

Ames Tester Strains

Art. No. Strain

PSS-0110 TA98

PSS-0111 TA100

PSS-0112 TA1535

PSS-0113 TA1537

PSS-0114 TA97a

PSS-0115 E.coli WP2 *uvrA*

PSS-0116 E.coli WP2 pKM101

PSS-0118 TA1535pSK1002

PSS-0119 E.coli WP2 *uvrA*[pKM101]

PSS-0120 TA102

PSS-0121 YG1041

PSS-0122 YG1042

PSS-0123 YG7108

For research Use Only. Not for Use in Diagnostic Procedures or in humans. For In Vitro Use Only.

Intended Use

The Ames Tester Strains are designated to be used in the Bacterial Reverse Mutation assay, also referred to as Ames Test for mutagenicity assessment. The Ames strains have been applied for the detection of mutagenic and potentially carcinogenic chemicals or pharmaceuticals for > 30 years according to Bruce Ames [1].

This product is for research use only and must not be used for diagnosis or treatment in humans or animals.

Reagent

Salmonella typhimurium and Escherichia coli strains include the following genotypes:

Strain	Mutation	Type	Target	Cell Wall	Repair	pKM101	pAQ1	pYG233	pYG620
TA98	<i>hisD3052</i>	Frameshift	GGCGGCGC	<i>rfa</i>	<i>uvrB</i>	✓	-	-	-
TA100	<i>hisG46</i>	BP substitution	GGG	<i>rfa</i>	<i>uvrB</i>	✓	-	-	-
TA1535	<i>hisG46</i>	BP substitution	GGG	<i>rfa</i>	<i>uvrB</i>	-	-	-	-
TA1537	<i>hisC3076</i>	Frameshift	+1 frameshift near C-C-C run	<i>rfa</i>	<i>uvrB</i>	-	-	-	-
TA102	<i>hisG428</i>	BP substitution	TAA	<i>rfa</i>	<i>uvrB</i>	✓	✓	-	-
TA97a	<i>hisD6610</i>	Frameshift	GGGGGG	<i>rfa</i>	<i>uvrB</i>	-	-	-	-
E.coli WP2 <i>uvrA</i> [pKM101]	<i>trpE65</i>	BP substitution	A.T	-	<i>uvrA</i>	✓	-	-	-
E.coli WP2 <i>uvrA</i>	<i>trpE65</i>	BP substitution	A.T	-	<i>uvrA</i>	-	-	-	-
pKM101	<i>trpE65</i>	BP substitution	A.T	-	-	✓	-	-	-
YG1041	<i>hisD3052</i>	Frameshift	GGCGGCGC	<i>rfa</i>	<i>uvrB</i>	✓	-	✓	-
YG1042	<i>hisG46</i>	BP substitution	GGG	<i>rfa</i>	<i>uvrB</i>	✓	-	✓	-
YG7108	<i>hisG46</i>	BP substitution	GGG	<i>rfa</i>	<i>uvrB</i>	-	-	-	✓

The Salmonella typhimurium and Escherichia coli strains used in the Ames tests and listed in the guideline OECD 471 derive from the LT2 strain and WP2 strain, respectively; and they do not contain any genes from other organisms except for the species Salmonella typhimurium and Escherichia coli, respectively. The mutations in the different strains were obtained by bacterial conjugation between the same species. The transfer of genetic material is through bacterial conjugation between the same species, and the Ames tester strains are therefore considered to be non-GMOs [2].

rfa mutation

This mutation leads to a defective lipopolysaccharide (LPS) layer that coats the cell surface, making the bacteria more permeable to bulky chemicals and non-pathogenic [1].

uvrB/uvrA mutations

The *uvrB/uvrA* deletion mutation eliminates the accurate excision repair mechanism, thereby allowing more DNA lesions to be repaired by error-prone DNA repair mechanisms. Deletion through the biotin gene makes the bacteria biotin-dependent.

Plasmid pKM101

pKM101 is derived from its clinically isolated parent R46 plasmid by an in vivo 14-kb deletion, carrying one drug resistance gene for Ampicillin [3]. R plasmids have a self-transmissible nature and pKM101 is a naturally occurring plasmid normally present in the members of the family Enterobacteriaceae including Salmonella and Escherichia [4]. The pKM101 plasmid included in the Ames tester strains is important for the enhancement of chemical and UV-induced mutagenesis via an error-prone recombinational DNA repair pathway.

Plasmid pAQ1 and Plasmid pSK1002

In addition to the pKM101plasmid, the S. typhimurium TA102 strain carries the plasmid pAQ1, which is a derivative of pBR322 and carries the target DNA sequence for reversion, *hisG428*, a part of the histidine biosynthetic operon originated from S. typhimurium. Consequently, pAQ1 is a self-cloned gene. The plasmid pAQ1 in the Ames tester strains does therefore not disturb the biological diversity of S. typhimurium, and pAQ1 plasmid can be generated via self-cloning technology and transferred to S. typhimurium LT2.

Plasmid pSK1002 is a pBR322 derivative that, in addition to the gene for β -lactamase (ampicillin resistance), contains genes *lacY* and *lacZ*, *umuC* and *umuD* from E.coli K12. The construct contains a merger of the *lacZ* gene to *umuC* of the *umu* operon, which consists of promoter and operator as well as the *umuC* and *umuD* genes. It is controlled by the SOS genes *recA* (co-protease) and *lexA* (repressor). After DNA damage, it is expressed more strongly and is involved in an imprecise DNA repair process.

Plasmid pYG233

YG1041 and YG1042 strains contain an additional plasmid pYG233, which includes genes encoding the two enzymes nitroreductase and acetyltransferase. The plasmid also confers resistance to kanamycin [5].

Plasmid pYG620

YG7108 Salmonella strain contains the plasmid pYG620, which encodes the E.coli O6-methylguanine DNA Methyltransferase [6].

Reconstitution and Preparation

The strains are reconstituted with Growth Medium (available from Xenometrix (art.no. PMM-GM00) and grown overnight in a shaking incubator at 37 °C with or without antibiotics depending on the Ames Tester Strain. A detailed protocol is delivered with each strain.

Certificate of Analysis

Each batch of strains is quality controlled, and a certificate of analysis can be obtained from info@xenometrix.ch.

Shipment and Storage Conditions

The Ames Tester Strains are validated for shipment at room temperature for 10 days. The product is shipped with cool packs.

References

- [1] Maron DM et al. *Mutat Res.* 1983;113:173–215. Revised methods for the Salmonella mutagenicity test.
- [2] Kei-ichi Sugiyama et al. *Genes Environ.* 2016; 38: 1-3. The strains recommended for use in the bacterial reverse mutation test (OECD guideline 471) can be certified as non-genetically modified organisms
- [3] Winans SC et al. *J Bacteriol.* 1985;161:402–10. Conjugal transfer system of the IncN plasmid pKM101.
- [4] Langer PJ et al. *Mol Gen Genet.* 1981;182:268–72. Restriction endonuclease cleavage map of pKM101: relationship to parental plasmid R46.
- [5] Hagiwara Y, Watanabe M, Oda Y, Sofuni T, Nohmi T. Specificity and sensitivity of Salmonella typhimurium YG1041 and YG1042 strains possessing elevated levels of both nitroreductase and acetyltransferase activity. *Mutat Res.* 1993 Jun;291(3):171-80. doi: 10.1016/0165-1161(93)90157-u. PMID: 7685058.
- [6] Yamada M, Sedgwick B, Sofuni T, Nohmi T. Construction and characterization of mutants of Salmonella typhimurium deficient in DNA repair of O6-methylguanine. *J Bacteriol.* 1995 Mar;177(6):1511-9. doi: 10.1128/jb.177.6.1511-1519.1995. PMID: 7883708; PMCID: PMC176767.

Available Kit Systems with Agar Plates

- MacroAmes1 98/100, 2 strains, art.no. R02-210-S2-P
- MacroAmes1 98/100, 2 strains, art.no. W02-210-S2-P (without components for min. Glucose Plates)
- MacroAmes1 Penta 3, 5 strains, art.no. S02-514-S2-P
- MacroAmes1 Penta 3, 5 strains, art.no. X02-514-S2-P (without components for min. Glucose Plates)
- MicroAmes6 98/100, 2 strains, art.no. L02-210-S2-P2

Ames Tester Strains IFU V 2.0 Short Protocol

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