Quantification and Comparison of Protein Content in Photosynthetic and Non-Photosynthetic Tissues of Six Plants at KCC

Jazmin Ausley and Farshad Tamari, Ph.D.
Kingsborough Community College, Brooklyn, NY, USA.

Introduction

Goal: In this investigation, we plan to determine the total protein content in photosynthetic tissues of six plant species, and in one species, we compared protein content of photosynthetic vs non-photosynthetic tissues.

Hypothesis:

1) Photosynthetic tissues of six different species will have the same relative protein content due to functional similarities
2) Non-photosynthetic tissues will have different levels of protein content due to different functional properties

Plant Species

Primula vulgaris is a slightly poisonous plant to some animals (NSCU) that is very colorful and small, so it is still commonly planted for its aesthetics (Li et al, 2020)

Pinus thunbergii is consumed for its number of health benefits such as anti-inflammation, hair-growth stimulation (Her et al, 2022) and fighting disorders like diabetes (Yoon et al, 2021).

Pinus strobus, the pine tree commonly used as Christmas trees, is native to North America and can grow up to 80 feet high (Gilman et al, 2015).

Taxus cuspidata, is shrubby and more commonly is known as Japanese yew (Zhao et al, 2022) with needles that have taxol used to fight cancer *(Kuang et al, 2019)*

Rhododendron ponticum originates from Iberia (Manzoor et al, 2020). It is considered an invasive species with its crippling ef fects on the natural fauna affecting European countries (Tyler et al, 2006).

Hydrangea macrophylla is a common shrub found in Asia or North America (Dilshara et al, 2013) that can come in a variety of colors (Negishi et al, 2012). It has many medicinal properties as well, like in China where the leaves of this plant are consumed as a tea for the phyllodulcin it naturally produces, an alternative sweetener substitute for people suffering from diabetes (Jung 2016).

Results

Figure One: Our results indicate that there is quite a bit of variability in the protein content of photosynthetic tissues when comparing the six species, ranging from 0.31 to 2.40 µg/µL of protein. *P. vulgaris* appears to have the lowest protein content, while *R. ponticum* appears to have the highest.

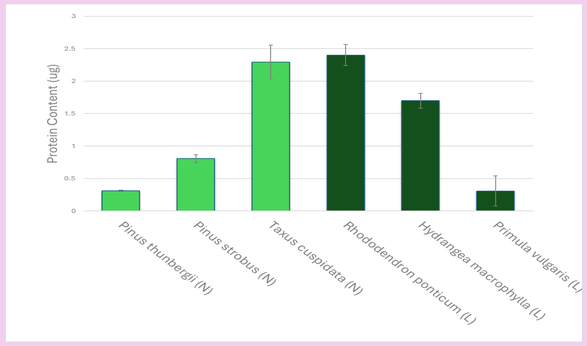
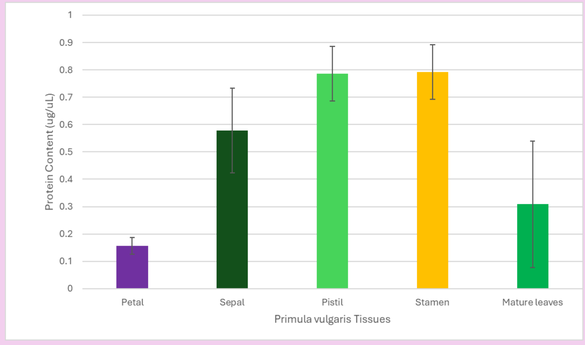


Figure Two: For non-photosynthetic tissues we used petal, pistil and stamen from *P. vulgaris* in addition to the photosynthetic tissues of leaves and sepals. The highest protein content was observed for stamens, with the lowest belonging to the petals. The range of protein content for *P. vulgaris* tissues was from 0.16 to 0.79 µg/µL.



Conclusion

1) Photosynthetic tissues of six different species will have the same relative protein content due to functional similarities (unsupported)
   - Varying levels show that just because all the selected tissue that was analyzed were photosynthetic, they did not all display a similar protein concentration
2) Non-photosynthetic tissues will have different levels of protein content due to different functional properties (supported)
   - Although all the samples were from the same *P. Vulgaris* plant, the amount of protein shown with each tissue were not similar to one another

Discussion

Error Analysis: The needles were fibrous and hard to work with, they were as easily able to be crushed the same as the leaf samples

Future Work: A greater sample size will be used for other experiments to yield more accurate results for each sample or tissue being investigated.

A statistical analysis was completed.

Materials & Methodology

All samples, with the exception of *P. Vulgaris* which was purchased separately, were collected from the Kingsborough campus with permission from Buildings and Grounds. Needles were collected from *P. thunbergii, P. strobus, T. cuspidata*. Leaves were collected from *R. ponticum and H. macrophylla*. *P. vulgaris,* in addition to leaves, had a collection of the sepal, petal, pistil and stamen.

Every extraction was made up of 50 µg of plant tissue crushed in 200µL of Phosphate Buffered Saline (PBS) until a homogeneous mixture was obtained. Between one to five microliters of tissue was used in triplicates for the quantification of total protein content. The Bradford Assay, is a colorimetric and spectrophotometric tool used to determine protein concentrations. While it has limitations, it is widely used to quantify protein content in a variety of settings due to its ease of use as well as its efficiency and accuracy in producing results quickly (Jones et al, 1989). It measures of 595 nanometers (λ = 595 nm). In this investigation, the Bradford Assay was used to quantify proteins from photosynthetic and non-photosynthetic tissues. A standard Bradford assay (BIO-RAD) with the xMark Microplate Absorbance Spectrophotometer (also BIO-RAD) generated results. Results were analyzed using Microsoft Excel and SPSS (IBM)

This research was made possible through funds made available to FT by CUNY's "CUNY Research Scholars Program"

References

- *Bradford Assay*. (n.d.). Bio-Rad Laboratories. https://www.bio-rad.com/en-us/feature/bradford-assay.html
- *Bradford Protein Assay*. (n.d.). Bio-Rad Laboratories. https://www.bio-rad.com/en-us/feature/bradford-protein-assay.html
- Gilman, E. F., & Watson, D. G. (1994, October). *ENH-632/ST473: Pinus strobus: Eastern White Pine*. Edis.ifas.ufl.edu. https://edis.ifas.ufl.edu/publication/ST473
- Her, Y., Lee, T.-K., Sim, H., Jae Sung Lee, Dae Won Kim, Soo Young Choi, Jun Sung Hong, Ji Hyun Lee, Jong Min Kim, Moo Ho Won, & Sung Wan Kim. (2022). *Pinus thunbergii bark extract rich in flavonoids promotes hair growth in dorsal skin by regulating inflammatory cytokines and increasing growth factors in mice*. *25*(3). https://www.spandidos-publications.com/10.3892/mmr.2022.12616
- Jung, C.-H., Kim, Y., Kim, M.-S., Lee, S., & Yoo, S.-H. (2016). The establishment of efficient bioconversion, extraction, and isolation processes for the production of phyllodulcin, a potential high-intensity sweetener, from sweet hydrangea leaves (Hydrangea macrophyllaThunbergii). *Phytochemical Analysis*, *27*(2), 140–147. https://analyticalsciencejournals.onlinelibrary.wiley.com/doi/10.1002/pca.2609
- Kuang, X., Sun, S., Wei, J., Li, Y., & Sun, C. (2019). Iso-Seq analysis of the Taxus cuspidata transcriptome reveals the complexity of Taxol biosynthesis. *BMC Plant Biology*, *19*(1). https://bmcplantbiol.biomedcentral.com/articles/10.1186/s12870-019-1809-8
- Li, L., Ye, J., Li, H., & Shi, Q. (2020). Characterization of Metabolites and Transcripts Involved in Flower Pigmentation in Primula vulgaris. *Frontiers in Plant Science*, *11*, 572517. https://www.frontiersin.org/journals/plant-science/articles/10.3389/fpls.2020.572517/full
- Manzoor, S. A., Griffiths, G., Obiakara, M. C., Esparza-Estrada, C. E., & Lukac, M. (2020). Evidence of ecological niche shift in Rhododendron ponticum (L.) in Britain: Hybridization as a possible cause of rapid niche expansion *Ecology and Evolution*, *10*(4). https://onlinelibrary.wiley.com/doi/10.1002/ece3.6036
- *Primula vulgaris (Primrose) | North Carolina Extension Gardener Plant Toolbox*. (n.d.). Plants.ces.ncsu.edu. Retrieved March 7, 2024, from https://plants.ces.ncsu.edu/plants/primula-vulgaris/
- Tyler, C., Pullin, A. S., & Stewart, G. B. (2006). Effectiveness of Management Interventions to Control Invasion by Rhododendron ponticum. *Environmental Management*, *37*(4), 513–522. https://doi.org/10.1007/s00267-005-0127-0
- Yoon, C. J., Choi, W. S., Kang, H. S., Kim, H. J., Lee, W. T., Lee, J. S., Lee, S., Son, S. Y., Lee, C. H., Sohn, U. D., & Lee, J. Y. (2021). Pinus thunbergii Parl. Extracts Reduce Acute Inflammation by Targeting Oxidative Stress. *Evidence-Based Complementary and Alternative Medicine*, *2021*, 1–10. https://doi.org/10.1155/2021/7924645
- YOSHIDA, K., OYAMA, K., & KONDO, T. (2021). Insight into chemical mechanisms of sepal color development and variation in hydrangea. *Proceedings of the Japan Academy. Series B, Physical and Biological Sciences*, *97*(2), 51–68. https://doi.org/10.2183/pjab.97.003
- Zhao, Z., Zhang, Y., Meng, H., Li, W., & Wang, S. (2022). Identification and Optimization of a Novel Taxanes Extraction Process from Taxus cuspidata Needles by High-Intensity Pulsed Electric Field. *Molecules*, *27*(9), 3010–3010. https://doi.org/10.3390/molecules27093010