

BAR

One Step Barbiturates Test Device Package Insert

A rapid, one step test for the qualitative detection of Barbiturates in human urine.

For healthcare professionals including professionals at point of care sites.

For in vitro diagnostic use only.

INTENDED USE

The *BAR* One Step Barbiturates Test Device is a lateral flow chromatographic immunoassay for the detection of Barbiturates in urine at a cut-off concentration of 300 ng/mL of secobarbital.

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

Barbiturates are central nervous system depressants. They are used therapeutically as sedatives, hypnotics, and anticonvulsants. Barbiturates are almost always taken orally as capsules or tablets. The effects resemble those of intoxication with alcohol. Chronic use of barbiturates leads to tolerance and physical dependence.

Short acting Barbiturates taken at 400 mg/day for 2-3 months produces a clinically significant degree of physical dependence. Withdrawal symptoms experienced during periods of drug abstinence can be severe enough to cause death.

Only a small amount (less than 5%) of most Barbiturates are excreted unaltered in the urine. The detection period for the Barbiturates in the urine is 4-7 days.¹

The *BAR* One Step Barbiturates Test Device is a rapid urine-screening test that can be performed without the use of an instrument. The test utilizes a monoclonal antibody to selectively detect elevated levels of Barbiturates in urine. The *BAR* One Step Barbiturates Test Device yields a positive result when the Barbiturates in urine exceed the cut-off level.

PRINCIPLE

The *BAR* One Step Barbiturates Test Device is an immunoassay based on the principle of competitive binding. Drugs that 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. Barbiturates, if present in the urine specimen below the cut-off level, will not saturate the binding sites of the antibody in the test device. The antibody coated particles will then be captured by immobilized Barbiturates 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 Barbiturates level exceeds the cut-off level because it will saturate all the binding sites of anti-Barbiturates 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 specimen containing a drug concentration lower than the cut-off concentration will generate a

line in the test line region because of the absence of drug competition.

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 device contains mouse monoclonal anti-Barbiturates antibody-coupled particles and Barbiturates-protein conjugate. A goat antibody is employed in the control line system.

PRECAUTIONS

- For healthcare professionals including professionals at point of care sites.
- For *in vitro* diagnostic use only. Do not use after the expiration date.
- The test device 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 test device 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 device is stable through the expiration date printed on the sealed pouch. The test device 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 testing. For long-term storage, specimens may be frozen and stored below -20°C. Frozen specimens should be thawed and mixed before testing.

MATERIALS

Materials Provided

- Test devices
- Disposable droppers
- Package insert

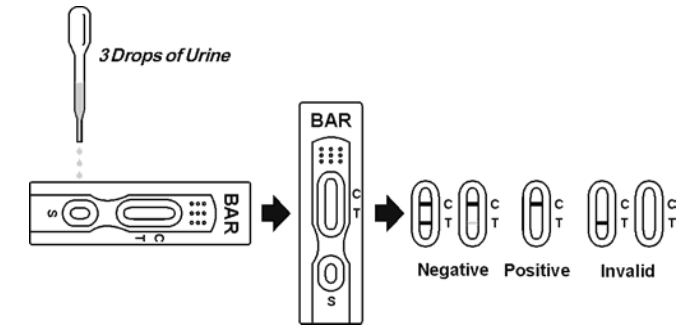
Materials Required But Not Provided

- Specimen collection container
- Timer
- External Controls

DIRECTIONS FOR USE

Allow the test device, 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 device from the sealed pouch and use it as soon as possible.
2. Place the test device on a clean and level surface. Hold the dropper vertically and transfer 3 full drops of urine (approx. 100µl) to the specimen well (S) of the test device, and then start the timer. Avoid trapping air bubbles in the specimen well (S). See the illustration below.
3. Wait for the red 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 above)

NEGATIVE: * **Two lines appear.** One red line should be in the control region (C), and another apparent red or pink line should be in the test region (T). This negative result indicates that the Barbiturates concentration is below the detectable cut-off level.

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

POSITIVE: **One red line appears in the control region (C).** No line appears in the test region (T). This positive result indicates that the Barbiturates concentration exceeds the detectable cut-off level.

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 with a new test device. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

QUALITY CONTROL

A procedural control is included in the test. A red line appearing in the control region (C) is considered an internal positive procedural control. It confirms sufficient specimen volume, adequate membrane wicking and correct procedural technique. This may be considered as the internal negative control.

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

LIMITATIONS

1. The *BAR* One Step Barbiturates Test Device 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) are the preferred confirmatory methods.^{2,3}
2. It is possible that technical or procedural errors, as well as other interfering substances in the urine specimen may cause erroneous results.
3. 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.
4. A Positive Result indicates presence of the drug or its metabolites but does not indicate level or intoxication, administration route or concentration in urine.
5. 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.
6. Test does not distinguish between drugs of abuse and certain medications.

PERFORMANCE CHARACTERISTICS

Accuracy

A side-by-side comparison was conducted using the BAR One Step Barbiturates Test Device and a commercially available BAR rapid test. Testing was performed on specimens previously collected from subjects presenting for Drug Screen Testing. Presumptive positive results were confirmed by GC/MS. The following results were tabulated:

Method	Other BAR Rapid Test		Total Results	
	Results	Positive		Negative
	BAR One Step Test Device	Positive		126
	Negative	0	165	165
Total Results		126	166	292
% Agreement with this commercial kit		>99%	99%	99%

When compared to GC/MS at the cut-off concentration of 300ng/mL, the following results were tabulated:

Method	GC/MS		Total Results	
	Results	Positive		Negative
	BAR One Step Test Device	Positive		122
	Negative	10	156	166
Total Results		132	160	292
% Agreement with GC/MS Analysis		92%	98%	95%

Eighty (80) of these samples were also run using the BAR One Step Barbiturates Test Device 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 Secobarbital at the following concentrations: 0 ng/mL, 150 ng/mL, 225 ng/mL, 300 ng/mL, 375 ng/mL and 450 ng/mL. The result demonstrates >99% accuracy at 50% above and 50% below the cut-off concentration. The data are summarized below:

Secobarbital Concentration (ng/mL)	Percent of Cutoff	n	Visual Result	
			Negative	Positive
0	0	30	30	0
150	-50%	30	30	0
225	-25%	30	20	10
300	Cutoff	30	13	17
375	+25%	30	8	22
450	+50%	30	0	30
600	100%	30	0	30

Specificity

The following table lists compounds that are positively detected in urine by the BAR One Step Barbiturates Test Device at 5 minutes.

Compound	Concentration (ng/mL)
Secobarbital	300
Amobarbital	300
Alphenol	150
Aprobarbital	200
Butabarbital	75
Butalbital	2,500
Butethal	100
Cyclopentobarbital	600
Pentobarbital	300
Phenobarbital	100

Precision

A study was conducted at 3 physicians' 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 Secobarbital, 25% Secobarbital above and below the cut-off and 50% Secobarbital above and below the 300 ng/mL cut-off was provided to each site. The following results were tabulated:

Secobarbital conc. (ng/mL)	n per site	Site A		Site B		Site C	
		-	+	-	+	-	+
0	15	15	0	15	0	15	0
150	15	13	2	15	0	15	0
225	15	2	13	8	7	6	9
375	15	2	13	1	14	2	13
450	15	0	15	0	15	0	15

Effect of Urinary Specific Gravity

Fifteen (15) urine samples with specific gravity ranges from 1.000 to 1.037 were spiked with 150 ng/mL and 450 ng/mL of Secobarbital respectively. The BAR One Step Barbiturates Test Device 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 1 pH unit increments and spiked with Secobarbital to 150 ng/mL and 450 ng/mL. The spiked, pH-adjusted urine was tested with the BAR One Step Barbiturates Test Device 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 Barbiturates positive urine. The following compounds show no cross-reactivity when tested with the BAR One Step Barbiturates Test Device at a concentration of 100 µg/mL.

Non Cross-Reacting Compounds

Acetaminophen	Estrone-3-sulfate	Oxolinic acid
Acetophenetidin	Ethyl-p-aminobenzoate	Oxycodone
N-Acetylprocainamide	Fenoprofen	Oxymetazoline
Acetylsalicylic acid	Furosemide	Papaverine
Aminopyrine	Gentisic acid	Penicillin-G
Amitriptyline	Hemoglobin	Pentazocine hydrochloride
Amoxicillin	Hydralazine	Perphenazine
Ampicillin	Hydrochlorothiazide	Phencyclidine
L-Ascorbic acid	Hydrocodone	Phenelzine
DL-Amphetamine sulfate	Hydrocortisone	Phentermine
Apomorphine	O-Hydroxyhippuric acid	Trans-2-phenylcyclopropylamine hydrochloride
Aspartame	p-Hydroxyamphetamine	L-Phenylephrine
Atropine	p-Hydroxy-methamphetamine	β-Phenylethylamine
Benzilic acid	3-Hydroxytyramine	Phenylpropanolamine
Benzoic acid	Ibuprofen	Prednisolone
Benzoyllecgonine	Imipramine	Prednisone
Benzphetamine	Iproniazid	Procaine
Bilirubin	(±) - Brompheniramine	Promazine
(±) - Brompheniramine	Caffeine	Promethazine
Caffeine	Cannabidiol	DL-Propranolol
Cannabidiol	Cannabinol	D-Propoxyphene
Cannabinol	Chloralhydrate	D-Pseudoephedrine
Chloralhydrate	Chloramphenicol	Levorphanol
Chloramphenicol		Quinacrine

Chlorothiazide	Loperamide	Quinidine
(±) - Chlorpheniramine	Maprotiline	Quinine
Chlorpromazine	MDE	Ranitidine
Chlorquine	Meperidine	Salicylic acid
Cholesterol	Meprobamate	Serotonin
Clomipramine	Methadone	Sulfamethazine
Clonidine	(L) Methamphetamine	Sulindac
Cocaethylene	Methoxyphenamine	Temazepam
Cocaine hydrochloride	(±) - 3,4-Methylenedioxyamphetamine hydrochloride	Tetracycline
Codeine	(±) - 3,4-Methylenedioxy-morphine-3-β-D glucuronide	Tetrahydrocortisone, 3-acetate
Cortisone	(-) Cotinine	Tetrahydrocortisone 3-(β-D-glucuronide)
Creatinine	Morphine Sulfate	Tetrahydrozoline
Deoxycorticosterone	Nalidixic acid	Thiamine
Dextromethorphan	Naloxone	Thioridazine
Diazepam	Naltrexone	DL-Tyrosine
Diclofenac	Naproxen	Tolbutamide
Diflunisal	Niacinamide	Triamterene
Digoxin	Nifedipine	Trifluoperazine
Diphenhydramine	Norcodein	Trimethoprim
Doxylamine	Norethindrone	Trimipramine
Ecgonine hydrochloride	D-Norpropoxyphene	Tryptamine
Ecgonine methylester	Noscapine	DL-Tryptophan
(-) -Ψ-Ephedrine	DL-Octopamine	Tyramine
[1R,2S] (-) Ephedrine	Oxalic acid	Uric acid
(L) - Epinephrine	Oxazepam	Verapamil
Erythromycin		Zomepirac
β-Estradiol		

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