

stance for trapping the analyte is immobilized to allow the analyte to be trapped by the trapping substance on the working electrode;

[0207] (2) forming a complex containing the analyte trapped on the working electrode in the process (1) and a label binding substance in which a labeling substance is retained via a modulator which generates an interaction with an electrolytic solution and a working electrode site except a site where the trapping substance are bound on a binding substance which binds to the analyte on the working electrode; and

[0208] (3) electrochemically detecting the labeling substance present on the working electrode obtained in the process (2).

[0209] A major characteristic of the method for electrochemically detecting an analyte according to the second embodiment of the present invention is that a complex containing an analyte and a label binding substance in which a labeling substance is attached to a binding substance via a modulator is formed on the working electrode.

[0210] In the method according to the second embodiment of the present invention, a photochemically or electrochemically active substance is used as the labeling substance. The photochemically active substance is detected using electrons released by excitation of the substance by light. On the other hand, the electrochemically active substance is detected using an oxidation reduction current and/or electrochemical luminescence based on the substance. Therefore, the method for the present invention can be divided broadly into the photoelectrochemical detection method (see FIGS. 23 and 25) and the oxidation reduction current/electrochemiluminescence detection method (see FIG. 26) depending on the type of detection technique of the labeling substance.

#### 1. Photoelectrochemical Detection Method

[0211] First, the photoelectrochemical detection method will be explained. In the photoelectrochemical detection method, the detector illustrated in FIG. 1 and the detection chip illustrated in FIG. 3 can be used, however, they are not limited thereto. Hereinafter, the method will be explained taking an example of the case of using the detector illustrated in FIG. 1 and the detection chip illustrated in FIG. 3.

[0212] Referring to FIG. 23, in the photoelectrochemical detection method, a user injects a sample containing the analyte S through the sample inlet 30b of the detection chip 20 [see the process of supplying a sample of FIG. 23A]. Thus, the analyte in the sample is trapped by the trapping substance 281 on the working electrode body 61 of the upper substrate 30 constituting the detection chip 20 [see the process of trapping an analyte of FIG. 6B]. In this case, substances (contaminants F) other than the analyte S in the sample are not trapped by the trapping substance 281.

[0213] As the trapping substance 281, the same substance as that of the trapping substance 81 described in the first embodiment can be used.

[0214] The process of trapping an analyte by the trapping substance 281 can be performed under the same conditions as those in the process of trapping an analyte by the trapping substance 81 described in the first embodiment.

[0215] Then, the user injects the label binding substance 290 into the detection chip 20 from the sample inlet 30b to allow the label binding substance 290 to be bound to the analyte S trapped by the trapping substance 281 on the working electrode body 61 [see the labeling process of FIG. 6C]. In

the labeling process, a complex containing the trapping substance 281, the analyte S, and the label binding substance 290 is formed on the working electrode body 61.

[0216] The label binding substance 290 is formed of a binding substance 291 to be bound to the analyte S, a modulator 292, and a labeling substance 293. In the label binding substance 290, the labeling substance 293 is fixed to the binding substance 291 via the modulator 292.

[0217] The binding substance 291 may be a substance which binds to a position or site in the analyte S, which is different from that of the trapping substance 281. The binding substance 291 is suitably selected depending on the type of the analyte S. For example, when the analyte S is a nucleic acid, a nucleic acid probe hybridizing to the nucleic acid, an antibody to the nucleic acid, a protein binding to the nucleic acid or the like can be used as the binding substance 291. When the analyte S is a protein or peptide, an antibody to the protein or peptide can be used as the binding substance 291.

[0218] The modulator 292 lies between the binding substance 291 and the labeling substance 293. The modulator 292 is a substance which interacts with the electrolytic solution and the working electrode body 61.

[0219] The modulator 292 is preferably selected from modulators having hydrophilicity and modulators having hydrophobicity depending on the electrolytic solution to be used in the detection process to be described later and the polarity of the surface of the working electrode.

[0220] In the method for electrochemically detecting an analyte according to the second embodiment of the present invention, it is preferable that the electrolytic solution contains an aprotic solvent and the surface of the working electrode and the modulator exhibit hydrophilicity, or the electrolytic solution contains a protic solvent and the surface of the working electrode and the modulator exhibit hydrophobicity.

[0221] Examples of the modulator having hydrophilicity include nucleic acids such as DNA and RNA; polyethylene glycol (a hydrophilic high molecular compound); and hydrophilic polypeptides mainly comprised of asparagine (a hydrophilic amino acid), serine, aspartic acid, glutamine, glutamic acid, threonine, arginine, histidine, ricin, tyrosine, and cysteine. Examples of the aprotic solvent include nitrils such as acetonitrile (CH<sub>3</sub>CN); carbonates such as propylene carbonate and ethylene carbonate; heterocyclic compounds such as 1,3-dimethylimidazolinone, 3-methylloxazolinone, and dialkyl imidazolium salt; dimethylformamide, dimethyl sulfoxide, and sulfolane. Among the aprotic solvents, acetonitrile is preferred. As an example of the working electrode having hydrophilicity, an electrode having a hydrophilic functional group on the surface is listed. Examples of the hydrophilic functional group include a functional group containing a hydroxyl group (for example, a silanol group), an amino group, and a thiol group. The working electrode having hydrophilicity can be obtained by, for example, treating the working electrode body with silane coupling agents such as aminopropyltriethoxysilane which provides an amino group and mercaptopropyltriethoxysilane which provides a thiol group. In the present invention, the electrode having a hydrophilic functional group on the surface may be subjected to hydrophilic treatment by binding polyethylene glycol, nucleic acid or the like to the electrode.

[0222] On the other hand, examples of the modulator having hydrophobicity include hydrophobic peptides mainly comprised of glycine (hydrophobic amino acid), tryptophan, methionine, proline, phenylalanine, alanine, valine, leucine,