

Effects of different colors of plastic-film mulching on soil temperature, yield, and metabolites in *Platostoma palustre*

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Abstract

Platostoma palustre is an annual herb and an important medicinal and edible plant in southern China. Plastic-film mulching is an effective agronomic practice in the cultivation system of *P. palustre*, of which the black film mulching is the most common. However, fewer researches have been focused on the use of the other colors of plastic films in *P. palustre* cultivation. In this study, different colors (white, black, red, and green) of plastic-film were adopted, and the effects of different colors of plastic-film mulching on the soil temperature, yield, and metabolites of *P. palustre* were investigated. The results showed that the fresh weight of single plant of the green film treatment was significantly higher than that of the white film treatment (n = top 28). Based on the results of three temperature measurements, the soil temperature was almost the highest in the red film treatment and lowest in the white film treatment. The metabolomic analysis revealed that a total of 103 differential metabolites were identified. Among these, the gluconic acid, deoxyribose, and N-Acetylmannosamine in the red film treatment presented the highest abundance compared with the other treatments, meanwhile, the abundances of the five monosaccharides in the red film treatment were significantly higher than those of the green film treatment. Moreover, the sucrose, trehalose, and D-(+)-trehalose in the green film treatment exhibited the highest abundance, and the abundances of eight different amino acids in red film treatment were almost the lowest while those in black film treatment were almost the highest. Further analysis of the membership function values indicated that the black and red film treatments might be more suitable for the cultivation and quality production of *P. palustre* in comparison with the other two treatments. This study will provide a theoretical basis for improving the efficient cultivation technology of *P. palustre* and forming a theoretical system of *P. palustre* film mulching cultivation.

Introduction

Plastic-film mulching is a common cultivation and management technology in agricultural production. Generally, this technique plays a role in regulating soil temperature, preserving soil moisture, and shortening crop growth period, and some special films have the functions of cooling, preventing weeds, avoiding aphids, and reflecting light ¹⁻⁴. Plastic-film mulching has a certain role in improving the yield and quality of crops and has been widely used in a variety of crop cultivation, such as corn ⁵, peanut ⁶, citrus ⁷, cucumber ⁸, potato ¹, etc.

Platostoma palustre is an annual herb belonging to the *Platostoma* genus of Labiatae, which is an important medicinal and edible plant in southern China ⁹⁻¹³. The active ingredients of *P. palustre* mainly include polysaccharide, quercetin, oleanolic acid, ursolic acid, caffeic acid, and so on ¹⁴, which possess multiple biological activities, like anti-hypertensive effect ¹⁵, hypolipidemic effect ¹⁶, hepatoprotective effect ¹⁷, antioxidation ¹⁸, and antibiosis ¹⁹. Nowadays, it is widely used as raw material in the food industry, with a wide range of uses and high market and economic value.

P. palustre is usually transplanted in the early spring season when the temperature is low in south China, therefore plastic-film mulching is an effective agronomic measure in the cultivation system of *P. palustre*.

Plastic-film mulching can be used to create good microclimate conditions under the film including heat, air, water and fertilizer conditions suitable for the growth and development of *P. palustre*²⁰. In the planting process of *P. palustre*, plastic-film mulching has been widely used in different production areas in China, especially the black plastic mulch is generally used for the cultivation of *P. palustre*. Studies have shown that black plastic-film mulching during the whole growth period can reduce labor intensity, lower production costs, and increase yields²¹.

Currently, different colors of plastic film are available on the market and vary in their effects of increasing temperature and moisture retention, thus affecting the growth, development, and yield of crops^{22,23}. As mentioned above, the research on the plastic-film mulching in *P. palustre* is mostly limited to the black plastic film, while fewer researches have been focused on the use of the other colors of plastic films in *P. palustre* cultivation. In this study, to screen out the most suitable types of mulching film for *P. palustre* cultivation, we intend to adopt different colors (white, black, red, and green) of plastic film to plant *P. palustre*. The effects of different color plastic-film mulching on the yield of *P. palustre* were studied, and the differences in metabolites between different treatments were also compared and analyzed using metabolomics. This study will provide a theoretical basis for improving the efficient cultivation technology of *P. palustre* and forming a theoretical system of *P. palustre* film mulching cultivation.

Materials and methods

Plant materials

In this study, we set up four colors of plastic-film mulching treatments (including red, green, black, and white) for growing *P. palustre* at the base of Lingshan, Guangxi, China. On April 21, 2022, we transplanted the cuttage seedlings of *P. palustre* into the field (the ridge length is 9 meters, the ridge width is 1.8 meters, and the planting density is 4200 plants/667 m²), with normal field management. The *P. palustre* test materials were obtained locally from Lingshan county, China, and complied with relevant institutional, national, and international guidelines and legislation.

Determination of plant fresh weight

On July 22, 2022, the measurement of the yield was performed in the field. For each treatment, we selected a certain number of plants to measure the fresh weight of individual plants. The weighed plants were then left to dry for 1 day, then stacked for 1–2 days, and spread out to dry until the moisture content was about 10–15%. Subsequently, a certain number of dried samples of *P. palustre* were selected, cut, and ground into powder, then screened with a 200 mesh sieve, and finally, the powder samples obtained were used for metabolome analysis.

Determination of soil temperature

We also measured the soil temperature in different treatments on May 18, May 25, and June 1, respectively. The measurement time was at 10:00 am and the soil temperature was measured at about 20

cm of the soil tillage layer.

Metabolite extraction

50 mg samples were accurately weighed, and the metabolites were extracted using a 400 μ L methanol: water (4:1, v/v) solution with 0.02 mg/mL L-2-chlorophenyl alanine as internal standard. The mixture was allowed to settle at -10 °C and treated by High throughput tissue crusher Wonbio-96c (Shanghai wanbo biotechnology co., LTD) at 50 Hz for 6 min, then followed by ultrasound at 40 kHz for 30 min at 5 °C. The samples were placed at -20 °C for 30 min to precipitate proteins. After centrifugation at 13000 $\times$ g at 4 °C for 15 min, the supernatant was carefully transferred to sample vials for LC-MS/MS analysis.

UHPLC-MS/MS analysis

The instrument platform for LC-MS analysis is the UHPLC-Q Exactive system of Thermo Fisher Scientific. The mass spectrometric data were collected using a Thermo UHPLC-Q Exactive Mass Spectrometer equipped with an electrospray ionization (ESI) source operating in either positive or negative ion mode. The chromatographic conditions, MS conditions, and data preprocessing and annotation referred to the literature²⁴.

Differential metabolites analysis

All analyses of metabolomics data were performed on the Majorbio Cloud platform (<https://cloud.majorbio.com>). Briefly, the R package ropls (Version 1.6.2) was used to perform principal component analysis (PCA) and orthogonal partial least squares discrimination analysis (OPLS-DA), and the selection of significantly different metabolites (DMs) was determined based on the Variable importance in the projection (VIP) obtained by the OPLS-DA model and the p -value of student's t test, and the metabolites with $VIP > 1$, $p < 0.05$ were significantly different metabolites²⁵.

Data analysis

SPSS v.17 software was employed for one-way variance analysis (ANOVA), correlation analysis, and membership function analysis. The data means were analyzed using the Duncan test for statistical significance ($p \leq 0.05$).

Results

Effect of different colors of plastic-film mulching on the fresh weight of *P. palustre*

In this study, four colors (black, white, red, and green) of plastic-film mulching treatments were adopted (Fig. 1A), and we randomly selected 34, 35, 36, and 36 individual plants from the black film, white film, red film, and green film treatments, respectively. Then the fresh weight of each individual plant was weighed. The results showed that there were no significant differences in the fresh weight of single plant between four treatments ($n \geq 34$) (Fig. 1B). However, when $n = 28$, the fresh weight of single plant of the green film treatment was significantly higher than that of the white film treatment (Fig. 1C). Compared with the

white film treatment, the fresh weight of the green film treatment increased by 14.15%. In addition, the white film treatment showed the lowest fresh weight of single plant, although it was not significantly different from the other two treatments (black and red films).

Effect of different colors of plastic-film mulching on the soil temperature

The soil temperature of different treatments was also determined. As shown in Fig. 2, on May 18, 2022, the soil temperatures in the black and red film treatments were significantly higher than those in the other two treatments. At the second measurement, the highest soil temperature was observed in the red film treatment. The third measurement showed that the highest soil temperatures were found in the red and green film treatments. Based on the results of three temperature measurements, the soil temperature was almost the highest in the red film treatment and lowest in the white film treatment.

Metabolites of different treatments based on LC-MS

In this study, we extracted and analyzed the metabolites from the whole plant of *P. palustre* with six replicates of each treatment. The base peak chromatogram (BPC) indicated that the obtained data could be used for subsequent analysis (Fig. 3). In total, 1588 and 1604 metabolites were identified under the positive and negative ion scanning modes, respectively (Supplementary Fig. 1). Further analysis of correlation, PCA, Venn, and PLS-DA showed that the data were reliable (Supplementary Fig. 2). KEGG compound classification revealed that these metabolites included several categories: antibiotics, carbohydrates, hormones and transmitters, lipids, nucleic acids, organic acids, peptides, vitamins and cofactors, and so on containing 177 metabolites (Supplementary Fig. 3A; Supplementary Table 1). KEGG pathway exhibited that these metabolites were involved in biosynthesis of other secondary metabolites, amino acid metabolism, lipid metabolism, carbohydrate metabolism and so on (Supplementary Fig. 3B).

Differential metabolites analysis

The different groups could be better distinguished by OPLS-DA analysis (Fig. 4). The model was considered valid when $Q^2 > 50\%$ and $R^2Y-Q^2 < 0.3$ ²⁶, and this criterion was satisfied in both positive and negative ion modes, so the OPLS-DA model fitted well in this study. To ensure the reliability of the results, the OPLS-DA model was also analyzed by permutation testing. As shown in Fig. 5, the Q^2 intercept of all comparison groups was < 0.05 in both positive and negative ion modes. This indicated that the OPLS-DA model was feasible and the metabolites differed significantly between treatments.

Further, according to the $VIP > 1$ and $p < 0.05$, all the identified metabolites were used for screening the differential metabolites, and the results of the different comparison groups were analyzed. The results showed that a total of 1775 differential metabolites were identified in this study. Of these, there were 867, 821, 775, 743, 810, and 872 differential metabolites were identified in Green_vs_Black, Red_vs_Black, Red_vs_White, Red_vs_Green, Green_vs_White, and White_vs_Black, respectively (Supplementary Fig. 4). Based on these differential metabolites, KEGG compounds classification showed that a total of 103 differential metabolites were identified in this study (Fig. 6). These differential metabolites were classified

into different categories, including phospholipids, monosaccharides, oligosaccharides, carboxylic acids, amino acids and so on (Supplementary Table S2). Among these, there were 42, 47, 42, 46, 57, and 39 differential metabolites in Green_vs_Black, Red_vs_Black, Red_vs_White, Red_vs_Green, Green_vs_White, and White_vs_Black, respectively (Supplementary Fig. 5). Further KEGG enrichment analysis revealed that the differential metabolites of different comparison groups were significantly enriched in different metabolic pathways (Supplementary Table 3). For example, in Green_vs_White, a total of 61 differential metabolites were involved in 7 metabolic pathways, which were the highest number of differential metabolites and metabolic pathways significantly enriched. This indicated the greatest difference between the two treatments. Conversely, the differential metabolites in Red_vs_Green were not significantly enriched in any metabolic pathway.

Monosaccharides

Polysaccharides are one of the most important quality indicators of *P. palustre*. Zhang, et al.²⁷ found that *P. palustre* polysaccharides consisted of eight monosaccharides including galacturonic acid, glucose, galactose, xylose, mannose, rhamnose, ribose, and glucuronic acid. In this study, five monosaccharides (gluconic acid, rhamnose, 2-deoxy-D-ribose, deoxyribose, and N-Acetylmannosamine) related to the quality of *P. palustre* were detected. As shown in Fig. 7, the gluconic acid, deoxyribose, and N-Acetylmannosamine in the red film treatment presented the highest abundance compared with the other treatments, meanwhile, the abundances of the five monosaccharides in the red film treatment were significantly higher than those of the green film treatment. It was inferred that the red film treatment could significantly increase the monosaccharide content and might be beneficial to the improvement of the quality of *P. palustre*.

Oligosaccharides

Oligosaccharide refers to any carbohydrate of from 3-6 units of simple sugars (monosaccharides). Here, we identified three oligosaccharides including sucrose, trehalose, and D-(+)-trehalose. The abundances of these three oligosaccharides in the black film treatment were significantly lower than those of the other treatments. On the contrary, these three oligosaccharides in the green film treatment exhibited the highest abundance (Fig. 8). Therefore, it was indicated that the green film treatment could promote the content of oligosaccharides of *P. palustre*. Meanwhile, the black film treatment was not conducive to the increase of oligosaccharide content in *P. palustre* in comparison with the other treatments.

Amino acids

In this study, we detected eight differential amino acids containing L-alanine, L-glutamine, L-phenylalanine, L-asparagine, L-(+)-arginine, L-tyrosine, Aspartic acid, and L-glutamic acid. As could be seen from Fig. 9, the abundances of eight different amino acids in red film treatment were almost the lowest, while those in black film treatment were almost the highest. Alanine, Serine, and Glycine are sweet amino acids in *P. palustre*, and Aspartic acid and Glutamic acid are delicious amino acids²⁸. This

suggested that the black film treatment was beneficial to the improvement of the nutrition and flavor levels of *P. palustre*.

Correlation analysis of various indicators

In this study, the correlation analysis on the 18 indicators (except soil temperature) analyzed above were further performed. As shown in Table 1, Rhamnose was positively correlated with 2-Deoxy-D-ribose and negatively correlated with Trehalose and Sucrose ($p<0.05$). Gluconic Acid was positively correlated with N-Acetylmannosamine and Aspartic Acid but negatively correlated with L-Glutamine ($p<0.05$). 2-Deoxy-D-ribose was negatively correlated with Trehalose, D-(+)-Trehalose, and Sucrose while positively correlated with L-Glutamic Acid ($p<0.05$). Deoxyribose and L-Phenylalanine were positively correlated with N-Acetylmannosamine and Aspartic Acid and L-Tyrosine, respectively, while Sucrose and L-Alanine were negatively correlated with L-Glutamic Acid and Aspartic Acid. L-Glutamic Acid and L-Alanine had a significantly negative correlation with Trehalose and D-(+)-Trehalose and N-Acetylmannosamine, respectively, while there was a significant positive correlation between Sucrose, Trehalose, and D-(+)-Trehalose. In addition, there was a significantly positive correlation between N-Acetylmannosamine and Aspartic Acid, with a correlation coefficient of 100%.

Analysis of the membership function values of various indicators

We calculated the D values of the indicators for each treatment using membership functions. The higher the D value, the better the treatment. It could be seen from Table 2, the ranking of different treatments was black>red>white>green, indicating that the black and red film treatments might be more suitable for the cultivation and quality production of *P. palustre* in comparison with the other two

Table 2
Comprehensive evaluation of different treatments.

Treatment	$\mu(x)$		D value	Ranking
	μ_1	μ_2		
Black	0.7580	1.0000	0.8311	1
Green	0.0000	0.1529	0.0462	4
Red	1.0000	0.0000	0.6978	2
White	0.2470	0.7085	0.3864	3

Discussion

In South China, the temperature in early spring is low and drought in spring often occurs. Plastic-film mulching plays important roles in increasing soil temperature and humidity, reducing weeds and improving crop yield ²⁹, which has been used in many crops such as corn, peanut, citrus, cucumber, potato, as well as *P. palustre* ^{1,5–8,30}. Different specifications and colors of mulch have different patterns of projection, absorption and reflection of the spectrum, resulting in different effects on plant absorption

of photosensitive pigments, soil environment, light environment and plant growth³¹. For example, compared to the control (no mulch), the yield of fresh ear of corn in transparent mulch, black mulch, and silver mulch increased by 9.25%, 14.47%, and 15.37%, respectively²³. Similarly, the yield of maize in black mulch and white mulch treatments was significantly higher than that of the control, with a yield increase of 18.77% and 15.28%, respectively, and the yield of black mulch was greater³². In this study, the fresh weight of single plant of the green mulch was significantly higher than that of the white mulch and increased by 14.15% in comparison with the white mulch (Fig. 1B). It was indicated that the white film mulch treatment might not be suitable for increasing the yield of *P. palustre* compared with the other treatments. The mulch could change the light environment of the plant by reflecting radiant light³¹. The green mulch reflected green light, which might fulfill the specific light required for *P. palustre* photosynthesis and promote *P. palustre* photosynthesis, resulting in higher yields.

Ngouajio and Ernest³³ showed that the color of the mulch can determine the amount of light energy and heat obtained, thus directly affecting the soil temperature. Different colors of mulch have different absorption and reflection bands of radiant light, which is reflected in the variability of light transmittance, red film > green film > silver-white film > black film³⁴. The greater the light transmittance of the mulch, the more significant the effect of soil warming and the lower the corresponding soil water content³⁴. The results of this study were similar to those of Zhong, et al.³⁴ on colored mulch for strawberry, where the soil temperature of the red film treatment was higher than that of the silver-white film.

Studies have showed that the main components in *P. palustre* mainly includes polysaccharides, flavonoids, phenols, terpenoids, amino acids, etc³⁵. Polysaccharide is one of the important quality indicators of *P. palustre*, and it is also the most abundant (about 20%) and widely used active ingredient in *P. palustre*, with antioxidant, immunomodulatory, antitumor and intestinal flora regulation effects^{36–40}. In our study, the abundance of five monosaccharides (gluconic acid, rhamnose, 2-deoxy-D-ribose, deoxyribose, and N-Acetylmannosamine) associated with the quality of *P. palustre* was higher under the red film mulch treatment than under the green film mulch treatment, while the abundance of three oligosaccharides (sucrose, trehalose, and D-(+)-trehalose) was highest under the green film mulch treatment (Fig. 7–8). There were also significant correlations between some monosaccharides and oligosaccharides. For example, rhamnose was negatively correlated with trehalose and sucrose ($p < 0.05$), and 2-deoxy-D-ribose was negatively correlated with sucrose, trehalose, and D-(+)-trehalose ($p < 0.05$) (Table 1). It was indicated that the red film mulch treatment could significantly influence the accumulation of monosaccharides and oligosaccharides and enhance the quality of *P. palustre*. This might be due to the facts that different colors of mulch films affect the light quality, except for black and white-black films, red and green films have higher light transmission at wavelengths of 350–622 nm, and the main light quality of red film treatment is red light, while the main light quality of green film treatment is blue, green, and blue-green light⁴¹. Similar to our previous study, red light promoted the growth and development of *P. palustre* compared to blue light and increased the content of soluble sugars and pectin in *P. palustre*¹³. This further confirmed that the red film treatment (red light) was beneficial to the quality improvement of *P. palustre*.

Nutritional flavor is also an important quality indicator of *P. palustre*. Amino acids are nutrient substances in *P. palustre*, which have an important influence on the taste of *P. palustre*⁴². Liu and Chen⁴³ determined amino acids in artificially cultivated and wild *P. palustre* from Jiangxi, and showed that *P. palustre* contained 18 amino acids, among which glutamic acid and aspartic acid had the highest content. Su, et al.²⁸ analyzed the differences in amino acid composition and content of *P. palustre* from different sources, and the results showed that all *P. palustre* contained 17 amino acids, with glutamic acid, aspartic acid, and leucine in the top three. In this study, eight differential amino acids were detected, including L-alanine, L-phenylalanine, L-(+)-arginine, L-tyrosine, aspartic acid, L-glutamic acid, L-glutamine and L-asparagine (Fig. 9). Human amino acid taste receptors have a particularly high affinity for glutamate, with other L-amino acids being second to none and D-amino acids being unable to bind⁴⁴. Among these differential amino acids, glutamine is an amino acid generated by amidation of the γ -carboxyl group of glutamate, which is interconvertible with glutamate and asparagine, and the other six amino acids have been detected in previous amino acid determinations of *P. palustre*. Glutamic acid and aspartic acid are the fresh tasting amino acids of *P. palustre* and have important pharmacological effects. Glutamic acid has antioxidant, anti-inflammatory and anti-cancer effects^{45–47}, while aspartic acid has anti-tumor and improves diabetic nephropathy^{47,48}. Alanine is the sweet amino acid of *P. palustre* that improves diabetic blood glucose levels⁴⁹. Tyrosine has antioxidant and analgesic effects^{50,51}. Phenylalanine and arginine, as essential amino acids, are related to the maintenance of nitrogen balance in the body and affect health⁵². Here, eight differential amino acids were almost the highest under the black film mulch treatment, indicating that the black film mulch treatment might be suitable to enhance the nutritional flavor of *P. palustre* compared to other treatments. Differences in soil conditions affect the type and content of amino acids in plants due to the reduced evaporation of soil water under black film mulch, which prevent soil surface caking, maintain soil looseness and increase the oxygen content in the soil, facilitating the development of cactus roots and the activity of soil microorganisms^{30,53}. Xiong and Zhu³⁰ showed that soil microorganisms were 10-fold higher under black film mulch treatment than without film in *P. palustre* cultivation. Therefore, we speculated that the red film treatment might have created a better soil environment, thereby increasing the amino acid content, nutritional flavor level, and potential pharmacological effects of *P. palustre*.

In general, based on the above results, the white film mulch treatment might not be suitable for the cultivation of *P. palustre*. The red film mulch treatment increased the content of monosaccharides, and green film mulch treatment increased the content of oligosaccharides, indicating that red film mulch treatment was beneficial to improve the quality of *P. palustre*. The eight differential amino acids were highest almost when under black film mulch treatment, indicating that the black film mulch treatment was suitable for improving the nutritional flavor of *P. palustre*. Therefore, in the actual cultivation of *P. palustre*, the use of black or red film treatment may be chosen according to the actual needs.

Conclusions

The white film mulch treatment was not suitable for the cultivation of *P. palustre*. The red film mulch treatment could significantly promote the content of monosaccharides, while the green film mulch treatment could increase the abundance of oligosaccharides. The black film mulch treatment could improve the abundance of amino acids. This study provided a reference for the application of different colors film in the cultivation and production of *P. palustre*, and also provided important data for improving the economic and medicinal value of this species.

Declarations

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Author Contributions: writing—original draft preparation, S.H., H.C., M.X., C.Q. and Z.C.; writing—review and editing, supervision, funding acquisition, D.T. and F.W. All authors have read and agreed to this version of the manuscript.

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Conflicts of Interest: The authors declare no conflict of interest.

Supplementary Materials: FIGURE S1 The RSD plot and total ion count and identification statistics. A, The RSD plot under positive ion mode. B, The RSD plot under negative ion mode. C, The total ion count and identification statistics. FIGURE S2 The correlation analysis, Venn plot, PCA, and PLS-DA plot. A-D, The correlation analysis, Venn plot, PCA, and PLS-DA plot, respectively. FIGURE S3 The KEGG compound classification and KEGG pathway. FIGURE S4 The number of compounds of different comparison groups. FIGURE S5 The number of compounds of different comparison groups based on KEGG compound classification. Table S1 KEGG compounds classification of the metabolites annotated in this study. Table S2 KEGG compounds classification of the DMs in this study. Table S3 The metabolic pathways significantly enriched in the DMs of different comparison groups.

References

1. Zhou, L. N. *et al.* The Influence of Different Colors of Plastic Films on Growth and Development of Potato. *Journal of Hebei Agricultural Sciences* 16, 18–21 (2012).
2. Wang, H. L., Zhang, X. C., Yu, X. F., Ma, Y. F. & Hou, H. Z. Effect of using black plastic film as mulch on soil temperature and moisture and potato yield. *Acta Ecologica Sinica* 36, 5215–5226 (2016).

3. Han, Z. H. Effects of Different Types of Plastic Film Mulching on PVY. *Journal of Anhui Agricultural Sciences* 46, 140–141 + 144 (2018).
4. Wang, H. *et al.* Effects of vapor-permeable reflective film mulching on citrus tree microenvironment, shoot growth and fruit development. *Journal of Fruit Science* 34, 996–1006 (2017).
5. Jiang, H. X., Wei, Z. B. & Shi, Z. H. Effectiveness of different mulches in corn inter-row applications. *Modernizing Agriculture*, 27–29 (2022).
6. Guo, S. Y. *et al.* Effects of Different Plastic Film Mulching on the Yield and Quality of Quanhua 557. *Fujian Agricultural Science and Technology* 53, 48–52 (2022).
7. Wu, S. H., Shi, X. G., Chen, J. W. & Wang, P. Effects of plastic film covering on improving illumination and fruit quality of *citrus* variety Bendizao. *Acta Agriculturae Zhejiangensis* 24, 826–829 (2012).
8. Yan, S. J., Zhang, J. N. & Liu, J. Effects of different mulching methods on growth and quality of cucumber. *China Cucurbits and Vegetables* 34, 74–79 (2021).
9. Tang, D. F. *et al.* Mitochondrial genome characteristics and phylogenetic analysis of the medicinal and edible plant *Mesona chinensis* Benth. *Front Genet* 13, 1056389 (2022).
10. Tang, D. F. *et al.* Characteristics and comparative analysis of *Mesona chinensis* Benth chloroplast genome reveals DNA barcode regions for species identification. *Functional & Integrative Genomics*, 1–13 (2022).
11. Tang, D. F. *et al.* Physio-morphological, biochemical and transcriptomic analyses provide insights into drought stress responses in *Mesona chinensis* Benth. *Front. Plant Sci.* 13, 203 (2022).
12. Tang, D. F. *et al.* Codon usage bias and evolution analysis in the mitochondrial genome of *Mesona chinensis* Benth. *Acta Physiologiae Plantarum* 44, 118 (2022).
13. Tang, D. F. *et al.* Identification of Differentially Expressed Genes and Pathways Involved in Growth and Development of *Mesona chinensis* Benth Under Red- and Blue-Light Conditions. *Front. Plant Sci.* 12, 761068 (2021).
14. Li, B. *et al.* Research progress on quality control of Xiancao. *Journal of Guangzhou University of Traditional* 33, 755–758 (2016).
15. Yeh, C. T., Huang, W. H. & Yen, G. C. Antihypertensive effects of Hsian-tsao and its active compound in spontaneously hypertensive rats. *J. Nutr. Biochem.* 20, 866–875 (2009).
16. Li, D. Y., Lu, G., Wang, D. D. & Wang, M. The Influence of Xiancao Hypolipidemic Tea on the TC and TG metabolism of the Experimental Rabbits. *Chinese General Practice* 13, 9–10 (2010).
17. Wang, W., Jiang, L., Ren, Y., Shen, M. & Xie, J. Characterizations and hepatoprotective effect of polysaccharides from *Mesona blumes* against tetrachloride-induced acute liver injury in mice. *Int. J. Biol. Macromol.* 124, 788–795 (2019).
18. Yen, G. & Hung, C. Effects of alkaline and heat treatment on antioxidative activity and total phenolics of extracts from Hsian-tsao (*Mesona procumbens* Hemsl.). *Food Res. Int.* 33, 487–492 (2000).
19. Liu, F. L. & Feng, C. L. In vitro antibacterial test of Hsian-tsao (*Mesona chinensis* Benth) against avian *Escherichia coli*. *Guangdong J. Anim. Vet. Sci.* 33, 17–43 (2008).

20. Wu, Q. H., Liang, G. J., Wei, R. C. & Xie, Y. J. A preliminary report on the comparative benefits of mulching and open-field cultivation of cool pink grass.. *Agricultural Research and Application* 2, 50–51 (2012).
21. Xiao, R. F., Han, G. D. & Wu, M. H. The black film planting technology of *Mesona chinensis* Benth.. *Fujian Agricultural Science and Technology* 3, 36 (2009).
22. Qu, X. C. Effects of Different Color Film Mulchings on Potato Growth and Yield. *Chinese Potato Journal* 29, 346–350 (2015).
23. Huang, S. C. & Chen, J. T. Effects of Different Colors of Plastic Film Mulching on Growth and Yield of Sweetcorn. *Modern Agricultural Science and Technology*, 11–14 + 18 (2022).
24. Hu, S. S. *et al.* Pu-erh tea increases the metabolite Cinnabarinic acid to improve circadian rhythm disorder-induced obesity. *Food Chem.* 394, 133500 (2022).
25. Liu, D. J. *et al.* Differential metabolites identification of Ma bamboo shoots under different low-temperature conditions based on non-target metabolomics. *Journal of Food Safety & Quality* 14, 212–220 (2023).
26. Eriksson, L., Kettanehworld, N., Trygg, J., Wikström, C. & Wold, S. Multi- and megavariate data analysis: part I: basic principles and applications. *Umetrics. Inc.* (2006).
27. Zhang, W. B., Wang, Z. C., Zhang, L. Y. & Qian, J. H. Analysis of Monosaccharide Composition and Content in *Mesona Chinensis* Benth Polysaccharides by Precolumn Derivatization Ultra-high Performance Liquid Chromatography-Tandem Quadrupole Mass Spectrometry. *Journal of Instrumental Analysis* 32, 143–149 (2013).
28. Su, H. L., Huang, Y. Z. & Chen, J. Y. Comparative Analysis of Amino Acids Content in *Mesona chinensis* from Different Producing Areas. *Chinese Wild Plant Resources* 30, 66–69 (2011).
29. Luo, X. *et al.* Carbon footprint of a winter wheat-summer maize cropping system under straw and plastic film mulching in the Loess Plateau of China. *Sci. Total Environ.* 794, 148590 (2021).
30. Xiong, X. L. & Zhu, H. S. High yield cultivation techniques of Xiancao covered with black film. *Fujian Science & Technology of Tropical Crops* 35, 45–47 (2010).
31. Zhang, H., Li, C., Huang, J. X., Yang, S. L. & Hu, H. Y. in *Research and Practice on Water Issues in Beijing (2018)*. 12–18.
32. Yang, S. J. *et al.* Effects of different color plastic film mulching on soil temperature, weed occurrence and corn yield of maize. *Jiangsu Agricultural Sciences* 47, 92–96 (2019).
33. Ngouajio, M. & Ernest, J. Changes in the Physical, Optical, and Thermal Properties of Polyethylene Mulches during Double Cropping. *Hortscience* 40, 94–97 (2005).
34. Zhong, P. L., Yang, S. P., Qiao, R. & Wang, T. W. An Application Study of Color Plastic Film Used for Strawberry Production in Greenhouse. *Guizhou Agricultural Sciences* 40, 77–80 (2012).
35. Ding, J., Huang, S., Zhang, B. M., Lv, W. Z. & Lai, X. P. Study on characteristic fingerprint of *Mesona chinensis*. *Chinese Traditional and Herbal Drugs* 43, 2284–2288 (2012).

36. Niu, R., Han, N. J. & Fang, H. L. Research progress on active ingredients and efficacy of *Mesona chinensis* Beath. Modern Communication, 254–255 (2018).
37. Huang, L. *et al.* *Mesona chinensis* Benth polysaccharides protect against oxidative stress and immunosuppression in cyclophosphamide-treated mice via MAPKs signal transduction pathways. Int. J. Biol. Macromol. 152, 766–774 (2020).
38. He, C. B. *et al.* Protective Effect of Polysaccharide from *Mesona blumes* on Oxidative Damage of Cells. Food Science 41, 160–168 (2020).
39. Chen, X. *et al.* Changes in polysaccharides structure and bioactivity during *Mesona chinensis* Benth storage. Current res. food sci. 5, 392–400 (2022).
40. Lu, H. *et al.* *Mesona chinensis* Benth Polysaccharides Alleviate DSS-Induced Ulcerative Colitis via Inhibiting of TLR4/MAPK/NF- κ B Signaling Pathways and Modulating Intestinal Microbiota. Mol. Nutr. Food Res. 66, e2200047 (2022).
41. Li, Z. M., Ma, Y. J., Liu, P., Cai, H. M. & Zhao, Y. C. Effects of Colored Plastic Mulches on Growth and Development of Morchella. Acta Edulis Fungi 30, 19–29 (2023).
42. Tang, D. F., Quan, C. Q., Huang, S. H. & Wei, F. Integrating LC-MS and HS-GC-MS for the metabolite characterization of the Chinese medicinal plant *Platostoma palustre* under different processing methods. Front. Nutr. 10, 1181942 (2023).
43. Liu, X. G. & Chen, M. M. Research on the exploitation and utilization of *Mesona Blume* in China. Food Research and Development, 109–112 (2004).
44. Efeyan, A., Comb, W. C. & Sabatini, D. M. Nutrient-sensing mechanisms and pathways. Nature 517, 302–310 (2015).
45. Pagire, S. H. *et al.* Design, synthesis and biological evaluation of glutamic acid derivatives as anti-oxidant and anti-inflammatory agents. Bioorg. Med. Chem. Lett. 28, 529–532 (2018).
46. Ali, I., Wani, W. A., Haque, A. & Saleem, K. Glutamic acid and its derivatives: candidates for rational design of anticancer drugs. Future Med. Chem. 5, 961–978 (2013).
47. Yamaguchi, Y. *et al.* Combination of aspartic acid and glutamic acid inhibits tumor cell proliferation. Biomed. res-tokyo 37, 153–159 (2016).
48. Ichikawa, S. *et al.* Aspartic acid supplementation ameliorates symptoms of diabetic kidney disease in mice. FEBS Open Bio. 10, 1122–1134 (2020).
49. Dandare, S. U. *et al.* L-alanine supplementation improves blood glucose level and biochemical indices in alloxan-induced diabetic rats. J. Food Biochem. 45, e13590 (2021).
50. Hung, C. H., Chiu, C. C., Liu, K. S., Chen, Y. W. & Wang, J. J. Subcutaneous L-tyrosine elicits cutaneous analgesia in response to local skin pinprick in rats. Eur. J. Pharmacol. 765, 457–462 (2015).
51. Lu, N. *et al.* Tyrosine can protect against oxidative stress through ferryl hemoglobin reduction. Toxicol. In Vitro 28, 847–855 (2014).
52. Wu, G. Amino acids: metabolism, functions, and nutrition. Amino acids 37, 1–17 (2009).

53. Shi, H. B., Ma, Y. J., Zhang, L., Liang, Y. Q. & Han, P. M. Effects of Nitrogen Reduction and Combined Application of Compound Microbial Fertilizer on Apple Yield and Quality in Dryland. Shaanxi Journal of Agricultural Sciences 69, 87–91 + 104 (2023).

Tables

Table 1 is available in the Supplementary Files section.

Figures

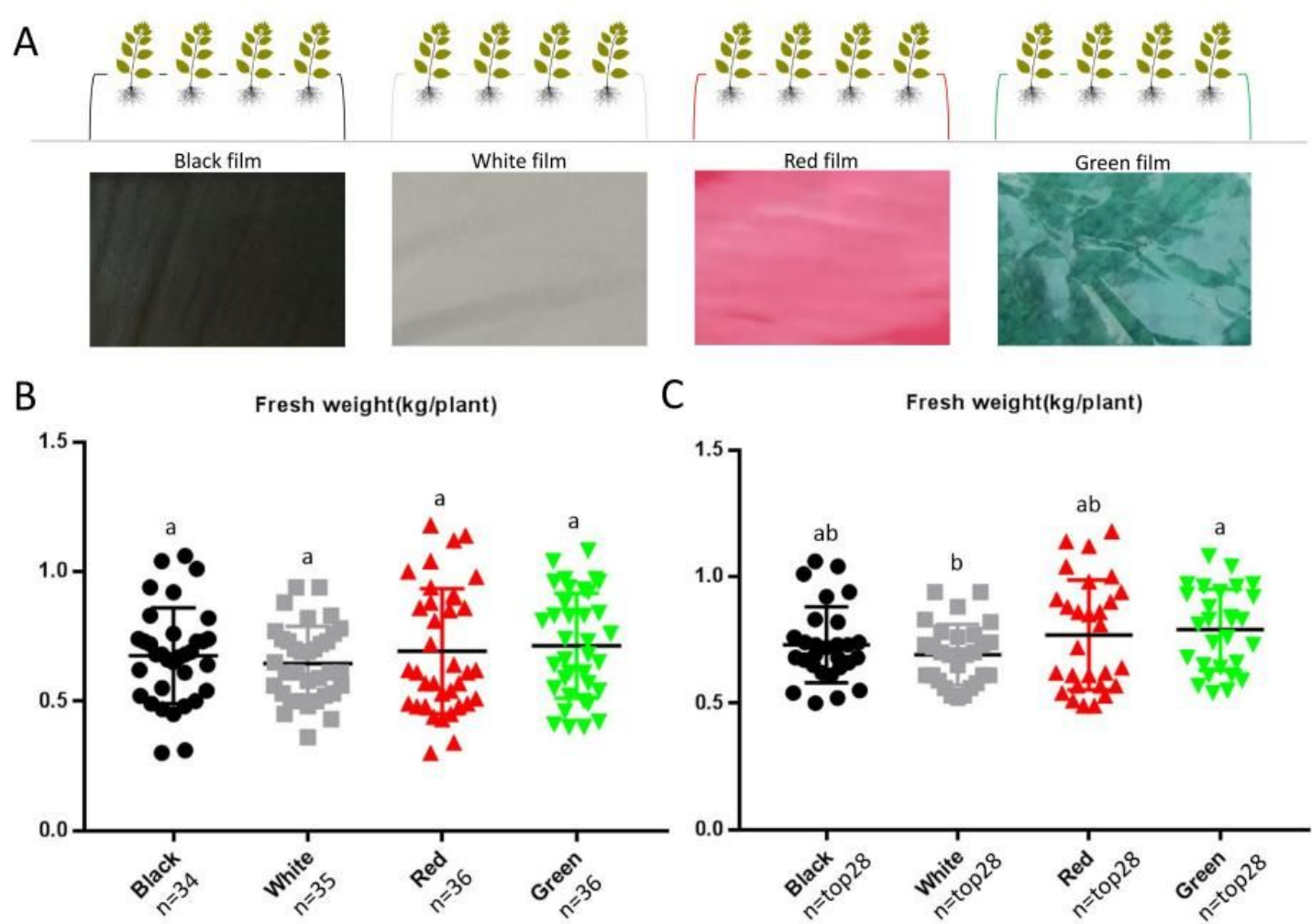


Figure 1

The comparison of fresh weight of individual plant under different treatments. A, The experimental treatment design; B, The fresh weight of individual plant when considering the total number of the samples; C, The fresh weight of individual plant when considering the top28 of the samples. Different letters indicated the significant difference ($p<0.05$).

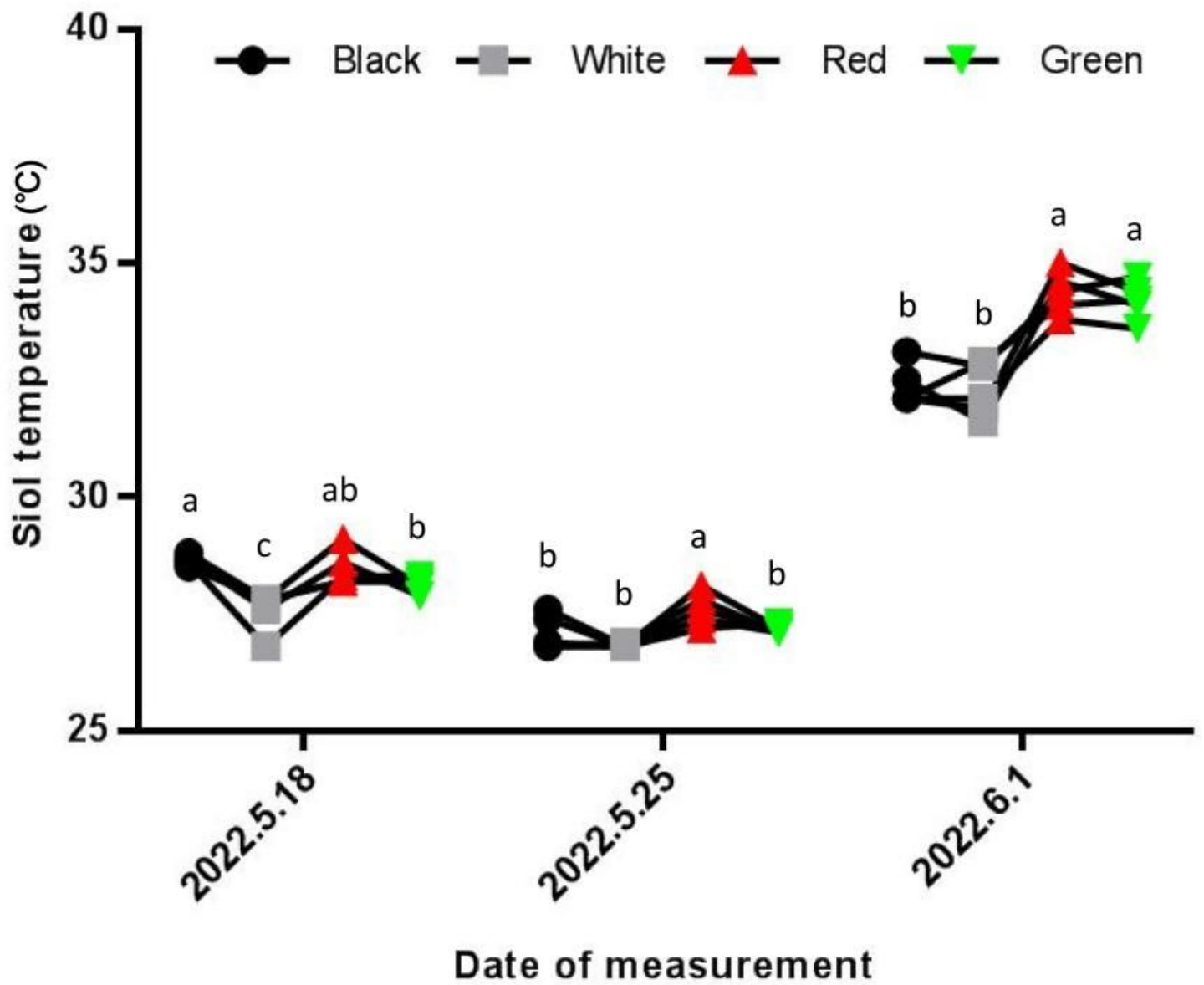


Figure 2

Changes in soil temperature under different treatments at different sampling times.

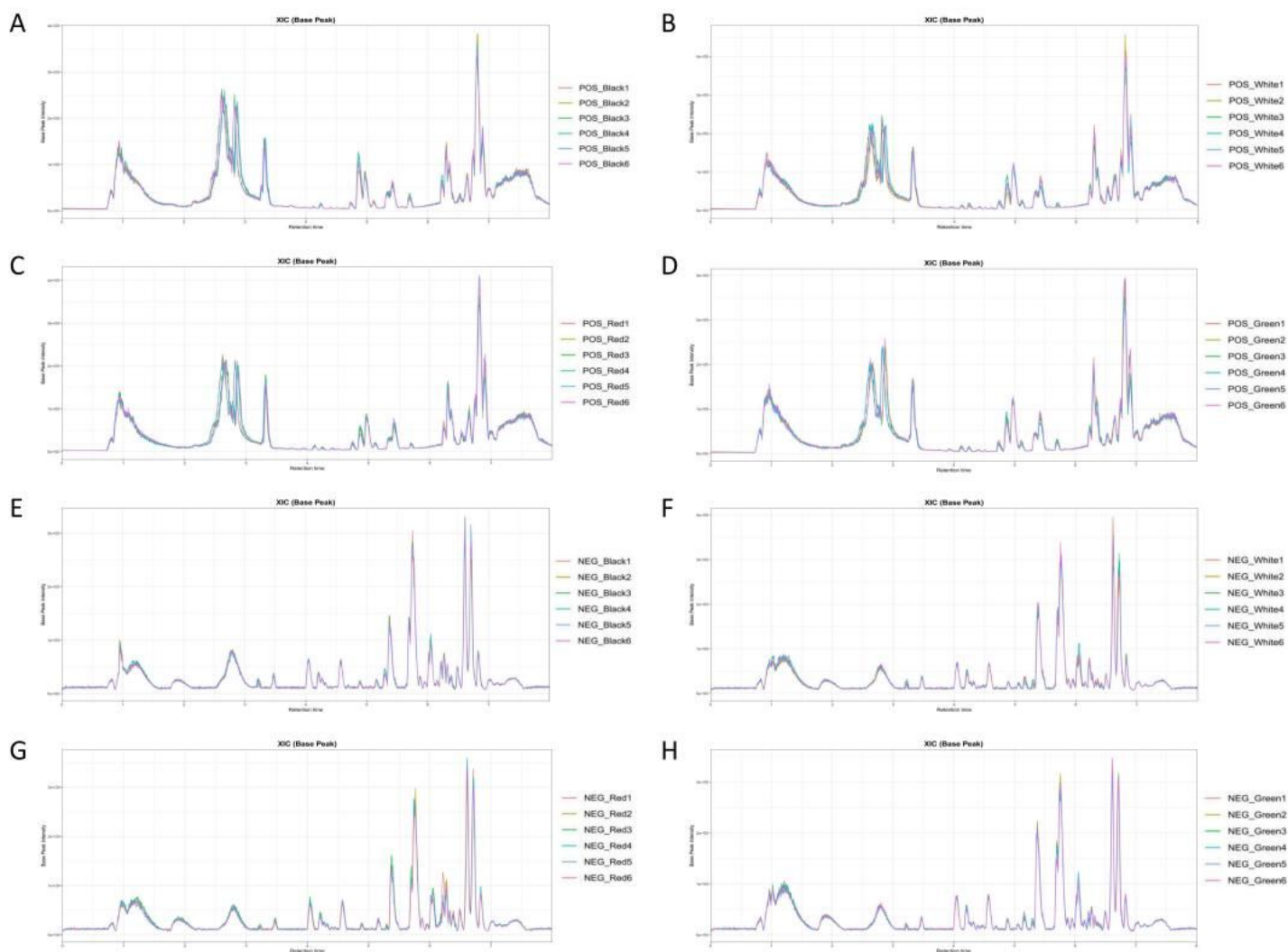


Figure 3

The base peak chromatogram (BPC) of different treatments under positive and negative ion modes. A-D, The base peak chromatogram (BPC) of Black, White, Red, and Green, respectively under positive ion mode. E-H, The base peak chromatogram (BPC) of Black, White, Red, and Green, respectively under negative ion mode.

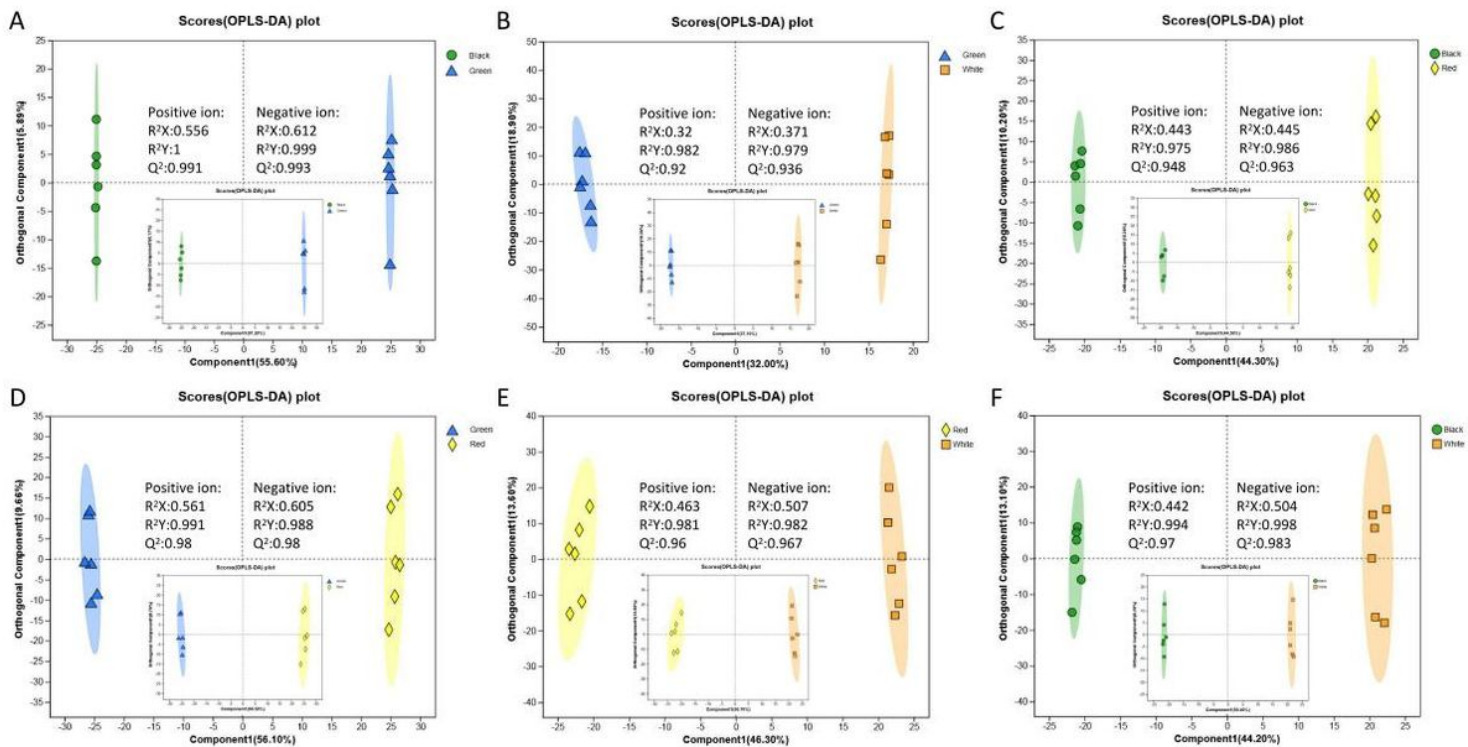


Figure 4

The OPLS-DA plot of different comparison groups. A-F, The OPLS-DA plot of Green_vs_Black, Green_vs_White, Red_vs_Black, Red_vs_Green, Red_vs_White, and White_vs_Black, respectively. The larger and smaller OPLS-DA plots indicated the positive and negative ion modes, respectively.

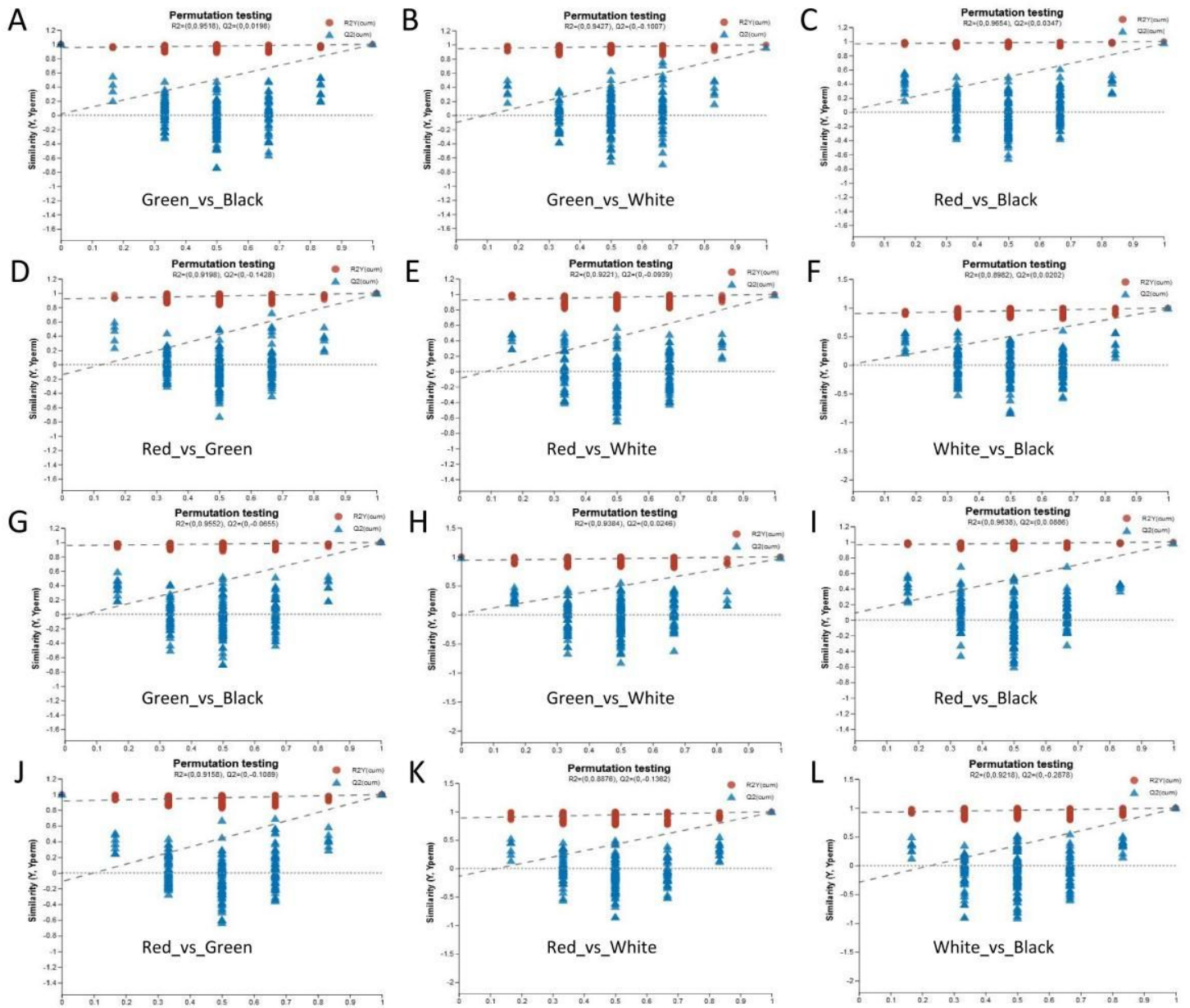


Figure 5

The permutation testing of different comparison groups. A-F, The permutation testing of Green_vs_Black, Green_vs_White, Red_vs_Black, Red_vs_Green, Red_vs_White, and White_vs_Black under positive ion mode, respectively; G-L, The permutation testing of Green_vs_Black, Green_vs_White, Red_vs_Black, Red_vs_Green, Red_vs_White, and White_vs_Black under negative ion mode, respectively.

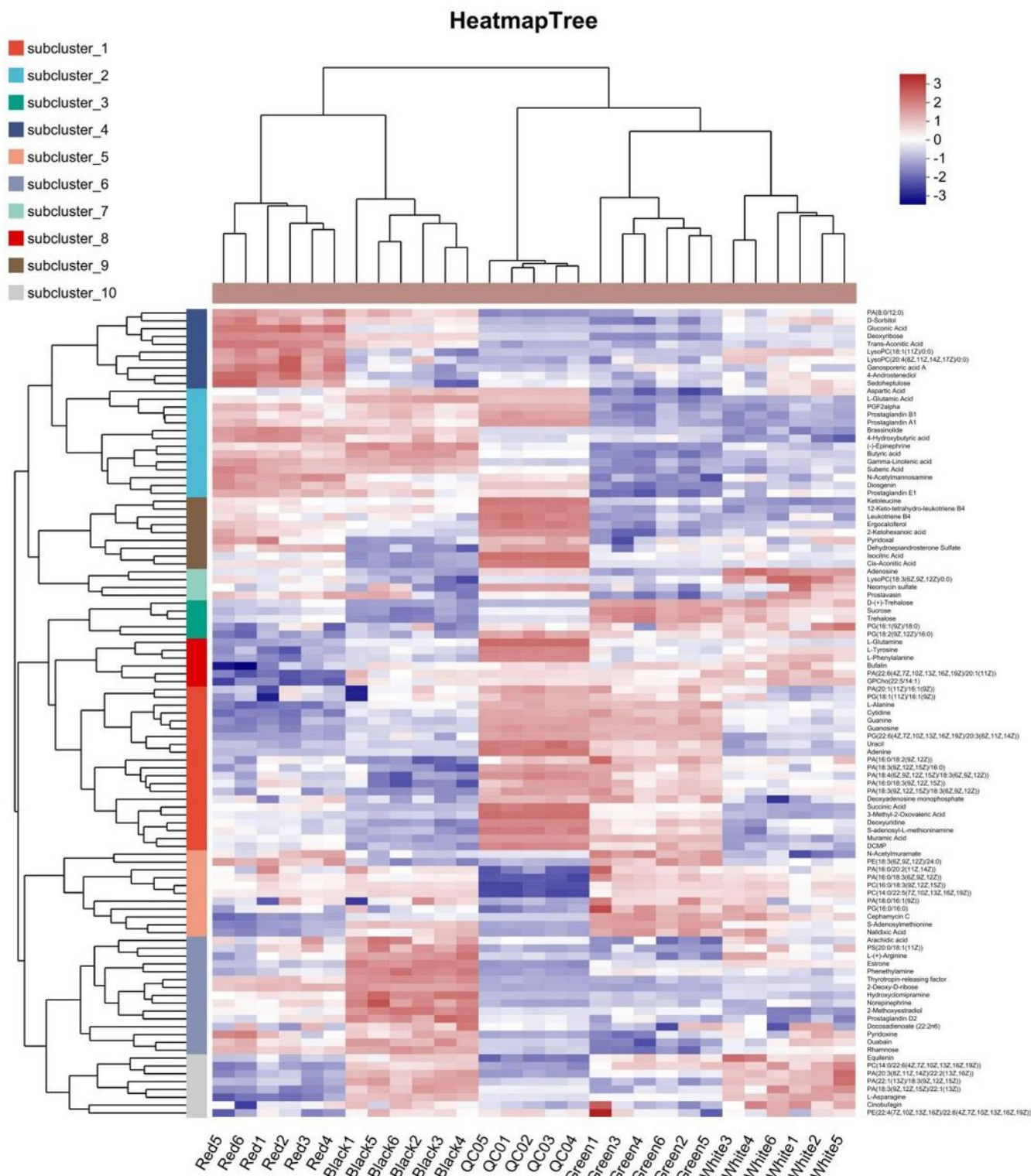


Figure 6

The 103 differential metabolites identified in this study.

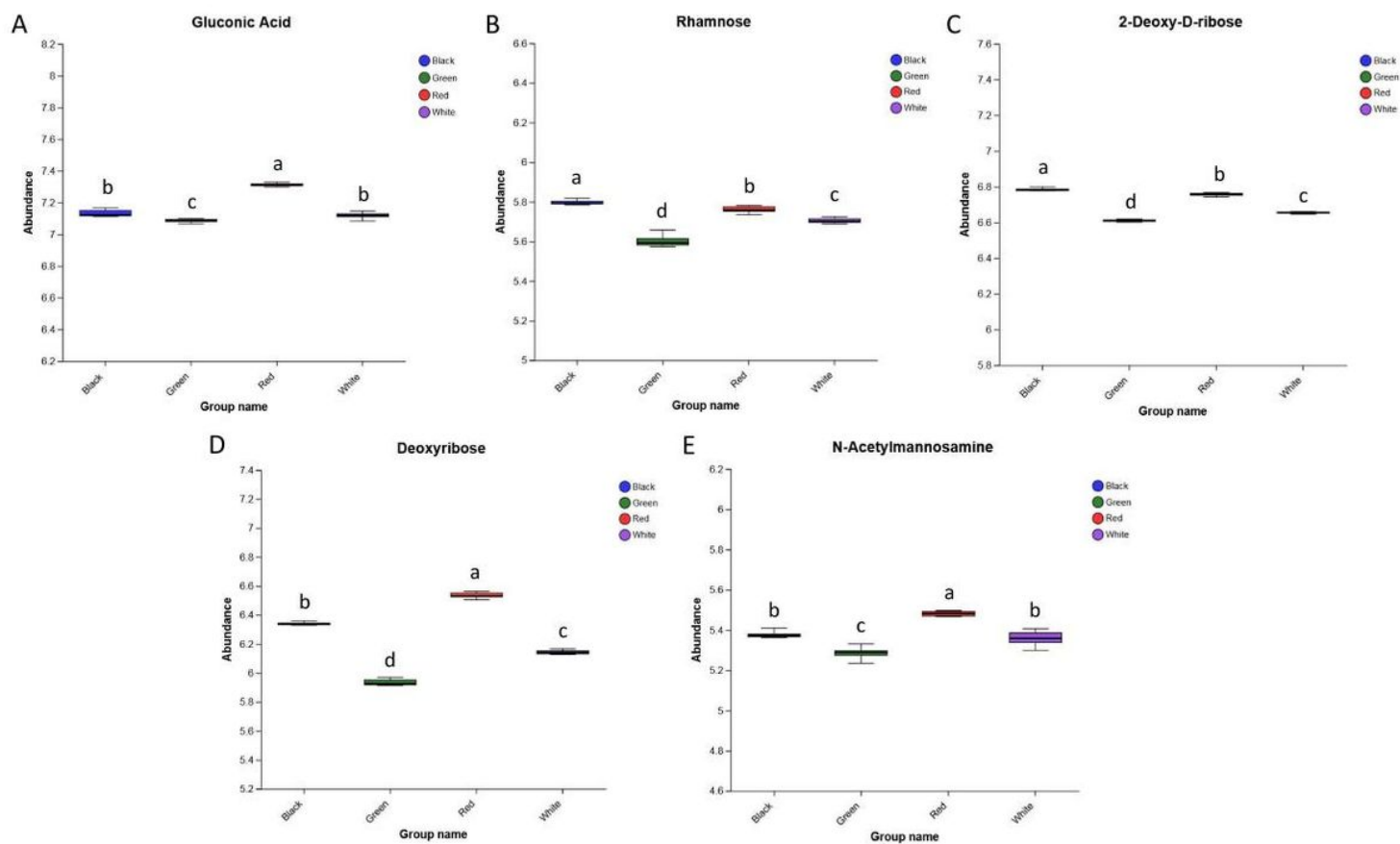


Figure 7

The abundance of differential monosaccharides. A-E, The abundances of gluconic acid, rhamnose, 2-deoxy-D-ribose, deoxyribose, and N-Acetylmannosamine, respectively.

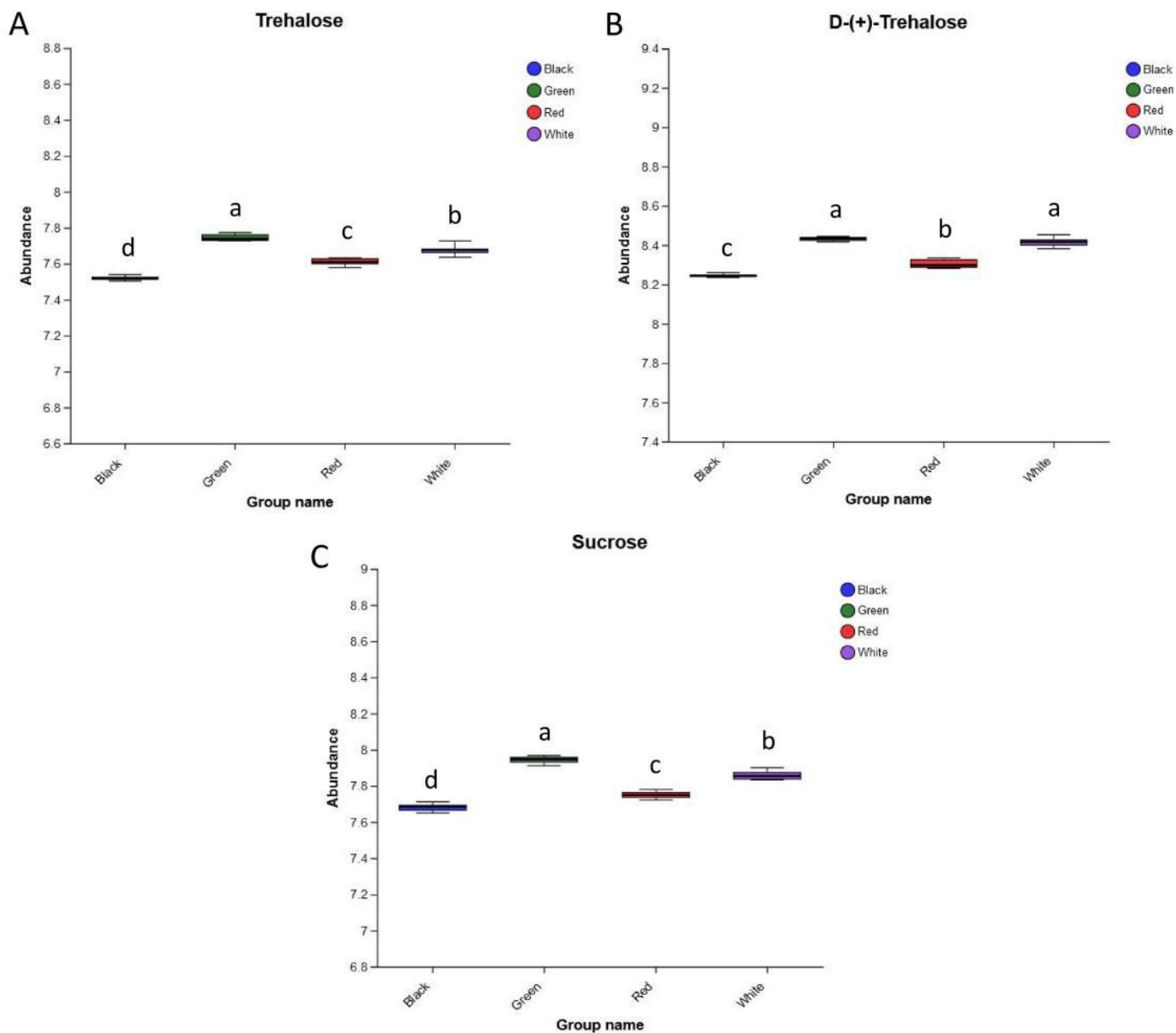


Figure 8

The abundance of differential oligosaccharide. A-C, The abundances of trehalose, D-(+)-trehalose, and sucrose, respectively.

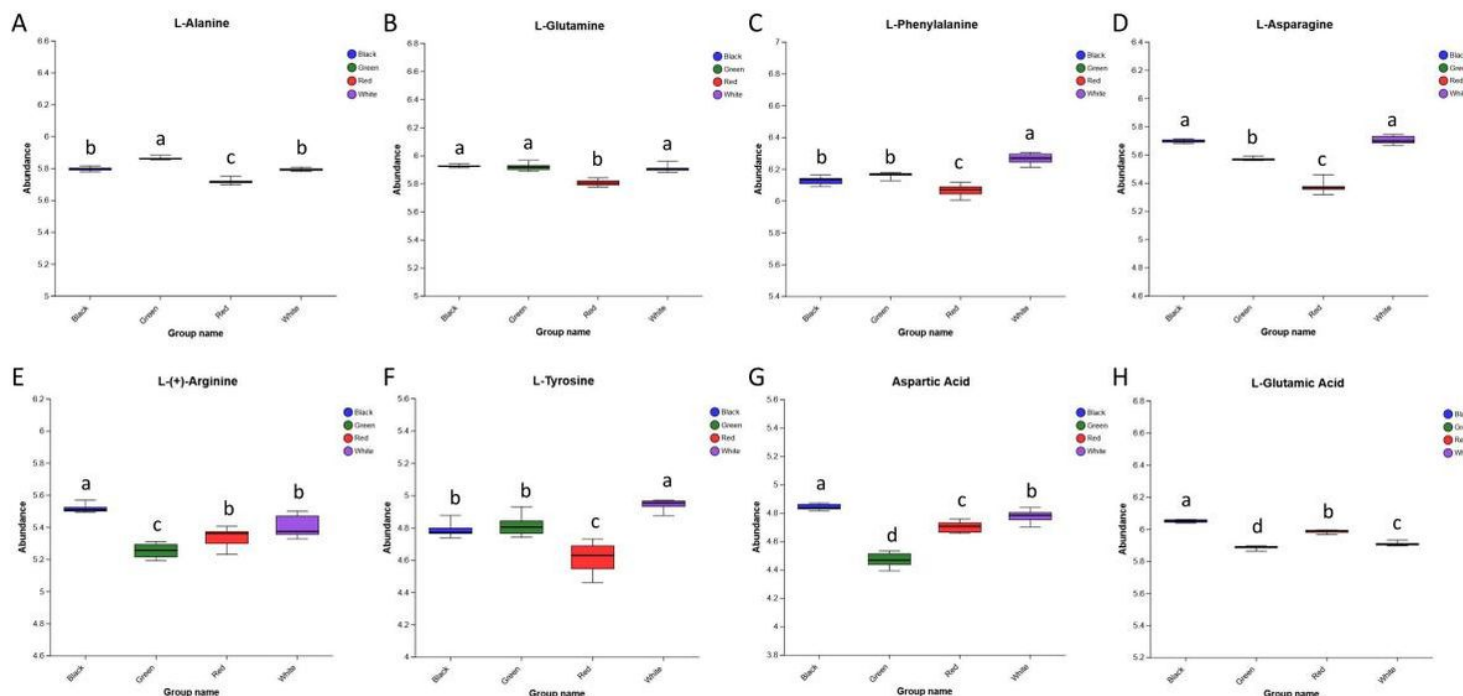


Figure 9

The abundance of differential amino acids. A-H, The abundances of L-alanine, L-glutamine, L-phenylalanine, L-asparagine, L-(+)-arginine, L-tyrosine, Aspartic acid, and L-glutamic acid, respectively.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [Table1.docx](#)
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