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**MECHANISMS FOR THE REGULATION OF CYTOSKELETAL
ORGANIZATION AND TENSION DEVELOPMENT DURING SMOOTH
MUSCLE CONTRACTION**


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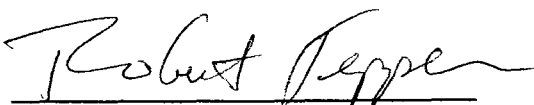

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ABSTRACT

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MECHANISMS FOR THE REGULATION OF CYTOSKELETAL ORGANIZATION AND TENSION DEVELOPMENT DURING SMOOTH MUSCLE CONTRACTION

There is growing evidence that actin polymerization and dynamic remodeling of the actin cytoskeleton play critical roles in the regulation of the physiologic properties of smooth muscle. The objective of these studies was to evaluate mechanisms for actin polymerization and cytoskeletal remodeling in smooth muscle tissues. Plasmids encoding target proteins or protein fragments were introduced into smooth muscle tissues, and tissues were incubated for two days to allow for protein expression. The role of the cytoskeletal protein α -actinin in the regulation of actin-integrin linkages during smooth muscle contraction was evaluated. The rod domain of α -actinin was expressed in smooth muscle tissues to compete with endogenous α -actinin for binding to β 1 integrin and thereby disrupt the connections between actin filaments and β integrins. Expression of the rod domain of α -actinin inhibited the redistribution of α -actinin to the cell membrane, inhibited its interaction with β integrin in response to ACh, and depressed active tension. These results suggest α -actinin contributes to the formation of connections between the integrin proteins and actin filaments required to transmit tension between the contractile apparatus and the extracellular matrix during smooth muscle contraction. In the second study, the roles of N-WASp and the Arp2/3

complex in regulating actin polymerization in smooth muscle in response to extracellular stimuli were determined. The CA domain of N-WASp was expressed in muscle strips to inhibit the activation of the Arp2/3 complex by N-WASp. Expression of the CA domain inhibited actin polymerization and tension development in response to ACh, but did not affect MLC phosphorylation. The association of the Arp2/3 complex with N-WASp in response to ACh was inhibited by the CA domain, as assessed by immunofluorescence and immunoprecipitation. These results suggest N-WASp-mediated activation of the Arp2/3 complex is necessary for agonist-induced actin polymerization and tension development in smooth muscle. In conclusion, the actin polymerization initiated by contractile stimulation in smooth muscle is mediated by N-WASp and the Arp2/3 complex, and contractile stimulation regulates connections between actin filaments and integrin proteins by α -actinin that enable the tension developed by contractile apparatus be transmitted from the contractile apparatus to the extracellular matrix.

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