EFFECTS OF BLOCKING OF ENDOGENOUS RHO FAMILY GTP-BINDING PROTEINS ON MORPHOLOGY, ADHESION AND LOCOMOTION OF *AMOEBA PROTEUS*

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Rho family GTP-binding proteins are known to control cellular processess associated with actin-based cytoskeleton such as cell migration, cytokinesis, endocytosis and exocytosis or muscle contraction [1]. While there is a number of data on the role of these proteins in higher Eukaryota, the studies on protozoans are practically limited to *Dictyostelium* that does not contain Rho-like and Cdc42-like proteins and *Acanthamoeba* which myosin I and actin have been found to be regulated by Rac1 and Cdc42 [2, 3]. By blocking of endogenous Rho family proteins of highly motile *Amoeba proteus* by the specific RhoA inhibitor, C3 transferase, and antibodies against human RhoA and Rac1 we tried to assess the *in vivo* effect of Rho-like proteins on amoeba morphology, locomotion and adhesion. Rho- and Rac-like proteins co-localize with F-actin, and are rather evenly distributed through the cytoplasm with more pronounced accumulation in the uroid part and in the middle-anterior body region corresponding to the adhesion zone of migrating cells. Blocking of Rac-like protein(s) resulted in significant inhibition of cell migration. Microinjected amoebae flattened, strongly adhered to the glass surface and developed few wide pseudopods that seemed to be more dense than of control cells. Microinjecting with anti-RhoA antibodies led to cells rounding up and producing numerous small hyaline protrusions that were not able to attach to the surface and were relatively quickly retracting. Surprisingly the adhesion of the entire cell body was even stronger than of the control cells. Amoebae exhibited a kind of atypical, apparent imperceptible locomotion that was statistically inhibited by about 60% in comparison with control cells. After treatment with C3 transferase cells rapidly contracted and almost completely rounded up, became refractile with the granules centrally beat into a dense mass. Within several minutes amoebae detached from the surface and died. These results indicate that Rho family-based regulation of *A. proteus* actin cytoskeleton plays the crucial role in this protozoan functions such as cell migration, adhesion to the substratum and pseudopod formation.

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