DRI® Fentanyl Assay

Thermo SCIENTIFIC

For Criminal Justice and Forensic Use Only

REF 10016006 (3x18 mL Kit)
10016005 (500 mL Kit)

Intended Use
The DRI® Fentanyl Enzyme Immunoassay is intended for the qualitative determination of Fentanyl in human urine.

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) and Liquid Chromatography/tandem mass spectrometry are the preferred confirmatory methods.[1]

Summary and Explanation of the Test
Fentanyl is a synthetic opiate analgesic similar to morphine. Fentanyl is 50-100 times more potent than morphine. It is prescribed mainly for patients with chronic pain and is generally used to manage pain after surgery. Fentanyl is prescribed as intravenous anesthetic (Sublimaze®), transdermal patch (Duragesic®), and transmucosal Lozenge form (Actiq®). The fentanyl dose in the Duragesic ranges from 2.5-10 mg and in Actiq, it ranges from 0.2 mg-1.6 mg. Half-life of Fentanyl is 3-12 hours.[2,3] Fentanyl is exclusively metabolized by N-dealkylation and hydroxylation.[4, 5] More than 90% of the dose is eliminated as norfentanyl and hydroxylated metabolites.[6] Less than 7% of the dose is excreted unchanged in the urine.[7,8]

The DRI Fentanyl Assay is supplied as liquid ready-to-use homogeneous enzyme immunoassay. The assay uses fentanyl specific monoclonal antibody that can detect fentanyl without any significant cross-reactivity to other opiate compounds.

The assay is based on competition between a drug labeled with glucose-6-phosphate dehydrogenase (G6PDH), and free drug from the urine sample, for a fixed amount of specific antibody binding sites. In the absence of free drug from the sample, the specific antibody binds the drug labeled with G6PDH and causes a decrease in enzyme activity. This phenomenon creates a direct relationship between the drug concentration in urine and enzyme activity. The enzyme activity is determined spectrophotometrically at 340 nm by measuring the conversion of nicotinamide adenine dinucleotide (NAD) to NADH.

Reagents
REAGENT ***Antibody/Substrate Reagent (R1):***
Contains mouse monoclonal anti-fentanyl antibody, glucose-6-phosphate (G6P), and nicotinamide adenine dinucleotide (NAD) in Tris buffer with sodium azide as a preservative.

REAGENT ***Enzyme Conjugate Reagent (R2):***
Contains fentanyl derivative labeled with glucose-6-phosphate dehydrogenase (G6PDH) in Tris buffer with sodium azide as a preservative.

Additional Materials Required (sold separately):

REF	Kit Description
1388	DRI® Negative Calibrator (25 mL)
10016023	DRI® Fentanyl 2 ng/mL Calibrator (10 mL)
10016022	DRI® Fentanyl Low Control 1 ng/mL (25 mL)
10016024	DRI® Fentanyl High Control 3 ng/mL (25 mL)

⚠ Precautions and Warning
DANGER: 1. The reagents are harmful if swallowed.
2. The reagents contain ≤0.2% bovine serum albumin (BSA) and ≤0.5% Drug-specific antibody (Mouse).
3. Reagents used in the assay components contain ≤0.09% sodium azide, which may react with lead or copper plumbing to form potentially explosive metal azides. When disposing of such reagents, always flush with large volumes of water to prevent azide build-up.

H317 - May cause allergic skin reaction.
H334 - May cause allergy or asthma symptoms or breathing difficulties if inhaled.

Avoid breathing mist or vapor. 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
The reagents are ready-to-use; no additional preparation is required. Reagents should be stored refrigerated. All assay components, opened or unopened, are stable until the expiration date indicated on their respective labels. Do not use the reagents beyond their expiration dates.

Specimen Collection and Handling
Collect urine specimens in plastic or glass containers. Fresh urine specimens should be used. Samples within a pH range of 4 to 9 are suitable for testing with this assay. An effort should be made to keep pipetted samples free of gross debris. Centrifuge highly turbid specimens before analysis. Adulteration may cause erroneous results. If adulteration is suspected, obtain another sample and forward both specimens to the laboratory for testing. **Handle all urine specimens as if they were potentially infectious.**

Assay Procedure
Clinical chemistry analyzers capable of maintaining a constant temperature, pipetting samples, mixing reagents, measuring enzymatic rates at 340 nm and timing the reaction accurately can be used to perform this immunoassay. Refer to specific application instructions for each analyzer for chemistry parameters before performing the assay.

Quality Control and Calibration
Good laboratory practice suggests the use of control specimens to ensure proper assay performance. Ensure that control results are within the established ranges, as determined by laboratory procedures and guidelines. If results fall outside of the established ranges, assay results are invalid. All QC requirements should be performed in conformance with local, state and/or federal regulations or accreditation requirements.

Results and Expected Values
The 2.0 ng/mL calibrator is used as a cutoff reference for distinguishing 'postive' from 'negative' samples. A sample that exhibits a change in absorbance values (ΔA) equal to or greater than that obtained with the cutoff calibrator is considered positive. A sample that exhibits a change in absorbance value (ΔA) lower than that obtained with the cutoff calibrator is considered as negative.

Limitations
1. A positive result from this assay indicates only the presence of fentanyl.
2. 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 Beckman Coulter Olympus AU680 analyzer are shown below. The results obtained in your laboratory may differ from these data.

Precision

Fentanyl	n = 240	Within-run		Total run	
Cal / Ctrls	Mean (mA/min)	SD	CV%	SD	CV%
1 ng/mL	462	2.9	0.63	3.6	0.78
2 ng/mL	476	2.5	0.53	3.5	0.74
3 ng/mL	497	2.6	0.52	3.2	0.65

Interference

Endogenous and exogenous substances were tested for potential interference in the DRI Fentanyl Assay. No interference was observed in urine samples containing the compounds up to the concentrations listed below. Urine sample pH levels from 4 – 9 were also studied for possible interference.

Compound	Concentration (mg/dL)
Acetaminophen	10
Acetone	500
Acetyl Salicylic Acid	10
Ascorbic acid	150
Caffeine	10
Creatinine	400
Ethanol	1000
Galactose	5
Glucose	1000
Hemoglobin	150
Human Serum Albumin	200
Ibuprofen	10
Oxalic Acid	50
Riboflavin	3
Sodium chloride	400
Urea	1000
pH	4-9

Specificity

The specificity of the assay was evaluated by testing various fentanyl analogs, major metabolite norfentanyl and various other opiate compounds (natural and synthetic). All compounds listed below produced negative result at the concentration tested.

Cross Reactants	Tested Concentration (ng/mL)
6-Acetyl morphine	50,000
Alfentanil	10,000
Buprenorphine	100,000
Buprenorphine glucuronide	100,000
Codeine	100,000
Despropionyl Fentanyl	100
Dihydrocodeine	200,000
EDDP	50,000
Heroin	50,000
Hydrocodone	100,000
Hydromorphone	100,000
Methadone	300,000
Levorphanol	20,000
Meperidine	100,000
Morphine	200,000
Morphine-3-Glucuronide	500,000
Norbuprenorphine	50,000
Naloxone	25,000
Naltrexone	25,000
Norfentanyl	10,000
Norcodeine	100,000
Normeperidine	50,000
Normorphine	100,000
Oxycodone	200,000
Oxymorphone	500,000
Sufentanil	100
Tapentadol	50,000
Tapentadol-O-Glucuronide	50,000
Tilidine	50,000
Tramadol	100,000
Tramadol-O-Desmethyl	100,000
Tramadol-N-Desmethyl	100,000

Various compounds that are concomitantly used and are structurally related or unreleated to Fentanyl were tested for their protential interference in the assay. All the compounds listed below producted a negative result at the concentrations tested.

Non-Critical cross reactants	Tested Concentration (ng/mL)
Acetaminophen	500,000
Acetylsalicyclic acid	500,000
Amitryptiline	25,000
Amoxicillin	100,000
Amphetamine	1,000,000
Aripiprazole	500
Benzoylecgonine	1,000,000
Caffeine	100,000
Carbamazepine	500,000
Cetrizine	50,000
Chlorpromazine	100,000
Clomipramine	25,000
Cimetidine	500,000
Dehydroaripiprazole	500
Desipramine	50,000
Dextromethorphan	200,000
Diphenhydramine	10,000
Doxepine	50,000
Doxylamine	100,000
Ephedrine	250,000
Gabapentin	100,000
Hydroxyzine	25,000
Loratidine	50,000
Fexofenadine	50,000
Fluoxethine	100,000
Fluphenazine	25,000
Ibuprofen	500,000
Imipramine	40,000
Maprotiline	500,000
Metronidazole	200,000
Meta- Chlorophenyl piperazine	100,000
3,4 Methylenedioxy pyrovalerone (MDPV)	100,000
Mirtazapine	50,000
Nalbuphine	500,000
Naproxen	25,000
Nortriptyline	100,000
Norfenfluramine	10,000
Oxazepam	100,000

Non-Critical cross reactants	Tested Concentration (ng/mL)
Phencyclidine	15,000
Phenobarbital	500,000
Pregbalin	100,000
Promethazine	50,000
Propoxyphene	15,000
Ranitidine	200,000
Ropinirole	50,000
Secobarbital	500,000
Talwin(Pentazocine)	10,000
Trazadone	10,000
Verapamil	12,500

Accuracy

A total of 170 clinical samples were tested in the DRI Fentanyl Assay in a qualitative mode and the results were compared to LC-MS/MS method.

		LC-MS/MS +	LC-MS/MS -
DRI Fentanyl Assay	+	102	0
	-	4*	64

* LC-MS/MS values ranged from 2.0 to 2.4 ng/mL.

Catalog Number

Contents


Manufacturer


Temperature Limitation


Caution

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References

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