

## PARASITES

### EFFICACY OF SANITIZERS AND DISINFECTANTS ON PATHOGENIC PARASITES

(Y.R. Ortega, M.P. Torres, S. Van Exel, L. Moss, and V. Cama)

Protozoan parasites have been associated with foodborne outbreaks. Parasite contamination has been of particular interest in produce and fruits that are consumed raw. The objective of this study is to identify disinfectants and sanitizers that can inactivate parasites. Hydrogen peroxide, ammonium hydroxide, N-alkyl dimethyl benzyl ammonium chloride and a peroxyacetic acid based sanitizers at various concentrations and treatment times were examined for its ability to inactivate *Encephalitozoon intestinalis* spores. Microsporidia spores could be efficiently inactivated at a 5% ammonium hydroxide concentration for 1 min. Prolonged exposure to lower dilutions did not significantly reduce spore counts as determined by *in vitro* cultivation. Tsunami at 40µg/ml partially inactivated microsporidial spores when incubated at 1 min but was not statistically significant. Complete inactivation was achieved with at least 5 min exposure, whereas at 20µg/ml, complete inactivation was achieved after 15 min incubation. In this study, hydrogen peroxide at all concentrations and ammonium hydroxide at 5% effectively inactivated microsporidial spores. Therefore, *E. intestinalis* spores are more susceptible to inactivation than *Cryptosporidium* oocysts when using these two chemicals. *Cryptosporidium parvum*, *Encephalitozoon intestinalis*, and *Cyclospora cayetanensis* were experimentally inoculated into lettuce and basil leaves and treated with gaseous chlorine dioxide at 4.1 mg/ml. After treatment, *Cryptosporidium* and microsporidia viability was determined by *in vitro* cultivation using HCT-8 and RK-13 cell lines respectively. *Cyclospora* viability was determined by sporulation for 2 week period at 23C. *C. parvum* log reduction of 2.75 to 3.26 was achieved. Microsporidia reduction was 3.6 and 3.58. No reduction was observed in *Cyclospora* sporulation, whereas a 2.6 to 3.2 log reduction of *E. coli* was achieved.

### PRESENCE OF *NEOSPORA CANINUM* SPECIFIC ANTIBODIES

#### ISOLATED FROM DAIRY FARMS IN GEORGIA AND TEXAS

(Y.R. Ortega, M.P. Torres, and K.D. Mena)

Bovine neosporosis is a parasitic disease produced by *Neospora caninum* which induces abortion in cows, and consequently has a negative impact on the herd's reproductive efficiency. This study demonstrated the presence of specific IgG to *Neospora* in milk and serum samples obtained from three dairy farms in Georgia and two in Texas. Using a western blot assay, samples from four hundred fourteen dairy cows were examined of which 362 were milk and 87 were serum. Samples with antibodies to *Neospora* were identified in 32.1% (105/327) of the examined animals in Georgia, whereas in Texas it was identified in 10.3% (9/87). Positive Georgia samples were found in 24 % from farm A (28/115), 21.6% from farm B (30/139), and 64.4% from farm C (47/73). In Texas, 13.5% (7/52) of animals in farm D and 5.71% (2/35) from farm E also had specific antibodies to *Neospora*. The number of animals from Georgia dairy farms with antibodies to *Neospora* was significantly higher than the Texas dairy farms. This may be related to the age of the animals examined in this study (more than 2 years old). Antibodies present in sera had excellent agreement with the antibodies present in milk. Collection of milk samples for serological testing is easier and less invasive than obtaining bovine sera, therefore offering an alternative for testing of animals.

### DETECTION OF *CYCLOSPORA* IN VEGETABLES

#### AND POTENTIAL CONTAMINATION SOURCES IN ENDEMIC AREAS

(Y.R. Ortega, A. Robertson, V.A. Cama, A. Mann, L. Cabrera, C. Taquiri, L. Xiao, and R.H. Gilman)

*Cyclospora cayetanensis* causes gastrointestinal illness and it is thought that people get infected via the food- or water-borne routes. In this study, we investigated the presence of *Cyclospora* in vegetables and hands of produce vendors in Pampas de San Juan in Lima Peru, a community where *Cyclospora* is endemic. We also surveyed other potential contamination factors, such as river irrigation water and soil from three agricultural fields. Vegetables and hand washes were collected from 20 vendors from 4 markets at 3 survey dates in 2004. All samples were analyzed by microscopy and PCR-RFLP for the presence of *Cyclospora*. It was identified in vegetables in the March and June surveys, but not in December. Hand washes were positive only in the March survey, one with *Cyclospora* and two with *Eimeria* spp. Irrigation water with *Cyclospora* was identified in March in 2 of 3 rivers, and 2 of 3 soil samples and, 1 of 3 rivers in August. Our findings show a seasonal trend in the detection of *Cyclospora*, which

mimics the presence of cyclosporiasis in the community. Detecting *Cyclospora* in vegetables, hands of vendors, irrigation water, and agricultural soil demonstrate the food and water-borne potential of this parasite and give new information to understand the dynamics of produce contamination.

#### **OUTBREAK INVESTIGATION OF CYCLOSPORIASIS IN NAVAL RECRUITS IN LIMA, PERU**

(C.C. Mundaca, P.A. Torres-Slimming, R.V. Araujo-Castillo, M. Morán, O. Colina,  
D. Bacon, Y. Ortega, David L. Blazes, and R. Gilman)

*Cyclospora cayetanensis* has been responsible for several epidemics in the last decade. In March 2005, an outbreak of diarrhea was identified in recruits at the Ancon Naval Base in Lima, Peru. A case-control study was carried out. Enrolled individuals in the study completed an epidemiological questionnaire including demographic data, eating habits, food items consumed, and symptoms. Complete data from 52 recruits was available for the analysis. 37 met the criteria for case and 15 for control. The epidemic curve indicated a point source transmission, with cases occurring over 9 days with a peak on the fifth day. There was no association between diarrhea and consumption of tap water or with water stored in plastic containers in the dormitories. PCR for *C. cayetanensis* detected 20/35 (57.1%) cases and 3/15 (20%) controls, demonstrating the improved diagnostic yield of this technique. This is the second report to characterize an outbreak of diarrhea due to *C. cayetanensis* in Peru among recruits. The epidemiology and clinical course are similar to other reported outbreaks in developed regions.

#### **DETECTION OF OOCYSTS OF *CYCLOSPORA CAYETANENSIS* IN HUMANS, DOGS AND SEWER SAMPLES**

(Y.R. Ortega, V.A. Cama, A. Robertson, A. Mann, L. Cabrera, C. Taquiri, L. Xiao, and R.H. Gilman)

Infections with *Cyclospora* have been primarily associated with foodborne transmission. In endemic areas, *Cyclospora* shows a defined seasonal pattern, and it is estimated that 7-15 days are required for it to sporulate and become infectious. We studied two other potential sources of *Cyclospora* oocysts in Pampas de San Juan, a location where we had a pediatric longitudinal cohort for endemic cyclosporiasis. Fecal specimens from local dogs and sewer samples from 10 different locations were examined for the presence of *Cyclospora* oocysts. Three *Cyclospora*-infected participants from three different households also had dogs with *Cyclospora* in their stools, with concurrent detection of parasites in all three episodes. The histo-pathological examination of tissues from one dog did not reveal infections in this animal and sequencing information demonstrated that the human and dog isolates were similar, suggesting a spurious infection. The sewer samples were collected on December 2005, January and March, 2006. Microscopy examination of the sewer pellets detected pathogenic parasites such as *Giardia*, *Ascaris*, *Trichuris* and *Ancylostoma* and the commensal parasites *Chilomastix mesnili*, *Endolimax nana*, and *Escherichia coli*. PCR testing identified *Cyclospora* in two sites, six other sites were positive at least once, and two sites were always negative. GPS mapping of the *Cyclospora*-negative sewer sites correlated with areas of low prevalence of *Cyclospora*, however several positive sites corresponded to areas where *Cyclospora* was not frequently detected in the study population. Our findings suggest that sewer samples can be used to determine the endemicity of *Cyclospora* in a community, that PCR detection is a more reliable method when testing sewer samples, that in endemic settings *Cyclospora* infections may not be restricted to young children, and that the observation of *Cyclospora* oocysts in animal stools is the result of spurious infections.