detecting a nucleic acid under visible light, the reagent is enabled to be dissolved in a solution containing an amplified nucleic acid without opening the reaction container; therefore, splashing and contamination of the sample can be prevented, and nucleic acid amplification can be easily and quickly detected under visible light. Thus, they have completed the present invention.

[0010]

Specifically, in one aspect, the present invention relates to a method of detecting a nucleic acid, comprising the steps of: contacting a nucleic acid with a detecting reagent containing a leuco dye in a closed system; wherein the leuco dye is initially colorless, and wherein the nucleic acid reacts as a color developer with the colorless leuco dye to produce a colored dye; and observing a color change of at least one of the nucleic acid and the detecting reagent under visible light. Also disclosed is a method of detecting a nucleic acid, including the steps of: contacting a nucleic acid with a detecting reagent in a closed system; and observing a color change of at least one of the nucleic acid and the detecting reagent under visible light.

[0011]

The method preferably further includes, prior to or after the contacting step, the step of amplifying the nucleic acid.

[0012]

The detecting reagent is preferably in a solid form.

[0013]

The detecting reagent preferably contains a leuco dye.

[0014]

The leuco dye is preferably a triarylmethane dye.

[0015]

The triarylmethane dye is preferably crystal violet or gentian violet.

[0016]

The detecting reagent preferably contains at least one of a nucleophilic agent and a stabilizer.

[0017]

The nucleophilic agent is preferably one or more nucleophilic agents selected from the group consisting of sodium borohydride, sodium cyanoborohydride, sodium bisulfite, sodium sulfite, sodium hydrosulfite, potassium pyrosulfite, sodium thiosulfate, glutathione, ascorbic acid, 2-mercaptoethanol, DL-dithiothreitol, 1-thioglycerol,

CLAIMS

Claim 1. A method of detecting a nucleic acid, comprising the steps of:

contacting a nucleic acid with a detecting reagent in a closed system; and

observing a color change of at least one of the nucleic acid and the detecting reagent under visible light.

Claim 2. The method of detecting a nucleic acid according to claim 1, further comprising, prior to or after the contacting step,

the step of amplifying the nucleic acid.

Claim 3. The method of detecting a nucleic acid according to claim 1 or 2,

wherein the detecting reagent is in a solid form.

Claim 4. The method of detecting a nucleic acid according to any one of claims 1 to 3,

wherein the detecting reagent contains a leuco dye.

Claim 5. The method of detecting a nucleic acid according to claim 4,

wherein the leuco dye is a triarylmethane dye.

Claim 6. The method of detecting a nucleic acid according to claim 5,

wherein the triarylmethane dye is crystal violet or gentian violet.

Claim 7. The method of detecting a nucleic acid according to any one of claims 4 to 6,

wherein the detecting reagent contains at least one of a nucleophilic agent and a stabilizer.

Claim 8. The method of detecting a nucleic acid according to claim 7,

wherein the nucleophilic agent is one or more nucleophilic agents selected from the group consisting of sodium borohydride, sodium cyanoborohydride, sodium bisulfite, sodium sulfite, sodium hydrosulfite, potassium pyrosulfite, sodium thiosulfate, glutathione, ascorbic acid, 2-mercaptoethanol, DL-dithiothreitol, 1-thioglycerol, cysteine, tributylphosphine, aminoethanethiol, and tris(2-carboxyethyl)phosphine.

Claim 9. The method of detecting a nucleic acid according to claim 7,

wherein the stabilizer is one or more stabilizers selected from the group consisting of α-cyclodextrin, β-cyclodextrin, γ-cyclodextrin, and ascorbic acid.

Claim 10. The method of detecting a nucleic acid according to claim 2,

wherein the nucleic acid is amplified by LAMP method.

Claim 11. The method of detecting a nucleic acid according to claim 10,

wherein a pyrophosphatase is used in the LAMP method.

Claim 12. The method of detecting a nucleic acid according to claim 11,

wherein the pyrophosphatase is a heat resistant pyrophosphatase.

Claim 13. A device or kit for detecting a nucleic acid, comprising:

a carrier carrying a detecting reagent; and

a lid and/or a reaction container which are to be provided with the carrier.

Claim 14. The device or kit for detecting a nucleic acid according to claim 13,

wherein the lid and/or reaction container further carries a nucleic acid amplification reagent.

Claim 15. The device or kit for detecting a nucleic acid according to claim 13 or 14,

wherein the detecting reagent is shielded from a nucleic acid by a coating material.

Claim 16. An apparatus, comprising the device or kit for detecting a nucleic acid according to any one of claims 13 to 15.

CLAIMS

Claim 1. A method of detecting a nucleic acid, comprising the steps of:

contacting a nucleic acid with a detecting reagent containing a leuco dye in a closed system; wherein the leuco dye is initially colorless, and wherein the nucleic acid reacts as a color developer with the colorless leuco dye to produce a colored dye; and

observing a color change of at least one of the nucleic acid and the detecting reagent under visible light.

Claim 2. The method of detecting a nucleic acid according to claim 1, further comprising, prior to or after the contacting step,

the step of amplifying the nucleic acid.

Claim 3. The method of detecting a nucleic acid according to claim 1,

wherein the detecting reagent is in a solid form.

Claim 4. The method of detecting a nucleic acid according to claim 1,

wherein the leuco dye is a triarylmethane dye.

Claim 5. The method of detecting a nucleic acid according to claim 4,

wherein the triarylmethane dye is crystal violet or gentian violet.

Claim 6. The method of detecting a nucleic acid according to claim 1,

wherein the detecting reagent contains at least one of a nucleophilic agent and a stabilizer that stabilizes the leuco dye.

Claim 7. The method of detecting a nucleic acid according to claim 6,

wherein the nucleophilic agent is one or more nucleophilic agents selected from the group consisting of sodium borohydride, sodium cyanoborohydride, sodium bisulfite, sodium sulfite, sodium hydrosulfite, potassium pyrosulfite, sodium thiosulfate, glutathione, ascorbic acid, 2-mercaptoethanol, DL-dithiothreitol, 1-thioglycerol, cysteine, tributylphosphine, aminoethanethiol, and tris(2-carboxyethyl)phosphine.

Claim 8. The method of detecting a nucleic acid according to claim 6,

wherein the stabilizer is one or more stabilizers selected from the group consisting of α-cyclodextrin, β-cyclodextrin, γ-cyclodextrin, and ascorbic acid.

Claim 9. The method of detecting a nucleic acid according to claim 2,

wherein the nucleic acid is amplified by LAMP method.

Claim 10. The method of detecting a nucleic acid according to claim 9,

wherein a pyrophosphatase is used in the LAMP method.

Claim 11. The method of detecting a nucleic acid according to claim 10,

wherein the pyrophosphatase is a heat resistant pyrophosphatase.

Claim 12. A device or kit for detecting a nucleic acid, comprising:

a carrier carrying a detecting reagent containing a leuco dye; wherein the leuco dye is initially colorless, and wherein the colorless leuco dye is reactable with a nucleic acid as a color developer to produce a colored dye;

a lid and/or a reaction container which are to be provided with the carrier; and

wherein the carrier is located within the lid and/or reaction container.

Claim 13. The device or kit for detecting a nucleic acid according to claim 12,

wherein the lid and/or reaction container further carries a nucleic acid amplification reagent.

Claim 14. The device or kit for detecting a nucleic acid according to claim 12,

wherein the detecting reagent is shielded from a nucleic acid by a coating material.

Claim 15. An apparatus, comprising the device or kit for detecting a nucleic acid according to claim 12 and means for automatically carrying out the detecting of the nucleic acid.

Claim 16. The method of detecting a nucleic acid according to claim 2, wherein the detecting reagent is in a solid form.

Claim 17. The method of detecting a nucleic acid according to claim 4, wherein the detecting reagent contains at least one of a nucleophilic agent and a stabilizer.