

Nucleotide sequences (Figure S1) and reads coverage (Figure S2) of pre-miRNAs sequences on the B⁺ assembled genome.

1>NODE_100054_length_260_cov_13.534616:1-255 - Mzebra scaffold_35 - 2593821-2593795 B+

ATGACCGTAAATATCTGGGTGACGGCTTAGGTTTCGCTTTTGGACAAGAGGCCTTGAATTGTATTTTTTTATTTATCCACCAGGTGGCGCCCTTGCTCCAAATTTGATCCGGGGCTCG
AAAACAGCCGAAAACAGCGGGCTGCTTAGAAAGACCTGCGGGGAATGAAGCCTCAATTCGGCGTGCAGCAGCAGCTTCGCCCGCTGATCGGGCGGGAG**CCATTTTCGGCCACTAGGA**GCTT
TTAGGGT

2>NODE_101487_length_1287_cov_10.790987:793-944 - Mzebra scaffold_440 98766-98750 B+

GGGCTAG**CTCCCTTTGATAGCTCAGC**TGGTAGAGCAGAGGACCACAACACGGATGCAACGTCATATCCTTaggtagtgaTTGGGTTGACGTCGTATCCGTGGCTATCCTCAGTCGCTGGTTCAAG
TC**CAGCTTGAAGGGAGATGTTAGTG**

3>NODE_301244_length_3618_cov_31.396351:2388-2560 - Mzebra scaffold_272 112777-112696 B+

CCGGTAT**CGCTTCTCGGCCCTTT**GGCTAAGATCAAGTGTAGTATCTGTTCTTATCAGTTTAATATCTGATACGTCCCTACCCGGGGACCatatattaaattgatttttgaacagggagaTGAATAGGGGCTTG
CTCCGTCCACTCC**ACGCATCGACCTGGTATTG**CAGT

4>NODE_930998_length_49431_cov_28.273998:36203-36369 - Mzebra scaffold_25 3400510-3400445 B+

AT**GGGAGCAATGTCAC**TTTTGTCTCCAGCACCAGCCAACTAGGGGAAAATAATTCTGGTATGTATAAGCAGAAACAAAACAGTTACTGGCACACAATCACAGCTCACTGCATACAACCTCAAA
TTATTTAATTACCGGGCCCCGGGTGAGTATATTTAAGCCATCATTATCTACCGAGTGCAATTGTTGTTAGGAGAAA**CTGCTGCGGCTCATAAAAATG**GTATTTAAAAGAATAGTCGTAATTCTACC
TTC

5>NODE_130674_length_183_cov_7716.606445:90-245 - Mzebra scaffold_26 1703851-1703905 B-

AATCAGGTGGGTGGTTGCTTTAATTTATTTGACTAAGCACCATAATCTTAGATACTATCTTTGGGTGCAAATTTGAAAAGGCTCAAATGTATGCAGCATTGGAGAGTAACAGAATACATGTACTG
CCGGATGCC**GCATGTAGTCTGATGAGCC**GGCCCCGGGTTCGAGTATGACCCGTACTTTAATTTTTAATTTTTTTTT

6>NODE_35549_length_678_cov_13.758112:33-199 - Mzebra scaffold_103 1015240-1015177 B-

GCTCGCATTTCTGTTTTTTTAAACAGAGGCAGCCAGTTACTGTTTTAACTGTTCTTGTGTTTTTTTT**CCAATCCCTCCTTTAGGTTCAAGG**CTGCCAGGCATCCACTGCATGTCTTCTGTGCA
GGTG**TTTGGGGCTGAC**AggaaggcatgtgtgttttgGCTTTGTCAATTTCTGCAAATGAAATGGAACATGTAGGGATTATTTCAAGTCTGTTTTGGTGAACGATTACAAAAAGAATT

7>NODE_319948_length_10366_cov_33.443951:1946-2097 - Mzebra scaffold_94 - 1629-1679 B-

readsTGTGAAGTACTCTGACTGCCAGTCACTCTGAGCAGGGTACAGGGTATAGTAGGTAGGGAATAGCGCTCAAACACAGGCTGTGTTTGAGAG**CTATACCCAGGCTACATTCAAG**TTTCATT
CATTACTTAATAGTTACACTCTTTGACCACT

8>NODE_80652_length_7592_cov_20.613541:1132-1289 - Mzebra scaffold_210 508222-608166 B-/B+

GGTCTTCTCACCCAGTCTCCCTTCCTGTGGACGATGCCGATCTCCGCTGAGCCAGACCGCAGCCGGAGCATCACCCATGGAGCACGACGCTCCGGCTGCGGTCCGACTCGCCGGAGACGT
GTCTGCTTCTGCATAAG**GTTTCCACCGAGGTTGCTGC**

9>NODE_120347_length_9887_cov_27.215435:4975-5136 - Mzebra scaffold_188 875231-875290 B-/B+

TTTTTTTTTT**CCTCCTTGCTGCTGTGTTG**GCAGCTCAGGGCAGCGTTCTGAACCAAGTCATCCGGAATAGGCTGTGCTTGTCTGAACAAACCTATCCTGGATTACTTGAATCAGGCCAGACC
CATCGTGTGTCATCGGACTCC**CAAGCAGCCGTCCTCAA**

10>NODE_313069_length_5831_cov_30.092779:266-423 - Mzebra scaffold_77 2572262-2572318 B-/B+

AATCAGAAGGTCGGTGGTTGATCCAGGCTGCCTCCTGGCTGCCAAATATCCACACTGGCAAGCCACAATGTAGCTTCAAACATAATGTAGCTTGCCAGTGTGAATATGTGTGTATGACT
GAATGTAGTGTGAAGC**GCTTTGGGGTCCTTAGG**

Figure S1 – Sequence descriptions: #primer>NODE_contigXXXX Mzebra scaffold_XX start:end B⁻/B⁺ reads. The primer binding sites are highlighted in bold.

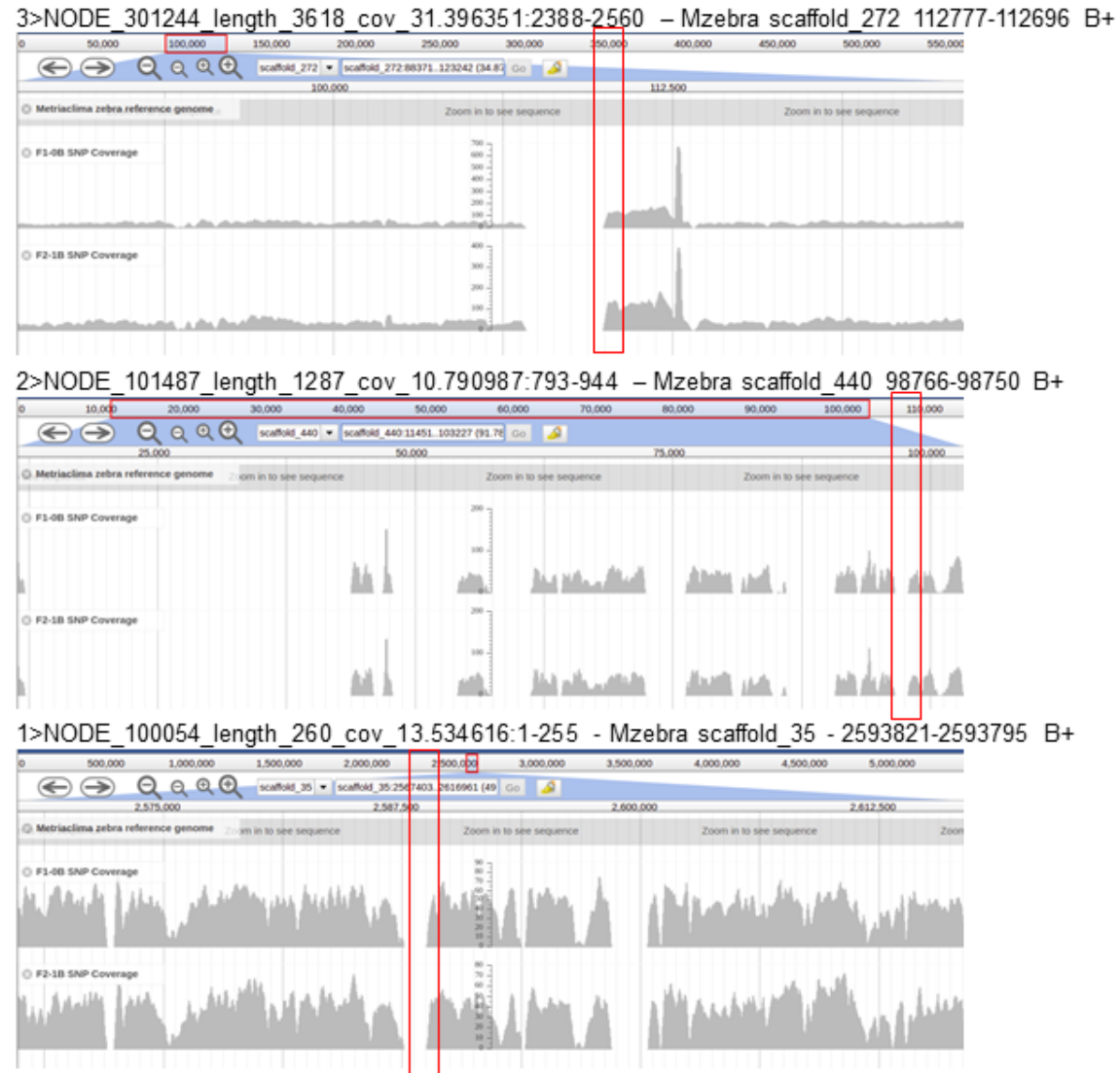


Figure S2 – Genome assembly bias. The red rectangles on the graphs highlight the pre-miRNAs regions in the *M. zebra* genome and *A. latifasciata* B⁻ and B⁺ genomic reads datasets. These miRNAs were predicted in the B⁺ assembly using the reads that failed to align in the B⁻ reference assembly expecting to find some B⁺ chromosome miRNA.