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Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Data Sheet

MAT2A Inhibitor Screening Assay Kit

Catalog: 71402
Size: 384 reactions

BACKGROUND: Methionine Adenosyltransferase 2A (MAT2A), also known as AdoMet Synthase 2, catalyzes the formation of S-adenosylmethionine from methionine and ATP. It is an important enzyme in cellular metabolism and is expressed in extrahepatic tissues. In liver, MAT2a expression is associated with growth, dedifferentiation, and cancer.

DESCRIPTION: The *MAT2a Inhibitor Screening Assay Kit* is designed to measure MAT2A activity for screening and profiling applications. The MAT2A assay kit comes in a convenient 384-well format, with purified recombinant MAT2A enzyme, L-Methionine, ATP, MAT2A assay buffer, and Colorimetric detection reagent for 384 enzyme reactions.

COMPONENTS:

Catalog #	Reagent	Amount	Storage	
71401	MAT2A	250 µg	-80°C	***Avoid multiple freeze/thaw cycles!***
	5x MAT2A Assay Buffer	3 ml	-20°C	
	ATP (750 µM)	1 ml	-20°C	
	L-Methionine (750 µM)	1 ml	-20°C	
74001	Colorimetric Detection Reagent*	2 x 10 ml	+4°C	
	Transparent 384-well plate	1	Room Temp.	

**Colorimetric Detection Reagent is used to measure the free phosphate from the MAT2A reaction. Any source of inorganic phosphate can interfere with the assay.*

MATERIALS OR INSTRUMENTS REQUIRED BUT NOT SUPPLIED:
UV/Vis spectrophotometer microplate reader capable of reading absorbance at 630 nm
Adjustable micropipettor and sterile tips
Rotating or rocker platform (optional)
Aluminum foil

APPLICATIONS: Great for studying enzyme kinetics and screening small molecular inhibitors for drug discovery and HTS applications.

STABILITY: Up to 6 months from date of receipt, when stored as recommended.
REFERENCE: Frau, M., *et al*., *Hepatology*, **56 (1),** 165-175 (2012)

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ASSAY PROTOCOL:
All samples and controls should be tested in duplicate.

1. Thaw **5x MAT2A assay buffer**, **ATP**, and **L-Methionine** on ice.

2. Prepare the master mixture (10 µl per well): N wells x (3 µl **5x MAT2A Assay Buffer** + 2.5 µl **ATP** + 2.5 µl **L-Methionine** + 2 µl water). Add 10 µl to every well. Store the remaining **ATP and L-Methionine** at -20°C in single use aliquots.

	Positive Control	Test Inhibitor	Blank
5x MAT2A Assay Buffer	3 µl	3 µl	3 µl
ATP	2.5 µl	2.5 µl	2.5 µl
L-Methionine	2.5 µl	2.5 µl	2.5 µl
Water	2 µl	2 µl	2 µl
Test Inhibitor	–	5 µl	–
5% DMSO in water (Inhibitor buffer)	5 µl	–	5 µl
1x MAT2A Assay Buffer	–	–	10 µl
MAT2A (60 ng/µl)	10 µl	10 µl	–
Total	25 µl	25 µl	25 µl

3. Add 5 µl of Inhibitor solution of each well labeled as “Test Inhibitor”. For the wells labeled “Positive Control" and “Blank”, add 5 µl of 5% DMSO in water (Inhibitor buffer). *Note: Do not dilute inhibitor in phosphate-based buffers, as phosphates will interfere with the Colorimetric Detection Reagent.*

4. Prepare **1x MAT2A Assay Buffer** by diluting **5x MAT2A Assay Buffer** with water. For 100 reactions, prepare 1 ml **1x MAT2A Assay Buffer** by mixing 200 µl of **5x MAT2A Assay Buffer** with 800 µl water. Dilute only enough buffer required for the assay. Store remaining **5x MAT2A Assay Buffer** at -20°C in single-use aliquots.

5. To the wells designated as "Blank", add 10 µl of **1x MAT2A Assay Buffer.**

6. Thaw **MAT2A** enzyme on ice. Upon first thaw, briefly spin tube containing enzyme to recover full contents of the tube. Calculate the amount of **MAT2A** required for the assay and dilute enzyme to ~60 ng/µl with **1x MAT2A Assay Buffer**. Aliquot remaining **MAT2A** enzyme into single-use aliquots. Store remaining undiluted enzyme in aliquots at -80°C. ***MAT2A*** *enzyme is sensitive to freeze/thaw cycles. Avoid multiple freeze/thaw cycles. Do not re-use thawed aliquots or diluted enzyme.*

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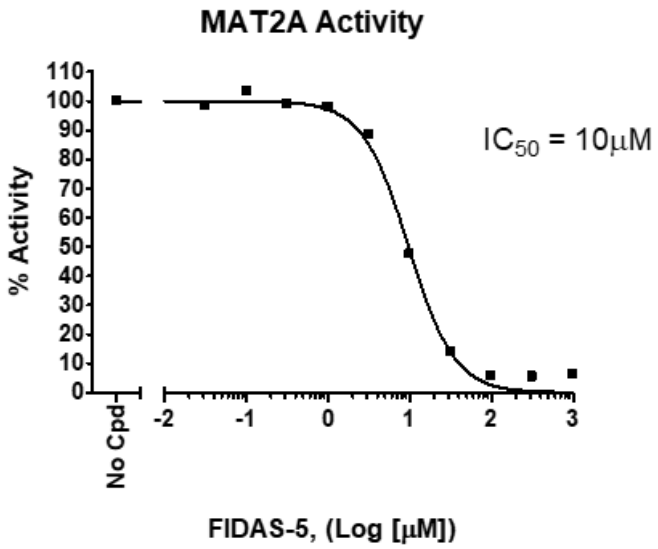
7. Initiate the reaction by adding 10 µl of diluted **MAT2A** enzyme to the wells designated "Positive Control" and "Test Inhibitor Control". **Incubate at room temperature for one hour**.

8. After the reaction, add 50 µl of **Colorimetric Detection Reagent** into each well. Cover the plate with aluminum foil and incubate the plate at room temperature for 15 minutes. During the 15 minute incubation, the plate can be placed on a rocker platform (optional).

9. Set the microplate reader and read Absorbance at 630 nm. Subtract "Blank" value from all other values.

Example of Assay Results:



MAT2A inhibition by FIDAS-5 (EMD Millipore, #504173), measured using the MAT2A Inhibitor Screening Assay Kit, BPS Bioscience Cat. 71402. The compound was pre-incubated with the MAT2a enzyme for 30 min before the reaction was initiated with the addition of master mix. The absorbance at 630 nm was measured using a Tecan Infinite M1000 microplate reader. *Data shown is lot-specific. For lot-specific information, please contact BPS Bioscience, Inc. at info@bpsbioscience.com*

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RELATED PRODUCTS:

Product Name	Catalog #	Size
MAT2A, His-tag	71401	100 µg
MTAP, GST-tag	50305	50 µg
S-Adenosylmethionine	52120	250 µl
S-adenosyl homocysteine, Fluorescein-labeled	52012	20 µl
Anti-Adenosyl Homocysteine Antibody	52030	1 ml
Adenosine Deaminase (ADA), His-tag	70016	100 µg
AHCY, GST-tag	50260	50 µg