Diagnostic Automation / Cortez Diagnostics, Inc.
IMMUNO DIAGNOSTICS

OneStep
K2 Urine
RapiDip™ InstaTest
RapiCard™ InstaTest

REF 121154-1
REF 121153-1

"For laboratory use only"

IVD See external Label 4-30°C 1 Test

Sensitivity	50 ng/ml

INTENDED USE

K2 Test is an immunochromatography based one step in vitro test. It is designed for qualitative determination of synthetic cannabis JWH-18 and JWH-73's major metabolites in human urine specimens at cut-off level of 50 ng/ml. This assay has not been evaluated in the point of care location and is for use by Healthcare Professionals only.

This assay provides only a preliminary analytical test result. A more specific alternative chemical method must be used in order to obtain a confirmed analytical result. Gas chromatography/mass spectrometry (GC/MS) has been established as the preferred confirmatory method by the Substance Abuse Mental Health Services Administration (SAMHSA). Clinical consideration and professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are indicated

SUMARRY AND EXPLANATION

Synthetic cannabis is a psychoactive herbal and chemical product that, when consumed, mimics the effects of cannabis. It is best known by the brand name K2 and Spice, both of which have largely become genericized trademarks used for refer to any systhetic cannabis product. The studies suggest that synthetic cannabinoid intoxication is associated with acute psychosis, worsening of previously stable psychotic disorders, and also may have the ability to trigger a chronic (long-term) psychotic disorder among vulnerable individuals such as those with a family history of mental illness. A large and complex variety of synthetic cannabinoids, most often cannabicyclohexanol, JWH-018, JWH-073, or HU-210, are used. As of March 1, 2011, five cannabinoids, JWH-018, JWH-073, CP-47, JWH-200 and cannabicyclohexanol are illegal in US because these substances have the potential to be extremely harmful and, therefore, pose an imminent hazard to the public safety.

TEST PRINCIPLE

K2 Test is based on the principle of specific immunochemical reaction between antibodies and antigens to analyze particular compounds in human urine specimen. The assay relies on the competition for binding antibody between drug conjugate and free drug which may be present in the urine specimen being tested. When drug is present in the urine specimen, it competes with drug conjugate for the limited amount of antibody-dye conjugate. When the amount of drug is equal or more than the cut-off, it will prevent the binding of drug conjugate to the antibody. Therefore, a positive urine specimen will not show a colored band on the test line zone, indicating a positive result, while the presence of a colored band indicates a negative result.

A control line is present in the test window to work as procedural control. This colored band should always appear on the control line zone if the test device is stored in good condition and the test is performed appropriately.

SPECIMEN COLLECTION AND PREPARATION

It is required that approximately 120 - 150 µl of sample for each test. Fresh urine specimens do not need any special handling or treatment. Specimens should be collected in a clean, dry, plastic or glass container. If the assay is not performed immediately, urine specimen may be refrigerated at 2-8ºC or frozen up to 7 days. Specimens should be thawed and brought to room temperature before testing. Urine specimens exhibiting a large amount of precipitate or turbidity should be centrifuged or allowed to settle before testing. Avoid contact with skin by wearing gloves and proper laboratory attire.

MATERIALS AND COMPONENTS

Materials provided with the test kits

1. Instructions for use.
2. K2 Test: The amount of each coated antigen and/or antibody on the strip is less than 1.0 mg for antigen conjugate and is less than 1.0 mg for goat anti-rabbit IgG antibody.
   Test zone: contains K2 protein antigen conjugates.
   Control zone: contains Goat anti-rabbit IgG antibody.
3. Conjugate pad: contains Mouse anti-K2 monoclonal antibody.

Materials required but not provided

1. Urine collection container.
2. Timer or clock.

ASSAY PROCEDURE

For Test Strip (Cat. No. 121154-1)

1. Bring all materials and specimens to room temperature.
2. Remove the test strip from the sealed foil pouch.
3. Dip the strip into the urine specimen with the arrow pointing toward the sample. The sample level should not be higher than the arrow pointed maximum line.
4. Hold the strip in the urine until a reddish color appears at the test area (approximately 20 seconds).
5. Withdraw the strip and place it face up on a clean, non-absorptive surface or leave the strip in urine if the urine level is not higher than arrow pointed maximum line.
6. Read the results at 5 to 10 minutes after adding the sample. **Test results may not be accurate after 10 minutes.**

For Test Card (Cat. No. 121153-1)

1. Bring all materials and specimens to room temperature.
2. Remove the test card from the sealed foil pouch.
3. Place the transfer pipette in the specimen and depress the bulb to

withdraw a sample.

4. Hold the pipette in a vertical position over the sample well of the test card and deliver 3 drops (120-150 µl) of sample in to the sample well.
5. Read the results at 5 to 10 minutes after adding the sample. **Test results may not be accurate after 10 minutes.**

RESULTS

- **Negative:** Two colored bands form. The appearance of two colored bands, one in test line zone and the other in control line zone, indicates negative results. The negative result indicates that the concentrations of metabolites of JWH-18 and/or JWH-73 in the specimen is either zero or less than cut-off level.
- **Positive:** One colored band forms. One colored band appears in control line zone. No colored band is found in test line zone. This is an indication that the concentrations of metabolites of JWH-18 and/or JWH-73 in the specimen is above the cut-off level.
- **Invalid:** If there is no colored band in control line zone, the test result is invalid. Retest the sample with a new device.
  Note: A borderline (+/-) in test line zone should be considered negative result.

QUALITY CONTROL

Good Laboratory Practice recommends the daily use of control materials to validate the reliability of device. Control materials should be assayed as clinical specimen and challenging to the assay cutoff concentration, e.g., 25% above and below cutoff concentration. If control values do not fall within established range, assay results are invalid. Control materials which are not provided with this test kit are commercially available.

The K2 Test provides a built-in process control with a different antigen/antibody reaction at the control region (C). This control line should always appear regardless the presence of drug or metabolite. If the control line does not appear, the test device should be discarded and the obtained result is invalid. The presence of this control band in the control region serve as 1) verification that sufficient volume is added, 2) that proper flow is obtained.

EXPECTED RESULTS

K2 Test is a qualitative assay. It identifies JWH-018 pentanoic acid or JWH-073 butanoic acid in human urine at a concentration of 50 ng/ml or higher. The concentration of the synthetic cannabis cannot be determined by this assay. The test is intended to distinguish negative result from presumptive positive result. All positive results must be confirmed using an alternate method, preferably GC/MS.

PERRFOMANCE CHARACTERISTICS

A. Sensitivity

The cut-off concentration (sensitivity level) of K2) Test is determined to be 50 ng/ml of JWH-018 5-pentanoic acid metabolite and JWH-73 4-butanoic acid metabolite respectively.

B. Accuracy

The accuracy of the K2 Test was evaluated in K2 spiked urine specimens. Forty (40) K2 urine specimens were spiked with JWH-018 pentanoic acid or JWH-73 butanoic acid from 10 to 150 ng/ml. 30 samples with JWH-018 pentanoic acid or JWH-073 butanoic acid concentration between 50 and 150 ng/mL were all found positive (100% agreement), 10 samples with JWH-018 pentanoic acid or JWH-073 butanoic acid concentration between 10 and 37.5 ng/mL were found negative.

C. Precision

The precision study was performed by three individuals observing the test result 5 times each day for 5 consecutive days to determine the random error of visual interpretation. Totally 25 devices for each control level were tested. The test results were found to have no significant differences between the three observers.

Device	Control Con. Ng/ml	No. of Tested	No. of positive			No. of negative		
			1*	2*	3*	1*	2*	3*
JWH-018 pentanoic acid	0	25				25	25	25
	25	25				25	25	25
	37.5	25				25	25	25
	62.5	25	25	25	25			
	75.0	25	25	25	25			

D. Specificity

The specificity for K2 Test test was tested by adding 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.

1. Interference testing

The K2 Test performance at cut-off level is not affected when pH and Specific Gravity ranges of urine specimen are at 5.0 to 8.0 and 1.005 to 1.035.

The following substances were tested and confirmed not to interfere with K2 Test at the listed concentrations.

Glucose	2000 mg/dL
Human albumin	2000 mg/dL
Human hemoglobin	10 mg/dL
Urea	4000 mg/dL
Uric acid	10 mg/Dl

2. Specificity

Diagnostic Automation, Inc. K2 Test is specific with JWH-018 pentanoic acid and JWH-073 butanoic acid.

Compounds	Concentration	Cross reactivity
JWH-018 pentanoic acid	50 ng/ml	100%
JWH-018 N-propanoic acid	25 ng/ml	200%
JWH-018 N-4-hydroxypentyl	2000 ng/ml	2.5%
JWH-018 N-5-hydroxypentyl	2000 ng/ml	2.5%
JWH-073 butanoic acid	25 ng/ml	200%
JWH-073 N-4-hydroxybutyl	1000 ng/ml	5%
JWH-073 N-2-hydroxybutyl	2000 ng/ml	2,5%
JWH-019 6-hydroxyhexyl	2000 ng/ml	2.5%
JWH-019 5-hydroxyhexyl	2000 ng/ml	2.5%
JWH122 N-4-hydroxypentyl	2000 ng/ml	2.5%
JWH-122 N-5-hydroxypentyl	5000 ng/ml	1%
JWH200 6-hydroxyindole	2000 ng/ml	2.5%
JWH210 N-5-carboxypentyl	200 ng/ml	25%
JWH-398 N-pentanoic acid	200 ng/ml	25%
MAM2201 N-pentanoic acid	100 ng/ml	50%
RCS4 N-5-carboxypentyl	750 ng/ml	15%

Each listed substance that may be found in the urine was evaluated and indicated negative result at concentration of 100 µg/ml or higher unless it is specified.

Diagnostic Automation/ Cortez Diagnostics, Inc.
21250 Califa St, Suite 102 and 116, Woodland Hills, CA 91367 USA Phone: 818-591-3030, Fax : 818-591-8383
Email: onestep@rapidtest.com Website: www.rapidtest.com

Acetaminophen	4-Acetamidophenol	Acetylsalicylic Acid
Amitriptyline	Amobarbital	Amphetamine
Aspartame	Ascorbic acid	Atenolol
Atrophine	Benzoyolecgonine	Buprenorphine
Butabarbital	Butalbital	Caffeine
Cannabidiol	Cannabinal	Cetirizine
Cortisone	Chlorpheniramine	Cocaine
Cotinine	Despiramine	Dextromethorphan
Digitoxin	Digoxin	Diphenhydramine
Doxylamine	Ecgonine	Ecgonine methyl ester
Ephedrine	Epinephrine	Fentanyl
Gentisic acid	Guaiacol glycer ester	Hertoine
Homatrophine	Hydrochlorothiazide	Hydroxyzine
Imipramine	Isoproterenol	Ketamine
MDA	MDMA	Meperidine
Methamphetamine	Methadol	Methadone
Morphine	Nalbuphine	Nalophine
Natrexone	Neomycin	Niacinamide
Noigertaline	Nortriptyline	Orphenadine
Oxycodone	Oxymorphone	Perphenazine
Pentobarbital	Phencyclidine (PCP)	Phenobarbital
Phenylpropanolamine	Promethazine	Propranolol
Protriptyline	Quetiapine fumarate	Quinine antidine
Salicylic acid	Secobarbital	Sertal
Tetracycline	Terahydrozoline	Theophyline
Trifluoperazine	Trihexyphenidyl	Trimipramine
Tyramine	Venlafaxine	Verapamil

11-nor-Δ^8-THC-9-COOH(10 µg/ml)* 11-nor-Δ^9-THC-9-COOH(10 µg/ml)*

Amikacin
Arterenol
Atenonal
Bup-3- β-glucuronide
Camphor
Chloroquine
Codeine
Deoxyephedrine
Diphenhydramine
EDDP
Fluoxetine
Histamine
Ibuprofen
Lidocaine
Methylphenidate
Methaqualone
Nalxone
Nicotine
Oxcarbazepine
Penicillin G
Phenylethylamine-α
Propoxyphene
Ranitidine
Sertraline
Thioridazine
Tryptophen

*The highest level to be tested is as indicated.

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 in certain foods and medicines may interfere with the test and cause false results. Please refer "SPECIFICITY" section for lists of substances that will produce either positive results, or that do not interfere with test performance. If a drug/metabolite is found present in the urine specimen, the assay does not indicate frequency of drug use or distinguish between drug of abuse and certain foods and medicines.

PRECAUTIONS

1. For in vitro diagnostic and forensic use only.
2. Do not use the product beyond the expiration date.
3. Handle all specimens as potentially infectious.
4. Humidity sensitive product, do not open foil pouch until it is ready to be tested.
5. Use a new urine specimen cup for each sample to avoid cross contamination.
6. The test device should be stored at 4 to 30ºC and will be effective until the expiration date stated on the package.
7. The product is humidity-sensitive and should be used immediately after being open.
8. Any improperly sealed product should be discarded.

REFERENCES

1. Urine testing for drugs of abuse, NIDA Research Monograph 73 (1986)
2. InfoFacts-Club drugs, NIDA , May 2006 . http://www.nida.nih.gov/infofacts/clubdrugs.html
3. Synthetic cannabis (2012) http://en.wikipedia.org/wiki/synthetic_cannabis

ISO 13485
ISO 9001

Diagnostic Automation/
Cortez Diagnostics, Inc.
21250 Califa St, Suite 102 and 116,
Woodland Hills, California 91367 USA

Date Adopted	2016-01-25
REF 121154-1 REF 121153-1	CORTEZ- OneStep K2 Urine RapiDip™ InstaTest RapiCard™ InstaTest

Revision Date: 2013-07-10