Comparative Metabolic Study of two types of R. Cohrysanthums under UV-B Stress

Wang Yu  
Jilin Normal University

Fushuai Gong  
Jilin Normal University

Li Sun  
Siping Central People's Hospital

Kun Cao  
Jilin Normal University

Hongwei Xu  
Jilin Normal University

Xiaofu Zhou (zhouxiaofu@jlnu.edu.cn)  
Jilin Normal University

Research Article

Keywords: metabolome, UV-B stress, R. Cohrysanthum, glyceric acid, amino acids, carbohydrates

Posted Date: May 31st, 2023

DOI: https://doi.org/10.21203/rs.3.rs-2988541/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License. 
Read Full License
Abstract

Background

*Rhododendron chrysanthum Pall (R. chrysanthum.)*, a plant with UV-B resistance mechanisms that can be adapted to alpine environments, has gained attention as an important plant resource with the ability to cope with UV-B stress. Research on the complex mechanism of plant adaptation to UV-B will be aided by these results.

Results

In this study, metabolomics analysis and physiological measurements were used to reveal the response of *Rhododendron chrysanthum* seedlings to UV-B stress. After UV-B irradiation, 404 metabolites were detected. The domesticated *R. chrysanthum* had high amino acid and SOD content.

Conclusions

The study shows that the domesticated *R. chrysanthum* showed significant UV-B resistance. 26 of these different metabolites were classified as UV-B-responsive metabolites. Glyceric acid is used as a potential UV-B stress biomarker.

Background

In recent years, due to the increase of greenhouse gas concentration, climate change is further aggravated, and stratospheric ozone is affected accordingly, leading to the increase of solar ultraviolet radiation (especially UV-B, 280-350nm) on the Earth surface[1, 2]. Although UV-B accounts for a small proportion of total UV, it is traditionally considered as a kind of stress, because it may have various harmful effects on plants, such as reduced growth rate, partial inhibition of photosynthesis, and changes in plant biochemistry[3–6].

*R. chrysanthum* is an evergreen shrub that can withstand low temperatures of around −40°C[7]. *R. chrysanthum* only grows in the highlands of Changbai Mountain, Jilin Province, China, at an altitude of 1,300 to 2,650 meters. The harsh climate and poor soil on the top of Changbai Mountain pose severe challenges for plants. *R. chrysanthum* has developed resistance to UV-B stress and other abiotic stress in the long process of evolution[8–10].

The large amount of metabolites produced within plants play a great role in maintaining their own growth and reproduction and resisting stress. With the progress of science and technology, metabolomics has become an effective method to study plants, revealing how plants respond to stress at the metabolic level[11, 12]. Numerous metabolomics studies have explored the ways in which different plants respond to UV-B stress, which affects the accumulation of many different metabolites in these plants[13–15]. There has been previous experiments involving the application of gas chromatography-time-of-flight
mass spectrometry (GC-TOFMS) in the identification of the metabolomics of R. chrysanthum after UV-B radiation treatment. [16].

This study examined the differences between two types of *R. chrysanthums* in response to UV-B. The purpose of this experiment was as follows ( ) Parallel and metabolic analysis was performed with GC-TOFMS; ( ) Identification of potential UV-B stress biomarkers and UV-B-responsive metabolites in *R. chrysanthum*.

**Materials and Methods**

*Plant Material and Treatment*

*R. crysanthum* belongs to the rhododendron family and grows in the highlands of Changbai Mountain. After being transferred to the lab, they were placed in an artificial climate chamber (18°C (14-h light)/16°C (10-h dark)) under the 50-µmol (photon) m\(^{-2}\) s\(^{-1}\) white light, with a relative humidity of 60%) and intelligent artificial incubator (25°C (14-h light)/18°C (10-h dark)), with a relative humidity of 60% under the white light. The seedlings were cultured for 8 months before the experiment. The plants were subjected to UV-B radiation in an artificial climate chamber to obtain artificially domesticated plants, and the seedlings cultivated in the intelligent artificial incubator were taken as incubator seedlings. The two types of seedlings were subjected to UV-B stress for 8 hours a day. After two days of stress treatment, leaf tissues were collected immediately and frozen in liquid nitrogen. The incubator seedlings without UV-B stress were used as control group (M), and the two seedlings subjected to UV-B (280–315 nm) and PAR (280–315 nm) stress were used as experimental group (N, Q). Mixed sampling was used to exclude errors between individuals, with 6 biological replicates (i.e., 18 plants) in each group.

The artificial UV-B and PAR used in this study were derived from UV-B fluorescent lamps (Philips, TL 20W/01 RS, The Netherlands) and white fluorescent lamps (Philips, T5 × 14W, The Netherlands). Different long pass filters are used to obtain two modes of target radiation. For PAR + UV-B treatment, 320-or 295-nm long-pass filters were placed on the culture flask, respectively. The 400nm long pass filter was placed on the flask to process the PAR. Corresponding to the transmittance function of the long pass filter, the actual UV-B and PAR irradiance of the sample was 2.3 W m\(^{-2}\) UV-B, and PAR of 50 µmol( photon) m\(^{-2}\) s\(^{-1}\).

*Detection of physiological indexes of experimental radiation treatment of R. chrysanthum*

The biological replicates of six sets of plant samples under liquid nitrogen cryopreservation were used for experimental studies. Soluble sugars, Amino acids and SOD content were analyzed by gas chromatography, tofms (Pegasus HT, USA), gas chromatography (7890B, Agilent, Santa Clara, CA, USA), and double-headed sample MPS2 (Gerstel, Muehlheim, Germany).

*Analyzing metabolites quantitatively and qualitatively with GC-TOFMS*
Metabolomics analysis was performed using the XploreMET platform (ver. 3.0, Metab—profile, Shanghai, China). As described above, samples were prepared in the same way and instruments were set up in the same way [17].

The metabolomic data were compared with the JiaLib metabolite database using XploreMET (metabolic - profile, version 3.0) software. This peak area represents the relative quantitative value obtained by integrating the chromatographic peaks of each metabolite.

**Analysis of metabolite data** The PCA method was a multidimensional statistical analysis method used to recognize patterns unsupervised. The filtered data was analyzed for principal component analysis (PCA) using R software ([www.r-project.org](http://www.r-project.org) (Email exchange on 11 May 2023))[18, 19]. MetaboAnalyst 5.0 software ([https://www.metaboanalyst.ca/](https://www.metaboanalyst.ca/), (accessed on 9 December 2023)) was used to analyze metabolite differences in the sample via kegg enrichment analysis. The Pearson correlation coefficient (PCC) and K-means group analysis were performed using Metware Cloud ([https://cloud.metware.cn](https://cloud.metware.cn)), accessed on 10 January 2022.

Interventionary studies involving animals or humans, and other studies that require ethical approval, must list the authority that provided approval and the corresponding ethical approval code.

**The analysis of statistical data**

IBM SPSS statistical software ([http://www.ibm.com/analytics](http://www.ibm.com/analytics)) was used for statistical analyses. Letters indicate the significance level in the results. The data were analysed for ANOVA, using 5% as the criterion for conducting Pearson's correlation test, followed by one-way ANOVA.

**Results**

**Metabolome analysis of R. chrysanthum**

The metabolites of *R. chrysanthum* treated with UV-B radiation were extracted. To investigate the metabolic response of chrysanthemum to UV-B stress, a non-targeted GC-TFORMS assay was used to detect. The result showed that 404 peaks were detected, of which 164 were known metabolites. These known metabolites were divided into 7 groups, including 40 Amino Acids, 37 Carbohydrates, 38 Organic Acids, 10 Nucleotides, 14 Fatty Acids, 10 Lipids, and 15 other metabolites (Fig. 1A).

PCA analysis reflected the characteristics of metabolomics multidimensional data through several principal components, among which the first principal component (PCA1) could explain 18.74% of the features of the original data set, and the second principal component (PCA2) could explain 11.44% of the features of the original data set (Fig. 1B). In addition, correlation analysis was conducted for 18 biological replicates, and Pearson correlation coefficient was used as the standard of biological replicate correlation. The heat map showed that the correlation of 6 biological replicates in group Q was weak, while that in group M and N was highly positive (Fig. 1C). Inconclusion, the experimental design and data were reliable and suitable for further analysis.
Detection of Differentially Metabolites (DMs) in R. chrysanthum in the presence of UV-B

Fold Change and P value were used to screen differential metabolites, and the metabolites meeting FC ≥ 1 or ≤ 0.67 and P < 0.1 were considered differential metabolites. The results showed that there were 22 DMs in the N and M groups, among which 9 DMs were up-regulated and 13 DMs were down-regulated. In the comparison between group Q and group M, there were 38 DMs, among which 26 DMs were up-regulated and 12 DMs were down-regulated. In the comparison between group Q and group N, there were a total of 34 DMs, among which 23 DMs were up-regulated and 11 DMs were down-regulated (Fig. 2A).

After removing the repeated DMs among the three groups, 54 DMs were obtained. Among the 54 DMs, 30% (16/54) Amino Acid, 20% (11/54) Carbohydrates, 18% (10/54) Organic Acids, 4% (2/54) Nucleotide, 6% (3/54) Fatty Acids, 9% (5/54) Lipids, and 13% (7/54) other metabolites (Fig. 2B).

In order to explore the dynamic changes of DMs between different groups, k-means cluster analysis was conducted. The result shows that 4 subclasses can be distinguished. Subclasses 1 to 4 contained 9, 18, 9 and 18 metabolites, respectively (Fig. 2C).

Exploration of UV-B-Responsive Metabolites in R. chrysanthum

Venn diagram was constructed to show the aggregation relationship of DMs in two strains under UV-B stress (Fig. 3). It is proposed to use glyceric acid as a potential UV-B stress biomarker. Duplicate DMs between the comparison groups were removed, and 26 DMs were obtained (Table 1). Among these DMs, amino acids and organic acids predominated and these DMs were treated as UV-B-response metabolites.
Table 1
UV-B-responsive metabolites between different comparison groups were determined using multidimensional statistical analysis.

<table>
<thead>
<tr>
<th>Class</th>
<th>Name</th>
<th>Kegg ID</th>
<th>P value</th>
<th>FC</th>
<th>type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alcohols</td>
<td>2-Hydroxypyridine</td>
<td>C02502</td>
<td>0.081</td>
<td>1.328</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>L-Tyrosine</td>
<td>C00082</td>
<td>0.0019</td>
<td>2.081</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>3-Nitrotyrosine</td>
<td>NA</td>
<td>0.0077</td>
<td>0.149</td>
<td>down</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>L-Serine</td>
<td>C00065</td>
<td>0.021</td>
<td>3.151</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>3-Aminoisobutanoic acid</td>
<td>C05145</td>
<td>0.022</td>
<td>2.037</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>3-Oxoalanine</td>
<td>NA</td>
<td>0.026</td>
<td>1.431</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>L-Methionine</td>
<td>C00073</td>
<td>0.031</td>
<td>0.646</td>
<td>down</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>N-Acetyl-L-aspartic acid</td>
<td>C01042</td>
<td>0.042</td>
<td>2.207</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>Homocysteine</td>
<td>NA</td>
<td>0.06</td>
<td>1.598</td>
<td>up</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>Lactulose</td>
<td>C07064</td>
<td>0.034</td>
<td>2.285</td>
<td>up</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>D-Glucose</td>
<td>C00031</td>
<td>0.059</td>
<td>0.596</td>
<td>down</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>L-Arabitol</td>
<td>C00532</td>
<td>0.08</td>
<td>2.245</td>
<td>up</td>
</tr>
<tr>
<td>Fatty Acids</td>
<td>Palmitoleic acid</td>
<td>C08362</td>
<td>0.007</td>
<td>0.466</td>
<td>down</td>
</tr>
<tr>
<td>Fatty Acids</td>
<td>Myristoleic acid</td>
<td>C08322</td>
<td>0.02</td>
<td>2.057</td>
<td>up</td>
</tr>
<tr>
<td>Hormone</td>
<td>Normetanephrine</td>
<td>C05589</td>
<td>0.028</td>
<td>2.103</td>
<td>up</td>
</tr>
<tr>
<td>Indoles</td>
<td>Indoleacetic acid</td>
<td>C00954</td>
<td>0.00053</td>
<td>0.288</td>
<td>down</td>
</tr>
<tr>
<td>Lipids</td>
<td>Phytol</td>
<td>C01389</td>
<td>0.0026</td>
<td>0.52</td>
<td>down</td>
</tr>
<tr>
<td>Lipids</td>
<td>Cortisol</td>
<td>C00735</td>
<td>0.036</td>
<td>1.563</td>
<td>up</td>
</tr>
<tr>
<td>Lipids</td>
<td>MG182</td>
<td>NA</td>
<td>0.075</td>
<td>2.398</td>
<td>up</td>
</tr>
<tr>
<td>Nucleotide</td>
<td>Uridine</td>
<td>C00299</td>
<td>0.026</td>
<td>16.377</td>
<td>up</td>
</tr>
<tr>
<td>Nucleotide</td>
<td>Guanine</td>
<td>C00242</td>
<td>0.074</td>
<td>0.394</td>
<td>down</td>
</tr>
<tr>
<td>Organic Acids</td>
<td>Glyceric acid</td>
<td>C00258</td>
<td>0.0078</td>
<td>1.833</td>
<td>up</td>
</tr>
<tr>
<td>Organic Acids</td>
<td>Fumaric acid</td>
<td>C00122</td>
<td>0.033</td>
<td>1.57</td>
<td>up</td>
</tr>
<tr>
<td>Organic Acids</td>
<td>Maleic acid</td>
<td>C01384</td>
<td>0.035</td>
<td>1.91</td>
<td>up</td>
</tr>
<tr>
<td>Organic Acids</td>
<td>3-Pyridylacetic acid</td>
<td>NA</td>
<td>0.048</td>
<td>1.963</td>
<td>up</td>
</tr>
<tr>
<td>Organic Acids</td>
<td>3-Hydroxybutyric acid</td>
<td>C01089</td>
<td>0.09</td>
<td>5.289</td>
<td>up</td>
</tr>
<tr>
<td>Class</td>
<td>Name</td>
<td>Kegg ID</td>
<td>P value</td>
<td>FC</td>
<td>type</td>
</tr>
<tr>
<td>------------</td>
<td>-----------------------------</td>
<td>---------</td>
<td>---------</td>
<td>------</td>
<td>------</td>
</tr>
<tr>
<td>Alcohols</td>
<td>2-Hydroxypyridine</td>
<td>C02502</td>
<td>0.081</td>
<td>1.328</td>
<td>up</td>
</tr>
<tr>
<td>Amino Acid</td>
<td>L-Tyrosine</td>
<td>C00082</td>
<td>0.0019</td>
<td>2.081</td>
<td>up</td>
</tr>
<tr>
<td>Vitamin</td>
<td>Alpha-Tocopherol</td>
<td>C02477</td>
<td>0.096</td>
<td>2.113</td>
<td>up</td>
</tr>
</tbody>
</table>

**Metabolic Pathways Enrichment Analysis**

In order to understand the metabolic network pathways of metabolites in *R. chrysanthum* under UV-B radiation, KEGG enrichment pathways of 54 DMs were analyzed (Fig. 4). The results showed that the DMs in response to UV-B stress, Alanine, aspartate and glutamate metabolism, Aminoacyl-tRNA biosynthesis (Table 2).

**Table 2**
The p-value of less than 0.05 is considered statistically significant for metabolic pathway enrichment

<table>
<thead>
<tr>
<th>ID</th>
<th>Pathway name</th>
<th>total</th>
<th>expected</th>
<th>hits</th>
<th>p-Value</th>
<th>FDR</th>
</tr>
</thead>
<tbody>
<tr>
<td>map00250</td>
<td>Alanine, aspartate and glutamate metabolism</td>
<td>28</td>
<td>0.529</td>
<td>5</td>
<td>0.0001</td>
<td>0.00771</td>
</tr>
<tr>
<td>map00970</td>
<td>Aminoacyl-tRNA biosynthesis</td>
<td>48</td>
<td>0.906</td>
<td>6</td>
<td>0.0002</td>
<td>0.00771</td>
</tr>
<tr>
<td>map00270</td>
<td>Cysteine and methionine metabolism</td>
<td>33</td>
<td>0.623</td>
<td>5</td>
<td>0.0003</td>
<td>0.00771</td>
</tr>
<tr>
<td>map00260</td>
<td>Glycine, serine and threonine metabolism</td>
<td>33</td>
<td>0.623</td>
<td>3</td>
<td>0.0226</td>
<td>0.459</td>
</tr>
<tr>
<td>map00524</td>
<td>Neomycin, kanamycin and gentamicin biosynthesis</td>
<td>2</td>
<td>0.0378</td>
<td>1</td>
<td>0.0374</td>
<td>0.459</td>
</tr>
<tr>
<td>map00350</td>
<td>Tyrosine metabolism</td>
<td>42</td>
<td>0.793</td>
<td>3</td>
<td>0.0424</td>
<td>0.459</td>
</tr>
<tr>
<td>map00040</td>
<td>Pentose and glucuronate interconversions</td>
<td>18</td>
<td>0.34</td>
<td>2</td>
<td>0.0437</td>
<td>0.459</td>
</tr>
<tr>
<td>map00500</td>
<td>Starch and sucrose metabolism</td>
<td>18</td>
<td>0.34</td>
<td>2</td>
<td>0.0437</td>
<td>0.459</td>
</tr>
</tbody>
</table>

**Changes in physiology of domesticated *R. chrysanthum***

The degree of alteration of soluble sugars and amino acids were measured separately and used to demonstrate their role in the domestication process. The results were as follows: the UV-B radiation significantly increased the amino acid content of domesticated *R. chrysanthum*, while soluble sugars...
decreased (Fig. 5). According to the results, domesticated *R. chrysanthum* was highly resistant to UV-B radiation.

Superoxide dismutase (SOD, EC 1.15.1.1) is an antioxidant enzyme that removes ROS and plays an important role in plant stress resistance. SOD includes three types: Fe-SOD, Mn-SOD, and Cu/Zn-SOD. SOD expression levels in two *R. chrysanthum* species were determined in this experiment (Fig. 6). The results showed that the expression levels of the three SODs were significantly increased in domesticated *R. chrysanthum*, indicating that domesticated *R. chrysanthum* is remarkably resistant to UV-B radiation.

**Discussion**

Due to the degradation of stratospheric ozone, the amount of UV-B radiation received by Earth and plant surfaces has increased[20]. In addition, in terms of growth, development, and morphology, UV-B strongly affects plants [21–23]. In light of this, the effect of V-B stress on *R. crysanthum* deserves further study. In this experiment, domesticated R. crysanthum was found to be more resistant to UV-B radiation [16]. In this study, it was observed that the biomarker of resistance to potential UV-B stress in *R. cruysanthum*'s UV-B response was glyceric acid, and the UV-B-response metabolites affecting the UV-B response of chrysanthemum were explored.

For the changes in the content of metabolites of both species under UV-B stress, especially the dynamics of amino acids and carbohydrates, osmoregulation is a form of regulation when plants are under stress[24, 25]. Under unfavorable conditions, to reduce the cytosol's osmotic potential and prevent excessive water loss, solutes are actively accumulated in the cell, and the accumulation of sugars plays a critical role in the adaptation of *R. chrysanthum* to UV-B stress, both from a growth and tolerance perspective, and Glucose can act as a signal during plant stress [26–28]. Two different pathways are involved in amylose metabolism and sucrose metabolism in *R. chrysanthum* [29, 30]. It is starch synthesis that takes place first in chloroplasts. As a result of the Calvin cycle, fructose 6-phosphate is converted into glucose 6-phosphate and starch follows. In the second pathway, the cytoplasm serves as a site for the synthesis and breakdown of sucrose [31]. The reduction in sugar transport and starch accumulation is likely to be related to the reduction in carbon flux for leaf sucrose synthesis [32]. The catabolism of starch can mitigate the effects of environmental factors on carbohydrates. [33]. Therefore, *R. chrysanthenum* can respond to U-VB through starch accumulation and catabolism [33]. Starch is one of the main nutritional components of *R. chrysanthum*. The growth, development and physiological state of chrysanthemum are greatly affected by the changes of starch metabolites, which affect the quality of *R. chrysanthum*. In abiotic stress situations, glycolysis increases ATP production, which helps plants adapt to their environment [34]. When in an environment unfavorable to the plant itself, the plant keeps the carbohydrate biosynthetic pathway from being disturbed through the TCA cycle [35]. Thus, the acquisition of greater UV-B resistance in domesticated *R. chrysanthum* may result from the activation of sucrose and starch metabolism.
Plants rely on amino acids to reduce the effects of UV-B radiation on themselves [36]. Amino acids are a vital metabolite in plants, capable of functioning as intermediates in the metabolic process [37–39]. In previous studies, plants treated with UV-B radiation showed a significant increase in the expression of many amino acids internally. As a result of UV-B radiation, the majority of amino acids increase significantly, indicating that *R. crysanthum* accumulates amino acids. In this study, prolonging the stress time significantly increased the serine content in the experimental material. Serine is essential for photorespiration, and the accumulation of Serine suggests that *R. cohrysanthum* can enhance photorespiration under UV-B stress [40]. Succinic acid, as an important intermediate in the TCA cycle, showed higher accumulation under UV-B stress, indicating that UV-B stress promoted the TCA cycle.

Superoxide dismutase (EC 1.15.1.1; SOD) catalyzes the dismutation of superoxide radicals to hydrogen peroxide and oxygen. In higher plants, SOD plays a major role in combating oxygen radical mediated toxicity. The sources of superoxide radical generation may be natural—that is, by-products of metabolic activities, including the electron-transport chain or induced by external agents—including ozone, UV B, gamma rays, light-induced photoinhibitory conditions, or chemicals like paraquat or methyl viologen. Three classes of SOD have been identified, depending on the metals present at the active site: copper/zinc SOD, manganese SOD, and iron SOD. In higher plants the Fe SOD has been isolated from plastids; Mn SOD, from the mitochondrial matrix; and Cu/Zn SOD, from the cytosol[41].

Glyceric acid(GA), i.e., 2,3-dihydroxypropanoic acid, is a simple but attractive derivative of glycerol because of its structure with two kinds of functional groups and chiral isomers. Moreover, GA is the oxidized product of glycerol, and its D-isomer is obtained as a phytochemical from tobacco leaves and some plant fruits[42]. Some biological effects of D-GA on ethanol metabolism have been reported. Ethanol metabolism in rat liver was accelerated by D-GA calcium salt[43]. Another aspect of GA is the protection of biological macromolecules. Radical scission of DNA was protected in the presence of 200 mM GA sodium salt[44]. In this study, glyceric acid was defined as UV-B-responsive metabolites by a comparative metabolomics analysis. Previous studies have shown that its transporter NPF8.4 is responsible for the isolation of photorespiratory carbon intermediate glycerol into vacuoles during nitrogen depletion, which elucidates a new function of photorespiration in nitrogen fusion[45]. The application of glyceric acid in UV-B response may be related to photorespiration and deserves further study.

**Conclusions**

In this study, *R. cohrysanthum* was used as the study subject for metabolomics analysis to study plant responses to UV-B caused by ozone hole. In order to clearly respond to the mechanisms of plant responses to UV-B stress, the pathway map was constructed in order to show the interactions between the DMs. Potential biomarkers and UV-B-responsive metabolites were screened by comparing DMs between groups. The results of this study further reveal the regulatory mechanism of *R. cohrysanthum*’s resistance to ultraviolet B radiation from a metabolomic perspective.
Declarations

Ethics approval and consent to participate

The Plant material was taken from Changbai Mountain in Jilin Province, China, picked with the permission of the local government, and is now stored in the form of tissue culture seedlings in Jilin Provincial Key Laboratory of Plant Resource Science and Green Production. All methods were implemented in accordance with relevant guidelines and regulations.

Consent for publication

Not applicable.

Availability of data and materials

The data used in this study are available from the corresponding author on submission of a reasonable request.

Competing interests

The authors declare no competing interests.

Funding

This work was supported by the Jilin Province Science and Technology Development Plan Project (YDZJ202301ZYTS517).

Authors’ contributions

Writing—original draft preparation, WY and FSG; writing—review and editing, KC; methodology, WY; software, WY and FSG; visualization, WY and FSG; supervision, KC and LS; project administration, HWX and XFZ. All authors have read and agreed to the published version of the manuscript.

Acknowledgments

We are grateful to Jingjie PTM Biolab (Hangzhou, China) Co., Ltd. for providing mass spectrometry support.

References


Figures

**Figure 1**

(A) The amount of metabolites and differential metabolites detected in different comparison groups. By using the hypergeometric test, the statistical significance was determined with * P < 0.1. (B) Principal component analysis (PCA) of different comparison groups. (C) Heatmap of Pearson’s correlation coefficient. Different colors are used to represent different groups.

**Figure 2**

(A) Statistics of up-regulated and down-regulated DMs. (B) DMs classification statistics. (C) The K−means analysis of DMs.
Figure 3

(A) Up-regulated DMs statistics. (B) Down-regulated DMs statistics

Figure 4
(A) Pathway Enrichment analysis of DMs. (B) The simplified metabolic model based on a heatmap of DMs. Screening DMs by log2 FC. The color of the border means the shift in expression of the corresponding DMs, with red indicating upregulation and green indicating downregulation.

Figure 5

The reaction of UV-B on *R. chrysanthum*'s physiological indicators. (A) The changes of amino acid content. (B) The changes of soluble sugars content.
Figure 6

Detection of three kinds of SOD expression changes. (A) The changes of amino Fe-SOD expression. (B) The changes of Mn-SOD expression. (C) The changes of Cu/Zn-SOD expression.