System Identification for Motion of Proteins using an AFM-based Nanorobotic Manipulation

Jungyul Park*, Deok-Ho Kim *, Byungkyu Kim* and Kyo-Il Lee**

* Microsystem Research Center, Korea Institute of Science and Technology (KIST), P.O.BOX 131, Cheongryang, Seoul 130-650, Korea,{ sortpark, kim-dh, bkim}@kist.re.kr
** School of Mechanical and Aerospace Engineering, Seoul National University, San 56-1, Shinlim-dong, Kwanak-gu, Seoul, 151-742, Korea, lki@snu.ac.kr

ABSTRACT

In biomanipulation research area, the reliable force measurement and modeling of biological molecules are essential techniques to estimate the material properties and a motion of the biological molecule. In this study, we introduce the system identification method to estimate motion of protein and material characteristics using AFM-based nanorobotic manipulation. Modeling of piezoresistive AFM cantilever and equivalent model for force-measurement system are presented. Based on equivalent, dynamic motion of a protein is efficiently estimated in real-time. In case of globular protein, damping coefficient and spring constant show good agreement between estimation results and actual values.

Keywords: Nanorobotic Manipulation, Protein, Force sensing, System identification.

1 INTRODUCTION

Sensing and manipulating in the nanoscale level are greatly important in challenging applications such as building nanomachines or biomanipulation. Specially, sensor-based manipulation is necessary to realize efficient and reliable handling of particles under uncertain mechanics in the nanoscale level.

As shown in Figure 1, AFM-based nanorobotic manipulation has been researched for several exploring applications such as nanoindenting/lithography [1], bio-material detection [2], characterization of nano-materials and devices [3], nanoasssembly [4,5], force sensing [13,16] etc. Our previous work [7] presented the implementation of a piezoresistive AFM cantilever for sensor-based nanorobotic manipulation. In particular, for single-molecule biomanipulation such as DNA extension, bending and twisting, protein domain motion, deformation and unfolding, we need to measure the interactive force between single molecule and AFM tip and to obtain dynamic equation to analyze a motion of biological molecules. Therefore, the system identification method is presented to estimate the motion of protein using AFM-based nanorobotic manipulation. In addition, modeling of piezoresistive AFM cantilever and equivalent model for dynamic force-measurement system are presented. The equivalent model enables to estimate dynamic motion of the protein efficiently in real-time.

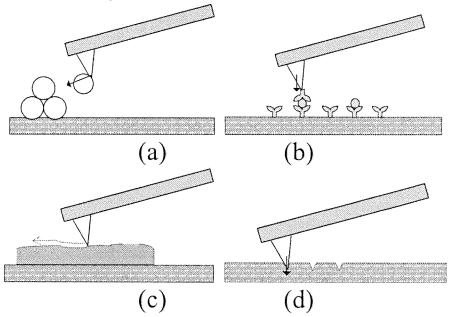


Figure 1: Possible applications using an Atomic Force Microscope (AFM) cantilever as a nanomanipulator nanoassembly, (b) biosensor, (c) nanotribological characterization, (d) nano-indenting/lithography

2 PIEZORESISTIVE AFM CANTILEVER AS A NANOMANIPULATOR

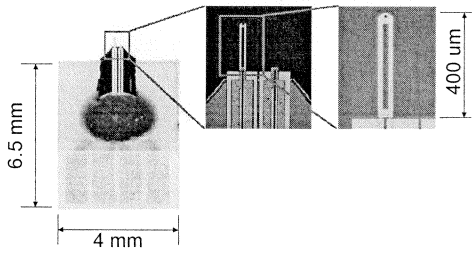


Figure 2: Piezoresistive AFM cantilever in contact-mode (Seiko Instruments Inc.)

In our previous work[7], a "self-sensing" piezoresistive AFM cantilever has been implemented for the sensory nanomanipulation. Figure 2 shows contact-mode AFM cantilever with approximately 20 nm radius of curvature, provided by Seiko Instruments Inc. Self-sensing piezoresistive AFM cantilever enables the sensing of gripping force in the nanoscale by measuring changes in

stress-induced electrical resistance. Table 1 shows electro-mechanical characteristics of a piezoresistive AFM cantilever for the use as the sensory manipulator.

Table 1. Electro-mechanical characteristics of a piezoresistive AFM cantilever

Items	Characteristics
Spring constant	1 N/m
Resonant frequency	38 KHz
Resistance	$550 \pm 150\ \Omega$
Sensitivity	2.6×10^{-5}(△R/R/nm)
Total cantilever length	400 um
Width	50 um
Thickness	5 um
Tip radius	< 20 nm

3 MODELING

3.1 Dynamic Equation of Motion of an AFM Cantilever

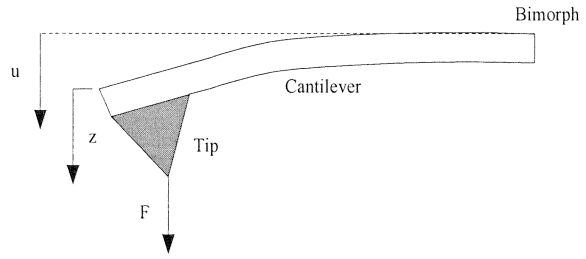


Figure 3: Schematic diagram of AFM cantilever

Modeling of the cantilever is very important for nano-force sensing as well as manipulation control due to its mechanical principle. We derive the dynamic equation of motion of an AFM cantilever with neglecting of tip-sample force. As shown in Figure 3, the position of the bimorph driving the cantilever is given by u, position of the tip is given by z, and the external force is F. Then, dynamic equation of motion of an AFM cantilever is

$$M_c \frac{d^2 z}{dt^2} + \eta_c \frac{dz}{dt} + k_c (z - u) = F \ . \qquad (1)$$

Here, M_c is the mass, η_c the damping coefficient, and k_c the elastic constant of the AFM cantilever.

3.2 Motion of Proteins

Proteins are cellular machines and constitute about 60% of a cell's dry weight. They perform most of the cellular functions such as transducing signals, providing structural support, transporting biomolecules, regulating biological responses, and catalyzing bio-chemical reactions for metabolism. An essential feature of proteins is that the functions of a protein are determined by its 3D confirmation, i.e., the spatial arrangement of the atoms in its folded structure. Protein molecules are deformable, thus their conformations could be altered by mechanical forces. Such alterations can affect protein-protein and protein-DNA recognition, binding and unbinding, leading to changes in downstream biochemical process that control cellular behavior and function. Protein deformation, therefore, is an important concept in molecular biomechanics [12]. Proteins are not static rigid structure. Rather, they are dynamic and undergo constant motions and structural changes in cells under normal physiological conditions. These changes include large-scale (~5-50 Å) movements of domains as well as small-scale (~0.5 Å) random movements of secondary structures or domains, or 'breathing'. The time scales for protein motion and deformation in biological process may span many orders of magnitude, ranging from one femtosecond (10^{-15} s) to a few seconds. Many proteins or protein domains have an approximately rounded shape, refer to as globular proteins.

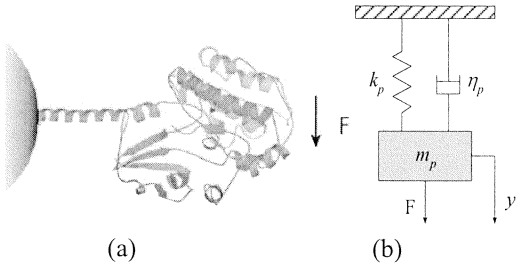


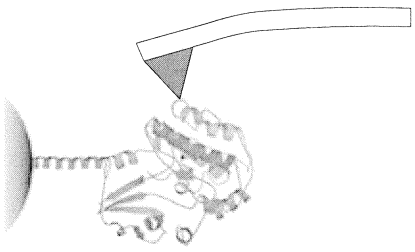
Figure 4: The motion of a protein under applied force F. (a) A globular protein immobilized on a surface through an α-helix. (b) The mass-spring-dashpot system as a model for protein motion.

To illustrate, consider a globular protein immobilized on a surface through a lever-arm (e.g., an α-helix), as shown schematically in Figure 4(a). Under an applied force F the small motion of the protein in vertical direction can be analyzed based on a mass-spring-dashpot system shown in Figure 4(b). The corresponding governing equation for displacement y is

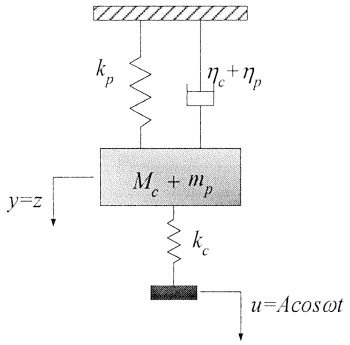
$$m_p \frac{d^2 y}{dt^2} + \eta_p \frac{dy}{dt} + k_p y = F \ , \qquad (2)$$

where m_p is the mass and η_p the viscous drag coefficient of the protein, and k_p is the elastic constant of the lever arm.

4 SYSTEME IDENTIFICATION FOR MOTION OF PROTEINS



(a)



(b)

Figure 5 : (a) Measurement system for motion of proteins using AFM cantilever. (b) The equivalent model for measurement system.

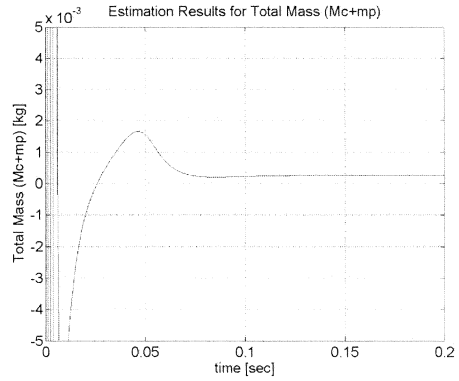
To perform system identification for motion of protein, the measurement system is constructed. Figure 5(a) shows the measurement system for motion of protein using AFM cantilever. Target protein is attached to AFM cantilever. Bimorph is controlled to apply sine wave shaped force to protein. Then, position of cantilever, that is equal position of lever-arm, is detected by piezoresistive sensing. Figure 5(b) shows the equivalent model for measurement system using AFM cantilever. Equation of this equivalent model is given by

$$(M_c + m_p)\frac{d^2z}{dt^2} + (\eta_c + \eta_p)\frac{dz}{dt} + (k_c + k_p)z = k_c u \,. \qquad (3)$$

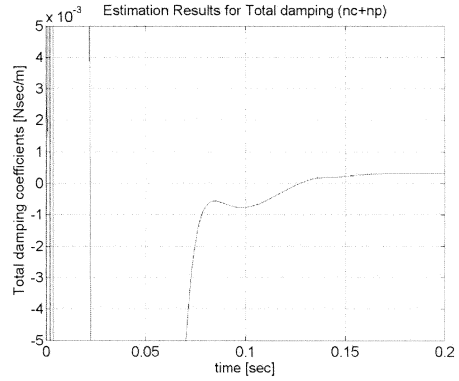
In this paper, we consider the recursive least square (RLS) algorithm to estimate parameters of protein. The RLS algorithm is technique for on-line identification. Using this technique, therefore, the dynamic characteristics of protein can be acquired in real-time. RLS algorithm is given by

$$\theta_{est}(t) = \theta_{est}(t-1) + R^{-1}(t)\varphi(t)[Y(t) - \varphi^T(t)\theta_{est}(t-1)]\,,$$
$$R(t) = \lambda(t)R(t-1) + \varphi(t)\varphi^T(t)\,, \qquad (4)$$

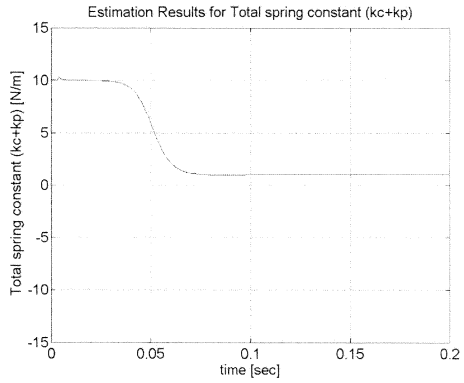
where $\varphi(t)$ is the regression vector, θ is the parameter vector, R is symmetric and R>0, and $\lambda(t)$ is weighting sequence [14].



(a)



(b)



(c)

Figure 6: Simulation results for system identification of protein motion. (a) Estimation results for total mass of AFM cantilever and protein. (b) Estimation results for total damping coefficient of AFM cantilever and protein. (c) Estimation results for total spring constant of AFM cantilever and protein

To apply the RLS algorithm, above equation for equivalent model must be reassigned. Modified equivalent equation is given by

$$Y(t) = \varphi^T(t)\theta\,, \qquad (5)$$

where $Y(t) = k_c u$, $\theta = [M_c + m_p \quad \eta_c + \eta_p \quad k_c + k_p]^T$, and $\varphi(t) = [\frac{d^2 z}{dt^2} \quad \frac{dz}{dt} \quad z]^T$.

System identification for motion of protein is performed using Eq.(4)-(5) with MATLAB. Simulation condition is like as followings. The mass of AFM cantilever is 0.2533 g, the damping coefficient is 3.183×10^{-4} N s/m, and spring constant is 1 N/m. The input signal is applied as

$$u = A \cos \omega t , \tag{6}$$

where A is the amplitude of input signal and ω is input angular frequency. Input frequency is applied by bimorph from 1 Hz to 1 KHz.

Figure 6 shows the simulation results for system identification of protein motion. Parameters are converged in 0.2 sec. The estimation result for drag coefficient of protein is 59.99 pN s/m, and spring constant is 5.99 pN/nm. . The actual globular protein has a mass of 100 kDa (1 Dalton = 1.66×10^{-24} g) and a drag coefficient of 60 pN s/m, and the lever arm has spring constant of 6pN/nm [15]. These results show good agreement with actual protein parameters as stated above. But the estimation result for mass of protein is -1.79 Gda, that is far from real protein value. Because protein mass is too small, differences of significant figures between mass of protein and mass of AFM cantilever can't easily classified in equivalent model due to numerical truncation error(see Eq.3). But, this methodology contributes to estimation of mechanical properties of protein, which are damping coefficient and spring constant, in real time. Previous results [13,16] can't estimate the damping coefficient of protein.

5 CONCLUSION

Protein deformation and motion are an important concept in molecular biomechanics. Previous researches didn't extract the dynamic motion of protein. They could only show model of the static or quasi-static motion of protein. This paper has three contributions in bio-mechanics. First, we construct the on-line measurement methodology to identify protein motion using AFM-based nanorobotic manipulation. Second, we introduce the mothod to derive dynamic motion of protein, not static motion. Third we present efficient system identification methodology using RLS to estimate the protein parameter in real-time. These contributions are available in other bio-mechanics.

Acknowledgements

This research, under the contract project code MS-02-324-01, has been supported by the Intelligent Microsystem Center(IMC: http://www.microsystem.re.kr), which carries out one of the 21st century's Frontier R & D Projects sponsored by the Korea Ministry of Science & Technology.

REFERENCES

[1] A.A.G. Requicha et al., "Massively parallel nanorobotics for lithography and data storage," *Int'l J. Robotics Research*, vol. 18. no.3, pp.344~350, 1999.

[2] David R. Baselt et al., "Biosensor based on force microscope technology," *Journal of Vac. Sci. Technol. B*, 14(2), pp.789~793, 1996.

[3] M. Sitti, "Nanotribological characterization system by AFM based controlled pushing," *Proc. IEEE-NANO 2001*, pp.99~104, 2001.

[4] M. Sitti, and H. Hashimoto, "Controlled pushing of nanoparticles: modeling and experiments," *IEEE/ASME Trans. on Mechatronics*, vol.5, pp.199~211, June 2000.

[5] L.Dong et al., "Three-dimensional nanoassembly of multi-walled carbon nanotubes through nanorobotic manipulation by using electron-beam-induced deposition," *Proc. IEEE-NANO 2001,* pp.93~98, 2001.

[6] D.H. Kim et al., "Motion planning of an AFM-based nanomanipulator in a sensor-based nanorobotic manipulation system," *Proc. Int'l Workshop on Microfactory*, Minnesota, USA, September, 2002.

[7] D.H. Kim et al., "Implementation of self-sensing MEMS cantilevers for nanomanipulation," *Proc. the 4th Korean MEMS conference*, pp.120~125, 2002.

[8] J.Colchero, E.Meyer, and O. Marti. "Friction on atomic scale," *Handbook of Micro/Nanotribology*, 2nd Edition, CRC Press, pp. 273~333, 1999.

[9] J. Israelachvili, *Intermolecular Forces*, 2nd Edition, Academic Press, San Diego, 1992

[10] Dror Sarid, *Exploring Scanning Probe Microscopy with MATHEMATICA*, John Willey & Sons, Inc., 1997.

[11] G. Bao, "Mechanics of biomolecules," *Journal of Mechanics and Physics of Solids*, vol. 50, pp. 2237-2274, 2002.

[12] G. Bao, "Single-molecule biomechanics : DNA and protein deformation," *Mechanics in Biology, ASME AMD*, vol. 242, pp. 25-35, 2000.

[13] Keita Mitsui, Ken Nakajima, Hideo Arakawa, Masahiko Hara, and Atsushi Ikai, "Dynamic measurement of single protein's mechanical properties," *Biochemical and Biophysical Research Communications*, vol. 272, pp. 55-63, 2000

[14] Lennart Ljung, *System Identification : theory for the user*, 2nd Edition, Prentice Hall

[15] J. Howard, *Mechanics of motor proteins and the cytoskeleton*, Sinauer Associates, Sunderland, MA. 2001

[16] C. S. Hodges, "Measuring forces with the AFM: polymeric surfaces in liquids," *Advances in colloid and Interface Science*, vol. 90, pp. 13-75, 2002