SHAPED DEFINED-PLGA MICROPARTICLES: THERAPEUTIC EFFICACY AND TRIBOLOGICAL BEHAVIOR IN OSTEOARTHRITIS

Agnese Fragassi[a,b], Antonietta Greco[a], Martina Di Francesco[a], Luca Cesarecciu[a], Aimar Abu Ammar[c], Haytam Kasem[c], Craig Duvall[d] and Paolo Decuzzi[a]

[a] Laboratory of Nanotechnology for Precision Medicine, Fondazione Istituto Italiano di tecnologia– Genoa, Italy
[b] Department of Chemistry and Industrial Chemistry, University of Genova, 16146 Genova, Italy
[c] Azrieli College of Engeneering, Jerusalem
[d] Department of Biomedical Engineering, Vanderbilt University, Nashville, Tennessee 37232, United States

Presenting author: agnese.fragassi@iit.it

Background

Osteoarthritis (OA) is the most common form of arthritis and chronic degenerative joint disease. It is characterized by subchondral bone sclerosis, synovial membrane inflammation and progressive cartilage degradation. All these alterations result in chronic pain, functional loss and, eventually, permanent disability [1]. Currently, there is no disease-modifying drug available to reverse the progression of the OA and conventional therapies provide only a temporary relief from the symptoms. Considering the localized nature of the disease, the direct injection of therapeutic agents into the joint cavity is a clinically valuable approach. However, intra-articular treatments can be rapidly cleared through the synovial vasculature (small molecules) or lymphatic drainage (macromolecule). In this context, the development of drug delivery systems, that can act as biometarial depot, represents a promising strategy for OA management.

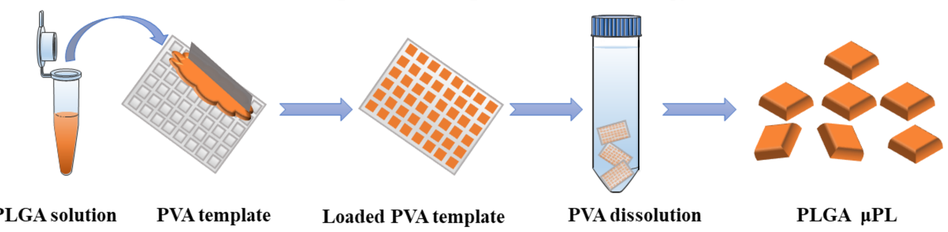
Research goal

Development of an intra-articular delivery system for OA treatment, able to provide a controlled and/or sustained drug release, enabling long- term treatment with a reduced number of injections.

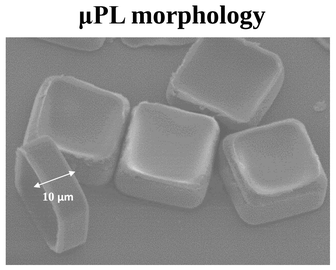
Fabrication and characterization of PLGA µPL

PLGA (poly(D,L-lactide-co-glycolide) microPLates (µPL), fabricated using a top-down approach, have been loaded with different cargo, including the anti-inflammatory molecule dexamethasone (DEX) and matrix metalloproteinase 13 (MMP-13) RNA interference nanoparticles (siMMP13-NPs). Note that MMP13 is upregulated in OA and degrades the key cartilage structural protein type II collagen. Both formulations achieved continuous release over a period of 30 days in biologically relevant, confined volumes [2,3].

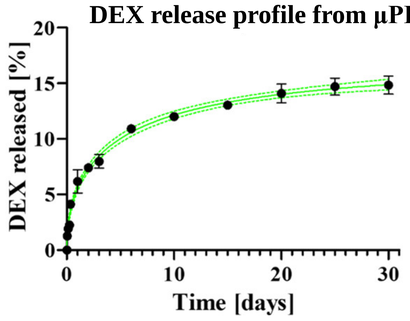
Top-down template-based strategy



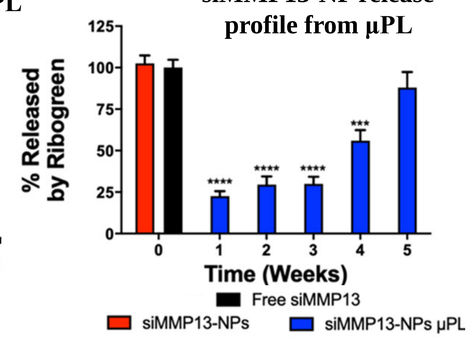
µPL morphology



DEX release profile from µPL



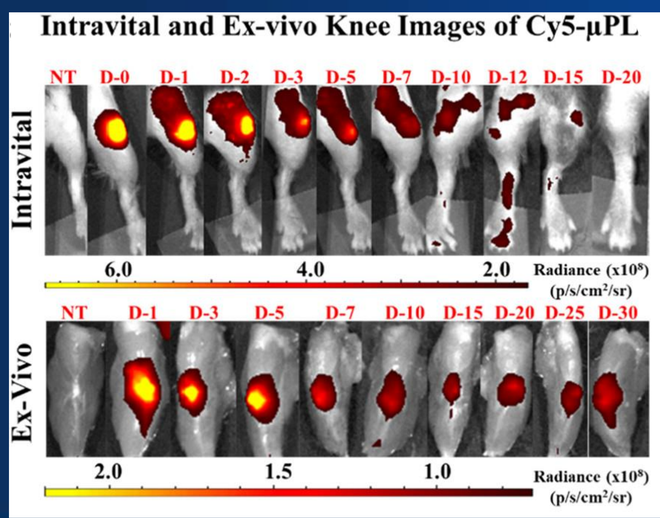
siMMP13-NP release profile from µPL



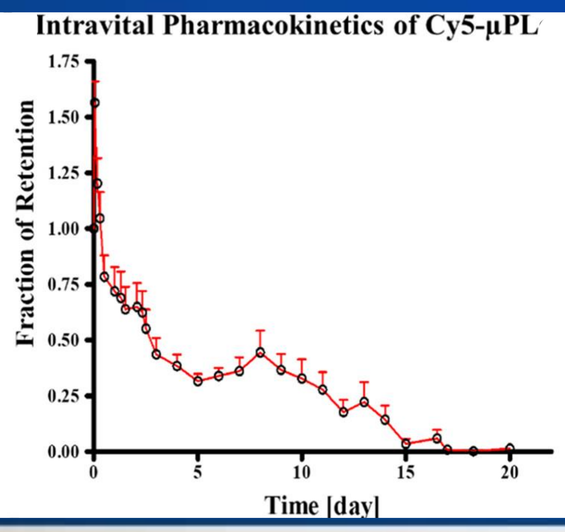
In vivo pharmacokinetic study of Cy5-µPL in a PTOA mouse model

After a single intra-articular injection of Cy5-µPL into a cohort of mice with mechanically induced osteoarthritis (PTOA), a time course of intravital imaging, *ex vivo* imaging, and confocal microscopy analyses were performed: fluorescent µPL were detected in the joint for up to 30 days.

Intravital and Ex-vivo Knee Images of Cy5-µPL



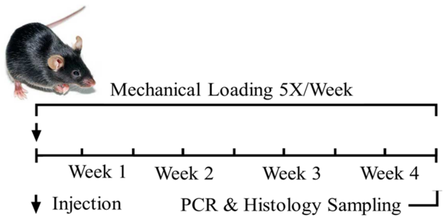
Intravital Pharmacokinetics of Cy5-µPL



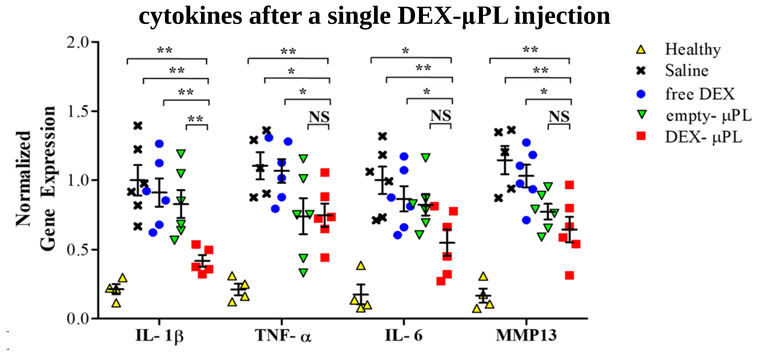
µPL therapeutic efficacy in a PTOA mouse model

A single injection of DEX-µPL decreased the expression of IL-1β, TNF-α, IL-6 and MMP-13 by approximately half compared to free DEX at 4 weeks post-treatment in a PTOA mouse model. In the same animal model treatment with siNP-µPL against MMP13 (siMMP13-µPL) provided potent (65−75%) MMP13 gene expression knockdown and reduced MMP13 protein production in joint tissues through 28 days.

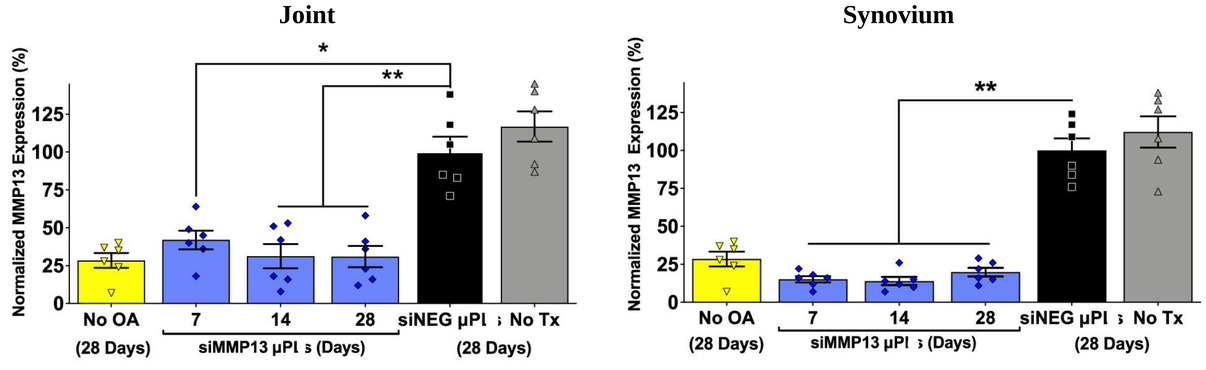
Mechanical loading regimen



In vivo expression of pro-inflammatory cytokines after a single DEX-µPL injection



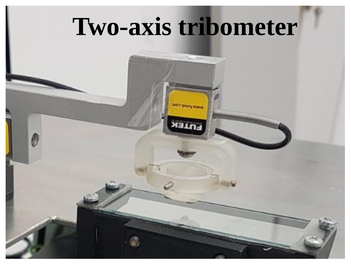
MMP13 gene expression after a single siMMP13-µPL injection

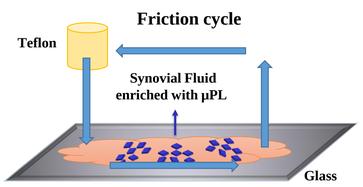


µPL Tribological behavior

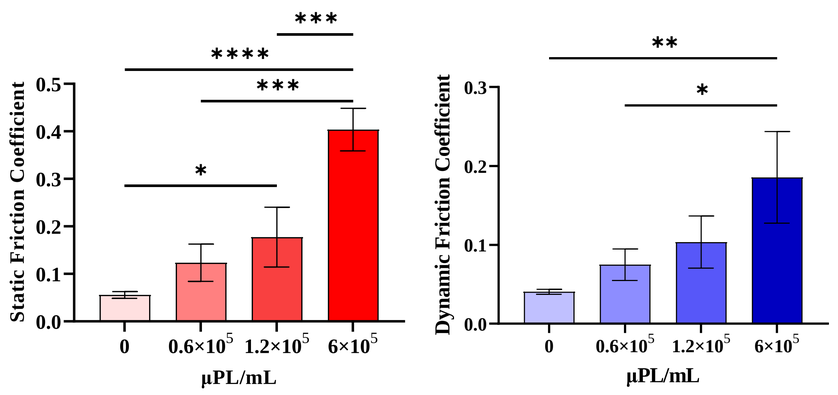
The tribological properties of the µPL were evaluated by using a customized two-axis tribometer designed and constructed at Azrieli College of Engineering Jerusalem. In this device, the micro-particles were suspended within artificial synovial fluid (SF) and squeezed in between two rigid surfaces (a flat Teflon pressed against a flat rigid glass) during sliding. Specifically, the SF was mixed with increasing amounts of particles to obtain different concentration and the friction coefficient was measured on the tribometer. Data showed that the friction coefficients (static and dynamic) increased with the local µPL concentration but, at sufficiently low concentration ($0.6x10^5$ µPL/mL), no statistically significant change in friction was observed. Under these conditions, µPL could protect the surface of the articular cartilage acting as cushions.

Two-axis tribometer





Friction coefficient for different µPL concentrations



Conclusion

In conclusion, shaped-defined Mpl, fabricated using a top-down approach, can be used as injectable drug delivery systems for OA treatment, able to act on both biomechanical and pharmacological disease aspects.

References

1. Martel-Pelletier et al., Osteoarthritis. *Nature Reviews Disease Primers*, 2.1 (2016): p. 16072.2
2. Di Francesco, Martina, et al. "Shape-defined microplates for the sustained intra-articular release of dexamethasone in the management of overload-induced osteoarthritis." *ACS Applied Materials & Interfaces* 13.27 (2021): 31379-31392.
3. Bedingfield, Sean K., et al. "Top-down fabricated microplates for prolonged, intra-articular matrix metalloproteinase 13 siRNA nanocarrier delivery to reduce post-traumatic osteoarthritis." *ACS nano* 15.9 (2021): 14475-14491