7th World Tribology Congress, WTC 2022
July 10-15, 2022, Lyon, France

Lubrication effect of ECM molecules and SF components evaluated with agarose-chondrocyte model

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In order to understand the mechanism of the extremely low friction between articular cartilages, the synovial joint lubrication was experimentally reproduced by using pseudo synovial fluid (PSF) and cartilage tissue model in which living chondrocytes were seeded in agarose gels and cultured to elaborate extracellular matrix (ECM). Friction tests were conducted using rheometer to evaluate the effect of synthesized ECMs and synovial fluid (SF) components on friction property. From results of the friction test, we concluded that the former reduced the shear resistance of the model surface, while the latter prevented the adsorption of hydrogel surface.

Keywords: Cartilage, Chondrocytes, Extracellular Matrix, Synovial fluid, hydrogel friction

1. Introduction

In recent years, research in the field of biomimetics has been aggressively carried out to apply highly efficient and high-performance biological functions as an engineering technology. Among them, biological joints are known to have an extremely low friction, but the details of lubrication mechanism are still the subject of active debate. We focused on two models for the synovial joint lubrication: the surface gel-hydrated lubrication model [1] with proteoglycans of extracellular matrix (ECM) and the boundary lubrication film model [2] formed by the interaction of ECM and hyaluronic acid (HA), dipalmitoyl phosphatidylcholine (DPPC) in synovial fluid (SF). In this study, we evaluated the friction reduction effect of ECM and synovial fluid components by using “living” cartilage tissue model.

2. Methods

2.1. Specimen

Fresh cartilage tissue was harvested from the metacarpal phalangeal joint of cattle, and digested by sequential enzyme treatments to isolate chondrocytes. Chondrocytes were seeded in 2 wt % agarose hydrogels at the initial cell concentration of 10^7/mL. Cylindrical specimen was prepared (φ = 8 mm, t = 2.5 mm) and subsequently cultured in the CO_2 incubator at 37°C and 5% CO_2 concentration for several weeks.

2.2. Fluorescence observation

To observe the distribution of synthesized ECM, keratan sulfate, a carbohydrate chain of proteoglycans, was fluorescently labeled with the antibody staining method and observed with the confocal laser microscope.

2.3. Friction test

The friction test under the constant vertical strain and coaxial unidirectional rotation were conducted using the rheometer (MCR-301, Anton Paar). Specimens were compressed by 15 % in the normal direction by the glass plate and the friction measurement was conducted after stress relaxation in lubricant for 5 minutes. Phosphate buffered saline (PBS) and pseudo synovial fluid (PSF) were used as test lubricants. The composition of PSF is PBS : HA : DPPC : Albumin : γ-globulin = 97.39 : 0.5 : 0.01 : 1.4 : 0.7 (wt %) .

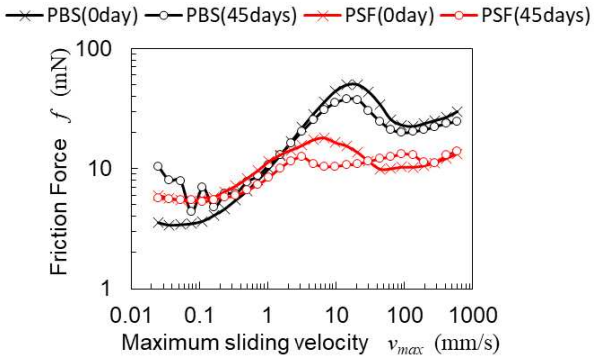

Figure 1: Velocity dependence of the friction force for 0day and 45days cultured specimens in PBS or PSF.

3. Results and discussions

The velocity depended frictional behavior of 0day and 45days cultured specimens in PBS or PSF is shown in Figure 1. In PBS, The maximum friction force of cultured specimen became lower compared with the 0day specimen. It would be due to the formation of the hydrated proteoglycans layer on the surface which could reduce the shear resistance of the interface. On the other hand, PSF showed certain lubrication effect on the cartilage model. The friction force peak was shifted to the lower sliding velocity and the peak friction was lowered even for the 0day specimen. It might be attributable to the viscosity of HA, which can increase viscosity and thickness of lubrication film. The adsorption of the agarose molecular chain on the glass surface is prevented by interposing HA with high molecular weight at the interface. In the case of 45days culture in PSF, the proteoglycan elaborated in the surface region attracted HA which locally increases the viscosity of the interface and further reduced the friction force.

4. References

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