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Photodecarboxylative Ring Annulation of α- and β-Functionalized Phthaloyl-GABA Derivatives: Bioactive Pyrroloisoindolinones with High Quantum Efficiency

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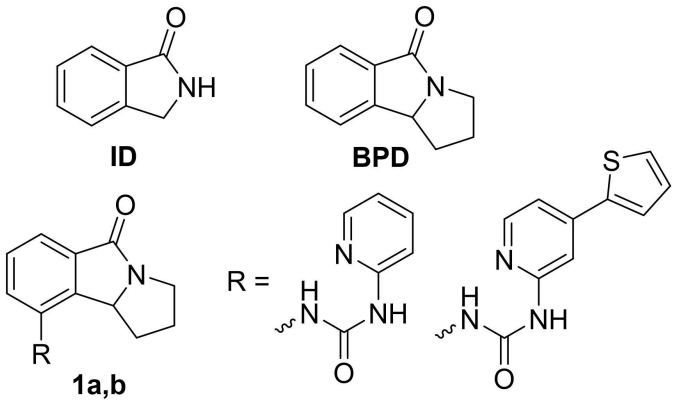
The triplet-sensitized (by the solvent acetone) as well as the direct (λ_{ex} = 300–320 nm) photochemical decarboxylation of N-phthaloylated γ-aminobutyric acid (GABA) derivatives are versatile and high-yielding routes to benzopyrrolizidines via intramolecular electron transfer initiated decarboxylation followed by radical coupling. The ß-mono- and ß,ß'-disubstituted N-phthaloyl GABA derivatives **7a**–**7g**, respectively, were applied as substrates. Decarboxylative photocyclization yielded hydroxy benzopyrrolizidines **8a**–**8g** in high chemical yields and with moderate diastereoselectivities from the ß-monosubstituted substrates. The analogous α-substituted GABA derivatives **11a**–**11c** were also applied as potential substrates for memory of chirality effects. The reaction quantum yields of the photodecarboxylation reactions for the parent GABA derivative **13** and for the new substrates **7h** and **11a** were determined by the quantum yield determination system (QYDS) and showed a remarkable concentration dependency indicating aggregation at higher substrate concentrations. Inhibition studies on the atherogenic human serine hydrolase cholesterol esterase showed derivatives **8a** and **8d** to exhibit a hyperbolic mode of inhibition with moderate IC_{50} values of about 60–80 μM.

The heterocyclic structures of isoindoline and isoindolinone (**ID**) are privileged structures in numerous natural alkaloids and synthetic derivatives.[1] Isoindolinones (phthalimidines) represent a growing class of benzofused γ-lactam natural compounds. Synthetic and natural isoindolinones are of high pharmacological relevance and valuable building blocks for materials synthesis.[2,3]

Another important class of nitrogen-containing heterocycles are pyrrolizidine alkaloids, plant metabolites that are produced by numerous species to repel insects.[4] Around 6000 known species contain pyrrolizidine alkaloids, resembling roughly 3% of the world's flowering plants.[5]

Many pyrrolizidine alkaloids are highly hepatotoxic and have shown to be genotoxic and show anti-tumor activities.[6] In the last decades, numerous synthetic approaches have been developed that often involve 1,3-dipolar cycloadditions,[7] ring-closing meta-thesis,[8] or proline-based structure modifications.[9] A more complex structural combination of ID and the pyrrolizidine motif is the benzopyrrolizidine or pyrrolo[2,1a]isoindolinone skeleton (BPD, Scheme 1). This compound class is much less studied than the components ID or



Scheme 1. The basic heterocyclic skeletons of isoindolinone (ID) and benzopyrrolizidine (BPD) and two pharmacologically active urea-functionalized BPD **1a**, **b**.

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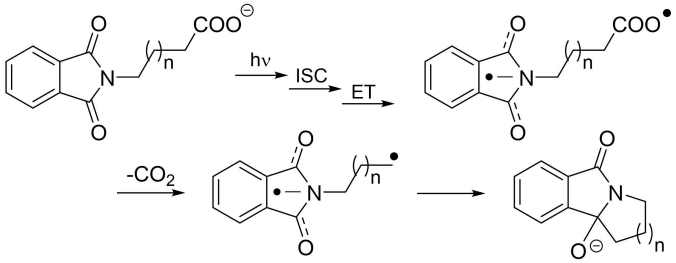
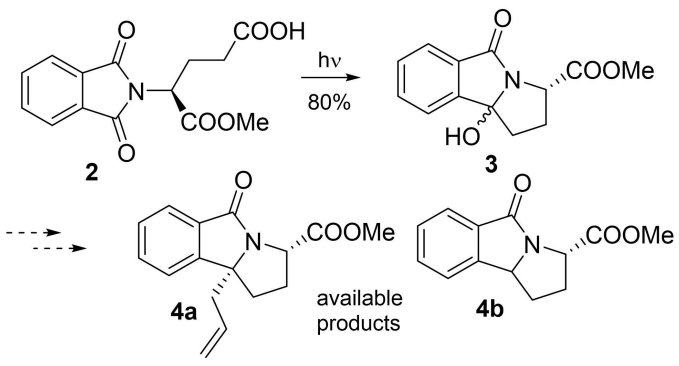
pyrrolizidines and only few synthetic approaches were published.[10]

Recently, structurally simple urea-linked BPD **1a**, **b** were identified as potential inhibitors of cyclin-dependent kinase (CDK).[11] CDK is believed to play a major target for new cancer therapies and efficient routes to these inhibitor structures are thus highly desirable.[12] Photochemical routes to these compounds often involve radical-type carbon-carbon bond formation. In this context, the generation of carbon-centered radicals from alkyl carboxylates by photoinduced oxidation and CO_2 cleavage is an unbeatably simple method.[13] The electronically excited acceptor in the photoinduced electron transfer (PET) redox process has to match the donor oxidation potential which is about 1.3 V (vs. SCE) for alkyl carboxylates.[14] We have investigated in recent years the phthalimide chromophore that exhibits a well-suited long-lived triplet excited state and very short-lived singlets.[15] The PET-induced intermolecular decarboxylation is efficient and, because of the extreme short singlet lifetimes (<200 ps), proofs the involvement of triplet states.[16] The intramolecular photoinduced decarboxylation/cyclization allows the synthesis of a very broad collection of heterocyclic ring systems with highly variable ring sizes ranging from small rings (4–6 ring atoms), medium rings (7–14 ring atoms) to large rings (15–26 ring atoms) with high chemical yields in nearly all cases.[17] Numerous functional groups are also tolerated as part of the donor-acceptor linker chains and thus cyclic ethers, thioethers, crown ethers, lactones, lactames and peptides are accessible by this method.



Scheme 2. Experimentally and spectroscopically established reaction mechanism of triplet excited ω-phthalimido alkylcarboxylate acceptor/donors couples resulting in radical combination and C–C bond formation.



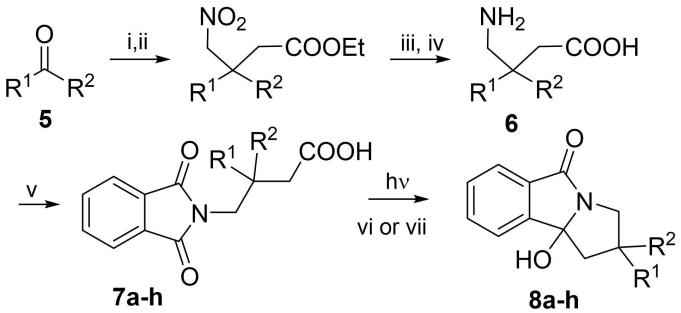
Scheme 3. Photochemical synthesis of the enantiopure benzopyrrolizidines **4** from the glutamic acid derivative ***2*** by photodecarboxylation, cyclization and acyl iminium cation allylation and reduction to **4a**, **b**.

The well-established mechanism of this process involves electron-transfer from the carboxylate to the triplet-excited phthalimide and subsequent decarboxylation and radical combination (Scheme 2). A specific intramolecular chiral pool version (Scheme 3) allows the efficient ring annulation of glutamic acid derivatives.[18] The initially formed hydroxyated BPD **3** can be appropriately modified at the hydroxyl aminal center using standard acyl iminium cation chemistry, e.g., allylation or hydride transfer to give products **4a** and **4b**, respectively.

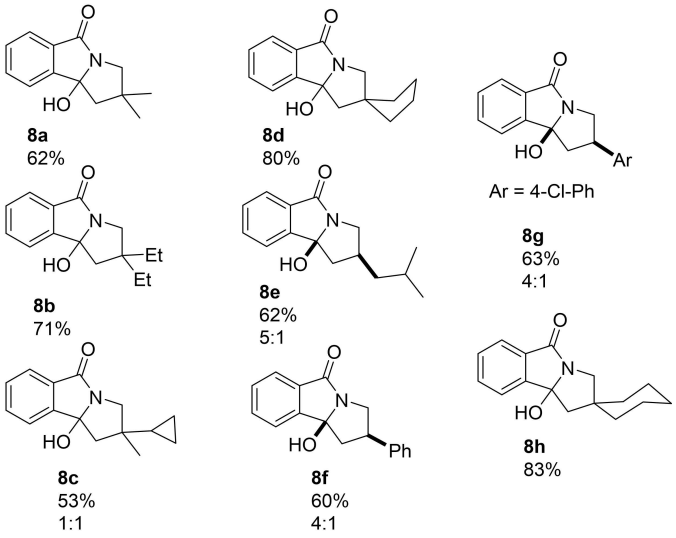
Photochemistry of β- and α-Substituted GABA Substrates

In contrast to this pool of chirally substituted substrates with α-substitution, β- and γ-substituted GABA (GABA = γ-aminobutyric acid) derivatives were not yet investigated. In a first set of experiments, eight β-branched phthalimides **7a–h** were synthesized (**7g** and **7h** from two commercial drugs, the anxiolytic *baclofen* **6g** and the analgesic *gabapentin* **6h**) following the nitronate/Michael addition route as shown in Scheme 4.[19] The β-substituted α-amino acids **6** were obtained in a four-step synthesis from the corresponding ketones or aldehydes, respectively, by a sequence of Horner-Wadsworth-Emmons (HWE) olefination, nitronate addition, catalytic hydrogenation and saponification. The phthalimides **7a–h** were synthesized by thermal condensation in acetic acid.

The direct (acetonitrile, λ_{max} (**7a–h**) $= 295 \pm 5$ nm with $\varepsilon = 1900 \pm 200\ L \cdot mol^{-1}\ cm^{-1}$) as well as the triplet-sensitized (in acetone solution) photolysis of **7a–h** delivered, after workup, the hydroxy benzopyrrolizidines in good chemical yields (Scheme 5). From the chiral substrates **8c**, **8e**, **8f**, and **8g**, cis/trans (with respect to the relative 1,3-configuration) mixtures with preferred formation of the cis-diastereoisomers resulted. Only in case of the β-cyclopropyl-substituted substrate, a 1:1 diastereoisoemric mixture was obtained after photolysis. The baclofen-derived stereoisomers could be separated and trans-



Scheme 4. Syntheses of β-substituted phthaloyl GABA derivatives **7a–h** and photolyses; i: HWE-olefination with triethylphosphonoacetate; ii: nitromethane/ Cs_2CO_3; iii: H_2, Raney-Ni; iv: HCl; v: phthalic anhydride, HOAc; vi: 20 mM **8** in CH_3CN, 0.5 eq. K_2CO_3, hν (300 nm, coated Hg low pressure lamps), or vii: 20–23 mM **8** in acetone/H_2O, 0.5 eq. K_2CO_3 or 1 eq. KOH, hν (300 nm).



Scheme 5. Products from the photodecarboxylative ring annulation of N-phthaloyl GABA derivatives **7a–7h** (major diastereoisomers are shown).

8g (minor diastereoisomer, 20%) was characterized by X-ray structure analysis (Figure 1 and SI).[20]

In a second series of substrates for this synthetic approach, we applied α-substituted GABA derivatives **11a–c**.[21] These starting materials were available by aldol condensation of aldehydes with the γ-butyrolactone enolate, subsequent hydrogenation, and nucleophilic ring-opening (Scheme 6). Triplet-sensitized photolysis delivered the *rac*-benzopyrrolizidines in high chemical yield as cis/trans mixtures. Even when starting with enantiomerically enriched **11a** (80% e.e.), a cis,trans-mixture of benzopyrrolizi-dines **12a** with $<5\%$ e.e. for both diastereoisomers resulted from room temperature photolysis (Scheme 7). This contrasts our results on the enantioselective photocyclization of amide derivatives of anthranilic acid derivatives[22] and shows that conformational biradical flexibility is much higher for the GABA derivatives investigated herein and leads to the rapid loss of memory of chirality.

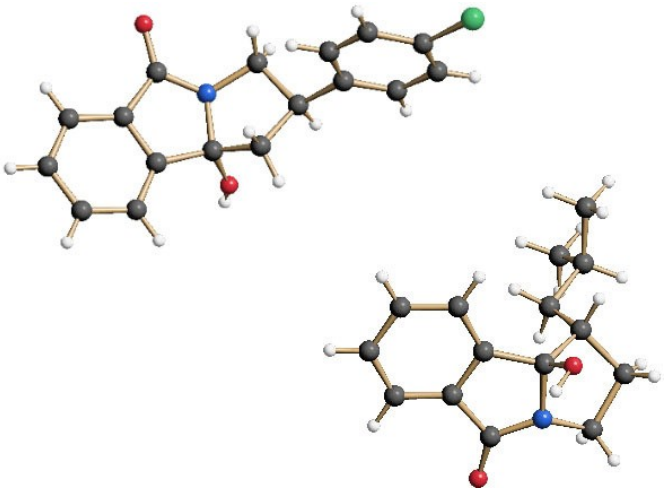
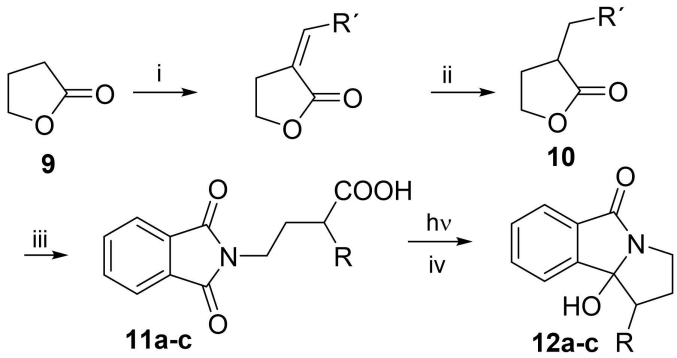
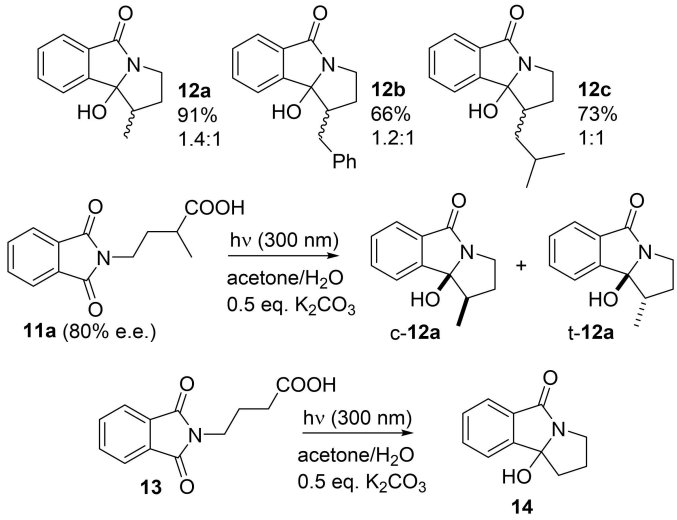


Figure 1. Structures of trans-**8g** and trans-**12c** in the crystal.



Scheme 6. Synthesis of α-substituted phthaloyl GABA derivatives **11a–c** and photolysis; i: LDA, R'CHO; ii: H_2, Raney-Ni; iii: phthalimide sodium; iv: acetone/H_2O, 20 mM solutions of **11**, 0.5 eq. K_2CO_3, hν (300 nm).



Scheme 7. Products **12a–c** from the photodecarboxylative ring annulation of N-phthaloyl GABA derivatives **11a–11c** and model compounds **13** and **14**.

Quantum Yield Determination

The high chemical yields of the photodecarboxylative cyclization of the substrates **7** and **11** indicate an efficient light-induced process. The reactive species for electron transfer is most probably the lowest triplet state with an intrinsic lifetime of 10 μs in acetonitrile and 30 μs in water.[15] These values were determined for the unreactive N-methylphthalimide. For **13** we measured a longest lifetime of 710 ns, attributed to the ring closure reaction. The reaction quantum yield for ISC of N-methylphthalimide was determined to 70% in acetonitrile/water (1:1).[23] For intermolecular decarboxylation processes, reaction quantum yields were determined by chemical actinometry.[24]

In the course of this study, we determined the absolute reaction quantum yield for the photodecarboxylation/cyclization process using substrates **7h** and **11a** together with the standard substrate N-phthaloyl GABA (**13**) by the QYDS (quantum yields determination setup) method developed by Riedle and coworkers.[25] The precisely measured illumination

power, spectral distribution of the LED light and the evolving molecular absorption were used in the evaluation. The conversion was determined from the UV-absorption changes of substrate vs. product and (Figure 2 for **13**, $\lambda_{ex} = 305$ nm, 1.39 mW LED) correlated with the changes in photon flux as determined by a solar cell detector.

All reaction quantum yields Φ_r were determined with a precision of better than 5% relative. The results are summarized in Table 1. During the initial measurements, the optimal concentration of the substrate for quantum yield determination was determined. This study revealed a surprising correlation between substrate concentration and the quantum yield of product formation. For low concentrations of the substrate (50–100 μM) clear isosbestic points are observed pointing at a clean conversion from the substrate to one product. A near unity chemical conversion at sufficiently long illumination time is observed. Accordingly, the final UV/vis spectrum was used as product spectrum. For the parent N-phthaloyl GABA derivative **13**, where substrate concentrations of 45 and 900 μM were tested, and no concentration effect on the quantum yield was found. Also the use of argon (in order to suppress bimolecular triplet quenching by oxygen) did only slightly increase Φ_r.

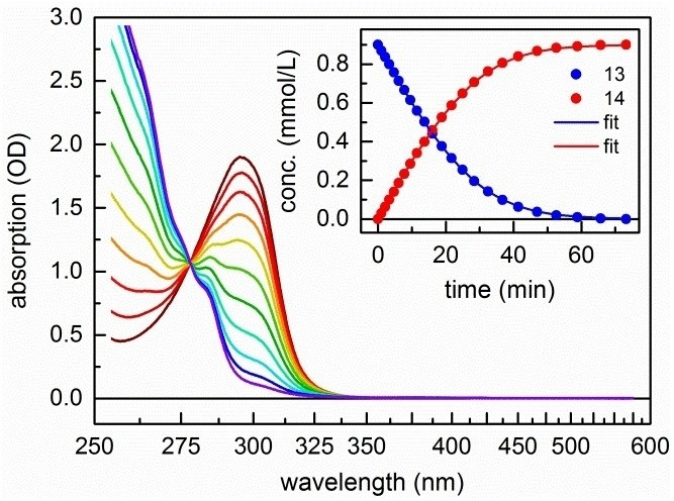


Figure 2. Photolysis of the parent GABA-derivative **13** (900 μM in CH_3CN/H_2O, 3:1, 450 μM K_2CO_3). Shown are the changes in UV absorption with initial 1 min irradiation steps and the isosbestic point at 278 nm.

Table 1. Reaction quantum yield for selected substrates and concentrations.

	λ_{exc} (nm)	concentration (μmol/L)	Pill (mW)	QY (%)
7h	305	85	7.90	10.4
"	305	837	10.28	14.6
11a	305	45	5.90	41.3
"	305	905	4.25	48.7
13	305	45	3.88	34.0
"	305	900	1.39	34.0
13[a]	305	950	4.24	34.4

[a] degassed with Ar.

In the case of **7h** and **11a** we found a significant increase of the quantum yield for higher concentrations approaching 1 mM. This is shown for substrate **7h** in Figures 3 and 4. This effect might originate from increased aggregation of the substrate at higher concentrations which we also have detected in the biological testing experiments (vide infra and SI) when increasing the concentration above 200 μM. In the actual preparative experiments, much higher substrate concentrations were used (> 20 mM) and thus aggregation effects are expected to become dominant in the synthesis. The aggregation might also explain the very high intramolecular cyclization efficiency even for long-chain separated phthalimido-alkyl carboxylates. In this case aggregation is expected to be stabilized by strong ground-state donor-acceptor interactions.

Inhibition of human cholesterol esterase. The serine hydrolase cholesterol esterase (CEase) is thought to be involved in the development of atherosclerosis, leading to coronary heart disease. Pharmacological inhibition of CEase might therefore have a beneficial effect on the course of this disease.[26] Recently, two ω-phthalimidoalkyl aryl ureas were identified as promising inhibitors of the human enzyme (hCEase).[27] In the present study, we therefore investigated the inhibitory activity of the related pyrroloisoindolinones **8a**–**8h** against hCEase.

An initial screening showed derivatives **8a** and **8d** (25 μM) to act as moderate inhibitors of hCEase decreasing the enzymatic activity to 65 ± 8% and 61 ± 5%, respectively (Figure 5, top). For further characterization, we determined the effect of increasing concentrations of **8a** and **8d** on the hCEase activity (Figure 5, bottom). We did not observe complete enzyme inhibition and therefore analyzed the data according to a model of hyperbolic inhibition[28] as recently done for inhibition of human and murine CEase by two thieno[1,3]oxazin-4-ones.[27] This analysis resulted in values of

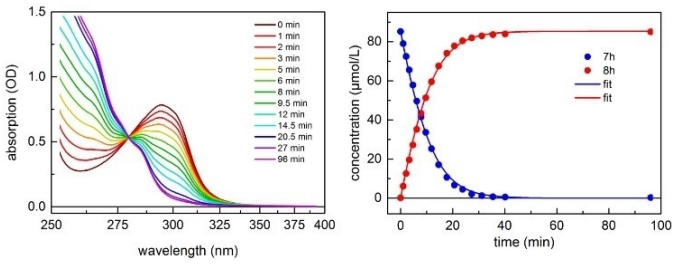


Figure 3. Photolysis of **7h** (85 μM in CH_3CN/H_2O, 3:1, 42.5 μM K_2CO_3): UV absorption with initial 1 min irradiation steps and concentration/time profile.

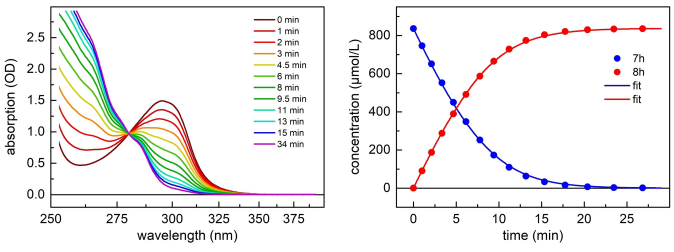


Figure 4. Photolysis of **7h** (837 μM in CH_3CN/H_2O, 3:1, 420 μM K_2CO_3): UV absorption with initial 1 min irradiation steps and concentration/time profile.

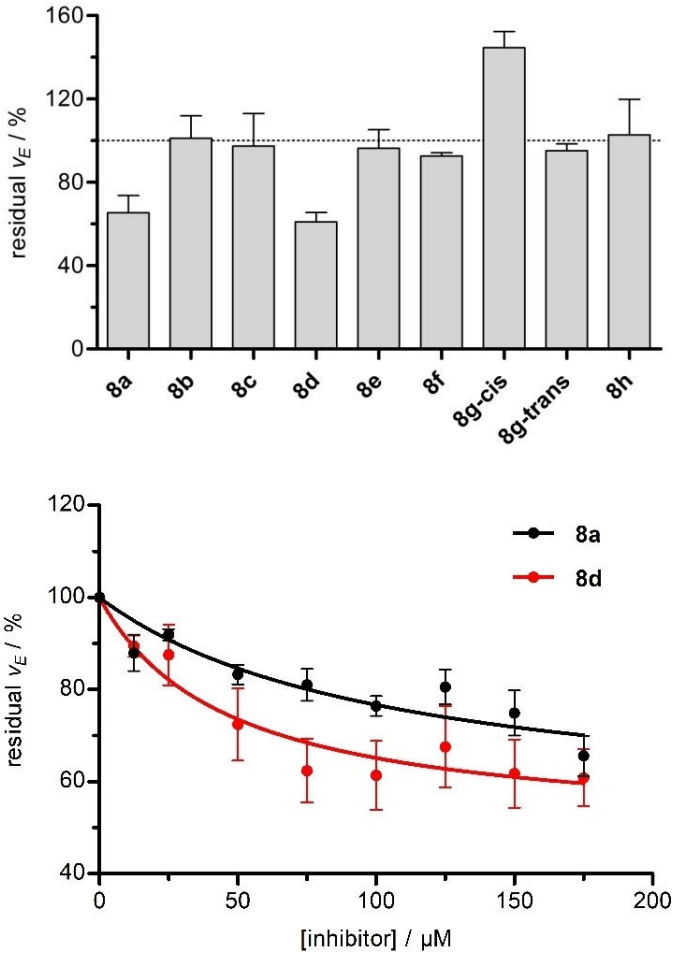


Figure 5. Inhibition of hCEase by pyrroloisoindolinones **8a**–**8h**. Shown are percentage residual enzymatic activities (mean ± SEM) from three to four independent experiments each performed in triplicate; **top**: screening for inhibition at 25 µM, **bottom**: determination of IC_{50} values of **8a** and **8d**, respectively, according to a hyperbolic mode of inhibition.

IC_{50} of 79 ± 38 µM (**8a**, n = 3) and 65 ± 23 µM (**8d**, n = 4), respectively, and additionally allowed for the calculation of the percentage residual enzyme activity at infinite high inhibitor concentrations ($v_{[I]\to\infty}$ of 57 ± 14% and 44 ± 11%, respectively). In contrast to the recently investigated ω-phthalimidoalkyl aryl ureas which completely inactivated hCEase at high concentrations,[27] the here observed hyperbolic inhibition by the pyrroloiso-indolinones points towards an active enzyme-substrate-inhibitor complex still capable of product formation.

Conclusions

The photodecarboxylative ring annulation for α- and β-substituted phthaloyl GABA derivatives is a high-yielding triplet process and leads to biologically active pyrroloisoindolinones. The reaction quantum yield Φ_r for direct photolysis is between 10% and 49%, depending on the substitution pattern. The unsubstituted GABA derivative **13** reacts with a concentration-independent Φ_r of 34%, whereas substrates **7h** (β,β'-disubstituted) and **11a** (α-monosubstituted) show concentration-dependent values of Φ_r that point to aggregation-induced acceleration of the reaction-determining electron transfer.

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Conflict of Interests

The authors declare no conflict of interest.

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

Keywords: Heterocycles · photochemistry · radicals · decarboxylation · quantum yields

[1] K. Speck, T. Magauer, *Beilstein J. Org. Chem.* **2013**, *9*, 2048–2078.
[2] W. Gao, M. W. Chen, Q. P. Ding, Y. Y. Peng, *Chem. Asian J.* **2019**, *14*, 1306–1322.
[3] a) P. J. Parsons, D. R. Jones, L. J. Walsh, L. A. T. Allen, A. Onwubiko, L. Preece, J. Board, A. J. P. White, *Org. Lett.* **2017**, *19*, 2533–2535; b) S. Hu, L. Yuan, H. Yan, Z. Li, *Bioorg. Med. Chem. Lett.* **2017**, *27*, 4075–4081; c) B. Flores, T. F. Molinski, *Org. Lett.* **2011**, *13*, 3932–3935; d) D. Augner, H.-G. Schmalz, *Synlett* **2015**, *26*, 1395–1397; e) S. Das, D. Addis, L. R. Knöpke, U. Bentrup, K. Junge, A. Brückner, M. Beller, *Angew. Chem.* **2011**, *123*, 9346–9350.
[4] J. Robertson, K. Stevens, *Nat. Prod. Rep.* **2017**, *34*, 62–89.
[5] S. Schramm, N. Köhler, W. Rozhon, *Molecules* **2019**, *24*, AN 498.
[6] a) N. Li, Q. S. Xia, J. Q. Ruan, P. P. Fu, G. Lin, *Curr. Drug Metab.* **2011**, *12*, 823–834; b) P. P. Fu, Q. S. Xia, G. Lin, M. W. Chou, *Drug Metab. Rev.* **2004**, *36*, 1–55.
[7] a) C. Najera, J. M. Sansano, *Pure Appl. Chem.* **2019**, *91*, 575–596; b) A. Brandi, F. Cardona, S. Cicchi, F. M. Cordero, A. Goti, *Chem. Eur. J.* **2009**, *15*, 7808–7821.
[8] R. Ben-Othman, M. Othman, K. Ciamala, M. Knorr, C. Strohmann, B. Decroix, *Tetrahedron* **2009**, *65*, 4846–4854.
[9] C. Bhat, S. G. Tilve, *RSC Adv.* **2014**, *4*, 5405–5452.
[10] a) Z. Huang, Q. Chen, X, Yang, Y. Liu, L. Zhang, T. Lu, Q. Zhou, *Org. Chem. Front.* **2017**, *4*, 967–971; b) F. Scorzelli, A. Di Mola, L. Palombi, A. Massa, *Molecules* **2015**, *20*, 8484–8498.
[11] T. Honma, K. Hayashi, T. Aoyama, N. Hashimoto, T. Machida, K. Fukasawa, T. Iwama, C. Ikeura, M. Ikuta, I. Suzuki-Takahashi, Y. Iwasawa, T. Hayama, S. Nishimura, H. Morishima, *J. Med. Chem.* **2001**, *44*, 4615–4627.
[12] a) M. Peyressatre, C. Prevel, M. Pellerano, M. C. Morris, *Cancers* **2015**, *7*, 179–237; b) M. D. Garrett, A. Fattaey, *Curr. Opin. Genet. Dev.* **1999**, *9*, 104–111; c) J. Wesierska-Gadek, M. P. Kramer, *Future Med. Chem.* **2012**, *4*, 395–424.
[13] J. Schwarz, B. König, *Green Chem.* **2018**, *20*, 323–361.
[14] L. Capaldo, L. Buzetti, D. Merli, M. Fagnoni, D. Ravelli, *J. Org. Chem.* **2016**, *81*, 7102–7109.
[15] A. Reiffers, C. Torres Ziegenbein, L. Schubert, J. Diekmann, K. Thom, R. Kühnemuth, A. G. Griesbeck, O. Weingart, P. Gilch, *Phys. Chem. Chem. Phys.* **2019**, *21*, 4839–4853.
[16] A. G. Griesbeck, J. Neudörfl, B. Goldfuss, S. Molitor, *ChemPhotoChem* **2017**, *1*, 355–362.

[17] A. G. Griesbeck, N. Hoffmann, K.-D. Warzecha, *Acc. Chem. Res.* **2007**, *40*, 128–140.
[18] A. G. Griesbeck, F. Nerowski, J. Lex, *J. Org. Chem.* **1999**, *64*, 5213–5217.
[19] P. G. Nantermet, J. C. Barrow, S. R. Lindsley, M. Young, S.-S. Mao, S. Carroll, C. Bailey, M. Bosserman, D. Colussi, D. R. McMasters, J. P. Vacca, H. G. Selnick, *Bioorg. Med. Chem. Lett.* **2004**, *14*, 2141–2145.
[20] Data for the X-ray structures analyses of compounds trans-**8 g**, **8 h**, and trans-**12** were deposited with CCDC numbers: CCDC 1983341 (**8 g**), CCDC 1983342 (**8 h**), and CCDC 1983343 (trans-**12 c**).
[21] K. Tanaka, N. Tamura, A. Kaji, *Chem. Lett.* **1980**, *9*, 595–598.
[22] A. G. Griesbeck, W. Kramer, J. Lex, *Angew. Chem. Int. Ed.* **2001**, *40*, 577–579.
[23] K.-D. Warzecha, H. Görner, A. G. Griesbeck, *J. Phys. Chem. A.* **2006**, *110*, 3356–3363.
[24] a) H. J. Kuhn, S. E. Braslavsky, R. Schmidt, *Pure Appl. Chem.* **2004**, *76*, 2105–2146; b) S. P. Pitre, C. D. McTiernan, W. Vine, R. DiPucchio, M. Grenier, J. C. Scaiano, *Sci. Rep.* **2015**, *5*, 16397; c) H. de Lasa, B. S. Rosales, J. Moreira, P. Valades-Pelayo, *Chem. Eng. Technol.* **2016**, *39*, 51–65.
[25] a) U. Megerle, R. Lechner, B. König, E. Riedle, *Photochem. Photobiol. Sci.* **2010**, *9*, 1400–1406; b) H. Volfova, Q. Hu, E. Riedle, *EPA Newslett.* **2019**, *96*, 51–69.
[26] J. E. Heidrich, L. M. Contos, L. A. Hunsaker, L. M. Deck, D. L. Vander Jagt, *BMC Pharmacol.* **2004**, *4*, 5.
[27] F. M. Dato, M. Sheikh, R. Z. Uhl, A. W. Schüller, M. Steinkrüger, P. Koch, J.-M. Neudörfl, M. Gütschow, B. Goldfuss, M. Pietsch, *ChemMedChem* **2018**, *13*, 1833–1847.
[28] I. H. Segel, *Enzyme Kinetics: Behavior and Analysis of Rapid Equilibrium and Steady-State Enzyme Systems*, Wiley, Hoboken, **1993**, 100–226.

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