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Lamellar slippage of bilayers—A hypothesis on low friction of natural joints

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The cartilage's amphoteric surface behavior is a physical phenomenon in biological lubrication. However, there is a lack of knowledge on amphoteric phospholipids bilayers and in overcoming friction in cartilage joints. In this paper, friction experiments were conducted, and the cartilage's surface was characterized using *p*H and wettability, while the interfacial energy and coefficients were determined. The lamellar slippage of bilayers and a short-range repulsion between the interfaces of negatively charged ($-PO_4^-$) cartilage surfaces resulted in low frictional properties of the joint. © *2014 American Vacuum Society*. [http://dx.doi.org/10.1116/1.4902805]

I. INTRODUCTION

An important difference between biological and man-made lubrication systems is that in the former, the lubricant is chemically attached to the surface of, for example, a cartilage joint.[1,2] The amphoteric[3,4] phospholipids (PLs) are the main solid-phase components on the surface of an articular cartilage (AC), which are responsible for the biological lubrication mechanism.[3,5] It has been well established that the PL bilayers mechanism, which essentially consist of a surface amorphous layer (SAL) surrounded by a 0.155 M electrolyte synovial fluid (SF) of *p*H ∼ 7.4 with high-molecular-weight charged biomacromolecules, supports low friction.[6–8]

The highly hydrated three-dimensional lamellar mechanism is electrically charged and is able to resist compressive forces during joint loading.[9,10] The negatively charged articular surface interacts electrostatically with the macromolecules of SF hyaluronate, lipids, and the glycoprotein lubricin.[11] Without this electrostatic charge, frictional forces can either deform or deplete the surface of the joint structure. A lamellar PL structure consisting of 5–7 bilayers was experimentally documented by electron microscopy and biochemical procedures.[12–18] It was observed that as friction increased, the damaged cartilage was prone to degenerate by losing its PL bilayers. The previous authors suggested that the PL bilayers on the surface and the PL lamellar aggregates in SF play a decisive role in the low friction of cartilage. Owing to the loss of the PL bilayers, the stiffness of cartilage increased[19,20] and in turn the friction coefficient was affected.[21,22]

The chemical and physical nature of the biological surfaces is seen in an entirely different light to that of engineering surfaces immersed in water.[4] The lubrication mechanisms in an animal's body, where the tissues slide over each other, the surfaces coated with PL bilayers and a lamellar structure negatively charged on articular surface with synovial fluid, have been referred to as a "lamellar-repulsive" mechanism.[27,39] The role played by hydration or structural force is believed to arise from a strongly bound and oriented first layer of the water molecules on charged surfaces.[23] A distinct polar charge distribution of the water molecule allows each molecule to participate in strong polar (electrostatic charge—dipole or hydrogen—bonding) interaction. The short-range repulsion often observed between biological surfaces is not due to layered structure of water but due to entropic repulsion.[23]

In this paper, the chemical and physical properties of the bovine cartilage surfaces, the interfacial energy of the PL bilayer, and the friction coefficient were found to respond in an amphoteric manner as the *p*H varied. The wettability of a normal articular surface was compared with its depletion of PL bilayers. Also, the cartilage's wettability effect on friction coefficient to support the lamellar-repulsive mechanism of lubrication was investigated.

II. MATERIALS AND METHODS

A. Materials

In the experiment, we used phosphatidylethanolamine (PE) as a phospholipid substance (estimated to be 99%

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pure), purchased from Fluka AG, Switzerland. To model the phospholipid membrane, we prepared a solution containing n-decane and 20 mg/ml phosphatidylethanolamine. The articular cartilage specimens were collected from bovine knees aged 15–20 months. Osteochondral plugs, 5 and 10 mm in diameter, were harvested from lateral and medial femoral condyles using a circular stainless steel cutter. The cartilage disks were cut into 3 mm plugs with underlying bone. Two types of samples were tested: untreated bovine cartilage and bovine cartilage treated with a Folch reagent[24] (a 2:1 v/v mixture of chloroform and methanol), and a lipid-rinsing solution to remove the lipids from the surface of the cartilage. After preparation, the specimens were stored at 253 K in saline of 0.155 M NaCl (pH = 6.9), and fully defrosted prior to testing. The cartilage disks were then glued to the disk and pin stainless steel surfaces, and friction tests were conducted in the saline. Bovine synovial fluid was collected from bovine ankle joints within 32 h of commercial slaughter. The fluid was filtered to remove cartilage debris and then stored frozen at 253 K.

The friction measurements of the cartilage joint versus pH (2.5–9.5) were carried out using a Britton–Robinson[25] universal buffer solution. It consisted of a mixture of 0.04 M H_3BO_3, 0.04 M H_3PO_4 and 0.04 M CH_3COOH that has been titrated to the desired pH with 0.2 M NaOH.

To obtain the required pH of the electrolyte solution, a Radiometer pH-meter with an electrode (Schott-BlueLine 16 pH type) was used in the experiment. This instrument was calibrated according to the recommendations made by IUPAC.[25]

B. Interfacial energy measurements

The interfacial energy (γ) of the phosphatidylethanolamine bilayer was determined by measuring the curvature radius, r, of the convex surface formed by applying a pressure difference, Δp, on its sides. The method used was based on Young's and Laplace's (Y–L) equation[4]

$$2\gamma = r\Delta p. \tag{1}$$

Gamma value obtained from (Y–L) equation was applied to Eq. (2), K_a and K_b was determined graphically, by using the least squares method. The dependence of interfacial energy on the pH using a Britton–Robinson universal buffer solution has the form[17,27]

$$\gamma = \gamma_{max} + 2sRT\ln\left(\sqrt{\frac{K_a}{K_b}} + 1\right) - sRT\ln\left[\left(\frac{K_a}{a_{H^+}} + 1\right)\left(\frac{a_{H^+}}{K_b} + 1\right)\right], \tag{2}$$

where K_a and K_b are the acid and base equilibrium constants, respectively, s (mol m^{-2}) is the surface concentration of phospholipids, $s = \frac{1}{N_A \cdot A}$; where A is surface area occupied by the phospholipid molecules, and N_A is the Avogadro constant, a_{H^+} is the hydrogen ion (H^+) concentration, R is the gas constant, T is temperature, and γ_{max} is the maximum interfacial energy of the lipid membrane.

The apparatus and the microelectrophoretic method used are described in Refs. 26 and 27. The value of (γ) was measured in 8–12 replicates with about seven instrumental readings of the lipid spherical cap. The results of interfacial energy (γ) as a function of pH are given in Fig. 1.

C. Delipidization procedure

In the delipidization procedure, a Folch reagent (2:1 v/v mixture of chloroform and methanol) was used to gradually remove the PL bilayers from the cartilage surface. The samples were immersed in the reagent mixture for 9, 13, and 17 min, at the same meniscus. After extraction, the sample was placed in saline solution for 1 h to remove the residue of the solvent and promote rehydration. These samples were used for the surface wettability and friction measurements. Other authors used isopropanol[28] and an enzymatic procedure with phospholipase A.[29,30] The delipidization procedure removed most of the PL although some amount of a hydrophobic proteolipid remained as a minor component.[9,30]

D. Contact angle measurements

The contact angle was measured using a KSV CAM100 computerized tensiometer. A drop of the 0.155 M saline solution was deposited on the air-dry cartilage surface. The contact angle measurements of the normal (not depleted), partial, and completely depleted cartilage samples were carried out under dry-air atmosphere at 295 ± 2 K and a relative

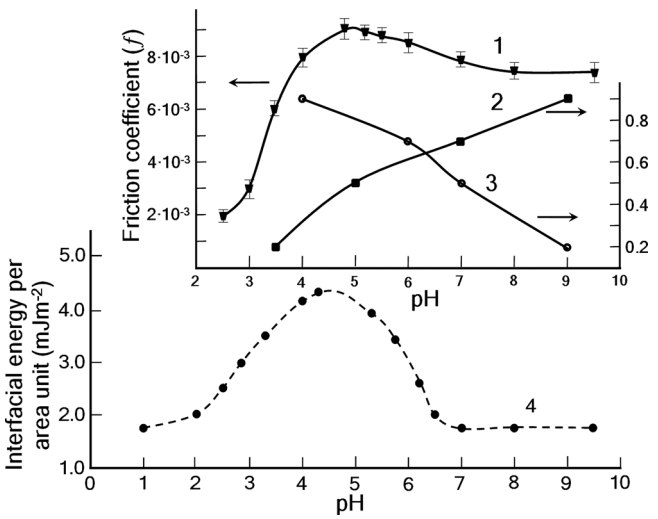

FIG. 1. Influence of the buffer solution pH on the friction coefficient of cartilage. The friction changes as the pH is raised toward to curve's maximum as can be expressed by reactions progressing on the cartilage surface: (curve 1) (-NH_3^+ → -NH_2), and after isoelectric point, IEP (-PO_4H → -PO_4^-) [this work]. To support our experiment, multilayers of nonamphoteric [poly(L-lysine)/hyaluronic acid] (Ref. 32); (curve 2) [L-lysine (-NH_3^+ → -NH_2)]; (curve 3) hyaluronic acid (-COOH → -COO^-) are shown. Additional support is provided by amphoteric character of phospholipidic cartilage and by interfacial energy of phospholipid bilayer formed by PE vs pH [this work], (curve 4) (-NH_3^+ → -NH_2, after IEP (-PO_4H → -PO_4^-)]. Curve 1 friction coefficient (%) standard deviation (SD) 10–17.

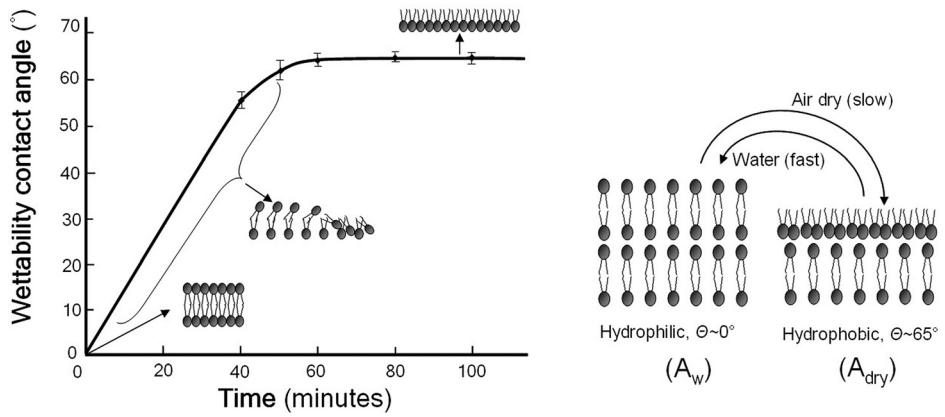

FIG. 2. Contact angle measured as a function of air-drying time of the articular surface of bovine patella with partially depleted surface phospholipid bilayer (contact angle of ~65°) compared to normal articular surface (contact angle ~ 100°). The transition from (HL → HB) is more likely due to the phospholipid translocation, or flip–flop, and has to occur of the cartilage surface. Superficial phospholipid bilayer of articular cartilage in water (A_w) and air-dry (A_{dry}) conditions. A change in surface energy leads to conformational changes in the surface of bovine patella from bilayer (super hydrophilic ~ 0° contact angle) to monolayer (hydrophobic) ~ 65°. Contact angle (%) SD 9–15.

humidity, HR ~ 50% ± 5%, between 40 and 100 min of the sample drying time (see Fig. 2 and Table I).[31] The contact angle test, on the normal, partial, and completely delipidized cartilage samples, was repeated at least five times.

E. Friction coefficient measurements in saline solution of 0.155 M NaCl (*p*H = 6.9)

The measurements were performed using a sliding pin-on-disk tribotester T-11 manufactured by the National Institute for Sustainable Technologies Research, Radom, Poland. The tests were conducted at room temperature, at a speed of 1 mm/s during 15 min, and under a load of 15 N (1.2 MPa), which correspond to the physiological lubrication condition.[27] Before each test, the cartilage samples were left in saline solution for 1 h. The results of the friction coefficient (*f*), as a function of wettability, are given in Table I and in Fig. 3 for the normal and delipidized cartilage/cartilage pairs, respectively. In each of the friction pairs, an increase in the friction coefficient and actual contact area (the parts rubbing) were observed with time. A total number of five tests were conducted, using fresh samples for each experimental setup with at least four repetitions per specimen pair, from which the mean and standard deviation were calculated.

1. Friction test in universal buffer solutions (pH 2.5–9.5)

Prior to the friction tests, the lubricants were prepared using the Britton–Robinson universal buffer solution[25] and its *p*H values were measured. The *p*H value depended on the volume quantity of the buffer solutions. The friction coefficients measurements of cartilage/cartilage tribopair were carried out within the *p*H values ranging between 2.5 and 9.5. The testing samples were equilibrated with each buffer under a load for 5 min, and the results of (*f*) as a function of *p*H are given in Fig. 1. A total number of five tests were conducted using fresh samples for each experimental setup with at least four repetitions per specimen pair, from which the mean and standard deviation were calculated.

III. RESULTS AND DISCUSSION

A. Solution *p*H versus friction of cartilage surface

The frictional forces, acting on the surface of the cartilage, are found to be sensitive to the *p*H values (ranging from 2.5 to 9.5) of the buffer solutions (lubricants) inserted between cartilage/cartilage tribopair surfaces. The experimental results in Fig. 1 display the relationship between friction coefficients and the *p*H solutions. Also shown in this

TABLE I. Friction coefficient (*f*) for the (cartilage/cartilage) tribopair during the run in saline solution of 0.155 M NaCl (*p*H 6.9) and wettability of the normal, partial, and completely depleted bovine cartilage surface measured for air-dry surface at ambient temperature and a relative humidity, HR ~ 50%.

Friction time run	Normal AC, and (*f*)[a]	Partially depleted AC, 9 min, and (*f*)[a]	Partially depleted AC, 13 min, and (*f*)[a]	Completely depleted AC, 17 min, and (*f*)[a]
−1 min	0.005	0.012	0.015	0.021
−15 min	0.007	0.015	0.018	0.023
5% SF[b], 5 min	0.004	—	—	—
15% SF[b], 5 min	0.003	—	—	—
Wettability (°)	100	65.0	54.0	36.7

[a](*f*) friction coefficient, (%) SD 10 to 17.
[b]Bovine synovial fluid, SF 5% and 15% diluted with 0.155 M NaCl solution.

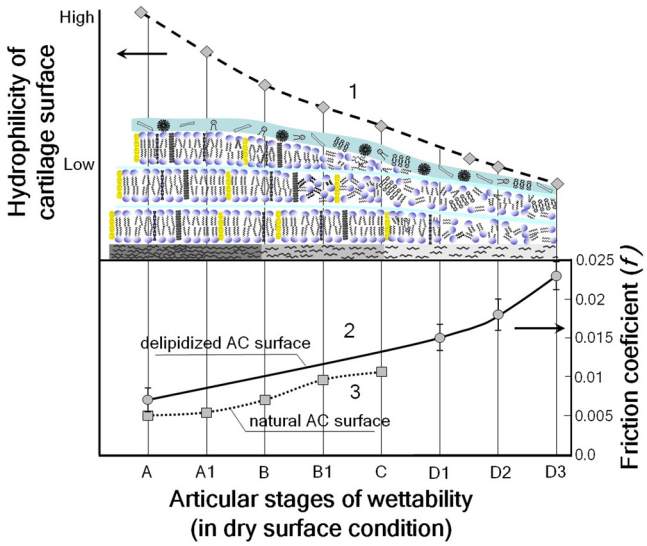

FIG. 3. Hydrophilicity of AC surface/or (friction coefficient) vs the stages of wettability of the AC during the active lifespan of animals: (A) human and bovine cartilage surface 103° (Ref. 25) and 100° [this work]; (A1) human knee 79.7° (Ref. 30); (B) unhealthy cartilage surface 65° (Refs. 27 and 30); (B1) human knee 63° (Ref. 30); (C) naturally degenerated hip 56.3° (Ref. 5 and 35); artificially partially depleted cartilage surface (D1) 65° [this work], partially depleted cartilage surface (D2) 54° [this work]; completely depleted cartilage surface (D3) 36.7° [this work]. Curve (1) changes of hydrophilicity of AC surface from stage A to D; curve (2) changes of friction coefficient of artificially depleted the bovine cartilage surface [this work]; curve (3) changes of friction coefficient of natural joints (Ref. 27). Note typical inter-lamellar aqueous spacing of 4.5 nm between bilayers (Ref. 3). High contact angle (in dry surface condition) corresponds to high hydrophilicity (when surface is wet), while low contact angle (for dry cartilage) corresponds to low hydrophilicity (when surface is wet). By illustration of the three bilayers, we outline a mechanism of lamellar frictionless lubrication wherein the low charged density the bilayer surface (Ref. 42) adsorbs biomacromolecules and lamellar aggregates (Ref. 9). (*f*) Friction coefficient (%) SD 10–17.

figure is a separate graph of the characteristic isoelectric point, IEP (curve 1). For comparative purposes, we used the interfacial energies of PL bilayer (curve 4) and multilayer of nonamphoteric [poly (L-lysine)/hyaluronic acid][32] [L-lysine ($-NH_3^+ \rightarrow -NH_2$)] (curve 2), and hyaluronic acid ($-COOH \rightarrow -COO^-$) (curve 3) to support the charged cartilage surface. As the *p*H is varied, the curves, friction coefficient (curve 1), and the interfacial energy (curve 4) show a remarkably similar amphoteric behavior. Below the isoelectric point (IEP) of the cartilage surface and PL bilayer, the surface is positively charged ($-NH_3^+$), with the gradual change of friction and interfacial energy, as the *p*H shifts toward the IEP. After passing through the IEP, the surface charge gradually changes from being positive ($-NH_3^+$) to negative ($-PO_4^-$), and the surface friction turns from the attractive to a repulsive state. The nonamphoteric effect (curves 2 and 3) confirms that the positively ($-NH_3^+$) and negatively ($-COO^-$) charged surfaces under friction, respond linearly to the *p*H range investigated. The relationship between surface friction and the *p*H solution was previously studied using amphoteric engineering material on a SiO_2 surface and similar results were also observed.[33]

The PE belongs to an amphoteric polyelectrolyte, amine ($-NH_2$) and phosphate ($-PO_4H$) functional group. The observed maximum on both curves was at the isoelectric point (*p*H 4.3) for PLs (PE) (pure phospholipid) and *p*H 4.8 for cartilage (mix of phospholipids and other biomolecules). This slow decrease in (*f*) after IEP suggests the existence of other anionic macromolecules beside ($-PO_4^-$). The maximum interfacial energy (γ_{max}) was found to be at 4.08 mJ m^{-2} while correspondingly on abscissa, the *p*H was noted to be 4.2. On the upper graph of Fig. 1, the maximum friction coefficient of 0.09 occurred when the *p*H was 4.8. When the *p*H ~ 2, amino groups of PLs occurs in the protonated form ($-NH_3^+$), while $-PO_4H$ is in its molecular form. As the *p*H of the solution is raised, the amino groups begin to lose their proton ($-NH_3^+ \rightarrow -NH_2$), leading to an increase in the interfacial energy toward a maximum value at the IEP, while the $-PO_4H$ group also tends to gradually lose its proton ($-PO_4H \rightarrow -PO_4^-$). At IEP, both surface constituents would carry no net electric charge (i.e., the negative and positive charges would be equal).[4,41] As the *p*H of the solution is increased, after IEP, the amino group would gradually lose its charge, while the $-PO_4H$ group loses its proton ($-PO_4H \rightarrow -PO_4^-$), leading to a negatively charged surface with decreased interfacial energy and decreased friction coefficient. The polyelectrolytes of nonamphoteric multilayers of PLL/HA [poly(L-lysine)/hyaluronic acid] illustrate the variation of the (*f*), which was found to be linear over the whole solution *p*H range of 3.5–9.5.

B. Cartilage surface wettability

The contact angle parameter is reflected in the charge density of the functional group on the surface, especially in the number of PLs bilayers on the cartilage surface. High contact angle (in dry surface condition) corresponds to high hydrophilicity (when surface is wet), while low contact angle (for dry cartilage) corresponds to low hydrophilicity (when surface is wet). Figure 2 shows a plot of the contact angle versus time on a partially depleted cartilage sample.

We measured the contact angle of sessile saline droplets on the surfaces of normal, partial, and totally depleted cartilage samples after 100 min of drying at room temperature. The biological tissue of the cartilage in its natural condition is superhydrophilic with a contact angle zero. The air-drying time is a process of transformation from the hydrophilic to the hydrophobic (HL→ HB) condition overturning phospholipid molecules (flip–flop), which is described by the surface reorganization of PL molecules of the bilayer into monolayer.[27,34] Here, the air-dry tissue loses surface water and electrostatic repulsive forces and transforms from a hydrophilic into a hydrophobic surface.

C. Cartilage surface wettability and friction

The bovine sample (cartilage/cartilage) pairs are used to study the effects of friction on the surfaces of articular joints. Various wettability states and their corresponding relationships with different levels of adsorption and hydration were considered. This includes the surface frictional and wettability properties of the *p*H dependent acid–base dissociation of

amphoteric PLs, that is, weak multilayer polyelectrolytes. When comparing the depleted cartilage with untreated normal samples, the results support observations made by Hills and other authors,[27,35–38,41] that both the friction and wettability are important factors in the assessment of biological surfaces.

In Fig. 3, the friction coefficients measured for a partial and completely depleted cartilage samples (curve 2) are compared with the results obtained for natural joints (curve 3) with healthy and naturally degenerated articular surfaces.[5,35,51,52] We interpret the increased friction coefficient values due to the number of bilayers available in SAL.[27,30,36] Both the friction and wettability show very similar behavior as the SAL thickness is varied.[27,39,49] The cartilage implication from osteoarthritis disease by a gradual losing of the surface amorphous layer has shown an increased friction coefficient.[35,36,38] The SAL, phospholipidic lamellar aggregates and biomacromolecules in SF may contribute to electrostatic repulsion during lubrication. The highly hydrated PL lamellar aggregates are expected to cover cartilage surfaces and support hydrophilic lamellar-repulsive lubrication.[9,24,35,40]

The parameters found to consistently influence friction were wettability, surface energy, *p*H, and effect of diluting synovial fluid. The saline lubricant spiked with 5% and 15% (v/v) of bovine synovial fluid resulted in decrease in friction coefficient from 0.005 to 0.003 (Table I). These observations indicate that the charged SF macromolecules were in contact with the surface of the cartilage and they functioned as a lubricant.[41,42]

1. Short-range hydration repulsion between the interfaces of negatively charged cartilage surfaces

Surfaces of articular cartilage joints coated with PL bilayers and surrounded by synovial fluid inherit charged macromolecules, proteins, and lipids. Low friction coefficient between articular cartilage surfaces in living joints, $f \sim 0.005$ under load 10 MPa, are described in this review as a "lamellar-repulsive" mechanism.[27,39] The major macromolecules lubricin, hyaluronan A^-, proteoglycan (PTG) form complex A^-PTG negatively charged groups (-COO^- and –SO_3^-). The cartilage surfaces experience weak van der Waals attractive forces and much stronger short range repulsive forces due to hydration repulsion.[4,23,43] Hydration repulsion dominates the interaction between charged cartilage surfaces at nanometer separations and ultimately prevents the sticking together of cartilage surfaces, even at high pressures of 100 MPa.[44,45] A layer of hydrated water strongly binds to the negatively charge cartilage surface, and when in contact with synovial fluid components (charged biomacromolecules, PL lamellar aggregates, and liposomes), this reduces the friction between cartilage surfaces.[45–52]

IV. CONCLUSIONS

This study has revived the importance of the amphoteric nature of an articular surface, and the surface amorphous layer, in reducing friction coefficient after the isoelectric point. The cartilage surface was characterized using wettability tests of fresh and depleted samples. Friction tests were conducted on normal, partial, and completely depleted bovine cartilage samples. The gradual removal of phospholipid bilayers was found to influence the friction coefficient. Saline fluid retained its lubricating properties when the friction test was implemented with 5% and 15% (v/v) synovial fluid in 0.155 M NaCl lubricant solution. We demonstrated experimentally showing that the cartilage *p*H sensitivity to friction introduced a novel concept in joint lubrication on charged surfaces. The possible lamellar-repulsive mechanism of lubrication and the influence of *p*H on friction coefficient have been discussed.

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[1]M. Urbakh, J. Klafter, D. Gourdon, and J. Israelachvili, Nature **430**, 525 (2004).
[2]J. Katta, Z. Jin, E. Ingham, and J. Fisher, Med. Eng. Phys. **30**, 1349 (2008).
[3]B. A. Hills, Proc. Inst. Mech. Eng., Part H **214C**, 83 (2000).
[4]J. N. Israelachvili, *Intermolecular and Surface Forces*, 2nd ed. (Academic, New York, 1991).
[5]B. A. Hills and M. K. Monds, Br. J. Rheumatology. **37**, 137 (1998).
[6]M. C. Corneci, F. Dekkiche, A. M. Trunfio-Sfarghiu, M. H. Meurisse, Y. Berthier, and J. P. Rieu, Tribol. Int. **44**, 1959 (2011).
[7]Z. Pawlak, Z. A. Figaszewski, A. Gadomski, W. Urbaniak, and A. Oloyede, Tribol. Int. **43**, 1719 (2010).
[8]Z. Pawlak, W. Urbaniak, and A. Oloyede, "Natural articular joints: Model of a lamellar-roller-bearings lubrication and the nature of the cartilage surface," in *Biomaterials and Medical Tribology: Research and Development*, Mechanical Engineering Series, edited by J. P. Davim (Woodhead Publishing Reviews, Cambridge, Philadelphia, New Delhi, 2013), Vol. 65, Chap. 6, p. 253.
[9]I. M. Schwarz and B. A. Hills, Br. J. Rheumatol. **35**, 821 (1996).
[10]B. A. Hills, J. Appl. Physiol. **73**, 1034 (1992).
[11]C. R. Flannery, C. E. Hughes, B. L. Schumacher, D. Tudor, M. B. Aydelotte, K. E. Kuettner, and B. Caterson, Biochem. Biophys. Res. Commun. **254**, 535 (1999).
[12]B. A. Hills, Intern. Med. J. **32**, 242 (2002).
[13]T. B. Kirk, A. S. Wilson, and G. W. Stachowiak, Orthop. Rheumatol. **6**, 21 (1993).
[14]D. Guerra, L. Frizziero, M. Losi, B. Bacchelli, G. Mazzadri, and I. Pasquali-Ronchetti, J. Submicrosc. Cytol. Pathol. **28**, 385 (1996).
[15]P. F. Williams, G. L. Powell, and M. LaBerge, Proc. Inst. Mech. Eng., Part H **207**, 59 (1993).
[16]J. S. Jurvelin, D. J. Müller, M. Wong, D. Studer, A. Engel, and E. B. Hunziker, J. Struct. Biol. **117**, 45 (1996).
[17]S. Kobayashi, S. Yonekubo, and Y. Kurogouchi, J. Anat. **187**, 429 (1996).
[18]P. Kumar, M. Oka, J. Toguchida, M. Kobayashi, E. Uchida, T. Nakamura, and K. Tanaka, J. Anat.**199**, 241 (2001).
[19]S. M. T. Chan, C. P. Neu, G. DuRaine, K. Komvopoulos, and A. H. Reddi, Osteoarthritis Cartilage **18**, 956 (2010).
[20]A. Oloyede, P. Gudimetla, R. Crawford, and B. A. Hills, Connect. Tissue Res. **45**, 86 (2004).
[21]D. Dean, L. Han, A. J. Grodzinsky, and C. Ortiz, J. Biomech. **39**, 2555 (2006).
[22]S. M. Chan, C. P. Neu, G. DuRaine, K. Komvopoulos, and A. H. Reddi, J. Biomech. **45**, 2426 (2012).
[23]J. Israelachvili and H. Wennerstrom, Nature **379**, 219 (1996).
[24]J. Folch, M. Lees, and G. H. Sloane-Stanley, J. Biol. Chem. **226**, 497 (1957).
[25]H. T. K. Britton and R. A. Robinson, J. Chem. Soc. **1931**, 1456.

[26]A. D. Petelska and Z. A. Figaszewski, Bioelectrochem. Bioenerg. **46**, 199 (1998).
[27]Z. Pawlak, W. Urbaniak, A. Gadomski, K. Q. Yusuf, I. O. Afara, and A. Oloyede, Cell Biochem. Biophys. **65**, 335 (2013).
[28]A. Oloyede, P. Gudimetla, R. Crawford, and B. A. Hills, Clin. Biomech. **19**, 534 (2004).
[29]K. Bagińska, J. Makowska, F. Kasprzykowski, W. Wiczk, and L. Chmurzyński, J. Pept. Sci. **14**, 283 (2008).
[30]B. A. Hills and M. K. Monds, Br. J. Rheumatol. **37**, 143 (1998).
[31]J. Chappuis, A. Sherman, and A. W. Neumann, Ann. Biomed. Eng. **11**, 435 (1983).
[32]S. E. Burke and C. J. Barrett, Biomacromolecules **4**, 1773 (2003).
[33]A. Marti, G. Hahner, and D. Spencer, Langmuir **11**, 4632 (1995).
[34]D. Leckband, Y. L. Chen, J. Israelachvili, H. H. Wickman, M. Fletcher, and R. Zimmerman, Biotechnol. Bioeng. **42**, 167 (1993).
[35]B. A. Hills, J. Rheumatol. **16**, 82 (1989).
[36]G. C. Ballantine and G. W. Stachowiak, Wear **253**, 385 (2002).
[37]H. Higaki, T. Murakami, Y. Nakanishi, H. Miura, T. Mawatari, and Y. Iwamoto, Proc. Inst. Mech. Eng., Part H **212**, 337 (1998).
[38]H. E. Ozturek, K. K. Stoffel, C. F. Jones, and G. W. Stachowiak, Tribol. Lett. **16**, 283 (2004).
[39]A. M. Trunfio-Sfarghiu, Y. Berthier, M. H. Meurisse, and J. P. Rieu, Langmuir **24**, 8765 (2008).
[40]M. Daniel, "Role of surface-active lipids in cartilage lubrication," in *Advances in Planar Lipid Bilayers and Liposomes*, edited by A. Iglic (Academic, Amsterdam, 2012), Chap. 7, p. 226.
[41]B. A. Hills, *The Biology of Surfactant* (Cambridge University, London, 1998).
[42]A. Minassian, D. O'Hare, K. H. Parker, J. P. Urban, K. Warensjo, and C. P. Winlove, J. Orthop. Res. **16**, 720 (1998).
[43]J. Israelachvili and H. Wennerstrom, Langmouir **6**, 873 (1990).
[44]R. P. Rand, Annu. Rev. Biophys. Bioeng. **10**, 277 (1981).
[45]E. Schneck, F. Sedlmeier, and R. R. Netz, Proc. Natl. Acad. Sci. U.S.A. **109**, 14405 (2012).
[46]B. N. Lourenço, G. Marchioli, W. Song, R. L. Reis, C. A. van Blitterswijk, M. Karperien, A. van Apeldoorn, and J. F. Mano, Biointerphases **7**, 46 (2012).
[47]A. C. Cowley, N. L. Fuller, R. P. Rand, and V. A. Parsegian, Biochemistry **17**, 3163 (1978).
[48]S. Garcia-Manyes and F. Sanz, Biochim. Biophys. Acta **1798**, 741 (2010).
[49]F. Dekkiche, M. C. Corneci, A. M. Trunfio-Sfarghiu, M. H. Meurisse, Y. Berthier, W. Kaabar, and J.-P. Rieu, Colloids Surf., B **80**, 232 (2010).
[50]S. Graindorge, W. Ferrandez, E. Ingham, Z. Jin, P. Twigg, and J. Fisher. Proc. Inst. Mech. Eng., Part H **220**, 597 (2006).
[51]K. Wierzcholski, Int. J. Appl. Mech. Eng. **11**, 515 (2006).
[52]H. Jing, H. Kim, D. Choi, D. R. Lee, J. S. Lee, C. J. Yu, and K. Shin, Biointerphases **6**, 73 (2011).