

# METHOD STATEMENT



## Determinand:

*Salmonella* Sp. (other than *Salmonella Typhi*)

## Matrix:

Raw and Potable waters, soils and sludges

## Principle of Method:

Isolation is based on concentration from water by membrane filtration or from sludge and soil samples by dilution, followed by pre-enrichment involving incubation in a non-selective medium (to recover environmentally-stressed organisms) and selective enrichment with subculture to a selective agar. Confirmation is achieved by MALDI TOF analysis, with identification given to genotype level, biochemical and serological techniques can be used in the absence of the MALDI TOF. Using MALDI TOF gives a confirmation within 4 days.

## Sampling and Sample Preparation:

Once taken, microbiological samples should be transferred immediately to dark storage conditions and kept at a temperature between 2 - 8°C for transport to the laboratory. If samples are not analysed immediately on receipt in the laboratory, they should be kept at a temperature between 2 - 8°C, in dark conditions until analysis commences.

Samples should be analysed as soon as practicable on the day of collection. In exceptional circumstances, if there is a delay, storage under the above conditions should not exceed 24 hours before the commencement of analysis.

## Interferences:

Chlorine and chloramines. Neutralise by adding sodium thiosulphate which at a concentration of 18mg l<sup>-1</sup> should counteract up to 5mg l<sup>-1</sup> of free and combined residual chlorine.

Plasticisers can interfere in the mass spectrometry analysis. Only certified, non-plasticising plastic materials should be used.

## Performance of Method:

Sample results are reported as ND (if *Salmonella* is not detected) and DET (if *Salmonella* is detected). For quantitative analysis, positive isolates are interpreted in accordance with the MPN (most probable number) tables.

## References:

The Microbiology of Drinking Water (2002) Part 9 - Methods for the isolation and enumeration of *Salmonella* and *Shigella* by selective enrichment, membrane filtration and multiple tube most probable number techniques. Methods for the Examination of Waters and associated Materials. Environment Agency.

The Microbiology of Recreational and Environmental Waters (2016) Part 8 - Methods for the isolation and enumeration of *Salmonella* and *Shigella*. Methods for the Examination of Waters and Associated Materials. Standing Committee of Analysts.

The Microbiology of Sewage Sludge (2004) Part 4 -Methods for the detection, isolation and enumeration of *Salmonellae*. Methods for the Examination of Waters and Associated Materials. Environment Agency.