STUDY OF THE ROOT TRANSCRIPTOME OF BREAD WHEAT USING HIGH-THROUGHPUT RNA SEQUENCING (RNA-SEQ)

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MOTIVATION AND AIM

- Bread wheat (*Triticum aestivum* L.) is the most important crop in the world. It provides about 20% of the total calories consumed by humans. For a long time, wheat selection was mainly based on phenotypic traits of the shoot, while the roots were given little attention. As a result, the root system of modern wheat varieties has weakened. Therefore, the study of genetic control of wheat roots development is an urgent issue. A convenient method to study the genetics of roots development is transcriptome profiling using high- throughput RNA sequencing (RNA-seq). The method allows us to evaluate gene expression across the entire genome, as well as to find specific genes responsible for roots development that will be used in the future during marker-assisted selection of wheat varieties with resistant root system.
- The aim of the current study is revealing differentially expressed genes between the root and shoot transcriptomes of allohexaploid wheat using RNA-seq analysis and identifying root-specific genes involved in the wheat root system development.

Plant material and RNA sequencing

RNA was extracted from 4-days roots and coleoptiles of seedlings of Russian spring wheat cultivar "Saratovskaya 29" in three biological replicas. One sample contained roots or coleoptile from six different plants. Sequencing was performed using Illumina NextSeq 550 platform in Institute of Cytology and Genetics SB RAS, 75-bp reads was obtained.

De novo assembly of transcriptome was performed using Trinity software. To evaluate quality of obtained transcriptome Transrate software was used. Quantification of reads was performed using Salmon software. To obtain different expressed transcripts edgeR package was used. For functional annotation Transrate, AgriGO and BlastKOALA services were used.

Data quality assessment

1.0

0.5

0.0

-0.5

-1.0

-1.5

C2_S2

-4

-2

0

Leading logFC dim 2

C3_S3

C1_S1

sample correlation matrix y_trans.isoform.TMM.EXPR.matrix.log

Color Key

0.8 Value

C2 S2

C1_S1

R2_S5

R1_S4

S6

R3_

C3_S3

Pipeline



R32S6

R1_S4

2

C1_S2, C2_S2, C3_S3 – coleoptile libraries, R1_S4, R2_S5, R3_S6 – root libraries

C2_S2

C1_S1

C3_S3

R2 S5

R1 S4

R3 S6

Transcriptome *de novo* assembly metrics

	Transcript number	Up- regulated in root	Down- regulated in root	Root- specific
Total	330,112	31,488	35,851	18,040
With ORF	112,934	20,793	21,094	7,216

DEG functional annotation (Agri GO)

	GO term	DEGs amount	FDR
Up- regulated	response to oxidative stress	287	3.8e-50
	oxidation-reduction process	1137	4.7e-33
	single-organism metabolic process	1712	1.1e-28
	response to stress	465	1.7e-21
Down- regulated	single-organism metabolic process	1612	1.3e-51
	single-organism process	2040	8.6e-43
	oxidation-reduction process	974	3e-31
	metabolicprocess	3914	5.9e-27

DEG functional annotation (KEGG)

Down-regulated genes



Up-regulated genes



Root-specific genes

Protein families: genetic information process Biosynthesis of other secondary metabolites Organismal Systems Carbohydrate metabolism Protein families: signaling and cellular proce Protein families: metabolism Genetic Information Processing Environmental Information Processing Lipid metabolism Unclassified: metabolism Metabolism of other amino acids