# Chemiluminescence enhancement by 6,8-dialkylated luminols

Chemiluminescence describes a chemical reaction which produces an electronically excited species that emits light. Only very few substrates are known to undergo such processes which find applications in the efficient detection of trace quantities of chemicals (e.g., luminol test for iron, nitric oxide test, DNA sequencing, Western blot). Many organisms utilize this phenomenon for signaling and communication. Since its discovery by Albrecht in 1928, the chemiluminescence of the eponymic luminol (5-amino-2,3-dihydro-1,4-phthalazinedione, **1**) has been intensively studied and widely used for forensic, environmental, and biomedical applications, including immunoassays (Western blot), the monitoring of metabolic pathways, and the detection of free radicals and trace amounts of reactive oxygen species. Chemiluminescence of **1** is even more effective in the presence of various metal ions which has led to numerous applications of combinations of **1** with metal ions, complexes, or nanoparticles, respectively, and oxidants. However, the detailed mechanism of chemiluminescence and its modulation by substituents or the presence of various additives are still elusive.

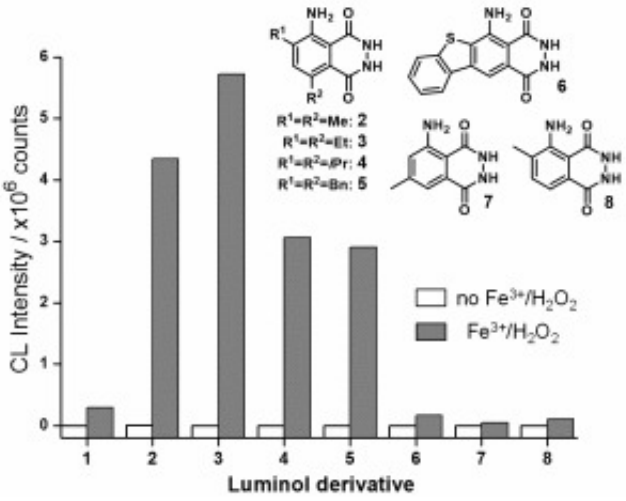


Fig. 1. Substituted 6,8-dialkyl luminol derivatives and CL enhancement of these substrates in the absence (open boxes) and presence (shaded boxes) of the catalyst/oxidant system.

Thus, the investigation of new luminol derivatives can provide structure–activity data which may foster a deeper understanding of the underlying photochemical processes and their application to demanding analytical problems. There are only few reports on substituted luminol derivatives, mainly due to a lack of robust and modular syntheses. However, the demonstration of significant chemiluminescence modulation induced by electron-rich substituents has fueled our endeavor into the evaluation of new luminol derivatives bearing alkyl substituents. The introduction of two

substituents vicinal to the amino and one carboxamide group at C6 and C8 is believed to have a pronounced effect on the structure of the excited states. Simple alkyl substituents are stable and lack the presence of polar sites amenable to strong solvent interactions (Fig. 1). Such steric modulation can easily be applied to other photosystems and is complementary to electronic perturbations by heteroatomic electron donating group substituents or chromophore extensions by benzannulation.

The chemiluminescence of the luminol derivatives **2-8** (vs. parent luminol, **1**) in the presence of metals as promotors and aqueous $H_2O_2$ as crucial oxidant has been investigated in detail. The results in this work reveal a surprising 20-fold chemiluminescence enhancement (Fig. 1) induced by simple alkyl substituents in positions 6 and 8 of the luminol skeleton (derivatives **2-5**). The mechanistic rationalization of the unprecedented chemiluminescence enrichment from these alkyl luminols has been studied by theoretical calculations where could be interpreted as a “steric gearing” effect on surface crossings (Fig. 2). This effect appears to be a direct consequence of vicinal steric gearing which improves the transition efficiency from an intermediary endoperoxide to the excited phthalate.

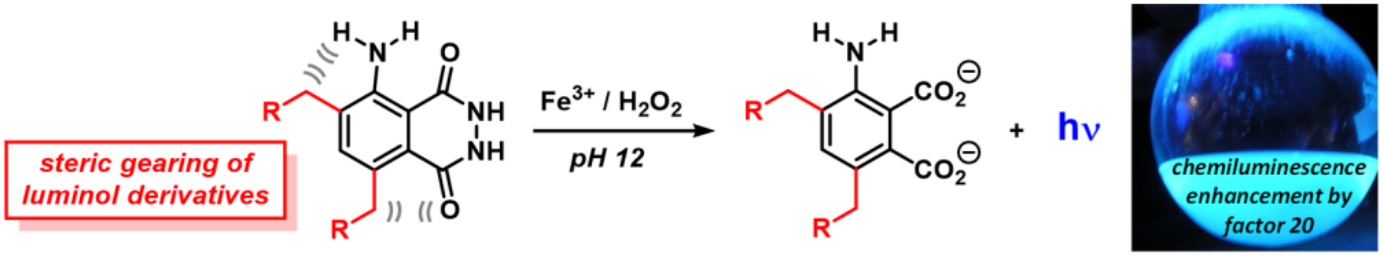


Fig. 2.

This concept of a mostly steric modulation complements those involving strong electronic variations of the luminophore or the extension of the conjugation length. The application of such rather subtle structural manipulation of photoactive molecules holds great potential for the design and amplification of various photophysical processes and eventually chemiluminescence properties. This might lead to improvements of the efficiency of current luminescence tools for the detection of small molecules or imaging.

## Publication

Steric Enhancement of the Chemiluminescence of Luminols.
Griesbeck AG, Díaz-Miara Y, Fichtler R, Jacobi von Wangelin A, Pérez-Ruiz R, Sampedro D

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