non-coding RNA

MDPI

Article

Deciphering microRNAs and Their Associated Hairpin Precursors in a Non-Model Plant, *Abelmoschus esculentus*

Kavitha Velayudha Vimala Kumar †, Nagesh Srikakulam †, Priyavathi Padmanabhan and Gopal Pandi *

Supplementary Fig. S1. Sliding pattern of Small RNA Reads within a precursor.

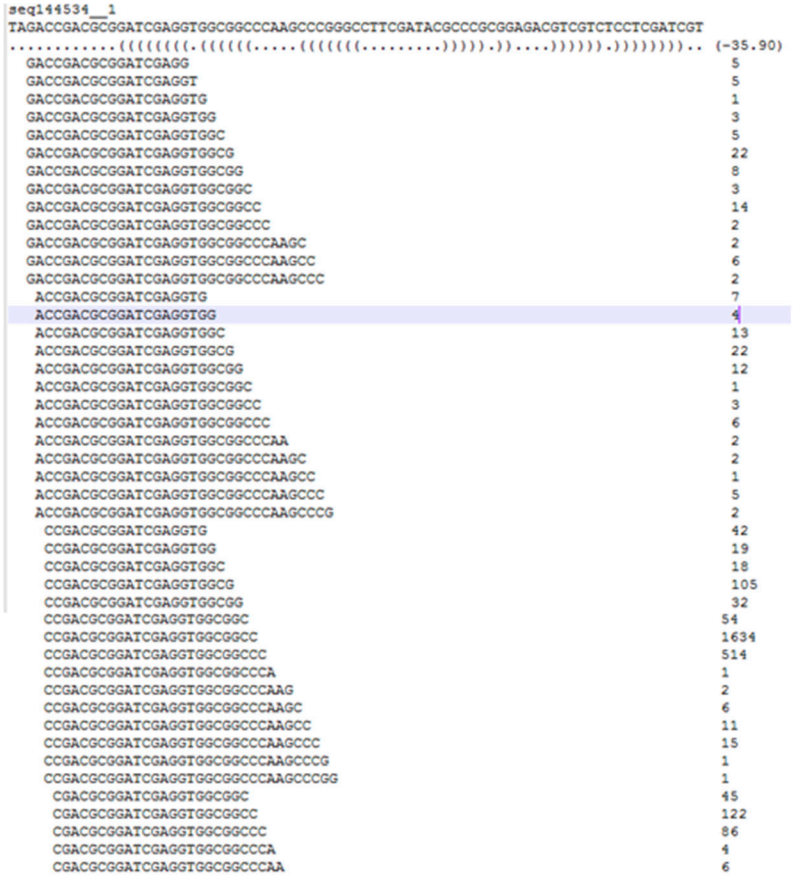

Fig. S1. Sliding pattern of Small RNA Reads within a precursor. Short reads from small RNA sequencing were mapped to the long reads of precursor sequencing by using perl scripts from miRGrep along with in-house built shell scripts. A sliding pattern of small reads was observed for many precursors as illustrated in figure S1.

Supplementary Fig. S2. Reverse Transcription PCR and restriction digestion of miRNAs

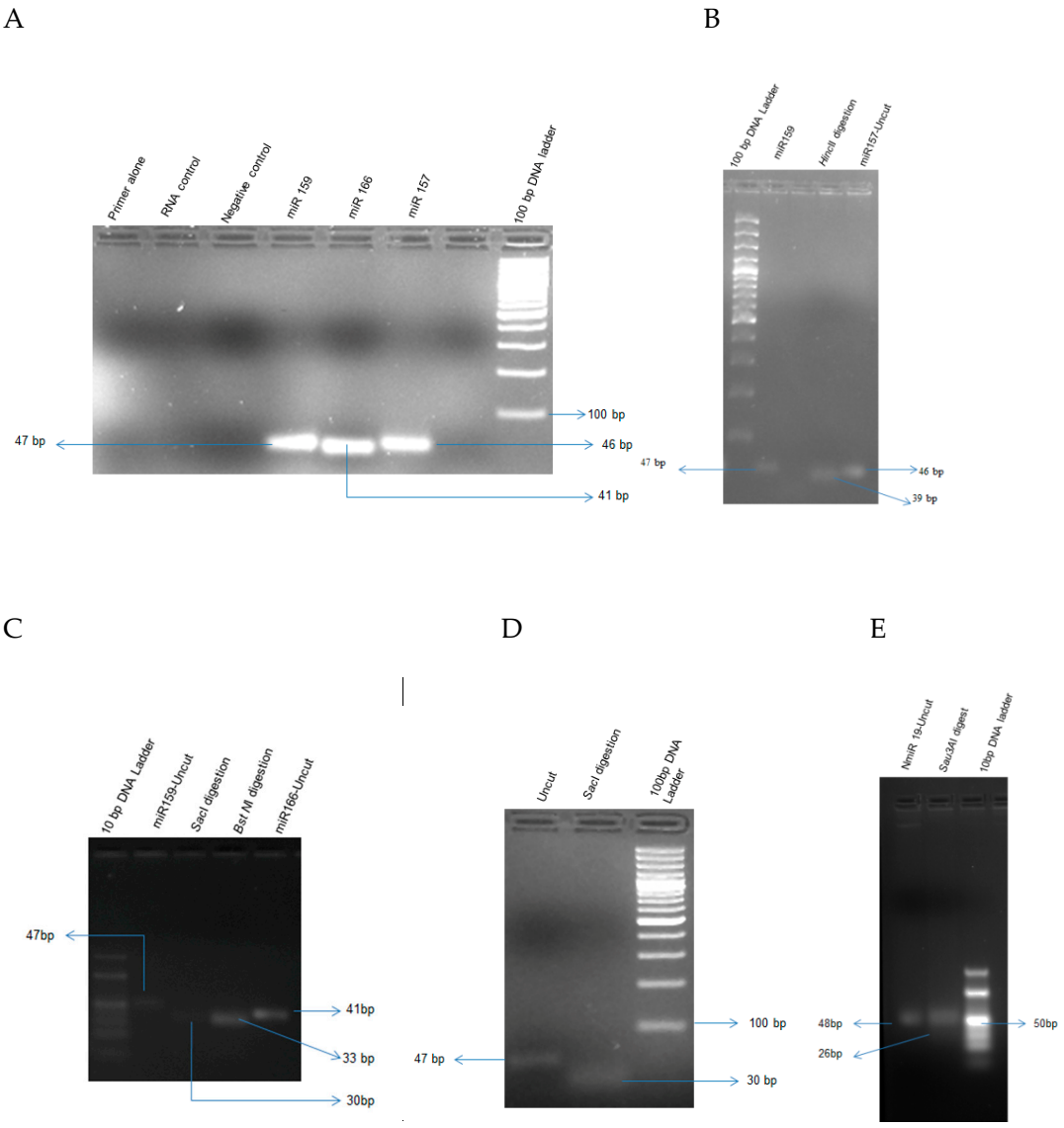

Fig. S2. Reverse transcription PCR and restriction digestion of miRNAs. PCR amplification was carried out for the randomly selected miRNAs from the reverse transcribed RNA (A). Amplified products were eluted and digested with specific restriction enzymes: *Hinc*II digestion for miRNA157 (B); *BstN*I digestion for miRNA166 (C); *Sac*I digestion for miRNA159 (D); and *Sau3A*I

digestion for NmiRNA19 (E), to ensure genuine amplification prior to qRT-PCR. All gels are 2.5%.

Table S1. List of Novel miRNAs and their precursors

miRNA ID	miRNA Sequence	Precursor Sequence	MFEI
NmiRNA1	TCTTTCCTACTCCTCCCATT	TCTTTCCTACTCCTCCCATTCCAGGGACGGAGGAGGCTAGGT	-0.750038
NmiRNA2	GCAAGGCGCAAGGAAGCTG	TTCCACAGCTTTCTTGAACTTGCGAGTCAACGGGTTAGCAAACCCGCAAGGC GCAAGGAAGCTGATTGGCGGGATCCCTCGCGGGTGCACCGCCGA	-0.750814
NmiRNA3	CGCGGATCGAGGTGGCGGC	ACGCGGATCGAGGTGGCGGCCCAAGCCCGGGCCTTCGATACGCCCGCGGAG ACGTCGTCTCCTCGATCTGCGTTGATGAAGCAATTTTATTGTGGACTAGAGTTT CTGATCTGG	-0.756122
NmiRNA4	TTTGGATTGAAGGGAGCTCTA	TTTGGATTGAAGGGAGCTCTATGCCATGGTGGTCCTGCAGAGATTATCGAACC TGGTATATCAGGGTTCCATATTGATCCA	-0.767551
NmiRNA5	TATTTCCCCAGATGATCGA	TCCATTGTCGTCCAGTGGTTAGGATATCTGGGGAAGTGATGATTATTTCCCCA GATGATCGAAGCTGAACAAACTGTGCGATTTT	-0.905393
NmiRNA6	TGTAGTATAGTGGTAAGTATTC	GTCGTTGTAGTATAGTGGTAAGTATTCCCGCGGCAAGGGATGAATATTTATCT TATCCTACACT	-0.715473
NmiRNA7	TTGTAGTATAGTGGTGAGTAT	GTCGTTGTAGTATAGTGGTGAGTATTCCTGGCAAGAGATGAATACTTATTCTA TCCTACACT	-0.770827
NmiRNA8	TGTGTTTGGAGAGGTGGAT	TTGGCATTCTGTCCACCTCCAGGTACTCAAGATATTGTGTTTGGAGAGGTGGA TAGATAGGACT	-0.813838
NmiRNA9	GTGGCTGTAGTTTAGTGG	GGTGGCTGTAGTTTAGTGGTGAGAATTCCACGTTGTTGGAATTCTCGGGTGCC AAGGAACTCCAGTCACGTCCGC	-0.827552
NmiRNA10	ATGTAGCCAAGTGGATCAAGGC	TGTTTTCTCTGCCTTGATCTATATGCGCATGGCGGATGTAGCCAAGTGGATCA AGGCAGTGG	-0.867742
NmiRNA11	TCGATAAACCTCTGCATCCAG	TCGATAAACCTCTGCATCCAGGAGCAATGAGGATAATCTGCTCTTGTGATGAT AGGGTTATC	-0.881449
NmiRNA12	CTTTTCTTTTTCTTATCC	CTTTTCTTTTTCTTATCCTACATTTGAGGATGATGAAAAAGATCAGACGGAT	-0.941243
NmiRNA13	GACCAAAGAGTCGGAGGGGGG	ACCAGCATTCTTAAGACCAAAGAGTCGGAGGGGGGAAAGCTCTCCGTTCCTG GTTCTCCTGTAGCTGGA	-0.700042

NmiRNA14	TTCGTCCCTCTAATATGT	TCTTTTATGGTTCATATTCTGGATTGGGTTCGTCCCTCTAATATGTAATATAAGATGTAATAATGGGATGAACTAAGTTATAGGCATGAAAGTGTAAGAACTCAACGG	-0.702688
NmiRNA15	ACTGGAGGTCTGTAGTTCG	TAAACTGGAGGTCTGTAGTTCGATCCTGCATGGGGGCACCTGGAATTCTCAGGTGCCAAGGAACTCCAGTCACGTCCGC	-0.704497
NmiRNA16	TTCGATACGCCCGCGGAGA	ACGTTCCCGTGGGCGGATCGAGGTGGCGGCCCAAGCCCGGGCCTTCGATACGCCCGCGGAGACGTCGTCTCCTCGATCGT	-0.744643
NmiRNA17	AGACGAAGCGGAAGCGAG	CCCTTTGTCGCTTCGATTCGTCGTTCCGCCTTAGGGGGAAGCACCCGCGTGAGCGGGAGTAGACGAAGCGGAAGCGAGAATGT	-0.746012
NmiRNA18	CCAGATATAGAGATGGCGAC	AAACCAGATATAGAGATGGCGACTAAGGTTGCTGTTTCCATTATTATATAATTTCAAGATCACAATGGATCTATGA	-0.748108

Novel miRNA sequences along with their corresponding precursor sequences are shown. NmiRNA denotes- Novel miRNA.

Table S2. List of Precursors of miRNAs from PMRD which are not present in miRBase

miRNA ID	**miRNA Sequence**	**Precursor Sequence**
MIR35-npr	GGAATATGAGTGTGTGACTT	GGTAGTTCGACCGCGAAATTTATTTGTTTCTGTATTTCCGGAATATGAGTGTGTGACTTGTTAGAATTGATC
MIRf10239-npr	CGCTTGGTGCAGGTCGGG	TCGCTTGGTGCAGGTCGGGAAATTACGATAGGTGTCAAGTGGAAGTGCA
MIRf10238-npr	TGCCTGGCTCCCTGTATGCCA	TGCCTGGCTCCCTGTATGCCACAATGTAGGCAAGGGAAGTCGGCAAAATGG
MIRf11025-akr	TGCATTTGCACCTGCACCT	TGCATTTGCACCTGCACCTTCTCATTACGATAGGTGTCAAGTGGAAGTGCA
MIRf10271-akr	TGGATTGAAGGGAGCTCT	TTTGGATTGAAGGGAGCTCTATGCCATGGTGGTCCTGCAGAGATTATCGAACCTGGTATATCAGGGTTCCATATTGATCCA
MIRf10082-akr	TGCATTTGCACCTGCACCT	TGCATTTGCACCTGCACCTTCTCATTACGATAGGTGTCAAGTGGAAGTGCA
MIRf11010-npr	TTCCACAGCTTTCTTGAAC	TTCCACAGCTTTCTTGAACTTATTACGATAGGTGTCAAGTGGAAGTGCA