Boosting Bioorthogonal Click-to-Release Chemistry by Intramolecular Control And Acceleration of Post-Click Tautomerization

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Among the chemical tools for bioorthogonal elimination reactions, the tetrazine-triggered cleavage of *trans*-cyclooctene conjugates (click-to-release chemistry) has outstanding click kinetics but the rate and efficiency of the subsequent release step vary significantly depending on the chemical structure of the applied tetrazine. The major drawback is that only poor release has so far been obtained when using the reagents of choice to achieve fast click reactions (highly reactive tetrazines such as bis-pyridyl-tetrazines), whereas less reactive tetrazines lead to complete and fast release.

Recently, reported mechanistic insights into the post-click reaction network of click-to-release chemistry (Fig. 1) uncovered the key role of post-click tautomerization in the overall outcome of the reaction. [1]

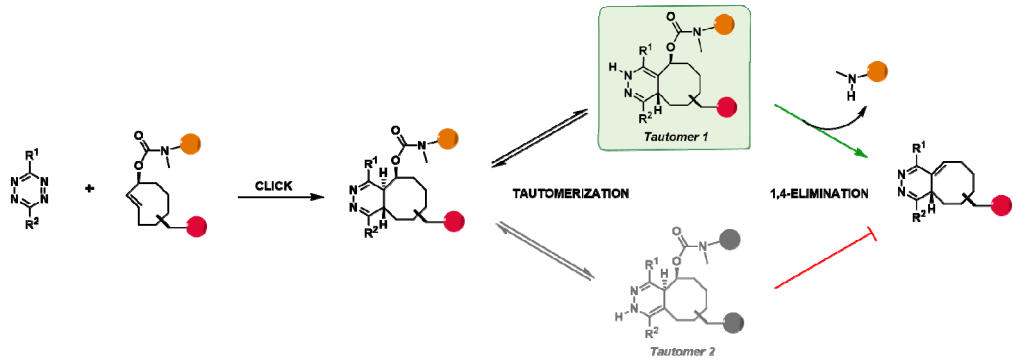

Fig. 1: Proposed mechanistical pathways of the post-click reaction, adapted from Carlson *et al.* [1]

Based on an improved understanding of the underlying mechanisms it has been suggested that directed and accelerated tautomerization leading to exclusive formation of *tautomer 1* displays the optimal route to significantly enhance release. [1]

To this end, we incorporated a functionality featuring directing effects tethered to the *trans*-cyclooctene and achieved a dramatic acceleration of the desired tautomerization pathway leading to a boost of the overall reaction by three orders of magnitude compared to the current state-of-the-art. Details regarding synthesis, measurements and results will be presented.

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[1] Jonathan C. T. Carlson, Hannes Mikula, Ralph Weissleder, *J. Am. Chem. Soc.* **2018**, *140*, 3603-3612.