Vanillylmandelic Acid (VMA) Assay Kit
Research Use Only (RUO)

【Product Name】

Vanillylmandelic Acid (VMA) Assay Kit

【Package Size】

Vanillylmandelic Acid (VMA) Assay Kit (35 mL)

Reagent R1	28 mL
Reagent R2	7 mL
Calibrators	6 × 1 mL
Quality Controls	2 × 3 mL

【Intended Use】

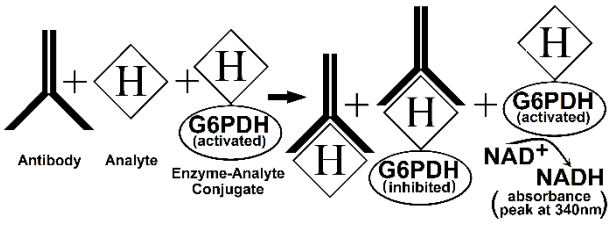
Vanillylmandelic Acid (VMA) Assay is a quantitative homogeneous enzyme immunoassay for measuring VMA in human urine. VMA is an end-stage metabolite of the catecholamines: epinephrine, and norepinephrine. Catecholamines are secreted by chromaffin cells of the adrenal medulla and the postganglionic fibers of the sympathetic nervous system.

Urinary VMA is elevated in patients with catecholamine secreting tumors including pheochromocytoma and neuroblastoma.

VMA levels in urine are also related to adrenal medulla hyperplasia (AMH). Research shows that medulla hyperplasia, hypertension, nocturnal hypoxemia and congestive heart failure may lead to elevated Vanillylmandelic acid in patients' urine.

【Assay Principle】

SJK Global VMA assay is a competitive homogeneous enzyme immunoassay, where antibody-bound and free analytes are not separated. The result is based on the competition for specific antibody binding sites between the free VMA compound in the sample and VMA conjugated to Glucose-6-Phosphate Dehydrogenase (G6PDH) enzyme. The more free VMA is present in a sample, the less antibody is available to bind enzyme-VMA conjugate, which speeds up conversion of NAD+ to NADH. Change in NADH concentration is detected by measuring absorbance at 340 nm.



【Reagent Composition】

Antibody/ Substrate Reagent 1 (R1): Contains rabbit polyclonal anti vanillylmandelic acid antibody, glucose-6-phosphate (G6P), nicotinamide adenine dinucleotide (NAD), stabilizers, sodium azide (0.09%) as preservative.

Enzyme-hapten Conjugate Reagent 2 (R2): Contains VMA labeled glucose-6-phosphate dehydrogenase (G6PDH) in buffer with sodium azide (0.09%) as preservative.

Calibrators/ Controls: Contain labeled concentration of VMA in buffer with sodium azide (0.09%) as preservative.

【Storage Condition and Term of Validity】

VMA Assay Kits should be stored in the dark at 2-8 °C. DO NOT FREEZE. Properly stored kits are valid until expiration date on the label. Properly stored open reagents, calibrators and controls are valid for 30 days after opening.

Do not mix kit components of different lots.

【Analyzers Application】

This product can be applied to run on chemistry analyzers including: Hitachi (7180, 7600); Beckman (AU640, AU5400, AU5800; Mindray (BS-200, BS-480); Roche (P800, Cobas701); Bayer-Siemens (ADVIA); Siemens Dimension RXL; and MEDICA EasyRA.

【Specimen Collection and Handling】

1. Sample is collected as urine sample.
2. Endogenous metabolites in urine samples will not statistically affect the test at the following concentrations:

Urine bilirubin	≤50 mg/L
Urine hemoglobin	≤1000 mg/L
Urinary albumin	≤100 g/L
Urinary ascorbic acid	≤176 mg/dL

3. Urine Samples are stable for 4 days at 2-8°C, and for 60 days at -20°C. Freeze and thaw may be performed up to three times. Frozen samples may be thawed at room temperature or in a 37°C water bath. Mix thawed samples well to ensure homogenous sample before testing.

【Assay Preparation】

VMA Assay kit is provided as ready to use; no special preparation is required.

【Assay Requirements】

Essential parameters (Beckman Coulter AU)

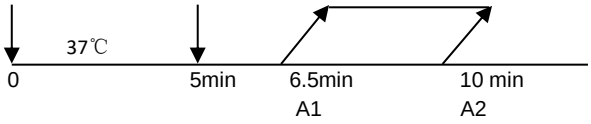
Method:	Terminal (End)	Sample Vol.:	10 µL
Measuring Point	12-24	Pre-Dilution Rate	1
Reagent 1 (R1):	200 µL	Reagent 2 (R2):	50 µL
Reaction Time:	10 Min	Main Wavelength:	340 nm
Temperature:	37℃	Sub Wavelength:	405 nm
Reaction Direction/ Indication:		Positive/Increasing	

Note: SJK will provide application parameters for specific chemistry analyzer per customer request.

Measurement:

R1: 200 µL R2: 50 µL Main wavelength: 340 nm

Sample: 10 µL Sub Wavelength: 405 nm



1. Calibration Procedure

Refer to Instructions for Use of Calibrators. Multi-point-calibration curve should be calculated using a non-linear model such as Polygonal or Line graph.

2. Quality Control Procedure

Refer to the Instruction for Use of Quality control. Daily quality control verification is recommended. General multi-point quality control model may be adopted to confirm calibration and to ensure the quality control results are within the acceptable range.

3. Results Calculation

Chemistry analyzers automatically calculate results. No additional data processing work is required.

【Reference Range】

24 hr Urine VMA reference range is ≤ 24 mg/24h.

Urine VMA was measured in 160 healthy donors without any VMA related diseases. Statistical method was used to calculate the mean ($\bar{x}$) and standard deviation (SD). Value of 95% one-sided confidence interval ($\bar{x}$+1.645*SD) resulted in the VMA range of ≤ 24 mg/24h. This result is for reference use only. Testing laboratories are encouraged to establish their own reference ranges.

【Further Explanation of Assay Result】

Assay result only reflects the specific sample VMA concentrations. Further testing of other markers may be required to assist for a clinical conclusion. Retesting, testing of the sample dilutions, or testing with a different methodology may be used to verify the result.

【Limitations】

For samples over the linear range, dilute the sample with 0.9% sodium chloride solution and retest.

【Product Properties】

Appearance

1. R1 and R2 are both transparent clear to light yellow liquid. Calibrator and quality controls are both transparent liquid.
2. Blank Absorbance ≤ 2.000 (1 cm; main wavelength is 340 nm and sub wavelength is 405 nm; 37)
3. At VMA concentration of 5.0 mg/L, the assay sensitivity range is [0.01-0.30]
4. Linear range of VMA assay is [2.0-200.0] mg/L, and r ≥ 0.990. For range of [2.0-10.0] mg/L the absolute deviation is ± 1.0 mg/L. For range of [10.0-200.0] mg/L the absolute deviation is ± 10.0%.
5. Precision:
   Intra lot variation CV ≤ 10.0%, with range R ≤ 10.0 %
6. Accuracy:
   Relative bias R ≤ 10.0 %
7. Calibrator Accuracy:
   Relative bias ≤ 10.0%; homogeneity CV ≤ 10.0%
8. Quality Control Accuracy :
   Relative bias ≤ 10.0%; homogeneity CV ≤ 10.0%

【Caution】

1. Turbid reagent should not be used and should be discarded.
2. All the test materials and devices should be well maintained and cleaned to avoid any contamination.
3. Proper personal protective equipment should be worn when working with laboratory test reagents. Care should be taken to avoid accidental ingestion, or contact with skin or mucosa. If the product contacted in the eyes, month, or skin, please rinse with plenty of water and seek medical attention if necessary.
4. Any result fails to meet the calibration and quality control is not acceptable
5. This product is for research use only (RUO).

【References】

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