metabolites between the two cells is essential for parasite survival and may be regulated by transporters at the host-parasite interface, i.e. the feeder organelle. We have localized a ~200 kD integral membrane protein, CpABC-1, from *C. parvum*, to the host-parasite boundary, possibly the feeder organelle. We have cloned and characterized the CpABC-1 gene. The predicted amino acid sequence of CpABC-1 has significant structural similarity with the cystic fibrosis conductance regulator (CFTR) and the multidrug resistance protein (MRP) subfamily of ATP-binding cassette (ABC) proteins. We have identified a second ABC protein gene, CpABC-2, in *C. parvum*. CpABC-2 is a single copy gene located on chromosome 2. The CpABC-2 gene identified a 5.3 kb mRNA in sporozoites. Preliminary sequence analysis identified CpABC-2 as a member of the MRP subfamily of ABC proteins. ATP-binding cassette proteins are associated with xenobiotic resistance phenotypes in many taxa.

74 ANTIBODY AND INTRAEPITHELIAL LYMPHOCYTE RESPONSES DURING *CRYPTOSPORIDIUM PARVUM* INFECTIONS IN SUCSEPTIBLE AND RESISTANT ATHYMIC NUDE MICE. Adjei AA, Curran BC, Castro M, Shrestha AK, Delsid LD, Velez M, Fritz H, and Enriquez FJ. Department of Veterinary Science and Microbiology, The University of Arizona, Tucson, AZ.

Differences in susceptibility to *Cryptosporidium parvum* infection between two strains of adult athymic nude mice prompted us to investigate the immune mechanism(s) that may control resistance to infection in these T-cell deficient mice. We studied fecal oocyst shedding, serum and fecal parasite-specific antibody responses, fecal immunoglobulin levels, and phenotypes of small intestinal Intraepithelial lymphocytes (IEL) in athymic C57BL/6J and BALB/cJ nude mice following oral inoculation with IC. parvumI oocysts. Inoculated C57BL/6J nude mice shed significantly fewer IC. parvumI oocysts than inoculated BALB/cJ nude mice from day 52 to day 63 post inoculation ($P < 0.05$). Inoculated C57BL/6J nude mice had significantly higher fecal parasite-specific IgA and IgM levels than inoculated BALB/cJ nude mice ($P < 0.05$) and significantly higher serum parasite-specific IgA levels at 63 days post inoculation ($P < 0.03$). In contrast, inoculated BALB/cJ nude mice had higher fecal levels of non parasite-specific IgA and IgM than inoculated C57BL/6J nude mice ($P < 0.05$). Analysis of IEL surface markers revealed that inoculated C57BL/6J mice had a higher percentage of +, CD4+, and both CD8+ + IEL than inoculated BALB/cJ nude mice. Conversely, inoculated C57BL/6J nude mice had a lower percentage of + IEL than inoculated BALB/cJ nude mice ($P <0.05$). We conclude that parasite-specific fecal IgA and IgM antibodies, +, CD4+, and/or CD8+ + IEL may be associated with resistance to C. parvum in C57BL/6J nude mice; and that thymus-independent regulatory and effector responses in the intestine may be a mechanism by which athymic nude mice resolve C. *parvum* infection.

75 MOLECULAR EPIDEMIOLOGY OF *CYCLOSPORA* AND *CRYPTOSPORIDIUM* INFECTIONS AMONG US MILITARY PERSONNEL IN INDONESIA. Higgins JA, Kerby S, Trout J, Fayer R, Xiao L, and Fryauff D. USDA-ARS, Beltsville, MD; GeoCenters/Special Pathogens Branch, USAMRIID, Ft Detrick; MD, CDC, Atlanta, GA; and NAMRU-2, Jakarta, Indonesia.

Upon posting to Indonesia, US military personnel can acquire acute or chronic diarrheal disease, in which *Cyclospora* and *Cryptosporidium* oocysts are observed in the feces. We isolated oocysts from fecal specimens, obtained from these non-indigenous patients during their visits to clinics in Jakarta, and extracted DNA for use in PCR assays. Using the Relman et al. 18S rRNA nested PCR, we amplified nested product (294 bp) from 3 of 6 specimens. Sequencing of these products indicated >99% homologies with the published sequence of *C. cayetanensis*. More recently, we have had some success in amplifying a larger (1 kb) 18S rRNA fragment from another fecal specimen by modifying the PCR parameters. The results of ongoing efforts to characterize these isolates at the molecular level will be presented. Some *Cyclospora* patients had additional oocyst-like objects present in their feces, with light microscopy morphology similar to *Cryptosporidium*. We will present data on our attempts to characterize these *Cryptosporidium* isolates as well. Our observations to date indicate that diarrheal disease among US military personnel in Indonesia may be due to co-infections with these two protozoal species, with obvious implications for diagnostic and therapeutic approaches.

76 EFFECTS OF ^{60}CO IONIZING RADIATION ON *TOXOPLASMA GONDII* TACHYZOITES. Hiramoto RM, Galisteo Jr AJ, Ferreira MA, and Andrade Jr HF. Lab. Protozoologia, Instituto de Medicina Tropical de São Paulo, USP, São Paulo, Brazil; Instituto de Pesquisas Energética e

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Toxoplasmosis is a protozoan disease which present acute infection with subsequent long lasting non-sterilizing immunity, with remaining tissue cysts. This immunity are capable to control new infections, providing the hope of the development of a vaccine, with parasite fractions or non reproductive organisms. Ionizing radiation are specially efficient in the induction of genetic damage that could abolish completely the reproduction of agents, but without affecting its immediate viability and physiology. Its use in *Schistosoma* experimental immunization schedules suggested that could be a useful tool for the development of vaccines for complex organisms. The effects of ^{60}Co γ-rays on the morphology, metabolism, infectivity, and antigenicity of *Toxoplasma gondii* was studied. The growth of tachyzoites on LLC-MK2 cells was completely blocked by 200 Gy ^{60}Co irradiation. We analyzed the metabolism of irradiated tachyzoites by MTT oxidative conversion, ^{3}H-proline protein incorporation and ^{3}H-Hypoxanthine nucleic acid synthesis in short term cultures. Infectivity was tested in cell cultures with Giemsa and Immunohistochemistry. Antigenicity was tested by serially antibody detection by ELISA and Western blotting, comparing with drug treated infected mice or in mice immunized with formolized parasites. *T. gondii* maintained its morphology after irradiation, as shown by electron microscopy. The irradiated parasites presented the same respiratory response, protein synthesis and nucleic acid incorporation as non irradiated parasites in short term cultures. Cell invasion was similar in irradiated and controls tachyzoites, but no reproduction and degeneration occurs in irradiated parasites. Mice immunized with irradiated tachyzoites show longer survival time after tachyzoite challenge and less cerebral disease after cyst challenge. Irradiated *T. gondii* tachyzoites maintain most of their metabolic function, without reproductive capacity, providing a new approach for experimental toxoplasmosis and vaccine development.

77 AMINOPEPTIDASES OF MICROSPORIDIA. Millership JJ, Chappell CL, Okhuysen PC, and Snowden KF. Department of Pathobiology, Texas A&M University, College Station, TX; and University of Texas - Houston Health Science Center, School of Public Health and Medical School, Houston, TX.

Microsporidia are eukaryotic, obligate intracellular parasites that infect a wide range of hosts. They are found with increasing frequency as opportunistic infections in immunocompromised individuals. *Encephalitozoon intestinalis* is among the most frequently diagnosed causes of diarrhea in immunocompromised humans (particularly AIDS patients). Presently, no broadly effective labeled drug treatment exists for all microsporidial species that infect humans. Aminopeptidases are proteinase enzymes that sequentially hydolyze N-terminal amino acids of oligopeptides. Many, but not all, of these enzymes are zinc metalloenzymes. Parasite aminopeptidases have been implicated in host cell invasion, immune evasion and host protein digestion. This study investigates the aminopeptidase activity of 4 human isolates of microsporidia, *Enc. intestinalis* , *Enc. cuniculi* , *Enc. hellem* and *Vittaforma corneae.* Aminopeptidase activity was assessed using a fluorometric assay that cleaves amino acid groups bound to a fluorescent substrate (7-amino-4-trifluoromethyl coumarin (AFC)). In total, 6 amino acid groups (Met, Leu, Ala, Phe, Arg and Gly) were utilized and the activity measured for each species of microsporidia. Amino acid cleavage varied among species and the enzyme activity observed for each amino acid also varied among species. No endopeptidase activity was measured using an N-terminal blocked substrate (Z-Met-AFC), for all 4 microsporidial species. The substrate cleaved most readily for each species of microsporidia was utilized in the characterization of aminopeptidase activity. The characterization of each enzyme included thermal stability, optimal pH, selected inhibitor profiles and partial purification. The purpose of this research was to identify potential targets for immunotherapeutic or chemotherapeutic agents against microsporidia.

78 JACOBUS PS-15, A DIHYDROFOLATE REDUCTASE INHIBITOR, CURED *PNEUMOCYSTIS CARINII* PNEUMONIA IN MICE. Bartlett MS, Shaw MM, Durant PJ, Smith JW, and Jacobus DP. Indiana University School of Medicine, Indianapolis, IN; and Jacobus Pharmaceutical Co., Inc., Princeton, NJ.