# Cryptosporidium in Water Supplies

Third Report of the Group of Experts to:
Department of the Environment, Transport and the Regions &
Department of Health

Third Report of the Group of Experts

Chairman – Professor Ian Bouchier

November 1998

Rt Hon John Prescott MP
Deputy Prime Minister and Secretary of State for the Environment, Transport and the Regions
Eland House
Bressenden Place
London SW1E 5DU

Rt Hon Frank Dobson MP
Secretary of State for Health
Richmond House
79 Whitehall
London SW1A 2NS

November 1998

Dear Secretaries of State

I have pleasure in submitting the Report of the Group of Experts on *Cryptosporidium* in water which was reconvened in response to an outbreak of cryptosporidiosis in north west London and Hertfordshire in March 1997.

We took as our starting base the information contained in the two Reports of the Group of Experts under the chairmanship of the late Sir John Badenoch. These Reports remain applicable to the problem of *Cryptosporidium* in water supplies. Indeed, as we evaluated waterborne outbreaks of cryptosporidiosis the relevance of many of the recommendations became highly apparent. Incidents do not just happen. There appears to be a strong correlation between outbreaks and situations where an inadequacy was identified in the treatment provided or in the operation of the treatment process, or where there was overloading of the treatment process. Furthermore the emphasis which these Reports placed on regular and close collaboration between the water utilities, the local authorities and the health authorities remains pertinent to supplying high quality water to the community and maintaining public confidence.

We paid particular attention to the issue of groundwater under the influence of surface water, a topic which had not received such prominence in the previous reports, because of its relevance to more recent outbreaks of cryptosporidiosis. The Group has made specific recommendations on risk assessment for groundwater sources.

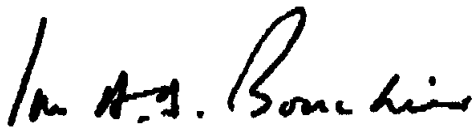
During our consultations and discussions it became apparent that there is some confusion amongst professionals involved in the water utilities and public health over the functions of Incident Management Teams and Outbreak Control Teams which form the basis for anticipating, identifying and managing incidents and outbreaks involving *Cryptosporidium* in water supplies. We hope that the functions of these Teams have been clarified and elaborated in this Report.

A number of other issues related to *Cryptosporidium* in water emerged and have been included in this Report, such as the need for high quality epidemiology when investigating outbreaks, water obtained from private water supplies, the risk posed to immunocompromised persons and *Cryptosporidium* in natural waters and beverages.

The Report has been written in a format which permits many of its sections to be issued as stand-alone documents or pamphlets, for example the Guidance on Epidemiological Investigations, Advice on the Management of Waterborne Outbreaks of Cryptosporidiosis and advice to Immunocompromised Persons.

I am most grateful to my colleagues who gave up so much time to the work of the Group; our meetings were always vigorous, stimulating and productive. Mr Michael Rouse of the Drinking Water Inspectorate and Dr Ailsa Wight of the Department of Health provided a great deal of helpful advice. I should also like to thank Dr Peter Gosling from the Department of Health and in particular Mr David Drury from the Drinking Water Inspectorate who worked so diligently and effectively on the production of the Report.

I am sending a copy of this letter to the Secretary of State for Scotland, the Secretary of State for Wales, the Secretary of State for Northern Ireland and to the Minister of Agriculture, Fisheries and Food.



Ian A D Bouchier

## MEMBERSHIP OF THE GROUP OF EXPERTS ON CRYPTOSPORIDIUM IN WATER SUPPLIES

### Chairman

**Professor Ian Bouchier CBE**

### Members

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| **Dr Catherine Benton**<br>Nominated by Scottish Water Authorities | *West of Scotland Water Authority* |
| **Dr David Casemore** | *Public Health Laboratory Service* |
| **Mr David Clapham** | *City of Bradford Metropolitan District Council* |
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1 Originally nominated by the Water Companies Association which became incorporated in Water UK on 1 April 1998.

2 Originally nominated by the Water Services Association which became incorporated in Water UK on 1 April 1998.

**Observers**

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| **Dr Judith Hilton**[3] | *Department of Health* |
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[5] Until July 1998

# Contents

# 1 Summary and recommendations

## 1.1 Summary

1.1.1 This is the Third Report of the Expert Group on *Cryptosporidium* in Water Supplies following those published in 1990 and 1995 under the chairmanship of the late Sir John Badenoch. Ministers decided to re-establish an Expert Group on *Cryptosporidium* in water supplies under the chairmanship of Professor Ian Bouchier following the outbreak of cryptosporidiosis in north west London and Hertfordshire in early spring 1997 which was associated with drinking water derived from underground strata.

1.1.2 The tasks of the Group as defined in its terms of reference were to:

- assess the lessons learned from suspected waterborne outbreaks of cryptosporidiosis;
- consider the results of research carried out since publication of the Second Report of the Group of Experts;
- consider whether there is a need for further advice on: protection of water resources, including surface and groundwaters; provision of additional water treatment; design of monitoring programmes and strategies; management of outbreaks of drinking water related illness;
- consider whether further research is appropriate; and
- report jointly to the Secretary of State for the Environment, Transport and the Regions and the Secretary of State for Health.

1.1.3 The Group addressed these requirements and came to the following important conclusions:

- Outbreaks of water related cryptosporidiosis do not just ‘happen’. There appears to be a strong correlation between outbreaks and situations where an inadequacy was identified in the treatment provided or in the operation of the treatment process, or where there was overloading of the treatment process.
- Turbidity monitoring through the water treatment process is a vital element in checking that treatment barriers are working properly. The unifying factor in all outbreak situations is the potential for peaks in turbidity to be present in the treated water leaving the works. The fact that turbidity events were not recognised in all cases could be a reflection of inadequacy in the continuity of turbidity monitoring, the interpretation of results or in the calibration and control of the equipment.

- Not all groundwater is consistently of high quality. Sources affected by the possibility of intermittent rapid transmission of water from the surface are potentially at high risk of contamination by *Cryptosporidium.*

- No reliance can be placed on disinfection at this time so physical barriers remain the form of treatment that will provide protection against *Cryptosporidium*. Conventional treatment is effective providing it is operated within design capacity and not bypassed.

- It is not possible to recommend a health-related standard for *Cryptosporidium* in drinking water.

- There is currently no cure for cryptosporidiosis. As a precautionary measure, to reduce the risk of waterborne cryptosporidiosis, immunocompromised persons should be advised to always bring to the boil all tap and bottled water used for drinking.

- There are lessons to be learnt from the experience gained from handling outbreaks, in particular, the need for local working partnerships and the importance of communications, planning and rehearsals.

- There is a need for more general recognition that water is not the only source of *Cryptosporidium* infection in humans. The organism can be acquired from food, milk, swimming pools, contact with farm and domestic animals and person to person transmission. Some epidemiological surveys following outbreaks have been deficient and there is a need for greater consistency in the quality of investigations.

- Making human cryptosporidiosis a laboratory reportable disease in England and Wales would enable earlier identification of outbreaks and greater consistency in their reporting and recording.

- There are a number of areas relating to *Cryptosporidium* that require further research including aspects of monitoring, analysis, water treatment and disinfection efficiency, transport and fate of oocysts in groundwater systems and therapy.

1.1.4 As a result of these findings the Expert Group has made a number of important recommendations with particular emphasis on advice to:

- water utilities;

- organisations and individuals concerned with the management of incidents and outbreaks; and

- those undertaking epidemiological surveys.

## 1.2 Recommendations

1.2.1 All of the Expert Group's recommendations are reproduced below with short explanations where necessary. The full background is given in the main body of this Report (numbers in brackets refer to the Report paragraph).

**Lessons learnt from analysis of outbreaks of waterborne crypotsporidiosis (Chapter 3)**

1.2.2 The Group concluded from its examination of these incidents that outbreaks of water related cryptosporidiosis do not just 'happen'. There appears to be a strong correlation between outbreaks and situations where an inadequacy was identified in the treatment provided or in the operation of the treatment process, or where there was overloading of the treatment process. A key factor in all situations was the potential for peaks in turbidity to have been present in the treated water leaving the works. The fact that turbidity events were not recognised in all cases could be a reflection of inadequacy in the continuity of turbidity monitoring or in the calibration and control of the equipment. Specific recommendations arising from these findings are included below under the heading Advice to Water Utilities but a general recommendation is given here.

*Recommendation*

***1.2.3 All water utilities should review their implementation of the Badenoch recommendations (3.2.1).***

1.2.4 The Group considers that there are four broad ways that a water utility could become aware of oocysts in the water supply:

- water utility detects oocysts in a sample of water;
- water utility detects a change of operational circumstance which potentially risks oocysts contaminating drinking water supplies;
- local health authority gets notification of an increase in stools containing *Cryptosporidium* oocysts; or
- the national disease surveillance centre is the first to detect a cluster of cases.

Arising from its consideration of these points, the Group has identified a number of recommendations to improve early recognition of potential problems, communications and reporting.

*Recommendations*

***1.2.5 Water utilities should investigate immediately when oocysts are detected in raw water to establish if any circumstances exist to allow Cryptosporidium to enter water supplies. Investigations should include review of recent treatment plant operational data (3.3.3).***

***1.2.6 Water utilities should develop local liaison arrangements with the local authority and health authority for rapid appraisal of the potential health risk, particularly when oocysts are detected in final water or in distribution (3.3.4).***

***1.2.7 Water utilities should ensure that employees operating assets producing drinking water are aware of the types of circumstance which can potentially put water supplies at risk of Cryptosporidium contamination. Procedures should be in place to ensure rapid recognition and appraisal of risks associated with any relevant change in operational circumstance (3.3.6).***

***1.2.8 Water utilities should provide copies of water supply zone maps to Consultants in Disease Control or Consultants in Public Health Medicine and health authorities should make early contact with the local water utility if an outbreak of cryptosporidiosis is suspected (3.3.9).***

***1.2.9 Human cryptosporidiosis should be made a laboratory reportable disease in England and Wales and consideration should be given to***

***making the disease notifiable. This should provide earlier identification of increased incidence of cryptosporidiosis and greater consistency in case reporting and recording (3.3.11).***

***1.2.10 Wherever possible, health authorities should make postcodes of cases of human cryptosporidiosis available to water utilities to help both organisations identify as early as possible if particular water sources are involved and to allow regional trends to be assessed (3.3.12).***

**Groundwater as a drinking water resource and its vulnerability to contamination by *Cryptosporidium* (Chapter 4)**

12.11 Groundwater provides over 30% of all water abstracted for public water supplies in England and Wales, 8% in Northern Ireland and 5% in Scotland. The possibility that an outbreak of cryptosporidiosis in March 1997 in north London was associated with a groundwater supply raised questions about *Cryptosporidium* as a possible groundwater contaminant. The Group concluded that not all groundwater is of consistently high quality. Water utilities should be especially vigilant for the possibility of intermittent rapid transmission of water from the surface into boreholes, wells and springs.

1.2.12 The isolation of oocysts in groundwater soon after rainfall recharge is a high risk circumstance which warrants immediate investigation. After making a risk assessment, water utilities should assess the possibility of minimising risk of contamination by reviewing catchment control options or by operational improvements to the security or integrity of the groundwater source. In rare cases the risk may be so unacceptably high that treatment installation is required. In the majority of cases, however, it will be necessary to carry out surveys and further investigations to confirm an unacceptable risk of groundwater contamination with *Cryptosporidium* before adopting a treatment solution. All risk assessments should be regularly reviewed, especially following any significant change in the catchment, the condition of the water supply source, or the demand on the source. The Group identified the following recommendations.

*Recommendations*

***1.2.13 Water utilities should systematically assess and rank the potential risk of groundwater contamination by Cryptosporidium by application of a tripartite approach which assesses source, catchment and hydrogeological factors (4.8.1).***

***1.2.14 Continued use should be made of existing national groundwater vulnerability maps and source protection zoning schemes to assess risk of contamination with Cryptosporidium (4.8.2).***

***1.2.15 For Cryptosporidium risk assessment, a fourth classification 'extreme vulnerability' is recommended for use with vulnerability maps and zoning schemes (4.8.3).***

***1.2.16 In order to ensure that groundwaters continue to be protected from agricultural activity, the Ministry of Agriculture, Fisheries and Food should promote further the application of the Code of Good Agricultural Practice – Water within the farming industry (4.8.4).***

***1.2.17 Careful attention should be given to the operational aspects of groundwater abstraction (4.8.5).***

**Advice to water utilities (Chapter 5)**

1.2.18 The Group concluded that 'incidents do not just happen'. Worldwide there is an increasingly strong correlation between outbreaks and inadequacies in drinking water supply. A key element in providing appropriate treatment is the assessment of risk from *Cryptosporidium*.

***Recommendation***

***1.2.19 The Group recommends that water utilities carry out an assessment of risk from Cryptosporidium for each source and put in place a procedure for updating periodically the review of the risk assessment. Water treatment requirements and monitoring systems should be reviewed against the level of risk (5.2.2).***

1.2.20 Most waterborne outbreaks occurred due to deficiencies in water supply including those in which the treatment was inadequate, or the works were operated above design capacity or some part of the treatment was bypassed. As recognised in the earlier Expert Group Reports a conventional treatment works (that is coagulation aided filtration) operated in accordance with good practice, is normally an effective barrier against *Cryptosporidium*. The Group makes the following recommendations on water treatment.

***Recommendations***

***1.2.21 Water treatment works should be designed to handle the typical peak turbidity and colour loadings in the source water (5.3.5).***

***1.2.22 Water treatment works should be operated at all times in a manner that minimises turbidity in the final water; attention should also be given to other parameters which reflect the performance of chemical coagulation, that is, coagulant metal concentration and colour (5.3.6).***

***1.2.23 Water treatment works should normally be operated within the design capacity and without by-passing of the solids-liquid separation processes which are responsible for removal of turbidity and coagulant solids; coagulation itself should never be by-passed or compromised (5.3.7).***

***1.2.24 In the event of an emergency, if it is necessary to overload or by-pass solid-liquid separation processes, a stringent monitoring regimen should be initiated to ensure that turbidity targets indicated in 1.2.32 below are not exceeded; if there is an indication that these targets will not be achieved, an immediate advice to boil notice should be issued (5.3.8).***

***1.2.25 For high risk sites, if minimisation of the effects of filter start up on final water quality cannot be achieved through more easily implemented changes (for example improved backwash or delayed start after backwash), modifications to the works should be made to allow the first flush to be run to waste or recycled to the works inlet (5.3.9).***

***1.2.26 Coagulation/flocculation processes should be checked regularly to meet changing conditions of source water quality and other environmental factors (5.3.10).***

***1.2.27 Only dedicated washwater mains should be used to carry the returned washwater flow (5.3.11).***

***1.2.28 Filters should be operated and maintained under optimum conditions with attention to the quality and depth of media and to the operation of the backwashing/air scouring system (5.3.12).***

***1.2.29 Treatment works staff should be trained to be aware of the potential effect on the final water quality of even very small changes in the catchment or the treatment stream (5.3.13).***

1.2.30 Investigation of waterborne outbreaks has shown that often there was a significant increase in turbidity at the time that the contaminated water was estimated to have gone into supply. The Group has made the following recommendations on monitoring.

*Recommendations*

***1.2.31 Water utilities should check that process monitoring systems are appropriate to the risk at each source (5.4.2).***

***1.2.32 For all sites at which Cryptosporidium might be a high risk, as determined by the risk assessment, monitoring should include continuous turbidity measurement on the outlet of each filter and on the final water using instruments capable of detecting changes of less than 0.1 NTU (5.4.3).***

***1.2.33 Water utilities should define for each of their treatment works the value and duration that constitute a significant deviation in turbidity of the final treated water irrespective of its relationship to the regulatory standard; for example it may be that at large water treatment works alarms should be set to be triggered by any increase in turbidity in the final water of greater than 50% of the normal average or suitably representative level; for small works, the increase of concern would vary and consideration should be given to the impact of the backwashing to individual filters (5.4.4).***

***1.2.34 Appropriate action procedures to react immediately to turbidity alarms, based on the level of risk and the history of the source/works should be in place; actions might include immediate sampling for Cryptosporidium, isolation of the filter(s) or source or, if suggested by history, the issue of advice to boil (5.4.5).***

1.2.35 Good experience has been reported with the operational use of particle counters which, when used in conjunction with turbidity monitors, can provide a more sensitive indication of particle breakthrough.

*Recommendation*

***1.2.36 The Group encourages the use of particle count monitors to provide additional information to that provided by turbidity measurements (5.4.7).***

1.2.37 Waterborne outbreaks occur even though oocysts cannot be detected in the water. This supports the general view that the 'contamination' occurs for only a few hours during which time it would be complete chance that routine samples coincided with the event. Random spot sampling is, therefore, unlikely to be effective for operational monitoring.

*Recommendation*

***1.2.38 Water utilities at high risk sites give consideration to either:***

***(i) continuous sampling for Cryptosporidium with analysis times linked to turbidity monitoring results; or***

***(ii) sampling triggered by turbidity events (5.4.9).***

1.2.39 Generally, good local working relationships and practices involving water utilities, health authorities and local authorities are in place. However, the Group wishes to emphasise the importance of both Incident Management Teams and Outbreak Control Teams and of the role of the water utilities in ensuring that the required local advice and support is in place to respond quickly to changing operational circumstances.

*Recommendations*

***1.2.40 Water utilities should review their working relationships with local health authorities and environmental health officers in the form of Incident Management Teams. Criteria should be established for identifying outbreaks and procedures put in place for activating Outbreak Control Teams (5.5.2).***

***1.2.41 Water utilities, in liaison with health authorities, should set out criteria for decision-making on the issue and the withdrawal of notice on advice to boil water and review these with experience (5.5.3).***

***1.2.42 Should there be an outbreak of cryptosporidiosis, the water utility, as a member of the Outbreak Control Team, should encourage the use of good epidemiology recommended in this Report to establish the source of the outbreak, including whether illness is associated with the drinking water supply (5.5.4).***

***1.2.43 Water utilities should encourage Incident Management and Outbreak Control Teams to review and rehearse regularly the response procedures to incidents and outbreaks (5.5.5).***

**Advice on management of waterborne outbreaks of cryptosporidiosis (Chapter 6)**

1.2.44 Since the First Report of the Expert Group, an increased understanding of cryptosporidial infection has been gained, as too has experience in incident and outbreak investigations and management. Incident Management Teams (IMT) and Outbreak Control Teams (OCT) are seen as part of the overall arrangements by health authorities for the control of communicable disease and as such are very practical teams whose aim is the protection of public health by the prevention of infection. If an incident leads to an outbreak of illness in the community or it is expected that this will occur, the IMT will evolve into an OCT. In practice, the function and membership of both these teams is similar, to protect public health and return the situation to normal as soon as possible. The exact stage when an IMT will become an OCT will depend on each incident and local circumstances and could be early in the incident.

1.2.45 It is important to remember however that there may be conflicting interests both within and outside the teams. The detail of information necessary to reach a conclusion for medical and public health needs may fail badly as legal evidence in a criminal prosecution and the resource required to collect different levels of evidence may vary considerably. It would be expected that, as the OCT is set up at the request of a Director of Public Health, it is primarily a team with a health objective.

*Recommendation*

***1.2.46 It is essential that Outbreak Control Teams are aware at the outset of the scope and purpose of their brief and that there is a clear understanding of the roles, responsibilities and standing of each member (6.1.5).***

1.2.47 An incident involving the breakthrough of *Cryptosporidium* oocysts through water treatment and into distribution may not be as obvious to identify as complete malfunctions such as disinfection or plant

breakdown. This is covered in more detail in Chapter 5 on advice to water utilities. It is possible that some water treatment plants may contribute to the background level of cryptosporidiosis in a community but there are other sources such as contact with animals, swimming pools, food and milk. Usually the sources of background levels are not investigated or identified.

***Recommendation***

***1.2.48 To facilitate recognition of an incident involving Cryptosporidium, there is a need for local studies to identify background levels of cryptosporidiosis, and for local risk assessments to be conducted so that any increased incidence can be identified easily (6.2.5).***

1.2.49 The need for systematic recording of events, particularly in the fast-moving early stages of an incident, should not be neglected and log-books will have an important role in the epidemiological study and in any subsequent review of lessons to be learned.

***Recommendation***

***1.2.50 All those involved at any stage of an incident should start a log-book immediately. This should include dates, times, key facts, summaries of telephone calls, and the actions taken by named staff (6.2.8).***

1.2.51 The Group recognises that there is a need for uniformity in the wording of advice on boiling water to ensure that the water is microbiologically safe whilst avoiding confusion to consumers and potential dangers with overheating electric kettles. It is necessary only to bring the water to the boil to kill *Cryptosporidium* oocysts. Water should be allowed to cool before use.

***Recommendation***

***1.2.52 All notices of advice to boil water issued to consumers should make it clear that it is only necessary to bring the water to the boil and then to allow it to cool before use (6.2.21).***

1.2.53 The Group emphasises the importance of liaison and team work in managing an incident or outbreak involving *Cryptosporidium* in the drinking water supply. The organisations concerned should meet regularly to discuss procedures and these should be rehearsed regularly.

***Recommendations***

***1.2.54 All parties likely to be involved in an IMT or OCT should establish a working dialogue and trust, preferably prior to the emergency situation, so that when a major incident occurs it will be dealt with more effectively (6.3.6).***

***1.2.55 All parties should regularly simulate incident and outbreak events to rehearse emergency procedures (6.3.7).***

1.2.56 There is a strong argument for some members of the team being appointed on account of their experience in dealing with water borne outbreaks previously or their knowledge of water treatment and distribution.

***Recommendation***

***1.2.57 The Expert Group recommends that a list of national experts who can be contacted in the event of an outbreak, be compiled. Consideration should be given to how the list should be compiled but it could include epidemiologists, public health microbiologists and water engineers with experience in the investigation of waterborne outbreaks of infection. Such experts would supplement local knowledge but not replace it (6.3.11).***

1.2.58 The Group recognises that the statutory powers for the investigation and control of communicable disease rests within the health authorities and the local authorities. However, it considers that OCT reports should be formally received and recommendations commented upon by the Drinking Water Inspectorate or its regulatory equivalent to ensure consistency and that any lessons learnt are communicated widely.

*Recommendation*

***1.2.59 OCT reports on waterborne outbreaks should be formally received and recommendations commented upon by the Drinking Water Inspectorate or its regulatory equivalents (6.3.14).***

**Guidance on the epidemiological investigation of outbreaks of infection (Chapter 7)**

1.2.60 The Group recognises that high quality epidemiological information is vital to the investigation of possible outbreaks of waterborne infection associated with mains water consumption because microbiological evidence of water contamination by pathogenic organisms is often difficult to obtain and even when it is available, such evidence is rarely conclusive. Some previous epidemiological surveys have been deficient and there is a need for greater consistency in the quality of investigations. The Group commends the use of the Guidance on the Epidemiological Investigation of Outbreaks of Infection (Appendix A4 of this Report) to assist in the conduct of epidemiological studies for CCDCs/CPHMs and other members of OCTs.

*Recommendation*

***1.2.61 The Group recommends the Chairman and members of the Outbreak Control Team use the Guidance on the Epidemiological Investigation of Outbreaks of Infection (Appendix A4 in this Report) in all outbreaks where waterborne infection is suspected (7.3.2).***

**Advice to the immunocompromised (Chapter 8)**

1.2.62 Cryptosporidiosis in immunocompromised people often results in a chronic life-threatening gastroenteritis with a high mortality. Whilst the Group recognises that the occurrence of *Cryptosporidium* in treated water is very rare it considers that the following recommendation will minimise the risk to immunocompromised people from drinking water.

*Recommendation*

***1.2.63 The Group recommends that all water, from whatever source, that might be consumed by immunocompromised persons should be brought to the boil and allowed to cool before use (8.3.7).***

**Current therapeutic approaches to cryptosporidiosis (Chapter 9)**

1.2.64 No antimicrobial agent has yet proved curative for cryptosporidiosis. However, there have been a number of encouraging reports on the use of paromomycin, and albendazole and nitazoxanide which may have some clinical use in cryptosporidiosis. A number of other agents including azithromycin, have shown some limited therapeutic effect. No drug regimens are known to be effective in preventing the recurrence of cryptosporidiosis.

*Recommendation*

***1.2.65 The Department of Health should continue to keep work in progress under review and encourage further controlled trials of new agents as they become available (9.6.2).***

**Review of recommendations in the Second Report of the Group of Experts (Appendix A1)**

1.2.66 Appendix A1 sets out those recommendations from the Second Report of the Expert Group, many of which originated in the First Report, which the current Expert Group considers to be of continuing relevance and worth emphasising. In addition the Group has added and amplified some of the recommendations where necessary and these are set out below.

*Further recommendations of the Expert Group*

***1.2.67 A national database should be established to provide comprehensive information on the occurrence of oocysts in both source and treated water (A1.3.1).***

***1.2.68 Research work on oocysts in sewage effluents should be directed at that work associated with typing and host specificity (A1.3.2).***

***1.2.69 The advice on storage and disposal of animal waste should be reaffirmed and efforts increased to encourage farmers to follow Codes of good practice (A1.3.3).***

***1.2.70 The inactivation of Cryptosporidium oocysts should be made one specific consideration in policy and practice in the disposal of sludges to land (A1.3.4).***

***1.2.71 Advice on personal hygiene in handling food, in preparation of ice and bottled waters should be reviewed and promoted by the new Food Standards Agency (A1.4.2).***

***1.2.72 It is considered that although disinfection has some effect, its contribution at time of most need (ie barrier breakthrough) has not been proven so in public health protection terms it cannot be relied upon (A1.5.2).***

***1.2.73 In light of some mistaken laboratory identifications of Cryptosporidium, consideration should be given to further training of laboratory staff and electronic links with expert laboratories (A1.5.4).***

**Future of the Expert Group**

1.2.74 The re-establishment of the Expert Group acknowledges the need to re-consider *Cryptosporidium* as a water supply issue in the light of experience gained in water treatment and outbreak management.

*Recommendation*

***1.2.75 The Expert Group should reconvene at two yearly intervals to consider, in the light of experience, whether additional advice should be issued and identify topics where further research is needed (2.9.2).***

**Recommendations for research (Chapter 11)**

1.2.76 The Group has identified the following as areas requiring further research:

***For groundwater:***

***(i) development of operational monitoring tools to improve the detection of rapid influence of surface water sources on the quality of groundwater;***

***(ii) transport and fate of Cryptosporidium and other pathogens in groundwater systems;***

***(iii) application of chemical and particulate tracers to investigate the transport and attenuation of pathogens in groundwater;***

***(iv) mechanisms causing, and the significance of, turbidity in groundwater to establish the role of rapid influence by surface water and assessing the use of turbidity as a monitoring tool; and***

***(v) attenuation rates for Cryptosporidium in soils and unsaturated zones following application of farm wastes and sewage sludge to land (4.7.2).***

***For treatment and monitoring:***

***(vi) application of continuous monitoring for Cryptosporidium in treated waters and investigation of correlation between Cryptosporidium and operating conditions that might lead to breakthrough of the organism;***

***(vii) investigations into the ways laboratory analytical procedures might affect the biological properties of oocysts;***

***(viii) development of a standardised approach to conducting disinfection trials;***

***(ix) development of reliable, routine tests for oocyst viability;***

***(x) further studies of the application of seroprevalence studies in assessing the impact of water treatment in reducing community exposure to Cryptosporidium;***

***(xi) investigation of the impact of operating filters under declining rate on the removal of Cryptosporidium;***

***(xii) evaluation of quality changes in treated waters and development of procedures to allow operators to identify Cryptosporidium risk associated with these changes for specific treatment works;***

***(xiii) development of techniques to specify and assess the performance of filtration systems for oocyst removal from groundwaters; and***

***(xiv) further evaluation and development of the use of bacterial spores to assess treatment performance (11.7.1).***

# 2 Introduction

## 2.1 Re-establishment of the Expert Group

2.1.1 In response to the outbreaks of cryptosporidiosis in Swindon and Oxfordshire in 1989 the Secretary of State for the Environment, jointly with the Secretary of State for Health, established an Expert Group to advise upon the significance of *Cryptosporidium* in water supplies. The Expert Group was chaired by the late Sir John Badenoch and reported in 1990 (Badenoch 1990). The Expert Group's recommendations were accepted by the Government and resulted in the establishment of a national research programme, which was steered by the Drinking Water Inspectorate (DWI) and has been reviewed in two reports (DOE 1992; DOE 1994). The Expert Group also made recommendations on reducing the risk of infection, ways to prevent *Cryptosporidium* getting into water supplies, monitoring strategies and investigation and management of outbreaks. In England and Wales DWI has monitored water utility implementation of these recommendations through its inspection process and similar checks have been carried out in Scotland by the Scottish Office.

2.1.2 The Expert Group was reconvened in 1994 to evaluate research findings and to assess the experiences of implementing its recommendations. The Expert Group's second report, which was published in 1995 (Badenoch 1995), included an authoritative and independent assessment of treatment requirements for *Cryptosporidium*. The report also made a number of recommendations on good practice, particularly on monitoring and operation of water treatment. The Expert Group concluded that properly operated conventional water treatment processes designed for removal of particulate material are usually very effective in removing *Cryptosporidium* oocysts from water.

2.1.3 The outbreak of cryptosporidiosis in north west London and Hertfordshire in early spring 1997 was associated with drinking water derived from underground strata. Such sources had generally been considered to be at very low risk of contamination by *Cryptosporidium*. It was important also to review the experience gained in water treatment and in outbreak management since the publication of the Second Report. Ministers therefore decided to re-establish an Expert Group on *Cryptosporidium* in water supplies under the chairmanship of Professor Ian Bouchier. Membership of the Expert Group is shown at the beginning of this Report.

## 2.2 Terms of reference

2.2.1 The terms of reference for the Expert Group are:

- to assess the lessons learned from suspected waterborne outbreaks of cryptosporidiosis;

- to consider the results of research carried out since publication of the Second Report of the Group of Experts;

- to consider whether there is a need for further advice on: protection of water resources, including surface and groundwaters; provision of additional water treatment; design of monitoring programmes and strategies; management of outbreaks of drinking water related illness;

- to consider whether further research is appropriate; and

- to report jointly to the Secretary of State for the Environment, Transport and the Regions and the Secretary of State for Health.

## 2.3 Format of the Report

2.3.1 Much of the advice given in the 1990 and 1995 Expert Group reports (Badenoch 1990; 1995) remains sound and is supported by the results of subsequent research. No attempt has been made in this Report to repeat the considerable body of information in the previous reports but rather to add to it where further data or information are available. The main recommendations from the two previous reports are listed in Appendix A1 and where necessary the Group has added comments.

2.3.2 The Report is written in the context of the administrative arrangements for water supply, health care and local government in England and Wales and in Scotland. The Water Supply Regulations are administered in England and Wales by DWI and the Scottish Office Environment Department and the Northern Ireland Environment and Heritage Department DWI have broadly similar functions in Scotland and Northern Ireland respectively. Much of the advice will be applicable outside the UK even where different administrative arrangements apply.

2.3.3 The main report is quite short and should be of value to both the technical and non-technical reader. Much of the detailed information is included as Appendices rather than in the body of the report. During its deliberations the Group has collected information on some topics which do not fall strictly within its terms of reference, such as the use of water by the food industry. Where the Group considers such information could be of use to a wider audience, and was not covered by the two previous reports, it is included. A paper on bottled water is included in Appendix A5.

2.3.4 This Report reviews: knowledge gathered and research carried out since the 1995 report; the lessons learnt from the analysis of outbreaks of waterborne cryptosporidiosis; the functioning of Incident Management Teams (IMT) and Outbreak Control Teams (OCT); the limitations of epidemiology and analysis; and the potential risks to groundwater. Where appropriate, recommendations for improvement are made. Advice is given to water utilities, IMTs and OCTs, immunocompromised people, the food industry and users of private water supplies. Topics for future research are identified.

2.3.5 Subsequent paragraphs in this section highlight very briefly the organism, the disease and the problems associated with water treatment and sampling and analysis of water supplies.

## 2.4 *Cryptosporidium parvum*

2.4.1 *Cryptosporidium* is a protozoan parasite found in man, many other mammals and also in birds, fish and reptiles. The only species known to infect both man and livestock is *Cryptosporidium parvum.* In the infected

animal, the parasite multiplies in the gastrointestinal tract. The animal then excretes oocysts of the parasite in its faeces in very large numbers, for example it is known that infected calves excrete approximately $10^{10}$ oocysts daily for up to 14 days and it is likely that humans shed a similar number.

2.4.2 *Cryptosporidium* oocysts are tiny spore-like structures, four to six micrometres in diameter, which carry within them the infective form, the sporozoites. When ingested by another host they can transmit the disease and set up a new cycle of infection. The oocysts are very resistant to adverse conditions in the environment and can survive dormant but viable for months in clean water or moist cool soil.

## 2.5 Cryptosporidiosis

2.5.1 In normal healthy individuals, cryptosporidiosis is usually characterised by an acute self-limiting diarrhoeal illness, commonly of two to three weeks duration, from which the patient recovers fully. In patients who are immunocompromised the disease is likely to be much more serious. The infective dose for humans is not known with any confidence but is thought to be quite low. As yet there is no effective specific treatment.

2.5.2 The prevalence of *Cryptosporidium* in livestock makes it likely that most oocysts in the environment derive from agricultural sources and wastewater discharges. However, recent studies have shown that human strains not derived from livestock are the prevalent form in at least some human waterborne outbreaks (see Appendix A7). All types of environmental water can become contaminated and oocysts may be present in low numbers in most waters from time to time. Drinking water is recognised as a vehicle for transmission although usually only when treatment is inadequate or compromised. There is always a low level of cryptosporidiosis in the community and it is unlikely that drinking water is a major cause of this background level. Sources other than human or livestock derived oocysts in water include contact with farm and domestic animals, swimming pools, food and milk. Cryptosporidiosis can be spread from animals to man and by person to person contact.

2.5.3 The evidence is now overwhelming that there are identifiably distinct ‘strains’ or sub-types (genotypes or lineages) of *C. parvum* and that one such sub-type appears on current evidence to be restricted to man. The evidence of a high prevalence of a ‘human genotype’ in three waterborne outbreaks has considerable significance in relation to transmission by the water route, suggesting that sewage effluent may have been the major source of these outbreaks. See Appendix A7.

2.5.4 Studies in healthy volunteers have shown that administration of 30-40 *C. parvum* oocysts is sufficient to initiate human infection (DuPont *et al* 1995). In view of the variable viability of oocysts in drinking water and the difficulty in confirming viability of oocysts obtained by filtration during routine surveillance of drinking water, it is unlikely that it will ever be possible to establish confidently a ‘safe limit’ of oocyst concentration.

## 2.6 *Cryptosporidium* and water treatment

2.6.1 The presence of *Cryptosporidium* oocysts in source water presents a particular challenge to water utilities because oocysts are resistant to the

standard chlorine disinfection regimens used for drinking water treatment and can be present in treated water in the absence of conventional bacterial indicators used to assess the efficiency of disinfection.

2.6.2 Conventional physical/chemical water treatment processes such as coagulation, sedimentation, dissolved air flotation (DAF), rapid gravity filtration and slow sand filtration were not designed to deal specifically with the problem of *Cryptosporidium* oocyst removal. However, such treatment can provide an effective barrier provided that the appropriate level of treatment for the raw water source has been installed and it is operated properly. However, if treatment is inadequate or compromised, significant numbers of oocysts may pass into the treated water supply. The use of membrane technologies can further improve oocyst removal.

## 2.7 Sampling and analysis

2.7.1 The analysis for most micro-organisms of relevance to the water industry involves starting from very low numbers, often one organism is sufficient, and growing them in artificial media or tissue culture until they are present in sufficient numbers to give a reliable result. Current routine analytical methods for *Cryptosporidium parvum* do not utilise any form of growth stage, so initially viability is not known. Oocysts have to be concentrated from large volumes of water, separated from contaminating debris, detected and, if practicable, tested for viability. Each stage can introduce large errors and recovery efficiencies as low as 1% have been reported.

2.7.2 Sampling and analysis for *Cryptosporidium* is presently the subject of considerable research worldwide. Significant advances have been made both in filtration and separation technologies and in the procedures for concentration of the organism prior to identification. Routine demonstration of oocyst viability is not reliable.

## 2.8 Proposed Regulation for *Cryptosporidium*

2.8.1 At the beginning of May 1998, the Department of the Environment, Transport and the Regions issued a consultation document, Preventing *Cryptosporidium* getting into Public Drinking Water Supplies (DETR 1998). This stated that Ministers had concluded that there is a case for increased monitoring at water treatment plants that are most at risk of releasing *Cryptosporidium* into drinking water supplies and have proposed amendments to the Water Supply (Water Quality) Regulations applying to England and Wales (Anon 1989). These propose a treatment standard of less than one oocyst in 10 litres based on continuously sampling 1000 litres of treated water per day. The document includes a number of issues for consultation. Although the consultation paper was not referred to the Group for consideration, the issues of particular interest to the Group are:

a) the rationale behind a treatment standard;
b) whether the proposals cover those sources of greatest risk; and
c) the use of continuous sampling.

2.8.2. It is understood that the proposed treatment standard has been derived from experience of routine samples in which the concentrations found in water treated according to accepted good practice were at least an order of magnitude lower than 1 oocyst in 10 litres and there was no increase in cryptosporidiosis in the community. The Group further

understands that the information available on infectivity, although limited, indicates that an infective concentration is at least an order of magnitude greater than 1 oocyst in 10 litres. The Group does not have any additional information on which to offer a different number to that proposed.

2.8.3. The analysis of outbreaks given later in the Report confirms that the greatest concern is on river sources which would be covered by the proposals as presented in the consultation paper. However there are other high risk situations, such as some groundwaters influenced by surface water, which do not have treatment appropriate for surface waters, and which do constitute an equivalent risk. The Group would like to see such sources included on a risk assessment basis.

2.8.4. The Group confirms, as concluded in the earlier Expert Group reports, that random spot sampling for *Cryptosporidium* is ineffective, as the chance of a sample being taken just as a pulse of *Cryptosporidium* is passing through the treatment works is very low. One solution is to introduce continuous sampling on high risk sites. The Group has received a preliminary report on the trials held on continuous sampling methods which show that one system is capable of processing 1000 litres of sample in a day for a variety of different treated waters with satisfactory and consistent recovery during analysis. This has confirmed the feasibility of continuous sampling for routine monitoring. The Group, although not commenting on the proposed regulations, recommends continuous sampling (see paragraphs 5.3.9 and 5.3.10) as one method of improving the effectiveness of monitoring in protecting public health.

## 2.9 Continuation of the Expert Group

2.9.1 The re-establishment of the Expert Group acknowledges the need to re-consider *Cryptosporidium* as a water supply issue in the light of experience gained in water treatment and outbreak management. The Group is aware that research and development is continuing around the world and it identifies an on-going need to consider whether additional advice is necessary and identify topics requiring further research. It considers that this would be best resolved by re-convening the Expert Group every two years.

***Recommendation***

***2.9.2 The Expert Group should reconvene at two yearly intervals to consider, in the light of experience, whether additional advice should be issued and identify topics where further research is needed.***

## References


Anon. (1989) The Water Supply (Water Quality) Regulations 1989 (Statutory Instrument No 1147) (as amended by SI No 1384 (1989), SI No 1837 (1991) and SI No 2790 (1991)). London. HMSO.

Badenoch, J. (1990) *Cryptosporidium* in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Badenoch, J. (1995) *Cryptosporidium* in water supplies. Second Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 108pp.

DETR (1998) Preventing *Cryptosporidium* getting into Drinking Water Supplies Consultation Document. Department of the Environment, Transport and the Regions, Welsh Office. London.

DOE (1992) *Cryptosporidium in water supplies*. Progress with the National Research Programme. Department of the Environment, Welsh Office. London: DWI.

DOE (1994) Proceedings of the workshop on *Cryptosporidium* in water supplies. Department of the Environment, Welsh Office, Department of Health. London: HMSO.

DuPont, H.L., Chappell, C.L., Sterling, C.R., Okhuysen, P.C., Rose, J.B., Jakubowski, W. (1995) The infectivity of *Cryptosporidium parvum* in healthy volunteers. *N Eng J Med* **332** 855-859.

# 3 Lessons learnt from outbreaks of waterborne cryptosporidiosis

## 3.1 Review of incidents

3.1.1 This section reviews incidents during the last ten years in which *Cryptosporidium* oocysts were detected in public drinking water supplies or where there was a suspected public drinking water related outbreak of cryptosporidiosis. The objective of the review is to establish whether there was any correlation between outbreaks of cryptosporidiosis and events which might pose a challenge to the integrity of the treatment or distribution process.

3.1.2 The Group is aware of twenty five outbreaks of cryptosporidiosis in the United Kingdom since 1988 that have been associated with the consumption of public drinking water supplies. These are listed in Table 3.1. The association with mains water varies from possible to probable to strong. Fourteen incidents have involved increased reporting of cases of cryptosporidiosis but in the absence of any reported detection of oocysts in water supplies. These may illustrate the limitations of current sampling and analytical techniques or indicate that the source was not the water supply. In eleven incidents, an increase in reported cases of cryptosporidiosis was associated with detection of oocysts in water supplies. The Group is also aware of eighteen notifications in England and Wales since 1990 involving detection of oocysts in water supplies but without any detectable increase in the level of cryptosporidiosis in the community which supports the practice of not over-reacting to low levels of oocysts in water supplies.

3.1.3 Most outbreaks occurred in situations where oocysts were not detected in water supplies. The tabulated data indicate that the majority of outbreaks of waterborne cryptosporidiosis occurred in situations in which the integrity of treatment had been compromised or where the treatment provided may have been less than adequate. In only two incidents was the correlation less than convincing.

3.1.4 Research funded by the Department of the Environment, Transport and the Regions (DETR 1998) on modelling the risk of infection has indicated that pathogens are non-randomly distributed in drinking water and may be associated with particles arising from turbidity events. It is significant that the majority of outbreaks identified were associated with situations in which turbidity increased although the regulatory standard for turbidity was not necessarily contravened. The groundwater incidents confirm that contamination of groundwater can be a significant risk factor. Time of year and climatic conditions may also be a risk factor, given that most incidents were reported during the late autumn to early spring period.

3.1.5 The Group concluded from its examination of these incidents that outbreaks of water related cryptosporidiosis do not just ‘happen’. There appears to be a strong correlation between outbreaks and situations where an inadequacy was identified in the treatment provided or in the operation of the treatment process, or where there was overloading of the treatment process. The unifying factor in all situations was the potential for peaks in

turbidity to have been present in the treated water leaving the works. The fact that turbidity events were not recognised in all cases could be a reflection of inadequacy in the continuity of turbidity monitoring or in the calibration and control of the equipment.

*Table 3.1* UK outbreaks of cryptosporidiosis associated with public drinking water supplies April 1988 – April 1998

| Date | Source/treatment characteristics (all water received normal chlorine based disinfection) | Oocysts detected in treated water | Approximate number of cases of cryptosporidiosis | Association with water[1] | Conclusions/ comments[5] |
|---|---|---|---|---|---|
| Apr 1988 | Surface water with coagulation and rapid gravity filtration | Yes | 27 | Strong[2] | Agricultural slurry contamination of water in distribution |
| Mar 1989 | Impounded reservoir supply, coagulation, rapid gravity filtration | Yes | 500+ | Strong[2] | Contamination of source water with animal wastes, breakthrough of treatment |
| Mar 1989 | Surface water, coagulation, rapid gravity filtration | Yes | Number of cases included in 500+ reported above | Strong[2] | River flows abnormally low, severe diarrhoea in cattle upstream of intake |
| Apr 1989 | Surface water, no filtration | Yes | 244 | Probable[4] | Unfiltered water, potential point source discharge from sewage treatment works and farm drains and non-point discharge from grazing animals |
| Dec 1989 | Lowland river with bankside storage, roughing filters and slow sand filtration | No | 477 | Strong[4] | Outbreak followed by-passing of filters |
| Dec 1990 | Lowland tidally influenced river, direct abstraction, gravity filtration | No | 47 | Probable[2] | Rapid fluctuations in source water quality at the time of the outbreak |
| Apr 1991 | Spring, well and stream supply, crude filtration | No | 5 | Possible[4] | Possible agricultural contamination of well supply, inadequate treatment |
| Apr 1992 | Surface water, no filtration | Yes | 50 | Probable[4] | Unfiltered water, potential point source discharge from sewage treatment works and farm drains and non-point discharge from grazing animals |
| Jul 1992 | Lowland river with direct abstraction, separation process and bankside infiltration with no filtration | No | 108 | Probable[3] | Possible link with groundwater turbidity |
| Nov 1992 | Upland reservoir and surface water supplying aqueduct, slow sand filtration | Yes | 125 | Strong[2] | Heavy rainfall in the catchment, high raw water turbidity, increase in treated water turbidity |
| Nov 1992 | Groundwater, no filtration | No | 47 | Probable[3] | Outbreak probably caused by faecal contamination from cattle housed adjacent to the well head |
| Apr 1993 | Stream source, no filtration | Yes | 3 | Probable[4] | Inadequate treatment, source open to potential animal contamination |
| Apr 1993 | Groundwater from fissured strata, no filtration | No | 40 | Probable[3] | Outbreak possibly caused by rapid recharge of surface water contaminated with oocysts |

*Table 3.1* UK outbreaks of cryptosporidiosis associated with public drinking water supplies April 1988 – April 1998 *(continued)*

| Date | Source/treatment characteristics (all water received normal chlorine based disinfection) | Oocysts detected in treated water | Approximate number of cases of cryptosporidiosis | Association with water[1] | Conclusions/ comments[5] |
|---|---|---|---|---|---|
| Apr 1993 | Upland reservoir, no filtration | No | 48 | Probable[4] | Possible run-off from grazing, very heavy rainfall |
| Jun 1993 | Upland reservoir and surface water supplying aqueduct, slow sand filtration | No | 97 | Probable[3] | Outbreak caused by poor operating practices and excessive head on the filters |
| Jun 1994 | Spring fed natural impoundment, upflow filtration | No | 8 | Probable[4] | Possible animal waste contamination following heavy rain |
| Feb 1995 | Spring supply, no filtration | No | 40 | Strong[4] | Heavy rain washed in waste animal material |
| Aug 1995 | Lowland river with direct abstraction, separation process and bankside infiltration with no filtration | Yes | 575 | Strong[3] | Plant operating above design output, evidence of turbidity peaks in the bankside infiltration water |
| Jan 1996 | Lowland river with coagulation and filtration | Yes | 126 | Strong[4] | Outbreak occurred when works was under strain with excess flow causing solids breakthrough |
| Mar 1996 | Surface water with bankside storage, rapid gravity filtration, no coagulant | No | 20 | Probable[4] | Outbreak caused by breakthrough of solids as a result of inadequate treatment during an algal bloom |
| Apr 1996 | Lowland river with full treatment | No | 80 | Probable[4] | Probable association with water but no evidence of plant operation problems |
| Feb 1997 | Groundwater from fissured strata, no filtration | Yes | 345 | Probable[4] | Outbreak caused by infiltration of surface water containing oocysts |
| Feb 1997 | Lowland river, coagulation and filtration | No | 22 | Probable[4] | Outbreak possibly associated with turbidity peak in filtered water |
| May 1997 | Spring supply, partial filtration | No | 34 | Possible[4] | Possible run-off from spring grazing |
| Apr 1998 | Surface water, no filtration | Yes | 303 | Possible[4] | Unfiltered water, potential point source discharge from sewage treatment works and farm drains and non-point discharge from grazing animals |

1 The definitions of strong, probable and possible association with water are given in Tillet *et al* (1998).
2 Tillet *et al* (1998).
3 Furtado *et al* (1998)
4 Personal communication from Expert Group Secretariat.
5 Conclusions and comments are those of the Drinking Water Inspectorate, health authorities, SCIEH, DWI NI or water utilities

3.1.6 The Group considers that the circumstances of these incidents do not alter the conclusion from the previous Expert Group reports (Badenoch 1990; 1995) that well operated appropriate conventional water treatment plants using processes designed for removal of particulate material minimise the risk of *Cryptosporidium* contaminating drinking water supplies. However, in the light of its assessment of incidents the Group has a number of recommendations. These are listed in Chapter 5, Advice to Water Utilities.

3.1.7 The Drinking Water Inspectorate (DWI) has published reports on four incidents in England where public drinking water supplies were associated with outbreaks of cryptosporidiosis (DWI 1993; 1997a; 1997b; 1998). One of these incidents is considered in the next section.

## 3.2 Cryptosporidiosis in north west London and Hertfordshire, spring 1997

3.2.1 The outbreak of cryptosporidiosis in north west London and Hertfordshire, in early spring 1997, with 345 confirmed cases, which was associated with drinking water derived from underground strata was one of the main reasons for re-convening the Expert Group. The DWI assessment of the incident was published in June 1998 (DWI 1998). It made recommendations specifically to the water utility involved on matters concerned with water quality monitoring, emergency procedures, risk assessment of groundwater sources, and implementation of recommendations contained in the two Reports of the Expert Group on *Cryptosporidium* in Water Supplies. It also made recommendations to all water utilities on matters concerned with risk assessment of groundwater sources and water quality monitoring; and to the Incident Management Team on matters associated with communications. The Group was concerned to note that it was necessary for the DWI report to refer to implementation of the recommendations from the two previous Expert Group reports. As most of these recommendations are still sound and relevant ***the Group recommends that all water utilities review their implementation of the Badenoch recommendations.*** The main Badenoch recommendations are listed in Appendix A1.

3.2.2 The DWI assessment of the north west London and Hertfordshire incident concluded that there were lessons to be learnt for all water utilities. ***It recommended that all water utilities:***

***(i) carry out risk assessments of groundwater sources to identify vulnerable sources requiring more rigorous surveillance;***

***(ii) consider the routine use of tracer tests as a measure of groundwater source vulnerability to pollution, especially those sources identified by risk assessment as most susceptible;***

***(iii) increase surveillance of shaft and adit chalk water sources vulnerable to fast infiltration of recharge; and***

***(iv) review their operational sampling programmes and establish where none exists or where the programme is shown to be deficient, a regular sampling and analytical programme for all separate sources of raw water.***

***3.2.3 The DWI report also recommended that a national survey of shaft and adit systems be undertaken to provide a better understanding of such systems, especially in chalk aquifers.***

***3.2.4 The Expert Group endorses these recommendations to water utilities as part of their risk assessment programme.***

### 3.3 Identifying and reporting the potential presence of oocysts in drinking water

3.3.1 The Group considers that there are four broad ways that a water utility could become aware of oocysts in the water supply. It notes that there are differences in the way this information would be handled in England and Wales, Scotland and Northern Ireland.

3.3.2 **Water utility detects oocysts in a sample of treated water.** In England and Wales and Northern Ireland the laboratory would normally notify the local operations manager or scientist. Further investigations would be made and the local authority environmental health department and local health authority Consultant in Communicable Disease Control (CCDC) or equivalent should be informed in accordance with locally agreed arrangements for notification. In Scotland, notification within the utility is the same but the local authority, Consultant in Public Health Medicine (CPHM) and the Scottish Office are informed.

*Recommendations*

***3.3.3 The Group recommends that the water utility should investigate immediately when oocysts are detected in raw water to establish if any circumstances exist to allow Cryptosporidium to enter water supplies. Investigations should include review of recent treatment plant operational data.***

***3.3.4 The water utility should develop local liaison arrangements with the local authority and health authority for rapid appraisal of the potential health risk, particularly when oocysts are detected in final water or in distribution.***

3.3.5 **Water utility detects a change of operational circumstance which potentially risks oocysts contaminating drinking water supplies.** Water utility response is essentially the same in England and Wales, Scotland and Northern Ireland with investigation by a treatment specialist or local operations manager. The most crucial step however, is early recognition and interpretation of the change of operational circumstance by a plant operative or local manager.

*Recommendation*

***3.3.6 The Group recommends that water utilities should ensure that employees operating assets producing drinking water are aware of the types of circumstance which can potentially put water supplies at risk of Cryptosporidium contamination. Procedures should be in place to ensure rapid recognition and appraisal of risks associated with any relevant change in operational circumstance.***

3.3.7 **Local health authority gets notification of an increase in stools containing *Cryptosporidium* oocysts.** There are important differences in reporting practice between England and Wales and Northern Ireland and Scotland. In Scotland details on **every** positive stool are sent by most CPHMs to the water utility for it to provide information on water supply source and treatment plant. The CPHM then collates these reports and comes to a view if there is evidence of a water-related clustering of cases. In England and Wales and Northern Ireland, the CCDC receives stool notification but rarely contacts the water utility immediately unless there is evidence accumulating to indicate clustering or patterns above expected background rates. Only when the CCDC is concerned about clusters or higher frequencies of isolation will the water utility be routinely asked to provide details of the water supply to each case. Ready availability to the CCDC/CPHM of local water supply zone maps will aid cluster identification.

3.3.8 The Group is anxious to avoid over or under reaction by health authorities and to not recommend a burdensome administrative process.

*Recommendation*

***3.3.9 The Group recommends that water utilities should provide copies of water supply zone maps to CCDCs/CPHMs or their equivalent and health authorities should make early contact with the local water utility if an outbreak of cryptosporidiosis is suspected.***

3.3.10 **The national disease surveillance centre is the first to detect a cluster of cases.** It is possible for the Centre for Disease Surveillance and Control (CDSC) for England and Wales and the Scottish Centre for Infection and Environmental Health (SCIEH), to establish case clustering when epidemiological data are pooled from each health authority area. It is usual for both centres to report findings to the local medical officer rather than directly to a water utility. The Group is aware that case reporting rates to CDSC and to SCIEH differ because cryptosporidiosis is a laboratory reportable disease in Scotland, but these centres routinely monitor outbreaks. For that reason, these centres do have a role to play in making drinking water safer. The national centres are able to show, over time, areas where there are higher than average rates of cryptosporidiosis incidence. However, in England and Wales, any attempt to link with water supplies is presently constrained, as cases are not coded by postcode. This makes integration of case locations into utility Geographic Information Systems (GIS) time consuming and inaccurate.

*Recommendations*

***3.3.11 Human cryptosporidiosis should be made a laboratory reportable disease in England and Wales and consideration should be given to making the disease notifiable. This should provide earlier identification of increased incidence of cryptosporidiosis and greater consistency in case reporting and recording.***

***3.3.12 The Group recommends that, wherever possible, health authorities should make postcodes of cases of human cryptosporidiosis available to water utilities to help both organisations identify as early as possible if particular water sources are involved and to allow regional trends to be assessed.***

### 3.4 Risk assessment

3.4.1 Research has highlighted the ability of oocysts of *Cryptosporidium* to survive in the aquatic environment and their tolerance of the disinfection process used in conventional water treatment. A consequence of this resistance to disinfection is that oocysts of *Cryptosporidium* can still be present when coliform bacteria, the traditional indicators of drinking water quality, have been inactivated.

3.4.2 To try and ensure freedom from risk of *Cryptosporidium*, other control strategies have had to be considered. Waterborne outbreaks occur even though oocysts cannot be detected in the water. This supports the general view that the ‘contamination’ occurs for only a few hours during which time it would be complete chance that routine samples coincided with the event. Random spot sampling is, therefore, unlikely to be effective for operational monitoring and a recommendation on continuous sampling or sampling triggered by turbidity events is made at 5.4.9. However, risk assessment, as recommended in the previous Expert Group reports (Badenoch 1990; 1995) has become an accepted strategy for the water industry.

3.4.3 Water utilities are now expected to make an assessment of the risk of contamination by oocysts for all water abstractions, including springs and groundwaters. If they judge there is a significant or unacceptable risk of contamination, utilities are expected to make an assessment of the effectiveness of the installed treatment in removing oocysts and the risk of an oocyst being present in the water supply. Utilities should review the assessments from time to time, particularly when there is a change in circumstances potentially affecting the risk.

3.4.4 Such assessments are not intended to predict *Cryptosporidium* outbreaks but they can highlight where they are most likely to occur. Changes in catchments and at treatment works can have significant affects on risk and it is important that these are reported and incorporated into assessments at regular reviews. Treatment works staff should be trained to be aware of the potential effect on the final water quality of even very small changes in the catchment or the treatment stream.

3.4.5 Risk assessment for water catchments and treatment works should cover anything in the catchment area that has the potential to allow *Cryptosporidium* into raw water. At the treatment works it should cover any factors which do not present a barrier to *Cryptosporidium*, or could contribute to *Cryptosporidium* breaking through filters, or which do not alert staff to filter breakthrough. Areas for consideration include the following.

■ **The type of water source**

- groundwaters and the likelihood of surface water influence
- security of underground springs
- surface water with direct abstraction or short duration storage facilities
- reservoirs and storage time
- river gravels

■ **Agricultural activity in the catchment**

- slurry and dung spreading
- sludge to land spreading
- slurry and dung stores
- abattoirs or livestock markets with land drainage

■ **Animals in the catchment**

- waste from animal housing
- cattle
- sheep
- deer herds
- pig farms
- wild animals
- birds (high numbers, wild or farmed)*

(*Probably only a source of *Cryptosporidium baileyi)*

■ **Sewage contamination of the raw water**

- sewage works
- septic tanks and cess pits
- storm water outlets

■ **Water treatment factors**

- full physical-chemical treatment
- partial physical-chemical treatment
- disinfection only
- is treatment process not used fully on every occasion
- is process known to be problematic
- does filter flow change suddenly
- is there a significant increase in turbidity before or after filter wash
- are there significant blips in turbidity during treatment runs
- are there signs of significant media loss or severe cracks in filter surface
- is backwash and/or sludge supernatant water recycled
- are turbidity meters on individual filters
- is turbidity alarm based on individual works performance
- are turbidity meters connected to alarm systems.

3.4.6 Microbiological risk assessment (MRA) is an emerging methodology to predict the risks to drinking water consumers from small numbers of pathogens breaking through into drinking water supplies. MRA models work by assessing the daily pathogen exposures to drinking water consumers and then translating those exposures into a risk by way of a dose-response curve. Limited dose-response data from human volunteer studies are available for *C.parvum* (DuPont *et al* 1995).

3.4.7 Current MRA models assume that pathogens are randomly distributed within the drinking water supply and therefore exposures to pathogens through drinking water is modelled by the Poisson distribution. There is considerable evidence however that bacteria are heterogeneously distributed in the drinking water supply and that treatment will cause clustering to occur. Intensive sampling studies using aerobic bacterial spores have shown that drinking water treatment removed 94–98% of spores but increased the clustering of those remaining. It is possible that oocysts are similarly affected. Through clustering a small proportion of consumers could be exposed to high doses through drinking water and models which do not consider rare high count of oocysts in samples could cause risk to human health to be underestimated (DETR 1998).

3.4.8 For these reasons the Group concludes that risk assessment should be based on a combination of factors including the degree of exposure of the catchment to oocysts, the treatment processes currently in place and the history of cryptosporidiosis in the community. Monitoring systems and water treatment requirements should be reviewed against the level of risk.

## References


Badenoch, J. (1990) *Cryptosporidium* in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Badenoch, J. (1995) *Cryptosporidium* in water supplies. Second Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 108pp.

DETR (1998) *Cryptosporidium*: Development of a risk assessment methodology for drinking water. Report No: DWI 4519. WRc plc for Department of the Environment, Transport and the Regions, London, UK.

DuPont, H.L., Chappell, C.L., Sterling, C.R., Okhuysen, P.C., Rose, J.B., Jakubowski, W. (1995) The infectivity of *Cryptosporidium parvum* in healthy volunteers. *N Eng J Med* **332** 855-859.

DWI. (1993) An outbreak of cryptosporidiosis in north Humberside, December 1989/January 1990: lessons for water supply. Drinking Water Inspectorate. London, UK.

DWI. (1997a) Assessment of water supply and associated matters in relation to the incidence of cryptosporidiosis in Torbay in 1992. Drinking Water Inspectorate. London, UK.

DWI.(1997b) Assessment of water supply and associated matters in relation to the incidence of cryptosporidiosis in Torbay in August and September 1995. Drinking Water Inspectorate. London, UK.

DWI. (1998) Assessment of water supply and associated matters in relation to the incidence of cryptosporidiosis in west Herts and north London in February and March 1997. Drinking Water Inspectorate. London, UK.

Furtado, C., Adak, G.K., Stuart, J.M., Wall, P.G., Evans, H.S. and Casemore, D.P. (1998) Outbreaks of waterborne infectious intestinal disease in England and Wales, 1992-5. *Epidemiol Infect* **121**, 109-119

Tillet, H.E., de Louvois, J. and Wall, P.G. (1998) Surveillance of outbreaks of waterborne infectious disease: categorizing levels of evidence. *Epidemiol Infect* **120,** 37-42**.**

# 4 Assessment of contamination risk for groundwater resources[6]

## 4.1 Groundwater use and protection

**Introduction**

4.1.1 The potential for contamination of groundwater by *Cryptosporidium* was addressed in the first and second reports of the Group of Experts (DOE/DH 1990 and 1995). These reports, however, only considered direct contamination at the wellhead via faulty, or poorly maintained, well or borehole construction. The likelihood that the outbreak of cryptosporidiosis in March 1997 in North London was associated with a groundwater supply raised questions about *Cryptosporidium* as a possible groundwater contaminant. The terms of reference for the re-convened Expert Group therefore included consideration of the need for protection of groundwater resources and sources.

4.1.2 For the purposes of this report, groundwater includes water abstracted from aquifers via springs, wells, boreholes and adits. There is a glossary of terms at the end of the chapter.

**Use of groundwater**

4.1.3 Groundwater provides over 30% of all water abstracted for public water supplies in England and Wales (DETR 1997), 8% in Northern Ireland and 5% in Scotland (Bell *et al* 1997). The regional differences reflect the distribution of aquifers and the more favourable geological conditions for surface water resource development in Northern Ireland and Scotland. It should be noted that in upland Britain while the proportion of the total supply derived from groundwater is low, the number of individual sources involved is large.

4.1.4 Over 80% of the total public supply in south-east England is derived from groundwater, while in the Severn and Trent basins, eastern England, the Thames Valley and the Wessex region the figure is between 30 and 50%. Extensive blending of groundwater and surface water further increases the extent of groundwater use. Industry and the agricultural community rely on groundwater in many areas and it is the predominant source for private water supplies. The total abstraction of groundwater in the UK, including that used by industry and agriculture, is some 2400 million $m^3$/year. About 85% is pumped from the two major aquifers, the Chalk and the Permo-Triassic sandstones which provide 60% and 25% respectively.

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**Protection of groundwater**

4.1.5 Groundwater source catchments are more difficult to define than the surface water equivalents as they cannot be as easily delineated by topography. Information from the Environment Agency's (EA) groundwater protection zone database shows that 875 of the 2,200 sources so far defined have a total catchment area exceeding 11,000 km$^2$. It therefore seems likely that as much as 15% of the land area of England and Wales may comprise catchment of an important potable groundwater supply source. The majority of groundwater-derived supplies is drawn from six major aquifer systems:

- the Chalk of southern and eastern England;
- the Permo-Triassic sandstones of central and northwest England and southern Scotland;
- the Lower Greensand of southern England;
- the Magnesian Limestone of central and northern England;
- the Jurassic limestones of southern, central and northern England; and
- the Carboniferous Limestone of southwestern and central England and south Wales.

4.1.6 More than one hundred other formations, notably fluvio-glacial gravels, provide locally important public and private potable supplies. The majority of important public supply groundwater sources are deep wells or boreholes, but there are several hundred licensed public spring supplies. Over a hundred of the wells have adit systems, mostly located in the Chalk of southern and eastern England. Many of these are highly productive: the average yield per source for 72 of these adited systems is over 10 Ml/d.

4.1.7 A technical and policy framework for groundwater protection in England and Wales is provided by the EA Policy and Practice for the Protection of Groundwater (NRA 1992). A similar strategy for Scotland was published subsequently (ADRIS 1995). A draft policy and practice document is under review for Northern Ireland. Central to each of these protection strategies is the principle of land surface zoning to protect aquifers as a whole and to safeguard specific sources of water supply.

4.1.8 The methodologies employed comprise:

- **A classification of groundwater vulnerability and a national set of maps showing areas of high, intermediate and low vulnerability to contamination.** A programme of mapping at 1:100,000 scale has been under way in England and Wales since 1992. Complete national coverage on 53 maps has now been published. In Scotland a national synoptic-level map at 1:625,000 has been published. Selective coverage of about seven important areas at a larger scale (1:100,000) is envisaged, one of which has been published. In Northern Ireland a national vulnerability map at 1:250,000 has been published. The maps all classify groundwater on the basis of intrinsic aquifer vulnerability. This addresses characteristics that determine vulnerability, rather than behaviour of specific contaminants.

- **Source protection zones defined by the travel time of potential pollutants and the overall source catchment areas**. These extended zones are superimposed on those zones defined on groundwater vulnerability criteria. The programme of defining protection zones around

supply sources in England and Wales also commenced in 1992, and of the 2,500 or so licensed public groundwater supplies yielding 0.25 Ml/d or more, some 2,200 now have delineated zones. However, due to the inherent uncertainties in the data used in groundwater catchment delineation techniques, there is generally a margin of error, sometimes significant, in the resultant zones. Relatively few groundwater public supplies have been zoned in Scotland, other than those required under the EC Nitrate Directive.

■ **A matrix of policy statements, relating to activities at the land surface and to the control of groundwater abstraction.** These allow the EA to exercise consistently its statutory powers. At the same time users and other interested parties can assess what practices are likely to be unacceptable in a given zone.

### 4.2 Microbiological contamination risk

**Vulnerability of groundwater to microbiological contamination**

4.2.1 Most infectious agents in human and animal wastes are potentially waterborne. Faecal matter contains very large numbers of non-pathogenic bacteria but may also contain helminth eggs, protozoa, bacteria and viruses which are capable of causing infectious diseases. Most protozoa and helminths are likely to be removed by filtration during passage through soil, or by predation by other micro-organisms. The protozoan *Cryptosporidium* may be atypical in that the 4-6μm diameter of the environmentally-resistant oocyst stage is smaller than many other protozoa.

4.2.2 While most microbial contaminants die off in passage through the soil, they may enter the subsurface directly via structures which by-pass the soil zone such as septic tanks, latrines or soakaway pits. As these structures are designed to dispose of liquid wastes quickly, they will almost invariably impose a higher hydraulic loading than pollutants entering at the top of the soil zone. This implies less potential for pathogen removal during subsurface flow. In agriculture, new or substantially modified farm waste storage facilities must comply with the Control of Pollution (Silage, Slurry and Agricultural Fuel) Regulations 1991, as amended, which requires them to be impermeable. An analogous infiltration process can occur along losing reaches of watercourses, especially where the flow regime and underlying drift deposits result in a silt-free permeable stream-bed.

4.2.3 Where by-passing of the sub-soil is common, the role of the unsaturated zone is especially important. Thus in rural communities using on-site domestic or agricultural wastewater disposal, the unsaturated zone offers an effective barrier for pathogen removal. Like the soil profile, the unsaturated zone exhibits aerobic conditions. As aerobic degradation is more effective than anaerobic decomposition for many organic compounds, this will favour pollutant removal as part of a natural biological process. However, soil and unsaturated zones can be overloaded. Furthermore, microorganisms may percolate most soils and rock pores, except in fine-grained strata where pore diameters are small. The proximity of highly permeable fluvio-glacial gravels to rivers creates a particular contamination risk for sources in such groundwater settings.

4.2.4 *Cryptosporidium* oocysts are larger than the typical 1μm pore size of the Chalk aquifer but they are within the pore size range of arenaceous aquifers like the Permo-Triassic sandstones. Oocysts are almost certainly

smaller than the fissure and micro-fissure/bedding plane aperture systems which dominate groundwater transmission in most important UK aquifers. Of the most productive UK formations referred to above, only the Lower Greensand is now regarded as a predominantly intergranular-flow aquifer. Thus the capacity of many UK aquifers to physically detain oocysts is limited. Intergranular unsaturated zone vertical flow is also typically slower than its saturated zone equivalent. However it should be noted that the effects of fissures can be even more important in the unsaturated zone than below the water table, because significant by-pass flow can occur along vertical fractures activated after major recharge events. This by-pass flow can be very fast compared to typical intergranular flow rates.

4.2.5 The fate of micro-organisms in the subsurface is also determined by the interaction of flow travel time with survival rates. At 20˚C, a 90% reduction in bacterial counts may be expected within about 10 days, although a few may persist for 200 days or more (Lewis *et al* 1982). Groundwater temperatures in England are typically about 10˚C and at that temperature, survival of micro-organisms may be at least twice as long. Viability of *Cryptosporidium* in the deep subsurface has not been studied but oocysts have been reported to survive dormant for months in moist soil or up to a year in clean water (Badenoch 1990). Oocysts may be expected to survive in the subsurface for periods that are at least two orders of magnitude longer than that of most bacterial cells (including pathogens) of faecal origin.

4.2.6 The groundwater vulnerability mapping and protection zone programmes undertaken by the environmental regulators provide the sound basis for assessing which British public groundwater supply catchments are likely to be most susceptible to contamination but there are some limitations when assessing risk from *Cryptosporidium.*

■ Although the vulnerability maps use soil leaching potential as a criterion, they do not take account of the other most important attenuating property: depth to water table. For a contaminant like *Cryptosporidium* this factor, which extends recharge travel time, could be significant, given its potential for retention in the unsaturated zone.

■ By-pass features that may allow the rapid passage of water from the land surface to the water table, are not taken into account in the vulnerability classification. Many important British carbonate aquifers, eg the Jurassic limestones, the Magnesian Limestone, the Carboniferous Limestone and the Chalk, contain zones where bypass flow is likely. Similarly, no account is taken of surface water-aquifer interactions, which occur most frequently in two distinct settings: valley bottoms, where natural or induced recharge from mature rivers occurs; and in the upper catchment where local surface drainage enters solution features.

■ Source protection areas must evolve to reflect temporal changes such as annual rainfall patterns or interference effects from adjacent pumping wells. It is necessary also to take account of better understanding of the groundwater setting of the source. It is possible to use statistical modelling to define catchment areas but in some cases boundaries of entry of water into an aquifer may be uncertain.

4.2.7 These limitations could be overcome by incorporating a fourth class of ‘extreme vulnerability’ into the existing schemes operated by the EA and equivalent bodies in Scotland and Northern Ireland. This would

apply to suspected rapid-access points such as solution features, sinkholes, karst or pseudo-karst features, as well as known areas of ground disturbed by human activities, such as mining and aggregate extraction. This zone, together with the presence of influent (losing) surface watercourses and water bodies could be mapped and would, in effect, define those areas in which groundwater is, or may be, under the rapid influence of surface water.

**Reported groundwater-related *Cryptosporidium* incidents**

4.2.8 Evidence is accumulating worldwide for the potential for contamination of groundwater by *Cryptosporidium* (Harvey 1992; National Cryptosporidium Survey Group 1992; Hancock *et al* 1998). Table 4.1 lists events associated with water supplies in England since 1990. The range of aquifer types varies but chalk, which is the UK's most productive and widely used aquifer, predominates. This may be more a consequence of its extensive exploitation as a water resource. Fissure flow, dual porosity and intergranular flow are represented. However, intergranular flow is only affected in settings where the residence time in the aquifer is likely to be very short, eg in river gravels close to a surface watercourse. Rural and part-rural/part-urban catchments are involved, although occurrences in the former are more commonly reported. Wells with adits, springs with galleries or former mines with adits appear to be particularly vulnerable settings, accounting for seven of the known events. This feature appears to be more significant than mere proximity to a watercourse.

*Table 4.1* Suspected *Cryptosporidium* groundwater contamination events in England since 1990

| Year of occurrence | Aquifer lithology | Aquifer flow type | Supply source type | Comments |
|---|---|---|---|---|
| 1990-91 | River gravels over chalk | Intergranular/dual porosity | Well with adits | Adjacent to river |
| 1992, 1995 | River gravels | Intergranular | Collector well used conjunctively with surface water | Adjacent to river |
| 1992 | Sandstone | Dual porosity | Well with adits | Contaminated grazed field runoff to wellhead, possible septic tank leakage in wellhead area |
| 1995 | Chalk | Dual porosity | Well with adits | – |
| 1995, 1996 | Sandstone | Fissure | Adited spring | – |
| 1997 | Chalk | Dual porosity | Single borehole | Grazed catchment, losing stream with sewage effluent discharge close to borehole |
| 1997 | Sandstone, karstic limestone | Fissure | Adit of former mine | Possible slurry pit leakage |
| 1997 | Chalk | Dual porosity | Well with adits | Adjacent to river |
| 1997 | Gravels | Intergranular | Collector well | Very shallow well in thin gravel on flood plain, seasonal flooding and gravel pits adjacent |

4.2.9 Table 4.2 lists factors and prioritises risk factors that water utilities should consider for modelling or when assessing the risk of groundwater contamination by *Cryptosporidium*. A more detailed treatment of groundwater risk assessment models is included in Lawrence *et al* 1996 and in a water industry research report (UKWIR 1998).

*Table 4.2* Factors for consideration in the risk assessment of groundwater contamination

| PREDISPOSING GROUNDWATER TO *CRYPTOSPORIDIUM* RISK | POSSIBLE VERIFICATION TECHNIQUES |
|---|---|
| **Well/raw water source factors:** | |
| Supply source tapping shallow flow systems eg adits, springs, mine galleries | Check site plans, tracing |
| Adits with upbores or construction-stage ventilation shafts | Check site plans, site inspection |
| Poor casing integrity | CCTV, geophysical logging |
| Masonry linings above pumping water level without additional sanitary seal | CCTV, check site plans |
| Sewer/septic tank/slurry pit systems near wellhead or above adits | Site inspection |
| Inadequately fenced source especially around spring boxes, catchpits, galleries | Site inspection |
| Old, poorly documented well construction | Site plans/BGS National Well Record Archive |
| **Hydrogeological factors:** | |
| Known or suspected river aquifer connection nearby | Flow gauging, modelling, hydrochemistry |
| Unconfined conditions with shallow water table | Well water-level monitoring |
| Karst or known rapid macro-fissure flow conditions, especially in shallow groundwater | Field mapping, farm surveys |
| Patchy drift cover associated with highly contrasting aquifer intrinsic vulnerabilities | Field mapping, shallow drilling |
| Solution features observed or inferred in catchment | Field mapping |
| Shallow flow cycles to springs | Tracing, hydrochemistry, water temperature logging |
| Fissure-dominant flow (as suggested by high transmissivity or specific capacity) | Downhole fluid/flow logging, pumping test analysis |
| **Catchment factors:** | |
| High wastewater returns, including sewage effluent to losing river reaches, especially under baseflow conditions | Hydrochemistry, microbiology, hydrometry |
| Livestock rearing in inner catchment, especially if intensive | Farm survey |
| Likely *Cryptosporidium* – generating activities in catchment eg abattoirs | Economic activity survey |
| Urbanising catchment | Cadastral survey |
| Livestock grazed/housed near wellhead patio/courtyard | Site inspection |

## 4.3 Catchment control to minimise groundwater contamination

**Introduction**

4.3.1 Catchment control measures can be broadly based on:

(i) avoiding the development or establishment of a hazardous activity within the catchment or zone (**prevention**);

(ii) removing, shutting down or banning hazardous activities (**elimination**); and

(iii) adopting measures which reduce the risk associated with hazardous activities (**mitigation**).

4.3.2 Prevention measures are available through statutory mechanisms involving the Town and Country Planning legislation and pollution control legislation. The possibilities for elimination of hazardous activities may be limited if the owner or occupier of affected land or property can claim for compensation. Possible approaches to mitigating a hazardous activity

include: statutory controls; codes of practice; and raising of awareness through education and promotion of good practice.

**Regulatory controls**

4.3.3 The Town and Country Planning legislation allows water utilities and the EA to comment on planning applications involving change of land use. However, water utilities are not statutory consultees and therefore rely on the goodwill of councils and the EA to bring relevant applications to their attention. While there is no obligation for the planning authorities to act on these comments, in practice they are taken seriously and can result in an application being refused, or approved subject to conditions. However, certain types of development, including many agricultural developments are exempt from planning requirements. Therefore, existing activities, many of which could be regarded as undesirable, fall outside the scope of planning controls, which are triggered mainly by applications to change land use.

4.3.4 The Water Resources Act 1991 requires the EA to consent all trade and sewage effluent discharges to controlled waters, which include inland surface and groundwater. The position for groundwater is complex as indirect discharges, ie those to the ground itself, may not need consent. The Control of Pollution (Silage, Slurry and Agricultural Fuel) Regulations 1991 require that new or substantially altered silage or slurry storage structures be built to a minimum standard, which must be notified to the EA before being brought into use. Legislation also provides powers for the EA to serve a notice requiring improvements where there is a risk of pollution. The European Community's Water Framework Directive may provide additional indirect controls. Ammonia is designated as a List II substance and as such its discharge into groundwater should be minimised. This offers the possibility of reducing the impact of organic effluents on groundwaters. The designation of Nitrate Vulnerable Zones may also provide some reduction in the risk of groundwater contamination from *Cryptosporidium* by obliging farmers to comply with a Programme which is based on the Code of Good Agricultural Practice for the Protection of Water (COGAP-Water) (MAFF 1991; 1998).

4.3.5 Groundwater contamination arising from waste disposal is controlled in a number of ways. Landfill sites require a waste management licence from the EA. For new sites, this will normally involve lining of the site and a comprehensive leachate management programme. The spreading of wastes 'for agricultural benefit' is permitted subject to the requirements of COGAP-Water. This practice is exempt from the licensing requirements but the EA must be notified. In practice this provides an opportunity for the EA to examine proposals and control those that do not meet the required criteria.

4.3.6 The conditions for application of sewage sludge to land are set out in the Sludge (Use in Agriculture) Regulations 1989. These set out sludge treatment requirements and application rates to protect public health and soil condition. The Regulations are supported by a code of practice. Sewage sludge spreading is expected to increase significantly when disposal at sea stops in 1998 in line with the European Community's Urban Wastewater Treatment Directive (EEC 91/271). Following recommendations by the Royal Commission on Environmental Pollution, the House of Commons Environment Committee and a report by independent consultants, the Government has agreed to amend the Regulations in order to phase out the use of untreated sludge on agricultural land by the end of 2001. Discussions between the British

Retail Consortium and the Water Industry are close to finalising an agreement, which will introduce additional controls on sludge use and cropping to further reduce perceived risks of pathogen transmission.

**Codes of Practice**

4.3.7 COGAP-Water, which was revised in 1998, includes guidance on avoiding water pollution from agricultural activity. The Code covers faecal contamination of water and *Cryptosporidium* in particular. The Code advises that special precautions, for example, on slurry and manure storage be taken on farms where cryptosporidiosis has been identified in livestock, in order to contain the spread of *Cryptosporidium* in the farm environment. The Code has a statutory designation code under section 97 of the Water Resources Act 1991. As such, any contravention can be taken into account if legal action follows a pollution incident. Raising awareness of this important Code among farmers remains vital for the continued protection of water resources (See Appendix A2).

**Awareness, education and partnerships**

4.3.8 Regional and national initiatives to promote good agricultural practice have achieved locally increased use of the Code by the farming industry. In respect of *Cryptosporidium*, a pilot initiative 'Wise Ways With Waste' (Water Services Association 1995) set out to promote safe slurry application to protect watercourses.

4.3.9 There is scope for innovative practice such as development of further incentive schemes within the framework of the EC Agri-Environment Regulation under the Common Agricultural Policy. This Regulation promotes schemes which aim to encourage farmers to undertake positive measures to conserve and enhance the rural environment. The latter scheme now covers 10% of the agricultural land in England and a reasoned case could be made to support inclusion of 'sustainability of water resources' as an additional threat to the environment which merits consideration within the ESA scheme.

**Communication**

4.3.10 A number of circumstances are now known to raise significantly the risk of groundwater contamination by *Cryptosporidium*. In any of the following situations, rapid communication is crucial if the water company and local public health professionals are to assess the impact on water supplies:

■ **Unusually large outbreaks of cryptosporidiosis among farm animals**. This is not a notifiable occurrence but there should be a line of communication between local vets, the MAFF veterinary service, the EA and the water company.

■ **Pollution incidents involving faecal contamination**. There is already communication between the water utilities and the EA over surface water pollution incidents. These arrangements should be broadened to ensure that incidents that might pose a risk to groundwater are included.

■ **Land spreading of exempt waste** (under the Waste Management Licensing Regulations 1994). These activities must be notified to the EA and, if spreading is taking place within a source protection zone, then the EA should also inform the water utility. When spreading is carried out on more than one occasion, arrangements should be agreed between the operator and the utility. Water utilities follow internal management procedures for their own sewage sludge spreading activities. Communication concerning the activities of water utility sub-contractors is important, particularly when spreading takes place in another utility's catchment area.

■ **Contamination from losing reaches of rivers adjacent to groundwater sources.** This represents a significant risk to public water supplies if oocysts are present. Water utilities need to be made aware of the possibility of such contamination and of the impact of seasonal changes in groundwater levels and river flows.

## 4.4 Operational aspects of groundwater abstraction

**Flow and level measurement**

4.4.1 Records of groundwater level, quantities abstracted and in the case of springs, total flow, provide useful information for assessing risk. Increased risks are likely if level or flow increase rapidly after rainfall, particularly if hydrological changes are also associated with deterioration in water quality. It is important to establish whether level or flow changes are a consequence of the influence of surface waters or a consequence of changes in pumping regimes. Where continuous fixed rate abstraction takes place, steady state conditions may be approached after a period although this can take months or even years. In some aquifers, a steady state may never be achieved because of seasonal variations in rainfall. Intermittent abstraction (eg to reflect changes in demand or take advantage of power tariffs) is being used increasingly. These factors cause short term, even diurnal, variations in water level in the vicinity of the abstraction point on top of the longer term variations in the aquifer as a whole. Where appropriate, data from observation boreholes remote from local influences should be considered.

4.4.2 Groundwater levels should be monitored regularly and compared with abstraction, rainfall and quality data. A rise in level will normally either be caused by reduced abstraction or by increased rainfall recharge. An unexpected rapid rise in level should be investigated and the possibility of ingress of water of recent surface origin should be considered, particularly if it can be correlated with recent rainfall or changes in water quality or water temperature. In the case of a spring, total flow variation will usually be the primary monitoring tool. However, this still needs to be considered in conjunction with groundwater level, quality and rainfall data.

**Construction characteristics**

4.4.3 Current practice for sinking new groundwater abstraction points involves drilling a borehole and casing with solid lining tubes. These are grouted in through drift and underlying strata until the lining reaches sufficient depth to create a sanitary seal. Below that point the borehole will continue with a solid or perforated casing, or as an open hole depending primarily on structural stability and hydrogeological factors. Such structures should present a low risk of contaminated surface water entering the borehole provided that linings and seals are maintained adequately, in accordance with recommendation 14.22 of the First Report of the Expert Group (Badenoch 1990).

4.4.4 At older abstraction sites, construction methods rarely reflect current standards, although many are in adequate sanitary condition. Some pose a higher risk because the fabric has deteriorated or because the nature of the recharge catchment has changed. In some cases the facilities were not originally intended for water abstraction, eg where abandoned mine workings are now used as a water resource. Many of these older abstraction points remain in use and even when abandoned can still present a pathway for contaminants to reach the aquifer. Shallow water-bearing strata present greater risks because they are more vulnerable to contamination and there is little or no opportunity of creating a secure

sanitary seal. An extreme example is the use of radial collector wells to abstract from gravel in a river valley. In these situations, the abstraction points should be considered as a surface water source. However, all sources with adits or galleries would comprise an inherently high-risk grouping.

4.4.5 Newer boreholes will have only limited data available for use in risk assessment, although they should have been constructed to a higher standard. Where intrusive maintenance has been carried out on older structures, existing records may no longer be representative. It may therefore be appropriate to assign a higher level of risk in these circumstances until sufficient relevant operating information has been obtained. The risks associated with construction factors should therefore be assessed on three broad sets of criteria: whether the location and surface geology favour a sound sanitary seal; whether a sound sanitary seal has actually been achieved; and the extent and reliability of knowledge of the abstraction structure and strata contributing water to the source.

## 4.5 Water quality testing

4.5.1 Monitoring raw (untreated) water from groundwater sources is essential for assessing risk from contamination by *Cryptosporidium*. Monitoring can vary from minimal surveillance for a few determinands to detailed investigations with automatic samplers and on-line continuous measurement. Table 4.3 and the following paragraphs review the application of monitoring activities in assessment of risk from *Cryptosporidium* in groundwater.

*Table 4.3* Application of monitoring activities in the assessment of risk

| EVIDENCE FOR *CRYPTOSPORIDIUM* RISK TO GROUNDWATER | SIGNIFICANCE |
|---|---|
| Detection of *Cryptosporidium* oocysts in source water | direct evidence of contamination |
| Detection of *Cryptosporidium* oocysts in distribution system sediments | evidence of recent or historic contamination |
| Regular detection of *E.coli* in source water | Indication of faecal contamination |
| Detection of *Clostridium perfringens* in source water | Possible surrogate for *Cryptosporidium* |
| Transient changes in turbidity of source water | Possible rapid influence of surface water |
| Micro-temperature or conductivity changes detected by down-well logging | May reveal influence of major inflows at shallow depths |
| Concentrations of certain characteristic dissolved inorganic species | May indicate recent surface water inflow |

4.5.2 Although there is no evidence for any consistent correlation between detection of *Escherichia coli* indicator bacteria and oocysts, the regular detection of *E.coli* does warrant investigation for *Cryptosporidium.* This is particularly the case where there is evidence of rapid recharge conditions or *Cryptosporidium*-generating activities within the catchment area. Nevertheless, *E.coli* does occur in the absence of oocysts and *vice versa*. This arises because: (i) the temporal distribution of *Cryptosporidium* in polluted groundwater recharge will be different to that of *E.coli*; (ii) differences in adsorption characteristics between oocysts and bacteria and (iii) greater persistence of oocysts in the subsurface compared with *E.coli*. Oocysts present a larger cross-section than the environmentally-stressed form of coliform bacteria and are likely to be

subject to stronger filtration effects in all but highly-fissured strata. The detection of oocysts in groundwater indicates that contamination has occurred and that further investigation to assess the overall risk is necessary. There is considerable interest in the feasibility of using simple and reliable surrogates for *Cryptosporidium* (Edberg *et al* 1997). There is some evidence that spores of the bacterium *Clostridium perfringens* may serve as a surrogate for *Cryptosporidium* in groundwater (DOE 1997) and in surface waters (Payment *et al* 1997). The former reference indicates also that monitoring for faecal streptococci provides a more reliable indication of faecal contamination than can be obtained from coliform monitoring, particularly at low sampling frequencies.

4.5.3 Several physical and chemical measures can help establish whether groundwater might be under the rapid influence of surface water. Of these, turbidity is most convenient and warrants further consideration. The presence of significant turbidity events in groundwater does not necessarily indicate the presence of oocysts, nor does low turbidity indicate the absence of oocysts. Sudden, unexplained peaks in groundwater turbidity should be investigated by use of particle size analysis and microscopic investigations to distinguish between aquifer material and organisms or debris derived from surface water bodies (Boutros 1992). There is a need for research into the mechanisms causing, and the significance of, turbidity in groundwater to establish the role of rapid influence by surface water and assessing the use of turbidity as a monitoring tool. Such investigations are currently limited by the restricted deployment of continuous turbidity monitoring at groundwater sources or the inadequate calibration and maintenance of installed equipment.

4.5.4 Logging of micro-temperature and micro-electrical conductivity in boreholes is a well established technique for identifying the depth and characteristics on major individual inflows (BSI 1988). Other chemical parameters such as chloride, potassium, phosphate, strontium, bromide and boron, as well as measurements of the concentrations of the stable oxygen and hydrogen isotopes have been used to determine the extent of surface water ingress (Edmunds *et al* 1976; Rodke 1981). However, in upper reaches of streams there may be little difference between surface and groundwater chemistry. Hydrogeological transport studies using tracers will greatly increase confidence in the diagnosis of rapid surface water ingress.

4.5.5 *Cryptosporidium* oocysts have been identified in mains and service reservoir sediments during investigation of suspected source contamination events. Such identification may provide evidence for intermittent contamination over a long time scale, although it is unlikely that oocysts in sediments will constitute an infection hazard. However, failure to identify oocysts in sediments does not provide confirmation that contamination of the water source has not taken place.

## 4.6 Guidance for water utilities

4.6.1 Water utilities should assess systematically the vulnerability of their groundwater sources to contamination by *Cryptosporidium* in the light of the risk factors set out in Tables 4.2 and 4.3 and take decisions concerning the need for provision of treatment or other appropriate actions. The assessment must take account of (i) the hydrogeological factors which render an aquifer prone to rapidly transmit oocysts; (ii) whether there are activities in the catchment which might constitute a

source of oocysts; and (iii) construction and design factors that might predispose the abstraction point to contamination. The risk assessment should be linked to a risk management programme as illustrated in Figure 4.1. The assessment of hazard and pathway factors should be carried out in liaison with the EA. Where there is doubt about the extent and reliability of hydrogeological information, appropriate surveys should be considered. Water utilities should also review at the same time whether groundwater assets comply with the recommendations in the First Report of the Expert Group for borehole seals and linings (Badenoch 1990).

4.6.2 Not all groundwater is consistently high quality. Utilities should be especially vigilant for the possibility of intermittent rapid transmission of water from the surface into boreholes, wells and springs. The catchment, resource and source characteristics should always be reviewed against water quality data. If it is necessary to undertake further work, sampling should include recharge periods or times when losing reaches of surface water are most contaminated.

4.6.3 Where an unacceptable risk is identified and treatment is already available, it is important to assess the effectiveness of the treatment process to remove or inactivate *Cryptosporidium*. Rapid gravity filtration (and granular activated carbon filtration) without chemical pre-treatment will be less effective than flocculation/sedimentation (or flotation) followed by filtration. Ion exchange and biological de-nitrification are unlikely to significantly remove *Cryptosporidium*. Ozone treatment for pesticide removal may reduce the viability of oocysts but effectiveness of treatment is dependent on temperature, ozone dose and contact time (Badenoch 1995). Only physical barriers will remove oocysts.

4.6.4 The isolation of oocysts in groundwater soon after rainfall recharge is a high risk circumstance which warrants immediate investigation. This should include an assessment of historical reported rates of human cryptosporidiosis. However, it is essential to ensure health authorities have used comparable reporting practices across the review period. A recommendation on making cryptosporidiosis laboratory reportable is made in paragraph 3.3.11. When assessing the significance for health of occasional exposure to low numbers of oocysts via groundwater, it is necessary to bear in mind that oocysts are not always absent from surface water derived drinking water. There is considerable evidence that occasional exposure to low numbers of oocysts does not lead to outbreaks of cryptosporidiosis (Parker *et al* 1993; National *Cryptosporidium* Survey Group 1992). For this reason identification of occasional low numbers of oocysts in water, in the absence of other risk factors, may not indicate the need for installation of treatment or other action. Local circumstances should be the defining factor.

4.6.5 After making a risk assessment, water utilities should assess the possibility of minimising risk of contamination by reviewing catchment control options or by operational improvements to the security or integrity of the groundwater source. In rare cases the risk may be so unacceptably high that treatment installation is required. In the majority of cases, however, it will be necessary to carry out surveys and further investigations to confirm an unacceptable risk of groundwater contamination with *Cryptosporidium* before adopting a treatment solution. All risk assessments should be regularly reviewed, especially following any significant change in the catchment, the condition of the water supply source, or the demand on the source.

## 4.7 Research needs

4.7.1 This section has concentrated on *Cryptosporidium* and its potential to contaminate groundwater. However, much of the preceding information also relates to the identification of general 'at risk' situations where there is potential for surface-derived contamination from a range of potential pathogens. **There appears to be a need for development of operational tools and a general guidance manual for the operation of groundwater abstraction and treatment processes.**

4.7.2 In addition, the following specific research needs have been identified:

(i) development of operational monitoring tools to improve the detection of rapid influence of surface water sources on the quality of groundwater;

(ii) transport and fate of *Cryptosporidium* and other pathogens in groundwater systems;

(iii) application of chemical and particulate tracers to investigate the transport and attenuation of pathogens in groundwater;

(iv) significance and nature of turbidity changes in groundwater and its role as a monitoring tool for rapid surface water ingress; and

(v) attenuation rates for *Cryptosporidium* in soils and unsaturated zones following application of farm wastes and sewage sludge to land.

## *4.8 Recommendations*

***4.8.1 Water utilities should systematically assess and rank the potential risk of groundwater contamination by Cryptosporidium by application of a tripartite approach which assesses source, catchment and hydrogeological factors.***

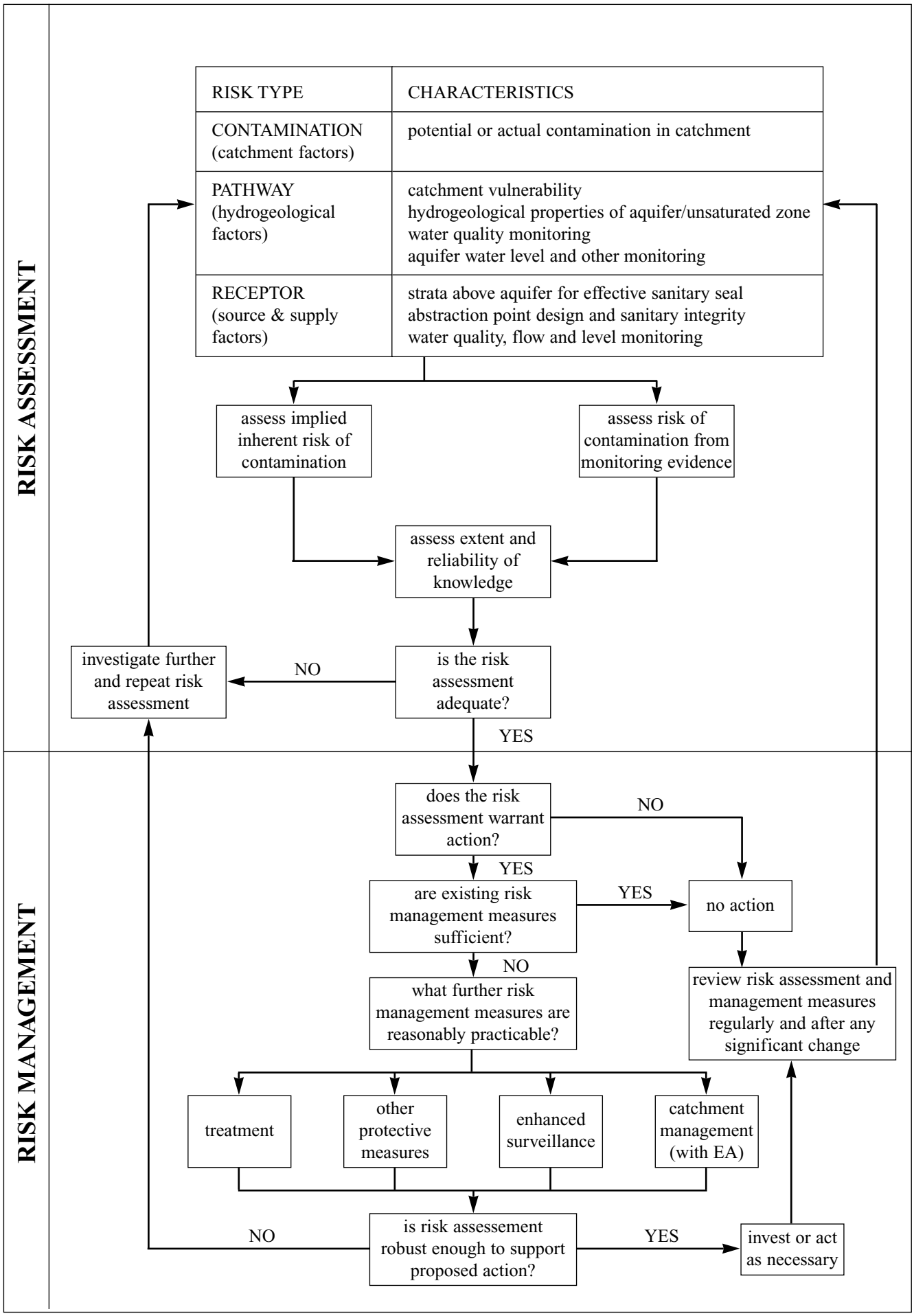
***4.8.2 Continued use should be made of existing national groundwater vulnerability maps and source protection zoning schemes to assess risk of contamination by Cryptosporidium.***

***4.8.3 For Cryptosporidium risk assessment, a fourth classification 'extreme vulnerability' is recommended for use with vulnerability maps and zoning schemes.***

***4.8.4 In order to ensure that groundwaters are properly protected from agricultural activity, the Ministry of Agriculture, Fisheries and Food should promote further the application of the the Code of Good Agricultural Practice – Water within the farming industry.***

***4.8.5 Careful attention should be given to the operational aspects of groundwater abstraction.***

## Figure 4.1: Groundwater risk assessment and management



## 4.9 Glossary of technical terms used in Chapter 4

**Abstraction –** The removal of water from surface water or groundwater, usually by pumping.

**Adit –** horizontal or near-horizontal tunnel extending outward from a well or shaft below the water table, designed to increase well productivity; passage from the surface by which a mine is entered and drained.

**Aquifer –** a permeable geological formation that is capable of both storing and transmitting water in significant amounts.

**Arenaceous –** a term applied to rocks that have been derived from sand or that contain sand.

**BGS** – British Geological Survey

**CCTV** – closed circuit television

**Collector well** – a particular design of well, usually constructed in unconsolidated alluvial or fluvio-glacial formations, comprising a central large-diameter shaft from which radial galleries or collectors have been driven to increase the production potential of the source.

**Dual porosity aquifer –** aquifer in which a certain proportion of the total storage capacity of the system is provided by the interstices in the rock matrix, while the fractures provide the dominant flow-path.

**Fissure flow –** the preferential flow of groundwater through dilated cracks, joints, bedding planes or other features of secondary porosity within an aquifer.

**Flow lines –** lines indicating the direction of groundwater movement.

**Fluvio-glacial –** pertaining to streams flowing from glaciers or to the deposits made by such streams.

**Groundwater –** naturally occurring sub-surface water in the saturated zone of a rock.

**Groundwater vulnerability –** the tendency or likelihood for contaminants to reach a specified position in the groundwater system after introduction at some location above the uppermost aquifer.

**Hazard** – a property or situation that in particular circumstances could lead to harm.

**High transmissivity** – capable of transmitting a large amount of water

**Hydraulic gradient –** the prevailing inclination of the water table or piezometric surface which provides the driving force to transmit groundwater through an aquifer.

**Intergranular flow –** flow occurring between the grains of a rock.

**Intrinsic aquifer vulnerability –** groundwater vulnerability determined without reference to the attributes and behaviour of particular contaminants.

**Karst (Karstic)** – an area of limestone or other highly soluble carbonate rock, in which the landforms are of dominantly solutional origin and in which the drainage is underground in solutionally enlarged fractures and conduits.

**Losing reaches of rivers –** locations in a watercourse where surface water is percolating through the bed of the watercourse into the underlying aquifer.

**Porosity –** the ratio of the volume of the interstices to the total volume of a rock, expressed as a fraction or a percentage. Effective porosity includes only the interconnected pore spaces available for groundwater transmission.

**Risk** – a combination of the probability, or frequency, of occurrence of a defined hazard and the magnitude of the consequences of the occurrence. **Risk estimation** is concerned with the outcome or consequences of an intention, taking account of the probability of occurrence; **risk evaluation** is concerned with determining the significance of the estimated risks for those affected, it therefore includes the element of risk perception; **risk perception** is the overall view of risk held by a person or group and includes both feeling and judgement; **risk assessment** consists of risk estimation and risk evaluation.

**Sinkhole** – the point at which a surface stream sinks underground.

**Soil leaching potential** – a composite measure of the ability of a soil to attenuate a diffuse source pollutant.

**Solution feature** – closed depressions a few metres to a few hundred metres in diameter and depth formed by solution action in soluble rocks, notably limestones.

**Source protection zones –** a series of concentric zones around an abstraction within which special policies apply to activities which might affect groundwater. The outermost zone covers the complete catchment area of the source, which is also called the well capture zone.

**Specific capacity** – the yield of a well per unit of draw down.

## References


ADRIS. (1995) Groundwater Protection Strategy for Scotland. Perth: Association of Directors and River Inspectors of Scotland.

Badenoch, J. (1990) *Cryptosporidium* in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Badenoch, J. (1995) *Cryptosporidium* in water supplies. Second Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 108pp.

Bell, D.F., McDonald, A.M., Morris, B.L., Lilly, A. (1997) Scotland's Minor Aquifers: A Scoping Study to Assess Groundwater Source Protection. BGS Technical Report WD/97/63. Edinburgh; BGS.

Boutros, S.N. (1992) Microscopic Particulate Analysis (MPA) in Studies of Groundwater. In *Proceedings of the Water Quality Technology Conference, Toronto, Ont.* Denver: American Water Works Association.

British Standards Institution. (1988) British Standard Guide for Geophysical Logging of Boreholes for Hydrogeological Purposes BSI 7022. BSI: London, UK.

DOE. (1997) *Cryptosporidium* Incidence in Private Water Supplies and Correlatory Indicators. Department of the Environment. London, UK.

DETR. (1997). Digest of Environmental Statistics. Department of the Environment, Transport and the Regions. London: TSO.

Edberg, S.C., LeClerc, H,. Robertson, J. (1997) Natural Protection of Spring and Well Drinking Water against Surface Microbial Contamination. *Critical Reviews in Microbiology* **23**(2), 179-206.

Edmunds, W.M., Owen, M., Tate, T.K. (1976) Estimation of Induced Recharge of River Water into Chalk. Boreholes at Taplow using Hydraulic Analysis, Geophysical Logging and Geochemical Methods. Report 76/5. London: Inst. Geol. Sciences.

Harvey, R.W. (1992) Transport of Protozoa Through an Organically Contaminated Sandy Aquifer. In *Proceedings of the First International Conference on Ground Water Ecology*. Tampa Fla: American Water Works Association.

Hancock, C.M., Rose, J.B. and Callahan, M. (1998) Crypto and Giardia in US groundwater. *JAWWA* **90** (3) 58-61.

Lawrence A.R., Williams A.T., Brewster L.J., Bowker J.A., Bird M.J., Ward R.S. (1996) Pathogen Transport in the Chalk Aquifer: an assessment of the risk to groundwater sources. British Geological Survey Report WD/96/48.

Lewis W.J., Foster S.S.D., Drasar B.S. (1982) The Risk of Groundwater Pollution by On-Site Sanitation in Developing Countries. Geneva: IRCWD.

MAFF. (1991) Code of Good Agricultural Practice for the Protection of Water. Ministry of Agriculture, Fisheries and Food. London, UK.

MAFF. (1998) Code of Good Agricultural Practice for the Protection of Water (The Water Code). Ministry of Agriculture, Fisheries and Food. London, UK.

National *Cryptosporidium* Survey Group. (1992) A survey of *Cryptosporidium* in Surface and Groundwaters in the UK. *Journal of the Institute of Water and Environmental Management.* **6** 697-703.

National Rivers Authority (1992) Policy and Practice for the Protection of Groundwater. Bristol: NRA.

Parker J.F.W., Smith H.V., Girdwood R.W.A. (1993) Survey of Loch Lomond to Assess the Occurrence and Prevalence of *Cryptosporidium* spp. Oocysts and their Likely Impact on Human Health. Report FR 0409. Marlow: Foundation for Water Research.

Payment P., Menard B., Prevost M. (1997) Enteric Viruses, *Giardia* and *Cryptosporidium* Level in the Raw Water of Water Treatment Plants along the Saint-Lawrence River. In *Proceedings of the Water Quality Technology Conference, Denver, Co.* Denver: American Water Works Association.

Rodke A. (1981) Spring Flood – Meltwater or Groundwater ? In *Nordic Hydrology* **12** 21-30.

UKWIR. (1998) Management of *Cryptosporidium* Risk in Groundwaters. UK Water Industry Research Limited, London, UK.

WSA. (1995) Wise Ways with Waste. Water Services Association. London, UK.

# 5 Advice to water utilities

## 5.1 Introduction

5.1.1 The purpose of this Chapter is to bring together the advice to water utilities contained in this Report for ease of reference. It should be read in conjunction with Chapter 3 and Chapter 4, section 6. The advice covers risk assessment, water treatment, monitoring and communications. Its aim is to minimise the possibility of *Cryptosporidium* oocysts entering drinking water supplies and to emphasise the need for water utilities to maintain good communications with health authorities and local authorities.

## 5.2 Risk assessment

5.2.1 The Group concluded that outbreaks of drinking water-related cryptosporidiosis do not just 'happen'. Worldwide there is an increasingly strong correlation between these outbreaks and inadequacies in drinking water treatment. A key element in providing appropriate treatment is the assessment of risk from *Cryptosporidium*. Risk assessment should be based on a combination of factors including the degree of exposure of the catchment to oocysts, the treatment processes currently in place and the history of cryptosporidiosis in the community. Monitoring systems and water treatment requirements should be reviewed against the level of risk.

*Recommendation*

***5.2.2 The Group recommends that water utilities carry out an assessment of risk from Cryptosporidium for each source and put in place a procedure for updating periodically the review of the risk assessment. Water treatment requirements and monitoring systems should be reviewed against the level of risk.***

***5.2.3 Water utilities should systematically assess and rank the potential risk of groundwater contamination by Cryptosporidium by application of a tripartite approach which assesses source, catchment and hydrogeological factors (4.8.1).***

## 5.3 Water treatment

5.3.1 As mentioned above, most waterborne outbreaks occurred due to deficiencies in water supply including those in which the treatment was inadequate or the works were operated above design capacity or some part of the treatment was bypassed. As recognised in the earlier Expert Group reports a conventional treatment works (that is, coagulation aided filtration) operated in accordance with good practice, is normally an effective barrier against *Cryptosporidium*.

5.3.2 The performance of chemical coagulation-based treatment for oocyst removal relies on the ability to maintain a suitable coagulant dose, governed by the raw water quality, particularly colour and turbidity. The subsequent solids-liquid separation processes must then be able to remove the coagulant solids to a very high degree. If a works is operated well in excess of its design capacity, or if some unit processes are by-passed, coagulant dosing and effective removal of the coagulant solids may be impaired, thus compromising the plant performance for oocyst removal.

However, some unit processes, such as second stage filters for manganese removal or post-filtration granular activated carbon adsorbers, are not designed for coagulant solids removal and overloading or by-passing of these should not compromise performance for oocyst removal.

5.3.3 The performance of filters is poorest immediately after restart, even if an effective backwash has been applied. Consistent with recommendations in the first two Expert Group reports, attempts should be made to minimise the impact on final water quality of increased turbidity after start up. A number of options are available to achieve this, the most effective being disposal of the first flush (perhaps with recovery for use as backwash water), or recycling to the head of the works. However, this may not be easy to retrofit at some works and more practical alternatives include improved backwash or delayed start (after backwash) of the filters.

5.3.4 United Kingdom Water Industry Research Limited has published a Guidance Manual supporting the water treatment recommendations from the first two reports of the Expert Group (UKWIR 1998). Its objective is to provide guidelines for water treatment works operation to minimise the risk from *Cryptosporidium*. The Expert Group endorses the general approach to water treatment recommended in this manual. The Group also makes the following recommendations on water treatment.

***Recommendations***

***5.3.5 Water treatment works should be designed to handle the typical peak turbidity and colour loadings in the source water.***

***5.3.6 Water treatment works should be operated at all times in a manner that minimises turbidity in the final water; attention should also be given to other parameters which reflect the performance of chemical coagulation, that is, coagulant metal concentration and colour.***

***5.3.7 Water treatment works should normally be operated within the design capacity and without by-passing of the solids-liquid separation processes which are responsible for removal of turbidity and coagulant solids; coagulation itself should never be by-passed or compromised.***

***5.3.8 In the event of an emergency, if it is necessary to overload or by-pass solid-liquid separation processes, a stringent monitoring regimen should be initiated to ensure that turbidity targets indicated in paragraph 5.4.4 below are not exceeded; if there is an indication that these targets will not be achieved, an immediate advice to boil notice should be issued.***

***5.3.9 For high risk sites, if minimisation of the effects of filter start up on final water quality cannot be achieved through more easily implemented changes (for example improved backwash or delayed start after backwash), modifications to the works should be made to allow the first flush to be run to waste or recycled to the works inlet.***

***5.3.10 Coagulation/flocculation processes should be checked regularly to meet changing conditions of source water quality and other environmental factors.***

***5.3.11 Only dedicated washwater mains should be used to carry the returned washwater flow.***

***5.3.12 Filters should be operated and maintained under optimum conditions with attention to the quality and depth of media and to the operation of the backwashing/air scouring system.***

***5.3.13 Treatment works staff should be trained to be aware of the potential effect on the final water quality of even very small changes in the catchment or the treatment stream.***

## 5.4 Monitoring

5.4.1 Investigation of waterborne outbreaks has shown that often there was a significant increase in turbidity at the time that the contaminated water was estimated to have entered supply. This is discussed in Chapter 3. The Group has made the following recommendations on monitoring.

*Recommendations*

***5.4.2 Water utilities should check that process monitoring systems are appropriate to the risk at each source.***

***5.4.3 For all sites at which Cryptosporidium might be a high risk, as determined by the risk assessment, monitoring should include continuous turbidity measurement on the outlet of each filter and on the final water using instruments capable of detecting changes of less than 0.1 NTU.***

***5.4.4 Water utilities should define for each of their treatment works the value and duration that constitute a significant deviation in turbidity of the final treated water irrespective of its relationship to the regulatory standard; for example it may be that at large water treatment works alarms should be set to be triggered by any increase in turbidity in the final water of greater than 50% of the normal average or suitably representative level; for small works, the increase of concern would vary and consideration should be given to the impact of the backwashing to individual filters.***

***5.4.5 Appropriate action procedures to react immediately to turbidity alarms, based on the level of risk and the history of the source/works should be in place; actions might include immediate sampling for Cryptosporidium, isolation of the filter(s) or source or, if suggested by history, the issue of advice to boil.***

5.4.6 Good experience has been reported with the operational use of particle counters which, when used in conjunction with turbidity monitors, can provide a more sensitive indication of particle breakthrough.

*Recommendation*

***5.4.7 The Group encourages the use of particle count monitors to provide additional information to that provided by turbidity measurements.***

5.4.8 Waterborne outbreaks occur even though oocysts cannot be detected in the water. This supports the general view that the 'contamination' occurs for only a few hours during which time it would be complete chance that routine samples coincided with the event. Random spot sampling is, therefore, unlikely to be effective for operational monitoring.

*Recommendations*

***5.4.9 The Group recommends that water utilities operating sites assessed as being at high risk give consideration to either:***

***(a) continuous sampling for Cryptosporidium with analysis times linked to turbidity monitoring results; or***

***(b) sampling triggered by turbidity events.***

## 5.5 Local working partnerships

5.5.1 The importance of good local working partnerships has been recognised in both of the earlier Expert Group Reports. Generally good working relationships and practices are in place. However, the Group wishes to emphasise again the importance of both Incident Management Teams and Outbreak Control Teams and of the role of the water utilities in ensuring that the required local advice and support is in place to respond quickly to changing operational circumstances. See also paragraphs 6.3.5 – 6.3.7.

*Recommendations*

***5.5.2 Water utilities should review their working relationships with local health authorities and environmental health officers in the form of Incident Management Teams. Criteria should be established for identifying outbreaks and procedures put in place for activating Outbreak Control Teams.***

***5.5.3 Water utilities, in liaison with health authorities, should set out criteria for decision-making on the issue and the withdrawal of notice on advice to boil water and review these with experience.***

***5.5.4 Should there be an outbreak of cryptosporidiosis, the water utility, as a member of the Outbreak Control Team, should encourage the use of good epidemiology recommended in this Report to establish the source of the outbreak, including whether illness is associated with the drinking water supply.***

***5.5.5 Water utilities should encourage Incident Management and Outbreak Control Teams to review and rehearse regularly the response procedures to incidents and outbreaks.***

**Reference**

UKWIR. (1998) Guidance Manual Supporting Water Treatment Recommendations from the Badenoch Group of Experts on *Cryptosporidium* (98/DW/06/5). UK Water Industry Research Limited. London, UK.

# 6 Advice on management of waterborne outbreaks of cryptosporidiosis

## 6.1 Introduction

6.1.1 The First Report of the Group of Experts on *Cryptosporidium* in Water Supplies 1990 (Badenoch 1990) made recommendations concerning the membership and terms of reference of Outbreak Control Teams (OCTs) in the event of an outbreak of cryptosporidiosis. The Second Report (Badenoch 1995) further recognised the need for an Incident Management Team (IMT).

6.1.2 Since that time an increased understanding of cryptosporidial infection has been gained, as too has experience in incident and outbreak investigations and management. IMTs and OCTs are seen as part of the overall arrangements by health authorities for the control of communicable disease and as such are very practical teams whose aim is the protection of public health by the prevention of infection. The teams are not collectors of evidence for legal purposes nor are they concerned with the maintenance of a commercial position by the water utility. The teams need to apply high standards of professional ethics and in particular ensure that patient confidentiality is maintained.

6.1.3 There are three components which underpin the management of waterborne outbreaks of cryptosporidiosis. The first is collaborative planning to develop response procedures, the second is recognising events which may put the population at risk and the third is early ascertainment by surveillance that there is an increase in human cases of cryptosporidiosis. The first of these requires advance collaborative planning; the second should trigger the convening of an IMT; the third should trigger the formal establishment of an OCT. In practice, the function and membership of both these teams is similar, to protect public health and return the situation to normal as soon as possible. The exact stage when an IMT will become an OCT will depend on each incident and local circumstances and could be early in the incident. This should be borne in mind in reading the following paragraphs. Relevant parts of the OCT checklist and meeting agenda in Appendix A3 will be useful equally to IMTs.

6.1.4 It is important to recognise however that there may be conflicting interests both within and outside the teams. The detail of information necessary to reach a conclusion for medical and public health needs may fail badly as legal evidence in a criminal prosecution and the resource required to collect different levels of evidence may vary considerably. It would be expected that, as the OCT is set up at the request of a Director of Public Health, it is primarily a team with a health objective. The Chair of the OCT should remind all team members of this at the outset.

*Recommendation*

***6.1.5 The Group recommends that it is essential that Outbreak Control Teams are aware at the outset of the scope and purpose of their brief and that there is a clear understanding of the roles, responsibilities and standing of each member.***

## 6.2 Incidents

6.2.1 Regulation 30(5) of the Water Quality (Water Supply) Regulations 1989 (SI No.1147) requires water utilities to inform district health authorities and local authorities as soon as possible of any event which gives rise to, or is likely to give rise to, a significant risk to health of persons residing in the authorities' areas. In practice this means that Consultants in Communicable Disease Control or Consultants in Public Health Medicine (CCDCs/CPHMs) and Environmental Health Officers (EHOs) are being informed of incidents during treatment and supply of drinking water. On receipt of this information the CCDC/CPHM and the EHO will have to decide its importance in public health terms and act accordingly. Whereas waterborne outbreaks are relatively rare, incidents during water treatment and supply are not, but except in rare instances they are not likely to be a serious public health hazard. In order to assess the risk an Incident Management Team (IMT), similar to the Outbreak Control Team (OCT), may need to be formed.

**Identifying an incident involving *Cryptosporidium***

6.2.2 An incident may be identified in a number of ways: by routine water sampling plus an increase in cryptosporidiosis in the community; sampling following operational work or repairs; by plant malfunction; or from consumer complaints. There is a legal requirement in the UK for public drinking water supplies to be monitored intensively by water utilities from source to tap. It must be done by bacteriological and chemical sampling, by visual inspection and organoleptic testing. Increasingly, continuous monitoring of treatment processes is carried out electronically at critical control points in the system. Despite this, circumstances can arise infrequently, where for short periods treatment of drinking water is not as effective as it should be.

6.2.3 Handling of water quality incidents is initially the responsibility of the water utility. Accordingly, utilities have internal procedures to deal with incidents. It is also likely that when serious incidents occur the local authority and the health authority will also become involved quickly. The key organisations likely to be involved in an incident should have written emergency procedures which can be put into action when an incident occurs.

6.2.4 An incident involving the breakthrough of *Cryptosporidium* oocysts through water treatment and into distribution may not be as obvious to identify as complete malfunctions such as disinfection or plant breakdown. This is covered in more detail in the section on advice to water utilities. It is possible that some water treatment plants may contribute to the background level of cryptosporidiosis in a community but usually the sources of background levels are not investigated or identified. The outcome of pre-outbreak assessments of potential problems, such as an increase in background numbers or presence of oocysts in water, should be explained when possible and the information disseminated to organisations likely to be represented on an OCT.

***Recommendation***

***6.2.5 To facilitate recognition of an incident involving Cryptosporidium, the Group considers there is a need for local studies to identify background levels of cryptosporidiosis, and for local risk assessments to be conducted so that any increased incidence can be identified easily.***

**Recording and communicating the incident**

6.2.6 All those involved at any stage of an incident should start a log-book immediately. This should include dates, times, key facts, summaries of telephone calls, and the actions taken by named staff. The need for systematic recording of events, particularly in the fast-moving early stages of an incident, should not be neglected as details will have an important role in the epidemiological study and in any subsequent review of lessons to be learned.

6.2.7 Communicating the incident is a key action. There is a statutory requirement to inform the local authority and health authority and the relevant Secretary of State (in practice DWI in England and Wales). Customers in the area affected should be advised as soon as possible and kept informed of progress. Circumstances will dictate who else needs to be informed. For example the fire brigade and the local renal dialysis coordinator must be told immediately of low water pressure or complete loss of supplies. Other outside organisations to be contacted may include the Environment Agency, the Ministry of Agriculture, Fisheries and Food, and the Ofwat regional customer services office. Important telephone messages to these organisations should be confirmed by facsimile transmission of the key points with the 'messages sent' output from these machines kept as objective evidence that a message was sent.

*Recommendation*

***6.2.8 The Group recommends that all those involved at any stage of an incident should start a log-book immediately. This should include dates, times, key facts, summaries of telephone calls, and the actions taken by named staff.***

**Assessment and management of incidents**

6.2.9 The next phase in managing an incident is the appointment of someone suitably senior at the water utility to act as Incident Manager to be in charge of taking remedial action to protect water supplies. The Incident Manager should check that sufficient human and equipment resources are available to cope, and that there are system drawings and road maps available in sufficient detail for the area affected. Key questions to be answered at this stage are who, and what, is at risk and from what kind of risk. Particular attention must be made to sensitive commercial and domestic water users and provision of alternative supplies may be needed, particularly for vulnerable groups such as nursing mothers, infants and the elderly and infirm.

6.2.10 The Incident Manager should consider the need to set up a local incident control room, appoint a duty officer and information officer, and assemble a support team. At the start of a major, or prolonged, incident it is common practice to draw up a shift system and if necessary send some of the team home immediately for rest before the first shift changeover.

6.2.11 The Incident Manager should regularly review the situation to ensure everything is under control. Evidence for this should be noted in the log-book. A local weather forecast can often help at this stage to see if actions planned for the next 72 hours will be affected. Resource requirements for the next 72 hours should be drawn up. Extra laboratory staff and work gangs may need to be placed on standby alert. Arrangements for out-of-hours working may need to be made.

6.2.12 External communications often become important at this phase of incident management. Dedicated telephone lines should be provided for use by the team dealing with the problem. Extra telephones and pagers should be provided if necessary: mobile phones can be invaluable during

an operational incident. This communication traffic must be kept separate from customer and press enquiries. Similarly, separate facsimile machine numbers should be dedicated for incoming and outgoing messages at the local incident room. Communications with outside organisations and customers, as well as other functions within the utility, should be dealt with by an information officer to allow the Incident Manager to concentrate on dealing with the situation.

6.2.13 The police force should be informed if it is likely that large numbers of water utility staff will be on the streets late at night either as part of the remedial action or delivering leaflets on 'boil water advice'. It is wise to provide utility clothing (for example, fluorescent jackets with the utility logo) to these employees for safety and security reasons. Plans to use large numbers of water utility staff on the streets should include details of local communications, transportation, provision of food, drink and petty cash, and issue of torches, notebooks and maps. If a very large number of people is to be advised to boil water it is sensible to warn the electricity utility of the likely extra power demand.

6.2.14 When an incident occurs the actions taken by public health physicians in the health authority will be similar to those undertaken by the water utility such as keeping a log-book and managing communications. It is important for the CCDC/CPHM to note in the log-book the time and date when first informed of the incident, and by whom, as the water utility has a statutory duty to inform the district health authority and the relevant Secretary of State promptly of any incident which is likely to give rise to a significant risk to health. The DWI will wish to know if the water utility has complied with this regulation when it subsequently investigates the incident. What constitutes a significant risk to health should be agreed in advance with all concerned.

**Establishment of an Incident Management Team**

6.2.15 Depending on the seriousness of the incident an IMT may need to be set up. A check should be made to see if other district health authorities are involved; if so they should be included in the team, and a decision made on which district takes the lead. Similarly when more than one district is involved the health authority (or health authorities, as the case may be) should be informed so they can take a coordinating role, although the investigation and management of the incident is a matter for the CCDCs/CPHMs involved as they have executive responsibility. If the incident is considered to be serious or a potential public health risk involving a large number of the population and where advice to boil water has been given or where there has been or is likely to be much media attention, the Chief Medical Officer of the relevant Government department, should be informed. In practice this can be done by telephoning the Senior Medical Officer responsible for drinking water at the relevant Government health department. These telephone numbers should be readily available and up-to-date.

6.2.16 The assessment of public health risk should be made as a matter of urgency by the CCDC/CPHM after notification from the water utility. It will be important to know at what stage in the treatment process, from source to tap, the incident has occurred. Is the problem in the raw water storage, in the treatment, or in the distribution system, including, post-treatment storage in service reservoirs? It is important to get as much information from the water utility as possible because this will help to establish how much treatment the water has had and where the potentially contaminated water is in the distribution system. It is also important to

clarify terms. For example, it is important to determine whether the term 'reservoir' relates to a raw water reservoir or a treated water, service reservoir.

6.2.17 It is also crucial to establish how long the problem had existed before its discovery and subsequent notification to the district health authority. Other questions to ask will include the possibility of mixing of the contaminated water with other sources, and the time it is likely to take for contaminated water to reach customers. Questions like these will enable the CCDC/CPHM to establish the degree of treatment the water has received – full, partial, or none at all – and for how long. The risk to health posed by the incident can then be assessed.

6.2.18 If the incident is sufficiently serious the CCDC/CPHM should set up an IMT, which should include the water utility, local authority and other relevant organisations such as the PHLS, CDSC, Environment Agency, Health and Safety Executive and the police. The members should either meet, or communicate frequently by telephone, to keep each other appraised of the situation. The use of pre-planned procedures for dealing with, for example, notification to customers of 'advice to boil water' is recommended. An important role of the IMT is to evaluate the possible outcomes of the incident, assess their impact on public health, and then plan for the best way to deal with each possible outcome. It may be necessary to seek the help and advice of others such as the regional epidemiologist, the local Public Health Laboratory director and possibly the PHLS Communicable Disease Surveillance Centre (CDSC). Medical colleagues at the Department of Health also provide help and advice to CCDCs/CPHMs both in and out of office hours.

6.2.19 The Expert Group recognises that the decision on advice to boil normally rests with the water utility and that it is impossible to lay down precise recommendations on the information to be made available because this will differ with each incident and local circumstances. However, as part of the decision-making process, criteria for removing the notice should be considered.

6.2.20 The Group recognises that there is a need for uniformity in the wording of advice on boiling water to ensure that the water is microbiologically safe whilst avoiding confusion to consumers and potential dangers with overheating electric kettles. It is necessary only to bring the water to the boil to kill *Cryptosporidium* oocysts. Water should be allowed to cool before use.

***Recommendation***

***6.2.21 The Expert Group recommends that all notices of advice to boil water issued to consumers should make it clear that it is only necessary to bring the water to the boil and then allow it to cool before use.***

6.2.22 The IMT should consider the need for press notices and briefing. If the media are used then information should be channelled through one central point. It is essential that any information given should be accurate. The spokesperson for the IMT is most likely to be the CCDC/CPHM or a member of the health authority management team. With regards to informing the public, a practical point to be borne in mind is that the use of loudspeaker vans can create confusion, as the message may be heard by people living in an unaffected area. If loudspeaker vans are used, the area affected should be clearly stated. The IMT should also prepare a report at the end of the incident. The CCDC/CPHM should take the lead in the

report's preparation and circulation to interested groups including the Chief Medical Officer of the relevant Government health department.

6.2.23 Other duties for the CCDC/CPHM will include checking that arrangements are in place for contact out-of-hours in case the situation deteriorates rapidly, or either the water utility or the local authority needs specialist advice. Checks should also be made to ensure that arrangements for hosting an IMT meeting are in place and that adequate communication links are available – if necessary at short notice.

6.2.24 If the response to the incident is to advise the potentially affected population to boil their drinking water, every effort must be made by the CCDC/CPHM to ensure that hospitals, residential institutions and vulnerable groups in the community, (for example, home dialysis patients) are informed as a matter of urgency. Arrangements for alternative water supplies may need to be made for these groups. Often the pre-planned 'boil water' procedures agreed between water companies, health authorities and local authorities will have set out arrangements for these tasks.

6.2.25 CCDCs/CPHMs will be informed about most incidents as soon as they occur and, if prompt action is taken by the district health authority, local authority and the water utility, it is unlikely that any illness will result. However, the CCDC/CPHM should consider the need to heighten surveillance for several weeks after the incident with the help of local general practitioners and microbiologists within the National Health Service (NHS) and PHLS.

6.2.26 All reported incidents, involving water supplies in England and Wales will be investigated by the DWI. The Inspectorate will wish particularly to know whether the health authority was informed promptly by the water utility, and if any illness resulted from the incident and whether the health authority considers that water unfit for human consumption was supplied. Its enquiries are usually made by correspondence with the health authority, local authority, and water utility involved. Although the Inspectorate will have received a copy of the IMT report, an individual response from each party is always sought. Responses from a health authority to the Inspectorate may be used in support of a criminal prosecution of a water utility, or individuals in the utility, so all correspondence may need to be cleared by regional health authority lawyers.

6.2.27 CCDCs/CPHMs should periodically undertake desktop exercises to check the working of any pre-planned procedures, whether for incidents or outbreaks, between the health authority, local authority, and the water utility. This is a useful way to ensure that arrangements for communications, especially out-of-hours, are kept up to date. A recommendation is made in paragraph 6.3.7 below. Liaison meetings with the water utility and the local authority should take place regularly to review data and plans.

**Role of local authorities when an incident occurs**

6.2.28 It is also a requirement of the Water Quality (Water Supply) Regulations 1989 (Anon 1989) that a local authority makes arrangements with the water utility to receive notification of incidents which may pose a significant risk to health. Written emergency procedures should be developed so that roles and responsibilities are clear. In practice this will require EHOs in local authorities to establish a working relationship with

the relevant offices of the water utility and district health authority. It is essential that contact telephone and fax numbers are exchanged, and 24 hour contact is available.

6.2.29 There should be prior consultation between the local authority, the district health authority and the water utility to determine which incidents may constitute a significant risk to health. A good working relationship should allow the water utility to notify the local authority as well as the district health authority in the early stages of an incident to enable EHOs to prepare for any necessary action. If the incident then develops to the extent that formal notification is required, the local authority will be better prepared to act.

6.2.30 Once an incident has been notified, local authorities will need to have their own internal procedures for action. The local authority will, on the basis of the information received, need to act accordingly. In most incidents, especially of a microbiological nature, the EHO must advise the occupiers of certain high risk premises, such as schools and premises dealing with food and, in conjunction with the CCDC/CPHM, other institutions such as nursing and residential homes. Additionally the EHO should inform the ward councillors and other relevant council officials. These arrangements should be worked out in advance and contact points listed in an Emergency Telephone Directory. The Food Hazard Warning System can be utilised by EHOs in appropriate circumstances to alert food businesses. The district health authority will normally alert hospitals and general practitioners but this must be agreed with the CCDC/CPHM in advance.

6.2.31 EHOs in conjunction with the CCDC/CPHM will need to consider the advice to give to high risk premises based on the incident. If advice to boil water is issued then serious consideration must be given to advising food-based businesses to stop using tap water in food preparation if the final product is not cooked or heat treated.

6.2.32 For schools and other institutions, boiling and cooling water may be impractical. Water utilities are likely to be able to provide safe alternative supplies by tanker. However, the microbiological quality of tanker water cannot be guaranteed and it too should be boiled. The provision of a supply of bottled water may be another alternative course of action. Immunocompromised people should note the advice in Chapter 8 about boiling bottled water.

6.2.33 The EHO supervising the incident for the local authority should keep a detailed timed log of the events as they unfold, including any notifications, and subsequent investigations, of illness from within the affected areas, as this will be useful in preparing a report to the regulators once the incident is over.

6.2.34 If, in the unlikely event that the incident poses such a serious risk to health that the IMT decides to suspend supply, the local authority will have to consider, along with the water utility and district health authority, the provision of alternative supplies. At this point it may be necessary to implement the local authority's overall emergency planning procedure. For further information on this topic see Dawson and West (1993).

**Closing down an incident**

6.2.35 The Incident Manager, in association with the IMT, should assemble the facts and establish the most probable explanation for the

incident. Alternative explanations should be considered but not eliminated until it is safe to do so, and then the reasons for their dismissal as possible causes recorded in the log-book.

6.2.36 Closing an incident should be deferred until it is safe to do so and until it is clear the situation will not deteriorate or the problem recur. A cardinal issue here is whether water supplies are judged to be back to normal. Without such a guarantee, it is not possible to close an incident down. Depending on the severity of the incident, it may be necessary to discuss progress with the CCDC/CPHM in the district health authority, the local EHOs, or the DWI before finally closing down an incident.

6.2.37 The final action once an incident is closed is often the preparation of a post incident report by the water utility. An important component in this will be a review of lessons learnt in order to minimise the risk of a recurrence of the incident. An Incident Control Team Report similar to an Outbreak Control Team Report may also be prepared especially if illness has resulted in the community.

## 6.3 Outbreaks

6.3.1 The first Badenoch Report (Badenoch 1990) recommended that the CCDC/CPHM should inform both the water utility and the local authority if there is sufficient evidence to suspect an outbreak of waterborne illness. These three organisations should have prepared jointly an Outbreak Control Plan (OCP). A water utility often covers much larger areas than either the health authority or the local authority and at first it might appear sensible that it should take the lead in arranging outbreak control meetings. Nonetheless, in matters of public health, especially hazards to human health in the community, it is the health authority and in particular the CCDC/CPHM who will normally take the lead role.

6.3.2 The purpose of the OCT is to ensure coordinated investigation, management and control of an outbreak of waterborne illness in the community. It must define the tasks of the OCT members and any relevant public and private bodies. It should include statements on lines of communication, when to call the team together, management and organisation of the team, control measures which may need to be taken, and plans for any epidemiological investigations.

**Establishment of the Outbreak Control Team**

6.3.3 Events or occurrences which could trigger the DPH or CCDC/CPHM to call together the OCT include:

- a greater number of cases of illness in a period of time than would normally be expected whether or not water-associated;
- the unexpected appearance of cases of cryptosporidiosis in more than one local authority district or more than one health authority district;
- a suspected, anticipated or actual major incident of drinking water contamination.

**Terms of reference for the Outbreak Control Team**

6.3.4 The responsibilities laid down in the first Expert Group Report (Badenoch 1990) are still relevant. They are:

- to review the evidence for an outbreak by examination of the epidemiological, microbiological and other data;

- to identify the population at risk to institute additional measures required to gather further relevant information;
- to decide on measures to control the outbreak and to protect other members of the community at risk;
- to make arrangements for the commitment of personnel and resources considered necessary;
- to monitor the implementation and effectiveness of the measures taken;
- to make arrangements for informing the public and media;
- to decide on the point at which the outbreak can be considered of no further significance;
- to prepare a report as soon as it is practicable on the outbreak and make recommendations for further action.

**Organisation and membership of the Outbreak Control Team**

6.3.5 In general, most members of an OCT should know each other and the organisations represented before an outbreak occurs. The Group emphasises the importance of liaison and team work in managing an incident or outbreak involving *Cryptosporidium* in the drinking water supply. The organisations concerned should meet regularly to discuss procedures and these should be rehearsed regularly (see also section 5.5).

***Recommendations***

***6.3.6 The Group recommends that all parties likely to be involved in an IMT or OCT should establish a working dialogue and trust, preferably prior to the emergency situation, so that when a major incident occurs it will be dealt with more effectively.***

***6.3.7 The Group also recommends that all parties regularly simulate incident and outbreak events to rehearse emergency procedures.***

6.3.8 The OCT membership should be kept to as small a number as possible while providing the necessary expertise and relevant representation. The core members of the team will include the CCDC/CPHM and a consultant microbiologist from the NHS or PHLS, a Regional Epidemiologist (England and Wales), the Chief Environmental Health Officer from the local authority, and a Water Supply or Water Quality Manager from the water utility based on local circumstances. The OCT will normally be chaired by the CCDC/CPHM. Minutes of meetings should be taken by a person assigned specifically for this duty who will also be capable of following up action points.

6.3.9 Relevant representation on the OCT will be interpreted in many ways but frequently as requiring every health authority and local authority to be represented by at least one person. This may not enhance the work of the OCT and the objective may be better achieved by a liaison group of all those concerned, with two representatives only on the OCT. These should be a CCDC/CPHM and an EHO of sufficient seniority to take decisions and allocate resources. Other participants who could be invited or co-opted to work with the core OCT are:

- Hospital manager
- Public Health Laboratory Service/Scottish Parasite Diagnostic Laboratory staff

- Communicable Disease Surveillance Centre/Scottish Centre for Infection and Environmental Health staff
- Consultant physicians
- Infection control nurse
- Community health visitors
- General Practitioner representative
- Hospital works engineers
- Press officer for the health authority or water utility
- Environmental Agency/Scottish Environmental Protection Agency representative
- Ministry of Agriculture, Fisheries and Food representative
- Health and Safety Executive representative

6.3.10 There is a strong argument for some members of the team being appointed on account of their experience in dealing with waterborne outbreaks previously or their knowledge of water treatment and distribution. This raises the question as to whether the management of outbreaks would be better served if there was a national panel of experts, members of which could take the lead role in outbreak investigation with the assistance of local health officials and the water company. Such a national panel could include epidemiologists, public health microbiologists and water engineers, all of whom have experience in the investigation of waterborne outbreaks of infection. A national panel of experts would supplement local knowledge, not replace it.

*Recommendation*

***6.3.11 The Expert Group recommends that a list of national experts who can be contacted in the event of an outbreak, be compiled. Consideration should be given to how the list should be compiled but it could include epidemiologists, public health microbiologists and water engineers with experience in the investigation of waterborne outbreaks of infection. Such experts would supplement local knowledge but not replace it.***

**Recognition, declaration and closure of an outbreak**

6.3.12 Circumstances will dictate how an outbreak may be recognised and there is always difficulty in deciding when to declare a genuine waterborne outbreak. The CCDC/CPHM is responsible on behalf of the district health authority for formal declaration of a waterborne outbreak after discussion with the local authority and water utility members of the OCT. Having made this difficult decision, the implementation of the previously agreed plan should follow automatically. Early consideration should be given to the criteria that will be required before the outbreak can be declared over. From the public relations standpoint it is important to inform the community that the outbreak is over as soon as this information is available.

6.3.13 It is normal for the CCDC/CPHM to prepare a final report on the outbreak with the help of the other members of the Team. An outline model for a report is given in Appendix A3. In addition to the report, the lessons learnt and how future outbreaks could be better managed must be

identified and recorded. This report should then be sent to key people in each organisation represented on the OCT. The Group recognises that the statutory powers for the investigation and control of communicable disease rests within the health authorities and the local authorities. However, it considers that OCT reports should be formally received and recommendations commented upon by the Drinking Water Inspectorate or its regulatory equivalents to ensure consistency and that any lessons learnt are communicated widely.

*Recommendation*

***6.3.14 The Group recommends that OCT reports on waterborne outbreaks should be formally received and recommendations commented upon by the Drinking Water Inspectorate or its regulatory equivalents.***

### 6.4 Press relations in management of an incident or outbreak

6.4.1 The media can play a valuable public information role during the course of an incident. Television and radio can be expected to issue announcements to back up the direct sources of information to consumers. However, at a fairly early stage there will also be investigative journalism which will have different objectives. It is important to distinguish between these two facets to ensure that necessary clear messages to consumers are not clouded by the premature conclusions of reporters.

6.4.2 Information for the public and media during an outbreak should be controlled from one location to be agreed by the Outbreak Control Team. However, it is becoming increasingly important for public health professionals to have a working knowledge of how the press operate and how to liaise with them, especially now that the general public are increasingly aware of medical issues through the media, whether it be newspaper, magazine, radio, television or the Internet.

6.4.3 Good relations should be established with the local press well before any incident or outbreak. This may be accomplished by inviting journalists to meet key members of staff at their place of work, taking the opportunity of explaining the nature of their work, including how they collaborate with other local agencies and departments in safeguarding the public health.

6.4.4 It is prudent that they place themselves in a position of being able to play an active part in setting any news story about water ‘problems’ in its correct context. However, a cautious approach should be adopted when providing journalists with information regarding a potential problem, to ensure that this is not misrepresented.

6.4.5 An initial enquiry by journalists is often best handled by obtaining the essential information being sought, and the background to the article or programme, and then for the relevant officer to inform them that expert advice will be taken after which the press will be contacted again.

6.4.6 It is prudent for the officers to make every effort to gain a breathing space to sort out their thoughts on the topic before giving permission to be quoted. The nature of the information being sought should be identified; ask whether anybody else is being approached about the topic; identify any deadlines and set an agreed time for a further interview.

### 6.5 Draft meeting agenda, checklist and outline report for Outbreak Control Teams

6.5.1 OCTs are unlikely to be convened often and it is possible that the Chairman and members will have little experience of dealing with any emergency. To help the team and to try to ensure a consistency of approach, the Group has drawn up a draft agenda for OCT meetings, a checklist as an *aide mémoire* for the OCT chairman and the outline of an OCT report. These are given in Appendix A3.

### 6.6 Questions asked frequently during an outbreak

6.6.1 Experience during outbreaks has shown that there are a number of questions asked frequently by members of the public of Outbreak Control Teams. To help the team the Group has drawn up a list of the most frequently asked questions together with some suggestions for briefing those dealing with the public. These are also given in Appendix A3.

## References

Badenoch, J. (1990) Cryptosporidium in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Dawson, A., and West, P., Editors. (1993) Drinking Water Supplies: A Microbiological Perspective. Department of Health, London:HMSO.

# 7 Guidance on the epidemiological investigation of outbreaks of infection

## 7.1 Introduction

7.1.1 There is a need for more general recognition that water is not the only source of *Cryptosporidium* infection in humans. The organism can be acquired from water, food, milk, swimming pools, contact with farm and domestic animals and person to person transmission. Some epidemiological surveys following outbreaks have been deficient and there is a need for greater consistency in the quality of investigations.

7.1.2 Outbreaks of illness associated with drinking water are uncommon but have the potential to affect large numbers of people. There are several problems unique to the investigation of outbreaks of infectious disease thought to be related to contaminated drinking water. These include:

- Some water treatment works have very large outputs of water which will supply a wide geographic area.

- The water supply to an area may be a blend from more than one treatment works, only one of which may be the source of contamination.

- By the time an outbreak is recognised the contamination incident may have passed and therefore pathogens are often absent from water samples.

- The presence of low numbers of *Cryptosporidium* oocysts in water does not imply that water is the source of the outbreak.

- General exposure of the population to mains water may obscure the actual source of contamination.

- Gastroenteritis, the illness most often caused by waterborne outbreaks, is not uncommon in the population and the majority of cases are not due to water.

- Many pathogens associated with water can also be transmitted in other ways such as food and milk, animals or person to person contact.

## 7.2 Guidance Manual

7.2.1 High quality epidemiological information is vital in the investigation of possible outbreaks of waterborne infection associated with mains water consumption because microbiological evidence of water contamination by pathogenic organisms is usually difficult to obtain. Epidemiological investigations of such outbreaks are not straightforward; they are relatively uncommon and may be statistically complex, such that individual consultants in communicable disease control (CCDCs) or consultants in public health medicine (CPHMs) may appreciate help with investigations.

7.2.2 Guidance has been produced to give CCDCs/CPHMs and other members of the Outbreak Control Team (OCT) practical advice to assist in the conduct of epidemiological studies. It is aimed at the investigation of outbreaks that may be associated with the consumption of mains water. As *Cryptosporidium* is the most commonly reported organism involved in such outbreaks the emphasis is on investigation of outbreaks caused by this organism. However, the general principles set out in this guidance will apply to all outbreaks of potentially waterborne infection and to the investigation of all alternative hypotheses. The manual is not intended to replace local outbreak control plans, but it is hoped that CCDCs/CPHMs will find it a useful supplement and that its use will lead to the establishment of an agreed best practice in the investigation of water associated infection. The guidance manual is given in Appendix A4.

## 7.3 The Group's advice

7.3.1 The Group recognises that high quality epidemiological information is vital to the investigation of possible outbreaks of waterborne infection associated with mains water consumption because microbiological evidence of water contamination by pathogenic organisms is often difficult to obtain and even when it is available, such evidence is rarely conclusive. Some previous epidemiological surveys have been deficient and there is a need for greater consistency in the quality of investigations. The Group commends the use of the Guidance on the Epidemiological Investigation of Outbreaks of Infection Associated with Mains Water (Appendix A4 of this Report) to assist in the conduct of epidemiological studies for CCDCs/CPHMs and other members of OCTs.

*Recommendation*

***7.3.2 The Group recommends the Chairman and members of the Outbreak Control Team use the Guidance on the Epidemiological Investigation of Outbreaks of Infection (Appendix A4 of this Report) in all outbreaks where waterborne infection is suspected.***

# 8 Advice to the immunocompromised individual

## 8.1 Introduction

8.1.1 *Cryptosporidium parvum* is a highly infectious protozoan parasite responsible for cryptosporidiosis in humans and many animals. *C.parvum* oocysts discharge sporozoites which then attach to and replicate in the intestinal epithelium, causing changes in electrolyte handling (Griffiths *et al* 1994). The initial attachment of the parasite to host cells is a pre-requisite for the pathophysiological events in infection (Joe *et al* 1998). Immunocompetent individuals experience a transient diarrhoea, while those with impaired immunity, such as AIDS patients, are unable to clear the infection and severe diarrhoea (McGowan *et al* 1993) and cholangitis (Forbes *et al* 1993) may result. Cryptosporidiosis in the immunocompromised subject often results in a chronic life-threatening gastroenteritis with a high mortality (Flanigan *et al* 1992; Blanshard *et al* 1992).

8.1.2 Immunocompetent hosts respond to infection with antibody production and the secreted antibodies appear to reduce parasite numbers in the intestine. Nevertheless, antibodies to *C.parvum* do not seem to be able to protect AIDS patients from heavy parasite burdens (Goodgame 1996) and it seems likely that cell-mediated immunity is important for recovery from *C.parvum* infection. In HIV-infected patients there is a clear relationship between disease severity and CD4 counts (Flanigan *et al* 1992; Blanshard *et al* 1992). However, in many other conditions which result in impaired immunity the outcome and severity of cryptosporidial infection has not yet been identified clearly. For example *C parvum* was identified recently as an important pathogen in boys with the hyper-immunoglobulin M (hyper-IgM) syndrome (Hayward *et al* 1997). Several other host defence factors are also thought to contribute to *C.parvum* immunity, including the cytokines IFN-γ and IL12. It is not surprising therefore that infection by *C.parvum* could readily occur in several primary and secondary immunodeficiency states (Cosyns *et al* 1998).

8.1.3 The following advice is aimed at immunocompromised individuals. This includes HIV infected persons and other patients immunocompromised as a result of conditions such as: hypo- or agammaglobulinaemia, hyperimmunoglobulin M syndrome, severe combined immunodeficiency, leukaemia (especially during aplastic crises); or as a result of therapy with immunosuppressive drugs, who may wish to take independent action to reduce the risk of waterborne cryptosporidiosis and may choose to take the precautions recommended below.

## 8.2 Prevention of exposure

8.2.1 Until an effective therapy for *C.parvum* is available, informing immunocompromised patients of potential exposure risks to *Cryptosporidium* may be the most useful course of action. They should be educated and counselled about the variety of ways *Cryptosporidium* can be transmitted. Modes of transmission include:

- contact with infected adults and nappy-aged children;
- contact with infected animals;
- drinking contaminated water;
- contact with contaminated water during recreational activities; and
- eating uncooked food and food (such as fruits and salad) that has been washed with contaminated water.

8.2.2 *Cryptosporidium* may be spread by the faecal-oral route of transmission. Person-to-person and animal-to-person transmission has long been recognised (Fayer and Ungar 1986). Soil contaminated with human or animal faeces and the water that drains through it to rivers, streams and shallow underground wells are also potential sources of cryptosporidial infection. Immunocompromised persons should avoid contact with human and animal faeces. They should be advised to wash their hands after contact with human faeces (eg after nappy changing), after handling pets and after gardening or other contact with soil. They should avoid sexual practices that may result in oral exposure to faeces (eg oral-anal intercourse).

8.2.3 Cryptosporidiosis occurs more commonly in young animals (Current 1987). Immunocompromised persons should be advised that newborn and very young pets may pose a small risk of cryptosporidial infection but generally they should not be advised to destroy or give away pets.

8.2.4 Immunocompromised persons contemplating the acquisition of a new pet should avoid:

- bringing any animal that has diarrhoea into their households;
- purchasing a dog or cat aged less than six months; and
- adopting stray pets.

8.2.5 Immunocompromised persons should also avoid exposure to farm animals such as calves and lambs and premises where these animals are raised.

### 8.3 Advice on the prevention of waterborne exposure

8.3.1 *Cryptosporidium* oocysts are found commonly in natural waters. Immunocompromised persons should not drink water directly from lakes and rivers. Waterborne infection may also result from swallowing water during recreational activities. Patients should be aware that many lakes, rivers, salt water beaches and some swimming pools (Anon 1994) and recreational water parks may be contaminated with human or animal waste that contains *Cryptosporidium*. Patients should avoid swimming in water that is likely to be contaminated and should avoid swallowing water during swimming.

8.3.2 Several outbreaks of cryptosporidiosis have been linked to public water supplies. During outbreaks, or in other situations in which ‘advice to boil water for drinking’ is issued, bringing the water to boiling point will eliminate the risk of cryptosporidiosis. Use of submicron personal use

filters (ie home/office types) may reduce the risk (Addiss *et al* 1996) but cannot be relied upon to eliminate it completely. Persons who opt for a personal use filter should be aware of the complexities involved in selecting appropriate products, the purchase and running costs of the products and the logistic difficulty in using them consistently. Manufacturer's instructions should always be followed.

8.3.3 The magnitude of the risk of acquiring cryptosporidiosis from drinking water in a non-outbreak situation is uncertain. As a precautionary measure, to reduce the risk of waterborne cryptosporidiosis, HIV infected persons with low CD4 counts should be advised to bring to the boil all drinking water from any source. Such individuals should always be advised to bring to the boil drinking water drawn from private domestic water supplies as these have a much higher risk than public supplies of contamination by *Cryptosporidium* and may have inadequate treatment (Clapham 1997). They should be aware that places such as campsites and remote holiday accommodation may rely on private water supplies. The limited evidence available suggests that bottled water cannot be regarded as universally safe for immunocompromised persons and should be boiled before drinking.

8.3.4 Immunocompromised persons should be advised that ice made from contaminated tap water might also be a source of *Cryptosporidium*. Ice made at home should be prepared from boiled water. Such persons should also be aware that fountain beverages served in restaurants, bars, theatres and other places may also pose a risk because these beverages, as well as the ice they contain, are made from tap water.

8.3.5 National distributed brands of frozen fruit juice concentrate are safe if the user reconstitutes them with boiled water. Fruit juices must be kept refrigerated from the time they are processed to the time of consumption; only those juices labelled as pasteurised should be considered free from *Cryptosporidium*. Outbreaks associated with unpasteurised apple cider have been reported (Mshar *et al* 1997). Other pasteurised beverages and beers are considered safe to drink. No data are available concerning survival of *Cryptosporidium* oocysts in wine.

8.3.6 Cryptosporidiosis in immunocompromised people often results in a chronic life-threatening gastroenteritis with a high mortality. Whilst the Group recognises that the occurrence of *Cryptosporidium* in treated water is very rare it considers that the following recommendation will minimise the risk to immunocompromised people from drinking water.

***Recommendation***

***8.3.7 The Group recommends that all water, from whatever source, that might be consumed by immunocompromised persons should be brought to the boil and allowed to cool before use.***

## References


Addiss, D.G., Pond, R.S., Remshak, M., *et al*. (1996) Reduction of risk of watery diarrhoea with point- of-use water filters during a massive outbreak of *Cryptosporidium* infection in Milwaukee, Wisconsin 1993. *Am J Trop Med Hyg* **54**, 549-53.

Anonymous. (1994) *Cryptosporidium* infections associated with swimming pools – Dane County, Wisconsin, 1993. *MMWR* **43**, 561-3.

Blanshard, C., Jackson, A.M., Shanson, D.C., *et al*. (1992) Cryptosporidiosis in HIV seropositive patients. *QJM* **85**, 8134-43.

Clapham, D. (1997) The incidence of *Cryptosporidium* and *Giardia* in private water supplies, correlatory indicators and the value of the coliform standard in assessing water quality**.** *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick.* ed. Morris, R. and Gammie, A., 220-225.

Cosyns, M., Tsirkin, S., Jones, M., *et al*. (1998) Requirement for CD40-CD40 ligand interaction for elimination of *Cryptosporidium parvum* from mice. *Infect Immun* **66**, 603-607.

Current, W. L. (1987) *Cryptosporidium*: its biology and potential for environmental transmission. *CRC Crit Rev Environ Control* **17** , 21-51.

Fayer, R. and Ungar, B.L. (1986) *Cryptosporidium* spp. and cryptosporidiosis. *Microbiol Rev* 50: 458- 83.

Flanigan, T., Whalen, C., Turner, J., *et al*. (1992) *Cryptosporidium* infection and CD4 counts. *Ann Intern Med* **116**, 840-2.

Forbes, A., Blanshard, C. and Gazzard, B. (1993) Natural history of AIDS related sclerosing cholangitis: a study of 20 cases. *Gut* **34**, 116-21.

Goodgame, R.W. (1996) Understanding intestinal spore-forming protozoa: cryptosporidia, microsporidia, isopora and cyclospora. *Ann Intern Med* **124**, 429-41.

Griffiths, J.K., Moore, R., Dooley, S., *et al*. (1994) *Cryptosporidium parvum* infection of Caco-2 cell monolayers induces an apical monolayer defect, selectively increases transmonolayer permeability and causes epithelia cell death. *Infect Immun* **62**, 4506-14.

Hayward, A.R., Levy, J., Facchetti, F., *et al*. (1997) Cholangiopathy and tumours of the pancreas, liver and biliary tree in boys with X-linked immunodeficiency with hyper-IgM (XHIM). *J Immunol* **158**, 977-83.

Joe, A., Verdon, R., Tzipori, S., *et al*. (1998) Attachment of *Cryptosporidium parvum* sporozoites to human intestinal epithelial cells. *Infect Immun* **66**, 3429-32.

McGowan, I., Hawkins, A.S. and Weller, I.A. (1993) The natural history of cryptosporidial diarrhoea in HIV-infected patients. *AIDS* **7**, 349-54.

Mshar, P.A., Dembek, Z.F., Cartter, M.L., *et al*. (1997) Outbreaks of *Escherichia coli* O157:H7 infection and cryptosporidiosis associated with drinking unpasteurised apple cider – Connecticut and New York, October 1996. *J Am Med Assoc* **277**, 781-2.

# 9 Current therapeutic approaches to cryptosporidiosis

## 9.1 Introduction

9.1.1 Infection by *Cryptosporidium parvum* causes self-limiting gastroenteritis of approximately two weeks duration in immunocompetent hosts. After an incubation period of 7-10 days the patient presents with watery diarrhoea, nausea and vomiting and abdominal pain in about 50% of cases. In a minority (36%) there is a febrile illness (Fayer and Ungar 1986). In immunosuppressed hosts, such as patients with AIDS and CD4 counts of ≤ 200 x 106/L, the disease is much more severe (Current and Garcia 1991; Flanigan *et al* 1992). The patients usually develop severe malabsorption and most patients with AIDS never clear the infection. Although asymptomatic infection of such patients has been documented, more than half develop a chronic illness, and about 10% develop fulminant disease (Blanshard *et al* 1992).

9.1.2 Specific treatment for cryptosporidial gastroenteritis is generally not needed for immunocompetent patients, but such therapy would potentially benefit immunosuppressed patients or those developing a febrile illness. At present there is no accepted specific therapy for cryptosporidiosis.

## 9.2 Pathogenesis

9.2.1 Infection is initiated by ingestion of the oocyst with subsequent excystation and release of sporozoites upon exposure to bile salts, although spontaneous excystation can occur. It is found primarily in the small bowel and is located at the luminal surface of the epithelial cells or occasionally just within the brush border of the intestinal epithelium (Clayton *et al* 1994).

9.2.2 Both cellular and humeral immunity play a role in infection. In adult BALB/c mice it has been shown that both CD4 + lymphocytes and interferon- are required to prevent initiation of infection, while CD4 cells also can limit duration and interferon-γ limits intensity (Ungar *et al*. 1991: Chen *et al*. 1993). Experiments have shown that both colchicine and vinblastine inhibit *C. parvum* infection in a concentration dependent manner, which suggests that microtubules are important in host-cell invasion and may represent targets for development of new therapeutic drugs (Wiest *et al* 1993).

## 9.3 Cell cultures and animal models

9.3.1 Cell cultures and animal models have been widely used to identify pathophysiologic mechanisms involved in human cryptosporidiosis and to screen candidate therapeutic agents (Woods *et al* 1996). *In vitro*, permissive cell lines provide useful models for the study of their interactions with the parasite, their regulatory consequences such as mediator secretion and the influence of other systems such as cells and mediators of the immune compartment. Ex vivo systems (for instance isolated ileum) provide useful clues to the understanding of alterations of

electrolyte secretions by intestinal mucosa infected with *C. parvum*. In rodent (mouse and rat) models of cryptosporidiosis, the role of immune response in the control of the infection has been established, although differences with human illness (such as the absence of diarrhoea) preclude direct comparisons. Screening of potential anti-cryptosporidial agents performed *in vitro* using enterocytic lines (HCT-8 or Caco-2) needs to be confirmed in rodent models which involve pharmacokinetics characteristics. Moreover, these models of intestinal, biliary or respiratory cryptosporidiosis mimic histological, but not functional alterations of human cryptosporidiosis. In this context, models of goat or calf cryptosporidiosis may provide a better approach for pathophysiologic and pharmacologic studies.

## 9.4 Therapy

9.4.1 Therapy can be either non-specific (for example fluid replacement) or specific (the addition of an antiprotozoal agent). The majority of cryptosporidial infections in immunocompetent patients are self-limiting and usually resolve spontaneously. However, there are exceptions which require the maintenance of fluid balance and such treatment for cryptosporidiosis as is currently available. The majority of immunocompromised patients (for example those with AIDS) have severe chronic diarrhoea and debilitating illness which will require specific drug therapy.

9.4.2 Although more than 95 compounds have been tested in patients with cryptosporidiosis, only a very limited number have been shown to have activity against *Cryptosporidium parvum*; these agents are usually suppressive rather than curative. Antiprotozoal agents with some clinical efficacy are described below.

- *Albendazole* – high dose albendazole (800 mg) twice daily for two weeks has been reported to improve symptoms and eradicate the parasite in four Zambian AIDS patients (Kelly *et al* 1998). This is a preliminary study and requires confirmation by larger controlled trials;

- *Azithromycin* – the first of the azalide antibiotics, has demonstrated good effectiveness to date in immunocompetent animal models of cryptosporidiosis. It also has shown modest activity at a dosage of 600 mg/d in HIV-positive patients who have active cryptosporidial infection, and a rapid clinical and parsitologic cure at a dosage of 1200 mg/d in an immunocompetent patient (Bessette and Amsden 1995).

- *Diclazuril* – a benzeneacetonitrile derivative which is used as an anti-coccidial agent in poultry. Its use was associated with anecdotal reports of subjective improvement, but eventually proved ineffective when tested in controlled studies.

- *Letrazuril* – a diclazuril analogue with enhanced bioavailability, has been shown to be somewhat effective but many patients experienced adverse reactions. Development of the drug has ceased and it is no longer available.

- *Nitazoxanide* – a 5-nitrothiazole derivative which has broad anti-parasitic spectrum which includes coccida and flagellate protozoa, amoeba, nematodes, cestodes and trematodes. In a preliminary open study in AIDS patients in Mali, nitazoxanide (500 mg) twice daily for seven days eradicated or produced a >90% reduction in *C. parvum* oocyst excretion in 7 of 12 patients with cryptosporidiosis and stage IV AIDS (Doumba *et al*, 1997). Further studies are underway to confirm the efficacy of this drug against cryptosporidiosis.

- *Octreotide* – a somatostatin synthetic analogue that inhibits secretory diarrhoea, has been associated only with symptomatic improvement, not parasitic cure.

- *Paromomycin* – a non-absorbable, oligosaccharide aminoglycoside, has demonstrated symptomatic improvement, and possible parasitological cure in a small series of patients, but patients required maintenance therapy to prevent relapse (see paragraph 9.4.4 below).

- *Spiramycin* – a macrolide antibiotic that has been used for various types of infections in Europe for the past 20 years, had anecdotal reports of putative cure or symptomatic improvement in enteric cryptosporidiosis patients by 1983, but controlled clinical trials in patients infected with HIV demonstrated poor efficacy of both oral and intravenous formulations.

9.4.3 Only paromomycin and, to a lesser extent, azithromycin have shown some benefit for patients. Paromomycin has been shown to have an anti-cryptosporidial effect in *in vitro* assays (Datry *et al* 1992; Marshall and Flanigan 1992), animal models (Fayer and Ellis 1993a,b, Regh 1994, Tzipori *et al* 1994, Verdon *et al* 1994), and uncontrolled clinical evaluations (Armitage *et al* 1992; Bissuel *et al* 1991; Fichtenbaum *et al* 1993; Gathe *et al* 1990). Paromomycin in a dose of 25 to 35 mg/kg/day has a beneficial but limited effect upon oocyst shedding and stool frequency in AIDS patients (White *et al* 1994). Paromomycin is probably the most promising compound for human treatment.

9.4.4 In the dexamethasone-treated rat model of cryptosporidiosis paromomycin has been shown to be effective at a dosage of 50mg/kg/day or more for ileal infection, and 200mg/kg/day or more for caecal infection. The effect was thus shown to differ according to the anatomical site of the infection. At one and three weeks after treatment, a persistent infection was demonstrated in all rats, indicating that no eradication of the parasite could be observed even when high-dosage regimens up to 400mg/kg/day were used (Verdon *et al* 1995). These results confirm the anti-cryptosporidial activity of paromomycin and underscore the limitations of this compound because of its potential toxicity at such high dosages and its inability to eradicate the infection. They suggest that only a beneficial effect on symptoms rather than a clearing of the infection may be expected from increasing the dose in humans. This was borne out in a prospective trial for cryptosporidiosis in forty four severely immunocompromised individuals with HIV-related cryptosporidiosis. Although almost half of all patients had a clear clinical response, only 4 (9%) had resolution of diarrhoea and clearance of oocysts. Paromomycin often resulted in symptomatic improvement, but rarely ‘cured’ infection (Flanigan *et al* 1996).

9.4.5 Azithromycin was active in the dexamethasone-treated rat model (Regh 1991), but few data for human patients are available (Vargas *et al* 1993). However, azithromycin treatment of four children with AIDS who had severe diarrhoeal illnesses in which *Cryptosporidium parvum* was the sole pathogen detected was reported recently to be favourable (Hicks *et al* 1996). Three of these children had a marked decrease in stool volume and frequency within 36 hours of initiating therapy and resolution of diarrhoea within five days; *Cryptosporidium* became undetectable on examination of stool or colonic biopsy or by both after therapy was discontinued. A fourth patient however required prolonged therapy with azithromycin to achieve clearance.

9.4.6 Other chemical compounds have been shown to reduce the intensity of the infection by *C. parvum* in animal models, but such agents are not available for use in humans (Brasseur *et al* 1993).

9.4.7 **Immunotherapy.** Preliminary data suggest that administration of hyperimmune colostrum can decrease diarrhoea and may in some instances result in oocyst eradication (Tzipori *et al* 1986; Greenberg & Cello, 1996). Further trials are required to confirm efficacy and wider applicability to the treatment of cryptosporidiosis.

### 9.5 Prevention of recurrence

9.5.1 While primary exposure frequently results in proven infection and symptomatic disease in healthy immunocompetent adults previously seronegative for *C.parvum* (DuPont *et al* 1995), the resulting susceptibility to reinfection and illness is unknown. Although seroconversion may occur, the *C.parvum* serum antibody response does not appear to correlate with the presence or absence of infection (Okhuysen *et al* 1998). Epidemiological data obtained for Brazilian children suggest that primary infection with *C.parvum* does not completely block reinfection upon subsequent exposure (Newman *et al* 1994) but may protect the host against clinical illness (Current and Bick 1989). However, in these studies, recurrent infections were described in healthy adults and in high risk populations in areas with high seroprevalence for the disease, suggesting that repeated infection may result in clinical disease. In immunocompromised patients the protective nature of the antibodies to *Cryptosporidium* that may still be present, remains uncertain. No drug regimens are known to be effective in preventing the recurrence of cryptosporidiosis.

### 9.6 Overall conclusion

9.6.1 No antimicrobial agent has yet proved curative in adequate randomised double-blind controlled trials. However, there have been a number of encouraging reports on the use of paromomycin, and albendazole and nitazoxanide may have some clinical use in cryptosporidiosis. A number of other agents including azithromycin, have shown some limited therapeutic effect. No drug regimens are known to be effective in preventing the recurrence of cryptosporidiosis.

*Recommendation*

***9.6.2 The Department of Health should continue to keep work in progress under review and encourage further controlled trials of new agents as they become available.***

## References


Armitage, K.T., Flanigan, J., Carey, J., *et al.* (1992) Treatment of cryptosporidiosis with paromomycin. A report of five cases. *Arch Intern Med* **152**, 2497-99.

Bessette, R.E. and Amsden, G.W. (1995) Treatment of non-HIV cryptosporidial diarrhoea with azithromycin. *Ann Pharmacother* **29**, 991-3.

Bissuel, F.L., Cotte, M., Rabodonirina, M. *et al*. (1994) Paromomycin: an effective treatment for cryptosporidial diarrhoea in patients with AIDS. *Clin Infect Dis* **18**, 447-49.

Blanshard, C., Jackson, A.M., Shanson, D.C., *et al*. (1992) Cryptosporidiosis in HIV-seropositive patients. *Quart J Med* **85**, 813-23.

Brasseur, P., Lemeteil, D. and Ballet, J.J. (1993) Curative and preventative anti-*Cryptosporidium* activities of sinefungin in an immunosupressed adult rat model. *Antimicrob Agents Chemother* **37**, 889- 92.

Chen, W., Harp, J.A. and Harmsen A.G. (1993) Requirements for CD4+ cells and gamma interferon in resolution of established *Cryptosporidium parvum* infection in mice. *Infect Immun* **61**, 3928-32.

Clayton, F., Heller, T. and Kotler, D.P. (1994) Variation in the enteric distribution of cryptosporidia in acquired immunodeficiency syndrome. *Am J Clin Pathol* 102,: 420-25.

Clezy, K.J., Gold, J., Blaze, J. and Jones, P. (1991) Paromomycin for the treatment of cryptosporidial diarrhoea in AIDS patients. *AIDS* **5**: 1146-47.

Current, W.L. and Garcia, L.S. (1991) Cryptosporidiosis. *Clin Microbiol Rev* **4**, 325-58.

Datry, A.O., Rogeaux, E., Caumes, E., *et al*. (1992) Effects of aminosidine sulfate on *Cryptosporidium parvum* in cell culture and in AIDS patients. *Proc 3rd Eur Conference on Clin Aspects and Treatment of HIV Infect*, Paris. 175.

Doumbo, O., Rossignol, J.F., Pichard, E., Traore, H.A., Dembele, M., Diakite, M., Traore, F., Diallo, D.A., (1997) Nitazoxanide in the treatment of cryptosporidial diarrhoea and other intestinal parasitic infections associated with acquired immunodeficiency syndrome in tropical Africa. *Am J Trop Med Hyg* **56** 637-639.

Du Pont, H.L., Chappell, C.L., Sterling, C.R., Okhuysen, P.C., Rose, J.B., and Jakubowski, W. (1995) The infectivity of *Cryptosporidium parvum* in healthy volunteers. *N Eng J Med*. **332**, 855-9.

Fayer, R. and Ellis, W. (1993a) Glycoside antibiotics alone and combined with tetracycline for prophylaxis of experimental cryptosporidiosis in neonatal Balb/c mice. *J Parasitol* **79**, 553-58.

Fayer, R. and Ellis, W. (1993b) Paromomycin is effective as prophylaxis for cryptosporidiosis in dairy calves. *J Parasitol* **79**, 771-74.

Fayer, R. and Ungar, B.L. (1986) *Cryptosporidium* spp. and cryptosporidiosis. *Microbiol Rev* **50**, 458- 83.

Fichtenbaum, C.J., Ritchie, D.J. and Powderly, W.G. (1993) Use of paromomycin for treatment of cryptosporidiosis in patients with AIDS. *Clin Infect Dis* **16**, 298-300.

Flanigan, T., Whalen, C., Turner, J. *et al*. (1992) *Cryptosporidium* infection and CD4 counts. *Ann Intern Med* **116**, 840-2.

Flanigan, T., Ramratnam, B., Graeber, C. *et al*. (1996) Prospective trial of paromomycin for cryptosporidiosis in AIDS. *Am J Med* **100**, 370-2.

Gathe, J., Piot, D., Hawkins, K. *et al*. (1990) Treatment of gastrointestinal cryptosporidiosis with paromomycin. *VIth Int Conference on AIDS, San Francisco*. abst 2121.

Greenberg, P.D., and Cello, J.P. (1996) Treatment of severe diarrhoea caused by *Cryptosporidium parvum* with oral bovine immunoglobulin concentrate in patients with AIDS. *J Acquir Immun Defic Syndr Hum Retrovirol* **13** 348-54.

Hicks, P., Zwlener, R.J., Squires, J. and Savell, V. (1996) Azithromycin therapy for *Cryptosporidium parvum* infection in four children infected with human immunodeficiency virus. *J Pediatr* **129**, 297- 300.

Kelly, P., Zulu, I., Sianongo, S., and Farthing, M.J.G., (1998) Eradication of intracellular intestinal protozoa in AIDS patients using high dose albendazole. *Gastroenterology* **114** (4) A1007.

Marshall, R.J. and Flanigan, T.P. (1992) Paromomycin inhibits *Cryptosporidium* infection of a human enterocyte cell line. *J Infect Dis* **165**, 772-74.

Okhuysen, P.C., Chappell, C.L., Sterling, C.R., *et al*. (1998) Susceptibility and serologic response of healthy adults to reinfection with *Cryptosporidium parvum*. *Infect Immun* **66**, 441-43.

Regh , J.E. (1991) Activity of azithromycin against Cryptosporidia in immunosuppressed rats. *J Infect Dis* **163**, 1293-96.

Regh, J.E. (1994) A comparison of anti-cryptosporidial activity of paromomycin with that of other aminoglycosides and azithromycin in immunosuppressed rats. *J Infect Dis* **170**, 934-38.

Tzipori, S., Rand, W., Griffiths, J. *et al.* (1994) Evaluation of an animal model system for cryptosporidiosis: therapeutic efficacy of paromomycin and hyperimmune bovine colostrum- immunoglobulin. *Clin Diagn Lab Immunol* **1**, 450-63.

Tzipori, S., Roberton, D., Chapman, C. (1986) Remission of diarrhoea due to cryptosporidiosis in an immunodeficient child treated with hyperimmune bovine colostrum. *Br Med J* **293** 1276-1277.

Ungar, B.L., Kao, T.C., Burris, J.A. and Finkelman, F.D. (1991) *Cryptosporidium* infection in an adult mouse model. Independent roles for IFN-gamma and CD4+ T lymphocytes in protective immunity. *J Immunol* **147**, 1014-22.

Vargas, S.L., Shenep, J.L., Flynn, P.M. *et al*. (1993) Azithromycin for treatment of severe *Cryptosporidium* diarrhoea in two children with cancer. *J Pediatr* **123**, 154-56.

Verdon, R.J., Polianski, C., Gaudebout, C. *et al.* (1994) Evaluation of curative anti-cryptosporidial activity of paromomycin in a dexamethasone-treated rat model. *Antimicrob Agents Chemother* **38**, 1681-82.

Verdon, R., Polianski, C., Gaudebout, C. *et al*. (1995) Evaluation of high-dose regimen of paromomycin against cryptosporidiosis in the dexamethasone-treated rat model. *Antimicrob Agents Chemother* **39**, 2155-7.

White, A.C., Chappell, C.L., Hayat, C.S. *et al.* (1994) Paromomycin for cryptosporidiosis in AIDS: a prospective, double-blind trial. *J Infect Dis* **170**, 419-24.

Wiest, P.M., Johnson, J.H. and Flanigan, T.P. (1993) Microtubule inhibitors block *Cryptosporidium parvum* infection of a human entrocyte cell line. *Infect Immun* **61**, 4888-90.

Woods, K.M., Nesterenko, M.V. and Upton, S.J. (1996) Efficacy of 101 antimicrobials and other agents on the development of *Cryptosporidium parvum in-vitro*. *Ann Trop Med Parasitol* **90**, 603-15.

# 10 Advice on private water supplies

## 10.1 Introduction

10.1.1 Private water supplies are defined as any supplies of water provided otherwise than by a statutorily appointed water utility. There are about 50,000 private water supplies in England and Wales supplying about a third of a million people with water for domestic purposes and 20,000 in Scotland supplying 130,000 people. Approximately 40,000 of these supplies serve people in a single dwelling. However, many more people will consume private water supplies used for food production purposes such as brewing or when it is supplied to places such as hospitals, hotels, schools or campsites.

10.1.2 Although there are some private supplies in urban areas, particularly those used serving industrial purposes, most private supplies are situated in the rural parts of the country. The source of the supply may be a well, a borehole, a spring or a stream. The supply may serve just one property or several properties.

## 10.2 Regulation of private water supplies

10.2.1 Private water supplies are regulated by local authorities under the Private Water Supplies Regulations. These contain the same water quality standards as those for public drinking water supplies but the frequency of monitoring and the parameters tested will vary according to how many people use the supply or the volume of water used daily. The regulations do not require private water supplies to be monitored specifically for *Cryptosporidium*. They rely on the presence of faecal coliform indicator bacteria to warn of possible microbiological contamination.

10.2.2 The regulations require only infrequent monitoring of small private water supplies and there is no specified sampling frequency for those supplies serving only a single property for domestic purposes. Therefore owners and users of private water supplies need to be aware of the potential for water contamination and what can be done to reduce the risk.

## 10.3 Quality of private water supplies

10.3.1 All private water supplies can pose a threat to health unless they are properly protected and treated. Although many private water supplies provide a safe source of water there are risks of contamination from micro-organisms including *Cryptosporidium* associated with them that do not apply to public water supplies. These are:

- farm animals may have unrestricted access to the source catchment and in some cases the wellhead or spring collecting chamber;
- many sources have inadequate protection from contamination from surface runoff; and
- the absence of treatment to many supplies and the inadequacy of many of the treatment systems that are installed.

10.3.2 For the reasons given above private water supplies can be more likely than public water supplies to contain *Cryptosporidium.* This has been demonstrated in a small study in northern England (Clapham 1997) where *Cryptosporidium* oocysts were detected in nine out of 15 private water supplies known to contain total and faecal coliform bacteria regularly. There was no known illness associated with the users of these supplies, but all those who drink contaminated water are at risk of infection. However, the risk is likely to be greater for the very young, the infirm and the immunocompromised and for those who do not drink the water regularly, such as visitors and holiday makers. It may be advisable for them to use boiled or bottled water for drinking.

## 10.4 Advice on protecting the supply

10.4.1 One of the best methods of evaluating the potential quality of a private water supply is a sanitary survey. Important elements to be considered are:

- the type of supply that is groundwater, surface water or spring;
- source protection;
- access of animals to the catchment; and
- the condition of collection chambers, tanks and pipework.

10.4.2 It is much better to protect the source of the supply to prevent contamination rather than trying to treat the water afterwards. Protection can be provided by using fencing to keep grazing animals away, having suitable drainage channels to divert rainwater, covering and sealing the tops of boreholes and wells and making sure collecting chambers are in good condition and protected from animal access.

10.4.3 If owners and users of private water supplies know or suspect that their water supply is contaminated with micro-organisms they should install treatment, or if practicable, consider the possibility of connecting to the public supply. Their local authority can give advice on the right type of treatment and the local water utility on the availability of a public supply. As an interim measure all water to be used for drinking and food preparation should be boiled. It is only necessary to bring the water to the boil, prolonged boiling is unnecessary.

## 10.5 Further advice for local authorities and owners and users of private water supplies

10.5.1 The Drinking Water Inspectorate has provided advice to local authorities on *Cryptosporidium* in Private Water Supplies (DWI 1996) and DWI (DWI 1998), the Scottish Office (SO 1997) and Northern Ireland Environment and Heritage Service (NIEHS 1997) have produced advice leaflets for owners and users of private water supplies. The Expert Group endorses the advice in these publications that source protection is the greatest safeguard for *Cryptosporidium* in private water supplies. The text of the DWI advice leaflet is reproduced in Appendix A6.

## References


Clapham, D. (1997) The incidence of *Cryptosporidium* and *Giardia* in private water supplies, correlatory indicators and the value of the coliform standard in assessing water quality**.** *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick.* ed. Morris, R. and Gammie, A., 220-225.

DWI. (1996) Removal of Oocysts of *Cryptosporidium* from Private Water Supplies – Assessment of Point-of-Use Filters. Drinking Water Inspectorate 3932/1. London, UK.

DWI. (1998) Private Water Supplies. Drinking Water Inspectorate. London, UK.

NIEHS. (1997) Is Your Private Water Supply Safe? Drinking Water Inspectorate, Northern Ireland Environment and Heritage Service, Belfast, UK.

SO. (1997) Is Your Private Water Supply Safe? The Scottish Office. Edinburgh, UK.

# 11 Review of research published since publication of the Second Report of the Group of Experts

## 11.1 Introduction

11.1.1 There have been several hundred research publications worldwide since the Second Report of the Group of Experts appeared in 1995. The abstracts of some 150 of these reports are reviewed in Appendix A8 and key findings are summarised in this Chapter. By far the greatest proportion of the research output has been funded in North America and the UK. More recently, research publications from mainland Europe and Australia have started to appear, reflecting a greater awareness of the significance of *Cryptosporidium* in water supplies.

## 11.2 Detection of the organism

11.2.1 Significant advances have been made both in filtration and separation technologies and in the procedures for concentration of the organism prior to identification. The combination of improved cartridge or membrane filters and concentration on immuno-magnetic beads has brought about significant improvements in the level of recovery and precision in recovery. Flow cytometry and laser scanning techniques now offer the potential to automate the processes of sorting, detection and confirmation of the organism. Current research promises further advances using continuous centrifugation, compressed foam filtration media or vortex flow filtration.

## 11.3 Viability and infectivity studies

11.3.1 There has been controversy over the role of surrogate tests for infectivity as applied to environmental isolates. Nevertheless, there is a consensus that such tests are essential for the interpretation of disinfection studies. Recent research funded by DETR and the American Water Works Association Research Foundation (AWWARF) has compared dye inclusion/exclusion techniques, *in vitro* excystation and neo-natal mouse infection in controlled disinfection studies. The results indicate that all surrogates gave unreproducible results and provided a poor basis for assessing efficiency of disinfection.

11.3.2 An insight into possible reasons for these findings has been provided by DETR funded research on the effects that cleaning and concentration procedures have on the surface properties of oocysts. It appears that the relatively extreme chemical environment and physical conditions cause unpredictable reactions at the oocyst surfaces and also influence the results of PCR analysis.

11.3.3 These findings, along with evidence for distinctive genotypes with different infection potential towards animals and humans (see 11.3.4 and Appendix A7), calls into question the reliability of published disinfection studies. While some studies may provide a general indication of disinfection capability, reliance on the data for treatment design or for modelling interaction of disinfectants appears unwise. Developments

involving tissue culture may offer a more reliable surrogate for human infectivity. However, it is likely that such approaches will also be sensitive to oocyst cleaning and concentration procedures.

11.3.4 Appendix A7 considers the evidence for identifiably distinct 'strains' or sub-types (genotypes or lineages) of *C. parvum* and concludes that this is very strong. One such sub-type appears to be restricted to man. There is no evidence that this results from the parasite changing within the host during the infection. Indeed, in a further outbreak in the UK where there was evidence of animal (sheep) contamination of the water, isolates from cases were of the animal genotype. The evidence of a high prevalence of 'human genotype' in three waterborne outbreaks has considerable significance in relation to transmission by the water route, suggesting that sewage effluent may have been the major source of these outbreaks.

## 11.4 Water treatment

11.4.1 There is now wide acceptance that one key to minimising exposure to *Cryptosporidium* is the consistent production of low turbidity water and the avoidance of peaks in turbidity. Much research has concentrated on the optimisation of turbidity and particle counts and also on the identification of a suitable surrogate for *Cryptosporidium* removal during treatment. The most promising tool appears to be measurement of bacillus spores and this approach has been validated in a number of pilot studies in North America and the UK.

11.4.2 Interest in removal of protozoan cysts has prompted the development of a variety of innovative filtration technologies, some of which have now been approved under regulation 25 of the Water Supply (Water Quality) Regulations 1989. A number of disinfection technologies based on high intensity UV radiation have also been introduced as treatment for *Cryptosporidium*. The exposure characteristics and design of these systems are such that they are only applicable to small water supply systems presently.

## 11.5 Risk assessment

11.5.1 DETR funded studies of microbiological risk assessment have addressed specifically the risk of exposure to *Cryptosporidium* via drinking water. Modelling and pilot scale work has confirmed that water treatment processes impose extremely non-random characteristics on the distribution of oocysts, probably through absorption of oocysts onto particulate matter. These findings may shed some light on the initiation of outbreaks, with a small number of consumers becoming exposed to extremely localised high concentrations of the organism.

11.5.2 An American Waterworks Association Research Foundation study has demonstrated the possibility of using seroprevalence of antibodies as an indication of a history of exposure to *Cryptosporidium* via drinking water. A study of serum from consumers receiving surface water derived drinking water showed a higher prevalence of antibodies than in a matched population receiving water from a deep groundwater source.

## 11.6 *Cryptosporidium* in the environment

11.6.1 The Ministry of Agriculture, Fisheries and Food and Scottish Office Agriculture and Food Department (MAFF – SOAFD) study on pathogens in the farm environment has contributed greatly to the understanding of the significance of agricultural activity as a source of the organism in environmental waters. The study has confirmed that good agricultural practice in relation to disposal of animal manures and slurries should not pose a particular pollution hazard. However, a related Environment Agency-MAFF study on wild animals in the farm environment has indicated that rats, mice and other animals carry the organism and may play a significant role in its dispersion in the environment. (See Appendix A2).

### *11.7 Recommendations for research*

***11.7.1 The Group has identified the following as areas requiring further research:***

***(i) application of continuous monitoring for Cryptosporidium in treated waters and investigation of correlation between Cryptosporidium and operating conditions that might lead to breakthrough of the organism;***

***(ii) investigations into the ways laboratory analytical procedures might affect the biological properties of oocysts;***

***(iii) development of a standardised approach to conducting disinfection trials;***

***(iv) development of reliable, routine tests for oocyst viability;***

***(v) further studies of the application of seroprevalence studies in assessing the impact of water treatment in reducing community exposure to Cryptosporidium;***

***(vi) investigation of the impact of operating filters under declining rate on the removal of Cryptosporidium;***

***(vii) evaluation of quality changes in treated waters and development of procedures to allow operators to identify Cryptosporidium risk associated with these changes for specific treatment works;***

***(viii) development of techniques to specify and assess the performance of filtration systems for oocyst removal from groundwaters; and***

***(ix) further evaluation and development of the use of bacterial spores to assess treatment performance.***

***11.7.2 The following recommendations for research relate to groundwater (see Chapter 4):***

***(i) development of operational monitoring tools to improve the detection of rapid influence of surface water sources on the quality of groundwater;***

***(ii) transport and fate of Cryptosporidium and other pathogens in groundwater systems;***

***(iii) application of chemical and particulate tracers to investigate the transport and attenuation of pathogens in groundwater;***

***(iv) mechanisms causing, and the significance of, turbidity in groundwater to establish the role of rapid influence by surface water and assessing the use of turbidity as a monitoring tool; and***

***(v) attenuation rates for Cryptosporidium in soils and unsaturated zones following application of farm wastes and sewage sludge to land (4.7.2).***

# 11 Review of research published since publication of the Second Report of the Group of Experts

## 11.1 Introduction

11.1.1 There have been several hundred research publications worldwide since the Second Report of the Group of Experts appeared in 1995. The abstracts of some 150 of these reports are reviewed in Appendix A8 and key findings are summarised in this Chapter. By far the greatest proportion of the research output has been funded in North America and the UK. More recently, research publications from mainland Europe and Australia have started to appear, reflecting a greater awareness of the significance of *Cryptosporidium* in water supplies.

## 11.2 Detection of the organism

11.2.1 Significant advances have been made both in filtration and separation technologies and in the procedures for concentration of the organism prior to identification. The combination of improved cartridge or membrane filters and concentration on immuno-magnetic beads has brought about significant improvements in the level of recovery and precision in recovery. Flow cytometry and laser scanning techniques now offer the potential to automate the processes of sorting, detection and confirmation of the organism. Current research promises further advances using continuous centrifugation, compressed foam filtration media or vortex flow filtration.

## 11.3 Viability and infectivity studies

11.3.1 There has been controversy over the role of surrogate tests for infectivity as applied to environmental isolates. Nevertheless, there is a consensus that such tests are essential for the interpretation of disinfection studies. Recent research funded by DETR and the American Water Works Association Research Foundation (AWWARF) has compared dye inclusion/exclusion techniques, *in vitro* excystation and neo-natal mouse infection in controlled disinfection studies. The results indicate that all surrogates gave unreproducible results and provided a poor basis for assessing efficiency of disinfection.

11.3.2 An insight into possible reasons for these findings has been provided by DETR funded research on the effects that cleaning and concentration procedures have on the surface properties of oocysts. It appears that the relatively extreme chemical environment and physical conditions cause unpredictable reactions at the oocyst surfaces and also influence the results of PCR analysis.

11.3.3 These findings, along with evidence for distinctive genotypes with different infection potential towards animals and humans (see 11.3.4 and Appendix A7), calls into question the reliability of published disinfection studies. While some studies may provide a general indication of disinfection capability, reliance on the data for treatment design or for modelling interaction of disinfectants appears unwise. Developments

involving tissue culture may offer a more reliable surrogate for human infectivity. However, it is likely that such approaches will also be sensitive to oocyst cleaning and concentration procedures.

11.3.4 Appendix A7 considers the evidence for identifiably distinct 'strains' or sub-types (genotypes or lineages) of *C. parvum* and concludes that this is very strong. One such sub-type appears to be restricted to man. There is no evidence that this results from the parasite changing within the host during the infection. Indeed, in a further outbreak in the UK where there was evidence of animal (sheep) contamination of the water, isolates from cases were of the animal genotype. The evidence of a high prevalence of 'human genotype' in three waterborne outbreaks has considerable significance in relation to transmission by the water route, suggesting that sewage effluent may have been the major source of these outbreaks.

## 11.4 Water treatment

11.4.1 There is now wide acceptance that one key to minimising exposure to *Cryptosporidium* is the consistent production of low turbidity water and the avoidance of peaks in turbidity. Much research has concentrated on the optimisation of turbidity and particle counts and also on the identification of a suitable surrogate for *Cryptosporidium* removal during treatment. The most promising tool appears to be measurement of bacillus spores and this approach has been validated in a number of pilot studies in North America and the UK.

11.4.2 Interest in removal of protozoan cysts has prompted the development of a variety of innovative filtration technologies, some of which have now been approved under regulation 25 of the Water Supply (Water Quality) Regulations 1989. A number of disinfection technologies based on high intensity UV radiation have also been introduced as treatment for *Cryptosporidium*. The exposure characteristics and design of these systems are such that they are only applicable to small water supply systems presently.

## 11.5 Risk assessment

11.5.1 DETR funded studies of microbiological risk assessment have addressed specifically the risk of exposure to *Cryptosporidium* via drinking water. Modelling and pilot scale work has confirmed that water treatment processes impose extremely non-random characteristics on the distribution of oocysts, probably through absorption of oocysts onto particulate matter. These findings may shed some light on the initiation of outbreaks, with a small number of consumers becoming exposed to extremely localised high concentrations of the organism.

11.5.2 An American Waterworks Association Research Foundation study has demonstrated the possibility of using seroprevalence of antibodies as an indication of a history of exposure to *Cryptosporidium* via drinking water. A study of serum from consumers receiving surface water derived drinking water showed a higher prevalence of antibodies than in a matched population receiving water from a deep groundwater source.

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***(iii) development of a standardised approach to conducting disinfection trials;***

***(iv) development of reliable, routine tests for oocyst viability;***

***(v) further studies of the application of seroprevalence studies in assessing the impact of water treatment in reducing community exposure to Cryptosporidium;***

***(vi) investigation of the impact of operating filters under declining rate on the removal of Cryptosporidium;***

***(vii) evaluation of quality changes in treated waters and development of procedures to allow operators to identify Cryptosporidium risk associated with these changes for specific treatment works;***

***(viii) development of techniques to specify and assess the performance of filtration systems for oocyst removal from groundwaters; and***

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***(iii) application of chemical and particulate tracers to investigate the transport and attenuation of pathogens in groundwater;***

***(iv) mechanisms causing, and the significance of, turbidity in groundwater to establish the role of rapid influence by surface water and assessing the use of turbidity as a monitoring tool; and***

***(v) attenuation rates for Cryptosporidium in soils and unsaturated zones following application of farm wastes and sewage sludge to land (4.7.2).***

# Appendix A1

# Review of recommendations in the Second Report of the Group of Experts

## A1.1 Introduction

A1.1.1 The Second Report of the Group of Experts made recommendations on:

(i) recovery, identification and typing of the organism;
(ii) how it should be controlled in the environment;
(iii) controlling the spread of infection in man;
(iv) water treatment and distribution;
(v) monitoring for oocysts in water; and
(vi) investigation and management of outbreaks.

A1.1.2 All the above are important and it is necessary to have a better understanding of the organism and how it can be controlled in the environment but ultimately public health protection rests largely on effective monitoring, effective water treatment and, in the event that something goes wrong, good outbreak management by all those involved locally.

A1.1.3 This appendix sets out those recommendations from the second report, many of which originated in the First Report, which the current Expert Group considers to be of continuing relevance and worth emphasising. In addition the Group has added and amplified some of the recommendations where necessary. The recommendation numbers refer to those used in the Second Report to which reference should be made.

## A1.2 The organism

**Recommendation 1** Further research work should be encouraged on methods of recovery and identification of *Cryptosporidium* oocysts such as cross-flow filtration, magnetisable particles, flocculation, electro-rotation assays and gene probes.

**Recommendation 2** Further research should be carried out to develop methods for identifying different species and strains of *Cryptosporidium.*

**Recommendation 3** The cross-checking of results with specialist laboratories, particularly in relation to research studies and where an outbreak has occurred, is seen as important. Laboratories are strongly advised to participate in a recognised external quality assurance scheme.

**Recommendation 5** Further work on typing and host specificity of *Cryptosporidium* oocysts should be encouraged.

### A1.3 Control of *Cryptosporidium* in the environment

**Recommendation 6** Water utilities should be encouraged to make results of monitoring available on a regular basis to a national database to provide further information on the occurrence of oocysts in water.

***Further recommendation of the Expert Group*** ***A1.3.1 A national database should be established to provide comprehensive information on the occurrence of oocysts in both source and treated water.***

**Recommendation 7** Surveys should be carried out on the concentration of oocysts in sewage effluents.

***Further recommendation of the Expert Group*** ***A1.3.2 Research work on oocysts in sewage effluents should be directed at that work associated with typing and host specificity.***

**Recommendation 8** The Codes of agricultural practice to prevent pollution of water sources should be reviewed regularly and the advice on storage and disposal of animal farm waste should be revised in the light of the results of research. Efforts should be made to encourage all farmers to follow these Codes.

***Further recommendation of the Expert Group*** ***A1.3.3 The advice on storage and disposal of animal waste should be reaffirmed and efforts increased to encourage farmers to follow Codes of good practice.***

**Recommendation 9** Regulations, codes of practice and enforcement procedures for the disposal of sludges which may contain *Cryptosporidium* should be reviewed and if appropriate harmonised.

***Further recommendation of the Expert Group*** ***A1.3.4 The inactivation of Cryptosporidium oocysts should be made one specific consideration in policy and practice in the disposal of sludges to land.***

### A1.4 Cryptosporidiosis in man

**Recommendation 10** Laboratories in England and Wales should be required to report the detection of *Cryptosporidium* oocysts in clinical samples to local public health officials and to the Public Health Laboratory Service (PHLS) Communicable Disease Surveillance Centre. Pending the introduction of a statutory requirement to report, the contract specification with laboratories should include the necessary details.

***Further recommendation of the Expert Group*** ***A1.4.1 This is covered in paragraph 3.3.10.***

**Recommendation 12** Recommendation R90/11 of the 1990 Report on advice to persons having contact with livestock is reiterated.

**Recommendation 13** The advice being produced on visits to farms and contact between farm animals and children should be brought to the attention of Environmental Health Officers and teachers.

**Recommendation 14** The effort made to date to communicate the advice on the control of person to person spread of infection is recognised, but this is seen to be a continuing requirement.

**Recommendation 15** Advice on personal hygiene should be brought to the attention of persons handling food, including those preparing ice and bottled waters.

***Further recommendation of the Expert Group*** ***A1.4.2 Advice on personal hygiene in handling food, in preparation of ice and bottled waters should be reviewed and promoted by the new Food Standards Agency.***

**Recommendation 16** The possibility of infection arising from pollution incidents in swimming pools should be brought to the attention of pool operators, engineers and designers. Checks should also be made that filtration systems are working effectively.

**Recommendation 17** Continued effort should be given to advising the public on the risks associated with accidental or deliberate ingestion of water in its raw state.

**Recommendation 18** The absence of *Cryptosporidium* oocysts in drinking water can never be guaranteed. In the light of a small risk of infection, it would be appropriate to advise people in whom cryptosporidiosis is likely to be a persistent and life-threatening illness as a result of impaired immunity.

**Recommendation 19** Where infectivity trials are carried out using human volunteers it is essential that a strain of *Cryptosporidium* is used which is known to be pathogenic to humans.

***Further recommendation of the Expert Group*** ***A1.4.3 This recommendation should now be linked to recommendation 5 above in relation to host specificity.***

**Recommendation 21** The Department of Health should keep work in progress under the Group review and encourage further controlled trials of treatment where appropriate. See also paragraph 9.6.2.

### A1.5 Water treatment and distribution

**Recommendation 22** Water utilities should ensure that the design and operation of treatment plant is optimised in a cost effective way for particle removal taking into account the level of risk identified at each plant.

**Recommendation 23** Advice given in the 1990 Report (recommendation R90/19 (ii)) on minimising rapid changes of flow is reiterated. However, ways of reducing the passage of particles into treated water following backwashing and after filter shut-down require further research.

**Recommendation 25** Strategies should be developed for each treatment plant whereby the optimum use can be made of turbidity and/or particle monitors to minimise passage of particles into supply at all stages in the filtration cycle.

***Further recommendation of the Expert Group*** ***A1.5.1 The Group has made further recommendations on water treatment in Chapter 5, Advice to Water Utilities.***

**Recommendation 27** Continuing attention is required to maintaining borehole linings and seals.

**Recommendation 28** A realistic assessment should be made, using published results, of the likely impact of disinfection strategies on reducing the risk of cryptosporidiosis in humans.

***Further recommendation of the Expert Group*** ***A1.5.2 It is considered that although disinfection has some effect, its contribution at time of most need (that is barrier breakthrough) has not been proven so in public health protection terms it cannot be relied upon.***

**Recommendation 29** Water utilities and manufacturers should be encouraged to publish the results of trials on the removal of *Cryptosporidium* oocysts by membranes and textile filters.

**Recommendation 30** Water utilities should confirm regularly, with the appropriate authorities, contingency arrangements for the disposal of contaminated sludge and process waste waters.

**Recommendation 31** Attention needs to be given to the effective design of systems for separating supernatant water from sludge and backwash solids.

**Recommendation 32** Water utilities and manufacturers should be encouraged to publish the results of using novel separation methods and disinfectants in recycling systems.

**Recommendation 33** Water utilities should continue to maintain good hygienic procedures for the repair and maintenance of distribution systems taking account of national guidelines.

**Recommendation 35** Manufacturers should be required to provide, within their instructions for filter units, guidance on the safe handling and disposal of used filter elements.

**Recommendation 36** Monitoring of raw water for *Cryptosporidium* should be related to an assessment of catchment risks and the nature of the treatment provided at individual sites.

**Recommendation 37** Monitoring of treated water for *Cryptosporidium* on a regular basis for health protection is not recommended but should be carried out where a significant disturbance to raw water quality or to a water treatment plant has occurred. Additional investigations will be required if an outbreak of cryptosporidiosis occurs.

***Further recommendation of the Expert Group*** ***A1.5.3 The Group has made further recommendations on monitoring in Chapter 5, Advice to water utilities.***

**Recommendation 38** Water utilities should continue to ensure that their staff or those of contract laboratories are up-to-date in their knowledge of sampling and examination procedures including quality assurance measures. There should be regular monitoring and testing for *Cryptosporidium* to maintain the expertise of staff.

**Recommendation 39** Consideration should be given to confirming the adequacy of the laboratory arrangements by simulation of emergency situations.

***Further recommendation of the Expert Group*** ***A1.5.4 In light of some mistaken laboratory identifications of Cryptosporidium, consideration should be given to further training of laboratory staff and electronic links with expert laboratories.***

### A1.6 Investigation and management of an outbreak

**Recommendation 40** Epidemiological studies to investigate significant local increases in the background incidence of cryptosporidiosis should be encouraged, even if the increase does not apparently constitute an outbreak. Comparable methodologies should be used in these investigations.

| | |
|---|---|
| ***Further recommendation of the Expert Group*** | ***A1.6.1 The Group has made further recommendations on epidemiological studies in Chapter 7, Guidance on the epidemiological investigation of outbreaks of infection associated with mains water. See also Appendix A4*** |
| **Recommendation 41** | Health authorities, local authorities and water utilities should continue to update and rehearse existing emergency plans, which should cover chemical or microbiological incidents as well as outbreaks of waterborne disease. |
| **Recommendation 42** | The circumstances where water supply zones overlap health and local authority areas, or where more than one water utility supplies a single authority, should be addressed by discussion between parties to agree protocols for dealing with emergencies. |
| **Recommendation 43** | Arrangements should be put in place to ensure regular liaison between the appropriate staff of water utilities, health authorities and local authorities. This should not be confined to periods where there are problems. |
| **Recommendation 48** | Key members from the Incident Management Team should agree procedures for the issuing and withdrawal of boil water advice. When considering imposing such advice there should be clear recognition of the need to agree criteria for its withdrawal. |
| **Recommendation 49** | Attention should continue to be given to effective communication with the public and media during incidents or outbreaks. This should be included as part of the Incident Management Team and Outbreak Control Team plans and should be rehearsed regularly. |
| **Recommendation 52** | The importance of adequately prepared and regularly rehearsed water utility plans as set out in the 1990 Report (R90/49) is confirmed. |
| **Recommendation 53** | Water utilities should collaborate with local authorities to identify more clearly which high-risk premises are fed by particular supply zones thus enabling appropriate emergency plans to be drawn up in advance. |

# Appendix A2

# Agriculture as a source of *Cryptosporidium* and measures to reduce risks[7]

## A2.1 Introduction

A2.1.1 As part of the work arising from the deliberations of the Expert Group set up in 1990 under the chairmanship of Sir John Badenoch, a research programme was initiated jointly by the Ministry of Agriculture, Fisheries and Food (MAFF) and the Scottish Office Agriculture and Food Department (SOAFD). The programme was titled 'Protozoan, Bacterial and Viral Pathogens, Farm Animal Wastes and Water Quality Protection, (MAFF Open Contract CSA 2064) (Kemp *et al* 1995; Svoboda *et al* 1997) and the work was undertaken by jointly by the Moredun Research Institute, Institute of Grassland and Environmental Research, Scottish Agricultural College and Scottish Parasite Diagnostic Laboratory. The final report from this contract was delivered in 1995 and includes:

- a literature survey titled 'Pathogenic Microorganisms in Livestock Waste and Factors Influencing Their Transport to the Aquatic Environment'; and

- reports of work to meet the following objectives:

  (i) determine the quantity and the rates at which *Cryptosporidium, Giardia, Salmonella* and Rotaviruses are released into the environment during the animal production cycle;

  (ii) determine the degree of partitioning between solids and liquid phases of animal wastes;

  (iii) identify the predisposing factors affecting the dissemination of *Cryptosporidium* oocysts through the environment and assess the risk of water contamination associated with various storage, handling and disposal practices;

  (iv) determine, with particular reference to protozoal oocysts and cysts, the survival of pathogens in animal wastes during storage and when released into the terrestrial and aquatic environment;

  (v) assess the efficacy of mesophyllic and thermophilic aerobic treatment for the removal of these pathogens; and

  (vi) produce guidelines to minimise the dissemination of viable *Cryptosporidium* oocysts into the environment.

[7]A note prepared for the Expert Group by Rob Robinson, Environment Agency, and Ian Davidson, Ministry of Agriculture, Fisheries and Food.

A2.1.2 Recommendation 8 of the Second Report of the Group of Experts (Badenoch 1995) states that 'The Codes of agricultural practice to prevent pollution of water should be reviewed regularly and the advice on storage and disposal of animal waste should be revised in the light of the results of research. Efforts should be made to encourage farmers to follow these Codes'. This recommendation is further endorsed by the Expert Group (see paragraphs 4.3.7 and 4.8.4).

A2.1.3 This chapter is primarily concerned with the results of research and development and their dissemination through the revised Codes of Good Agricultural Practice for the Protection of Water (COPGAP – Water) (MAFF 1998), which applies in England and Wales and 'Prevention of Environmental Pollution from Agricultural Activity – A Code of Good Practice' (PEPFAA) (SO 1997), which applies in Scotland.

A2.1.4 An additional research contract – titled '*Cryptosporidium* in Farmed and Wild Animals and the Implications for Water Contamination' (CSA 2783) (Sturdee *et al* 1998), has been jointly funded by MAFF and the Environment Agency (EA) and undertaken by Coventry University. The objectives of this research were to:

- produce a quantitative and comprehensive account of the parasite's occurrence in livestock and wild animals by analysis of faecal samples;
- explore routes and reservoirs of infection amongst the animal groups tested; and
- identify the potential pathways for the contamination of watercourses.

## A2.2 Key points from the MAFF research and development

A2.2.1 Key points from the summary of the report for CSA 2064 (Kemp *et al* 1995) are reproduced in the following paragraphs and comments of particular relevance to the consideration of Codes of Good Agricultural Practice are highlighted.

A2.2.2 On farms with a recognised infection problem, up to 96% of the calves present have become clinically infected, resulting in large numbers of oocysts being shed – in excess of 1010 oocysts per animal is possible. Low grade infections occur in adult cattle, and while numbers of oocysts shed per gram of faeces are very low, making detection difficult, the combination of volume of faeces produced and the persistent nature of these infections can result in large numbers of oocysts being shed over a period of months.

A2.2.3 When infections occur in housed calves, some oocysts may be leached out of the bedding and washed into the drainage system, but the majority remain in the bedding, which may be treated subsequently. Oocysts produced by housed adult cattle will usually be channelled into the slurry storage system.

A2.2.4 Oocysts have been detected throughout the farm in all types of animal derived waste, including dirty water (which is usually contaminated with fresh faeces). **Correct handling of farm wastes, such as composting of manures and prolonged storage of slurries reduces oocyst viability markedly, while aerobic treatment of slurries can rapidly reduce oocyst viability to insignificant levels.**

A2.2.5 Oocysts are lost from wastes after application to land by both leaching and in runoff, though the numbers lost by both of these routes represents only a small percentage of the total numbers produced and of these only a small percentage may still be viable if the wastes have been handled correctly. **Fresh faeces from grazing animals, yard and dairy washings and bedding leachates seem the most likely agricultural sources of viable oocysts.** While the latter may be controlled, the former is impossible to regulate.

A2.2.6 As long as there is no effective antiprotozoal agent to treat cryptosporidial infection, its eradication within the farm environment is impossible, given its persistence and low infective dose. **Similarly, while adherence to the Codes of Good Practice for handling and disposal of farm wastes should minimise contamination of watercourses with viable oocysts, there are no practicable measures which will eradicate contamination completely.**

A2.2.7 Project CSA2783 was undertaken within an estate where farming operations were considered to be a model of good management. The major outputs were:

- confirmation that *Cryptosporidium* is ubiquitous amongst mammals (including wild species); and

- the establishment of a benchmark for what may be the irreducible minimum background levels of the organism which can be expected in the UK countryside.

### A2.3 Guidelines to minimise the dissemination of viable *Cryptosporidium* oocysts into the environment

A2.3.1 Objective 6 of project CSA2064 was the production of guidelines to minimise the dissemination of viable *Cryptosporidium* oocysts into the environment. The guidelines were intended to inform revisions of the Codes such as those undertaken recently by SOAFD and MAFF.

A2.3.2 The draft guidelines reflect the principal that minimisation of dissemination of *Cryptosporidium* oocysts should involve all aspects of good husbandry and should aim to reduce the incidence of outbreaks of cryptosporidiosis in farm animals as well as focusing on waste handling procedures.

A2.3.3 The revised COGAP – Water and PEPFAA take account of the researcher's draft guidelines wherever these are practical. The Codes emphasise good farm waste management planning and extra prudence in the storage and spreading of manures when *Cryptosporidium* is diagnosed on the farm (COGAP – Water, paragraph 9 and PEPFAA, section 4.3). The draft guidelines on animal purchase and rearing are generally in keeping with current good practice for the prevention and control of zoonotic diseases in general. The Government is currently planning the dissemination of such advice in the context of *Salmonella* in livestock.

A2.3.4 MAFF has undertaken a three year promotional campaign 'Practice the Codes' which has involved an educational pack sent to every agricultural college, over 100 articles in national and local press and around 30 presentations per year at relevant farmers' meetings by ADAS

advisors. The revised COGAP – Water is to be launched in October 1998 with a leaflet summarising key messages. The aim of the leaflet is to reach a wider audience than the Codes themselves and encourage more farmers to read and use the Codes. A publicity strategy is also planned after the relaunch.

### A2.4 Additional guidance

A2.4.1 An initiative involving water utilities, MAFF, the Agricultural Development and Advisory Service (ADAS) and the former Water Services Association produced a template leaflet entitled 'Wise ways with waste' (WSA 1995). This is used by water utilities to promote good farm waste management practice amongst farmers in catchments supporting public water supply sources. There is scope for the revision and reissue of a similar leaflet.

### A.2.5 Contact with farm animals

A2.5.1 Recommendation 12 of the Second Report of the Group of Experts (Badenoch 1995) covered the provision of advice to persons having contact with livestock of the importance of personal hygiene to protect themselves and their families from cryptosporidial infection. Recommendation 13 related to advice relating to visits to farms by school children.

A2.5.2 Relevant advice covering these issues has been produced by the Health and Safety Executive (HSE) in its Information Sheets on 'Common zoonoses in agriculture' and 'Occupational health risks from cattle' (HSE 1996; 1996).

## References


Badenoch, J. (1995) *Cryptosporidium* in water supplies. Second Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 108pp.

HSE. (1996) Information Sheet No2. Common zoonoses in agriculture. Health and Safety Executive. London, UK.

HSE. (1996) Information Sheet No 19. Occupational health risks from cattle. Health and Safety Executive. London, UK.

Kemp, J.S., Wright, S.E., Coop, R.L., Mawdsley, J.L., Barget, R.L., Merry, R.J., Pain, B.F., Theodorou, M.K., Read, I.A., Svobota, I. F., Bukhari, Z., and Smith, H.V. (1995) Protozoan, bacterial and viral pathogens, farm animal wastes and water quality. Ministry of Agriculture Fisheries and Food Project CSA2064 Final Report. London, UK.

MAFF. (1998) Code of Good Agricultural Practice for the Protection of Water (The Water Code). Ministry of Agriculture, Fisheries and Food. London, UK.

SO. (1997) Prevention of Environmental Pollution from Agricultural Activity. The Scottish Office Agriculture, Environment and Fisheries Department. Edinburgh, UK.

Sturdee, A.P., Bodley-Tickell, A.T. and Kitchen, S.E. (1998) Cryptosporidium in Farmed and Wild Animals and the Implications for Water Contamination. Ministry of Agriculture, Fisheries and Food Project CSA2783 Final Report. London, UK.

Svoboda, I.F., Read, I., Kemp, J.S., Wright, S.E., Coop, R.L., Mawdsley, J.L., Merry, R.J., Bargett, J.L., Pain, B.F., Theodoru, M.K., Bukhari, Z. and Smith, H.V. (1997) In Cryptosporidium in Water – the Challenge to Policy Makers and Water Managers, Proceedings of Symposium by Chartered Institute of Water and Environmental Management, Glasgow, UK. December 1997, 3-20.

WSA. (1995) Wise Ways with Waste. Water Services Association. London, UK.

# Appendix A3

# Advice for Outbreak Control Teams

## A3.1 Draft meeting agenda for Outbreak Control Teams

### A3.1.1 Minutes

The Chairman should ensure that minutes of each meeting are taken by a person not directly involved and that these are circulated with action points to all members as soon as possible after the meeting.

### A3.1.2 Agenda

1 Chairman's introduction including terms of reference

2 Minutes of last meeting (if applicable)

3 Review membership (are there lines of contact to all appropriate organisations ?)

4 Outbreak resumeè and update

- 4.1 General situation report
- 4.2 Case report
- 4.3 Water utility report
- 4.4 Microbiological report
- 4.5 Environmental health report
- 4.6 Other relevant reports

5 Management of outbreak

- 5.1 Control measures
- 5.2 Implications for public health
- 5.3 Advice to boil water
- 5.4 Provision of alternative water supplies
- 5.5 Care of patients (hospital and community)
- 5.6 Microbiological aspects (specimens, analysis and resources)

6 Issuing advice

- 6.1 Advice to the public (need for press release)
- 6.2 Advice to professionals (GPs, hospital doctors and nurses)

7 Agree content of press release and press arrangements

8 Nominate others to assist Chairman in press conferences and interviews (if required)

9 Consider arrangements for enquiries from the public (the need for a Helpline)

10 Obtain all relevant telephone and fax numbers and other contact details for all key personnel

11 Agree action points and timetable, identify individuals responsible for the agreed actions

12 Date, time and location of next meeting

### A3.2 Checklist for outbreak control teams

A3.2.1 The following is a suggested prompt list for actions by the Chairman of an Outbreak Control Team (OCT) for dealing with microbiological incidents affecting drinking water supplies. It is not meant to imply that each action must follow the one preceding it or that all steps are needed on every occasion. In practice some steps will be carried out simultaneously and others will be required throughout the outbreak.

**1 Ongoing routine surveillance**

1.1 Analysis of routine data
1.2 Is there a mechanism for early identification of change in data?

**2 Recognition of a possible outbreak/potential for an outbreak**

2.1 Notification of an incident by a water utility
2.2 Investigation of the potential for the incident to cause an outbreak
2.3 Change in background level of cases
2.4 Determine if there is an outbreak
2.5 Consider whether or not cases have the same illness

**3 Assessing the public health significance of the incident**

3.1 Obtain as much information as possible about:
- 3.1.1 the nature of the incident;
- 3.1.2 the exact area affected in terms of water supply zone(s) and geographical distribution;
- 3.1.3 the action being taken by the water utility;
- 3.1.4 whether more than one water utility, local authority or health authority is involved.

3.2 Decide from the available evidence whether the incident may result in an outbreak

**4 If no potential for an outbreak** – no further investigation, resume ongoing surveillance.

**5 If potential for an outbreak, or outbreak recognised**

5.1 When convening the outbreak control team, consider:
- 5.1.1 membership
- 5.1.2 optimum size
- 5.1.3 lines of communication to all organisations not represented directly
- 5.1.4 accommodation, communications and catering
- 5.1.5 deputies to provide cover for long hours
- 5.1.6 accurate records of meetings

5.2 Start a log book noting the time and date of the first notification of the incident and maintain this record throughout the incident

5.3 Assess the seriousness of the incident from details provided by the water utility
5.4 Seek advice from professionals eg national experts, CDSC, the Director of the local public health laboratory
5.5 Make contact with the Drinking Water Inspectorate and the Department of Health

**6 Decide on control measures and determine the necessary commitment of personnel and resources**

6.1 Initiate immediate control measures
6.2 Consider the need for the issue of 'Advice to boil water' and the special provisions needed if this action is carried out
6.3 Liaise with and advise the water utility who ultimately issue the notice of advice, appropriately
6.4 Consider with the water utility the provision of alternative water supplies
6.5 Ensure high risk premises eg businesses concerned with food production, hospitals, schools and residential homes have been contacted
6.6 Ensure water sampling regimen covers appropriate area and samples are analysed for appropriate parameters
6.7 Consider if there is an ongoing public health risk

**7 Communications with the media and the public**

7.1 Consider best routes of communication for informing the media and the public
7.2 Ensure accuracy and timeliness
7.3 Use the media constructively
7.4 Nominate one or more spokespersons

**8 Decide when the outbreak is over**

8.1 Decide at an early stage the criteria for lifting 'Advice to boil water'

**9 Prepare and issue the OCT report as soon after the event as practicable**.

9.1 Include a review of lessons learnt for the future

### A3.3 Draft outline for Outbreak Control Team report

The following is a list of suggested headings which is not exhaustive. Each report should be tailored to the circumstances of the individual incident.

Terms and abbreviations

Summary

1 Introduction

2 Background to the outbreak

2.1 Population demographics
2.2 Background rates of cryptosporidiosis

2.3 Water treatment and distribution
2.4 Water quality monitoring

3 Outbreak control

3.1 Co-ordination and management of outbreak
3.2 Action taken by the water utility
3.3 Advice to boil water
3.4 Media
3.5 Advice to the public and to business

4 Epidemiology

4.1 Surveillance
4.2 Descriptive epidemiology
4.3 Case Control Study

5 Other investigations

5.1 Water microbiology
5.2 Catchment area studies
5.3 Climatic conditions
5.4 Hydrogeology

6 Discussion

6.1 Epidemiology
6.2 Water treatment and distribution
6.3 Control measures
6.4 Other recent outbreaks

7 Actions by the water utility

8 Lessons learned, recommendations and conclusions

9 References

10 Appendices

10.1 Chronology of events
10.2 General background on cryptosporidiosis
10.3 *Cryptosporidium* and water supplies
10.4 The Outbreak Control Team – membership and terms of reference
10.5 Detailed epidemiology
10.6 Maps

**A3.4 Questions and answers on cryptosporidiosis – briefing for those dealing with enquiries from the public**

**Q** What is cryptosporidiosis?

**A** Cryptosporidiosis is a diarrhoeal disease of humans and animals, caused by a microscopic parasite called *Cryptosporidium*. The organism is common in farm and domestic animals, and is passed on through their manure. It is present in the environment at low levels all the time, but higher levels are common in springtime, particularly when heavy rain washes the parasite into the ground and into rivers and lakes.

**Q** How is cryptosporidiosis spread?

**A** It can spread to humans by a variety of routes. Person-to-person spread is an important source of infection if care is not taken with personal hygiene. Contact with farm animals (and sick pets) can put people at risk of infection. Water supplies may be vulnerable to contamination with *Cryptosporidium*, and the infection can be spread via drinking water.

**Q** What precautions can be taken against cryptosporidiosis?

**A** Personal hygiene is very important, particularly if there is someone with diarrhoea in the household or if farm animals or sick pets have been handled. Hand washing is important after using the toilet, or as soon as possible after touching potential sources of infection (ie farmyard animals or sick pets, manure on the ground, or people with diarrhoea). Good hygiene is particularly important before handling food.

**Q** Why have I been told to boil drinking water? [if appropriate]

**A** *Cryptosporidium* is killed by boiling. A boil water notice has been issued because [as appropriate] *Cryptosporidium* has been found in testing of the water supply or the pattern of illness in the community suggests that water may be a factor in transmission of the disease at present, and the boil water notice has been issued as a precaution.

**Q** How long do I have to boil water for?

**A** It is sufficient to bring the water to the boil and then allow it to cool. Using an electric kettle is fine.

**Q** What about brushing teeth, ice cubes, bathing and washing up?

**A** Brushing teeth – use cooled boiled water for brushing teeth.

Ice cubes – if you made ice cubes prior to the boil water notice, discard them. Make ice cubes with cooled boiled water or fizzy drinks.

Washing and bathing – it is quite safe to wash and take a bath: the route of infection is by swallowing the water.

Washing up – dishes should be washed using boiled water if possible, but it is probably sufficient to rinse washed dishes with boiled and cooled water before they are dried. Dishwashers are unaffected if used on a hot wash cycle.

**Q** Are pets affected?

**A** As household pets can become infected with *Cryptosporidium*, it is advisable to use water for them to drink that has been boiled and cooled.

**Q** Can I use water filters or water purification tablets instead of boiling?

**A** Domestic filters should not be relied upon unless they are designed for the purpose of removing micro-organisms (ie not just designed to improve palatability). Please refer to the manufacturer's brochure if in any doubt. Domestic filter cartridges may trap contamination, and particular care is required in hygienic disposal of these. Water purification tablets are not recommended as an alternative to boiling.

**Q** What are the public health authorities doing about this?

**A** The public health authorities, the environmental health officers and the water suppliers are working together to ensure that any possible risks to the public are kept to a minimum, and that the need to boil water is removed as quickly as possible.

**Q** What should I do if I become ill?

**A** This infection will usually clear up by itself in a healthy person, although this may take several days. You should consult your GP if diarrhoea is unduly prolonged or if you are at all concerned. A GP should also be consulted if the person affected is very young, elderly or frail. It is generally recommended to increase your fluid intake if you have diarrhoea.

**Q** Is anyone particularly at risk from this infection?

**A** People who have problems with their immune systems are more at risk of serious illness or very prolonged illness. These people include those with HIV infection or AIDS, those on chemotherapy for illnesses such as cancer or leukaemia, transplant patients, and people with hereditary immune disorders. These people may be advised to boil drinking water before consumption at all times, even if it is bottled. Please contact your doctor if you think you might be affected.

**Q** What treatment is available for cryptosporidiosis?

**A** There is no effective treatment for cryptosporidiosis, which is why people with immune problems are particularly advised to avoid contracting the infection in the first place if at all possible. The infection will usually clear up by itself in a healthy person, and taking antibiotics is not normally advised. It is generally good advice to increase your fluid intake when you have diarrhoea.

**Q** Are there special dangers to pregnant women?

**A** Not specifically related to *Cryptosporidium*. Pregnant women should follow the boil water advice for drinking water. Pregnant women who develop continuing diarrhoea should seek advice from their GP. *Cryptosporidium* is not a virus (rubella and other viruses can cause damage to the fetus if caught by mothers in pregnancy): it is a protozoan parasite which is not transmitted to the fetus.

**Q** If I am ill with cryptosporidiosis, can I pass it on to someone else?

**A** Yes, this is probably a common means of passing the infection on. It is important that you and your relatives/carers pay particular attention to hygiene and hand-washing, particularly before eating. If you are unwell, it is advisable to avoid handling or preparing food for others to eat.

**Q** Should drinks vending machines be used?

**A** Use of drinks dispensers is not recommended unless the water is heated to at least 70°C. Ideally, they should be turned off and disconnected from the water supply. When the water supplies are back to normal, the machine should be cleaned out before re-use, and any filters replaced as recommended in the manufacturer's operating manual. Care should be exercised in the handling and disposal of filters in case they are contaminated with *Cryptosporidium*.

Do not use cold drinks from vending machines.

**Q** I am a dialysis patient. Can you offer advice?

**A** Cryptosporidiosis is a gut disease and there is no risk of contracting it during dialysis. However, you should boil your drinking water as directed. If you have any other concerns please contact your dialysis administrator.

**Q** Does the boil water advice have any implications for swimming pool operation?

**A** The risk of illness from using a swimming pool should be minimal. Closure of swimming pools is not normally considered necessary during a boil water notice period. It is important that people who are symptomatic should be advised not to use public swimming pools.

**Q** I am a dentist. How can I protect my patients?

**A** Precautions should be taken to minimise the ingestion of drill bit cooling water, and mouth rinses should be made from boiled water that has been cooled. Dentists will normally have a separate tank feed for drill water, and in such circumstances the mains feed can be turned off and alternative safe supplies added directly.

**Q** I run a bar/restaurant. What precautions should I take?

**A** All water for drinking and food preparation should be brought to the boil and cooled as necessary. Bottled water may be used as normal. Fizzy drink dispensers should not be used unless they can be adapted to use sources other than tap water. Washing up should be carried out in water which has been heated to at least 70°C and allowed to cool, or in a dishwasher which heats to 70°C. Ice making machines should be disconnected, but ice may be made from water which has been brought to the boil and cooled.

**Q** I am a food/drink producer. How can I protect my products?

**A** If an alternative water source is not available, the mains water should be treated at the point of use by means of an appropriate filtration system. Only micro-straining filters capable of removing particles down to 1 micrometre in diameter should be used. Filters in this category include reverse osmosis units and those labelled 'absolute' 1 micrometer filters. Care should be taken in handling and disposal of used filter cartridges. Your local authority environmental health department will be able to give further advice.

# Appendix A4

# Guidance on the epidemiological investigation of outbreaks of infection[8]

**Contents**



[8] This Appendix is based upon a report prepared by the Public Health Laboratory Service under contract to the Drinking Water Inspectorate. The principal authors were:

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References

Sub-appendices

Sub-appendix A4.1 Investigation of sporadic cases of cryptosporidiosis

Sub-appendix A4.2 Regulation 29 – Requirement on water companies to prepare and maintain information on public record

Sub-appendix A4.3 Guidance to be given to interviewers

Sub-appendix A4.4 Case control study of (pathogen x) in (name of) health authority: protocol (example)

Sub-appendix A4.5 Cryptosporidiosis outbreak investigation questionnaire (example)

## A4.1 Introduction

1.1 High quality epidemiological information is vital in the investigation of possible outbreaks of waterborne infection associated with mains water consumption because microbiological evidence of water contamination by pathogenic organisms is usually difficult to obtain. Epidemiological investigations of such outbreaks are not straightforward; they are relatively uncommon and may be statistically complex, such that individual consultants in communicable disease control (CCDCs) or consultants in public health medicine (CPHMs) may appreciate help with investigations.

1.2 This manual has been produced to give CCDCs/CPHMs and other members of the Outbreak Control Team (OCT) practical advice to assist in the conduct of epidemiological studies. It is aimed at the investigation of outbreaks that may be associated with the consumption of mains water. As *Cryptosporidium* is the most commonly reported organism involved in such outbreaks the emphasis is on investigation of outbreaks caused by this organism. However, the general principles set out in this manual will apply to all outbreaks of potentially waterborne infection and to the investigation of all alternative hypotheses. The manual is not intended to replace local outbreak control plans, but it is hoped that CCDCs/CPHMs will find it a useful supplement and that its use will lead to the establishment of an agreed best practice in the investigation of water associated infection.

1.3 Investigation of suspected waterborne outbreaks involves four complementary activities.

- Epidemiological investigation of persons at risk.
- Microbiological investigation of patients, water, treatment works, animals and the environment.
- Engineering investigations.
- Investigation for evidence of a contamination incident at the water treatment works.

1.4 All four activities are important and need to be carried out in parallel and with close liaison between the investigating agencies. This guidance document will deal only with the epidemiological aspect of the investigation.

## A4.2 Recognition of an outbreak

2.1 High quality surveillance is the cornerstone of outbreak recognition and investigation. Surveillance includes:

- recognition by clinicians, especially general practitioners (GPs), of clinical cases if diarrhoeal disease and taking of stool samples;
- appropriate microbiological investigation of all diarrhoeal samples submitted to laboratories;
- reporting of laboratory results from cases;
- maintaining a current health authority (HA) database with regard to water associated events;
- regular review of the incidence of pathogens which might be associated with transmission by the waterborne route;
- regular liaison with water utilities concerning water quality data, for example, faecal indicator organisms, increased turbidity;
- prompt reporting of suspected outbreaks by CCDCs/CPHMs to the regional/national surveillance centres; and
- maintenance of national databases of water associated events.

2.2 Although *Cryptosporidium* is the most commonly reported organism in outbreaks associated with mains water contamination, other organisms may be responsible for outbreaks of waterborne disease, for example *Campylobacter, Giardia, Escherichia coli O157* and enteric viruses. Water should be considered as one of a number of possible vehicles of infection whenever an outbreak due to these organisms is being investigated.

2.3 Particular attention should be paid to monitoring and investigating cases of cryptosporidiosis (Furtado *et al* 1998). Sporadic cases of cryptosporidiosis should be interviewed to identify common risk factors using a short questionnaire. An example is given in Sub-appendix A4.1. Interviews by EHOs are already standard practice in many districts.

2.4 Thresholds for investigation according to level of *Cryptosporidium* incidence above baseline rates of disease should be agreed (Table 1).

*Table 1:* Suggested thresholds for investigation

| | **Observed number of cases in one week** | |
|---|---|---|
| Usual weekly rate* | Check | Alert |
| =1 | 3 | 4 |
| 2 | 5 | 6 |
| 3 | 6 | 8 |

* Allowing for seasonal variation

These numbers have been calculated to have only a 1 in 20 ‘check’ or a 1 in 100 ‘alert’ probability of occurring by chance assuming that the baseline numbers of cases per week follow a Poisson distribution (and could be used for any organism). Reaching the check or alert level in any one week is suggested as an indication to review recent cases, and to take further action if appropriate. Reaching the alert level for two consecutive weeks (which should only occur by chance about 1/10,000 weeks without an outbreak) is a suggested indicator for calling an OCT meeting. The CCDC/CPHM may decide that responses are appropriate at other levels of incidence.

## A4.3 Initial investigation

### 3.1 The Epidemiological Study Team

3.1.1 One of the major responsibilities of the OCT is to carry out appropriate epidemiological studies of the outbreak. These will normally be dealt with by a sub-group of the OCT. Typically, this will be led by the CCDC/CPHM, with input from EHOs and other members of the team. The regional epidemiologist (RE) or Scottish Centre for Infection and Environmental Health (SCIEH) representative will usually be invited to join the OCT for large or multi-district outbreaks.

3.1.2 Specific resources will be specifically required to undertake epidemiological studies effectively. These include access to relevant information e.g. water supply zones, population densities, HA databases for choice of controls; computer software, including maps and statistical packages to store and analyse the data; and staff trained to use them; access to expert statistical advice; data entry clerks and trained staff to carry out interviews for the questionnaire. Interviewers may be the most difficult resource to obtain, particularly if it is a large outbreak. Interviews may be conducted by EHOs and staff of the Department of Public Health Medicine. In England and Wales the RE may be able to recruit public health staff from elsewhere within or outwith the region to assist. However, the number of interviewers should be kept as small as possible to ensure that interviewing style is as uniform as possible throughout the whole data collecting exercise. The Information that is kept by water utilities on their public records is detailed in Sub-appendix A4.2.

3.1.3 Outbreaks can dramatically increase in size during the progress of an investigation. The burden of interviewing all new cases can be great, and the possible need for additional interviewers should be assessed periodically during an outbreak. It may be necessary to recruit interviewers from outside the groups recommended above. Other potential sources include hospital infection control nurses, health visitors and school nurses. However, as these groups are likely to be less familiar with the use of the relevant questionnaires, it is vital that they are appropriately briefed. Some of the factors to be considered in briefing interviewers are summarised in Sub-appendix A4.3.

### 3.2 Obtaining outside support

3.2.1 The RE, Public Health Laboratory Service (PHLS) Communicable Disease Surveillance Centre (CDSC), or SCIEH will be able to provide support. This support could include:

- providing regional/national incidence data on the relevant pathogen;
- advising or assisting with epidemiological studies;
- seeking or providing specialist registrar support;
- seeking or providing statistical support;
- co-ordinating epidemiological investigations if several districts are involved.

3.2.2 Issues in the design and analysis of such epidemiological investigations are complex. Expert statistical support is recommended, for example, from the PHLS Statistics Unit, as well as advice from experts in waterborne disease at national centres such as CDSC, SCIEH, the PHLS Cryptosporidium Reference Unit or the Scottish Parasite Diagnostic Reference Laboratory.

### 3.3 Definition of the study population

3.3.1 It is necessary to identify and define as precisely as possible the population considered to be at risk. This may be restricted to a specific age range, or by other factors, as well as to an identifiable administrative area. Definitions involving a potential risk factor, such as 'those living in given "water supply" areas' should be avoided because that could pre-empt the result. The population defined as 'those on the lists of health or local authorities in which cases have occurred' has proved useful in the past and is probably the safest definition to use unless there are particular local reasons against it. Less rigorous, but sometimes useful is the population defined by 'the combined practice lists of the GPs of the cases'. The cases and controls to be included in the epidemiological study must be recruited in such a way as to ensure that they are members of this population. For outbreaks in rural or seaside areas during holiday periods the population at risk is likely to include visitors who are temporary residents. Inclusion in the case definition should be considered but follow up may be difficult especially when urgent investigation is required.

### 3.4 Definition of cases

3.4.1 The initial case definition should be designed to include all those who could reasonably be part of the outbreak. It needs to define geographical, clinical and temporal parameters and whether temporary residents are included.

3.4.2 An example of an appropriate case definition is:

- **Probable case**: Any person resident in the area _____ who became ill with symptoms of _____ with date of onset on or after_____.
- **Confirmed case**: As probable case with stool specimen positive for _____.

### 3.5 Case finding

3.5.1 Cases should be actively sought through enhanced surveillance as the cases detected initially usually represent only a small proportion of the total cases occurring. Alerting local microbiology laboratories, GPs and

hospital doctors, and other public health departments, should help to identify additional cases. Regional and national case finding may be helpful especially if temporary residents are included in the case definition.

### A4.4 Descriptive epidemiology

4.1 The cases should be described by age, occupation, sex, time, place of residence and work/school, and by relevant known exposures obtained from initial interviews, for example, farm visits, recreational water contact, drinking water source and supply zones, food consumed and milk supply (see Sub-appendix A4.1).

4.2 If local microbiological screening for *Cryptosporidium* includes samples from adults, positives during waterborne outbreaks will usually be distributed throughout the age range. The median age for primary cases is often increased in comparison to the median for sporadic cases. A predominance of cases in school-aged children may indicate that infection has arisen from farm visits or swimming pools.

4.3 An epidemic curve should be created, and kept up-to-date, by plotting a bar chart indicating the numbers of cases, as the height of the bars on the horizontal scale according to the date on which symptoms first appeared. The pattern of the epidemic curve may suggest whether the outbreak originated from a point source, continuing source or as a result of person to person spread. The attack rates by area, eg local authority, water supply zone should be calculated. The denominator is the population at risk in the relevant area. The proportion of cases exposed to each of the suspected risk factors should be examined.

4.4 **Water supply zone.** Cases should be plotted on a map of water supply zones to determine whether distribution of cases matches the distribution of water supplies. Such maps should be provided by the water utility. Water from different treatment works is often blended so supply maps should ideally include information on the percentage of water from each treatment centre supplied to each zone. Statistical tests can be used to estimate the associations between infection rates and the percentage from each centre and the probability that differences in rates of illness in the different water supply zones occurred by chance.

4.5 **Proceeding to analytical study.** The OCT should review the evidence for water contamination and other suspected risk factors. Finding a statistically significant association between being ill and residence in a particular water supply zone does not necessarily mean that illness was caused by the water. However, in conjunction with evidence of mains water contamination, this can provide strong support for a hypothesis that mains water is the vehicle of infection and for the immediate implementation of control measures. In the absence of clear evidence to identify a vehicle of infection, an individual based analytical study is recommended. This may need to be done rapidly. In the presence of clear evidence of the vehicle of infection, if a boil water notice has been issued and/or population immunity is high, these factors should be borne in mind when deciding whether to proceed with an analytical study (see paragraph 5.2.6 of this Appendix).

## A4.5 Analytical study

### 5.1 Setting the objectives

5.1.1 The objectives of the epidemiological study should be clearly defined. These are usually to determine the size and extent of the outbreak and to identify the source of infection and its mode of transmission so that appropriate measures can be implemented to control the current outbreak and prevent a recurrence.

5.1.2 In the protocol for the analytical study it is necessary to specify aims and objectives which identify the hypotheses to be tested, the effects to be estimated and the information needed for both.

### 5.2 Hypothesis generation

5.2.1 In epidemiological studies it is usual to begin with the premise that there is no association between the exposure most under suspicion and the illness (null hypothesis) and then assess the extent to which the evidence is in conflict with this by carrying out an analytical study. The hypothesis will generally take the form of specifying that 'There was no association between exposure to a specified risk factor and an increased risk of infection'.

5.2.2 To generate hypotheses the value of good descriptive epidemiology cannot be overemphasised. The descriptive epidemiology from early cases should be reviewed in conjunction with the results of microbiological investigations, information on water quality and treatment and on any possible contamination of water sources.

5.2.3 More sensitive hypotheses for the investigation of waterborne infections use dose-response relationships. These take the form of specifying that 'There was no increase in the infection rate associated with increasing exposure to a specific risk factor'. Testing these hypotheses requires that data on habitual levels of exposure are collected for those factors which could have a dose-response relationship with the rate of infection.

5.2.4 Ideally, cases used to identify a risk factor and for generating a hypothesis should not be used in a subsequent analytical study to test that hypothesis. However, if there are few cases, the power of the study may be compromised by excluding these early cases. They can be included if full data can be collected from them. The final statistical analysis should be performed with and without these particular cases.

5.2.5 As soon as an outbreak is identified in which mains drinking water may be the vehicle of infection, investigators should use a full outbreak protocol and questionnaire. Examples derived from documents which have been used successfully to investigate previous waterborne outbreaks subsequently identified as due to *Cryptosporidium*, are contained in Sub-appendices A4.4 and A4.5. If the same questionnaire is used for cases and controls throughout the investigation, then cases from the initial interviews can be included in any subsequent case control study without it being compromised. Controls should be recruited as early in the investigation as possible.

5.2.6 In carrying out analytical studies, it may be helpful to recognise that:

- the power of epidemiological studies may be reduced if the population at risk has a higher level of immunity due to previous contamination incidents; and

- introduction of an 'advice to boil-water' notice may influence responses of cases and controls.

Dose-response relationships (and water supply zone attack rates) should be less affected by the above factors.

### 5.3 Cohort studies

5.3.1 A cohort study is one which includes all of a complete cohort of individuals who may have been at risk whether or not they have been ill and will generally have more statistical power than a case control study. As outbreaks associated with mains water mostly have large at risk populations, a cohort study is usually not feasible. Although a sample of the at risk population could be taken, attack rates are unlikely to be high enough for adequate statistical power. As a general rule, a cohort study should only be undertaken if:

(a) most of the cases have arisen from a readily identifiable cohort;

(b) it is feasible to identify and interview all members of the cohort, and

(c) it is plausible that the exposure responsible for the illness was limited to the members of the cohort.

5.3.2 An example of this situation would be an outbreak arising as a result of contamination of a private water supply. If a cohort study is undertaken, analysis follows much the same lines as that for case-control studies, which is described fully below.

## A4.6 Case control studies

### 6.1 General points

6.1.1 Where an analytical study is done and where the population at risk is large, that is in most outbreaks where mains water is a possible vehicle of infection, a case-control study is the most appropriate method of analytical investigation. The numbers of cases and controls should be sufficient to ensure that associations between potential risk factors and infection, which are large enough to explain the outbreak, can be shown to be statistically significant (see paragraph 6.5 below). The assumption that the vehicle of infection is water must always be tested.

6.1.2 Enquiries about the period of exposures normally refer to the incubation period before illness for the cases and to an equivalent period before interview for controls. Occasionally it may be known that exposures occurred during a specific calendar period. In that situation if the investigation is sufficiently timely, information should be requested on exposures during that period for both cases and controls. It is important that epidemiological studies should start as early in an outbreak as possible.

6.1.3 The case definition may be revised, if necessary, to exclude those individuals who were ill for reasons not related to the suspected source.

This would include travel abroad in the incubation period, or a household contact being ill during the incubation period prior to onset of symptoms in the case.

6.1.4 If possible, dose-response relationships should be assessed for all appropriate parameters, e.g. consumption of water and milk, frequency of immersion in swimming pool etc. This means collecting quantitative information on these exposures. Nearly all the controls and cases will have been exposed to some tap water. It is therefore important to collect detailed information on the amounts of unboiled water consumed by cases and controls, as well as its source. Questions on water consumption should be related to usual consumption (Sub-appendix A4.5, Q26).

## 6.2 Bias

6.2.1 Bias is defined in epidemiology as the 'deviation of results or inferences from the truth, or processes leading to such deviation'. There are many ways in which the design, execution, analysis and interpretation of epidemiological studies can introduce bias (Table 2). A major task in epidemiology is the recognition and avoidance of such biases. Incompleteness or changing completeness of case ascertainment is unlikely to introduce bias in the results of case control studies unless a substantial proportion of cases is misclassified as controls.

*Table 2* Examples of bias

| **Type** | **Explanation** | **How to reduce** |
|---|---|---|
| Selection bias | Controls may not be representative of the population at risk | Selection of controls (See section 6.4) |
| Information bias<br>(a) Recall bias | Cases may be more motivated to answer carefully than controls. If the suspected risk factor is known to cases and controls, this may influence response e.g. if an advice to boil water notice has been issued. (NB. This should not affect information on other risk factors and would be less likely to affect a dose response relationship) | Undertake interviews of both cases and controls within the same time frame as early as possible in the investigation. Use prompts and memory aids such as big local or national events |
| (b) Interviewer bias | The interviewer may question cases and controls in a different way thereby influencing the response. | Keep the number of interviewers to a minimum and ensure that they receive adequate instruction (Sub-appendix A4.3). Use the same interviewers and same interview method for cases and controls |

## 6.3 Confounding

6.3.1 Confounding occurs when the relationship of an exposure to a disease is distorted by another (confounding) exposure. This occurs when the confounding exposure is not only associated with the exposure under investigation but is also an independent risk factor for the disease.

6.3.2 Confounding can be minimised by careful study design. The effect of confounding factors can be managed in the statistical analysis of the study, but only if information on the confounding variable is collected. Therefore if any confounding factors are identified, data on them must be collected in the questionnaire.

6.3.3. For example if some cases had acquired their infection while travelling abroad their exposure patterns to other risk factors might well be the same as the controls, but not the other cases. This means that combining them with non-travellers in the analysis will make the exposures to tap water (say) of the cases more similar to the exposures of the controls and hence tend to hide the effect of this exposure. Such confounding with travel is usually dealt with by excluding travellers from the study.

6.3.4 It is important to remember that any one outbreak may have two or more vehicles of infection. For example, a higher incidence of cryptosporidiosis from farm animal contact may coincide with an episode of mains water contamination.

## 6.4 Selection of controls

6.4.1 Controls must be selected in a way that ensures they are an unbiased sample of the unaffected members of the population at risk. Controls should be free of the disease being studied, and should be within the same overall age range and area of residence as the majority of cases. They do not need to be the same sex unless the cases are predominantly of one sex. Inclusion or exclusion criteria, which apply to the cases (except their disease status), should apply equally to the controls.

6.4.2 **Matching**. This is a traditional way of dealing with confounding devised before statistical software for stratified multivariable analyses was widely available. Other than to ensure a roughly even distribution of age and sex between cases and controls (frequency matching), matching in studies of water associated outbreaks of infection where controls are relatively easy to obtain is rarely of much benefit. Matching can lead to wasted data, and confounding variables are best handled in the analysis of the data. Formal matching hardly ever increases the power and reliability of this kind of study and should only be used when there is clear evidence that it will be of benefit.

6.4.3 In investigating outbreaks of waterborne illness health authorities (HA) controls are recommended where feasible and especially if rapid recruitment is not compromised. These will be associated with particular cases and may be kept to a similar age range, but should not be considered as formally matched. However, although matching should generally be avoided ‘case nominated’ controls are a useful alternative when rapid recruitment is necessary. The analysis must then take full account of the matching between the cases and their ‘nominated’ controls.

6.4.4 **Health Authority/GP controls.** Controls are selected from the same health authority or general practice and from the same age band, that is <6 years, 6-15 years and over 15 years, as the case. This method is more feasible now that HAs have computerised population registers and can produce a more random sample of the population at risk. The main disadvantage in an urgent investigation by telephone interviews is that this method of control selection involves ‘cold calling’ and may reduce the response rate of controls. Random selection from the HA population is preferable to controls from the same GP list who would have to be regarded as matched controls. Delays can also arise from the need to gain approval of the GP.

6.4.5 **Case nominated community controls**. Each case is asked to nominate friends of similar age who live in the same community and who would be willing to act as controls. This is a quick and convenient method of obtaining controls when they might otherwise be difficult to obtain, and may control for some confounders. This method avoids 'cold calling' and overcomes the difficulties of contacting those with numbers not listed in the telephone directory. The main disadvantage is that such controls may be so similar to cases in relation to the exposure of interest that a true association with disease is not found. This is known as overmatching. This is more likely to occur in relation to social activity such as swimming pool exposure or farm visits than in relation to drinking water consumption but there is also a risk of overmatching on water supply.

6.4.6 Reluctance on the part of cases to give names of controls can often be overcome by suggesting that the case contacts potential controls for permission and that the interviewer phones the case again to confirm this before contacting controls.

6.4.7 Ideally, cases and controls should be interviewed in a standardised way, using the same interview method by a limited number of experienced interviewers as soon as possible after the identification of the outbreak.

**6.5 Sample size calculations**.

6.5.1 The main aim of the study will be to test for an association between the risk of becoming infected and a range of risk factors such as 'household pets', 'farm visits' or 'tap-water consumption'. These associations are generally measured by odds ratios (ORs). The size of the study will determine how much power it has for detecting odds ratios of different magnitudes.

6.5.2 Since the size of the outbreak will not be easy to predict the number of cases available for the study will be uncertain. However, it is wise to calculate the likely power of the study for differing numbers of cases covering the numbers likely to be available.

6.5.3 Small outbreaks where the number of cases are in the range 10 to 50 will only have sufficient power to detect large ORs, but studies of this size will still give estimates of the effects of the various risk factors and can help identify which risk factors are and are not likely to be associated with the infection.

6.5.4 Unless there are many cases they should all, as far as is possible, be included in the study, but the number of controls is a matter of choice. Where the number of cases is small recruiting up to three controls for each case will increase the power of the study. Conversely if there are many cases sufficient power may be achieved with more cases than controls up to a ratio of three to one. Increasing either ratio to more than three to one is generally not very useful because the power of a study is largely limited by the size of the smaller group.

6.5.5 In general it is sensible to start recruiting two or three controls per case and review this strategy as the outbreak progresses.

6.5.6 Finally for a given study size, the power to detect a dose-response relationship between the risk of infection and increasing tap-water consumption is somewhat greater than that for detecting an equivalent

association with an all or nothing exposure. Thus quantitative data on tap-water consumption (and on other exposures where dose response can be readily measured) should be collected in the questionnaire to ensure that this type of analysis is possible. The volume estimation may be inaccurate and some guidance in the questionnaire is recommended (for example, one glass is equivalent to a third of a pint or about 200 millilitres). See Sub-appendix A4.5, Q26.

### 6.6 Data collection

6.6.1 **Questionnaire design.** Questions should be short, clear and designed to obtain unambiguous answers that are free from bias. Details of illness in the case and household members, of recent travel abroad, and of all likely known risk factors should be included. Postcodes are important for mapping to water supply zones.

6.6.2 For investigation of outbreaks of cryptosporidiosis suspected of being related to drinking water contamination the same questionnaire (for example, see Sub-appendix A4.5) should be used to interview all cases and controls. Additional questions generated by local circumstances or the preliminary interviews should be included. Local points of relevance e.g. names of swimming pool, farms open to public should be added where appropriate. Questions should be worded so that they are completely unambiguous. Optional responses must be exhaustive and mutually exclusive. 'Not applicable', negative, and 'Don't know' responses should be recorded in a distinguishable way. Apart from 'skip sections', no answer space should be left blank. It may be helpful to consult with regional/national epidemiologists and a statistician before the questionnaire is finalised.

6.6.3 **Questionnaire administration.** Questionnaires can be administered in face to face interviews, by telephone or by post. Face to face interview is the best way of administering a questionnaire and is recommended in preliminary interviews but is frequently not possible for use in the main case control study because of resource constraints. Telephone administered questionnaires are likely to be the best available option unless telephone ownership in the study population is unusually low. The interviewer can ensure the questions are understood and answered correctly, but does not have to travel to find the person. Evening or weekend calls are more likely to produce an unbiased group of controls than daytime calls. Considerable efforts should be made to ensure that all questions are answered so that the interaction between the effects of several exposures can be thoroughly investigated. Clear interviewing instructions must be given for both face to face and telephone interviewers (Sub-appendix A4.3) to increase the reliability of the data collected.

6.6.4 Questionnaires sent by post are less likely to be completed correctly and fully than those completed by interview and have longer return times. Postal questionnaires should be considered if the other suggested methods are not practicable, or if large numbers of people have to be contacted and there is no urgency.

6.6.5 **Data entry.** Data should be entered into a computerised database. Direct input of interview data via a data entry screen representing the questionnaire is the best way to avoid data transcription errors. Accurate data entry should where possible be facilitated by double entry comparison together with range and consistency checks at entry.

## A4.7 Statistical analysis of case-control study data.

### 7.1 Introduction

7.1.1 The analysis must produce and the results be presented to give:

(i) a clear summary of the data available in terms of the age and sex distributions of cases and controls and the symptomatology, microbiology and outcomes of the cases;

(ii) a set of simple estimations and tests of the associations with infection for all the risk factors considered individually as single risk variables; and

(iii) a multivariable analysis assessing the affects of various risk factors considered together to allow for the possibility of confounding.

7.1.2 The magnitudes of the associations found should be indicated by ORs or 95% confidence limits. The actual *p* values from tests of ORs against 1.0 should also be given to indicate the strength of the evidence for a genuine association. Formal testing of primary hypotheses stipulated in the protocol (for example, there is no dose-response relationship between the rate of infection and the consumption of unboiled tap water) is usually made at the 5% significance level. If multiple variables are being tested, *p* values should be treated with caution.

7.1.3 If a matched case-control study design has been used the analyses differ slightly from those described below, in that the case-control pairs must be kept as a unit throughout and the important information is whether and how the members of each pair differ in their exposures. Strong positive associations will lead to a large proportion of pairs with the cases exposed and the controls not exposed. A full discussion of the analysis of matched and unmatched case-control data is given in Schlesselman (1982), and Tillet (1986) gives a good overall summary. The analyses described below assume that the controls were not formally matched with specific cases, for example in HA selected controls.

### 7.2 Single risk variable analysis

7.2.1. This should consist of a sequence of analyses estimating the associations between illness and exposure for each exposure considered individually, including investigations of dose-response relationships and simple stratified analyses to assess and adjust for confounding between two exposure variables.

7.2.2. These analyses should be performed by generating two-by-two tables summarising the numbers of cases and controls exposed and not exposed to each of the risk factors. The tables should be used to obtain ORs (or relative risks for cohort studies) to estimate the magnitude of associations with 95% confidence limits and *p* values to indicate the strength of evidence for such associations. Exposure variables with k categories (k being the number of categories) where $k \geq 2$, e.g. types of occupation or categories representing increasing exposure or dosage, will require k by 2 tables and odds ratios will need to be obtained for each category against one chosen as a reference category. This is usually the first in an ordered sequence of categories.

7.2.3. The analysis should include $\chi^2$ tests of dose-response trends in such k x 2 tables, where the categories represent a steadily increasing exposure such as increasing consumption of tap water. These can readily be performed with statistical software using a method originally due to Armitage (1955) for testing linear trends in a sequence of proportions. In this case the proportions are, for each dosage category, (no. of cases)/(no. of cases + no. of controls). Note that this must not be considered or presented as independent information to that obtained from a test of association with the same risk factor dichotomised, for example, tap water consumption into 'no tap-water' and 'any tap-water' unless the zero consumption category is omitted from the trend analysis. In the single variable analysis the latter approach will generally be the most appropriate.

7.2.4. It may be possible to demonstrate an association between illness and another factor such as age or area of residence. In that case it will be useful to investigate the association between illness and tap water consumption separately in two or more strata representing different age groups or areas. The associations if found would then be adjusted for any confounding due to age group or area effects.

7.2.5. Such a stratified analysis can be particularly useful to assess how dose-response trends with tap water consumption differ between individuals living in areas with water supplies which differ in the proportions of water they receive from a suspect source.

### 7.3 Multivariable logistic regression analysis

7.3.1 This method of analysis provides a comprehensive way of assessing associations with a variety of exposures and of estimating and testing dose-response relationships allowing for the possibility of the effects of one factor being confounded with another. To ensure confounding of any sort does not bias the results this type of analysis is essential. However, since it requires complete questionnaire responses for all risk factors included in each regression it may not always be possible to use all the relevant data in estimating the effect of an individual risk factor. In addition there are rarely more than one or two risk factors in acute outbreaks of infectious disease. For these reasons it is generally best, as long as their direction and magnitude are confirmed by the regression results and not influenced by confounding, to consider the single variable results, which use the maximum information on that variables association with disease, as the most reliable. Logistic regression can be performed using the most professional statistical software including the public domain software EPI-Info which was designed for outbreak investigations with an additional logistic regression module. Both are available from a World Health Organization Internet site. See References.

7.3.2. The logistic regression analysis should endeavour to include all the risk factors where the single variable result was insufficient to exclude them as associated with illness. Since studies may include many potential risk factors and questionnaire responses are often incomplete, some strategy is needed for restricting which risk factors are selected. One such strategy which has proved effective is to omit potential risk variables with odds ratios from the single variable analysis with $p>0.2$ excepting, at this stage, any risk factors known to have been associated with this infection in the past if a substantial proportion of the cases have been exposed. The results of the logistic regression including the selected factors can then be used to restrict the selection further by identifying, for omission, those

factors with no evidence of an association once other factors were taken into account and the process repeated. In this sequence factors with any substantial proportion of responses missing should be omitted as soon as possible unless they show strong evidence of an association. If this is not done the analysis will suggest conclusions based only on part of the data. If this is not a substantial part of the data set, ie if it is not at least 70% of both the cases and controls the results cannot be considered reliable. The resulting model can then be used to identify those associations that are reduced to insignificance when other factors are taken into account and those that persist.

7.3.3. As stated above, because the logistic regression is generally unable to use all of the data, the final conclusions will be better based on the single variable odds ratios, given that they are supported by the logistic regression.

7.3.4. In general the conclusions will convey whether or not there was a dose-response relationship between tap water consumption and infection rates, what the evidence was for associations between infection rates and other factors and what, based on the analytic study results, was the most likely vehicle by which the infection was spread.

### A4.8 Report writing

8.1 A preliminary report should be written once an outbreak is recognised, ideally within 24 hours. Follow-up reports should be written as required. The final report should include the study protocol, questionnaire and examples of press releases and letters used in the study for the benefit of other investigators. Report writing is the responsibility of the OCT. However, the epidemiology study team should prepare the report of the epidemiological study. A copy of the report should be sent to regional/national epidemiological centres to ensure that information on outbreaks is collated and used to inform future practice.

### Useful references


Armitage, P. (1955) Tests for linear trends in proportions and frequencies. *Biometrics* **11**, 375-386

CDC Atlanta Epi Info software download site: http://www.cdc.gov/epo/epi/software.htm

Furtado, C., Adak, G.K., Stuart, J.M., Wall, P.G., Evans, H.S. and Casemore, D.P. (1998) Outbreaks of waterborne infections intestinal disease in England and Wales, 1992-1995. *Epidemiology and Infection* **121**, 109-119.

Golding, A.M.B., Noah, N,. Stanwell-Smith, R. eds (1994) Water and Public Health. London: Smith Gordon.

Hunter, P.R. ed (1997) Waterborne Disease. Chichester: John Wiley.

Meinhardt, P.L., Casemore, D.P. and Millar, K.B. (1997) Epidemiological aspects of human cryptosporidiosis and the role of waterborne transmission. *Epidemiological Reviews* **18**, 118-136.

Schlesselman, J.J. (1982) Case-Control Studies – Design, Conduct, Analysis. Oxford:OUP.

Tillett, H.E. (1986) Statistical Analysis of Case-Control Studies of Communicable Diseases. *International Journal of Epidemiology* **15**, 126-133.

UN WHO Epi Info and Logistic regression module download site: ftp://ftp.unaids.org/inet/ftp/epi/index.html

# Sub-appendix A4.1

**INVESTIGATION OF SPORADIC CASES OF CRYPTOSPORIDIOSIS**

**Case Number**.....

Form completed by ..... Date.....

**NB:** If you are answering on behalf of a child please remember that the questions relate to the child and not to you.

1 First name .....Surname.....

2 Address .....

3 Postcode.....Telephone .....

4 Sex: **Male/Female** Date of Birth .....

5 Please state occupation (both parents if case is a child) .....

6 If case is at school/preschool group, give name and location.....

.....

7 Date of onset of illness .....

8 Did anyone in your household have diarrhoea (three or more loose stools in 24 hours) in the two weeks before illness started?

**Yes/No**

**Travel away from home**

9 In the two weeks before the onset of illness did **you/your child** spend one or more nights away from home?

**Yes/No/Not sure**

If yes was this: **Local/Other/UK Abroad**

| Place visited | Date returned |
|---|---|
| | |
| | |
| | |

**Food and drink**

10 In the two weeks before the onset of illness did **you**/**your child** drink:

| **Drink** | **Yes** | **No** | **Not sure** | **Name of shop or supplier** |
|---|---|---|---|---|
| Unpasteurised cows milk | | | | |
| Goats/sheep milk | | | | |
| Cold pasteurised milk (exclude hot milk, include cold milk on cereal) | | | | |

11 What is the source of your drinking water?

Mains: **Yes**/**No** If yes, specify water company.....

Private: **Yes**/**No**

Both: **Yes**/**No**

Other: **Yes**/**No** Specify.....

Do you have a pet/animal that has been ill in the last two weeks? **Yes**/**No**

12 In the two weeks before the onset of illness did **you**/**your child** swim in a pool or paddling pool?

**Yes**/**No**/**Not sure**

If yes, please name pool and location .....

.....

13 In the two weeks prior to illness, did **you**/**your child** take part in any activity involving contact with water (eg swimming, or playing in a river or pond)?

**Yes**/**No**/**Not sure**

If yes, please specify name and location .....

.....

14 Did **you**/**your child** have any contact with farm or zoo animals in the month prior to the onset of illness?

**Yes**/**No**/**Not sure**

If yes, please give name and address of farm or zoo .....

.....

Do you have or have you been in contact with a pet/animal that has had diarrhoea in the last two weeks?

**Yes**/**No**

Comments: .....

.....

.....

# Sub-appendix A4.2

**Regulation 29 – Requirement on water utilities to prepare and maintain information on Public Record**

A5.2.1 The Water Supply (Water Quality) Regulations 1989 define a water supply zone as an area that is designated by a water undertaker whether by reference to a source or sources of supply or the number of people supplied in which not more than 50,000 people reside. A source in this context could be the outlet of a water treatment works or a number of water treatment works supplying water of similar quality, a pumping station, a blending point or a service reservoir. Sizes of zones vary from about 50 people to the maximum although very small zones are gradually disappearing

A5.2.2 Regulation 29 requires water companies to prepare and maintain, in respect of each of its water supply zones, a public record containing:

(i) the name of the zone;

(ii) the name of any water treatment works supplying the zone;

(iii) the population of the zone;

(iv) details of any relaxations of water quality standards applying to the zone;

(v) details of any undertakings applying to the zone (ie remedial or improvement action affecting any water treatment works or service reservoirs supplying the zone or any part of the distribution system within the zone which is linked to an undertaking given to the Secretary of State);

(vi) results of all individual compliance samples taken from water treatment works, service reservoirs and the distribution system; and

(vii) annual summary analytical information including, where appropriate, a commentary on water quality.

A5.2.3 This public record must be available at all offices normally open to the public. Anyone can view it and obtain free of charge information on the zone in which they reside. A nominal charge may be made to provide other information from the record. Analytical information can be deleted after five years.

A5.2.4 There is no requirement for maps to be provided with the public record but in practice most water companies include these so that people using the record may identify the zone where they live. Under an Information Direction, water companies are required to provide the Inspectorate with detailed maps of water supply zones and update these annually but at present there is no requirement for these to be provided to local authorities or health authorities.

# Sub-appendix A4.3

# Guidance to be give to Interviewers

### A5.3.1 The Interviewer's Responsibilities

The interviewer's job is to see that the interview is completed honestly and accurately according to the instructions specified during training. Specifically this means:

- Clearly understanding the nature of and content of the questions before starting the interview.
- Being familiar with all instructions.
- Making sure the interview is conducted with the correct respondent.
- Recording a true picture. This means neither adding nor deleting any information.
- Executing the work clearly and accurately.
- Striving for maximum efficiency without sacrificing quality.
- Being courteous, friendly, and wearing a smile.

### A5.3.2 Basic Interviewing Rules

- Be sure that you speak with the appropriate respondent.
- Read all questions word for word.
- Never explain, interpret or add to a question.
- Read all questions in the exact order in which they appear.
- Do not skip appropriate questions even if you feel you know the answer.
- Never hurry the interview.
- Remain objective. Do not indicate surprise, pleasure or disapproval at any respondent's answers.
- Be prepared to probe on some answers.

(Source: CDC, Atlanta)

### A5.3.3 Telephone interviews – things to consider

**Voice Personality**

■ Be courteous and polite

■ Sound confident

■ Do not sound bored

■ Sound interested in response/s

**Probing and Clarification**

■ Probe for accurate information

■ Know when to probe

■ Use neutral probe/s

**Enunciation of Questionnaire**

■ Speak clearly

■ Pronounce words properly

**Handling Difficult Respondents**

■ Answer respondent's questions as fully as possible

■ Alleviate confidentiality concerns

■ Encourage responses when respondent is reluctant

■ Alleviate length of interview concerns

**Interviewing Techniques**

■ Verify phone number

■ Read question(s) verbatim

■ Follow skip pattern smoothly

■ Go from introduction to questions smoothly

■ Close interview smoothly

■ Make appointments properly

■ Provide neutral feedback

**General Knowledge of Survey**

■ Recognize need for data quality

■ Know survey objectives

■ Know rationale for questions

(Source: CDC Atlanta)

# Sub-appendix A4.4

## Case control study of (pathogen X) in (name of) Health Authority: Protocol

### A5.4.1 Introduction

An increase in cases of (pathogen X) is being investigated in (name of health authority). Descriptive epidemiology of the (number of cases) cases has revealed no specific risk factors. The emergence of new cases without any clear risk factor has prompted a case control study.

### A5.4.2 Aims of the case control study

To identify the source of (pathogen X) causing the increase in cases in order to inform appropriate control measures.

### A5.4.3 Objectives of the case control study

- To provide descriptive epidemiology of cases
- To determine if there is an association between specific risk factors and infection with (pathogen X)

### A5.4.4 Case definition

A case is defined as a person:

- with diarrhoea (three or more loose stools in 24 hours); **and**
- the presence of (pathogen X) in a faecal specimen; **and**
- no other faecal pathogen isolated; **and**
- onset after (date the outbreak started)

Cases are excluded from the case control study if they:

- have a history of travel outside the UK since the date the outbreak started; and or
- had a household contact with new onset severe diarrhoea in the two weeks prior to onset of symptoms

### A5.4.5 Case finding

Enhanced surveillance and case finding are underway. Local laboratories and GPs have been alerted to the increase in cases. GPs have been asked to request stool specimens on any patient with diarrhoea. Laboratories are testing all stool specimens for (pathogen X) and are reporting positive stools to the CCDC/CPHM by fax and phone.

GPs of the patients with positive stool specimens will be contacted by the health authority to inform them of the investigation and to check that there is no reason why the case should not be contacted. The case will be visited at home, or telephoned by a CCDC/CPHM/EHO who will administer the questionnaire.

### A5.4.6 Selection of controls

The study will be unmatched. Controls will be taken at random from the HA register. Three controls will be sought from the same practice list as each case. In order to facilitate stratified sampling, controls will be of the same sex as the case and within the same age band as the case. The age bands are age 5 years or less, age 6 to 15, and over age 15.

Local GPs will be informed that a case control study is being carried out. Once control names have been taken from the FHSA register the GP will be contacted to ask if there is any reason why a particular control should not be included in the study.

Controls will be excluded if they have:

■ had any diarrhoea since (date outbreak started); and or

■ a history of travel outside the UK since (date outbreak started)

### A5.4.7 Questionnaires

Questionnaires will be administered face to face, or by telephone, by environmental health officers or public health doctor. Cases will be asked about risk factors in the two weeks immediately prior to onset of symptoms. Controls will be asked about risk factors for the two week period before interview.

The questionnaire will be the same for cases and controls apart from details of illness and will include the questions which were used for the initial descriptive epidemiological studies.

### A5.4.8 Data analysis

Data will be entered and analysed by (name of person undertaking data entry and analysis).

When to stop the study will depend on the pattern of the outbreak and the ability to recruit cases and controls. The minimum sample size calculated for an association to be found with an odds ratio of around three will be at least (number derived from sample size calculation) cases.

# Sub-appendix A4.5

## Cryptosporidiosis outbreak investigation

**Health Authority:**.....**Identification No** .....

*Please ring as appropriate, tick box or write in the space provided.*

**Case $_1$/Control $_2$**

Interview date...../...../.....Interviewer .....

**Interview method: Telephone $_1$/Face to face $_2$/Postal $_3$**

**Personal details**

1 Forename.....Surname.....

2 Sex: **Male $_1$/Female $_2$**

3 Age: Years.....Months (if aged less than one year) .....

4 Date of Birth...../...../.....

5 Address.....

Postcode.....Telephone .....

6 If case/control is an adult – occupation .....

7 If case control is a child, does he or she attend

School: **Yes $_1$/No $_2$**

Nursery or playgroup: **Yes $_1$/No $_2$**

Address of school or nursery .....

.....Postcode.....

Parent's occupation (Mother). .....

Parent's occupation (Father) .....

8 How many people live in your household

| Adults (16 years and older) | Children (5–15 years) | Children (under 5 years) |
|---|---|---|
| | | |

9 GP name.....

GP address.....

GP telephone number.....

10 Health Authority.....

11 Local Authority .....

12 Water supply zone (from water company information).....

**CONTROLS ONLY (QUESTIONS 13-15)**

13 Method of control selection: **Case nominated/GP nominated/HA Register**

Nominated for which case.....

**Exclusion criteria**

14 Have **you/your child** been ill with diarrhoea (three or more loose stools in 24 hours) at any time since (date of onset of outbreak as defined in case definition.....)

**Yes** $_1$/**No** $_2$

If **YES**, thank and end interview, if **NO**, continue.

15 In the two weeks prior to this interview have **you/your child** travelled outside the UK?

**Yes** $_1$/**No** $_2$

If **YES**, thank and end interview, if **NO**, continue.

**CASES ONLY (QUESTIONS 16-23)**

16 Have **you/your child** been ill with diarrhoea (three or more loose stools in 24 hours) since (date of onset of outbreak as defined in case definition.....)

**Yes** $_1$/**No** $_2$

If **NO**, thank and end interview, if **YES**, continue.

17 In the two weeks before the diarrhoeal illness did **you/your child** travel outside the UK?

**Yes** $_1$/**No** $_2$

If **YES**, thank and end interview, if **NO**, continue.

18 Did anyone in your household have diarrhoea in the two weeks before **you/your child** became ill?

**Yes** $_1$/**No** $_2$

If **YES** thank and end interview, if **NO** continue.

19 When did **you/your childs** diarrhoea start?...../...../.....

20 For how many days approximately were **you/your child** ill?.....

21 Did **you**/**your child** have any of the following symptoms?

| Sympton | Yes | No | Not sure |
|---|---|---|---|
| (a) Fever | 1 | 2 | 3 |
| (b) Abdominal pain | 1 | 2 | 3 |
| (c) Vomiting | 1 | 2 | 3 |
| (d) Blood in stools | 1 | 2 | 3 |
| (e) Other | 1 | 2 | 3 |

If Other please specify (1).....

(2).....

22 Were **you**/**was your child** admitted to hospital for this illness? **Yes** $_1$/**No** $_2$

If **YES**, which hospital? .....

Admission dates...../...../.....to...../...../.....

23 Laboratory confirmed case? **Yes** $_1$/**No** $_2$ Specimen date...../...../.....

Laboratory.....Lab No (1).....(2).....(3).....

**CASES AND CONTROLS**

*Cases should be asked about their exposure in the two weeks prior to the onset of their symptoms. Controls should be asked about exposure during the two weeks prior to the interview.*

**Background information**

24 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, how many nights did **you**/**your child** spend away from home?.....

Where did **you**/**your child** visit?

**Local (within 10 miles of home)** $_1$/**Other UK** $_2$

| Place visited | Number of nights away |
|---|---|
| | |
| | |
| | |

**Consumption of water**

25 Do **you**/**your child** drink any cold tap water or drinks containing cold tap water (without boiling it first)?

**Yes $_1$/No $_2$/Not sure $_3$**

If **YES** continue, if **NO**, go to question 27.

26 About how much cold (unboiled) tap water (including water used to dilute in squash or fruit juice) do **you**/**your child usually drink per day**? *Answer as **number of glasses per day on average** (NB one glass is about 1/3 of a pint or about 200 millilitres).*

(i) From the tap at home .....

(ii) From the tap at work/nursery/school.....

Address and/or post code .....

(iii) From the tap elsewhere.....

Address and/or post code .....

27 In the two weeks prior to illness/interview, did you drink any cold (unboiled) tap water (including water used to dilute in squash or fruit juice) in places not usually visited (public houses, parties, restaurants etc)?

Place..... Number of glasses drunk.....

Place..... Number of glasses drunk.....

28 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, did **you**/**your child** have any drinks with ice added?

**Yes $_1$/No $_2$/Not sure $_3$**

If **YES**: **at home $_1$/at work or nursery or school $_2$/elsewhere $_3$**

If elsewhere, please specify.....

29 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, did **you**/**your child** drink any bottled water?

**Yes $_1$/No $_2$/Not sure $_3$**

**carbonated $_1$/still $_2$/both $_3$**

If **YES** Brand. .....

How many glasses of bottled water do you drink on average per day?.....

30 Do you use a water filter at home?

**Yes $_1$/No $_2$/Not sure $_3$**

If **YES:** Type. .....

31 In the two weeks before the onset of **illness (for cases)/date of interview (for controls)**, did **you/your child** drink:

| Type of water | Yes | No | Not sure | Please specify where |
|---|---|---|---|---|
| (a) Any untreated water (private well, borehole, spring, lake, river or seawater) | 1 | 2 | 3 | |
| (b) Water from a drinks dispenser | 1 | 2 | 3 | |
| (c) Water from a drinking fountain | 1 | 2 | 3 | |

32 Type of water supply at home. **Mains $_1$/Other $_2$/Other $_3$**

If not mains, please specify.....

33 Was there any disruption to the mains supply in the two weeks preceding **illness (for cases)/date of interview (for controls)**?

**Yes $_1$/No $_2$/Not sure $_3$**

If **YES**, give details.....

**Contact with water**

34 In the two weeks before the onset of **illness (for cases)/date of interview (for controls)**, did **you/your child** visit a swimming pool?

**Yes $_1$/No $_2$/Not sure $_3$**

Name of pool..... About how many times?.....

Name of pool..... About how many times?.....

Name of pool..... About how many times?.....

About how many times was the head immersed on each occasion?

| None | 1 | 1-2 | 2 | 3-7 | 3 | >7 | 4 | Not sure | 5 |
|---|---|---|---|---|---|---|---|---|---|

Did **you/your child** swallow water during a swim?

**Yes $_1$/No $_2$/Not sure $_3$**

35 In the two weeks before the onset of **illness (for cases)/date of interview (for controls)**, did **you/your child** go boating/windsurfing/water skiing/canoeing/swimming in a river or lake.

**Yes $_1$/No $_2$/Not sure $_3$**

If **YES**, where.....About how many times?.....

How many times was the head immersed on each occasion?

| None | 1 | 1-2 | 2 | 3-7 | 3 | >7 | 4 | Not sure | 5 |
|---|---|---|---|---|---|---|---|---|---|

Did **you**/**your child** swallow water?

**Yes** $_1$/**No** $_2$/**Not sure** $_3$

36 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, did **you**/**your child** have any other contact with fresh water (eg other water sports, fishing)?

**Yes** $_1$/**No** $_2$/**Not sure** $_3$

If **YES**, please give details.....

**Consumption of food**

37 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, about how often did you eat the following:

| Food | not at all | 1–2 times | 3–7 times | most days | not sure | Brand (if appropriate) and source |
|---|---|---|---|---|---|---|
| Lettuce | 1 | 2 | 3 | 4 | 5 | |
| Other green salad | 1 | 2 | 3 | 4 | 5 | |
| Tomatoes | 1 | 2 | 3 | 4 | 5 | |
| Coleslaw | 1 | 2 | 3 | 4 | 5 | |
| Raw vegetables | 1 | 2 | 3 | 4 | 5 | |
| Fresh fruit | 1 | 2 | 3 | 4 | 5 | |
| Undercooked burgers | 1 | 2 | 3 | 4 | 5 | |
| Another undercooked or raw meat | 1 | 2 | 3 | 4 | 5 | |
| Raw shellfish | 1 | 2 | 3 | 4 | 5 | |
| Soft cheese, uncooked | 1 | 2 | 3 | 4 | 5 | |
| Hard cheese, uncooked | 1 | 2 | 3 | 4 | 5 | |
| Yoghurt | 1 | 2 | 3 | 4 | 5 | |
| Ice cream | 1 | 2 | 3 | 4 | 5 | |
| Cream | 1 | 2 | 3 | 4 | 5 | |

38 Before eating do you normally wash the following foods with tap water:

| Type of food | Yes | No | Not sure |
|---|---|---|---|
| (a) Lettuce | 1 | 2 | 3 |
| (b) Raw vegetables | 1 | 2 | 3 |
| (c) Fruit | 1 | 2 | 3 |

39 In the two weeks before the onset of **illness (for cases)/date of interview (for controls)**, did **you/your child** eat or drink any unusual or new foods?

**Yes** $_1$/**No** $_2$/**Not sure** $_3$

If **YES**:.....Food.....Supplier.....

40 Do **you/your child** drink unheated **pasteurised** milk (eg with cereal or as milkshake)?

**Yes** $_1$/**No** $_2$

If **YES**, how many glasses on average do **you/your child** drink per day?.....

Supplier .....

41 Do **you/your child** drink unheated **unpasteurised** milk (including goats and sheep milk)?

**Yes** $_1$/**No** $_2$

If **YES**, how many glasses on average do **you/your child** drink per day?.....

Supplier .....

**Contact with pets/animals**

*NB Contact with animals refers to touching, feeding, being licked or other such close contact, and not to being in the same room or house.*

42 In the two weeks before the onset of **illness (for cases)/date of interview (for controls)**, did **you/your child** have contact with animals at home (pets)?

**Yes** $_1$/**No** $_2$

If **YES**, (a) type of pet .....

(b) did the pet have diarrhoea **Yes** $_1$/**No** $_2$

43 In the two weeks before the onset of **illness (for cases)/date of interview (for controls)**, did **you/your child** have contact with any:

(a) zoo animals **Yes** $_1$/**No** $_2$

If **YES**, name of zoo .....

(b) farm animals **Yes** $_1$/**No** $_2$
(*prompt with farm visits, animals brought to school, visits to pet shops*)

If **YES**, name of farm from where animals originated .....

44 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, did **you**/**your child** have any visits to a farm:

**Yes** $_1$/**No** $_2$

If **YES**, name and address of farm .....

Details of visit (eg contact with slurry, animal faeces) .....

.....

45 In the two weeks before the onset of **illness (for cases)**/**date of interview (for controls)**, did **you**/**your child** have contact with any other animals or birds (excluding farm and zoo animals) eg at a friend's or neighbour's house?

**Yes** $_1$/**No** $_2$

If **YES**, name and address of household .....

.....

**Additional questions relevant to individual outbreaks**

**Thank you for completing this questionnaire.**

If you have any comments you think may be useful, please use the space below.

Questionnaire completed by:

Name.....Status.....Date.....

# Appendix A5

## *Cryptosporidium* in natural mineral waters and beverages[9]

### A5.1 Water and the food industry

A5.1.1 The food industry utilises large volumes of either public water supplies or private treated borehole-derived water for its manufacturing and ancillary processes. Uses of water include: direct incorporation into foods as an ingredient; washing of food containers (eg cans prior to passing into high risk processing areas); washing raw vegetables, raw fruits and animal carcasses etc; transport of materials eg in flumes; factory cleaning; and in cooling systems (eg in canneries) and for boiler systems and steam raising.

A5.1.2 All water used in direct food contact situations must be at least of potable quality and should be free of pathogenic microorganisms.

A5.1.3 Conservation of water resources is important for commercial reasons in the food industry, and where possible, systems have been devised to use water in economical ways (eg counter-current washing of vegetables where the produce is moved from the 'dirty' end of the tank towards the incoming 'fresh' water at the 'clean' end), and filtration of and recycling of water used for preliminary soil removal from root vegetables destined for further processing.

A5.1.4 Of paramount importance to the food industry is the consistent availability of food-safe water for use in direct contact with foods and food contact surfaces.

A5.1.5 Cleaning water, which has the potential to become contaminated with pathogens washed from produce, can be discharged to waste, with or without treatment. In 1982, the food industry used about 660 million tonnes of water, of which about 40% was from public water supplies and which cost approximately £150 million for supply and disposal (Whitman, 1982). Alternative sources include boreholes and rivers as well as canals, estuaries and lakes with the majority of it being non-potable. Water used for cleaning is now recognised as a limited resource and interest has been identified in its reconditioning, especially in the use of reconditioned water for the preparation and processing of food, given that the microbiological safety and quality of each food is also assessed (Palumbo *et al*., 1997).

A5.1.6 From the above, it can be seen that public water supply usage in the food industry is variable, being dependent upon both the industry and the specific manufacturer.

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[9] A paper by Professor Huw Smith, Scottish Parasite Diagnostic Laboratory

## A5.2 Consumption of bottled water in the UK

A5.2.1 In the last seven years, the consumption of bottled water in the UK has nearly doubled, rising from 480 x $10^6$ litres in 1991 to 895 x $10^6$ litres in 1997. In 1991, bottled water accounted for a 5.8% share of the soft drinks market, by 1997 this had risen to 8.7%. Currently, bottled water represents the highest growth sector in the soft drinks industry, with the 1997 figures representing an increase of 12.6% over the 1996 figures. This equates to an increase in *per capita* average annual consumption from 8.3 litres in 1991 to 15.2 litres in 1997, with the retail value estimated at over £400 million in 1997.

A5.2.2 Changing lifestyles, effective marketing and distribution, increase in prosperity and travel as well as concerns about tap water are all factors identified for this increase in consumer demand. Bottled waters (consisting of water packaged in retail packs or in water coolers), can be still or sparkling and can be subdivided into mineral, spring and table waters. Of the 895 x $10^6$ litres of bottled water consumed in 1997, 56% of demand was accounted for by still water, 30% by sparkling water and 14% by still water coolers. Of the 895 x $10^6$ litres of bottled water consumed in 1997, 73% of demand was home produced with 23% accounted for by imported brands. Mineral water accounted for 69% of this demand whereas sparkling and table (and others) water accounted for 27% and 4% respectively.

A5.2.3 In 1986, the demand for sparkling water was twice that for still water, with the majority of product being sourced from outside the UK. Ten years later, the situation has been largely reversed with a greater demand for still rather than sparkling water. Spring water accounted for a 22% share in the market and water coolers for a 12% share. By the end of 1997, 165,000 water coolers had been installed, predominantly in offices, accounting for 14% of the bottled water volume.

A5.2.4 The only approved treatments for bottled natural mineral waters (and bottled spring waters under the new regulations) are filtration or decanting as well as the addition or removal of carbon dioxide (the new regulations, currently in draft, will permit the use of ozone treatment). Section 7.4.a of the Natural Mineral Waters Regulations, 1985 states that 'At source and thereafter, up to and including the point of sale, a natural mineral water shall be free from parasites and pathogenic microorganisms ....'. Both mineral and spring waters are derived from managed sources which must comply with current regulations. Table waters can be produced from either groundwater or potable water sources and can be subjected to both chemical and physical treatment as well as disinfection.

## A5.3 Other trends

A5.3.1 900 x $10^6$ litres of pure (100% fruit) fruit juice, 420 x $10^6$ litres of (<100% fruit) fruit drinks and 2850 x $10^6$ litres of dilutables were consumed in the UK in 1997. All are increases above 1996 figures. Soft drinks account for 73% of all liquid consumption by children between the ages of 5 and 12 and under 20% of adult consumption. A 1997 survey by Saatchi & Saatchi identified that children influence 60% of family soft drink purchases and that by the age of 11, over 50% buy their own drinks. A bottled water aimed at consumption by babies was introduced in 1997.

A5.3.2 There is increasing concern in both the bottled water and food industry of the potential for waterborne *Cryptosporidium* oocysts to contaminate the product. In situations where water for drinking, bottling or vending can be treated prior to use, microfiltration membranes or similar can be incorporated by Industry to reduce further the likelihood of *Cryptosporidium* contamination. However, where the product receives no treatment whatsoever prior to bottling, currently the case for natural mineral waters, there is intense interest in developing a reliable method for detecting and characterising parasites, especially *Cryptosporidium*, in support of the HACCP based systems operated in producing companies.

## A5.4 Natural mineral waters

A5.4.1 Natural mineral waters are derived from officially recognised sources and are governed by the Natural Mineral Water Regulations (Anon 1985). Prior to recognition as a mineral water, the source must be protected against pollution and be subjected to a hydrogeological survey. The source water must be described physico-chemically and must meet a number of defined microbiological and chemical standards.

A5.4.2 The annual consumption of bottled natural mineral waters in the UK rose from <5 million litres in 1975 to 200 million litres in 1988 (Stickler, 1989). Since that time, marketing of both the perceived and aesthetic quality of the product have encouraged demand. In 1992, demand amounted to a market of £297 million per annum (Richards *et al.*, 1992). In 1993, as published by the European Commission in their official journal (Anon., 1993), 'the UK recognised 58 natural mineral water trade names as being derived from, and exploited as, natural mineral waters'. In addition, two 'third' country trade names are recognised. UK guidelines governing the microbiological quality of natural mineral waters for human consumption (Anon., 1989) identify that eukaryotic parasites should be absent from the sample tested however, there appears to be no consensus method(s) for demonstrating the presence of eukaryotic parasites in these waters. Furthermore, neither the volume to be analysed nor the biological status of the parasite is identified.

A5.4.3 The increasing concerns regarding the likelihood of the occurrence of protozoan parasites in natural mineral waters, at Government, consumer and Industry levels, led the Ministry of Agriculture, Fisheries and Food to fund a research project (FS1223) to develop a method for detecting *Cryptosporidium parvum* oocysts and *Giardia duodenalis* cysts in natural mineral waters.

## A5.5 Survival of *Cryptosporidium* in water

A5.5.1 In the majority of studies on the survival of *C. parvum* oocysts in various water types, the fluorogenic vital dyes assay of Campbell *et al*. (1992), the *in vitro* excystation method of Robertson *et al.* (1993) or both have been used to assess oocyst viability. Robertson *at al*. (1992) studied the survival of three oocyst isolates within semi-permeable membranes in selected environments including raw water, tap water and reverse osmosis laboratory grade 1 water. In all environments, including the grade 1 water control, the proportion of dead oocysts increased gradually over time with death being most rapid in oocysts placed in river and tap water. The table below indicates that, for two UK oocyst isolates stored in tap water, a reduction of between 13 and 32% was determined over a 7 week period.

*Table 5.1* Alterations in the proportion of percentage potentially viable oocysts over time in various aquatic environments

| | ISOLATE A | | | ISOLATE B | | |
|---|---|---|---|---|---|---|
| Time (days) | Grade 1 water | Tap | River | Grade 1 water | Tap | River |
| 0 | 77.7 | 77.7 | 77.7 | 83.8 | 83.8 | 83.8 |
| 11 | 75.2 | 75.0 | 85.4 | 70.9 | 71.9 | 64.1 |
| 19 | 73.7 | 81.1 | 47.0 | 82.3 | 58.8 | 81.1 |
| 33 | 74.3 | 69.7 | 55.7 | 71.7 | 60.4 | 60.0 |
| 47 | 77.5 | 64.4 | 62.9 | 66.0 | 51.0 | 45.1 |

The % of potentially viable oocysts [DAPI(+) PI(-) and DAPI (-) PI(-)] was demonstrated using the fluorogenic viability assay of Campbell *et al*, (1992).

## A5.6 Survival of *Cryptosporidium* in natural still mineral waters

A5.6.1 Storage of bovine-derived oocysts at 4˚C in various commercially produced natural mineral waters does not appear to affect their viability as determined both by the fluorogenic vital dye and the *in vitro* excystation assays, with viability levels after 9 weeks of storage being similar to those determined at day 0. Some decline in the viability (10–30%) of human-derived *C. parvum* oocysts stored in mineral waters at 20˚C occurred after 3 weeks of storage. The reduction in viability appears to be temperature-related rather than related to the (mineral) water type in which the oocysts are immersed, since the same source of human-derived oocysts stored at 4˚C does not demonstrate a comparable decline in viability even after storage for seven weeks.

## A5.7 Survival of *C. parvum* oocysts in beverages and infant formula

A5.7.1 Friedman *et al.* (1997) assessed the survival of five week old oocysts following storage in orange juice (pH 3.87), cola beverage (pH 2.46), bottled beers (pH 3.8) and infant formula (pH 6.65) for between one and seven days. Oocyst survival was assessed at 4˚C in orange juice and infant formula and at both 4˚C and 22˚C in cola and beer. Both the fluorogenic vital dyes assay and morphological assessment were used to determine oocyst viability and experiments were performed in duplicate. Morphological assessment of viability did not correlate with the fluorogenic vital dyes assay.

A5.7.2 Water was used as a control and 70% of oocysts stored in water at 4˚C were viable after 24h, while 59% of oocysts stored at 22˚C remained viable after 24h.

A5.7.3 By 48h, a large percentage of oocysts stored in beer were deemed to be empty and/or non-viable. Between 32% and 46% of oocysts remained ‘full’ (8–20% viable) after 24h storage in beer at 4˚C, whereas between 20 and 22% of oocysts were ‘full’ (7–12% viable) after 24h storage in beer at 22˚C. On average 14% of oocysts remained viable when stored at 4˚C in carbonated beverages compared with 9% viable when stored in carbonated beverages at 22˚C. After 24h storage in orange juice, 65% of oocysts were viable, whereas 89% remained viable in infant formula.

## A5.8 Survival of *C. parvum* oocysts in bottled waters and beverages

A5.8.1 Waters and beverages which are carbonated or have high acid (and $SO_2$) content (eg fruit juices) are expected to affect oocyst survival adversely. There are no long term (>2 weeks) studies which have been conducted on the survival of oocysts in bottled waters (excluding data presented above on still mineral waters), beverages, soft drink concentrates or fruit juices.

A5.8.2 *Effect of carbonation*: Carbonation can reduce the pH of the product to between approximately pH 4 – 6, depending both on the degree of carbonation and the pH of the still product and should have the likely effect of reducing the long term survival of oocysts.

A5.8.3 *Effect of extremes in pH, and the effect of acid*: Acid, a major constituent of stomach secretions, does not affect the survival of oocysts over short time periods and can be used to good effect for maximising the effects of *in vitro* surrogates of *C. parvum* viability (Campbell *et al*., 1992; Robertson *et al.*, 1993). In addition, at extremes of pH (e.g. 2.75 and 10), the short term survival (1 to 2 hours) of oocysts appears to be unaffected. Storage for longer periods of time is expected to reduce oocyst survival although the exact kinetics are currently unknown.

A5.8.4 *Effect of adjuncts and storage*: The term adjuncts covers a variety of natural and artificial components added to the water base. Included are natural and artificial flavourings and enhancers, alcohol and other preservatives, stabilisers, etc. Some adjuncts are likely to exert a protective or null effect while others may reduce oocyst survival. As for still natural mineral waters, an increase in oocyst death is likely to occur during storage in products with long shelf lives (and which may contain some form of preservative) with storage at higher (20˚C) temperatures increasing the rate of die off.

## A5.9 Description of method developed during MAFF project FS1223 for detecting *Cryptosporidium* spp. oocysts in natural mineral waters

A5.9.1 Four mineral water types, which demonstrate interferences/interactions with various steps in the proposed method have been chosen as gold standards. Deionised water serves as control. Three methods are being developed: one for large volumes (>1,000 litres), and two for small volumes (1 litre and 10 to 20 litres) of product. The methods take note of equipment already in analytical laboratories and, where possible, attempt to avoid the use of expensive equipment (e.g. large centrifuges, flow cytometers, laser scanners, etc.). The current procedure can be divided into two sections, namely: (a) sampling, elution and concentration and (b) identification.

**(a) sampling, elution and concentration**

A5.9.2 *Large volume sampling*: The filters identified for large volume filtration can filter between 1,000 and 20,000 litres of sample. Recovery efficiencies range from 20% to approx. 60%.

A5.9.3 *Small volume sampling*: Between 1 and 20 litres of product can be filtered through flat bed 47mm or 13mm membranes and oocyst recoveries (using seeds of 50 oocysts) range from 56.1% ± 11.9 to 106.4% ± 11.1. Recoveries of seeded oocysts from 1 litre volumes of mineral waters range from 32.8 ± 7.3 to 59.0 ± 13.1%. Small volume

centrifugation (≤250 mL) recovers >75% of oocysts. Recovery using immunomagnetisable separation is no better than recovery by filtration and/or centrifugation and introduces a further manipulation (where oocysts could be lost) and cost for no identifiable benefit.

**(b) identification**

A5.9.4 *Microscopy*: Oocysts can be identified and enumerated by epifluorescence microscopy on glass microscope slides or 13 mm membranes using standard methods. The enhanced fluorogenic morphological method of Grimason *et al.,* (1994), using the nucleic acid intercalator (DAPI) enables easier recognition of intact oocysts either on slides or on membranes.

A5.9.5 *PCR*: A method for DNA release has been developed and optimised for *Cryptosporidium* which causes the release of up to 90% of nuclei from oocysts. The polymerase chain reaction (PCR) method has been developed for detecting small numbers (1.9 ± 1.1 to 55.1 ± 7.8) of oocysts in 1000x concentrates of the mineral waters tested. Restriction fragment length polymorphism (RFLP) of the PCR amplified product (amplicon) can differentiate *C.parvum*, *C.muris* and *C.baileyi*. The amplicon contains two endonuclease sites which can be digested after PCR to produce products of varying sizes. An internal control which co-amplifies with the *Cryptosporidium* primers has been developed and the inclusion of this in the PCR allows estimation of the amount of DNA amplified and to extrapolate it to the number of oocysts in the sample (current range for the internal control = 1–19 oocysts).

### A5.10 Other issues

A5.10.1 *Exploitation*: Issues such as hydrogeology, catchment type and catchment control are primary concerns prior to exploitation. The hydrogeology of groundwaters and the influence of surface water sources on groundwater quality are dealt with by the subcommittee on groundwater protection.

A5.10.2 *Catchment control*: The control of oocyst contributors both in the catchment as well as areas which impinge on the catchment is of prime importance. The principles of source protection as well as the necessary procedures required to maintain standards are identified in the guide to good bottled water standards (1995) published by the British Soft Drinks Association. This guide is currently being revised. Specific information on *Cryptosporidium* and catchment control, identified in Badenoch (1990; 1995) and Smith *et al*., (1995) may also prove useful when identifying oocyst contributors and best practice for exploiters of natural mineral waters.

## References


Anon (1985). The Natural Mineral Waters Regulations. Ministry of Agriculture, Fisheries and Food. London, UK. HMSO.

Anon (1989). Guidelines for the recognition and exploitation of natural mineral waters. Draft document. Ministry of Agriculture, Fisheries and Food. 11pp.

Anon (1993). List of natural mineral waters recognised by the UK. (93/C224/12). *Official Journal of the European Communities* No. C224 (19.08.93) 18-20.

Badenoch, J. (1990). *Cryptosporidium* in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Badenoch, J. (1995). *Cryptosporidium* in water supplies. Second Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 108pp.

Campbell, A.T., Robertson, L.J. and Smith, H.V. (1992). Viability of *Cryptosporidium parvum* oocysts: correlation of *in vitro* excystation with inclusion/exclusion of fluorogenic vital dyes. *Journal of Applied and Environmental Microbiology* **58:** 3488-3493.

Friedman, D.E., Patten, K.E., Rose, J.B. and Barney, M.C. (1997). The potential for *Cryptosporidium parvum* oocyst survival in beverages associated with contaminated tap water. *Journal of Food Safety* **17:** 125-132.

Grimason, A.M., Smith, H.V., Parker, J.F.W., Bukhari, Z., Campbell, A.T. and Robertson, L.J. (1994). Application of DAPI and immunofluorescence for enhanced identification of *Cryptosporidium* spp. oocysts in water samples. *Water Research* **28:** 733-736.

Palumbo, S.A., Rajkowsi, K.T. and Miller, A.J. (1997). Current approaches for reconditioning process water and its use in food manufacturing operations. *Trends in Food Science and Technology* **8:** 69-74.

Ritter, W.F. (1997). Water sanitation in the food processing industry. *Journal of the Association of Food and Drug Officials*. **61:** 30-37.

Richards, J., Stokely, D. and Hipgrave, P. (1992). Quality of drinking water. *British Medical Journal* **304:** 571.

Robertson, L.J., Campbell, A.T. and Smith H.V. (1993). *In vitro* excystation of *Cryptosporidium parvum*. *Parasitology* **106:** 13-19.

Smith, H. V., Robertson, L. J. and Ongerth. J. E. (1995). Cryptosporidiosis and giardiasis: the impact of waterborne transmission. *Journal of Water Supply Research and Technology – Aqua* **44(6):** 258-274.

Stickler, D.J. (1989). Microbiology of bottled natural mineral waters. *Journal of the Royal Society of Hygiene* **4:** 118-24

Whitman, W.E. (1982). How much water does the food industry use? Presented at the FDIC Water Quality Seminar, London, 1982. Leatherhead Food Research Association, Technical Note No. 1. 5 pp.

# Appendix A6

## Keeping your private water supply safe[10]

### A6.1 Protecting your water supply

A6.1.1 The purpose of this leaflet is to alert the owners and users of private water supplies to the risks of contamination of their water. Although most private water supplies are probably safe to drink most of the time, this leaflet gives details of the risks and of what you can do to protect your supply. A number of serious illnesses can be contracted from contaminated water supplies.

### A6.2 What is a private water supply?

A6.2.1 In general terms a private water supply is any water supply which is not provided by a water company. It is not a 'mains' supply. About 1% of the population of England and Wales have private water supplies to their homes. Most private supplies are situated in the more remote, rural parts of the country. The source of the supply may be a well, borehole, spring, stream, river, lake or pond. The supply may serve just one property or several properties through a network of pipes.

### A6.3 What is the problem?

A6.3.1 Safe drinking water is essential to good health. All private water supplies can pose a threat to health unless they are properly protected and treated. They may become contaminated with bacteria, protozoa, parasites and viruses (micro-organisms) or other substances. Many of these are harmless, but some may cause serious illness or even death in vulnerable people. You may not be able to tell whether your water is safe as contamination may not show by smell, taste or colour of the water. Unlike public supplies, many private supplies are not treated to remove contamination.

A6.3.2 This leaflet explains a number of things you can do to protect your supply and reduce the risk of contamination. It also gives you information about different types of private water supply and things which may contaminate them.

### A6.4 What should I do?

A6.4.1 Find out about your water supply.

- Who is responsible for the upkeep and maintenance? If this is not clear, consider reaching an agreement with other users.

- Where is the source?

[10] This Appendix contains the text of the leaflet on Private Water Supplies produced by the Drinking Water Inspectorate

■ Where does it go to get to your property?

■ Is it treated in any way?

■ Is the treatment equipment in good order and serviced regularly?

### A6.5 Keep your supply safe

A6.5.1 Make sure you inspect all parts of your supply, including the catchment area, regularly to check it is in good order and has not been interfered with or damaged. Any defects found should be put right as quickly as possible.

### A6.6 Supplies from springs, wells or boreholes

A6.6.1 Check that the source is adequately protected to stop surface water getting into your supply, particularly at times of heavy rain.

### A6.7 Supplies from streams, rivers, lakes or ponds

A6.7.1 The collection arrangement should include a settlement pond to allow larger particles to settle out before the water flows into your supply.

A6.7.2 The collection arrangement should also include a sand or gravel filter after the settlement pond to remove organic material and small animals. These filters may not remove all micro-organisms and will not remove chemical contamination.

A6.7.3 Ensure that the water being collected is not contaminated by discharges from a septic tank, any sewage discharge upstream or run-off from farmland.

### A6.8 Supplies from farmland where animals graze or manure is spread

A6.8.1 Divert rain water run-off so it does not flow into your supply (for example, with a small ditch leading away from your supply).

A6.8.2 Check that the farmer is aware of the drinking water supply and the need to avoid contaminating it by farming activity.

A6.8.3 Fencing may be necessary to stop farm and other animals from gaining access to areas where they could contaminate the water source.

### A6.9 If your supply has water collection chambers and/or storage tanks

■ These should have watertight and vermin-proof walls and lids.

Tops of chambers or tanks should be above ground level to prevent water from surrounding land flowing into them.

■ Any overflow pipes or vents in chambers and tanks should be designed to stop animals and debris from entering them.

■ The collection chamber should not be close to any soakaway or drain.

### A6.10 If you supply water to others

A6.10.1 If you supply water to other people in the course of a business, for example by renting out holiday accommodation or using water for food production, you have a duty of care towards customers for the safety of water you supply. For example, if you think your water may be unsafe, you may have to:

- advise boiling water for drinking and food production; or
- use or suggest others use bottled water.

### A6.11 Should I get my supply checked?

A6.11.1 Local councils monitor the quality of all private supplies, although depending on how many people use the supply, this may not happen often. If you want to know the results of previous tests of your supply contact your local council to obtain a copy of the analysis.

A6.11.2 You can also contact your council if you:

- would like to have an additional test done; or
- are the sole user of a supply and would like to have your water tested for the first time.

A6.11.3 The council is likely to charge you for this service. Alternatively, you can arrange for your own first time or additional tests using a private laboratory. Remember that a water test can only tell you about the quality of the water at the time of the test. The quality of your water may change at different times. Your local council can advise you what the result of the test on your water supply means.

### A6.12 Should I get my supply treated?

A6.12.1 If your supply is contaminated or may become contaminated, you can and should install treatment for your private water supply. If it serves more than one household it is better (and probably more economical) to install treatment for the supply as a whole rather than in individual households.

A6.12.2 Many different forms of treatment are available and your choice must suit your particular supply and the type of contamination. Your local council will be able to give advice about treatment methods or other improvements to your water supply. If you know your supply is contaminated with micro-organisms you should boil all water to be used for drinking and food production until suitable treatment is installed.

### A6.13 What else can I do?

A6.13.1 If you no longer want to use your private water supply, you can ask your local water company about the possibility of connecting to the public supply (‘the mains supply’). You will probably have to pay all the costs. Contact your local water company for further information (see your phone book under ‘Water’).

## A6.14 Sources of contamination

A6.14.1 **Micro-organisms.** A number of serious illnesses can be contracted from water that is contaminated with certain micro-organisms. The most likely source of contamination of the water will be animal droppings. Water supplies drawn from farmed land where animals graze or where manure is spread are most at risk. The risk is particularly high at times of heavy rainfall when water may run directly off farmland and carry micro-organisms into private water supplies. Discharges from cess pits or septic tanks are another likely source of contamination.

A6.14.2 All those who drink contaminated water are at risk of infection. However, the risk for those who do not drink the water regularly and are not used to it, such as visitors and guests, is likely to be greater. It may be advisable to use boiled or bottled water for drinking.

A6.14.3 **Chemicals.** Private water supplies may be affected by chemicals:

- used in farming or forestry, for example nitrate and sheep-dip chemicals and other pesticides; nitrate is only a problem in some parts of England and Wales, water containing high nitrate is particularly unsuitable for bottle-fed babies;

- used in industrial or commercial premises or workshops, for example solvents from metal finishing industries or dry cleaning premises; and/or

- occurring naturally in the catchment area, for example iron or manganese or colour from peat; these may affect the appearance, taste or smell of the water, but on the whole, these are not dangerous to health.

A6.14.4 If you are not sure whether your water is affected, ask your local council for advice about the level of nitrate or other chemicals in your water supply.

A6.14.5 **Lead.** This can be particularly harmful to very young children. Many private water supplies in England and Wales are naturally acidic and may dissolve lead. If your water supply passes through a lead tank or pipes, either outside or inside the home, it is likely there will be a significant amount of dissolved lead in your water and you may need to replace your tank or pipes.

## A6.15 Types of private water supply

A6.15.1 **Springs, boreholes and wells** that draw water from **deep** underground sources are less likely to be contaminated than supplies from other sources but water may become contaminated where:

- the spring emerges;

- the water collects in the boreholes or well.

A6.15.2 In farmland, underground water can pick up nitrate (from fertilisers) or pesticides which may be harmful to health. Water from **shallow** wells and springs which draw water from **close** to the surface of the ground is usually less satisfactory and more likely to be contaminated.

A6.15.3 **Streams, rivers, lakes and ponds.** The quality of water from these sources will generally not be as good as that from springs, boreholes and wells. The quality of water will also vary depending on the weather conditions. It is most likely to be contaminated, particularly with micro-organisms, at times of:

- high rainfall;
- warm weather.

A6.15.4 Water that runs across the land into streams, rivers and lakes picks up contamination from various sources, for example, from the soil and from the droppings of farm animals, wild animals and birds. Some of this contamination may be dangerous to health.

### A6.16 Further questions or advice

A6.16.1 If you have any questions or want any advice about your private water supply, contact the environmental health department of your local council.

# Appendix A7

## Molecular and antigenic aspects of *Cryptosporidium* and cryptosporidiosis (a brief review)[11]

### A7.1 Introduction

A7.1.1 *Cryptosporidium parvum* is widely distributed in livestock animals, wildlife and humans. In man it is generally considered to be a zoonotic infection. Although there is considerable evidence for zoonotic transmission (Casemore *et al* 1997) evidence from early epidemiological studies on cryptosporidiosis led, partly intuitively, to the view that there were two cycles of infection, zoonotic and urban, with the latter predominating (Casemore & Jackson 1984; Casemore *et al* 1987). It has become clear in the UK and elsewhere that water provides a major route of transmission (Badenoch 1990; Badenoch 1995; Casemore 1990; Meinhardt *et al* 1996; Furtado *et al* 1998; Nichols & Rutter 1998). The dynamics of these outbreaks is complex, reflecting a number of variables, including, water treatment and distribution factors, parasite characteristics and immunity of an exposed community, both individual and population (Meinhardt *et al* 1996).

A7.1.2 The major source of *C. parvum* oocysts in water was assumed to be agricultural livestock. Phenotypic (antigenic and enzymatic) and molecular genetic studies have permitted the study of phylogenetic and 'strain' or isolate characterisation and comparison (Webster 1993; Jenkins & Petersen 1997; Morgan & Thompson 1998; Widmer 1998) and the study of immunological factors (Frost *et al* 1998; McLauchlin *et al* 1998) which are beginning to shed light on these complexities.

### A7.2 Characterisation and comparison of isolates for epidemiological purposes

A7.2.1 **Phenotypic Methods.** Early attempts to characterise *Cryptosporidium* species, particularly *C. parvum,* by use of phenotypic polymorphisms indicated that species and individual isolates or 'strains' could be distinguished (Nichols et al 1991; Nina *et al* 1991; 1992a,b). Subsequently, Moran *et al* (1995) and McLauchlin *et al* (1998), used antigens of various molecular weights in a Western blotting method, with a monoclonal antibody probe, enhanced chemiluminescence and percentage similarity matrix analysis of banding patterns. By this means it was possible to distinguish between isolates of *C.parvum* in a way which was of value in epidemiological investigations (see below).

A7.2.2 A significant development with a different phenotypic characterisation method was the demonstration by Awad El-Kariem *et al* (1993; 1995; 1996) that isoenzyme (zymodeme) studies subdivided *C. parvum* into two phenotypes, one associated with animals and the other

[11] Paper prepared by Dr DP Casemore, Public Health Laboratory Service Cryptosporidium Reference Unit, Rhyl PHLS, Wales, UK.

with humans. The authors proposed that this supported the occurrence of two transmission cycles within animal and human hosts. Similar zymodeme studies in the 1970s, subsequently confirmed by molecular genetic studies, had demonstrated that the parasite *Entamoeba histolytica* comprised two morphologically identical species, one pathogenic and the other a commensal (Clark 1998) – a finding with far-reaching public health implications.

A7.2.3 **Genotypic Methods.** Genetically based molecular studies using polymerase chain reaction (PCR) supported and expanded on these phenotypic findings. These included random amplification of DNA polymorphisms or of specific polymorphic genetic loci, while, for example, the 18S small subunit ribosomal RNA has been completely sequenced and up to eight chromosomes have been identified. Numerous reports have confirmed the distinction of the main species and indicated 'strain' or isolate variation (Awad El-Kariem 1994, 1998; Bonnin *et al* 1996; Carraway *et al* 1996; Cai et al 1992; Chrisp & LeGendre 1994; Elwin *et al* 1998; Gibbons *et al* 1998; Kilani & Fayer 1994; Kilani & Wenman 1994; Laxer *et al* 1991; Leng *et al* 1996; Morgan *et al* 1995, 1997, 1998; Nichols *et al* 1991; Nina *et al* 1991, 1992; Ortega *et al* 1992; Petesen *et al* 1992; Patel *et al* 1998; Peng *et al* 1997; Pozio *et al* 1992; Spano *et al* 1997, 1998; Vasquez *et al* 1996).

A7.2.4 Thus, for example, Leng *et al* (1996), using restriction enzyme analysis, showed that *C. parvum, C. baileyi* and *C. muris* could be distinguished. Bonnin *et al* (1996) used PCR restriction fragment length polymorphism (PCR-RFLP) assay to develop markers that distinguish *C. parvum* isolates. Ten out of ten calf isolates were identical while two patterns were observed among human isolates – seven out of thirteen were indistinguishable from the calf isolates, while six out of thirteen presented a different profile.

A7.2.5 Carraway *et al* (1996) used PCR for two markers, based on polymorphisms in random amplified DNA, and ribosomal RNA nucleotide sequences, to show two subdivisions among isolates, one associated with various host species and one from humans alone. Awad-El-Kariem *et al* (1996; 1998) used arbitrary-primed PCR to support their earlier isoenzyme marker findings and suggested that a variety of phenotypic and genotypic markers could be used to distinguish *Cryptosporidium* species and also *C. parvum* isolates of human and animal origin. Spano *et al* (1997) used PCR-RFLP analysis with a *Cryptosporidium* oocyst wall protein (COWP) gene to show that *C. parvum* differed from *C. wrairi*, and that isolates of *C. parvum* of human and animal origin could be distinguished.

A7.2.6 Peng *et al* (1997) looked at 39 isolates of human (including some from waterborne infection) and bovine origin. They identified two banding patterns, which they referred to as genotypes 1 and 2. Type 1 was observed only in human isolates, while genotype 2 pattern was observed in calf isolates and in human isolates from cases known to have been exposed to calf excreta. The authors viewed their results as supporting the concept of two distinct transmission cycles in humans. This concept was given further support by Morgan *et al* (1998) who used PCR incorporating analysis of two gene sequences to examine a variety of isolates to confirm the existence of more than one genotype of *C. parvum*. One of these was found only in man, while the other type found in man was also found in calves.

A7.2.7 Vasquez *et al* (1996) described the nucleotide sequence of the *C. parvum* dihydrofolate reductase (DHFR) gene, showing differences between two isolates, one from an AIDS patient and one from a calf. Gibbons et al (1998) used a PCR endonuclease restriction method to study the nucleotide sequence of the DHFR gene of *C. parvum*. This method showed complete correlation with earlier studies, including the demonstration of animal/human and apparently human-specific subtypes of this parasite.

A7.2.8 Patel *et al* (1998) and Pedraza-Diaz *et al* (1998) used PCR-RFLP with the COWP gene (Spano *et al* 1997) to study isolates from a large number of cases involved in waterborne outbreaks, and from sporadic infection in humans and animals, in the UK (see below). They confirmed the presence of the animal/human and apparently human-specific types. Elwin et al at the PHLS Cryptosporidium Reference Unit have developed a rapid typing system based on previous published sequence of the TRAP-C2 gene (Peng *et al*, 1997), and have confirmed the presence of three genotypes (Elwin *et al*, 1998).

### A7.3 Infectivity

A7.3.1 Of particular importance is the finding by several workers that isolates of the apparently human-specific type of *C. parvum* do not infect mice or calves (Awad-El-Kariem *et al* 1998; Peng et al 1997; Pozio *et al* 1992; McLauchlin *et al*, unpublished). The findings described by Pedraza-Diaz *et al* (1998) suggest the possibility of a selective process leading to development of infection by genotype 1 (human type) when humans are exposed to oocysts from mixed origins. Looking at the small amount of data available, including infectivity studies (Meinhardt *et al* 1996), one can speculate that the infective dose for homologous host transmission is lower than for transmission between different hosts.

### A7.4 Molecular and antigenic epidemiological studies

A7.4.1 Molecular methods developed to characterise isolates have been used to study waterborne outbreaks (McLauchlin *et al* 1998; Peng *et al* 1997; Pedraza-Diaz *et al* 1998; Patel *et al* 1998). Surface-derived water sources are likely to lead to exposure to isolates from a variety of sources, both human and animal. Hence, such episodes may not involve an identifiable ‘outbreak strain’ but many strains, the occurrence of an outbreak being determined by a complex set of conditions. Infections in individuals may also be with one or more isolates.

A7.4.2 McLauchlin *et al* (1998) used Western blots with a monoclonal antibody probe (Moran *et al* 1995) to characterise isolates from individuals in some waterborne outbreaks in the UK. By this method, isolates divided into eight band pattern groups. Their findings provided support for several possibilities, including that:

- reproducible marker (antigenic) banding patterns could be identified and analysed for ‘relatedness’ or clustering;
- there was evidence for restricted band patterns (single ‘strain’) in some samples from individuals, families and outbreaks; and
- there is evidence for multiple (ie mixed infection) types occurring in outbreaks, in families and in individuals.

A7.4.3 Genetically based methods (PCR-RFLP with the COWP gene) were used by Patel *et al* (1998) and Pedraza-Diaz *et al* (1998) to study isolates from waterborne outbreaks in the UK. Of 133 stool isolates from three outbreaks, 130 (98%) were genotype 1 (human) and 3 were genotype 2 (animal/human), with one identified as mixed and one of a third genotype pattern. In contrast, among 87 sporadic cases 47 (54%) were genotype 1 and the remainder genotype 2; 100% of animal isolates were genotype 2. A further group of isolates from cases in another small outbreak associated with a small rural private supply were all of the animal type, genotype 2 (Pedraza-Diaz *et al*, unpublished data). From these data it may be inferred that the primary source of oocysts in the three former outbreaks was likely to have been sewage effluent.

## A7.5 Serological aspects

A7.5.1 The evidence from the studies in the UK and elsewhere shows that a picture has emerged of the complex dynamics, which includes several key variables, including parasite variability and the effect of individual and population immunity (immuno-prevalence) in the exposed community (Meinhardt *et al* 1996; McLauchlin *et al* 1998). Using a series of low and high molecular weight oocyst-derived antigens, (McLauchlin *et al* 1998) showed antibody responses in sera from subjects in an outbreak area, collected within nine months of the outbreak, and in control groups. The subjects included confirmed cases, persons from the outbreak area who were not known to have been symptomatic and others from a geographically distinct source. Reactions to the selected antigens were found in sera of >88% of nine known convalescent cases, in 32-49% (depending on antigen) of 37 persons living in the area of the outbreak; in 15-21% of 34 from an adjacent area, and <7% of 58 geographically distinct control subjects.

A7.5.2 Further work is being done to characterise these responses but the differences between the groups were most notable with the small molecular weight antigens, which are believed to be those most indicative of recent infection. Frost *et al* (1998) in the USA have also studied sero-prevalence in communities subject to differing exposure histories, including water source types and have noted significant differences which appear to reflect their epidemiological patterns.

## A7.6 Conclusions

A7.6.1 The evidence is now overwhelming that there are identifiably distinct ‘strains’ or sub-types (genotypes or lineages) of *C. parvum* and that one such sub-type appears on current evidence to be restricted to man. There is no evidence that this results from the parasite changing within the host during the infection. Indeed, in a further outbreak in the UK where there was evidence of animal (sheep) contamination of the water, isolates from cases were of the animal genotype. The evidence of a high prevalence of genotype 1 in three waterborne outbreaks has considerable significance in relation to transmission by the water route, suggesting that sewage effluent may have been the major source of these outbreaks.

A7.6.2 A notable feature in outbreaks is commonly a marked increase in adults among early primary cases and the amplification of such outbreaks by secondary propagation, especially amongst children. This is most likely to be seen in areas of a low background level of infection prior to an outbreak, leading to low frequency of antibody (sero-prevalence). One

consequence of sero-prevalence is that those areas supplied from sources perceived to be safe may tend to experience a greater impact (attack rate) if a contamination event occurs, compared with those supplied from poorer quality sources. In addition the integrity of ground water supplies cannot be assumed to be safe from either human or animal faecal contamination (DWI 1998). Thus, the assumptions about the likely source of waterborne outbreaks can now be challenged and the earlier view of the occurrence of a human or 'urban' (ie non-zoonotic) cycle of infection has been supported. The potential usefulness of sero-epidemiology has also been underlined.

## References


Awad El Kariem, F.M., Robinson, H.A., McDonald, V., Evans, D., Dyson, D.A. (1993) Is human cryptosporidiosis a zoonotic disease? *The Lancet* **341,** 1535.

Awad-El-Kariem, F.M., Warhurst, D.C., McDonald, V. (1994) Detection and species identification of *Cryptosporidium* oocysts using a system based on PCR and endonuclease restriction. *Parasitology* **109**, 19-22.

Awad El Kariem, F.M., Robinson, H.A., Dyson, D.A., Evans, D., Wright, S., Fox, M.T., McDonald, V. (1995) Differentiation between human and animal strains of *Cryptosporidium parvum* using isoenzyme typing. *Parasitology* **110**, 129-132.

Awad El Kariem, F.M. (1996) Significant Parity of Different Phenotypic and Genotypic Markers between Human and Animal Strains of *Cryptosporidium parvum*. *J Euk Microbiol* **43**, No 5, 70S.

Awad El Kariem, F.M., Robinson, H.A., Petry, F., McDonald, V., Evans, D., Casemore, D.P. (1998) Differentiation between human and animal isolates of *Cryptosporidium parvum* using molecular and biological markers. *Parasitol Res* **84**, 297-301.

Badenoch, J. (1990) *Cryptosporidium* in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Badenoch, J. (1995) *Cryptosporidium* in water supplies. Second Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 108pp.

Bonnin, A., Fourmaux, M.N., Dubremetz, J.F., Nelson, R.G., Gobet, P., Harly, G., Buisson, M., Puygauthier-Toubas, D., Gabriel-Pospisil, F., Naciri, M., Camerlynck, P. (1996) Genotyping human and bovine isolates of *Cryptosporidium parvum* by polymerase chain reaction-restriction fragment length polymorphism analysis of a repetitive DNA sequence. *FEMS Microbiology Letters* **137:**207- 211.

Cai, J., Collins, M.D., McDonald, V., Thompson, D.E. (1992) PCR cloning and nucleotide sequence determination of the 18SrRNA genes and the internal transcribed spacer 1 of the protozoan parasites *Cryptosporidium parvum* and *Cryptosporidium muris*. *Biochim Biophys Acta* **1131**:317.

Carroway, M., Tzipori, S., Widmer, G. (1996) Identification of genetic heterogeneity in the *Cryptosporidium parvum* ribosomal repeat. *Applied and Environmental Microbiology* Vol 62;**2:**712- 716.

Casemore, D.P. (1990) Epidemiological aspects of human cryptosporidiosis. *Epidemiol Infect*; **104**:1- 28.

Casemore, D.P., Jackson, F.B. (1984) Hypothesis: cryptosporidiosis in human beings is not primarily a zoonosis. *Journal of Infection* **9:**153-156.

Casemore, D.P., Palmer, S.R., Biffin, A., (1987) Cryptosporidiosis PHLS *Microbiological Digest* **4** *1.7*.

Casemore, D.P., Wright, S.E., Coop, R. (1997) Cryptosporidiosis – human and animal epidemiology. In: Ed. R Fayer. Cryptosporidium and Cryptosporidiosis. CRC Press, Boca Raton, Pp 65-92.

Chrisp, C.E., Le Gendre, M. (1994) Similarities and differences between DNA of *Cryptosporidium parvum* and *C. wrairi* detected by the polymerase chain reaction. *Folia Parasitol* **41:**97-100.

Clark, C.G. (1998) Amoebic disease. *Trans Roy Soc Trop Med Hyg* **92**:361-4.

DWI. (1998) Assessment of water supply and associated matters in relation to the incidence of cryptosporidiosis in west Herts and north London in February and March 1997. Drinking Water Inspectorate. London, UK. 59 Pp.

Elwin, K., Casemore, D.P., Guy, E.C. (1998) A modified method for identification of type-specific polymorphisms in the TRAP-C2 gene of *Cryptosporidium parvum*. (in preparation).

Frost, F., Calderon, R.L., Muller, T.B. *et al*. (1998) A two year follow-up of antibody to *Cryptosporidium* in Jackson County, Oregon, following an outbreak of waterborne disease. *Epidemiol Infect* **121**:213-7.

Gibbons, C.L., Gazzard, B.G., Ibrahim, M.A.A., Morris-Jones, S., Ong, C.S.L., Awad El Kariem, F.M. (1998) Correlation between markers of strain variation in *Cryptosporidium parvum*: Evidence of clonality. *Parasitology International* **47:**139-147.

Jenkins, M.C. and Petersen, C. (1997) Molecular Biology of *Cryptosporidium*. In: Ed R Fayer, *Cryptosporidium and Cryptosporidiosis*. CRC Press, Boca Raton.

Kilani, R.T. and Fayer, R. (1994) Geographical variation in 18Sr RNA gene sequence of *Cryptosporidium parvum*. *Int J Parasitol* **24**:303 *et seq*.

Kilani, R.T. and Wenman, W.M. (1994) Geographical variation in 18S rRNA gene sequence of *Cryptosporidium parvum*. *Int J for Parasitol* **24**:303-6.

Laxer, M.A., Timblin, B.K., Patel, R.J. (1991). DNA sequences for the specific identification of *C.parvum* by the polymerase chain reaction. *American J Trop Med Hyg* **45**: 688-94.

Leng, X., Mosier, D.A., Oberst, R.D. (1996) Differentiation of *Cryptosporidium parvum, C. muris* and *C. baileyi* by PCR-RFLP analysis of the 18S rRNA gene. *Vet Parasitol* **62**:1 *et seq*.

McLauchlin, J., Casemore, DP., Moran, S., Patel, S. (1998) The epidemiology of cryptosporidiosis: application of experimental sub-typing and antibody detection systems to the investigation of water-borne outbreaks. *Folia Parasitologica* **45:**83-92.

Meinhardt, P.L., Casemore, D.P., Miller, K.B. (1996) Epidemiologic aspects of human cryptosporidiosis and the role of waterborne transmission. *Epidemiol Reviews* **18**:118-36.

Moran, S., Casemore, DP., Mc Lauchlin, J., Nichols, GL. (1995) Detection of Cryptosporidium Antigens on SDS-PAGE Western Blots using Enhanced Chemiluminescence. Proceedings: *Protozoan Parasites and Water*, Betts WB, Casemore DP, Fricker C, Smith H, Watkins J, Eds. Royal Society of Chemistry, Cambridge 168-171.

Morgan, U.M., Thompson, R.C.A. (1998) PCR detection of Cryptosporidium: The way forward? *Parasitology Today* Vol 4;**6:**241-245.

Morgan, U.M., Sargent, K.D., Deplazes, P., Forbes, D.A., Spano. F., Hertzberg, H., Elliot, A., Thompson, R.C.A. (1998) *Parasitology* **117:**31-37.

Morgan, U.M., Constantine, C.C., Donoghue, P., Meloni, B.P., O'Brien, P.A., Thompson, R.C.A. (1995) Molecular characterisation of *Cryptosporidium* isolates from humans and other animals using RAPD (Random Amplified Polymorphic DNA) analysis. *American Journal of Tropical Medicine and Hygiene* **52:**559-564.

Morgan, U.M., Constantine, C.C., Forbes, D.A., Thompson, R.C.A. (1997) Differentiation between human and animal isolates of *Cryptosporidium parvum* using rDNA sequencing and direct PCR analysis. *Journal of Parasitology* **83:**825-830.

Morgan, U.M., Sargent, K.D., Deplazes, P., Forbes, D.A., Spano, F., Hertzberg, H., Elliot, A., Thompson, R.C.A. (1998). Molecular characterisation of *Cryptosporidium* from various hosts. *Parasitology* **117**, 31-37.

Nichols, G.L., McLauchlin, J., Samuel, D. (1991) A technique for typing *Cryptosporidium* isolates. *J Protozool* Vol 38;**6:**237S-240S.

Nichols, G.L., Rutter, M. (1998) Review of cryptosporidiosis in England and Wales 1983-1996. *WW- 07 UKWIR Operators Collaborative Research Programme* 1997-8; Pp 41.

Nina, J., McDonald, V., Deer, R., Dyson, D.A., Chiodini, P.L., McAdam, K. (1991) Antigenic differences between oocyst isolates of *Cryptosporidium parvum. Trans R Soc Trop Med* **85** Pp315.

Nina, J.M.S., McDonald, V., Deer, R.M.A., Wright, S.E., Dyson, D.A., Chiodini, P.L., McAdam, K.P.W.J. (1992) Comparative study of the antigenic composition of oocyst isolates of *Cryptosporidium parvum* from different hosts. *Parasitology Immunology* **14:**227-232.

Nina, J.M.S., McDonald, V., Dyson, D.A., Catchpole, J., Uni, S., Iseki, M., Chiodini, P.L., McAdam, K.P.W.J. (1992) Analysis of oocyst wall and sporozoite antigens from three *Cryptosporidium* species. *Infection and Immunity* Vol 60;**4:**1509-1513.

Ortega, Y.R., Sheehy, R.R., Cama, V.A. *et al*. (1991) Restriction fragment length polymorphism analysis of *Cryptosporidium parvum* isolates of bovine and human origin. *J Protozool* **38**:540-1.

Palmer, S.R., Biffin, A and Casemore, D.P. (1987) Cryptosporidiosis. Public Health Lab Serv Microbiol Dig **4:**4-7.

Patel, S., Pedraza-Diaz, S., McLauchlin, J., Casemore, D.P. (1998) The molecular characterisation of *Cryptosporidium parvum* from two large suspected waterborne outbreaks. *Communicable Disease and Public Health* (in-press).

Pedraza-Diaz, S., McLauchlin, J., Patel, S. *et al*. (1998) The molecular characterisation of *Cryptosporidium parvum* from waterborne outbreaks. (Poster and abstract) PHLS Annual Scientific Conference University of Warwick .

Peng, M.M., Xiao, L., Freeman, A.R., Arrowood, M.J., Escalante, A.A., Weltman, A.C., Ong, C.S.L., MacKenzie, W.R., Lal, A.A., Beard, C.B. (1997) Genetic polymorphism among *Cryptosporidium parvum* isolates: Evidence of two distinct human transmission cycles. *Emerging Infectious Diseases* Vol 3;**4:**567-573.

Petersen, C., Gut, J., Leech, J.H., Nelson, R.G. (1992) Identification and initial characterisation of five *Cryptosporidium parvum* sporozoite antigen genes. *Infect Immunol* **60**:2343 *et seq*.

Pozio, E., Gomez Morales, M.A., Barbieri, FM., La Rosa, G. (1992) *Cryptosporidium:* different behaviour in calves of isolates of human origin. *Trans R Soc Trop Med Hyg* **86**, 636-638.

Ranucci, L., Spano, F., Muller, H.M. (1994) Cloning of *Cryptosporidium* oocyst wall protein COWP – 190 (Abst). *J Eukaryot Microbiol* Vol 41; **1**, 3A.

Spano, F., Putignani, L., Mc Lauchlin, J., Casemore, D.P., Crisanti, A. (1997) PCR-PFLP analysis of the *Cryptosporidium* oocyst wall protein (COWP) gene discriminates between *C. wrairi* and *C. parvum*, and between *C. parvum* isolates of human and animal origin. FEMS Microbiology Letters **150**, 209- 217.

Spano, F., Putignani, L., Naitza, S., Puri, C., Wright, S., Crisanti, A. (1998) Molecular cloning and expression analysis of a *Cryptosporidium parvum* gene encoding a new member of the thrombospondin family. *Molecular and Biochemical Parasitology* **92**, 147-162.

Vásquez, JR., Goozé, L., Kim, K., Gut, J., Petersen, C., Nelson, RG. (1996) Potential antifolate resistance determinants and genotypic variation in the bifunctional dihydrofolate reductase-thymidylate synthase gene from human and bovine isolates of *Cryptosporidium parvum. Molecular and Biochemical Parasitology* **79**, 153-165.

Webster, K.A. (1993) Molecular methods for the detection and classification of *Cryptosporidium*. *Parasitol Today* **9**, 263-266.

Widmer, G. (1998) Genetic Heterogeneity and PCR Detection of *Cryptosporidium parvum*. *Advances in Parasitology* **40**, 224-236.

# Appendix A8

## Summary of research documents on *Cryptosporidium* abstracted between 1995 and 1998 [12]

### A8.1 Introduction

A8.1.1 This Appendix contains a summary of research papers on *Cryptosporidium* and *Giardia* which have been published worldwide between the beginning of 1995 and the present date. The papers have been summarised in broad subject areas and the full references are given after the text. This summary is reproduced here for information only, the Expert Group does not necessarily endorse the work or findings.

A8.1.2 A portion of the work is centred around water treatment, in particular the optimisation of existing coagulation and flocculation practices. Novel treatments such as ultra-filtration and nano-filtration also appear. Disinfection studies have looked at combinations of disinfectants to obtain synergistic effects and the application of new technologies such as mixed oxidant species and ultra-violet light (UV). A variety of viability assays are available, from mouse infectivity, tissue culture, excystation and dye exclusion. Of all of these perhaps tissue culture may be a valuable technique for the future.

A8.1.3 Waterborne outbreaks of cryptosporidiosis have been reported from around the world associated with the consumption of contaminated drinking water or the ingestion of contaminated recreational waters. The reports indicated that outbreaks have been associated with filtered and unfiltered surface waters, groundwaters and contaminated distribution systems. A number of outbreaks have involved swimming pools, usually after faecal contamination of the water. Distributions of cysts and oocysts in surface waters, groundwaters, sewage effluents and slurries have been studied, and in particular efforts have been made to collect data other than through routine sampling programmes. It is not always possible to know when oocysts are present in high numbers, but efforts should be made to collect data when environmental conditions suggest deterioration of raw water quality. In addition a clear understanding of the likely input of parasites to a raw water catchment is helpful.

A8.1.4 Methods for detection continue to improve. Many authors highlight the need to understand recovery efficiencies and the factors that can lead to poor recoveries. Perhaps collation of this data might be useful as a ‘How to improve your analysis’ guide. Emphasis for the most part is on simplicity with reproducibility and the need for quality control. There are many variables in any detection method and it is important that a laboratory knows what these are. Better concentration techniques, the use of immunomagnetic separation (IMS) and flow cytometry have all helped

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to improve detection and reduce analytical time. Determination of viability by tissue culture is an interesting development. The viability of a single oocyst can be determined from replication of sporozoites. Detection may be by means of enzyme-linked immunosorbent assay (ELISA) or using molecular techniques, with viability assay times ranging from 24 to 48 hours. Molecular techniques are based on either DNA or RNA amplification, or fluorescent *in situ* hybridisation (FISH). These techniques also demonstrate potential viability and infectivity. The Standing Committee of Analysts and the United States Environment Protection Agency both have draft revised methods for the analysis of water samples. The rate of progress with development will probably mean that they will be out of date before too long.

A8.1.5 Studies of survival in the environment continue to demonstrate that whilst *Giardia* is a poor survivor, *Cryptosporidium* can survive well, although aged oocysts are more susceptible to environmental factors and the effects of disinfectants.

### A8.2 Sampling

A8.2.1 Walker *et al*. (1997) have described a sampling device, operating under a gravity head, for sampling streams during periods of moderated sediment-generated turbidity.

### A8.3 Detection

A8.3.1 Shepherd and Wyn-Jones (1996) compared membrane filtration (142-mm) with flocculation and cartridge filtration for recovering *Giardia* and *Cryptosporidium* from tap and river water. Flocculation gave the best recoveries of both parasites from both water types. A 1.2-µm cellulose acetate membrane was found to be best for *Cryptosporidium* and a 3.0-µm cellulose nitrate membrane best for *Giardia*. Ongerth (1997) evaluated membrane filtration, cartridge filtration and chemical flocculation for the recovery of *Giardia* and *Cryptosporidium* from seeded river, reservoir and tap water samples. Chemical flocculation and membrane filtration gave comparable results whilst cartridge filtration gave poor recoveries. Investigation of losses during sample processing are discussed by Grimason *et al*. (1997). Highest losses occur during filtration with wound yarn cartridges (up to 36% pass through) and flotation (up to 41%). Filter elution and centrifugation also create losses. A coefficient of variance of up to 40% can be observed.

A8.3.2 Clancy *et al* (1997a) have compared two types of membrane filter, a stainless steel cartridge filter, a polysulphone cartridge filter and vortex flow filtration for recovering oocysts from 10-litre volumes of seeded tap water. Fricker *et al*. (1997b) describe the use of vortex flow filtration which can achieve >90% recovery, together with Immunomagnetic Separation (IMS) which can give similar recoveries, as a rapid and reliable detection system for *Giardia* and *Cryptosporidium* in water. Vortex filtration, Nuclepore filters (ongoing) and Envirocheck filters (Gelman) were examined by Matheson *et al*. (1997) as alternatives to flocculation and flat bed membrane filtration. Recoveries for each of the membranes was >50% for both parasites and gave significantly less variation than wound fibre cartridges.

A8.3.3 Clancy *et al*. (1997b) describe trials in the USA and the UK using filtration with either vortex flow, Gelman Envirocheck, capsule

filters or Costar 5-inch filters. The filters gave >70- 96% recovery. Samples were further concentrated by IMS and could be stained with monoclonal antibody and DAPI/PI. Parton *et al*. (1997) describe an open cell reticulated foam filter which can concentrate oocysts from water with an efficiency of >95%. Samples are recovered in 5-10 minutes in volumes of 250 ml and the process requires minimum equipment and operator skill. Monro (1998) described a filter that achieved enhanced recovery of *Cryptosporidium* oocysts. It consisted of discs of foam compressed into a small cylinder. It is estimated that a recovery of at least 80% of oocysts could be achieved, and up to 2000 litres of water could be passed through the unit without clogging or breakthrough. An immuno-affinity separator, selective for *Cryptosporidium*, is being developed. Johnson *et al*. (1995) compared two methods for concentrating parasites from 300-litre seeded samples of marine waters. The Filterite negatively-charged filter was found to be simpler and faster, with reduced interference from algae, than the wound polypropylene cartridge.

A8.3.4 New approaches to detection are described by Whitmore (1997), including hydrocyclones, magnetically stabilised fluidised beds, dynamic membranes and flow cytometry using two-colour sorting. Graczyk *et. al*. (1997a) outline the importance of using an eluting fluid for recovery of residual oocysts from sample containers. Trials showed a mean of 34.7% retention in sample containers and recoveries increased from 44.1% without elution to nearly 78%. They also demonstrated (Graczyk *et al*. 1997b) that dissolution of cellulose acetate membranes with acetone and parasite concentration using ethanol does not affect stain intensity with monoclonal antibodies or infectivity in BALB/c mice.

A8.3.5 Campbell and Smith (1997) describe an inter-laboratory trial using IMS for the concentration of *Cryptosporidium* from seeded water samples. With low turbidity waters, IMS was found to be better than microscopy and flow cytometry. With increased turbidity, the efficiency of IMS diminished. Campbell *et al* (1997b) describe an improved immunomagnetic procedure for concentrating *Cryptosporidium* from high turbidity samples. Selective concentration of oocysts was achieved from waters with turbidities ranging from 60 to 6,000 NTU and for a wide range of different water types.

A8.3.6 In a recent study comparing counting methods, Klonicki *et al*. (1997) found that there were significant variations between haemocytometer, cellulose acetate and well-slide counting. Recovery after cleaning with percoll/sucrose also varied. Hoffmann *et al*. (1997) discussed the use of flow cytometry compared with direct microscopy for the detection of *Giardia* and *Cryptosporidium* in water samples. Flow cytometry was found to take less time, cost less, and could analyse a greater volume of sample. An increase in sensitivity of almost three times was observed for both parasites. A review of flow cytometry has been published by Deere *et al*. (1996). Medema (1997a) describes the detection of *Cryptosporidium* and *Giardia* in river and reservoir water using flow cytometry. Viability was demonstrated by DAPI/PI staining before sorting and could easily be assessed. A number of oocysts were observed to be DAPI and PI negative but internal contents could not be resolved.

A8.3.7 Deere *et al*, (1997) discuss the use of two antibodies specific for *Cryptosporidium* labelled with different coloured fluorochromes to minimise non-specific sorting in flow cytometry together with a specific DNA probe to identify at genus or sub-species level and demonstrate

viability. Vesey *et al*. (1997) describe the development of an IgG1 monoclonal antibody which is less sticky and reduces the amount of non-specific binding. This helps microscopy and sorting with a flow cytometer. The authors also provide some guidance on how to evaluate antibodies and improve existing reagents. A flow-cytometric assay was developed by Vesey *et al.* (1997) to evaluate *Cryptosporidium*-specific antibodies for testing water samples. There were significant differences in performance between the five different monoclonal antibodies used in the study.

A8.3.8 Rodgers *et al*. (1995) tested 54 algal species and found that 24 showed some fluorescence. Two species showed bright green fluorescence. Blocking with goat serum was found to be successful. Dowd and Pillai (1997) describe the use of propidium iodide with immunofluorescence to detect oocysts and determine viability. Three different inactivation methods were used in the study.

A8.3.9 Clancy (1997a) reports that in studies, laboratories produce highly variable data, and in some trials cannot detect seeds as high as 4,000 oocysts, and may therefore not detect oocysts in an outbreak. Health officials may misuse data particularly when advising AIDS patients about risk. Smith and Fricker (1997) discuss the variabilities that current analytical techniques have. They provide data on a range of analytical techniques, from filtration to drying on a slide, which can enable analysts to optimise some of their methodology and improve recoveries. Fricker *et al*. (1997a) discuss some factors which can produce variations in assessing the recovery efficiency of various analytical methods. Spikes should be counted in replicates of 10 and the whole recovered pellet examined. The source, age and condition of oocysts will have an effect as will the time that the oocysts have been in water. Where laboratories are comparing methods they should use the same type of water. Veal *et al*. (1997) describe rigorous quality control procedures for *Cryptosporidium* and *Giardia* analysis where one sample in ten is seeded for recovery. The information allows optimisation of different stages of analysis, comparison of recoveries from different water types and modification of results to present values corrected for recovery.

A8.3.10 Sartory (1997) was unable to find any relationship between sulphite-reducing clostridia, enterococci and the presence of *Cryptosporidium* in surface and groundwater in samples taken for routine analysis over a two year period.

### A8.4 Molecular techniques

A8.4.1 Mayer and Palmer (1996) used immunofluorescence, PCR and nested PCR to investigate the removal of *Giardia* and *Cryptosporidium* from wastewater. Immunofluorescence demonstrated a 3-log reduction for *Giardia* and a 2-log reduction for *Cryptosporidium*. PCR gave 100% correlation for *Giardia* but slightly less for *Cryptosporidium.* Stinear *et al*. (1996) describe an RT-PCR technique capable of detecting a single oocyst in reticulated, reservoir, borehole and river water. No product was obtained in oocysts fixed with formalin. *Cryptosporidium parvum* was grown in Caco-2 cells by Rochelle *et al*. (1997b). PCR was used to detect the heat-shock protein 70. A single infectious oocyst could be detected using this procedure. Additional data is given on *in vitro* infectivity with *in situ* PCR and probe hybridisation for the specific quantification of infectious *Cryptosporidium parvum* in water (Rochelle *et al.* 1997b). A comparative evaluation of different polymerase chain reaction (PCR) primers was

undertaken by Sluter *et al.* (1997), using several published protocols, with the aim of optimising PCR detection of low concentrations of *Cryptosporidium parvum* oocysts in raw and finished water.

A8.4.2 Battigelli (1997) describes an integrated ELISA/RT-PCR tissue culture procedure using human adenocarcinoma (HCT-8) cells. ELISA could assess viability down to 20 oocysts and RT-PCR could reduce this to one oocyst. A simple membrane filter procedure is described by Wiedenmann *et al*. (1997) whereby filtered oocysts are stained and counted, the membrane is dissolved in acetone and excystation of sporozoites is followed by PCR. The PCR can currently detect 10-20 viable oocysts but sensitivity is expected to increase. Recent research on DNA electrochemical sensors for a number of pathogens, including the use of an oligonucleotide probe unique to *Cryptosporidium parvum* DNA, has been reviewed by Wang *et al.* (1997).

A8.4.3 Chappell *et al*. (1997a) describe a study to assess whether DNA sequence variations were related to infectivity *in vivo*. The Harley Moon strain was compared with the TAMU isolate and current data suggest that genetic variability revealed by random amplified polymorphic DNA does not correlate with differences in oocyst infectivity in mice and humans.

A8.4.4 Vesey *et al*. (1997) used fluorescence *in situ* hybridisation (FISH) with a Cryl probe targeted to 18S ribosomal RNA to produce sporozoite fluorescence. The technique was shown to correlate with excystation and be *parvum* specific, and could be combined with immunofluorescence for detection. A rapid method for the extraction, amplification and detection of *Cryptosporidium parvum* DNA is described by Wick (1997). Strand displacement amplification uses biotin labelled primers and the amplicon is captured on microtitre plates using complimentary oligonucleotides. Detection is with horseradish-peroxidase streptavidin. Calomiris (1997) describes the use of compound D7 to enhance the recovery of oocysts from seeded turbid waters. As well as improving percoll/sucrose flotation recovery by 2 to 6 fold, compound D7 improves the sensitivity of DNA-based detection by preventing inhibition. Chung *et al*. (1997) describe an improved method using 293-mm diameter, 8.0μm pore size cellulose acetate membrane filtration followed by solution of the membrane in acetone, DNA extraction and amplification. The overall sensitivity of the method was 100 oocysts per 100 litres. Smith *et al*. (1997) used the combination of IMS to remove oocysts from inhibitory water concentrates with PCR to detect low levels (0.003-0.015 per litre) in raw waters.

A8.4.5 Deere *et al*. (1997) describe a fluorescent *in-situ* hybridisation (FISH) technique, requiring only 1 hour for labelling. The technique is species specific and can demonstrate viability by targeting 18S ribosomal RNA.

## A8.5 Viability

A8.5.1 Fricker *et al*. (1997c) describe an AWWARF and UK DWI joint funded project to assess methods for the determination of viability using CD-1 mice, excystation and vital dyes during disinfection studies. The project is being undertaken in the USA and UK during 1997 and 1998. Smith *et al*. (1997) investigated the criteria for using neonatal CD-1 mice for infectivity studies. Variability of response to infection was demonstrated and parameters such as variation in infective dose, choice of

the method for mouse analysis and stress could all influence the results obtained. Slifko *et al*. (1997) describe a foci detection method (FDM) using slide cultures of human ileocaecal adenocarcinoma (HCT-8) cells. Infection was determined by immunofluorescence. As few as 10 oocysts have been observed to set up infections.

A8.5.2 Jenkins *et al*. (1997), in comparing dye permeability using DAPI/PI with excystation and mouse infectivity, demonstrated that the dye permeability assay as an indicator of potential viability and infectivity was a useful tool. Belsoevic *et al*. (1997) described a procedure for determining viability using SYTO-9, hexidium and MPR7 1059. Viability was related to animal infectivity but not excystation. Staining was not affected by disinfectants. Black *et al*. (1996) used dye exclusion with DAPI and PI, excystation and mouse infectivity to assess viability after disinfection. Dye exclusion and excystation were found to give comparable results but overestimated viability. Mouse infectivity was considered the most reliable measure. Campbell *et al*. (1997a) found that following exposure of *Cryptosporidium* oocysts to low levels of ozone, the vital dye assay significantly over-estimated the viability of oocysts. With excystation the released sporozoites were non-motile and misshapen suggesting non-viability. Sporozoite to oocyst ratios were also found to be variable.

### A8.6 Methodology reviews

A8.6.1 Watanabe (1996) discussed the validity of current test procedures for *Cryptosporidium* and *Giardia* and the newer test methods of flow cytometry, electrorotation assay, IMS and PCR. Jakubowski *et al*. (1996) also review methods in a report by the Working Group on Waterborne Cryptosporidiosis. Viability assessment, surrogate indicators, sampling and processing techniques are reviewed. The report also presents relevant information on available antibodies. Smith and Hayes (1997) review the limit of current isolation techniques, suggest modifications and additions and examine the development of new methods and the assessment of oocyst viability. Fricker and Clancy (1998) briefly review advances that are being made in *Cryptosporidium* detection methods including new filter technologies, immunomagnetic separation, flow cytometry, nucleic acid probes and viability testing. They conclude that whilst significant advances have been made there is much still to be done before a standard protocol for all water types can be produced.

### A8.7 Raw and treated water contamination

A8.7.1 Ong *et al*. (1996) studied two adjacent catchments for *Giardia* and *Cryptosporidium*. Headwaters were not contaminated but creeks and water intakes contained *Giardia*, consistent with multiple source contamination. Both *Giardia* and *Cryptosporidium* were found in one catchment coinciding with calving activity. Wallis *et al*. (1996) found *Giardia* in 73% of raw sewage samples, 21% of raw water samples and 18.2% of treated water samples. The corresponding values for *Cryptosporidium* were 6.1%, 4.5% and 3.5%.

A8.7.2 Karanis and Seitz (1996) analysed raw, treated and backwash water from six treatment plants in Germany. Of the drinking water samples, 21% were positive for *Giardia* and 36.4% for *Cryptosporidium*, while 84% of backwash waters were positive for *Giardia* and 82% for *Cryptosporidium*. In the course of epidemiological studies of raw and

treated water in southern Germany, Wagner-Wiening *et al.* (1998) found *C. parvum* oocysts in 35 out of 89 raw water samples and in 10 out of 29 treated water samples, all of which were derived from lake waters. In all cases the concentrations were below 10 oocysts per 100 litres of water, and there was no recorded outbreak of cryptosporidiosis. Chauret *et al*. (1995) analysed raw waters for the parasites together with a range of indicators. There was some correlation between *Cryptosporidium* and enterococci, and between *Giardia* and somatic coliphages and algae. These were not general but were catchment-specific. There was no correlation between *Cryptosporidium* and *Clostridium perfringens.*

A8.7.3 LeChevallier and Norton (1995) provide results of the American Water System's monitoring for surface and potable waters. A prevalence rate of 53.9% for *Giardia* and 60.2% for *Cryptosporidium* in surface waters is reported. The validity of the test procedures is examined and the importance of the Disinfectants/Disinfection By-Products Rule not jeopardising microbiological quality is stressed. A South African study of sewage, raw water and drinking water by Kfir *et al*. (1995) revealed that the average values for *Giardia* in 10 litres of sewage, effluent, surface water and treated water were 130, 120, 30, and 2 respectively. Values for *Cryptosporidium* were approximately one quarter of these.

A8.7.4 Hancock *et al.* (1997) note that in the 12 most recent outbreaks of *Cryptosporidium*, 33% were traced to contaminated wells. In addition, in 1993-94, 40% of outbreaks with both parasites were traced to groundwater. Their most recent survey showed that 7 of 74 wells contained *Giardia* (18 cysts per 100 litres, average) and 17 contained *Cryptosporidium* (41 oocysts per 100 litres, average).

A8.7.5 Norton (1997) increased the frequency of monitoring of a raw water from monthly to weekly/fortnightly to assess whether low frequency monitoring underestimated oocysts levels. This was found to be the case in that more than 50% of the short-term evaluation samples for *Cryptosporidium* had greater levels than the monthly samples (up to 14 times higher).

A8.7.6 In a study of waters and effluents in Israel, Zuckerman *et al*. (1997) found 12 of 15 stream samples were positive for *Cryptosporidium* (0.04-1.9 oocysts per litre) and eight for *Giardia* (0.05-0.78 cysts per litre). Four out of 6 samples of a drinking water reservoir were also positive for *Cryptosporidium* (0.3-1.09 oocysts per litre) and five were positive for *Giardia* (0.135-16.2 cysts per litre). Sewage samples were positive and one sample of cowshed effluent contained 3,630 oocysts per litre.

A8.7.7 Crockett and Haas (1997) describe a systematic approach to sources of protozoa in catchments, and those conditions which could lead to increases in the concentrations of protozoa in surface waters, and to challenges to water treatment. In addition they discuss the variations in sample collection and analysis in relation to the collection of data for the Enhanced Surface Water Treatment Rule (Crockett and Haas 1995). The authors note that the consequences of poor monitoring could result in expensive and unnecessary water treatment. States *et al*. (1997) detail the monitoring of two rivers in the US for *Giardia* and *Cryptosporidium.* The parasites were detected in more than 50% of river samples and although *Giardia* was not detected in the treated water, small numbers of *Cryptosporidium* were occasionally found, and higher numbers were

found in the backwash water. Atherholt *et al* (1998) studied the effect of rainfall on the concentrations of *Cryptosporidium* oocysts and *Giardia* cysts in the Delaware river. Rainfall was a significant factor increasing concentrations of both organisms. These increases were associated with higher turbidity caused by resuspension of river and storm drain sediments and surface run-off.

A8.7.8 LeChevallier *et al*. (1997) examined the inlet and outlet of six open finished-water reservoirs. Results for parasites and indicator bacteria increased through the reservoir. Nearly all the cysts and oocysts detected were either empty or the internal structures were poorly defined. They concluded that the health risks were low. Craun *et al.* (1997) compared the outbreaks of waterborne disease in areas where the USEPA maximal contaminant level (MCL) for total coliforms had been exceeded. The violation rate was not significantly different between community systems that experienced an outbreak and those that did not. They conclude that to safeguard the public against waterborne disease, microbiological monitoring must be supplemented with periodic sanitary surveys and activities that ensure adequate water quality.

A8.7.9 Jarmey-Swan *et al* (1997) tested raw and treated waters, effluents and sludge for *Cryptosporidium* in KwaZulu-Natal. Flocculation and immunofluorescence were used for detection. Faecal samples from two hospitals were also examined. Raw water data ranged from 0-80 oocysts per litre with the highest concentrations in the summer months (rainfall). A wastewater effluent contained up to 150 oocysts per litre, a pre-thickened sludge $7.0 \times 10^5$ and a post-thickened sludge $0.25 \times 10^4$ oocysts per litre. About 10% of patients' stools were positive, and 35% of children's, with the highest incidence during summer rainfall.

## A8.8 Water treatment

A8.8.1 Daniel *et al.* (1996) describe a risk assessment made in the USA, studies of *Cryptosporidium* and *Giardia* removal at activated sludge and drinking water plants in France, and Japanese research examining the risk of *Echinococcus multilocaris* infection in water. The French study concluded that membrane filtration was needed for completely reliable removal of parasites and the Japanese study concluded that risk of infection was low. Hancock *et al.* (1996) suggest using microscopic particulate analysis (MPA) to assess water treatment plant performance as an alternative to parasite detection. This can be done by centrifugate pellet and particulate count reduction between raw and treated water samples. Powell (1996) describes a membrane-based filtration for water treatment using 0.2-µm membranes. The technique is suitable for surface waters, groundwaters and backwash water, removing colour and suspended solids as well as bacteria and parasites. Bernhardt and Clasen (1996) discuss the optimisation of water treatment regimes to prevent breakthrough of *Giardia* and *Cryptosporidium* into treated water supplies.

A8.8.2 Ongerth and Pecoraro (1995) used laboratory-based experiments to look at parasite removal from alum coagulant dosed waters using optimal and sub-optimal doses. Dosed waters were fed to a triple-media filter of anthracite coal, silica sand and garnet sand. Removal of *Giardia* ranged from 2.7 to 3.1 logs and *Cryptosporidium* from 3.05 to 3.6 logs. Halving the coagulant reduced the removal to 1.3 and 1.5 logs respectively. Jacangelo *et al.* (1995) investigated microfiltration and ultrafiltration for the removal of *Giardia*, *Cryptosporidium* and MS2

coliphage. Removal was increased by coating membranes with kaolinite. The influence on coagulation practices on the elimination of particles is reviewed by Lind (1997) in four case studies. Polyaluminium chloride out-performed competitor coagulants in terms of filter run-length, build-up of headloss, length of time to breakthrough, and particle count reduction. Dissolved air flotation with iron dosing was shown to remove 3.7 logs of *Cryptosporidium* under optimum conditions by Plummer *et al.* (1995).

A8.8.3 By addressing problems such as poor coagulant dosing control, inadequate rapid mixing processes, poor monitoring of individual filters, insufficient wastage after backwashing, unreliable turbidity measurement, inadequate operator knowledge; the use of dirty filters in part-time units, poor process monitoring, and using filter run time as the sole criterion for backwashing, Consonery *et al.*(1997) raised the acceptable performance of treatment plants in Pennsylvania from 39% to 91% over an eight-year period. Positive presumptive *Cryptosporidium* samples fell from 35% to below 5%. A post-filtration ninetieth percentile particle count below 10 per ml in the 3-18-µm size range was considered necessary to minimise breakthrough of pathogenic protozoa.

A8.8.4 Turbidity, UV absorption and dissolved organic carbon were suggested as surrogate parameters for analysis. Yates *et al*. (1997) used bench-scale and pilot- scale studies to optimise coagulant and polymer doses for particle and turbidity removal. Further pilot-scale work will include dual or tri-filtration media. Aerobic spores will be used as a surrogate for determining optimisation. *Cryptosporidium* seeding and removal will be used once conditions are optimised. Additional testing will include evaluation of pre-oxidant dosing (chlorine or ozone). Using a large filter pilot plant in Israel, Hatukai *et al.* (1997) monitored the effect of different filtration velocities, grain sizes, pre-oxidant types and flocculant doses. Approximately 1.7 and 2 log removals of *Giardia*-sized particles were achieved through filtration alone and through sedimentation and filtration, respectively. For *Cryptosporidium*, these values were 1.5 and 1.9 logs respectively. The most important parameters were pre-oxidant type, and dosing rate of alum and a cationic polymer. Chlorine dioxide was more efficient than chlorine. Ongerth and Hutton (1997) used laboratory-scale tests to determine the effectiveness of diatomaceous earth (DE) filtration in removing *Cryptosporidium* oocysts from raw water. Reductions were higher with finer, less permeable DE, and with a higher filtration rate for all DE grades.

A8.8.5 Payment (1998) suggests that the spores of *Clostridium perfringens* could be used as indicators of the presence of *Cryptosporidium* oocysts because they were an indicator of faecal pollution. Additionally, since they were relatively resistant to inactivation they were also a good indicator of the efficiency of treatment. Rice *et al.* (1996) assessed the value of aerobic spores as indicators of treatment efficiency. It was concluded that these spores were useful surrogates although being smaller than oocysts they would underestimate removal by filtration processes. The use of aerobic spores for assessing the efficiency of drinking water treatment has recently been reviewed by United Kingdom Water Industry Research Limited (UKWIR, 1998). Particle counting, turbidity and bacillus spores were compared as surrogates for *Cryptosporidium* by Fox (1997b). Jar tests and a pilot-scale plant were used to evaluate flocculation and filtration. Spiking with *Bacillus subtilis* spores and *Cryptosporidium* oocysts was also done. Scott *et al.* (1997) are studying a full-scale treatment plant to determine the removal of

*Cryptosporidium,* aerobic spore formers and particles to optimise treatment processes and to provide methods for treatment plant evaluation. Hijnen *et al.* (1997) used spores of sulphite-reducing clostridia in raw water as a surrogate for parasite removal in a water treatment plant. Flotation, filtration, ozone and GAC gave an approximate 3-log reduction.

A8.8.6 Gregory (1998) argues that the possible presence of *Cryptosporidium* means that even low levels of particles in drinking water are of concern, and that traditional turbidity measurements are insensitive in the size range of oocysts. He discusses the forms of turbidity measurements, and the alternatives which have greater sensitivity. Particle counters were evaluated by Hall and Croll (1997) as tools for monitoring and improving plant performance with regard to *Cryptosporidium* risk at a pilot-plant facility. Particle counters gave a more sensitive indication of particle breakthrough from filters compared with turbidimeters. The Diverse fine particle monitor has been described by Roth (1998). This instrument continuously monitors particles in the range 2-20 μm and if the particle concentration exceeds the norm an alarm sounds. Li *et al.*(1997) observed in field-scale systems a high linear correlation was between removals of 4-6 μm polystyrene microspheres and *Cryptosporidium parvum* oocysts. Other potential surrogates, including 4-6 μm particle counts, 1-25 μm particle counts and turbidity, proved less reliable.

A8.8.7 Frederiksen (1997) augmented conventional water treatment with wound fibre nanofiltration and monitored particle counts, turbidity, spore and plate counts over a 15-month period. Membranes remained intact despite over 50 acid washings. Further studies will include parasite monitoring. Drodz and Schwartzbrod (1997) used a pilot tangential microfiltration system (0.2 μm) for the removal of oocysts added to large volumes of river water. Removal of >4.3 logs was observed in nine trials. The viability of the oocysts in the filter concentrate was unaltered but washing the membrane with sodium hydroxide and nitric acid significantly reduced viability. The development of a new two-stage process for *Cryptosporidium* removal has been described by Bell and Pearce (1997). The oocysts were removed from the water supply by a spirally-wound backwashable depth filter and then destroyed by *in situ* vacuum steam pasteurisation of the filter element. Johnson (1998) described the Memtec membrane control strategy. Monitoring of process integrity and the identification and isolation of faulty modules was required to ensure 4 log unit (or better) removal of *Giardia* or *Cryptosporidium*.

A8.8.8 Drury and Lloyd (1997) discuss the difficulties of enumerating oocysts, the use of laboratory or pilot-plant studies for 'real-life' situations and the failure of surrogates to mimic *Cryptosporidium.* Good water treatment regimes and good liaison between water utilities, the Environment Agency and Environmental Health officials are stressed. They also describe work which shows that conventional water treatment may cause oocysts to clump, and consider survival in agricultural wastes. Oxenford *et al* (1997) have produced a report based on findings of an AWWARF sponsored research project and other international research to provide water treatment managers with 'bottom line' information on *Cryptosporidium.*

## A8.9 Disinfection

A8.9.1 Fayer *et al* (1996) used saturated gaseous atmospheres of ammonia, carbon monoxide, ethylene oxide, formaldehyde and methyl

bromide to challenge purified oocysts of *Cryptosporidium parvum* at 21-23˚C for 24 hours. Oocysts exposed to ammonia, ethylene oxide or methyl bromide were non-infective for BALB/c mice whilst formaldehyde and carbon monoxide exposed oocysts were infective. Quinn *et al* (1996) used dielectrophoresis at two frequencies to demonstrate ozone inactivation of *Cryptosporidium* oocysts. Aqueous chlorine, chlorine dioxide, sodium thiosulphate, chlorite and chlorate were assessed by Liyanage *et al* (1997a,b,c) as disinfectants against *Cryptosporidium parvum* at pH 8.0 and 22˚C. Infectivity was assessed using CD-1 mice. Only chlorine dioxide was shown to be effective. Gyurek *et al* (1997) examined chlorine and monochloramine inactivation of *Cryptosporidium parvum* oocysts at pH 6.0 and 8.0 and 22˚C. CD-1 mice were used to assess infectivity. Design graphs were produced to aid engineers to establish disinfection requirements for controlling *Cryptosporidium* in drinking water.

A8.9.2 Venczel *et al* (1997) compared an electrochemically produced oxidant solution (MIOX, LATA Inc.) and free chlorine as disinfectants against *Cryptosporidium* and *Clostridium perfringens* spores at pH 7.0 and 25˚C. Doses of 5mg.$l^{-1}$ were used with contact times of up to 24 hours. The mixed oxidant gave a 3-log inactivation of oocysts and spores in four hours whilst free chlorine had no effect on oocysts and gave a 1.4-log reduction of *Clostridium perfringens.*

A8.9.3 Studies of the relationship between the inactivation of *Cryptosporidium* in natural waters and different combinations of different disinfectants are being done by Oppenheimer *et al* (1997). Ozone, chloramine and chlorine are being tested over a wide range of temperatures. Mouse infectivity will be used to assess viability. In bench-scale experiments, Finch *et al* (1997a) found that pre-treatment of oocysts with ozone enhanced the disinfectant activity of free chlorine. A similar effect was found for pre-treatment with free chlorine followed by monochloramine. Viability was assessed by mouse infectivity. Additional data is described in Liyanage *et al* (1997b). Studies of the inactivation by sequential addition of ozone and chlorine dioxide of *Cryptosporidium parvum* in bench scale experiments are reported Liyanage *et al* (1997c). Suspensions of *C. parvum* were exposed to chlorine dioxide or to ozone followed by chlorine dioxide. Oocyst viability was determined from infectivity studies and showed that inactivation was synergistic.

A8.9.4 LeChevallier (1997) proposes to look at *Cryptosporidium* inactivation using a new system for generating pure chlorine dioxide. Two pH levels (6 and 8), two disinfectant concentrations (0.5 and 1.5mg.$l^{-1}$) and two temperatures (10 and 20˚C) will be assessed. Viability will be assessed by DAPI/PI staining, excystation and tissue culture. Pilot studies will also examine the levels of disinfection by-products. Miltner *et al* (1997), working with a pilot-scale ozone plant, found that *Bacillus subtilis* spores were the most difficult micro-organisms to inactivate followed by indigenous spores>*Cryptosporidium*>*Giardia*>*poliovirus.* Inactivation of spores was found to be temperature dependent.

A8.9.5 Campbell *et al* (1995) studied ultraviolet radiation in a novel apparatus. Oocysts were removed by filtration, exposed to ultraviolet, backflushed from the filter and the process repeated. A reduction in viability by a factor of 100 was noted. Clancy (1997b) examined pulse and medium intensity UV, plasma sparker technology (sonoluminescence), electron beam and pulsed electric field systems as alternative disinfection technologies. Viability was determined by mouse infectivity. Only UV

methods were found to inactivate oocysts. In a further study using a full scale plant treating 400 gpm, UV gave a 4.1 log reduction in oocyst viability using mouse infectivity (Clancy *et al* 1997b). Assays by DAPI/PI and excystation gave only a two log reduction.

### A8.10 Prevention of infection

A8.10.1 A Working Group on Waterborne Cryptosporidiosis (WGWC) has produced guidelines on issuing and rescinding boil-water orders (Pontius 1996). The paper recommends forming a local task force to evaluate factors such as source water quality, treatment effectiveness, distribution system integrity, finished water quality and epidemiological evidence before issuing or removing boil water advice.

### A8.11 Risk assessment

A8.11.1 Gale (1996) discusses the wide range of pathogens which may be in water and which should be modelled. Complications arise around pathogen densities in water and whether organisms are randomly distributed or clumped. The author suggests that emphasis should be shifted away from dose-response curves and towards defining exposures to pathogen doses when making risk assessment models. The major contributing factors for human infection from drinking water are discussed by Teunis *et al* (1997). They conclude that the uncertainty in the estimated removal efficiency of treatment processes is more important than other factors. The aptness of water quality models which assume that waterborne pathogens are randomly dispersed in treated water, and that therefore their presence in any one sample could be expected to be mirrored in any other from the same batch, is challenged by Gale *et al* (1997), following experimental work using aerobic spores as surrogates for *Cryptosporidium* oocysts. In samples taken from an operational works there was evidence of clustering such that assessments of risk to human health were invalidated. Le Blanq (1997) considers that risk assessment for infection from drinking water can be based on seroprevalence or epidemiological data. Her risk assessment results suggest that tap water has a minor role to play for *Giardia* but a potentially significant role for *Cryptosporidium.*

A8.11.2 Medema (1997b) assesses all the relevant data necessary to build a risk model. Factors include concentration of the parasite in the raw water, recovery efficiency of the detection method, treatment removal and daily consumption of unboiled water. He concludes that in general risk is low but there are a few instances when risk may be high and here, reliability of water treatment needs to be controlled. Miller *et al* (1997) collected data on risk factors for *Cryptosporidium* infection in New York. Of 475 cases in 1995, >80% had a compromised immune function and 69.8% were listed in the AIDS registry. The data are not case controlled.

### A8.12 Outbreaks

A8.12.1 A large number of outbreaks have now been documented. The earliest ones occurred in the early 1980s (Badenoch 1990) but outbreaks still continue and despite our improved understanding of the parasite, its distribution in the environment and its removal by water treatment, outbreaks continue. A number of surface water derived outbreaks have occurred in the United Kingdom. Two early ones are reported in Hull and in Sheffield. The cause of the latter was never documented but

environmental investigations by Chapman *et al* (1997*)* suggested surface water contamination by demonstration of the parasite in the catchment to the supply. A large outbreak in Oxford and Swindon in 1989 (Richardson *et al* 1991) focused attention on the parasite as a waterborne problem in the United Kingdom. A smaller outbreak occurred in Bradford in 1992 (Atherton *et al* 1995) in a part of the city which received treated water from a moorland reservoir. A case control study demonstrated an association between illness and consumption of tap water from the source and cryptosporidial oocysts were recovered from the treated water. During 1997 the Drinking Water Inspectorate identified 5 notifications of increases in cases of cryptosporidiosis as being associated with water supply (DETR 1998). The most serious was in north London where there were 345 confirmed cases which appeared to be related to a ground water supply (DWI 1998).

A8.12.2 Fox and Lytle (1996) report on the USEPA investigations into the Milwaukee outbreak. Factors contributing to the outbreak are discussed and recommendations for improving the operation of the treatment works are summarised. Roefer *et al* (1996) report the investigations into an outbreak of cryptosporidiosis in the HIV-infected population in Las Vegas in 1994. There were no apparent treatment deficiencies or breakdowns. AIDS patients were considered to be at greater risk of infection through drinking tap water as opposed to bottled or filtered water.

A8.12.3 Solo-Gabrielle and Neumeister (1996) review cryptosporidiosis outbreaks in the USA. Most people affected received surface water supplies and all treatment facilities were complying with federal and local regulations. Interestingly, wastewater was implicated as the source of contamination of raw or treated waters for about half the outbreaks. A case-control study of adults with HIV infection revealed that those who drank tap water were four times more likely to have cryptosporidiosis than those who drank bottled water (Goldstein *et al* 1996). Weidenmann *et al* (1996) note that so far there have been no recorded outbreaks of *Cryptosporidium* in Germany. The investigation suggested that consumption of raw milk and contact with animals were major sources of infection rather than consumption of contaminated drinking water. Fewtrell and Delahunty (1995) report that between 1987 and 1992, there were 497 laboratory confirmed cases of *Cryptosporidium* in Blackpool, Wyre and Fylde. There was no correlation with water supply but water sport participation and animal contact were risk factors.

A8.12.4 Bridgman *et al* (1995) describe an outbreak in north-west England in 1993 giving 47 cases of *Cryptosporidium*. One groundwater source drained water from a field contaminated with animal faeces where there were fissures from mining subsidence. Water analysis was negative but a case-control study showed significant association with drinking unboiled tap water. Maguire *et al* (1995) describe the investigation of an outbreak of 44 cases of *Cryptosporidium* in south London in 1991. Fifteen primary cases were supplied by one water utility. There was no association with the amount of tapwater drunk and no water quality problems had been identified by the utility. Steiner *et al* (1997) review *Cryptosporidium parvum, Giardia, Entamoeba histolytica* and *Cyclospora cayetanensis* as causes of waterborne diarrhoeal disease. Fox (1997a) has researched waterborne outbreaks in the USA and the treatment lapses which allowed oocysts into the drinking water. The data can be used to assist utilities to manage water treatment systems to minimise the risk of outbreaks.

A8.12.5 Frost (1997) plans to study seroprevalence of antibodies against two specific *Cryptosporidium* antigens. Of the two populations selected, one receives water from a heavily contaminated surface source and the other from a deep well source. Five hundred sera will be collected and tested over a five-month period. An additional study is reported using surplus sera from NHANES III involving seven cities (Frost 1997). Statistically significant differences were observed possibly due to geographical variation in endemic levels of infection and there was possibly a significant contribution from waterborne transmission. Craun *et al* (1998) reviews 35 outbreaks of cryptosporidiosis in the United States of America and the United Kingdom. They conclude that available epidemiological data is inadequate to assess endemic waterborne risks and that analytical rather than ecological epidemiological surveys must be carried out to assess these risks.. They also suggest that protective immunity is an important consideration when assessing endemic risks. This immunity may be acquired from sporadic low-level exposure to oocysts in the drinking water which do not cause an outbreak. Craun *et al* (1998*)* believe that evidence in support of protective immunity can be obtained from clinical studies in which less severe symptoms were observed in volunteers re-challenged with oocysts.

## A8.13 Infection

A8.13.1 Chappel *et al* (1997b) infected volunteers and used the immunological response to understand the host-parasite interaction. A challenge to 29 serologically negative adults with 30-1,000,000 oocysts (Iowa strain) resulted in 18 with oocysts in faeces and of these, seven had diarrhoeal symptoms. Faecal IgA was positively associated with faecal shedding. Serum response did not correlate with shedding or illness. Haas *et al* (1996) evaluated data from a study of infection and illness in human volunteers subjected to controlled exposure to oocysts of *Cryptosporidium parvum.* The apparent acceptable oocyst concentrations in potable waters is 0.003 per 100 litres. Over 73% were positive for IgM, 45% for IgA and 21% for IgG. One year on, 19 were re-challenged with 500 oocysts. Only three had evidence of oocyst shedding and seven had diarrhoea. Swabby-Cahill and Cahill (1997) used C57B1/6 mice for routine passage and stock production of *Cryptosporidium* for investigating minimum infective dose. A full review of the epidemiologic aspects of human cryptosporidiosis has been published by Meinhardt *et al* (1996).

## A8.14 Survival

A8.14.1 Heisz *et al*. (1997) suspended oocysts in river water in the dark at different temperatures. Oocysts were counted using a counting chamber and viability assessed by excystation. At higher temperatures there was a 2.5 log reduction in viability at 30 days and a 1.2 log reduction at lower temperatures in the same period. Abbeaszadegan (1997) studied the survival of *Cryptosporidium parvum* and *Giardia muris* in natural waters, sludges and sediments. *Giardia* rapidly became undetectable in river water (3 weeks) whereas *Cryptosporidium* numbers were only reduced by 0.6 logs. Aged oocysts and cysts were more susceptible to chlorine than fresh ones. *Giardia* was very susceptible to freezing. Viability was assessed by excystation.

A8.14.2 Medema *et al* (1997a) found that the time required for a 1-log reduction of *Cryptosporidium* in river water was 40-160 days at 15˚C and 100 days at 5˚C. Die-off of *Escherichia coli* and enterococci was faster

than *Cryptosporidium* but *Clostridium perfringens* die-off was slower. Johnson *et al* (1997) found that *Giardia* cysts were inactivated by high salinity, where the contents hyperplasmolyse, and by light (cysts survive for 72 hours in the dark and three hours in the light). Two month old oocysts required 13-16 days for 90% reduction whereas four-month old oocysts required three to four days. The order of survival was *Cryptosporidium*>poliovirus>*Giardia*>*Salmonella.*

### A8.15 Private water supplies

A8.15.1 Clapham (1997) examined 15 private water supplies. *Cryptosporidium* was found in 20 (14%) of samples taken and nine of the 15 supplies (60%) contained oocysts during the survey. *Giardia* was present in eight of the supplies. Enterococci and sulphite reducing clostridia were significantly correlated to *Cryptosporidium*.

A8.15.2 An outbreak of *Cryptosporidium* and *Campylobacter* was described by Duke *et al* (1996) in a private water supply with 43 people affected. *Campylobacter jejuni* was isolated from five cases, *Cryptosporidium* from four cases and both pathogens from two cases. Heavy rainfall had occurred several days before the outbreak and water samples had contained high levels of *Escherichia coli*. Three dead lambs were found in a collecting chamber from a spring supplying the supply but these were removed before they could be investigated.

### A8.16 Swimming pools

A8.16.1 The recent occurrence of cryptosporidiosis at five public pools led Kebabjian (1995) to make a number of suggestions on the management of faecal contamination in pools. Closure for up to one day is suggested to permit proper filtration and disinfection of pool water. An increase of *Cryptosporidium* incidence from 0.5-1% to 15-17% was investigated by Medema (1997c). Drinking water, distribution systems and water treatment operations were satisfactory but a case control study revealed that swimming in pools was the only risk factor. MacKenzie *et al* (1995) detail an outbreak of cryptosporidiosis involving 51 people at a hotel. Use of the swimming pool was the only significant risk factor. Unrecognised faecal accidents were suggested as the cause.

### A8.17 Wastewater

A8.17.1 Rider *et al* (1996) described a wastewater treatment system which removes 99.9% of *Giardia* cysts and *Cryptosporidium* oocysts. The filter system consisted of a dual-stage, deep bed sand filter. Stadterman *et al* (1995) found that a laboratory activated sludge plant removed 98.6% of seeded *Cryptosporidium parvum* oocysts. In a comparison of different treatment regimes, activated sludge and anaerobic digestion were found to be the most effective means of removing oocysts, the latter destroying 99.9% in 24 hours. Hirata *et al* (1997) looked at raw sewage, and primary, secondary and final effluents for *Giardia* and *Cryptosporidium*. *Cryptosporidium* was only found in one sample (28 oocysts per litre) whereas *Giardia* was found in all samples (125-4,500 cysts per litre). Conventional activated sludge reduced cysts by 2 logs. *Clostridium perfringens* was suggested as a good surrogate. Bukhari *et al* (1997) produced data to show small numbers of oocysts in both the influent and effluent samples from sewage works whereas *Giardia* cysts were detected more frequently and at higher concentrations. Oocysts were only detected

at one site in sewage sludge whereas cysts were found at all the sites examined.

A8.17.2 Madireddi *et al* (1997) constructed a pilot plant to treat a municipal secondary effluent for augmentation of a lake used as a drinking water source. Extensive treatment including ultrafiltration and nanofiltration gave 21-22 log removal of bacteriophages and 8-10 log removal of *Giardia* and *Cryptosporidium*. During a one-year study at a water reclamation facility employing biological treatment, sand filtration and chlorination, Rose *et al* (1996) found that total and faecal coliforms were reduced by >7 logs, coliphages and enteroviruses by >5 logs and *Giardia* and *Cryptosporidium* by >3 logs. The risk of infection by exposure to 100 ml of water was calculated as between $10^{-6}$ and $10^{-8}$.

## A8.18 Faecal material

A8.18.1 Bukhari and Smith (1997) discuss ways in which agricultural wastes could pollute water, and provide information on survival of oocysts in naturally contaminated materials. Bodley-Tickell *et al* (1997) found *Cryptosporidium* in almost 70% of rural surface waters tested. Numbers were found to be higher in autumn, coinciding with calving, slurry spreading and rainfall. Levels ranged from 0-16.7 oocysts per litre with a mean value of approximately 1.0. The results indicate that wildlife may have a substantial input to small rural waters.

## A8.19 Soil

A8.19.1 Mawdsley *et al* (1996) dosed soil cores in the laboratory with high numbers ($10^8$) of *Cryptosporidium* oocysts. Small numbers of oocysts were detected in the leachate from clay loam and silty loam but not from a loamy sand soil. Variations in leaching were observed with replicate cores. The majority of oocysts were found in the top 2 cm of soils.

## A8.20 Food

A8.20.1 Harp (1996) demonstrated that oocysts were killed by heating in water and milk to 71.7˚C for 5, 10 and 15 seconds. Viability was assessed by infectivity in mice. Water as a vehicle for various foodborne agents is discussed by Palumbo *et al* (1997). An overview of wastewater treatment processes is presented and approaches to reconditioning plant processing water for reuse in food processing are included. Chalmers *et al* (1997) claim the first report of *Cryptosporidium* oocysts in mussel tissue and suggest that this might be a source of food poisoning.

## A8.21 Statistical analysis

A8.21.1 Nahrstedt and Gimbel (1996, 1997) and Gimbel and Nahrstedt (1996) describe a statistical method for determining the reliability of analytical results and a strategy for the improvement of analytical methods. Medema *et al* (1997b) used a chemical pollutants model to calculate the concentration of oocysts and cysts in surface waters receiving domestic sewage effluent. Calculated figures were found to be in good agreement with actual measurements although the model underestimates the concentrations in agricultural discharges where concentrations may be significantly higher. Sakaji and Chun (1997) outline an integrated action plan for when oocyst counts in waters are

higher than historical data. Particular reference is paid to good communication systems.

A8.21.2 Stuart (1997) modelled the effect of body-contact recreation on the concentration of pathogens, including *Cryptosporidium,* at the outlet to a reservoir under construction. The effects of boating (limited activity) and full recreational activities were modelled.

### A8.22 Typing *Cryptosporidium*

A8.22.1 McLauchlin *et al* (1997) describe the use of SDS-PAGE Western-blotting analysis to sub-type *Cryptosporidium parvum.* This and a similar system can be used to recognise multiple types of the parasite and antibodies in sera. The technique has been used to investigate waterborne outbreaks.

A8.22.2 Bonnin *et al* (1996) used PCR with restriction-fragment length polymorphism (RFLP) to type 23 isolates of *Cryptosporidium parvum.* Ten calf isolates were shown to have the same profile but 13 human isolates had two patterns, one identical to the calf isolates but the second different. Spano *et al* (1997) also used PCR with RFLP to distinguish *Cryptosporidium wrayii* from *Cryptosporidium parvum* and were able to distinguish two isolates of *parvum,* one associated with animal and one with human infections. Carroway *et al* (1996) and Morgan *et al* (1995) were also able to differentiate human and animal groups, the latter using RADP. In addition, Carroway *et al* (1997*)* were able to show that there was a change in the genetic profile of *Cryptosporidium parvum* following transmission from cattle to humans.

## References


Abbaszadegan, M. (1997) Survival of *Cryptosporidium* oocysts and *Giardia* cysts in watersheds. Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Atherholt,T.B, LeChevalier, M.W., Norton, W.D. and Rosen, J.S. (1998) Effect of rainfall on *Giardia* and *Crypto*. *Journal of American Water Works Association*, **90**, No.9, 66-80.

Atherton, F., Newman, C.P.S. and Casemore, D.P. (1995) An outbreak of waterborne cryptosporidiosis associated with a public water supply in the UK. *Epidemiology and Infection* **115**, 123-131.

Badenoch J (1990) Cryptosporidium in water supplies. Report of the Group of Experts; Department of the Environment, Department of Health. London, UK. HMSO. 230pp.

Battigelli, D.A. (1997) Viability assessment of *Cryptosporidium parvum* by an integrated cell culture/RT-PCR method**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Bell, J.A. and Pearce, G.K. (1997) Filter process removes cryptosporidial oocysts from water supplies. *Water Engineering & Management* **144**, No.8, 18-19.

Belsoevic, M., Guy, R.A., Neumann, N.F. and Finch, G.R. (1997) Viability assessment of *Cryptosporidium parvum* oocysts using nucleic acid dyes. Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Bernhardt, H. and Clasen, J. (1996) Elimination of micro-organisms illustrated with reference to the treatment of reservoir water. *GWF-Wasser/Abwasser* **137**, 109-116.

Black, E.K., Finch, G.R., Taghi-Kilani, R. and Belosevic, M. (1996) Comparison of assays for *Cryptosporidium parvum* oocysts viability after chemical disinfection**.** *FEMS Microbiology Letters* **135**, 187-189.

Bodley-Tickell, A., Kitchen, S. and Sturdee, A. (1997) *Cryptosporidium parvum* in rural surface waters. *Proceedings of the Second Symposium on Health-Related Water Microbiology* University of Warwick, ed. Morris, R. and Gammie, A., 138-143.

Bonnin, A., Fourmax, M.N., Dubremetz, J.F., Nelson, R.G., Gobet, P., Harly, G., Buisson, M., Puygauthier-Toubas, D., Gabriel-Pospisil, F., Naciri, M. and Camerlynck, P. (1996) Genotyping human and bovine isolates of *Cryptosporidium parvum* by polymerase chain reaction-restriction fragment length polymorphism analysis of a repetitive DNA sequence. *FEMS Microbiology Letters* **137**, 207- 211.

Bridgman, S.A., Robertson, R.M.P., Syed, Q., Speed, N., Andrews, N. and Hunter, P.R. (1995) Outbreak of cryptosporidiosis associated with a disinfected groundwater supply. *Epidemiology and Infection* **115**, No. 3, 555-566.

Bukhari, Z. and Smith, H.V. (1997) The impact and significance of agricultural waste-derived *Cryptosporidium* sp. oocysts on contamination of water. Submitted as an abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.

Bukhari, Z., Smith, H.V., Sykes, N., Humphries, S.W., Paton, C.A., Girdwood, R.W.A. and Fricker, C.R. (1997) Occurrence of *Cryptosporidium* spp oocysts and *Giardia* spp cysts in sewage influents and effluents from treatment plants in England. *Water Science and Technology* **35**, No. 11/12, 385-390.

Calomiris, J.J. (1997) Enhanced recovery of *Cryptosporidium parvum* from turbid surface waters for detection by DNA amplification. Submitted as an abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.

Campbell, A.T., Robertson, L.J., Snowball, M.R. and Smith, H.V. (1995) Inactivation of oocysts of *Cryptosporidium parvum* by ultraviolet irradiation. *Water Research* **29**,. 2583-2586.

Campbell, A.T. and Smith, H.V. (1997) Immunomagnetic separation of *Cryptosporidium* oocysts from water samples: round robin comparison of techniques**.** *Water Science and Technology* **35**, No. 11/12, 397-402.

Campbell, A.T., Anderson, R.L., Robertson, R.L., Parker, J.F.W. and Smith, H.V. (1997a.) Viability of *Cryptosporidium* oocysts*:* assessment following disinfection with ozone. *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver. Co., 97-102.

Campbell, A.T., Gron, B. and Johnsen, S.E. (1997) Immunomagnetic separation of *Cryptosporidium* oocysts from high turbidity water samples. concentrates *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 91-96.

Carroway, M., Tzipori, S. and Widmer, G. (1996) Identification of genetic heterogeneity in the *Cryptosporidium parvum* ribosomal repeat. *Applied and Environmental Microbiology* **62**, No. 2, 712- 716.

Carroway, M., Tzipori, S. and Widmer, G. (1997) A new restriction fragment length polymorphism from *Cryptosporidium parvum* identifies genetically heterogenous parasite populations and genotypic changes following transmission from bovine to human hosts**.** *Infection and Immunity* **65,** No. 9, 3958- 3960.

Chalmers, R.M., Sturdee, A.P., Mellors, P., Nicholson, V., Lawlor, F., Kenny, F. and Timpson, P. (1997) *Cryptosporidium parvum* in environmental samples in the Sligo area, Republic of Ireland: a preliminary report. *Letters in Applied Microbiology* **25**, No.5, 380-384.

Chappel, C.L. (1997a) *Cryptosporidium parvum*: comparison of two isolates by RAPD analysis and infectivity in mice and humans. Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Chappel, C.L. (1997b) *Cryptosporidium* volunteer studies: what have we learned and where are we going? Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Chauret, C., Armstrong, N., Fisher, J., Sharma, R., Springthorpe, S. and Sattar, S. (1995) Correlating *Cryptosporidium* and *Giardia* with microbial indicators**.** *Journal of American Water Works Association* **87**, No.11, 76-84.

Chung, E., Yee, A., DeGrandis, S., Aldom, J., Chagla, A., Palmateer, G., Unger, S., Boleszczuk, M., Brodsky, P., Trevors, J.T. and Lee, H. (1997) Detection of *Cryptosporidium parvum* oocysts in municipal water samples using the polymerase chain reaction and the Digene SHARP signalTM system**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 71-78.

Clancy, J.L. (1997a) *Cryptosporidium* monitoring – what do the data mean? Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Clancy, J.L. (1997b) Innovative electrotechnologies for inactivation of C*ryptosporidium***.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Clancy, J.L., Hargy, T.M. and Schaub, S. (1997a) Improved sampling methods for the recovery of *Giardia* and *Cryptosporidium* from source and treated waters**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 79-86.

Clancy, J.L., Marshall, M.M. and Dyksen, J.E. (1997b) Innovative electrotechnologies for the inactivation of *Cryptosporidium. Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick*. ed. Morris, R. and Gammie, A., 192-199.

Clapham, D. (1997) The incidence of *Cryptosporidium* and *Giardia* in private water supplies, correlatory indicators and the value of the coliform standard in assessing water quality**.** *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick.* ed. Morris, R. and Gammie, A., 220-225.

Consonery, P.J., Greenfield, D.N. and Lee, J.J. (1997) Pennsylvania's filtration evaluation programme. *Journal of American Water Works Association* **89**, No.8, 67-77.

Craun, G.F., Berger, P.S. and Calderon, R.L. (1997) Coliform bacteria and waterborne disease outbreaks. *Journal of American Water Works Association* **89**, No.3, 96-104.

Craun, G.F., Hubbs, S.A., Frost, F., Calderon, R.L. and Via, S.H. (1998) Waterborne outbreaks of cryptosporidiosis. *Journal of American Water Works Association*, **90**, No.9, 81-91.

Crockett, C.S. and Haas, C.N. (1995) Protozoan monitoring: from the ICR to the ESWTR**.** *Journal of American Water Works Association* **87,** No. 8, 50-59.

Crockett, C.S. and Haas, C.N. (1997) Understanding protozoa in your watershed**.** *Journal of American Water Works Association* **89**, No. 9, 62-73.

Daniel, P.A., Dumoutier, N., Mandra, V., Tambo, N. and Kamei, T. (1996.) Pathogenic protozoa in raw and drinking water: occurrence and removal**.** *Water Supply* **14**, No. 3/4, 387-401.

Deere, D., Veal, D., Fricker, C. and Vesey, G. (1997) Automating analytical microbiology. *Water and Environment Management* **2,** No.1, 13.

Deere, D., Vesey, G. and Veal, D. (1996) Rapid microbial analysis using flow cytometry**.** *Australasian Biotechnology* **6**, No.3, 174-177.

Deere, D., Vesey, G., Dorsch, M., Ashbolt, N., Williams, K. and Veal, D. (1997) Optimisation of *in- situ* ribosomal RNA labelling of *Cryptosporidium parvum* in suspension**.** *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick.* ed. Morris, R. and Gammie, A., 144-156.

Dowd, S.E. and Pillai, S.D. (1997) A rapid viability assay for *Cryptosporidium* oocysts and *Giardia* cysts for use in conjunction with indirect fluorescent antibody detection. *Canadian Journal of Microbiology* **43**, No. 7, 658-662.

DETR. (1998) Drinking Water 1997 – Chief Inspector's statement. Department of the Environment, Transport and the Regions. London, UK: The Stationary Office.

Drodz, C. and Schwartzbrod, J. (1997) Removal of *Cryptosporidium* from river water by crossflow microfiltration: a pilot-scale study. *Water Science and Technology* **35**, No. 11/12, 391-396.

Drury, D.F. and Lloyd A. (1997) *Cryptosporidium* – a Regulator's viewpoint**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 369-376.

Duke, L.A., Breathnach, A.S., Jenkins, D.R., Harkis, B.A. and Codd, A.W. (1996) A mixed outbreak of *Cryptosporidium* and *Campylobacter* infection associated with a private water supply. *Epidemiology and Infection* **116**, 303-308.

DWI. (1998) Assessment of water supply and associated matters in relation to the incidence of cryptosporidiosis in west Herts and north London in February and March 1997. Drinking Water Inspectorate. London, UK.

Fayer, R., Graczyk, T.K., Cranfield, M.R. and Trout, J.M. (1996) Gaseous disinfection of *Cryptosporidium parvum* oocysts. *Applied and Environmental Microbiology* **62**, No. 10, 3908-3909.

Fewtrell, L. and Delahunty, A. (1995) The incidence of cryptosporidiosis in comparison with other gastro-intestinal illnesses in Blackpool**,** Wyre and Fylde**.** *Journal of Chartered Institution of Water and Environment Management* **9**, No. 6, 598-601.

Finch, G.R. (1997a) Inactivation of *Cryptosporidium* using single and sequential application of ozone and chlorine species**.** Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Fox, K.R. (1997a) Recent waterborne cryptosporidiosis outbreaks: what have we learned**.** Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Fox, K.R. (1997b) Monitoring surrogates for controlling *Cryptosporidium* removal**.** Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Fox, K.R. and Lytle, D.A. (1996) Milwaukee's crypto outbreak: investigation and recommendations**.** *Journal of American Water Works Association* **88**, No.9, 87-94.

Frederiksen, M.M. (1997) Comparison of membrane filtration and conventional water treatment processes for the removal of *Cryptosporidium***.** Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Fricker, C.R., Clancy, J.L. and Smith, H.V. (1997a) Factors involved in the variation of performance data collected during trials of new methods for protozoan parasites**.** abstract in *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick*. ed. Morris, R. and Gammie, A., 238.

Fricker, C.R, Jonas, A., Crabb, J., Turner, N. and Smith, H.V.(1997b) The concentration and separation of *Cryptosporidium* oocysts and *Giardia* cysts using vortex flow filtration and immunomagnetic separation. *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 1-8.

Fricker, C.R., Smith, H.V., Marshall., M.M. and Clancey, J.L. (1997c) Comparison of viability/infectivity of *Cryptosporidium* in disinfection studies. abstract in *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick*. ed. Morris, R. and Gammie, A., 240.

Fricker, C.R. and Clancy, J.L (1998) Crypto's protocol prospects. *Water Quality International* **May/June 1998**, 11-14.

Frost, F. (1997) A two-city study of *Cryptosporidium* infection Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Frost, F., Muller, T. and Caldreron, R. (1997) A survey of *Cryptosporidium* antibodies using surplus NHANES sera. Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Gale, P. (1996) Developments in microbiological risk assessment models for drinking water – a short review**.** *Journal of Applied Bacteriology* **81**, No. 4, 403-410.

Gale, P., van Dijk, P.A.H. and Stanfield, G. (1997) Drinking water treatment increases micro-organism clustering: the implication for microbiological risk assessment. *Aqua* **46**, No.3, 117-126.

Gimbel, R. and Nahrstedt, A. (1996) Predictive capability of analytical data for the occurrence of *Giardia* and *Cryptosporidium* in water**.** *GWF-Wasser/Abwasser* **137**, No.2, 101-104.

Goldstein, S.T., Juranek, D.D., Ravenholt, O., Hightower, A.W., Martin, D.G., Mesnik, J.L., Griffiths, S.D., Bryant, A.J., Reich, R.R. and Herwaldt, B.L. (1996) Cryptosporidiosis: an outbreak associated with drinking water despite state-of-the-art water treatment**.** *Annals of Internal Medicine* **124**, No. 5, 459-468.

Graczyk, T.K., Cranfield, M.R. and Fayer, R. (1997a) Recovery of waterborne oocysts of *Cryptosporidium* from water samples by the membrane-dissolution method**.** *Parasitology Research* **83,** No. 2, 121-125.

Graczyk, T.K., Fayer, R., Cranfield, M.R. and Owens, R. (1997b) *Cryptosporidium parvum* oocysts recovered from water by the membrane filter dissolution method retain their infectivity. *Journal of Parasitology* **83**, No. 1, 111-114.

Gregory, J., (1998) Turbidity and beyond. *Filtration & Separation* **35**, No.1, 63-67.

Grimason, A.M., Humphries, S., Bukhari, Z., Smith, H.V. and Fricker, C.R. (1997) Where have all the oocysts gone? Losses of *Cryptosporidium* oocysts during sample preparation**.** Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Gyurek, L.L., Finch, G.R. and Belsoevic, M. **(**1997) Modelling chlorine inactivation requirements of *Cryptosporidium parvum* oocysts. *Journal of Environmental Engineering* – ASCE **123**, No. 9, 865-875.

Haas, C.N., Crockett, C.S., Rose, J.B., Gerba, C.P. and Fazil. A.M. (1996) Assessing the risk posed by oocysts in drinking water. *Journal of American Water Works Association* **88**, No.9, 131-136.

Hall, T. and Croll, B., (1997) Particle counters as tools for managing *Cryptosporidium* risk in water treatment *Water Science & Technology* **36**, No.4, 143-149.

Hancock, C.M., Ward, J.V., Hancock, K.W., Klonicki, P.T. and Sturbaum, G.D. (1996) Assessing plant performance using MPA**.** *Journal of American Water Works Association* **88,** No.12, 24-34.

Hancock, C., Rose, J.B. and Callahan, M.(1997) The prevalence of *Cryptosporidium* and *Giardia* in U. S. groundwaters. *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 147-152.

Harp, J.A., Fayer, R., Pesch, B.A. and Jackson, G.J. (1996) Effect of pasteurisation on infectivity of *Cryptosporidium parvum* oocysts in water and milk. *Applied and Environmental Microbiology* **62,** No. 8, 2866-2868.

Hatukai, S., Ben-Tzur, Y. and Rebhun, M. (1997), Particle counts and size distribution in system design for removal of turbidity by granular deep bed filtration. *Water Science & Technology* **36**, No.4, 225- 230.

Heisz, M., Chauret, C., Chen, P., Springthorpe, S. and Sattar, S.A. (1997) In vitro survival of *Cryptosporidium* oocysts in natural waters**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 171-176.

Hijnen, W.A.M., Houtepen, F.A.P., van den Speld, W.M.H. and van der Kooij, D. (1997) Spores of sulphite reducing clostridia: a surrogate parameter for assessing the effects of water treatment processes on protozoan (oo)cysts? *1997 International Symposium on Waterborne Cryptosporidium Proceedings*, ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 115-126.

Hirata, T. and Hashimoto, A. (1997) A field survey on occurrence of *Giardia* cysts and *Cryptosporidium* oocysts in sewage treatment plants**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 183-194.

Hoffman, R.M., Standridge, J.H., Prieve, A.F., Cucunato, J.C. and Bernhardt, M. (1997) Using flow cytometry to detect protozoa**.** *Applied and Environmental Microbiology* **89**, No. 9, 104-111.

Jacangelo, J.G., Adham, S.S and Laine, J.M. (1995) Mechanism of *Cryptosporidium, Giardia* and MS2 removal by MF and UF**.** *Journal of American Water Works Association* **87,** No. 9, 107, 121.

Jakubowski, W., Boutros, S., Faber, W., Fayer, R., Ghiorse, W., Lechevallier, M., Rose, J., Schaub, S., Singh, A. and Stewart, M. (1996) Environmental methods for *Cryptosporidium***.** *Journal of American Water Works Association* **88.** No. 9, 107-121.

Jarmey-Swan, C., Bailey, I.W and Howgrave-Graham, A.R. (1997) Detection, occurrence and epidemiology of *Cryptosporidium* in KwaZulu-Natal, South Africa**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 159-170..

Jenkins, M.B., Anguish, L.J., Bowman, D.D., Walker, M.J. and Ghiorse, W.C. (1997) Assessment of a dye permeability assay for determination of inactivation rates of *Cryptosporidium parvum* oocysts**.** *Applied and Environmental Microbiology* **63**, No. 1, 3844-3850.

Johnson, D.C., Enriquez, C.E., Pepper, I.L., Davis, T.L., Gerba, C.P. and Rose, J.B. (1997) Survival of *Giardia, Cryptosporidium, poliovirus* and *Salmonella* in marine waters. *Water Science and Technology* **35**, No. 11/12, 261- 268.

Johnson, D.C., Reynolds, K.A., Gerba, C.P., Pepper, I.L. and Rose, J.B. (1995) Detection of *Giardia* and *Cryptosporidium* in marine waters. *Water Science and Technology* **31,** No. 5/6, 439-442.

Johnson, W.T. (1998) Predicting log removal performance of membrane systems using in-situ integrity testing. *Filtration & Separation* **35**, No.1, 26-29.

Karanis, P. and Seitz, H.M. (1996) Occurrence and distribution of *Giardia* and *Cryptosporidium* in raw and finished waters produced from surface waters. *GWF-Wasser/Abwasser* **137,** No.2, 94-100.

Kebabjian, R.R. (1995) Disinfection of public pools and management of faecal accidents**.** *Journal of Environmental Health* **58**, No. 1, 8-12.

Kfir, B., Hilner, C., Du Preez, M. and Bateman, B. (1995) Studies on the prevalence of *Giardia* cysts and *Cryptosporidium* oocysts in South African water**.** *Water Science and Technology* **31,** No. 5/6, 435- 438.

Klonicki, P.T., Hancock, C.M., Straub, T.M., Harris, S.I., Hancock, K.W., Alyaseri, A.N., Meyer, C.J. and Sturbaum, G.D. (1997) Crypto research: are fundamental data missing. *Applied and Environmental Microbiology* **89**, No. 9, 97-103.

Le Blanq, S.M. (1997) A framework for assessing the risks of waterborne *Cryptosporidium* and *Giardia*. Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

LeChevallier, M.W. (1997) Inactivation of *Cryptosporidium* by chlorine dioxide: balancing the risks**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

LeChevallier, M.W. and Norton, W.D. (1995) *Giardia* and *Cryptosporidium* in raw and finished water. *Journal of American Water Works Association* **87**, No. 9, 54-68.

LeChevallier, M.W., Norton, W.D. and Atherholt, T.B. (1997) Protozoa in open reservoirs**.** *Journal of American Water Works Association* **89**, No. 9, 84-96.

Li, S.Y., Goodrich, J.A., Owens, J.H., Willeke, G.E., Schaefer, F.W. and Clark, R.M. (1997) Reliability of surrogates for determining Cryptosporidium removal. *Journal of American Water Works Association* **89**, No.5, 90-99. Lind, C.B. (1997) Particle counts can benefit from changes in coagulant programs. *Water Engineering & Management* **144**, No.5, 24 and 26-28.

Liyanage, L.R.J., Finch, G.R. and Belosevic, M. (1997a) Effect of aqueous chlorine and oxychlorine compounds on *Cryptosporidium parvum* oocysts. *Environmental Science and Technology* **31**, No. 7, 1992-1994.

Liyanage, L.R.J., Finch, G.R. and Belosevic, M. (1997b) Sequential disinfection of *Cryptosporidium parvum* by ozone and chlorine dioxide *Ozone Science & Engineering* **19**, No.5, 409-423.

Liyanage, L.R.J., Finch, G.R. and Belosevic, M. (1997c) Synergistic effects of sequential exposure of *Cryptosporidium* oocysts to chemical disinfectants**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 41-52.

MacKenzie, W.R., Kazmierczak, J.J. and Davis, J.P. (1995) An outbreak of cryptosporidiosis associated with a resort swimming pool. *Epidemiology and Infection* **115**, No. 3, 545-553.

Madireddi, K., Babcock, R.W., Levine, B., Huo, T.L., Khan, E., Ye, Q.F., Neethling, J.B., Suffet, I.H. and Stenstrom M. K. (1997) Wastewater reclamation at lake Arrowhead, California-an overview**.** *Water Environment Research* **69**, No. 3, 350-362.

Maguire, H.C., Holmes, E., Hollyer, J., Strangeways, J.E.M., Foster, P., Holliman, R.E. and Stanwell-Smith, R. (1995), An outbreak of cryptosporidiosis in South London: what value the *p* value? *Epidemiology and Infection* **115**, No. 2, 279-287.

Matheson, Z., Jonas, A., Smith, H.V., Hargy, T., Clancy, J. and Fricker, C.R. (1997) Evaluation of new concentration procedures for the detection of *Cryptosporidium* and *Giardia***.** Abstract in *Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick,* ed. Morris, R. and Gammie, A., 244.

Mawdsley, J.L., Brooks, A.E. and Merry, R.J. (1996) Movement of the protozoan pathogen *Cryptosporidium parvum* through three contrasting soil types**.** *Biology and Fertility of Soils* **21**, No. 2, 30-36.

Mayer, C.L. and Palmer, C.J. (1996) Evaluation of PCR, nested PCR and fluorescence antibodies for detection of *Giardia* and *Cryptosporidium* species in wastewater**.** *Applied and Environmental Microbiology* **62**, No.6, 2081-2085.

Mclauchlin, J., Casemore, D.P., Moran, S. and Patel, S. (1998) The epidemiology of cryptosporidiosis: application of experimental sub-typing and antibody detection systems to water-borne outbreaks. *Folia Parasitologica* **45**, 83-92.

Medema, G.J. (1997a) Detection of viable *Cryptosporidium* and *Giardia* in water by flow cytometry and propidium iodide staining**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Medema, G.J. (1997b) A stochiastic method to assess the risk of *Cryptosporidium* and *Giardia* in drinking water. Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Medema, G.J. (1997c) An outbreak of cryptosporidiosis in the Netherlands: correlation with swimming pools. Submitted as an *abstract to 1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Medema, G.J., Bahar, M. and Schets, F.M. (1997a) Survival of *Cryptosporidium parvum, Escherichia coli*, faecal enterococci, and *Clostridium perfringens* in river water: influence of temperature and autochthonous microorganisms. *Water Science and Technology* **35**, No. 11/12, 249-253.

Medema, G.J. Schijven, J.F., De Nijs, A.C.M. and Elzenga, J.G. (1997b) Modelling the discharge of *Cryptosporidium* and *Giardia* by domestic sewage and their dispersion in surface water. *1997 International Symposium on Waterborne Cryptosporidium Proceedings, ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A.* American Water Works Association, Denver, Co., 177-182.

Meinhardt, P.L., Casemore, D.P. and Killer, K.B. (1996) Epidemiologic aspects of human cryptosporidiosis and the role of waterborne transmission. *Epidemiologic Reviews* **18**, No. 2, 118-136.

Miller, J. R., Ashendorf, A., Seeley, A., Mikol, Y.B., Faber Jr, W.W., Calder, J. and Layton, M. (1997) Descriptive epidemiology of cryptosporidiosis in New York City**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 317-328.

Miltner, D., Shukairy, H.M., Rice, E.W., Owens, J.H., Schaefer III, F.W. and Dahling, D.R. (1997) Comparative ozone inactivation of *Cryptosporidium* and other microorganisms**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co.,229-242.

Monro, M. (1998) Filter cracks crypto problem. *Water Bulletin* No.791, 11.

Morgan, U.M., Constantine, C.C., O'Donoghue, P., Melone, B.F., O'Brien, P.A.. and Thompson, R.C.A. (1995) Molecular characterisation of *Cryptosporidium* from humans and other animals using random amplified polymorphic DNA analysis. *American Journal of Tropical Medicine and Hygiene* **52**(b), 559-564.

Nahrstedt, A. and Gimbel, R. (1996) A statistical method for determining the reliability of analytical results in the detection of *Cryptosporidium* and *Giardia* in water. *Aqua* **45**, No3, 101-111.

Nahrstedt, A. and Gimbel, R. (1997) The meaningfullness of analytical results in the detection of *Cryptosporidium* and *Giardia* in water**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Norton, W.D. (1997) Short-term variability of *Giardia* cyst and *Cryptosporidium* oocyst concentrations in a surface water source used as for potable water**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Ong, C., Moorehead, W., Ross, A. and Isaac-Renton, J. (1996) Studies of *Giardia* spp. and *Cryptosporidium* spp. in two adjacent watersheds**.** *Applied and Environmental Microbiology* **62,** No. 8, 2798-2805.

Ongerth, J.E. (1997) Comparison of *Cryptosporidium* and *Giardia* analysis methods-lab and field studies**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Ongerth, J.E. and Hutton, P.E. (1997) DE filtration to remove *Cryptosporidium*. *Journal of American Water Works Association* **89**, No.12, 39-46.

Ongerth, J.E. and Pecoraro, J.P. (1995) Removing *Cryptosporidium* using multimedia filters**.** *Journal of American Water Works Association* **87**, No.12, 83-89.

Oppenheimer, J., Aieta, E.M., Jacangelo, I.J., Najm, I., Selby, D. and Rexing, D.(1997) Disinfection of *Cryptosporidium*: design criteria for North American water agencies. *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 275-280.

Oxenford, J.L., Frey, M.L., Logsdon, G.S. and Hancock, C. (1997) Synthesis report on *Cryptosporidium***.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Palumbo, S.A., Rajkowski, K.T. and Miller, A.J. (1997) Current approaches for reconditioning process water and its use in food manufacturing operations**.** *Trends in Food Science and Technology* **8**, No. 3, 69-74.

Parton, A., Mendez, F. and Sartory, D.P. (1997). Evaluation of a novel filter for the rapid capture and concentration of *Cryptosporidium* oocysts from drinking water. Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick. ed. Morris, R. and Gammie, A., 185-191.

Payment, P. (1998) Elimination of *Cryptosporidium* oocysts: potential indicators. *Gas, Wasser, Abwasser* **78**, No.1, 42-44.

Plummer, J.D., Edzwald, J.K. and Kelley, M.B. (1995) Removing *Cryptosporidium* by dissolved air floatation. *Journal of American Water Works Association* **87**, No. 9, 85-95.

Pontius, F.W. (1996) Guidelines for boil-water advisories**.** *Journal of American Water Works Association* **88,** No.12, 18-102.

Powell, N. (1996) Membrane microfiltration for municipal water treatment. *Membrane Technology* No. 71, 7-9.

Quinn, C.M., Archer, G.P., Betts, W.B. and O'Neill, J.G. (1996) Dose-dependent dielectrophoretic response of *Cryptosporidium* oocysts treated with ozone**.** *Letters in Applied Microbiology* **22,** No.3, 224-228.

Rice, E.W., Fox, K.R., Miltner, R.J., Lytle, D.A. and Johnson, C.H. (1996) Evaluating plant performance with endospores. Journal of the American Water Works Association **88** No 9, 122-130.

Rider, D., Suozzo, J. and Collins, C. (1996) Dual-stage filter removes protozoan cysts from effluent**.** *Water Environment and Technology* **8**, No.6, 15-16.

Richardson, A.J., Frankberg, R.A., Buck, A.C., Selkon, J.B., Colbourne, J.S., Parsons, J.W. and Mayon-W|hite, R.T. (1991) An outbreak of cryptosporidiosis in Swindon and Oxfordshire, Epidemiology and Infection **107**, 485-495

Rochelle, P.A., Ferguson, D.M., Handojo, T.J., Deleon, R., Stewart, M.H. and Wolfe, R.L. (1997b) An assay combining cell culture with reverse transcriptase PCR to detect and determine the infectivity of waterborne *Cryptosporidium parvum*. *Applied and Environmental Microbiology* **63**, No. 5, 2029-2037.

Rochelle, P.A., DeLeon, R., Ferguson, D.M., Stewart, M.H. and Wolfe, R.L. (1997a) Optimisation of an infectivity assay, combining cell culture and PCR for waterborne *Cryptosporidium parvum***.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 31-40

Rodgers, M.R., Flanigan, D.J. and Jakubowski, W. (1995) Identification of algae which interfere with the detection of *Giardia* cysts and *Cryptosporidium* oocysts and a method for alleviating this interference. *Applied and Environmental Microbiology* **61,** No. 10, 3759-3763.

Roefer, P.A., Monscvitz, J.T. and Rexing, D.J. (1996) The Las Vegas cryptosporidiosis outbreak. *Journal of American Water Works Association* **88**, No.9, 95-106.

Rose, J.B., Dickson, L.J., Farrah, S.R. and Carnahan, R.P. (1996) Removal of pathogenic and indicator microorganisms by a full-scale water reclamation facility**.** *Water Research* **30**, N0.11, 2785-2797.

Roth, S., (1998) Shining a bright new light on crypto. *Water Bulletin* No.784, 10-11.

Sakaji, R.H. and Chun, D.G. (1997) Oh-oh cysts**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 213-218

Sartory, D.P. (1997) Relationship between the occurrence of *Cryptosporidium* and bacterial indicators in surface and groundwaters. Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick. ed., R. and Gammie, A., 119-125

Scott, K.N. (1997) Evaluation of *Cryptosporidium* and surrogate removal through a full-scale treatment plant. Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California*

Shepherd, K.M. and Wyn-Jones, A.P. (1996.) Evaluation of methods for the simultaneous detection of *Cryptosporidium* and *Giardia* from water**.** *Applied and Environmental Microbiology* **62**, No. 4, 1317-1322.

Slifco, T.R., Friedman, D.E., Rose, J.B., Upton, S.J. and Jakubowski, W. (1997) Unique cultural methods to detect viable *Cryptosporidium parvum* oocysts in environmental samples**.** *Water Science and Technology* **35,** No. 11/12, 363-368.

Sluter S.D., Tzipori, S. and Widmer, G. (1997), Parameters affecting polymerase chain reaction detection of waterborne *Cryptosporidium parvum* oocysts. *Applied Microbiology and Biotechnology* **48**, No.3, 325-330.
Smith, H.V. and Fricker, C.R. (1997) How effective is the current method for the analysis of waterborne *Cryptosporidium*? Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick. ed. Morris, R. and Gammie, A., 170-184

Smith, H.V. and Hayes, C. (1997) The status of UK methods for the detection of *Cryptosporidium* spp. oocysts and *Giardia* spp. cysts in water**.** *Water Science and Technology* **35**, No. 11/12, 369-376.

Smith, H.V., Marshall, M.M., O'Grady, J., Korich, D.G., Olfers, S., Millan, D., Clancy, J., Fricker, C.R., Campbell, B., Bukhari, Z. and Rosen, J. (1997) Usefulness of the CD1 neonatal mouse model for determining the infectivity of *Cryptosporidium parvum* oocysts. Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick. ed. Morris, R. and Gammie, A., 200-206.

Smith, H.V., Webster, K.A., Parker, J.F.W., Giles, M. and Bukhari, Z. (1997) Development of an immunomagnetisable particle separation (IMS) PCR for the detection of *Cryptosporidium parvum* oocysts in water concentrates**.** Submitted as an abstract to *1997 International Symposium on Cryptosporidium, Newport Beach, California.*

Solo-Gabrielle, H. and Neumeister, S. (1996) U.S. outbreaks of cryptosporidiosis**.** *Journal of American Water Works Association* **88**, No. 9, 76-86.

Spano, F., Putignani, L., Mclauchlin, J., Casemore, D.P. And Crisanti, A. (1997) PCR-RFLP analysis of the *Cryptosporidium* oocyst wall protein (COWP) gene discriminates between *C. wrayii* and *C. parvum* and between *C. parvum* isolates of human and animal origin**.** *FEMS Microbiology Letters* **150**, 209-217.

Stadterman, K.L., Sninsky, A.M., Sykora, J.L. and Jakubowski, W. (1995) Removal and inactivation of *Cryptosporidium* oocysts by activated sludge treatment and anaerobic digestion**.** *Water Science and Technology* **31**, No. 5/6, 97-104.

States, S., Stadterman, K., Ammon, L., Vogel, P,. Baldizar, J., Wright, D., Colney, L. and Sykora, J. (1997) Protozoa in river water: sources, occurrence, and treatment. *Journal of American Water Works Association* **89**, No. 9, 74-83.

Steiner, T.S., Thielman, N.M. and Guerrant, R.L. (1997) Protozoal agents – what are the dangers for the public water supply? *Annual Review of Medicine* **48**, 329-340.

Stewart, M., Yates, M., Anderson, M., Gerba, C., DeLeaon, R. and Woldfe, R.(1997) Modelling the impact of body-contact recreation on *Cryptosporidium* levels in a drinking water reservoir. 1997 International Symposium on Waterborne *Cryptosporidium Proceedings*, ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 137-146.

Stinear, T., Matusan, A., Hines, K. and Sandery, M. (1996) Detection of a single viable *Cryptosporidium parvum* oocyst in environmental water concentrates by reverse transcriptase-PCR. *Applied and Environmental Microbiology* **62**, No. 9, 3385-3390.

Swabby-Cahill K.D. and Cahill, A.R. (1997) Infection of immune supressed adult C57BL6 mice as a biologic model to study infectivity and viability of *Cryptosporidium parvum* oocysts**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 133-136

Teunis, P.F.M., Medema, G.J., Kruidenier, L. and Havelaar, A.H. (1997) Assessment of the risk of infection by *Cryptosporidium* or *Giardia* in drinking water from a surface water source**.** *Water Research* **31**, No.6, 1333-1346.

UKWIR (1998) Use of aerobic spores for assessing the efficiency of drinking water treatment United Kingdom Water Industry Research Limited Project WW-07, 16 pp.

Veal, D., Vesey, G., Fricker, C., Ongerth, J., Le Moenic S., Champion, A., Rossington, G. and Faulkner, B. (1997) Routine cytometric detection of *Cryptosporidium* and *Giardia:* recovery rates and quality control. *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 9-20

Venczel, L.V., Arrowood, M., Hurd, M. and Sobsey, M.D. (1997) Inactivation of *Cryptosporidium parvum* oocysts and *Clostridium perfringens* spores by a mixed-oxidant disinfectant and free chlorine. *Applied and Environmental Microbiology* **63**, No. 4, 1598-1601.

Vesey, G,. Deere, D., Weir, C,. Ferrari, B., Gauci, M., Williams, K. and Veal, D. (1997) Optimising the use of monoclonal antibodies for the detection of *Cryptosporidium* in water samples. Proceedings of the Second Symposium on Health-Related Water Microbiology, University of Warwick. ed. Morris, R. and Gammie, A., 214-219.

Vesey, G., Deere, D., Dorsch, M., Veal, D., Williams, K. and Ashbolt, N (1997) Fluorescent In-situ labelling of viable *Cryptosporidium parvum* in water samples**.** 1997 International Symposium on Waterborne *Cryptosporidium* Proceedings, ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 21-30.

Vesey, G., Deere, D., Weir, J.C., Ashbolt, N., Williams, K.L. and Veal, D.A., (1997), A simple method for evaluating *Cryptosporidium* -specific antibodies used in monitoring environmental water samples. *Letters in Applied Microbiology* **25**, No.5, 316-320.

Wagner-Wiening, C., Kimmig, P. and Sacre, C., (1998) Epidemiological and methodological studies of Cryptosporidia in water samples *Gas, Wasser, Abwasser* **78**, No.1, 37-41.

Walker, M.J., Walter, M.F. and Mcdonough, K. (1997) Large-volume samples for *Cryptosporidium* and *Giardia* from small streams**.** Journal of Environmental Engineering **123**, No.1, 89-92.

Wallis, P.M., Erlandsen, S., Isaac-Renton, J.L., Olson, E., Robertson, W.J. and Van Keulen, H. (1996.) Prevalence of *Giardia* cysts and *Cryptosporidium* oocysts and characterisation of *Giardia* spp. isolated from drinking water in Canada**.** *Applied and Environmental Microbiology* **62,** No.8, 2789-2797.

Wang, J., Rivas, G., Cai, X., Palecek, E., Nielson, P., Shiraishi, H., Dontha, N., Luo, D., Parrado, C., Chicharro, M., Farias, P.A.M., Valera, F.S., Grant, D.H., Ozsoz, M. and Flair, M.N. (1997) DNA electrochemical biosensors for environmental monitoring. A review. *Analytica Chimica Acta* **347**, No. 2, 1-8.

Watanabe, M E. (1996.) New *Cryptosporidium* testing methods**.** *Environmental Science and Technology* **30**, No.12, 532A-535A.

Whitmore, T.N. (1997) Improved techniques for the recovery of *Cryptosporidium* from water. 1997 International Symposium on Waterborne *Cryptosporidium* Proceedings, ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 103-108.

Wick, J.F. (1997) Detection of *Cryptosporidium* using strand displacement amplification and a colorimetric probe capture system**.** *Abstract for the International Symposium on Cryptosporidium, Long Beach, California.*

Wiedenmann, A., Rohn, S., Hauser, A.C. and Botzenhart, K. (1996) Transmission pathways for *Cryptosporidium* in Germany**.** *GWF-Wasser/Abwasser* **137,** No.2, 105-108.

Wiedenmann, A., Steuer, S. Krüger, P. and Botzenhart, K. (1997) A simple procedure for an extract evaluation of the selective detection of viable *Cryptosporidium* oocysts by PCR**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 109-114.

Yates, R.S., Green, J.F., Liang, S., Merlo, R.P. and Deleon, R. (1997) Optimising coagulation/filtration processes for *Cryptosporidium* removal**.** *1997 International Symposium on Waterborne Cryptosporidium Proceedings,* ed. Fricker, C.R., Clancy, J.L. and Rochelle, P.A. American Water Works Association, Denver, Co., 281-290.

Zuckerman, U., Gold, D., Shelef, G. and Armon, R. (1997) The presence of *Giardia* and *Cryptosporidium* in surface waters and effluents in Israel**.** *Water Science and Technology* **35**, No. 11/12, 381-384.

# Appendix A9

## Abbreviations and Glossary

### A9.1 Abbreviations used in this Report

**ADAS** – The Agricultural Development and Advisory Service, now trading as ADAS

**AIDS** – Acquired Immune Deficiency Syndrome

**AWWARF** – American Waterworks Association Research Foundation

**BGS** – British Geological Survey

**CCDC** – Consultant in Communicable Disease Control or Consultant in Public Health Medicine

**CCTV** – Closed circuit television

**CDSC** – Communicable Disease Surveillance Centre

**COPGAP-Water** – Code of Good Agricultural Practice for the Protection of Water

**CPHM** – Consultant in Public Health Medicine

**DAF** – Dissolved air flotation

**DAPI** – 4'6-diamidino-2-phenyl indole (blue emitting fluorogen)

**DETR** – Department of the Environment, Transport and the Regions

**DWI** – Drinking Water Inspectorate

**DOE** – Department of the Environment (now DETR)

**EHO** – Environmental Health Officer

**ELISA** – enzyme-linked immunosorbent assay

**GIS** – Geographical Information System

**GP** – General Practitioner

**HIV** – Human immunodeficiency virus

**HSE** – Health and Safety Executive

**IMT** – Incident Management Team

**MAFF** – Ministry of Agriculture, Fisheries and Food

**MRA** – Microbiological risk assessment

**NTU** – Nephelometric Turbidity Unit (a measure of turbidity of a water sample)

**OCP** – Outbreak control plan

**OCT** – Outbreak Control Team

**PCR** – polymerase chain reaction (a laboratory method for amplifying DNA or RNA of an organism to aid identification)

**PEPFAA** – Prevention of Environmental Pollution from Agricultural Activity – A Code of Good Practice

**PHLS** – Public Health Laboratory Service

**SCIEH** – Scottish Centre for Infection and Environmental Health

**SOAFD –** Scottish Office Agriculture and Fisheries Department

**SPDL** – Scottish Parasite Diagnostic Laboratory

**UKWIR** – United Kingdom Water Industry Research Limited

**UV** – Ultraviolet radiation

**WRc** – formerly the Water Research Centre, now trading as WRc plc

### A9.2 Glossary

This glossary is a general aid to the reader of this Report and is not intended to be definitive.

**Abstraction –** The removal of water from surface water or groundwater, usually by pumping.

**Adit** – horizontal or near-horizontal tunnel extending outward from a well or shaft below the water table, designed to increase well productivity; passage from the surface by which a mine is entered and drained.

**Antibody** – a specific substance produced by the body's immune system in response to a particular infection.

**Aquifer** – a permeable geological formation that is capable of both storing and transmitting water in significant amounts.

**Arenaceous –** a term applied to rocks that have been derived from sand or that contain sand.

**Backwash** – cleaning water treatment filters by reversing the water flow.

**Catchment** – the area of land which drains into a watercourse.

**CD4** – a group of lymphocytes which are important in mediating the immune response; counting CD4 cells provides a guide to the potential for mounting an immune response to foreign substances and organisms.

**Clostridium** – an anaerobic spore-forming bacterium.

**Coagulant** – a substance added in water treatment to cause coagulation of particles.

**Collector well** – a particular design of well, usually constructed in unconsolidated alluvial or fluvio-glacial formations, comprising a central large-diameter shaft from which radial galleries or collectors have been driven to increase the production potential of the source.

***C. parvum*** – a species within the genus *Cryptosporidium*.

**Cryptosporidiosis** – the illness produced by infection with *Cryptosporidium.*

**Cytokines** – biologically active, soluble fractions secreted by lymphocytes and other cells.

**Dual porosity aquifer –** aquifer in which a certain proportion of the total storage capacity of the system is provided by the interstices in the rock matrix, while the fractures provide the dominant flow-path.

**Epidemiology** – a study of factors affecting health and disease in a population.

**Final water** – fully treated drinking water at the point where it leaves a water treatment works.

**Fissure flow** – the preferential flow of groundwater through dilated cracks, joints, bedding planes or other features of secondary porosity within an aquifer.

**Flocculation** – the aggregation of very fine organic or inorganic particles to form larger particles (floc) which can be moved by separation purposes, such as sedimentation, flotation or filtration, as part of the treatment of drinking water. Flocs are generally produced by the addition of chemicals.

**Flow lines –** lines indicating the direction of groundwater movement.

**Fluorescent labelled monoclonal antibody –** a monoclonal antibody with an attached fluorescent dye to aid oocyst identification when using a microscope.

**Fluvio-glacial** – pertaining to streams flowing from glaciers or to the deposits made by such streams.

**Genotype** – the genetic constitution of an organism.

***Giardia*** – a protozoal parasite capable of infecting man and causing diarrhoea.

**Groundwater** – naturally occurring sub-surface water in the saturated zone of a rock.

**Groundwater vulnerability** – the tendency or likelihood for contaminants to reach a specified position in the groundwater system after introduction at some location above the uppermost aquifer.

**Hazard** – a property or situation that in particular circumstances could lead to harm.

**Health authority** – an authority established within the National Health Service to provide or secure the provision of health services to its area on behalf of the Secretary of State for Health.

**High transmissivity** – capable of transmitting a large amount of water.

**Hydraulic gradient –** the prevailing inclination of the water table or piezometric surface which provides the driving force to transmit groundwater through an aquifer.

**IFN-gamma** – a soluble factor which activates certain white cells to increase their bactericidal activity.

**ILN-1** – interleutenin 1, important in the immune process.

**Immunocompromised** – individuals with an impaired or inefficient immune response.

**Immunoglobulins** – proteins of importance to the mounting of an immune response.

**Intergranular flow –** flow occurring between the grains of a rock.

**Intrinsic aquifer vulnerability** – groundwater vulnerability determined without reference to the attributes and behaviour of particular contaminants.

**Karst (Karstic)** – an area of limestone or other highly soluble carbonate rock, in which the landforms are of dominantly solutional origin and in which the drainage is underground in solutionally enlarged fractures and conduits.

**Losing reaches of rivers –** locations in a watercourse where surface water is percolating through the bed of the watercourse into the underlying aquifer.

**Monoclonal antibody** – an antibody produced in a laboratory which recognises one specific part of a specific micro-organism.

**Oocyst** – the environmentally resistant transmissable form of *Cryptosporidium* excreted in the faeces of an infected person or animal.

**Outbreak –** two or more linked cases of disease.

**Pathogen –** a micro-organism capable of causing disease

**Phenotype** – the sum of the observable characteristics of an organism.

**Porosity** – the ratio of the volume of the interstices to the total volume of a rock, expressed as a fraction or a percentage. Effective porosity includes only the interconnected pore spaces available for groundwater transmission.

**Private water supply** – any supplies of water provided otherwise than by a statutorily appointed water utility.

**Recycling** – the return of water which cannot enter the supply system, for example the initial filtrate after backwashing, to the treatment plant inlet.

**Risk** – a combination of the probability, or frequency, of occurrence of a defined hazard and the magnitude of the consequences of the occurrence. **Risk estimation** is concerned with the outcome or consequences of an intention, taking account of the probability of occurrence; **risk evaluation** is concerned with determining the significance of the estimated risks for those affected, it therefore includes the element of risk perception; **risk perception** is the overall view of risk held by a person or group and includes both feeling and judgement; **risk assessment** consists of risk estimation and risk evaluation.

**Sinkhole** – the point at which a surface stream sinks underground.

**Slurry** – animal wastes in a semi-liquid form.

**Soil leaching potential** – a composite measure of the ability of a soil to attenuate a diffuse source pollutant.

**Solution feature** – closed depressions a few metres to a few hundred metres in diameter and depth formed by solution action in soluble rocks, notably limestones.

**Source protection zones** – a series of concentric zones around an abstraction within which special policies apply to activities which might affect groundwater. The outermost zone covers the complete catchment area of the source, which is also called the well capture zone.

**Specific capacity** – the yield of a well per unit of draw down.

**Sporozoite** – the motile stage of *Cryptosporidium* which is released after excystation.

**Surveillance** – the process of monitoring the number of cases of disease in the community.

**Water supply zone** – A discrete area in the community, of not more than 50,000 population, generally supplied from a single source of water, for example water treatment works or service reservoir.

**Zoonosis** – a disease transmitted naturally from animals to man.