**Uracil Glycosylase Inhibitor (UGI)**

**Application:** Uracil Glycosylase Inhibitor (UGI) inhibits uracil-DNA glycosylase (UDG). Since UDG remains partially active following heat treatment at 95°C, UGI can be used to prevent subsequent degradation of product DNA.

Supplied in: 50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM DTT, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C).

**Reagents Supplied with Enzyme:**
10X Uracil DNA Glycosylase Buffer.

**Reaction Conditions:** 1X Uracil DNA Glycosylase Buffer. Incubate at 37°C.

**Quality Control Assays**

- **Exonuclease Activity:** Incubation of 50 units for 4 hours at 37°C in 50 µl of assay NEBuffer 1 with 1 µg [3 H] DNA (10^5 cpm/µg) released < 0.1% radioactivity.

**Unit Definition:** One unit of UGI is defined as the amount of protein required to inhibit one unit of *E. coli* UDG in 1 hour at 37°C in a total reaction volume of 50 µl. One unit of UDG is the amount of enzyme which will catalyze the release of 60 pmol of uracil per minute from double-stranded, uracil-containing DNA.

**Unit Assay Conditions:** 1X Uracil DNA Glycosylase Buffer, 1 unit of Uracil DNA Glycosylase, 0.2 µg [3 H]-uracil DNA (10^4-10^5 cpm/µg) for 30 minutes at 37°C in a total reaction volume of 50 µl.

**Endonuclease Activity:** Incubation of 50 units of UGI with 1 µg of φX174 RF I DNA for 4 hours at 37°C in 50 µl NEBuffer 1 resulted in < 5% conversion to RF II.

**Heat Inactivation:** No

**Note:** UGI is extremely thermostable, retaining > 95% activity after boiling for 10 minutes.

**References:**