PLANT SIGNALING & BEHAVIOR
2018, VOL. 13, NO. 3, e1445933 (3 pages)
https://doi.org/10.1080/15592324.2018.1445933

Taylor & Francis
Taylor & Francis Group

MINI-REVIEW

DELLA-dependent and -independent gibberellin signaling

Takeshi Ito, Kanako Okada, Jutarou Fukazawa, and Yohsuke Takahashi

Department of Biological Science, Graduate School of Science, Hiroshima University, Kagamiyama, Higashi-Hiroshima, Japan

ABSTRACT

DELLA proteins act as negative regulators in gibberellin (GA) signal transduction. GA-induced DELLA degradation is a central regulatory system in GA signaling pathway. Intensive studies have revealed the degradation mechanism of DELLA and the functions of DELLA as a transcriptional regulator. Meanwhile, recent studies suggest the existence of a DELLA-independent GA signaling pathway. In this review, we summarized the DELLA-independent GA signaling pathway together with the well-analyzed DELLA-dependent pathway.

ARTICLE HISTORY
Received 31 January 2018
Accepted 22 February 2018

KEYWORDS
Gibberellin; DELLA; calcium

Gibberellins (GAs) are phytohormones that control diverse aspects of plant growth and development, including seed germination, stem elongation, leaf expansion, and flower and seed development.[1] DELLA proteins act as growth repressors by inhibiting GA signaling in response to developmental and environmental cues.[2] Rice (*Oryza sativa*) contains one DELLA, SLENDER1, while *Arabidopsis thaliana* contains five DELLAs, GIBBERELLIN-INSENSITIVE (GAI), REPRESSOR OF *ga1–3* (RGA), RGA-LIKE1 (RGL1), RGL2, and RGL3, which display partially overlapping but distinct functions in repressing GA responses.[3-7] SLEEPY1 (SLY1) and GIBBERELLIN INSENSITIVE DWARF2 (GID2) are F-box proteins in *Arabidopsis* and rice, respectively.[8-10] Upon GA-binding to a soluble GA receptor, GIBBERELLIN INSENSITIVE DWARF1 (GID1), DELLAs are recruited to the $SCF^{SLY1/GID2}$ ubiquitin E3 ligase complex for polyubiquitination and is subsequently degraded by the 26S proteasome.[11-13] Proteolysis-independent regulations of DELLA have been also reported. GID1 overexpression in *sly1* mutant partially rescues dwarf and infertility phenotypes without degradation of DELLAs suggesting the proteolysis-independent downregulation of DELLAs.[14] A portion of DELLAs is conjugated to the small ubiquitin-like modifier (SUMO) protein.[15] GA-independent interaction of SUMOylated DELLAs with GID1 causes an accumulation of non-SUMOylated DELLAs, restraining plant growth.

DELLAs modulate gene expression by interacting with transcription factors (reviewed in refs[16,17]). For example, DELLAs interact with PHYTOCHROME INTERACTING FACTORs (PIFs) and BRASSINAZOLE RESISTANT1 (BZR1) to inhibit their DNA-binding activity, resulting in repression of hypocotyl elongation.[18-20] DELLAs also interact with jasmonic acid ZIM-domain proteins (JAZs), causing the release of MYC2 from JAZs.[21] Moreover, DELLAs are recruited to the promoters by DNA-binding transcription factors, such as INDETERMINATE1 domain proteins (IDDs) and type-B ARABIDOPSIS RESPONSE REGULATORS (ARRs), and act as transcriptional coactivators.[22-26] SPINDLY (SPY) and its paralog, SECRET AGENT (SEC), affect the binding affinity of DELLA for PIFs and BZR1. SPY was identified in a genetic screen for negative regulators of GA signaling in *Arabidopsis*.[27] Loss-of-function *spy* alleles are able to germinate in the presence of GA biosynthesis inhibitors, and to partially rescue the dwarf phenotype of GA-deficient mutants. Based on sequence comparison, *SPY* and *SEC* were predicted to encode *O*-linked *N*-acetylglucosamine (*O*-GlcNAc) transferases.[28,29] Recent studies revealed that SPY and SEC act as an *O*-fucosyltransferase and an *O*-GlcNAc transferase of DELLA proteins, respectively.[30,31] *O*-fucosylation by SPY promotes the interaction of DELLA with PIFs and BZR1, whereas *O*-GlcNAcylation by SEC inhibits these interaction. In addition to these glycosylation, phosphorylation and dephosphorylation are reported to inhibit and promote the degradation of DELLA, respectively,[32,33] but it is unclear whether phosphorylation state affects *O*-fucosylation and *O*-GlcNAcylation of DELLA.

GA-induced degradation of DELLA proteins acts as a central regulatory switch for GA signal transduction. However, some studies suggest the existence of a DELLA-independent GA pathway. Fruit growth in *Arabidopsis* is partially determined by a DELLA-independent GA response, and this response requires both GID1-mediated GA reception and 26S proteasome activity.[34] A microarray study using the *Arabidopsis* quadruple-DELLA mutant, which lacks *GAI, RGA, RGL1*, and *RGL2*, suggested that some GA-regulated genes are not regulated by DELLAs.[35] Tomato (*Solanum lycopersicum*) has only one DELLA gene, *PROCERA* (*PRO*). RNA sequencing using the *pro* mutant suggests that 5% of all GA-regulated genes in tomato are DELLA independent.[36]

Besides these studies, we also found a DELLA-independent GA pathway.[37] Ca^{2+} is a ubiquitous second messenger involved in signal transduction of various environmental and developmental stimuli in eukaryote.[38,39] One physiological response of plant cells to GAs is increase in cytosolic Ca^{2+} ($[Ca^{2+}]_{cyt}$).[40] An increase in $[Ca^{2+}]_{cyt}$ was observed previously after several hours of GA application.[41] We reexamined the effects of GAs on $[Ca^{2+}]_{cyt}$ using the Ca^{2+} sensor protein aequorin in *Arabidopsis*.[37] $[Ca^{2+}]_{cyt}$ increased

CONTACT Takeshi Ito itotakes@hiroshima-u.ac.jp Department of Biological Science, Graduate School of Science, Hiroshima University, Kagamiyama 1-3-1, Higashi-Hiroshima City, 739-8526, Japan.

within a few minutes of GA treatment, even in transgenic plants expressing a mutated degradation-resistant version of RGA (RGAΔ17) and in *della* pentuple mutant plants. These results suggested that the GA-induced increase in $[Ca^{2+}]_{cyt}$ occurs via a DELLA-independent pathway.

Ca^{2+}-dependent protein kinases (CDPKs) are multifunctional Ser/Thr protein kinases possessing both a Ca^{2+}-sensing function and kinase activity within a single gene product and found only in plants and some protozoans.[42-44] Tobacco (*Nicotiana tabacum*) NtCDPK1 is involved in GA feedback regulation through the phosphorylation of the transcription factor, REPRESSION OF SHOOT GROWTH (RSG).[45-47] The translocation of RSG from the nucleus to the cytoplasm is promoted by NtCDPK1.[47,48] We previously found that NtCDPK1 is phosphorylated in response to GAs in plants and autophosphorylated in vitro.[47,49,50] NtCDPK1 is possibly activated by the increase in $[Ca^{2+}]_{cyt}$ via a DELLA-independent GA pathway.

Unlike DELLA proteins, GID1 GA receptors are localized in both the nucleus and the cytoplasm.[11] Therefore, GID1 could be involved in GA-induced increase in $[Ca^{2+}]_{cyt}$. GID1-GA complex in the cytoplasm could cause the activation of Ca^{2+} channels or promote the degradation of a Ca^{2+} channel repressor via the ubiquitin-proteasome pathway, similar to DELLAs, leading to the GA-induced increase in $[Ca^{2+}]_{cyt}$. Another possibility is that an unidentified accessory GA receptor other than GID1 is involved in the GA-induced increase in $[Ca^{2+}]_{cyt}$. Although GID1 plays essential roles in GA signaling, physiological studies suggest an alternative signaling pathway related to a membrane-localized GA receptor.[41,51] Therefore, determining the involvement of GID1 in the GA-induced increase in $[Ca^{2+}]_{cyt}$ is an important issue. In addition to GID1 receptors, SPY is also reported to be a nucleocytoplasmic protein. Nuclear-export signal-fused SPY complements the *spy* mutation, but nuclear-localization signal-fused SPY does not.[52] This result suggests a DELLA-independent function of cytosolic SPY. It is unlikely that SPY is involved in GA reception, but GA-induced increase in $[Ca^{2+}]_{cyt}$ might possibly affect SPY activity in cytoplasm. The importance of DELLA-independent pathway in GA signaling is still largely unknown. Further studies of DELLA-dependent and -independent GA signaling will not only provide new insights into each signaling pathway but also promote better understanding of each other.

Disclosure of potential conflicts of interest

No potential conflicts of interest were disclosed.

Funding

This work was supported by the Ministry of Education, Culture, Sports, Science and Technology (24118004) Japan Society for the Promotion of Science (17K15143) Japan Society for the Promotion of Science (17K07449) Japan Society for the Promotion of Science (15H04392).

ORCID

Takeshi Ito http://orcid.org/0000-0002-5648-1371
Jutarou Fukazawa http://orcid.org/0000-0002-0048-4821
Yohsuke Takahashi http://orcid.org/0000-0002-3851-8783

References

1. Yamaguchi S. Gibberellin metabolism and its regulation. Annu Rev Plant Biol. 2008;59:225–51. doi:10.1146/annurev.arplant.59.032607.092804.
2. Sun TP. The molecular mechanism and evolution of the GA-GID1-DELLA signaling module in plants. Curr Biol. 2011;21:R338–45. doi:10.1016/j.cub.2011.02.036.
3. Dill A, Sun T. Synergistic derepression of gibberellin signaling by removing RGA and GAI function in *Arabidopsis thaliana*. Genetics. 2001;159:777–85.
4. King KE, Moritz T, Harberd NP. Gibberellins are not required for normal stem growth in *Arabidopsis thaliana* in the absence of GAI and RGA. Genetics. 2001;159:767–76.
5. Lee S, Cheng H, King KE, Wang W, He Y, Hussain A, Lo J, Harberd NP, Peng J. Gibberellin regulates Arabidopsis seed germination via RGL2, a GAI/RGA-like gene whose expression is up-regulated following imbibition. Genes Dev. 2002;16:646–58. doi:10.1101/gad.969002.
6. Cheng H, Qin L, Lee S, Fu X, Richards DE, Cao D, Luo D, Harberd NP, Peng J. Gibberellin regulates *Arabidopsis* floral development via suppression of DELLA protein function. Development. 2004;131:1055–64. doi:10.1242/dev.00992.
7. Tyler L, Thomas SG, Hu J, Dill A, Alonso JM, Ecker JR, Sun TP. DELLA proteins and gibberellin-regulated seed germination and floral development in Arabidopsis. Plant Physiol. 2004;135:1008–19. doi:10.1104/pp.104.039578.
8. Sasaki A, Itoh H, Gomi K, Ueguchi-Tanaka M, Ishiyama K, Kobayashi M, Jeong DH, An G, Kitano H, Ashikari M, et al. Accumulation of phosphorylated repressor for gibberellin signaling in an F-box mutant. Science. 2003;299:1896–8. doi:10.1126/science.1081077.
9. McGinnis KM, Thomas SG, Soule JD, Strader LC, Zale JM, Sun TP, Steber CM. The Arabidopsis *SLEEPY1* gene encodes a putative F-box subunit of an SCF E3 ubiquitin ligase. Plant Cell. 2003;15:1120–30. doi:10.1105/tpc.010827.
10. Dill A, Thomas SG, Hu J, Steber CM, Sun TP. The Arabidopsis F-box protein SLEEPY1 targets gibberellin signaling repressors for gibberellin-induced degradation. Plant Cell. 2004;16:1392–405. doi:10.1105/tpc.020958.
11. Ueguchi-Tanaka M, Ashikari M, Nakajima M, Itoh H, Katoh E, Kobayashi M, Chow T, Hsing YC, Kitano H, Yamaguchi I, et al. GIBBERELLIN INSENSITIVE DWARF1 encodes a soluble receptor for gibberellin. Nature. 2005;437:693–8. doi:10.1038/nature04028.
12. Ueguchi-Tanaka M, Nakajima M, Katoh E, Ohmiya H, Asano K, Saji S, Hongyu X, Ashikari M, Kitano H, Yamaguchi I, et al. Molecular interactions of a soluble gibberellin receptor, GID1, with a rice DELLA protein, SLR1, and gibberellin. Plant Cell. 2007;19:2140–55. doi:10.1105/tpc.106.043729.
13. Griffiths J, Murase K, Rieu I, Zentella R, Zhang ZL, Powers SJ, Gong F, Phillips AL, Hedden P, Sun TP, et al. Genetic characterization and functional analysis of the GID1 gibberellin receptors in *Arabidopsis*. Plant Cell. 2006;18:3399–414. doi:10.1105/tpc.106.047415.
14. Ariizumi T, Murase K, Sun TP, Steber CM. Proteolysis-independent downregulation of DELLA repression in *Arabidopsis* by the gibberellin receptor GIBBERELLIN INSENSITIVE DWARF1. Plant Cell. 2008;20:2447–59. doi:10.1105/tpc.108.058487.
15. Conti L, Nelis S, Zhang C, Woodcock A, Swarup R, Galbiati M, Tonelli C, Napier R, Hedden P, Bennett M, et al. Small Ubiquitin-like Modifier protein SUMO enables plants to control growth independently of the phytohormone gibberellin. Dev Cell. 2014;28:102–10. doi:10.1016/j.devcel.2013.12.004.
16. Davière JM, Achard P. A pivotal role of DELLAs in regulating multiple hormone signals. Mol Plant. 2016;9:10–20. doi:10.1016/j.molp.2015.09.011.
17. Van De Velde K, Ruelens P, Geuten K, Rohde A, Van Der Straeten D. Exploiting DELLA Signaling in Cereals. Trends Plant Sci. 2017;22:880–3. doi:10.1016/j.tplants.2017.07.010.
18. de Lucas M, Davière JM, Rodríguez-Falcón M, Pontin M, Iglesias-Pedraz JM, Lorrain S, Fankhauser C, Blázquez MA, Titarenko E, Prat S. A molecular framework for light and gibberellin control of cell elongation. Nature. 2008;451:480–4. doi:10.1038/nature06520.

19. Feng S, Martinez C, Gusmaroli G, Wang Y, Zhou J, Wang F, Chen L, Yu L, Iglesias-Pedraz JM, Kircher S, et al. Coordinated regulation of *Arabidopsis thaliana* development by light and gibberellins. Nature. 2008;451:475–9. doi:10.1038/nature06448.
20. Bai MY, Shang JX, Oh E, Fan M, Bai Y, Zentella R, Sun TP, Wang ZY. Brassinosteroid, gibberellin and phytochrome impinge on a common transcription module in *Arabidopsis*. Nat Cell Biol. 2012;14:810–7. doi:10.1038/ncb2546.
21. Hou X, Lee LYC, Xia K, Yan Y, Yu H. DELLAs modulate jasmonate signaling via competitive binding to JAZs. Dev Cell. 2010;19:884–94. doi:10.1016/j.devcel.2010.10.024.
22. Fukazawa J, Teramura H, Murakoshi S, Nasuno K, Nishida N, Ito T, Yoshida M, Kamiya Y, Yamaguchi S, Takahashi Y. DELLAs function as coactivators of GAI-ASSOCIATED FACTOR1 in regulation of gibberellin homeostasis and signaling in *Arabidopsis*. Plant Cell. 2014;26:2920–38. doi:10.1105/tpc.114.125690.
23. Fukazawa J, Ito T, Kamiya Y, Yamaguchi S, Takahashi Y. Binding of GID1 to DELLAs promotes dissociation of GAF1 from DELLA in GA dependent manner. Plant Signal Behav. 2015;10:e1052923. doi:10.1080/15592324.2015.1052923.
24. Fukazawa J, Mori M, Watanabe S, Miyamoto C, Ito T, Takahashi Y. DELLA-GAF1 complex is a main component in gibberellin feedback regulation of GA20 oxidase 2. Plant Physiol. 2017;175:1395–406. doi:10.1104/pp.17.00282.
25. Yoshida H, Hirano K, Sato T, Mitsuda N, Nomoto M, Maeo K, Koketsu E, Mitani R, Kawamura M, Ishiguro S, et al. DELLA protein functions as a transcriptional activator through the DNA binding of the indeterminate domain family proteins. Proc Natl Acad Sci U S A. 2014;111:7861–6. doi:10.1073/pnas.1321669111.
26. Marín-de la Rosa N, Pfeiffer A, Hill K, Locascio A, Bhalerao RP, Miskolczi P, Gronlund AL, Wanchoo-Kohli A, Thomas SG, Bennett MJ, et al. Genome wide binding site analysis reveals transcriptional coactivation of cytokinin-responsive genes by DELLA proteins. PLoS Genet. 2015;11:e1005337. doi:10.1371/journal.pgen.1005337.
27. Jacobsen SE, Olszewski NE. Mutations at the *SPINDLY* locus of Arabidopsis alter gibberellin signal transduction. Plant Cell. 1993;5:887–96. doi:10.1105/tpc.5.8.887.
28. Jacobsen SE, Binkowski KA, Olszewski NE. SPINDLY, a tetratricopeptide repeat protein involved in gibberellin signal transduction in *Arabidopsis*. Proc Natl Acad Sci U S A. 1996;93:9292–6. doi:10.1073/pnas.93.17.9292.
29. Hartweck LM, Scott CL, Olszewski NE. Two *O*-linked N-acetylglucosamine transferase genes of *Arabidopsis thaliana* L. Heynh. Have overlapping functions necessary for gamete and seed development. Genetics. 2002;161:1279–91.
30. Zentella R, Hu J, Hsieh WP, Matsumoto PA, Dawdy A, Barnhill B, Oldenhof H, Hartweck LM, Maitra S, Thomas SG, et al. *O*-GlcNAcylation of master growth repressor DELLA by SECRET AGENT modulates multiple signaling pathways in *Arabidopsis*. Genes Dev. 2016;30:164–76. doi:10.1101/gad.270587.115.
31. Zentella R, Sui N, Barnhill B, Hsieh W, Hu J, Shabanowitz J, Boyce M, Olszewski NE, Zhou P, Hunt DF, et al. The Arabidopsis *O*-fucosyltransferase SPINDLY activates nuclear growth repressor DELLA. Nat Chem Biol. 2017;13:479–85. doi:10.1038/nchembio.2320.
32. Dai C, Xue H. Rice early flowering1, a CKI, phosphorylates DELLA protein SLR1 to negatively regulate gibberellin signalling. EMBO J. 2010;29:1916–27. doi:10.1038/emboj.2010.75.
33. Hussain A, Cao D, Cheng H, Wen Z, Peng J. Identification of the conserved serine/threonine residues important for gibberellin-sensitivity of Arabidopsis RGL2 protein. Plant J. 2005;44:88–99. doi:10.1111/j.1365-313X.2005.02512.x.
34. Fuentes S, Ljung K, Sorefan K, Alvey E, Harberd NP, Østergaard L. Fruit growth in *Arabidopsis* occurs via DELLA-dependent and DELLA-independent gibberellin responses. Plant Cell. 2012;24:3982–96. doi:10.1105/tpc.112.103192.
35. Cao D, Cheng H, Wu W, Soo HM, Peng J. Gibberellin mobilizes distinct DELLA-dependent transcriptomes to regulate seed germination and floral development in Arabidopsis. Plant Physiol. 2006;142:509–25. doi:10.1104/pp.106.082289.
36. Livne S, Lor VS, Nir I, Eliaz N, Aharoni A, Olszewski NE, Eshed Y, Weiss D. Uncovering DELLA-independent gibberellin responses by characterizing new tomato *procera* mutants. Plant Cell. 2015;27:1579–94. doi:10.1105/tpc.114.132795.
37. Okada K, Ito T, Fukazawa J, Takahashi Y. Gibberellin induces an increase in cytosolic Ca^{2+} via a DELLA-independent signaling pathway. Plant Physiol. 2017;175:1536–42. doi:10.1104/pp.17.01433.
38. Sanders D, Pelloux J, Brownlee C, Harper JF. Calcium at the crossroads of signaling. Plant Cell. 2002;14:S401–17. doi:10.1105/tpc.002899.
39. Schuster S, Marhl M, Höfer T. Modelling of simple and complex calcium oscillations. From single-cell responses to intercellular signalling. Eur J Biochem. 2002;269:1333–55. doi:10.1046/j.0014-2956.2001.02720.x.
40. Bush DS, Biswas AK, Jones RL. Gibberellic-acid-stimulates Calcium accumulation in endoplasmic reticulum of barly aleurone: Calcium transport and steady-state levels. Planta. 1989;178:411–20. doi:10.1007/BF00391870.
41. Gilroy S, Jones RL. Gibberellic acid and abscisic acid coordinately regulate cytoplasmic calcium and secretory activity in barley aleurone protoplasts. Proc Natl Acad Sci U S A. 1992;89:3591–5. doi:10.1073/pnas.89.8.3591.
42. Harper JF, Sussman MR, Schaller GE, Putnam-Evans C, Charbonneau H, Harmon AC. A calcium-dependent protein kinase with a regulatory domain similar to calmodulin. Science. 1991;252:951–4. doi:10.1126/science.1852075.
43. Suen KL, Choi JH. Isolation and sequence analysis of a cDNA clone for a carrot calcium-dependent protein kinase: homology to calcium/calmodulin-dependent protein kinases and to calmodulin. Plant Mol Biol. 1991;17:581–90. doi:10.1007/BF00037045.
44. Harper JF, Harmon A. Plants, symbiosis and parasites: a calcium signalling connection. Nat Rev Mol Cell Biol. 2005;6:555–66. doi:10.1038/nrm1679.
45. Fukazawa J, Sakai T, Ishida S, Yamaguchi I, Kamiya Y, Takahashi Y. Repression of shoot growth, a bZIP transcriptional activator, regulates cell elongation by controlling the level of gibberellins. Plant Cell. 2000;12:901–15. doi:10.1105/tpc.12.6.901.
46. Fukazawa J, Nakata M, Ito T, Yamaguchi S, Takahashi Y. The transcription factor RSG regulates negative feedback of *NtGA20ox1* encoding GA 20-oxidase. Plant J. 2010;62:1035–45.
47. Ishida S, Yuasa T, Nakata M, Takahashi Y. A tobacco calcium-dependent protein kinase, CDPK1, regulates the transcription factor REPRESSION OF SHOOT GROWTH in response to gibberellins. Plant Cell. 2008;20:3273–88. doi:10.1105/tpc.107.057489.
48. Ishida S, Fukazawa J, Yuasa T, Takahashi Y. Involvement of 14-3-3 signaling protein binding in the functional regulation of the transcriptional activator REPRESSION OF SHOOT GROWTH by gibberellins. Plant Cell. 2004;16:2641–51. doi:10.1105/tpc.104.024604.
49. Ito T, Nakata M, Fukazawa J, Ishida S, Takahashi Y. Scaffold function of Ca^{2+}-dependent protein kinase: tobacco Ca^{2+}-DEPENDENT PROTEIN KINASE1 transfers 14-3-3 to the substrate REPRESSION OF SHOOT GROWTH after phosphorylation. Plant Physiol. 2014;165:1737–50. doi:10.1104/pp.114.236448.
50. Ito T, Ishida S, Oe S, Fukazawa J, Takahashi Y. Autophosphorylation affects substrate-binding affinity of tobacco Ca^{2+}-dependent protein kinase1. Plant Physiol. 2017;174:2457–68. doi:10.1104/pp.17.00515.
51. Hooley R, Beale M, Smith S. Gibberellin perception at the plasma membrane of *Avena fatua* aleurone protoplasts. Planta. 1991;183:274–80. doi:10.1007/BF00197799.
52. Maymon I, Greenboim-Wainberg Y, Sagiv S, Kieber JJ, Moshelion M, Olszewski N, Weiss D. Cytosolic activity of SPINDLY implies the existence of a DELLA-independent gibberellin-response pathway. Plant J. 2009;58:979–88. doi:10.1111/j.1365-313X.2009.03840.x.