**Supporting information**

# Autoinhibitory Feedback Control Over Photodynamic Action

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## 1. General Methods

All reagents and solvents were purchased from commercial sources and used without further purification. Compounds **BOD1** and **BOD2** were prepared using the literature procedures.(1, 2) Column chromatography was carried out using silica stationary phase (230–400 mesh, SiliCycle Inc., Canada). Analytical thin layer chromatography was performed on 0.25 mm thick precoated silica gel plates (60F254, Merck, Germany). Compounds were visualized under UV light. All $^{1}$H NMR spectra were recorded on a Varian Inova instrument (400 MHz) at Selçuk University, Konya. Chemical shifts (δ) are reported in parts per million (ppm) and referenced to the residual solvent peak. Coupling constants (*J*) are reported in hertz (Hz). Standard abbreviations indicating multiplicities are given: br = broad, d = doublet, m = multiplet, s = singlet, t = triplet. High-resolution mass spectrometry was carried out using Agilent 6530 Accurate-Mass Q-TOF LC/MS of the Eastern Anatolia Advanced Technology Research and Application Centre (DAYTAM, Erzurum, Turkey). For PDT, LED from Bright LED Electronics Corp. and model BL-BG43V4V with peak absorption value at 506 nm was used as a light source. For cell culture experiments DLD – 1 human colorectal adenocarcinoma cells (ATCC) were used.

## 2. Additional Figures and Tables

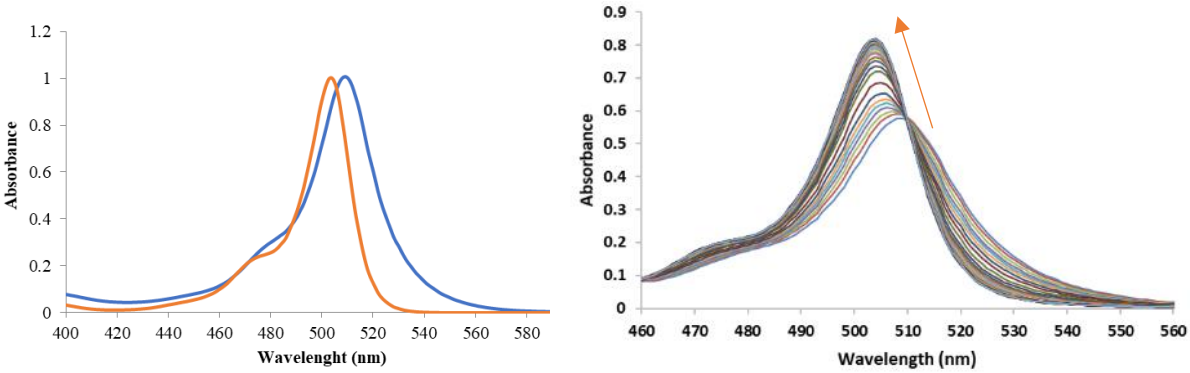


**Figure S1.** Normalized absorbance spectra of **BOD1** (blue) and **BOD2** (orange) in isopropanol (left). Change in the UV-Vis Absorption spectra of **BOD1** (10 μM) during PDT (right). Absorption peak located at 510 nm shifts to 505 nm which overlaps with the spectrum of **BOD2**. Sample is irradiated for 140 min with 506 nm LED Lamp.

In order to demonstrate that major effect of sodium ascorbate on PDT efficiency is not singlet oxygen scavenging, a control experiment was run. In the experiment, **BOD2** is used as PDT agent. Since this compound lacks boronate in the structure, it is not responsive to hydrogen peroxide. Hence, hydrogen peroxide generated by the reaction of ascorbate is not expected to alter photosensitizer chemically. In this case, change in $^1O_2$ generation efficiency would only be attributed to $^1O_2$ quenching ability of the ascorbate. Figure S2 shows that presence of ascorbate barely changes the PDT efficiency of **BOD2**, indicating that major role of this compound on PDT action is via feedback loop (chemical modification of **BOD1** through reactions series).

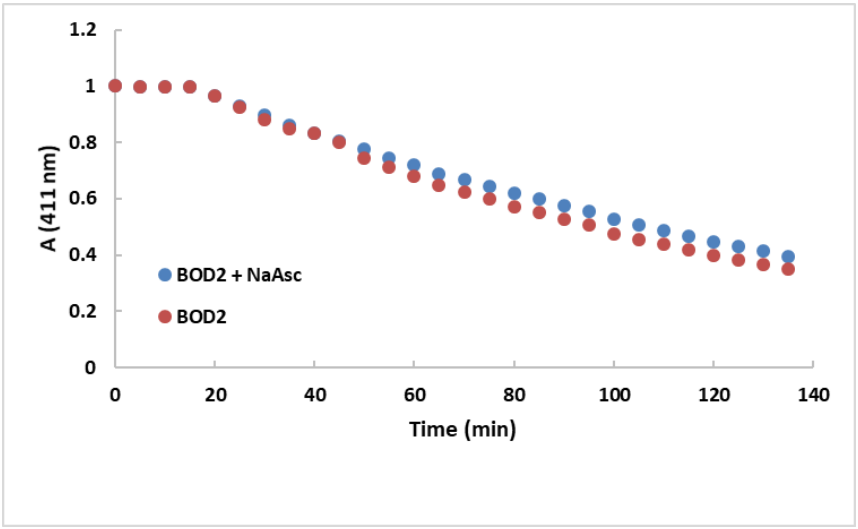


**Figure S2.** Singlet oxygen generation efficiencies of **BOD2** in the presence and absence of sodium ascorbate. Change in DPBF (50 μM) absorbance at 411 nm is followed in the presence of 5 μM of **BOD2** only (red) and 10 μM of **BOD2** in the presence of 0.2 mM of sodium ascorbate (blue). Small difference in the singlet oxygen generation ability is attributed to scavenging the $^1O_2$ by ascorbate.

**Table S1.** Photophysical properties of **BOD1**, **BOD2** and **BOD1** (10 μM) irradiated with 506 nm light for 150 min in the presence of 0.2 mM sodium ascorbate. All measurements were obtained in isopropanol.

| Compound | $\lambda_{max, abs}$ (nm) | $\lambda_{max, fluo}$ (nm) | $\phi_F$ |
|---|---|---|---|
| **BOD1** | 510 | 515 | 0.04 |
| **BOD1** + NaAsc + Light | 505 | 515 | 0.16 |
| **BOD2** | 504 | 515 | 0.31 |

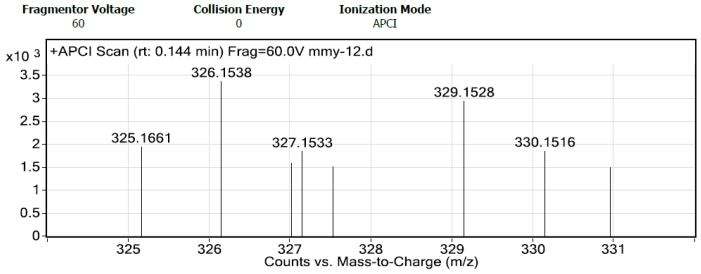


**Figure S3.** High Resolution ESI-MS spectrum of **BOD1** after irradiation with 506 nm LED light source for 150 min. Peaks corresponding to **BOD2** appears suggesting the formation of it from **BOD1** during PDT action. $(M+H)^+$ theoritical: 326.1640 ; experimental:326.1538; Δ: 31 ppm

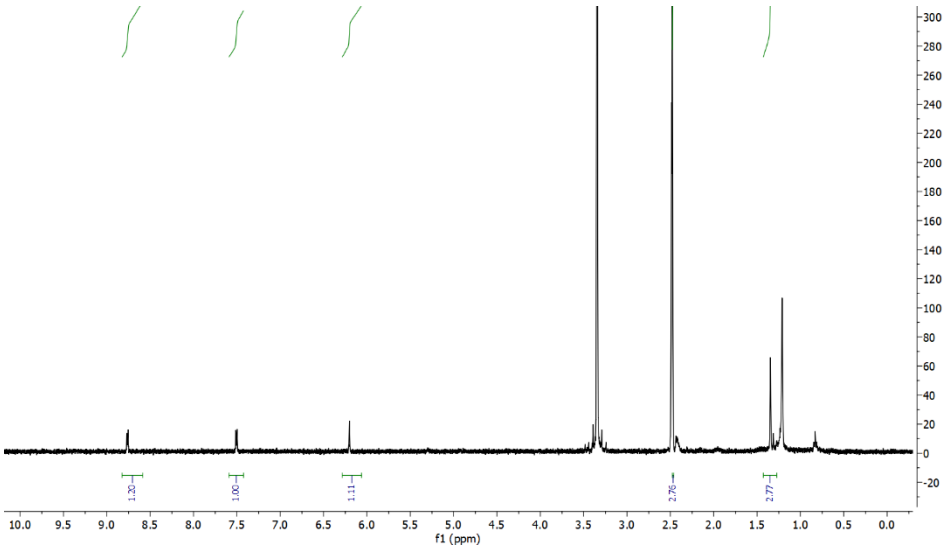


**Figure S4.** $^1$H NMR Spectrum (400 MHz, *d*-DMSO) of the samples in the organic phase of the extraction. After $^1$H NMR analysis during PDT action is performed (results of which is given in Figure 5), sample is extracted with chloroform and water. $^1$H spectrum of organic phase of the sample has peaks only corresponding to **BOD2**.

Stability of **BOD1** in aqueous solutions (PBS buffer) in biologically relevant pH values are analysed and it has been shown that within experimental time scale (less than 24h), there is no significant change in fluorescence (Figure S5). At day 3, fluorescence increases to a certain value at neutral pH, indicating that around that time **BOD2** may be generated spontaneously.

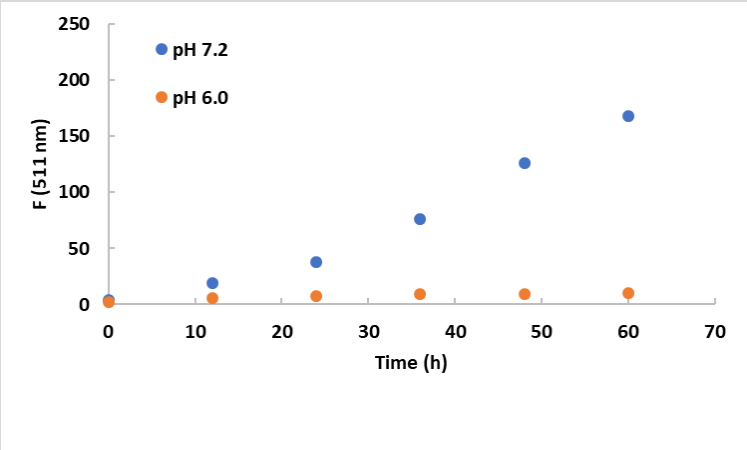


**Figure S5**. Stability of **BOD1** in PBS buffer at pH 7.2 (blue) and pH 6.0 (red) followed by change in fluorescence at 511 nm. Fluorescence change is not significant until 24h and all experiments, including cell culture analysis, are done within this time.

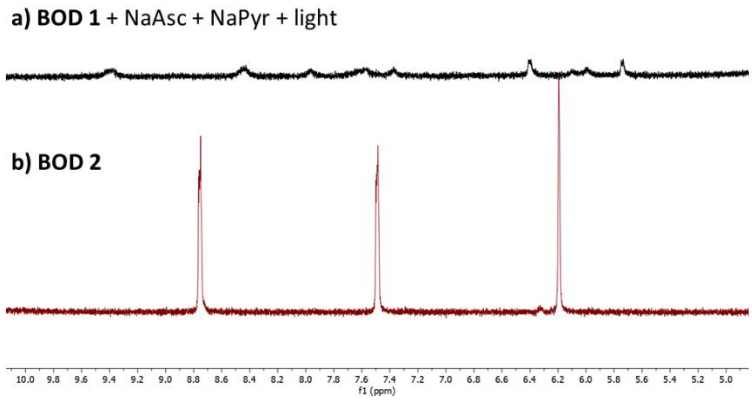


**Figure S6.** $^{1}$H NMR Spectra (400 MHz, *d*-DMSO) of **BOD 1** (25 mM) in the presence of saturated sodium ascorbate and sodium pyruvate (a) and **BOD 2** (b). NMR spectrum of BOD 1 was recorded after irradiation with 506 nm LED light for 80 min.

Peaks corresponding to **BOD 2** does not appear in spectrum shown in Figure S2a where as in the absence of hydrogen peroxide scavenger (pyruvate), **BOD 2** peaks are apparent (Figure 5). This result indicates scavenger interferes with the formation of **BOD 2**, in other words hydrogen peroxide mediates the loop reaction.

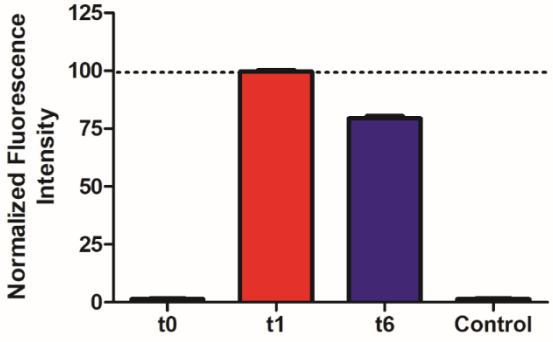


**Figure S7.** Change in the fluorescence intensity of DLD-1 cells incubated with **BOD 1** (10 μM). Cells are exposed to 506 nm light and fluorescence is recorded at times 0-6 hours. Control cells are kept in dark during 6h and fluorescence is recorded. Within the first hour, fluorescence of irradiated cells increases significantly indicating the formation of fluorescent but inactive **BOD 2** photosensitizer.

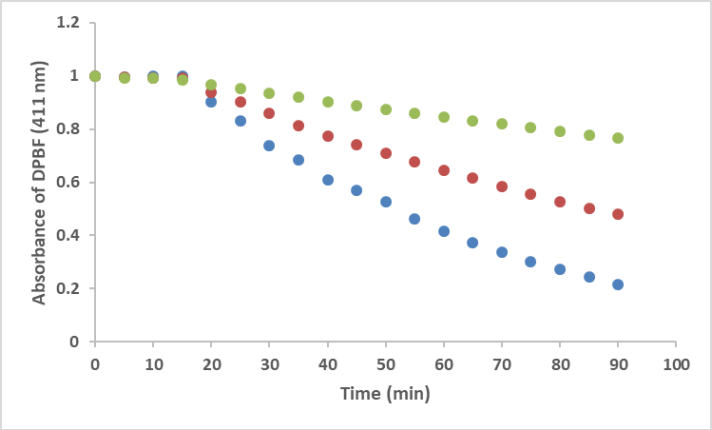


**Figure S8.** $^1O_2$ generation by **BOD1** (1, 5 and 10 μM shown in green, red and blue respectively, in isopropanol) followed by change in the absorption of DPBF (50 μM). Samples are kept in dark for 15 min and illuminated by 506 nm LED light for 75 min.

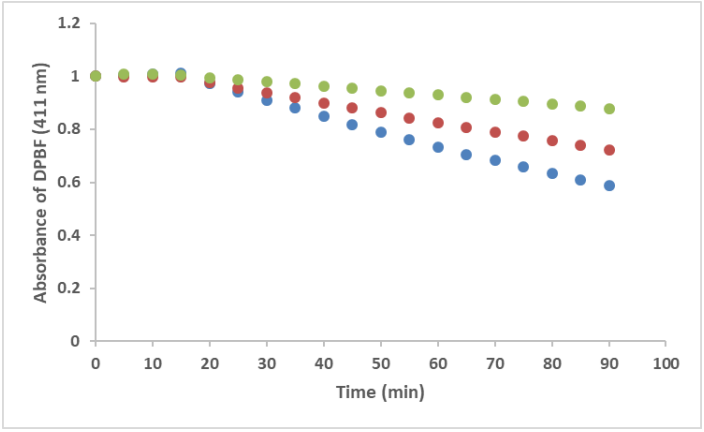


**Figure S9.** $^1O_2$ generation by **BOD2** (1, 5 and 10 μM shown in green, red and blue respectively, in isopropanol) followed by change in the absorption of DPBF (50 μM). Samples are kept in dark for 15 min and illuminated by 506 nm LED light for 75 min

## 3. Additional Characterization Data

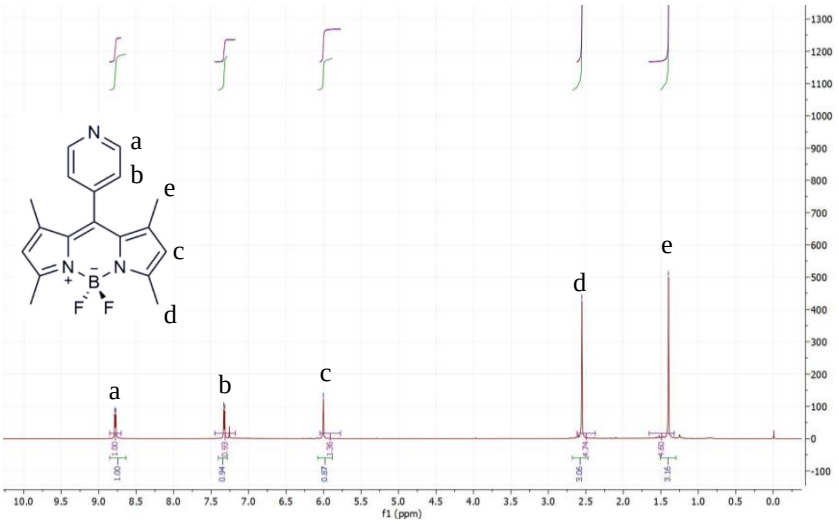


**Figure S10.** $^{1}$H NMR spectra of **BOD2** (400 MHz, Chloroform-*d*).[2]

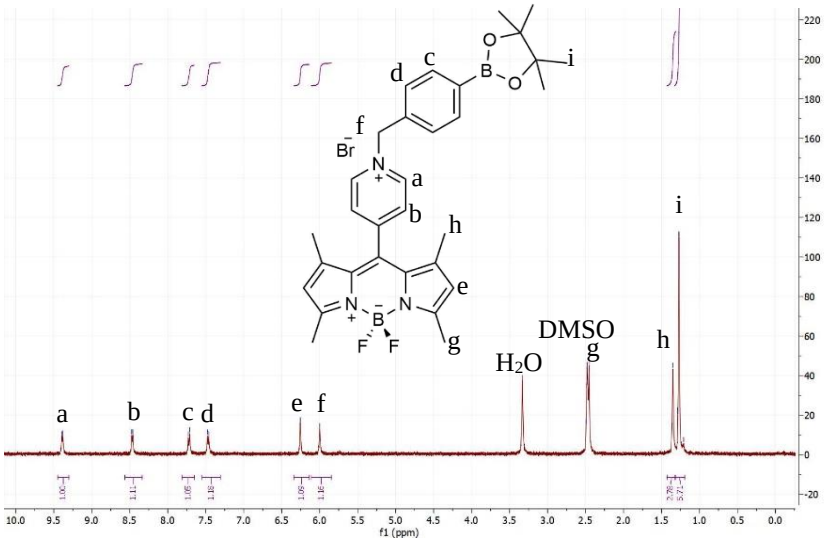


**Figure S11.** $^{1}$H NMR Spectra of **BOD1** (400 MHz, DMSO-$d_6$).[2]

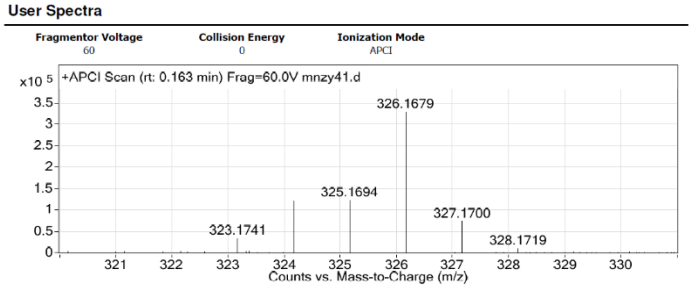


**Figure S12.** High Resolution ESI-MS Spectrum of **BOD2**.

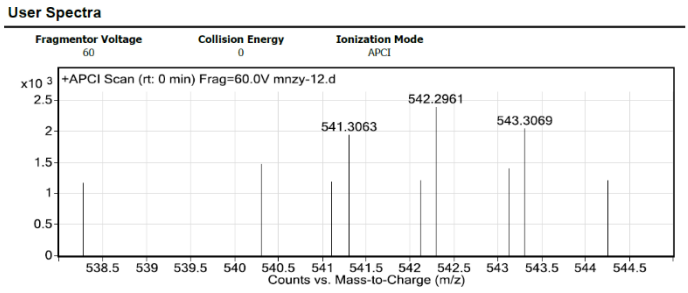


**Figure S13.** High Resolution ESI-MS Spectrum of **BOD1**.

## 4. References

(1) Xu, J.; Li, Q.; Yue, Y.; Guo, Y.; Shao, S. A Water-Soluble BODIPY derivative as Highly selective "Turn-On" fluorescent sensor for $H_2O_2$ sensig in vivo. *Biosense. Bioelectron*. **2014**, *56*, 58-63.

(2) Harriman, A.; Mallon, L J.; Ulrich, G.; Ziessell, R. Rapid Intersystem Crossing in Closely-Spaced but Orthogonal Molecular Dyads. *ChemPhysChem*, **2007**, *8*, 1207-1214.