

SUPPLEMENTARY INFORMATION

Supplementary Table 1 Data collection, phasing and refinement statistics

	GA ₄ -GID1A-DELLA Native	GA ₃ -GID1A-DELLA Native	GA ₃ -GID1A-DELLA SeMet
Data collection			
Space group	<i>P</i> ₄ ₃ ₂ ₁ ₂	<i>P</i> ₄ ₃ ₂ ₁ ₂	<i>P</i> ₄ ₃ ₂ ₁ ₂
Cell dimensions			
<i>a</i> , <i>b</i> , <i>c</i> (Å)	81.8, 81.8, 129.9	82.0, 82.0, 130.1	82.1, 82.1, 129.8
			<i>Peak</i>
Wavelength (Å)	1.00000	0.99533	0.97938
Resolution (Å)	50-1.8 (1.86-1.80)*	50-1.8 (1.86-1.80)	20-2.0 (2.07-2.00)
<i>R</i> _{sym}	6.5 (6.8)	5.9 (12.9)	6.4 (11.9)
<i>I</i> / <i>σ</i> <i>I</i>	71.4 (58.8)	65.8 (28.6)	72.2 (39.7)
Completeness (%)	99.0 (98.2)	100 (98.4)	100 (100)
Redundancy	13.3 (11.2)	13.4 (14.4)	15.4 (15.3)
Refinement			
Resolution (Å)	50.0-1.8	50.0-1.8	
No. reflections	40,855	39,256	
<i>R</i> _{work} / <i>R</i> _{free}	19.8/22.1	20.5/22.9	
No. atoms			
Protein	3,156	3,134	
Ligand/ion	24	25	
Water	267	216	
B-factors			
Protein	19.10	22.83	
Ligand/ion	11.04	17.75	
Water	24.32	27.28	
R.m.s deviations			
Bond lengths (Å)	0.005	0.005	
Bond angles (°)	1.21	1.18	

One crystal was used for each data set.

*Values in parenthesis are for highest resolution shell.

Supplementary Table 2 Lists of hydrogen bonds between GA and GID1A

GA atom	GID1A residue/atom	GA ₃ -GID1A-DELLA	GA ₄ -GID1A-DELLA
		distance for GA ₃	distance for GA ₄
O3-1	H ₂ O	2.80 Å	2.89 Å
O3-1	Y127/OH	2.83 Å	2.75 Å
O7-1	S116/N	3.01 Å	2.96 Å
O7-1	S116/O _γ	2.68 Å	2.71 Å
O7-1	S191/O _γ	3.28 Å	3.23 Å
O7-2	S191/O _γ	2.95 Å	3.17 Å
O7-2	H ₂ O	2.65 Å	2.71 Å
O19-1	G320/N	2.87 Å	2.90 Å
O13	F238/O	3.25 Å	- (GA ₄ has no O13)
O13	H ₂ O	3.15 Å	- (GA ₄ has no O13)

Figure S1 Structural comparison of the GA₃- and GA₄-GID1A-DELLA complexes.

a, Chemical structures of bioactive GA₁ and GA₇. The bioactive GA molecules conserve one 7-carboxylate, 3-hydroxyl and one lactone (a cyclic carboxylester) moiety. The 3-hydroxyl group is introduced by GA 3-oxidase at the final step to produce active GA derivatives.

b, Structural comparison of the GA₃- and GA₄-GID1A-DELLA complexes. The GA₃-GID1A-DELLA complex is superimposed on the GA₄-GID1A-DELLA complex (grey). The GA₃-GID1A-DELLA complex is colored in pink (GAI DELLA domain), light blue (GID1A) and green (GA₃). The GA₄-GID1A-DELLA complex is colored in light green (GAI DELLA domain), yellow-orange (GID1A) and magenta (GA₄). The overall mean rms deviation is extremely small, 0.18 Å, over all the C_α atom positions of aligned residues. The GID1A molecules superimpose with an averaged rms deviation of 0.14 Å over all the C_α atom positions and the DELLA domains superimpose with an averaged rms deviation of 0.25 Å.

c, Molecular surface of the GA₃-GID1A-DELLA complex with the GAI DELLA domain (pink), the GID1A N-terminal extension (cyan) and the GID1A α/β core domain (light blue).

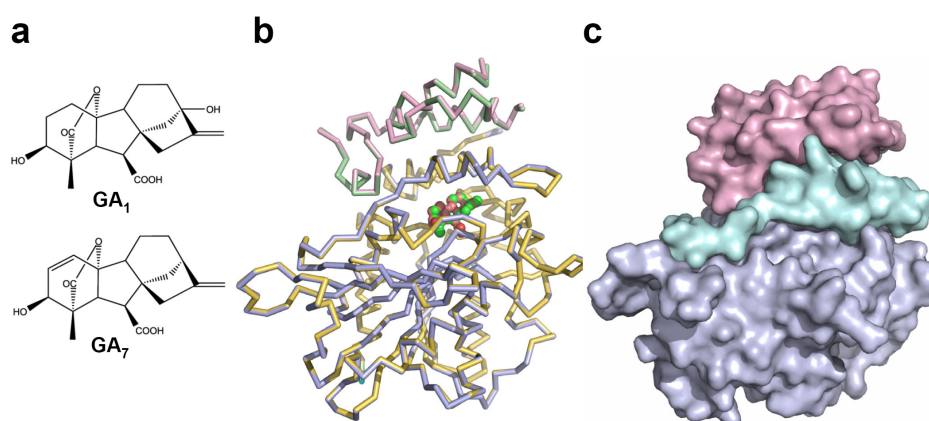


Figure S2 Structural comparison of GID1A and plant carboxylesterase CXE1. *Arabidopsis thaliana* GID1A is superimposed on *Actinidia eriantha* carboxylesterase CXE1 (from kiwifruits, PDB code 2O7V). The core domains superimpose well with an averaged rms deviation of 1.61 Å over the C_α atom positions of 148 out of 345 aligned residues without insertion and deletion segments. Tube diagrams of GID1A and CXE1 are shown in light blue and grey, respectively. Insertions in GID1A and CXE1 are highlighted in blue and red, respectively. Compared with CXE1, GID1A has 3 insertion and 4 deletion sites, where the N-terminal extension residues play a role in DELLA binding, although most of the other sites are located at the C-terminal edge of the β-sheet away from either the DELLA- or GA-binding sites. CXE1 lacks the corresponding helices found in the GID1A N-terminal extension, and instead contains a short pair of antiparallel strands that fail to completely cover the pocket.

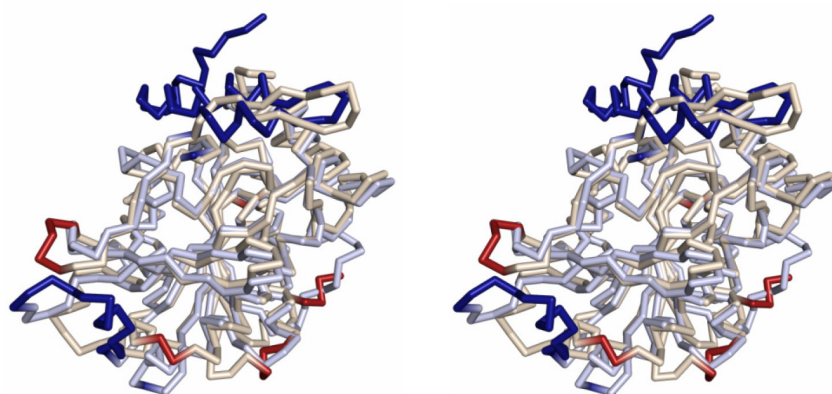


Figure S3 Sequence alignment of GID1s and a plant carboxylesterase.

Structural-based sequence alignment of GID1 proteins, *A. thaliana* GID1A, B and C, *Oryza sativa*, (rice) GID1 (Os GID1), and *A. eriantha* (kiwifruit) carboxylesterase CXE1 (Ae CXE1, PDB code 2O7V). The secondary structures of GID1A are shown at the top with α -helices (yellow bars), 3_{10} -helices (orange bars), β -strands (green arrows) and disordered residues (dotted lines). The secondary structures of CXE1 are shown at the bottom. The N-terminal extension of GID1A comprises three helices, α_a (residues 9-13), α_b (18-34) and α_c (43-49), followed by loop α_c - β_1 (residues 51-62). The GID1A core domain consists of a central eight-stranded mixed β -sheet (β_1 - β_8) and seven α -helices (α_1 - α_7) and two 3_{10} helices (η_1 and η_2). GID1A lacks three α -helices corresponding to α_3 , α_5 , and α_8 (grey bars) of CXE1 and one 3_{10} -helix corresponding to η_1 of CXE1. Instead of α_3 of CXE1, GID1A has a 3_{10} -helix η_1 . Bold letters indicate important residues for GA₃ and GA₄ binding (blue circles), DELLA binding (black circles), and the catalytic triad and oxyanion hole found in carboxylesterase (red letters). The triad residues are Ser191 (loop β_5 - α_3), Val319 (loop β_8 - α_7) and Asp289 (at loop β_7 - α_6). GA-interacting residues are highlighted in light green and boxed with bold lines. Identical residues to GID1A are highlighted in pale yellow. Accession numbers: GID1A, At3g05120; GID1B, At3g63010; GID1C, At5g27320; Os GID1, AB211399. GID1A and CXE1 share 22.5% sequence identity, whereas no homology at the N-terminal extension (62 residues) is detected. Among GID1 members, however, the N-terminal extensions are highly homologous, implying a specific function as the gibberellin receptor.

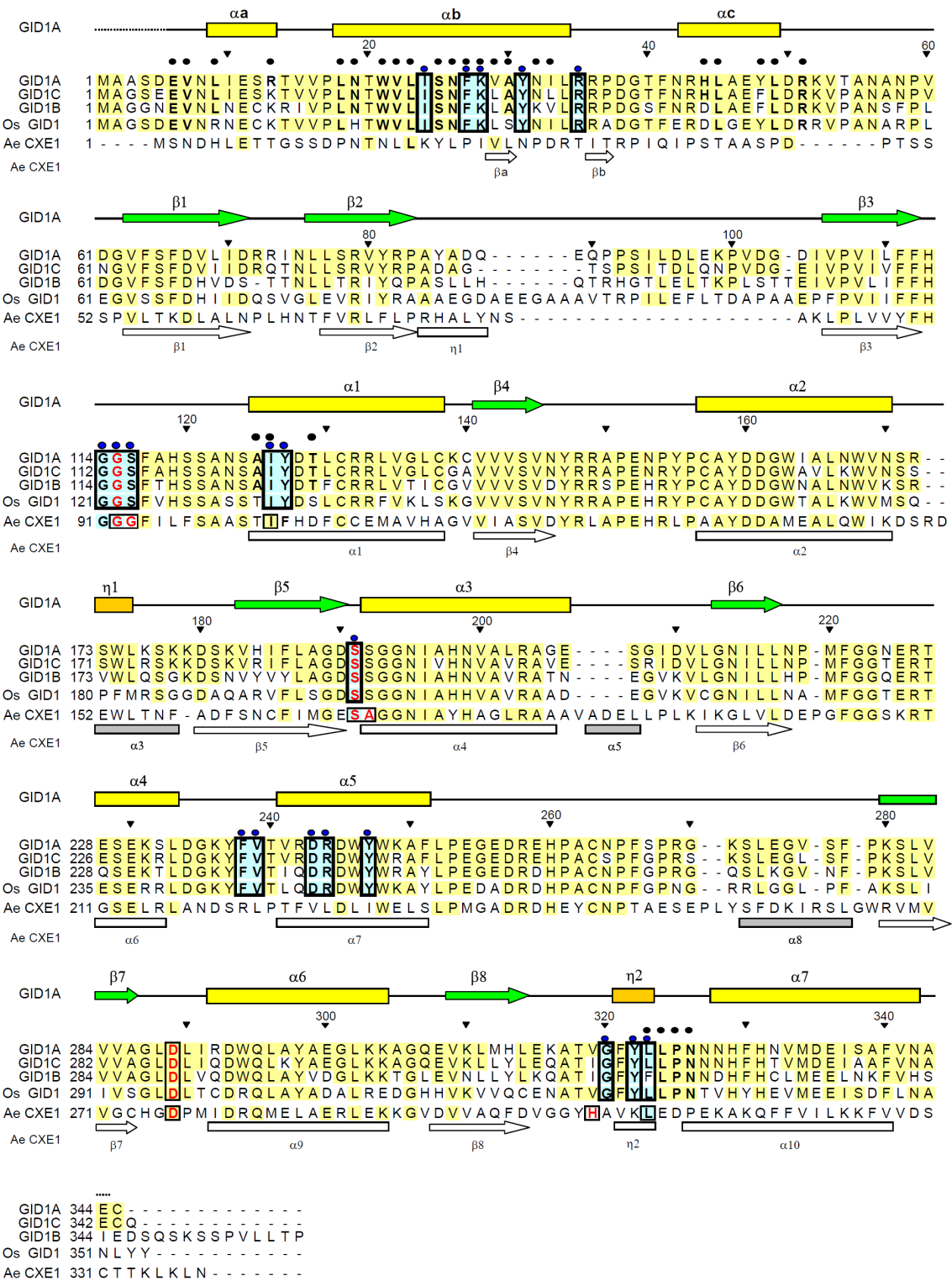


Figure S3

Figure S4 Schematic representation of the interactions. Summary of intermolecular interactions between GID1A and GA₃ in the GA₃-GID1A-DELLA complex. Van der Waals contacts (dotted lines), hydrogen bonds (arrows) and a salt bridge (an arrow with a dotted line) are shown. Residue side chains that participate in the interactions are indicated by boxes with bold lines. The N-terminal extension residues of GID1A are highlighted (cyan). All the interactions are conserved in those with GA₄ in the GA₄-GID1A-DELLA complex. Basically, contacts between hydrogen donor and acceptor atoms are nominated for hydrogen bonds with the typical distances (for example, 2.9 Å for H-bond between N-H and O) plus/minus 0.2 Å and the angles (for example, C-N---O and N---O-C) larger than 90°.

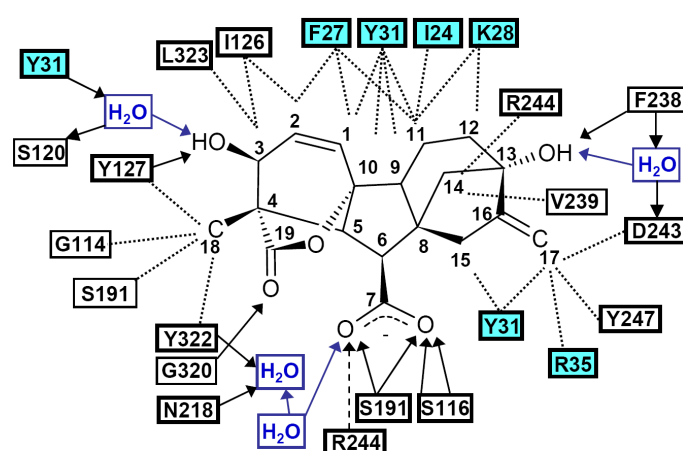


Figure S5 A stereo-view of GA perception in the GA₄-GID1A-DELLA complex. GID1A residues that contact GA₄ are highlighted in yellow (the core domain) or cyan (the N-terminal extension) and shown as stick models with van der Waals surfaces represented by dots. Water molecules are shown as blue spheres. Hydrogen bonds are indicated with dotted lines. Atom codes are oxygen in red and nitrogen in blue.

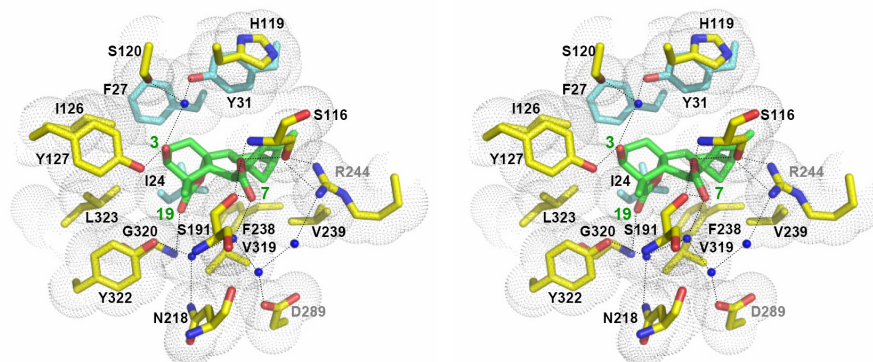


Figure S7 Effects of mutation of the DELLA domain on GID1A binding. Pull-down binding assays of mutated DELLA domains of GAI with GA₃-bound GID1A were performed with single mutations of the DELLA (Leu31, Tyr36 and Val38) and the LExLE (Met43, Glu51 and Glu54) motifs, and double mutations of Leu31 and Val38, Tyr36 and Met43, Glu51 and Glu54. Eluted samples analyzed by SDS-PAGE.

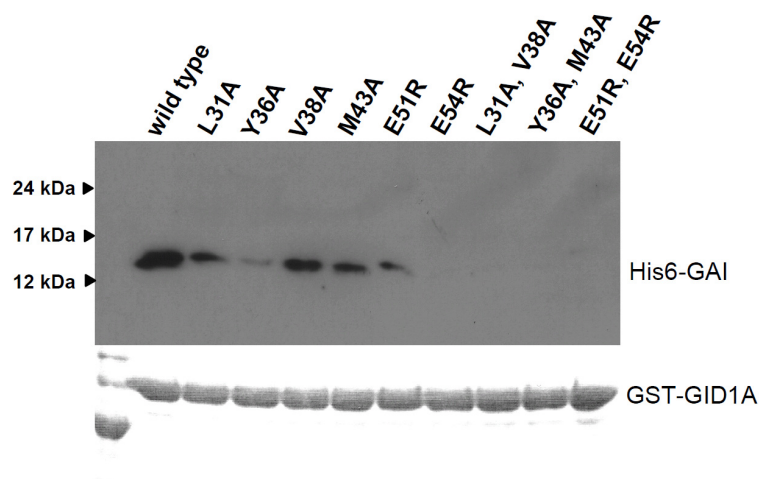


Figure S8 Conformational flexibility of the GID1A and the DELLA domain of GAI.

a, Conformational flexibility of the GID1 N-terminal extension. Proteolysis of GID1A in the free, GA₃-bound and DELLA-bound states. GID1A (residues 14-345) produced by trypsin-treatment of GST-GID1A was degraded rapidly within 10 min in the GA-unbound state, whereas resistant over 20-30 min in the GA-bound and DELLA-bound states.

b, Conformational flexibility of the DELLA domain of GAI. CD spectra of the DELLA domain. No significant changes were observed in the spectra at 20, 37, 50 and 70°C. Secondary structure estimations suggest a 100% random coil structure.

