

ROJAN[®] MDMA

One Step Ecstasy Test Strip (Urine) Package Insert

REF: R-1107

English

A rapid, one step test for the qualitative detection of Methylenedioxymethamphetamine (MDMA) in human urine. For healthcare professionals including professionals at point of care sites. For professional *in vitro* diagnostic use only.

INTENDED USE

The MDMA One Step Ecstasy Test Strip is a lateral flow chromatographic immunoassay for the qualitative detection of Methylenedioxymethamphetamine (MDMA) in human urine at a cut-off concentration of 500 ng/mL.

This assay provides only a preliminary analytical test result. A more specific alternate chemical method must be used in order to obtain a confirmed analytical result. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method. Clinical consideration and professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are used.

SUMMARY

Methylenedioxymethamphetamine (ecstasy) is a designer drug first synthesized in 1914 by a German drug company for the treatment of obesity.¹ Those who take the drug frequently report adverse effects, such as increased muscle tension and sweating. MDMA is not clearly a stimulant, although it has, in common with amphetamine drugs, a capacity to increase blood pressure and heart rate. MDMA does produce some perceptual changes in the form of increased sensitivity to light, difficulty in focusing, and blurred vision in some users. Its mechanism of action is thought to be via release of the neurotransmitter serotonin. MDMA may also release dopamine, although the general opinion is that this is a secondary effect of the drug (Nichols and Oberlander, 1990). The most pervasive effect of MDMA, occurring in virtually all people who took a reasonable dose of the drug, was to produce a clenching of the jaws. Methylenedioxymethamphetamine Test Strip yields a positive result when the Methylenedioxymethamphetamine in urine exceeds 500 ng/mL.

PRINCIPLE

The MDMA One Step Ecstasy Test Strip is an immunoassay based on the principle of competitive binding. Drugs which may be present in the urine specimen compete against the drug conjugate for binding sites on the antibody.

During testing, a urine specimen migrates upward by capillary action. Methylenedioxymethamphetamine, if present in the urine specimen below 500 ng/mL, will not saturate the binding sites of antibody-coated particles in the test strip. The antibody-coated particles will then be captured by immobilized Methylenedioxymethamphetamine conjugate and a visible colored line will show up in the test line region. The colored line will not form in the test line region if the Methylenedioxymethamphetamine level exceeds 500 ng/mL because it will saturate all the binding sites of anti-Methylenedioxymethamphetamine antibodies. A drug-positive urine specimen will not generate a colored line in the test line region because

of drug competition, while a drug-negative urine specimen or a specimen containing a drug concentration less than the cut-off will generate a line in the test line region.

To serve as a procedural control, a colored line will always appear at the control line region, indicating that proper volume of specimen has been added and membrane wicking has occurred.

REAGENTS

The test strip contains mouse monoclonal anti Methylenedioxymethamphetamine antibody-coupled particles and Methylenedioxymethamphetamine-protein conjugate. A goat antibody is employed in the control line system.

PRECAUTIONS

- For healthcare professionals including professionals at point of care sites.
- For professional *in vitro* diagnostic use only. Do not use after the expiration date.
- The test strip should remain in the sealed pouch until use.
- All specimens should be considered potentially hazardous and handled in the same manner as an infectious agent.
- The used test strip should be discarded according to federal, state and local regulations.

STORAGE AND STABILITY

Store as packaged in the sealed pouch at 2-30°C. The test strip is stable through the expiration date printed on the sealed pouch. The test strip must remain in the sealed pouch until use. **DO NOT FREEZE.** Do not use beyond the expiration date.

SPECIMEN COLLECTION AND PREPARATION

Urine Assay

The urine specimen must be collected in a clean and dry container. Urine collected at any time of the day may be used. Urine specimens exhibiting visible particles should be centrifuged, filtered, or allowed to settle to obtain a clear specimen for testing.

Specimen Storage

Urine specimens may be stored at 2-8°C for up to 48 hours prior to assay. For long-term storage, specimens may be frozen and stored below -20°C. Frozen specimens should be thawed and mixed before testing.

PROCEDURE

Materials Provided

- Test strips
- Package insert

Materials Required But Not Provided

- Specimen collection container
- Timer
- External controls

DIRECTIONS FOR USE

Allow the test strip, urine specimen, and/or controls to reach room temperature (15-30°C) prior to testing.

1. Bring the pouch to room temperature before opening it. Remove the test strip from the sealed pouch and use it as soon as possible.
2. With arrows pointing toward the urine specimen, immerse the test strip vertically in the urine specimen for at least 10 to 15 seconds. Do not pass the maximum line (MAX) on the test strip when immersing the strip. See the illustration below.
3. Place the test strip on a non-absorbent flat surface, start the timer

and wait for the colored line(s) to appear. The result should be read at 5 minutes. Do not interpret the result after 10 minutes.

INTERPRETATION OF RESULTS

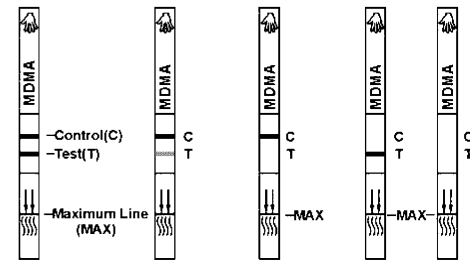
(Please refer to illustration below)

NEGATIVE: * **Two lines appear.** One colored line should be in the control region (C), and another colored line should be in the test region (T). This negative result indicates that the Methylenedioxymethamphetamine concentration is below the detectable level (500 ng/mL).

* **NOTE:** The shade of the color in the test line region (T) will vary, but it should be considered negative whenever there is even a faint line.

POSITIVE: **One colored line appears in the control region (C).** No line appears in the test region (T). This positive result indicates that the Methylenedioxymethamphetamine concentration exceeds the detectable level (500 ng/mL).

INVALID: **Control line fails to appear.** Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test using a new test strip. If the problem persists, discontinue using the lot immediately and contact your local distributor.



Negative

Positive

Invalid

QUALITY CONTROL

A procedural control is included in the test. A colored line appearing in the control region (C) is considered an internal procedural control. It confirms sufficient specimen volume, adequate membrane wicking and correct procedural technique.

Control standards are not supplied with this kit; however it is recommended that positive and negative controls be tested as good laboratory practice to confirm the test procedure and to verify proper test performance.

LIMITATIONS

1. The MDMA One Step Ecstasy Test Strip provides only a qualitative, preliminary analytical result. A secondary analytical method must be used to obtain a confirmed result. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method.^{1,2}
2. It is possible that technical or procedural errors, as well as other interfering substances in the urine specimen may cause erroneous results.

- Adulterants, such as bleach and/or alum, in urine specimens may produce erroneous results regardless of the analytical method used. If adulteration is suspected, the test should be repeated with another urine specimen.
- A Positive Result indicates presence of the drug or its metabolites but does not indicate level of intoxication, administration route or concentration in urine.
- A Negative Result may not necessarily indicate drug-free urine. Negative results can be obtained when drug is present but below the cutoff level of the test.
- Test does not distinguish between drugs of abuse and certain medications.
- A positive test result might be obtained from certain foods or food supplements.

PERFORMANCE CHARACTERISTICS

Accuracy

A side-by-side comparison was conducted using the MDMA One Step Ecstasy Test Strip and a leading commercially available MDMA rapid test. Testing was performed on 240 clinical specimens. Ten percent of the positive specimens employed were either at -25% or +25% level of the cut-off concentration of 500ng/ml Methyleneoxy methamphetamine. Presumptive positive results were confirmed by GC/MS. Approximately 10% of the negative clinical samples were also confirmed with GC/MS. The following results were tabulated:

Method	Other MDMA Rapid Test			Total Result
	Result	Positive	Negative	
MDMA One step test strip	Positive	90	1	91
	Negative	0	149	149
Total result		90	150	240
%Agreement		>99%	>99%	

MDMA One Step Ecstasy Test Strips were also tested with 93 MDMA positive and 147 MDMA negative urine samples in a clinical study. Nine of these positive urine samples in the +/- 25% cutoff range were derived from the concentrated MDMA clinical specimens; the rest were true clinical specimens. All positive samples used in this study were confirmed by GC/MS. Negative clinical samples were screened by a commercial MDMA rapid test kit. Approximately 10% of these negative samples were confirmed by GC/MS. The following results were tabulated.

		Negative	-25% cutoff to cutoff	Cutoff to +25% cutoff	>+25% of Cut-off
MDMA Strip	Positive	0	3	0	82
	Negative	147	2	5	0

Eighty (80) of these samples were also run using the MDMA One Step Ecstasy Test Strip by an untrained operator at a different site. Based on GC/MS data, the operator obtained a statistically similar Positive Agreement, Negative Agreement and Overall Agreement rate as the laboratory personnel.

Analytical Sensitivity

A drug-free urine pool was spiked with Methyleneoxy

methamphetamine at the following concentrations: 0ng/mL, 250 ng/mL, 375 ng/mL, 500 ng/mL, 625 ng/mL and 750 ng/mL. The result demonstrates >99% accuracy at 50% above and 50% below the cut-off concentration. The data are summarized below:

Methyleneoxy methamphetamine Conc. (ng/ml)	Percent of cut-off	n	Visual Result	
			Negative	Positive
0	0	30	30	0
250	-50%	30	30	0
375	-25%	30	26	4
500	Cut-off	30	17	13
625	25%	30	4	26
750	50%	30	0	30

Analytical Specificity

The following table lists compounds that are positively detected in urine by the MDMA One Step Ecstasy Test Strip at 5 minutes.

Compound	Concentration (ng/mL)
D,L-3,4-Methylenedioxyamphetamine HCl (MDMA)	500
3,4-Methylenedioxyamphetamine HCl (MDA)	3,000
3,4-Methylenedioxyethyl-amphetamine (MDE)	300

Precision

A study was conducted at 3 physician's offices by untrained operators using 3 different lots of product to demonstrate the within run, between run and between operator precision. An identical panel of coded specimens containing no Methyleneoxyamphetamine, 25% Methyleneoxyamphetamine above and below the cut-off and 50% Methyleneoxyamphetamine above and below the 500 ng/mL cut-off were provided to each site. The results are given below:

Methyleneoxy methamphetamine Conc. (ng/ml)	n	Site A		Site B		Site C	
		-	+	-	+	-	+
0	15	15	0	15	0	15	0
250	15	15	0	15	0	15	0
375	15	15	0	15	0	15	0
625	15	6	9	4	11	7	8
750	15	0	15	0	15	0	15

Effect of Urinary Specific Gravity

Fifteen (15) urine samples of normal, high, and low specific gravity ranges were spiked with 250 ng/mL and 750 ng/mL of Methyleneoxyamphetamine respectively. The MDMA One Step Ecstasy Test Strip was tested in duplicate using the fifteen neat and spiked urine samples. The results demonstrate that varying ranges of urinary specific gravity does not affect the test results.

Effect of the Urinary pH

The pH of an aliquoted negative urine pool was adjusted to a pH range of 5 to 9 in 1pH unit increments and spiked with Methyleneoxyamphetamine to 250 ng/mL and 750 ng/mL. The spiked, pH-adjusted urine was tested with the MDMA One Step Ecstasy Test Strip in duplicate. The results demonstrate that varying ranges of pH does not interfere with the performance of the test.

Cross-Reactivity

A study was conducted to determine the cross-reactivity of the test with compounds in either drug-free urine or Methyleneoxyamphetamine positive urine. The following

compounds show no cross-reactivity when tested with the MDMA One Step Ecstasy Test Strip at a concentration of 100 µg/mL.

Non Cross-Reacting Compounds

4-Acetamidophenol	β-Estradiol	Pentobarbital
Acetophenetidin	Estrone-3-sulfate	Perphenazine
N-Acetylprocainamide	Ethyl-p-aminobenzoate	Phencyclidine
Acetylsalicylic acid	Fenoprofen	Phenelzine
Aminopyrine	Furosemide	Phenobarbital
Amitypyline	Gentisic acid	Phentermine
Amobarbital	Hemoglobin	Trans-2-phenylcyclopropylamine hydrochloride
Amoxicillin	Hydralazine	L-Phenylephrine
Ampicillin	Hydrochlorothiazide	β-Phenylethylamine
L-Ascorbic acid	Hydrocodone	Phenylpropanolamine
D-Amphetamine	Hydrocortisone	Prednisolone
DL-Amphetamine sulfate	O-Hydroxyhippuric acid	Prednisone
L-Amphetamine	p-Hydroxyamphetamine	Procaine
Apomorphine	p-Hydroxy-methamphetamine	Promazine
Aspartame	3-Hydroxytyramine	Promethazine
Atropine	IBuprofen	DL-Propriolol
Benzilic acid	Imipramine	D-Propoxyphene
Benzoic acid	Iproniazid	D-Pseudoephedrine
Benzoylcegonine	(±) - Isoproterenol	

BIBLIOGRAPHY

- Baselt RC. Disposition of Toxic Drugs and Chemicals in Man. 2nd Ed. Biomedical Publ., Davis, CA. 1982; 488
- Hawks RL, CN Chiang. *Urine Testing for Drugs of Abuse*. National Institute for Drug Abuse (NIDA), Research Monograph 73, 1986
- A Handbook of Drug and Alcohol Abuse, Gail Winger, Third Edition, Oxford Press, 1992, page 146.

Index of Symbols	
Attention, see instructions for use	Tests per kit
For in vitro diagnostic use only	Use by
Store between 2-30°C	Lot Number
Manufacturer	Do not reuse

ISO, CE, GMP

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