

Claims

We claim:

1. A method for detection of a target nucleic acid, the method comprising steps of:
contacting a CRISPR-Cas complex comprising:
 - (a) a Cas protein with collateral cleavage activity that is thermostable at temperatures above at least 56 °C, wherein the Cas protein has an amino acid sequence according to any one of SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NOs: 8-10, SEQ ID NO: 3, or SEQ ID NO: 4; and
 - a guide RNA selected or engineered to be complementary to a target nucleic acid sequence; with a sample potentially comprising the target nucleic acid sequence, wherein the step of contacting comprises contacting the CRISPR-Cas complex and sample with a reporter susceptible to cleavage by the Cas protein collateral activity; and
 - (b) amplifying nucleic acid present in the sample.
2. The method of claim 1, wherein the step of contacting comprises incubating for a period of time above the temperature.
3. The method of claim 1, wherein the step of amplifying utilizes a thermostable nucleic acid polymerase.
4. The method of claim 1, wherein the steps of amplifying and contacting are performed in a single vessel.
5. The method of claim 1, wherein the thermostable collateral cleavage activity is thermostable above a temperature of about 60°C.
6. The method of claim 1, wherein the thermostable collateral cleavage activity is thermostable above a temperature of about 65°C.