

Interaction between a Blood Vessel-Inducing Protein Angiogenin and  
Its Binding Protein Actin

Seung-Ho So, Byung-Cheol Ahn, Seung-Bum Paik, and Soo-Ik Chang  
Department of Biochemistry, College of Natural Sciences, Chungbuk  
National University, Cheongju 361-763, Korea

Bovine angiogenin (bAng) is a potent blood vessel inducing protein purified from cow milk. Fluorescence spectroscopy has been used to study the interaction of bAng with actin in 50 mM Tris-HCl, pH 7.5, and 1 mM CaCl<sub>2</sub> at 25 °C. Actin contains four tryptophans but bAng contains no tryptophans. A 50% decrease in intrinsic fluorescence accompanied formation of the bAng/actin complex. By contrast, the interaction of RNase A, a homologous protein to bAng, with actin results in about 10% quenching of the fluorescence. Fluorescence titration experiments were performed by adding increasing concentrations of bAng (0-1.0 μM) to a constant concentration of actin (0.1 μM), and the dissociation constant K<sub>d</sub> for the bAng/actin complex and the stoichiometry n were measured as 20 ± 1 nM and 1.0 ± 0.1, respectively. These results suggest that the interaction between bAng with actin is specific and that quenching of actin fluorescence has occurred in the bAng/actin complex. The bAng binding sites of actin are discussed in the results of this study, and we propose that Trp-80 in the small domain of bovine actin is responsible for the bAng/actin binding.