iCS-digital™ PSC test report

CONTACT PERSON		
Name: Christiaan Arendzen	Phone: +31643293652	E-mail: c.h.arendzen@lumc.nl
INSTITUTION / COMPANY		
Name: LUMC - hiPSC Hotel		
Address: MiraiHouse- Silviusweg 62- 2333BC- Leiden		

PURCHASE ORDER (DATE)	REMAINING TEST
0001336887 (04/07/2023)	**10**

TEST DATE	CONTROL VALIDATION
September 20, 2023	**Normal DNA control for the 24 probes: Passed**

SUMMARY OF THE DETECTED COPY NUMBER VARIATIONS

Sample	Passage	dsDNA (ng/μL)	dsDNA quantity	CNVs	Sex *
LUMC0289iCTRL07	p8	14.3	Good	Not detected	F

The results of this test are for research use only.
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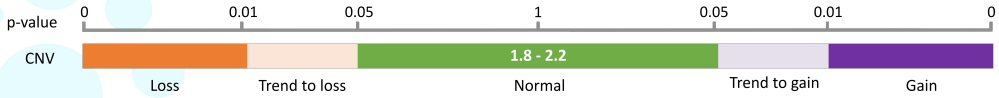
* If unknown, sex is deducted from the ChrX copy number; note that one ChrX can be detected in female cells that lost one ChrX, and two ChrX in male cells that gained one copy.

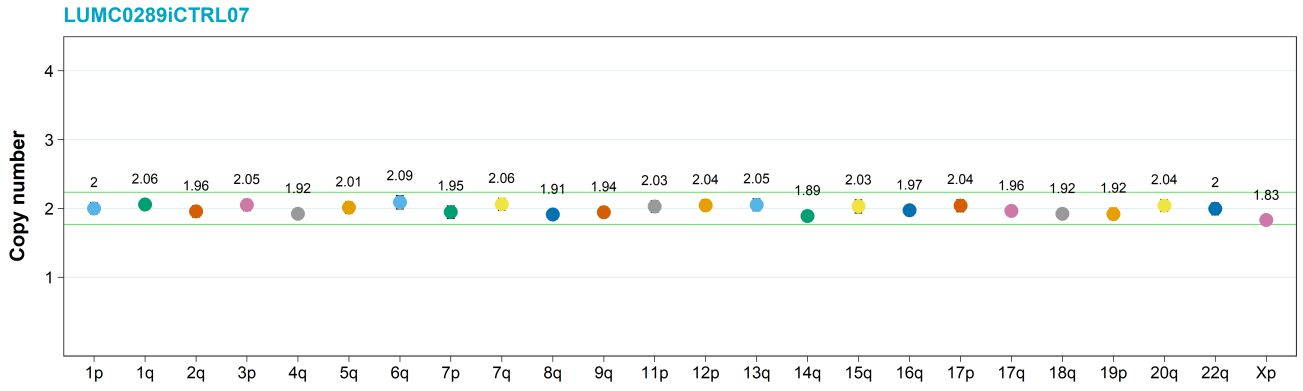
Copy number values:



SUMMARY OF THE DETECTED COPY NUMBER VARIATIONS

CHROMOSOME	1p	1q	2q	3p	4q	5q	6q	7p	7q	8q	9q	11p	12p	13q	14q	15q	16q	17p	17q	18q	19p	20q	22q	Xp
LUMC0289iCTRL07																								

COPY NUMBER VARIATION



Juline VINCENT
R&D Manager

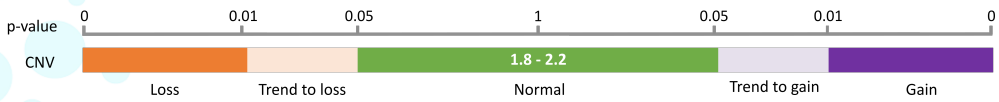
Appendix: iCS-digital™ interpretation method

Our technology allows for the detection of Copy Number Variations (CNVs) that is to say, DNA segments of one kilobase (kb) or larger that are present at an abnormal copy number in comparison with a reference genome. Normal copy number should be equal or close to the value of 2 at all the 24 recurrent regions that we analyze (except for the Xp position since it depends on the sex of the cell line studied: XX or XY). However, due to intrinsic variation caused by multiple factors (DNA concentration, quality, etc.), some samples will present higher copy number fluctuation than others over the 24 positions.

Our interpretation method takes into account the difference in CNV fluctuation observed among samples. More specifically, our statistical analysis is based on normal distribution and is adapted to the overall variability of each sample in an independent manner. P-values are then assigned to each probe and the detection of anomalies is calculated based on their specific p-values and CNVs. The confidence limits (p-value threshold) are displayed as green lines on each graph.

Therefore, for a high quality sample, the fluctuation will be weaker and all copy number values closer to 2 compared with a lower quality samples. The confidence limits (green lines) will be closer or more distant accordingly.

Also, a visual is shown describing the p-value and CNV thresholds detected at each position:



A sample is considered normal by default if its copy number values are strictly between 1.8 and 2.2, or if its p-values are strictly above 0.05. A trend (Trend to loss or Trend to gain), corresponds to a position detected with a p-value between 0.01 and 0.05. Trends are not anomalies but are defined as suspicion of anomalies. It could be linked to the quality of the samples, to the run, or the limit of sensitivity of the test (<20% mosaicism). In these cases we advise to keep an eye on the samples involved and potentially re-test them few weeks/passages later.

An anomaly (CNV = Loss or Gain) is detected if a position presents a p-value strictly below 0.01.

The CNV value gives an information on the proportion of abnormal cells. For example:

- CNV = 2.2 means that 20% of the clonal population has acquired a third copy of the target region.
- CNV = 2.3 means that 30% of the clonal population has acquired a third copy of the target region.
- CNV = 3.0 means that 100% of the clonal population has acquired a third copy of the target region.
- CNV = 3.2 means that 100% of the clonal population has acquired a third copy of the target region and 20% of them has acquired a fourth copy.

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LUMC0289iCTRL10	p8	11.2	Good	Not detected	F

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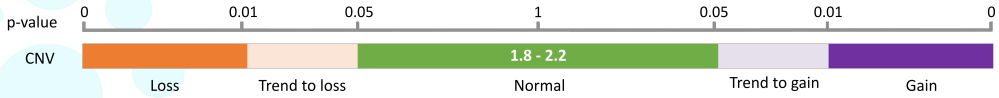

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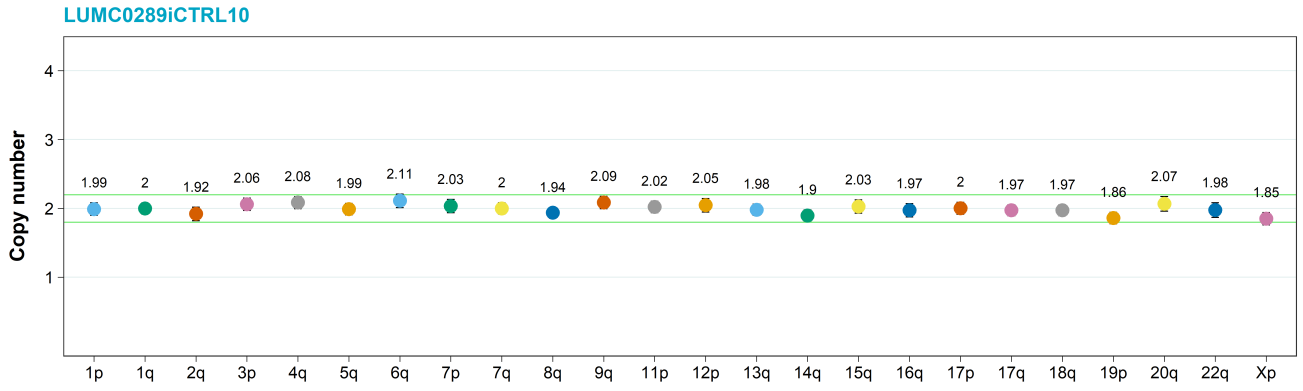
LUMC0289iCTRL10																								

COPY NUMBER VARIATION



Juline VINCENT
R&D Manager

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Our interpretation method takes into account the difference in CNV fluctuation observed among samples. More specifically, our statistical analysis is based on normal distribution and is adapted to the overall variability of each sample in an independent manner. P-values are then assigned to each probe and the detection of anomalies is calculated based on their specific p-values and CNVs. The confidence limits (p-value threshold) are displayed as green lines on each graph.

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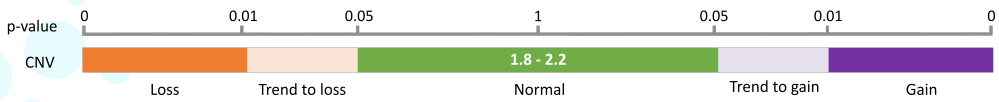

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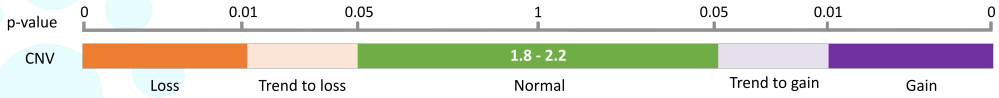

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LUMC0289iCTRL12																								

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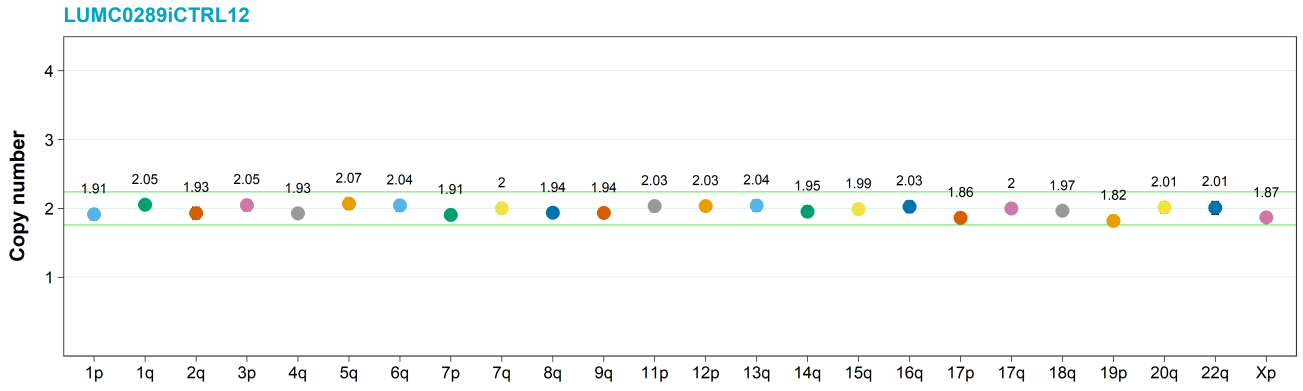

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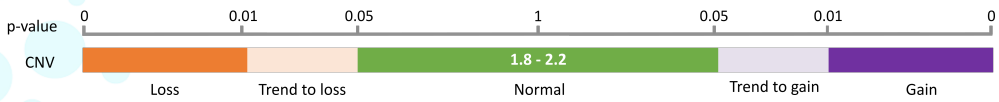

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