3 - 53 Determination Anthocyanin Content in Donghuaxiaocao by pH Differential Method and HPLC

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Anthocyanins are a colored class of flavonoids that are responsible for the pink, red, violet and blue colors of flowers. The anthocyanins are water-soluble and unstable which are extracted from grapes, berries, red cabbage, apples, radishes, tulips, roses and orchids. Donghuaxiacao is a mutant of Wandering Jew induced by carbon ions radiation. The previous research found that light pink patches appeared on the basal part of adaxial surface of the young leaves since the first 2 weeks of September. Meanwhile, purple occurred on abaxial surface. Pink patch enlarged gradually, and it would not fade until March the next year^[1]. All above accounts indicated that the anthocyanins are primary pigment in purple leaves of Donghuaxiacao. In order to determination the anthocyanin content in Donghuaxiaocao, pH differential method and HPLC were used in this study.

Total anthocyanin content in Donghuaxiacao was measured using the pH differential method^[2]. The anthocyanin extract was dissolved in 0.025 mol/L potassium chloride buffer, pH 1.0 and 0.4 mol/L sodium acetate buffer, pH 4.5 with a pre-determined dilution factor. The absorbance was measured at 520 nm and 700 nm. The absorbance (A) of the diluted sample was then calculated as follows:

$$A = (A520 - A700)_{pH \ 1.0} - (A520 - A700)_{pH \ 4.5}$$

The Donghuaxiaxao and its control anthocyanin concentration in the original sample was calculated in cyanidin equivalents according to the following formula:

Anthocyanin content= $A \times MW \times DF \times 1000/\epsilon \times 1$

As shown in Table 1, total anthocyanin content was significantly different between Donghuaxiacao and its control. The total anthocyanin content of Donghuaxiacao is reached to 0.051 mg/g. However, it is hardly found some anthocyanin in control. The study indicated that anthocyanins were distributed in purple leaves of Donghuaxiacao. The result suggested that anthocyanins are responsible for the color of purple leaves of Donghuaxiacao. However, the color of purple leaves of Donghuaxiacao, especially the color stability, is not only related to the total content of anthocyanins, but also related to the type of anthocyanins. Hence, analysis of individual anthocyanins would be critical for the understanding of the pigment stability of purple leaves of Donghuaxiacao for processing purposes.

Table 1 Total anthocyanin content in Donghuaxiacao(n=3)

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	CK	Donghuaxiacao
Anthocyanin content(mg/g)	0.001 ± 0.001	0.051±0.004*

Results were the average of three analyses and expressed as mean \pm SD; * the difference of anthocyanin content was significant different between control and Donghuaxiacao, p < 0.05.

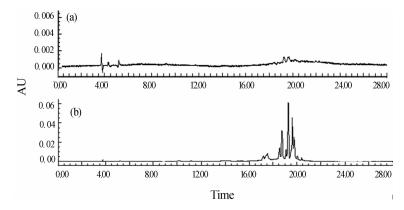


Fig. 1 HPLC chromatograms of anthocyanins in leaves of control(a) and Donghuaxiacao(b).

Individual anthocyanins in Donghuaxiacao and its control were identified using the HPLC method. Fig. 1 demonstrates the separation of Donghuaxiacao and control leaves by using the HPLC method. Under our experimental conditions, the most pigment identified in purple leaves of Donghuaxiacao, and incontrol's

green leaves, neither pigment was detected by HPLC. This result showed that increases in the intensity of anthocyanin metabolites in Donghuaxiacao. However, the exact individual anthocyanins were not identified.

These results encouraged us to identify the exact individual anthocyanins and isolate the genes involved in anthocyanin metabolism from purple leaves of Donghuaxiacao in the future.

References

- [1] H. J. Yu, W. J. Li, H. M. Xie, IMP & HIRFL Annual Report, (2010)141.
- [2] Z. L. Huang, B. W. Wang, Paul Williams, et al., LWT-Food Science and Technology, 42(2009)819.

3 - 54 Study on Cd²⁺-resistant and Adsorption Capacity of a Bacterium Mutated by Heavy Ions

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Heavy metal pollution is a serious environmental problem in recent years. Besides the toxic and harmful effects to organisms living in water, heavy metals also accumulate throughout the food chain and may affect human beings. The traditional method for removing heavy metals, such as ion exchange, chemical precipitation, reverse osmosis, evaporation, membrane filtration, suffer from some drawbacks, high capital and operational cost or the disposal of the residual metal sludge. An emerging and attractive method, biosorption involves sorption of dissolved substances by a biomaterial, of which advantages is low operating cost, short operation time, and no production of secondary compounds which might be toxic^[1].

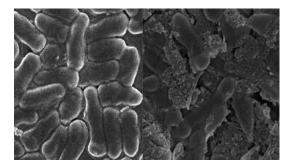


Fig. 1 Scanning Electron Microscopy(SEM) of C2 cultivated on the concentration of Cd^{2+} was 0 and 100 mg/L(\times 10⁴).

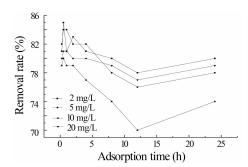


Fig. 2 Effect of Cd^{2+} concentration on the biosorption efficacy of the bacteria powder.

A bacterial strain C2 isolated from sludge was mutated by heavy ion beams, which was tolerated with Cd^{2+} . C2 was able to grow on Cd^{2+} concentration $\leq 100 \text{ mg/L}$, while the growth was inhibited with the Cd^{2+} concentration increased and a part of bacteria reshaped. Scanning Electron Microscopy (SEM) of C2 indicated that the extracellular products were produced by the microorganism, which could combine with Cd^{2+} formed complex, to maintain growth environment of C2 appropriate.

Research showed that bacteria powder of C2 could be used as biosorbent for the removal of Cd^{2+} from aqueous solution and the adsorption capacity of bacteria powder was good. when the concentration of Cd^{2+} increased from 2 to 20 mg/L and the oncentration of bacteria powder was 1.0 g/L, the removal rate of Cd^{2+} reached 90% by the bacteria powder and the optimum adsorption time was approximately 30 min.

Reference