

An outbreak of waterborne cryptosporidiosis caused by post-treatment contamination

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SUMMARY

An outbreak of waterborne cryptosporidiosis affecting 27 persons, diagnosed stool positive, occurred in Ayrshire in April 1988. Twenty-one of the 27 confirmed cases required some form of fluid replacement therapy. Local general practitioners indicated a two- to fivefold increase in diarrhoeal disease during the outbreak, and following enquiries made by Environmental Health Officers it became apparent that many hundreds of people had suffered a diarrhoeal illness at that time. *Cryptosporidium* spp. oocysts were detected in the treated chlorinated water supply system, in the absence of faecal bacterial indicators. Oocyst contamination of a break-pressure tank containing final water for distribution was the cause of this waterborne outbreak. An irregular seepage of oocyst-containing water, which increased during heavy rains, was the cause of the break-pressure tank contamination, rather than a failure of the water-treatment processes. The waterborne route should be considered when clusters of cryptosporidiosis associated with potable water occur. Waterborne cryptosporidiosis can occur in the absence of other faecal indicators of contamination.

INTRODUCTION

Cryptosporidium spp. are coccidian parasites which have been recognized as a cause of diarrhoeal disease in man since 1976 (1, 2). They are widely distributed in nature and can infect numerous animal hosts. Fayer & Ungar (3) in a review on *Cryptosporidium* spp. found that this protozoan parasite was responsible for 0.12-23% of human diarrhoeal cases worldwide. Infection is spread by the faecal-oral route. Zoonotic transmission from infected calves to humans, and person-to-person transmission have also been described (4, 5). Recently, crypto-

sporidiosis has been recognized as a waterborne disease in the United States of America. D'Antonio and colleagues (6) reported a waterborne outbreak following sewage contamination of a chlorinated well, and Hayes and colleagues (7) described an outbreak in 1987 affecting an estimated 13000 people exposed to a sand-filtered chlorinated public water supply. Evaluation of the water treatment plant showed that it was operating within regulations but that suboptimal flocculation and filtration may have allowed the passage of the organism.

Here we report the first recognised outbreak of cryptosporidiosis associated with a fully treated UK public water supply.

THE OUTBREAK

The towns of Saltcoats and Stevenston lie adjacent to each other on the coast of Ayrshire, and their approximate populations are 13000 and 11000 respectively (Fig. 1).

On 5 April 1988, the Community Medicine Department of the Ayrshire and Arran Health Board was informed that *Cryptosporidium* spp. had been detected in faecal samples from five patients within a few days of each other. Three of these patients resided in Saltcoats, one of the other two was employed in Saltcoats and the other visited a relative regularly in that town. By 11 April, 19 cases had been recorded in Saltcoats and adjacent towns, and by the end of April this number had increased to 27 (Fig. 2). The number of cases in each town appears in Table 1.

INVESTIGATION OF THE OUTBREAK

Methods

Epidemiological

The outbreak was investigated on the lines laid down in the Scottish Home and Health Department memorandum *The Investigation and Control of Food Poisoning in Scotland* and used the standard investigation form recommended in that memorandum (8).

Environmental

Normal routine operating procedures were carried out by Strathclyde Regional Council Water Department before the outbreak as laid down by Scottish Development Department Water Supply guidelines (SDD Memorandum No. 13/1979).

Extensive examination of treated water transmission pipelines and storage tanks was initiated during the outbreak. The possibility of sewage contamination of the water supply was investigated in conjunction with the Sewerage Department of the Strathclyde Regional Council.

Farming activities in the raw water reservoir catchments were noted. Rainfall data for the period preceding the outbreak were collected.

Microbiological

Direct stool smears were examined routinely for the presence of *Cryptosporidium* spp. oocysts using the modified Ziehl-Neelsen staining method (9). Wet preparations for the detection of ova, cysts and parasites were not made routinely.

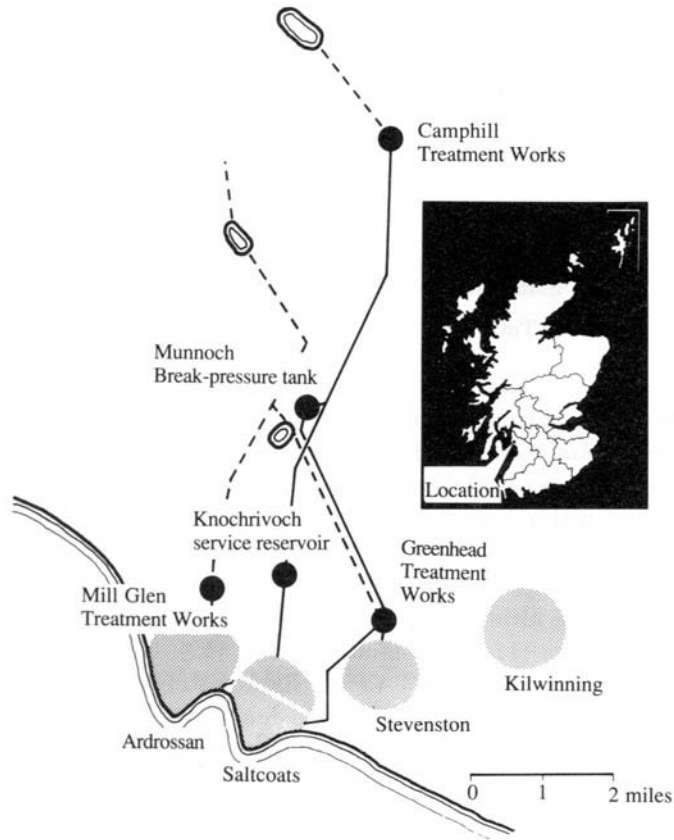


Fig. 1. Schematic diagram of the Saltcoats/Stevenston water supply system with adjacent towns in South Ayrshire. ----, Raw water; —, treated water.

Irvine, Kilwinning, West Kilbride



Ardrossan



Saltcoats, Stevenston



24 25 26 27 28 29 30 31 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23
 March April

Fig. 2. Date of onset of symptoms. ■, 'Confirmed' case preceded by unconfirmed case of similar illness in same household. □, 'Laboratory confirmed case'. ▨, 'Unconfirmed' case followed by confirmed case in same household.

Table 1. *Numbers of cases involved in the waterborne outbreak according to their place of residence*

Town	No. of cases
West Kilbride	1
Kilwinning	2
Irvine	4
Stevenston	5
Ardrossan	7
Saltcoats	8
Total	27

Water samples from various points in the system were tested for the presence of coliforms, *Escherichia coli*, faecal streptococci and bacterial plate counts and enumerated by procedures laid down in Report 71 (10).

Raw and treated water samples were examined for the presence of *Cryptosporidium* spp. oocysts by passing up to 500 l of water through a 1 µm polypropylene cartridge filter (Micro-Wynd 11 DPPPY, AMF/CUNO Corp.). Sediment trapped in the filter was eluted and processed according to the method of Musial and colleagues (11) with modifications (12). The shredded filter was washed in 0.1% Tween 80 (BDH) in distilled water in a large beaker and mechanically agitated for 10 min in order to facilitate the removal of particles trapped within the filter. The detergent solution was expressed from the shredded filter and was allowed to settle overnight, together with the backflush sediment. The settled sediment was subjected to sucrose density flotation in sterile 50 ml conical centrifuge tubes (Corning, UK). The settled sediment was underlayered with a 1.18 sp. gr. sucrose solution, centrifuged (1000 g, 10 min), and the material trapped in the interface was washed in distilled water to remove the sucrose, centrifuged and examined by microscopy with a variety of stains to detect *Cryptosporidium* spp. oocysts. Modified Ziehl-Neelsen (9), Wright-Giemsa (Diff-Quik, Travenol, Compton, UK), auramine-phenol, (Lempert) (9) safranin methylene blue (13) stains and a fluorescein-labelled monoclonal antibody (Northumbria Biologicals Ltd) (14) were used to detect the presence of *Cryptosporidium* spp. oocysts in the water samples (15).

Waterworks sludges, works filter backwashes and soil and grass were also analysed for the presence of *Cryptosporidium* spp. oocysts, using modifications of the above method to facilitate the initial suspension of particulate material. The presence of oocysts was recorded when at least three of the staining procedures outlined above were positive.

RESULTS

Epidemiological

In three households a confirmed case of cryptosporidiosis had been preceded by an unconfirmed case of similar illness (24 March, unconfirmed, 27 March, confirmed; 27 March, unconfirmed, 3 April, confirmed; and 28 March, unconfirmed, 4 April, confirmed), and in one household a confirmed case had been preceded by two successive unconfirmed cases (29 March and 7 April, unconfirmed,

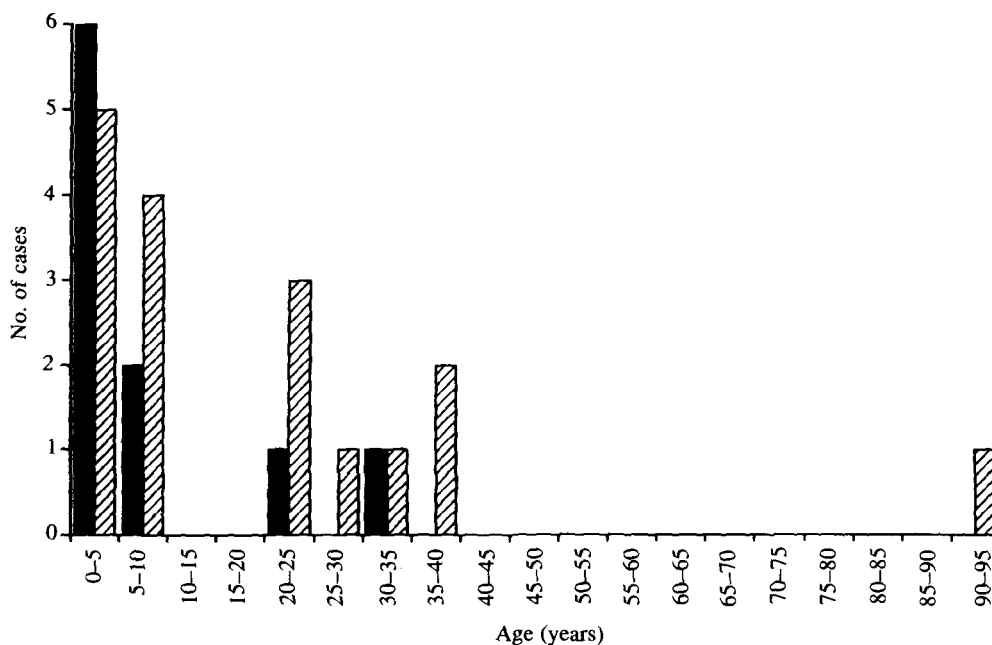


Fig. 3. Sex and age distribution of cases. ■, Male; ▨, Female.

14 April, confirmed) (Fig. 2). In three households a confirmed case was followed by an unconfirmed case (28 March, confirmed, 3 April, later unconfirmed; 3 April, confirmed, 11 April, later unconfirmed; and 18 April, confirmed, 22 April, later unconfirmed). In none of the later unconfirmed cases was a faecal sample analysed. In two of these households, the diagnosis in the index case was not made until several days after the onset of the symptoms and, by the time the index case was reported and investigated, the second patient had already recovered from a brief illness of only 1 or 2 days duration. In the third household, the second patient was so severely ill as to require admission to hospital, but by some unexplained oversight, the investigations performed did not include the examination of a faecal sample.

On no occasion during this outbreak were two confirmed cases recorded in the same household. The interval between successive cases in the same household varied from 3 to 9 days but, was most usually 7 days. Telephone enquiries to local general practitioners indicated that during the period of the outbreak, the incidence of patients presenting with diarrhoeal illness was between two and five times the normal level.

Seventeen of the 27 confirmed cases were female. Seventeen were children aged from 4 months to 8 years and 10 were adults over 20 years (Fig. 3). Twelve were admitted to hospital, 11 of whom required intravenous fluid replacement, mainly because of prolonged or recurrent vomiting, while 10 of the 15 treated at home required oral rehydration. In no instance was the illness life-threatening.

No common foodstuff or premises seemed to be implicated, however most of the affected individuals had some connection with Saltcoats or Stevenston. When the residences of the affected cases were mapped it became apparent that all the

households concerned in Stevenston and Saltcoats were served by potable water supplied from the Greenhead Treatment Works at Stevenston. The cases in Ardrossan had connections with lower Saltcoats as did other cases in Irvine and West Kilbride.

At the time of the outbreak, approximately 20000 people from Stevenston and the lower part of Saltcoats received treated water from the Camphill Treatment Works through the storage tanks at the Greenhead works in Stevenston (Fig. 1). The treated supply to the Greenhead works was routed through a break-pressure tank situated beside the Munnoch reservoir, from which tank treated water was piped into tank 7B at Greenhead. As the Camphill water was already treated it received no further treatment at Greenhead. Around 4000 people in the upper part of Saltcoats received treated Camphill water via the Knockrivoch storage tank which drew its supply from a point above the break-pressure tank at Munnoch reservoir. In addition, Munnoch reservoir provided a separate raw water supply to the Greenhead works which received full treatment and was distributed from storage tank 4.

Environmental

On 5 April the local area office of the Water Department was advised that a routine distribution water sample taken by Cunninghame District Council in Saltcoats on 22 March and the resample (29 March), failed routine bacteriological examination (10 *E. coli*, 31 coliforms; and 0 *E. coli*, 4 coliforms, per 100 ml respectively). Tank 7B at Greenhead, fed from the break-pressure tank at Munnoch, distributed the water in which the bacteriological failure occurred. Following notification of the possible involvement of the water supply, on 6 April booster chlorination procedures were introduced at the Greenhead Treatment Works and extensive routine sampling commenced. In addition, an extensive examination of the Camphill and Greenhead Treatment Works, all treated water transmission pipelines and storage tanks in the area was initiated. Numerous water samples were obtained throughout the supply system for microbiological analyses.

Microbiological

Bacteriological findings. On the week commencing 3 April all routine samples from the treatment works distribution pipelines and tanks were negative for *E. coli* and total coliforms. No failures were found until 14 April when 2 faecal streptococci per 100 ml were reported in a sample from tank no. 6 at Greenhead Treatment Works and 3 faecal streptococci per 100 ml in an ex-works sample. No coliforms or *E. coli* were found in either of the above samples. On 19 April six distribution samples in the Saltcoats and Stevenston area were negative for faecal bacterial indicators. A sample from Greenhead no. 7B service tank contained 5 *E. coli*, 8 coliforms and 2 faecal streptococci per 100 ml. Tank nos. 4 and 6 were negative. The failure in tank no. 7B suggested possible ingress within the transmission pipeline from Camphill Treatment Works. A re-inspection of all air valves and the break-pressure tank within the transmission pipeline was carried out on 20 April and samples were again taken throughout the total system.

A sample from the break-pressure tank was found to be unsatisfactory with 66

E. coli, 66 coliforms and 18 faecal streptococci per 100 ml. Samples taken from the inlet and outlet of tank no. 7B were also found to be unsatisfactory, with a much lower level of contamination. Tank no. 6 was found to be satisfactory.

Subsequent re-inspection of the break-pressure tank revealed an old 52 cm fire-clay pipe discharging into it with the pipe collecting run-off from the surrounding catchment area. This inspection had been preceded by heavy rainfall. Records showed, incorrectly, that the pipe had been sealed off many years previously. A sample of water from the pipe showed evidence of gross faecal contamination (403 coliforms, 244 *E. coli* and 58 faecal streptococci per 100 ml) as did water from a neighbouring stream (152 coliforms, 107 *E. coli* and 34 faecal streptococci per 100 ml).

Parasitological findings. A total of 27 samples including 14 final waters, 6 raw waters, 2 backflush waters, 4 sludges and 1 soil and grass were submitted for examination for the presence of *Cryptosporidium* spp. oocysts (Table 2). Of these, 6 final waters, 5 raw waters, 2 backflush waters, 2 sludge and the soil and grass were examined. Two final waters, 1 raw waters, 1 backflush water, 2 sludges and the soil and grass samples were positive for oocysts, and the numbers of oocysts detected ranged from 0.13 to 1000 per litre for liquid samples, and from 3 to 32 per g for solid samples (Table 2).

On 7 April selected samples of raw and final waters, sludge and treatment works filter backwash samples were submitted for oocyst analyses. No oocysts were detected in any of the 20 l samples submitted, and additional 500 l samples were requested. On 12 April samples were taken from Camphill and Greenhead works, from raw water, backwash water, pressed sludge cake and water entering and leaving tank 7B at Greenhead works. The presence of *Cryptosporidium* spp. oocysts was reported in the sludge samples from both Camphill and Greenhead works and in the water leaving tank 7B taken at Ardeer Golf Club (Table 2). Further samples taken on 15 and 19 April from sites within Greenhead works and from the distribution system were negative for oocysts. Samples from the break-pressure tank, the neighbouring stream and soil and grass collected from the immediate vicinity of the fire clay pipe were all positive for oocysts.

Scouring among calves on farms in the vicinity of Greenhead and Camphill had been reported to the Veterinary Investigation Centre at the West of Scotland Agricultural College, Ayrshire. Further questioning of local farmers revealed that muck spreading and the spraying of cattle slurry had occurred in the vicinity of the fire clay pipe draining into the break-pressure tank prior to the outbreak.

The record of rainfall in the water supply catchment areas feeding the Greenhead Treatment Works for the months of February, March and April 1988 showed that the latter part of February was very dry with little or no rainfall. However, three very wet days (19, 22 and 26 of March respectively), were recorded for Irvine at the weather station, Department of Environment Health, Cunninghame District Council (Fig. 4). The first week of April (with the exception of 1 April) was dry.

Table 2. Analysis of water related samples for *Cryptosporidium spp. oocysts*

Date (1988)	Volume of water (l) filtered		Source	Sampling point	Raw final	No. of oocysts detected
	Taken	Received				
April	7	8	Camphill	Ex works	Raw	0
	7	8	Camphill	Ardeer Golf Club	Final	0
	12	13	Camphill	Filter backwash water	Raw	300/l
	12	13	Camphill	Pressed sludge cake	—	3/g
	12	13	Camphill	Sludge	—	1000/l
	13	14	Camphill	Ex works	Raw	0
	13	14	Camphill	At Greenhead	Final	
	13	14	Camphill	Ardeer Golf Club	Final	
	14	15	Camphill	No. 4 tank	Final	4.8/l
	15	18	Munnoch	Millglen	Raw	
	15	18	Millglen	Millglen	Final	
	15	18	Millglen	Munnoch Reservoir	Raw	
	15	18	Camphill	Ex works at Greenhead	Final	
	15	18	Camphill	No. 5 tank (NIV) at Greenhead	Final	
	15	18	Camphill	No. 4 tank at Greenhead	Final	
	15	18	Camphill	No. 6 tank	Final	
	19	20	Camphill	Cuthbertson's Dairy factory at Saltcoats	Final	0
	19	20	Greenhead	Harvies Sports Centre Stevenstons Road	Final	0
19	20	Camphill	Ardeer Golf Club	Final	0	
21	22	Camphill	BPT	Final	0.04/l	
21	22	Munnoch Stream	Adjacent to BPT	Raw	0.13/l	
21	22	Munnoch Stream	Soil & grass	—	32/g	
21	22	Greenhead	Sludge tank 5	—		
21	22	Greenhead	Sludge tank 7	—		
May	10	11	Greenhead	Greenhead inlet	Raw	0
	10	11	Greenhead	Ex works	Final	0
	10	11	Greenhead	Filter backflush	Raw	0

N/A. Not applicable; BPT, Break Pressure Tank (Munnoch).

* Sampling is time consuming and laboratories may be unable to test all samples submitted.

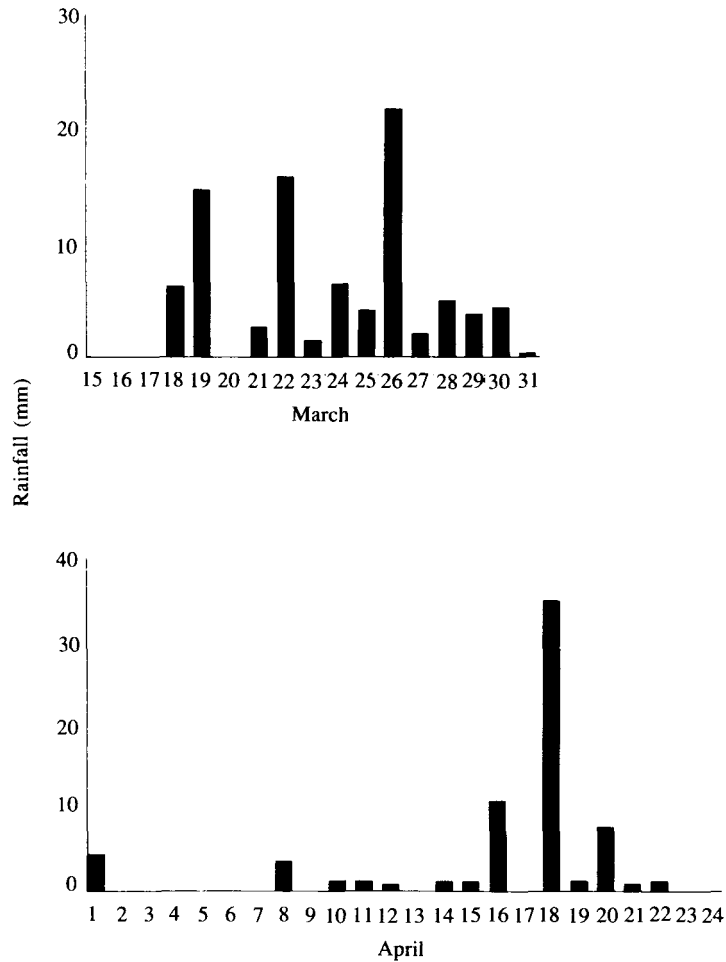


Fig. 4. Rainfall data for March and April taken at Irvine, Ayrshire.

CONTROL MEASURES

On 6 April emergency chlorination procedures were introduced at Greenhead Treatment Works, and were maintained throughout the period. The fire clay pipe draining into the break-pressure tank was sealed off with a concrete plug and the pipe broken out. As a consequence of the microbiological findings, the Camphill Treatment Works supply through the storage tanks at Greenhead Treatment Works was isolated on the afternoon of 21 April and replaced with the Bradan supply. All storage tanks containing Camphill water were isolated and were subsequently drained, cleaned and disinfected. Mains flushing was also extensively carried out in Saltcoats and Stevenston and emergency booster chlorination at the Greenhead Treatment Works was maintained throughout the period of the outbreak.

All samples taken since 21 April 1988 have been found to be satisfactory in respect of routine microbiological examination and no further cases of waterborne cryptosporidiosis have been reported in the Saltcoats and Stevenston area.

DISCUSSION

Here we report what we believe to be the first UK waterborne outbreak of cryptosporidiosis where oocysts were detected in finished water supplying the affected area. This outbreak consisted of 27 confirmed cases who were served by, or drank water from a contaminated public potable water supply. The waterborne nature of this outbreak was elucidated through the close co-operation of the many disciplines involved and by using the standard questionnaire for the investigation and control of food poisoning in Scotland (8). Although *Cryptosporidium* spp. had not been considered as a waterborne zoonosis at that time, this document proved useful in directing effort into defining the waterborne nature of cryptosporidiosis.

Although only 27 confirmed cases were identified, local practitioners reported a two to fivefold increase in diarrhoeal disease during the outbreak and the enquiries made by local Environmental Health Officers indicated that many hundreds of people had suffered a diarrhoeal illness at that time. In the two previously recorded waterborne outbreaks, 117 and 13000 cases respectively were affected (6, 7). In the second outbreak, the estimated figure of 13000 was arrived at following a telephone questionnaire of people at risk and was based on the assumption that all water-attributable illness at the time of the outbreak was due to *Cryptosporidium*. Based, retrospectively, on the date of onset of symptoms of the cases supplied by the affected water supply, and the decline in cases to normal levels, the outbreak was deemed to have lasted 31 days (24 March to 23 April) (Fig. 2), with the majority of cases occurring in the first 14 days. It is generally accepted that the incubation period for human cryptosporidiosis is about 7 days, however it is difficult to define the duration of the waterborne outbreak as secondary faecal-oral transmission within households may have occurred.

The symptoms of cryptosporidiosis can be protracted and severe in some patients and in the Ayrshire incident 21 of the 27 confirmed cases required some form of fluid replacement therapy. Hayes and colleagues (7) state that 13% of their patients with laboratory-confirmed cryptosporidiosis did not have diarrhoea as defined by three or more loose stools per day. Presumably a similar spectrum of disease occurred in Ayrshire, again indicating a far larger number of affected people than the 27 confirmed cases imply.

A higher proportion of females than males (1.7:1) was affected (Fig. 3), but the reasons for this are unknown. After controlling for age and water consumption, Hayes and colleagues (7) reported a higher attack rate in females.

Water intake levels are difficult to quantify exactly. The infective dose of oocysts also is important in the initiation of the disease. This has not been well documented for *Cryptosporidium* spp. Ernest and colleagues (16) reported a 22% infection rate in mice given 100 oocysts *per os*, while Miller and colleagues (17) reported that as few as 10 oocysts could initiate infection in primates. From Table 2 it can be deduced that 10 oocysts would be contained in 2 l of contaminated finished water sampled at Ardeer Golf Club, whereas 10 oocysts would be contained in over 200 l of the contaminated sample from the break-pressure tank. Whether the numbers of contaminating oocysts were higher prior to sampling is not known as both these positive samples were taken towards the end of the outbreak. Current (18) suggested that the virulence of different isolates may vary.

Added to this is the capacity of the host to counteract the infection immunologically. A bias in the age distribution of cases reflecting the above phenomenon in the young and old would be expected in an outbreak. In the Ayrshire outbreak 22.2% of confirmed cases were either below 2 years or above 90 years of age (Fig. 3).

The treatment works at Camphill produced finished waters which met EEC requirements (80/778/EEC) throughout the duration of the incident. Oocysts were detected in final waters on two separate occasions (Table 2) at 0.04 and 4.8 oocysts per litre, in the absence of faecal coliform indicators. This is in agreement with the findings of Hayes and colleagues (7) who detected *Cryptosporidium* in final waters in the absence of faecal bacterial indicators. They stated the need to consider new standards and practices to prevent future contamination of major public water supplies. The engineering evaluation following that outbreak identified several pieces of equipment and operational procedures, which previously had been shown to be adequate for water treatment, that would not provide optimal removal and destruction of small protozoan cysts and oocysts such as *Giardia intestinalis* and *Cryptosporidium*.

In the Ayrshire outbreak the filtration and purification treatment at plant appeared to be effective because although oocysts were detected in filter sludges and backwashes, none was detected in final waters leaving the plant. The outbreak appears to have been initiated by irregular seepage of *Cryptosporidium* spp. containing water from the ground in the vicinity of the fire clay pipe. The difficulty in detecting the source of contamination was primarily due to this irregular ground seepage which increased following the heavy rains of late March (Fig. 4) and secondarily due to incorrect records which stated that the pipe had been sealed off many years previously. Thus oocyst contamination of the break-pressure tank which contained final water for distribution, rather than a failure of the water treatment processes appears to be the cause of the outbreak. Circumstantial evidence of the spraying of cattle slurry in the vicinity of the break-pressure tank would enhance the likelihood of oocysts being present in the soil and the heavy rainfall experienced might be expected to enhance their elution from soil and grass into the fire-clay pipe. There is little information on the viability of oocysts in the environment, but Blewett (personal communication) states that purified bovine oocysts, under experimental conditions, can remain viable for 12 months at 4 °C with a loss of up to 70% viability when stored in Hanks' balanced salt solution. Thus it seems likely that oocysts trapped in a soil or water environment can remain viable for many months. Although direct transmission of *Cryptosporidium* spp., from livestock to human beings has been documented (4, 5), a causal relationship between cattle and humans cannot be established in this outbreak. Circumstantial evidence, such as spraying of cattle slurry, and the outbreak occurring at the same time as the spring peak of bovine cryptosporidiosis in that area, enhances the association but until more specific and powerful techniques such as strain specific monoclonal antibodies or DNA probes become available for epidemiological research such associations must remain tenuous. Coupled with the environmental robustness of the oocyst is its resistance to free chlorine. Campbell and colleagues (19) reported that the incubation of oocysts for up to 18 h in a 3% sodium hypochlorite solution (undiluted bleach,

10–14% available chlorine) failed to prevent infection in mice. However, the concentration of free chlorine initially and at the end of the 18 h incubation period was not determined. Recently, Smith and colleagues (20) demonstrated that levels of free chlorine attainable for water disinfection have no oocysticidal effects on *Cryptosporidium* spp. isolated from clinical cases. Levels of up to 16000 mg per litre were necessary to reduce viability to zero, as assessed by *in vitro* excystation. Under the conditions which occurred during the Ayrshire incident oocysts could survive routine disinfection procedures whereas coliform bacteria could not.

Numerous reports have identified the presence of oocysts in water-related samples. Rose (21) found 77% of 107 water-related samples positive for *Cryptosporidium* spp. It is not known how significant the waterborne route of transmission for human cryptosporidiosis is, partly due to the difficulty in its isolation and the significance thereof. In water related samples it is necessary to detect low numbers of oocysts in large volumes of water. In the first two 20 l samples received no oocysts were detected, and further analyses were performed on 500 l samples (Table 2). Twenty-litre samples may not be sufficiently large to enable the detection of oocysts. Larger volumes are recommended (21) because of the small size of the oocysts (4–6 μm) and the inefficiency of cartridge filtration. Such methods rely on oocyst entrapment in a 1 μm (nominal pore size) yarn wound cartridge filter and were developed for the isolation of *G. intestinalis* cysts (typical diameter 10–14 μm) from water samples. Current work in the Scottish Parasite Diagnostic Laboratory indicates that such filters are between 67–96% efficient at trapping *G. intestinalis* cysts, and as *Cryptosporidium* spp. oocysts are smaller it seems likely that they would be trapped less efficiently. Methods are being developed to study the occurrence of *Cryptosporidium* spp. in water, but the recovery efficiencies of filters used, the small numbers of oocysts present in most samples (< 1 oocyst per litre) and an inability to assess viability in such samples are major limitations in determining the prevalence of waterborne *Cryptosporidium*. In addition, oocyst survival in the environment and their removal by conventional processes used in water treatment at present are unknown factors. The isolation and detection of oocysts in water samples is a laborious procedure, taking up to 10 h to process one sample. This reduces the number of samples which can be tested and regular sampling is outwith the capabilities and resources of most laboratories. Such was the case in this outbreak (Table 2). In order to overcome the above problems it is recommended that definitive procedures for the isolation and identification of *Cryptosporidium* spp. oocysts in water should be implemented in order that water samples can be tested for the presence of oocysts at regular intervals, bearing in mind that neither faecal bacteria nor viruses appear to be an indicator of the presence of oocysts. This incident highlights the need to review catchment control measures and to agree satisfactory policies with the farming community with particular emphasis on cattle and sheep grazing and sludge, slurry and manure spreading.

The potential hazard associated with drinking raw or unfiltered waters and the possibility that *Cryptosporidium* oocysts can penetrate water treatment processes and survive chlorination and the importance of the integrity of water pipes has highlighted the need for further research to be carried out in this area. In view of the annual number of laboratory confirmed cases (400–500) for Scotland alone, future combined epidemiological and environmental investigations may indicate

that the waterborne route may be more significant in disease transmission than was previously acknowledged. In Ayrshire a prospective study of all cases of cryptosporidiosis has been undertaken from 1 January 1989.

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