

Characterizing single fibronectin-integrin complexes

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SUPPLEMENTARY MATERIAL

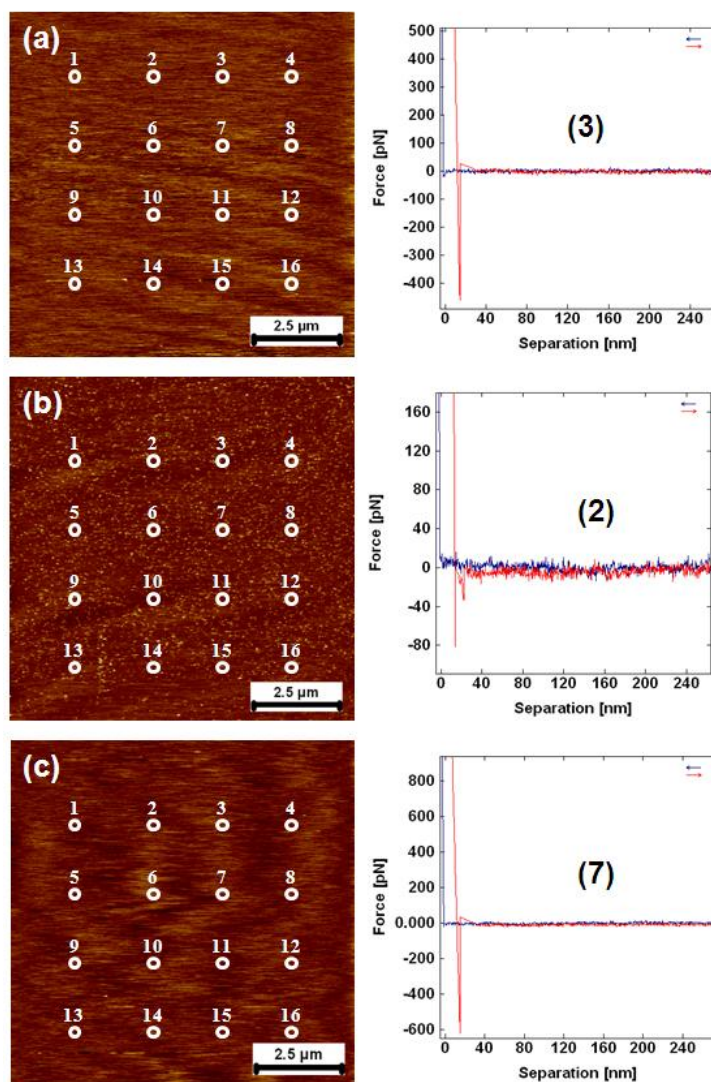


Fig 1 Experimental protocol to determine effect of potential tip contamination and confirming that integrin molecules remain on the surface. Atomic force microscopy images of bare mica before the adsorption of integrin (a), integrin on mica (b) and bare mica subsequent to the adsorption of integrin (c). Numbered points on the images correspond to individual force distance measurements. One such curve is shown on the right of each image in which corresponding to point on the images. Force distance curves obtained before and after the adsorption are identical and are characteristic for bare mica confirming that the integrins remain on the surface and are not “picked up” by the tip. This is in contrast to force distance curves on the integrin sample which show the characteristic fingerprint of the adsorbed molecules.

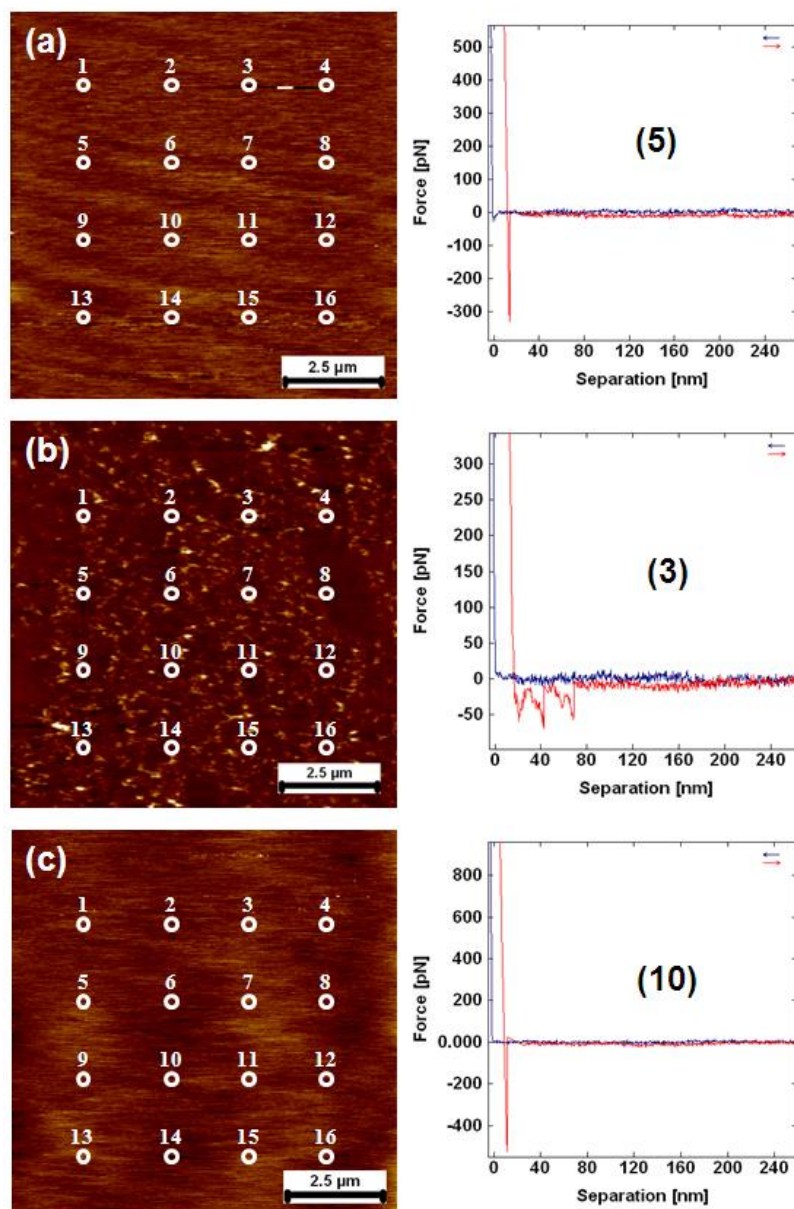


Fig 2 Experimental protocol to determine effect of potential tip contamination by fibronectin-integrin and that the fibronectin/integrin remains on the surface. Atomic force microscopy images of bare mica before the adsorption of fibronectin-integrin (a), fibronectin-integrin on mica (b) and bare mica after the adsorption of fibronectin-integrin (c). Numbered points on the images correspond to individual force distance measurements. One such curve is shown on the right of each image in which corresponding to point on the images. Force distance curves obtained before and after the adsorption are identical, characteristic of bare mica, and show no sawtooth pattern (unfolding of protein). Force distance curves on the fibronectin integrin complexes exhibit the characteristic saw tooth pattern.

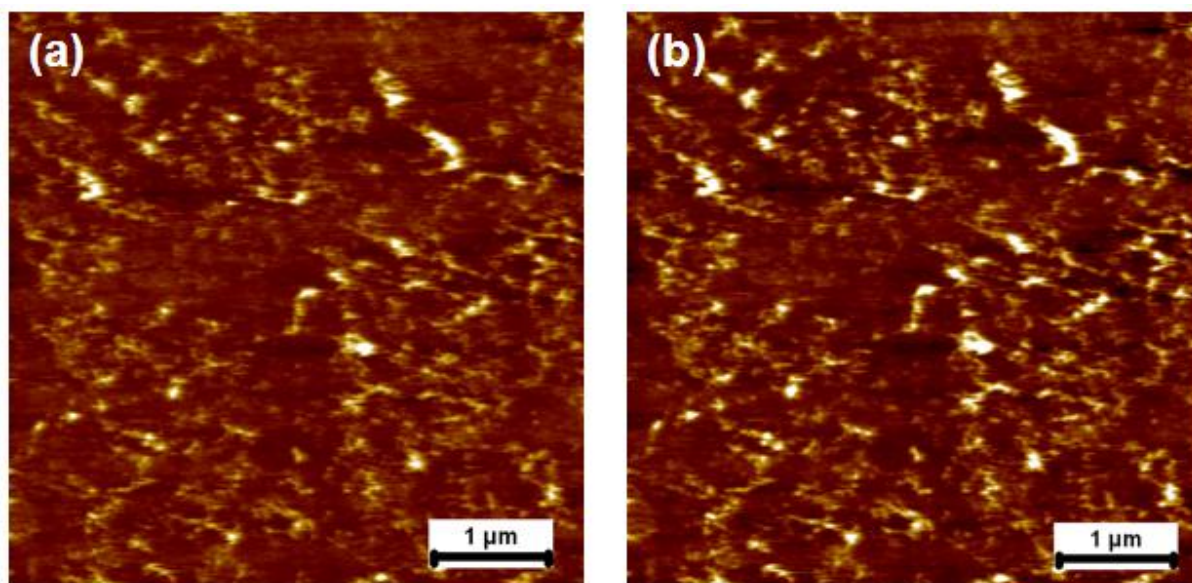


Fig 3 Determination of the effect of drift and force spectra collection on lipid fibronectin-integrin imaging. AFM image taken before force spectroscopy measurements (a). AFM image taken immediately after force distance measurements (b). Drift is seen to have a negligible effect within the time frame of the experiments.

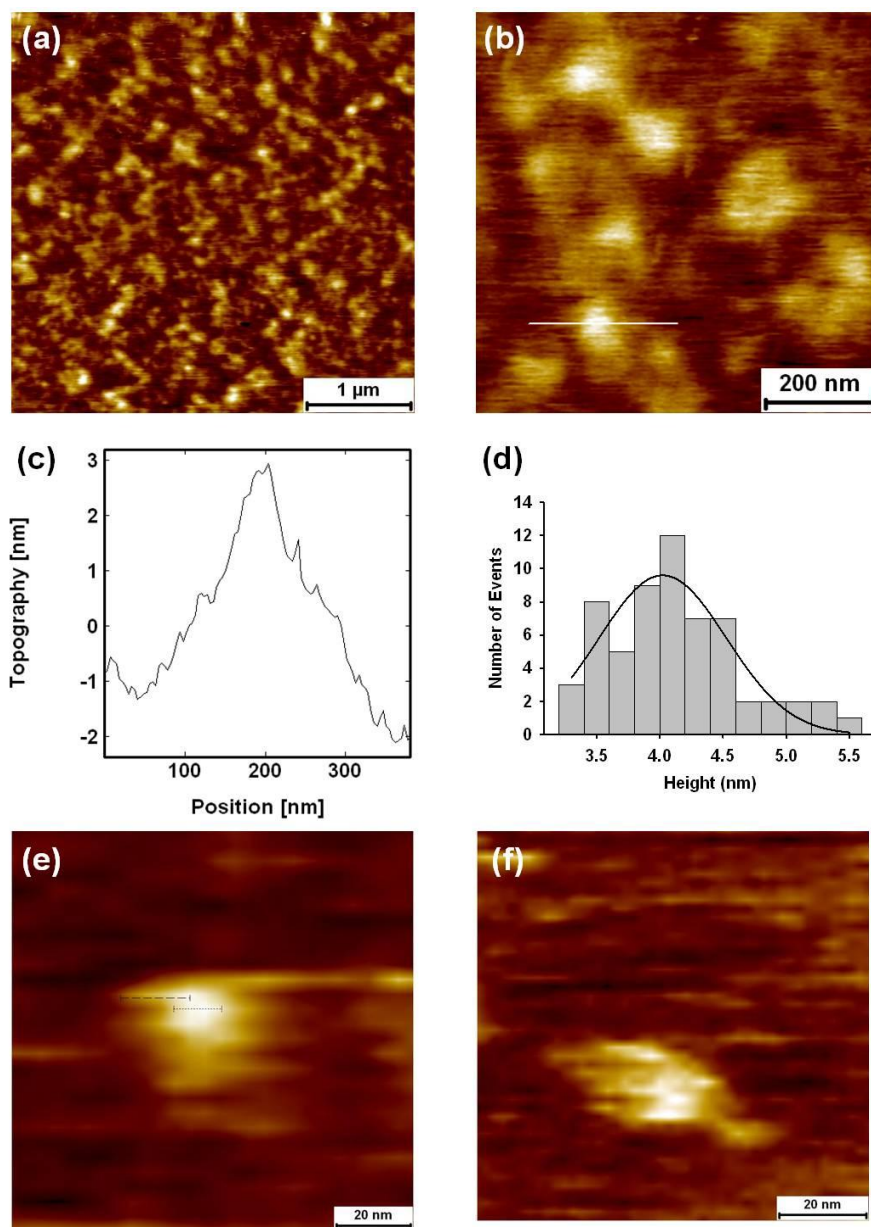


Fig 4 Preformed complexes of fibronectin-integrin. AFM images of complexes adsorbed on mica at a concentration of 5 µg/mL (a and b). Corresponding height profile (c) and distribution (d). High resolution AFM images taken with carbon nanotube tips at a concentration of 1 µg/mL (e and f). Upper dashed line corresponds to the tail region of the integrin whilst the lower dotted line highlights the head region.