

CEDIA® Cannabinoids OFT Assay (ANZ)

Thermo
SCIENTIFIC

IVD For In Vitro Diagnostic Use Only
(For Use with Oral-Eze® Oral Fluid Collection System)

REF 10010883 (3 x 18 mL Kit)
10010888 (65 mL Kit)

Intended Use

The Thermo Scientific CEDIA Cannabinoids OFT Assay is intended for use in the semi-quantitative determination of Cannabinoids in human oral fluid at the diluted target concentration of 8 ng/mL. The specimen must be collected exclusively with the Oral-Eze® Oral Fluid Collection System which collects approximately 1 mL of neat oral fluid and dilutes with 2 mL buffer. The assay is calibrated against Δ^9 THC. This in vitro diagnostic device is intended for clinical laboratory use only.

The CEDIA® Cannabinoids OFT Assay provides only a preliminary analytical test result. A more specific alternative method must be used to obtain a confirmed analytical result. Gas Chromatography/Mass Spectrometry (GC/MS) and Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS) are the preferred confirmatory methods.⁽¹⁻³⁾ Clinical consideration and professional judgment should be applied to any drug of abuse test result particularly when preliminary positive results are used.

Summary and Explanation of the Test

The collection of oral fluid is less invasive and no special facilities are required. Oral fluid contains mostly parent drug and therefore is a better indicator of recent drug use.

Cannabinoids and minor amounts of cannabinoid metabolites could be detected in oral fluid immediately within 30 minutes after administration. Therefore, detection of cannabinoids and their metabolites in oral fluids is a good indicator of recent use.^(4,5) Detection levels and duration of detection of cannabinoids in oral fluid are dependent up on pH and amount of drug consumed.

CEDIA Cannabinoids OFT Assay uses recombinant DNA technology to produce a unique homogeneous enzyme immunoassay system.⁽⁶⁾ The assay is based on the bacterial enzyme β -galactosidase, which has been genetically engineered into two inactive fragments. These fragments spontaneously re-associate to form fully active enzyme that, in the assay format, cleave a substrate, generating a color change that can be measured spectrophotometrically.

In the assay, analyte in the sample competes with analyte conjugated to one inactive fragment (enzyme donor) of β -galactosidase for antibody binding site. If analyte is present in the sample, it binds to antibody, leaving the inactive enzyme fragment free to form active enzyme. If the analyte is not present in the sample, antibody binds to analyte conjugated on the inactive fragment, inhibiting the re-association of inactive β -galactosidase fragments, and no active enzyme is formed. The amount of active enzyme formed and resultant absorbance change are directly proportional to the amount of analyte present in the sample.

Reagents

- 1. EA Reconstitution Buffer**
Contains buffer salts, 0.2 mg/L rabbit monoclonal anti-THC antibody, stabilizer and preservative.
- 1a EA Reagent**
Contains 0.171 g/L Enzyme Acceptor (microbial), buffer salts and preservative.
- 2. ED Reconstitution Buffer**
Contains buffer salts, stabilizers, and preservative.
- 2a ED Reagent**
Contains 0.175 nM Enzyme Donor (microbial) conjugated to cannabinoid derivative, 1.67 g/L chlorophenol red- β -D-galactopyranoside, stabilizers, detergent and preservative.

Additional Materials Required (sold separately)

REF	Kit Description
10016643	CEDIA THC OFT Negative Calibrator
10016700	CEDIA THC OFT Calibrator 1
10016701	CEDIA THC OFT Calibrator 2
10016702	CEDIA THC OFT Calibrator 3
10017702	CEDIA THC OFT Control 1
10017703	CEDIA THC OFT Control 2
10017704	CEDIA THC OFT Control 3
96100-050	Oral-Eze Collection Device (50/Box)
96100-500	Oral-Eze Collection Device (500/Box)
96105-050	Oral-Eze Sample Extractor (50/Box)
96105-500	Oral-Eze Sample Extractor (500/Box)

Precautions and Warnings

1. This test is for in vitro diagnostic use only. The reagents are harmful if swallowed.
2. Do not use the reagents beyond their expiration dates.

DANGER: Powder reagent contains $\leq 55\%$ w/w bovine serum albumin (BSA), $\leq 1\%$ w/w Sodium azide and $\leq 0.5\%$ w/w Drug-specific antibody (Rabbit). Liquid reagent contains $\leq 0.5\%$ bovine serum (FBS), $\leq 0.15\%$ Sodium azide and $< 0.1\%$ Drug-specific antibody (Rabbit).

H317 – May cause allergic skin reaction.

H334 – May cause allergy or asthma symptoms or breathing difficulties if inhaled.

EUH032 – Contact with acids liberates very toxic gases.

Avoid breathing dust/mist/vapors/spray. Contaminated work clothing should not be allowed out of the workplace. Wear protective gloves/ eye protection/face protection. In case of inadequate ventilation wear respiratory protection. If on skin: Wash with plenty of soap and water. IF INHALED: If breathing is difficult, remove victim to fresh air and keep at rest in a position comfortable for breathing. If skin irritation or rash occurs: Get medical advice/attention. If experiencing respiratory symptoms; Call a POISON CENTER or doctor/physician. Wash contaminated clothing before reuse. Dispose of contents/ container to location in accordance with local/regional/national/international regulations.

Reagent Preparation and Storage

For preparation of the solutions, refer to the section below. Remove the kit from refrigerated storage (2-8°C) immediately prior to preparation of the solutions.

In the case of accidental spill, clean and dispose of material according to your laboratory's SOP, local, and state regulations.

In the case of damaged packaging on arrival, contact your technical support representative (refer to back page of this PI).

Prepare the solutions in the following order to minimize possible contamination.

R2 Enzyme donor solution

Connect Bottle 2a (ED reagent) to Bottle 2 (ED Reconstitution Buffer) using one of the enclosed adapters. Mix by gentle inversion, ensuring that all the lyophilized material from Bottle 2a is transferred into Bottle 2. Avoid the formation of foam. Detach Bottle 2a and adapter from Bottle 2 and discard. Cap Bottle 2 and let stand approximately 5 minutes at room temperature (21-25°C). Mix again. Record the reconstitution date on the bottle label. Place the bottle directly into the reagent compartment of the analyzer or into refrigerated storage (2-8°C) and let stand 30 minutes before use.

R1 Enzyme acceptor solution

Connect Bottle 1a (EA reagent) to Bottle 1 (EA Reconstitution Buffer) using one of the enclosed adapters. Mix by gentle inversion, ensuring that all the lyophilized material from Bottle 1a is transferred into Bottle 1. Avoid the formation of foam. Detach Bottle 1a and adapter from Bottle 1 and discard. Cap Bottle 1 and let stand approximately 5 minutes at room temperature (21-25°C). Mix again. Record the reconstitution date on the bottle label. Place the bottle directly into the reagent compartment of the analyzer or into refrigerated storage (2-8°C) and let stand 30 minutes before use.

NOTE 1: The components supplied in this kit are intended for use as an integral unit. Do not mix components from different lots.

NOTE 2: Avoid cross-contamination of reagents by matching reagent caps to the proper reagent bottle. The R2 solution (Enzyme Donor) should be yellow-orange in color. A red or red-purple color indicates that the reagent has been contaminated and must be discarded.

NOTE 3: The R1 and R2 solutions must be at the reagent compartment storage temperature of the analyzer before performing the assay. Refer to the analyzer specific application sheet for additional information.

Store reagents at 2-8°C. **DO NOT FREEZE.** For stability of the unopened components, refer to the box or bottle labels for the expiration date.

R1 Solution: 60 days refrigerated on analyzer or at 2-8°C.

R2 Solution: 60 days refrigerated on analyzer or at 2-8°C.

Specimen Collection and Handling

Oral fluid samples are suitable for use in the CEDIA Cannabinoids OFT Assay. Collect oral fluid samples using the Oral-Eze Oral Fluid Collection System. Care should be taken to preserve the chemical integrity of the oral fluid sample from the time it is collected until the time it is assayed by securely capping the samples, storing the samples at 2-8°C or at room temperature (21-25°C), and testing within 21 days after collection.

Handle oral fluid samples as if they were potentially infectious.

Samples within a pH range of 5-9 are suitable for testing with this assay.

Oral-Eze Sample Processing Procedure

1. Label the sample collection vial with proper identification.
2. Check the sample collection date on the vial to ensure that the sample is within 21 days from the date of collection.
3. Open the cap and compress the pad to express the sample.
4. Recap the vial and the sample is ready for testing.
5. Ensure that the oral fluid samples are maintained between 4°C and 37°C during shipping.
6. Samples can be stored at room temperature (21-25°C) for 21 days. They should be stored at 2-8°C.

Assay Procedure

The Oral-Eze Oral Fluid Collection Device contains a preservative buffer that dilutes the neat oral fluid sample. The calibrator and control levels are set at diluted levels.

NOTE: To correlate the Oral-Eze result from the assay or the associated LC-MS/MS confirmation result to a neat oral fluid value, the result from the Oral-Eze sample should be multiplied by a factor of 3.

1. Pipet the processed oral fluid samples and controls into labeled sample cups and place the cups into the sample ring of the analyzer.
2. Load reagents (reagent 1 and reagent 2) into the reagent compartment of the analyzer.
3. Pipet calibrators into labeled cups and load the cups into the sample rack of the analyzer.
4. Program the run setup using 540 nm as primary wavelength and 660 nm as the secondary wavelength. Refer to the parameter sheet for detailed instructions on how to program the analyzer.

Quality Control and Calibration

Good laboratory practice suggests that controls be tested each day patient samples are tested and each time calibration is performed. Recalibrate the test if reagents are changed or if control results are outside of established limits. Ensure that control results are within the established range, as determined by laboratory procedures and guidelines. If results fall outside of the established ranges, assay results are invalid. All quality control requirements should be performed in conformance with local, state and/or federal regulations or accreditation requirements. Each laboratory should establish its own calibration and control frequency.

Results and Expected Values

Semi-quantitative Results

A standard curve is run using all the calibrators to estimate relative concentrations of the drug in the samples. Refer to the analyzer specific application sheet for detailed information.

Limitations

A positive result from this assay indicates only the presence of cannabinoids and does not necessarily correlate with the extent of physiological and psychological effects.

It is possible that other substances and/or factors (e.g.: technical or procedural), other than those investigated in the specificity study may interfere with the test and cause false results.

Specific Performance Characteristics

Typical performance results obtained on the Olympus AU680 analyzer are shown below. The results obtained in your laboratory may differ from these data.

Precision

The samples containing various amounts of *l*-isomer Δ^9 THC were tested in semi-quantitative mode using CLSI (EP05-A2) precision protocol. The samples were randomized and tested in replicates of six, twice a day for five days, total N=60. The results are summarized in the table below.

Semi-quantitative Analysis:

Samples	Result Mean (ng/mL)	Within-Run		Total Run	
		SD	%CV	SD	%CV
Control 1	6.1	0.2	3.1	0.3	4.3
Control 2	8.3	0.2	2.9	0.3	3.4
Control 3	10.5	0.3	2.5	0.3	3.2

Specificity and Cross-Reactivity

Cannabinoid compounds and metabolites were tested for cross-reactivity in the assay. The potential cross-reactant compound was spiked into a negative oral fluid at the listed concentrations and tested in semi-quantitative mode. Then concentrations listed below produced a result approximately equal to the cutoff calibrator.

Compounds	Tested Concentration (ng/mL)	Cannabinoids OFT Assay Negative / Positive
<i>l</i> -11-nor- Δ^9 THC-COOH	7.0	Positive
11-OH- Δ^9 THC	8.5	Positive
Δ^8 THC	14.0	Positive
Cannabinol	16.0	Positive
Cannabidiol	35000	Positive

Various common over-the-counter medications and structurally unrelated compounds were tested for cross-reactivity in the assay. The cross-reactant solutions were prepared by adding the compound to negative oral fluid at the concentrations listed in the table below. All the compounds tested negative and did not show any cross-reactivity.

Compounds	Tested Concentration (ng/mL)	Cannabinoids OFT Assay Negative/Positive
Acetaminophen	80,000	Negative
Acetylsalicylic Acid	80,000	Negative
Alprazolam	10,000	Negative
Amobarbital	10,000	Negative
Amoxicillin	80,000	Negative
Amphetamine	80,000	Negative
Ampicillin	10,000	Negative
Atropine	10,000	Negative
Benzoylcegonine	40,000	Negative

(Con't)

Compounds	Tested Concentration (ng/mL)	Cannabinoids OFT Assay Negative/Positive
Butabarbital	10,000	Negative
Butalbital	10,000	Negative
Caffeine	8,000	Negative
Captopril	40,000	Negative
Chlorpromazine	10,000	Negative
Chorazepate	10,000	Negative
Chordiazepoxide	8,000	Negative
Cimetidine	40,000	Negative
Clonazepam	10,000	Negative
Cocaethylene	10,000	Negative
Cocaine	500	Negative
Codeine	80,000	Negative
Cyclizine	10,000	Negative
Dextromethorphan	80,000	Negative
Diazepam	40,000	Negative
Digoxin	8,000	Negative
Diphenhydramine	10,000	Negative
Enalapril	40,000	Negative
Fluoxetine	40,000	Negative
Gentisic Acid	10,000	Negative
Hydrocodone	10,000	Negative
Hydromorphone	10,000	Negative
Ibuprofen	40,000	Negative
Imipramine	10,000	Negative
/-Ephedrine	10,000	Negative
Levothyroxine	4,000	Negative
Lidocaine	10,000	Negative
Loperamide	10,000	Negative
Medazepam	10,000	Negative
Meperidine	80,000	Negative
Methadone	80,000	Negative
Methamphetamine	80,000	Negative
Metoprolol	10,000	Negative
Morphine	16,000	Negative
Naproxen	80,000	Negative
Niacinamide	10,000	Negative
Nifedipine	40,000	Negative
Norchlordiazepoxide	10,000	Negative
Oxazepam	40,000	Negative
Penicillin	10,000	Negative
Phencyclidine	80,000	Negative
Phenethylamine	10,000	Negative
Phenobarbital	80,000	Negative
Phenylephrine	10,000	Negative
Phenylpropanolamine	10,000	Negative
Procainamide	10,000	Negative
Procaine	10,000	Negative
Propoxyphene	80,000	Negative
Pseudoephedrine	10,000	Negative
Quinidine	10,000	Negative
Ranitidine	40,000	Negative
Salbutamol	10,000	Negative
Salicylic Acid	40,000	Negative

(Con't)

Compounds	Tested Concentration (ng/mL)	Cannabinoids OFT Assay Negative/Positive
Secobarbital	80,000	Negative
Temazepam	10,000	Negative
Theophylline	10,000	Negative
Tolmetin	40,000	Negative
Verapamil	40,000	Negative
Zomepirac	10,000	Negative

Endogenous, Exogenous Substances and pH Interference

The potential interference from several endogenous and exogenous substances, and pH on the detection accuracy of the samples containing *Δ*⁹ THC at the control 1 and control 3 concentrations were tested in the assay. The interfering substances were added to the negative oral fluid at the concentrations listed in the table below. The samples were tested in semi-quantitative mode. No interference was observed with the interfering substances and pH samples at the control 1 and control 3 concentrations.

Substances	Tested Concentration (ng/mL)	Cannabinoids OFT Assay	
		Control 1	Control 3
Control 1	6.0	Negative	N/A
Control 3	10.0	N/A	Positive
Cotinine	0.01 mg/mL	Negative	Positive
Nicotine	0.01 mg/mL	Negative	Positive
Hemoglobin	0.10 mg/mL	Negative	Positive
Human Serum Albumin	2 mg/mL	Negative	Positive
Sodium Chloride	6.0 mg/mL	Negative	Positive
Cholesterol	0.15 mg/dL	Negative	Positive
Acetaminophen	0.1 mg/mL	Negative	Positive
Acetylsalicylic Acid	0.1 mg/mL	Negative	Positive
Caffeine	0.1 mg/mL	Negative	Positive
Ibuprofen	0.1 mg/mL	Negative	Positive
Coffee	2% v/v	Negative	Positive
Milk	0.5 % v/v	Negative	Positive
Orange Juice	2% v/v	Negative	Positive
Cranberry Juice	2% v/v	Negative	Positive
Soft drink (Coke)	2% v/v	Negative	Positive
Toothpaste	2% v/v	Negative	Positive
Mouthwash	2% v/v	Negative	Positive
Tea	2% v/v	Negative	Positive
Denture Adhesive	2% v/v	Negative	Positive
pH	5-9	Negative	Positive

Method Comparison

Forty-two oral fluid samples from rehabilitation clinic were collected using the Oral-Eze Oral Fluid Collection Device. The oral fluid samples were tested in the CEDIA Cannabinoids OFT Assay and by LC-MS/MS method.

Semi-quantitative Analysis: The overall concordance between CEDIA Cannabinoids OFT Assay and LC-MS/MS is 95.0%. Then comparison of sample results by the CEDIA Cannabinoids OFT Assay to LC-MS/MS showed 100.0% sensitivity and 91% specificity.

		Semi-quantitative LC-MS/MS (ng/mL)	
		+	-
CEDIA	+	20	*2
	-	0	20

*Discordant samples were border negative by LC-MS/MS.

References

1. Cone, E. J., et al. (2002). Oral Fluid Testing for Drugs of Abuse: Positive Prevalence Rates by Intercept (TM) Immunoassay Screening and GC-MS-MS Confirmation and Suggested Cutoff Concentrations. *Journal of Analytical Toxicology*, 26, 541-547.
2. Maurer, H. H. (2005). *Advances in Analytical Toxicology*: The Current Role of Liquid Chromatography-Mass Spectrometry in Drug Quantification in Blood and Oral Fluid. *Analytical and Bioanalytical Chemistry*, 381, 110-118.
3. Lambert, W. E., et al. (2002). Simultaneous, quantitative determination of opiates, amphetamine, cocaine and Benzoylcegonine in oral fluid by liquid chromatography quadrupole-time of-flight mass spectrometry. *Chromatogr B Analyt Technol Biomed Life Sci.*, 779, 321-30.
4. Niedbala, R. S., et al. (2001). Detection of marijuana use by oral fluid and urine analysis following single dose administration of smoked and oral marijuana. *Journal of Analytical Toxicology*, 25, 290-303.
5. Samyn, N., et al. (1999). *Analysis of Drugs of Abuse in Saliva. Forensic Science Review*, 11, 1-19.
6. Henderson, D. R., et al. (1986). CEDIA, A New Homogeneous Immunoassay System *Clin. Chem.*, 32, 1637-1641.



Microgenics Corporation
46500 Kato Road
Fremont, CA 94538 USA
US Customer
and Technical Support:
1-800-232-3342



EC REP

Thermo Fisher Scientific Oy
Ratastie 2, P.O. Box 100
01621 Vantaa, Finland
Tel: +358-9-329100
Fax: +358-9-32910300



For insert updates go to:
www.thermoscientific.com/diagnostics

Other countries:

Please contact your Thermo Fisher Scientific representative.

AU680® is a trademark of Beckman, Coulter Inc. and its subsidiaries.
CEDIA® is a registered trademark of Roche Diagnostics.

100181231NS-3
2015 09

Thermo
SCIENTIFIC