Methyl jasmonate modulates metabolism of mulberry leaves: Effects on saccharides and phenols content

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Mulberry leaves produced by the mulberry tree are traditionally used in silkworm rearing as well as herbal medicine, contain a variety of active ingredients with antioxidant, hypoglycemic, anti-cholesterol, anti-obesity, anti-inflammatory and anticancer activities (Afzal et al., 2021). The phytochemistry and pharmacology of mulberry leaves have given them both traditional and current uses as medicines, food, feed and cosmetics (Ma G et al., 2022). The current research focuses on the enrichment of gamma-aminobutyric acid in mulberry leaves (Tu et al., 2022; Li et al., 2018; Han et al., 2012). There are also studies on the determination of flavonoids and phenolic acids in mulberry leaves collected at different times (Levickiene et al., 2017). However, there are few studies on the enrichment of other active components of mulberry, such as saccharides, protein, phenols, and flavonoid, so it is of great significance to enrich many active components of mulberry leaves.

Elicitor treatment is an effective way to regulate plant metabolites, among which jasmonic acid, salicylic acid and ethylene, as key hormone signals of plant immune pathway, play a vital role in plant growth and metabolism (Pieterse et al., 2009). It has been shown that the use of methyl jasmonate on tomato fruits can induce the production of amino acids, fatty acids, tocopherols, and phytosterols, and that ethylene can increase sugar accumulation (Meza et al., 2022). The use of methyl jasmonate in kiwi fruit can promote secondary metabolism, improve secondary metabolites and enhance disease resistance (Li et al., 2022). Salicylic acid sprayed on labiaceae increased the contents of hydroxycinnamic acid, total phenolic compounds and flavonoids, and also increased the hypoglycemic activity and antioxidant activity (Skrypnik et al., 2022). Therefore, methyl jasmonate was used in this study to induce mulberry leaf metabolism and improve its application.

Firstly, water content was determined, and then water extract of mulberry leaf was prepared to determine saccharides and protein of mulberry leaf, alcohol extract of mulberry leaf was prepared to determine phenolss and flavonoids of mulberry leaf. Saccharidess were determined by phenol-sulfuric acid method (Nielsen et al., 2022), soluble proteins were determined by Coomash bright blue method (Wang et al., 2022), phenolss were determined by folin-ciocalteu method (Vingrys et al., 2022), and flavonoids were determined by sodium nitrite-aluminum nitrate-sodium hydroxide colorimetry method.

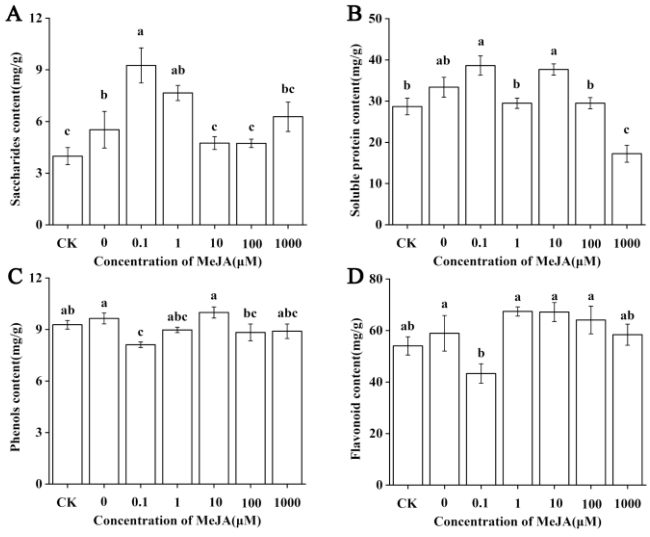

Figure.1 After spraying mulberry leaves with different concentrations of methyl jasmonate, the dry weight content of mulberry leaves.
(A) Saccharides content; (B) Soluble protein content; (C) Phenols content; (D) Flavonoid content.

The results showed that (1) Spraying water could promote the growth of mulberry leaves and increase the contents of all components, among which saccharidess increased up to 38.45%; (2) In mulberry leaves sprayed with 0.1μM methyl jasmonate, saccharides content was significantly increased by 132.05%, protein content was significantly increased by 29.76%, phenols content was significantly reduced by 12.51%, and flavone content was

significantly reduced by 19.87%; (3) Mulberry leaves sprayed with 10μM methyl jasmonate increased phenolss content by 7.70%, flavonoids content by 24.30%, protein content significantly by 26.87%, and saccharides content by 19.02%. (4) With the concentration of methyl jasmonate increased from 0.1μM to 10μM, primary metabolism decreased, secondary metabolism increased, primary metabolites saccharides and protein decreased, secondary metabolites phenolss and flavonoids increased. This is similar to the result of jasmonic acid signaling promoting secondary metabolism (Rubio-Rodriguez et al., 2021). When the concentration of methyl jasmonate is too high, the content of phenols, flavonoid and protein decreases, which affects the normal physiological metabolism of plants. The highest protein content was 0.1μM and 10μM methyl jasmonate, which may be the result of the increase of primary metabolism-related proteins and secondary metabolism-related proteins.

In conclusion, after spraying mulberry leaves with 0.1μM methyl jasmonate, mulberry leaves with high content of saccharides and protein, low content of anti-nutrition factors phenolss and flavonoids can be obtained, which is suitable for feed industry. After spraying 10μM methyl jasmonate, the contents of phenolss and flavonoids of mulberry leaves were higher, and the antioxidant properties were stronger. Further evaluation is needed for the variation of phenols species and the difference of application function in treated mulberry leaves.

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References

[1] Afzal, F., Khalid, W., Asif, M., Jabeen, A., Jha, R., Khalid, M.Z., Fizza, C., Aziz, A., Akram, R., Bashir, A., Younas, S., Nayyer, F., Yasin, R., Ahmad, M., 2021. 'Role of Mulberry Leaves in Human Nutrition: A Review'. *ACTA SCIENTIFCI NUTRITIONAL HEALTH*, 4: 43-50.

[2] Ma, G., Chai, X., Hou, G., Zhao, F., Meng, Q., 2022. 'Phytochemistry, bioactivities and future prospects of mulberry leaves: A review'. *FOOD CHEMISTRY*, 372: 131335.

[3] Tu, J., Liu, G., Jin, Y., Tang, C., Yao, T., Zhuo, J., Li, Q., Liu, L., Wang, J., 2022. 'Enrichment of gamma-aminobutyric acid in mulberry leaves and the inhibitory effects of the water extract on ACE and alpha-glucosidase activity'. *INDUSTRIAL CROPS AND PRODUCTS*, 177: 114485.

[4] Li, E., Luo, X., Liao, S., Shen, W., Li, Q., Liu, F., Zou, Y., 2018. 'Accumulation of gamma-aminobutyric acid during cold storage in mulberry leaves'. *INTERNATIONAL JOURNAL OF FOOD SCIENCE AND TECHNOLOGY*, 53 (12): 2664-2672.

[5] Han, K., Lee, Y., Song, J., Hwang, Y., Lee, W., Kim, M., Kim, S., 2012. 'Enhanced production and secretion of rutin and GABA in immobilized cells of mulberry tree (Morus bombycis K.)'. *PLANT CELL TISSUE AND ORGAN CULTURE*, 108 (3): 513-520.

[6] Levickiene, D., Jariene, E., Gajewski, M., Danilcenko, H., Vaitkeviciene, N., Przybyl, J.L., Sitarek, M., 2017. 'Influence of Harvest Time on Biologically Active Compounds and the Antioxidant Activity in Leaves of Mulberry Grown in Lithuania'. *NOTULAE BOTANICAE HORTI AGROBOTANICI CLUJ-NAPOCA*, 45 (2): 431-436.

[7] Pieterse, C.M.J., Leon-Reyes, A., Van der Ent, S., Van Wees, S.C.M., 2009. 'Networking by small-molecule hormones in plant immunity'. *NATURE CHEMICAL BIOLOGY*, 5 (5): 308-316.

[8] Meza, S., Tobaruela, E.D.C., Pascoal, G.B., Magalhaes, H.C.R., Massaretto, I.L., Purgatto, E., 2022. 'Induction of Metabolic Changes in Amino Acid, Fatty Acid, Tocopherol, and Phytosterol Profiles by Exogenous Methyl Jasmonate Application in Tomato Fruits'. *PLANTS-BASEL*, 11: 3663.

[9] Li, S., Xiao, L., Chen, M., Cao, Q., Luo, Z., Kang, N., Jia, M., Chen, J., Xiang, M., 2022. 'The involvement of the phenylpropanoid and jasmonate pathways in methyl jasmonate-induced soft rot resistance in kiwifruit (Actinidia chinensis)'. *FRONTIERS IN PLANT SCIENCE*, 13: 1097733.

[10] Skrypnik L., Golovin A., Savina T., 2022. 'Effect of salicylic acid on phenolic compounds, antioxidant and antihyperglycemic activity of Lamiaceae plants grown in a temperate climate.'. *FRONTIERS IN BIOSCIENCE (ELITE EDITION)*, 14 (1): 1-3.

[11] Nielsen S.S., Nielsen S.S., 2017. 'Total carbohydrate by phenol-sulfuric acid method'. *FOOD ANALYSIS LABORATORY MANUAL*, 137-141.

[12] Wang, Y., Yu, Q., Li, Y., Li, J., Chen, J., Liu, Z., Huang, J., Al-Harbi, M.S., Ali, E.F., Eissa, M.A., 2021. 'Mechanisms of Nitric Oxide in the Regulation of Chilling Stress Tolerance in Camellia sinensis'. *HORTICULTURAE*, 7: 41010.

[13] Vingrys, K., Mathai, M., Ashton, J.F., Stojanovska, L., Vasiljevic, T., McAinch, A.J., Donkor, O.N., 2022. 'The effect of malting on phenolic compounds and radical scavenging activity in grains and breakfast cereals'. *JOURNAL OF FOOD SCIENCE*, 87 (9): 4188-4202.

[14] Song F., Wang L., Mu J., Ma, H., 2022. 'Protective Effects and Molecular Mechanism of Total Flavonoids from Lycium Barbarum Leaves on Photoaged Human Dermal Fibroblasts'. *EVIDENCE-BASED COMPLEMENTARY AND ALTERNATIVE MEDICINE*, 2022: 4156330.