KromaTiD

Wild Type and Edited Cells Comparison *QYYMMDD*

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Customer: Project Lead

Project Overview

Company X is editing human cells and testing the resulting batches against unedited controls (also referred to as WT) for chromosome ploidy and structural rearrangements. For this project, KromaTiD:

- Received two cryopreserved samples.
- Recovered cells into culture and harvested the two samples.
- Stained and imaged metaphase slides for each sample.
- Analyzed 100 metaphase spreads from each sample and generated independent reports of genomic integrity.
- Generated this summary report comparing the unedited to the edited sample; NOTE: % of Cells equals # of Cells since 100 cells were analyzed.

Procedure

See individual sample reports (**Appendix A**) for method information.

Results

The two samples compared in this summary include an unedited reference (WT) and an edited sample (Treated Z). Independent reports for each sample (see **Appendix A**) comprise 100 cells scored using industry-standard protocols (ISCN 2020: An International System for Human Cytogenomic Nomenclature). **Appendix B** contains a list of all chromosomal variants scored and their abbreviations.

Table 1: Sample List

Sample Name	LIMS ID
WT	S######
Treated Z	S######

Table 2: Summary of Ploidy for WT and Treated Z

Category	WT	Treated Z	Fisher's Exact*	*p*-value significant
	% of Cells	% of Cells	*p*-value	< .05
Diploid	100	100	N/A	N/A

*Fisher's exact test assumes the null hypothesis that the two variables are independent, that is, the values of one do not depend on the values of the other. *P*-value significance threshold < .05.

Table 3: Summary of Events by Category for WT and Treated Z

Category	WT	Treated Z	Fisher's Exact*	*p*-value significant
	% of Cells	% of Cells	*p*-value	< .05
No Events	84	74	0.1175	No
Chromosome Aneuploidy	13	20	0.2528	No
Marker Chromosomes	1	2	1	No
Additional material of unknown origin	1	0	1	No
Deletions	1	2	1	No
Inversions	0	1	1	No
Translocations	0	2	0.4975	No

*Fisher's exact test assumes the null hypothesis that the two variables are independent, that is, the values of one do not depend on the values of the other. *P*-value significance threshold < .05.

Both samples are fully diploid with the treated sample having a higher rate of aneuploidy. Both samples have marker chromosome and deletion events. The WT sample has additional material of unknown origin that was not observed in the treated sample. The treated sample has inversion and translocation events that were not observed in the WT sample. None of the differences are significant by Fisher's Exact Test.

Table 4: Percent of Cells in Each Sample with Specific Events

Event Type	WT % of Cells	Treated Z % of Cells	Fisher's Exact* *p*-value	*p*-value significant < .05
−X	0	1	1	No
−2	4	4	1	No
−3	0	1	1	No
−4	3	0	0.2462	No
−5	2	2	1	No
−7	1	0	1	No
−9	0	1	1	No
−10	1	0	1	No
−11	2	0	0.4975	No
−15	1	2	1	No
−14	1	0	1	No
−16	1	1	1	No
−18	0	1	1	No
−21	2	6	0.279	No
−22	1	0	1	No
9	0	1	1	No
+M	1	2	1	No
add(2)(q10)	1	0	1	No
del(5)(q24)	1	0	1	No
del(9)(q15)	0	1	1	No
del(21)(q25)	0	1	1	No
inv(7)(p13)	0	1	1	No
t(2;9)(p15.3;q22)	0	1	1	No
t(8;12)(q15;q12)	0	1	1	No

*Fisher's exact test assumes the null hypothesis that the two variables are independent, that is, the values of one do not depend on the values of the other. *P*-value significance threshold < .05.

Table 5: Event Rates for Edit Target Chromosomes

Sample Name	WT			Treated Z		
Sample ID	S######			S######		
Category	# Events	# of Cells	% of Cells	# Events	# of Cells	% of Cells
Chr 9 Events	0	0	0	4	4	4
Chr 21 Events	2	2	2	7	7	7

Appendix A:

QYYMMDD Company X Genomic Integrity G-band Final Report S###### (WT)

QYYMMDD Company X Genomic Integrity G-band Final Report S###### (Treated)

Appendix B: Table of karyotyping abbreviations

Abbreviation	Definition
ace	acentric fragment
add	additional material of unknown origin
c	constitutional anomaly
chrb	chromosome break
chtb	chromatid break
cth	chromothripsis
del	deletion
der	derivative chromosome
dic	dicentric
dmin	double minute
dup	duplication
end	endoreduplication
fra	fragile site
h	heterochromatin,constitutive
hsr	homogeneously staining region
i	isochromosome
idic	isodicentric
ins	insertion
inv	inversion
mar or M	marker chromosome
minus sign (-)	loss
multiplication sign (x)	multiple copies of same abnormality
plus sign (+)	gain
qr	quadriradial
r	ring chromosome
t	translocation
tr	triradial

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KromaTiD Genomic Integrity G-Band Assay is for research use only and is not a medical diagnostic test