Claims

1. A polypeptide selected from the group: B-GEn.1 according to SEQ ID NO: 1 or SEQ ID NO: 39, and B-GEn.2 according to SEQ ID NO: 2, any polypeptide sequence that is at least 80% identical to any of the above, or any nucleic acid encoding the same.
2. A composition comprising a polypeptide according to claim 1 and a sgRNA comprising a sequence selected from SEQ ID NOs: 40, 41, 42, 43, and 45.
3. A composition comprising

    (i) a polypeptide according to claim 1, and

    (ii) one or more single heterologous guide RNA(s) (sgRNA) or DNA(s) that allow the generation of such one or more sgRNA(s) *in situ,* each sgRNA or DNA encoding an sgRNA comprising:

    a. an engineered DNA targeting segment that can hybridize to a target sequence in a polynucleotide locus,
    b. a tracr mate sequence, and
    c. a tracr RNA sequence,

    wherein the tracr mate sequence can hybridize to the tracr sequence, and wherein (a), (b), and (c) are arranged in a 5' to 3' orientation.

4. A composition according to claim 2 or 3, wherein the engineered DNA targeting segment is on its 3' end directly adjacent to the PAM sequence on the targeted DNA segment, or such PAM sequence is part of the targeted DNA sequence in its 5' portion, whereby the PAM sequence comprises the sequence motif “DTTN” with “D” representing “A” or “T” or “G”. and “N” representing any nucleotide.

5. Method of targeting, editing, modifying, or manipulating a target DNA at one or more locations *in vitro,* the method comprising

    (i) introducing a polypeptide according to claim 1 or a nucleic acid encoding the same into the *in vitro* environment; and
    (ii) introducing one or more single heterologous guide RNA(s) (sgRNA) or DNA(s) encoding such one or more sgRNA(s) in the *in vitro* environment, each sgRNA or DNA

encoding the sgRNA comprising:

a. an engineered DNA targeting segment comprising an RNA and capable of hybridizing to a target sequence in a polynucleotide locus,
b. a tracr mate sequence comprised of RNA, and
c. a tracr RNA sequence comprised of RNA,

wherein the tracr mate sequence hybridizes to the tracr sequence, and wherein (a), (b), and (c) are arranged in a 5' to 3' orientation; and

(iii) creating one or more nicks or cuts or base edits in the target DNA, wherein the B-GEn polypeptide is directed to the target DNA by the sgRNA in its processed or unprocessed form.

6. Use of a composition comprising

i. a polypeptide according to claim 1 or a nucleic acid encoding the same; and/or
ii. one or more single heterologous guide RNA(s) (sgRNA) or DNA(s) suitable for the generation of such one or more sgRNA in situ, each comprising:
    a. an engineered DNA targeting segment comprised of RNA and capable of hybridizing to such target sequence in a polynucleotide locus,
    b. a tracr mate sequence comprised of RNA, and
    c. a tracr RNA sequence comprised of RNA,
    wherein the tracr mate sequence hybridizes to the tracr sequence, and wherein (a), (b), and (c) are arranged in a 5' to 3' orientation;

in the manufacture of a medicament for targeting, editing, modifying, or manipulating a target DNA at one or more locations in a cell.

7. A composition according to claim 3, for use in targeting, editing, modifying, or manipulating a target DNA at one or more locations in a cell.

8. A cell comprising

i. a polypeptide according to claim 1, or a nucleic acid encoding encoding the same; and
ii. one or more single heterologous guide RNA(s) (sgRNA) or DNA(s) suitable for the generation of such one or more sgRNA *in situ,* each comprising:
    a. an engineered DNA targeting segment that can hybridizing to a target sequence in a polynucleotide locus,

b. a tracr mate sequence, and
c. a tracr RNA sequence,

wherein the tracr mate sequence that can hybridize to the tracr sequence, and wherein (a), (b), and (c) are arranged in a 5' to 3' orientation.

9. A kit comprising

I. a nucleic acid encoding a B-GEn polypeptide according to claim 1, wherein the nucleic acid encoding theB-GEn is operably linked to a promoter; and

II. one or more single heterologous guide RNA(s) (sgRNA) or DNA(s) suitable for the generation of such one or more sgRNA *in situ,* each sgRNA comprising:
   a. an engineered DNA targeting segment that can hybridize to a target sequence in a polynucleotide locus,
   b. a tracr mate sequence, and
   c. a tracr RNA sequence,

wherein the tracr mate sequence can hybridize to the tracr sequence, and wherein (a), (b), and (c) are arranged in a 5' to 3' orientation.

10. A nucleic acid or any nucleic acid comprising a nucleic acid selected from SEQ ID NO: 40-44.