RESEARCH AREAS

Single-cell technology can be used in a **wide range of research areas**:

- Cancer biology
- Neuroscience
- Inmunology
- Developmental biology
- Biology of stem cells

SAMPLE REQUIREMENTS

Single-cell technology requires cells to be **individualized**.

	Single-cell gene expression	Fixed RNA profiling
Initial number of total cells	-	>300.000 cells >500.000 nuclei
Cell viability	>95%	>80% [1]
Optimal concentration (cells/µl)	700-1.200	400-4.000 [2] (single plex) 500-6.000 [2] (4 plex) 500-10.000 [2] (16 plex)
Recommended cell number per sample	500-10.000	500-10.000 (single plex) 500-10.000 (4 plex) 500-8.000 (16 plex)
Cell size	<30 µm	<30 µm
Recommended reads per cell	20.000	10.000
Organisms	Eukaryotes	Human and mouse
Shipping to NIMGenetics	Fresh [3] / Cryopreserved [4]	>300.000 cells >500.000 nuclei

[1] *Cell viability before fixation.*
[2] *Depending on the target.*
[3] *10X recommends immediate cold shipping, preferably within 30 min of individualization. Consult.*
[4] *In cryopreserved cells, a loss of 50% of viable cells over the original number of cells is assumed.*

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SINGLE-CELL
Individual sequencing for global thinking

NIMGenetics
New Integrated Medical Genetics

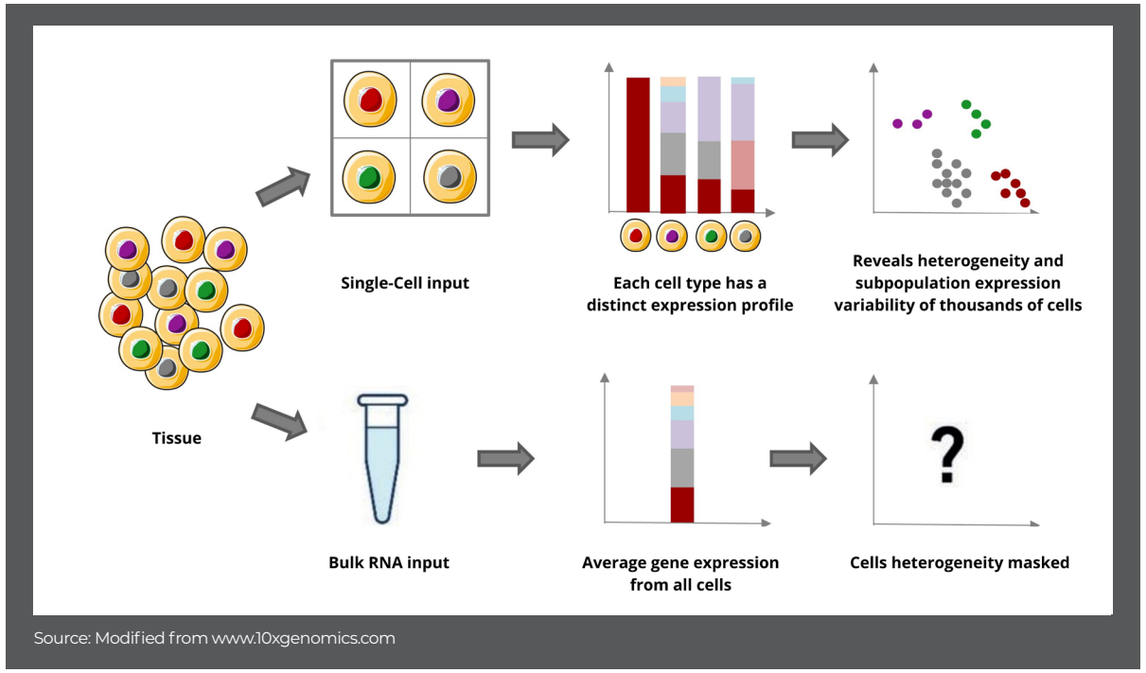
***Single-cell* RNA-Seq** technology allows the analysis of transcriptomes at the level of a single cell to discover sample heterogeneity. This solution facilitates the study of previously inaccessible information since it can analyze, in addition to transcriptomes, epigenomes and immune repertoires with the **resolution of individual cells**.

What are the **advantages** of *single-cell* RNA-seq compared to conventional RNA-seq?

Sequencing technologies require a cell lysis step in which the genetic material of all the cells contained in the sample is mixed in the same tube to make the sequencing libraries.

Most samples have a heterogeneous cellular composition, therefore, the results obtained from a conventional sequencing, and specifically from a bulk RNA-Seq, represent the average expression of all cells.

However, the single-cell RNA-Seq allows to individualize the cells so that the specific expression of each of the cells can be analyzed which permits to identify all the cell types contained in a sample.

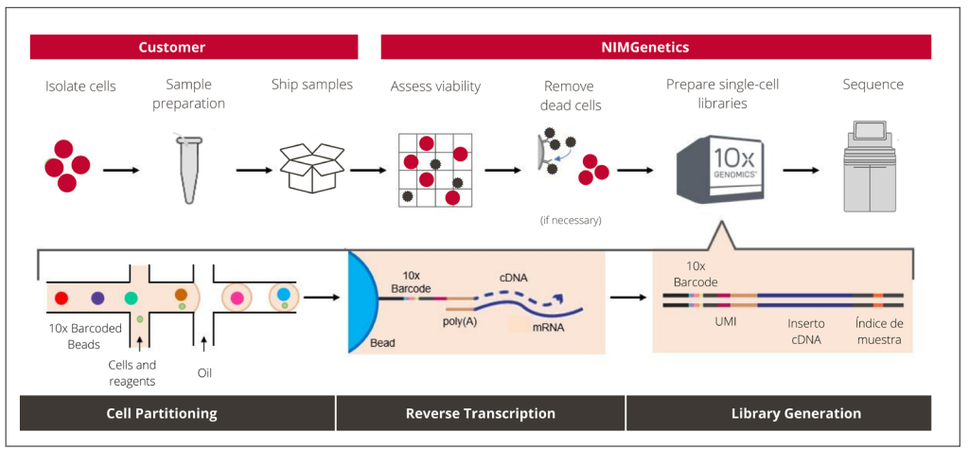


Source: Modified from www.10xgenomics.com

*NIMGenetics offers to researchers the **single-cell service**, both in **fresh** and **fixed** cells.*

Single-Cell Gene expression

The ***single-cell* RNA-Seq** application provides transcriptional profiles that facilitate researchers to understand at single-cell level the individual gene expression profiles and how they differ among the thousands of cells contained in a sample.



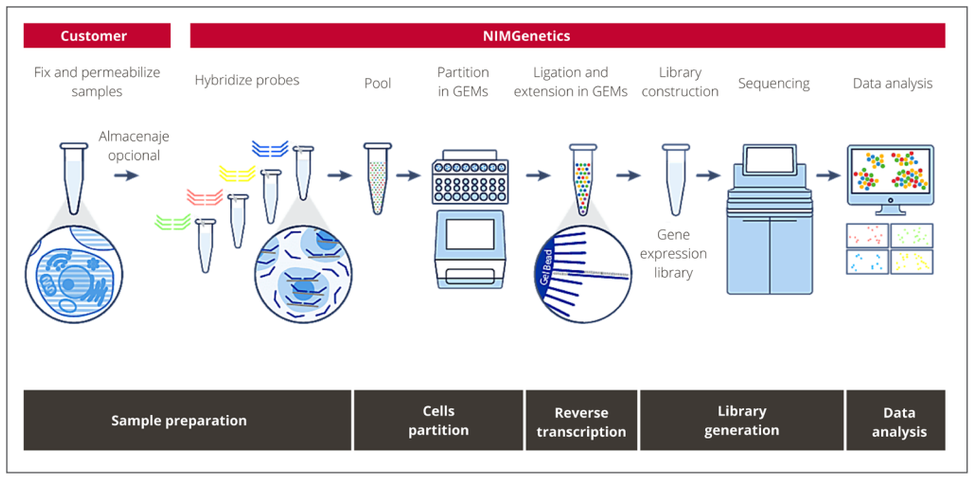
Source: Own design and www.10xgenomics.com

- In this application the fresh cells are encapsulated in GEMs where once cell lysis occurs the polyA tails of the mRNA molecules are captured.
- Subsequently, a cDNA labeled with a barcode that identifies the original cell and a unique molecular identifier (UMI) that refers to each mRNA transcript is generated.
- Finally, the cDNA of each cell is amplified and an Illumina® adapter containng a sample identifier is added. The generated libraries are then sequenced in our NovaSeq® 6000 equipment and the data is easily analyzed by specific software.

Fixed RNA-profiling

The ***fixed RNA-profiling technique*** brings the opportunity to study the complete transcriptome from single cells based on probe hybridization.

The **Chromium iX** instrument enables to work with cells fixed at the time of collection thus solving one of the main barriers that single-cell technology presented until now.



Source: Modified from www.10xgenomics.com

- This modality manages to preserve that biological information most susceptible of being deteriorated.
- This allows the study of single-cell gene expression in samples that were previously inaccessible due to logistical challenges in handling, transporting and storing samples.