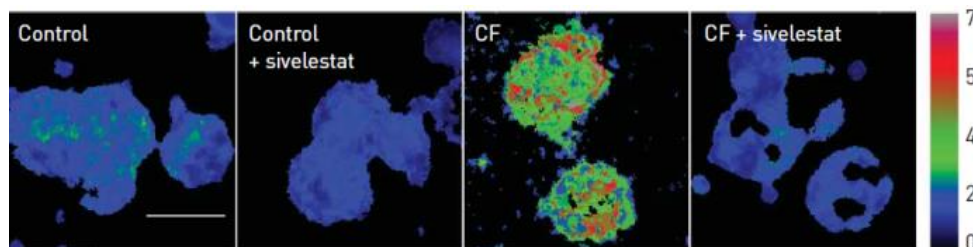


Nemo FRET reporter



Neutrophil elastase (NE) activity is increased on the surface of cystic fibrosis (CF) sputum neutrophils: Representative ratio images calculated from donor and acceptor fluorescence of sputum neutrophils from a healthy nonsmoker (control) and a patient with CF. Scale bar=15 μ m.

Neutrophil elastase (NE) is a novel biomarker in patients with cystic fibrosis (CF) and potentially other chronic neutrophilic lung diseases such as COPD. NEMo is a FRET-based reporter system that detects NE activity on cell surfaces or in solution.

NE is considered a key risk factor for severity of **cystic fibrosis (CF)** lung disease ②. Surface-bound NE activity is associated with lung disease severity in adult CF patients who exhibit high levels of neutrophilic airway inflammation ③.

To measure NE activity in solution, use the highly sensitive FRET reporter **NEMo-1**. On cell surfaces use the lipidated reporter **NEMo-2E**.

NEMo-1 was successfully used to assess the potential of native bronchoalveolar lavage fluid (BALF) from CF children.

NE activity is consistently increased on the surface of CF sputum neutrophils and is considered a marker for severity of lung disease in patients with CF. Recently, Nemo-2E was successfully used for a FRET readout by FACS (①). In addition, the probes are useful to monitor NE inhibitors on the microscope stage or in a plater reader format.

The two reporters are available at these sizes:

FRET reporter NEMo-1:

SC-0200: 10 μ L: 200 EUR
 50 μ L: 600 EUR

Lipidated FRET reporter NEMo-2E:

SC-0201: 10 μ L: 200 EUR
 50 μ L: 600 EUR

- ▶ 10 μ L are sufficient for approx. **8 FACS- OR 12 microscopy** experiments
- ▶ soluble in buffer
- ▶ or use a stock solution in DMSO

For your experiments

Sputum was separated from saliva and homogenised using 10% sputolysin. Sputum inflammatory cells were isolated and total immune cell count was performed. **Free NE-activity** was quantified in sputum supernatants using the soluble FRET probe **NEMo-1** and compared with activity levels determined by the chromogenic substrate MeO-Suc-AAPV-pNA.

Surface-bound NE-activity on sputum neutrophils was measured using the lipidated FRET reporter **NEMo-2E** and determined from the change in ratio of donor to acceptor fluorescence over time.

For more detailed information just have a look at the JoVE-Video (62193):

NEMo: New FRET-based reporter system that detects NE activity



play video

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Publications

- ❶ New method for rapid and dynamic quantification of elastase activity on sputum neutrophils from patients with cystic fibrosis using flow cytometry// PMID: 32139467 (2020)
- ❷ Elastase activity on sputum neutrophils correlates with severity of lung disease in cystic fibrosis PMID: 29545279 (2018)
- ❸ Elastase Exocytosis by Airway Neutrophils Is Associated with Early Lung Damage in Children with Cystic Fibrosis PMID: 30281324 (2019)
- ❹ Lack of neutrophil elastase reduces inflammation, mucus hypersecretion, and emphysema, but not mucus obstruction, in mice with cystic fibrosis-like lung disease PMID: 24678594 (2014).
- ❺ Macrophage PD-1 associates with neutrophilia and reduced bacterial killing in early cystic fibrosis airway disease PubMed ID: 35732550 (2022)
- ❻ Pilot study of inflammatory biomarkers in matched induced sputum and bronchoalveolar lavage of 2-year-olds with cystic fibrosis PubMed ID: 35637404 (2022)
- ❼ Relationship between airway dysbiosis, inflammation and lung function in adults with cystic fibrosis PubMed ID: 33431308 (2021)
- ❽ Transcriptional firing represses bactericidal activity in cystic fibrosis airway neutrophils PubMed ID: 34573044 (2021)
- ❾ Elastase Exocytosis by Airway Neutrophils Is Associated with Early Lung Damage in Children with Cystic Fibrosis PubMed ID: 33948572 (2019)

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