

他誌発表論文 (国内)

Rapid and Accurate Diagnosis Based on Real-Time PCR Cycle Threshold Value for the Identification of *Campylobacter jejuni*, *astA* Gene-Positive *Escherichia coli*, and *eae* Gene-Positive *E. coli*

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We previously developed a multiplex real-time PCR assay (Rapid Foodborne Bacterial Screening 24 ver.5, [RFBS24 ver.5]) for simultaneous detection of 24 foodborne bacterial targets. Here, to overcome the discrepancy of the results from RFBS24 ver.5 and bacterial culture methods (BC), we analyzed 246 human clinical samples from 49 gastroenteritis outbreaks using RFBS24 ver.5 and evaluated the correlation between the cycle threshold (CT) value of RFBS24 ver.5 and the BC results. The results showed that the RFBS24 ver.5 was more sensitive than BC for *Campylobacter jejuni* and *Escherichia coli* harboring *astA* or *eae*, with positive predictive values (PPV) of 45.5-87.0% and a kappa coefficient (KC) of 0.60-0.92, respectively. The CTs were significantly different between BC-positive and -negative samples ($p < 0.01$). All RFBS24 ver.5-positive samples were BC-positive under the lower confidence interval (CI) limit of 95% or 99% for the CT of the BC-negative samples. We set the 95% or 99% CI lower limit to the determination CT (d-CT) to discriminate for assured BC-positive results (d-CTs: 27.42?30.86), and subsequently the PPVs (94.7%-100.0%) and KCs (0.89-0.95) of the 3 targets were increased. Together, we concluded that the implication of a d-CT-based approach would be a valuable tool for rapid and accurate diagnoses using the RFBS24 ver.5 system.

Determination of Trace Hydrazine in Environmental Water Samples by in situ Solid Phase Extraction

Toshikuni KATO, Hiroshi KAMIYA

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A simple and rapid in situ method for the determination of hydrazine based on the concentration of aldazine compound formed by the reaction of hydrazine with *p*-dimethylaminobenzaldehyde was developed. This method was based on solid-phase extraction using a Sep-Pak C18 cartridge, followed by the quantification of hydrazine using a spectrophotometric method. To a sample solution of environmental water, *p*-dimethylaminobenzaldehyde solution was added to form aldazine by the reaction with hydrazine. The solution was passed through a Sep-Pak C18 cartridge for the adsorption of aldazine. In the laboratory, the aldazine adsorbed on the Sep-Pak C18 cartridge was eluted by passing a hydrochloric acid–ethanol (1:10) solution through the cartridge, and the color intensity of the solution was measured at 457 nm. The limit of detection for the new method was 0.2 mgN L⁻¹ of hydrazine. The determination of hydrazine in solution was not influenced even by hydrogen sulfide and organic matter. This method was then applied to the brackish water of Lake Nakaumi in the eastern area of Shimane Prefecture, Japan. This method was used to determine hydrazine in freshwater, sea water and waste water.

Sensitive Method for the Oxidation-determination of Trace Hydroxylamine in Environmental Water Using Hypochlorite Followed by Gas Chromatography

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We developed a method for quantifying trace NH₂OH in brackish- and sea-water samples. Previously reported methods applicable to fresh water cannot be applied to such samples. We determined that interference in seawater owing to the bromide ion can be removed by the addition of phenol. In our procedure, phenol and hypochlorite solutions were added to a sample solution to oxidize NH₂OH to N₂O. N₂O in the sample was then quantified by headspace analysis. The method is not affected by the salt content or ammonia, nitrate, or nitrite at concentrations of 300 µgN L⁻¹ or less. It has a limit of detection of 0.2 µgN L⁻¹, and can quantify NH₂OH in natural water samples with a wide range of salinity. It was applied to samples from Lake Nakaumi, a brackish lake located in the eastern part of Shimane Prefecture, Japan.