What is claimed is:

1. A method for improving the replication of a covalently closed circular plasmid comprising the following steps:
    a. providing a covalently closed circular plasmid comprising:
        i. a Pol I-dependent origin of replication, and
        ii. an insert comprising a structured DNA sequence selected from the group consisting of inverted repeat sequence, direct repeat sequence, homopolymeric repeat sequence, eukaryotic origin of replication or eukaryotic promoter enhancer sequence, wherein the structured DNA sequence is located at a distance of less than 1000 bp from the Pol I-dependent origin of replication in the direction of replication; and
    b. modifying the covalently closed circular recombinant molecule of a. such that the Pol I-dependent origin of replication is replaced with a Pol III-dependent origin of replication whereby the resultant Pol III-dependent origin of replication covalently closed circular plasmid has improved replication.

2. The method of claim 1, wherein said Pol I-dependent origin of replication is selected from the group consisting of pUC origin, pMB1 origin, and ColE1 origin.

3. The method of claim 1, wherein said Pol III-dependent origin of replication is an R6K gamma replication origin.

4. The method of claim 1, wherein said Pol III-dependent origin of replication is an R6K gamma replication origin with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 1, SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4 and SEQ ID NO: 18.

5. The method of claim 1, wherein said structured DNA sequence is selected from the group consisting of polyA repeat, SV40 origin of replication, viral LTR, Lentiviral LTR, Retroviral LTR, transposon IR/DR repeat, Sleeping Beauty transposon IR/DR repeat, AAV ITR, transposon ITR, PiggyBac transposon ITR, CMV enhancer, and SV40 enhancer.

6. The method of claim 1, wherein said improved replication is selected from the group consisting of reduced production of replication intermediates and increased plasmid copy number.

7. A method for improving the replication of a covalently closed circular plasmid comprising the following steps:
    a. providing a covalently closed circular plasmid comprising:
        i. a bacterial replication-selection region comprising a Pol I-dependent origin of replication and an antibiotic selectable marker, and
        ii. an insert comprising a structured DNA sequence selected from the group consisting of inverted repeat sequence, direct repeat sequence, homopolymeric repeat sequence, eukaryotic origin of replication, and eukaryotic promoter enhancer sequence, wherein the structured DNA sequence is located at a distance of less than 1000 bp from the Pol I-dependent origin of replication in the direction of replication; and
    b. modifying the covalently closed circular recombinant molecule of a. such that the antibiotic selectable marker is replaced with an RNA selectable marker and the Pol I-dependent origin of replication is replaced with a Pol III-dependent origin of replication, whereby the resultant Pol III-dependent origin of replication covalently closed circular plasmid has improved replication.

8. The method of claim 7, wherein said Pol I-dependent origin of replication is selected from the group consisting of: pUC origin, pMB1 origin, and ColE1 origin.

9. The method of claim 7, wherein said Pol III-dependent origin of replication is an R6K gamma replication origin.

10. The method of claim 7, wherein said Pol III-dependent origin of replication is an R6K gamma replication origin with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 1, SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4 and SEQ ID NO: 18.

11. The method of claim 7, wherein said RNA selectable marker is an RNA-IN regulating RNA-OUT functional variant with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 5, and SEQ ID NO: 7.

12. The method of claim 7, wherein said RNA selectable marker is an RNA-OUT RNA selectable marker that encodes an RNA-IN regulating RNA-OUT RNA with at least 95% sequence identity to SEQ ID NO: 6.

13. The method of claim 7, wherein said bacterial replication-selection region comprising a Pol I-dependent origin of replication and an antibiotic selectable marker is replaced with a Pol III-dependent R6K origin-RNA-OUT RNA selectable marker bacterial replication-selection region with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 16, and SEQ ID NO: 17.

14. The method of claim 7, wherein said structured DNA sequence is selected from the group consisting of polyA repeat, SV40 origin of replication, viral LTR, Lentiviral LTR, Retroviral

LTR, transposon IR/DR repeat, Sleeping Beauty transposon IR/DR repeat, AAV ITR, transposon ITR, PiggyBac transposon ITR, CMV enhancer, and SV40 enhancer.

15. The method of claim 7, wherein said improved replication is selected from the group consisting of reduced production of replication intermediates, and increased plasmid copy number.

16. An antibiotic marker free covalently closed circular recombinant DNA molecule comprising:

    a. an antibiotic marker free insert comprising a structured DNA sequence selected from the group consisting of inverted repeat sequence, direct repeat sequence, homopolymeric repeat sequence, eukaryotic origin of replication, and eukaryotic promoter enhancer sequence;
    b. a Pol III-dependent origin of replication comprising an R6K gamma replication origin with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 1, SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4 and SEQ ID NO: 18; and
    c. an RNA-OUT RNA selectable marker comprising an RNA-IN regulating RNA-OUT RNA with at least 95% sequence identity to SEQ ID NO: 6.

17. The recombinant DNA molecule of claim 16, wherein said RNA-OUT RNA selectable marker is an RNA-IN regulating RNA-OUT functional variant with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 5, and SEQ ID NO: 7.

18. The recombinant DNA molecule of claim 16, wherein said R6K gamma replication origin and said RNA-OUT RNA selectable marker comprise a R6K origin-RNA-OUT RNA selectable marker bacterial replication-selection region with at least 95% sequence identity to a sequence selected from the group consisting of SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 16, and SEQ ID NO: 17.

19. The recombinant DNA molecule of claim 16, wherein said structured DNA sequence is selected from the group consisting of polyA repeat, SV40 origin of replication, viral LTR, Lentiviral LTR, Retroviral LTR, transposon IR/DR repeat, Sleeping Beauty transposon IR/DR repeat, AAV ITR, transposon ITR, PiggyBac transposon ITR, CMV enhancer, and SV40 enhancer.

20. The recombinant DNA molecule of claim 16, wherein said recombinant DNA molecule is selected from the group consisting of viral vector, Lentiviral vector, Retroviral vector, AAV vector, Ad vector, non-viral transposon vector, Sleeping Beauty transposon vector, PiggyBac transposon vector, Tol2 transposon vector, and polyA containing mRNA vector.