

DSPP-silencing in oral cancer cells suppresses SIBLING-partnering MMPs and VEGF

Shivani Patel

The SIBLINGs (Small integrin-binding ligand N-linked glycoproteins), which includes dentin sialophosphoprotein (DSPP), have emerged as molecular tools facilitating various stages of cancer progression. DSPP upregulation in human oral squamous cell carcinomas (OSCC) is associated with tumor aggressiveness. Here we investigate the effects of DSPP-silencing on the profiles of MMP-2, MMP-3, and MMP-9 known to interact with and activate specific SIBLINGs along with the profile of the proangiogenic factor, VEGF, known to play major roles in oral cancer progression. Stable lines of DSPP-silenced oral cancer cells, OSC2, were established following lentiviral-mediated DSP-shRNA interference, and the status of MMP-2, MMP-3, MMP-9, and VEGF investigated by western blot analysis. Controls consisted of parental OSC2 cells and stable lines of scrambled sequence. Quantitation of western blot and Least Square Regression analyses showed significantly reduced levels of pro- and activated MMP-2 $\{(y=0.850x, p<0.001) (y= 1.156x, p<0.001)\}$, MMP-3 $\{(y= 0.0.994x, p<0.001) (y= 1.324x, p= 0.004)\}$, and MMP-9 $\{(y= 1.248x, p= 0.005, y= 0.809, p= 0.013)\}$ with significantly reduced levels of VEGF following DSPP-silencing, compared to controls. Equally significant is a direct correlation between the degree of DSPP-silencing and MMP suppression; however, the level of MMP suppression did not differ significantly between pro- and cleaved forms. The down-regulation of SIBLING-partnering MMPs and VEGF in DSPP-silenced OSC2 cells suggests these as downstream targets of DSPP, and that DSPP may play a key regulatory role in oral carcinogenesis via mechanisms involving MMPs and VEGF. These findings provide a framework for studying the mechanisms of DSPP-MMP interaction in oral carcinogenesis.