

# Detection of balanced reciprocal translocations in pigs using short read sequencing

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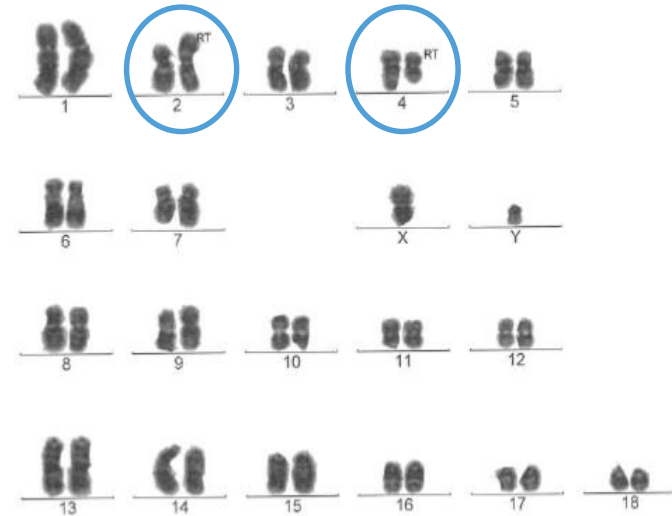


# Acknowledgement

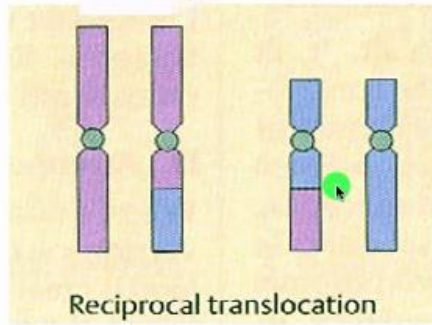


# Balanced reciprocal translocations

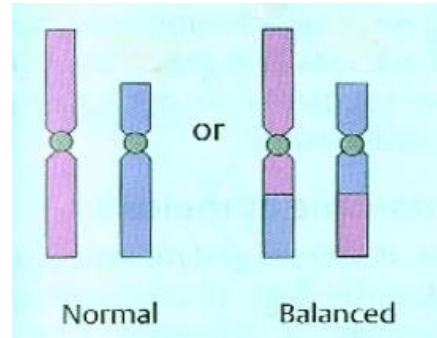
- Part of non-homologous chromosomes exchanged
- No gain nor loss of chromosomal material
- Healthy individuals
- Karyotyping: chromosome staining



# Reduced fertility



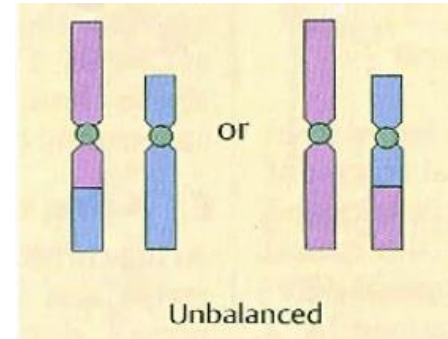
## Gametes



25%

25%

↓  
Carriers



50%

↓  
Unviable  
Malformed

# Karyotype screening AI boars

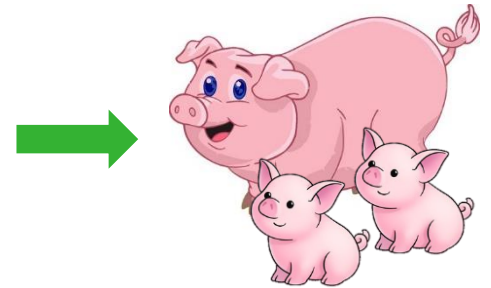
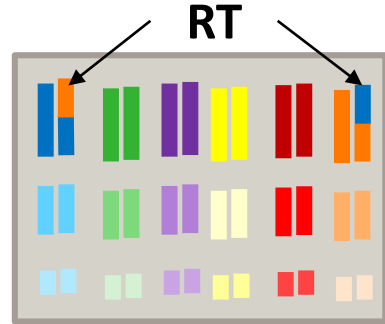
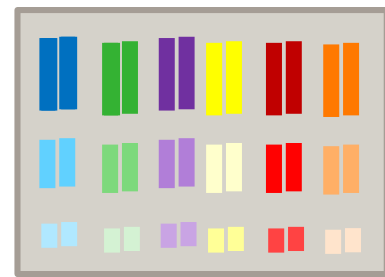
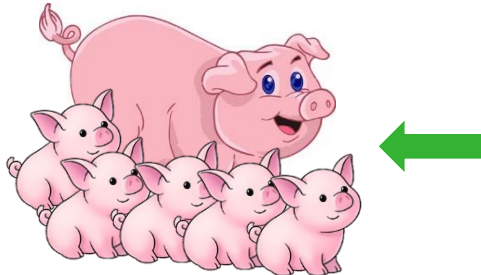
AI boars



normal

0.47% abnormal karyotype

- Mosaicism
- **Reciprocal translocations (RT)**



**Subfertility**

# Issue

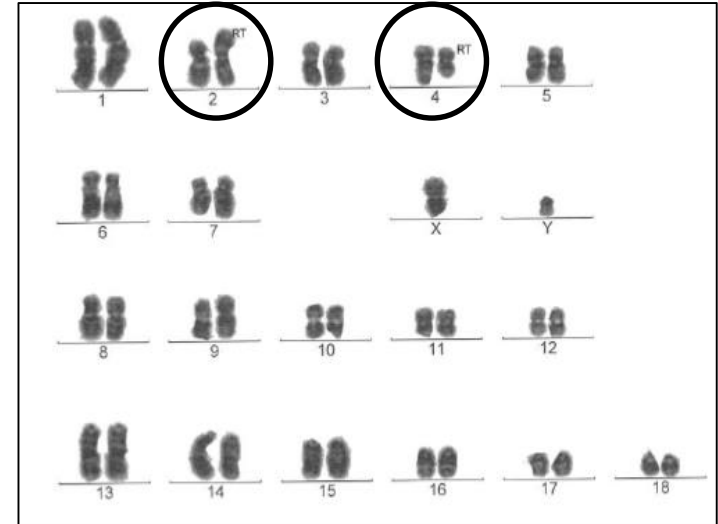
- Chromosome staining sensitive and labour-intensive process
- Translocations smaller than bandsize remain undetected
- SNP genotypes can detect unbalanced karyotypes and inherited RTs
  
- Expand use of sequence data & detect smaller translocations

# Aim

**Test the possibilities of paired end short read sequencing  
for detection of balanced reciprocal translocations**

# Data

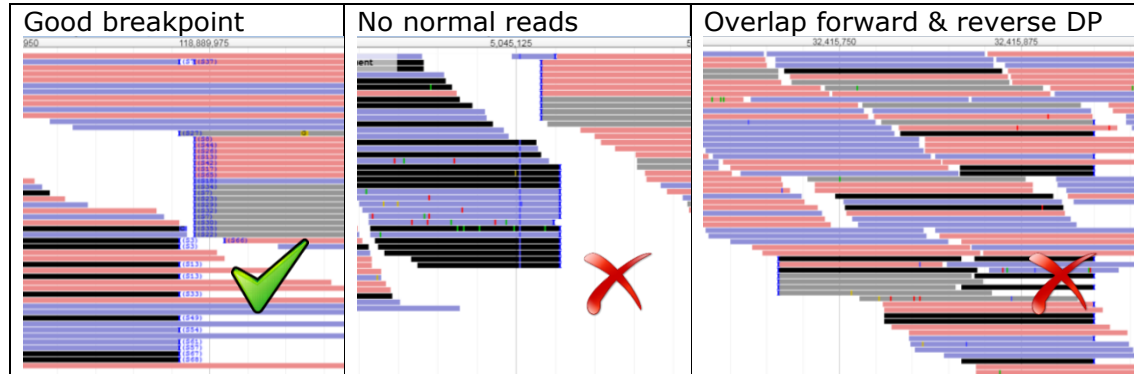
- 7 boars with positive RT test
- 31 control pigs
- 30x Illumina paired end 150bp reads
- Fragments 300-450bp
- Aligned against SusScrofa 11.1





# Detection in sequence

- Software: Delly (DP&SR)
- Basic filtering: contigs, low quality, imprecise
- Final filtering: mapping quality, read support, reciprocal breakpoint present  
Using reference & 10 filter animals: genome build & common issues
- Visual inspection (Jbrowse)



# Results cases

	PIG1	PIG2	PIG3	PIG4	PIG5	PIG6	PIG7
RT chr	2/4	6/8	2/4	2/4	7/14	2/4	1/16
Coverage	32.6	37.9	33.4	35.1	37.2	31.1	30.3
Delly out	73,923	96,378	85,597	94,223	94,683	75,368	78,946
Basic filt	1,127	1,426	1,185	1,175	1,503	1,067	1,116
Final filt	30(15)	56(28)	34(17)	38(19)	68(34)	38(19)	44(22)
Visual insp	<b>1(2,4)</b>	<b>1(6,8)</b>	<b>1(2,4)</b>	<b>1(2,4)</b>	<b>1(7,14)</b>	<b>1(2,4)</b>	<b>0(FN)</b>

- Pig 3 & 4 FS, Pig 6 HS, Pig 1 related -> same RT
- Pig 4: visual inspection debatable
- Pig 7: nothing passed visual inspection -> **false negative**

# Results controls (n=31)

	Average	Min	Max
<b>Coverage</b>	31.7	27.4	38.4
<b>Delly out</b>	66,167.6	42,281	100,641
<b>Final filt</b>	38.3	20(10)	64(32)
<b>Visual insp</b>	<b>0</b>	0	0

- Specificity = 100% (31/31)
- Sensitivity = 85.7% (6/7)

# Reduced coverage

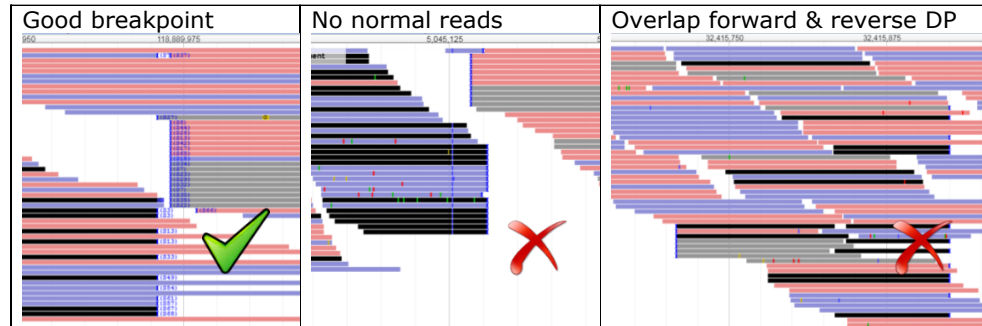
	PIG1	PIG2	PIG3	PIG4	PIG5	PIG6	PIG7
RT chr	2/4	6/8	2/4	2/4	7/14	2/4	1/16
10x	<b>4(2)</b>	<b>12(6)</b>	<b>0</b>	<b>2(1)</b>	<b>4(2)</b>	<b>4(2)</b>	<b>6(3)</b>
15x	<b>8(4)</b>	<b>16(8)</b>	<b>12(6)</b>	<b>18(9)</b>	<b>24(12)</b>	<b>4(2)</b>	<b>14(7)</b>
20x	<b>20(10)</b>	<b>30(15)</b>	<b>18(9)</b>	<b>28(14)</b>	<b>30(15)</b>	<b>6(3)</b>	<b>30(15)</b>
10x relaxed	<b>32(16)</b>	<b>24(12)</b>	<b>28(14)</b>	<b>26(13)</b>	<b>40(20)</b>	<b>8(4)</b>	<b>28(14)</b>
15x relaxed	<b>46(23)</b>	<b>62(31)</b>	<b>46(23)</b>	<b>54(27)</b>	<b>60(30)</b>	<b>44(22)</b>	<b>52(26)</b>
20x relaxed	<b>54(27)</b>	<b>68(34)</b>	<b>48(24)</b>	<b>66(33)</b>	<b>62(31)</b>	<b>50(25)</b>	<b>54(27)</b>

- With 10x some RT breakpoints were not detected by Delly
- Filter criteria need to be adjusted to the coverage

# Next

## Automated visual inspection

- Bioinformatics: read flags
- DeepLearning: CNN



# Conclusions

- Detectable using short reads
- Specificity 100%, sensitivity 85.7%
- 20x or more required



Post-graduate course:

# Characterization, management and exploitation of genomic diversity in animals

Wageningen, 9-13 December 2019

## Course content

- Measures of genomic diversity
- Diversity across breeds
- Adaptive introgression
- Functional genomics
- Management of small populations
- Optimal contributions

## Lecturers

- Mirte Bosse, Wageningen University
- Martien Groenen, Wageningen University
- Gabor Meszaros, BOKU, Vienna
- Christian Reimer, Georg-August-University, Göttingen
- Michele Tixtier, INRA Jouy-en-Josas
- Jack Windig, Wageningen Research

For more information and registration please check: [www.wur.eu/wias](http://www.wur.eu/wias)