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# Содержание

## МАТЕРИАЛЫ VII МЕЖДУНАРОДНОЙ НАУЧНО-ПРАКТИЧЕСКОЙ КОНФЕРЕНЦИИ «БИОТЕХНОЛОГИЯ: НАУКА И ПРАКТИКА»

## СЕКЦИЯ 1. ПИЩЕВАЯ, СЕЛЬСКОХОЗЯЙСТВЕННАЯ И ЛЕСНАЯ БИОТЕХНОЛОГИЯ

## СЕКЦИЯ 4. БИОИНФОРМАТИКА И IT

Binding sites of 11 miRNAs have been found in 5'UTR mRNAs of 13 candidate genes: *ABCC1, ADNP, ATP11B, ACADM, ARID3A, ASXL1, ARID1A, ARHGAP17, ASH1L, ASTN2, ARRB1, ANKRD11* and *AMFR*. The mRNAs of *ARL4C, ADARB1, ATXN7L1, ACSL6, AKT2, AGPAT6* and *APBB1IP* genes have ten miRNAs binding sites in 3'UTR.

Only three genes of the 20 candidate genes with a free interaction energy greater than -120 kJ/mol predicted as targets of two or more miRNAs: *APBB1IP* (miR-4763–3p, miR-3960, miR-328–5p); *ASTN2* (miR-4763–3p, miR-4707–5p); *AGPAT6* (miR-1273d, miR-1226–5p). Among miRNAs that bind with high energy to mRNA genes with nucleotide repeats, we choose four miRNAs that have binding sites in mRNAs of two or more genes: miR-619–5p *(ACSL6, AKT2*); miR-6089 (*ACADM, ARID3A, ASXL1*); miR-4787–5p (*AGPAT6, ARHGAP17, ASH1L*); miR-4763–3p (*APBB1IP, ASTN2*).

The mRNAs of *AGPAT6, ARHGAP17* and *ASH1L* genes having nucleotide repeats (GGAGUU**GCCGCCGCCGCC**CCCAC)$_{GCC4}$, (GGG**CCGCCGCCG**CCA**CCGCCG**C)$_{CCG5}$ and (GGAGCU**GCCGCCGCCACCGCC**GC)$_{GCC4}$ in 3’ and 5’ UTRs are a targets of miR-4787–5p. The obtained results of the mRNA of *ASXL1* and *ARID1A* genes indicated that the miR-6089 and miR-6081 bind with (**CCGCCGCCG**CCCCUCCCCCACCGCC)$_{CCG3}$/ (GGCGCCUCG**GCCGCCGCCGCCGCC**U)$_{GCC5}$ repeats in 5’ UTR. miR-3960 and miR-1908–3p bind with mRNA of *APBB1IP* and *ARRB1* genes in sites with (CCC**CCGCCGCCGCCGCCG**CC)$_{CCG5}$ and (**CGGGGGCGGCGGCGGCGG**CCGG)$_{CGG5}$ repeats in 3’ UTR and in 5’UTR, respectively.

mRNAs of non-polyglutamine disorders genes (*ATXN10, AFF2, CNBP, DMPK, FMR1, FXN, JPH3* and *PPP2R2B*) have 202 miRNAs binding sites in 5'UTR and 3'UTR. The ΔG/ΔGm values equal to 87 %. *AFF2* and *FMR1* mRNAs bind with miR-3960 and miR-4258 in regions with nucleotide repeat sequences. miR-3960 and miR-4258 bind with (**GCCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCC**)$_{GCC15}$ and (**CGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGGCGG**)$_{CGG19}$ repeats with ΔG/ΔGm values equal or more than 85 % in 5’UTR.

Thus, miRNAs binding sites with mRNA genes of having nucleotide repeats have been predicted. Studying the binding characteristics of miRNA and genes will help identify associations of miRNAs with genes with nucleotide repeats for recommending for the diagnosis of nucleotide repeat expansion disorders.

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# CHARACTERISTICS OF INTERACTION OF MIRNAS WITH MRNA GENES OF HEAT STRESS TRANSCRIPTION FACTORS

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Heat stress transcription factors (HSF) play a key role in plants response to several abiotic stresses by controlling the expression of stress-responsive genes, such as heat shock proteins [1, 2]. In recent years, the important role of miRNAs in regulating the expression of many plant genes has been established [3, 4]. However limited studies has been reported for the association of miRNA with the expression of plant HSF genes. In the present work, the task is to determine the characteristics of the interaction of miRNA with the mRNA genes of the HSF family Arabidopsis thaliana, Oryza sativa, Triticum aestivum, Zea mays. The nucleotide mRNA sequences of the HSF family were taken from Plant Transcription Factor Database v4.0 (http://planttfdb.cbi.pku.edu.cn/index.php; https://www.ncbi.nlm.nih.gov/). miRNAs were borrowed from miRBase v. 22 (http://www.mirbase.org/). The search for miRNA target genes was carried out in the MirTarget program [5, 6]. This program defines the start of miRNA binding sites with mRNA; the localization of miRNA binding sites in the 5’ – untranslated region (5’UTR), the protein-coding region (CDS) and the 3’ – untranslated region (3’UTR) of mRNA; the free energy of hybridization (ΔG, kJ/mole) and schemes of nucleotide interactions between miRNA and mRNA. The miRNA binding sites were selected to have the ΔG/ΔGm ratio of more than 85 %, taking into account the ΔG value and miRNA length.

Table 1 shows the characteristics of the miRNA binding sites in the mRNA of the HSF genes of *A. thaliana, O. sativa, T. aestivum, Z. mays. A. thaliana* is a model plant organism for modern molecular research and we found binding sites between miRNA and mRNA of their target genes. The binding of 428 ath-miRNAs to mRNA of 24 HSF genes of *A. thaliana* was studied. Only 14 genes were targets for 18 miRNAs. miR5021–5p and miR5658–5p had three target genes with a ΔG/ΔGm value from 85 % to 91 %. Two miRNAs of the miR172c, d-3p family, were bound with mRNA of the *AT5G03720.1* gene. The remaining miRNAs had only one target genes. The miRNA binding sites were located in 5'UTR, CDS and 3'UTR mRNA HSF genes of *A. thaliana*.

Table 1 – Characteristics of miRNA binding sites in mRNA of *A. thaliana, O. sativa, T. aestivum, Z. mays* HSF genes

| Gene | miRNA | Start of site, nt | Region | ΔG, kJ/mole | ΔG/ΔGm, % | Length, nt |
|---|---|---|---|---|---|---|
| Arabidopsis thaliana | | | | | | |
| AT4G11660.1 | miR5021–5p | 215 | 5'UTR | -89 | 91 | 20 |
| AT4G18880.1 | miR415–5p | 752 | 5'UTR | -96 | 90 | 21 |
| AT4G17750.1 | miR5631–3p | 728 | CDS | -93 | 90 | 21 |
| AT4G18870.1 | miR827–3p | 877 | CDS | -93 | 90 | 21 |
| AT1G46264.1 | miR834–3p | 653 | CDS | -100 | 89 | 21 |
| AT5G16820.1 | miR4245–3p | 591 | CDS | -87 | 87 | 21 |
| AT1G67970.1 | miR5021–5p | 227÷230 | CDS | -85 | 87 | 20 |
| AT3G63350.1 | miR5021–5p | 590 | CDS | -85 | 87 | 20 |
| AT5G03720.1 | miR5658–5p | 1067 | CDS | -89 | 87 | 21 |
| *Oryza sativa* | | | | | | |
| LOC_Os06g35960.1 | miR5075–3p | 547 | CDS | -113 | 91 | 21 |
| LOC_Os06g36930.1 | miR5077–5p | 486 | CDS | -96 | 90 | 19 |
| LOC_Os10g28340.1 | miR529a-3p | 47 | 5'UTR | -96 | 90 | 20 |
| LOC_Os08g36700.1 | miR5075–3p | 974 | CDS | -110 | 90 | 21 |
| LOC_Os01g53220.1 | miR5809–3p | 233 | CDS | -104 | 89 | 20 |
| LOC_Os08g43334.1 | miR2907a, b, c, d-3p | 987 | CDS | -117 | 89 | 22 |
| LOC_Os09g28200.1 | miR2925–5p | 1071 | CDS | -100 | 89 | 19 |
| LOC_Os02g29340.1 | miR164d-5p | 76 | 5'UTR | -102 | 87 | 21 |
| LOC_Os09g28354.1 | miR169d-5p | 1053 | CDS | -98 | 87 | 21 |
| LOC_Os10g28340.1 | miR1862f, g-3p | 758 | CDS | -87 | 87 | 20 |
| LOC_Os05g45410.1 | miR2097–3p | 896 | CDS | -98 | 87 | 22 |
| LOC_Os03g12370.1 | miR396e-5p | 1042 | CDS | -96 | 87 | 21 |
| LOC_Os03g63750.1 | miR530–3p | 538 | CDS | -100 | 87 | 21 |
| LOC_Os01g43590.1 | miR2102–5p | 616 | CDS | -106 | 88 | 20 |
| LOC_Os03g53340.1 | miR2102–5p | 961 | CDS | -106 | 88 | 20 |
| LOC_Os06g36930.1 | miR2102–5p | 395 | CDS | -106 | 88 | 20 |
| LOC_Os09g35790.1 | miR2102–5p | 937 | CDS | -106 | 88 | 20 |
| LOC_Os03g53340.1 | miR414–5p | 1525 | CDS | -98 | 88 | 21 |
| LOC_Os07g44690.1 | miR5075–3p | 302 | CDS | -108 | 88 | 21 |
| LOC_Os08g43334.1 | miR5075–3p | 1397 | CDS | -108 | 88 | 21 |
| LOC_Os09g28200.1 | miR5075–3p | 292 | CDS | -108 | 88 | 21 |
| LOC_Os09g28354.1 | miR5075–3p | 199 | 5'UTR | -108 | 88 | 21 |
| LOC_Os09g35790.1 | miR5075–3p | 1410 | CDS | -108 | 88 | 21 |
| *Triticum aestivum* | | | | | | |
| TRAES3BF071100100CFD_t1 | miR9778–5p | 39 | CDS | -100 | 89 | 21 |
| TRAES3BF021000010CFD_t1 | miR9676–5p | 41 | CDS | -104 | 87 | 22 |
| Traes_4BL_B64C157DC.1 | miR9657b, c-3p | 651 | CDS | -102 | 87 | 21 |
| Zea mays | | | | | | |
| GRMZM2G301485_P01 | miR164a-3p | 488 | CDS | -102 | 91 | 21 |
| GRMZM2G118453_P01 | miR164a-3p | 215 | 5'UTR | -100 | 89 | 21 |
| GRMZM2G089525_P01 | miR172c-5p | 1181 | CDS | -93 | 88 | 20 |
| GRMZM2G179802_P01 | miR164d-3p | 1214 | CDS | -96 | 88 | 20 |
| GRMZM2G002131_P01 | miR169c, r-5p | 1102 | CDS | -102 | 87 | 21 |
| GRMZM2G173090_P01 | miR164f-3p | 687 | CDS | -102 | 87 | 21 |
| GRMZM2G173090_P01 | miR167d-3p | 1473 | 3'UTR | -110 | 87 | 23 |

The quantitative characteristics of 738 osa-miRNAs binding to mRNA of 25 HSF family genes of *O. sativa* were established. 21 genes were under the control of miRNA. 47 miRNAs were bound to mRNAs HSF, of which the largest number of binding sites had miR5075–3p. It bound to mRNA of nine genes *(LOC_Os04g48030.1, LOC_Os05g45410.1, LOC_Os06g35960.1, LOC_Os07g44690.1, LOC_Os08g36700.1, LOC_Os08g43334.1, LOC_Os09g28200.1, LOC_Os09g28354.1, LOC_Os09g35790.1)* with free energy from -106 kJ/mole to -113 kJ/mole and ΔG/ΔGm value from 86 % to 90 %. miR2102–5p had binding sites in mRNA of six genes with ΔG/ΔGm value from 86 % to 88 %. miR5809–3p interacted with mRNA of three genes (*LOC_Os01g53220.1, LOC_Os06g35960.1, LOC_Os07g44690*) with a ΔG/ΔGm value of 85 %-89 %. Two miRNA of the miR1846a, b-3p family, were bound with mRNA of *LOC_Os04g48030.1, LOC_Os08g43334.1, LOC_Os09g28200.1, LOC_Os09g28354.1* genes. Each miRNA of miR1428e, f, g-5p, miR169b, c, d-5p, miR1862f, g-3p, miR2907a, b, d-3p, c-5p, miR5539a, b-5p, miR814a, b, c-5p had one binding site in the mRNA of *the LOC_Os09g35790.1, LOC_Os09g28354.1, LOC_Os10g28340.1, LOC_Os08g43334.1, LOC_Os05g45410.1, LOC_Os03g63750.1* genes, respectively. miR529a-3p, miR530–3p, miR535–5p, miR5819–5p, miR6249a, b-5p had two binding sites in the mRNA of the HSF genes. miR1435–5p, miR164d-5p, miR2091–5p, miR2097–3p, miR2863b-3p, miR2866–5p, miR2870–3p, miR2924–3p, miR2925–5p, miR396e-5p, miR3982–5p, miR414–5p, miR5076–3p, miR5077–5p, miR5338–5p, miR5530–5p, miR5544–3p, miR5820–3p, miR5837.2–5p had only one target gene. The miRNA binding sites were located in 5'UTR and CDS mRNA HSF family genes of *O. sativa*.

To identify miRNAs that are targets genes of the HSF transcription factor family, a search was made for the binding sites of 125 tae-miRNAs with the mRNA of 51 genes of the *T. aestivum* HSF family. As a result of the search, it was found that six genes of the HSF family were targets of eight miRNAs.

miR5384–3p, miR9654b-3p, miR9657a, b, c-3p, miR9670–3p, miR9676–5p, miR9778–5p interacted with the mRNA of these genes. miR9654b-3p had binding sites in the mRNA of *Traes_5AL_16AD8DEEC.1, Traes_5BL_8D23DFA4A.1* genes. Three miRNAs of miR9657a, b, c-3p family, were bound with mRNA of the *Traes_4BL_B64C157DC.1* gene. miR5384–3p and miR9676–5p interacted with mRNA *TRAES3BF021000010CFD_t1* gene. One miRNA was bound with mRNA of *Traes_1AL_A4B5C1474.2* and *TRAES3BF071100100CFD_t1* genes. All discovered miRNA binding sites in mRNA of HSF family genes of *T. aestivum* were located only in the CDS. In the absence of complete complementarity of the interaction of miRNA with mRNA of genes, unpaired nucleotides appear, which leads to decrease of free energy of interaction of miRNA and mRNA.

As a result of studying the binding of 325 zma-miRNAs to mRNA 28 HSF family genes of *Z. mays* it was revealed that only only 13 targets targets for 20 miRNA. With mRNA of target genes miR156j, l-3p, miR156k-5p, miR164a, d, f-3p, miR166k-5p, miR167d, g-3p, miR169a, b, c, p, r-5p, miR172b, c, d-5p, miR444a, b-3p, miR482–3p were interacted. The miR164a, d, f-3p family had the largest number of target genes (four). They bound to the mRNA of *GRMZM2G118453_P01, GRMZM2G301485_P01, GRMZM2G179802_P01, GRMZM2G173090_P01* genes with free energy of -96 kJ/mole to -102 kJ/mole and ΔG/ΔGm value from 87 % to 91 %. miR167d, g-3p interacted with the mRNA of three genes: *GRMZM2G173090_P01, GRMZM2G089525_P01* and *GRMZM2G164909_P01*. In the mRNA of the *GRMZM2G002131_P01* gene, binding sites for miR156j, l-3p and miR169a, b, c, p, r-5p were found. miR482–3p bound with the mRNA of two target genes – *GRMZM2G086880_P01* and *GRMZM2G115456_P01*. miR444a, b-3p, miR172b, c, d-5p, miR166k-5p had one target gene. The miRNA binding sites were located in 5'UTR, CDS and 3'UTR mRNA HSF genes of *Z. mays*.

In the present work using the bioinformatics approach the interactions of miRNA with mRNA of HSF genes *A. thaliana, O. sativa, T. aestivum, Z. mays* were studied. The results of the following research are: a) the miRNA binding sites for the mRNA genes of the HSF family of *A. thaliana, O. sativa, T. aestivum, Z. mays* were established; b) The miRNA binding sites are located in 5'UTR, CDS and 3'UTR mRNA target genes; c) specific miRNAs for different plant species were detected which affect the expression of heat stress transcription factors.

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