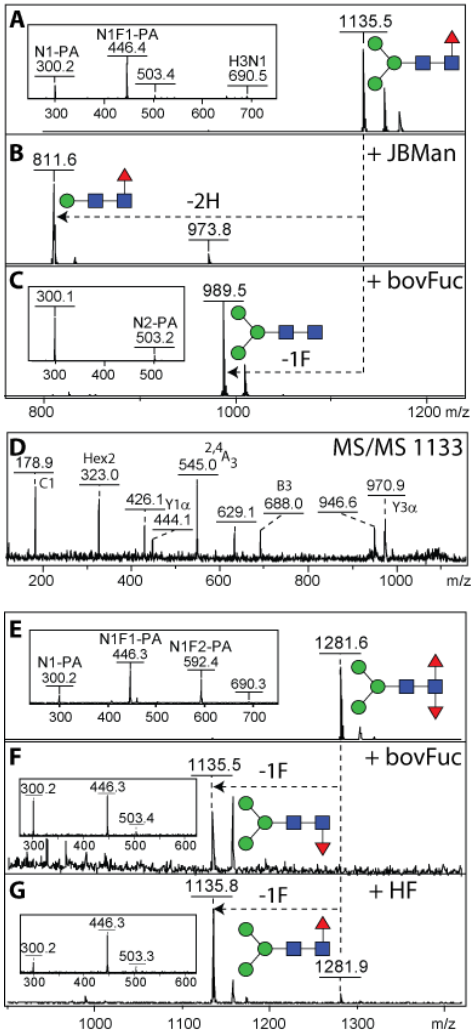


**Fig. S1** Analysis of fucosylated paucimannosidic N-glycans from *T. suis.* Two selected N-glycan fractions containing paucimannosidic structures were subject to α-mannosidase, α-fucosidase or hydrofluoric acid treatments. (A-C) Positive ion mode MALDI-TOF MS of the major $Man_3GlcNAc_2Fuc_1$-PA glycan (9.4 g.u.) before and after jack bean α-mannosidase or bovine α-fucosidase digestion, the latter resulting in a loss of the *m/z* 446 core fragment ion (see insets) and a shift to 7.2 g.u. on HPLC (i.e., co-elution with $Man_3GlcNAc_2$-PA); (D) negative ion mode MALDI-TOF MS/MS of the $Man_3GlcNAc_2Fuc_1$-PA glycan (*m/z* 1133 as $[M-H]^-$) showing different cleavages as those in positive mode, including a cross-ring *A* cleavage of the distal GlcNAc, in addition to a $HexNAc_1Fuc_1$-PA *Y1*α-fragment (*m/z* 444) comparable to the dominant *m/z* 446 fragment in positive mode. (E-G) Positive ion mode MALDI-TOF MS of the $Man_3GlcNAc_2Fuc_2$-PA glycan (6.8 g.u.) before and after bovine α-fucosidase or hydrofluoric acid treatment; the key core MS/MS fragments of *m/z* 300, 446 and 592 ($HexNAc_1Fuc_{0-2}$-PA) are shown in the insets and show different ratios of the *m/z* 300 and 446 fragment intensities in panels F and G depending on the remaining fucose linkage. Abbreviations for monosaccharides and their substitutions are: F, fucose; H, hexose; N, *N*-acetylhexosamine; PA, 2-aminopyridine.

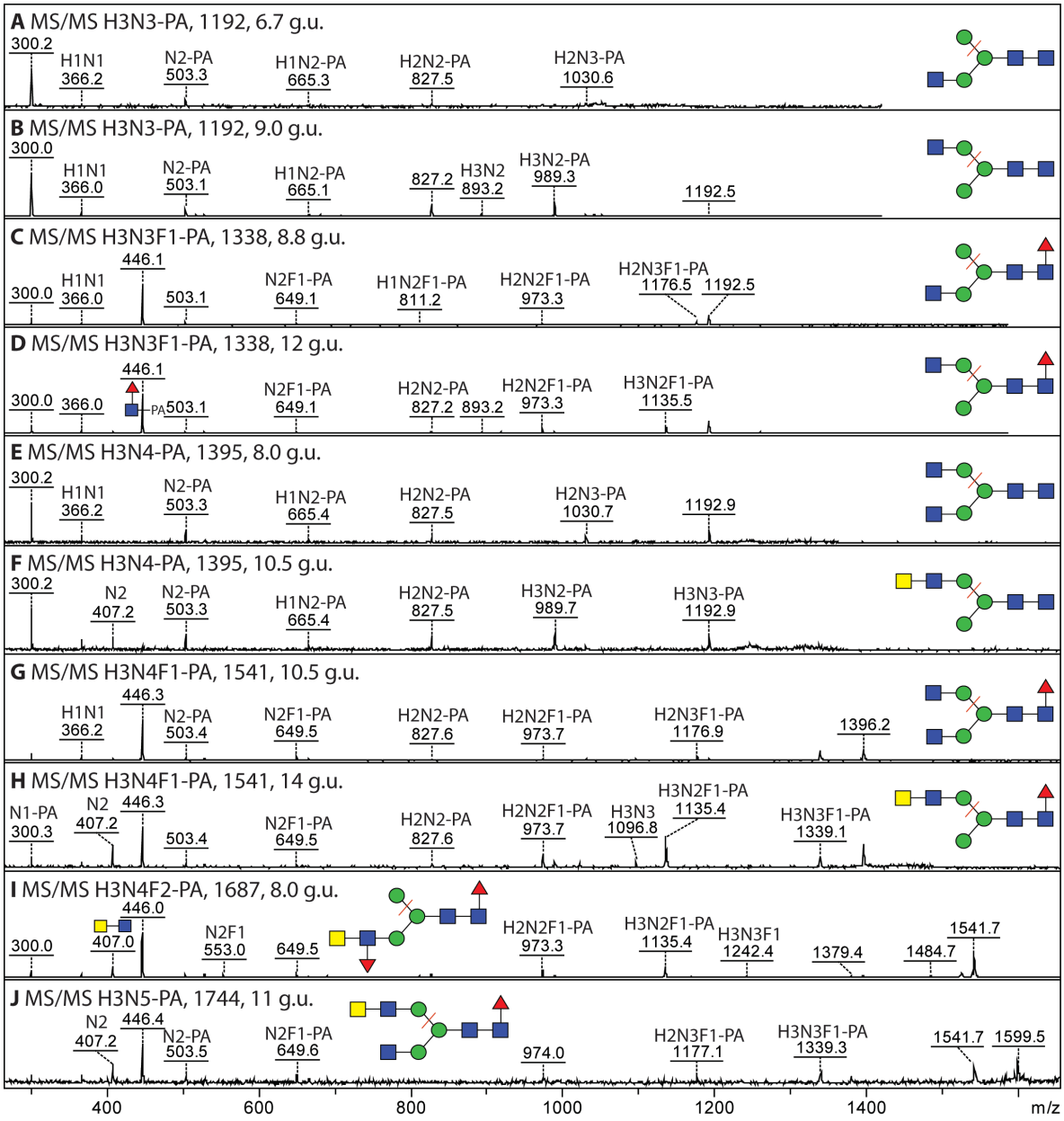


**Fig. S2** MS/MS analysis of N-glycans from *T. suis* with terminal *N*-acetylhexosamine residues. (A-J) Positive ion mode MALDI-TOF MS/MS of various glycans, including isomeric structures, eluting at the indicated glucose units on the RP-amide HPLC column. Key fragments are annotated with an abbreviated composition and the entire predicted structure is shown with a key fragmentation position (the α1,6-substitution of the core β-mannose; a *Y3* fragment) indicated as a red bar; this results in fragment ions of, e.g., *m/z* 827, 973, 1030 and 1176 whose occurrence and intensities depend on the structure and which can correlate with the substitution with HexNAc or HexdiNAc (presumed to be GlcNAc or LacdiNAc) on the upper or lower arms. All core fucosylated glycans show an MS/MS core fragment ion of *m/z* 446 ($HexNAc_1Fuc_1$-PA) of an intensity typical for α1,6-fucose. In some cases, the assignments are also supported by mass spectrometric and HPLC elution shifts after enzymatic digestion (see Figures 2 and 3 as well as Fig. S3).

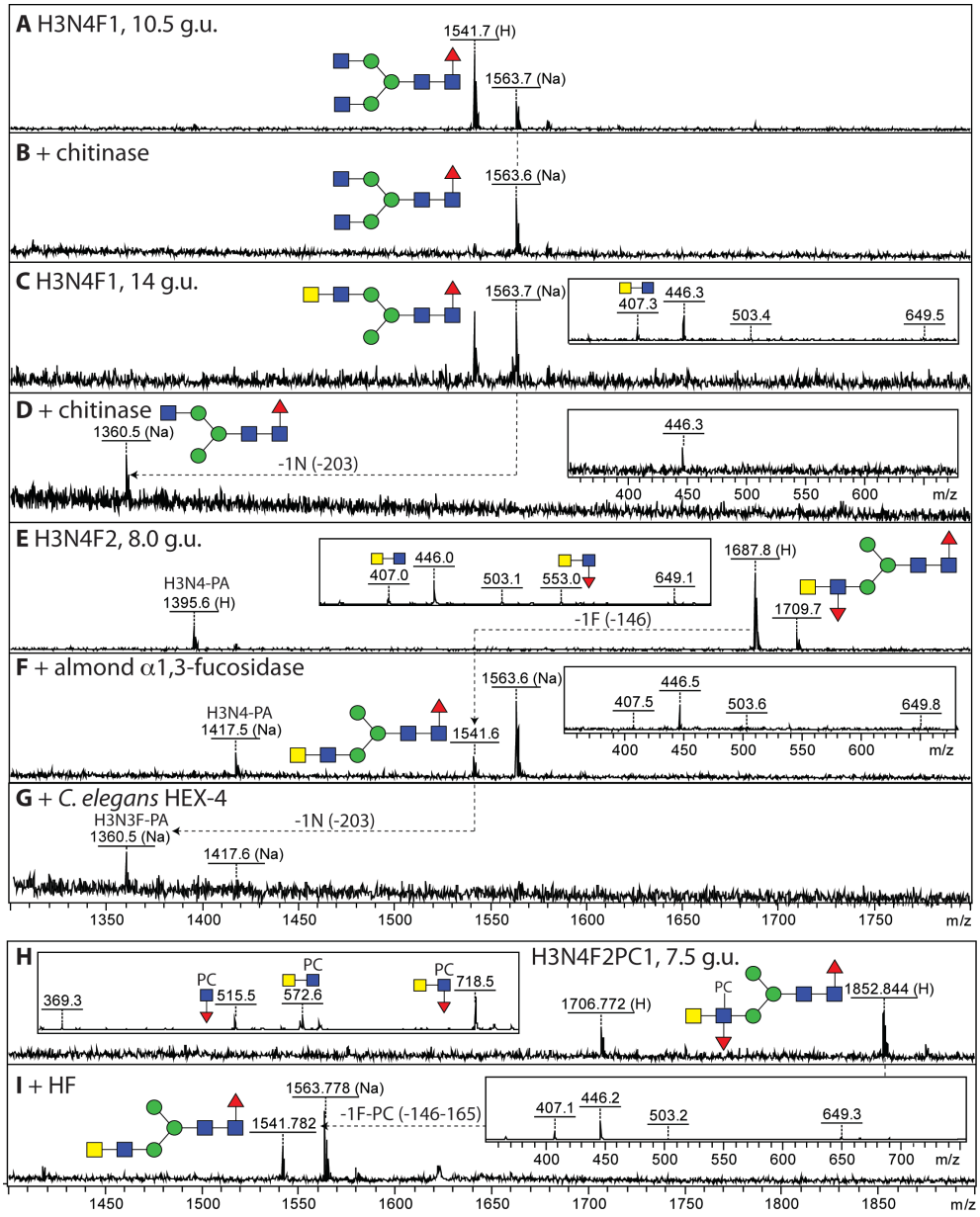


**Fig. S3** Enzymatic and chemical treatments of HexdiNAc-containing glycans from *T. suis*. (A-D) Two isobaric forms of $Hex_3HexNAc_4Fuc_1$ (10.5 and 14 g.u.) were incubated with a β1,3/4/6-specific hexosaminidase (chitinase) showing that only the latter was sensitive to this treatment resulting in the loss of the antennal *m/z* 407 HexdiNAc fragment ion (insets). (E, F, G) MALDI-TOF MS analysis of a difucosylated glycan (8.0 g.u.) before and after serial treatment with almond α1,3-fucosidase (resulting in the loss of the antennal *m/z* 553 $HexNAc_2Fuc_1$ fragment ion) and *C. elegans* GalNAc-specific HEX-4. (H, I) MALDI-TOF MS analysis of a difucosylated phosphorylcholine-modified glycan (7.5 g.u.) before and after treatment with hydrofluoric acid, which resulted in the loss of the antennal *m/z* 369, 515, 572 and 718 ($HexNAc_{1\text{-}2}Fuc_{0\text{-}1}PC_1$) *B*-fragment ions and the appearance of an *m/z* 407 ion ($HexNAc_2$) as well as core *Y*-fragment ions (*m/z* 446, 503 and 649; $HexNAc_{1\text{-}2}Fuc_{0\text{-}1}$-PA; see insets). The components in the enzymes used tended to result in a shift to sodiated adducts (annotated with Na); however, only the protonated quasimolecular ions (annotated with H) were fragmented.

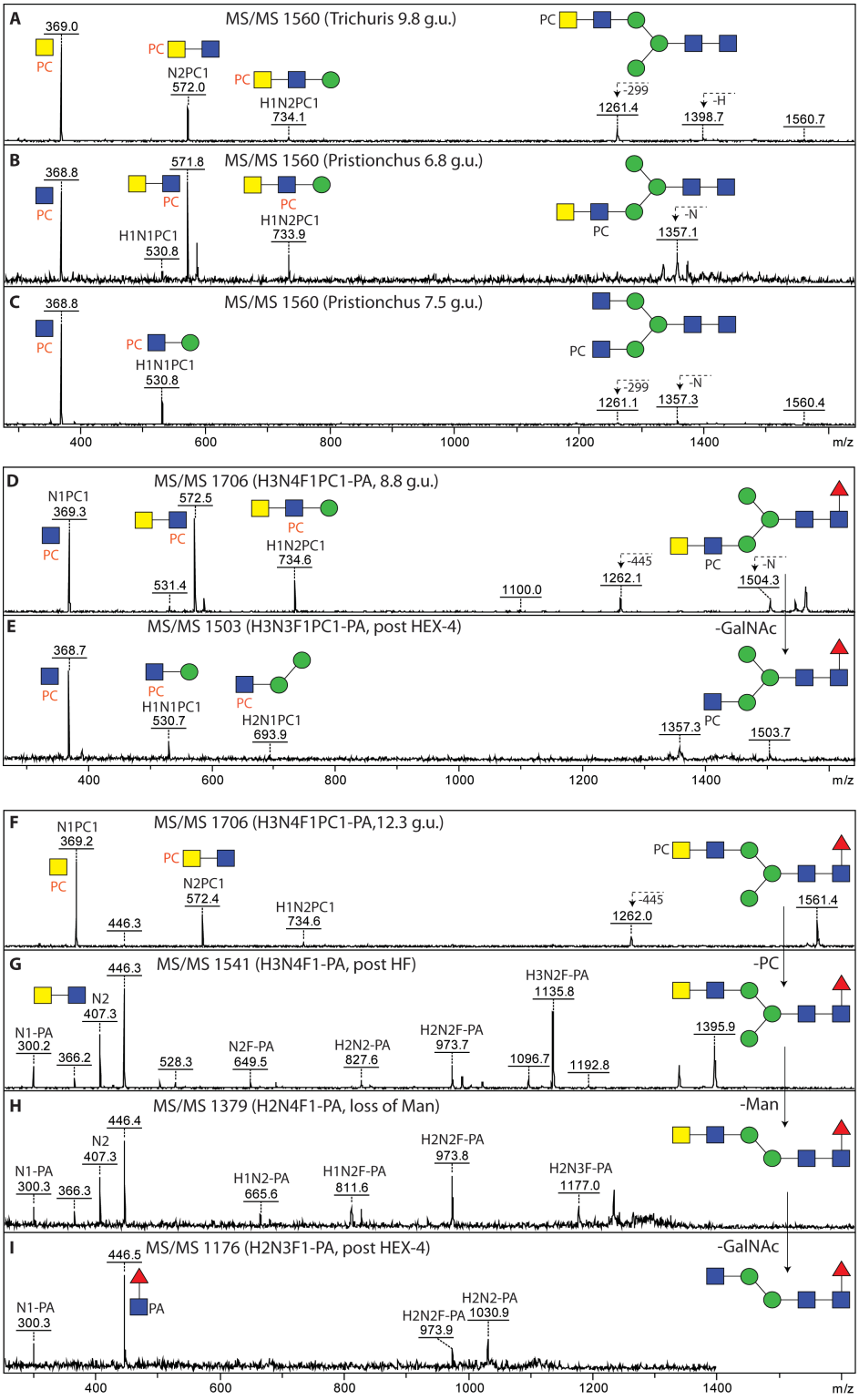


**Fig. S4** Comparisons of MS/MS fragmentation patterns for phosphorylcholine-modified N-glycans. (A-C) MS/MS of the *m/z* 1560 isomers from *T. suis* and *P. pacificus* highlighting the differences in the intensities and occurrence of the *m/z* 369, 531, 572 and 734 fragment ions. (D, E) MS/MS of the $Hex_3HexNAc_4Fuc_1PC_1$ glycan eluting at 8.8 g.u. before and after HEX-4 treatment. (F-I) MS/MS of the $Hex_3HexNAc_4Fuc_1PC_1$ glycan eluting at 12.3 g.u. before and after serial loss of phosphorylcholine (HF treatment), mannose and GalNAc (HEX-4 treatment). The occurrence of [M+H-HexNAc] fragment ions in B and D as well as the ability to digest the 8.8 g.u. glycan with HEX-4 without prior hydrofluoric acid treatment (E) is indicative of a subterminal phosphorylcholine modification, whereas the absence of [M+H-HexNAc] fragment ions in A and F is consistent with a terminal position for the zwitterion.