

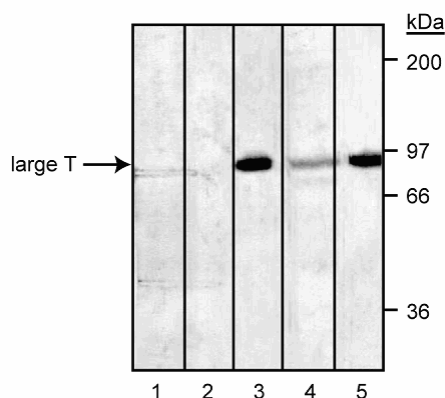
Technical Data Sheet

Purified Mouse Anti-SV40 Large T Antigen**Product Information**

Material Number:	554149
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	PAb 101
Immunogen:	SV40-transformed BALB/c mouse cell lines
Isotype:	Mouse IgG2a
Reactivity:	Viral-transformed cells
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

Description

Simian virus 40 is a small DNA virus encoded by 5.2 kb of double-stranded DNA. SV40 large T antigen (T-ag) is a multifunctional ~85 kD phosphoprotein, which is the sole viral protein required for SV40 replication. All other factors are provided by the infected host cell. In addition to its role in SV40 DNA replication, T-ag also causes transformation of susceptible cell lines. Studies of various mutant T-ag proteins have shown that the replication and transformation fractions of T-ag can be separated. The multifunctional nature of this protein has resulted in its use as a model system in a wide variety of disciplines. SV40 T-ag exercises negative regulation on the transcription of SV40 early mRNA by feedback inhibition and exerts positive regulation on transcription from the late promoter. In addition to transcriptional regulation, T-ag is involved in viral DNA replication. Specific biochemical functions required for DNA synthesis that are inherent to the T-ag include high-affinity binding to sites within the viral origin of DNA synthesis, and ATPase and helicase activities. Other functions attributed to T-ag include cellular transformation, induction of cellular DNA synthesis, induction of rRNA synthesis and provision of a host-range function for viral replication. However, all functions of T-ag are influenced by a wide range of post-translational modifications including phosphorylation, glycosylation, acetylation, acylation and adenylation. T-ag exists in monomeric as well as polymeric forms and associates with the tumor suppressor proteins p53 and Rb (retinoblastoma protein). Most of T-ag is transported to the nucleus, while a small fraction is localized at the cell surface. PAb 101 recognizes a C-terminal epitope within the last 190 amino acids of T-ag. PAb 101 was originally known as Clone 7. Studies have suggested that PAb 101 binds the strongest to mature T-ag. PAb 101 (i.e., Clone 7) was developed along with a panel of monoclonal antibodies where SV40-transformed BALB/c mouse cell lines (SVT2 or B4) were used as immunogens. The specificity of the antibody was originally characterized by a variety of techniques using SV40-infected and SV40-transformed cells.



Western blot analysis of large T in COS-7 cells using a panel of monoclonal antibodies. Large T was identified by PAb 101 (lane 3, Cat. No. 554149), 108 (lane 4) and 114 (lane 5). Mouse IgG2a (lane 1) and PAb 100 (lane 2) were used as negative controls. Note that PAb 100 can be used for immunoprecipitation, but not for western blotting of large T.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4°C.

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Application Notes

Application

Western blot	Routinely Tested
Immunoprecipitation	Reported
Immunofluorescence	Reported
Immunohistochemistry-formalin (antigen retrieval required)	Reported

Recommended Assay Procedure:

T-ag purified with PAb 101 has been shown to retain helicase activity. T-ag is immunoprecipitated as a single or multiple bands between about 80-95 kD depending on post-translational modifications. SV40-transformed cells such as COS-7 (ATCC CRL 1651) are suggested as positive controls. Any cell line that is not SV40-transformed or SV40-infected can be used as a negative control.

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

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