

BACKGROUND AND PURPOSE OF THE STUDY

Osteoarthritis (OA) is a prevalent musculoskeletal disease. Molecular mechanisms of OA are not well understood, and no treatments are available to restore degraded cartilage or decelerate disease progression. Representative novel in vitro models are needed to advance OA pathology research, allowing development of efficient disease-modifying OA drugs (DMOADs). To reply to this unmet need, we present uKnee, a novel human mechanically active 3D OA cartilage-on-chip model.

uBEAT®: THE BEATING ORGAN-ON-CHIP TECHNOLOGY

uBeat® Platform (Figure 1) enables highly controlled and easily modified mechanical stimulation to form 3D microtissues.^[1] Integration of uBeat® enables the generation of organ-on-chip models with improved functionality and resemblance to pathological states.^[2, 3, 4]

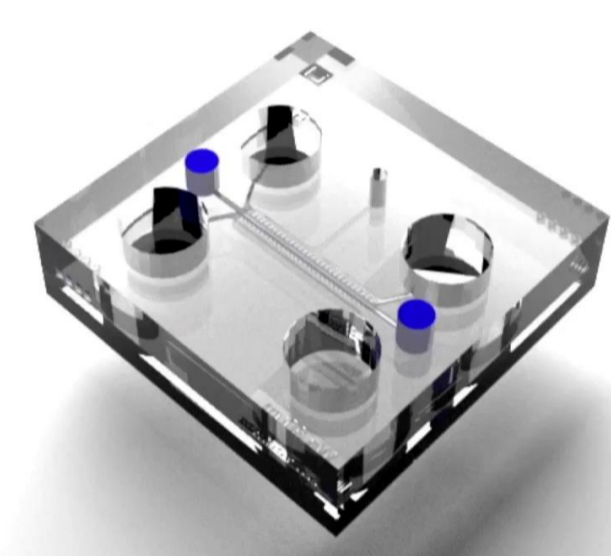


Figure 1: uBeat® Platform

A HUMAN HEALTHY CARTILAGE-ON-CHIP (CoC) MODEL

Human primary chondrocytes from healthy donors were embedded in hydrogel matrix and grown for 14 days in standard chondrogenic medium in the uBeat® Platform without mechanical stimulus.^[4] After 2 weeks in culture, the CoC model represents a gene expression profile correlating with healthy human cartilage and articular cartilage-like ECM deposition (Figure 2).

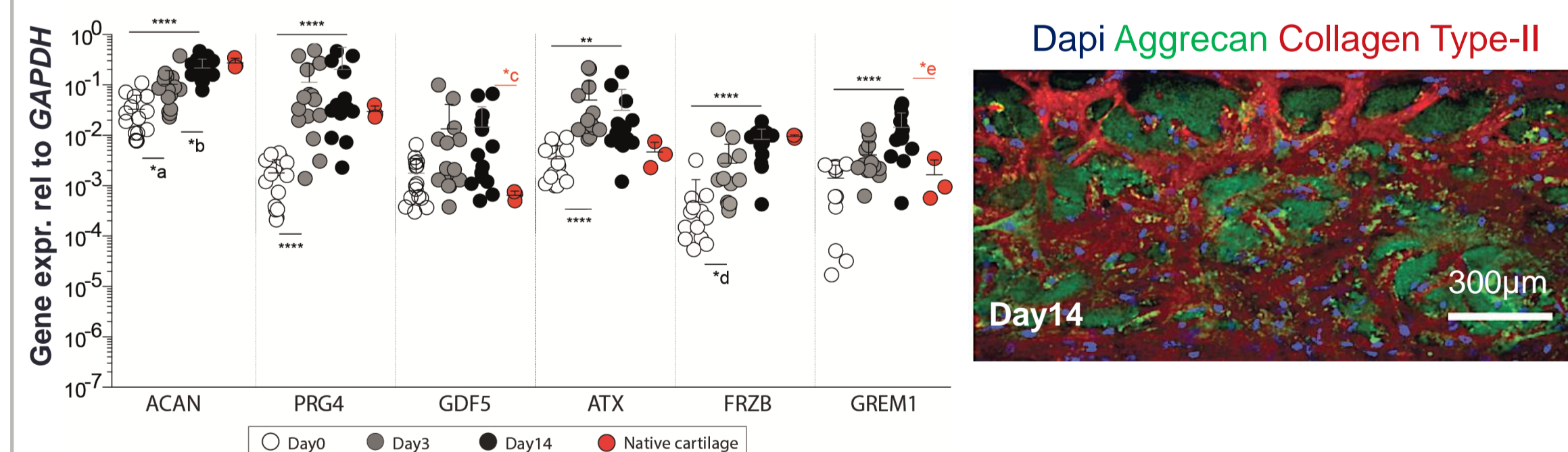


Figure 2: During the culture period, the expression of cartilage-related genes (ACAN, PRG4, GDF5, ATX, FRZB, GREM1) resembles their expression in healthy human (native) cartilage (in red). After 14 days, the cultures demonstrate organized matrix as shown by aggrecan and type-II collagen staining.

uBEAT® APPLICATION TO uKNEE MODEL: INDUCTION OF OA-LIKE TRAITS

The obtained healthy CoCs are subjected to mechanical stimulus by applying hyper-physiological 30% confined compression (HPC) using uBeat® Platform. The mechanical stimulus induced the following osteoarthritic changes: i) enhancement of catabolic and inflammatory responses and ii) switching towards hypertrophic cartilage phenotype (Figure 3).

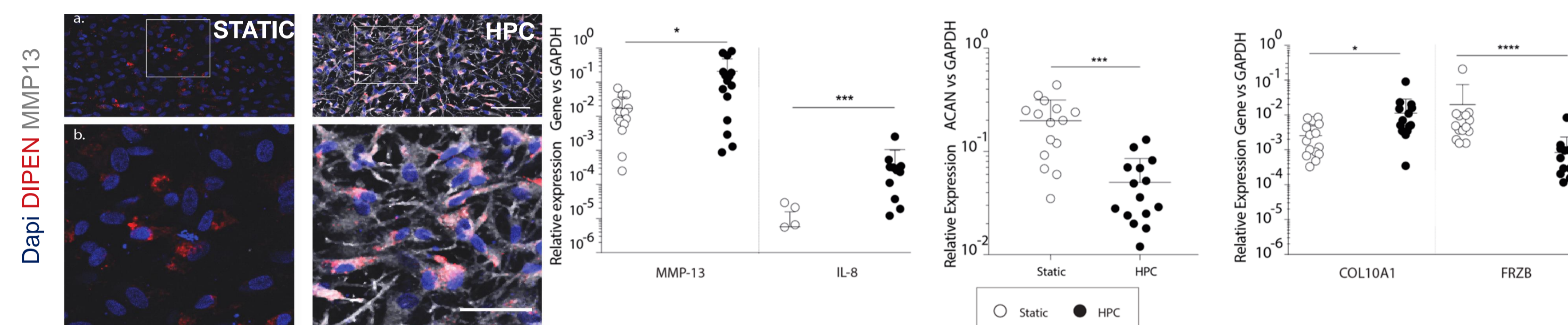


Figure 3: HPC induced osteoarthritic changes in the CoCs, including enhancement of catabolic and inflammatory responses demonstrated by increased deposition of MMP13, upregulation of IL-8 and downregulation of ACAN gene expression, and a swift towards hypertrophic cartilage demonstrated by upregulation of COL10A1 and downregulation of FRZB gene expression.

uKNEE AS A TOOL TO SCREEN DMOADs AND MEDICAL DEVICES

uKnee demonstrates treatment responses to anti-inflammatory and anti-catabolic clinically used standard-of-care (SOC) therapies (Rapamycin, Celecoxib, IL-1Ra and dexamethasone) and to two hyaluronic acid-based medical devices (Hymovis® and Hyalgan®, Fidia Farmaceutici) (Figure 4).

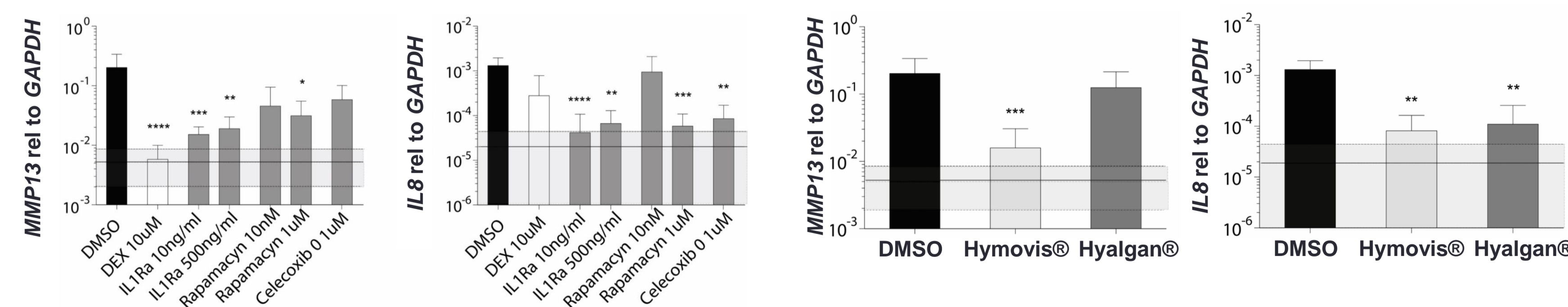


Figure 4: Treatment responses are examined as changes in MMP13 and IL-8 gene expression. The horizontal line in all figures represents the expression levels in healthy cartilage.

Summary of uKnee: a mechanically active in vitro cartilage-on-chip model of human OA

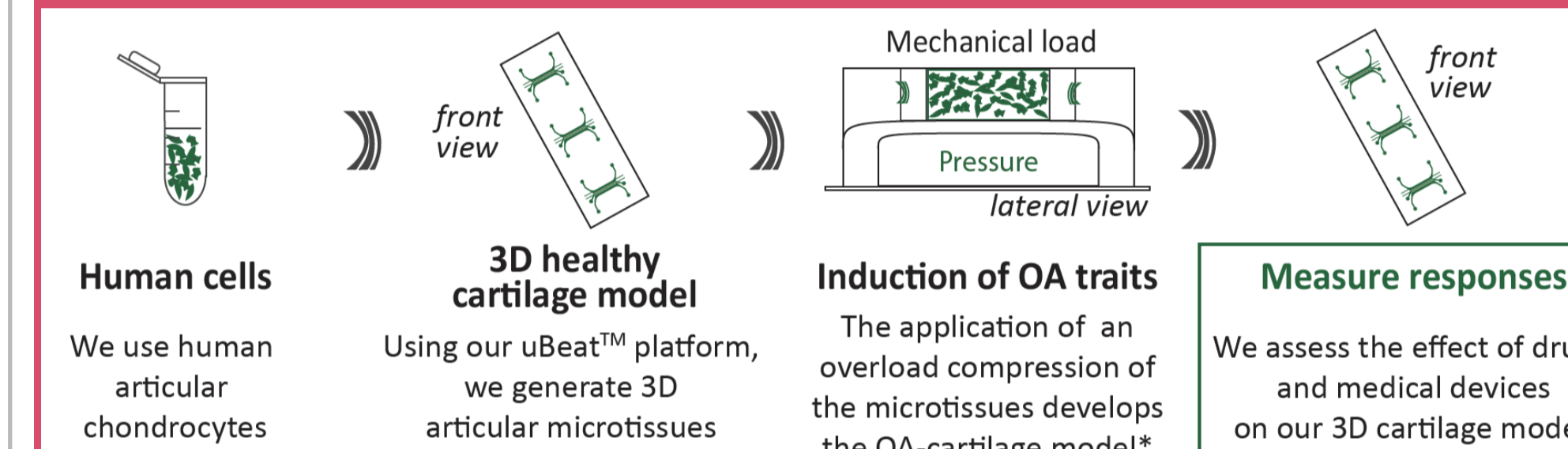


Figure 5: Summary of the uKnee OA CoC model. Human primary chondrocytes are cultured in 3D organ-on-chips. After maturation of the cartilage constructs, they are subjected to HPC stimulus using uBeat® Platform that stimulates osteoarthritic changes. The uKnee model can be effectively used to test effects of DMOADs and medical devices.

In uKnee, 3D healthy human cartilage-like constructs are subjected to a hyper-physiological OA resembling mechanical microenvironment provided by the proprietary uBeat® technology.

The uKnee model can be exploited to analyse gene expression profiles and molecular mechanisms corresponding to human OA onset and study effects of SOC anti-OA therapies.

CONCLUSION AND PERSPECTIVE EXPLOITATION OF uKNEE

The HPC provided by the uBeat® Platform is fundamental for eliciting OA pathogenesis in the uKnee CoC model, enabling predictive screening of DMOADs and medical devices. In the next phase of research, the uBeat® Platform will be utilized for more complex representation of a joint-on-a-chip model, integrating subchondral bone and synovial compartments.