

Supporting Information for

Confinement Effect on Structure and Elasticity of Proteins Interfacing Polymers

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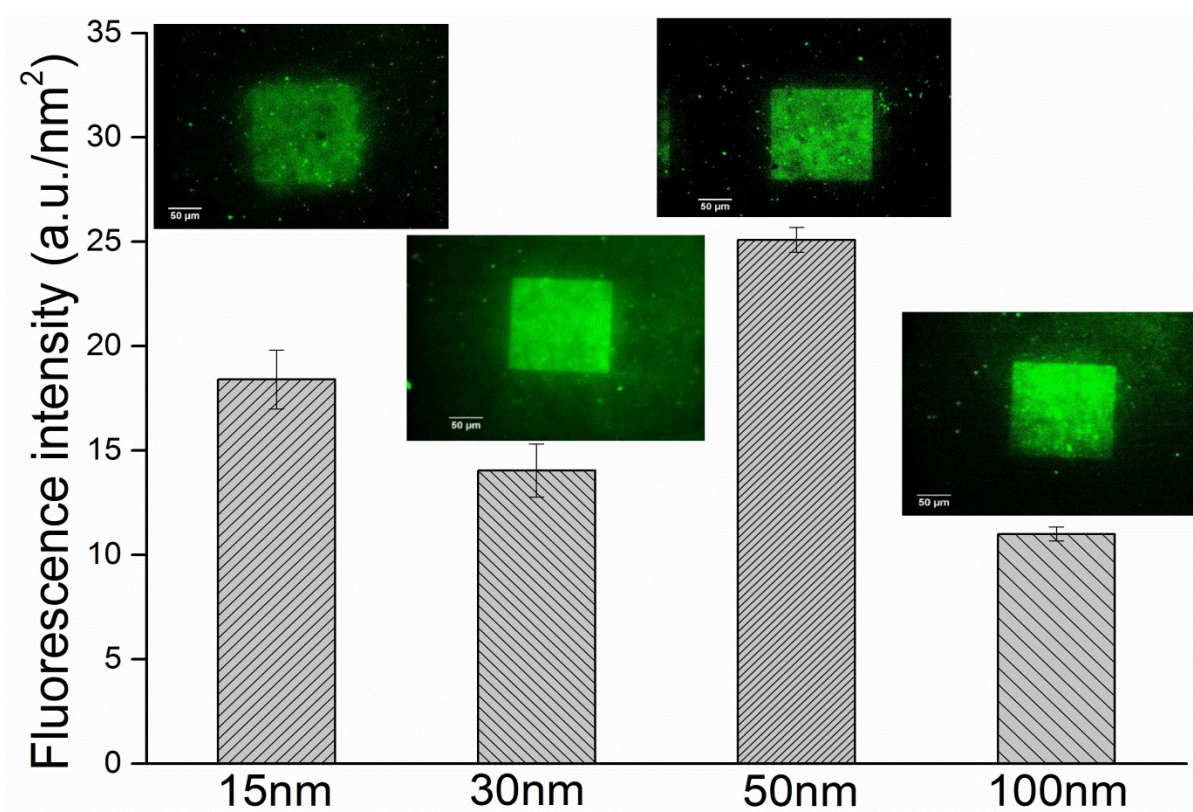


Fig S1. Averaged fluorescence intensity per area (a.u./nm²) is used to compare fibrinogen density in different pore sizes.

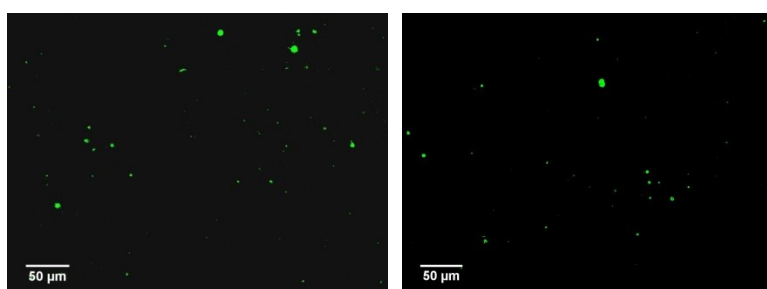


Fig S2. Fluorescence images of protein solution washed open pores (left: 50nm, right: 100nm) without silane functionalization.

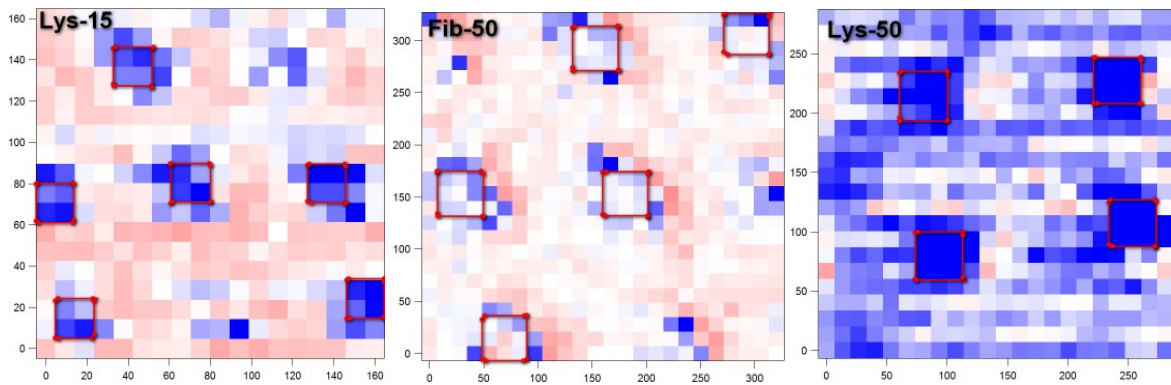


Fig S3. Adhesion forces of labeled regions are averaged over many pores to calculate the net adhesion force change after protein attachments. Due to the resolution limitation of the technique, we were careful in setting size of the pores in adhesion force maps and compare these areas with the real pore sizes from the AFM topography images.

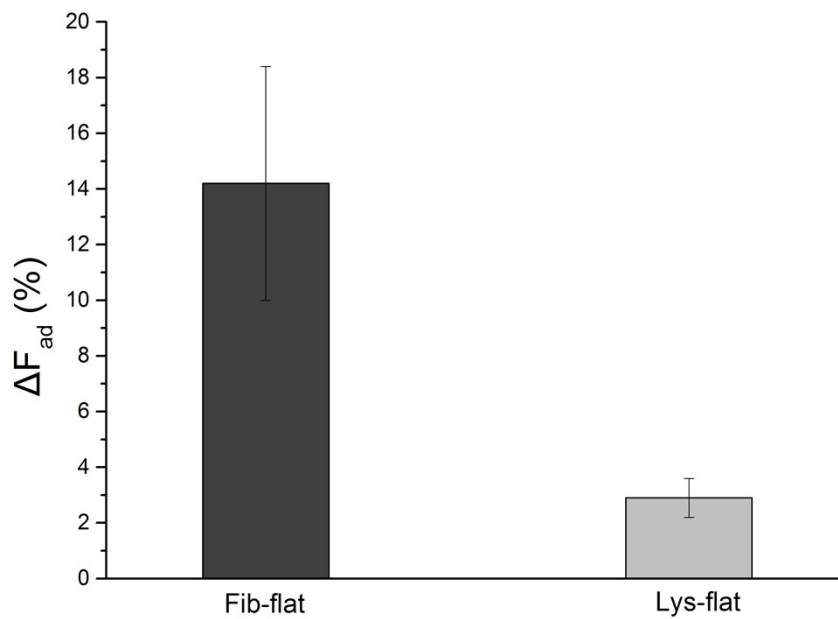
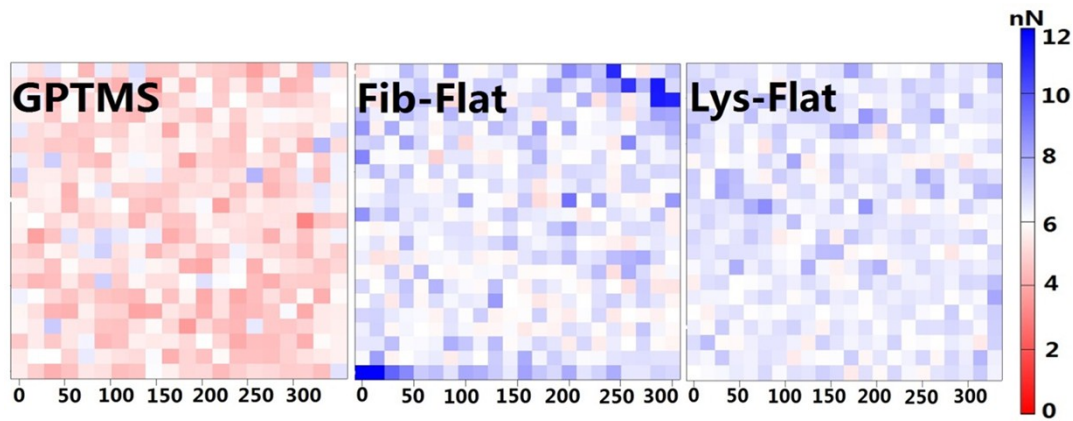


Fig S4. Adhesion force maps of GPTMS surface, fibrinogen and lysozyme functionalized flat surfaces and difference in adhesion force before and after protein addition on flat surfaces.

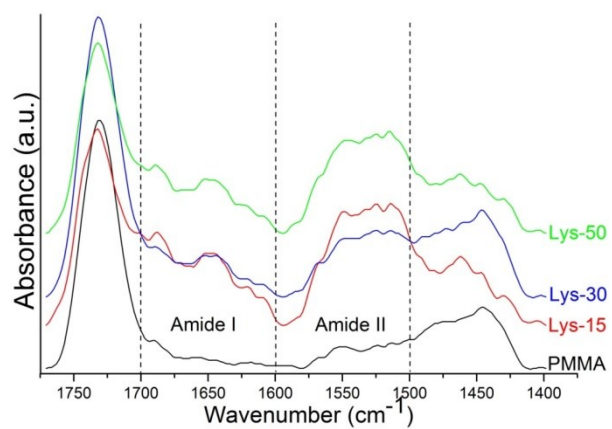
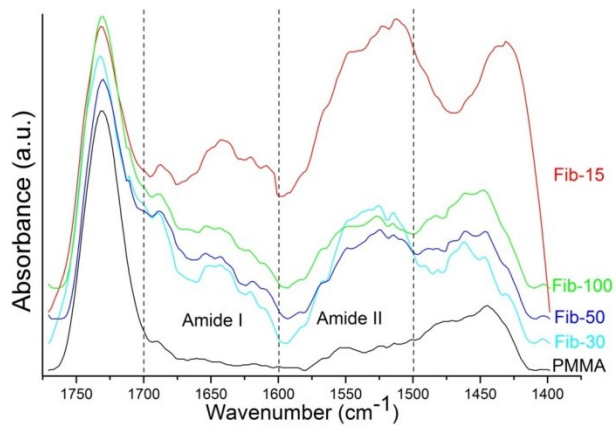


Fig S5. IR spectra of fibrinogen (top), lysozyme (bottom) functionalized PMMA porous films.

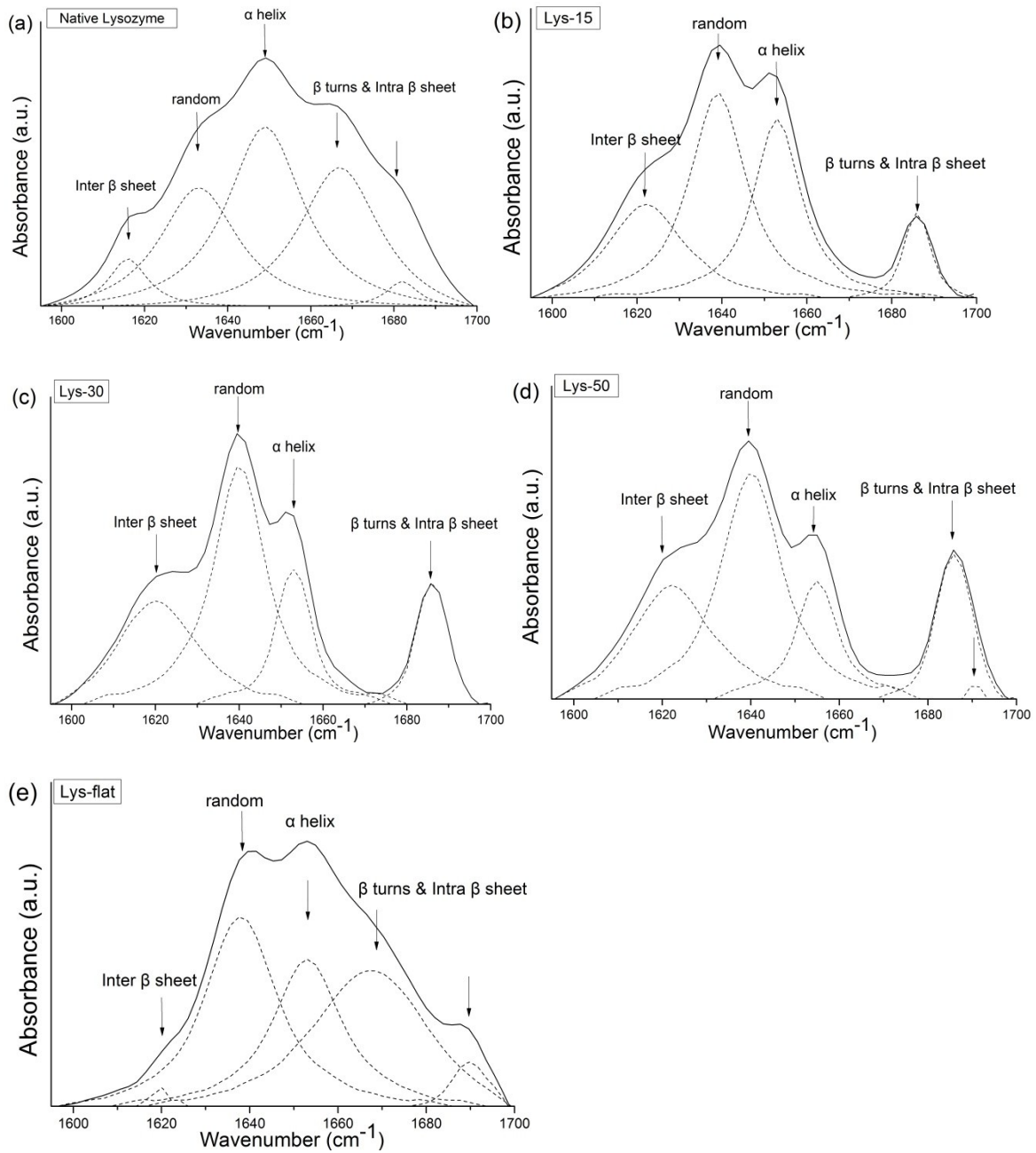


Fig S6. Separation of the amide I bands of (a) native lysozyme, lysozyme tethered onto (b) 15nm, (c) 30nm and (d) 50nm pores of PMMA thin films and (e) silanized flat surfaces.

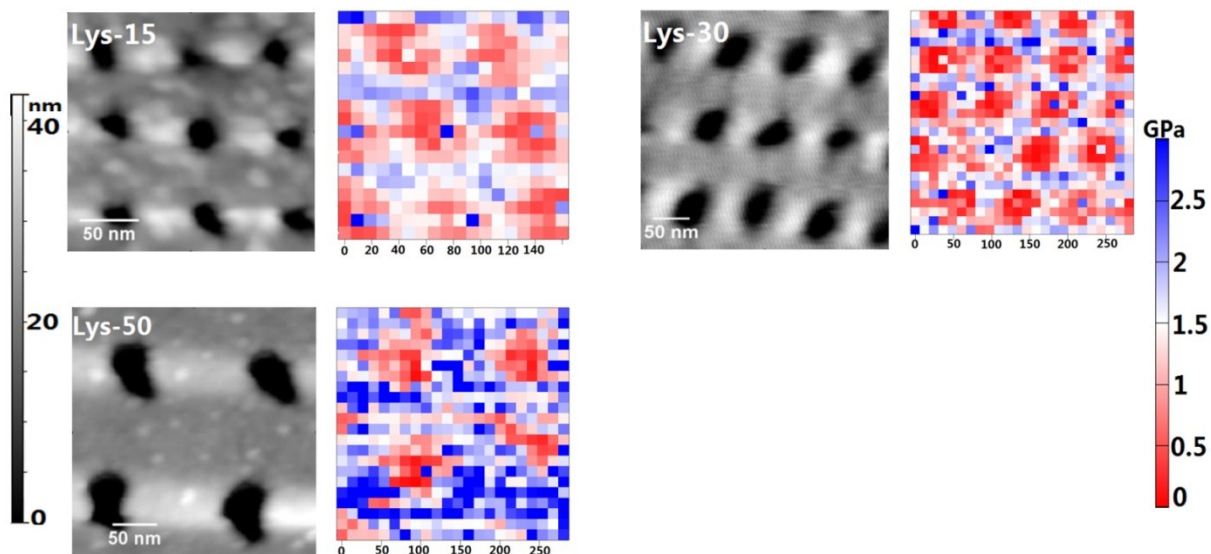


Fig S7. Height images (left) and corresponding elastic modulus maps (right) of lysozyme, functionalized nanoporous PMMA films. Force and modulus maps obtained at resolution of 22x22 points in contact mode.

% structure	Fib-15	Fib-30	Fib-50	Fib-100
Inter β sheet (1615-1635 cm^{-1})	16 \pm 2	17 \pm 6	6 \pm 4	8 \pm 1
β turns & Intra β sheet (1670-1690 cm^{-1})	14 \pm 3	8 \pm 4	24 \pm 2	5 \pm 2
Random (1635-1645 cm^{-1})	59 \pm 2	51 \pm 13	23 \pm 12	54 \pm 9
α helix (1645-1665 cm^{-1})	12 \pm 2	25 \pm 3	47 \pm 5	33 \pm 5

Table S1. Spectroscopic analysis of secondary structures of fibrinogen tethered to nanopores of different sizes in hydrated state.

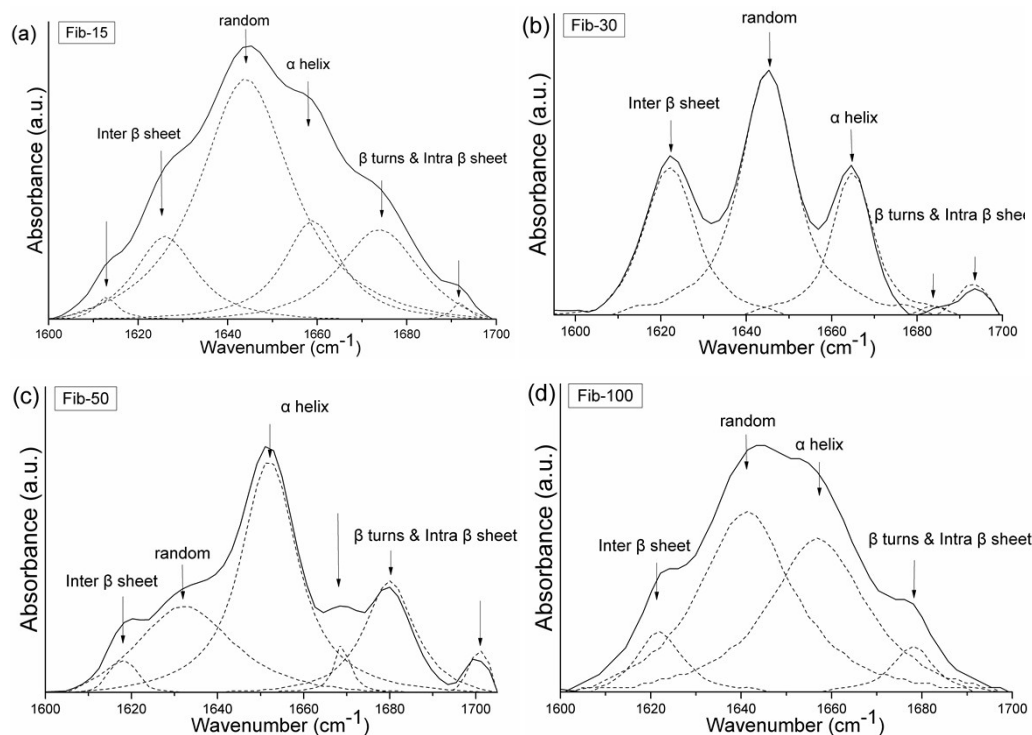


Fig S8. Structural analysis of the amide I bands of fibrinogen tethered on (a) 15nm, (b) 30nm, (c) 50nm and (d) 100nm nanoporous PMMA thin films in hydrated state. Deuterated water peak at $\sim 1650\text{cm}^{-1}$ was subtracted from all spectra before the analysis.

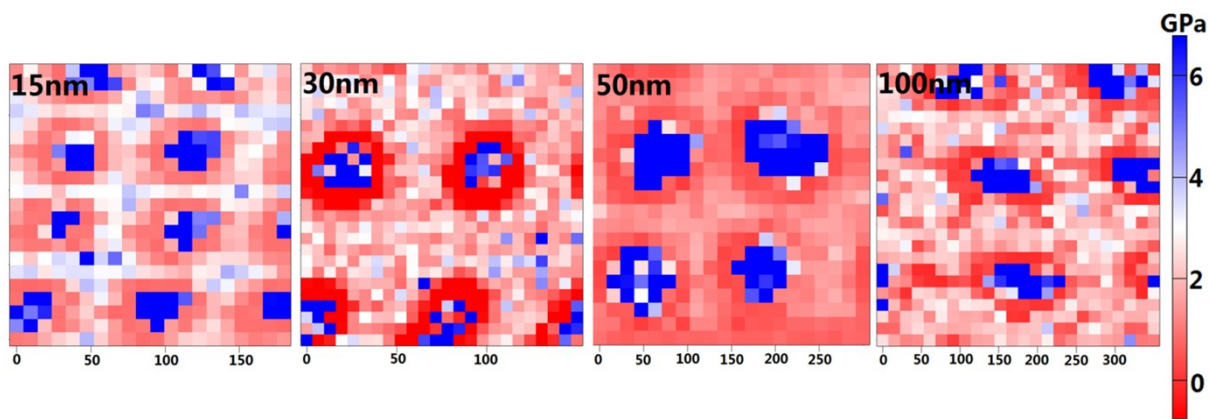


Fig S9. Elastic moduli maps of pores before protein functionalization.

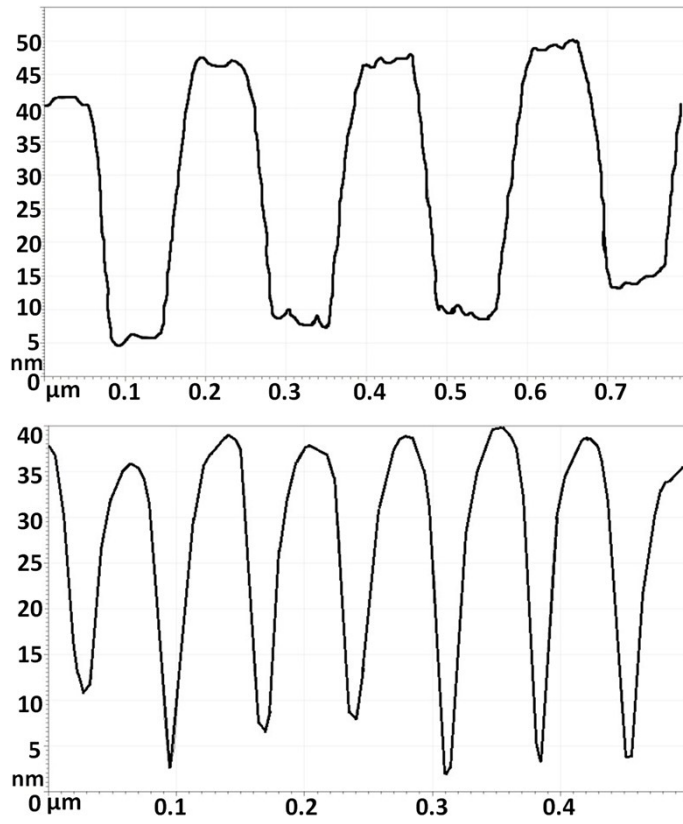
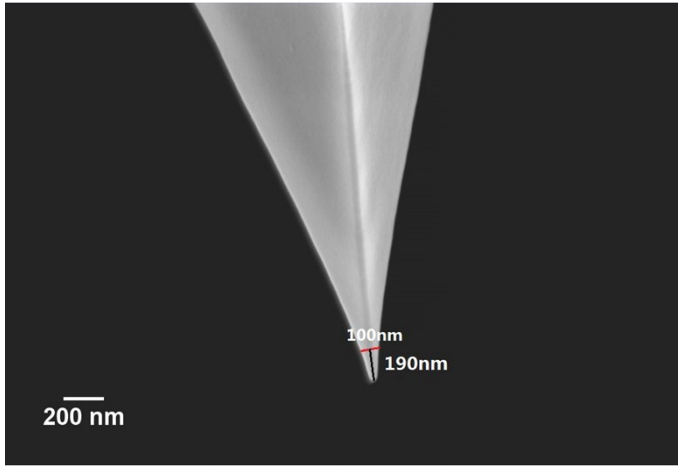


Fig S10. SEM image of the AFM probe used in nanoindentations. Averaged pore depth is measured to be around 35nm for the 15 nm and 100nm pores from the AFM height profiles.