Abstract

Protein CAS is a major tyrosine-phosphorylated protein in cells transformed by *v-crk* and *v-src* oncogenes. It is a multidomain adaptor protein, which serves as a scaffold for assembly of signalling complexes which are important for migration and invasiveness of Src-transformed cells. A novel phosphorylation site in N-terminal SH3 domain was identified – tyrosine 12 located on binding surface of CAS SH3 domain. To study biological importance of tyrosine 12 phosphorylation, non-phosphorylable (Y12F) and phosphomimicking (Y12E) mutant of CAS were prepared. We found that phosphomimicking mutation Y12E leads to decreased interaction of CAS SH domain with kinase FAK a phosphatase PTP-PEST and also reduce tyrosine phosphorylation of FAK. Using GFP-tagged CAS protein, we show that Y12E mutation caused delocalization of CAS from focal adhesion but has no effect on localization of CAS to podosome-type adhesion. Non-phosphorylable mutation Y12F cause hyperphosphorylation of CAS substrate domain and decrease turnover of focal adhesion and associated cell migration of mouse embryonal fibroblasts (MEFs) independent to integrin singalling. Analogically to migration, CAS Y12F decrease invasiveness of Src-transformed MEF. The results of this diploma thesis show that phosphorylation of Tyr12 in CAS SH3 domain is critically important for focal adhesion dynamics and associated migration and invasiveness of MEF.