

Uromonitor® (PRTPCR25, PRTPCR50)

Product Type	Uromonitor® - Real-Time PCR kit for TERT, FGFR3 and KRAS mutations detection (#Urokit3, includes mastermix)
Species	Human
Catalog number	PRTPCR25, PRTPCR50
Batch number	See box and tubes
Shipping	Dry Ice
Storage	-20°C

Storage and stability: This product is shipped in dry ice. Upon arrival “Uromonitor® - Real-Time PCR kit for TERT, FGFR3 and KRAS mutations detection” (PRTPCR25, PRTPCR50) reagents must be stored at -20°C and are stable for up to 2 years. Once opened, each reagent is stable to 5 freeze-thaws.

Expiry: The expiration date is printed on the box and on each tube label. This product will maintain its performance until that date. Its performance is not guaranteed after the expiration date.

Quality control: The quality of “Uromonitor® - Real-Time PCR kit for TERT, FGFR3 and KRAS mutations detection” (PRTPCR25, PRTPCR50) is tested on a lot-to-lot basis. Triplicate amplification of independent DNA’s for each of the alterations ensures accuracy and specificity of the batch.

Notes: For *In Vitro* Diagnostic Use.

Product description

The **Uromonitor®** is an *in vitro* diagnostic kit (IVD) intended to detect and monitor non-muscle invasive bladder cancer (BCa) recurrence in conjunction with cystoscopy in previously diagnosed bladder cancer patients. It provides all the components for a non-invasive detection of a set of hotspot mutations in the TERT, FGFR3 and KRAS genes, highly prevalent in BCa, in DNA derived from exfoliated bladder cells present in urine. The **Uromonitor®** kit is composed of three subunits, including all the tools and reagents needed for sample processing and analysis. The **Uromonitor®** kit consists of three subunits:

1. Uromonitor® - Urine filtration kit (#Urokit1, PUF01/PUF25/PUF50)

Provides the components needed for sample collection. The principle of the procedure relies on the filtration of urine samples. Using a syringe, urine samples will pass through a filter, with a proprietary preservative where exfoliated bladder cells are trapped for subsequent procedures.

2. Uromonitor® - DNA extraction and preparation kit (#Urokit2, PADNP25/PADNP50)







Provides an efficient and fast method for the purification of high-quality cell DNA extracted from cells exfoliated in urine. Eluted purified DNA is suitable for use with Uromonitor®’s Real-Time PCR procedure.

3. Uromonitor® - Real-Time PCR kit for TERT, FGFR3 and KRAS mutations detection (#Urokit3, PRTPCR25/PRTPCR50)

Provides an efficient method for the detection of mutations in the TERT promoter (-124 and -146), FGFR3 codons 248, 249, 372 and 375, and KRAS codons 12/13 and 61, through a highly sensitive and specific Real-Time PCR method. It includes a mastermix and 4 independent assays that use allele-specific primers in multiplex reactions to identify the presence of TERTp, FGFR3 and KRAS mutations in a total of 9 reactions per sample (see table below). Each reaction contains primer/probes sets for mutations detection, as well as internal control (IC), negative and positive DNA controls for sample validation and qualitative analysis.

	Target	Reporter
●	TERT_124_Mut	FAM
	TERT_124_IC	HEX
	TERT_146_Mut	FAM
	TERT_146_IC	HEX
●	FGFR3_248_Mut	FAM
	FGFR3_249_Mut	FAM
	FGFR3_IC	FAM
●	FGFR3_372_Mut	FAM
	FGFR3_372_IC	HEX
	FGFR3_375_Mut	FAM
	FGFR3_375_IC	HEX
●	KRAS_12/13_Mut	FAM
	KRAS_12/13_IC	HEX
	KRAS_61_Mut	FAM
	KRAS_61_IC	HEX

Kit components (#Urokit3, PRTPCR25/PRTPCR50)

Assay	Component	PRTPCR25	PRTPCR50
TERT 124/146 	RTA-124/146	58 µL	116 µL
	RTB-124/146	58 µL	116 µL
	R1-124	13 µL	26 µL
	R2-124	13 µL	26 µL
	R1-146	13 µL	26 µL
	R2-146	13 µL	26 µL
FGFR3 248/249 	RFA-248/249	130 µL	260 µL
	RFB-248/249	39 µL	78 µL
	R1-248	17 µL	34 µL
	R2-248	17 µL	34 µL
	R1-249	17 µL	34 µL
	R2-249	17 µL	34 µL
	R1-IC	17 µL	34 µL
	R2-IC	17 µL	34 µL
FGFR3 372/375 	RFA-372	29 µL	58 µL
	RFB-372	29 µL	58 µL
	R1-372	13 µL	26 µL
	R2-372	13 µL	26 µL
	RFA-375	29 µL	58 µL
	RFB-375	29 µL	58 µL
	R1-375	13 µL	26 µL
	R2-375	13 µL	26 µL
KRAS 12/13/61 	RKA-12/13	29 µL	58 µL
	RKB-12/13	29 µL	58 µL
	R1-12/13	13 µL	26 µL
	R2-12/13	13 µL	26 µL
	RKA-61	29 µL	58 µL
	RKB-61	29 µL	58 µL
	R1-61	13 µL	26 µL
	R2-61	13 µL	26 µL
Negative and Positive Controls 	TERT/FGFR3/KRAS-NC	50 µL	100 µL
	TERT_124/146+	15 µL	30 µL
	FGFR3_248/249+	15 µL	30 µL
	FGFR3_372/375+	15 µL	30 µL
	KRAS_12/13/61+	15 µL	30 µL
	Mastermix	2 x 1500 µL	4 x 1500 µL
	H2O	2 x 1500 µL	4 x 1500 µL

User guidelines

- The use of this product is limited to personnel trained in Real-time PCR technique and in the use of recognized and validated Real-time PCR equipment and software's.
- The Uromonitor® (PRTPCR25/PRTPCR50) is a qualitative test. The test is not for quantitative measurements of percent mutation.
- Reliable results are dependent on adequate specimen sampling, filtration/centrifugation, transport, storage, and processing. Always follow the procedures described in the instructions.

- The presence of PCR inhibitors in the samples may lead to false negative or invalid results.
- The Uromonitor® (PRTPCR25/PRTPCR50) has been validated for use with 2.5-25 ng of DNA per replicate. DNA input amounts below 2.5 ng or above 25 ng per replicate are not recommended.
- The Uromonitor® (PRTPCR25/PRTPCR50) detects down to 3.125% mutant sequences in a background of wild-type DNA for the TERT promoter mutations.
- The Uromonitor® (PRTPCR25/PRTPCR50) detects down to 1.56% mutant sequences in a background of wild-type DNA for the FGFR3 hotspot mutations.
- The Uromonitor® (PRTPCR25/PRTPCR50) detects down to 3.125% mutant sequences in a background of wild-type DNA for the KRAS hotspot mutations.

Please, always follow the procedures described in “Uromonitor® - Real-Time PCR kit for TERT, FGFR3 and KRAS mutations detection” Instructions. Digital and printed versions of the instructions are provided with the product.

Handling requirements

- Allow reagent/controls to thaw on ice immediately before use. Keep them on ice while in use. In case reagents were not previously aliquoted for single use, store the remaining immediately at -20°C until next use.
- All reagents and mixes must be prepared and stored protected from direct light.
- To prevent DNA/RNA contamination, use disposable plasticware. Automatic pipettes and non-disposable glassware or plasticware should be sterile/DNA/RNA free and should only be used for pre-amplification procedures. Gloves should be always worn, when handling/performing the full protocol.
- Change the tip when pipetting different components during reaction mixes preparation as well as when pipetting samples/controls into a different well.
- As with any test procedure, good laboratory practices are essential for the proper performance of this assay. Due to the high analytical sensitivity of this test, care should be taken to keep reagents and amplification mixtures free from contamination.

Safety precautions

The “Uromonitor® - Real-Time PCR kit for TERT, FGFR3 and KRAS mutations detection” (#Urokit3, PRTPCR25/PRTPCR50) contains no substances which at their given concentration, are hazardous to health. We recommend handling all reagents with caution, always wear a suitable lab coat, disposable gloves, and protective glasses. For more information, please refer to Uromonitor® Material Safety Data Sheet (MSDS) available for download at www.u-monitor.eu.

Technical support

For any technical enquiries, please contact our Technical Support team via email at lab@uromonitor.com.