Instant Drug Test Card (IDTC)-Nicotine
Professional Use Package Insert

A rapid test for the qualitative detection of nicotine metabolite in human urine
For in vitro diagnostic use including point-of-care. (CLIA Exempt)

【INTENDED USE】

The IDTC-Nicotine Test Card is a lateral flow immunochromatographic assay for the qualitative detection of Nicotine metabolite (cotinine) in human urine with 3 cutoff levels (50, 200 and 500 ng/mL). The IDTC-Nicotine Test Card is intended for prescription professional, in-vitro diagnostic use in near-patient (POC), centralized testing locations and for testing in employment settings. The device can consist of any combination of the 3 assays in 1, 2 or 3-test formats.

The test provides a preliminary result only; preliminary positive results should be confirmed using an alternate chemical methodology (GC/MS, GC/MS/MS, LC/MS or LC/MS/MS) if the donor does not admit to nicotine use or anytime your test policies require.

【SUMMARY】

Cotinine is the predominant metabolite of nicotine. Cotinine is used as a biomarker for exposure to tobacco. In a 24-hour urine, approximately 5% of a nicotine dose is excreted as an unchanged drug with 10% as cotinine and 35% as hydroxy cotinine; the concentrations of other metabolites are believed to account for less than 5%.[1] In urine, values between 11 ng/mL and 30 ng/mL may be associated with light smoking or passive exposure, and levels in active smokers typically reach 500 ng/mL or more.[2] Nicotine is rapidly metabolized and has a short half-life, but cotinine is metabolized and eliminated at a much lower rate. Because of the resulting increase with time in the cotinine to nicotine ratio in the body, (including the brain), it is of interest to examine the effect of cotinine on nicotine-induced changes.

【PRINCIPLE】

The IDTC-Nicotine Test Card is a rapid chromatographic 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. Cotinine, the nicotine metabolite, if present in the urine specimen below 50 ng/mL, 200 ng/mL or 500 ng/mL will not saturate the binding sites of the antibody coated particles in the test. The antibody coated particles will then be captured by immobilized cotinine 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 cotinine level exceeds the cutoff levels of each test because it will saturate all the binding sites of anti-cotinine antibodies.

A drug-positive urine specimen will not generate a colored line in the test line region, 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 contains anti-cotinine antibody-coupled particles and cotinine-protein conjugate. A goat antibody is employed in the control line system.

【PRECAUTIONS】

- Do not use after the expiration date.
- The test 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 should be discarded according to local regulations.

【STORAGE AND STABILITY】

- Store as packaged in the sealed pouch or canister either at room temperature or refrigerated (2-30°C). The test is stable through the expiration date printed. **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 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

- Dip Cards
- Package insert

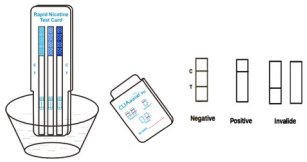
Materials Required But Not Provided

- Specimen collection container
- Timer

【DIRECTIONS FOR USE】

If refrigerated, allow the test, urine specimen, and/or controls to reach room temperature (15-30ºC) prior to testing.

1. Bring the pouch to room temperature before opening it. Remove the IDTC-Nicotine Test Card from the sealed pouch and use it within one hour.
2. With arrows pointing toward the urine specimen, **immerse the dip card vertically in the urine specimen for at least 20 seconds**. Place the dip card on a non-absorbent flat surface, start the timer and wait for the colored line(s) to appear. **Alternatively, the dip card can be left in the urine specimen throughout the testing process**.
3. Negative results can be interpreted as soon as they are apparent. Read positive results at 5 minutes. The results remain stable for 60 minutes.



【INTERPRETATION OF RESULTS】

(Please refer to the illustration above)

NEGATIVE: * Two lines appear. One colored line should be in the control line region (C), and another apparent colored line should be in the test line region (T). This negative result indicates that the cotinine concentration is below the detectable levels.

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

POSITIVE: One colored line appears in the control line region (C). No line appears in the test line region (T). This positive result indicates that the cotinine concentration exceeds the detectable level of the specific strip (50 ng/mL, 200 ng/mL or 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. If the problem persists, discontinue using the lot immediately and contact your local distributor.

【QUALITY CONTROL】

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

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

【LIMITATIONS】

1. The IDTC-Nicotine Test Card Drug Screen provides only a qualitative, preliminary analytical result. A secondary analytical method should be used to obtain a confirmed result if the donor doesn't admit to nicotine use or anytime your policies require. GC/MS, GC/MS/MS, LC/MS and LC/MS/MS are the preferred confirmation methods.
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 in urine specimens may produce erroneous results regardless of the analytical method used. If adulteration is suspected, the test should be repeated with another sample.
4. A positive result indicates presence of the drug or its metabolites but does not indicate level of intoxication, administration route or exact concentration in urine.
5. A negative result may not necessarily indicate cotinine-free urine. Negative results can be obtained when drug is present but below the cut-off levels of the tests.
6. Test does not distinguish between tobacco and nicotine-based smoking cessation products.

【EXPECTED VALUES】

This negative result indicates that the cotinine concentration is below the detectable level of each strip. Positive result means the concentration of cotinine is above the cutoff level of each strip (50 ng/mL, 200 ng/mL and 500 ng/mL).

【PERFORMANCE CHARACTERISTICS】

Accuracy

Blind testing was conducted using the IDTC-Nicotine Test Card Drug Screen and clinical specimens with confirmed levels of cotinine by GC/MS. The following results were tabulated:

Method		GC/MS		Accuracy
	Results	Positive	Negative	
COT 50 Rapid Test	Positive	40	0	>99%
	Negative	0	40	>99%
COT 200 Rapid Test	Positive	40	0	>99%
	Negative	0	40	>99%
COT 500 Rapid Test	Positive	37	0	>99%
	Negative	1	42	97.67%

Analytical Sensitivity

A drug-free urine pool was spiked with cotinine at the following concentrations: 25 ng/mL, 37.5 ng/mL, 50 ng/mL, 62.5 ng/mL and 75 ng/mL (50 ng/mL cutoff), 100 ng/mL, 150 ng/mL, 200 ng/mL, 250 ng/mL and 300 ng/mL (200 ng/mL cutoff), and 250 ng/mL, 375 ng/mL, 500 ng/mL, 625 ng/mL and 750 ng/mL (500 ng/mL cutoff). All assays were also tested with a drug-free urine pool. All assays demonstrated >99% correlation for the -50%, +50% and drug free solutions.

Analytical Specificity

The following table lists compounds that are positively detected in urine by the IDTC-Nicotine Test Card Urine Drug Screen at 5 minutes.

Compound	Concentration (ng/mL)	Reactivity (%)
COT 50		
Cotinine	50	100%
Trans-3´-hydroxycotinine	30	14.28%
(R,S)-Norcotine	2000	0.67%
S(-)-Nicotine	90,000	0.05%
COT 200		
Cotinine	200	100%
Trans-3´-hydroxycotinine	200	4%
(R,S)-Norcotine	30,000	0.2%
S(-)-Nicotine	>100,000	<0.2%
COT 500		
Cotinine	500	100%
Trans-3´-hydroxycotinine	500	3.33%
(R,S)-Norcotine	>100,000	<0.5%
S(-)-Nicotine	>100,000	<0.5%

Cross-Reactivity

A study was conducted to determine the cross-reactivity of the test with compounds in cotinine drug control solutions 50% below and 50% above their respective cutoff concentrations. The following compounds show no cross-reactivity when tested with the IDTC-Nicotine Test Card Urine Drug Screen at a concentration of 100 μg/mL.

Acetaminophen	Diphenhydramine	(+/-)-Norephedrine
Acetone	Dopamine	Oxalic acid
Albumin	(+/-)-Isoproterenol	Penicillin-G
Ampicillin	1R,2S(+)-Ephedrine	Pheniramine
Ascorbic acid	Erythromycin	Phenothiazine
Aspartame	Ethanol	L-Phenylephrine
Aspirin	Furosemide	B-Phenylethylamine
Atropine	Glucose	Procaine
Benzocaine	Guaiacol glyceryl ether	Quinidine
Bilirubin	Hemoglobin	Ranitidine
Caffeine	Ibuprofen	Riboflavin
Chloroquine	(+/-)-Isoproterenol	Sodium chloride
(+)-Chlorpheniramine	Ketamine	Sulindac
(+/-)-Chlorpheniramine	Levorphanol	Theophylline
Creatine	Lidocaine	Tyramine
Dexbrompheniramine	(1R,2S)-(-)-n-Methylephedrine	
Dextromethorphan	(+)-Naproxen	
4-Dimethylaminoantipyrine	Niacinamide	

【BIBLIOGRAPHY】

1. Baselt RC. Disposition of Toxic Drugs and Chemicals in Man. 10th Ed. Biomedical Publ., Seal Beach, CA. 2014
2. Moyer TP, Charlson JR, Enger RJ, et al: Simultaneous analysis of nicotine, nicotine metabolites, and tobacco alkaloids in serum or urine by tandem mass spectrometry, with clinically relevant metabolic profiles. Clin Chem 2002;48:1460-1471