

Supplementary materials

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1 Supplementary methods

1.1 Bioinformatics workflow

1.1.1 Schema

1.1.2 Alignment, filtering and count: Pipeline single-meiosis.sh

Tetrad's analysis was carried out by applying the overall strategy proposed by Anderson (2011) and implemented in the single-meiosis.sh pipeline available at .

- For each tetrads, sequences from the four individuals were mapped against a merged reference genome containing both the Columbia (TAIR10) and Landsberg (Zapata et al. (2016)) sequences. BWA (Li and Durbin (2009)) was used to align sequences with a set of parameters positioned to neither allow mismatches (or gap) nor multiple hits.
- Alignment were filtered out to remove non specific hit and PCR duplicates. Such parameters ensure only the maintenance of reads that are specific to one of the parental genomes and also specific to a single locus This parameters discards reads that span conversion but allowing mismatches introduced two much noises in the results. This settings was the best compromise between specificity and sensitivity.
- Each alignment file was then separated by parental genomes: Col-0 and Ler-0 to compute the number of reads covering each genome position thanks to the samtools mpileup programm.

1.1.3 Cross with SNV: variantutils R functions [author: Charif D.]

VCF files and SNPs list (Zapata et al., 2016) were cross referenced and used as input for the HMM

1.1.4 Individuals and Tetrad genotyping: HMM_model R functions [author: Robin S.]

An HMM model was implemented by S. Robin to obtain both the individuals and tetrad genotypes at each SNV position from files that compile the number of reads at each position for each individual in the tetrad (M1, M2, M3, M4).

Notations:

- $I = 4$ individuals forming a tetrade ($i = 1 \dots I$);
- $C = 5$ chromosomes per individuals ($c = 1 \dots C$);
- T markers (T_c markers in chromosomes c : $\sum_c n_c = n$, $t = 1 \dots n_c$);
- R_{ict} = number of reads mapped onto marker t in the Col + Ler genome from chromosome c in individual i ;
- Y_{ict} = number of reads mapped onto marker t in the Col genome from chromosome c in individual i .

1.1.4.1 Individual level: Because of the experimental setting, at each marker, the ratio Y_{it}/R_{it} is expected to be close to either:

- 1/2 (heterozygous genotype Col/Ler) or
- 1 (homozygous genotype Col)

This suggest a model with $K = 2$ hidden states with respective emission distributions:

- $Y_{it} \sim \mathcal{B}(R_{it}, \gamma_1)$, where $\gamma_1 \simeq 1/2$;
- $Y_{it} \sim \mathcal{B}(R_{it}, \gamma_2)$, where $\gamma_2 \simeq 1$.

The first goal is to recover for each individual at each marker the hidden state

$$Z_{it} \in \{1, 2\}$$

1.1.4.2 Tetrade level Tetrad genotype is determined by the states of the four individuals at a given marker. Therefore, we consider the joint hidden state $S_t \in \{1, \dots, K^I\} = \{1, \dots, 16\}$ for the whole tetrade at position t , which is related to the four individual hidden states Z_{it} according to Table. 1. In the same table H_t is the number of heterozygous individuals at marker t . These states can be interpreted as follows:

- States $S_t = 1$ and 16 ($H_t = 4$ and 0) are expected to be absent;
- States $S_t = 4, 6, 7, 10, 11$ and 13 ($H_t = 2$) are expected to be predominant;
- States $S_t = 2, 3, 5, 8, 9, 12, 14$ and 15 ($H_t = 1$ or 3) correspond to NCO.

| S_t | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 |
|----------|---|---|---|---|---|---|---|---|---|----|----|----|----|----|----|----|
| Z_{1t} | 1 | 2 | 1 | 2 | 1 | 2 | 1 | 2 | 1 | 2 | 1 | 2 | 1 | 2 | 1 | 2 |
| Z_{2t} | 1 | 1 | 2 | 2 | 1 | 1 | 2 | 2 | 1 | 1 | 2 | 2 | 1 | 1 | 2 | 2 |
| Z_{3t} | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 2 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 2 |
| Z_{4t} | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 |
| H_t | 4 | 3 | 3 | 2 | 3 | 2 | 2 | 1 | 3 | 2 | 2 | 2 | 1 | 2 | 1 | 1 |

Table 1: Correspondence between the tetrade hidden state S_t and each of the four individual hidden states Z_{it} , as defined by the R instruction: `expand.grid((1:2),(1:2),(1:2),(1:2))`.

We used the hidden Markov model (HMM) framework where:

- the hidden states Z_{it} or S_t are supposed to form a Markov chain and where
- the observations Y_{it} are supposed to be independent conditionally on the hidden states with distribution controlled by the corresponding hidden state.

1.1.4.3 HMM parameters

- The hidden states of each individual have been considered jointly. Both the success probabilities (γ_1, γ_2) and the transition rates (λ, μ) are common to all individuals, resulting in:

$$\hat{\gamma}_1, \hat{\gamma}_2, \hat{\lambda}, \hat{\mu} \rightarrow 4 \text{ parameters.}$$

- To take into account the non homogeneous distance separating the marker, we allowed heterogeneous transition probabilities (continuous time Markov chain). The frequencies of changes from one state to another are given by transition rates

$$\lambda_{k\ell} : k \rightarrow \ell.$$

In the binary case ($Z \in \{1, 2\}$), denoting

$$\lambda : 1 \rightarrow 2, \quad \mu : 2 \rightarrow 1,$$

1.1.4.4 Classification: The inference on the hidden states is carried based on their conditional distribution given the observations $Y = (Y_{it})$, that is

$$p_{\hat{\theta}}(Z|Y).$$

The classification (that is the inference of the hidden state) had been made marker by marker. For each individual and each marker, we obtained the $\{posterior\ probability\}$ provided by the EM alorithm.

$$\tau_{itk} = P_{\hat{\theta}}\{Z_{it} = k|Y\}.$$

A natural classification rule consists in using the $\{maximum\ a\ posteriori\}$ (MAP) rule:

$$MAP_{it} = \arg \max_k \tau_{itk}.$$

1.1.4.5 Results:

- Chromosome
- Position
- MAP
- $-\log_{10}(1 - MAP_{it})$
- MAP classification

1.1.5 COs detection and classification: crossover programm

Thanks to the marker genotype obtained by the HMM model, we then used the crossover programm Anderson (2011) to detect and classify the CO event. To be considered as 2 independent COs, the SNPs involved had to be more than 5 kilobases apart

1.1.6 NCOs detection:

NCOs correspond to markers with the following states $St = \{2, 3, 5, 8, 9, 12, 14, 15\}$. Contiguous markers with a genotype corresponding to only of this states and less than 2 kb apart were considered as a single event belong to the same event.

1.1.7 Tetrads analysis iteration

1. A first analysis of all tetrads was done considering all of the SNV markers from Zapata et al. (2016) (All_SNV=545481)
2. SNVs where then filtered according to the followig criterions:
 - Removing of SNVs associated to COs that are found in several tetrads (file Liste_FAUX_CO.txt)
 - Removing of markers with a non 2:2 segregation in most of the tetrads (fichier df_Genotype_SNP_count.txt)
3. Re-analysis of all Tetrads with a reducing list of SNV (Gold_Variants=522658).
4. Sensibility and specificity analysis (?)

1.1.8 Gold variants, CO and NCO annotation

SNP have been annotated thanks to TxDb.Athaliana.BioMart.plantsmart28 database and the locateVariants function from the VariantAnnotation package: Coding, Intron, FiveUTR, ThreeUTR, Promoter up to 1000 bases.

Regarding, heterochromatine, centromere and transposable elements, we used a bed file (Giraut (2011))

1.1.9 Track Length estimation

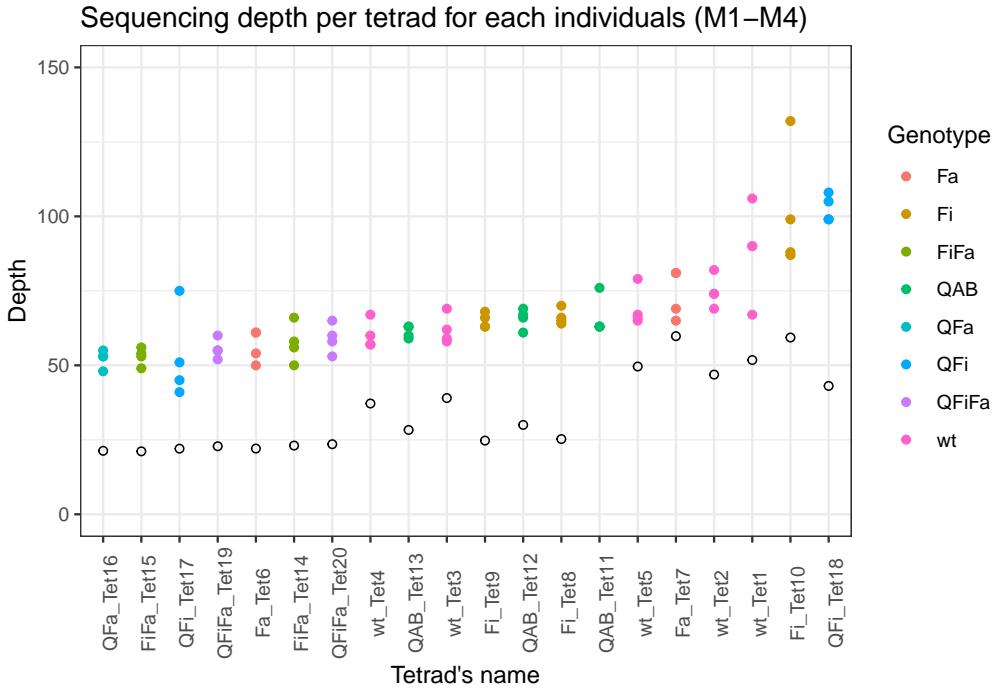
- median TL: Each end of the track is defined as the midpoint between the two converted SNPs with opposite genotypes. Tract length is found by calculating the distance between these two midpoints
- min TL: between the last converted SNPs
- max TL: between the 2 flanking SNPs

1.1.10 Detection power

- For each window size (20,50,75,100,200,300,400,500,600,700, 800,900,1000,1200,1500,2000,2500,3000, 4000,5000,8000,10000)
 - 10000 sequences of this length were sampled from the genome.
 - We computed the following statistics
 - * mean converted SNP
 - * number of window with at least one converted SNP

2 Supplementary Results

2.1 Overview of sequencing data:



```
## 
##   Fa    Fi   FiFa    QAB    QFa    QFi  QFiFa     wt
##   2     3     2     3     1     2     2       5
```

2.2 Sensibility and specificity of the overall strategy

2.2.1 The Score is highly correlated to the tetrads sequencing depth

The score is positively correlated to the rate of true positive (TP) NCO.

The more the tetrad's depth is important, the less the number of event to check is high and the highest is the rate of TP NCO.

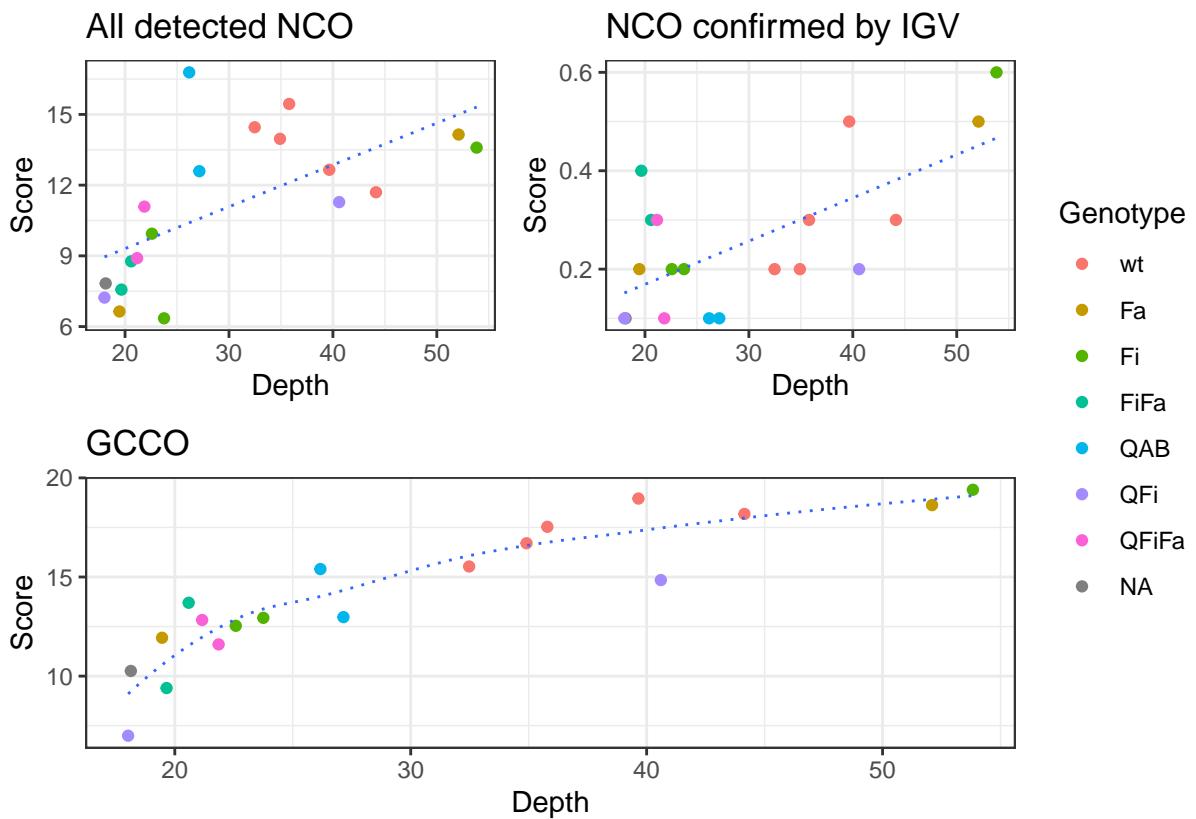
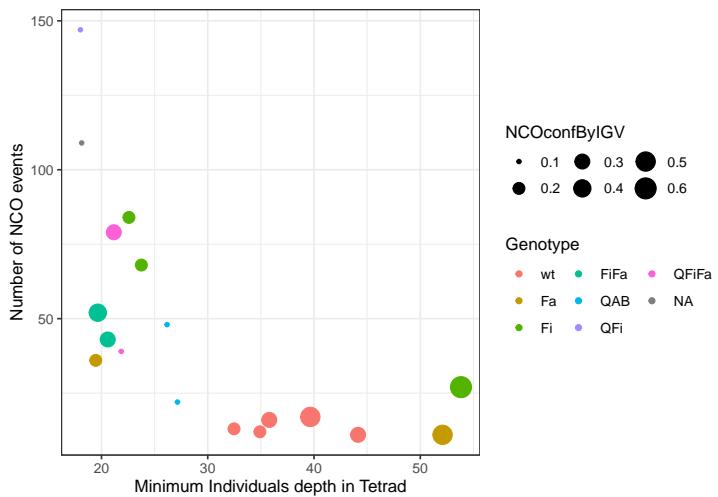


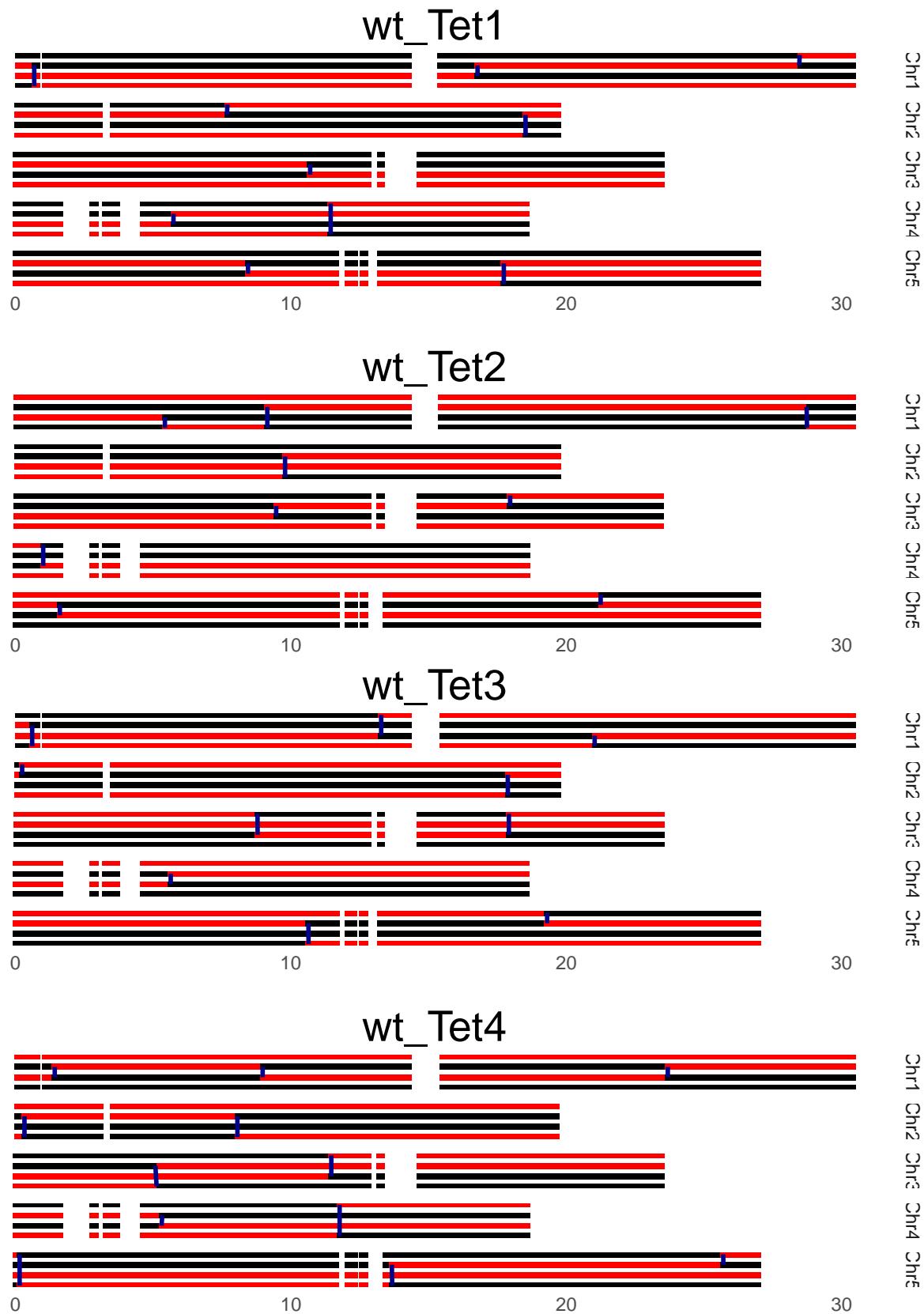
Figure 1: Supp Fig 2



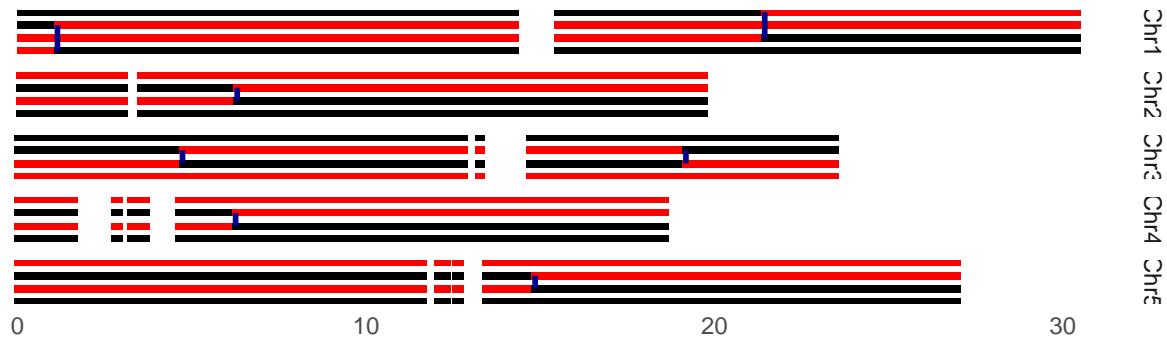
2.3 Gold SNPs descriptiv statistics:

```
## # A tibble: 5 x 4
##   Chr     max   mean median
##   <chr> <int> <dbl>  <dbl>
## 1 Chr1    996316 220.    43
## 2 Chr2    459751 208.    50
## 3 Chr3   1376607 233.    48
## 4 Chr4   1170202 231.    45
## 5 Chr5    519179 204.    43
```

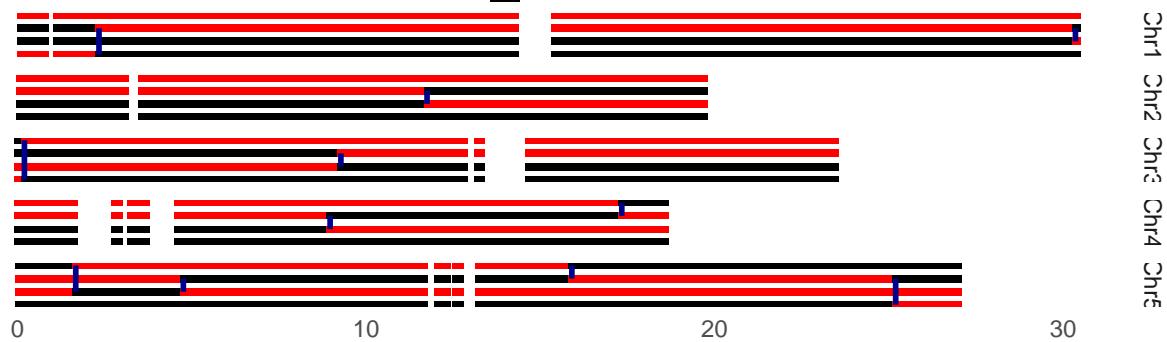
2.4 Tetrads Genotypes:



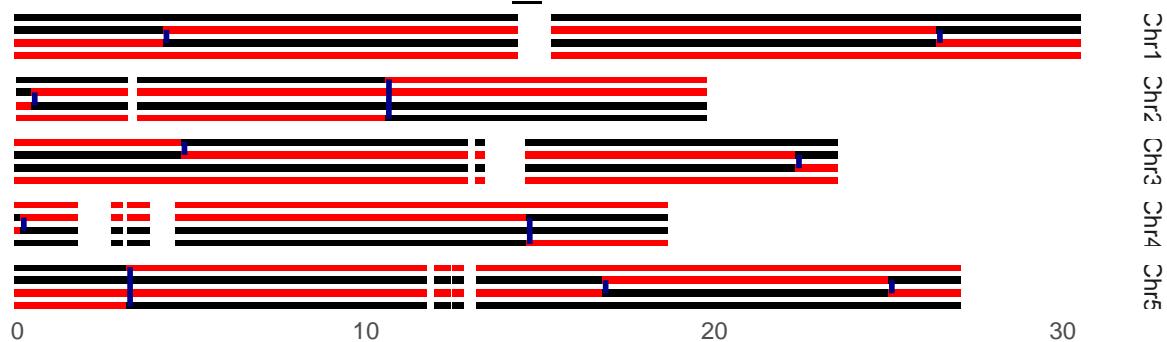
wt_Tet5



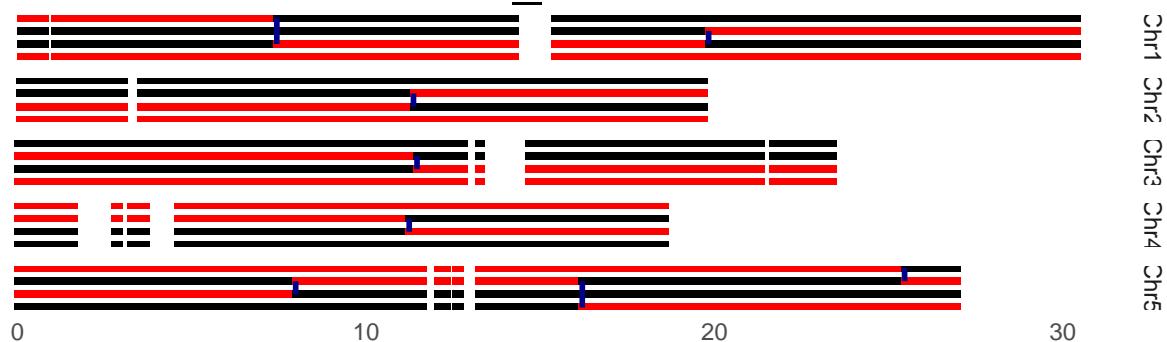
Fi_Tet10



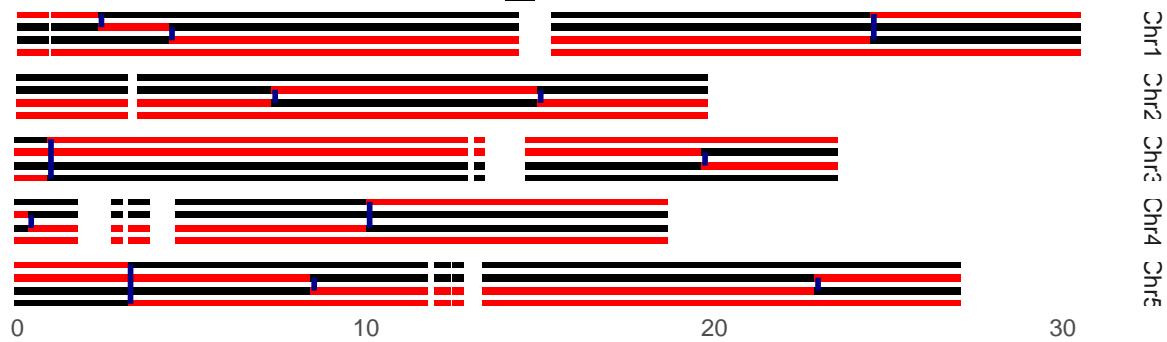
Fa_Tet6



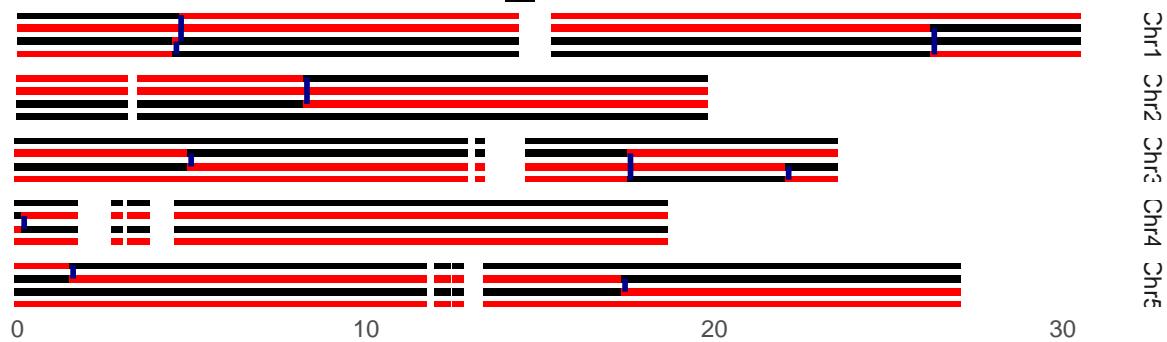
Fa_Tet7



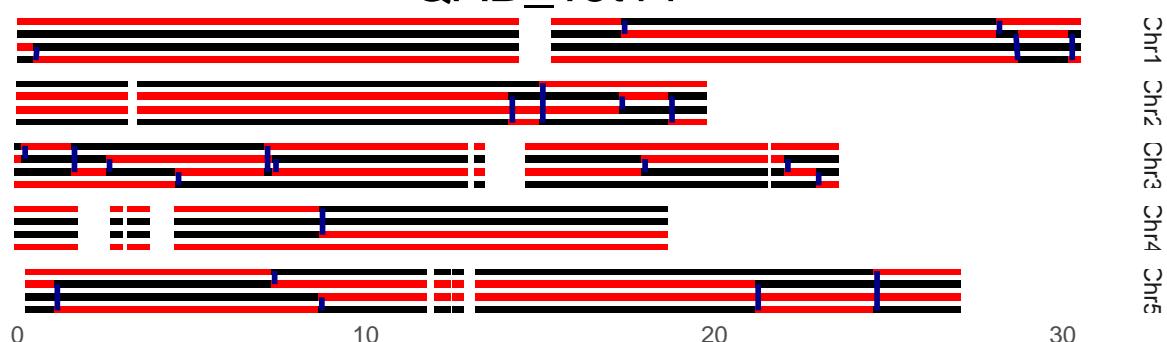
Fi_Tet8



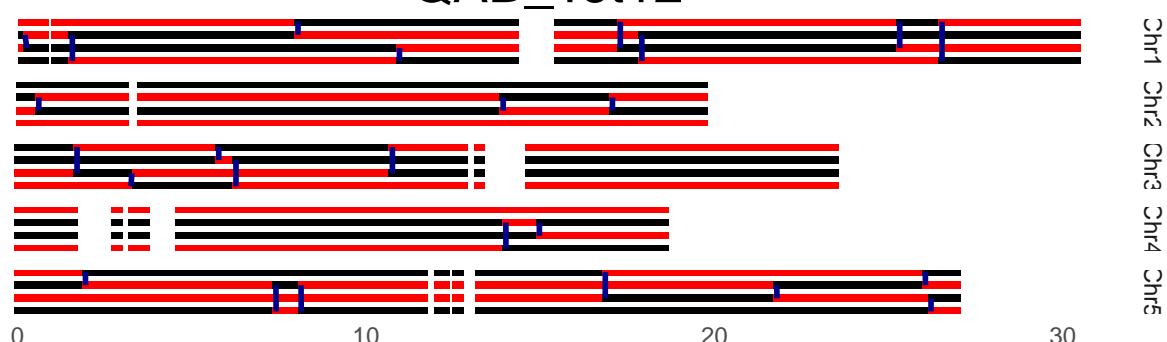
Fi_Tet9



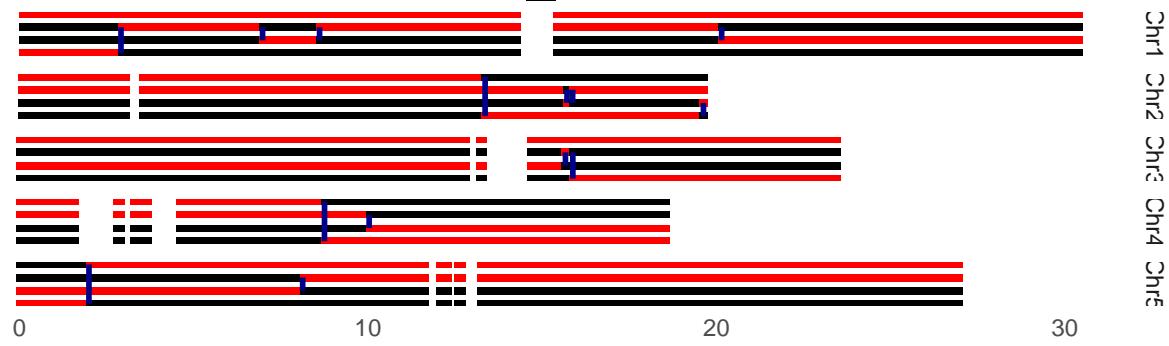
QAB_Tet11



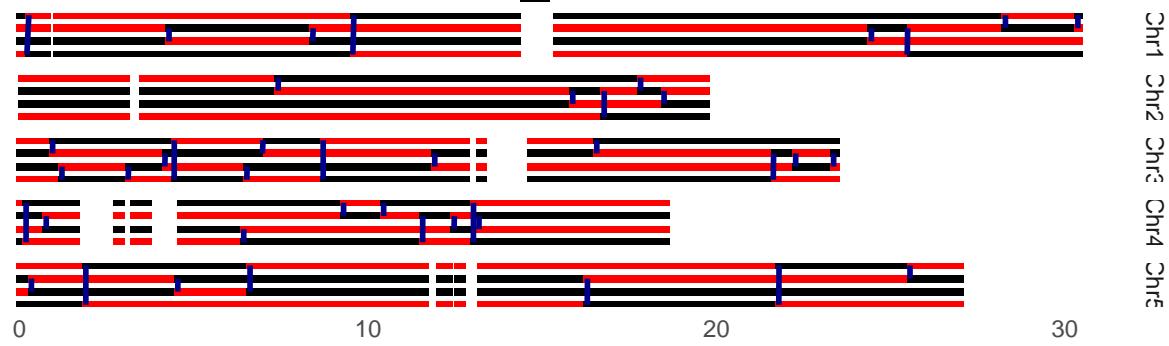
QAB_Tet12



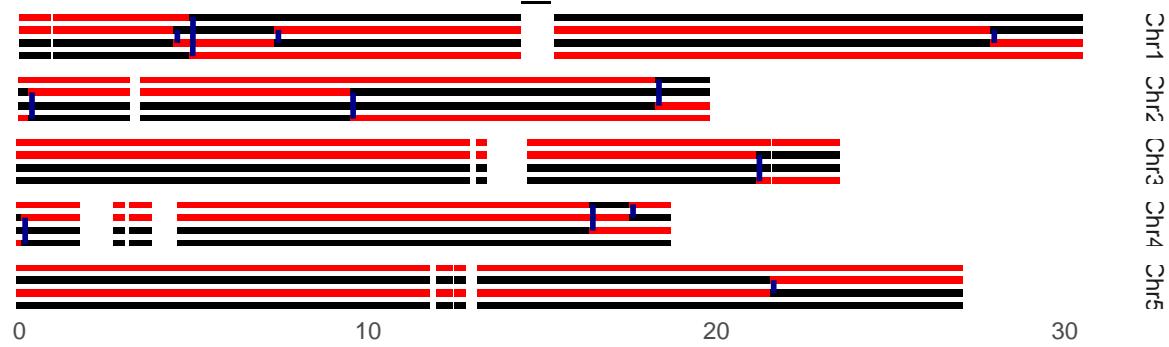
QAB_Tet13



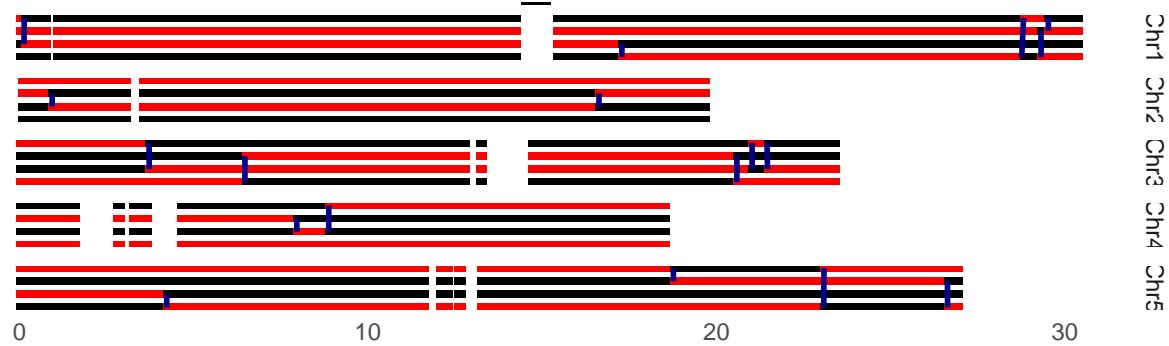
QFa_Tet16



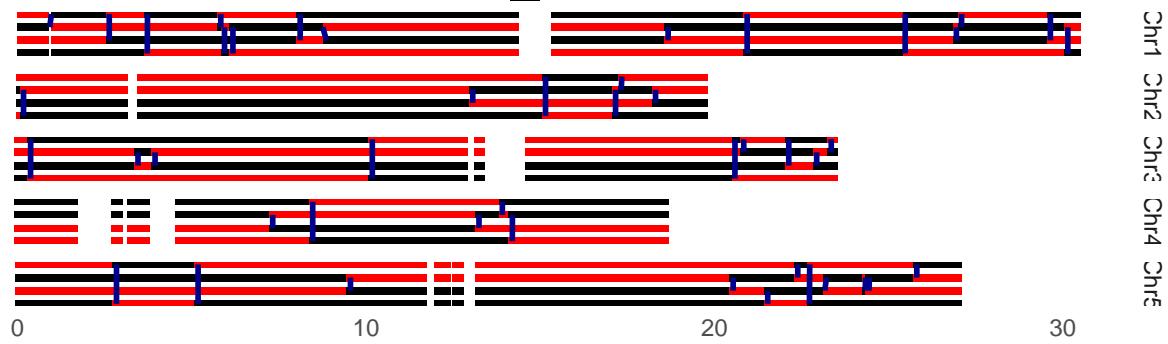
FiFa_Tet14



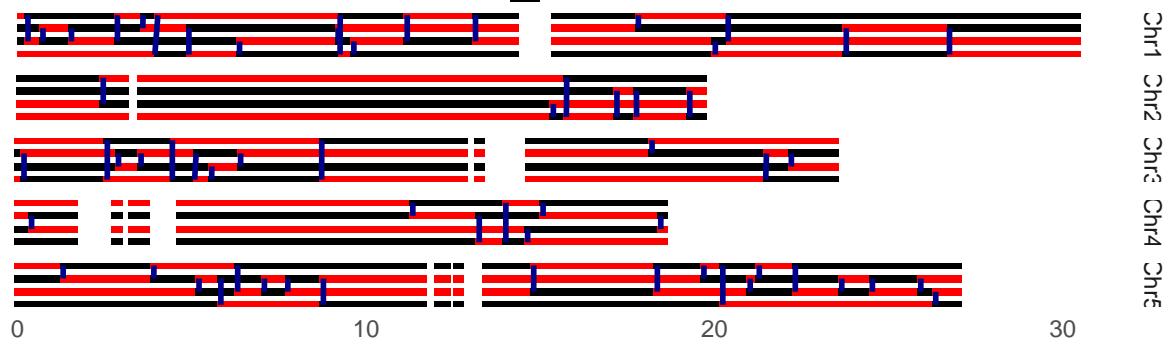
FiFa_Tet15



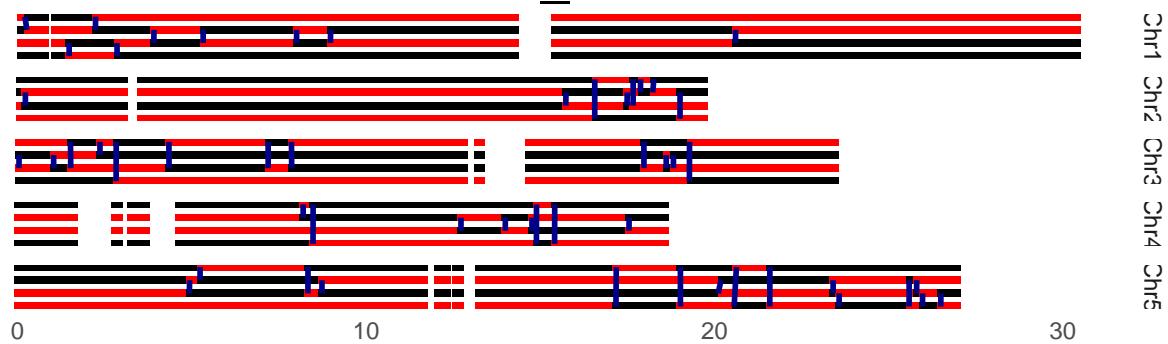
QFi_Tet17



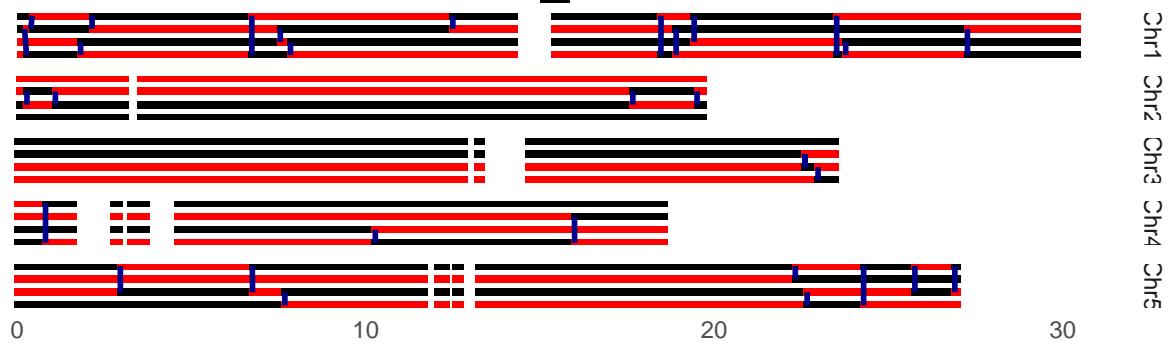
QFi_Tet18



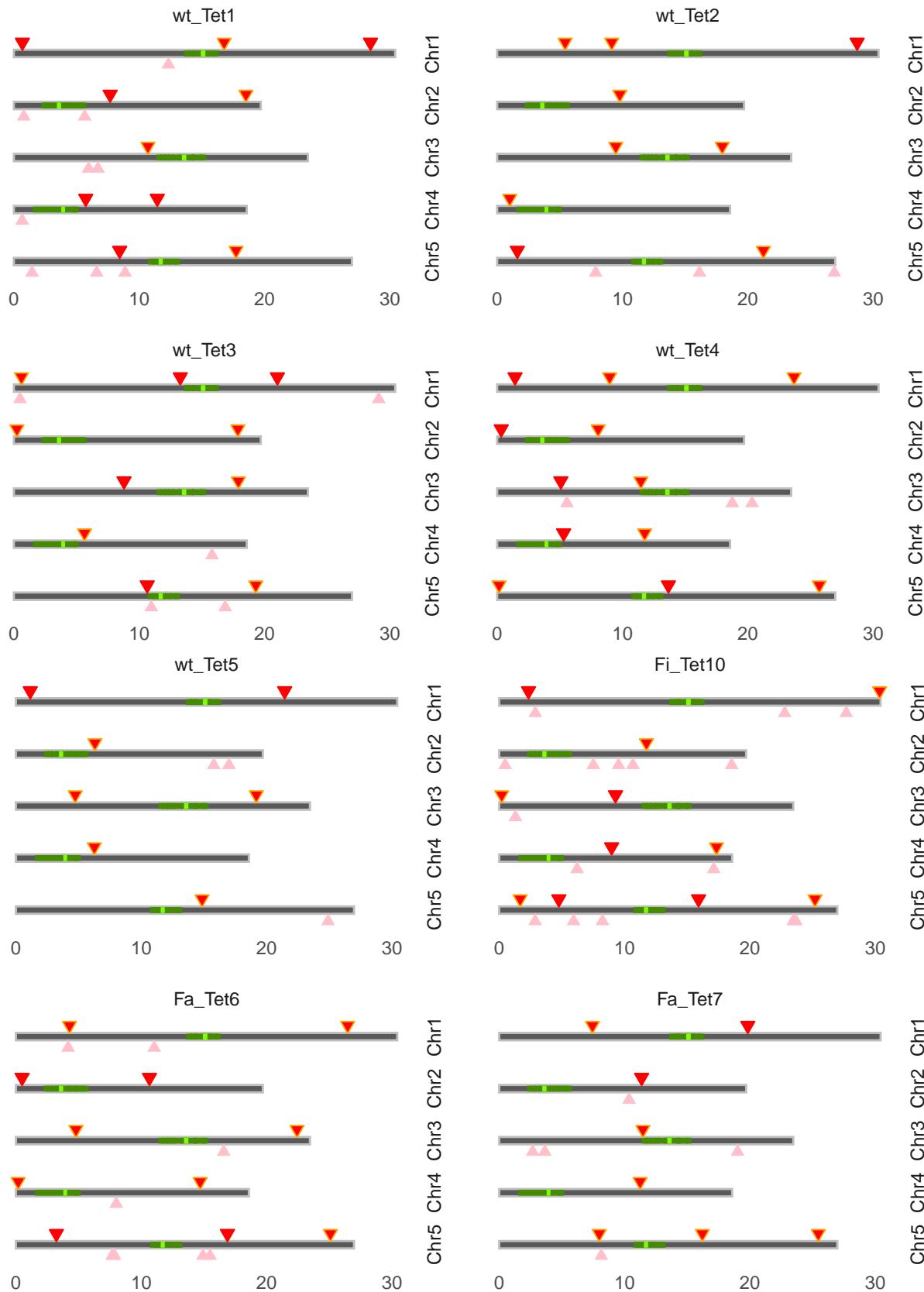
QFiFa_Tet19

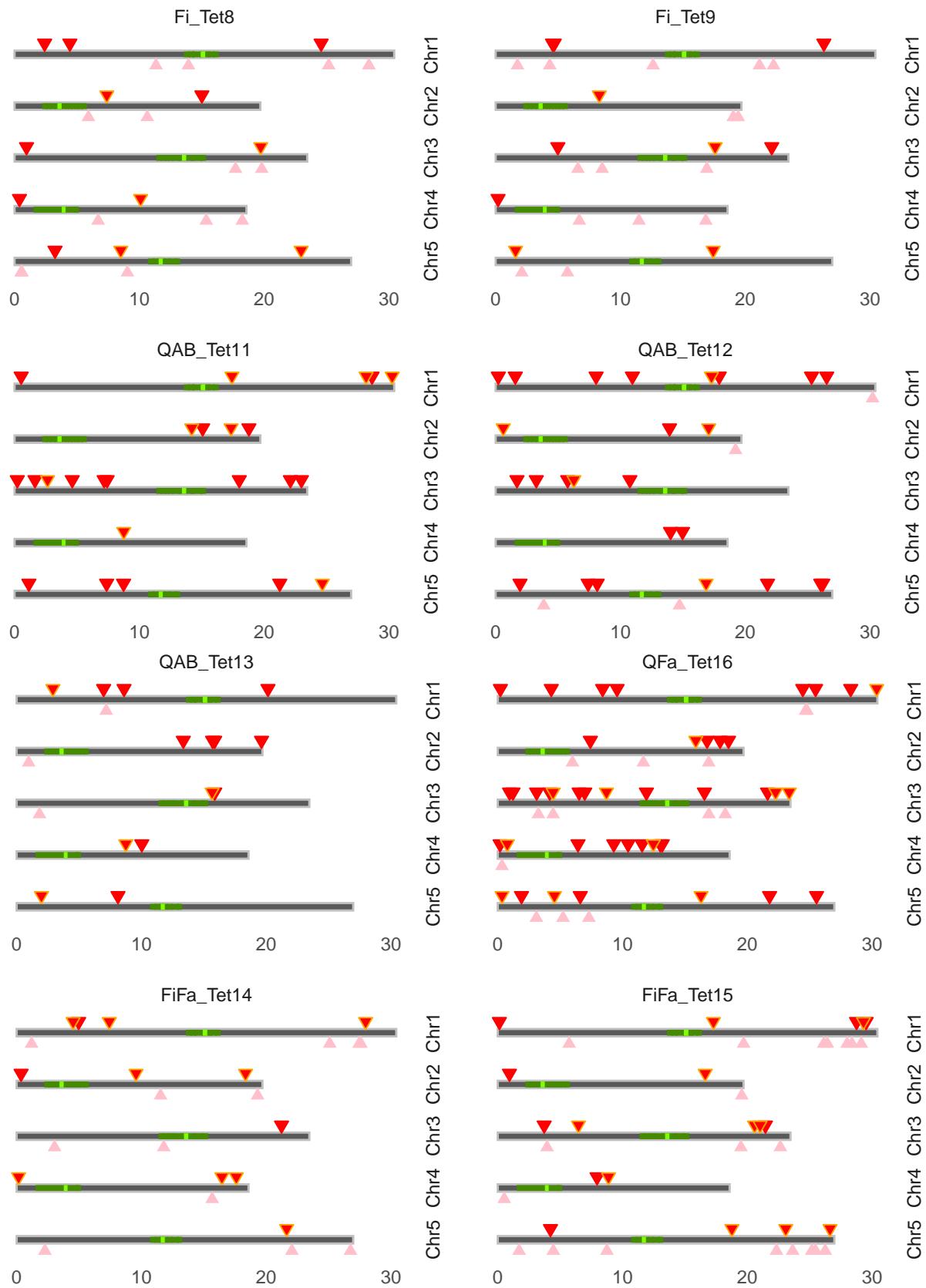


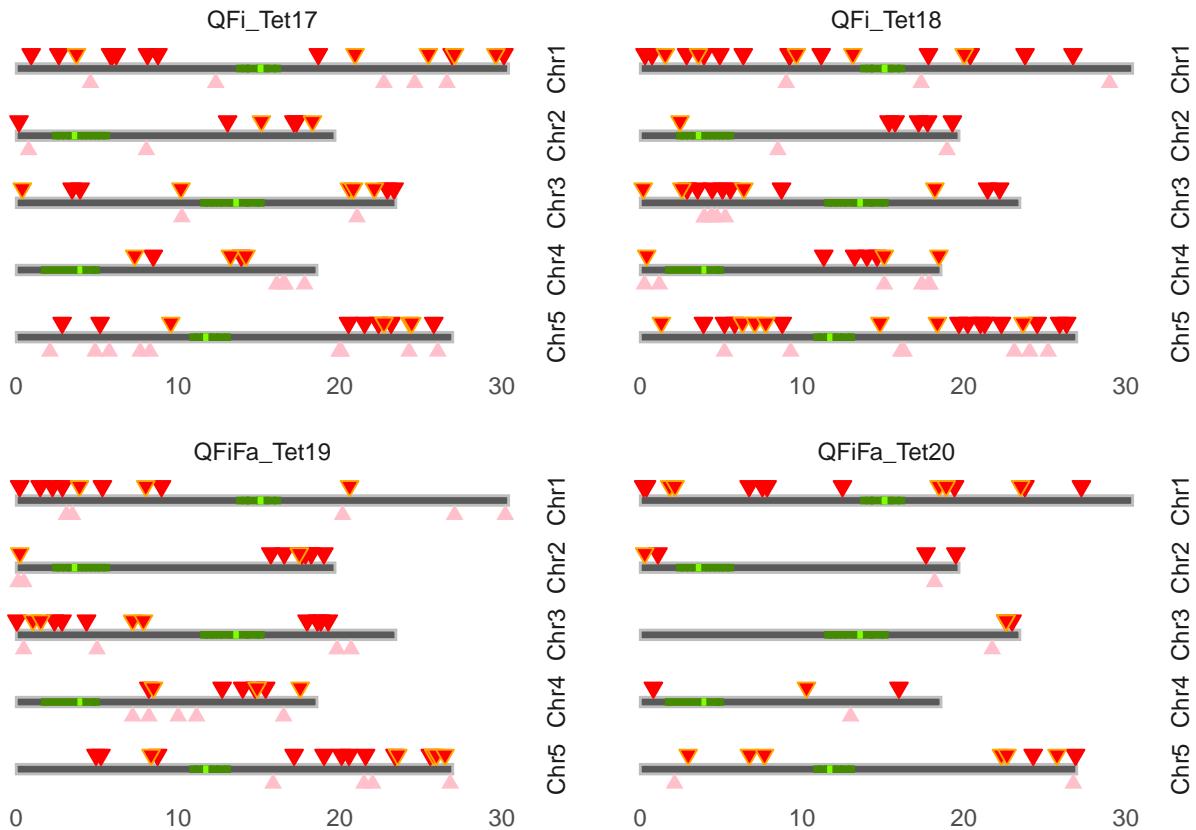
QFiFa_Tet20



2.5 Distribution of detected CO and NCO along the genome:







2.6 Number of detected events per tetrads and genotypes

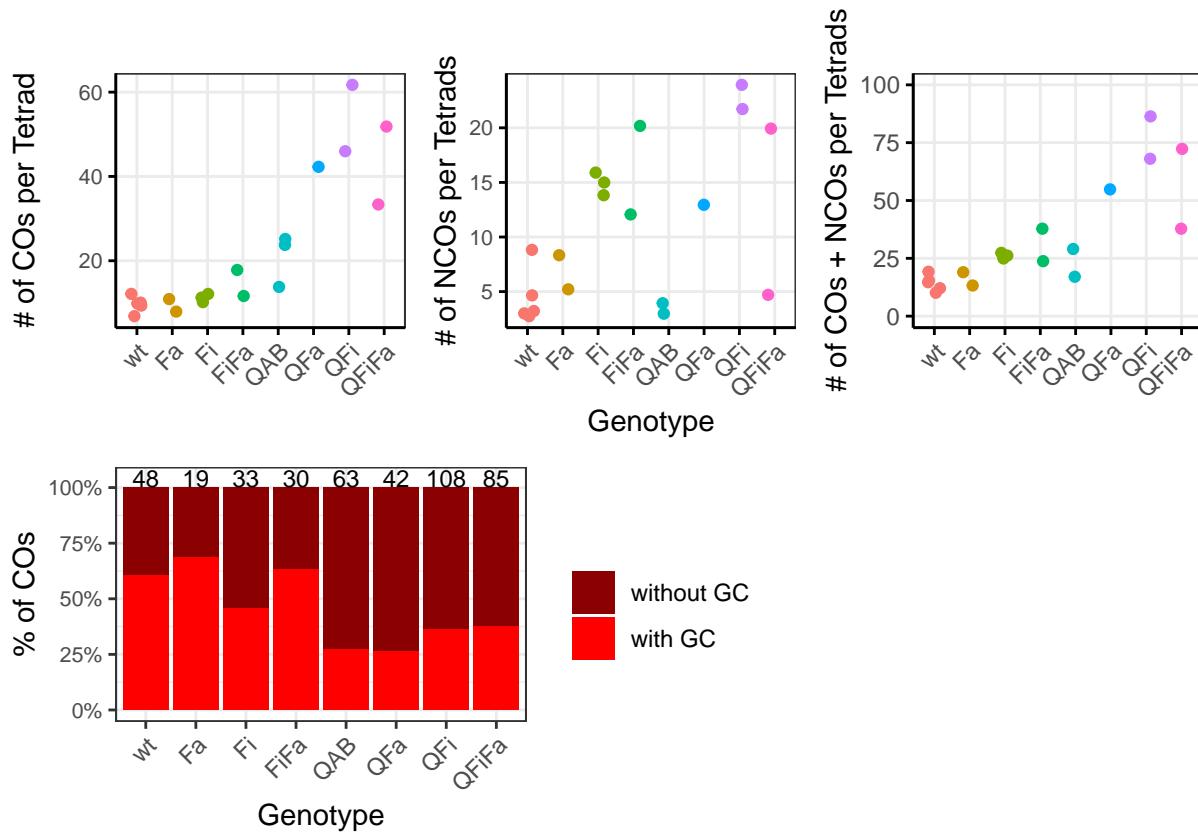


Figure 2: fig2

2.7 Number of events and converted SNP per Tetrad

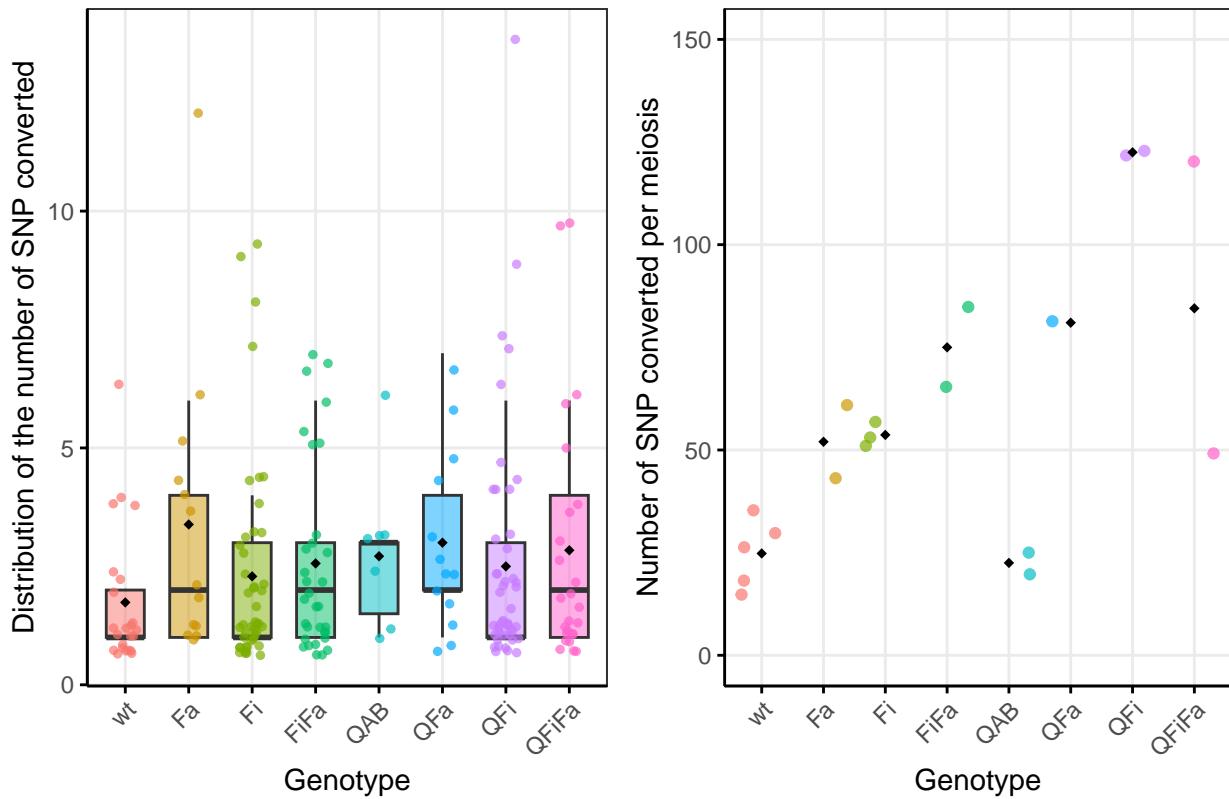


Figure 3: Supp Fig4

2.8 Track Length analysis

Dans l'analyse précédente, on avait pris cette décision:

When the two SNPs flanking the conversion events are at a cumulativ distance greater than 1 kb (ie twice the distance separating on average 2 snps) then we consider that the estimate of the size of the conversion is not precise enough to be kepted for the analysis.

2.8.1 Descriptiv statistics:

2.8.1.1 NCO % latex table generated in R 4.4.0 by xtable 1.8-4 package % Tue Sep 24 11:27:20 2024

| | Genotype | n | median.mean | minTL.mean | maxTL.mean |
|---|----------|----|-------------|------------|------------|
| 1 | wt | 23 | 2018.74 | 185.26 | 3853.22 |
| 2 | Fa | 13 | 595.65 | 26.00 | 1166.31 |
| 3 | Fi | 45 | 1149.77 | 83.44 | 2217.09 |
| 4 | FiFa | 32 | 1113.94 | 271.12 | 1957.75 |
| 5 | QAB | 7 | 859.50 | 89.43 | 1630.57 |
| 6 | QFa | 13 | 3094.85 | 492.38 | 5698.31 |
| 7 | QFi | 46 | 1370.43 | 215.96 | 2525.91 |
| 8 | QFiFa | 25 | 1501.34 | 532.12 | 2471.56 |

Table 2: table1

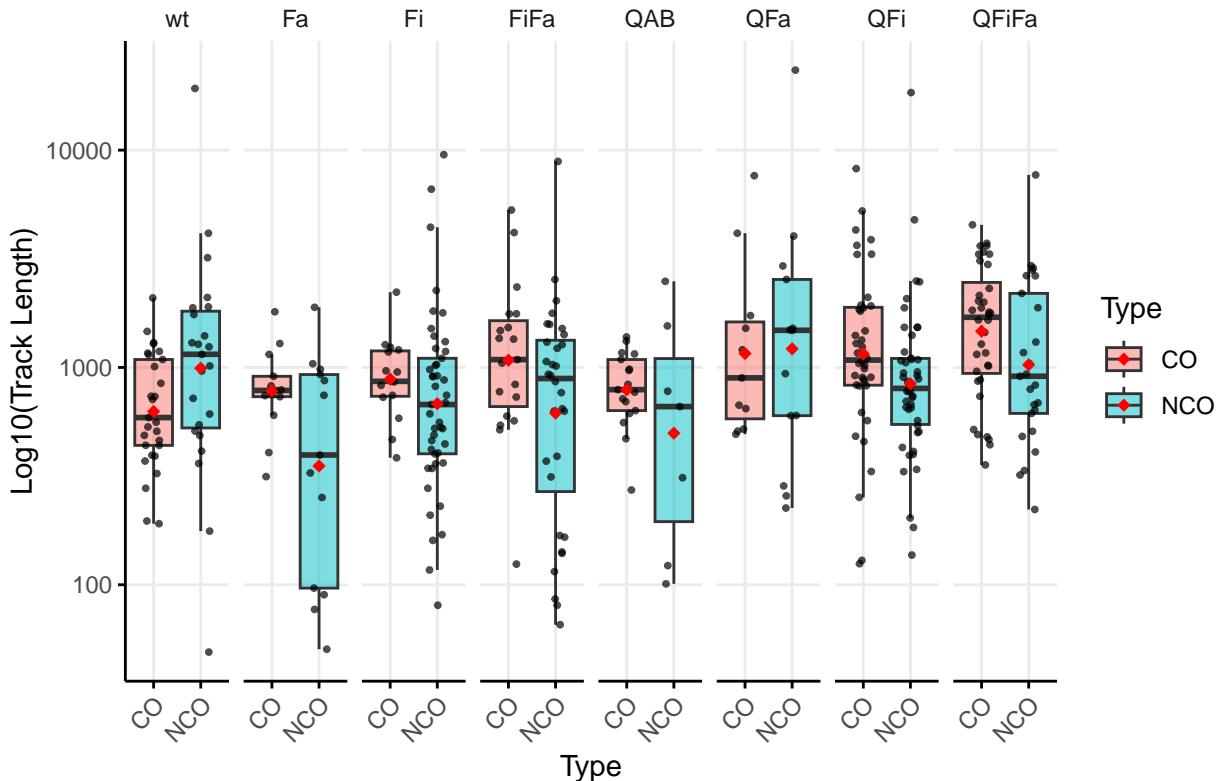
2.8.1.2 CO % latex table generated in R 4.4.0 by xtable 1.8-4 package % Tue Sep 24 11:27:20 2024

| | Genotype | n | median.mean | minTL.mean | maxTL.mean |
|---|----------|----|-------------|------------|------------|
| 1 | wt | 29 | 742.78 | 255.90 | 1230.66 |
| 2 | Fa | 14 | 870.14 | 294.07 | 1447.21 |
| 3 | Fi | 15 | 1019.17 | 430.27 | 1609.07 |
| 4 | FiFa | 19 | 1659.61 | 896.68 | 2423.53 |
| 5 | QAB | 18 | 1406.83 | 738.11 | 2076.56 |
| 6 | QFa | 13 | 2746.00 | 1866.85 | 3626.15 |
| 7 | QFi | 49 | 1923.40 | 803.76 | 3044.04 |
| 8 | QFiFa | 40 | 2300.95 | 1160.45 | 3442.45 |

Table 3: table1

2.8.1.3 Comparison of CO and NCO TL within Genotype

Track Length by Genotype and Type



% latex table generated in R 4.4.0 by xtable 1.8-4 package % Tue Sep 24 11:27:21 2024

| | Genotype | statistic | p.value | alternative | p.adjust | significant |
|---|----------|-----------|---------|-------------|----------|-------------|
| 1 | Fa | 110.00 | 0.20 | two.sided | 0.20 | |
| 2 | Fi | 416.00 | 0.19 | two.sided | 0.19 | |
| 3 | FiFa | 380.00 | 0.14 | two.sided | 0.14 | |
| 4 | QAB | 71.00 | 0.49 | two.sided | 0.49 | |
| 5 | QFa | 71.50 | 1.00 | two.sided | 1.00 | * |
| 6 | QFi | 1178.00 | 0.03 | two.sided | 0.03 | * |
| 7 | QFiFa | 573.00 | 0.07 | two.sided | 0.07 | |
| 8 | wt | 222.00 | 0.04 | two.sided | 0.04 | * |

2.8.2 Comparaison of the CO TL: between wt and each genotype

% latex table generated in R 4.4.0 by xtable 1.8-4 package % Tue Sep 24 11:27:21 2024

| | Genotype | statistic | p.value | alternative | p.adjust | significant |
|---|----------|-----------|---------|-------------|----------|-------------|
| 1 | Fa | 143.00 | 0.22 | two.sided | 0.22 | |
| 2 | Fi | 143.00 | 0.07 | two.sided | 0.07 | |
| 3 | QAB | 185.00 | 0.17 | two.sided | 0.17 | |
| 4 | FiFa | 147.00 | 0.01 | two.sided | 0.01 | * |
| 5 | QFa | 93.00 | 0.04 | two.sided | 0.04 | * |
| 6 | QFi | 314.00 | 0.00 | two.sided | 0.00 | * |
| 7 | QF1Fa | 198.00 | 0.00 | two.sided | 0.00 | * |

2.8.3 Comparaison of the NCO TL: between wt and each genotype

% latex table generated in R 4.4.0 by xtable 1.8-4 package % Tue Sep 24 11:27:21 2024

| | Genotype | statistic | p.value | alternative | p.adjust | significant |
|---|----------|-----------|---------|-------------|----------|-------------|
| 1 | Fa | 222.00 | 0.02 | two.sided | 0.02 | * |
| 2 | Fi | 657.00 | 0.07 | two.sided | 0.07 | |
| 3 | FiFa | 446.00 | 0.19 | two.sided | 0.19 | |
| 4 | QAB | 104.00 | 0.27 | two.sided | 0.27 | |
| 5 | QFa | 138.00 | 0.72 | two.sided | 0.72 | |
| 6 | QFi | 613.00 | 0.29 | two.sided | 0.29 | |
| 7 | QF1Fa | 291.00 | 0.95 | two.sided | 0.95 | |

Table 4: table1

2.9 Distribution of CO and NCO in features

2.10 nb SNP annot

```
##      UTR3 UTR5 Intron Coding TE Hetero Promoter
## GCCO    15   16    120    123  79      2     417
## NCO     2    5    29     24  50      7     111
```

2.11 nb Event Annot

```
##      UTR3 UTR5 Intron Coding TE Hetero Promoter
## GCCO    13   12    50     59  25      3     121
## CO      31   21    84    101  40      0     195
## NCO     1    4    14    10  14      4     40
```

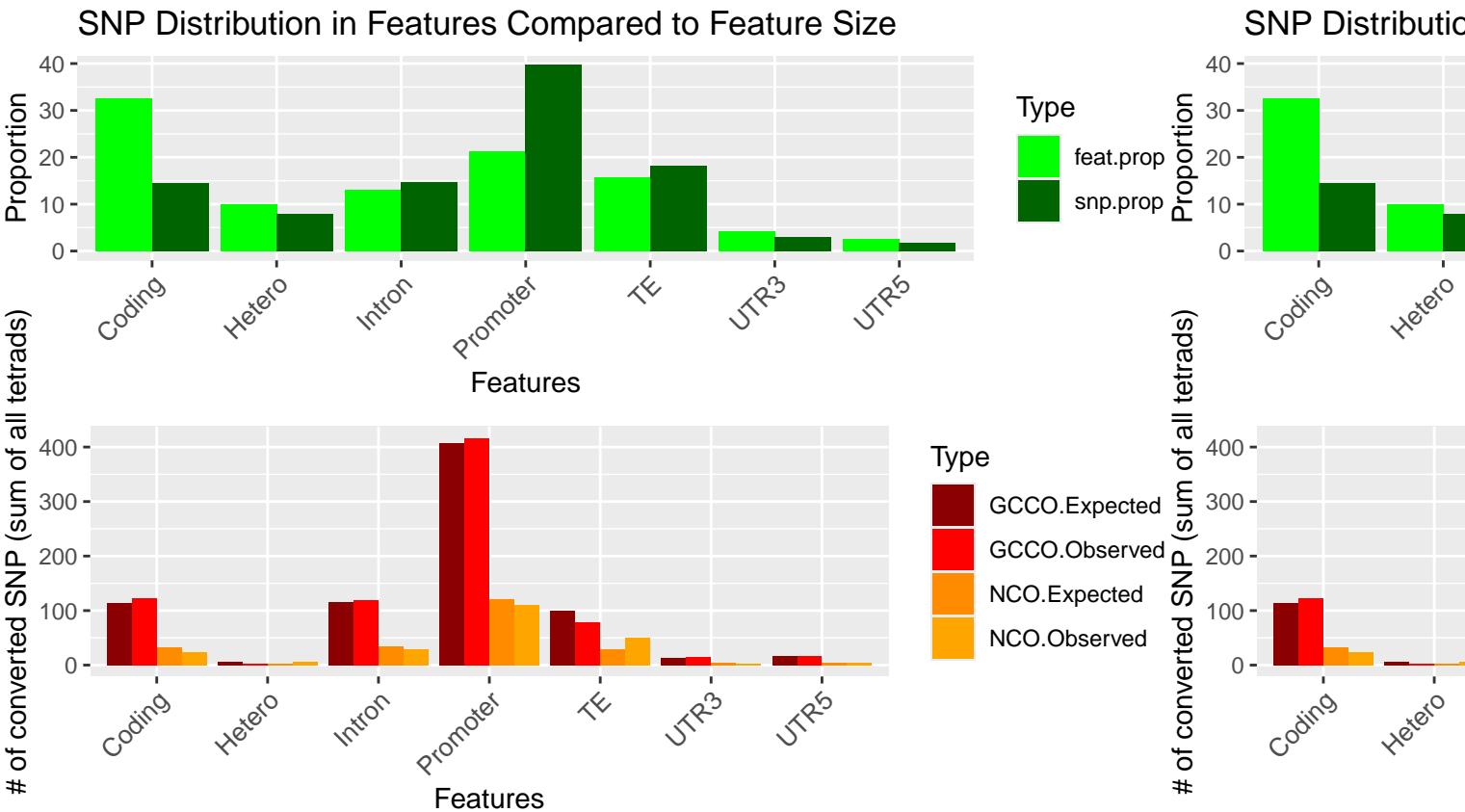
2.12 Distribution of SNP in features

The datamart **TxDb.Athaliana.BioMart.plantsmart28** was used to annotate the SNP in the different features of the genome thanks to the locateVariants function from the VariantAnnotation package.

% latex table generated in R 4.4.0 by xtable 1.8-4 package % Tue Sep 24 11:27:32 2024

| | Compartments | SumLength | NbSNP | feat.prop | snp.prop | GCCO | NCO | GCCO.event | CO.event | NCO.event |
|----------|--------------|-------------|--------|-----------|----------|--------|--------|------------|----------|-----------|
| Intron | Intron | 20610303.00 | 99430 | 13.04 | 14.62 | 120.00 | 29.00 | 50.00 | 84.00 | |
| Coding | Coding | 51600369.00 | 97879 | 32.66 | 14.40 | 123.00 | 24.00 | 59.00 | 101.00 | |
| UTR5 | UTR5 | 4123096.00 | 12522 | 2.61 | 1.84 | 16.00 | 5.00 | 12.00 | 21.00 | |
| UTR3 | UTR3 | 6636022.00 | 20902 | 4.20 | 3.07 | 15.00 | 2.00 | 13.00 | 31.00 | |
| Hetero | Hetero | 15711413.00 | 54382 | 9.94 | 8.00 | 2.00 | 7.00 | 3.00 | 0.00 | |
| TE | TE | 24932539.00 | 124024 | 15.78 | 18.24 | 79.00 | 50.00 | 25.00 | 40.00 | |
| Promoter | Promoter | 33602000.00 | 269863 | 21.27 | 39.69 | 417.00 | 111.00 | 121.00 | 195.00 | |

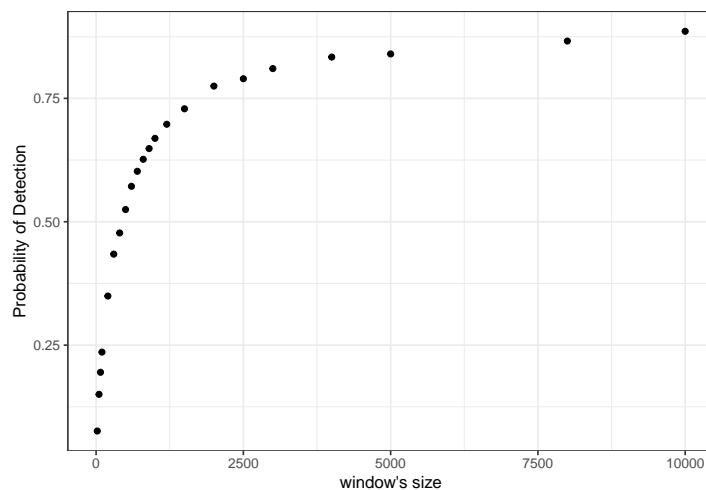
The number of converted SNP per features (summing up all tetrads results) is compared to the number of SNP awaited for each feature if the distribution follow the SNP distribution in features.



```
##  
## Pearson's Chi-squared test  
##  
## data: rr[2:3, ]  
## X-squared = 40.701, df = 6, p-value = 3.317e-07
```

2.13 Probability of conversion detection

Given the gold SNP distribution in the genome, we computed the probability of detecting a SNP in a window of a given size. We sampled 10000 windows of different sizes and computed the mean number of SNP detected and the number of windows with at least one SNP.



3 Bibliographie

4 Session info

```
sessionInfo()
```

```
## R version 4.4.0 (2024-04-24)
## Platform: aarch64-apple-darwin20
## Running under: macOS Ventura 13.1
##
## Matrix products: default
## BLAS:    /Library/Frameworks/R.framework/Versions/4.4-arm64/Resources/lib/libRblas.0.dylib
## LAPACK:  /Library/Frameworks/R.framework/Versions/4.4-arm64/Resources/lib/libRlapack.dylib; LAPACK version
##
## locale:
## [1] en_US.UTF-8/en_US.UTF-8/en_US.UTF-8/C/en_US.UTF-8/en_US.UTF-8
##
## time zone: Europe/Paris
## tzcode source: internal
##
## attached base packages:
## [1] stats4      stats       graphics   grDevices  utils      datasets   methods
## [8] base
##
## other attached packages:
## [1] TxDb.Athaliana.BioMart.plantsmart28_3.2.2
## [2] GenomicFeatures_1.56.0
## [3] AnnotationDbi_1.66.0
## [4] Biobase_2.64.0
## [5] GenomicRanges_1.56.0
## [6] GenomeInfoDb_1.40.1
## [7] IRanges_2.38.0
## [8] S4Vectors_0.42.0
## [9] BiocGenerics_0.50.0
## [10] patchwork_1.2.0
## [11] cowplot_1.1.3
## [12] broom_1.0.6
## [13] gridExtra_2.3
## [14] xtable_1.8-4
## [15] plyr_1.8.9
## [16] lubridate_1.9.3
## [17] forcats_1.0.0
## [18] stringr_1.5.1
## [19] dplyr_1.1.4
## [20] purrr_1.0.2
## [21] readr_2.1.5
## [22] tidyverse_2.0.0
##
## loaded via a namespace (and not attached):
## [1] DBI_1.2.3                  bitops_1.0-7
## [3] rlang_1.1.4                 magrittr_2.0.3
## [5] matrixStats_1.3.0            compiler_4.4.0
## [7] RSQLite_2.3.7                mgcv_1.9-1
## [9] png_0.1-8                  systemfonts_1.1.0
```

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## [11] vctrs_0.6.5                  pkgconfig_2.0.3
## [13] crayon_1.5.2                 fastmap_1.2.0
## [15] backports_1.5.0              XVector_0.44.0
## [17] labeling_0.4.3                utf8_1.2.4
## [19] Rsamtools_2.20.0             rmarkdown_2.27
## [21] tzdb_0.4.0                   UCSC.utils_1.0.0
## [23] ragg_1.3.2                  tinytex_0.52
## [25] bit_4.0.5                   xfun_0.44
## [27] zlibbioc_1.50.0              cachem_1.1.0
## [29] jsonlite_1.8.8              blob_1.2.4
## [31] highr_0.11                  DelayedArray_0.30.1
## [33] BiocParallel_1.38.0          parallel_4.4.0
## [35] R6_2.5.1                    stringi_1.8.4
## [37] rtracklayer_1.64.0           Rcpp_1.0.12
## [39] SummarizedExperiment_1.34.0 knitr_1.47
## [41] Matrix_1.7-0                 splines_4.4.0
## [43] timechange_0.3.0            tidyselect_1.2.1
## [45] abind_1.4-5                 rstudioapi_0.16.0
## [47] yaml_2.3.8                  codetools_0.2-20
## [49] curl_5.2.1                  lattice_0.22-6
## [51] withr_3.0.0                 KEGGREST_1.44.0
## [53] evaluate_0.23                Biostrings_2.72.1
## [55] pillar_1.9.0                 MatrixGenerics_1.16.0
## [57] generics_0.1.3               RCurl_1.98-1.14
## [59] hms_1.1.3                   munsell_0.5.1
## [61] scales_1.3.0                 glue_1.7.0
## [63] tools_4.4.0                 BiocIO_1.14.0
## [65] GenomicAlignments_1.40.0    XML_3.99-0.16.1
## [67] grid_4.4.0                  colorspace_2.1-0
## [69] nlme_3.1-165                GenomeInfoDbData_1.2.12
## [71] restfulr_0.0.15             cli_3.6.2
## [73] textshaping_0.4.0            fansi_1.0.6
## [75] S4Arrays_1.4.1              gtable_0.3.5
## [77] digest_0.6.35                SparseArray_1.4.8
## [79] rjson_0.2.21                farver_2.1.2
## [81] memoise_2.0.1               htmltools_0.5.8.1
## [83] lifecycle_1.0.4              httr_1.4.7
## [85] bit64_4.0.5

```

Anderson, Stacy Y. AND Dimon, Carol M. AND Chen. 2011. “ReCombine: A Suite of Programs for Detection and Analysis of Meiotic Recombination in Whole-Genome Datasets.” PLOS ONE 6 (10): 1–17. <https://doi.org/10.1371/journal.pone.0025509>.

Giraut, Matthieu AND Drouaud, Laurène AND Falque. 2011. “Genome-Wide Crossover Distribution in Arabidopsis Thaliana Meiosis Reveals Sex-Specific Patterns Along Chromosomes.” PLOS Genetics 7 (11): 1–15. <https://doi.org/10.1371/journal.pgen.1002354>.

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