

## Product Information

## Duolink® flowPLA Detection Kit - Violet

Duolink® PLA kit for Flow Cytometry with Violet Detection

**DUO94005**

### Product Description

Duolink® flowPLA Detection Kit - Violet contains all the necessary Duolink® PLA reagents to perform the amplification and detection of bound PLA probes by flow cytometry. The detection oligonucleotides contain a fluorophore ( $\lambda_{\text{ex}} = 390 \text{ nm}/\lambda_{\text{em}} = 476 \text{ nm}$ ), which may be excited using the 405 nm violet laser line.

Experiments conducted using Duolink® flowPLA reagents can detect protein interactions, protein expression levels, and post-translational modifications at the single molecule level in fixed, suspended cells.

### Components

Sufficient components are provided for 40 tests, based on 100  $\mu\text{L}$  total reaction volume covering 100,000 cells.

Component	Volume
5x Ligation Buffer: Contains oligonucleotides that hybridize to the PLA probes and all components needed for ligation except the ligase. DUO82009-40 TST	800 $\mu\text{L}$
Ligase (1 unit/ $\mu\text{L}$ ) DUO82027	100 $\mu\text{L}$
Polymerase (10 units/ $\mu\text{L}$ ) DUO82028	50 $\mu\text{L}$
5x Amplification Buffer: Contains all components needed for rolling-circle amplification (RCA) except the polymerase. DUO82050-40 TST	800 $\mu\text{L}$
5x Detection Solution Violet: Contains oligonucleotides labeled with a fluorophore that hybridize to the RCA product. DUO824005-40 TST	800 $\mu\text{L}$

### Reagents Required

(But not provided)

To perform a complete Duolink® flowPLA experiment, one will need two primary antibodies (IHC or ICC/IF validated) that recognize two target epitopes.

Additional reagents include a pair of PLA probes (one PLUS and one MINUS) and flowPLA detection reagents of choice. Recommended reagents include Duolink® Wash Buffers and PBS.

### Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

### Preparation Instructions

Thaw the 5x Ligation, 5x Amplification, and 5x Detection Violet buffers at room temperature and vortex before use. Dilute the required volumes of each 5x solution 5-fold with ultrapure water immediately before use. Do not store diluted reagents. The Duolink® Detection solutions are light-sensitive. Protect from light.

**Note:** The 5x Ligation Buffer contains DTT that may precipitate at  $-20^\circ\text{C}$ . Make sure the DTT is completely dissolved and vortexed before use.

The ligase and polymerase enzymes should be kept cold ( $-20^\circ\text{C}$ ) at all times; use a freezing block when removing them from the freezer. Quick spin the vial before pipetting. Add the enzyme to the appropriate reaction mix immediately before use. Vortex the mix after addition of enzyme. Do not store diluted reagents.

## Storage/Stability

Store the flowPLA reaction components at  $-20^{\circ}\text{C}$ . The enzymes should be kept cold ( $-20^{\circ}\text{C}$ ) at all times, use a freezing block when removing them from the freezer.

## Procedure

The experimental procedures for Duolink® PLA Flow Cytometry application can be found at [sigma.com/duolink](https://sigma.com/duolink).

**Note:** Duolink® PLA reagent volumes are based on 40  $\mu\text{L}$  reaction volume for a 1  $\text{cm}^2$  sample on a microscope slide or 100  $\mu\text{L}$  reaction volume at  $\sim 1,000$  cells/ $\mu\text{L}$  for flow cytometry. However, volumes may need to be adjusted according to the sample size or number of cells of your sample.

## Notice

We provide information and advice to our customers on application technologies and regulatory matters to the best of our knowledge and ability, but without obligation or liability. Existing laws and regulations are to be observed in all cases by our customers. This also applies in respect to any rights of third parties. Our information and advice do not relieve our customers of their own responsibility for checking the suitability of our products for the envisaged purpose.

The information in this document is subject to change without notice and should not be construed as a commitment by the manufacturing or selling entity, or an affiliate. We assume no responsibility for any errors that may appear in this document.

## Technical Assistance

Visit the tech service page at [SigmaAldrich.com/techservice](https://SigmaAldrich.com/techservice).

## Standard Warranty

The applicable warranty for the products listed in this publication may be found at [SigmaAldrich.com/terms](https://SigmaAldrich.com/terms).

## Contact Information

For the location of the office nearest you, go to [SigmaAldrich.com/offices](https://SigmaAldrich.com/offices).

The life science business of Merck operates as MilliporeSigma in the U.S. and Canada.

Merck, Duolink, PLA, and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. All other trademarks are the property of their respective owners. Detailed information on trademarks is available via publicly accessible resources.

© 2021 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.

DUO94005dat Rev 06/21

The Merck logo, consisting of the word "MERCK" in a bold, red, sans-serif font.