Figure S5. Rho-dependent pyknosis requires the effector-interacting sequences in Rho loop6. A. Rho-induced pyknosis and influence of Bcl-2 were measured as in figure 4. The ability of active Rho to increase pyknosis was eliminated by inclusion of Rho>Rac mutations in the effector binding loop6 region of RhoA i.e. using RhoA Q63N/D87V/D90A (“RhoA L63VA”). Means ± SEMs are shown (n = 3). † indicates a significant difference from control without RhoA Q63L and * indicates a significant difference from corresponding sample without Bcl-2, by t-test (P<0.05 or better). B. Glutamate-evoked pyknosis was not affected by 10-100 µM of Y-27632, an inhibitor of the loop6-dependent Rho effector Rock Means ± SEMs are shown (n = 3).

Figure S6. Scheme depicting the proposed role of Rho in regulation of p38α by glutamate. Glutamate acts on NMDA receptors (MK-801 sensitive) leading to calcium influx into the cell. Different stimuli (depolarization by elevating extracellular KCl) leading to calcium influx result in Rho activation and activation of p38α. Activation of p38α by glutamate depends on Rho as it is blocked by C3 toxin. The glutamate evoked apoptosis requires NMDA receptors, Rho and p38 as it is blocked by inhibitors of all these proteins as well as by Bcl-2. Rho activity is sufficient to activate this pathway as expression of activated Rho leads to p38 activation and apoptosis. This response depends on loop 6, one of the effector binding regions of Rho.