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# Volatile organic compound emissions from straw-amended agricultural soils and their relations to bacterial communities: A laboratory study

Juan Zhao<sup>1</sup>, Zhe Wang<sup>1</sup>, Ting Wu<sup>1,2,\*</sup>, Xinming Wang<sup>2,\*</sup>, Wanhong Dai<sup>1</sup>, Yujie Zhang<sup>1</sup>, Ran Wang<sup>1</sup>, Yonggan Zhang<sup>1</sup>, Chengfei Shi<sup>1</sup>

1. College of Environmental Science and Engineering, Anhui Normal University, Wuhu 241000, China

2. State Key Laboratory of Organic Geochemistry, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences, Guangzhou 510640, China

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## ABSTRACT

A laboratory study was conducted to investigate volatile organic compound (VOC) emissions from agricultural soil amended with wheat straw and their associations with bacterial communities for a period of 66 days under non-flooded and flooded conditions. The results indicated that ethene, propene, ethanol, i-propanol, 2-butanol, acetaldehyde, acetone, 2-butanone, 2-pentanone and acetophenone were the 10 most abundant VOCs, making up over 90% of the total VOCs released under the two water conditions. The mean emission of total VOCs from the amended soils under the non-flooded condition (5924 ng C/(kg·hr)) was significantly higher than that under the flooded condition (2211 ng C/(kg·hr)). One “peak emission window” appeared at days 0–44 or 4–44, and over 95% of the VOC emissions occurred during the first month under the two water conditions. Bacterial community analysis using denaturing gradient gel electrophoresis (DGGE) showed that a relative increase of Actinobacteria, Bacteroidetes, Firmicutes and  $\gamma$ -Proteobacteria but a relative decrease of Acidobacteria with time were observed after straw amendments under the two water conditions. Cluster analysis revealed that the soil bacterial communities changed greatly with incubation time, which was in line with the variation of the VOC emissions over the experimental period. Most of the above top 10 VOCs correlated positively with the predominant bacterial species of Bacteroidetes, Firmicutes and Verrucomicrobia but correlated negatively with the dominant bacterial species of Actinobacteria under the two water conditions. These results suggested that bacterial communities might play an important role in VOC emissions from straw-amended agricultural soils.

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## Introduction

Volatile organic compounds (VOCs) have received special attention because of their important impact on atmospheric photochemistry by the formation of tropospheric ozone (O<sub>3</sub>) and secondary organic aerosols (SOA) (Singh et al., 1995;

Atkinson, 2005). They can also affect biogeochemical processes in soil through altering the rates of carbon and nitrogen cycling (Smolander et al., 2006; Asensio et al., 2012) and stimulating or inhibiting the growth and activity of bacteria and fungi (Xu et al., 2004; Ramirez et al., 2010), and thus affect plant growth (Farag et al., 2006).

\* Corresponding authors. E-mails: [wuting19@mail.ahnu.edu.cn](mailto:wuting19@mail.ahnu.edu.cn) (Ting Wu), [wangxm@gig.ac.cn](mailto:wangxm@gig.ac.cn) (Xinming Wang).

Agricultural emissions are considered to be an important source of atmospheric VOCs. Previous field and laboratory measurements have quantified the VOC emissions from planted and unplanted agricultural soils, including non-methane hydrocarbons (NMHCs) (Redeker et al., 2003), volatile organic sulfur compounds (VOSCs) (Nouchi et al., 1997; Liu et al., 2010; Yi et al., 2013) and oxygenated volatile organic compounds (OVOCs) (Schade and Custer, 2004; Leff and Fierer, 2008). The available data suggest that the VOC emissions from agricultural fields are largely produced by the aboveground living crops, and change significantly with their growth stages (Nouchi et al., 1997; Redeker et al., 2003; Yi et al., 2013). Compared to aboveground living vegetation, dead crop straws have received less attention with respect to their influence on the VOC emissions from agricultural fields when they return into soils (Nouchi et al., 1997), although a recent study on VOC emissions from forest soils and leaf litter suggested that the decomposition of dead plant matter contributed substantially to VOC emissions from natural soils (Hellén et al., 2006).

Agricultural production generates a large amount of crop straws, approximately 2.5 billion tons per year globally (FAO, 2013) and 0.8 billion tons per year in China (Jiang et al., 2012). The crop straw is commonly incorporated into cultivated fields to provide plant nutrients and maintain soil fertility. It is estimated that 46.1% of crop straws are returned to agricultural soils every year in China (Jiang et al., 2012). A laboratory experiment on the VOC emissions from rice and maize straws showed that the emission of six VOCs (ethane, ethene, propane, propene, n-pentane and methyl chloride) totally reached 1.73 ng/gdw (g dry weight) per hour at 20°C and increased to 18650 ng/(gdw·hr) at 70°C (Derendorp et al., 2011). Therefore, the decomposition of the returned crop straw could release a mass of VOCs, and thus may influence the VOC emissions from agricultural soils.

Microorganisms play a critical role in soil VOC emissions (<http://bioinformatics.charite.de/mvoc/>) (Lemfack et al., 2014). Soil bacteria are capable of producing a diverse array of VOCs, including alcohols, ketones, aldehydes, esters, carboxylic acids, lactones, terpenes, sulfur and nitrogen compounds, and aliphatic and aromatic hydrocarbons during the decomposition of organic substances (Leff and Fierer, 2008; Gray et al., 2010; Ramirez et al., 2010). Some VOCs are characteristically produced by specific phylogenetic groups or species. For instance, the VOC spectra of *Pseudomonas* species are dominated by alcohols, aldehydes, ketones, alkanes and alkenes, whereas *Aspergillus* and *Penicillium* species release distinct alcohols, ketones and furans, and *Streptomyces*, *Bacillus* and *Pseudomonas* are known to produce S-containing volatiles (Gerber, 1968; Medsker et al., 1968, 1969; Dickschat et al., 2005). On the other hand, soil microbes can readily consume a wide range of VOCs and may represent an important sink for VOCs in terrestrial ecosystems (Smolander et al., 2006; Leff and Fierer, 2008). Furthermore, the microbial production or consumption of soil VOCs also depends on the presence of substrates and environmental conditions in the soils (Blom et al., 2011). Several studies have revealed that straw return shifted the microbial diversity and structure in cultivated fields (Tanahashi et al., 2005; Asari et al., 2007; Huang et al., 2012), with an increase of  $\gamma$ -Proteobacteria and a decrease of Sphingobacteria and Verrucomicrobia in the amended soil compared to the control

soil. However, the corresponding changes in the VOC emissions from straw-amended agricultural soils are still unknown, so a study on the relations of the VOC emission to bacteria communities would be useful for clarifying the role of bacteria communities in the VOC emission from straw-amended agricultural soils, as well as understanding the microbial mechanism of soil VOC emissions.

Cultivated soils used for upland crop production are mostly under aerobic conditions, and soils for irrigated rice production are flooded for various lengths of time. In our previous study (Wang et al., 2015), we measured the C<sub>2</sub>–C<sub>5</sub> NMHC emissions from straw-amended agricultural soils under non-flooded and flooded conditions, and found substantially higher light alkene emissions from agricultural soils after straw return, particularly under non-flooded conditions. We also found a significant correlation of the C<sub>2</sub>–C<sub>5</sub> NMHC fluxes with microbial biomass C, respiration rate and population in amended agricultural soils. However, the emissions of other VOCs and the roles of specific microbial species remain unidentified. In the present study, a more specific and comprehensive survey was conducted on VOC emissions, including alkanes, alkenes, alcohols, aldehydes, ketones, furans, esters, sulfides and halocarbons, as well as the bacterial community structure in straw-amended agricultural soils under non-flooded and flooded conditions. The goals were to determine how the VOC release patterns and bacterial community structures varied with incubation time in straw-amended soils under two water conditions, and to study the relationship between soil VOC emissions and bacterial communities in straw-amended agricultural soils.

## 1. Materials and methods

### 1.1. Soil and straw collection

The soils used in the experiment were sampled from the 0–5 cm layer of a typical subtropical paddy field under rice-wheat crop rotation in May 2013 after wheat harvest in Wuhu, China (N 31°16.804', E 118°35.164'). The soil is characterized as gley paddy soil with a clay loam texture (22.4% clay, 40.1% silt, 37.5% sand), bulk density of 1.30 g/cm<sup>3</sup>, pH of 5.18, water holding capacity of 80%, total-C content of 18 g/kg, total-N content of 5.1 g/kg, total-S content of 1.7 g/kg, available-S content of 36.8 mg/kg and available-P content of 7.35 mg/kg. After collection, soils were air-dried, ground and passed through a 2-mm sieve, then stored in a cool and dry place for use.

Fresh wheat straw was collected immediately after harvesting in the same field at the same time. The wheat straw characteristics were as follows: total-C content of 395 g/kg, total-N content of 4.5 g/kg, total-S content of 0.65 mg/kg, and water content of 7%. After collection, the wheat straw was air-dried and cut to 0.5 cm for use.

### 1.2. Incubation experiment

The experiment was performed in the laboratory. Self-made reactors were employed to incubate agricultural soils before and after amendment of wheat straw, and dynamic chambers were used to collect gas samples. The design and operation of the reactors and the chambers has been described in detail

elsewhere (Wang et al., 2015). Briefly, each reactor was made of a cubic box (35 × 35 × 35 cm) without cover but with a collar. Each chamber contained an air inlet and an air outlet. Two small fans were installed inside the chamber to ensure that the headspace air was well mixed. Both the reactor and chamber were made of stainless steel plates with their inner walls coated with Teflon films. All of the connecting tubes were made of Teflon.

Two treatments were conducted in triplicate in the study: (1) soils amended with wheat straw under non-flooded condition (non-flooded SW), (2) soils amended with wheat straw under flooded condition (flooded SW). For the experiment, 27 kg dry paddy soil was first filled into each incubator, and its moisture was adjusted with distilled water to 80% (w/w) for the non-flooded SW treatment and 120% (w/w, water depth of 8 ± 2 cm above the soil surface) for the flooded SW treatment. Then, 1.35 kg dry wheat straw with the same moisture (80% and 120% for the non-flooded and flooded SW treatment, respectively) was put into each reactor and mixed homogeneously with the soil in the two treatments, with a straw-to-soil ratio of 1:20. All of the treatments were incubated at room temperature (25 ± 0.5°C). Except for sampling, the reactors were exposed to air saturated with moisture. Measurements were simultaneously performed on the VOCs and bacterial communities at day 0 before and after straw return, and on days 4, 7, 12, 17, 24, 34, 44, 56 and 66 during the incubation.

### 1.3. VOC flux measurements

At 11 time points during the 66-day incubation period, VOC fluxes were measured by a dynamic flow-through chamber technique. Briefly, dry clean air (with VOC levels comparable to that in ambient air) from a gas cylinder instead of ambient air passed through the chamber after being filtered with activated carbon humidified by deionized water at a rate of 0.5 L/min. After 4 hr (over 5 cycles of residence time), when a steady state was reached, air samples were collected from the outlet with 1 L Teflon sampling bags (SKC Inc., USA). The emission fluxes ( $F$ , ng/kg·hr) were calculated as below:

$$F = Q \times [C_o(t) - C_i(t)] / M_w \quad (1)$$

where  $Q$  (L/hr) is the airflow rate of compressed air through the chamber,  $C_o(t)$  (ng/L) is the concentration in the outgoing air, and  $C_i(t)$  (ng/L) is the concentration in the incoming air. For VOCs,  $C_i(t)$  is zero.  $M_w$  (kg) is the dry mass of unamended or amended soils used for the simulation study.

VOCs were analyzed by an Entech 7100 Preconcentrator (Entech Instruments Inc., CA, USA) coupled with an Agilent 5973 N gas chromatography-mass selective detector/flame ionization detector (GC-MSD/FID, Agilent Technologies, USA), including 10 NMHCs, 35 OVOCs, 4 VOSCs and 1 halocarbon (Table 1). Details about sample analysis and standard preparation and calibration were similar to those presented previously (Yi et al., 2007; Wu et al., 2010; Wu and Wang, 2015; Zhang et al., 2013). Briefly, VOCs in air samples were concentrated using an Entech Preconcentrator with three stages of cryo-trapping, and then transferred into the GC-MS/FID system for analysis. The mixture was separated first by a HP-1 capillary column (60 m × 0.32 mm × 1.0 μm, Agilent Technologies, USA) with

helium as carrier gas, and then split into two paths: one to a PLOT-Q column (30 m × 0.32 mm × 20 μm, Agilent Technologies, USA) followed by FID detection; the other to a 65 cm × 0.10 mm I.D. stainless steel line followed by MSD detection. The GC oven temperature was programmed to be initially at -10°C, holding for 3 min, increasing to 120°C at 5°C/min, and then to 250°C at 10°C/min and holding for 10 min. The MSD was used in scan mode with mass range from 35 to 350 amu, and the ionization method was electron impact (EI).

Target compounds were identified based on their retention times and mass spectra, and quantified by the external calibration method. All VOCs except C<sub>2</sub>-C<sub>3</sub> NMHCs were determined based on MSD signals. C<sub>2</sub> and C<sub>3</sub> NMHCs were detected based on FID signals. Photochemical Assessment Monitoring Stations (PAMS) standard mixtures containing 57 NMHCs (supplied by Sigma-Aldrich Corp., St. Louis, USA) were used for the identification of 10 C<sub>2</sub>-C<sub>5</sub> NMHCs, while TO-15 standard mixtures including 65 compounds (purchased from Spectra Gases Inc., NJ, USA) were employed for the detection of chloromethane, carbon disulfide and 6 OVOCs (ethanol, acetone, i-propanol, 2-butanone, 2-hexanone and ethyl acetate). The calibration standards of these compounds were prepared by dynamically diluting the 100 ppbv standard gas mixtures with pure nitrogen to 0 (pure nitrogen), 0.2, 1, 5, 25, 50 and 100 ppbv using mass flow controllers and a mixing chamber. For the remaining 29 OVOCs and 3 VOSCs, their pure liquids (bought from Sigma-Aldrich Corp., St. Louis, USA) were first diluted with pure nitrogen to 1000 ppmv as primary standard mixtures, then their standard mixtures were further dynamically diluted with pure nitrogen to 0 (pure nitrogen), 0.2, 1, 5, 25, 50 and 100 ppbv. The calibration curves were obtained by running the diluted standards in the same way as the gas samples. For all target VOCs except methanol and ethanol, good dose-response correlation ( $R > 0.99$ ) was found in the range 0–100 ppbv. The analytical system was challenged daily with a one-point (typically 1 ppbv) calibration before running air samples. If the response was beyond ±10% of the initial calibration curve, recalibration was performed. The method detection limits (MDLs) for each VOC ranged from 0.006 to 0.056 ppbv with a sample volume of 400 mL. The relative standard deviations (RSDs) were less than 8% after 10 replicate analysis of standard mixtures (1 ppbv) in 10 consecutive days. The recoveries of spiked samples (1 ppbv) were 86%–107%. Ethanol, due to its high solubility in the water phase, was easily removed with water during the preconcentration process, and its recovery was therefore 46%. Thus, its quantitative result might be underestimated. Nevertheless, the RSD of ethanol was 5% after 10 replicate analyses of a standard mixture (1 ppbv), which was humidified similarly to the incubation samples.

### 1.4. Bacterial community analysis

A five hundred milligram soil sample was collected for bacterial analysis from each incubator at each time. Each sample consisted of 5 sub-samples. Bacterial communities of soils were analyzed by nested polymerase chain reaction-denaturing gradient gel electrophoresis (PCR-DGGE). Details about sample deoxyribonucleic acid (DNA) extraction, amplification and denaturing gradient gel electrophoresis (DGGE) analysis were similar to those presented previously (Zhao et al., 2012). Briefly, about 0.5 g (wet weight) of the sample was used to extract total genomic DNA

**Table 1 – VOCs detected in the two treatments.**

Alkanes	Alcohols	i-Pentanal	2-Heptanone	Methyl butyrate
Ethane	Ethanol	Hexanal	3-Heptanone	Ethyl butyrate
Propane	i-Propanol	Octanal	6-Methyl-5-hepten-2-one	1-Methylethyl butyrate
n-Butane	1-Butanol	Nonanal	2-Octanone	Butyl butyrate
i-Butane	2-Butanol	Benzaldehyde	3-Octanone	Sulfides
n-Pentane	i-Butanol	Ketones	Acetophenone	Dimethyl sulfide
Alkenes	2-Ethyl-1-hexanol	Acetone	Furans	Carbon disulfide
Ethene	Aldehydes	2-Butanone	2-Methyl furan	Dimethyl disulfide
Propene	Acetaldehyde	2-Pentanone	3-Methyl furan	Dimethyl trisulfide
1-Butene	Butanal	2-Methyl-3-pentanone	2-Ethyl furan	Halohydrocarbons
1-Pentene	i-Butanal	2-Hexanone	Esters	Chloromethane
Isoprene	Pentanal	5-Methyl-2-hexanone	Hexyl acetate	

with FastDNA® SPIN Kit for Soil (MP Biomedicals, CA, USA) according to the manufacturer's procedure. The extracted DNA was used as template for the two successive PCR amplifications. For the first PCR cycle, 16S ribosomal ribonucleic acid (rRNA) was amplified with primers 5'-AGAGTTTGATCMTGGCTCAG-3' (*Escherichia coli* bases 8–27) and 5'-ACGGTTACCTTGTACGACTT-3' (*E. coli* bases 1487–1507). PCR was performed in a thermal cycler with the following sequence: initial denaturation at 94°C (5 min); 30 cycles consisting of denaturation at 94°C (45 sec), annealing at 50°C (45 sec) and an extension at 72°C (1 min); final extension at 72°C (10 min); and soak at 4°C. A final volume of 30 µL reaction mixture contained 19 µL ddH<sub>2</sub>O, 1 µL of each primer (10 µmol/L each), 2 µL dNTP mixture (2.5 µmol/L), 5 µL 10 × Taq buffer, 1 µL Taq enzymes (2.5 U/µL) and 1 µL (about 15 ng) DNA template. During the second PCR cycle, the 16S rRNA V3 region was amplified using the primer 341F (5'-CCTACGGGAGGCAGCAG-3') with a GC-clamp (5'-CGCCCCGCGCGCGCGGGCGGGCGGGGCGGGG GCACGGGGGG-3') at the 5' end and the primer 534R (5'-ATTACC GCGGCTGCTGG-3') (Muyzer et al., 1993). The touch-down PCR program was as follows: initial denaturation at 94°C (10 min); 10 cycles performed at a denaturation temperature at 94°C (1 min) and then an annealing temperature of 65°C (1 min) with a touchdown temperature of 1°C per cycle and an extension at 72°C (1 min); a further 20 cycles with the annealing temperature changed to 55°C (1 min); final extension at 72°C (10 min); and soak at 4°C. The mixture with total volume of 30 µL contained 18.5 µL ddH<sub>2</sub>O, 1 µL of each primer (10 µmol/L each), 2 µL of 2.5 µmol/L dNTP mixture, 5 µL of 10× Taq buffer, 1 µL Taq enzymes (2.5 U/µL), 0.5 µL BSA (2 µg/µL) and 1 µL of products of the first PCR cycle.

The DGGE analysis was performed with 12 µL of the products from the second PCR using the DCode Universal Mutation Detection System (Bio-Rad, USA). The electrophoresis was performed on polyacrylamide gel (8% acrylamide/bisacrylamide, *w/v* = 37.5/1) with a denaturing gradient of 30% to 55%. 100% denaturant contained 7 mol/L urea with 40% formamide (*v/v*). All gels were run in approximately 7 L of 1 × TAE buffer (0.04 mol/L Tris base, 0.02 mol/L sodium acetate, and 0.01 mM EDTA; pH adjusted to 8.0) at 100 V and 60°C for 9 hr, and then were stained by silver staining and photographed by a Geldoc XR system (Bio-Rad Laboratories, Inc., Hercules, CA). The intensities of bands on the gels were analyzed using Quantity One software (version 4.6.2, BIORAD Laboratories).

DGGE bands were excised from the gels, and then eluted in 30 µL sterile ddH<sub>2</sub>O overnight at 4°C to dissolve the DNA. The

dissolved DNA was used as a template for PCR amplification with primers 341F without GC-clamp and 534R. The amplification products were purified with TIANquick Midi Purification Kit (TIANGEN, Beijing, China) based on the manufacturer's instruction, and then sequenced by Sangon Biotechnology Company (Shanghai, China). The nucleotide sequences of the 16s rRNA V3 fragments determined in the study were submitted to the NCBI database limited to sequences from type material under the accession numbers from KP277120 to KP277208.

### 1.5. Statistical analysis

Data analysis was performed using SPSS (version 16.0 for Microsoft Windows). Linear regression analysis was used to investigate the correlation between the emission fluxes of the VOCs and the band intensities of the bacterial communities. One-way analysis of variance (ANOVA) was performed to compare the differences in the VOC emissions between the treatments and between the incubation times with a post hoc LSD test, and the significance level was set at *p* < 0.05.

## 2. Results and discussion

### 2.1. VOC emissions

Fifty VOCs were identified and quantified, namely, five alkanes, five alkenes, six alcohols, nine aldehydes, twelve ketones, three furans, five esters, four sulfides and one halohydrocarbon, during the 66-day incubation period (Table 1). The total VOC emissions from the amended soils averaged 5924 and 2211 ng C/(kg·hr) in the non-flooded and flooded SW treatments, respectively. The OVOCs and NMHCs dominated and contributed, respectively, 95.5% and 4.36% of the total VOCs in the non-flooded SW treatment and 95.9% and 3.95% in the flooded SW treatment. Ten dominant compounds in the two treatments accounted for over 90% of the total emitted VOCs (Table 2), among which acetone was the most abundant and accounted for 49.9% and 35.8% in the non-flooded and flooded SW treatments, respectively. It was followed by ethanol and 2-butanone, accounting for 23.5% and 9.04% in the non-flooded SW treatment and 28.0% and 13.5% in the flooded SW treatment, respectively. This was consistent with their identification as major contributors to VOC emissions during the degradation of dead plant materials or organic matter, such as plant litter (Warneke et al., 1999; Gray et al., 2010) and yard

**Table 2 – Contribution (%) of individual VOC to total VOCs emitted from amended agricultural soils in the two treatments during 66-day incubation.**

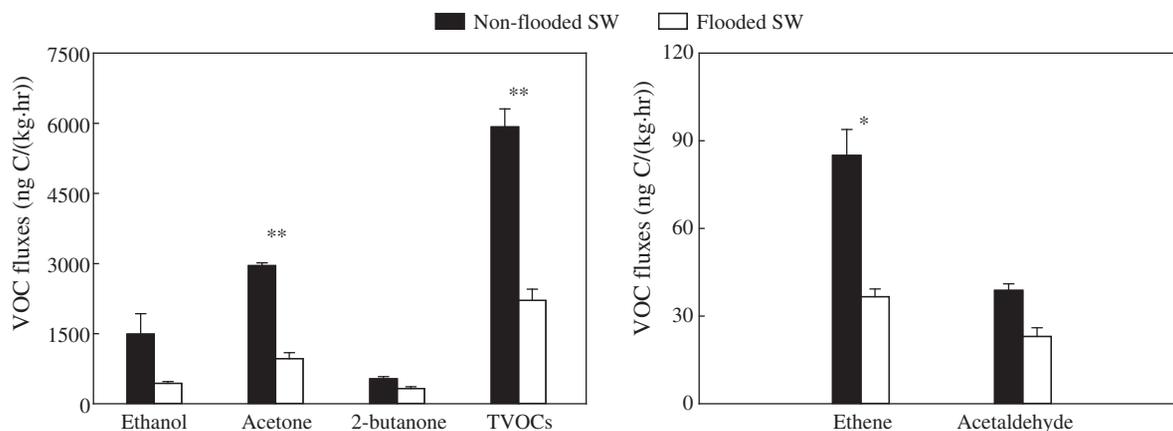
Compounds	Non-flooded SW	Flooded SW	Compounds	Non-flooded SW	Flooded SW
Ethane	0.02 ± 0.00	0.02 ± 0.00	Acetone	49.9 ± 8.86	35.8 ± 7.87
Propane	0.40 ± 0.01	0.57 ± 0.01	2-Butanone	9.04 ± 1.86	13.5 ± 2.13
n-Butane	0.19 ± 0.00	0.25 ± 0.08	2-Pentanone	2.81 ± 0.67	4.86 ± 0.96
i-Butane	0.03 ± 0.00	0.04 ± 0.00	2-Methyl-3-pentanone	0.37 ± 0.13	0.34 ± 0.10
n-Pentane	0.62 ± 0.03	0.60 ± 0.06	2-Hexanone	0.04 ± 0.00	0.20 ± 0.12
Ethene	1.44 ± 0.04	1.79 ± 0.23	5-Methyl-2-hexanone	0.20 ± 0.08	0.35 ± 0.13
Propene	1.10 ± 0.01	0.43 ± 0.00	2-Heptanone	0.06 ± 0.02	0.20 ± 0.08
1-Butene	0.32 ± 0.10	0.12 ± 0.01	3-Heptanone	0.26 ± 0.08	0.71 ± 0.31
1-Pentene	0.12 ± 0.03	0.06 ± 0.00	6-Methyl-5-hepten-2-one	0.07 ± 0.00	0.10 ± 0.02
Isoprene	0.11 ± 0.04	0.07 ± 0.01	2-Octanone	0.06 ± 0.01	0.10 ± 0.03
Ethanol	23.5 ± 5.47	28.0 ± 5.79	3-Octanone	0.05 ± 0.03	0.12 ± 0.07
i-Propanol	3.15 ± 0.77	4.04 ± 0.64	Acetophenone	1.33 ± 0.02	0.32 ± 0.00
1-Butanol	0.06 ± 0.00	1.33 ± 0.42	2-Methyl furan	0.22 ± 0.10	0.25 ± 0.11
2-Butanol	0.65 ± 0.00	1.93 ± 0.25	3-Methyl furan	0.25 ± 0.07	0.26 ± 0.09
i-Butanol	0.15 ± 0.08	0.21 ± 0.13	2-Ethyl furan	0.09 ± 0.04	0.09 ± 0.05
2-Ethyl-1-hexanol	0.56 ± 0.12	0.75 ± 0.21	Hexyl acetate	0.08 ± 0.00	0.10 ± 0.02
Acetaldehyde	0.66 ± 0.00	1.18 ± 0.02	Methyl butyrate	0.12 ± 0.05	0.04 ± 0.00
Butanal	0.05 ± 0.01	0.15 ± 0.11	Ethyl butyrate	0.09 ± 0.03	0.02 ± 0.00
i-Butanal	0.12 ± 0.05	0.10 ± 0.00	1-Methylethyl butyrate	0.33 ± 0.13	0.02 ± 0.01
Pentanal	0.08 ± 0.02	0.10 ± 0.02	Butyl butyrate	0.19 ± 0.12	0.05 ± 0.00
i-Pentanal	0.17 ± 0.10	0.06 ± 0.01	Dimethyl sulfide	0.07 ± 0.01	0.06 ± 0.00
Hexanal	0.10 ± 0.05	0.12 ± 0.06	Carbon disulfide	0.08 ± 0.03	0.04 ± 0.01
Octanal	0.10 ± 0.02	0.11 ± 0.05	Dimethyl disulfide	0.00 ± 0.00	0.01 ± 0.00
Nonanal	0.09 ± 0.01	0.14 ± 0.09	Dimethyl trisulfide	0.00 ± 0.00	0.01 ± 0.00
Benzaldehyde	0.57 ± 0.21	0.22 ± 0.12	Chloromethane	0.02 ± 0.00	0.02 ± 0.00

Mean ± Standard error (S.E.). Non-flooded SW or flooded SW: soils amended with wheat straw under non-flooded condition or flooded condition.

waste (Staley et al., 2006). The mean emissions of both total VOCs (TVOCs) and some major VOCs, such as ethene and acetone, from the non-flooded SW treatment were significantly larger than those from the flooded SW treatment (Fig. 1). This could be attributed to the significantly higher microorganism abundance, resulting in more abundant VOC production (Wang et al., 2015) and a greater gas diffusion rate (Peñuelas et al., 2014) under non-flooded conditions than under flooded conditions. Actually, the average microbial biomass C, respiration and population in non-flooded SW were obviously higher than those from flooded SW (Table 3). Similarly, Liu et al. (2010)

observed that the emission rates of volatile sulfur compounds from two paddy soils decreased with increased soil water content. It is also worth noting that the removal of VOCs in the flushing gas before passing through the chamber might enhance the emission fluxes of VOCs.

As demonstrated in Fig. 2, the emission fluxes of VOCs from soils varied with incubation time. In two treatments, the emissions VOCs of the total and individual VOCs before straw addition were significantly lower than those after straw addition in the beginning. At day 0, the mean fluxes of the total VOCs in non-flooded (16174 ng C/(kg·hr)) and flooded



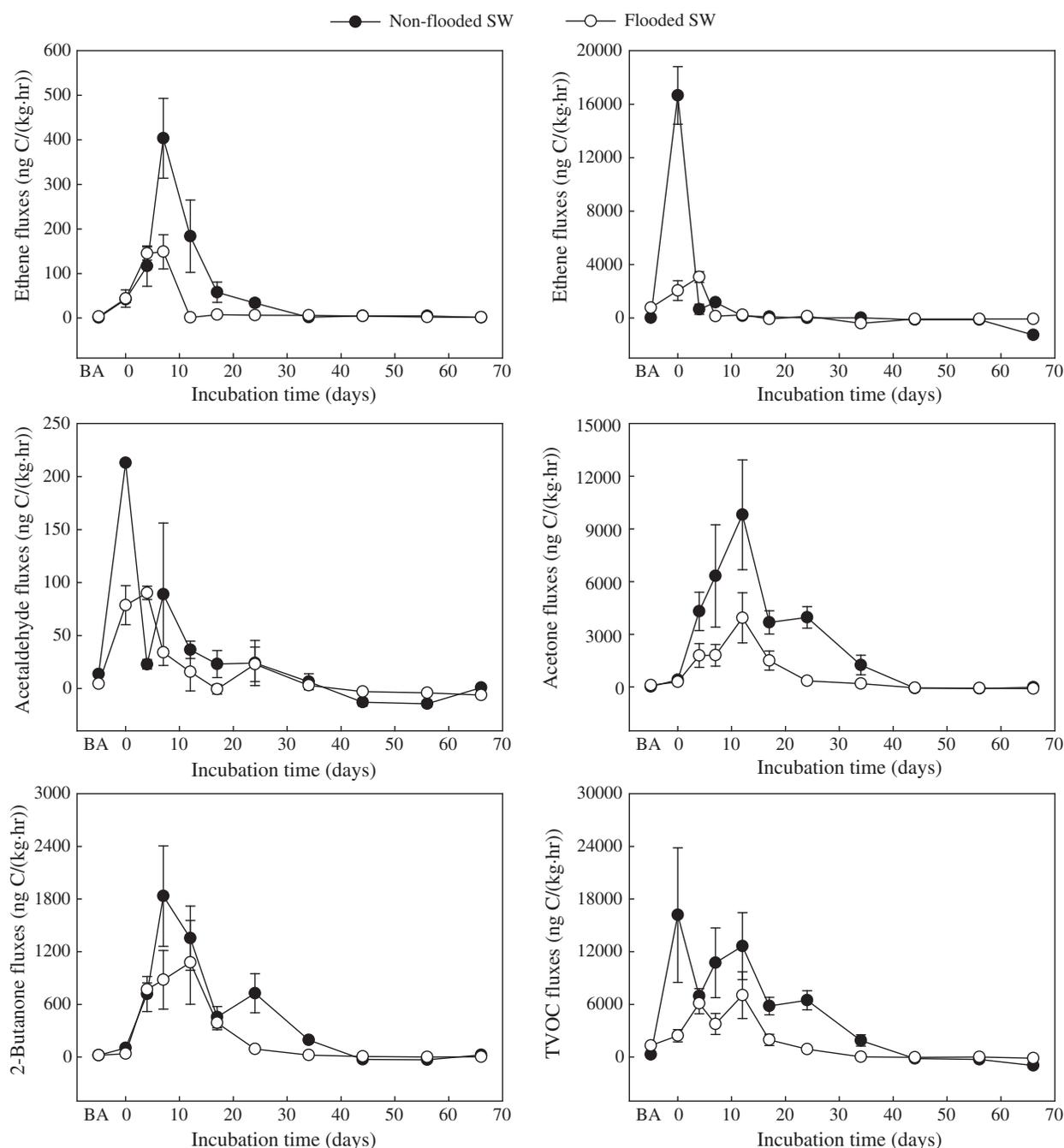
**Fig. 1 – Emission fluxes (n = 3) of some major VOCs from amended agricultural soils in the two treatments. Error bars denote the standard error. TVOCs, total VOCs. \*Significant difference at p < 0.05 level; \*\* significant difference at p < 0.01 level. VOCs: volatile organic compounds.**

**Table 3**–The microbial biomass C, respiration and population in four treatments during the incubation experiments.

Microbial characteristics	Non-flooded SW	Flooded SW
MBC (mg/g)	0.82 ± 0.18	0.54 ± 0.17
CO <sub>2</sub> flux (mg/(kg·hr))	13.6 ± 2.97	9.44 ± 2.94
Bacteria (×10 <sup>6</sup> CFU/gdw)	36.3 ± 16.1	4.87 ± 1.03
Fungus (×10 <sup>5</sup> CFU/gdw)	64.1 ± 22.1	10.5 ± 3.26
Actinomyces (×10 <sup>6</sup> CFU/gdw)	17.1 ± 0.72	15.4 ± 0.47

MBC; microbial biomass carbon, DW; dry weight.

(2429 ng C/(kg·hr)) SW treatments after straw-return were, respectively, 56 and 2 times those (291 and 1303 ng C/(kg·hr)) before straw-return. This finding suggested that the returned crop straw contributed substantially to the soil emissions of VOCs and that the VOCs in the two SW treatments after straw-return at day 0 were mainly derived from wheat straw rather than from soils, although both straw and soil were sources of VOCs. As reported by previous studies, OVOCs and NMHCs are metabolites formed largely from fatty or amino acid precursors in both living and dead plant materials, and can be emitted transiently upon cell wounding (Derendorp et

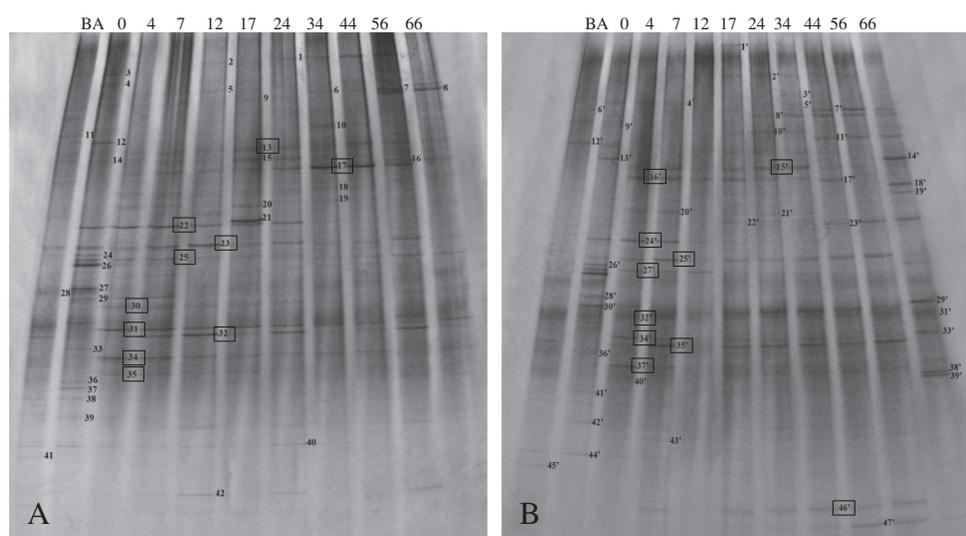
**Fig. 2** – Some major VOC fluxes ( $n = 3$ ) changing with incubation time in the two treatments. Error bars represent the standard errors. BA, samples at day 0 before amendments. TVOC, total VOCs.

al., 2011). Crop straw, as a pool of VOCs, was cut before incubation and was wet at the beginning of experiment, making these compounds unable to be locked in clumps but instead to volatilize rapidly due to the increase of surface area and the replacement of compound molecules with water in the crop residues (Warneke et al., 1999). In the first 4 days after straw return, the emission rates of some VOCs, including ethanol, i-butanol, acetaldehyde, i-butanol, pentanal, i-pentanal, 2-pentanone, 2-methyl-3-pentanone, 6-methyl-5-hepten-2-one, 3-octanone, three furans, four sulfides and chloromethane in the non-flooded SW treatment and i-pentanal, hexanal, nonanal, benzaldehyde, 2-hexanone, 3-heptanone, 6-methyl-5-hepten-2-one, 3-octanone and chloromethane in the flooded SW treatment, decreased sharply, implying that they were mainly derived from the direct evaporation of VOC from the straw storage pool or soil solution. Then, their emissions increased again to form one “peak emission window” at days 4–44, suggesting secondary formation. For the remaining VOCs listed in Table 1, such as three major VOCs (ethene, acetone and 2-butanone) in both treatments and two major VOCs (ethanol and acetaldehyde) in the flooded treatment, emissions were trivial or below detection limits in the beginning and increased immediately to form one “peak emission window” with a maximum at days 4–12, and then decreased sharply until leveling off after 44 days (Fig. 2). The results indicated that these VOCs would be secondarily formed during the incubation rather than inherited, probably by the microbial activity described in detail in part 2.3. In both treatments, the total VOCs emitted during days 4–44, which were probably secondarily formed, were significantly higher in their amounts when compared to those emitted from the amended soils in the first four days, which were probably inherited. Furthermore, the emission rates of the total and most of the individual VOCs were close to zero after 44 days, and over 95% of the emissions occurred

during the first 34 days in both treatments, suggesting that VOCs were mainly emitted from straw-amended agricultural soils during the early decomposition stage. The result was consistent with the findings of previous studies, in which other plant residues, such as orange waste and food waste, mainly consisting of fruit peelings and vegetable trimmings, gave off considerable amounts of VOCs in the early disposal (Wang and Wu, 2008; Wu et al., 2010). This can be explained by the fact that crop residues decompose in two distinct phases, an initial rapid phase followed by a slower phase, and in the initial phase of decomposition, easily available compounds, such as sugar and starch, are strongly transformed by microorganisms into low molecular weight compounds (Wang et al., 2004).

## 2.2. Bacterial community structure

The community structures of the bacteria in the amended agricultural soils in both treatments were analyzed by PCR-DGGE. The triplicate DGGE profiles for each treatment were highly reproducible, so only the one-time DGGE patterns are displayed (Fig. 3). The DGGE patterns showed significant changes over the experimental period. There was an increase in the bacterial population from the unamended soil to the amended soil in both treatments. At day 0, two DGGE bands (bands 28 and 41) that existed in the soils before amendment disappeared, but 11 new bands (bands 12, 22, 24, 25, 26, 27, 29, 33, 34, 36 and 39) appeared after amendment in the non-flooded SW treatment, whereas 12 new bands (bands 9', 13', 24', 26', 27', 28', 30', 32', 36', 37', 41' and 42') occurred in the amended soils when compared to the unamended soils in the flooded SW treatment. On the other hand, the bacterial communities also varied greatly with incubation time after straw return. The band numbers of the bacteria in the amended soils in the non-flooded SW treatment fluctuated



**Fig. 3** – DGGE patterns of bacterial communities during 66-day incubation in non-flooded (A) and flooded (B) SW treatments, respectively. BA denotes samples at day 0 before amendments in the two treatments. The numbers above the lanes designate the sampling days (0–66). The numbers in profiles denote the position of the band in gels. The assignment of numbers to bacteria is shown in Table 4. Boxed numbers represent the dominant bands with intensity of more than 2% and appearance frequency of more than 5 samples. DGGE: denaturing gradient gel electrophoresis.

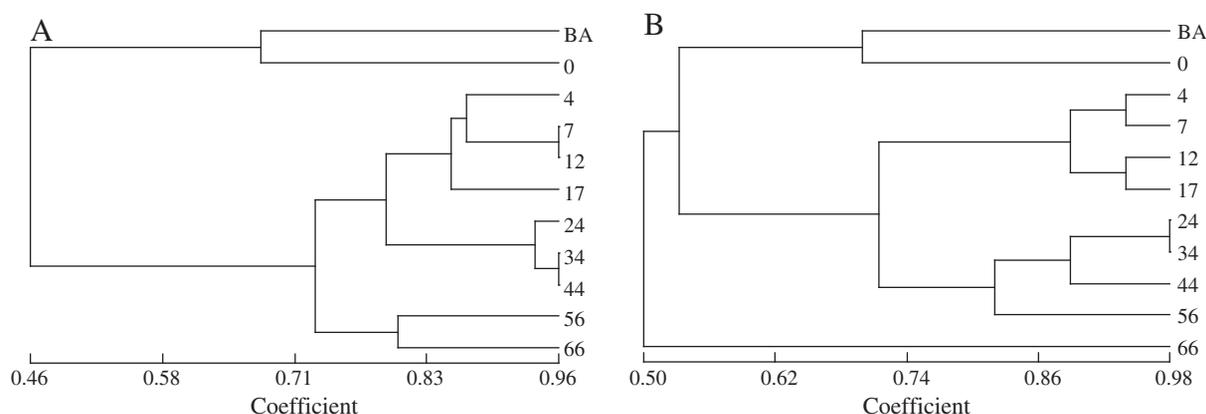
with incubation time and ranged from 10 to 20, with the occurrence of only three bands (bands 23, 25 and 32) during the whole incubation process. In the flooded SW treatment, the DGGE band numbers of bacteria in the amended soils increased with incubation time and were maximized at day 24 (25 bands), with the existence of only three bands (bands 24', 25' and 37') during the whole incubation process. Moreover, cluster analysis separated, respectively, the soil bacterial communities into four groups in the two treatments: at day 0 before amendment, day 0 after amendment, days 4–44 and days 56–66 in the non-flooded SW treatment; and at day 0 before amendment, day 0 after amendment, days 4–56 and day 66 in the flooded SW treatment (Fig. 4), which was consistent with the aforementioned trend of VOC emissions. These results were consistent with the changes in the bacterial communities with the decomposition process of rice straw left on the paddy soil surface or in the paddy field (Tanahashi et al., 2005; Asari et al., 2007).

All bands marked in Fig. 3 were successfully excised and sequenced, and their closest relatives are shown in Table 4. In the non-flooded SW treatment, forty-two sequenced bands belong to 11 phylogenetic groups, including *Acidobacteria*, *Actinobacteria*, *Bacteroidetes*, *Chloroflexi*, *Fibrobacteres*, *Firmicutes*, *Nitrospirae*, *Planctomycetes*, *Proteobacteria*, *Spirochaetes* and *Verrucomicrobia*. Compared to the non-flooded SW treatment, the flooded SW treatment had fewer phylogenetic groups, with the disappearance of four groups (*Chloroflexi*, *Fibrobacteres*, *Planctomycetes* and *Spirochaetes*), although it presented more bands (47 bands). As presented in Fig. 5, the structure and composition of the bacterial communities varied with incubation time. At day 0, three groups (*Bacteroidetes*, *Fibrobacteres* and *Planctomycetes*), which were lacking in the agricultural soils before amendment, emerged after amendment in the non-flooded SW treatment, and there was a relative increase of *Firmicutes* from 16.5% to 31.9% and from 2.36% to 16.4%, and  $\gamma$ -*Proteobacteria* from 4.02% to 20.3% and from 0.00% to 20.2%, but a relative decrease of *Acidobacteria* from 16.2% to 3.06% and from 43.4% to 13.2%, and *Nitrospirae* from 5.60% to 2.57% and from 20.2% to 4.66% was observed before and after straw application in the non-flooded and flooded SW treatments,

respectively. During the 66 day incubation period after straw amendments, the relative abundance of *Actinobacteria*, *Bacteroidetes*, *Firmicutes* and  $\gamma$ -*Proteobacteria* was enhanced with time and maximized at days 34, 66, 4 and 7 in the non-flooded SW treatment, and at days 56, 12, 66 and 44 in the flooded SW treatment, respectively, while those of *Acidobacteria* decreased with time in the two treatments. This confirmed the finding by Cleveland et al. (2007), who observed that labile carbon additions caused a relative reduction of *Acidobacteria* and a relative enhancement of  $\gamma$ -*Proteobacteria* and *Firmicutes*, which may be largely responsible for the decomposition of organic substrates and be particularly adept at responding to increases in labile soil C. Tanahashi et al. (2005) also reported that  $\gamma$ -*Proteobacteria* prevailed during the decomposition of rice straw compost in a Japanese rice paddy field. As indicated by Fierer et al. (2007), in low C environments, oligotrophic groups such as *Acidobacteria* were most dominant, but in high C soils, copiotrophic groups such as *Proteobacteria* and *Bacteroidetes* increased in relative abundance.

### 2.3. Correlations between the VOC emissions and bacterial communities

Correlations between the fluxes of the top 10 VOCs and the intensities of the dominant DGGE bands were performed to study their interactions during the 66-day incubation after straw amendment in both treatments. As shown in Table 5, the correlations for the two treatments showed a similar trend, such that most of the top 10 VOCs were positively related to the predominant bacterial species of *Bacteroidetes*, *Firmicutes* and *Verrucomicrobia* but negatively to those of *Actinobacteria*. In detail, the non-flooded SW treatment had a significantly positive correlation between *Bacillus niacin* and three VOCs (ethene, 2-butanol and acetaldehyde), between *Caldilinea aerophila* and acetophenone, between *Desulfobacterium dehalogenans* and propene, between *Desulfobacterium metallireducens* and i-propanol, between *Desulfovibrio oxamicus* and five VOCs (ethene, i-propanol, 2-butanol, acetone and 2-butanone), between *Methylacidimicrobium tartarophylax* and propene, between *Oceanirhabdus sediminicola* and four VOCs



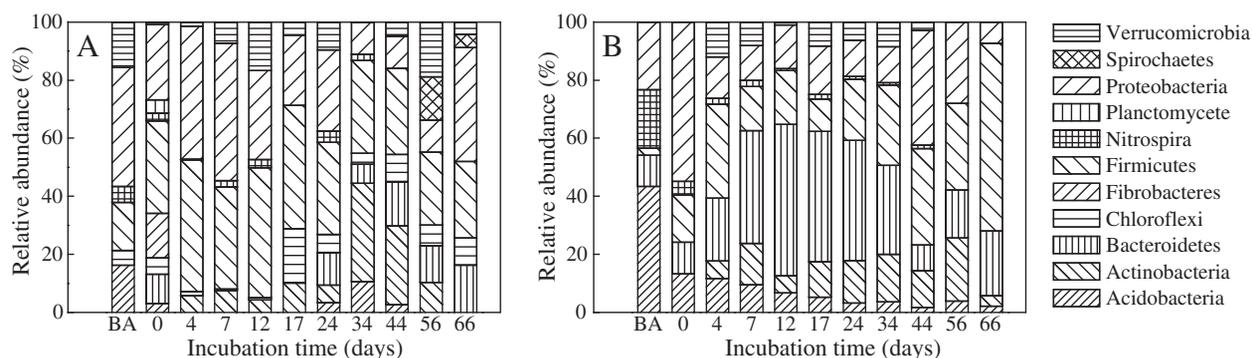
**Fig. 4 – Cluster analysis of DGGE patterns of bacterial communities in non-flooded (A) and flooded (B) SW treatments during 66-day incubation. BA, samples at day 0 before amendments in the two treatments. DGGE: denaturing gradient gel electrophoresis.**

**Table 4 – Closest relatives of the DGGE bands in two treatments.**

Non-flooded SW				Flooded SW			
Phylogenetic group (closest relative)	Band no.	Accession no.	Similarity (%)	Phylogenetic group (closest relative)	Band no.	Accession no.	Similarity (%)
Acidobacteria				Acidobacteria			
<i>Acidipila rosea</i>	38	KP277157	96	<i>Acidicapsa ligni</i>	12'	KP277163	93
	14	KP277127	93	<i>Bryobacter aggregatus</i>	43'	KP277204	93
	6	KP277150	94	<i>Holophaga foetida</i>	24'	KP277168	85
Actinobacteria				Actinobacteria			
<i>Adlercreutzia equolifaciens</i>	9	KP277146	89	<i>Corynebacterium callunae</i>	38'	KP277195	84
<i>Gaiella occulta</i>	17	KP277137	97	<i>Gaiella occulta</i>	35'	KP277178	97
	18	KP277138	96	<i>Lechevalieria atacamensis</i>	46'	KP277206	99
Bacteroidetes				Bacteroidetes			
<i>Alkaliflexus imshenetskii</i>	16	KP277154	90	<i>Alkaliflexus imshenetskii</i>	7'	KP277187	91
<i>Chryseobacterium daecheongense</i>	1	KP277156	90	<i>Chitinophaga pinensis</i>	6'	KP277162	96
<i>Mucilaginibacter angelicae</i>	33	KP277133	95	<i>Flavitalea populi</i>	16'	KP277175	95
<i>Chloroflexi</i>				<i>Mucilaginibacter boryungensis</i>	33'	KP277201	94
<i>Anaerolinea thermophila</i>	37	KP277135	88	<i>Mucilaginibacter oryzae</i>	27'	KP277170	98
<i>Caldilinea aerophila</i>	13	KP277136	87	<i>Nonlabens dokdonensis</i>	36'	KP277173	92
Firmicutes				<i>Paludibacter propionigenes</i>	32'	KP277194	96
<i>Fibrobacter succinogenes</i> subsp. <i>elongatus</i>	26	KP277131	88	<i>Solitalea koreensis</i>	14'	KP277188	87
Firmicutes				<i>Tenacibaculum jejuense</i>	10'	KP277185	90
<i>Bacillus niacini</i>	25	KP277130	98	Firmicutes			
<i>Caloramator mitchellensis</i>	19	KP277152	94	<i>Anaerostipes butyraticus</i>	47'	KP277207	99
<i>Clostridium acetobutylicum</i>	15	KP277147	98	<i>Anaerobacter polyendosporus</i>	31'	KP277192	100
	21	KP277149	99	<i>Bacillus berkeleyi</i>	18'	KP277189	96
<i>Clostridium hungatei</i>	10	KP277151	85	<i>Bacillus niacini</i>	21'	KP277186	99
<i>Clostridium puniceum</i>	27	KP277132	100	<i>Bacillus persicus</i>	4'	KP277165	99
	36	KP277134	100		9'	KP277166	99
<i>Clostridium sticklandii</i>	4	KP277125	88		34'	KP277177	99
<i>Oceanirhabdus sediminicola</i>	34	KP277142	97	<i>Bacillus vireti</i>	23'	KP277193	100
<i>Desulfotobacterium dehalogenans</i>	23	KP277122	96	<i>Butyrivibrio fibrisolven</i>	28'	KP277171	89
<i>Desulfotobacterium metallireducens</i>	30	KP277140	93	<i>Clostridium aciditolerans</i>	40'	KP277180	99
<i>Propionispora hippei</i>	2	KP277144	92	<i>Clostridium cavendishii</i>	37'	KP277179	92
Nitrospira				<i>Clostridium hungatei</i>	20'	KP277176	99
<i>Nitrospira japonica</i>	40	KP277159	98	<i>Clostridium polysaccharolyticum</i>	1'	KP277181	100
Planctomycete				<i>Clostridium sticklandii</i>	5'	KP277199	88
<i>Desulfovibrio alaskensis</i>	24	KP277129	85	<i>Desulfotobacterium metallireducens</i>	29'	KP277191	93
Proteobacteria				<i>Eisenbergiella tayi</i>	39'	KP277196	98
<i>Azoarcus indigens</i>	28	KP277123	96	<i>Paenibacillus alginolyticus</i>	19'	KP277190	99
<i>Burkholderia ambifaria</i>	20	KP277148	100	<i>Thermincola carboxydiphila</i>	8'	KP277184	95
<i>Desulfovibrio marinus</i>	39	KP277158	87	Nitrospira			
<i>Desulfovibrio oxamicus</i>	31	KP277141	88	<i>Nitrospira japonica</i>	44'	KP277205	97
<i>Novosphingobium fuchskuhlense</i>	8	KP277155	99	Proteobacteria			
<i>Novosphingobium lindaniclasticum</i>	42	KP277160	96	<i>Desulfonatronum alkalitolerans</i>	22'	KP277183	86
<i>Ochrobactrum anthropi</i>	41	KP277161	100	<i>Desulfovibrio marinus</i>	45'	KP277208	87
<i>Pantoea anthophila</i>	22	KP277128	98	<i>Dokdonella fugitiva</i>	11'	KP277197	98
<i>Pantoea rwandensis</i>	3	KP277120	99	<i>Methylobacterium radiotolerans</i>	2'	KP277182	99
<i>Pseudomonas entomophila</i>	5	KP277145	99	<i>Nitrosospira multiformis</i>	3'	KP277198	93
<i>Rhizomicrobium electricum</i>	35	KP277143	98	<i>Pantoea rwandensis</i>	42'	KP277203	98
<i>Sphingomonas roseiflava</i>	29	KP277139	100	<i>Pseudomonas mediterranea</i>	13'	KP277167	99
<i>Wolinella succinogenes</i>	11	KP277121	88		30'	KP277172	100
<i>Yersinia kristensenii</i>	12	KP277126	99	<i>Pseudomonas putida</i>	17'	KP277200	99
Spirochaetes				<i>Rahnella aquatilis</i>	26'	KP277169	98
<i>Treponema caldaria</i>	7	KP277153	93	<i>Sphingobium suberifaciens</i>	25'	KP277164	99
Verrucomicrobia				<i>Sphingomonas wittichii</i>	41'	KP277202	100
<i>Methylacidimicrobium tartarophylax</i>	32	KP277124	83	Verrucomicrobia			
				<i>Chthoniobacter flavus</i>	15'	KP277174	84

(ethene, i-propanol, 2-butanol and 2-butanone), between *Pantoea anthophila* and acetaldehyde, and between *Rhizomicrobium electricum* and 2-butanone, but a substantially negative correlation of *Gaiella occulta* to 2-pentanone. In the flooded SW treatment, significantly positive correlations were found between *Bacillus persicus* and two VOCs (ethene and ethanol),

between *Clostridium cavendishii* and three VOCs (ethene, ethanol and acetaldehyde), between *Chthoniobacter flavus* and propene, between *Flavitalea populi* and four VOCs (ethene, propene, 2-butanol and 2-butanone), between *Holophaga foetida* and three VOCs (ethene, ethanol and acetaldehyde), and between *Mucilaginibacter oryzae* and acetaldehyde, whereas negative



**Fig. 5 – Relative abundance of 16S ribosomal ribonucleic acid (rRNA) gene phylotypes before and after amendments in non-flooded (a) and flooded (b) SW treatments. BA, samples at day 0 before amendments.**

correlations were observed between *Lechevalieria atacamensis* and acetaldehyde, and between *Sphingobium suberifaciens* and *i*-propanol, and no significant correlation of *Gaiella occulta* and *Paludibacter propionigenes* with all top 10 VOCs was found.

As reported by previous studies, positive correlations can occur through VOC production from microbes and/or enhancement of microbial growth by VOCs (Mayrhofer et al., 2006; Ramirez et al., 2010). Species of *Bacillus*, *Clostridium*, *Desulfotobacterium*, *Desulfovibrio* and *Pantoea* were reported to emit predominantly alkenes, alcohols, esters, ketones, aldehydes, fatty acids, ethers and sulfur-containing compounds (reviewed by Rappert and Müller, 2005), which might result in their significantly positive correlations with most of the top 10 VOCs in the two treatments. For instance, *Bacillus licheniformis* produced a considerable amount of acetaldehyde, ethanol, acetate and lactate during anaerobic fermentation in chemostats and batch-cultures (Bulthuis et al., 1991). *Desulfotobacterium dichloroeliminans* strain DCA1 isolated from a 1,2-dichloroethane-polluted soil matrix selectively converted 1,2-dichloroethane and all possible vicinal dichloropropanes and dichlorobutanes into completely dechlorinated end products, such as ethene, propene and butene (De Wildeman et al., 2003). The genus *Desulfovibrio*, together with other *Dehalobacter* inoculated with groundwater in the reactive iron barrier, reduced 1,2-dichloroethane to ethene (Zemb et al., 2010), and *Desulfovibrio* IsBdl, isolated from a sand sample of Lake Kasumigaura, oxidized 2-propanol to acetone and 2-butanol to 2-butanone (Tanaka, 1992). The spoilage bacteria *Pantoea agglomerans*, grown on mixed-lettuce agar, metabolized sugars to produce a range of VOCs, including ethanol, 2-methyl-1-propanol, 2-methyl-1-butanol, 3-methyl-1-butanol, 2,3-butanedione, 1-butanol, 3-methyl-1-pentanol and 1-hexanol during storage at 7°C in air and a low-oxygen atmosphere (Ragaert et al., 2006). On the other hand, negative correlations may also result if VOCs are consumed by microorganisms and/or if the growth and activity of VOC-producing microbes are inhibited by other microorganisms (Mayrhofer et al., 2006; Ramirez et al., 2010; Asensio et al., 2012). *Sphingobium* sp. was reported to employ ethanol or *i*-propanol as sacrificial electron donors when it participates in the regeneration of reduced nicotinamide cofactors in alcohol dehydrogenase (ADH)-catalyzed reactions (Kara et al., 2014), which might lead to negative correlations between *Sphingobium suberifaciens* and *i*-propanol in the flooded SW treatment. For *Gaiella occulta* in the

non-flooded SW treatment and *Lechevalieria atacamensis* in the flooded SW treatment, their negative correlations with most of the top 10 VOCs could be explained by the fact that actinomycetes can produce antibiotics, which are primarily active against bacteria (reviewed by Bérdy, 2005), and thus inhibit the bacterial VOC production. For example, Furumai et al. (2002) reported that kosinostatin, a quinocycline antibiotic with antitumor activity from an actinomycete strain (*Micromonospora* sp. TP-A0468), strongly inhibited the growth of Gram-positive bacteria (MIC = 0.039 µg/mL), and Gram-negative bacteria and yeasts moderately (MIC = 1.56 similar to 12.5 µg/mL). Sun et al. (2007) also found that chemomicin A, an angucyclinone antibiotic produced by actinomycete strains (*Nocardia mediterranei* subsp. *kanglensis* 1747–64), had antimicrobial activity against *Enterococcus faecium*, *Bacillus subtilis* and *Proteus vulgaris*, with MIC values of 120, 60 and 60 mmol/L, respectively.

### 3. Conclusions

The present work was designed to investigate the VOC emissions from straw-amended agricultural soils and their relations to the bacterial communities in non-flooded and flooded SW treatments. Our results indicated that the total VOC emissions from the amended soils averaged 5924 and 2211 ng C kg/hr in non-flooded and flooded SW treatments, respectively. Ethene, propene, ethanol, *i*-propanol, 2-butanol, acetaldehyde, acetone, 2-butanone, 2-pentanone and acetophenone were the top 10 VOCs, and together accounted for over 90% of the total VOCs released in the two treatments. One “peak emission window” presented at days 0–44 or 4–44, and over 95% of the VOC emissions occurred during the first month in the two treatments. The DGGE analysis demonstrated that the fractions of *Actinobacteria*, *Bacteroidetes*, *Firmicutes* and  $\gamma$ -*Proteobacteria* increased, but the fractions of *Acidobacteria* decreased with incubation time after straw return in the two treatments. Cluster analysis showed that the alteration of the bacterial communities was consistent with the change of the VOC emissions in two treatments. Correlation analysis revealed that the top 10 VOCs correlated positively with *Bacillus niacin*, *Caldilinea aerophila*, *Desulfotobacterium dehalogenans*, *Desulfotobacterium metallireducens*, *Desulfovibrio oxamicus*, *Methylacidimicrobium tartarophylax*, *Oceanirhabdus sediminicola*, *Pantoea anthophila* and *Rhizomicrobium*

**Table 5 – Correlation between the fluxes of top 10 VOCs and the intensities of dominant DGGF bands in two treatments. a**

Treatments	Closest relatives (band no.) <sup>a</sup>	Ethene	Propene	Ethanol	i-Propanol	2-Butanol	Acetaldehyde	Acetone	2-Butanone	2-Pentanone	Acetophenone
Non-flooded SW	<i>Bacillus niacini</i> (25)	0.780*	-0.318	0.436	0.683	0.725*	0.729*	0.399	0.653	0.637	-0.196
	<i>Calditinea aerophila</i> (13)	-0.376	-0.091	-0.318	-0.213	-0.183	-0.382	-0.150	-0.147	0.247	0.896**
	<i>Desulfobacterium dehalogenans</i> (23)	0.234	0.748*	0.006	0.417	0.271	0.174	0.500	0.338	0.156	0.024
	<i>Desulfobacterium metallireducens</i> (30)	0.619	0.638	-0.285	0.760*	0.701	0.016	0.432	0.563	-0.012	-0.086
	<i>Desulfovibrio oxamicus</i> (31)	0.854**	0.150	-0.501	0.903**	0.865**	-0.121	0.904**	0.944**	0.212	-0.031
	<i>Gaiella occulta</i> (17)	-0.312	-0.166	-0.425	-0.443	-0.376	-0.636	-0.422	-0.433	-0.834**	-0.387
	<i>Methylacidimicrobium tartarophylax</i> (32)	-0.160	0.785*	-0.288	0.053	-0.086	-0.273	0.217	-0.015	-0.200	0.147
	<i>Oceanirhabdus sedimicola</i> (34)	0.945**	-0.060	-0.356	0.907**	0.943**	0.031	0.568	0.841	0.151	-0.233
	<i>Pantoea anthophila</i> (22)	0.647	-0.129	0.453	0.559	0.525	0.707*	0.528	0.584	0.553	-0.361
	<i>Rhizomicrobium electricum</i> (35)	0.680	-0.109	-0.486	0.696	0.699	-0.157	0.625	0.746*	0.263	0.065
	<i>Bacillus persicus</i> (34)	0.671*	0.590	0.825**	-0.030	0.563	0.574	0.241	0.400	0.400	-0.017
	<i>Clostridium cavendishii</i> (37')	0.778*	0.535	0.885**	-0.048	0.600	0.697*	0.162	0.373	0.373	-0.062
	<i>Chthoniobacter flavus</i> (15')	0.516	0.710*	0.255	-0.170	0.293	0.178	0.033	0.191	0.191	-0.252
	<i>Flavitailea populi</i> (16')	0.747*	0.891**	0.535	0.277	0.755*	0.412	0.545	0.706*	0.206	0.074
<i>Holophaga foetida</i> (24')	0.235	0.277	-0.280	-0.148	0.084	-0.406	-0.152	0.019	-0.300	-0.347	
<i>Lechevaleria atacamensis</i> (46')	-0.439	-0.336	-0.637	-0.459	-0.591	-0.669*	0.236	0.471	0.471	-0.004	
<i>Mucilaginibacter oryzae</i> (27')	0.379	0.013	0.592	0.314	0.416	0.780*	-0.612	-0.633	-0.514	-0.466	
<i>Paludibacter propionigenes</i> (32')	-0.099	0.471	-0.494	0.111	-0.045	-0.371	0.134	0.303	0.335	0.342	
<i>Sphingobium suberifaciens</i> (25')	0.001	-0.217	0.114	-0.728*	0.214	0.091	-0.252	0.106	0.106	0.047	
											-0.371
											0.188

<sup>a</sup> The closest relatives of bands were determined according to Table 4.

\* Significant correlation at  $p < 0.05$  level.

\*\* Significant correlation at  $p < 0.01$  level.

electricum in the non-flooded SW treatment and *Bacillus persicus*, *Clostridium cavendishii*, *Chthoniobacter flavus*, *Flavitalea populi*, *Holophaga foetida*, *Mucilaginibacter oryzae* and *Paludibacter propionigenes* in the flooded SW treatment, but negatively with *Gaiella occulta* in the non-flooded SW treatment and *Lechevalieria atacamensis* and *Sphingobium suberifaciens* in the flooded SW treatment. The results of this study could help us to gain a better understanding of the impact of agriculture on atmospheric VOCs, as well as the microbial mechanism of soil VOC emissions. However, it is worth noting that the results from this lab-based study, with straw-amended paddy soils incubated under controlled temperature and moisture conditions, cannot be fully used to predict the specific VOC emission rates and bacterial community structures in agricultural soils in the field; thus further study should be conducted on the VOC emissions from different types of agricultural soils amended with different straws and their associations with bacterial communities in the field.

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