

CRLstat™ Onsite Drug Cup

FOR IN VITRO DIAGNOSTIC USE

INTENDED USE

The CRLStat™ Onsite Drug Cup is a one-step immunoassay for the qualitative detection of multiple drugs and drug metabolites in human urine at the following cutoff concentrations:

Test	Calibrator	Cut-off (ng/ml)
AMP	Amphetamine	1000
BAR	Secobarbital	300
BZO	Oxazepam	300
COC	Benzoylcegonine	300
MDMA	3,4-methylenedioxymethamphetamine	500
MET	Methamphetamine	1000
MTD	Metadone	300
OPI2000	Morphine	2000
OPI300	Morphine	300
OXY	Oxycodone	100
PCP	Phencyclidine	25
TCA	Nortriptyline	1000
THC	11-nor- Δ^9 -THC-9-COOH	50

The configurations of the CRLStat™ Onsite Drug Cup consist of any combination of the drugs listed above. The CRLStat™ Onsite Drug Cup is used to obtain a visual, qualitative result and is intended for professional use only.

This assay provides only a preliminary result. Clinical consideration and professional judgment must be applied to any drug of abuse test result, particularly in evaluating a preliminary positive result. In order to obtain a confirmed analytical result, a more specific alternate chemical method is needed. Gas Chromatography/Mass Spectroscopy (GC/MS) is the preferred confirmation method.

SUMMARY AND EXPLANATION

Amphetamine/Methamphetamine, amphetamine, and metabolites are potent central nervous system stimulants. Acute higher doses induce euphoria, alertness, and sense of increased energy and power. More acute responses produce anxiety, paranoia, psychotic behavior, and cardiac dysrhythmias. Methamphetamine is excreted in urine as amphetamine and oxidized as deaminated and hydroxylated derivatives. However, methamphetamine is also excreted to some extent unchanged. Thus the presence of the parent compound in the urine indicates methamphetamine use.

Barbiturates are classified as central nervous system depressants. These products produce a state of intoxication that is similar to alcohol intoxication. Symptoms include slurred speech, loss of motor coordination and impaired judgment. Depending on the dose, frequency, and duration

of use, one can rapidly develop tolerance, physical dependence and psychological dependence on barbiturates. Barbiturates are taken orally, or by intravenous and intramuscular injections. They are excreted in urine as parent compound as well as metabolites.

Benzodiazepines are central nervous system (CNS) depressants commonly prescribed for the short-term treatment of anxiety and insomnia. In general, benzodiazepines act as hypnotics in high doses, as anxiolytics in moderate doses and as sedatives in low doses. The use of benzodiazepines can result in drowsiness and confusion. Psychological and physical dependence on benzodiazepines can develop if high doses of the drug are given over a prolonged period. Benzodiazepines are taken orally or by intramuscular or intravenous injection, and are extensively oxidized in the liver to metabolites. Parent compounds, as well as metabolites are excreted in the urine.

Cocaine is a potent central nervous system stimulant and a local anesthetic found in the leaves of the coca plant. The psychological effects induced by using cocaine are euphoria, confidence and sense of increased energy. These psychological effects are accompanied by increased heart rate, dilation of the pupils, fever, tremors and sweating. Cocaine is excreted in the urine primarily as benzoylecgonine in a short period of time. Benzoylecgonine has a biological half-life of 5 to 8 hours, which is much longer than that of cocaine (0.5 to 1.5 hour), and can be generally detected for 24 to 60 hours after cocaine use or exposure.

3,4-methylenedioxymethamphetamine is classified as both a stimulant and a hallucinogen. Like methamphetamine, adverse effects of 3,4-methylenedioxymethamphetamine use include jaw clenching, teeth grinding, dilated pupils, perspiring, anxiety, blurred vision, vomiting, and increased blood pressure and heart rate. Overdose of 3,4-methylenedioxymethamphetamine may cause heart failure or extreme heart stroke. 3,4-methylenedioxymethamphetamine is taken orally in tablets or capsules and excreted in urine as parent compound as well as metabolic.

Metadone is a synthetic analgesic drug originally used for the treatment of narcotic addiction. The psychological effects induced by using methadone are analgesia, sedation, and respiratory depression. Overdose of methadone may cause coma or even death. Methadone is taken orally or intravenously and is metabolized in the liver and has a biological half-life of 15-60 hours.

Opiates, such as heroin, morphine, and codeine, are central nervous system (CNS) depressants. The use of opiates at high doses produces euphoria and release from anxiety. Physical dependence is apparent in users and leads to depressed coordination, disrupted decision making, decreased respiration, hypothermia and coma. Heroin is quickly metabolized to morphine, morphine glucuronide and 6-acetylmorphine. Thus, the presence of morphine (or the metabolite, morphine glucuronide) in the urine indicates heroin, morphine, and/or codeine use.

Oxycodone is a semi-synthetic opioid with a structural similarity to codeine. It produces potent euphoria, analgesic and sedative effects, and has a dependence liability similar to morphine. Oxycodone is most often administered orally and is metabolized by demethylation to noroxycodone and oxymorphone followed by glucuronidation and excreted in urine. The window of detection for oxycodone in urine is expected to be similar to that of other opioids such as morphine.

Phencyclidine, commonly known as “angel dust” and “crystal cyclone”, is an arylcyclohexylamine that is originally used as an anesthetic agent and a veterinary tranquilizer. The drug is abused by oral or nasal ingestion, smoking, or intravenous injection. It produces hallucinations, lethargy, disorientation, loss of coordination, trance-like ecstatic states, a sense of euphoria and visual distortions. It is well absorbed following all routes of administration. Unchanged PCP is excreted in urine in moderate amounts (10% of the dose).

Tetrahydrocannabinol is generally accepted to be the principle active component in marijuana. When ingested or smoked, it produces euphoric effects. Abusers exhibit central nervous system effects, altered mood and sensory perceptions, loss of coordination, impaired short term memory, anxiety, paranoia, depression, confusion, hallucinations and increased heart rate. When marijuana is ingested, the drug is metabolized by the liver, the primary metabolite of marijuana excreted in the urine is 11-nor- Δ^9 -tetrahydrocannabinol-9-carboxylic acid. Therefore, the presence of detected cannabinoids, including the primary carboxyl metabolite, in the urine indicates marijuana/cannabis use.

Tricyclic antidepressants (TCAs) have been prescribed for depression and compulsive disorders. Because of the possibility of causing serious cardiac complications, TCAs can be lethal if misused at high doses. TCAs are taken orally or sometimes by injection. TCAs are metabolized in the liver. Both TCAs and their metabolites are excreted in urine mostly in the form of metabolites for up to ten days. The length of time following drug use of which a positive result may occur is dependent upon several factors, including the frequency and amount of drug, metabolic rate, excretion rate, drug half-life, and the drug user's age, weight, activity and diet.

Adulteration of urine samples may cause erroneous results in drugs of abuse test by either interfering with the drug screening test and/or destroying the drugs in the urine. Dilution of urine with water is probably the simplest urine adulteration method. Bleach, vinegar, Visine[®], sodium bicarbonate, sodium nitrite, Drano[®], soft drinks and hydrogen peroxide are the examples of adulterants used to adulterate the urine sample. It is important to insure the integrity of urine samples in drugs of abuse test.

TEST PRINCIPLE

The CRLStat[™] Onsite Drug Cup is based on the principle of competitive immunochemical reaction between a chemically labeled drug (drug-protein conjugate) and the drug or drug metabolites which may be present in

the urine sample for the limited antibody binding sites. The test contains a nitrocellulose membrane strip pre-coated with drug-protein conjugate in the test region and a pad containing colored antibody-colloidal gold conjugate. During the test, the urine sample is allowed to migrate upward and rehydrate the antibody-colloidal gold conjugate. The mixture then migrates along the membrane chromatographically by the capillary action to the immobilized drug-protein band on the test region. When drug is absent in the urine, the colored antibody-colloidal gold conjugate and immobilized drug-protein bind specifically to form a visible line in the test region as the antibody complexes with the drug-protein. When drug is present in the urine, it will compete with drug-protein for the limited antibody sites. The line on the test region will become less intense with increasing drug concentration. When a sufficient concentration of drug is present in the urine, it will fill the limited antibody binding sites. This will prevent attachment of the colored antibody-colloidal gold conjugate to the drug-protein on the test region. Therefore, the presence of the line on the test region indicates a negative result for the drug and the absence of the test line on the test region indicates a positive result for the drug.

A visible line generated by a different antigen/antibody reaction is also present at the control region of the test strip. This line should always appear, regardless of the presence of drugs or metabolites in the urine sample. This means that a negative urine sample will produce both test line and control line, and a positive urine sample will generate only control line. The presence of control line serves as a built-in control, which demonstrates that the test is performed properly.

REAGENTS & MATERIALS SUPPLIED

- 25 individually wrapped test devices. Each device consists of different test strips in a plastic test strip holder. The test strip contains a colloidal gold pad coated with antibody and rabbit antibody. It also contains a membrane coated with drug-bovine protein conjugate in the test band and goat anti-rabbit antibody in the control band adulterant pads when applicable.
- One instruction sheet

MATERIAL REQUIRED BUT NOT PROVIDED

- Timer
- Specimen collection container
- External positive and negative controls

WARNINGS AND PRECAUTIONS

- For professional in vitro diagnostic use only
- Urine specimens may be potentially infectious. Proper handling and disposal methods should be established.
- Avoid cross-contamination of urine samples by using a new specimen collection container for each urine sample.
- Test device should remain sealed until ready for use.

- Do not use the test kit after the expiration date.
- A positive test result does not always mean an individual has taken the drug illegally as the drug can be administered legally. Do not store and or expose reagent kits at temperature greater than 30°C. Do not freeze.

STORAGE

The CRLStat™ Onsite Drug Cup should be stored at 2-30°C (36-86°F) in the original sealed pouch. Do not freeze. Do not store and or expose reagent kits at temperature greater than 30°C.

SPECIMEN COLLECTION AND HANDLING

Fresh urine does not require any special handling or pretreatment. A fresh urine sample should be collected in the container provided. Alternately, a clean, dry plastic or glass container may be used for specimen collection. If the specimen will not be tested after the specimen collection, the specimen may be refrigerated at 2-8°C up to 2 days or frozen at -20°C for longer period of time. Specimens that have been refrigerated must be equilibrated to room temperature prior to testing. Specimens previously frozen must be thawed and mixed thoroughly prior to testing.

Note: Urine specimens and all materials coming in contact with them should be handled and disposed as if capable of transmitting infection. Avoid contact with skin by wearing gloves and proper laboratory attire.

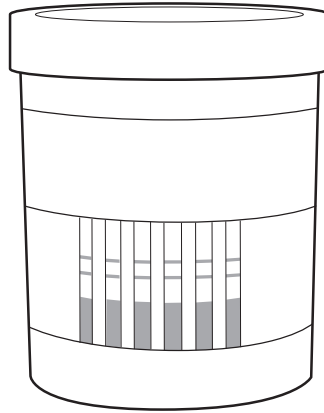
ASSAY PROCEDURE

Preparation

1. If specimen, control, or test devices have been stored at refrigerated temperatures, allow them to warm to room temperature before testing.
2. Do not open test device pouch until ready to perform the test.

Testing

1. Remove the test cup from the sealed pouch.
2. Issue the cup to the individual to be tested.
3. Collect the urine into the cup. Ensure the specimen is above the minimum level.
4. The cup must be returned immediately to the collector. Authorized personnel should remove the tear-off label.
5. Read results of drugs of abuse tests in 5 minutes. Do not interpret result after 10 minutes.



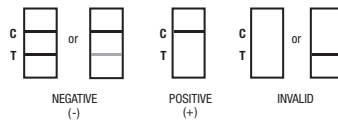
Drug Screen Cup

INTERPRETATION OF RESULTS

Negative (-): Two colored lines should be observed in the viewing window. The line in the test region is the drug probe line. The line in the control region is the control line, which is used to indicate proper performance of the device. The test line may have varying intensity either weaker or stronger in color than that of the control line.

Positive (+): Only one colored line appears in the control region. The complete absence of a test line with a clear white background indicates a positive result.

Invalid: No colored line appears in the control region. If the control line does not form, the test result is inconclusive and should be repeated.



QUALITY CONTROL

An internal procedural control is included in the test device. A line must form in the Control band region regardless of the presence or absence of drugs or metabolites. The presence of the line in the Control region indicates that the proper sample volume has been used and that the reagents are migrating properly. If the line in the Control region does not form, the test is considered invalid.

To ensure proper kit performance, it is recommended that the test devices be tested once a week with external controls. External controls are available from commercial sources. It is important to make sure that the control values are within established limits. If the values of external control do not fall within established limits, the test results are invalid. Additional controls may be tested according to guidelines or requirements of local, state, and/or federal regulations or accrediting organizations.

LIMITATIONS OF PROCEDURE

- The assay is designed for use with human urine only.
- A positive result with any of the tests indicates only the presence of a drug/metabolite and does not indicate or measure intoxication.
- There is a possibility that technical or procedural error as well other substances as factors not listed may interfere with the test and cause false results. See SPECIFICITY for lists of substances that will produce positive results, or that do not interfere with test performance.
- If adulteration is suspected, the test should be repeated with new sample.

PERFORMANCE CHARACTERISTICS

Accuracy

The accuracy of the CRLStat™ Onsite Drug Cup was evaluated in comparison to commercially available drug screen tests. Sixty (60) negative urine samples collected from presumed non-user volunteers were tested by both CRLStat™ Onsite Drug Cup and commercially available drug screen tests. Of these negative urine samples tested, all were found negatives by both methods. In a separate study, positive urine samples, obtained from clinical laboratories where the drug concentrations were determined by GC/MS, were tested by CRLStat™ Onsite Drug Cup and commercial drug screen tests. The results of accuracy study are presented below:

Drug Test		GC/MS (<-50% C/O)	GC/MS (-50% C/O to C/O)	GC/MS (C/O to +50% C/O)	GC/MS (>+50% C/O)	% Agreement with GC/MS
AMP	(+)	0	1	11	57	100
	(-)	15	8	0	0	95.8
BAR	(+)	0	1	4	83	96.7
	(-)	15	7	3	0	95.7
BZO	(+)	0	3	12	37	98.0
	(-)	18	17	1	0	92.1
COC	(+)	0	2	8	70	100
	(-)	15	6	0	0	91.3
MDMA	(+)	0	1	6	37	100
	(-)	24	6	0	0	96.8
MET	(+)	0	1	6	58	100
	(-)	20	7	0	0	96.4
MTD	(+)	0	0	6	65	98.6
	(-)	15	5	1	0	100
OPI300	(+)	0	1	6	77	100
	(-)	16	6	0	0	95.7
OPI2000	(+)	0	2	9	45	100
	(-)	15	6	0	0	91.3
OXY	(+)	0	1	6	47	100
	(-)	15	7	0	0	95.7

PCP	(+)	0	1	5	36	100
	(-)	15	3	0	0	94.7
TCA	(+)	0	1	12	9	100
	(-)	23	11	0	0	97.1
THC	(+)	0	4	24	32	100
	(-)	15	5	0	0	83.3

Precision

The precision of the CRLStat™ Onsite Drug Cup was evaluated by testing three lots of the test devices at four study sites with spiked drug sample solutions on three consecutive days. Sample concentrations were confirmed by GC/MS.

AMP (ng/ml)	0	500	750	1000	1250	1500
(+/-)	0/135	0/135	34/101	72/63	99/36	135/0
BAR (ng/ml)	0	150	225	300	375	450
(+/-)	0/135	0/135	31/104	68/67	101/34	135/0
BZO (ng/ml)	0	150	225	300	375	450
(+/-)	0/135	0/135	33/102	76/59	104/31	135/0
COC (ng/ml)	0	150	225	300	375	450
(+/-)	0/135	0/135	35/100	71/64	98/37	135/0
MDMA (ng/ml)	0	250	375	500	625	750
(+/-)	0/135	0/135	29/106	73/62	102/33	135/0
MET (ng/ml)	0	500	750	1000	1250	1500
(+/-)	0/135	0/135	32/103	71/64	98/37	135/0
MTD (ng/ml)	0	150	225	300	375	450
(+/-)	0/135	0/135	30/105	76/59	101/34	135/0
OPI300 (ng/ml)	0	150	225	300	375	450
(+/-)	0/135	0/135	33/102	69/66	96/39	135/0
OPI2000 (ng/ml)	0	1000	1500	2000	2500	3000
(+/-)	0/135	0/135	32/103	70/65	103/32	135/0
OXY (ng/ml)	0	50	75	100	125	150
(+/-)	0/135	0/135	30/105	73/62	102/33	135/0
PCP (ng/ml)	0	12.5	18.75	25	31.25	37.5
(+/-)	0/135	0/135	35/100	70/65	100/35	135/0
THC (ng/ml)	0	25	37.5	50	62.5	75
(+/-)	0/135	0/135	31/104	68/67	103/32	135/0
TCA (ng/ml)	0	500	750	1000	1250	1500
(+/-)	0/135	0/135	26/109	62/77	97/38	135/0

Specificity

The specificity for the CRLStat™ Onsite Drug Cup was determined by testing various drugs, drug metabolites, and other compounds that are likely to be present in urine. All compounds were prepared in drug-free normal human urine.

The following compounds produce positive results when tested at levels greater than the concentrations listed below.

Compound	Conc. (ng/ml)	Compound	Conc. (ng/ml)
Amphetamine			
d-Amphetamine	1,000	d-Methamphetamine	50,000
dl-Amphetamine	2,500	(+/-)3,4-MDMA	50,000
(+/-)3,4-MDA	1,250		
Barbiturates			
Secobarbital	300	Butobarbital	400
Allobarbital	600	Butalbital	300
Alphenal	200	Butethal	450
Amobarbital	1500	Pentobarbital	400
Aprobarbital	300	Phenobarbital	450
Barbital	1500		
Benzodiazepines			
Oxazepam	300	Flunitrazepam	300
Alprazolam	400	Flurazepam	300
Bromazepam	250	Lorazepam	500
Chlordiazepoxide	300	Medazepam	300
Clobazam	1000	Nitrazepam	250
Clonazepam	500	Nordiazepam	150
Clorazepate	150	Prazepam	500
Desalkylflurazepam	200	Temazepam	200
Diazepam	450	Triazolam	450
Estazolam	300		
Cocaine			
Benzoyllecgonine	300	Cocaine	300
Methamphetamine			
d-Methamphetamine	1000	(+/-)3,4-MDMA	3,000
d-Amphetamine	50,000	l-Methamphetamine	10,000
l-Amphetamine	>100,000	Ephedrine	>100,000
(+/-)3,4-MDEA	50,000	Mephentermine	75,000
(+/-)3,4-MDA	100,000		
MDMA			
(+/-)3,4-MDMA	500	(+/-)3,4-MDA	4,000
(+/-)3,4-MDEA	450		
Methadone			
(+/-) Methadone	300	Methadol	1,500

Compound	Conc. (ng/ml)	Compound	Conc. (ng/ml)
Opiates (300)			
Morphine	300	Hydrocodone	500
Codeine	300	Hydromorphone	500
Ethylmorphine	300	Morphine-3-glucuronide	300
Heroin	750	Nalorphine	5,000
Opiates (2000)			
Morphine	2,000	Hydrocodone	4,000
Codeine	2,000	Hydromorphone	5,000
Ethylmorphine	1,000	Morphine-3-glucuronide	2,500
Heroin	5,000	Nalorphine	5,000
Oxycodone			
Oxycodone	100	Morphine	>100,000
Hydrocodone	5000	Codeine	50,000
Hydromorphone	50,000	Heroin	>100,000
PCP			
Phencyclidine	25	Tenocyclidine	2,000
THC			
11-nor- Δ^9 -THC-9-COOH	50	Δ^9 -tetrahydrocannabinol	5,000
11-hydroxy- Δ^9 -THC	1,000	Cannabinol	10,000
Δ^8 -tetrahydrocannabinol	5,000	Cannabidiol	>100,000
Tricyclic Antidepressant			
Nortriptyline	1,000	Promazine	1,500
Nordoxepin	2,000	Desipramine	400
Trimipramine	2,000	Doxepin	3,000
Amitriptyline	1,500	Maprotiline	2,000

Interference

Two pools of drug-free urine were spiked with drug standards to 50% below and 50% above cutoff concentrations. The drug concentrations were confirmed by GC/MS. The following compounds were evaluated for potential positive and/or negative interference with the CRLStat™ Onsite Drug Cup. All compounds were dissolved in the spiked sample solutions and tested with CRLStat™ Onsite Drug Cup. An unaltered sample was used as a control. No positive interference or negative interference was found for the following compounds when tested at concentrations up to 100 ng/ml.

Acetaminophen	Ibuprofen
Acetone	(+/-)-Isoproterenol
Albumin	Ketamine
Ampicillin	Levorphanol
Ascorbic Acid	Lidocaine
Aspartame	(+)-Naproxen
Aspirin	Niacinamide
Atropine	Nicotine
Benzocaine	(+/-)-Norephedrine
Bilirubin	Oxalic Acid
Caffeine	Penicillin-G
Chloroquine	Pheniramine
(+)-Chlorpheniramine	Phenothiazine
(+/-)-Chlorpheniramine	I-Phenylephrine
Creatine	β-Phenylethylamine
Dexbrompheniramine	Procaine
Dextromethorphan	Quinidine
Diphenhydramine	Ranitidine
Dopamine	Riboflavin
(+/-)-Epinephrine	Sodium Chloride
Erythromycin	Sulindac
Ethanol	Theophylline
Furosemide	Tyramine
Glucose	4-Dimethylaminoantipyrine
Guaiaccol Glyceryl Ether	(1R,2S)-(-)-N-Methyl-Ephedrine
Hemoglobin	

Effect of Specimen pH

Drug sample solutions with 50% below and 50% above cutoff concentrations were adjusted to pH 4-9 and tested using CRLStat™ Onsite Drug Cup. An unaltered sample was used as a control. The results demonstrate that varying ranges of specimen pH do not interfere with the performance of the test.

Effect of Specimen Specific Gravity

Drug sample solutions with 50% below and 50% above cutoff concentrations were adjusted to specific gravity 1.003-1.04 and tested using CRLStat™ Onsite Drug Cup. An unaltered sample was used as a control. The results demonstrate that varying ranges of specimen specific gravity do not interfere with the performance of the test.

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