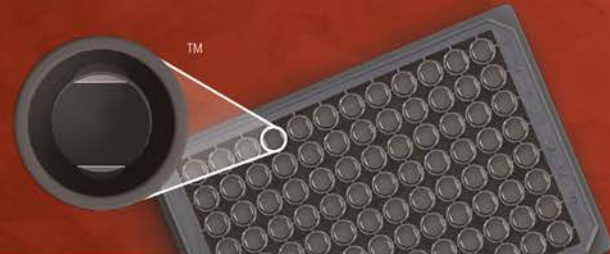


MSD® Rat B2M Assay Kit

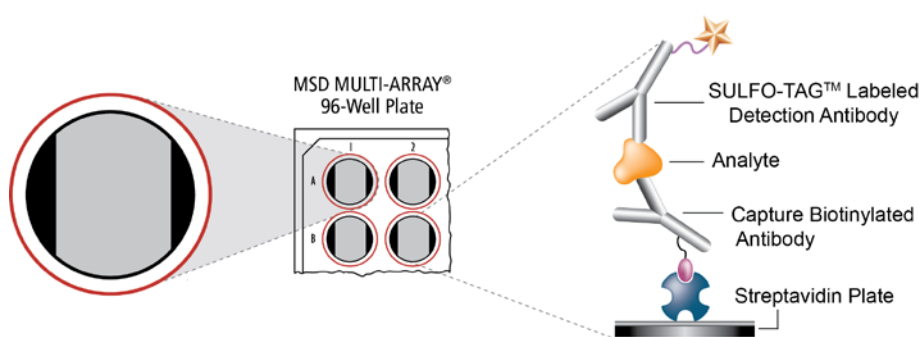
For quantitative determination in rat urine



Alzheimer's Disease
BioProcess
Cardiac
Cell Signaling
Clinical Immunology
Cytokines
Hypoxia
Immunogenicity
Inflammation
Metabolic
Oncology
Toxicology
Vascular

Catalog Numbers

Rat B2M Assay Kit	
Kit size	
1 plate	K153KAC-1
5 plates	K153KAC-2
25 plates	K153KAC-4



Beta-2-Microglobulin (B2M) is an 11.8 kDa secreted and ubiquitously expressed protein that is involved in the presentation of peptide antigens to the immune system. Mature rat B2M is a 99-amino acid polypeptide containing an IgC beta2m region (24-116) corresponding to class 1 major histocompatibility complex (MHC). B2M is normally filtered in the kidneys through the glomerulus, reabsorbed, and metabolized in the cells of the proximal tubules.¹ Under normal conditions, trace amounts of B2M are excreted in the urine. The levels of B2M rise in the event of kidney toxicity associated with glomerular injury and/or renal tubular damage.²⁻⁴ Elevated serum B2M levels have also been reported in conditions involving reduced Glomerular Filtration Rate (GFR) and are reportedly an indicator of allograft rejection in patients undergoing kidney transplant.^{5,6}

The MSD Rat B2M Assay is available on 96-well Streptavidin coated plates. The performance of this kit meets the levels of consistency and robustness as determined by methods based on the principles outlined in "Fit-for-Purpose Method Development and Validation for Successful Biomarker Measurement" by Lee, J.W. et al.⁷

This datasheet outlines the performance of the MSD Rat B2M Assay. Presented below are representative data from the assay qualification. The actual kit lot-specific standard curve and measured limits of quantification can be found in the Certificate of Analysis (C of A) supplied with the kit. A copy of the lot-specific C of A can be downloaded from www.mesoscale.com.

Ordering information

MSD Customer Service
Phone: 1-301-947-2085
Fax: 1-301-990-2776
Email: CustomerService@mesoscale.com

Assay Sensitivity

	B2M (ng/mL)
LLOD	0.00500
LLOQ	0.0488
ULOQ	20.0

The lower limit of detection (LLOD) is measured as the concentration at 2.5 standard deviations over the background.

The lower limit of quantification (LLOQ) is determined as the lowest concentration where the %CV of the calculated concentration is less than 20% and the percent recovery of the standard is between 80% and 120%.

The upper limit of quantification (ULOQ) is determined as the highest concentration where the %CV of the calculated concentration is less than 20% and the percent recovery of the standard is between 80% and 120%.

Company Address

MESO SCALE DISCOVERY®
A division of
Meso Scale Diagnostics, LLC.
9238 Gaither Road
Gaithersburg, MD 20877 USA

www.mesoscale.com®

MSD Advantage

- **Multiplexing:** Multiple analytes can be measured in one well using typical sample volumes of 25 µL or less without compromising speed or performance
- **Large dynamic range:** Linear range of up to five logs enables the measurement of native levels of biomarkers in normal and diseased samples without multiple dilutions
- **Minimal background:** The stimulation mechanism (electricity) is decoupled from the signal (light)
- **Simple protocols:** Only labels near the electrode surface are detected, enabling no-wash assays
- **Flexibility:** Labels are stable, non-radioactive, and conveniently conjugated to biological molecules
- **High sensitivity and precision:** Multiple excitation cycles of each label enhance light levels and improve sensitivity

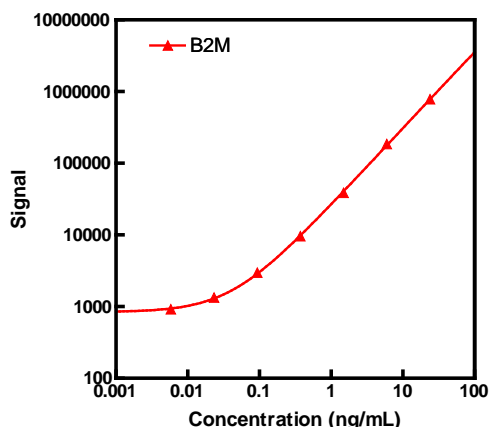
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procedures.

For a complete list of products, please visit our website at www.mesoscale.com.

MSD Toxicology Assays

Typical Standard Curve

The following standard curve is an example of the wide dynamic range of the Rat B2M Assay.



B2M		
Conc. (ng/mL)	Average Signal	%CV
0	759	4.7
0.00583	915	4.8
0.0233	1338	3.8
0.0934	2973	5.2
0.373	9605	2.3
1.49	38714	4.6
5.98	185089	3.6
23.9	780993	2.5

Spike Recovery

Normal rat urine was spiked with the calibrator at multiple levels throughout the range of the assay. The samples were diluted 1000-fold and then spiked with calibrator at the levels indicated in the table below.

% Recovery = measured / expected x 100%

B2M				
Sample	Spike Conc. (ng/mL)	Measured Conc. (ng/mL)	Measured Conc. %CV	% Recovery
Urine	0	0.877	4.5	
	0.488	1.34	3.3	98
	1.95	2.89	2.9	102
	7.81	9.55	3.0	110

Linearity

To assess linearity, urine samples were diluted 500-fold, 1000-fold, 2000-fold, and 4000-fold prior to testing. The diluted samples were prepared as follows:

- 25-fold dilution: 10 μ L of sample in 240 μ L of 1X PBS
- 20-fold dilution: 10 μ L of the 25-fold diluted sample in 190 μ L of Diluent 5 (500-fold dilution)
- 2-fold dilution: 100 μ L of 500-fold diluted sample in 100 μ L of Diluent 5 (1000-fold dilution)
- Repeat 2-fold serial dilutions 2 additional times to generate 2000- and 4000-fold dilutions.

The concentrations shown below have been corrected for dilution (concentration = measured x dilution factor). Percent recovery is calculated as the measured concentration divided by the concentration measured from the previous dilution (expected).

% Recovery = (measured x dilution factor) / expected x 100

B2M				
Sample	Fold Dilution	Conc. (ng/mL)	Conc. %CV	% Recovery
Urine	500	963	2.3	
	1000	890	2.6	92
	2000	867	2.6	97
	4000	842	4.1	97

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MSD Toxicology Assays

Precision

Control samples of high, mid, and low levels of were measured on each plate. Controls were made as follows:

High control: 275-fold diluted normal rat urine in Diluent 5

Mid control: High control samples diluted 4-fold in Diluent 5

Low control: Mid control samples diluted 4-fold in Diluent 5

All three controls were run neat.

The controls were run in quadruplicate on each of 3 plates run across multiple days (n>3). Average intra-plate %CV is the average %CV of the control replicates within an individual plate. Inter-plate %CV is the variability of controls across 9 plates over 3 days. Inter-lot %CV is the variability of controls across 2 kit lots.

	Control	Plates	Average Conc. (ng/mL)	Average Intra-plate %CV	Inter-plate %CV	Inter-lot %CV
B2M	High	16	4.61	3.1	3.9	4.3
	Mid	16	0.981	2.6	4.3	4.3
	Low	16	0.245	3.3	6.5	5.5

Samples

Urine samples collected from normal Sprague-Dawley rats were tested at 1000-fold dilution in the Rat B2M Assay. Shown below are the median and range of concentrations for each sample set. Concentrations have been corrected for sample dilution.

Sample	Statistic	B2M
Urine (male rat)	Median (ng/mL)	1004
	Range (ng/mL)	606-2036
	N	20
Urine (female rat)	Median (ng/mL)	365
	Range (ng/mL)	155-953
	N	20

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References using MSD's platform for the measurement of toxicology biomarkers

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