

CLAIMS

1. A recombinant UvsX comprising an amino acid sequence that is at least 60%, 70%, 80%, 90%, 95%, 99% identical to SEQ ID NO: 1, which recombinant UvsX comprises an amino acid substitution mutation at the position functionally equivalent to Pro256 in the RB49 UvsX amino acid sequence, preferably in which the substitution mutation comprises a mutation to a charged residue, more preferably in which the substitution mutation comprises a mutation to a basic residue, most preferably in which the substitution mutation comprises a mutation to a lysine residue.
2. A recombinant UvsX according to Claim 1, which comprises an amino acid sequence that is at least 80% identical to SEQ ID NO: 1.
3. A recombinant UvsX comprising the amino acid sequence of any one of SEQ ID NOs: 2 and 22 to 35.
4. A recombinant UvsX comprising a substitution mutation to the semi-conserved domain comprising the amino acid sequence of any of SEQ ID NOs: 3 to 5 in which the substitution mutation comprises a mutation selected from a substitution at position 7 to any residue other than Phe, Pro, Asp, Glu or Asn, preferably in which the substitution mutation comprises a mutation to a charged residue, more preferably in which the substitution mutation comprises a mutation to a lysine residue.
5. A recombinant UvsX comprising a substitution mutation to the semi-conserved domain comprising the amino acid sequence of SEQ ID NO: 6 or 7 in which the substitution mutation comprises a mutation selected from a substitution at position 12 to any residue other than Phe, Pro, Asp, Glu or Asn, preferably in which the substitution mutation comprises a mutation to a charged residue, more preferably in which the substitution mutation comprises a mutation to a lysine residue.
6. A recombinant UvsX according to any preceding claim, in which the recombinant UvsX further comprises a substitution mutation at a position functionally equivalent to His63 in the RB49 UvsX amino acid sequence, preferably in which the substitution mutation comprises a mutation to a serine residue.
7. A recombinant UvsX according to any preceding claim, in which the recombinant UvsX further comprises a mutation selected from the group consisting of: the addition of one or

more glutamic acid residues at the C-terminus; the addition of one or more aspartic acid residues at the C-terminus; and a combination thereof.

8. A recombinant UvsX according to any of preceding claim, in which the recombinant UvsX is derived from a myoviridae phage selected from the group consisting of: T4, T6,
5 Rb69, Aeh1, KVP40, Acinetobacter phage 133, Aeromonas phage 65, cyanophage P-SSM2, cyanophage PSSM4, cyanophage S-PM2, Rb32, Vibrio phage nt-1, Rb16, Rb43, Rb49, T2, Rb14, Aeromonas phage 25, phi-1, Phage 31, phage 44RR2.8t, phage Rb3, and phage LZ2.
9. A nucleic acid molecule encoding recombinant UvsX as defined in any preceding claim, preferably in which the nucleic acid molecule is part of an expression vector, more preferably
10 in which the nucleic acid molecule is part of an expression vector contained in a host cell.
10. A recombinase polymerase amplification process of amplification of a target nucleic acid molecule, comprising the steps of: (a) contacting the recombinant UvsX according to any of Claims 1 to 8 with a first and second nucleic acid primer to form a first and second
15 nucleoprotein primer, in which the nucleic acid primer comprises a single stranded region at its 3' end; (b) contacting the first and the second nucleoprotein primer to the target nucleic acid molecule thereby forming a first double-stranded structure at a first portion of the first strand and forming a second double stranded structure at a second portion of the second strand such that the 3' ends of the first nucleic acid primer and the second nucleic acid primer are oriented toward one another on the same double-stranded template nucleic acid molecule; (c)
20 extending the 3' end of the first and second nucleic acid primer with one or more polymerases and dNTPs to generate a first and second double-stranded nucleic acid and a first and second displaced strands of nucleic acid; and (d) continuing the reaction through repetition of (b) and (c) until a desired degree of amplification is reached.
11. A process according to Claim 10, in which the target nucleic acid molecule comprises
25 double stranded nucleic acid, and/or a single stranded adaptor region.
12. A process according to Claim 10, in which the process is performed in the presence of a recombinase loading protein, preferably in which the recombinase loading protein is selected from the group consisting of T4 UvsY, *E. coli* recO, *E. coli* recR, or a combination thereof.

13. A process according to Claim 10, in which the process is performed in the presence of a single strand stabilizing agent selected from the group consisting of gp32, *E. coli* SSB protein, T4 gp32 protein, and derivatives thereof.
14. A process according to Claim 10, in which the process is performed in the presence of a crowding agent selected from the group comprising polyethylene glycol, polyethylene oxide, polystyrene, Ficoll, dextran, PVP, and albumin such that the crowding agent stimulates amplification.
15. A process according to Claim 10, in which the process is performed on an array of amplification sites, preferably in which each amplification site comprises a plurality of amplification primers for amplification of the target nucleic acid.
16. A process according to Claim 15, in which the array of amplification sites comprises an array of features on a surface, preferably in which the features are non-contiguous and are separated by interstitial regions of the surface that lack the amplification primers.
17. A process according to Claim 15, in which the array of amplification sites comprises beads in solution, beads on a surface or an emulsion.
18. A process according to Claim 10, in which the process occurs isothermally.
19. A kit for performing a recombinase polymerase reaction comprising: a recombinant UvsX as defined in any of Claims 1 to 8 and one or more of the following:
- a single stranded DNA binding protein;
 - a DNA polymerase;
 - dNTPs or a mixture of dNTPs and ddNTPs;
 - a crowding agent;
 - a buffer;
 - a reducing agent;
 - ATP or ATP analog;
 - a recombinase loading protein;
 - a first primer and optionally a second primer.