

Advancing the Science of Water: AwwaRF and *Giardia* and *Cryptosporidium*

The intestinal parasites *Giardia* and *Cryptosporidium* are single-cell protozoa that cause gastrointestinal infections in humans as well as in wild and domestic animals. These pathogens can cause disease outbreaks when individuals ingest *Giardia* cysts or *Cryptosporidium* oocysts in fecally contaminated water or food. Person-to-person transmission, which happens most commonly in daycare centers and healthcare institutions, occurs by hand-to-mouth transfer of cysts from the feces of an infected person.

Infected individuals often experience acute or chronic symptoms, primarily diarrhea and abdominal cramping, but some may be asymptomatic. Acute symptoms of giardiasis typically disappear after five to seven days but may continue for months or even years; individuals infected with cryptosporidiosis usually stop experiencing symptoms within 30 days. However, people with compromised immune systems (for example, AIDS patients, cancer patients receiving chemotherapy, and organ transplant recipients) may have prolonged infections contributing to death.

The *Giardia* species most frequently identified with waterborne disease is *Giardia lamblia*. Individual cases of giardiasis often involve hikers or campers who ingested untreated water in the backcountry, and some incidences of the disease have been associated with foreign travel. The first waterborne outbreak of giardiasis in the United States was documented in the early 1960s. Although most outbreaks associated with public water supplies have involved surface water sources, protozoa have also been found in groundwater and in treated water.

The *Cryptosporidium* species first associated with waterborne disease was *Cryptosporidium parvum*, but other species can also infect humans. Contamination of the Milwaukee, Wisconsin, water system in April 1993 caused some 400,000 cases of cryptosporidiosis; more than 1,000 people were hospitalized, and at least 100 deaths were attributed to the outbreak. Although smaller waterborne outbreaks of cryptosporidiosis had previously been documented in the United States and the United Kingdom, the size of the Milwaukee outbreak shocked water suppliers and revolutionized water treatment practices.

As researchers began to study *Cryptosporidium* in depth, they discovered a number of disturbing characteristics associated with this pathogen. As summed up by Jennifer

Clancy, president of Clancy Environmental Consultants, "*Cryptosporidium* occurs in surface water supplies worldwide, it can be carried by a variety of animal hosts, the infection it causes can be fatal to immunocompromised patients, it is difficult to filter out of water because of its small size, and it has been responsible for hundreds of waterborne disease outbreaks throughout the world, in large part because of its resistance to chlorine-based disinfectants, the final tool in the multiple barrier approach to water treatment."

Understanding Protozoan Pathogens

AwwaRF has been funding *Giardia* studies since 1984 and *Cryptosporidium* studies since 1988. These studies can be grouped into four major categories: occurrence, detection methods, health effects, and treatment techniques.

Within these four categories, AwwaRF's fundamental contributions to our understanding of these protozoan pathogens include:

- Documenting the occurrence of *Giardia* cysts and *Cryptosporidium* oocysts in both untreated and treated water supplies,
- Developing analytical methods to determine the presence of these protozoa in water supplies as well as their ability to cause infection,
- Pinpointing environmental conditions that affect protozoan viability,
- Identifying various *Cryptosporidium* species through genotyping,
- Evaluating the ability of conventional filtration and disinfection processes to control protozoan pathogens in drinking water,
- Establishing the value of particle counting in optimizing filter performance for parasite removal,
- Determining the amount of disinfectant and contact time required to inactivate *Giardia* cysts,
- Assessing the ability of alternative disinfectants and combinations of disinfectants to inactivate *Cryptosporidium*,
- Discovering the effectiveness of ultraviolet (UV) irradiation at inactivating *Cryptosporidium* oocysts,
- Demonstrating that regulations aimed at protecting consumers from pathogens and disinfection by-products (DBPs) should be considered in tandem, and
- Providing data to support the inclusion of UV disinfection as a technique for controlling *Cryptosporidium* in the Long Term 2 Enhanced Surface Water Treatment Rule.

Ascertaining Occurrence

The first AwwaRF-funded study on the occurrence of *Giardia* and *Cryptosporidium* in North American surface water supplies was a small unsolicited project from American Water, then called American Water Works Service Company ("*Giardia* and *Cryptosporidium* in Water Supplies," Project 430, funded 1988, published 1991, order number 90583). In this study, Mark LeChevallier, now director of innovation and environmental stewardship at American, and Joan Rose, then at the University of South Florida, analyzed some 200 water samples from 66 water treatment plants. They found

the two protozoa in most of these supplies, particularly the major industrialized rivers like the Mississippi and Missouri.

"At this time, most people thought these pathogens were only a problem in Colorado and Pennsylvania," said LeChevallier. "The study concentrated on American's operation areas—New England and the Midwest—not just pristine mountain supplies, and this little project showed there was a lot of crypto and *Giardia* in these surface supplies."

"This study gave us the first indication of how widespread the problem of *Giardia* and crypto in water supplies was," said Paul Rochelle, principal microbiologist at Metropolitan Water District of Southern California. "It should have been a wake-up call for the industry, but it wasn't until the Milwaukee outbreak that everyone woke up."

American submitted the proposal to AwwaRF because of a cryptosporidiosis outbreak that had occurred in Carrollton, Georgia, in January 1987 and had been investigated by the Centers for Disease Control and Prevention (CDC) in nearby Atlanta. "Carrollton had a conventional treatment plant—coagulation, sedimentation, rapid sand filters, and disinfection," said LeChevallier. "The flocculator arms had broken, and chlorine-resistant crypto were getting through the plant. Because occasional equipment breakdowns occur in all treatment plants, we wanted to know what level of *Giardia* and crypto occurrence would be of concern to our operations."

When results of the study were published, however, they had more wide-ranging effects than anticipated. At this time, the U.S. Environmental Protection Agency (USEPA) was holding its first discussions in preparation for regulating DBPs in drinking water. When American's AwwaRF-funded study showed that *Giardia* occurrence was much more extensive than previously thought, USEPA conducted new risk assessments, which indicated that the disinfection requirement for inactivating *Giardia* specified in the Surface Water Treatment Rule (SWTR) allowed a higher risk of infections from this pathogen than the agency had realized when the rule was promulgated in 1989.

"Our study was a key indicator that reducing microbial risk and reducing DBPs had to go hand in hand," LeChevallier said.

After the Milwaukee outbreak, water suppliers across the United States and Canada became concerned about *Cryptosporidium*. "Milwaukee was a major wake-up call for the drinking water industry, and AwwaRF was instrumental in following up on that," says LeChevallier. "Having a centralized, science-driven research group that could follow up on a crisis like this was fundamental. The foundation pulled together the industry's concerns and focused on applied research in a coordinated fashion. The progress that resulted probably wouldn't have happened without AwwaRF serving as a clearinghouse."

The year after the Milwaukee event, AwwaRF funded a project to study the effects of various environmental conditions—temperature, age, physical stress—on the viability of protozoan pathogens in water supplies ("*Giardia* Cyst and *Cryptosporidium* Oocyst Survival in Watersheds and Factors Affecting Inactivation," Project 151, funded 1994,

published 1999, order number 90761). Led by Syed Sattar at the University of Ottawa, the study also examined the susceptibility of these parasites to disinfection.

Giardia and crypto were included in a subsequent project that documented the occurrence of fecal contaminants in six North American watersheds—one in Waterloo, Canada, and five in the States ("Source Water Assessment: Variability of Pathogen Concentrations," Project 488, funded 1997, published 2002, order number 90906). This project, co-sponsored by AwwaRF and USEPA through the Microbial/DBP (M/DBP) Research Council, was based on an earlier study conducted by LeChevallier and Tom Atherholt in New Jersey and involved American Water, Clancy Environmental Consultants, and McGuire Environmental Consultants.

"We analyzed 100 samples from each watershed—more than 600 samples—enough to make sure that if something was there, we found it," said Clancy. "We looked at protozoa, bacteriophages, coliphages, *E. coli*, *Clostridium perfringens*, and total and fecal coliforms, collecting data on indicators of fecal contamination as well as actual pathogens. The results gave us a good database on how much *Giardia* and crypto and other fecal contaminants are out there."

Once researchers had a better understanding of *Giardia* and crypto occurrence in source water supplies, the next step was to determine whether they were present—and still infectious—in treated water. In a current AwwaRF study, Rochelle is using a cell culture method to look for infectious *Cryptosporidium* in filtered drinking water ("Detection of Infectious *Cryptosporidium* in Filtered Drinking Water," Project 3021, funded 2004, ongoing). The study was designed to assess the results of an internally funded project at American Water, where, according to LeChevallier, "We found live *Cryptosporidium* in 1.4 percent of the filtered water samples we analyzed."

Developing Detection Methods

The cell culture technique used by Rochelle and LeChevallier represents the most recent development in a long evolution of analytical methods aimed at detecting both the presence and viability of *Giardia* and crypto in water supplies. "Advances in methodology continue," says LeChevallier. "And AwwaRF funded various projects that led to better analytical methods."

Immunomagnetic Separation. Clancy's lab conducted several studies that enhanced our ability to detect protozoan pathogens, including the first study on immunomagnetic separation ("New Approaches for Isolation of *Cryptosporidium* and *Giardia*," Project 364, funded 1996, published 2000, order number 90814). "This study improved on the fluorescent monoclonal antibody method first used to measure *Giardia*," said Clancy. "We looked at each step—from sample collection, through separation, to the antibodies—to come up with what basically became USEPA Method 1623, now used worldwide."

Method 1623, a combined test for *Giardia* and crypto that's part of USEPA's 1600 series of microbiological methods, "isn't perfect," according to LeChevallier, "but it has higher

recoveries and is more consistent than earlier analytical methods. With microscopic methods like this, you just see an organism—you can't tell if it's alive or dead."

A more recent AwwaRF project led by Zia Bukhari (then in Clancy's lab) examined methods for recovering small concentrations of oocysts from large-volume water samples ("Recovery of *Cryptosporidium* Oocysts From High-Volume Water Samples," Project 2595, funded 2000, published 2003, order number 90964F). "These samples were 1,000 liters in volume," said Clancy, "so this study gave us a detection method for finished water as well as a method for source water."

Mouse Infectivity. AwwaRF has also funded some significant viability and infectivity studies, according to Rochelle. "It's one thing to know crypto are present, but what's important is to know how infectious they are," he says. Charles Sterling's lab at the University of Arizona did some of the first work to develop viability assays and evaluate how well they correlated with infectivity in animals ("Development of a Test to Assess *Cryptosporidium parvum* Oocyst Viability: Correlation With Infectivity Potential," Project 609, funded 1990, published 1993). "This study introduced the concept of using infectivity models—mouse models—in drinking water applications," said Rochelle.

The Clancy and Arizona labs continued this work in a project funded by AwwaRF and the UK Drinking Water Inspectorate ("*Cryptosporidium* Viability and Infectivity Methods," Project 395, funded 1996, published 2000, order number 90799). This study involved two UK labs as well as the two U.S. labs, allowing identical trials to be conducted in both countries. The mouse infectivity measurements proved more effective at demonstrating oocyst inactivation than the other methods tested, according to Clancy.

Although the mouse model was the preferred method for some years, it also has limitations. "Mouse assays are terribly labor intensive," Rochelle points out. "Because it's hard to introduce any kind of automation into them, every step involves manual labor, and this makes them expensive. In addition, the results are not always reproducible. You can't guarantee the way an animal will respond to a particular infectious agent."

Cell Culture. Rochelle, Clancy, and LeChevallier all favor the more recently developed cell culture method. "Cell culture allows a much higher number of replicates," says Rochelle. "It's more amenable to automation, you get higher throughput, and you don't have the ethical issue associated with experiments involving animals."

Rochelle conducted the first thorough comparison of the cell culture method and the mouse assay ("Comparison of Cell Culture and Mouse Assays for Measuring Infectivity of *Cryptosporidium*," Project 2589, funded 1999, published 2004, order number 90127F). "Up to this point, many analysts considered the mouse model the only reliable method for measuring infectivity," said Rochelle. "We demonstrated that cell culture was equivalent to the standard mouse model in terms of sensitivity and reproducibility."

Rochelle's study "marked another milestone in methodology," according to Clancy. "It allowed us to move away from the mouse model, which was very expensive. Now

everyone uses the cell culture method." LeChevallier also considers the new method an important advance. "Molecular testing allows us to identify the organism and to determine whether it is infectious in human intestinal cells," he said.

Genotyping. AwwaRF's work on genotyping methods constitutes another significant advance in our understanding of *Cryptosporidium*. "Some of the genetic analyses have shown that *Cryptosporidium* is more complex than we realized, that other strains from different animals exist," says LeChevallier.

"Through a variety of avenues in many countries, we've discovered that at least two species of *Cryptosporidium* are equally important in terms of waterborne disease," said Rochelle. "With the immunofluorescent assay, the two species are indistinguishable; only with molecular methods can we tell them apart."

Rochelle says AwwaRF funding has been instrumental in what we've learned about the speciation of *Cryptosporidium* over the past six to eight years. "Until the late 1990s, we thought of *Cryptosporidium parvum* as the big issue," he said. "Now *Cryptosporidium hominis*, a human-specific species, is recognized as well. Prior to five years ago, we didn't know this second species existed."

In AwwaRF's first project on crypto genotyping, Rochelle used a molecular fingerprinting technique to describe the genetic characteristics that identify the source of some crypto oocysts ("Evaluation of Genotyping Techniques for *Cryptosporidium parvum*," Project 366, funded 1996, published 2001, order number 90850). The study also described how water utilities could incorporate this technique into source water monitoring programs.

A subsequent project led by Lihua Xiao at CDC assembled genotyping information from diverse sources ("Development and Standardization of a *Cryptosporidium* Genotyping Tool for Water Samples," Project 2675, funded 2000, published 2006, order number 91101). The Xiao study helped to standardize the genotyping method, making it easier to compare studies from different labs, according to Rochelle.

Documenting Health Effects

Of course waterborne disease outbreaks caused by *Giardia* and crypto, including the one in Milwaukee, have been instructive about the health effects of these pathogens, but epidemiologic studies have produced data about the occurrence of drinking water-related gastrointestinal (GI) illness under controlled experimental conditions. Pierre Payment at the University of Quebec directed an epidemiologic study funded by AwwaRF and a number of research partners, including Compagnie Générale des Eaux and several governmental agencies in Canada ("An Epidemiological Study of Gastrointestinal Health Effects of Drinking Water," Project 919, funded 1993, published 2000, order number 90772).

"Payment's studies were the first to link the increased incidence of GI illness with consumption of tap water that was in compliance with regulations," said Stig Regli, a USEPA policy analyst who had primary responsibility for developing the SWTR. In these studies, river water entering the treatment plant contained parasites, viruses, and bacteria at concentrations typical of fecally contaminated water supplies throughout the world, and the treatment plant produced finished water that met or exceeded Canadian and U.S. water quality standards at the time.

USEPA recently used Payment's data to generate a national estimate of the risk of GI illness, according to Regli. "Payment's studies were unique in that they showed the magnitude of the potential risk posed by a mixture of pathogens that could be in drinking water. Other studies only provided data for risk estimates related to single organisms," Regli said.

Removing or Inactivating Protozoa During Water Treatment

"Anything that's done to protect water supplies from *Cryptosporidium* generally protects them from *Giardia* as well," Rochelle says. "Since Milwaukee, protozoan research has focused predominantly on *Cryptosporidium*, and AwwaRF's impact has been significant."

Controlling *Giardia*. Before Milwaukee, however, two groundbreaking AwwaRF projects funded in the mid-1980s studied *Giardia* removal and inactivation. Focusing on removal techniques, Jerry Ongerth at the University of Washington evaluated the ability of full-scale filtration systems to remove *Giardia* cysts and cyst-size particles under normal operating conditions ("A Study of Water Treatment Practices for the Removal of *Giardia lamblia* Cysts," Project 074, funded 1984, published 1989, order number 90542). The study also provided design and operation guidelines for maximizing cyst removal and concluded that cyst removal was likely to be comparable to the effectiveness of turbidity removal.

Focusing on inactivation, Charles Hibler at Colorado State University observed that cold water temperatures prolonged the life span of *Giardia* cysts, allowing survival times of up to two months, and slowed the reaction of some disinfectants ("Inactivation of *Giardia* Cysts With Chlorine at 0.5°C to 5°C," Project 107, funded 1985, published 1987, order number 90526). The study's most noteworthy contribution, however, was delineating various chlorine concentrations and contact times required to inactivate *Giardia* cysts in low-temperature water supplies.

USEPA used the concept of *CT*—the product of disinfectant concentration (*C*) and contact time (*T*)—to define the disinfection requirements specified in the SWTR. Under this regulation, all surface water supplies had to be filtered unless they could meet filtration avoidance requirements by inactivating cysts with disinfection. "The results of Hibler's study establishing *CT* values for *Giardia* inactivation were used to support the criteria for avoiding filtration," said Regli.

Evaluating Filtration Performance. "*Giardia* and *Cryptosporidium* research is really about the treatment process—the integrity of filtration, the adequacy of disinfection, and balancing disinfection with minimizing DBP formation," LeChevallier says. He points to particle counting as "the first step toward optimizing filter performance to reduce particle breakthrough" and cites an AwwaRF project led by Nancy McTigue and Dave Cornwell of Environmental Engineering and Technology, Inc. ("National Assessment of Particle Removal by Filtration," Project 908, funded 1993, published 1998, order number 90757). "The research team took a trailer all over the country, found *Cryptosporidium* at concentrations similar to those found in [American's] first study, and used particle counting to assess filtration's effectiveness at removing these organisms," LeChevallier said.

These particle counting data were also used in developing two water quality regulations. "The Interim Enhanced SWTR and the Long Term 1 Enhanced SWTR were structured around tightening up the filtration process, which enhances removal of *Giardia* and *Cryptosporidium*," said Regli. "These rules made use of the particle counting work done by Cornwell and McTigue, plus their work showing the frequency with which spikes occur during the filtration cycle. The Interim Enhanced rule contains provisions for identifying when upsets in treatment occur. These provisions facilitate the recognition of elevated turbidity events and how significant they are."

Before the Filter Backwash Rule was proposed, Cornwell led an AwwaRF-funded project to analyze the microbiology of filter backwash water ("Treatment Options for *Giardia*, *Cryptosporidium*, and Other Contaminants in Recycled Backwash Water," Project 352, funded 1996, published 2001, order number 90832). "This was one of the first studies in which we used the cell culture method to look for infectious *Cryptosporidium*," said LeChevallier. "This study told us that live *Cryptosporidia* were not only in the source water but were also making it through the treatment process to the filters and into the backwash water. The next question was if they're making it *to* the filters, can they make it *through* the filters? We addressed this question in our internally funded project, which is currently being followed up by the AwwaRF project led by Rochelle [Project 3021, discussed previously]."

Looking for Alternative Disinfectants. AwwaRF has also supported numerous studies to assess the ability of various disinfection processes to inactivate protozoan pathogens. Once it became clear that none of the chlorine-based disinfectants was totally effective at inactivating *Cryptosporidium* oocysts (despite chlorine's ability to inactivate *Giardia* cysts), researchers began to explore the efficacy of alternative disinfection schemes.

"The early AwwaRF-funded work on ozone disinfection was critical to gaining an understanding of whether *Cryptosporidium* could be controlled at the treatment plant," said Rochelle. "In 1991, Gordon Finch at the University of Alberta did the earliest groundbreaking work on ozone disinfection of protozoa ["Ozone Disinfection of *Giardia* and *Cryptosporidium*," Project 731, funded 1991, published 1994, order number 90661]. By then, we knew that chlorine wouldn't do it, but Finch showed that we have alternatives, that ozone was more effective."

Finch also examined the effects of sequential and simultaneous applications of multiple disinfectants on protozoa ("Synergistic Effects of Multiple Disinfectants," Project 273, funded 1995, published 2000, order number 90778, and "Sequential Disinfection Design Criteria for Inactivation of *Cryptosporidium* Oocysts in Drinking Water," Project 348, funded 1996, published 2001, order number 90831). Clancy says "Finch's work with ozone and chlorine dioxide was interesting" but points out that these bench-scale experiments were conducted with laboratory water samples. "When the work was repeated with samples of natural water supplies, we didn't see the same effects," Clancy said.

Joan Oppenheimer at Montgomery Watson conducted AwwaRF's follow-up study with natural water supplies in conjunction with the Dutch organization Kiwa Water Research ("Evaluation of *Cryptosporidium* Inactivation in Natural Waters," Project 375, funded 1996, published 2000, order number 90797). "This was the first big study of the effectiveness of a wide range of disinfectants at inactivating *Cryptosporidium* under fairly realistic conditions," said Rochelle. "It was a bench-scale study—and very thorough."

Stumbling Onto UV. The big breakthrough in crypto inactivation, however, came in the mid-1990s when, as Clancy phrases it, "we stumbled onto UV." She describes the use of UV irradiation as the newest weapon in the war against *Cryptosporidium* but says this technology was almost overlooked in the search for ways to inactivate oocysts.

In a 2003 essay called "Let There Be Light—UV Light," Clancy recounts what she calls "the serendipitous discovery that UV inactivated crypto." AwwaRF and the Electric Power Research Institute jointly funded a project in which a research team from her lab and Sterling's lab at the University of Arizona evaluated the effectiveness of several electrotechnologies at inactivating crypto ("Innovative Electrotechnologies for *Cryptosporidium* Inactivation," Project 282, funded 1995, published 1998, order number CR-111090 available from EPRIAMP Customer Assistance Center, 800.432.0267).

"Because the University of Arizona team had shown in a 1990 study that chlorine was ineffective against *Cryptosporidium*, we undertook this new research to determine if any commercially available electrotechnologies had the potential to inactivate *Cryptosporidium* oocysts in drinking water. The study did not assess inactivation levels versus treatment doses, nor did it attempt to optimize any technology—it was simply a quick look-see to screen the technologies for further study if warranted," Clancy explained. For these initial experiments, the analysts used staining and excystation methods to measure oocyst inactivation because these assays were inexpensive and simple to perform.

"Two electrotechnologies appeared to be successful at inactivating oocysts—pulsed UV and advanced UV (low-pressure UV over an extended period of exposure). Conventional low-pressure UV appeared to have no effect on oocyst viability," Clancy said. But in retrospect, she concedes, measuring inactivation with the surrogate assays instead of

mouse infectivity probably caused the researchers to miss the inactivation potential of the lower UV doses administered by the advanced and pulsed systems.

The serendipitous discovery happened during another AwwaRF-funded project conducted by the same researchers at about the same time ("*Cryptosporidium* Viability and Infectivity Methods," Project 395, discussed earlier). In this study, the research team used UV disinfection to try to identify a surrogate assay that could predict the results of the animal infectivity model for determining oocyst viability, but all the surrogate assays significantly underestimated inactivation compared with the mouse infectivity measurements, according to Clancy.

"We discovered that although there was no surrogate for animal infectivity, UV was inactivating all the crypto anyway—even at low doses," Clancy said. As a result, the research team realized they had probably made a mistake by reporting that low-pressure UV in the previous study had been ineffective at inactivating *Cryptosporidium*. "We conducted additional studies using the mouse model and found that low-pressure UV is highly effective at oocyst inactivation," Clancy said.

By the late 1990s, the Clancy–University of Arizona team had conducted dozens of UV inactivation studies using mouse infectivity, and, according to Clancy, they were confident that UV was effective at inactivating *Cryptosporidium* oocysts. Other researchers, many funded by AwwaRF, repeated their work and corroborated the results, she said. But Clancy doesn't view her group's discovery as something only they could have done. "UV was waiting to be discovered, and it was only a matter of time before someone gave it a second look," she said.

Exploring Additional Questions About UV. Once UV disinfection was recognized as a viable method of inactivating crypto, mounting interest in the technology prompted researchers to address any lingering reservations about its use. Two of the most frequently asked questions were whether UV could inactivate more than one crypto strain and whether the protozoan could repair itself after being subjected to UV irradiation.

"All of the previous work had been done with the Iowa strain of *Cryptosporidium*, and people wanted to know if other strains were resistant to UV," Clancy said. AwwaRF's number-one ranked project in 2000 was an unsolicited proposal submitted by Clancy and Marilyn Marshall at the University of Arizona ("*Susceptibility of Multiple Strains of C. parvum* to UV Light," Project 2721, funded 2000, published 2002, order number 90916). Clancy and Marshall tested four strains of *Cryptosporidium parvum* for their susceptibility to inactivation by low-pressure UV light at various doses. "This project put an end to questions related to the Iowa strain," Clancy said. "We confirmed that low-dose UV works on other strains as well and that all these strains are equally susceptible to UV inactivation."

A Rochelle study, also funded in 2000, confirmed that UV inactivated multiple strains of crypto ("*An Investigation of UV Disinfection and Repair in Cryptosporidium parvum*," Project 2669, funded 2000, published 2004, order number 91015F). Working with Steve

Upton at Kansas State University, Rochelle also determined that although crypto has the genetic material necessary to repair itself from UV damage, for some reason it's not able to do this. "We don't know why," Rochelle said. "Many other organisms can repair varying degrees of UV-induced damage—humans, for example."

Conclusion

As a result of research demonstrating that alternative disinfectants such as UV and ozone can inactivate chlorine-resistant *Cryptosporidium* oocysts, water utilities have better tools for protecting the public against protozoan pathogens. AwwaRF data also enabled USEPA to promulgate more practical and affordable regulations for controlling waterborne protozoa. "AwwaRF played a crucial role in the discovery of UV as a solution to the problem of *Cryptosporidium* control in drinking water and in moving the technology into widespread acceptance by all facets of the water supply community," Clancy said.

The acceptance of UV disinfection allows large and small public water systems to enhance their treatment processes without incurring the higher construction, energy, and operations costs associated with some other treatment methods. Water suppliers are now in a good position to evaluate the feasibility of incorporating alternative disinfectants into their own treatment schemes, taking into account factors such as cost, water quality characteristics, and other site-specific conditions.