Check the product label for actual catalog number, lot and expiry date.

UDGin™ PCR Cleaner Mix, 20X

CAT.#	SIZE	COMPONENTS	COMPONENT COMPOSITION
UDG0101	500 r of 20 µl	0.5 ml - UDGin™ PCR Cleaner Mix, 20X	20X master mix for PCR carry-over contamination prevention, contains optimized concentration of dUTP, Uracil DNA Glycosylase (UDG), and stabilizers.

Storage *In the dark at -20°C.*

APPLICATIONS

Efficient prevention of carry-over contamination:

- in qPCR
- in PCR

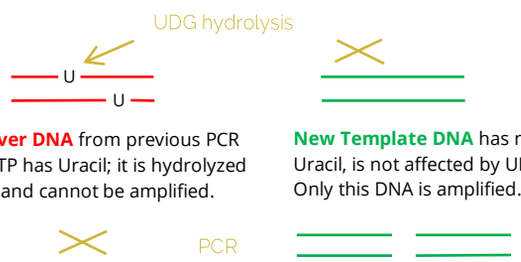
BENEFITS

- Time saving ready-to-use mix with UDG and dUTP
- Compatible with all highQu ORA™ qPCR Mixes and PCR master mixes

PRODUCT DETAILS

UDGin™ PCR Cleaner Mix is an efficient tool for carry-over contamination prevention in qPCR or PCR. It is optimized for the use in conjunction with highQu qPCR or PCR products.
UDGin™ PCR Cleaner Mix, is a ready to use 20X mix of optimally combined Uracil-DNA Glycosylase (UDG) and dUTP in a buffer with stabilizers. 1 µl of UDGin™ PCR Cleaner Mix added into each 20 µl PCR reaction prevents amplification of DNA carried over from previous PCR performed with dUTP.
During 10 minutes incubation before the PCR start, UDG hydrolyzes the N-glycosilic bond between uracil and sugar leaving apyrimidinic sites in uracil containing DNA which is then cleaved by heat during the first PCR cycle, thus only the newly added template is amplified. UDG is inactivated during the initial PCR denaturation step and does not destroy newly synthesized dUTP containing DNAs. PCR products obtained when UDGin™ PCR Cleaner Mix was used contain uracil, therefore they will be destroyed again by UDG before the next PCR start.

PCR CARRY-OVER CONTAMINATION PREVENTION



If you use UDGin™ PCR Cleaner Mix in every PCR, only the newly added template DNA will be amplified. All PCR products will contain uracil. Therefore, even if you carry them over into the next PCR they will not be amplified, as the uracil containing DNA will be destroyed during UDG treatment before each PCR. Only new added DNA containing no uracil will serve as PCR template.

Reference: Longo, M.C., et al., Use of uracil DNA glycosylase to control carry-over contamination in polymerase chain reactions, Gene 93, 125-8, 1990.

PROTOCOL

Follow the protocol of ORA™ qPCR Mix or ALLin™ PCR Enzyme or Mastermix that you use. Only three additional steps are generally required for preventing carry-over contamination in PCR using UDGin™ PCR Cleaner Mix:

1. Before adding water into PCR reactions, add 1µl of UDGin™ PCR Cleaner Mix, 20X into each 20 µl reaction. Use less water accordingly.
2. Set your qPCR/PCR instrument to perform at the beginning one cycle of 37°C incubation for 10 min.
3. After incubation always perform one cycle of longer initial denaturation (and UDG inactivation) for 5 minutes at 95°C.

Note:

- Store PCR reactions on ice.
- Before subsequent applications, consider that your PCR products contain uracil. This has no influence on electrophoresis and sequencing, but might have effect on cleavage with certain restriction enzymes (check enzyme performance on U containing sites). When cloning, use only *ung*- bacterial hosts for transformations.

Note: Use of UDG in certain countries for certain applications may be covered by patents and may require a license.

Example protocol guidelines (qPCR, dye based)

✓ Prepare a 20 µl reaction:

Reverse Primer	100 - 400 nM final c.
Forward Primer	100 - 400 nM final c.
cDNA Template *or* gDNA Template	<100ng *or* <1 µg
UDGin™ PCR Cleaner Mix, 20X	**1 µl (to final 1X conc.)**
PCR Water	to 10 µl
ORA™ Green ROX L Mix, 2X	10 µl

✓ Mix gently, avoid bubbles.

✓ Place into the instrument (SYBR® Green or FAM channel), set like:

UDG treatment	**1 cycle: 37°C - 10 min** (uracil containing DNA hydrolysis)
Initial denaturation	**1 cycle: 95°C - 5 min** (DNA denaturation, UDG inactivation, Hot-start Polymerase activation)
Denaturation	40 cycles: 95°C - 5 sec
Annealing/extension	40 cycles: 60-65°C – 20-30 sec

Follow instrument instructions for melting curve analysis.

IN VITRO RESEARCH USE ONLY

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