# Product Information Sheet for ARP-2850

## *Toxoplasma gondii* Transformation Vector (pminCAT/HXGPRT+), Recombinant in *Escherichia coli*

## Catalog No. ARP-2850

This reagent is the tangible property of the U.S. Government.

## Lot No. 95030

## Manufacturing Date: Unknown; before 1998

## For research use only. Not for human use.

### Contributor:

David S. Roos, E. Otis Kendall Professor of Biology, Department of Biology, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA and Bruce K. Brown, Project Director, National Institutes of Health AIDS Reagent Program, Germantown, Maryland, USA

### Manufacturer:

David S. Roos, E. Otis Kendall Professor of Biology, Department of Biology, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA

### Product Description:

ARP-2850 is an *Escherichia coli (E. coli)* DH5α stock expressing a transformation vector clone, pminCAT/HXGPRT+, containing a chloramphenicol acetyltransferase (CAT) coding sequence (GenBlock cassette; Pharmacia) flanked by 5' and 3' sequences derived from the *Toxoplasma gondii* dihydrofolate reductase-thymidylate synthase *(T. gondii dhfr-ts)* gene cloned into Bluescript pKS+ (Stratagene) polylinker *Eco*RV-*Sma*I sites.[1] Both *Eco*RV-*Sma*I sites were destroyed during the cloning process. A unique *Bgl*II site is located upstream of CAT, and a unique *Pst*I site is downstream. A *T. gondii* hypoxanthine-xanthine-guanine phosphoribosyl transferase (HXGPRT) minigene containing 5' genomic flanking sequences fused to cDNA encoding isoform I was cloned into the polylinker *Xho*I site upstream of CAT and in the same orientation. The *Xho*I fragment also contains some polylinker sequences from pGEM (Promega).

ARP-2850 is suitable for transient expression of CAT in transfected *T. gondii* parasites and/or stable transformation of HXGPRT-deficient *T. gondii* host strains (ARP-2857 and ARP-2860). The HXGPRT gene can be removed as a ~ 2000 base pair *Xho*I fragment for use as a selectable marker in other vectors; clone pminiHXGPRT (ARP-2855) works slightly better for this purpose. Clone pminCAT/HXGPRT+ is identical to pminCAT/HXGPRT- (ARP-2851) except for the orientation of the HXGPRT-containing *Xho*I fragment. Clone pminCAT/HXGPRT+ exhibits higher transformation frequencies than ARP-2851, however, possibly because of the strong hairpin in ARP-2851 produced by the polylinker sequences introduced with the HXGPRT minigene.

### Material Provided:

Each vial contains approximately 500 µL of *E. coli* DH5α with pminCAT/HXGPRT+ in Luria Bertani (LB) broth supplemented with 10% glycerol.

### Packaging/Storage:

ARP-2850 was packaged aseptically in plastic cryovials. The product is provided frozen on dry ice and should be stored at -60°C or colder immediately upon arrival. Freeze-thaw cycles should be minimized.

### Growth Conditions:

Media:
LB broth or agar
pminCAT/HXGPRT+ contains the gene required for chloramphenicol resistance. The standard concentration of chloramphenicol in culture is 20 µg per mL.
Incubation:
Temperature: 37°C
Atmosphere: Aerobic
Propagation:
Incubate the tube, slant and/or plate at 37°C for 1 day.

### Citation:

Acknowledgment for publications should read "The following reagent was provided by the NIH AIDS Reagent Program for distribution by BEI Resources, NIAID, NIH: *Toxoplasma gondii* Transformation Vector (pminCAT/HXGPRT+), Recombinant in *Escherichia coli*, ARP-2850, contributed by Dr. David S. Roos."

### Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

### Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at www.beiresources.org.

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

**BEI Resources**
www.beiresources.org
E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI Resources are not liable for damages arising from the misidentification or misrepresentation of products.

## Use Restrictions:

**This material is distributed for internal research, non-commercial purposes only.** This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

## References:

1. Roos, D. S., et al. "Molecular Tools for the Genetic Dissection of the Protozoan Parasite *Toxoplasma gondii*." Methods Cell Biol. 45 (1994): 27-63. PubMed: 7707991.
2. Pfefferkorn, E. R. and S. E. Borotz. "*Toxoplasma gondii*: Characterization of a Mutant Resistant to 6-Thioxanthine." Exp. Parasitol. 79 (1994): 374-382. PubMed: 7957757.



Support Provided by NIAID

**BEI Resources**
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898