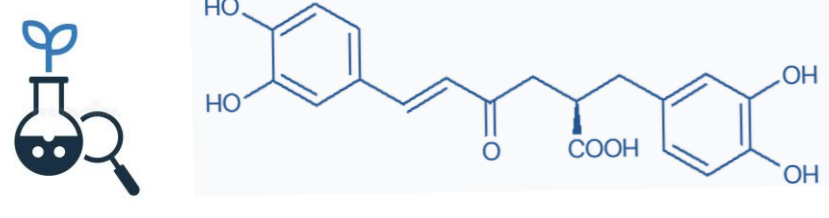


# Comparative study of rosmarinic acid content in methanolic extracts of Bulgarian *Thymus* species, section *Hyphodromi*



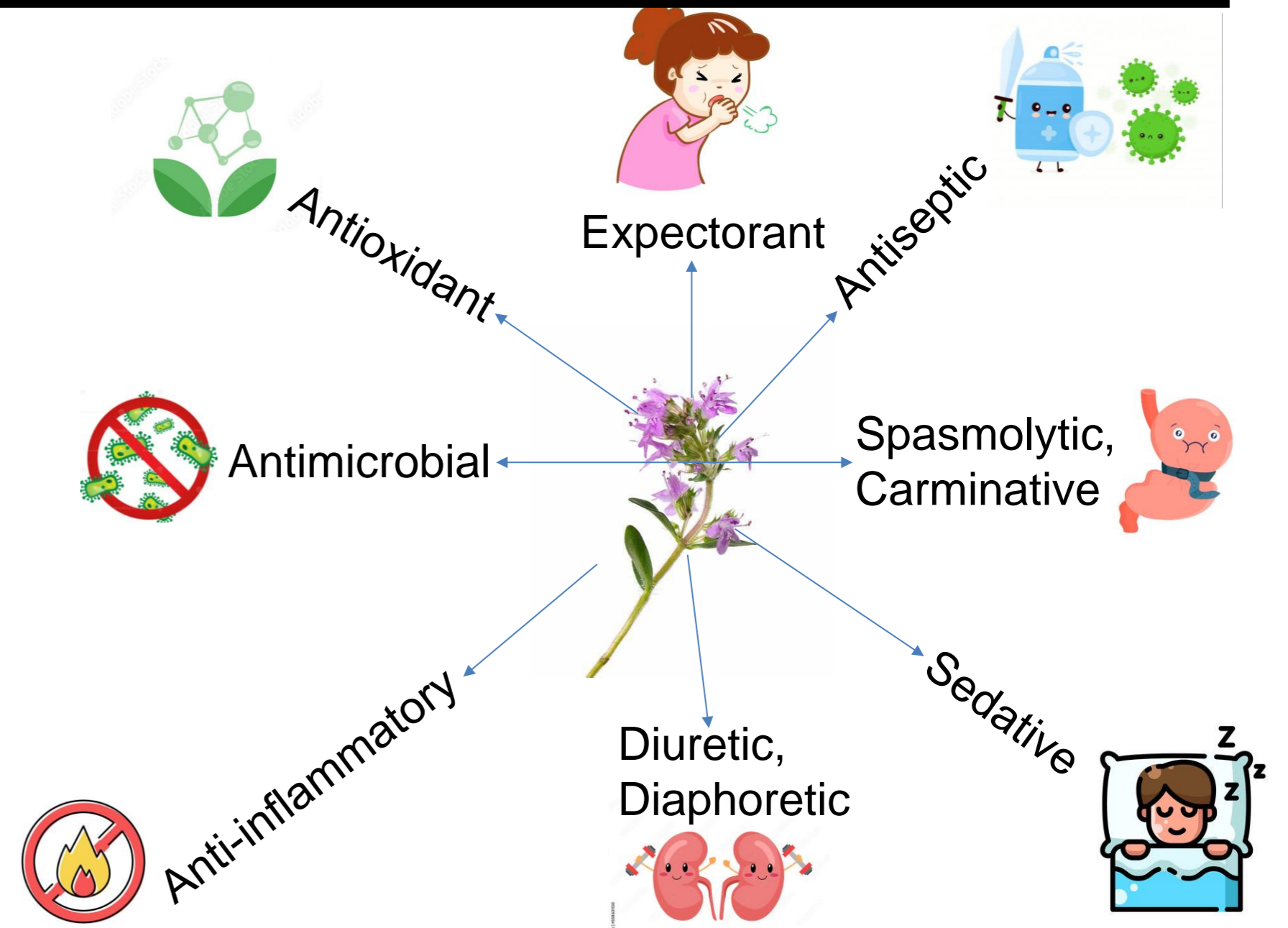
Denitsa Kancheva, Milena Nikolova, Ina Aneva

Department of Plant and Fungal Diversity and Resources, Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia, Bulgaria  
kanchevadenitsa@gmail.com

## INTRODUCTION & AIM

The genus *Thymus*, belonging to the *Lamiaceae* family, comprises around 250 aromatic species, widespread across Eurasia and North Africa. Twenty-one representatives occur naturally in Bulgaria, with eight belonging to the section *Hyphodromi*: two are local endemics: *T. jalasianus* Stoyanov & Marinov. and *T. perinicus* (Velen.) Jalas; *T. aznavourii* Velen. and *T. comptus* Friv. are Balkan endemics, while *T. atticus* Čelak is sub-endemic; *T. leucotrichus* Halácsy, *T. striatus* Vahl., and *T. zygioides* Griseb., are widespread [1].

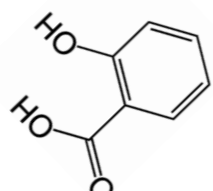
Pharmacological activities:



Thymes are highly valued medicinal plants. Their diverse biological activities are mostly related to the content of essential oils and phenolic compounds.

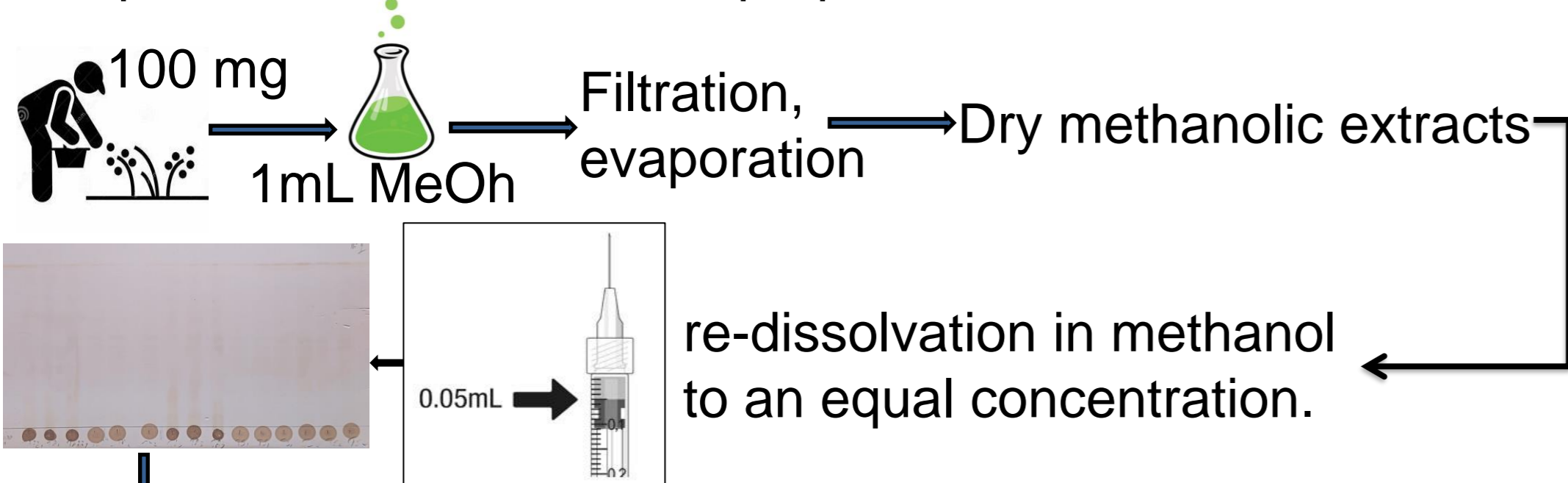
Due to the species' limited distribution and the overall complexity of the genus, extensive studies are lacking. Rosmarinic acid is a phenolic compound with highly valued biological activities, including antioxidant, anti-inflammatory, and antimicrobial properties. Reports identify many thyme species as sources of this polyphenol, highlighting their potential beneficial properties [2].

This study aimed to estimate the rosmarinic acid content by a quick, accurate, and readily available method such as HPTLC in the methanolic extract of the *Thymus* species section *Hyphodromi* distributed in Bulgaria.



## METHOD

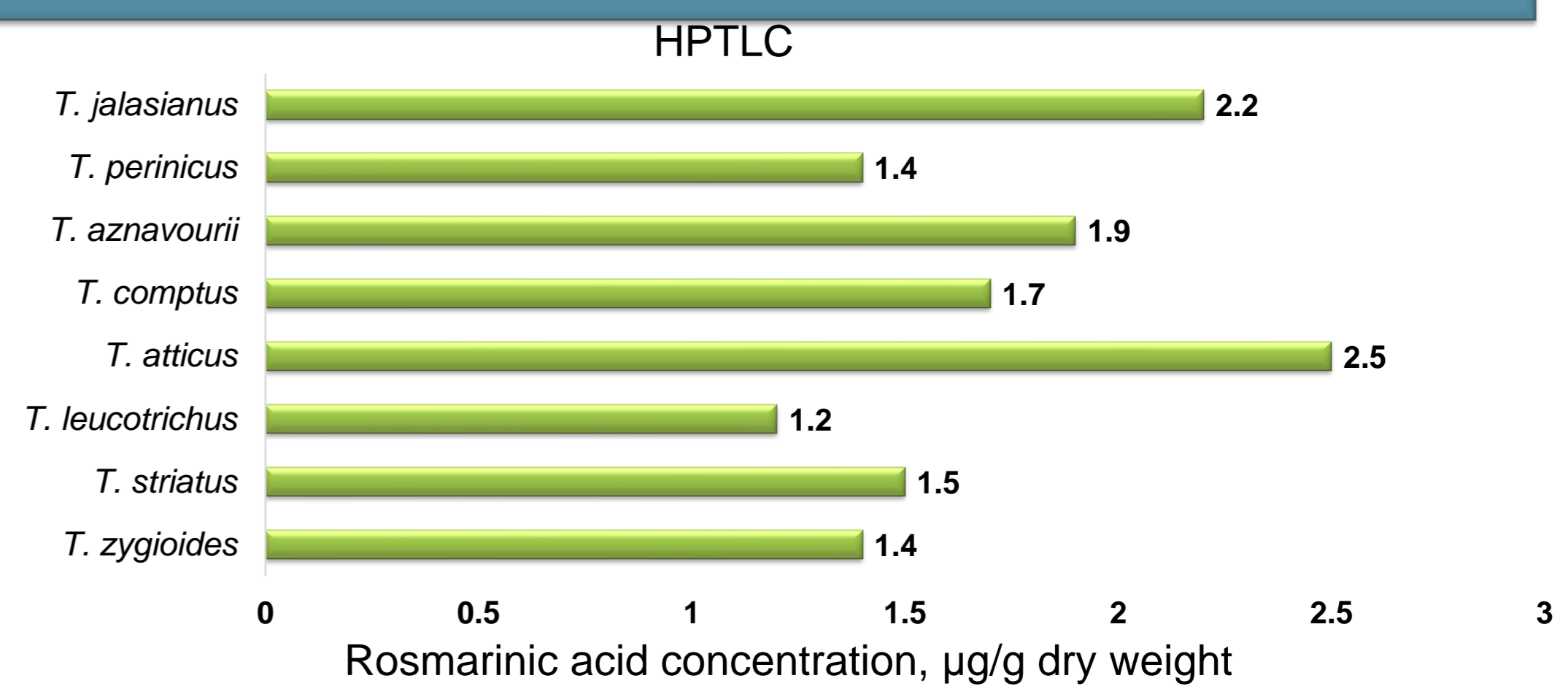
Sample collection and extract preparation:



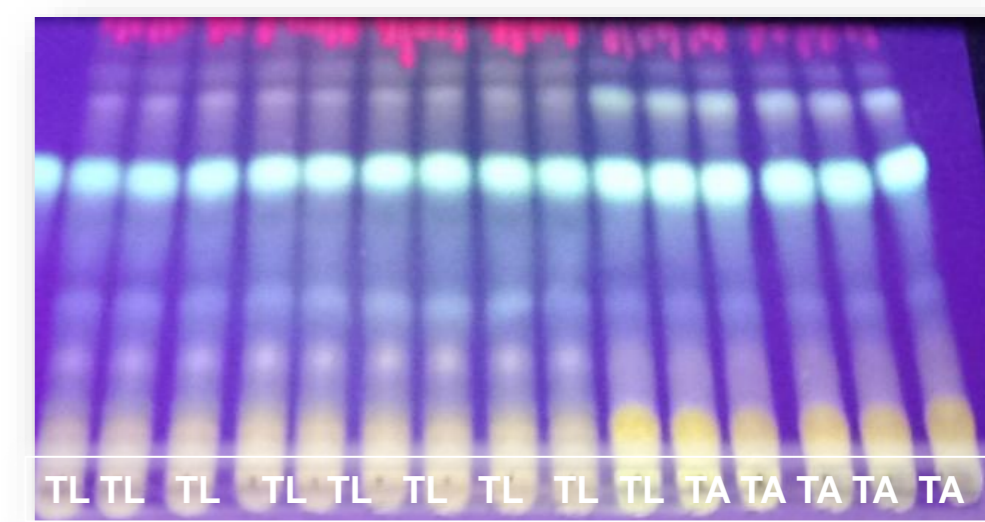
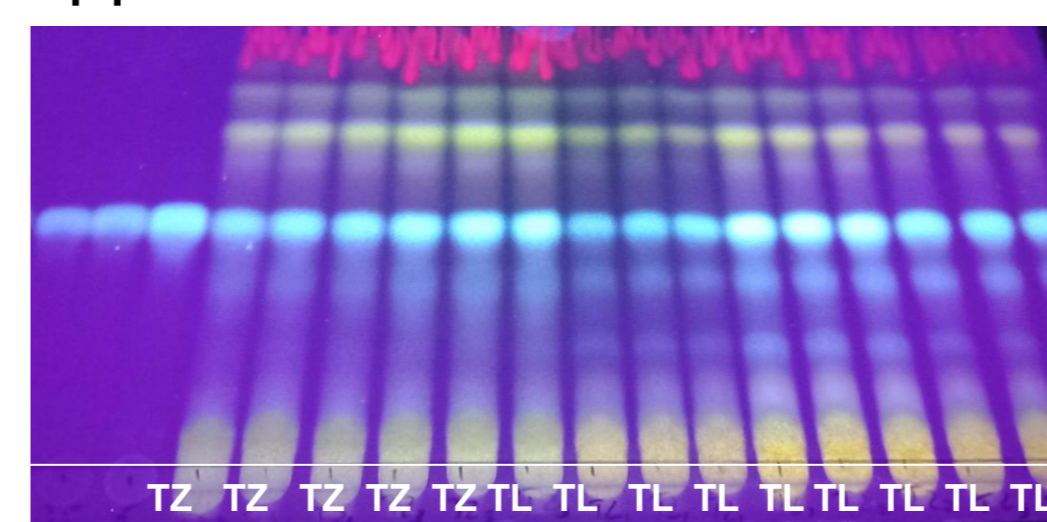
Chromatographic condition: stationary phase: HPTLC silica gel 60 F254 plates. Mobile phase: chloroform/ethyl acetate/formic acid (50:40:10). The compounds were visualized by spraying with 'Naturstoffreagenz A', a 1% solution of diphenylboric acid ethanolamine complex in methanol.

The fluorescence emission of rosmarinic acid was recorded under UV radiation at 336 nm, using a digital camera. The images were analyzed by QuantiScan 2.1 Biosoft software.

## RESULTS & DISCUSSION



Rosmarinic acid was present across all extracts, from 1.20 to 2.50 µg/g of dry weight. Among the species, *Thymus atticus* and *T. jalasianus* exhibited significant abundance, while *T. comptus*, *T. aznavourii*, and *T. striatus* showed moderate quantity levels. The tested species are sources of valuable polyphenols, with potential applications in various industries.



Figures 1&2. TLC plates for rosmarinic acid  
TZ – *T. zygioides*, TL – *T. leucotrichus*, TA – *T. atticus*

## CONCLUSION

The obtained data showed *Thymus atticus* and *T. jalasianus* as promising for cultivation when plant material with a high rosmarinic acid content is needed.

## FUTURE WORK / REFERENCES

The initial stages of cultivating the endangered and economically valuable *Thymus* species have begun. The following phase involves an investigation of the reproductive biology of these plants, which is essential for developing strategies that advocate for their sustainable use and long-term conservation.

References:

- Aneva I et al., 2022. In *Plants* <https://doi.org/10.3390/plants11030270>.
- Afonso A. et al., 2017. In *Int J Mol Sci.* <https://doi.org/10.3390/ijms18091879>.