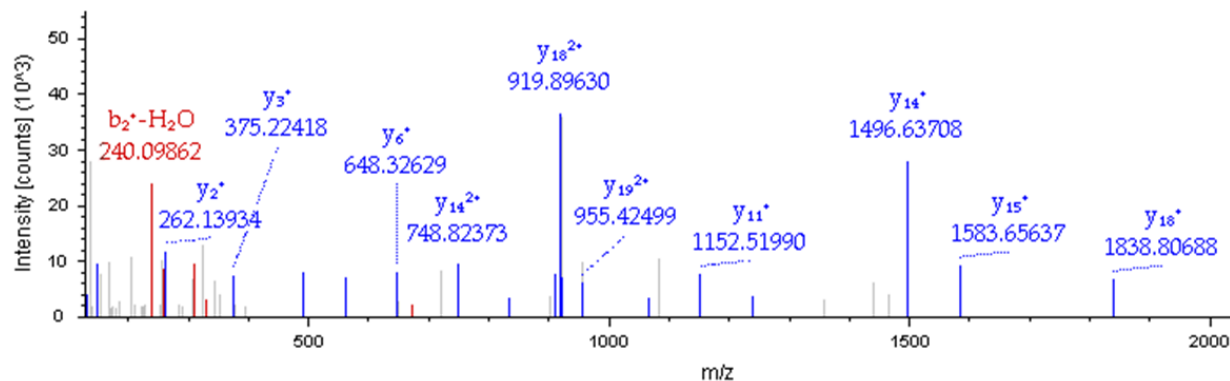


Figure S3: Identification of sFN14 derived tryptic peptides by immunoaffinity liquid chromatography tandem mass spectrometry (IA-LC-MS/MS).

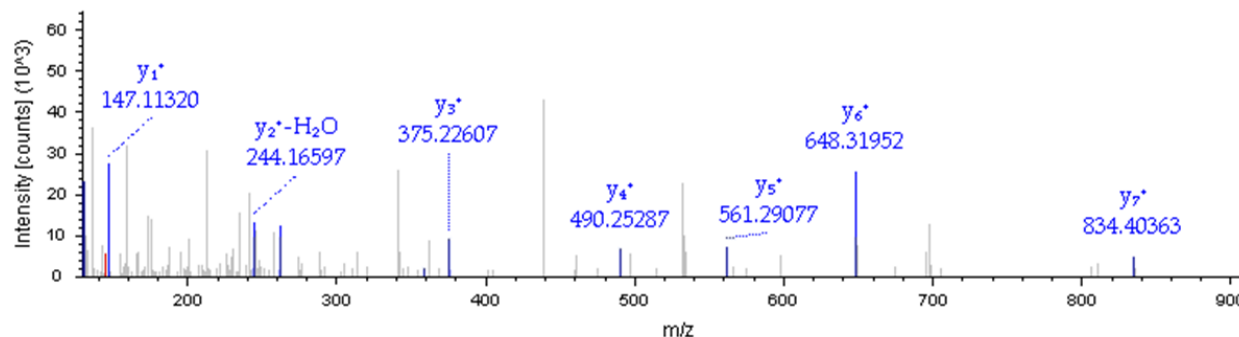
A Tryptic peptide of sFn14 in mouse serum

FTMS, HCD@27.00, z=+2, Mono m/z=1083.96582 Da, MH+=2166.92436 Da, Match Tol.=0.04 Da



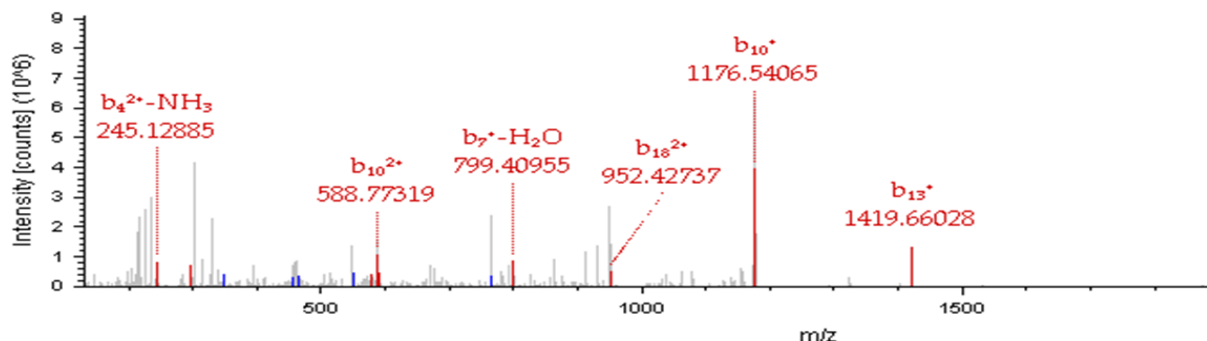
B Tryptic peptide of sFN14 in human serum

FTMS, HCD@27.00, z=+2, Mono m/z=533.24597 Da, MH+=1065.48467 Da, Match Tol.=0.04 Da



C Tryptic peptide of sFN14 in human serum

FTMS, HCD@27.00, z=+3, Mono m/z=783.39642 Da, MH+=2348.17472 Da, Match Tol.=0.04 Da



D Tryptic peptides of FN14 in urine of human DN samples

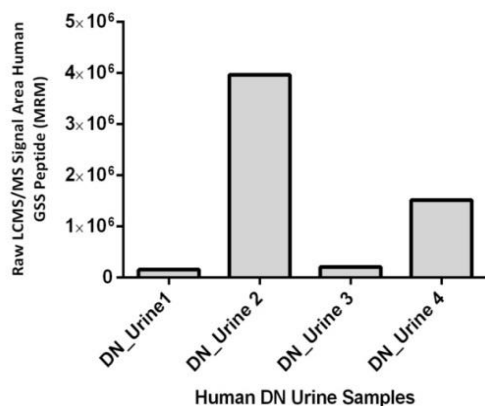


Figure S3: Tandem mass spectra of sFN14 derived tryptic peptides acquired following FN14 immunoaffinity enrichment and trypsin digestion. **(A)** Mouse Fn14 tryptic peptide: ²⁸EQAPGTSPC*SSGSSWSADLDK (*C carbamidomethylated, Uniprot ID Q9CR75) corresponding to the ectodomain of mouse Fn14. **(B)** Human FN14 tryptic peptide: ³⁹GSSWSADLDK (Uniprot ID Q9NP84), corresponding to the ectodomain of FN14; **(C)** Human FN14 tryptic peptide: ¹⁰⁸EKFTTPIIETGGEGC*PAVALIQ* (*C carbamidomethylated, *Q deamidated, Uniprot ID Q9NP84). Note this tryptic peptide originates from the cytoplasmic domain of FN14. **(D)** LCMS detection of Fn14 tryptic peptide (GSSWSADLDK) in the urine of human DN samples- Briefly 200ul of urine sample was incubated overnight with P4A8 antibody at 4°C and tryptic peptide measured by LCMS assay.