FIN219-FIP1: Two Vital Proteins Involved in Cross-talk Between FR Light Signaling and JA Response

A FIP1-mediated FIN219 conformational change increases the rate of biosynthesis of JA-Ile that is an important JA derivative for plant-defense response.

In *Arabidopsis thaliana*, far-red (FR) insensitive 219 (FIN219) belongs to family Gretchen Hagen 3 (GH3) of amido synthetases and serves as a positive regulator in phytochrome A (phyA)-mediated FR signaling. FIN219, also called as jasmonic acid or jasmonate resistant 1 (JAR1; AtGH3.11), catalyzes conjugation between JA and amino acids, such as isoleucine (JA-Ile) and leucine (JA-Leu), in JA signaling for a response to plant defence. How FIN219/JAR1 couples FR light and JA signaling is still elusive. Preceding work showed that FIN219-interacting protein 1 (FIP1), a member of the GST tau family, interacts with FIN219 for further control of the FR light signaling,[1] whereas some other experiments (*e.g.* knockdown and knockout of FIP1, and microarray assays) indicate that FIP1 is strongly associated with JA signaling. Taken together, FIN219 and FIP1 are considered to be two key components to connect the FR light signaling and the JA response.[1,2] The structure of complex FIN219-JA-Ile that has been solved indicates that FIN219 belongs to an adenylate-forming enzyme and consists of an N-terminal domain containing an active site, a flexible hinge linker tuning the dynamic C-terminal domain for substrate binding. How FIN219 interacts with FIP1, and how FIN219-FIP1 is involved in the JA response

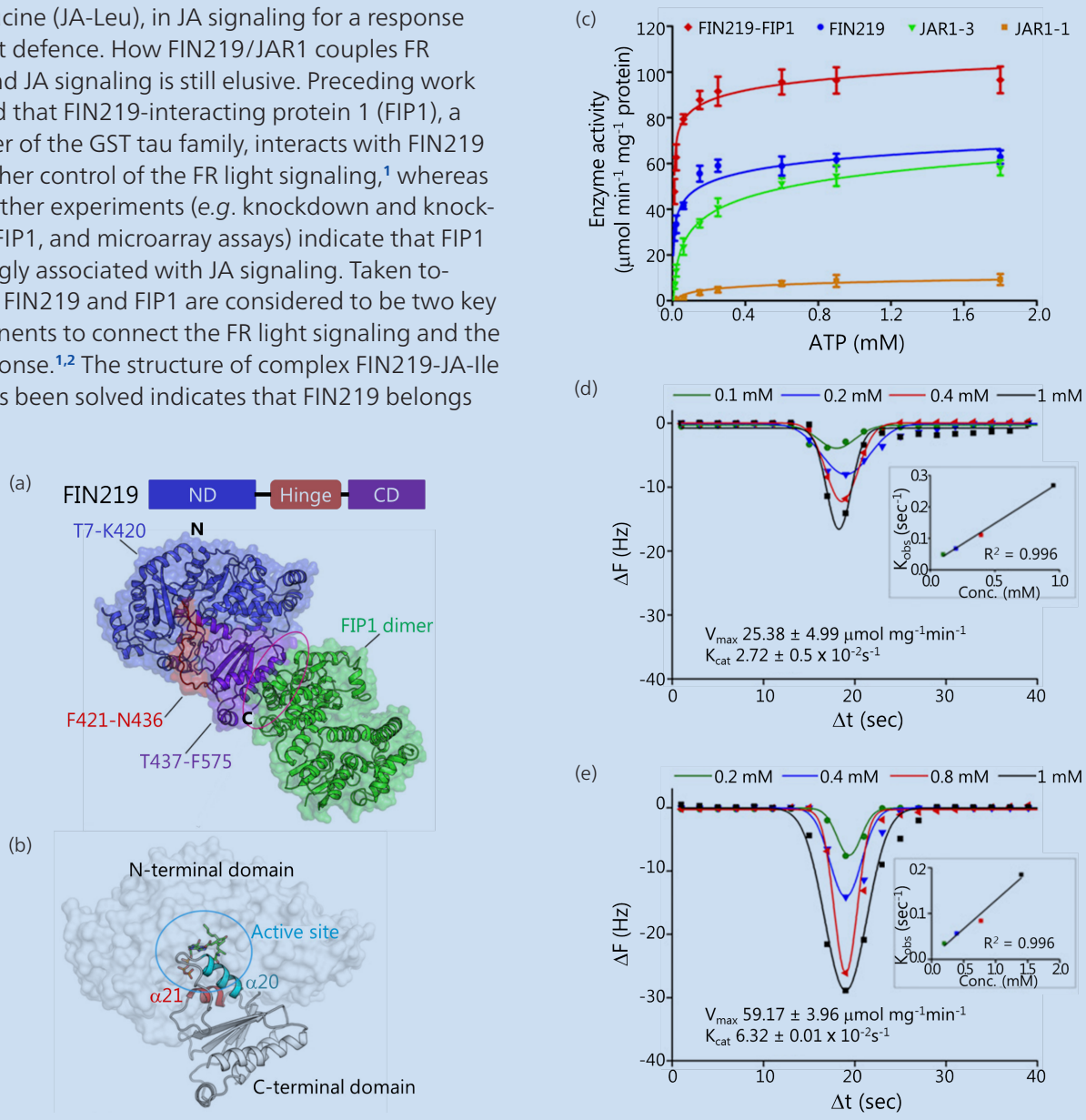

Fig. 1: (a) Overall structure of the FIN219-FIP1 complex. (b) The active site of FIN219 was occluded by its two helices, α20 and α21, of the C-terminal domain. (c) Adenylation activity assay of FIN219-FIP1, FIN219 and two mutants, JAR1-3 (E334K) and JAR1-1 (S101F). (d) & (e) Kinetic assays of adenylation of FIN219 or the FIN219-FIP1, respectively. [Reproduced from Ref. 3]

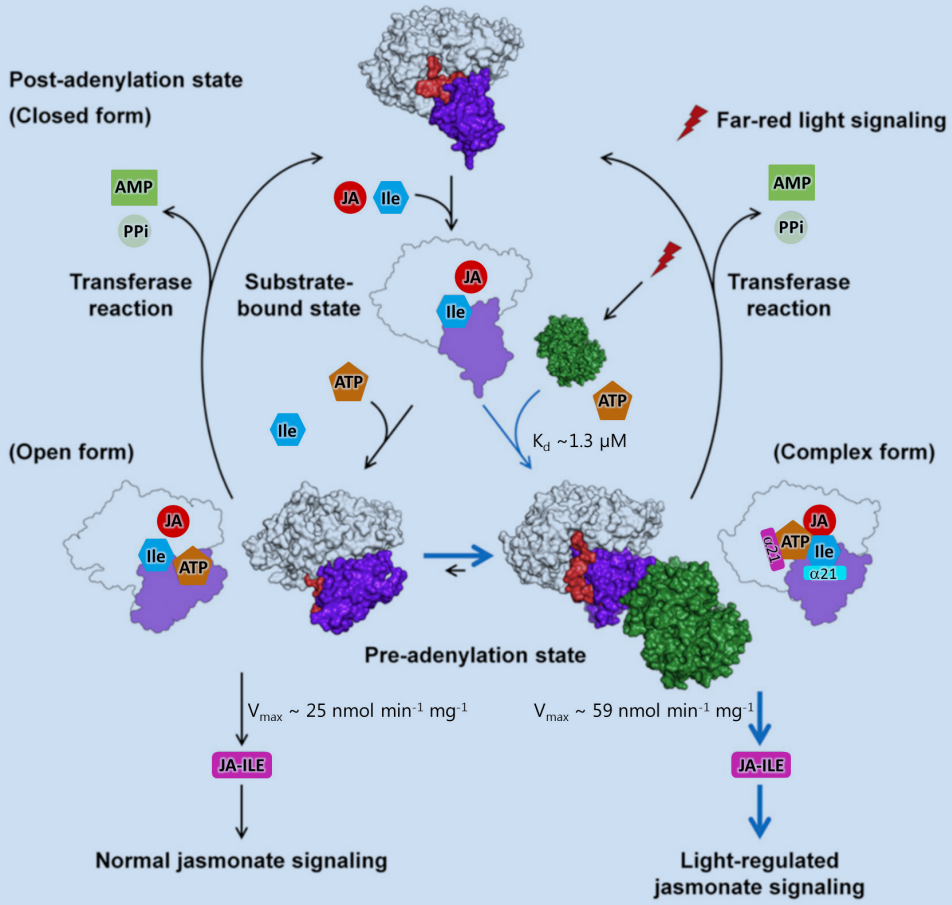

Fig. 2: Possible working model of how FIN219-FIP1 increases JA signaling under the FR light condition. [Reproduced from Ref. 3]

are still unclear. To investigate the binding mode between FIN219 and FIP1, and the catalytic mechanism, a research team led by Yi-Sheng Cheng (Department of Life Science, National Taiwan University) determined the structures of complex FIN219-FIP1 with various ligands – JA, ATP, Ile, Leu, Val, Met and Mg – at resolution 1.54–2.25 Å. The structure of FIP1 alone with native form was solved at resolution 1.65 Å. All diffraction data sets were collected at beamline **TLS 13C1** of NSRRC.[3]

According to **Fig. 1(a)**, the binding mode depicts clearly that the FIP1 dimer contacts the rotatable C-terminal domain (T437-F575) of FIN219. To distinguish the FIP1-bound FIN219 fold from the AMP- and ATP-bound FIN219 fold (closed and open forms), the authors named it the complex form. **Figure 1(b)** represents that a change in the orientation of the FIN219 C-terminal domain was observed upon FIP1 binding, which gives rise to the active site of FIN219 becoming blocked by two helices, α20 and α21, of the FIN219 C-terminal domain. After that, the re-orientated α20 structurally pushed the bound ATP into the interior of the active site for a more effective adenylation reaction. To understand whether FIP1 binding enhances the FIN219 adenylation activity, a kinetic assay was performed; the result confirms that FIN219-FIP1 has greater adenylation activity than FIN219 alone (**Fig. 1(c)**). The conformational change of FIP1-mediated FIN219 hence accelerates the rate of JA-Ile biosynthesis. The binding and catalytic assays demonstrate also that FIN219-FIP1 has a greater activity than FIN219 alone (**Figs. 1(d) and 1(e)**). On the basis of these data, the unique FIN219-FIP1 binding mode is critical for the improvement of the synthetase activity of FIN219.

Based on the structural information and the biochemical results, a working model was proposed to elucidate the FR light-coupled JA response (**Fig. 2**). Briefly, the proposed model shows that FIP1 (shown in green) is up-regulated under phyA-mediated FR light signaling followed by binding to FIN219 for higher JA-Ile catalytic activity *via* C-terminal domain switching. In summary, the unique binding between FIN219 and FIP1 provides an alternative path to enhance the JA signaling efficiently under a continuous FR light condition. (Reported by Chun-Hsiang Huang)

This report features the work of Yi-Sheng Cheng and his co-workers published in PNAS ***114****, E1815 (2017).*

TLS 13C1 Protein Crystallography

- MR, SIR, MIR
- Protein Crystallography

References

1. I. C. Chen, I. C. Huang, M. J. Liu, Z. G. Wang, S. S. Chung, and H. L. Hsieh, Plant Physiol. **143**, 1189 (2007).
2. D. P. Dixon, M. Skipsey, and R. Edwards, Phytochemistry **71**, 338 (2010)
3. C. Y. Chen, S. S. Ho, T. Y. Kuo, H. L. Hsieh, and Y. S. Cheng, PNAS. **114**, E1815 (2017).