

Overlapping transcripts - RPKM NAN

Dear Sammeth,

I run flux-capacitor in the following options: flux-capacitor -i bamfile -a annotation_file -o ouput --threads nthreads.

When I run it with an gff annotation file that have some transcripts that overlap each other, but not exons, such as I show in the following example with 3 transcripts or in the attached image [dmel.overlap.jpg](#).

```
2L    selenoprofiles_exonerate    exon    4939284    4939788    .    +    .    transcript_id
"FBtr0304686.74.homologue"; gene_id "FBgn0304686.74.homologue";

2L    selenoprofiles_exonerate    exon    4891851    4892080    .    +    .    transcript_id
"FBtr0304686.74.homologue"; gene_id "FBgn0304686.74.homologue";

2L    selenoprofiles_genewise     exon    4893955    4893994    .    +    .    transcript_id
"FBtr0077391.1.homologue"; gene_id "FBgn0077391.1.homologue";

2L    selenoprofiles_genewise     exon    4901402    4901917    .    +    .    transcript_id
"FBtr0077391.1.homologue"; gene_id "FBgn0077391.1.homologue";

2L    selenoprofiles_genewise     exon    4902025    4902395    .    +    .    transcript_id
"FBtr0077391.1.homologue"; gene_id "FBgn0077391.1.homologue";

2L    selenoprofiles_genewise     exon    4902461    4902593    .    +    .    transcript_id
"FBtr0077391.1.homologue"; gene_id "FBgn0077391.1.homologue";

2L    selenoprofiles_genewise     exon    4902664    4902905    .    +    .    transcript_id
"FBtr0077391.1.homologue"; gene_id "FBgn0077391.1.homologue";

2L    selenoprofiles_genewise     exon    4902974    4903150    .    +    .    transcript_id
"FBtr0077391.1.homologue"; gene_id "FBgn0077391.1.homologue";

2L    selenoprofiles_genewise     exon    4937441    4937468    .    +    .    transcript_id
"FBtr0077393.27.homologue"; gene_id "FBgn0077393.27.homologue";

2L    selenoprofiles_genewise     exon    4937534    4938182    .    +    .    transcript_id
"FBtr0077393.27.homologue"; gene_id "FBgn0077393.27.homologue";

2L    selenoprofiles_genewise     exon    4938324    4938866    .    +    .    transcript_id
"FBtr0077393.27.homologue"; gene_id "FBgn0077393.27.homologue";

2L    selenoprofiles_genewise     exon    4940964    4941363    .    +    .    transcript_id
"FBtr0077393.27.homologue"; gene_id "FBgn0077393.27.homologue";
```

The ouput results obtained for this example are the following:

```
2L    selenoprofiles_genewise     transcript 4893955 4903153 . + . transcript_id "FBtr0077391.1.homologue"; locus_id "2L:
4891851-4941366W"; gene_id "FBgn0077391.1.homologue"; reads NaN; length 1482; RPKM NaN

2L    selenoprofiles_genewise     transcript 4937441 4941366 . + . transcript_id "FBtr0077393.27.homologue"; locus_id
"2L:4891851-4941366W"; gene_id "FBgn0077393.27.homologue"; reads NaN; length 1623; RPKM NaN

2L    selenoprofiles_exonerate     transcript 4891851 4939788 . + . transcript_id "FBtr0304686.74.homologue"; locus_id
"2L:4891851-4941366W"; gene_id "FBgn0304686.74.homologue"; reads NaN; length 735; RPKM NaN
```

As it can be seen the reads and RPKM values are inconsistent. Some parts of the ouput of the execution are shown here. It can be seen that are some errors and warnings that are responsible for these NAN values.

```
PROFILE] Scanning the input and getting the attributes.

profiling OK (00:23:52)

first round finished .. took 1432 sec.

12325 single transcript loci

377309560 mappings in file

43276718 mappings fall in single transcript loci

4615251872 mappings in annotation-mapped pairs
```

```
81,121 min/max read length

[SOLVE] Deconvolving reads of overlapping transcripts.

decomposing [WARN] Fraction inconsistency FBtr0077393.27.homologue -2.304722170272954

...

[ERROR] Failed to set lp output to:
null
java.lang.NullPointerException
  at java.io.FileOutputStream.<init>(FileOutputStream.java:206)
  at java.io.FileOutputStream.<init>(FileOutputStream.java:136)
  at barna.flux.capacitor.reconstruction.GraphLPSolver.run(GraphLPSolver.java:950)
  at barna.flux.capacitor.reconstruction.FluxCapacitor$LocusSolver.call(FluxCapacitor.java:1097)
  at barna.flux.capacitor.reconstruction.FluxCapacitor.explore(FluxCapacitor.java:2631)
  at barna.flux.capacitor.reconstruction.FluxCapacitor.call(FluxCapacitor.java:1886)
  at barna.flux.capacitor.reconstruction.FluxCapacitor.call(FluxCapacitor.java:74)
  at barna.commons.launcher.Flux.main(Flux.java:198)

unusual normalization factor: 0.03384014257154664
[WARN] Unsolved system: 2L:4891851-4941363W
[WARN] Fraction inconsistency FBtr0302984.1.homologue -2.313103458721854
Model name: FBtr0080341.1.homologue
```

Is there a solution for this overlapping case?

Thank you.

Cheers,

Miquel [dmel.overlap.jpg](#)