**Bis-Rhodamines Bridged with a Diazoketone Linker: Synthesis, Structure, and Photolysis**

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**ABSTRACT:** Two fluorophores bound with a short photoreactive bridge are fascinating structures and remained unexplored. To investigate the synthesis and photolysis of such dyes, we linked two rhodamine dyes via a diazoketone bridge ($-\text{COCN}_2-$) attached to position 5′ or 6′ of the pendant phenyl rings. For that, the mixture of 5′- or 6′-bromo derivatives of the parent dye was prepared, transformed into 1,2-diarylacetylenes, hydrated to 1,2-diarylethanones, and converted to diazoketones Ar$_1\text{COCN}_2\text{Ar}_2$. The high performance liquid chromatography (HPLC) separation gave four individual regioisomers of Ar$_1\text{COCN}_2\text{Ar}_2$. Photolysis of the model compound $\text{C}_6\text{H}_5\text{COCN}_2\text{C}_6\text{H}_5$ in aqueous acetonitrile at pH 7.3 and under irradiation with 365 nm light provided diphenylacetic acid amide (Wolff rearrangement). However, under the same conditions, Ar$_1\text{COCN}_2\text{Ar}_2$ gave mainly $\alpha$-diketones Ar$_1\text{COCO}\text{Ar}_2$. The migration ability of the very bulky dye residues was low, and the Wolff rearrangement did not occur. We observed only moderate fluorescence increase, which may be explained by the insufficient quenching ability of diazoketone bridge ($-\text{COCN}_2-$) and its transformation into another (weaker) quencher, 1,2-diarylethane-1,2-dione.

**INTRODUCTION**

The possibility to modify two fluorophores (and change the emission parameters of two dye residues) in the course of one photochemical reaction is intriguing and remained unexplored. If we consider two masked (caged) fluorophores bound with a linker (Scheme 1), the assembly may include two photoconvertible caging groups, one for each fluorophore (Scheme 1A). In this case, the photoactivation is stepwise, and the whole structure represents only a bare aggregate of two caged dyes. Alternatively, if a single photoreactive group efficiently suppresses the emission of the whole compound, and this group can be transformed into a nonquenching state, then both fluorophores may be activated in one step (Scheme 1B). This option is particularly challenging, as the quenching efficiencies of energy or electron transfer strongly depend on the distance. Therefore, we have chosen a potential fluorescence quencher and used it as a linker directly connecting two (identical) fluorophores.

The literature survey revealed that the fluorescein derivatives incorporating benzil fragments (Ar$_1\text{COCO}\text{Ar}_2$) are essentially nonfluorescent (due to photoinduced electron transfer). Therefore, we applied photoconvertible 2-diazo-1,2-diarylethanones Ar$_1\text{COCN}_2\text{Ar}_2$ closely related to Ar$_1\text{COCO}\text{Ar}_2$, prepared bis-fluorophores bridged with a diazoketone linker, and studied their photolysis. Our motivation was to clarify whether the short diazoketone bridge (COCN$_2$) incorporated between two dyes will suppress their emission, and whether a Wolff rearrangement will take place. As fluorophores, we have used $N,N'$-bis(2,2,2-trifluorethyl)-substituted rhodamines, which have absorption and emission spectra very similar to those of fluorescein. The structures of newly prepared compounds are given in Figure 1.

**Scheme 1. Combination of Two Caged Fluorophores Bound with a Linker (A) and an Alternative Based on a Single Photoreactive Caging Group Incorporated into a Linker (B)**

- **A**. Combination of two caged fluorophores with a linker under photochemical activation.
- **B**. Combination of one caged fluorophore with a linker under photochemical activation.
RESULTS AND DISCUSSION

**Synthesis.** The synthesis of bromorhodamines 6a,b from aminophenol 5 is given in Scheme 2. In the condensation reaction leading to compounds 6a,b, we compared two sets of conditions (see legend to Scheme 2). Higher yields (43–47%) were achieved when the first step was carried out without a solvent. Due to high temperature (160 °C) and the presence of water in the gas phase, the partial cleavage of the 2,2,2-trifluoroethyl amino group and the formation of the rhodol byproduct—a dye with the hydroxyl group instead of one CF₃CH₂NH residue—were observed. Under drastic condensation conditions, the undesired reaction was inevitable; it decreased the yields of the target compounds and complicated the isolation of pure dyes 6a,b. For isolation of compounds 6a,b, we applied chromatography on reversed-phase (C₁₈ silica gel) because crystallization or chromatography on regular silica was not successful. The mixture of bromides 6a and 6b was stable by storing at −18 °C but slowly decomposed at room temperature. A high degree of purity (>95% HPLC area) was required for the success of the next coupling step (Scheme 3). Only by applying highly pure bromides 6a,b, we were able to obtain acetylenes 7a–c in synthetically useful amounts.

At the next step (Scheme 3), bromides 6a,b were coupled with bis(tributylstannyl)acetylene and, as expected, provided a mixture of 3 compounds (7a–c). Isolation was performed by chromatography on reversed-phase and afforded a mixture of 5,5-, 5,6-, and 6,6-regioisomers in an overall yield of 81%.

The acetylene-bridged systems consisting of two fluorescent dyes linked directly through the triple bond belong to the family of through-bond energy transfer cassettes (TBET-Cs). The reaction conditions in Scheme 3 (for details, see the Experimental Section) may be applied for the synthesis of other TBET-Cs.

The reported conditions of hydration reaction (Scheme 4) were first checked with diphenylacetylene (tolane 9) in Scheme 5 as a model. Transformation of tolane to deoxybenzoin 10 catalyzed by NaF/(F₃SO₃)₃ or CF₃SO₃H in CF₃CH₂OH proceeded smoothly and with good yields. However, under all of these conditions, hydration
The Regitz diazotransfer with tosyl azide affords the target diazoketones 1a−d (Figure 1) were prepared according to the modified and optimized procedure of M. Regitz using p-toluen sulfonyl azide and DBU as a base of regioisomeric ketones (Scheme 4). The combinatorial fashion of the reaction sequence 6a,b−7a−c−8a−d increased the number of regioisomers on each step. The hydration reaction proceeded through the corresponding vinyl esters formed from acetylenes and TfOH. Further optimization was required, to fully hydrolyze these esters to ketones 8a−d. The HPLC analysis was difficult, due to numerous peaks with similar retention times. However, we managed to isolate a mixture of 8a−d and then separate it to individual components 1a−d so that the overall yield was about 80%. For that, we used preparative HPLC on reversed phase with a gradient of acetonitrile in the basic aqueous buffer.

**Scheme 2. Synthesis of Regioisomeric Bromorhodamines 6a and 6b Containing N,N′-Bis(2,2,2-trifluoroethyl) Groups**

Conditions: (i) pyridine, CH₂Cl₂, rt, overnight; (iii) 48% aq. HBr, AcOH, reflux, 6 h; (iv) method A: 160 °C, 3 h; addition of 5 (2nd equiv), 85% aq. H₃PO₄, 160 °C, 3 h (47%); method B: 1,2-dichlorobenzene, 160 °C, 3 h, addition of 5 (2nd equiv), 160 °C, 3 h (31%).
Structure Elucidation of Diazoketones 1a–d. The regularities of $^1$H NMR spectra reported for 5- and 6-substituted (in the pendant phenyl ring) rhodamines allowed us to assign structures to compounds 1a–d (Figure 1). Additionally, we used gCOSY and gHMBCAD spectra showing $^1$H–$^1$H and multibond (optimized for three bonds) $^1$H–$^13$C correlations, respectively. In the proton spectra, we observed six 1-proton multiplets corresponding to two 3-substituted benzene rings: one with CO and one with CN$_2$. For isomer 1 (lowest retention time in HPLC), these structures were sensitive to acids and decomposed under acidic conditions. They were isolated in milligram amounts and purified by means of preparative HPLC with acetonitrile and basic aqueous buffers (e.g., AcON$_4$ at pH 8.6). The overall preparative yield of all compounds 1a [5(N$_2$),5(CO)], 1b [6(N$_2$),5(CO)], 1c [5(N$_2$),6(CO)], and 1d [6(N$_2$),6(CO)] was about 40%. To avoid decomposition, the products were stored at $-18$ °C in the dark.

The photolysis of azibenzil 22,23 was performed under irradiation with 365 nm light in acetonitrile–water mixtures (80/20) in the presence of HEPES buffer (pH 6.5) or HCOON$_4$ buffer (pH 7.3–7.4). The reaction mixtures were analyzed by means of HPLC with a UV–vis absorption (diode array) spectrometer and a mass spectrometric detection (LC-MS). The expected product of the photolysis (in the absence of amines in the reaction solution)—diphenylacetic acid (13)—was detected along with deoxybenzoin (10), benzil (14), and traces of diphenylmethane (15) (Scheme 5). These compounds were identified by comparison with commercial reference substances (retention times, UV, and mass spectra).

In some experiments, we also detected products with higher masses: an oxazole formed upon [2 + 3] cycloaddition from acetonitrile and ketene 12b; as well as small amounts of 3,3,6,6-tetraphenyl-1,2,4,5-tetroxane, the peroxide related to the photocyclization product of diphenylacetic acid.

Photolysis of the solutions containing aqueous HEPES buffer provided complex mixtures with diphenylacetic acid (13) as one of the main products (Figure S1). Irradiation in the presence of aqueous HCOON$_4$ was found to be “cleaner” (Figure 2) and resulted in the formation of diphenylacetic acid.
amide (16; Scheme 5). Azibenzil 11 and amide 16 had the same retention times under conditions of HPLC separation. Unlike azibenzil (11) and benzil (14), amide 16 did not display the absorption maximum at about 320 nm. The composition of amide 16 was confirmed by HRMS data obtained for the reaction mixture (see Figure S2). The origin of amide 16 is obvious: it formed from ketene 12b and ammonia, as the strongest nucleophile present in the

### Table 1. Chemical Shifts ($\delta$, ppm) and Coupling Constants ($J$, Hz) of Aromatic Protons H-4—H-7 and H-4′—H-7′ in Compound 8c (Scheme 4) and Diazoketones 1a—d (Figure 1) in [D$_4$]MeOH

| Compound       | H-4       | H-5       | H-6       | H-7       | H-4′      | H-5′      | H-6′      | H-7′      |
|----------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| 8c, [5(CH$_2$),6(CO)] | 7.76 d (0.7) | 7.32 d (0.15) | 7.52 d (0.3) | 7.00 d (0.3) | 8.11 d (0.7) | 7.99 d (0.7) | 7.69 d (0.7) | 7.44 d (0.7) |
| Isomer 4       | Isomer 3   | Isomer 1  | Isomer 2  |           |           |           |           |           |
| 1a, [5(N$_2$),6(CO)] | 8.24 d (1.4) | 7.93 d (0.3) | 7.29 d (0.3) | 8.24 d (1.4) | 7.93 d (0.3) | 7.29 d (0.3) | 8.24 d (1.4) | 7.93 d (0.3) |
| Isomer 4       | Isomer 3   | Isomer 1  | Isomer 2  |           |           |           |           |           |
| 1b, [5(N$_2$),5(CO)] | 7.81 d (2.6) | 7.35 s    | 7.35 s    | 7.81 d (2.6) | 7.35 s    | 7.35 s    | 7.81 d (2.6) | 7.35 s    |
| Isomer 4       | Isomer 3   | Isomer 1  | Isomer 2  |           |           |           |           |           |
| 1c, [5(N$_2$),6(CO)] | 7.90 d (0.7) | 7.80 d (0.7) | 7.35 s    | 7.90 d (0.7) | 7.80 d (0.7) | 7.35 s    | 7.90 d (0.7) | 7.80 d (0.7) |
| Isomer 4       | Isomer 3   | Isomer 1  | Isomer 2  |           |           |           |           |           |
| 1d, [6(N$_2$),6(CO)] | 8.06 d (3.3) | 8.06 d (3.3) | 8.06 d (3.3) | 8.06 d (3.3) | 8.06 d (3.3) | 8.06 d (3.3) | 8.06 d (3.3) | 8.06 d (3.3) |
| Isomer 4       | Isomer 3   | Isomer 1  | Isomer 2  |           |           |           |           |           |

**Figure 2.** Irradiation of azibenzil (PhCOCN$_2$Ph) dissolved in aqueous acetonitrile (80% acetonitrile, 20% water, v/v) with HCOONH$_4$ buffer (pH 7.3) results in full conversion to a new substance (amide 16, see Figure S2) with the same retention time but without absorption maximum at 319 nm. (A) Absorption changes upon irradiation; inset: transient at 319 nm. (B) Chromatograms (2D maps) of the sample before (left) and after (right) irradiation. (C) Chromatogram at 260 nm (a shift was introduced for clarity); inset: absorption spectra of the main peaks.
equilibrium in aqueous ammonium formate (2 mM) at pH 7.3–7.4 (the initial concentration of azibenzil was 0.1 mM). At physiological pH, ammonia may be considered as an analogue of biogenic amines, which have basicity similar to ammonia.

Having in mind the encouraging results obtained with model diazoketone 11, we performed the photolysis of diazoketones 1a–d (12 μM) in aqueous acetonitrile (acetonitrile/water = 80/20; v/v) in the presence of ammonium formate buffer (pH 7.3–7.4) (Scheme 6). Surprisingly, in this solvent, diketones 1a–d photolysis products were always 12 Da lower than the diazoketone residue turned out to be an inefficient quencher by the presence of the diazoketone bridge. The product of the starting diazoketones 1a–d, which diﬀer from each other considerably (Figure S4b). As expected, isomers 1 and 3 (compounds 1b and 1c in Figure 1) gave the same diketone 5-ArCOCOAr-6. The products’ retention times (Figure S3a) and emission gains were very similar: 30 and 20%, respectively (Figure 3). For all diazoketones, the photoactivation ratios (1.2–2.4) are moderate, if compared with dyes having two 2-nitrobenzoylcarbonyl residues attached to the nitrogen atoms in one fluorophore, photoactivatable rhodamine spiroamides, or rhodamines incorporating the spiro-diazoketone fragment.

This result may be explained if we assume that the quenching ability of diazoketone COCN2 is higher than that of α-diketone COCO, but the former does not completely inhibit the emission, while the latter does not allow to unfold the full fluorescence signal pertinent to two fluorophores. In addition, the quenching ability of the COCN2 residue toward “left” (Ar′) and “right” (Ar″) aryl groups in Ar′COCN2Ar″ is expected to be diﬀerent, and may also depend on the substitution pattern of the aromatic ring (i.e., 5′ or 6′). The Wolff rearrangement is unfavored because the migration ability of the very bulky dye residue is low.

**CONCLUSIONS**

We prepared and studied the photolysis of assemblies consisting of the two identical fluorophores directly bound with a short, compact, and photoconvertible diazoketone bridge (−COCN−). Structurally, this approach to compounds in which two fluorophores can be activated with one photon is simpler than the design of sophisticated assemblies containing one photoconvertible unit (FRET acceptor) bound with two fluorescent dyes (FRET donors). In the course of photolysis, we observed only a moderate fluorescence increase. However, this method may be easily extended to compounds with other, more eﬃcient quenchers linking two fluorescent dyes and undergoing photoconversion into another, essentially nonquenching state.

**EXPERIMENTAL SECTION**

**General Remarks.** The reactions were performed with magnetic stirring under argon. Oil baths were used for heating the reaction mixtures, and the bath temperatures are given as reaction temperatures. Evaporations in vacuum were performed in a rotary evaporator with bath temperature not exceeding 40 °C. NMR spectra were recorded at 25 °C on an Agilent 400-MR (400 MHz 1H and 100.5 MHz 13C). All spectra are referenced to tetramethylsilane (δ = 0 ppm) using the signals of the residual protons of CHCl3 (7.26 ppm) in CDCl3, CH2OD (3.31 ppm) in CD2OD (49.15 ppm for 13C), CH2CN (1.94 ppm) in CD3CN (1.39 and 189.69 ppm for 13C), or [D6]DMSO (2.50 ppm for 1H); 39.5 ppm for 13C in [D6]DMSO.

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**Figure 3.** Relative fluorescence of isomers 1–4 (Figure 1) in MeCN (80% v/v) and 10 mM HCOONH4 buffer, pH = 7.4 (20% v/v). Yellow bars: starting materials. Green bars: after complete photolysis of the starting diazoketones. The numbers on top of the bars show the fluorescence quantum yields for the starting compounds and their increase upon photolysis to mixtures containing α-diketones as the main products (see Figure S3).
Multiplicities of signals are described as follows: s = singlet, br. = broad signal, d = doublet, t = triplet, q = quartet, m = multiplet, dd = doublet of doublets. Coupling constants (J) are given in hertz. Structural assignments for asymmetric acetylenes 7a–c, ketones 8a–d, and diazoketones 1a–d were made with additional information from gCOSY, gHSQC, and gHMBC experiments. Mass spectra with electrospray ionization (ESI-MS) were recorded on a Varian 500-MS spectrometer (Agilent). ESI-HRMS were measured on a MICRO-TOF spectrometer (Bruker) equipped with an Apollo ion source and a direct injector with an LC-autosampler Agilent RR 1200. Analytical HPLC separations (reversed phase) were performed on MERCK ready-to-use plates with silica gel 60 (F254). Preparative HPLC separations (reversed phase) were otherwise performed on LC-Shimadzu LCMS-2020. 5′-Bromo-3′-(bis(2,2,2-trifluoroethyl)rhodamine (6a) and 6′-Bromo-4′- (bis(2,2,2-trifluoroethyl)rhodamine (6b). In a pear-shaped flask, phosphoric pentafluoride (500 mg, 2.20 mmol, 1.0 equiv) and powdered aminophenol 5 (317 mg, 1.66 mmol, 0.75 equiv) were well mixed and heated under argon at 170 °C for 3 h. After cooling to rt, the red mixture was then taken up in ethyl acetate, washed with aqueous NaHCO3 (2 × 50 mL), sat. NaCl solution (50 mL), dried over MgSO4, and evaporated to get 602 mg (48%) of the title compound as a bright red solid. TLC (SiO2) hexane/AcOEt 1:2, Rf (mixture 6a, 6b) = 0.33. TLC (reversed phase): MeCN:H2O, 7:3; Rp (mixture 6a, 6b) = 0.19. Analytical HPLC (Knauer, A/B = 70/30–0/100 in 20 min, λ = 530 nm): tR = 11.3 min and 11.6 min (1:1; sum of peak areas 100%). As a byproduct, we isolated 172 mg (14%) of compounds with one oxygen atom instead of one CF2CH2NH group (dark orange solid). For additional purification, the product was dissolved in a minimal amount of aqueous MeCN and subjected to preparative HPLC (Interchrom; gradient MeCN/H2O: 20/80–70/30 with 0.1 v/v% of HCOOH in both components); detection interval 200–600 nm, column Knauer (see the General Remarks section). The pure fractions were pooled and evaporated; the residue dissolved in 1,4-dioxane and filtered through a 0.2 μm PTFE membrane filter. The filtrate solution was freeze-dried to yield 530 mg (86%) of compounds 6a,b as red solid. Mixture of 5′- and 6′-COOH isomers in ca. 1:1 ratio. 1H NMR (CD3CN, 400 MHz) δ 8.11 (d, 0.5H, J = 1.8, H-4′ in 5′-isomer), 7.89–7.75 (m, 1H, H-6′ in 5′-isomer and H-4′ in 6′-isomer), 7.41 (d, 0.5H, J = 1.5, H-7′ in 6′-isomer), 7.11 (d, 0.5H, J = 8.2, H-7′ in 5′-isomer), 6.64–6.55 (m, 4H), 6.46 (m, 2H), 5.27 (t, 2H, J = 7.0, NH), 3.88 (m, 4H, CH2CF3). 13C{1H} NMR (CD3CN, 101 MHz) δ 169.7, 169.1 (COOH), 156.1, 154.0, 153.0, 151.1, 133.8 (all Cq), 139.4 (CH3), 134.6 (CH), 130.9 (Cq), 130.6 (Cq), 130.3 (2 × CH), 128.8 (CH), 128.5 (CH), 128.3 (Cq), 127.6 (Cq), 127.2, 127.2 (q = 280, CF2), 124.5 (Cq), 111.6 (2 × CH), 109.3 (Cq), 99.9 (2 × CF3), 44.3 (2 × CH2CF3). [M + Na]0+ calcd for C50H32F12N4O6Na 861.1720 (99.9, 90.6, 89.9, 89.1, 88.4, 87.8), 87.5 (2 × 44.3 CH2CF3), 40.3 (2 × CH3). HRMS (ESI-TOF) m/z: [M + H]+ calc for C24H14BrF5N2O4 573.0243; found 573.0224; [M + Na]+ calc for C24H14BrF5N2O4Na 595.0622; found 595.0424.

Compounds 7a–c A 1:1 mixture of 5′- and 6′-bromorhodamines 6a,b (2.82 g, 4.9 mmol, 2 equiv) and Ph(Pd(PPh3)3) (284 mg, 0.25 mmol, 0.1 equiv) was transferred into a screw-cap 100 mL pressure tube and purged with argon for 5 min. Degassed dioxane (45 mL) and bis(trifluoromethyl)acetone (1.29 mL, 1.48 g, 24.6 mL, 1.0 equiv) were added, and the reaction mixture was purged with argon for 10 min. The reaction vial was closed, and the red reaction solution was heated to 110 °C with stirring for 5 h. The course of the reaction was monitored by TLC, LCMS, or HPLC. After the reaction was complete, the reaction mixture was cooled to rt, water (50 mL) was added, and the mixture was extracted with ethyl acetate (3 × 50 mL). The combined organic solutions were washed with brine and dried over MgSO4. The solvents were removed under reduced pressure. The residue (3.5 g) was dissolved in ethyl acetate, applied onto Celite, and subjected to flash chromatography in two portions. Cartridge with 40 g of regular SiO2 (Puriflash, 15 μm); eluent CH2Cl2:MeOH = 90:10 to 35:65 to 50:50 (v/v). The fractions containing compounds 7a–c were pooled and concentrated under reduced pressure, excluding light and atmospheric oxygen. The residue was dissolved in dioxane, filtered through a 0.2 μm PTFE filter, frozen, and lyophilized. Yield 2.01 g (81%) of the mixture 7a, 7b and 7c (red solid). TLC (SiO2), CH3Cl2/AcOEt 1:1; tR = 0.28, 0.20, 0.13. Analytical HPLC (Knauer, A/B = 80/20–30/70 in 30 min, λ = 530 nm): tR = 20.4 min (peak area 34%), 21.5 min (peak area 40%), 22.3 min (peak area 26%). Compound 7a [isomer 5,5′]. 1H NMR (CD3CN, 400 MHz) δ 8.16 (dd, 2H, J = 1.5, 0.8, H-4′, 4′), 7.90 (dd, 2H, J = 8.0, 1.5), 6.66 (dd, 2H, J = 8.0, 0.7), 6.63 (d, 4H, J = 8.7), 6.59 (d, 4H, J = 2.4), 6.48 (dd, 4H, J = 8.7, 2.4), 5.28 (t, 4H, J = 8.7, NH), 3.89 (qd, 8H, J = 9.3, 6.8, CH3CF2). 13C{1H} NMR (CD3CN, 101 MHz) δ 169.7 (COOH), 154.2, 154.0, 151.1, 139.4, 130.9, 129.1, 126.9 (q = 276, CF2), 125.9, 125.5, 111.6, 109.85, 99.9, 90.6, 45.8 (q = 33, CH2CF3). HRMS (ESI-TOF) m/z: [M + H]+ calc for C50H32F12N4O6Na 861.1720 (99.9, 90.6, 89.9, 89.1, 88.4, 87.8), 87.5 (2 × 44.3 CH2CF3), 40.3 (2 × CH3). [M + Na]+ calc for C50H32F12N4O6Na 883.1803; found 883.1667. Compound 7b [isomer 5,6′]. 1H NMR (CD3CN, 400 MHz) δ 8.17 (dd, 2H, J = 1.5, 0.8, H-4′, 4′), 8.09 (dd, 2H, J = 8.0, 0.7, H-7), 7.90 (dd, 1H, J = 8.0, 1.5, H-6), 7.86 (dd, 1H, J = 8.0, 1.5, H-5′), 7.46 (dd, 1H, J = 1.4, 0.7, H-7′), 7.25 (dd, 1H, J = 8.0, 0.7, H-7), 6.81 (d, 2H, J = 8.8), 6.75 (d, 2H, J = 8.8), 6.71 (d, 4H, J = 8.4, 2.4), 6.62 (dd, 2H, J = 8.8, 2.4), 6.59 (dd, 2H, J = 8.8, 2.4), 5.74 (br. s, 4H, NH), 3.96 (m, 8H, CH2CF3). 1H NMR (CD3OD, 400 MHz) δ 8.36 (m, 1H, H-4),
lyophilized to yield 163 mg (80%) of a dark red solid containing all.

Under stirring at room temperature, the following reagents were
dried over MgSO₄, and concentrated. The residue was dissolved in
fl.

were carefully transferred with aqueous dioxane into a 1 L Erlenmeyer
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(0.1 mL), and CF₃SO₃H (0.50 mL) were added; and heating was
additional portions of CF₃SO₃H (0.50 mL each time) were added
Compound 7c [isomer 6,6

δ

J

1H,

(dd, 4H,

J

7a

= 8.0, 1.4; H-5

m

97.3 (CH), 91.1 (Cq), 89.4 (Cq), 45.1 (m, CH₂CF₃). HRMS (ESI-

TOF) m/z: [M + H]⁺ calcd for C₅₀H₃₂F₁₂N₄O₇Na 1051.1972; found 1051.2039. m/z: [M + Na]⁺ calcd for C₅₀H₃₃F₁₂N₄O₇Na 1073.2092; found 1073.2146. Compound 8a [S(CH₃)₅-S(CH₅)₅]. ¹H NMR (CD₃CN, 400 MHz) δ 8.48 (dd, 1H, J = 1.6, 0.7; H-4 [CO]), 8.24 (dd, 1H, J = 1.8, 1.6; H-4 [CO]), 7.77 (m, 1H, H-7 [CH]), 7.28 (dd, 1H, J = 8.1, 0.7; H-5 [CH]), 7.11 (dd, 1H, J = 1.4, 0.7; H-7 [CH₃]), 6.61 (2H, J = 8.7), 6.58–6.57 (m, 3H), 6.52 (m, 1H), 6.45 (dd, J = 8.9, 6.7 and 2.4, 4H), 5.23 (2 x t, J = 7, 4H, NH), 4.56 (s, 2H, CH₂CO), 4.47 (s, 2H, CH₂CO). ¹³C[H] NMR (CD₃CN, 101 MHz) δ 197.1 (CO), 170.5, 169.6 (COOH), 158.3, 154.9, 153.9, 151.1, 150.9, 144.2, 139.6, (all C), 136.1 (C), 133.2 (CH), 130.3 (CH₂), 128.9 (C₈), 128.3 (C), 126.9 (q, J = 282, CF₃), 126.8 (CH₂), 126.1 (CH₂), 125.7 (CH₂), 125.6, 116.1, 116.1, 110.0, 109.1 (all C), 99.94 (CH₉), 99.90 (CH₇), 46.7 (CH₂). 48.5 (2 x q, J = 34, CH₂CF₃). HRMS (ESI-TOF) m/z: [M + H]⁺ calcd for C₅₀H₃₃F₁₂N₄O₇Na 1073.2092; found 1073.2146. m/z: [M + Na]⁺ calcd for C₅₀H₃₄F₁₂N₄O₇Na 1095.2172; found 1095.2172 m/z: [M + 2H]²⁺ calcd for C₅₀H₃₄F₁₂N₄O₇Na 1095.2172; found 1151.1100 Compound 8c [S(CH₅)₅-S(CH₅)₅]. ¹H NMR (CD₃CN, 400 MHz) δ 8.26 (dd, 1H, J = 8.0, 1.4; H-4 [CO]), 8.09 (d, 1H, J = 8.1; H-3 [CO]), 7.88 (d, 1H, J = 1.1; H-7 [CO]), 7.75 (d, 1H, J = 1.5; H-7 [CH₃]), 7.50 (dd, 1H, J = 8.0, 1.6; H-6 [CH₃]), 7.08 (d, 1H, J = 7.9; H-6 [CH₃]), 6.63–6.49 (m, 8H), 6.45 (m, 4H), 5.27 (m, 4H, NH), 4.48 (s, 2H, CH₂CO), 3.88 (m, 8H, CH₂CO). ¹³C[H] NMR (CD₃CN, 101 MHz) δ 198.0 (CO), 170.4, 169.7 (COOH), 154.7, 154.0, 153.9, 152.9, 151.1, 150.9, 143.8, 138.5 (all C), 138.1 (CH), 131.8 (C), 130.4 (CH₂), 128.4 (C), 128.3 (C), 127.0 (C₁₂), 126.5 (CH₂), 126.5 (q, J = 282, CF₃), 125.9 (CH₂), 124.6 (CH₂), 124.6 (CH), 111.5 (2 x CH₃), 99.90 (CH₇), 46.8 (CH₂). 48.5 (2 x q, J = 45, CH₂CF₃). ²⁹F NMR (CD₃CN, 376 MHz) δ −72.85 (t, J = 9.3), −72.84 (t, J = 9.3). HRMS (ESI-TOF) m/z: [M + H]⁺ calcd for C₅₀H₃₄F₁₂N₄O₇Na 1095.2172; found 1095.2172. m/z: [M + Na]⁺ calcd for C₅₀H₃₅F₁₂N₄O₇Na 1151.1100; found 1151.1100.

Compounds 8d [6(CH₃)₅-6(CO)]. ¹H NMR (CD₃CN, 400 MHz) δ 8.09 (dd, 1H, J = 8.0, 1.4; H-5 [CO]), 8.00 (dd, 1H, J = 8.0, 0.8; H-4 [CO]), 7.83 (dd, 1H, J = 7.9, 0.7; H-4 [CH₃]), 7.69 (m, 1H, H-7 [CH₃]), 7.41 (dd, 1H, J = 7.9, 1.4; H-5 [CH₃]), 6.91 (m, 1H, H-7 [CH₃]), 6.56 (m, 4H), 6.48 (dd, J = 8.7, 2.3, 4H), 6.41 (dt, J = 8.7, 2.6, 4H), 5.23 (m, 4H, CH₂CO), 3.88 (m, 8H, CH₂CO). ¹³C[H] NMR (CD₃CN, 101 MHz) δ 197.9 (CO), 170.4, 169.6 (COOH), 154.7, 154.6, 154.0, 153.8, 151.0, 150.8, 144.1, 143.7 (all C), 133.1 (CH), 131.7 (C), 130.8, 130.4 (CH₂), 130.3 (CH₂), 128.3 (C₁₂), 126.9 (C), 126.9 (q, J = 282, CF₃), 126.7 (CH₂), 126.4 (CH₂), 126.4 (CH), 125.7 (CH₂), 125.6, 125.3 (CH₁₁), 111.5 (2 x CH₃), 99.90 (CH₇), 46.2 (CH₂). 48.5 (2 x q, J = 45, CH₂CF₃).
**ASSOCIATED CONTENT**

**Supporting Information**

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.joc.1c01721.

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