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Emission of odor pollutants and variation in microbial community during the initial decomposition stage of municipal biowaste

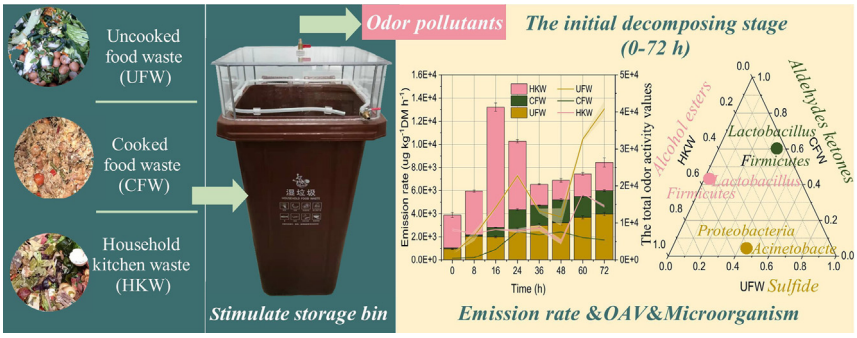
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HIGHLIGHTS

- The highest emission was observed in household kitchen waste with alcohol esters.
- The highest total odor activity values was observed in uncooked food waste.
- *Lactobacillus* was the dominant genus in household kitchen waste and cooked food waste.
- *Acinetobacter* was the dominant genus in uncooked food waste.
- The variation of odorants composition kept consistent with microbial composition.

GRAPHICAL ABSTRACT



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ABSTRACT

Odor pollution often occurs in the initial decomposition stage of municipal biowaste, including throwing/collection and transportation. However, this aspect of odor impact from municipal biowaste has not been well studied. In this study, a practical dustbin (120 L) equipped with flux chamber and filled with three types of municipal biowaste was used to simulate garbage storage conditions. The result indicated that the emission rate of odor pollutants for uncooked food waste (UFW) represented a nearly linear growth trend, reaching the maximum (3963 ± 149 μg kg^{-1} DM h^{-1}) at 72 h. Cooked food waste (CFW) increased rapidly from 8 h to 24 h, and then remain fluctuated, reached the maximum (2026 ± 77 μg kg^{-1} DM h^{-1}) at 72 h. Comparatively, household kitchen waste (HKW) reached the maximum emission rate ($10{,}396 \pm 363$ μg kg^{-1} DM h^{-1}) at 16 h. Sulfide and aldehydes ketones were identified as dominant odor contributor to UFW and CFW, respectively. While aldehydes ketones and sulfides were both dominant odor contributor to HKW. Moreover, the microbial diversity analysis suggests that Acinetobacter was the dominant genus in UFW, and Lactobacillus was the dominant genus in CFW and HKW. In addition, it was evident that each odorous pollutant was significantly associated with two or more bacterial genera, and most bacterial genera such as Acinetobacter, were also significantly associated with multiple odorous pollutants. The variation of odorants composition kept consistent with microbial composition. The present study could provide essential evidence for a comprehensive understanding of odorant generation in the initial decomposition stage of municipal biowaste. It could contribute to setting out strategies for odor control and abatement in municipal biowaste management systems.

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1. Introduction

Municipal biowaste, including restaurant food waste (FW) and household kitchen waste (HKW), is principally recognised as being related to odorants nuisance complaints due to its high organic portion as well as high moisture, which could provide favourable conditions for the emission of odorants during the decomposing stage (He et al., 2020; Liu et al., 2020). In addition to odor nuisance, odorous compounds could also have adverse effects on the surrounding environment (Nicell, 2009). Furthermore, prolonged exposure to odorants could lead to adverse psychological and physiological reactions, such as headache, anxiety and depression (Tan et al., 2017). Although extensive studies have focused on odor pollution to address these questions, these studies primarily focus on dustbin/house (Shi et al., 2020; Statheropoulos et al., 2005), transfer station (Guo et al., 2017; Li et al., 2013), composting plant (Cheng et al., 2019; Mustafa et al., 2017; Nie et al., 2018) and anaerobic digestion plant (Orzi et al., 2010; Yao et al., 2019).

Indeed, the treatment and disposal of municipal biowaste is a relatively long process chain. It not only includes downstream treatment but also the source throwing/collection and midstream transportation (Xiao et al., 2020), particularly the processes of throwing, collection, and transfer, which would occur in the initial decomposition stage, and these sites are commonly located near residential zones, which then frequently confront strong opposition from urban residents due to the irritating odors emitted (Liu et al., 2020). Unfortunately, the research on odor pollution in the initial decomposition stage of municipal biowaste is relatively insufficient, and lacking research on the emission rate of odor pollutants and the amount and composition of microbial communities. Therefore, regardless of the downstream treatment facilities, the impact of source separation, collection and transportation should not be neglected.

In addition, the municipal biowaste is commonly stored in open or semi-sealed dustbin during the initial decomposition stage (including source separation, throwing, and collection). In this stage, the odor pollutants emitted primarily included direct volatilization, aerobic degradation and anaerobic degradation simultaneously (Wu et al., 2010). This suggests the waste composition and environment conditions play a crucial role in fugitive emissions of odor pollutants. Although there are some studies investigating the profiles of odor pollutants in 24 h (24 h) (Tan et al., 2017; Wu et al., 2020), they didn't quantify the odor emission rate and didn't take into account the impact of waste composition on the emission of odor pollutants.

Accordingly, the present study aimed to investigate odor pollution in the 72-h initial decomposing stage of municipal biowaste, including uncooked food waste (UFW) cooked food waste (CFW) and household kitchen waste (HKW). The 120-L practical dustbin that filled with waste was designed to simulate storage conditions in the early stage. The composition and emission rate of the odor pollutants were determined by the flux chamber method, simultaneously, an odor impact assessment was also conducted. Furthermore, the microbial community structure in different stages (0–72 h) were investigated by both microbial composition and quantities, and its effect on the emission of odor pollutants was also evaluated. The results could provide further understanding for the emission characteristics of odorous compounds in the initial decomposition stage of different biowaste and thereby propose practical advice on the management of these municipal biowaste from the point of controlling odor pollution during the waste collection and transportation, e.g. the best frequency for waste collection, and which odorous compound should be focused for targeted treatment.

2. Materials and methods

2.1. Materials and experimental set-up

The UFW and CFW used in this study were collected from the campus canteens at Tongji University, Shanghai, China. The UFW primarily included vegetables, vegetables leave, melon skins, seafood processing residues and shells. Comparatively, the main ingredients of CFW are leftover dishes and rice. The HKW was collected from a residential community that implemented waste source separation (Liao et al., 2022), which primarily included vegetables leave, leftover dishes and rice, regarding as the mixture of CFW and HKW. The municipal biowaste was collected three times from the different sampling locations during August 2021. The UFW, CFW, and HKW at each sampling location were adequately mixed respectively in the dustbin, and then approximately 120 L of raw materials were taken back to the lab for experiment immediately.

The simulating device included a reactor and a closed dynamic chamber above the reactor. The reactor was constructed from a plastic dustbin with a practical volume of 120 L. The dynamic chamber was constructed by chamber polymethyl methacrylate, and the size was 43.0 cm × 43.5 cm × 15.0 cm (length × width × height). The diagram of the chamber is shown in Fig. S1 (see Supplementary material). Briefly, the gas path along the inner wall was made of silica gel hose (inner diameter, 0.8 cm), and the holes were opened on the hose every 30 cm to facilitate the uniform mixing of gas. The dynamic chamber method was applied to quantify the odor pollutants effluxes. The reactor was placed in thermostatic room (35 °C) and capped with dynamic chamber to simulate the initial decomposing stage of waste. The experiments were conducted for 72 h, in order to simulate the process of waste collection and transportation typically in urban areas (48 to 72 h) (Wang et al., 2015).

2.2. Collection of solid and gaseous samples

The raw biowaste and the solid samples (approximately 2 kg) were collected from the surface and inside (depth of 50 ± 5 cm) of the reactor every 24 h during the experiments. The shovels and food clips were sterilized with 75 % alcohol and washed with sterile water before sampling. The solid samples were removed of bones and impurities, and then homogenized in a shredding machine. The samples were stored in a freezer (−40 °C) for subsequent microbial analysis.

The dynamic chamber method (Lucernoni et al., 2017) was implemented to quantify the odor pollutants effluxes. Before sampling, the dynamic chamber was placed on a dustbin and sealed with PVC hot melt adhesive. The high-purity air (99.999 %, Shanghai Chunyu Special Gas Co., Ltd.) cylinder, flowmeter (FM-6, Darhor Instrument INC, USA) and air inlet with silicone hoses were connected sequentially, and the initial flow was set to 5.0 L min^{-1} as suggested in Lucernoni et al. (2017). To prevent sampling errors derived from the disturbance generated by the disposition of the sampling device, the dynamic chamber was equilibrated for 30 min (30 min) before each measurement. Two sampling methods were used to collect odors samples, including airbag collection and sulfuric acid absorption, as shown in Fig. S1 (see Supplementary material). Among them, the samples that collected by Teflon FEP membrane air bag were used to detect sulfides, alcohol esters, aldehydes ketones, terpenes, benzene. While the sulfuric acid solution absorption (0.01 M and a volume of 10.0 mL) was used to detect ammonia and amine. A total of eight gaseous sampling times were carried out at 0, 8, 16, 24, 36, 48 60, and 72 h, respectively during the experiments. The sampling duration was 1 min, and the sampling was triplicate. The gas samples were all analyzed within 8 h. The blank was carried out when there was no raw material in the dustbin. In addition, although volatile organic compounds (VFAs) are also the typical odorants for putrescible waste (Fang et al., 2013), they were not detected in our preliminary experiments (below the detection limit). This is probably due to the relatively short fermentation process and lack of external microorganism introduced, which weakened the microbial pathway of mixed acid fermentation (Nie et al., 2022).

2.3. Analytical methods

The gas samples collected by gas bag in triplicate were quantitatively analyzed on a preconcentrator GC (450-GC, Varian, USA) platform. The preconcentrator (MARKES International, UK) consists of a sampling module (CIA Advantage-XR), a dewatering module (Kori-XR), a thermal desorption module (Unity-XR), and a cold trap model U-T14HS-2S. The related

operating parameters including pipeline temperature 80 °C, sample purge time 3 min, sample purge and sample flow rate 50 mL min^{-1}, Kori cold trap low temperature −30 °C, high temperature 300 °C, cold trap purge time 2 min. The pulsed-flame photometric detector (PFPD) (Varian, USA) was used for sulfide analysis. The corresponding column (CP-SIL 5 CB, ThermoFisher Scientific, USA) was 30 m × 0.32 mmID × 4.00 μm. Alcohol esters, aldehydes, ketones, terpenes and benzenes were analyzed using Flame Ionization Detector (FID) (Varian, USA). The corresponding column (CP-WAX 52 CB, Thermo Fisher Scientific, USA) was 60 m × 0.32 mmID × 1.20 μm. The standard gases of sulfide, alcohol esters, aldehydes ketones and benzene were prepared by Dalian Datte Gas Co., LTD., and terpenes were prepared by Sichuan Zhong standard Technology Co., LTD., The performance of this method was shown in Table S1 (see Supplementary material).

The sulfuric acid absorption solution was analyzed by ion chromatography (ECO IC, Metrohm, Switzerland) to quantify ammonia, methylamine, dimethylamine, trimethylamine and isopropyl amine. The target substances were separated by cationic chromatographic column (Metrosep C 6-250/4.0, Metrohm, Switzerland). Ammonium chloride, methylamine hydrochloride, dimethylamine hydrochloride, trimethylamine hydrochloride and isopropyl amine hydrochloride were used as reference materials to establish standard curves. The eluent consisted of 4.5 mmol nitric acid and 50 mL BNitrile (chromatographic purity) was prepared in a constant volume of ultrapure water to 1 L. The parameters of ion chromatography were set as follows: injection volume 50 μL, the flow rate of eluent was 0.9 mL min^{-1}, and the recording time was 32.0 min.

2.4. 16S rDNA sequencing and Real-time PCR quantification

DNA extraction was performed on the ultra-clean workbench following instructions on the Kit (DNeasy PowerSoil Pro Kit, Qiagen, Germany). The extracted DNA samples were stored in a −40 °C before high-throughput sequencing analysis. After qualified samples were tested, the 16S universal primers ArBa515F_Arch806R (Bates et al., 2011) of prokaryotes (including bacteria and archaea) were used for PCR amplification of the samples. After amplification, sequencing was performed on MiSeq platform (Illumina, USA), which was performed at Major Bio Bio-Pharm Technology Co. Ltd. (Shanghai, China) using a MiSeq platform (PE300). The detailed information was reported in our previous studies (Nie et al., 2021). Quantitative real-time PCR (qPCR) was used to amplify the 16S rRNA gene to quantify the total bacteria. The reactions were conducted on a thermocycler (Mastercycler @ realplex 2, Eppendorf, Germany). The detailed qPCR procedure and the primer pairs were used to determine the copy numbers of bacteria was described in our previous study (Nie et al., 2021).

2.5. Data analysis

To assess the odorous contribution of a single odorous compound, the threshold dilution of each odorous pollutant was calculated according to Eq. (1)

$$D_i = \frac{C_i}{C_i^T} \quad (1)$$

where, D_i is the dilution multiple of odorous compound *I* (non-dimensional); C_i is the concentration of individual odorous compound (ppmv or mg L^{-1}); C_i^T is the olfactory threshold (ppmv or mg L^{-1}) of compound. In this study, the olfactory threshold was shown in Table S2 (see Supplementary material).

The total odor active value (OAV_T) of odorous compound was calculated according to the summation model method while neglecting the synergistic and/or masking effects in complex VOCs mixture, as shown in Eq. (2),

$$OAV_T = \sum_{i=1}^{n} D_i \quad (2)$$

In addition, Principal Component Analysis (PCA) and figures were performed by Origin Pro (2021, OriginLab, USA). Mantel test was performed between microbial composition (based on phylum) and odor pollutants with vegan package "ggcor" in R.

3. Results

3.1. The compositions of odorous compound emissions in the initial decomposition stage

According to the performance of the GC analysis methods, a total of 33 most widely recognised odor pollutants were detected in the initial decomposition stage (0–72 h) of three types of municipal biowaste, which can be compiled into 6 categories, including alcohol esters, aldehydes ketones, benzenes, terpene, sulfides, and ammonia amines. The qualitative and quantitative results of odor pollutants emission in temporal variation were depicted in Fig. 1. In general, the composition of odor pollutants emitted varied with the types of municipal biowaste. For example, the alcohol esters and aldehydes ketones were the dominant compound in UFW and accounted for over 50 % of the total emissions of odor pollutants at 0–36 h. However, with the interval of waste removal time continues (48–72 h), the proportion of alcohol ester gradually decreased accompanied by an increase of ammonia amines and sulfides. The proportion of ammonia amine compounds reached a maximum of 46.0 % at 60 h, and the proportion of sulfides gradually increased from 2.5 % at 0 h to 19.1 % at 72 h, compared with the above compound, benzenes and terpenes had lower proportion (Fig. 1a).

Similarly, alcohol esters and aldehydes ketones were also the dominant compound in CFW, approximately accounting for over 95 % of the total odorous compound at 0–72 h (Fig. 1b). Moreover, although other odor pollutants composition, such as ammonia amines, sulfides, benzene and terpenes were also detected, the total proportion was <5.0 %. For HKW, alcohol esters were the most abundant compound, accounted for 80.8–90.2 % (0–24 h), and then represented an apparent decrease at 36–72 h (46.8–53.9 %), accompanied by the increase of aldehydes ketones (reaching the maximum proportion of 42.9 % at 36–72 h). Due to the low concentration levels, the proportions of ammonia amines, sulfides and terpenes were all unremarkable (<1.50 %) at 0–24 h, while it slightly increased at 36–72 h, particularly ammonia amines, reaching 11.5 % at 72 h. Furthermore, surprisingly, toluene, ethylbenzene and o-xylene were also detected in small amounts of some samples (Fig. 1c).

The emission rate of odor pollutants for UFW was 960.1 ± 55.9 μg kg^{-1} DM h^{-1} at 0 h, and subsequently it remained growing almost linearly until 72 h, reaching 3963.3 ± 148.6 μg kg^{-1} DM h^{-1}, which increased approximately 4 times. For CFW, the total emission rate increased rapidly (135.8 ± 3.5–1790.8 ± 79.3 μg kg^{-1} DM h^{-1}) at 0–24 h and then remained fluctuated within 24–72 h (1790.8 ± 79.3–2026.1 ± 76.6 μg kg^{-1} DM h^{-1}). Comparatively, the emission rate of odor pollutants for HKW showed an obvious increased trend at 0–16 h, reaching the peak of 10,395.6 ± 362.4 μg kg^{-1} DM h^{-1}, and following accompanied by the rapidly decrease until 72 h (10,395.6 ± 362.4–2451.1 ± 394.5 μg kg^{-1} DM h^{-1}). Overall, these above results represented an obvious phenomenon that the highest total emission rate of odor pollutants was observed in HKW, in addition, this was in stark contrast to the pattern of consistently elevated emission rates exhibited by UFW and CFW. In order to identify the cause of the difference in total emission rates, the dominant odor pollutants (the emission rate > 50 μg kg^{-1} DM h^{-1}) were used to calculate the amount of emission in half-day ($d_{1/2}$), the first day (d_1), day 2 (d_2) and day 3 (d_3), respectively. As shown in Fig. 1d, although the

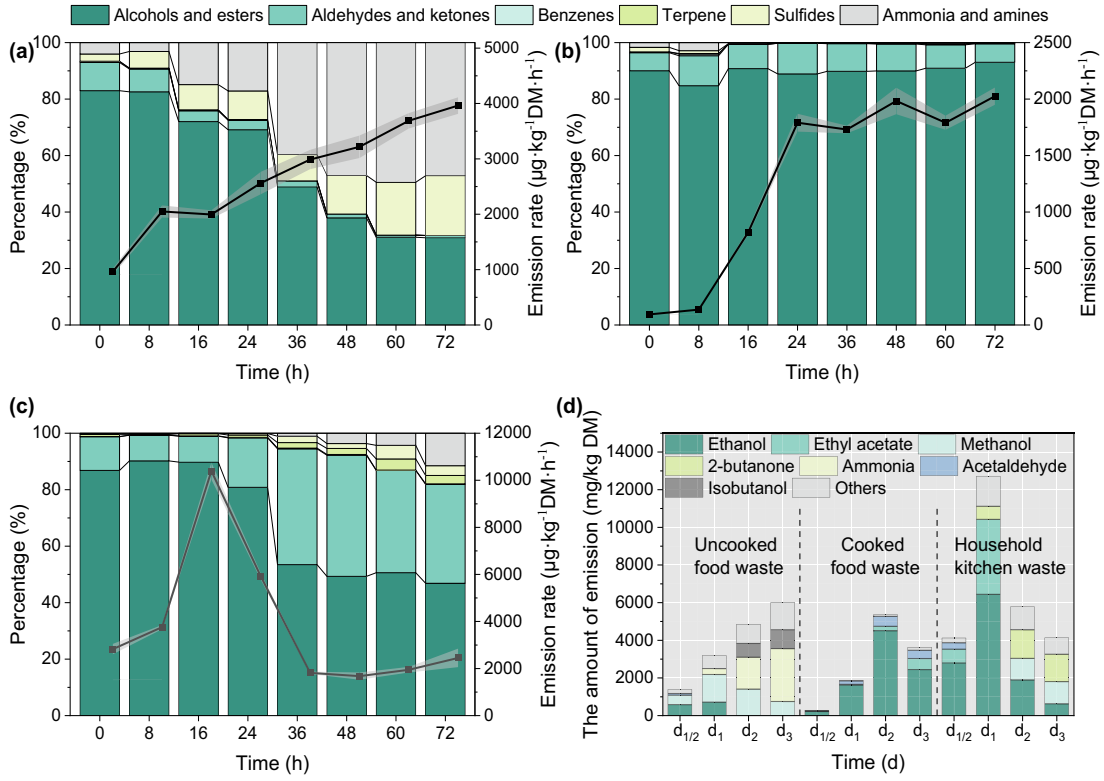


Fig. 1. The composition and emission rate of odor pollutants from three municipal biowaste. (a) Uncooked food waste; (b) Cooked food waste; (c) Household kitchen waste; and (d) The amount of selected primary odor pollutants emitted. The shade of each line represents standard deviation. $d_{1/2}$, d_1, d_2 and d_3 means 12, 24, 48 and 72 h, respectively.

daily emission for UFW (1388.5–6008.8 mg kg^{-1} DM) and CFW (264.8–3625.8 mg kg^{-1} DM) were both increased over time, the types of primary odor pollutants represent apparent difference, for example, UFW was dominated by ammonia and methanol, while CFW was dominated by ethanol, ethyl acetate and acetaldehyde. Furthermore, the daily emission of odorous pollutants for HKW represented an

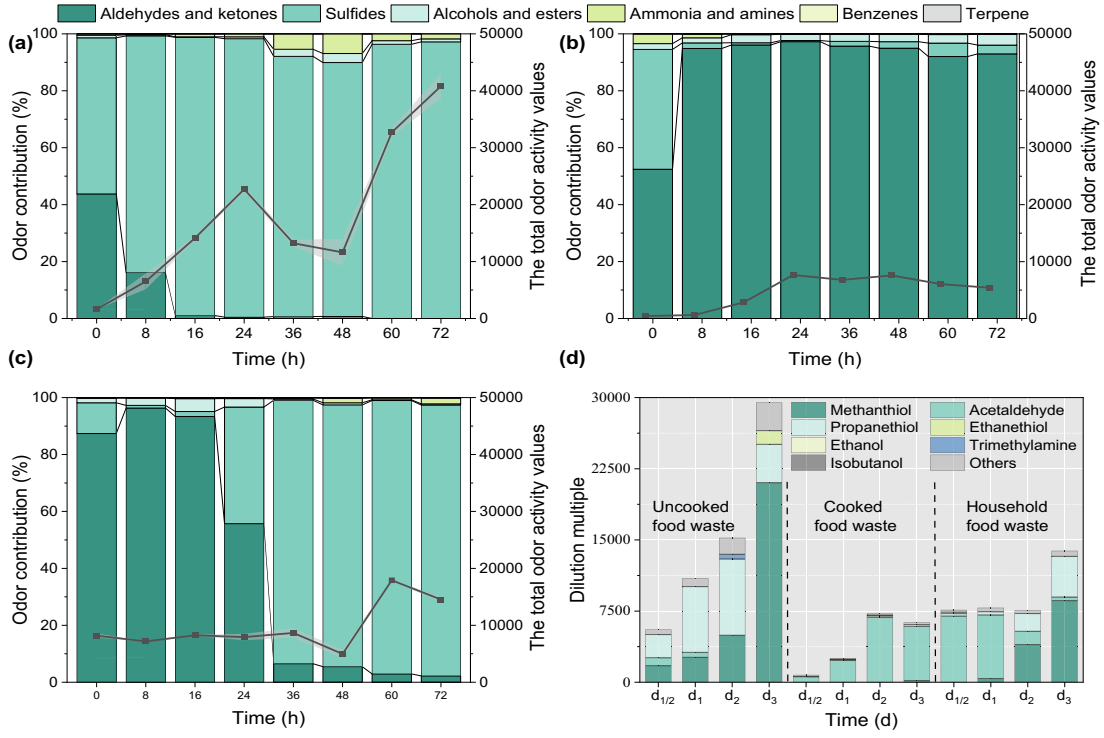


Fig. 2. The odor contribution and the total odor active value from three municipal biowaste. (a) Uncooked food waste; (b) Cooked food waste; (c) household kitchen waste; and (d) the dilution multiple of dominant odor pollutants. The shade of each line represents standard deviation. $d_{1/2}$, d_1, d_2 and d_3 means 12, 24, 48 and 72 h, respectively.

increasing trend from $d_{1/2}$ to d_1 (4119.8–12,719.2 mg kg^{-1} DM), reaching a peak on d_1, and then decreased (5781.1–4132.5 mg kg^{-1} DM).

3.2. The odor active value of odor pollutants in the initial decomposition stage

In order to evaluate the odor contribution of an individual odor pollutants species or chemical category, the total odor active value of odor pollutants was calculated, and simultaneously only components with $D_i > 1$ was regarded as odorants. It was evident that aldehydes ketones were the essential odorant for UFW at 0 h, and the contribution reached 43.8 %. Subsequently, the sulfide became the dominant odor contributor, reaching over 50.0 %, especially at 16–72 h, its contribution was as high as 89.2–97.8 % (Fig. 2a). Indicating that although the concentration of sulfide comprised <20 % of the total odor pollutants emissions (Fig. 1a), its odor contribution accounted for over 85 % due to its relatively low olfactory threshold (for example, Methanethiol, 6.7×10^{-11} ppm), which also demonstrated the fact that most of the nuisances were usually caused by sulfur compounds. In addition, it should be noted that aldehydes ketones were the dominant odor contributor for CFW, and their contribution was higher than 90.0 % within 72 h except for 0 h (the odor contribution was 52.4 %) (Fig. 2b). Although aldehydes ketones had relatively high olfactory threshold (for example, acetone, 7.2×10^{-6} ppm) and which was not generally identified as the priority nuisance compound. However, it still expressed a great odor effect, when it had large emissions and mixed with other compounds. In addition, surprisingly, the dominant odor contributor for HKW were aldehydes ketones at 0–24 h, as the interval of waste removal time continues, complex sulfur compounds become the dominant contributors at 36–72 h and the nuisance of oxygenated compounds transitions to a slight contributor (Fig. 2c).

To quantify the perceived strength of odor sensation, the total odor active value was introduced. Noticeably, the total odor active value of three types of municipal biowaste all represented a different variation trend within 72 h. For example, UFW showed an increasing trend from 0 to 24 h (1628 ± 242–22,707 ± 521), decreasing from 24 to 48 h (22,707 ± 521.9–11,607 ± 2356), and increasing again from 48 to 72 h, reached the maximum value of 40,806 ± 2188 at 72 h. Comparatively, the total odor active value of HKW increased slightly from 0 to 36 h (7175 ± 66–8656 ± 848), decreased to 4963 ± 713 at 48 h, and then increased rapidly to 17,914 ± 173 at 60 h. This phenomenon indicated that the odor effect would aggravate with the interval of waste removal time continues, except for 24–48 h. However, it should be noted that surprisingly the total odor active value of CFW showed a slightly increasing trend at 0–24 h and then decreasing from 24 to 72 h, this probably attributed to the relatively stable compound composition with composed primarily of alcohol esters (Fig. 1b). In addition, the highest total odor active value was observed in UFW, which was approximately four times higher than CFW and HKW in the corresponding time. Indicating that the compounds in UFW gaseous emissions could probably cause odor pollution to nearby people through inhalation.

Based on the fact that the frequency of waste removal time, the dilution multiples of the three top critical compounds in $d_{1/2,}$ d_1, d_2, and d_3 were calculated with detection thresholds. These compounds included most of the critical compounds in terms of the amount of emission (Fig. 1d), such as ethanol, isobutanol, acetaldehyde and trimethylamine. This finding was mainly ascribed to compounds with relatively high detection thresholds at magnitudes. In contrast, most sulfur compounds, especially organic sulfur compounds, such as methanethiol, propanethiol and ethanethiol, presented very low detection thresholds. Hence, very low concentrations of these compounds could cause intense olfaction, resulting in serious odor pollution. In this regard, the initial decomposing of HKW mainly happens in neighborhoods and communities should be removed in time. In the catering industry that adopts UFW and CFW waste classification, special attention should be paid to UFW. The most appropriate strategy was to remove the UFW within 24 h, which may have a certain effect on reducing odor pollution. On the other hand, more targeted deodorization measures should be taken based on the primary odor compound species at the collection sites.

3.3. Microbial diversity and dynamics

The result in phylum level was illustrated in Fig. S2 (see Supplementary material). It was obvious that the bacteria in and on the surface of UFW were dominated by Proteobacteria. The relative abundances in the interior and surface layers decreased from 78.1 % to 48.8 % and 79.5 % to 51.4 %, respectively, at 0–72 h, accompanied by the increase of Firmicutes. Comparatively, Firmicutes was the dominant bacteria in CFW, accounting for 90.6 % and 88.9 % in the interior and surface, respectively. In addition, a similar result was observed in HKW, Firmicutes was also the dominant bacteria, and the relative abundance reached 76.2 % and 69.2 % in internal and external, respectively. This result probably suggesting that due to the difference of dominant microorganisms and waste composition lead to the variation of odorous pollutants, previous studies also reported that Proteobacteria were indigenous microorganisms of fresh fruit and vegetable wastes (Huang et al., 2014), while Firmicutes prefer to use various substance in food waste, such as fats, oils, and protein, to produce volatile fatty acids (Zhang et al., 2020). This was consistent with the composition of different types of biowaste (see Table S3 in Supplementary material), which also explained why the oxygenated compounds was the dominant odorous pollutants emitted by CFW (Fig. 1).

The result of three types of municipal biowaste in genus level is illustrated in Fig. 3. It was obvious that *Acinetobacter* was the dominant genus in UFW (accounting for 30.4 % and 33.9 % in the interior and surface, respectively), followed by *Pseudomonas* (accounting for 30.4 % and 33.9 % in the interior and surface, respectively). *Acinetobacter* has been reported that which could use sulfur-containing compounds as sulfur sources and participate in the oxidation of thiosulfate (Luo et al., 2013), and *Pseudomonas* plays a crucial role in the production of various odorous compounds such as methyl mercaptan, dimethyl disulfide, and ethyl acetate (Mayr et al., 2003; Parlapani et al., 2014). Therefore, this could explain a large amount of sulfides emitted in the early stage of UFW (Figs. 1a and 2a). In comparison, *Lactobacillus* was the most abundant genus in CFW, accounting for 0.2 % and 0.6 % in interior and surface, respectively at 0 h, and then represented an obviously increased trend (67.8 % and 47.8 %, respectively at 72 h), followed by *Weissella* and *Leuconostoc*, which remained above 10.0 % at 0–72 h. Previous studies have reported that both *Weissella* and *Leuconostoc* could produce ethanol and acetic acid by degrading organic wastes such as carbohydrates (Cicotello et al., 2018). In addition, largely accumulated lactic acid and acetic acid can also be converted to ethanol by *Lactobacillus* and *Weisseria*. (Zhang et al., 2020). Therefore, the high content of the above bacterial genera was an essential reason for the high emission rate of ethanol in the initial decomposing of CFW.

Compared with CFW, *Lactobacillus* was also the dominant genus in HKW, with a relative abundance of 49.0 % and 42.3 % in the interior and surface, respectively. In addition, *Acinetobacter*, *Kurthia*, *Weissella*, *Megasphaera*, *Leuconostoc* and *Comamonas* were also observed in HKW, which were both over 3.0 %. It should be noted that *Megacoccus* as a strictly anaerobic gram-negative coccus, which represented an obvious increased trend with time, reached 18.8 % at 72 h. This indicated that the internal environment of HKW during the experiment was beneficial to the growth and proliferation of anaerobic bacteria. Accordingly, *Macrococcus*, as an aerobic bacterium, represented an opposite result, which had a low relative abundance in internal, and a high relative abundance of surface (reaching 13.0 % at 72 h), which suggests that the differences within and on the surface of the waste lead to differences in the composition of microbial communities and their evolutionary trends.

3.4. Real-time PCR quantification of bacteria in municipal biowaste

In addition to focus on microbial diversity, the total amount of microorganisms was quantified using qPCR (Fig. 4). It was obvious that the total amount of bacteria in the surface for UFW (4.24E + 12 copies/mL) and HKW (8.51E + 11 copies/mL) were all higher than that for CFW (5.27E + 12 copies/mL) in the corresponding time, except for 48 h, combined

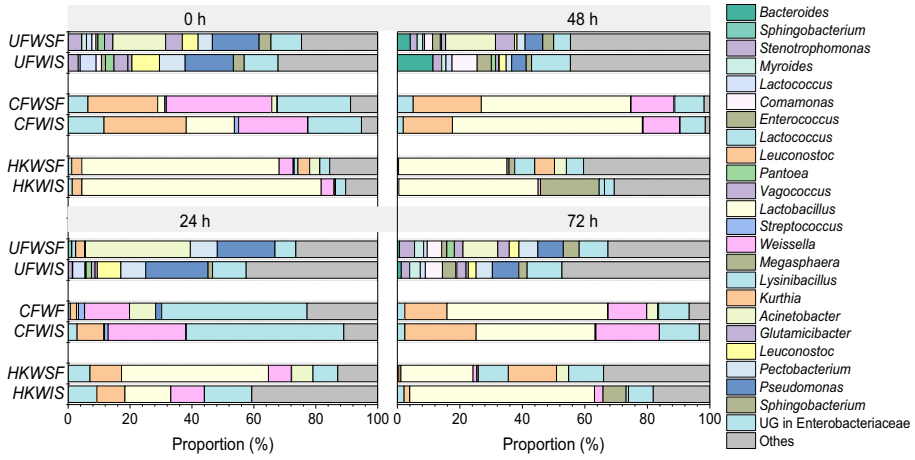


Fig. 3. Taxonomic distribution of bacteria at genus level as revealed by 16S rDNA analysis in different decomposing stages of municipal biowaste. SF: surface, IS: inside.

with the results of microbial composition (Fig. 3), this phenomenon probably attributed to the anoxic condition that developed by over prolonged storage period, which highlighted the *Lactobacillus* obtained the competence, leading to the sudden increase of *Lactobacillus*. This phenomenon was also supported by our previous study (Nie et al., 2022), which reported CFW would produce a lot of lactic acid (>10 g/L) in the storage process (48 h). Moreover, the apparent reduction at 72 h probably due to the accumulation of lactic acid which inhibits the activity of the microorganisms. Meanwhile, it was worth noting that the number of microorganisms in both UFW and HKW all remained relatively high in the whole decomposing stage (0–72 h). In addition, it was also observed that the total amount of bacteria in UFW and HKW all remained a high level whether in inside or on the surface, particularly at 24 h and 72 h, which generated two peaks. Taken together, this suggested that the variation of microorganisms' amount from the waste throwing/ collections sites were associated with the frequency of waste removal.

3.5. Correlations between microbial diversity and the odorant emissions in the initial decomposition stage

The non-metric multidimensional scaling (NMDS) plot also provides evidence that the composition of odorous pollutants represents an obvious trend of change with time (Fig. 5a). For example, the composition of odorous pollutants within 72 h had significant differences in NMDS1 and NMDS2. The composition of odorous pollutants in UFW and HKW represented an apparent disperse and enlarged the distance with the change of time. In particular, from 24 h to 72 h, the composition of odorous pollutants apparently clustered together and enlarged the distance from other odorous pollutants. It suggests that short-term storage (>24 h) had substantially changed the composition of odorous pollutant, which also correspond well with the result of odorant composition (Fig. 1a and c). Combined with the increasing emission rate of odorant, particularly the result of odor impact (Fig. 2), it possibly indicated that the interval of waste removal time had significant influence on the changes of odorous pollutants composition, and in turn potentially impact on the residential community. By contrast, the composition of odorous pollutants in CFW was relatively stable, which apparently clustered together and had part of the overlap with HKW. Indicating that the odorant composition in CFW remain stable at 0–72 h, this was also supported by the above result that with the alcohol esters and aldehydes ketones as the primary compound (Fig. 1b), this phenomenon was mainly attributed to the rapid hydrolysis that conversion to oxygen compounds.

Furthermore, the related microbial composition was also performed NMDS analysis based on the relative abundance of bacterial genera (Fig. 5b). The bacterial composition of UFW and HKW in the interior and surface showed significant difference, which represented distinct

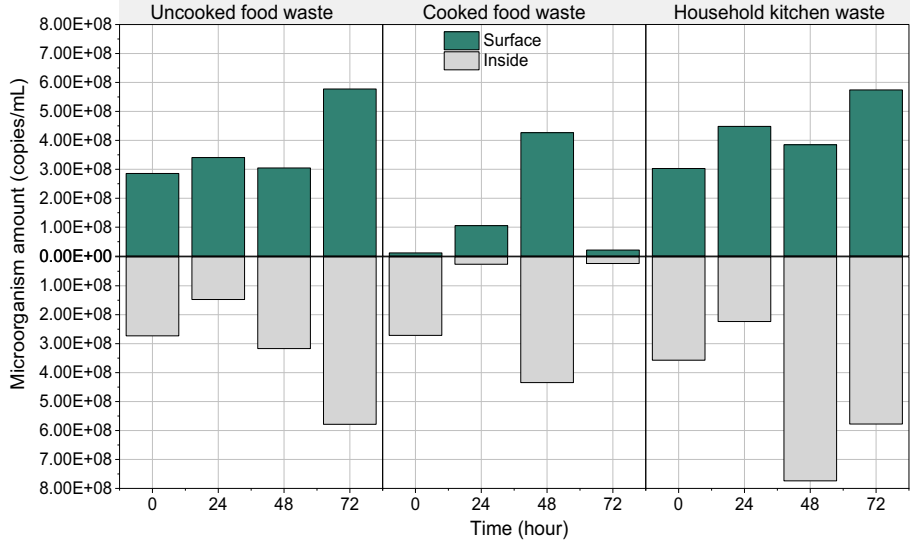


Fig. 4. Total amounts of bacterial based on the real-time PCR at various different types of municipal biowaste.

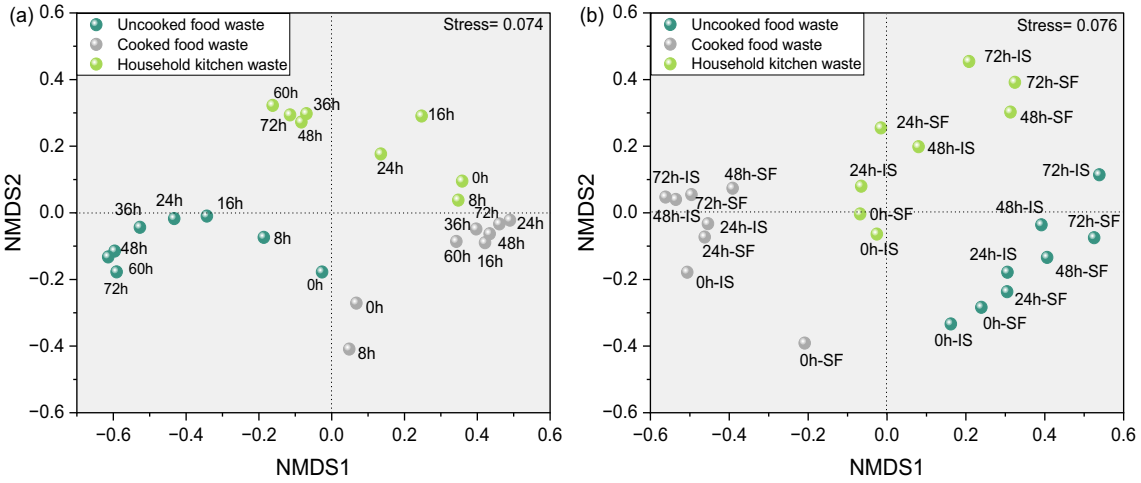


Fig. 5. The non-metric multidimensional scaling analysis of all samples based on the odor pollutants and microbial composition. (a) Odor pollutants; (b) Microbial composition. SF: surface, IS: inside.

dispersion cluster in NMDS1 and NMDS2. Comparatively, the microbial composition in CFW clustered closely. This indicates that as the interval of waste removal time continues, the microbial composition of UFW and HKW in interior and surface has changed greatly, while the microbial composition of CFW remained relatively stable. This difference probably due to the obvious difference in compositions and properties of municipal biowaste, leading to the different evolution directions of microbial communities. Moreover, the differences of microbial in inside and surface of the same type of municipal biowaste may be due to different oxygen levels at different depths. Surprisingly, the discrepancy in odorous pollutants composition was consistent with the certain degree of change in microbial composition. Consequently, the above phenomenon further demonstrated that microorganisms are the driving factor for the emission of odorous pollutants, and changes in the microbial community will directly affect the composition of odorous pollutants.

In order to further analyze the relationship between microbial and odorous pollutants, mantel test was carried out based on the microbial composition (in phylum level) and odorous pollutants (Fig. 6), it was evident that each odorous pollutant was significantly associated with two or more bacterial genera, and most bacterial genera such as *Acinetobacter*, were also significantly associated with multiple odorous pollutants Fig. S5 (see Supplementary material). This was attributed to that a single bacterial species can produce a variety of odor pollutants (Verginer et al., 2010), and different bacteria could also produce the same odor pollutant (Korpi et al., 2009). In general, a significant positive correlation between microorganisms and odorous pollutants suggested that the microorganisms can

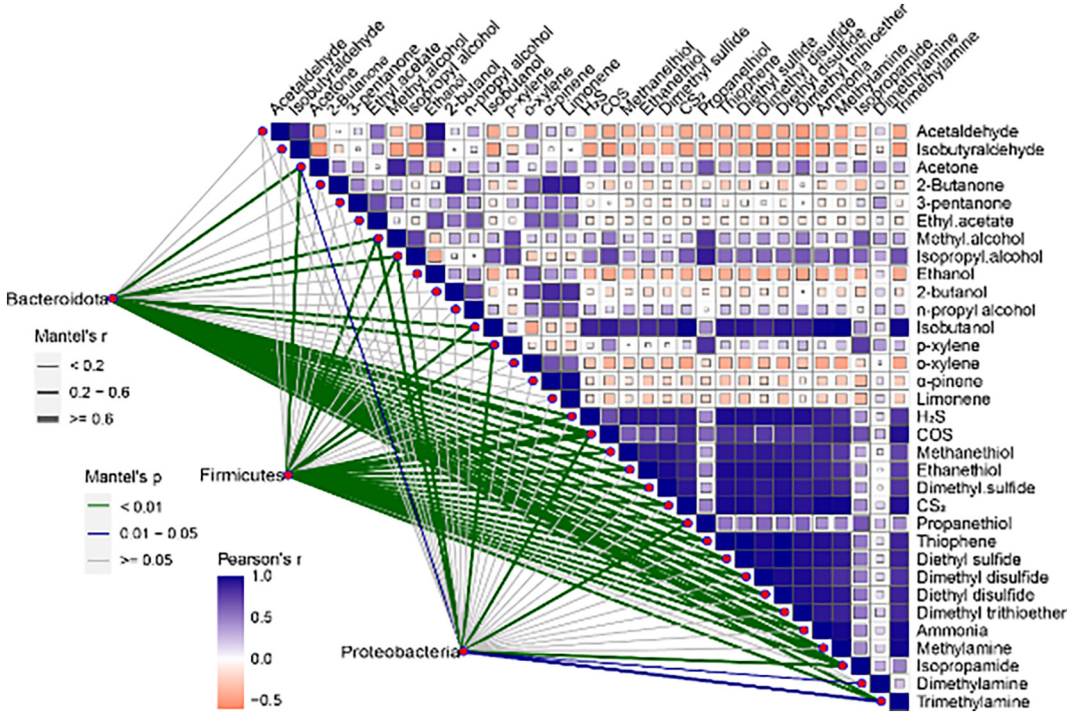


Fig. 6. Mantel test of odor pollutants and microbial composition (based on phylum). The correlation within physiochemical parameters was marked with a cross ($P > 0.05$). The distance matrix of the odor pollutants was calculated based on Euclidean distance, and the microbial composition was based on Bray-Curtis distance. The color and width of the edge denote statistical significance and Mantel's r statistic.

produce the odorous pollutants or the odorous pollutants could promote the growth and proliferation of microorganisms, while a significant negative correlation may be that the microorganisms use the odorous pollutants as carbon. The source or the odorous pollutant could inhibit the growth and proliferation of related microorganisms (Mayrhofer et al., 2006). For example, *Acinetobacter* is significantly positively correlated with various sulfides such as dimethyl disulfide, methyl mercaptan, methyl sulfide, carbonyl sulfide, carbon disulfide, dimethyl trisulfide, etc. It plays an essential role in the metabolism of sulfide (Xia et al., 2015). *Weissella* and *vibrio* are significantly positively correlated with ethanol and acetaldehyde, and significantly negatively correlated with various sulfides such as dimethyl disulfide, while *Corynebacterium* and *sphingosine* are significantly negatively correlated. Conversely, *Sphingobacterium*, *Microbacterium*, *Myroides*, *Stenotrophomonas*, *Vagococcus* and other bacterial genera have significant negative correlation with ethanol and acetaldehyde, and had a significant positive correlation with various sulfides such as dimethyl disulfide Fig. S5 (see Supplementary material). This further verifies the conjecture that various sulfides such as dimethyl disulfide and ammonia may originate from the same or closely related microorganisms, and these microorganisms are significantly different from those that produce acetaldehyde.

4. Discussion

4.1. Emission characteristics of odor pollutants and the related microbial diversity

In contrast to most previous reports focus on the odor emission characteristics of downstream treatment facilities, the present study observed an interesting phenomenon that compared with CFW and HKW, the emission rate and the total odor active value of UFW all represented a relatively high level. Noticeably, an apparent increase of sulfide in the initial decomposing stage (8–72 h) was observed simultaneously in terms of proportion. According to the previous studies, although the concentration of sulfide comprised less, which was still recognised as the dominant odor contributor in the odor pollution of municipal biowaste, due to its relatively low olfactory threshold (Liu et al., 2020). Moreover, UFW was commonly collected from restaurants, industrial area, institutional canteens etc., (Begum et al., 2016), and these locations are usually densely populated work areas. Therefore, it suggests that this UFW throwing/collection sites would cause some disturbance to nearby urban residents due to the irritating odorants emitted. Moreover, it was obvious that oxygenated compounds are the key elements for CFW, whether in emission rate (alcohol esters) or the dominant odor contribution (aldehydes ketones). This result could be attributed to the relatively fast-aerobic decomposition of organic components, which was consistent with our previous study (Nie et al., 2022) where CFW would occur fast hydrolyzes during the collection/transportation (48 h). Comparatively, another interesting phenomenon was observed that aldehydes ketones (0–24 h) and sulfides (36–72 h) were the dominant odor contributor for HKW in temporal variation, respectively. These two compounds were recognised as the dominant odor contributor of UFW (sulfides) and CFW (aldehydes ketones), respectively. This presumably due to the composition of HKW, which included leftovers, vegetable leaves, fruit peels, eggshells and some fruit waste. Furthermore, although compared with UFW, the emission rate and the total odor active value of CFW and HKW all represented a relatively low level to a certain degree, these locations are usually located near residential areas, particularly HKW throwing/collection sites.

Besides, it was worth to mention that this present study observed that the variation of microbial composition and odor composition represented a reasonable consistency in temporal sequence (Fig. 5). It indicated that the emission of various odorous pollutants was closely related to the activities of microorganisms and the microorganisms was probably the primary driving factors for the emission of odorous pollutants (Cui et al., 2022). In addition, the diversity of bacterial communities was enhanced in UFW and HKW during the initial decomposing stage (Fig. 3) and the total amount of bacteria in UFW and HKW all remained a high level whether in inside or on the surface, particularly at 24 h and 72 h (Fig. 4), in addition to odor pollution, these microorganisms also pose a serious health risk in their own right by acting as respiratory sensitizers, they also leave people more vulnerable to infection by live microbial bioaerosols. Therefore, it could be effective that shortening the duration of waste transfer and increasing the frequency of waste collection would relieve the odor nuisance at the initial decomposition stage as far as under ample economic conditions for waste collection. On the other hand, it suggested the necessity of placing waste at a fixed time.

4.2. Significance and perspective

As the implementation of the waste source-segregation policy, the throwing, collection, transportation and transfer (if any) of municipal biowaste is non-avoidable in megacities because of Not-In-My-Back-Yard (Cui et al., 2022). The present study was carried out in the context of the implementation of waste source-segregation policy in China, and investigated the emission characteristics of odorous compounds in the initial decomposing stage of three types of municipal biowaste through simulation experiments in a practical dustbin (120 L) using the flux chamber method. The results indicated that from the perspective of the emission of odor pollutants, HKW > CFW > UFW, while followed the pattern of UFW > HKW > CFW in the perspective of the total odor active value. Moreover, the odorous compounds emitted in the throwing/collection and transportation process would aggravate with prolonging interval of the waste removal, it should be carefully deliberated to avoid the resident complaints. In addition, this study screened out the primary odorous contributor in the temporal variation, and the production mechanism was revealed from the perspective of microorganism.

Therefore, the present study could provide an essential reference for a comprehensive understanding of odorant generation in the initial decomposition stage of three types of municipal biowaste. Furthermore, it could contribute to setting out strategies for odor control and abatement in municipal biowaste management systems. For example, setting a fixed collection schedule and increasing the frequency of waste collection on the basis of considering the differences in the release characteristics of odorous pollutants of different types of municipal biowaste. Certainly, other deodorization technologies, such as the application of biological deodorant and odor liquid adsorbent, also deserved to be explored in further studies so as to alleviate the emission of odorous compounds in the initial decomposing stage of municipal biowaste.

5. Conclusion

The emission rate of odor pollutants for UFW represented a nearly linear growth trend, reaching the maximum (3963 ± 149 $\mu g\ kg^{-1}$ DM h^{-1}) at 72 h. Alcohol esters and ammonia amines had the highest emission rates at 0–36 h and 48–72 h, respectively. Sulfide was the dominant odor contributor. CFW increased rapidly from 8 h to 24 h, and then remain fluctuated, reached the maximum (2026 ± 77 $\mu g\ kg^{-1}$ DM h^{-1}) at 72 h, alcohol esters had the emission ratee and aldehydes ketones was the dominant odor contributor. Comparatively, HKW represented a fluctuating trend, reached the maximum (10,396 ± 363 $\mu g\ kg^{-1}$ DM h^{-1}) at 16 h, and the release rate of alcohol esters was the highest. Aldehydes ketones and sulfides were the most important odor contributor from 0 to 24 h and 36 to 72 h, respectively. Additionally, in phylum level, the UFW was dominated by Proteobacteria, CFW and HKW were all dominated by Firmicutes. In the genus level, *Acinetobacter* was the main type of UFW, and *Lactobacillus* was the main type of CFW and HKW. The variation of microbial composition and odor composition represented a consistency in temporal sequence, indicating that the microbial was probably the primary driving factors for odorous pollutants emission. These results provided important reference information for potential monitoring and control of odorant emissions in management facilities.

CRediT authorship contribution statement

Erqi Nie: Formal analysis, Investigation, Writing - Original Draft, Visualization; **Wei Wang**: Methodology, Validation, Formal analysis, Writing - Review & Editing, Data Curation; **Haowen Duan**: Software, Visualization; **Hua Zhang**: Resources, Validation; **Pinjing He**: Conceptualization, Methodology, Funding acquisition. **Fan Lü**: Conceptualization, Methodology, Data curation, Writing - Review & Editing, Funding acquisition. All authors read and approved the final manuscript.

Data availability

Data will be made available on request.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2022.160612.

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