

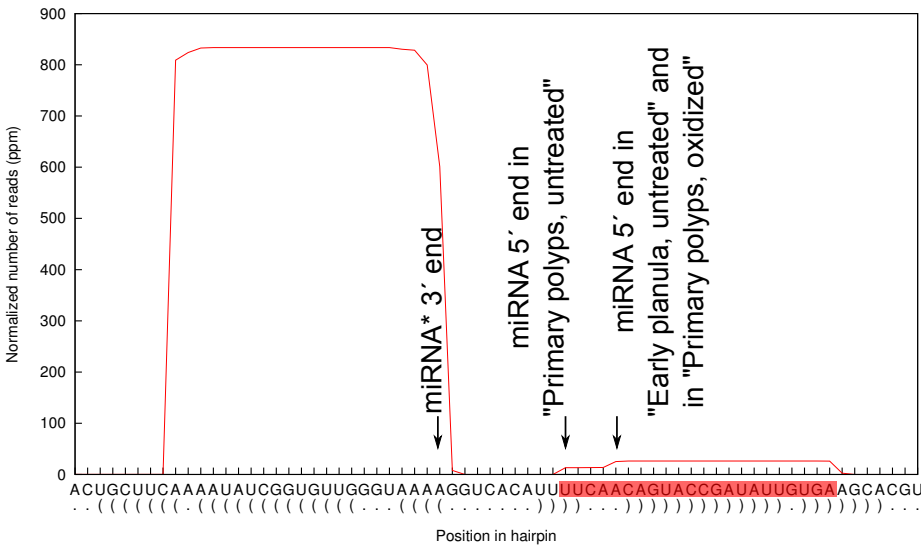
# Supplemental Figure S2

**Figure legend:** In each of the following graphs, the *x*-axis indicates nucleotide position along the hairpin sequence (predicted secondary structure is represented by dots and brackets, with dots indicating unpaired nucleotides and opening and closing brackets representing paired nucleotides). The *y*-axis indicates the number of reads covering each nucleotide in the pooled 18 deep-sequencing libraries. For miRNA candidates mapping several unbranched hairpins on the *Nematostella* genome, the read profile is shown for each genomic locus (one graph per locus).

The “cumulated abundance” given for each miRNA is the total read count across all 18 libraries, including alternative miRNA isoforms encompassing the published isoform sequence.

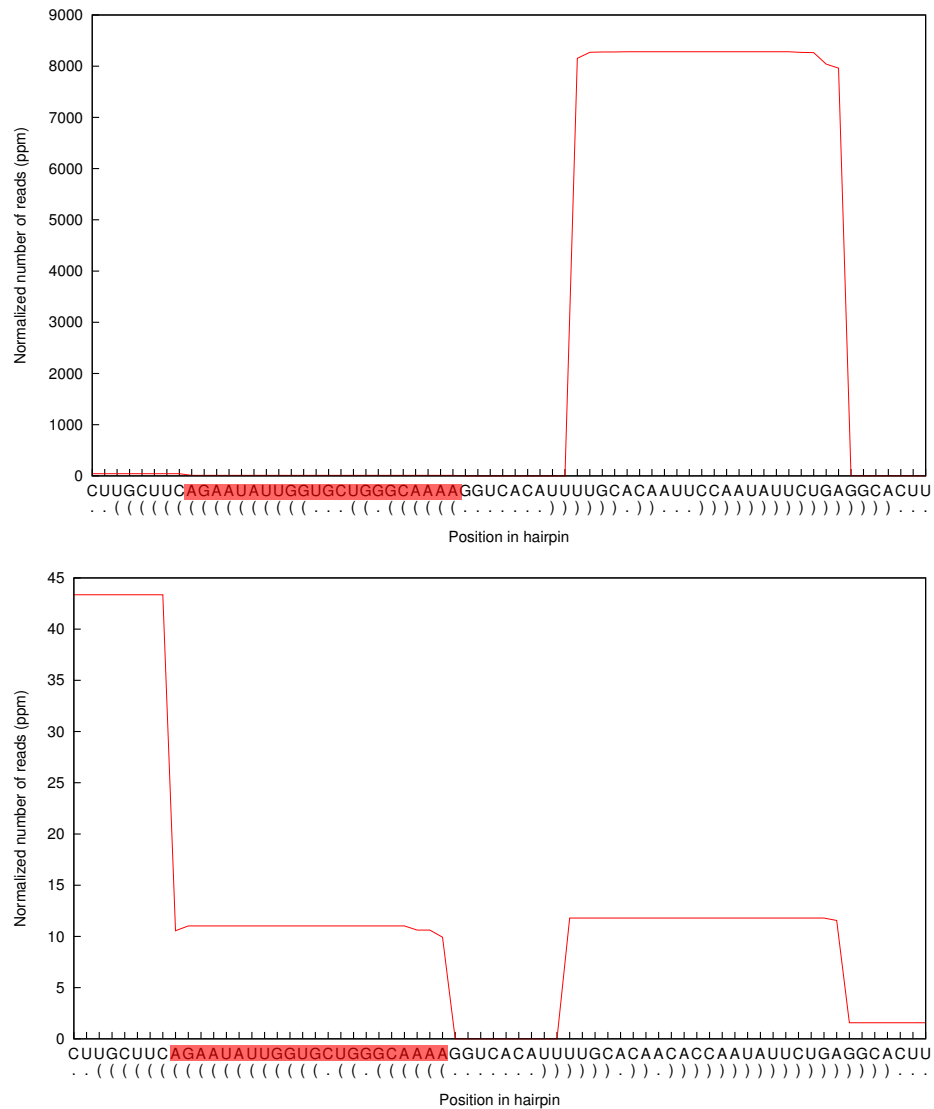
Within the hairpin sequence below the *x*-axis, the sequence of the published miRNA is shown in red.

## 1 Published miR-2024b: TTCAACAGTACCGATATTGTGA, cumulated abundance=48 reads



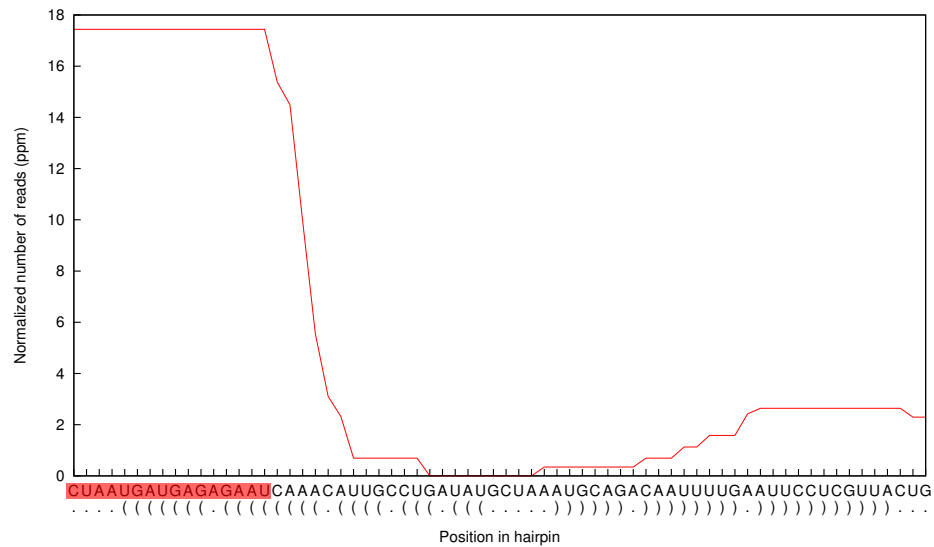
**Small RNA coverage of the pre-miR-2024b hairpin.** 3' overhang on the Dicer cleavage site is unusually long in two out of three libraries where miR-2024b read count  $\geq 10$ . *N.B.*: the predicted miRNA\* is identical to the miRNA\* of miR-2024a, hence the high number of reads for miR-2024b\* is probably partly due to miRNA\*s originating from the miR-2024a locus.

## 2 Published miR-2024d: AGAATATTGGTGCTGGGCAAAA, cumulated abundance=31 reads



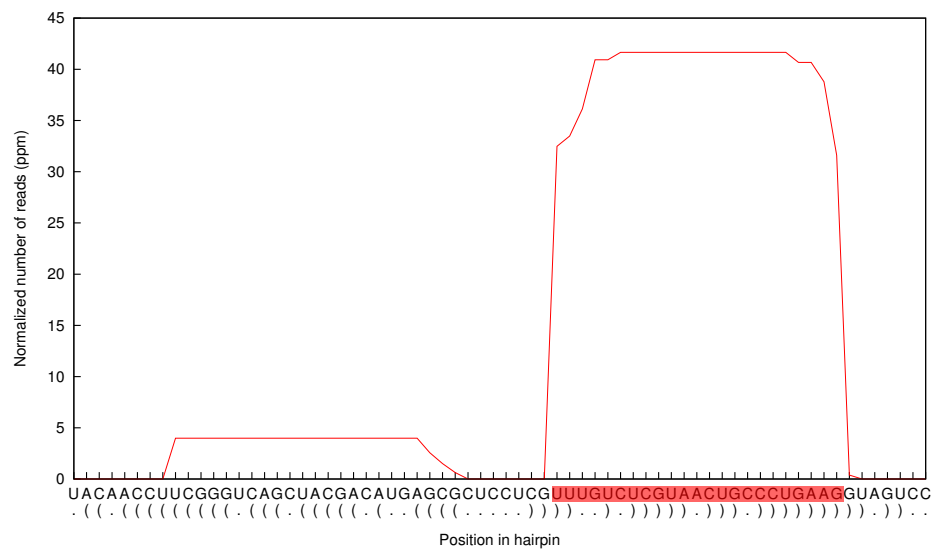
**Small RNA coverage of the possible pre-miR-2024d hairpins.** RNAs deriving from the base of the stem are more abundant than the miRNA or the miRNA\*.

### 3 Published miR-2033: AGCTAATGATGAGAGAAT, cumulated abundance=39 reads



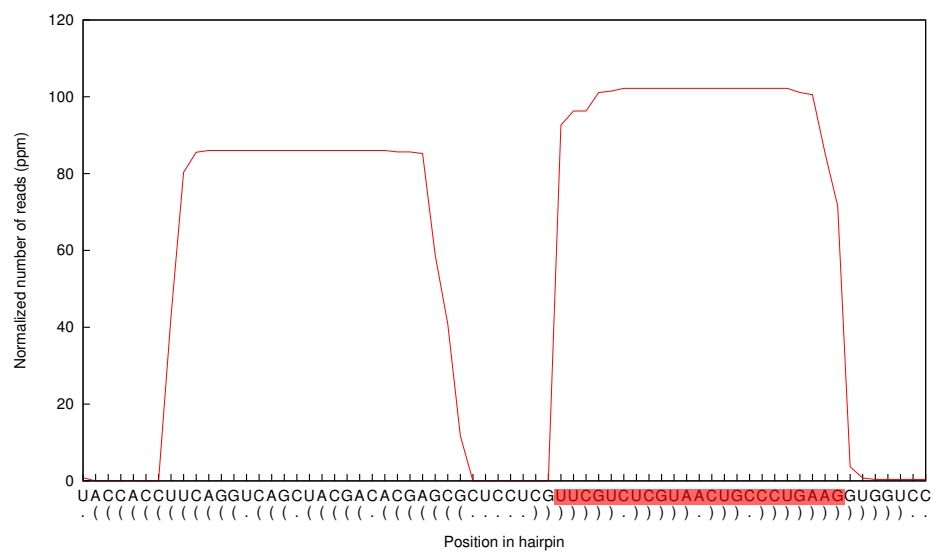
**Small RNA coverage of the pre-miR-2033 hairpin.** The distance between the Dicer cleavage site and the apical loop is unusually long.

### 4 Published miR-2040a: TTTGTCTCGTAACTGCCCTGAAG, cumulated abundance=87 reads

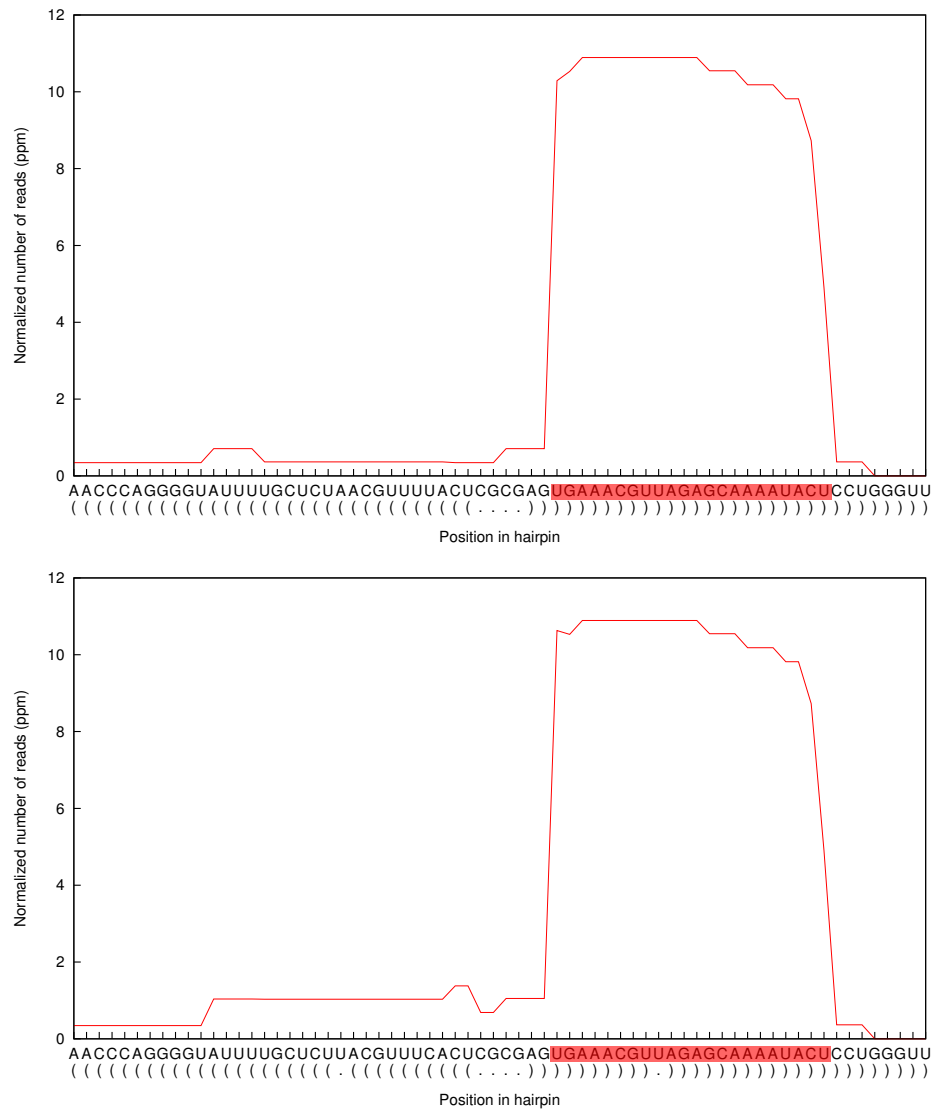


**Small RNA coverage of the pre-miR-2040a hairpin.** The presumptive miRNA has a heterogeneous 5' end.

## 5 Published miR-2040b: TTCGTCTCGTAACTGCCCTGAAG, cumulated abundance=185 reads

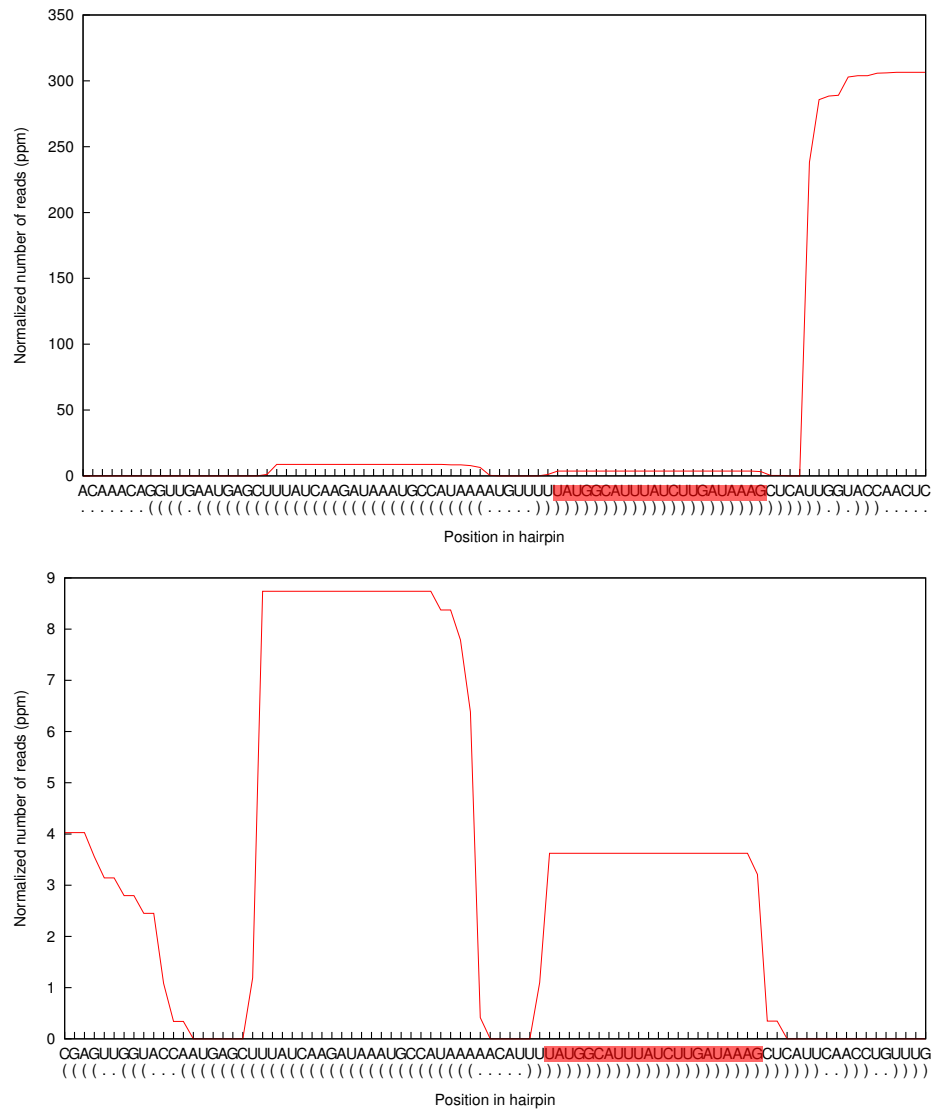


## 6 Published miR-2041: TGAAACGTTAGAGCAAAATACT, cumulated abundance=12 reads



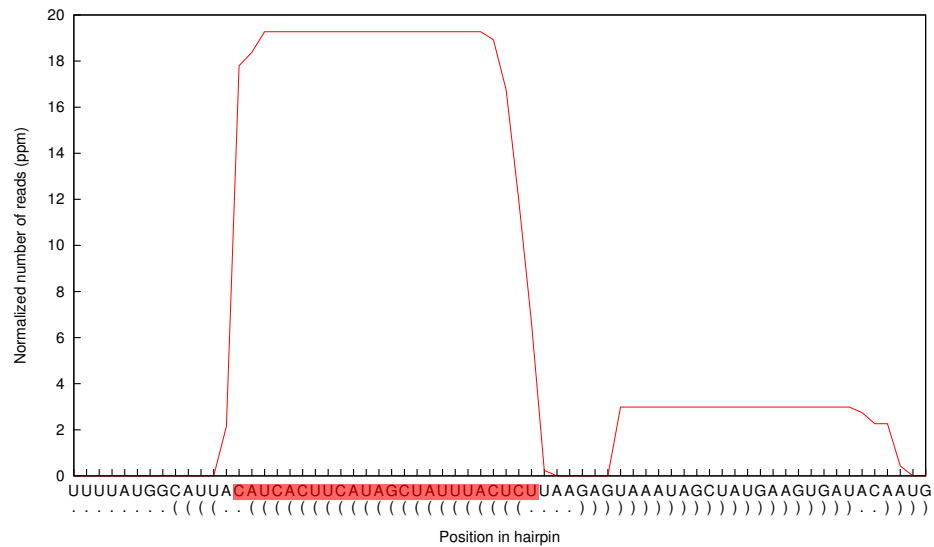
**Small RNA coverage of the possible pre-miR-2041 hairpins.** The presumptive miRNA has a heterogeneous 5' end.

## 7 Published miR-2044: TATGGCATTATCTTGATAAAG, cumulated abundance=11 reads



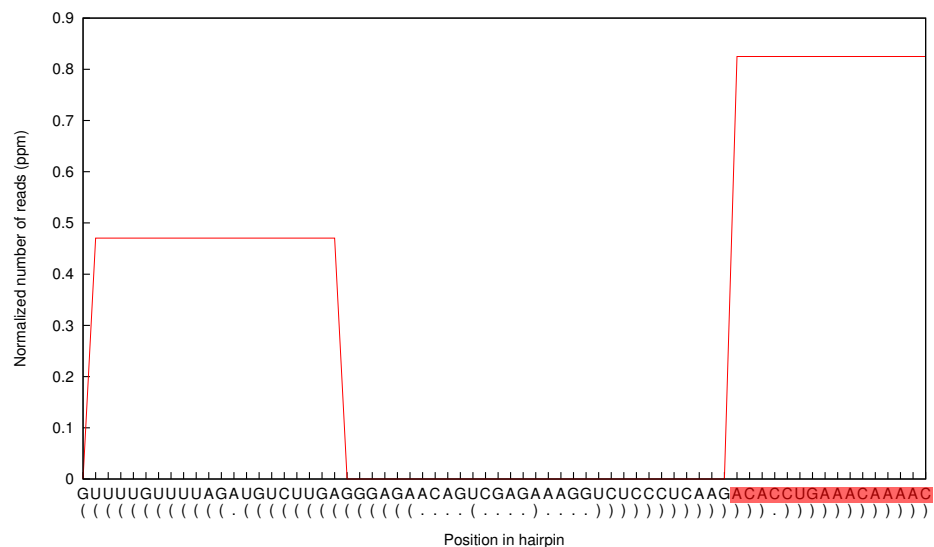
**Small RNA coverage of the possible pre-miR-2044 hairpins.** These hairpins generate multiple small RNA species.

## 8 Published miR-2046: CATCACTTCATAGCTATTTACTCT, cumulated abundance=21 reads



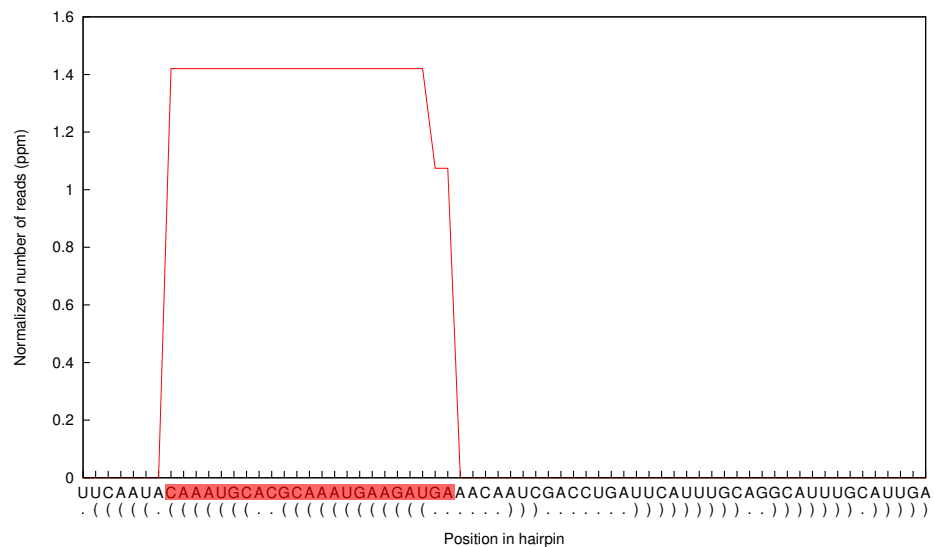
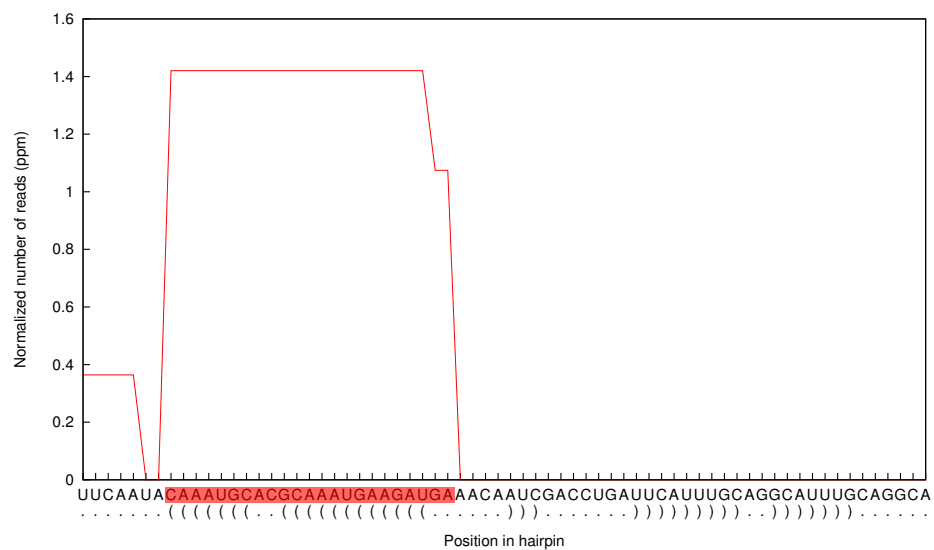
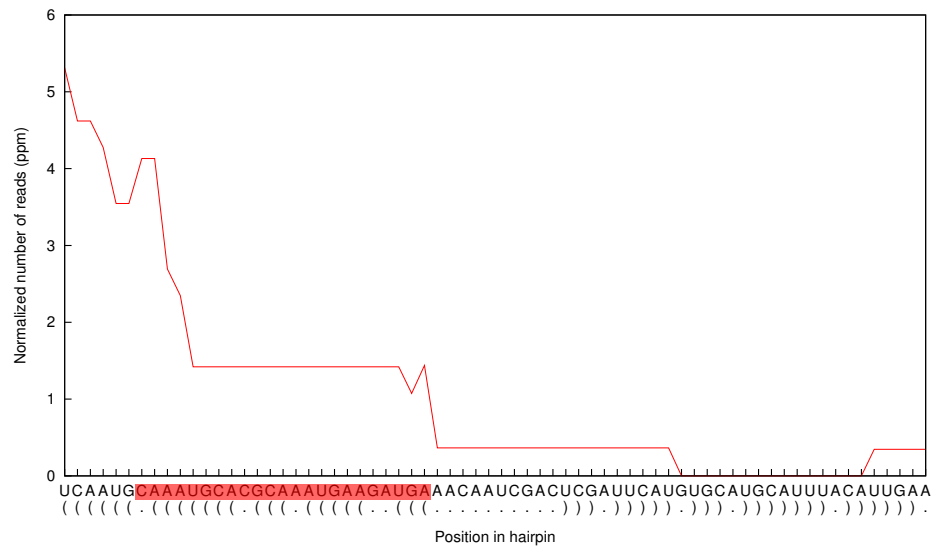
**Small RNA coverage of the pre-miR-2046 hairpin.** The presumptive miRNA has a heterogeneous 5' end.

## 9 Published miR-2048: ACACCTGAAACAAAACGCAA, cumulated abundance=3 reads



**Small RNA coverage of the pre-miR-2048 hairpin.** Low abundance does not allow a reliable assessment of small RNA distribution.

## 10 Published miR-2049: CAAATGCACGCAAATGAAGATGA, cumulated abundance=3 reads



**Small RNA coverage of the possible pre-miR-2049 hairpins.** Low abundance does not allow a reliable assessment of small RNA distribution.