



T4 gp32 Protein

Manual

Catalog #	3511	3512	3515	3518	3516
Package Size	100μg	200µg	500µg	1000µg	500μg
Volume	20μΙ	40μl	100μΙ	200μΙ	50μΙ
Concentration	5μg/μl				10μg/μl



Important!

-20°C Storage Required

- * Immediately inspect packages
- * Freeze upon receipt

India Contact:

Life Technologies (India) Pvt. Ltd. Ph: +91-11-42208000, 42208111, 42208222 Mobile: +91-9810521400

Email: customerservice@lifetechindia.com

www.lifetechindia.com

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Description:

Intact Genomics (ig®) T4 gp32 is a single-stranded DNA binding protein required for T4 DNA replication, recombination, and repair (1, 2). It improves the efficiency of reverse transcriptase (RT) during RT-PCR (3), enhances T4 DNA polymerase activity (4), as well as increases the yield of PCR products (5).

Protein Purity:

The physical purity of this enzyme is ≥98% as assessed by SDS-PAGE with Coomassie® blue staining (Fig. 1).

Product Source:

E. coli BL21 (DE3) strain expressing T4 gp32 gene.

Product Components:

- T4 gp32 protein
- 10X gp32 reaction buffer

1x GP 32 Reaction Buffer Composition:

20 mM Tris-acetate100 mM Potassium acetate 10 mM Magnesium acetate, 1 mM DTT pH 7.8 @ 25°C

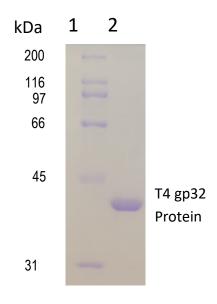


Fig. 1: Lane 1. Protein Marker Lane 2. gp32 Protein

Storage:

50 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA 50% Glycerol, pH 7.5 @ 25°C

Heat Inactivation:

65ºC for 20 min



Enzyme Concentration:

IG uses orthogonal, 3-part approaches to determine the enzyme concentration to provide you with consistent and reliable enzymes for your needs. The quantity of a protein sample is assessed using densitometry with polyacrylamide gel electrophoresis (PAGE), UV absorbance spectra of native protein, and using a protein standard assay such as bicinchoninic acid assay (BCA) using bovine serum albumin (BSA) as a standard (Figure 1).

Why does IG use all three approaches?

- Each method above has limitations. The limitations include experimental noise, accuracy, and susceptibility to buffer and/or enzyme conditions.
- 2. Each enzyme has unique physical properties that make a single approach to analyzing proteins a challenge. Each enzyme has a different protein sequence, different requirements to be stable in solution, and different requirements to retain its maximal activity. These differences can interfere with or convolute results, especially when compared to other enzymes. When used together, however, each method provides the scientist with independent measures of both enzyme and buffer purity and quality.

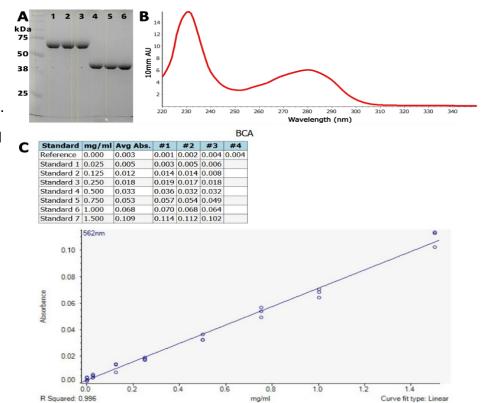


Figure 1: Enzyme quantitation methods used by IG. A) SDS-polyacrylamide gel electrophoresis. Ladder in 1st lane, 2 μ g BSA (~67 kDa) as a standard in lanes

1-3, and IG enzymes (~40 kDa) in lanes 4-6. The yellow boxes are the areas evaluated by densitometry. The integrated band intensities of IG enzymes are compared with integrated band intensities from BSA to assay concentration. B) UV spectrum of a clean IG enzyme with protein peaks at 230 nm and at 280 nm. An extinction coefficient at 280 nm is typically used to quantify protein using these spectra with buffer subtraction at 330 nm. C) BCA standard curve for BSA. The curve is used to calculate an IG enzyme concentration using BSA as the standard.



Quality Control:

T4 gp32 Protein is free from detectable nuclease activities.

References:

- 1. Chase, J. W. and Williams, K. R. (1986) Ann. Rev. Biochem. 55, 103-136
- 2. Sinha, N. K. and Snustad, D. P. (1971) J. Mol. Biol. 62, 267-271.
- **3.** Baugh, L.R. et al. (2001). Nucl. Acids Res. 29, e29.
- 4. Topal, M.D. and Sinha, N.K. (1983). J. Biol. Chem. 258, 12274-12279.
- 5. Schwartz, K. et al. (1990). Nucl. Acids Res. 18, 1079.

Related Products:

- T4 UvsX DNA Recombinase (Cat.# 3562)
- T4 UvsY Protein (Cat.# 3572)
- Bsu DNA Polymerase (Cat.# 3585)
- Sau DNA Polymerase (Cat.# 3595)
- Exonuclease III (Cat.# 3415)
- Exonuclease IV (Nfo) (Cat.# 3425)

Ordering Information:

- Order online within the USA. Place orders on www.intactgenomics.com using our secure Shopping Cart.
- Order by email, phone, or fax.

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Intact Genomics, Inc.

11840 Westline Industrial Drive, Suite 120, St. Louis, MO. 63146, USA

Phone: (314) 942-3655 | Toll-free: 855-835-7172 | Fax: (314) 942-3656

Email: sales@intactgenomics.com | ig@intactgenomics.com

Website: www.intactgenomics.com



India Contact: Life Technologies (India) Pvt. Ltd. Ph: +91-11-42208000, 42208111, 42208222 Mobile: +91-9810521400 Email: customerservice@lifetechindia.com

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