Cleavage of Apolipoprotein E by Membrane-Type Matrix Metalloproteinase-1 Abrogates Suppression of Cell Proliferation

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Apolipoprotein E (apoE) in a human fetal brain cDNA library was identified, using the expression cloning method, as a gene product that formed a complex with latent matrix metalloproteinase (MMP)-2. Co-expression of membrane-type MMP-1 (MT1-MMP) with apoE in HEK293T cells reduced the amount of apoE secreted into the culture medium, whereas cell-associated apoE core protein was not affected. Incubation of native apoE protein with recombinant MT1-MMP resulted in the cleavage of apoE. Recombinant apoE protein fused to glutathione S-transferase (apoE-GST) was cleaved by MT1-MMP at the following peptide bonds; T^{85}-M^{86}, K^{93}-S^{94}, R^{246}-L^{247}, A^{255}-E^{256} and G^{296}-L^{297}. HT1080 cells transfected with the apoE gene, which express endogenous MT1-MMP, secreted a low level of apoE protein and its cleaved fragments, and treatment with MMP inhibitor BB94 induced accumulation of apoE and retardation of cell proliferation. Addition of apoE-GST protein to the culture of HEK293T cells suppressed cell proliferation, and stable transfection of the MT1-MMP gene partly abrogated the suppression. These results suggest that cleavage of apoE protein by MT1-MMP abrogates apoE-mediated suppression of cell proliferation.

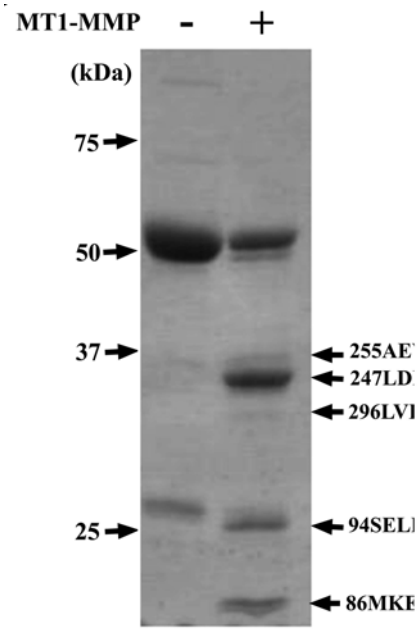

Fig. 1. Cleavage of apoE by MT1-MMP. ApoE-GST fusion protein (5 μg) was incubated with recombinant MT1-MMP catalytic domain (0.5 μg), separated on 15% SDS-polyacryamide gel, and then blotted onto PDF membrane. The N-terminal amino acid sequence of each fragment was determined with a Beckman Coulter LF300 amino acid sequencer.