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# $CF_3$-Bis-TEMPO-Vis: New Visible Light Active Bis-Benzimidazolequinone Alkoxyamine

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**Abstract:** Alkoxyamines of TEMPO usually dissociate thermally at >100 °C; however, room temperature homolysis, activated by visible light, occurs with benzimidazolequinone derivatives. 1,1′-Dimethyl-2,2′-bis{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-6-(trifluoromethyl)-1*H*,1′*H*-[5,5′-bibenzimidazole]-4,4′,7,7′-tetrone (**$CF_3$-Bis-TEMPO-Vis**) is prepared in a 59% yield through NBS/$H_2SO_4$ oxidative demethylations of the dimethoxybenzimidazole-benzimidazolequinone precursor with aqueous work up. The alternative basic work up in air gave the epoxide derivative of **$CF_3$-Bis-TEMPO-Vis**. Unlike the latter $CF_3$-epoxide, both alkoxyamine residues are labile under green light (470–600 nm), and the rate of TEMPO release is three times slower than **Bis-TEMPO-Vis**.

**Keywords:** epoxide; homolysis; kinetics; nitroxide; quinones; radicals



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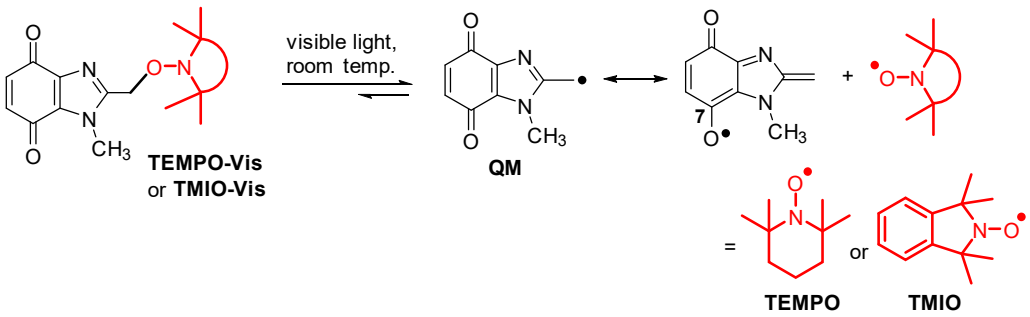
## 1. Introduction

Alkoxyamines usually dissociate thermally at >100 °C to bench-stable five or six membered cyclic α-tetraalkyl-substituted aminoxyl (nitroxide) and reactive carbon-centered radical [1,2]. Recently, we reported homolysis of benzimidazolequinone alkoxyamines of (2,2,6,6-tetramethylpiperidin-1-yl)oxyl (TEMPO) [3], and 1,1,3,3-tetramethylisoindolin-2-yloxyl (TMIO) [4], activated by visible light (Scheme 1). Room temperature homolysis is favored through extensive delocalization of the quinone methide (QM) radical, including onto the quinone 7-O atom. The facile radical reaction provides a useful and benign alternative to Nature's bioreductive activation of heterocyclic quinones (prodrugs) to give cytotoxic QMs [5–7].




**Scheme 1.** Visible light activation of benzimidazolequinone alkoxyamines [3,4].

Visible light activated the release of both TEMPO residues from **Bis-TEMPO-Vis** at the same rate (Figure 1) [3]. **TEMPO-Vis** dissociated faster than **Bis-TEMPO-Vis** in blue LED (420–520 nm); however, rates of dissociation were reversed in green LED (470–600 nm). Removal of conjugation from one of the quinone chromophores of **Bis-TEMPO-Vis** to

give epoxide derivative **1**, limited release to <1 equiv. of TEMPO over the same period [3]. Herein, we circumvent the epoxidation to give the new visible light active bis-alkoxyamine, 1,1′-dimethyl-2,2′-bis{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-6-(trifluoromethyl)-1*H*,1′*H*-[5,5′-bibenzimidazole]-4,4′,7,7′-tetrone (**$CF_3$-Bis-TEMPO-Vis**). The homolysis kinetics of **$CF_3$-Bis-TEMPO-Vis** under green light are disclosed and compared with bis-alkoxyamines, **Bis-TEMPO-Vis** and $CF_3$-epoxide **1**.

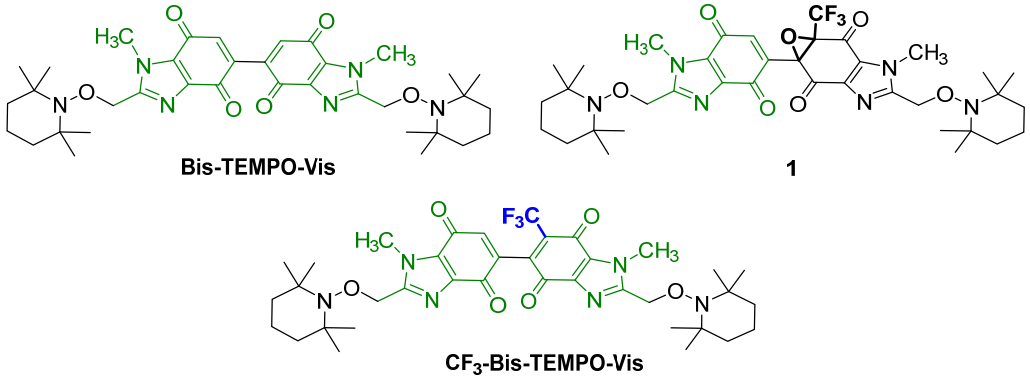
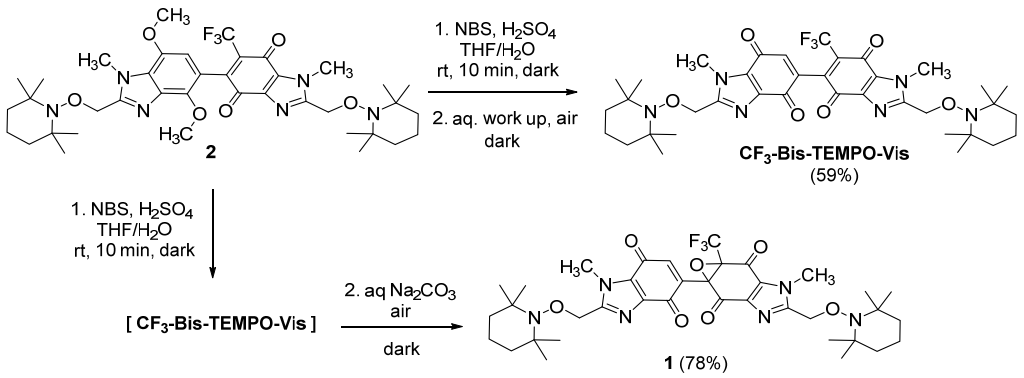



**Figure 1.** Bis-alkoxyamines [3] and new **$CF_3$-Bis-TEMPO-Vis**.

## 2. Results and Discussion

### *2.1. Synthesis*

NBS/$H_2SO_4$-mediated oxidative demethylations of the *p*-dimethoxybenzimidazole moiety of **2** to quinone, led to further functionalization to give the $CF_3$-epoxide **1** in 78% yield (Scheme 2), after aq. $Na_2CO_3$ workup in air [3]. The use of neutral conditions for workup, avoids epoxidation to give trifluoromethyl-bis-benzimidazolequinone, **$CF_3$-Bis-TEMPO-Vis** in a 59% yield. The key spectral difference between **1** and **$CF_3$-Bis-TEMPO-Vis** is the downfield shift of the *C*-$CF_3$ quartet in the $^{13}C$ NMR spectrum from 63.4 ppm to 132.3 ppm (Figure 2).




**Scheme 2.** Regioselectivity determined by the work up.

The epoxidation is likely to involve site-specific addition of hydroxide onto the highly electrophilic quinone *C*-$CF_3$ of **$CF_3$-Bis-TEMPO-Vis** followed by air oxidation (Scheme 3). This is supported by base-mediated Michael additions onto naphthoquinones [7], and Zhang et al. reported an epoxidation of a trifluoromethylated naphthoquinone using $H_2O_2/Na_2CO_3$ [8].

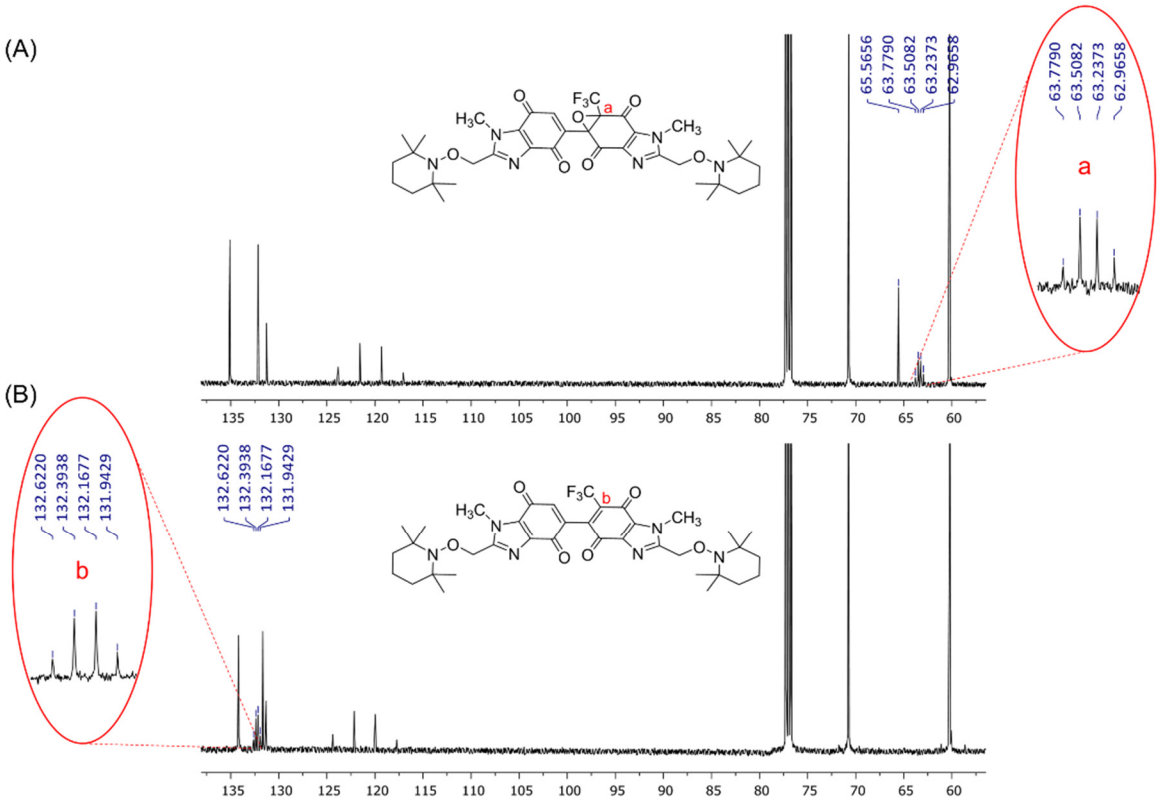
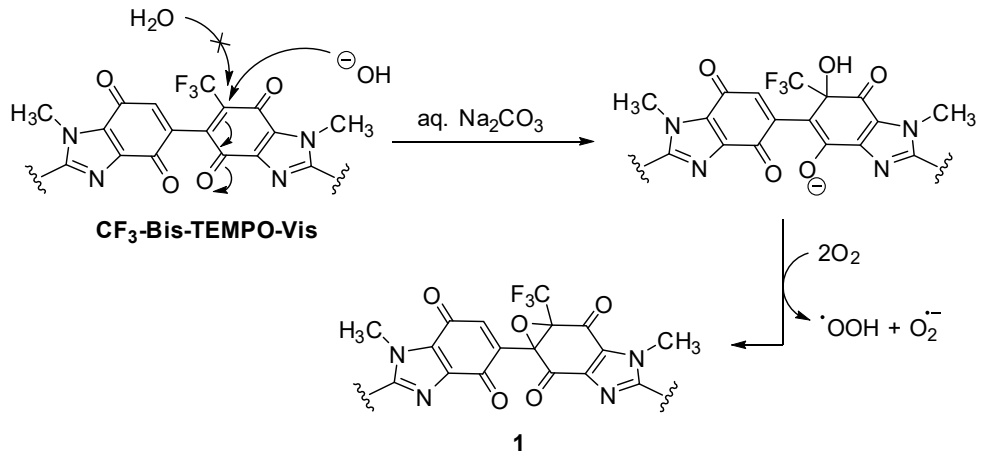



**Figure 2.** $^{13}$C NMR spectra of (**A**) $CF_3$-epoxide **1** and (**B**) **$CF_3$-Bis-TEMPO-Vis**.




**Scheme 3.** Proposed mechanism for epoxidation using base-mediated air-oxidation.

### *2.2. Kinetics of Homolysis*

The rate of homolysis of **$CF_3$-Bis-TEMPO-Vis**, **Bis-TEMPO-Vis**, and $CF_3$-epoxide **1** was determined at room temperature using green LED activation under an $O_2$ atmosphere to trap generated QM radicals [3]. HPLC monitored alkoxyamine decay and TEMPO release. Using initial bis-alkoxyamine concentrations of 5 mM in DCE, 130% and 152% TEMPO (based on the starting bis-alkoxyamine) were released from **$CF_3$-Bis-TEMPO-Vis** and **Bis-TEMPO-Vis**, respectively, after 348 min (Table 1). However, the decay of **$CF_3$-Bis-TEMPO-Vis** and **Bis-TEMPO-Vis** deviated from first order kinetics after about 20 min (Figure 3A). When the quinone moiety was removed, as in the case of $CF_3$-epoxide **1**, first order decay in green LED was observed, with the slope of the linear decay plot corresponding to a $k_d$ of 0.0162 min$^{-1}$ for 5 mM initial bis-alkoxyamine concentration.

Less than 1 equiv. of TEMPO (73% after 180 min) was released from bis-alkoxyamine **1** due the presence of one chromophore. The $k_d$ of **1** increases 2-fold when the homolysis solution was diluted to 0.25 mM, which is expected due to the relative increase in photon to molecule ratio using the same LED setup.

**Table 1.** Kinetics of Bis-Alkoxyamine Homolysis [a].

| Bis-Alkoxyamine | [Bis-Alkoxyamine]$_0$ (mM) | mol% TEMPO Released (time, min) [c,d] | $k_d$ (min$^{-1}$) [b] |
|---|---|---|---|
| **$CF_3$-Bis-TEMPO-Vis** | 5.00 | 130 (348) | - |
| **Bis-TEMPO-Vis** | 5.00 | 152 (348) | - |
| **1** | 5.00 | 73 (180) | 0.0162 ± 0.0010 |
| **$CF_3$-Bis-TEMPO-Vis** | 0.25 | 125 (80) | 0.0472 ± 0.0018 |
| **Bis-TEMPO-Vis** | 0.25 | 162 (15) [e] | 0.1480 ± 0.0020 |
| **1** | 0.25 | 43 (65) [e] | 0.0321 ± 0.0070 |

[a] Conditions: Bis-alkoxyamine in 1,2-dichloroethane (DCE) using green LED illumination (2 × 9 W bulbs) in the presence of $O_2$. [b] Homolysis rate ($k_d$) derived from the slope of the first-order decay plot (Figure 3). [c] HPLC yield based on starting bis-alkoxyamine. [d] TEMPO versus time plots, see Figures S1 and S2. [e] TEMPO versus time plot [3].

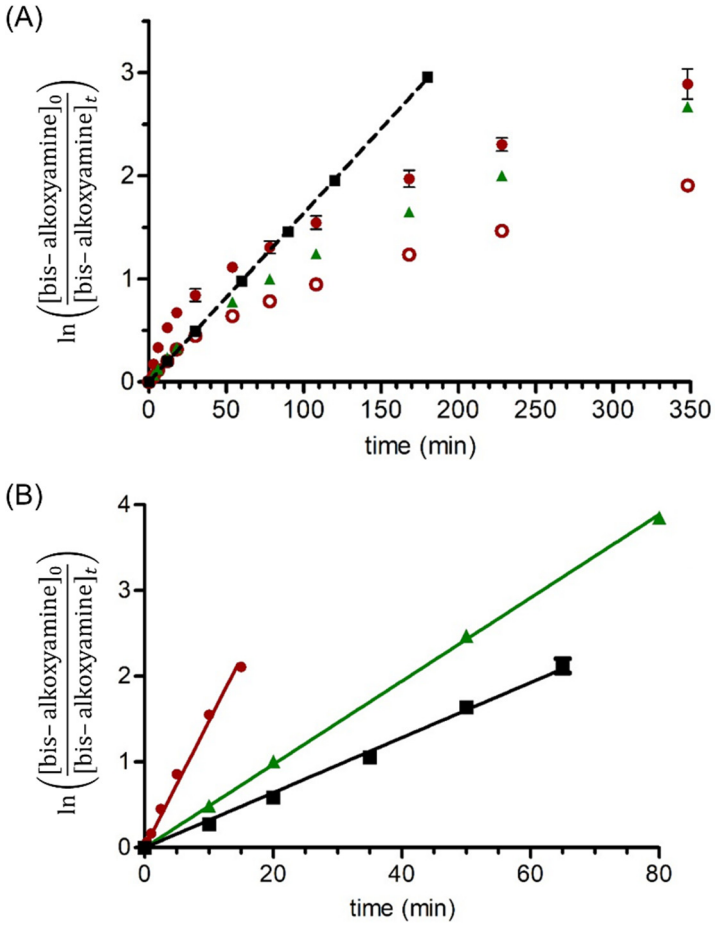



**Figure 3.** Kinetics for room temperature green-LED (2 × 9 W bulbs) induced decomposition in DCE at (**A**) [bis-alkoxyamine] = 5 mM and (**B**) [bis-alkoxyamine] = 0.25 mM, where **$CF_3$-Bis-TEMPO-Vis** (green triangles, green line), **Bis-TEMPO-Vis** (red filled circles, red line), **Bis-TEMPO-Vis** with added free TEMPO (1.3 equiv., red open circles), and $CF_3$-quinone epoxide **1** (black squares and black line).

The rate of visible light induced decay of alkoxyamines and bis-alkoxyamines of TEMPO is influenced by the concentration of released nitroxide (i.e., free TEMPO) [4]. The addition of 1.3 equiv. of free TEMPO to the **[Bis-TEMPO-Vis]**$_0$ = 5 mM experiment

slowed the rate of homolysis (Figure 3A). TEMPO significantly absorbs in the visible region (Figure 4) [4], and quenches excited states by electron transfer [9]. However, the addition of free TEMPO will also slow alkoxyamine decay through increased recombination. The diluting of the starting concentrations of the bis-alkoxyamines by 20-fold (to 0.25 mM), nullified the effects of light absorption and quenching by released TEMPO, with decay plots following first order kinetics (Figure 3B). The rate of decay of **$CF_3$-Bis-TEMPO-Vis** ($k_d$ = 0.0472 $min^{-1}$) was about three times slower than **Bis-TEMPO-Vis** ($k_d$ = 0.1480 $min^{-1}$), demonstrating a stabilizing effect by the $CF_3$ group. For thermal decomposition of alkoxyamines, strongly electron-withdrawing groups (EWGs) stabilize the alkyl radical fragment, promoting alkoxyamine bond homolysis [10]. Our observation (tentative, given only one example is presented) is a reversal of the observed thermal trend with visible light induced photolysis slower, when the highly EWG ($CF_3$) is present. Given that alkoxyamine fragments are remote (six or nine bonds away) from the $CF_3$ group, **$CF_3$-Bis-TEMPO-Vis** and **Bis-TEMPO-Vis** have similar DFT calculated bond dissociation energies (BDEs) [3,11]. The slower rate of homolysis compared to **Bis-TEMPO-Vis** may, thus, be due to the marginally lower absorbance of **$CF_3$-Bis-TEMPO-Vis** in the green region of the visible spectrum (Figure 4) or a $CF_3$ enhancement in the reductive quenching of the excited triplet state by released TEMPO.

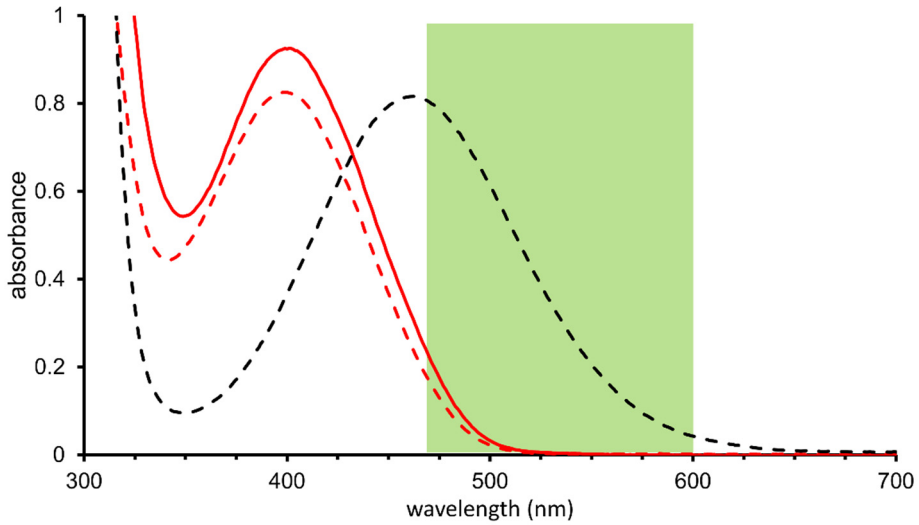



**Figure 4.** UV-Visible absorbance spectra in DCE of **Bis-TEMPO-Vis** (red continuous, 0.33 mM), and **$CF_3$-Bis-TEMPO-Vis** (red dashed, 0.33 mM) and TEMPO (black dashed, 75.0 mM) with the green LED emission region (470–600 nm) shaded. The higher concentration of TEMPO is to demonstrate free TEMPO absorption, and the inadequacy of TEMPO as a radical trap in kinetic studies [4].

## 3. Experimental

### *3.1. Materials*

The preparation of 4′,7′-dimethoxy-1,1′-dimethyl-2,2′-bis{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-6-(trifluoromethyl)-1*H*,1′*H*-[5,5′-bibenzimidazole]-4,7-dione (**2**), 3-methyl-6*a*-(1-methyl-4,7-dioxo-2-{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-4,7-dihydro-1*H*-benzimidazol-5-yl)-4-{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-1*a*-(trifluoromethyl)-1*H*-oxireno[f]benzimidazole-2,6-(3*H*,6a*H*)-dione (**1**), and 1,1′-dimethyl-2,2′-bis{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-1*H*,1′*H*-5,5′-bibenzimidazole-4,4′,7,7′-tetrone (**Bis-TEMPO-Vis**) is reported [3]. *N*-Bromosuccinimide (NBS, Lancaster, PA, USA, 99%), $H_2SO_4$ (BDH, 98%), THF (Sigma-Aldrich, St. Louis, MO, USA, ≥99%), $CH_2Cl_2$ (Fischer Scientific, Loughborough, UK, ≥99%), $MgSO_4$ (Alfa Aesar, Haverhill, MA, USA, 99.5%), EtOAc (Fischer Scientific, ≥99%), hexanes (Fischer Scientific, bp 40–60 °C), and 1,2-dichloroethane (DCE, Sigma-Aldrich, MI, USA, anhydrous, 99.8%) were used as received. Dry-column vacuum chromatography (with Apollo Scientific, Stockport, UK, silica gel ZEOprep 60 and

15–35 μm particle size) [12] was preferable for the purification of light-active compounds, due to the convenience of light exclusion by covering the apparatus with Al-foil during elution.

### *3.2. Measurements*

The melting point was measured on a Stuart Scientific melting point apparatus SMP3 (Staffordshire, UK). Ultraviolet-Visible (UV-Vis) spectra were recorded using a Varian (Cary 100) UV-Vis spectrometer (Palo Alto, CA, USA). Infrared spectra were recorded using a Perkin-Elmer Spec 1 (Perkin-Elmer, Waltham, MA, USA) with ATR attached. NMR spectra were recorded using a Varian 500 MHz instrument (Varian Medical Systems, Palo Alto, CA, USA). The chemical shifts were in ppm relative to $Si(CH_3)_4$. $^{13}C$ NMR data were collected at 125 MHz with complete proton decoupling. NMR assignments were supported by DEPT. $^{19}F$ NMR data were collected at 470 MHz. High-resolution mass spectrometry (HRMS) was carried out using ESI time-of-flight mass spectrometer (TOFMS, Waters, Milford, MA, USA) in positive mode using a Waters LCT Mass Spectrometry instrument at the School of Chemistry, NUI Galway, Ireland.

### *3.3. The Photoreactor*

The reactor used green (2 × 9 W) LED bulb(s) placed inside an aluminum container with a diameter of 10 cm (Figure S3). A vial (with capacity of 1.5 mL), containing a solution of alkoxyamine under a balloon of $O_2$, was placed in the center of the reactor opposite the bulb for specific time periods. The reactor was air cooled using a 2.5-W fan, which ensured that the interior remained at ambient temperature. The emission spectra of the LED was narrow and confined to the visible region (Figure 4).

### *3.4. Analytical HPLC*

The Agilent 1100 Series HPLC (Santa Clara, CA, USA) was equipped with a UV detector operating at 254 nm and a Phenomenex® BondClone™ 10 μm C18, 250 × 4.6 mm column. The mobile phase programs for **Bis-TEMPO-Vis** and $CF_3$-epoxide **1** were as reported [3], with analysis of **$CF_3$-Bis-TEMPO-Vis** homolysis in $O_2$ performed using the same conditions as for **Bis-TEMPO-Vis** homolysis in $O_2$.

### *3.5. Homolysis Rates ($k_d$)*

A solution of bis-alkoxyamine in DCE (1 mL, 5.00 mM or 0.25 mM) in a 1.5-mL clear glass vial was prepared in the absence of light. A sample was taken prior to insertion into the photoreactor, and analyzed by HPLC (representing time 0). A sample was taken and [TEMPO] and [alkoxyamine] were measured by HPLC during photolysis. During the run, the remainder of the solution was placed in the dark, which stopped the reaction, as evidenced by the on/off experiment [3]. Illumination of the solution was resumed, with continued sampling over time. Experiments were performed in triplicate. Rate constants ($k_d$) were derived using the first-order plot describing the decay of [alkoxyamine] over time fitting (Equation (1)).

$$\ln\left(\frac{[bis-alkoxyamine]_0}{[bis-alkoxyamine]_t}\right) = k_d \quad (1)$$

### *3.6. Synthesis of 1,1′-Dimethyl-2,2′-bis{[(2,2,6,6-tetramethylpiperidin-1-yl)oxy]methyl}-6-(trifluoromethyl)-1H,1′H-[5,5′-bibenzimidazole]-4,4′,7,7′-tetrone (**$CF_3$-Bis-Tempo-Vis**)*

*N*-Bromosuccinimide (NBS, 39 mg, 0.22 mmol) was added to dimethoxybenzimidazole-benzimidazolequinone **2** (0.152 g, 0.20 mmol), $H_2SO_4$ (18 μL, 0.34 mmol), and THF/$H_2O$ (6 mL, 2/1) at rt, and stirred for 10 min in the absence of light. $H_2O$ (10 mL) was added and extracted with $CH_2Cl_2$ (2 × 20 mL). The combined organic layers were dried ($MgSO_4$) and evaporated, and the residue purified by dry column vacuum chromatography with EtOAc and hexanes as eluent to give **$CF_3$-Bis-TEMPO-Vis** (86 mg, 59%) as a yellow solid;

mp 136–140 °C (deg); $R_f$ 0.31 (3:1 EtOAc:hexanes); $\lambda_{max}$ (DCE, nm) 399 ($\varepsilon = 2.21 \times 10^3$), 252 ($\varepsilon = 1.89 \times 10^4$); $\nu_{max}$ (neat, $cm^{-1}$) 2974, 2932, 1665, 1518, 1479, 1361, 1334, 1276, 1252, 1207, 1181, 1151, 1122, 1100, 1032; $\delta_H$ (500 MHz, $CDCl_3$) 1.10 (12H, s, $CH_3$), 1.23–1.25 (12H, m, $CH_3$), 1.34–1.38 (2H, m), 1.44–1.57 (10H, bs), 4.12 (3H, s, $NCH_3$), 4.13 (3H, s, $NCH_3$), 5.03 (2H, s), 5.04 (2H, s), 6.45 (1H, s); $\delta_C$ (125 MHz, $CDCl_3$) 16.9 ($CH_2$), 20.1 ($CH_3$), 32.9 ($CH_3$), 33.2 ($CH_3$), 39.7 ($CH_2$), 60.2 (C), 70.7 ($CH_2$), 121.1 (q, $^1J_{C\text{-}F}$ = 276.4 Hz, $CF_3$), 131.3, 131.7 (both C), 132.3 (q, $^2J_{C\text{-}F}$ = 28.3 Hz, C6), 134.2 (CH), 140.0, 140.1, 140.3, 140.8, 151.7, 153.0 (all C), 172.8, 176.6, 176.8, 177.5 (all C=O); $\delta_F$ (470 MHz, $CDCl_3$) −56.83; HRMS (ESI) *m/z* $[M + H]^+$, $C_{37}H_{48}N_6O_6F_3$ calcd. 729.3587, observed 729.3581.

## 4. Conclusions

The synthesis and homolysis kinetics of **$CF_3$-Bis-TEMPO-Vis** are established. This is a new visible light activated bis-alkoxyamine, which releases two TEMPO free radicals under green LED. The room temperature TEMPO release is remarkable given literature alkoxyamines dissociate at >100 °C; however, alkoxyamine decay is only first order under dilute conditions.

**Supplementary Materials:** The following are available online at: $^1H$, $^{13}C$, and $^{19}F$ NMR spectra for **$CF_3$-Bis-TEMPO-Vis**. Figure S1: TEMPO release at $[\text{Bis-alkoxyamine}]_0$ = 5.00 mM, Figure S2: TEMPO release at $[\text{Bis-alkoxyamine}]_0$ = 0.25 mM, and Figure S3: a photograph of the photoreactor.

**Author Contributions:** P.K. was the only experimentalist, who obtained and analyzed all data; P.K. wrote the manuscript with research director and supervisor, F.A.; D.A.S. obtained the research funding with F.A. and acted as joint project supervisor; P.F. became the official supervisor, when F.A. departed NUI Galway for Kingston University. All authors have read and agreed to the published version of the manuscript.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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