METHOD STATEMENT

Determinand:
Male-specific (F+) Coliphage

Scope:
Water and sediments

Principle of Method:
The test sample is mixed with a small volume of semi-solid nutrient medium to which a culture of host strain is also added. This is mixed and poured on to a solid plating medium which when set, is incubated under specified conditions. After incubation, the F-specific bacteriophage will infect the host strain to produce visible plaques (zones of clearance) which can be counted.

Sampling and Sample Preparation
Samples should be taken in sterile containers and transported to the laboratory under cool, dark conditions as quickly as possible.
On receipt, samples may be stored at 5°C ± 3°C and analysed within 24 hours. Alternatively, samples may be frozen until the day of analysis when added to TYE broth. Add 1 volume of TYE broth to 4 volumes of well mixed sample and freeze at -70°C ± 10°C. Immediately prior to analysis, frozen samples must be defrosted at room temperature.

Interferences:
Once started, the speed of analysis is critical. If the sample and host bacteria remain in contact for too long prior to agar pouring, then the number of plaque forming units may be over-estimated. The entire plating process should be carried out in less than 20 minutes.

Reporting of Results:
F+ Coliphage are expressed as plaque forming units, per volume of sample (usually 1ml) analysed.

References:
British Standard 6068. Water Quality Part 4. Microbiological methods. Section 4.11
Detection and enumeration of bacteriophages – Enumeration of F–specific RNA bacteriophages.

Detection and quantitation of F-specific RNA bacteriophages by double agar layer assay using *Escherichia coli* Famp or *Salmonella typhimurium* WG49 as the host bacterium. ALS method COR-MM510. I. Bolch, 14/10/10.

US EPA Method 1602: Male-specific (F+) and somatic Coliphage in water by single agar layer (SAL) procedure. 2001.