# An investigation of flavonoid diversity in *Scutellaria* to identify species with medicinal potential

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### Abstract

Scutellaria baicalensis is a medicinal plant whose root extracts have been widely used in Asian medicine for more than 2,000 years. Although *S. baicalensis* has been well-studied, the majority of the 470 species in the *Scutellaria* genus have not been analyzed. This study aims to extend the current knowledge of *Scutellaria* by analyzing flavonoid profiles and genome sizes of a large set of species representing the diversity in the genus. Targeted metabolite analysis via High Performance Liquid Chromatography (HPLC) was used to quantify concentrations of 15 medicinally relevant flavones. The genome sizes of eight of the species analyzed were also estimated with flow cytometry. A comparison of phytochemical profiles indicate diversity in site and identity of accumulation for multiple species when compared to *S. baicalensis*. Inconsistences in site of accumulation imply significant differences in biosynthetic pathway exists for several species. Researchers studying *Scutellaria* can use the results of this work to identify previously uncharacterized species with medicinal potential, aiding in the development of new drugs and treatments.

# Background

#### Scutellaria baicalensis is a medicinal plant

- Root extracts are used in traditional Chinese medicine
  - Huang Qin commercially sold powder of *S. baicalensis* roots

#### The medicinal properties of *S. baicalensis* are well-studied

- Proven medicinal effects: anti-Alzheimer's, anxiolytic, anti-cancer, and angiogenic
- Specific phytochemicals responsible have been identified
- Reference genome recently completed
- Proposed flavonoid biosynthetic pathway explains organ-specific accumulation

#### However, the Scutellaria genus contains >470 species of plants

- Little is known about the medicinal relevance of most of the genus
  - Phytochemical profiles are not well-studied/not studied at all
- Uncharacterized species could possibly accumulate medicinally valuable compounds at higher concentrations than species currently studied

**Goal:** Analyze a large set of *Scutellaria* plants representing the diversity in the genus to identify species with medicinal potential for further study

#### Objectives:

- I. Identify species of interest by quantifying medicinally relevant flavonoids in total (i.e. not organ-specific) samples
- II. Analyze organ-specific samples from species of interest to determine site of accumulation of medicinally relevant flavonoids
- III. Estimate genome size for species of interest to evaluate ease of further genetic study/improvement

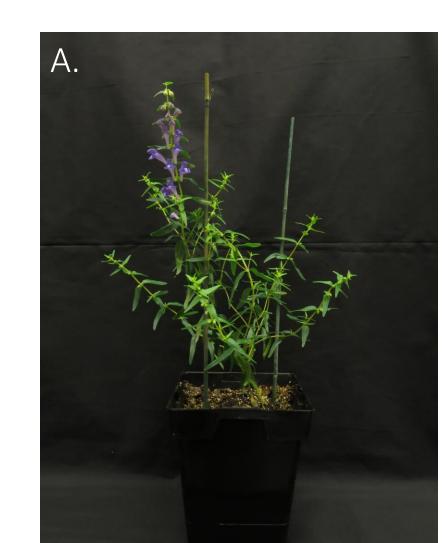
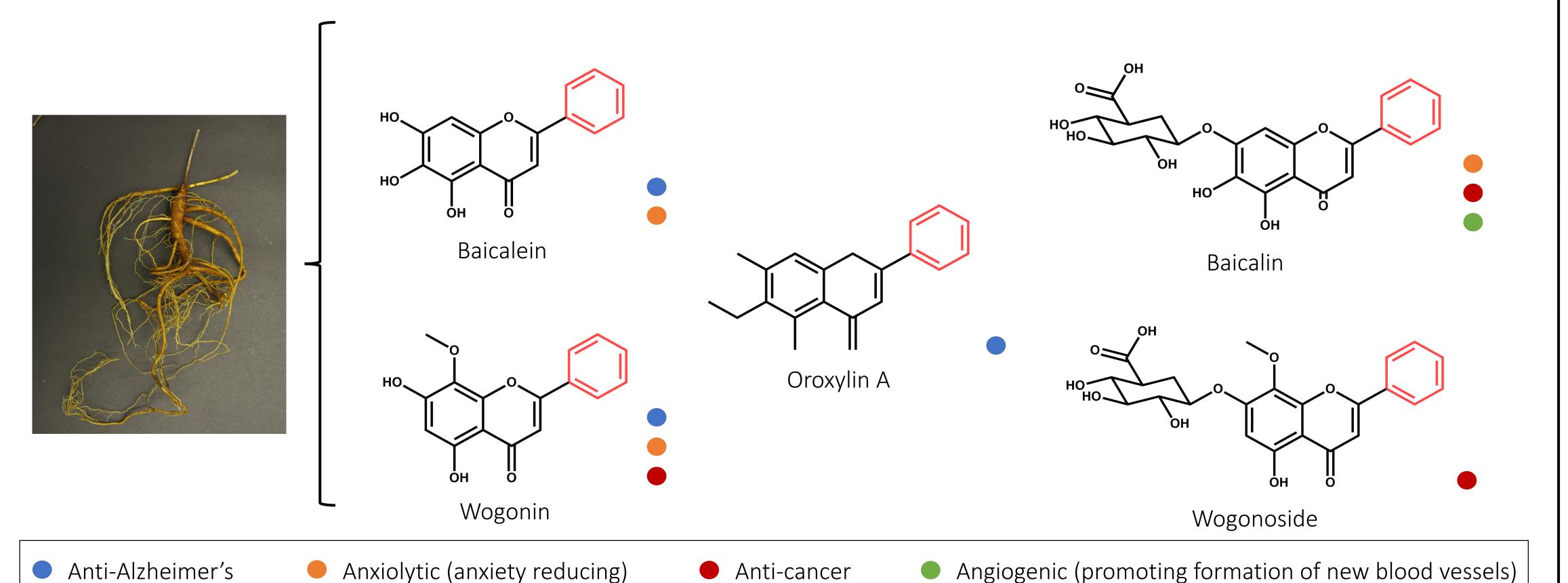






Figure 1. Morphological differences between (A) Scutellaria baicalensis, (B) S. lateriflora, and (C) S. racemosa plants illustrate the diversity of the Scutellaria genus.



Methods and Results

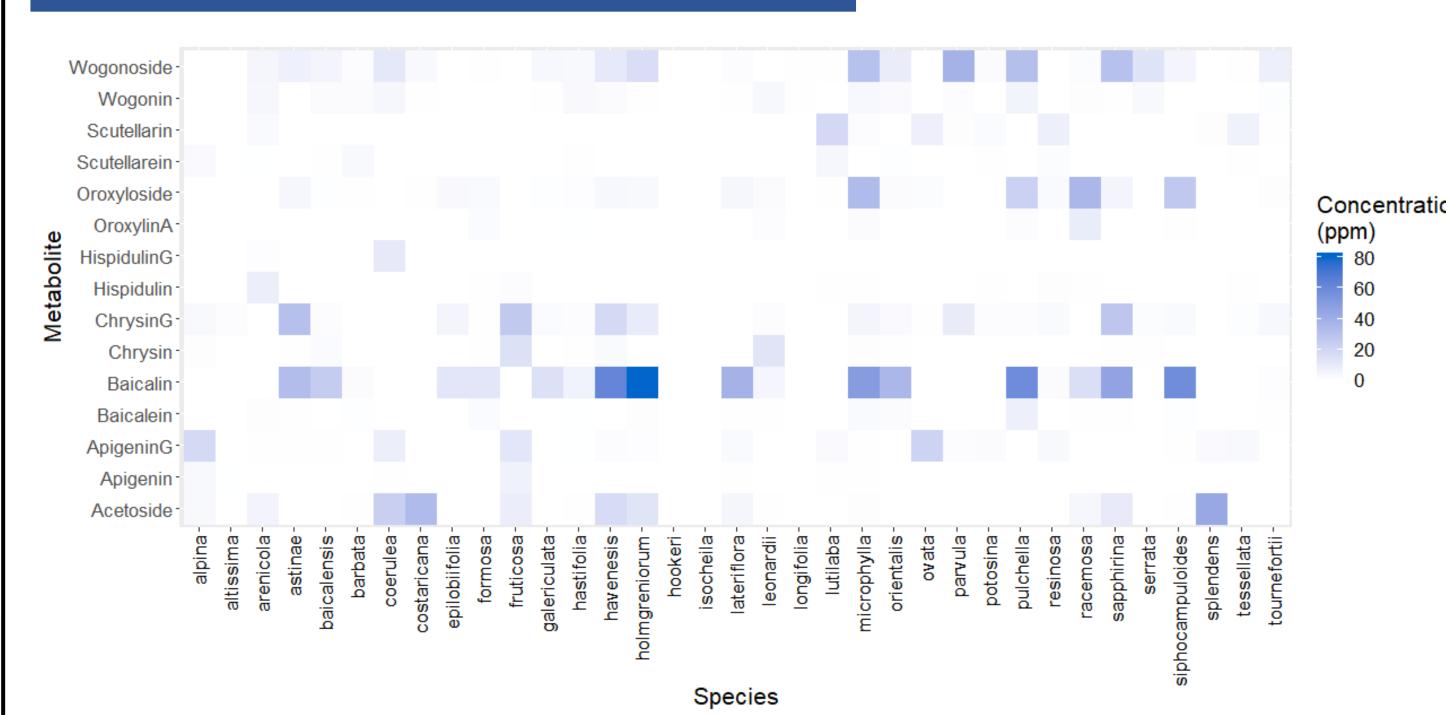


Figure 3. Heatmap of flavonoid concentrations for 34 *Scutellaria* species analyzed.

#### Chemical analysis of total samples

- Targeted metabolite analysis performed with High Performance Liquid Chromatography
  - Concentrations of 15 medicinally relevant flavonoids were quantified
- 34 species of *Scutellaria* were analyzed
- Dried (herbarium) samples prepared at 1000 ppm (1 mg sample / 1 mL solvent)
- Fresh samples prepared at 5000 ppm (5 mg sample / 1 mL solvent)

#### Organ-specific chemical analysis

- 6 species were selected for further analysis
- HPLC used to quantify flavonoids in different organs of plant leaves, shoots, and roots
- Differences in site of accumulation imply differences in flavonoid biosynthetic pathway

#### Estimation of genome size

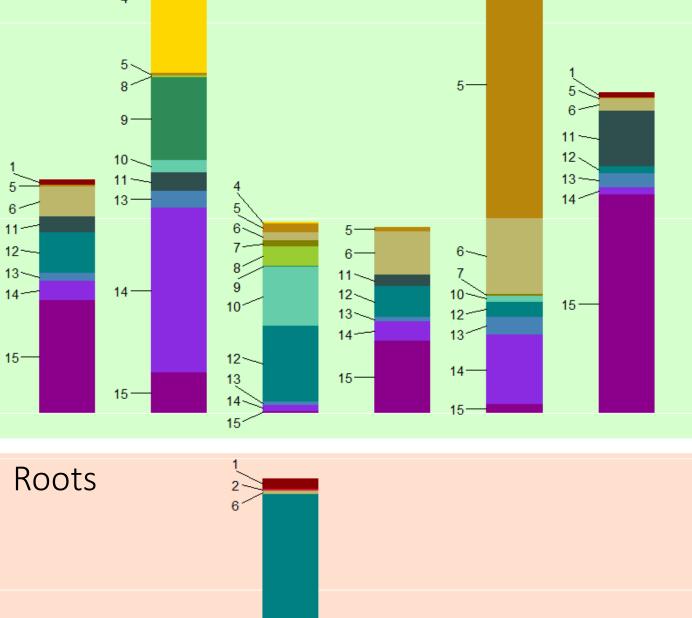
- Genome sizes of selected species were measured with flow cytometry
- 4 species have similarly sized genomes to *S. baicalensis*
- Allows comparison to *S.*baicalensis reference genome
- Overall genome sizes are small
- Ex: *Artemisia annua* (source of anti-malaria drugs) has a 1.74 GBP genome

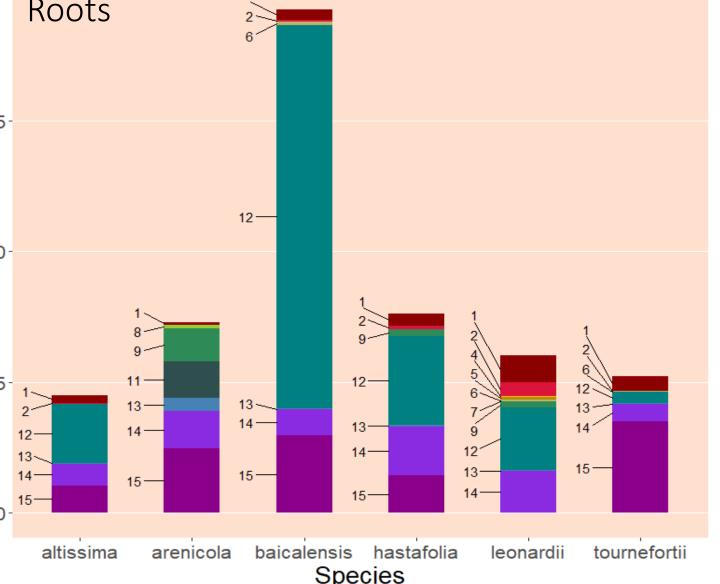
**Table 1.** Genome sizes for selected species.

Species	Standard plant	1C (Mean)	SD	Genome size (GBP)
altissima	Solanum	0.40	0.02	0.39
arenicola	Glycine	0.87	0.02	0.85
baicalensis	Solanum	0.55	0.00	0.54
hastifolia	Solanum	0.39	0.04	0.39
leonardii	Glycine	0.51	0.02	0.50
tournefortii	Solanum	0.40	0.01	0.39

Flavonoid

1. Oroxyloside
2. OroxylinA
3. Hispidulin
6. Chrysin
9. Acetoside
10. Scutellarein
11. Scutellarein
14. Wogonin
15. Wogonosid
15. Wogonosid
15. Wogonosid
16. Chrysin
17. Apigenin
18. Apigenin
18. Apigenin
19. Acetoside
10. Scutellarein
19. Acetoside
10. Scutellarein
11. Scutellarein
14. Wogonin
15. Wogonosid
15. Wogonosid
16. Chrysin
17. Apigenin
18. Apigenin
19. Acetoside
10. Scutellarein
19. Acetoside
10. Scutellarein
19. Acetoside
10. Scutellarein
10. Scutellarein
11. Scutellarein
11. Scutellarein
12. Baicalein
15. Wogonosid
15. Wogonosid
16. Chrysin
16. Chrysin
17. Apigenin
18. Apigenin
19. Acetoside
10. Scutellarein
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19. Acetoside
19





**Figure 4.** Results of organ-specific flavonoid analysis for selected species.

## Conclusions

A diverse range of flavonoid profiles were observed in the 34 species of *Suctellaria* studied. For the 6 species selected for organ-specific analysis, differences in site of accumulation suggest distinct biosynthetic gene expression between different species. Genome sizes for 4 of the species analyzed are similar to that of *S. baicalensis*, indicating that the published reference genome for *S. baicalensis* can potentially be used in comparative genomic and transcriptomic studies with these species. Future work will include analysis of an even larger set of *Scutellaria* species, phylogenetic study to identify clades of plants with medicinal potential, and transcriptomics to pinpoint genes involved in the biosynthetic pathway.

# References and Acknowledgements

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