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Basic study on nanofabrication of biodegradable plastics applying biochemical machining

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ABSTRACT

Plastics are regarded as proper materials for microfluidic chips, and, in particular, biodegradable plastics will be more appropriate from the viewpoint of environmental friendliness. In addition, when they are utilized as chip substrate, biochemical machining can be applied. Biochemical machining is a process, which utilizes enzymatic biodegradation. This paper describes studies on application of biochemical machining to nanofabrication of poly(L-lactic acid) (PLLA), which is one of the most widely used biodegradable plastics industrially. The degradation performance of enzyme (proteinase K) was experimentally evaluated to extrapolate a suitable condition for nanofabrication. Additionally, in order to make arbitrary channels with nano-order depth on PLLA, mask fabrication was proposed. It is a fabrication method to control degraded regions and obtain desired shapes with a mask, which has penetrating grooves. Using this method, we achieved fabrication of straight grooves with nano-scale depth. In conclusion, it is clear that biochemical machining can realize an effective process of arbitrarily shaped nanogrooves on PLLA.

1. Introduction

In the field of medical science and biological chemistry, μTAS (micro total analysis system) has been rapidly developed and drawing much attention. Furthermore, as miniaturization of such analysis system, demands for development of nanofluidic chips are increasing. Although glass and polydimethylsiloxane (PDMS) are conventionally used for chip materials, they are not appropriate enough for mass production resulted from practical use of the chips in the near future. Meanwhile, plastic is regarded as a proper material due to its high productivity, and in particular, biodegradable plastic can be more appropriate for mass disposal arising from mass production. Biodegradable plastic is a material defined as polymer, which is degraded into low molecular substances by bacteria. It is not harmful to environment and can reduce environmental load of mass disposal of used chips [1]. Therefore, development of an efficient and accurate nanofabrication method for biodegradable plastic is required for fabrication of disposable and environmental friendly nanofluidic chips. Conventional nanofabrication processes of plastic have some problems. Although mold injection is usually used for fine processing of plastic, achievement of accurate nanofabrication is difficult since the sizes and shapes of the molds are limited in order to prevent products from deformation in demolding. In addition, photolithography and etching processes require hazardous chemicals, great amount of energy, and much time.

In order to solve these problems, an application of biochemical machining to nanofabrication of plastic chips is proposed. Biochemical machining is a wet etching style fabrication of biodegradable materials, which utilizes biochemical reactions of enzymatic hydrolysis, using enzyme solution as etchant [2]. It was originally proposed as one of biological fabrication techniques. Biological fabrication has been proposed as a new fabrication method possessing feasibility to gain advantages which conventional ones cannot obtain [3–6]. Table 1 shows comparison of conventional micro and nanofabrication methods and biochemical machining [7]. The value of processing scale is based on the result of this paper. Biochemical machining has potential for precise nanofabrication, and moreover has the advantage that huge apparatuses and hazardous chemicals are unnecessary since it basically requires only naturally derived enzyme solution.

In the past studies, the performance of biochemical machining in macro scale has been evaluated. Meanwhile, biochemical degradation generally progresses at an atomic unit, and thereby, biochemical machining is regarded as material removal process on nanometric scale. In this study, we especially propose an application of biochemical machining to nanofabrication of plastic for nanofluidic chips utilizing extended nano space, which consists of only interfacial regions in 100 nm scale. Since fluid shows specific

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Table 1
Comparison between conventional micro and nanofabrication methods and biochemical machining.

	Processing scale	Processed area	Processing flexibility	Environmental friendliness
Micro machining	1 μm	100 mm	○	○
Etching (photo resist)	0.1 μm	100 mm	△	×
Micro molding/nano imprint	1 μm/10 nm	10 mm	×	△
Biochemical machining	10 nm	100 mm	△	◎

behaviors in interfacial regions, it is available for effective analysis of distinctive fluidic behaviors and physical phenomenon in extended nano space. Therefore, development of nanofabrication methods for extended nano space are now rapidly required. The demands for the fabrication of the fluidic chips are as follows:

1. Machining of 100 nm scale with error under 10%.
2. Completing of 100 nm scale fabrication for a controllable short time (in the range of processing time from minutes to tens of minutes).

In this paper, properties of biochemical machining is evaluated and an appropriate condition is extrapolated in order to satisfy these demands. And then, application possibility of biochemical machining is proposed and examined to make desired shaped channels.

2. Principle of biochemical machining of PLLA

In this study, poly(L-lactide) (PLLA) is used for test pieces. PLLA is the most widely used biodegradable plastic because of its high fusion point, high stiffness, and, particular eco-friendliness due to biomass feedstock [1]. The chemical structure of PLLA is shown in Fig. 1. The degradation of PLLA is progressed by cleavages of ester bonds as shown in Fig. 2. The cleavages are progressed by hydrolysis and accelerated tremendously by catalytic ability of proteinase K, which is a serine protease derived from a fungus stain *Tritirachium album* [8]. The enzymatic degradation of PLLA with proteinase K is conducted in a buffer solution. At the start of the enzymatic degradation of PLLA, proteinase K in the solution approaches the surface of PLLA and adsorbs. The catalytic ability begins to work as soon as the adsorption [9]. As the degradation reaction progresses, PLLA molecule chains are separated into oligomers or monomers of lactic acid and solve in the solution from the surface of PLLA. Therefore, it is regarded as a nanoremoval process like wet etching. Although the degradation reaction without the enzyme progresses through bulk erosion mechanism by autocatalytic hydrolysis and results in fragmentation of PLLA, the enzymatic degradation is progressed by surface erosion [10]. Moreover, the velocity of the enzymatic degradation is relatively so high that the influence of the autocatalytic hydrolysis is negligible. Hence, biochemical machining realizes only surface fabrication of PLLA without the internal fragmentation.

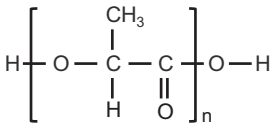

Fig. 1. Chemical structure of PLLA.

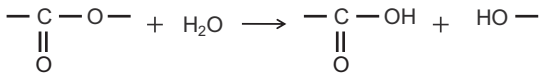

Fig. 2. Chemical equation of cleavage of ester bond.

Regarding the enzyme activity, proteinase K keeps its activity in the range of pH 7.5–12.0 and temperature less than 328 K. The enzyme degradation reaction can be easily stopped as planned by ablution with 40% ethanol solution [9]. This fact indicates the progression of the degradation can be controlled.

The hydrolysis acts preferentially on amorphous regions of PLLA [11]. Thus, amorphous PLLA samples are required for biochemical machining in order to realize homogeneous degradation.

3. Experimental procedure

3.1. Sample of PLLA film

A thin amorphous PLLA film stuck on a glass plate was required for the experimental use. The PLLA films were obtained from PLLA pellets (poly(L-lactide), MW 85,000–160,000, Sigma–Aldrich) by the following process. The PLLA pellets were dissolved in chloroform solution and casted horizontally in a petri dish. After 24 h, only the chloroform had been completely evaporated and a thin film of PLLA was obtained. In order to make an amorphous PLLA sample, the film was fixed between a glass plate and a silicon wafer, and then heated to 440 K, which is the melting point of PLLA. After the film was sufficiently melted, it was soaked in water and quenched to avoid crystallization [12]. Then, the silicon wafer was removed and a thin amorphous PLLA film sample was obtained on the glass plate as shown in Fig. 3 because PLLA has a greater tendency to be pasted on glass surfaces than on silicon surfaces.

Fig. 4 shows an observational result of the PLLA film surface by 3D optical profiler (New View 6200, Zygo). The vertical and lateral resolutions are 0.1 nm and 1.16 μm. The surface roughness (*Ra*) of the PLLA film was measured less than 1 nm, which was the almost same value of that of the silicon wafer. This fact indicates

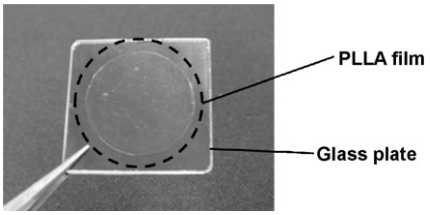

Fig. 3. Sample of PLLA film stuck on a glass plate.

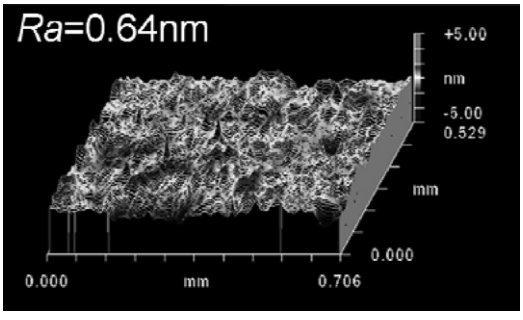

Fig. 4. Surface of PLLA film observed by 3D optical profiler before experimental degradation.