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Prevention and Control of Ginger Blast by Two Fumigants and Their Effects on a Soil Bacterial Community and the Metabolic Components of Ginger

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Abstract: A two-year field trial was conducted in order to assess techniques to control ginger blast and explore the effects of fumigants on soil bacterial microorganisms and ginger metabolites. This study examined the effects of dazomet and chloropicrin on the control of ginger blast and their influence on ginger yield in Luoping County, Yunnan Province, China. The results showed that in 2022, the control effectiveness of dazomet and chloropicrin treatments on ginger blast was 84.33% and 94.67%, respectively. The corresponding yields were 50,154.40 kg/hm^2 and 50,296.90 kg/hm^2. In 2023, the control effectiveness of dazomet and chloropicrin treatments on ginger blast were 86.33% and 93.67%, respectively, and the yields were 65,115.83 kg/hm^2 and 65,337.93 kg/hm^2. In both years, the incidence of ginger blast in the control group reached nearly 100%, leading to the near extinction of the crop. Additionally, in 2023, 16S rRNA high-throughput sequencing and non-targeted metabolomics techniques were used to analyze the effects of the fumigants on soil bacterial microorganisms and the metabolites in ginger. The results showed that the diversity and richness of soil bacterial communities were lower than those in the control group at 0 and 120 days after treatment with two fumigants, but the relative abundances of beneficial bacteria such as *Pseudomonas* increased at 60 days, and the relative abundances of *Actinobacteria*, *Gemmatimonadetes*, and *Bacillus* increased at 120 days. The abundance of *Firmicutes* also increased after 120 days of chloropicrin treatment. The non-targeted metabolic LC–MS results showed that the production of phenols and terpenoids was upregulated after dazomet and chloropicrin treatments. The contents of amino acids and their derivatives were also upregulated. This upregulation of metabolites was beneficial to the flavor quality of ginger and enhanced its anti-inflammatory, anti-tumoral, and antioxidant effects.

Keywords: dazomet; chloropicrin; soil microbial community; metabolites; yield

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1. Introduction

Ginger (*Zingiber officinale* Roscoe), a perennial herb of the ginger family, holds high medicinal and edible value and is an important cash crop globally [1]. Many studies have reported that various compounds extracted from ginger have anti-cancer, antioxidant, anti-inflammatory, and other beneficial properties [2–5].

Ginger has been cultivated in China for over 2000 years. Today, China is a major player in the global ginger market, being one of the largest producers, consumers, and exporters of ginger [6]. With more than 2700 years of cultivation history, China has a rich variety of ginger types in terms of form, color, flavor quality, and yield traits. Among the cultivars,

little yellow ginger is particularly notable for its small tuber, high gingerol content, and greater medicinal value compared to ordinary ginger [7]. Luoping County in Yunnan Province in China is the core producing area for this high-quality ginger. In 2016, Luoping small yellow ginger was registered under China's National Geographical Indication Certification trademark. In 2020, it was included in the first batch of mutual recognition products listed in the China–EU Geographical Indication agreement and became a Sino–European geographical indication protected brand. However, with the increase in planting years and the low resistance of ginger to continuous cropping, Luoping is now facing the challenge of finding suitable land to grow small yellow ginger due to continuous cropping being an obstacle.

Ginger blast is one of the primary factors preventing the continuous cultivation of ginger and is the most severe disease affecting the ginger industry. In addition to the common characteristics of soil-borne diseases, ginger blast is highly infectious, aggressive, and devastating. Once ginger blast occurs in fields, the infection will spread rapidly, leading to a sharp reduction in ginger yield or to no yield at all [8,9]. Ginger blast is caused by *Ralstonia solanacearum*, a bacterium that lives in the soil. Contact chemical agents and biological agents cannot completely eradicate it because they struggle to penetrate the deep soil and distribute evenly. When the climate becomes favorable, the surviving *R. solanacearum* will rapidly multiply and infect ginger plants, resulting in another outbreak of ginger blast. Current cultivation practices, disease-resistant varieties, and fungicides are not effective at controlling ginger blast [10,11].

Soil fumigation is one of the most effective techniques for controlling ginger blast. It involves using fumigants that release gases into the soil, which can be deeply and evenly distributed to kill soil pathogens before the planting of crops [12]. Since 1950, methyl bromide, chloropicrin, metham-sodium, dazomet, and other fungicides have been used for soil fumigation around the world [13]. Currently, the commercially available soil fumigants in China include chloropicrin, dazomet, metham-sodium, dimethyl disulfide (DMDS), and allyl isothiocyanate (AITC) [14].

Dazomet and chloropicrin are both broad-spectrum soil fumigants widely used on fruit and vegetable crops, effective against nematodes, fungi, bacteria, and weeds [15–18]. Locascio et al. [19] found that dazomet had a notable effect on controlling soil-borne diseases of tomatoes, effectively inhibiting fungal growth, killing nematodes, and significantly increasing tomato yield. A study by de Cal et al. [20] showed that dazomet could effectively reduce the pathogenic fungus population in strawberry soil. Wang et al. [21] reported that chloropicrin treatment could help overcome the continuous cropping obstacle and promote the growth of *Panax notoginseng*. In another study, Wang et al. [22] found that chloropicrin could significantly reduce the relative abundances of pathogens in soil and improve the survival rate of continuously cropped *P. notoginseng*. Regarding the safety of fumigants, Cao et al. [23] concluded that chloropicrin does not have significant long-term effects on soil microbial communities and stated that chloropicrin and its degradation products have no teratogenic effects.

At present, dazomet and chloropicrin have achieved good results in the prevention and control of soil-borne diseases in various crops. This study introduces the novel approach of using these two fumigants to control ginger blast. It also examines the changes in the soil bacterial community during the cultivation process and the flavor quality of Luoping small yellow ginger after harvest. The findings of this study can provide a theoretical and scientific basis for the efficient use of soil fumigants to control ginger blast in the field, thereby improving the yield and flavor quality of Luoping small yellow ginger.

2. Materials and Methods

2.1. Test Materials

The ginger variety used in this study was Chinese Luoping small yellow ginger. The soil fumigants were 99.5% chloropicrin, produced by Dalian Lufeng Chemical Co., Ltd., Dalian, China, and 98% dazomet particulate agent, produced by Nantong Shizhuang

Chemical Co., Ltd., Nantong, China. The tarp covering the soil after fumigation was a 0.04 mm PVC tarp produced by Shandong Longxing Plastic Film Technology Co., Ltd., Weifang, Shandong, China.

2.2. Overview of the Test Site

The experimental site was located in a continuous ginger cropping field in Qingcaotang Village, Lushan Street, Luoping County, Qujing City, Yunnan Province, China (24.90472318° N, 104.33795941° E). The field test was conducted at two locations over two years. The physical and chemical properties of the test site are given in Table 1.

Table 1. Soil physical and chemical properties at different experimental sites.

Physical and Chemical Property	Test Site in 2022	Test Site in 2023
pH (water-to-soil ratio = 2.5:1)	5.58	6.14
Organic material (g/kg)	40.78	38.14
Moisture (%)	20.06	19.13
Ammonium nitrogen (mg/kg)	6.45	5.50
Total nitrogen (g/kg)	2.22	2.10
Available potassium (mg/kg)	487	348.00

2.3. Test Treatment

Over the two years of field trials, three treatments were established as the following: 75 g/m^2 of dazomet, 52 g/m^2 of chloropicrin, and a control group without any treatment. Three replicates were arranged for each treatment at each site. Each replicate area was 333 m^2. All replicate fields were randomly arranged, and deep trenches (50 cm deep and 30 cm wide) were dug between different replicates to ensure isolation. In 2022, the fumigation operation was carried out on 29 March and the PVC tarp was removed on 13 April. In 2023, the fumigation operation was carried out on 13 March, and the tarp was removed on 28 March. The fumigation operation was carried out according to the "NY/T3129-2017 dazomet soil disinfection technical specification" and "NY/T2725-2015 technical specification for chloropicrin soil disinfection".

Dazomet was mechanically mixed into the soil [24,25], and chloropicrin was administered using an injection method [26,27]. After fumigation, planting, fertilization, watering, and weeding were carried out according to conventional planting methods.

2.4. Sample Collection

In 2022, this study focused on the effect of the fumigants in controlling ginger blast and determining ginger yield. In 2023, experiments were conducted to verify the results, as well as to determine the effects of fumigation on the microflora in rhizospheric soil and the ginger quality (metabolome).

In 2023, rhizospheric soil was collected using a 5-point sampling method from the control site, the dazomet treatment site, and the chloropicrin treatment site, at 0, 60, and 120 days after tarp removal. The soil samples were transported in a dry ice box and then stored at −80 °C for 16S rRNA high-throughput sequencing. Information on the collected soil samples is given in Table 2.

Table 2. Serial number information for soil samples collected in 2023.

Treatment	0 Days after Tarp Removal	60 Days after Tarp Removal	120 Days after Tarp Removal
Dazomet	DA 1	DA 2	DA 3
Chloropicrin	PS 1	PS 2	PS 3
Control	CK 1	CK 2	CK 3

Regarding fresh ginger sample collection, on harvest day in 2023, 5 ginger rhizomes were randomly collected for each repeat for the control, dazomet treatment, and chloropicrin treatment, placed in a dry ice box, and taken back to the laboratory. The ginger rhizomes from each repeat were cut into small pieces and mixed evenly. A 0.5 kg sample from each mixture was taken and stored in the refrigerator at −80 °C for metabolomics analysis.

2.5. Investigation of Ginger Blast and Yield

In 2022 and 2023, the incidence of ginger blast was investigated at the early stage of ginger blast occurrence, at the outbreak, and at the harvest stage. A 5-point sampling method was adopted for each replicate, with 20 ginger plants investigated at each point. A total of 300 ginger plants were investigated for each treatment.

$$\text{Incidence} = \text{Number of infected plants/total number of investigated plants} \times 100\%.$$

$$\text{Prevention and treatment effectiveness} = (\text{incidence in control area} - \text{incidence in treatment area})/\text{incidence in control area} \times 100\%.$$

The investigation of small yellow ginger agronomic characteristics and yield was carried out on harvest day. The traits investigated included the plant height, stem diameter, branch number, and fresh weight of the ginger rhizome per plant. The investigation involved a 5-point sampling method for each repeat and taking 2 plants from each point for measurements.

For yield determination, the fresh weight of ginger rhizomes was determined by sampling all plants within 10 m^2 at each location. The final yield (kg/hm^2) was adjusted according to the incidence of ginger blast in the fields. The ginger rhizomes were graded according to the CODEX standard [28].

We summarized and organized the collected data, and the significance of differences in the data for the ginger blast, agronomic trait, and yield were tested by one-way analysis of variance (ANOVA) with SPSS 26.0 software, Duncan's new multiple range method, and Student's *t*-test ($p = 0.05$). Plots were generated using GraphPadPrism 9 software.

2.6. Sequencing and Data Analysis of the 16S rRNA of Soil Microorganisms

For 16S rRNA sequencing, the total DNA of soil microorganisms was extracted using an OMEGA Soil DNA Kit (M5635-02) (Omega Bio-Tek, Norcross, GA, USA). The 16S rRNA V3–V4 region was amplified using the bacterial universal primer sequences 338F (5′-ACTCCTACGGGAGGCAGCA-3′) and 806R (5′-GGACTACHVGGGTWTCTAAT-3′). After recovery and purification, the PCR products were sent to Shanghai Baipu Biotechnology Co., Ltd. (Shanghai, China), for high-throughput sequencing on the Illumina MiSeq platform.

For data analysis, after sequencing, the DADA2 plugin was used for quality filtering, denoising, merging, and chimera removal. Then, 97% of non-chimera sequences were clustered, and OTU representative sequences and OTU tables were generated. The alpha diversity index was calculated using QIIME2 (2019.4) to compare the diversity and richness of different samples. Principal coordinate analysis (PCoA) was performed with R (4.0.3) and QIIME2 (2019.4) software to study the soil bacterial community compositions under different treatments according to the weighted UniFrac algorithm.

2.7. Determination and Data Analysis of the Metabolome of Small Yellow Ginger

For metabolome determination, the non-targeted metabolomic testing of small yellow ginger was conducted by Shanghai Baipu Biotechnology Co., Ltd.; a SHIMADZU-LC30 ultra-high performance liquid chromatography system (UHPLC) was used to isolate the metabolome. Chromatography was performed on an ACQUITY UPLC® HSS T3 (2.1 × 100 mm, 1.8 μm) column (Waters, Milford, MA, USA) with a QE Plus mass spectrometer (Thermo Scientific, MA, USA). Each sample was analyzed in both positive ion (+) and negative ion (−) modes using electrospray ionization (ESI), with ionization performed using a HESI source.

For data analysis, MSDIAL (4.9) software was used for peak alignment, retention time correction, and peak area extraction for the raw data. Metabolite structures were identified by precise mass number matching (mass tolerance < 10 ppm) and secondary spectrum matching (mass tolerance < 0.01 Da). Public databases such as HMDB, MassBank, GNPS, and self-established BP-DB were searched. The total peak area of the positive and negative ion data was normalized, and the peaks of positive and negative ions were integrated. Pattern recognition was carried out using the Python programming language. After processing the data by unit variance (UV) scaling, subsequent data analysis was conducted.

3. Results and Analysis

3.1. Effects of Two Fumigation Treatments on the Control of Ginger Blast in the Field

The incidence of ginger blast in 2022 was surveyed on 15 July, 15 August, and 12 November. The incidence of ginger blast in 2023 was surveyed on 20 July, 20 August, and 13 November.

Figure 1a shows the incidence of ginger blast in 2022. Figure 1b shows the control effectiveness of the two fumigants against ginger blast. On 15 July 2022, the incidence of ginger blast in the control group was 26.67%. The incidence in the dazomet and chloropicrin treatment groups was 1.67% and 1.33%, respectively. There was no significant difference between the two fumigation treatments ($p = 0.861$), but both were significantly different compared with the control group ($p < 0.001$). On 15 August 2022, the incidence of ginger blast in the control group was 49.33%. The incidence in the dazomet and chloropicrin treatment groups was 8.67% and 2.67%, respectively, and both were significantly different compared with the control group ($p < 0.001$); there was also a significant difference between the two fumigation treatments ($p = 0.01$). On 12 November 2022, during the harvest period, the incidence of ginger blast in the control site reached 100%, resulting in complete crop loss. The incidence in the dazomet treatment and chloropicrin treatment groups was 15.67% ($p < 0.001$) and 5.33% ($p < 0.001$), respectively, at that time; both values were significantly different compared with the control group. The control effects of dazomet and chloropicrin on ginger blast were 84.33% and 94.67%, respectively, and the difference between them was significant ($p < 0.001$).

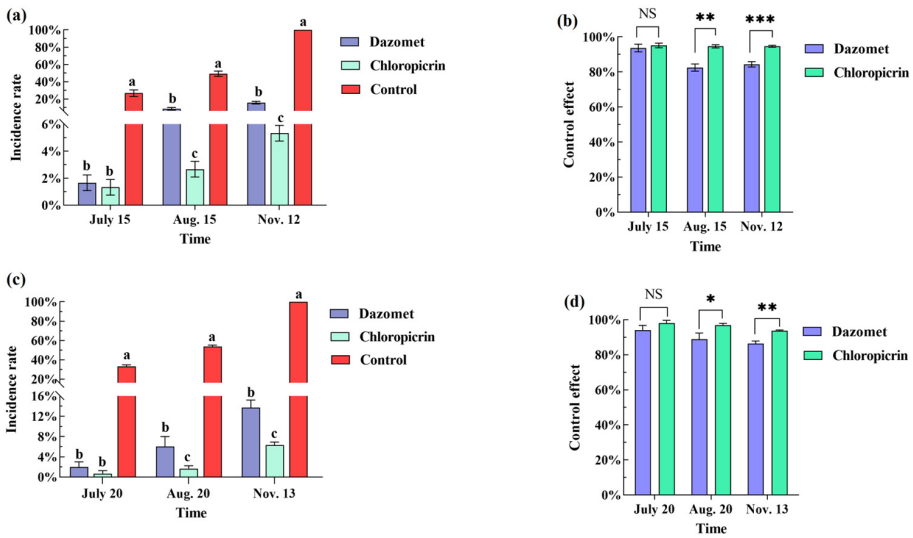


Figure 1. Incidence and control effects of ginger blast after fumigation with dazomet and chloropicrin: (**a**) incidence of ginger blast after treatment with two fumigants in 2022; (**b**) efficacy of two fumigants against ginger blast in 2022; (**c**) incidence of ginger blast after treatment with two fumigants in 2023; (**d**) efficacy of two fumigants against ginger blast in 2023. Different lowercase letters indicate significant differences between treatments (* $p < 0.05$). Double asterisks (**) mean $p < 0.01$, have significant difference; triple asterisks (***) mean $p < 0.001$, have very significant difference; NS means no significant difference.

Figure 1c,d show the incidence of ginger blast in 2023 and the control effectiveness of the two fumigants against ginger blast. On 20 July 2023, the incidence in the control group was 33.00%. The incidence in the dazomet and chloropicrin treatment groups was 2.00% ($p < 0.001$) and 0.67% ($p < 0.001$), respectively, which were both significantly lower than that of the control group, but there was no significant difference between the two fumigation treatments ($p = 0.223$). On 20 August 2023, the incidence in the control group was 53.67% and the incidence in the dazomet treatment group was 6.00%, showing significant difference from the control group ($p < 0.001$). The incidence in the chloropicrin treatment group was 1.67%, which was significantly different from that of the control group ($p < 0.001$). There was a significant difference between the two fumigation treatments ($p = 0.012$). On 13 November 2023, the incidence in the control group was 100%, the incidence in the dazomet treatment group was 13.67% ($p < 0.001$), and the incidence in the chloropicrin treatment was 6.33% ($p < 0.001$). Both were significantly different compared with the control group. There were also significant differences between the two fumigation treatment groups ($p < 0.001$). The control effects of dazomet and chloropicrin were 86.33% and 93.67%, respectively; they were significantly different between the two treatment groups ($p = 0.002$). The results of two years of field experiments showed that both fumigants were effective against ginger blast, with chloropicrin demonstrating a better control effect than dazomet.

3.2. Effects of Two Fumigation Treatments on Agronomic Traits and Yield of Ginger

On 12 November 2022 and 13 November 2023, the harvest days for ginger, the incidence of ginger blast in the control area had reached nearly 100%, resulting in almost no harvest. Therefore, no agronomic trait or yield evaluation was carried out for the control area.

Tables 3 and 4 show the agronomic traits and yields of ginger in 2022. In terms of agronomic traits, the plant height and fresh weight of ginger rhizomes per plant treated with dazomet in 2022 were significantly better than those treated with chloropicrin. Other indices were not significantly different between the two treatments. Considering the incidence of ginger blast under different treatments in 2022, there was no significant difference between the corrected yields of dazomet and chloropicrin treatments, which were 50,154.40 kg/hm^2 and 50,296.90 kg/hm^2, respectively.

Table 3. Effects of dazomet and chloropicrin treatments on agronomic traits of ginger in 2022.

Treatment	Agronomic Trait			
	Plant Height (cm)	**Stem Diameter (cm)**	**Number of Branches (pcs)**	**Fresh Weight of Rhizomes (kg)**
Dazomet	87.07 ± 0.45	1.04 ± 0.01	11.83 ± 0.15	0.92 ± 0.02
Chloropicrin	83.10 ± 0.53	1.03 ± 0.01	11.67 ± 0.50	0.86 ± 0.03
t	−9.882	−2	−0.549	−3.043
p-value	0.001	0.116	0.612	0.038

Table 4. Effects of dazomet and chloropicrin treatments on yield of ginger in 2022.

Treatment	Corrected Yield (kg/hm^2)		
	First-Grade Rhizome Yield	**Second-Grade Rhizome Yield**	**Total Yield**
Dazomet	43,391.17 ± 135.44	6763.27 ± 84.35	50,154.40 ± 106.61
Chloropicrin	43,651.10 ± 511.51	6841.47 ± 54.62	50,296.90 ± 218.68
t	−0.851	−1.348	−1.015
p-value	0.443	0.249	0.368

Tables 5 and 6 show the agronomic traits and yields of small yellow ginger in 2023. The single-rhizome fresh weight of the dazomet treatment group was significantly better than that of the chloropicrin treatment group. The other indices showed no significant difference. After adjusting for the ginger blast incidence rates, there was no significant

difference between the corrected yields in the dazomet and chloropicrin treatment groups, which were 65,115.83 kg/hm^2 and 65,337.93 kg/hm^2, respectively.

Table 5. Effects of dazomet and chloropicrin treatments on agronomic traits of ginger in 2023.

Treatment	Agronomic Trait			
	Plant Height (cm)	Stem Diameter (cm)	Number of Branches (pcs)	Fresh Weight of Rhizomes (kg)
Dazomet	95.23 ± 0.91	1.13 ± 0.06	11.87 ± 0.47	1.08 ± 0.04
Chloropicrin	93.80 ± 0.89	1.10 ± 0.01	11.47 ± 0.31	0.99 ± 0.01
t	1.955	0.985	1.231	4.372
p-value	0.122	0.380	0.286	0.012

Table 6. Effects of dazomet and chloropicrin treatments on yield of ginger in 2023.

Treatment	Corrected Yield (kg/hm^2)		
	First-Grade Rhizome Yield	Second-Grade Rhizome Yield	Total Yield
Dazomet	55,642.57 ± 623.58	9473.30 ± 345.49	65,115.83 ± 799.74
Chloropicrin	55,708.67 ± 1041.31	9036.03 ± 180.05	65,337.93 ± 859.57
t	−0.094	1.944	−0.328
p-value	0.929	0.124	0.760

The results of field trials in 2022 and 2023 showed that both dazomet and chloropicrin treatments could effectively improve the agronomic traits and yields of small yellow ginger. The single-rhizome fresh weight after dazomet treatment was significantly higher than that after chloropicrin treatment. However, because the incidence of ginger blast in the dazomet group was higher than that in the chloropicrin group, there was no significant difference in yield between the two fumigation treatments after correction.

3.3. Effects of Different Treatments on Soil Bacterial Community Diversity

3.3.1. Sparse Curve

A total of 2,584,226 valid sequences were obtained from all soil samples after denoising, and 1,622,093 high-quality sequences were obtained after removing chimeras. Figure 2 shows the sparse curves for all samples. The flatness of the curves indicates whether the sequencing depth was sufficient enough to capture the diversity of the bacterial communities in the samples. The flatter the curves become, the more likely it is that the sequencing depth was adequate to reflect the true diversity present in the samples. As shown in the figure, the curve tends to flatten out at a certain sequencing depth, suggesting that the sequencing was sufficient at reflecting the diversity of bacterial communities and could be used for subsequent analysis.

3.3.2. Venn Diagram

In microbial diversity analysis, each OTU represents a species. The OTU partitioning of all sequences can be used to observe the differences in the number of species among different samples. As shown in Figure 3, at 0 day after fumigation, there were 6014, 232, and 5363 unique bacterial OTUs in the control (CK), dazomet (DA), and chloropicrin (PS) groups. There were 23 common bacterial OTUs shared among the three groups. At 60 days after fumigation, there were 2912, 7093, and 5383 unique bacterial OTUs in the CK, DA, and PS groups, respectively. The number of OTUs in the CK decreased significantly, while it increased significantly in the DA group and remained relatively unchanged in the PS group. The total number of common bacterial OTUs in the three groups increased to 146. At 120 days after fumigation, there were 7693, 4086, and 2976 unique bacterial OTUs in the CK, DA, and PS groups, respectively, with significant differences among the three groups. The number of unique OTUs increased in the CK group and decreased in the DA and PS groups, and the total number of common bacterial OTUs continued to rise to 257.

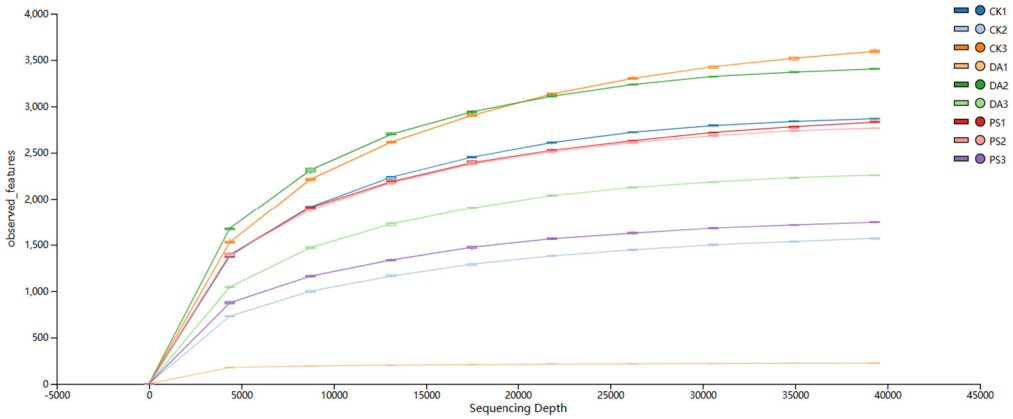


Figure 2. Sparse curve for bacterial communities in soil samples after fumigation with dazomet and chloropicrin.

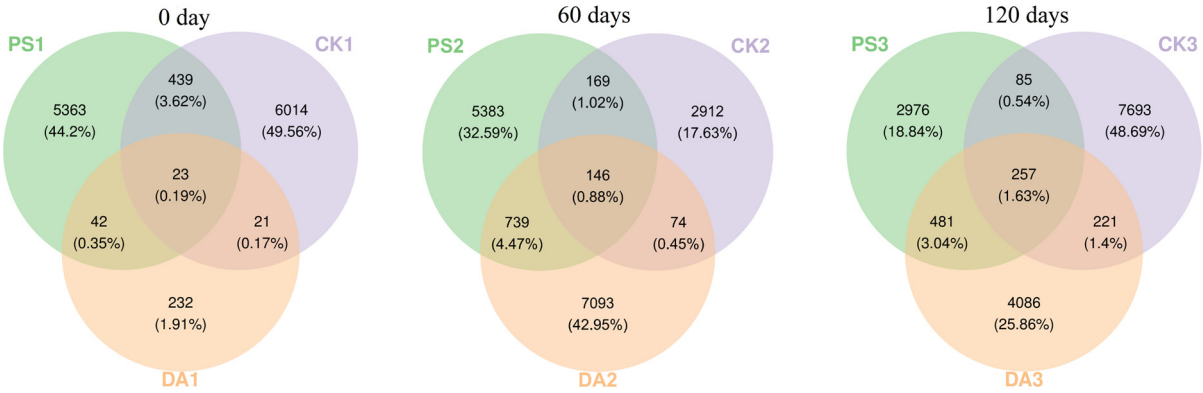


Figure 3. Venn diagram of OTU classifications of bacterial communities in soil samples after fumigation with dazomet and chloropicrin. Note: Different petals represent different samples. The overlapping areas represent the common OTUs shared among samples, and the non-overlapping areas represent the OTUs unique to the corresponding sample.

The resulting Venn diagram showed that the OTU count of endemic bacteria in soil treated with dazomet and chloropicrin was lower than that of the control at 0 day and 120 days after fumigation, but the OTU count of soil-specific bacteria under both treatments was higher than that in the control at 60 days. Over time, the common bacterial OTUs of the three treatments showed an increasing trend, and the difference in species numbers tended to decrease.

3.3.3. Alpha Diversity of Soil Bacterial Communities

To evaluate the alpha diversity of microbial communities, the Shannon index was used to characterize the diversity, and the Chao index was used to characterize the community richness. As shown in Figure 4, at 0 day after fumigation, the Shannon index and Chao index were lower under the dazomet and chloropicrin treatments compared to the control group, although there was no significant difference between the chloropicrin treatment and the control. At 60 days, the Shannon index and Chao index were significantly higher in the dazomet and chloropicrin treatment groups compared to the control, with a significant difference in the Chao index between the two fumigation treatments. At 120 days, compared with the control, both the Shannon index and Chao index of the two fumigation treatments decreased significantly, with a significant difference in the Shannon index between the two fumigation treatments, but no significant difference in the Chao index.

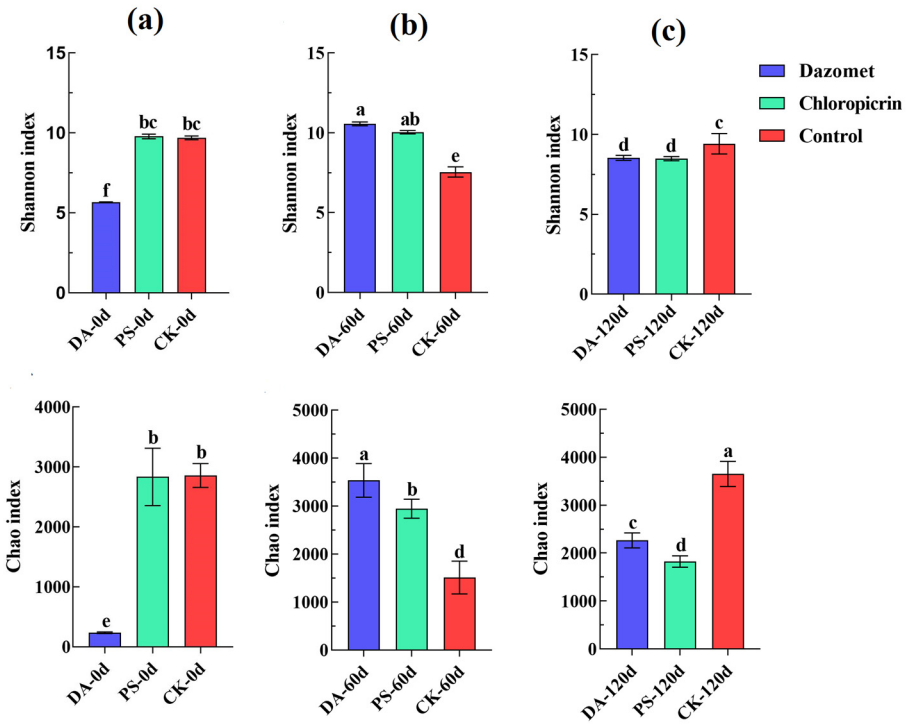


Figure 4. Effects of fumigation with dazomet and chloropicrin treatments on the alpha diversity of bacterial communities: (**a**) 0 day after fumigation; (**b**) 60 days after fumigation; (**c**) 120 days after fumigation. Different lowercase letters indicate significant differences between treatments ($p < 0.05$).

The results of the alpha diversity assessment showed that the two fumigants significantly affected the diversity and richness of the soil bacterial community. Compared with the control group, the diversity and richness of the soil bacterial community decreased at 0 and 120 days and increased at 60 days.

3.3.4. Beta Diversity of Soil Bacterial Communities

To further reveal the effects of different fumigation treatments on the composition of soil bacterial communities, the differences or similarities between microbial communities were visualized using beta diversity, and a PCoA analysis was conducted based on the weighted UniFrac distance. As shown in Figure 5, Axis1 and Axis2 explain 34.7% and 19.5% of the differences in sample composition, respectively. At 0 day, the CK group was clearly separated from the DA and PS groups along Axis2, and the DA group was clearly separated from the PS group along Axis1. At 60 days, the CK group was separated from the DA and PS groups along Axis1, and the DA group was separated from the PS group along Axis2. At 120 days, the CK group was separated from the DA and PS groups along Axis1. However, there was no significant difference in the distance between the two fumigation groups at that time.

The beta diversity results showed that both fumigation treatments altered the composition of the bacterial community structure. The structural composition of the bacterial community after chloropicrin and dazomet treatments differed from that of the control at 0, 60, and 120 days. The community structure composition of the dazomet and chloropicrin treatment groups differed at 0 and 60 days, but became closer at 120 days, indicating that the community composition between the two fumigation treatments tended to be similar at 120 days as time progressed.

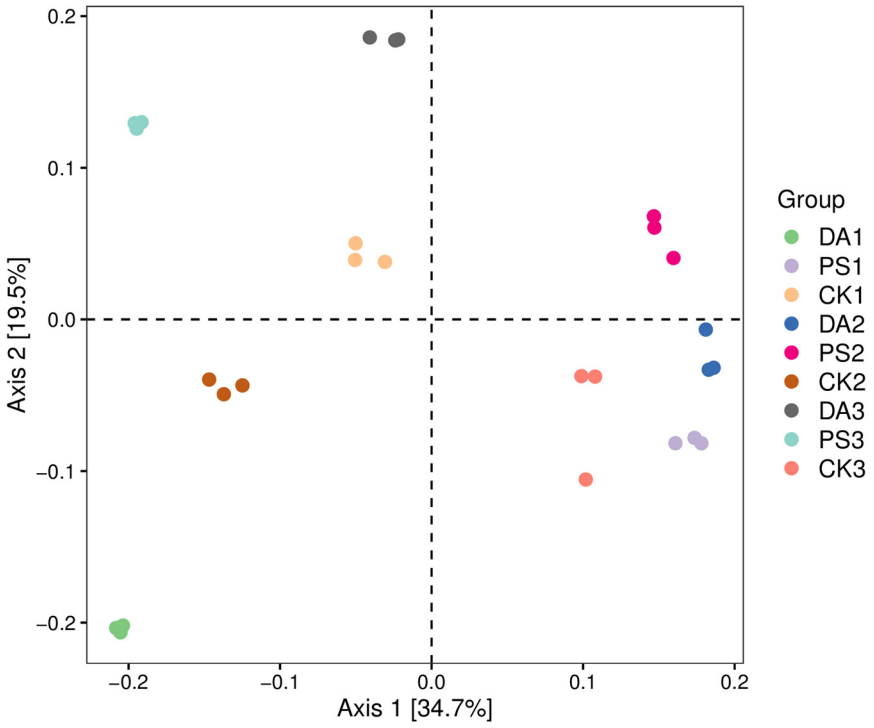


Figure 5. Principal coordinate analysis (PCoA) of soil bacterial communities after fumigation with dazomet and chloropicrin.

3.3.5. Changes in the Composition of Soil Bacterial Communities at the Phylum and Genus Levels

Stacked bar charts of the bacterial community composition of the soil samples from different treatments at the phylum level are shown in Figure 6. *Actinobacteria*, *Proteobacteria*, *Gemmatimonadetes*, *Firmicutes*, *Chloroflexi*, and *Acidobacteria* were the dominant phyla in all samples. At 0 day after fumigation, compared with the CK group, the relative abundances of *Actinobacteria* and *Firmicutes* in the DA- and PS-treated soil increased, but it decreased at 60 days. Specifically, compared to the CK at 60 days, the *Actinobacteria* and *Firmicutes* relative abundances decreased by 9.71% and 90.65% in the DA treatment, and by 8.91% and 83.27% in the PS treatment, respectively. At 0 day in the PS treatment group, the relative abundances of *Chloroflexi* and *Acidobacteria* increased by 196.74% and 7.44%, respectively. The relative abundance of *Proteobacteria* treated with DA and PS was lower than that of the CK at all sampling times, indicating that this bacterial group was more sensitive to DA and PS fumigants. At 120 days, the relative abundances of *Actinobacteria* and *Gemmatimonadetes* were higher in both the DA and PS treatments compared to the CK group, while the relative abundance of *Firmicutes* was also higher in the PS treatment group than in the CK group.

Stacked bar charts of the bacterial community composition of soil samples from different treatments classified at the genus level are shown in Figure 7. At the genus level, Unclassified-*Micromonosporaceae*, Unidentified-Ellin5301, Unidentified-*Gaiellaceae*, Unidentified-Micromonosporaceae, and *Bacillus* were the dominant species. At 0 day after fumigation, Unclassified-*Micromonosporaceae* decreased by 85.21% in the PS treatment compared with the CK; after DA treatment, it increased by 224.77% compared with the CK. At 60 days, the abundance of Unclassified-*Micromonosporaceae* was lower than that of CK after DA treatment, and it was higher than CK after PS treatment. The abundance of Unidentified-Ellin5301 decreased at 0 day after PS treatment and disappeared after DA treatment but recovered at 60 days. The relative abundance of Unidentified-*Micromonosporaceae* was higher than that of the CK at 0 day and lower than that of the CK at 60 days. At 120 days, the relative abundance of Unidentified-*Gaiellaceae* in both fumigation treatments

was lower than that in the CK, and the relative abundance of the other four dominant genera was higher than that in the CK.

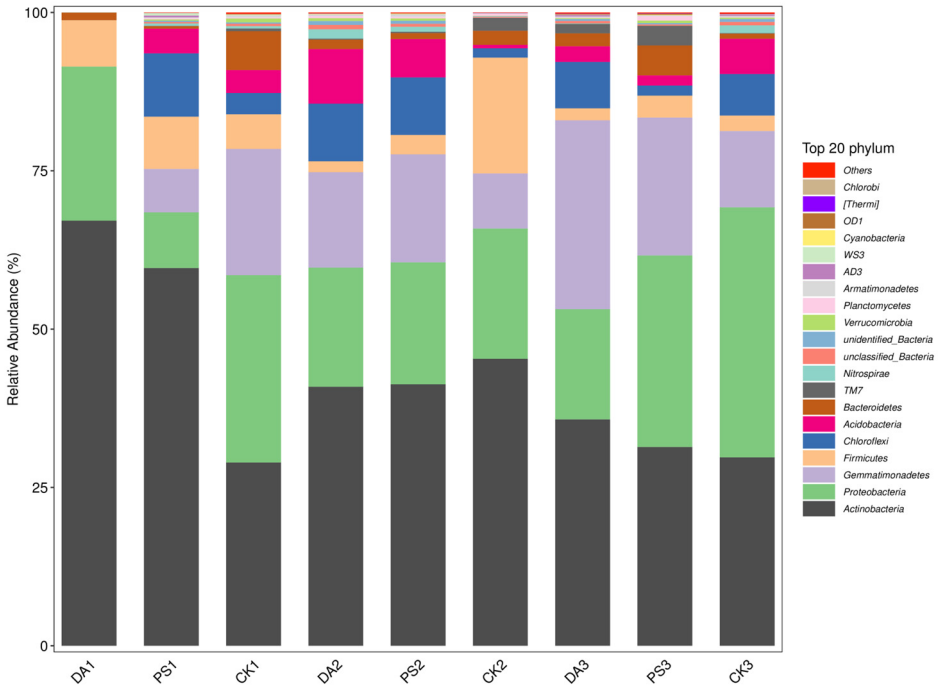


Figure 6. Differences in bacterial community compositions at the phylum level after fumigation with dazomet and chloropicrin.

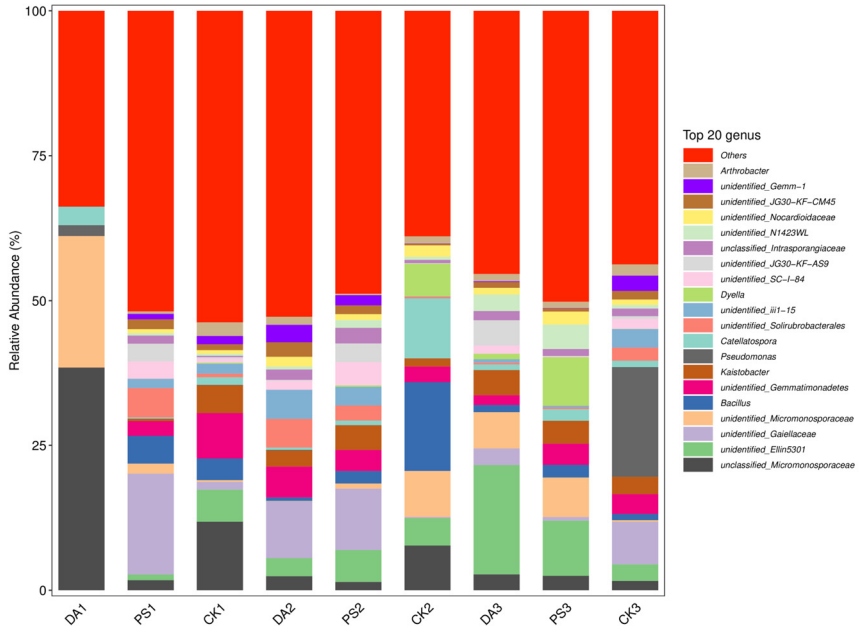


Figure 7. Differences in bacterial community compositions at the genus level after fumigation with dazomet and chloropicrin.

3.4. Effects of Different Treatments on Ginger Metabolites

3.4.1. PCA Analysis

The PCA score plots mainly reflect the similarities or differences in the metabolites of small yellow ginger between the different treatments. As shown in Figure 8, in both positive and negative ion modes, QC samples are closely clustered together, indicating that the test had good repeatability and reliability. The first principal component (PC1)

explained 51.67% of the features of the original data set, showing a significant separation between the control and fumigation treatments. The second principal component (PC2) explained 18.33% of the features of the original data set and indicated that the chloropicrin treatment was clearly separated from the other treatments.

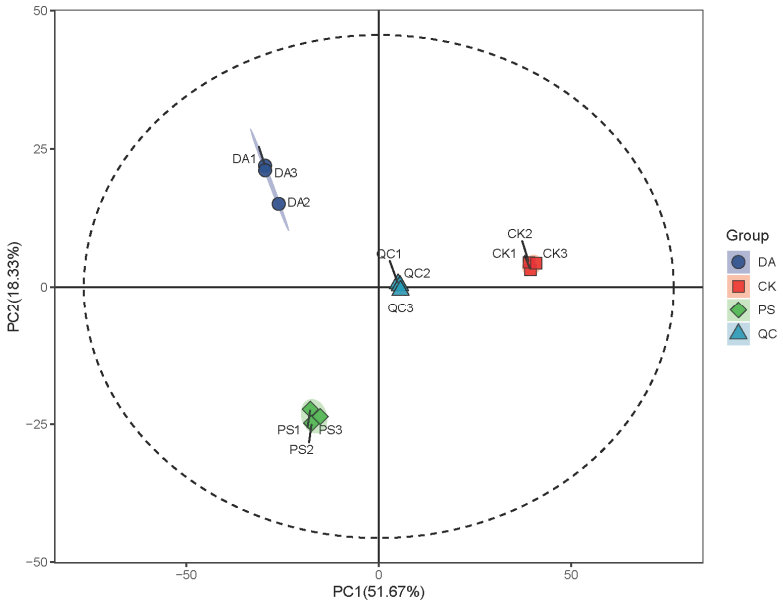


Figure 8. PCA score of the population sample in positive and negative ion mode after fumigation with dazomet and chloropicrin. Note: The QC sample represents a sample of all samples mixed in equal amounts, used to balance the chromatography–mass spectrometry system and determination of the instrument status.

The results of the PCA analyses showed that both fumigants affected the metabolics of small yellow ginger. The greatest differences in metabolites were observed between the control treatment and the fumigation treatments, with the chloropicrin treatment showing the most distinct separation from the other treatments.

3.4.2. Differential Metabolite Screening

The differential metabolites were screened using a combination of multivariate statistics and univariate statistical analysis. The threshold was set as VIP > 1.0, and $p < 0.05$ was used as the screening criteria to select metabolites with significant differences. As shown in the volcano diagrams (Figure 9), a total of 943 different metabolites were identified in the dazomet–control group, with 519 upregulated and 424 downregulated. In the chloropicrin–control group, 916 different metabolites were identified, of which 551 were upregulated and 365 were downregulated. This is shown in the differential metabolite classification pie chart (Figure 9). Lipids and lipid-like molecules, organic acids and derivatives, and organoheterocyclic compounds were the main differential metabolites in both groups.

3.4.3. Differential Analysis of Functional Related Metabolites of Ginger

Ginger contains a variety of bioactive metabolites. In order to further explore the effects of fumigation on the metabolites of ginger, several kinds of bioactive metabolites were selected for differential analysis. As shown in Table 7, in the dazomet–control group, the number of upregulated differential metabolites was lower than the number of downregulated metabolites for amino acids and derivatives, flavonoids, and diarylheptanoids. In the chloropicrin–control group, diarylheptanoid and terpenoid upregulation was less common than the downregulations, and flavonoid upregulation and downregulation were equal. For terpenoids and phenolic compounds in the dazomet–control group, the number of upregulated differential metabolites was higher than the number of downregulated ones,

with five upregulated and four downregulated for terpenoids, and eleven upregulated and seven downregulated for phenolic compounds. Among phenolic compounds, the levels of gingerol, 6-gingerdione, and zingerone were all upregulated.

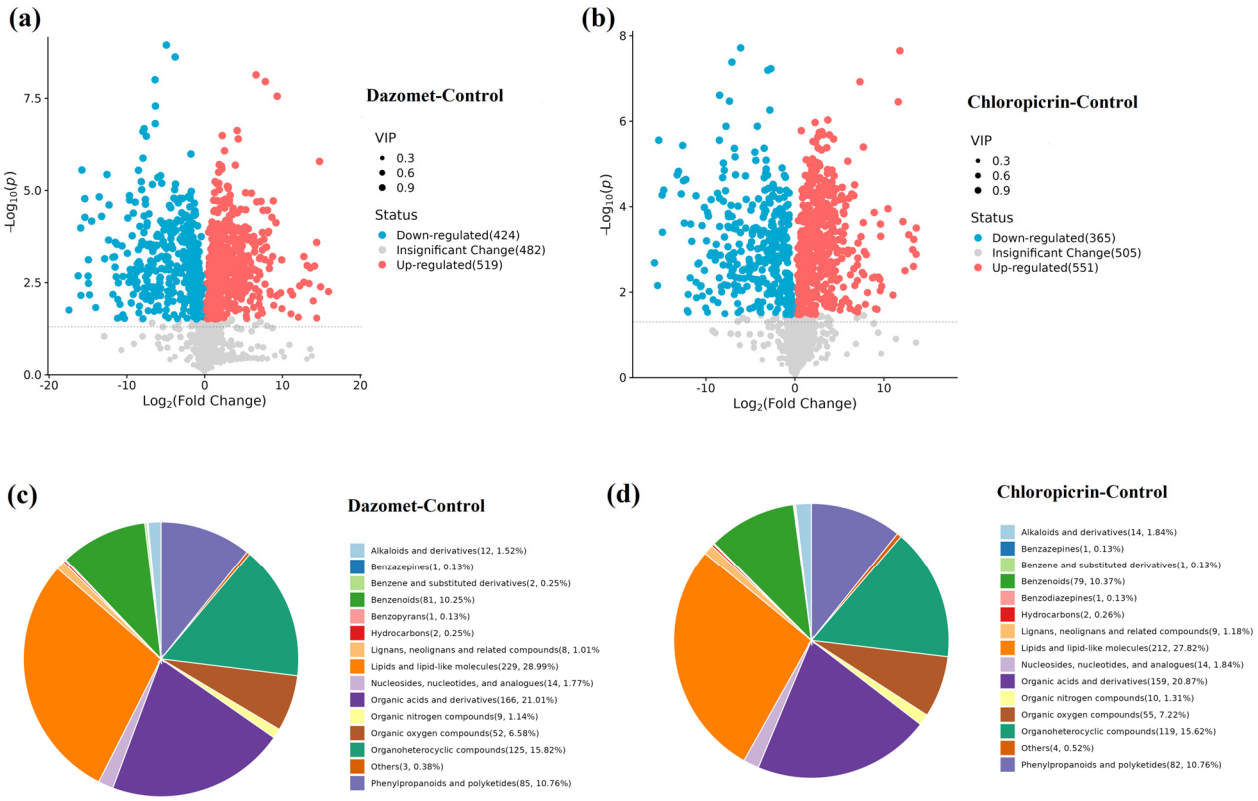


Figure 9. Screening results for different metabolites after fumigation with dazomet and chloropicrin: (**a**) volcanic maps of dazomet and control groups; (**b**) volcanic maps of chloropicrin and control groups—red dots indicate upregulated metabolites and blue dots indicate downregulated metabolites; (**c**) pie chart of differential metabolite classification between dazomet and control groups; (**d**) pie chart of differential metabolite classification between chloropicrin and control groups.

Table 7. Changes in different functional metabolites in ginger after fumigation with dazomet and chloropicrin.

Metabolites	Dazomet VS CK			Chloropicrin VS CK		
	Total Variance	Up	Down	Total Variance	Up	Down
Amino acids and derivatives	40	16	24	36	19	17
Terpenoids	9	5	4	11	5	6
Flavonoids	24	8	16	24	12	12
Phenols	18	11	7	16	11	5
Phenols (gingerol)	2	2	0	2	2	0
Phenols (6-gingerdione)	1	1	0	1	1	0
Phenols (6-shogaol)	0	0	0	1	1	0
Phenols (zingerone)	1	1	0	1	1	0
Diarylheptanoids	10	2	8	11	3	8

In the chloropicrin–control group, the number of upregulated differential metabolites was higher than the number of downregulated ones for amino acids and their derivatives and phenolic compounds, with nineteen upregulated and seventeen downregulated for amino acids and their derivatives, and eleven upregulated and seven downregulated for phenolic compounds. Among these, the levels of gingerol, 6-gingerdione, zingerone, and 6-shogaol were all increased.

The results of the differential analysis of ginger bioactive metabolites showed that both fumigants affected the levels of these valuable metabolites. The increase in these metabolites after fumigation treatment enhanced the pharmaceutical properties and improved the flavor quality of small yellow ginger.

4. Discussion

4.1. Prevention and Control Effects of Two Fumigants on Ginger Blast

The use of chemical fumigants is one of the most effective methods for the control of ginger blast, and studies have shown that dazomet treatment can achieve results similar to those of methyl bromide in terms of disease index, mortality rate, and yield, significantly inhibiting the occurrence of ginger blast [29]. Tian et al. [30] showed that dazomet fumigation could effectively alleviate the problems associated with the continuous cropping of ginger and could effectively control ginger blast. The research of Zhou et al. [31] showed that both chloropicrin and dazomet treatments were effective in suppressing ginger blast and stem basal rot and significantly increased the yield of ginger. In this study, the control effectiveness of dazomet and chloropicrin treatments on ginger blast was, respectively, 84.33% and 94.67% in the first year and 86.33% and 93.67% in the second year, when the control went into a complete loss. In both years, chloropicrin had a better control effect on ginger blast than dazomet. The fresh weight of a single ginger rhizome after dazomet treatment was significantly higher than that after chloropicrin treatment. This observation can be attributed to the fact that dazomet could promote crop nitrogen absorption and crop growth, as reported in the study of Yang et al. [32]. The yield-enhancing effects of dazomet and chloropicrin are in agreement with the observations of Wang et al. [33], who reported that dazomet fumigation could increase the yield of *Codonopsis* seedlings. Mao et al. [34] and Yan et al. [35] also reported increases in ginger yield and strawberry yield after chloropicrin fumigation.

4.2. Effects of Two Fumigants on Soil Bacterial Communities

The occurrence of soil-borne diseases is closely related to the rhizospheric microorganisms in soil, and the incidence of soil-borne diseases is usually low in balanced microecosystems [36]. After the fumigation treatments, the Venn diagram results showed a pattern where unique bacterial OTUs decreased initially, then rose, and finally reduced again during the sampling period. The shared bacterial OTUs increased over time; the alpha diversity results indicated that the diversity and richness of bacterial communities showed a similar pattern during the sampling period. Both the Venn diagram and alpha diversity results indicated that there were significant differences in the structural compositions of the bacterial communities across different treatments. While the field trial was complex and susceptible to weather conditions, the fumigation treatments comprised the primary factor responsible for these changes in microbial compositions. Additionally, the growth process of the ginger likely played a role, as root exudates released by the plant during growth and development play a crucial role in plant–microbe interactions [37]. Studies have shown that plant growth status can lead to changes in rhizospheric microorganisms [38]. It is also known that allelopathy in ginger, where the plant produces allelochemicals, can influence the composition of soil microorganisms [39].

The PCoA results showed that the main factor affecting the soil bacterial community composition with significant differences was the fumigation treatment. A stacked bar diagram at the phylum and genus levels showed that the presence of *Proteobacteria* was inhibited by two fumigants during fumigation and sampling. The relative abundance of *Pseudomonas* increased at 60 days. At 120 days, the relative abundances of *Actinobacteria*, *Gemmatimonadetes*, *Bacillus*, and *Firmicutes* increased. *Bacillus*, *Pseudomonas*, and *Actinobacteria* are beneficial bacteria that can effectively inhibit the occurrence of soil-borne diseases [40–43]. *Gemmatimonadetes* can produce some antibacterial substances and promote crop growth [44]. Studies have shown that *Firmicutes* can resist harsh soil environments [45–47] and may participate in the disease resistance processes of microbial communities [48,49]. Huang

et al. [50] reported that *Actinobacteria* and *Bacillus* have important roles in soil improvement and disease resistance, promoting soil nutrient cycling and maintaining the soil micro–ecological balance. Li et al. [51] and Chen et al. [52] found that after the fumigation stress of chloropicrin and dazomet was relieved, the number of *Bacillus* individuals in soil increased. Li et al. [53] reported that the relative abundances of *Actinobacteria* and *Bacillus* increased after fumigation by chloropicrin and dazomet. Zhang et al. [54] showed that chloropicrin fumigation increased the relative abundance of *Actinomycetes*, which played a key role in the suppression of ginger blast. These reports are consistent with the results of this study.

4.3. Effects of Two Fumigants on Metabolites of Ginger

The results of non-targeted metabolomics showed that both dazomet and chloropicrin fumigation treatments significantly altered the metabolic profile of small yellow ginger. Both treatments increased the content of phenolic compounds such as gingerol, zingerone, and 6-gingerdione in small yellow ginger. Phenolic compounds in ginger, such as gingerol, shogaol, and zingerone have been reported as having anti-cancer, anti-inflammatory, and antioxidant effects [55–58]. Ohnishi et al. [59] suggested that shogaol had good neuroprotective, antioxidant, and anti-inflammatory effects. The levels of terpenoids and amino acids and their derivatives also increased after dazomet and chloropicrin fumigation. These compounds contribute to ginger's unique aromatic flavor and provide amino acids needed by the human body [60–62]. Yang et al. [63] found that soluble solids in the strawberry fruit increased after fumigation using dazomet. Wang et al. [64] reported that using chloropicrin to treat continuous cropping soil for strawberries, combined with adding edible fungal cultivation wastes, improved strawberry fruit quality. In this study, both dazomet and chloropicrin improved the flavor quality of ginger, consistent with previous findings that these fumigants can enhance crop quality.

5. Conclusions

The results of this study showed that dazomet and chloropicrin could effectively control ginger blast, overcome the challenges of continuous cropping, and significantly improve the yield and quality of ginger. Treatment with both fumigants increased the relative abundance of some beneficial bacteria associated with plant resistance to soil-borne diseases. Additionally, both fumigants upregulated phenolic compound and terpenoid production, and chloropicrin treatment also increased the levels of amino acids and their derivatives. The upregulation of these substances can effectively improve flavor quality and enhance antioxidant, anti-inflammatory, and anti-tumoral properties in ginger.

For a long time, ginger blast has affected the healthy growth of small yellow ginger in Luoping County, Yunnan, China, causing the local ginger industry to shrink continuously. After two years of large-scale fumigation experiments, disinfecting soil via fumigation opened up a promising future, and the yield and quality of small yellow ginger improved. In the future, by combining soil fumigation with the addition of beneficial bacteria and bio-organic fertilizers, and integrating chemical fumigation into biological control technologies, the amount of chemical fumigants can be further reduced to achieve sustainable development in the Luoping small yellow ginger industry.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agriculture14091439/s1.

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