

EEQYNSTYR (1189.5120 Da)

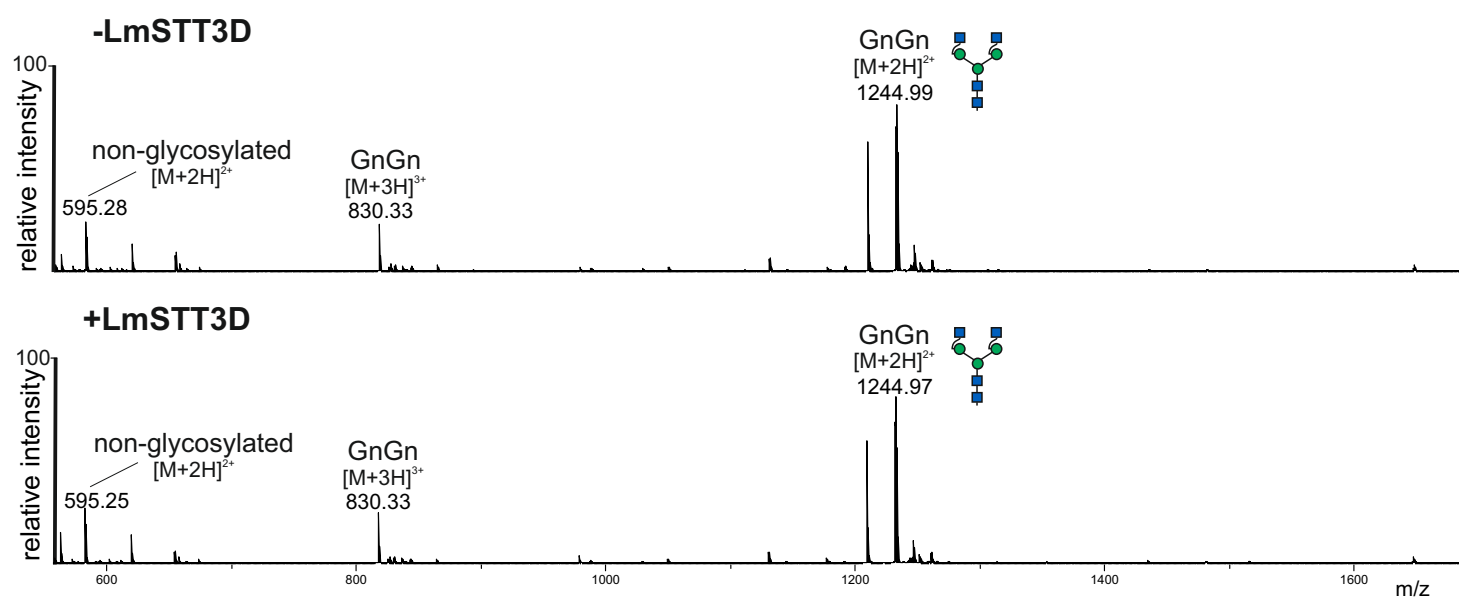


Figure S1 Mass spectra of (glyco)peptides from IgG. IgG was transiently expressed in *N. benthamiana* Δ XT/FT in the presence or absence of LmSTT3D-GFP. Purified IgG was trypsin digested and analysed by LC-ESI-MS.

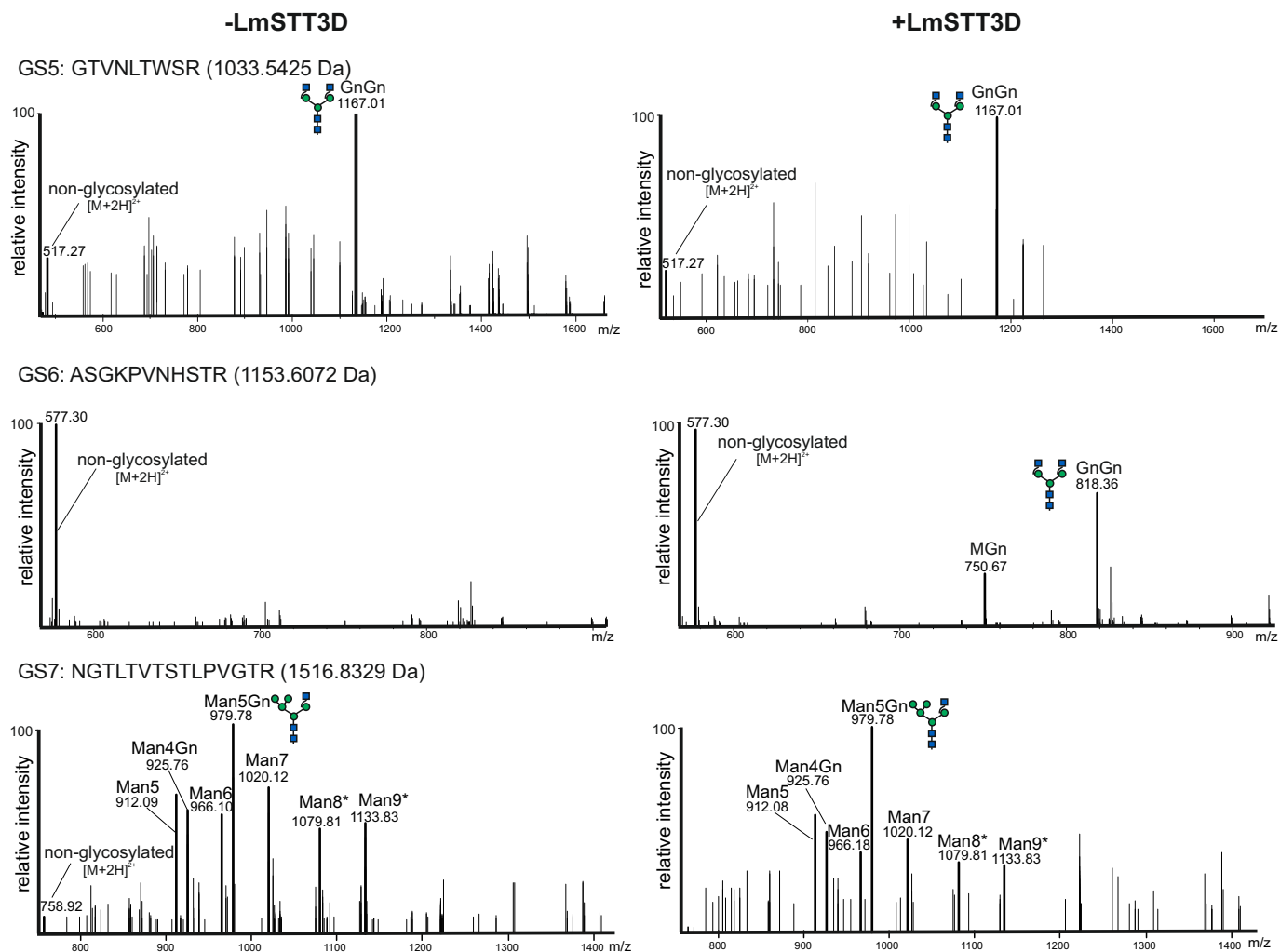


Figure S2 Mass-spectra of IgE (glyco)peptides harbouring glycosylation sites (GS) 5, 6 and 7. IgE was transiently expressed in *N. benthamiana* Δ XT/FT in the presence or absence of LmSTT3D-GFP. Purified IgE was trypsin digested and analysed by LC-ESI-MS. The mass range displays the major glycoforms. Except for the glycopeptides harbouring GS5, the peaks corresponding to triple charged glycopeptides ($[M+3H]^{3+}$) are indicated. The major glycoform on GS5 as well as all non-glycosylated peptides are shown in their double charged ($[M+2H]^{2+}$) state. Ammonia adducts are denoted by an asterisk.

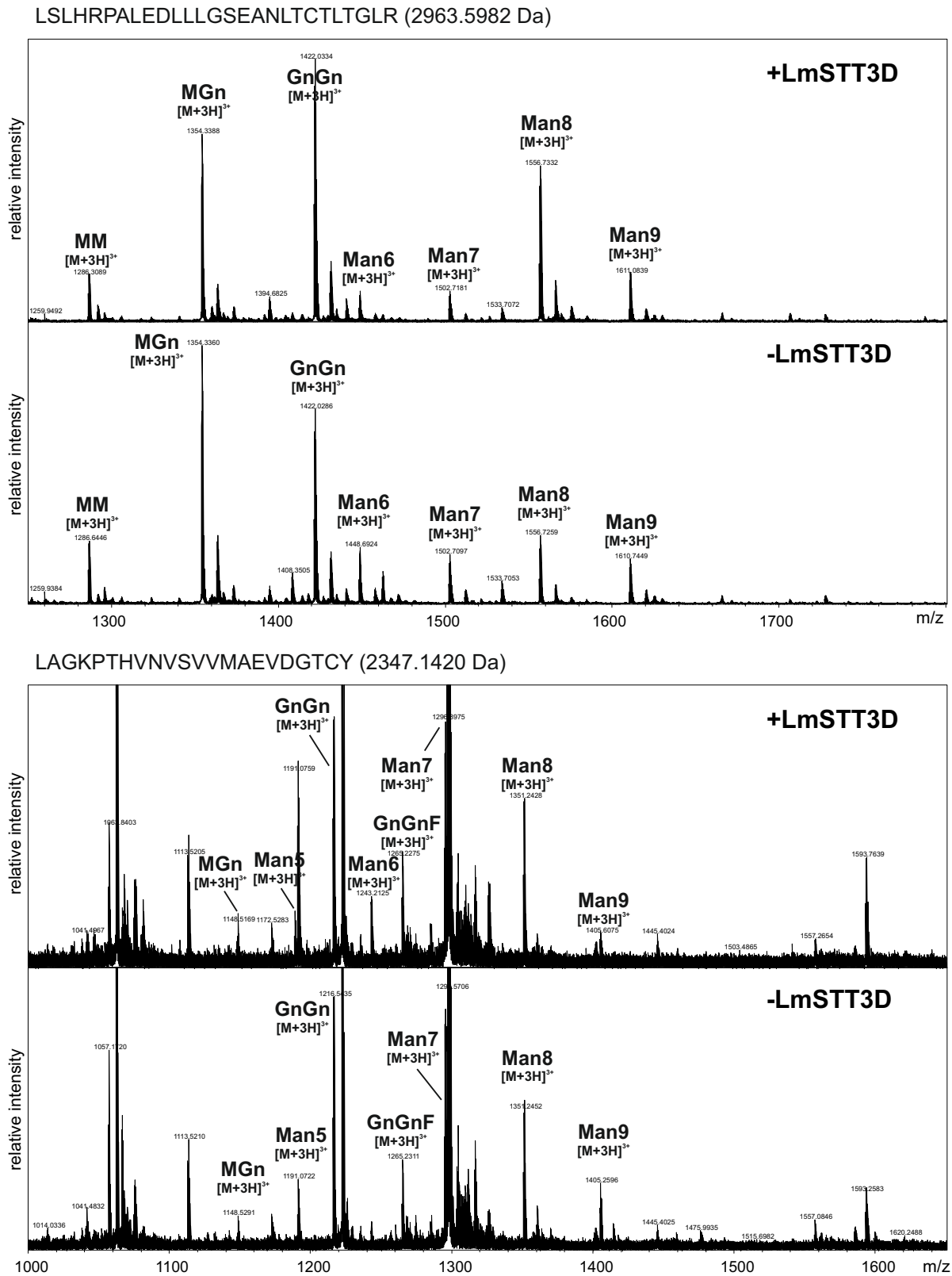


Figure S3 Mass-spectra of IgA1 (glyco)peptides. IgA1 was transiently expressed in *N. benthamiana* Δ XT/FT in the presence or absence of LmSTT3D-GFP. The purified IgA1 heavy chain was trypsin digested and analysed by LC-ESI-MS.

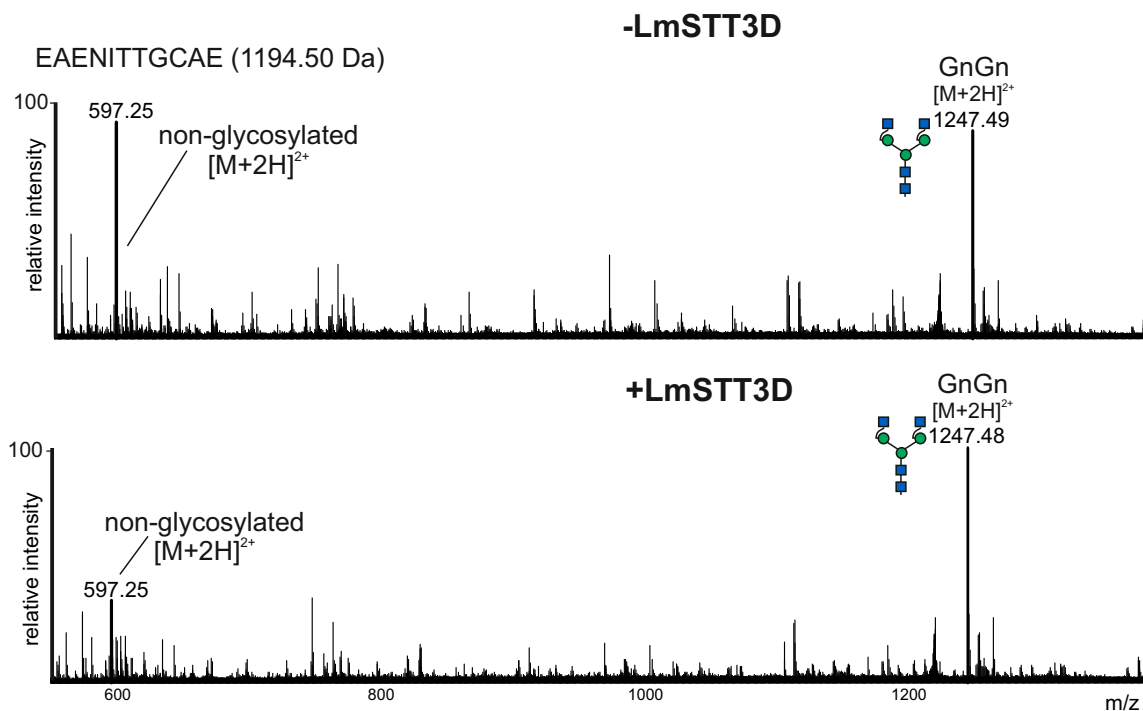
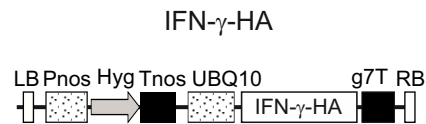


Figure S4 Mass-spectra of EPO-Fc (glyco)peptides. EPO-Fc was transiently expressed in *N. benthamiana* Δ XT/FT in the presence or absence of LmSTT3D-GFP. Purified EPO-Fc was trypsin+GluC digested and analysed by LC-ESI-MS. The mass range displaying the major glycoform from the peptide corresponding to glycosylation site 1 (due to the iodoacetamide treatment the cysteine is present as carbamidomethyl-cysteine) is shown.

(a)



(b)

MANKHLSLSLFLVLLGLSASLAQDPYVKEAENLKKYFNAGHSDVADNGTLFLGILKNWKEESDRKIMQ
SQIVSFYFKLFKNFKDDQSIQKSVETIKEDMNVKFFNSNKKKRDDFEKLTNYSVTDLNVQRKAIHELI
QVMAELSPAAGTGRKRKRSQMLFRGRRASQGSYPYDVPDYASLYPYDVPDYASLYPYDVPDYASL

Figure S5 Schematic illustration of the IFN- γ -HA expression construct (a) and the IFN- γ -HA amino acid sequence (b). (a) LB: left border; Pnos: nopaline synthase gene promoter; Hyg: hygromycin B phosphotransferase gene; Tnos: nopaline synthase gene terminator; UBQ10: *A. thaliana* ubiquitin-10 promoter; IFN- γ -HA: coding sequence of human interferon γ fused to a 3x hemagglutinin (HA) tag; g7T: agrobacterium gene 7 terminator; RB: right border. (b) Amino acid sequence of IFN- γ -HA. The signal peptide from barely alpha-amylase is marked in green. The two N-glycosylation sites (NGT and NYS) are marked in blue. A short dipeptide linker is marked in red and the 3x HA tag is highlighted in grey.