

**USE OF ATOMIC FORCE MICROSCOPY (AFM) TO MEASURE INTEGRIN – FIBRONECTIN (FN) ADHESION FORCE IN VASCULAR SMOOTH MUSCLE CELLS (VSM)**

**Zhe Sun**<sup>1</sup>, Ulrich Pohl<sup>2</sup>, Gerald A. Meininger<sup>1</sup>; <sup>1</sup>Texas A&M Univ. Sys. HSC, Dept. of Medical Physiology, College Station, Texas 77843-1114, <sup>2</sup>Ludwig Maximilians Univ., Germany

The adhesion forces of FN and monoclonal antibodies (Mab) to  $\beta 1$  or  $\alpha 5$  integrins were measured for VSM with AFM. VSM were isolated and cultured from rat skeletal muscle arterioles (70–100  $\mu\text{m}$  dia). AFM probes were coated with FN or Mab using a polyethylene glycol linker system. To measure binding strength, the AFM probe was programmed to contact and retract from the cell surface at 550 nm/s. During retraction, detachment events were quantified as binding force. The binding forces (mean $\pm$ sd; pN) were 66 $\pm$ 11 and 55 $\pm$ 8 for Mab HM $\beta 1$ -1 and Ha2/5 to  $\beta 1$  integrin, respectively; 63 $\pm$  13 for HM $\alpha 5$ -1to  $\alpha 5$  integrin; and 62 $\pm$ 14 with serum and 28 $\pm$ 7 without serum for FN. Specificity was examined by addition of soluble  $\beta 1$  Mab to the VSM media.  $\beta 1$  blocking Mab reduced binding incidence but not average binding force. Changes in cell surface rigidity (Young's modulus) were also measured. The rigidity of the cell above the nucleus increased by approximately 40% in response to the FN coated probe, but decreased by about 30% in response to a  $\beta$  tubulin Mab coated probe. These results indicate the feasibility of measuring attachment strength between FN and integrins and show that cortical membrane stiffening, cytoskeletal remodeling, occurs in response to FN interaction with VSM.