

## Strain Restriction Phenotypes

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Strain	<i>mcrA</i>	<i>mcrBC</i>	EcoK	
			R	M
C600	-	+	+	+
C600 <i>hfl</i>	-	+	+	+
DH5 $\alpha$ <sup>TM</sup>	+	+	-	+
DH10B	-	-	-	-
HB101	+	-	-	-
JM109	(-)	(+)	-	+
JM109(DE3)	(-)	(+)	-	+
KW251	-	-	-	+
LE392	-	+	-	+
NM538	-	+	-	+
NM539	-	+	-	+
Stbl2 <sup>TM</sup>	-	-	-	-
Stbl4 <sup>TM</sup>	-	-	-	-
SURE <sup>®</sup>	-	-	-	-
TOP10	-	-	-	-
Y1089	(-)	+	+	+
Y1090	(-)	+	+	+

#### Key:

- ( ) = phenotype inferred from that of ancestral or descendant strains
- + = wildtype
- = mutant

Bacteria possess restriction and modification systems that allow them to distinguish between foreign DNA and the host cell's own genome. These systems use combinations of methylases and restriction enzymes and result in the destruction of foreign DNA. Restriction/modification systems can cause problems when a vector containing DNA from another organism is introduced into a bacterial host. Many mutant bacterial strains that have defective restriction systems are now available to counteract this problem.

There are two *mcr* (methyl-cytosine restricting) systems in *E. coli*—*mcrA* and *mcrBC*. Another system, *mrr* (modified adenine recognition and restriction), restricts particular DNA sequences that include methyladenine (m6A) or methylcytosine (m5C) residues. Most laboratory strains are positive for this system. In general, the *mcrA*, *mcrBC* and *mrr* methylation-restricting systems are sequence-specific and attack DNA only when it is methylated at these specific sequences. The EcoK I restriction system attacks DNA that is not protected by adenine methylation at the appropriate recognition site. The DNA is modified by CpG methylases (M. Sss I) to contain methylcytosine in CpG dinucleotides, and a variety of adenine methylases, which modify adenine nucleotides, also exist. The Dam (DNA adenine methyltransferase) enzyme modifies GATC sequences, while the Dcm (DNA cytosine methyltransferase) enzyme modifies CC(A/T)GG sites. The *mcrA*, *mcrBC* and *mrr* systems do not restrict DNA modified at *dcm* sites, while the *mrr* system does not restrict DNA methylated at *dam*, EcoK I or EcoR I sites (1).

Mammals, higher plants and many prokaryotes contain methylcytosine in their genomic DNA (2). To increase the recovery of such genomic sequences from propagation in bacteria after cloning experiments, a strain lacking the *mcr* and *mrr* systems should be used.

#### References

1. Neidhardt, F.C. *et al.* (1996) *Escherichia coli and Salmonella Cellular and Molecular Biology*, ASM Press, Washington, D.C.
2. Woodcock, D.M. *et al.* (1989) *Nucl. Acids Res.* **17**, 3469.