CRYPTOSPORIDIUM PARVUM

Date of document: July 2004

elika

Fundación Vasca para la
Seguridad Agroalimentaria

Nekazaritzako Elikagaien
Segurtasunarako
Euskal Fundazioa

1.- INTRODUCTION

Taxonomically, members of the *Cryptosporidium* genus belong to the *Apicomplexa* phylum, *Coccidia* sub class*, Eucoccidiida* order and *Cryptosporidiidae* family **(1-3)**. They are small parasites that infect the digestive and respiratory tract of vertebrates and, although many species of the ***Cryptosporidium*** **genus** have been described***,*** they seem to be specific for each type of vertebrates **(2)**:

- *C. parvum*, *C. muris, C. felis* and *C. wrairi* for mammals
- *C. baileyi* and *C. meleagridis* for birds
- *C. serpentis* for reptiles
- *C. nasorum* for fish

Infections in **human beings** are caused almost exclusively by ***Cryptosporidium parvum***, which is frequently found in cows and sheep, and infects many other species of mammals **(1, 4)**.

The **life cycle** of the *Criptosporidium parvum* ends in the gastrointestinal tract of the host. The life cycle includes several stages of asexual and sexual reproduction, in which gametes are formed. After fertilisation. these generate cygotes and later oocysts. Two kinds of oocysts are formed: Some with a thick wall, which are released in large numbers through faeces, and others with a thin wall which re-invade the cells of the host's intestine, giving rise to a chronic and persistent infection. The oocysts of *Criptosporidium parvum* have a diameter of 4-6 µm (smaller than other protozoes) and split in the host's small intestine, giving rise to **sporozoites**, which are infective structures. Each mature oocyte contains 4 sporozoites **(1, 3, 4)**.

Criptosporidium parvum is widespread in nature, mainly in the **aquatic medium**. It can be found frequently in rivers and lakes, especially when there have been cattle nearby. Once eliminated through faeces, the oocytes of *Criptosporidium parvum* can **survive** 18 months under wet and cold conditions, in which they remain viable. They are sensitive to heat. A temperature of 65ºC renders the oocytes inactive in 5-10 minutes. They are generally sensitive to **freezing**, although it depends on how the cooling process is begun. Rapid freezing destroys the oocytes, but in a slow freezing process, oocytes have been reported to survive in temperatures of under 22 °C **(1)**. There are studies that show that the freezing of meat products does not render all the oocytes present in the sample inactive **(5)**. On the other hand, oocytes die if left to dry for a sufficient amount of time (more than two hours). They are also extraordinarily resistant to many common **disinfectants**, including **chlorine based compounds (1)**. Recently it has been seen that the treatment of water with ozone or with UV light can be effective in order to render these parasites inactive, although these treatments are not suitable for application on a large scale.

2.- TRANSMISSION

The parasite is transmitted by fecal-oral means and the infection can be acquired in the following way:

- ✓ Contaminated water.
- ✓ Direct contact with the faeces of infected animals that may contain in large number of oocysts (during acute clinical infection, up to 10^{10} oocysts are eliminated every day).
- ✓ Person-person contact, of special importance in schools for young children.
- ✓ Contamination of uncooked foods, such as meat or unpasteurised milk fruit and fresh vegetables.

Most outbreaks of *Cryptosporidium* are due to the consumption of drinking water or exposure to oocysts in swimming pools, rivers, aquatic parks, lakes for sailing or other aquatic activities (**Table 1**: Annex). The largest reported outbreak was in 1993 in Milwaukee (USA), when the main municipal water system was contaminated. During this outbreak, more than 400,000 persons were infected and 100 died. In July-August 1998, in Sydney (Australia) the municipal water system was also contaminated and although no infections were reported, the populace were recommended to boil water before use. This recommendation, which was kept in place for 1 week, affected 3 million residents **(4)**.

Due to its resistance to chlorine, *Criptosporidium parvum* has been a threat for distributors of drinking water who generally depend on natural purification by geological processes or use coagulation or filtering mechanisms as a sole barrier against micro organisms. When these measures fail and/or the number of micro organisms in nature increases, the drinking water can be contaminated and cause sporadic disease or outbreaks of the disease **(4)**.

In the United States, it has been reported that *Cryptosporidium* oocysts can be found in between 17 and 26.8% of samples of treated drinking water, in concentrations of 0.005 to 0.017 oocysts per litre, and in between 9.5 and 22% of samples of underground water **(6)**.

Cases due to cut direct **contact with animals** have also been described among veterinary students and during school visits to farms and due to person-person contact in kindergartens, day care centres, hospitals and old people's homes.

This parasite has also been recognised as a possible cause of **traveller's diarrhoea** as many cases have been detected in patients who had recently travelled to countries where water reserves are frequently contaminated, health levels are not adequate and/or there is a direct contact with animals **(6).**

While water is a well-known vector for the transmission of *Cryptosporidium*, recently it has been seen that **foods** may play a very important role in the transmission of this parasite. Outbreaks linked to the consumption of dairy products, ciders and apple juices and sausages have been described. Meat products, including chicken salads, frozen tripe and raw sausages have also been associated with the cryptosporidiosis. **Table 2** (Annex) shows a large number of outbreaks transmitted by foods and their suspected sources. Generally, it is difficult to identify the source of infection due to the delay between the moment when the food is consumed and the appearance of the symptoms as well as to the lack of an adequate methodology to detect the parasites in foodstuffs. Products treated with heat have never been the cause of outbreaks of *C. parvum*, as the oocysts are rendered inactive during heat treatment **(2)**.

Moreover, *Cryptosporidium parvum* oocysts have been isolated in different geographical areas of **vegetables and foods (6)**, such as the roots and leaves of coriander, lettuce, radish, tomato, pepper, carrot, pumpkin, basil, parsley, cucumber, leak, green onions, green pepper, sprouts of alfalfa, green beans, **seafood products**, such as mussels, clams, cockles and oysters, and **meat products (2).**

3.- DETECTION METHODOLOGY

The available detection methodology is difficult to carry out and cannot be used for routine tests. As it is a parasite, it cannot be cultured or developed outside the host and therefore concentration in foods and faecal and environmental samples is difficult. In water it concentrates by means of filtration and subsequent centrifugation. In order to detect this parasite it is necessary to apply microscopy and dyeing techniques. For serodiagnostics, ELISA methods can be used. Another difficulty of the methodology relates to sensitivity, has a detection methods must be sufficiently sensitive to detect just a few oocysts. Currently, there are specific methods for water and clinical samples, but these do not work well for foodstuffs. More research is necessary into routine methodologies to isolate and detect *Criptosporidium parvum* in water, food and environmental samples **(7)**.

Over the last 3-5 years research has been conducted into the development of methods to isolate and detect the parasite in foodstuffs and apply these methods in order to assess the prevalence and persistence of this parasite in foods. Until now, methods have been described to isolate and detect *Cryptosporidium parvum* in milk, vegetables, fruit, seafood, yoghurt and drinks such as apple juice. These methods include agitation in detergents or traditional bacteriological homogenisation to extract the oocysts from solid matrices, followed by immunomagnetic separation (IMS) with visualisation by immunofluorescent microscopy or the detection of the genetic material of the parasite by PCR. The limitations in the use of these methods in order to isolate *C. parvum* in foods include variations in the recovery of the oocysts. Although these work well for liquid samples (extractions of around 84%), for solid samples, extraction is about 42% **(8)**.

4.- LEGISLATION

Through **Directive 98/83/CE** of the Council of November 3, relating to the quality of water for human consumption **(9)**, the European Union establishes a regulation based on two categories: water for public consumption and packaged water. Both in community and state legislation, these had been treated differently. Taking into consideration the advisability of common health criteria for both types of water, Spanish legislation has maintained regulations with two different but concurrent provisions. Thus, there is **Royal Decree 140 (10)**, of February 7 2003, which establishes health criteria for the quality of water for human consumption and **Royal Decree 1074** of October 18 2002, which regulates the processing, distribution and sale of packaged drinking water **(11)**.

Royal Decree 140/2003 establishes the health criteria to be complied with by water for human consumption and the installations used to collect this, as well as establishing responsibilities and powers, the controls that must be carried out, the analytical parameters and values that water must have in order to be considered to be apt for consumption. This decree relates to water which, whether it is treated or not, is used for drinking, cooking, preparing food, for personal hygiene and for other domestic uses. As far as the food industry is concerned, this decree affects the water used for manufacturing, treating, conserving, commercialising, cleaning surfaces and materials that have to be in direct or indirect contact with foods. Thus, Article 5 states that "water for human consumption is considered to be healthy and clean when it does not contain any kind of micro organism, parasite or substance, in an amount or concentration that might represent a risk for human health". The Annex to this Royal Decree sets out microbiological parameters whereby the count of *Escherichia coli*, Enterococci and *Clostridium perfringens* (including spores) must be 0 ufc in

100 ml. Moreover, it establishes that "when the determination of *Clostridium perfringens* is positive and there is a turbidity of over 5 FNU (Formazin Nephelometric Unit), it will be necessary to determine the existence of *Cryptosporidium* or other micro organisms or parasites, at the outlet from the drinking water treatment plant or tank, if the health authorities consider this to be necessary". Therefore, the regulations do not establish any obligatory value for *Cryptosporidium* **(10)**.

On the other hand, **Royal Decree 1074/2002**, regulates the preparation, circulation and sale of packaged drinking water, this being understood to be natural mineral waters, spring water, prepared water and packaged water for public consumption, which must comply with the following microbiological requirements: Absence of parasites and pathogenic micro organisms, absence of *Escherichia coli*, other coliforms and *Pseudomonas aeruginosa* in 250 ml of sample examined and the absence of Clostridium sulphite reducers in 50 ml of sample examined **(11)**.

European legislation does not make it obligatory to disinfect water or to maintain residual disinfection in the water distribution system and allows each member state to establish its own position on this matter. Article 10 of Royal Decree 140/2003, which refers to the water purification treatment, makes it obligatory to filter water through sand or other appropriate media should the average turbidity of the water collected be over 1 UNF.

In the United Kingdom, Water Supply (Water Quality) legislation in effect since June 1999, requires that water supply companies carry out daily sampling to verify that the amount of *Cryptosporidium* oocysts is less than 1 for every 10 litres of water **(12).**

Directive 2003/99/CE of the European Parliament and the Council of November 17 2003, on the surveillance of zoonoses and zoonotic agents **(13)**, includes cryptosporidiosis and its agents among the parasitic zoonoses that must be controlled in accordance with the epidemiological situation (Annex I, List B).

On the other hand, the Decision of the Commission of March 19 2002, establishes the definitions of cases for the notification of transmissible diseases to the community network, including cryptosporidiosis **(14).**

5.- CRIPTOSPORIDIOSIS

Infection from *Cryptosporidium parvum* is called **cryptosporidiosis**. In **animals** it usually develops in asymptomatic manner, although cattle release a large number of oocysts in faeces. In **young cattle** it may cause **diarrhoea** and is a disease of veterinary importance. Studies carried out in lambs in Castilla-León show that cryptosporidiosis represents a serious problem on a large number of sheep farms and although mortality is not high (4%), there is a major delay in the growth of affected animals of around 23%. Ewes also start milking late and milk production decreases. The predisposition to develop mammitis increases due to the retention of milk in udders **(15)**.

According to published data, an examination of faeces samples in the developed countries has shown that prevalence in animals is between <1% and 4.5%, when many infections are subclinical. (Eurosurveillance, 1998). In Denmark, annual Zoonosis reports indicated the prevalence in cattle of between 13.2% in 2000, 11.4% in 2001 and 10.9% in 2002 (**Table 3**: Annex)**(16)**.

These studies show that animal waste must be kept during a sufficient period of time or be treated actively in order to eliminate pathogenic micro organisms, before being applied to crops **(2)**.

In **humans**, the most common **symptom** of infection by *Criptosporidium parvum* is **diarrhoea**, followed by abdominal pain, nausea and vomiting, fever and loss of appetite and weight. The illness has a longer symptomatic period than most gastrointestinal bacterial infections. In healthy individuals, complete recovery generally comes within 2-3 weeks, although it may take to 6 weeks. In **immunodepressed individuals**, the illness may be more severe and persistent, with the invasion of other organs, including lungs and the bile duct, endangering the life of the patient. In this risk group, it has been described that the risk of mortality stands at between 50-60% of infected individuals. The first case of this disease in humans was reported in 1976, although until the beginning of the eighties, it was not recognised as a human disease, being associated with immunodepressed patients **(1, 4, 7)**.

C. parvum sporozoites attack the ephitetelium of the stomach initiating the infection and, after a incubation period of from 2 to 3 days, the pathogen causes symptoms in humans (1-3). Studies carried out among healthy human volunteers show a clear relationship between the likelihood of infection and the doses of C. parvum oocysts (Dupont et al, 1995). With the lowest dosage of 30 oocysts, the likelihood of infection was 20% and, when the dose was increased to 1000 oocysts, the likelihood increased to 100%. This approach assumes that the ingestion of a single oocyte results in a probability of infection of 0.5% **(3)**.

The incidence of cryptosporidiosis in humans is difficult to determine, because it is not obligatory to declare this disease. The incidence in the population has been described as being between 0.6 and 20% according to geographical area. It is more prevalent in Asia, Australia, Africa and South America, while in the **United States** it has been associated with 0.4-1% of cases of diarrhoea **(6),** some 10 million cases per year. In **England, Wales and Northern Ireland**, some 6000 cases of infections by this parasite are reported on a yearly basis (see **Table 4**). Of these cases, almost 50% occur in children of under 9 years of age. In Northern Ireland, it is the second cause of gastroenteritis in this region, affecting more than 50% of cases of children of under four years of age (see **Graph 1) (17).** In **Scotland**, in 1998, an increase in the number of cases in humans was reported **(18).** In **Denmark**, few cases have been notified, standing at 39 in 2000, 84 in 2001 and 38 in 2002 **(16)**. **In developing countries,** the prevalence among immunocompetent individuals has been described at being between 20 and 30 % **(7)**.

In addition to reported cases of the illness, **serological controls** have shown that *Cryptosporidium parvum* antibodies are present in the blood of **25-30%** healthy individuals tested in Europe and North America **(6, 7)**, and in **65-85%** of the population of developing countries. This indicates high levels of exposure to the pathogen in spite of the fact that this disease is not diagnosed in most individuals **(7)**.

In spite of the fact that some drugs can eradicate the parasite, there is no effective pharmacological treatment to eliminate the disease. (**1, 7**)

In the **Spanish state**, cryptosporidiosis is not currently subject to any control although it is expected to be included in the list of diseases to control, given that the European Union recently decided to do so. In spite of the fact that there is no official control of this pathogen, the National Epidemiological Surveillance Network has information about this disease in two of its basic systems: The Epidemiological Information System and the Outbreaks System **(19)**.

The **Microbiological Information System (MIS)** is based on a voluntary weekly notification by clinical microbiological laboratories, mainly in hospitals, of the diagnoses they carry out. From 1995 to 2002, 823 cases of infection by *Cryptosporidium* were notified to the system, representing a yearly average of 103 cases. The distribution of cases according to age groups shows the higher number of notifications in the 1-4 years group With regard to patient immunity level, results show *Cryptosporidium* as a causal causal agent of diarrhoea irrespective of the immunity status of the host. During 2003, 63 cases were notified, although the data is provisional as of December 11 **(Table 5)**.

The **Outbreaks System** obtains and analyses the results of research into outbreaks or academic situations in Spain including those that affect foreign tourists who have been in this country. From 1995 to 2003 (data for 2003 is provisional), 11 outbreaks were notified with a total of 1455 cases and an average of 132.3 cases per outbreak. The environments where cases were most frequently notified were schools (12.2% of cases), hotels (29.1% of cases) and towns (58.4% of cases). The transmission mechanism was water in all outbreaks in which this detail was known and the source was the water supply network on three occasions and a swimming pool on another two occasions **(Table 6)**.

In addition to these outbreaks, the National Epidemiological Centre has been notified about 15 outbreak alerts of *Cryptosporidium* in foreign tourists who had visited Spain, with a total of 95 cases (34 confirmed and 61 suspected). These alerts have not been confirmed as outbreaks by the Autonomous Communities (1 in the Canary Islands, 1 in the Community of Valencia, 2 in Catalonia and 11 in the Balearic Islands). The 15 alerts were declared by the Scottish Centre for Infection and Environmental Health **(19)**.

The MIS detects infections by this parasite, although this system is not completely developed and many laboratories in this country do not include cryptosporidiosis diagnosis within their routine investigations. It would be necessary to promote the SIM declaration by laboratories. Although insufficient, the results of the Outbreak System oriented us towards the detection of outbreaks in two specific environments, namely schools and hotels. In outbreaks with a known vehicle, the main source of the outbreak in schools is the water supply network and in hotels this is the swimming pool. It would be necessary to promote a control of this pathogen (declaration in the MIS and search for outbreaks), as well as to insist upon the control of water supply systems and swimming pools **(19)**.

6.- RISK ASSESSMENT STUDIES

A study made in **Holland** by TNO-Nutririon and Food Research **on the Evaluation of the quantitative risk of *Cryptosporidium* in food and water,** reached the conclusion that more data is required in order to assess the risk in a quantitative manner, but proposed measures in order to improve the current situation:

- ✓ With regard to the risk from consuming tap water, the most important factors to be controlled are the initial contamination of the water before treatment and the efficiency of the treatments carried out in water purification plants. With regard to raw fruit and vegetables for consumption, the most important risk factor is the faecal contamination of products in the field. It would be recommendable to treat fertilisers (sludge, liquid manure and slurry) in order to render the *Cryptosporidium* oocysts inactive before being applied to crops.
- ✓ The industrial treatment (preparation and cleaning) of fruit and vegetables is also important.

- ✓ In meat products, the greatest risk exists in the contamination with the faecal material or crossed contamination between carcasses in slaughterhouses, although generally these products had treated with heat, thereby rendering the *Cryptosporidium* oocysts inactive **(20).**

In 2003, a **Risk Assessment of *Cryptosporidium parvum*** in the food chain **in Europe** was carried out in a project in which 5 European countries took part (Ireland, United Kingdom, Denmark, Italy and Holland). The general conclusion of the study is that the risk of infection through the consumption of drinking water for the population in general is low. The risk is also very low in vegetables and meat. This study recommends that when water comes from areas with a highly risk of contamination, a stricter treatment should be made of these. With regard to vegetables, it is recommendable to clean these thoroughly and eliminate peels, external release, etc. and keep them in low-temperature storage. Meat should be cooked adequately and cross contamination with vegetables should be avoided **(21)**

7.- BIBLIOGRAPHICAL REFERENCES

1 Khan O.A. **A Review of Cryptosporidiosis**. Carlo Denegri Foundation - Atlas on Medical Parasitology.

2 Duffy G, Moriarty, EM (2003) Cryptosporidium and its potential as food-borne pathogen. Animal Health Research Reviews 4(2):95-107.

3 Fricker CR. Protozoan parasites (Cryptosporidium, Giardia, Cyclospora). Guidelines for drinking-water quality.

4 Dawson D. (2003) Foodborne Protozoan Parasites. ILSI-International Life Sciences Institute. Report prepared under the responsibility of the ILSI Europe Emerging Pathogen Task Force.

5 McEvoy JM, Moriarty EM, Duffy G, Sheridan JJ, Blair IS, McDowell DA. (2004) Effect of a commercial freeze/tempering process on the viability of *Cryptosporidium parvum* oocysts on lean and fat beef trimmings. Meat Science, 67:559-564.

6 Rose JB, Slifko TR (1999) *Giardia*, *Cryptosporidium* and *Cyclospora* and their Impact on Foods: A Review. Journal of Food Protection, 62(9):1059-1070.

7 Duffy G. ***Cryptosporidium parvum* – an Emerging Pathogen in the Water and Food Industry**. Irish Agriculture and Food Development Authority.

8 Moriarty EM, McEvoy JM, Duffy G, Sheridan JJ, Blair IS, McDowell DA (2004) Development of a novel method for isolating and detecting *Cryptosporidium parvum* from lean and fat beef carcass surfaces. Food Microbiology, 21:275-282.

9 Directive 98/83/CE

10 Royal Decree 140 of February 7 2003, which establishes the health criteria for the quality of water for human consumption.

11 Rural Decree 1074/October 18 2002 which regulates the processing, distribution and commercialisation of package drinking water.

12 Hunter PR (March 2000) **Advice on the response for public and environmentalhealth to the detection criptosporidial oocysts in treated drinking water** . Communicable Disease and Public Health, 3 (1) 24-27.

13 The Directive 2003/99/CE of the European Parliament and Council, of November 17 2003, on the control of zoonosis and zoonotic agents.

14 Decision of the Commission of March 19 2002, establishing the definition of cases of diseases transmittable to the community network, the columns with Decision no. 2119/98/CE of the European Parliament and Council.

15 Gutierrez J, Martín S, Manteca Ch, Rojo-Vázquez FA (Mayo 2004) Ovine Cryptosporidiosis. Mundo Ganadero nº166.

16 Ministry of Food, Agriculture and Fisheries Annual Report on Zoonosis in Denmark: **2000**, **2001** y **2002**.
17 Health Protection Agency. (May 2003) ***Cryptosporidium*** – Epidemiological data
18 Christie P (1998) Increased cases of *Cryptosporidium* infection in central Scotland. Eurosurveillance Weekly, 2(19).
19 National Epidemiology Centre–Carlos III Health Institute (2003) Epidemiological Surveillance of Cryptosporidiosis in Spain. Epidemiological Bulletin, 11(24):277-284.
20 Hoornstra E, Hartog B. A quantitative Risk Assessment on Cryptosporidium in food and water. TNO Nutrition and Food Research, Holland.
21 Duffy G et al (2003) A Risk Assessment on Cryptosporidium parvum, an emerging pathogen in the food and water chain in Europe. QLK1-1999-CT-00775.

8.- ANNEX: TABLES AND GRAPHICS

Table 1: Main outbreaks of *C. parvum* transmitted by water **(4)**

YEAR	Place	Source	No. cases
1987	Carrolton, Georgia (USA)	Surface Water	13,000
1989	Oxford/Swindon (UK)	Surface Water	500*
1993	Milkwaukee/Wisconsin (USA)	Surface Water	403,000
1996	Ogose (Japón)	---	9,000
1997	North London (UK)	Underground Water	345*

*Confirmed by laboratory

Table 2: Main outbreaks of *C. parvum* transmitted by foods **(2)**

YEAR	Place	Source	No. cases
1985*	United Kingdom	Frozen tripe	1
1986*	Mexico	Milk (Canadian tourists)	22
1986*	Australia	Unpasteurised goat's milk	2
1986*	Wales	Sausages	19
1986*	Mexico	Salads	1
1993	Maine (USA)	Unpasteurised cider	154
1996	New York (USA)	Unpasteurised apple juice	31
1996	USA	Chicken salads	15
1997	Washington (USA)	Green onions	54
1997*	United Kingdom	Cow's milk	50
2000*	USA	Fruit/vegetables	148
2002*	Queensland	Unpasteurised cow's milk	8

*Year outbreak was published

Table 3: Cases of *C. parvum* in livestock in Denmark **(16)**.

YEAR	2000		2001		2002	
	No. determ.	Positive	No. determ.	Positive	No. determ.	Positive
Cows	2730	13.2	2509	11.4	2825	10.9
Goats	544	0.9	492	0	625	0.2
Pigs	2390	0.5	1503	0.3	933	0.9
Horses	734	0.7	477	0.2	397	0
Dogs	197	1	170	1.2	199	1
Cats	38	0	52	0	107	1.9
Others	716	0.3	751	0.5	622	5.7

Table 4: Identifications of *Cryptosporidium sp* in laboratories in England, Wales and Ireland **(17)**

Year	England and Wales	Ireland
1986	3560	--
1987	3277	--
1988	2750	--
1989	7768	--
1990	4682	--
1991	5165	--
1992	6164	58
1993	4753	177
1994	4502	89
1995	5701	81
1996	3587	98
1997	4393	82
1998	3670	180
1999	5045	181
2000	5774	417
2001	3625	360
2002	2992	126
2003*	5532	140

*Provisional data

Graph 1: Cases of *Cryptosporidium sp* identified in laboratories in Northern Ireland according to age range. 1998-2003 **(17)**

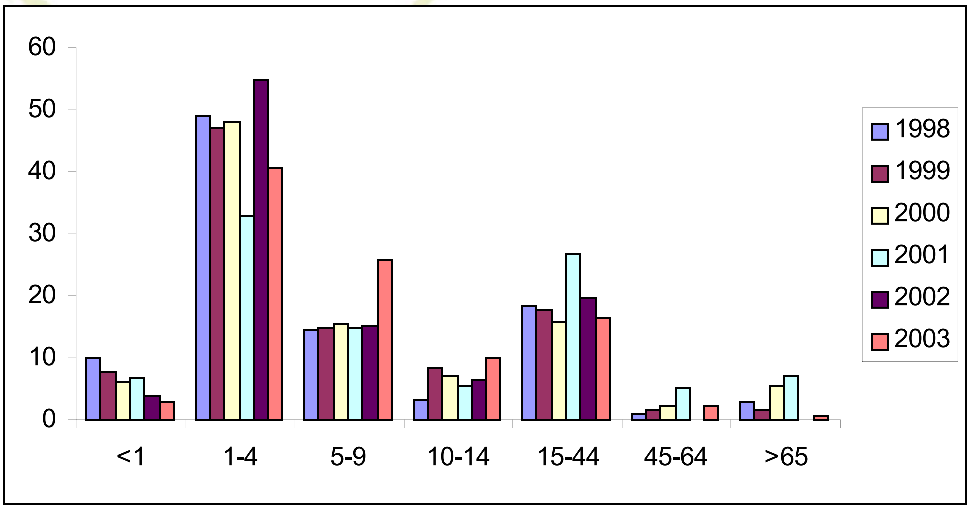

Table 5: Cases of cryptosporidiosis notified to the MIS in Spain, distributed according to Autonomous Communities, (1995-2003) **(19)**

CC.AA.	1995	1996	1997	1998	1999	2000	2001	2002	2003	TOTAL
Aragón	48	58	68	39	82	36	74	98	45	**548**
Balearic Islands		1	1	1						**3**
Canary islands	18	9	22	7	7	11	7	15	12	**108**
Castilla La Mancha	1		1	1						**3**
Castilla León		2	4	1		1		1	3	**12**
Catalonia	27	8								**35**
Valencia		2								**2**
Madrid	3	11	1				4	4	1	**24**
Basque Country	47	47	15	12	8	6	3	3		**141**
La Rioja	5		2	1						**8**
TOTAL	**149**	**138**	**114**	**62**	**97**	**54**	**88**	**121**	**61**	**884**

Table 6: Outbreaks of cryptosporidiosis notified to the National Epidemiological Surveillance Network in Spain (1995-2003) (*Outbreaks notified initially by European surveillance services) **(19)**

Year	AA.CC.	Environment	Exposed	Cases	Symptoms	Vehicle	Source	Observations
1997	Andalusia	School	200	66	October	Water	Water supply network	Breakdown/works
1998	Madrid	School	519	62	April	Water		Contamination installation
1998*	Andalusia	Hotel	2500	3	July			Tourists
1999	Madrid	School	138	36	October			
2000	Aragón	Town		750	January	Water	Water supply network	Contamination surface water farming land
2000	Aragón	Town		100	May	Water	Water supply network	Insufficient water treatment
2000*	Balearic Islands	Hotel		25	May	Water	Swimming pool	Tourists
2000	Catalonia	School	45	13	October			
2001	Madrid	Picnic	80	5	July	Water	Well	Untreated water, farm
2003*	Balearic Islands	Hotel	2000	391	July	Water	Swimming pool	Tourists
2003*	Balearic Islands	Hotel		4	July			Tourists