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THE C-TERMINAL PEPTIDE OF H-RAS AS A TARGET FOR THE COVALENT BINDING OF DRUGS MODULATING RAS-DEPENDENT PATHWAYS

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Ras proteins are crucial players in differentiation and oncogenesis and thus constitute important drug targets. The localization and activity of Ras proteins are highly dependent on PTMs at their C-termini. In addition to an isoprenylated cysteine, H-Ras, but not other Ras proteins, possesses two cysteine residues (Cys181 and Cys184) in the C-terminal hypervariable domain that act as palmitoylation sites in cells. Therefore, the enzymes responsible for these modifications of H-Ras and the target residues are the subject of intense study as sites for therapeutic intervention. Here we describe the potential of several endogenous and exogenous small molecules to bind to a synthetic peptide from the hypervariable domain of H-Ras proteins. Cyclopentenone prostaglandins (cyPG) are reactive lipidic mediators that may bind covalently to proteins and activate H-Ras dependent pathways. Our results indicate that the dienone cyPG 15-deoxy-$\Delta^{12,14}$-PGJ_2 and Δ^{12}-PGJ_2 can bind simultaneously Cys181 and Cys184 of H-Ras *in vitro*, thus possibly inducing important conformational changes of the hypervariable domain. In contrast, single enone cyPG bind the two cysteines independently, whereas cyclopentenone, which lacks the lateral chains of the PG, is a very poor modifier of this peptide and does not activate Ras-dependent pathways in cells. Interestingly, among several small molecules currently being tested, phenylarsine oxide, a widely used tyrosine phosphatase inhibitor, effectively binds H-Ras hypervariable domain. These observations open new perspectives for the study of molecules potentially modulating Ras-dependent pathways.