STRUCTURAL
BIOLOGY

Volume 72 (2016)

Supporting information for article:

Crystal structure of the fluorescent protein from *Dendronephthya sp.* in both green and photoconverted red forms

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Molar extinction coefficients of the proteins were determined as following:

$$\varepsilon_A^{490} = \frac{A^{490}}{C_{Tot}} \text{ and } \varepsilon_P^{380} = \frac{A^{380}}{C_{Tot}},$$

where A^{490} and A^{380} the intensity of the protein 490 and 380 nm absorbance bands at pH 9 (all deprotonated) and 5 (all protonated), respectively and C_{Tot} is the total concentration of the protein determined by Ward method as:

$$C_{Tot} = \frac{A^{447}(pH13)}{44,000},$$

where $A^{447}(pH13)$ is the absorbance of the fully denatured protein at pH 13 (0.2 M NaOH) and 44,000 $M^{-1}cm^{-1}$ is the molar extinction coefficient of a fully denatured protein.

Protonation degree (α) of the protein at pH 7.5 was determined as:

$$\alpha = \frac{C_P}{C_{Tot}} = 1 - \frac{C_A}{C_{Tot}} = 1 - \frac{A^{490}(pH7.5)}{\varepsilon_A^{490} C_{Tot}},$$

where C_P and C_A correspond to the concentration of protonated and deprotonated DendFP at pH 7.5. We have used the value C_{Tot} of determined by Ward method since the protein in this experiment was freshly prepared and not exposed to any UV-blue light causing its photoconversion and photobleaching.

Capability of the mutant to absorb the UV-light at physiological pH:

$$\frac{C_P}{C_{Tot}}\varepsilon_P^{380} = \frac{A_{380}^{pH7.5}}{C_{Tot}} = \frac{A_{380}^{pH7.5}}{\frac{A_{447}^{pH13}}{\varepsilon_{pH13}^{447}}} = \frac{A_{380}^{pH7.5}}{A_{447}^{pH13}} \times 44,000$$

UV absorption capability of mutant relative to wild type (RA_{380}):

$$RA_{380} = \frac{A_{380}^{pH7.5}(mut)}{A_{447}^{pH13}(mut)} \Big/ \frac{A_{380}^{pH7.5}(wt)}{A_{447}^{pH13}(wt)}$$

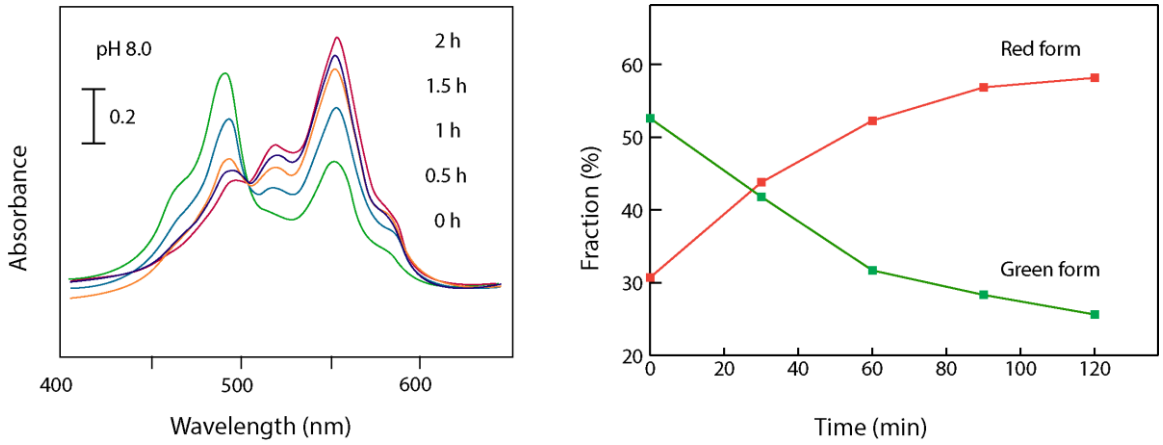

Figure S1 Photoconversion of DendFP. Absorbance spectra and change of green and red fractions of DendFP with time. 1 mg/ml green DendFP solution in 20 mM Tris pH 8.0, 200 mM NaCl exposed to 365 nm light (UVLMS-38 EL Series 3UV Lamp).

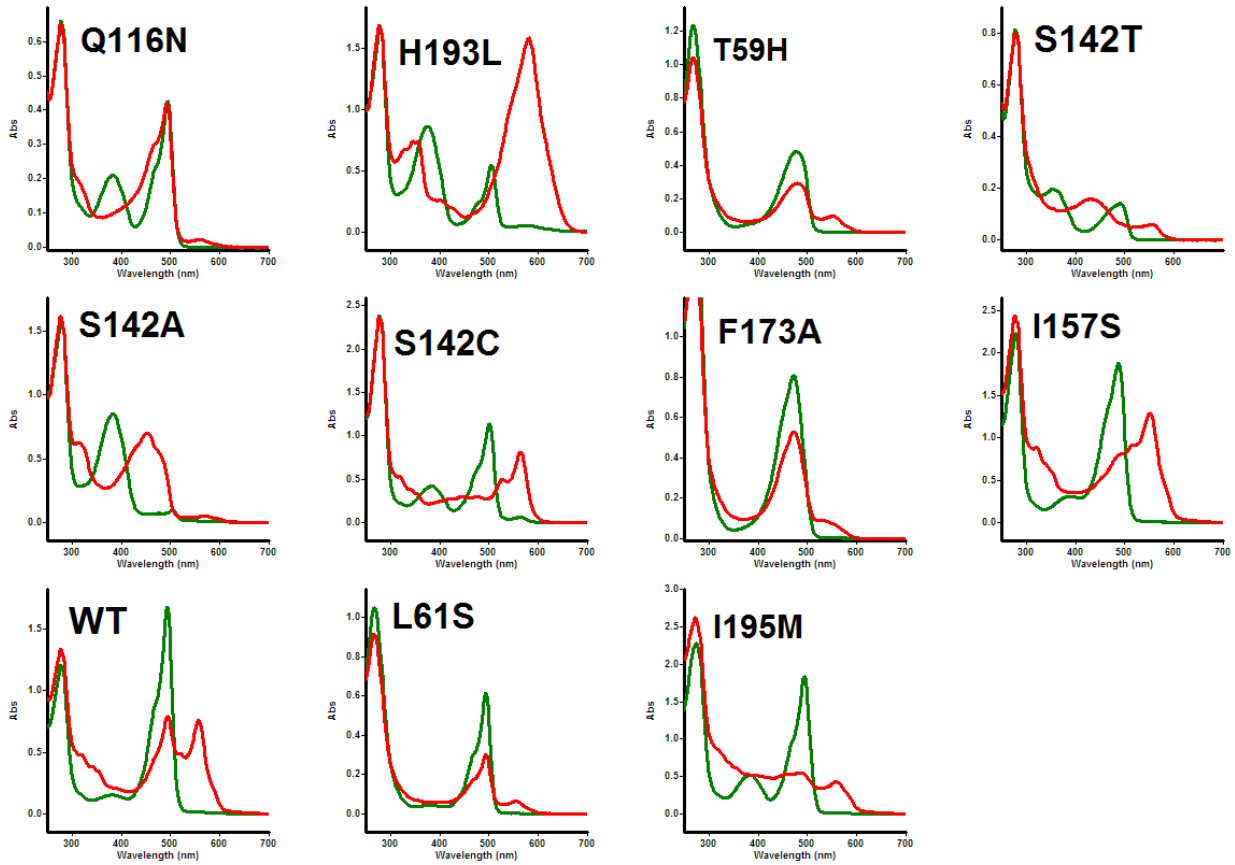

Figure S2 Absorption spectra of DendFP and its mutants before (green) and after (red) exposure to UV-light (3 h 4°C).

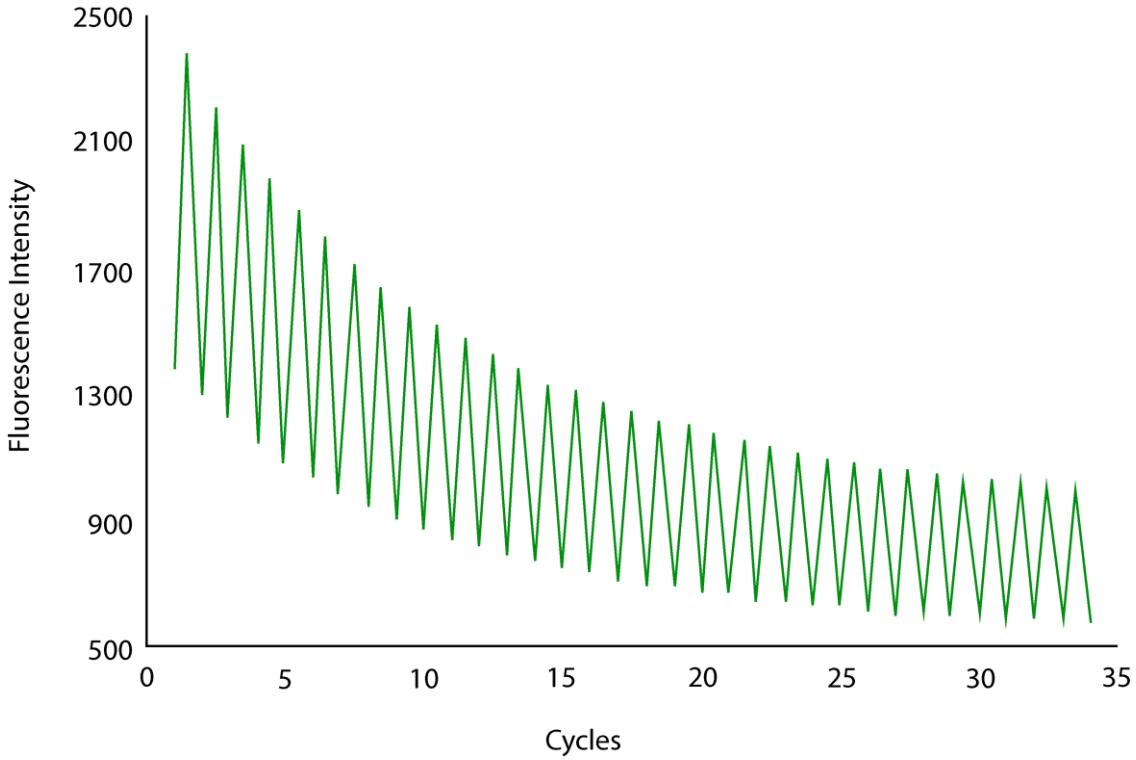

Figure S3 Photochromic behavior of Phe173Ala DendFP variant. The green species of Phe173Ala DendFP was switched OFF with 488 nm light and back ON with 405 nm light. A gradual decrease of the fluorescence intensity is, presumably, due to concomitant protein photobleaching.