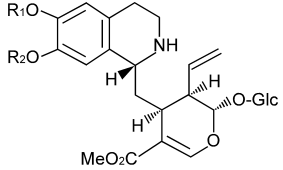
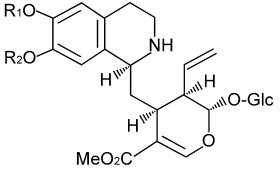
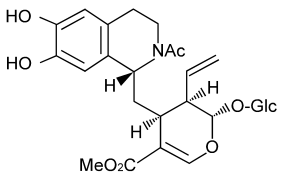


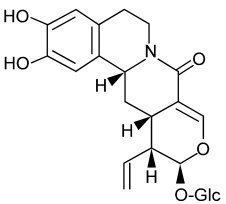
R_1 = H, R_2 = H : *N*-Deacetylipecoside
R_1 = Me, R_2 = H : 6-*O*-Methyl-*N*-deacetylipecoside
R_1 = H, R_2 = Me : 7-*O*-Methyl-*N*-deacetylipecoside
R_1 = Me, R_2 = Me : 6,7-*O,O*-Dimethyl-*N*-deacetylipecoside



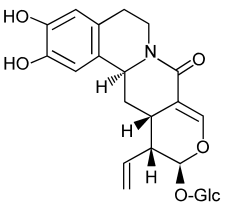
R_1 = H, R_2 = H : *N*-Deacetylisoipecoside
R_1 = Me, R_2 = H : 6-*O*-Methyl-*N*-deacetylisoipecoside
R_1 = H, R_2 = Me : 7-*O*-Methyl-*N*-deacetylisoipecoside
R_1 = Me, R_2 = Me : 6,7-*O,O*-Dimethyl-*N*-deacetylisoipecoside



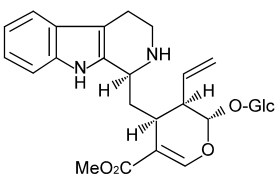
Ipecoside



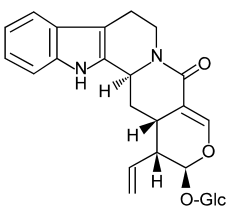
Demethylalangiside



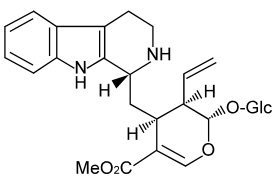
Demethylisoalangiside



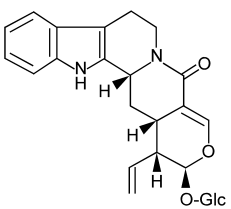
Strictosidine



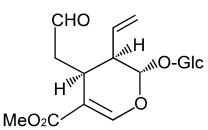
Strictosidine lactam



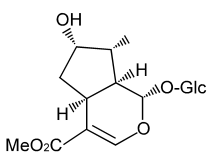
Vincoside



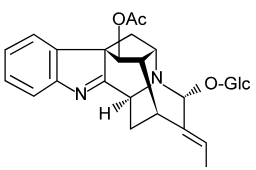
Vincoside lactam



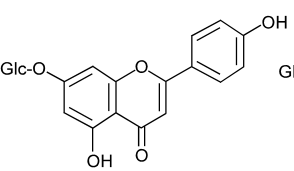
Secologanin



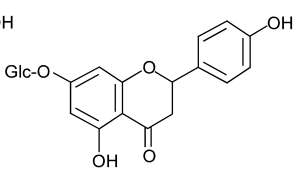
Loganin



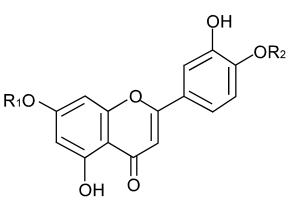
Raucaffricine



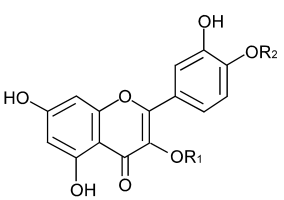
Apigenin-7-*O*-Glc



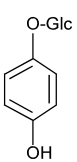
Naringenin-7-*O*-Glc



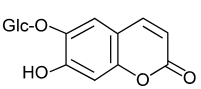
R_1 = Glc, R_2 = H : Luteolin-7-*O*-Glc
R_1 = H, R_2 = Glc : Luteolin-4'-*O*-Glc



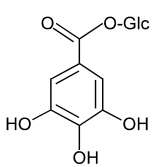
R_1 = Glc, R_2 = H : Quercetin-3-*O*-Glc
R_1 = H, R_2 = Glc : Quercetin-4'-*O*-Glc



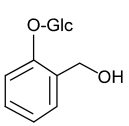
Arbutin



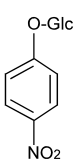
Esculin



Galloyl-Glc



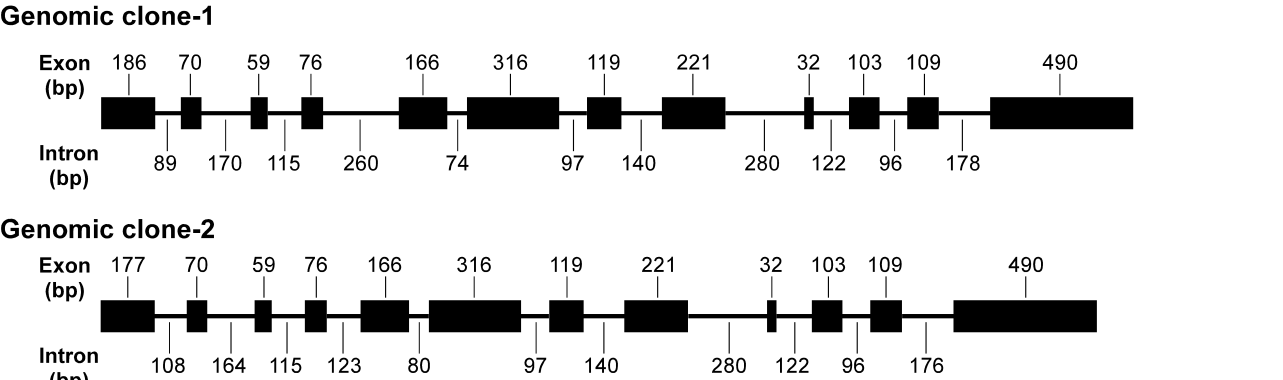
Salicin



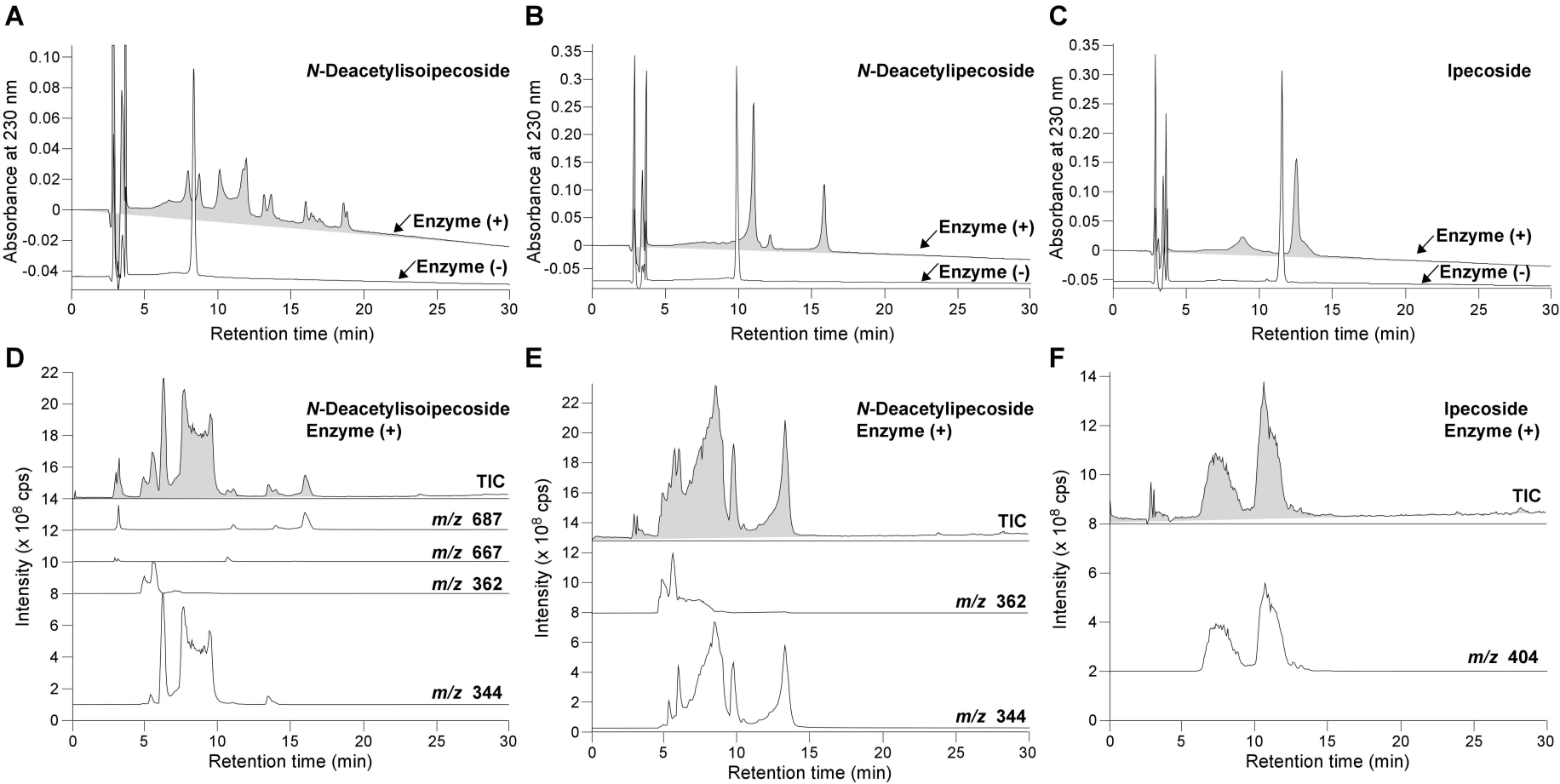
pNP-Glc

Supplemental Figure 1. Chemical structures of the compounds tested for enzyme assay of IpeGlu1.

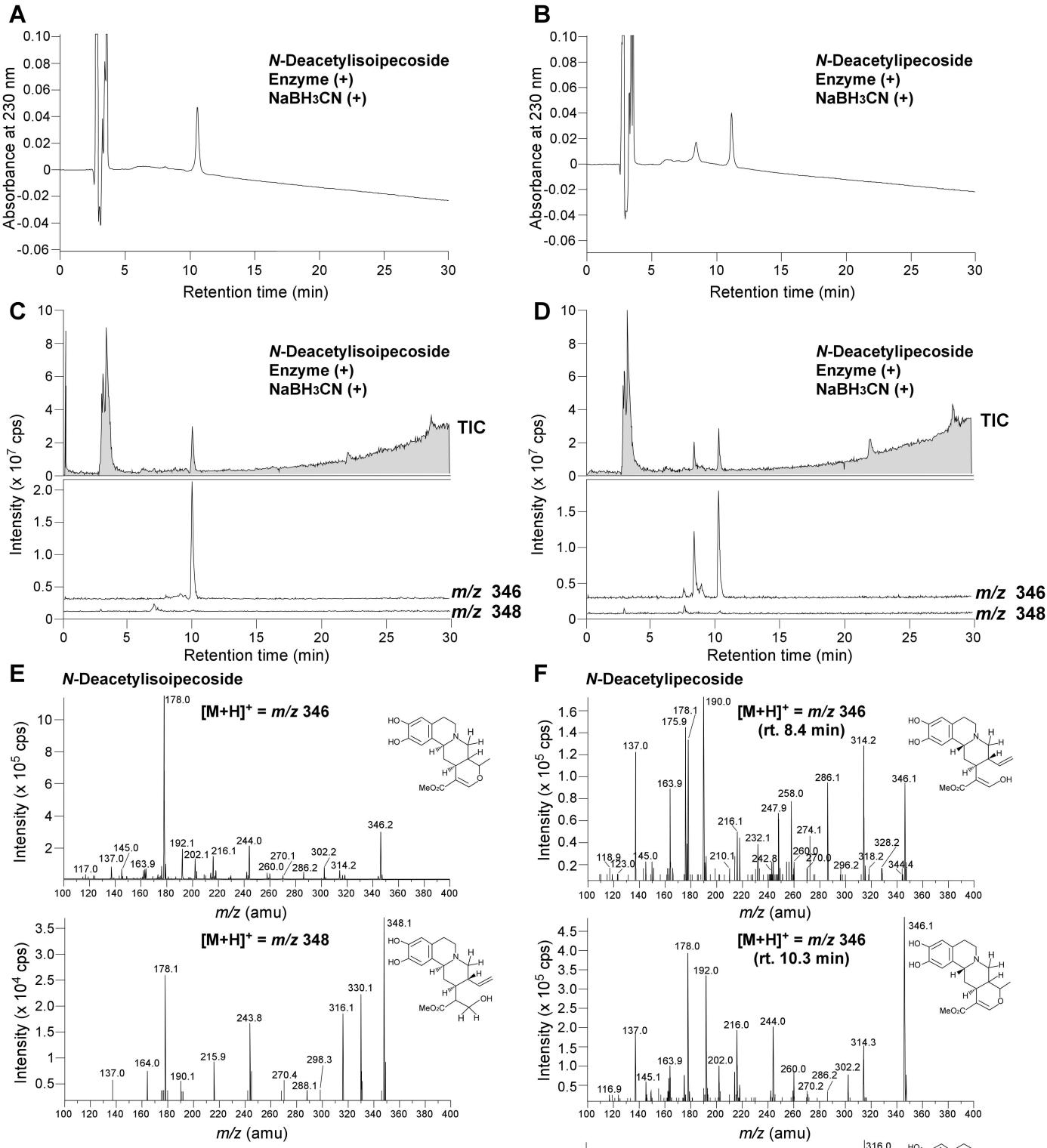
Supplemental Figure 2. Sequence alignment of deduced amino acids of Ipecac alkaloid β-glucosidases (IpeGlu1 and IpeGlu9) with other β-glucosidases of alkaloid biosynthesis. Raucaffricine β-glucosidase (*R. serpentina*, GenBank accession no. AF149311), strictosidine β-glucosidase (*R. serpentina*, AJ302044) and strictosidine β-glucosidase (*C. roseus*, AF112888). Conserved sequence motifs among family 1 glycosyl hydrolases are boxed. Amino acid residues involved in the recognition of the aglycon moiety in strictosidine β-glucosidase of *R. serpentina* (32) are marked by black triangles.



Supplemental Figure 3. Exon-intron structure of the genomic clones of Ipecac alkaloid β-glucosidase. Exon and intron are shown by black box and line, respectively. The number shows the nucleotide length of each exon/intron. Genomic PCR products amplified using the same primers as used for the cloning of the *Ipeglu2-Ipeglu9* cDNAs were cloned into pCR-Blunt II-TOPO (Invitrogen) and sequenced. Genomic clone-1 (GenBank accession no. AB455585) is 3,568-bp long consisting of 1,947-bp exons and 1,621-bp introns. Genomic clone-2 (AB455586) is 3,439-bp long consisting of 1,938-bp exons and 1,501-bp introns. The exon sequences of the genomic clones-1 and -2 were identical to the *Ipeglu3* and *Ipeglu5* cDNA sequences, respectively. Both genomic clones comprised 12 exons and 11 introns.



Supplemental Figure 4. HPLC (A-C) and LC-MS/MS (D-F) analyses of the IpeGlu1 reaction mixture with *N*-deacetylisoipecoside (A and D), *N*-deacetylipecoside (B and E), and ipecoside (C and F). Chromatograms of the reaction mixture with (Enzyme +) or without (Enzyme -) enzyme are shown for HPLC analysis. The total ion chromatogram (TIC) and the chromatogram of individual molecular ions included in the reaction mixture are shown for LC-MS/MS analysis. Fragmentation of each molecular ion is not shown. Differences in the peak retention time between HPLC and LC-MS/MS analyses are due to the different HPLC systems.



Supplemental Figure 5. HPLC (A and B) and LC-MS/MS (C-F) analyses of the IpeGlu1 reaction mixture under reduced conditions ($NaBH_3CN$) with *N*-deacetylisoipecoside (A, C and E) and *N*-deacetylipecoside (B, D and F) as substrate. The total ion chromatogram (TIC) and the chromatogram of individual molecular ions included in the reaction mixture are shown in C and D. Fragmentation of each molecular ion is shown in E and F. Differences in the peak retention time between HPLC and LC-MS/MS analyses are due to the different HPLC systems.