Phosphorylation of cortactin by cyclin-dependent kinase 5 modulates actin bundling by the dynamin 1-cortactin ring-like complex and formation of filopodia and lamellipodia in NG108-15 glioma-derived cells

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Abstract. Dynamin copolymerizes with cortactin to form a ring-like complex that bundles and stabilizes actin filaments. Actin bundle formation is crucial for generation of filopodia and lamellipodia, which guide migration, invasion, and metastasis of cancer cells. However, it is unknown how the dynamin-cortactin complex regulates actin bundle formation. The present study investigated phosphorylation of cortactin by cyclin-dependent kinase 5 (CDK5) and its effect on actin bundle formation by the dynamin-cortactin complex. CDK5 directly phosphorylated cortactin at T145/T219 *in vitro*. Phosphomimetic mutants in which one or both of these threonine residues was substituted by aspartate were used. The three phosphomimetic mutants (T145D, T219D and T145DT219D) had a decreased affinity for F-actin. Furthermore, electron microscopy demonstrated that these phosphomimetic mutants could not form a ring-like complex with dynamin 1. Consistently, the dynamin 1-phosphomimetic cortactin complexes exhibited decreased actin-bundling activity. Expression of the phosphomimetic mutants resulted in not only aberrant lamellipodia and short filopodia but also cell migration in NG108-15 glioma-derived cells. These results indicate that phosphorylation of cortactin by CDK5 regulates formation of lamellipodia and filopodia by modulating dynamin 1/cortactin-dependent actin bundling. Taken together, these findings suggest that CDK5 is a potential molecular target for anticancer therapy.