific or Educational Purposes of 15 January 2015 (Journal of Laws 2015, No. 266) in force in Poland, the present research did not require the consent of the Ethics Committee for Animal Experiments because sample collection did not require direct contact with animals, was stress-free, and did not cause any negative impact on their welfare.

Sample treatment

Preparation of the bat guano samples was performed according to the method developed by Martin et al. (2023). The samples were lyophilized, homogenized, ground into powder and sieved (<100 μm). A total of 1 g of the sample was put into a 12-mL glass tube and spiked with 100 μL of a methanol solution of BP-d16 (250 ng/mL). The samples were then subjected to extraction by ultrasonic solvent extraction using 7 mL of methanol (0.5% v/v, formic acid) for 5 min and then centrifuged (4,050 g, 5 min). This procedure was repeated three times, and the resulting supernatants

were combined. Matrix interferences were removed by a cleaning dispersive solid-phase extraction procedure using 0.3 g of C18 sorbent, followed by shaking for 2 min and centrifugation for 5 min (4050 g). The solvent was evaporated under a nitrogen stream at room temperature to a dry residue and reconstituted in 0.25 mL of a mixture of methanol and water (50:50 v/v) and filtered through a 0.22-μm nylon filter. A total of 10 μL of the extract was injected into a liquid chromatography-mass spectrometry (LC-MS/MS) system.

Liquid chromatography coupled with mass spectrometry

An Agilent 1260 Infinity II instrument (Agilent, Santa Clara, CA, United States) was used in the present study. Analyses were performed according to the method described by Martin et al. (2023). A HALO C18 Rapid Resolution analytical column (50 × 4.6 mm i.d., 2.7 μm) (Teknokroma, Barcelona, Spain) was used for separation. The chromatographic process was made up in a gradient manner with methanol (solvent A) and a buffer solution of acetic acid/ammonium acetate (pH 4.4) (solvent B). The elution program was performed as follows: 0–14 min, linear gradient from 28% to 70% of solvent A, increased to 80% of A in 5 min and to 100% of A in 6 min, and held for 2 min. The flow rate was 0.6 mL/min.

A 6495 triple quadrupole mass spectrometer with an electrospray ionization source operating in the negative mode was coupled to the LC system. Two multiple-reaction-monitoring (MRM) transitions, for identification and quantification purposes, were used for each substance that was analyzed. The analytical determination parameters are presented in Supplementary Table S2.

Validation method

The method was validated using the matrix effect, linearity, sensitivity, accuracy, and specificity. Supplementary Table S3 contains a summary of the main analytical features of the method. The method was thoroughly validated, and all parameters were described by Martin et al. (2023). A matrix-matched calibration was used to overcome the matrix effect. Certified reference materials are not commercially available for the determination of the selected compounds in guano. Therefore, spiked samples were prepared. Commercial guano ("SuperGuano" from Top Crop, www.topcropfert.com) samples were enriched with eight different concentrations of the compounds analyzed, including 25, 50, 100, 500, 750, 1,000, 1,250, and 1,500 ng/g dry weight (dw) for LASs and 0.01, 0.05, 0. 10, 2.00, 5.00, 25.0, 50.0, and 100 ng/g dw for NP. A calibration curve was built from the method quantification limits (MQLs) to 1,500 ng/g dw in the case of LASs and from the MQLs to 100 ng/g dw in the case of NP.

The method detection limits (MDLs) and MQLs were calculated as the concentrations of each compound corresponding to signal-to-noise ratios of 3:1 and 10:1, respectively, using guano-spiked samples at low concentrations.

The method's accuracy (both trueness and precision) was assessed with commercial guano samples enriched at three concentrations: 100, 500, and 1,000 ng/g dw for LASs, and 5, 25, and 50 ng/g dw for NP, with each sample tested in triplicate. Accuracy was evaluated by a recovery control throughout the entire procedure, which included extraction from the matrix, d-SPE, and concentration steps. Precision was expressed as the relative standard deviation (%, RSD) of the measurements taken on different days.

Statistical analysis

The statistical analysis was performed with GraphPad Prism version 9.2.0 (GraphPad Software, San Diego, CA, United States). Data were analyzed using descriptive statistics. Moreover, due to the non-normal distribution of the data in some bat colonies, as confirmed by the Shapiro–Wilk test, a nonparametric Kruskal–Wallis test was used to compare levels of compounds in specific bat colonies. Differences were considered statistically significant at $p < 0.05$. NP concentrations lower than the MQL were included in the statistics as MQL/2 (1 ng/g).

Results

All substances studied were found in each guano sample included in the study (Table 1; Supplementary Table S4). Significant differences in the concentrations of the substances studied were also observed between particular guano samples collected from the same colony (Supplementary Table S4). The highest concentrations were observed in the case of LAS C12. Its levels ranged from 436 ng/g to 1,254 ng/g, with a median of 560.5 ng/g and a mean (±SD) of 662 ± 227.2 ng/g. The second in terms of concentrations was LAS C11, whose levels ranged from 139 to 345 ng/g (median 165 ng/g, mean 195.5 ± 59.4). LAS C13 and LAS C10 were found at lower concentrations in the studied guano samples. The levels of LAS C13 ranged from 64.8 mg/g to 198 ng/g, with a median of 79.4 ng/g and a mean of 94.3 ± 35.7 ng/g. In turn, levels of LAS C10 ranged from 55.3 ng/g to 160 ng/g, with a median of 75.1 ng/g and a mean of 87 ± 28.5 ng/g. Among the substances included in the study, NP was found at the lowest levels. In 35% of samples studied, NP levels were below the MQL (<2.00 ng/g), and the highest level of this substance amounted to 223 ng/g. The mean concentration level of NP was 65.2 ± 74.9 ng/g, and the median was 23.5 ng/g. Cumulative data on the presence of LASs and NP are shown in Table 1.

During this study, differences in the levels of the studied substances were visible between particular bat colonies (Figure 1; Supplementary Table S5).Concentrations of all LASs studied were higher in colony no. 1 than in the other colonies (Supplementary Table S5). The median concentration of LAS C10 in colony no. 1 was 133.5 ng/g and was significantly higher ($p < 0.05$) than that of other colonies (74.7 ng/g in colony no. 2, 76.5 ng/g in colony no. 3, and 62.7 ng/g in colony no. 4) (Figure 1A). The differences between colonies 2, 3, and 4 were not statistically significant. A similar situation was noted in the case of LAS C11. Its median level in colony no. 1 was 296.5 ng/g and was statistically significantly higher than the values noted in colony no. 2 (159 ng/g), colony no. 3 (165 ng/g), and colony no. 4 (161 ng/g), which, in turn, did not

TABLE 1 Concentration (ng/g) and frequency of detection (%) of LASs and NP in the analyzed guano samples (n = 40)—cumulative data.

Compound	Range	Arithmetic mean ± SD	Geometric mean	25% percentile	Median	75% percentile	% Samples > MQL
LAS C10	55.3–160	87 ± 28.5	83.2	65.8	75.1	107.3	100
LAS C11	139–345	195.5 ± 59.4	188.4	159.3	165	217	100
LAS C12	436–1,254	662 ± 227.2	630.4	495.8	560.5	796.8	100
LAS C13	64.8–198	94.3 ± 35.7	89.1	69.3	79.4	103.8	100
NP	<MQL–223	65.2 ± 74.9	14.4	<MQL	23.5	123.5	65

statistically differ from each other (Figure 1B). Samples taken from colony no. 1 also showed the highest median concentration of LAS C12, which was 1,021 ng/g. In colonies 2, 3, and 4, the median levels of this substance were 558 ng/g, 568.5 ng/g, and 458.5 ng/g, respectively. Statistically significant differences were noted between colony no. 1 and the other colonies and between colonies 3 and 4 (Figure 1C). In turn, the median levels of LAS C13 were 146 ng/g in colony no. 1, 73.9 ng/g in colony no. 2, 79.8 ng/g in colony no. 3, and 67.4 ng/g in colony no. 4. Statistically significant differences were found between colonies 1 and 2 and between colonies 1 and 4 (Figure 1D).

The distribution of median concentrations of NP, in particular in bat colonies, was completely different from levels of LASs (Figure 1E). In colony no. 1, the median value of NP was the lowest and was below the MQL. However, it was calculated for statistical purposes (values of NP below the MQL were included in the statistics as MQL/2). The median value calculated in this way was 1 ng/g. In colony no. 2, the median concentration of NP was higher and amounted to 10.5 ng/g, but there were no statistically significant differences between colonies 1 and 2. Significantly higher medians of NP were observed in colonies 3 and 4, where these values were 101.8 ng/g and 118 ng/g, respectively. The differences between colony no. 1 and colonies 3 and 4 were statistically significant. Moreover, statistically significant differences were also noted between colonies 2 and 4 (Figure 1E).

Data concerning the levels of surfactants studied in bat guano collected from specific colonies are summarized in Supplementary Table S5.

Discussion

The present investigation clearly demonstrates that wild bats are extensively exposed to LASs and NP. This exposure is linked to the widespread use of these substances in many everyday products and their subsequent penetration into the environment (Hampel et al., 2012). Because this work is the first to describe the presence of LASs and NP in the feces/guano of terrestrial animals, it is impossible to compare the present results with previous observations on humans and aquatic organisms (Pirard et al., 2012; Lu et al., 2021). Guano is a completely different matrix than the bodies of aquatic organisms or human urine and blood serum, in which the described surfactants have been detected thus far (Jiang et al., 2022; Xu et al., 2022).

Previous studies have demonstrated that LAS and NP concentrations vary widely by region (Pirard et al., 2012; Li X. et al., 2013). Regional differences in NP and LAS concentrations result from various factors, including urbanization and industrialization, human population density, agricultural development, and wastewater treatment (Xu et al., 2022). Moreover, it is known that the levels of surfactants in the environment can change seasonally. For example, it has been shown that levels of NP in the wastewater are higher in winter than in other seasons, which is explained by the likely influence of temperature and insolation on the biological and/or abiological transformation of this compound (Gao et al., 2017). Seasonal differences in NP levels have also been noted in surface water. Interestingly, the majority of studies have described higher levels of NP in rivers and lakes in the summer (Li et al., 2004; Li Z. et al., 2013; Wu et al., 2013). Seasonal differences have also been found in the levels of LASs in surface water. For example, Wang et al. (2010) found that LAS levels were the lowest in the fall compared to summer or winter. It is likely that seasonal differences in the levels of surfactants in the surface water depend on many factors, including seasonal human activity, agriculture, insolation, and water microorganisms; however, temperature appears to be the major factor influencing the biodegradation of these substances (Wang et al., 2010; Wu et al., 2013; Li Z. et al., 2013).

Regional variations in the levels of surfactants studied in the environment were confirmed by the present study, which revealed differences in NP and LAS levels between particular bat colonies. The reasons for these differences are difficult to explain without comprehensive studies on surfactants in the environment. The highest levels of LASs were noted in bat colony no. 1, which is located in rural areas, which suggests that exposure to LASs is connected with agriculture, that is, the use of pesticides and the fertilization of the fields with sewage sludge, and/or the contamination of water used for field irrigation. High LAS levels may also result from the rather low percentage (approximately 50%) of residential buildings equipped with sewage systems in rural regions of Poland (Supplementary Table S1), which increases the risk of untreated sewage entering the environment. Higher levels of LASs in bat guano from colony no. 1 align with prior studies that have documented the presence of LASs in rural regions (Xu et al., 2022).

On the other hand, in colony no. 2, which is also located in a rural region, LAS levels were clearly lower than in colony no. 1. This difference may result from the fact that colony no. 2 is located in a region where the human population density is less than half as much as in the area where colony no. 1 is located (Supplementary Table S1). Moreover, the area where colony no. 1 is located is a tourist

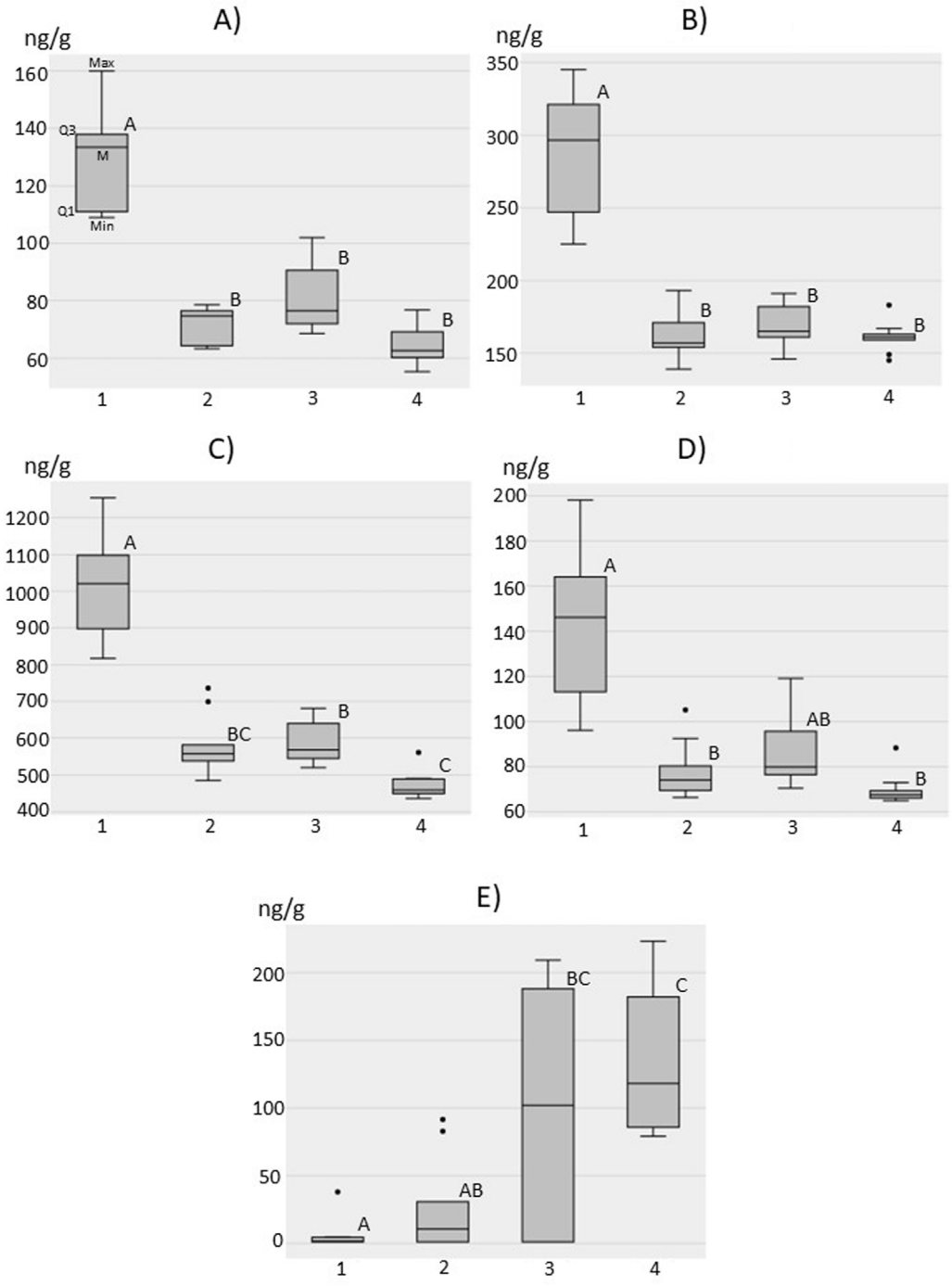

FIGURE 1
Levels of LAS C10 **(A)**, LAS C11 **(B)**, LAS C12 **(C)**, LAS C13 **(D)**, and NP **(E)** in bat colonies (no. 1, no. 2, no. 3, and no. 4). Statistically significant differences ($p < 0.05$) are marked with different letters, and values not showing such differences are marked with the same letter. M, median; Max, maximum value; Min, minimum value; Q1, quartile 1; Q3, quartile 3. Black dots represent outliers (values lying beyond the upper boundary of Q3 +1.5 IQR and the lower boundary of Q1-1.5 IQR, where IQR = Q3–Q1).

region connected with higher human activity, which may affect LAS concentrations in the environment (Okbah et al., 2013). Interestingly, in the present study, high levels of LASs were not positively correlated with high levels of NP. The highest levels of NP were observed in colonies 3 and 4, which are located in an urban environment with a relatively high human population density. These results strongly suggest that the sources of NP and LASs in the environment are different and that various factors influence environmental pollution with these compounds.

Another interesting topic is the sources of bat exposure to LASs and NP. However, without comprehensive environmental studies, the sources of this exposure cannot be clearly determined and

remain speculative. Water is likely the primary source of bat exposure to LAS and NP, as previous studies have identified these surfactants in surface water (Zgoła-Grześkowiak et al., 2009). Another source of bat exposure to surfactants may be connected with food. Greater mouse-eared bats are insectivorous, and previous studies have reported that both NP and LASs may accumulate in the bodies of insects (Huerta et al., 2015). Although previous studies have focused on aquatic insects, and greater mouse-eared bats feed mainly on beetles from the carabid family, it is also known that the described surfactants also pollute soil and affect the insects living there (Sørensen and Holmstrup, 2005). This information suggests that ground insects may also contain surfactants. It cannot be excluded that bats are also exposed to surfactants through the skin and respiratory systems, especially since greater mouse-eared bats establish colonies in the immediate vicinity of human habitations.

Another important issue is whether the levels of surfactants observed in bat guano pose a danger to bats. This study does not provide a clear answer to this question. There are no studies on the surfactant metabolism in bats, so it is unclear how LAS and NP concentrations in guano reflect the levels of these compounds in bat tissues. Nevertheless, previous studies, primarily conducted on experimental animals, have reported that even low doses of surfactants, particularly NP, can have a negative impact on living organisms (Xie et al., 2019). It is known that long-term exposure to even relatively low environmental doses of NP (below the no observed adverse effect level'—NOAEL) can result in changes in the liver and adipose tissue (Ribeiro et al., 2023), and the toxic effect on the liver is likely connected to NP-induced oxidative stress reactions (Kazemi et al., 2016). It is also known that low doses of NP may affect the nervous system, which may, in turn, induce depressive behavior and cause disturbances in memory performance, learning, general activity, emotionality, and sexual behavior (Kawaguchi et al., 2015; Capela et al., 2018; Pan et al., 2024). Moreover, NP exposure may affect female and male reproductive systems (Gong and Han, 2006; Yu et al., 2011; Goktepe et al., 2023). It is also a relatively well-known toxic factor that negatively affects the development of fetuses and young organisms (Chamard-Jovenin et al., 2017; Yan et al., 2025). Although the toxicity of LASs is relatively well understood, the majority of previous studies concern aquatic organisms, such as fish and invertebrates. These studies have shown that aquatic organisms are highly sensitive to the toxic effects of LASs (Jackson et al., 2016). It is known that LASs, even in relatively low doses, negatively affect, among others, the testes (where LASs promote inflammatory processes and cause a delay in sexual maturity), liver (where LASs cause the vacuolization of hepatocytes, inhibit the activity of several enzymes, and affect iron storage), and heart (where LASs cause alterations in heart function) (Gupta et al., 1989; Kumar et al., 2007; Jones-Costa et al., 2018; Franco-Belussi et al., 2021). Moreover, LASs cause changes in hematological parameters (Gouda et al., 2022), induce oxidative stress reactions and intensify apoptosis (Shukla and Trivedi, 2018), and show genotoxic effects (Zhao et al., 2015). Given the above facts, it can be inferred that the levels of LASs and NP noted in the present study may influence bat health. This is even more likely because bats are exposed to many different environmental pollutants, which often have a synergistic effect (Gonkowski et al., 2023). Therefore, the primary biological implication of the present study is that the content of surfactants in the guano of bats indicates their exposure to these compounds. In turn, such long-term exposure, even to relatively low environmental doses, may result in alterations in the functioning of various internal systems. These alterations may not cause any clinical symptoms in specific individuals but may affect the life expectancy of animals and even the size of the entire population (e.g., through reproductive disorders). Therefore, eventually, exposure to surfactants and other environmental pollutants should be taken into account in activities aimed at protecting bats. Another implication of this study is that bat guano may be a potential source of human exposure to surfactants. Bat nursery (summer) colonies are located in the immediate vicinity of human settlements (in the attics of schools, churches, and houses), and humans may come into contact with guano. Those who clean areas with bat colonies after the summer season (when bats move to winter colonies) and carry out maintenance work there are particularly vulnerable to inhalation exposure to substances contained in guano.

It should be noted that this study has some limitations. One of them is that surfactant levels have been analyzed only in bat guano. No analyses were carried out to determine the levels of these substances in surface waters located in the vicinity of bat colonies, dust from areas where the colonies were located, soil, or insects. The lack of such studies makes it impossible to determine the main sources of bat exposure to LASs and NP. Therefore, determining these sources requires further comprehensive research. Another limitation is connected with the matrix used in the study. Analysis of LASs and NP in guano collected from the colony makes it impossible to determine the exposure of individual animals. Moreover, as the study was conducted only in one season, it was not possible to determine whether there were seasonal differences in the exposure of bats to the surfactants that were studied. On the other hand, the choice of the time of year for collecting the samples was not accidental. Summer (nursery) colonies of greater mouse-eared bats are typically found in the attics of various buildings. Humans need to access attics to carry out renovations, and these spaces are cleaned at the end of summer, when the bats move to their winter colonies. During winter, bats hibernate in isolated places, and it is crucial not to disturb them because awakening a bat from hibernation may pose a threat to its life. Moreover, it can be assumed that due to hibernation, bat exposure to environmental pollutants in winter is marginal.

In summary, the present study has shown that greater mouse-eared bats are exposed to LASs and NP. This is the first study on the exposure of wild terrestrial mammals to these surfactants. LASs and NP were found in all guano samples included in this research, which confirms the common significant pollution of the natural environment by these substances. The study showed the presence of LASs and NP in samples collected from bat colonies located in various regions, both in rural and urban areas. Differences between particular bat colonies suggest that the presence of LASs and NP in the environment depends on local factors.

Moreover, the present study showed that guano/feces is a suitable matrix to evaluate the exposure of wild animals to surfactants polluting the environment. This matrix appears to be particularly useful for studies on protected animals because guano/feces sample collection does not require capturing and stressing the animals. However, several issues related to the exposure of wild bats to surfactants remain unclear. The most important of these include

determining the exact sources of bat exposure to surfactants, the correlations between surfactant concentrations in guano and tissues, and surfactant metabolism in the bats. Therefore, further studies are needed to address these issues.

Data availability statement

The original contributions presented in this study are included in the article/Supplementary Material; further inquiries can be directed to the corresponding author.

Ethics statement

Ethical approval was not required for this study involving animals in accordance with the local legislation and institutional requirements because according to the Act for the Protection of Animals for Scientific or Educational Purposes of 15 January 2015 (Journal of Laws 2015, No. 266) in force in Poland, the present research did not require consent from the Ethics Committee for Animal Experiments because sample collection did not require direct contact with animals, was stress free, and did not involve any negative impact on their welfare.

Author contributions

SG: Conceptualization, Formal Analysis, Resources, Supervision, Writing – original draft. JM: Investigation, Validation, Writing – review and editing. IA: Investigation, Writing – review and editing. JS: Investigation, Writing – review and editing. EA: Investigation, Writing – review and editing. WS: Investigation, Writing – review and editing. LR: Conceptualization, Writing – review and editing.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fenvs.2025.1607016/full#supplementary-material

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