

FROM SPATIAL MAPS TO MOLECULAR NETWORKS: CONNECTOGENOMICS AS THE NEXT GENERATION OF SPATIAL BIOLOGY

Raghularajan Govindarajan¹, Kaliyammal Krishnan¹, Dr. Senthil Natesan¹, Dr. V. Rajan Babu^{1*}.

¹Department of Plant Biotechnology, Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore – 641003.

*Corresponding authors. Dr. V. Rajan Babu, Email: rajanbabu.v@tnau.ac.in

ABSTRACT

Spatial omics has transformed biological research by enabling molecular profiling while preserving the spatial organization of tissues. Unlike conventional omics approaches that often lose spatial context during sample processing, spatial omics integrates molecular information with tissue architecture to investigate cellular heterogeneity, cell–cell communication, and tissue organization. Connectogenomics is an emerging field that combines spatial omics, connectomics, and network biology to characterize molecular and cellular interaction networks within biological systems. By representing biological entities as interconnected nodes and edges, connectogenomics provides a systems-level framework for understanding tissue organization, development, and disease progression. This review examines fundamental concepts of spatial omics, including cellular heterogeneity and cell–cell communication, and discusses limitations of traditional omics that have contributed to the development of spatially resolved approaches. Major sequencing-based technologies, including volumetric DNA microscopy, single-cell Hi-C, and proximity network assays, are discussed alongside computational approaches for dimensionality reduction and network analysis. Applications in cancer research, neuroscience, developmental biology, plant biotechnology, and precision medicine are highlighted. Current challenges include high-dimensional data analysis, computational complexity, limited spatial resolution, multi-omics integration, and experimental validation. Finally, emerging opportunities involving artificial intelligence and high-resolution spatial technologies are considered for advancing systems-level understanding of complex biological networks.

KEYWORDS: Connectogenomics; Spatial omics; Spatial transcriptomics; Molecular interaction networks; Cell–cell communication; Network biology; Multi-omics; Systems biology.

1. INTRODUCTION

In biological systems, cells have complex molecular networks, and Cells communicate with one another either through direct, short-range contact-dependent interactions or by releasing signalling molecules that convey information to neighbouring cells. The physical connection and the surface-level communication between cells by direct or indirect means are called “cell-cell interactions.” These interactions regulate essential biological processes such as development, regeneration, immune responses, and disease progression (1). The intracellular information of individual cells is influenced by their surrounding multicellular environment (2).

Therefore, understanding how cells interact within tissues requires both molecular and spatial information. The interaction between cells depends on several factors, and most of the existing methods that are considered only gene expression information of neighbouring cells and spatial location information are somewhat limited (3). The traditional omics technologies include genomics, transcriptomics, proteomics, and metabolomics have greatly expanded our knowledge of biological systems at the molecular level, which analyse large numbers of molecules simultaneously (4). Although, they often lack spatial information because tissues are homogenized before molecular analysis.

Spatial omics technologies were developed to underlying address these challenges by integrating molecular measurements with spatial information (5). These technologies enable researchers to get a better knowledge of the structure and function of biological systems, revealing mechanisms that underpin disease development, medication effects, and complex regulatory networks within organisms.

2. Spatial omics technologies

Spatial omics integrates molecular analysis with spatial information, improving our understanding of cellular function and organization within their native environments. It also enables us to achieve accurate molecular localization within biological tissues. It has a various technologies throughput method such as spatial transcriptomics, translomics, genomics, and metabolomics to understand the spatial distribution, interaction, and regulation of various biomolecules within biological systems (6).

Understanding the fundamental ideas of spatial omics is critical for conducting accurate and successful molecular localization studies. Spatial transcriptomics investigates gene expression patterns and regulatory networks to understand the spatial arrangement of mRNA within cells or tissues. (7). Spatial proteomics and translomics

analyse protein expression levels, relationships, and the translation of mRNA into proteins, providing insights into the proteins' spatial distribution and regulatory mechanisms. (8).

Spatial genomics and epigenomics investigate DNA's three-dimensional (3D) structure, chromatin organization of the genome and its regulatory landscape (9). Metabolomics investigates metabolite distribution and variation within cells or tissues, completing the spatial omics landscape by understanding the spatial distribution and dynamics of cellular metabolites. (10). Spatial omics system also reveals cellular interaction and organization by maintaining the spatial context of tissues, which unravels complex biological processes. By integrating these diverse information, spatial omics technologies offer novel prospective and methodologies for understanding cellular functions and identify cell types within their native microenvironment, and investigate tissue heterogeneity. A general overview of this spatial omics workflow, illustrating the transition from tissue preservation to computational analysis, is provided in **Figure 1**.

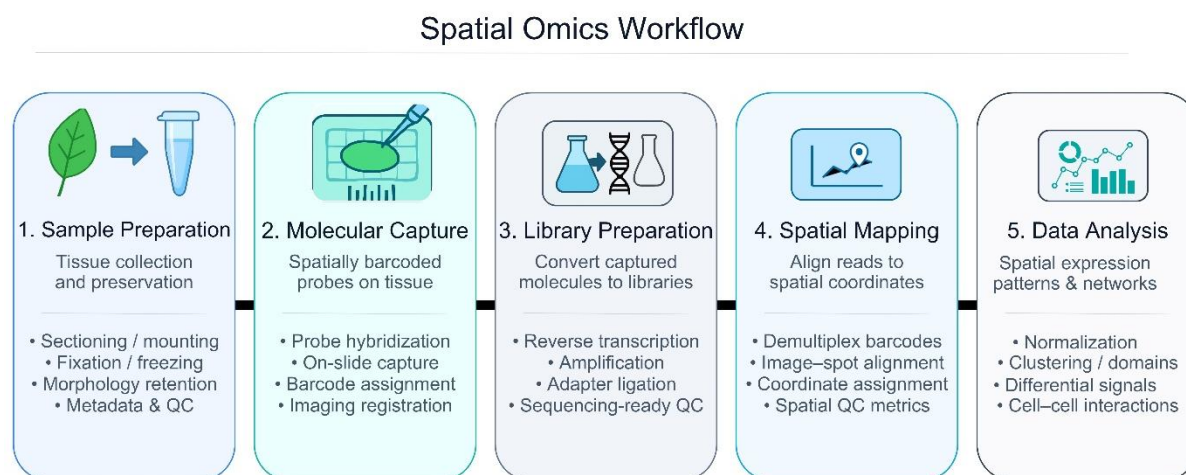


Figure 1. Overview of the spatial omics workflow. Tissue samples are first collected and preserved to maintain structural and molecular integrity. Biomolecules (RNA, proteins, or metabolites) are then captured using spatially barcoded probes, followed by library preparation for sequencing. The sequencing data are mapped back to their original spatial coordinates within the tissue, and computational analysis is performed to identify spatial expression patterns, cellular heterogeneity, and cell–cell interaction networks. This figure is an originally conceptualized by the authors and created using Canva (raghularajan, 2026).

2.1. CONCEPTS IN SPATIAL OMICS

2.1.1. Heterogeneity

Cellular heterogeneity can have different properties for the population if it ensures the robustness of the biological system to fluctuating environments and allows the population to adapt to a wider variety of environments, such as cellulose heterogenetic diversity in clonal populations due to multiscale organization.

Clonal heterogeneity occurs when there is an existence of the genetically distinct cell populations within the same tissue. This phenomenon is commonly seen in case of cancer, where the tumour cells accumulate mutations over time (11).

Spatial heterogeneity refers to variations in gene expression and cellular composition across different regions of tissue. For example, cells located near blood vessels, may exhibit different metabolic activities compared to cells located in oxygen—poor regions (12).

Temporal heterogeneity explains variations in cellular states such as components, structure or processes of a system over time during development, regeneration, or disease progression (13). Therefore, spatial omics technologies enable researchers to study these different forms of heterogeneity simultaneously.

2.1.2. Cell-cell communication:

Cells interact with each other through signalling molecules that regulate biological processes. Generally, signal molecules are referred as the chemical substances which involved in intracellular signal transduction or intercellular signal communication, while the small molecules that carries biological information are called messenger molecules (14). These interactions often involve ligand-receptor binding events that activate intracellular signalling pathway.

In plant systems, hormones such as auxin and cytokinin plays a pivotal role in regulating cell division, differentiation involves the binding of auxin molecules to the TIR1 receptor, which triggers transcriptional changes that sing immune response influence plant development. While cytokines are initiating diversified signalling pathways within specific tissue or activating a variety of cell types within a specific tissue (15) . In animal systems, cytokines and growth factors regulate various physiological processes, encompassing immune responses, and tissue repair. Understanding, these signalling networks are essential for studying disease mechanisms and therapeutic interactions.

2.1.2.1. Molecular mechanisms of cell-cell communication:

It is fundamental for cooking biological processes in multicellular organisms. Cell-signalling involves in transmitting the biological information between cells or subcellular components. Cellular communication comprises of essential processes include cell signal conduction and cell signal transduction. Communication occurs through several mechanisms including direct cell contact, chemical signalling, and extracellular vesicle exchange.

Most common form of cellular communication is chemical signalling, where cells communicate by releasing chemical messengers (ligand) that travel to and bind with specific receptors on other or same calls to trigger a response. This binding activates intracellular signalling cascades that regulate gene expression and cellular responses. Therefore, chemical signalling and ligand-receptor signalling are intrinsically linked(16). Examples of such signalling pathways include growth factor signalling in animal cells. Auxin signalling is mediated through receptors such as TIR1 which regulate transcription factors involved in cell division and differentiation. On the other hand, cytokinin signalling influences cell proliferation and organ development.

In animal biology, cytokines, chemokines, and growth factor mediate communication between immune cells, epithelial cells, and stomal cells. For instance, in the immune system, cytokine signalling regulates immune responses against pathogens and tumours. The signalling molecules produces with initiation of gene expression (17). Specific stimuli, such as extracellular signals or changes in intracellular conditions, activate the transcription and translation of specific genes, resulting in the creation of proteins or small molecules that function as signalling molecules. These signalling molecules are subsequently secreted or transported to target cells, where they bind to specific receptors and initiate intracellular signalling cascades that regulate diverse cellular responses. The overall process of cell–cell communication in both animal and plant systems, including signalling molecule production, secretion, receptor recognition, signal transduction, and downstream biological responses, is illustrated in **Figure 2**. Understanding these communication mechanisms is crucial for elucidating how cellular networks coordinate tissue development, physiological functions, and responses to environmental cues.

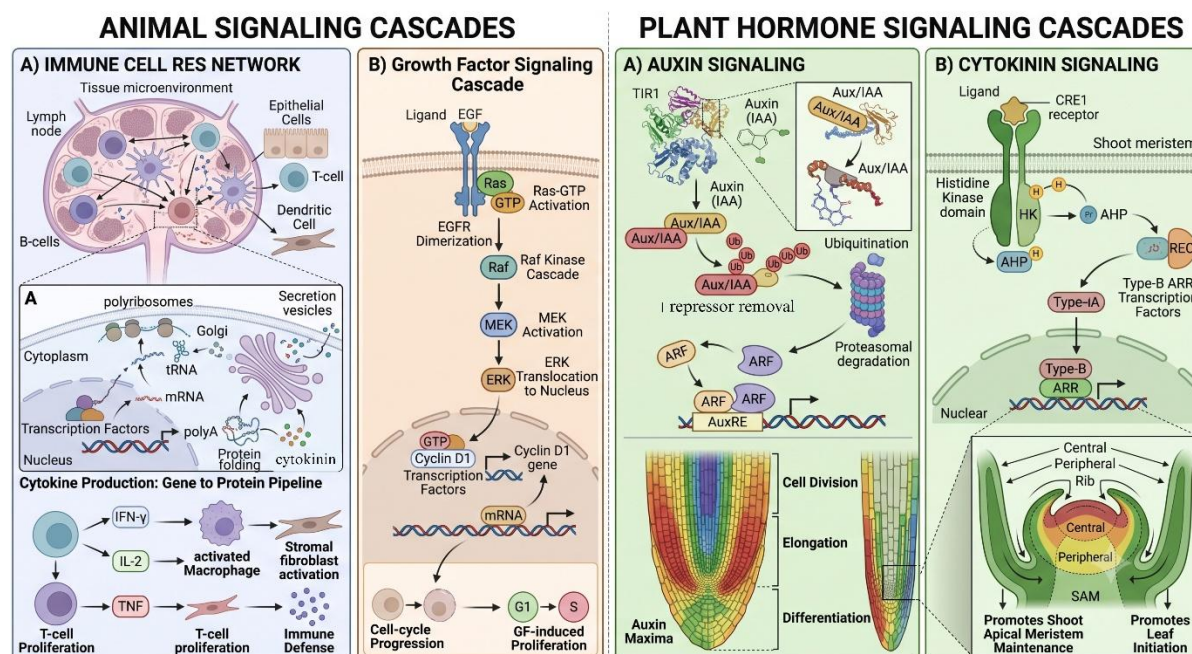


Figure 2. Overview of cell–cell communication through ligand–receptor signalling in animal and plant systems. External or internal stimuli induce gene transcription and translation, resulting in the production of signalling molecules. These molecules are secreted and bind to specific receptors on target cells, triggering intracellular signalling cascades that regulate gene expression and cellular responses. Representative examples include cytokine and growth factor signalling in animals, and auxin and cytokinin signalling in plants, ultimately controlling cell proliferation, differentiation, tissue development, and stress responses. This figure is an originally conceptualized by the authors and created using Canva (raghularajan, 2026).

3. Limitations of traditional omics approaches

Though traditional omics techniques play significant role in biological research, they have several limitations in the point of spatially organized tissues. Genomics remains restricted to DNA-level analysis, offering no functional insight and ignoring epigenetic and environmental influences, while transcriptomics demonstrates poor correlation with protein abundance, and proteomics involves highly complex analytical workflows (18).

Metabolomics and epigenomics further compound these challenges by generating highly dynamic datasets that are difficult to interpret in isolation (19).

One of such major limitations is the loss of spatial information, as of traditional omics system, tissues are typically homogenised during sample preparation. Due to the mixture of different regions of cell, the molecular measurements cannot be linked to their original locations within the tissue. In case of bulk transcriptomics, the average gene expression can be measured across many cells, which may obscure important differences between individual cells or rare cell populations (20). Traditional omics approaches also struggle to capture cellular heterogeneity. Many tissues contain multiple cell types with distinct functions, but bulk analysis cannot accurately resolve this distinctness.

Finally, conventional omics cannot directly reveal cell-cell interaction networks while they can identify molecules present in the tissue, they cannot determine how the cell communicate or influence each other (21). These limitations show the need for technologies that preserve spatial information while measuring the molecular data.

4. Importance of Spatial biology:

Spatial biology offers unique insights into genes, proteins, RNA, and therapeutic targets within a tissue's context. Even though the conventional omics technologies provided comprehensive molecular inventories, they lack spatial context. Understanding the spatial organization of these biological molecules is essential for interpreting within tissues are not randomly distributed; instead, they form structured environments where their position influences their behaviour and interactions (22). For example, in multicellular organisms, cells located near blood vessels receive higher oxygen and nutrient supply compared to those located in hypoxic regions. These environmental differences influence gene expression, metabolic activity, and cellular differentiation.

Furthermore, cellular interactions are highly dependent on spatial proximity. Cells communicate through direct physical contacts, ligand-receptor signalling, and secreted signalling molecules. This communication is essential for growth and regulate numerous biological processes such as immune responses, tissue repair, and developmental patterning (22). In the extracellular space, short-range movement of signals occurs through apoptotic pathway or directly from cell-cell via the symplastic pathway. Without spatial information, it is difficult to determine which cells interact with each other and how these interactions influence biological outcomes (23). Spatial omics technologies, provide powerful framework for studying the framework of life including tumour microenvironments, intricate neural circuitry, spatiotemporal immune dynamics and developmental trajectories. By combining molecular profiling with spatial mapping, researchers can analyse how gene expression patterns vary across tissue regions and how cellular interactions contribute to tissue organization.

Understanding spatial context is particularly important in diseases such as cancer. Tumour tissues consist of heterogeneous cell populations that interact with immune cells and stromal cells within the tumour microenvironment. Mapping these spatial interactions helps researchers understand tumour progression and identify potential therapeutic targets.

5. Connectogenomics:

Connectogenomics is an emerging field that integrates spatial omics with connectomics and network biology. It denotes the sequencing methods which records the physical interactions among cells or molecules directly as a network of edges (17). The goal of connectogenomics is to understand how molecular interactions form complex networks within tissue. In connectogenomics analysis, biological entities such as cells, genes or proteins are represented as nodes, while the interactions between them are represented as edges (24). These nodes and edges form networks that describe how biological systems are organized and it reveals how gene influences the biological components by linking the patterns of gene expression with the formation, organization and activity.

Connectogenomics has two main approaches. **Coordinates-first Approach-** begins with spatial gene expression data and predicts interactions based on spatial proximity and ligand-receptor analysis. **Edges-First Approach-** focuses on measuring the molecular interactions directly before constructing connectivity networks (24). Together these approaches provide a complete understanding of how gene expression influences the biological system.

5.1. Technologies in Connectogenomics:

Connectogenomics relies on advanced molecular technologies that can detect interactions between biological components within their spatial context. These technologies are sequencing-based analysis that enable researches to measure connecting between genes, proteins, and cells rather than measuring molecular abundance (24). Some of the most important technologies used in connectogenomics includes volumetric DNA Microscopy (vDM), Proximity Network Analysis/ Molecular Pixelation (PNA/ MPX). An overview of the experimental workflow and data outputs of the major sequencing-based connectogenomics technologies is presented in **Figure 3**.

ADVANCED SEQUENCING-BASED SPATIAL AND INTERACTION MAPPING TECHNOLOGIES

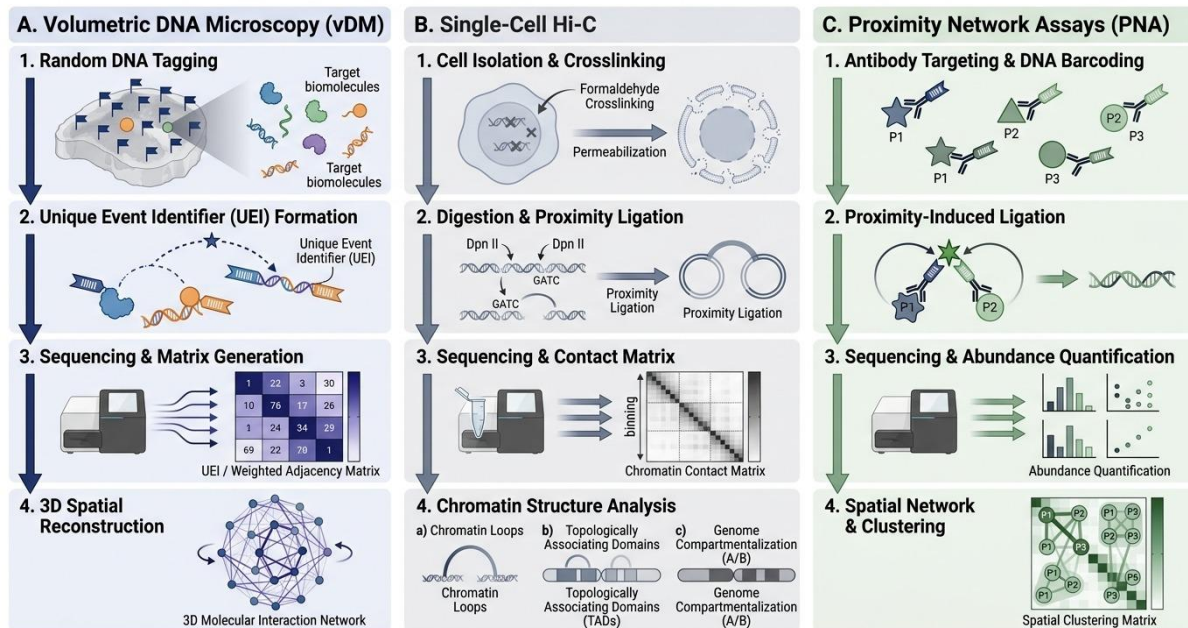


Figure 3. Overview of advanced sequencing-based spatial and interaction mapping technologies used in connectogenomics. (A) Volumetric DNA microscopy (vDM) randomly tags biomolecules with DNA barcodes, generates unique event identifiers (UEIs) through proximity-dependent amplification, and reconstructs three-dimensional molecular interaction networks from sequencing-derived weighted adjacency matrices. (B) Single-cell HI Throughput chromosome conformation capture (Hi-C) maps chromatin interactions by formaldehyde crosslinking, restriction enzyme digestion, proximity ligation, and sequencing, producing chromatin contact matrices that reveal chromatin loops, topologically associating domains (TADs), and genome compartmentalization. (C) Proximity Network Assays (PNA) employ DNA-barcoded antibodies to detect protein–protein interactions, followed by proximity-induced ligation, sequencing, abundance quantification, and spatial network clustering to characterize protein interaction networks at single-cell resolution. This figure is an originally conceptualized by the authors and created using Canva (raghularajan, 2026).

5.1.1. volumetric DNA Microscopy (vDM):

volumetric DNA Microscopy is an innovative sequencing-based technology of nucleotide-level genetic information and spatial organization from within the specimen itself. It also reconstructs spatial relationship between molecular without using conventional optical microscopy (25). Instead of capturing images, the technology encodes spatial proximity information directly into DNA sequences.

Volumetric DNA Microscopy starts by randomly tagging biomolecules inside a cell with unique molecular identifiers (DNA barcodes). These barcodes undergo amplification reactions that generate chimeric DNA (dimers) sequences when molecules are spatially close. The formation of dimers is recorded by integrating random nucleotides, called unique event identifiers (UEIs). By sequencing these DNA products, researchers can determine which molecules were physically near each other in the original tissue sample. After sequencing, the resulted data form a UEI matrix, which can be represented as a weighted adjacency matrix, where nodes represent molecules and edges represent spatial proximity relationships. Computational algorithms then reconstruct three-dimensional molecular interaction networks from the sequencing data (26).

This technology is particularly useful for studying complex molecular systems because it allows high-throughput measurement of molecular interactions without the limitations of optical imaging resolution. Volumetric DNA microscopy has been applied to investigate RNA-RNA spatial relationships and cellular organization in tissues through in-situ cDNA synthesis, spatial encoding, proximity bridging, decoding, and reconstruction of IVT (Invitro transcription) production.

5.1.2. Single-cell HI Throughput chromosome conformation capture:

It is a genome wide chromatin interaction mapping technique used to study the 3D structure of genome within individual cells. Similar to the traditional Bulk Hi-C, single-cell also includes chromatin crosslinking, cells permeabilization, DNA digestion, proximity ligation and library preparation(27). But the Bulk Hi-C represents an average of millions of cells, failing to show structural variation between the individual cells. The chromatin arrangements play a crucial role in regulating gene expression, because genes that are spatially close within the nucleus can influence each other's activity. The Hi-C technique works by crosslinking the cells using formaldehyde, which results in chromatin interactions within the nucleus. Then, DNA is digested by restricted enzyme such as Don II which cleaves at the four-letter palindromic motif GATC. Finally, the DNA fragments that

are physically close to each other are ligated. These ligated fragments represent chromatin contacts and are subsequently sequenced to determine which regions of the genome interact(28).

In single-cell Hi-C experiments, this process is performed on individual cells, allowing researchers to study chromatin architecture at single-cell resolution. The resulting data will be processed by optimal pre-processing of reads like trimming, demultiplexing, etc., read mapping, optional restriction fragment assignment, filtration of contacts and duplication and binning with generation of single-cell Hi-C maps. The interaction data can be represented as a chromatin contact matrix that reveals the patterns of genome organization(29). Single-cell Hi-C has provided important insights into chromatin loops, topologically associating domains (TADs), and genome compartmentalization. In connectogenomics, chromatin interaction matrices can be interpreted as connectivity networks that describes regulatory relationships between genomic regions.

5.1.3. Proximity network Assays (PNA):

Proximity Network Assays are the molecular technique designed to detect protein-protein interactions within cells. These assays convert physical interactions between proteins into DNA barcodes that can be sequenced. It is a DNA-based method to construct the three-dimensional spatial interaction networks of proteins in individual cells(30). In a typical proximity network assay, antibodies targeting specific proteins are conjugated with unique DNA tags.

When two proteins interact, their attached DNA tags are brought into close proximity. A ligation or amplification reaction then generates a DNA product that represents the protein-protein interaction. The resulting data structure supports analysis of protein abundance, and spatial clustering matrices which allows for a system-level view of protein organization(31). Proximity network assays allow high-throughput mapping of protein interaction networks directly within cells and tissues. This information is valuable for understanding signalling pathways, regulatory complexes, and molecular communication networks.

5.2. Computational Analysis:

Connectogenomic data are high-dimensional and require advanced computational methods. Complex datasets can be shown using dimensional reduction techniques like Principal Component Analysis (PCA), Uniform Manifold Approximation and Projection (UMAP), and t-distributed Stochastic Neighbour Embedding (t-SNE). Graph analysis methods identify network hubs and community structures. Software platforms used in spatial data analysis include **Seurat**, **Scanpy**, **Bioconductor**, and **Imaris**. The computational workflow and major analytical approaches employed in connectogenomics are summarized in Figure 4.

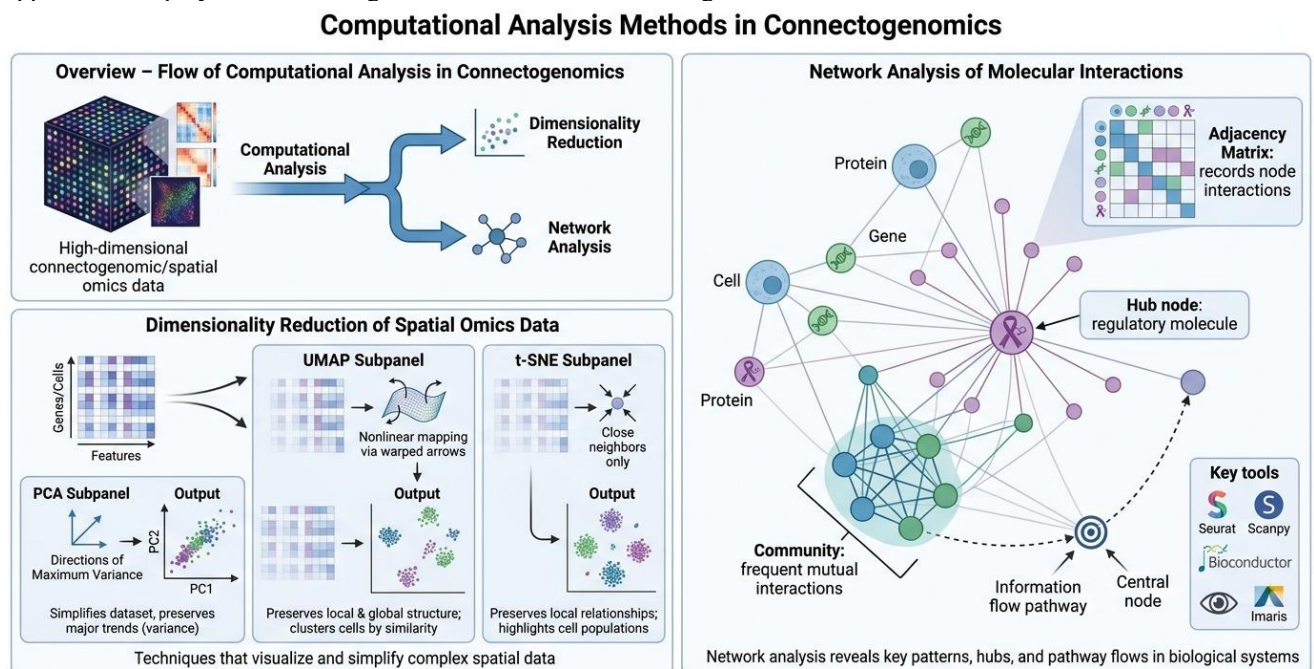


Figure 4. Computational analysis workflow in connectogenomics. High-dimensional connectogenomic and spatial omics datasets are processed using computational approaches that integrate dimensionality reduction and network analysis. Principal Component Analysis (PCA), Uniform Manifold Approximation and Projection (UMAP), and t-distributed Stochastic Neighbour Embedding (t-SNE) simplify complex datasets while preserving biologically meaningful patterns for visualization and clustering. Network analysis represents biological entities, including cells, genes, and proteins, as nodes connected by interaction edges stored in adjacency matrices. Graph-theoretical approaches identify hub nodes, community structures, centrality, and information flow pathways,

facilitating the interpretation of molecular interaction networks. Commonly used software platforms include Seurat, Scanpy, Bioconductor, and Imaris. This figure is an originally conceptualized by the authors and created using Canva (raghularajan, 2026).

5.2.1. Dimensionality Reduction in Spatial data:

Spatial omics datasets are typically high-dimensional because they contain information about thousands of genes measured across many spatial locations. Dimensionality reduction techniques are used to simplify these datasets while preserving the most important patterns(32).

5.2.2. **Principal component Analysis (PCA)** is a popular dimensionality reduction method that reduces a huge dataset while preserving key patterns and trends. It determines the directions of maximum variation in the data and projects it onto a lower-dimensional space.(33).

5.2.3. **Uniform Manifold Approximation & Projection (UMAP)** is an another widely used method that preserves both local and global structure in high-dimensional datasets. It is used for visualization similar to t-SNE but also for general non-linear dimension reduction(34). UMAP is particularly useful for visualizing clusters of cells in spatial transcriptomics data.

5.2.4. **t-distributed Stochastic Neighbour Embedding (t-SNE)** is also used for visualization high-dimensional biological data. It focuses on preserving local relationships between data points and it is commonly used to identify cell populations in single-cell datasets(35). These dimensionality reduction techniques allow researchers to visualize complex spatial data and identify patterns in gene expression and cellular connectivity.

5.3. Network Analysis in Connectogenomics:

In connectogenomics research, network analysis is a significant component, in which nodes represent the biological entities such as cells, genes, or proteins, while edges represent molecular interactions between them. These networks can be analysed using graph theory and computational algorithms to identify key connectivity patterns. One commonly used network representation is the adjacency matrix, which records the presence or absence of interactions between nodes. In connectogenomics, adjacency matrices may represent gene-gene interactions, protein-protein interactions, or cell-cell communication networks.

Graph-based analysis methods allow researchers to identify important features within biological networks. For example, **hub nodes** are nodes with many connections and often represent important regulatory molecules. **Community detection algorithms** identify clusters of nodes that interact more frequently with each other than with the rest of the network (24). Network analysis also enables the study of shortest of shortest paths and network centrality, which help determine how information flows within biological systems. These approaches are widely used in systems biology to understand complex regulatory networks. As illustrated in Figure 4, network analysis complements dimensionality reduction by identifying molecular interaction networks, hub nodes, community structures, and information flow within biological systems.

6.Applications of Connectogenomics

Connectogenomics has emerged as a transformative approach for understanding the spatial organization and interaction networks within biological systems by integrating spatial omics technologies with network analysis. This framework enables the reconstruction of cell–cell communication networks and molecular interactions at high spatial resolution, thereby providing insights into tissue organization, development, and disease progression (36). In cancer research, connectogenomics has become a valuable tool for characterizing the tumour microenvironment (TME), where malignant cells interact dynamically with immune, stromal, endothelial, and fibroblast populations. Spatial transcriptomics integrated with connectogenomic analysis enables the identification of ligand–receptor signalling networks, immune evasion mechanisms, and spatial communication niches that influence tumour progression, metastasis, and therapeutic response, thereby facilitating the discovery of prognostic biomarkers and personalized treatment strategies(36, 37). In neuroscience, connectogenomics combines molecular profiling with neural connectivity maps to investigate how gene expression regulates neuronal circuit formation and synaptic communication. This integrated approach provides valuable insights into the molecular basis of neurological disorders such as Alzheimer's disease, Parkinson's disease, epilepsy, and autism spectrum disorders by identifying changes in neuronal connectivity and gene regulatory networks that contribute to disease pathogenesis (21, 22). During embryonic development, connectogenomics enables the investigation of dynamic cell–cell communication and spatial regulatory networks that control tissue patterning, lineage specification, organogenesis, and morphogenesis. By integrating spatial transcriptomics with computational network analysis, researchers can reconstruct developmental trajectories and identify signalling pathways responsible for cellular differentiation and tissue organization, thereby supporting advances in developmental biology and regenerative medicine (5). In plant biotechnology, connectogenomics has gained increasing importance for studying plant growth, regeneration, and responses to environmental stresses. Spatial transcriptomic studies in species such as *Populus* and *Arabidopsis* have revealed spatially coordinated auxin- and cytokinin-mediated signalling networks that regulate root regeneration, vascular differentiation, and organ

development (38). These findings improve our understanding of plant developmental biology and provide opportunities to enhance micropropagation, genetic transformation, crop improvement, and the development of stress-tolerant cultivars. Furthermore, connectogenomics is expected to play a pivotal role in precision medicine by generating patient-specific maps of cellular interactions within tissues, enabling the identification of disease-specific biomarkers, molecular signatures, and dysregulated signalling pathways. Such comprehensive spatial interaction maps can improve disease diagnosis, predict therapeutic response, facilitate targeted drug development, and support individualized treatment strategies, making connectogenomics an emerging cornerstone of next-generation precision healthcare (36).

Application Area	Major Application	Key Outcomes	References
Cancer Research	Mapping tumour microenvironment and cell–cell communication	Identification of ligand–receptor interactions, immune evasion pathways, biomarkers, and therapeutic targets	(36, 37)
Neuroscience	Integrating gene expression with neural connectivity	Understanding neural circuit organization and mechanisms underlying neurological disorders	(21)
Developmental Biology	Reconstruction of developmental communication networks	Identification of regulatory pathways controlling cell differentiation, tissue patterning, and organogenesis	(22)
Plant Biotechnology	Analysis of hormone signalling and regeneration networks	Improved micropropagation, crop improvement, stress tolerance, and regeneration studies	(39)
Precision Medicine	Patient-specific spatial interaction mapping	Biomarker discovery, disease diagnosis, targeted therapy, and personalized medicine	(38)

Table 1: Major Applications of Connectogenomics Across Biological and Medical Sciences.

It provides an overview of the principal applications of connectogenomics in cancer research, neuroscience, developmental biology, plant biotechnology, and precision medicine. It outlines the major biological applications, key outcomes, and representative references, illustrating the broad potential of connectogenomics in deciphering spatial cellular communication and advancing both biomedical and agricultural research

7.Importance of Connectogenomics in System Biology:

Connectogenomics plays a crucial role in advancing system biology by enabling the study of biology systems as interconnected networks rather than isolated components. Traditional molecular biology approaches often focus on individual gene or proteins, but biological functions are typically regulated by complex interaction networks. By integrating spatial omics data with network analysis, connectogenomics allows researchers to reconstruct interaction networks that describe how cells communicate within tissues. These networks provide insights into how biological systems maintain homeostasis and respond to environmental changes.

In addition, connectogenomic analysis helps identify regulatory modules within biological networks. These modules represent groups of interacting genes or proteins that perform specific biological functions. Identifying such modules can help researchers understand disease mechanisms and develop targeted therapies. Overall, connectogenomics represents an important step toward a system-level understanding of biological processes.

8. challenges in connectogenomics

Connectogenomics has enormous potential for revealing spatial cellular communication and tissue organization; however, several technical, computational, and biological challenges currently limit its widespread application. The integration of spatial omics datasets with network-based analytical frameworks generates highly complex, high-dimensional data that require sophisticated computational methods and substantial computing resources(37, 40). A single spatial transcriptomics experiment can produce expression profiles for thousands of spatial locations, and subsequent network reconstruction may involve millions of potential cell–cell interactions, making data storage, processing, visualization, and interpretation computationally demanding(40). In addition, connectogenomic analyses involve multiple computational steps, including data preprocessing, normalization, dimensionality reduction, spatial clustering, ligand–receptor interaction prediction, graph construction, and network analysis, all of which require specialized bioinformatics expertise. Many currently available analytical tools were originally developed for single-cell transcriptomics and are not optimized for spatial interaction networks, highlighting the need for new algorithms specifically designed for connectogenomic applications (41). Another important limitation is the spatial resolution of existing technologies. Although recent advances have greatly improved spatial profiling, several platforms still measure gene expression from groups of cells rather than individual cells, making it difficult to accurately identify direct cellular interactions and signalling events. Achieving true single-cell and subcellular spatial resolution remains a major technological objective for future

connectogenomics research (40). Furthermore, integrating multiple omics layers—including transcriptomics, proteomics, epigenomics, metabolomics, and spatial imaging data—remains a significant analytical challenge because each dataset differs in scale, resolution, data structure, and technical variability. Developing standardized analytical pipelines, common data formats, and robust multimodal integration frameworks is essential for constructing comprehensive and biologically meaningful interaction networks (42). Finally, many interaction networks identified through computational approaches represent predicted rather than experimentally validated biological interactions. Therefore, complementary experimental techniques such as multiplexed fluorescence imaging, immunohistochemistry, CRISPR-based perturbation studies, and functional validation assays are required to confirm predicted signalling pathways and ensure biological accuracy(37). Addressing these challenges through advances in spatial technologies, computational methods, standardized analytical frameworks, and experimental validation will be essential for realizing the full potential of connectogenomics in biological research, precision medicine, and agricultural biotechnology (42).

Table 2. Major Challenges in Connectogenomics and Potential Solutions

Challenge	Description	Impact on Connectogenomics	Possible Solutions	References
High data complexity	Massive spatial omics datasets with millions of potential cellular interactions	Difficult data storage, processing, visualization, and interpretation	High-performance computing, AI and machine learning algorithms	(37, 40)
Computational limitations	Complex workflows requiring specialized bioinformatics tools	Limited accessibility and slower data analysis	Development of dedicated connectogenomic software and optimized algorithms	(42)
Resolution limitations	Many platforms capture multiple cells instead of true single-cell resolution	Reduced accuracy in identifying direct cell–cell interactions	Higher-resolution spatial transcriptomics and subcellular imaging technologies	(40)
Multi-omics data integration	Difficulty integrating transcriptomic, proteomic, epigenomic, and metabolomic datasets	Incomplete reconstruction of biological interaction networks	Standardized data formats and multimodal integration frameworks	(42)
Validation of predicted interactions	Computationally inferred networks require biological confirmation	Risk of false-positive interactions	Experimental validation using imaging, CRISPR, and functional assays	(37)

It summarizes the major challenges associated with connectogenomics, including technical, computational, and biological limitations. It highlights the impact of each challenge on connectogenomic research and outlines potential solutions and future directions for improving spatial multi-omics integration, network analysis, and experimental validation.

9. Future Prospects of Connectogenomics:

Connectogenomics is a rapidly evolving field that combines spatial omics technologies with network biology to understand molecular interactions within tissues. Although the field is still emerging, ongoing technological and computational advancements are expected to significantly expand its capabilities and applications in the coming years. It also uses Higher Resolution Spatial Technologies which is One of the most important future directions in the development of higher resolution spatial technologies. Current spatial omics methods often analyse gene expression at the level of groups of cells rather than individual cells. Advances in sequencing technologies, imaging methods, and molecular barcoding strategies are expected to enable true single-cell and subcellular resolution. Such improvements will allow researchers to map molecular interactions with greater accuracy and reconstruct detailed cellular connectivity networks within tissues. This field has a higher chance of developing by Integration with Artificial Intelligence which is a promising area that integration of artificial intelligence and machine learning in connectogenomic analysis. Connectgenomics datasets are highly complex and involve large-scale interaction networks. Machine learning algorithms can help identify hidden patterns in these datasets, predicts interaction networks, and detect important regulatory pathways. AI- based tools may also improve the improve the interpretation of spatial omics data and accelerate discoveries in system biology. The integration of multi-omics data represents another important future prospect. Biological systems are regulated by multiple layers of molecular information, including gene expression, protein interactions, epigenetic modifications, and metabolic pathways. Future connectogenomics studies are expected to integrate spatial transcriptomics to generate comprehensive models of cellular communication networks. Precision Medicine Application Connetogenomics also holds great potential for precision medicine and disease research. By mapping molecular interactions within

diseased tissues, researchers can identify biomarkers and therapeutic targets associated with specific disease states. In cancer research, for example, connectogenomic analysis may help reveal interaction networks between tumour cells and immune cells, leading to improved strategies for immunotherapy and personalized treatment. Whole-Tissue Connectivity Maps Ultimately, the long-term goal of connectogenomics is to create comprehensive cellular connectivity maps of tissues and organs, similar to the connectome maps used in neuroscience. These connectivity maps will provide a detailed understanding of how molecular interactions shape biological structure and function.

10. CONCLUSION:

Spatial omics technologies have significantly transformed the study of biological systems by enabling the measurement of molecular information within the spatial context of tissues. Unlike traditional omics approaches, which often lose spatial information during sample processing, spatial omics preserves tissue architecture and allows researchers to investigate cellular heterogeneity, tissue organization, and molecular signalling pathways in their natural environment. These technologies have provided valuable insights into complex biological processes such as development, regeneration, and disease progression.

However, many existing spatial analysis methods infer cellular interactions indirectly, often relying on spatial proximity or ligand-receptor co-expression. While these approaches are useful, they may not fully capture the true molecular interactions occurring within the tissues. The emerging field of connectogenomics addresses these limitations by integrating spatial omics with network biology and connectogenomics to study genome-wide interaction networks.

Connectogenomics focuses on identifying biological entities as nodes and their interactions as edges to reconstruct connectivity networks within the tissues. Advanced technologies such as volumetric DNA microscopy, single-cell Hi-C, and proximity network assays enable the direct measurement of molecular interactions. Combined with computational tools and network analysis methods, these technologies allow researchers to map cellular communication and regulatory pathways more accurately.

Applications of connectogenomics in cancer research, neuroscience, and plant biotechnology demonstrate its potential to uncover previously hidden interaction networks. Although challenges remain, including data complexing, computational demands, and experimental costs, ongoing, technological and analytical advancements are expected to overcome these limitations. Overall, connectogenomics represents a promising approach for understanding cellular connectivity and the spatial organization of biological systems, offering new opportunities for research in medicine, agriculture, and system biology.

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