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# Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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## Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

## Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand


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# Data Sheet

## *Fluorogenic MMP12 Assay Kit*

**Catalog #78017**
**Size: 96 reactions**

**BACKGROUND:** MMP12 (matrix metalloproteinase 12) is a member of the matrix metalloproteinase (MMP) family involved in the degradation of the extracellular matrix. MMP12 is also associated with the regulation of cytokines and chemokines, suggesting a role for MMP12 in inflammation. MMP12 is also essential for arterial stiffening in mice and is a highly prognostic marker of arterial stiffness, making MMP12 a therapeutic target for cardiovascular disease.

**DESCRIPTION:** The *Fluorogenic MMP12 Assay Kit* is designed to measure MMP12 activity for screening and profiling applications, in a homogeneous assay with no time-consuming washing steps. The kit comes in a convenient 96-well format, with purified MMP12 enzyme, fluorogenic substrate, and MMP assay buffer for 100 enzyme reactions.

**COMPONENTS:**

| Catalog # | Component | Amount | Storage | |
|---|---|---|---|---|
| | MMP12 | 3 µg | -80°C | ***Avoid freeze/ thaw cycles!*** |
| 79919 | MMP Substrate (1 mM) | 10 µl | -80°C | |
| 79917 | 1X MMP Assay Buffer 1 | 25 ml | -20°C | |
| 79685 | Black, low binding black microtiter plate | 1 | Room Temperature | |

**APPLICATIONS:** Great for studying enzyme kinetics and HTS applications.

**STABILITY:** At least one year from date of receipt when stored as directed.

**REFERENCE(S):**

1. Morris, D.G., *et al.* 2003. "Loss of integrin αvβ6-mediated TGF-β activation causes MMP12-dependent emphysema." *Nature* **422(6928):** 169-173.
2. Hunninghake, G.M., *et al.* 2009. "MMP12, lung function, and COPD in high-risk populations." *New England Journal of Medicine* **361(27):** 2599-2608.

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**ASSAY PROTOCOL:**

***All samples and controls should be tested in duplicate.***

1) Dilute 1 mM MMP substrate 1:100 in 1X assay buffer, to make a 10 µM solution. Dilute only enough as is required for the assay. Store remaining 1 mM substrate in aliquots at -80°C.

2) Prepare the substrate solution: N wells × (20 µl 1X assay buffer + 5 µl diluted (10 µM) MMP Substrate).

3) Add 25 µl of the substrate solution to each well (Final concentration of the MMP substrate in a 50 µl reaction is 1 µM).

| Component | Positive Control | Test Sample | Blank |
|---|---|---|---|
| Substrate solution | 25 µl | 25 µl | 25 µl |
| Test Inhibitor | – | 5 µl | – |
| Inhibitor buffer (usually 10% DMSO in assay buffer) | 5 µl | – | 5 µl |
| MMP12 (1.25 ng/µl) | 20 µl | 20 µl | – |
| 1X Assay Buffer | – | – | 20 µl |
| **Total** | **50 µl** | **50 µl** | **50 µl** |

4) Prepare the inhibitor solution.

   The final concentration of DMSO in the assay should not exceed 1%. If the inhibitor compound is dissolved in DMSO, make a 100-fold higher concentration of the compound than the highest concentration you want to test in DMSO. Then make a 10-fold dilution in 1X assay buffer (at this step the compound concentration is 10-fold higher than the final concentration in 10% DMSO). To determine an IC50 or to test lower concentrations of the compound, prepare a series of further dilutions in 1X assay buffer containing 10% DMSO (the final concentration of the DMSO will be 1% in all samples).

   If the inhibitor compound is dissolved in water, make a solution of the compound 10-fold higher than the final concentration in 1X assay buffer.

5) Add 5 µl inhibitor solution to each well designated “Test Sample.” Add 5 µl of the same solution without the inhibitor (inhibitor buffer, usually 10% DMSO in assay buffer) to “Blank” and “Positive Control” wells.

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6) Thaw MMP12 on ice. Upon first thaw, briefly spin tube containing enzyme to recover the full content of the tube. Aliquot MMP12 into single use aliquots. Store remaining undiluted enzyme in aliquots at -80°C. Note: MMP12 enzyme is sensitive to freeze/thaw cycles. Do not re-use diluted enzyme.

7) Dilute MMP12 in 1x assay buffer at 1.25 ng/µl (25 ng per reaction).

8) Add 20 µl diluted MMP12 enzyme solution to wells designated as "Positive Control" and "Test Sample." Add 20 µl 1X assay buffer to the "Blank" wells.

9) Incubate at room temperature for 2 hours. Measure the fluorescence intensity in a microtiter plate-reading fluorimeter capable of excitation at wavelength 328 nm ±10 nm and detection of emission at wavelength 393 nm ±10 nm. The fluorescence intensity can also be measured kinetically. "Blank" value is subtracted from all other values.

**MATERIALS OR INSTRUMENTS REQUIRED BUT NOT SUPPLIED:**
Fluorescent microplate reader capable of reading exc/em=328 nm/393 nm

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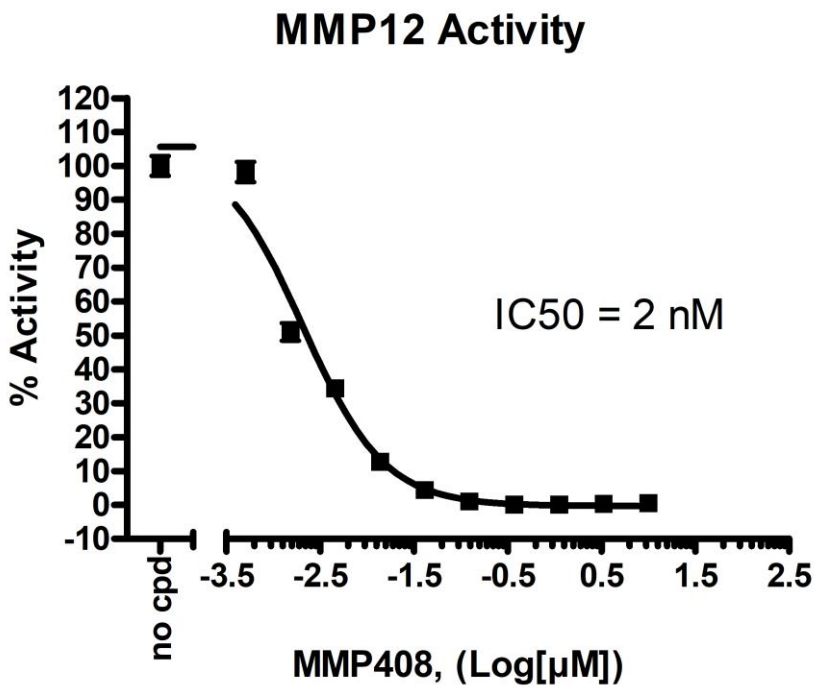
**EXAMPLE OF ASSAY RESULTS:**




Inhibition of MMP12 enzyme activity by MMP408 (Sigma #444291), measured using the *Fluorogenic MMP12 Assay Kit (BPS Bioscience #78017).* Fluorescence intensity was measured using a Tecan fluorescent microplate reader. *Data shown is lot-specific. For lot-specific information, please contact BPS Bioscience, Inc. at info@bpsbioscience.com*

**RELATED PRODUCTS**

| Product | Cat. # | Size |
|---|---|---|
| MMP1, His-Tag (Human) | 80214 | 20 µg |
| MMP2, His-Tag (Human) | 80213 | 20 µg |
| MMP3(K45E), His-Tag (Human) | 11346 | 100 µg |
| MMP8, His-Tag (Human) | 100552 | 100 µg |
| MMP9(Q279R), His-Tag (Human) | 80215 | 20 µg |
| Fluorogenic MMP3 (K45E) Assay Kit | 79907 | 384 rxns. |
| Fluorogenic MMP2 Assay Kit | 79918 | 96 rxns. |
| Fluorogenic MMP9 (Q279R) Assay Kit | 79915 | 96 rxns. |
| Fluorogenic MMP10Assay Kit | 79986 | 96 rxns. |
| Fluorogenic MMP13 Assay Kit | 79991 | 96 rxns. |
| Fluorogenic MMP14 Assay Kit | 79993 | 96 rxns. |