Purification and Characterization of Membrane-Type Matrix Metalloproteinase-1

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Membrane-type matrix metalloproteinase-1 (MT-MMP-1) is a new member of the MMP gene family and activates proMMP-2 on the cell surfaces. We purified a deletion mutant Δ536-582 of MT-MMP-1 lacking the COOH-terminal transmembrane domain (ΔMT-1) from the culture media of the stable transfectants. ΔMT-1 was secreted in a complex form with TIMP-2 in the media and the complex was separated by anti-TIMP-2 IgG-Sepharose column. The proteinase showed a new NH2-terminus of Ala113-Ile-Gln-Gly-Leu-Lys-Try, indicating the cleavage at the Tyr112-Ala113 bond. Transfection of a mutant ΔMT-1 in which RRKR sequence in the first insertion was replaced with the furin-resistant site failed in processing, suggesting that the furin-recognition site is required for removal of the prodomain. The proteinase degraded extracellular matrix (ECM) components including gelatins, type I, II, III collagens, proteoglycan, fibronectin and vitronectin. A human breast carcinoma cell line (MDA-MB-231) also secreted MT-MMP-1 in a complex form with TIMP-2 into the media and it was also purified to homogeneity. The native MT-MMP-1 showed ECM-degrading activity similar to ΔMT-1. These data indicate that MT-MMP-1 is an ECM-degrading enzyme and suggest that the proteinase may play an important role in tissue destruction by directly cleaving the ECM components as well as through activation of proMMP-2.