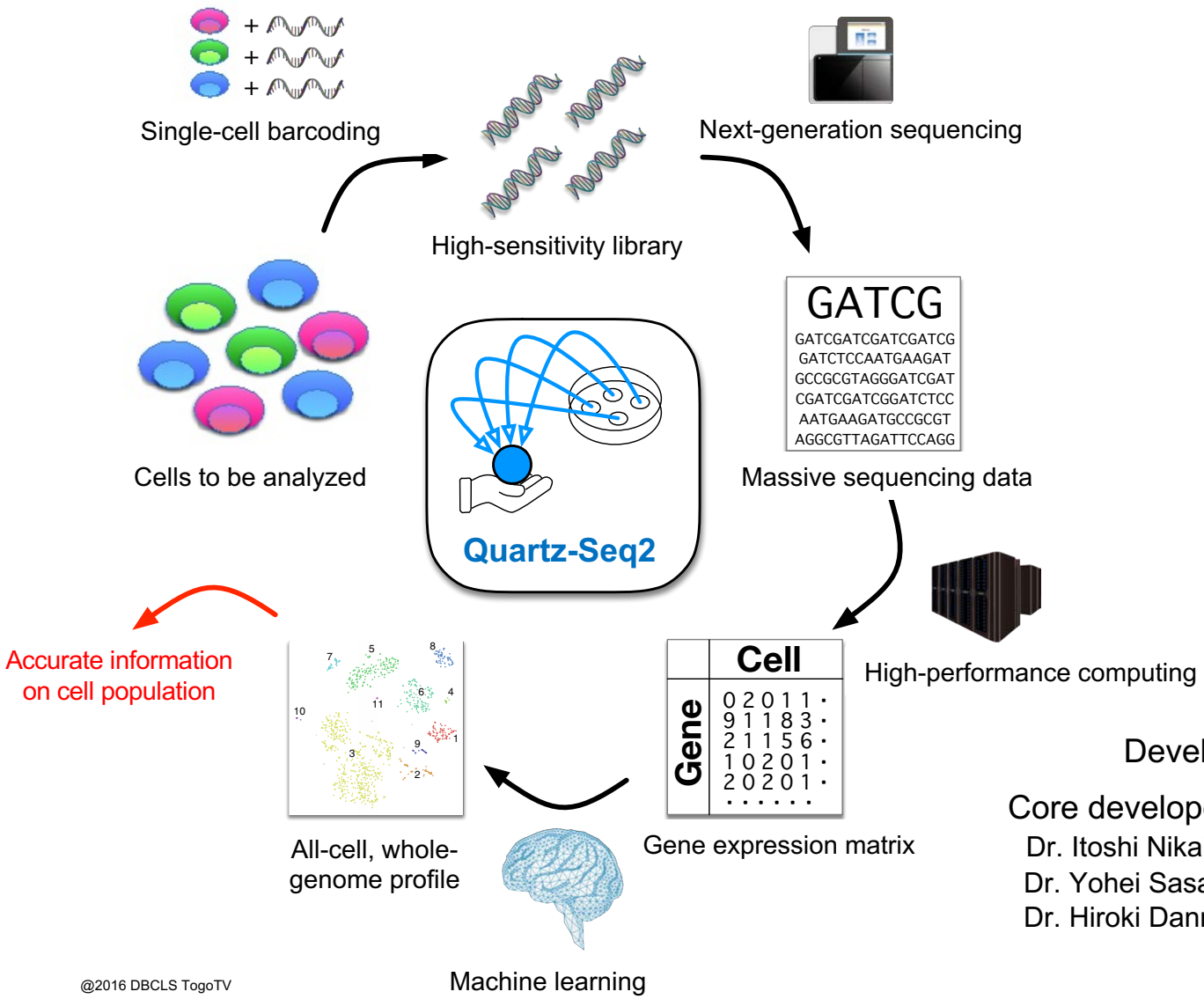
Knowledge Palette, Inc.

Company Introduction Materials

June 2021

knowledge palette

Single-cell transcriptome analysis technology developed by RIKEN



This technology combines single-cell barcoding, next-generation sequencing, high-performance computing, machine learning, and bioinformatics to enable the determination of expression levels of all genes in each cell for populations of thousands of cells

Developed by Nikaido Lab of RIKEN

Core developers:

Dr. Itoshi Nikaido (RIKEN and Adviser for Knowledge Palette)
Dr. Yohei Sasagawa (RIKEN and Adviser for Knowledge Palette)
Dr. Hiroki Danno (RIKEN → Knowledge Palette Founder)

Three Features of KP's Core Technologies

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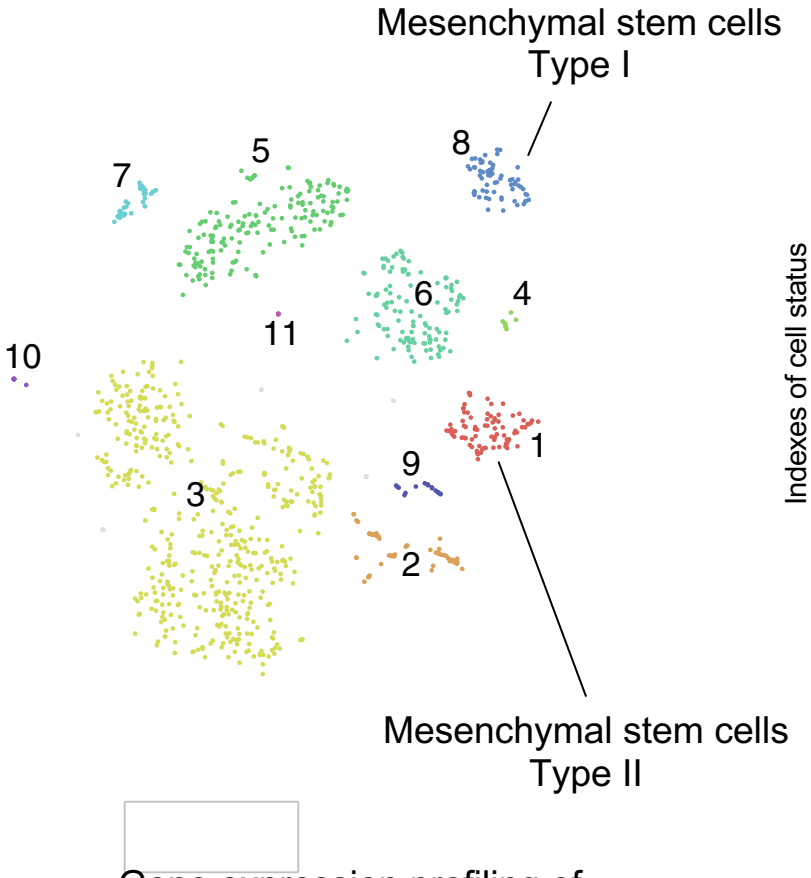
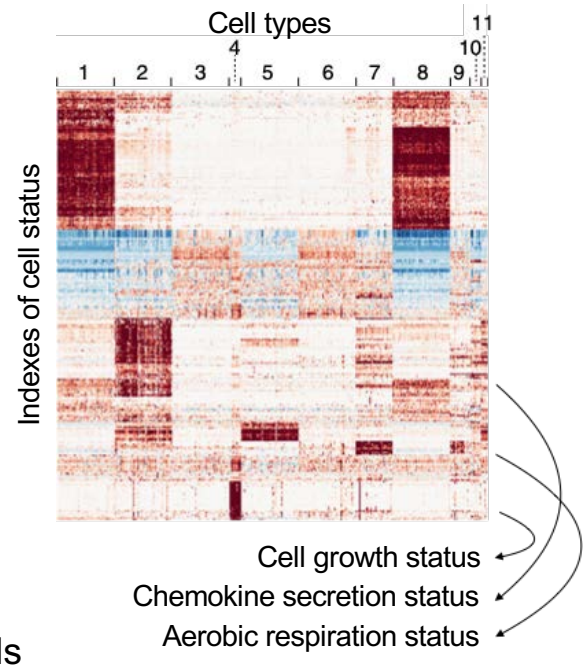
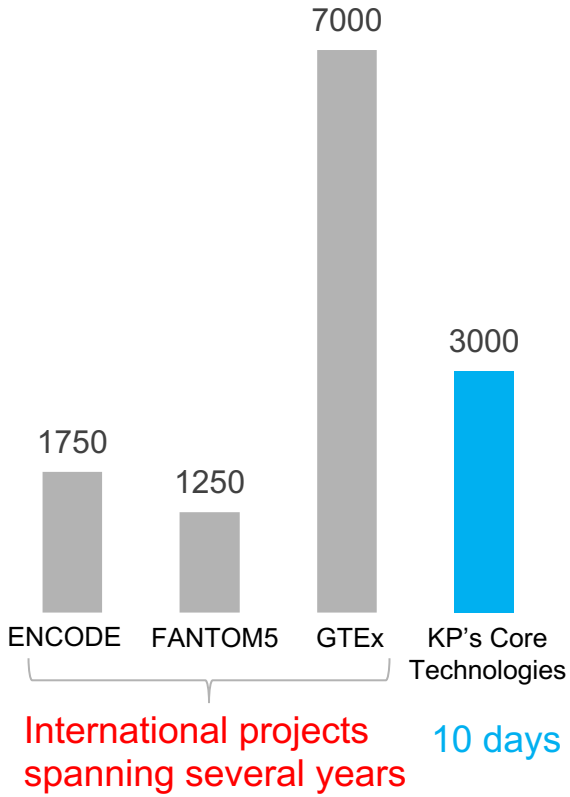
Resolution × Precision × High throughput



Gene expression profiling of individual cells



Accurate assessment of status of individual cells



Acquisition of transcriptome data for 3000 samples within 10 days at a single lab

nature biotechnology

ANALYSIS

https://doi.org/10.1038/s41587-020-0469-4

Check for updates

Benchmarking single-cell RNA-sequencing protocols for cell atlas projects

Elisabetta Mereu[1,26], Atefeh Lafzi[1,26], Catia Moutinho[1], Christoph Ziegenhain[2], Davis J. McCarthy[3,4,5], Adrián Álvarez-Varela[6], Eduard Batlle[6,7,8], Sagar[9], Dominic Grün[9], Julia K. Lau[10], Stéphane C. Boutet[10], Chad Sanada[11], Aik Ooi[11], Robert C. Jones[12], Kelly Kaihara[13], Chris Brampton[13], Yasha Talaga[13], Yohei Sasagawa[14], Kaori Tanaka[14], Tetsutaro Hayashi[14], Caroline Braeuning[15], Cornelius Fischer[15], Sascha Sauer[15], Timo Trefzer[16], Christian Conrad[16], Xian Adiconis[17,18], Lan T. Nguyen[17], Aviv Regev[17,19,20], Joshua Z. Levin[17,18], Swati Parekh[21], Aleksandar Janjic[22], Lucas E. Wange[22], Johannes W. Bagnoli[22], Wolfgang Enard[22], Marta Gut[1], Rickard Sandberg[2], Itoshi Nikaido[14,23], Ivo Gut[1,24], Oliver Stegle[3,4,25] and Holger Heyn[1,24] ✉

Single-cell RNA sequencing (scRNA-seq) is the leading technique for characterizing the transcriptomes of individual cells in a sample. The latest protocols are scalable to thousands of cells and are being used to compile cell atlases of tissues, organs and organisms. However, the protocols differ substantially with respect to their RNA capture efficiency, bias, scale and costs, and their relative advantages for different applications are unclear. In the present study, we generated benchmark datasets to systematically evaluate protocols in terms of their power to comprehensively describe cell types and states. We performed a multicenter study comparing 13 commonly used scRNA-seq and single-nucleus RNA-seq protocols applied to a heterogeneous reference sample resource. Comparative analysis revealed marked differences in protocol performance. The protocols differed in library complexity and their ability to detect cell-type markers, impacting their predictive value and suitability for integration into reference cell atlases. These results provide guidance both for individual researchers and for consortium projects such as the Human Cell Atlas.

Mereu, et al. *Nature Biotechnology* (2020)

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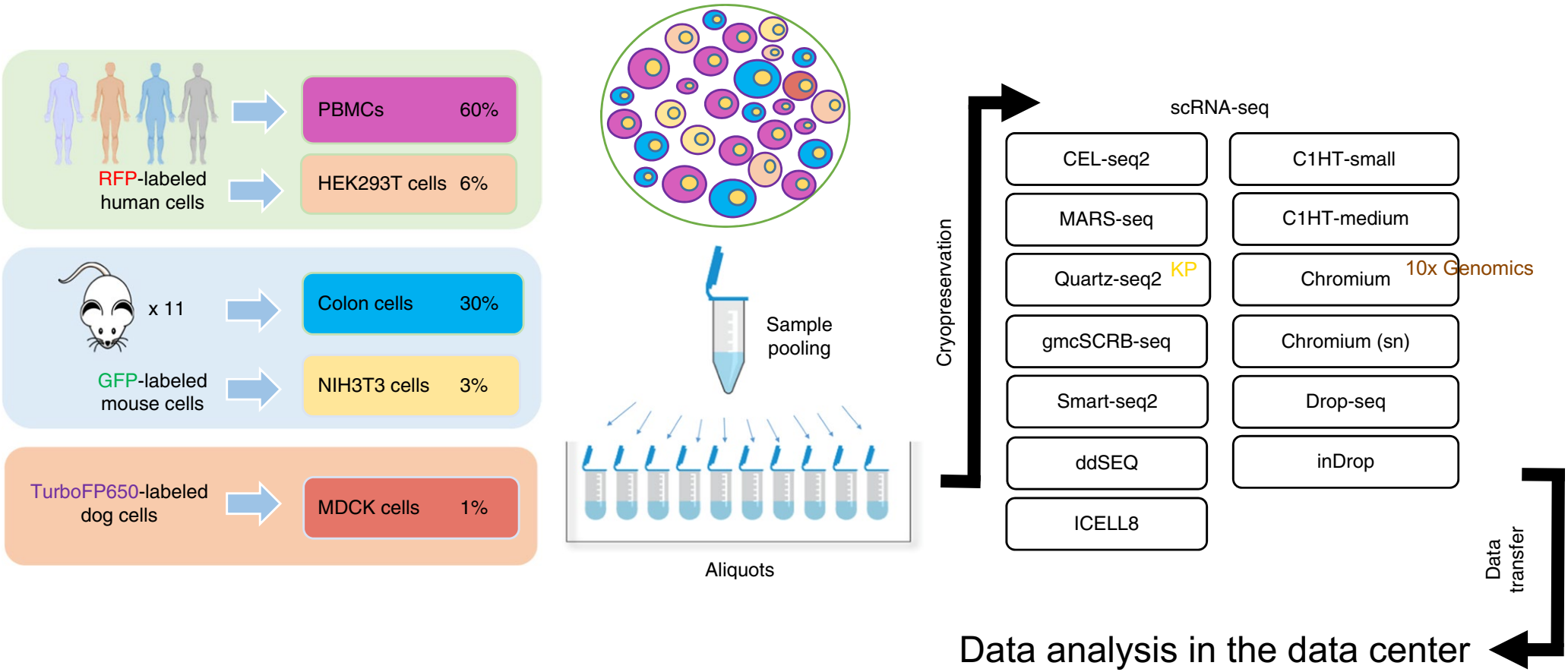
Cell mixture preparation



Mereu, et al. *Nature Biotechnology* (2020)

Quartz-Seq2 Reaches Top International Benchmark

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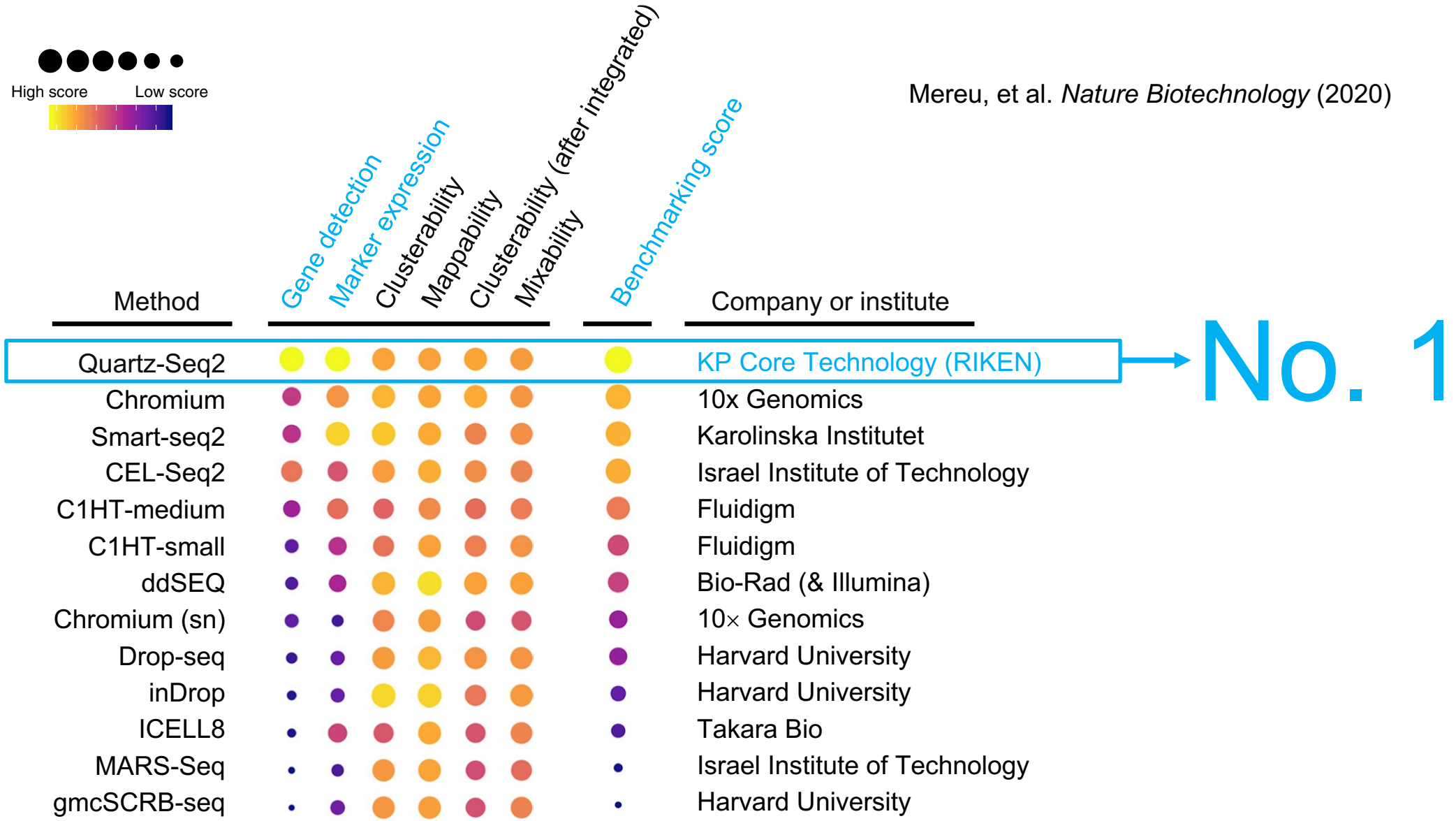
In international benchmarking of single-cell transcriptome analysis in the Human Cell Atlas project, our core technology received 1st place in both accuracy scores and overall scores.



Mereu, et al. *Nature Biotechnology* (2020)

Quartz-Seq2 is Particularly Advanced in Gene Detection

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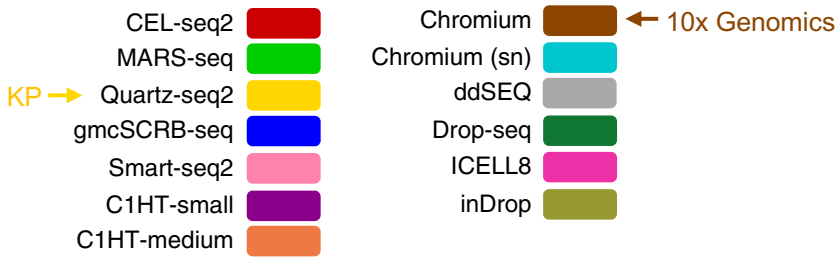
Numbers of genes detected (=index of accuracy) by each technology were compared in an international benchmarking effort. Quartz-Seq2 technology ranked number one for the number of genes detected in each of the 5 types of cell considered.

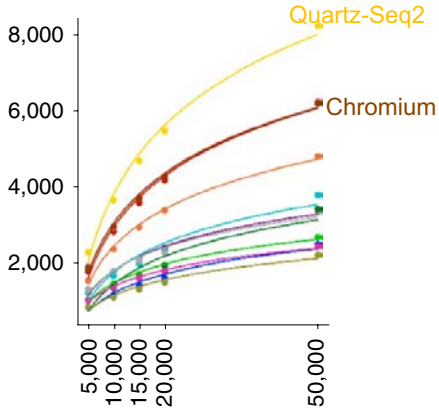
Colors used in plots



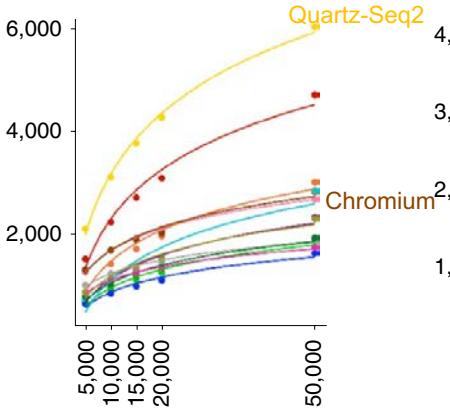
Number of detected genes

Cultured cells | Blood cells | Cells from tissue

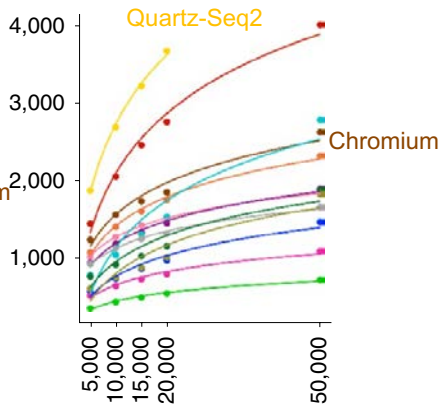
HEK293T



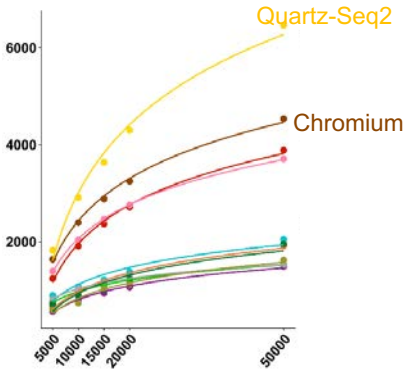
Monocytes



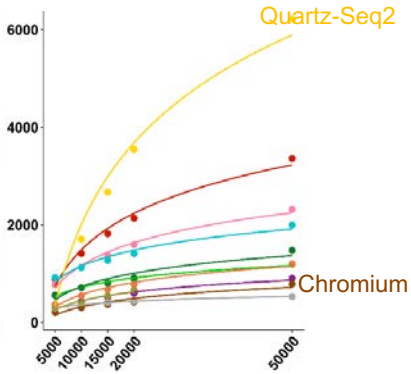
B cells

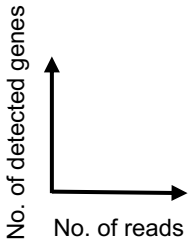


Secretory cells



Transit amplifying cells





High accuracy
Low cell-type dependency

Figures from Mereu, et al. *Nature Biotechnology* (2020)

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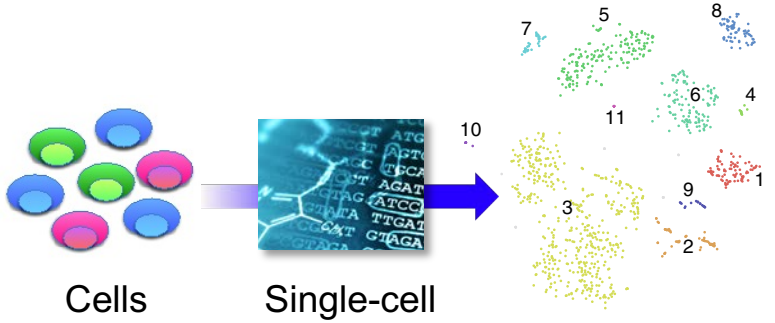
Single-cell Quartz

- High performance single-cell transcriptome technology



Cells

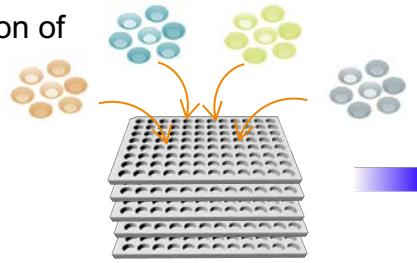
Single-cell level gene expression profiling

Specification of cell types and functions

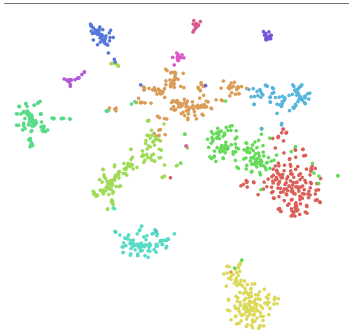
Bulk Quartz

- Ultra high throughput bulk transcriptome technology
  – Applicable for phenotypic drug discovery

Cells after administration of various compounds



384-well plate x 20~50

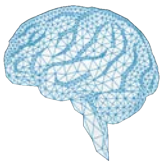


compounds/conditions profiling

Quartz AI

- High performance computing
- In house large transcriptome database and machine learning method

High performance computing



Machine learning

In 2018, Novartis developed DRUG-seq technology for whole transcriptome-based phenotypic drug discovery

ARTICLE

DOI: 10.1038/s41467-018-06500-x OPEN

DRUG-seq for miniaturized high-throughput transcriptome profiling in drug discovery

Chaoyang Ye[1,7], Daniel J. Ho[1], Marilisa Neri[2], Chian Yang[3], Tripti Kulkarni[4], Ranjit Randhawa[1], Martin Henault[3], Nadezda Mostacci[2], Pierre Farmer[2], Steffen Renner[2], Robert Ihry[1], Leandra Mansur[5], Caroline Gubser Keller[2], Gregory McAllister[6], Marc Hild[3], Jeremy Jenkins[6] & Ajamete Kaykas[1]

[1] Neuroscience Research, Novartis Institutes for Biomedical Research, 250 Massachusetts, Cambridge, MA 02139, USA. [2] Chemical Biology & Therapeutics Informatics, Novartis Institutes for Biomedical Research, Fabrikstrasse 22, 4056 Basel, Switzerland. [3] Chemical Biology & Therapeutics, Novartis Institutes for Biomedical Research, 250 Massachusetts, Cambridge, MA 02139, USA. [4] Scientific Computing, Novartis Institutes for Biomedical Research, 250 Massachusetts, Cambridge, MA 02139, USA. [5] Analytical Sciences & Imaging, Novartis Institutes for Biomedical Research, 250 Massachusetts, Cambridge, MA 02139, USA. [6] Chemical Biology & Therapeutics Informatics, Novartis Institutes for Biomedical Research, 250 Massachusetts, Cambridge, MA 02139, USA. [7] Present address: Blueprint Medicines, 45 Sidney St, Cambridge, MA 02139, USA. Correspondence and requests for materials should be addressed to A.K. (email: ajamete.kaykas@novartis.com)

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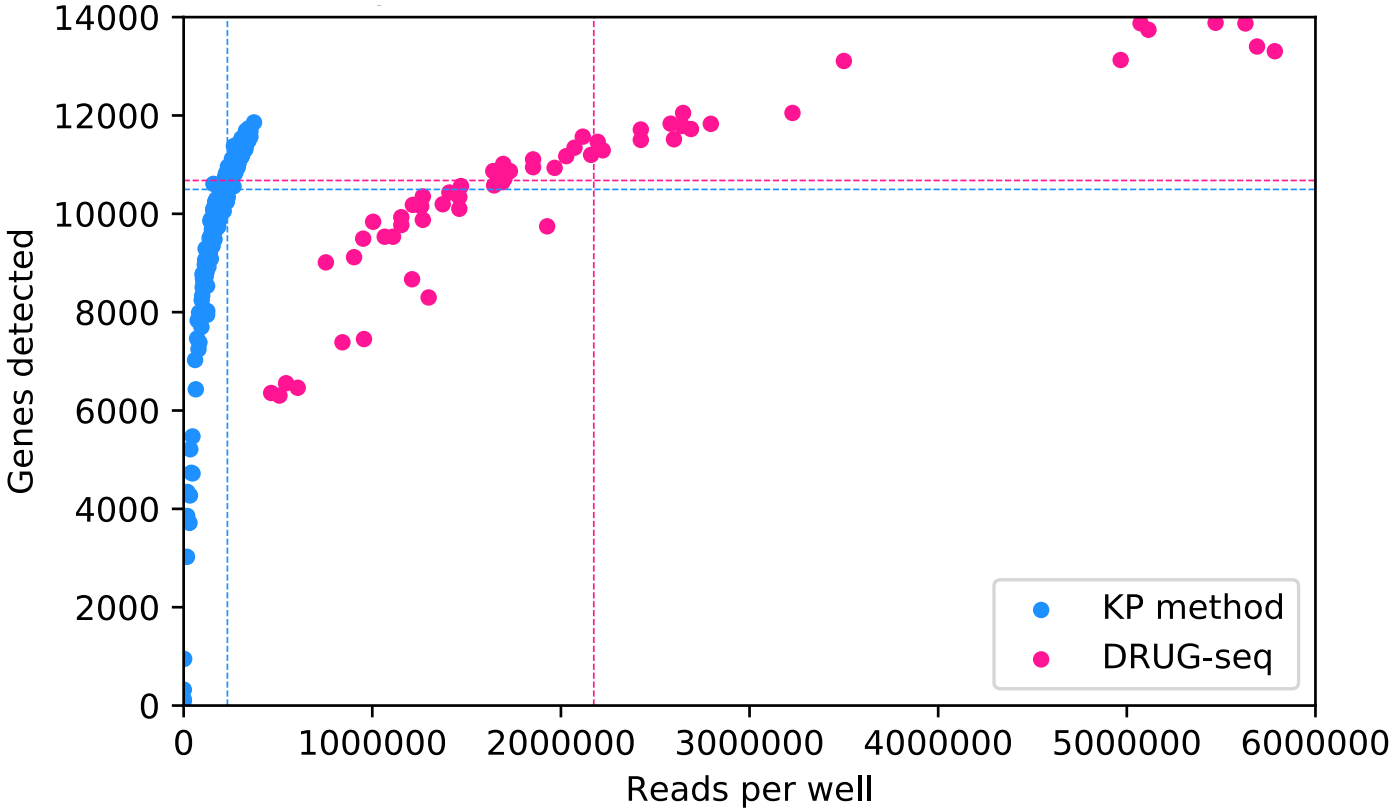
Data was obtained in the same condition (Cell line: U-2 OS, Conc.: 10 μM, time: 12 h)



(Ye, et al., Nat. Commun 2018 and our data)

KP's technology

230,000 reads per well

→ ~10,496 genes were detected

DRUG-seq

2,170,000 reads per well

→ ~10,678 genes were detected

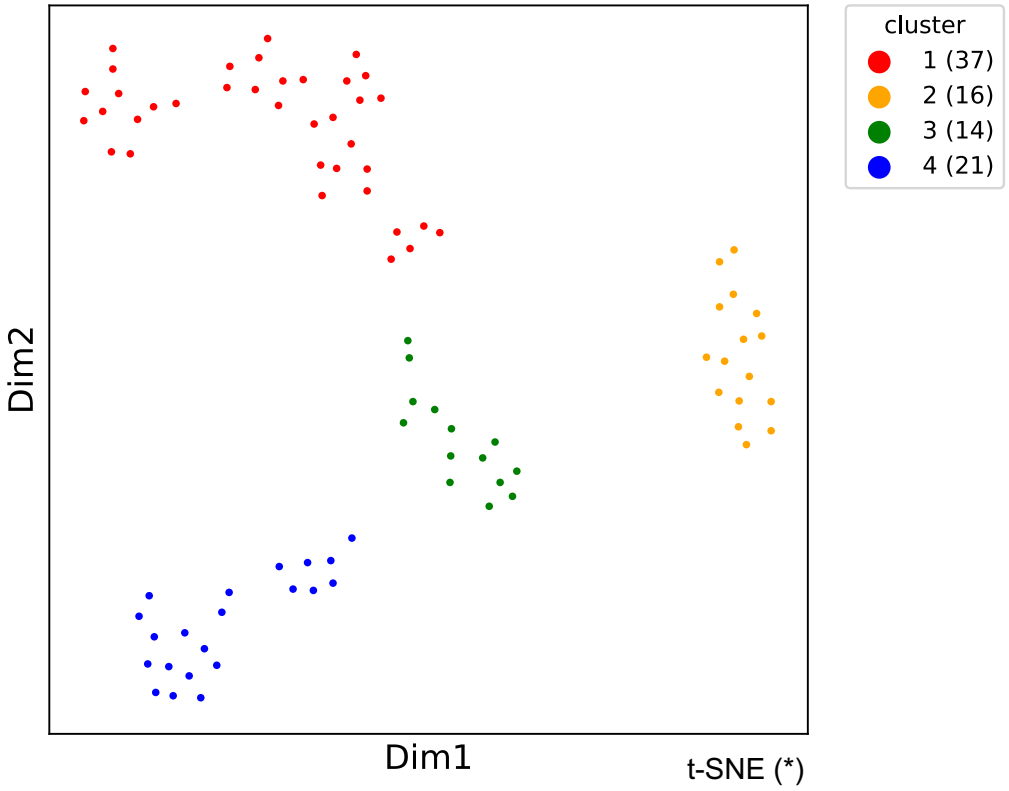
KP's technology can analyze 10 times more compounds in 1 run of NGS

knowledge palette

Data was obtained in the same condition (Cell line: U-2 OS, Conc.: 10 μM, time: 12 h)

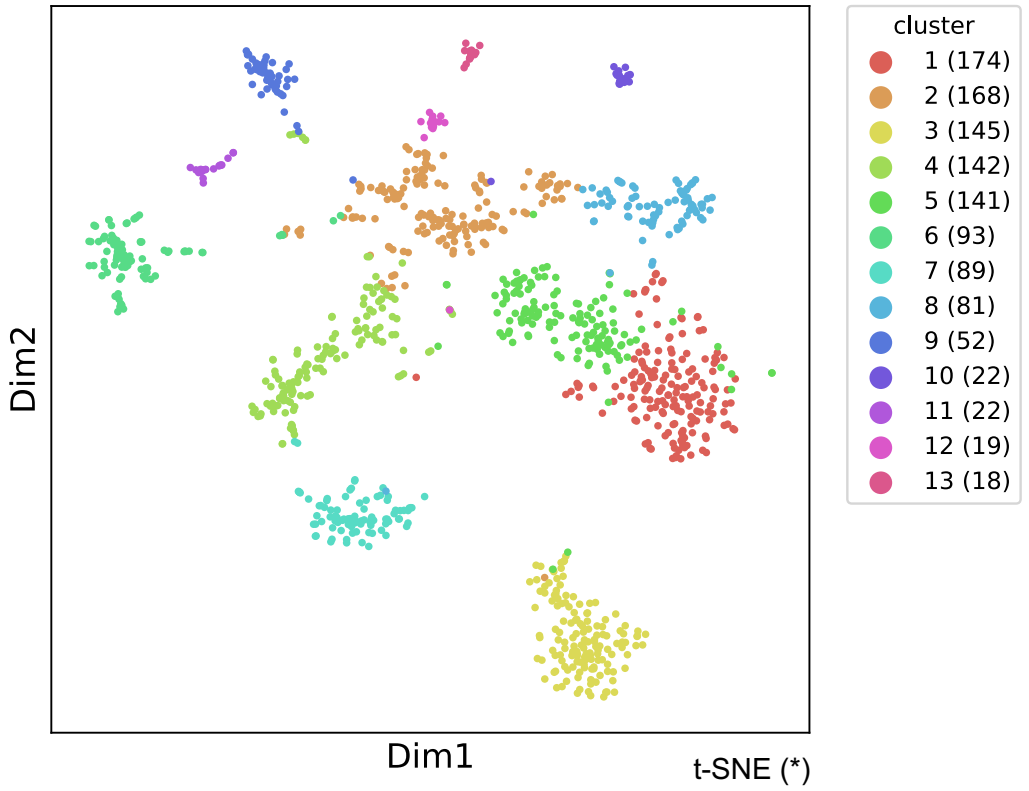
DRUG-seq

(Ye, et al., Nat. Commun 2018)



433 compounds (n=3)
→ 88 compounds caused signature change
→ 4 clusters identified (n=1 plotted)

KP's technology



1300 compounds (n=3)
→ 468 compounds caused signature change
→ 13 clusters identified (n=3 plotted)

＊ t-SNE

2 Identifying Stratification Biomarkers using Our Core Technologies

As our core technology enables us to analyze gene profile of cell population **by single cell with high accuracy**, we plan to identify **stratification biomarkers** which can distinguish successful patients precisely and design clinical trial

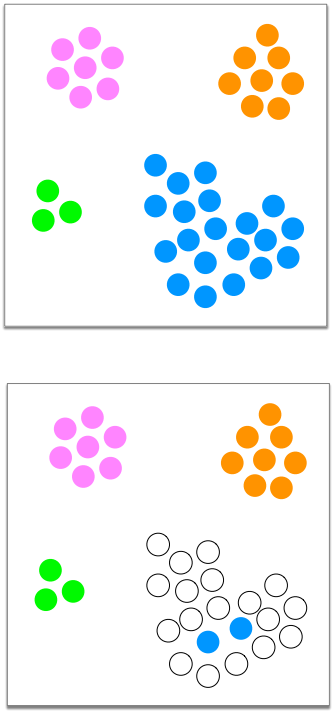
Identification flow of stratification biomarkers

Samples from patients / animals

Patients who show drug effectiveness

Patients who do NOT show drug effectiveness

High-precision single-cell transcriptome analysis



(example) Constituents of cell populations are compared at the resolution of a single cell

Optimization of clinical trial

Cell ● identified as drug target

Marker genes for cell ● identified

Design stratification and clinical trial