Molecular Systems Architecture of Human Knee Osteoarthritis

A Research Collaboration Between

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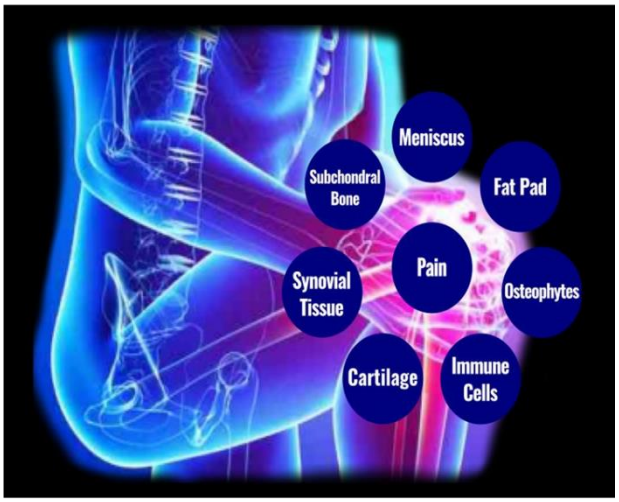
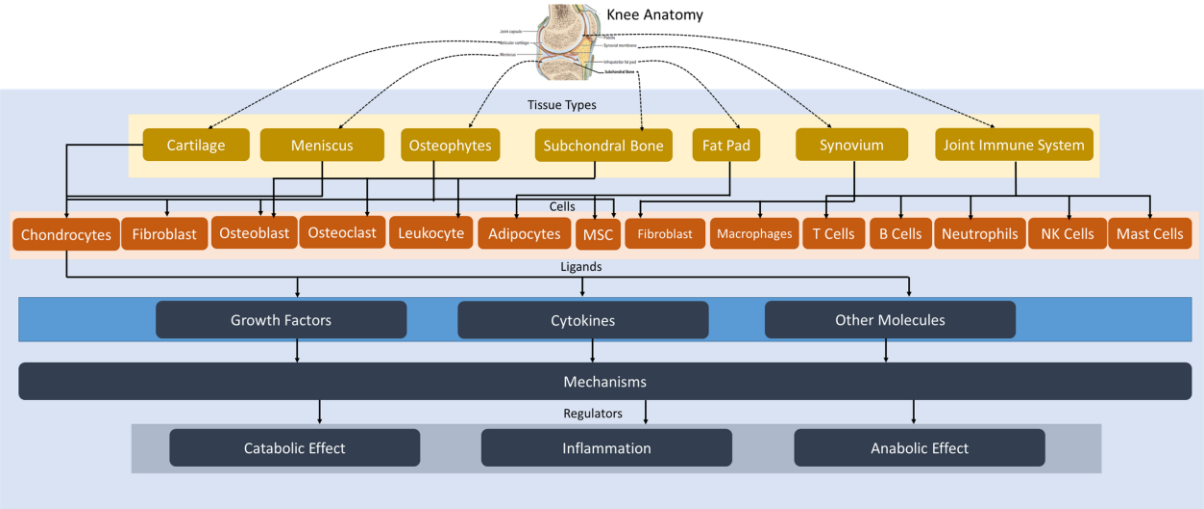


A collaboration between the Division of Orthopedics, Arthritis Program (UHN) and CytoSolve, Inc., a multi-scale molecular systems architecture has been created to provide a comprehensive and integrative molecular systems understanding of human knee osteoarthritis (OA). The architecture is constructed through a supervised bioinformatics process beginning with nearly 20,000 scientific papers curated to a refined set of 5,243 papers from which molecular interactions are extracted. The systems architecture is accessed through an easy-to-use interface to traverse the complexity of knee OA systems biology from tissue to cell to an ensemble of molecular interactions whereby one may discover the particular paper from which an interaction is derived. The systems architecture maintains its currency by enabling the research community to provide feedback that can be reviewed, refereed and incorporated. The architecture provides a framework for developing new educational tools as well as to support research that aims to investigate disease mechanisms, and identify potential therapeutic targets for the treatment of knee OA.

"3D" viewer for systems architecture of knee OA:



The first layer is the knee with its anatomical components. The second layer allows traversal from a particular anatomical component to cell types within that component i.e. chondrocytes within cartilage. The third to the nth layer allow one to progress downward to the ensemble as well as individual molecular reactions. The final layer provides clickable access to the particular paper from which that particular reaction is derived.

Search criteria to build the system architecture: In this effort, PubMed was the main source database to search for scientific articles published until June 2017. An initial search is performed to identify as many relevant citations. The resultant initial list included 20,231 research papers. The process is delineated in the PRISMA flowchart in Figure 1.

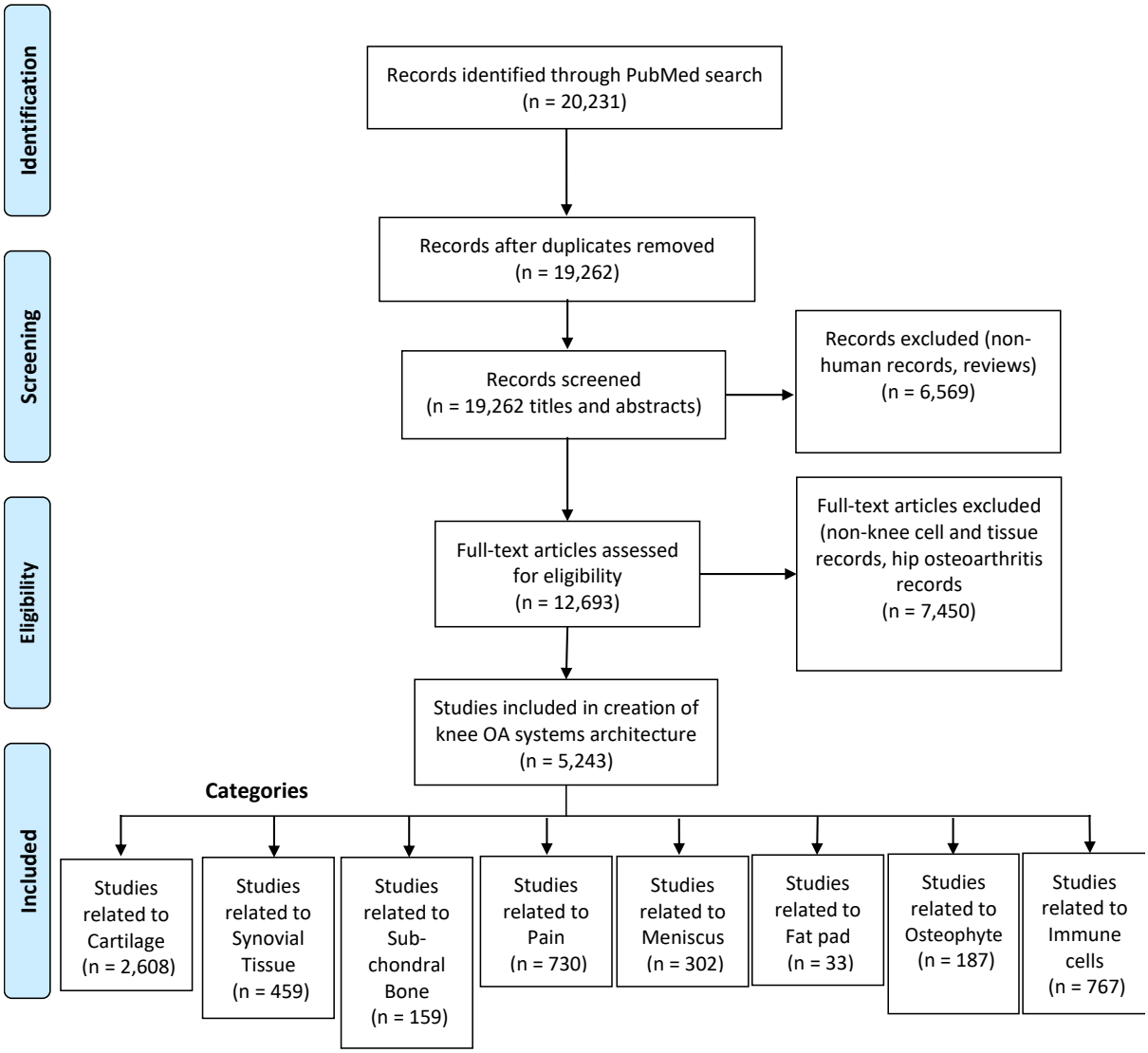


Figure 1. PRISMA Flow diagram for literature search

After removing 969 duplicate records, 19,262 records were screened to exclude non-human studies and review articles, which resulted in 12,693 records. We further screened 12,693 records to exclude non-knee cell/tissue and hip/spine osteoarthritis studies. We included a total of 5,243 studies that were reviewed to construct the systems architecture of knee osteoarthritis. The MeSH keywords used in the identification of literature are listed in Appendix 1.

Rules for Construction of the Systems Architecture of Human Knee Osteoarthritis

From each relevant paper, molecular pathway information is extracted. This process requires following particular agreed upon filtering rules. These filtering rules ensure quality control of the molecular mechanisms being derived from the refined list of articles. For example, the experiments in the study to be used for extraction of molecular mechanisms have to be performed on the OA cells and not from normal cells. If the study has data from different species including humans, only the relevant data from human experiments should be used. The full set of filtering rules to ensure quality are listed below:

Definitions:

Independent Variable: A biological molecule that affects the outcome of an experiment, e.g. IL-1 is an Independent Variable if it affects the experimental outcome such as MMP-13 (mRNA and/or Protein) in knee OA joint cells.

Types of Independent Variables:

- Ligands
- Inhibitors
- Single molecule drugs

Dependent Variable: A biological molecule that is the outcome of an experiment affected by the Independent Variable, e.g. catabolic biomarker such as MMPs, ADAMTS5.

Types of Dependent Variables:

- Enzymes
- Transcription factors
- mRNA

- Expressed proteins
- Small molecules (byproducts of metabolism e.g. nitrite)

Table 1: Graphical Notations for Molecular Pathway Representation

Variable Type	Entity	Graphical Notation	Position in the Cell	Example
Independent Variable	Small Molecule/ Drug/ Inhibitor		Outside the membrane	
	Ligand		Outside the membrane	
Dependent Variable	Receptor		Transmembrane	
	Enzyme	Name of entity without Background	Cytosol	ERK1/2
	Outcome Entity	OUTCOME	Cytosol	MMP-13
	Outcome Phenomenon	Phenomenon	Cytosol	CATABOLISM

Table 2: Rules for Selecting Data from Papers

Human
Cell type: Only From OA Knee (Not from normal cells)
If no distinction between cells from OA patients and normal, use all the relevant data
If Hip/ Knee OA cells are indistinguishable, use the data
Only choose human data in the event more than one species is studied
Do not use data from referenced studies

<u>Rules for Pathway Extraction and Representation</u>

1. Select information present only in the figures of within the RESULTS section, if and only if such data is coming from within the paper and not any referenced work.

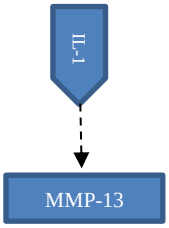
2. Making the connections in the pathway

    a. If there is **direct evidence** of two molecules interacting with each other to form a complex, represent it by solid black arrow

    This will ONLY be used in following case:

        i. receptor – ligand interaction

    b. If there is evidence of an Independent Variable increasing activity/expression a Dependent Variable through an unspecified mechanism, represent the interaction with dashed black arrow (e.g. IL-1 increasing the expression of MMP-13)



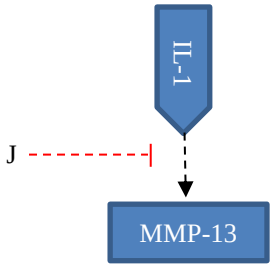
    c. If an Independent Variable is inhibiting/ reducing expression or levels of either the molecule or a process, there can be two different variations for this rule:

        i. For a small molecule/inhibitor that is inhibiting/reducing expression or levels a Dependent Variable, via unspecified mechanisms, the graphical representation is as follows:

Example: Results showing “Inhibitor I lowered the expression levels of MAPK” should be represented as follows

ii. For a molecule inhibiting/reducing expression or levels in a process, the graphical representation is as follows:

Example: Results showing “Independent Variable J reduced expression of IL-1 induced MMP-13”



d. If an Independent Variable is inhibiting a Dependent Variable and the current study results DO show Independent Variable binding to the dependent variable and subsequent inhibition, then use the solid red arrow with flat end to represent the interaction.

Example: Results showing “Inhibitor I inhibited MAPK” should be represented as follows

e. Represent ALL the interactions studied in the Results section related to human data.

Appendix A

MeSH keywords used to identify relevant literature.

Cartilage	
	human osteoarthritis cartilage chondrocytes MMP-13 NOT review
	human osteoarthritis cartilage chondrocytes apoptosis NOT review
	human osteoarthritis chondrocytes Autophagy NOT review
	human osteoarthritis chondrocytes BMP NOT review
	human osteoarthritis chondrocytes fibroblast growth factor NOT review
	human osteoarthritis chondrocytes GM-CSF growth factor NOT review
	human osteoarthritis chondrocytes microRNA NOT review
	human osteoarthritis chondrocytes mTOR NOT review
	human osteoarthritis chondrocytes Adipogenesis NOT review
	human osteoarthritis chondrocytes ADAMTS4 NOT review
	human osteoarthritis chondrocytes ADAMTS5 NOT review
	human osteoarthritis chondrocytes COX-2 NOT review
	human osteoarthritis chondrocytes INOS NOT review
	human osteoarthritis chondrocytes ZIP8-MTF1 NOT review
	human osteoarthritis chondrocytes PPARg NOT review
	human osteoarthritis chondrocytes wnt NOT review
	human osteoarthritis chondrocytes TGF-β NOT review
	human osteoarthritis chondrocytes IL-6 NOT review
	human osteoarthritis chondrocytes TNF NOT review
	human osteoarthritis chondrocytes hedgehog NOT review
	human osteoarthritis knee cartilage chondrocytes metabolism NOT review
	human osteoarthritis chondrocytes Aggrecan NOT review
	human osteoarthritis chondrocytes Type II Collagen NOT review
	human osteoarthritis chondrocytes interleukin-1 NOT review
	human osteoarthritis chondrocytes IGF-1 NOT review
	human osteoarthritis chondrocytes AKT NOT review
	human osteoarthritis chondrocytes type X collagen NOT review
	human osteoarthritis chondrocytes cartilage oligomeric matrix protein NOT review

Knee Tissue Type/ Function	Search Query
Subchondral Bone	
	human osteoarthritis osteoblast NOT review
	human osteoarthritis leukocytes inflammation NOT review
	human osteoarthritis osteoclast NOT review
Synovial Tissue	
	human osteoarthritis TNF (synovial tissue or synovium) NOT review
	human osteoarthritis TGF (synovial tissue or synovium) NOT review
	human osteoarthritis Autophagy (synovial tissue or synovium) NOT review
	human osteoarthritis Ephrin B2 (synovial tissue or synovium) NOT review
	human osteoarthritis microRNA (synovial tissue or synovium) NOT review
	human osteoarthritis mTOR (synovial tissue or synovium) NOT review
	human osteoarthritis NF-κB (synovial tissue or synovium) NOT review
	human osteoarthritis IL-6 (synovial tissue or synovium) NOT review
	human osteoarthritis JAK/STAT (synovial tissue or synovium) NOT review
	human osteoarthritis Type I Collagen (synovial tissue or synovium) NOT review
Pain	
	human osteoarthritis nerve growth factor NOT review
	human osteoarthritis brain neurotrophic factor NOT review
	human osteoarthritis GDNF NOT review
	human osteoarthritis PGE2 NOT review
	human osteoarthritis prostanoid receptors NOT review
	human osteoarthritis Autotaxin NOT review
	human osteoarthritis Lysophosphatidic acid NOT review
	human osteoarthritis "Leukotriene B4" NOT review
	human osteoarthritis Kinins NOT review
	human osteoarthritis kinins receptors NOT review
	human osteoarthritis substance P NOT review
	human osteoarthritis serotonin NOT review
	human osteoarthritis Catecholamines NOT review
	human osteoarthritis Norepinephrine NOT review
	human osteoarthritis metanephrines NOT review
	human osteoarthritis epinephrine NOT review
	human osteoarthritis pain fibroblast growth factor NOT review
	human osteoarthritis vascular endothelial growth factor NOT review
	human osteoarthritis PKCδ NOT review
	human osteoarthritis ATP NOT review
	human osteoarthritis histamine NOT review
	human osteoarthritis pain IL-6 NOT review
	human osteoarthritis pain IL-1 NOT review
	human osteoarthritis pain TNF NOT review
	human osteoarthritis IL-17 NOT review
	human osteoarthritis IL-18 NOT review
	human osteoarthritis CCL2 NOT review
	human osteoarthritis MCP-1 NOT review
	human osteoarthritis CCR2 NOT review
	human osteoarthritis Fractalkine NOT review
	human osteoarthritis CX3CR1 NOT review

Meniscus	
	human osteoarthritis meniscus NOT review
	human osteoarthritis meniscus Apoptosis NOT review
	human osteoarthritis meniscus Autophagy NOT review
	human osteoarthritis meniscus NF-κB NOT review
	human osteoarthritis meniscus IL-6 NOT review
	human osteoarthritis meniscus IL-1β NOT review
	human osteoarthritis meniscus TNF NOT review
	human osteoarthritis meniscus matrix metalloproteinases 13 NOT review
	human osteoarthritis meniscus COX-2 NOT review
	human osteoarthritis meniscus iNOS NOT review
	human osteoarthritis meniscus PPAR NOT review
	human osteoarthritis meniscus TGF NOT review
	human osteoarthritis meniscus Wnt NOT review
Infrapatellar Fat Pad	
	human osteoarthritis fat pad adipogenesis NOT review
	human osteoarthritis fat pad leptin NOT review
	human osteoarthritis fat pad adiponectin NOT review
	human osteoarthritis fat pad IFN-γ NOT review
	human osteoarthritis fat pad adipocytes NOT review
	human osteoarthritis fat pad mesenchymal stem cells NOT review
Immune Cells	
	human osteoarthritis macrophages NOT review
	human osteoarthritis plasma cells NOT review
	human osteoarthritis T cells NOT review
	human osteoarthritis B cells NOT review
	human osteoarthritis Neutrophils NOT review
	human osteoarthritis Mast Cells NOT review
	human osteoarthritis NK Cells NOT review
Osteophyte	
	human osteoarthritis osteophyte NOT review

Appendix B

List of literature used to create the systems architecture

https://docs.google.com/spreadsheets/d/1O7o2IeEBLF7Q-BKlG-ehc4wdrjFDQEpC9MN346w3oGM/edit?usp=sharing

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