

# COLOR DE LYS®

## HDAC colorimetric activity assay kit

### Simplify HDAC activity analysis with the fastest colorimetric assay on the market

Useful for assaying lysates, immunoprecipitates or inhibitor screening using the nuclear extract provided.- Includes HeLa nuclear extract, a rich source of HDACs 1 & 2 for use as a positive control or as a source of HDAC activity for screening.

Compatible with class I & IIb HDAC and sirtuins (with addition of NAD<sup>+</sup>). The COLOR DE LYS® system (Colorimetric Histone de Acetylase Lysyl Substrate/Developer) is a sensitive and convenient alternative to protocols utilizing radiolabeled, acetylated histones or peptide/HPLC methods for the assay of histone deacetylases. The assay procedure has two steps. First, the COLOR DE LYS® substrate which comprises an acetylated lysine side chain, is incubated with a sample containing HDAC activity (HeLa nuclear or other extract, purified enzyme, bead bound immunocomplex, etc.). Deacetylation of the substrate sensitizes the substrate so that, in the second step, mixing with the COLOR DE LYS® developer causes an increase in yellow color intensity, and absorption at 405 nm.

**Note:** The COLOR DE LYS® substrate is efficiently deacetylated by HDAC1 and HDAC2, the major contributors to HDAC activity in HeLa nuclear extracts. It is however, a poor substrate for HDAC3 and recombinant human HDAC8 (BIOMOL International, unpublished results). The activities of other HDAC isotypes with the COLOR DE LYS® substrate have yet to be investigated. HDAC3 and HDAC8 do deacetylate the FLUOR DE LYS® substrate, which is the basis for the HDAC fluorometric activity assay kit (Prod. No. BML-AK500).

Citations: 14

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### Ordering Information

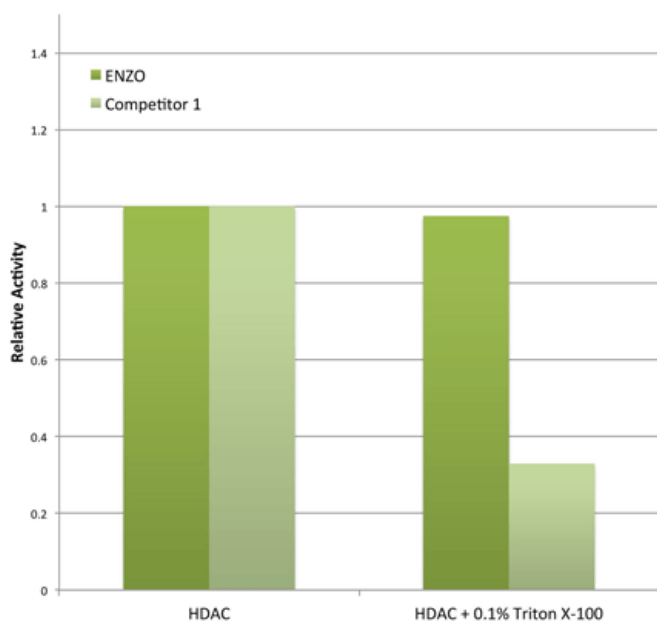
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BML-AK501-0001	96 wells
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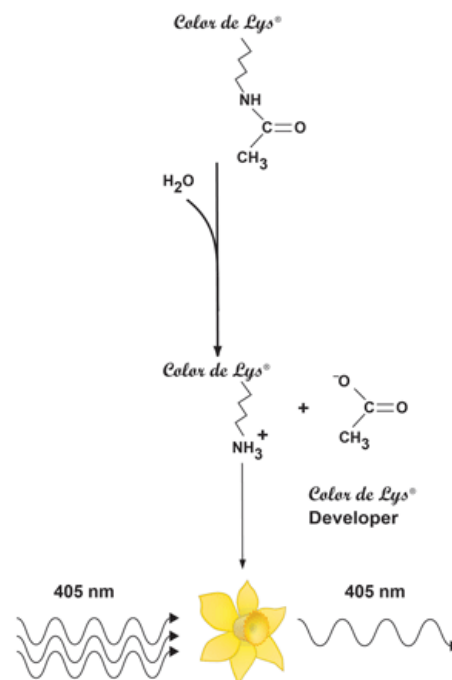
### Manuals, SDS & CofA

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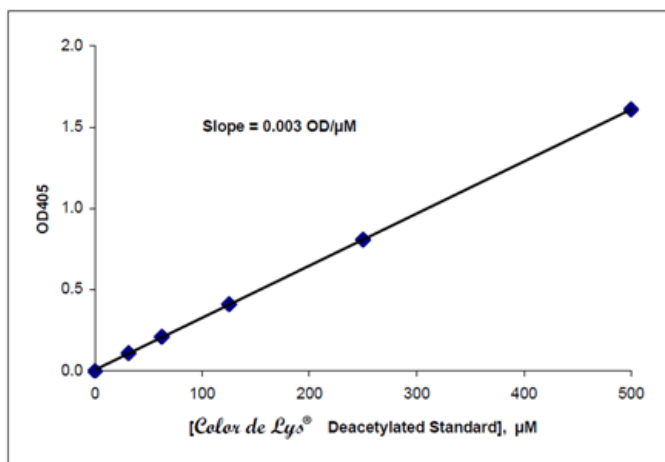
- Simple two-step protocol with < 1 hour time to answer
- Colorimetric readout at 405 nm compatible with most plate readers
- Resistant to detergent interference common to antibody-based assays
- Eliminates need for radioactivity, extractions, and/or chromatography
- Suitable for high throughput analysis



Our Color-de-Lys HDAC colorimetric assay is less sensitive to detergents than competitive antibody-based assays. HeLa nuclear extract (8.3µg) was added to the substrate and buffer recommended by the manufacturer in the presence or absence of 0.1% Triton X-100. After 60 minutes at 37°C, the reaction was stopped and processed as recommended by the manufacturers. Triton X-100 showed little or no effect on the Color-de-Lys reaction, but caused an apparent 70% inhibition of the antibody-based assay.



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Reaction Scheme of the HDAC Colorimetric Activity Assay (patent pending). Deacetylation of the substrate sensitizes it to the developer, which causes an increase in yellow intensity and absorption at 405 nm.

# Handling & Storage

**Use/Stability** Store all components except the microtiter plate at -80°C for the highest stability. The HeLa Nuclear Extract must be handled with particular care in order to retain maximum enzymatic activity. Defrost it quickly in a RT water bath or by rubbing between fingers, then immediately store on an ice bath. The remaining unused extract should be refrozen quickly, by placing at -80°C. If possible, snap freeze in liquid nitrogen or a dry ice/ethanol bath. To minimize the number of freeze/thaw cycles, aliquot the extract into separate tubes and store at -80°C. The COLOR DE LYS® Substrate, when diluted in Assay Buffer, may precipitate after freezing and thawing. It is best, therefore, to dilute only the amount needed to perform the assays of that day.

**Long Term Storage** -80°C

**Shipping** Dry Ice

**Regulatory Status** RUO - Research Use Only

## Product Details

**Alternative Name** Histone deacetylase colorimetric assay kit

**Application** Activity assay, Colorimetric detection, HTS

**Compatibility** This product is compatible with the [Absorbance 96 Plate Reader](#).

## Contents

**Nuclear Extracts from HeLa Cells** (human cervical cancer cell line) (Prod. No. BML-KI137)  
(500 µl)

Storage: avoid freeze/thaw cycles!

**COLOR DE LYS® Substrate** (Prod. No. BML-KI138)  
(50 µl; 50mM in DMSO)

Storage: -80°C

**COLOR DE LYS® Developer Concentrate** (20x) (Prod. No. BML-KI139)

(100 µl; 20x stock solution, dilute in HDAC assay buffer before use)

Storage: -80°C

**Trichostatin A (HDAC Inhibitor)** (Prod. No. BML-GR309-9090)

(100 µl; 0.2mM in DMSO)

Storage: -80°C

**COLOR DE LYS® Deacetylated Standard** (Prod. No. BML-KI141)

(30 µl; 10mM in DMSO)

Storage: -80°C

**HDAC Assay Buffer** (Prod. No. BML-KI143)

(50mM TRIS-HCl, pH 8.0, 137mM NaCl, 2.7 mM KCl, 1mM MgCl<sub>2</sub>)

(20 ml)

Storage: -20°C

**1/2 volume microplate** (Prod. No. BML-KI101)

Storage: Ambient

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