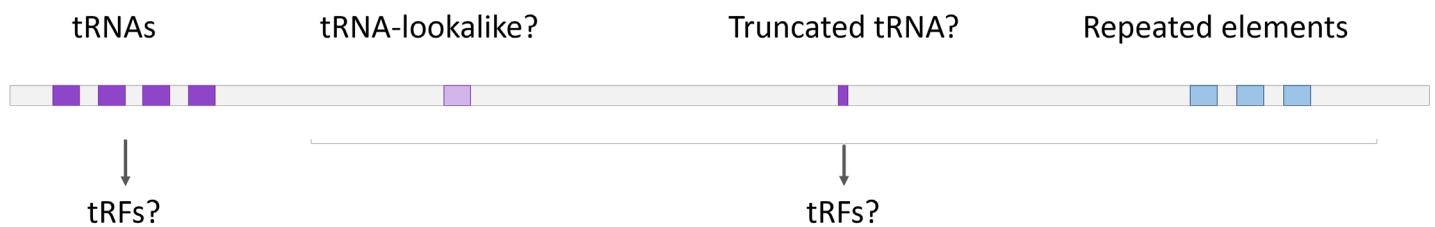
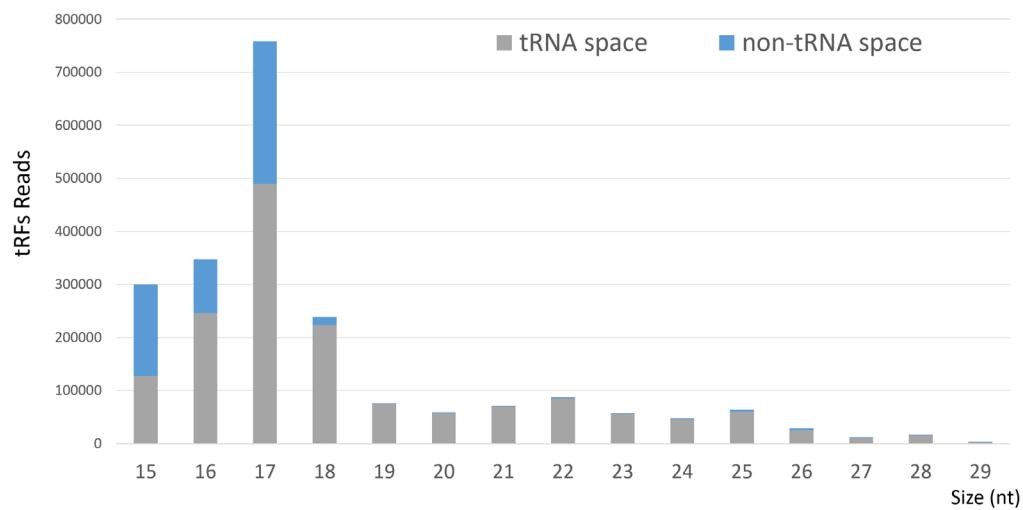
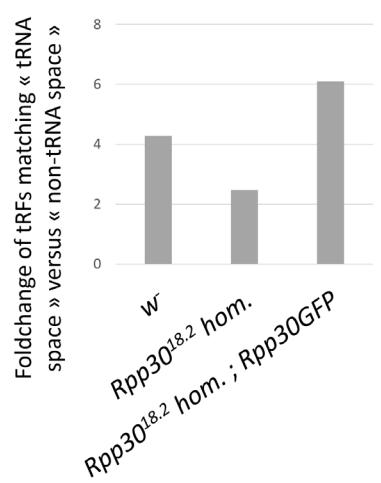
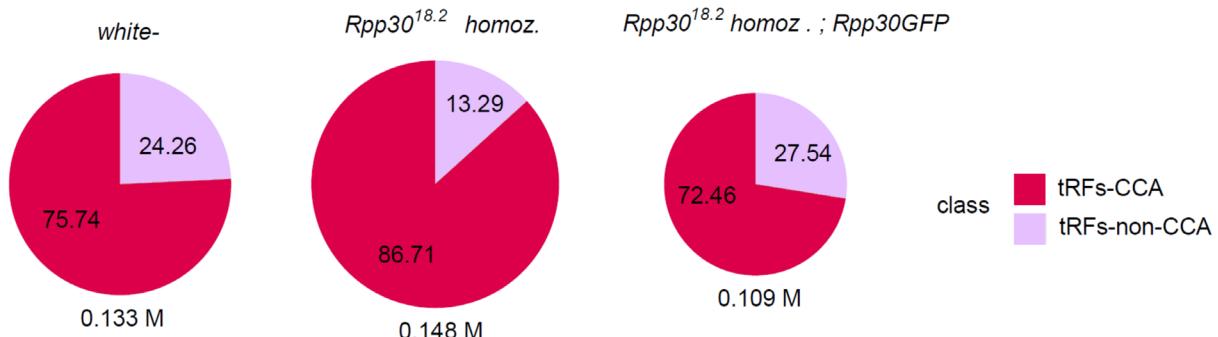
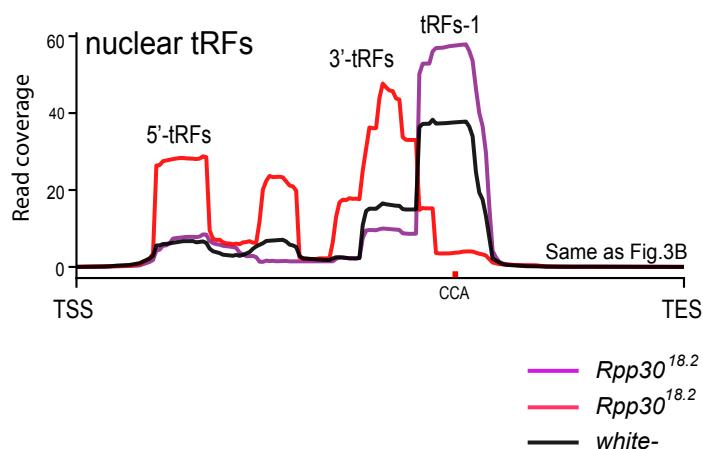
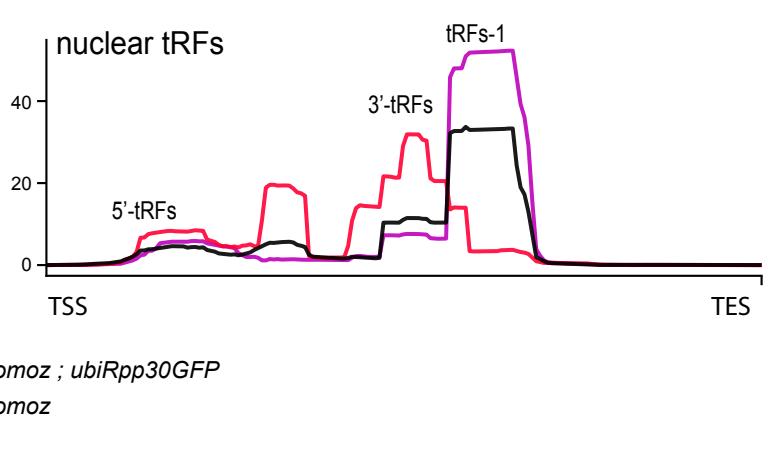
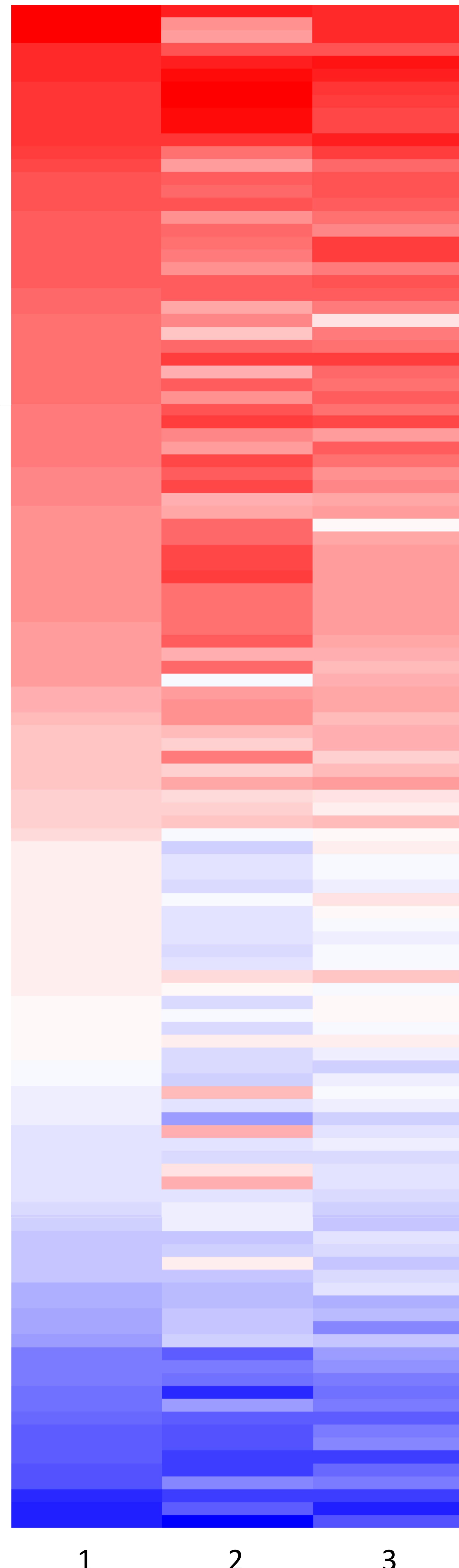


A Possible tRFs origins:**B tRFs size distribution:****C tRFs fold change****D tRFs matching to non-tRNA space****E Bam coverages tRFs populations 15-29nt****Bam coverages tRFs populations 17-29nt**

A tRNA expression heatmap

Unique tRNA mature CCA-edited seq.

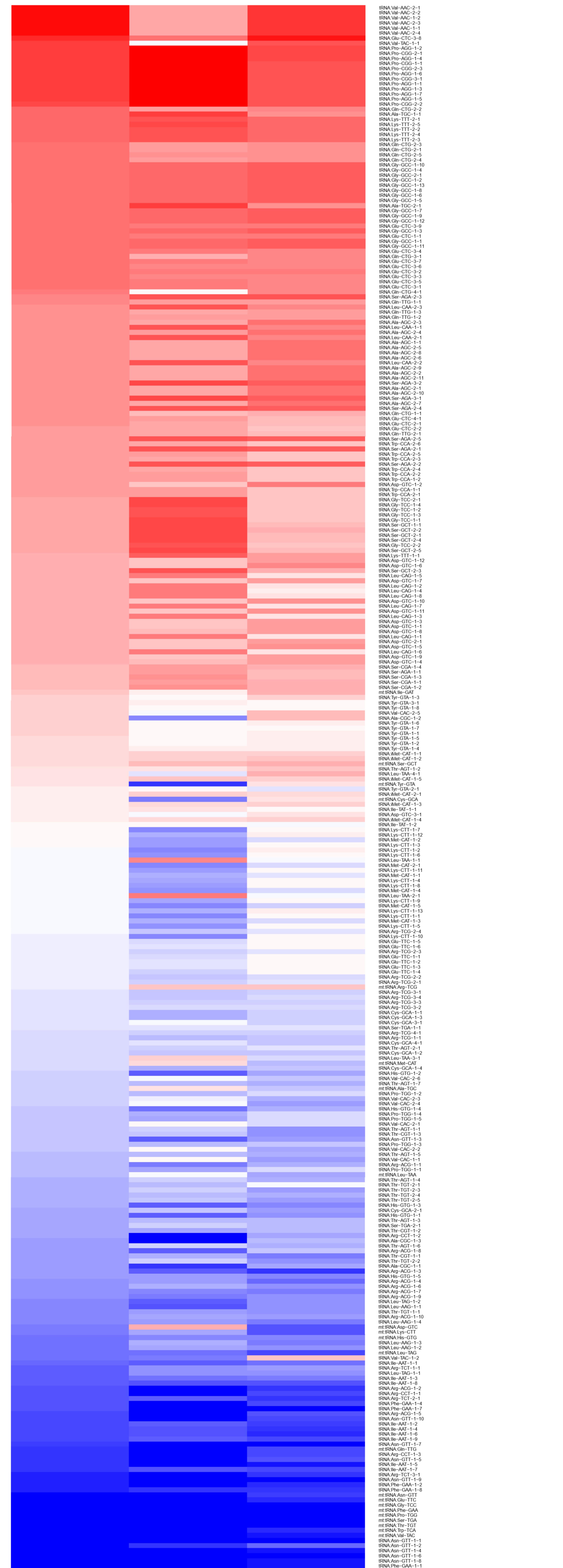
1 White⁻2 Rpp30^{18.2} hom.3 Rpp30^{18.2} hom. ; Rpp30GFP (rescue)**B Ratio of counts $Rpp30^{18.2}/w^-$**

Unique tRNA mature CCA-edited seq.

tRNA:Leu-TAA-2-1	8,96	mt:tRNA:Arg-TCG	0,95
tRNA:Leu-TAA-1-1	8,16	tRNA:Arg-TCT-1-1	0,91
mt:tRNA:Glu-TTC	7,41	tRNA:Thr-CGT-1-1	0,90
mt:tRNA:Cys-GCA	6,68	tRNA:Ile-AAT-1-1	0,84
tRNA:Thr-AGT-1-1	6,22	mt:tRNA:Thr-TGT	0,82
tRNA:Leu-TAA-3-1	5,22	tRNA:Trp-CCA-2-1	0,82
tRNA:Ser-GCT-1-1	4,59	tRNA:Ile-TAT-1-2	0,80
tRNA:Ser-GCT-2-1	4,58	mt:tRNA:Val-TAC	0,79
mt:tRNA:Lys-CTT	3,91	mt:tRNA:Gln-TTG	0,79
tRNA:Leu-TAA-4-1	3,87	mt:tRNA:Leu-TAG	0,79
tRNA:Gly-TCC-2-1	3,29	tRNA:Asp-GTC-3-1	0,78
tRNA:Gly-TCC-1-1	3,21	tRNA:Ile-TAT-1-1	0,77
tRNA:Ser-AGA-3-1	3,20	tRNA:Trp-CCA-1-1	0,75
tRNA:Ala-CGC-1-1	3,17	tRNA:Arg-ACG-1-1	0,75
tRNA:Ser-AGA-2-1	3,13	tRNA:Cys-GCA-4-1	0,72
mt:tRNA:Met-CAT	3,09	tRNA:Asn-GTT-1-1	0,68
tRNA:Pro-CGG-1-1	3,06	tRNA:Glu-CTC-1-1	0,66
mt:tRNA:Phe-GAA	3,00	tRNA:Gln-TTG-2-1	0,65
tRNA:Pro-AGG-1-1	2,98	tRNA:Glu-CTC-3-1	0,64
tRNA:Pro-CGG-3-1	2,82	mt:tRNA:Ala-TGC	0,62
tRNA:Pro-CGG-2-1	2,78	tRNA:Ala-AGC-2-1	0,59
mt:tRNA:Trp-TCA	2,69	mt:tRNA:Ala-AGC-1-1	0,57
tRNA:Ser-TGA-1-1	2,51	tRNA:Gln-TTG-1-1	0,56
tRNA:Leu-TAG-1-1	2,36	tRNA:Tyr-GTA-2-1	0,54
tRNA:Tyr-GTA-3-1	2,36	tRNA:Leu-AAG-1-1	0,54
tRNA:Leu-CAA-2-2	2,27	tRNA:Arg-TCT-2-1	0,53
tRNA:Gly-GCC-1-1	2,26	tRNA:Asp-GTC-1-1	0,52
tRNA:Lys-TTT-1-1	2,24	tRNA:Asp-GTC-2-1	0,51
tRNA:Leu-CAA-2-3	2,24	tRNA:Arg-TCG-4-1	0,50
tRNA:Ala-TGC-1-1	2,23	tRNA:Met-CAT-1-1	0,49
tRNA:Leu-CAA-2-1	2,22	tRNA:Phe-GAA-1-1	0,49
tRNA:Leu-CAA-1-1	2,20	tRNA:Tyr-GTA-1-6	0,48
tRNA:Ala-TGC-2-1	2,19	tRNA:Val-TAC-1-1	0,46
tRNA:Thr-TGT-1-1	1,99	tRNA:Gln-CTG-1-1	0,45
mt:tRNA:Asn-GTT	1,95	tRNA:Tyr-GTA-1-4	0,44
tRNA:Arg-CCT-1-1	1,93	tRNA:Arg-TCG-2-1	0,44
mt:tRNA:Pro-TGG	1,85	mt:tRNA:His-GTG	0,44
tRNA:Ser-AGA-1-1	1,66	tRNA:Arg-TCG-1-1	0,43
tRNA:Cys-GCA-2-1	1,64	tRNA:Tyr-GTA-1-3	0,43
tRNA:Ser-CGA-1-1	1,57	tRNA:Tyr-GTA-1-7	0,43
tRNA:Cys-GCA-3-1	1,56	tRNA:Arg-TCG-3-1	0,43
tRNA:Leu-CAG-1-1	1,55	tRNA:Tyr-GTA-1-8	0,42
tRNA:iMet-CAT-2-1	1,41	tRNA:Tyr-GTA-1-1	0,40
tRNA:Thr-TGT-2-1	1,39	tRNA:His-GTG-1-1	0,39
tRNA:Lys-CTT-1-1	1,29	tRNA:Tyr-GTA-1-2	0,37
mt:tRNA:Ile-GAT	1,27	tRNA:Gln-CTG-2-1	0,35
mt:tRNA:Gly-TCC	1,27	tRNA:Tyr-GTA-1-5	0,35
tRNA:Cys-GCA-1-1	1,24	tRNA:Glu-TTC-1-1	0,34
tRNA:Ser-TGA-2-1	1,21	tRNA:Pro-TGG-1-1	0,30
mt:tRNA:Asp-GTC	1,19	tRNA:SeC-TCA-1-1	0,28
tRNA:Lys-TTT-2-5	1,17	tRNA:Gln-CTG-3-1	0,26
mt:tRNA:Ser-GCT	1,16	tRNA:Val-CAC-2-1	0,20
tRNA:iMet-CAT-1-1	1,15	mt:tRNA:Leu-TAA	0,18
tRNA:Glu-CTC-2-1	1,10	tRNA:Arg-TCT-3-1	0,15
mt:tRNA:Tyr-GTA	1,10	tRNA:Val-CAC-1-1	0,15
tRNA:Lys-TTT-2-1	1,07	tRNA:Gln-CTG-4-1	0,12
tRNA:Thr-AGT-2-1	1,07	tRNA:Val-AAC-2-1	0,05
tRNA:Gly-GCC-2-1	1,00	tRNA:Val-AAC-1-1	0,04
tRNA:Met-CAT-2-1	0,97	mt:tRNA:Ser-TGA	0,00

C tRNA expression heatmap

tRNA extended CCA-edited



1

2

3

Profiles of highly expressed tRFs in control *white-* ovaries
compared to *Rpp30* mutant:

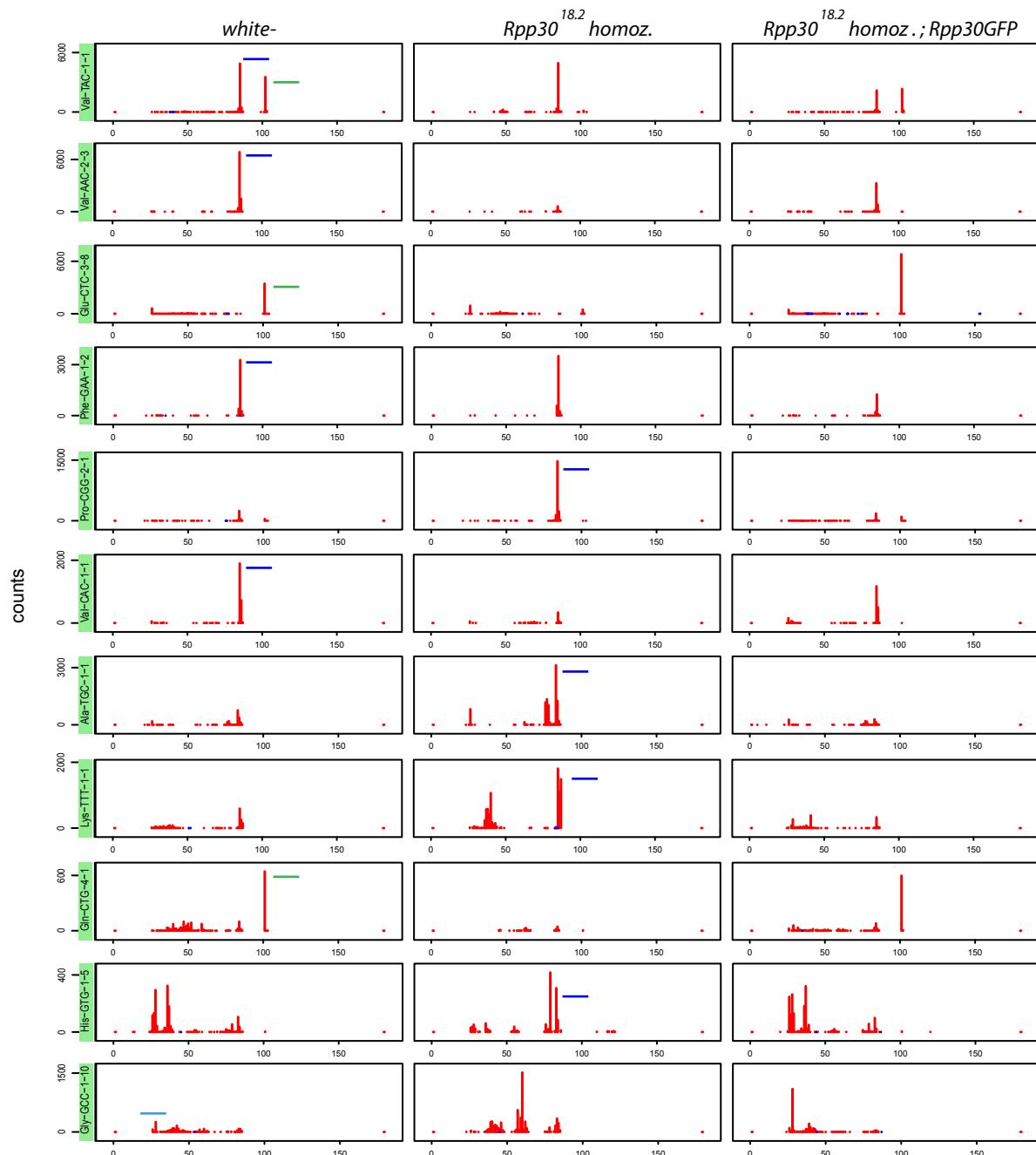
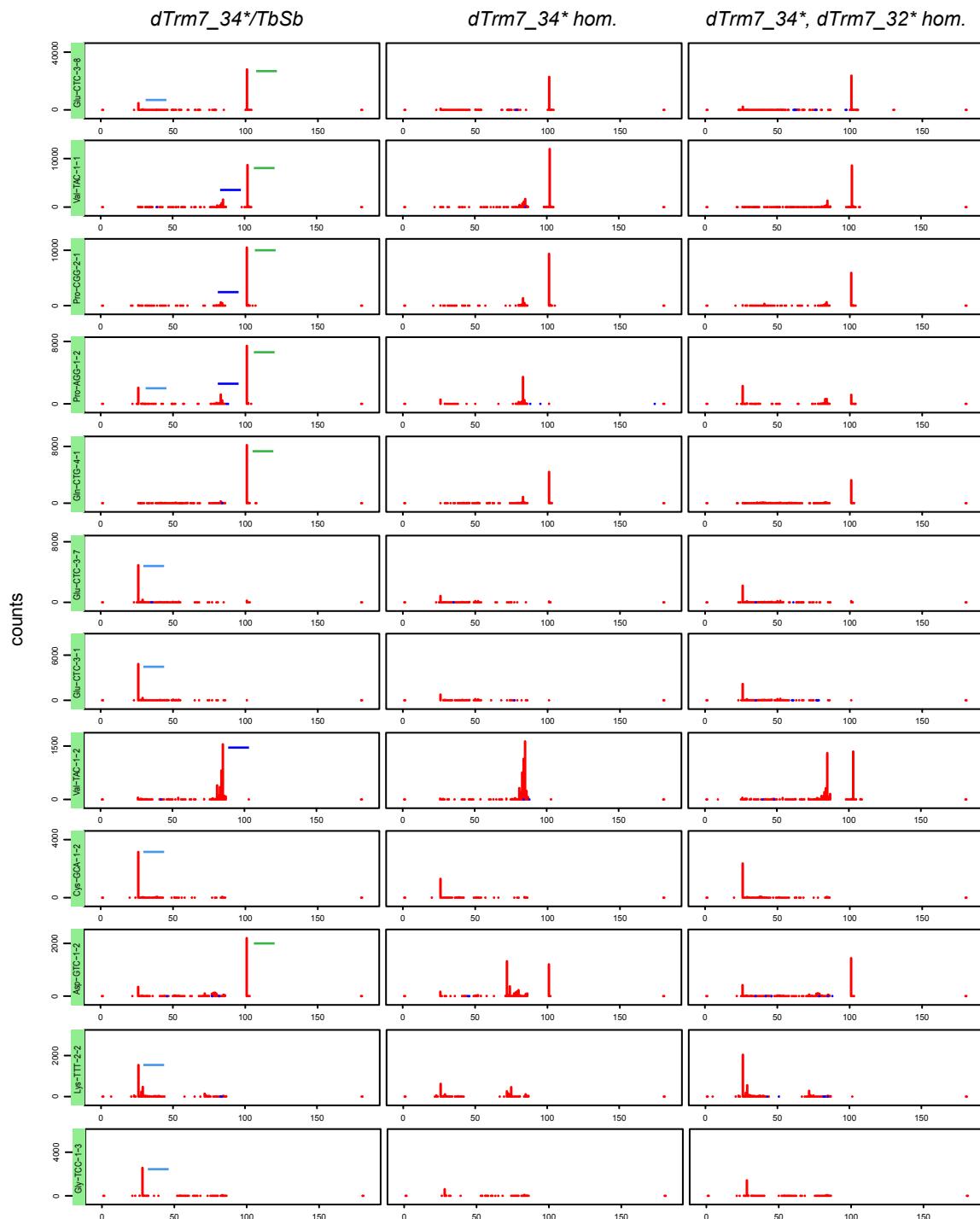


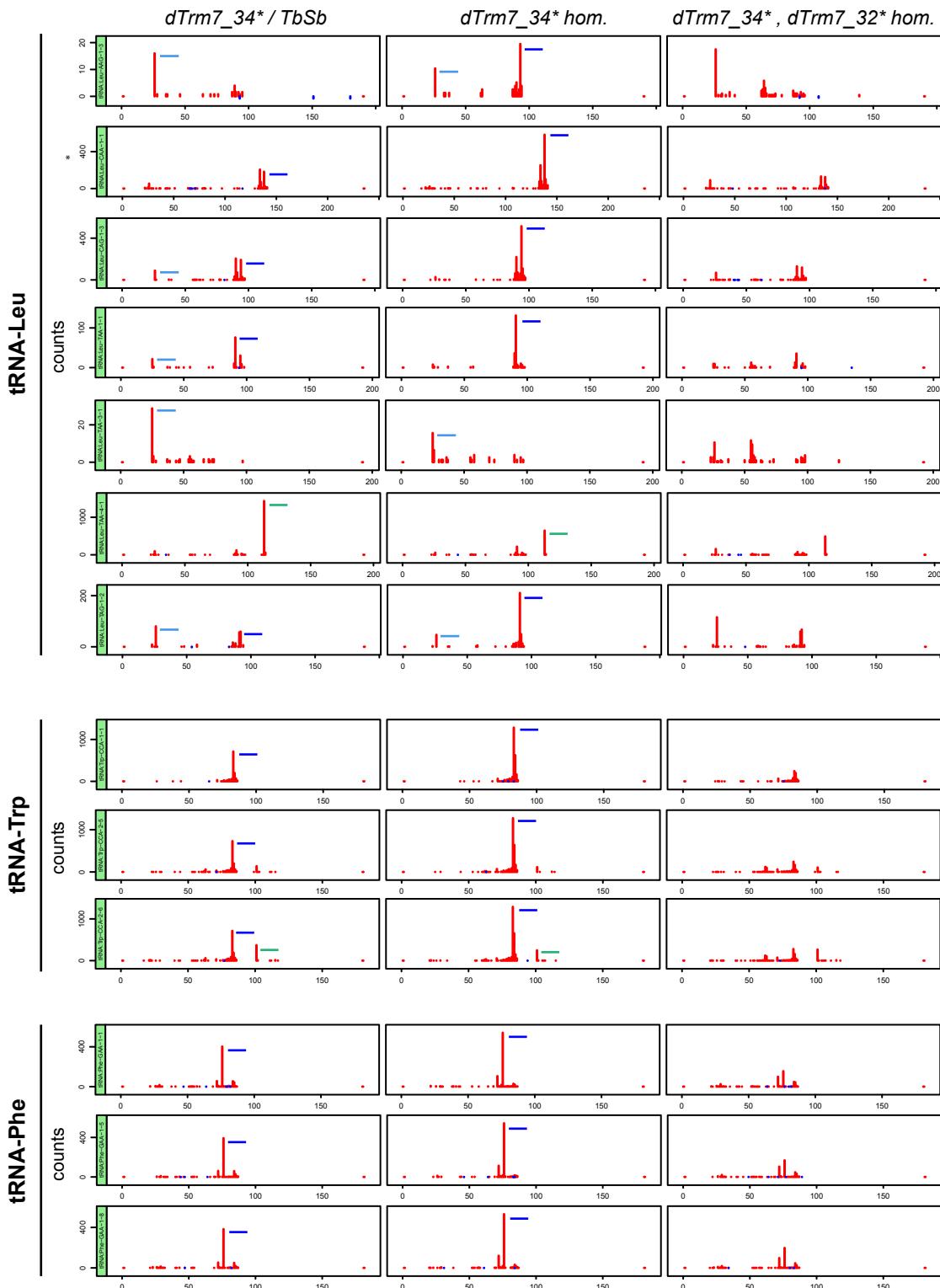
Fig.Sup.4

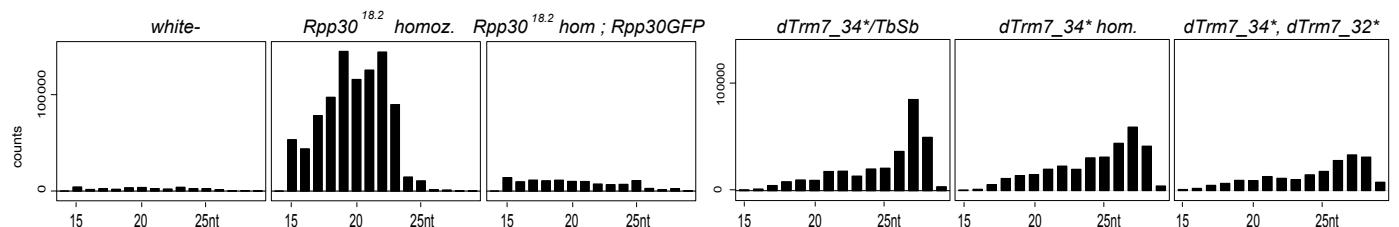
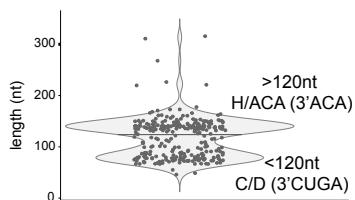
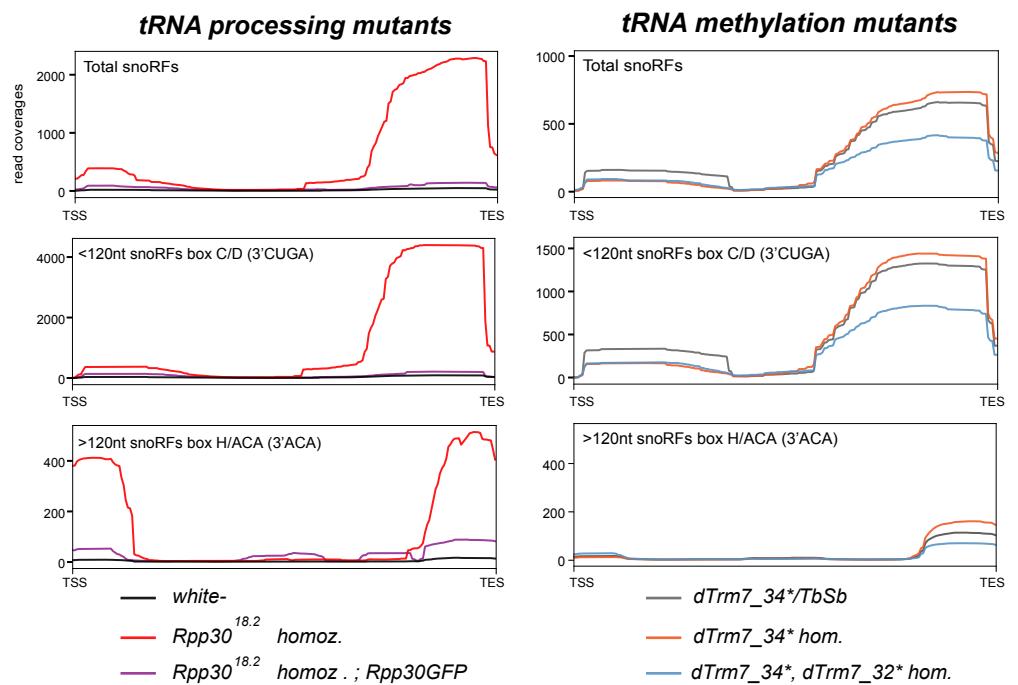
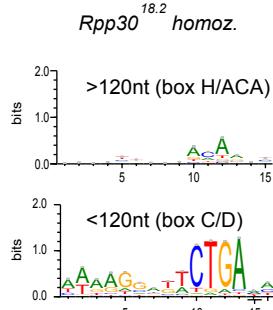


Profiles of highly expressed tRFs in control ovaries
compared to tRNA methylation mutants:



tRFs profiles of tRNAs methylated by dTrm7_34 and dTrm7_32:



A snoRFs size distributions:**B snoRNA length:****C snoRNA fragments (snoRFs) coverage:****D Logo last 15nt snoRFs**

A**miRNA normalization:**

- small RNA reads input fasta files
- miRcounts Counts miRNA alignments from small RNA sequence data (Galaxy Version 1.3.2)
- Column Join on Collections (Galaxy Version 0.0.3) of miRcounts tabular files
- DESeq2 normalization of hit lists (geometric method) (Galaxy Version 1.0.1)
- Compute an expression on every row (Galaxy Version 1.2.0)

**B****normalization factors:**

Genotype	NF= Norm. factors	1/NF
<i>Rpp30</i> ^{18.2} , <i>mnk</i> ^{P6} homoz	0.50	2.00
<i>Rpp30</i> ^{18.2} homoz.; <i>Rpp30GFP</i>	2.39	0.42
<i>Rpp30</i> ^{18.2} / <i>Rpp30</i> ^{PE}	1.23	0.81
<i>Rpp30</i> ^{18.2} homoz.	0.43	2.33
w-	1.76	0.57
<i>dTrm7_34*/TbSb</i>	1.26	0.80
<i>dTrm7_34*</i> homoz.	0.77	1.30
<i>dTrm7_34*</i> , <i>dTrm7_32*</i> hom.	1.89	0.53
<i>dTrm7_34*/Def9487</i>	0.56	1.80

C**Annotate datasets:**

- small RNA reads input fasta files deleted from rRNA.
- Annotate smRNA dataset by iterative alignments with sRbowtie (Galaxy Version 2.4.0).
- 0 mismatch allowed.
- Alignment steps: in the following order tRNA, tRNA-CCA-edited, miRNA, TE-derived, all-ncRNA, all genes, all intergenic.
- Generation of a cascade annotation analysis and a barplot.



A**tRNA fragments (tRFs) Bam Coverage:**

tRFs input fasta files

sRBowtie (for small RNA short reads (Galaxy Version 2.1.1)
Matched on DNA, multiple mappers, randomly matched at a single position
0MM
Ref: tRNA-CCA-edited-extended

BamCoverage generates a coverage bigWig file from a given BAM or CRAM file (Galaxy Version 3.1.2.0.0)
Scale factors were used for each genotype.

ComputeMatrix prepares data for plotting a heatmap or a profile of given regions (Galaxy Version 3.1.2.0.0)
Regions to plot: Nuclear tRNAs (all, 5' or 3') or Mitoch. tRNAs Bed files.

↓ plotProfile creates a profile plot for score distributions across genomic regions (Galaxy Version 3.1.2.0.0)

B**tRF Expression workflow:**

tRF collection list

sRBowtie (for small RNA short reads (Galaxy Version 2.1.1)
Matched on DNA, multiple mappers, randomly matched at a single position
0MM
Ref: tRNA-CCA-edited

Parse items in sRbowtie alignment (Galaxy Version 1.0.6)

DESeq2 normalization of hit lists (geometric method) (Galaxy Version 1.0.1)

Cut columns from a table (Galaxy Version 1.0.2)

Sort data in ascending or descending order (Galaxy Version 1.1.1)

Plot heatmap with high number of rows (Galaxy Version 1.0.0).

Data transformation: Log2(value+1)

Cut columns from a table (Galaxy Version 1.0.2)

Compute an expression on every row (Galaxy Version 1.2.0)

C**snoRNA fragment (snoRF) Bam Coverage:**

snoRNA input fasta files

sRBowtie (for small RNA short reads (Galaxy Version 2.1.1)
Matched on DNA, multiple mappers, randomly matched at a single position
0MM
Ref: snoRNA

BamCoverage generates a coverage bigWig file from a given BAM or CRAM file (Galaxy Version 3.1.2.0.0)
Scale factors were used for different genotypes.

ComputeMatrix prepares data for plotting a heatmap or a profile of given regions (Galaxy Version 3.1.2.0.0)
Regions to plot:
snoRNA Bed description file, or snoRNA>120nt or snoRNA<120nt.

↓ plotProfile creates a profile plot for score distributions across genomic regions (Galaxy Version 3.1.2.0.0)