# Analyze single cell genomes in under 24 hours with Agilent SurePrint G3 8x60K CGH microarrays

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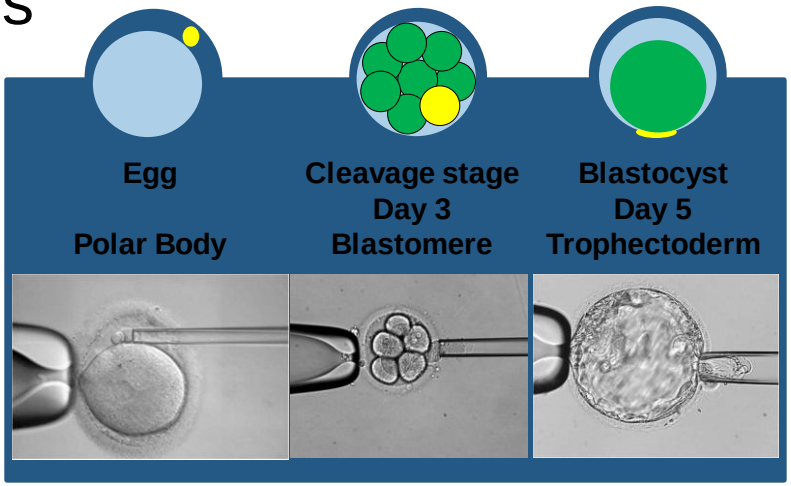
Agilent Technologies

# Single cell analysis is important for specific sample studies

1. Preimplantation samples: Comprehensive chromosome analysis including aneuploidy analysis of all 24 chromosomes

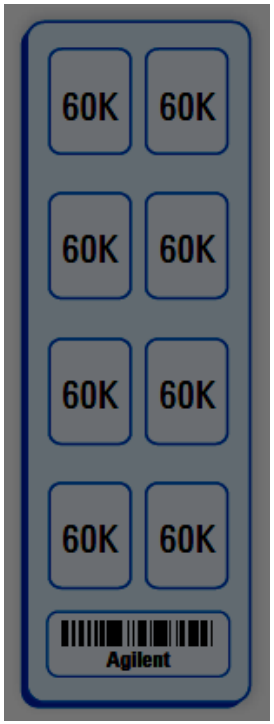



2. Cancer samples: Tumor heterogeneity, micrometastases, minimal residual disease

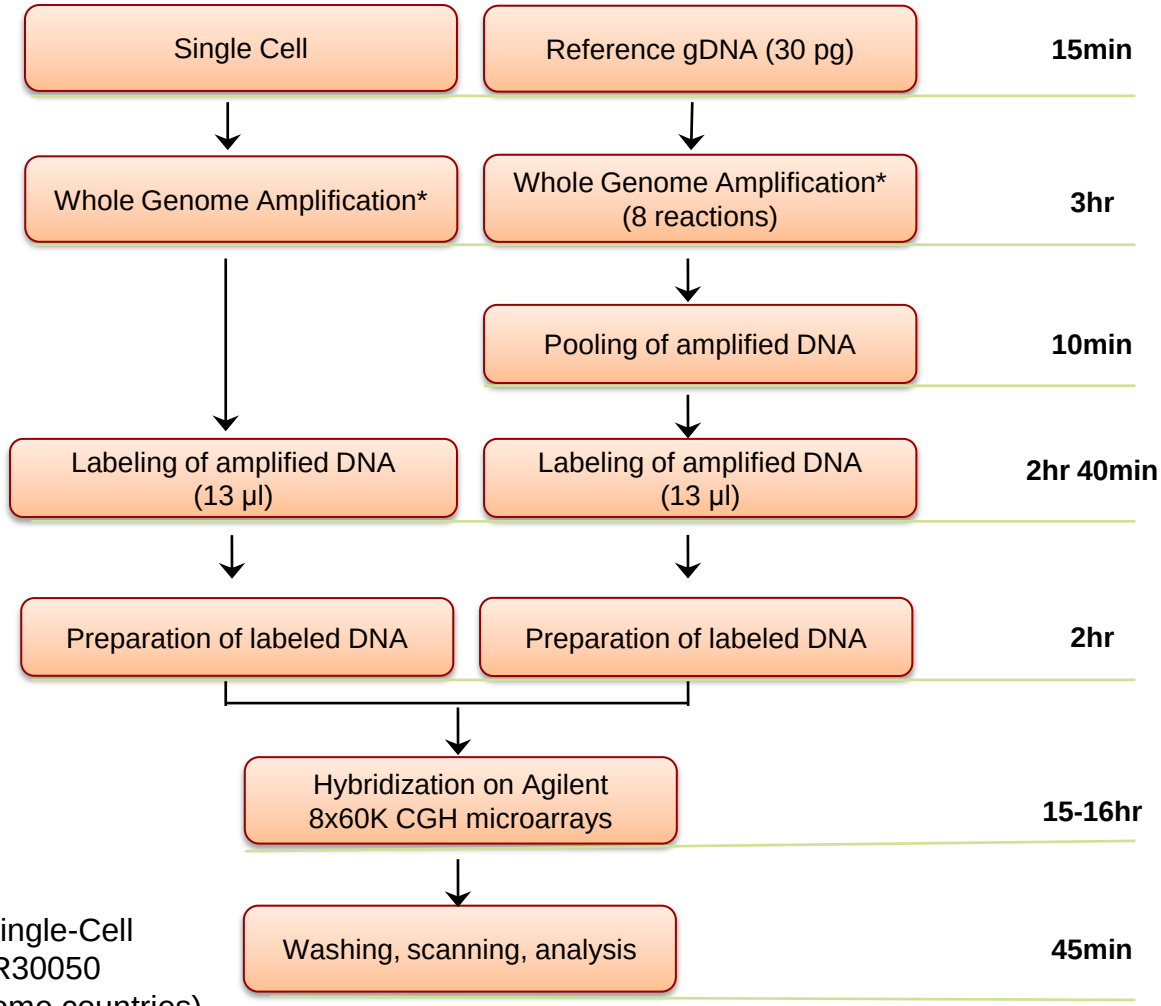
3. Noninvasive prenatal samples

# Single-cell array comparative genomic hybridization on Agilent 8x60K CGH microarray

- Optimized for time sensitive applications
- More cost effective and higher performance than BAC arrays and FISH

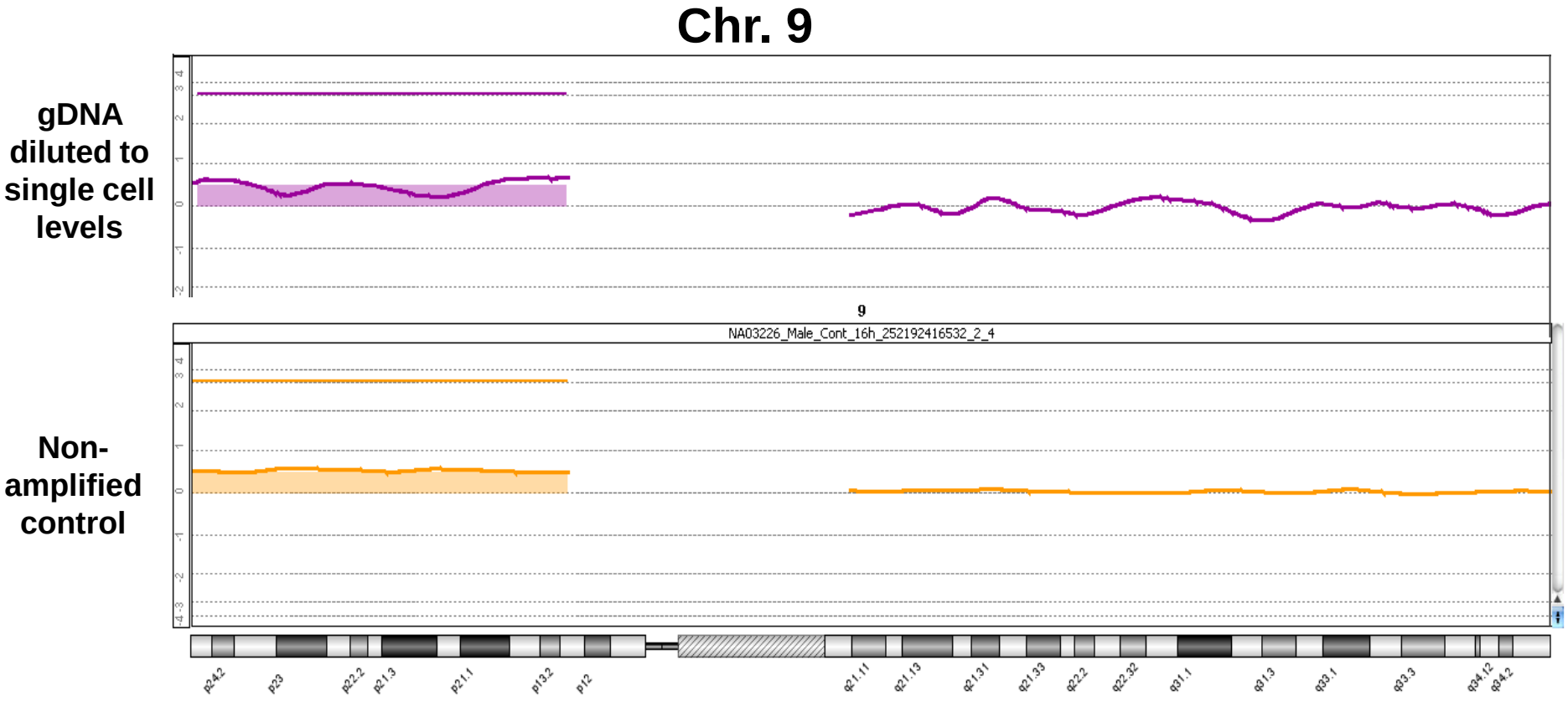


# Protocol: Optimized single cell workflow takes less than 24 hours (manual P/N G4410-90012)




*PicoPlex WGA Kit for Single-Cell
Rubicon Genomics p/n R30050
(distributed by NEB in some countries)

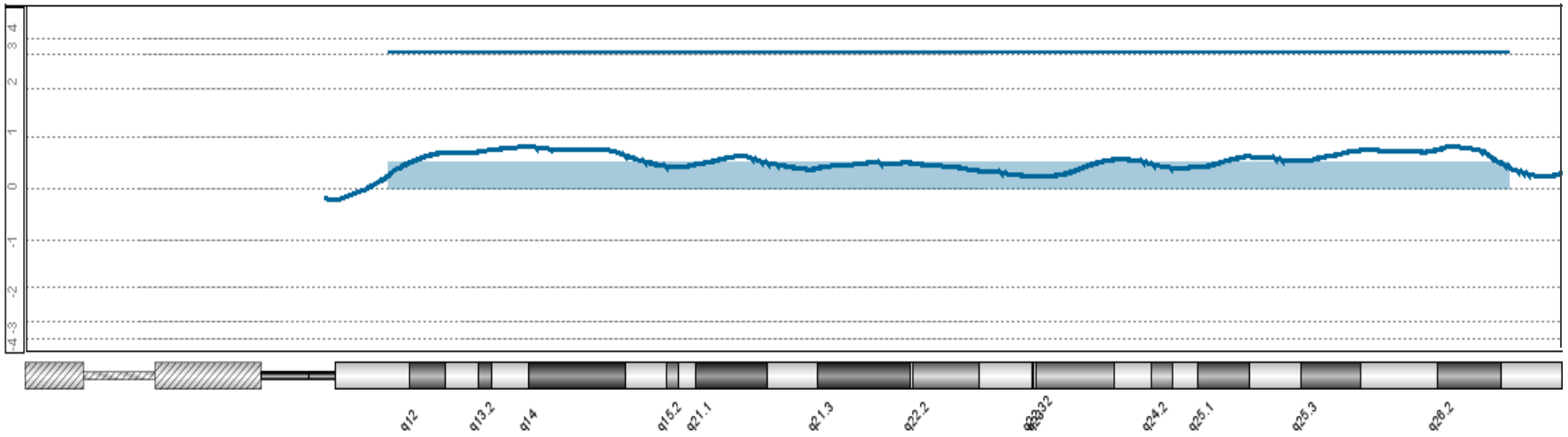
# Expected copy change detected on Agilent 8x60K CGH microarrays in less than 24 hours




**Expected copy number (CN) change identified in amplified DNA (NA03226) with a known aberration on chromosome 9, co-hybridized with a pooled reference amplified from 30 pg of gDNA. The same aberration could be detected in the non-amplified control hybridization.**

# Single cell hybridized on Agilent 8x60K CGH microarrays: Entire Chr. 15 amplification

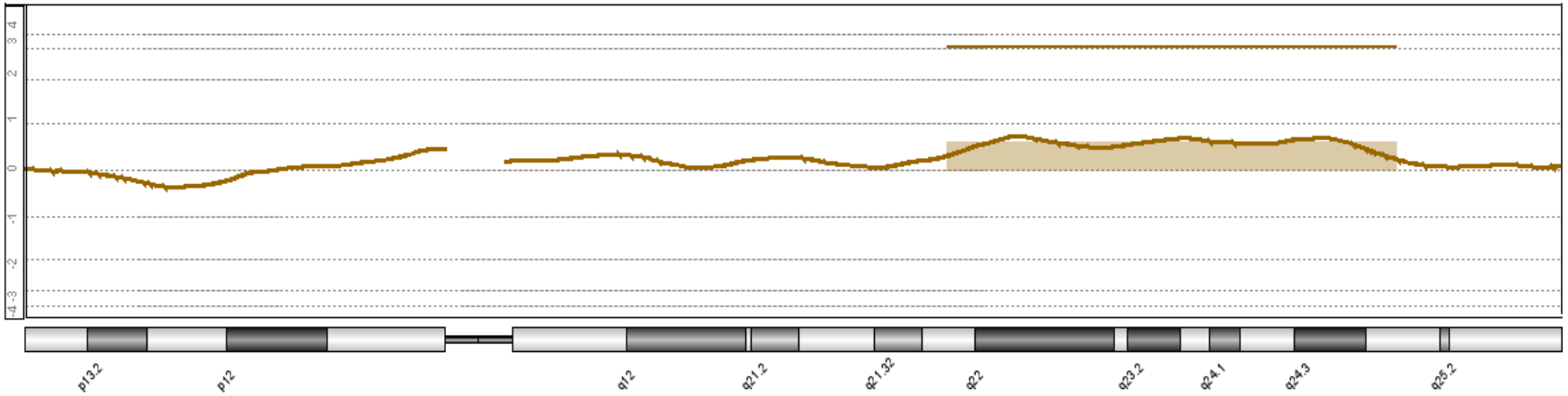
## Chr. 15



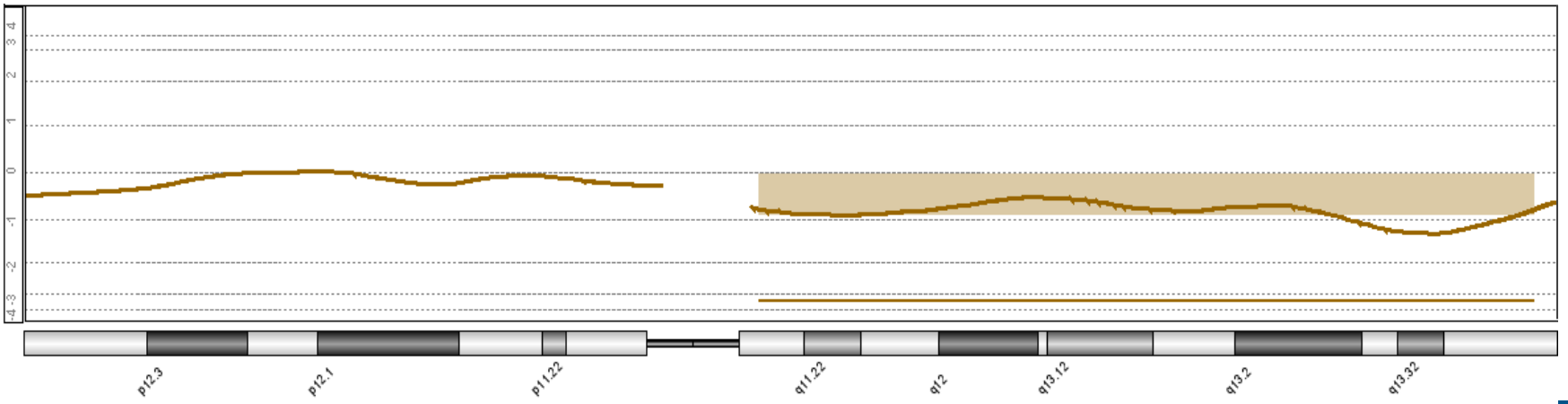

**Recommended single cell analysis settings: Log ratio moving average: data points smoothed to 5 Mb windows, Aberration filter: ADM2, threshold 6, minimum of 5 Mb aberrations, ≥0.3 log ratio for amplifications and ≥0.55 log ratio for deletions**

# Single cell hybridized on Agilent 8x60K CGH microarrays: Partial chromosome aberrations

## Chr. 17

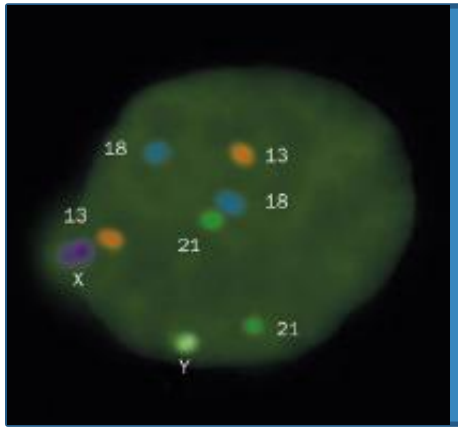



## Chr. 20



# Agilent 8x60K CGH microarrays vs. FISH on single cells

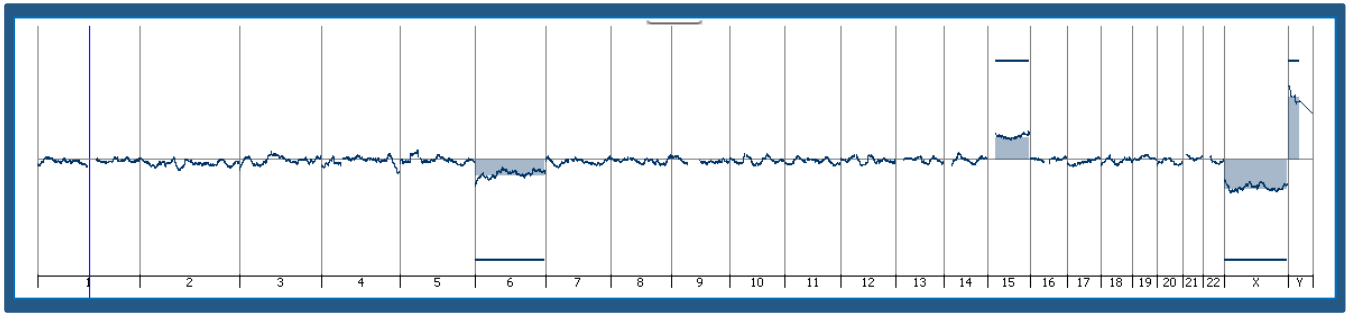
**FISH**: The number of chromosomes that can be analyzed simultaneously is **limited** by the number of fluorochromes available



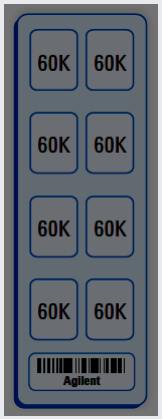
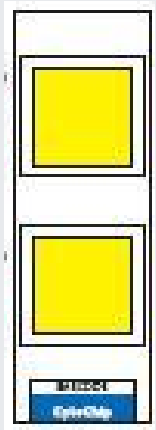

Result of five-color FISH applied to nuclei of single blastomere

Lancet 2004; 363: 1633–41

**Oligo aCGH:** The **entire genome** can be interrogated at once

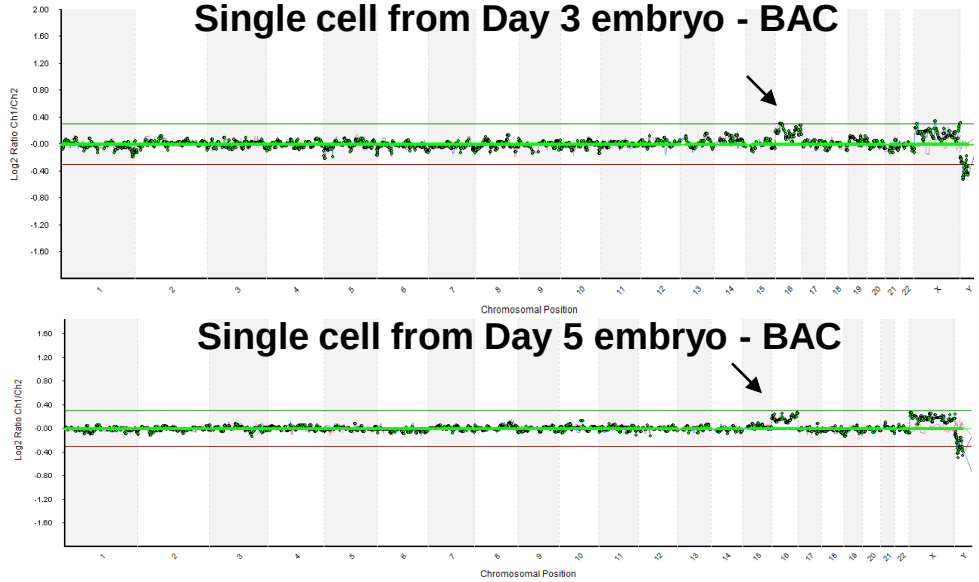


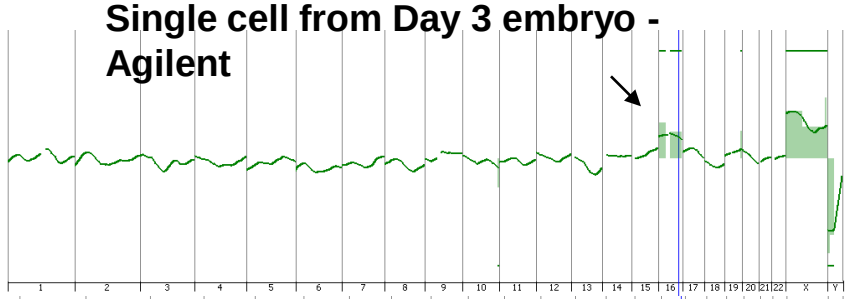
# Agilent 8x60K CGH microarrays vs. BAC

| Description | Agilent 8x60K CGH microarrays | BAC arrays |
|---|---|---|
| Number of probes – **higher resolution** | 60,000 | 3,000 |
| Number of samples per slide – **cost effective** | 8   | 2  |
| Type of probe – **robust** | *In-situ* synthesized 60-mer probes | Spotted BAC probes |
| Customization possible | **Yes** (through eArray) | No |

# Data comparison Agilent CGH arrays vs. BAC (data generated by Dr. Richard Choy CUHK)

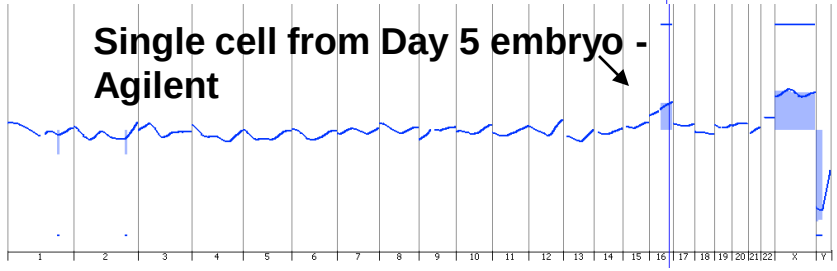
## Aneuploid embryo (47,XX,+16)



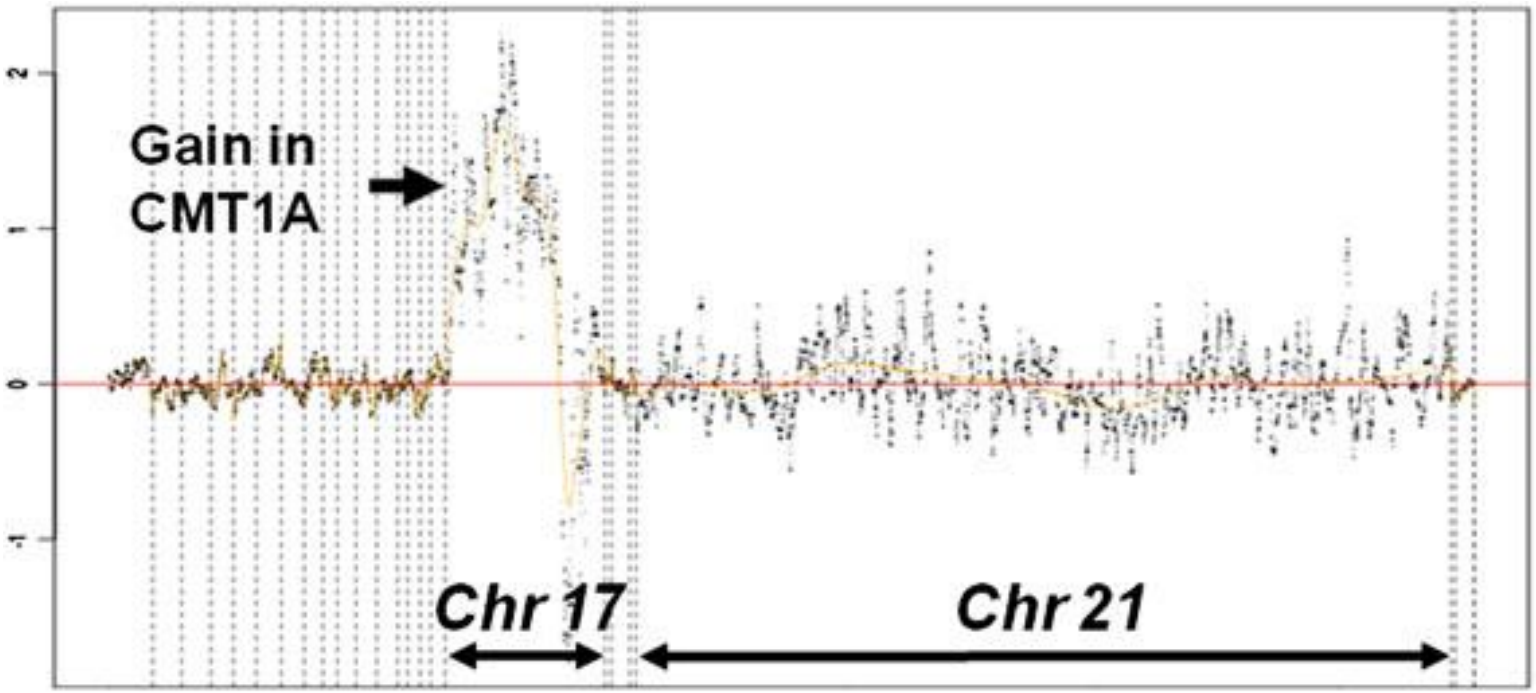







**Consistency of the results is demonstrated across the platforms, as well as among the cells derived from the same embryo.**

# Example of detection of smaller aberration compared to BAC arrays: 1.3Mb dup in CMT1A



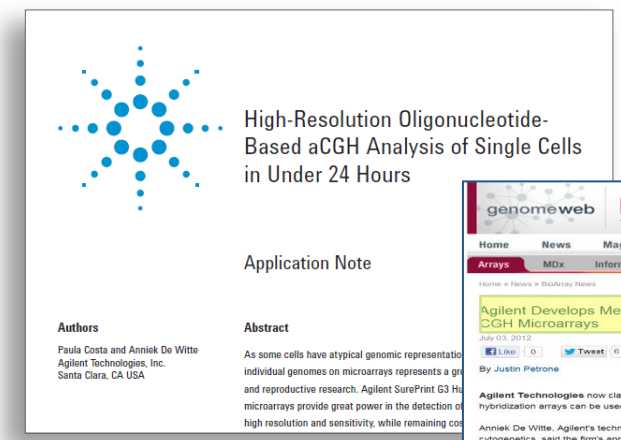

**Array CGH of a single cell with a 1.3Mb CMT1A duplication on the customized NIP2 array that has 20,000 probes in this region. The detection of the 1.3Mb copy number gain in a single CMT1A cell was clearly visualized after customized data analysis. The X-axis represents alignment of chromosomes 1 to 22; X and Y are not shown on this rendering. The Y-axis shows the normalized log ratios of the hybridization signals. Note that chromosome 21 takes up a disproportionately large area on the graph, because the array has ~100 K probes covering this chromosome. Data generated by Bi *et al.* Baylor College of Medicine.**

# Conclusions

- Genome-wide copy-number changes of **single cells** can be successfully profiled **within 24 hours** with Agilent oligo-based SurePrint G3 8x60K CGH microarrays
- The high-resolution genome-wide profiling allows for the cost-effective analysis of single cell whole genomes, overcoming the limitations associated with FISH and BAC arrays

# Additional Information

Poster

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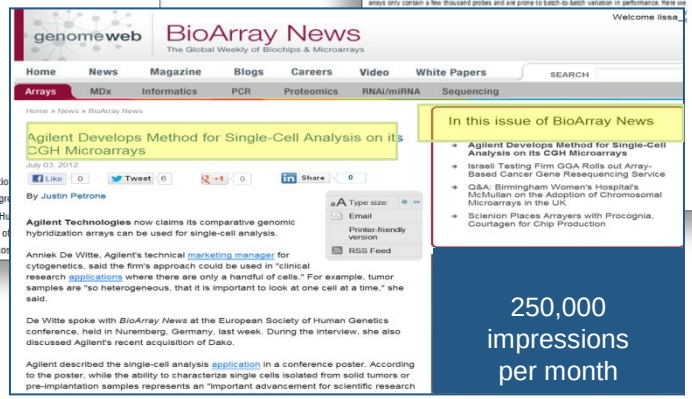
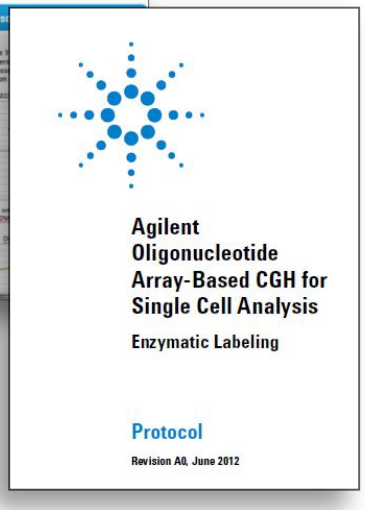



GenomeWeb article




P/N G4410-90012

## Peer Reviewed Papers

- Bi *et al.* Prenatal Diagnosis 2012;32:10–20.
- Cheng *et al.* Genome Biology 2011;12:R80.
- Hellani *et al.* Reproductive BioMedicine 2008;17(6):841-847.
- Traversa *et al.* Reproductive Biology 2011;11(3):51

Example data available on request

# Live demo of analysis of single cell data in Agilent CytoGenomics 2.5 software

# Questions