

# Betalain extraction for photometry

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**Tags:** *Betalain HPLC Photometry*

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## Material

- ~ 30 mg material
- 275 µl H<sub>2</sub>O (minimum)
- 250 µl methanol (HPLC grade with internal standard)
- 500 µl chloroform (HPLC grade)
- Liquid nitrogen
- 1.5 ml safe-lock low protein binding tubes

## Procedure

- Weight plant material, increase solvent volume based on initial weight to ensure all samples are comparable (similar fresh weigh per volume solvent ratio)
- Homogenize plant material in liquid nitrogen using pistil, cool pistil using liquid nitrogen also
- Always keep tubes on ice between steps
- Add H<sub>2</sub>O, incubate at 1,200 rpm for 60 min on shaker at 8 °C
- centrifuge at 13,000 rpm for 10 min at 4 °C, transfer 250 µl of supernatant to new tube
- add methanol and chloroform, incubate at 1,200 rpm for 10 min on shaker at 8°C
- centrifuge at 13,000 rpm for 10 min at 4 °C, transfer 400 µl of upper phase to new tube
- Upper phase represents the final extract



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