# S35. Uncovering Novel Targets for Cancer Chemoprevention

E. Dmitrovsky[1,4,5], K.H. Dragnev[1,5], Q. Feng[4], Y. Ma[4], S.J. Shah[4], C. Black[3,5], V. Memoli[3,5], W. Nugent[2], J.R. Rigas[1,5], K. Sutisak[4], S. Freemantle[4]

*Departments of [1]Medicine, [2]Surgery, [3]Pathology and [4]Pharmacology and Toxicology, and [5]Norris Cotton Cancer Center, Dartmouth Medical School, Hanover, NH, 03755 and Dartmouth-Hitchcock Medical Center, Lebanon, NH 03756, United States of America*

Tobacco carcinogen-treatment of immortalized human bronchial epithelial (HBE) cells uncovered novel targets for cancer chemoprevention. Studies with all-trans-retinoic acid and other chemopreventive agents highlighted D-type and E-type cyclins as molecular targets. Aberrant expression of G1 cyclins is reported in bronchial pre-malignancy and lung cancers, implicating these species as targets for clinical cancer chemoprevention. To better understand roles of D-type cyclins in chemoprevention, pre-clinical studies and clinical trials were undertaken. Chemoprevention of HBE cells by retinoids and other agents was linked to induced proteasomal degradation of cyclin D1. Threonine 286 mutation of cyclin D1 stabilized this protein, implicating a phosphorylation event in this chemoprevention. A phospho-specific anti-cyclin D1 antibody that recognized phosphorylation changes at this residue confirmed this hypothesis. Glycogen synthase kinase inhibitors revealed a role for this kinase in the post-translational regulation of cyclin D1, but not other D-type cyclins. Transcriptional and post-transcriptional mechanisms were engaged to alter stability of individual D-type cyclins. A role for these cyclins in chemoprevention was highlighted using small interfering RNAs (siRNAs) that targeted individual D-type cyclins. Gene profiling experiments were conducted using carcinogen-transformed and chemoprevented HBE cells. These as well as biochemical and immunohistochemical studies highlighted the E1-like ubiquitin-activating enzyme (UBE1L) in retinoid regulation of cyclin D1. Treatment with the epidermal growth factor (EGF) stimulated HBE growth and cyclin D1 expression and an epidermal growth factor receptor (EGFR) tyrosine kinase inhibitor (TKI) was used to block these EGF-mediated effects. Findings were extended into the clinic through proof of principle trials with agents that targeted D-type cyclins. These trials proved useful to monitor changes in pharmacological targets in pre-treatment and post-treatment tumor biopsies, and to relate pharmacodynamic changes to intratumoral and plasma drug levels. Studies were built upon using a targeted combination regimen with agents that independently affected D-type cyclin expression. Taken together, pre-clinical and clinical findings that will be presented strongly implicate cyclin D1 as a novel molecular pharmacological target for cancer chemoprevention.