

Cell cell communications in spatial transcriptomics

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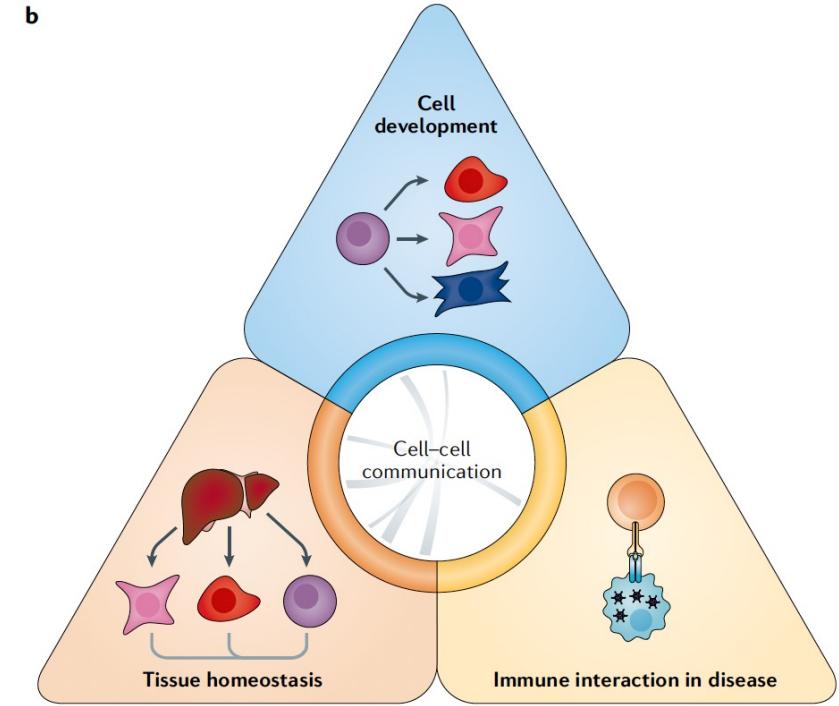
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Cell–cell interactions

- **Cell–cell interactions** orchestrate organismal development, homeostasis and single-cell functions.
- When cells do not properly interact or improperly decode molecular messages, **disease** ensues.
- Thus, the identification and quantification of **intercellular signalling pathways** has become a common analysis performed across diverse disciplines.
- The expansion of protein–protein interaction databases and recent advances in RNA sequencing technologies have enabled routine analyses of intercellular signalling from gene expression measurements of bulk and single-cell data sets.
- In particular, **ligand–receptor pairs** can be used to infer intercellular communication from the coordinated expression of their cognate genes.

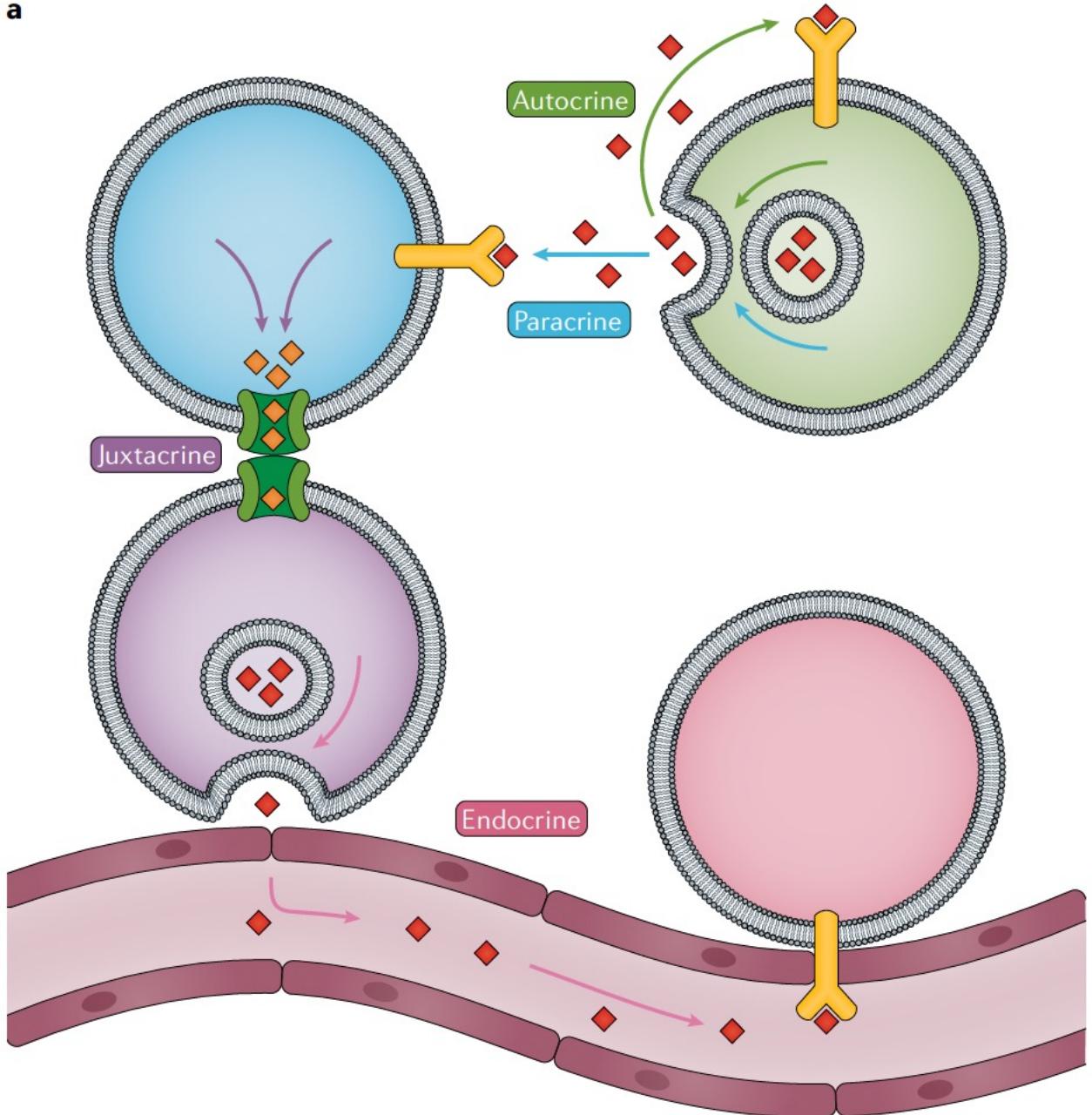
Interpretable

Armingol, E., Officer, A., Harismendy, O. et al. Deciphering cell–cell interactions and communication from gene expression. *Nat Rev Genet* 22, 71–88 (2021).

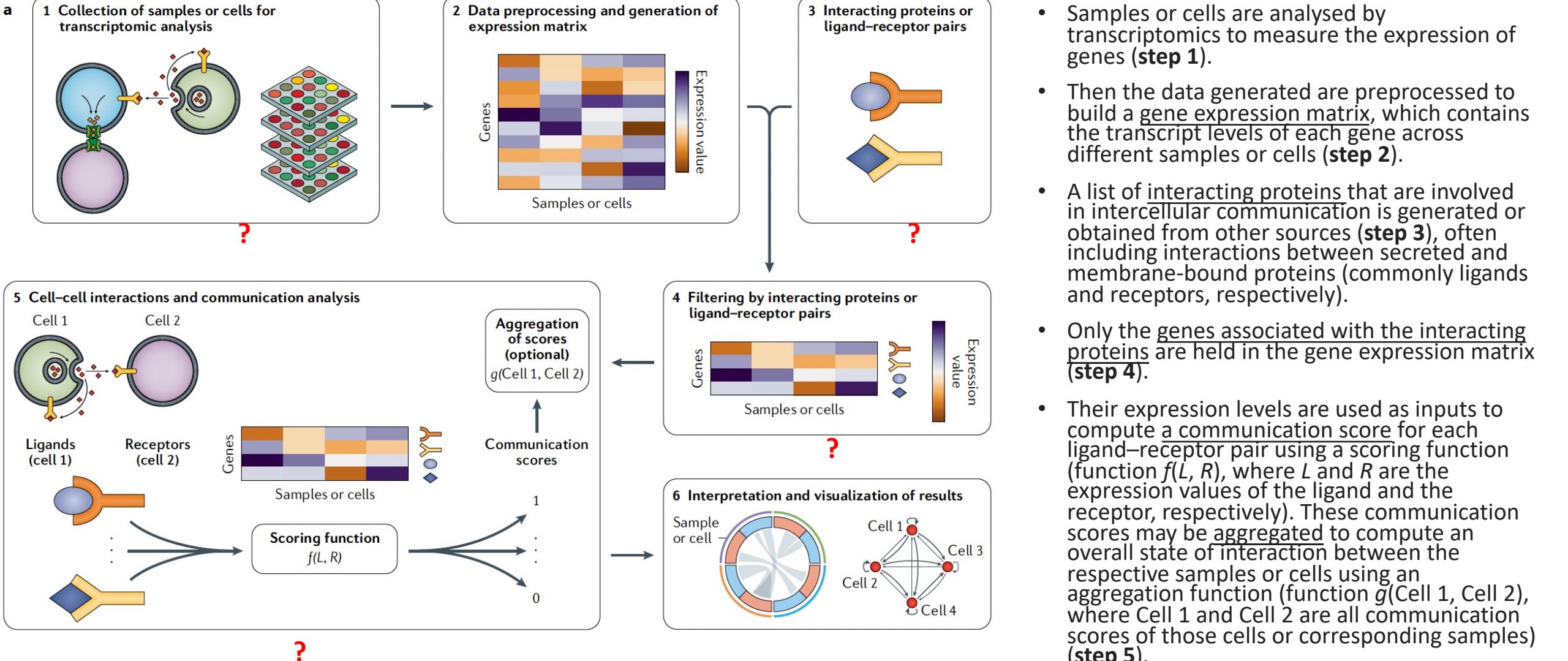


Overview of the main applications of cell–cell interaction methods: cell development, tissue and organ homeostasis, and immune interactions in disease

a



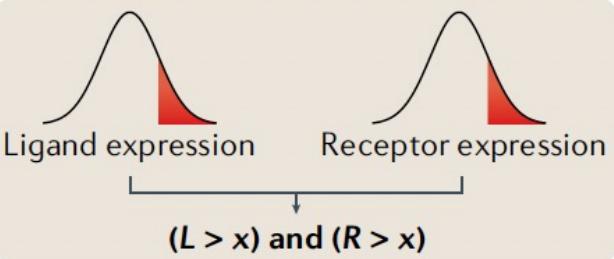
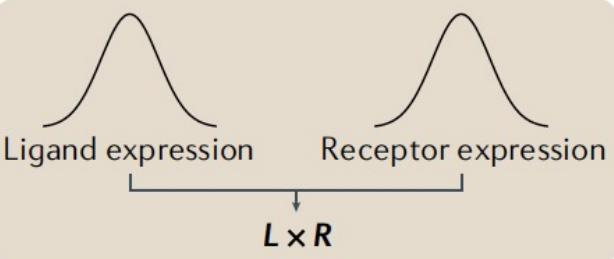
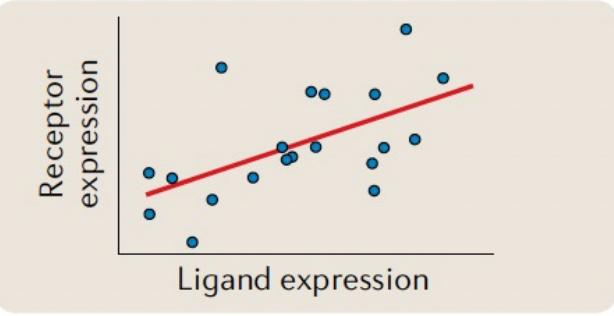
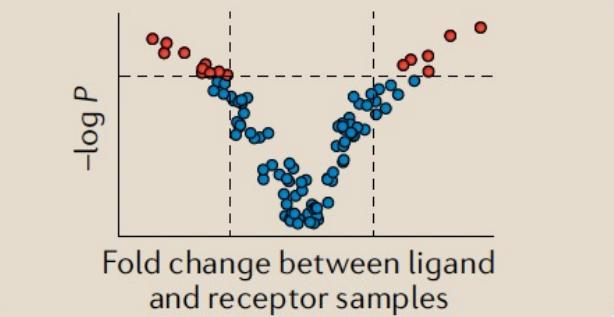
- a | ‘**Autocrine signalling**’ refers to intracellular communication whereby cells secrete ligands that are used to induce a cellular response through cognate receptors for those molecules expressed on the same cell. **Paracrine** cell–cell communication does not require cell–cell contact, rather depending on the diffusion of signalling molecules from one cell to another after secretion. **Juxtacrine**, that is, contact-dependent, cell–cell communication relies on gap junctions or other structures such as membrane nanotubes to pass signalling molecules directly between cells, without secretion into the extracellular space. **Endocrine** cell–cell communication represents intercellular communication whereby signalling molecules are secreted and travel long distances through extracellular fluids such as the blood plasma; typical mediators of this communication are hormones.



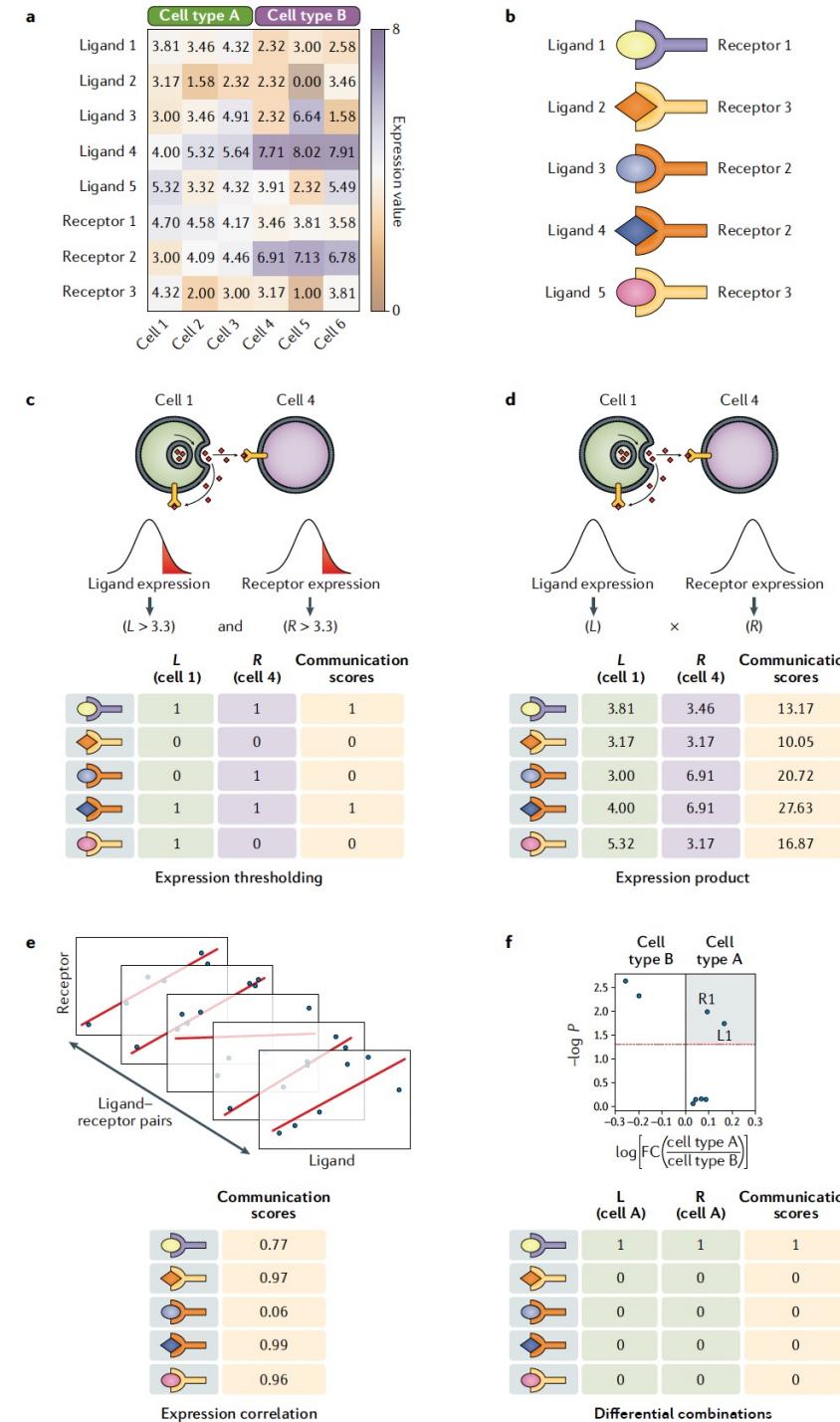
- Samples or cells are analysed by transcriptomics to measure the expression of genes (**step 1**).
- Then the data generated are preprocessed to build a gene expression matrix, which contains the transcript levels of each gene across different samples or cells (**step 2**).
- A list of interacting proteins that are involved in intercellular communication is generated or obtained from other sources (**step 3**), often including interactions between secreted and membrane-bound proteins (commonly ligands and receptors, respectively).
- Only the genes associated with the interacting proteins are held in the gene expression matrix (**step 4**).
- Their expression levels are used as inputs to compute a communication score for each ligand–receptor pair using a scoring function (function $f(L, R)$, where L and R are the expression values of the ligand and the receptor, respectively). These communication scores may be aggregated to compute an overall state of interaction between the respective samples or cells using an aggregation function (function $g(\text{Cell 1}, \text{Cell 2})$, where Cell 1 and Cell 2 are all communication scores of those cells or corresponding samples) (**step 5**).
- Finally, communication and aggregated scores can be represented by, for instance, Circos plots and network visualizations to facilitate the interpretation of the results (**step 6**).

Routine SRT analysis protocol

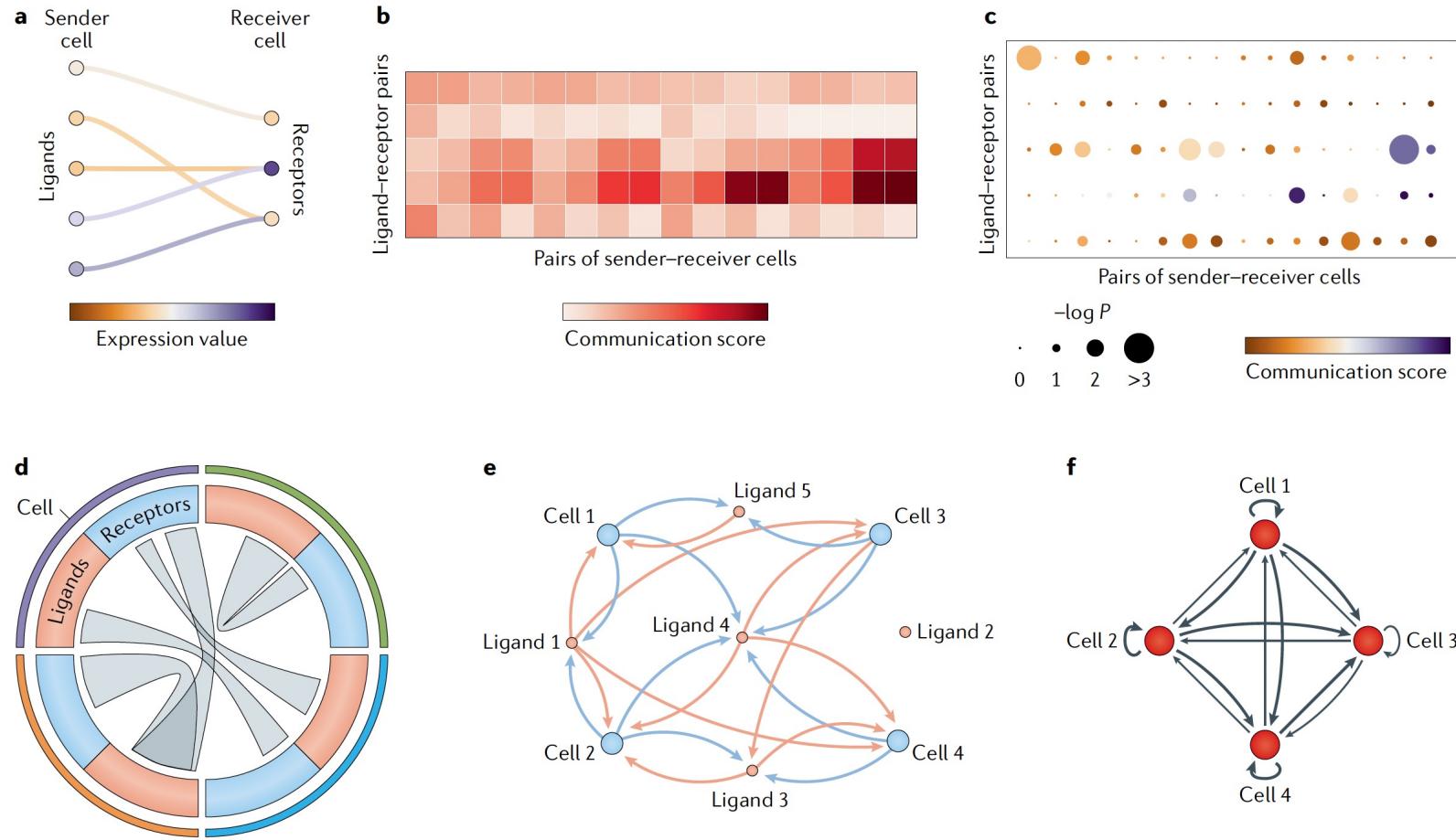
b

Expression thresholding		Recommended data	Communication score
Expression product		Bulk, single cell	Binary
Expression correlation		Single cell	Continuous
Differential combinations		Bulk, single cell	Binary

- Main scoring functions of communication pathways based on the expression of their components. Recommended data to use with these functions and the type of their resulting communication score are indicated.



- Two primary inputs are used for quantifying communication scores: a preprocessed gene expression matrix (part a) and a list of interacting proteins to supervise the analysis (for example, ligand–receptor pairs) (part b).
- Then a **communication score (CS)** can be computed for every ligand–receptor pair in a given pair of cells. Here, we show how to perform these calculations for four core functions (parts c–f). These are applied to elucidate **paracrine** (parts c,d) and **autocrine** (parts e,f) communication.
- To assess cell–cell communication, a CS can be computed for each ligand–receptor pair by accounting for the presence of both partners if their expression is greater than a given threshold, which for demonstrative purposes was **set arbitrarily to a value of 3.3** (part c), or by **multiplying their expression values** (part d).
- Similarly, the CS for each ligand–receptor pair can be the **correlation score** obtained from their expression across all cell types for **autocrine communication** (part e). To reveal non-autocrine interactions, the correlation can be computed across pairs of different cells. Particular signatures of each cell type can be extracted through analysing differentially expressed ligands and receptors. Using the cell type-specific differentially expressed genes, we can assign a **binary CS** and study the ligand–receptors used for **autocrine communication** (part f).
- In this example, autocrine communication is evaluated for cell type A by using its differentially expressed genes with respect to cell type B (cell type A-specific genes are located in the coloured quadrant). Analogously to the correlation score, for non-autocrine communication we would need to consider differentially expressed genes in each of the cell types or samples.
- For a given pair of cells, we can say that a communication pathway is active when the ligand is differentially expressed in one cell and its cognate receptor is differentially expressed in the other.



- **a** | A **Sankey diagram** for connecting key ligands from a sender cell to cognate receptors in the receiver cell. Node colour (ligand or receptor) indicates the expression level. **b** | **Heatmap** to represent the communication scores for each ligand–receptor interaction in each cell pair. **c** | **Dot plot** to show the communication score (colour of dots) and at the same time its significance (size), often obtained from a statistical model or permutation analysis. **d** | **Circos plot** or **chord diagram** to show key communication pathways used by different cell types to communicate. The links start from a ligand (red) and end in a receptor (blue), which are grouped for each cell type (coloured outer arcs). **e** | **Bipartite network** where nodes can be either cells or ligands. Edges can be directed only from a cell to a ligand it produces or from a ligand to a cell that expresses its cognate receptor. **f** | **Cell–cell interaction network** to represent the potential of cells to interact. Nodes correspond to cells and edges correspond to their interactions. These are directed from a sender cell to a receiver cell, and their thicknesses are proportional to the respective global cell–cell communication scores (for example, number of active ligand–receptor pairs).

CCC methods on scRNA-seq

- A major form of cell–cell interaction is **cell–cell communication (CCC)**, mainly mediated by biochemical signaling through ligand–receptor binding that induces downstream responses that shape development, structure and function.
- CellPhoneDB⁵, ICELLNET⁷ and CellChat⁶ account for the multi-subunit composition of protein complexes;
- SoptSC⁸, NicheNet⁹ and CytoTalk¹⁰ utilize downstream intracellular gene–gene interactions
- scTensor¹¹ examines higher-order CCC represented as hypergraphs.

Parts of the CCC methods

Tools	Method	Subunit	Prior knowledge	Language	Ref.
Statistical-based tools					
CellCall	Embedded pathway activity analysis for activity score; hypergeometric testing for significance of pathway activity	Single subunit	Ligand-receptor pairs; downstream TF regulation	R	[15]
CellChat	Law of mass action for communication probability; permutation test for significant interactions	Multi-subunit	Ligand-receptor pairs; signaling cofactors and pathways	R	[11]
CellPhoneDB	The mean of average ligand and receptor expression values for interaction enrichment; permutation test for significant interactions	Multi-subunit	Ligand-receptor pairs	Python	[10]
ICELLNET	Product of ligand and receptor expression values for communication score; geometric mean for multi-subunit complexes; Wilcoxon statistical test for highly potential interactions	Multi-subunit	Ligand-receptor pairs	R	[16]
iTALK	Finding differentially expressed ligand and receptor genes between cell types	Single subunit	Ligand-receptor pairs	R	[17]
SingleCellSignalR	Regularized product of ligand and receptor for lr-score; estimate lr-score cutoff for filtering interactions	Single subunit	Ligand-receptor pairs	R	[18]
Network-based tools					
Connectome	Cell types as nodes, interactions as edges; gene-wise z-score of ligand and receptor expression values as edge weights; system-wide Wilcoxon rank sum test for significant edges filtering	Single subunit	Ligand-receptor pairs	R	[19]
CytoTalk	Integrate two de novo intracellular signaling networks by known ligand-receptor interactions; optimal subnetwork searching for significant interactions	Single subunit	Ligand-receptor pairs	R	[20]
Domino	Construction global signaling network; cluster specific signaling subnetwork for prediction	Multi-subunit	Ligand-receptor pairs; TF regulation	R	[21]
NATMI	Cell types as nodes, interactions as edges; mean expression or specificity for edge weights; edge weight ranks for confident interactions	Single subunit	Ligand-receptor pairs	Python	[22]
NicheNet	Weighted network prior knowledge model; compute ligand activity and regulatory potential score using network propagation; select interactions by potential score	Single subunit	Ligand-receptor pairs; ligand-target pairs; receptor-target pairs	R	[12]
scMLnet	Construct primary ligand-receptor, TF-target, receptor-TF subnetworks using highly expressed genes; merge three subnetworks as final output	Single subunit	Ligand-receptor pairs; receptor-TF pairs; TF-target pairs	R	[23]
ST-based tools					
CellPhoneDB v3	L-R expression for enrichment; permutation test for significance; filter interactions based on spatial microenvironment	Multi-subunit	Ligand-receptor pairs; spatial microenvironment	Python	[13]
Giotto	Spatial proximity for interacting cell types; spatial co-expression for interactions	Single subunit	Ligand-receptor pairs; cell type colocalization; L-R co-expression	R	[24]
stLearn	Identify interactions by L-R co-expression and cell type density	Single subunit	Ligand-receptor pairs; cell type colocalization; L-R co-expression	Python	[25]

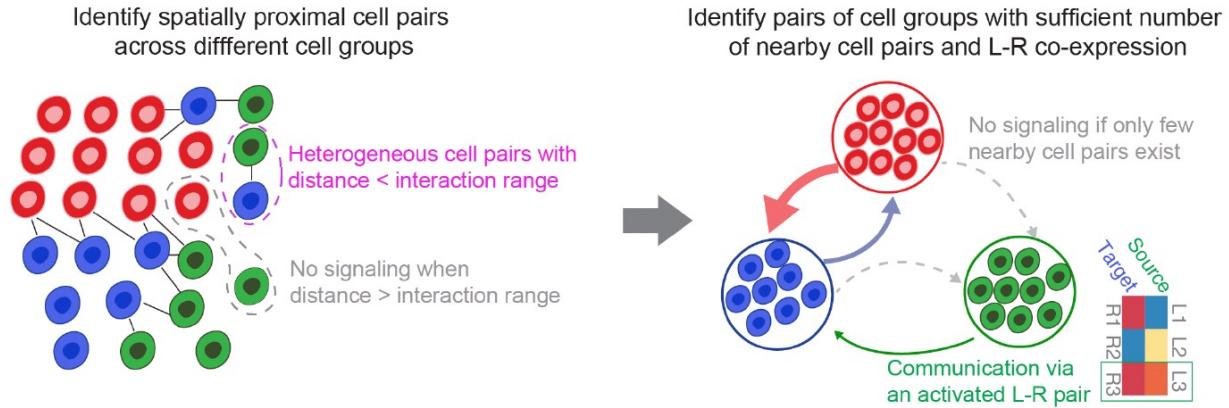
However, these non-spatial studies often contain significant false positives given that CCC takes place **only within limited spatial distances** that are not measured in scRNA-seq datasets.

Liu, Z., Sun, D. & Wang, C. Evaluation of cell-cell interaction methods by integrating single-cell RNA sequencing data with spatial information. *Genome Biol* **23**, 218 (2022).

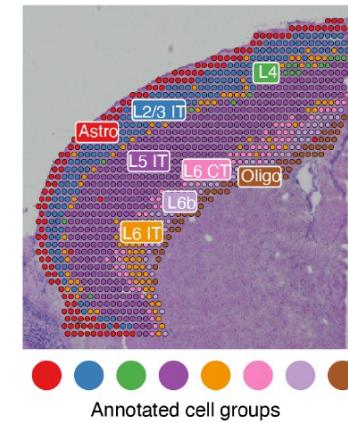
Spatial?

- Inference of cell-cell communication can be naturally extended to spatial context by **first identifying pairs of cells that are physically close to one another to have biologically realistic interactions based on maximal possible molecular interaction/diffusion ranges**, and then identifying combinations of cell groups that have enough nearby cell-cell pairs.
- The diffusive spatial distance of molecules depends on **many factors**, including molecule size, its covalent modifications, touristy of the spatial tissues, and the molecule's regulators on the cell membrane and in the extracellular environments. All these factors usually reduce diffusion. For example, large molecules have shorter diffusion distance, leading to more restricted spatial range in diffusion.
- CellChat v2 uses the ideal diffusion range in a free medium, that is the maximally allowable transport distance for small diffusive molecules (by default $250 \mu\text{m}$). In this way, CellChat v2 will not remove any interactions that are spatially plausible. For the contact-dependent signaling, the interaction range is restricted to the nearest neighbors of each cell, such that signaling and target cells are in direct contact.

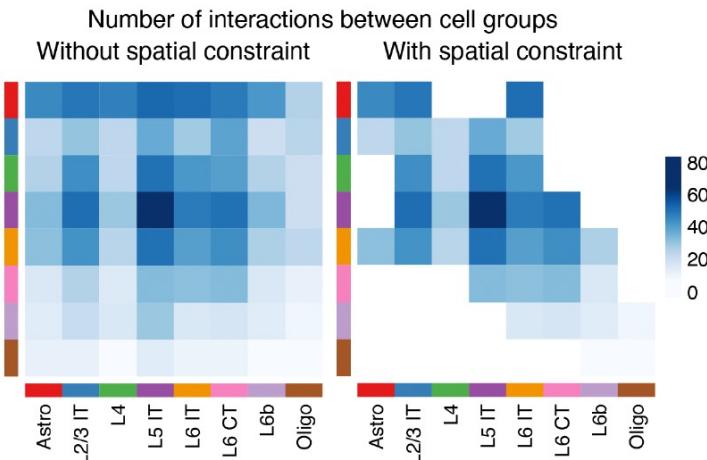
a Inference of spatially-proximal communication between cell groups



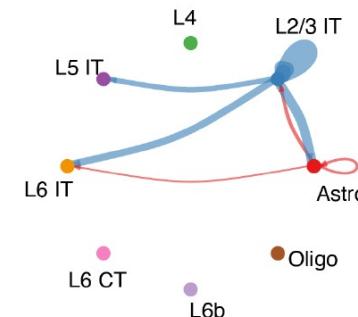
b Mouse cortex (10X Visium)



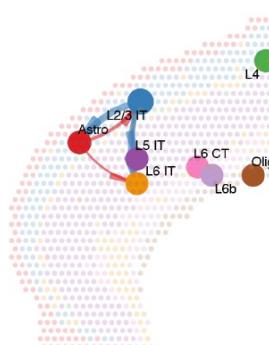
c



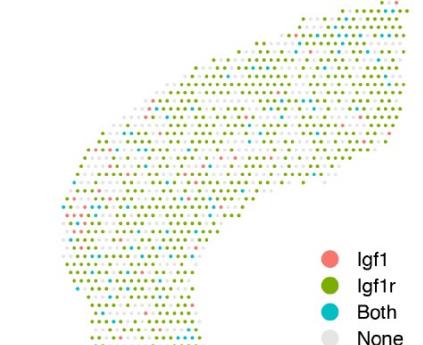
d Circle plot of IGF signaling



Spatial plot of IGF signaling

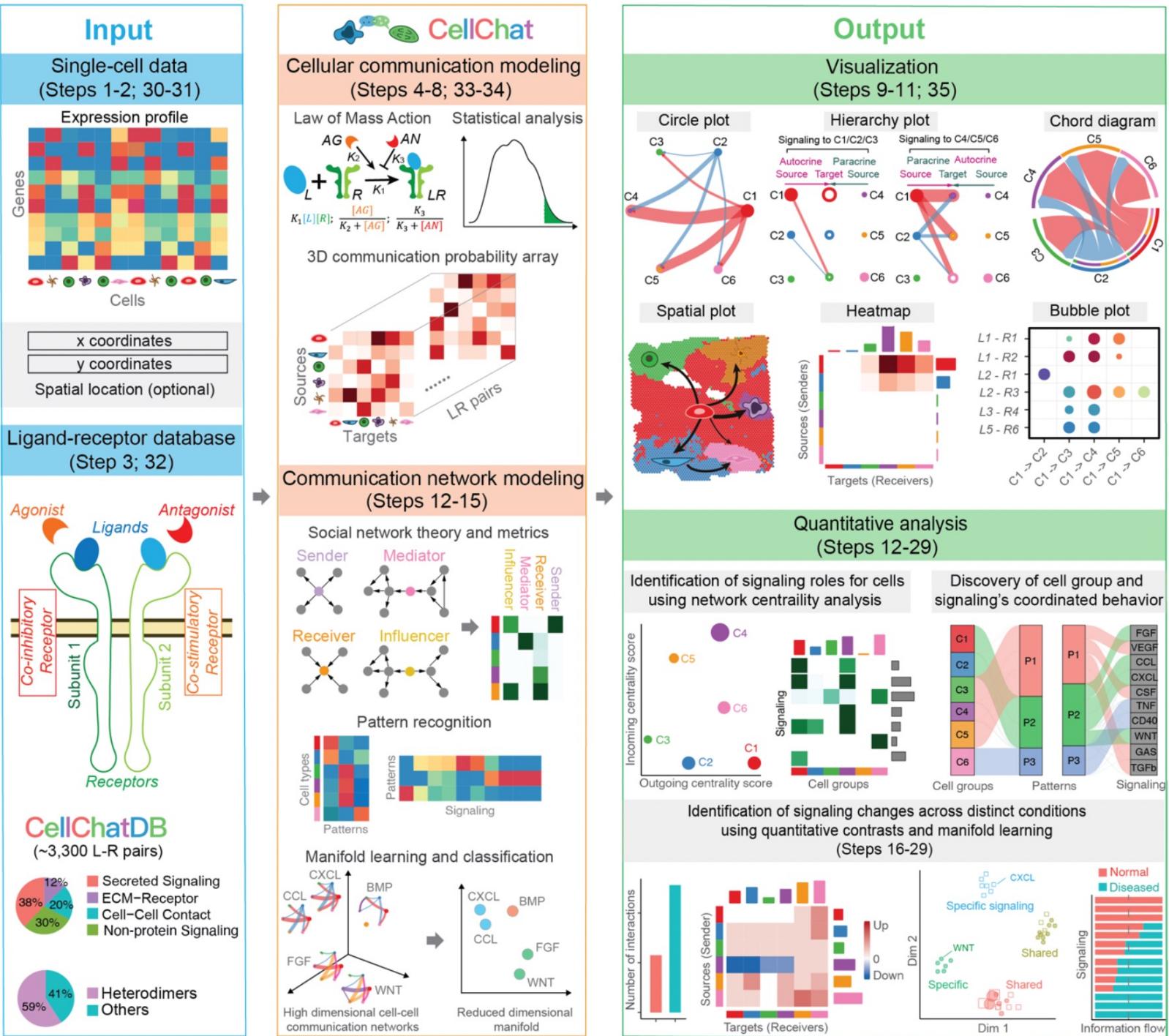


Expression of L-R pair IGF1_IGF1R



CellChat, CellChatDB, CellChat V2

- CellChat is an R package designed for inference, analysis, and visualization of cell-cell communication from single-cell and spatially resolved transcriptomics. CellChat aims to enable users to **identify and interpret cell-cell communication** within an easily interpretable framework, with the emphasis of clear, attractive, and interpretable visualizations.
- CellChatDB is a **manually curated database** of literature-supported ligand-receptor interactions in multiple species, leading to a comprehensive recapitulation of known molecular interaction mechanisms including multi-subunit structure of ligand-receptor complexes and co-factors.
- CellChat V2:
 - inference of spatially proximal cell-cell communication between interacting cell groups from spatially resolved transcriptomics
 - expanded database CellChatDB v2 by including more than 1000 protein and non-protein interactions (e.g. metabolic and synaptic signaling) with rich annotations. A function named updateCellChatDB is also provided for easily updating CellChatDB.
 - new functionalities enabling easily interface with other computational tools for single-cell data analysis and cell-cell communication analysis
 - interactive web browser function to allow exploration of CellChat outputs of spatially proximal cell-cell communication
 - [Suoqin Jin et al., CellChat for systematic analysis of cell-cell communication from single-cell and spatially resolved transcriptomics, bioRxiv 2023 \[CellChat v2\]](#)
 - [Suoqin Jin et al., Inference and analysis of cell-cell communication using CellChat, Nature Communications 2021 \[CellChat v1\] Citation:1717](#)

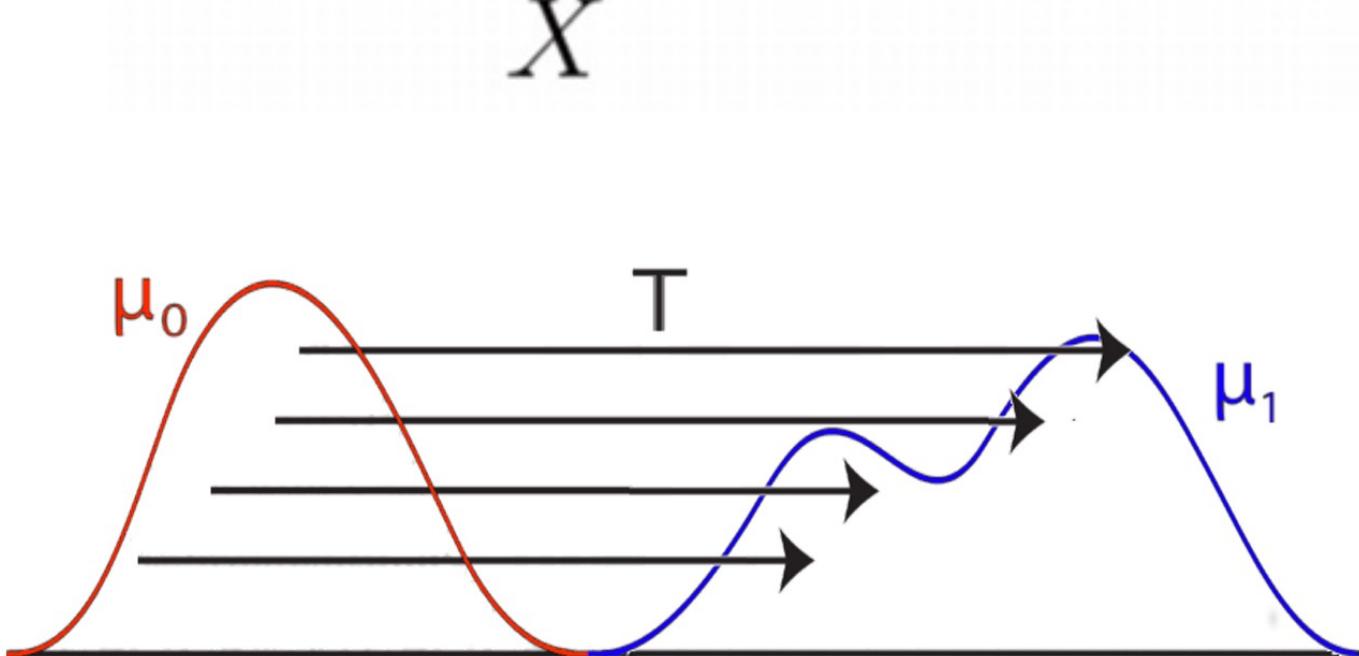
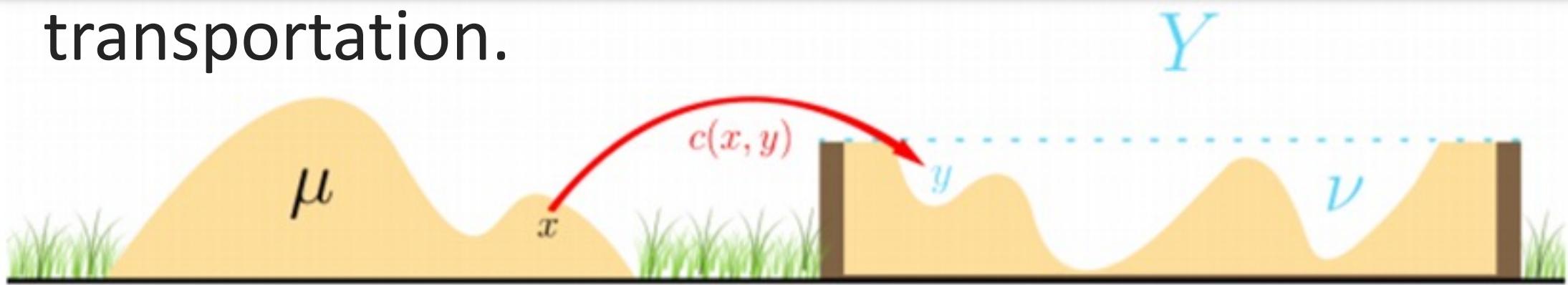


- Overview of CellChat along with the Procedure step numbers.
- Left: Required input data and the ligand-receptor interaction database CellChatDB. CellChat's input data consist of gene expression data and cell group information. When inferring spatially-proximal cell-cell communication, spatial locations of cells are also required. CellChatDB takes into account known composition of the ligand-receptor complexes, including complexes with multimeric ligands and receptors, as well as several cofactor types: soluble agonists, antagonists, co-stimulatory and coinhibitory membrane-bound receptors.
- Middle: CellChat models the communication probability based on the law of mass action and identifies significant communications using permutation tests. The inferred communication probabilities among all pairs of cell groups across all pairs of ligand receptor are represented by a three-dimensional array. CellChat analyzes the inferred networks by leveraging social network metrics, pattern recognition methods and manifold learning approaches.
- Right: CellChat offers several intuitive visualization outputs to facilitate data interpretation of different analytical tasks. In addition to analyzing individual dataset, CellChat also delineates signaling changes across different conditions. The Procedure step numbers in different parts are also indicated to link the Procedure sections with the overall scheme.

CCC on spatial transcriptomics

- Giotto builds a spatial proximity graph to identify interactions through membrane-bound ligand–receptor pairs²³;
- CellPhoneDB v3 restricts interactions to cell clusters in the same microenvironment defined based on spatial information²⁵;
- stLearn relates the co-expression of ligand and receptor genes to the spatial diversity of cell types²⁴;
- SVCA²⁶ and MISTy²⁷ use probabilistic and machine learning models, respectively, to identify the spatially constrained intercellular gene–gene interactions;
- NCEM fits a function to relate cell type and spatial context to gene expression²⁸.

Optimal Transport: is an optimization problem which goal is to minimize the cost of transportation.



- How to move pile X to the shape of pile Y with minimal effort? i.e. given a cost function $c(x,y)$ of moving a grain $x \in X$ to the position $y \in Y$, what is the optimal displacement of all X to Y ?

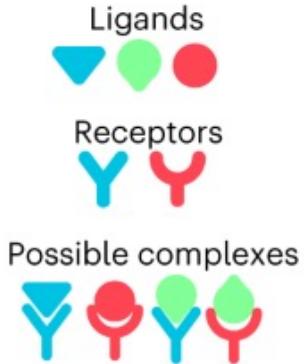
French Mathematician: Gaspard Monge

Elegant solution: OT

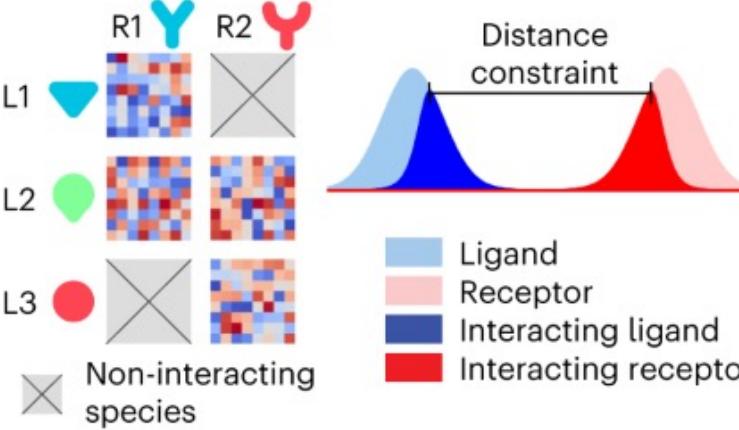
- Current methods examine CCC locally and on cell pairs independently, and focus on information between cells or in the neighborhoods of individual cells. As a result, collective or global information in CCC, such as competition between cells, is neglected.
- Naturally, one can form an optimal transport problem by viewing ligand and receptor expression as two distributions to be coupled with a cost based on spatial distance
- **Collective optimal transport**, which is capable of preserving the comparability between distributions, ensuring that the total signal does not exceed the individual species amounts (ligand or receptor), enforcing spatial range limits of signaling, and handling multiple competing species.

a

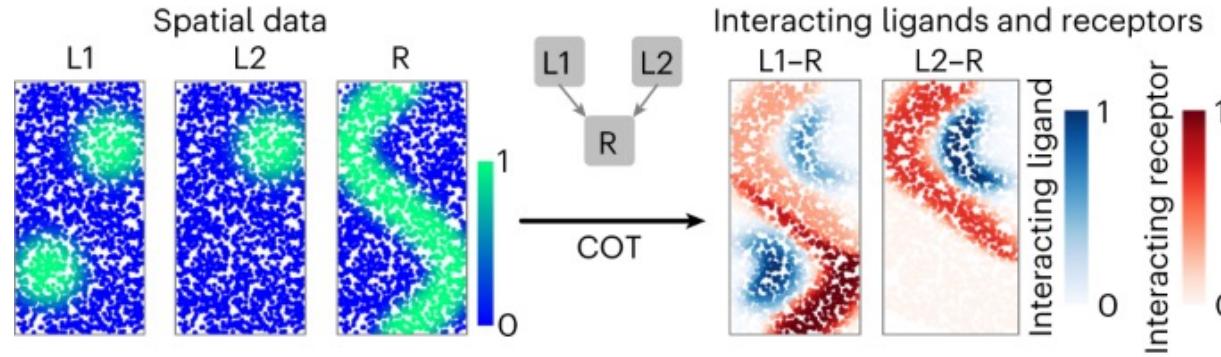
Multi-species ligand-receptor interactions

**b**

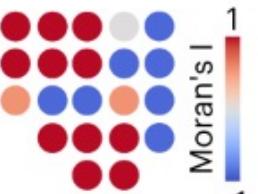
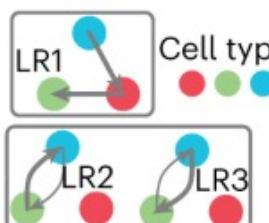
COT for CCC inference with multiple species and spatial constraints

**c**

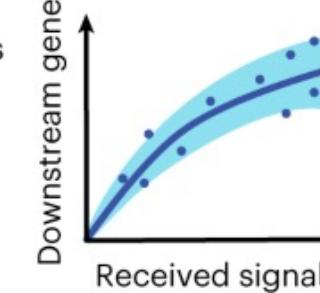
Infer interacting ligands and receptors

**d**

Identify differential CCC dir.

Downstream analysis
Group CCC networks

Identify DEG w.r.t. CCC

**COMMOT**

(COMMunication analysis by Optimal Transport)

- Ligands and receptors often interact with multiple species and within limited spatial ranges (Fig. 1a).
- Considering this, we present **collective optimal transport** (Fig. 1b) with three important features:
 - first, the use of non-probability mass distributions to control the marginals of the transport plan to maintain comparability between species;
 - second, enforcement of spatial distance constraints on CCC to avoid connecting cells that are spatially far apart;
 - last, the transport of multi-species distributions (ligands) to multi-species distributions (receptors) to account for multi-species interactions (Fig. 1c).

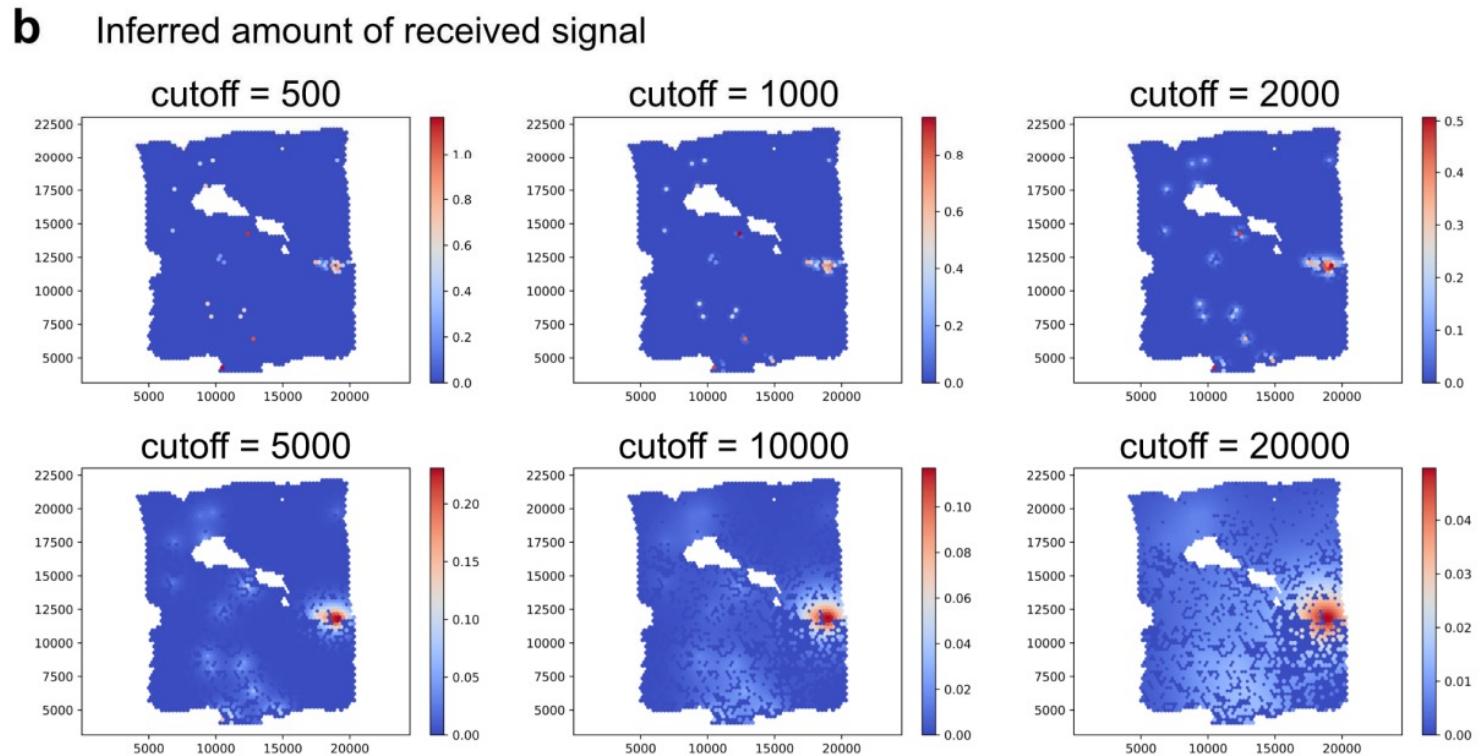
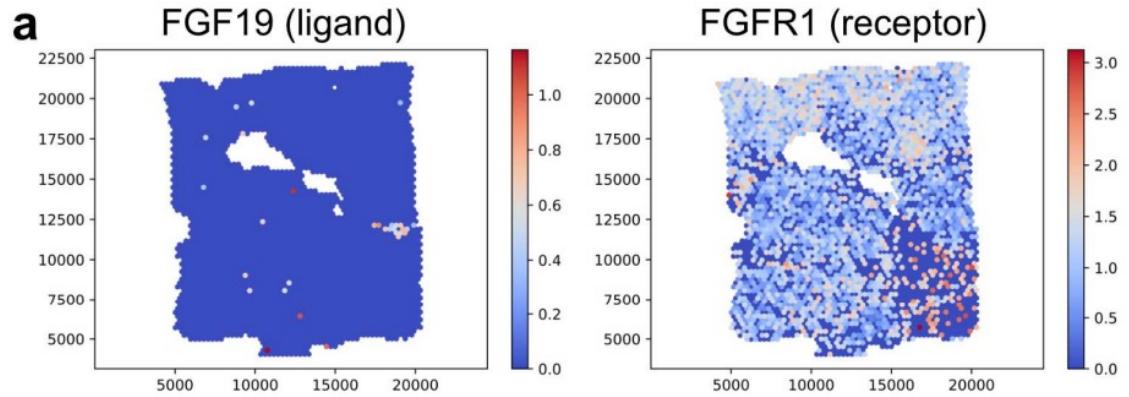
Given a spatial transcriptomics dataset of n_s cells or spots and n_l ligand species and n_r receptor species, the collective optimal transport determines an optimal multi-species coupling $\mathbf{P}^* \in \mathbb{R}_+^{n_l \times n_r \times n_s \times n_s}$ where $\mathbf{P}_{i,j,k,l}^*$ scores the signaling strength from sender cell k to receiver cell l through ligand i and receptor j . This is achieved by solving a minimization problem, $\min_{\mathbf{P} \in \Gamma} \sum_{(i,j) \in I} \alpha_{(i,j)} \langle \mathbf{P}_{i,j,\cdot,\cdot}, \mathbf{C}_{(i,j)} \rangle_F$ where

- Formulation

$$\Gamma = \left\{ \mathbf{P} \in \mathbb{R}_+^{n_l \times n_r \times n_s \times n_s} : \mathbf{P}_{i,j,\cdot,\cdot} = 0 \text{ for } (i,j) \notin I, \right.$$

$$\left. \sum_{j,l} \mathbf{P}_{i,j,k,l} \leq \mathbf{X}_{i,k}, \sum_{i,k} \mathbf{P}_{i,j,k,l} \leq \mathbf{X}_{j,l} \right\},$$

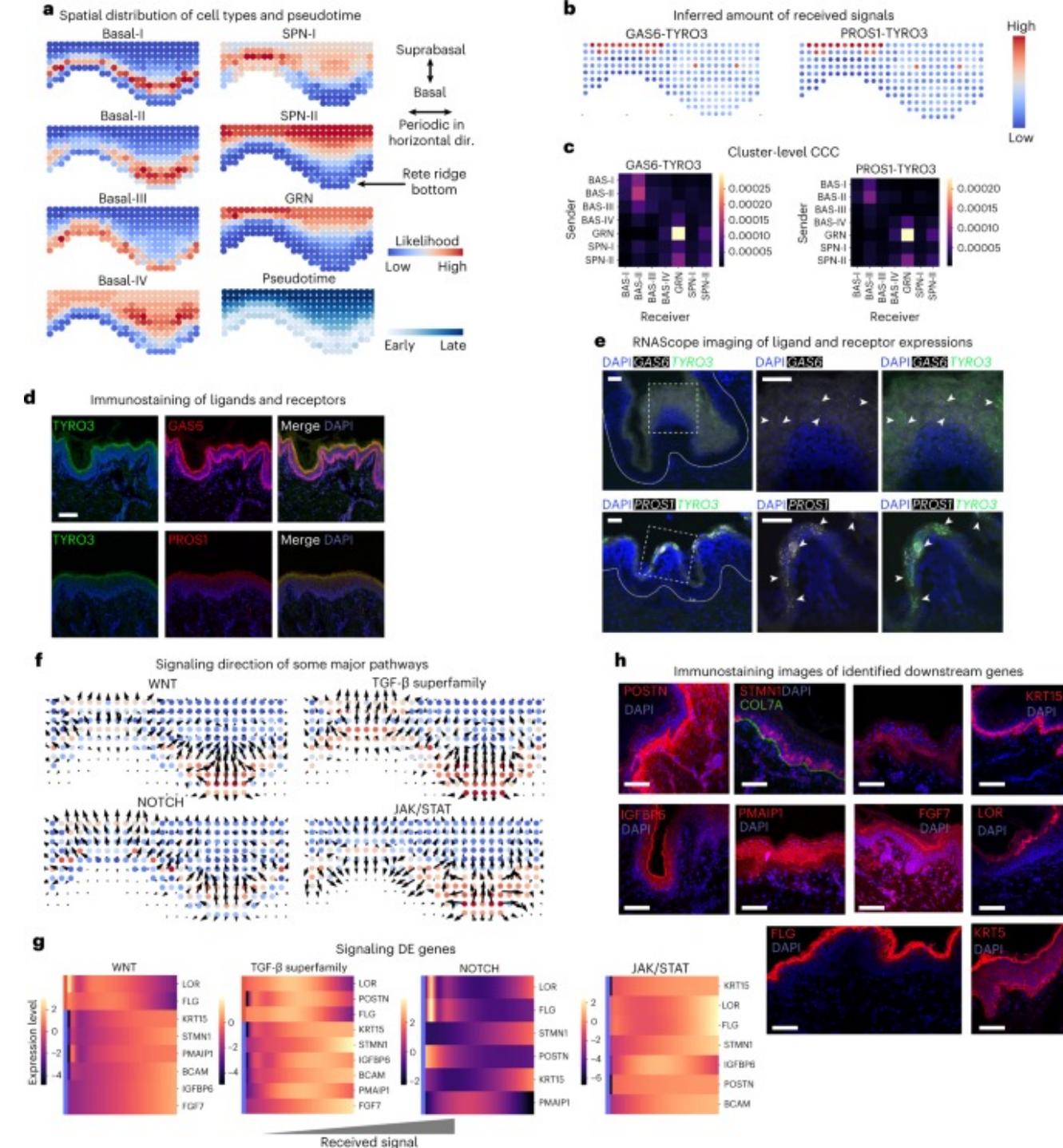
I is the index set for ligand and receptor species that can bind together, and $\mathbf{X}_{i,k}$ is the expression level of gene i on spot k . The species-specific cost matrix $\mathbf{C}_{(i,j)}$ is a modified distance matrix for between-spot distance that replaces distances exceeding the spatial range of ligand i by infinity. The competitions between molecule species and cells are considered by assuming that a given receptor species or cell has limited capacity for interactions, such that a stronger inferred interaction with one ligand species or cell reduces the potential of interaction with other ligand species or cells (see the Methods and Supplementary Note for detailed formulations and algorithm derivations).



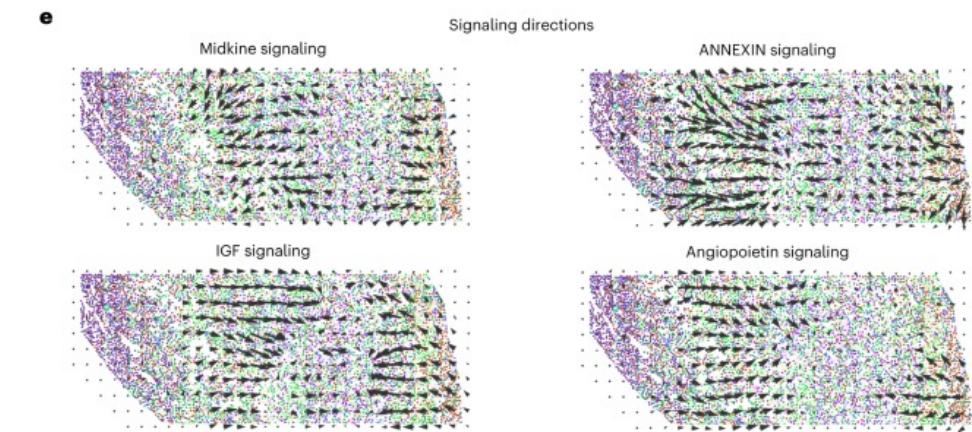
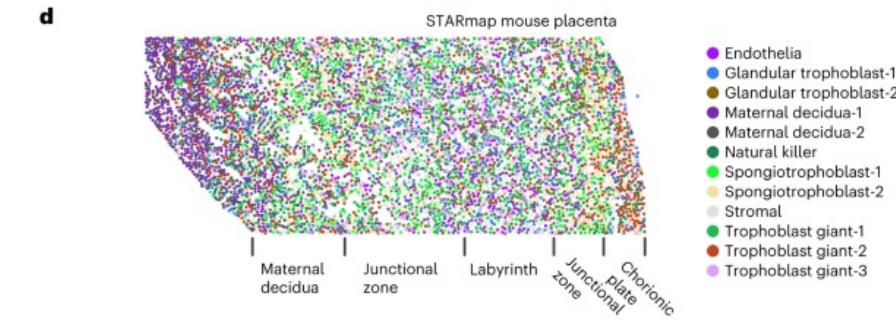
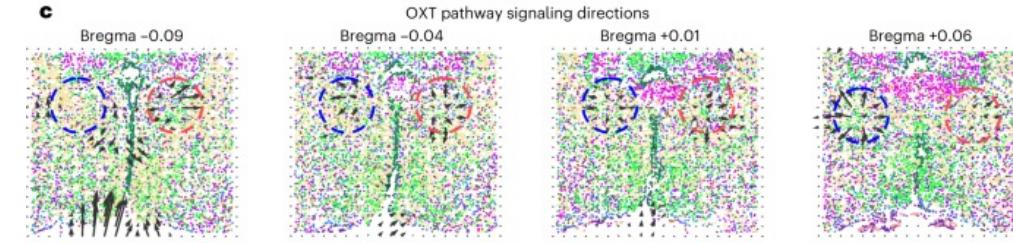
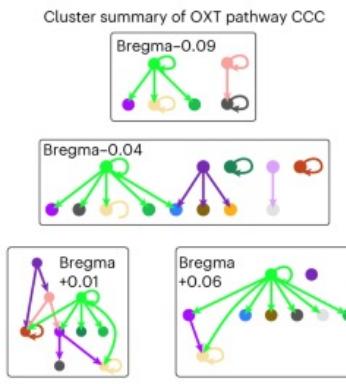
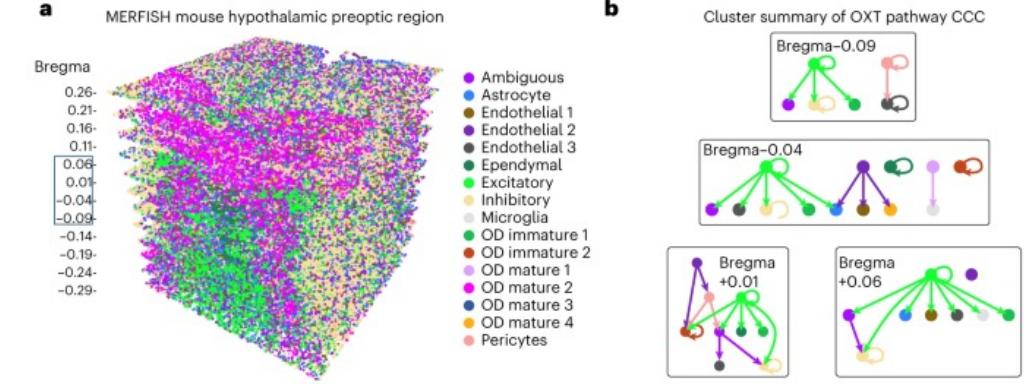
The spatial distance constraint used to capture the effect of ligand diffusivity is usually determined by several factors, including protein weight and tortuosity of extracellular space⁶².

It is difficult to accurately estimate this parameter for every pair in the database.

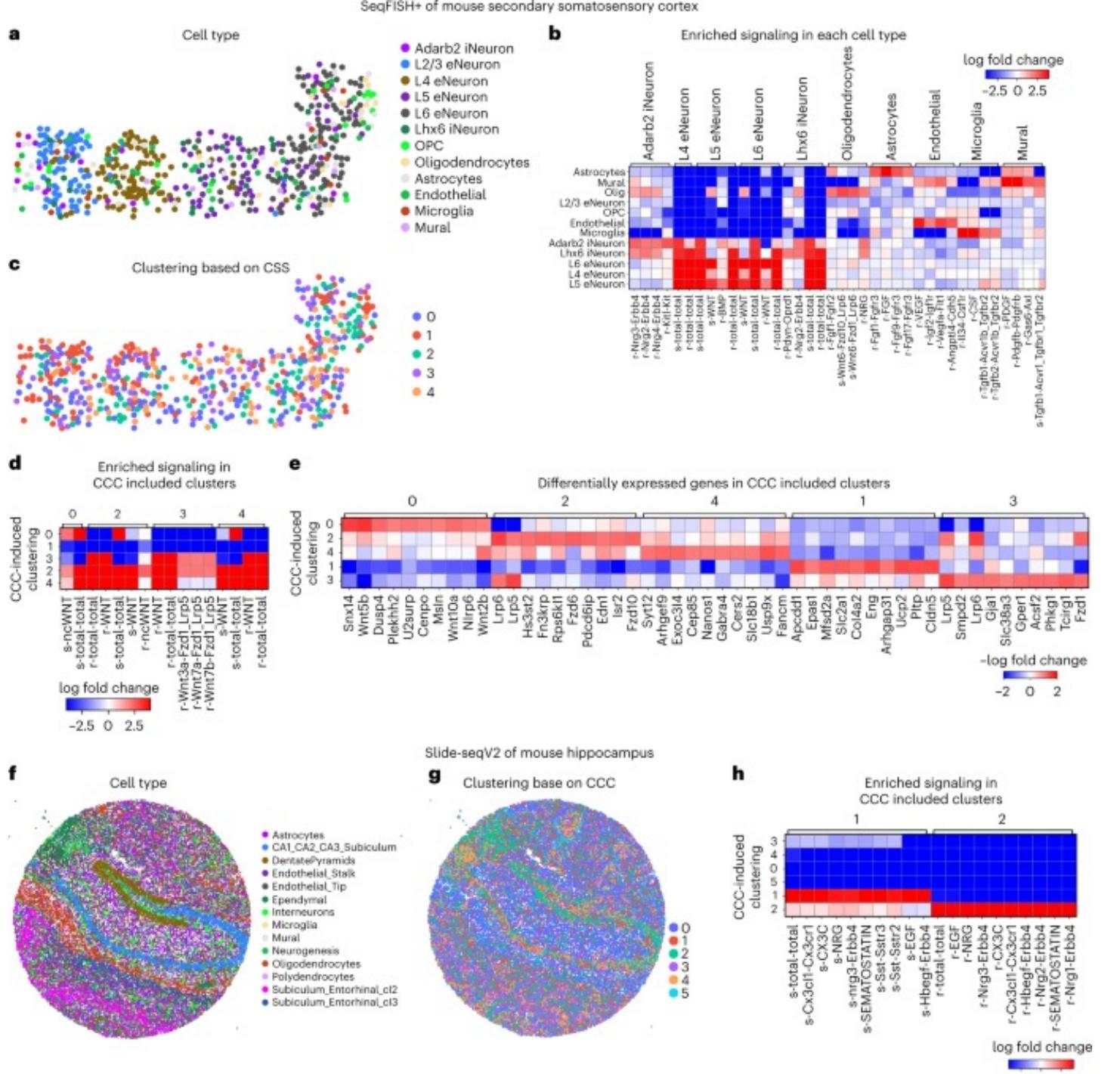
In our model the local short-range interactions are emphasized even when the spatial distance range is increased (Supplementary Fig. 36). Thus, when screening many ligand–receptor pairs a uniform and relatively large spatial distance limit may be used to avoid missing important interactions. Once the important interactions are identified, an accurate estimation of this parameter would further refine the prediction to remove false-positive CCC links.



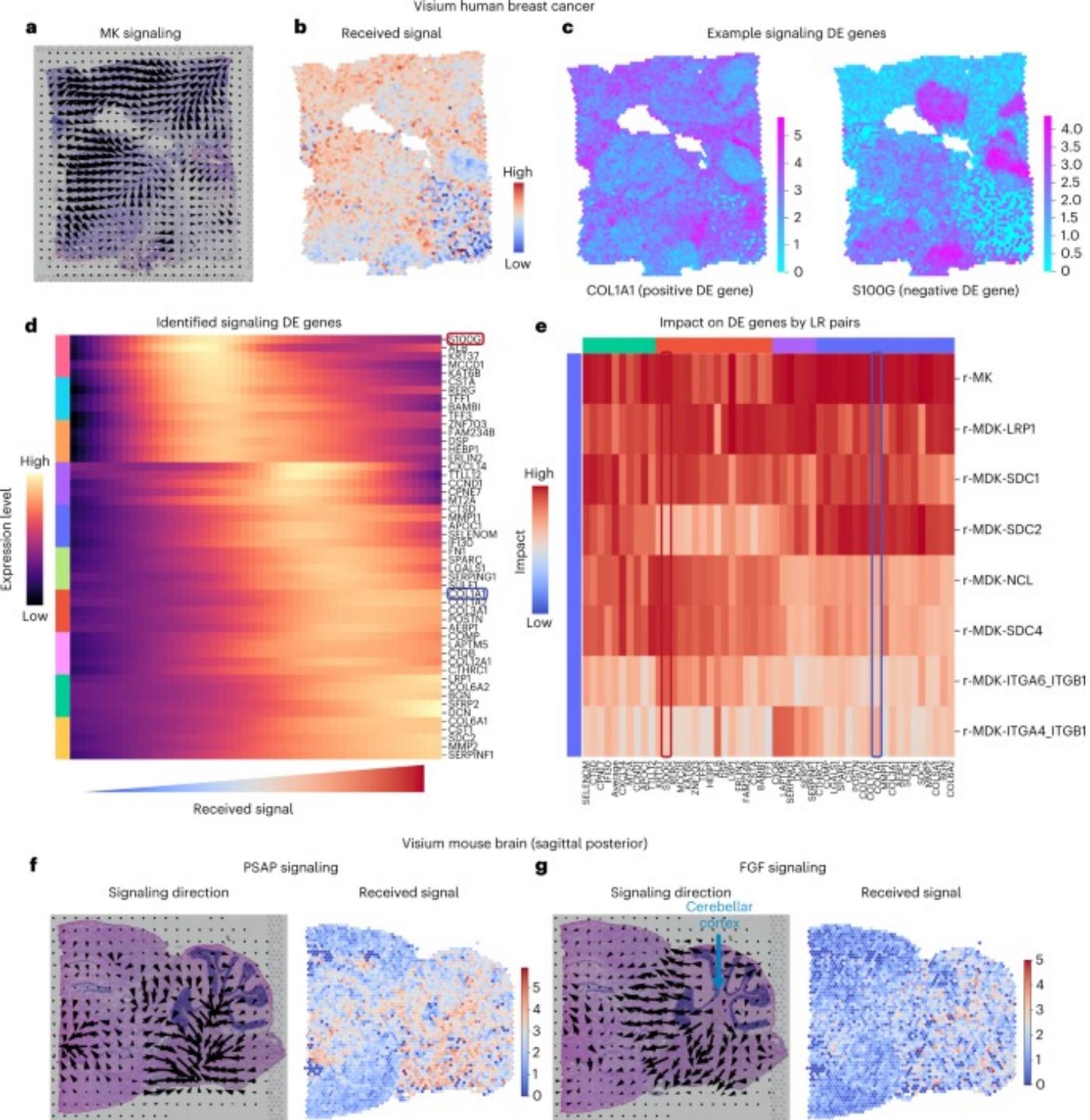
- **a**, Predicted spatial origin of the skin subtypes of cells in intact tissue and the pseudotime projected to space. GRN, granular cell cluster; SPN, spinous cell cluster. **b,c**, The inferred amount of received signals of two example ligand–receptor pairs, GAS6-TYRO3 and PROS1-TYRO3 at the cell level (**b**) and cluster level (**c**). **d**, Immunostaining of proteins for GAS6, TYRO3 and PROS1. **e**, Fluorescent *in situ* hybridization against RNA molecules for predicted ligand–receptor interactions in human epidermis (solid white outline; regions of interest are marked by a white dashed square). The top row shows expression patterns of GAS6 (white) and TYRO3 (green); the bottom row shows expression patterns for PROS1 (white) and TYRO3 (green). In both cases, the middle and right panels show ligand–receptor signals, some of which colocalize to the stratum granulosum (white arrowheads). In merged images, the brightness of the GAS6 channel was increased to improve clarity against the prominent TYRO3 (green) signal. Experiments were repeated four times independently with consistent results. **f**, The signaling directions of four major signaling pathways. **g**, Heatmaps of selected signaling differentially expressed genes of the four signaling pathways, respectively. **h**, Immunofluorescence staining images of the identified signaling differentially expressed genes supporting the identified correlation between WNT signaling and the expression of these genes. Scale bars: **d,e,h**, 100 μ m. The immunostaining experiments in **d** and **h** were repeated three times independently with consistent



- **a**, MERFISH data of the mouse hypothalamic preoptic region with multiple slices across the anterior-posterior axis⁴⁴. **b**, Cluster-level summary of CCC through the OXT signaling pathway. **c**, Signaling directions of the OXT pathway. **d**, STARmap data of the mouse placenta⁴⁶. **e**, Signaling directions of the midkine, IGF, annexin and angiopoietin pathways.

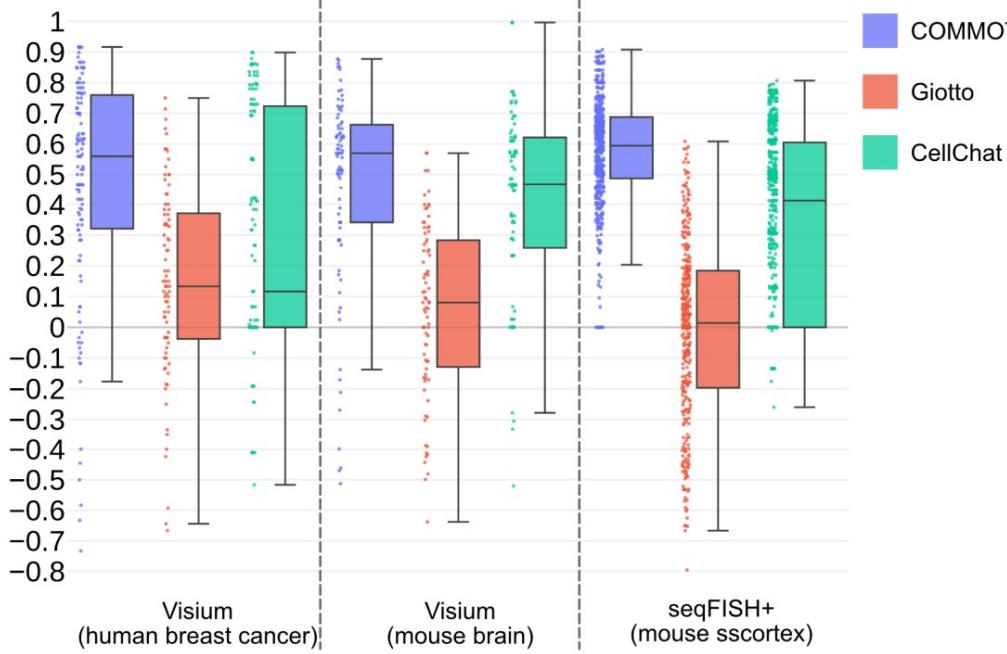


- **a–e**, CCC analysis of seqFISH+ data of mouse secondary somatosensory cortex. **a**, Clustering of cell type based on gene expression. OPC, oligodendrocyte precursor cells. **b**, Enriched signaling in each cell type. **c**, **Clustering based on inferred CCC**. **d**, Enriched signaling in CCC-induced clusters. **e**, Differentially expressed genes in the CCC-induced clusters. **f–h**, CCC analysis of Slide-seq (v2) data of mouse hippocampus. **f**, Clustering of cell type based on gene expression. **g**, Clustering based on inferred CCC. **h**, Enriched signaling in CCC-induced clusters.



- **a–e**, Midkine (MK) signaling in human breast cancer tissue. **a**, Spatial signaling direction. **b**, Amount of received signal by each spot. **c**, Two examples of differentially expressed (DE) genes due to signaling. **d**, Identification of the differentially expressed genes due to the total amount of received signal in the MK signaling pathway. **e**, Unique impact on the identified differentially expressed genes by the individual ligand–receptor pairs. **f,g**, Signaling in mouse brain tissue. The signaling direction (left) and the level of received signal (right) are shown for PSAP signaling (**f**) and FGF signaling (**g**).

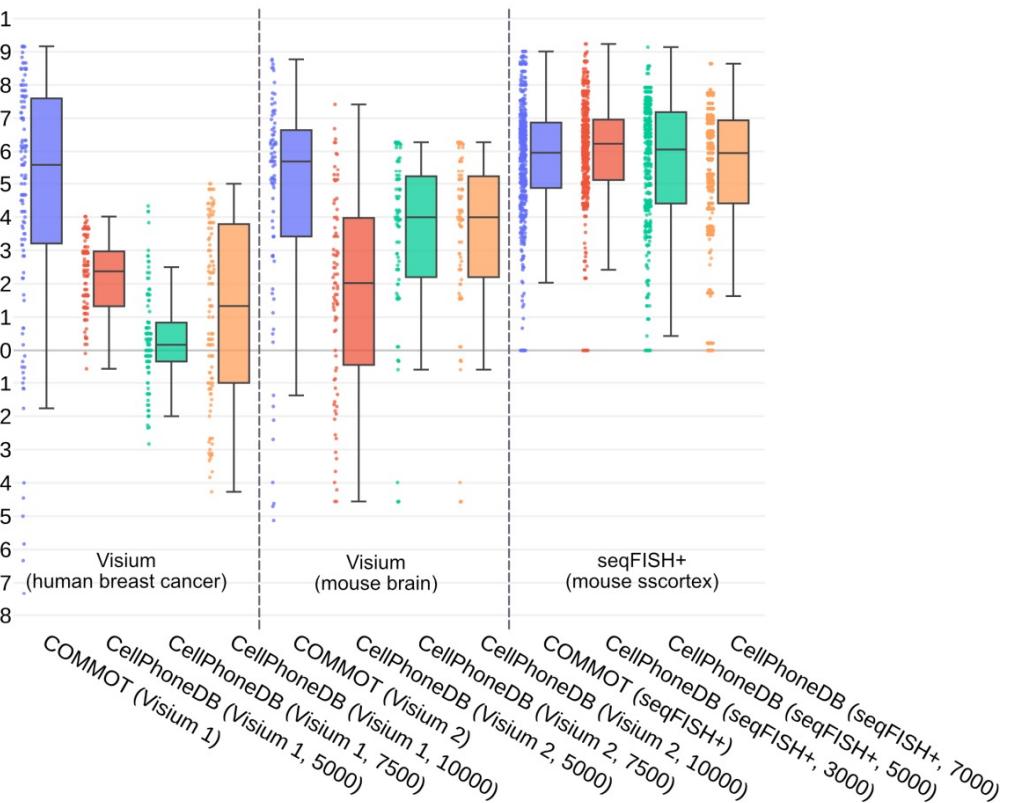
Cluster-level Spearman's r

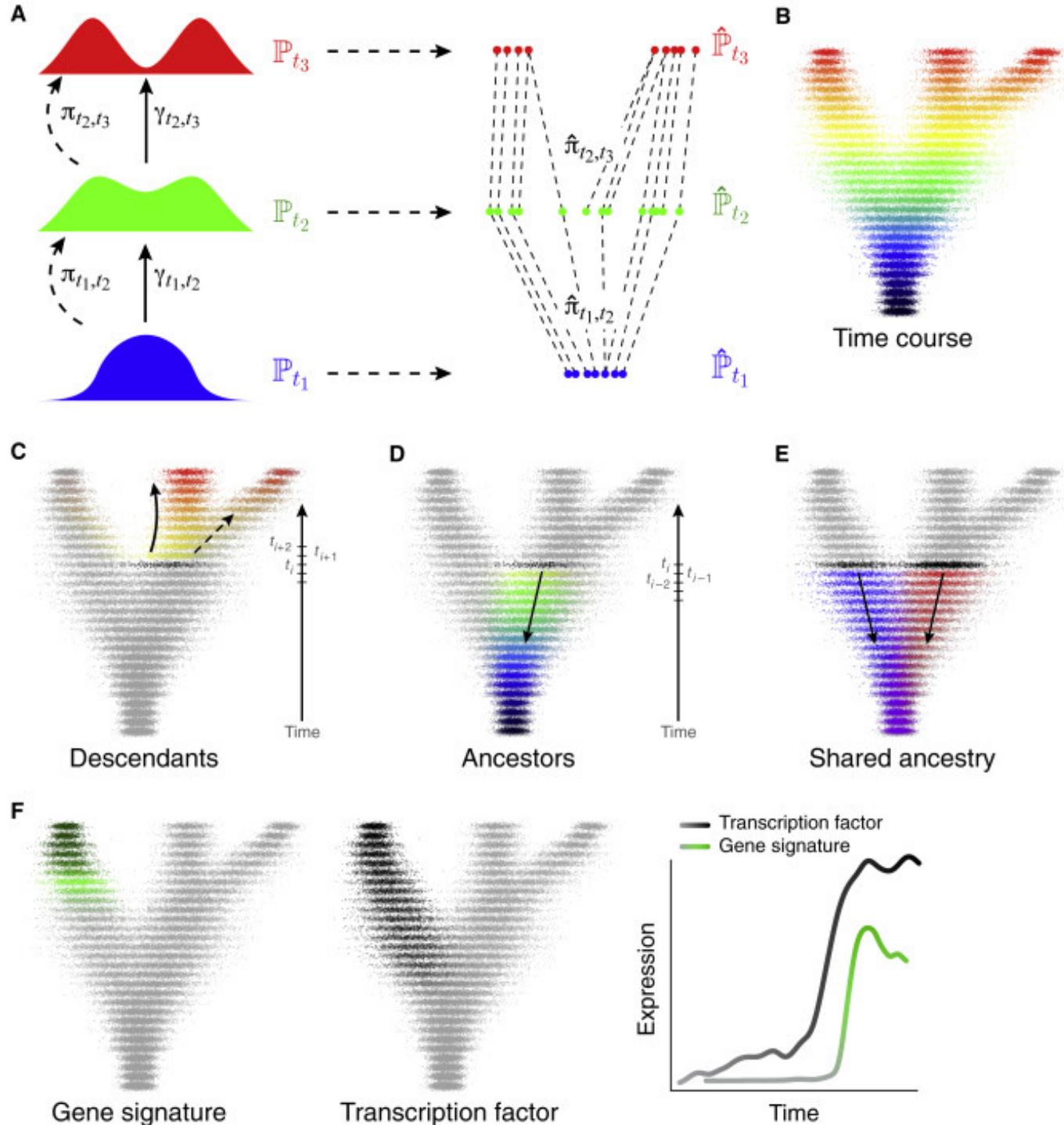


Cluster-level correlation between inferred signaling and activity of known downstream genes (comparison with CellPhoneDB v3)

- Cluster-level correlation between inferred signaling and activity of known downstream genes (comparison with CellChat and Giotto)

Cluster-level Spearman's r

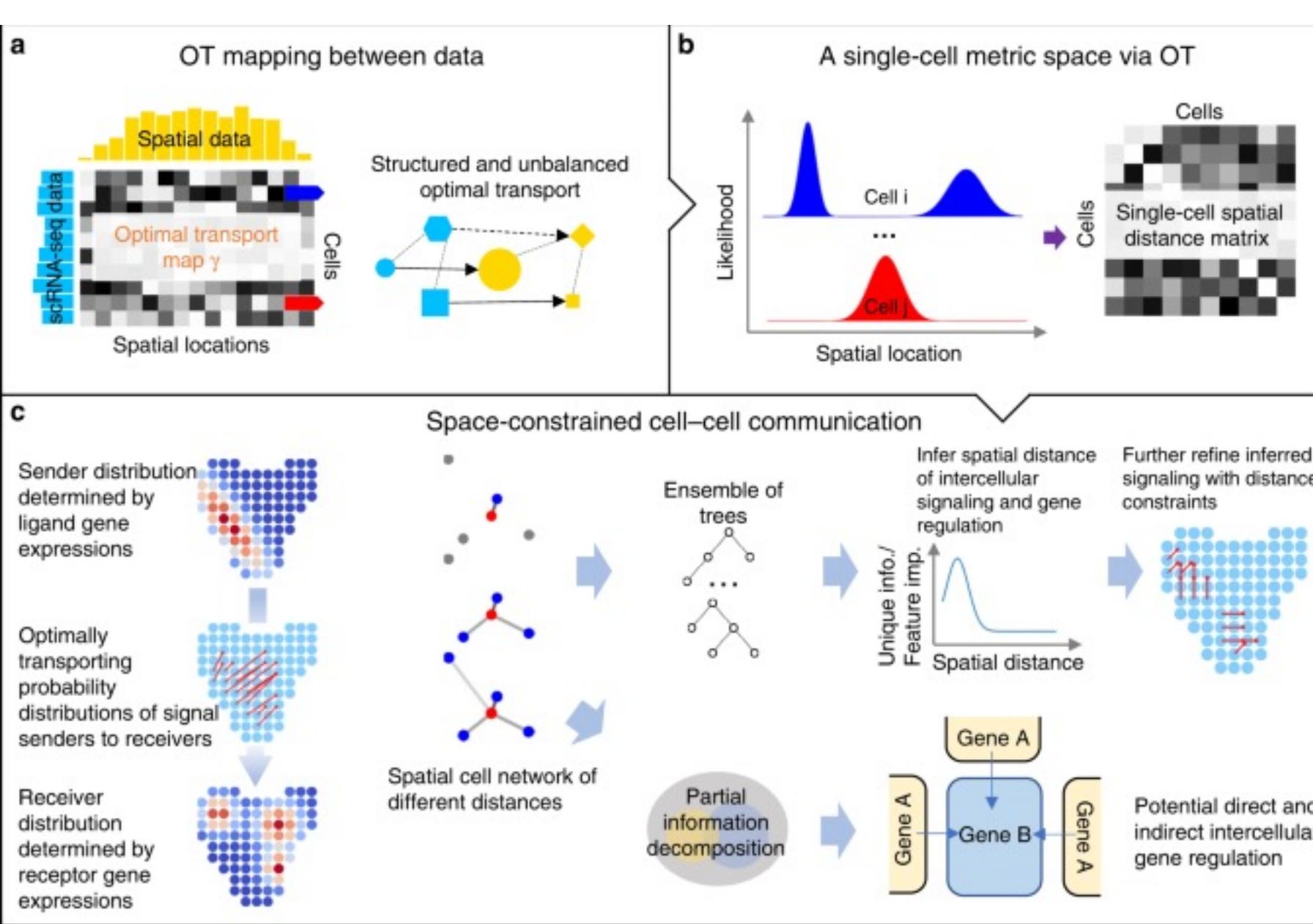




Other OT applications

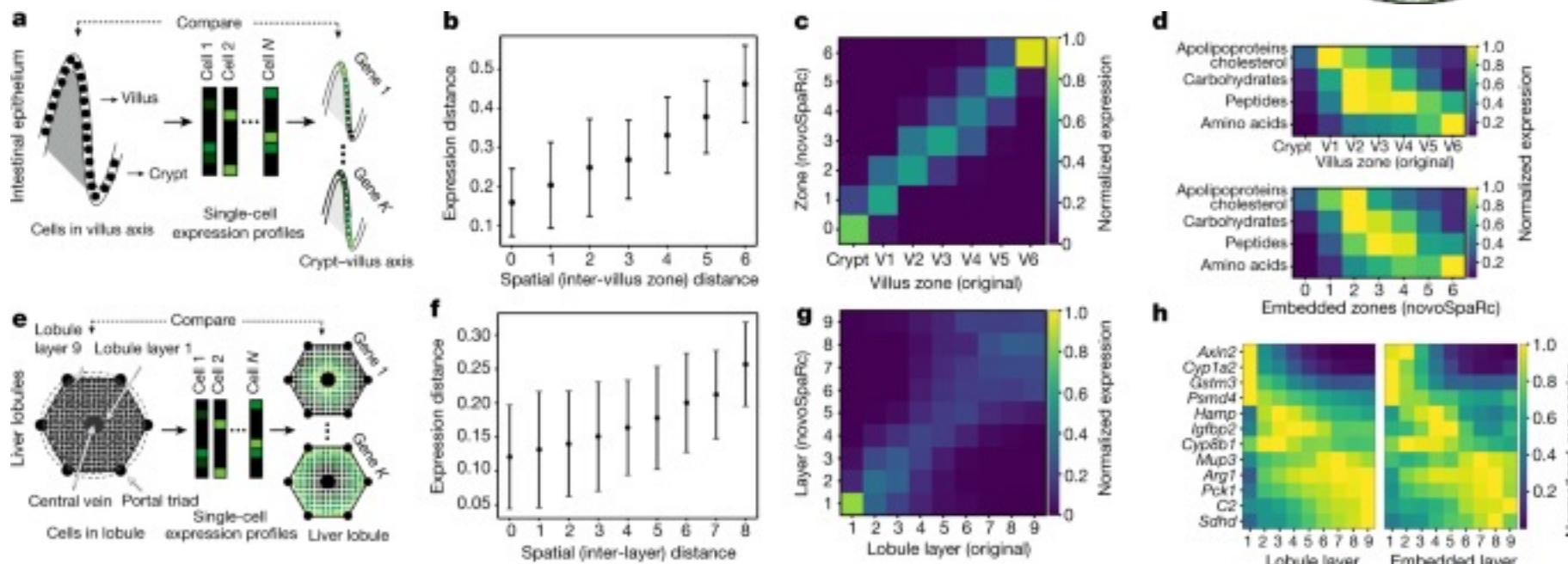
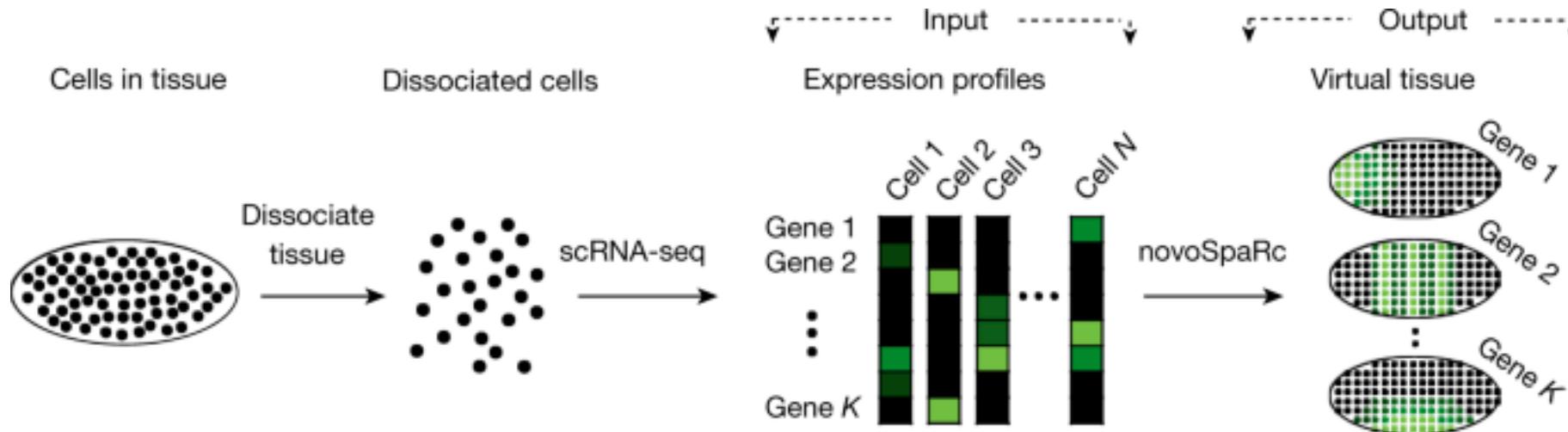
- OT for developmental trajectory reconstruction

Schiebinger, G. et al. Optimal-transport analysis of single-cell gene expression identifies developmental trajectories in reprogramming. *Cell* **176**, 928–943 (2019)



OT for spatial annotation of scRNA-seq data

Cang, Z. & Nie, Q. Inferring spatial and signaling relationships between cells from single cell transcriptomic data. *Nat. Commun.* **11**, 2084 (2020).



OT for spatial annotation of scRNA-seq data

Nitzan, M., Karaïkos, N., Friedman, N. & Rajewsky, N. Gene expression cartography. *Nature* **576**, 132–137 (2019)

Other methods to study CCC with spatial transcriptomics data

- SpatialDM evaluates the co-expression of ligand and receptor genes;
- SpaTalk and stMLnet are focused on signaling target genes;
- HoloNet studies the joint impact from different combinations of CCC events;
- DeepLinc constructs de novo cell–cell interaction landscapes without the need for annotated ligand and receptor genes.

- 1.Li, Z., Wang, T., Liu, P. & Huang, Y. SpatialDM: Rapid identification of spatially co-expressed ligand-receptor reveals cell-cell communication patterns. Preprint at <https://doi.org/10.1101/2022.08.19.504616> (2022).
- 2.Shao, X. et al. Knowledge-graph-based cell-cell communication inference for spatially resolved transcriptomic data with SpaTalk. *Nat. Commun.* **13**, 4429 (2022).
- 3.Cheng, J., Yan, L., Nie, Q. & Sun, X. Modeling spatial intercellular communication and multilayer signaling regulations using stMLnet. Preprint at <https://doi.org/10.1101/2022.06.27.497696> (2022).
- 4.Li, H., Ma, T., Hao, M., Wei, L. & Zhang, X. Decoding functional cell–cell communication events by multi-view graph learning on spatial transcriptomics. Preprint at <https://doi.org/10.1101/2022.06.22.496105> (2022).
- 5.Li, R. & Yang, X. De novo reconstruction of cell interaction landscapes from single-cell spatial transcriptome data with DeepLinc. *Genome Biol.* **23**, 124 (2022).

Limitations

- It is possible that there are **missing ligand-receptor interactions** not covered in CellChatDB. A tutorial is provided on how to update CellChatDB by adding user-defined ligandreceptor pairs or other resources. There are several other limitations for the original CellChat and its v2.
- First, CellChat infers **potential interactions between cell groups without considering heterogeneity within the defined cell groups**. To address this limitation, users can refine cell grouping (e.g., spatially heterogenous cells) or define mixed cell types as “super cell types” before applying CellChat v2.
- Second, similar to other methods, CellChat is **limited to hypothesis generation** and employs heuristics to guide interpretation of cell-cell communication outputs. With limited benchmarking studies, the question of how to better validate the inferred signaling networks and their downstream gene outputs remains to be answered.
- Third, cross-condition analysis in CellChat is largely restricted to **pairwise comparisons**. Identification of signaling changes across multiple conditions and time-series is valuable.
- Fourth, for molecules that are not directly related to genes **measured in scRNA-seq**, CellChat v2 estimates the expression of ligands and receptors using those molecules’ key mediators or enzymes for potential communication mediated by non-proteins.
- Fifth, given that cell-cell communication **occurs at the protein level**, newly emerging data modalities from singlecell multi-omics, such as single-cell proteomics and epigenomics, can be used to improve the inference of cell-cell communication.
- Sixth, CellChat employs a **simplified mass action-based model** to quantify communication probability between a given ligand and its cognate receptor, and models with more biochemical details can potentially improve inference predictions.
- Finally, **incorporation of the downstream signaling** events of activated receptors on receiving cells could further improve the overall inference accuracy.

Summary

- The complex structures and functions of multicellularity are achieved through the coordinated activities of various cells.
- Cells make decisions and accomplish their goals by interacting with an environment consisting of external stimuli and other cells.
- There are huge needs and spaces in CCC

Expert opinion

“With COMMOT, Cang et al. present an elegant mathematical solution to the problem of inferring cell–cell communication from spatial transcriptomics data based on a variant of optimal transport. The method is applied to spatial datasets of different sizes and technologies, and robustness of results is shown. Further, the authors show how their method can be used in different biological contexts, including human breast cancer and mouse brain samples.” **Fabian Theis and Marius Lange, Helmholtz Munich, Germany**

CellChatDB

<http://www.cellchat.org/cellchatdb/>



Species: mouse

Enter a pathway: ACTIVIN

Show 100 entries

Search:

	interaction_name	pathway_name	ligand	receptor	agonist	antagonist	co_A_receptor	co_L_receptor	evidence	annotation	interaction_name_2
1	INHBA_ACVR1B_ACVR2A	ACTIVIN	Inhba	ACVR1B_ACVR2A	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04350	Secreted Signaling	Inhba - (Acvr1b+Acvr2a)
2	INHBA_ACVR1B_ACVR2B	ACTIVIN	Inhba	ACVR1B_ACVR2B	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04351	Secreted Signaling	Inhba - (Acvr1b+Acvr2b)
3	INHBB_ACVR1B_ACVR2A	ACTIVIN	Inhbb	ACVR1B_ACVR2A	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04352	Secreted Signaling	Inhbb - (Acvr1b+Acvr2a)
4	INHBB_ACVR1B_ACVR2B	ACTIVIN	Inhbb	ACVR1B_ACVR2B	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04353	Secreted Signaling	Inhbb - (Acvr1b+Acvr2b)
5	INHBB_ACVR1C_ACVR2A	ACTIVIN	Inhbb	ACVR1C_ACVR2A	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04354	Secreted Signaling	Inhbb - (Acvr1c+Acvr2a)
6	INHBB_ACVR1C_ACVR2B	ACTIVIN	Inhbb	ACVR1C_ACVR2B	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04355	Secreted Signaling	Inhbb - (Acvr1c+Acvr2b)
7	INHBC_ACVR1B_ACVR2A	ACTIVIN	Inhbc	ACVR1B_ACVR2A	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04356	Secreted Signaling	Inhbc - (Acvr1b+Acvr2a)
8	INHBC_ACVR1B_ACVR2B	ACTIVIN	Inhbc	ACVR1B_ACVR2B	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04357	Secreted Signaling	Inhbc - (Acvr1b+Acvr2b)
9	INHBC_ACVR1C_ACVR2A	ACTIVIN	Inhbc	ACVR1C_ACVR2A	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04358	Secreted Signaling	Inhbc - (Acvr1c+Acvr2a)
10	INHBC_ACVR1C_ACVR2B	ACTIVIN	Inhbc	ACVR1C_ACVR2B	ACTIVIN antagonist		ACTIVIN inhibition receptor		KEGG: mmu04359	Secreted Signaling	Inhbc - (Acvr1c+Acvr2b)

<http://www.cellchat.org/wound/>

