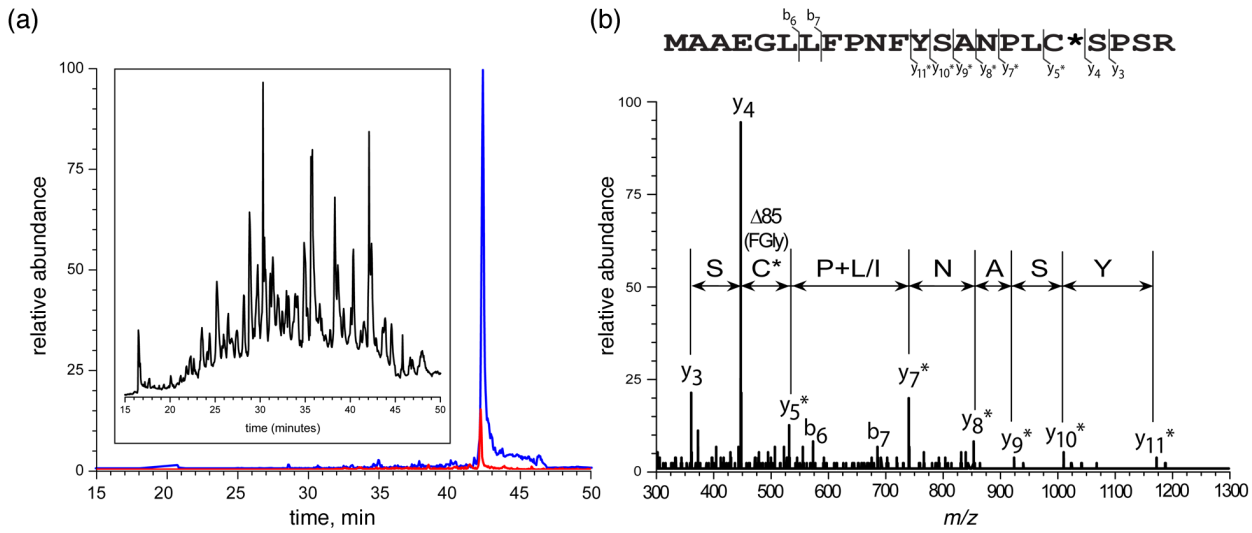
Supplemental Fig. 1. Mass spectrometry of GALNS

GALNS was reduced, alkylated, and subsequently digested with trypsin. The peptide mixture was separated by nano-LC and analyzed using online mass spectrometry for peptide identification and simultaneous CAD for residue specific information. (a): NanoLC separation of peptides is shown by the total ion chromatogram (black) and extracted ion chromatograms of the +3 charge state for tryptic peptides containing Cα-formylglycine (*m/z* 790.1, blue) or carboxymethylated Cys (*m/z* 815.5, red) at residue 79. (b): The tryptic-peptide ion containing formylglycine was subjected to CAD. Various fragment ions were observed including the y_4 and y_5 ions, representing cleavage of peptide bonds on either side of the formylglycine residue.

Supplemental Fig. 2. Structure-based sequence alignment of human sulfatases

A structure-based sequence alignment of human GALNS (PDB ID 4FDI), ASA (PDB ID 1AUK), ASB (PDB ID 1FSU), and ASC (PDB ID 1P49) is shown with amino acid sequence identities in cyan, conservative substitutions in orange, and active site residues in red. Helices and strands are shown as cylinders and arrows, respectively, colored as in Fig. 1. Potential N-linked glycosylation sites and disulfide connections are indicated.



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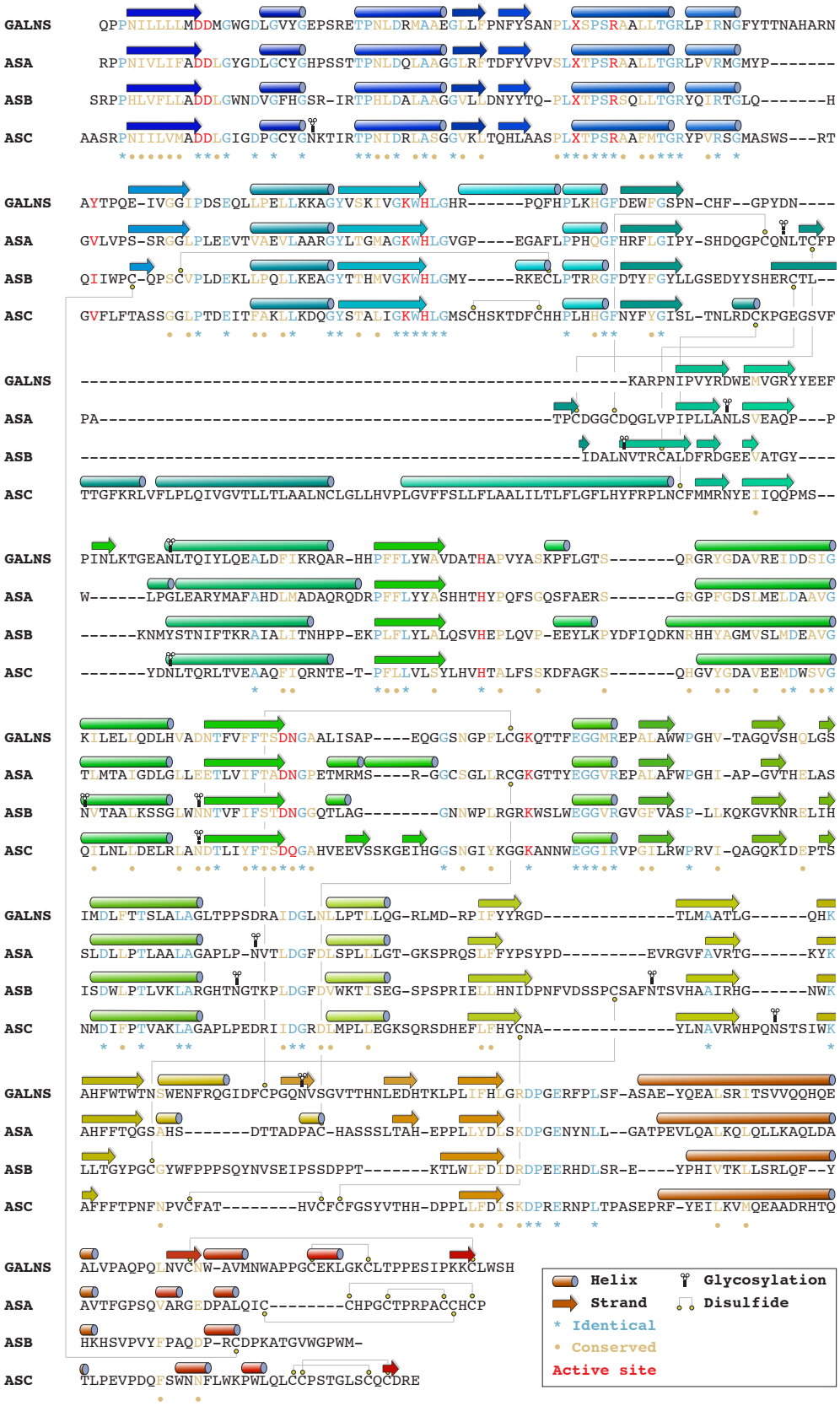

Rivera-Colón et al., Supplemental Figure 2