Osteoarthritis and Cartilage 25 (2017) 1143–1149

Osteoarthritis and Cartilage

Reinforcement of articular cartilage with a tissue-interpenetrating polymer network reduces friction and modulates interstitial fluid load support

B.G. Cooper † ‡, T.B. Lawson ‡ §, B.D. Snyder ‡ || ¶ # **, M.W. Grinstaff † || ¶ *

† Department of Chemistry, Boston University, Boston, MA, USA
‡ Center for Advanced Orthopaedic Studies, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, MA, USA
§ Department of Mechanical Engineering, Boston University, Boston, MA, USA
|| Department of Biomedical Engineering, Boston University, Boston, MA, USA
¶ Department of Medicine, Boston University, Boston, MA, USA
# Department of Orthopaedic Surgery, Boston Children's Hospital, Boston, MA, USA

ARTICLE INFO

Article history:
Received 24 November 2016
Accepted 1 March 2017

Keywords:
Articular cartilage
Friction
Biolubrication
Biomaterials
Tissue reinforcement

SUMMARY

Objective: Osteoarthritis (OA) is associated with increased articular cartilage hydraulic permeability and decreased maintenance of high interstitial fluid load support (IFLS) during articulation, resulting in increased friction on the cartilage solid matrix. This study assesses frictional response following *in situ* synthesis of an interpenetrating polymer network (IPN) designed to mimic glycosaminoglycans (GAGs) depleted during OA.

Methods: Cylindrical osteochondral explants containing various interpenetrating polymer concentrations were subjected to a torsional friction test under unconfined creep compression. Time-varying coefficient of friction, compressive engineering strain, and normalized strain values ($\varepsilon/\varepsilon_{eq}$) were calculated and analyzed.

Results: The polymer network reduced friction coefficient over the duration of the friction test, with statistically significantly reduced friction coefficients (95% confidence interval 14–34% reduced) at equilibrium compressive strain upon completion of the test ($P = 0.015$). A positive trend was observed relating polymer network concentration with magnitude of friction reduction compared to non-treated tissue.

Conclusion: The cartilage-interpenetrating polymer treatment improves lubrication by augmenting the biphasic tissue's interstitial fluid phase, and additionally improves the friction dissipation of the tissue's solid matrix. This technique demonstrates potential as a therapy to augment tribological function of articular cartilage.

Introduction

Articular cartilage is the smooth, hydrated hyaline cartilage that supports compressive and shear forces applied to diarthrodial joint surfaces, providing low coefficients of friction (COF) and resisting material failure for many decades of use in healthy individuals. Several general models for articular cartilage are described, including a biphasic framework (with tissue comprised of a *solid matrix* and an *interstitial fluid phase)* and a triphasic framework (incorporating an *ionic phase* of fixed and mobile charges). The *solid matrix* is comprised of an extracellular biopolymer matrix of predominantly type II collagen, along with hyaluronan, proteoglycan complexes (glycosaminoglycan (GAG) chains attached to a peptide backbone), and chondrocytes. Porosity and hence permeability of the solid matrix are key defining characteristics of this phase[1], as are various other elements of structure including compressibility, anisotropy due to collagen fibril orientation[2], and tensile strength of the matrix[3]. An aqueous solution of proteins, hyaluronan, and

* Address correspondence and reprint requests to: M. W. Grinstaff, Department of Chemistry, Boston University, Boston, MA 02215, USA.
** Address correspondence and reprint requests to: B. D. Snyder, Department of Orthopaedic Surgery, Boston Children's Hospital, Boston, MA, USA.
E-mail addresses: bgcooper@bu.edu (B.G. Cooper), tlawson@bu.edu (T.B. Lawson), bsnyder@bidmc.harvard.edu, Brian.Snyder@childrens.harvard.edu (B.D. Snyder), mgrin@bu.edu (M.W. Grinstaff).

http://dx.doi.org/10.1016/j.joca.2017.03.001
1063-4584/

other solutes, known as synovial fluid, constitutes the tissue's *interstitial fluid phase*. As typically 65–85 w/w% of cartilage is water, the fluid phase plays an important physiologic role with respect to diffusion-based transport, and it also contributes significantly to tissue compressive properties. As cartilage is compressed, water (being incompressible) is expelled from the tissue; the rate of this expulsion is limited largely by the solid matrix's permeability, as well as by the GAGs which attract and retard the outward flow of water molecules. Thus, when the tissue is initially loaded in compression, nearly 100% of the load is supported by the fluid phase, and as water molecules flow out of the matrix as the tissue creeps to equilibrium under constant compressive load, the interstitial fluid load support (IFLS) decreases from near 100% to 0%[4]. Over this transition, the proportion of total load supported by the tissue's solid phase likewise increases from near 0% to 100% upon compressive equilibration. In a configuration of migrating rather than stationary loading contact, such as in various reciprocating friction testing experiments, IFLS may be maintained at a high value near 100% pending certain conditions, e.g., a substantially high ratio of time unloaded to time loaded, or a substantially low compressive force[5]. It should be noted that not all water molecules are mobile, and only "free" water in the fluid phase is able to exude from the tissue upon compression while "bound" water is electrostatically immobilized by fixed charges within the tissue[6] (hence "bound" water is more accurately classified as part of the solid phase than the fluid phase). The *ionic phase* of the tissue plays a role in triphasic theory of cartilage material and mechanical properties; fixed negative charges within the solid matrix, arising from sulfate and carboxylate functional groups of GAGs, are balanced by mobile sodium, potassium, and calcium cations through Donnan equilibrium, and exhibit an electrostatic resistance to being compressed as the anionic groups repel one another upon loading[7]. Furthermore, the ionic phase gives rise to cartilage's dependence of compressive properties on salt concentration, with increasing presence of salts causing a charge screening effect and thereby decreasing the fixed negative charges' (1) electrostatic resistance to compression and (2) attraction to mobile and non-mobile water molecules[8].

As human knee and hip articular cartilage experiences about 1 million articulation cycles in a typical year, its lubrication is essential in maintaining low friction and wear. Owing to the biphasic nature of cartilage as well as its viscoelasticity and variability in loading conditions under different physiologic circumstances (e.g., alterations in synovial fluid viscosity with aging or disease, and various loading magnitudes caused by body weight and carrying load), several lubrication mechanisms have been proposed over the last half century[4,9–12]. The majority of investigations of cartilage COF have been conducted at equilibrium (either in creep or stress-relaxation), when the IFLS is ~0%[12–14]. Under such conditions when nearly 100% of the load is supported by the tissue's solid phase, lubrication of cartilage is approximated as occurring analogous to that of a monophasic elastic solid, with classical modes of *boundary* and elastohydrodynamic (*fluid film*) lubrication being operative. An alternative mechanism, known as "*boosted*" lubrication, occurs as water molecules in the interposed fluid film between apposing cartilage surfaces are driven into the tissue matrix; this causes the local concentration of lubricating macromolecules in synovial fluid (hyaluronic acid, lubricin, and phospholipids) to be increased or "boosted," forming a lubricious gel at the tissue interface. In contrast to a 0% IFLS scenario, the presence of substantial IFLS greatly reduces the tissue's friction and causes alternate stress dissipation phenomena. Upon initial compressive loading concurrent with sliding, negligible solid–solid frictional forces exist between the apposing cartilage surfaces, since the tissue's interstitial fluid phase supports nearly 100% of the applied load. Under these ~100% IFLS conditions, the expulsion of interstitial fluid from the tissue's bulk and into the interfacial region between the tissues, known as "*weeping*," allows the two surfaces to be kept apart via a self-pressurized interfacial fluid in a hydrostatic mode of lubrication[10,15,16]. In these types of studies, the COF (as well as the creep deformation) varies directly with the IFLS[4,17,18], and the ability to maintain near 100% IFLS has been postulated as a characteristic critical for affording low COF and high wear resistance over a synovial joint's many decades of use.

A primary disease of articular cartilage occurring with advanced age is osteoarthritis (OA), associated with degradation of cartilage material properties and causing wear. Consequently, significant research activities are directed at treating, augmenting, or repairing degraded cartilage[19–23] including, for example, polymer scaffolds for filling tissue defects[24–28], lubricants for improving tribological properties[29,30], stimulating growth factors for promoting healing[31], cell transplantation[32,33], and gene therapy for restoring biological activity[34]. An early hallmark of OA is the loss of GAGs with a resultant decrease in the dynamic and equilibrium compressive moduli of the cartilage by allowing increased rate of water exudation and decreased quantity of GAG-bound water, respectively. Furthermore, the hydraulic permeability of cartilage is increased, causing IFLS to decrease at a faster rate upon loading, exacerbating cartilage degeneration by increasing the occurrence of solid–solid contact-derived friction[35].

Currently, there are no therapies that mitigate the loss of GAGs or effectively replace lost GAGs. We hypothesize that a treatment which restores high IFLS may improve cartilage function by reducing solid–solid interfacial friction. We recently reported a new cartilage-reinforcing technique—administration of a GAG-inspired zwitterionic polymer 2-methacryloyloxyethyl phosphorylcholine (pMPC) that reconstitutes cartilage matrix hydrophilicity[36]. The treatment involves forming, through *in-situ* photopolymerization, a semi-synthetic interpenetrating polymer network (IPN) entangled with native collagen fibrils [Fig. 1(a)] that increases the compressive stiffness and wear resistance of treated cartilage during accelerated wear testing against stainless steel when IFLS began high and was allowed to subside. The present study investigates the frictional response of IPN-treated cartilage sliding against IPN-treated cartilage and the effects of the interpenetrating polymer on the relationship between normalized strain values ($\varepsilon/\varepsilon_{eq}$) and COF.

Methods

Sample preparation and IPN treatment

Nine pairs of osteochondral cylindrical plugs (7 mm diameter, cartilage thickness ranging approximately 1–1.5 mm) were cored from the stifle joints of skeletally mature cows in a procedure similar to those reported previously[12,37] using a diamond-tipped coring bit (Starlite Industries, Bryn Mawr, PA), irrigated with 0.9% saline at room temperature. Throughout experimentation, plugs were stored at 4°C in 400 mOsm sodium chloride solution containing protease inhibitor benzamidine hydrochloride (5 mM), GIBCO Antibiotic/Antimycotic (Invitrogen, Grand Island, NY), and calcium ion chelating agent ethylenediamine tetraacetic acid (5 mM). The nine plug pairs were randomly sorted into three groups of $N = 3$ pairs. Two groups of $N = 3$ osteochondral plugs were incubated in the dark for 24 h at 25°C in 400 mOsm saline containing 2-methacryloyloxyethyl phosphorylcholine (either 20 or 60 w/v%), ethylene glycol dimethacrylate (1% mol/mol 2-methacryloyloxyethyl phosphorylcholine), eosin Y (0.1 mM), triethanolamine (115 mM), and N-vinylpyrrolidone (94 mM). Plugs were suspended upside down with articular surface exposed to solution to allow diffusion of solutes. Plugs were removed from