

Poster Presentations

[MS5-P04] Structures of States of a Photoconvertible and Photoswitchable Fluorescent Protein Engineered from Dronpa.

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The structures of several states of a new photoconvertible and photoswitchable fluorescent protein (FP) engineered from Dronpa [1], called pcDronpa (photoconvertible Dronpa), were determined. Like IrisFP [2] and NijiFP [3], pcDronpa possesses both photoconvertible and photoswitching properties. However, pcDronpa shows the success of introducing photoconvertibility into the photoswitchable FP. The green fluorescent state (green-on state) of pcDronpa can switch reversibly to the nonfluorescent state (green-off state) by illumination with 488-nm and 405-nm light, respectively. The structural basis for this switching was proven by the crystal structure determination of the green-on and green-off states. The photoswitching of pcDronpa is as expected not different from that of Dronpa [4], because there is no alternation in the part of the chromophore responsible for the photoswitching, as well as in its proximate environment. Remarkably, pcDronpa can also convert to the red fluorescent state (red-on state) by illumination with 405-nm light. This property was introduced by rationally mutating the chromophore from Cys-Tyr-Gly to His-Tyr-Gly.

The structure of green-on pcDronpa was solved to a resolution of 1.95 Å (Rwork and Rfree values of 0.1840 and 0.2351, respectively). The green-on chromophore created from the His62-Tyr63-Gly66 triad is well defined, especially the new

imidazole moiety from the His62 substitution. As expected the original part of the chromophore displays the cis-planar configuration. The imidazole moiety is accepted by its environment without a significant rearrangement of the nearby residues. However, compared to the other photoconvertible FPs, the chromophore environment of pcDronpa is different at two positions, Met40 and Tyr116, which are placed in the proximate environment of the imidazole moiety. It is thus supposed that they play a role in the photoconvertible properties of pcDronpa.