

Supplementary Materials

Table S1. Primer sets used for the construction of plasmid DNA.

| Primer Set | Sequences |
|----------------------|---------------------------------------------------------------------------|
| Asn127Ala | 5'-ACTACATTAGAGCATGCATCATTGGT-3' 5'-ACCAATGATGCATGCTCTAATGTAGT-3' |
| Val140Ala | 5'-TACAAGGGAACAGCATCTATCACTAAGA-3' 5'-TCTTAGTG ATAGATGCTGTTCCCTTGTA-3' |
| Lys144Ala | 5'-TCTATCACTGCAAGTGGCATCA-3' 5'-TGATGCCACTTGCACTGATAGA-3' |
| NK1 fragment for HGF | 5'-ATGTGGGTGACCAAACCTCCT-3' 5'-TTCAACTTCTGAACACTGAGGAAT-3' |
| in-fusion cloning-1 | 5'-GACAATAACCCCTGATAAATGCTTC-3' 5'-CATTTATCAGGGTTATTGTCTCATG-3' |
| in-fusion cloning-2 | 5'-TTTGGTCACCCACATGGT-3' 5'-TG TTCAGAAAGTTGAATGCATGA-3' |

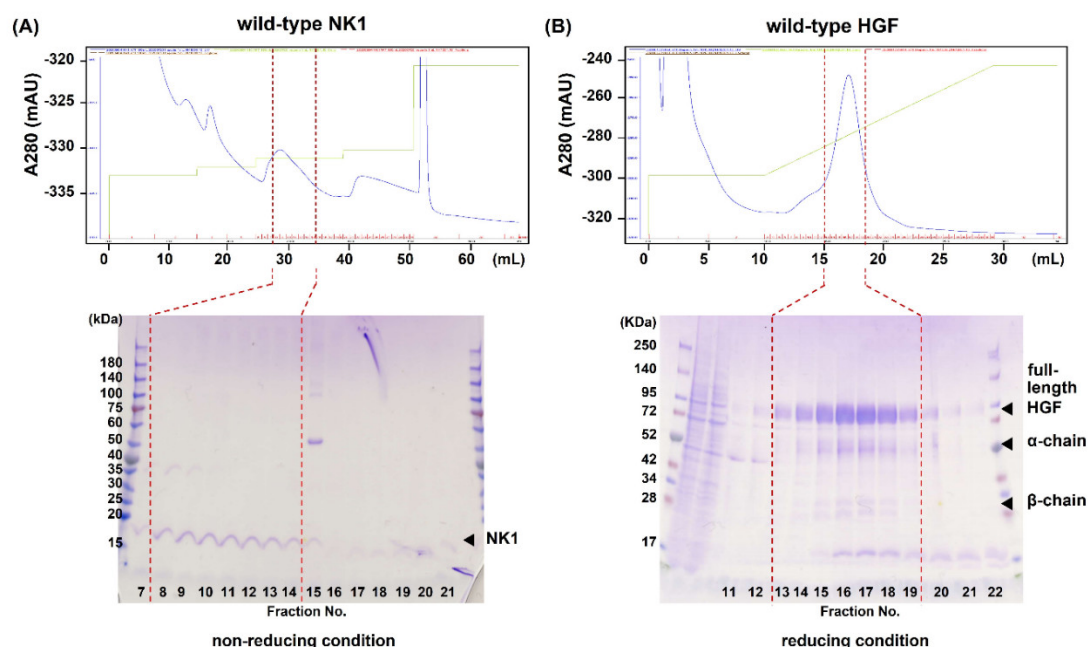


Figure S1. Elution profiles and purification of wild-type NK1 (A) and wild-type HGF (B) in heparin-affinity chromatography. CBB staining of proteins after SDS-PAGE is shown in the bottom panels. Elution profiles of mutant NK1 and HGF were similar to those of wild-type NK1 and HGF.

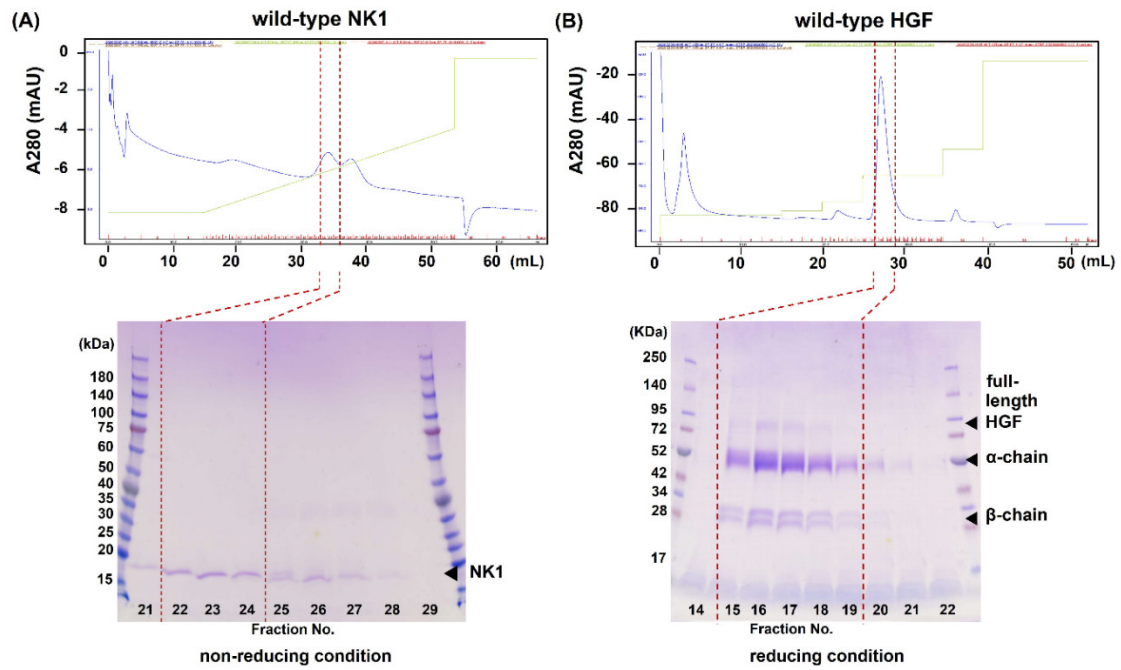


Figure S2. Elution profiles and purification of wild-type NK1 (A) and wild-type HGF (B) in cation exchange chromatography. CBB staining of proteins after SDS-PAGE is shown in the bottom panels. Elution profiles of mutant NK1 and HGF were similar to those of wild-type NK1 and HGF.

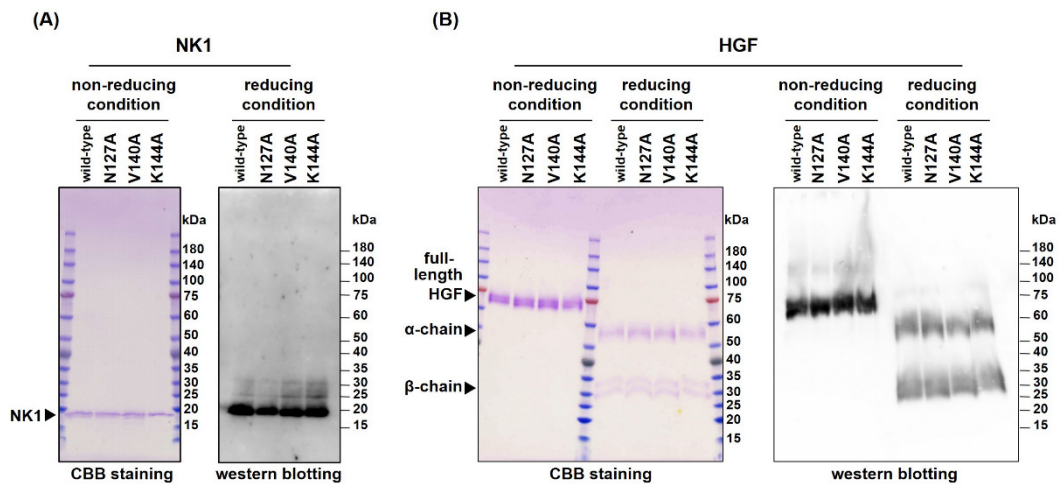


Figure S3. Recombinant NK1 (A) and HGF (B) confirmed by protein staining and Western blotting. Recombinant proteins were electrophoresed on a 5–20% gradient gel under non-reducing and reducing (by 2-mercaptoethanol) conditions. Proteins were stained with Coomassie Brilliant Blue (CBB) (left) and detected by Western blot analysis (right). Wild-type and mutant HGF show a single band and two bands in non-reducing and reducing conditions, respectively.

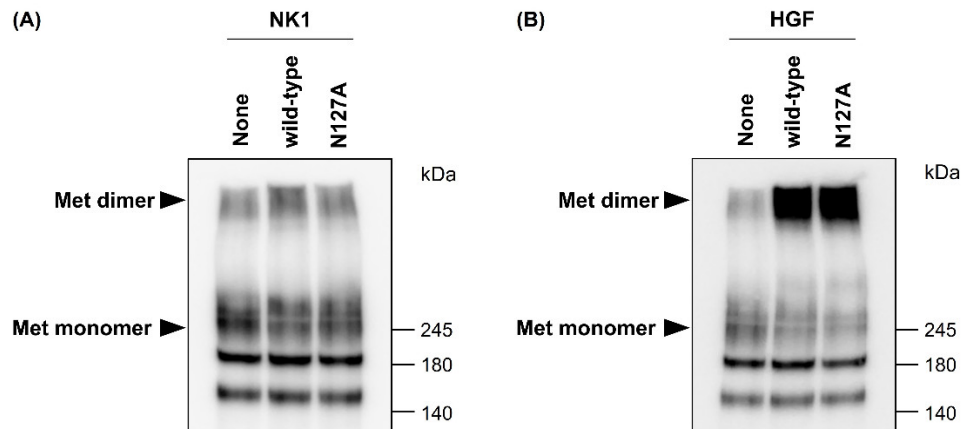


Figure S4. Met dimer formation promoted by NK1 and HGF. EHMES-1 cells were left untreated or treated with 30 nM wild-type and mutant NK1 or 1 nM wild-type and mutant HGF. Met dimer formation was analyzed by chemical cross-linking, immunoprecipitation, SDS-PAGE, and Western blotting.