

## De Novo Transcriptome Sequencing of Ramonda serbica: Identification of the Candidate Genes Involved in the Desiccation Tolerance

# <u>Vidovic M</u><sup>1</sup>, Stevanovic S<sup>1</sup>, Pantelic A<sup>2</sup>, Veljovic-Jovanovic S<sup>3</sup>



1) Institute of Molecular Genetics and Genetic Engineering, Laboratory for Plant Molecular Biology, University of Belgrade, Serbia

- 2) University of Belgrade, Faculty of Chemistry, Studentski trg 12-16, 11000 Belgrade, Serbia
- 3) Institute for Multidisciplinary Research, University of Belgrade, Belgrade, Serbia



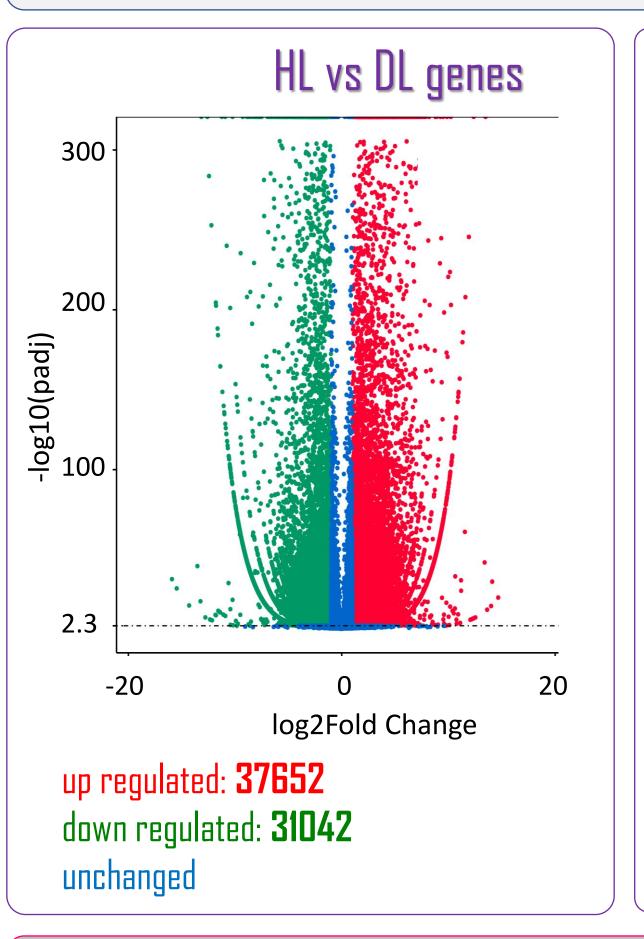
Introduction: Ramonda serbica Panc. is a resurrection plant that can survive a long period of severe dehydrationdesiccation. Desiccation induces cellular membrane integrity loss, protein aggregation, and denaturation, as well as accelerated generation of reactive oxygen species. However, R. serbica can fully recover its metabolic functions already one day upon watering (1).

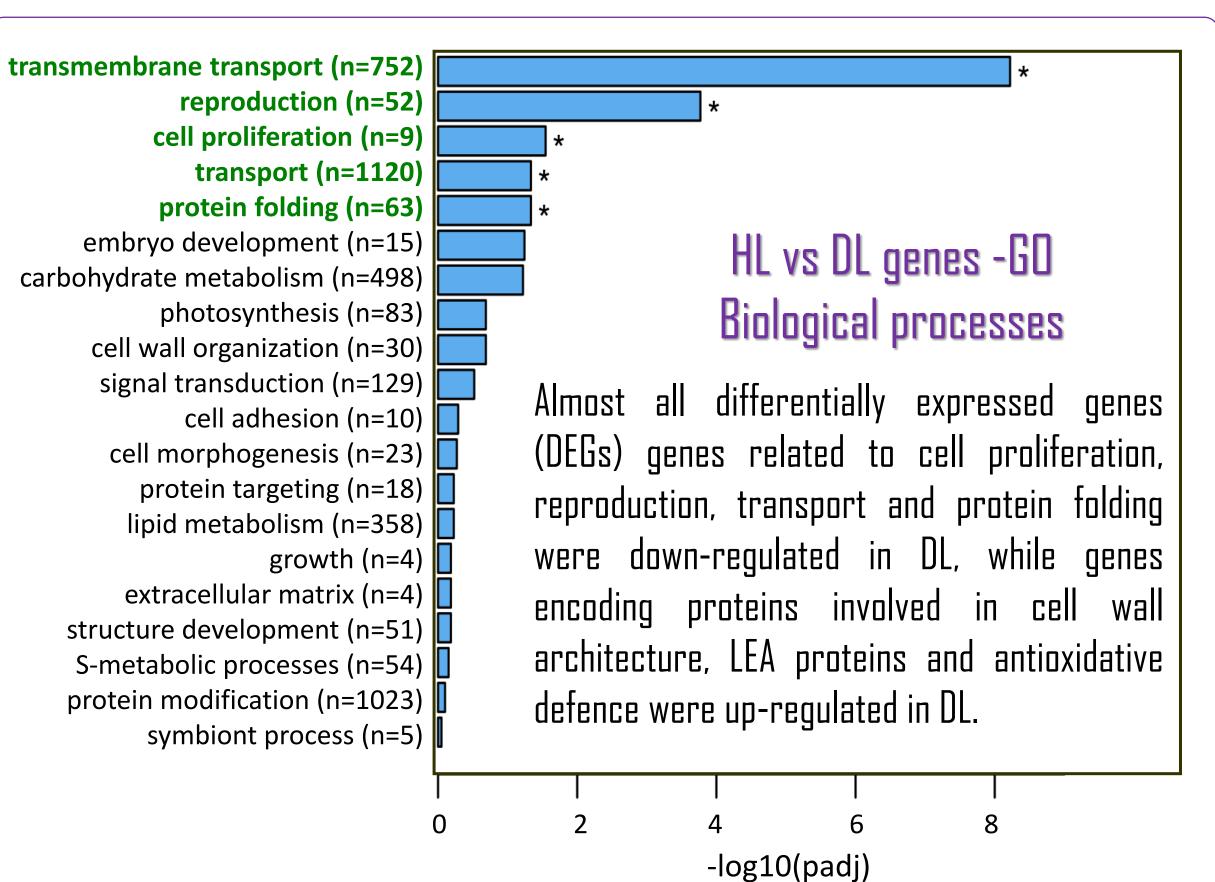
Aim: to obtain more insight into the mechanisms of desiccation tolerance in R. serbica by differential de novo transcriptomics of hydrated (HL) and desiccated leaves (DL).

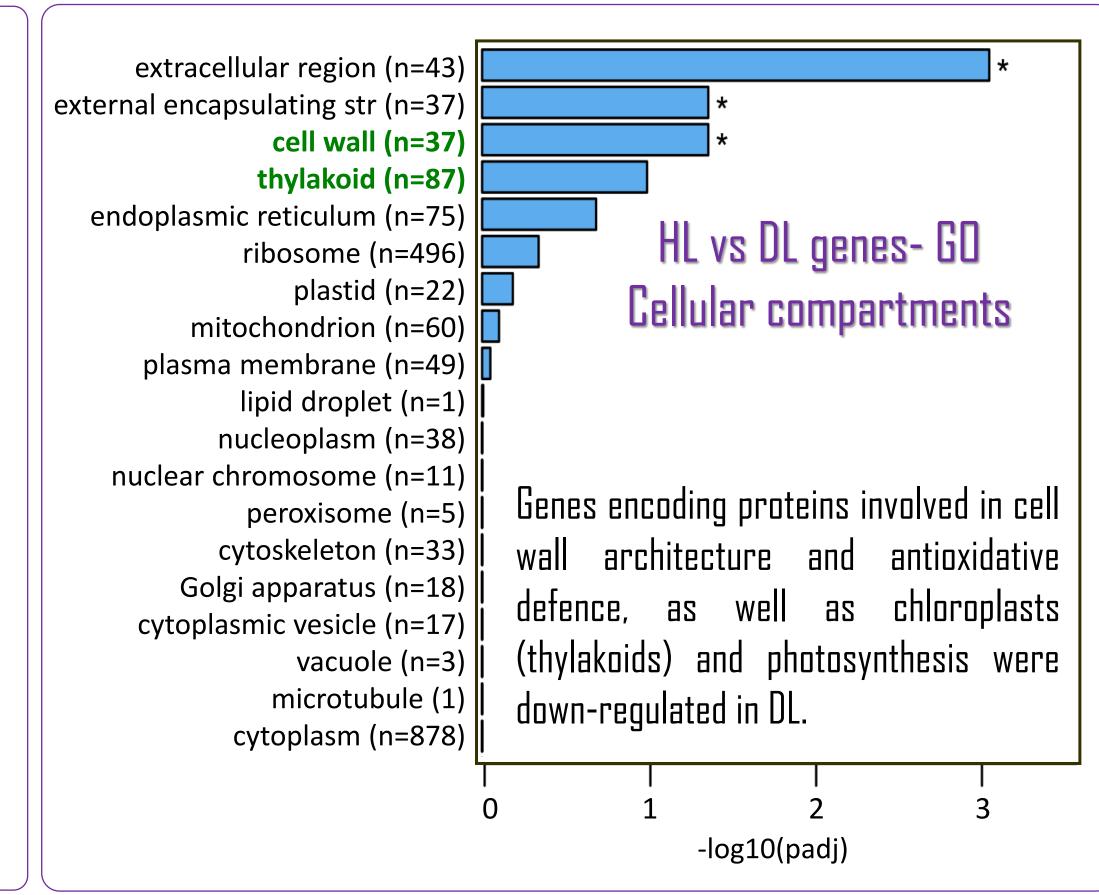


#### Results:

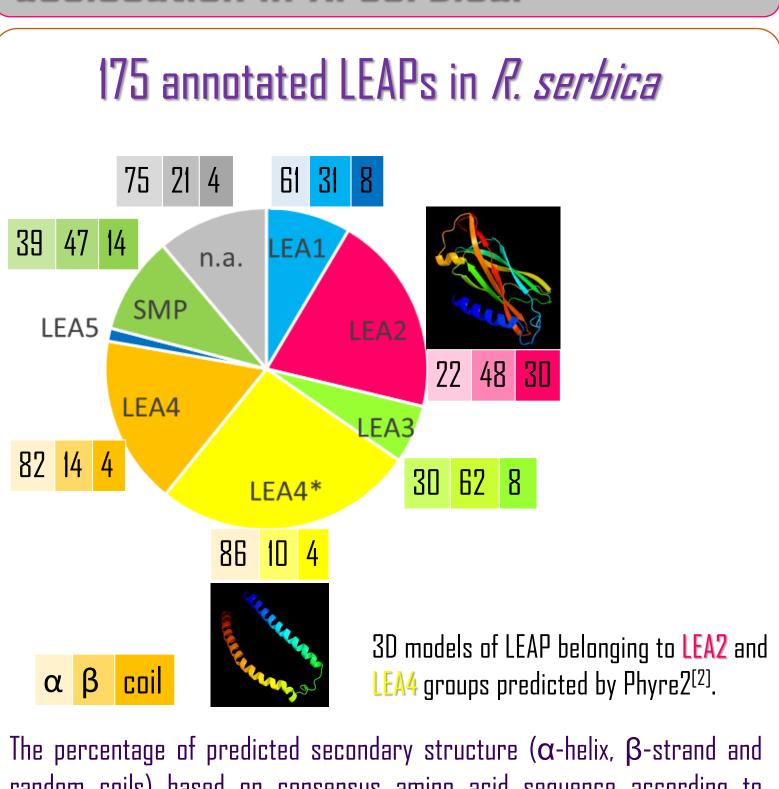
After de novo transcriptome analysis, 189456 transcripts (32.6% with the length between 500-1kbp) with 189003 unigenes were annotated with seven common databases. Among obtained unigenes, 64.6% and 42.3% were annotated by NCBI non-redundant protein and nucleotide sequences database (db), 23% by PFAM db, 22.5% by Clusters of Orthologous Groups of proteins db, 48.02% by Swiss-Prot db, 23 % KEGG db and 13.73 by Gene Ontology db.

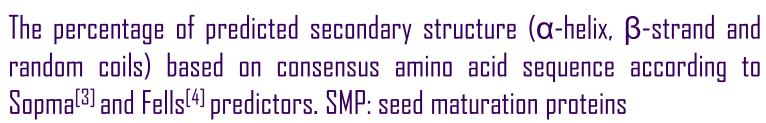


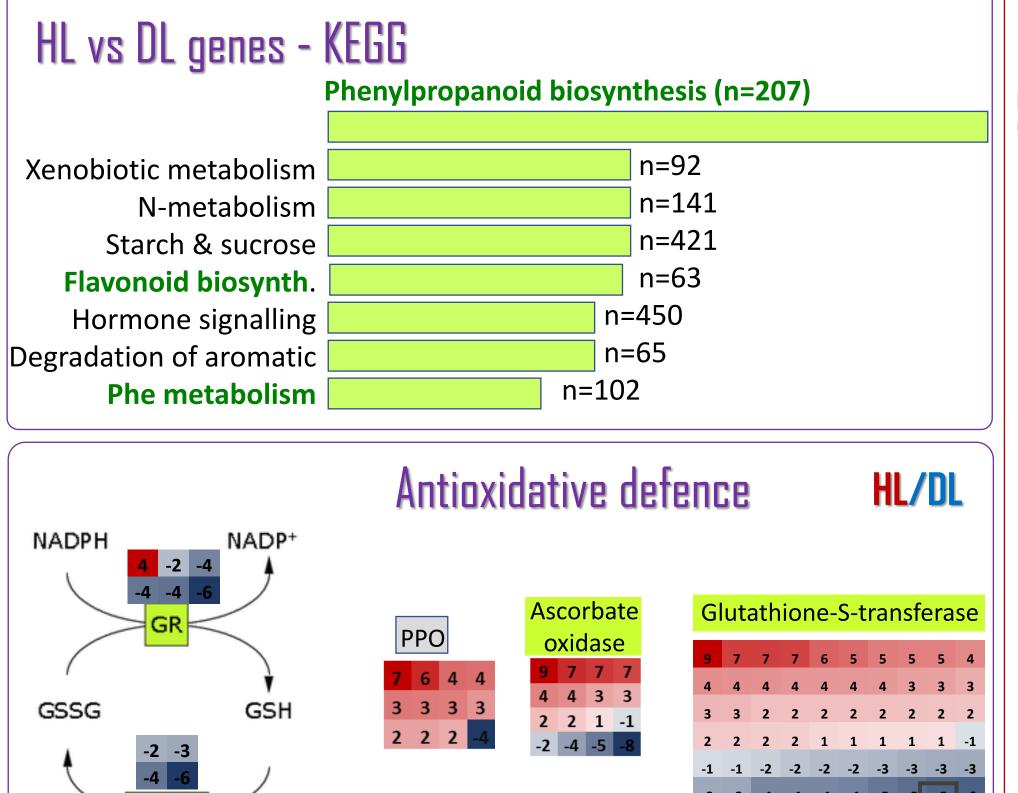


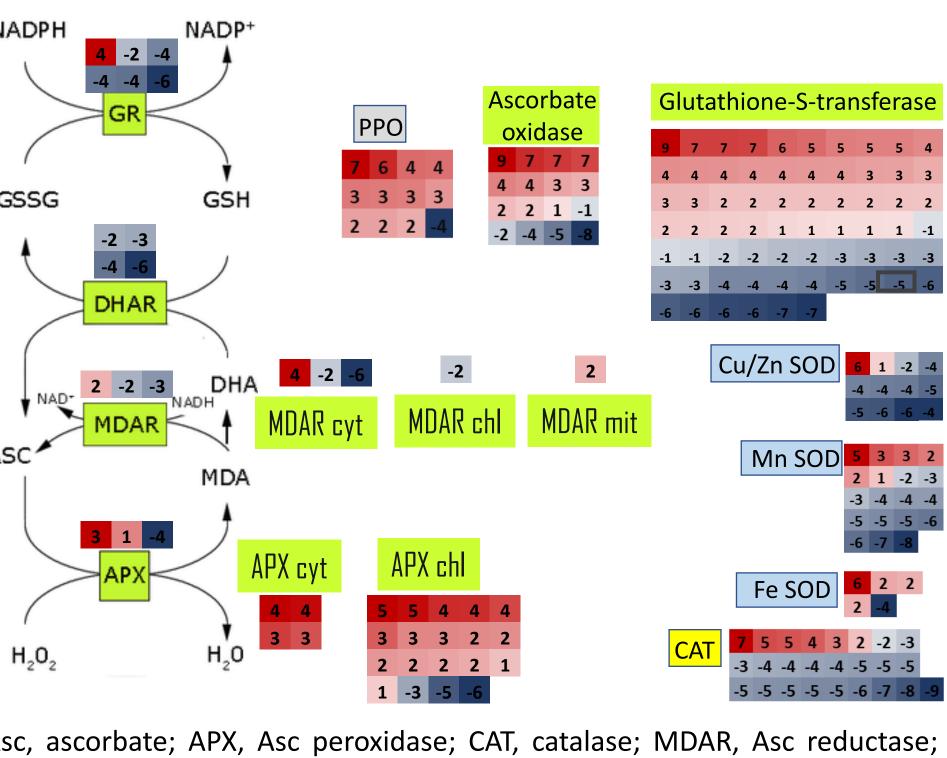


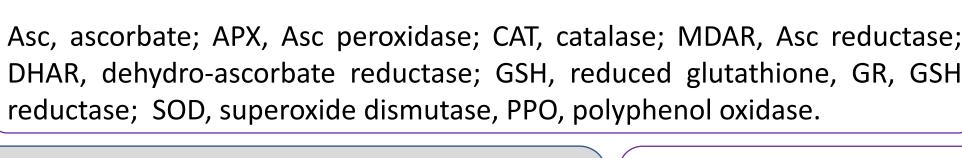
Our results imply an important role cell wall structural proteins, GLPs, late embryogenesis abundant polyphenol (LEAPs), oxidases (PPOs), and Cu/Zn SODs protective mechanism against desiccation in *R. serbica*.

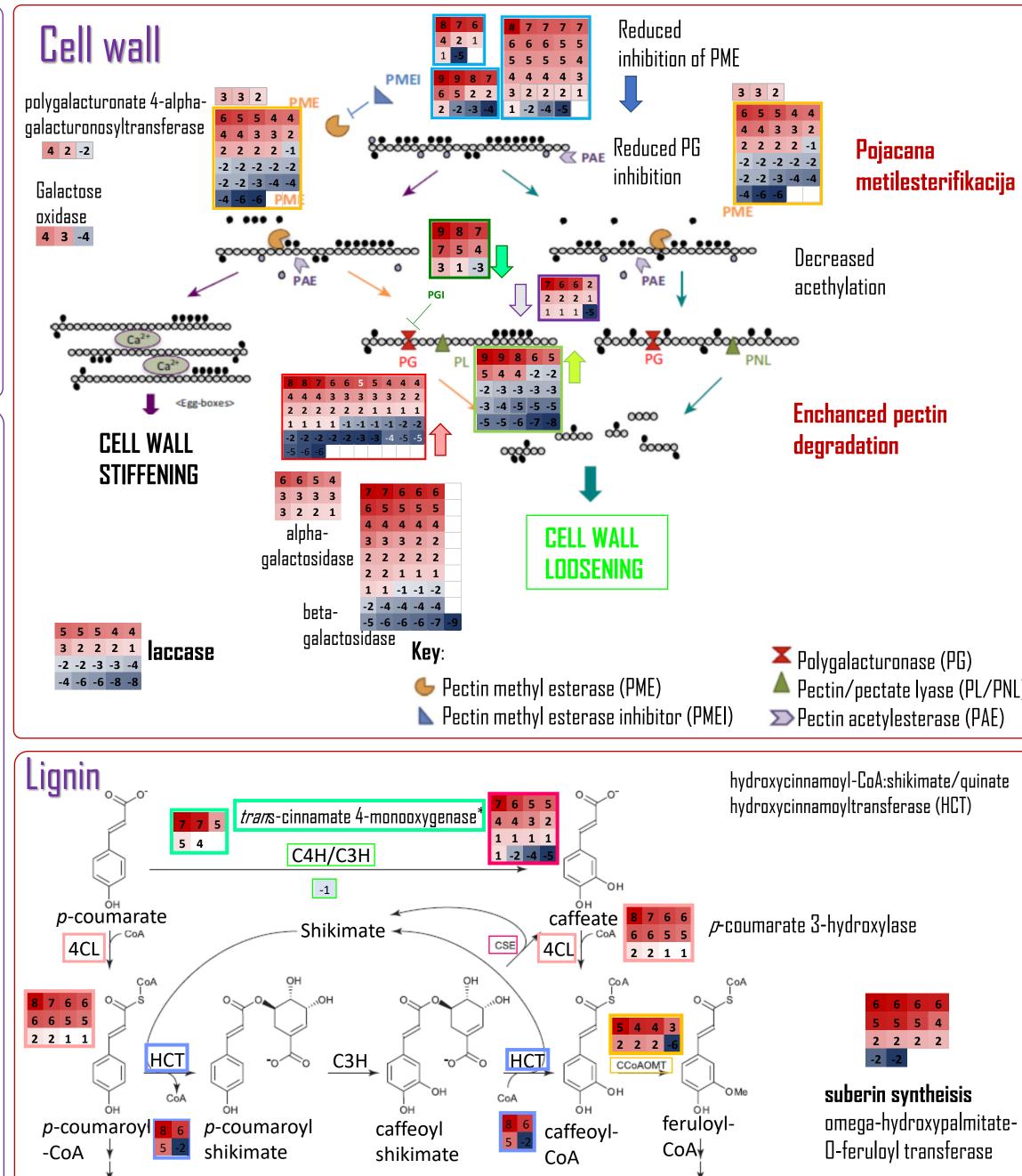












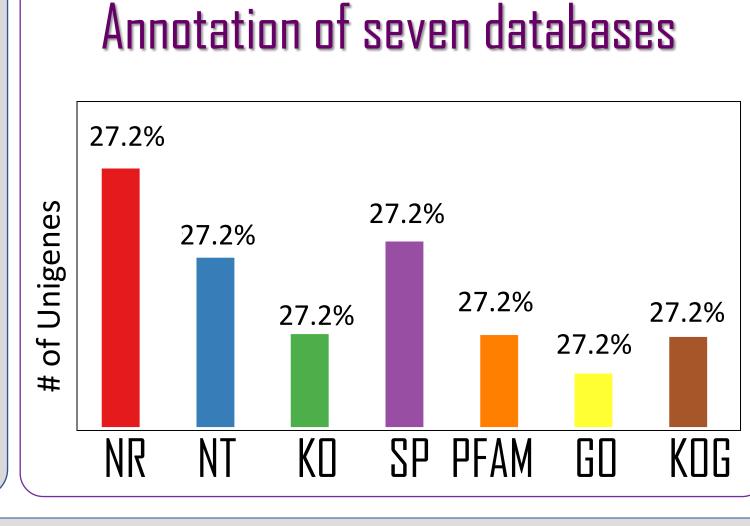
### Workflow (wet lab)

TRIzol-based extraction of high-quality RNA from *Ramonda serbica* [1]

Quality control and quantification [1]

Synthesis and sequencing of cDNA libraries [1]

## Transcriptome reconstruction <sup>[5]</sup> Annotation [6] Quantification [7] Differential Expression Analysis [8] Enrichment Analysis [9]



H-unit

#### References:

(9) GO Seq, v. 1.32.0, topGO

[1] Vidović M, Ćuković K. 2020. 3 Biotech. 10(6):286. (2) Kelley et al. 2015 Nature Protocols 10: 845-58. (3) Combet et al. 2000. Trends Biochem Sci 291: 147-150 [4] Piovesan et al. 2017 Bioinformatics 33: 1889-91. [5] Trinity (Inchworm, Chrysalis, Butterfly); v. r20140413p1. min\_kmer\_cov=2, min\_glue=2; k-mer=25 [6] NR, NT, Swiss-Prot, KOG/COG, KEGG, Pfam, GO [7] Bowtie2-2.2.2, mismatch=0, RSEM-v1.3.0 [8] DESeq2, v. 1.10.1 Normalzation method DESeq2, BH FDR estimation method, negative binomial distribution, padjust<0.05

G and S-units

Ministry of Education, Science and Technological Development, RS (451-03-9/2021-14/200042).

Acknowledgments

Ministry of Education, Science and Technological Development, RS; Bilateral project (no. 451-03-01963/2017-09/09)





## Science Fund of the Republic of Serbia

Program for Excellent Projects of Young Researchers LEAPSyn-SCI #6039663