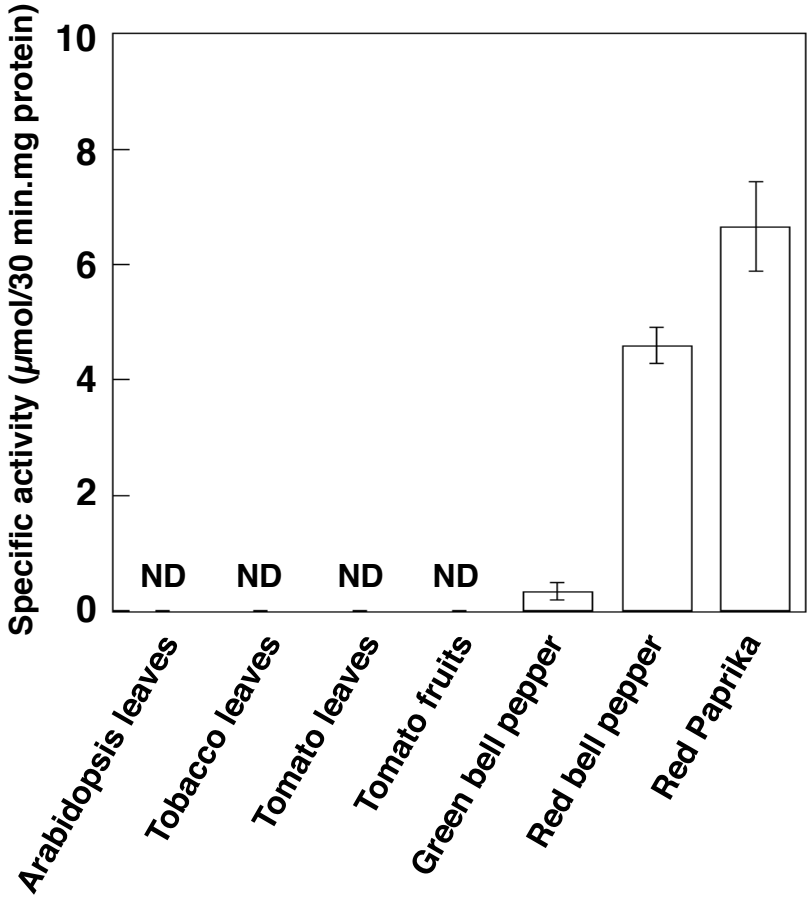

Fig. S1. Screening of plant materials. HI activity in crude extract prepared from each plant material was determined by standard assay. Data are means ± SE (*n*=3). ND, not detected.

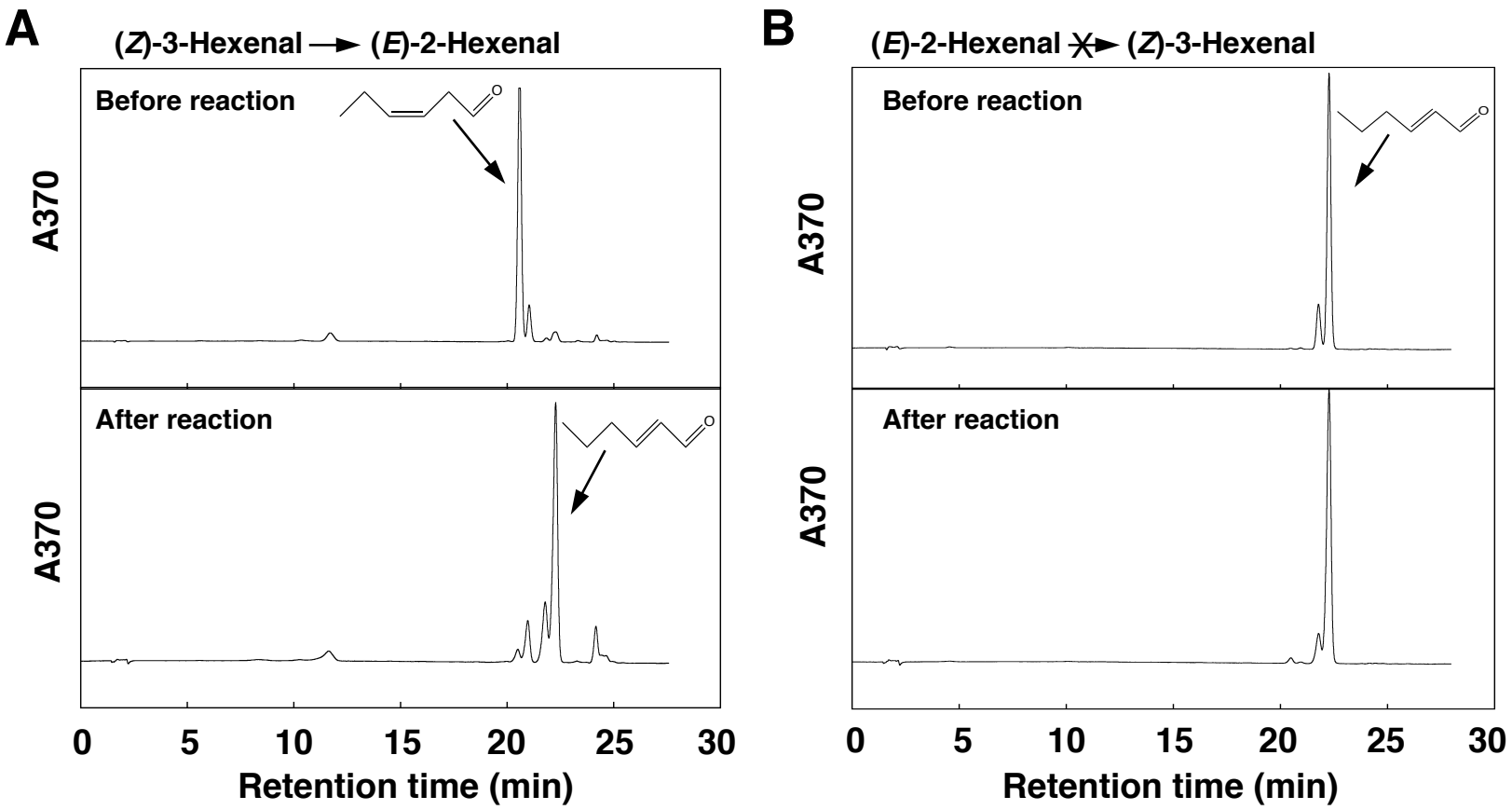

Fig. S2. Detection of HI activity by HPLC. Typical chromatograms of DNP-derivatized samples prepared from reaction mixtures before and after reaction are shown. Arrows indicate peaks of which retention time were identical to those of authentic compound-DNPs, respectively. Substrates are (*Z*)-3-hexenal (*A*), (*E*)-2-hexenal (*B*), (*E*)-2-nonenal (*C*), and (*Z*,*Z*)-3,6-nonadienal (*D*). (*E*)-2-Hexenal was not isomerized (*B*).

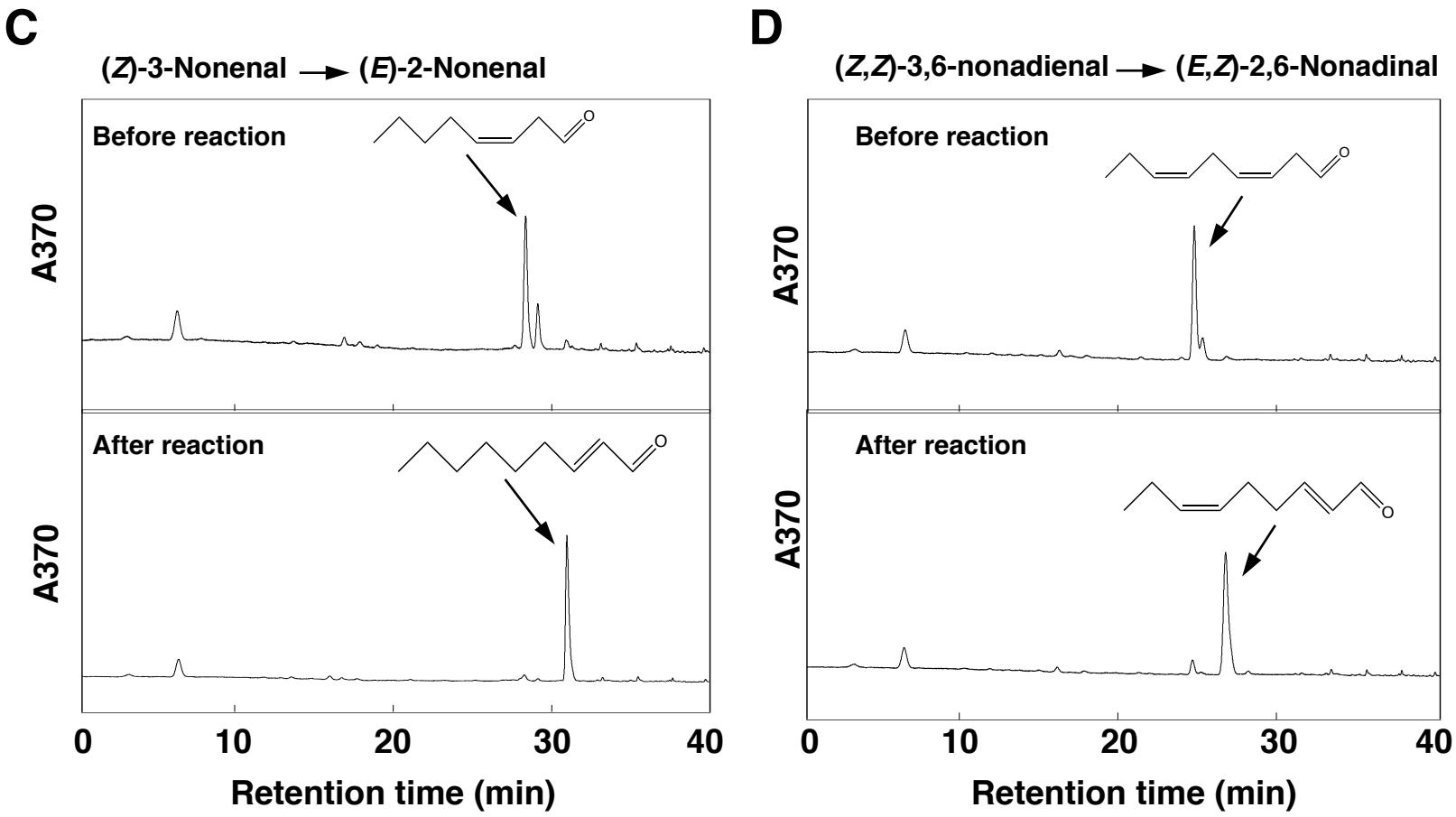

Fig. S2. (continued)

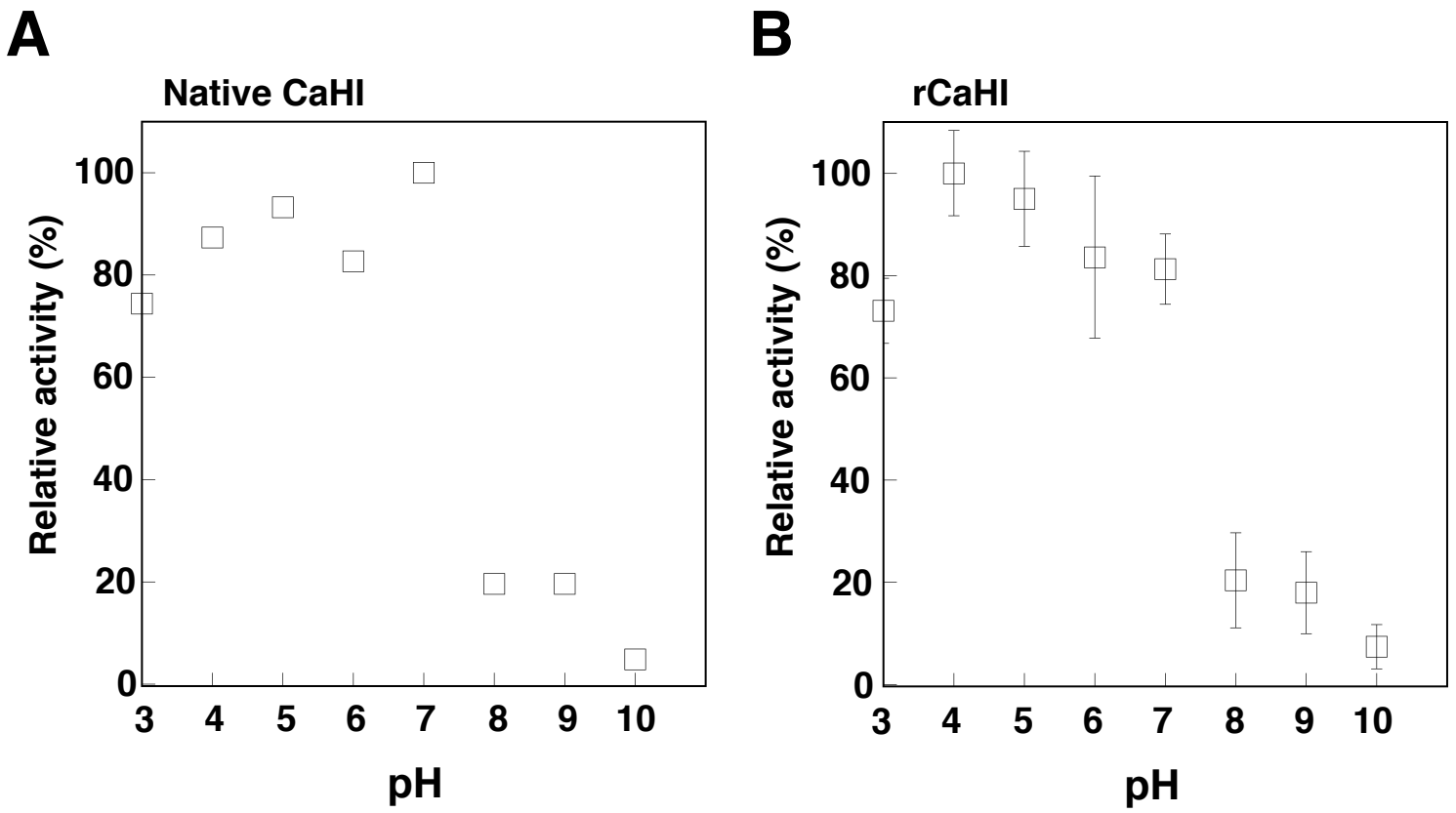

Fig. S3. pH dependence of activities of native CaHI (A) and rCaHI (B). Enzyme activity was determined by standard assay in 50 mM Na-acetate buffer (pH 3.0-7.0) or 50 mM Tris-HCl buffer (pH 8.0-10.0). Data of native CaHI and rCaHI are from single assay and triplicate assays (means ± SE), respectively. Maximum activity was set to 100%.

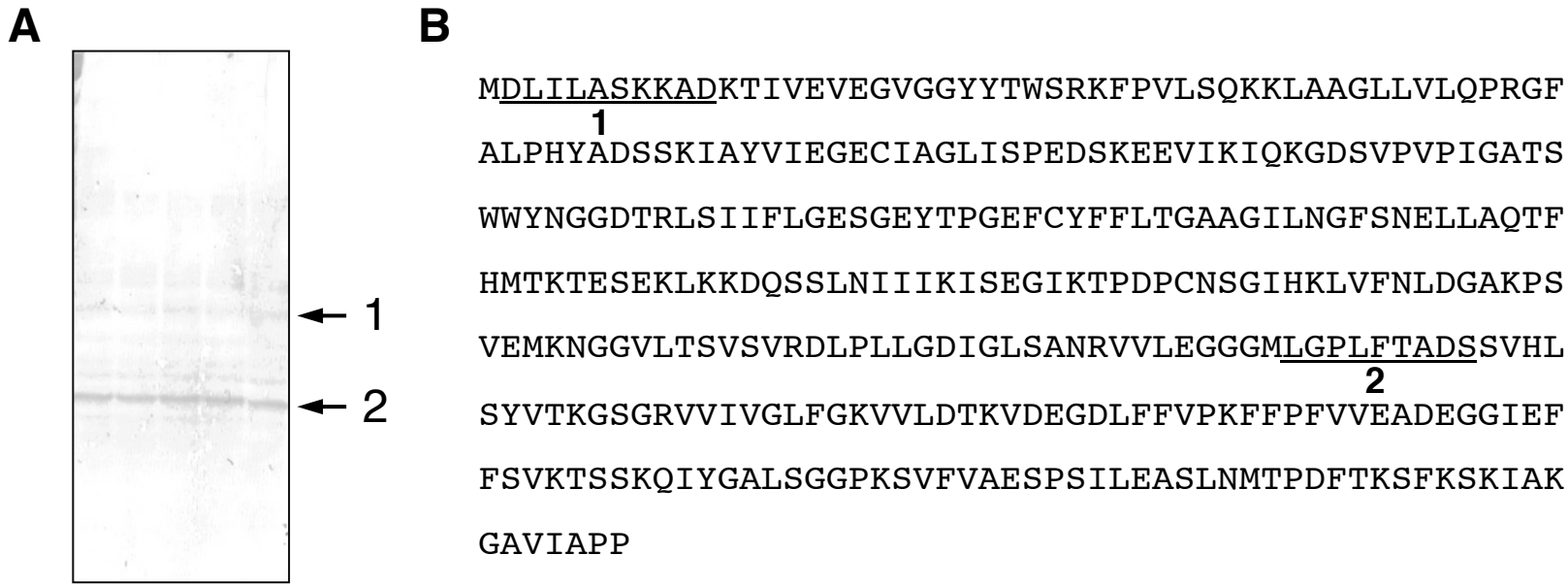

Fig. S4. Determination of internal amino acid sequence of CaHI. (*A*) Purified CaHI was cleaved by BrCN and then the resultant peptide fragments were separated with Tricine-SDS-PAGE. Peptides blotted onto PVDF membrane were stained by CBB R-250. Arrows indicate peptides of which sequences could be determined. (*B*) Determined internal amino acid sequences are underlined on the whole amino acid sequence of CaHI.

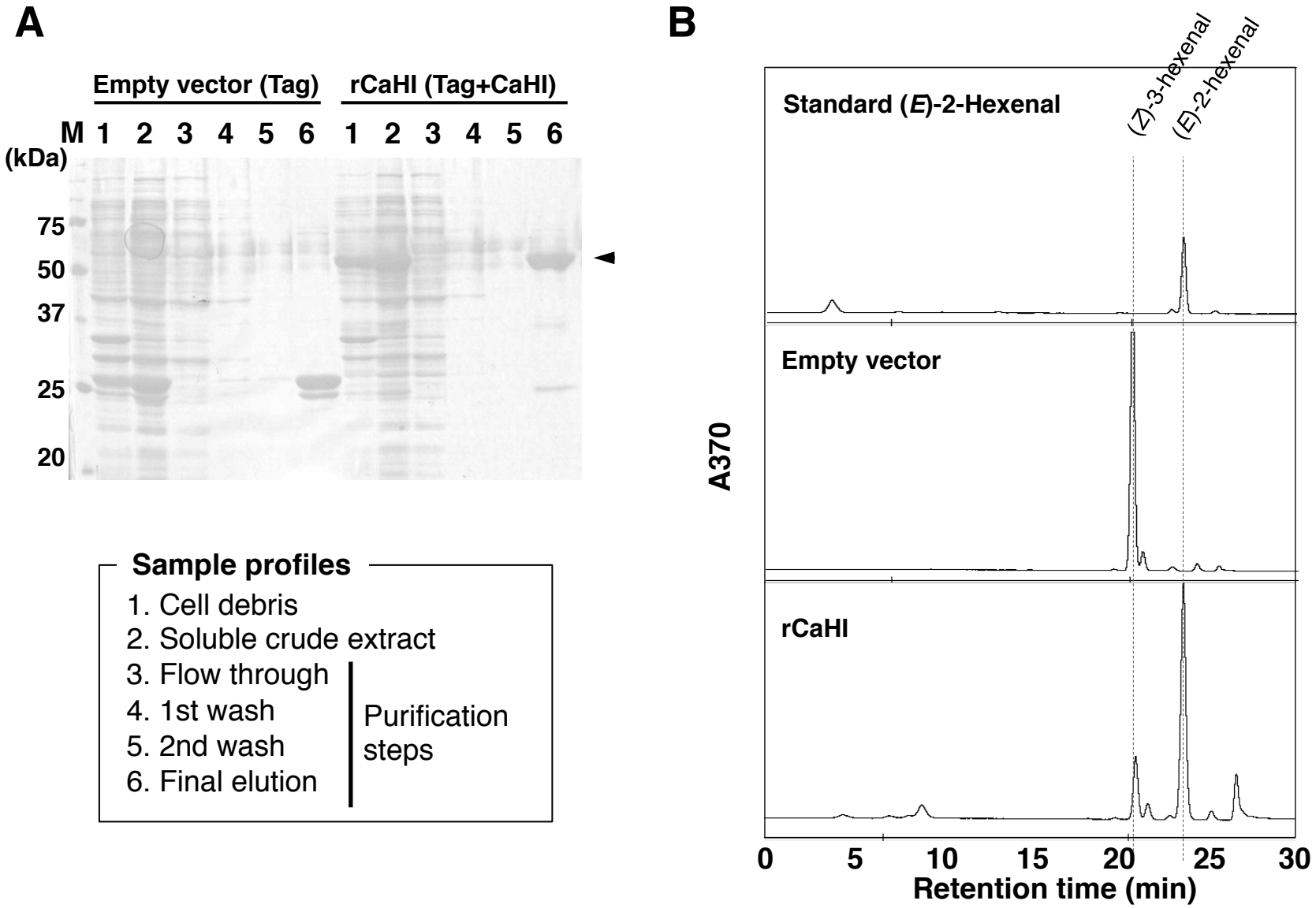

Fig. S5. Production of recombinant HI by heterogeneous expression in *E. coli*. (*A*) Purification of recombinant CaHI (rCaHI). Proteins in each fraction were electrophoresed by SDS-PAGE. Purified rCaHI is indicated by an arrowhead (60 kD = 35 kD (CaHI) + 25 kD (Tag)). (*B*) Activity of the purified rCaHI was confirmed by the production of (*E*)-2-hexenal. (*C*) By same strategy, activities of recombinant HIs from tomato (rSlHI1) and potato (rStHI1 and 2) were confirmed, on the contrary, recombinant HI-like protein from tomato (rSlHI-like1) did not show activity.

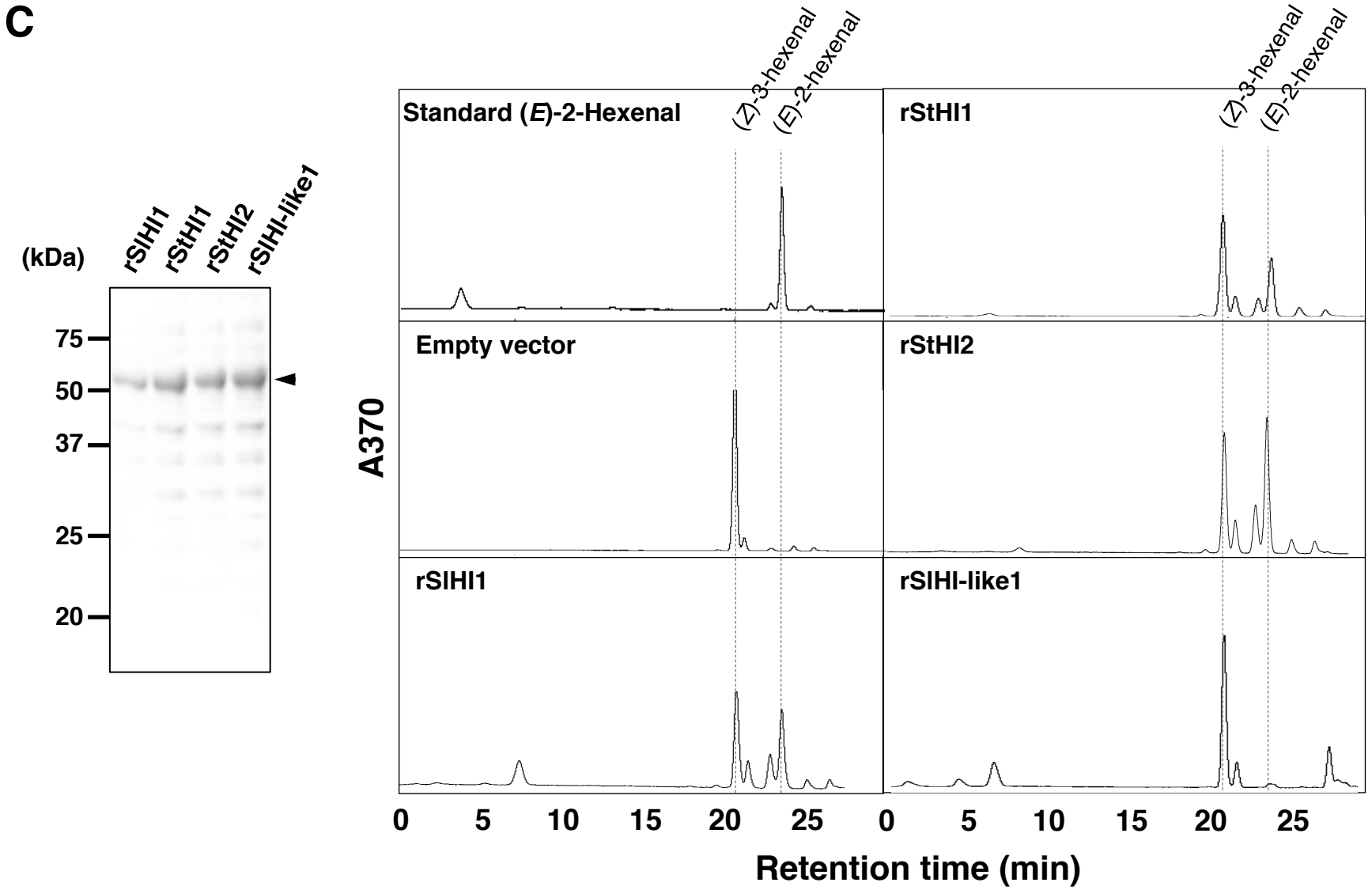

Fig. S5. (continued)

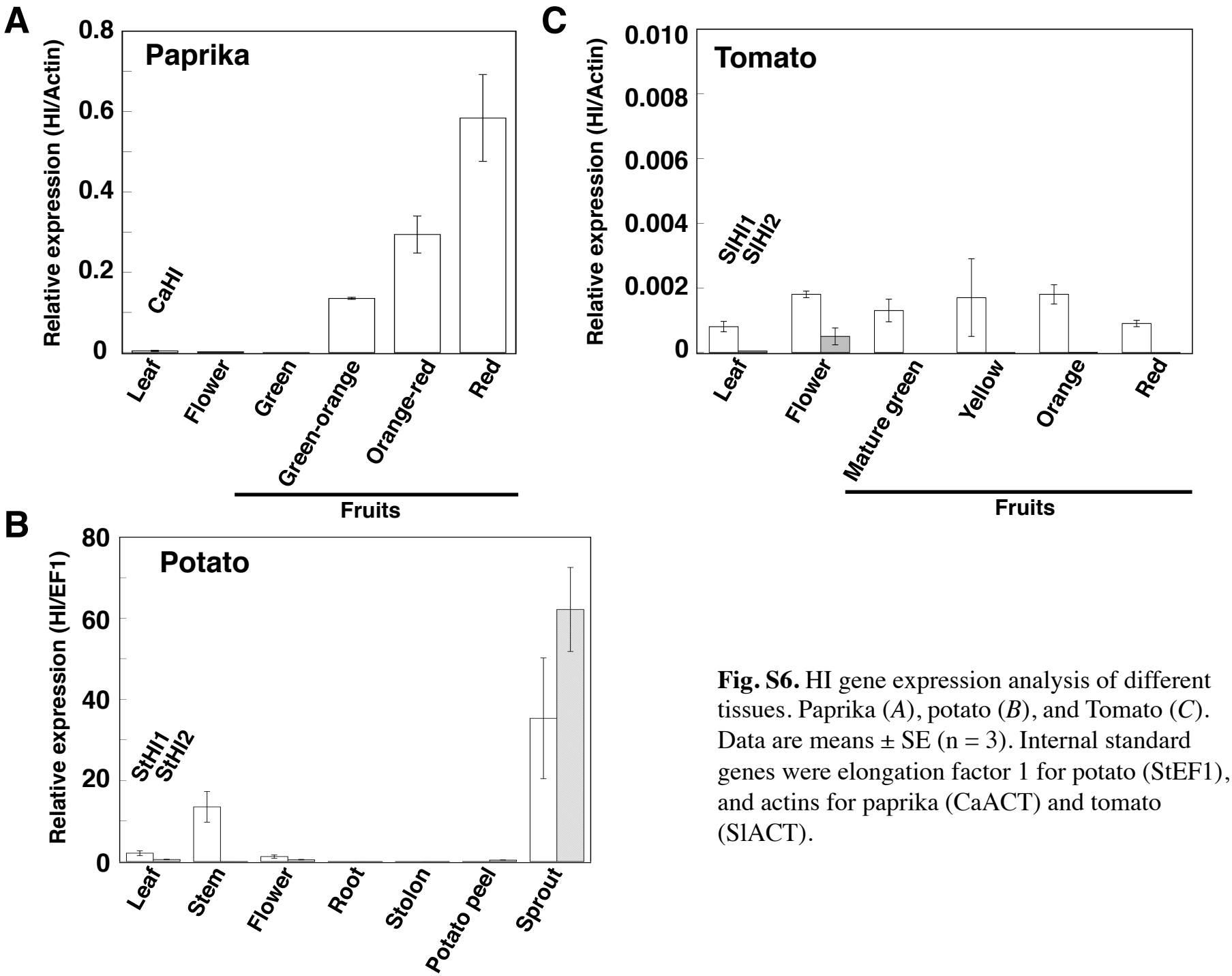

Fig. S6. HI gene expression analysis of different tissues. Paprika (*A*), potato (*B*), and Tomato (*C*). Data are means ± SE (n = 3). Internal standard genes were elongation factor 1 for potato (StEF1), and actins for paprika (CaACT) and tomato (SlACT).

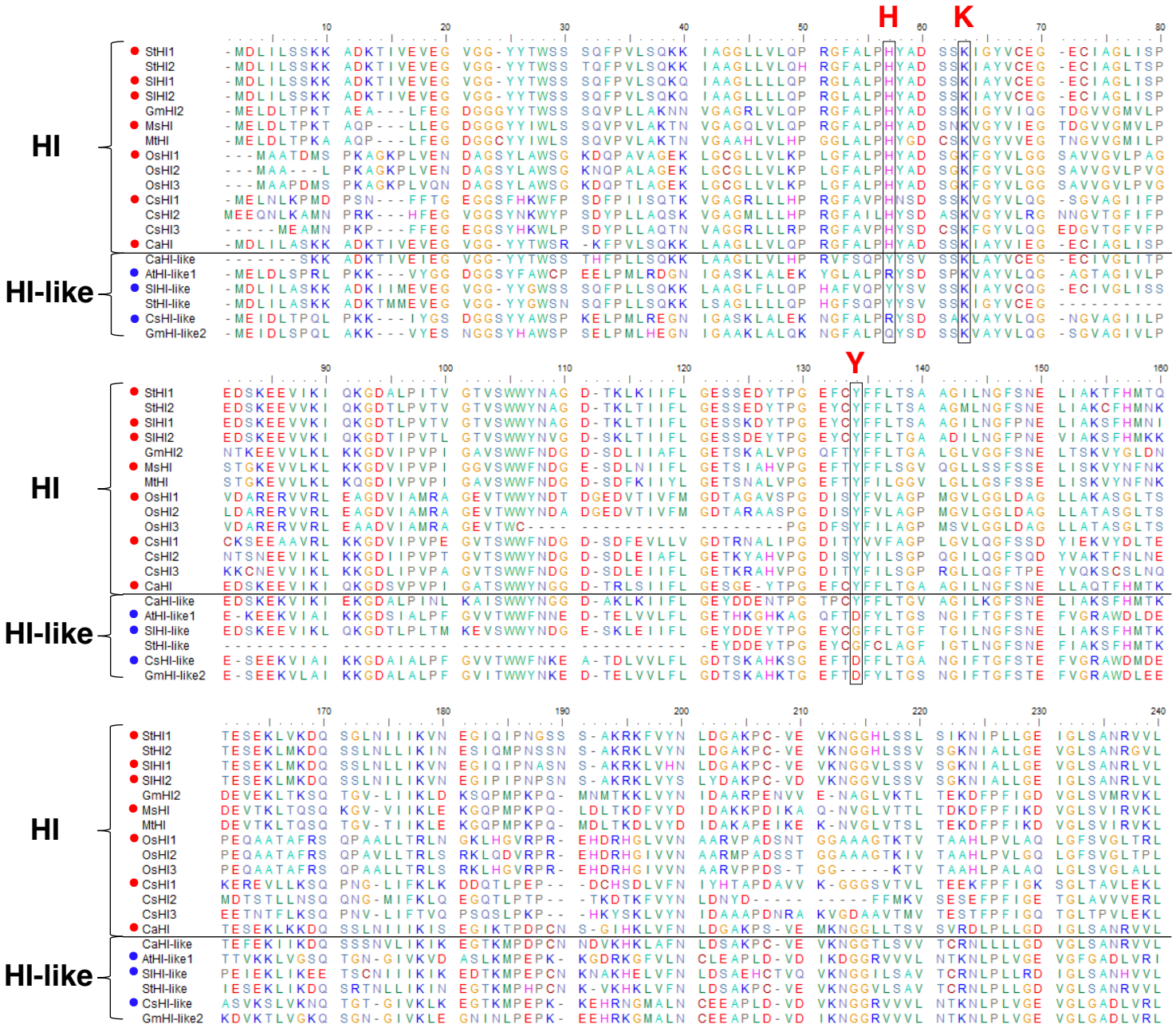

Fig.S7. Alignment of HI and HI-like proteins. Positions of catalytic HKY are boxed. Red and blue dots indicate that their recombinant proteins showed activities and not, respectively.

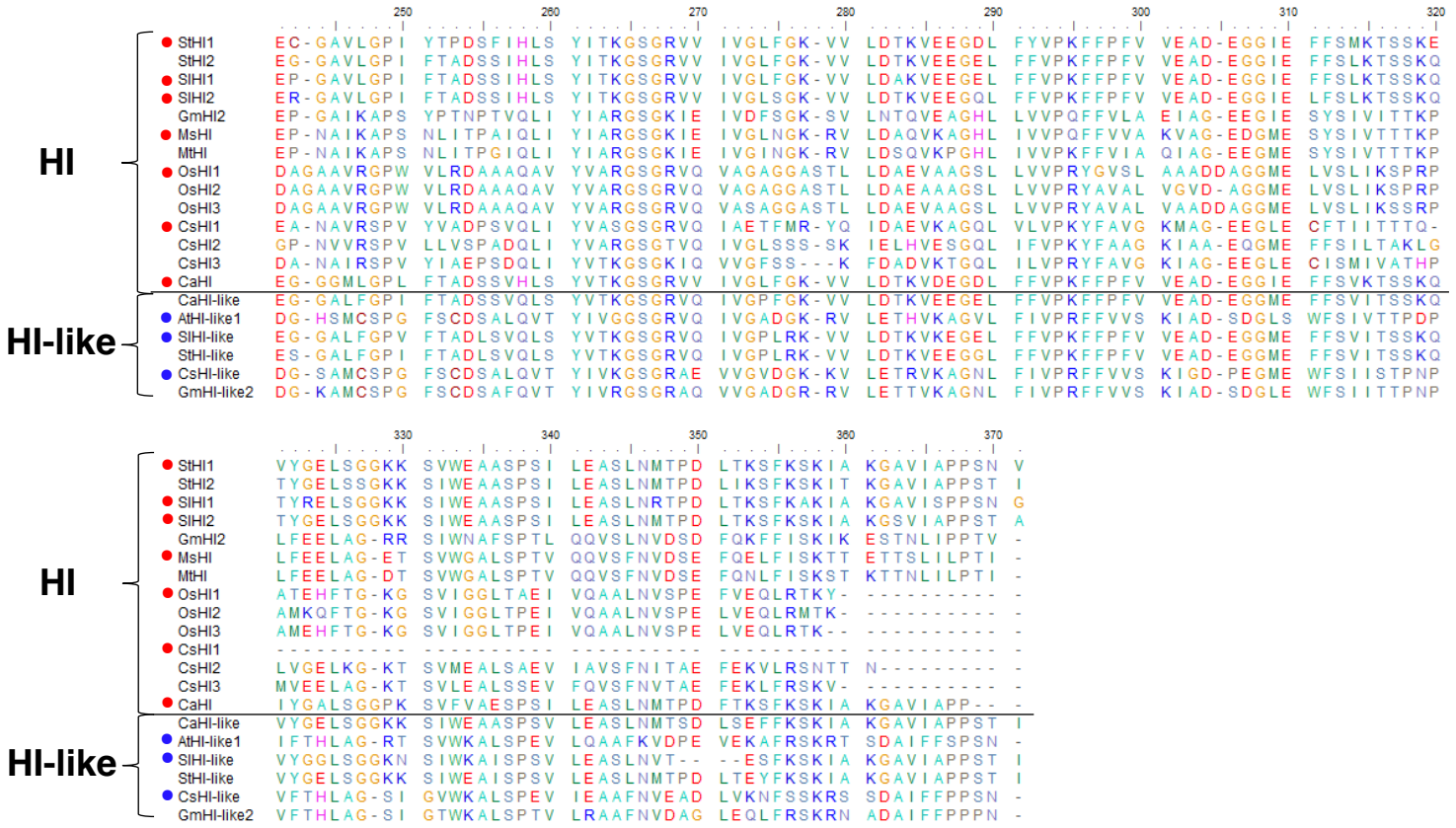

Fig. S7. (continued)

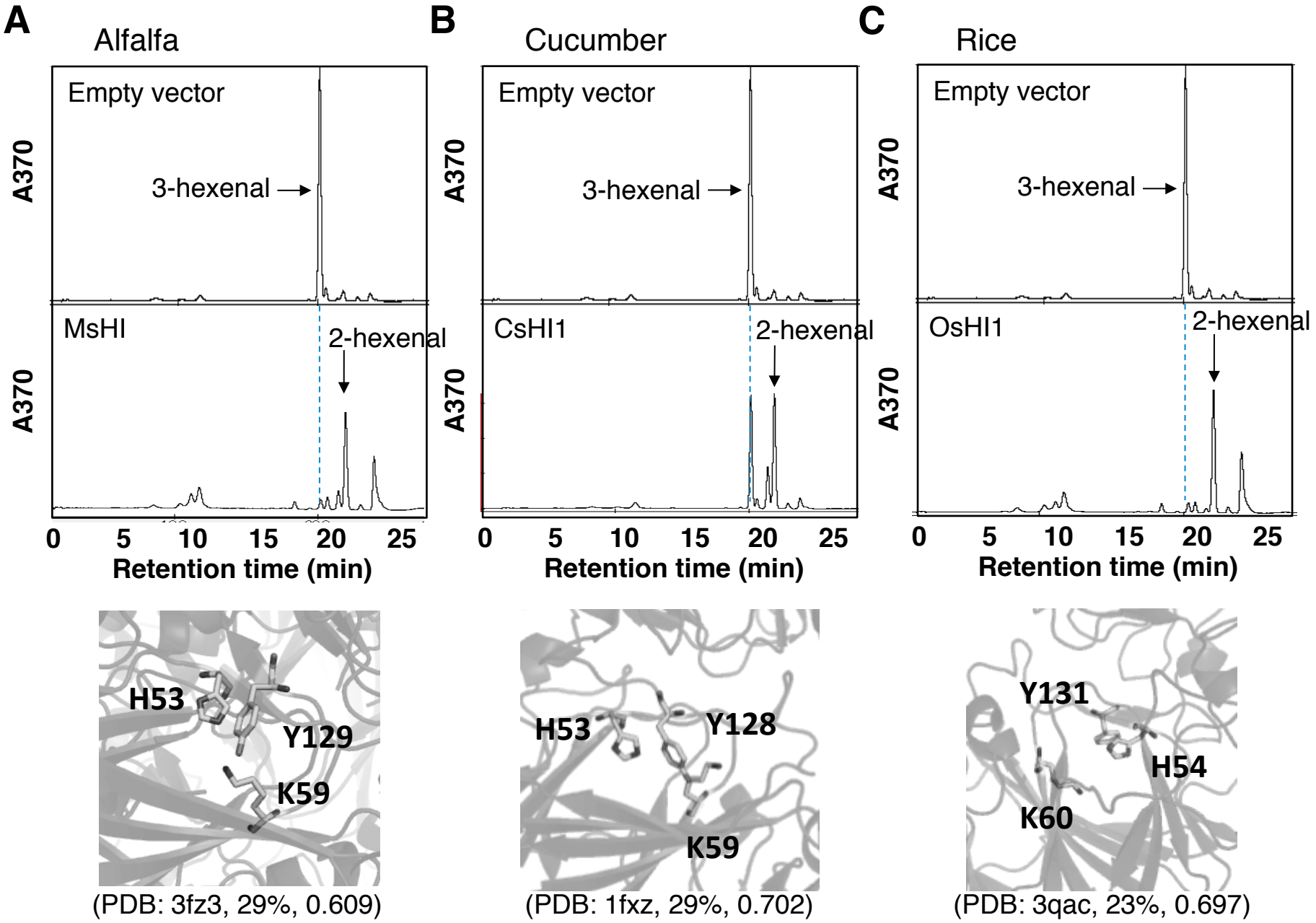

Fig. S8. HI homologs of alfalfa (*A*), cucumber (*B*), and rice (*C*) showed HI activity. Catalytic pocket of each HI by homology modeling is shown in lower panel. Catalytic HKY locate in the same pocket. PDB ID of template protein, identity between CaHI and template protein, and QMEAN score are shown in parentheses.

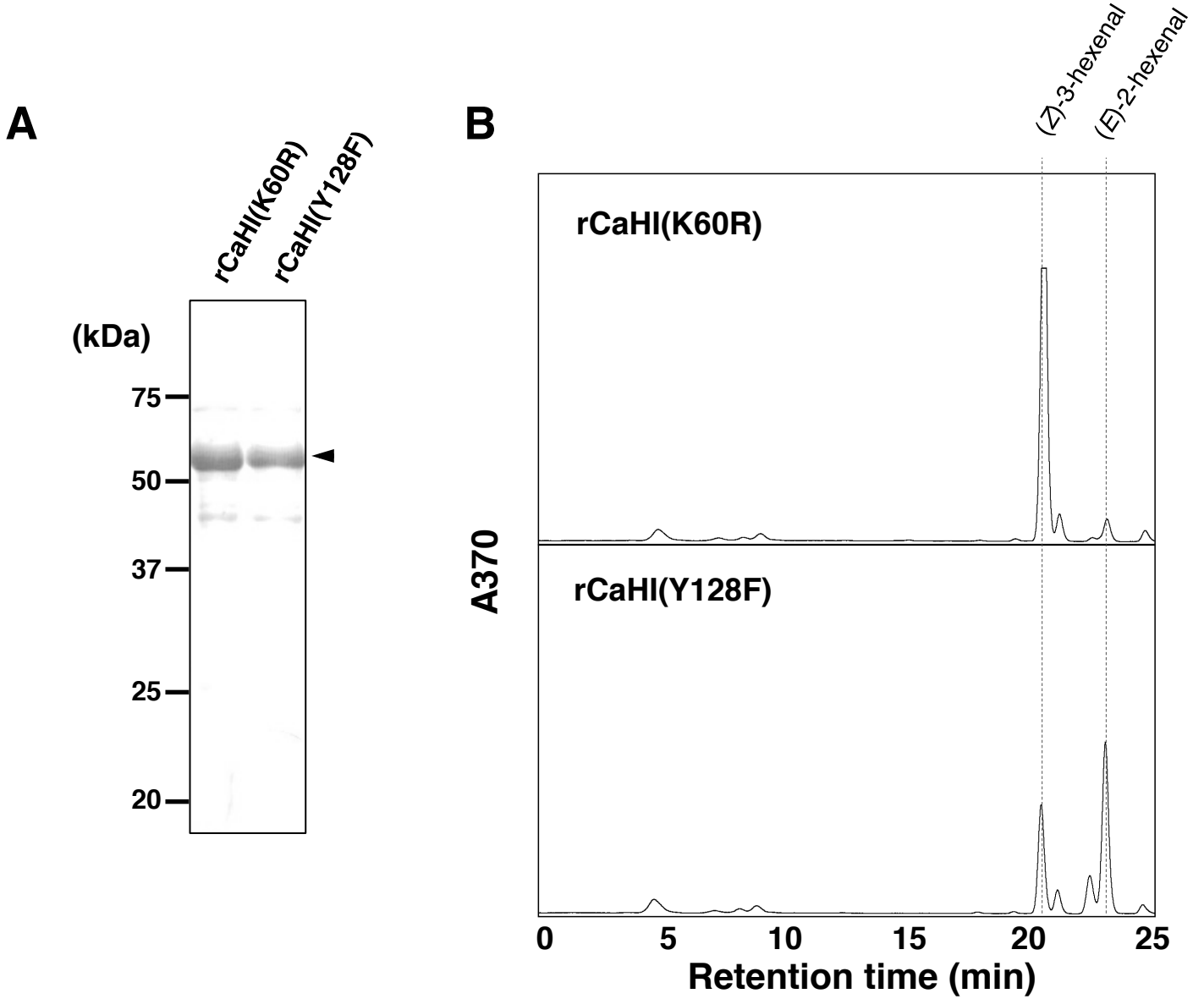

Fig. S9. Production of point-muted rCaHI by heterogeneous expression in *E. coli*. (*A*) Purified rCaHI recovered in soluble fractions were electrophoresed by SDS-PAGE. Purified rCaHI is indicated by an arrowhead (60 kD = 35 kD (CaHI) + 25 kD (Tag)). (*B*) Activity of the purified rCaHI was determined by the production of (*E*)-2-hexenal.