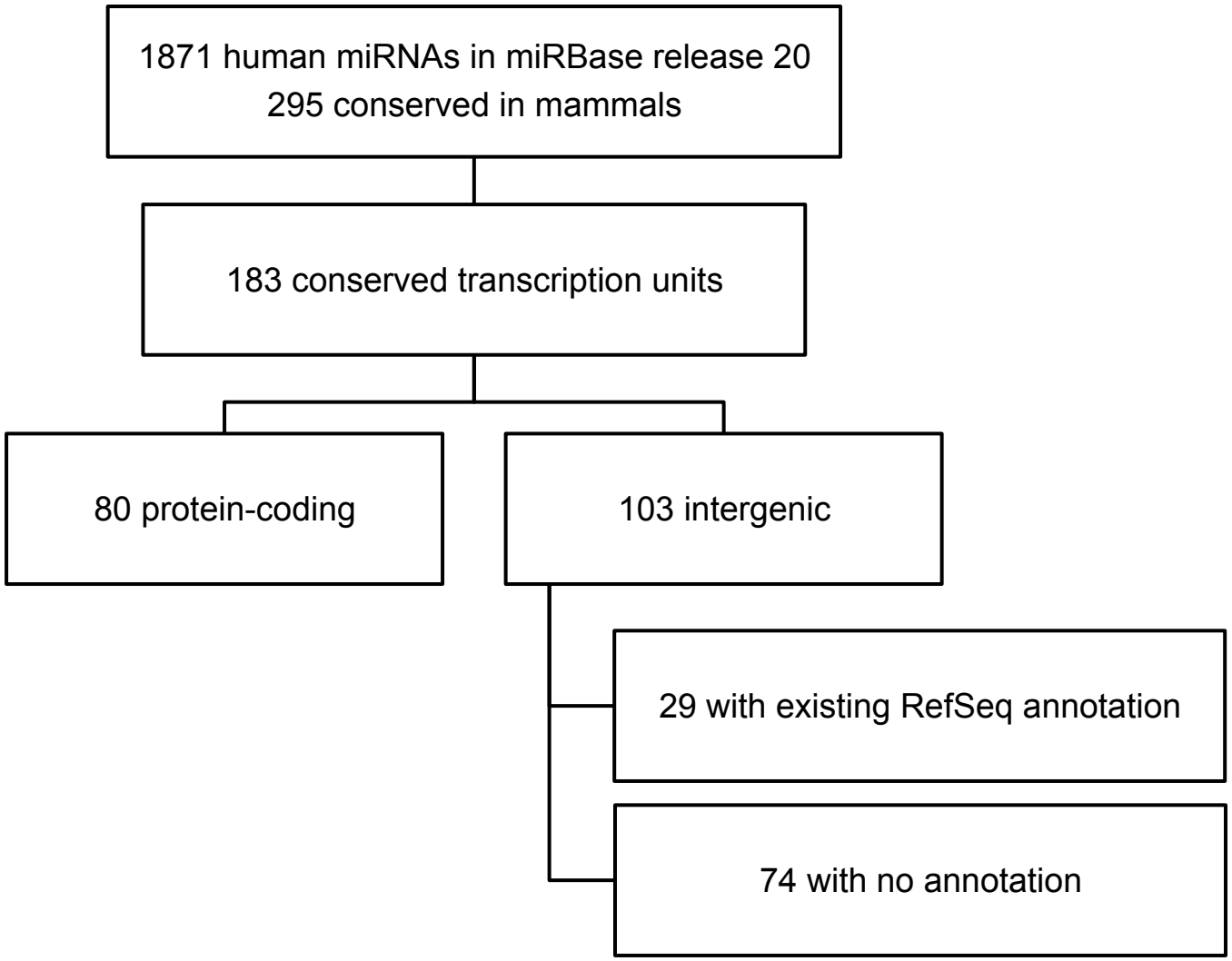
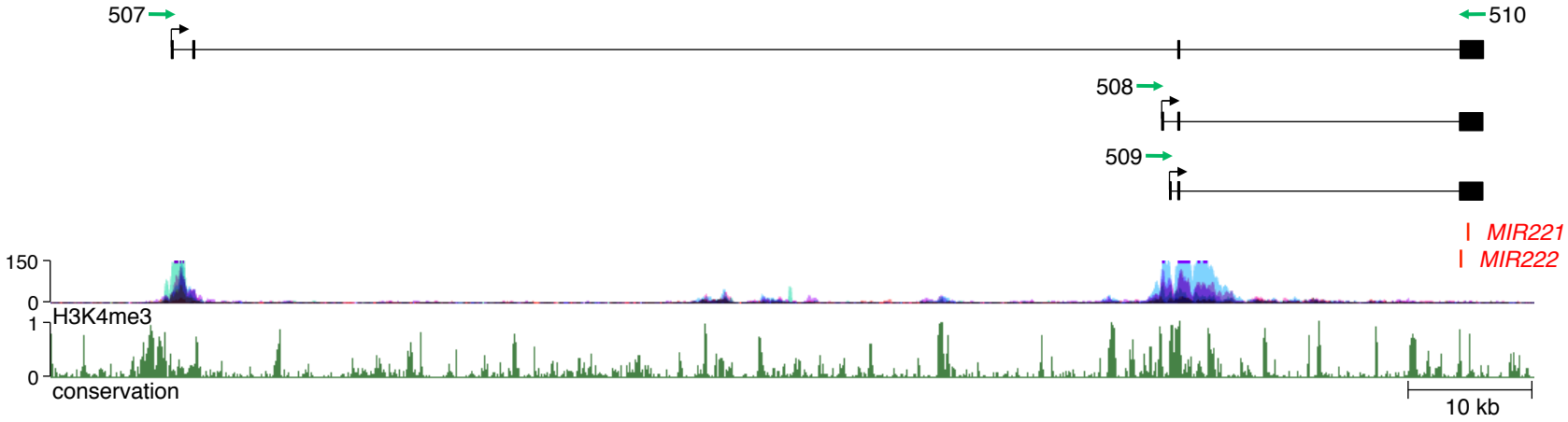


Figure S1. Overview of the organization and existing annotation of conserved human miRNA genes.



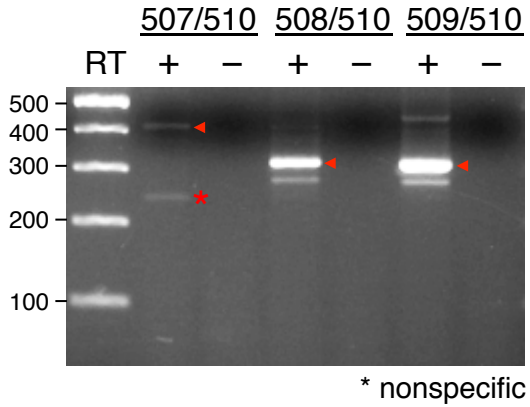


Figure S2. RT-PCR validation of newly assembled primary transcripts encoding human miR-221 and miR-222. Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing. Nonspecific PCR product indicated with asterisk.

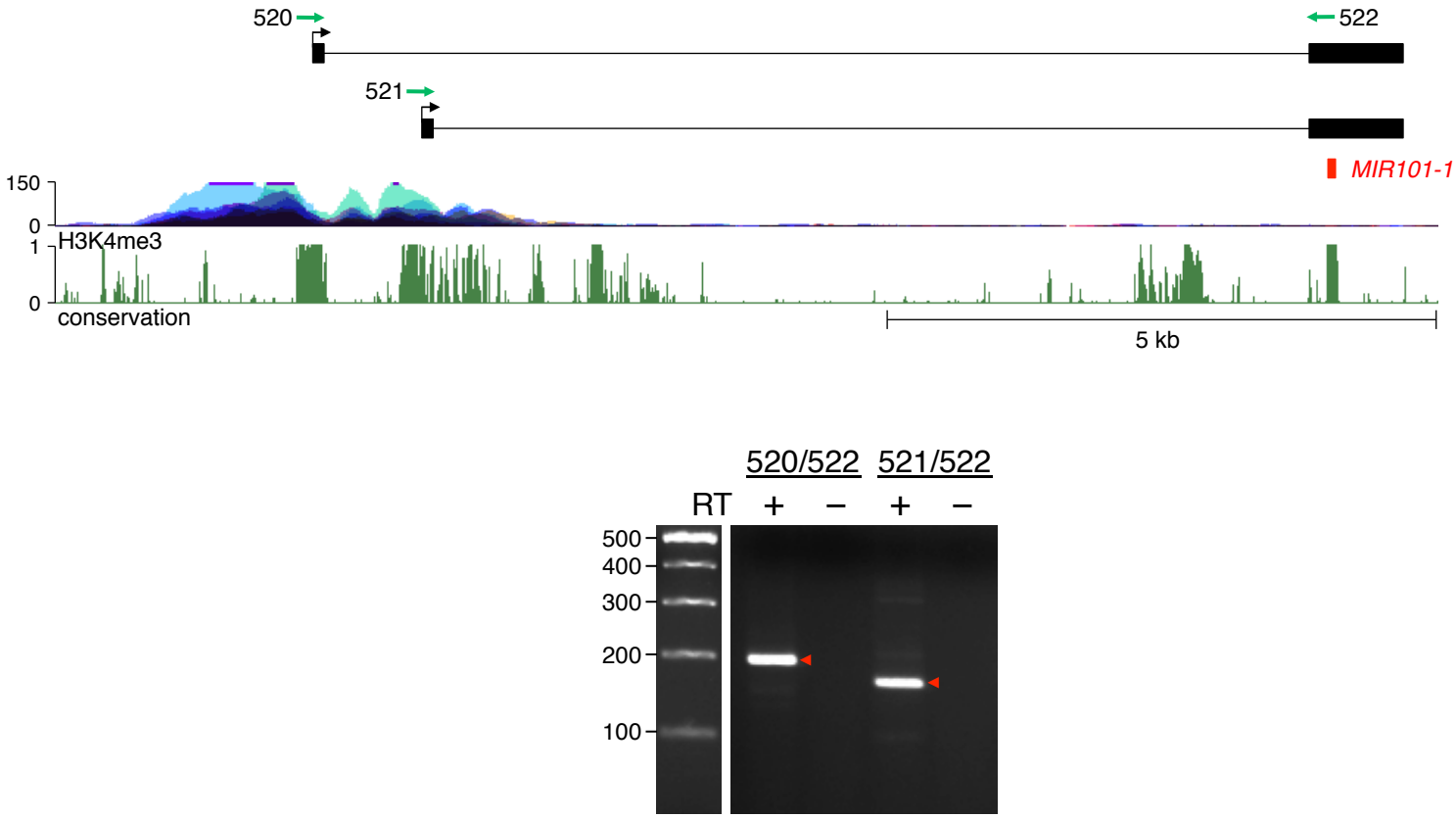


Figure S3. RT-PCR validation of newly assembled primary transcripts encoding human miR-101-1.
Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing.

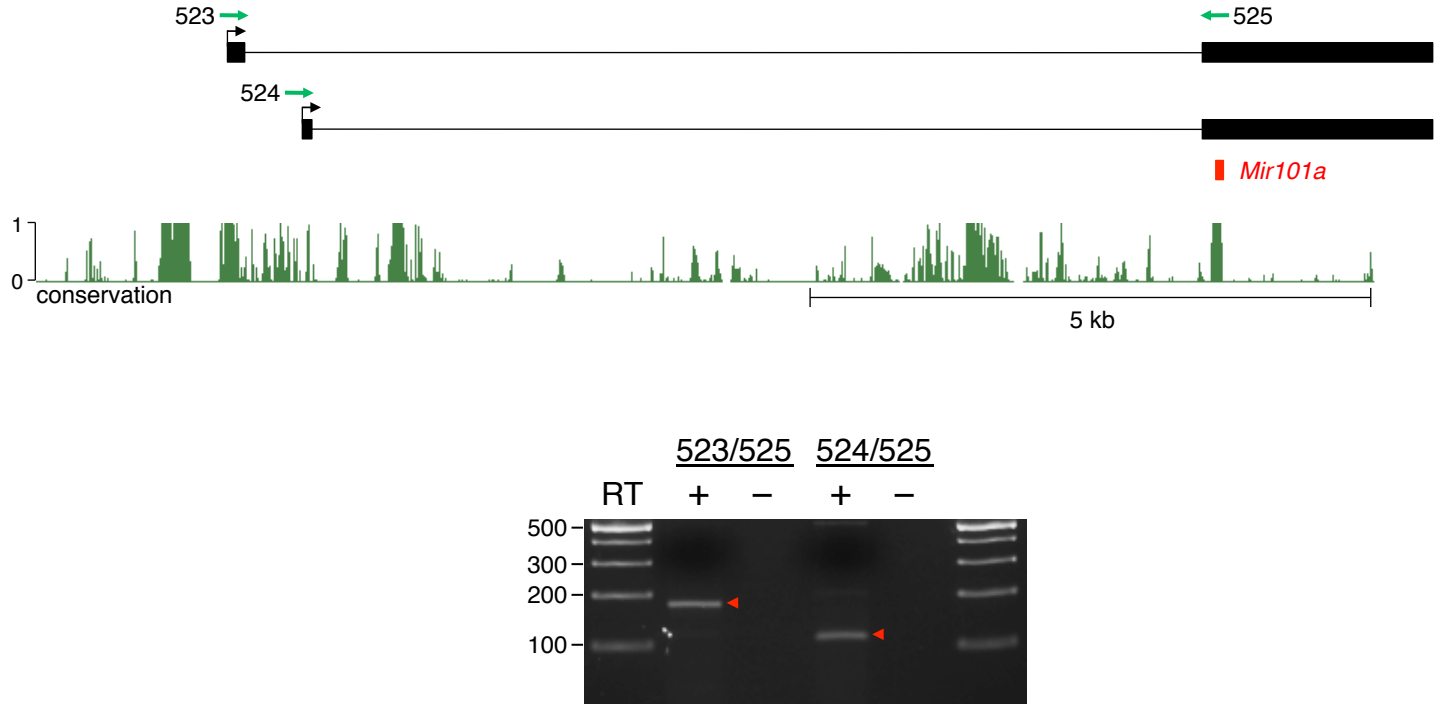


Figure S4. RT-PCR validation of newly assembled primary transcripts encoding mouse miR-101-1.
Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing.

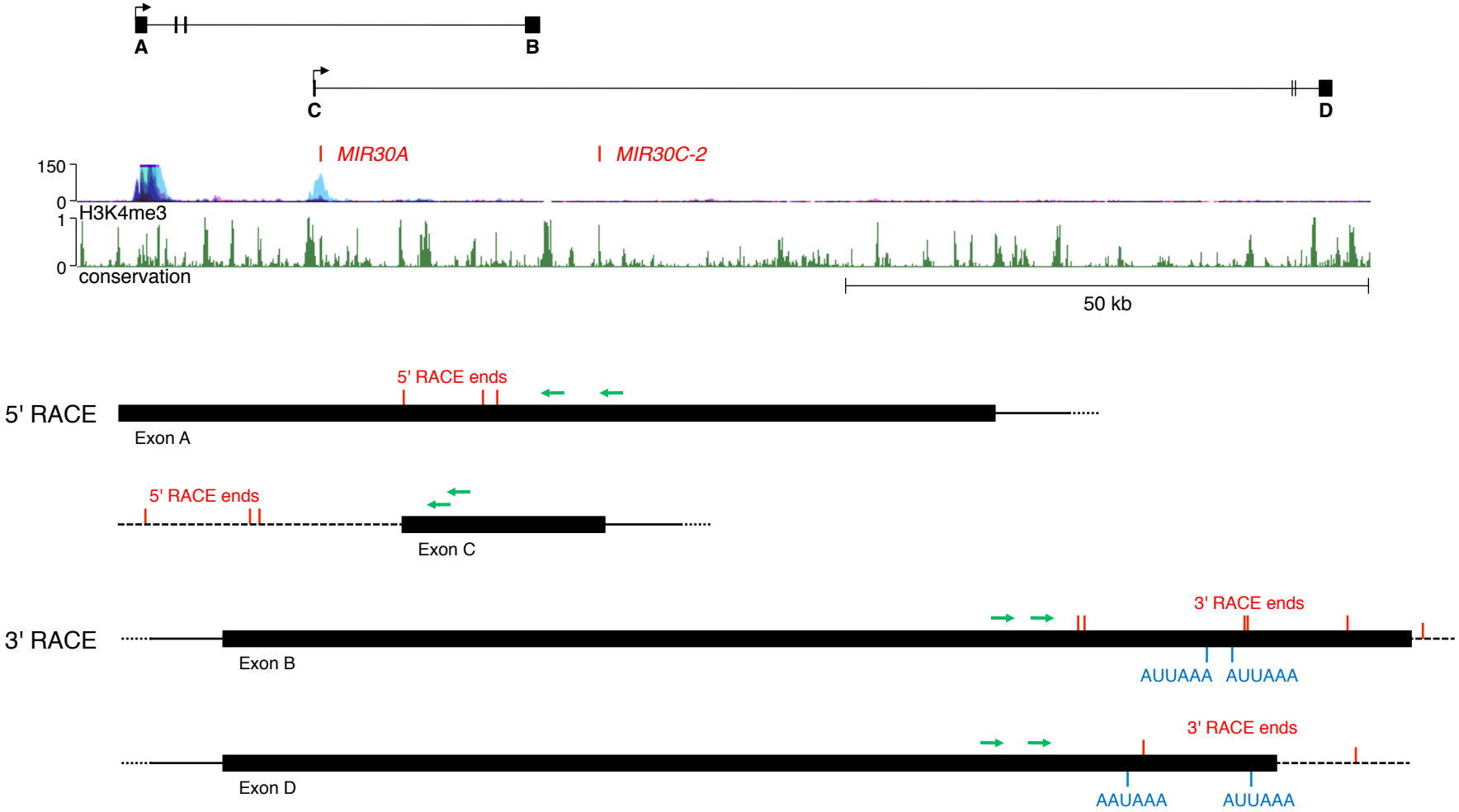


Figure S5. 5′ and 3′ RACE analysis of newly assembled primary transcripts encoding human miR-30a and miR-30c-2. The upper panel summarizes the overall transcript structures while the lower panel shows primer locations (green arrows) with red ticks indicating the end of each individual sequenced RACE clone. Putative polyadenylation signals are shown in blue.

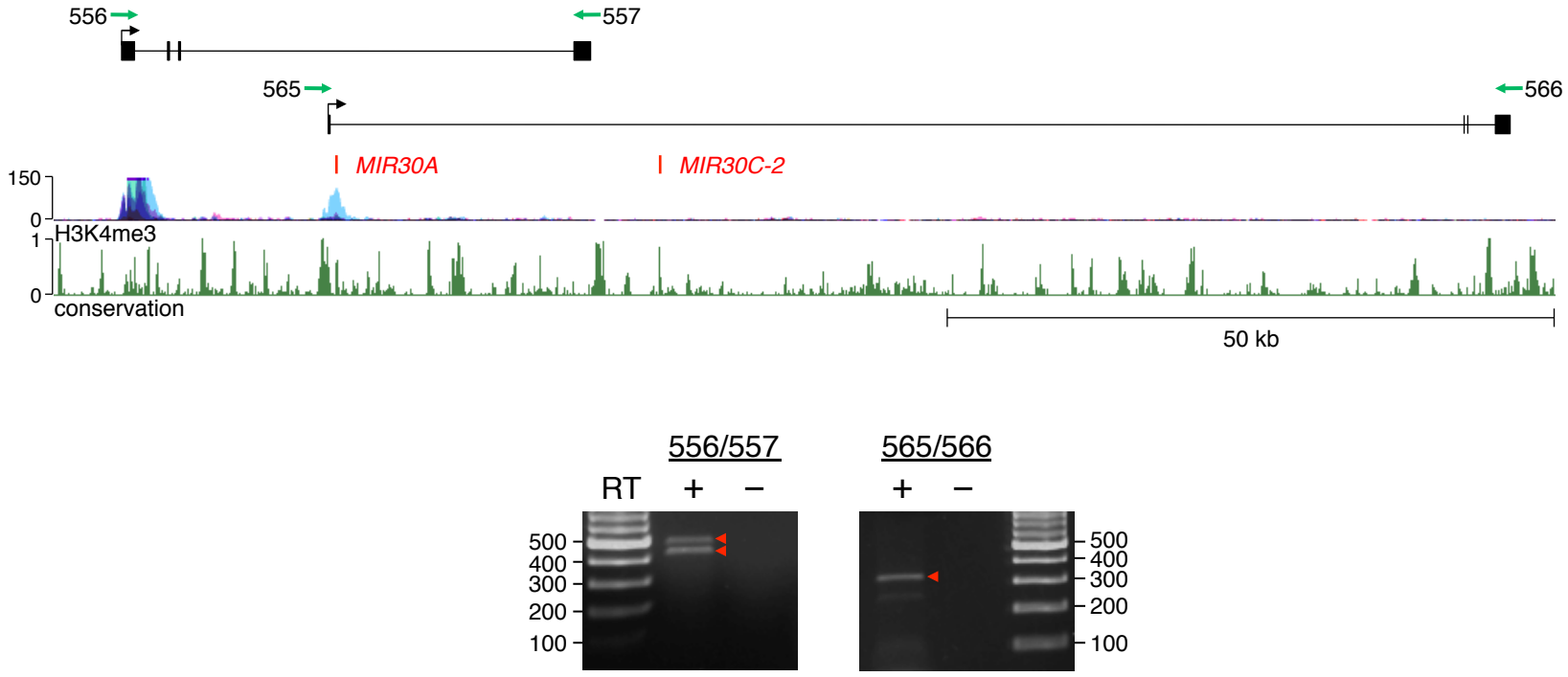


Figure S6. RT-PCR validation of newly assembled primary transcripts encoding human miR-30a and miR-30c-2. Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing. The two PCR products generated with primer pair 556/557 result from alternative splicing.

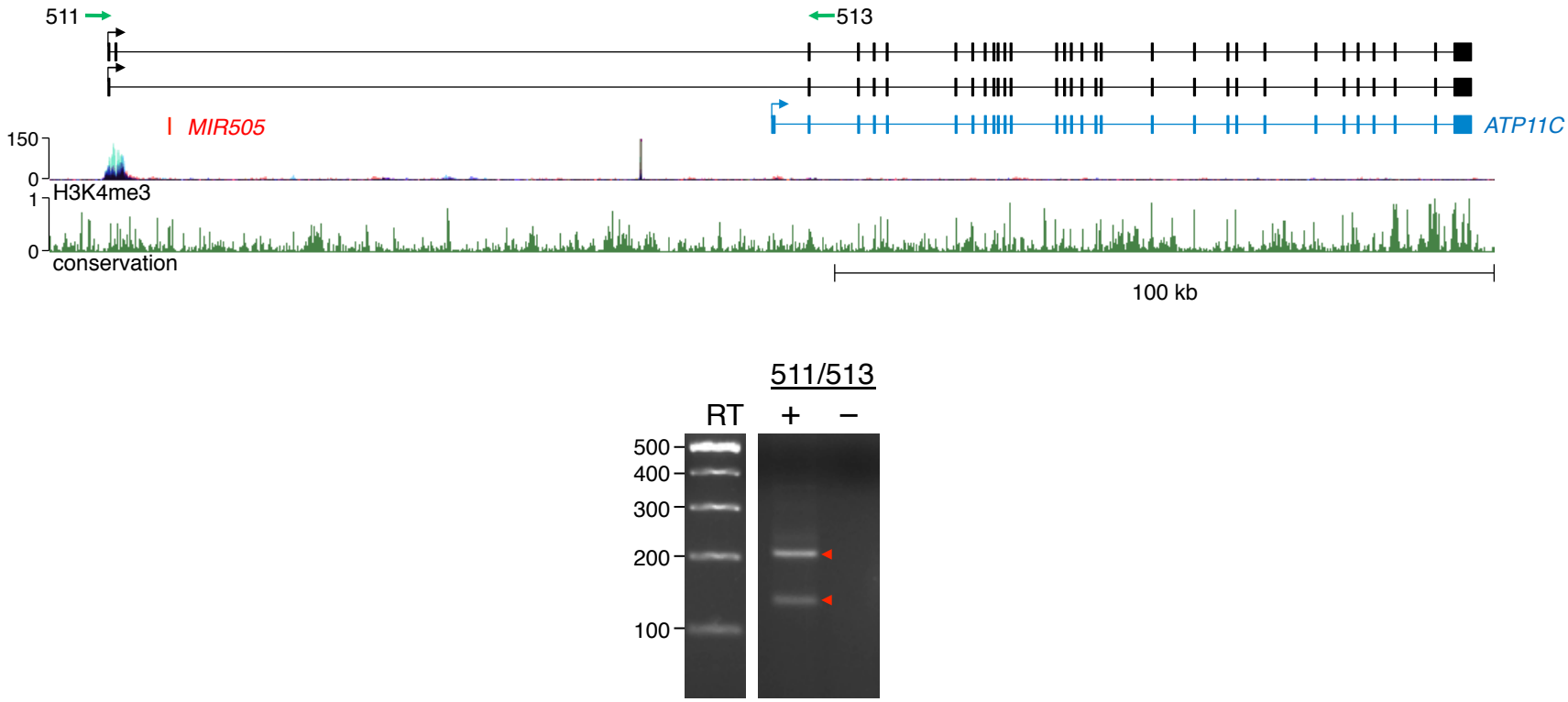


Figure S7. RT-PCR validation of newly assembled primary transcripts encoding human miR-505.
Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing. The two PCR products represent alternatively spliced isoforms.

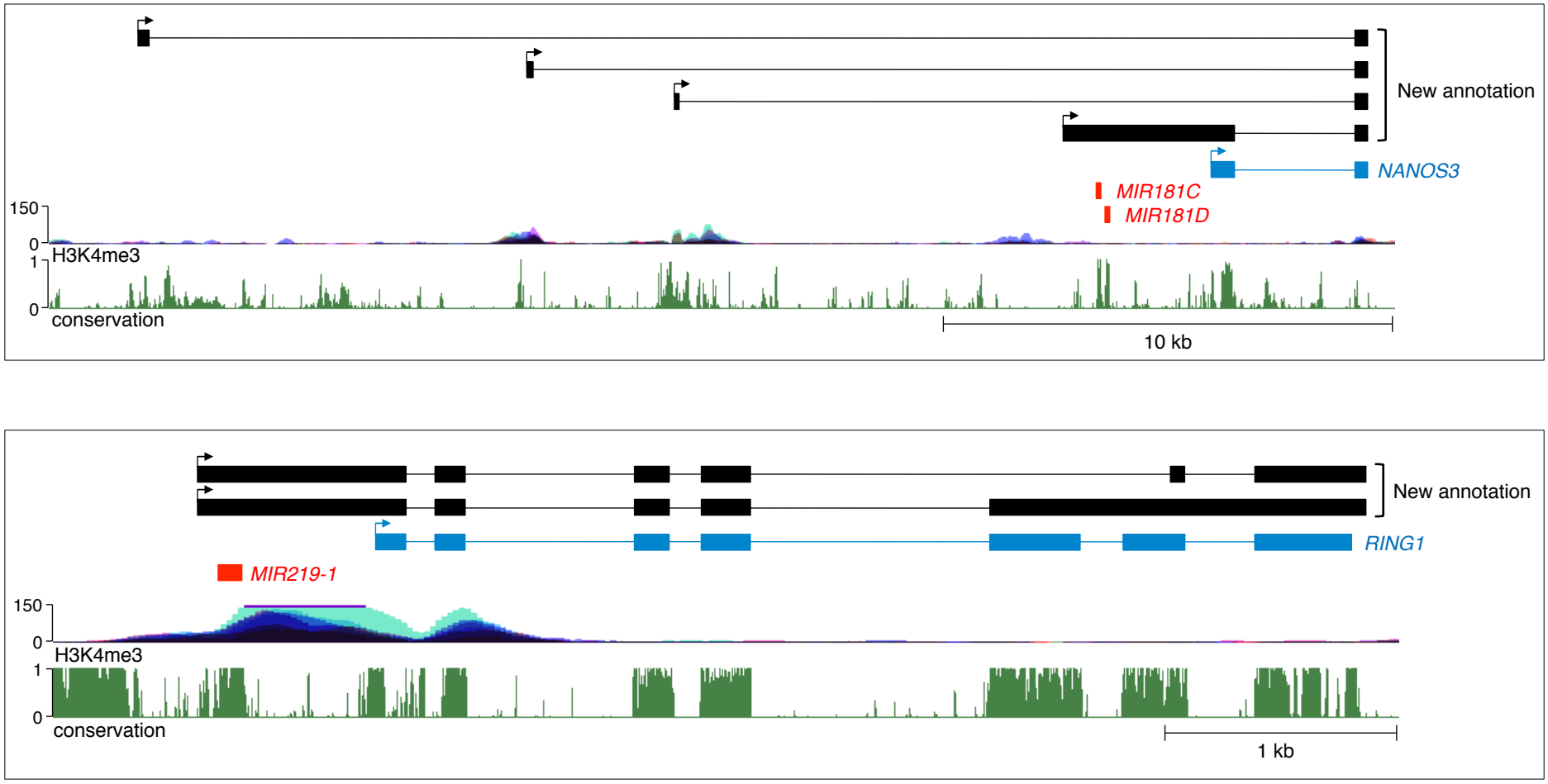


Figure S8. Additional examples of human miRNAs that are transcribed as extensions of annotated protein-coding genes.

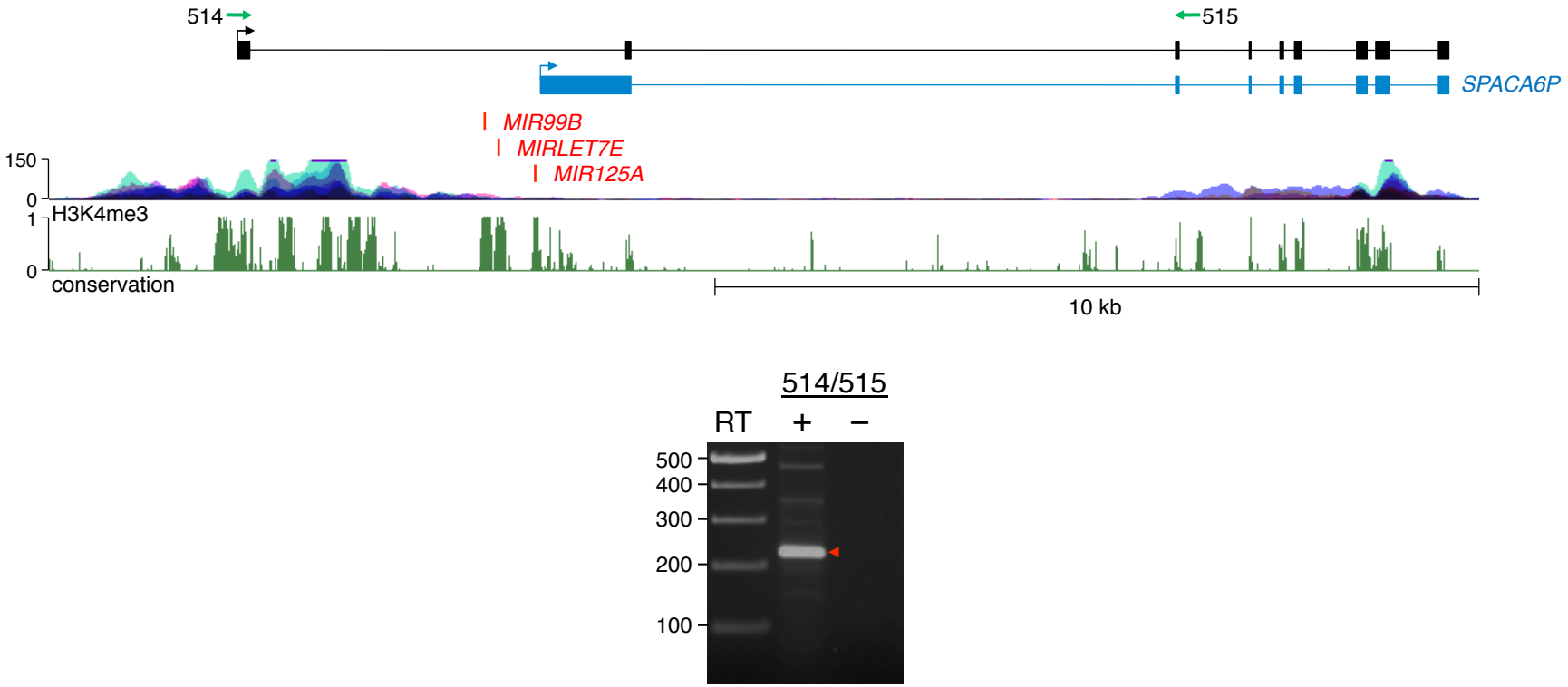


Figure S9. RT-PCR validation of the newly assembled primary transcript encoding human miR-99b, let-7e, and miR-125a. Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with the PCR product corresponding to the assembled transcript highlighted with a red arrowhead. The identity of the PCR product was verified by DNA sequencing.

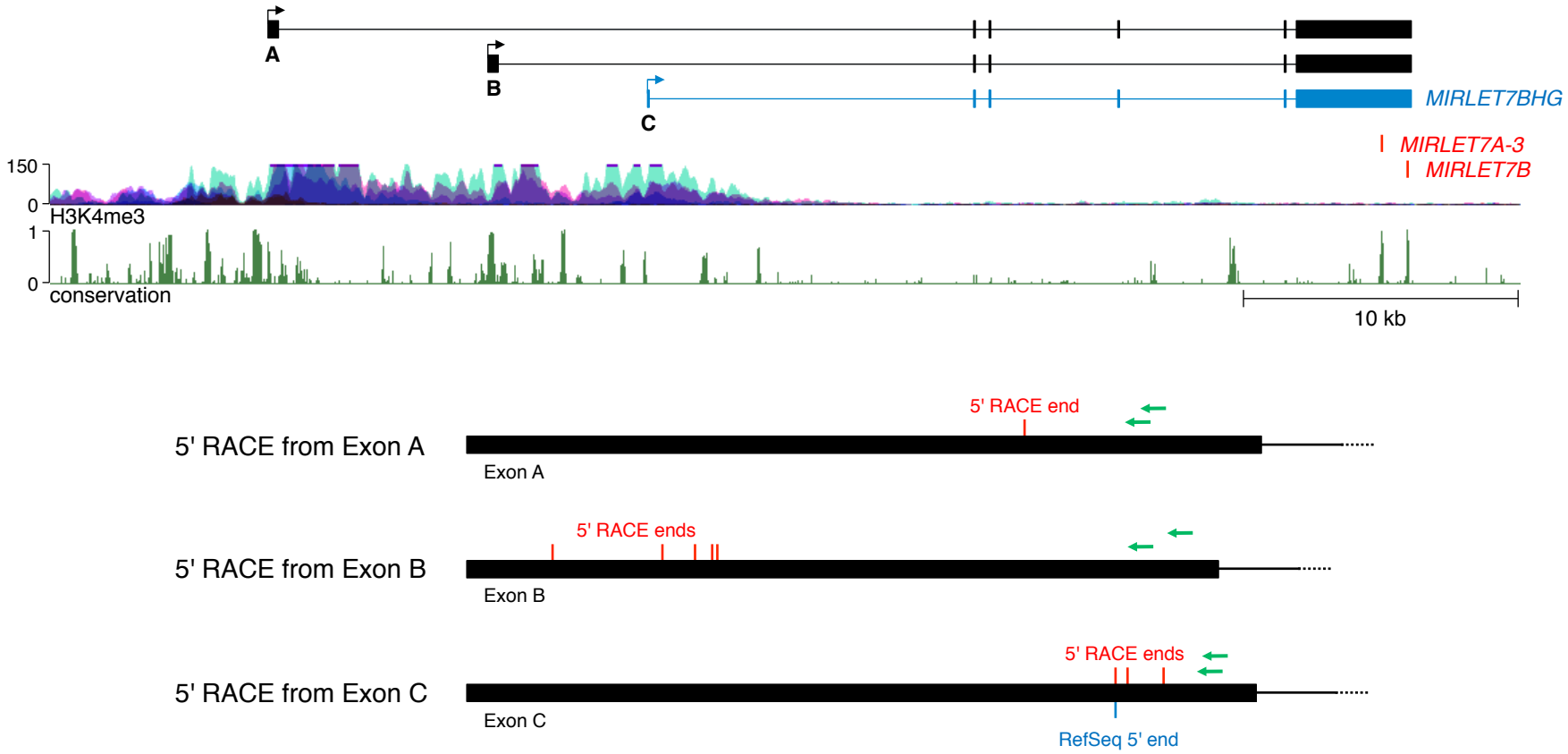


Figure S10. 5′ RACE analysis of primary transcripts encoding human let-7a-3 and let-7b.
The upper panel summarizes the overall transcript structures while the lower panel shows primer locations (green arrows) with red ticks indicating the end of each individual sequenced RACE clone.

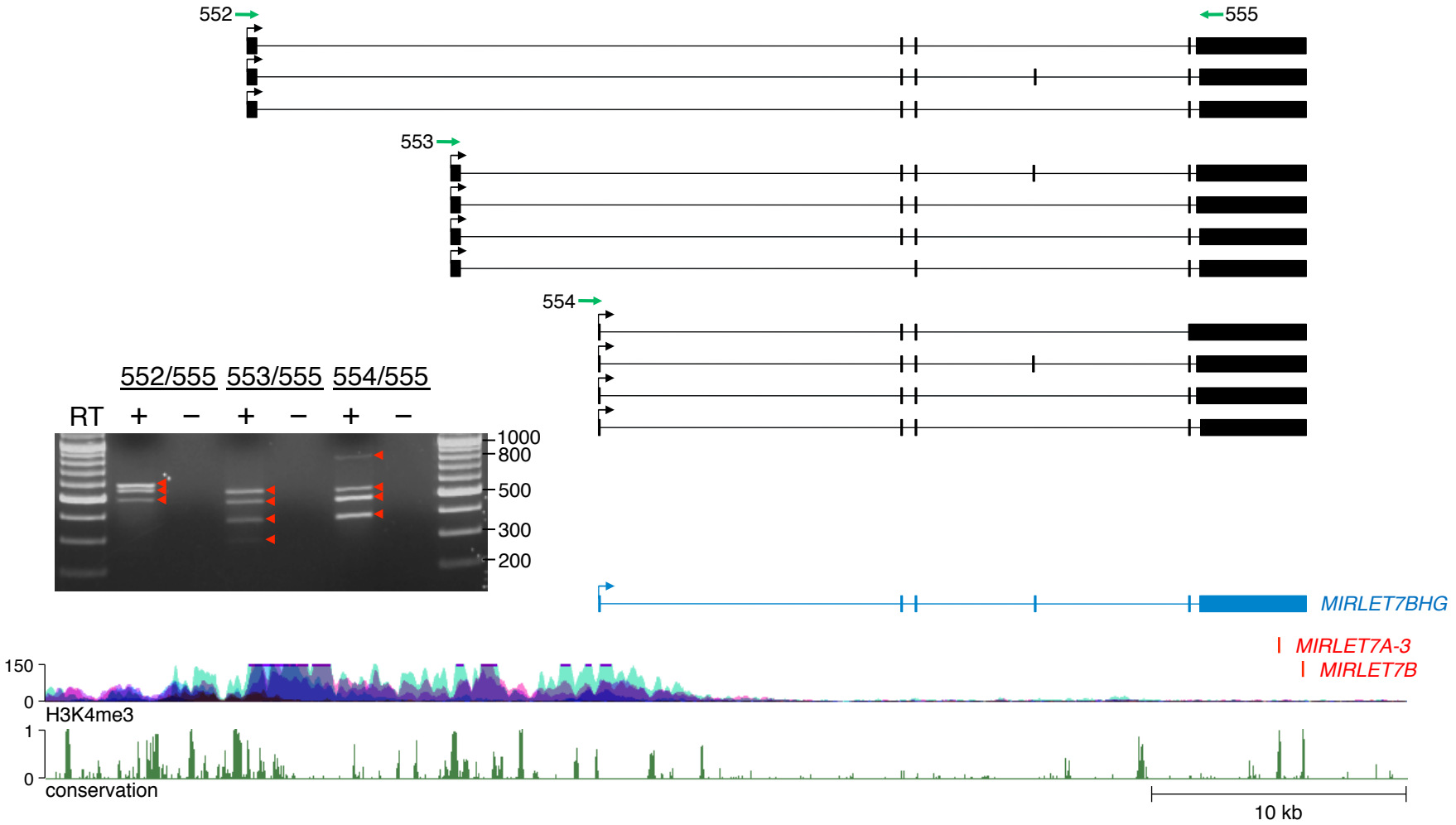


Figure S11. RT-PCR validation of newly assembled primary transcripts encoding human let-7a-3 and let-7b. Green arrows indicate the location of primers. RT-PCR results are shown to the left of the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing. The multiple PCR products generated with each primer pair represent alternatively spliced isoforms.

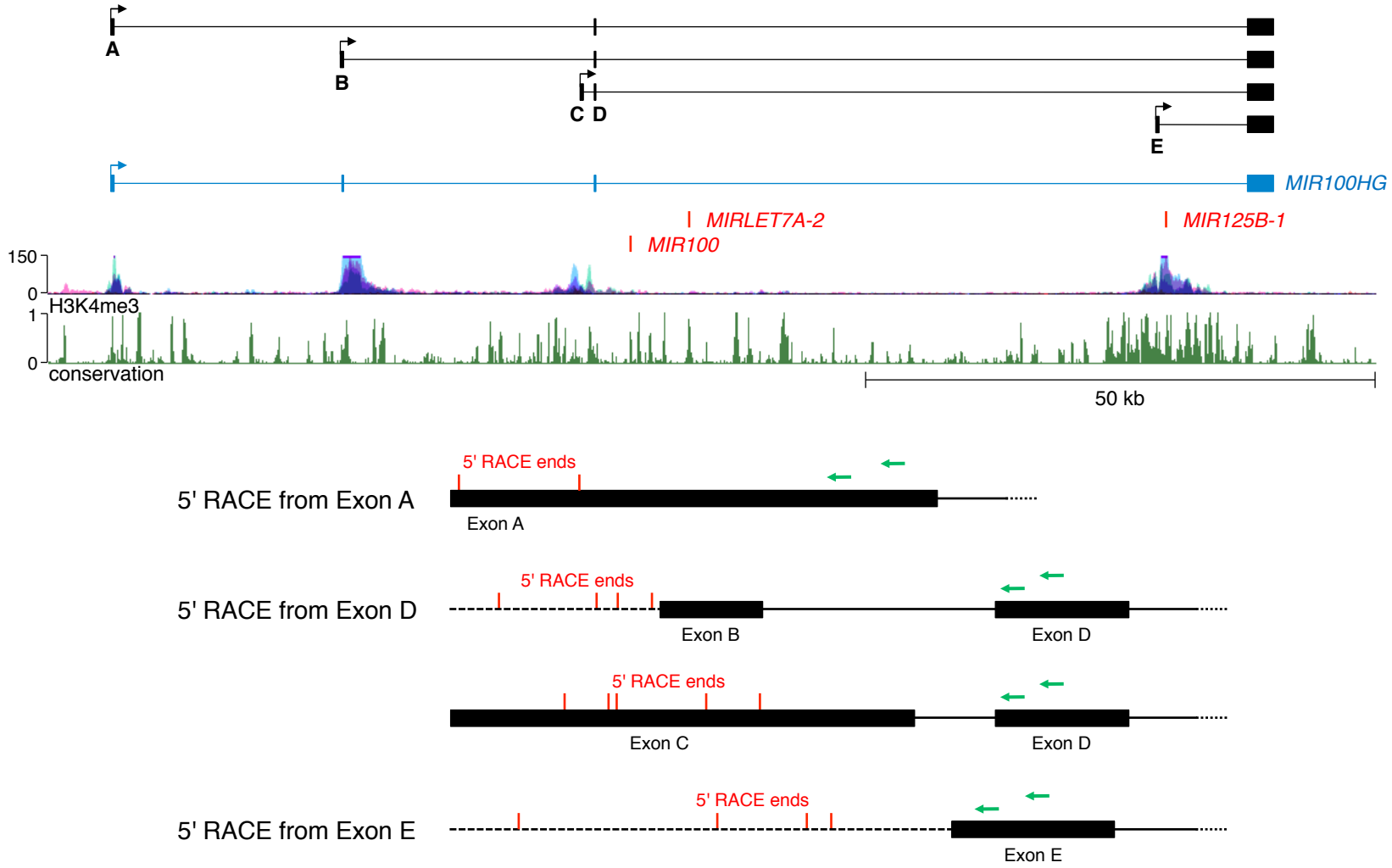


Figure S12. 5′ RACE analysis of primary transcripts encoding human miR-100, let-7a-2, and miR-125b-1.
The upper panel summarizes the overall transcript structures while the lower panel shows primer locations (green arrows) with red ticks indicating the end of each individual sequenced RACE clone.

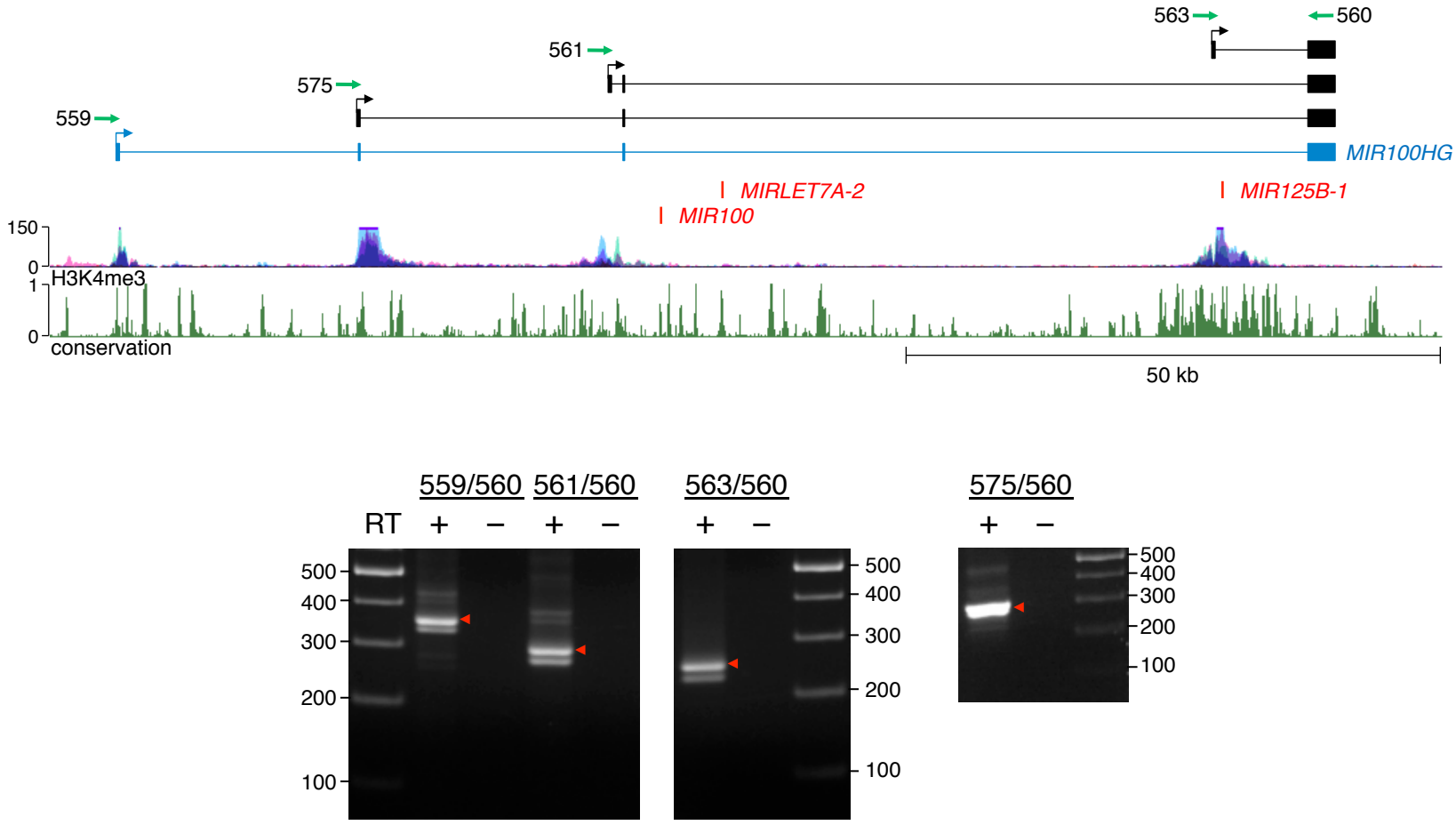


Figure S13. RT-PCR validation of newly assembled primary transcripts encoding human miR-100, let-7a-2, and miR-125b-1. Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing.

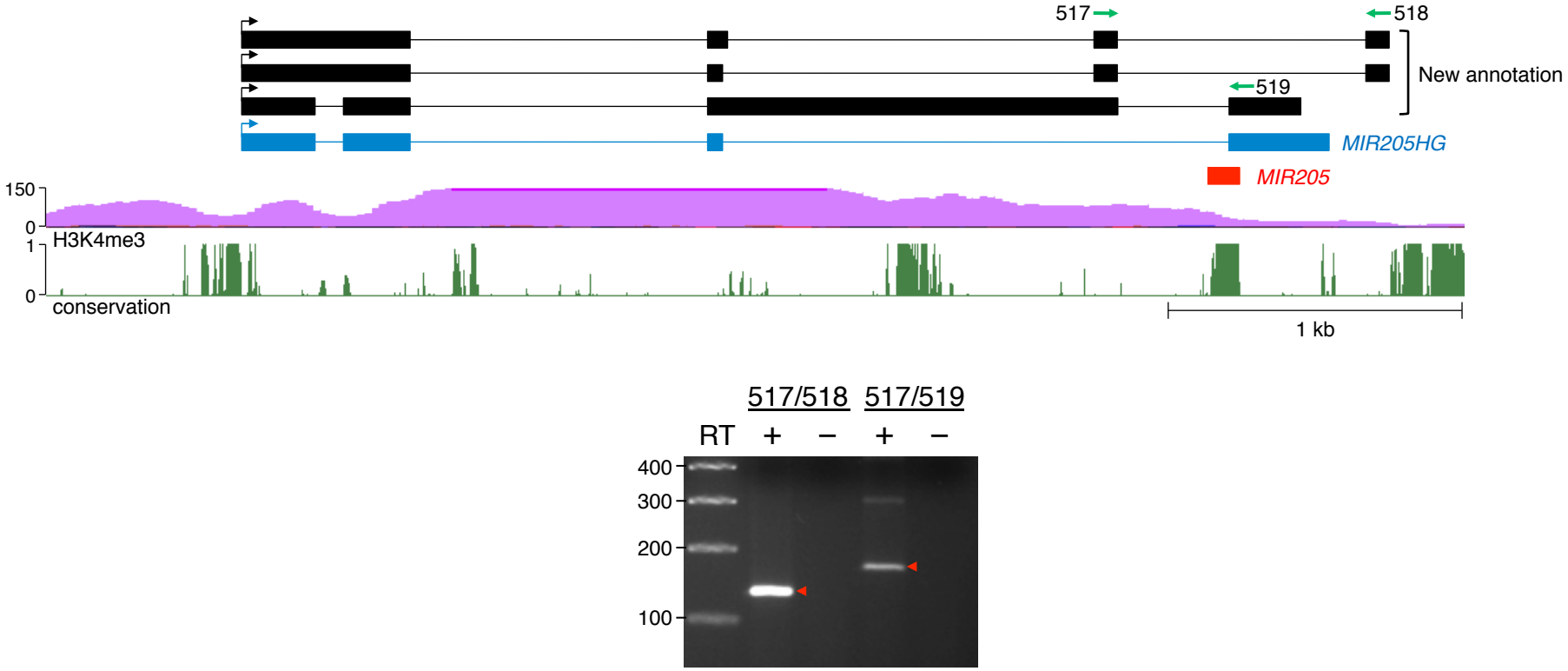


Figure S14. RT-PCR validation of primary transcripts encoding human miR-205.
Green arrows indicate the location of primers. RT-PCR results are shown below the transcript alignments with PCR products corresponding to the assembled transcripts highlighted with red arrowheads. Identities of all PCR products were verified by DNA sequencing.