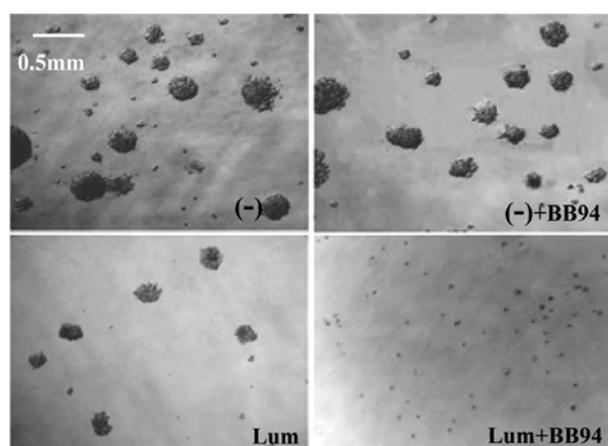


## Cleavage of Lumican by Membrane-Type Matrix Metalloproteinase-1 Abrogates This Proteoglycan-Mediated Suppression of Tumor Cell Colony Formation in Soft Agar

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The small leucine-rich proteoglycan lumican was identified from a human placenta cDNA library by the expression cloning method as a gene product which interacts with membrane-type matrix metalloproteinase-1 (MT1-MMP). Co-expression of MT1-MMP with lumican in HEK293T cells reduced the concentration of lumican secreted into culture medium, and this reduction was abolished by addition of the MMP inhibitor BB94. Lumican protein from bovine cornea and recombinant lumican core protein fused to glutathione S-transferase were shown to be cleaved at multiple sites by recombinant MT1-MMP. Transient expression of lumican in HEK293 cells induced expression of tumor suppressor gene product p21/Waf-1, which was abrogated by the co-expression of MT1-MMP concomitant with a reduction in lumican concentration in culture medium. Stable expression of lumican in HeLa cells induced expression of p21 and reduction of colony formation in soft agar, which were both abolished by the expression of MT1-MMP. HT1080 fibrosarcoma cells stably transfected with the lumican cDNA (HT1080/Lum), which express endogenous MT1-MMP, secreted moderate levels of lumican, however, treatment of HT1080/Lum cells with BB94 resulted in accumulation of lumican in culture medium. The expression levels of p21 in HT1080/Lum were proportional to the concentration of secreted lumican, and showed reverse co-relation with colony formation in soft agar. These results suggest that MT1-MMP abrogates lumican-mediated suppression of tumor cell colony formation in soft agar by degrading this proteoglycan which down-regulates it through the induction of p21.



**Fig. 1.** Degradation of lumican by HT1080 cells enhances colony formation in soft agar. Control HT1080 (-) or HT1080 cells transfected with lumican plasmid (Lum) were plated into 0.3% agarose with or without 0.1  $\mu$ M BB94, and colonies were observed under microscopy 2-weeks after incubation.