

## Suppression of MT1-MMP-Mediated MMP-2 Activation and Tumor Invasion by Testican 3 and its Splicing Variant Gene Product N-Tes.

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Using expression cloning to screen a human fetal kidney cDNA library for regulator(s) of pro-MMP-2 processing mediated by membrane-type-1 matrix metalloproteinase (MT1-MMP), we isolated a cDNA whose product interfered with pro-MMP-2 activation. It encodes the N-terminal 313 amino acids region of a calcium-binding proteoglycan, testican 3 with a 3 amino acids substitution at the C terminus, and thus was named N-Tes. N-Tes comprises a signal peptide, a unique domain, a follistatin-like domain and a  $Ca^{2+}$  binding domain, but lacks C-terminal thyroglobulin domain and two putative glycosaminoglycan attachment sites of testican 3 (1). Pro-MMP-2 activation by MT3-MMP was also inhibited by the co-expression of N-Tes. Immunoprecipitation analysis demonstrated direct interaction of N-Tes with either MT1-MMP or MT3-MMP. Expression of testican 1 or testican 3 but not testican 2 also inhibited pro-MMP-2 activation by either MT1-MMP or MT3-MMP. Deletion and substitution of amino acids residues in N-Tes revealed that the unique N-terminal domain of N-Tes is responsible for the inhibition of pro-MMP-2 activation by MT-MMPs. Expression of N-Tes and testican 3 was detected in normal brain, but was down-regulated in glioma tissues. Transfection of either N-Tes or testican 3 gene into U251 glioma cells or MDCK cells transformed by erbB2 suppressed their invasive growth in collagen gel. These results suggest that both N-Tes and testican 3 would interfere with tumor invasion by inhibiting MT-MMPs.

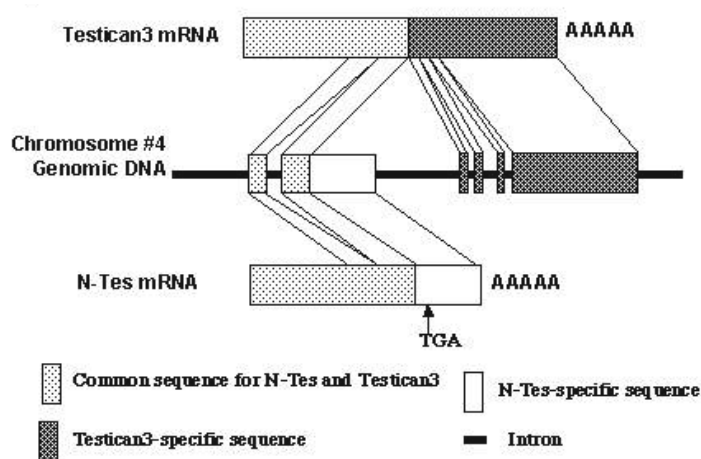


Fig. 1. Genome construction of N-Tes and testican 3 genes.

### Reference:

Nakada, M., Yamada, A., Takino, T., Miyamori, H., Takahashi, T., Yamashita, J., and Sato, H. (2001) Suppression of MT1-MMP-mediated MMP-2 activation and tumor invasion by testican 3 and its splicing variant gene product N-Tes. *Cancer Res.*, 61, 8896-8902.