Figure S1 - Zoom into the part of the tree where *C. scindens* DesB is located in the large scale phylogenetic analysis of DesB. The full maximum-likelihood tree contains 38,186 NCBI non-redundant database protein sequences with lengths between 200 and 800 amino acids that matched *C. scindens* DesB. Branch colors reflect taxonomic affiliation as shown in figure 1. Numbers on nodes are FastTree-calculated support values, with only values above 0.5 being shown.

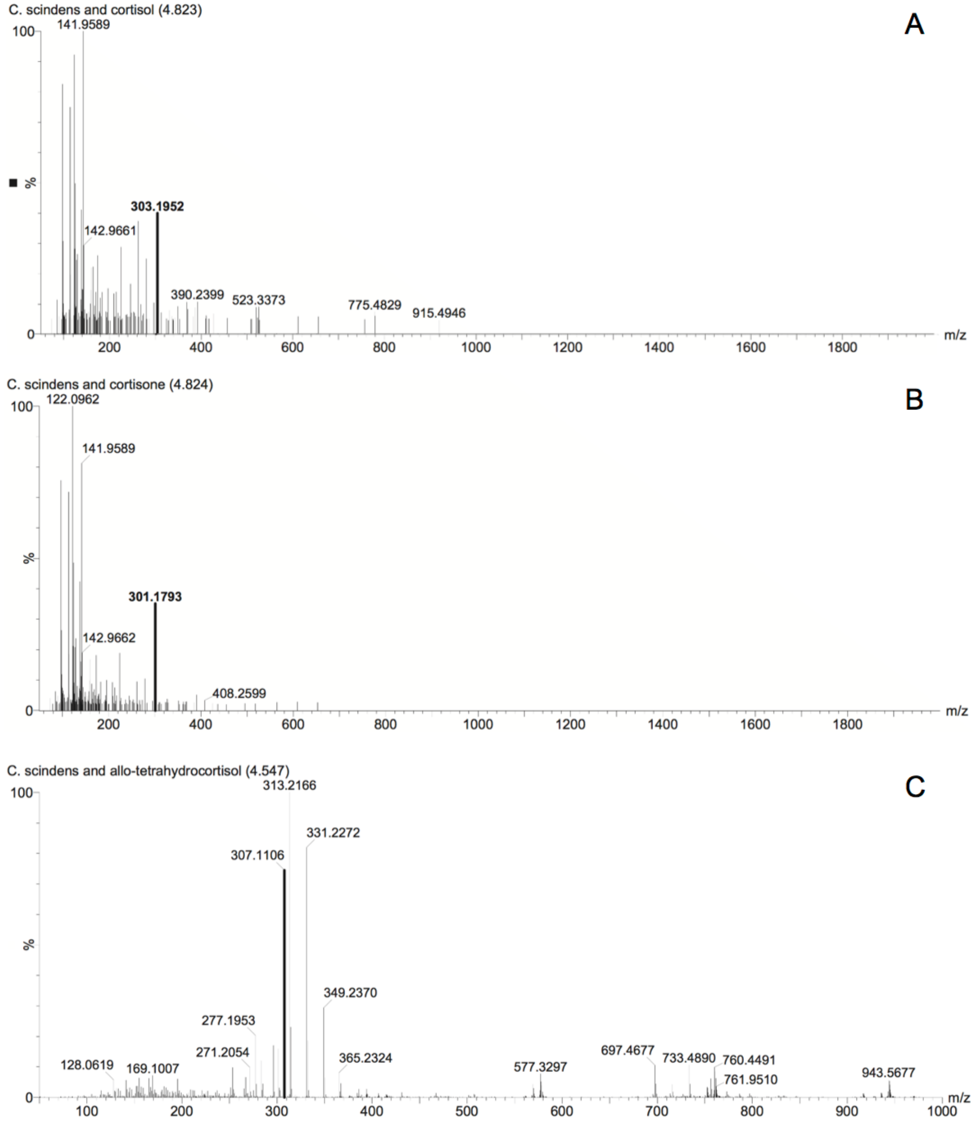

Figure S2. Mass spectra of side chain cleaved product of endogenous glucocorticoids screened after incubation with *C. scindens*. Bolded mass ions represent -60.02 amu less than the original m/z of (a) cortisol, (b) cortisone, and (c) allo-tetrahydrocortisol.

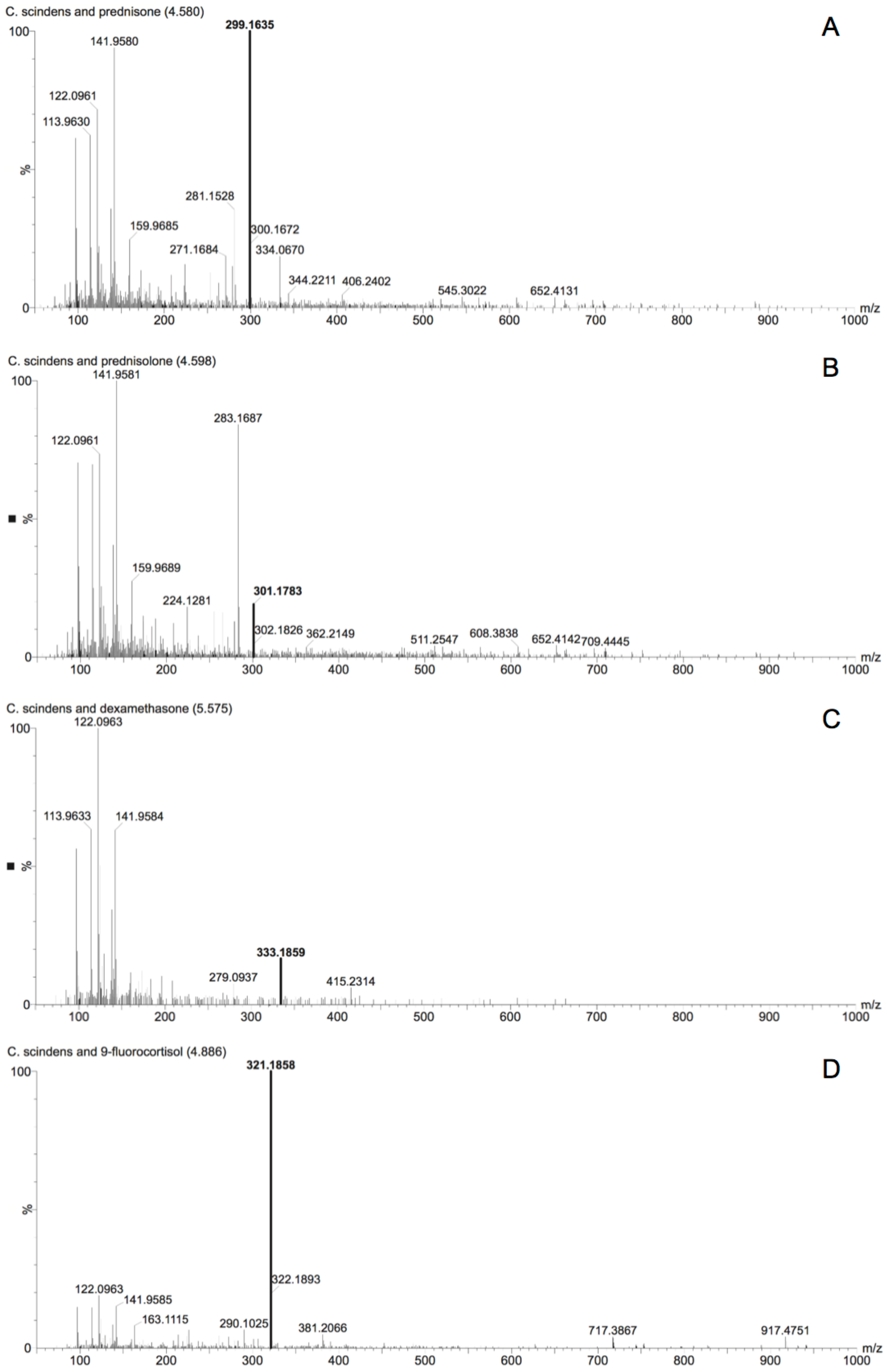

Figure S3. Mass spectra of side chain cleaved product of exogenous glucocorticoids screened after incubation with *C. scindens*. Bolded mass ions represent -60.02 amu less than the original m/z of (a) prednisone, (b) prednisolone, (c) dexamethasone, and (d) 9-fluorocortisol.

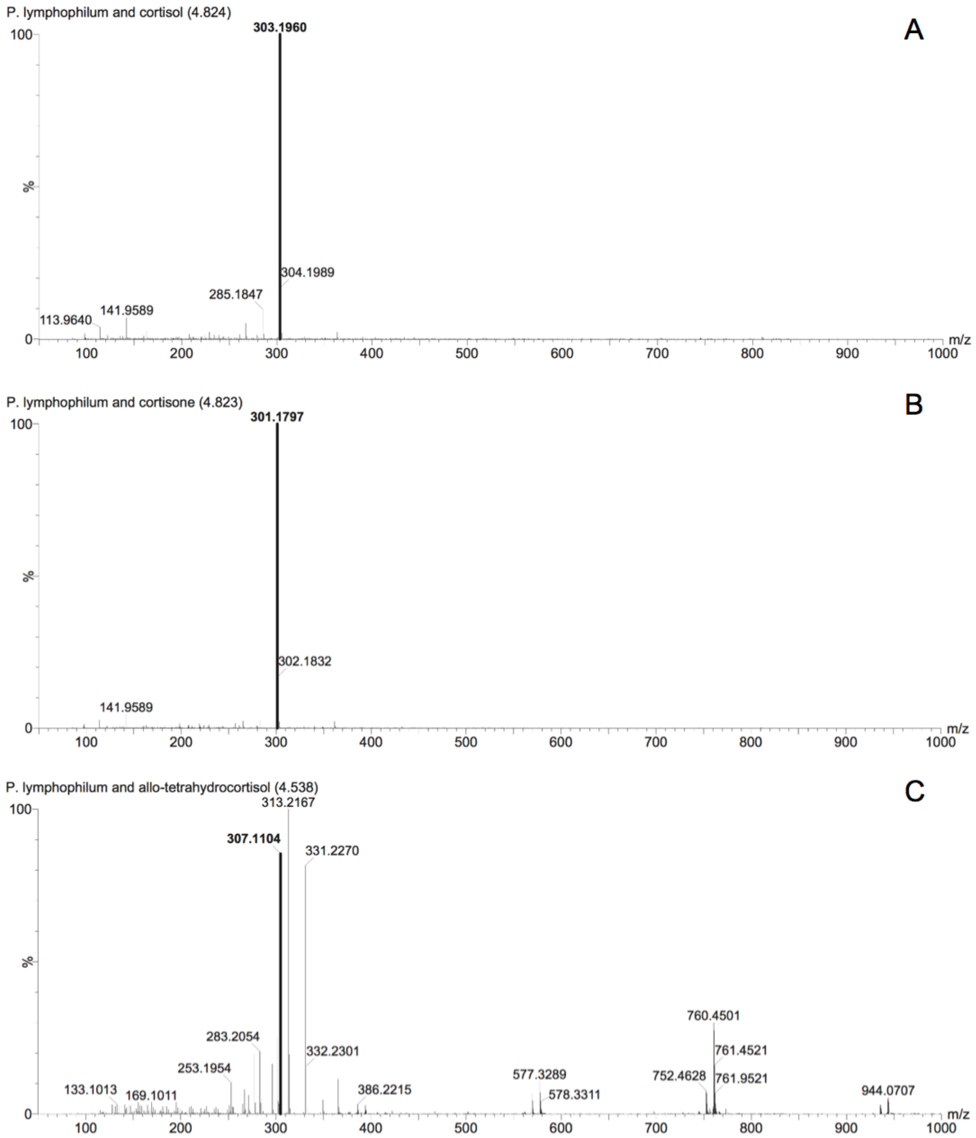

Figure S4. Mass spectra of side chain cleaved product of endogenous glucocorticoids screened after incubation with *P. lymphophilum*. Bolded mass ions represent -60.02 amu less than the original m/z of (a) cortisol, (b) cortisone, and (c) allo-tetrahydrocortisol.

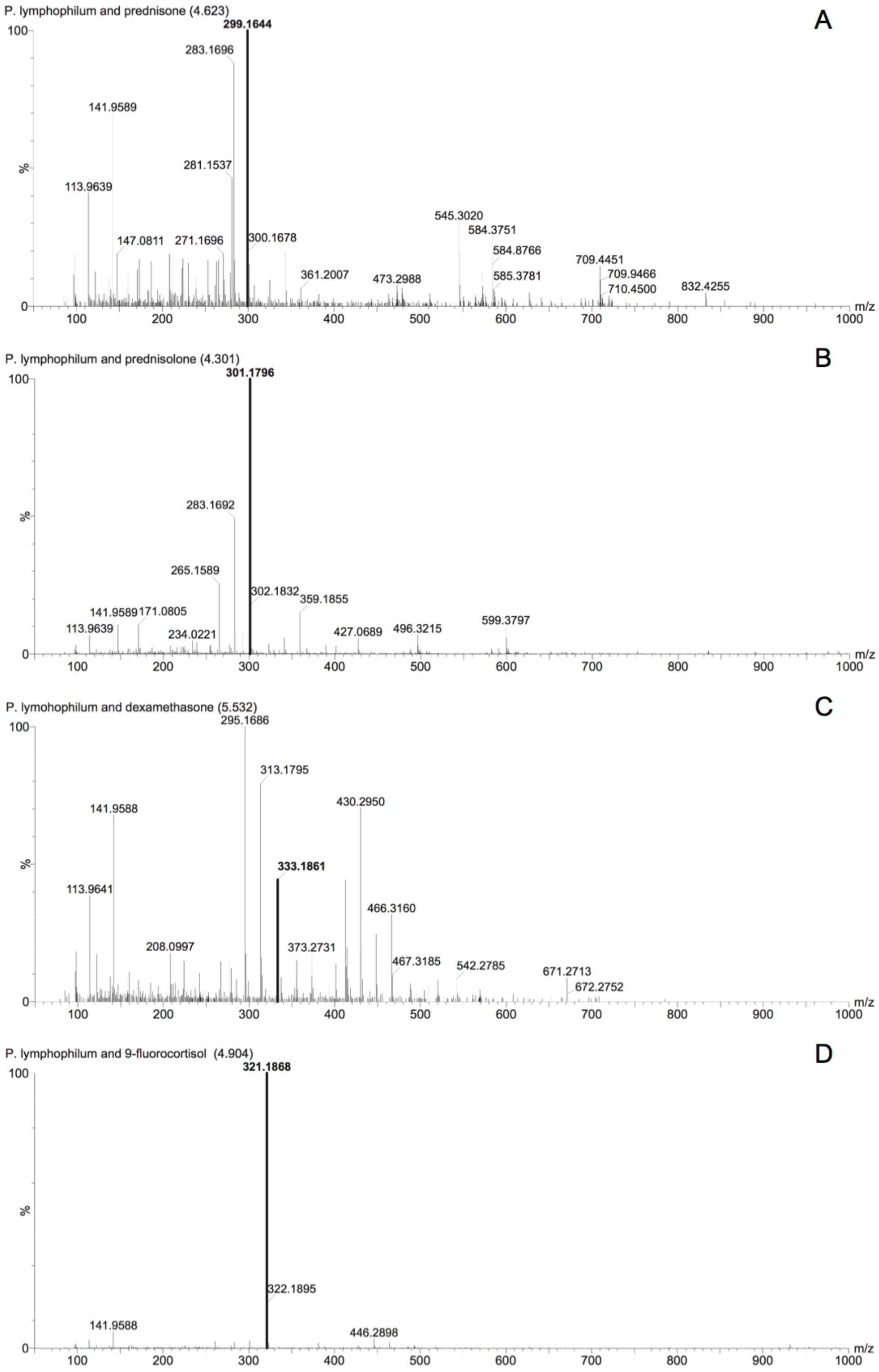

Figure S5. Mass spectra of side chain cleaved product of exogenous glucocorticoids screened after incubation with *P. lymphophilum*. Bolded mass ions represent -60.02 amu less than the original m/z of (a) prednisone, (b) prednisolone, (c) dexamethasone, and (d) 9-fluorocortisol.

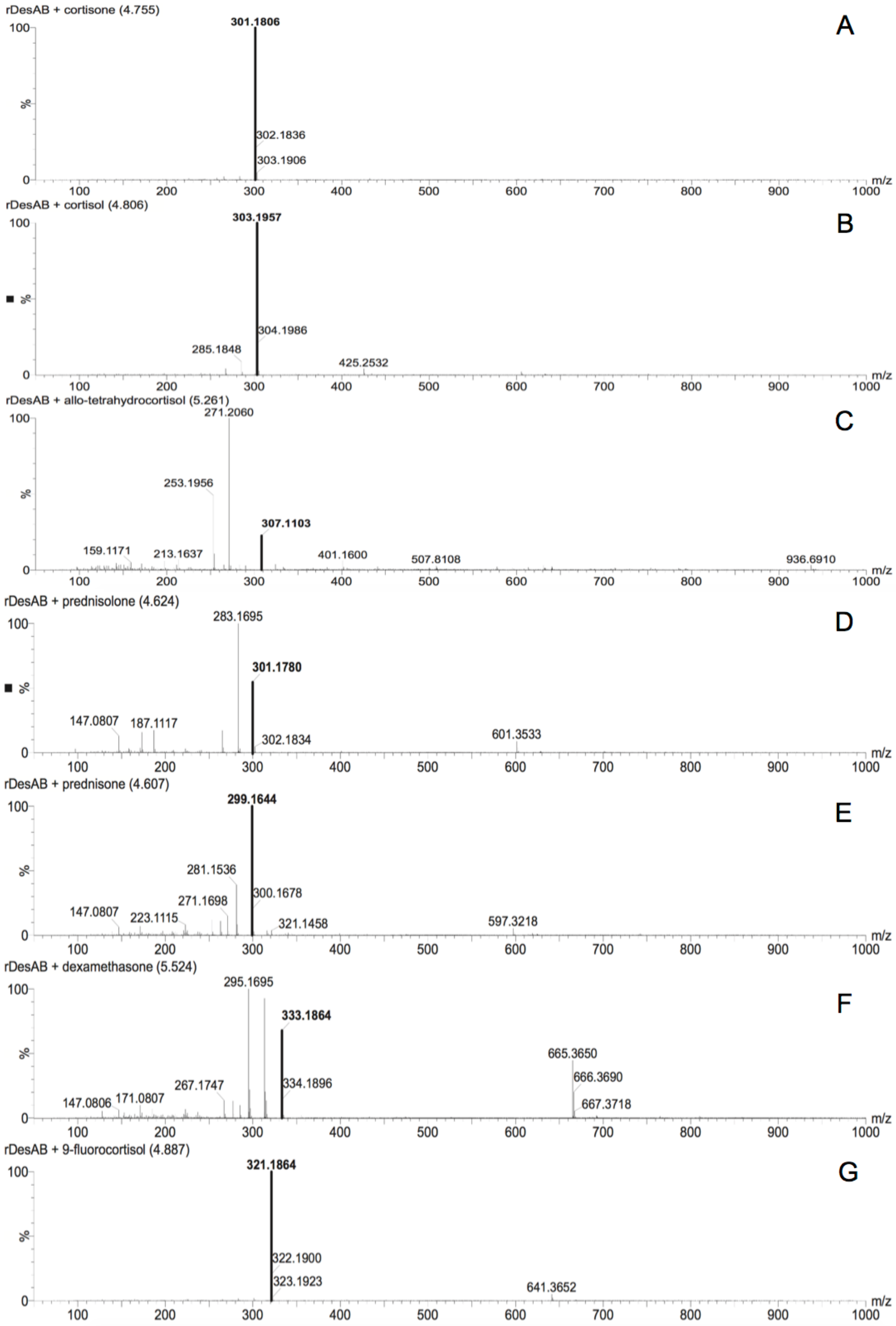

Figure S6. Mass spectra of side-chain cleaved product of all glucocorticoids screened after incubation with pure recombinant DesAB. Bolded mass ions represent -60.02 amu less than the original m/z of (a) cortisone, (b) cortisol, (c) allo-tetrahydrocortisol, (d) prednisolone, (e) prednisone, (f) dexamethasone, and (g) 9-fluorocortisol.