[0011] ~~Therefore, a principal object~~Embodiments of the present invention ~~is to~~ provide a microchip for a nucleic acid amplification reaction which allows high-precision analysis by a simple method.

Means for solving the Problem

[0012] ~~In order to solve the above-mentioned problem,~~ According to an aspect of the present invention, ~~provides~~ there is provided a microchip for a nucleic acid amplification reaction, ~~including~~ comprising: an entrance through which a liquid enters from the outside; a plurality of wells configured to function as reaction sites of the nucleic acid amplification reaction; and a flow channel through which the liquid entered through the entrance is fed into each of the wells, ~~in which~~ wherein a plurality of reagents needed for the reaction are laminated and anchored in a prescribed order in each of the wells, wherein an anchored layer of an oligonucleotide primer is laminated over an anchored layer of an enzyme.

In an embodiment of the microchip for the nucleic acid amplification reaction, an anchored layer of an oligonucleotide primer may be laminated over an anchored layer of an enzyme, or, in an opposite manner, an anchored layer of an enzyme may be laminated over an anchored layer of an oligonucleotide primer. Between the anchored layer of the enzyme and the anchored layer of the oligonucleotide primer, an anchored layer of a reaction buffer solute may be laminated.

[0013] ~~Also, the present invention provides a method of manufacturing a microchip for a nucleic acid amplification reaction, including a first step of laminating and anchoring a plurality of reagents needed for the reaction in a prescribed order in each of a plurality of wells configured to function as reaction sites of the nucleic acid amplification reaction, formed on a substrate layer.~~

~~The method of manufacturing the microchip for the nucleic acid amplification reaction further includes a second step of activating and adhering surfaces of substrate layers which the reagents are laminated and anchored to, in which the first step includes processes of dropping and drying an enzyme~~

~~solution in each of the wells, and then dropping and drying an oligonucleotide primer solution.~~

According to another aspect, there is provided a method of manufacturing a microchip for a nucleic acid amplification reaction, comprising:

a first step of laminating and anchoring a plurality of reagents needed for the reaction in a prescribed order in each of a plurality of wells configured to function as reaction sites of the nucleic acid amplification reaction, formed on a substrate layer, wherein the first step includes processes of dropping and drying an enzyme solution In each of the wells, and then dropping and drying an oligonucleotide primer solution,

further comprising:

a second step of activating and adhering surfaces of the substrate layers to which the reagents are laminated and anchored.

In the method of manufacturing the microchip for the nucleic acid amplification reaction, the first step favorably includes processes of dropping and drying a reaction buffer solute solution after the dropping and drying of the enzyme solution in each of the wells, and before the dropping of the oligonucleotide primer solution.

In addition, in the method of manufacturing the microchip for the nucleic acid amplification reaction, the second step may include process of activating the surfaces of the substrate layers with an oxygen plasma treatment or a vacuum ultraviolet light treatment.

[0014] In the present invention, "a nucleic acid amplification reaction" includes a conventional PCR (polymerase chain reaction) method performing a temperature cycle, and a variety of isothermal amplification methods involving no temperature cycle. Examples of the isothermal amplification methods include an LAMP (Loop-Mediated Isothermal Amplification) method, an SMAP (Smart Amplification Process) method, an NASBA (Nucleic Acid Sequence-Based Amplification) method, an ICAN (Isothermal and Chimeric primer-initiated Amplification of Nucleic acids) method (trademark), a TRC (transcription-reverse transcription

concerted) method, an SDA (strand displacement amplification) method, a TMA (transcription-mediated amplification) method, a RCA (rolling circle amplification) method and the like. Besides, "the nucleic acid amplification reaction" widely includes nucleic acid amplification reactions for amplifying nucleic acids at an alternating temperature or at a constant temperature. Also, "the nucleic acid amplification reaction" includes the reactions involving quantification of amplified nucleic acid strands such as a real time PCR (RT-PCR) method and an RT-RAMP method.

[0015] In addition, "reagents" include the reagents needed for obtaining the amplified nucleic acid strands in the above-mentioned nucleic acid amplification reaction, and specifically include an oligonucleotide primer having a complementary base sequence to target nucleic acid strands, a nucleic acid monomer (dNTP), an enzyme, a reaction buffer solution (buffer) solute and the like.

Effect of the Invention

[0016] ~~The~~ Embodiments of the present invention ~~provides~~ provide a microchip for a nucleic acid amplification reaction which allows high-precision analysis by a simple method.

Brief Description of Drawings

[Fig. 1] A schematic top view of a microchip for a nucleic acid amplification reaction according to the present invention.

[Fig. 2] A schematic cross-sectional view of the microchip (Fig. 1, p-p cross-section).

[Figs. 3] Schematic views illustrating anchored layers of reagents laminated in each of the wells of the microchip.

[Fig. 4] A schematic view illustrating an alternative example of anchored layers of reagents laminated in a well of a microchip.

[Fig. 5] A flowchart illustrating a method of manufacturing a microchip for a nucleic acid amplification reaction according to the present