Claims

1. A method for conducting primer extension, comprising: contacting a polynucleotide template with a DNA polymerase selected from the group consisting of SEQ ID NOS: 85, 87, 89, 147, 149, 151, 153, 155, 157, 159, 161, 170, 174, 176, 178 and 180, an RNase H2 enzyme, a blocked-cleavable rhPCR primer and nucleoside triphosphates; and performing a primer extension method under suitable conditions, thereby producing an extended primer.

2. The method of claim 1, wherein the primer extension method comprises a method for conducting polymerase chain reaction (PCR).

3. The method of claim 2, wherein the method for conducting PCR comprises allele-specific PCR.

4. The method of claim 2, wherein the method for conducting PCR comprises detecting a rare allele at a level of discrimination of > 1:10,000.

5. A mutant Taq DNA polymerase selected from the group consisting of SEQ ID NOS: 85, 87, 89, 147, 149, 151, 153, 155, 157, 159, 161, 170, 174, 176, 178 and 180.

6. A kit for producing an extended primer, comprising: at least one container providing a DNA polymerase selected from the group consisting of SEQ ID NOS: 85, 87, 89, 147, 149, 151, 153, 155, 157, 159, 161, 170, 174, 176, 178 and 180; a container containing an RNase H2; and a container containing a blocked-cleavable rhPCR primer.

7. A reaction mixture comprising:

a polynucleotide template;

a DNA polymerase selected from the group consisting of SEQ ID NOS: 85, 87, 89, 147, 149, 151, 153, 155, 157, 159, 161, 170, 174, 176, 178 and 180;

an RNase H2 enzyme;

at least one block-cleavable rhPCR primer; and

nucleoside triphosphates.

8. A method for performing rhPCR, comprising performing primer extension with the reaction mixture according to claim 7.

9. The method according to claim 1 or 7, wherein the blocked-cleavable rhPCR primer is selected from the group consisting of a RDDDDx blocked-cleavable rhPCR primer and a RDxxD blocked-cleavable rhPCR primer,

wherein the RDDDDx blocked-cleavable rhPCR primer consists of an oligonucleotide complementary to the polynucleotide template and having at its 3'-terminus the sequence RDDDDx, wherein R is an RNA base, D is a DNA base and x is a C3 spacer group, and

wherein the RDxxD blocked-cleavable rhPCR primer consists of an oligonucleotide complementary to the polynucleotide template and having at its 3'-terminus the sequence RDxxD, wherein R is an RNA base, D is a DNA base and x is a C3 spacer group.

10. The kit according to claim 6, wherein the blocked-cleavable rhPCR primer is selected from the group consisting of a RDDDDx blocked-cleavable rhPCR primer and a RDxxD blocked-cleavable rhPCR primer,

wherein the RDDDDx blocked-cleavable rhPCR primer consists of an oligonucleotide complementary to the polynucleotide template and having at its 3'-terminus the sequence RDDDDx, wherein R is an RNA base, D is a DNA base and x is a C3 spacer group, and

wherein the RDxxD blocked-cleavable rhPCR primer consists of an oligonucleotide complementary to the polynucleotide template and having at its 3'-terminus the sequence RDxxD, wherein R is an RNA base, D is a DNA base and x is a C3 spacer group.

11. The reaction mixture according to claim 7, wherein the at least one blocked-cleavable rhPCR primer is selected from the group consisting of a RDDDDx blocked-cleavable rhPCR primer and a RDxxD blocked-cleavable rhPCR primer,

wherein the RDDDDx blocked-cleavable rhPCR primer consists of an oligonucleotide complementary to the polynucleotide template and having at its 3'-terminus the sequence RDDDDx, wherein R is an RNA base, D is a DNA base and x is a C3 spacer group, and

wherein the RDxxD blocked-cleavable rhPCR primer consists of an oligonucleotide complementary to the polynucleotide template and having at its 3'-terminus the sequence RDxxD, wherein R is an RNA base, D is a DNA base and x is a C3 spacer group.