One Step K2/Spice Test
Package Insert

A rapid, one step test for the qualitative detection of Synthetic Cannabis (K2/Spice) in human urine.

For forensic use only.

INTENDED USE

The One Step K2/Spice Test is a lateral flow chromatographic immunoassay for the qualitative detection of synthetic cannabis (K2/Spice) JWH-018 and JWH-073's major metabolites in human urine at a cut-off concentration of 50 ng/mL. This assay has not been evaluated in the point of care field, is for forensic use only.

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) or Liquid Chromatography/ Mass Spectrometry (LC/MS) are the preferred confirmatory methods. Clinical consideration and professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are used.

SUMMARY

Synthetic cannabis is a psychoactive herbal and chemical product that, when consumed, mimics the effects of cannabis. It is best known by the brand names **K2** and **Spice**, both of which have largely become genericized trademarks used to refer to any synthetic 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. As of March 1, 2011, five cannabinoids, JWH -018, JWH-073, CP-47, JWH-200 and cannabicyclohexanol are now illegal in the US because these substances have the potential to be extremely harmful and, therefore, pose an imminent hazard to the public safety.

The One Step K2/Spice Test yields a positive result when the concentration of JWH 018 and JWH-073's metabolites in urine exceeds 50 ng/mL.

PRINCIPLE

The One Step K2/Spice Test 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. JWH 018 and JWH-073's metabolites, if present in the urine specimen below 50 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 K2/Spice conjugate and a visible colored line will show up in the test region. The colored line will not form in the test region if the JWH 018 and JWH-073's metabolites level exceeds 50 ng/mL because it will saturate all the binding sites of anti-K2/Spice antibodies.

A drug-positive urine specimen will not generate a colored line in the test region because of drug competition, while a drug-negative urine specimen or a specimen containing a drug concentration lower than the cut-off will generate a line in the test region.

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

REAGENTS

The test contains mouse monoclonal anti-K2/Spice antibody-coupled particles and K2/Spice -protein conjugate. A goat antibody is employed in the control line system.

PRECAUTIONS

- For professional use only.
- For forensic use only.
- 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 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℃. The test is stable through the expiration date printed on the sealed pouch. The test must remain in the sealed pouch until use. DO NOT FREEZE. Do not use beyond the expiration date. The product is humidity-sensitive and should be used immediately after being open.

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 obtain a clear specimen for testing.

Specimen Storage

Urine specimens may be stored at 2-8℃ for up to 96 hours prior to assay. For long-term storage, specimens may be frozen and stored below -20℃. Frozen pecimens should be thawed and mixed before testing.

MATERIALS

Materials Provided

- Test Devices
- Package insert

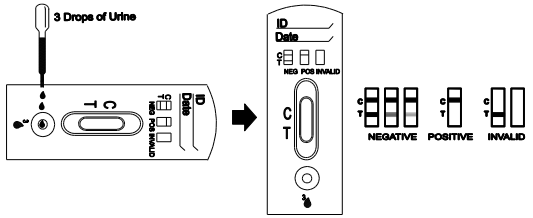
Materials Required But Not Provided

- Specimen collection container
- Timer
- External controls

DIRECTIONS FOR USE

Allow the test device, urine specimen, and/or controls to equilibrate to room temperature (15-30℃) 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. 100 μL) 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 colored line(s) to appear. The result should be read at 5 to 10 minutes. It is important that the background is clear before the result is read. Do not interpret the result after 10 minutes.



INTERPRETATION OF RESULTS

(Please refer to the illustration above)

NEGATIVE:* Two lines appear. One colored line should be in the control region (C), and another apparent colored line should be in the test region (T). A negative result indicates that the K2/Spice concentration is below the detectable level of 50 ng/mL.
*** NOTE:** The shade of color in the test region (T) will vary, but it should be considered negative whenever there is even a faint pink line.

POSITIVE: One colored line appears in the control region (C). No line appears in the test region (T). A positive result indicates that the K2/Spice concentration exceeds the detectable level (50 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 the manufacturer.

QUALITY CONTROL

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

LIMITATIONS

1. The One Step K2/Spice Test 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) or Liquid Chromatography/Mass Spectrometry (LC/MS) are the preferred 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, 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 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 cut-off level of the test.
6. Test does not distinguish between drugs of abuse and certain medications.

EXPECTED RESULTS

The K2/Spice test is a qualitative assay that detects JHW-018, JHW-073's major metabolites in human urine at a concentration of 50 ng/mL or higher. The assay provides only a preliminary analytical results. **A more specific alternate chemical method must be used in order to obtain a confirmed analytical result.**

PERFORMANCE CHARACTERISTICS

Analytical Sensitivity

A drug-free urine pool was spiked with JHW-018 N-(5-hydroxypentyl) metabolite at the following concentrations: 0 ng/mL, 25 ng/mL, 37.5 ng/mL, 62.5 ng/mL and 75 ng/mL .. The result demonstrates 99% accuracy at 50% above and 50% below the cut-off concentration. The data are summarized below:

JHW-018 N-(5-hydroxypentyl) metabolite Concentration (ng/mL)	Percent of Cut-off	n	Visual Result		Positive (%)
			Negative	Positive	
0	0%	20	20	0	0%
25	50%	20	20	0	0%
37.5	75%	20	18	2	10%
62.5	125%	20	2	18	90%
75	150%	20	0	20	100%

Analytical Specificity

Cross-reactivity was established by spiking various concentrations of similarly structure drug compounds into drug-free urine or negative urine. Analyzing various concentrations of each compound by using K2/Spice drug test, the concentration of each compound that produces a response approximately equivalent to the cut-off concentration of the assay was determined. The result is summarized in the table below:

Drug Compound	**Response equivalent to cut-off in ng/mL**
JWH-018 N-(5-hydroxypentyl) metabolite	50
JWH-073 N-(4-hydroxybutyl) metabolite	50
JWH-018 pentanoic acid	50

JWH-073 butanoic acid	50
JWH-018	15,000
JWH-073	15,000

Precision/Reproducibility Study

Precision/Reproducibility was determined by replicating tests on five different concentrations of JWH-018 N-(5-hydroxypentyl) metabolite in urine specimens: negative, 50% below cut-off, 25% below cut-off, 25% above cut-off and 50% above cut-off with total 20 assays at each concentration. The data are summarized below:

JWH-018 N-(5-hydroxypentyl) metabolite Concentration (ng/mL)	Total Numbers of Determinations	Results #Neg/#Pos	Precision (%)
0	20	20/0	100%
25	20	20/0	100%
37.5	20	18/2	90%
62.5	20	2/18	90%
75	20	0/20	100%

Interfering compounds

The following compounds in both drug-free urine and K2/Spice positive urine show no cross-reactivity when tested with One Step K2/Spice Test at a concentration of 50ng/mL.

Common Substances:

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	L-Phenylephrine
Creatine	B-Phenylethylamine
Dexbrompheniramine	Procaine
Dextromethorphan	Quinidine
Diphenhydramine	Ranitidine
Dopamine	Riboflavin
(+/-)-Epinephrine	Sodium Chloride
Erythromycin	Sulindac
Ethanol	Theophylline
Furosemide	Tyramine
Glucose	4-Dimethylaminoantipyrine
Guaiacol Glyceryl Ether	(1R,2S)-(-)-N-Methyl-Ephedrine
Hemoglobin	

Biological Materials:

Albumin	Vitamin(L-Ascorbic Acid)
Bilirubin	Uric Acid
Creatine	Urine pH 4.5-9.0
Hemoglobin	Urine Specific Gravity 1.002-1.035g/mL
Glucose	

There is a possibility that other substances and/or factors not listed above may interfere with the test and cause false results.

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3. Synthetic Cannabis (2012), http://en.wikipedia.org/wiki/Synthetic_cannabis

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