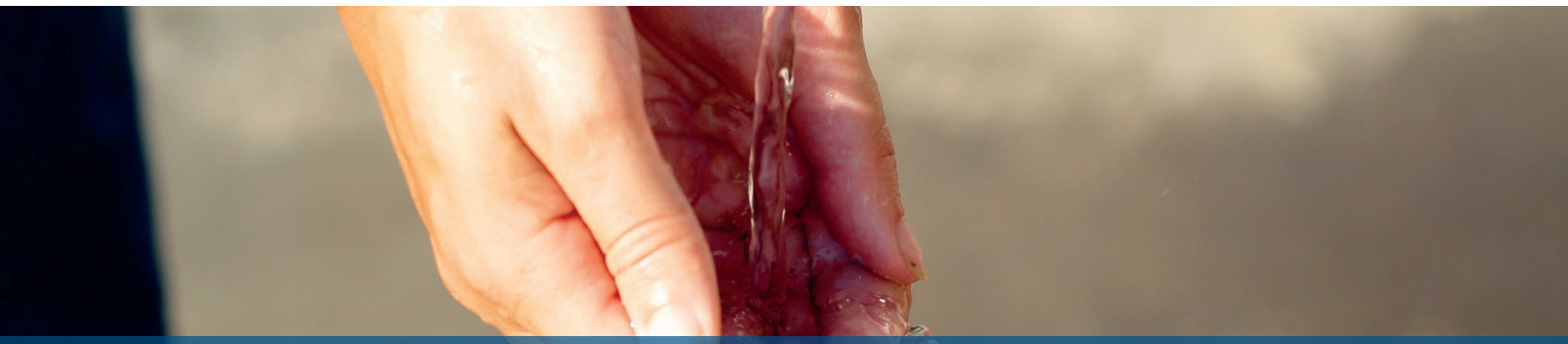

TECHNICAL DATASHEET

Cryptosporidium and Giardia Analysis in Water

Overview

Cryptosporidium is a waterborne, protozoan, parasitic human pathogen that infects a wide range of vertebrates. Infection is primarily caused by ingestion of water from a contaminated source, for example drinking or recreational water. Additional infection pathways have been described including person-to-person, zoonotic, foodborne and airborne transmission of Cryptosporidium oocysts.

In recent years there have been a number of significant Cryptosporidium outbreaks across the UK and Ireland. Notable cases as a result of contaminated Drinking Water were found in North Wales and Southern England and a case of infection from contaminated swimming pools in Galway, Ireland.

Infection from Cryptosporidium is termed Cryptosporidiosis and typical symptoms include diarrhoea, weight loss, abdominal pain, flu-like symptoms with headache, malaise, fever and muscle aches. The incubation period is typically one week but this may depend on the health of the patient and previous exposure. The symptoms may persist for up to 21 days and in severe cases in the immunocompromised, Cryptosporidiosis can be fatal. With an unknown infection dose there is a potential that one oocyst may be sufficient in causing Cryptosporidiosis.

Human Cryptosporidiosis constitutes one of the most common causes of protozoal diarrhoea worldwide. As a direct cause a significant morbidity and mortality rate in both the developing and developed world can be seen.

At least seven species (C. hominis, C. parvum, C. meleagridis, C. felis, C. canis, C. suis, and C. muris) and two genotypes (monkey and cervine) of Cryptosporidium are associated with human disease. Molecular approaches have enabled a greater understanding of the contributions of humans and livestock as reservoirs of infection. C. parvum and C. hominis are the most commonly occurring species related to infection by cryptosporidiosis.

Guidelines

The sampling and analysis for Cryptosporidium and Giardia is undertaken on a risk based approach and in the event of a suspected outbreak.

Table 1 provides some indication to the volume of water that may be sampled in order to acquire a representative sample from a range of sources. This is discussed in more detail in the Microbiology of Drinking Water series which is listed in the reference section of this leaflet. It states that

"Small volumes of samples may provide an indication of water quality at the time the sample is taken. However, due to the varying recovery efficiencies of the methods used, it may be very difficult to reliably determine low numbers of (oo)cysts in small volumes of samples. Large volumes of samples should provide an overall indication of water quality covering a longer period of time. When suspected waterborne outbreaks are investigated, the volume of sample taken may need to be a compromise between competing factors, for example the time taken for sample collection versus the urgency of requiring the results.

This may require strategic sampling of large volumes of water, together with more frequent sampling of smaller volumes of water (so that results may be obtained more rapidly in suspect areas)."

Table 1
Typical sample volumes for Cryptosporidium and Giardia monitoring

Sampling environment Sample range* (litres)
Distribution system waters 10 to 1000
Post-filter and final waters 10 to 1000
Backwash waters 1 to 10**
Surface waters of low turbidities 10 to 100**
Other surface waters 10 to 50**
Ground waters 10 to 1000**
Swimming pool waters 100 to 1000**
Treated wastewater effluents 10 to 20
Treated wastewater effluents 10 to 20
Solid samples (for example filter sand) 100 to 1000***
* Whilst it is desirable to use large volumes of sample, small volumes may need to be taken to facilitate management of outbreak situations where results are required quickly. ** Depending on the particulate loading of the sample. *** Amount in grams

TECHNICAL DATASHEET

Cryptosporidium and Giardia Analysis in Water

Sampling Equipment

ALS are able to provide sampling equipment which can be used in the field or appropriate sampling location, for example a swimming pool. Our equipment is capable of sampling large volumes of water. The larger the sample volume the more accurate the analytical result which in turn allows for a more accurate representation of the sampling location.

The sampling kit consists of a Water Meter to determine the sample volume analysed, the sampling device (housing within which is a sponge filter), interconnecting hoses that connect the housing to the meter and the sampling point and a complete set of instructions on the set up and use.

Analytical Method

Our method for Cryptosporidium analysis is both UKAS and DWTS accredited. Cryptosporidium oocysts appear as fluorescent apple green spherical objects when stained with a specific antibody stain under a microscope illuminated by a UV lamp at 465nm excitation. The oocysts have a typical morphology under a microscope as pictured in Figures 1-3 and summarised in table 2.

Figures 1-3

Figure 1 - FITC Stain

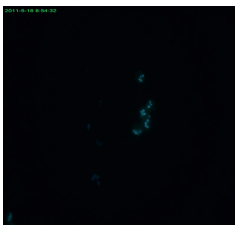

Figure 2 - DAPI Stain

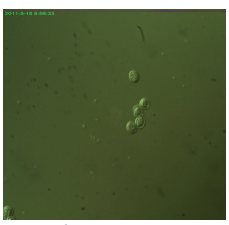

Figure 3 - DIC

Table 2

Morphology type	Typical Characteristics
Shape	Roughly Spherical
Size	4-6um
Reaction to Stain (FITC)	Bright Apple Green Flourescence
Oocyst wall	Entire
Internal Contents (DAPI)	Up to 4 DAPI stained nuclei, typically 1 um in diameter
DIC	Sporozoites can be observed within the oocyst or gape

The internal contents fluoresce under UV light whereby the nuclei may become visible as blue dots within the oocyst wall or in close proximity to the oocysts (see figure 1). Sporozoites (see figure 3) may be visible under Nomarski differential interference contrast (DIC) microscopy.

Reference

The Microbiology of Drinking Water (2010) - Part 14 - Methods for the isolation, identification and enumeration of Cryptosporidium oocysts and Giardia cysts

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