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Molecular Characterization and Genetic Diversity of *Cryptosporidium spp.* in Human Infections: A Review

Iltefat Amer Al-Tameemi [1,*], Samiya Shamkhi Jebur [2] and Ayoob Radhi Al-Zaalan [1]

[1] *Department of Medical Lab Technology, College of Health and Medical Technology, Southern Technical University, Basrah, Iraq.*
[2] *Ministry of Education, General Directorate of Education in Basra.*

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Abstract

The *Cryptosporidium spp*. are protozoan parasites of significance in the world and the major cause of the diarrheal disease (cryptosporidiosis) in humans, which presents a significant risk to young children, immunocompromised people, and livestock. The introduction of molecular methods has completely transformed the view on the taxonomy, genetic diversity, and the mode of transmission of the parasite that was not initially evident due to the morphological homogeneity of the oocysts. This review summarizes the existing information and knowledge regarding the molecular characterization of species of Cryptosporidium that infect human beings based on genetic diversity. We touch upon the development of diagnostic and typing methods, starting with the classical microscopy and all the way to the current state of multilocus sequence typing (MLST) and whole-genome sequencing (WGS). The review addresses the two most common human-pathogenic species, C. hominis and C. parvum, includes the description of the population structure, genetic recombination role and the public health importance of different subtype families. We look at the specific epidemiological trends where anthroponotic is more likely in low- and middle-income nations than is zoonotic in the high-income nations. In addition, the clinical consequences of this genetic diversity, especially in the vulnerable groups like the HIV/AIDS patients are discussed. This review brings together the results of the most important molecular epidemiological research and points out how advancing knowledge about the genetic landscape of Cryptosporidium can be very essential in crafting an effective disease management approach, outbreak surveillance and population health interventions.

Keywords: Cryptosporidiosis; *Cryptosporidium parvum*; Genetic Diversity; Zoonotic Transmission

1. Introduction

Cryptosporidium is an obligatory intracellular protozoan parasite that is a globally widespread well-established pathogenic organism causing the diarrheal illness cryptosporidiosis and belongs to the phylum Apicomplexa [1,2]. It was first described in 1907 in the gastric mucosa of laboratory mice by the scientist Ernest Edward Tyzzer, but its importance as a human pathogen was not realized until it was first identified in diarrheal disease of the human host in 1976 [2].

The parasite was first highly recognized in the 1980s as an etiological agent of severe, life-threatening diarrhea in victims with acquired immunodeficiency syndrome (AIDS) and first became widely known through a massive water-borne outbreak in 1993 in Milwaukee, Wisconsin, which had an estimated incidence of 403000 cases [3]. *Cryptosporidium* is currently recognized as one of the principal causes of moderate-to-severe diarrhea and mortality related to the condition, especially in children below the age of five years in low- and middle-income countries (LMICs)

* Corresponding author: Iltefat Amer Al-Tameemi

and in immunocompromised patients around the globe [2,4,5]. *Cryptosporidium* is transmitted mostly through fecal-oral route, that is, through ingestion of hardy, environmentally resistant oocysts. The ways of getting infected include person-to-person contact, zoonotic diseases that infect, and ingestion of infected water or food [1,2]. Clinical manifestation of cryptosporidiosis is diverse, and it may manifest as asymptomatic infection in immunocompetent individuals to acute self-limiting diarrhea and chronic debilitating disease in the case of immunocompromised people [6,7].

The microscopic size of the parasite hindered the progress of the *Cryptosporidium* epidemiology and taxonomy study in decades because the oocysts of various species were not separated by a distinct morphology [1], this morphological homogeneity that ensured that species could not be differentiated using microscopy itself and posed extremely difficult problems in determining the transmission dynamics, host specificity, and the public health importance of different *Cryptosporidium* species, Molecular biology has significantly changed the discipline with its introduction, the use of molecular tools especially DNA sequencing and genotyping has revealed a startling amount of genetic diversity in the genus, which resulted in the recognition of a large number of species and genotypes with different host-preferences and implications on public health [1,8,9]. Today, more than 40 species of *Cryptosporidium* are identified, and no less than 20 are capable of infecting human beings [2,8]. Nevertheless, two species are considered the primary sources of human infections in the world, including *C. hominis* (anthroponotic transmission) and *C. parvum* (zoonotic transmission) [2,4,9]. The capacity to differentiate them and other species and the capability to describe genetic variation among them (subtyping) is vital in tracking the sources of infection, outbreaks, and adjusting appropriate control actions.

The present review offers an overall view of the present body of knowledge on the molecular characterization and genetic diversity of *Cryptosporidium spp*. in human infections. We are going to see how molecular tools have developed since the ancient genotyping techniques, how genetic diversity and population structure of the major human-pathogenic species evolves, and what the epidemiological and clinical consequences of this genetic variation may be. Through a synthesis of the most significant discoveries of the past 20-years of molecular studies, this review is expected to offer a clear understanding of the complicated genetic map of *Cryptosporidium* and its significance to the health of people.

2. Taxonomy and Species of Public Health Importance

Since its original description, the taxonomy of the genus *Cryptosporidium* has been modified substantially. The species classification has been a matter of confusion over many years and has mostly been attributed to the fact that the parasite has subtle morphological variations and was considered to have no rigid host specificity In the past, the classification of species was based on a mixture of the morphology of the oocyst and the host of origin and the site of infection, however, proved inadequate in the correct identification of oocysts because the discovery that the oocysts in the various species frequently have overlapping size ranges and similar shapes [1].

Molecular data integration has been useful in the elucidation of the taxonomic relationships in the genus. A new system of nominating new *Cryptosporidium* species currently needs a polyphasic system, combining oocyst morphology, natural host specificity and an effective genetic characterization, It has resulted in the rapid increase in the number of identified species, to more than 40 nowadays, whereas in 2004, there were 13 [1,2,8]. Although an increasing number of species has been reported to cause occasional cases of infections in humans, *C. hominis* and *C. parvum* cause most of the world disease burden [2,4]. *C. hominis* is highly host adapted to humans and is one of the main agents of anthroponotic (human-to-human) transmission and is particularly prevalent in overcrowded regions and low-income countries with inadequate sanitation [5]. Conversely, *C. parvum* has a wide host range with one of the important reservoirs being cattle. It is the main causative agent of zoonotic cryptosporidiosis, which is often associated with rural outbreaks and high-income countries in contact with a sick animal or in an infected environment [4,9].

Other species are responsible of lesser, nonetheless, considerable share of human infections. The third most prevalent species in humans worldwide is *C. meleagridis* that is mostly recognized as a turkey pathogen [8]. It is also known that others, including *C. canis* (dog-derived), *C. felis* (cat-derived), *C. ubiquitum* (ruminant and rodent-derived), and *C. cuniculus* (rabbit-derived), are also considered as zoonotic agents, but not detected as often [8,9]. The proper identification of these species is not just an academic game, but it is essential to measure the risks to the population, the routes of transmission, and specific intervention measures. As an example, the *C. parvum* outbreak in a waterborne outbreak would lead to a search of the agricultural sources of contamination, but an outbreak of *C. hominis* would lead to a human sewage source [9].

3. Molecular Tools for Characterization and Typing

Correct identification and distinguishing of *Cryptosporidium* species and subtypes are vital in epidemiological surveillance, investigation of outbreaks, and risk evaluation. These shortcomings of the traditional approaches have led to the creation and use of a collection of potent molecular tools.

3.1. Conventional Diagnostic Methods

Over a long period, cryptosporidiosis diagnosis was based on the microscopic observation of oocysts in feces. The most common has been the modified acid-fast staining technique in which the oocysts present as bright red spheres on a blue or green background [6]. Although precise in the hands of an expert microscopist, this technique is tedious, lengthy, with a low sensitivity, especially when the oocyst shedding is low [2]. Immunofluorescence assays (IFA) and enzyme-linked immunosorbent assays (ELISA) which are used to detect *Cryptosporidium*-specific antigens are more sensitive and less operator-dependent when compared to microscopy. Nevertheless, such immunological techniques are usually unable to distinguish species which is an essential constraint of molecular epidemiological investigations [2,4].

3.2. PCR-Based Detection and Key Molecular Markers

Application of Polymerase Chain Reaction (PCR) has been one of the greatest achievements in diagnosis as well as research. Assays based on PCR are much more sensitive and specific compared to microscopy or antigen detection techniques [2]. Moreover, they give the DNA template to be used in the genetic characterization of the sample to be performed later with the help of various molecular markers.

3.2.1. The 18S rRNA Gene: The Gold Standard for Species Identification

The small subunit (SSU) rRNA gene is the most commonly used genetic marker to identify a species. It is a perfect target due to its universal occurrence in eukaryotes, and the conserved and variable architecture. These conserved regions can then be used to construct broad-range primers that will amplify DNA of most *Cryptosporidium* species and the variable regions consist of species-specific sequences hence differentiation by sequence analysis or Restriction Fragment Length Polymorphism (RFLP) is possible [1]. It is the foundation of *Cryptosporidium* molecular diagnosis and taxonomy as it allows the most important and crucial step, and that is, the identification of the species of the organism involved in an infection.

3.2.2. The gp60 Gene: The Standard for Subtyping and Epidemiological Studies

Although the 18S rRNA gene is a great species-level identification, it is not always resolute enough to differentiate between isolates of the same species. A more variable marker is required to monitor transmission pathways and learn more about population structures. The gp60 (or gpa) gene is a 60-kDa glycoprotein that has become the gold standard of *C. hominis* and *C. parvum* subtyping [8,9]. The *gp60* gene expresses a surface glycoprotein that is crucial in host cell binding and invasion and the degree of polymorphism, especially in a serine-rich repeat region, is indicative of the selective pressures of the host the immune system [8]. The developed subtyping system using gp60 sequences variation has been used to connect human cases with animal reservoirs, source of outbreaks and to explain the unique anthroponotic and zoonotic process of spread that characterize the epidemiology of cryptosporidiosis.

3.2.3. The COWP Gene: A Supplementary Diagnostic and Genotyping Marker

Cryptosporidium oocyte wall protein (COWP) gene is another significant genetic marker. This is a structural protein gene of the tough oocyst wall. COWP gene has been extensively used as a molecular detection and a genotyping target gene and commonly in a nested PCR method to enhance sensitivity [10]. Although the gene is not as polymorphic as the gp60 gene, variation in the sequence in the COWP gene can be taken advantage of in PCR-RFLP analysis to distinguish between some species and even isolates of *C. parvum* of various host origins. It is mostly useful in supporting molecular diagnosis and in giving further information of genotyping in relation to data on the 18S rRNA and gp60 genes, particularly in experiments exploring the genetic diversity of isolates of diverse sources, such as waterborne outbreaks [10].

3.3. Advanced Genomic Approaches

In more recent years, the field has shifted to even more finer resolution techniques. Multilocus sequence typing (MLST) is another technique that uses a sequence variation of a set of housekeeping genes to obtain more robust data on population genetics and recombination compared to single locus typing [5,9].

Most importantly, the emergence of the next-generation sequencing (NGS) has facilitated whole-genome sequencing (WGS) of *Cryptosporidium* isolates directly off clinical samples [11]. WGS provides the final solution to isolate differentiation, to investigate genes that are under selection, to study genome structure, and to reveal complicated evolutionary mechanisms, e.g., genetic recombination and introgression [9,11]. By 2024, more than 70 *Cryptosporidium* genome assemblies were publicly availed, which is an invaluable source of comparative genomics. These new genomic methods are transforming the knowledge of the biology, virulence, and epidemiology of the parasite, thus opening the way to the creation of new diagnostics, therapeutic methods, and vaccinations [11].

4. Genetic Diversity and Population Structure

Molecular subtyping has identified that *C. hominis* and *C. parvum* are not homogeneous populations but are made up of different genetic lineages with diverse biological and epidemiological properties. The dynamic of their population structure is the centre of study to comprehend the host adaptation, virulence, and patterns of transmission.

4.1. *Cryptosporidium hominis*

C. hominis has been regarded as a human-adapted species and anthroconotic transmission is a major determinant in its population structure. International research has discovered that many prominent subtype families, namely Ia, Ib, Id, Ie, and If, among others, exist [5]. There is unique geographic distribution of some of these subtype families. Indicatively, the Ib subtype family is so widespread in most regions of the world, yet the IbA10G2 subtype specifically has been linked to high levels of virulence and is a significant contributor to the occurrence of cryptosporidiosis in children in LMICs [5,9]. It seems that genomic recombination in *C. hominis* is less common as compared to *C. parvum* and is predominantly clonal. Nevertheless, instances of recombination have been found especially in the virulent subtype of IbA10G2, which means that genetic exchange happens on occasion and thus contributes to its evolutionary success [9]. Geographic segregation also affects the population structure of *C. hominis* resulting in the separation of the populations in the various regions. Interestingly, nonhuman primates and equine animals were found to harbor *C. hominis* populations which are genetically different, and it is uncertain whether this parasite could be cross-species transmitted and how this human-adapted parasite evolved [9].

4.2. *Cryptosporidium parvum*

Unlike *C. hominis, C. parvum* possesses a wide host spectrum and a more intricate population dynamics with two types of transmission i. e. zoonotic and anthroponotic. The most common subtype family is IIa that is typical of pre-weaned calves and is frequently a common cause of zoonotic infections in humans, particularly in agricultural areas with a high income [4,9]. There is a subtype that is very extensive IIaA15G2R1 and regarded as hyper transmissible [9]. Nevertheless, an alternative scenario has been depicted in molecular studies in LMICs, In these areas, the human infection of *C. parvum* is most transmitted by the IIc subtype family, which seems to be anthropologically transmitted, like *C. hominis* [5]. This has important public health consequences, which indicate that control interventions against *C. parvum* need to target human-to-human transmission (e.g. sanitation and hygiene) as well as zoonotic sources in many regions of the globe. The genetic relatedness of *C. parvum* suggests that genetic recombination of the species is also frequent, specifically in the zoonotic IIa subtype family, which leads to its great genetic diversity and adaptability [9]. Anthroconotic subclade in *C. parvum*, which has presumably been formed as a result of the introgression of the DNA of other *Cryptosporidium* species, has also been detected through comparative genomic studies, further emphasizing the complicated evolutionary dynamics of the discussed pathogen [11].

5. Clinical Manifestations and Vulnerable Populations

The immune status of the host is very critical in the clinical spectrum of cryptosporidiosis. Although the genetic properties of the parasite, including species and subtype, can be used to determine the severity of the symptoms, the host immune response is the main determinant of the clinical outcome [4,6].

5.1. Immunocompetent Individuals

Cryptosporidium infection is an acute self-limiting condition in individuals with a normal immune system. Profuse and watery diarrhea is the most frequent symptom that may be accompanied by abdominal cramps, nausea, vomiting, low-grade fever, and weight loss [6]. The symptoms are normally evident after incubation of approximately one week and then they may take one to two weeks and fade away on their own. There are also cases of asymptomatic infections, especially in instances where adults in endemic regions may have acquired partial immunity because of a prior exposure [4]. Even though the disease is usually self-limiting, it may be disabling and cause severe morbidity and financial loss through absenteeism at work or in school.

5.2. Immunocompromised Individuals

Cryptosporidiosis poses a significantly better and chronic risk to immunocompromised patients. Patients with HIV/AIDS are the most researched of these groups. In such patients, especially those who have lower than 200 cells of CD4+ T-cells count, the infection is usually chronic and life-threatening [1,6]. The diarrhea may become incessant and voluminous causing severe dehydration, malabsorption, wasting, and even death. Although the gastrointestinal tract is the main site of infection, in severely immunocompromised persons, the parasite may spread to other areas, such as the biliary tract (to cause sclerosing cholangitis and acalculous cholecystitis), ducts of the pancreas, and the respiratory tract (to cause cough and shortness of breath) [6]. Cryptosporidiosis was a significant killer of AIDS patients before the presence of effective antiretroviral treatment (ART). Although the incidence is lower in those areas that have access to ART, it is still a prominent opportunistic infection in resource-limited areas and among patients with no idea about their HIV status or with poor adherence to treatment [5,6,12].

5.3. Children in Low- and Middle-Income Countries

Another regionally poor population is the young children, particularly children under the age of two years. *Cryptosporidium* was also known as one of the most significant pathogens causing moderate-to-severe diarrhea among the age group, only followed by rotavirus [4]. Recurring cases of cryptosporidiosis during early childhood are closely linked with malnutrition, retarded growth, and cognitive retardation.The malnutrition and infection is a vicious cycle since malnutrition compromises the immune system, making one more prone to infection and the diarrheal disease affects malnutrition by inhibiting the absorption of nutrients. This is a significant, frequently mis-acknowledged, societal healthcare cost [4,5,13].

6. Transmission Dynamics: Anthroponotic vs. Zoonotic Cycles

In order to generate proper public health interventions, it is essential to understand how *Cryptosporidium* is transmitted. Molecular tools have played a key role in breaking down the complicated transmission cycles and show two different patterns in the transmission of the two major human pathogens, *C. hominis* or *C. parvum*.

6.1. Anthroponotic Transmission

The *C. hominis* and the *C. parvum* IIc subtype family are predominantly transmitted by anthroponotic transmission, or human-to-human transmission [5,9]. The cycle is very effective in places that are poorly sanitized and hygienic where human sewage often contaminates water and food. *C. hominis* has always been the most common species in LMICs, whereas anthroponotic transmission has been a major determinant of childhood diarrhea globally in the GEMS study and other large-scale epidemiological studies [4,5]. Anthroponotic subtypes are also common, which implies the relevance of water, sanitation, and Hygiene (WASH) interventions. Improving the availability of safe drinking water, setting up an effective wastewater treatment process, promoting safe food handling and washing of hands is something that is required to break this transmission cycle. *C. hominis* outbreak is usually associated with contaminated municipal water, swimming pools or community meetings where one human source may cause the whole community to contract the disease [2].

6.2. Zoonotic Transmission

Zoonotic transmission is based on the transmission of the parasite to humans by animals. It is the main path of transmission of the *C. parvum* IIa subtype family, and the main reservoir is young livestock, especially the pre-weaned calves [9]. Human beings can be infected by direct contact with infected animals (ex: at farms or petting zoos) or in the more frequent case, by ingesting oocysts that have contaminated the environment, water sources or food products. Zoonotic transmission is the transmission of the parasite of animals to humans. This is the main path of transmissions of the *C. parvum* IIa subtype family with the main reservoir being young livestock, especially pre-weaned calves [9]. Human beings may be infected by direct contact with infected animals (e.g. in farms or petting zoos), or, more frequently, by ingesting oocysts that have been deposited in the environment or water bodies, in foodstuffs, Cryptosporidiosis in rural and agricultural areas of high-income countries is largely caused by zoonotic transmission [4].

The disease has repeatedly proven to have been transmitted through the pollution of drinking water reservoirs by farming wastes that have animal feces. *Cryptosporidium* oocysts pose a significant danger to the water systems of people since they are resistant to common gatekeeping disinfection procedures, including chlorination [14]. Although *C. hominis* is the most prevalent of the zoonotic species globally, there are other species that cause human illnesses, such as *C. meleagridis*, *C. canis*, and *C. felis,* which are also important in identifying the source of an infection or an epidemic [8].

7. Conclusion

The use of molecular tools in the last 25 years has revolutionized the research of *Cryptosporidium*. A single, morphologically indistinct parasite is now known to be a complex genus with more than 40 species, each having different degrees of host specificity and public health importance. The development of molecular characterization has rendered the field much farther beyond the constraints of the old microscopy because it is now essential to the precise diagnosis, epidemiological surveillance, and outbreak detection. Of particular interest is the genetic diversity of the *C. hominis* and *C. parvum* which are the major human-pathogenic species. Host adaptation, geographical segregation and genetic recombination have been shown to produce different population structures through the use of gp60 subtyping and more recently through multilocus and whole-genome methods. The apparent differentiation between the human-dominated fanatic spread of *C. hominis* and the zoonotic prospects of *C. parvum* have significant resourceful impacts on the general health, which direct actions towards the most probable source of infection. The fact that anthroponotic *C. parvum* subtypes (e.g. IIc) are predominant pathogens in most LMICs has also made our knowledge even more specific, with control measures being required to be specific to local epidemiology. This genetic diversity has the greatest clinical importance in susceptible groups. The disastrous effect of chronic cryptosporidiosis on immunocompromised patients and association of frequent infections and malnutrition in small children highlight the urgency of finding effective control measures. As the pace of the discovery of new drug and vaccine targets is increasing due to new innovations brought on by genomics, there are major challenges. The only FDA-approved medication to treat cryptosporidiosis is nitazoxanide, yet its effects are not effective in immunocompromised patients. The complicated life cycle of the parasite and absence of a strong in vitro culture system over time persist in obstructing research. The work ahead should be the incorporation of sophisticated genomic information into international surveillance so that the onset and propagation of virulent or drug resistant sub-types can be tracked. The multifaceted problem of zoonotic transmission requires a One Health approach, which acknowledges the interdependence between human health, animal health and environmental health. The scientific community can help to better inform the public health policy, devise new interventions, and ultimately decrease the burden of the disease by further unravelling the genetic complexities of *Cryptosporidium* in the world.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

References

[1] Xiao L, Fayer R, Ryan U, Upton SJ. Cryptosporidium taxonomy: Recent advances and implications for public health. Clinical Microbiology Reviews. 2004 Jan;17(1):72-97.

[2] O'Leary JK, Sleator RD, Lucey B. *Cryptosporidium spp*. diagnosis and research in the 21st century. Food and Waterborne Parasitology. 2021 Sep;24:e00131.

[3] MacKenzie WR, Hoxie NJ, Proctor ME, Gradus MS, Blair KA, Peterson DE, et al. A massive outbreak in Milwaukee of Cryptosporidium infection transmitted through the public water supply. New England Journal of Medicine. 1994 Jul;331(3):161-167.

[4] Checkley W, White AC, Jaganath D, Arrowood MJ, Chalmers RM, Chen XM, et al. A review of the global burden, novel diagnostics, therapeutics, and vaccine targets for Cryptosporidium. The Lancet Infectious Diseases. 2015 Jan;15(1):85-94.

[5] Yang X, Guo Y, Xiao L, Feng Y. Molecular epidemiology of human cryptosporidiosis in low- and middle-income countries. Clinical Microbiology Reviews. 2021 Apr;34(2):e00087-19.

[6] Hunter PR, Nichols G. Epidemiology and clinical features of Cryptosporidium infection in immunocompromised patients. Clinical Microbiology Reviews. 2002 Jan;15(1):145-154.

[7] Bouzid M, Hunter PR, Chalmers RM, Tyler KM. Cryptosporidium pathogenicity and virulence. Clinical Microbiology Reviews. 2013 Jan;26(1):115-134.

[8] Robinson G, Chalmers RM, Elwin K, Guy RA, Bessonov K, Troell K, et al. Deciphering a cryptic minefield: A guide to Cryptosporidium gp60 subtyping. Current Research in Parasitology & Vector-Borne Diseases. 2025;7:100257.

[9] Feng Y, Ryan UM, Xiao L. Genetic diversity and population structure of Cryptosporidium. Trends in Parasitology. 2018 Nov;34(11):997-1011.

[10] Pedraza-Díaz S, Amar C, Nichols GL, McLauchlin J. Nested polymerase chain reaction for amplification of the Cryptosporidium oocyst wall protein gene. Emerging Infectious Diseases. 2001 Jan;7(1):49-56.

[11] Agyabeng-Dadzie F, Shinko J, Engel JC, Kissinger JC. Cryptosporidium genomics—Current understanding, advances, and applications. Current Tropical Medicine Reports. 2024 Jun;11(2):92-103.

[12] Ahmadpour E, Safarpour H, Xiao L, Zarean M, Hatam-Nahavandi K, Barac A, et al. Cryptosporidiosis in HIV-positive patients and related risk factors: A systematic review and meta-analysis. Parasite. 2020;27:27.

[13] Korpe PS, Valencia C, Haque R, Mahfuz M, McGrath M, Houpt E, et al. Epidemiology and risk factors for cryptosporidiosis in children from 8 low-income sites: Results from the MAL-ED study. Clinical Infectious Diseases. 2018 Dec;67(11):1660-1669.

[14] Korich DG, Mead JR, Madore MS, Sinclair NA, Sterling CR. Effects of ozone, chlorine dioxide, chlorine, and monochloramine on Cryptosporidium parvum oocyst viability. Applied and Environmental Microbiology. 1990 May;56(5):1423-1428.