

NOVEL ADDRESSABLE CHOLESTEROL ANALOGUES FOR LIVE IMAGING OF CELLULAR MEMBRANES

Tristan Wegner^a, Lena Roling^a, David Grill^b, Anna L. L. Matos^b, Stephanie Wulff^c,
Da Wang^c, Jonas Börgel^a, Martin Körsgen^d, Heinrich F. Arlinghaus^d,
Hans-Joachim Galla^c, Volker Gerke^b, Frank Glorius^a

^aInstitute of Organic Chemistry

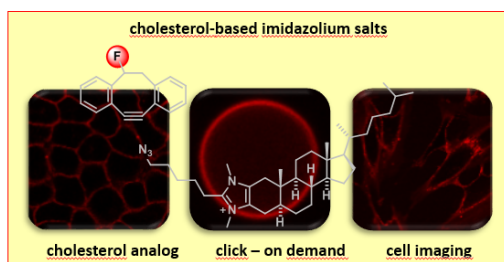
^bInstitute of Medical Biochemistry

^cInstitute of Biochemistry

^dInstitute of Physics

University of Münster, 48149 Münster, Germany

Cholesterol is an essential component of most biological membranes and serves important functions in controlling membrane integrity, organization and signaling.^[1] However, probes to follow the dynamic distribution of cholesterol in live cells are scarce and so far show only limited applicability.^[2] Herein, we addressed this problem by synthesizing and characterizing a novel class of versatile and clickable cholesterol-based imidazolium salts.^[3] We show that these cholesterol analogues faithfully mimic the biophysical properties of natural cholesterol in phospholipid mono- and bilayers and that they integrate into the plasma membrane of cultured and primary human cells. The membrane-incorporated cholesterol analogues can be specifically labelled by click chemistry and visualized in live cell imaging experiments that show a distribution and behavior comparable to that of endogenous membrane cholesterol. These results indicate that the novel cholesterol analogues can be used to reveal the dynamic distribution of cholesterol in live cells. We envision that our highly modifiable analogues will ultimately serve as a flexible toolbox for the investigation of cellular membrane processes.



[1] Ikonen, E., *Nat. Rev. Mol. Cell. Biol.* **2008**, *9*, 125; Brown, D. A., London, E., *J. Biol. Chem.* **2000**, *275*, 17221; Simons, K., Ikonen, E., *Science* **2000**, *290*, 1721.

[2] Gimpl, G., Gehrig-Burger, K., *Biosci. Rep.* **2007**, *27*, 335; Maxfield, F. R., Wüstner, D., *Methods Cell Biol.* **2012**, *108*, 367.

[3] Rakers, L., Grill, D., Matos, A. L. L., Wulff, S., Wang, D., Börgel, J., Körsgen, M., Arlinghaus, H. F., Galla, H.-J., Gerke, V., Glorius, F., *Cell Chem. Biol.* **2018**, *25*, 952.