Claims

1. An *in vitro* or *ex vivo* method of altering a eukaryotic cell comprising

providing to the eukaryotic cell guide RNA sequence GN_{19} GUUUUAGAGCUAGAAAUAGCAAGUUAAAAUAAGGCUAGUCCGUUAUCAACUUGA AAAAGUGGCACCGAGUCGGUGCUUUU,

wherein the GN_{19} sequence is complementary to a target nucleic acid sequence and binds to the target nucleic acid sequence and

providing to the eukaryotic cell a Cas 9 protein that interacts with the guide RNA sequence, and

wherein the method does not comprise a process for modifying the germ line genetic identity of human beings and wherein the cell is not a human embryo.

2. The method of claim 1 wherein the Cas 9 protein is a Cas9 enzyme and the target nucleic acid sequence is cleaved.

3. The method of any one of claims 1 to 2 wherein the guide RNA sequence is provided to the cell by introducing to the cell a first nucleic acid encoding the guide RNA sequence,

wherein the Cas 9 protein is provided to the cell by introducing to the cell a second nucleic acid encoding the Cas9 protein, and

wherein the cell produces the guide RNA and the Cas9 protein.

4. The method of claim 3 wherein the first nucleic acid is introduced into the cell using a viral delivery method.

5. The method of claim 3 wherein the first and/or second nucleic acid are introduced into the cell by transfection.

6. The method of claim 3 wherein the first nucleic acid is introduced into the cell using an adeno-associated virus.

7. The method of any one of claims 1 to 6 wherein the Cas9 protein is a Cas9 enzyme, a Cas9 nickase, a nuclease null Cas9, a modified Cas9 or a homolog of Cas9.

8. The method of any one of claims 1 to 7 wherein the eukaryotic cell is a yeast cell, a plant cell or a mammalian cell.

9. The method of any one of claims 1 to 8 wherein the cell is a human cell.

10. The method of any one of claims 1 to 9 wherein a plurality of guide RNA sequences complementary to different target nucleic acid sequences are provided to the eukaryotic cell, wherein the guide RNA sequence binds to the different target nucleic acid sequence,

and the Cas 9 protein interacts with the plurality of guide RNA sequences.

11. The method of any one of claims 1 to 10 wherein the guide RNA is a crRNA-tracrRNA fusion.

12. The method of any one of claims 1 to 11 wherein the Cas9 protein is human codon-optimized.

13. The method of any one of claims 1 to 12 wherein guide RNA sequences complementary to target nucleic acid sequences in a locus are provided to the cell, wherein the guide RNA sequences bind to the target nucleic acid sequences and the Cas9 protein is a Cas9 enzyme that cleaves the target nucleic acid sequences and an intervening nucleic acid sequence is deleted from the locus.

14. The method of any one of claims 1 to 13 wherein the Cas9 protein is a Cas9 enzyme that cleaves the target nucleic acid sequence and a donor nucleic acid sequence provided to the eukaryotic cell is inserted into the target nucleic acid sequence.

15. The method of claim 1 wherein the Cas9 protein is nuclease null and interacts with the guide RNA sequence and binds to the target nucleic acid sequence,

wherein the Cas 9 protein has a FokI nuclease domain attached thereto,

wherein a target nucleic acid sequence is cleaved upon FokI nuclease domain dimerization of adjacent guide RNA sequence and Cas9 protein co-localization complexes.

16. The method of claim 1 wherein the Cas9 protein is nuclease null and includes a transcriptional activation or repression domain and wherein expression of a target gene is modulated.

17. The method of claim 1 wherein the Cas9 protein is nuclease null and includes a fluorescent protein and wherein the fluorescent protein is visualized.

18. The method of claim 1 wherein the Cas9 protein is nuclease null and includes a protein bound organic fluorophore, a nucleic acid bound organic fluorophore, a quantum dot, a molecular beacon, an echo probe or a multivalent ligand-binding protein domain.

19. An RNA-guided genome editing system comprising:

guide RNA sequence GN_{19} GUUUUAGAGCUAGAAAUAGCAAGUUAAAAUAAGGCUAGUCCGUUAUCAACUUGA

AAAAGUGGCACCGAGUCGGUGCUUUU, wherein the GN_{19} sequence is complementary to a target nucleic acid sequence and binds to the target nucleic acid sequence; or a first nucleic acid sequence encoding the guide RNA sequence; and

a Cas9 protein that interacts with the guide RNA sequence, or a second nucleic acid sequence encoding the Cas9 protein, wherein the Cas9 protein includes a C-terminus SV40 nuclear localization signal.

20. The RNA-guided genome editing system of claim 19 for use in gene therapy.

21. An eukaryotic cell containing the RNA-guided genome editing system of claim 19, wherein the cell is not a human germ cell and not a human embryo.